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Quantitative Assessment of Floristic Diversity in Large Cardamom based Traditional Agroforestry System Across Altitudinal Gradient in Darjeeling Himalaya, India

Vineeta, Bisleshna Tamang, Gopal Shukla and Sumit Chakravarty

Department of Forestry, Uttar Banga Krishi Vishwavidyalaya, Pundibari- 736165, India
E-mail: babra.vini@gmail.com

***Abstract*:** The present study was carried out in Darjeeling Himalayas region of West Bengal, India from January, 2019 to April, 2021. The study area was classified into three altitudinal class i.e., low, mid and high with eleven, nine and five holdings, respectively. The important value index (IVI) of the documented species at the studied systems varied from 0.15-17.11 irrespective of altitude classes. IVI estimated for large cardamom was 12.74 while, at low-, mid- and high-altitude class it was 9.41, 13.88 and 18.59 respectively. Overall, the most important species based on IVI was fern *Selaginella denticulata* followed by herb *Amomum subulatum*. The least important species based on IVI was a tree *Litsea glutinosa* (0.15). Based on the distribution of plant species in the system five species distribution models (SDM) were evaluated and the Preemption SDM give best fit to the data with highest Akaike Information Criterion (AIC) score.

***Keywords*:** Large cardamom, Darjeeling Himalayas, Traditional, Altitude, Distribution model

Traditional agroforestry systems are unique and culturally sound land use systems which are still practiced globally by indigenous people since the time immemorial (Fischer et al 2014, Rendón-Sandoval et al 2020) including the Himalayas (Thakur et al 2017, Bhusara et al 2016). Eastern Himalayan region of India is a part of Indo Malayan Biodiversity Hotspot (Myers et al 2000) and thus home of the different types of traditional agroforestry systems (Sharma et al 2016a-c). Himalayan region of India is categorized by highly complex socio-ecological systems due to dominance of ethnic communities with rich cultural diversity directly associated with rich species diversity (Ramakrishnan 2007, Kumar et al 2012) which is seen as the foundation for safeguarding human security in these socio-ecologically fragile mountain systems. A number of workers studied the farming systems of Himalayan including agroforestry systems (Sharma et al 2007, 2016a-c, Kumar et al 2012, Pandey et al 2017) but the quantitative information on vegetation analysis, plant biomass and other aspect of traditional agroforestry systems are very limited and scanty (Maikhuri et al 2000, Kumar et al 2012). Traditional agroforestry systems have been influenced by many factors such as religious, social and economic since long time (Nath et al 2016, Pandey et al 2017) as well as these systems are also part of our tradition and culture (Kumar et al 2012). Indigenous agroforestry systems display a great complexity with regards to their component tree, shrub, climber and herb species (Thakur et al 2005, 2017). One such system is large cardamom (*Amomum subulatum* Roxb.) based traditional agroforestry in the north eastern Indian states including the Darjeeling Himalayas (Mehta et al 2015, Shrestha 2018, Vineeta et al 2021). Large cardamom, the oldest of spices is native to Sikkim and Darjeeling Himalaya including eastern hills of Nepal (Shrestha et al 2018). The distribution of large cardamom is very limited and mainly found in Eastern Himalayan region of India, Nepal and Bhutan (Mehta et al 2015). Large cardamom is a shade loving perennial cash crop traditionally inter-mixed as understorey of natural forest tree on marginal lands and slopes with high moisture in areas of high rainfall between 1500-3500 mm at an altitude of 600 and 2000 m above mean sea level (Gudade et al 2013, Yadav et al 2015). This complex traditional large cardamom based agroforestry systems of Darjeeling Himalayas are still not clearly understood in terms of their bio-physical and socio-cultural factors that determine its floristic composition, species diversity and distribution. Therefore, this study was carried out to generate precise and systematic quantitative data on the potential of large cardamom based traditional agroforestry in the Darjeeling Himalayas along the altitudinal gradient for its Vegetational analysis.

MATERIAL AND METHODS

Study site: The present study was carried out in Kalimpong and Darjeeling districts, Darjeeling Himalayan region of West Bengal, India from January, 2019 to April, 2021 (Fig. 1).

The study site covering 3149 km^2 extends between 26° 27'

05"-27° 13' 10" N latitude and 87° 59' 30"-88° 53' E longitude with altitude of 132-3660 m. The region is humid and sub-tropical to sub-alpine with dry winter where normal temperature of coldest month is -3-18°C and warmest month is above 22°C (Saxena 2005). Soils of the region categorized as mountain and glacial soil, brown hill soil, forest soil, brown forest soil, tea soil, cinchona soil and Terai soil types (Kawosa 1988, Talwar 1988, De and Bera 1990) are acidic, yellow to red-brown in colour, silty loam to sandy loam textured and poor in calcium, magnesium, nitrogen, potassium, phosphate and organic matter (De and Bera 1990, Froehlich and Sarkar 2000). Tropical (below 800 m), subtropical (800-1600 m), temperate (1600-2400 m), cold-temperate (2400-3200 m) and sub-alpine (3200-4000 m) types of vegetation are found in the Darjeeling Himalayas (Bhujel 1996, Moktan and Das 2013, Cajee 2018). The forests in the region are mostly reserved and protected which were classified into five altitudinal zones by Basu (2000) as tropical moist deciduous (300-1000 m), tropical evergreen lower montane (1000-2000 m), tropical evergreen upper montane (2000-3000 m), temperate coniferous (3000-3500 m) and sub-alpine forest (> 3500 m).

Reconnaissance survey was conducted in the study area to explore the large cardamom based traditional agroforestry systems. The traditional large cardamom-based agroforestry farming in Darjeeling and Sikkim Himalayas were reported as the systems where large cardamom are cultivated under the canopy of reserved or protected forest leased out to the growers by the State Forest Department with no rights to cut the trees (Sharma et al 2009). Not many traditional large cardamom-based agroforestry systems/holdings were found in the study area. The size of the large cardamom holdings found were about 1-3 ha similar to the size reported from Sikkim (Sharma et al 2000, 2009). There was large cardamom under canopy cultivation in agricultural landscapes also which were not considered for the present study because these cultivations were adopted not more than a decade ago that too under the canopy of planted trees.

Sampling design: A total of 25 traditional large cardamom based agroforestry holdings found during the reconnaissance survey of the study area were distributed in the elevations between 700-2000 m, of which 17 were in Kalimpong district and only eight were in Darjeeling district. The geographical locations of the holdings were recorded with Garmin 72. Following the altitudinal chrono-sequence for distribution of vegetation in Darjeeling Himalayas by Das and Chanda (1987), Bhujel (1996), Moktan and Das (2013), Cajee (2018) and Sarkar (2020) the available traditional large cardamom holdings in the study area was also classified into three altitude class as low (700-1200 m asl), mid (1200-1700 m asl) and high (> 1700 m asl) with eleven, nine and five holdings, respectively. All the large cardamom holdings were analyzed for their phyto-sociological attributes adopting stratified random nested quadrate sampling method (Sarkar 2020, Tamang et al 2021). In these large cardamom holdings 1-4 quadrates were laid for vegetation analysis depending on its size. Holdings with size less than one hectare were laid with only quadrate in its centre and so on.

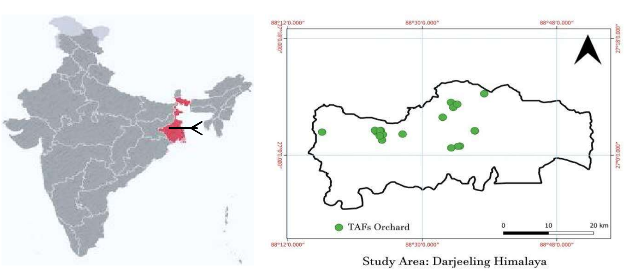

Fig. 1. Study area

Phyto-sociological analysis: In each large cardamom holding, 10 m × 10 m quadrates were laid down for trees within which two 5 m × 5 m sub-quadrates were laid at diagonal corners and five 1 m × 1 m sub-quadrates at four corners and center of the main quadrate. In each large cardamom holding the plant community was studied for their quantitative character. The trees, shrubs, herbs, climbers or any other vegetation was noted to keep an account of the floral composition of the plot.

Life form: Vegetation were stratified in to different layers according to its life forms like trees, shrubs, herbs, climbers, orchids and ferns (Johnson 1983) and every documented species were individually assigned a symbol like T, S, H, C, O and F, respectively.

Frequency or occurrence/presence: The degree of dispersion of an individual species in a community, i.e. chance of occurrence of species in each habitat is frequency (F) and expressed in per cent.

$$F=\frac{\text{No. of quadrates in which a species occurs}}{\text{Total no. of quadrates}}\times 100$$

Species documented from the large cardamom holdings were categorized as very less/rare (r), seldom present/less frequent (s), often present/frequent (o) and mostly or generally present/abundant (f) based on ≤10, 10-25, 25-75 and ≥ 75 % occurrence/presence, respectively of the total sampled homegardens.

Relative frequency: The frequency of a species relative to frequency of all other species in a community is relative frequency (RF).

$$RF=\frac{\text{No. of occurrence of a species}}{\text{No. of occurrence of all species}}\times 100$$

Raunkiaer's law of frequency: The frequency values are grouped in frequency classes to study the homogenous/heterogeneous nature of vegetation (Raunkiaer 1934). The law of frequency states that the numbers of species of a community in the five twenty per cent classes are A, B, C, D and E distributed as 0-20, 20-40, 40-60, 60-80 and 80-100 %, respectively.

Density: Abundance of a species in a unit area is expressed as density (D).

$$D=\frac{\text{Total no. of individuals of a species}}{\text{Total no. of quadrates}}$$

Relative density: Relative density (RD) is per cent representation of a species in term of number of individuals relative to all other species in a community.

$$RD=\frac{\text{No. of individuals of the species}}{\text{No. of individuals of all species}}\times 100$$

Abundance: Individuals of a species present in sampled area are expressed as abundance (A) of a species that also reflect commonality of the species in a studied habitat. Species abundance distribution (SAD) indicates most abundant and rare species in an ecosystem and was described through a number of models. Likelihood and Akaike Information Criterion (AIC) was used to fit the best SAD model out of the five models used i.e. Log-normal, Mandelbrot, Zipf, Null and Preemption models using abundance data of plant species documented from the 150 homegardens across an altitudinal gradient (Baldridge et al 2016).

$$A=\frac{\text{No. of individuals of the species in sampled quadrates}}{\text{No. of quadrates in which a species occurred}}$$

Log-normal model, a log-normal distribution is defined as a distribution whose variate conforms to the normal laws of probability. For SADs, the log-normal distribution characterizes a sample with relatively low abundance or very rare species (Matthews and Whittaker 2014). Preston (1948) introduced the log-normal SAD by demonstrating a good fit to a large number of data sets covering a number of different communities.

Log-normal (a^r) = exp [log (μ) + log (σ) Φ]

Zipf and Zipf-Mandelbrot model was originally developed for information systems, assessing the cost of information. In plant communities, the presence of a species can be seen as dependent on previous physical conditions and previous species presences.

Zipf (a^r) = Np^1rγ

Zipf-Mandelbrot a^r = (r + β)

Neutral-theory model was proposed by Hubbell (2001) and noted that the relative abundance of species within and the species diversity of a community can be explained through neutral drift of individual species abundances.

$$\text{Neutral - theory}\ \phi n = \theta + \frac{J!}{n!(J-n)!}\frac{r(y)}{r(J+y)!}\int_0^y \frac{r(n+y)}{r(1+y)}\frac{r(J-n+y-y)}{r(y-y)}\exp\left(\frac{y\theta}{dy}\right)$$

Niche-preemption model was proposed by (Motomura 1932) and assumes that the percentage of the total niche occupied by the first species is α, the second species occupied a percentage α of the reminder, (1 –a), and so on.

Niche-preemption (a^r) = (1 −a) −1

a^r is the expected abundance of species at rank r, *S* is the number of species, *N* is the number of individuals, Φ is a standard normal function, p^1 is the estimated proportion of the most abundant species, and α, σ, γ, β and *c* are the estimated parameters in each model. In neutral-theory model, where Γ(z) = ∫ t∞ z-1 0 e-t dt which is equal to (z-1)!, for integer z and γ = m(J-1) 1-m, θ is fundamental diversity number, *m* is migration rate.

Relative abundance: Abundance of a particular species relative to total number of individuals of all species in the sampled area is relative abundance (RA).

$$RA=\frac{\text{Abundance of a species}}{\text{Sum of the abundance of all species}}\times 100$$

Basal area: Total ground area occupied by a tree is its basal area (BA) which is estimated by measuring its diameter at breast height (1.37 m). BA of a tree species gives an idea on the proportion of its dominance in the community along with its relative size, volume and biomass. Basal area is calculated by the following formula (Chauhan et al 2009).

$BA = 0.7854 \times (DBH)^2$

Importance value index: Importance value index (IVI) reflects the sociological structure of a species in a community as it indicates its importance in the community. The summation of RF, RD and RA of a species is its IVI (Curtis 1959, Kershaw 1973).

RESULTS AND DISCUSSION

Vegetation analysis: Overall plant species enlisted for the large cardamom based traditional agroforestry systems in the Darjeeling Himalayas was 130 species included 37 tree species 25 shrubs species, 46 herbs species, eight ferns species, 11 climbers species and three orchids species (Table 1, 2). Low- (700-1200 m asl), mid- (1200-1700 m asl) and high- (< 1700 m asl) altitude class was documented with 76 plant species, 60 species and 52 species. The overall range of plant species richness range recorded was 8-50 species with 16-50 species at low altitude class, 8-30 species

Table 1. Relative frequency and relative density of plant species in large cardamom based traditional agroforestry systems

Plant species	RF				RD			
	O_a	L	M	H	O_a	L	M	H
Acalypha accedens	0.7	-	2.1	-	0.29	-	1	-
Acmella uliginosa	1	2.3	-	-	1.33	2.9	-	-
Adiantum capillus	1.3	2.9	4.5	-	2.29	4.9	7.5	-
Ageratina adenophora	4.2	5.4	2.9	3.1	2.7	4.3	1.1	1.29
Ageratum conyzoides	2.9	3.2	1.6	4.14	3.25	3.1	0.7	5.74
A. houstonianum	2.6	2	3.7	2.07	2.28	1.9	2	2.93
Albizia odoratissima	0.3	0.5	-	-	0.24	0.5	-	-
A. procera	0.3	0.6	-	-	0.15	0.3	-	-
Alnus nepalensis	0.5	0.3	0.4	0.72	0.67	0.2	1.2	0.94
Alsophila dregei	0.3	0.6	-	-	0.03	0.1	-	-
Amomum subulatum	7.8	5.7	8.2	10.3	4.77	3.4	5.7	5.5
Anaphalis triplinervis	1	-	-	4.14	1.86	-	-	6.67
Ardisiamacrocarpa	0.2	-	-	0.83	0.09	-	-	0.31
Artemesia vulgaris	1.6	2.3	1.6	-	0.89	1.6	0.5	-
Begonia palmata	1	-	-	4.14	0.44	-	-	1.6
B. tessaricarpa	0.9	-	-	3.62	0.38	-	-	1.37
Biden pilosa	1.6	3.4	-	-	2.43	5.2	-	-
Bischofia javanica	0.2	-	0.7	-	0.09	-	0.3	-
Boehmeria cylindrica	0.4	0.9	-	-	0.14	0.3	-	-
B. platyphylla	0.5	0.6	0.8	1.03	0.73	0.9	0.5	1.13
Brachiaria reptans	2.9	2.9	1.6	4.14	4.06	3.3	2.1	6.91
Brassaiopsis mitis	0.1	-	0.3	-	0.17	-	0.6	-
Brugmansia suaveolens	0.1	0.2	-	-	0.07	0.1	-	-
C. tetrandra	0.2	0.3	-	-	0.08	0.2	-	-
Carex sylvatica	0.9	-	-	3.62	1.48	-	-	5.31
Castanopsis indica	0.2	0.5	-	-	0.13	0.3	-	-
Cayratia geniculate	0.3	0.7	-	-	0.17	0.4	-	-
Centella asiatica	1.8	-	2.5	4.14	3.16	-	3.7	7.53
Citrus spp	0.2	0.3	-	-	0.1	0.2	-	-
Colebrookea oppositifolia	0.3	-	1	-	0.68	-	2.4	-
Commelina suffruticosa	0.4	-	-	1.55	0.18	-	-	0.66
Croton caudatus	0.2	0.3	-	-	0.39	0.8	-	-
Cryptomeria japonica	0.6	0.3	1	0.72	0.76	0.1	1.6	0.86
Cupressuscashmeriana	0.5	0.3	0.3	1.03	0.52	0.3	0.3	1.13
Dichroa febrifuga	0.7	0.6	2.1	1.55	0.27	0.2	2.7	0.59
Diplazium esculentum	2.6	0.6	2.1	6.72	4.3	1.3	2.8	10.4
Drymaria cordata	2.7	1.7	2.9	4.14	4.12	2.9	3.7	6.13
Drypetes lancifolia	0.2	0.3	-	-	0.2	0.4	-	-
Equisetum arvense	0.7	1.4	-	-	0.26	0.6	-	-
E. debile	1.8	2.3	2.5	-	1.48	1.7	2.4	-
Erythrina variegate	0.4	0.5	0.5	-	0.22	0.2	0.4	-
Euonymus attenuatus	0.2	-	-	0.31	0.17	-	-	0.62

Cont...

Table 1. Relative frequency and relative density of plant species in large cardamom based traditional agroforestry systems

Plant species	RF				RD			
	O_a	L	M	H	O_a	L	M	H
Exbucklandia populnea	0.1	-	0.3	-	0.04	-	0.2	-
Ficus auriculata	0.4	0.8	-	-	0.41	0.9	-	-
F. lacor	0.3	0.6	-	-	0.25	0.5	-	-
F. semicordata	0.6	0.8	0.7	-	0.42	0.5	0.6	-
F. spp.	0.2		0.6		0.09		0.3	-
Firmiana colorata	0.3	0.4	0.5	-	0.38	0.6	0.4	-
Floscopa scandens	0.9	2	-	-	1.3	2.8	-	-
Fragaria nubicola	2.2	1.3	2.5	-	1.97	2.8	2.4	-
Girardinia diversifolia	0.2	-	-	0.62	0.12	-	-	0.43
G. palmata	0.5	0.1	0.5	1.03	0.37	0.1	0.1	1.05
Goodyera oblongifolia	0.7	1.4	-		0.35	0.7	-	-
Gynocardia odorata	0.1	-	0.4	-	0.1	-	0.3	-
Gynura cusimbua	0.2	-	-	1.55	0.16	-	-	0.59
Hydrocotyle javanica	0.5	-	-	2.07	0.85	-	-	3.04
Hydrocotyle nepalensis	0.5	-	1.6	-	0.27	-	0.9	-
Hypoestes phyllostachya	0.5	1.2	-	-	0.56	1.2	-	-
Juniperus indica	0.3	0.7	-	-	0.21	0.4	-	-
Justicia prostrata	1.3	0.9	2.9	-	0.2	0.1	0.5	-
Lantana camara	0.6	0.7	1	-	0.93	1	1.5	-
Laportea bulbifera	0.1	-	-	0.41	0.13	-	-	0.47
Lepidagathis incurva	1.6	1.4	2.5	-	1	1	1.8	-
Leucosceptrum canum	0.2	-	1	-	0.11	-	1.5	-
Lindenbergia grandiflora	1.2	1.2	2.1	-	0.81	0.8	1.6	-
Litsea glutinosa	0	-	-	0.1	0.01	-	-	0.04
L. monopetala	0.2	0.5	-	-	0.28	0.6	-	-
Lycopodium japonicum	2.2	2.3	3.7	-	2.18	2.1	4.1	-
Lygodium flexuosum	0.1	-	-	0.41	0.03	-	-	0.12
Macaranga denticulata	0.4	0.5	0.6	-	0.3	0.3	0.6	-
Machilus edulis	0.1	-	-	0.52	0.08	-	-	0.27
Magnolia doltsopa	0.1	-	-	0.21	0.03	-	-	0.12
M. grandiflora	0.1	-	-	0.31	0.03	-	-	0.12
M. lanuginosa	0.2	-	-	0.83	0.1	-	-	0.35
Mallotus tetracoccus	0.1	-	0.4	-	0.07	-	0.2	-
Matteuccia struthiopteris	2.2	2.3	3.7	-	1.12	1.2	1.9	-
Melastoma malabathricum	0.1	-	-	0.41	0.09	-	-	0.31
Mikania micrantha	0.4	0.9	-	-	0.12	0.3	-	-
Myrsine semiserrata	0.3	-	0.8	-	0.36	-	1.2	-
Nephrolepsis cordifolia	2.9	3.2	4.5	-	3.04	3.1	5.4	-
Oenanthe thomsonii	1	-	3.3	-	0.75	-	2.6	-
Oroxylum indicum	0.3	0.7	-	-	0.15	0.3	-	-
Ostodes paniculata	0.1	0.3	-	-	0.08	0.2	-	-
Oxalis corniculata	1.6	3.4	-	-	2.44	5.2	-	-
O. latifolia	0.7	1.4	-	-	0.5	1.1	-	-
O. martiana	1.4	3.2	-	-	2.21	4.7	-	-

Cont...

Table 1. Relative frequency and relative density of plant species in large cardamom based traditional agroforestry systems

Plant species	RF				RD			
	O_a	L	M	H	O_a	L	M	H
Peliosanthes griffithii	0.5	-	-	3.1	0.27	-	-	0.47
Persicaria capitata	0.5	1.2	-	-	0.61	1.3	-	-
P. chinesis	0.2	0.5	0.7	-	0.61	1.3	1.1	-
Phaulopsis dorsiflora	0.7	1.4	-	-	0.49	1	-	-
Phlogacanthus thyrsiformis	0.5	0.6	0.8	1.03	0.47	0.6	0.7	0.98
Pilea cordifolia	0.5	1.2	-	-	0.38	0.8	-	-
P. involucrate	0.1	-	-	0.41	0.16	-	-	0.59
P. melastomoides	0.2	-	-	0.83	0.4	-	-	1.44
P. nummulariifolia	0.5	-	1.6	-	0.94	-	3.2	-
Pinus wallichiana	0.2	-	-	0.72	0.12	-	-	0.43
Piper attenuatum	0.2	0.5	-	-	0.18	0.4	-	-
P. boehmeriaefolium	0.7	0.7	0.7	0.83	0.67	0.5	0.8	0.86
P. peepuloides	0.3	0.6	-	-	0.29	0.6	-	-
Plantago asiatica	1.2	-	2.1	2.07	0.61	-	1.4	0.7
P. major	0.4	-	-	1.55	0.09	-	-	0.31
Plumbagoauriculata	0.5	-	1.6	-	0.44	-	1.5	-
Pogostemon andersonii	0.5	-	1.6	-	0.71	-	2.4	-
Polygonum rude	0.2	-	0.5	-	0.17	-	0.6	-
Pouzolzia zeylanica	0.8	1.7	-	-	0.76	1.6	-	-
Prunus cerasoides	0.1	-	0.3	-	0.08	-	0.3	-
Rhododendron griffithianum	0.2	-	-	0.72	0.08	-	-	0.27
Rubia cordifolia	0.1	-	0.3	-	0.07	-	0.2	-
Rubus diffusus	0.3	-	1	-	0.34	-	1.2	-
R. holosericeus	0.2	-	-	0.62	0.17	-	-	0.62
R. spp	0.2	-	0.5	-	0.04	-	0.2	-
Schima wallichii	0.7	0.7	0.6	0.83	0.81	1	0.7	0.59
Scutellarialateriflora	0.4	-	-	1.55	0.18	-	-	0.66
Selaginella denticulata	6.1	5.7	5.7	6.72	10.5	9.9	8.5	12.3
Senecio densiflorus	0.3	-	-	1.03	0.37	-	-	1.33
Smilax ovalifolia	0.3	-	-	1.24	0.13	-	-	0.47
Solena amplexicaulis	0.3	0.6	-	-	0.13	0.3	-	-
Spathoglottius plicata	0.9	0.9	-	2.07	0.52	0.5	-	0.98
Stephaniajaponica	0.4	0.9	-	-	0.51	1.1	-	-
Strobilanthes exserta	1.3	1.2	2.1	-	1.05	0.9	2.1	-
Synedrella nudiflora	0.7	1.4	-	-	0.2	0.4	-	-
Terminalia myriocarpa	0.5	0.6	0.4	0.31	0.41	0.5	0.4	0.27
Tetrastigma serrulatum	0.3	-	-	1.03	0.26	-	-	0.94
Thujaplicata	0.1	-	-	0.21	0.02	-	-	0.08
Thysanolaena latifolia	0.5	1.4	-	-	0.3	0.9	-	-
Toona ciliata	0.4	0.6	0.4	-	0.3	0.5	0.3	-
Vitex negundo	0.4	0.8	-	-	0.09	0.2	-	-
Vitis pedata	0.5	1.2	-	-	0.3	0.7	-	-
Zanthoxylum piperitum	0.2	-	0.5	-	0.08	-	0.3	-
Zephyranthes carinata	1.6	-	2.9	2.59	1.64	-	4.5	1.17

RF- Relative frequency; RD- Relative density; Oa- overall (700-1930 m asl); L- low (700-1200 m asl); M- mid (1200-1700 m asl); H- high (> 1700 m)

Table 2. Relative abundance and importance value index of plant species in large cardamom based traditional agroforestry systems

Plant species	RA				IVI			
	O_a	L	M	H	O_a	L	M	H
Acalypha accedens	0.1	-	0	-	1.07	-	3.08	-
Acmella uliginosa	0.4	0.6	-	-	2.73	5.7	-	-
Adiantum capillus	0.5	0.8	0.1	-	4.08	8.53	12.1	-
Ageratina adenophora	0.2	0.4	0	0.8	7.04	10.1	4	5.21
Ageratum conyzoides	0.3	0.4	0	1.1	6.43	6.73	2.33	11
A. houstonianum	0.2	0.4	0	0.6	5.12	4.3	5.72	5.54
Albizia odoratissima	0.2	0.4	-	-	0.77	1.47	-	-
A. procera	0.2	0.2	-	-	0.59	1.19	-	-
Alnus nepalensis	0.4	0.3	0.2	0.2	1.54	0.7	1.71	1.85
Alsophila dregei	0	0.1	-	-	0.33	0.7	-	-
Amomum subulatum	0.2	0.3	0	2.8	12.7	9.41	13.9	18.6
Anaphalis triplinervis	0.5	-	-	1.1	3.39	-	-	11.9
Ardisiamacrocarpa	0.1	-	-	0.2	0.41	-	-	1.36
Artemesia vulgaris	0.2	0.3	0	-	2.61	4.23	2.1	-
Begonia palmata	0.1	-	-	1.1	1.6	-	-	6.84
B. tessaricarpa	0.1	-	-	1	1.41	-	-	5.95
Biden pilosa	0.4	0.7	-	-	4.42	9.31	-	-
Bischofia javanica	0.1	-	0	-	0.41	-	0.98	-
Boehmeria cylindrica	0.1	0.2	-	-	0.63	1.32	-	-
B. platyphylla	0.4	0.7	0	0.3	1.64	2.14	1.29	2.44
Brachiaria reptans	0.4	0.5	0.1	1.1	7.31	6.62	3.83	12.1
Brassaiopsis mitis	0.6	-	0.1	-	0.87	-	0.97	-
Brugmansia suaveolens	0.2	0.3	-	-	0.34	0.64	-	-
C. tetrandra	0.1	0.2	-	-	0.37	0.72	-	-
Carex sylvatica	0.5	-	-	1	2.84	-	-	9.89
Castanopsis indica	0.2	0.3	-	-	0.51	1.01	-	-
Cayratia geniculate	0.2	0.2	-	-	0.64	1.3	-	-
Centella asiatica	0.5	-	0.1	1.1	5.46	-	6.18	12.8
Citrus spp	0.2	0.3	-	-	0.43	0.82	-	-
Colebrookea oppositifolia	0.6	-	0.1	-	1.6	-	3.46	-
Commelina suffruticosa	0.1	-	-	0.4	0.71	-	-	2.63
Croton caudatus	0.7	1.1	-	-	1.24	2.27	-	-
Cryptomeria japonica	0.3	0.2	0.1	0.2	1.72	0.64	2.63	1.77
Cupressus cashmeriana	0.3	0.4	0.1	0.3	1.31	0.98	0.56	2.44
Dichroa febrifuga	0.1	0.2	0.1	0.4	1.04	0.99	4.8	2.55
Diplazium esculentum	0.5	1	0.1	1.8	7.36	2.8	4.95	18.9
Drymaria cordata	0.4	0.8	0.1	1.1	7.27	5.35	6.62	11.4
Drypetes lancifolia	0.4	0.5	-	-	0.7	1.3	-	-
Equisetum arvense	0.1	0.2	-	-	1.02	2.16	-	-
E. debile	0.2	0.3	0.1	-	3.52	4.29	4.89	-
Erythrina variegate	0.2	0.2	0	-	0.75	0.92	0.9	-
Euonymus attenuatus	0.3	-	-	0.1	0.64	-	-	1.02

Cont...

Table 2. Relative abundance and importance value index of plant species in large cardamom based traditional agroforestry systems

Plant species	RA				IVI			
	O_a	L	M	H	O_a	L	M	H
Exbucklandia populnea	0.1	-	0	-	0.26	-	0.5	-
Ficus auriculata	0.3	0.5	-	-	1.09	2.18	-	-
F. lacor	0.3	0.4	-	-	0.78	1.52	-	-
F. semicordata	0.2	0.3	0.1	-	1.2	1.6	1.34	-
F. spp.	0.1	-	0	-	0.4	-	0.9	-
Firmiana colorata	0.3	0.6	0	-	1.03	1.58	0.94	-
Floscopa scandens	0.4	0.6	-	-	2.61	5.41	-	-
Fragaria nubicola	0.3	1	0.1	-	4.43	5	4.85	-
Girardinia diversifolia	0.2	-	-	0.2	0.49	-	-	1.21
G. palmata	0.2	0.4	0	0.3	1.06	0.57	0.61	2.36
Goodyera oblongifolia	0.2	0.2	-		1.15	2.41	-	-
Gynocardia odorata	0.2	-	0	-	0.44	-	0.79	-
Gynura cusimbua	0.3	-	-	0.4	0.61	-	-	2.55
Hydrocotyle javanica	0.5	-	-	0.6	1.82	-	-	5.66
Hydrocotyle nepalensis	0.1	-	0	-	0.94	-	2.6	-
Hypoestes phyllostachya	0.3	0.5	-	-	1.39	2.82	-	-
Juniperus indica	0.2	0.3	-	-	0.7	1.42	-	-
Justicia prostrata	0	0.1	0	-	1.54	1	3.4	-
Lantana camara	0.4	0.7	0.1	-	1.97	2.41	2.59	-
Laportea bulbifera	0.4	-	-	0.1	0.58	-	-	0.99
Lepidagathis incurva	0.2	0.3	0	-	2.74	2.8	4.24	-
Leucosceptrum canum	0.2	-	0.1	-	0.46	-	2.59	-
Lindenbergia grandiflora	0.2	0.3	0	-	2.18	2.21	3.65	-
Litsea glutinosa	0.1	-	-	0	0.15	-	-	0.17
L. monopetala	0.4	0.6	-	-	0.87	1.65	-	-
Lycopodium japonicum	0.3	0.4	0.1	-	4.67	4.84	7.8	-
Lygodium flexuosum	0.1	-	-	0.1	0.22	-	-	0.64
Macaranga denticulata	0.2	0.3	0.1	-	0.92	1.08	1.18	-
Machilus edulis	0.2	-	-	0.1	0.37	-	-	0.93
Magnolia doltsopa	0.2	-	-	0.1	0.26	-	-	0.38
M. grandiflora	0.1	-	-	0.1	0.23	-	-	0.51
M. lanuginosa	0.1	-	-	0.2	0.44	-	-	1.4
Mallotus tetracoccus	0.1	-	0	-	0.33	-	0.66	-
Matteuccia struthiopteris	0.1	0.2	0	-	3.47	3.73	5.61	-
Melastoma malabathricum	0.2	-	-	0.1	0.42	-	-	0.84
Mikania micrantha	0.1	0.1	-	-	0.62	1.3	-	-
Myrsine semiserrata	0.4	-	0.1	-	1	-	2.13	-
Nephrolepsis cordifolia	0.3	0.4	0.1	-	6.19	6.7	10	-
Oenanthe thomsonii	0.2	-	0	-	1.99	-	5.89	-
Oroxylum indicum	0.2	0.2	-	-	0.59	1.26	-	-
Ostodes paniculata	0.2	0.3	-	-	0.37	0.7	-	-
Oxalis corniculata	0.4	0.7	-	-	4.44	9.34	-	-
O. latifolia	0.2	0.3	-	-	1.36	2.83	-	-
O. martiana	0.4	0.7	-	-	4.07	8.56	-	-

Cont...

Table 2. Relative abundance and importance value index of plant species in large cardamom based traditional agroforestry systems

Plant species	RA				IVI			
	O_a	L	M	H	O_a	L	M	H
Peliosanthes griffithii	0.1	-	-	0.8	0.94	-	-	4.39
Persicaria capitata	0.3	0.5	-	-	1.45	2.95	-	-
P. chinesis	0.8	1.3	0.1	-	1.63	3.03	1.86	-
Phaulopsis dorsiflora	0.2	0.3	-	-	1.35	2.8	-	-
Phlogacanthus thyrsiformis	0.3	0.5	0	0.3	1.24	1.61	1.53	2.28
Pilea cordifolia	0.2	0.3	-	-	1.1	2.27	-	-
P. involucrate	0.4	-	-	0.1	0.7	-	-	1.11
P. melastomoides	0.5	-	-	0.2	1.15	-	-	2.49
P. nummulariifolia	0.5	-	0.1	-	1.97	-	4.98	-
Pinus wallichiana	0.2	-	-	0.2	0.48	-	-	1.35
Piper attenuatum	0.3	0.4	-	-	0.64	1.24	-	-
P. boehmeriaefolium	0.3	0.3	0.1	0.2	1.66	1.45	1.46	1.91
P. peepuloides	0.3	0.5	-	-	0.87	1.69	-	-
Plantago asiatica	0.1	-	0	0.6	1.92	-	3.5	3.32
P. major	0.1	-	-	0.4	0.54	-	-	2.28
Plumbagoauriculata	0.2	-	0.1	-	1.2	-	3.21	-
Pogostemon andersonii	0.4	-	0.1	-	1.6	-	4.14	-
Polygonum rude	0.3	-	0.1	-	0.64	-	1.15	-
Pouzolzia zeylanica	0.3	0.4	-	-	1.81	3.77	-	-
Prunus cerasoides	0.2	-	0	-	0.38	-	0.63	-
Rhododendron griffithianum	0.1	-	-	0.2	0.37	-	-	1.19
Rubia cordifolia	0.2	-	0	-	0.34	-	0.59	-
Rubus diffusus	0.3	-	0.1	-	0.95	-	2.2	-
R. holosericeus	0.3	-	-	0.2	0.64	-	-	1.41
R. spp	0.1	-	0	-	0.28	-	0.66	-
Schima wallichii	0.3	0.6	0.1	0.2	1.84	2.26	1.35	1.63
Scutellarialateriflora	0.1	-	-	0.4	0.71	-	-	2.63
Selaginella denticulata	0.5	0.8	0.1	1.8	17.1	16.4	14.3	20.8
Senecio densiflorus	0.4	-	-	0.3	1.02	-	-	2.64
Smilax ovalifolia	0.1	-	-	0.3	0.56	-	-	2.04
Solena amplexicaulis	0.1	0.2	-	-	0.53	1.07	-	-
Spathoglottius plicata	0.2	0.3	-	0.6	1.59	1.67	-	3.59
Stephaniajaponica	0.3	0.5	-	-	1.27	2.54	-	-
Strobilanthes exserta	0.2	0.3	0.1	-	2.58	2.37	4.22	-
Synedrella nudiflora	0.1	0.1	-	-	0.93	1.98	-	-
Terminalia myriocarpa	0.2	0.4	0.1	0.1	1.13	1.44	0.83	0.67
Tetrastigma serrulatum	0.3	-	-	0.3	0.8	-	-	2.25
Thujaplicata	0.1	-	-	0.1	0.19	-	-	0.34
Thysanolaena latifolia	0.2	0.3	-	-	0.99	2.65	-	-
Toona ciliata	0.2	0.4	0	-	0.91	1.44	0.7	-
Vitex negundo	0.1	0.1	-	-	0.52	1.09	-	-
Vitis pedata	0.2	0.3	-	-	0.99	2.05	-	-
Zanthoxylum piperitum	0.1	-	0	-	0.37	-	0.78	-
Zephyranthes carinata	0.3	-	0.1	0.7	3.49	-	7.46	4.44

RF- Relative frequency; RD- Relative density; Oa- overall (700-1930 m asl); L- low (700-1200 m asl); M- mid (1200-1700 m asl); H- high (> 1700 m) richness at mid-altitude class and 9-21 species at high altitude class. The frequency of species irrespective of altitude classes documented was 1.7-100.0. The most frequent species was *Amomum subulatum* and the least frequent species was *Litsea glutinosa* while *Amomum subulatum* and *Selaginella denticulata* (100.0 each) were the most frequent species at low-altitude. At mid-altitude, the most frequent species was *Amomum subulatum* (100.0) and the least frequent species was *Rubia cordifolia* (10.0). At high-altitude class *Amomum subulatum* was the most frequent species with frequency of 100.0 and the least frequent species class was *Litsea glutinosa* (5.0 each).The species documented with higher occupancy status i.e. frequency exhibited wider spatial distribution at multiple altitudinal levels were widely representing biodiversity across a broad landscape (Gbedomon et al 2017, Sarkar 2020). Overall, these systems across the altitudinal gradient with varying frequencies but multi-strata vegetation with herbs, shrubs and trees were contributing heterogeneity with temporal and spatial dynamism. Moreover, these Darjeeling Himalayan forest based below canopy large cardamom farming systems being close to natural forest ecosystems due to minimal disturbance can be considered as efficient and viable systems of biodiversity conservation intrinsically similar to its traditional homegardens (Sarkar 2020) and traditional homegardens elsewhere (Cruz-Garcia and Struik 2015, Gbedomon et al 2017). The species richness of 130 documented from these large cardamoms based traditional agroforestry farming systems of Darjeeling Himalayas can be sufficient enough to represent regional species richness (van der Wal and Bongers 2013) and thus are important to conserve regional plant diversity *in situ* (Galluzzi et al 2010, Sarkar 2020).

The overall density estimated for the documented species in Darjeeling Himalayan large cardamom based TAFs was in the range of 0.01-3.23 quadrate^{-1}. The densest species was *Selegnella denticulata* while, *Lygodium flexuosum* and *Alsophila dregei* were sparse. The plant density estimated for low-, mid- and high-altitude class was 0.03-4.26 (*Alsophila dregei-Selegnella denticulata*), 0.06-2.30 (*Rubia cordifolia- Selegnella enticulata*) and 0.03-3.16 (*Lygodium flexuosum-Selegnella denticulata*), respectively while, density of *Amomum subulatum* at these classes were 1.47, 1.50 and 1.41, respectively. Overall, abundance of species was in the range of 0.30-7.0. The least abundant species was *Alsophila dregei* and the most abundant species found was *Persicaria chinensis* while, for large cardamom the value was 1.47. The abundance for low-, mid- and high-altitude class was 0.27-7.0 (*Justicia prostrata-Persicaria chinensis*), 0.40-6.20 (*Justicia prostrata-Alnus nepalensis*) and 0.40-5.33 (*Peliosanthes griffithii-Euonymus attenuates).*

The important value index (IVI) of the documented species at the studied systems varied from 0.15-17.11 irrespective of altitude classes (Table 1, 2). IVI estimated for large cardamom was 12.74 while, at low-, mid- and high-altitude class it was 9.41, 13.88 and 18.59 respectively. Overall, the most important species based on IVI was a fern *Selaginella denticulata* followed by an herb *Amomum subulatum*. The least important species based on IVI was a tree *Litsea glutinosa* (0.15). The IVI of species estimated for low-altitude class was 0.57-16.39. The most important species based on IVI at this altitude was a fern *Selaginella denticulata* and the least important was a shrub *Girardinia palmata* (0.57). At mid-altitude class the IVI estimated was in the range of 0.50 (*Exbucklandia populnea*)-14.30 (*Selaginella denticulata*). The most important species based on IVI at this altitude found was a fern and the least important species was a tree (0.50). Similarly, IVI estimated for high-altitude class 0.17 (*Litsea glutinosa*)-20.84 (*Selaginella denticulata*). The plant assemblages in the large cardamom based traditional at low-, mid- and high-altitude class were more or less similarly distributed. Species with higher IVI signifies its ecological importance in the system. The species growing at a particular altitude in the Darjeeling Himalayas was primarily influenced by site factors and ecological conditions that they best adapted the natural selection and evolution . Similar vegetation analysis with comparable density, abundance, frequency and IVI of traditional agroforestry systems was reported in earlier studies also (Taran and Deb 2019, Sarkar 2020). Phyto-sociology of the plant communities varies with agroforestry systems, slope and land quality (Sharma et al 2010).

Model: The distribution of plant species in the large cardamom based traditional agroforestry systems was evaluated using Log-normal, Mandelbrot, Zipf, Null and Preemption species distribution models (SAM). The Preemption SDM give best fit to the data with highest Akaike Information Criterion (AIC) score (Fig. 2).Similarly, distribution of species using these species distribution models was also evaluated for the homegardens of Darjeeling Himalayas across the altitudinal gradient (Sarkar 2020). Among the models SAD was unable to distinguish the species with any degree of certainty due to limited information (Volkov et al 2005). Therefore, it was recommended that the models should be evaluated for their ability to simultaneously explain multiple macro-ecological data in order to obtain sufficient information on ecological processes (Xiao et al 2015).

The conservation status of the Darjeeling large cardamom based traditional agroforestry can be

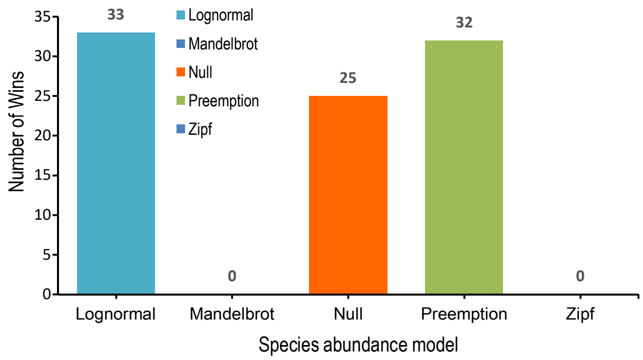

Fig. 2. Number of cases model provided best fit to plant species abundance data

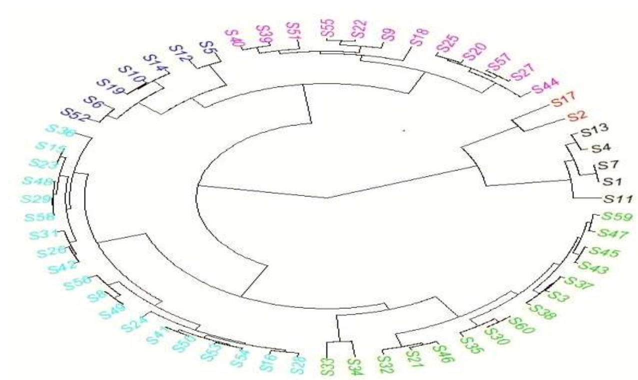

Fig. 3. Clustering based on plant species richness and population of the Darjeeling Himalayan large cardamom based traditional agroforestry systems

satisfactorily recognised only when its ecosystem services are fully realised in addition to factors affecting its spatial distribution, evolution and temporal resiliency understood and regularly monitored for any evolutionary changes there in (Agbogidi and Adolor 2013). These traditional large cardamom-based agroforestry systems of Darjeeling Himalayas are also diverse plant species with variable population size across the altitudinal gradient over reasonably wider landscape like many other traditional agroforestry systems (Abebe et al 2013, Gebrewahid and Meressa 2020) including its home gardens also (Sarkar 2020). Based on species richness and its plant population across the altitudinal gradient, large cardamom based traditional agroforestry farming systemswere grouped into six clusters illustrated with six different colors (Fig. 3). Similar cluster analysis of homegardens across the altitudinal gradient in Central (Vibhuti et al 2018) and Darjeeling (Sarkar 2020) Himalayas, India were also reported. These traditional agroforestry systems as are very close to natural ecosystems with potential to offer variety of ecosystem services from provisional to cultural services like NTFPs, biodiversity conservation, water regulation and purification, biomass production, carbon sequestration, nutrient cycling and socio-cultural service for the well-being of the society (Nath et al 2016, Vineeta et al 2021).

CONCLUSION

The heterogeneity of the system can be attributed to richness of 130 plant species include 37 were tree species, 25 shrub species, 46 herb species; eight fern species, 11 climber species and three orchid species. Overall, the most important species based on IVI was a fern *Selaginella denticulata* followed by an herb *Amomum subulatum*. The least important species based on IVI was a tree *Litsea glutinosa* (0.15). The farming systems also varied on the basis of their species richness and plant population which were grouped into six clusters. Based on the distribution of plant species in the system five species distribution models (SDM) were evaluated and the Preemption SDM was provided best fit to the data with highest Akaike Information Criterion (AIC) score. This study recommends further in-depth analysis of structure and composition with respect to altitudinal location will generate more information on functional diversity, structure, composition and productivity.

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Changes in Soil Physico-Chemical Properties in different Land Use Systems of Manipur, Northeast India

Ng. Polbina Monsang, Ngangbam Somen Singh, Keshav Kumar Upadhyay and Shri Kant Tripathi*

Department of Forestry, Mizoram University, Tanhril Aizawl-796 004 India
**E-mail: sk_tripathi@rediffmail.com*

Abstract: The current study examined the soil physico-chemical properties of five different land uses (e.g. Mixed pine forest, MPF; Pine plantation, PP; *Lithocarpus* forest, LF; *Quercus* forest, QF; and *Dipterocarpus* forest, DF) in Manipur, India. Replicated soil samples from three soil depths (i.e., 0-10 cm, 10-20 cm, and 20-30 cm) were collected from each land use type and analyzed for soil physical and chemical properties. Sand percentage was highest in DF and lowest in QF. The soil pH varied between 4.3 and 5.3 in different land uses. The highest water holding capacity (84.86%) was in PP and lowest (55.46%) in LF. The bulk density was highest in LF and QF (1.04 g/cm^3) and lowest in DF (0.85 g/cm^3). The highest soil organic carbon and organic matter were in PP (30.6 and 59.5 Mg/ha) and lowest in MPF (6.6 and 11.2 Mg/ha). The stocks of nitrogen, phosphorus and potassium in different systems ranged 250.5-438.7 kg/ha, 12.7-54.8 kg/ha and 102-236.20 kg/ha, respectively. Higher accumulation of soil organic carbon and nutrients in PP within a short period of time (40 years) compared to others are because slow decomposition of pine needle.

Keywords: Land use, Soil physico-chemical properties, Mixed pine forest, Pine plantation, Northeast India

Land use change is among the major global change processes responsible for affecting the structure and functioning of natural and modified ecosystems (Tripathi et al 2008, Wapongnungsang et al 2018). Soil physico-chemical properties are important determinants of the structure and functioning of different land uses, which are strongly influenced by the biota through recycling of organic matter and nutrients (Singha et al, 2020, Manpoong et al 2020). Soil contains organic matter, minerals, water, air and micro-organisms that determine the physico-chemical characters and provide a natural environment for the growth of terrestrial plants and animals (Velayutham and Bhattacharyya 2000, Singh et al 2020a, Singh et al 2021a, b). Soil and micro-fauna provide several environmental services such as soil erosion control, pest control, depletion of greenhouse gases, pollutants, improve soil composition and help retain nutrient (Kibblewhite et al 2008, Baer and Birgé 2018) thus keep nature in balance. However, vegetation has also affected soil compaction and cycling via litter fall, organic matter, nutrient recycling, weathering process, erosion, etc. (Binkley et al 1992, Nkongolo and Plassmeyer 2010, Singh and Tripathi 2020a, b). Decomposition of litter and presents of organic matter affects soil formation and fertility by adding humus and nutrients to the soil (Singh et al 2022).

Plants are major source of organic matter and are essential elements for a healthy forest environment. In addition, plant species composition has contributed to forest ecosystems to maintain soil organic matter and to support the biogeochemical cycle which in turn disrupts soil structures through rooting, nutrient uptake, and root growth (Marcet et al 2006, Akintola et al 2020). Tropical forest has the potential to sequester 1.1 to 1.8 Gt C per year in 50 years (Makundi et al 1998). However, carbon sequestration potential of these forests is strongly influenced by deforestation for commercialization, conversion to plantation and other land use types, development activities and climate change (Foley et al 2005, Yang et al 2009).

Degradation of the forest ecosystem as a result of anthropogenic activities has been well reported in several studies (Devi and Yadava 2015, Tripathi et al 2016, 2017, Singh et al 2020a, b). This has led to increase in soil erosion, nutrient loss, decreased soil microbes which are responsible for soil structure and quality (Slam and Weil 2000, Chen et al 2001, Singh and Tripathi 2020b). The varying soil structures due to changing vegetation and a strong relationship between vegetation and soil in the forest. Therefore, the physico-chemical properties of soils are important in determining soil production and the quality of the forest site that provide sustainable forest and ecological services. The main aim of the present study was to analyze the physico-chemical soil properties of different land use types and to understand the potential of different land use soils in sequestering carbon and nutrients in Manipur.

MATERIAL AND METHODS

Study sites: Manipur is in the Northeastern part of India between 23°50' N - 25 ° 42' N lat. and 92°59' E - 94°46' E long. with an area of 22,327 km^2 (ISFR 2019). The state has a tropical climate with moderate temperatures ranging from 14.5 °C to 38 °C and an average annual rainfall of 1200 to 2700 mm (ISFR 2019). There are five major forest types which were further sub-divided into 11 forest types. The present study was conducted in five different land uses under the three districts (Chandel, Senapati and Tengnoupal districts) of Manipur (Fig. 1). Mixed pine forest (MPF) and *Lithocarpus* forests (LF) were in the Chandel district. Pine plantation (PP) and *Quercus* forest (QF) were in the Senapati district and *Dipterocarpus* forest (DF) in the Tengnoupal district (Table 1).

Sampling and analysis of soil: Soils of the selected forests were collected randomly from nine locations and three depths (0-10, 10-20 and 20-30 cm) and three samples were composited to make one and kept in a well labeled zip polybags for analysis of its physico-chemical properties. These soils were sieved through 2 mm mesh and parted into fresh and air-dried soil. Soil texture was assessed through hydrometer method (Bouyoucos 1962) and the result was subsequently classified according to the United States Department of Agriculture (USDA) soil texture classification. Soil moisture content (SMC) was determined following a gravimetric method by oven drying the fresh soil samples at 105°C for 24 hours (Verstraeten et al 2008). Soil pH was measured using a digital pH meter with soil and water ratio of 1: 2.5 (Bandyopadhyay et al 2012). The water holding capacity (WHC) was measured using the Keen Raczkowski box process (Piper 1966). Bulk density (BD) of soil was measured using the stainless-steel tube of known inner as per the method Anderson and Ingram (1993). Soil organic carbon (SOC) was analyzed by Walkley and Black (1947). Soil organic matter (SOM) was estimated following Van Bemmelen factor 1.724. Available nitrogen (N) is estimated using the kjeldahl method (Jackson 1973), available phosphorus (P) was obtained by the Bray and Kurtz method (1945), and exchangeable potassium (K) was estimated using flame photometry method (Jackson 1973).

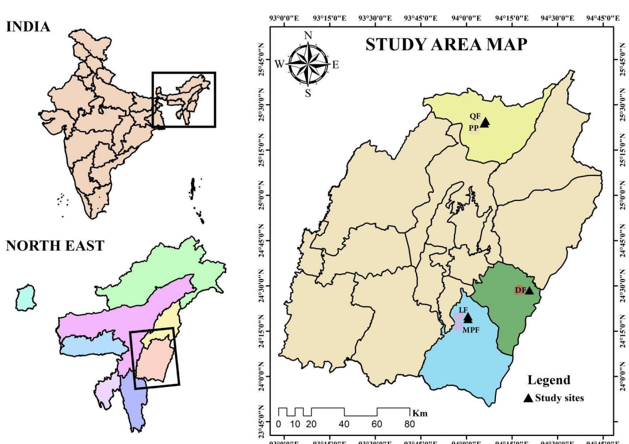

Fig. 1. Location map of the study area

Statistical analysis: Pearson coefficient (r) correlation was performed to assess significant differences among various soil physico-chemical properties. All analyses were conducted using SPSS statistics v18.0 software.

RESULTS AND DISCUSSION

In the present study, soil was sandy with sandy loam and loamy sand in texture (Table 2). In different land uses, the amount of sand, silt and clay ranged from 51.33 - 82.75%, 5.17 - 34.18% and 12.08 - 18.16%, respectively. Highest amount of sand, silt and clay contents were in *Dipterocarpus* forest, *Quercus* forest and *Lithocarpus* forest. The values of sand, silt and clay content of 70.90, 17.90 and 12.01%, respectively reported by Devi and Yadava (2015) in *Dipterocarpus tuberculatus* forest of Manipur were broadly comparable. Niirou et al (2015) also reported sandy soil type in different land use types in Manipur. Soil texture in different land uses is caused by movement and deposition of soil particles (Sand, silt and clay) due to rains and undulating site conditions at altitudinal gradients in forested lands (Saeed et al 2014).

Soil BD values ranged from 0.85 to 1.04 g/cm^3 in different land uses. Soil BD increase with soil depth (Table 2). In all study sites, the highest value was observed in the bottom soil layer (20-30cm) of *Lithocarpus* forest and *Quercus* forest. Niirou et al (2015) in various land use system in Senapati

Table 1. Location, age, GPS co-ordinates, elevations and districts of the study sites

Land use types	Age (Years)	Co-ordinate	Elevation (m amsl)	District
Mixed pine forest (MPF)	100	24°19`05``N, 94°00`31``E	1019	Chandel
Pine plantation (PP)	40	25°23`51``N, 94°05`52``E	1325	Senapati
Lithocarpus forest (LF)	100	24°19`58``N, 94°00`30``E	960	Chandel
Quercus forest (QF)	100	25°24`43``N, 94°06`15``E	1318	Senapati
Dipterocarpus forest (DF)	100	24°28`31``N, 94°21`04``E	536	Tengnoupal

district of Manipur also observed same trend. The soil BD was lowest in upper soil layer (0-10 cm) and highest in lower depths (20-30 cm). Low BD in upper soil layer is related to the presence of higher organic matter. The lowest BD (0.85 g/cm^3) was in *Dipterocarpus* forest in upper soil layer (0-10 cm). Higher organic matter has been reported lower the soil BD in different forest soils of Japan (Morisada et al 2004). Yinga et al (2020) reported a high amount of BD in the *Emblica* base agroforestry system due to a few crop coverings. In another study, the natural forest had a low amount of BD caused by higher accumulation of vegetation litters and the growth of densely populated roots reduces the soil BD of bamboo forest (Manpoong and Tripathi 2019).

Soil MC, an important regulator of plant growth, affected by the soil texture and organic matter contents. The decrease in soil moisture content with increasing soil depth in the present study is related to decreasing soil organic matter. Zheng et al (2015), reported decreasing soil moisture content with depths during the study period (June to September) in *Larix* spp and *Quercus mongolica* the forest. Authors argued that greater retention of soil moisture in *Q. mongolia* forest was due high soil organic matter and nutrients. The highest (37.25%) soil moisture was recorded in *Dipterocarpus* forest followed by Pine plantation and *Quercus* forest (having 34.95% MC), and the lowest (29.66%) in Mixed pine forest (Table 2). Variation in soil moisture was related to rainfall because precipitation and evapo-transpiration significantly recharged the soils water supply (Zheng et al 2015). Lesser ground vegetation and open canopy in Mixed pine forest may result in lower soil MC. Similar result was reported by Bargali et al (2018) where an open-bed pine chir forest had a lower MC.

WHC ranged from 55.5 to 84.9% with the highest WHC in pine plantation and lowest in *Lithocarpus* forest (Table 2), which may be due to the presence of high soil organic matter and higher content of clay and silt in the site. The higher content of organic carbon and clay contents in the soil have been reported to increase soil water holding capacity (Gupta et al 2010). The study sites were acidic in nature where soil pH ranged from 4.3 to 5.3 (Table 3), which broadly like the range (pH of 4.0 to 5.4) in different forest soils of Manipur (Sahoo et al 2020). The difference in soil pH value between the different land used types may be due to the kind of deposited organic debris (Mishra et al 2018).

In all study sites, the SOM content was very high in the topsoil layer (0-10 cm) which decreased with soil depth. The highest value of SOM (59.5 Mg/ha) was at the topsoil layer (0-10 cm) of pine plantation while the lowest (11.22 Mg/ha) at the bottom layer (20-30 cm) of mixed pine forest (Table 3). The current result is similar to the findings of Oladoye (2015).

Table 2. Depth wise soil physical properties from various forest types

Soil parameters	Depth (cm)	MPF	PP	LF	QF	DF
Sand (%)	0-10	64.67^a	62.03^a	63.33^a	58.33^a	82.75^a
	10-20	61^b	57^b	66^a	61^a	78.25^b
	20-30	59.67^b	62^{ac}	63^a	51.33^b	78.59^b
Silt (%)	0-10	22.84^a	25.57^a	18.51^a	27.34^a	5.17^a
	10-20	23.17^a	26.67^a	19.84^a	25.84^a	8.67^b
	20-30	23.67^a	22.84^a	21.67^a	34.18^b	6.83^{ab}
Clay (%)	0-10	12.49^a	12.50^a	18.16^a	14.33^a	12.08^a
	10-20	15.83^b	16.33^b	14.16^b	13.16^a	13.08^{ab}
	20-30	16.66^b	15.16^b	15.33^b	14.49^a	14.58^b
BD (g/cm^3)	0-10	0.95^a	0.89^a	0.92^a	0.90^a	0.85^a
	10-20	1.02^a	0.98^b	1.02^{ab}	0.95^{ab}	0.95^{ab}
	20-30	1.03^a	1.03^c	1.04^b	1.04^c	1.02^{bc}
MC (%)	0-10	29.66^a	34.95^a	31.61^a	34.95^a	37.25^a
	10-20	29.15^a	32.27^{ab}	26.71^b	33.19^{ab}	31.92^b
	20-30	29.07^a	28.64^c	25.05^{bc}	32.27^b	30.59^b
WHC (%)	0-10	61.26^a	84.86^a	59.33^a	60.47^a	58.08^a
	10-20	64.89^a	74.92^b	55.46^b	66.36^b	60.73^a
	20-30	63.19^a	76.51^b	57.41^a	65.85^b	68.08^b

Different superscript letter indicates significant differences ($P < 0.05$) among soil depth. (n-3, Mean ± 1SE. Abbreviation; MC-moisture content, WHC-water holding capacity and BD-bulk density. MPF=Mixed pine forest, PP=Pine plantation, LF=*Lithocarpus* forest, QF=*Quercus* forest, DF=*Dipterocarpus* forest

The decrease in the organic matter was due decline in the amount of litter (leaf and root) inputs in lower soil depths. Similarly, the amount of SOC was estimated to be significantly higher in the upper soil layer (0-10 cm) than the lower soil layers. The reports are in consistent with Niirou et al (2015). The variation in SOC in different land uses were in the order: Pine plantation> *Quercus* forest> *Lithocarpus* forest> *Dipterocarpus* forest> Mixed pine forest. The highest value (30.2 Mg/ha) was observed in the topsoil layer (0-10 cm) of pine plantation while the lowest (6.6 Mg/ha) at bottom layer (20-30 cm) of mixed pine forest (Table 3). The high concentration of SOC in the surface soil is due to the higher inputs of litter biomass which accelerates the amount of SOM and SOC in the soil through the process of decomposition (Wapongnungsang et al 2017, Hauchhum and Tripathi 2017, Shah et al 2021).

In the present study, the concentration of available N ranged from 242.5-440.7 µg/g. The highest amount of N was in *Dipterocarpus* forest followed by *Lithocarpus* forest and *Quercus* forest, and the lowest in mixed pine forest. The availability of P in different land uses ranged from 12.2 - 61.8 µg/g. The available P concentration was highest in the pine plantation followed by *Lithocarpus* forest and mixed pine forest, and least in the *Dipterocarpus* forest. Similarly, the highest exchangeable K concentration ranged from 112.3 - 257.3 µg/g) in different land uses. The highest exchangeable K was in *Lithocarpus* forest (180.7 -257.3 µg/g) in different depths. However, the other land uses had comparable exchangeable K concentrations (112.3 - 144.6 µg/g) (Fig. 2). Singh et al (2014) found 184.1 and 157.3 µg/g concentration of available N, 8.5and 7.9 µg/gof available P, 159.9and 136.1 µg/g of exchangeable K in the natural forest land and plantation land which are lower than our reported values in all the land use types except exchangeable K of the plantation which is higher than our values of pine planation. The study on different forests of Kamuan Himalayan had reported available P ranging from 213-267 µg/g in the Banj-oak forest, 93 -167 µg/g in the Chir pine forest and 160 -220 µg/g in the Sal forest which are higher than present study. Similarly, exchangeable K of these study (41 - 54 µg/g in Banj-oak forest; 56 - 62 µg/g chir in pine forest; 25 - 59 µg/g in sal forest) were quite lower than present values.

The N, P and K are the major soil nutrients and their availability in soil play a major role in plant growth and production (Pandey et al 2018). Plants use available form of nitrogen (NO_3-N and NH_4-N) for their growth and development. In the current study, available N varied between different land uses and depth, and ranged from 282.81 to 438.65 kg/ha. The range of available N in the present study is broadly comparable to N availability (219.80

Table 3. Depth wise chemical properties of the soil from various forest types

Soil parameters	Depth (cm)	Land use					LSD (p=0.05)
		MPF	PP	LF	QF	DF	
SOM (Mg/ha)	0-10	29.9^{a}	59.5^{a}	36.5^{a}	38.2^{a}	32.3^{a}	9.27
	10-20	11.8^{b}	42.4^{b}	18.2^{b}	33.8^{ab}	19.3^{b}	5.04
	20-30	11.2^{bc}	27.4^{c}	16.8^{bc}	26.7^{b}	16.7^{bc}	9.52
SOC (Mg/ha)	0-10	16.5^{a}	30.6^{a}	19.3^{a}	19.9^{a}	16.2^{a}	4.98
	10-20	7^{b}	24.3^{b}	10.9^{b}	18.7^{a}	10.6^{ab}	3.25
	20-30	6.6^{bc}	16.4^{c}	10.1^{bc}	16.1^{b}	10.1^{bc}	5.50
N (kg/ha)	0-10	282.8^{a}	343.6^{a}	376.3^{a}	360.5^{a}	376.1^{a}	152
	10-20	345.4^{b}	346.0^{ab}	438.7^{b}	406.4^{b}	344.5^{b}	172
	20-30	250.5^{c}	376.7^{c}	314.1^{c}	407.8^{b}	282.4^{c}	255
P (kg/ha)	0-10	49.2^{a}	54.8^{a}	52.1^{a}	15.1^{a}	26.8^{a}	2.2
	10-20	42.4^{b}	47.6^{b}	49.9^{b}	13.0^{ab}	41.0^{b}	1.8
	20-30	29.1^{c}	37.6^{c}	34.9^{c}	12.7^{ab}	16.3^{b}	1.9
K (kg/ha)	0-10	125.8^{a}	109.9^{a}	236.2^{a}	122.3^{a}	120.6^{a}	1.3
	10-20	116.4^{b}	122.7^{b}	195.4^{b}	109.2^{b}	103.9^{b}	1.3
	20-30	113.0^{c}	109.5^{a}	187.1^{c}	115.0^{c}	102.3^{b}	1.8
pH	0-10	4.4^{a}	4.8^{a}	4.3^{a}	5.1^{a}	4.6^{a}	0.54
	10-20	4.4^{ab}	4.8^{ab}	4.5^{ab}	5.1^{a}	4.8^{b}	0.19
	20-30	4.9^{b}	5.3^{b}	4.8^{b}	5.2^{b}	4.9^{b}	0.13

See Table 2 for details

to 878.1 kg/ha) in different land use types of Manipur (Watham et al 2018, Singh and Athokpham 2018). In contrast to the SOC, the highest soil N content (438.65 kg/ha) was recorded in the middle soil layer (10-20 cm) in *Lithocarpus* forest. This reflects the movement of available N from the upper soil layer to the lower soil layer due to the process of leaching. The low N value (250.45 kg/ha) in mixed pine forest of the bottom layer (20-30 cm) corresponds to the low SOC value in mixed pine forest (Table 3). SOC content has been well correlated with the amount soil available N in different forest soils (Bhuyan and Sharma 2017, Haobijam et al 2020).

Available P helps to promote plant growth through the proliferation of roots and thereby improving the nitrogen fixation process (Watham et al 2018). The available P was the highest in all the soil depths except DF (Table 3). Between sites, high P concentrations were observed at pine plantation (37.64 to 54.75 kg/ha) which may have the effect of litter decay and high concentrations of organic matter. The low P was in *Quercus* forest (12.74 to 15.14 kg/ha), which is in the reported range (11-30 kg/ha) for disturbed mixed oak forest in Manipur (Niirou et al 2015).

Similarly, the exchangeable K is most abundant in the topsoil layer at all sites except Pine plantation which may be due to the leaching effect of the nutrient transfer to the bottom soil layer. The nutrient cycling brings back the nutrient to the surface forest floor through litterfall. Thus, the release of K from the decomposition of organic matter in its high concentration occurred in the topsoil of forest (Kumar et al 1998). In all sites, the highest K values were observed at *Lithocarpus* forest (187.13 to 236.20 kg/ha) which may be due to the input of high amount of leaf litter from very deep trees (Table 3).

The significant interaction was observed at soil depth with pH and BD but negative interactions with MC, SOM, BD, SOC, and P. Soil MC shows a significantly positive interaction with SOM and SOC but a negative interaction with BD. Soil pH had a very good correlation with silt content but significantly negative correlation to P and K. WHC shows positive interaction with sand content but significantly negative interaction with K content, clay and silt. SOM indicates a negative interaction with BD and sand. BD was very well matched with clay and silt. The available P and K also show significantly positive interactions (Table 4).

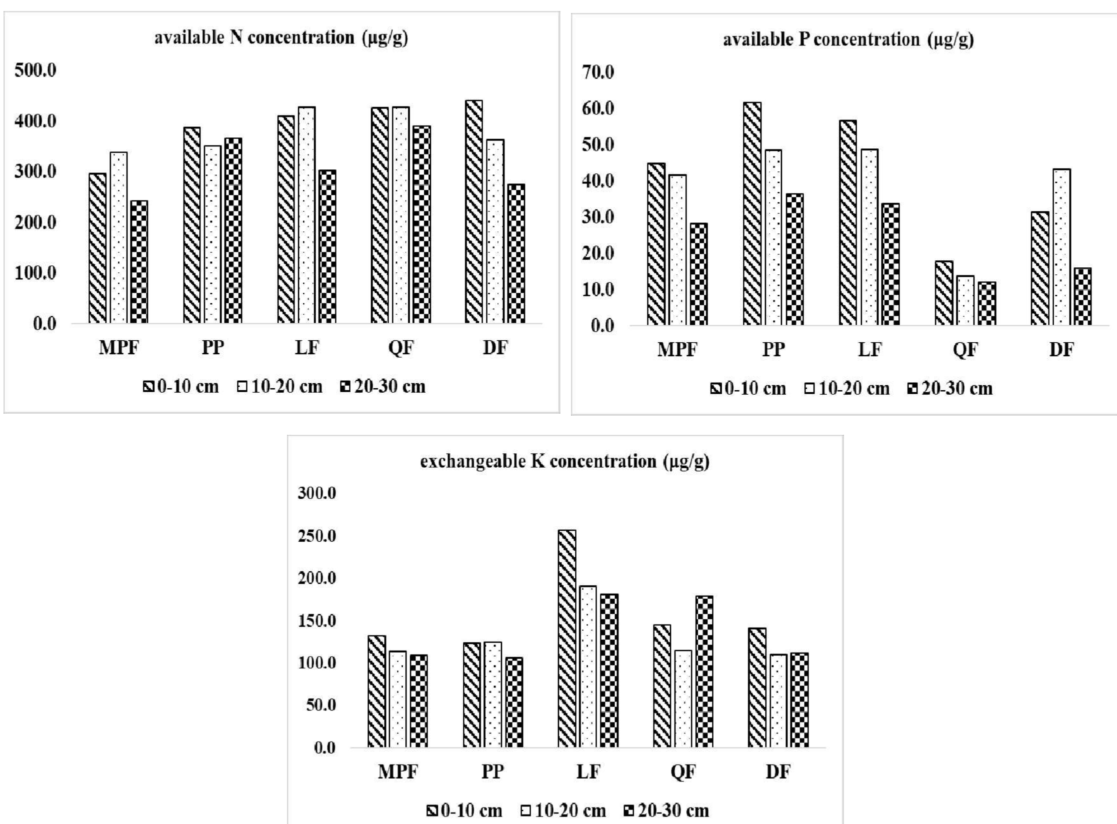

Fig. 2. Spatial changes in the concentration of available NPK (µg/g) in different land-use systems in Manipur. Abbreviation: MPF=Mixed pine forest, PP=Pine plantation, LF=*Lithocarpus* forest, QF=*Quercus* forest, DF=*Dipterocarpus* forest

Table 4. Correlation coefficient (r) of various soil physico-chemical properties in different land use

	Soil depths	MC	pH	WHC	SOM	BD	SOC	N	P	K	Clay	Silt
MC	-.538**											
pH	.489**	-.003										
WHC	.070	.155	-.105									
SOM	-.604**	.562**	.012	-.151								
BD	.730**	-.509**	.199	-.090	-.565**							
SOC	-.527**	.526**	.061	-.181	.988**	-.444**						
N	-.176	.187	.136	-.169	.244	-.126	.262					
P	-.376*	-.199	-.595**	-.144	.217	-.108	.207	-.037				
K	-.186	-.270	-.470**	-.327*	-.039	.012	-.044	.271	.422**			
Clay	.273	-.252	-.100	-.356*	-.190	.298*	-.160	-.130	.189	.442**		
Silt	.224	-.216	.296*	-.588**	.018	.334*	.087	.203	-.179	.053	.296*	
Sand	-.072	.091	-.218	.552**	-.338*	-.179	-.386**	-.139	-.081	-.046	-.348*	-.834**

* Correlation is significant at the 0.05 level (2-tailed); ** Correlation is significant at the 0.01 level (2-tailed)

CONCLUSIONS

The *Lithocarpus* forest and pine plantation having higher soil chemical properties (soil organic carbon, soil organic matter, available NPK) that may be attributed to higher nutrient availability in the region and to the type of the vegetation grown that accumulated higher contain of litter biomass that improve the soil structure. The forests with high soil organic carbon have great potential for mitigating climate change. Therefore, there is a need to promote and conserve such forests.

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Assessment of Leaf Fodder Quality of *Melia dubia* Genetic Resources for Proximate and Mineral Composition

T.M. Akhilraj, K.T. Parthiban[1] and Sneha S. Kambli[2]

Department of Silviculture and Agroforestry, College of Forestry, Sirsi, Uttara Kannada-581 401, India
[1]Department of Agroforestry, Forest College and Research Institute, Mettupalayam-641 301, India
[2]Karnataka Forest Department, Bengaluru-574 221, India
E-mail: akhil.rajtm@gmail.com

Abstract: The present investigation was carried out to evaluate the leaf fodder quality of thirty superior *Melia dubia* genetic resources. *Melia dubia* Cav. is a member of Meliaceae family and it's a multipurpose agroforestry tree used for pulpwood, timber, fuel wood, plywood and afforestation purposes. *Melia dubia* plantation was raised during January, 2010 under National Agriculture Innovation Project at Forest College and Research Institute, Mettupalayam, Tamil Nadu. Selection of superior genetic resources was carried out in the year 2016 by comparison tree method. Further screening has been done and thirty plus trees have been finally screened for leaf fodder quality estimation. In the current study among thirty genetic resources evaluated, the superiority of MTPMD 1 was evident due to higher crude protein (17.30%) coupled with increased dry matter production (37.95%). Among the thirty Melia genetic resources evaluated, high proportion of nutritional constitution viz. (N, P, K, Ca and Mg) was registered by the selection of MTPMD 1. This study is essential for successful breeding program and will help in development of varieties with superior fodder quality. This will help to meet the growing demand of fodder for livestock.

Keywords: *Melia dubia*, Leaf fodder quality, Animal feed, Proximate analysis, Mineral composition

Livestock rearing is one of the major occupations in India that provides dairy products, manure, draught power for agriculture and local transportation. It forms an important source of cash income to millions of households across various parts of the country. Significance of the livestock sector can be appreciated from the fact that it contributes about 8.5- 9% to the country's GDP. With world's largest livestock population, India faces problem in meeting its fodder requirement. Inadequate feed supply to ruminants during the dry season is the basis for poor performance of livestock. Cultivated fodders occupy only 4% of the entire cultivable land in the country. Presently, the country faces a net shortfall of 35.6% green fodder, 10.5% dry crop leftovers, and 44% concentrate feed ingredients (Singh et al. 2022). The option for increasing land area under fodder cultivation is very limited. Hence feed and fodder availability in India is not sufficient to meet the fast-growing ruminant population. There is an immediate need to explore new feed resources which do not compete with human food chain. Tree leaves are an alternative feed source for small ruminants and can help to minimize the wide gap between demand and supply of nutrients (Bakshi and Wadhwa 2007). The interest in search for alternative/additional feed ingredients is of great importance mainly because of the global demand for grains which has exceeded the production and stiff competition between man and the livestock industry for existing food and feed material. There is an urgent need to increase the productivity of cultivated fodder crops on the same piece of land in order to meet the fodder needs of rising number of cattle. In addition to vertical expansion from arable lands, utilization of non-arable land area for pastures is a viable option to balance the demand. There are a number of lesser-known and under-utilized plants that adapted to local, harsh conditions and have tremendous potential as livestock feed (Sukhadiya et al. 2022). Trees have greater adaptability to varied climatic condition and ability to produce biomass throughout the year. As trees require minimal post planting care, the cost of production will be low. In dry season, leaves as fodder is an important source of nutrients to cattle. Fodder trees are highly nutritious, easy to grow, improve soil fertility and are relatively easy to manage. Fodder trees do not compete with food crops, can be intercropped and once matured can be fed to live stock for up to 20 years. Utilization of protein rich fodder trees has been recognized to be one of the most effective means of improving both the supply and the quality of forage in tropical small holder livestock systems, especially during the dry season. In general, fodder tree leaves contain higher crude protein and calcium content compared to grasses and straws and can act as major feeding resources to the animals (Manoj et al 2017).

Melia dubia is one of the best trees that provides fodder to cattle during the off seasons. With its superior characteristics like fast growing nature, easy coppicing and high leaf biomass, it's a best alternative fodder resource. The processing and conservation of fodder trees is increasingly being recognized and explored, while the cultivation is minimal and insignificant. The role and importance of fodder trees for livestock production, their nutritional quality and factors that limit their use in livestock production are reviewed. The need for increased cultivation and integration of fodder trees into local farming systems through agroforestry is imperative in order to promote livestock production and also to support rural livelihoods. However, fodder quality studies in *Melia dubia* have not been evaluated. Hence the current investigation was to determine the fodder quality of *Melia dubia* genetic resources.

MATERIAL AND METHODS

Leaves were obtained from the felled thirty genetic resource of *Melia dubia* from the 6-year-old existing plantation located at Forest College and Research Institute, Mettupalayam. These leaves were dried and powdered by using Willey mill passed through 60 mesh and analysis have been carried out. The leaves samples were subjected to analysis for proximate and mineral composition. All the analysis were carried out in the research laboratory present in the Agroforestry department, Forest College and Research Institute, Mettupalayam.

Proximate analysis: Proximate analysis includes dry matter, ash, crude fat, crude protein, which are essential in determining the quality of the fodder. This analysis was carried out as per the guidelines of AOAC (1990).

Moisture content: Determination of moisture was done by; one gram of each sample was taken in a petri-dish and placed in an oven at 100 °C for four hours. It was then cooled in desiccators and weighed. The samples were heated again in the oven for another two hours and the process was repeated, till a constant weight obtained. The moisture content was calculated by using the following formula

$$\text{Moisture (\%)} = \frac{\text{Weight of fresh sample} - \text{Weight of dried sample}}{\text{Weight of the sample}} \times 100$$

.

Ash content: Determination of ash was done by one-gram dried sample was taken in a crucible and charred over a low flame and kept in a muffle furnace set at 550 °C until white ash was obtained. The ash was moistened with water, dried on steam and then on hot plate. The crucible was again placed in the muffle furnace at 550°C, till a constant weight was obtained. The per cent ash was calculated as:

$$\text{Ash (\%)} = \frac{\text{Weight of sample of ter ash}}{\text{Weight of the sample}} \times 100$$

Crude fat: Determination of crude fat done by the dried sample was taken and crushed. Two gram of the sample was taken in a paper thimble and connected to a soxhlet extractor. Then 300 ml of petroleum ether was poured on the flask and refluxed for 12 hours with a heating mantle. Crude fat was extracted in a flask. The flask was cooled in a desiccator and the weight was taken. Crude fat was determined by using the formula:

$$\text{Crude fat (\%)} = \frac{\text{Weight of flash with fat} - \text{Weight of empty flask}}{\text{Weight of original sample}} \times 100$$

Crude fiber: Determination of crude fiber was done by one gram of the defatted plant material was taken in beakers and boiled in 200 ml of 1.25% sulphuric acid for 30 minutes. The content was then filtered and washed with distilled water to neutralize the content. The content was transferred again to the beaker and boiled in 200 ml of 1.25% sodium hydroxide for 30 minutes. They were again filtered and washed with distilled water for neutralization. A Gooch crucible was prepared with an asbestos mat and the contents of the beakers were placed on the mat and washed with 15 ml of ethyl alcohol. The crucible was dried in an oven at 110 °C to a constant weight. The crucible having crude fiber was cooled and weighed (W1). The content of the crucibles were ignited over a low flame until charred and then kept in a muffle furnace at 550 °C and weighed (W2). The Percentage fiber was determined by the following formula:

$$\text{Crude fibre (\%)} = \frac{W1 - W2}{\text{Weight of the sample}} \times 100$$

Crude protein: Determination of crude protein is done by micro Kjeldahl's method. It involves digestion, distillation and titration of the samples. Digestion is done by one gram of dried plant material of each species was taken in the digestion flask. To this, 10ml diacid was added. The solution was heated until it became clear and frothing ceased. It was then boiled gently for another 2 hours and then it was cooled down. This digested material was mixed with 30 ml of water with constant mixing. The digest was transferred to 100 ml volumetric flask and necessary amount of water was added up to the mark of the flask. Distillation is carried out in the Kjeloplus distillation unit. 25 ml of 2% boric acid was taken and double indicator was added, by which wine red colour could be observed. Then 10 ml of the digest was transferred to the distillation assembly and 10 ml of 40% sodium hydroxide solution was added on it. The distillation was completed in 10 minutes indicating the change of color of boric acid to blue due to the formation of ammonium borate. Titration is carried out by The boric acid having trapped ammonia was titrated with 0.02N sulphuric acid, the colour of boric acid having ammonia changed again to pink. The percent nitrogen was calculated by the formula

$$N\ (\%) = \frac{0.00028 \times X \times 100 \times 100 \times 100}{10 \times 1 \times 100 - x}$$

x – moisture content of the leaf,

From this crude protein was found by using the formula

CP (%) = 6.25 x nitrogen

Mineral composition: Mineral composition includes Nitrogen, Phosphorous, Potassium, Calcium, Magnesium and Carbohydrate content present in the sample, which is important constituent of nutritional feed. All this analysis were carried out as per the guidelines of AOAC (1990).

Nitrogen: Total nitrogen is estimated by micro Kjeldahl's method.

Phosphorous: Total phosphorous estimated by Vanadomolybdate yellow colour method. Pipette out 5ml of triacid extract into 25 ml volumetric flask, add 5ml Barton's reagent and make up volume. Allow 30 min. to develop yellow colour. Measured the intensity of colour in UV spectrophotometer (470nm).

Potassium: Potassium is estimated by pipette out 5ml of triacid extract into 25 ml volumetric flask, neutralize the acid with ammonium hydroxide, make up volume. Measured the concentration of K in the solution by using Flame photometer.

Calcium and Magnesium: Calcium and Magnesium get complexed by EDTA in order of Ca first followed by Mg. Ca is estimated first by using murexide indicator in the presence of sodium hydroxide at pH 12. The Ca + Mg is estimated using Erichrome block-T in the presence of ammonium chloride and ammonium hydroxide buffer solution at pH 10.

Carbohydrate: Carbohydrate is measured by weighed 0.1g sample in to boiling tube. Hydrolysed it with 5ml of 2.5 N HCl. Neutralize it with sodium carbonate. Made up the volume 100ml and centrifuge. Collect supernatant, prepare standards. Add 4ml anthrone to the samples, heat for eight min. and take reading in spectrophotometer (630nm).

RESULTS AND DISCUSSION

Nutritive quality of leaves are very important for the fodder crops. Thirty *Melia dubia* genetic resources were subjected to the analysis for fodder quality. Both proximate as well as mineral content of the leaves were determined (Table 1 & 2). Dry matter is the actual amount of feed material leaving water and volatile acids and bases. Among the genetic resources the highest leaf moisture content was registered by MTPMD 6 (66.11%) and the lowest moisture content was registered by MTPMD 1 (62.01%). The highest dry matter was found in MTPMD 1 (37.95%) whereas the lowest was in MTPMD 6 (33.85%), highest ash content was registered by MTPMD 12, MTPMD 25 and MTPMD 42 (7.5%) and lowest was registered by MTPMD 6 (7.20 %). The results indicated that higher moisture content leads to lower dry matter yield and vice versa. Species containing more than 30% dry matter, 50% organic matter digestibility and less than 10% total ash in the dry matter generally considered as good fodder (Mandal and Gautam 2012). All the Melia genetic resources registered higher dry matter as well as lower ash content, which implies their suitability as a good fodder, among 30 genetic resources, the superiority of MTPMD 1 was very well witnessed. Similar work has been carried out Azim et al. (2011), Berhe and Tanga (2013), Haftay and Kebede (2014) and Awotoye et al (2016) in several tree species and found superiority of few species, which extent support to the current situation.

Crude protein (CP) content is the most important criterion for judging feed and fodder. Among the genetic resources, highest CP was registered by MTPMD 1 (17.30 %) and the lowest CP was registered by MTPMD 2 (16.80 %) which indicated that MTPMD 1 leaves as a good protein supplementn. This supports the findings of Onwuka (1980) who reported that main features of fodder are high crude protein and mineral content, and added that average trees and shrubs are richer in crude protein and lower ash than tropical grasses. The findings of this study were in line with those of Bakshi and Wadhwa (2004). They also reported high CP in the *Melia azedarach* and *Morus alba*. Srivastava et al. (2006) reported high CP contents of *Morus alba* (15.31-30.91%) on dry matter basis. Ayodele et al. (2014) and Amanulla et al. (2006) reported *Albizzia lebbeck* is very rich in crude protein with mean value of 20%.

Ether extract or fat is also a measure of energy levels of feedstuffs. Crude fat content was highest in MTPMD 42 (3.61 %) and lowest was in MTPMD 25 (2.81 %), carbohydrate content was maximum in MTPMD 30 (17.50 %) and minimum in MTPMD 14 and MTPMD 19 (15.00 %). This indicated that MTPMD 42 leaves could be a good source of energy that can be utilized by ruminants for body maintenance and production. Similar variations are also reported by other research group (Brandis et al. 1978, Malla 2004, Kayastha et al. 1998, Panday and Tiwari 2003).

Nitrogen is very essential for plant growth and serves as a source of supplementary energy for animal feeding, MTPMD 1 registered highest N content (2.77 %) and MTPMD 2 and MTPMD 11 registered lowest N content (2.69 %). Carew (1982) from his findings reported that *Gliricidia sepium* had a higher Nitrogen content with values ranging between 3.2% and 4.21%. Similar work was carried out in *Quercus serrata* (Migita et al 2007). Maximum phosphorus content was found in MTPMD 1 and MTPMD 30 (0.34 %), minimum was in

MTPMD 11 (0.28 %). Highest potassium content was registered by MTPMD 1 and MTPMD 30 (0.09 %) lowest was in MTPMD 11 and MTPMD 25 (0.06 %).

Highest calcium content was found in MTPMD 30 (2.30 %) and lowest was in MTPMD 11 (2.16 %); maximum magnesium content was found in MTPMD 1, MTPMD 13, MTPMD 22, MTPMD 30, MTPMD 42 (0.33 %) and minimum was in MTPMD 12 & MTPMD 26 (0.29 %). The Melia genetic resource in the current study registered relatively higher calcium level, which appreceable to the results of Abdulrazak et al (2000) and Aganga et al. (2000). The leaves of *Melia* examined in the present study exceeded the recommended level of calcium for lactating ewes (11g/Kg DM) suggested by NRC (1985). However concentration of phosphorous was extremely low in the leaves of genetic resources studied. Potassium is also an essential element for growth and development of animals. Deficiencies in K in cattle can result in reduced intake, weight lose and stiff joints. Cattle stressed owing to long transport

Table 1. Proximate analysis of *Melia dubia* genetic resources for leaf fodder

Genetic resource no.	Moisture content %)	Dry matter (%)	Ash (%)	Crude fat (%)	Crude protein (%)	Carbohydrate (%)
MTPMD 1	62.01	37.95*	7.35	3.21*	17.30*	17.10*
MTPMD 2	65.31*	34.65	7.25	2.96	16.80	16.30
MTPMD 3	62.65	37.31	7.40	3.01	16.95	16.00
MTPMD 4	62.41	37.55*	7.45*	2.76	17.10	16.50*
MTPMD 5	63.11	36.85*	7.21	3.11	17.00	15.50
MTPMD 6	66.11*	33.85	7.20	2.81	17.20	16.00
MTPMD 7	63.21	36.75*	7.26	3.01	16.85	16.8*
MTPMD 11	62.91	37.05	7.25	2.86	16.82	16.85*
MTPMD 12	64.26*	35.70	7.50*	2.96	17.10	16.90*
MTPMD 13	63.71	36.25	7.25	2.91	17.20	16.00
MTPMD 14	62.56	37.40*	7.46*	3.01	17.15	15.00
MTPMD 15	62.91	37.05*	7.35	3.06	17.15	16.50*
MTPMD 17	63.63	36.33*	7.26	3.16	17.20	17.30*
MTPMD 19	64.36*	35.60	7.30	2.96	17.02	15.00
MTPMD 21	64.26*	35.70	7.35	2.91	17.18	16.00
MTPMD 22	64.71*	35.25	7.39	3.01	17.05	17.00*
MTPMD 23	65.41*	34.55	7.30	3.26*	16.97	17.30*
MTPMD 24	65.56*	34.40	7.25	2.86	16.98	16.90*
MTPMD 25	65.91*	34.05	7.50*	2.81	17.05	15.90
MTPMD 26	62.41	37.55*	7.40	2.96	16.95	16.00
MTPMD 29	64.85*	35.11	7.38	3.01	17.12	16.00
MTPMD 30	65.11*	34.85	7.26	2.91	17.25	17.50*
MTPMD 32	63.31	36.65*	7.40	3.11	16.85	16.00
MTPMD 34	62.91	37.05*	7.37	3.06	16.89	15.40
MTPMD 39	63.45	36.51*	7.25	3.01	17.20	15.20
MTPMD 40	64.36*	35.60	7.35	2.96	16.99	15.80
MTPMD 42	64.40*	35.56	7.50*	3.61*	17.05	17.00*
MTPMD 43	65.11*	34.85	7.28	2.86	17.10	15.40
MTPMD 44	64.59*	35.37	7.39	2.91	17.11	16.10
MTPMD 46	63.21	36.75*	7.36	3.21*	17.20	16.20
Mean	63.95	36.00	7.34	3.01	17.06	16.25
Sed	0.08	0.11	0.04	0.09	0.11	0.12
CD (p=0.05)	0.15	0.22	0.08	0.18	0.21	0.25

*Significant at 5% level

Table 2. Leaf mineral composition of *Melia dubia* genetic resources

Genetic resource No.	Nitrogen %	Phosphorus %	Potassium %	Calcium %	Magnesium %
MTPMD 1	2.77*	0.34*	0.09*	2.29*	0.33
MTPMD 2	2.69	0.29	0.07	2.21	0.31
MTPMD 3	2.71	0.32	0.08	2.20	0.32
MTPMD 4	2.74	0.30	0.08	2.24	0.30
MTPMD 5	2.72	0.31	0.07	2.23	0.30
MTPMD 6	2.75	0.30	0.08	2.20	0.32
MTPMD 7	2.70	0.30	0.07	2.20	0.30
MTPMD 11	2.69	0.28	0.06	2.16	0.28
MTPMD 12	2.74	0.30	0.08	2.24	0.29
MTPMD 13	2.75	0.31	0.08	2.23	0.33
MTPMD 14	2.74	0.31	0.08	2.20	0.32
MTPMD 15	2.74	0.29	0.08	2.20	0.30
MTPMD 17	2.75	0.33*	0.08	2.29*	0.31
MTPMD 19	2.72	0.31	0.07	2.20	0.30
MTPMD 21	2.75	0.30	0.08	2.23	0.32
MTPMD 22	2.73	0.30	0.07	2.24	0.33
MTPMD 23	2.71	0.30	0.07	2.20	0.31
MTPMD 24	2.72	0.32	0.08	2.26	0.31
MTPMD 25	2.73	0.31	0.06	2.24	0.30
MTPMD 26	2.71	0.33*	0.08	2.19	0.29
MTPMD 29	2.74	0.31	0.07	2.22	0.30
MTPMD 30	2.76*	0.34*	0.09*	2.30*	0.33
MTPMD 32	2.70	0.31	0.08	2.21	0.32
MTPMD 34	2.70	0.31	0.08	2.19	0.30
MTPMD 39	2.75	0.32	0.08	2.20	0.30
MTPMD 40	2.72	0.30	0.07	2.27*	0.31
MTPMD 42	2.73	0.33*	0.08	2.26	0.33
MTPMD 43	2.74	0.30	0.08	2.25	0.32
MTPMD 44	2.74	0.31	0.08	2.24	0.30
MTPMD 46	2.75	0.29	0.07	2.20	0.30
Mean	2.73	0.31	0.08	2.23	0.31
Sed	0.02	0.01	0.01	0.02	0.01
CD (p=0.05)	0.03	0.02	0.01	0.04	0.03

*Significant at 5% level

distance may require increased levels of K to replenish lost body reserves (NRC 1980). In the present study, potassium level was comaparatively low, which may be due to the age factors. Young tissue posses higher concentrations of N, P, K and Mg and lower calcium concentrations on dry matter basis.

CONCLUSION

Among thirty *Melia dubia* genetic resource MTPMD 1 exhibited the better values for proximate analysis as well as mineral composition thus it proved to be superior genetic material with improved leaf fodder quality and extended scope for further breeding programme and immediate deployment in industrial agroforestry plantations.

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Evaluation of Half-sib Progeny of *Grewia optiva* Drummond under Nursery Conditions

H.P. Sankhyan, Jyoti Dhiman and Krishan Chand

College of Forestry, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan-173 230, India
E-mail: sankhyanhp@gmail.com

***Abstract*:** Significant variability was observed for different years and between selected populations for all seedling growth characteristics in two-year-old half-sib progenies of *Grewia optiva*. Phenotypic coefficient of variation (PCV) found higher than the genotypic coefficient of variation (GCV) for all the characters. GCV and PCV values were observed at their maximum for the number of branches (GCV: 20.97% and PCV: 24.06%) and branch angle (GCV: 22.30% and 24.92%). The highest heritability was recorded for leaf area, i.e., 91.22, and highest genetic advance (64.20) for seedling height and highest genetic advance as a percent of mean (41.12%) were observed for branch angle. The root/shoot ration had the lowest values of heritability, genetic advance, and genetic advance as a percentage of mean. The principal components (PCI-PCIII) cumulatively accounted for 71.07 per cent of the total variation. PCA I explained 44.43% of the total variance. The genetic divergence divided thirty-five populations into two major clusters. The maximum inter-cluster distance was observed between population Dharkyari (SI-5) and population Gangal (MA-2).

Keywords: *Grewia optiva,* Genetic gain, Genetic divergence, Variability, Heritability

Grewia optiva is locally called Bhimal, Beul, or Dhaman belongs to the Tiliaceae family and is native to India. It is a unique medicinal tree of the sub-Himalayan terrain, often used for fibre and fodder by local farmers. The genus *Grewia* was named after Nehemiah Grew (1964-1712), the founder of plant physiology. Bhimal (*Grewia optiva*) is a medium-sized multipurpose nutritious fodder tree species growing in sub-tropical climates of the north-western Himalayas, generally raised on terrace risers and fairly well distributed up to middle elevations (500-2500 masl) in India, Pakistan, and Nepal (Semwal et al 2002). It is preferentially grown for fodder in the hills of Uttarakhand, Himachal Pradesh, and Nepal, etc., due to its high palatability, faster growth, easy propagation and high forage yield over other tree species (Mukherjee et al 2018) and its ability to retain an appreciable amount of nutrients in its leaves (Katoch et al 2017). Livestock rearing combined with agriculture is a common practise in the hilly areas of the country and it plays an important role in the economy of the country. The availability of nutritious fodder is primarily dependent on the availability of tree fodder (Roder 1992), particularly during the winter months when the availability of quantity (Khanal and Subba 2001) and quality (Vishvakarma et al 1998, Roder et al 2003) of green fodder is limited. *Grewia optiva* leaf fodder is almost as nutritious as that of leguminous crops, containing high digestibility, good vitamins and minerals, and it also improves the microbial growth and digestion of cellulosic biomass in the rumen of livestock (Singh 1982). Its leaves are fairly rich with 17.4-21.0% crude protein, 17-21.5% crude fiber, 10.4-21.5% total ash, 4.2-6.0% ether extract, and 40.4-50.2% nitrogen free extract (Sankhyan and Bhagta 2016) and do not contain tannins (Orwa et al 2009). Crude protein is highest in young leaves and in winter leaves but decreases during the rainy season. Verma et al. 2014, Orwa et al 2009).The high calorific value (4920 kcal kg^{-1}) of the tree wood makes it an excellent fuel wood and alternative energy source. It provides fibre, edible fruits and is also used as a traditional medicinal tree for treating various diseases like cough, dysentery, diarrhoea, small pox, malaria, typhoid, intestine and bladder with irritable conditions, rheumatism and eczema (Chopra et al 1956). Besides producing valuable products, the tree also provides a variety of ecological functions and associated services (Verma et al 2014). A considerable amount of organic matter is also added to the soil through the litter fall of Bhimal leaves (Kar *et al* 2019).

Progeny testing is a prerequisite to estimating the genetic worth of parents while screening the naturally available genetic variations so as to isolate good genotypes rather than merely selecting good phenotypes and to achieve maximum gain per unit area. The aim of the present study was to delineate genetically divergent best nutritive strains of *G. optiva* Drummond in different geographical regions of Himachal Pradesh and generate promising breeding material or heterotic vigour through hybridization between genetically distant families of *G. optiva.* Hence, keeping in view the above aim, the present investigation was executed.

MATERIAL AND METHODS

The present investigation on the evaluation of half-sib progeny of *G. optiva* Drummond was carried out for two years, *i.e.*, 2019-20 and 2020-21 at College of Forestry, Dr. Y. S. Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh. Taking into account the rich genetic diversity and phenotypically superior plant populations of *G. optiva,* thirty-five populations of the 20 cm-30 cm diameter class (five populations in each district) (Table 1) were selected from seven districts of Himachal Pradesh, namely Kangra, Mandi, Bilaspur, Solan, Sirmaur, Una, and Hamirpur. To study the growth performance of the progeny under nursery conditions, seeds were collected, processed, and maintained separately on an individual tree basis. Seeds obtained from different selected trees were depulped by soaking in lukewarm water, dried for 3-4 days and sown during April–May 2019 in poly bags (in three replications) under Shilli nursery conditions. Shilli nursery is situated at 30.904486° N, 77.096733° E at an altitude of 1300 m above mean sea level, with an average annual rainfall of 1262 mm. The germination of seeds takes place in 10-15 days of sowing.

Different growth characteristics of progenies, *viz.,* seedling height, root/shoot ratio, internodal length, leaf length, and leaf breadth, were measured with the help of a scale; basal diameter with a vernier calliper, branch angle with a protractor; number of branches, number of leaves visually counted and leaf area measured with the help of a leaf area meter. The data on seedling growth characteristics was recorded for two years, and pooled data for two years is presented under the present investigation. The genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) analysis (Allard 1960) and genetic advancement and heritability as per Burton (1952). Genotypic correlation coefficients and principal component analysis were calculated using OP-STAT (Sheoran *et al* 1998) and PAST (Hammer *et al* 2001). A cluster analysis was performed on PAST.

RESULTS AND DISCUSSION

The analysis of variance (Table 2) was conducted for all the progeny characteristics of thirty-five selected populations of *Grewia optiva*. Significant differences were observed for all seedling characteristics between different years and for different populations. A significant interaction was also observed between population/treatments and years.

Genetic variability parameters revealed that the phenotypic coefficient of variation (PCV) was higher than the genotypic coefficient of variation (GCV) for all the characters under observation (Table 3). A narrow difference existed between PCV and GCV in most characters, showing that they were comparatively stable to environmental pressure, which means environmental influences were very low and hence the phenotypic performance of traits can be used as a

Table 1. Details of thirty-five populations selected from seven districts of Himachal Pradesh

Population No.	Population name	Population code	District
1	Kothi kanwal	SO-1	Solan
2	Unchagaon	SO-2	Solan
3	Nerikalan	SO-3	Solan
4	Gaddo	S0-4	Solan
5	Devera	SO-5	Solan
6	Machair	SI-1	Sirmaur
7	Jajjar	SI-2	Sirmaur
8	Neharbag	SI-3	Sirmaur
9	Badon	SI-4	Sirmaur
10	Dharkyari	SI-5	Sirmaur
11	Kant	UN-1	Una
12	Navami	UN-2	Una
13	Kharunibangana	UN-3	Una
14	Thanakalan	UN-4	Una
15	Lamlehri	UN-5	Una
16	Katoi	KA-1	Kangra
17	Balugloa	KA-2	Kangra
18	Purana kangra	KA-3	Kangra
19	Dohan	KA-4	Kangra
20	Balla	KA-5	Kangra
21	Janhen	HA-1	Hamirpur
22	Jhinkari	HA-2	Hamirpur
23	Harbalneri	HA-3	Hamirpur
24	Anu khurd	HA-4	Hamirpur
25	Bhaleth	HA-5	Hamirpur
26	Patta	MA-1	Mandi
27	Gangal	MA-2	Mandi
28	Bagla	MA-3	Mandi
29	Balt	MA-4	Mandi
30	Bharnoi	MA-5	Mandi
31	Ghumarwin	BI-1	Bilaspur
32	Barthi	BI-2	Bilaspur
33	Kuthera	BI-3	Bilaspur
34	Jukhala	BI-4	Bilaspur
35	Nehari	BI-5	Bilaspur

criterion for selection. GCV and PCV were observed at their maximum for the number of branches (GCV: 20.98% and PCV: 24.07%) and branch angle (GCV: 22.31 and 24.93%). Genetic parameters permit identifying the action nature of involved genes as well as evaluating the efficiency of different selection methods and strategies (Cruz et al 2014). The highest heritability was recorded for leaf area, i.e., 91.22; the highest genetic advance (64.204) for seedling height and genetic advance as per cent of mean (41.127%) was observed for branch angle (Table 3). The root/shoot ration had the lowest values of heritability, genetic advance, and genetic advance as a percentage of mean. The relationship between heritability and genetic advance of a trait aids breeders in predicting the performance of those traits in future generations as well as their response to selection. Similar findings were reported by Kundal et al (2020) in their study on half-sib progeny evaluation of *Toona ciliata.*

The Pearson genotypic and phenotypic correlation was also observed for all the characters under study. The seedling height showed a highly significant and positive genotypic and phenotypic correlation (Table 4, Fig. 1) with basal diameter (rg: 0.702, rp: 0.540), number of branches (rg: 0.564, rp: 0.404), leaf area (rg : 0.708, rp: 0.647), leaf length (rg: 0.554, rp: 0.437), leaf breadth (rg: 0.422, rp: 0.328) and with the number of leaves (rg: 0.865, rp: 736). Root: shoot showed a strong genotypic correlation with branch angle (rg : 0.366). There was a strong genotypic correlation observed between basal diameter and the number of branches and leaf length. Basal diameter also showed a strong genotypic and phenotypic correlation with leaf area (rg: 0.569, rp: 0.467), and with the number of leaves (rg: 0.706, rp: 0.454). Strong and positive genotypic and phenotypic correlations were

Table 2. Anova for different seedling/half sib progenies growth characteristics of *Grewia optiva*

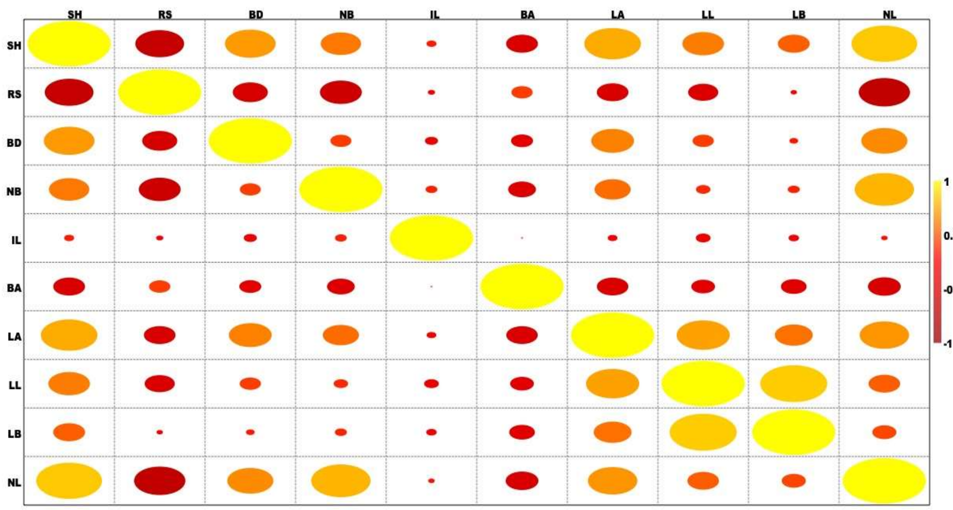
Source	DF	SH	RS	BD	NB	IL	BA	LA	LL	LB	NL
		MS	MS	MS	MS	MS	MS	MS	MS	MS	MS
Rep/year	4	0.898	0.000	0.071	0.087	0.013	0.655	1.593	0.045	0.007	8.277
Treatments	34	786.27**	0.015**	11.80**	28.78**	2.364**	540.010**	181.92**	5.707**	3.964**	859.56**
Year	1	17407.78**	0.0005	2531.0**	6.97**	44.96**	17431.07**	7324.26**	42.33**	25.36**	158148.24**
Treat* year	34	50.25**	0.004**	3.36**	3.92**	0.340**	59.68**	8.35**	0.38**	0.3203**	206.04**
Pooled error	136	2.346	0.00	0.058	0.064	0.007	1.109	1.721	0.036	0.014	6.732
Total	209										
CV		2.583	2.742	2.757	2.603	2.521	2.625	2.686	2.595	2.713	3.033
CD 5% Y		0.418	NS	0.066	0.069	0.023	0.287	0.358	0.052	0.032	0.708
CD 5% T		8.318	0.080	2.152	2.326	0.684	9.065	3.391	0.729	0.664	16.842
CD 5% Y*T		2.473	0.021	0.389	0.408	0.138	1.700	2.118	0.305	0.192	4.189

Significant at 5 % level, Where; SH-seedling height, R/S-root/shoot ratio, BD-basal diameter, NB- number of branches, IL-internodal length, BA- branch angle, LA-leaf area, LL- leaf length, LB- leaf breadth, NL- number of leaves, CV-coefficient of variation, CD-critical difference, Y-year, T-Treatment

Table 3. Genetic estimates for growth characteristics of *Grewia optiva* progenies

Parameters	Range	Coefficient of variability		H^2 (Heritability)	GA (Genetic advance)	GAM (Genetic advance as percent of mean)
		GCV	PCV			
SH (cm)	30.99-87.44	18.674	19.908	87.985	64.204	36.084
R/S	0.4-0.61	8.907	12.185	53.426	0.191	13.411
BD (mm)	3.78-13.99	13.578	18.201	55.655	5.468	20.867
NB	5.61-13.67	20.978	24.066	75.983	10.965	37.669
IL (cm)	1.68-5.14	17.157	19.831	74.846	3.105	30.577
BA (˚)	15.54-66.66	22.308	24.927	80.094	49.486	41.127
LA (cm^2)	31.45-62.45	11.014	11.532	91.22	31.746	21.671
LL (cm)	5.3-8.99	12.925	13.832	87.319	5.438	24.88
LB (cm)	3.02-5.87	17.79	19.291	85.047	4.441	33.797
NL	45.13-137.21	12.202	15.581	61.328	50.509	19.684

See Table 2 for details. GCV-genotypic coefficient of variability, PCV-phenotypic coefficient of variability recorded between the number of branches and leaf area, i.e., rg: 0.459, rp: 0.396, and between the number of branches and number of leaves per seedling. Non-significant and negative correlations were observed between internodal length and all other observed characteristics. There was a negative and significant genotypic and phenotypic correlation observed between branch angle and all other characteristics. Leaf area showed a highly significant correlation with leaf length (rg: 0.688, rp: 0.585), leaf breadth (rg: 0.496, rp: 0.399) and with the number of leaves per seedling (rg: 0.663, rp: 0.536). Strong phenotypic and genotypic correlations were observed between leaf length



See Table 2 for details

Fig. 1. Presentation of phenotypic and genotypic correlation between different seedling growth characteristics

Table 4. Genotypic and phenotypic correlation among growth characters of *Grewia optiva* progenies

		SH	RS	BD	NB	IL	BA	LA	LL	LB	NL
SH	rg	1.000									
	rp	1.000									
RS	rg	-0.669^{**}	1.000								
	rp	-0.533^{**}	1.000								
BD	rg	0.702^{**}	-0.545^{**}	1.000							
	rp	0.540^{**}	-0.344^{**}	1.000							
NB	rg	0.564^{**}	-0.603^{**}	0.317^{**}	1.000						
	rp	0.404^{**}	-0.424^{**}	0.188^{NS}	1.000						
IL	rg	0.133^{NS}	-0.057^{NS}	-0.147^{NS}	0.135^{NS}	1.000					
	rp	0.083^{NS}	-0.073^{NS}	-0.139^{NS}	0.119^{NS}	1.000					
BA	rg	-0.415^{**}	0.366^{**}	-0.277^{*}	-0.369^{**}	-0.049^{NS}	1.000				
	rp	-0.335^{**}	0.176^{NS}	-0.235^{*}	-0.284^{*}	0.050^{NS}	1.000				
LA	rg	0.708^{**}	-0.457^{**}	0.569^{**}	0.459^{**}	-0.098^{NS}	-0.373^{**}	1.000			
	rp	0.647^{**}	-0.319^{**}	0.467^{**}	0.396^{**}	-0.100^{NS}	-0.363^{**}	1.000			
LL	rg	0.554^{**}	-0.498^{**}	0.335^{**}	0.154^{NS}	-0.170^{NS}	-0.312^{**}	0.688^{**}	1.000		
	rp	0.437^{**}	-0.255^{*}	0.178^{NS}	0.162^{NS}	-0.153^{NS}	-0.239^{*}	0.585^{**}	1.000		
LB	rg	0.422^{**}	-0.083^{NS}	0.145^{NS}	0.131^{NS}	-0.121^{NS}	-0.339^{**}	0.496^{**}	0.833^{**}	1.000	
	rp	0.328^{**}	-0.042^{NS}	0.047^{NS}	0.126^{NS}	-0.101^{NS}	-0.262^{*}	0.399^{**}	0.775^{**}	1.000	
NL	rg	0.865^{**}	-0.689^{**}	0.706^{**}	0.951^{**}	0.148^{NS}	-0.418^{**}	0.663^{**}	0.491^{**}	0.356^{**}	1.000
	rp	0.736^{**}	-0.576^{**}	0.454^{**}	0.545^{**}	0.003^{NS}	-0.360^{**}	0.536^{**}	0.280^{*}	0.221^{NS}	1.000

See Table 2 for details

and leaf breadth (rg: 0.833, rp: 0.775) and the number of leaves (rg: 0.491, rp: 0.280). These results find support with the findings of Deepanjli (2018) in different seed sources of *Toona ciliata,* Thakur and Thakur (2015) in *Melia azedarach*, Singh et al (2015) in *Populus deltoides.*

PCA (principal component analysis) results showed that the principal components I (PC I), principal component II (PC II) and principal component (PC III) gave eigenvalues >1.0. PC I accounted for 44.43% of the total variation (Table 5). PC II accounted for 16.088 % of total variation and PC III accounted for 10.553 % of total variation. PC I was positively associated with the characteristics viz., seedling height, basal diameter, number of branches, leaf area, leaf length, leaf breadth, and number of leaves. The PC II was strongly associated with the characteristics, *viz.,* number of branches and internodal length. PC III is associated positively with internodal length, leaf length, and leaf breadth. Thus, the use of these characteristics will help in saving a considerable amount of time for the identification and selection of the best genotypes of *Grewia optiva*. The principal component analysis (PCA) is one of the powerful statistical methods widely applied to classify phenotypic traits in tree germplasm into groups based on similarities. PCA guides the choice of parents for genetic improvement (Afuape et al 2011, Beheshtizadeh et al 2013). PCA reduce the original variables into a new set of uncorrelated variables known as principal components (PCs). These PCs clarify the connections between traits and divide the total variance of original traits into a small number of uncorrelated new variables (Wiley and Lieberman 2011).

A biplot (Fig. 2) was also drawn using the values of PCA I and PCA II. The PCA allows visual differentiation among entries and identify possible associations by providing a two-dimensional scatter plot consisting of individual entries. The geometric distance among individuals in this plot reveals the genetic distance among them. Amalgamation of individuals in a similar quadrant of plot may indicate a group of genetically related individuals. (Warburton et al 2002). The higher the coefficients of particular characters, more it is related to the respective principal component axis. Four grouping of seedling growth characteristic was observed in Biplot and some overlapping occurred within groups demonstrating the relatedness of the seedling growth characteristic of different seed sources.

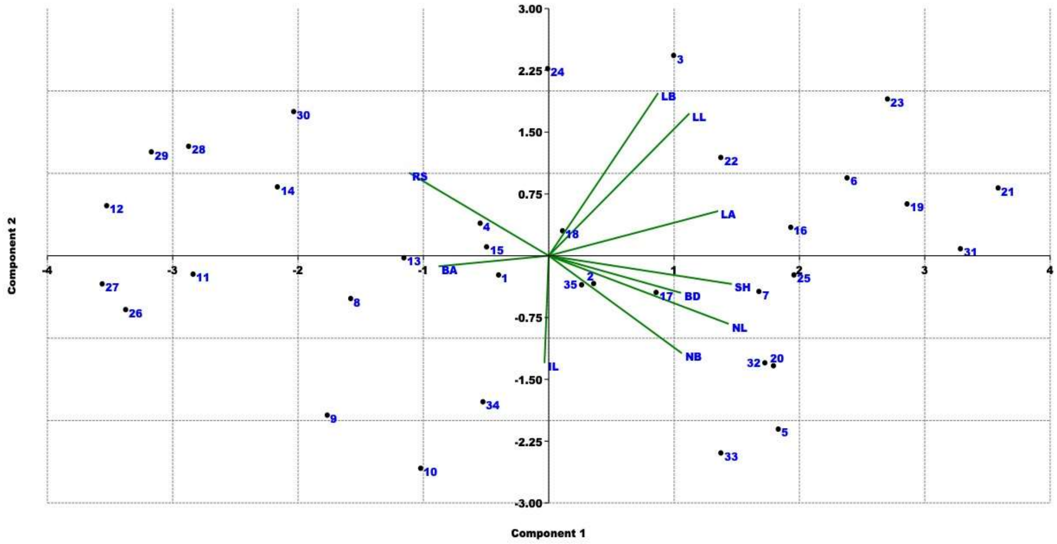
In the biplot graph of PCA, the quadrant I (+, +) consisting of 9 different populations formed the cluster 1, which was highly influenced by three growth characteristics, viz., leaf breadth, leaf length, and leaf area. The cluster II, corresponding to the quadrant II (-, +), contained 9 different populations and was highly influenced by seedling growth characteristics, viz., seedling height, basal diameter, number of leaves, and number of branches. Similarly, the cluster III corresponding to quadrant III (-, -) consisted of nine different populations and was influenced by two growth characteristics, i.e., internodal length and branch angle. The cluster IV corresponding to quadrant IV (-, -) consisted of eight different populations and was influenced by one growth characteristic, i.e., root: shoot ration. The quadrants III and IV are least influenced by the seedling growth characteristics under study.

Genetic divergence analysis divided thirty-five populations into two major clusters and three sub-clusters based on different seedling growth characteristics under consideration and presented through a dendrogram (Fig. 3) using Ward's method. Cluster I and Cluster II comprised of 23

Table 5. Principal component analysis of different growth characteristics of *Grewia optiva* progenies

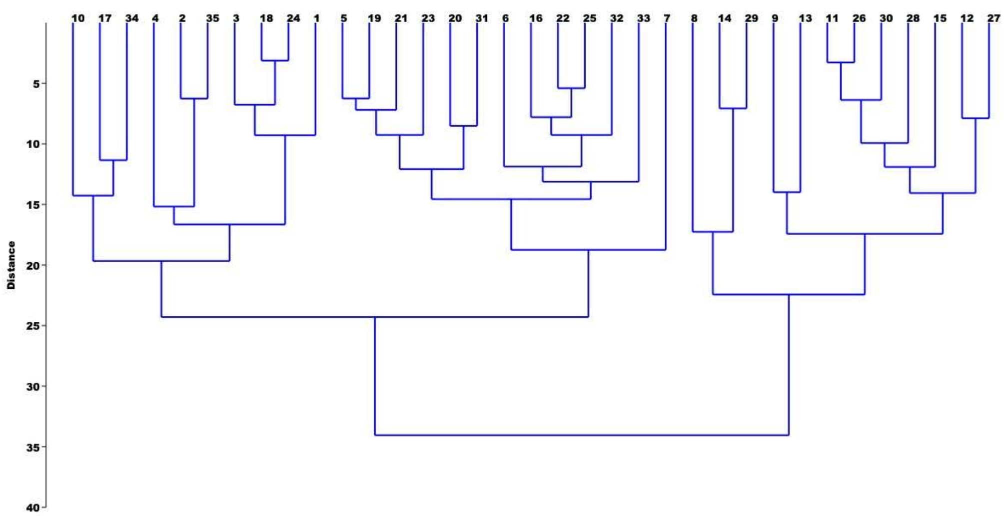
Parameters	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6	PC 7	PC 8	PC 9	PC 10
SH	0.417	0.098	-0.005	0.187	-0.215	0.05	0.156	0.625	-0.533	0.186
RS	-0.318	-0.287	0.064	-0.206	-0.288	0.747	0.138	0.078	0.055	0.317
BD	0.301	0.129	-0.527	0.122	-0.487	0.072	0.239	-0.546	-0.049	0.032
NB	0.303	0.338	0.181	-0.234	0.54	0.368	0.012	-0.368	-0.331	0.186
IL	-0.01	0.371	0.731	0.286	-0.442	0.009	-0.067	-0.2	0.081	0.006
BA	-0.251	0.037	-0.155	0.844	0.327	0.294	0.058	-0.019	-0.019	-0.05
LA	0.385	-0.154	-0.073	0.062	-0.103	0.352	-0.758	0.04	0.085	-0.317
LL	0.32	-0.492	0.143	0.23	0.097	-0.222	-0.118	-0.17	0.178	0.67
LB	0.249	-0.563	0.319	0.047	0.051	0.066	0.432	-0.158	-0.151	-0.526
NL	0.409	0.236	-0.007	-0.01	0.134	0.186	0.34	0.273	0.73	-0.04
Eigenvalue	4.443	1.609	1.055	0.800	0.697	0.545	0.362	0.258	0.138	0.093
% variance	44.433	16.088	10.553	8.004	6.974	5.450	3.616	2.578	1.378	0.928
Cumulative variance	44.433	60.521	71.074	79.078	86.051	91.501	95.117	97.695	99.072	100.000

See Table 2 for details



Where, 1-Kothi kanwal population (SO-1), 2- Unchagaon population (SO-2), 3- Nerikalan (SO-3), 4- Gaddo (SO-4), 5-Devwra- (SO-5), 6-Machair (SI-1), 7-Jajjar(SI-2), 8-Neherbhag (SI-3), 9- Badon (SI-4), 10-Dharkyari (SI-5), 11- Kant (UN-1), 12- Navami (UN-2), 13-Kharunibangana (UN-3), 14- Thanakalan (UN-4), 15-Lamlehri (UN-5), 16- Katoi, (KA-1), 17-Balugloa (KA-2), 18- Purana kangra (KA-3), 19- Dohan (KA-4), 20-Balla (KA-5), 21- Janhen (HA-1), 22- Jhinkari (HA-2), 23-Harbalneri (HA-3), 24-Anu khurd (HA-4), 25- Bhaleth (HA-5), 26- Patta (MA-1), 27- Gangal (MA-2), 28- Bagla (MA-3), 29- Balt (MA-4), 30-Bharnoi (MA-5), 31- Ghumarwin (BI-1), 32- Barthi (BI-2), 33- Kuthera (BI-3), 34- Jukhala (BI-4), 35- Nehari (BI-5)

Fig. 2. Biplot between principal component 1 and 2



See Fig 2 for details, SH-seedling height, R/S-root/shoot ratio, BD-basal diameter, NB- number of branches, IL-internodal length, BA- branch angle, LA- leaf area, LL- leaf length, LB- leaf breadth, NL- number of leaves

Fig. 3. Dendrogram depicting genetic divergence of thirty – five populations of Grewia optiva

and 12 different populations, respectively. The maximum inter-cluster distance was observed between population no. 10 (Dharkyari, Sirmaur District) and population no. 27 (Gangal, Mandi District). Whereas, the maximum intra-cluster distance was observed between population no. 7 (Jajjar, Sirmaur District) and population no. 10 (Dharkyari, Sirmaur District) in cluster I and population no. 8 (Neharbag, Sirmaur District) and population no. 27 (Gangal, Mandi district) in cluster II. In a study conducted by Navya et al (2021) in sorghum (*Sorghum bicolor* L) reported that 20 genotypes were classified into four distinct clusters. The maximum (6) and lowest (4) number of genotypes were in clusters I and IV respectively. Clusters II and IV (150.99) and cluster III and IV (150.99) had the greatest and smallest inter cluster. Therefore, hybridization between the progenies of distant populations may produce more hybrid vigour. Similar results were reported by Sehgal et al (1995) in chir pine, Behera et al (2017) in *Eucalyptus,* Kumar et al (2016) in *Dalbergia sissoo,* Kundal et al (2020) in Toona ciliata and Mohanraj et al (2022) in *Toona ciliata.*

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Studies on Natural Regeneration of Dollar Earning Parasite (*Santalum album* Linn.) in Himachal Pradesh

Sachin Verma, Aditi Sharma and Vimal Chauhan[1]

Department of Biodiversity and Climate Change, Indian Council of Forestry Research and Education Dehradun-248 006, India

[1]Department of Silviculture and Agroforestry, Dr. Yashwant Singh Parmar University of Horticulture and Forestry Nauni, Solan-173 230, India

E-mail: sachinsilvi@gmail.com

***Abstract*:** The present investigation was conducted in two different sites *viz.,* Jawalaji (Kangra) and Dholra (Bilaspur) during 2020-2022. Seeds of *Santalum album* L. are dormant due to the presence of hard seed coat which results in poor natural regeneration and slow growth rate. The *Santalum album* L. was found to be the dominant tree species in selected sites and co-dominated species were *Dalbergia sissoo, Acacia catechu* or *Lannea coromendelica*. The dominant shrub species were *Lantana camara* and *Carrisa carandus*. *Cymbopogon martini* and *Chrysopogon montanus* were dominant herbaceous vegetation of Jawalaji (Kangra) and Dholra (Bilaspur) site. The maximum successful regeneration (8.50%) was recorded at Dholra (Bilaspur) site.

***Keywords*:** Regeneration, Sandalwood, Dominant, Parasite

Santalum album L. belongs to family *Santalaceae*, family consists of 29 genera with more than 400 species, out of which 19 species are useful for oil aromatic purpose. One species *Santalum fernandezianum* has been reported to be extinct (Harbaugh 2007, Harbaugh and Baldwin 2007, Harbaugh et al 2010, Butaud, 2015). The global distribution of the sandalwood is between 30˚N and 40˚S from Indonesia in West to Juan Fernandez Island in the north to New Zealand in the South. The species is mainly found in India, Indonesia and Australia. In India, its distribution is mostly in the deciduous forests of the Deccan region of Peninsular India and mainly growing naturally in the states of Karnataka and Tamil Nadu. In Himachal Pradesh, the sandalwood is found growing naturally in districts of Kangra, Bilaspur and Sirmour. There were 3,000 fully grown sandalwood trees on a forest land near Jwalamukhi temple by the end of 2011 and the number touched 3,998 by December 2018.

The main reason for the economic and cultural value of sandalwood is the oil contained mainly in the heartwood. Heartwood oil content varies, widely between species and even within species. The Indian sandalwood is valued for its oil, which is highly rated for its sweetness, fragrance, persistent aroma and the fixative property and demanded by the perfumery industry. Heartwood oil priced at 22,000 Indian rupees per kg (Jain et al 2003). Being a root hemi-parasite, sandalwood depends upon host plants for nutrients and water for survival and growth. Host plants with the nitrogen-fixing ability and light shade appear to be the most suitable for good sandalwood growth (Silva et al 2016). Production of *Santalum album* L. has been decreased due to mismanagement (over-exploitation) in the past, inadequate plantation and establishment techniques and lack of support of the local community in planting and maintaining sandalwood trees. Stringent legal control and centralized authority on sandalwood resource, management, utilization and trade are the factors which have resulted in the low interest of the private sector towards investing in sandalwood plantation (Butar et al 2007).

The tree is now on the verge of extinction due to over exploitation, though much of work has been done by the government to protect the tree plantation by applying strict laws. But, still the use of new techniques and biotechnological methods like that of rapid mass propagation such as *in-vivo, in-vitro* and micro grafting are required. In addition to that, proper storage of seeds is also an important factor because large number of seeds fails to develop due to improper storage and poor cultivation methods (Solanki et al 2015). The major constraints in raising large-scale plantations of sandalwood are the poor germination rate, prolonged seed germination period and slow rate of field establishment (Anandalakshmi et al 2019). Das and Tah (2013) reported physical dormancy due to hard seed coat which results in poor germination of sandalwood. Therefore keeping in view, the above facts, the present investigation has been carried out to study on natural regeneration of *Santalum album* L. in low hills of Himachal Pradesh.

MATERIAL AND METHODS

Climate and Location: The study area falls under sub-tropical sub montane low hill zone with 31.8756˚N latitude and 76.3243˚E longitude, which receives about 518mm precipitation annually and the major part of which is received during July and August (monsoon period). Jawalaji (Kangra) is situated at 650m amsl, April to June are the hottest months with temperature ranges up to 38˚C, whereas mid-November to mid-March are the coldest with temperature sometimes reaching to freezing point (Fig. 1). The sandalwood trees were grown naturally near the Jawalaji temple and in the private lands. The soils of the location were shallow, embedded with stones. Bilaspur district has a hilly terrain. The district is situated in the Shivalik range of the lower Himalayas. Dholra is situated, along the left bank of river Govind Sagar Lake. The study area falls under sub-tropical sub montane low hill zone, with 31.3330˚N latitude and 76.7584˚E longitude with an elevation of 600m amsl, which experiences warm summers and cool winters but is protected from the temperature extremes of the surrounding mountains and its situation in valley. The monsoons are received from July to September. The highest temperature months are May and June when the temperature reaches upto 38˚C and sometimes exceeding up to 40˚C (Fig. 1).

Floristic composition and phytosociological analysis of vegetation: Floristic and phytosociological analysis of the existing vegetation was carried out at two sites *viz.* Jawalaji (Kangra) and Dholra (Bilaspur). At each site, five quadrates each of 31.62m×31.62m for trees, 5m×5m for shrubs and 1m×1m for herbaceous components were randomly laid out. Percent frequency, density, basal area, relative frequency, relative density, relative dominance and important value index were calculated for each species (Raunkiaer 1934, Mishra 1968, Menon and Balsubramanyan 1985).

Regeneration studies: The regeneration survey of two sites was carried out in all the major sample plots. Within each sample plot (31.62m×31.62m) three sub-quadrat per plot of size (2m×2m) for regeneration studies were laid down. To express satisfactory regeneration 2500 established plant per hectare were desired. The quadrate was considered fully stocked when it contained on established plant (Chacko 1965). The regeneration survey was conducted from recruits (r) which may be defined as current years seedlings, unestablished regeneration (u) seedlings other than recruits

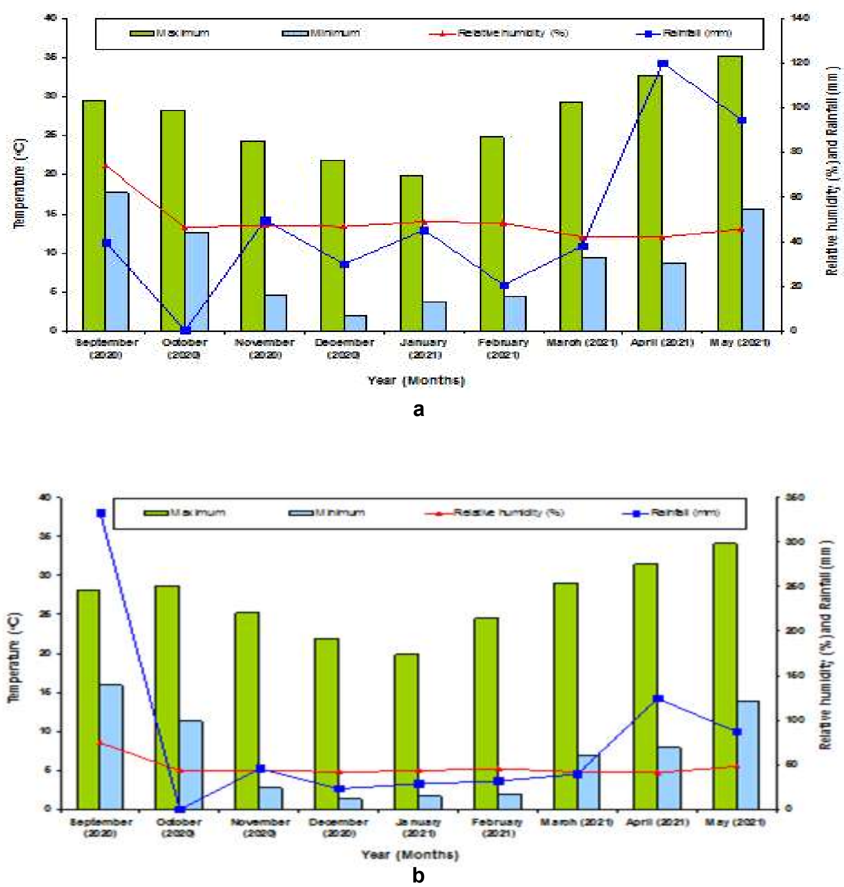

Fig. 1. Meteorological data of a) Jawalaji (Kangra) b) Dholra (Bilaspur) site on monthly basis during period of study (September 2020 - May 2021)

which has not established and whose height was less than 2 m, here four established plants were taken equivalent to one established plant and established regeneration (e) having height more than 2 m.

RESULTS AND DISCUSSION

Floristic composition and phytosociology: The maximum number (9) of tree species were observed in Dholra (Bilaspur) site and the minimum number (7) of species were in Jawalaji (Kangra) site (Table 1). The maximum number (7) of shrubs species were recorded in Jawalaji (Kangra) site and the minimum number (6) of shrubs species in Dholra (Bilaspur) site. The maximum numbers (7) of herbaceous species were in Jawalaji (Kangra) site and minimum numbers (6) of species were in Dholra (Bilaspur) site.

Two different sites had varied floristic composition of trees, shrubs and herbaceous indicating common species. Generally common species for trees found in both the sites were *Santalum album, Acacia catechu, Mallotus philipensis, Leucaena Leucocephala, Dalbergia sissoo* and *Cassia*

Table 1. Floristic composition of Jawalaji (Kangra) and Dholra (Bilaspur) sites in Himachal Pradesh

Name of species	Family	Jawalaji (Kangra)	Dholra (Bilaspur)	Total
Trees				
Santalum album	Santalaceae	+	+	2
Acacia catechu	Legumeaceae	+	+	2
Mallotus philipensis	Spurgeaceae	+	+	2
Bombax ceiba	Bombacaeae	+	-	1
Albizzia lebbeck	Legumeaceae	-	+	1
Lannea coromandelica	Anacardiaceae	-	+	1
Leucaena Leucocephala	Legumeaceae	+	+	2
Dalbergia sissoo	Legumeaceae	+	+	2
Cassia fistula	Legumeaceae	+	+	2
Grewia optiva	Malvaceae	-	+	1
Total		7	9	16
Shrubs				
Lantana camara	Verbenaceae	+	+	2
Murraya koengii	Rutaceae	+	-	1
Justacia adhatoda	Acanthaceae	+	+	2
Carissa carandas	Apocynaceae	+	+	2
Zizyphus numularia	Rhamnaceae	+	+	2
Asparagus adscendens	Asparagaceae	-	+	1
Dodonea viscosa	Sapindaceae	+	+	2
Agave Americana	Asparagaceae	+	-	1
Total		7	6	13
Herbaceous				
Ageratum conyziodes	Asteraceae	+	+	2
Bidens pilosa	Asteraceae	+	+	2
Eupatorium adenophorum	Asteraceae	+	+	2
Achyranthus aspera	Amaranthaceae	-	+	1
Ageratum houstonianum	Asteraceae	+	-	1
Chrysopogon montanus	Poaceae	+	+	2
Cymbopogon martini	Poaceae	+	-	1
Heteropogon contortus	Poaceae	+	-	1
Dicanthium anulatum	Poaceae	-	+	1
Total		7	6	13

+ = present; - = not present

fistula. Common species of shrubs were *Lantana camara, Justacia adhatoda, Carissa carandas, Zizyphus numularia* and *Dodonea viscosa.* In herbaceous, the common species consisted of *Ageratum conyziodes, Bidens pilosa, Eupatorium adenophorum* and *Chrysopogon montanus.* In the present investigation, species diversity in tree component was recorded highest in Dholra (Bilaspur) site. The species diversity of shrubs and herbaceous component was observed highest in Jawalaji (Kangra) site. Seven species of trees, seven species of shrubs and seven species of herbaceous in which four species of herbs and three species of grasses were in Jawalaji (Kangra) site (Table 2). The tree species in all reported a total density of 294.00 no. ha^{-1}, percent frequency 280.00, basal area of 12.71 $cm^2 ha^{-1}$ and IVI of 300. Maximum density (212.00 no. ha^{-1}), per cent frequency (100.00), basal area (10.14 $cm^2 ha^{-1}$) and IVI (187.58) in case of trees were in *S. album* and minimum density (2.00 no. ha^{-1}), percent frequency (20.00), basal area (0.08 $cm^2 ha^{-1}$) and IVI (8.42) were recorded in *Bombax cieba* whereas *L. leucocephala* (0.03 $cm^2 ha^{-1}$) showed minimum basal area (0.05 $cm^2 ha^{-1}$) followed by *C. fistula* . The shrub species in all reported a total density of 2416.00 no. ha^{-1}, percent frequency 304.00, basal area of 23046.00 $cm^2 ha^{-1}$ and IVI of 300 . Maximum density (736.00 no. ha^{-1}), percent frequency (72.00) were in *Murraya koengii* whereas, maximum basal area (5726.07 $cm^2 ha^{-1}$) and IVI (64.43) were recorded in *Lantana camara*. Minimum density (80.00 no. ha^{-1}) and IVI (25.17) were in *Agave Americana*. Minimum percent frequency (16.00) was observed in *Carrisa carandas* while minimum basal area (987.42 $cm^2 ha^{-1}$) was in *J. adhatoda.*

Table 2. Phytosociological parameters of vegetation in sandalwood forest at Jawalaji (Kangra) site in Himachal Pradesh

Name of species	Density (No. ha^{-1})	Frequency (%)	Basal area (m^2/cm^2 ha^{-1})	RD	RF	RBA	IVI
Trees							
Santalum album	212.00	100.00	10.14	72.11	35.71	79.76	187.58
Acacia catechu	18.00	40.00	1.08	6.12	14.29	8.51	28.92
Mallotus philipensis	24.00	40.00	0.12	8.16	14.29	0.92	23.37
Bombax ceiba	2.00	20.00	0.08	0.68	7.14	0.60	8.42
Leucaena Leucocephala	10.00	20.00	0.03	3.40	7.14	0.21	10.75
Dalbergia sissoo	20.00	40.00	1.22	6.80	14.29	9.61	30.70
Cassia fistula	8.00	20.00	0.05	2.72	7.14	0.40	10.26
Total	294.00	280.00	12.71	100.00	100.00	100.00	300.00
Shrubs							
Lantana camara	416.00	68.00	5726.07	17.22	22.37	24.85	64.43
Murraya koengii	736.00	72.00	1037.79	30.46	23.68	4.50	58.65
Justacia adhatoda	608.00	60.00	987.42	25.17	19.74	4.28	49.19
Zizyphus numularia	208.00	44.00	2159.72	8.61	14.47	9.37	32.45
Carissa carandas	160.00	16.00	7160.88	6.62	5.26	31.07	42.96
Dodonea viscosa	208.00	24.00	2452.29	8.61	7.89	10.64	27.14
Agave Americana	80.00	20.00	3522.16	3.31	6.58	15.28	25.17
Total	2416.00	304.00	23046.34	100.00	100.00	100.00	300.00
Herbaceous							
Ageratum conyziodes	8000.00	63.20	2930.82	3.86	18.99	5.62	28.46
Bidens pilosa	6160.00	28.00	3626.98	2.97	8.41	6.95	18.33
Eupatorium adenophorum	8400.00	57.60	3949.98	4.05	17.31	7.57	28.92
Ageratum houstonianum	5280.00	42.40	2039.22	2.54	12.74	3.91	19.19
Chrysopogon montanus	67040.00	61.60	10680.98	32.31	18.51	20.46	71.28
Cymbopogon martini	67280.00	39.20	19800.09	32.42	11.78	37.94	82.14
Heteropogon contortus	45360.00	40.80	9163.53	21.86	12.26	17.56	51.68
Total	207520.00	332.80	52191.60	100.00	100.00	100.00	300.00

*m^2= Basal area (trees)
*cm^2= Basal area (shrubs and herbaceous component)

The herbaceous vegetation reported a total density of 207520.00 no. ha^{-1}, percent frequency 332.80, basal area of 52191.60 cm^{2}ha^{-1} and IVI of 300. Maximum density (8400.00 no. ha^{-1}), basal area (3949.98 cm^2 ha^{-1}) and IVI (28.92) in f herbs were observed in *Eupatorium adenophorum* whereas, maximum percent frequency (63.20.00) were in *Ageratum conziodes* and minimum density (5280.00 no. ha^{-1}), basal area (2039.22 cm^2 ha^{-1}) were observed in *Ageratum houstanianum* whereas minimum percent frequency (28.00) and IVI (18.33) were recorded in *Bidens pilosa.* In case of grasses maximum density (67280.00 no. ha^{-1}), basal area (19800.00 cm^2 ha^{-1}) and IVI (82.14) were observed in *Cymbopogon martinii* whereas, percent frequency (61.60) were observed in *C. montanus* and minimum density (45360.00 no. ha^{-1}), basal area (9163.53 cm^2 ha^{-1}) and IVI (51.68) were recorded in *Heteropogon contatus* whereas minimum percent frequency (39.32) were observed in *C. martini.*

Trees species (9), shrubs (6) and herbaceous (6) were found in Dholra (Bilaspur) site. The tree species reported a total density of 272.00 no. ha^{-1}, percent frequency 400.00, basal area of 7.51 cm^{2}ha^{-1} and IVI of 300 (Table 3). Maximum density (148.00 no. ha^{-1}), percent frequency (100.00), basal area (5.97 cm^2 ha^{-1}) and IVI (158.94) in *S. album.* The minimum density (6.00 no. ha^{-1}), percent frequency (20.00), basal area (0.01 cm^2 ha^{-1}) and IVI (7.23) were recorded in *Grewia optiva.* The shrub species reported a total density of 6160.00 no. ha^{-1}, percent frequency 192.00, basal area of 80522.08 cm^{2}ha^{-1} and IVI of 300 (Table 3). Maximum density (1760.00 no. ha^{-1}), percent frequency (68.00), basal area

Table 3. Phytosociological parameters of vegetation in sandalwood forest at Dholra (Bilaspur) site in Himachal Pradesh

Name of species	Density (No. ha^{-1})	Frequency (%)	Basal area (m^2/cm^2 ha^{-1})	RD	RF	RBA	IVI
Trees							
Santalum album	148.00	100.00	5.97	54.41	25.00	79.52	158.94
Acacia catechu	10.00	40.00	0.09	3.68	10.00	1.26	14.94
Mallotus philipensis	18.00	60.00	0.03	6.62	15.00	0.43	22.05
Albizzia lebbeck	16.00	40.00	0.04	5.88	10.00	0.59	16.47
Lannea coromandelica	28.00	40.00	0.44	10.29	10.00	5.88	26.17
Leucaena leucocephala	18.00	40.00	0.03	6.62	10.00	0.36	16.97
Cassia fistula	8.00	20.00	0.06	2.94	5.00	0.80	8.74
Dalbergia sissoo	20.00	40.00	0.84	7.35	10.00	11.14	28.49
Grewia optiva	6.00	20.00	0.01	2.21	5.00	0.02	7.23
Total	272.00	400.00	7.51	100.00	100.00	100.00	300.00
Shrubs							
Lantana camara	960.00	40.00	18179.16	15.58	20.83	22.58	58.99
Carissa carandas	1760.00	68.00	42807.50	28.57	35.42	53.16	117.15
Justacia adhatoda	1360.00	32.00	1916.81	22.08	16.67	2.38	41.13
Zizyphus numularia	400.00	12.00	6725.14	6.49	6.25	8.35	21.10
Asparagus adscendens	720.00	24.00	593.16	11.69	12.50	0.74	24.92
Dodonea viscosa	960.00	16.00	10300.30	15.58	8.33	12.79	36.71
Total	6160.00	192.00	80522.08	100.00	100.00	100.00	300.00
Herbaceous							
Ageratum conyziodes	4200.00	35.20	812.63	2.18	13.71	1.23	17.12
Bidens pilosa	2933.33	17.60	1647.31	1.52	6.85	2.50	10.88
Eupatorium adenophorum	4733.33	44.00	2051.50	2.45	17.13	3.12	22.70
Achyranthus aspera	4266.67	40.00	892.50	2.21	15.58	1.36	19.14
Chrysopogon montanus	133360.00	76.00	59478.17	69.14	29.60	90.36	189.10
Dicanthium anulatum	43401.60	44.00	938.17	22.50	17.13	1.43	41.06
Total	192894.93	256.80	65820.28	100.00	100.00	100.00	300.00

*m^2= Basal area (trees)
*cm^2= Basal area (shrubs and herbaceous component)

(42807.50 cm^{2}ha^{-1}) and IVI (117.15) in shrubs were observed in *C. carandus*. The minimum density (400.00 no. ha^{-1}), percent frequency (12.00) and IVI (21.10) were in *Z. numularia* and the minimum basal area (593.16 cm^2 ha^{-1}) were observed in *Asparagus adscendens.* The herbaceous species in all reported a total density of 192894.93 no. ha^{-1}, frequency 256.80 per cent, basal area of 65820.28 cm^2 ha^{-1} and IVI of 300 (Table 3). Maximum density (4733.00 no. ha^{-1}), percent frequency (44.00), basal area (2051.50 cm^{2}ha^{-1}) and IVI (22.70) in case of herbs were observed in *Eupatorium adenophorum* and minimum density (2933.33 no. ha^{-1}), percent frequency (17.60) and IVI (10.88) were in *B. pilosa* whereas *A. conyziodes* showed minimum basal area (812.63 cm^2 ha^{-1}). In case of grasses maximum density (133360.00 no. ha^{-1}), percent frequency (76.00), basal area (59478.17 cm^{2}ha^{-1}) and IVI (189.10) were observed in *C. montanus* and minimum density (43401.60 no. ha^{-1}), percent frequency (44.00), basal area (938.17 cm^{2}ha^{-1}) and IVI (41.06) were in *Dicanthium anulatum.*

Dutt et al (2021) also revealed that the Jawalamukhi site showed maximum dominance of *S. album* L. Maximum IVI for *S. album* L. is attributed to its higher frequency, basal area and density. Among shrubs, *C. carandus* dominated the site. *C. montanus* was found to be the most dominant grass with an IVI (186.04) which is due to its high frequency and basal area.

In Bilaspur *S. album* dominated the vegetation with maximum IVI (87.51) which is attributed to its higher frequency, basal area and density. The most dominant shrub at Bilaspur site was *L. camara* (IVI 51.99). *C. montanus* (IVI 145.49) dominated the different grass species, owing to its high density, frequency and basal area. Sreejith et al (2016) studied the flora in Marappalam forest in Kerala and reported that the forest includes a total of 20 species in which sandalwood excels all other species, indicating 100 per cent frequency of distribution, total density more than 38 per cent, total basal area more than 36 per cent with 31 per cent of importance value index (IVI). Huish et al (2015) reported the population densities of *Santalum. yasi* ranging from 19 to 63 trees ha^{-1} in Fiji and Tonga. Bahadur, (2019) reported that sandalwood showed maximum relative density (21.67%) and relative frequency (16.67%).

Regeneration status: In Jawalaji (Kangra) site, *S. album* maximum number of recruits ha^{-1} (150.00), unestablished ha^{-1} (250.00), established ha^{-1} (50.00), establishment stocking percent (3.32%) and successful regeneration (4.50%) (Table 4). *L. leucocephala* showed number of unestablished ha^{-1} (100.00) and successful regeneration (1.00%) whereas recruit's ha^{-1}, established ha^{-1} and establishment stocking percent were found absent in *L. leucocephala.* In Dholra (Bilaspur) site *S. album* showed maximum number of recruits ha^{-1} (450.00), unestablished ha^{-1} (450.00), established ha^{-1} (100.00), establishment stocking percent (4.59%) and regeneration successful (8.50%) as compared to other species followed by *L. leucocephala,* which showed unestablished ha^{-1} (250.00), established ha^{-1} (100.00), establishment stocking percent (4.93%) and regeneration successful (6.50%). *A. lebbeck* showed recruits ha^{-1} (250.00), unestablished ha^{-1} (200.00), established ha^{-1} (50.00), establishment stocking percent (2.28%) and regeneration successful (4.00%). *M. phillipensis* showed recruits ha^{-1} (200.00), established ha^{-1} (50.00), establishment stocking percent (2.00%) and regeneration successful (2.00%) whereas recruit's ha^{-1} in *L. leucocephala* and unestablished ha^{-1} in *M. phillipensis* were found absent. The maximum recruits ha^{-1} (900.00), unestablished ha^{-1} (900.00), established ha^{-1} (300.00), establishment stocking percent (13.81%) and regeneration successful (21.00%) were recorded in Dholra (Bilaspur) site and minimum recruits ha^{-1} (150.00), unestablished ha^{-1}

Table 4. Regeneration status at Jawalaji (Kangra) and Dholra (Bilaspur) site in Himachal Pradesh

Name of species	Recruits (ha^{-1})	Unestablished (ha^{-1})	Established (ha^{-1})	Establishment stocking percent (%)	Regeneration successful (%)
Jawalaji (Kangra)					
Santalum album	150.00	250.00	50.00	3.32	4.50
Leucaena leucocephala	-	100.00	-	-	1.00
Total	150.00	350.00	50.00	3.32	5.50
Dholra (Bilaspur)					
Santalum album	450.00	450.00	100.00	4.59	8.50
Leucaena leucocephala	-	250.00	100.00	4.93	6.50
Mallotus phillipensis	200.00	-	50.00	2.00	2.00
Albizzia lebbeck	250.00	200.00	50.00	2.28	4.00
Total	900.00	900.00	300.00	13.81	21.00

(350.00), established ha^{-1} (50.00), establishment stocking percent (3.32%) and regeneration successful (5.50%) in Jawalaji (Kangra) site.

CONCLUSION

Santalum album was dominant tree species in selected sites and co-dominated species were *Dalbergia sissoo*, *Acacia catechu and Lannea coromandelica*. The dominant shrub species were *Lantana camara* and *Carrisa carandus*. *Cymbopogon martini* and *Chrysopogon montanus* were dominant herbaceous vegetation in Jawalaji (Kangra) and Dholra (Bilaspur) site. The regeneration of sandalwood was found maximum in Dholra (Bilaspur) site as compared to the minimum regeneration observed at Jawalaji (Kangra) site.

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Floristic Composition and Distribution of Plant Communities Under different Traditional Agroforestry Systems in Takoli Gad Watershed of Garhwal Himalayas

Jitendra Singh, D.S. Chauhan and Atul Negi

Department of Forestry and Natural Resources, HNB Garhwal University
Srinagar Garhwal-246 174, India
E-mail: singhjitu264@gmail.com

Abstract: The study was carried out in Lower (300-1200 masl) and Middle (1200-2000 masl) altitudinal ranges of the Takoli Gad Watershed of Garhwal Himalaya. Quadrats of 10×10 m, 5×5 m and 1×1 m were randomly laid out for trees, shrubs, crops and herbaceous plants in each system, respectively. Agri-silviculture, silvi-pasture, and homegardens were recognized as traditional Agroforestry systems. A total of 19 tree species, 10 agricultural crops and 6 herb species were recorded in Agri-silvicultural system, whereas 23 tree species, 15 agricultural crops which include cash crops and 6 herb species were recorded in home garden. In silvi-pastoral systems, 19 tree, 13 shrubs, 9 herb and 10 grass species were documented. Maximum diversity was in the home garden system owing to the inclusion of fruit trees and other multipurpose tree species. Agri-silviculture systems are generally utilized for food production, whereas home garden systems are mostly used for subsistence purposes and silvi-pasture systems are mostly used for fodder and fuelwood production. From the current study it can be conclude that the traditional agroforestry systems are important for the livelihood support of local people and biodiversity conservation to protect the depletion of natural resources.

Keywords: Traditional Agroforestry, Agri-silviculture, Home garden, Silvipasture, Phytosociology

Recent population development has put more strain on natural resources, such as land availability for maintaining lifestyles. Additionally, due to overuse and extraction of natural resources, ecosystems are becoming unstable and fragile (Sundriyal and Sharma 1996). In this context, agroforestry plays an important role not only to sustain the natural balance but also providing livelihood securities to the local people. Agroforestry is commonly considered as 'a low-hanging fruit" due to its multifarious outputs such as tangible benefits (food, timber, fuelwood, fertilizer, NTFPs) and intangible (ecosystem services controlling soil erosion, water conservation, carbon sequestration, increasing climate resiliency, etc.) benefits, with the 6Fs, i.e. food, fruit, fodder, fuel, fertilizer, and fiber (Chavan et al 2015, 2016, 2022). Agroforestry has potential to enhance livelihoods in India, where people have a long history and have gathered local knowledge. India is notably noteworthy for its ethnoforestry practices and indigenous tree-growing knowledge systems. Shifting cultivation, taungya, and homegardens are three significant traditional systems that have changed through time (Sharma et al 2007, Newaj et al 2016). Several traditional Agroforestry practices from Himachal Pradesh and Uttarakhand, one of India's Himalayan regions, have been recorded (Arunachalam et al 2019, Thakur et al 2005, Thakur et al 2007). In Himachal, these practices include the three most often used Agroforestry systems: Agri-silviculture, agri-horticulture, and gri-horti-silviculture (Singh and Dagar 1990). Similarly, in the Mussoorie hills of Western Himalayas identified an Agri- silviculture system, Agri-horticulture system, Agri-horti- silviculture system, Silvi-pastoral system and Homesteads.

The production potentials in Agroforestry systems can be accessed under particular site circumstances through phytosociological study. The top layer has a bigger influence on the structure of understory species. When there is no competition, a species reacts to external stimuli differently than when there is competition. The abandonment of conventional, regionally appropriate crop types and intercropping in favor of high-yielding monocultures may have an impact on the biodiversity of the agroecosystem (Chappell and LaValle 2011, Sunderland 2011). Guillerme et al (2011) observed that introduction of exotic fast-growing multipurpose trees and the conversion of agroforestry systems (including home gardens or their parts) to monocropping production systems has resulted in a decline in the diversity of native multipurpose trees and shrubs as well as herbaceous components like traditional vegetable crops and ornamental plants. Therefore, by preserving tree species in their natural habitat on farms, easing pressure on remnant forests, and providing optimal habitat for plant and animal species on fields, traditional agroforestry methods promote biodiversity. Keeping the potential of traditional agroforestry in view, the present study was conducted with objective of to assess the biodiversity of traditional agroforestry systems.

MATERIAL AND METHODS

Study area: The current study was conducted in the 'Takoli Gad Watershed' of Uttarakhand's Tehri District. The digital elevation map of study area is shown in the Figure 1 (located between 30° 14' to 30° 23' N latitude and 78° 37' to 78° 46' E longitude). This watershed consists of 67 villages on an area of approximately 131.43 km^2. The region is in the Garhwal of the lesser Himalaya and is distinguished by gentle and mature landscape (Parmar et al 2012). Two altitudes *viz*. lower altitude (300-1200 m) and middle altitude (1200-2000 m) were taken for present study (Fig. 1). The soil of the study area is mostly acidic to neutral in nature with pH range from 6.4 to 7.3 (Parmar and Negi 2017).

Community analysis: Thorough reconnaissance field survey was carried out for identifying Traditional Agroforestry systems and species distribution. Different useful parameters viz village, altitudes, GPS locations, existing agroforestry systems, tree species, DBH, agricultural crops, grasses and weeds were recorded in each Agroforestry systems. Quadrats of 10×10m, 5×5m and 1×1m quadrats were laid out randomly for trees, shrubs, agricultural crops and herbaceous plants respectively in each system.

Quantitative analysis: The Important Value Index (IVI) which is an integrated measure of the relative frequency,

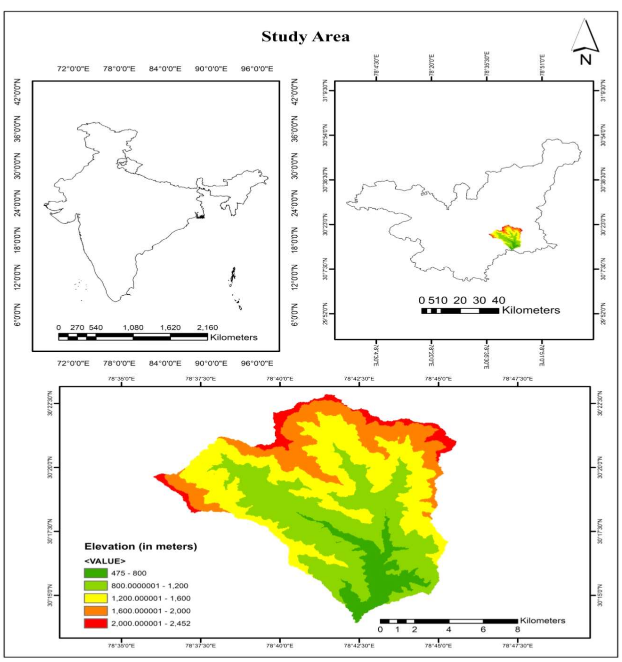

Fig. 1. Digital elevation map of study area

relative density and relative dominance/abundance was calculated for each species. The ratio of abundance to frequency indicates regular random (<0.050), contagious (0.050-1.00) and clump (>1.00) distribution patterns. The quantitative analyses for frequency, density, and abundance were done by following methodology developed by Curtis and MacIntosh (1950). Other parameters such as relative frequency, relative density, relative dominance was calculated by following Phillips (1959). The importance value index (IVI) at species level was calculated from the sum of relative frequency, relative density, and relative dominance (Curtis 1959). The ratio of abundance to frequency is generally used to interpret the distribution pattern of species (Whitford 1949). The ratio of abundance to frequency indicates regular distribution if below 0.025, random distribution between 0.025-0.05 and contagious if it is >0.05 (Curtis and Cottam 1956). The basal area of trees was calculated using the formula Basal area = $\pi d^2/4$ or $G^2/4\pi$ or πr^2 (Chaturvedi and Khanna1984).

RESULTS AND DISCUSSION

Among the different traditional agroforestry models, the three most common in the study area were agri-silviculture, silvi-pasture and homegardens. Bijalwan (2013) and Vikrant et al (2016, 2018) also reported agri-silviculture, agri-horticulture and agri-hortisilviculture in Tehri district of Garhwal Himalaya, Uttarakhand. Similarly, Kumari et al (2008) reported agri-horticulture, agri-silviculture, agri-silvi-pastoral, silvi-pastoral and horti-pastoral systems in the similar climatic conditions of Kinnaur district, Himachal Pradesh. A total of 19 tree species, 10 agricultural crops and 6 herb species were present in agri-silvicultural system whereas 23 tree species, 15 agricultural crops which include cash crops too and 6 herb species were recorded in aomegarden and 19 tree species, 13 species of shrubs, 9 herb species and 10 grass species were recorded in silvi-pastoral systems. Lower altitude shows more plant diversity than the middle altitude.

Agri-silvicultural system: In agri-silvicultural system, lower altitude has more biodiversity with 13 tree species, 5 herb species and 10 agricultural species than the middle altitude which has 12 trees, 4 herb and 6 agricultural species (Table 1). In both altitudes, the most abundant tree species were *G. optiva* and Celtis australis. Manzoor and Jazib (2020) also reported *G. optiva* as the most frequent tree species followed by *Pyrus persica* and *C. australis* in the agroforestry systems of Poonch District of Jammu & Kashmir. Farmers grow these species on the farm bunds because of their high fodder value. Middle altitude contains more density of trees, agricultural crops and herbs per hectare. Highest IVI for trees in middle and lower altitude was recorded for *G. optiva* in both lower

Table 1. Phytosociological attributes of Agri-silviculture system

Trees	Botanical name	Lower altitude (300-1200 masl)				Middle altitude (1200-2000 masl)			
		Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F	Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F
Trees									
Bhimal	*Grewia optiva*	191	41815.27	102.63	0.02	200	15652.00	87.25	0.03
Kharik	*Celtis australis*	118	40240.00	75.17	0.03	78	8571.44	40.34	0.04
Toon	*Toona ciliata*	36	17792.27	29.73	0.05	11	972.67	6.97	0.09
Khaina	*Ficus cunia*	9	560.18	5.14	0.11	0	0	0	0
Timla	*Ficus roxburghii*	9	765.91	5.30	0.11	33	6482.44	25.54	0.03
Kachnar	*Bauhinia variegata*	18	2412.91	8.43	0.22	22	5381.22	18.40	0.05
Subabool	*Leucaena leucocephala*	18	8077.45	15.66	0.06	0	0	0	0
Khair	*Acacia catechu*	18	1729.91	10.75	0.06	0	0	0	0
Sandhan	*Ougeinia oojeinense*	9	2541.09	6.67	0.11	22	1383.78	9.63	0.18
Dainkan	*Melia azaderach*	27	6937.36	16.63	0.08	0	0	0	0
Ruina	*Mallotus phillipensis*	18	3873.91	12.41	0.06	0	0	0	0
Kathgular	*Ficus hispida*	9	1193.09	5.63	0.11	0	0	0	0
Asan	*Terminalia elliptica*	9	1471.55	5.85	0.11	0	0	0	0
Mulberry	*Morus alba*	0	0	0	0	33	6482.44	15.52	0.07
Chanchari	*Ficus subincisa*	0	0	0	0	11	1680.33	7.89	0.09
Panyan	*Prunus cerasoides*	0	0	0	0	33	5300.89	24.00	0.03
Banj	*Quercus leucotrichophora*	0	0	0	0	33	5156.89	16.67	0.27
Phalsa	*Grewia asiatica*	0	0	0	0	11	17819.89	28.90	0.09
Jamun	*Syzygium cumini*	0	0	0	0	33	6863.89	18.89	0.27
Herbs									
Kumarr	*Bidens pilosa*	5455	18.18	34.65	0.17	20000	55.56	75.87	0.06
Tipatti	*Oxalis spp.*	13636	18.18	70.84	0.41	28889	33.33	98.98	0.26
Billygoat weed	*Ageratum conyzoides*	28182	81.82	110.01	0.04	30000	66.67	99.25	0.07
Kana	*Commelina benghalensis*	10909	36.36	53.87	0.08	0	0	0	0
Asthma plant	*Euphorbia hirta*	4545	18.18	30.63	0.14	0	0	0	0
Yellow foxtail	*Setaria pumila*	0	0	0	0	3333	11.11	25.90	0.27
Agricultural crops									
Toor	*Cajanus cajan*	2727	9.09	11.37	0.33	11111	22.22	31.17	0.23
Chaulai	*Amaranthus viridis*	18182	45.45	45.17	0.09	23333	44.44	53.79	0.12
Mandua	*Eleusine coracana*	39091	36.36	66.37	0.30	63333	44.44	100.30	0.32
Himalayan Navrangi	*Vigna umbellata*	9091	18.18	24.27	0.28	17778	33.33	43.29	0.16
Jhangora	*Echinochloa esculenta*	20000	9.09	51.72	2.42	20000	11.11	56.48	1.62
Till	*Sesamum indicum*	7273	9.09	21.99	0.88	3333	11.11	14.97	0.27
Gahat	*Macrotyloma uniflorum*	10909	18.18	27.12	0.33	0	0	0	0
Arvi	*Colocasia esculenta*	3636	9.09	13.49	0.44	0	0	0	0
Urad	*Vigna mungo*	10909	18.18	27.12	0.33	0	0	0	0
Soyabean	*Glycine max*	2727	9.09	11.37	0.33	0	0	0	0

Table 2. Phytosociological attribute of Silvi-pasture system

Trees	Botanical name	Lower altitude (300-1200 masl)				Middle altitude (1200-2000 masl)			
		Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F	Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F
Plants									
Kharik	*Celtis australis*	36	8464.09	24.42	0.03	89	15110.22	45.17	0.03
Banj	*Quercus leucotrichophora*	9	2033.36	6.03	0.11	100	23912.00	57.07	0.03
Timla	*Ficus roxburghii*	36	4854.45	18.57	0.05	89	11227.78	40.71	0.03
Bhimal	*Grewia oppositifolia*	36	2995.64	19.25	0.03	67	11750.89	35.03	0.03
Toon	*Toona ciliata*	45	22467.18	39.31	0.03	22	4018.11	10.89	0.18
Kachnar	*Bauhinia variegata*	36	6706.82	17.88	0.11	0	0.00	0.00	0.00
Harad	*Terminalia chebula*	27	8351.82	17.77	0.08	56	4924.22	19.99	0.11
Aonla	*Phyllantus emblica*	9	339.73	4.43	0.11	22	879.22	9.99	0.05
Sandhan	*Ougeinia oojeinense*	55	4215.91	21.30	0.07	33	2200.11	13.29	0.07
Khair	*Syzygium cumini*	55	8557.64	25.40	0.07	0	0.00	0.00	0.00
Amaltas	*Cassia fistula*	18	1962.64	10.07	0.06	0	0.00	0.00	0.00
Haldu	*Haldina cordifolia*	45	10706.36	25.76	0.06	0	0.00	0.00	0.00
Ruina	*Mallotus phillipensis*	64	7212.91	28.24	0.05	11	1347.67	6.04	0.09
Kanndi	*Bauhinia retusa*	36	8011.27	21.55	0.05	0	0.00	0.00	0.00
Khaina	*Ficus cunia*	36	8987.55	20.03	0.11	22	1112.22	7.55	0.18
Chanchari	*Ficus subincisa*	0	0	0	0	22	858.00	9.96	0.05
Khinna	*Falconeria insignis*	0	0	0	0	44	6403.22	25.31	0.02
Panyan	*Prunus cerasoides*	0	0	0	0	22	2091.22	11.38	0.05
Jamun	*Syzygium cumini*	0	0	0	0	22	1160.44	7.61	0.18
Shrubs									
Hisalu	*Rubus ellipticus*	73	9.09	14.94	0.22	356	33.33	40.58	0.08
Kingora	*Berberis aristata*	218	27.27	27.58	0.07	222	22.22	29.23	0.11
Lantana	*Lantana camara*	255	27.27	30.50	0.09	178	11.11	27.91	0.36
Tungla	*Rhus parviflora*	255	27.27	30.50	0.09	400	44.44	46.06	0.05
Sakina	*Indigofera tinctoria*	73	9.09	14.94	0.22	133	11.11	22.25	0.27
Dhaud	*Woodfordia fruticosa*	400	45.45	42.57	0.05	89	11.11	16.59	0.18
Kari patta	*Murraya koenigii*	509	54.55	50.95	0.04	89	11.11	16.59	0.18
Khakshu	*Boehmeria macrophylla*	327	27.27	36.36	0.11	0	0	0	0
Kharanu	*Carissa spinarum*	218	27.27	27.58	0.07	0	0	0	0
Mimosa bush	*Vachellia farnesiana*	73	9.09	14.94	0.22	0	0	0	0
Ber	*Ziziphus jujuba*	36	9.09	9.14	0.11	0	0	0	0
Kala bansa	*Eupatorium adenophorum*	0	0	0	0	800	55.56	72.88	0.06
Ameda	*Rumex hastatus*	0	0	0	0	178	11.11	27.91	0.36
Herbs									
Kumarr	*Bidens pilosa*	14545	66.67	74.29	0.07	16667	44.44	89.35	0.08
Mamira	*Thalictrum foliolosum*	909	33.33	46.08	0.16	5556	22.22	41.51	0.11
Billygoat weed	*Ageratum conyzoides*	4545	11.11	23.13	0.45	5556	11.11	45.53	0.45

Cont...

Table 2. Phytosociological attribute of Silvi-pasture system

Herbs	Botanical name	Lower altitude (300-1200 masl)				Middle altitude (1200-2000 masl)			
		Density (plants/ha)	Frequency (%)	IVI	A/F	Density (plants/ha)	Frequency (%)	IVI	A/F
Herbs									
Lechkumar	*Cynoglossum lanceolatum*	7273	44.44	58.07	0.12	5556	22.22	41.51	0.11
Ashthma plant	*Euphorbia hirta*	2727	22.22	29.03	0.18	7778	22.22	51.45	0.16
Gaajar ghaas	*Parthenium hysterophorus*	9091	11.11	23.13	0.45	0	0	0	0
Kaliko plant	*Euphorbia heterophylla*	3636	11.11	30.28	0.63	0	0	0	0
Tridex daisy	*Tridex procumbens*	7273	11.11	15.98	0.27	0	0	0	0
Kunja	*Artemisia vulgaris*	0	0	0	0	3333	11.11	30.65	0.27
Grasses									
Tachlu	*Apluda mutica*	8182	18.18	28.40	0.25	32222	66.67	74.29	0.07
Birachu	*Pennisetum species*	8182	18.18	28.40	0.25	17778	33.33	46.08	0.16
Dhaddu	*Arundinella nepalensis*	18182	36.36	47.74	0.14	24444	44.44	58.07	0.12
Gurla	*Crysopogon montanus*	44545	72.73	92.89	0.08	8889	22.22	29.03	0.18
False brome	*Brachypodium sylvaticum*	9091	27.27	30.29	0.12	5556	11.11	23.13	0.45
Black speargrass	*Heteropogon contortus*	2727	9.09	14.92	0.33	7778	11.11	30.28	0.63
Yellow bluestem	*Bothriochloa ischarmum*	2727	9.09	14.92	0.33	3333	11.11	15.98	0.27
Finger grass	*Digitaria spp.*	3636	9.09	18.50	0.44	0	0	0	0
Naaru		6364	18.18	23.94	0.19	0	0	0	0
Nut grass	*Cyprus rotundus*	0	0	0	0	5556	11.11	23.13	0.45

(102.63) and middle altitude (87.25) followed by *C. australis* (75.17 in lower and 40.34 in middle altitude). While lowest IVI was recorded for *Ficus cunia* (5.14) in lower and *Toona ciliata* (6.97) in middle altitude. Among the herb species, highest IVI was recorded for *Ageratum conyzoides* in lower altitude (110.01) as well as in middle altitude (99.25) while the minimum IVI was recorded for *Euphorbia hirta* (30.63) in lower altitude and *Setaria pumila* (25.90) in middle altitude. Maximum IVI for agricultural crop was recorded for *Eleusine coracana i.e.* 66.37 for lower altitude and 100.30 for middle altitude. While minimum IVI value was recorded for *Glycine max* (11.37) in lower altitude and *Sesamum indicum* (14.97) in middle altitude.

Silvi-pasture system: A total of 15 tree, 11 shrub, 8 herb and 9 grass species were recorded in lower altitude and 14 tree, 9 shrub, 6 herbs and 8 grass species were found in middle altitude in silvi-pasture system (Table 2). In tree layer, highest IVI was recorded for *Toona ciliata* (39.31) in lower altitude and for *Quercus leucotrichophora* (57.07) in middle altitude while the lowest IVI was recorded for *Phyllanthus emblica* (4.43) in lower altitude and for *Mallotus phillipensis* (6.04) in middle altitude. In shrub layer, *Murraya koenigii* (54.55) showed highest IVI value in lower altitude and *Eupatorium adenophorum* (72.88) showed highest IVI in middle altitude. Among the herb layer, highest IVI value was recorded for *Bidens pilosa* in both altitudes *i.e.* 74.29 in lower and 89.35 in middle altitude while the minimum IVI was recorded for *Tridex procumbens* (15.98) in lower altitude and for *Artemisia vulgaris* (30.65) in middle altitude. Among grasses maximum IVI was recorded for *Crysopogon montanus* (92.89) in lower altitude and for *Apluda mutica* (74.29) in middle altitude while the least (14.92) was recorded for *Heteropogon contortus* and *Bothriochloa ischarmum* simultaneously in lower altitude and 15.98 for *Bothriochloa ischarmum* in middle altitude. Dominance of *Murraya koenigii* in shrubs and *Crysopogon montanus* in herbs was also documented by Thakur et al (2004, 2005) in the silvi-pastoral system of Western Himalayas.

Home garden: Home garden consists of higher biodiversity than other agroforestry systems. It is due to the presence of fruit (horticultural) trees and multipurpose trees. Home garden is the system used to meet the subsistent requirements of the households in study area. As Linger (2014) suggested that the home gardens, ecologically sustainable and diversifies sustainability of local community and therefore, considered as exceptional tools for

Table 3. Phytosociological attributes of homegarden

Trees	Botanical name	Lower altitude (300-1200 masl)				Middle altitude (1200-2000 masl)			
		Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F	Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F
Tree									
Mango	*Mangifera indica*	118	18800.00	59.99	0.02	56	3622.33	25.60	0.05
Malta	*Citrus sinensis*	55	2819.18	20.33	0.04	156	12110.56	71.08	0.02
Mulberry	*Morus alba*	36	2563.45	16.94	0.03	22	1131.78	12.13	0.04
Guava	*Psidium guajava*	73	6770.27	33.94	0.02	89	6755.11	40.30	0.04
Kagzi Nimbu	*Citrus aurantiifolia*	9	771.91	4.39	0.11	0	0.00	0	0
Bhimal	*Grewia optiva*	9	1868.27	5.72	0.11	67	6888.89	33.73	0.06
Banana	*Musa paradisica*	55	7596.27	26.09	0.04	100	12974.89	50.99	0.09
Peach	*Prunus persica*	9	505.91	4.07	0.11	33	2358.22	19.46	0.03
Papaya	*Carica papaya*	73	5998.27	31.08	0.02	11	527.89	5.99	0.09
Phalsa	*Grewia asiatica*	9	7491.45	12.50	0.11	0	0.00	0	0
Chanchari	*Ficus subincisa*	9	498.73	4.06	0.11	0	0.00	0	0
Chulu	*Prunus armeniaca*	9	835.45	4.47	0.11	11	219.78	5.40	0.09
Anaar	*Punica granatum*	18	414.55	7.42	0.06	0	0.00	0	0
Kathal	*Artocarpus heterophyllus*	9	4981.27	9.47	0.11	0	0.00	0	0
Dainkan	*Melia azaderach*	27	8951.00	19.26	0.08	22	3069.67	15.86	0.04
Chabutra	*Citrus paradisi*	27	1984.91	10.86	0.08	11	896.78	6.70	0.09
Dhaura	*Anogeissus latifolia*	9	1756.82	5.58	0.11	0	0.00	0	0
Akhrot	*Juglans regia*	9	4346.82	8.71	0.11	0	0.00	0	0
Pear	*Pyrus communis*	9	1931.09	5.79	0.11	0	0.00	0	0
Nimbu	*Citrus limon*	9	1090.18	4.78	0.11	0	0.00	0	0
Apple	*Mallus domestica*	9	892.00	4.54	0.11	0	0.00	0	0
Kharik	*Celtis australis*	0	0	0	0	11	1208.33	7.30	0.09
Narangi	*Citrus aurantium*	0	0	0	0	11	249.67	5.46	0.09
Herbs									
Kumarr	*Bidens pilosa*	9091	27.27	49.85	0.12	10000	33.33	75.59	0.09
Tipatti	*Oxalis spp.*	8182	18.18	49.13	0.25	12222	22.22	83.52	0.25
Billygoat weed	*Ageratum conyzoides*	20909	63.64	91.96	0.05	7778	22.22	61.23	0.16
Gallant soldier	*Galinsoga parviflora*	1818	9.09	19.14	0.22	4444	11.11	43.93	0.36
Kana	*Commelina benghalensis*	10000	36.36	53.66	0.08	3333	11.11	35.72	0.27
Asthma plant	*Euphorbia hirta*	5455	18.18	36.26	0.17	0	0	0	0
Agricultural crops									
Mirch	*Capsicum annuum*	39091	81.82	92.91	0.06	32222	77.78	72.81	0.05
Baingan	*Solanum melongena*	1818	9.09	13.04	0.22	7778	33.33	26.77	0.07
Lauki	*Lagenaria siceraria*	909	9.09	8.08	0.11	1111	11.11	8.14	0.09
Maize	*Zea mays*	4545	18.18	21.33	0.14	4444	22.22	18.63	0.09
Maize	*Zea mays*	4545	18.18	21.33	0.14	4444	22.22	18.63	0.09
Adrak	*Zingiber officinale*	5455	18.18	24.35	0.17	13333	33.33	38.51	0.12

Cont...

Table 3. Phytosociological attributes of homegarden

Trees	Botanical name	Lower altitude (300-1200 masl)				Middle altitude (1200-2000 masl)			
		Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F	Density (Plants/ha)	Total basal cover (cm²/ha)	IVI	A/F
Arvi	*Colocasia esculenta*	14545	63.64	47.95	0.04	16667	66.67	47.12	0.04
Haldi	*Curcuma longa*	10000	45.45	35.99	0.05	5556	22.22	21.56	0.11
Karela	*Memordica charantia*	909	9.09	8.08	0.11	0	0	0	0
Tomato	*Solanum lycopersicum*	1818	18.18	12.28	0.06	0	0	0	0
Bhindi	*Abelmoschus esculentus*	2727	9.09	18.00	0.33	0	0	0	0
Sugarcane	*Saccharum officinarum*	2727	9.09	18.00	0.33	0	0	0	0
Beans	*Phaseolus vulgaris*	0	0	0	0	4444	22.22	18.63	0.09
Rai	*Brassica juncea*	0	0	0	0	2222	11.11	12.82	0.18
Reddish	*Raphanus sativus*	0	0	0	0	1111	11.11	8.14	0.09
Pudina	*Mentha spicata*	0	0	0	0	5556	11.11	26.89	0.45

biodiversity conservation. Lower altitude reported 21 trees, 6 herb and 11 agricultural species, whereas middle altitude recorded 13 trees, 5 herb, and 11 agricultural species (Table 3). Plant density was higher at the middle altitudes of the tree layer and agricultural crop layer. The highest IVI value was recorded for *Mangifera indica* (59.99) in lower altitude and for *Citrus sinensis* (71.08) in middle altitude. While the minimum value of IVI was found for *Prunus persica* (4.07) in lower altitude and for *P. armeniaca* (5.40) in middle altitude. *A. conyzoides* (91.96) in lower altitude and *Oxalis* spp. (83.52) in middle altitude had the highest IVI values in the herb layer. Minimum IVI value for herbs was recorded for *Galinsoga parviflora* (19.14) in lower altitude and *Commelina benghalensis* (35.72) in middle altitude. *Capsicum annuum* was the most prominent agricultural crop in both altitudes, with an IVI value of 92.91 in lower altitude and 72.81 in middle altitude.

CONCLUSION

Traditional agroforestry systems found in the study area support a diverse range of vegetation. People manage these systems based on their requirements. The home garden supports maximum tree density. Trees are cultivated on agricultural bunds in agri-silviculture systems, whereas in homegardens, they are grown during the farm. However, the density of herbs in the home garden is lower due to constant weeding by households. Agri-silviculture systems showed higher density of agricultural crops than homegardens owing to difference in structural compositions. Silvi-pasture system includes trees and grasses with high fodder value. The system, altitudinal gradient and climatic conditions influence the kind of species and their distribution. The current study reveals that traditional Agroforestry systems are essential not just for meeting people's basic needs for livelihood support, but also for conserving the area's biodiversity.

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Assessment of Wetland Ecosystem Services (RAWES approach) in Urban Settlement Area: A case study of Bilaspur, Chhattisgarh, India

Abhishek Maitry, Damini Sharma, Preety Shah, Gunja Baretha, Ramesh and Gunjan Patil

Department of Forestry, Wildlife & Environmental Sciences, Guru Ghasidas Vishwavidyalaya
Bilaspur, Chhattisgarh-495 009, India
E-mail: abhimaitry@gmail.com

Abstract: The wetland ecosystem provides several ecosystem services that support the life on earth and improve wellbeing of humans. In the era of rapid urbanization, the growing urban population can get benefit by implementing appropriate planning and management of wetland ecosystem services that are of prior importance. In order to comprehend the significance of the wetland ecosystem in an urban area, a study was conducted to evaluate the ecosystem services offered by 20 urban wetlands of Bilaspur, Chhattisgarh. This paper considers a case study from urban wetlands in order to understand the level of ecosystem services that they deliver. Based on the findings the highest significant positive contribution of any ecosystem service was local climate regulation (++ = 8), and primary production (+ = 15 each) was the most frequent ecosystem service making a positive contribution. In contrast, the most detrimental ecosystem services were livestock disease regulation and human disease regulation (- = 16 each), and waste disposal, water purification, and waste treatment (-- = 1). The highest Ecosystem Service Index among different categories was observed in Regulating services (0.66) and the lowest in Provisioning services (0.32). Trees growing in and around the ponds affects the ecosystem services provided by wetlands directly or indirectly. It has been observed that wetlands with the highest levels of recreation and tourism services have diversified tree species. The top 5 tree species most frequently observed around the wetlands are *Ficus religiosa, Acacia nilotica, Ficus benghalensis, Azadirachta indica* and *Peltophorum pterocarpum*. The Rapid Assessment of Wetland Ecosystem Services (RAWES) technique is significant in evaluating the deteriorating state of Bilaspur's urban wetlands as a result of disturbance caused by human settlements, which in turn diminished the urban wetlands' capacity to deliver ecosystem services.

Keywords: Wetland, Urban settlement, Ecosystem services, Rapid assessment, Degradation

Urban wetlands have been one of the most important tools in the life of Indian cities. Wetlands make essential positive contributions (McInnes et al 2016) to multiple dimensions of human wellbeing (Ghermandi et al 2010). According to the National Wetland Inventory and Assessment (Compiled by the Indian Space Research Organisation), in India, wetlands cover over 1,52,600 square kilometres that comprise 4.63 per cent of the total geographical area of the country (Bassi et al 2014). Their importance in human and urban lives grew as the population and the population-based pressures have increased recently (Mitsch and Gosselink 2000, Avishek and Nathawat 2004). The urban wetland provides a wide range of diverse benefits like basic biophysical needs (food, fresh water *etc*.), regulation of the environment, cultural enrichment and also support internal processes to ecosystems that maintain their functioning, resilience and capacities to produce more directly consumed services. Natural ecosystems provide benefits that are both generally acknowledged and poorly understood (Sharma et al 2022a) however, these benefits are not sufficiently recognised due to lack in decision-making (McInnes 2013), compromising the welfare (Faulkner 2004, Russi et al 2013) of ecosystems and many human beneficiaries such as yield benefits and economic value (Patil 2022) linked with it (Chu et al 2020, Rana and Bhardwaj 2022). The processes and activities that enable ecosystems to sustain and fulfil human life are referred to as ecosystem services (Baretha et al 2022). To evaluate these ecosystem services within the local context and at relevant scales, the Rapid Assessment of Wetland Ecosystem Services (RAWES) approach is presented as a systemic approach (McInnes and Everard 2017) for the assessment that is essential to avert prejudgements about which services are important and to assess the positive or negative contribution of these ecosystem services at local, regional, or global scales. The outputs from RAWES process can provide a qualitative assessment of the wide range of ecosystem services obtained from the wetlands and a comprehensive and rapid overview of the several benefits provided by the same across a large geographic area (Everard et al 2019).

The present study deals with the assessment of plurality of benefits or ecosystem services including provisioning services, regulating services, cultural services and supporting services provided by the selected 20 wetlands in reference to Bilaspur, Chhattisgarh, India, an area under urban settlement by implementation of RAWES approach.

MATERIAL AND METHODS

Study area: The study was conducted in the Bilaspur district of northern Chhattisgarh. One of the state's larger cities, Bilaspur, is situated 270 metres above mean sea level at 22.0797º N Latitude and 82.1409º E Longitude (Tiwari et al 2022).

The complete assessment was conducted during the period of October 2022 to December 2022. The site experienced largely dry and humid weather during the study period, with the lowest and highest temperatures of 13.40°C and 38.50°C, respectively, and an average rainfall of 5.8 mm. (Source: Climate department, TCB College of Agriculture & Research Station, Bilaspur, Chhattisgarh). Few rainy days were observed during the study period with light precipitations. The study site has several wetlands out of which the survey was conducted in 20 different wetland sites (Fig. 1) based on their characteristics of presence near urban settlements of Bilaspur, Chhattisgarh. All the selected wetlands (Fig. 2) are situated at approximately 25 kilometres from each other covering overall urban settlement areas.

Assessment of ecosystem services*:* A consensus was done to finalize the list of ecosystem services prior to conducting field assessments through consultation with resource persons who have an idea about the wetlands. As defined in the Millennium Ecosystem Assessment (2005), a total of 37 services grouped into four functional categories (Namely provisioning, regulating, cultural and supporting services) were selected for the analysis. The complete assessment was done by the assessors from October 2022 to December 2022 in 20 selected wetlands of the urban settlement area through regular visits to collect data about the site and other details of the wetland that needed to be assessed. The assessors used a combination of field observations and visual signs or indicators, combined with their ability to pose and answer a series of questions in order to evaluate the relative importance of each ecosystem service listed on the RAWES field assessment sheet. A five-point scale (Table 1) was used to record the importance of each ecosystem service. This scale is non-dimensional, insofar as there is no standard unit or measure between different points on the five-point scale.

Calculation of ESI: Obtained scores were numerically transformed for all ecosystem services, or alternatively for assessed ecosystem services within each service category

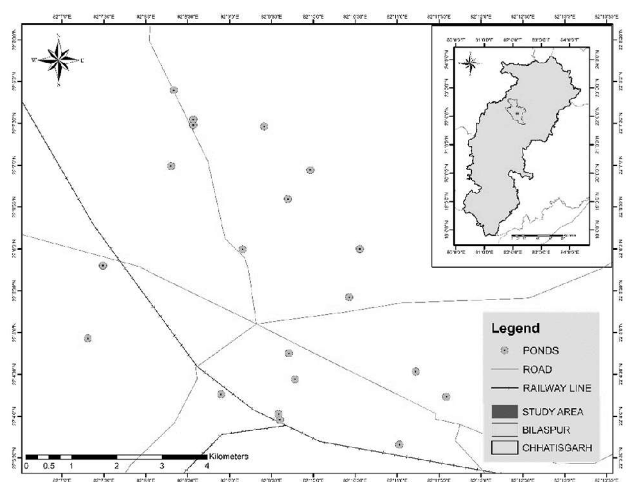

Fig. 1. Geographical location of study area locating 20 selected wetlands in Bilaspur, Chhattisgarh

Table 1. Five-point scale used to record the importance of each ecosystem service

Importance score	Numerical value	Assessment of ecosystem service	Rationale
+ +	2.0	Significant positive contribution (>1,000 people benefitting)	• Significant service provided by the wetland and a key element of its ecological character • Large number of beneficiaries (relative to wetland context)
+	1.0	Positive contribution (1-1,000 people benefitting)	• One of many services provided by the wetland and an element of its ecological character • Limited number of beneficiaries (relative to wetland context)
0	0	Negligible contribution	• No obvious beneficiaries or benefits • Not an important known part of the wetland's ecological character
-	-1.0	Negative contribution (1-1,000 people dis-benefitting)	Limited number of dis-beneficiaries
- -	-2.0	Significant negative contribution (>1,000 people dis-benefitting)	Large number of dis-beneficiaries
?	Remove from analysis	Gaps in evidence	Further evidence needs to be obtained

(provisioning, regulating, cultural and supporting), which was further analysed by deriving a comparable Ecosystem Services Index (ESI). An ESI is an index of observed ecosystem service production against potential maximum service production.

$$ESI = \frac{\sum(n_{+2.0} + n_{+1.0}) + \sum(n_{-2.0} + n_{-1.0})}{\sum(n_{Total})}$$

Assessment of scale of ESI: Through the application of RAWES approach, the benefits that wetlands provide accrue at a range of geographic scales, ranging from within the wetland itself (such as soil formation) through local, regional and up to international levels (McInnes and Everard 2017). Three scales of benefits delivery applied when conducting the RAWES assessment are:

Local benefits: Those experienced by individuals, households or communities living and working in the immediate vicinity of the wetland. (*viz.* storm buffering)

Regional benefits: Those delivered to individuals, households or communities living and working in the wider catchment of the wetland. (*viz.* flood or drought buffering across a catchment)

Global benefits: Those that extend beyond national boundaries. (*viz.* regulation of global carbon cycles)

Vegetation analysis: A vegetation survey was performed in and around the wetlands by enumerating the number of tree species present within the 15-meter radius of wetlands. The method provided a comprehensive data on vegetation composition and structure around the sites that can contribute to different functioning and services of wetlands.

Fig. 2. Water bodies selected for RAWES analysis in urban settlement areas of Bilaspur, Chhattisgarh (1. Ashok Nagar Pond, Birkona, 2. Bandhawapara Pond, 3. Bilasatal Pond, 4. Chhathghat Pond, 5. Chingrajpara Pond, 6. Deepupara Pond 1, 7. Deepupara Pond 2, 8. GGV Pond 1, 9. GGV Pond 2, 10. GGV Pond 3, 11. Ghuru Pond, Ameri, 12. Jorha Pond, Sarkanda, 13. Kalimandir Pond, Birkona, 14. Karbala Pond, 15. Mama Bhanja Pond, 16. Morum Pond, 17. Nag Nagin Pond, 18. Putha Pond, Mangla, 19. Smriti Van Pond, 20. Talapara Pond, Vyapaar Vihar)

RESULT AND DISCUSSION

Assessment of ecosystem services: Based on the field assessment and data analysis, ecosystem services making a significant positive contribution were recorded less frequently than those making a positive contribution (Table 2). The highest significant positive contribution of any ecosystem service was the local climate regulation (++ = 8) followed by aesthetic value and Nutrient cycling (++ = 6 each) and spiritual & religious value (++ = 4) also made a significant positive contribution in the study site. Water regulation and primary production (+ = 15

Table 2. Count data for the frequency of the ecosystem service scores

Ecosystem service	n	++	+	0	-	- -	L	R	G
Fresh Water	20	0	12	0	8	0	4	8	0
Food	20	0	11	9	0	0	10	1	0
Fuel	20	0	12	8	0	0	10	2	0
Fibre	20	0	1	19	0	0	1	0	0
Genetic resources	20	0	0	20	0	0	0	0	0
Natural medicines	20	0	2	18	0	0	2	0	0
Ornamental	20	0	2	18	0	0	0	2	0
Clay mineral, aggregate harvesting	20	0	6	14	0	0	2	4	0
Waste disposal	20	0	2	11	6	1	2	0	0
Energy harvesting from	20	0	1	19	0	0	1	0	0
Air quality regulation	20	0	12	3	5	0	3	7	2
local climate regulation	20	8	8	0	4	0	16	0	0
Global climate regulation	20	0	8	11	1	0	0	0	8
Water regulation	20	0	15	3	2	0	13	2	0
Flood hazard regulation	20	2	13	4	1	0	10	4	0
Storm hazard regulation	20	0	0	20	0	0	0	0	0
Pest regulation	20	0	3	5	12	0	3	0	0
Disease regulation human	20	0	1	3	16	0	1	0	0
Disease regulation livestock	20	0	0	4	16	0	0	0	0
Erosion regulation	20	0	12	6	2	0	12	0	0
Water purification	20	0	3	7	9	1	3	0	0
Pollination	20	2	8	8	2	0	1	9	0
Salinity regulation	20	0	3	17	0	0	3	0	0
Fire regulation	20	0	14	5	1	0	13	1	0
Noise visual buffering	20	0	12	6	2	0	4	8	0
Cultural heritage	20	0	3	17	0	0	0	3	0
Recreation and tourism	20	3	3	10	4	0	1	5	0
Aesthetic value	20	5	6	5	4	0	8	3	0
Spiritual and religious	20	4	9	7	0	0	3	10	0
Inspirational value	20	1	3	16	0	0	4	0	0
Social relation	20	2	12	4	2	0	14	0	0
Education and research	20	1	3	16	0	0	0	4	0
Soil formation	20	0	5	14	1	0	4	1	0
Primary production	20	2	15	2	1	0	15	2	0
Nutrients cycling	20	5	8	6	1	0	2	11	0
Water recycling	20	0	4	12	4	0	0	4	0
Provision of habitat	20	2	8	6	4	0	3	7	0

*L – Local benefits, R – Regional benefits, G – Global benefits each) were the most frequently occurring ecosystem service making a positive contribution. Fire regulation (+ = 14), flood hazard regulation (+ = 13 each) and freshwater, fuel, air quality, erosion regulation, noise and visual buffering and social relation (+ = 12 each) also made a positive contribution at more than half of all the study sites. Livestock disease regulation and human disease regulation (- = 16 each) made the most negative contributions followed by pest regulation (- = 12) in the study sites whereas waste disposal and water purification and waste treatment (-- = 1 each) made the most significant negative contributions. Out of all 37 ecosystem services, a total of 23 ecosystem services contributed negatively whereas 34 ecosystem services contributed positively in the functioning of studied urban wetlands.

Assessment of scale of ES: The benefits derived from the ecosystem services are delivered across a range of scales. For several services, there was sufficient information to make a judgement on the scale of the benefits (Fig. 3), as most of the ecosystem services were contributing to the local benefits whereas, services like nutrient cycling and facilitation of pollination were having broader ecological importance (Table 2). On the other hand, global climate regulation had a positive global benefit in the studied wetlands followed by air quality regulation. However, there were several services, where insufficient information was available to undertake an assessment of the spatial scale of benefit limiting the utility of the approach. The analysis of the information on the scale of benefit was less comprehensive than the significance of individual ecosystem services. The study reveals (Figure 4) that out of 20 total selected wetlands, GGV Pond 2 (26) provided best ecosystem service score followed by GGV Pond 1 (25), Bandhwapara Pond (22), Bilasatal Pond (20) whereas the most disturbed ecosystem service score was observed in Karbala Pond (-11) followed by Deepupara Pond 2 and Mama Bhanja Pond (-8 each). Similarly, Talapara Pond (Vyapaar Vihar) also showed negative ecosystem service scores and the rest of the studied wetlands showed positive ecosystem service scores. All these disturbed ponds are situated near the settlement area of Bilaspur city. One study was conducted in the major ponds of Bilaspur city and found that the pond near populated areas was contaminated exceeding the maximum permissible limit of WHO (Shrivastav et al 2008).

Calculation of ESI: The study of Ecosystem Services Index (Fig. 6) indicating the potentials of different ecosystem service categories that they possess in wetlands. The highest ESI was achieved by the Regulating services (0.66) which show its positive contribution to wetlands whereas the lowest ESI was observed in Provisioning services (0.32) which defines the least contribution in the services provided by the studied wetlands. However, both Cultural services (0.46) and Supporting services (0.6) achieved an ESI near about 0.5 showing their positive contribution to overall services provided by the studied urban wetlands of Bilaspur, Chhattisgarh.

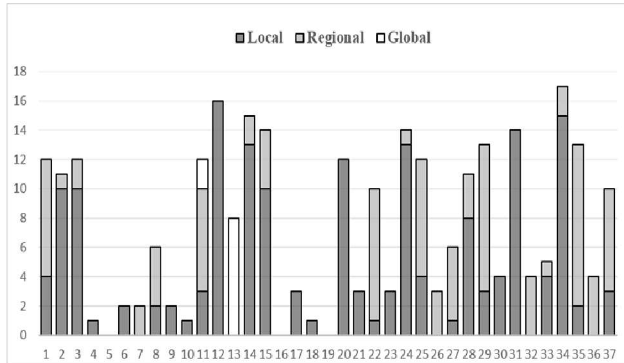

Fig. 3. Graphical representation of the scale of impact by 37 different ecosystem services on wetlands of Bilaspur, C.G.

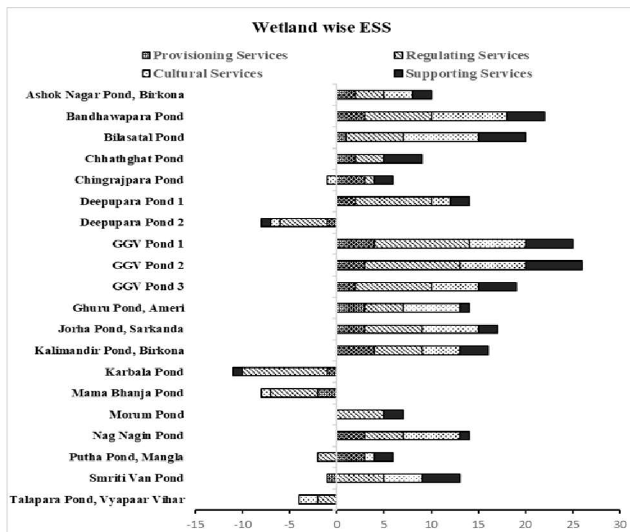

Fig. 4. Graphical representation of wetland wise ecosystem service scores of Bilaspur, C.G.

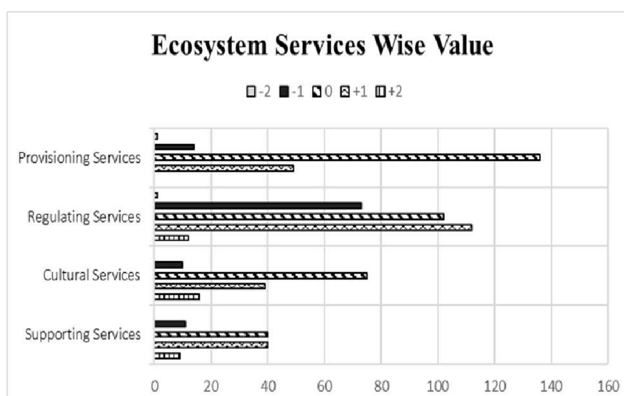

Fig. 5. Importance scores wise valuation of different ecosystem service categories

Assessment of importance scores: The evaluation of importance scores achieved by different ecosystem service categories in the 20 studied urban wetlands in which the provisioning services were dominated by negligible contributions with the least negative contributions whereas the regulating services were dominated by positive contributions but also with a maximum number of negative contributions (more than 50% of the total) when compared to other services as shown in Figure 5. Many most significant positive contributions were also observed in provisioning services whereas provisioning services and regulating services possessed an equal number of significant negative contributions (1 each).

Classifying sites based on ecosystem services: Agglomerative hierarchical clustering was done using SPSS software (version 25.0) for classifying the wetlands based on similar and dissimilar services provided by them (Fig. 7). The wetlands were divided into 4 clusters (A, B, C, D). Class B is the largest cluster (n=9) while cluster A is the smallest (n=3) and there is a high degree of similarity among them.

Cluster A represents the sites which provide a high level of regulating services score as compared to other clusters and site 10 is distinct among the two sites. Cluster B sites deliver the average ecosystem services score and represent the sites which are partially degrading. Wetlands fall in cluster C was the places with high cultural services for recreation and tourism and aesthetic value while cluster D shows the wetlands which are highly degraded and show negative services scores due to deposition of domestic waste from nearby settlement areas. The quantity and nature of domestic waste generation are influenced by the people living in a house as well as seasonal factors including summer, winter, and rainy weather (Sharma et al 2022).

Vegetation analysis: In total 65 tree species belonging to 29 families were observed near the 20 studied wetlands (Table 3). The most frequently occurring tree is *Ficus religiosa* due to its religious and spiritual values (Rutuja et al 2015) and its high invasion ability in various types of climatic and edaphic conditions (Sitaramam et al, 2009, Kumari et al 2022). The second most frequent species were *Acacia nilotica* which is arid-adapted species, and a study found the existence of most arid-adapted species in urban wetlands are drought-tolerant and need less water for growth (Avishek et al 2012) followed by *Ficus benghalensis* and *Azadirachta indica*. Vegetation analysis provided comprehensive data on vegetation composition and structure around the sites which showed that the highest number of tree species were observed in Bilasatal Pond (52) constituting 80% of the total observed species (Fig. 8) followed by Smriti Van Pond (39) and Bandhawapara Pond

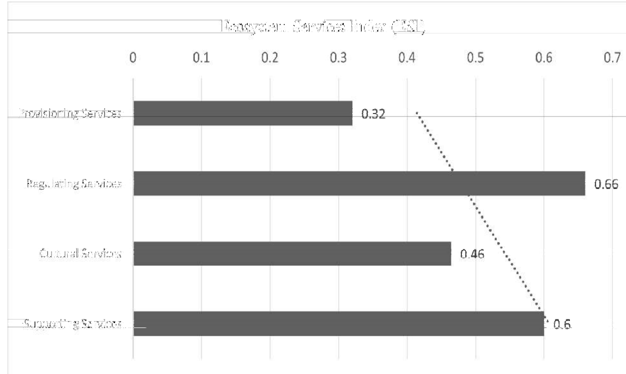

Fig. 6. Ecosystem Services Index (ESI) of different ecosystem service categories

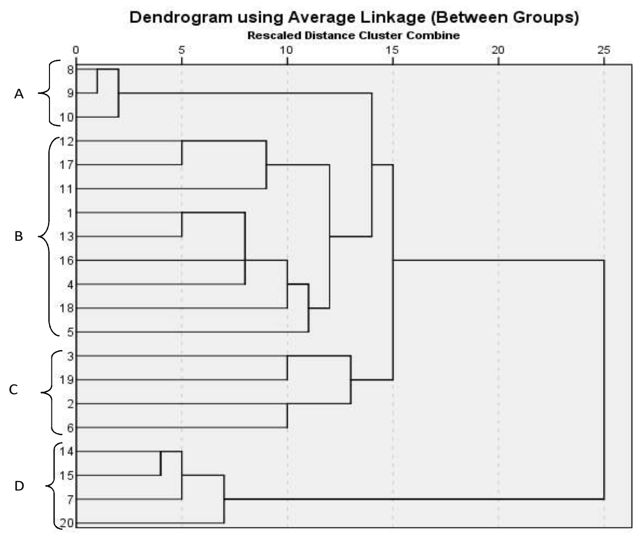

Fig. 7. Agglomerative hierarchical clustering of different wetlands sites

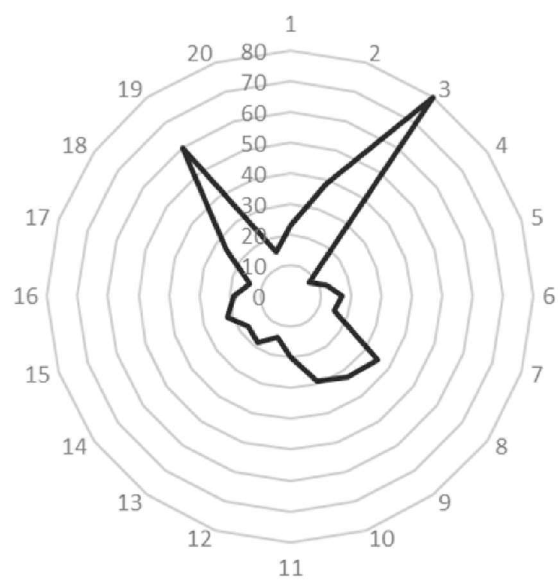

Fig. 8. Number of species observed per wetland

Table 3. Tree species distribution around studied wetlands of Bilaspur, Chhattisgarh

Common name of tree	Botanical name	Family	No. of wetlands	Frequency (%)
African Tulip	*Spathodea campanulate*	*Bignoniaceae*	5	25
Amaltash	*Cassia fistula*	*Fabaceae*	6	30
Amla	*Phyllanthus emblica*	*Phyllanthaceae*	6	30
Arjun	*Terminalia arjuna*	*Combretaceae*	2	10
Austalian Babool	*Acacia auriculiformis*	*Fabaceae*	3	15
Babool	*Acacia nilotica*	*Fabaceae*	18	90
Badam	*Prunus dulcis*	*Rosaceae*	11	55
Buddha Belly Bamboo	*Bambusa ventricose*	*Poaceae*	1	5
Male Bamboo	*Dendrocalamus strictus*	*Poaceae*	5	25
Banana	*Musa spp.*	*Musaceae*	1	5
Banyan	*Ficus benghalensis*	*Moraceae*	17	85
Beal	*Aegle marmelos*	*Rutaceae*	4	20
Ber	*Ziziphus mauritiana*	*Rhamnaceae*	9	45
Black Siris	*Albizia lebbeck*	*Fabaceae*	3	15
Bottle Brush	*Callistemon acuminatus*	*Myrtaceae*	2	10
Champa	*Magnolia champaca*	*Magnoliaceae*	2	10
Chikoo	*Manilkara zapota*	*Sapotaceae*	1	5
Euclayptus	*Eucalyptus spp.*	*Myrtaceae*	6	30
False Ashoka	*Monoon longifolium*	*Annonaceae*	7	35
Gangaimli	*Pithecellobium dulce*	*Fabaceae*	2	10
Goolar	*Ficus racemose*	*Moraceae*	6	30
Guava	*Psidium guajava*	*Myrtaceae*	5	25
Gulmohar	*Delonix regia*	*Fabaceae*	6	30
Imli	*Tamarindus indica*	*Fabaceae*	1	5
Jamun	*Syzygium cumini*	*Myrtaceae*	7	35
Jharul	*Lagerstroemia speciosa*	*Lythraceae*	1	5
Kachnar	*Bauhinia variegate*	*Fabaceae*	3	15
Kadam	*Neolamarckia cadamba*	*Rubiaceae*	8	40
Kalmi/Haldu	*Haldina cordifolia*	*Rubiaceae*	1	5
Kapok	*Ceiba pentandra*	*Malvaceae*	2	10
Karanj	*Millettia pinnata*	*Fabaceae*	11	55
Kashi	*Bridelia retusa*	*Phyllanthaceae*	2	10
Kasood	*Senna siamea*	*Fabaceae*	4	20
Katahal	*Artocarpus heterophyllus*	*Moraceae*	1	5
Khamhar	*Gmelina arborea*	*Lamiaceae*	3	15
Krishna Fig	*Ficus benghalensis var krishnae*	*Moraceae*	1	5
Lemon	*Citrus limon*	*Rutaceae*	1	5
Litchi	*Litchi chinensis*	*Sapindaceae*	1	5
Mahaneem	*Ailanthus excelsa*	*Simaroubaceae*	6	30
Mahua	*Madhuca longifolia*	*Sapotaceae*	2	10
Malshree	*Mimusops elengi*	*Sapotaceae*	3	15
Mango	*Mangifera indica*	*Anacardiaceae*	11	55
Mudhi	*Mitragyna parvifolia*	*Rubiaceae*	2	10

Cont...

Table 3. Tree species distribution around studied wetlands of Bilaspur, Chhattisgarh

Common name of tree	Botanical name	Family	No. of wetlands	Frequency (%)
Munga	*Moringa oleifera*	*Moringaceae*	12	60
Neem	*Azadirachta indica*	*Meliaceae*	17	85
Oleander	*Nerium oleander*	*Apocynaceae*	2	10
Palash	*Butea monosperma*	*Fabaceae*	7	35
Palm	*Arecales*	*Arecaceae*	5	25
Parijat	*Nyctanthes arbor-tristis*	*Oleaceae*	3	15
Peepal	*Ficus religiosa*	*Moraceae*	19	95
Peltophorum	*Peltophorum pterocarpum*	*Fabaceae*	13	65
Putranjeeva	*Putranjiva roxburghii*	*Putranjivaceae*	2	10
Rohina	*Swietenia febrifuga*	*Meliaceae*	1	5
Rudraksh	*Elaeocarpus angustifolius*	*Elaeocarpaceae*	1	5
Saja	*Terminalia elliptica*	*Combretaceae*	1	5
Samea	*Samanea saman*	*Fabaceae*	3	15
Saptparni/ Chatim	*Alstonia scholaris*	*Apocynaceae*	7	35
Semal	*Bombax ceiba*	*Malvaceae*	4	20
Sissoo	*Dalbergia sissoo*	*Fabaceae*	10	50
Sitaphal	*Annona reticulata*	*Annonaceae*	5	25
Subabool	*Leucaena leucocephala*	*Fabaceae*	11	55
Teak	*Tectona grandis*	*Lamiaceae*	3	15
Weeping Fig	*Ficus benjamina*	*Moraceae*	2	10
White Siris	*Albizia procera*	*Fabaceae*	6	30
Yellow Kaner	*Cascabela thevetia*	*Apocynaceae*	2	10

(25). All three-study site comes under the public park and hence plantations with aesthetic and fruiting species were seen there which contributed to increased species diversity in the area. Similarly, in all the three ponds of Guru Ghasidas Vishwavidyalaya *viz.*, GGV Pond 1, GGV Pond 2 and GGV Pond 3 being an institutional campus, a total of 23, 21, and 19 species were observed respectively. Whereas, Chhathghat Pond (5) and Chingrajpara Pond (8) had the least species diversity in comparison to other urban wetlands with 7.70% and 12.31% species constitution respectively. Thus, due to less biotic pressure, the GGV ponds are good in quality and support biodiversity which leads to the highest ecosystem services scorer among all wetlands of the study area. The Guru Ghasidas Vishwavidyalaya campus has a high species richness, as evidenced by the number of species there (Anand et al 2021). As a result, it can say that the small water structure can be preserved and kept alive with the least amount of human interference and effort. As we all know, nature is self-sufficient in improving itself, so it is crucial to preserve it in order to support biodiversity, human health, and to get benefitted from the area's ecosystem services.

CONCLUSION

The current investigation of the pond ecology reveals various functions that have never been examined in the region. Outcomes are helpful to gain an understanding of the various advantages of a small inland ecosystem that is subject to heavy residential pressure, both directly and indirectly, in terms of concrete and intangible benefits. Results have indicated about the impacts of disturbance in a pond on various services connected to that ecosystem and human life. For instance, dumping rubbish into a pond reduces its cultural value and causes pests and diseases to proliferate, which has an impact on regulatory services. The removal of wastes can also cause an algal bloom, have an impact on the cycling of nutrients, reduce biological oxygen demand (BOD) and affect supporting services. Consequently, interlinked services have a significant impact on our environment. Thus, the assessment helps to comprehend the condition of the ponds and their advantages on a local, regional, and worldwide scale.

AUTHORS CONTRIBUTION

A.M., D.S., P.S., G.B., and R. contributed to the design of the research and also carried out the implementation. A.M., D.S., and P.S. analyzed the data. D.S., G.B. and R. performed the calculations. A.M. and P.S. wrote the manuscript with input from all authors. G.P. conceived the study and was in charge of overall direction and planning. All authors provided critical feedback and helped shape the research, analysis and manuscript.

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Distribution and Phenology of *Syzygium stocksii* (Duthie) Gamble an Endangered Tree Species in South Konkan

A.P. Pawar, V.K. Patil, M.D. Patil, A.D. Rane, V.M. Mhaiske and V.D. Tripathi

College of Forestry, Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth Dapoli-415 712, India
E-mail: anaghapawar521@gmail.com

Abstract: *Syzygium stocksii* (Duthie) Gamble is an endangered, evergreen tree species, endemic to southwestern states of India. An assessment was conducted on distribution and phenology of *S. stocksii* in South Konkan viz. Ratnagiri and Sindhudurg districts. A snowball sampling was used in the study to identify the primary database on location of these species. Tree density and population structure were accessed by laying a quadrat of 10 x10 m around the individual tree and minimum 3 similar quadrats were laid around mature trees for the population. At each site, 3 adult individuals of these species were marked permanently for recording phenological observations. General BBCH (Biologische Bundesanstalt, Bundessortenamt und CHemische Industrie) was used to record phenophase of the selected trees. Total 78 individual trees were recorded in which maximum number of mature stems was recorded from the study area. Present study revealed that good sized populations of *S. stocksii* were is present in the study area. In total, seven principal stages - *i.e.*, bud development, leaf development, inflorescence emergence, flowering, fruit development, fruit maturity, senescence and beginning of dormancy are described with respect to their seasonal progression and relative dominant.

Keywords: Endangered, Endemic, Distribution, Phenology

Syzygium is the largest woody genus of flowering plants, with 1200-1800 species found throughout the Old World tropics and subtropics (Ahmad et al 2016). The genus is found naturally in subtropical and tropical regions of Africa and Madagascar, Asia, and Oceania and the Pacific region. The majority of *Syzygium* species range from medium to large evergreen trees. The forests of the Western Ghats, particularly tropical wet evergreen, and high-altitude Shola peaks, are ideal habitats for the *Syzygium* (Sreekumar et al 2020). It was reported that many species in this area have not yet received the proper taxonomic classification, and it is likely that new species will be classified in the future (Ahmed et al 2016). *S. stocksii* is a tall, evergreen tree which belongs to the family Myrtaceae. It is an eendangered tree species (WCMC 1998), endemic to southwestern states of India. Page (2017) reported the species as rare along forest streams and swamps within 40-80 m elevation range and put it into Data Deficient category. It was published by Duthie as *Eugenia stocksii* based on plant collection of J. E. stocks from Konkan region of Maharashtra in 1879. Despite being first described from Konkan region, this species is a less-known rarity in Maharashtra. This species is found only in few localities of Konkan region of Maharashtra, Uttara Kannada in Karnataka, Wayanad district of Kerala and Tamil Nadu (Hooker 1880, Cooke 1905). It is classified as endangered in IUCN red list category (IUCN 1998). There have been no attempts to document phenological growth stages in this species so far. The study was therefore conducted to study the distribution and phenology of *S. stocksii* in Ratnagiri and Sindhudurg districts in south Konkan region of Maharashtra. This study also attempts to provide photographic phenophases charts for the *S. stocksii* for the first time.

MATERIAL AND METHODS

Study site: The present investigation was conducted in the southern Konkan region, comprising Ratnagiri and Sindhudurg districts, located between 15°36' N to 18°50' N latitude, and 74°36' E to 75°50' E longitudes. The areas of Ratnagiri and Sindhudurg districts are 8208 and 5207 km^2, respectively. The South Konkan is bounded in the west by the Arabian Sea, in the east, it shares boundary of the Sahyadri escarpment, in the north by the river Savitri and in the south by the Union Territory of Goa and the Terekhol estuary. South Konkan experiences tropical warm, humid or maritime climate throughout the year. Atmospheric temperature of South Konkan varies from 25 to 35°C with the average humidity ranging between 60 and 90 %. As the districts (Sindhudurg and Ratnagiri) of South Konkan belong to coastal region, the variation in temperature during the day and through the seasons is not large (Dhawal et al 2013).

Collection of data and field survey: Rare tree species are distributed sparsely and the knowledge of existence of individual trees or populations of such species is often with the researchers, forest department officials and some local people. There are no databases of locations of this species. Hence, snowball sampling or chain referral sampling was used in the study (David 2008). The initial small pool of informants included some known researchers and forest department personnel. They were contacted over phone and email and a simple questionnaire following Tantiado (2012) was provided to them to get the locations of the study species. They were also requested to nominate further sources of information for finding more populations/individuals of the study species. Once, the information on a few sites was obtained, the sites were visited personally and the claim of existence of the species were verified. Trees were identified in the field as much as possible, with the help of local people or departmental persons. Interactions were also conducted with local people to identify any more nearby trees/populations. For the unidentified plants, photos and sample parts were collected and identification was done with the help of experts or books like Flowers of Sahyadri (Ingalhalikar 2001) and Further Flowers of Sahyadri (Ingalhalikar 2007), Endemic Woody plants of the Western Ghats (Page 2017), Flora of Maharashtra (Singh et al 2001).

Tree density and ppopulation sstructure: At some sites, there were single individuals while at others, there were populations. Wherever single individuals occurred, a quadrat of 10 m x10 m was laid around the individual tree. For all mature trees taken at the centre of the quadrat, location parameters (village, lat-long, elevation, slope, aspect), physical parameters (height, GBH), social parameters (ownership) and population/stand parameters (total number of individuals in a population) were recorded. Ravi altimeter was used for measuring top height of tree and ordinary measuring tape was used for measuring girth and crown spread. Photographs of trees and its phenophases were also taken. Girth at Breast Height (GBH) was taken for all the stems of *S. stocksii*. Based on the girth class, populations were grouped into three classes. Stems <30 cm gbh were classified as saplings. Stems between 30-60 cm gbh were treated as sub adults. Stems >60 cm gbh were classified as mature trees (Irwin et al. 2013).

Mapping: Precise locations of *S. stocksii* in South Konkan were mapped using Garmin eTrex 10 Global Positioning System (GPS) handheld receiver. The GPS data were plotted using QGIS software and a distribution map was prepared.

Phenology of *S. stocksii:* Three adult individuals of *S. stocksii* were marked permanently at each site for recording phenological observations. On each marked individual, 20 branches (five in each direction) were selected and tagged. General BBCH scale for phenology of tree and woody species was used to record phenophase of the selected trees (Finn et al 2017). The General BBCH scale modified for woody species uses eight of the ten principle stages, beginning with sprouting/bud development (stage 0) and ending with dormancy (stage 9). Stage 2 and Stage 4 were excluded as they were irrelevant to woody species. Secondary stages were numbered 0 to 9 and correspond to ordinal or percentile growth stages. Appropriate corresponding photographs were taken to develop a pictorial guide to phenophases of the selected species. On every visit, the predominant phenophase was recorded for each of the tagged branch. The observation interval was approximately 30 days. Secondary information and photographs of phenological events was collected from the informants. The duration of phenological events such as leaf fall, completion of leaf fall, leaf initiation, beginning of flowers, completion of flowering, beginning of fruiting and completion of fruiting of the selected species were recorded.

RESULTS AND DISCUSSION

Locations were sought from experts in snowball sampling method. Initially two experts were contacted and later the number increased to fifteen. From them, total 5 locations for *S. stocksii* were received *viz*. Aavere, Barsu, Dhamapur, Medhe, and Parule in the study area. It is seen growing between 7-84 m altitude, exclusively in 50-100 m vicinity of streams. The tree grows to a height of about 22 m, attains maximum girth of 425 cm and maximum crown spread of 19.5 m. The trees were observed as the fastest growing tree as the 6 months old tree in study site shows the 22 cm girth and approximately 3 m height. It was also observed as a good coppicer tree species. Total 78 trees were enumerated from five different locations in study areas. Among all 5 locations, highest number of individuals was found at the study site Parule followed by Barsu, Dhamapur and least was found at Medhe. Among all identified sites an individual tree was recorded at the study site Aavere. Average girth of trees at different locations revealed that in Kalse, Dhamapur comparatively higher GBH were recorded followed Rajapur and least was recorded in Medhe and Parule (Table 1). On the other hand, average height of trees at different locations revealed that study area in Barsu recorded highest average height followed by Dhamapur while least was recorded Parule followed by Medhe. Both highest average GBH and height values were recorded for a single tree in Aavere (Table 1). This species is found growing in association with *Cocos nuciifera, Caryota urens, Areca catechu, Mangifera indica, Callophyllum inophyllum, Macaranga peltata, Hydnocarpus pentandrus, Artocarpus heterophyllus, Holigarna arnottiana, Vitex negundo, Tabernaemontana alternifolia, Tectona* *grandis, Mammea suriga, Garcinia indica* and *Strychnos-nux-vomica*. The species is threatened in the study area due to harvesting of trees for the timber, clearance for the agricultural lands and farms. It was reported earlier as rare in the evergreen forest of Konkan (Cooke 1905), whereas Page (2017) also reported it as rare along forest streams and swamps within 40-80 m elevation range. Recently Sathish et al (2020) reported it as critically endangered in two locations of the *Kaanu* forests in Karnataka having perennial stream. But observations showed that large populations of this species are scattered along the streams over Sindhudurg and Ratnagiri districts.

Among all five locations, five adults were recorded from the study area in Medhe while only a single stem was recorded from Aavere. Of the five locations, population sites which were highly disturbed due to anthropogenic interference show very poor number of saplings and sub-adults (Fig. 2). Saplings were completely absent at the study area in Barsu and Dhamapur. The absence of saplings, despite good adult population, indicates poor germination of seeds and establishment of seedlings. The reasons for this phenomenon need to be explored through more intensive field studies. In Parule site, good number of sapling (10), sub-adult (13) and mature stems (15) were recorded. Occurrence of more saplings in this site proves that if an area is left undisturbed, regeneration of species is promoted naturally, especially of endemic species. On the other hand, occurrence of more saplings may be due to the high rate of seed germination. Similar observations have been made for *Elaeocarpus venustus* in Agastyamalai Biosphere Reserve, Kanyakumari District (Irwin et al 2013).

Phenology of *S. stocksii*: Phenological data for *S. stocksii* was collected from January to June 2022 in all the 3 sites viz. Barsu, Dhamapur and Parule. Throughout the season, trees at all the three sites showed similar progression of phenophases. Overall, the trees remained in the vegetative flush phenophases till Meteorological Week (MW) 9. Inflorescence emergence phenophases started during MW 10 and peaked during MW 14. Flowering phenophases prevailed dominantly during MW 18 along with simultaneous fruit development. Fruit maturity was maximum during MW 23. Despite the large distance between Barsu, Dhamapur, and Parule sites, no obvious delay in inflorescence emergence was observed. In *Syzygium caryophyllatum*, Nadarajan and Pujari (2018) found that flowering occurred during the summer season (second week of March to May) with massive flowering in the month of April (peak photoperiod). Synchronous flowering was observed in this species. In the present study, similar observations regarding flower emergence and peak flowering were recorded. Further, there was synchronous flowering even in very distant populations. A pictorial depiction of phenophases in

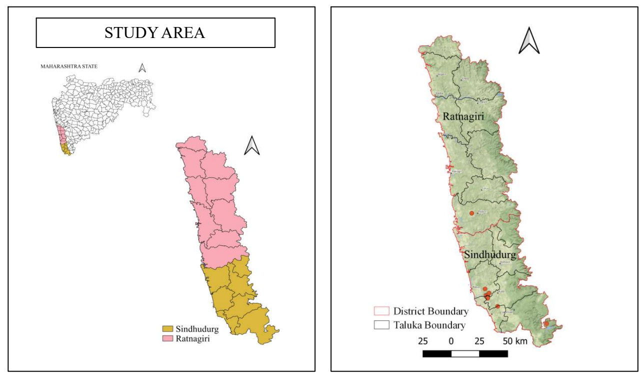

Fig. 1. Population sites of *S. stocksii* in Ratnagiri and Sindhudurg districts

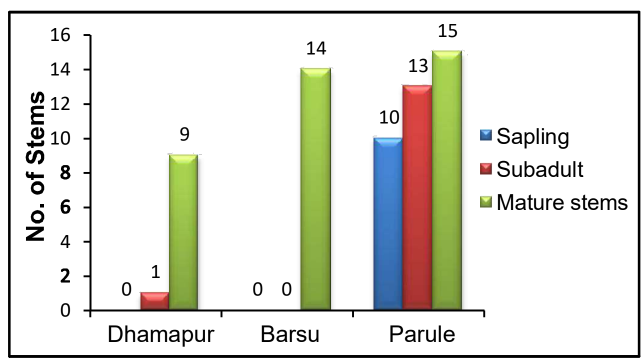

Fig. 2. Population structure of *S. stocksii* in Ratnagiri and Sindhudurg districts

Table 1. Current population status of *S. stocksii* in Ratnagiri and Sindhudurg districts

Village	Taluka, District	No. of trees	Av. GBH (cm)	Av. Height (m)
Aavere	Vengurla, Sindhudurg	1	270.00	19.0
Barsu	Rajapur, Ratnagiri	14	213.02	9.9
Dhamapur	Malvan, Sindhudurg	10	86.00	7.5
Medhe	Dodamarg, Sindhudurg	5	89.91	8.1
Parule	Vengurla, Sindhudurg	48	199.23	12.3
Total		78	143.57	9.6

Fig. 3. a– Habit and Habitat of tree, b– Quadrangular branches of *S. stocksii*, c- 6 months old *S. stocksii* growing in study area, d– Coppicing shoots of *S. stocksii*

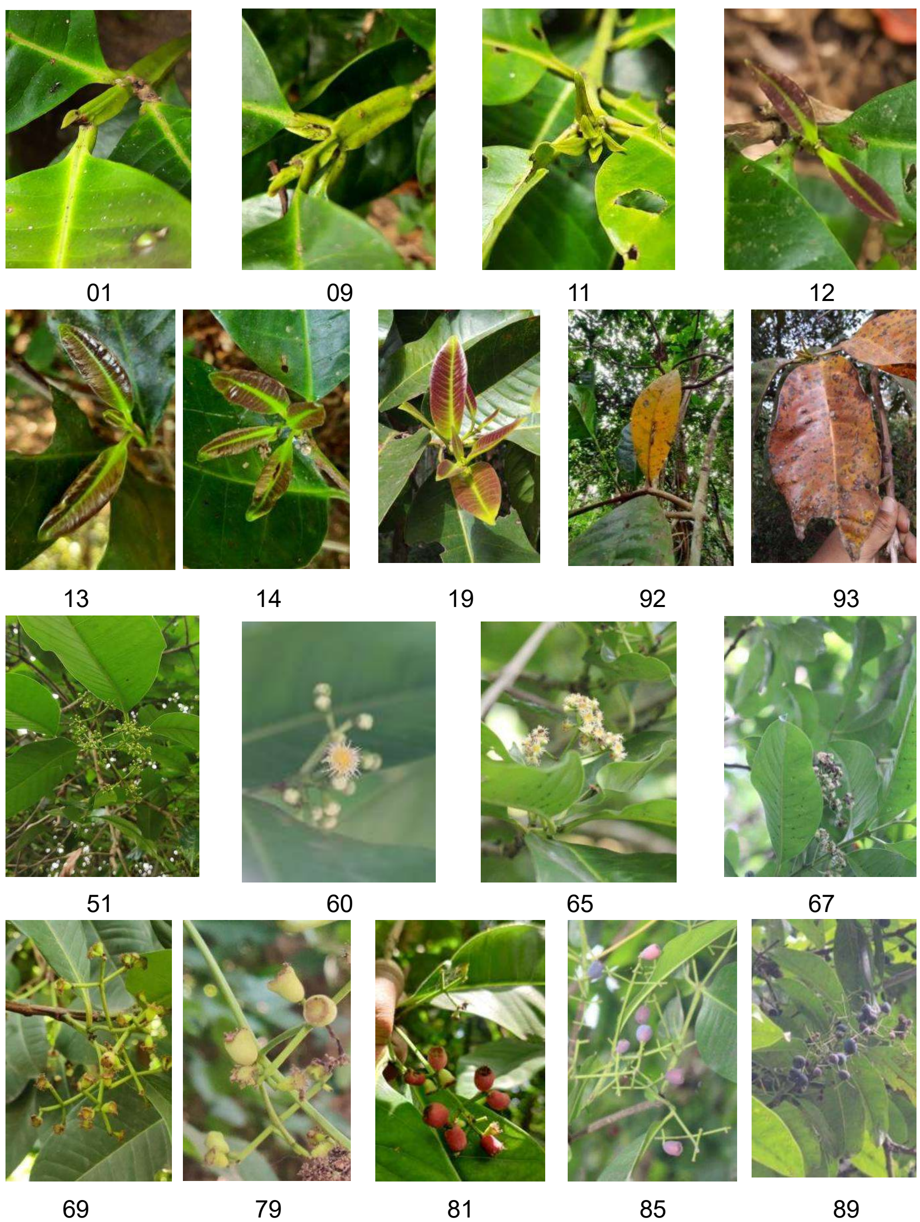

Fig. 4. Phenological growth stages of *S. stocksii* according to the BBCH scale

Table 2. BBCH scale developed for phenological growth stages of *S. stocksii*

Code	Stage/ sub-stages and description/ timing of occurrence
Principal growth stage 0: Bud development	
00	Dormancy: Buds closed and covered by green scale
01	Beginning of bud swelling
03	End of bud swelling
07	Beginning of sprouting or bud breaking
09	Bud shows green tips
10	First leaves separated
Principal growth stage 1: Leaf development	
11	First leaves unfolded
12	2 true leaves unfolded
13	3 true leaves unfolded
14	4 true leaves unfolded
15	5 true leaves unfolded
16	6 true leaves unfolded
17	7 true leaves unfolded
18	8 true leaves unfolded
19	more true leaves unfolded and expanded
Principal growth stage 5: Inflorescence emergence	
51	Inflorescence or green flower buds visible
55	First individual flowers visible (still closed)
59	First flower petals visible (in petalled forms)
Principal growth stage 6: Flowering	
60	First flowers open (sporadically), A tiny white color flower open
61	Beginning of flowering: 10% of flowers open
62	20% of flowers open
63	30% of flowers open
64	40% of flowers open
65	Full flowering: 50% of flowers open
67	Flowering finishing: majority of petals fallen or dry
69	End of flowering: fruit set visible
Principal growth stage 7: Development of fruit	
72	20% of fruits have reached final size
75	50% of fruits have reached final size
78	80% of fruits have reached final size
79	Fruit final size. The small light green fruits are seen
Principal growth stage 8: Maturity of fruits and seeds	
81	Beginning of ripening or fruit coloration, pericarp turns into pinkish red
85	Advanced ripening or fruit coloration, pericarp turns into pinkish purple
87	Fruit begins to soften
89	Fruit fully ripe, pericarp turns into dark purple
Principal growth stage 9: Leaf senescence and beginning of dormancy	
91	Shoot growth completed; foliage still green and terminal bud developed
92	Beginning of leaf discoloration, majority of leaves turn yellow
93	Beginning of leaf-fall, majority of leaves turn brown
95	50% of leaves fallen
	End of leaf fall

S. stocksii is presented in Fig. 4 and description of these phenophases is provided in Table 2.

CONCLUSION

S. stocksii is confined to riparian habitat which is a specialized ecological niche and is assessed as Endangered in the IUCN Red List of Threatened Species (WCMC 1998). The species were having good natural populations whereas some were sparsely distributed as individuals. This study provides a unified standard for describing the phenology of *S. stocksii*. In contrast to other procedures, application of the extended BBCH scale allows for more accurate and scientific description of the morphological characteristics of *S. stocksii* in different developmental stages. Detailed stand structure and regeneration studies need to be conducted for this species in future.

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Influence of Biofertilizers on Early Stage Seedling Growth, Biomass and Vigour of *Anthocephalus cadamba* (Roxb.) Miq.

Punamkumari Chauhan, L.K. Behera, M.B. Tandel, N.S. Thakur, R.S. Chauhan and C.A. Dholariya

College of Forestry, Navsari Agricultural University, Navsari-396 450, India
E-mail: punamcchauhan@gmail.com

Abstract: The present investigation was conducted to know the influence of biofertilizers on seedling growth and vigour in *Anthocephalus cadamba* (Roxb.) Miq., for which it was laid out in a Completely Randomized Design with three repetitions and seven biofertilizer treatments. An application of VAM @ 10 ml plant^{-1} exhibited significantly maximum collar diameter (9.62 mm), root length (33.04 cm) and found at par with Azospirillum @ 10 ml plant^{-1} at 150 DAT. However, Azospirillum @ 10 ml plant^{-1} exhibited maximum shoot length (80.52 cm), number of leaves per plant (13.33) and total leaf area (cm^2) per plant (1770.29 cm^2), and found at par with VAM @ 10 ml plant^{-1} having 76.50 cm, 13.25, 1692.92 cm^2 for same growth characters of *A. cadamba* seedlings at 150 DAT. Further, VAM @ 10 ml plant^{-1} produced significantly maximum fresh weight, dry weight and seedling quality index of 96.58 g, 19.67 g and 1.77, respectively whereas the maximum root to shoot ratio of 0.33 and the lowest improved sturdiness quotient of 7.53 were observed in seedlings treated with Pseudomonas @ 10 ml plant^{-1}, which was statistically at par with VAM @ 10 ml plant^{-1} at 150 DAT. Overall result showed that application of VAM @ 10 ml plant^{-1} improved the growth and vigour of *A. cadamba* seedlings, so this treatment can be used for raising and production of good quality seedlings in the forest nursery.

Keywords: *Anthocephalus cadamba,* Biofertilizers, Seedling quality index, Seedling vigour, VAM

Anthocephalus cadamba (Roxb.) Miq., synonymously named as *Neolamarckia cadamba* (Roxb.) Bosser and *Anthocephalus chinensis* (Lam.) A. Rich ex Walp. and popularly known as Kadamb or Kadam tree belongs to Rubiaceae family. The Kadam is a large tropical tree with broad crown and straight cylindrical bole having average height about 15 meters and in the favourable climatic condition it attains a height of 20 meters or more with a clean bole of about 9 meter and a diameter of 40 to 60 cm. The tree is particularly aesthetic and deciduous in nature however sometimes evergreen and semievergreen in nature (Bijalwan et al 2014). *A. cadamba* is native to South and Southeast Asia, including India, Indo-Malayan region, Java, Sumatra, China, Indonesia, Malaysia, Bangladesh, Sri Lanka, Cambodia, Papua, New Guinea, Philippines and Singapore. In India, it occurs in the Sub-Himalayan tract from Nepal eastward to West Bengal and Assam, Bihar, Chhattisgarh, Madhya Pradesh, Andhra Pradesh and evergreen forest of Karnataka to Kerala (Anonymous 1985). The species is considered suitable for soil conservation, agroforestry, jhum and land reclamation. The rotation period or harvesting of Kadam depends upon the production purposes, normally for pulpwood and matches, harvesting can start at 4-5 years whereas for wood production, felling of trees can start approximately from the age of 10 years (Bijalwan et al 2014). *A. cadamba* wood used in the ceiling boards, light construction work, packing cases, planking, carving and turnery. It makes good veneers and plywood suitable for the manufacture of grade IV commercial plywood and tea chest plywood. It is highly suitable for the manufacture of pencils, match boxes and splints and also suitable for making printing and wrapping paper (Chaturvedi et al 2017). There is high demand for the species in pencil industry, plywood and match splints and adopted in plantation, particularly in agroforestry with a spacing of at least 5 m × 5 m or 6 m × 6 m and farm forestry practices in humid tropics for industrial and income generation (Bijalwan et al 2014). Further, *A. cadamba* has minimum shade effect and no allelopathic effect on the agricultural crop advocates its suitability for the agroforestry purpose.

Forest nurseries play a vital role in the development of plantations of forest trees. There is large demand for quality forest planting materials, so it has become imperative that forest nurseries should be managed professionally to produce the desirable quality of seedlings (Shreedher and Mohan 2016). The improved nursery seedlings stock can be obtained by application of biofertilizers. Biofertilizers are group of microorganisms consisting of bacteria, fungi, algae *etc.* These alone or in combinations are known to be increasing plant growth by way of various biochemical activities in the soil such as Nitrogen fixation, breakdown of organic matter, secretion of plant growth hormones and increase of available mineral nutrients in soil. They are also helpful to build up other beneficial microorganisms and in turn improve soil health. The biofertilizers also control soil or root borne diseases, parasitic nematodes and maintain soil structure. An application of these microorganisms to seed, soil or composting areas with the objective of increasing the numbers of such microorganisms and accelerating certain microbial processes to augment the extent of the availability of nutrients in a form which can be easily assimilated by plants (Sharma and Chaubey 2015). Looking into the economical as well as environmental importance of Kadam and to produce quality planting materials in nursery, the present trial was carried out to study the effect of biofertilizers on seedling growth, biomass and vigour in kadam, *A. cadamba* (Roxb.) Miq. in early stage.

MATERIAL AND METHODS

The present investigation was conducted at Net House, College of Forestry, Navsari Agricultural University (NAU), Navsari, (Gujarat) from December 2019 to August 2020. For the experiment, orange coloured ripened fruits of Kadam were collected from healthy, well developed trees distributed in the campus of NAU, Navsari. The collected fruits were allowed to rot for three to four days and pulp was then washed off by hand in a small bucket or pot of water, after tiny seeds settled at the bottom were taken out and dried well (Vijayaraghavan 2014). These seeds were sown in germination bed containing soil and sand for raising seedlings. Germinated seedlings growth was very slow for which they kept in the same condition for two months and then transplanted into the 10" × 8" size perforated black polythene bags containing growing media of soil, sand and FYM (2:1:1) having a seedling height of about 5-6 cm. After one month of establishment, different biofertilizers such as $T_{1:}$ Control (No biofertilizer), $T_{2:}$ Azotobacter, $T_{3:}$ Azosprillum, $T_{4:}$ Acetobacter, $T_{5:}$ PSB (Phosphate Solubilising Bacteria), $T_{6:}$ Pseudomonas and $T_{7:}$ VAM (Vesicular Arbuscular Mycorrhiza) at the rate of 10 ml per plant were directly applied to soil in the polythene bag. Various growth parameters such as shoot length, collar diameter, number of leaves per plant, total leaf area per plant, root length, total fresh and dry weight of plant were recorded at 150 DAT (Days After imposing Treatment). Further, seedling vigour indices like root: shoot ratio, sturdiness quotient (SQ) and Seedling Quality Index (SQI) were calculated. The data obtained from the experiment were processed and fed to the data sheet in MS Excel and subjected to statistical analysis using DOS based software developed by Department of Agricultural Statistics, NAU, Navsari in experimental design CRD (Completely Randomized Design). The ANOVA was constructed for further inference. The appropriate standard error of mean [SEm (±)] was calculated in each case and critical difference (CD) at 5 per cent level of probability was worked out to compare the treatment means, where the treatment effects were significant (Panse and Sukatme 1985).

RESULTS AND DISCUSSION

The findings of present study showed that effect of biofertilizers such as VAM and Azospirillum on growth and development of Kadam seedlings was found considerably maximum as compared to other treatments with respect to shoot length, collar diameter, number of leaves plant^{-1}, total leaf area plant^{-1}, root length, fresh and dry biomass (Table 1). Among different biofertilizers, Azospirillum @ 10 ml plant^{-1} (T_3) showed maximum shoot length (80.52 cm), number of leaves per plant (13.33), total leaf area per plant (1770.29 cm^2) in *A. cadamba* seedlings and found at par with VAM @ 10 ml plant^{-1} (T_7) at 150 DAT. However, application of VAM @ 10 ml/plant (T_7) exhibited the maximum collar diameter (9.62 mm) followed by Azospirillum (8.90 mm) @ 10 ml plant^{-1} (T_3) at 150 DAT. Further, maximum root length (33.04 cm) was recorded in VAM @ 10 ml plant^{-1} (T_7) and found at par with Azospirillum (32.20 cm) @ 10 ml plant^{-1} (T_3). Similarly, VAM @ 10 ml plant^{-1} (T_7) exhibited significantly maximum fresh and dry weight of plant (96.58 g) and (19.67 g) followed by Azospirillum @ 10 ml plant^{-1} (T_3) (89.18 g) and (18.25 g) respectively in *A. cadamba* at 150 DAT.

The increase in shoot length and collar diameter can be credited to VAM and Azospirillum application in the media as it helps in increasing nutrient and water uptake capacity which are necessary for the better growth and also helps in maintenance of good physical and chemical properties of the media (Chiranjeevi et al 2018). The growth of seedlings in early stages in nursery normally depends upon the growing media: its type, nature along with the types of inoculants or biofertilizers applied. The growth of seedlings in early stages also depends upon the tree species type and growth characteristics. Normally, certain biofertilizers enhance the growth of seedlings due to their symbiotic and positive interaction with the seedlings (Duponnoisa et al 2005, Wu et al 2010). The rhizosphere is a dynamic soil environment formed by living plant roots and their associated microorganisms and fauna. Among different microbes, plant growth promoting microbes like AM fungi, Azospirillum, Azotobacter, phosphobacteria in rhizosphere are able to exert a beneficial effect upon plant growth. These microbes have multiple functions like nitrogen fixation, phosphorus solubilization and mobilization (Sen and Paul 1957) and stimulating root development by producing metabolites like IAA and other growth hormones (Lynch 1990). Normally, the effect of plant growth promoting microbes on the growth performance varied with the treatments and the host plant species (Shreedher and Mohan 2016). In the present study, all the growth parameters were found maximum in VAM and Azospirillum treatments. It may be due to the stimulatory effect exerted by these biofertilizers. AM have great importance as it efficiently enhances the nutrient uptake in infertile soils, water uptake and drought resistance in plants as well as improves the disease resistance (Nowak 2004). Similarly, Azospirillum has been attributed to several mechanisms including secretion of phytohormones (auxins and gibberellins), biological nitrogen fixation and enhancement of mineral uptake of plants (Mrkovack and Milic 2001).

Present findings of growth parameters of seedlings in Kadam inoculated with VAM and Azospirillum are in accordance with the reports of Budi and Christiana (2012), Shreedher and Mohan (2016). Budi and Christiana (2012) recorded maximum plant height, collar diameter, shoot and root dry weight when treated with Arbuscular Mycorrhizal Fungi (AMF) in *A. cadamba* seedlings whereas Shreedher and Mohan (2016) found in seedlings of *Neolamarckia cadamba* treated with VAM and Azospirillum in combination exhibited maximum shoot length, collar diameter, leaf area ratio. Similarly, inoculation of VAM and Azospirillum enhanced the seedling growth in other tree species. As reported by Vairamani and Rajendran (2021) in seedlings of *Casuarina junghuhniana* inoculated with the combined application of *Azospirillum* + *Paenibacillus polymyxa* + AM fungus produced the maximum growth and biomass; Mohan and Rajendran (2014) in *Feronia elephantum* treated with combined inoculation of Azospirillum + AM fungi + Pseudomonas showed shoot length, root length, collar diameter and biomass increased above 77.47 per cent than control; Parveen and Kumar (2020) used *Azospirillum brasilense* + *Bacillus polyxyma* + *VAM* in production of quality seedlings of *Acacia nilotica*. Further, Mohan and Rajendran (2017) observed increased shoot length, root length, collar diameter and biomass in combined inoculation of Azospirillum + AM fungi + Pseudomonas in *Aegle marmelos* whereas in *Casuarina equisetifolia* after six months of inoculation of biofertilizers, significantly maximum root length, shoot length, collar diameter, root weight and shoot weight were recorded in combination of Frankia + Azospirillum + Phosphobacterium (Saravanan et al 2012). Moreover, AM treated seedlings increased in plant biomass and height to the extent of 34 and 24 per cent respectively in *Dalbergia sissoo*, 126 and 50 per cent in *Acacia auriculiformis*, 48 and 24 per cent in *D. latifolia* and 100 and 112 per cent in *A. nilotica* (Uniyal and Thapar 1995). Similarly, Chiranjeevi et al (2018) also found the maximum seedling height (24.13 cm), root length (16.33 cm), seedling girth (0.63 cm), number of leaves per plant (18.86), fresh weight (26.9 g) and dry weight (3.07 g)in aonla when treated with VAM as an inoculant.

The present result of higher biomass production with application of VAM and the same findings was supported by Shreedhar and Mohan (2016) in Kadam where seedlings inoculated with AM fungi and PGPR exhibited higher biomass production. VAM normally enhances the surface area of roots which helps in absorption of available nutrient and water to larger extent as compared to other biofertilizer. So it may ultimately enhance the growth and biomass production of seedlings (Saini 2019). Furthermore, the increase in seedling biomass production may be strongly correlated with improved accumulation of N due to Azospirillum and Azotobacter and P due to AM fungi and PSB inoculation (Ratha Krishnan et al. 2004). Moreover, Kandasamy et al (1987) recorded, in *Ailanthus excelsa* and two other species inoculation with AM fungus, increased the

Table 1. Influence of biofertilizers on the growth parameters of *A. cadamba* seedlings at 150 DAT

Treatments	Shoot length (cm)	Collar diameter (mm)	Number of leaves/plant	Total leaf area /plant (cm2)	Root length (cm)	Fresh weight /plant (g)	Dry weight /plant (g)
Control	65.40	7.47	12.17	1372.25	26.47	72.75	13.99
Azotobacter	68.22	8.43	11.42	1384.68	28.02	77.96	16.99
Azospirillum	80.52	8.90	13.33	1770.29	32.02	89.18	18.25
Acetobacter	74.73	8.45	12.25	1502.76	29.00	81.58	17.89
PSB	56.12	6.86	12.08	1469.79	27.66	49.61	9.54
Pseudomonas	57.31	7.61	10.42	1326.90	26.42	48.60	8.42
VAM	76.50	9.62	13.25	1692.92	33.04	96.58	19.67
Mean	68.40	8.19	12.13	1502.80	28.95	73.75	14.96
SEm(±)	1.78	0.13	0.27	43.85	0.68	1.77	0.42
CD (0.05)	5.38	0.40	0.80	133.02	2.06	5.35	1.26

shoot and root dry weights of 62.08 per cent and 43.18 per cent, respectively. Further, Balasubramanian and Srinivasan (1995) found that seedlings inoculation with four AM fungi in *A. excelsa*, *Tectona grandis* and *Dalbergia sissoo* significantly increased the total biomass; Madan et al (1995) reported that seedlings inoculated with AM fungal showed better shoot and root dry weight in *A. excelsa*, *Pongamia glabra* and *Cassia siamea*; Rahangdale and Gupta (1998) also recorded that *Gmelina arborea* and other five tree species exhibited significant increase in root and shoot biomass when treated with AM fungi.

Results also revealed that seedlings treated with PSB @ 10 ml plant^{-1} and Pseudomonas @ 10 ml plant^{-1} recorded lower value than control in most of the growth parameters. This may be due to the nature and functions of both biofertilizers and their effect on the growth characters of treated seedlings. Further, both these biofertilizers avail the P to host seedlings in most of conditions but might compete with the host plants for N as food supplements in the present situation as a result of which growth parameters were inhibited and found lower. Present findings were supported by Saini (2019) in *Swetenia macrophylla* seedlings treated with PSB @ 10ml plant^{-1} and Pseudomonas @ 10 ml plant^{-1} resulted in lower value of growth parameters as compared to control at 90 DAT.

The vigour parameters such as root: shoot ratio, sturdiness quotient and seedling quality index showed significant variation among seven treatments in the seedlings of *A. cadamba* at 150 DAT (Table 2). Among various treatments, Pseudomonas @ 10 ml plant^{-1} (T6) produce maximum root: shoot ratio (0.33) and found at par with T_7, T_2, T_4 and T_5 in the seedlings of *A. cadamba.* Further, the lowest SQ value was recorded in Pseudomonas @ 10 ml plant^{-1} (7.53) which was statistically at par with VAM @ 10 ml plant^{-1} (7.95). Whereas, VAM @ 10 ml plant^{-1} (T_7) was recorded significantly maximum SQI (1.77) followed by T_2, T_4, T_3 at 150 DAT.

Table 2. Influence of biofertilizers on vigour parameters of *A. cadamba* seedlings at 150 DAT

Treatments	Root:shoot ratio	Sturdiness quotient	Seedling quality index
Control	0.26	8.75	1.11
Azotobacter	0.31	8.09	1.50
Azospirillum	0.29	9.04	1.46
Acetobacter	0.30	8.84	1.47
PSB	0.30	8.17	0.82
Pseudomonas	0.33	7.53	0.80
VAM	0.31	7.95	1.77
Mean	0.30	8.34	1.28
SEm(±)	0.01	0.15	0.03
CD (0.05)	0.03	0.45	0.09

The root: shoot ratio is an important measure for seedling survival. It relates the water absorbing area of roots to the transpiring area of shoot. A good ratio indicates a healthy plant (Jaenicke 1999). It may be due the fact that nitrogen deficiency in soil media may result in an increased root: shoot ratio (Harris 1992). Similar finding was recorded in *Swietenia macrophylla* seedlings treated with Pseudomonas at 90 DAT (Saini 2019). Moreover, kadam seedlings recorded higher value of root: shoot ratio when treated with AMF and Azospirillum (Shreedher and Mohan 2016). The result was further supported by Firuzsalari et al (2012) where significant difference in root: shoot ratio was found maximum in treatment involving Nitragin (a local biofertilizer containing Azospirillum, Azotobacter and Pseudomonas as its content).

The lowest value of SQ signifies the sturdiness *i.e.* ability to withstand in stress condition of the seedlings in terms of field establishment. Present finding of minimum SQ value was in accordance with the result of *S. macrophylla* seedlings inoculated with Pseudomonas (Saini 2019). Further, Shreedhar and Mohan (2016) reported that the biofertilizer inoculated seedlings showed better sturdiness quotient value as compared to uninoculated seedlings. Moreover, Maharana et al (2018) reported the lowest SQ of 11.75 in treatment inoculated with Azospirillum + Novel in *Gmelina arborea* seedlings whereas Chandra and Ujjaini (2002) recorded lowest result in forest tree seedlings treated with AM fungi + organic manure.

Seedling Quality Index is considered a promising integrated measure of morphological traits and a good indicator of seedling quality as its computes robustness and biomass distribution of seedlings as compared to individual growth parameters like shoot length, collar diameter *etc*. (Binotto et al. 2010). In the trial, VAM @ 10 ml plant^{-1} was recorded significantly maximum SQI (1.77) at 150 DAT. Higher the value of SQI, better the ability of the seedling to survive in the field. VAM inoculated seedlings exhibited better result in terms of SQI which may be due the availability of nutrients such as N and P increased with the application of it. However, the high percentage of root colonization in AM fungi treated seedlings is found to be directly correlated with an improved growth and physiology. Further, the presence of AM fungi significantly increases root surface area by production of extensive hyphae, increase transpiration, reduce leaf temperature and restrain the decomposition of chlorophyll and improves the seedling quality (Ajeesh et al 2015). Moreover, inoculation of VAM in seedlings of woody species at nursery stage improves the seedling growth and survivability (Rashmi and Bhavana 2015). According to Shreedhar and Mohan (2016), the higher SQI value was reported in *N. cadamba* seedlings when inoculated with AM fungi and PGPRs. The results for *A. cadamba* seedlings inoculated with VAM produced maximum SQI and this finding was further supported by Sumana and Bagyaraj (2002) in neem treated with VAM (maximum SQI of 0.85), Raj et al. (2010) in teak seedlings inoculated with VAM (maximum SQI of 0.44) and Maharana et al. (2018) in seedlings of *Gmelina arborea* treated with AM + PSB + Novel (maximum SQI of 1.82). Moreover, current findings are in line with the findings of earlier researchers on native tree species like, *Tectona grandis* (Ayswarya 2008, Rajeshkumar et al 2009), *Melia azedirach* (Rajeshkumar et al 2009) as well as exotic like *Eucalyptus* hybrid (Sastry et al 2000), *Simarouba glauca* (Ratha Krishnan et al 2004) and *Acacia auriculiformis* and *A. mangium* (Tamilselvi 2005).

CONCLUSION

VAM inoculated seedlings of *Anthocephalus cadamba* resulted in maximum SQI value, collar diameter, fresh as well as dry biomass with higher shoot length, number of leaves and total leaf area with improved SQ value than other biofertilizer treatments and control. Thus, VAM inoculation is found best for the growth and vigour of seedlings of *A. cadamba* in early stage and can be used for production of quality seedlings of *Anthocephalus cadamba* in forest nursery.

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Effect of Climate Change on Seed Germination and Seedling Attributes of *Calophyllum inophyllum* L.

Supriya K. Salimath, Ramakrishna Hegde* and Clara Manasa P.A.

College of Forestry, Ponnampet, Kodagu-571 216, India
Keladi Shivappa Nayaka University of Agricultural and Horticultural Sciences, Shivamogga-577 201, India
**E-mail: ramakrishnahegde@uahs.edu.in*

Abstract: The acclamatory responses of plants to the changing climate are a subject of debate over the past two decades. Hence, it is prudent to understand the response of tree species in the initial stages, as seed and seedlings, to the elevated CO_2 conditions. The study on seed germination, seedling growth and stomatal density analysis of *Calophyllum inophyllum* L. revealed that, the elevated CO_2 negatively affected the germination of the species. But, after the germination, elevated CO_2 condition positively influenced the seedling height growth. Stomatal density of the seedlings was negatively affected by the elevated CO_2 condition.

Keywords: Climate change, Elevated CO_2, Seed germination, Seedling growth, Stomata

Global climate change induced by the anthropogenic activities is the major challenge faced by the world in this 21st century. Humans are having long-term cumulative impacts on earth's ecosystems through a range of consumptive, exploitive and indirect mechanisms, even to the extent of influencing the global climate (Yumnam and Ronald 2022). According to the IPCC (2015) the total contribution of the agriculture, forestry and other land use systems towards the greenhouse gases emissions was 24 per cent. Moreover, the city transport sector, industrialization and reduction in green space are leading more to climate change (Pradhan et al 2022). Under this situation observing and predicting the forest tree seed germination and seedling growth under elevated CO_2 conditions is an urgent need. Although seed germination is under genetic control (Gutterman 2000, Nordan et al 2009) and parental environment (e.g. availability of light, temperature, water, and nutrients) can significantly influence germination characteristics (Marty and Bassirirad 2014, Maharana Rashmiprava et al 2018, Chauhan et al 2019). Although the evidences in the literature show species specific response of seed germination to elevated CO_2, the data on the major species is lacking. Further, increasing atmospheric carbon dioxide concentration has profound effects on growth and development of tree seedlings. A doubling of CO_2 generally stimulates photosynthesis and can lead to substantial increase in growth. The response of seedlings to elevated CO_2 greatly depend on the nutrient reserves of the soil too. Short-term experiments in tree species exposed to elevated CO_2 levels have shown that increased photosynthetic rate up to 40-80 per cent in *Pinus ponderosa, P. radiata, Quercus coccinea* and *Populus deltoides* seedlings (increased net production by 20 per cent (Couteaux and Bottner 1996). Since the studies on response of seed germination and seedling growth to elevated CO_2 of commercially important seedlings are scanty, it is prudent to understand the response of tree species in the initial stages, as seed and seedlings, to the elevated carbon dioxide conditions from the point of climate change and global warming in the future. Further, the acclamatory responses of seedlings to elevated CO_2 conditions changes stomatal density. The variations in stomatal density are attribute to the total water use efficiency of plants and are major considerations in species recommendations in the future. *Calophyllum inophyllum* L. of family Guttifereae (Clusiaceae) is a tree species native to India, East Africa, South East Asia, Australia and South Pacific and is commonly called as 'Indian laurel'. It is a broad leaved evergreen tree occurring as a littoral species along the beach crests, although sometimes occurring inland and adjacent lowland forest and widely planted throughout the tropics and is naturalized in the main Hawaiian Islands. Oil from the nuts has been traditionally used for medicine and cosmetics and is today being commercially used for the production of biodiesel. Annual yield of 20-100 kg/tree of whole fruits have been reported. The nut kernel contains 50-70 per cent oil and the mature tree may produce 1-10 kg of oil per year depending upon the productivity of the tree and the efficiency of extraction process (Chavan et al 2013). In the present study, an attempt was made to understand the response of *C. inophyllum* seeds and seedlings to the elevated CO_2 condition.

MATERIAL AND METHODS

The experiment was carried out at College of Forestry, Ponnampet, Kodagu, Karnataka. Seeds from matured fruits were collected and subjected to mechanical scarification by removing seed coat by hammer. A polytunnel (10.56 m X 0.71 m X 1.26 m) was erected to create elevated carbon dioxide conditions. The floor of the tunnel was spread with the farm yard manure which on decomposition releases the CO_2 and the quantity of farm yard manure to be added was decided as per the procedure given by Devakumar et al (1996). In all, 800 polythene bags were filled with the potting mixture, one batch having 400 polythene bags (100 polythene bags/replication) was kept in open condition as control, and similarly, another batch with 400 polythene bags (100 bags/replication) was placed in the CO_2 enriched condition. One seed per polythene bag was dibbled under both the conditions. Everyday observation of temperature and CO_2 concentrations in the polytunnel were recorded at 9.30 AM and 4.00 PM using CO_2 analyzer (GC 2028) and monthly average was computed. In the control condition average monthly CO_2 concentration was found to be 401.94 ppm with average temperature 25.25ºC whereas in elevated conditions CO_2 concentrations were elevated to 832.53 ppm with 26.12ºC temperature (Fig. 1).

Observation on seed germination were recorded till the end of germination. Growth parameters like seedling collar diameter and the seedling height were measured at the end of the experiment (6 months). Estimation of variations in stomatal density in the open and elevated CO2 concentrationwas carried out after 90 days of sowing. Every third leaf from the top of the plant was harvested, and its area was determined using the leaf area meter. The selected leaves were applied with a quick fix (glue) at both the surfaces which on drying, the glue containing the epidermal cell was removed to observe under the microscope. The number of stomata in the field of view was counted, and their density was calculated per unit leaf area. The observed parameters were:

Days taken for initiation of germination: Number of days taken to initiate germination in each treatment was recorded.

Germination Energy (G.E.): The per cent of seeds in a given sample that germinated up to the time of peak germination.

Germination: The germination was recorded when cotyledons emerged out from the potting mixture and expressed in percentage.

$$\text{Germination (\%)} = \frac{\text{Number of Seed germinated}}{\text{Number of seeds sown}} \times 100$$

Mean Daily Germination (MDG):

$$\text{Mean daily germination} = \frac{\text{Cumulative per cent germination}}{\text{Total number of days for germination}}$$

Peak Value (PV): Maximum mean daily germination reached at any stage of germination period.

Germination Value (GV): This is calculated by formula given by Czabator (1962)

Germination value (GV) = Average mean daily germination × Germination × Peak value

Stomatal density:

$$\text{Number of stomata per unit leaf area} = \frac{\text{Number of stomata in entire leaf}}{\text{Leaf area}}$$

Response Index (RI):

Response index for all the germination parameters under elevated CO_2 was calculated using the formula (Hegde et al 1993):

$$Response\ Index = \frac{Treatment\ mean - Control\ mean}{Control\ mean} \times 100$$

RESULTS AND DISCUSSION

The seed germination in the open area was 82.50 per cent which was significantly higher than the seed germination under CO_2 elevated condition (49.00 %). A significant negative response index value for the all the germination parameters revealed the existence of the deleterious effect of elevated CO_2 on seed germination. The number of days taken to initiate germination in open condition (22 days) was significantly lesser than the elevated CO_2 condition (27 days). The high positive response index value for the initiation of germination (0.23) indicated that the germination was prolonged under

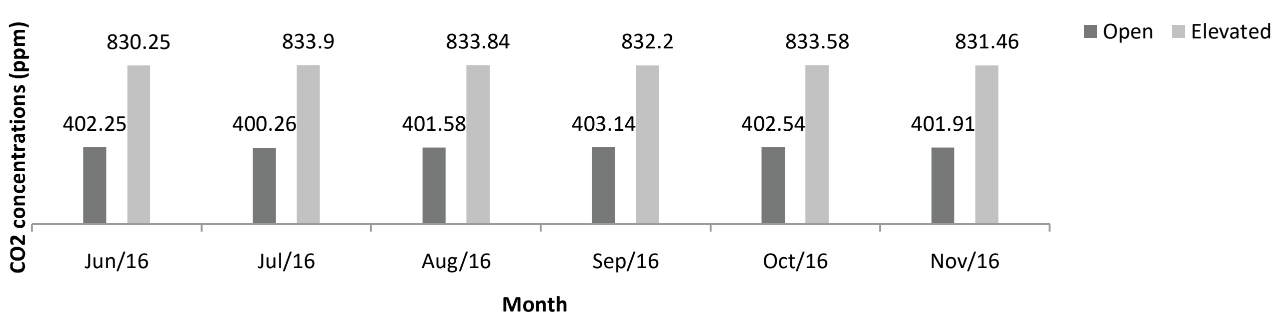

Fig. 1. Mean monthly concentration of CO_2 in open and elevated conditions

elevated CO_2 conditions. Further, the peak value (-0.05), germination value (-0.15) and the mean daily germination (-0.11) had shown a negative response index value depicting the deleterious effect of elevated CO_2 conditions on germination parameters. Germination energy, which needs to be lower for ideal germination showed a high positive response index to elevated condition (0.29) depicting that elevated CO_2 condition has a significant undesirable effect on seed germination of *C. inophyllum*. There was significant increase in the seedling height after six months was observed under the elevated CO_2 condition was indicated by the positive response index value (26.07). In contrast, the collar diameter of the seedlings was negatively affected by the elevated conditions. The study on the stomatal analysis revealed that the stomatal density was reduced under the elevated CO_2 conditions (179.98) than open condition (113.46). However, the results are not significant (Table 1).

Seed germination and speed of germination largely governs plant regeneration from seed. Although seed germination is under genetic control, parental environment can significantly influence germination characteristics. One of the environmental parameters which affect the seed germination and its rate is CO_2 concentration. In the present study, a significant effect of elevated CO_2 on germination of *C. inophyllum* was evident (Table 1). Poor germination of seeds under the elevated CO_2 condition as evidenced by the negative response index could be considered as the negative or deleterious effect of increased CO_2 in the atmosphere (Fig. 2). Seed germination was reduced by 40.6 per cent in elevated CO_2 conditions when compared to open conditions. The elevated CO_2 condition considerably reduced all the germination parameters. The major gases which play a key role in seed germination are ethylene, CO_2 and oxygen. The deprivation of oxygen by increased CO_2 and ethylene coupled with increased temperature has an inhibitive effect on seed germination. Even the higher temperature affects the sensitivity of the seed coat to the surrounding gaseous environment thereby impacting the seed germination negatively (Kigel and Galili 1995). The results were in accordance with the meta analysis carried out by Marty and Bassirirad (2014) on the impact of elevated CO_2 on tree seed germination where percentage germination responses to

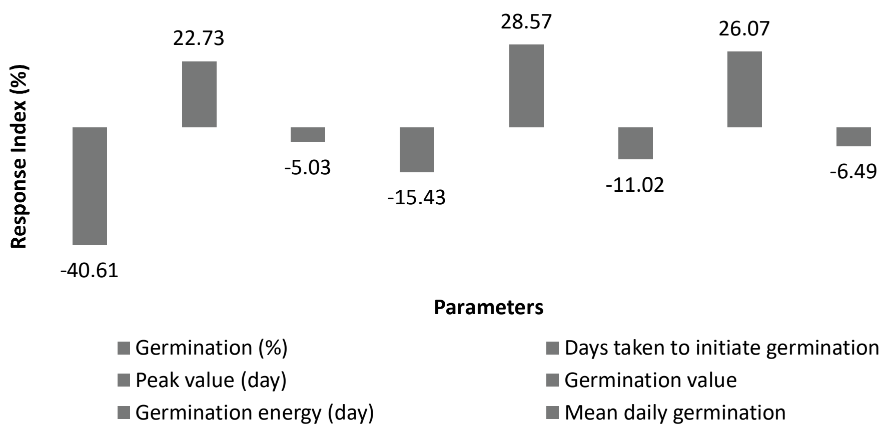

Fig. 2. Response index of the species to elevated CO_2

Table 1. Effect of elevated CO_2 on seed germination and seedling growth

Parameters	Control/Open	Elevated CO_2	t (α=0.05)	p-value
Germination (%)	82.50 (65.27)*	49.00 (44.23)	9.10	0.0001
Days taken to initiate germination	22 (4.77)+	27 (5.22)	-5.64	0.001
Peak value (day)	1.59	1.51		
Germination value	1.88	1.59		
Germination energy (day)	28	36		
Mean daily germination	1.18	1.05		
Seedling height (cm)	25.47	32.11	-2.72	0.035
Collar diameter (mm)	6.01	5.62	0.74	0.488
Stomatal density	179.98 (13.42)+	113.46 (10.65)	2.33	0.066

*Values in parenthesis indicate arcsine transformed values; +Values indicate square root transformed values increased CO_2 concentration were highly variable, ranging from a 60 per cent decrease in *Abutilon theophrasti* to a 239 per cent increase in *Pinus taed* (Marty and Bassirirad 2014).

Mean daily germination was also manipulated by the elevated CO_2 (Table 1). Maximum mean daily germination was recorded under open condition (1.18) when compared to elevated conditions (1.05). It may be due to the effect of elevated CO_2 which inhibited the seed respiration (Kigel and Galili 1995) which in turn decreased the mean daily germination (Fig. 1). Time taken to attain peak daily germination is considered as germination energy or energy period. Higher germination energy which is an indicator of slower germination was found to be more for seeds subjected to the elevated CO_2 condition. Thus, carbon dioxide increase in the atmosphere would reduce the rate of germination. Generally, seeds with higher germination and minimum energy period are ideal to produce uniform and quality seedlings. Seeds with longer energy period required more nursery retention time compared to the seeds of shorter energy period (Varsha, 2016). The delayed germination energy in case of elevated conditions could be due to the prolonged initiation of germination in elevated conditions (27 days) when compared to open conditions (22 days). Germination value of the seeds was found to be higher in the open condition than the elevated CO_2 condition. The maximum germination occurred per day (Peak value) was marginally higher at open conditions when compared to elevated CO_2 conditions. Thus, the elevated CO_2 condition has significantly impacted the seed germination and its associated parameters in *C. inophyllum*.

The present study revealed the increased seedling height under elevated CO_2 condition. The elevated CO_2 increase the carboxylation efficiency relative to oxygenation resulting in reduced photorespiration. The increase in seedling height might be due to increased use of CO_2 for carbon assimilation. Evidence from literature showed that it is possible to increase plants height under elevated CO_2concentration (Kumar et al 2001, Salimath et al 2018). But the decrease in the collar diameter of the seedlings under elevated CO_2 conditions may be attributed to the fact that, higher biomass allocation towards the height growth of the seedlings which has reduced the growth towards the lateral regions of the stem. Further, the nutrient deficit condition may also result in the production of thinner/leaner seedlings under elevated CO_2 condition. The reduction of stomatal density under elevated CO_2 condition could be attributed to the fact that, as the CO_2 level in the air surrounding the leaf increases, a lesser volume of air is necessary to obtain the requisite amount of CO_2 for the photosynthesis which implies that fewer stomata are sufficient to satisfy the need for gas exchange in the leaf. The present findings were also similar to the observations made by Ainsworth and Rogers (2007).

CONCLUSION

The results revealed that elevated CO_2 condition had significantly impacted the seed germination of *C. inophyllum.* The elevated CO_2 condition negatively influenced the germination and its parameters. But, after the germination, elevated CO_2 condition positively influenced the seedling height growth. The stomatal density of the seedlings was negatively affected by the elevated CO_2 condition. The supplementation of nutrients may increase the growth potential of the seedlings under elevated CO_2 conditions.

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Wood Properties and Utilization of Pollard Shoots of Indian Tulip Tree (*Thespesia populnea* (L.) Sol. ex Corrêa)

Navneet M. Kantariya and Satish Kumar Sinha*
College of Forestry, Navsari Agricultural University, Navsari-396 450, India
**E-mail: sksinha@nau.in*

Abstract: Forests are declining in India under severe socio-economic pressure resulting short-rotation plantation wood species as a major choice for industrial raw material. Due to shortage of raw materials, industries have to raise the fast-grown plantations of suitable wood species. Indian tulip tree (*Thespesia populnea* (L.) Sol. ex Corrêa) is one of the fast growing lesser-known tree species which is heavily pollarded and its shoots are used as fuelwood in coastal regions of South Gujarat. Since, *T. populnea* has potential for many industrial applications; therefore, a study was carried out to evaluate the physical and mechanical properties of pollard shoots of this species to check the wood quality for effective utilization. Total 20 wood samples of pollard shoots (5-10 cm diameter) from 5 trees of *T. populnea* in the girth class of 110-160 cm were collected from road side plantation in Navsari, Gujarat. The physical and mechanical properties were evaluated in air-dry condition and compared them with corresponding published values for teak (*Tectona grandis* L.f.) and 3, 4- & 5-year-old Malabar neem (*Melia dubia* Cav.). Considering physical properties, very low wood density and high volumetric shrinkage were reported in *T. populnea* compared to teak and *M. dubia*. Among mechanical properties, static bending, compressive strength and hardness of wood from pollard shoots of *T. populnea* were found to be better than 3 years old *M. dubia*. This indicates that wood from pollard shoots of thin to moderately thick diameter of this tree species can be utilized for tool handles, light construction, light packing cases and furniture.

Keywords: Static bending, Compressive strength, Volumetric shrinkage, Hardness, Wood density

Forests are declining in India under severe socio-economic pressure. Providentially, the declining rate of India's forests has come down with the enactment of the Forest Conservation Act, 1980 (Saravanan 2014). Now, the total forest and tree cover in the country is 24.62 per cent and in the state of Gujarat it is about 10.41 per cent which is low against the country's target of bringing 33.00 per cent of its geographical area under forest and tree cover as envisaged in the National Forest Policy, 1988 (ISFR 2021). The mean annual increment (MAI) of India's forest is scanty of 0.5 to 0.7 m^3 per hectare in comparison to the global average of about 2.1 m^3 per hectare (Saravanan 2014). The low productivity of Indian forest (4% of the total demand) has increased the widening gap between the demand and supply of both domestic and industrial requirement of timbers. The demand for raw materials in the furniture, construction, plywood and pulpwood industries is constantly rising, and is projected to increase further due to economic growth and a rise in population. Currently, the potential of timber production in India is 45% of the total demand. This indicates that there is a need for more timber production on a sustainable basis through agroforestry and plantation programmes to meet the domestic demand and reduce the reliance of India on imports of timbers. Due to shortage of raw materials, the industries have to establish the short-rotation fast-grown plantations of suitable wood species with tree improvement programmes for maximum yield (Sujatha et al 2023). Furthermore, there is also a need to explore the wood properties of lesser-known tree species for various end use applications. Among several lesser-known species, *Thespesia populnea* is one such species which is currently used as fuelwood in coastal regions of South Gujarat; however, this species may be suitable for timber, pulpwood and many other industrial applications after knowing its wood properties.

Thespesia populnea [(L.) Soland ex Correa] belongs to the family Malvaceae is well-known as Indian tulip tree and locally known as Paras-pipal (Troup, 1921). In India, it is a common species in the coastal tracts of the Indian Peninsula and in mangrove swamps. It is a fast-growing evergreen shrub or medium sized tree up to 20 m height with a broad dense crown with often crooked stem. The tree grows in short twists and turns with numerous limbs; therefore, lumber is only found in short lengths. Clear bole is around 2-2.5 m and girth at the breast height (GBH) is around 0.6-1.2 m. It is easily recognized by fissured grey dark brown bark, large yellow flowers with purple centre and a dense crown of green glossy heart shaped leaves (Anonymous 2022). The timber of this species is supposed to be of great local utility, being used for furniture and agricultural implements. It is an easy timber to saw and work and can be brought to a smooth surface and also takes a high polish. The wood can be carved into bowls, tools and figures. Timber is in demand for turnery and toys. It is also suitable for helves, tool handles, shuttles and other textile accessories. Wood is used for food containers, slit drums and cabinetry. It is also used as fuel wood (Anonymous 2022).

T. populnea is available in plenty around the coastal belts of the South Gujarat. It responds well to pollarding and coppices well. Currently, the pollard shoots of this species are mostly used in fuelwood. However, this species is lesser-known for various industrial applications. Therefore, a study was carried out to evaluate the physical and mechanical properties of wood from pollard shoots of *T. populnea* and compared them with corresponding published values for 'standard teak' (*Tectona grandis* L. f.) and 3, 4 and 5-year-old Malabar neem (*Melia dubia* Cav.) to check its wood quality for effective utilization.

MATERIAL AND METHODS

Total of 20 wood samples of pollard shoots (5-10 cm diameter) from 5 trees of *T. populnea* in the girth class of 110-160 cm were collected from the different blocks of Matwad road side plantation in Navsari, Gujarat. The area is located at coastal region of South Gujarat at 20° 95' N latitude, 75° 90' E longitude and at an altitude of 12 m above the mean sea level. The climate of Navsari is tropical warm with fairly hot summer, moderately cold winter and warm humid monsoon with average annual rainfall of about 1600 mm. In the current study, destructive method of sampling was adopted to collect wood samples from pollard shoots of *T. populnea*. The pollard shoots were converted into scantlings of 2 x2 x 40 cm cross section to prepare test samples from pith to periphery in four radial direction after air drying. Physical and mechanical tests were conducted as per the Indian Standard Specification IS 1708 (Part 1-18):1986 (Fig. 1a-b). Considering the physical properties, moisture content and volumetric shrinkage were determined by oven-dry method, while basic density was determined by water displacement method. The mechanical properties such as static bending, compressive strength parallel to grain, compressive strength perpendicular to grain, hardness and nail & screw holding power of of wood were tested by the Universal Testing Machine (Fig.1c-e). The data of all the parameters generated in the study were subjected to the basic statistical analysis .

RESULTS AND DISCUSSION

Considering the physical properties, the mean basic density and volumetric shrinkage based on oven-dry weight of wood samples from pollard shoots of *T. populnea* were 0.249 g/cm^3 and 47.0 %, respectively (Table 1). Orwa et al (2009) reported that the wood of *T. populnea* is light to medium in weight and basic density varied from 0.400 to 0.770 g/cm^3 at 15% moisture content in the main stem and very low to low shrinkage in this species. Similar results were also reported by Anon (2022) in an ITTO report. On contrary, very low wood density and high volumetric shrinkage in *T. populnea* were reported in the present study which may be due to the thin diameter of juvenile wood of pollard shoots. In comparison with teak and *M. dubia*, wood density of *T. populnea* recorded 37 per cent value of teak wood density and 50 per cent value of *M. dubia* wood density, while volumetric shrinkage of this species was seven times more than teak and three times more than *M. dubia*.

Considering the mechanical properties, the mean value of MOR (modulus of rupture) and MOE (modulus of elasticity) in static bending were 786.8 kg/cm^2 and 72.6 x10^3 kg/cm^2, respectively. In compressive strength parallel to grain, the

Fig. 1a-f. Test samples and testing of mechanical properties of *T. populnea* by Universal Testing Machine (a) samples for physical test (b) samples for mechanical test (c) static bending (d) compressive strength (e) hardness (f) nail and screw holding power

Table 1. Comparative profile of physical and mechanical properties of wood from pollard shoots of *T. populnea* with teak (*T. grandis*) and Malabar neem (*M. dubia*) in air-dry condition

Wood properties		*T. populnea*	Teak*	*M. dubia***		
				3 years	4 years	5 years
Moisture content (%)		15.9 ±2.14	12	12	12	12
Basic density (g/cm^3)		0.249±0.04	0.672	0.418	0.485	0.500
Volumetric shrinkage (%)		47.0±4.16	6.5	-	14.6	17.0
Modulus of rupture, MOR (kg/cm^2) in static bending		786.8±200.01	941	492.6	808.3	851.9
Modulus of elasticity, MOE (1000kg/cm^2) in static bending		72.6±23722.94	117	52.9	63.2	68.4
Maximum crushing stress (kg/cm^2) in compressive strength parallel to grain		358.3±78.45	520	-	-	-
Compressive stress at elastic limit (kg/cm^2) in compressive stren perpendicular to grain		33.3±8.87	99	31.8	68.8	104.2
Hardness (kg)	Side	444.2±64.51	500	173.7	264.1	324.2
	End	490.6±75.96	480	234.9	270.4	400.4
Nail holding power (kg)	Side	42.3±10.56	89	77.45	137.8	171.9
	End	32.3±10.12	71	59.4	65.4	91.9
Screw holding power (kg)	Side	60.4±11.80	398	248.2	325.6	366.4
	End	50.5±6.56	294	223.0	231.2	270.0

Source: * Shukla et al (2007) and ** Saravanan et al (2014)

mean value of maximum crushing stress was 358.3 kg/cm^2 and in compressive strength perpendicular to grain, the mean compressive stress at elastic limit was 33.3 kg/cm^2. The mean f side and end hardness were 444.2 kg and 490.6 kg, respectively. The mean nail holding power on side and end surfaces were 42.3 kg and 32.3 kg while, screw holding power on side and end surfaces were 60.4 kg and 50.5 kg, respectively. in an revealed that MOR and MOE of *T. populnea* in static bending were 1202 kg/cm^{2,} 119019 kg/cm^2, respectively according to ITTO report (Anonymous 2022). However, the compressive stress parallel to grain was 581 kg/cm^2. Warrier (2010) observed more bending strength, compressive stress, hardness and nail and screw holding power in the main stem of *T. populnea* in comparison to teak. In the current study static bending strength (MOR and MOE), compressive strength, and hardness of wood from pollard shoots of *T. populnea* were lower than teak but better than 3 years old *Melia dubia*. However, the nail and screw holding power was less than teak and *M. dubia* which may be due to the thin diameter of juvenile wood of pollard shoots in *T. populnea*.

CONCLUSION

The physical properties and mechanical strength of wood from pollard shoots of *T. populnea* is lower than teak wood. Among mechanical properties, static bending, compressive strength and hardness of wood from pollard shoots are better than 3- year-old *M. dubia*. This indicates that wood from pollard shoots of thin to moderately thick diameter of this tree species can be utilized for tool handles, light construction, light packing cases and furniture. However, it is necessary to evaluate the wood of main stem in *T. populnea* for better understanding of its physico-mechanical properties, so that, this lesser known species can be explored extensively for various end uses.

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Development and Performance of Mahua (*Madhuca longifolia*) Seed Decorticator

F.M. Sahu, S.H. Suthar[1], V.K. Sharma[2] and H.T. Hegde[3]

Centre of Excellence on Post Harvest Technology, ASPEE College of Horticulture
Navsari Agricultural University, Navsari, Gujarat-396 450, India
[1]Department of Processing and Food Engineering, CAET, NAU, Dediapada-393 040, India
[2]Department of Agricultural Engineering, College of Agriculture, NAU, Waghai -394 730, India
[3]Department of Forest Products and Utilization, College of Forestry, NAU, Navsari-396 450, India
E-mail: fmsphtc@nau.in

***Abstract*:** The manual decortication process of Mahua (*Madhuca longifolia*) is a tedious, costly, low output, time consuming and labour intensive process and injurious to human beings. To overcome this problem, the present investigation was carried out to develop a mahua seed decorticator and evaluate its performance. The performance parameters of developed seed decorticator were tested and evaluated with independent variables, namely four levels of seed moisture content *i.e.* 9, 12, 15 and 18 % (dry weight basis), and four levels of concave clearance *i.e.* 9, 11, 13 and 15 mm. The best performance of mahua seed decorticator was obtained at seed moisture content of 9% and a concave clearance of 11 mm, which resulted in the maximum percentage of whole kernel recovery (67.25%) with decorticating efficiency of 98.18% and overall machine efficiency of 88.63% and a desirability value of 0.932.

Keywords: Concave clearance, Decorticator, Performance, Seed moisture content

Mahua (*Madhuca longifolia*) seed is a tree-borne oilseed that is found in the states of Jharkhand, Chhattisgarh, Odisha, Madhya Pradesh, Maharashtra, and Gujarat in India, with an annual fruit production of nearly 1.81 million metric tonnes. It is one of the most important tree seed oil sources in tribal areas of India (Jha and Vaibhav 2013, Nayak and Sahoo 2020). Mahua is a medium size, deciduous tree, having height up to 16-20 m, usually with a spreading, dense with shady canopy. The tree matures from age of 8-15 years and fruits up to 60 years (Hegde et al 2018). Mahua is mainly cultivated or harvested for three major F's *i.e*. food, fodder, and fuel (Patel et al 2011, Hegde et al 2019). The tree yields several products of daily utilities. The seeds are primarily processed for their oil which is used in various food and non-food industries. Mahua fruits are green at maturity and turn reddish-yellow when ripen. Fruits are 2.5-5.0 cm long, ovoid, fleshy and have 1- 4 elongated seeds (2.0-3.5 cm) of brown colour with shiny appearance (Ramadan and Moersel 2006).

The seed contains two kernels and is highly sensitive to desiccation and freezing, indicating the recalcitrant nature of the seed. Fruits open after 45-60 days of flowering period and seeds get matured by the month of June and are available up to mid-July. The mature fruits fall on the ground on ripening and are tribal people collect the fruits and separate the seed by pressing the fruit wall manually (Patel et al 2011, Gupta et al 2012). However, they are commercially harvested during June and July in rainy season. Harvested matured fruits from the tree can also be kept for ripening and seeds are extracted by depulping the fruit. It is an important tree-based oilseed valued for its high content (33-61%) of pale yellow semi-solid fat (Ghadge and Raheman 2005). Mahua seed oil is the largest source of natural hard fat commercially known as mahua butter or mowrah butter, hence Mahua is also called as an Indian butter nut tree. The oil is edible and consumed in various rural areas and also beneficial for curing of skin diseases and smoothening of skin (Bakhara et al 2016). Because of its many uses, collection of Mahua seeds is an important activity in the annual calendar of tribal families (Yadav et al 2011, Ramadan et al 2016).

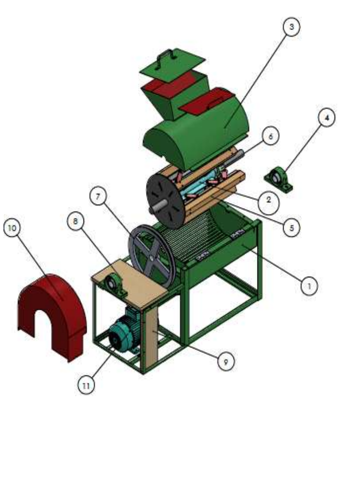
Due to a lack of efficient post harvest management, the seeds are low priced and have less demand in the market, which makes this seed underutilized. One of the important post harvest processing operations for oilseeds is decortication. Traditionally, tribal people and farmers use the manual method of decorticating the seed by applying impact force on the seed with a wooden mallet. The manual decortication process is a tedious, costly, low output, time consuming and labour intensive process and injurious to human beings. Quantity and quality of kernel out-turn fully depends upon the skill of the person. This limits the availability of mahua seed kernel and seed oil in the market. Thus, the quest for a satisfactory, effective, and economical means such as mechanized decortication technique is highly required for small and marginal farmers in the rural and tribal areas of India to reduce the excessive drudgery in mahua seed decortications. To overcome this problem, a mahua seed decorticator was designed and developed and evaluated for its performance.

MATERIAL AND METHODS

The study was conducted in Centre of Excellence on Post Harvest Technology, ASPEE College of Horticulture, Navsari Agricultural University, Navsari, Gujarat- 396450, India.

Moisture content: The moisture content of raw material was determined by following the procedure of AOAC (2000).

Design and development of the machine: Mahua seed decorticator was designed employing SOLIDWORKS software (Version 2020, Dassault Systems) and fabricated at the Centre of excellence of PHT. The machine comprises of the main frame, feed hopper, decorticating casing, a mechanism for adjusting clearance between concave and rotor assembly and a power transmission unit etc (Fig. 1). All fabrication parts were made of mild steel (MS) because it is cheap, durable, and readily available. Standard components such as belts, pulley, bearing, bearing housing *etc*. were purchased from the local market. The capacity of the developed machine was 100 kg/h. The decorticating casing comprised of a rotor assembly with six wooden rasp bars of 500 mm long and 300 mm cylinder diameter mounted on a 30 mm central mild steel shaft that rotates inside a stationary metal concave sieve. The main function of a rotating wooden rasp bar is to apply compression force on the surface of the seeds surface which helps the frangible seed coat to crack. Due to the continuous rotating action of the wooden rasp bar, impact and shearing forces inside the chamber are created, which caused the cracked seed coat to break and detach from the kernel. A feed hopper with dimensions 305 (Length) × 40 (Width) × 120 mm (Height) feeds the bulk mahua seeds into the decorticating casing at the desired feed rate of 1.66 kg/min. The feed hopper and discharge outlets were designed at a 45° angle of inclination to ensure the free flow of the seed during loading and unloading conditions. The concave was made of 26 numbers of Ø 10 mm mild steel bars at an equal spacing of 10 mm to form slotted apertures. The semicircular concave screen was made of mild steel and acted as impacting surface. A single phase 1 HP electric motor with 1440 rpm was used as a drive to rotate the cylinder at 310 rpm. The overall dimensions of the decorticator were 1000 (Length) × 420 (Width) × 850 mm (Height) having a net weight of 101 kg excluding the motor.



Item no.	Part number	Nos.
1	Concave assembly with frame	01
2	Major bush	01
3	Upper concave with	01
4	Bearing P206	02
5	Wooden rasp bar	06
6	Minor bush	01
7	Pulley	01
8	Wood ply 1	01
9	Wood ply 2	01
10	Protection guard	01
11	Motor	01

Fig. 1. Mahua seed decorticator assembly (Left) with Part details (Right)

Performance evaluation of mahua seed decorticator: The performance parameters of the mahua seed decorticator were tested and evaluated with independent variables, namely four levels of seed moisture content, i.e. 9, 12, 15 and 18 %, and four levels of concave clearance, i.e. 9, 11, 13 and 15 mm. Mahua seeds were introduced into the decorticating casing with known quantity (N0) and decortication experiments were carried out. With Factorial Composite Randomized Design (FCRD), 16 combinations were considered as treatments and each treatment had three replications. Sample of 5 kg mahua seeds was taken in each replication of 16 treatments for the decortications in the mahua seed decorticator. The speed (310 rpm) and feed rate (1.67 kg/min) were kept constant for the machine for the entire duration of decortications operation.

After each experiment, the decorticated samples were carefully collected from the outlet and grouped into four categories, namely whole kernel (N1), broken kernel and powder (N2), partially decorticated seed (N3) and un-decorticated seed (N4) and subsequently weighed. The decorticating performance was measured in terms of dependent variables, namely Percent of the whole kernel (η_k), Percent of the broken kernel and powder (η_b), Percentage of partially decorticated seed (η_{pd}), Percentage of un-decorticated seed (η_{ud}), Decorticating efficiency ((η_{de}), Overall machine efficiency (η_{me}) which were determined using (Eq. 1 - 6) using method described by Pradhan et al (2010) and Sobowale et al (2015).

Percent of whole kernel (η_k)

$$\eta_k = \frac{N1}{N0} \times 100 \qquad (1)$$

Percent of broken kernel and powder (η_b)

$$\eta_b = \frac{N2}{N0} \times 100 \qquad (2)$$

Percentage of partially decorticated seed(η_{pd})

$$\eta_{pd} = \frac{N3}{N0} \times 100 \qquad (3)$$

Percentage of un-decorticated seed (η_{ud})

$$\eta_{ud} = \frac{N4}{N0} \times 100 \qquad (4)$$

Where, N0 = amount of seed fed into the hopper, kg

Decorticating efficiency ((η_{de})

$$\eta_{de} = \left[1 - \frac{(N3 + N4)}{N0}\right] \times 100 \qquad (5)$$

Overall machine efficiency (η_{me})

$$\eta_{me} = \left[1 - \frac{(N3 + N4)}{N0}\right] \times \left[\frac{N1}{N1 + N2}\right] \times 100 \quad (6)$$

RESULTS AND DISCUSSION

The fractions (Per cent) and decortications efficiency and overall machine efficiency which were calculated using equations 1-6 (Table 1). Independent parameters like seed moisture content (M) and concave clearance (C) significantly affected the decorticating performance of the mahua seed decorticator.

At any concave clearance from 9 to 11 mm, the per cent of the whole kernel decreased as moisture content increased from 9% to 18% (db). But, at any moisture content from 9 to 18% (db), the per cent of whole kernel recovery increased for concave clearance of 9 to 11 mm but decreased for 11 to 15 mm concave clearance. This may be due to the maximum compression obtained for 11 mm concave clearance with minimum breakage. It is because of the seed was more brittle at low moisture content and it is more susceptible to mechanical damage. The reported results were well supported by the study conducted by Pradhan et al (2010) conducted study on jatropha seeds decortication at different moisture content and concave clearance. At any concave clearance from 9 to 11 mm, the per cent of the broken kernel and powder decreased as moisture content increased from 9 to 18% (db) whereas it decreased with an increase in concave clearance from 9 to 15 mm.

At any concave clearance from 9 to 11 mm, the percentage of partially decorticated seed and per cent un-decorticated seed decreased as moisture content increased from 9 to 18% (db). But, at any moisture content from 9 to 18% (db), the per cent of the partially decorticated seed and per cent of un-decorticated seed increased with an increase in concave clearance from 9 to 15 mm. The results are in line with the observation made by Shashikumar et al (2016) while testing the sal seed decorticator who reported that at higher

Table 1. Effect of the seed moisture content and concave clearance on the performance parameters of the mahua seed decorticator

Treatments	Whole kernel (%)	Broken kernel and powder (%)	Partially decorticated seed (%)	Un-decorticated seed (%)	Decorticating efficiency (%)	Overall machine efficiency (%)	Desirability
T_1 (M_1C_1)	58.87	13.19	0.79	0.56	98.65	80.59	0.312
T_2 (M_1C_2)	67.25	7.26	1.03	0.79	98.18	88.63	0.932
T_3 (M_1C_3)	63.05	7.45	2.31	1.12	96.57	86.36	0.732
T_4 (M_1C_4)	61.26	6.33	2.62	2.13	95.25	86.34	0.615
T_5 (M_2C_1)	58.12	12.83	1.14	0.91	97.95	80.24	0.080
T_6 (M_2C_2)	65.45	7.11	1.33	1.56	97.11	87.60	0.880
T_7 (M_2C_3)	64.13	6.67	2.69	2.27	95.04	86.09	0.679
T_8 (M_2C_4)	60.79	6.15	2.98	2.35	94.67	85.97	0.561
T_9 (M_3C_1)	57.79	12.66	1.56	1.45	96.99	79.56	0.037
T_{10} (M_3C_2)	64.67	6.45	1.81	1.93	96.26	87.53	0.828
T_{11} (M_3C_3)	62.04	6.11	2.87	2.56	94.57	86.10	0.625
T_{12} (M_3C_4)	60.38	6.00	3.12	2.64	94.24	85.72	0.496
T_{13} (M_4C_1)	57.12	12.40	1.65	1.57	96.78	79.52	0.134
T_{14} (M_4C_2)	62.90	6.00	2.15	2.64	95.21	86.92	0.763
T_{15} (M_4C_3)	60.93	5.90	2.70	2.65	94.40	86.07	0.553
T_{16} (M_4C_4)	59.01	5.82	3.33	2.90	94.02	85.59	0.036
S. Em. ±	0.40	0.23	0.10	0.07	0.11	0.29	-
CD at 5%	1.16	NS	0.29	0.19	0.31	NS	-
CV %	1.13	4.89	8.05	6.16	0.20	0.59	-

Where, M= Mahua seed moisture content with M_1 = 9%; M_2 = 12%; M_3 = 15% and M4 = 18%, C = Concave clearance with C_1 = 9 mm; C_2 = 11 mm; C_3 = 13 mm and C_4 = 15 mm

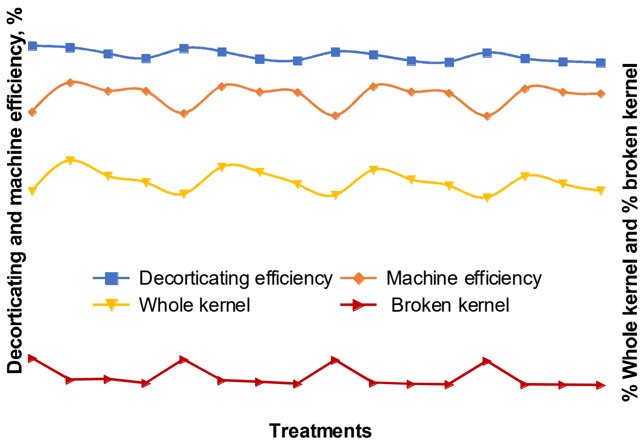


Fig. 2. Evaluation of different parameters for selection of best treatment combination of moisture content and concave clearance

Table 2. Results of optimization mahua seed decorticator with actual and predicted data at 9% (db) moisture content and 11 mm concave clearance [Treatment: $T_2 (M_1C_2)$]

Response (%)	Actual	Predicted	SE Mean
Whole kernel	67.25±0.74	66.19	0.43
Broken kernel and powder	7.26±0.61	7.24	0.16
Partially decorticated seed	1.03±0.05	1.13	0.11
Un-decorticated seed	0.79±0.07	1.87	0.19
Decorticating efficiency	98.18±0.09	97.85	0.23
Overall machine efficiency	88.63±0.73	88.22	0.18

moisture content and concave clearance, the percentage of un-decorticated seed was highest. The overall machine efficiency (%) decreased with an increase in moisture content from 9% to 18% (db) and moisture content from 9 to 18% (db). The overall machine efficiency (%) did not show any particular trend with respect to concave clearance.

Two factor interactions (2FI) were used for predicating the response. The different runs of experimental data were fitted in the 2FI model and there was 0.932 desirability of response for accrual value at 9% (db) moisture content with 11 mm concave clearance (Treatment–T_2, M_1C_2) (Figure 2). The actual and predicted values for different responses were similar to each other (Table 2). Hence, the two factorial interaction model was the best suitable model for this experiment.

CONCLUSIONS

The best performance of the mahua seed decorticator was obtained at seed moisture content of 9% (db) and concave clearance of 11 mm which resulted in maximum percentage of whole kernel recovery, decorticating efficiency, overall machine efficiency) and desirability value. The installation cost of developed mahua seed decorticator was estimated to be Rs 32,000/- with a throughput capacity of 100 kg/h. The decortication cost per kg of mahua seed was Rs. 0.88 per kg compared to Rs. 7.50 per kg of mahua seeds by manual method. Hence the developed machine was economically feasible to set up a small-scale industry in rural areas.

AUTHORS CONTRIBUTION

This work was carried out collectively by all the authors. The first author F. M. Sahu designed the study, formulated the protocol, methodology, collected the data resources, formal analysis, investigation, visualization and wrote the first draft of manuscript. The third author V. K. Sharma checked design and fabrication of equipment, managed the analysis of the data, figures and the tables. The fourth author H. T. Hegde managed the visualization, literature resources and reviewed the editing. The second author S. H. Suthar supervised the complete experiment and approved the final manuscript. All authors had read and approved the final manuscript.

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Performance of Mustard Crop Under Citrus Based Agroforestry Systems in Vidarbha Region of Maharashtra

Aarti P. Deshmukh, V.M. Ilorkar, P.D. Raut and Lalji Singh

Department of Forestry, Indira Gandhi Krishi Vishwavidyalya Raipur-492 012, India
E-mail: aartideshmukh776@gmail.com

***Abstract*:** Traditional agroforestry is very common in Vidarbha region of Maharashtra with forest tree crops. Now fruit tree crops are also introduced to increase the farmer's income. Hence, the experiment was conducted at AICRP on Agroforestry research farm at College of Agriculture, Nagpur under citrus based agroforestry systems. The citrus fruit trees were planted at 6 x 6 m and forest species including *Tectona grandis, Eucalyptus teretocornis* and *Ailanthus excelsa* at equilateral distance of 3 meter in each treatment between two mandarin trees, where, Mustard crop (*Brassica juncea* var. Pusa bold) was cultivated as traditional agri-horti-silviculture system during 2020-21. The growth performance and yield of Mustard under different set of treatments viz. T_1 (Sole mustard), T_2 (Sole mandarine + mustard), T_3 (Mandarin + *Tectona grandis* + mustard), T_4 (Mandarin + *Eucalyptus teretocornis* + mustard) and T_5 (Mandarin + *Ailanthus excelsa* + mustard) was recorded. The growth parameter and yield of mustard crop was found maximum in open field crop (Sole Mustard) than the treatment under citrus based agroforestry systems and it was 25.57, 56.35, 44.00 and 63.12 per cent higher as compared to treatment T_2, T_3, T_4 and T_5, respectively. The grain and straw yield of mustard was 5.75 and 11.90 q ha^{-1} in crop of open field (Sole mustard). As regard the soil fertility, the most important parameter i.e., the organic carbon was significantly increased in sole cropping (0.57), sole Mandarin (0.53) and Mandarin + *Ailanthus excelsa* (0.48). Whereas, it decreased under Mandarin + *Tectona grandis* (0.48) and Mandarin + *Eucalyptus tereticornis* (0.44) after the harvesting of *rabi* mustard crop.

Keywords: Citrus based agroforestry system, Mustard, Productivity*, Ailanthus*

Agroforestry is a land use system that integrates trees, crops and animals in a way that is scientifically sound, ecologically desirable, practically feasible, and socially acceptable to the farmers (Nair 1993). Agroforestry systems can be advantageous over conventional agricultural and forest production methods through increase productivity, economic benefits, social outcomes and the ecological goods and services provided. Agroforestry is one of the best option to increase the tree cover outside the forest. An efficient agroforestry system not only maximizes the benefit it provides but also ensures the link to climate change mitigation. Trees are important carbon warehouses that filters massive quantities of carbon from the atmosphere, trapping it in their biomass (Montagini and Nair 2004). An average tree can remove about 23kg of carbon dioxide from the atmosphere annually. In agroforestry systems, the amount of carbon sequestration is further increased. The interaction of the different components of agroforestry systems can help absorb and sequester carbon dioxide and other greenhouse gasses from the atmosphere (Pandey 2002). Thus, trees in an agroforestry system make it a potential strategy in mitigating climate change (Patra and Behera 2014). XV finance Commission in its report submitted in July 2020, mentioned that in 2017 total consumption of wood in India was 65 million cum of which 3 million cum was produced from forests, around 47 Million cum was produced from plantations (mainly agro forestry systems), and balance 15 million cum was imported (Anon 2020). This clearly brings out the fact that around 25% of the demand for industrial wood is met from imports. Of the balance met with through domestic production around 94% is from ToF, primarily from agroforestry (Bansal 2021). Keeping this in view, an investigation was carried out to study the performance of mustard crop (*Brassica juncea* var. Pusa bold) under citrus based agroforestry systems in Vidarbha Region of Maharashtra.

MATERIAL AND METHODS

Nagpur tract fall in sub-tropical zone, at a latitude of 21° 14' N and longitude 79° 08' E at an elevation of 310 meter above mean sea level. The climate of the area is semiarid characterized by three distinct seasons i.e., summer, rainy and winter. The normal mean annual precipitation is 1064.1mm and the major share of precipitation is received during the period of June to September. Winter rains are few and uncertain. The normal mean monthly temperature varies from 27.7°C to 41.7°C during the hottest month (May), while the mean monthly minimum temperature is ranges from

14.5°C to 29.5°C in the coldest month (December). The selected level field of 0.29 ha in which horti-silviculture-system including Mandarin, *Tectona grandis, Eucalyptus tereticornis* and *Ailanthus excelsa* planted during 2015 was selected. The experiment was laid out in Randomized Block Design (RBD) with four replications. There were 20 treatments. The size of unit plot was 6 X 6 m^2. The Mandarin was planted at a distance of 6 x 6 m^2, whereas the forest species were planted at a distance of 3m between two mandarin plantations.

Mustard (*Brassica juncea*) Variety (Pusa bold) was cultivated as per recommended package and practices in rabi season. The treatments includes T_2 - Sole Mandarin + Mustard, T_3 - Mandarin + *Tectona grandis*+Mustard, T_4-Mandarin + *Eucalyptus tereticornis* + Mustard and T_5-Mandarin + *Ailanthus excels* + Mustard were frame, while T_1-sole mustard field area without trees were laid down to accesses the growth and yield performances of mustard crop affected by microclimate condition available to mustard crop at different growth stages, the observation were made for plant population m^{-2}, height of plant (cm), number of branches plant^{-1} number of leaves plant^{-1} number of siliqua plant^{-1}, seed siliqua^{-1}, length of siliqua (cm), grain yield (q ha^{-1}), straw yield (q ha^{-1}), harvest index (%), test weight (gm)1000 seed^{-1}, fresh weight (kg m^{-2}), dry weight (kg m^{-2}), fresh root weight plant^{-1} (gm) and root length (cm). The dry weight of sample was estimated after drying of sample at 75°c for 24 hours. The microclimate observation i.e., solar radiation, atmospheric temperature, humidity where recorded at different growth by wind LICOR-photo meter, digital thermometer & hygrometer at 9.00 to 10.00, 12.00 to 13.00 and 15.00 to 16.00 hours in each replication of the treatments during crop period. The soil of the crop area was analyzed for in organic carbon, pH, electric conductivity (EC), nitrogen, phosphorus and available potassium after harvesting of crop as per standard methods of Jackson (1957), Piper (1967), Olsen et al (1954) and AOAC (2002).

RESULTS AND DISCUSSION

The highest plant population per square meter of mustard (30 m^{-2}) was recorded under sole cropping followed by sole Mandarin (29.50 m^{-2}) based agroforestry system (Table 1). Whereas, the minimum plant population per square meter (27.25 m^{-2}) was recorded under Mandarin + *Eucalyptus tereticornis* based agroforestry system. No significant differences in plant population were found among different agroforestry systems. The per cent reduction in plant population of mustard was 1.66, 7.5, 5, 9.16 per cent under sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus tereticornis* and Mandarin + *Ailanthus excelsa* as compared to sole cropping. Rahangdale et al. (2014) recorded highest reduction in plant population in soyabean (21.61%) and the lowest in paddy (12.35%). They also noticed that the moong and til showed almost the similar trend of reduction (16.06% and 17.63%, respectively) under bamboo based agri-silviculture system over the control that is sole crop. In mustard, significant difference in plant height were observed at 30 and 60 DAS (Table 1). Significantly maximum plant height was recorded in sole mustard i.e. 47.00 and 106.25 cm at 30 and 60 DAS. However, no significant difference in plant height was recorded at 90 DAS. Amongst the different citrus based agroforestry system, maximum plant height was observed in sole Mandarin. Less height under Mandarin + *Ailanthus excelsa* agroforestry systems may be primarily due to reduced light intensity. Rahangdale et al (2014) also recorded reduction in plant height of paddy (3.90 %) and sesame (2.84%) as compared to soybean (8.83 %) and moong (7.57%) under bamboo based agri-silviculture system over the control that is sole crop which may be because bamboo canopy could have even affected the proper penetration of light on the understory annual crop. Significant differences in number of branches plant^{-1} of mustard were observed under citrus based agroforestry systems at 60 and 90 DAS of mustard (Table 1). However, no significant difference was observed in number of branches plant^{-1} at 30 DAS of mustard. Significantly highest number of branches plant^{-1} at 60 DAS (4.17) and 90 DAS (4.91) was recorded under sole cropping followed by sole Mandarin (3.88 and 4.68), Mandarin + *Eucalyptus tereticornis* (3.67 and 4.55), Mandarin + *Tectona grandis* (3.60 and 4.41) and Mandarin + *Ailanthus excelsa* (3.36 and 4.33). Rahangdale et al (2014) noticed that among the *kharif* crop highest reduction in soybean (33.66 %) fro number of branches per plant and the lowest was in paddy (23.75 %) for number of effective tillers per plant. They also observed that the moong and sesame showed the almost similar trend of reduction (31.32 % and 30.41 %, respectively) for number of branches per plant under bamboo based agri-silvicuture system over control that is sole crop. In mustard, no significant difference was observed in number of leaves plant^{-1} at 30 and 90 DAS (Table 2). The significantly maximum number of leaves plant^{-1} was recorded in mustard at 60 DAS (9.08) under sole cropping. Kumar and Nandal (2004) found that the entire test crop sown in the interspaces of *Eucalyptus teretocornis* showed reduced plant vigour in terms of plant height, stem diameter, number of branches, number of leaves and yield attributes as compared to sole cropping. There were no significant differences in number of siliqua plant^{-1} amongst the different treatments. However, maximum number of siliqua plant^{-1} was recorded in sole cropping in mustard (130.00) and lowest was in Mandarin + *Ailanthus excelsa* agroforestry system i.e., 101.20. Kumar et al (2013) found that parameters such as plant running meter row lay, spike length, grains spike^{-1} and test weight was significantly less under *Eucalyptus tereticornis* than sole cropping. In mustard, seeds siliqua^{-1} was less under *Eucalyptus tereticornis* than sole cropping. Yield parameters such as secondary siliqua plant^{-1} and test weight were also significantly higher in sole cropping.

The effect of agroforestry system on seed siliqua^{-1} of mustard as compared to sole cropping indicated that number of seed siliqua^{-1}was significantly maximum in sole cropping in cowpea (16.45) and the lowest seed siliqua^{-1} were in Mandarin + *Ailanthus excelsa* agroforestry system (11.05). The percent reduction in seed siliqua^{-1}was 9.73, 26.26, 16.41 and 32.83 in mustard under sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus tereticornis* and Mandarin + *Ailanthus excelsa*, respectively as compared to sole cropping of intercrops. This may be due to competition of light among the annuals and perennials. Kumar *et al.* (2013) observed that parameters such as plant running meter row lay spike length, grains per spike and test weight was significantly less under *Eucalyptus tereticornis* than sole cropping. In mustard, seeds siliqua^{-1} was less under *Eucalyptus tereticornis* than sole cropping. Yield parameters such as secondary siliqua per plant and test weight were also significantly higher in sole cropping.

Highest length of silica (5.23 cm) of mustard was in sole cropping and lowest was in Mandarin + *Ailanthus excelsa* agroforestry systems (4.30 cm). All the treatments were at par with each other. The yield attributing parameter i.e. length of siliqua (cm) of mustard was highest under sole cropping as compared to sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus tereticornis* and Mandarin + *Ailanthus excelsa* agroforestry systems, respectively may be due to competition of light among the annuals and perennials. Kumar et al (2013) observed that plant running meter row lay, spike length, grains per spike and test weight of wheat was significantly less under *Eucalyptus tereticornis* than sole cropping. In mustard, seeds siliqua^{-1} was less under

Table 1. Effect of tree crops on plant population m^{-2}, height of plant (cm), number of branches plant^{-1}, number of leaves plant^{-1}, number of siliqua plant^{-1}, seed siliqua^{-1} and length of siliqua (cm)of cowpea under different agroforestry systems

Treatment	Plant population m^{-2}	Height of plant (cm)			Number of branches cowpea plant^{-1}			Number of leaves plant^{-1}			No. of siliqua plant^{-1}	Seed siliqua^{-1}	Length of siliqua (cm)
		30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS			
Sole cropping	30.00	47.00	106.25	138.75	1.28	4.17	4.91	5.46	9.08	9.98	130.00	16.45	5.23
Mandarin	29.50	46.00	102.00	135.25	1.25	3.88	4.68	5.42	8.43	9.31	128.50	14.85	5.03
Mandarin + *Tectona grandis*	27.75	42.00	98.00	126.75	1.13	3.60	4.41	5.15	7.75	8.61	107.55	12.13	4.38
Mandarin + *Eucalyptus tereticornis*	28.50	44.25	99.75	130.50	1.22	3.67	4.55	5.28	7.85	8.82	122.10	13.75	4.67
Mandarin + *Ailanthus excels*	27.25	40.50	94.50	122.75	1.08	3.36	4.33	5.00	7.45	8.11	101.20	11.05	4.30
SE(m) ±	1.65	2.66	3.50	2.44	0.09	0.18	0.19	0.29	0.45	0.58	54.46	1.42	0.38
CD @ 5%	NS	8.21	10.79	NS	NS	0.57	0.59	NS	1.40	NS	NS	4.38	1.19

Table 2. Effect of tree crops on grain yield(q ha^{-1}), straw yield(q ha^{1}), harvest index (%),test weight (gm) 1000 seed^{-1}, fresh weight (kg m^{-2}), dry weight (kg m^{-2}), fresh root weight plant^{-1} (gm) and root length (cm) mustard under citrus based different agroforestry systems

Treatments	Grain yield (q ha^{-1})	Straw yield (q ha^{-1})	Harvest Index (%)	Test weight (gm) 1000 seed^{-1}	Fresh weight (kg m^{-2})	Dry weight (kg m^{-2})	Fresh root weight plant^{-1} (gm)	Root length (cm)
Sole cropping	10.31	20.48	33.48	4.50	1.57	0.67	8.80	14.50
Sole Mandarin	8.51	18.05	32.04	4.23	1.46	0.60	8.53	14.25
Mandarin + *Tectona grandis*	7.45	16.24	31.44	3.90	1.11	0.44	8.10	12.95
Mandarin + *Eucalyptus tereticornis*	7.93	17.04	31.75	4.15	1.37	0.57	8.33	13.15
Mandarin + *Ailanthus excels*	6.82	15.00	31.25	3.70	0.79	0.35	7.94	11.65
SE(d) ±	0.87	1.45	1.06	0.47	0.19	0.13	2.92	3.44
CD @ 5%	NS	NS	NS	1.45	0.59	NS	NS	NS

Eucalyptus tereticornis than sole cropping. Yield parameters such as secondary siliqua plant^{-1} and test weight were also significantly higher in sole cropping.

Grain and straw yield (q ha^{-1}) of mustard under different agroforestry systems: It was observed that the due to competition for moisture, light and nutrients among the annual crops, trees and fruit plants, the observed values for different yield attributing parameters were lesser than the sole cropping system. As regards mustard yield, highest grain and straw yield was recorded under sole cropping i.e. 10.31 and 20.48 q ha^{-1}which was 17.46, 27.74, 23.08 and 33.85 per cent higher in grain and 11.87, 21.78, 15.63 and 26.76 per cent higher in straw than the yield recorded under sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus tereticornis* and Mandarin + *Ailanthus excelsa* agroforestry systems, respectively (Table 2). Highest harvest index of mustard intercrop was in sole cropping, whereas, lowest harvest index was recorded under Mandarin + *Ailanthus excelsa* agroforestry systems. In mustard, the harvest index under sole cropping was 4.19, 5.08, 5.98 and 6.58 per cent higher as compared to sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus tereticornis* and Mandarin + *Ailanthus excelsa* agroforestry systems, respectively and may be due to competition of light among the annuals and perennials. Rahangdale et al. (2014) recorded that the soyabean (67.88%) and moong (61.30%) showed relatively higher reduction in grain and straw yield as compared to sesame (49.25%) and paddy (34.00%) in old bamboo based agri-silviculture system over the sole crops and this reduction in grain yield may be due to less PAR (photosynthesis active radiation) interception and available energy below the canopy of bamboo species in comparison to sole crop (open condition). These results are also in conformity with the findings earlier workers (Kaushik et al 2002, Kiran et al 2002, Swamy et al 2003, Yadav et al 2005 , Bijalwan et al 2009).

Highest test weight of mustard was recorded under sole cropping (4.50 gm 1000 seed^{-1}) (Table 2). All the treatments were at par with each other. The test weight of mustard in sole cropping was 6.0, 13.33, 7.77 and 17.77 per cent higher as compared to sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus tereticornis* and Mandarin + *Ailanthus excelsa*. Johar et al (2017) observed the effect of *Eucalyptus tereticornis* based agri-silvi-horticultural system on growth and yield of wheat and test weight of wheat was significantly reduced under Kinnow + wheat (46.1 gm) and Kinnow + Eucalyptus + wheat agroforestry model as compared to sole cropping .

Significantly highest fresh weight of mustard (1.57 kg m^{-2}) was under sole cropping followed by sole Mandarin (1.46 kg m^{-2}) and lowest was in Mandarin + *Ailanthus excelsa* agroforestry systems (0.79 kg m^{-2}). All the agroforestry systems were at par with each other for fresh weight of mustard except sole Mandarin and sole cropping. The dry weight, fresh root weight plant^{-1} and root length of mustard was not significantly affected by different agroforestry systems. However, highest dry weight, fresh root weight plant^{-1} and root length was recorded under sole cropping i.e., 0.67 kg m^{-2}, 8.80 gm and 14.50 cm, respectively. The lowest values of all parameters i.e., dry weight, fresh root weight plant^{-1} and root length were in Mandarin + *Ailanthus excelsa* agroforestry system i.e., 0.35 kg m^{-2}, 7.94 gm and 11.65 cm, respectively.

Available soil nutrient content: The organic carbon, available soil nitrogen (N) and available phosphorus (P) and available potassium (K) was ranged between 0.44-0.53 (%), 173.05-231.10 (kg ha^{-1}), 16.03-17.88 (kg ha^{-1}) and 268.80-314.50 (kg ha^{-1}), respectively (Table 3). The highest soil available nitrogen, phosphorus and potassium were recorded in sole cropping and lowest values of soil available nitrogen (173.05 kg ha^{-1}) and phosphorus (16.03 kg ha^{-1}) were in Mandarin+ *Eucalyptus tereticornis* agroforestry system. Lowest value of soil available potassium (268.80 kg ha^{-1}) was in Mandarin + *Ailanthus excelsa.*

The fertility status of soil after harvest of cowpea crop infers that organic carbon and available soil nitrogen (N) were significantly, whereas, available phosphorus (P) and available potassium (K) was non-significantly affected by the different citrus based agroforestry systems as compared to sole cropping. The organic carbon was significantly increased in sole cropping (0.57), sole Mandarin (0.53) and Mandarin + *Ailanthus excelsa* (0.48) and decreased under Mandarin + *Tectona grandis* (0.48) and Mandarin + *Eucalyptus tereticornis* (0.44) after the harvesting of rabi crop. The initial available nitrogen status under sole cropping, sole Mandarin, Mandarin + *Ailanthus excelsa* was 231.10, 225.15 and 199.95 kg ha^{-1} and increased to 239.00, 230.23 and 207.90 kg ha^{-1}, respectively after harvesting of rabi season mustard crop. In Mandarin + *Tectona grandis* and Mandarin + *Eucalyptus tereticornis*, the nitrogen status before sowing of rabi mustard crop was 195.73 and 173.05 kg ha^{-1} and significantly reduced to 187.08 and 166.63 kg ha^{-1}. Similar results were also observed for potassium (K) which was initially observed as 314.50, 27, 275.85, 282.80, 280.00 and 268.80 kg ha^{-1} under sole cropping, sole Mandarin, Mandarin + *Tectona grandis*, Mandarin + *Eucalyptus teretocornis* and Mandarin + *Ailanthus excelsa*, respectively significantly reduced to 311.75, 273.50, 274.58, 273.10 and 263.75 kg ha^{-1} after harvest of rabi mustard crop. This may be due to over utilization of the nutrients by the different components in the agroforestry system as returned to soil in the form of litter fall and its decomposition. There were no significant differences in the available status of phosphorus in soil before sowing and after harvesting of the rabi mustard crop. Soil moisture was significantly highest during kharif season as compared to rabi season.

Nutrients are made available to plants in agroforestry mainly by atmospheric nitrogen fixation and mineralization of nutrients from organic forms (Muthuri et al 2005, Fang et al 2008, Jose 2009). The intercropping of trees with crops that are above to biologically fix nitrogen is common in tropical agroforestry systems. Non N-fixing trees

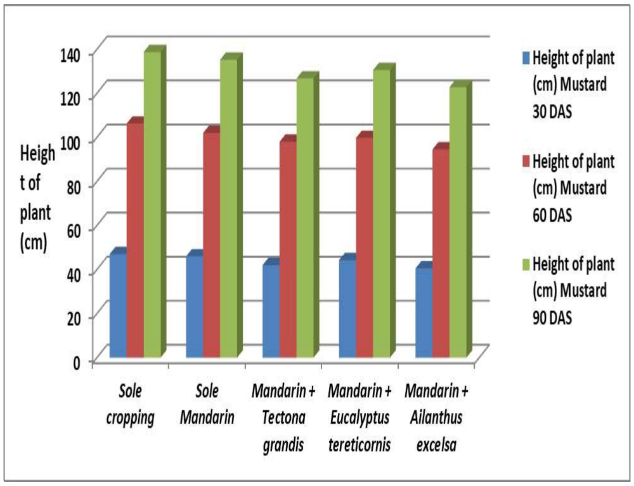

Fig. 1. Effect of tree crops on height of plant (cm) of mustard under different citrus based agroforestry systems

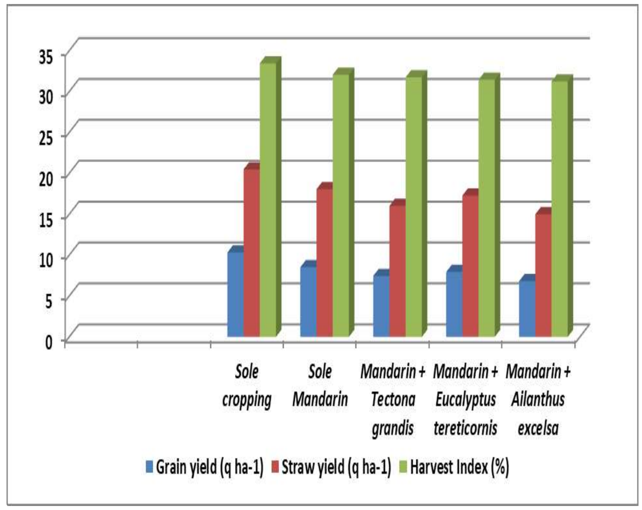

Fig. 2. Effect of tree crops on grain and straw yield (q ha^{-1}) of mustard under different Citrus based agroforestry systems

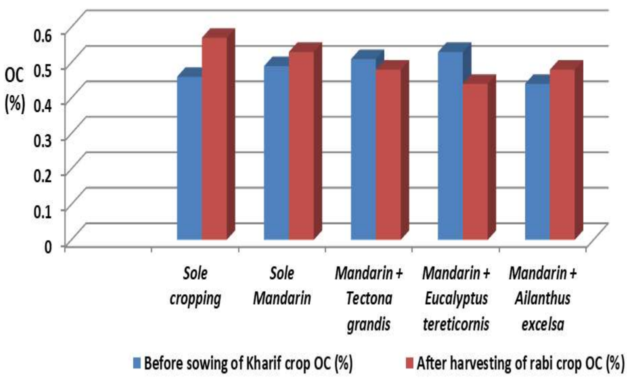

Fig. 3. Effect of tree crops on soil organic carbon content before sowing and after harvest of rabi crop

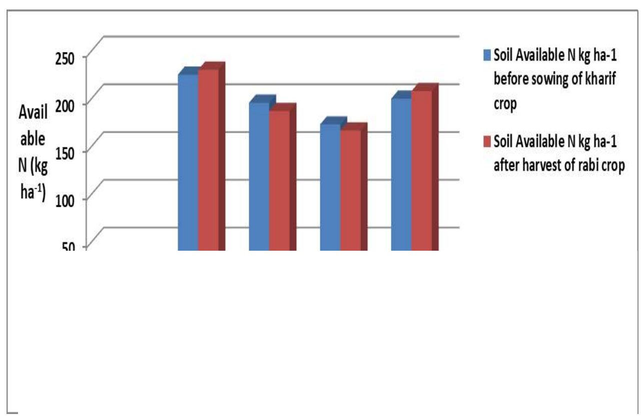

Fig. 4. Effect of tree crop on soil available nitrogen status under different citrus based agroforestry system

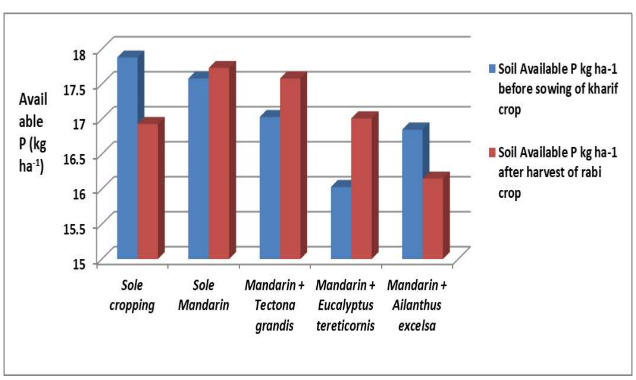

Fig. 5. Effect of tree crop on soil available phosphorus status under different citrus based agroforestry system

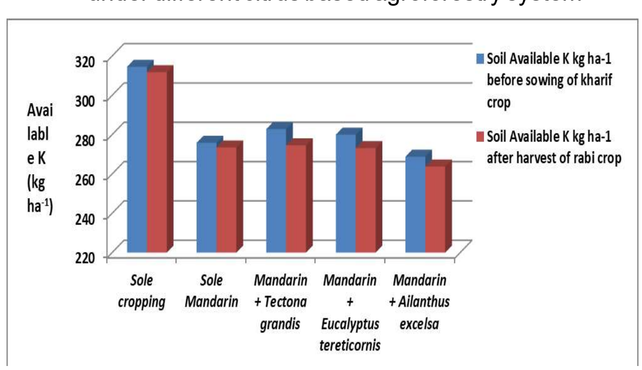

Fig. 6. Effect of tree crop on soil available potassium status under different citrus based agroforestry system

Table 3. Initial and final soil fertility status of soil under different agroforestry systems

Treatment	Before sowing of *kharif* crop						After harvest of *kharif* crop					
	pH (1:2)	EC (dSm^{-1})	OC (%)	Available Nutrients ($kg\ ha^{-1}$)			pH (1:2)	EC (dSm^{-1})	OC (%)	Available Nutrients ($kg\ ha^{-1}$)		
				N	P	K				N	P	K
Sole cropping	7.14	0.38	0.46	231.10	17.88	314.50	7.17	0.36	0.57	239.00	16.93	311.75
Sole Mandarin	7.16	0.32	0.49	225.15	17.58	275.85	7.10	0.31	0.53	230.23	17.73	273.50
Mandarin + *Tectona grandis*	7.21	0.29	0.51	195.73	17.03	282.80	7.24	0.32	0.48	187.08	17.58	274.58
Mandarin + *Eucalyptus tereticornis*	7.25	0.30	0.53	173.05	16.03	280.00	7.22	0.31	0.44	166.63	17.01	273.10
Mandarin + *Ailanthus excelsa*	7.15	0.29	0.44	199.95	16.85	268.80	7.17	0.30	0.48	207.90	16.15	263.75
SE(d) ±	-	-	-	-	-	-	0.06	0.03	0.08	16.82	1.41	20.13
CD (0.05)	-	-	-	-	-	-	NS	NS	0.26	51.85	NS	NS

can also affect soil physical, chemical and biological properties by adding some amount of organic matter and releasing and recycling of nutrients in agroforestry system (Paoli et al 2008, Yadav et al 2008).

CONCLUSION

The growth parameters and yield of mustard crop was maximum in sole cropping as compare to other citrus based agroforestry systems. The grain yield and straw yield of mustard was higher in sole cropping (sole mustard). The organic carbon significantly increase in sole cropping, mandarin +mustard (and mandarin + *Ailanthus excelsa* +mustard whereas it was decrease under mandarin + *Tectona grandis* + Mustard 0.48% and mandarin + *Eucalyptus tereticornis* + mustard. Similar trend also observed with regards to other nutrients.

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Effect of Different Spacing of Eucalypts (*Eucalyptus tereticornis*) based Agroforestry System on Performance of *Rabi* Field Crops

V. Dalal, K.S. Ahlawat, Rajesh Kathwal and K.K. Bhardwaj
Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004, India
E-mail: v.dalal1979@gmail.com

Abstract: The shrinkage of land is a big problem in the present time and to harvest the full opportunity of the land under different agroforestry system seems to be one the solution to the spatial and temporal arrangement of the different crops. The field experiment based on agroforestry system was conducted to find out the suitable crop combinations for *rabi* season and its economics at Farm Forestry, CCSHAU, Hisar, in which, eucalypts (P-23) planted in October 2018 was taken as forest tree with spacing: 6×3, 7×3, 8×3, 9×3m and *Rabi* crops *viz.,* wheat (HD 2967), barley (BH -393), raya (RH-30) as agricultural crops. The results revealed that three year old eucalypts planted at a spacing of 9×3 m exhibited significantly higher plant height (9.13 m) as compared to 8×3, 7×3 and 6×3 m spacing. The edaphic factors like pH (7.92-8.02), $EC_{1:2}$ (1.7-2.1dS/m), organic carbon (OC) ranged from 0.30-0.39 per cent of the field with different spacing were inclined to salinity, However, minimum grain yield reduction (11.34%) was observed in barley at eucalypts spacing 9×3m followed by 8×3m as compared to sole crop. In addition to this raya crop was found suitable under eucalypts based agroforestry system in 9×3m spacing. Maximum B: C ratio was obtained with eucalypts + mustard agroforestry system under two spacing 8×3m and 9×3 m *i.e.,* 1.66 and 1.67, respectively.

Keywords: Edaphic conditions, Eucalypts, Organic Carbon, *Rabi* crops and spacing

Haryana with geographical area of 4.42 million hectare is agrarian state carrying forward with intensive and mechanical agriculture, thusly, Forest cover limited to 1603 km^2 which makes 3.63 per cent of its geographical area (State of Forest Report 2021). Substantially, there is need to integrate trees and shrubs on farmlands and rural landscapes to enhance productivity, profitability, diversity and ecosystem sustainability (National Agroforestry Policy 2014). Agroforestry currently meets 50 per cent of the demand for fuel wood, 60 to 70 per cent of the demand for small timber, 70 to 80 per cent for plywood, 60 per cent of the raw material for paper pulp, and 9-11 per cent of the demand for green fodder, in addition to meeting the subsistence needs of households for grain, fruit, fibre, medicine, and so on (Arya et al 2018).

In India salt-affected soils occupy about 6.73 million hectares. Indo-Gangetic plains that lie between 21°55′-32° 39′N and 73°45′-88°25′E comprising of the states of Punjab, Haryana, Uttar Pradesh and part of Bihar (North), West Bengal (South) and Rajasthan (North) have about 2.7 million hectare salt-affected soils (Singh 2017).

MATERIAL AND METHODS

The present study was conducted at the Research Farm, Department of Forestry, CCS HAU, Hisar, situated at 29^0 09' N latitude and 75^0 43' E longitudes at an elevation of 215.2 m above mean sea level situated in the arid region of North Western India. The climate is subtropical-monsoon with an average annual rainfall of 350-400 mm, 70-80 per cent of which occurs during July to September. The summer months are very hot with maximum temperature ranging from 40 to 45°C in May and June whereas, December and January are the coldest months (lowest January temperature may reach as low as 0°C). The mean monthly values of weather parameters *viz.,* temperature, relative humidity, evaporation and rainfall were recorded during the period of experimentation. The weather data during the study period 2019 to 2021 like maximum temperature ranged from 30.4 to 31.1°C, minimum temperature 16.6 to 17.4°C, morning relative humidity ranged from 83 to 85 per cent, evening relative humidity 48 to 49 per cent, average wind speed 4.6 to 4.7 km per hour, bright sun shine hours 5.9 to 6.7, pan evaporation 4.0 to 4.1 mm, total rain fall 474.8 to 795.0 mm and total rainy days 31-34 days (Table 1). There were four spacing of Eucalypts (P-23) spacing *i.e.*, (i) 6×3m, (ii) 7×3m, (iii) 8×3m, (iv) 9×3m. The plantation of eucalypts trees was done during October, 2018. In the *rabi* season, wheat, barley and raya were sown in different eucalypts based agro forestry system during 2020-21. The seeds of variety for wheat was HD 2967, BH 393 for barley and RH 30 for raya were procured Department of Plant Breeding, CCSHAU, Hisar, Haryana. The respective varieties of the crops were included in the research for their peculiar characteristics which fit in the Agro forestry system as the experimental site falls under the semi-arid zone. There were three replications in the experiment with randomized block design. Among different spacing, soil pH ranged from 7.92 to 8.02, EC 1.7 to 2.1 dS/m, soil organic carbon 0.30 to 0.39 per cent, available N 102.4 to 112.8 kg ha^{-1}, available P 9.8 to 12.4 kg ha^{-1} and K_2O 212.4 to 228.2 kg ha^{-1} under different tree spacing and control (Table 5).

Soil data: Organic carbon was determined by Walkley and Black (1934) method, soil pH and electrical conductivity by Antil et al. (2002), available nitrogen by Asija and Subbiah (1956) method, available phosphorus by Olsen et al (1954) and available potassium by Jackson (1973) method.

Plant height: Eucalypts plant height was measured in meters with Clinometer from the base up to the tip of the tree. Measurements were done at sowing time and after harvesting of wheat, barley and raya crops.

Girth at breast height (GBH): Circumference (C) of ten randomly selected trees were measured from 1.37 m height from the ground level and converted into dbh by using the formula:

$$dbh = C/\pi$$

It was measured in cm with the help of measuring tape. The measurements were taken at sowing time and after harvest of wheat, barley and raya.

Crop parameters: The various yield attributes and yield of wheat, barley and raya were measured with the standard techniques of each crop. *B: C ratio:* Benefit cost ratio was worked for each crop with the given formula:

$$B{:}C = \frac{Gross\ returns(Rs.)}{Total\ cost(Rs.)}$$

RESULTS AND DISCUSSION

Growth studies: Three year old eucalypts planted at a spacing of 9×3 m exhibited significantly higher plant height (9.13 m) than other spacing's *i.e.,* 8×3, 7×3 and 6×3 m spacing plantation however, gbh was statistically at par with 8 x 3 m spacing (Table 2).

Yield attributes and yield: The yield attributes in wheat like grains/ spike, number of tillers /m^2 and grain yield/ ha were found less under 9×3 m (41.93, 409.98 and 41.65 q/ha) and 8×3 m (41.00, 367.66 and 41.15 q/ha) as compared to control *i.e.*, sole cropping system (44.62, 410.00 and 52.08 q ha^{-1}), respectively (Table 3 and Table 4). The yield attributes in barley like grains/spike, number of tillers/m^2 and grain yield /ha were found lower under spacing's 9×3m (40.65, 353.54 and 37.46 q ha^{-1}) and 8 × 3 m (39.50, 349.10 and 37.39 q ha^{-1}) as compared to control *i.e.,* sole cropping system (42.80, 357.50 and 42.25 q ha^{-1}), respectively. The yield attributes in mustard like plant height, number of primary and secondary branches, siliqua/ plant, number of seeds/ siliqua and grain yield/ ha were found less under tree spacing 9×3 m (167.60, 5.00, 9.00, 240.00, 12.16 and 18.15 q ha^{-1}) and 8×3m (167.30, 5.00, 8.30, 230.60, 11.90 and 18.03 q ha^{-1}) with control *i.e.*, sole cropping system (168.70, 5.25, 9.00, 263.00, 13.10 and 20.58 q ha^{-1}), respectively. The yield attributes and yield of wheat, barley and raya were found maximum under spacing 9×3 and 8×3 m spacing might be due to the reason that under broader spacing less light competition, more availability of nutrients and less competition for moisture among the field crops and eucalypts. Deep-rooted plants consume more deep soil moisture than conventional cropland due to their well-developed and deeper root systems and higher evapotranspiration rates (Zhang et al 2018, Arora et al 2021).

Soil studies: Among different spacing, organic carbon was found maximum under 6×3m (0.39%) and 7×3m (0.38) spacing. Soil pH (7.92 – 8.02) and electrical conductivity (1.7-2.1 dS/m) were also being lowered under different spacing as compared to sole crop (pH 8.01, EC 2.1 dS/m). However, the

Table 2. Growth performance of eucalypts tree under different spacing

Tree spacing (m)	Plant height (m)	GBH (cm)
6 × 3	5.68^{C}	24.3^{C}
7 × 3	7.12^{B}	33.15^{B}
8 × 3	7.97^{B}	34.45^{AB}
9 × 3	9.13^{A}	40.3^{A}
CD (0.05)	1.11	6.46

Table 1. Weather data during the study

Year	Temperature (°C)		Relative humidity (%)		Average wind speed (km/h)	Bright sun shine hours (h)	PAN evaporation (mm)	Rainfall (mm)	Rainy days
	Maximum	Minimum	M	E					
2019	30.4	17.0	83	48	4.6	5.9	4.0	474.8	31
2020	30.6	16.6	85	48	4.7	6.7	4.1	501.1	34
2021	31.1	17.4	84	49	4.6	6.3	4.0	795.0	34

magnitude of decrease was more in closer spacing. The available soil N, P and K increased significantly under different spacing of tree-based agroforestry system in comparison to control (sole crop). In the present study, the status of soil under study was EC >0.8 dS/m are saline soils; OC < 0.4% as low; N < 250 kg ha^{-1} as low; 10-20 kg ha^{-1} P as medium; K_2O 125-300 kg ha^{-1} as medium. The highest available soil N, P and K were recorded under 6×3m spacing (112.8, 12.4 and 228.20 q ha^{-1}, respectively). Soil pH and EC properties and did not differ significantly in different spacing and sole crop under study (Table 5).

Increase in soil OC under 6×3 and 7×3m tree spacing under eucalypts agroforestry system may be due to considerable addition of organic matter by more number of trees, which on decomposition release weak acids. Similar findings were observed by Bhupender (2021), Sirohi and Bhangrwa (2017) .

Benefit cost ratio: Maximum benefit cost ratio was estimated in raya under 9×3m tree spacing (1.67) and 8×3m (1.66) as compared to wheat (1.32), barley (1.13) under 9×3m tree spacing (Table 6). The tree spacing system 9×3m and 8×3m were found more economical as compared to other tree spacing system. This might be due to the reason of higher prices of grain of raya as compared to wheat and barley resulting in higher benefit cost ratio.

Table 4. Effect of different spacing of eucalypts based cropping system on yield of wheat, barley and raya

Tree spacing (m)	Grain yield (q ha^{-1})		
	Wheat	Barley	Raya
6 × 3	34.42^{C}	32.69^{C}	14.36^{C}
7 × 3	37.24^{C}	34.36^{C}	16.12^{C}
8 × 3	41.15^{B}	37.39^{B}	18.03^{B}
9 × 3	41.65^{B}	37.46^{B}	18.15^{B}
Control (Sole)	52.08^{A}	42.25^{A}	20.58^{A}
CD (0.05)	5.33	3.04	2.35

Table 6. Effect of different spacing of eucalypts based cropping system on benefit cost ratio in wheat, barley and raya

Tree spacing (m)	Wheat	Barley	Raya
6×3	1.09	0.98	1.32
7×3	1.18	1.03	1.49
8×3	1.30	1.12	1.66
9×3	1.32	1.13	1.67
Control (Sole)	1.64	1.27	1.90

Excluding economics of eucalypts plantation

Table 3. Yield attributes of *Rabi* field crops under eucalypts based agroforestry system

Tree spacing (m)	Wheat		Barley		Raya			
	Number of grains/spike	Number of tillers m^{-2}	Number of grains /spike	No. of tillersm^{-2}	No. of primary branches	No. of secondary branches	Siliqua/ plant	No. of seeds/ siliqua
6×3	36.58^{D}	325.54^{C}	35.30^{D}	325.23^{C}	4.00^{C}	8.00^{C}	209.95^{C}	9.93^{D}
7×3	39.51^{C}	352.87^{B}	38.33^{C}	338.83^{B}	5.00^{B}	8.11^{BC}	214.72^{C}	11.18^{C}
8×3	41.00^{B}	367.66^{B}	39.50^{BC}	349.10^{A}	5.00^{B}	8.30^{B}	230.60^{B}	11.90^{B}
9×3	41.93^{B}	409.98^{A}	40.65^{B}	353.54^{A}	5.00^{B}	9.00^{A}	240.00^{B}	12.16^{B}
Control	44.62^{A}	410.00^{A}	42.80^{A}	357.50^{A}	5.25^{A}	9.00^{A}	263.00^{A}	13.10^{A}
CD (0.05)	1.86	24.99	1.86	12.19	0.14	0.23	12.5	0.6

Table 5. Soil chemical property of field under different spacing of eucalypts

Tree spacing (m)	pH	$EC_{1:2}$ (dSm^{-1})	OC (%)	Available nutrients (kg ha^{-1})		
				N	P	K_2O
6×3	7.94	1.7	0.39	112.8	12.4	228.2
7×3	7.92	1.9	0.38	111.4	12.3	219.5
8×3	8.01	2.1	0.32	105.7	10.2	216.6
9×3	8.02	2.0	0.30	104.8	10.1	212.4
Control (Sole)	8.01	2.1	0.30	102.4	9.8	215.1
Mean	7.98	1.96	0.34	107.4	10.9	218.4
CD (0.05)	NS	NS	0.05	5.3	1.4	NS

CONCULSION

Minimum grain yield reduction was observed in barley at eucalypts spacing 9×3m followed by 8×3m. However, mustard also found to be suitable under eucalypts based agroforestry system in 9×3m spacing. Maximum B:C ratio was obtained with eucalypts + mustard agroforestry system under two spacing 9 × 3 m followed by 8×3 m and 7×3 m.

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Response of Different Organics Spray on Yield and Biochemical Characteristics of Sapota Fruits

V.R. Zala, A.K. Pandey* and P.J. Jadhav[1], D.Y. Gamanbhai, Y.N. Tandel B.V. Lakhamanbhai[2] and Shivanand Koti

Department of Fruit Science, ASPEE College of Horticulture, Navsari Agricultural University and Forestry Navsari-396 450, India
[1]Division of Fruit Science, Indian Agricultural Research Institute, New Delhi-110 012, India
[2]Department of Horticulture, B. A. College of Agriculture, Anand Agricultural University, Anand-388 110, India
**E-mail: avnish.hort2010@gmail.com*

***Abstract*:** An experiment was conducted to study the effect of different organic spray on yield and bio-chemical attributes of sapota [*Manilkara achras* (Mill.) Fosberg] cv. Kalipatti. Significant variation was noticed among the different treatments for all the characters studied. Result revealed highest physical and yield parameters like fruit weight (83.60g), fruit length (6.15cm), fruit diameter (5.78cm), number of marketable fruits (2553), marketable fruit weight (170.30kg/tree), min. number of damage of fruits (54.25), min. damage fruit weight (4.0 kg), total fruit yield (174.30 kg/tree) recorded in T_9 Novel Organic Liquid Fertilizer 5% (5 spray) also recorded minimum PLW% (2^{nd}, 4^{th} and ripening stage (3.34%, 5.09%, 8.09% respectively), acidity (0.121%), spoilage (6.25%), while maximum fruit firmness (13.81 kg cm^{-2}), TSS (23.84^0 Brix), ascorbic acid (10.80 mg 100 g^{-1}), reducing sugar (11.28 %), total sugar (19.47 %) and total phenol (133.87 mg 100 g^{-1}) .

Keywords: Novel, SPD, PLW, Absolute, Cow-urine, *Panchgavya*

Sapota botanically known as *Manilkara achras* (Mill.) Fosberg belongs to family Sapotaceae. It is popularly known as "*chikoo*" and important fruit crop of the tropical region. It is native to Tropical America especially Southern Mexico or Central America. However, it is commercially cultivated in India, The Philippines, Sri Lanka, Mexico, Venezuela, Guatemala and other countries of Central America. It is not much known when it was first introduced into India but Gholwad village of Maharashtra state is credited to have the first plantation of sapota in 1898 (Chadha 1992) and it is spread to the other states like Karnataka, Gujarat, Andhra Pradesh, West Bengal, Maharashtra and Tamil Nadu and now it occupies a significant position among the fruit crops in India. However, South Gujarat, Coastal Maharashtra and Karnataka are the major areas where, it is extensively cultivated. India is considered to be the largest producer of sapota in the world. The area and production of sapota cultivation in India is 97 thousand ha and 11.76 lakh million tonnes, respectively with productivity of 12.1 MT ha^{-1}. In Gujarat, the area under sapota cultivation is 27.83 thousand ha with production of 3.1 lakh tonnes, with productivity of 11.0 MT ha^{-1}. While in south Gujarat, the area under sapota cultivation is 13.03 thousand ha with 1.58 lakh tonnes of annual production wherein its cultivation is concentrated mainly in Navsari, Valsad and Surat districts.

The foliar application of cow urine, *panchagavya* and Novel Organic Liquid Nutrients plays a vital role in improving the quality and comparatively more effective for rapid recovery of plants. The foliar feeding of fruit tree has gained much importance in recent years, as nutrients applied through soil are needed in higher quantity because some amount leaches down and some become unavailable to the plant due to complex soil reactions. The yield parameters like average fruit weight, number of fruits per tree and yield per tree are increased by the spray of micronutrients.

MATERIAL AND METHODS

The present investigation was undertaken during the years 2020-21 at Instructional farm, ASPEE College of Horticulture and Forestry, NAU, Navsari (Gujarat). Experiment out in SPD analysis (in CRD) was done taking season as subplot factor. The treatments comprises ten treatments such as T_1: Cow-urine 2% (3 sprays), T_2: Cow-urine 2% (4 sprays), T_3: Cow-urine 2% (5 sprays) T_4: *Panchgava* 3% (3 sprays), T_5: *Panchgava* 3% (4 sprays), T_5: *Panchgava 3*%(5 sprays), T_7: Novel 5% (3 spray), T_8: Novel 5% (4 spray), T_9: Novel 5% (5 spray) and T_{10}: Absolute control. The experiment was carried out on 32 years old trees. Observation recorded on following parameters (1) Physical and Yield parameters like fruit length (cm), fruit diameter (cm), fruit volume(cc), fruit weight(g), Number of marketable fruits per tree, Marketable fruits weight kg per tree, Number of damage fruits per tree, damage fruits weight kg per tree, total fruit yield kg per tree (2) physico-chemical attributes like PLW%, fruit firmness (kg cm^{-2}), TSS (°Brix), Acidity (%), Ascorbic acid (mg $100g^{-1}$), Reducing sugar (%), Total sugar (%) and Total phenol (mg $100g^{-1}$). The two season data were pooled and statically analysed.

3 Sprays	1^{st} March, 1^{st}May and 15^{th} October
4 Sprays	1^{st} March, 15^{th} March, 1^{st}May and 15^{th} October
5 Sprays	1^{st} March, 15^{th} March, 1^{st}May, 15^{th} May and 15^{th} October
Absolute Control	No spray

RESULT AND DISCUSSION

Physical and yield parameters: The data on different physical parameters characters are presented in Table 1. The data revealed that maximum fruit weight (83.60 g), fruit length (6.15 cm), fruit diameter (5.78 cm) and fruit volume (76.19cc) observed in T_9 (5 Sprays of Novel Organic Liquid Nutrients @ 5%) whereas, the lowest fruit weight (69.49 g), fruit length (4.55 cm), fruit diameter (4.54 cm) and fruit volume (60.75cc) was noted in T_{10} (absolute control). Similar findings were reported by (Parmar 2016, Bhatt et al 2012, Gurjar et al 2017). The variation in fruit weight, length, diameters and volume may be due to the Zn and Fe nutrition received by the plants via foliar application of 5 Sprays of Novel Organic Liquid Nutrients @ 5% which play vital role to promote starch formation and suitable cell enlargement cell division in plants, respectively thus, the cumulative effect of micronutrients Fe and Zn might have proved beneficial for fruit growth (Nehete et al 2011).

The data on different yield parameters characters are presented in Table 1. The data revealed that maximum number of marketable fruits per tree (2553), marketable fruits weight per tree (170.30 kg tree-1), number of damage fruits (54.25) and min. damage fruits weight per tree (4.0 kg tree^{-1}) and fruit yield per tree (174.30 kg tree^{-1}) was observed in T_9(5 Sprays of Novel Organic Liquid Nutrients @ 5 %) whereas, the lowest number of marketable fruits per tree (2104.50), marketable fruits weight per tree (124.81 kg tree^{-1}), number of damage fruits (67.0) and damage fruits weight per tree (5.17 kg tree^{-1}) and fruit yield per tree (130 kg tree^{-1}) was noted in T_{10} (absolute control).It might be due to Novel Organic Liquid Nutrients contain zinc which plays a vital role to promote carbonic anhydrase is a metalloenzyme that requires Zn as a cofactor and is involved in diverse biological processes including pH regulation, CO_2 transfer, ionic exchange, respiration, CO_2 photosynthetic fixation and stomatal closure. Starch formation and iron are required for suitable cell enlargement and cell division in plants. Similar findings were reported by (Parmar et al 2017).

Physico-chemical attributes: The physico-chemical attributes characters viz., minimum acidity (0.121 %), spoilage (6.25 %) and PLW % at 2nd day, 4th day, and at ripening stage respectively recorded (3.34 %, 5.09 % and 8.09 %), It might be due to Ca present in Novel Organic Liquid Fertilizer reduce the decaying and maintain the firmness of fruit and minimizing the rate of respiration, protein break down and disease incidence (Gupta et al. 1980). while, maximum fruit firmness (13.81 kg cm^{-2}) and TSS (23.84°B) it might be due to the influence of nutrients on physiological processes such as respiration and photosynthesis, which enhanced the supply of dry matter, minerals and carbohydrates towards the developing fruits (Rani et al 2017). Ascorbic acid (10.80 mg 100 g^{-1}) content was increase it might be due to the catalytic activity of zinc and iron on its bio-synthesis from its precursor (glucose-6-phosphate) or inhibition of its conversation into dehydro ascorbic acid by enzyme ascorbic acid oxidation or both. Maximum total sugar (19.47 %) and reducing sugar (11.28 %) This might be due to zinc released from Novel Organic Liquid Nutrients promote hydrolysis of starch into sugars and acts as a catalyst in oxidation-reduction processes in plants. Sapota possesses climacteric phenomenon which triggers the dramatic changes in respiration. Potassium content in Novel Organic liquid Nutrients could be involved to enhance photosynthetic efficiency of the leaves and a possible increase in translocation of assimilates into the fruit. Sugar content might also be affected by respirational demand and adequate supply of nutrients, synthesis of invertase and starch splitting enzymes (Ram and Prasad 1988). Total phenol (133.87 mg 100 g^{-1}) It might be due to Novel Organic Liquid Nutrients regulate the DNA, RNA, protein synthesis, gene action and cell division in plants. All parameters were found maximum with 5 sprays of 5 % Novel Organic Liquid Nutrients. Whereas, the maximum acidity (0.13%), spoilage (16.56 %) and PLW % at 2nd day, 4th day, and at ripening stage respectively recorded

Table 1. Effect of different organic spray on physical and yield parameters of sapota cv. Kalipatti (pooled data of a winter and summer seasons)

Treatments	Fruit weight (g)	Fruit length (cm)	Fruit diameter (cm)	Fruit volume (cc)	Number of marketable fruit tree^{-1}	Marketable fruit weight kg tree^{-1}	Number of damage fruit tree^{-1}	damage fruit weight kg tree^{-1}	Total fruit yield kg tree^{-1}
T_1:Cow urine 2 % (3 spray)	71.73	4.75	4.68	63.05	2166.75	144.63	58.50	4.37	149.00
T_2: Cow urine 2 % (4 spray)	73.65	4.97	4.83	66.30	2220.75	148.23	53.00	3.69	151.93
T_3:Cow urine 2 % (5 spray)	79.38	5.75	5.53	71.51	2394.25	159.78	49.75	3.55	163.33
T_4: *Panchagavya* 3 % (3 spray)	75.19	5.43	4.99	66.99	2268.75	151.45	61.75	4.93	156.39
T_5: *Panchagavya* 3 % (4 spray)	76.72	5.55	5.30	69.45	2347.25	156.66	60.25	5.02	161.69
T_6: *Panchagavya* 3 % (5 spray)	82.13	5.90	5.62	74.93	2436.00	162.64	56.50	4.16	166.81
T_7: Novel Organic Liquid Nutrients 5 % (3 spray)	76.31	5.48	5.14	67.95	2298.50	153.44	64.75	4.84	158.29
T_8: Novel Organic Liquid Nutrients 5 % (4 spray)	82.31	6.05	5.69	75.24	2501.00	166.89	64.00	4.72	171.61
T_9: Novel Organic Liquid Nutrients 5 % (5 spray)	83.60	6.15	5.78	76.19	2553.00	170.30	54.25	4.00	174.30
T_{10}: Absolute control	69.49	4.55	4.54	60.75	2104.50	124.81	67.00	5.17	130.00
S. Em. ±	1.15	0.072	0.057	0.47	50.07	3.27	1.75	0.13	3.59
C.D. at 5 %	3.34	0.213	0.165	1.34	144.60	9.46	5.06	0.38	10.37
C.V. %	4.24	3.70	3.09	1.93	4.30	4.26	5.95	5.96	4.54

Table 2. Effect of different organic spray on physico-chemical parameters of sapota cv. Kalipatti (pooled data of a winter and summer seasons)

Treatments	PLW % at 2^{nd} day	PLW % at 4^{th} day	PLW % at ripening	Fruit firmness (kg cm^{-2})	TSS (˚B)	Acidity (%)	Ascorbic acid (mg 100 g^{-1})	Total sugar (%)	Reducing sugar (%)	Total phenol (mg 100 g^{-1})
T_1:Cow urine 2 % (3 spray)	4.69	6.55	8.93	10.56	19.62	0.127	9.96	16.49	10.37	121.17
T_2: Cow urine 2 % (4 spray)	4.45	6.50	8.79	11.42	19.68	0.128	10.30	16.95	10.52	123.39
T_3:Cow urine 2 % (5 spray)	3.79	5.63	8.49	13.34	21.21	0.125	10.50	18.28	10.97	129.02
T_4: *Panchagavya* 3 % (3 spray)	4.35	6.32	8.74	10.67	19.95	0.129	10.21	17.16	10.51	125.12
T_5: *Panchagavya* 3 % (4 spray)	3.94	6.17	8.57	11.58	20.19	0.127	10.39	17.21	10.63	128.23
T_6: *Panchagavya* 3 % (5 spray)	3.51	5.27	8.36	13.29	23.09	0.124	10.59	18.66	11.00	131.26
T_7: Novel Organic Liquid Nutrients 5 % (3 spray)	4.19	6.27	8.65	10.90	20.94	0.126	10.35	17.55	10.79	126.17
T_8: Novel Organic Liquid Nutrients 5 % (4 spray)	3.44	5.15	8.32	13.58	23.39	0.122	10.66	19.15	11.13	133.06
T_9: Novel Organic Liquid Nutrients 5 % (5 spray)	3.34	5.09	8.09	13.81	23.84	0.121	10.80	19.47	11.28	133.87
T_{10}: Absolute control	5.10	6.77	9.36	9.42	19.28	0.130	9.32	16.11	9.84	118.70
S. Em. ±	0.05	0.06	0.07	0.13	0.18	0.0025	0.066	0.19	0.28	0.68
C.D. at 5 %	0.14	0.18	0.22	0.38	0.53	0.0072	0.190	0.55	0.82	1.97
C.V. %	3.46	2.93	2.53	3.15	2.49	6.44	1.81	3.06	7.58	1.52

(5.10 %, 6.77 % and 9.36 %) while, minimum firmness (9.42 kg cm-2), TSS (19.28˚B), ascorbic acid (9.32 mg 100 g-1) total sugar (16.11 %) and reducing sugar (9.84 %),Total phenol (118.70 mg 100 g^{-1}) was observed in T_{10} (absolute control). Similar results were also observed by (Bhowmick and Banik 2011, Nehete et al 2011, Bhatt et al 2012, Chandra and Singh 2015).

CONCLUSION

Result of present study revealed that among the investigation of two seasons, it can be concluded that 5 foliar sprays of Novel Organic Liquid Nutrients at 5 %was most effective for improving fruit physical, yield, as well as reducing spoilage, maintaining firmness and extending the shelf life with optimal retention of fruit quality in sapota cv. Kalipatti.

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Ecotype Diversity Assessment of Autumn Olive (*Elaeagnus umbellata* Thunb.) in Himachal Pradesh

Shreya Chauhan, Anita Kumari*, Pratiksha Saini, Suman Kumar Jha[1] and G.B. Rawale

Department of Tree Improvement and Genetic Resources, Dr Yashwant Singh Parmar University of Horticulture and Forestry, Nauni, Solan-173 230, India
[1]Department of Forest Biology and Tree Improvement, College of Forestry Navsari Agricultural University, Gujarat-396 450, India
**E-mail: anita_78@ymail.com*

Abstract: *Elaeagnus umbellata* Thunb. commonly known as autumn olive (Elaeagnaceae) is a multipurpose actinorhizal shrub of the Western Himalayas. It is distinctly distributed at a range of about 1200 m to 2100 m, withstanding the eroded areas owing to renowned nitrogen fixing ability. The shrub was recorded at the three altitudinal ranges, at two locations from each range. *Elaeagnus* invasively outcompetes *Myrisine africana, Lantana camara, Coraria nepalensis* in chirpine forest and *Sambuca nigra, Ruscus, Indigofera, Lonicera* in kail forest. *E. umbellata* enhanced the levels of soil nitrogen in forest land, due to the presence of nodules on its roots that houses nitrogen fixing actinomycetes. This wild shrub is acclaimed for plantation to revamp soil fertility in a forest for hosting numerous herbs and shrubs in its vicinity.

Keywords: Wild species, Diversity, Distribution, Phytosociology, Nodules

Globally, forests cover nearly one third of the land area and contain over 80% of terrestrial biodiversity. Forests play a significant role in offering a multitudinal range of habitats for plants, animals, as well as micro-organisms (Jendresen and Rasmussen 2022). As a result of changing climatic patterns and altering human behavior, forests serve as a reserve for future needs (Devi et al 2018, Pichura et al 2017). More than 2/3rd (1.6 billion) people throughout the world, living in low and middle-income nations, reside within 5 kilometers of a forest periphery (Newton et al 2020). Further around 40% of the world's extremely poor population, rely on products and services that forest provide (IUFRO 2020). Forest aids in benefitting the humans by providing firewood, housing materials, high value forest products and food. The Himalayas are lavished with the rich flora, flourishing across vast range of habitats in distinct altitudinal ranges. Himalayan region, geographically covers 18% and accounts for more than 50% of India's forest cover. 30-40% species are endemic from the total, circumscribed under the Himalayan region many wild species of economic importance suffer from obscurity and of being ascertained. In natural or semi-natural habitats, the term "wild plant species" refers to those that develop spontaneously in self-maintaining populations and are unaffected by human activity. Wild resources in general are frequently disregarded and receive little or no appreciation. Lack of knowledge about their scope of use and significance, economic worth, global markets and a lack of quality standards are the major causes of this neglect.

Among the wild species, *Elaeagnus* is the largest shrub genus of Elaeagnaceae family (Paudel et al 2020). Around 90 species, have been reported in *Elaeagnus,* inclusive of which *E. umbellata* Thunb (Ahmad et al 2006)*, E. angustifolia* Thunb.*, E. multiflora* Thunb. and *E. pungens*, are used as medicinal plants (Paudel et al 2020). *Elaeagnus umbellata* Thunb. commonly known as autumn olive, autumn *elaeagnus* and Japanese silverberry (Gamba et al. 2020) belonging to family Elaeagnaceae is a multipurpose actinorhizal shrub of the Western Himalayas. *E. umbellata* Thunb. is widely distributed in Shimla, Solan, Sirmour, Chamba, Kangra, and Kinnaur districts in Himachal Pradesh at an altitudinal range of 1000-3000m. It is spiny-branched, deciduous shrub that grows upto 3.5 to 5.5m tall and wide along with light green foliage. The leaves are alternate with size ranging from 1 to 4 cm and are petiolated in lateral clusters on twigs (Ahmad et al 2005). *E. umbellata* Thunb. develops root nodules as a result of symbiotic association with actinomycetes (*Frankia*) in the soil that enables fixation and utilization of atmospheric nitrogen (Kim et al 1993). It is planted in eroded areas of mountainous zones to re-establish and develop vegetation cover. The fixation of atmospheric nitrogen by actinorhizal plants represented a contribution to global nitrogen cycle (Baker et al 1979).

There is a meager information on the diversity status of wild species. The appraisal of diversity and phytosociological studies constituting a forest could prove essential for exploration of certain unexplored wild species that may prove beneficial for several future purposes. Phytosociology is a study of vegetation of a plant community, certainly focused on assemblage of plants in forest stands. It emphasizes on characterizing vegetation types on basis of floristic composition of stands. There are various topographic and climatic factors that affect the plant species diversity (Malik and Bhatt 2015). The diversity of species in a forest differs throughout the altitudinal range depending upon the set of factors characterizing the habitat of particular species (Gairola et al 2011, Joshi et al 2021). In the hill slopes, the physiographic factors widely influence the plant microhabitats (Sharma et al 2010, Slobodkin and Sanders 1969). These studies frame an outlining for the wild species for establishing their status and distribution patterns.

MATERIAL AND METHODS

The present study was confined to three altitudinal ranges *i.e.,* <1200 m, 1200-1800 and >1800 m, containing *Elaeagnus umbellata* Thunb. in three districts of Himachal Pradesh (Fig. 1). From each altitudinal range two locations were selected comprising of five shrubs. At altitudinal range less than 1200 m, two sites *viz.,* Dilman (1157 m) (30º48´39"N, 77º08´36"E) and Kujji (1046 m) (30º49´02"N, 77º09´41"E) in Sirmaur district, between 1200-1800 m, two sites *viz.,* Kalaghat (1320 m) (30º51´57"N, 77º11´03"E) and Nauni (1252 m) (30º51´39"N, 77º10´09"E) in Solan district, at an elevation range greater than 1800 m sites *viz.,* Dhar (2141 m) (31º06´11"N, 77º41´37"E) and Shari (2010 m) (31º06´27"N, 77º41´11"E) in Shimla district were appraised.

The vegetation analysis was carried out by randomly placed 20 quadrats of 5×5 m for shrubs and 1×1 m for herbaceous growth in the six locations bearing shrubs along with one control site (*E. umbellata* absent). Quadrates were laid out randomly by species area curve method (Mishra 1968). The vegetation data was analyzed quantitatively for phytosociological parameters *viz.,* basal area, density and frequency (Curtis and McIntosh 1950). The Importance Value Index (IVI), (Philips 1959), species diversity indices and dominance indices were determined as per the methods outlined by Magurran (1988), Shannon-Wiener (1963) and Simpson (1949). The soil parameters (pH, electrical conductivity, N, P, K) were evaluated at three depths *i.e.,* 0-10 cm, 11-20 cm and 21-30 cm.

RESULTS AND DISCUSSION

Around 32 species of shrubs and 50 herbaceous species were recorded during the sampling of the three habitats. The species diversity observed in the chir pine and kail forest, was comparable with the previous studies (Sharma 2006, Gupta et al 2009, Attri et al 2017). Variations in the phytosociological attributes, among the different habitats in Himachal Pradesh, are primarily due to the changing environmental, geographic and edaphic conditions. *E. umbellata* dominated the three habitats, ranging at different altitudes (Table 1). Due to its invasive nature, outcompetes species *viz., Myrisine africana, Lantana camara, Coraria nepalensis, Zanthoxylum aratum, Bidens pilosa* and *Setaria glauca* in chir pine forest, *Sambuca nigra, Ruscus, Indigofera, Lonicera*, *Eunymus europaeus, Urena lobata, Viola* and *Liparis* in kail forest (Table 2). These species dominated in its absence. During the early phases of regeneration, it replaces the forest with invasive thickets and limits the light reaching forest ground (Chittka and Schurkens 2001). As being a vigorous individual with sufficient light in open grasslands habitat, it provides a large source of seeds to understory, where probability of successful recruitment increases with increased propagule pressure (Brym et al 2011).

The diversity indices (Shannon, Simpson, equitability and species richness) were highest in the habitat bearing *E. umbellata* (Table 3). The luxuriant richness of species was present at an altitudinal range below 1200 m, whereas at a higher altitudinal range greater than 1800 (Dhar and Shari), the species were scattered and equally distributed. Our findings were supported by earlier investigations that

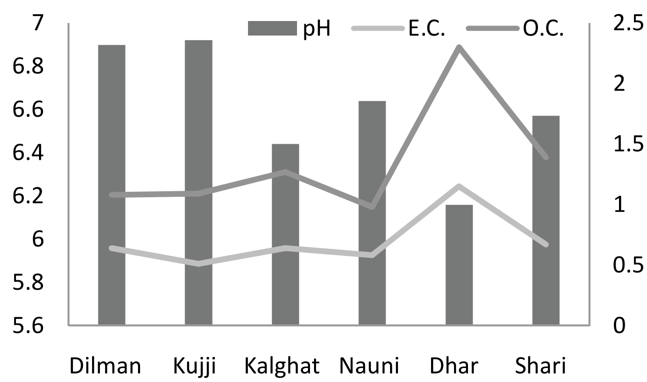

Fig. 1. Soil parameters (pH, EC, O.C.)

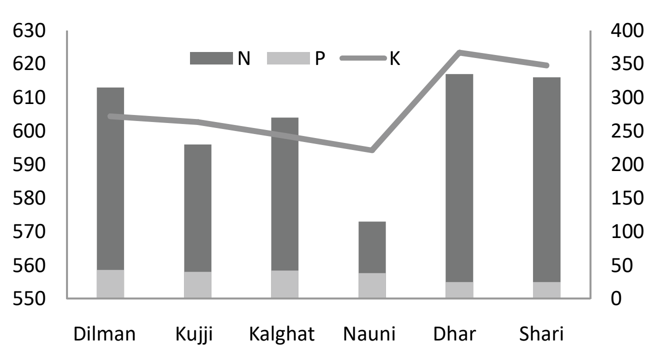

Fig. 2. Soil parameters (N, P, K)

demonstrated species richness increasing from the higher elevation towards the lower elevation (Kumar and Thakur 2008, Sharma et al 2009, Raturi 2012, Singh 2013).

E. umbellata alters the soil chemistry owing to the presence of root nodules that house nitrogen fixing actinomycetes (Baer et al 2006). The nodules favor pH above 5 and no nodulation is observed below pH 4 (Mohebbi and Mahler 1989). Electrical conductivity in the soils depicted significantly higher values ($P<0.05$) at a range of >1800 m, it is linked with the high nutrient pumping and deposition of organic matter on soil surface. *E. umbellata* Thunb. is a source of nitrate overload in both terrestrial and aquatic ecosystems, thus increasing levels of nitrogen in forest soils (Goldstein et al 2009) (Fig. 1). Significantly higher nitrogen levels ($P<0.05$) were observed at a range of >1800 m (Fig. 2). A single plant of *E. umbellata* Thunb. is sufficient to induce a change in composition of soil microbial communities. It releases higher than normal nitrate in surrounding soil environment that is a form of nitrogen used by plants for their growth (Brym et al 2011).

Table 1. Shrub species diversity in all the six locations and their IVI values

Species	Dilman		Kujji		Kalaghat		Nauni		Dhar		Shari	
	EP	EA	EP	EA	EP	EA	EP	EA	EP	EA	EP	EA
Berberis aristata	-		-	-	-	-	-	-	102.59	72.43	27.54	44.87
Berberis lycium	38.67	40.79	43.59	-	70.07	45.95	65.39	84.14	-	-	-	-
Carissa spinarum	24.48	20.22	21.48	33.49	12.50	-	-	-	-	-	-	-
Cassia floribunda	-	15.34	17.90	28.12	-	-	-	-	-	-	-	-
Coriaria nepalensis	-	16.43	-	-	-	-	-	-	-	-	-	-
Caryopteris wallichiana	15.22	20.93	10.16		16.44	-	-	-	-	-	-	-
Daphne spp.	-	-	-	-	-	-	-	-	-	28.59	15.40	19.66
Elaeagnus umbellata	76.87	-	63.22	-	41.96	-	74.98	-	135.91	-	59.06	-
Euonymus europaeus	-	-	-	-	-	-	-	-	-	94.91	-	-
Hypericum oblongifolium	15.23	-	31.68	-	13.81	26.67	29.50	15.21	-	-	-	-
Indigofera spp.	-	-	-	-	-	-	-	-	-	35.24	24.07	-
Lantana camara	-	40.29	-	60.30	-	34.90	-	31.72	-	-	-	-
Lonicera angustifolia	-	-	-	-	-	-	-	-	-	88.43	19.13	29.32
Lonicera interrupta	-	-	-	-	-	-	-	-	-	-	8.64	-
Murraya koenigii	-	-	-	-	-	-	24.71	-	-	-	-	-
Myrsine africana	-	46.33	-	61.90	-	-	29.33	-	-	-	-	-
Peritoma arborea	-	-	-	-	-	-	9.89	-	-	-	-	-
Plectranthus rugosus	-	-	-	-	-	-		-	-	-	18.60	-
Prinsepia utilis	-	-	18.50	-	18.26	22.36	31.36	50.06	43.12	48.31	34.60	30.53
Pseudocaryopteris bicolor	-	-	-	-	27.39	15.87	5.99	-	-	-	-	-
Rhamnus spp.	-	-	-	-	20.56	10.68	-	-	-	-	-	-
Rosa moschata	55.23	44.34	24.49	43.29	45.22	50.32	35.92	58.73	23.33	28.39	26.64	39.37
Rosa mulliganii	-	-	7.77	-	-	-		-	75.02	33.28	-	-
Rubus leucodermis	-	-	-	-	-	-	-	-	62.23	-	-	-
Rubus ellipticus	47.92	33.02	43.58	53.22	33.46	53.43	32.15	33.56	-	72.68	29.31	41.41
Rubus niveus	-	-	-	-	-	-	-	-	52.05	-	-	-
Ruscus aculeatus	-	-	-	-	-	-	-	-	30.91	-	-	30.66
Sambuca nigra	-	-	-	-	-	-	-	-	-	38.10	-	15.38
Sarcococca saligna	-	-	-	-	-	-	-	-	-	-	16.26	-
Symphoricarpos orbiculatus	-	-	-	-	-	-	-	-	-	79.25	-	-
Viburnum prunifolium	-	-	-	-	-	-	-	-	13.33	-	20.75	38.27
Zanthoxylum armatum	26.38	22.31	17.65	19.68	-	-	-	26.58	-	-	-	-

Here EP represents *E. umbellata* present, EA represents *E. umbellata* absent

Table 2. Herb species diversity in all the six locations and their IVI

Species	Dilman		Kujji		Kalaghat		Nauni		Dhar		Shari	
	EP	EA	EP	EA	EP	EA	EP	EA	EP	EA	EP	EA
Alternanthera pungens	-	-	-	-	-	-	-	-	-	-	27.51	-
Apluda mutica	14.09	20.56	11.94	11.39	-	13.11	-	16.49	-	-	-	-
Ageratum conyzoides	-	-	-	-	-	-	-	-	15.47	-	-	-
Artemisia	44.68	19.79	37.38	-	29.11	73.94	60.05	24.40	-	-	-	-
Arundinella	9.85	-	-	-	-	-	-	-	-	-	-	-
Athyrium attenuatum	5.64	6.09	10.48	27.43	-	-	-	-	21.91	18.08	-	-
Atriplex glauca	-	-	-	-	-	-	-	-	32.19	-	-	-
Avena	-	-	-	-	-	-	6.68	-	-	-	-	-
Barleria cristata	34.79	6.89	42.73	-	14.47	-	-	-	-	-	-	
Bidens Pilosa	-	58.61	-	32.19	77.72	51.19	44.20	82.28	-	-	-	-
Brixly oxtongue	17.09	29.29	+21.91	108.99	16.85	6.41	29.80	-	12.22	19.31	-	-
Cenchrus	-	-	-	-	-	-	-	-	13.51	-	-	-
Chrysopogon montanus	5.14	13.72	13.44	-	7.89	3.40	5.12	-	-	-	-	-
Clematis	-	-	-	16.04	10.63	4.88	5.03	6.30	-	-	-	17.92
Climber	-	-	-	-		-	-	-	-	-	7.63	
Convolvulus arvensis	-	-	-	-	-	-	-	-	10.40	-	-	-Contd.
Crepidium acuminatum	-	-	-	-	-	-	-	-	11.16	40.07	-	-
Crepis tectorum	-	-	-	-	-	-	-	-	-	-	12.01	-
Cirsium horridulum	-	-	-	-	10.21	-	9.17	-	-	-	-	-
Deutzia gracilis	-	-	-		25.22	-	-	-	-	-	-	-
Eleusine indica	-	-	39.81	21.46	-	-	-	-	19.10	9.38	37.52	39.28
Erigeron canadensis	61.31	18.85	25.02	19.88	-	33.45	25.33	49.69	23.17	-	16.67	62.83
Euphorbia hirta	-	-	-	-	-	-	-	-	-	18.31	-	-
Galinsoga parviflora	14.74	-	21.78	-	9.18	-	-	-	-	-	-	-
Habenaria intermedia	-	-	-	-	-	-	-	-	-	46.80	-	-
Hedera helix	-	-	-	-	-	-	-	-	11.24	26.51	8.61	-
Heteropogon contortus	14.62	8.23	4.23	-	-	17.05	3.34	25.52	-	-	-	8.35
Jasminum humile	-	-	-	-	-	-	14.40	-	-	-	-	-
Justicia simplex	-	-	-	-	8.31	-	-	-	-	-	-	-
Koeleria macrantha	-	-	-	-	10.65	-	6.71	-	-	-	-	-
Liparis nervosa	-	-	-	-	-	-	-	-	-	36.45	-	-
Oxalis corniculate	-	-	-	-	-	-	-	-	5.62	-	5.39	-
Panicum maximum	-	-	-	-	8.41	6.71	-	-	-	-	-	-
Parthenium hysterophorus	-	-	-	-	-	43.77	-	-	-	-	-	-
Paspalum notatum	15.48	-	-	-	-	-	3.35	-	-	-	-	-
Pennisetum purpureum	-	-	-	-	-	-	-	-	8.52	-	-	-
Pteris cretica	-	-	-	-	-	-	-	-	-	30.57	48.24	18.36
Rhynchosia minima	-	-	-	-	-	-	-	-	-	-	14.94	-
Rumex nepalensis	-	-	-	-	-	-	9.31	34.45	17.49	-	18.95	35.57
Saccharum filifolium	-	-	-	-	8.16	3.65	-	-	-	-	-	-
Setaria glauca	-	12.84	-	16.04	18.97	13.41	6.69	-	-	-	-	-
Smilax aspera	21.63	17.99	16.17	-	12.85	17.01	18.43	8.99	-	-	10.07	16.44
Spartina patens	-	-	-	-	-	-	-	-	-	-	13.77	-
Thalictrum foliolosum	10.56	-	12.16	-	4.69	-	11.65	-	-	-	-	-
Themeda anathera	12.69	42.83	-	-	11.61	10.34	24.36	18.90	-	-	-	-
Trifolium repens	-	-	15.67	21.24	-	-	18.11	27.87	23.20	26.34	26.84	35.16
Urena lobata	-	-	-	-	-	-	-	-	-	8.15	16.10	47.46
Urtica dioca	17.67	44.31	27.28	25.33	-	-	-	-	28.28	12.09	20.83	-
Vicia hirsuta	-	-	-	-	14.08	6.08	+	-	-	-	-	-
Viola	-	-	-	-	-	-	-	-	-	7.93	14.89	18.63

Here EP represents *E. umbellata* present, EA represents *E. umbellata* absent

Table 3. Vegetation indices in all the six locations

Population	Plant category	Simpson index (S)		Shannon Weiner (H)		Equitability index (E)		Species richness index	
		EP	EA	EP	EA	EP	EA	EP	EA
Dilman	Shrubs	0.902	0.85	2.32	2.009	1.008	0.9663	2.53	1.68
	Herbs	0.930	0.934	2.64	2.757	1.003	0.9906	3.12	3.45
Kujji	Shrubs	0.856	0.885	1.939	2.27	0.996	0.947	1.38	2.03
	Herbs	0.924	0.92	2.31	2.6	1.006	0.988	1.93	2.60
Kalaghat	Shrubs	0.890	0.8693	2.19	2.21	1.001	0.922	2.06	2.26
	Herbs	0.93	0.946	2.65	2.86	1.005	1.012	3.05	3.69
Nauni	Shrubs	0.811	0.864	1.802	2.084	0.9258	0.9485	1.49	1.87
	Herbs	0.904	0.9225	2.399	2.68	0.9653	0.9459	2.51	3.71
Dhar	Shrubs	0.900	0.882	2.358	2.22	0.9835	0.9658	2.19	1.97
	Herbs	0.900	0.923	2.38	2.61	0.96	0.9892	2.54	2.70
Shari	Shrubs	0.8828	0.882	2.106	2.242	1.013	0.9351	1.74	2.11
	Herbs	0.9185	0.865	2.57	2.052	0.9768	0.9868	1.57	2.91

Here EP represents *E. umbellata* present, EA represents *E. umbellata* absent

CONCLUSION

Several worthwhile wild species face obscurities due to dearth of study and anonymity. The present study has illustrated the significance of *E. umbellata* in distinct habitats, at three altitudinal gradients in north-west Himalayas. The shrub occupies various forests, hosting numerous shrubs, herbaceous vegetation and soil microbes underneath its canopy. Juxtaposing habitats bearing *Elaeagnus* with the non-bearing ones, edaphic and phytosociological attributes were more inclined towards the *Elaeagnus* bearing habitats. *E. umbellata* Thunb. is renowned for its soil binding and nitrogen fixing characteristics simultaneously. Therefore, we recommend *E. umbellata* for plantation in degraded landscapes and reforesting mountainous regions. The wild shrub provides anchorage to multitudinal species, that are unable to establish themselves in stress conditions in a particular habitat.

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Standardization of Macro-Propagation Protocol of *Toona ciliata* M. Roem under Punjab Conditions

Mohit Kundal and Aman Deep Ranga[1]

College of Forestry, Department of Tree Improvement and Genetic Resources and [1]College of Horticulture, Department of Vegetable Science, Dr Yashwant Singh Parmar University of Horticulture and Forestry, Nauni, Solan-173 230, India
E-mail: mohit-fnr@pau.edu

***Abstract*:** *Toona* species are amongst the most valuable timbers of tropics and the backbone of forest based industries in many countries throughout the world. The study was conducted during 2018 to standardize the macropropagation protocol of *Toona ciliata* under Punjab conditions. Two types of cuttings (stem and coppiced) were grown under two different growing media *viz*., vermiculite and Soil: Sand: FYM (1:1:1) treated with 4 treatments (500, 1000, 1500 and 2000 ppm each of IBA and NAA including control and were evaluated for 6 characters under two different planting conditions *viz*., mist chamber and greenhouse. The experiment was conducted in three different growing seasons viz., mid-March, mid-July and mid-November. Season mid-March provide better performance followed by season mid-July whereas season mid-November was poorest. Vermiculite media gave better results than Soil: Sand: FYM media, IBA than NAA, mist chamber conditions than greenhouse conditions and stem cuttings gave better results than coppiced cuttings. Stem cuttings planted in mid-March season, treated with IBA 500 ppm, planted in vermiculite media and grown under mist chamber conditions came out to be the best combination to get significant and fruitful results.

***Keywords*:** IBA, Macropropagation, NAA, *Toona ciliata*

Vegetative propagation in forest trees is emerging as a strong alternative to the seed propagation method and now employed for operational planting in many forest species. It aims to reproduce 'true to type' progeny identical to the parent source while maintaining selected elite trees indefinitely without pre-developing true-breeding stock through inbreeding and hence enables the tree breeder to shorten a breeding program considerably. It speeds up the reproductive cycle for accelerated breeding and testing while producing more stable improved material. Vegetative propagation has the potential to capture more genetic gain to obtain greater uniformity of the tree crops than through seed regeneration as both additive and non-additive traits are readily accessible and can be exploited as compared to sexual propagation where only additive traits are accessible. Moreover, it is a good method to evaluate the genotypes and their GxE interactions. *T. ciliata* is a fast growing large deciduous tree attaining a height of upto 20-30 m with a clean bole of up to 9-12 m height. It is native to Australia and has been distributed naturally in India, Burma, Laos, Pakistan, Thailand, Malaysia, Indonesia, and China (Haines et al 2016). This is a least concerned species found majorly in hills of Tamil Nadu, Andhra Pradesh and Odisha upto an elevation of 900 m (Rao 2016). In the western sub-Himalayan tract recorded in moist localities, in sheltered ravines, along streams and even in swamp forest, while in the Western Ghats, mostly in wet evergreen forests; with scattered occurrences in moist deciduous forests (Khare 2007). It is also a characteristic tree of Eastern alluvial secondary semi-evergreen forests in Assam and in Punjab *T. ciliata* is distributed in Kandi region which mainly comprises of dry deciduous scrub forest. *T. ciliata* is moderate light demander, however, the young plants require some side protection from direct sun (Orwa et al 2009). Although it favours moist localities, if tended and worked in the early stages, it can be cultivated under dry localities receiving as low as 750 mm rainfall with 2-6 dry months with a maximum temperature as high as 49°C.

Toona species are amongst the most valuable timbers of tropics and in fact the backbone of forest-based industries in many countries throughout the world owing to its high quality timber and for the ease with which they can be grown in plantations (Bufalino 2012). Toon possesses important economic characteristics including a relatively short 15-year cycle; straight clean bole, good yields, and high value in the internal and external markets (Murakami 2008). The colour of the heartwood ranges from pinkish to dark reddish brown with a good natural texture whereas sapwood is pale yellow to brown by which it is easily demarcated from heartwood. The timber is rated as moderately durable i.e. it is moderately resistant to shoot borer attack with moderate weight, strength and hardness. The versatile timber is used for building houses and ships, furniture, musical instruments, carvings, and numerous other uses. *T. ciliata* is also planted avenue tree and firebreaks along roadsides in north India and are often looped for fodder. The technique of propagation through vegetative means have been standardized for many species, yet limited work on *T. ciliata* under Punjab conditions has been undertaken. In this experiment, macropropagation technique has been standardized through scientific improvement for large scale production of planting materials using low-cost conventional methods.

MATERIAL AND METHODS

This experiment was conducted during 2019 at College of Horticulture and Forestry, Punjab Agricultural University, Ludhiana, Punjab, India to standardize the macropropagation protocol of *T. ciliata* under Punjab conditions. Best planting season, best planting conditions, best type of planting material for cuttings (Fig. 1), best growing media and best growth regulator for significant macropropagation of *T. ciliata* were assessed under this study.

Methodology: Mid-March (season 1), mid-July (season 2) and mid-November (season 3) were chosen as the three planting to assess the best planting season for macropropagation. Two types of cutting *viz.*, juvenile stem cuttings from one year old plants along with one year old coppicing shoots from coppiced tree were used in the study. The cuttings having dimension of 8-10 cm length and 0.4-0.8 cm diameter having single node were selected. These cuttings were pre-treated with Bavistin (3g l^{-1} water) for 10 minutes and washed thrice with running water for protection against fungal diseases. Two media *viz.*, vermiculite and Soil: Sand: FYM (1:1:1) along with two auxins *viz.* IBA and NAA with 500, 1000, 1500 and 2000 ppm concentrations each along with a control each were used for planting cuttings.

The experiment was conducted in hyco-trays in two planting conditions *viz*., mist chamber and greenhouse. The temperature and humidity range of the mist chamber ranged from 29-32°C and 65-70%, respectively. In greenhouse, the temperature and humidity ranged from 17-30°C and 45-50% in mid-March, 27-36°C and 55-60% in Mid-July and 17-31°C and 30-35% in mid-November, respectively. The water sprinkler timings were set for 30 seconds at an interval of five minutes in mist chamber whereas, in greenhouse, water sprinkling was carried out manually by rose can at an interval of an hour from 0800 hrs to 1700 hrs. Hyco-trays were also washed with Bavistin (3g l^{-1}) to reduce the further chances of fungal attack. These trays were tagged and respective growing media were filled in these trays. A horizontal cut at basal end and slanting cut at apical end of the cuttings were made and the basal end of each cutting was dipped in respective auxin concentrations for a quick dip of 1 minute. Twenty cuttings each of one year old seedling as well as coppiced shoots were used per growth regulator per growing media per planting location and were replicated thrice. These cuttings were inserted 6-7 cm deep in the trays keeping in view that the buds are outside the media.

Observations were recorded for sprouting per cent (%), survival per cent (%), rooting per cent (%), average root length (cm), collar diameter (cm) and time taken for sprouting (days) at an interval of one month. Mean data of the observations were estimated and their respected critical differences (CD) and coefficient of variation (CV 0.05%) were calculated using (SPSS 2006).

RESULTS AND DISCUSSION

Both the growing media, vermiculite (under mist chamber conditions than greenhouse conditions) gave better results for all the characters observed as compared to Soil: Sand: FYM media with similar planting conditions for both stem cuttings and coppicing shoots in season 1 (mid-march) (Table 1, 2). The overall maximum sprouting percentage of 71.67 from stem cuttings and 23.33 from coppicing shoots was observed in cuttings treated with IBA 500 ppm (Fig. 2 & 3). Similarly, maximum rooting and survival of 66.67 and 25% was observed from stem cuttings whereas it was 11.67% each for cuttings from coppicing shoots. Moreover, maximum root length and collar diameter of 16.83 and 27 mm was observed from stem cuttings whereas it was 14.23 and 23 mm for cuttings from coppicing shoots. Additionally minimum days taken to sprout were 9 in case of stem cuttings whereas cuttings from coppicing shoots took 12 days to sprout. The season 1 (mid-March), both the growing media, vermiculite (under mist chamber conditions than greenhouse conditions) again gave better results for all the characters observed as compared to Soil: Sand: FYM media with similar planting conditions for both stem cuttings and coppicing shoots for season 2 (mid-July). The overall maximum sprouting of 36.67% from stem cuttings and 23.33% from coppicing shoots was observed in cuttings treated with IBA 500 ppm. Similarly, maximum rooting and survival percentage of 23.33 and 10% was observed from stem cuttings and coppicing shoots. Moreover, maximum root length and collar diameter of 10.32 cm and 23mm was observed from stem cuttings, whereas, it was 6.37 cm and 17 mm for cuttings from coppicing shoots. Additionally minimum days taken to sprout were 10 in case of stem cuttings whereas cuttings from coppicing shoots took 12 days to sprout (Table 3, 4). The season 3 was poorest among all the seasons but similar fashion for all the growth parameters. The stem cuttings from

Table 1. Effect of stem cuttings planted in mid-March season on various parameters

Stem cuttings	Sprouting per cent				Rooting per cent				Survival per cent				Root length				Collar diameter				Days taken to sprout			
	Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2	
	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G
Control	58.33	51.67	30.00	33.33	55.00	45.00	43.33	30.00	53.33	36.67	38.33	18.33	10.03	7.49	6.42	4.67	23.00	21.00	23.00	17.00	10.00	12.00	12.00	14.00
IBA 500 ppm	71.67	56.67	48.33	43.33	66.67	48.33	51.67	38.33	63.33	43.33	48.33	36.67	16.83	10.93	9.40	6.31	27.00	26.00	30.00	24.00	9.00	10.00	12.00	13.00
IBA 1000 ppm	55.00	45.00	25.00	35.00	51.67	38.33	30.00	35.00	50.00	30.00	28.33	25.00	10.00	8.48	3.96	3.74	13.00	17.00	23.00	22.00	10.00	12.00	12.00	14.00
IBA 1500 ppm	51.67	33.33	26.67	23.33	45.00	31.67	30.00	23.33	41.67	28.33	28.33	11.67	11.22	6.25	3.42	3.42	20.00	18.00	20.00	18.00	10.00	12.00	13.00	15.00
IBA 2000 ppm	50.00	21.67	23.33	23.33	41.67	18.33	21.67	21.67	38.33	18.33	18.33	8.33	7.77	4.13	6.75	2.73	23.00	15.00	17.00	16.00	10.00	13.00	12.00	15.00
NAA 500 ppm	55.00	20.00	36.67	20.00	48.33	16.67	31.67	11.67	43.33	8.33	28.33	5.00	12.15	3.74	7.71	2.10	10.00	16.00	33.00	17.00	9.00	13.00	13.00	15.00
NAA 1000 ppm	31.67	18.33	28.33	11.67	25.00	16.67	25.00	8.33	21.67	3.33	23.33	0.00	9.75	3.22	3.86	0.00	27.00	14.00	23.00	0.00	10.00	13.00	13.00	16.00
NAA 1500 ppm	26.67	15.00	18.33	10.00	21.67	8.33	13.33	0.00	16.67	0.00	10.00	0.00	14.33	0.00	5.67	0.00	17.00	0.00	20.00	0.00	10.00	13.00	14.00	16.00
NAA 2000 ppm	28.33	11.67	10.00	3.33	23.33	1.67	6.67	0.00	18.33	0.00	6.67	0.00	10.79	0.00	3.10	0.00	20.00	0.00	17.00	0.00	10.00	14.00	15.00	16.00
SEM	4.99	1.63	1.39	1.27	1.69	1.58	1.43	1.27	1.58	1.26	1.63	0.94	0.16	0.07	0.09	0.04	0.38	0.22	0.28	0.21	0.14	0.23	0.19	1.00
CD (0.5%)	14.96	4.88	4.16	3.82	5.07	4.75	4.29	3.82	4.75	3.77	4.89	2.82	0.47	0.22	0.27	0.12	1.14	0.67	0.83	0.63	0.41	0.68	0.57	3.01
CV%	18.95	9.11	8.78	9.68	6.78	10.89	8.80	11.67	7.12	11.65	11.06	13.77	2.35	2.57	2.83	2.69	3.31	2.76	2.09	2.88	2.43	3.17	2.56	24.02

Media 1: Vermiculite, Media 2: Soil: Sand: FYM (1:1:1), M: Mist chamber conditions, G: Greenhouse conditions

Table 2. Effect of coppiced shoot cuttings planted in mid-March season on various parameters

Coppiced shoots	Sprouting per cent				Rooting per cent				Survival per cent				Root length				Collar diameter				Days taken to sprout			
	Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2	
	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G
Control	11.67	3.33	10.00	1.67	13.33	1.67	8.33	0.00	11.67	1.67	6.67	0.00	10.82	7.62	7.62	2.37	20.00	14.00	16.00	13.00	13.00	14.00	14.00	15.00
IBA 500 ppm	23.33	11.67	16.67	8.33	25.00	11.67	20.00	10.00	23.33	11.67	18.33	8.33	14.23	9.15	12.47	5.41	23.00	17.00	20.00	18.00	12.00	12.00	13.00	13.00
IBA 1000 ppm	20.00	8.33	3.33	5.00	13.33	5.00	3.33	3.33	15.00	3.33	3.33	5.00	7.42	6.11	6.62	4.33	10.00	11.00	19.00	13.00	14.00	14.00	14.00	15.00
IBA 1500 ppm	6.67	0.00	0.00	0.00	8.33	0.00	0.00	0.00	8.33	0.00	0.00	0.00	6.61	0.00	4.83	0.00	17.00	0.00	19.00	0.00	14.00	0.00	15.00	0.00
IBA 2000 ppm	5.00	0.00	0.00	0.00	5.00	0.00	0.00	0.00	3.33	0.00	0.00	0.00	4.72	0.00	4.44	0.00	15.00	0.00	14.00	0.00	15.00	0.00	16.00	0.00
NAA 500 ppm	18.33	6.67	5.00	3.33	5.00	1.67	8.33	3.33	10.00	3.33	8.33	3.33	7.33	3.16	3.15	1.82	16.00	13.00	16.00	12.00	15.00	16.00	17.00	18.00
NAA 1000 ppm	11.67	0.00	1.67	0.00	3.33	0.00	3.33	0.00	5.00	0.00	5.00	0.00	4.17	0.00	2.93	0.00	14.00	0.00	15.00	0.00	16.00	0.00	17.00	0.00
NAA 1500 ppm	5.00	0.00	0.00	0.00	1.67	0.00	0.00	0.00	3.33	0.00	0.00	0.00	2.59	0.00	0.00	0.00	13.00	0.00	0.00	0.00	17.00	0.00	0.00	0.00
NAA 2000 ppm	3.33	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
SEM	1.43	1.16	1.00	0.88	1.40	1.00	1.16	0.81	1.30	1.00	1.06	0.81	0.16	0.04	0.09	0.06	0.21	0.16	0.20	0.15	0.19	0.14	0.15	0.18
CD (0.5%)	4.29	3.48	3.00	2.63	4.20	3.00	3.48	2.43	3.91	3.00	3.17	2.43	0.48	0.12	0.26	0.17	0.64	0.47	0.61	0.45	0.57	0.43	0.45	0.55
CV%	20.58	57.21	42.58	74.69	29.15	72.06	41.80	75.75	24.87	66.91	39.57	75.75	4.33	2.33	3.20	6.16	2.59	4.47	2.67	4.14	2.58	4.04	2.21	4.72

Media 1: Vermiculite, Media 2: Soil: Sand: FYM (1:1:1), M: Mist chamber conditions, G: Greenhouse conditions

Table 3. Effect of stem cuttings planted in mid-July season on various parameters

Stem cuttings	Sprouting per cent				Rooting per cent				Survival per cent				Root length				Collar diameter				Days taken to sprout			
	Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2	
	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G
Control	16.67	13.33	11.67	8.33	8.33	3.33	11.67	0.00	8.33	3.33	11.67	0.00	6.73	3.41	4.39	0.00	20.00	13.00	16.00	0.00	11.00	12.00	13.00	0.00
IBA 500 ppm	36.67	21.67	25.00	15.00	23.33	18.33	20.00	10.00	23.33	18.33	20.00	8.33	10.32	7.63	9.28	9.28	23.00	16.00	20.00	20.00	10.00	10.00	12.00	12.00
IBA 1000 ppm	16.67	18.33	8.33	8.33	13.33	8.33	8.33	3.33	13.33	8.33	25.00	3.33	7.64	4.28	4.53	4.53	10.00	11.00	19.00	19.00	11.00	12.00	13.00	13.00
IBA 1500 ppm	11.67	3.33	0.00	0.00	11.67	5.00	0.00	0.00	11.67	3.33	0.00	0.00	6.23	2.19	0.00	0.00	17.00	10.00	19.00	0.00	0.00	0.00	0.00	0.00
IBA 2000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	4.17	0.00	0.00	0.00	15.00	0.00	14.00	0.00	0.00	0.00	0.00	0.00
NAA 500 ppm	11.67	13.33	11.67	3.33	8.33	3.33	8.33	0.00	8.33	1.67	6.67	0.00	3.36	3.13	6.15	0.00	16.00	13.00	16.00	0.00	11.00	13.00	13.00	0.00
NAA 1000 ppm	10.00	3.33	5.00	0.00	3.33	0.00	1.67	0.00	3.33	0.00	1.67	0.00	2.62	0.00	3.97	0.00	14.00	0.00	15.00	0.00	11.00	0.00	14.00	0.00
NAA 1500 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	13.00	0.00	0.00	0.00	10.00	0.00	0.00	0.00
NAA 2000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	10.00	0.00	0.00	0.00
SEM	1.32	1.55	1.00	1.02	1.43	1.06	1.13	0.56	1.39	1.13	1.18	0.81	0.11	0.02	0.01	0.03	0.30	0.136	0.24	0.12	0.11	0.13	0.13	0.13
CD (0.5%)	3.95	4.64	3.00	3.06	4.29	3.17	3.38	1.67	4.16	3.38	3.53	2.43	0.34	0.07	0.04	0.08	0.90	0.41	0.73	0.34	0.32	0.39	0.39	0.38
CV%	19.56	32.88	24.65	45.46	32.62	43.01	35.18	64.95	31.68	50.25	36.74	108.21	4.31	1.72	0.71	2.86	3.64	3.35	3.20	4.62	2.25	4.29	3.14	7.99

Media 1: Vermiculite, Media 2: Soil: Sand: FYM (1:1:1), M: Mist chamber conditions, G: Greenhouse conditions

Table 4. Effect of coppiced shoot cuttings planted in mid-July season on various parameters

Coppiced shoots	Sprouting per cent				Rooting per cent				Survival per cent				Root length				Collar diameter				Days taken to sprout			
	Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2	
	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G
Control	11.67	3.33	8.33	0.00	3.33	3.33	3.33	0.00	5.00	1.67	1.67	0.00	3.11	0.00	2.83	0.00	15.00	0.00	11.00	13.00	14.00	0.00	14.00	0.00
IBA 500 ppm	23.33	15.00	13.33	10.00	10.00	10.00	10.00	5.00	1.00	8.33	8.33	5.00	6.37	9.15	5.14	5.41	17.00	17.00	14.00	18.00	12.00	12.00	13.00	13.00
IBA 1000 ppm	8.33	5.00	3.33	5.00	3.33	3.33	3.33	3.33	3.33	3.33	3.33	1.67	4.38	6.11	4.16	4.33	13.00	11.00	12.00	13.00	13.00	14.00	15.00	15.00
IBA 1500 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
IBA 2000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 500 ppm	3.33	3.33	1.67	3.33	3.33	0.00	0.00	0.00	3.33	0.00	0.00	0.00	4.41	0.00	0.00	0.00	14.00	0.00	0.00	0.00	15.00	0.00	0.00	0.00
NAA 1000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 1500 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 2000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
SEM	1.06	1.02	1.16	0.56	0.96	0.73	1.27	0.56	0.81	1.00	1.00	0.56	0.04	0.07	0.04	0.02	0.12	0.04	0.13	0.09	0.09	0.05	0.18	0.04
CD (0.5%)	3.17	3.06	3.48	1.67	2.88	2.20	3.82	1.67	2.43	3.00	3.00	1.67	0.13	0.22	0.11	0.07	0.37	0.12	0.39	0.28	0.27	0.17	0.56	0.14
CV%	34.11	53.03	67.92	47.23	75.00	68.74	99.22	103.9	58.27	117.0	117.0	129.9	3.72	7.56	4.79	3.67	3.28	2.28	5.53	3.31	2.62	3.46	6.91	2.51

Media 1: Vermiculite, Media 2: Soil: Sand: FYM (1:1:1), M: Mist chamber conditions: Greenhouse conditions

one year old seedlings poorly sprouted whereas cuttings from coppicing shoots completely failed to sprout (Table 5). Cuttings planted in greenhouse failed to sprout regardless of the cutting type and growing media. The maximum sprouting percentage, rooting percent, survival per cent, root length and collar diameter observed was 11.67, 4.67, 3.33, 4.35 and 16mm, respectively. After one month interval, all of the survived cuttings from each season were transferred to polybag (5 cm x 7 cm) containing Soil: Sand: FYM (1:1:1). These cuttings were then placed under observation in greenhouse conditions for one month. 100% survival of cuttings was observed.

In forest trees, a large number of chemicals have been tested and reported to positively influence the rooting ability of tree species. The success of enhanced root proliferation may be attributed to transformation of chemical or a physiological response of the cuttings to the applied chemical or induced hydrolysis and mobilization of nutritional factors to the site of application thereby promoting root initiation in the cuttings. Among chemicals, auxin is essential for the propagation of cuttings in many plant species used in horticultural and in forestry industries. Usually, Indole-3-butyric acid (IBA) is found to be the most effective root promoting auxin and least toxic for plant tissues (Mazzini-guedes et al 2017). It is considered to be the best and most effective root promoting substance owing to its ability to produce a strong fibrous root system, non-toxic nature over a wide range of concentration, chemical stability and high degree of effectiveness in various plant species.

Mid-January season gave the best result among all three seasons whereas mid-November gave the worst results. This was because trees have great capability to grow in spring season and they retard their growth due to dormancy induction during autumn season. Season mid-July gave the intermediate results for all the parameters among all the seasons. Vegetative propagation of *Swietenia macrophylla* through branch cuttings treated with 0.4% IBA gave highest rooting per cent, sprouting per cent and survival per cent with 0.4% IBA treatment (Azad and Matin 2015). Vegetative propagation of *T. ciliata* revealed that maximum sprouting percent (52.22%) and rooting percent (36.61%) was in cuttings treated with 8000 ppm IBA. All the concentrations of IBA gave better results than the respective NAA concentrations (Thakur 2014). Rooting success in IBA 6000 ppm was (5.72% and was higher compared to control and IBA 4000 ppm. The survival in air layering was 11%, whereas, in root suckers (27.78% in *Tecomella undulata*. The coppice shoots emerged well on stumps of one year old seedlings during March. (Kaur et al 2019). IBA (250 mg L^{-1}) showed best results with sand in terms of rooting percent (80%), number of roots (70.63), root length (11.13) and number of leaves (5.25) per rooted mini-cuttings of *Azadirachta indica* under mist chamber conditions (Gehlot et al 2014).

Fig. 1. Sprouted cuttings

Fig. 2. One month old stem cuttings treated with IBA 500 and planted in vermiculite media under mist chamber conditions

Fig. 3. One month old stem cuttings treated with IBA 500 and planted in vermiculite media under mist chamber conditions

Table 5. Effect of stem cuttings planted in mid-November season on various parameters

Coppiced shoots	Sprouting per cent				Rooting per cent				Survival per cent				Root length				Collar diameter				Days taken to sprout			
	Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2		Media 1		Media 2	
	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G	M	G
Control	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
IBA 500 ppm	11.67	8.33	5.00	3.33	4.67	0.00	0.00	0.00	3.33	0.00	0.00	0.00	4.35	0.00	0.00	0.00	16.00	0.00	0.00	0.00	18.00	0.00	0.00	0.00
IBA 1000 ppm	3.33	3.33	1.67	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
IBA 1500 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
IBA 2000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 500 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 1000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 1500 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
NAA 2000 ppm	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
SEM	0.81	0.73	0.56	0.56	0.11	0.00	0.00	0.00	0.56	0.00	0.00	0.00	0.02	0.00	0.00	0.00	0.01	0.00	0.00	0.00	0.04	0.00	0.00	0.00
CD (0.5%)	2.43	2.20	1.67	1.67	0.33	0.00	0.00	0.00	1.67	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.02	0.00	0.00	0.00	0.13	0.00	0.00	0.00
CV%	75.75	98.20	129.9	259.8	37.12	0.00	0.00	0.00	259.8	0.00	0.00	0.00	6.05	0.00	0.00	0.00	0.54	0.00	0.00	0.00	3.79	0.00	0.00	0.00

Where, Media 1: Vermiculite, Media 2: Soil: Sand: FYM (1:1:1), M: Mist chamber conditions, G: Greenhouse conditions

CONCLUSION

T. ciliata respond well to macropropagation and could be a great option to propagate this species. Mid-march came out to be the best planting season for vegetative propagation of *T. ciliata*. Stem cuttings had better survival rate and longer roots than cuttings obtained from coppiced shoots. Vermiculite gave better results than Soil: Sand: FYM irrespective of the planting season or planting conditions. Similarly, IBA gave better results than NAA with respective concentration irrespective of the planting season, planting conditions or growth media. Overall, stem cuttings planted in mid-March season, treated with IBA 500 ppm, planted in vermiculite media and grown under mist chamber conditions came out to be the best combination to get significant and fruitful results.

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Evaluation of Primary Metabolites during Different Developmental Stages in *Alhagi maurorum* and *Caralluma edulis* from Arid Zone

Rajesh Kumar and Pawan K. Kasera*

Laboratory of Plant Ecology, Department of Botany, Centre of Advanced Study
Jai Narain Vyas University, Jodhpur-342 005, India
**E-mail: pkkasera1963@gmail.com*

Abstract: The present study deals with evaluation of primary metabolites in two important medicinal plants, *viz. Alhagi maurorum* (Camelthorn) and *Caralluma edulis* (Pimpa) from the Indian Thar desert. Primary metabolites such as leaf pigments, osmotic potential, proline, total sugars, crude protein and phosphorus were estimated during different developmental stages, *i.e.* vegetative, flowering and fruiting. The leaf pigments, total sugars, phosphorus and crude protein were maximum during vegetative stage, whereas proline and osmotic potential during flowering and fruiting stages, respectively in *A. maurorum.* In *C. edulis,* maximum amount of leaf pigments and proline were observed during fruiting stage; whereas osmotic potential, total sugars and phosphorus during flowering stage; and crude protein in vegetative stage.

Keywords: *Alhagi maurorum, Caralluma edulis,* Primary metabolites, Arid zone

In recent times, focus on plant research has increased all over the world and evidence has been collected to show the immense potential of medicinal plants used in traditional systems (Aali et al 2010). Plant medicines are ideal tools to restore health and treat disease precisely because they consist of a multiplicity of chemical components. The medicinal value of these plants lies in the bioactive phytochemical constituents that produce definite physiological effects on human body. These natural compounds formed the base of modern drugs as in use today (Rout et al 2009). Medicinal plants are of great importance not only for their biologically active secondary metabolites but also for their primary metabolites such as carbohydrates, proteins and lipids. These primary metabolites are essential for the growth and development of a plant and are carriers of chemical energy to the successive trophic levels of the food chain (Borkataky et al 2014).

Alhagi maurorum Medikus (Family: Fabaceae), commonly known as Camelthorn and whose distribution is restricted to rocky and gravelly soils of north-west Rajasthan, is a small erect shrub, armed with sharp and long spines and grows with massive rhizomatous systems, which extends up to 5-6 feet into the ground. It grows horizontally and has a potential to allow new shoots to grow upwards (Plate 1, Photo 1). The plants are used to treat numerous diseases such as gastro-entroenteritis (Varshney and Singh 2008), headache, toothache and cancer (Zou et al 2012), liver disorders, kidney stone and urinary tract infections (Badshah and Hussain 2011). *Caralluma edulis* Edgew. (Family: Apocynaceae), popularly known as Pimpa is an erect, succulent, branched, perennial herb with viscous watery sap (Plate 1, Photo 2). Traditionally, this plant is used to treat parasitic infections, Alzheimer disease, rheumatism, hypertension, gastric problems, diabetes, leprosy and is enriched in pregnane & megastigmane glycosides, flavone and esters (Adnan et al 2014). Thus, the present study was conducted to investigate estimation of primary metabolites such as leaf pigments, proline, osmotic potential, total sugars, crude protein and phosphorus during different developmental stages, *i.e.* vegetative, flowering and fruiting for evaluating suitable stage to harvest these plants for obtaining the maximum amount of these products.

MATERIAL AND METHODS

For chemical analyses of plants, leaf samples of *A. maurorum* were collected from Kanana, village of the Barmer district (25^0 49' 17.5944'' N and 72^0 25' 59.5416'' E), which is located 105 km away in south-west direction from the University Campus, Jodhpur during three developmental stages, *viz.* vegetative (August to February), flowering (March to May) and fruiting (April to July). The shoot samples of *C. edulis* were collected during vegetative (July and August), flowering (September to January) and fruiting (January to May) from Jaisoorana village of the distric Jaisalmer (27.0556^0 N and 71.1330^0 E), which is located 308.3 km away in western direction from the University Campus, Jodhpur.

The samples of both medicinal plants were collected during 2020 and 2021. The identification of plants were confirmed from the Botanical Survey of India, Jodhpur and specimens have been deposited in BSI herbarium. Plant materials were washed with running tap water to remove the adherent foreign particles, air-dried and used for chemical analyses. Leaf pigments, osmotic potential and proline were estimated in fresh leaf samples, while other parameters from oven-dried leaves in *A. maurorum,* but in *C. edulis* all parameters were estimated from the shoots. Fresh leaves and shoots were extracted with 80% acetone for estimation of leaf pigments as per Arnon (1949). Free proline and osmotic potential were estimated as per Bates et al (1973) and Janardhan et al (1975), respectively. The total sugars were estimated as per standard methods given by Plummer (1971), while nitrogen by micro-kjeldahl apparatus as suggested by Peach and Tracey (1955) and phosphorus as per Allen et al (1976). The data collected during both the years (2020 and 2021) were subjected to analyses of variance (ANOVA) as suggested by Gomez and Gomez (1984).

RESULTS AND DISCUSSION

The total chlorophylls and carotenoids ranged from 1.27 to 1.82 and 0.001373 to 0.002523 mg g^{-1} f. wt., respectively,

Plate 1. *Alhagi maurorum* with massive rhizome system (1), and *Caralluma edulis* in vegetative stage (2) growing in natural habitats

being the maximum during vegetative stage in *A. maurorum* (Table 1, Fig. 1). Daiya and Kasera (2016) reported highest values of total chlorophylls during vegetative stage in *Corbichonia decumbens*, which support present findings. The proline values ranged from 24.31 to 26.87 µg g^{-1} f. wt. during three stages, being the maximum in flowering stage and the minimum in fruiting stage (Table 1, Fig. 2). Daiya and Kasera (2016) reported maximum proline content at flowering stage in *C. decumbens*. Values for osmotic potential varied from 1.97 to 2.63 -MPa, being highest at fruiting while the lowest in flowering stage (Table 1, Fig. 2). Proline accumulation in *Trianthema triquetra* is accompanied by a decrease in osmotic potential (Mohammed et al 2000). In the present investigations, proline was maximum with minimum values of osmotic potential and *vice versa*, which is in accordance with present observations.

Total sugars ranged from 5.5 to 20.84 mg g^{-1} d. wt. during three phases, being the maximum in vegetative stage (Table 1, Fig. 3). Sagar and Kasera (2016) reported the maximum amount of total sugars during vegetative stage in *D. erythraeum.* The maximum crude protein were observed during vegetative stage (11.23% d. wt.) while non-significant variations were observed during flowering and fruiting stages. The phosphorus content was the highest (0.245% d. wt.) during vegetative followed by flowering and minimum at fruiting stage (Table 1, Fig. 4). Sagar and Kasera (2016) documented the highest content of crude protein along with phosphorus during vegetative stage in *Dipcadi erythraeum*. The present finding also support the above observations. The maximum content of total chlorophylls and carotenoids were reported during fruiting stage in *C. edulis*. The total chlorophylls and carotenoid values ranged from 0.348 to 0.404 and 0.00036 to 0.000508 mg g^{-1} f. wt., respectively

Table 1. Primary metabolic parameters in *A. maurorum* during different developmental stages

Parameters	Stages			CD
	Vegetative	Flowering	Fruiting	
Chlorophyll *a* (mg g^{-1} f. wt.)	1.26	0.890	0.914	0.2866^{ns}
Chlorophyll *b* (mg g^{-1} f. wt.)	0.557	0.382	0.500	0.1413^{ns}
Total chlorophylls (mg g^{-1} f. wt.)	1.82	1.27	1.41	0.4136^{ns}
Carotenoids (mg g^{-1} f. wt.)	0.002523	0.001466	0.001373	0.00044^{ns}
Proline (µg g^{-1} f. wt.)	24.81	26.87	24.31	3.8252*
Osmotic potential (-MPa)	2.34	2.63	1.97	0.6941^{ns}
Soluble sugar (mg g^{-1} d. wt.)	8.59	2.14	5.75	3.9463^{ns}
Insoluble sugar (mg g^{-1} d. wt.)	12.25	3.36	3.41	5.9409*
Total sugars (mg g^{-1} d. wt.)	20.84	5.5	9.16	6.9155*
Crude protein (% d. wt.)	11.23	8.58	8.58	3.5452^{ns}
Phosphorus (% d. wt.)	0.245	0.219	0.196	0.0327*

ns = Non-significant; and * = Significant at (<0.05) level

(Table 2, Fig. 5). Daiya and Kasera (2016) noted higher amount of carotenoids during fruiting stage in *C. decumbens*, which is in accordance with present studies.

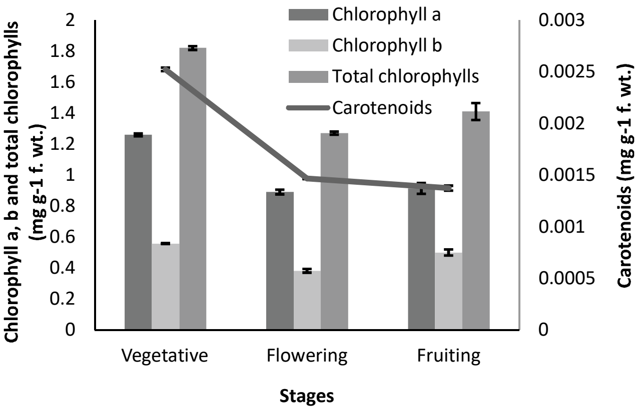

Fig. 1. Leaf pigment (mg g^{-1} f. wt.) contents in *A. maurorum* leaves during different growth stages (Mean values and standard error are presented)

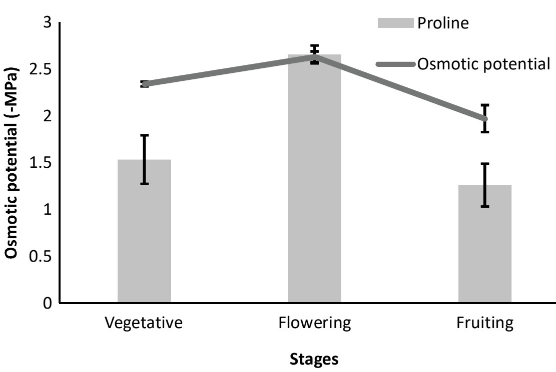

Fig. 2. Proline (µg g^{-1} f. wt.) and osmotic potential (-MPa) values in *A. maurorum* leaves during different growth stages (Mean values and standard error are presented)

In the present studies, the proline level increases with the decreases in osmotic potential in *C. edulis*, which can be correlated with its tolerance to environmental stress. The

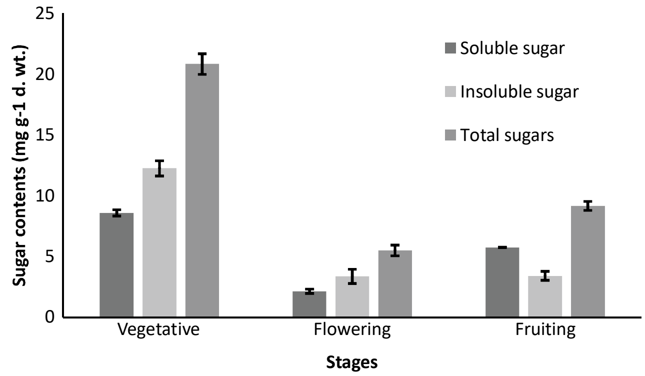

Fig. 3. Sugar (mg g^{-1} d. wt.) contents in *A. maurorum* leaves during different growth stages (Mean values and standard error are presented)

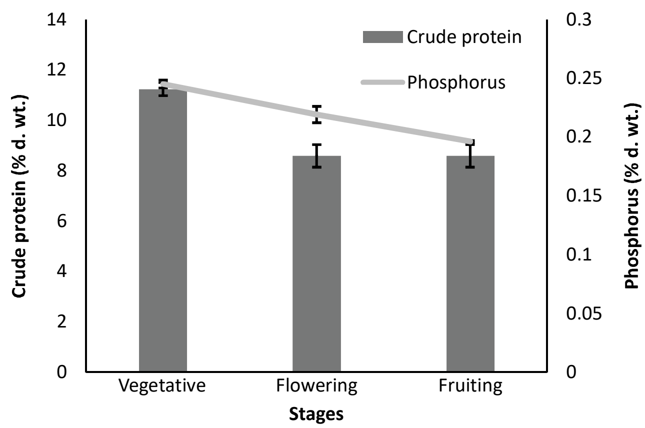

Fig. 4. Crude protein and phosphorus (% d. wt.) contents in *A. maurorum* leaves during different growth stages (Mean values and standard error are presented)

Table 2. Primary metabolic parameters in *C. edulis* during different developmental stages

Parameters	Stages			CD
	Vegetative	Flowering	Fruiting	
Chlorophyll *a* (mg g^{-1} f. wt.)	0.226	0.229	0.259	0.0515^{ns}
Chlorophyll *b* (mg g^{-1} f. wt.)	0.136	0.118	0.144	0.0315^{ns}
Total chlorophylls (mg g^{-1} f. wt.)	0.362	0.348	0.404	0.0775^{ns}
Carotenoids (mg g^{-1} f. wt.)	0.00036	0.00042	0.000508	8.3640*
Proline (µg g^{-1} f. wt.)	0.793	0.698	1.33	1.223*
Osmotic potential (-MPa)	0.985	0.834	1.13	0.3985^{ns}
Soluble sugar (mg g^{-1} d. wt.)	11.70	10.64	8.79	8.911^{ns}
Insoluble sugar (mg g^{-1} d. wt.)	2.26	6.93	7.22	4.358^{ns}
Total sugars (mg g^{-1} d. wt.)	13.97	17.56	16.01	9.299^{ns}
Crude protein (% d. wt.)	9.23	9.0	7.55	2.764^{ns}
Phosphorus (% d. wt.)	0.298	0.373	0.349	0.0997^{ns}

ns = Non-significant; and * = Significant at (<0.05) level

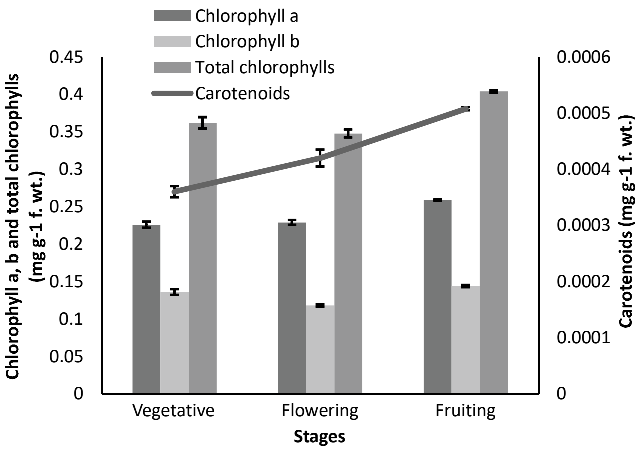

Fig. 5. Leaf pigment (mg g^{-1} f. wt.) contents in *C. edulis* shoots during different growth stages (Mean values and standard error are presented)

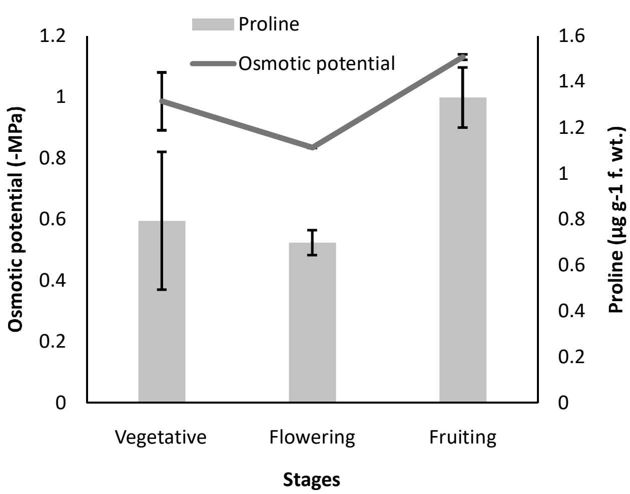

Fig. 6. Proline (µg g^{-1} f. wt.) and osmotic potential (-MPa) values in *C. edulis* shoots during different growth stages (Mean values and standard error are presented)

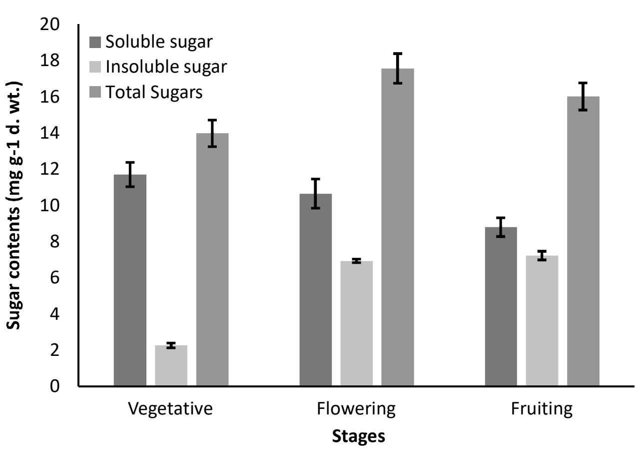

Fig. 7. Sugar (mg g^{-1} d. wt.) contents in *C. edulis* shoots during different growth stages (Mean values and standard error are presented)

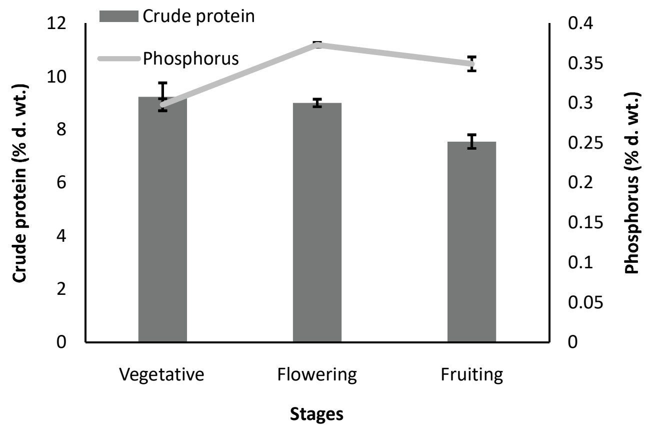

Fig. 8. Crude protein and phosphorus (% d. wt.) contents in *C. edulis* shoots during different growth stages (Mean values and standard error are presented)

higher values of proline were reported during fruiting stage, while osmotic potential in flowering stage (Table 2, Fig. 6). The values of proline and osmotic potential ranged from 0.698 to 1.33 µg g^{-1} f. wt. and 0.834 to 1.13 -MPa, respectively. The maximum proline and osmotic potential in *C. decumbens* were recorded during fruiting and flowering stages by Kasera et al (2018) and Daiya and Kasera (2016), respectively. Saharan et al (2001) also reported the lowest value of osmotic potential when proline content was at peak in *Evolvulus alsinoides,* which support the present findings. Hence, a positive correlation between proline and osmotic potential was observed in the present study. The maximum content of total sugars was reported during flowering stage, while the minimum in vegetative stage in *C. edulis*. The total sugars ranged from 13.97 to 17.56 mg g^{-1} d. wt. (Table 2, Fig. 7). Sagar and Kasera (2021) reported the maximum amount of total sugars during flowering stage in *Drimia indica*, which is supported by the present investigations. In *C. edulis*, crude protein and phosphorus values ranged from 7.55 to 9.23 and 0.298 to 0.373% d. wt., respectively (Table 2, Fig. 8). The maximum values of crude protein and phosphorus were noted during vegetative and flowering stages, respectively. Daiya and Kasera (2016) reported higher content of crude protein during vegetative stage in *C. decumbens.* Swami (2006) observed the maximum amount of phosphorus during flowering stage in *Solanum surattense.* The above observations are in accordance with the present investigations. The data on proline, total sugars and phosphorus were significant, while remaining parameters were non-significant in *A. maurorum,* whereas in *C. edulis*, except carotenoids and proline remaining all parameters were non-significant.

CONCLUSIONS

The leaf pigments, total sugars, phosphorus and crude protein were accumulated in maximum amount during vegetative stage, while proline during flowering and osmotic potential in fruiting stage in *A. maurorum.* In *C. edulis*, fruiting stage was the most favourable for obtaining the maximum production of leaf pigments and proline, while osmotic potential, total sugars and phosphorus during flowering and crude protein in vegetative stage. Thus, on the basis of present findings we can find out the most suitable developmental stage to obtain the maximum amount of these products for commercial utility.

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Macrophytes Diversity in Wetlands of North Dinajpur District, West Bengal, India

Sudeshna Mukherjee and Sujit Kumar Mandal*

Taxonomy of Angiosperms and Biosystematics Laboratory, Department of Botany
Sidho Kanho Birsha University, Purulia-723 104, India
**E-mail: smondal.bot@gmail.com*

***Abstract*:** Aquatic macrophytes comprise of taxonomically most diverse groups of macroscopic angiosperms that have great ecological impact to aquatic ecosystem. Macrophytes perform significant role in aquatic food webs and serve as host for others different ecological function as these are the most important element of aquatic ecosystem. The aim of the present study is to analysis the floristic composition and diversity of macrophytes from10 selected study areas along with their proper documentation for further research. The configuration of entire work are survey of selected wetlands, information gathered from local people, collection of macrophytes, documentation, description, and identification of collected species followed by checking their accepted names. The 66 species of vascular plants under 50 genera belonging to 27 families were recorded. Poaceae is the most dominant family with 11species.This study may consider as an important database of documented aquatic macrophytes from selected wetlands in North Dinajpur district which enables the future research work regarding this field and also may generate the global awareness about conservation of biodiversity of the concerned district.

***Keywords*:** Macrophytes, Diversity, Wetlands, North Dinajpur

Wetlands are the transitional zone between terrestrial and aquatic ecosystem, where the land is covered with shallow water and water table is at superficial level. The aquatic ecosystems are also considered as most productive ecosystem that play a major role in maintaining the wide range of biodiversity (Paul 2022). Aquatic macrophytes are the integral bioindicator of aquatic ecosystem which provide substratum for different invertebrates, serve as primary producers (Pagag and Borthakur 2020) and also involve in different ecological processes such as biomineralization, transpiration, sedimentation, nutrient recycling, accumulation of heavy metals, shaping and structuring of the ecosystem (Tiwari and Sandya 2022), food chain as the basic source of energy, shelter for other invertebrates, amphibians and birds (Chaudhury and Devkota 2021, Maybel 2022). The macrophytes include angiosperms, bryophytes and pteridophytes and all of they are may be of different categories like submerged, emergent, floating, free-floating. Various anthropogenic activities like urbanization, industrialization, human disturbances etc detrimentally influencing both the quantitative and qualitative parameters of wetland ecosystem (Germ et al 2021, Deka et al 2022, Khan et al 2022). Except some ethnobotanical studies of terrestrial plants no considerable studies have been carried out in the selected district about the relatable field. The present research may consider as blue print to investigate and analysis the overall diversity and distribution of the aquatic macrophytes from different selected wetlands of a totally unexplored district which may be unique.

MATERIAL AND METHODS

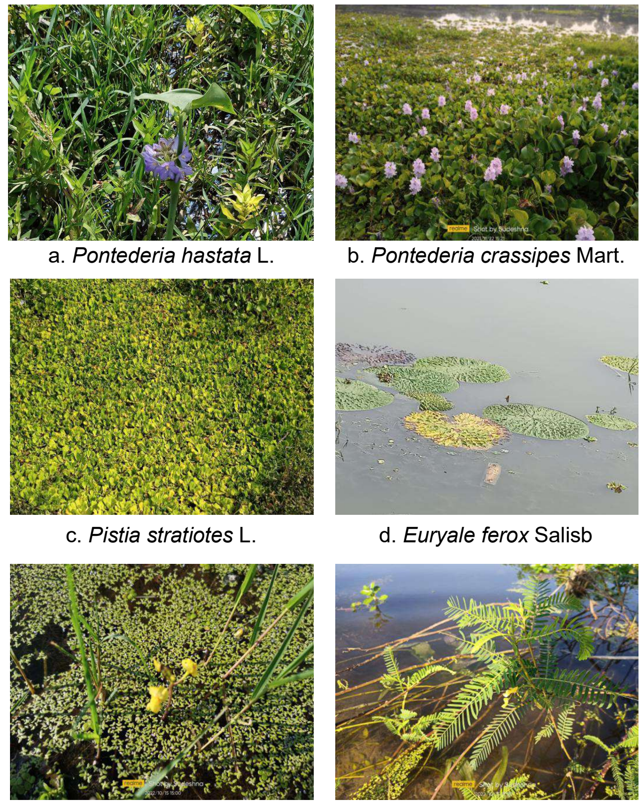
Study area: The selected study area is North Dinajpur district of West Bengal. North Dinajpur district lies between 25. 11°N to 26.49° N latitude and between 87.49° E to 90.00° E longitude occupying the total area of 3142 sq. km. The main rivers of this district are Kulik, Mahananda, Nagar etc. The district is rich in alluvial soil and mostly sandy to sandy-loam in texture and porous along with thick forest. Organic content of alluvial soil is medium to high. Due to presence of high rich organic containing soil, it helps to grow paddy, jute and sugarcane etc. Climate of this district is characterized by hot-summer with high humidity, abundant rainfall and cold winter.

Samples collection methods and identification: The selected wetlands from study area were surveyed and visited at different season from November (2021) to December (2022). The collected macrophytes were dried properly by changing papers at regular interval and worked out of the specimen in the laboratory. For proper identification of the collected specimens different standard taxonomic literatures(Cook 1996, Mandal and Mukherjee 2012, 2017, Manjunatha et al 2019, Panchal et al 2019, Anyinkeng et al 2020, Chanda and Bhowmik 2020, Humane 2020, Metwally et al 2020, Ravi et al 2020, Badole et al 2021, Patel and Sahoo 2021, Philippov and Komarova 2021, Rahangdale and Rahangdale 2021, Reshi et al 2021, Badra et al 2022, Chowdhury and Chowdhury 2022, Jeffry et al 2022, Jogd and 2022, Mishra and Singh 2022,Sahu and Roy 2022). POWO (Plants of the World Online) was used for the verification of the accepted names of the identified specimen. Plant specimens were mounted on the herbarium sheets for the preservation after drying properly. The prepared herbarium sheets were deposited in the herbarium of Taxonomy of Angiosperms and Biosystematics Laboratory, Department of Botany, Sidho Kanho Birsha University, Purulia.

RESULTS AND DISCUSSION

The present study reveals the documentation of macrophytes from different wetlands of research area to cover the vegetational distribution. For this study 10 wetlands were selected primarily for diversity analysis from different blocks of the North Dinajpur District (Table 1). A total of 66 species of macrophytes having 50 genera, belonging to 27 families were collected from the selected wetlands (Table 2). Among 27 families Poaceae was the most dominant family comprising of 11 species subsequently by Cyperaceae with 9 species (Fig. 2). Some of the macrophytes like *Pontederia hastata, Ipomoea aquatica, Acmella uliginosa* showed very common occurrences in 10 of the study site. Contradictorily *Najas indica, Neptunia oleracea, Marsilea minuta, Scirpus expansus* and *Utricularia inflexa* were very rare in their distribution. According to the habitat four different category of macrophytes were documented among which emergent species were 43, showing the highest percentage i. e. 65 %. Similarly lowest percentage was shown by rooted with floating leaves i. e. 4 % only.

Present status of macrophytes: Maximum number of macrophytes were documented from Chaitan Beel (22



a. *Pontederia hastata* L. b. *Pontederia crassipes* Mart.

c. *Pistia stratiotes* L. d. *Euryale ferox* Salisb

e. *Lemna minor* L. f. *Aeschynomene aspera* L.

Fig. 1. A few macrophytes of North Dinajpur district

Table 1. General information of wetlands in North Dinajpur District, West Bengal

Name of the wetlands	Block	Latitude	Longitude	Nearest village/town	Size in acres	Ownership pattern	Types of Wetland
Atkora Beel	Hemtabad	25.59 °N	88.22° E	Atkora, Malgram	62	Pr	P
Bagbari Beel	Itahar	25.55 °N	88.19° E	Chalunia, Gothlu	8.75	G	S
Chaitan Beel	Raiganj	25.66 °N	88.12° E	Malgram, Malancha	12	Pr	P
Chingri Beel	Itahar	25.54° N	88.19° E	Tilla, Mahagachi	187.5	G	P
Gidisha Beel	Kaliaganj	25.55 °N	88.19° E	Rainagar, Puyaltore	250	Pr	P
Gobra Beel	Raiganj	25.59 °N	88.13° E	Kuitore, Kalitala	312	Pr	S
Gothlu Beel	Itahar	25.47 °N	88.16° E	Gothlu, Bankur	32	Pr	P
Jogdol Beel	Raiganj	25.58°N	88.14° E	Dangapara Mahisbathan	100	G	P
Noyapara Beel	Raiganj	25.64 °N	88.12° E	Gheshor, Taherpur	19	Pr	P
Ranipur Beel	Itahar	25.55° N	88.19° E	Tilna, Bhadrashila	625	G	P

Pr- Private, G- Government, P- Perennial, S- Seasonal

Table 2. An enumeration of macrophytes in wetlands of North Dinajpur District

Scientific names	Family	1	2	3	4	5	6	7	8	9	10	Availability
Acmella uliginosa (Sw.) Cass.	Asteraceae	+		+	+	+		+			+	C
Actinoscirpus grossus (L.f.) Goetgh.& D.A. Simpson	Cyperaceae		+			+					+	C
Aeschynomene aspera L.	Fabaceae		+		+				+			C
Albidella oligococca (F. Muell.) Lehtonen	Alismataceae			+								LC
Alopecurus arundinaceus Poir.	Poaceae	+										LC
Alternanthera philoxeroides (Mart.) Griseb.	Amaranthaceae			+								LC
A.sessilis(L.)DC.	Amaranthaceae								+			LC
Ammannia auriculata Willd.	Lythraceae		+	+								LC
Arenaria serpyllifolia L.	Caryophyllaceae										+	LC
Azolla filiculoides Lam.	Salviniaceae			+			+		+			C
A. pinnata R.Br.	Salviniaceae								+			LC
Caesulia axillaris Roxb.	Asteraceae										+	LC
Calamagrostis epigejos (L.) Roth	Poaceae							+				LC
Ceratophyllum muricatum Cham.	Ceratophyllaceae				+							LC
Colocasia esculenta(L.) Schott	Araceae						+					LC
Commelina diffusa Burm.f.	Commelinaceae				+							LC
Cyanotis axillaris (L.) D. Don ex Sweet	Commelinaceae					+			+			LC
Cynosurus cristatus L.	Poaceae		+		+				+			C
Cyperus amabilis Vahl	Cyperaceae	+										LC
C. difformis L.	Cyperaceae										+	LC
C. fuscus L.	Cyperaceae							+			+	LC
C. tenuispica Steud.	Cyperaceae		+									LC
Dactylis glomerata L.	Poaceae		+									LC
Echinochloa colonum (L.)Link	Poaceae			+							+	LC
Eclipta prostrata (L.) L.	Asteraceae			+								LC
Eleocharis atropurpurea (Retz.) J. Presl & C. Presl	Cyperaceae		+									LC
Euryale ferox Salisb.	Nymphaeaceae								+			LC
Holcus lanatus L.	Poaceae								+			LC
Hydrilla verticillata (L.f.) Royle	Hydrocharitaceae	+	+						+			C
Hygroryza aristata (Retz.) Nees ex Wright & Arn.	Poaceae			+								LC
Hypericum hirsutum L.	Hypericaceae		+									LC
Ipomoea aquatica Forssk.	Convolvulaceae	+			+		+	+	+	+	+	C
I. quamoclit L.	Convolvulaceae		+		+						+	C
Lemna gibba L.	Araceae				+		+					LC
L. minor L.	Araceae				+				+			LC
Limnophila heterophylla (Roxb.) Benth.	Plantaginaceae		+	+	+						+	C
L. indica (L.) Druce	Plantaginaceae						+					LC
Lindernia dubia (L.) Pennell	Linderniaceae				+							LC
Ludwigia adscendens (L.) H. Hara	Onagraceae			+	+		+	+	+	+	+	C
L. perennis L.	Onagraceae	+		+				+		+		C
Marsilea coromandelina Willd.	Marsileaceae							+				LC
M. minuta L.	Marsileaceae	+										LC
M. quadrifolia L.	Marsileaceae		+	+	+				+	+		C

Cont...

Table 2. An enumeration of macrophytes in wetlands of North Dinajpur District

Scientific names	Family	1	2	3	4	5	6	7	8	9	10	Availability
Murdannia pauciflora (G. Bruckn.) G. Bruckn.	Commelinaceae			+						+		LC
Najas indica (Willd.) Cham.	Hydrocharitaceae				+							LC
Neptunia oleracea Lour.	Fabaceae								+			LC
Nymphoides hydrophylla (Lour.) Kuntze	Menyanthaceae		+				+					LC
Panicum repens L.	Poaceae			+				+			+	C
Paspalum distichum L.	Poaceae		+									LC
Persicaria hydropiper (L.) Delarbre	Polygonaceae				+		+	+			+	C
P. longiseta (Bruijn) Kitag.	Polygonaceae						+					LC
Phleum pratense L.	Poaceae						+	+	+	+	+	C
Pistia stratiotes L.	Araceae	+	+	+			+	+		+		C
Pontederia crassipes Mart.	Pontederiaceae			+					+	+		C
P. hastata L.	Pontederiaceae		+	+	+	+	+	+	+		+	C
Rumex maritimus L.	Polygonaceae			+								LC
Salvinia molesta D. Mitch.	Salviniaceae							+	+			LC
Schoenoplectiella articulata (L.) Lye	Cyperaceae	+		+		+				+	+	C
S. corymbosa (Roth ex Roem.& Schult.) J.R. Starr & Jim. Mejias	Cyperaceae					+						LC
Scirpus expansus Fernald	Cyperaceae				+							LC
Setaria geminate (Forssk.)Veldkamp	Poaceae			+						+		LC
Succisa pratensis Moench	Caprifoliaceae					+			+		+	C
Torenia crustacea (L.) Cham. & Schltdl.	Linderniaceae		+	+	+	+		+	+		+	C
Utricularia aurea Lour.	Lentibulariaceae	+		+	+					+	+	C
U. australis R.Br.	Lentibulariaceae					+						LC
U. inflexa Forssk.	Lentibulariaceae		+									LC

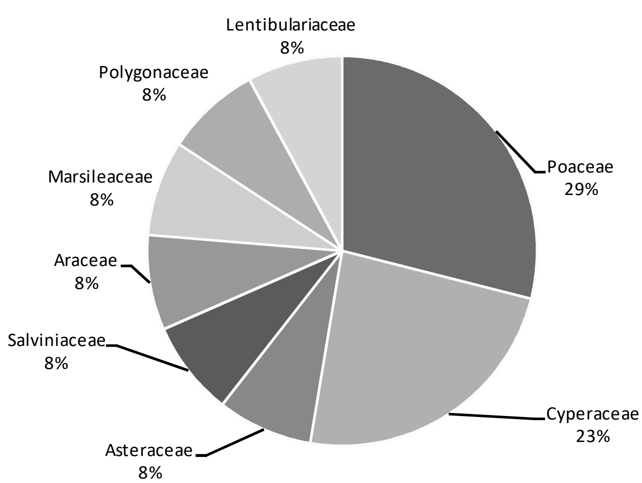


Fig. 2. Dominant families of macrophytes

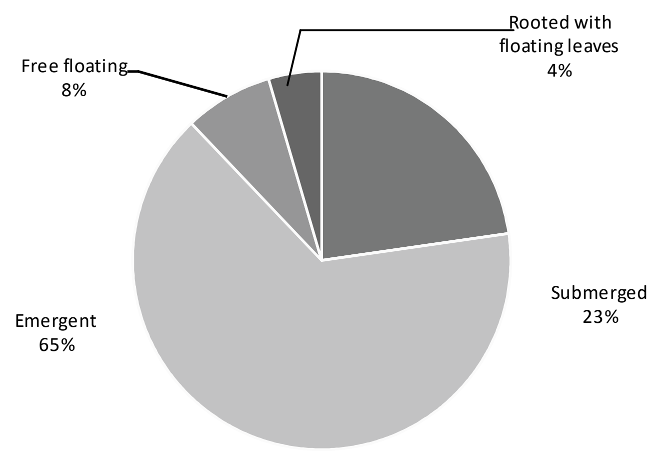


Fig. 3. Distribution of species according to habitat

species), Jogdal Beel (20 species). Chingri Beel and Ranipur Beel (each contain 19 species). Out of 66 species, *Pontederia hastata* was most dominant species as it was present in 8 different wetlands among the selected 10 wetlands. Other common species were *Ipomoea aquatica, Ludwigia adscendense* and *Torenia crustace*a all three were present in 7 of the 10 wetlands, followed by *Acmella uliginosa, Pistia stratiotes* were reported from 6 wetlands, and *Marsilea quadrifolia*, *Schoenoplectiella articulata, Utricularia aurea* were from 5 wetlands. There are some species of macrophytes also documented in this study which were confined to only a particular wetland from 10 of the study sites such as *Utricularia inflexa* from Bagbari Beel, *Utricularia australis* from Gidisha Beel, *Persicaria longiseta* from Gobra Beel, *Najas indica* from Chingri Beel, *Marsilea minuta* from Atkora Beel, *Marsilea coromandelina* from Gothlu Beel and so on.

The analysis of vegetational diversity and distribution of macrophytes revealed the great fluctuation of environmental factors that have a major impact on the growth, abundance as well as physiognomy of the macrophytes community. The occurrence and agglomeration of particular macrophytes are significantly influenced by some natural factors confined to a particular environment. Similarly, some macrophytes are rare in their occurrence in certain wetlands in response to their growth and adaptation according to the environmental variation.

CONCLUSION

The overall assessment of the study also indicates the gradual degradation of the wetlands along with their rich macrophytes vegetation due to various anthropogenic activities like eutrophication, encroachment for building construction, efflux of industrial and drainage water, pollution, etc. Therefore, immediate necessary action should be taken for the conservation of macrophytes in their native region from degradation and sustainable use of the wetland.

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Provenance Variation in Fruit and Seed Morphometric Characteristics of *Dysoxylum binectariferum* across Distribution in India

Suraj R. Hosur, A. Krishna, B.H. Shivakumar, M.R. Jagadish, R. Vasudeva and P. Mohanakumara[1]

College of Forestry Sirsi, University of Agricultural Sciences, Dharwad-581401, India
[1]KRC College of Horticulture, Arabhavi (University of Horticultural Sciences), Bagalkot-587 104, India
E-mail: srj1419@gmail.com

Abstract: *Dysoxylum binectariferum* is a rich source of rohitukine among known natural source, a billion-dollar anti-cancer drug. During January-March 2022, a pioneer study on provenance variation in fruit and seed morphometric traits in recalcitrant *D. binectariferum* species across its distribution in India was conducted. Seeds were collected from superior trees selected through eye ball screening from 7 provenances covering Assam, Arunanchal Pradesh, Karnataka and Maharashtra states. Seed morphometric characteristics were determined using digital caliper and electronic weighing balance and differences in performance of characteristic among provenances were compared using analysis of variance. Analysis of variance revealed significant variation in seed morphometric traits among 7 provenances. All fruit traits and seed traits of Jog provenance were found to be significantly superior over other provenances. Among trees, JT1 found to be best in fruit parameters and seed parameters over the other trees. The superior trees identified in the present study can further used for tree improvement work in order to get maximum genetic gain in timber and metabolite production.

Keywords: *Dysoxylum binectariferum*, Seed morphometric traits, Provenance variation, Superior tree

Dysoxylum binectariferum (Roxb.) Hook. (*Meliaceae)* is a medicinally important species grows medium to large-sized tree found in tropical and subtropical climates. The evergreen tree *D. binectariferum* is native to India, China, and other parts of Asia . *D. binectariferum* has gained international importance because of the presence of dysobinin and rohitukine. Dysobinin, a tetranortriterpenes isolated from the fruits, is a general central nervous system (CNS) depressant and also is reported to have mild anti-inflammatory activity (Singh et al 1976). Among all known botanical sources of rohitukine, *D. binectariferum* is reported to yield the highest amounts of rohitukine (Naik et al 1988). Further, the bark of the tree is also reported to be used for the treatment of leprosy and foul ulcers (Jain and DeFilipps 1991). Because of its economic value, *D. binectariferum* has been excessively harvested in many parts of the Western Ghats (Nath et al 2005). Despite its high value, only small-scale research on fruit and seed morphometric variation is conducted, and no literature on seed germination and propagation is available. Seed characters study is necessary for successful establishment and long-term secondary metabolite supply through selection of superior seed sources as well as superior mother trees.

Seed collected from same geographical region vary in their morphological characteristics, which intern reflects the genetic control of formation of seed (Abdelkheir et al 2003). *Dysoxylum binectariferum* has vast diversity between trees as well as among provenances. However, species distribution mainly affected by the extrinsic factors such as rodent's predation, seeds which are infected by the fungus on surface litter and adverse climatic conditions like lack of moisture for germination of fallen seeds just after the winter are the major causes that hinders seed germination (Gunaga et al 2015). Therefore, the identification of superior tree, source is necessary to get maximum benefits. With this background study was undertaken to know the intra-species variation in fruit and seed morphological traits of *D. binectariferum* and to identify the best seed source for better multiflication.

MATERIAL AND METHODS

Seed sources: Clues on the natural populations of *D. binectariferum* were obtained by consulting various published literatures Mohanakumara et al (2010), Gunaga et al (2015) and consulting various scientist who worked on this species as well as from forest department staffs. Extensive filed surveys were conducted to locate exact location of the populations. Natural populations of *D. binectariferum* are scanty and always small. Extensive and repeated visits were made to locate populations. Overall, seven populations from four states were located *viz.*, four populations (Jog, Kargal, Benagaov and Kathagal) from Karnataka, one population (Lonavala) from Maharashtra, one population (Manas) from Assam and another one population (Phasighat) from Arunanchal Pradesh (Fig. 1).

Seed collection: In each population, five superior trees (in girth class 50-150cm) were selected based on eye ball screening method (Hanumanth 2020). The trees were marked and their height, gbh and crown spread were recorded using altimeter and tape respectively. Since maturity of fruits vary in Western Ghat (2nd Fortnight of January) and North-East region (1st Fortnight of March). So matured 75 fruits from 5 superior trees from each source were collected during January from Western Ghat source and March from North-East source using tree pruner. Trees were coded with source and tree number ex. JT1 means First numbered tree from Jog source. Further fruit and seed parameters like length, width and weight were recorded using digital caliper and electronic weighing balance.

Statistical analysis: One way analysis of variation for tree growth parameters and two-way analysis of variation for fruit and seed morphometric traits was done using Operational Statistics (opstat) software.

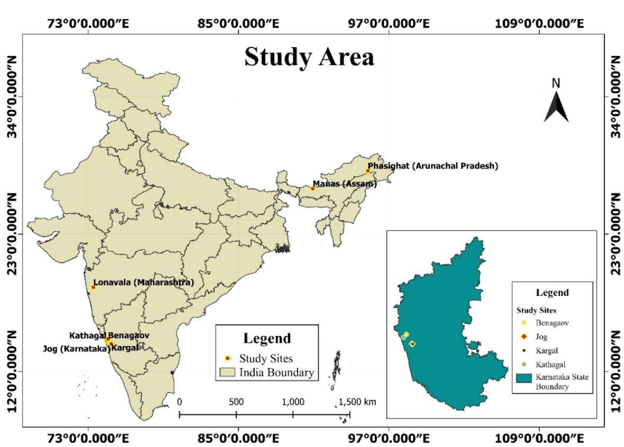

Fig. 1. Location of the study sites

RESULTS AND DISCUSSION

Variation in growth parameters of superior trees: Significant differences across population were observed for tree height, gbh and crown diameter of *D. binectariferum* is shown in Table 1. Highest gbh was observed for Jog (1.17m) and lowest for Kathagal and Lonavala (0.86m and 0.80m respectively), highest height was observed for Kargal (14.00m) and lowest for Manas and Phasighat (7.58m and 7.20m, respectively) and highest crown diameter was recorded for Benagaov and lowest for Phasighat. Overall tree girth is similar in Western Ghats and North-eastern regions. But height (7.20-7.58m) and crown diameter (2.80-3.10m) of North-Eastern region were significantly lower than Western Ghat regions height (11.00-14.00m) and crown diameter (4.50-5.50m). These variations might be contributed by the genetic makeup of the selected trees across different sites (Senthil Kumar et al 2010).

Effect of provenance and tree variation on fruit parameters: Significant variations were recorded for fruit traits across population as well as between trees. Among population, the fruit traits, namely fruit weight (71.58 g), fruit length (49.56 mm) and fruit diameter (54.45 mm) from Jog were found to be superior over the other followed by Manas and Phasighat, the second best sources, whereas least was observed in Lonavala population for all fruit parameters. Average fruit weight (75.90 g and 72.85g) respectively from Manas and Phasighat were on par with Jog population. Among superior trees, highest fruit weight (95.20g) and highest fruit length (56.68mm) recorded for JT1, highest fruit diameter (57.77mm) for JT4 and least was recorded for LT2 tree in all fruit parameters (Table 2). The variation existed among population as well as between trees for fruit traits is expected since the species naturally grows over wide range of climatic conditions. The results were conformity with the reports of Patil (2012) in *Zanthoxylum rhetsa*, Uma (2015) on *Buchanania lanzan*, Kallaje (2000) in *Garcinia indica*, Tomar and Rattan (2012) on *Hippophae salicifolia* and Jamaludeen et al (2015) in *Lagestromia speciosa*.

Table 1. Variation in growth parameters of selected trees of *D. binectariferum*

Mountain ranges	Sl. No.	Girth (m)	Height (m)	Crown diameter (m)
Western Ghat	Jog	1.17^{a}	11.20^{b}	4.70^{c}
	Kargal	1.07^{b}	14.00^{a}	4.90^{c}
	Benagaov	1.00^{b}	12.80^{ab}	5.50^{a}
	Kathagal	0.86^{c}	11.00^{b}	5.20^{b}
	Lonavala	0.80^{c}	11.30^{b}	4.50^{d}
North-East	Manas	1.06^{b}	7.58^{c}	3.10^{e}
	Phasighat	1.07^{b}	7.20^{c}	2.80^{f}
	SE(m)±	0.03	0.81	0.89
	CD (p=0.05)	0.09	2.37	0.30

Table 2. Effect of seed source and tree variation on fruit parameters of *Dysoxylum binectariferum*

Source	Trees	Fruit weight (g)		Fruit length (mm)		Fruit diameter (mm)	
Joga	JT1	95.20		56.68		57.10	
	JT2	51.44		39.82		51.45	
	JT3	51.57		48.24		54.39	
	JT4	86.66		55.62		57.77	
	JT5	73.04		47.43		51.52	
	Mn	71.58		49.56		54.45	
Kargal	KT1	51.92		45.78		46.19	
	K T2	52.38		42.01		56.62	
	K T3	45.38		39.22		45.75	
	K T4	57.47		41.94		55.22	
	K T5	53.07		40.02		49.51	
	Mn	52.05		41.79		50.66	
Kathagal	KtT1	56.81		45.28		48.15	
	Kt T2	38.86		40.89		49.70	
	Kt T3	61.53		42.43		54.05	
	Kt T4	61.52		47.06		49.60	
	Kt T5	50.96		40.29		48.18	
	Mn	53.94		43.19		49.94	
Benagav	T1	31.88		35.96		41.84	
	T2	37.68		38.85		49.42	
	T3	53.97		43.66		46.85	
	T4	38.98		37.35		44.14	
	T5	55.01		40.40		49.73	
	Mn	43.51		39.24		46.40	
Lonavala	LT1	44.89		39.52		42.23	
	LT2	6.94		27.58		21.84	
	LT3	43.37		38.90		42.18	
	LT4	46.07		39.52		43.51	
	LT5	18.49		32.11		30.38	
	Mn	31.95		35.03		36.03	
Manas	MT1	79.51		51.81		42.37	
	MT2	68.01		41.59		58.13	
	MT3	93.04		44.27		51.57	
	MT4	42.80		50.04		50.09	
	MT5	96.13		49.55		56.89	
	Mn	75.90		47.45		51.81	
Phasighat	PT1	43.02		52.27		48.17	
	PT2	92.37		46.62		49.09	
	PT3	75.28		53.56		53.73	
	PT4	64.45		39.82		42.36	
	PT5	89.14		47.29		56.84	
	Mn	72.85		47.91		50.04	
		SE(m)±	CD@5%	SE(m)±	CD@5%	SE(m)±	CD@5%
	Source	2.596	7.338	0.554	1.565	0.955	1.908
	Trees	2.194	6.202	0.468	1.323	0.807	1.613
	Source x Trees	5.805	16.409	1.238	3.499	2.135	4.267

Table 3. Effect of seed source and tree variation on seed parameters of *Dysoxylum binectariferum*

Source	Trees	Seed weight (gm)		Seed length (mm)		Seed diameter (mm)	
Joga	JT1	9.68		33.48		22.28	
	JT2	7.51		33.04		20.17	
	JT3	8.67		30.54		20.81	
	JT4	8.13		31.89		20.45	
	JT5	8.40		31.46		20.82	
	Mean	8.48		32.08		20.91	
Kargal	KaT1	6.96		28.22		19.47	
	Ka T2	7.07		26.77		19.70	
	Ka T3	6.18		23.99		18.87	
	Ka T4	7.59		27.31		20.34	
	Ka T5	6.51		27.35		19.60	
	Mean	6.86		26.73		19.60	
Kathagal	KtT1	7.19		28.64		20.10	
	Kt T2	7.24		26.64		20.63	
	Kt T3	6.49		24.38		26.57	
	Kt T4	6.62		25.40		19.64	
	Kt T5	4.70		24.08		17.03	
	Mean	6.45		25.83		20.79	
Benagav	BT1	3.69		23.24		15.91	
	BT2	7.20		26.77		19.45	
	BT3	3.88		25.84		16.25	
	BT4	3.91		25.03		15.78	
	BT5	8.11		27.54		20.21	
	Mean	5.36		25.68		17.52	
Lonavala	LT1	3.65		25.18		15.65	
	LT2	2.19		27.07		13.25	
	LT3	4.46		26.18		17.00	
	LT4	3.15		22.12		14.93	
	LT5	2.01		19.73		12.46	
	Mean	3.09		24.06		14.66	
Manas	MT1	8.15		25.49		20.64	
	MT2	9.03		31.79		21.54	
	MT3	7.67		28.92		19.95	
	MT4	9.17		29.15		20.42	
	MT5	6.39		31.68		21.88	
	Mean	8.08		29.41		20.89	
Phasighat	PT1	7.46		32.37		18.59	
	PT2	8.97		27.32		19.24	
	PT3	6.18		27.51		20.44	
	PT4	7.83		28.74		19.67	
	PT5	8.89		28.58		19.98	
	Mean	7.87		28.90		19.58	
		SE(m)±	CD@5%	SE(m)±	CD@5%	SE(m)±	CD@5%
	Source	0.196	0.555	0.418	1.182	0.372	1.051
	Trees	0.166	0.469	0.353	0.999	0.314	0.888
	Source x Trees	0.439	1.241	0.935	2.642	0.831	2.350

Effect of provenance and tree variation on seed parameters: Seed is a propagule which depict the growth and performance of a particular plant species. Seed polymorphism is a common feature for adaptation, which includes production of seeds of different size, weight, seed coat patterns etc. Selection for seed source with larger seeds may result in vigour and healthy seedlings. The significant differences in various seed morphological characteristics of *D. binectariferum* provenance is indicative of the possibility of selecting phenotypically superior plant within the species for further improvement work. But environmental factors also play a role in changing the component of the seed size as the species grow in wide range of ecological conditions and hence, populations can be expected to experience markedly selection pressure on seed characteristics.

Statistical parameters for various seed traits among seven provenances of *D. binectariferum* showed significant differences among the geographic source and trees of same source with regards to seed weight, seed length and seed diameter. Jog provenance was superior over other provenance in terms of seed weight (8.48g), seed length (32.08mm) and seed diameter (20.91mm) (Table 3). Jog seed diameter is on par with Manas (20.89mm) and Kathagal (20.79mm), similarly seed weight of Jog is on par with Manas (8.08g). There was significant variation among thirty-five superior trees from seven provenances for all seed traits, seed weight varied from 2.01g (LT5) to 9.68g (JT1), seed length varied from 19.73mm (LT5) to 33.48mm (JT1) and seed diameter varied from 12.46mm (LT5) to 26.57mm (KtT3) (Table 3).

Seed and fruit morphometric traits of this tree species are important because seed kernels and fruits contain considerable amount of rohitukine (Mohanakumara et al 2010). In the present study, among the various populations it was identified that, Jog source was best over others in terms of fruit weight (55.36%), fruit length (29.32%), fruit diameter (33.83%), seed weight (63.56%), seed length (27.24%) and seed diameter (42.63%) higher than Lonavala source. JT1 is superior tree over others in terms of fruit weight (92.71%), fruit length (51.34%), fruit diameter (61.75%) higher than LT2 and seed weight (79.23%), seed length (41.06%), seed diameter (44.07%) higher than LT5. Evidently, greater population variations and tree to tree variations were observed for fruit and seed morphometric traits which intern helps in the selection of superior source and superior types selection. Such large variations obviously indicate existing genetic diversity level in the species (Arjun 2017).

Similar kind of significant variation was found between *D. binectariferum* trees from Honnavar geographical sources were seed weight varied from 0.32-0.59g, seed length from 21.3-27.2mm and seed diameter 16.7-20.3mm among ten trees studied (Gunaga et al 2015). This variation may be due to wide range of ecological conditions quoted by Gunaga et al (2020) in *Saraca asoca.* Wide range of rainfall, temperature and soil type of seed sources with superior seed length and width possessed higher seed weight in *Zanthoxylum rhetsa* (Patil 2012) and also variations for reproductive traits are highly controlled by genetic factors (Zobel and Talbert 1984). Similar findings were observed by Basappa (2011) in *Emblica officinalis*, Manjunath (2003) in *D. malabaricum,* Abraham et al (2006) in *Cordia africana* and Hareesh et al (2008) in *Nathapodytes nimmoniana.*

Table 4. Correlation for fruit traits of *D. binectariferum*

Character	Fruit length	Fruit diameter	Fruit weight
Fruit length	-	0.77**	0.92**
Fruit diameter	0.76*	-	0.84**
Fruit weight	0.90*	0.84*	-

**: Genotypic correlations *: Phenotypic correlations

Table 5. Correlation for seed traits of *D. binectariferum*

Character	Seed length	Seed diameter	Seed weight
Seed length	-	0.64**	0.73**
Seed diameter	0.46*	-	0.89**
Seed weight	0.67*	0.71*	-

**: Genotypic correlations *: Phenotypic correlations

Correlation for fruit and seed traits: From the correlation matrix of different fruit traits (Table 4), it is evident that fruit length increases with fruit diameter (r = 0.77), fruit weight (r = 0.92). A positive correlation was observed between fruit diameter vs fruit weight (r = 0.84). Similarly seed length increases with seed diameter (r = 0.64), seed weight (r = 0.73). A positive correlation was observed between seed diameter and seed weight (r = 0.89) (Table 5). Correlation among traits is important to be studied since selection for one trait influences the other based on the nature and strength of correlations. Selection of provenances with larger seeds may result in sturdy and better seedlings (Arjun 2017). This kind of relation can be attributed to provision of larger amounts of soluble sugars required for germination and radicle protrusion and other energy expending developmental processes (Mamo et al 2006).

CONCLUSION

In the present study fruit parameters and seed parameters varied significantly among provenance, trees and their interaction. All fruit traits and seed traits of Jog provenance were found to be superior over other provenances. Among trees, JT1 found to be best in fruit parameters and seed parameter over the other trees followed by JT4. Greater tree to tree and population variations existing in this species, that can be utilised to select superior types for tree improvement work in order get maximum genetic gain.

AUTHOR CONTRIBUTION

Suraj R Hosur, conceived and designed the experiment, performed the experiment, analysed and interpreted the data, wrote the paper. Shivakumar BH perormed the experiment. Krishna A, Jagadish MR, Vasudeva R and Mohanakumar, conceived and designed the experiments.

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Morphological Variation of Fruits and Seeds of *Garcinia indica* (Thouars) Choisy in Western Ghats Region of Karnataka, India

Anila M. Sunny, T. Hamalton, R. Hegde[1], S.L. Renjini and N. Ravi*

Institute of Wood Science and Technology, Bengaluru- 560 003, India
[1]College of Forestry, University of Agriculture and Horticultural Science, Shivamogga- 571 216, India
**E-mail: nravi@icfre.org*

Abstract: *Garcinia indica* is an important indigenous tree, which grows in tropical and evergreen forests of India and has a tremendous potential to be utilised in various value-added applications. The present study aims at understanding the variation in fruit and seed characters among 16 populations in Western ghats region of Karnataka. The large variation exists in economically important characters such as seed weight, fruit weight and rind weight and a significant variation was observed in morphological characters of different populations. The samples collected from Gundya region shown highest fruit and seed traits except in rind weight and rind thickness which was high in the fruit samples collected from Belthangady. A strong dependence of morphological characters on geoclimatic factors was revealed in correlation studies. The cluster analysis carried indicated that these16 population divided into two major groups based on the different morphological characters studied. The results concluded that *G. indica* showed a high level of phenotypic plasticity and these results can be used as a foundation for selective breeding.

Keywords: *Garcinia indica*, Fruit and seed characters, Morphological variation, Principal component analysis (PCA), Cluster analysis

Garcinia indica (Thouars) Choisy (Kokum) is a polygamodioceous tree, belongs to the family Clusiaceae. It has multifarious uses both traditionally and commercially. Kokum has been wildly used by the local communities for therapeutic, culinary and beverage purposes. Commercially, Kokum butter and rinds has been used by the cosmetic, pharmaceutical, nutraceutical and beverage industries (Swami et al 2014). The red coloured antioxidant dye present in rinds of kokum is being used as a natural food colourant. The bioactive compounds present in *G. indica* such as Hydroxy citric acid and Garcinol are known for its anti-obesity and anti-oxidant properties respectively (Ravi et al 2022). It is an under exploited tree, which grows in a dissipate manner in forest land, riverside, roadside, waste land and generally does not require much rainfall for their growth (Swami et al. 2014). This tree can flourish at coastal belts and thrive well upto an elevation of 800 metres above mean sea level (Nayak et al 2010, Braganza et al 2012). The wide variation in morphological characters of a tree species is primarily govern by geo-specific environmental conditions (Ji et al 2016). Studying of variation in phenotypic characters are the markers for the genotypic selection of a tree, which point towards the adaptive evolution (Pigliucci et al 2006). The variation in morphological traits of a same species is controlled by the genetical composition of the tree species and environmental factors or through their interaction. Morphological variation in an ecological zone is controlled or induced by micro climatic factors viz., rainfall, humidity, temperature etc. (Gouwakinnou et al 2011, Atefe et al 2015). It is vital to assess the intra specific variation among populations in a robust manner to determine the effect of geoclimatic and genetic parameters on morphological variation (Franks et al 2013). Recently few studies have been carried out to understand the morphological variation in *Garcinia* species. In *Garcinia* species the morphological variation exists within the species in same ecosystem (Parthasarathy 2014). *Garcinia gummi-gutta* grows highlands, coastal belts, and river banks however, the tree grown near river bank had high productivity when compared to other habitats (Aswathi et al 2018). The existence of tree in wide range of altitude, the dioecious and cross - pollinated nature of *G. indica* harbour a wide genetic diversity in morphological and biochemical characters among different populations (Joseph et al 2015). However, there was no studies on morphological variation on fruit and seed characters in *G. indica* in Western ghat region of Karnataka, Southern India. The present study aims to elucidate the magnitude of morphological variation in economically important fruit and seed characters of *G. indica* Choisy among 16 populations in Karnataka based on the climate and geographical parameters.

MATERIAL AND METHODS

Study area: The study was carried out in Western ghats region of Karnataka, Southern India, based on occurrence and abundance of tree and area consists of 16 different populations of Western ghats namely, Puttur, Udupi, Mangalore, Bhatkal, Honnavar, Karwar, Belthangady, Madikeri, Dandeli, Dharwad, Yellapur, Londa, Sirsi, Charmady, Kukke and Gundya. The selected site was separated from each other by a distance of at least 50 km to avoid sampling error.

Data collection: The latitude, longitude and altitude of study site were measured using Garmin GPS device. The mean annual rainfall from March 2021-May 2022 data were obtained from https://mausam.imd.gov.in/ (Table 1). The sample selection sites were marked in digital elevation map (DEM) by using QGIS (3.4) software (Fig. 1). Fruits were

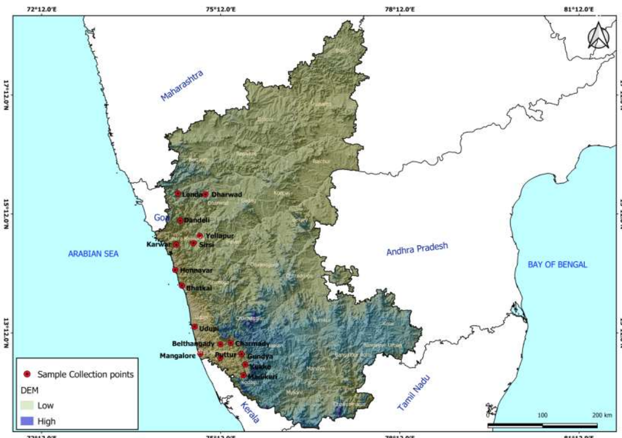

Fig. 1. Location map of population selected in Western ghats, Karnataka

collected from the selected matured trees having total height and breast diameter of 11-50 m and 15.55-70.7cm respectively were selected. The former was measured using range finder (Forestry proII) and later by a measuring tape. In each site the fruits were collected from 10-15 selected trees and bulked for further study. Physiologically matured, disease and pest free fruits were selected. Fruits of 100 numbers from the bulk were selected randomly for the morphological variation analysis. The morphological data such as fruit weight (FW), total seed weight (SW), rind thickness (RT), rind weight (RW), fruit length (FL), fruit diameter (FD), seed length (SL) and seed diameter (SD) was recorded. The RT, FL, FD, SL, SD were measured using vernier caliper with an accuracy of 0.01mm and FW, SW and RW were using weighing balance with an accuracy of 0.001 g.

Statistical analysis: The data was analysed using SPSS 20 software (IBM company, New York, USA). Significant difference among means were analysed using Tukey's post hoc test. Karl Pearson's correlation were calculated and analysed among the fruit and seed characters with geoclimatic parameters. The effect of fruit trait with seed traits were also deliberated using Karl Pearson's correlation. Principal component analysis (PCA), a multivariate statistical tool was used for elucidating the relative variation of different fruit and seed traits for estimating the total variability using Originpro 9.0 (Origin Lab company, Northampton, MA, USA) software. In principal component analysis, morphological data sets of different units were normalised with a mean of 0

Table 1. Geographical coordinates (latitude, longitude), altitude and rainfall of study area

Sample site	Latitude	Longitude	Altitude (msl)	Mean annual rainfall (MAR) (mm)
Honnavar	14º15'33.8"	74º26'12.1"	4	4613
Karwar	14º41'0.65"	74º27'21.6"	19	4293
Udupi	13º18'18.8"	74º45'47.0"	13	3609
Bhatkal	13º59'43.3"	74º32'55.2"	26	4569
Mangalore	12º51'11.7"	74º51'24.3"	32	3382
Belthangady	13°00'34.6"	75º11'37.1"	77	4383
Puttur	12º46'53.9"	75º11'34.7"	79	3609
Kukke	12º40'10.29"	75º36'36.2"	133	1122
Charmady	13º01'55.1"	75º21'55.4"	125	4383
Gundya	12º50'40.9"	75º32'52.4"	143	3609
Madikeri	12º29'25.7"	75º34'49.4"	229	3556
Dandeli	15º05'20.2"	74º31'22.1"	448	2064
Yellapur	14º50'0.38"	74º50'56.6"	522	2205
Sirsi	14º42'39.2"	74º44'35.5"	514	3194
Londa	15º32'40.9"	74º28'58.5"	687	2394
Dharwad	15º31'48.3"	74º56'40.0"	702	1060

and variance of 1. Hierarchical cluster analysis was used for constructing dendrogram between the population and was carried out using group linkage method according to squared Euclidean distance in Originpro 9.0 Software.

RESULTS AND DISCUSSION

Fruit and seed characteristics: The minimum, maximum, mean, median and data distribution of the morphological parameters among the population is represented in Figure 2 (a-h). There was significant variation in fruit and seed traits between the different population. The samples collected from population of Gundya shown highest fruit and seed characters except in RT and RW which was highest in population of Belthangady. The mean FW varied from 15.71

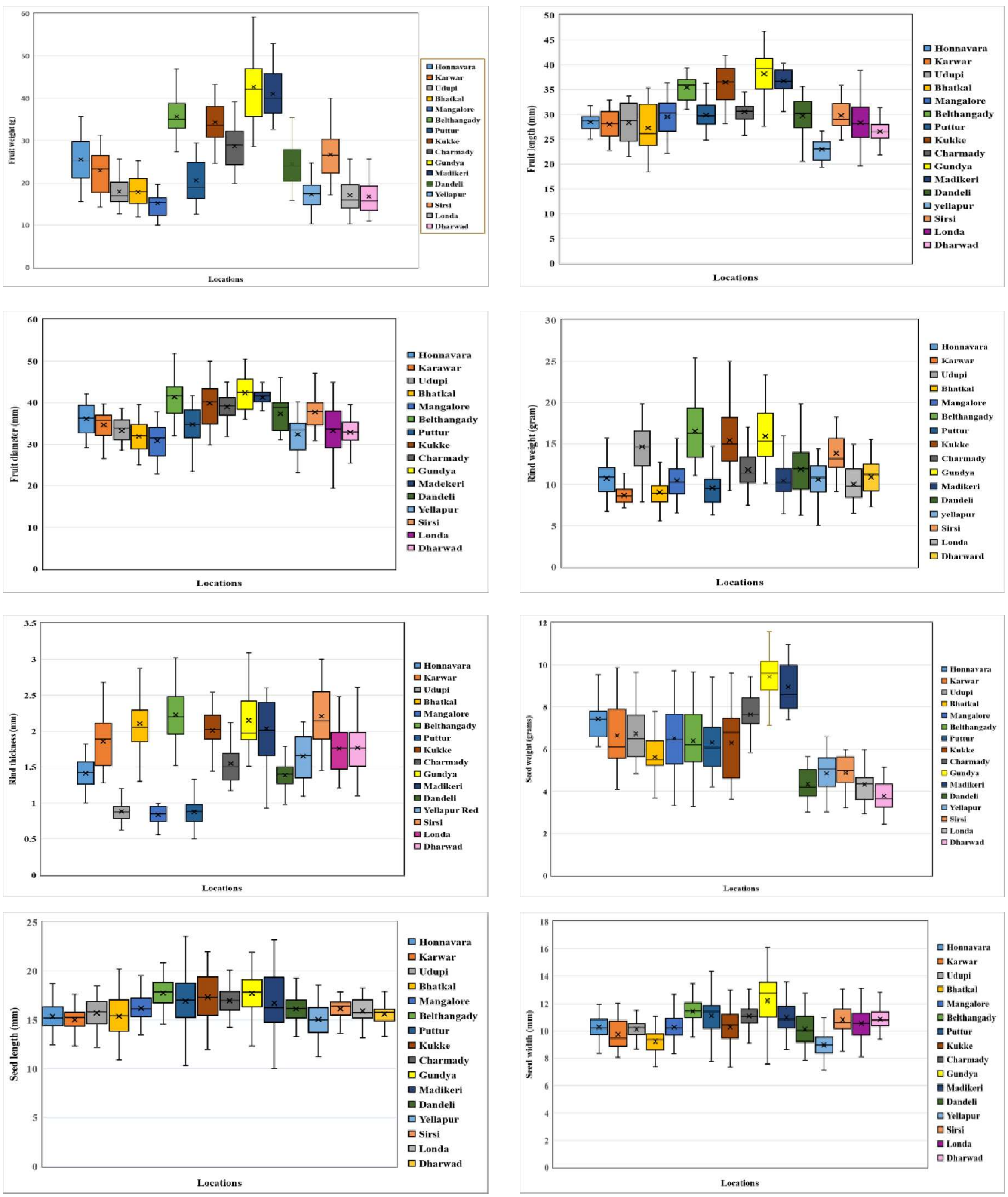

Fig. 2. The variation in morphological characters namely FW (a), FL (b), FD (c), RW (d), RT(e), SW (f), SL (g), SD (h) in *Garcinia indica* among 16 populations

to 41.50 g, with highest fruit weight in samples collected from Gundya region. The fruit length and fruit diameter of *Garcinia indica* was maximum in populations of Gundya followed by Madikeri. The highest fruit length to diameter was also observed in populations of Gundya (0.90) followed by Madikeri. The maximum RW and RT was observed in population of Belthangady with an average of 16.69 g and 2.38 mm respectively. Followed by the population Belthangady, the RW and RT were observed to be the most for the samples collected from Gundya and Kukke region. The highest total seed weight was observed in population of Gundaya (9.30 g) whereas, the least total seed weight was observed in population of Dandeli (3.50 g). The population of Gundya was also observed highest seed length (17.70 mm) and seed diameter (12.24 mm) (Table 2). *G. indica* have vulnerable status according to IUCN (Gowthami et al. 2021) however, the seed and fruits have different industrial uses and are one of the important characters for selection (Baliga et al 2011). The existence of variation in *Garcinia indica* was reported in Western ghat region of Maharashtra, the fruit weight ranges from 25.4 g to 58.38 g, the maximum fruit length and width was 4.28cm to 4.75cm and the fresh fruit rind ranges from m 13.93 g to 35.2 g (Gawankar et al 2001). Similarly, in Goa the variation within the 264 accessions was reported in tree canopy shape, leaf shape, fruit, and seed characters. The fruit characters between 11 taluks shown that the maximum fruit weight was 47.60 g in natural conditions, the fruit rind was 0.48 cm, the fruit length and diameter ranges from 1.19 to 4.36 cm and 1.80 to 5.51 cm in natural conditions (Priya devi et al 2013). Similar findings were also reported from study conducted in *Garcinia gummi-gutta* at Andaman Nicobar Islands by Bohra and Waman (2019), confirming the existence of morphological variation between the population.

Influence of locations and fruit traits: The FW, FL, FD, SL and RW had a positive correlation with longitude, but these are negatively correlated with latitude (Table 3). The significant r^2 were observed for FW, FL, FD, SL and RW with longitude i.e., 0.71, 0.75, 0.70, 0.83 and 0.50 respectively. Mean annual rainfall had a positive relation with fruit weight, seed weight, fruit length and fruit diameter, but a significant correlation was seen only with seed weight (r^2 =0.60). The altitude or elevation does not have any effect on the morphological variation in fruit and seed characters. It is negatively correlated with all the fruit and seed characters expect rind thickness, which was not significant. The correlation among fruit and seed characters were represented in Table 4. The FW is significantly correlated with FL, FD, SL, SD, SW, RW and RT. A significant positive correlation was observed for SW with FW, SL and SD.

Table 2. Relationship between geoclimatic factors and fruit traits

Morphological parameters		Geo-climatic parameters			
		Latitude	Longitude	Altitude	Mean Annual Rainfall (MAR)
Fruit characters	Fruit weight	-0.56	0.71*	-0.37	0.16
	Fruit length	-0.6	0.75*	-0.34	0.07
	Fruit diameter	-0.43	0.70*	-0.19	0.01
	Rind weight	-0.36	0.50*	-0.12	-0.11
	Rind thickness	0.14	0.27	0.24	-0.14
Seed characters	Seed weight	0.38	0.54*	-0.77	0.60*
	Seed length	-0.79	0.83*	-0.50	0.14
	Seed diameter	0.09	0.09	-0.09	-0.01

*Denotes values are significant at 95% confidence interval

Table 3. Relationship between fruit and seed traits

Morphological parameters	Fruit weight	Fruit length	Fruit diameter	Rind weight	Rind thickness	Seed weight	Seed length	Seed diameter
Fruit weight	1							
Fruit length	0.94*	1						
Fruit diameter	0.95*	0.59*	1					
Rind weight	0.57*	0.60*	0.59*	1				
Rind thickness	0.62*	0.54*	0.65*	0.50*	1			
Seed weight	0.54*	-0.23	-0.08	-0.36	-0.03	1		
Seed length	0.72*	0.71*	0.70*	0.32	0.18	0.59*	1	
Seed diameter	0.92*	0.04	-0.06	0.42	0.02	0.58*	0.06	1

*Denotes values are significant at 95% confidence interval

Similarly, RW was significantly correlated with RT, FW, FL and FD. Wang et al (2020) and Ji et al. (2016) reported that the variation in phenotypic character was largely influenced by local environmental characters. The variation caused by climatic variation may led to adaptive evolution and phenotypic plasticity among the species (Aitken et al 2008, Hoffmann and Sgro 2011, Alberto et al 2013, Franks et al 2014). Topographical (latitude, longitude, altitude) or abiotic factors (rainfall, temperature, precipitation) stimulate the variation in morphological traits of tree species between the population (Liu et al 2011, Soper Gorden et al 2016, Jesus et al 2017, Mojzes et al 2018, Wu et al 2018).

Principal component analysis (PCA) and cluster analysis: The PCA is a statistical tool used to elucidate total variation caused by different morphological traits such as seed and fruit characters. The results of the multivariate analysis for principal component revealed that maximum amount of variation was for PC1, PC2 and PC3 constituting total of 88.27%, which was evident in screen plot (Fig. 3). The PC1, PC2 and PC3 causing 64.56, 13.47 and 10.23% respectively accounted for major variation. The PC3 point in screen plot is known as elbow point after which the variation is slight. The component scores of variables contributing to coefficient of PC were depicted in Table. 5. The major morphological traits causing variation in PC1 are FW, FL and FD, whereas, PC2 is contributed by RT and SW contributes to PC3. The biplot represented in interpret the relationship among different fruit and characters and 16 different populations using extracted eigen value of coefficient of PC1 and PC2 (Fig. 4).

The hierarchical dendrogram between different population based on Euclidean distance from Principal component 1 and 2. The dendrogram forms two main clusters between 16 different populations of *G. indica* based on the variation in morphological characters. The cluster I consist of Honnavar, Karwar, Bhatkal, Yellapur, Dandeli, Londa, Dharwad, Udupi, Mangalore, Puttur whereas, the cluster II consist of Belthangady, Kukke, Charmady, Sirsi, Gundya and Madikeri. The cluster II populations had showed superior various morphological traits when compared with cluster I based on the study. From the dendrogram it was evident the

Table 4. Component scores of the first three factors of the total variance

Variables	Component 1	Component 2	Component 3
Fruit weight	0.41	0.03	0.31
Fruit length	0.41	-0.02	-0.06
Fruit diameter	0.40	0.13	0.17
Rind Weight	0.31	0.22	-0.48
Rind thickness	0.22	0.72	0.33
Seed weight	0.25	-0.54	0.55
Seed length	0.38	-0.15	-0.36
Seed diameter	0.34	-0.27	-0.28

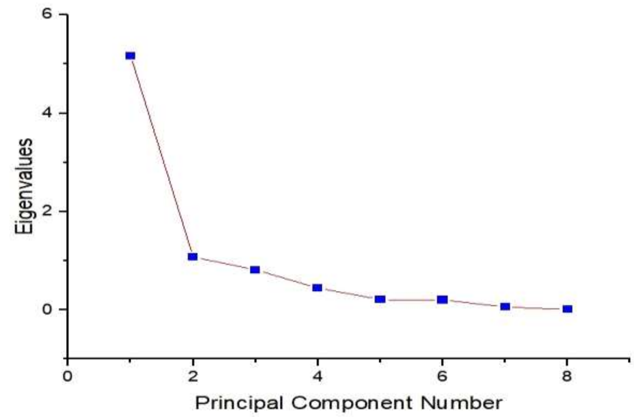

Fig. 3. Screen plot for PCA of different fruit and seed variable

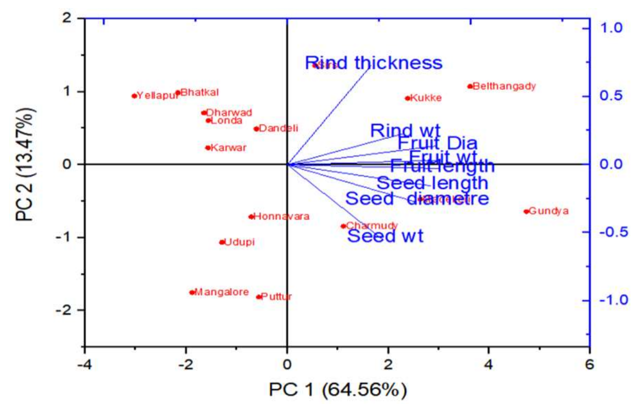

Fig. 4. Two dimensional bi plot of PCA of 16 different populations of *G. indica*

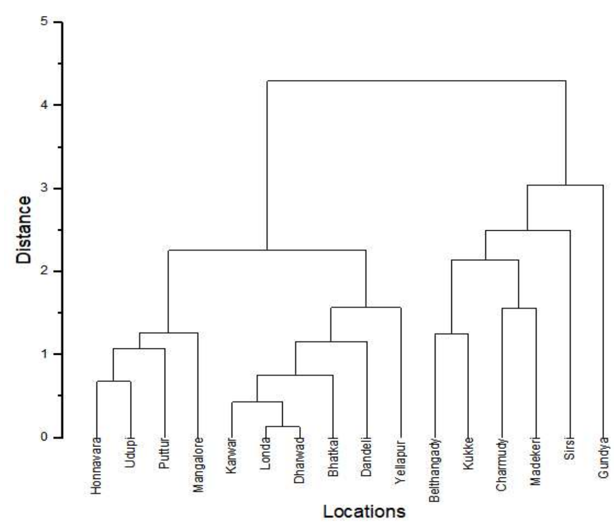

Fig. 5. Hierarchical dendrogram between different populations of *G. indica*

least representative was in population of Gundya whereas the most representative population was Dandeli. Despite, the Gundya population was distinct because FW, FL, FD, SW, SL and SD character was higher than other populations.

CONCLUSIONS

The study was conducted to understand the variation present in morphological characters of *Garcinia indica* in 16 different populations of Karnataka. Based on Karl Pearson correlation study it is evident that the morphological parameters had a strong relationship with longitudinal geoclimatic parameters. The PCA and dendrogram analysis revealed that the population in cluster II have distinguished morphological characters. The study also revealed that samples collected from population of Gundya showed a prominent character in fruit and seed character except rind thickness and rind weight which was highest in the samples collected from Belthangady. Thus, the variations between the population provide us an abundant source for breeding material based on the need of industry.

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Assessment of Groundwater Quality for Irrigation Purposes using Hydrochemical Diagrams and Chemical Indexes

S. El hasini, H. Dakkak[1], M. Azzouzi, H. Iaaich[1], A. Douaik[1], O. Iben Halima[1] H. Sehlaoui[2] and A. Zouahri[1]

Laboratory of Spectroscopy, Molecular Modeling, Materials, Nanomaterials, Water, and Environment, Department of Chemistry, Faculty of Sciences, BP 1014, Rabat-Morocco
[1] National Institute of Agricultural Research (INRA), Regional Center of Rabat Avenue Mohamed Belarbi Alaoui B.P 6356, Morocco
[2]Laboratory of Biology, Mycology and Environment, Department of Biology, Faculty of Sciences, BP 1014, Rabat-Morocco
E-mail: elhasinisoukaina3@gmail.com

Abstract: This work deals with groundwater quality under the effect of aridity. The case study is the region surrounding a sabkha named Sed Masjoun, located in the province of Kalâat des Sraghna, central Morocco. For this purpose, 67 hung-dug and borehole wells were investigated in the dry seasons in summer. Water samples were analyzed for chemical characteristics such as electrical conductivity, sodium absorption ratio, and sodium percentage. The results were also exploited using statistical analyses, hydrochemical diagrams, distribution maps and chemical indexes. The groundwater was severely affected by salinity but minorly by sodicity. The widespread salt was halite. The suitability of water for irrigation was also evaluated through several indexes. It follows that groundwaters are hard in nature, and have high sodium hazard, but no permeability problem was recorded. The concentration of nitrate and chloride were above the acceptable limit for irrigation in most wells, as shown by the dataset and the distribution map. In such a situation, any strategy promoting best management practices of irrigation and soil management, is highly encouraged to ensure sustainable use of these resources in the context of the severe aridity in Sed Masjoun area.

Keywords: Groundwater, Sed Masjoun, Quality, Salinity, Assessment

Morocco is basically an arid and desert country (93% of the global area) (Debbarh and Badraoui 2002) and rainfall is irregular, either in time or space. The average annual rainfall rate over the entire territory is fated to 150 billion m^3 unevenly distributed and is only 15% of the total area acquires nearly 50% of the precipitations. The northern mountainous areas receive more than 1000 mm, while less than 300 mm is shared annually between the basins of Tensift, Souss-Massa, and Moulouya, and the areas of South Atlas and Sahara (Debbarh and Badraoui 2002, Dahan 2012). This condition makes irrigation an instrumental imperative in the agricultural sector. Effectively, since independence, the Moroccan government has adopted an irrigation policy, as one of the hydro-agricultural management strategies. The 45% of agricultural production comes from irrigated area (13% of the total cultivated areas) (Debbarh and Badraoui 2002). However, excessive and uncontrolled irrigation water use inevitably leads to soil degradation, mainly to salinization and sodification risks (Kielen and Tanji 2002). Among the 350000-ha affected by salinity in Morocco), irrigated land occupies about 160000 ha (Badraoui 2006). The Bahira plain, the subject area of this study, represents the third affected area by salinization, with an area of 21000 ha, after Low Moulouya and Haouz basins. Irrigation water with poor quality, especially salt-rich waters, can bind sodium to the soil adsorbent medium, destroying its structure by dispersion of clays, leading to the loss of permeability, which affects plant growth. Moreover, the accumulation of chemical constituents of polluted waters in surface soils constitutes a serious threat to human health by the contaminating food, drinking water, and air. As a consequence of agriculturally degraded lands due to irrigation water in Morocco, the cost corresponds to 940 Million MAD annually (Croitoru and Sarraf 2010). Irrigated agriculture in the world depends mainly on groundwater resources (Foster and Shah 2012), especially in arid regions where surface water resources are restricted. In this regard, assessing and monitoring groundwater resources have become a priority worldwide. Numerous types of research have been performed in many basins around the world (Moujabber et al 2006, Baghvand et al 2010, Gouaidia et al 2013, Gautam et al 2015 and Abu-alnaeem et al 2018). Groundwater resources present an important legacy in Morocco. However, intensive use and strong evaporation in arid and semi-arid regions pose qualitative and quantitative problems, such as the degradation and the scarcity of freshwater resources. The Bahira plain is considered one of the most representative aquifers of this situation in Morocco. The arid climate and the succession of drought years lead consequently to the overuse of groundwater resources by the rural population, agricultural and domestic activities. This work's objective is to evaluate groundwater quality around the sabkha of Sed Masjoun and its suitability for irrigation.

MATERIAL AND METHODS

Study area: This study concerns the area surrounding the Sabkha of Sed Masjoun, (also named Sed El Mejnoun, Sed El Mesjoune, and Sed El Majnoon in previous studies), located in the central Bahira, and corresponded to the lowest altitude (404 m) of the great Bahira. Sed Masjoun is a seasonal lake extending over 32 Km^2 (El Mokhtar et al 2012). The Bahira plain is a vast central Morocco area 35 km north of Marrakech. It occupies a surface of about 5000 Km^2, bounded by the wavy uplands of Rhamna in the north, the mountain series of Jbilet in the south, the alluvial cone of Oued Tassaoute in the east and the Gantour uplands in the west, (Karroum et al 2014) (Fig. 1 , 2).

The water runoff coming from the surrounding elevations, mainly Jbilet (estimated at 33 Mm^3 / year) (Tensift Watershed Agency (ABHT)), is the main component of the natural recharge of the groundwater in the Plain and also the origin of the submersion during the winter of the depressions of Sed El Mesjoune at the central Bahira and Zima at the western Bahira. These two sites are considered evaporative machines during hot seasons, which salinize the water table. Under the plain of Bahira, waters circulate in the recent formations of Plio-Quaternary and Lutetian limestones (unconfined aquifer) and in the Upper Cretaceous (confined aquifer). The climate is arid to semi-arid with Saharan influences, the rainfall is low, irregular, and random, with a marked decline over the years (250 and 200 mm/an in 1995 and 2014, respectively) (Karroum et al 2014). Temperatures are high in winter and low in summer (48 and -3,6° C as maximum and minimum, respectively). The water balance is negative, the destocking of water is -4 Mm^3 due to the weak infiltration, the increase of the agricultural levies, and the evaporation. This last reached 2618,7 mm in 2014 (Karroum et al 2014). Cereals (barley, soft wheat, durum wheat, maize) and fallow land make up more than 90% of the utilized agricultural area. Fruit trees and other crops are exclusively present in the irrigated area.

Methodology: To evaluate groundwater quality and its suitability for irrigation in the area surrounding Sed Masjoun dry lake, a field survey was conducted during the dry season in June, July, and August, through the investigation of 67 wells, including boreholes and hand-dug wells, with depths ranging from 25 to 450 m. The levy was performed after the identification of sampling points using of a topographic map and GPS receiver (Fig. 3).

Before taking the samples, the well water pumps were opened sufficiently so that the sample became representative of the concerned well. Groundwater samples were placed into pre-conditioned bottles cleaned previously with nitric acid, washed afterward with distilled water, and rinsed finally by the water sample. In the field, samples were analyzed for some of their physicochemical parameters, such as electrical conductivity (EC), pH, temperature, and piezometric level, using portable conductivity, pH meter, a thermometer, and a level probe, respectively. Samples were kept airtight, over ice in an icebox, and transported to the laboratory for analysis. The physicochemical analyses, including ionic balance, pH and EC, were carried out in the Laboratory of Soil Chemistry at the National Institute of Agricultural Research (INRA) of Rabat (Table 1). The results were expressed in milliequivalent per liter (meq/l) except NO_3^- and NH_4^+ (ppm), pH, and EC (dS/m).

Statistical analysis: Data measured and calculated were submitted to statistical analysis using the SPSS software. Distribution maps were generated using the IDW spatial interpolation using ArcGIS. Water quality diagrams were drawn using Diagrammes - hydrochemistry software, 4.0 version (Simler 2007).

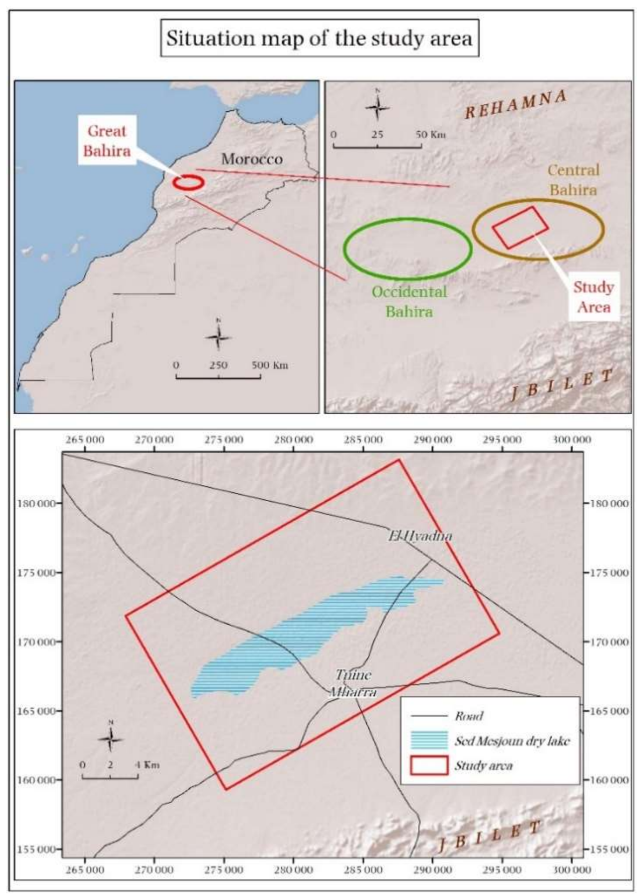

Fig. 1. Site map of the study area

RESULTS AND DISCUSSION

Salinity and chemical composition: Electrical conductivity (EC) is one of the most important criteria to assess irrigation water quality. An excessive amount of salts in irrigated soil adversely affects soil permeability, bulk density, and structural stability (Tejada and Gonzalez 2005), ultimately impacting plant growth. The groundwater in the surrounding area of Sed Masjoun show that EC varies between 1.3 and 77.2 dS/m with an average of 5.7 dS/m. The 61% of the wells violated the permissible limit for irrigation use (Wilcox 1955), with 37 and 24% of the wells doubtful and not suitable for irrigation, respectively. In contrast, only 39% of the wells were classified as permissible. Consequently, none of the samples belonged to the good or the excellent categories. The majority of wells with no suitable water are located on the south side of Sed Masjoun, especially toward the southeast part except wells P67 and P65, which have questionable water quality. This category also includes some wells on the north side (P27) in the sabkha borders and certain wells in the eastern part (P43, P44, P49 and P50). The area with doubtful groundwater to use is extended in the north and the southwest, whereas the water with permissible use occupies some locations in the far north, the north-south, and the southwest areas (Fig. 4).

Pearson correlation (Table 5) was significant between EC and Cl^- (0.83), Na^+ (0.85), SO_4^{2-} (0.76), and Ca^{2+} (0.58), which highlights the responsibility of these three elements for water salinity. Cl^- showed a strong correlation with Na^+ (0.99), presuming the existence of the halite (NaCl) in the water. The correlation between Mg^{2+} and SO_4^{2-} (0.81), indicate existence of specific quantities of magnesium sulfate ($MgSO_4$) precipitant. Considering the anionic and the cationic groups separately, a significant correlation was between Cl^- and SO_4^{2-} (0.74) in part and between Ca^{2+} and Mg^{2+} (0.50) in the other part, indicating the probable common source of every two ions.

The skewness coefficients (Table 3) show that all parameters were positively skewed (especially Na^+(7,2) and Cl^- (6,9)), except for pH (0,0) and HCO_3^{2-} (0,5), where the data was pretty symmetrical. The kurtosis values were high for the majority of parameters, which means the profusion of outliers, this was more pronounced for Na^+ (55.1) and Cl^- (52.2) where the presence of some abnormally high (circles) or some extreme (stars) values (Fig. 5).

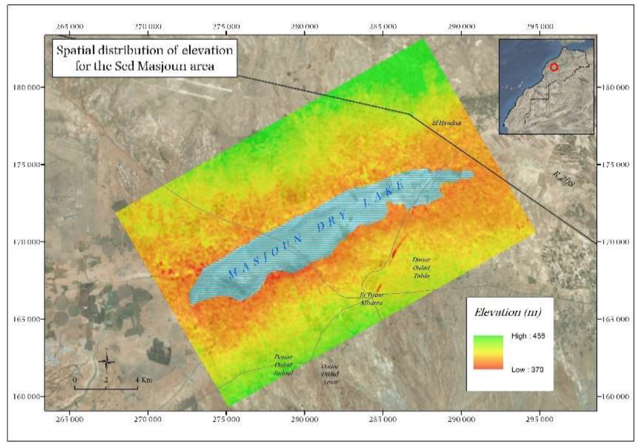

Fig. 2. Spatial distribution of elevation for the studied area

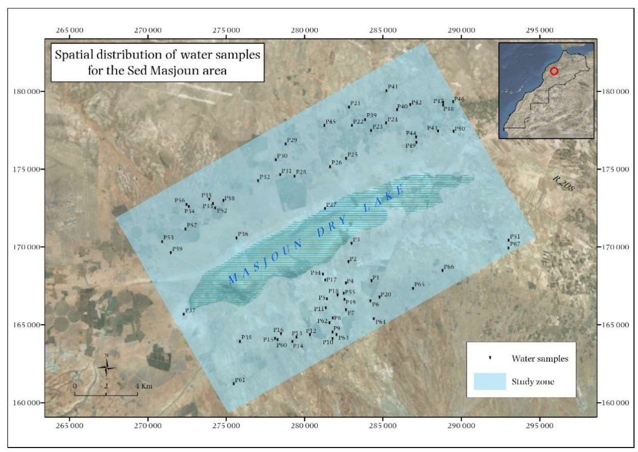

Fig. 3. Spatial distribution of water samples for the Sed Masjoun area

Table 1. Measured physicochemical parameters and the corresponding methodology

Parameters	Method
Electrical conductivity (EC)	Orion laboratory conductivity meter, model 162
pH	Metrohm pH meter, model 691
Sodium and potassium (Na^+ and K^+)	Flame photometry (Jackson 1967), photometer Jenway, PFP7 model
Calcium and magnesium (Ca^{2+} and Mg^{2+})	Complexometric titration with EDTA (Page 1982)
Carbonate and bicarbonate (CO_3^{2-} and HCO_3^-)	Acid–base titration (Rodier 1984).
Sulfate (SO_4^{2-})	Nephelometric method of precipitating sulfates as barium sulfate in HCl medium (Rodier 1984)
Chlorides (Cl^-)	Mohr's method (Rodier 1984)
Nitrates and amonium (NO_3^- and NH_4^+)	Steam distillation (Büchi unit B-323 model)

The abundance, in average, was in the order $Na^{+}>Mg^{2+}>Ca^{2+}>NH^{4+}>K^{+}$ for the cations and $NO_3^{-}>Cl^{-}>SO_4^{2-}>HCO_3^{-}>CO_3^{-}$ for the anions. The anion classification showed no difference compared with some studies in countries with the same or similar climatic conditions. However, cation abundance was slightly dissimilar. In fact, by studying 60 samples of groundwater taken from the Hail region in Saudi Arabia, Abdel-Satar et al (2017) found that Ca^{2+}, followed by Na^{+}, constitute the predominant cations, while Ebrahimi et al (2016) pointed out the prevalence of Na^{+} followed by Ca^{2+}, after analyzing 15 samples of groundwater collected from the Damghan basin in Iran. Considering variability, Cl^{-} is the most variable concerning anions (CV = 255%), with values ranging from 6.8 to 880 meq/l with a mean of 43.2. Na^{+} shows the highest variability among cations, with values ranging from 4.3 to 1036, and a mean value of 43.4 meq/l. High variability was also for K^{+} (CV=260%) with values ranging between 0.0084 and 16.4 meq/l and a mean of 0.93. However, it constitutes the minor element that did not play any role in groundwater hydrochemistry and didn't show any correlation with the other elements.

The Piper trilinear diagram (Piper 1944) was used to bring out the chemical relationships among water samples, and

Table 2. Calculated indices and their corresponding equations

Indice	Equation
Sodium percentage (Na %)	$Na\% = 100 * \frac{Na}{Ca+Mg+Na+K}$
Sodium adsorption ratio (SAR)	$SAR = \frac{Na}{\frac{Ca+Mg}{2}}$
Residual sodium carbonate (RSC)	RSC= CO3+ HCO3 - (Ca+ Mg)
Permeability index(PI)	$PI = 100 * \frac{Na+\sqrt{HCO3}}{Ca+Mg+Na}$
Kelly's ratio (KR)	$KR = \frac{Na}{Ca+Mg}$
Total hardness (TH)	TH = 50 * (Ca+ Mg)
Magnesium hazard(MR)	$MR = 100 * \frac{Mg}{Ca+Mg}$

Table 4. Descriptive statistics of the calculated indices

Index	Maximum	Minimum	Mean	SD	CV%
% Na	96,1	24,7	51,5	17,4	33,9
PI	95,4	25,4	52,1	17,0	32,7
RSC	0 ,0	-123,8	-15,4	23,1	-150,2
KR	24,7	0,3	2,1	3,7	177,2
TH	6316,0	270,0	1121,2	1192,0	106,3
MR	90,3	16,9	54,8	17,5	31,9
SAR	142,3	1,4	11,6	22,8	196,3

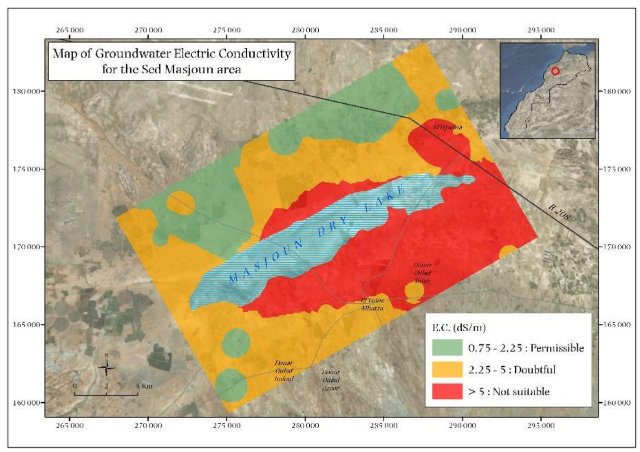

Fig. 4. Distribution map of groundwater electrical conductivity for the Sed Masjoun area

Table 3. Descriptive statistics of water level and major ion chemistry in the study area

Parameter	Mean	Median	SD	CV (%)	Min	Max	Skewness	Kurtosis
Well depth (m)	112,1	104,0	79,4	7,6	25	450	1,9	5,7
pH	7,7	7,8	0,4	4,7	7,0	8,5	0,0	-0,9
EC (dS/m)	5,7	2,7	10,9	193,5	1,3	77,2	5,2	30,0
NH^{4+} (ppm)	6,1	3,6	5,7	93,4	0,0	18,0	1,0	-0,2
NO_3^{-} (ppm)	49,1	49,6	34,1	69,5	3,9	161,2	1,1	1,9
Cl^{-} (meq/l)	43,2	20,4	110,2	254,8	6,8	880,0	6,9	52,2
CO_3^{2-} (meq/l)	0,5	0,0	0,7	156,8	0,0	2,5	1,5	1,4
HCO_3^{-} (meq/l)	6,6	5,5	3,9	59,6	1,0	15,0	0,5	-1,1
SO_4^{2-} (meq/l)	16,9	3,3	40,9	242,8	0,1	268,9	4,5	23,2
Ca^{2+} (meq/l)	8,1	5,6	6,9	84,9	2,6	43,4	2,9	11,1
Mg^{2+} (meq/l)	14,3	6,4	19,7	137,1	1,4	111,3	3,1	10,8
Na^{+} (meq/l)	43,4	15,6	129,0	297,4	4,3	1036,3	7,2	55,1
K^{+} (meq/l)	0,9	0,1	2,4	260,4	0,01	16,4	4,9	27,2

define hydrochemical facies. Most facies are sodium chloride, calcium chloride and magnesium (Fig. 6). This finding corroborates those reported by El Mokhtar et al (2012). El Mokhtar et al (2012) explained the magnesian component by a cation exchange between groundwater and the clay complex, rich in Mg, well-developed in plioquaternary deposits when flow gradients are low.

To sum up, groundwater in the basin surrounding the sabkha of Sed Masjoun generally has high saline charge; many wells are brackish to saline. The analyzes were carried out by El Mokhtar et al (2012) using stable isotopes of Oxygen 18 (^{18}O) and Deuterium (^{2}H) after sampling at different depths and salinity levels. The results showed the effect of evaporation on the salinization process, whether in the surface or deep water. This phenomenon occurs habitually by water from rain and runoff that undergo evaporation before and during infiltration.

Sodicity: The sodicity measures the quantity of sodium in water. Sodium hazard can be quantified by sodium abundance with regard to either Ca^{2+} and Mg^{2+} contents using SAR or to the number of total cations existing in water using Na%. These two indices are both connected with EC by two diagrams.

Sodium percent: The percentage of sodium (Na%) with respect to the other cations expressed by Na% is defined by equation 1 (Table 2). An excessive amount of sodium in irrigation water may affect soil permeability and cause osmotic effects leading to a restriction in agricultural yields (Raju 2007). This parameter ranged between 25 and 96% in the study area, with an average of 51%. However, none of the samples belongs to the excellent category (Table 6).

The combined consideration of Na% and EC by Wilcox diagram (Fig. 7) showed that 27 wells out of the 67 (40.3%) fell in the range of the excellent category, 25.4,26.9 and 7.4 % in the good, the permissible limit and the doubtful categories, respectively.

Sodium absorption ratio: The Sodium Absorption Ratio (SAR) is an important index to evaluate the suitability of water for irrigation purposes. This parameter is defined by equation 2 (Table 2). A high concentration of Na^{+} in the soil-soluble state may replace most frequently Ca^{2+} and Mg^{2+} exchangeable cations in the absorbing complex. This displacement leads to the dispersion of sodium- saturated soil particles and then the destruction of soil structure. The average SAR of groundwater of the study area was 11. 6 (meq$^{1/2}$. l$^{-1/2}$), varying between 1.4 to 142.4 (Table 4) Majority (81%) of wells present low SAR values, while 9, 4, and 6% show medium, high and very high rates, respectively. High SAR values are concentrated in the easternmost and the south sides, distributed gradually toward the dry lake (Fig. 8).

The evaluation of EC and SAR together by plotting the obtained results graphically on the diagram proposed by Richards (1954) (Figure 9) reveals that the majority of samples fall in C3S1, C2S1, and C3S2 classes, meaning their good to moderate quality for irrigation use.

Residual sodium carbonate: Residual sodium carbonate *(*RSC), calculated by equation 3 (Table 2), is another index to evaluate sodium hazard. This is supported by the fact that sodium amount in water with a high level of bicarbonates

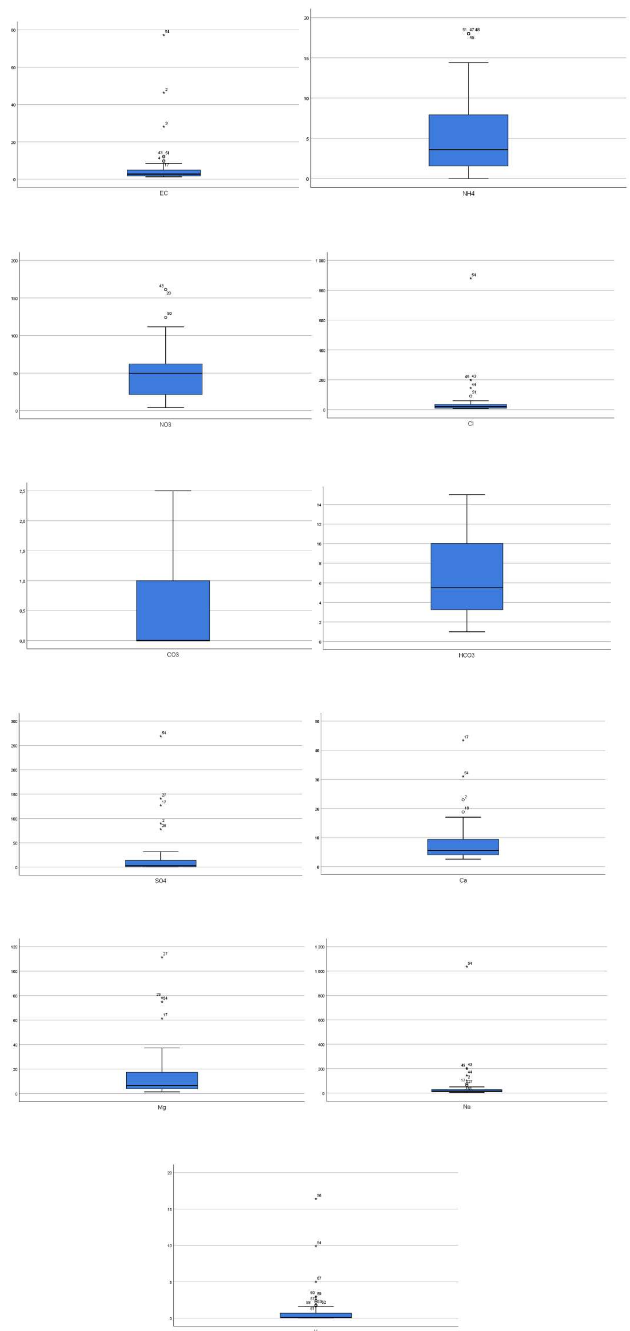

Fig. 5. Boxplots of the groundwater measured parameters

increases in the $NaHCO_3$ state, leading, as a consequence, to the precipitation of the divalent cations of Ca^{2+} and Mg^{2+} (Reddy 2013). The presence of carbonate and bicarbonate in groundwater is mainly owed to the dissolution of carbonate weathering and carbonic acid in the aquifers (Kumar et al 2009). All water samples had negative RSC values except one with a null value, which is the maximum value, while -15.39 and -123.82 are the mean and the minimum values, respectively. The negative values reveal that the level of carbonate and bicarbonate in water were lower than calcium and magnesium levels, which implies the nonexistence of a residual amount of carbonate to react with sodium and then intensify the alkali hazard in the soil.

pH and piezometric level: The pH had the lowest variation (CV of 4.7%), with values ranging from 7.0 to 8.5 and an average value of 7.8. The distribution map of pH (Fig. 10) indicates that most wells in the area around Sed Masjoun had alkaline pH, except for some sporadic spots, where the pH is slightly alkaline. This finding is also asserted by the Spearman correlation, where a strong correlation was found between pH and CO_3^{2-} (0.72), rather than NH_4^+ (0.54).

The piezometric level in the study area ranged between 268 and 408 m, with 360 m as the mean value. The distribution map of the piezometric level (Fig. 11) showed relatively lower values in the northeast side of Sed Masjoun, especially in the easternmost part. In contrast the high values were recorded on the south side, more strongly toward the middle part.

Indices of Water Suitability for Irrigation

Permeability index: Permeability problems are ascribed to the joint effect of irrigation water prolongedly remaining, and the content of sodium, calcium, magnesium, and bicarbonate in soil). The permeability index (PI) is calculated using equation 4 (Table 2). This parameter varied in our case between 25.4 and 95.4%, with 52% as a mean value. According to Doneen (1966), 10% of wells were considered excellent for irrigation (PI>75%), and 90% were good (25<PI<75); however, none of the samples belonged to the not favorable category (PI<25).

Magnesium ratio: Excessive amounts of magnesium in irrigation water can deteriorate soil quality, leading to reduced crop yields. The magnesium hazard is evaluated using the magnesium ratio (MR), calculated according to

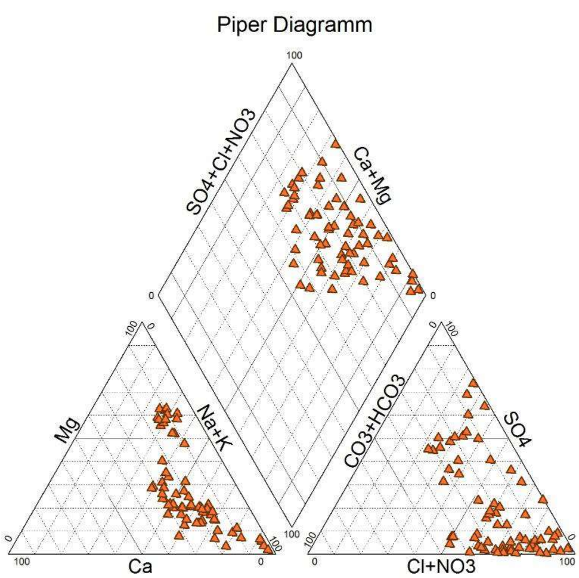

Fig. 6. Groundwater samples plotted in the Piper-Trilinear diagram

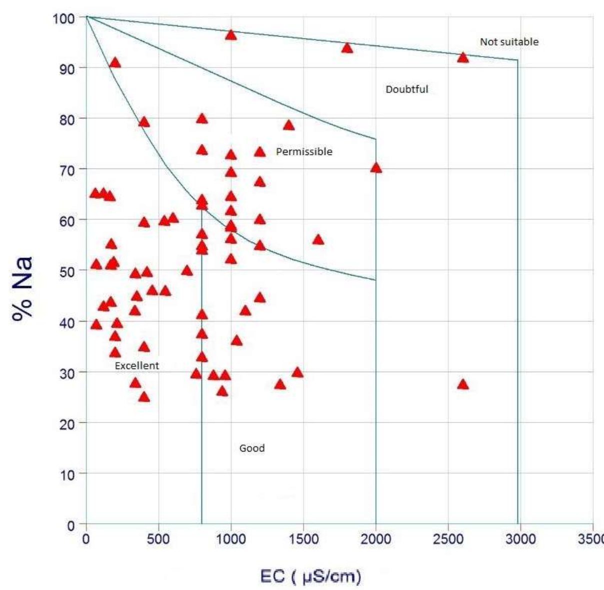

Fig. 7. Suitability of groundwaters for irrigation based on Wilcox diagram

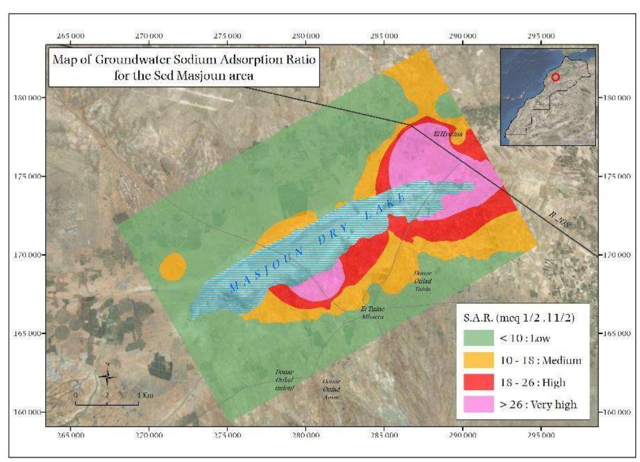

Fig. 8. Distribution map of SAR of groundwater in the study area

equation 7 (Table 2). In the study area, MR varied from 17 to 90%, with a CV of 32% and a mean value of 55%. According to Raghuanth (1987), 43% of wells were classified, in the suitable category (MR<50%), while 57% were not suitable for irrigation (MR>50%).

Total hardness: Total hardness (TH), defined by the equation 6 (Table 2), is an index to evaluate the abundance of the divalent cations of Ca^{2+} and Mg^{2+} in water. TH varied widely (CV=106%) with values ranging from 270 to 6316 mg/l and an average value of 1121. Since all samples exceeded 180, groundwater of the studied area is considered very hard in nature, according to Durfor and Becker (1964), owing to the excessive presence of Ca^{2+}, Mg^{2+}, and HCO_3^- ions.

Kelley's ratio: Kelley's ratio (KR) is a parameter suggested by Kelley et al (1940) to report the ratio of sodium to the sum of calcium and magnesium, according to equation 5 (Table 2). This index should not exceed one. If so, water becomes not suitable for irrigation owing to the excessive amount of sodium. KR had a wide range between 0.3 and 24.5 with a mean value of 2.1, and 58% of wells were unsuitable for irrigation purposes.

Risk related to chloride: Chloride is a common element in

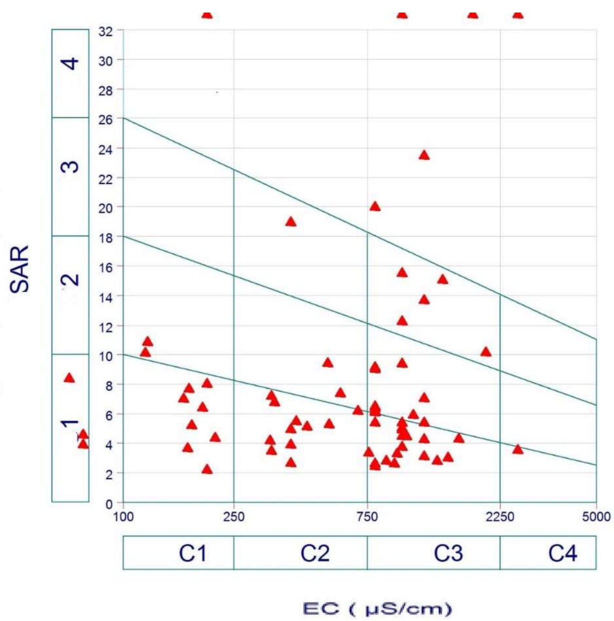

Fig. 9. USSL classification of groundwater in the study area

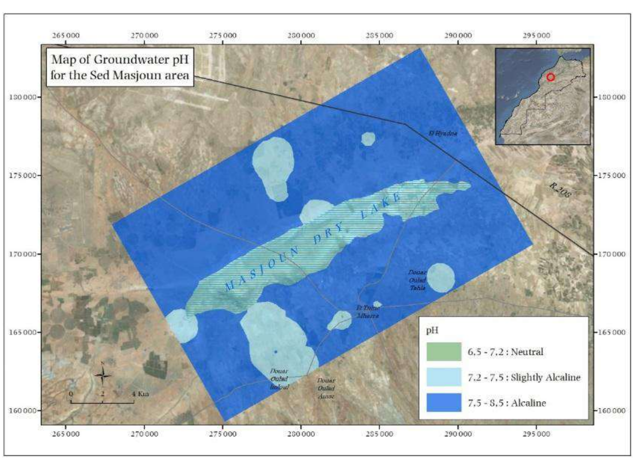

Fig. 10. Distribution map of pH in the study area

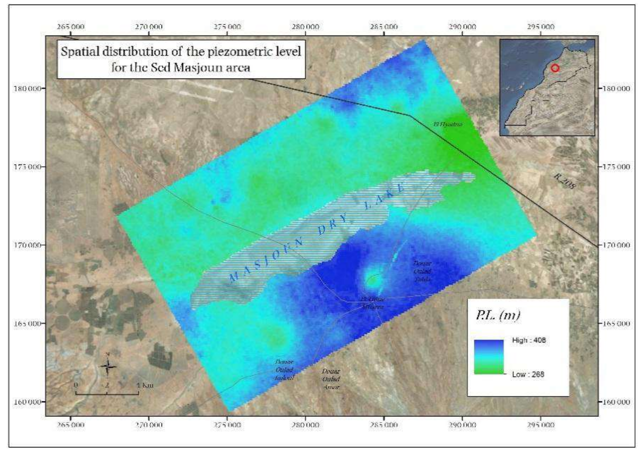

Fig. 11. Distribution map of piezometric level of the study area

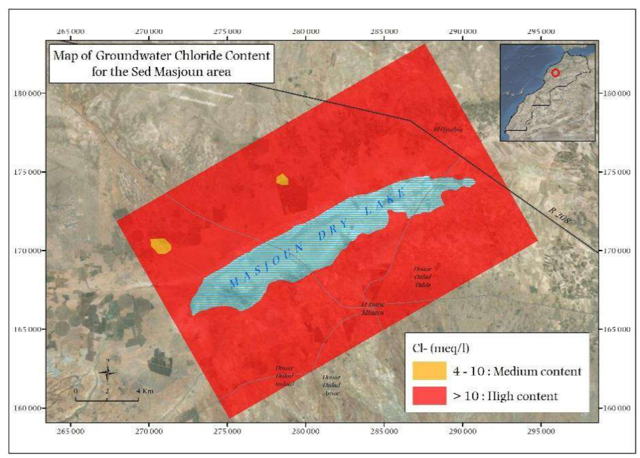

Fig. 12. Distribution map of chloride in the study area

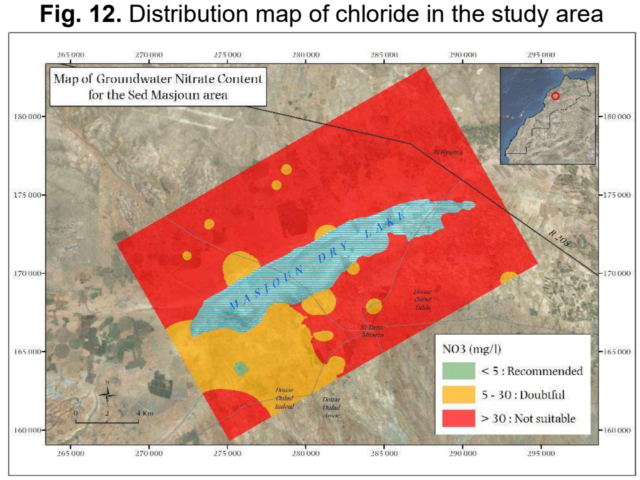

Fig. 13. Distribution map of nitrates in the study area

Table 5. Pearson correlation coefficients for groundwater parameters

Parameter	pH	CE	NH^{4+}	NO_3^-	Cl^-	CO_3^{2-}	HCO_3^-	SO_4^{2-}	Ca^{2+}	Mg^{2+}	Na^+	K^+
pH	1											
CE	0,01	1										
NH_4^+	0,54**	0,01	1									
NO_3^-	0,43**	-0,05	0,49**	1								
Cl^-	-0,02	0,83**	0,11	-0,02	1							
CO_3^{2-}	0,69**	0,01	0,58**	0,34**	-0,04	1						
HCO_3^-	-0,62**	-0,16	-0,28*	-0,19	-0,14	-0,42**	1					
SO_4^{2-}	-0,18	0,76**	-0,05	-0,13	0,74**	-0,18	0,03	1				
Ca^{2+}	-0,42**	0,58**	-0,30*	-0,30*	0,45**	-0,26*	0,18	0,66**	1			
Mg^{2+}	-0,36**	0,35**	-0,08	-0,11	0,38**	-0,26*	0,31*	0,81**	0,50**	1		
Na^+	-0,02	0,85**	0,10	-0,03	0,99**	-0,04	-0,13	0,78**	0,47**	0,41**	1	
K^+	0,08	0,32**	-0,10	-0,03	0,40**	-0,12	-0,32**	0,36**	0,09	0,09	0,41**	1

**. * Correlation is significant at the 0.01 0.05 level (bilateral)

Table 6. Groundwater classification based on percent Na in the study area according to Wilcox (1955)

Percent Na	Classification	Percentage of wells
<20	Excellent	0
20-40	Good	27
40-60	Permissible	46
60-80	Doubtful	21
>80	Unsuitable	6

all-natural water, generally in low concentrations (<10 meq/l). However, when the level increases, Cl^- may intoxicate plants by moving with soil water to accumulate in leaves since it is not absorbed by the soil (Hussain et al 2010). Data from the present study indicate the risk of chloride in the study area. All samples exceeded 4 meq/l, which means, according to Bigak and Nielsen (1972), a tendency to Cl^- toxicity, particularly for sensitive crops, while 85% of wells exceeded the high limit (10 meq/l) enormously. The range was between 6.8 and 880 meq/l, with mean 43.2 meq/l and CV of 255%. The risk related to chloride was also reported by Zouahri et al (2015), where 68% of wells in the Skhirat region (northwest) had Cl^- >10meq/l, while only 3% had Cl^- between 2 and 4. Based on the distribution map (Figure 12). There were only two small localities with $4<Cl^-<10$ meq/l situated in the north area of Sed Masjoun. Chlorides could come mainly from the percolation of irrigation return water through salty soils.

Risk related to nitrate: At varying levels, the nitrogen compounds NO_3^- and NH_4^+ were present in the groundwater samples. For NO_3^- the mean was 49.1, ranging from 3.9 to 161.2, while for NH_4^+ the mean was 6.1 mg/l with a range of 0 - 18 mg/l. However, the highly soluble form is the nitrate that constitutes an index of pollution when present at a high level (Prasad and Ramesh 1997). According to irrigation water quality guidelines (Ayers 1994), high nitrate concentration in water can affect sensitive crops, while the accepted level should not exceed 5 mg/l. When the concentration is between 5 and 30 mg/l, water should be used with specific caution. Regarding the present dataset, only 3 wells out of 67 (45%) respected the recommended level of use, while 43 wells (42,2%) violated the permissible limit by exceeding 30 mg/l. The rest of the samples should be used with moderate to slight restriction. This last category was concentrated essentially in the southwest part of Sed Masjoun (Figure 13). An excessive amount of NO_3^- in water can be ascribed mainly to the overuse of manure, nitrates fertilizers in agriculture, and the domestic wastes likely to convert into nitrates in soil (Kumar et al 2009).

CONCLUSIONS

Due to the arid climatic conditions of the Bahira region, the quality of available water resources is an additional problem to water scarcity. The quality of groundwater around Sed Masjoun, constituting the deepest point of the Bahira basin, depends on the basin geochemistry, evaporation and the mode of exploitation. The primary threat affecting a large amount of these waters is the salinity; that is, only 39% of samples were permissible for irrigation purposes. The salinity problem is mainly due to evaporation, accentuated especially in the southern area of Sed Masjoun, where the piezometric level is higher. The mean ions responsible for the salinity were the chlorides foremost, followed by sodium and calcium, while the most widespread salt was halite. According to Piper trilinear diagram, the dominant facies were sodium chloride, and calcium chloride and magnesium. Since the sodium presence is secondary, the sodicity problem was found to be minor. Regarding % Na, SAR, and RSC values, the majority of wells were grouped within the permissible and good categories to be used in irrigation. The suitability of groundwater for irrigation was equally assessed by using the permeability index, the magnesium ratio, the total hardness, and Kelly's index. It follows that no permeability problem was present; however, the hardness and the magnesium hazards struck a considerable quantity of water. High quantities of chlorides and nitrates severely affected groundwater in the Sed Masjoun area. Shallow water wells are considered the most easily attenable and contaminated. Therefore, many of them are definitively abandoned. In front of this situation, starting from the fact that groundwater resources constitute the major water provider in this area, and also considering the probable direct risk on human health, since the majority of this water is used equally for drinking and domestics purposes, continuous monitoring should be achieved regularly for sustainable development in this area.

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Estimation of Carbon Stock of the Trees in the Herbal Garden of Telangana State Medicinal Plant Board, Aziznagar, Hyderabad, India

D. Veeranjaneyullu, D. Md. Shareef, K. Jyothi, A. Sonibala Devi[1] and M. Suneela

ENVIS, Environment Protection Training and Research Institute, Hyderabad
[1]Telangana State Medicinal Plant Board, Hyderabad
E-mail: hanveerobu@gmail.com

Abstract: Carbon staock estimation studies were carried out during November 2019 of the tree species with ≥10-30cm Girth at Breast Height (GBH) and ≥31cm GBH at Herbal Garden of Telangana State Medicinal Plant Board (TSMPB). A total of 1.5-hectare (ha) area was covered 6.39% of the total geographic area of 23.47 ha. The study area is dominated by *Leucaena leucocephala*, *Phyllanthus emblica*, *Azadirachta indica*, *Pterocarpus santalinus, Gliricidia sepium*, *Aegle marmelos*, *Dalbergia sissoo*, *Terminalia arjuna*, *Albizia lebbeck* and *Bauhinia purpurea.* A total of 50 species were documented. Total standing crop biomass was estimated based on volume and specific gravity. Total carbon accumulation was estimated using carbon factor 0.47. A total of 326.388 Megagram **(**Mg tonnes) was accumulated in the sampling plots.

Keywords: Carbon staock, Crop biomass, Geographical area, Herbal garden, Telangana state

The Intergovernmental Panel on Climate Change (IPCC) recommends that actions be taken to limit the earth's temperature increase to no more than 2.0°C, and preferably to no more than 1.5°C above preindustrial levels. An increase of just 1.5 to 2.5ºC could spell extinction for 20-30% of all species, and above 3.5ºC, 40-70% of global species could be wiped out (IPCC, AR6-SCOP/2017). Forest vegetation contains over 350,000 teragrams (Tg) of carbon and plays a major role in the global carbon cycle. Estimation of above-ground biomass (AGB) is an essential aspect of studies of carbon sequestration, carbon stocks and to study the effect of deforestation on global carbon balance because it constitutes about 60% of total phytomass and is involved in the regulation of atmospheric carbon concentration (Ketterings et al 2001). Tree inventories are an efficient way of assessing forest carbon stocks and emissions to the atmosphere during deforestation (Chave et al 2004). Biomass represents the largest organic carbon pool in a mature tropical forest ecosystem. Despite its importance, it is treated as a poorly quantified stock, because changes in biomass are dramatically induced by gap dynamics and succession after natural and human-induced disturbances. Moreover, biomass shows wide variability within and between forest communities (Sarmiento et al 2005). The change in forest biomass has been considered a key characteristic of the forest ecosystem. Biomass variability can be explained by several factors like climate, topography, soil fertility, water supply, and wood density, distribution of tree species, tree functional type, and forest disturbance (Luizao et al 2004, Sicard et al 2006). At present, not much information is available on the estimates of carbon sequestration potential in urban areas. Hence, the study was conducted to estimate carbon stock of tree species at herbal garden, Aziz nagar, Hyderabad.

Study area: The study area is government land located at Aziznagar, Rajendranagar Mandal, Rangareddy district ($17^0$20'03.17" N to $78^0$21'07.58" E and $17^0$21'05.04" N to $78^0$21'05.04" E) of Telangana and measuring 23.47 ha of Telangana State Medicinal Plants Board (TSMPB), Health, Medical, and Family Welfare Department are developing the greenery and maintaining the area. In a portion of this area, more than 500 varieties of medicinal plants are conserved *ex-situ*, with the financial aid of the National Medicinal Plants Board (NMPB), Ministry of Ayush, Government of India and Government of Telangana under various schemes in different years. TSMPB maintains a model nursery in this study area to supply quality planting materials to farmers as well as for distributing medicinal plants to institutions, households, etc. Mother stock groves of 20 medicinal species have been raised in different years. Apart from these, a few trees of *Azadirachta indica, Hardwickia binata, Leucaena leucocephala* are existing in the area naturally. The species existing in the garden are utilised for propagation in model nursery. About 80 varieties of medicinal plants are being raised and maintained at any point of time to distribute to the people to create awareness of medicinal

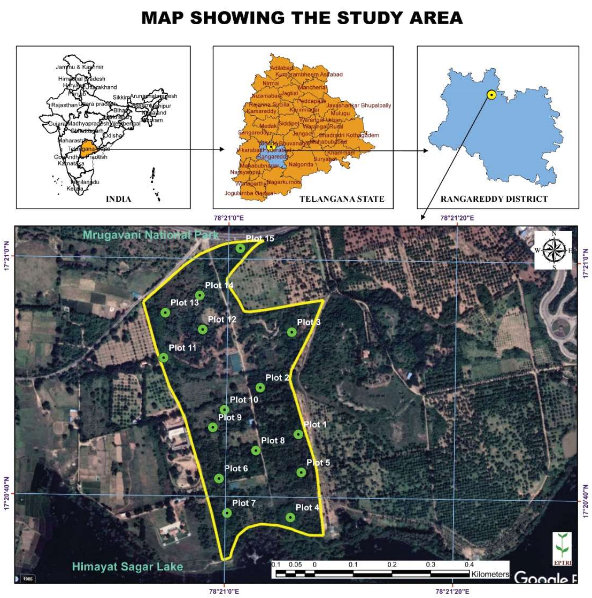

Fig. 1. Map of the study area

plants and implement various schemes like home herbal garden, school herbal garden.

MATERIAL AND METHODS

The primary data was collected from the study area in 15 randomly stratified sample sites as shown in the map. Thus, a total of 1.5 ha area was sampled covering 6.39% of the total 23.47 ha study area. A total of 15 sample sites of 31.62 m x 31.62 m size were identified based on Normalized Difference Vegetation Index (NDVI) values. The said methodology was a comprehensive format design of Vegetation Carbon Pool (VCP) Assessment by National Carbon Project, Indian Institute of Remote Sensing (IIRS) (Singh and Dadhwal, 2008) and the methodology was adopted for the current study.

The following data were collected from each plot

1. A stratified random sampling plot was laid with the size of 0.1 ha (31.62 m x 31.62 m)
2. In the sample plot all tree individuals of more than or equal to 10 cm GBH were documented by measuring girth at the height of 1.3 m.
3. The data for each species was classified based on GBH classes: ≥10-30cm, ≥31-60cm, ≥61-90cm, ≥91-120cm, ≥121-150cm, ≥151-180cm, ≥181-210cm, etc.

Primarily the team has documented the species available in the plots. All the plant taxa present in the sample plots were photographed during the field survey. The total number of individuals of all species together was also recorded. Each species was considered in categories, one between 10cm and 30cm girth class and the other above 30cm girth classes. The estimation of volume, biomass, and carbon stocks of trees in the study area was calculated in 15 stratified sample sites.

Basal area: Basal area of each tree was calculated by using the following standard formula:

dbh(cm) = gbh(cm) / π , dbh(m) = dbh(cm)/ 100, Radius(r) = dbh(m)/2. Basal Area ($m^2\ ha^{-1}$) = πr^2

Volume estimation: Volume of each tree of ≥10cm diameter on above was estimated using the selected volumetric equation developed and compiled by FSI (1996). The formula was selected based on the availability of equations developed by Forest Survey of India.

Criteria 1: Species specific volumetric equation of the same study area.

Criteria 2: If criteria one is not available the species specific volumetric equation of neighboring area of same phytogeographical zone were considered.

Criteria 3: If criteria one and two are not available the volumetric equation belongs to the same species or same genus of the same state or other states has been selected by checking equations of different regions and finally the appropriate equation was selected.

Criteria 4: If criteria one, two and three are not available for those species, the common equation of the same study area is selected.

Specific gravity: Specific gravity values of different species were selected from literature. These values are available for 75-80% of species. For stems with unknown specific gravity, the arithmetic mean of all known species was substituted and used in a particular sample plot.

The estimated volume was converted into biomass by multiplying with specific gravity .Biomass of all the trees was summed up to obtain biomass in a 0.1 ha area.

Biomass (tonnes) = Volume (m^3) × Specific Gravity

Below-ground biomass (BGB): In the present study, 20% of the above-ground biomass was considered as root biomass as per the procedure adopted by Achard et al (2002), Houghton et al (2001), Montagu et al (2002), Ramankutty et al (2007).

Total biomass (TB): Total biomass of each plot was calculated by the addition of total above ground biomass (TAGB) and below ground biomass (BGB).

Estimation of carbon stock : Estimation of carbon stocks from the biomass is done by multiplying the total biomass by a conversion factor that represents the average carbon content in biomass. In the present study, the default 0.47 carbon fraction was used for estimation of carbon (McGroddy et al 2004).

Carbon (tonnes) = Biomass (tonnes) × Carbon %

RESULTS AND DISCUSSION

There are 50 species (41 genera and 23 families) with a total strength of 1936 tree individuals (<30cm GBH 1602 individuals and >30cm GBH 334 individuals) in the study area. The mean stem density is 129 stems ha^{-1}. The top ten dominant tree species are *Leucaena leucocephala* (549) followed by *Phyllanthus emblica*, *Azadirachta indica*, *Pterocarpus santalinus*, *Gliricidia sepium* , *Aegle marmelos* , *Dalbergia sissoo* , *Terminalia arjuna* , *Albizia lebbeck* and *Bauhinia purpurea* which shares 81.06% of the total population of the sampled inventory. However, different sampling plots (0.1ha) showed diversity as per the species as well as total number of individuals is concerned. This ranges from 33 to 282 individuals in different plots.

Trees ≥10-30 GBH: Trees having ≥10-30cm GBH, share 66.39% in all sampling plots. The values range between 1.07-17.41 m^2 ha^{-1} for different sampling plots**.**

Basal area: The basal area ranged between 0.53-9.21 m^2 ha^{-1} in the sampled plots. The top ten dominant tree species are *Leucaena leucocephala* (17.95 m^2 ha^{-1}) followed by *Phyllanthus emblica*, *Azadirachta indica* , *Pterocarpus santalinus*, *Hardwickia binata*, *Gliricidia sepium*, *Aegle marmelos* , *Dalbergia sissoo*, *Terminalia arjuna* and *Bauhinia purpurea* . The top ten dominant tree species shared 84.36% of the total basal area.

Volume: The volume ranges between 2.23-78.16 m^3 ha^{-1} in the sampled plots. The top ten dominant species are *Pterocarpus santalinus* (80.85 m^3 ha^{-1}) followed by *Leucaena leucocephala*, *Azadirachta indica*, *Phyllanthus emblica*, *Terminalia arjuna*, *Gliricidia sepium*, *Aegle marmelos*, *Albizia lebbeck*, *Dalbergia sissoo and Hardwickia binata*. The top ten dominant tree species shared 87% of the total volume.

Biomass: The biomass ranges between 1.5-75.16 Mg ha^{-1} in the sampled plots. The top ten dominant species are *Pterocarpus santalinus* (78.18 Mg ha^{-1}) followed by *Leucaena leucocephala*, *Azadirachta indica*, *Phyllanthus emblica*, *Terminalia arjuna*, *Gliricidia sepium*, *Aegle marmelos*, *Pterocarpus marsupium*, *Dalbergia sissoo* and *Albizia lebbeck*. The top ten dominant tree species shared 90.37% of the total Biomass.

Trees ≥31 GBH: Trees having ≥31cm GBH, share 33.6% in all sampling plots. The values range between 0.8-6.2 m^2 ha^{-1} for different sampling plots while the mean value stands at 3.528±1.78 m^2 ha^{-1}.

Basal area: The basal area ranges between 1.0-10.4 m^2 ha^{-1} in the sampled plots while the mean was 5.56 m^2 ha^{-1}. The top ten dominant tree species are *Azadirachta indica* (31.4 m^2 ha^{-1}) followed by *Hardwickia binata*, *Albizia lebbeck*, *Pterocarpus santalinus*, *Leucaena leucocephala*, *Terminalia arjuna*, *Ficus religiosa*, *Dalbergia sissoo*, *Tamarindus indica* and *Phyllanthus emblica*. The top ten dominant tree species shared 81.42% of the total basal area.

Volume: The volume ranges between 1.3-175.8 m^{3}ha^{-1} in the sampled plots while the mean value stands at 42.23m^3 ha^{-1}. The top ten dominant volume species are *Azadirachta indica* (407 m^3 ha^{-1}) followed by *Albizia lebbeck*, *Leucaena leucocephala*, *Pterocarpus santalinus*, *Hardwickia binata*, *Ailanthus excelsa*, *Terminalia arjuna*, *Ficus religiosa*, *Terminalia bellirica* and *Ficus benghalensis*. The top ten

Table 1. Quantitative attributes of ≥10 to ≤30 cm GBH tree individuals

Plot No.	Tree Individuals	Diameter	BA	Volume	Biomass	BGB	TB	Carbon
1	65	7.33	6.529	14.6539	9.38089	1.876	11.257	5.291
2	117	7.75	4.283	78.1664	75.5869	15.117	90.704	42.631
3	171	7.49	3.161	7.85375	4.78571	0.957	5.743	2.699
4	273	17.41	9.217	16.1594	9.71368	1.943	11.656	5.479
5	38	2.54	1.493	2.23513	1.49565	0.299	1.795	0.844
6	215	12.1	5.858	14.4421	11.1106	2.222	13.333	6.266
7	113	8.1	4.742	11.1184	7.98298	1.597	9.580	4.502
8	67	4	1.979	6.95662	5.18369	1.037	6.220	2.924
9	19	1.07	0.537	4.21967	2.46675	0.493	2.960	1.391
10	115	9.61	4.065	8.61461	5.40523	1.081	6.486	3.049
11	131	8.03	4.099	7.94158	4.81141	0.962	5.774	2.714
12	83	5.27	2.803	7.61789	5.64005	1.128	6.768	3.181
13	109	6.68	3.541	7.6258	5.01832	1.004	6.022	2.830
14	36	2.56	1.524	6.46335	4.13889	0.828	4.967	2.334
15	70	4.61	2.569	6.04083	3.88944	0.778	4.667	2.194
	1602		56.401	200.11	156.61	31.322	187.932	88.328

*BA=Basal Area; ABG=Above Ground Biomass; BGB=Below Ground Biomass; GBH=Girth at Breast Height; TB=Total Biomass dominant tree species shared 96.96% of the total volume.

Biomass: The biomass ranges between 0.9-121.67 Mg ha^{-1} in the sampled plots while the mean value stands at 28.14 Mg ha^{-1}. The top ten dominant Biomass species are *Azadirachta indica* (282.58 Mg ha^{-1}) followed *Albizia lebbeck*, *Pterocarpus santalinus*, *Leucaena leucocephala*, *Hardwickia binate*, *Terminalia arjuna*, *Ailanthus excelsa*, *Pterocarpus marsupium*, *Terminalia bellirica* and *Tamarindus indica*. The top ten dominant tree species shared 94.5% of the total Biomass.

Correlation of basal area and biomass of trees with ≥31cm diameter revealed a determination coefficient of R^2 is 0.9457. Rao et al (2013) reported R^2=0.944, 0.97 & 0.982 for trees outside forests of Kurnool district, Andhra Pradesh. Srinivasa Rao et al (2012a) reported trees outside forests of Kadapa district, Andhra Pradesh (R^2=0.986 & 0.9939). Srinivasa Rao et al (2012b) reported R^2 values from trees outside forests of Prakasam district, Andhra Pradesh (R^2=0.952, 0.986 & 0.871). Bhakthavathsalam (2010) reported R^2 values from Vengalammacheruvu reserved forest of Anantapur district, Andhra Pradesh (R^2=0.961). Patil, (2009) reported R^2 values from forests of Surat district (R^2 = 0.9061), Kashmir Valley (R^2 = 0.9232), Uttarkashi district (R^2 = 0.934) and Panna district (R^2 = 0.903) in North West India. A significant R^2 value of 0.98 by using basal area and 0.94 by using height and basal area was reported from tropical dry evergreen forests of peninsular India by Mani and Parthasarathy (2007). All above R^2 values are greater than obtained in the present study.

Total biomass (TB): The total above-ground biomass of ≥10-30cm (TAGB) and below ground biomass (BGB) ranged between 1.8-90.7 Mg ha^{-1} in the sampled plots with a mean of 12.53Mg ha^{-1}. It accounts for 187.932 Mg in the study area (Table 2). The total above-ground biomass ≥31cm (TAGB) and below ground biomass (BGB) ranged between 1.1-146 Mg ha^{-1} in the sampled plots with a mean value of 33.77Mg ha^{-1}. It accounts for 506.509 Mg in the study area (Table 2). The total biomass in study area (all the individuals) is 694.441 Mg.

Above ground biomass widely varies due to regional differences in stem size distribution, soil fertility, topography and disturbance (Rolim et al 2005, Sarmiento et al 2005, Castilho et al 2006, Malhi et al 2006, Muller-Landau et al 2006, Urquiza-Haas et al 2007). Above ground biomass varies from plot to plot in a forest area due to different stages

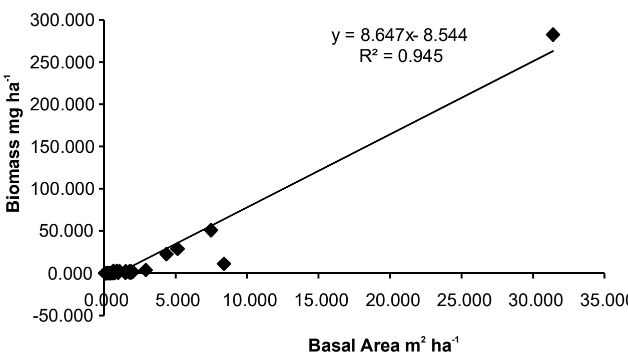

Fig. 2. Correlation between basal area and biomass of ≥31cm diameter

Table 2. Quantitative attributes of ≤31 cm GBH tree individuals

Plot number	Tree individuals	Diameter	BA	Volume	Biomass	BGB	TB	Carbon
1	22	4	6.535	30.471	20.651	4.130	24.781	11.647
2	39	6.2	10.382	82.426	53.113	10.623	63.735	29.956
3	5	0.8	1.193	2.552	1.562	0.312	1.874	0.881
4	9	1.65	3.433	37.407	22.520	4.504	27.024	12.701
5	22	2.91	3.244	2.239	1.433	0.287	1.720	0.808
6	27	4.11	7.414	124.117	85.824	17.165	102.989	48.405
7	15	1.77	1.637	1.297	0.897	0.179	1.076	0.506
8	38	6.22	9.090	20.571	13.738	2.748	16.485	7.748
9	14	3.72	10.191	175.808	121.669	24.334	146.002	68.621
10	27	3.76	5.501	58.083	40.087	8.017	48.104	22.609
11	17	3.17	5.296	21.419	12.064	2.413	14.477	6.804
12	39	6.15	8.763	24.903	18.060	3.612	21.672	10.186
13	5	0.81	0.963	1.818	1.225	0.245	1.470	0.691
14	24	3.24	3.608	6.154	5.004	1.001	6.005	2.822
15	31	4.41	6.146	44.161	24.245	4.849	29.094	13.674
	334		83.395	633.426	422.091	84.418	506.509	238.059

See Table1 for details

of forest growth cycles, habitat variation and tree density. Biomass variability can be explained by several factors like climate, topography, soil fertility, water supply and wood density, distribution of tree species, tree functional type and forest disturbance (Sicard et al 2006).

Carbon stock: The carbon pool ranges between 0.8-42.6 Mg ha^{-1}in the sample plots with a mean value of 5.9Mg ha^{-1} of ≥10-30cm GBH trees the total carbon pool in the study area is 88.328 Mg (A) (Table 1). The carbon pool ranges between 0.5-68.62 Mg ha^{-1} in the sample plots. The total carbon pool in the study area is 238.059 Mg (B) (Table 2). The total carbon pool in the study area (all the individuals A+B) is 326.388 Mg.

CONCLUSION

The study area is dominated by *Leucaena leucocephala*, *Phyllanthus emblica*, *Azadirachta indica*, *Pterocarpus santalinus, Gliricidia sepium*, *Aegle marmelos*, *Dalbergia sissoo*, *Terminalia arjuna*, *Albizia lebbeck* and *Bauhinia purpurea.* A total of 50 species were documented. These species were classified into two girth classes *i.e.* ≥10-30cm GBH and ≥31cm GBH. Total standing crop biomass was estimated based on volume and specific gravity. Carbon accumulations were estimated using carbon factor 0.47 and it estimated that a total of 326.388 Megagrams (Tonnes) (equals to 0.000326 million tonnes) of carbon was found sequestered in the sampling plots and which is equal to 27.2 Mg of carbon dioxide accumulated and release 10.2 Mg of oxygen into atmosphere. India submitted its Intended Nationally Determined Contribution (INDC) to the United Nations Framework Convention on Climate Change (UNFCCC) in October 2015, committing to cut the emissions intensity of GDP by 33–35% by 2030 from 2005 levels. In recognition of the growing problem of Climate Change, India declared a voluntary goal of reducing the emissions intensity of its GDP by 20–25%, over 2005 levels, by 2020, despite having no binding mitigation obligations as per the Convention. A slew of policy measures was launched to achieve this goal. As a result, the emission intensity of our GDP has decreased by 12% between 2005 and 2010. To reach the target by 2030 more number of urban parks and should be developed and also other GHG gas reduction systems should be established. With this it concluded that at present, information on the estimation of sequestered carbon potential in natural way in urban areas is not available. Therefore, more detailed studies are needed for assessing the ultimate changes that are happening in both forest and urban areas.

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Analysis of Wood Anatomical Features of Shisham (*Dalbergia sissoo*) from Rajasthan

Shishpal Choudhary, Kanica Upadhyay[1], Amol Vasishth and Chandramohan Singh

College of Forestry VCSG Uttarakhand University of Horticulture and Forestry, Ranichauri-249 199, India
[1]CH&F (AU, Kota), Jhalawar-326 023, India
E-mail: spl8239990476@gmail.com

Abstract: In this study Shisham (*Dalbergia sissoo*) wood samples were collected from twelve districts of Rajasthan -Baran, Barmer, Bhilwara, Bikaner, Jaipur, Jhalawar, Kota, Nagaur, Rajsamand, Sikar, Sri Ganganagar and Tonk for analyzing anatomical features. The results revealed that the vessel length (0.195 mm), vessel diameter (0.153 mm), fibre length (1.017 mm) and fibre diameter (20.237 µm) in the Shisham wood were significantly highest from Jhalawar district of Rajasthan. Among different locations, the vessel frequency in Shisham wood was highest (7 per mm^2) from Nagaur district of Rajasthan. Among different districts of Rajasthan, wood samples obtained from Jhalawar district showed desirable anatomical properties that may be best suited for the wood industry.

Keywords: Shisham, Fibre length, Fibre diameter, Vessel length, Vessel diameter

Shisham (*Dalbergia sissoo* Roxb. ex DC.) is an important and substantial forest species distributed in tropical and subtropical regions of Africa and Asia. In addition to these regions, it is also found in Java, Nigeria, Mauritius, Sri Lanka, Kenya, northern Zimbabwe, Palestine, and South Africa. This deciduous tree is native to the foothills of the Himalayas, where it mainly grows along canal banks, roadsides, railway lines, and water canals in agricultural fields. The height of the trees ranges from 30 to 80 feet. Shisham is well-known in the international market for its lumber. On average, the heartwood is golden to dark brown and the sapwood is white to brown, which is durable, tough, and resilient. It can be grown in a variety of soils (Orwa et al 2009).

Wood consists of a matrix of fiber walls and air spaces. From the pith to the bark, from the trunk to the top and from the trunk to the branches and roots, its structural properties vary. The tracheids or fiber cells are the primary structural component of wood. There are wide range of sizes and lengths of these cells, from 16 to 42 microns in diameter and 870 to 4000 microns in length. The properties of a wood vary with the distribution patterns of its microstructures, the arrangement of its component cells, and the size and dimension of its components. Anatomically, hardwood consists of vessels, fibers, parenchyma cells, and rays that make up its anatomical structure. Fiber is the main component responsible for the strength of wood (Sinha et al 2019). Ocloo and Laing (2003) observed that these anatomical properties correlate positively with the strength properties of the wood. Fiber length is one of the quality parameters for sawn timber, plywood, pulpwood (Jorge et al 2000, Dhaka et al 2020). Anatomical properties of wood included studies of vessel, ray and axial parenchyma properties and their derived values. The study of wood fiber determines its use in the industries, such as making the pulp and paper, carbonated wood, and bioethanol (Adi et al 2011, Chaudhary et al 2017). The importance of the Shisham can be characterized as being a fast-growing and multi-purpose tree. It can fix nitrogen, is easy to propagate, and offers a high economic return to the stakeholders. It is one of the most demanding tree species in timber industry. This multipurpose tree is economically significant as it is used as a medicinal tree to treat various diseases (Qurashi 2004). Shisham is a money-making tree because farmers earn good economic return by selling wood which is used in the manufacture of furniture, plywood industries, construction, and fuel. It is an inexpensive source of indigenous medicines to treat various ailments (Azaizeh et al 2003). It is perfect for railroad ties, but is rarely used for sleepers due to its usefulness in construction and furniture making. Shisham, like teak and rosewood, is considered one of the best cabinet and furniture wood in India, due to its rich brown color, attractive shape, good grain. The wood properties vary from species to species and according to edaphoclimatic conditions. Therefore, it is important to study wood properties of one species according to the climatic conditions of its place of occurrence. Therefore, present study was undertaken to ascertain the wood properties of wood from different regions of Rajasthan, India.

MATERIAL AND METHODS

The *D. sissoo* (Shisham) wood samples (n=3) were collected from twelve districts (taken as treatments) of Rajasthan Baran, Barmer, Bhilwara, Bikaner, Jaipur, Jhalawar, Kota, Nagaur, Rajsamand, Sikar, Sri Ganganagar and Tonk from tree of similar diameter class. The recorded data were subjected to one way ANOVA for statistical analysis.

D. sissoo logs were converted into different sample sizes according to the test specifications. The samples were properly planed and sanded to maintain smoothness. The size of the specimens prepared for anatomical studies was 20 mm x 20 mm x 20 mm. Wood samples were preserved in 100 ml formalin-acetic acid (FAA) solution consisting of 90 ml 70% ethyl alcohol, 5 ml glacial acetic acid and 5 ml formaldehyde (Johanson 1940). The wood sections were macerated by dipping the wood shavings in Jefferys solution (10% chromic acid and 10% nitric acid) for 48 hours (Pandey et al 1968). The anatomical parameters like vessel parameters (Vessel length, Vessel diameter and number of vessel) and fibre dimensions (fibre length and Fibre diameter) were studied under the microscope.

Vessel parameters (length, diameter and frequency) were measured by macerating the wood in Jefferys solution (10% chromic acid and 10% nitric acid) for 48 hours (Pandey et al 1968). The macerated woods havings were then thoroughly washed, stained with safranine, and roughened with a needle in 10% glycerine before mounting on slides. Straight and complete fibers were selected and measured under a stereomicroscope fitted with a 10X eyepiece. Vessel element length was observed from the macerated wood samples by using ocular and stage micrometer. Vessel element was observed on the macerated wood samples using an eyepiece and a stage micrometer. Vessel frequency was observed under the microscope in transverse sections of the wood. The number of vessels was counted at 10x magnification in a microscopic field. The diameter of the microscopic field was measured using a bench top micrometer, and then the area of the microscopic field was calculated using the formula: $A = \pi r^2$, Where, π=3.143, r=radius of microscopic field standardized with stage micrometer. Fiber length (mm) and diameter (µm) were measured after maceration using Jeffery's method determined by placing the wood chips in Jefferys solution, i.e.10% chromic acid and 10% nitric acid, were soaked (Pandey et al 1968). The shavings were then thoroughly washed, stained with safranine and roughened with a needle in 10% glycerine before mounting on slides. Straight and complete fibers were selected and measured under a trinocular microscope fitted with a 10X eyepiece. The fiber measurements (7-10 readings for each replication) were made in each slide using an ocular micrometer attached to the eyepiece of a microscope at 10X magnification and standardized using a micrometer.

RESULTS AND DISCUSSION

Vessel parameters: Significant variation for vessel length of Shisham wood collected from different districts of Rajasthan were observed (Table 2). Vessel element length of Shisham wood from Jhalawar district was maximum (0.195 mm) followed by wood from Kota (0.186 mm), whereas the minimum (0.162 mm) was in Rajsamand was statistically at par with vessel element length of wood (0.164 mm) from Tonk region. On contrary to the present study, Pandey and Singh (2005) observed that variation in anatomical features such as vessel length, vessel diameter, and wall thickness of wood in a tree in vertical or radial and axial direction and at different locations were not significant in *Shorea robusta*. Similar results were also reported by Sunny (2017) in *D. sissoo.* Vessel diameter showed significant differences in Shisham wood from different districts of Rajasthan (Table 1). The vessel diameter of Shisham wood from Jhalawar district was maximum (0.153 mm) followed by wood from Sikar (0.150 mm), whereas the minimum vessel diameter was observed in wood samples from Bikaner and Barmer districts (0.128 mm). Karimanisha et al (2020) reported a radial variation in vessel diameter from pith to periphery region of *D. sissoo*. As regards vessel frequency, highest number of vessels were in the wood samples of Nagaur which was statistically equivalent to Bhilwara, Jaipur, Rajsamand, Sikar and Sri Ganganagar wood samples. The lowest number of vessels (2 mm^2) was in the wood samples from the Kota area, which was statistically equal to the samples from Barmer (3 mm^2).

Table 1. Geographic location of collected *D. sissoo* (Shisham) wood samples

Location	Latitude	Longitude
Baran	25° 6'4.1220"N	76° 30'47.3796"E
Barmer	25° 45' 11.3580"N	71° 25'5.0160"E
Bhilwara	25° 19' 36.7536"N	74° 36'49.2948"E
Bikaner	28° 1'37.6968"N	73° 18'7.7580"E
Jaipur	26° 55' 19.4520"N	75° 46'43.9860"E
Jhalawar	24° 35' 50.4564"N	76° 9'39.5280"E
Kota	25° 9'46.7928"N	75° 50'43.1592"E
Nagaur	27° 12' 25.2216"N	73° 44'32.2584"E
Rajsamand	25° 4'0.1200"N	73° 52'59.8836"E
Sikar	27°36'55.4616"N	75° 7'33.1860"E
Sri Ganganagar	29°54'13.8204"N	73° 52'37.8840"E
Tonk	26°13'45.7536"N	75° 46'51.4020"E

Variation in fibre dimensions: The significant difference was observed in fiber length of Shisham wood samples collected from different districts of Rajasthan (Table 3). Longest fiber length (1.017 mm) was recorded in samples collected from Jhalawar which was statistically at par with samples from Baran , Bhilwara, Kota, Rajsamand, Sri Ganganagar and Tonk. The shortest fiber length (0.877 mm) was recorded in samples collected from Sikar and Bikaner, which was statistically at par with specimens from Nagaur, Barmer and Jaipur. Sykes et al (2006) observed that fiber length is genetically controlled and not subject to the influence of the environmental fluctuations. Similar results in fiber length variation were reported by Krisdianto and Damayanti (2007) in *Acacia nilotica* and Saravanan et al (2014) and Sinha et al (2019) in *Melia dubia* wood. Mahmood et al (2016) also found the fiber length of 1470 mm in the samples of *A. nilotica* collected from Pakistan. Sunny (2017) reported large fiber length (1.66 mm) of *D. sissoo*. Karimanisha et al (2020) reported the variation in fiber morphology between different radial positions of the *D. sissoo* wood samples. Tavares et al (2011) observed that in *Eucalyptus* globule wood, fiber length decreases from the base to the top of the tree and increases from the pith to the bark. Similar results have been reported by Panda et al (2021) in Teak, Meghwal and Chauhan (2020) in *D. sissoo*.

Among the Shisham wood samples (Table 3), the maximum fiber diameter (20.23 µm) was from Jhalawar, which was statistically at par with samples from Kota and Tonk. Smallest fiber diameter (19.87 µm) was observed in the wood samples from Nagaur district, which was statistically at par with samples from Baran , Barmer , and Bikaner . Fibers are the main elements responsible for the strength of wood. In the present study, variations in fiber length and diameter may be due to genetic (chemical composition, age, elevation, location of trees, *etc*.) and environmental factors (soil composition, mean annual precipitation, seasonality, and temperature). David et al (2009) reported a decrease in fiber diameter in response to decreased water availability. Similarly, Karimanisha et al (2020) observed average fiber diameter of 24.23 µm in *D. sissoo* wood and fiber length and fiber diameter both increases from pith to the periphery. Wani

Table 3. Variation in fibre length (mm) and fibre diameter (µm) of *D. sissoo*

Treatment	Location	Fibre length (mm)	Fibre diameter (µm)
T_1	Baran	0.983	19.960
T_2	Barmer	0.890	19.953
T_3	Bhilwara	0.980	20.027
T_4	Bikaner	0.877	19.950
T_5	Jaipur	0.897	19.990
T_6	Jhalawar	1.017	20.237
T_7	Kota	0.973	20.233
T_8	Nagaur	0.887	19.870
T_9	Rajsamand	0.987	20.007
T_{10}	Sikar	0.877	20.087
T_{11}	Sri Ganganagar	0.977	20.063
T_{12}	Tonk	0.993	20.187
CD (p=0.05)		0.058	0.107
SE		0.028	0.052

Table 2. Variation in vessel length (mm), diameter (mm) & number of vessels per mm^2 of *D. sissoo*

Treatment	Location	Vessel element length (mm)	Vessel element diameter (mm)	Vessel frequency (No. of Vessels per mm^2)
T_1	Baran	0.168	0.131	4
T_2	Barmer	0.170	0.128	3
T_3	Bhilwara	0.178	0.149	6
T_4	Bikaner	0.168	0.128	5
T_5	Jaipur	0.173	0.132	6
T_6	Jhalawar	0.195	0.153	4
T_7	Kota	0.186	0.146	2
T_8	Nagaur	0.167	0.133	7
T_9	Rajsamand	0.162	0.140	6
T_{10}	Sikar	0.169	0.150	6
T_{11}	Sri Ganganagar	0.168	0.137	6
T_{12}	Tonk	0.164	0.145	5
CD (p=0.05)		0.005	0.005	1.376

and Khan (2010) reported significant differences in wood fiber diameter of *Populus nigra* from different locations. Similar results were also reported by Chauhan et al (2022) in Himalayan conifers.

CONCLUSION

The present research was conducted to evaluate the variation in some anatomical properties of Shisham wood from different regions of Rajasthan. Wood samples of *D. sissoo* collected from Jhalawar showed maximum fiber dimensions as compared to the samples from other sites of Rajasthan, which ensures the strength provided by the fibers to the wood. Results showed that the Shisham wood from Jhalawar, Baran and Kota sites has shown better performance as far as wood anatomical characteristics are concerned. The results of the present study may provide some insights for effective utilization of Shisham wood from different locations for different end uses.

AUTHOR CONTRIBUTIONS

Conceptualization, K.U.; Methodology, K.U. and S.C.; Data collection, S.C.; Validation and Formal Analysis, S.C.; Writing and Original Draft Preparation, S.C.; Rewriting, Review and Editing, K.U., A.V. and C.S.; Supervision, K.C. and A.V.

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Impact Assessment of Check Dams on Irrigation Potential and Effect on Socio Economic Condition of Farmers in Eastern Uttar Pradesh

Rajat Mishra, Arpan Sherring, P.V. Singh[1], Shekhar Singh[1], A.K. Mishra and Vishnu ji Awasthi

Vaugh Institute of Agricultural Engineering & Technology, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj-211007, India
[1]Department of Soil and Water Conservation Engineering, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar-263 145, India
E-mail: rajatmishra826@gmail.com

***Abstract*:** This study analyzed the impact of check dams on irrigation availability and socio-economic conditions in Meja tehsil of Prayagraj district. Check dams were built in drought-prone areas to retain rainwater, providing water for agriculture and domestic use and recharging groundwater. Crop water requirements were calculated using crop-evapotranspiration and potential evapotranspiration was assessed using the Penman-Monteith method. Socio-economic analysis was done using Focus Group Discussion and Household Questionnaire. The study found a 31% increase in irrigated cropped areas and positive changes in household and economic conditions. Additionally, the overall cropping area for wheat and paddy crops has experienced an average growth of 32% and 51%, respectively. Check dams have a significant positive impact on agriculture and socio-economic life in the study region.

***Keywords*:** Irrigation water requirement, Check dam, Socio-economics, Penman-Monteith

Water is a necessary natural resource for the successful and healthy growth of crops. With the inadequate or uneven distribution of rainfall during the growing span of a crop because of climate change and mountain effects, it becomes essential to apply additional water to the soil for plant use in the form of irrigation (Liebe et al 2005). India is an agricultural country having 70% of its population economically dependent on agriculture, however per capita, arable land is quite small. The solution to the problem to meet the rising demand for food, fodder, feed and fuel for the human and livestock population can only be achieved by increasing the production per unit of land. Agricultural production can be increased with proper management practices, particularly water management (Alfonso-Torreño et al 2022). The irrigation water is ensured through check dams, reservoirs, ponds, tube wells, and canals. Irrigation planning assumes great significance in the wake of growing concern for the conservation, proper utilization, and development of natural resources (Polyakov et al 2014). A check dam is a small obstruction that is placed across a river or channel to mitigate sediment transportation and reduce the adverse effect of water velocity to reduce channel erosion (Xu et al 2013). In catchments with a high susceptibility to soil and water contamination through erosion, check dams could trap water and sediments before they reach the river or reservoir (Das et al 2021). Due to undulating topography, Meja tehsil faces water scarcity as well as Vindhyan Super group soil in which Shale and sandstone rocks in subsurface strata limit the groundwater use and recharge (CGWB report, 2007). The usage of groundwater is also being restricted by the application of fertilizer and crop manure in the field (Singh et al 2021). In many cases, farmers complain that irrigation water is not available to them in an inadequate amount and at the appropriate time, causing a reduction in yield (Raskar et al 2020). Sometimes the supply of water is in excess causing water logging while during some periods of crop growing season, check dam irrigation projects do not carry water at all despite high project supply requirements resulting in low crop production (Pari et al 2021). Most of the check dam irrigation projects do not store adequate water so farmers of the command area do not get sufficient water, which often results in crop failure (Ramathilagam et al 2017). On the other hand, authorities claim that water is being supplied according to the requirement of the command area of the project. But they also accept deficient water supply to meet their requirement due to erratic rainfall, heavy losses in conveyance system caused by weeding, silting of the waterway, heavy water losses in field channels, improper methods of irrigation, lack of training and awareness of farmers for water management and political interference (Singh et al 2019). The water scarcity conditions affect crop growth and result in a decrease in the crop production and socio-economic condition of farmers. ASA India organization conducted a study to determine the impact of a stop dam on irrigation water availability through various irrigation programs (ASA, 2008). Water is a scarce resource in the drought-prone area of the Prayagraj district of Uttar Pradesh. The main focus of the study was to assess the impact of check dams on irrigation water availability by analyzing the storage capacity of a check dam with water requirements in various crops in the growing season, and the assessment of the socio-economic conditions of the farmer.

MATERIAL AND METHODS

Study area: The study area is comprised of three check dams under Gadarnala of Salaiya Khurd village which is situated in the micro watershed “2A7D2d3e" in Meja tehsil of Prayagraj district. Prayagraj district is a part of the Vindhyan plateau, with the predominant rock structures made of shale and sandstone, also known as Deccan traps. Salaiya Khurd village is spread over an area of 771.37 hectares on the catchment of river Tamas. The area of catchment and command area of check dams lies approximately between 25°14'26.69"N 81°95'21.07 "E, 25°10'37.68"N 81°95'08.64"E. The first check dam (CD-1) is located between Chainage 0-600 m in Gadarnala and covers the command area of 15.99 hectares. The second check dam (CD-2) is situated between chainage 600-1100 m and covers the command area of 8.24 hectares. The third check dam (CD-3) is situated between chainage 1100-1600 m and covers the command area of 7.54 hectares. The average annual rainfall in the Prayagraj district is 1181 mm on average 49 rainy days and the mean temperature varies between 44.3°C in the hot season and 4.9°C in the cold season (Statistical Diary Uttar Pradesh 2022).

The contemplated affecting appraisal of the check dam, actual confirmation of various pressure-driven and underlying measurements were planned and checked, and diverse parametric perceptions were recorded like the state

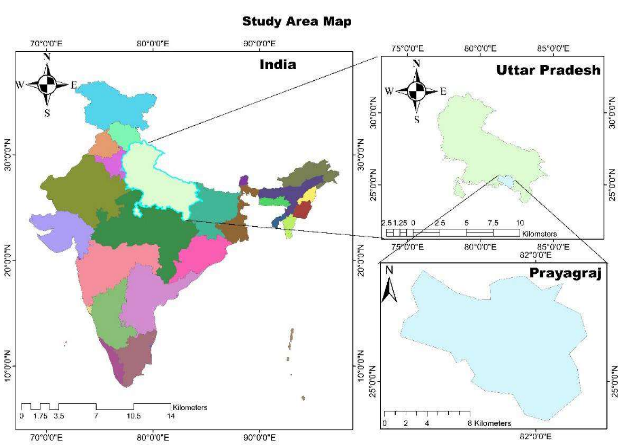

Fig. 1. Study area map

of being of the check dams, soil type, culturing works on, trimming design, and water asset potential. Detailed information regarding the design of check dams was collected from the Minor Irrigation Department, Prayagraj, and existing parameters i.e. crest length, width, and storage depth were measured to calculate the storage volume. The storage capacity of the check dams was calculated using the volumetric method between chainage at different cross-sections and storage depths and total storage capacity was calculated by summing all the storage volumes in different cross-sections between chainage. Focus group discussion and household questionnaire methodology were adopted for a detailed survey in the command area of these checks dams to assess the impact of a check dam on irrigation water availability and socio-economic conditions of farmers. For impact assessment a questionnaire was developed to collect the data, the randomly selected beneficiary farmers from the command area of each check dam were also interviewed and data were collected regarding the variation in cropping pattern adopted before and after the construction of the check dams, improvement in irrigation facilities, and productivity of crops, scheduling, and socio-economic conditions of the farmer and other, marketing and storage facilities, etc. The storage volume of water at selected storage depth = Chainage length* cross-section of the check dam.

It was assumed that the reservoir water loss is only through the surface evaporation losses and seepage through channel crest length. To calculate evaporation loss the shape of the check dam reservoir was considered trapezoidal the following formula was used to calculate the evaporation losses from the reservoir.

$$V = E^{\circ} \times A \qquad (1)$$

V=the total volume of water evaporated (m^3); E°=average evaporation rate (mm)

A= surface area of the reservoir at a particular storage depth (m^2)

For the estimation of crop water requirement crop evapotranspiration was calculated with the help of potential evapotranspiration and crop coefficient. The crop water requirement (ET_{Crop}) for different crops grown in command areas was calculated by the following equation:

$$CWR = ET_{Crop} - P_{eff.} \qquad (2)$$

$$ET_{Crop} = K_C . ET_P \qquad (3)$$

Where,

CWR= Crop water requirement in mm; ET_P= reference/potential evapotranspiration, mm/day

ET_{Crop} = crop water requirement, mm/day; $P_{eff.}$= Effective rainfall, mm

Potential evapotranspiration was calculated by using the

Penman-Montieth equation:

$$ET_p = \frac{0.408\Delta.40 \ -G) + \gamma \dfrac{900}{T+273} u2(es - ea)}{\Delta + \gamma(1 + 0.34u2)}$$

Where,

ET_P = reference evapotranspiration [mm day^{-1}]; Rn = net radiation at the crop surface [MJ m^{-2} day^{-1}]

G = soil heat flux density [MJ m^{-2} day^{-1}]; T = air temperature at 2 m height [°C];

u_2= wind speed at 2 m height [m s^{-1}]; e_s = saturation vapor pressure [kPa]; e_a= actual vapor pressure [kPa]; e_s-e_a= saturation vapor pressure deficit [kPa]; Δ = slope of vapor pressure curve [kPa °C^{-1}]

γ= psychometric constant [kPa °C^{-1}]

Various climatological parameters were collected from the Department of Forestry of Sam Higginbottom University of Agriculture Technology and Sciences and the value of the average crop coefficient was calculated for the study period 2016-2018 by using different growth stages of the crop namely initial, development, mid, and end-stage (FAO 56). To calculate the net irrigation requirement following field water balance model was employed to compute the net irrigation requirement.

NIWR = Σ CWR_i. S_i

Where,

NIWR = Net irrigation water requirement; CWR_i = Crop water requirement for crop i

S_i = Area cultivated with the crop i in ha,

RESULTS AND DISCUSSION

Actual design dimensions of check dams (CDs) were measured using a field survey and enumerated in Table 1. By using the measured data the storage capacities of the check dam 1, 2 and 3 at various stages were calculated and shown in Table 2. This available storage in different chainages was mainly concerned to meet the various household and irrigation demand of the adjacent beneficiaries and their areas.

Table 1. Design dimensions of CDs constructed between chainage 0-1600 m

Design parameters	The measured value of the parameter (meter)
Height of crest	2.00
Top width of crest	1.00
Base width of crest	2.75
Length of crest	15.00
Length of wing wall	25.95
Top width of wing wall	0.50
Base width of wing wall	1.25
Hight of wing wall	2.50
Length of cistern	4.00
Depth of cistern	0.30
U/S floor	3.00
D/S floor	8.00

During the monsoon periods, the storage reservoir of the check dams is at full reservoir level and thus providing the total storage volume of 12480, 4555.6, and 2640 m^3 for irrigation in the respective command areas of the check dams. This storage volume is generally available till the beginning of the rabi crop growing season and the depletion in storage takes place as per the evaporation rate and water usage during the rabi crop growing season.

For post-monsoon season when storage volume was in depletion condition, various storage calculation was done for different reservoir level to assess the impact of check dams on water availability. In post-monsoon season at ¾ reservoir level, the total storage available for irrigation was estimated as 9360, 3416.70 and 1980 m^3, in ½ reservoir levels 6240, 2277.8 and 1080 m^3; and ¼ reservoir condition, 3120, 1138.9 and 660 m^3 for CD-1, CD-2 and CD-3 respectively.

Evaporation loss: For estimating evaporation losses from the reservoir surface, the pan evaporation data is used with the open surface area of the reservoir check dam. The 4593.5, 3214.56, and 2649.84 m^3/year reservoir water loss through the evaporation in the first, second, and third check dam respectively; which was estimated with the help of channel surface area and pan evaporation data of the study area. The highest evaporation loss was estimated in June followed by May and the minimum evaporation loss was estimated in January (Fig. 2).

Monthly crop evapotranspiration (ET_c) (mm) from various crop fields in the command area of check dams: There was maximum evapotranspiration for the wheat crop in December and minimum evapotranspiration in March (12.59 mm) at the harvesting stage (Table 3). At the germination and its critical stage as the CRI stage, the water required for crop evapotranspiration was 81.2 mm in November. For the mustard crop, the maximum crop water required in December was 143 mm and the minimum evapotranspiration was 41.29 mm in February. For the paddy crop, the maximum crop water requirement was 176.11 mm in August month and minimum crop water was required at the harvesting stage i.e. 16.39 mm in November.

Net irrigation requirement: Irrigation requirement in the command area of the check dam was estimated to develop a comparative study between water availability and water requirement before and after the construction of the check dam. The irrigation requirement in the rabi season was

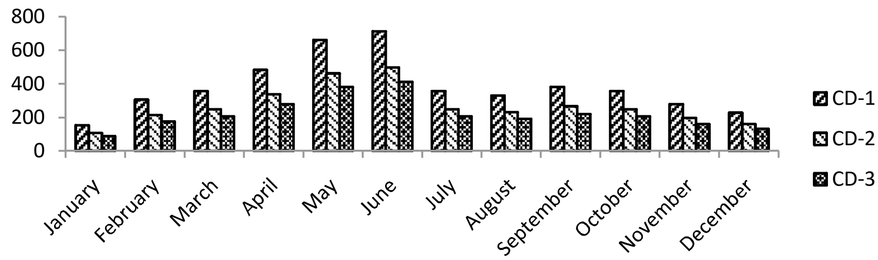

Fig. 2. Monthly evaporation from Check Dams reservoir

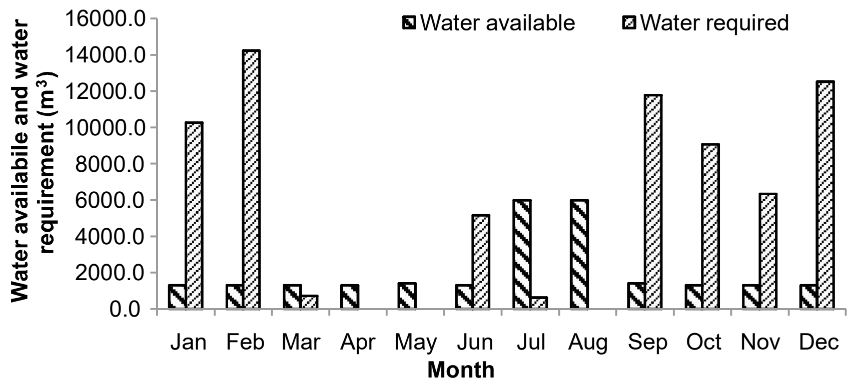

Fig. 3. Water availability and water requirement in the command area of CD-1

Table 2. Storage capacity of CDs

Chainage (m)	Avg. width (m)	Avg. depth (m)	Storage at full reservoir level (m^3)	Storage at 3/4 reservoir level (m^3)	Storage at 1/2 reservoir level (m^3)	Storage at 1/4 reservoir level (m^3)
Storage capacity of CD-1						
0-100	14	1.8	2520	1890	1260	630
100-200	14	1.6	2240	1680	1120	560
200-300	14	1.6	2240	1680	1120	560
300-400	14	1.5	2100	1575	1050	525
400-500	13	1.4	1690	1267.5	845	422.5
500-600	13	1.3	1690	1267.5	845	422.5
Total			12480	9360	6240	3120
Storage capacity of CD-2						
600-700	13	1.2	135.6	101.7	67.8	33.9
700-800	13	1	1300	975	650	325
800-900	12	1	1200	900	600	300
900-1000	12	0.9	1080	810	540	270
1000-1100	12	0.7	840	630	420	210
Total			4555.6	3416.7	2277.8	1138.9
Storage capacity of CD-3						
1100-1200	12	1	1200	900	360	300
1200-1300	12	0.5	600	450	300	150
1300-1400	12	0.4	480	360	240	120
1400-1500	12	0.3	360	270	180	90
1500-1600	12	0	0	0	0	0
Total			2640	1980	1080	660

58,523 cubic meters and 41380.27 cubic meters in the Kharif season. In CD-2 for the rabi season, there were 36,857.67 cubic meters of water and for the Kharif season 25112.75 cubic meters of irrigation water is required to fulfil the different water needs of crops. For the CD-3 command area 16,201.9 cubic meters of water is required to fulfil the rabi crop needs and 10592.56 cubic meters of water is required for the Kharif season to meet the irrigation requirement of the adopted cropping pattern (Table 4).

Impact of check dam on irrigation water availability: During the rabi season water requirement is at its peak and additional sources such as groundwater are taken to fulfil the various usage. During the Kharif season, from excess rainfall, the check dams are overtopped by water as most of the rain occurs during this period. The total net irrigation requirement for an adopted cropping pattern in CD-1 was 99,903.27 cubic meters (Fig. 3). In the rabi season for the wheat crop net irrigation requirement is 45772.59 cubic meters and in the mustard crop, there were 12750.41 cubic meters of net irrigation required. The total net irrigation requirement for an adopted cropping pattern in CD-2 was 61970.31 cubic meters. In the rabi season for the wheat crop net irrigation requirement is 29322.9 cubic meters and in the mustard crop, there were 7534.87 cubic meters of net irrigation required (Fig. 4). In the Kharif season, for the paddy crop, 25112.6 cubic meters of net irrigation was calculated. In

Table 3. Monthly ET_c (mm) from various crop fields in the command area of check Dams

Month	Wheat	Mustard	Paddy
January	141.75	121.91	-
February	101.46	41.29	-
March	12.59	-	-
April	-	-	-
May	-	-	-
June	-	-	-
July	-	-	169.32
August	-	-	176.11
September	-	-	160.76
October	-	-	121.85
November	81.21	60.01	16.39
December	156.79	143.41	-

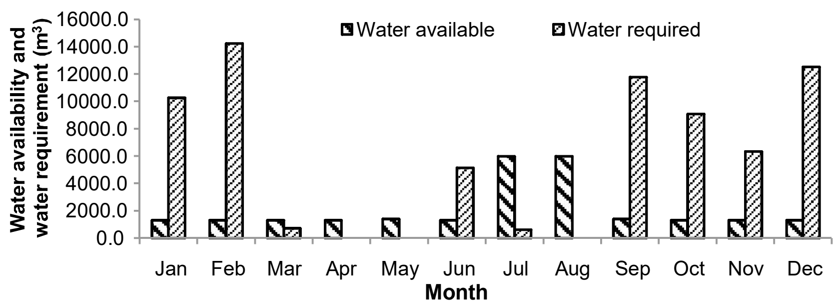

Fig. 4. Water availability and water requirement in the command area of the CD-2

Table 4. NIWR of various crops in the command CD

Crop	Et_o (mm)	Kc	Et_c (mm)	ER (mm)	IR (mm)	Area (ha.)	NIWR (m^3)
Command area of CD-1							
Wheat	437.63	1.15	503.27	38.53	464.74	9.84	45772.59
Mustard	364.13	1.04	378.69	19.73	358.96	3.55	12750.41
Paddy	581.93	1.34	779.78	449.8	329.98	12.54	41380.27
Command area of CD-2							
Wheat	437.63	1.15	506.49	38.53	467.96	6.266	29322.8
Mustard	364.13	1.04	381.98	19.73	362.25	2.08	7534.87
Paddy	581.93	1.34	780.66	449.8	330.86	7.59	25112.75
Command area of CD-3							
Wheat	437.63	1.15	506.33	38.53	467.80	2.72	12724.38
Mustard	364.13	1.04	381.97	19.73	362.24	0.96	3477.52
Paddy	581.93	1.34	779.78	449.8	329.98	3.21	10592.56

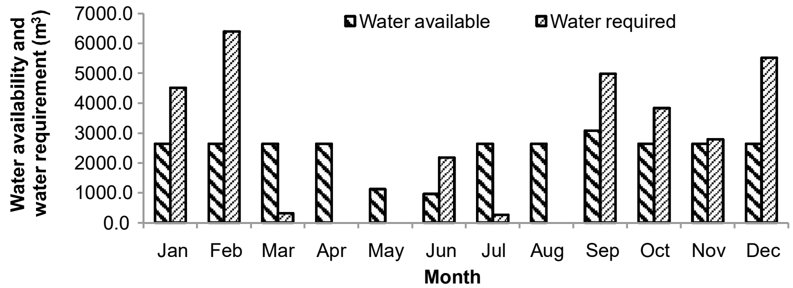

Fig. 5. Water availability and water requirement in the command area of the CD-3

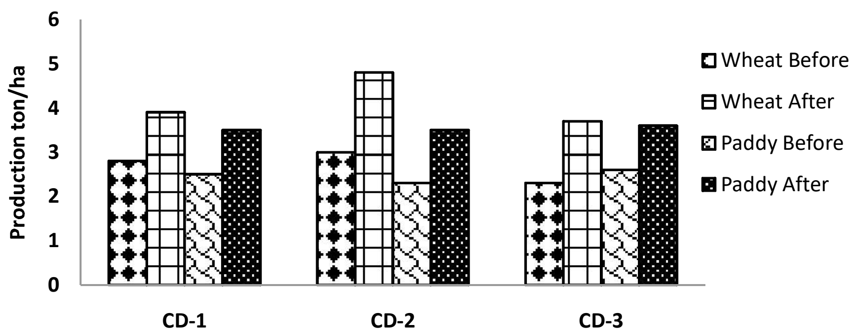

Fig. 6. Impact of the check dams on crop production of command area

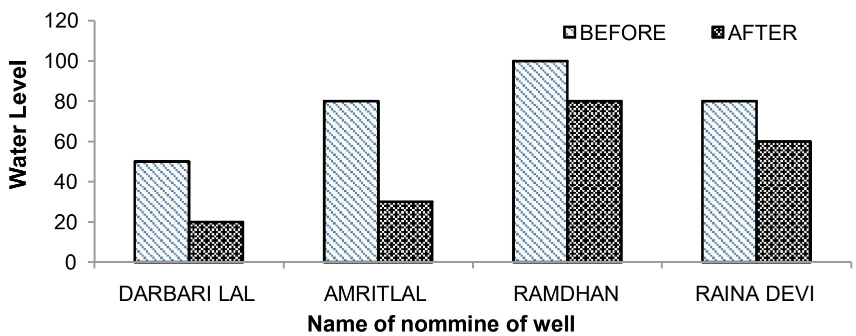

Fig. 7. Water level variation before and after the construction of the check dam (m)

Figure 5 the results were shown that the Wheat crop requires 12728.7 cubic meters of net irrigation, the Mustard crop required 4217.76 cubic meters of net irrigation and for the Paddy crop, 10620.7 cubics meter of net irrigation water is required for optimum production in the CD-3 command area.

Area under irrigation: After the construction of the check dam, the area under irrigation significantly increased which reduces water constraints from cultivation as result expanded remarkably in the area under cultivation. For the Rabi season 8, 14 and 70 % increase in cropping area under CD-1, CD-2 and CD-3 respectively. For the Kharif season 21% under CD-1, 66% under CD-2, and 66% under CD-3 increase in cropping area were recorded after the construction of the check dam.

Impact of check dam on socio-economic condition: There was 28, 56 and 49% increase in agricultural production in CD-1, CD-2, and CD-3 respectively, after the construction of the check dams (Fig. 6). The result shows that there was a significant increase in the socio-economic condition of beneficiaries' farmer of the study area and also in groundwater level (Fig. 7). The developed comparative study reveals that the Farmers living in the command area of the dams were making changes to their house. 90 % of farmers have amended their roofs from mud to concrete. 64 % of farmers had changed their small mud huts to bigger houses made of bricks. Almost 100% of the farmers reported a significant increase in their income and the value of the land as reported 5,75,000 Rs/ha to 31,25,000 Rs/ha. A significant increase of 45% was recorded in the literacy percentage. An average 56.33 % decrease in migration was observed from the farmers living in the nearby villages after the construction of the check dam. After the construction of check dams, the main impact on the industry's stabilization as NTPC establish one hydrothermal power plant in the concerned area for electricity generation which is creating abundant employment and increased the social value of the study area.

CONCLUSIONS

Construction of the check dams resulted in a 31% increase in irrigated cropped area, as well as significant increases in cropping area for wheat and paddy crops (32% and 51% respectively) and agricultural production (44%). Positive changes were also observed in the living conditions of farmers, with many upgrading their roofs and housing. The construction of check dams also contributed to a decrease in migration from nearby villages and an increase in literacy percentages. In the Gadarnala watershed, the check dams have reduced soil erosion, but siltation in the reservoirs continues to be a concern that needs monitoring. Addressing this issue can improve the efficiency of the check dams and ensure water availability during times of high demand.

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Analysis of Trend Using Nonparametric Test for Rainfall and Rainy-Days in Jodhpur Zone of Rajasthan

Deepshikha, Shashi Ranjan[1], Lakshman Singh[2], Anoop Bahuguna[3] and Amit Rawat[4*]

Department of Civil Engineering, Graphic Era Deemed to be University, Dehradun-248 002, India
[1]Department of Civil Engineering, NIT Patna-800 005, India
[2]Department of Civil Engineering, Dev Bhoomi Uttarakhand University, Dehradun-248 007, India
[3]Department of Civil Engineering, Graphic Era Hill University, Dehradun-248 002, India
[4]Department of Civil Engineering, Govind Ballabh Pant Institute of Engineering and Technology, Pauri-246 194, India
[*]E-mail: rawat.amit.gpr@gmail.com

***Abstract*:** The planning, development, and management of water resources are significantly influenced by meteorological processes, which are significantly impacted by climate change. Trends become apparent as a result of these effects. Using nonparametric Mann-Kendall (MK), modified Mann-Kendall (MMK), and Theil-Sen slope estimator tests based on 60 years of data. The current study was conducted to evaluate the historical patterns of annual rainfall and rainy days in the Jodhpur zone of Rajasthan, India (1957-2016). Out of 27 stations, the annual rainfall at seven stations had a significant increasing trend. In the case of annual rainy days, only two stations showed a significant increasing trend. The dramatically increasing trend in rainfall at seven stations in the Jodhpur zone has shown that arid state of Rajasthan has been affected by climate change. The findings of this study could be used as preliminary data for planning and management of agricultural and water resources, as well as for designing soil and water conservation structures to increase the region's water availability.

Keywords: Trend analysis, Mann-Kendal Test, Sen's slope estimator, Rajasthan

Life on Earth depends on water in order to exist. It is the fundamental component on which many meteorological systems rely. Changes in rainfall frequency and pattern have a direct impact on the moisture levels of the soil, stream flows, groundwater levels, and the availability of freshwater reserves. The impacts of anthropogenic and climatic activities are continuously reflected in trends in rainfall, temperature, evapotranspiration, stream flows, and also other meteorological processes (Sen 2013). For the planning and management of water resources at the watershed scale, agricultural engineers, hydrologists, and water scientists must have a comprehensive understanding of rainfall trends. Given that the majority of the nation's agriculture is rain fed in nature, studying rainfall trends is of utmost significance in India. In India, the southwest monsoon, which occurs from June to September, is responsible for nearly 80% of all rainfall (Meshram et al 2017). The management of water resources, agricultural output, and finally the nation's economy are all greatly impacted by fluctuations in the southwest monsoon over India. In certain sections of the country, the monsoon months' significant concentration of rainfall causes a water deficit throughout the other months. A meteorological time series' trend is a deterministic element. The average long-term or regular variation (increasing or decreasing) in a time series is referred to as the trend component. Many recent studies have been conducted on the rainfall and temperature trend analysis in India as well as other parts of the worlds (Rahman and Dawood 2018, Rana et al 2019, Agarwal et al 2021, Harka et al 2021, Sharma and Adhikari 2022). Farooq et al (2021) analysed monthly, seasonal and annual trends in temperature using the non-parametric method for Kazakhstan from 1970 to 2017 and concluded that significant increasing trend in the mean annual temperature for the studied period. Harshavardhan et al (2020) investigated the spatiotemporal trends in rainfall in Krishna River Basin in India from 1965 to 2012.and observed that annual, monsoon and post-monsoon precipitation exhibited a significant negative trend in magnitude from the normal. Salehi et al (2020) examined patterns and identified turning points in Iran's yearly and seasonal rainfall. To conduct the study, the author applied the Mann-Kendall and modified Mann-Kendall tests. Ay and Kisi (2015) found rainfall patterns using ITA and Sen's slope approach for six Turkish. Based on the above review of literature this study is made to determine the trend in annual rainfall & rainy-day and determine the trend magnitude in rainfall & rainy days for Jodhpur zone of Rajasthan provinces.

MATERIAL AND METHODS

Study area: With a total area of 342,239 km^2, Rajasthan is the largest state in India and accounts for over 10% of the overall land area. It is located in the north-western part of the country extending from 23°03′ to 30°12′ north latitude and 69°30′ to 78°17′ east longitude. In the north and northeast, it is bordered by the Indian states of Punjab, Haryana, and Uttar Pradesh; in the southeast, Madhya Pradesh; in the southwest, Gujarat; and in the northwest and west, it has an international boundary with Pakistan. In this study Jodhpur zone of Rajasthan has been selected for study which covers six districts namely Jodhpur, Nagpur, Pali, Barmer, Jalor and Sirohi districts (Fig. 1). The daily data of rainfall and the number of rainy days for 60 years (1957–2016) were obtained from the Department of Water Resources, Govt. of Rajasthan. The location and elevation of all the stations are shown in Table 1.

Methodology: Mann-Kendall test and modified Mann-Kendall test were used to detect trend along with Sen's slope test for magnitude of trend.

Mann-Kendall and modified Mann-Kendall Test: Mann-Kendall test (Mann 1945 and Kendall 1975) has been broadly used to test monotonic trend in hydrological and metrological data (Patra et al 2012, Sa'adi et al 2019, Deoli et al 2020). Mann-Kendall test is suitable for time series data such as hydrological data, metrological data which are not normally distributed (Hameed 2008, Deoli et al 2022).

The MK test for a time series P_1, P_2 P_3…. P_n of length n is given as

$$S = \sum_{j=1}^{n-1} \sum_{k=j+1}^{n} sgn(P_k - P_j) \quad (1)$$

$$sgn(P_k - P_j) = \begin{cases} 1 if P_k > P_j \\ 0 if P_k = P_j \\ -1 if P_k < P_j \end{cases} \quad (2)$$

where, sgn ($p_k - P_j$) enotes the signum function of P_k and P_j. P_j and P_k are the rainfall values at time j and k for j < k. The value of S is generally positive when trend is increasing and negative when trend is decreasing. The variance of the MK statistic is given as

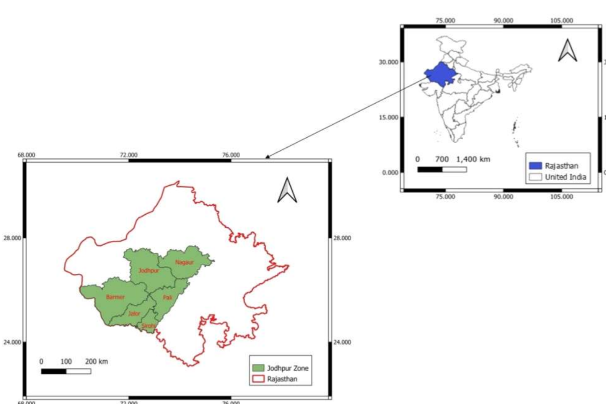

Fig. 1. Study area

$$Var(S) = \frac{n(n-1)(2n+5) - \sum_{m=1}^{q} t_m (t_m - 1)(2t_m + 5)}{18} \quad (3)$$

where q is the number of tied group and t_m is the total data values in m^{th} group.

The Z value is calculated after the calculation of the variance of time series data by following formula

$$\begin{cases} \dfrac{S-1}{\sqrt{\text{var}(s)}} & \text{if } S > 0 \\ 0 & \text{if } S = 0 \\ \dfrac{S+1}{\sqrt{\text{var}(s)}} & \text{if } S < 0 \end{cases} \quad (4)$$

The modified Mann-Kendall (Hamed and Rai 1998) is used to consider the effect of autocorrelation in time series.

Table 1. Location and elevation of all the stations

District	Station	Latitude	Longitude	MSL (m)
Barmer	Barmer	25.75	71.40	190
	Chohtan	25.48	71.07	172
	Siwana	25.48	72.42	170
	Sheo	26.18	71.25	236
Jodhpur	Jodhpur	26.30	73.03	258
	Bilara	26.19	73.71	271
	Osian	26.72	72.92	334
	Shergarh	26.33	72.30	252
	Phalodi	27.13	72.37	229
Pali	Pali	25.78	73.33	219
	Desuri	25.28	73.55	375
	Bali	25.18	73.28	304
	Jaitaran	26.20	73.94	302
	Raipur	26.05	74.05	351
Nagpur	Nagpur	27.20	74.75	385
	Nawa	27.00	75.00	366
	Jayal	27.22	74.18	309
	Degana	26.90	74.33	345
Sirohi	Abu Road	24.48	78.80	300
	Sirohi	24.88	72.88	320
	Mt. Abu	24.53	72.74	1186
	Pindwara	24.78	73.06	368
	Sheoganj	25.16	73.07	269
Jalore	Jalore	25.35	72.62	163
	Ahore	25.33	72.75	192
	Bhinmal	25.02	72.29	149
	Sanchore	24.75	71.77	60

MK test produces false results due to the presence of autocorrelation in time series data, in this case Modified MK test give correct results. Hence, accounting for autocorrelation in time series becomes very important.

The auto correlation coefficient has been calculated by t-test as

$$t = |\rho_1| \sqrt{\frac{n-2}{1-\rho_1^2}} \quad (5)$$

where 't' is the t-value and n is the number of observations. P is the auto correlation coefficient which is given by

$$\rho = \frac{\sum_{t=1}^{n-k}(x_t - \bar{x}_t)(x_{t+k} - \bar{x}_{t+k})}{\sqrt{[\sum_{t=1}^{n-k}(x_t - \bar{x}_t)^2 \sum_{t=1}^{n-k}((x_{t+k} - \bar{x}_{t+k})^2)]}} \quad (6)$$

where x_t is the observed value, X_t is the mean of first 'n-k' terms and X_{t+k} is the mean of last 'n-k' terms.

If the serial correlation in the rainfall data has been determined the trend in data has been calculated by MMK test and a correction factor is given as follow

$$= 1 + \frac{2}{n(n-1)(n-2)} \sum_{k=1}^{n-1}(n-k)(n-k-1)(n-k-2)\rho_k) \quad (7)$$

where n is the total observation, n^*_s is the effective observation for autocorrelation and p_k is the function of autocorrelation. Variance of MMK test has been calculated by multiplying the variance of MK test by correction factor as

$$Var(S*) = Var(S)\frac{n}{n_S^*} \quad (8)$$

Z- value of MMK test is computed as MK test by using modified variance var(S*).

Sen's Slope estimator: Sen's slope estimator test (Sen

Table 2. Annual rainfall and rainy-days characteristics over the study area

District	Station	Minimum		Maximum		Mean		CV%	
		Rainfall	Rainy days	Rainfall	Rainy days	Rainfall	Rainy days	Rainfall	Rainy days
Barmer	Barmer	29.6	3	484	25	283.3	14.5	60.5	36.5
	Chohtan	38.8	2	654.8	26	291.9	14.5	64.8	48.9
	Siwana	35.6	3	753.6	28	360.5	14.6	54.2	41.5
	Sheo	30.8	3	515.5	25	226.1	16.9	41.3	38.4
Jodhpur	Jodhpur	92	10	819.4	31	364.7	22.1	49.8	42.5
	Bilara	128.1	9	756	34	426.8	20.5	44.6	42.3
	Osian	94	8	652.9	28	615.8	17.9	46.6	36.4
	Shergarh	42.6	5	569	36	251.8	16	61.5	57.4
	Phalodi	65.5	6	421	31	234.5	15.8	56.2	29.4
Pali	Pali	91	7	818	37	418.5	25.2	37.8	36.4
	Desuri	199.5	12	1233.3	39	640	27.9	65.4	50.5
	Bali	174	11	1178.5	47	588.8	25.6	31.2	50.1
	Jaitaran	122.7	12	888.5	42	423.5	22.9	52.3	47.8
	Raipur	116	10	970	40	485.4	23.5	42.1	41.3
Nagpur	Nagpur	107	10	660.5	40	380.7	23	66.6	36.8
	Nawa	188.5	15	875	39	455.9	23.2	36.6	39.7
	Jayal	70	6	698.4	41	370.8	23.1	33.3	33.3
	Degana	120.5	8	778.5	40	422.5	23.2	45.7	38.9
Sirohi	Abu Road	116	12	1306.5	55	670.5	29.9	49.9	43.5
	Sirohi	191.4	8	1514.4	42	703.9	31.6	41.6	44.4
	Mt. Abu	370.5	25	3102.5	91	1586.5	51	50.6	22.5
	Pindwara	145.7	17	1435.5	51	708.5	31.5	59.7	29.7
	Sheoganj	106.6	7	1119.2	41	552.6	22.9	40.3	36.5
Jalore	Jalore	52	6	795	27	420	19.8	25.4	42.6
	Ahore	105.6	8	817.8	34	399.5	20.1	18.7	39.9
	Bhinmal	49	4	837.5	37	406.6	18.8	36.4	39.8
	Sanchore	29	3	938	38	398	18.8	42	25.5

1968) is used to calculate magnitude of trend as a trend slope which is a nonparametric test. The total slope has been estimated by computing slope for all time point pair using the median of all slopes.

In this method, first the slope (S_i) of all data pairs are calculated by

$S_i = (y_j - y_k)/(j-k)$ for i=1, 2, 3……N (9)

Where y_j and y_k are values at j and k for j > k respectively. The median of these N values is Sen's slope estimator and calculated as

$$S_{\frac{N+1}{2}} \quad for\ N\ is\ odd$$

$$\frac{1}{2}(S_{\frac{N}{2}} + S_{\frac{N+2}{2}} \quad for\ N\ is\ even \tag{10}$$

RESULTS AND DISCUSSION

Variation of rainfall: The mean annual rainfall at different stations varied from 226.1 for Sheo to 1586.5 for Mt. Abu (Table 2). The highest rainfall (370.5mm) was d at Mt. Abu station of Sirohi whereas the minimum rainfall (29 mm) was at Sanchore station of Jalore. The mean annual rainy-days were varying from 14.5 for Barmer station to 51 for Mt. Abu station. The minimum number of annual rain days 2 was observed at Chohtan station of Barmer whereas the maximum number of rainy days 91 was observed at Mt. Abu station.

Autocorrelation test: In annual rainfall and rainy-days no station showed autocorrelation in data hence all the trends were calculated using the Mann-Kendall test only.

Table 3. Result of Mann Kendall Test for annual rainfall and rainy-days

District	Station	Z-statistics		Significant trend	
		Annual rainfall	Annual rainy days	Annual rainfall	Annual rainy days
Barmer	Barmer	1.23	1.53	No	No
	Chohtan	2.32	1.5	Yes	No
	Siwana	1.30	1.58	No	No
	Sheo	1.54	2.45	No	Yes
Jodhpur	Jodhpur	1.74	1.22	No	No
	Bilara	1.05	1.54	No	No
	Osian	1.12	1.83	No	No
	Shergarh	2.01	1.32	Yes	No
	Phalodi	-0.35	0.95	No	No
Pali	Pali	2.15	0.54	Yes	No
	Desuri	1.31	1.98	No	Yes
	Bali	1.99	1.53	Yes	No
	Jaitaran	2.10	1.64	Yes	No
	Raipur	1.98	0.9	Yes	No
Nagpur	Nagpur	0.98	1.64	No	No
	Nawa	1.10	1.52	No	No
	Jayal	0.54	1.44	No	No
	Degana	0.62	1.25	No	No
Sirohi	Abu Road	-0.12	-0.68	No	No
	Sirohi	0.58	-0.75	No	No
	Mt. Abu	-0.44	-0.52	No	No
	Pindwara	-0.95	-0.47	No	No
	Sheoganj	0.12	0.46	No	No
Jalore	Jalore	0.35	1.02	No	No
	Ahore	1.26	1.21	No	No
	Bhinmal	1.96	1.32	Yes	No
	Sanchore	-0.35	1.2	No	No

Table 4. Trend magnitude and percentage change in annual rainfall and rainy-days

District	Station	Trend magnitude		Percentage change (%)	
		Annual rainfall	Annual rainy days	Annual rainfall	Annual rainy days
Barmer	Barmer	1.98	0.065	32.5	36.5
	Chohtan	2.45	0.073	54.6	33.9
	Siwana	3.60	0.07	81.6	37.8
	Sheo	1.84	0.082	54.3	31.2
Jodhpur	Jodhpur	1.32	0.047	21.5	39.4
	Bilara	1.55	0.050	23.5	35.8
	Osian	2.02	0.112	29.7	11.2
	Shergarh	1.21	0.077	47.1	33.5
	Phalodi	-0.8	0.032	-18.9	36.7
Pali	Pali	1.83	0.021	33.6	17.8
	Desuri	2.05	0.034	54.8	12.6
	Bali	3.21	0.063	74.3	15.8
	Jaitaran	2.55	0.132	42.9	17.9
	Raipur	1.5	0.065	33.3	19.7
Nagpur	Nagpur	1.4	0.032	29.4	18.7
	Nawa	0.2	0.038	47.2	28.4
	Jayal	0.68	0.098	51.3	22.3
	Degana	0.86	0.043	22.2	14.5
Sirohi	Abu Road	4.5	0.088	32.6	16.6
	Sirohi	-0.65	-0.135	-29.8	-15.5
	Mt. Abu	-0.5	-0.074	-23.6	-18.7
	Pindwara	1.15	0	36.8	35.7
	Sheoganj	-1.4	0.023	-25.2	33.8
Jalore	Jalore	3.35	0.035	32.7	27.4
	Ahore	2.49	0.048	59.7	39.9
	Bhinmal	1.25	0.066	40.5	21.5
	Sanchore	0.96	0.041	29.8	13.5

Trend analysis in annual rainfall and rainy-days: Out of 27 stations 22 station shows an increasing trend in rainfall whereas 5 station shows decreasing rainfall trend from which 7 stations namely Chohtan (2.32), Shergarh (2.01), Pali (2.15), Bali (1.99), Jaitaran (2.10), Raipur (1.98) and Bhinmal (1.96) show significant increasing trend at (Table 2) whereas no station shows significant negative trend. On annual rainy days, 23 stations show an increasing trend and 4 stations show a decreasing trend. Only 2 stations namely Sheo (2.45) and Desuri (1.98) stations show a significant increasing trend whereas no stations show a significant negative trend.

Magnitude of trend and percentage change in annual rainfall and rainy-days: In annual rainfall the lowest positive magnitude of trend was 0.2 mm/year for Nawa station and highest positive was 4.5 mm for Abu Road Station. Stations, varied between -0.5 to -1.4 mm/year. In annual rainy-days the magnitude of trend varies between -0.135 mm/year and 0.132 mm/year (Table 4). The maximum percentage positive change in rainfall is detected 81.6% for Siwana Station and minimum positive percentage change is detected for Jodhpur Station (21.5%) whereas the maximum negative change was -29.8% for Sirohi Station and minimum negative percentage change (-18.9%) for Phalodi Station. In annual rainy-days the percentage changes have been varied from -18.7% for Mt. Abu Station to 39.9% for Ahore Station.

CONCLUSIONS

The present study successfully attempted to assess the temporal trend in annual rainfall and rainy days for the Jodhpur zone of Rajasthan. Out of the 27 stations, the annual rainfall at seven stations Chohtan, Shergarh, Pali, Bali, Jaitaran, Raipur) and Bhinmal show a significant increasing trend (1.96 mm/year to 2.32 mm/year). Similarly, on annual rainy days, Sheo and Desuri show an increasing rainfall trend with 2.45 mm/year and 1.98 mm/year. No station in annual rainfall as well as in annual rainy days showed a significant negative trend. The results of this study will be beneficial for water resource information and planning and designing of soil and water conservation structures for improving water availability in the area.

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AMMI Model based Stability of Little millet [*Panicum sumatrense* Roth. Ex. Roem. & Schult.] Advanced Lines Evaluated across Eighteen Environments in India

T.E. Nagaraja, Nandini C.[1], Sujata Bhat, S. Gazala Parveen D.N. Vinutha and I.S. Tilak

ICAR-All India Coordinated Research Project (Small Millets), University of Agricultural Sciences GKVK, Bangalore-560 065, India
[1]ZAHRS, Hiriyuru, Keladi Shivappa Nayaka University of Agricultural and Horticultural Sciences Shivamogga-577 412, India
E-mail: tenagaraja@gmail.com

***Abstract*:** Identification of superior and stable genotype for commercial cultivation is constrained majorly by the existence of genotype × environment interaction (GEI). Nine little millet advanced lines with checks, were employed over nine Indian locations throughout two rainy seasons during 2017 and 2018 to access the patterns of GEI governing traits *viz*., days to 50% flowering, early flowering and yield (seed & fodder). Statistical analysis (AMMI model and best linear unbiased predictors (BLUP) was performed. The variance due to genotype, environment and GEI was highly significant for all three traits. Environment attributed to a higher proportion of the variation (28.68%-73.44%), while genotypes contributed 1.41-47.30% of the total variation. The GEI contributed 24.00-27.79% of the total variation for all three traits. The testing environments were partitioned into four, three and two mega-environments for seed, fodder yield and days to 50% flowering, respectively. The environments E9, E13 and E6 were representative and discriminative for days to 50% flowering, seed and fodder yield, respectively and can be used to recognize superior early flowering genotypes with high seed and fodder yield adapted to specific agro-ecology. Check (OLM203) performed better than all the genotypes except the advanced line DHLT28-4 for seed and fodder yield, but it was late flowering. DHLT28-4 which is early flowering and most stable with high seed and fodder yielding cultivar can be commercialized in India as a better substitute for the existing varieties.

***Keywords*:** Genotype × environment interaction, Multi-environment trial, Yield stability, GGE biplot, AMMI

Globally, 7000 crop species are grown (Khoshbakht and Hammer 2008), yet the majority of research and breeding is focused on a few crops (Hammer et al 2001), resulting in ignorance about many crops, particularly Little millet. Little millet (*Panicum sumatrense* Roth. Ex. Roem. & Schult.) is a tetraploid (2n = 4x = 36) minor cereal grown in the tropics and sub-tropics is nutritionally comparable to rice and wheat (Saha et al 2016). A 100 grams of little millet seeds include 4.70 g of fat, 7.70 g of crude fibre, 9.30 mg of iron, and 220.00 mg of phosphorus, which is equivalent to cereals and other millets (Gopalan et al 2010). Quite apart from being nutritionally dense, it is a short-duration crop with low water requirements and is also used as livestock fodder, making it more appealing to be cultivated in crop-pasture-based farming in areas with little to no rainfall. It is critical to identify stable and high seed and fodder yielding little millet genotypes for farmer direct use that can replace current cultivars. Before recommending any cultivar for commercial cultivation, it is essential that they be evaluated in varying environments to identify consistent and high seed and fodder-yielding cultivars. Consequently, quantification of the interaction of these cultivars with the target environment under which they are evaluated is essential, this aids in determining the breeding objectives, identifying ideal test environments and recommending regional cultivars with better adaptation (Yan et al 2000). To quantify the impact of GEI and recognize stable and adaptable cultivars across different environments, various statistical tools such as joint regression (Finlay and Wilkinson 1963), stability models (Eberhart and Russell 1966), additive main effects and multiplicative interaction (AMMI; Gauch 1992) and genotype main effects in addition to genotype by environment interaction (GGE) biplots (Yan et al 2000) are employed. The two most popular and highly effective multivariate models to analyze the stability, adaptability, rank genotypes and mega environments (ME) are the AMMI and GGE biplots (Gauch 1992). The farmer interprets the effects of genotypes and environments as an additive and the interaction between them as multiplicative by principal component analysis (PCA). The latter group the additive genotypic effects in the AMMI analysis, together with the multiplicative effects of GEI and analyzes these effects by principal components (PC).

The current study aimed to evaluate the stability and adaptability of nine little millet advanced lines including checks across eighteen environments for days to 50% flowering, seed yield and fodder yield using the AMMI methodology and GGE biplot. For simultaneous identification of high seed fodder-yielding genotypes that were also early flowering with good stability and adaptability, the best linear unbiased prediction (BLUP)-based simultaneous selections, such as the harmonic mean of genotypic values (HMGV), the relative performance of genotypic values (RPGV) and harmonic mean of the relative performance of genotypic values (HMRPGV) are used (de Resende 2004).

MATERIAL AND METHODS

Data of little millet initial and advanced varietal trials (LIAVT) from All India Coordinated Research Project (AICRP) on Small Millets, in which nine little millet advanced lines including three checks (Table 1) evaluated across nine locations (Table 2) in the rainy seasons of 2017 and 2018 is used in this study. The testing locations represented seven states of India. Depending on the onset of monsoon across the test locations of this study, the crop was sown during June-July. The experiment was conducted in a randomized complete block design with three replications. The plot size of each replicate was 6.75 square meters with 10 rows of 3-meter length. A spacing of 22.50 cm × 10 cm was followed. Crop management was followed as recommended in the package of practices. Observations on days to 50% flowering, seed and fodder yield were recorded from each plot. At physiological maturity, seed yield was recorded, further plot size was used as a factor to convert plot yield data to kg ha^{-1}.

Statistical analysis: A combination of a single year and a single location made up eighteen test environments in this study (Table 3). The phenotypic data of days to 50% flowering, seed and fodder yield collected from the nine little millet genotypes evaluated across eighteen environments was confirmed for the homogeneity of variance by Bartlett's test (Bartlett 1937). To determine the significance level of genotypes (G), environments (E) and GEI, combined analysis of variance using a mixed linear model (R Core Team 2020) was used. To determine the GEI effects to assess the adaptability and stability of the little millet genotypes across the eighteen test environments, the AMMI model was used. The genotypes were treated as fixed variables, while the environments as random. The AMMI amalgamates ANOVA for genotype and environment main effects with PCA of the GEI with the axes of the principal components of interactions (Gauch 1988; Yan et al 2007). The AMMI model used is as follows:

$$Y_{ij} = \mu + g_i + e_i + \sum_{k=1}^{n} \lambda_k a_{ik} \gamma_{jk} + e$$

Y_{ij}	=	Trait mean of the ith genotype in the jth environment
μ	=	Experimental genotype mean
g_i and e_j	=	Genotype and environment deviations from the grand mean
k	=	Eigen value of the PCA analysis axis k
α_{ik} and γ_{jk}	=	Genotype and environment principal component scores for axis k
n	=	Number of principal components retained in the model
e_{ij}	=	Error term

Genotype + Genotype × environment (GGE) bi-plot is a subjective/qualitative means to characterize patterns of GEI and assess the relative stability of test genotypes. The first two principal components (PC1 and PC2) derived using adjusted trait mean value from ANOVA are used to construct the GGE biplot (Yan 2001, Yan 2002).

The GGE bi-plot is suggestive of visual interpretation of the GEI patterns, representativeness and discriminating ability of the environments and relative stability of test genotypes. In the current study, the biplots were based on singular-value partitioning = 2, transformed (transform = 0), environment-centered (centering = 2) and standard deviation-standardized (scaling = 0).

The BLUP-based stability parameters such as HMGV (to infer both yield and stability), RPGV (to investigate the mean yield and genotypic adaptability) and HMRPGV (to evaluate stability, adaptability and yield simultaneously; de Resende (2004) and (2016) were estimated. The analysis was computed using the metan package in R software version 4.2.1 (Olivoto et al 2020).

RESULTS AND DISCUSSION

Analysis of variance: The combined analysis of variance emphasized that the sources of variations were significant for days to 50 % flowering, seed yield and fodder yield and supported the existence of environmental heterogeneity and stipulated significant differences between the genotypes since their responses were not coincident in the test environments (Table 3). A significant GEI is suggestive of the need to further analyze the data for AMMI analysis of variance.

AMMI analysis of variance: The most important source of variation for yield was environment, accounting for 73.44% and 60.50% of the total variance (G+E+GE) for seed and fodder yield respectively, contrastingly 28.68% for days to 50% flowering (Table 4). The selection of AMMI as the appropriate model for analyzing the multi-environment trials (METs) data is justified by a large variation due to the environment that is impertinent to evaluate cultivars. The variation due to the GEI accounted 24.00, 25.13 and 27.79%

Table 1. Pedigree information of the little millet genotypes used in the study

Genotype	Developing center	Pedigree
WV 126	Waghai (Gujarat)	Local Collection from Dangs Taluka, Dangs District
DHLT 28-4	Hanumanamatti (Karnataka)	CO 2 x TNAU 26
OLM 217	Berhampur (Orissa)	OLM 217 Selection from Udayagiri Local Bhubaneswear
IIMR LM 7162	Indian Institute of Millets Research (Hyderabad)	Selection from GPMR 1153
TNPSu 186	Athiyandal (Tamil Nadu)	MS 507 x MS 1211
WV 125	Waghai (Gujarat)	Local Collection from Waghai Taluka, Dangs District
JK 8 (Check)	Rewa (Madhya Pradesh)	Selection from local germplasm
OLM 203 (Check)	Berhampur (Orissa)	Pureline selection from Lakshmipur local
BL 6 (Check)	Jagdalpur (Chhattisgarh)	Paiyur 1 x OLM 29

Table 2. Geographical identity and climate variables of the locations during the crop growth period

Location	2017			2018			Latitude	Longitude	Altitude (ft)
	T Max.	T Min.	Rainfall (mm)	T Max.	T Min.	Rainfall (mm)			
Athiyandal	35.52	26.63	18.94	34.57	26.58	18.30	12.07° N	78.99° E	561
Berhampur	35.55	24.35	69.72	35.26	24.37	86.70	19.31° N	84.79° E	78
Dindori	34.52	25.69	49.63	32.59	26.98	54.23	22.94 ° N	81.06° E	2099
Jagdalpur	30.78	22.88	74.91	28.13	20.85	62.09	19.08 ° N	82.02 ° E	1811
Nandyal	33.66	25.77	40.30	34.90	24.98	16.80	15.47 ° N	78.48 ° E	666
Perumallapalle	34.72	25.75	44.88	35.33	25.96	28.07	13.60° N	79.35° E	1000
Ranchi	28.61	19.83	75.00	29.53	19.60	54.23	23.34° N	85.30 ° E	2135
Vizianagaram	30.55	27.83	39.88	30.78	28.75	60.10	18.10° N	83.39° E	242
Waghai	31.42	24.28	126.80	27.59	23.29	55.00	20.77° N	73.49° E	830

T Max.: Maximum temperature during crop period; T Min.: Minimum temperature during crop period

Table 3. Description of combination of a single year and a single location making up eighteen test environments

Code	Description
E1	Rainy season-2017, Athiyandal
E2	Rainy season-2017, Berhampur
E3	Rainy season-2017, Dindori
E4	Rainy season-2017, Jagdalpur
E5	Rainy season-2017, Nandyal
E6	Rainy season-2017, Perumallapalle
E7	Rainy season-2017, Ranchi
E8	Rainy season-2017, Vizianagaram
E9	Rainy season-2017, Waghai
E10	Rainy season-2018, Athiyandal
E11	Rainy season-2018, Berhampur
E12	Rainy season-2018, Dindori
E13	Rainy season-2018, Jagdalpur
E14	Rainy season-2018, Nandyal
E15	Rainy season-2018, Perumallapalle
E16	Rainy season-2018, Ranchi
E17	Rainy season-2018, Vizianagaram
E18	Rainy season-2018, Waghai

for days to 50% flowering, seed yield and fodder yield, respectively. The variance due to genotypes was relatively meager in comparison to the other two sources of variation, 1.41 and 11.70% for seed and fodder yield, respectively (Table 4). On contrary, 47.30% of the genotypic variance was observed for days to 50% flowering. Considerable differences in the response of genotypes across environments are indicated by a higher magnitude of GEI sum of squares than genotypes alone for seed and fodder yield (Tonk et al 2011, Alam et al 2015, Vaezi et al 2017). These pieces of evidence are suggestive for the possible existence of different mega-environments in our study (Yan and Hunt 2002, Mohammadi et al 2009). The multiplicative variance of the treatment sum of squares due to interaction was partitioned into eight significant interaction principal components for days to 50% flowering and seed yield, whereas, six for fodder yield (Table 4). The first two PCs explained 73.70, 75.30 and 76.90% of the total variation for days to 50% flowering, seed yield and fodder yield, respectively. The contribution of PC1 and PC2 was 42.00 and 31.70% for days to 50% flowering, 53.00 and 22.30% for seed yield and 59.10 and 17.80% for fodder yield.

GGE Biplot: the GGE biplot has not been implemented to analyze the MET data of little millet yield trials. Approximately, 73% of the variability of the three traits studied was captured by the first two principal components. Consequently, the number of PCs used in this study is reasonable, especially when coupled with Gollob's F-test which also suggested the usage of two PCs (Zobel et al 1988, Yan 2000).

Mean performance and stability of the genotypes across environments: The mean days to 50% flowering of the nine genotypes across the eighteen environments varied from 49.10 (JK8) to 88.40 days (OLM 217), while the seed yield varied from 1311 (JK8) to 1742 kg/ha (DHLT28-4), and the fodder yield varied from 3940 (JK8) to 8355 kg/ha (OLM 203) (Fig. 1). The range of days to 50% flowering varied from 47 (E1) to 91 (E18) days (Fig. 2). The mean seed yield per environment varied from 346 (kg/ha) in E4 to 3737 (kg/ha) in E13 (Fig. 2). The mean fodder yield per environment varied from 988 (kg/ha) in E13 to 11790 (kg/ha) in E7. The "mean *vs.* stability" biplot enables visualization of the mean performance of genotypes in addition to their stability, this is aided by the average environment coordination (AEC) abscissa that bears a single arrowhead. It also serves as a marker for the average environment and points towards a higher mean. The perpendicular lines on the AEC are referred to as AEC ordinates. The stability of a genotype is inversely proportional to the length of the AEC ordinate. The genotypes are arranged along the average environment axis based on their average seed or fodder performance across all the environments with the arrow pointing to the highest value of yield. The genotypes JK8, TNPSu186 and DHLT28-4 took least time to flower while, the genotypes OLM217, OLM203 and WV125 were late flowerings (Fig. 1 and Fig. 3). The genotypes TNPSu186, WV125 and IIMRLM7162 were highly stable for flowering. The genotypes DHLT28-4, BL6 and OLM203 produced higher seed yields, while the genotypes JK8 and TNPSu186 were the poorest seed yielders. The genotypes IIMRLM7162, OLM203 and DHLT28-4 were stable for seed yield, whereas, the genotypes WV126, WV125 and OLM217 were highly unstable. The genotypes OLM203, OLM217 and IIMRLM7162 produced higher fodder yield, while the genotypes JK8, TNPSu186 and WV125 were poor fodder yielders (Fig. 1 and Fig. 3). Most stable for fodder yield were OLM203, JK8 and WV126, while the genotypes WV125, OLM217 and BL6 were highly unstable. The genotype OLM203 produced higher seed and fodder yield, besides being stable across all the environments, it took maximum duration to flower. While the genotype DHLT28-4 was early flowering and high seed yielding with good stability across all the environments. On the contrary, the genotype

Table 4. Combined analysis of variance for days to 50% flowering, seed yield and fodder yield of little millet genotypes across eighteen environments

Source of variation	df	SS	MSS	'F' value	Probability	SS	MSS	'F' value	Probability	SS	MSS	'F' value	Probability
		Days to 50% flowering				Seed yield				Fodder yield			
Environment (E)	17	5.87×10^4	3.45×10^3	2.32×10^3	<0.001	3.63×10^8	2.13×10^7	562.45	<0.001	3.19×10^9	1.88×10^8	351.22	<0.001
Replication (Environment)	36	130.14	3.61	2.42	<0.001	3.10×10^6	8.62×10^4	2.27	<0.001	2.32×10^7	6.46×10^5	1.20	<0.001
Genotype (G)	9	9.68×10^4	1.07×10^4	7.23×10^3	<0.001	1.10×10^7	1.23×10^6	32.41	<0.001	7.01×10^8	7.79×10^7	145.47	<0.001
G × E Interaction (GEI)	135	4.91×10^4	3.63×10^2	2.44×10^2	<0.001	1.21×10^8	8.96×10^5	23.59	<0.001	1.49×10^9	1.10×10^7	20.63	<0.001
Residuals	288	428.51	1.48			1.09×10^7	3.80×10^4			1.54×10^8	5.35×10^5		

df: Degrees of freedom; SS: Sum of squares; MSS: Mean sum of squares

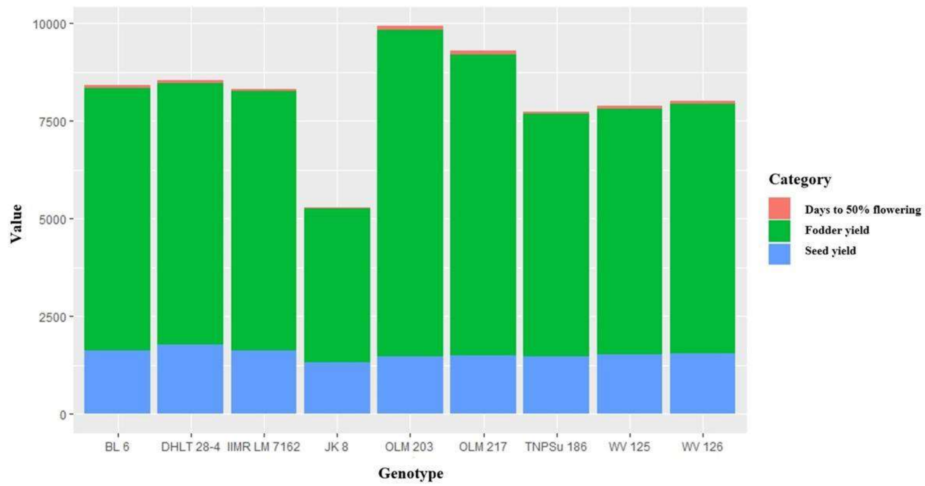

Fig. 1. Grand mean of days to 50% flowering, seed yield and fodder yield of nine little millet genotypes evaluated across eighteen environments

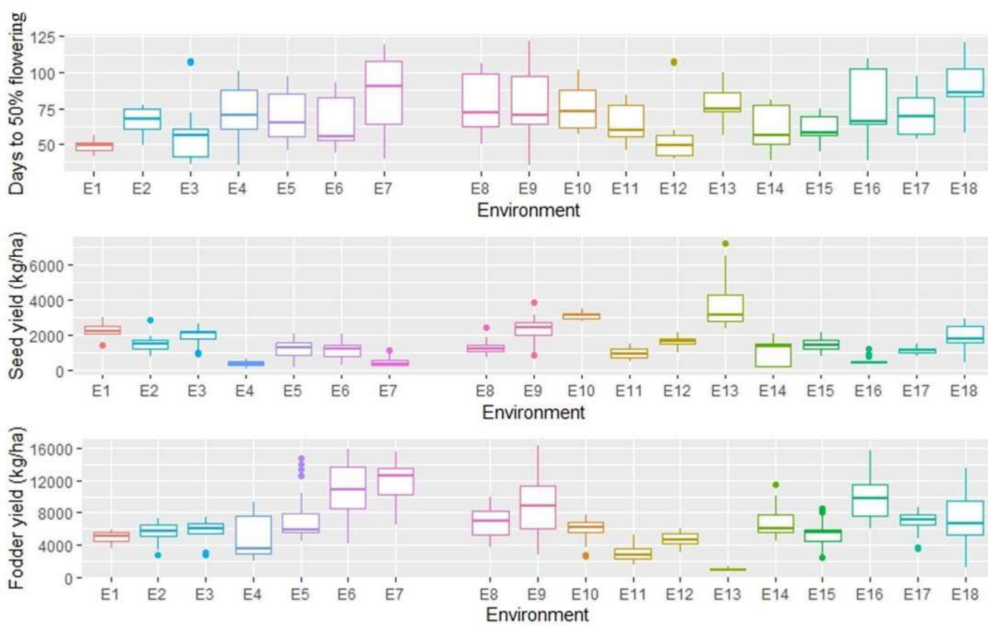

Fig. 2. Mean of days to 50% flowering, seed yield and fodder yield of nine little millet genotypes in each of the eighteen environments

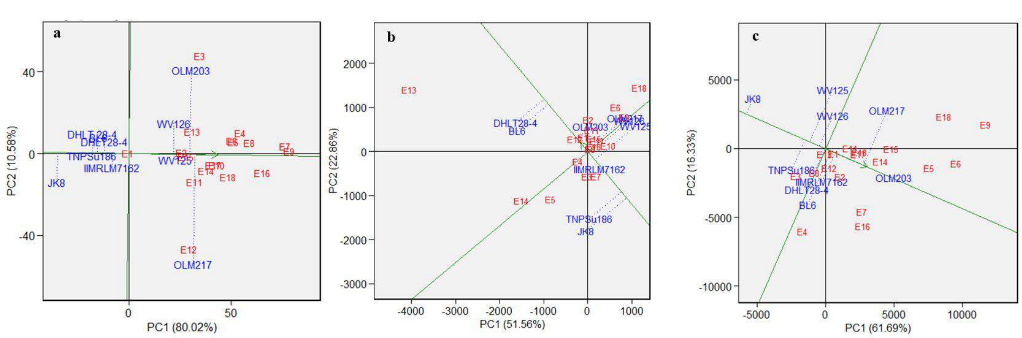

Fig. 3. Average environment coordination view of GGE-biplot based on environment-focused scaling for the mean performance *vs.* stability of nine little millet genotypes for (a) days to 50% flowering (b) seed yield and (c) fodder yield

IIMRLM7162 and JK8 were highly stable but poor seed and fodder yielders, respectively. For days to 50% flowering, wide variability was observed, thus indicating that a genotype stable for one trait may not necessarily be stable for the other. Perhaps, each trait is governed by different genes and the influence of the environment on the expression of different genes varies substantially, this is visualized by way of varying levels of stability of genotypes for seed yield, fodder yield and flowering time.

Ideal genotype: An ideal genotype is the one with a high mean yield and good stability within a mega-environment. It is present at the center of concentric circles with AEC passing through it in the positive direction and has a vector length equal to the longest vector of the genotype on the positive side of AEC (Yan and Tinker 2006). Genotypes located closer to the 'ideal genotype' are more desirable than others. The genotypes WV125 and WV126 were positioned towards the ideal genotype for flowering, although their mean days to 50% flowering across all the environment were as high, making them late flowering types and not desirable by farmers (Fig. 4a). DHLT28-4, BL6 followed by OLM203 were close to ideal genotypes for seed yield (Fig. 4b). The first two genotypes (DHLT28-4, BL6) had highest grand mean for seed yield performance. The genotypes OLM203 and OLM217 were ideal for fodder-yielding, also evident by higher grand mean for fodder-yield (Fig. 4c). The genotype OLM203 was ideal for seed and fodder yield.

Discriminativeness vs. Representativeness: To reduce the cost of genotype evaluation, it is essential to better understand the environments and determine the most

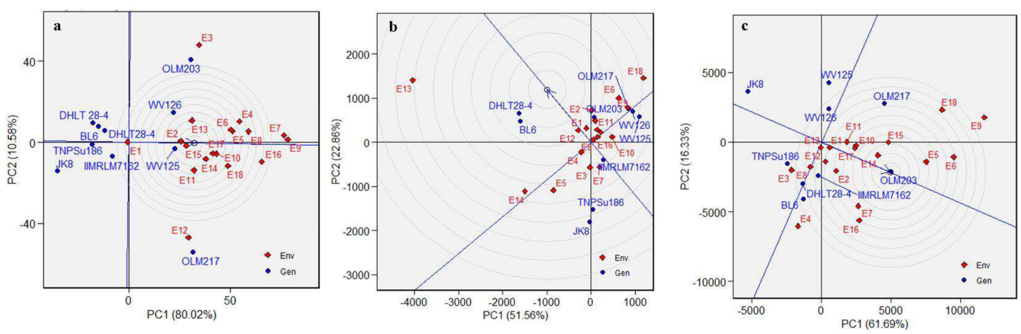

Fig. 4. GGE-biplot showing the ideal little millet genotypes based on mean (a) days to 50% flowering (b) seed yield and (c) fodder yield performance across eighteen environments

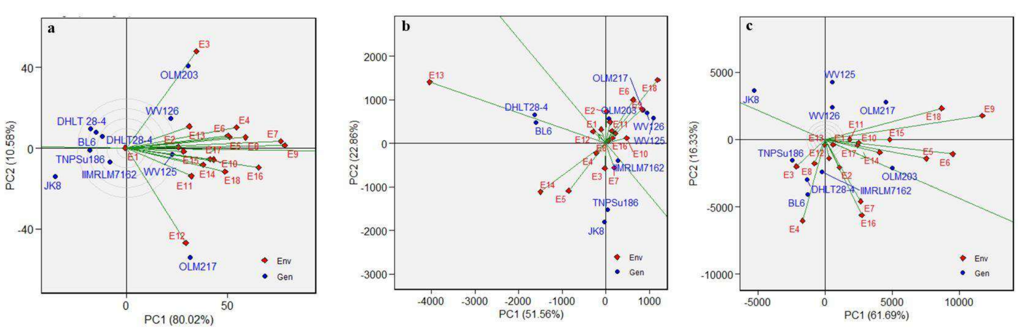

Fig. 5. Discriminative *vs.* representativeness view of GGE biplot for (a) days to 50% flowering (b) seed yield and (c) fodder yield of nine little millet genotypes evaluated across eighteen environments

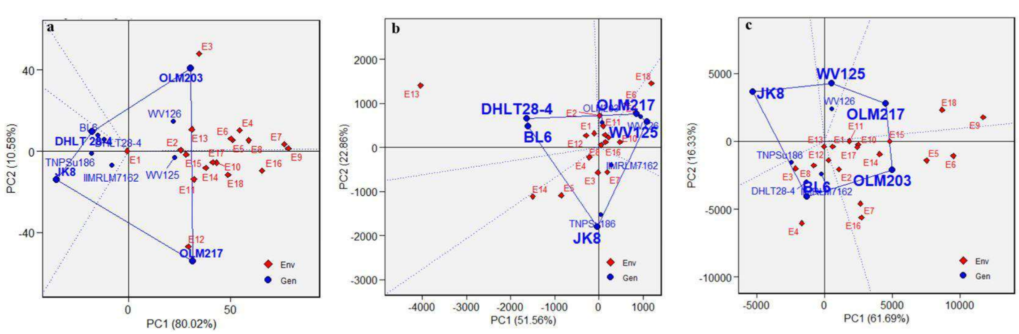

Fig. 6. Polygon view of GGE-biplot based on the symmetrical scaling for "which won-where" pattern of nine little millet genotypes and eighteen environments for (a) days to 50% flowering (b) seed yield and (c) fodder yield

Table 5. AMMI analysis of variance for days to 50% flowering, seed yield and fodder yield of little millet genotypes evaluated across eighteen environments

Source of variation	DF	Days to 50% flowering					Seed yield				
		SS	MSS	F value	Probability	Proportion (%)	SS	MSS	F value	Probability	Proportion(%)
Environment (E)	17	58724.74	3454.39	955.51	<0.0001		362211924	21306583.80	235.79	<0.0001	
Replication (Environment)	36	130.14	3.61	2.42	<0.0001		3252979	90360.52	1.98	<0.0001	
Genotype (G)	8	96839.04	10759.89	7231.54	<0.0001		6966008	870751.00	19.09	<0.0001	
G × E interaction (GEI)	136	49135.39	363.89	244.61	<0.0001		123965993	911514.65	19.99	<0.0001	
PC1	24	20702.71	828.10	556.56	<0.0001	42.00	65465318	2727721.57	59.82	<0.0001	53.00
PC2	22	15647.65	680.33	457.24	<0.0001	31.70	27576565	1253480.24	27.49	<0.0001	22.30
PC3	20	7935.08	377.86	253.95	<0.0001	16.10	13876588	693829.38	15.22	<0.0001	11.20
PC4	18	2580.55	135.81	91.28	<0.0001	5.20	5876552	326475.10	7.16	<0.0001	4.80
PC5	16	1314.42	77.31	51.96	<0.0001	2.70	5473620	342101.23	7.50	<0.0001	4.40
PC6	14	441.52	29.43	19.78	<0.0001	0.90	2528828	180630.55	3.96	<0.0001	2.00
PC7	12	354.68	27.28	18.34	<0.0001	0.70	1573295	131107.91	2.88	<0.0001	1.30
PC8	10	181.22	16.47	11.07	<0.0001	0.40	1104286	110428.58	2.42	<0.0001	0.90
Residuals	288	130.32	14.48	9.73		0.30	13131601	45595.84			
Total	621	428.51	1.48				633003554	1019329.40			
Fodder yield											
Environment (E)	17	3261623410	191860200.60	282.37	<0.0001		PC6	14	27057107	1932650.50	3.57
Replication (Environment)	36	24460446	679456.80	1.25	<0.0001		PC7	12	10864087	905340.50	1.67
Genotype (G)	8	630822910	78852863.70	145.62	<0.0001		PC8	10	3346012	334601.20	0.62
G × E interaction (GEI)	136	1498046976	11015051.30	20.34	<0.0001		Residuals	288	155949718	541492.10	
PC1	24	884824126	36867671.90	68.09	<0.0001	59.10	Total	621	7068526988	11382491.10	
PC2	22	265954092	12088822.40	22.33	<0.0001	17.80					
PC3	20	144008560	7200428.00	13.30	<0.0001	9.60					
PC4	18	123826208	6879233.80	12.70	<0.0001	8.30					
PC5	16	37743335	2358958.40	4.36	<0.0001	2.50					

DF: Degrees of freedom; SS:Sum of squares; MSS: Mean sum of squares

Table 6. Estimates of BLUP-based stability parameters of little millet genotypes evaluated under eighteen test environments and their ranks indicated in parenthesis

Genotype	Days to 50% flowering			Seed yield			Fodder yield		
	HMGV	RPGV	HMRPGV	HMGV	RPGV	HMRPGV	HMGV	RPGV	HMRPGV
BL6	59.00 (6)	0.86 (6)	0.85 (6)	944 (4)	1.01 (5)	0.93 (3)	5083 (2)	1.05 (3)	1.01 (4)
DHLT28-4	57.30 (7)	0.84 (7)	0.83 (7)	1048 (2)	1.08 (2)	1.03 (2)	5041 (3)	1.04 (4)	1.01 (3)
IIMRLM7162	60.605 (5)	0.90 (5)	0.89 (5)	1130 (1)	1.08 (1)	1.06 (1)	4607 (5)	1.01 (5)	0.98 (5)
JK8	47.20 (9)	0.71 (9)	0.67 (9)	867 (6)	0.91 (8)	0.72 (8)	2887 (9)	0.62 (9)	0.54 (9)
OLM203	81.40 (2)	1.24 (2)	1.20 (2)	748 (7)	0.89 (9)	0.79 (6)	5535 (1)	1.23 (1)	1.17 (1)
OLM217	81.80 (1)	1.25 (1)	1.21 (1)	566 (9)	0.96 (6)	0.64 (9)	4770 (4)	1.16 (2)	1.05 (2)
TNPSu186	57.00 (8)	0.83 (8)	0.82 (8)	1034 (3)	1.03 (3)	0.93 (4)	4493 (6)	0.97 (6)	0.93 (6)
W125	79.70 (3)	1.18 (3)	1.17 (3)	724 (8)	0.95 (7)	0.74 (7)	4184 (8)	0.95 (8)	0.91 (8)
W126	77.60 (4)	1.17 (4)	1.15 (4)	919 (5)	1.03 (4)	0.87 (5)	4189 (7)	0.96 (7)	0.91 (7)

discriminative and representative environments (Yan and Kang 2002). It helps to cull out the inferior genotypes from the superior ones. A discriminative environment has the ability to discriminate between test genotypes, while a representative environment represents an average of the eighteen test environments. A lower and higher discriminative ability of the environments is indicated by a shorter and longer environment vector, respectively. The most and least representative environments are indicated by smaller and larger angles between environment vectors, respectively (Yan and Tinker 2006). The environments with long vectors like E3, E12 and E9 for days to 50% flowering (Fig. 5a), while E13, E18 and E14 for seed yield (Fig. 5b) and E9, E6 and E18 for fodder yield (Fig. 5c) were most discriminating. Whereas, the environments E17, E2 and E1 for days to 50% flowering while, E12, E1 and E15 for seed yield and E17, E14 and E1 for fodder yield were nearer to the average environment indicating their representativeness. The environment E1 was representative for days to 50% flowering, seed yield and fodder yield. The environments E9, E13 and E6 were most discriminating and representative for days to 50% flowering, seed yield and fodder yield, respectively. Therefore, these environments can be used jointly as discriminative environments during early-generation testing. On the other hand, the environments that were, being discriminating and non-representative are useful for selecting specifically adapted genotypes.

Which-won-where and mega-environment identification: Polygon of the fodder yield is relatively well distributed than the polygon for days to 50% flowering and seed yield, hence making its biplot most informative as it could discriminate environments more effectively (Fig. 6c). With fewer vertices, the polygons for days to 50% flowering (Fig. 6a) and seed yield (Fig. 6b) depicted that the environments were not well separated and hence, being less informative are not discussed further. The polygon of fodder yield has genotypes JK8, WV125, OLM217, DLM203 and BL6 at the vertices. The equality lines divided the seed and fodder yield polygon into five sectors effectively, while four sectors for days to 50% flowering. Therefore, the eighteen testing environments were spread in two, four and three MEs for days to 50% flowering, seed yield and fodder yield, respectively. The ME-I of fodder yield, included the environments E9 and E18 with OLM217 as the winner, while the ME-II encompassed the environments E6, E5, E15, E14, E10, E11, E17, E1, E2, E7 and E16 with the genotype OLM203 as the winner. The third ME had the environments E13, E12, E8, E3 and E4 with BL6 as the winner. Although, METs are conducted in numerous environments, evaluation in one or two representatives of mega-environments also shall give the same results, thereby reducing the cost incurred in conducting METs. The genotypes present in a sector devoid of any environment, signified that these genotypes are not productive in any environment for any of the trait evaluated.

BLUP-based stability parameters to identify stable genotypes: The BLUP-based stability parameters such as HMGV, RPGV, and HMRPGV further represent robust statistical approaches for predicting stability coupled with adaptability and higher trait mean (Pires et al 2011; Anuradha et al 2022). An attempt was made to identify stable high seed and fodder yielding, preferably early flowering types using BLUP-based stability indices. The chief advantage of biometric approaches, such as HMGV, RPGV and HMRPGV is to disclose the randomness of the genotypic effects and to allow the ranking of genotypes in relation to their performance based on the genetic effects (Resende et al 2001). Based on all three BLUP-based stability estimates (HMGV, RPGV and HMRPGV) for both days to 50% flowering and fodder yield the genotypes OLM217 and

OLM203 were the top rankers (Table 5), whereas for seed yield, the genotype IIMRLM7162 was the top ranker, followed by DHLT28-4 (Table 5), as evident by the results of biplots and high mean performance in the field across the environments. Although the BLUP-based stability parameters were applied to various crops to estimate the stability and adaptability, but none in little millet.

CONCLUSIONS

The current study deciphered the effects of genotype × environment interaction for days to 50% flowering, seed yield and fodder yield in little millet advanced lines and checks. Identified the most stable and high seed and fodder yielding genotypes that were also early in flowering, discerned the representativeness and discriminativeness of eighteen environments for the traits evaluated. The environment and genotype × environment interaction components significantly affected the days to 50% flowering, seed yield and fodder yield. For obvious reasons, the check OLM203 produced higher seed and fodder yield but was late flowering. We recommend DHLT28-4 as an early flowering and most stable with high seed and fodder yielding ability that could be commercialized in India and can be a potential substitute for contemporary cultivars. The environments E9, E13 and E6 were both most representative and discriminative for days to 50% flowering, seed yield and fodder yield, respectively and hence can be used to recognize superior early flowering genotypes with high seed and fodder yielding adapted to specific agro-ecologies.

CONTRIBUTION OF AUTHORS

SB and NC performed data compilation and analysis. TEN, SGP, DNV and IST drafted the manuscript.

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Stability of Valepotriate Specific Valerian Chemotypes using Eberhart and Russell, AMMI, BLUP, WAAS and MTSI

Neha Mishra, Rakesh Kumar Gupta, Ashu Chandel and Subhash Sharma[1]

Department of Basic Sciences, [1]Department of Social Sciences
Dr YSP University of Horticulture and Forestry Nauni, Solan -173 230, India
E-mail: mneha6893@gmail.com

Abstract: Due to over-exploitation of *Valeriana jatamansi* rhizomes this species is now on the verge of extinction in India. For sustainable production and supply to the market/herbal industries, present study was conducted on twenty one chemotypes for four years, considering the four valepotriates. Analysis of variance based on Eberhart and Russell, additive multiplicative mean interaction was performed for all the valepotriates, which indicated highly significant Chemotype × Environment interaction for the valepotriates valtrate, acevaltrate, didrovaltrate, IVHD valtrate. The chemotypes A/F/38, D/B/10, D/B/15, U/B/1 were superior for valtrate and for acevaltrate the stable chemotypes were A/B/31, D/B/5, A/F/38, A/F/15, D/B/16, D/F/19. For didrovaltrate A/B/31, A/F/38, D/F/19, D/B/13 these four chemotypes are identified as superior and highly stable chemotypes. Seven chemotypes namely A/B/7, A/F/8, A/B/40, U/B/46, U/B/1, D/B/16 and A/B/42 were superior and highly stable chemotypes for IVHD valtrate. The three chemotypes namely A/B/31, A/B/42 and U/B/1 were superior and stable for all the valepotriates. The stable valepotriate specific chemotypes could be used for valerian varietal development programme and could ensure the preservation of the species

Keywords: Indian Valerian, Valepotriates, Stability analysis, Superior chemotypes

Valeriana jatamansi Jones is a temperate medicinal plant native to the Himalayas and found from Afghanistan to southwest China, India, Nepal, Bhutan, and Myanmar at elevations ranging from 1,000 to 3,000 meters above sea level (Jugran et al 2013). This herb, which belongs to the family Valeriaceae, was first mentioned in the 9th century by an Indian physician under the common name Indian Valerian, which is derived from the word Velo, which means "powerful drug." Although the term "Valerian" comes from the Latin word "valere," which means having aromatic or clinical properties (Bhatt et al 2012). This is a preferred sedative herb over modern medications, and can sometimes enhance the therapeutic effects of other generic drugs (Rivera et al. 2013, Jugran et al 2019). The presence of valepotriates in valerian roots/rhizome is responsible for the sedative and tranquillizing properties. These potent medicinal properties are concentrated in the herb's underground parts, such as rhizomes and roots, rather than the plant's aboveground biomass.

V. jatamansi has been collected from forests for decades, as have other Himalayan herbs, and overexploitation is causing this herb to disappear from its natural habitat. The IUCN, however, has not yet designated this herb as endangered. If this herb is not protected, it will become extinct very soon. As a result, there is an urgent need for this species' replenishment and cultivation. This species chemical diversity, in addition to its genetic diversity, has been documented (Mathela and Dev 2003, Sati et al 2005, Raina and Negi 2015). Knowledge of chemically stable genotypes (Stable chemotypes) can be applied in crop breeding programmes for varietal development, which is important in commercial cultivation of the species. The Additive Multiplicative Mean Interaction (AMMI) model has been the most commonly used (Jain et al 2017) so far for capturing chemotype and environment interaction regression models. WAAS (Weighted Average of Absolute Scores) was recently used in the AMMI I biplot instead of the first PC to obtain the entire G x E interaction variance.

Linear Mixed-effects Models (LMM) include a prediction component called Best Linear Unbiased Predictor (BLUP), which predicts a genotype's performance in a given environment. Weighting between Weighted Average Absolute Scores from BLUP (WASSB) and mean value (Y) allows for the selection of yield and stability at the same time via a superior index called WASSBY (Olivoto et al 2019a). Throughout the most situations, genotype stability is assessed using a single trait. The selection of a stable superior genotype for a greater number of traits is required in combination breeding. MTSI (Multi Trait Stability Index) identifies stable genotypes by taking into account all traits (Olivoto et al 2019b). In this paper, these analyses are used to find out the chemotypic stability of Valerian chemotypes and selection of stable superior chemotypes based on various Valepotriates for four years evaluation.

MATERIAL AND METHODS

The Department of Medicinal and Aromatic Plants conducted the experiment in Medicinal and Aromatic Plants Research Farm, Shilly, Distt. Solan, at an altitude of 1550m amsl. The geographical coordinates of the research field are latitude-N 30^0 54´ 30´´ and longitude E 77^0 07´ 30´´ Himachal Pradesh India. Plants (400-500) of different germplasms uniformly kept under the same climatic condition at Shilly farm were critically observed for different morphological features viz plant habit, type of leaf stem and root, inflorescence type and floral characteristics for quantitative assessment of distinct valepotriates from the roots of *V. jatamansi*. Plants with at least one distinguishing morphological feature were labelled and named A/B/7, U/B/1, U/F/8, and so on. Because the species is gynodioecious the first letter indicates the parent germplasm, the second indicates the plant sex (female, bisexual), and the third number indicates the plant's serial number (Raina and Srivastava 1992). A four-year quantitative study of the distinct valepotriates extracted from rhizomes of *Valeriana jatamansi* chemotypes, namely valtrate, acevaltrate, didrovaltrate, and isovaleroxyhydroxyldidro valtrate (IVHD valtrate), was conducted for four years, 2015 to 2018.

Eberhart and Russell model: Linear regression was performed using the Eberhart and Russell method. The pooled analysis of variance over the four years was done to test the chemotype(C) and the environment (E) differences against the C x E interactions for all the Valepotriates under study.

Additive multiplicative mean interaction model (AMMI): The recorded data were subjected to the AMMI analysis according to the statistical model provided by (Zobel et al1988).

Weighted average absolute scores (WAAS): The sum of squares for the C×E interaction was decomposed into single values. The IPCAs of C×E interaction from AMMI ANOVA were used to calculate Weighted Average Absolute Scores (WAAS). WAAS took the place of IPCA1 in the traditional AMMI1 biplot (Olivoto et al 2019a).

Best linear unbiased predictor (BLUP): BLUP is a prevalent method that uses a linear mixed model and treats chemotype effect as random. The components of variance were estimated using restricted maximum likelihood (Dempster et al 1977). To determine the significance of C×E and chemotype, the likelihood ratio test was used (Random effects). The chemotype's BLUP is the sum of the general mean across the chemotypic effect and the environments.

Multi trait stability index (MTSI): MTSI is calculated using factor analysis scores and the genotype-ideotype distance (Euclidian) (Olivoto et al 2019b). MTSI was computed using all valepotriates and the final loadings were calculated using the Varimax rotation criterion. WAASBY means were used to calculate the chemotype scores. The ideotype scores were calculated using the assumption that an ideotype has the highest WAASBY values (100) for all observed traits.

Software: An open-source software R 4.2.0 was used for performing all the statistical analysis utilizing package 'metan' developed by Olivoto (2019).

RESULT AND DISCUSSION

Eberhart and Russell stability model: This model was used to identify the stable chemotypes among the high valepotriates content rich chemotype by partitioning the C×E interaction into two parts (Eberhart and Russell 1966). Significant variance due to chemotypes, environment, chemotype × environment indicated the presence of variation in the mean performance of all the chemotypes over different environment and in the environment mean. C×E interaction variance was significant that suggested the differential performance of chemotypes under varying environment (Table 1). The data on the three stability parameters, i.e., phenotypic index (P_i), regression coefficient (b_i) and deviation from regression (σ^2_{di}) for different valepotriates are presented in Error: Reference source not found and the stable chemotypes for different valepotriates are presented in Table 3.

AMMI model analysis: The result of AMMI ANOVA showed that the chemotype, environment and C × E interaction effects were highly significant for the various valepotriates (Table 4). A high environment effect of 22.30% of the total sum of squares was observed for Didrovaltrate followed by IVHD valtrate (19.82%), valtrate (11.99%), acevaltrate (7.94%). Medium chemotype effect was observed for valtrate (33.70% of the total sum of squares) while high chemotype effect was observed for acevaltrate (57.25%). The interaction effect (C× E) ranged from medium (22.71%) for didrovaltrate to high (53.65%) for valtrate. The C× E was further partitioned into Interaction Principal Component Axes (IPCA) and residuals. The partitioning of C× E shows that the top three interaction principal component analysis (IPCA) scores from the AMMI model best represents the C× E patterns for valepotriates.

Weighted average absolute scores (WAAS) plot: WAAS plots mean values versus WAAS scores, with mean values on the X-axis and WAAS scores on the y-axis. WAAS is regarded as one of the most effective and efficient tools for identifying superior and stable genotypes (Abdelghany et al

2021). The WAAS index computed using GEI variance is used to estimate stability in this model (Olivoto et al. 2019b). Quadrant I (unstable), Quadrant II (unstable), Quadrant III (stable), and Quadrant IV (stable) comprise the WAAS plot (Fig. 1). Quadrant IV is the most preferred quadrant for selecting stable and superior genotypes, whereas genotypes from Quadrant III are stable but low yielders. Quadrants I and II, on the other hand, are made up of unstable but high-yielding genotypes. If the WAAS values of the genotypes are close to zero, the genotype is considered most stable (Olivoto et al. 2019b). The WAAS biplot for valtrate represents A/F/38, D/B/15, U/B/1 as the most stable chemotypes having WAAS values near to zero. The WAAS biplot of acevaltrate represented D/B/5, A/F/38, A/F/15, D/F/19 and U/F/6. The chemotypes having mean value greater than 0.5 and WAAS value near to zero was A/B/31 is considered as stable and better performing chemotype for acevaltrate. The chemotypes A/F/38, D/F/19, A/B/31 and D/B/13 are considered as stable and better performing chemotypes for didrovaltrate. The WAAS biplot for IVHD valtrate represents A/B/7, A/F/8, A/B/40, U/B/46, U/B/1 as the stable chemotypes. The chemotypes D/B/16 and A/B/42 are considered as stable and better performing chemotypes for IVHD valtrate (Fig. 1).

Table 1. Pooled analysis of variance for the valepotriates content estimated from the rhizomes of higher yielder chemotypes

Source of variation	d.f.	MSS			
		Rhizome valtrate	Rhizome acevaltrate	Rhizome didrovaltrate	Rhizome IVDH valtrate
Chemotype	20	0.571	0.030	0.437	0.553
Environment	3	1.354	0.027	1.188	1.634
C × E	60	0.303	0.006	0.061	0.142
Environment + C × E	63	0.353	0.007	0.114	0.213
Environment(linear)	1	4.063	0.082	3.565	4.902
C × E (linear)	20	0.411	0.011	0.086	0.205
Pooled deviation	42	0.237	0.003	0.046	0.105
A/B/7	2	0.150	0.003	0.023	0.102
A/B/31	2	0.034	0.002	0.008	0.050
A/B/33	2	0.538	0.011	0.367	1.436
A/B/40	2	0.146	0.000	0.001	0.006
A/B/42	2	1.217	0.002	0.030	0.028
A/F/8	2	0.048	0.001	0.012	0.018
A/F/10	2	0.064	0.002	0.050	0.076
A/F/15	2	0.129	0.001	0.084	0.191
A/F/36	2	1.319	0.019	0.128	0.043
A/F/38	2	0.079	0.004	0.010	0.006
D/B/5	2	0.007	0.001	0.001	0.007
D/B/10	2	0.184	0.000	0.016	0.004
D/B/13	2	0.065	0.000	0.006	0.006
D/B/15	2	0.087	0.001	0.117	0.059
D/B/16	2	0.065	0.002	0.061	0.065
D/F/9	2	0.223	0.005	0.003	0.024
D/F/19	2	0.259	0.002	0.008	0.001
U/B/1	2	0.199	0.001	0.000	0.050
U/B/46	2	0.156	0.000	0.003	0.024
U/F/6	2	0.009	0.005	0.003	0.004
U/F/47	2	0.003	0.001	0.026	0.011
Pooled error	168	0.001	0.000	0.000	0.002
Total	251				

Table 2. Stability parameters for different valepotriates contents estimated from the rhizomes of high yielder **c**hemotypes

Chemotypes	Valtrate				Acevaltrate			
	Mean	b_i	P_i	σ^2_{di}	Mean	b_i	P_i	σ^2_{di}
A/B/7	1.395	-1.108	0.055	0.148	0.294	-1.38	0.146	0.003
A/B/31	1.218	0.913	-0.122	0.032	0.358	0.90	0.210	0.001
A/B/33	1.506	5.322	0.166	0.537	0.260	7.28	0.112	0.011
A/B/40	1.124	1.629	-0.216	0.144	0.054	0.48	-0.094	0.000
A/B/42	2.247	1.476	0.907	1.215	0.088	1.01	-0.060	0.002
A/F/8	1.060	0.230	-0.280	0.046	0.107	1.27	-0.040	0.000
A/F/10	1.221	-0.048	-0.119	0.063	0.116	-0.71	-0.032	0.002
A/F/15	1.074	3.044	-0.266	0.127	0.178	2.42	0.030	0.001
A/F/36	2.291	2.101	0.952	1.317	0.194	2.49	0.046	0.019
A/F/38	1.474	0.131	0.134	0.078	0.188	0.66	0.040	0.004
D/B/5	1.115	0.900	-0.225	0.005	0.175	1.18	0.027	0.001
D/B/10	1.342	1.293	0.002	0.183	0.044	0.64	-0.104	0.000
D/B/13	0.985	0.324	-0.355	0.064	0.037	0.36	-0.110	0.000
D/B/15	1.524	-0.461	0.184	0.086	0.060	0.04	-0.088	0.001
D/B/16	0.906	0.970	-0.434	0.064	0.148	1.10	0.001	0.002
D/F/9	1.199	0.149	-0.141	0.221	0.149	0.58	0.001	0.005
D/F/19	1.526	-1.223	0.186	0.258	0.161	-0.30	0.013	0.002
U/B/1	1.706	0.718	0.366	0.198	0.066	0.47	-0.082	0.001
U/B/46	1.219	2.380	-0.121	0.155	0.062	0.29	-0.086	0.000
U/F/6	0.896	0.593	-0.444	0.007	0.221	1.95	0.073	0.005
U/F/47	1.110	1.667	-0.230	0.002	0.145	0.28	-0.003	0.001
A/B/7	0.286	0.022	-0.299	0.022	0.991	0.33	0.26	0.101
A/B/31	0.628	1.119	0.042	0.007	0.722	0.54	-0.01	0.048
A/B/33	0.625	1.415	0.039	0.367	1.502	3.68	0.77	1.434
A/B/40	0.170	0.327	-0.416	0.000	1.014	1.71	0.28	0.004
A/B/42	0.663	1.495	0.078	0.030	0.859	1.12	0.12	0.026
A/F/8	0.480	0.515	-0.106	0.012	0.872	0.58	0.14	0.016
A/F/10	0.504	0.440	-0.081	0.050	0.481	0.13	-0.25	0.074
A/F/15	0.647	1.303	0.061	0.084	1.235	2.59	0.50	0.189
A/F/36	1.360	2.968	0.774	0.128	0.579	0.98	-0.16	0.041
A/F/38	0.760	1.106	0.174	0.010	0.281	0.29	-0.45	0.005
D/B/5	0.445	0.719	-0.140	0.001	0.471	0.78	-0.26	0.005
D/B/10	0.411	0.879	-0.174	0.016	0.167	0.25	-0.57	0.002
D/B/13	0.726	1.271	0.141	0.006	0.488	0.44	-0.25	0.004
D/B/15	1.416	2.200	0.830	0.117	0.502	0.49	-0.23	0.058
D/B/16	0.368	0.769	-0.218	0.061	0.938	1.29	0.20	0.064
D/F/9	0.278	0.116	-0.308	0.003	0.505	0.20	-0.23	0.022
D/F/19	0.856	1.209	0.271	0.008	0.362	0.30	-0.37	-0.001
U/B/1	0.233	0.301	-0.352	0.000	1.406	1.75	0.67	0.048
U/B/46	0.198	0.290	-0.387	0.003	1.099	2.39	0.36	0.022
U/F/6	0.548	1.038	-0.038	0.003	0.510	0.56	-0.23	0.002
U/F/47	0.695	1.499	0.110	0.025	0.458	0.60	-0.28	0.009

LMM-Best linear unbiased predictor: The chemotype (C) and C × E variances were significant for all the valepotriates based on likelihood ratio test (Table 5). The contribution of environment variation was high for Acevaltrate (42.86%) followed by Didrovaltrate (36.51%). The coefficient of determination (R^2_{CEI}) of G × E interaction was found to be moderate to low.

The accuracy of selection was very high for the valepotriates didrovaltrate (0.93) followed by acevaltrate. High selection accuracy was found for IVHD valtrate (0.86) and moderate accuracy was observed for valtrate (0.68). The genotypic correlation among environments (r_{Ce}) was high for Valtrate (0.84), IVHD Valtrate (0.65) while was low for Didrovaltrate (0.32) and Acevaltrate (0.30).

The predicted mean values of the chemotypes are presented in Figure 2. Eight chemotypes had above-average predicted mean value for valtrate. A/F/36 followed by A/B/42, U/B/1 and D/F/19 had the highest predicted mean values for valtrate. Out of 11 chemotypes that had above-average predicted mean value, A/B/31 followed by A/B/7 had the highest predicted means for acevaltrate. Similarly, 10 chemotypes for didrovaltrate, 11 chemotypes for IVHD

Table 3. Stability of various chemotypes over different years for different valepotriates

Valepotriates	Suitable For				
	Rich environment		Poor environment		Stable
	Below average stability	Above average stability	Below average stability	Above average stability	
Valtrate	A/B/33, A/B/42, A/F/36,	A/B/7, A/F/38, D/B/15, D/F/19,	A/B/40, A/F/15, U/B/46, U/F/47	A/B/31, A/F/8, A/F/10, D/B/5, D/B/13, D/B/16, D/F/9, U/F/6	D/B/10, U/B/1,
Acevaltrate	A/B/33, A/F/36, U/F/6,	A/B/7, A/B/40, A/F/15, A/F/38, D/F/9, D/F/19	A/B/42, A/F/8, U/F/47	A/F/10, D/B/10, D/B/13, D/B/15, U/B/1, U/B/46,	A/B/31, D/B/5, D/B/16,
Didrovaltrate	A/B/33, A/B/42, A/F/15, A/F/36, D/B/13, D/B/15, D/F/19, U/F/47	----	----	A/B/7, A/B/40, A/F/8, A/F/10, D/B/5, D/B/10, D/B/16, D/F/9, U/B/1, U/B/46, U/F/6,	A/B/31, A/F/38,
IVHD Valtrate	A/B/33, A/B/40, A/F/15, D/B/16, U/B/46, U/B/1,	A/B/7, A/F/8,	---	A/B/31, A/F/10, A/F/36, A/F/38, D/B/5, D/B/10, D/B/13, D/B/15, D/F/9, D/F/19, U/F/6, U/F/47	A/B/42,

Table 4. AMMI anova for the various valepotriates of *Valeriana* chemotypes

Source	Df	Valtrate			Acevaltrate		
		SS	MSS	%TV	SS	MSS	%TV
Environment (E)	3	4.062	1.354*	11.99	0.08248	0.02749*	7.94
Chemotype (C)	20	11.420	0.571*	33.70	0.59488	0.02974*	57.25
CXE	60	18.180	0.303*	53.65	0.35381	0.00590*	34.05
PC1	22	12.450	0.566*	68.48	0.23600	0.01073*	66.60
PC2	20	4.400	0.220*	24.20	0.08600	0.00430*	24.40
PC3	18	1.330	0.074*	7.32	0.03200	0.00178*	9.00
Pooled error	168	0.227	0.001		0.00793	0.00005	
Source	df		Didrovaltrate			IVHD Valtrate	
Environment (E)	3	3.5651	1.1884*	22.30	4.902	1.634*	19.82
Chemotype (C)	20	8.7380	0.4369*	54.65	11.052	0.553*	44.69
CXE	60	3.6311	0.0605*	22.71	8.523	0.142*	34.46
PC1	22	2.1500	0.0977*	59.20	7.248	0.329*	85
PC2	20	1.2500	0.0625*	34.40	0.877	0.044*	10.3
PC3	18	0.2300	0.0128*	6.40	0.399	0.022*	4.7
Pooled error	168	0.0549	0.0003		0.256	0.002	

*= Significant at 0.001 probability significance level

valtrate had above-average predicted mean values. D/B/15 followed by A/F/36; A/B/31 followed by A/B/7had the highest predicted mean values for Didrovaltrate and IVHD valtrate, respectively.

BLUP (Best Linear Unbiased Predictor) is more advantageous to plant breeders in a mixed model approach because it results in more accurate predictions of chemotypes future mean values. It predicts random effects

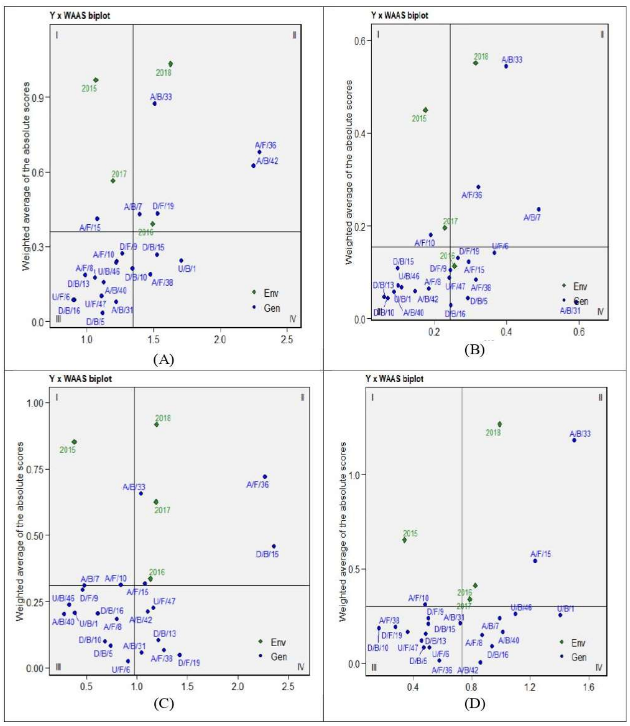

Fig. 1. Mean vs. WAAS biplot from AMMI anova for (A) Valtrate, (B) Acevaltrate, (c) Didrovaltrate, (D) IVHD valtrate

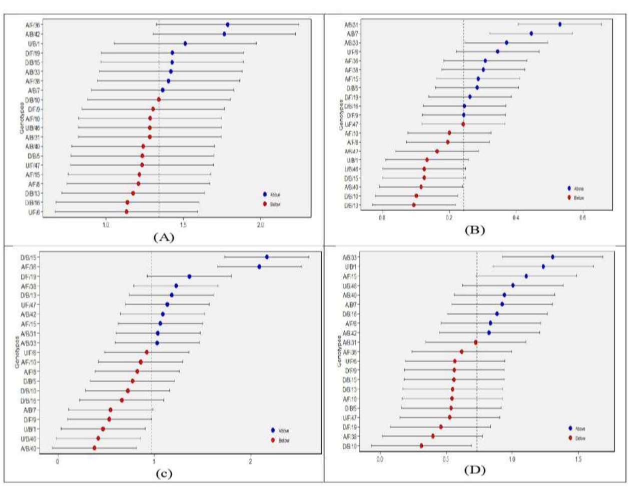

Fig. 2. BLUP mean values of Chemotypes for (A) Valtrate, (B) Acevaltrate, (c) Didrovaltrate, (D) IVHD valtrate

more accurately and works better with unbalanced or incomplete data (Smith et al. 2005). A Linear Mixed Model analysis of variance revealed significant differences between chemotypes and their interactions with an environment, similar to AMMI anova. Except for acevaltrate, the per cent of chemotypic variation was greater than the per cent of environmental variation in phenotypic variation. Albeit the contribution of the environment is less for other valepotriates, it was having a significant effect on the phenotypic expression of valepotriates. Low R^2_{CEI} of acevaltrate and didrovaltrate and moderate R^2_{CEI} of Valtrate and IVHD Valtrate demonstrated the presence of high residual variation in C X E interaction component contrasting to AMMI anova, which explained high proportion of C X E interaction through first two IPCAs.

The genotypic accuracy of selection (As), also known as model predictive accuracy, is the correlation between observed and predicted values (Olivoto et al 2019a). Moderate to very high A_s values for all the traits indicated the reliability of the model in the selection of superior genotypes. The high genotypic correlation among environments (r_{ce}) valtrate and IVHD valtrate suggested the similar trend across all the environments and easy identification of stable and superior genotypes while the low r_{ce} for Didrovaltrate and IVHD valtrate indicated difficulties in the selection of superior stable chemotypes for these valepotriates and need for detailed accurate information for selecting stable superior chemotypes.

Multi trait stability index (MTSI): MTSI was calculated based on all the valepotriates viz., valtrate, acevaltrate,

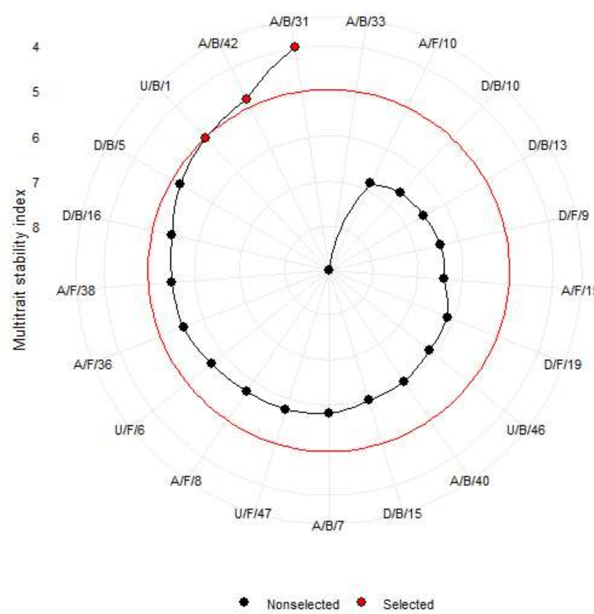

Fig. 3. MTSI values of the chemotypes

Table 5. Estimation of variance components from LMM

	Valtrate	Acevaltrate	Didrovaltrate	IVHD Valtrate
CHEM	0.066**	0.016**	0.261**	0.103**
CHEM: ENV	0.283**	0.008**	0.099**	0.121**
Error	0.055	0.018	0.207	0.064
% CV in PV	16.30	38.09	46.03	35.89
% IV in PV	69.88	19.05	17.46	42.16
% EV in PV	13.82	42.86	36.51	21.95
PV	0.405	0.042	0.567	0.287
R^2_{CEI}	0.700	0.188	0.175	0.420
A_s	0.684	0.908	0.928	0.862
r_{ce}	0.837	0.305	0.324	0.653

CV = chemotypic variation; IV = Interaction variance; PV = Phenotypic variation; EV = Environment variation ; R^2_{CEI} = co-efficient of determination of CE interaction; A_s = Accuracy of selection; r_{ce}=chemotypic correlation across environments; **=significant at 1%level based on likelihood ratio test

Table 6. Communalities and selection differential of various characters based on MTSI

VAR	FAI	Communality	Uniqueness	X_0	X_s	SD	SD (%)
Valtrate	-0.795	0.653	0.347	1.34	1.72	0.381	28.3
Acevaltrate	-0.742	0.550	0.450	0.244	0.283	0.039	16.0
Didrovaltrate	-0.696	0.851	0.149	0.976	0.847	-0.129	-13.2
IVHD valtrate	-0.155	0.922	0.0785	0.735	0.996	0.260	35.4

didrovaltrate and IVHD valtrate. A Varimax rotation criterion was applied for calculating the final loadings. The first two factors having eigenvalue more than 1.00 i.e. PC1 representing 43.5% and 30.9% variation was selected (Table 6). After Varimax rotation, the communality was ranged from 0.550 for acevaltrate to 0.922 for IVHD valtrate with a mean value of 0.743. Figure 6 shows the MTSI values of the chemotypes. The chemotypes indicated in red colour dots were selected based on their MTSI values at a selection intensity of 15 %. The stable and selected chemotypes in the order were A/B/31 followed by A/B/42, U/B/1. A selection differential of 28.3, 16.0, -13.2 and 35.4% for valtrate, acevaltrate, didrovaltrate and IVHD valtrate was observed respectively by the selection of these three chemotypes.

The lower values of MTSI indicate stable genotypes based on multiple traits. The stable and selected genotypes in the order were A/B/31 followed by A/B/42, U/B/1. It was supported by Table 4 as these were the genotypes classified as stable or fairly stable for all the traits. Besides, among these three, A/B/31 had the highest predicted mean (BLUP) for Acevaltrate, A/B/42, U/B/1 had second highest predicted mean (BLUP) for valtrate and IVHD valtrate, respectively. Therefore, the selection of these chemotypes was justified. The selection of these chemotypes would greatly benefit the improvement in mean performance as reflected by the high per cent of selection differentials.

CONCLUSIONS

A multi-year study for four years for twenty one chemotypes of *Valeriana jatamansi* using various multivariate analyses like Eberhart and Russell, AMMI, WAAS, BLUP and MTSI evaluation inculcate the stable and superior chemotypes identified through individual analysis and were combined, which identified a total of four superior and highly stable chemotypes for valtrate i.e., A/F/38, D/B/10, D/B/15, U/B/1. Similarly, A/B/31, D/B/5, A/F/38, A/F/15, D/B/16, D/F/19 in total six chemotypes are identified as superior and highly stable chemotypes for acevaltrate. For didrovaltrate A/B/31, A/F/38, D/F/19, D/B/13, these four chemotypes are identified as superior and highly stable chemotypes. Seven chemotypes namely A/B/7, A/F/8, A/B/40, U/B/46, U/B/1, D/B/16 and A/B/42 are superior and highly stable chemotypes for IVHD valtrate. BLUP identified eight above valtrate yielding chemotypes and eleven above acevaltrate yielding chemotypes. Similarly, ten chemotypes are identified as above didrovaltrate and eleven above IVHD valtrate Yielding chemotypes. Multi-trait stability index analysis identified three chemotypes (A/B/31, A/B/42 and U/B/1) as superior and stable for all the valepotriates. The selected chemotypes can be utilized for *V. jatamansi* varietal development programme and could be recommended for commercial cultivation. The multivariate techniques used in the present study proved to be very efficient to discover superior and stable genotypes while MTSI is the new technique for evaluating stability based on multiple traits for wider adaptations.

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Influence of Sorghum and Pearl Millet Mixture on Emergence Count, Yield, Competition Indices and Productivity of Crops

Manpreet Singh Brar, Naveen Kumar[1] and Shakshi Mankotia[2]

Department of Agronomy, Punjab Agricultural University, Ludhiana-141 004, India
[1]Department of Agronomy, Forages and Grassland Management, [2]Department of Soil Science
CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur-176 062, India
E-mail: msbrarbrar@gmail.com

Abstract: Forage intercropping integrates crops and livestock production because forages can be grown as intercrops with grain crops. It is characterized by rotation and diversification in time and space dimensions. The experiment on performance of sorghum in mixture with pearl millet was conducted to study the benefits of intercropping/mixed cropping. The experiment included two sowing methods (line sowing and broad cast sowing) and five seed proportions (100:0, 75:25, 50:50, 25:75 and 0:100) of sorghum and pearl millet hybrids. The sowing of sorghum pearl millet under line sowing in 25:75 seed proportion resulted in better emergence count, higher herbage and crude protein yields, higher land equivalent ratio, more net returns and B:C ratio as compared to other treatments.

Keywords: Broadcast, Emergence, Herbage, Intercrop, Mixture, Seed

Intercropping is a cropping system useful for more efficient use of resources, for stable yields particularly in problematic environments and also a method to reduce problems with weeds, nitrogen losses etc. It is a common practice among small and marginal farmers using traditional farming systems (Torkaman et al 2018). It provides farmers with a variety of returns from land and labor, and often increases the efficiency with which scarce resources are used and reduces the risk associated with a single crop that is susceptible to environmental and economic fluctuation (Khan et al 2005). Intercropping is also known to intercept more solar energy and provide comparatively higher yield stability (Tsubo et al 2013) and yield insurance during aberrant weather conditions compared with sole cropping. There is also an increasing scientific interest in intercropping systems in temperate regions for developing sustainable farming systems for forage or grain production (Neugschwandtner et al 2014). The type of inter/mixed crop and the spatial arrangement of inter/mixed crop have significant effects on the balance of competition between the components of the crop and their productivity. In order to achieve the best results, a rational approach is needed to obtain information on the appropriate inter/mixed crop population (Guleria et al 2018). Cereals intercropping with legumes result in increased resource capture by component crops and improve soil microbial activity along with better efficiency of resource conversion which triggers higher biomass production (Iqbal et al 2019). Among cultivated fodder crops sorghum (*Sorghum bicolor* L.) is an important *kharif* season crop which gives good biomass in first cut in most of the cultivars, but the regeneration capacity of this crop is not so good. However, pearl millet [*Pennisetum glaucum* (L.) is one of the important millets crop of hot and dry areas of arid and semi-arid climatic condition (Yadav et al 2018). It has good regeneration capacity after first cut and gives good tonnage in subsequent cuts compared to sorghum. This is the best meal for animals during the summer season and is usually grown in rain-fed areas of Punjab and excellent in providing a lot of dry matter (Islam et al 2018). In forage production, profitability is of the utmost importance and cereal- cereal or fodder intercropping has been reported to increase economic returns. Greater productivity per unit area by sorghum-soybean intercropping systems resulted in 46% higher monetary returns than their sole cropping (Iqbal et al 2017). Sowing methods as well as seed rates or seed proportions of different component crops in mixed cropping play integral role in the production potential of crops as well as entire cropping system. Most of the farmers in hilly areas due to lack of mechanization and ease of operation opt for broadcast sowing of crops. Different planting patterns and varying seed rates/proportions for inter/mixed cropping have pronounced results on the potential yield advantage of mixed cropping over sole cropping of each component crop. Generally, both sorghum and pearl millet are sown as sole crops for fodder or as main crops, being intercropped with legumes with a view point of enhancing overall production of the system and soil fertility. However, the information on the mixed cropping of sorghum and pearl millet particularly with respect to quantity of seed of each species to be used as well under different methods of sowing is lacking. Hence, in this study, the multicut sorghum with pearl millet hybrids under different sowing methods and seed proportions have been investigated.

MATERIAL AND METHODS

Site of experiment: The present study was conducted at CSKHPKV, Palampur, Himachal Pradesh during kharif season of 2016 & 2017 to evaluate the yield and quality attributed of fodder mixture sown under different sowing methods. The experimental site was located at 32° N latitude and 76° E longitude at an elevation of about 1227 meters above mean sea level in North-Western Himalaya. The site falls in the mid-hills sub-tropical zone of Himachal Pradesh.

Soil characteristics: Before the commencement of the experiment, soil samples from 0-15 cm depth were collected randomly from several spots and composite soil sample was prepared. This composite soil sample was dried, ground and sieved through 2 mm sieve and then subjected to chemical analysis to determine the fertility of the experimental site. Soil of the experimental field was high in organic carbon, acidic in reaction, low in available nitrogen, medium in available phosphorus and high in available potassium (Table 1). In treatments comprised of sorghum and pearl millet, the seed of each crop was computed as per the respective proportion of seed in each treatment. In pure stand, sorghum and pearl millet were sown as per the sowing methods, whereas, in mixed treatment, seed of sorghum and pearl millet as per respective proportion was mixed and then sown in lines at 30 cm spacing or by broadcast as per the treatments.

A) Sowing method (2)		B) Seed proportion (5)		
		Sorghum	:	Pearl millet
		100	:	0
Line sowing	x	75	:	25
Broadcast sowing		50	:	50
		25	:	75
		0	:	100

Emergence count: The emergence count in each plot of each crop was taken 15 days after sowing. In line sown plots, the count was taken from 1m row length from two places and average value of two was expressed as count m^2. In broadcast sowing, plant count was taken using a quadrant measuring 25 cm x 25 cm and expressed as emergence count m^2.

Green fodder yield: In all, two cuts of both the crops were taken. The crop from net plot was harvested and weighed. The total yield for each plot was adjusted by including the fresh weight of samples, taken for various observations. The cut wise as well as total yield of all cuts was converted into quintals per hectare.

Quality studies: The crude protein yield (q ha^{-1}) was calculated:

Crude protein yield = per cent crude protein content × dry fodder yield (q ha^{-1})

Net returns: The treatment wise net returns were worked out by subtracting the cost of cultivation from the gross returns of the respective treatment expressed as net returns in Rs. ha^{-1}.

Net returns = Gross returns - Cost of cultivation

Benefit cost ratio: The benefit cost ratio was obtained by dividing the net returns with the treatment wise cost of production in Rs. ha^{1}.

Land equivalent ratio: The land equivalent ratio was obtained by dividing the intercrop yield of each crop by the yield of pure crop.

Relative crowding coefficient: The relative crowding coefficient is calculated by the following formula:

$$\text{RCC (Sorghum)} = \frac{\text{Mixture yield of sorghum x proportion of pearl millet sown}}{\text{(Yield of pure sorghum-mixture yield of sorghum) x proportion of sorghum sown}}$$

$$\text{RCC (Pearl millet)} = \frac{\text{Mixture yield of millet x proportion of sorghum sown}}{\text{(Yield of pure pearl millet-mixture yield of pearl millet) x proportion of pearl millet sown}}$$

Here, RCC= Relative crowding coefficient

Per day productivity: Per day productivity is calculated by

Table 1. Chemical properties of soil of the experimental field

Particulars	Values	Methods used for analysis
Chemical analysis		
Organic carbon (%)	1.11	Rapid titration method (Walkley and Black 1934)
pH	4.5	Glass electrode pH meter (Jackson 1967)
Available nutrients (kg ha^{-1})		
Nitrogen	272.0	Alkaline permanganate method (Subbiah and Asija 1956)
Phosphorus	12.0	Ammonium molybdate blue colour method (Olsen et al 1954)
Potassium	283.0	Neutral normal ammonium acetate extraction method (AOAC 1970)

dividing the total green fodder yield by number of days taken by crops season.

RESULTS AND DISCUSSION

Emergence count: The emergence count of sorghum and pearl millet (m^2) at 15 days after sowing was significantly more under line sowing than broadcast sowing (Table 2). The emergence count for sorghum and pearl millet line sowing was 39 & 48, respectively as compared to 33 and 44 under broadcast sowing which indicated 18.2 and 9.1 per cent higher emergence count when sown in lines than the broadcast sowing for sorghum and pearl millet, respectively. In line sowing, proper placement of seed in soil would have resulted in better germination under line sowing which would have caused better emergence count than broadcast sowing. Ayub and Shoaib (2009) also observed better plant density of sorghum + cluster bean in line sowing over broadcast sowing due to unequal depth of sowing under broadcast sowing. In case of seed proportions, the mean emergence count of both the crops decreased significantly and consistently with decreasing seed proportion of crops in mixture, which agreed with the seed proportion of crops used for sowing in each treatment *i.e.* emergence directly corresponds to seed count of particular crop used under a specific seed proportion. The emergence count of sorghum was significantly higher under sole sorghum followed by 75:25 seed proportion and being lowest sorghum count per square meter under 25:75 seed proportion while vice versa was true for pearl millet with highest emergence count under sole stand of pearl millet and least emergence count under 75:25 seed proportion.

Total green and total dry fodder yield: The interaction effect of treatments on total green fodder and dry fodder yields was non-significant. The pooled data analysis indicated that the line sowing of crops using 25:75 seed proportion produced significantly higher mean green and dry fodder yields which were statistically at par with sole pearl millet sown under line sowing (Table 3) while the other seed proportions including 50: 75:25 & sole sorghum incurred the lower green and dry fodder yields, respectively symbolizing that the crop mixture sown with higher proportion of pearl millet was best due to regenerative and fast growing capability of pearl millet. Moreover, this could be attributed to higher growth indices like plant height, shoot population etc. under the respective treatment. Significantly minimum mean green and dry fodder yields were obtained in broadcast sown sole sorghum observed under the pooled analysis of both the years. Reza et al (2013) in Tehran (Iran) observed higher fresh and dry weight of sorghum when grown with lima bean (*Phaseolus lunatus*) in additive series at different planting proportions of crops. Ganvit et al (2018) conducted a field experiment and studied the performance of forage-based intercropping of oat (*Medicago sativa* L.) – lucerne under different row proportions which resulted in significantly higher green fodder yield (991.14 q/ha) of oat and lucerne along with significantly higher dry fodder yield (114.12 q/ha) of oat under 2:1 row proportion of oat + lucerne.

Crude protein yield: The pooled data analysis depicted significantly higher crude protein yield (12.02 q ha^{-1}) in line sown sole pearl millet as compared to other seed proportions, being statistically at par with line sown sorghum + pearl millet in 25:75 seed proportion which incurred a crude protein yield

Table 2. Effect of sowing methods and seed proportions of crops on emergence count of sorghum and pearl millet (m^{-2}) 15 days after sowing, land equivalent ratio (LER), relative crowding coefficient (RCC) indices and per day productivity of green fodder under the crop mixture (Pool data for 2 years)

Treatment	Sorghum	Pearl Millet	LER	RCC (Sorghum)	RCC (Pearl millet)	Per day productivity (q ha^{-1} day^{-1})
Sowing methods						
Line sowing	39	48	1.07	1.15	1.75	3.89
Broadcast sowing	33	44	0.90	0.74	0.97	3.26
CD (p=0.05)	2.82	1.66	0.07	0.18	NS	0.17
Seed proportions						
100:0	54	--	--	--	--	2.68
75:25	43	21	0.99	0.78	1.35	3.37
50:50	28	41	0.90	0.72	1.00	3.39
25:75	18	50	1.07	1.34	1.73	4.31
0:100	--	73	--	--	--	4.10
CD (p=0.05)	3.98	2.36	0.09	0.23	NS	0.27

of 11.82 q ha^{-1} (Table 3). The line sown 25:75 seed proportion resulted in 31.2 per higher mean crude protein yield as compared to the nearest seed proportion of 50:50 under line sowing while the lowest quality fodder i.e., pooled crude protein yield was attained in sole sorghum sown by broadcast sowing (Table 3) which was even 19.6 per cent lower in protein yield as compared to the similar sole crop stand of sorghum practiced under the line sowing. This might be attributed to the fact that the pearl millet fodder is more palatable and nutritious than sorghum crop being more dry and hard fodder which resulted in higher mean crude protein yields under high pearl millet seed proportions while the line sowing resulted in proper placement & distribution of seed count indicating high protein yields in line sown crops as compared to the broadcast sowing. Khalatbari et al (2009) also reported higher percentage of digestible dry matter and carbohydrate in 75 per cent sorghum + 25 per cent pearl millet combination but the highest percentage of crude protein was observed in sole sorghum. Sobkowiz et al (2016) also indicated the increased protein yields when oat, wheat, triticale were sown in intercrop mixtures as compared to the respective sole stand of each crop. Ganvit et al (2018) observed that oat and lucerne in 2:1 row proportion recorded significantly higher crude protein and crude fibre content as compared to other treatments at first and second cut, respectively.

Economics: The perusal of data clearly indicated that planting of sorghum + pearl millet in lines using 25:75 seed proportion was the most remunerative treatment in terms of net returns and B:C ratio. The pooled net returns of Rs. 75,669 per hectare and the respective highest B:C ratio of 3.33 was observed (Table 3) under the line sowing of 25:75 seed proportion which was closely followed by line sown sole pearl millet incurring the net returns of Rs. 72,151 per hectare and B:C ratio of 3.23. Pure sorghum sown by broadcast method appeared least profitable treatment which recorded a huge decrease in net returns by 78.3 per cent as compared to the respective pure crop of pearl millet sown under broadcast sowing. On the contrary, Guleria (2013) observed that broadcast sowing of sorghum and cowpea crops using 75 per cent of recommended seed rate of cowpea resulted in higher profitability. The results could be pertained to the higher green fodder yield under respective treatment resulting in higher gross returns which indicated the higher net returns and B:C ratio under 25:75 seed combination sown in lines. Similarly, Deori *et al.,* (2019) recorded significantly higher net profit and benefit-cost ratio under sole cropping of setaria and intercropping of hybrid napier with setaria in alternate row and column method. Iqbal et al (2017) observed the greater profitability of sorghum-soybean intercropping systems obtaining more than 40 per cent higher monetary returns than their sole crop stand.

Competition indices: Line sowing of crops produced significantly higher mean LER and RCC values than broadcast sowing. There was 18.9 per cent higher LER and 55.4 per cent higher RCC of sorghum under line sown crops as compared to broadcasting the seed mixture (Table 2). Sowing of sorghum + pearl millet using 25:75 seed ratio had the highest land equivalent ratio of 1.07 due to higher green & dry fodder yield in 25:75 seed combination under mixture, which was closely followed by sorghum + pearl millet sown with seeding ratio of 75:25 in land equivalent ratio. The minimum f land equivalent ratio was observed in broadcast sown sorghum + pearl millet in 50:50 seed proportion. The higher mean values of crowding coefficient of pearl millet

Table 3. Effect of sowing methods and seed proportions of crops on total green fodder yield , total dry fodder yield, total crude protein yield, net returns and B:C ratio of crop mixture (Pool data for 2 years)

Treatment	Total green fodder yield (q ha^{-1})	Total dry fodder yield (q ha^{-1})	Total crude protein yield (q ha^{-1})	Net returns (Rs. ha^{-1})	B:C
T_1- Sole sorghum- Line sown	258.15	61.72	7.07	39068	1.78
T_2- Sole sorghum- Broadcast sown	218.27	51.78	5.91	31528.5	1.57
T_3- S:PM ::75:25- Line sown	320.71	73.45	8.44	54289	2.52
T_4- S:PM ::75:25- Broadcast sown	279.13	63.93	7.49	45057.5	2.16
T_5- S:PM ::50:50- Line sown	330.48	74.05	9.01	56471	2.63
T_6- S:PM ::50:50- Broadcast sown	272.04	61.16	8.11	43307	2.08
T_7- S:PM ::25:75- Line sown	417.88	93.24	11.82	75669	3.33
T_8- S:PM ::25:75- Broadcast sown	349.63	78.16	10.25	60748	2.80
T_9- Sole pearl millet- Line sown	401.32	88.33	12.02	72151.5	3.23
T_{10}- Sole pearl millet- Broadcast sown	327.79	72.64	10.0	56221.5	2.67
CD (p=0.05)	33.45	7.71	1.11	7951.5	0.36

under different seed proportions indicated dominance of pearl millet crop in comparison to sorghum in a mixed cropping. Relative crowding coefficient of 1.73 in 25:75 seed proportion indicated highest dominance of pearl millet in this treatment with higher seed count of pearl millet under 25:75 seed proportion followed by significantly higher green as well as dry fodder yields under the respective crop mixture proportion. Similarly, the greater productivity per unit area or higher competition indices like LER were obtained by sorghum-soybean intercropping systems as compared to the sole cropping by Iqbal et al (2017)**.**

Per day productivity: The line sowing of crops resulted in 19.33 per cent higher pooled per day productivity as compared to the broadcast sown mixture (Table 2). Better crop stand & growth of plants followed by significantly higher total green fodder yield under line sowing would have resulted in better per day productivity on mean basis as the productivity on per day basis directly corelates to every single growth and yield attribute throughout the crop cycle. Among seed proportions, sowing of crops in 25:75 seed proportion had significantly higher per day mean productivity of 4.31 q $ha^{-1}day^{-1}$ and was statistically at par with sole stand of pearl millet exhibiting the pooled per day productivity of 4.10 q $ha^{-1}day^{-1}$. The d per day productivity of 25:75 seed proportion was 5.12, 27.14, 27.89 and 60.82 per cent higher than sole pearl millet, 50:50, 75:25 and sole sorghum, respectively due to higher green fodder yield, better shoot population and other yield attributes being higher under 25:75 seed proportion. Similarly, Ganvit et al (2018) recorded a higher per day productivity of intercropping of oat (*Medicago sativa* L.) – lucerne under 2:1 row proportion of oat + lucerne as compared to the sole stand of forage crops.

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Performance of Special Rice Varieties under Different Transplanting Dates in Lower Altitude Conditions of North Kashmir

Haseena Shabnam, S. Sheraz Mahdi*, Rukhsana Jan, Waseem Raja, Mohammad Anwar Bhat, Shaista Nazir[1], Faheem Jeelani[2], Owais Ali Wani[1] and Bhagyashree Dhekale[2]

Division of Agronomy, [1]Division of Soil Science, [2]Division of Agricultural Statistics, Faculty of Agriculture, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir (SKUAST-K), Wadura-193 201, India
*E-mail: syedapbau@gmail.com

Abstract: Sowing environment and varieties are the two important factors that influence growth and final yield of rice (*Oryza sativa* L.). In this study, a field experiment was conducted during *Kharif* -2020 at experimental farm, Faculty of Agriculture, SKUAST-K, Wadura to study the performance of special rice varieties to different sowing windows under lower altitude conditions of Kashmir. The treatments comparing of three high value rice cultivars viz. Mushkbudji, SKAU-494 and Zag/Red rice, transplanted at three dates viz. 06th June (early), 16th June (mid) and 26th June (late), laid out in a factorial randomized complete block design. The rice crop transplanted early (6th June) did better than those transplanted in latter weeks of June in terms of growth, phenology, grain yield and quality. Variations among cultivars were also observed with SKUAST-494 registered statistically significant higher grain yield of 58.92q/ha. The cultivar also took longer time to attain different phenological stages than the other two cultivars. However, rice-*Mushkbudji* recorded better aroma score and less amylose content compared to other two cultivars. This high value rice varieties when transplanted early (June 6) under lower altitudes reached to their higher grain yields with better quality parameters and could be best alternative options to traditionally grown rice in the region due to their aroma and taste and will also be beneficial to farmers for realizing the higher income in the market.

Keywords: Special rice, Lower altitude, Sowing environment, Growth and phenology, Yield and quality

Rice (*Oryza sativa* L.) is accounting for 21% of global calorie intake while using 11% of global cropland (FOA 2020). In India, where rice is a staple food for more than 70% of population is bestowed with enormous genetic diversity of aromatic and non-aromatic rices and is home to at least 50,000 rice land races (Shikari et al 2014). But in recent decades, north-west Himalayan region witnessed significant genetic erosion and a loss in area under local scented and non-scented landraces (Rana et al 2000). Scented rice constitutes a small but important subgroup of rice genotypes traditionally grown by farmers, accounting for approximately 4% of total global rice. Of these, temperate regions produce 2.2% and the expression of aroma and texture is considered to be richer (Jena and Mackill 2008). The global scented rice market is growing at the rate of 12% per annum and there is a scarcity of scented rice other than Basmati that the country has to offer. Kashmir Valley being temperate region has bestowed with more than 100 land races (Parray & Shikari 2008). Amongst, Mushkbudji rice, Kamad, Nun-beoul and Zag are the most popular and enjoy enormous demand in the market due to aroma and are effective means to enhance and reform the farmer's income. At present, the area under these valuable heritage rice varieties which happens to be around 10, 000 ha during 1970s across Kashmir is dwindling and has reached to 400 ha only (MRCFC 2014). In addition, their cultivation has been confined in some specific mid-altitude (1750-1990 meters) pockets of Kashmir (Village Sagam of south Kashmir) and current yields obtained by farmers are not satisfactory (1.5-2.0 q/ha) compared to earlier yields (3.0-3.5 q/ha) due to climate variability and susceptibility of these rices to blast which accounts for 70% yield loss (Ali et al 2009).

Yield and aroma of aromatic rices is mostly determined by environmental conditions, which in turn vary in Kashmir valley due to rugged topography. Temperature is believed as most critical factors affecting yield and aroma particularly at flowering, grain filling and maturity (Chakrabarti et al 2010, Pradhan et al 2017). This challenge become more greater in a region that is increasingly becoming warmer. As Kashmir valley is now witnessing clear signs of climate change as evident by increasing temperatures and decreasing precipitations (Mahdi et al 2022). Increasing temperatures due to climate change affects rice productivity and quality (Okada et al 2011, Wu et al 2015). Thus, present study aims to explore options for cultivation of some special rice varieties beyond their niche areas and to determine the optimum transplanting date and its effect on growth, phenology, yield and quality of these high value rices under lower altitude conditions of Kashmir valley.

MATERIAL AND METHODS

The field experiment was carried out at the Crop Research Farm, Division of Agronomy, Faculty of Agriculture, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Wadura, Sopore, during *Kharif* 2020, located in the northern part of the Jammu and Kashmir region (Fig. 1) at lower altitude (1690 meters). The treatments included three transplanting dates viz. D_1: June 6 (early), D_2: June 16 (mid) and D_3: June 26 (late) and three special rice varieties viz. V_1: Mushkbudji, V_2: SKAU-494 and V_3: Zag (red rice). The experiment was laid out in a factorial randomized complete block design (RCBD) with three replications. The soil of the experimental site was silty clay loam in texture, neutral in reaction with medium available nitrogen (432.88 kg ha^{-1}), phosphorus (9.1 kg ha^{-1}) and potassium (218.60 kg ha^{-1}). Weather data were obtained from a weather station set up at the experimental site during the period of May to October of 2020, indicating that the crop had experienced mean maximum and minimum temperature of 28 and 11°C, average precipitation of 12.74 mm and mean maximum and minimum relative humidity of 74 and 50% during the growth period (Fig. 2).

The thirty days old treated seedlings (Mancozeb 75 WP @ 3g/litre of solution for 48 hrs as seed treatment) were transplanted on a well puddled soil at a hill spacing of 20 × 15 cm with two-three seedlings per hill. Fertilizer with a uniform dose of 120 kg N, 60 kg P_2O_5 and 30 kg K_2O and 25 kg $ZnSO_4$/ha was applied in SKAU-494 variety and 70 kg N, 60 kg P_2O_5 and 30 kg K_2O and 20 kg $ZnSO_4$/ha was applied in remaining two varieties. N was applied through urea, P through DAP and potash through MOP. One third dose of nitrogen and full dose of phosphorus, potassium and zinc

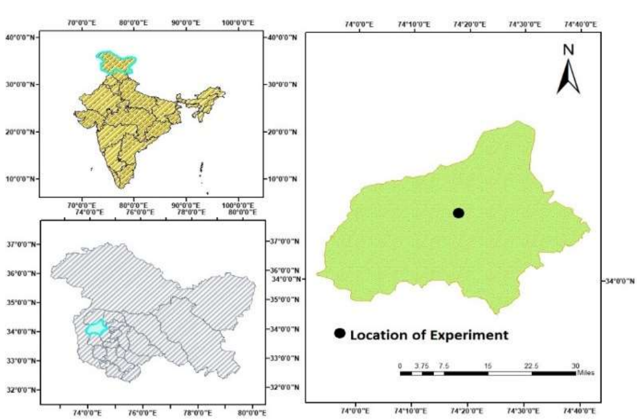

Fig. 1. The Geographical location of the experimental site

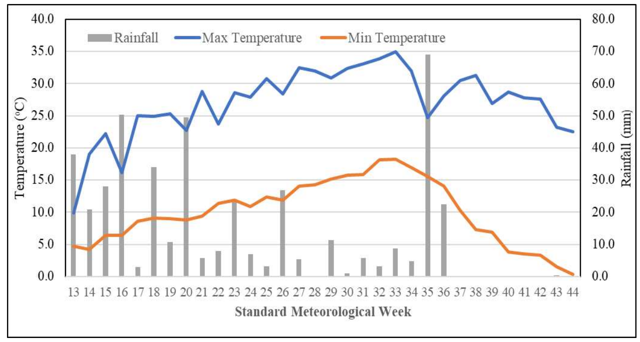

Fig. 2. Weather conditions prevailed during the crop growth period of *Kharif* 2020

was applied as basal application just before transplanting. The remaining dose of nitrogen was applied in two equal splits at 22 and 43 days after transplanting (DAT). To control the blast disease, three sprays of Tricyclazole 75 WP @ 6g/10 litre of water at 25-30 days interval after transplanting were also applied. The crop was harvested at maturity and threshed as per schedule. The data on growth (plant height, leaf area index, yield attributes viz. number of panicles/m2, grains/panicle, 1000-grains weight, yield and quality of rice were recorded following standard procedures. Data from crop samples were analyzed using SAS 9.0 (SAS 2003).

RESULTS AND DISCUSSION

Rice growth parameters: Various transplanting dates and cultivars caused substantial variations in growth parameters. The crop transplanted early recorded maximum plant height and LAI values compared to mid and late transplanting (Table 1). Decrease in plant height and LAI in late transplanting might be due to shorter growing period. Early transplanting crop may have enjoyed the better environmental conditions especially the temperature and solar radiation which resulted maximum height and LAI (Hossain and Sikdar 2009, Suleiman et al 2014). Among the cultivars *Mushkbudji* rice at par with *Zag* (red rice) recorded maximum plant height. However, LAI values were significantly higher in SKUA-494 than other two cultivars. This might be due to the inherent character of the cultivar as also supported by Murshida et al (2017).

Rice yield attributes: Yield attributes viz. number of effective tillers m^{-2}, panicle length, panicle weight and 1,000-grain weights showed that crop transplanted on June 6 was significantly superior over late transplanting (June 16 and June 26) in respect of all yield contributing characters (Table 1). Less number of effective tillers, panicle length, weight and grain weight in late transplanting was due to less production of photosynthates due to shorter growing period (Yawinder et al 2006). The varieties also differ significantly in respect of yield contributing characters. SKUA-494 recorded the maximum number of effective tillers m^{-2}, panicle length, panicle weight and 1,000-grain weight than other two varieties. This differences in yield contributing characters among varieties might be attributed to their genetic variability (Diaz et al 2000).

Rice phenology: Figure 3 shows the influence of the sowing date and rice varieties on the number of days required to complete different wheat growth stages: maximum tillering, panicle initiation, anthesis, and physiological maturity. Crop sown transplanted early prolongs maximum tillering, panicle initiation, anthesis, and physiological maturity compared to crop transplanted in mid to end June. Wani et al (2016) reported that the overall growth and phenology period decreased with delayed in transplanting in rice. Among the three genotypes, SKUA-494 took maximum number of days to attain different phenological stages viz. maximum tillering, panicle initiation, anthesis, and physiological maturity compared to other two rice varieties. This might be due to genotype makeup which varies from one variety to other (Rezaei et al 2018).

Rice yield: A comparison of the transplanting date and varieties showed significant variation (Table 2). Crop transplanted early recorded significantly higher grain yield (59.29 q ha^{-1}), straw yield (88.92 q ha^{-1}), and harvest index (40.54%) as compared to mid to late June transplanting. The average grain yield superiority of early transplanted rice crop was 22.41 and 36.29% over mid to late June transplanted crop respectively. Such yield penalty beyond June 16 could be mainly explained in terms of reduction in yield–determining attributes primarily caused by shortened duration of various phenophases of crop development, and higher temperatures during the milking to grain–filling stage (Wani et al 2016). The genotypes, SKAU-494 recorded a significantly higher yield for grain (58.92 q ha^{-1}), straw yield (91.80 qha^{-1}), and harvest index (39.51 %) over *Mushkbudji*

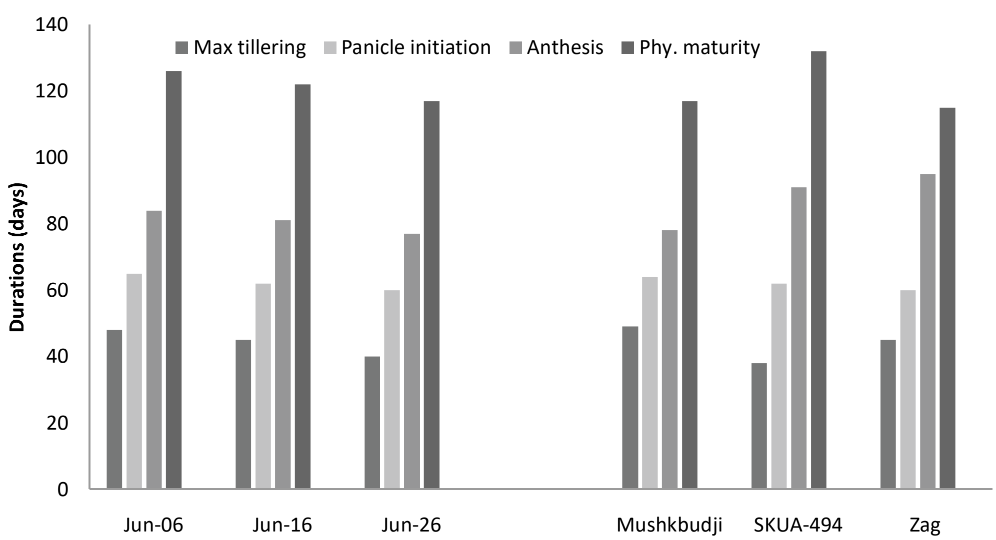

Fig. 3. Effect of transplanting dates and varieties on different phenological stages of rice

Table 1. Growth and yield attributing characters of special rice varieties influenced by transplanting dates

Treatments	Plant height (cm)	Leaf area index	No. of effective tillers m^{-2}	Panicle length (cm)	Panicle weight (g)	1000-grain weight (g)
Transplanting dates						
6 June	124.21	1.90	329.55	25.85	3.43	21.15
16 June	114.12	1.81	326.33	23.08	2.81	18.95
26 June	103.84	1.76	299.88	21.66	2.58	17.62
CD (p=0.05)	1.28	0.02	5.31	1.85	0.36	0.41
Varieties						
Mushkbudji	125.68	1.69	310.11	21.38	2.62	19.30
SKAU-494	103.45	2.16	340.88	28.94	3.61	20.01
Zag	113.04	1.62	304.77	20.27	2.59	18.41
CD (p=0.05)	1.28	0.02	5.31	1.85	0.36	0.41

Table 2. Yield and harvest index of special rice varieties influenced by transplanting dates

Treatments	Grain yield (q ha^{-1})	Straw yield (q ha^{-1})	Harvest index (%)
Transplanting dates			
6 June	59.29	88.92	40.54
16 June	46.00	78.19	36.70
26 June	37.77	68.20	34.99
CD (p=0.05)	5.31	6.22	4.39
Varieties			
Mushkbudji	45.39	74.38	37.44
SKAU-494	58.92	91.80	39.51
Zag	38.75	69.13	35.28
CD (p=0.05)	5.31	6.22	4.39

Table 3. Quality parameters of special rice varieties influenced by different dates of transplanting

Treatments	Amylose content (%)	Aroma score	Gelatinization temperature (°C)	Alkali spread value
Transplanting dates				
6 June	21.57	2.00	76.89	2.18
16 June	20.44	1.77	76.44	2.42
26 June	19.17	1.22	75.33	3.05
CD (p=0.05)	0.42	0.39	0.63	0.05
Varieties				
Mushkbudji	17.50	3.00	76.89	1.63
SKAU-494	25.24	1.89	75.22	3.34
Zag	18.43	0.11	76.56	2.68
CD (p=0.05)	0.39	0.39	0.63	0.05

rice and Zag (Table 2). This variation in yield in different genotypes could be due to genetic makeup of a variety (Ganajaxi et al 2001, Hoque et al 2003).

Rice quality: Significantly higher amylose content (21.57%), aroma score (2) and gelatinization temperature (76.89°C) and lower alkali spread value (2.18) were observed in crop transplanted in early June than mid to late June transplanting (Table 3). Higher temperatures at the time grain filling stage experienced by crop transplanted late reduced the quality characters including aroma (Singh et al 2013, Kumar et al 2014). Among varieties investigated, SKAU-494 reported significantly higher amylose content (25.24%) than other two varieties. However, significantly higher gelatinization temperature (76.89^{0}C), lower alkali spread value (1.63) and high aroma score (3.0) was recorded in Mushkbudji rice compared to SKAU-494 and Zag rice and is attributed to inherit traits of a particular variety (Arif et al 2019).

CONCLUSIONS

The three widespread special rice varieties under different transplanting dates responded to their growth, phenology, yield and quality. These high value rice varieties viz. *Mushkbudji* rice, SKUA-494 and Zag / red rice, when transplanted early (June 6) reached to their higher grain yields with better quality parameters. For this reason, if these varieties were to be sown under lower altitude conditions at the proposed time, they could be best alternative options to traditionally grown rice in the region due to their aroma and taste and will also be beneficial to farmers for realizing the higher income in the market.

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Prospects and Challenges of Emerging Insect Pest Problems *vis-à-vis* Climate Change

Yousra Mukhtar, Uma Shankar* and Zahid Abass Wazir

Division of Entomology, Faculty of Agriculture, Sher-e-Kashmir University of Agricultural Sciences & Technology Chatha, Jammu-180 009, India
**E-mail: shankaruipm@gmail.com*

***Abstract*:** Emerging insect pests are a constant threat to mankind and their sudden upsurge and outbreak not only affects the crop yield and biodiversity but also poses a constant threat to global trade in agriculture. According to an estimate, these pests cause diverse losses ranging from 18-20 per cent in Indian agriculture. It is evident from the literature that American bollworm, *Helicoverpa armigera*; Brown plant hopper; *Nilaparvata lugens*, Diamondback moth, *Plutella xylostella*; Pink stem borer, *Sesamia inferens*; Whitefly, *Bemisia tabaci*; Wheat aphid, *Macrosiphum miscanthi* and many more have emerged as key pests on various crops in recent years. Recent incidence of Fall Armyworm (*Spodoptera frugiperda*) on maize and other crops has also drawn attention of researchers and policy makers to issue a nation-wide advisory to the farming community to safeguard their produce as well as to combat against this dreaded pest. Various activities such as excessive usage of fertilizers, faulty cropping pattern, introduction of high yielding and hybrid varieties, indiscriminate and injudicious use of pesticides, etc. have had a tremendous impact on resurgence, resistance and secondary outbreak of insect pests. However, one of the main reasons, which has usually been neglected by us, for changing status and scenario of insect pest species is Climate aberration or in broader perspective 'Climate Change'. This article discusses the issues concerned to some emerging insect pests, their probable reasons of outbreak and providing the way out to devise timely and effective ecological based Integrated Pest Management solutions to mitigate the losses throughout our country.

***Keywords*:** Emerging insect pest, Outbreak, Invasive pest, Pesticide, Climate change

Frequent outbreaks of emerging insect pests have drawn attention of scientific community as well as policy makers to mitigate the damage inflicted to the cultivated crops and thereby, enhance the crop productivity to feed the rapidly growing population. The status of the pest changing from minor to major or from secondary to primary is labelled as an Emerging Insect Pest. Due to certain anthropogenic activities like changes in crop cultivation practices, low key crop management practices or due to natural reasons which are not in favour of a particular pest, the other insect pests find enough quantity of resources to feed on in absence of the major pest and reproduces more rapidly, thus, emerging as major or key pests.

Global losses due to pests: Since the dawn of agriculture, the constant interaction of crops with pests has been a cause of concern to farmers as they inflict considerable loss and destruction to cultivated crops, livestock, stored products, cattle feed, etc. These pests can either be insects, pathogens, weeds, virus, fungi, nematodes, protozoa, or any other plant which is not needed in a particular crop ecosystem at that specific time. Among various categories of pests, maximum crop losses are done by insects (34 per cent) followed by pathogens (31 per cent), weeds (27 per cent) and virus (8 per cent) respectively. In terms of monetary value, Indian agriculture suffers an annual loss of about US$ 36 billion (Dhaliwal et al 2015). These destructive organisms reduce the quality and quantity of crop produce, hinder the viability of cultivated plants and thus, threaten the sustainability of human beings.

Change in insect pest scenario: During the pre-green revolution era (1960's), insect pests damaged food crops to a greater extent. Among the crops, maximum losses were reported in cotton (18 per cent) followed by groundnut, pulses, rice, maize, millets, wheat and sugarcane, respectively, thus accounting to an average loss of 7.2 per cent in the food crops due to insect pests. However, in the Post-Green revolution era, the losses due to insect pests gained an upsurge and thus increasing the average loss in food crops to 23.3 per cent with almost 50 per cent damage in Cotton crop alone due to insect pests (Dhaliwal et al 2010). Certain reasons have been attributed to this changing insect pest scenario *viz*., monoculture practices (Andow 1983), excessive use of fertilizers (Butler et al 2012), use of high yielding varieties (Halder and Rai 2021), non-judicious use of pesticides (Gangwar et al 2014), absence of natural enemies (Bommarco et al 2011), favourable weather conditions (Harish et al 2015) and climate change (Peace 2020). Among all these reasons, climate change is the most indubitable reason behind gradual change in the status of pests.

Climate change and projections: Climate change is no hoax. The on and off fluctuations in day-to-day weather phenomena are vividly visible. As defined by Intergovernmental Panel on Climate Change (IPCC 2007), 'Climate Change is any change in Earth's climate over time, whether due to natural conditions or because of human activity'. The upsurge in the figure of greenhouse gases *viz*., CO_2, CH_4, N_2O, etc. leads to greenhouse effects which promotes global warming. This is the prime reason for Climate change. Temperature, carbon-dioxide and precipitation are important climate projections wherein the fluctuations drastically lead a direct impact on climate change. The constant increase in various climate projections alters the incidence of crop pests' accordingly, thus impairing the sustenance of ecosystem. For example, the average global temperature has increased by 0.6°C and is expected to reach 1.1-5.4°C by the end of next century. Similarly, the carbon-dioxide concentration has increased from 280ppm to 360 ppm and is expected to reach 655 ppm by 2070. The number of rainy days has decreased by more than 15 days following a more erratic distribution. The anthropogenic emission of greenhouse gases has upsurged over a period of forty years (1970-2010). In 2010, the greenhouse gas emission has reached to 49 ± 4.5 $GtCO_2$-eq/yr. Of this, fossil fuel combustion and industrial processes contributed about 78 percent of the total increase in greenhouse gas emissions (IPCC 2014). Various policy makers and scientists have been assembled across the globe to frame out timely strategies and policies to mitigate the deleterious effects of climate change.

Climate change-Role in emerging pest problems in India: As a reason of changing climatic conditions, the insects have become much more serious in the crop ecosystem and have developed the status of major or key pests. The insect pests that have emerged as or are likely to emerge as key or serious pests due to climate change are tabulated in Table 1.

There have been certain instances of emerging insect pest problems as reported by Sharma (2016) at ICRISAT, Hyderabad. For instance, *Helicoverpa armigera* damage in pigeon pea has prevailed following wet weather conditions in Sep-Oct; Mealy bug, *Ceroplastodes cajaninae* infestation in pigeon pea has been reported under prolonged hot and dry conditions; Beet army worm, *Spodoptera exigua* damage in chickpea has been reported to be triggered by winter rains on Oct-Nov and Pink stem borer, *Sesamia inferens* damage in sorghum has been reported to emerge due to hot and dry conditions post-rainy season.

Recent pest outbreaks in relation to climate change in India: India has witnessed several instances of insect pest outbreak in the past two decades which has caused immense loss to agriculture and horticulture sector (Table 2, Fig. 1).

Sugarcane woolly aphid: Sugarcane woolly aphid, *Ceratovacuna lanigera* Zehntner (Aphididae: Hemiptera), otherwise known as 'white sugarcane aphid' is a prime pest of the family Poaceae; however, it has been found to feed on plants of other families as well like Bixaceae, Theaceae and Combretaceae. In India, its presence has been witnessed on ten different plant species. It undergoes parthenogenetic reproduction and has an anholocyclic life cycle without any

Table 1. Insects pests that have emerged and are likely to emerge as key or serious insect pests

Insect pest	Scientific name	Crop(s)
American bollworm	*Helicoverpa armigera*	Cotton, chickpea, tomato, etc.
Beet armyworm	*Spodoptera exigua*	Chickpea in southern India
Spotted pod borer	*Maruca vitrata*	Pigeonpea, cowpea.
Diamondback moth	*Plutella xylostella*	Cabbage, cauliflower
Pink stem borer	*Sesamia inferens*	Maize, sorghum, wheat
Whitefly	*Bemisia tabaci*	Cotton, tobacco
Brown planthopper	*Nilaparvata lugens*	Rice
Green leafhopper	*Nephotettix spp.*	Rice
Serpentine leaf miner	*Liriomyza trifolii*	Cotton, tomato, several other vegetables
Fruit fly	*Bactrocera spp.*	Fruits and vegetables
Mealy bugs	*Paracoccus marginatus*	Several crops
Thrips	Several species	Groundnut, cotton, chillies
Wheat aphid	*Macrosiphum miscanthi*	Wheat, barley, oats
Pod sucking bugs	*Clavigralla* spp.	Pigeon pea

Courtesy: Sharma (2016)

sexually producing generation. This pest was first reported in India from West Bengal in 1958 as a minor pest on Sugarcane (Basu and Banerjee 1958). Afterwards, it spread to Tripura, Assam, Uttar Pradesh, Arunachal Pradesh and Sikkim till 1979 but was still considered a minor pest. This pest damages the plant by congregating on both sides of the leaves and sucking up the plant sap and as a result, the plant turns yellow, loses its vigour resulting in reduced crop yield. Under severe infestation, the plant wilts and dies off. The infested leaves develop sooty mould and as a result turn black which causes reduced photosynthesis, thus, affecting the yield of the crop (Patil 2002). In 2002, the outbreak of sugarcane woolly aphid was noticed on sugarcane in Maharashtra from where it spread to Karnataka and gained the status of a major pest (Joshi and Viraktamath 2004). Very soon, this pest damaged almost entire of the crop in these two states. By 2003-04, in Maharashtra, 2,67,000 ha area under sugarcane got infested, wherein parts of Pune and Solapur were affected the most. In Karnataka, an area of approximately 61,000 ha under sugarcane was affected during the same year wherein Belgaum and Bidar were severely affected (Sharanabasappa et al 2009). The introduction of bio-control agents viz., *Dipha aphidivora*, *Chrysoperla* spp., Coccinellids, Syrphid flies and some spiders in Maharashtra and Karnataka provided effective control over woolly aphids.

Rice plant hoppers: Brown plant hopper, *Nilaparvata lugens* Stal and White-backed plant hopper, *Sogatella furcifera* Hovarth (Delphacidae: Hemiptera) are two categories of plant hoppers that are important pests in rice ecosystem. Plant hoppers are sucking insect pests that suck the sap out of the plant, thus reducing its vitality and vigour. The most important characteristic symptom of plant hopper is 'hopper burn' which is characterized by dried, dead necrotic patches in the crop field. Its first serious epidemic occurred in 1973 over half a million-ha resulting in 10-70 per cent loss in grain yield in Indian condition. It was then succeeded by a series of Brown Plant Hopper outbreaks in Karnataka (1975 and 1985), Andhra Pradesh (1976-1983), Telangana (1980), Madhya Pradesh and Orissa (1976) and Tamil Nadu (1977 and 1982). However, in 2008-09, the epidemic form of plant hoppers occurred in Northern India and as a result, more than 3,33,000 ha rice crop failed badly (Rathee and Dalal 2018). The outbreak of plant hoppers has been attributed to several reasons viz., high humidity (>90%), high temperature (25-32°C), overuse of nitrogenous fertilizers, reduced spacing (15x10 cm), non-judicious use of pesticides, hoppers tolerance to neonicotinoids and increased mortality of natural enemies (Chander and Patel 2009). Randhawa et al (2015) reported that temperatures of 26.4-30.0℃, relative humidity between 55-99% accompanied with few showers of rain are conducive for Brown Plant Hopper population build-up. Prasannakumar et al (2012) studied the impact of elevated carbon-dioxide on brown plant hopper population on Pusa Basmati-1 during 2010-11 and opined that at an elevated carbon-dioxide concentration (570±25 ppm), the population of nymphs and adults, fecundity rate and honey dew production was three to four times more than at ambient concentration of carbon-dioxide (380±25 ppm). Similarly, Guru Pirasanna et al (2016) also studied the impact of elevated carbon dioxide and temperature on brown planthopper population in rice ecosystem in open-top

Table 2. Recent insect pest outbreak with respect to climate change in India

Insect pest	Host plants	Region/ Location	Probable reasons	Impact	Reference
Sugarcane woolly aphid, *Ceratovacuna lanigera*	Sugarcane	Maharashtra and Karnataka	Abnormal weather condition.	30% yield losses	Joshi and Viraktamath (2004)
Plant hoppers, *Nilparvata lugens; Sogatella furcifera*	Rice	North India	Abnormal weather conditions	Crop failure on >33,000 ha	IARI (2008)
Mealybug, *Phenacoccus solenopsis*	Cotton, Vegetables	Punjab, Haryana	Hot and dry weather	30-40% loss	Dhawan and Saini (2009)
Tobacco caterpillar, *Spodoptera litura*	Soybean	Maharashtra	Abnormal weather conditions	25-100% yield loss	Singh et al (2012)
Spiralling whitefly, *Aleurodicus disperses*	Tapioca	Kerala	Abnormal weather conditions	Significant yield loss	Palaniswami et al (1995)
Fall armyworm, *Spodoptera frugiperda*	Maize	Karnataka	Abnormal weather conditions	About 58% yield loss	Dhar et al (2019)
Knot grass moth, *Acronicta rumicis*	Apple	Kashmir	Abnormal weather conditions	Significant yield loss	Dar et al (2019)
Thrips, *Thrips parvispinus*	Chilli	Andhra Pradesh	Overuse of insecticides	Significant yield loss	Sireesha et al (2021)

A. Brown plant hopper infestation on paddy stem and leaves B. Hopper burn symptoms C. Infestation of *Spodoptera frugiperda* on maize D. *Phenococcus solenopsis* on Malvaceae family E. Spiralling whiteflies F. Infestation of Spiralling whiteflies on guava G. *Spodoptera litura* predation by *Eocanthecona furcellata* on soybean H. *S. litura* infestation during flowering period on soybean

Fig. 1. Emerging insect pests in India

chambers during rainy season of 2013 and found that at elevated carbon-dioxide condition (570±25 ppm+>3℃), the mean brown plant hopper population and fecundity rate of females after seven weeks of adult release was relatively more than at ambient carbon-dioxide concentration (397±25 ppm+>3℃). Use of resistant varieties like Daya, Lalat, Jyothi, Karthika, Makon, Remya, Mansarovar, etc. in hopper endemic areas, augmentation of natural enemies viz., parasitoids like *Gotnatocerus* spp. (egg parasitoid), *Pseudogonatopus* spp. (Larval parasitoid), and predators like long-jawed and orb spider and mirid bug, *Cyrtorhinus lividipennis*, crop rotation, early planting, sensible and split application of nitrogenous fertilizers can mitigate the infestation of rice plant hoppers (Prakash et al 2014).

Cotton mealy bug: The cotton mealybug, *Phenacoccus solenopsis* Tinsley (Hemiptera: Pseudococcidae) is a serious polyphagous pest of approximately 21 different agricultural and horticultural crop families (Arif et al 2009). The damage is caused by both nymphs and adults by sucking up the plant sap from leaf tissues resulting into drying and wilting of plant followed by significant yield loss. They also secrete honeydew which causes sooty mould to develop leading to reduced photosynthesis (Wang et al 2009). *P. solenopsis* is an exotic insect pest species that got introduced from USA (Nagrare et al 2009). In 2006, it was reported from Pakistan and subsequently from Gujarat, followed by Punjab and Haryana (Monga et al 2009). In 2007, the infestation by two mealybug species viz., *Maconellicoccus hirsutus* and *Phenacoccus solenopsis* was reported in nine cotton-growing states of India wherein *P. solenopsis* was the predominant species (Nagrare et al 2009). Dhawan et al (2007) reported 30-40% losses in cotton yield due to mealybugs in Punjab. There has been severe economic damage to *Gossypium sp.* with 50% reduction in yields during 2006 due to infestation of mealy bug in Gujarat (Jhala 2008). In 2011 and 2014, the mealybug damage was reported on Jute in West Bengal, India (Satpathy et al 2016). The probable reasons of its outbreak on jute as reported by Gotyal et al (2014) were warm and dry conditions during seedling stage, sudden upsurge in the maximum and minimum temperature and reduced rainfall and number of rainy days from Jan-May. This pest can be effectively controlled by suitable Integrated Pest Management programmes. Proper field sanitation and removal of infested plants from field helps to reduce the pest establishment and infestation. Use of biological control agents like *Anagyrus kamali* Mani, *Cryptolaemus montrouzieri* (Mulsant), *Verticillium lecanii* (Zimmermann), *Beauveria bassiana* (Vuillemin), *Aenasius bambawalei* Hayat and *Chrysoperla carnea* (Stephens) can prove beneficial in successfully reducing the infestation by this pest on cotton (Joshi et al 2010).

Tobacco caterpillar: Tobacco caterpillar, *Spodoptera litura* Fabricus (Lepidoptera: Noctuidae) is a polyphagous pest and has emerged as a major pest of tobacco, cotton, rice, maize, soybean and groundnut over the years. This pest causes economic crop losses ranging from 25.8 to 100 per cent based on crop stage and its infestation level in the field (Natikar and Balikai 2015). There was an outbreak of tobacco caterpillar (*Spodoptera litura*) on soybean crop causing disease epidemic in Vidarbha region of Maharashtra in August 2008 (Singh et al 2012). Srinivasa Rao et al (2013) studied the impact of elevated CO_2 on tobacco caterpillar, *Spodoptera litura* on peanut, *Arachis hypogea* and reported that at 550 and 700 ppm of CO_2, the relative consumption rate and total consumption of larva was significantly more than at ambient CO_2 level. The impact of elevated CO_2 on growth parameters of *Spodoptera litura* on peanut revealed that at 500 and 700 ppm of CO_2, larval weight (g), larval duration (days) and pupal weight (g) was significantly more than at ambient concentration. Anurag et al (2020) reflected eco-friendly management of tobacco caterpillar by installing pheromone traps in the field to trap the males of *S. litura* along with spray of bio-pesticide formulation, *Bacillus thuringiensis* var. *kurstaki* for effective control of this pest.

Spiralling whitefly: The spiralling whitefly, *Aleurodicus dispersus* (Aleyrodidae; Hemiptera) is true to its name due to characteristic egg laying spiral pattern. It is a polyphagous pest that paved its way from Sri Lanka into India in 1993 on tapioca plantations of Kerala (Palaniswami et al 1995). Soon, it spread to all of the southern states of India followed by Orissa and north-eastern regions (Pradhan and Borthakur 2020). Nymphs and adults cause damage by sucking up the cell sap from the leaves, thus inflicting significant damage to different crops including cassava, chillies, mulberry, guava, banana, papaya, groundnut etc. in peninsular India (Mani and Krishnamoorthy 1999). The infestation of whiteflies depends directly on weather conditions (Mani 2010). Due to wide host range, it becomes very difficult to control this pest. Chandel et al (2010) proposed integrated Pest Management techniques to control whitefly by integrating environmental manipulation, enhancement of natural enemies and area-wide control programmes. Biological control is quite effective means of controlling whiteflies. Sugiyama et al (2011) documented three parasitoid species viz., *Eretmocerus mundus*, *E. eremicus* and *Encarsia formosa* in reducing the number of whiteflies. Very recent research has found a mite, *A. swirskii* as an effective bio-control agent of thrips and whiteflies in many crops (Messelink et al 2008). The use of yellow sticky traps is advocated to trap the adults of whitefly due to their attraction towards yellow colour (Barbedo 2014).

Fall armyworm: Fall Armyworm, *Spodoptera frugiperda* Smith (Lepidoptera: Noctuidae) is the latest threat that the agriculture sector is facing. It is a polyphagous pest attacking almost every known crop family. This pest is characterized by four spots arranged in a square or trapezoid form on its eighth abdominal segment. The neonate larvae initially damage the crop by scrapping the leaf chlorophyll. The mature larvae feed on the older leaves and create window-pane like symptoms. The continuous feeding by the larvae produces moist saw dust-like frass in funnel and as a result, there is no cob formation. The outbreak of fall armyworm was first noticed in Africa in 2016 wherein 43 countries were severely affected (Dively 2018). Sisay et al (2019) surveyed various districts of Ethiopia, Tanzania and Kenya in 2017 for mean percent of fields infested by fall armyworm (FAW) along with its level of infestation in these three mentioned countries of Africa and reported that almost 100 per cent fields were infested in all the surveyed districts of the above three countries. Being highly migratory in nature, this pest rapidly spread to the nearby continents and in 2018, its outbreak was seen in Asia wherein 9 countries were severely infested including India (Dively 2018). Dhar et al (2019) reported 6 to 58 per cent yield loss in maize in India due to infestation by this pest and reported that high humidity and moderately high temperature is suitable for the spread of fall armyworm. Extreme hot areas are not suitable for the survival of this pest and it requires an optimum temperature of about 28^{o}C for spread (CABI 2019). The emergence of pest in soil is directly proportional to temperature and inversely proportional to relative humidity (CABI 2019). Firake et al (2019) suggested hand picking and egg mass destruction as an effective control measure against fall armyworm. He further demonstrated the importance of intercropping maize with leguminous crops in breaking the food chain of fall army worm. FAO (2018) reported that bio-pesticide formulations of *Bacillus thuringiensis*, *Beauveria bassiana* and Baculo viruses can effectively control this pest.

Knot grass moth: Knot grass moth, *Acronicta rumicis* Linnaeus (Noctuidae: Lepidoptera) is emerging pest of Apple plantation in Kashmir valley. This pest is widely distributed throughout the world- India (Kashmir, Himachal Pradesh), Europe, Campbellpur, Eurasia, etc. Knot grass moth larvae actively damage newly planted apple trees. They infest the terminal shoots of the developing seedling resulting in undesired apical dominance and stunted growth of the plants. In heavy infestation, heavy economic losses are inflicted. Dar et al (2019) surveyed apple plant nurseries of various districts of Kashmir from 2014-16 growing season to determine the severity of this pest in Kashmir valley. The studies indicated that out of 634 plants observed, 197 were found infested and 29 plants were severely damaged, thus revealing a total of 31.07 and 14.03 per cent incidence and severity, respectively. The computation of percentage incidence and severity contributions by each district towards the total incidence and severity in the region revealed that Ganderbal district had highest incidence and severity percentage followed by Srinagar while least incidence and severity of this pest was reported from Pulwama.

Chilli thrips: Recent outbreak of *Thrips parvispinus* (Thysanoptera: Thripidae) on chilli in south India has drawn attention of scientists and policy makers towards the emerging and invasive insect pest in posing threat to the biosecurity of Indian agriculture. In this context, Tobacco thrips, *T. parvispinus* paved its way into India in 2015 on *Carica papaya* L. plantations, wherein its invasion caused appreciable losses to the agriculture sector. Since its invasion, this pest has been reported in many fruit, ornamental and field crops. However, a recent outbreak of chilli thrips, *Thrips parvispinus* (Thripidae; Thysanoptera) has been witnessed in Andhra Pradesh which created havoc among chilli growers. Earlier, *Scirtothrips dorsalis* was the only species of thrips affecting chilli and no species of flower thrips was seen infesting this crop till 2020. However, in January 2021, several areas of Guntur district in Andhra Pradesh noticed a new species of thrips in chilli ecosystem which was later identified to be *Thrips parvispinus*. This pest primarily infests the leaves where it scraps the chlorophyll on lower leaves and sucks up the cell sap due to which the corresponding part of upper side of leaves turns yellow and the undersurface of the leaves turn reddish-brown. The leaf lamina becomes distorted and exhibits necrotic areas. Besides leaves, the floral parts are also affected by this pest where it scraps the petals causing brownish streaks on them. They may also affect pollination by feeding on the flower pollen, which affects the fruit set (Sireesha et al 2021). Scientists suggested that indiscriminate and non-judicious application of insecticides in chilli ecosystem caused this pest to resurge in epidemic form. However, it is not hidden that the changing climatic conditions favoured the survival and development of this pest and different climatic projections and their fluctuations can be attributed as a concerning reason for epidemic upsurge in *T. parvispinus.* As such, planting of pest free seedlings should be advocated. Consequently, regular monitoring and proper surveillance of this crop is required to avoid further spread of this pest to uninfested areas. Botanicals like neem oil, Pongamia oil and microbial pesticides like *Beauveria* sp. may be the potential drivers in mitigating the resurgence of this emerging insect pest and add value in sustainable cultivation of commercial chilli.

CONCLUSION

Climate change through fluctuations in climate projections *viz*., warm temperature, undistributed precipitation and increased CO_2 concentration drastically affects the insect pest population dynamics, leading to the emergence of new pests. Sucking insect pests which were earlier considered to be minor pests have now gained the status of major pests in the past two decades such as mealy bugs, plant hoppers, thrips, etc. If required measures are not taken, most pests will become ubiquitous and disperse within the places where the climate is favourable and the preferred hosts are available. Thus, there is an urgent need to modify crop production and protection measures with changing climate to tackle the problems of emerging pests in modern agriculture so as to ensure better crop protection and productivity.

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Impact of Institutional Development Programmes on Rural Livelihoods of North-Western Himalayan State, India

Gagan Mehta, Manoj K Vaidya and Pravidhi Sharma[1]

Department of Social Sciences, [1]Department of Environmental Sciences
Dr. Yashwant Singh Parmar University of Horticulture and Forestry Nauni, Solan-173 230, India
E-mail: vidhusharma461@gmail.com

Abstract: For uplifting the rural sector of our country, the Ministry of Rural Development and the Government of India in coordination with Department of Rural Development and Department of Land Resources have been carrying forward various schemes. These schemes are formulated to benefit the citizens of rural India who will eventually become the pillars of Indian Economy in the long run. Therefore and attempt has been made in this paper to analyze the impact of different institutional development programmes on the livelihood of rural households of Himachal Pradesh. Primary data was collected based on stratified multistage random sampling from 360 households. To test the significant impact of different Governmental development programmes on sampled households Response Priority Index (RPI) was used. Among all the government sponsored schemes, the schemes which had significant impact on livelihood of sample households were Kisan Credit Card Scheme (KCC), Horticulture Training and Extension Service, Sub-Mission on Agricultural Mechanization, Mahatma Gandhi National Rural Employment Guarantee Act (MGNREGA). So the rural people should be made more aware of these schemes in order to accomplish their overall growth.

Keywords: Government policies, Rural households, RPI, Awareness

Government interventions are required to correct any distortions in the distributive mechanism resulting from a variety of imperfections coming into play while any programme is in actual implementation. The State of Himachal Pradesh has reoriented its approach and has endeavored to build an environment favorable for realizing the objective of overall development on sustainable basis. Role of the State has undergone a change from 'mere policing' till the early forties of the twentieth century to being a facilitator in the rapidly emerging market oriented, highly competitive and relatively open environment. Till late eighties of the twentieth century, the State had been 'enforcing' multiple interventions affecting almost all the aspects of life when the process of liberalization and structural reforms started gathering momentum (Anonymous 2015). The multiple interventions not only made people heavily dependent on these, but State's policy was also dependent on the outcome of its own policy without having any consideration for the active market forces. Constantly improving indicators of availability of health and education services as reflected in high ranks among Indian States is the result of serious commitment of the State Government in this regard. Support in the form of sector specific capacity building programmes enabling rural population enhance their capabilities is also available. Support in the form of sector specific subsidies and grants are available for the vulnerable Strata of the society pursuing livelihoods in these sectors. This support helps vulnerable sections compete in the relatively open and competitive markets. Direct interventions are also available through various poverty alleviation and wage employment programmes as are available in other parts of the country. To generate employment, alleviation of poverty, providing higher standard of living and to improve the socio-economic life of people as a whole, many development schemes have been launched by Central and State Government with the object of making self-employment programmes more effective. In order to determine the effectiveness of these schemes/programmes on the livelihood it is important to examine the impact and performance of these programmes, so that various effective development programmes further may be designed for the betterment of the rural communities.

MATERIAL AND METHODS

The present study was conducted in the state of Himachal Pradesh located in the Northern region of India, surrounded by Jammu & Kashmir, Ladakh, Punjab, Haryana, Uttarakhand and China on the six sides. Geographically, it extends 30°22"40” to 33°12"20” N latitudes and 75°45'55" to 79°04"20” E longitudes. Selection of the study area was based upon the contribution of districts to the Gross State Domestic Product (GSDP) or state income. These districts were then classified into three Strata viz. Strata-1 i.e. highly developed districts (greater than 10 percent contribution), Strata-2 moderate developed districts (5 to 10 per cent contribution) and Strata-3 least developed districts (less than 5 per cent contribution) as per District Domestic Product of Himachal Pradesh (Economics and Statistics 2016). Stratified multistage random sampling was used for the present study. State was firstly divided into three Strata i.e. Strata-1, Strata-2 and Strata-3. At the first stage one district was selected randomly from each of the above classified Strata. Thus comprises of district Solan from Strata-1, Chamba from Strata-2 and Kinnaur district from Strata-3 (Fig. 1). At the second stage two blocks were selected randomly from each selected districts. At the third stage three panchayats were selected randomly from each selected blocks. At the fourth stage two villages were selected randomly from each selected panchayats. At the last stage ten households were selected from each selected villages for the collection of primary data. Thus, 120 respondents from each district were selected which constitute a sample of 360 respondents for the present study. Further, to evaluate the impact of the different development schemes on the livelihood, fifteen schemes have been analyzed through Response Priority Index and difference between priorities was determined.

Responses-Priority index (RPI) : In the quantification of impact of government policies on livelihood as expressed by

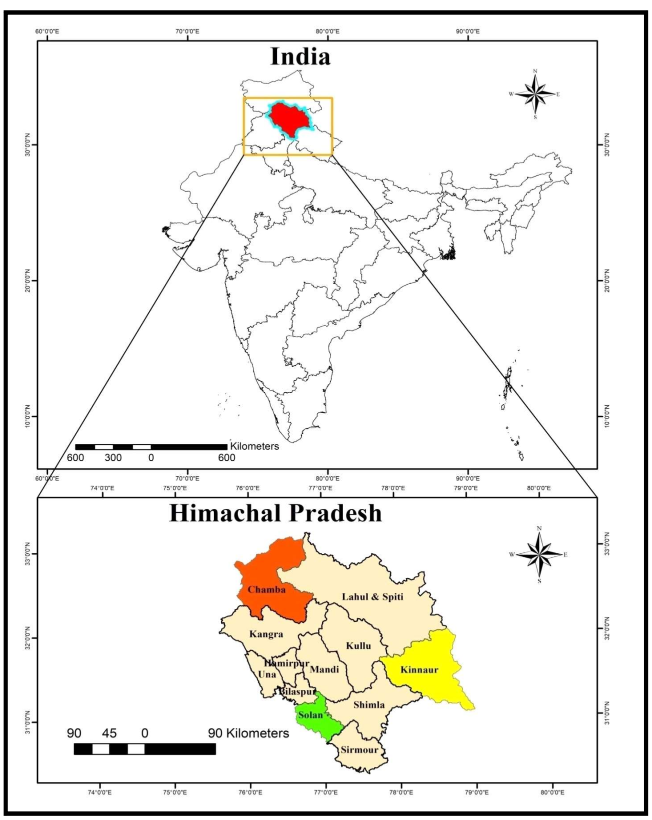

Fig. 1. Location of study area

the respondents, there was a problem, whether emphasis should be given for the number of responses to a particular priority or to the highest number of responses to a policy/programme in the first priority as both lead to different conclusions. Thus to resolve this, a Responses-Priority Index (RPI) was constructed as a product of Proportion of Responses (PR) and Priority Estimate (PE), where PR for the i^{th} policy/programme gave the ratio of number of responses for a particular policy/ programme to the total responses as (Ramarao IVY 2011) and is estimated as:

$$RPI = \frac{\sum_{j=1}^{k} f_{ij} X_{[(k+1)-j]}}{\sum_{i=1}^{1} \sum_{j=1}^{k} f_{ij}} \qquad 0 \leq RPI \leq 5$$

Where,

RPI_i = Response Priority Index for i^{th} policy/ programme

f_{ij}=Number of responses for the j^{th} priority of the i^{th} policy/programme

$\sum_{j=1}^{k} f_{ij}$ =Total number or responses for the i^{th} policy/programme

k= Number of priorities i.e. 5

$X_{[(k+1)-j]}$= Scores for the j^{th} priority (5, 4, 3, 2 and 1)

$\sum_{i=1}^{1} \sum_{j=1}^{k} f_{ij}$=Total number of responses to all the policies/programmes

$\sum_{i=1}^{n} RPI_i$ =Summation of RPI for all policies/programmes

Thus, larger the Responses Priority Index Higher was the impact of Government policies/programmes on livelihood.

RESULTS AND DISCUSSION

Impact of institutional programmes in the Solan district: The Kisan Credit Card (KCC) scheme has considerable effect on the livelihood of people with an RPI index score of 0.216 (Table 1). Vashisht and Vashisht (2019) observed 100 percent of the respondents in Solan have reported increase in opportunities for livelihood after introduction of this scheme. Furthermore, Himachal Pradesh Horticulture Development Project **(**HPHDP) programme contributed substantially in creating better livelihood options by providing quality planting material to farmers, with index of 0.210 and was ranked second. The Horticulture training and extension service scheme provided by the government also contributed effectively as obtained an index of 0.205 and ranked third among all the schemes studied in Solan district. However, the objective of strengthening the livelihood resource base of rural poor by land development under MGNREGA scheme was ranked fourth with an index value of 0.195. The effect of Sub- Mission on Agricultural Mechanization in providing subsidy on farm implements was also substantial with an index score of 0.185. However, no significant difference between the priorities was found as indicated by p (0.12) and f_{cal} (1.89) values.

Impact of institutional programmes in the Chamba district: The Mahatma Gandhi National Rural Employment Guarantee Act (MGNREGA) has large effect on the livelihood of people by helping in employment generation, with an RPI score of 0.216 (Table 2). In this scheme the resource base of rural poor by land development was strengthened as it obtained the second rank under Response Priority Index with value of 0.214. Furthermore, Sub- Mission on Agricultural Mechanization scheme contributed substantially in providing subsidy on farm implements. The Horticulture training and extension service scheme provided by the government also contributed effectively with index value of 0.205 and ranked fourth among all the schemes studied in Chamba district. Moreover, the KCC scheme was ranked fifth with an index value of 0.195. However significant difference between the priorities was found as determined by p (0.44) and f_{cal} (0.95) values.

Impact of institutional programmes in the Kinnaur district: As evident from the results presented in Table 3, the HPHDP programme obtained maximum RPI value of 0.204 against its objective of providing quality planting material to the farmers and was ranked first among all the programmes studied in Kinnaur district. By providing respondents with the best planting material of one major cash crop in this district i.e. apple, the quality and productivity of the produce is improved which would ultimately provide best market prices to the farmers, thereby having considerable impact on the livelihood of people. Moreover, the Watershed Development Programmes was also found to have significant impact as it was ranked second and obtained an index score of 0.202. Similar results were also observed in a study conducted by Mehta et al (2022), where maximum respondents reported significant impact of this scheme on the livelihood of tribal households residing in the district .The Horticulture Training

Table 1. Prioritization of impacts of different institutional programmes on livelihood in Solan district

Name of schemes	Numbers in respective priorities					Total recorded responses	RPI	Rank
	5	4	3	2	1			
MNREGA								
Strengthening the livelihood resource base of rural poor by land development	47	30	15	16	12	120	0.195	IV
Improvement in irrigation facilities	28	20	12	21	39	120	0.148	XV
Helps in employment generation	37	26	22	16	19	120	0.178	VI
IRDP								
Helps in employment generation	32	22	22	18	26	120	0.165	VIII
Financial assistance to the families in form of govt. subsidies.	19	20	30	12	39	120	0.144	XI
PMFBY (Pradhan Mantri Fasal Bima Yojna)	16	9	31	34	30	120	0.135	XVI
NRLM(National Rural Livelihood Mission)	6	5	10	40	59	120	0.096	XIX
PMAY (Pradhan Mantri Awaas Yojna)	22	21	25	22	30	120	0.150	XIII
Watershed Development Programme (IWDP/IWMP/PMKSY)	9	17	29	33	32	120	0.131	XVII
KCC (Kisan Credit Card)	58	26	28	7	1	120	0.216	I
PMEGP (Pradhan Mantri Employement Generation Programme)	11	5	33	24	47	120	0.118	XVIII
Mukhyamantri Swavalmban Yojna	21	19	40	24	16	120	0.160	X
Himachal Pradesh Horticulture Development Project (HPHDP)								
Helps in providing quality planting material to famers	59	23	18	17	3	120	0.210	II
Helps in providing training	14	26	36	24	20	120	0.154	XII
Sub- Mission on Agricultural Mechanization								
Provide subsidy on farm implements	39	27	21	22	11	120	0.185	V
Himachal Pradesh Khumb Vikas Yojna	11	19	50	23	17	120	0.151	XIV
Horticulture Training & Extension Service	57	16	29	13	5	120	0.205	III
Mahila Mandal Protsahan Yojna	35	12	26	25	22	120	0.164	IX
HIMCARE	34	29	22	16	19	120	0.177	VII

Table 2. Prioritization of impacts of different institutional programmes on livelihood in Chamba district

Name of schemes	Numbers in respective priorities					Total recorded responses	RPI	Rank
	5	4	3	2	1			
MNREGA								
Strengthening the livelihood resource base of rural poor by land development	55	36	15	11	3	120	0.214	II
Improvement in irrigation facilities	45	20	26	18	11	120	0.189	VII
Helps in employment generation	72	22	9	11	6	120	0.221	I
IRDP								
Helps in employment generation	29	24	11	29	27	120	0.157	XI
Financial assistance to the families in form of govt. subsidies.	15	5	51	25	24	120	0.141	XII
PMFBY (Pradhan Mantri Fasal Bima Yojna)	7	12	45	23	33	120	0.130	XIII
NRLM(National Rural Livelihood Mission)	5	4	33	31	47	120	0.109	XVII
PMAY (Pradhan Mantri Awaas Yojna)	36	29	26	19	10	120	0.185	IX
Watershed Development Programme (IWDP/IWMP/PMKSY)	16	19	23	48	14	120	0.147	XIV
KCC (Kisan Credit Card)	43	29	26	18	4	120	0.197	V
PMEGP (Pradhan Mantri Employement Generation Programme)	8	15	4	48	45	120	0.111	XVIII
Mukhyamantri Swavalmban Yojna	35	22	36	12	15	120	0.180	VIII
Himachal Pradesh Horticulture Development Project (HPHDP)								
Helps in providing quality planting material to famers	15	19	8	43	35	120	0.130	XVI
Helps in providing training	9	12	32	21	46	120	0.121	XV
Sub- Mission on Agricultural Mechanization								
Provide subsidy on farm implements	56	25	19	7	13	120	0.204	III
Himachal Pradesh Khumb Vikas Yojna	0	0	0	27	93	120	0.064	XIX
Horticulture Training & Extension Service	35	42	21	13	9	120	0.193	IV
Mahila Mandal Protsahan Yojna	54	9	29	18	10	120	0.193	VI
HIMCARE	24	19	30	26	21	120	0.157	X

Table 3. Prioritization of impacts of different institutional programmes on livelihood in Kinnaur district

Name of schemes	Numbers in respective priorities					Total recorded responses	RPI	Rank
	5	4	3	2	1			
MNREGA								
Strengthening the livelihood resource base of rural poor by land development	21	15	31	32	21	120	0.150	IX
Improvement in irrigation facilities	19	9	34	35	23	120	0.143	XIV
Helps in employment generation	21	19	22	31	27	120	0.147	X
IRDP								
Helps in employment generation	10	21	31	22	36	120	0.135	XII
Financial assistance to the families in form of govt. subsidies.	7	12	45	35	21	120	0.136	XVI
PMFBY (Pradhan Mantri Fasal Bima Yojna)	19	22	17	26	36	120	0.141	XI
NRLM (National Rural Livelihood Mission)	6	5	29	31	49	120	0.109	XVIII
PMAY (Pradhan Mantri Awaas Yojna)	22	8	27	25	38	120	0.136	XIII
Watershed Development Programme (IWDP/IWMP/PMKSY)	53	21	25	16	5	120	0.202	II

Cont...

Table 3. Prioritization of impacts of different institutional programmes on livelihood in Kinnaur district

Name of schemes	Numbers in respective priorities					Total recorded responses	RPI	Rank
	5	4	3	2	1			
KCC (Kisan Credit Card)	46	19	31	8	16	120	0.189	IV
PMEGP (Pradhan Mantri Employement Generation Programme)	8	6	31	26	49	120	0.113	XVII
Mukhyamantri Swavalmban Yojna	14	9	41	43	13	120	0.144	XV
Himachal Pradesh Horticulture Development Project (HPHDP)								
Helps in providing quality planting material to famers	55	18	28	15	4	120	0.204	I
Helps in providing training	36	28	19	25	12	120	0.180	VI
Sub- Mission on Agricultural Mechanization								
Provide subsidy on farm implements	31	36	28	10	15	120	0.183	V
Himachal Pradesh Khumb Vikas Yojna	0	0	17	44	59	120	0.087	XIX
Horticulture Training & Extension Service	58	14	24	9	15	120	0.198	III
Mahila Mandal Protsahan Yojna	36	14	9	25	36	120	0.153	VIII
HIMCARE	37	12	35	14	22	120	0.170	VII

Table 4. Prioritization of impact of different institutional programmes on livelihood in the study area

Name of schemes	Numbers in respective priorities					Total recorded responses	RPI	Rank
	5	4	3	2	1			
MNREGA								
Strengthening the livelihood resource base of rural poor by land development	123	81	61	59	36	360	0.187	IV
Improvement in irrigation facilities	92	49	72	74	73	360	0.160	IX
Helps in employment generation	130	67	53	58	52	360	0.182	V
IRDP								
Helps in employment generation	71	67	64	69	89	360	0.152	XII
Financial assistance to the families in form of govt. subsidies.	41	37	126	72	84	360	0.140	XV
PMFBY (Pradhan Mantri Fasal Bima Yojna)	42	43	93	83	99	360	0.135	XVI
NRLM (National Rural Livelihood Mission)	17	14	72	102	155	360	0.105	XVIII
PMAY (Pradhan Mantri Awaas Yojna)	80	58	78	66	78	360	0.157	XI
Watershed Development Programme (IWDP/IWMP/PMKSY)	78	57	77	97	51	360	0.160	XIII
KCC (Kisan Credit Card)	147	74	85	33	21	360	0.201	I
PMEGP (Pradhan Mantri Employement Generation Programme)	27	26	68	98	141	360	0.114	XVII
Mukhyamantri Swavalmban Yojna	70	50	117	79	44	360	0.161	X
Himachal Pradesh Horticulture Development Project (HPHDP)								
Helps in providing quality planting material to famers	129	60	54	75	42	360	0.181	VI
Helps in providing training	59	66	87	70	78	360	0.152	XIV
Sub- Mission on Agricultural Mechanization								
Provide subsidy on farm implements	126	88	68	39	39	360	0.190	III
Himachal Pradesh Khumb Vikas Yojna	11	19	67	94	169	360	0.101	XIX
Horticulture Training & Extension Service	150	72	74	35	29	360	0.199	II
Mahila Mandal Protsahan Yojna	125	35	64	68	68	360	0.170	VIII
HIMCARE	95	60	87	56	62	360	0.168	VII

and Extension Service ranked third with an index value of 0.198 also contributed effectively. However, the KCC scheme was ranked fourth with an index value of 0.195. Furthermore, Sub-Mission on Agricultural Mechanization scheme contributed substantially in providing subsidy on farm implements. However, significant difference between the priorities was found as represented by p (0.02) and f_{cal} (2.95) values.

At overall level the Kisan Credit Scheme (KCC) contributed substantially in augmenting the livelihood status of the people by providing them financial security at reasonable interest rates (Table 4). Furthermore, Horticulture Training and Extension Service, ranked second with an index value of 0.199 also contributed effectively. Also, the Sub-Mission on Agricultural Mechanization scheme contributed substantially in providing subsidy on farm implements and recorded an index score of 0.190. The Mahatma Gandhi National Rural Employment Guarantee Act (MGNREGA) has large effect on the livelihood of people by helping them in employment generation, with an RPI score of 0.187. Also, under this scheme the resource base of rural poor by land development was strengthened as it obtained the fifth rank under Response Priority Index with value of 0.182. However, significant difference between the priorities was found as indicated by p (0.04) and f_{cal}(2.57) values.

CONCLUSIONS

The majority of respondents in Solan district reported increase in the options for livelihood after the introduction of KCC scheme, which provided them with the financial stability whereas in Chamba district profound impact of Mahatma Gandhi National Rural Employment Guarantee Act (MGNREGA) was on the livelihood of people as it helped in generating employment opportunities for the rural households. The HPHDP programme obtained maximum RPI value among all the programmes studied in Kinnaur District. However, at overall level the Kisan Credit Card scheme (KCC) contributed substantially in augmenting the livelihood status of the people by providing them financial security at reasonable interest rates. The focus group discussions showed that there were many problems in implementation of these schemes, especially identification of wrong beneficiaries, delay in disbursal of subsidy, underutilization of subsidized inputs, misallocation of resources. Therefore, in order to address these issues and ensure that the maximum benefits reach the ultimate beneficiaries, there is a need to popularize about various government schemes among the households through frequent visits by local officers and sensitization in Gram Sabha.

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Biology of Tea Mosquito Bug, *Helopeltis antonii* Signoret in *Moringa olifera*

K. Aravinthraju, K. Suresh[1] and A.V. Moorthy[2]

Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore-641 003, India
[1]Krishi Vigyan Kendra, Tamil Nadu Agricultural University, Madurai-625 104, India
[2]Department of Entomology, G.B Pant University of Agriculture and Technology, Pantnagar-263 145, India
E-mail: aravinth4697@gmail.com

Abstract: Biology of tea mosquito bug, *Helopeltis antonii* Signoret was studied under field and laboratory conditions. The primary survey on incidence of tea mosquito bug in moringa at different locations showed higher incidence of tea mosquito bug s during November, 2019 and till January, 2020 at all locations. The primitive symptom was formation of necrotic lesions on young flush of moringa due to feeding of tea mosquito bug, and on continuous feeding by nymphs and adults of tea mosquito bug wilting of young flush along with gummy exudes occurs. The duration of incubation period, nymphal instars and male and female adult longevity was 5.55, 16.75, 5.16 to 6.65 days of adult longevity (males and females respectively). The total life cycle was 27.45 and 29.20 days for males and females, respectively.

Keywords: Tea mosquito bug, *Helopeltis antonii*, *Moringa olifera*, Symptoms, Biology

Moringa, *Moringa olifera* (Moringaceae) is one of the most important vegetable crop in many of the countries including India, Ethiopia, Phillliphines etc. Moringa plants suffer from major pests pod fly, Gitona distigma; budworm, Noorda moringae; hairy caterpillar, Eupterote mollifera etc. (Mridha and Baraka 2017). The tea mosquito bug, *Helopeltis antonii* Signoret (Miridae: Hemiptera) is one of the new pest in Moringa (Aravinthraju et al 2022). Tea mosquito bug was first reported in India at Cachar District of Assam, attacking *Camellia sinensis* (L.) Kuntze (Theaceae) (Aravinthraju et al 2021). After which is co-evolving in the agricultural ecosystem by feeding many tropical hosts like *Psidium guajava* (Lithomyrtus), *Azadirachta indica* (Meliaceae), *Anacardium occidentale* (Anacardiaceae) etc. The present studies on nature of damage and biology of tea mosquito bug in moringa were undertaken.

MATERIAL AND METHODS

Survey: A survey was conducted in moringa fields of Andipatti (9.99508° N latitude, 77.66715° E longitude) and Natham (10.20284° N latitude, 78.15778° E longitude) blocks of Theni and Dindigul districts respectively, Tamil Nadu, India, during 2019-2020. The collection of adults and nymphs were done for mass culturing purpose. The specimens have been maintained in the Department of Entomology, Agriculture College and Research Institute, Madurai.

Mass culturing of tea mosquito bug: Mass rearing methods of tea mosquito bug described by Sundararaju and John (1992) was followed. Adult and nymphs collected from the moringa fields and released in one month old moringa seedlings kept in the aluminum rearing cage of size 74 X 74 X 74 cm. Three sides and top of the cage were closed and one side was provided with movable lid. The seedlings were replaced once in three days based on feeding punctures and drying of leaves and seedling in adult cage was observed for presence of egg. This mass rearing setup was maintained in normal atmospheric condition under shade at Insectary, Department of Agricultural Entomology, Agricultural College and Research Institute, Madurai.

Biology of tea mosquito bug: Biology of TMB was studied based on the procedure reported by Mohapatra and Satapathy (1999). The adults were released on one month old moringa seedlings and observed regularly for presence of eggs and egg hatching. Moringa seedlings with eggs were isolated in separate cage of size 45 X 45 X 45 cm. Observations on egg hatching, nymph duration, adult longevity, adult sex and female oviposition were made daily. Five replications with five nymphs per replication were maintained for recording observations. The study was carried out in normal atmospheric condition at Insectary, Department of Agricultural Entomology, Agriculture College and Research Institute, Madurai during 2019-2020.

RESULTS AND DISCUSSION

Incidence of tea mosquito bug in moringa: The incidence of tea mosquito bug starts from November, 2019 and continued till January, 2020 at all locations in moringa (Table

Table 1. Survey on the incidence of tea mosquito bug in moringa at different locations

Block	Village	Population (No./ 15 flush)		
		November 2019	December 2019	January 2020
Andipatti	Maravapatti	5.67	9.83	13.17
	Thimmarasanaikanur	6.33	12.83	12.00
Natham	Mulaiyur	4.00	8.00	10.00
	Panniyamalai	5.67	9.67	9.67

Table 2. Biology of tea mosquito bug, *H. antonii* in moringa

Life stages		Duration (Days)*
Egg period		5.55 $\pm$ 1.17
Nymphal period	I instar	3.29 $\pm$ 0.41
	II instar	3.53 $\pm$ 0.87
	III instar	3.14 $\pm$ 0.36
	IV instar	3.30 $\pm$ 0.50
	V instar	3.49 $\pm$ 0.23
	Total	16.75 $\pm$ 1.13
Adult longevity	Male	5.16 $\pm$ 0.74
	Female	6.65 $\pm$ 0.68
Total life cycle	Male	27.45 $\pm$ 2.09

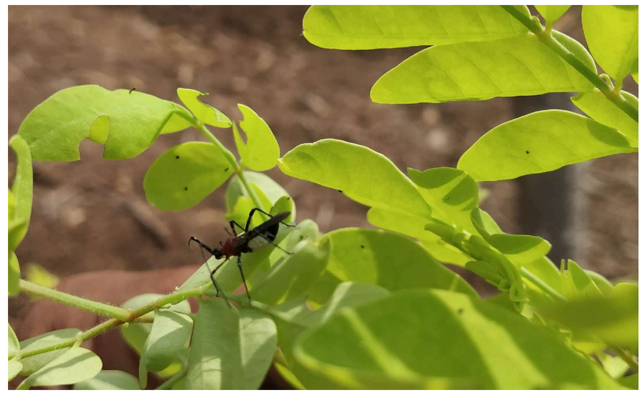

Plate 1. Tea mosquito bug in moringa

Plate 2. Severe infestation of moringa tea mosquito bug

1). The incidence ranged was 4.00 and 13.17 bugs/ 15 flush in moringa. The peak incidence of tea mosquito bug in moringa was during January. Kumar et al (2002) and Thirumalaraju (2003) also observed that the tea mosquito bug was active from June to February. But peak incidence was during September to November. Patil and Naik (2010) observed the mosquito bug incidence in guava was at its peak during September (24.3%).

Symptoms of damage: The nymphs and adults suck sap from young shoots of moringa, forming necrotic lesions on shoots. Due to continuous feeding by the bugs the lesions coalesce, result in drying of the whole twigs. The young instars mainly congregate on growing shoot tips, which dry first. Later due to continuous feeding of bugs the whole twig become dry and the leaves will fall off. There were gummy exude on the feeding sites of tea mosquito bug, which will resemble bubbles on the stem. Under severe infestation of bugs, entire leaves will fall off and the plant resembles the snag. In some occasions the bugs will even feed on pods (Plate 1, 2).

Biology of tea mosquito bug: Life cycle of tea mosquito bug consists of an egg stage, five nymphal stages and an adult stage. The egg duration of tea mosquito bug in moringa was as 5.55 days. Eggs are oval with presence of two extra chorionic processes above it. As eggs are inserted inside plant tissues only these two extra chorionic processes are visible with an eggs exit side over the surface of the plant part. Durations of I, II, III, IV and V instar nymphs were 3.29, 3.53, 3.14, 3.30 and 3.49 days, respectively. The total nymphal duration was 16.75. Early nymphal instars show less movement and hence the congregate on the young twigs, but the later instars spreads actively. The longevity of adult male and female was 5.16 and 6.65 days respectively. Adults are poor flyers, mostly sessile while feeding and oviposition, but escapes actively during disruption. The total life cycle of tea mosquito in moringa was 27.45 and 29.20 days for males and females, respectively (Table 2).

CONCLUSION

Tea mosquito bug in moringa causes severe damage by sucking sap from young flushes, forming necrotic lesions on young shoots. These lesions on continuous feeding coalesce and result in wilting of twigs. The duration of egg and nymphal stage was 5.55 and 16.75 days. The longevity of male and female was 5.16 and 6.65 days. The total life cycle was 27.45 and 29.20 days for males and females, respectively.

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Effect of Sulphur Dioxide Inhalation on Oxidative and Histopathological Damage in Kidney and Spleen of *Rattus rattus*

P. Sood and N. Singla

Department of Zoology, College of Basic Sciences and Humanities, Punjab Agricultural University, Ludhiana-141 004, India
E-mail: prernasood@pau.edu

Abstract: Effect of different concentrations of SO_2 (5, 10 and 15ppm) on antioxidant enzymes and histopathological changes were investigated in kidney and spleen of rats of both the sexes under natural and experimental conditions. Naturally exposed, house rat, *Rattusrattus*were collected from urban residential areas at Ludhiana, India. Laboratory rats were exposed to 5, 10 and 15 ppm of SO_2 for 5 hours /day for 28 days in 1 m^3exposure chamber. All the three concentrations of SO_2 showed significant increase in lipid peroxidation levels followed by significant decrease in the activities of superoxide dismutase, catalase and glutathione reductase and non-significant decrease in glutathione peroxidase and glutathione levels in kidney and spleen of rats of both the sexes. The results showed that SO_2 is an oxidative damage causing agent to kidney and spleen of rats of both the sexes.

Keywords: Sulphur dioxide (SO_2), Rats, Lungs, Brain, Histopathology, Oxidative damage, Air pollutant

In most of the developing countries, air pollution is one of the major problems that arise due to industrial activities and fossil fuel consumption (Ghorani et al 2016). Sulphur dioxide (SO_2) is the air pollutant formed by the coal fired power plants, industrial processes and motor vehicle operations. It is present in high concentrations in urban and industrial locations and produces damaging effect on human health (Shen et al 2020). High concentrations of SO_2 can affect functioning of vital organs of animals and humans. In developed countries, SO_2 at 2 ppm concentration is the industrial maximal limit for 8 hour per day. According to facts, air pollution has contributed to premature death, diseases of cardiovascular system (Al-Kindi et al 2020), respiratory system, neurology system, cancer, diabetes mellitus (DM) and fertility disorders (Li et al 2019, Liu et al 2022). Above 3 ppm concentration, SO_2 gas has a strong, nauseating odour. In developed countries, SO_2 at 2 ppm concentration is the industrial maximal limit for 8 hour per day. In this study three doses of SO_2 i.e. 5, 10 and 15 ppm were taken and these were above the maximum industrial permissible limits of SO_2 present in the atmosphere. 5 ppm and 10 ppm concentration of SO_2 represents 10 and 20 fold greater than the typical urban concentration (0.5ppm) and is known to induce harmful effects to respiratory system of healthy individuals. Third concentration is 15 ppm which is beyond the natural exposure and is used to examine the effects of this higher concentration of SO_2 on the health of individuals.On the basis of this exposure limit three different concentrations of SO_2 were selected for the present study. In this study the kidney and spleen of rats were examined for oxidative damage and stress and histopathological damage instigated by SO_2 inhalation.

MATERIAL AND METHODS

The male and female rats of 100-150 g were taken. Naturally exposed (Group I), house rat, *Rattusrattus*were collected from urban residential areas at Ludhiana, India and acclimatized for 1 month in the laboratory. Laboratory rats were divided into four groups. Each group was further subdivided into two subgroups having 6 male and 6 female rats. Group II (control rats) was exposed to filtered air in exposure chamber for 28 days, Group III, IV and V were treated with 5, 10 and 15 ppm of SO_2 for 5 hours /day for 28 days in 1 m^3 exposure chamber. SO_2 gas was provided to treated rats through a tube located at the top of each chamber and was distributed with the help of a fan in each chamber. Rats were kept in cages under standard conditions of humidity and temperature with light-dark cycle. Food (loose mixture of cracked wheat grains, powdered sugar and edible vegetable oil in ratio 96:2:2) and water were provided to control and treated rats *ad libitum*. The weight of individual rat was recorded weekly. After 28 days of exposure rats were dissected. The experiment was performed after the approval of Institutional Animal Ethical Committee, Guru Angad Dev Veterinary and Animal Sciences University Ludhiana, India under protocol No. (GADVASU/2022/IAEC/64/17).

Assay of antioxidant enzymes: After dissection kidney and spleen of male and female rats were removed and weighed. The tissues were sheared in 0.9% saline (chilled) and homogenization was done in 0.1 M phosphate buffer pH 7.4. The homogenates were centrifuged for 30 min at 1000 rpm at 4^0C to obtain supernatants. The tissue supernatants were used for the assay of different antioxidant enzymes like superoxide dismutase (SOD), glutathione S-transferase (GST), catalase (CAT), glutathione (GSH), glutathione peroxidase (GPx), glutathione reductase (GR) and lipid peroxidation (LPO).

SOD activity: The kidney and spleensupernatant was used for estimating the activity of SOD by standard method of Marklund and Marklund (1974).

Glutathione-S-transferase (GST) activity: GST activity in tissue homogenate was estimated by the method of Habiget al (1974).

GSH-Px activity: The GSH-Px activityin tissue homogenate was determined by the method of Hafeman et al(1984).

Glutathione reductase (GR) activity: GR enzyme activity was estimated by the method given by Carlberg and Mannervik (1985) in tissue homogenate.

CAT activity: Catalase enzyme activity was estimated by the standard method of Aebi (1983).

LPO activity: Lipid peroxidation (LPO) activity was estimated by the method of Stocks and Dormandy (1971).

GSH activity: GSH activity was estimated by standard method of Jollow et al (1974).

Protein assay: In the estimations of all the antioxidant enzymes the total soluble protein content was estimated by Lowry et al (1991) taking BSA as standard.

Histological studies: Kidney and spleen of rats were cleared and fixed in 10% formaline for 24 hours. Then the tissues were dehydrated in different grades of ethanol, clearing was done in xylene and embedding was done in paraffin wax for the preparation of blocks. The 5-7µm thick sections were cut and stained in haematoxylin-eosin stain and mounted in DPX.

Statistical analysis: Statistical analysis software (SPSS) was used to analyse the data.

RESULTS AND DISCUSSION

There were two important considerations for the planning of this experiment. Firstly, rats were exposed to SO_2 for a regular period (5 h/day for 7 days with 19 hours between exposures) with relief periods in between the exposure. Secondly, rats are nose breathers and most of inhaled gas is trapped in nasal chambers and only some amount is reaching to lungs that is why higher concentration of SO_2 (i.e.15 ppm) was used. Both the considerations may provide a repercussion to persons exposed to the gas in an occupational or industrial setting. SO_2 exposure caused decrease in SOD and CAT activities at all the three concentrations but the decrease was significant at 15 ppm as compared to control rats in both the sexes (Table 3 and 4). GSH level showed non-significant decrease at three

Table 1. Effect of SO_2 on net body weight of male and female rats

Group	Treatment	Male rats		Female rats	
		Initial weight (g)	Final weight (g)	Initial weight (g)	Final weight (g)
I	Naturally exposed rats	103.66± 1.85	-	107.66 ± 3.84	-
II	Control	118.33 ± 7.26	110.33 ± 6.38	115.00± 2.88	106.66 ±1.66
III	5 ppm SO_2	111.66± 9.27	106.66± 9.27	121.66 ± 1.66	116.00 ± 3.05
IV	10 ppm SO_2	105.66± 3.48	100.00 ± 3.78	114.00 ± 3.05	109.33 ± 1.76
V	15 ppm SO_2	105.33± 1.76	102.33 ± 1.45	125.66 ± 6.98	120.00 ± 5.77

Values are shown as mean±SE

Table 2. Effect of SO_2 on weights (g/100g b.w.) of kidney and spleen in male and female rats

Group	Treatment	Male rats		Female rats	
		Initial weight (g)	Final weight (g)	Initial weight (g)	Final weight (g)
I	Naturally exposed rats	0.40 ± 0.01^{b}	$0.27 \pm .01^{a}$	0.42 ± 0.005^{ab}	0.30 ± 0.006^{ab}
II	Control	0.38 ± 0.005^{ab}	0.29 ± 0.01^{a}	0.43 ± 0.01^{b}	0.29 ± 0.01^{ab}
III	5 ppm SO_2	0.37 ± 0.01^{ab}	0.24 ± 0.02^{a}	0.43 ± 0.02^{a}	0.36 ± 0.02
IV	10 ppm SO_2	0.36 ± 0.01^{a}	0.24 ± 0.02^{a}	0.36 ± 0.01	0.24 ± 0.02^{a}
V	15 ppm SO_2	0.36 ± 0.01^{a}	0.24 ± 0.02^{a}	0.37 ± 0.01^{ab}	0.24 ± 0.02^{a}

Values are shown as mean±SE
^{ab} represents significant difference between treatments at p≤0.05 as compared to control

concentrations of SO_2 and significant decrease at 15 ppm. GST, GPx and GR activities showed non-significant decrease at all the three concentrations when compared with control rats. LPO level was increased at all the three concentrations in the kidney and spleen of rats of both the sexes (Table 5, 6) compared with control rats. But the increase in LPO level was statistically significant at 15 ppm and it was more significant in spleen of male rats as compared to female rats.

Superoxide dismutase (SOD) helps in protecting the cells from molecular oxygen and also removes superoxide radicals and decreased SOD level in brain and lungs may lead to free radical damage at large scale because SOD is the first security against toxicity of molecular oxygen.The decrease in the level of GST, GSH and GPx at higher concentration of SO_2 was found to be dose dependent. A significant increase in Lipid peroxidation (LPO) activity was observed at 15 ppm concentration in brain and lungs from rats of both the sexes as compared to control rats. Higher SO_2 concentration decreased the activities of SOD, CAT, GST, GPx, GSH and increased lipid peroxidation indicating the increased oxidative damage and stress in cells and tissues in

Table 3. Effect of SO_2 on antioxidant parameters of kidney of male rats

Group	Treatment	Antioxidant parameters						
		Superoxide dismutase (SOD)	Catalase (CAT)	Glutathione-S-transferase (GST)	Glutathione Peroxidase (GPx)	Glutathione (GSH)	Glutathione reductase (GR)	Lipid peroxidation (LPO)
I	Naturally exposed rats	12.66 ± 0.01^d	11.90 ± 0.14^c	0.40 ± 0.004^b	0.82 ± 0.03^b	46.60 ± 0.39^d	0.06 ± 0.003^c	0.47 ± 0.01^a
II	Control	13.72 ± 0.01^d	12.63 ± 0.06^d	0.41 ± 0.009^b	0.85 ± 0.009^b	48.75 ± 0.13^d	0.06 ± 0.001^c	0.46 ± 0.01^a
III	5ppm SO_2	9.45 ± 0.39^b	11.38 ± 0.19^c	0.34 ± 0.01^a	0.79 ± 0.007^b	41.49 ± 0.14^b	0.04 ± 0.002^b	0.76 ± 0.01^c
IV	10ppm SO_2	10.96 ± 0.02^c	10.16 ± 0.14^b	0.33 ± 0.01^a	0.81 ± 0.007^b	39.27 ± 0.22^a	0.05 ± 0.0007^b	0.52 ± 0.008^a
V	15ppm SO_2	9.66 ± 0.05^b	8.51 ± 0.19^a	0.33 ± 0.015^a	0.60 ± 0.04^a	40.79 ± 0.12^b	0.05 ± 0.001^b	0.61 ± 0.02^b

Values expressed as mean±SE
[abcd] represents significant difference between treatments at p≤0.05 as compared to control
Units: SOD (U/mg protein), CAT (μmole of H_2O_2decomposed/min/mg protein), GPx(U/mg protein), GST (μmoles of GSH-CDNB conjugate formed/min/mg protein) GR (μmoles of NADPH oxidized/min/mg protein), Lipid peroxidation (nmol MDA/100 mg tissue)

Table 4. Effect of SO_2 on antioxidant parameters of kidney of female rats

Group	Treatment	Antioxidant parameters						
		Superoxide dismutase (SOD)	Catalase (CAT)	Glutathione-S-transferase (GST)	Glutathione Peroxidase (GPx)	Glutathione (GSH)	Glutathione reductase (GR)	Lipid peroxidation (LPO)
I	Naturally exposed rats	10.83 ± 0.07^d	11.57 ± 0.04^a	0.51 ± 0.04^a	0.71 ± 0.01^d	46.01 ± 0.12^c	0.06 ± 0.008^c	0.46 ± 0.01^a
II	Control	11.73 ± 0.07^d	12.86 ± 0.03^a	0.54 ± 0.06^a	0.74 ± 0.016^d	48.47 ± 0.12^c	0.066 ± 0.0008^c	0.47 ± 0.011^a
III	5ppm SO_2	6.08 ± 0.15^a	10.95 ± 0.31^b	0.58 ± 0.04^a	0.66 ± 0.02^b	41.33 ± 0.40^b	0.061 ± 0.003^{bc}	0.93 ± 0.01^c
IV	10ppm SO_2	10.67 ± 0.04^c	11.40 ± 0.28^b	0.44 ± 0.001^{ab}	0.68 ± 0.02^{bc}	41.76 ± 0.30^b	0.05 ± 0.001^b	0.52 ± 0.008^a
V	15ppm SO_2	7.39 ± 0.23^b	10.68 ± 0.07^b	0.44 ± 0.14^a	0.76 ± 0.016^d	41.45 ± 0.92^b	0.05 ± 0.001^b	0.95 ± 0.01^c

See Table 3 for details

Table 5. Effect of SO_2 on antioxidant parameters of spleen of male rats

Group	Treatment	Antioxidant parameters						
		Superoxide dismutase (SOD)	Catalase (CAT)	Glutathione-S-transferase (GST)	Glutathione Peroxidase (GPx)	Glutathione (GSH)	Glutathione reductase (GR)	Lipid peroxidation (LPO)
I	Naturally exposed rats	12.83 ± 0.27^c	9.57 ± 0.20^d	0.51 ± 0.03^b	0.85 ± 0.005^c	46.01 ± 0.17^c	0.05 ± 0.001^c	0.45 ± 0.01^a
II	Control	13.03 ± 0.19^c	9.99 ± 0.20^d	0.50 ± 0.001^b	0.87 ± 0.005^c	48.12 ± 0.18^c	0.06 ± 0.001^c	0.48 ± 0.019^a
III	5ppm SO_2	7.82 ± 1.30^a	8.48 ± 0.16^c	0.42 ± 0.001^b	0.82 ± 0.01^{bc}	42.33 ± 0.20^b	0.054 ± 0.001^b	0.73 ± 0.01^{bc}
IV	10ppm SO_2	9.71 ± 0.05^b	6.36 ± 0.12^a	0.43 ± 0.003^b	0.84 ± 0.007^{bc}	42.68 ± 0.38^b	0.052 ± 0.0006^b	0.52 ± 0.01^a
V	15ppm SO_2	7.82 ± 1.30^a	7.56 ± 0.12^b	0.45 ± 0.002^b	0.81 ± 0.007^b	42.00 ± 0.44^b	0.053 ± 0.0007^b	0.75 ± 0.01^c

See Table 3 for details

present case. Increased lipid peroxidation seems to be indicator of several disorders in cells, tissues or organs and it might be involved in different diseased state of cell such as aging, nervous disorders etc (Meng et al 2002a, Meng et al 2002b). Increased lipid peroxidation may have harmful effects on composition and function of biological membranes. SO_2 toxicity may involve oxidative stress to cells and tissues because of the production of free radicals during oxidation process (Meng 2003).

Histologically, kidney section of control and naturally exposed rats displayed normal bowman's capsule, proximal convoluted tubule and distal convoluted tubule while the 5 ppm SO_2 exposed group showed enlarged parietal layer of glomerulus and degenerated tubules and 10 ppm SO_2 exposed group showed blood congestion, degenerated tubules and degenerated glomerulus, 15 ppm SO_2 exposed group showed distorted bowman's capsule, degenerated tubules and collecting duct as compared to control rats (Fig. 1). Spleen tissue section of control rats and naturally exposed rats displayed normal morphologies, the boundary of the red pulp and white pulp is clear, the central artery is obvious, the trabeculae is clear, the white pulp lymphocytes and the macrophages are abundant. 5 ppm SO_2 exposed group showed white pulp hyperplasia and red pulp congestion and 10 ppm exposed group showed red pulp area having splenic cord hyperplasia and lymphatic nodules multiplied and 15 ppm SO_2 exposed group showed number of lymphocytes and macrophages decreased significantly compared with the control group (Fig. 2) (Gao et al 2018). Decreased antioxidant enzymes influence the organs (lungs and kidney) to increase free radical damage which in turn affects the defense system of body (Bakurt et al 2004). The main target organ for SO_2 intoxication are the lungs as it is

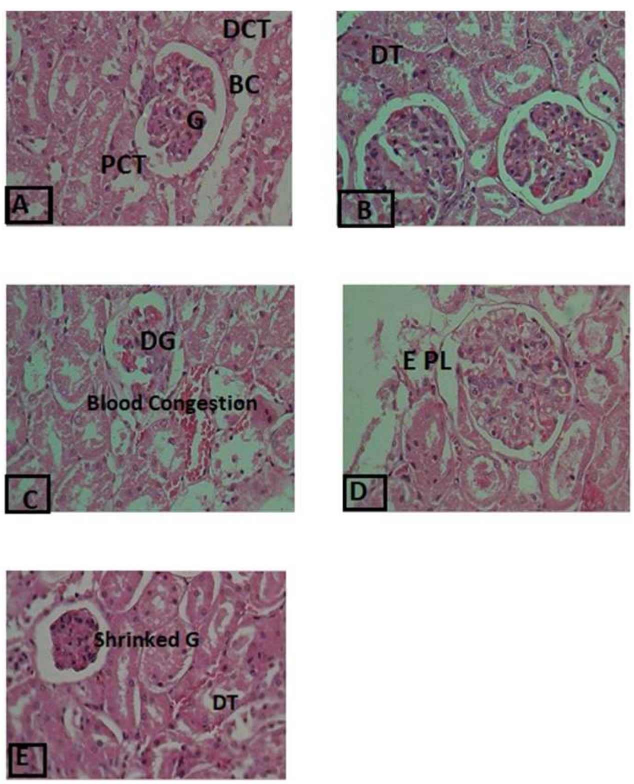

Fig. 1. Kidney section of control and naturally exposed rats (A&B) displayed normal Bowman's capsule (BC), Proximal Convoluted Tubule (PCT) and Distal Convoluted Tubule (DCT) (Fig 1(C) 5ppm SO_2 exposed group showed degenerated glomerulus (G), blood congestion and degenerated tubules(DT) Fig 1(D) exposed group showed Enlarged parietal layer(EPL) of glomerulus, blood congestion and degenerated tubules Fig 1(E) 15 ppm exposed group showed shrinked or distorted bowman's capsule, degenerated tubules and collecting duct as compared to control rats observed by light microscopy with X400 magnification

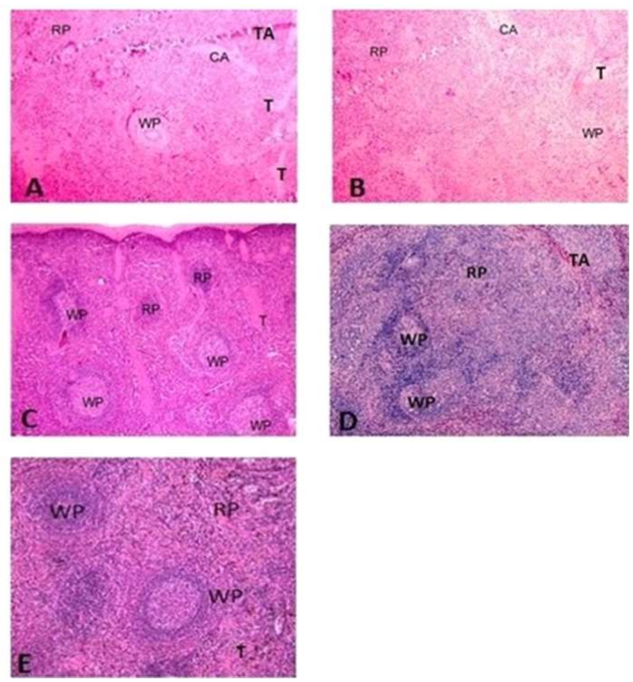

Fig. 2. Spleen tissue section of control group and naturally exposed rats (A&B) displayed normal morphologies, the boundary of the red pulp (RP) and white pulp(WP) is clear, the central artery(CA) is obvious, the trabeculae (T) is clear, the white pulp lymphocytes are abundant and the macrophages are abundant. 5 ppm SO_2 exposed group (C) showed white pulp hyperplasia and red pulp congestion and 10 ppm exposed group (D) showed red pulp area having splenic cord hyperplasia and lymphatic nodules multiplied and 15 ppm SO_2 (E) exposed group showed number of lymphocytes and macrophages decreased significantly compared with the control group observed by light microscopy with X400 magnification

Table 6. Effect of SO_2 on antioxidant parameters of spleen of female rats

Group	Treatment	Antioxidant parameters						
		Superoxide dismutase (SOD)	Catalase (CAT)	Glutathione-S-transferase (GST)	Glutathione Peroxidase (GPx)	Glutathione (GSH)	Glutathione reductase (GR)	Lipid peroxidation (LPO)
I	Naturally exposed rats	13.80 ± 0.26[c]	8.64 ± 0.30[c]	0.51 ± 0.003[a]	0.87 ± 0.01[a]	47.09 ± 0.19[c]	0.06 ± 0.001[c]	0.49 ± 0.02[a]
II	Control	14.73 ± 0.13[d]	9.97 ± 0.20[d]	0.50 ± 0.001[a]	0.88 ± 0.007[b]	48.73 ± 0.13[c]	0.06 ± 0.001[c]	0.48 ± 0.03[a]
III	5ppm SO_2	8.85 ± 0.67[b]	8.59 ± 0.12[c]	0.69 ± 0.08[c]	0.843 ± 0.01[a]	40.86 ± 0.14[b]	0.04 ± 0.002[b]	0.92 ± 0.004[c]
IV	10ppm SO_2	11.81 ± 0.01[c]	7.90 ± 0.23[bc]	0.48 ± 0.008[ab]	0.841 ± 0.006[a]	42.89 ± 0.26[c]	0.05 ± 0.001[bc]	0.56 ± 0.02[a]
V	15ppm SO_2	8.85 ± 0.67[b]	7.61 ± 0.12[b]	0.45 ± 0.01[ab]	0.83 ± 0.01[a]	41.56 ± 0.13[b]	0.05 ± 0.001[c]	0.94 ± 0.009[c]

See Table 3 for details

shown by oxidative damage and histopathological alterations in lungs., Besides lungs it also effects other organs like brain, kidneys, spleen and reproductive organs.

CONCLUSIONS

The SO_2 inhalation have toxic effects in brain and lungs of rats of both the sexes. Brain and lungs were affected in terms of SO_2 induced oxidative stress by elevation of LPO and diminution of antioxidant enzymes markers in rats of both the sexes in dose dependent manner.

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Farmers' Knowledge and Perceptions of Cotton Insect Pests and their Management Practices in Haryana

Pritam Kumari, Anil Jakhar, Deepak Kumar[1] and Sindhu Sheoran

Department of Entomology, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004, India
[1]Maharishi Markendaswar University, Mullana, Ambala-133 203, India
E-mail: primehra36@gmail.com

***Abstract*:** A survey was carried out in major cotton growing districts namely, Hisar, Sirsa and Bhiwani of Haryana state for acquiring information on farmers' knowledge, perception and practices of insect pest management in *Gossypium hirsutum* during *kharif*, 2019. Majority of respondents (70.0%) belonged to the age group of 40-60 years with an overall literacy rate of 87 per cent and 56 per cent of the respondents had more than 15 years of experience in cotton cultivation. RCH 773 BG II, RCH 776 BG II, US 51 BG II, Ankur 3028 BG II and US 81 BG II were the most commonly grown hybrids by the farmers. Whitefly, *Bemisia tabaci* and leafhopper, *Amrasca biguttula biguttula* were identified as the most important pests among sucking pests in *Bt* cotton. All the farmers targeted whitefly and leafhopper for control whereas none targeted the dusky cotton and red cotton bug for control. The incidence, severity and yield losses caused by whitefly and leafhopper were estimated to be high by 100.0, 86.6 & 82.7; 76.6, 74.0 & 64.6 per cent farmers, respectively. Cent per cent farmers adopted control measures at 61-90 days after sowing as highest pest incidence was estimated by cent per cent farmers in this duration. Majority of the farmers (44.6%) were dependent on agriculture input dealers for information on cotton cultivation followed by CCS HAU, Hisar/ICAR-CICR-RS, Sirsa (34.6%). Higher cost of insecticides, extreme weather conditions, lack of knowledge about bioagent and poor efficacy of insecticides were identified as major constraints faced by farmers for insect pest management in cotton. Insecticides like imidacloprid, thiamethoxam, emamectin benzoate, fipronil, dimethoate and monocrotophos were mainly used by farmers to manage sucking pests. Besides, newer insecticides like flonicamid, dinotefuran, diafenthiuron, spiromesifen and spinetoram were also used by farmers.

Keywords: *Bt* cotton, Farmers, Whitefly, Leafhopper, Information, Constraints

Cotton, *Gossypium* spp. popularly known as "White Gold" is a major fiber crop of the world and is used by about 75 per cent of world's population for textile purposes. It is native to tropical and subtropical regions around the world, including America, India and Africa. In India all four cotton species *viz*., old-world cotton, *G. arboreum* L., *G. herbaceum* L. and new world cotton, *G. barbadense* L. and *G. hirsutum* L. as well as some hybrids are cultivated commercially. The American cotton, *G. hirsutum* accounts for about 90% of the hybrid cotton genotypes grown in India (Hong-Bin et al 2008). Cotton is grown in an area of more than 38 million hectares (m ha) in the world, of which approximately 24 per cent is covered in India. The major producers of cotton are China, India, USA, Pakistan, Uzbekistan, Argentina, Australia, Greece, Brazil, Mexico and Turkey contributing about 85% to global cotton production. India is the second-largest cotton producer globally after China (Anonymous 2021). In India, it is cultivated over an area of 12.35 m ha with production and productivity of 34.06 million bales and 468.87 kg ha^{-1}, respectively (Kiruthika et al 2022). Among Indian states, Haryana is the 6^{th} largest cotton-producing state with an area of 0.74 m ha, production of 1.82 million bales and productivity of 419 kg ha^{-1} (Anonymous 2021). Cotton crop is attacked by several insect pests from germination to harvesting stage. These insect pests may be classified into sap sucking insects (aphids, jassids and whitefly) and chewing insects (bollworms, leaf-eating caterpillars, *etc*.). Before the introduction of *Bacillus thuringiensis* (*Bt*) cotton, insecticides were the only option to manage these insect pests (Razaq et al 2013) and nearly 50 per cent of the pesticides were sprayed on cotton for the control of bollworms, which accounts for major damage (Anonymous, 2014). Although the introduction of *Bt* cotton reduces bollworms problem in cotton but the problem of sucking pests remain as such. Therefore nowadays the insecticides are mainly applied to manage sucking pests. But the information on insecticide use patterns in different cotton-growing areas in the Haryana state is limited. Therefore, the present study is devised to gather information on farmers' knowledge, perception and practices of insect pest management in *Bt* cotton.

MATERIAL AND METHODS

Designing of survey schedule: The farmers' knowledge regarding cotton insect pest management was evaluated through a questionnaire. For this purpose, a preliminary survey was performed to develop a questionnaire. Major cotton-growing districts namely, Hisar, Sirsa and Bhiwani of Haryana state were selected and in each district five block and two villages per block were selected purposively based on total cotton area coverage and production. Personal interviews were conducted with five selected farmers from each village and thus, the total sample size of the respondents was 150 comprising 50 from each district. The blocks selected were namely, Hisar- I, Adampur, Hisar- II, Agroha and Barwala; Rania, Nathusera Chopta, Baragudha, Ellenabad and Sirsa and; Bhiwani, Loharu, Bawani Khera, Tosham and Kairu from Hisar, Sirsa and Bhiwani districts respectively. The villages selected were Dhansu and Shikarpur; Sadalpur and Kohli; Kaluwas and Kirtan; Shamsukh and Kirara and; Khedar and Iserheri from Hisar- I, Adampur, Hisar- II, Agroha and Barwala blocks respectively. The villages selected were Mangalia and Dhottar; Ding and Gudia Khera; Baragudha and Karamgarh; Beharwala Khurd and Poharka and; Darbi and Shahpur Begu from Rania, Nathusera Chopta, Baragudha, Ellenabad and Sirsa blocks respectively. Similarly, the villages selected were Manheru and Gauripur; Dhigawa Shamyan and Singhani; Milakpur and Barsi; Chhapar Rangran and Chhapar Jogian and; Shimliwas and Bhangarh from Bhiwani, Loharu, Bawani Khera, Tosham and Kairu blocks respectively.

Collection of data: The farmers were interviewed individually in the appropriate local language using a structured questionnaire. Questions were focused first on farmers' age, level of education, farm size and history of cotton cultivation. Subsequently, questions bordered on tests of farmers' knowledge of cotton insect pests, their damage to cotton, control measures adopted, farmers' perception of incidence, severity and yield loss, use of insecticides/ biopesticides, crop stage and frequency of application and constraints faced in insect pest management of cotton. On an average, each questionnaire took 30-40 minutes of the interview with each farmer.

Statistical analysis: Survey data were summarised and Chi-square test with Cramer's *V* for independent attributes was used to indicate the association between farmers in different districts using SPSS software (version 23).

RESULTS AND DISCUSSION

Farmers' profile: Details of farmers profile are given in Table 1. All the farmers (100.0%) were growing *Bt* cotton across the selected districts and none was reported to be growing non-*Bt* cotton. Yadav and Goel (2019) in survey during 2015-16 also observed same trend in Sirsa and Fatehabad districts of Haryana state. The major *Bt* hybrids cultivated by the respondents were BG II cotton hybrids namely, RCH 773, RCH 776, US 51, Ankur 3028 and US 81. The majority of the respondents (34.0%) were growing RCH 773 followed by RCH 776 which is cultivated by 31.3% of the respondents. The proportion of farmers growing cotton hybrids like US 51, Ankur 3028 and US 81 was 13.3, 13.3 and 8.0%, respectively. Rani and Selvaraj (2009), Singh et al (2013) and Hoshmath et al (2012) confirmed that majority of the farmers adopted *Bt*-cotton technology mainly because of more yield, less number of labour, high producer price, lower pest attack and decrease in volume of insecticides sprayed are supporting the present findings. Statistical analysis of data revealed that age of farmer, educational status of farmers, experience in cotton cultivation, type of *G. hirsutum* and hybrid use pattern among farmers is non-significantly associated within the district.

Farmers' knowledge and perception: Among the sucking pests, all the respondents were having knowledge of whitefly and leafhopper and targeted for control whereas in case of dusky cotton and red cotton bugs, all the respondents were having knowledge about these pests but no one was targeting for their control (Table 2). It may be because of less economic damage caused by these pests. Similarly, among other insect pests, despite knowing about tobacco caterpillar by 82.7% farmers, only 32.0% apply insecticides for control as it is sporadic pest and it is only considered as the important pest on cotton at some locations. The farmers were also having knowledge of bollworms but no one was found to target for control. Previous studies have also reported that the introduction of *Bt* cotton resulted in the reduction in the population of bollworms, damage of cotton squares and bolls (Rani and Selvaraj 2009). There is a significant association between farmers' knowledge of thrips and targeting for control (V=0.22 and 0.26) among different districts. Similarly, in the case of pink bollworm among bollworms and tobacco caterpillar among defoliators, there is a significant association between farmers in having knowledge among different districts.

Insect pests' incidence, severity, and yield losses: There is less variation in insect pests' incidence, severity and yield losses among three districts of Haryana (Table 3). On the mean basis, the incidence, severity and yield losses were categorized as high for whitefly and leafhopper by farmers (100.0, 86.6, 82.7%; 76.6, 74.0, 64.6%, respectively) among sucking pests. These findings are in agreement with the those of Sharma and Pampapathy (2006) wherein *Bt* cotton-growers reported a high incidence of sucking pests including whitefly and jassid, as *Bt* cotton hybrids are not possessing resistance for sucking insect pests. The incidence, severity and yield losses by other sucking pests like thrips, mealybug, aphid, dusky cotton bug and red cotton bugs were categorized into low to medium.

Table 1. Profile of cotton growing farmers in selected districts of Haryana (n=150)

Parameters	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
Age of farmer					
<40 years	13.00 (26.00)	12.00 (24.00)	10.00 (20.00)	11.67 (23.30)	NS
40-60 years	35.00 (70.00)	34.00 (68.00)	36.00 (72.00)	35.00 (70.00)	
>60 years	2.00 (4.00)	4.00 (8.00)	4.00 (8.00)	3.33 (6.70)	
Educational status					
Illiterate	7.00 (14.00)	7.00 (14.00)	6.00 (12.00)	6.67 (13.30)	NS
Primary	11.00 (22.00)	9.00 (18.00)	7.00 (14.00)	9.00 (18.00)	
Middle	14.00 (28.00)	13.00 (26.00)	15.00 (30.00)	14.00 (28.00)	
Matric	13.00 (26.00)	15.00 (30.00)	14.00 (28.00)	14.00 (28.00)	
Graduation & above	5.00 (10.00)	6.00 (12.00)	8.00 (16.00)	6.33 (12.70)	
Literacy rate	(86.00)	(86.00)	(88.00)	(87.00)	
Experience in cotton cultivation					
<5 years	5.00 (10.00)	4.00 (8.00)	10.00 (20.00)	6.33 (12.70)	NS
5-15 years	18.00 (36.00)	13.00 (26.00)	16.00 (32.00)	15.67 (31.30)	
> 15 years	17.00 (54.00)	33.00 (66.00)	24.00 (48.00)	24.67 (56.00)	
Type of *Gossypium hirsutum*					
Bt cotton	50.00 (100)	50.00 (100)	50.00 (100)	50.00 (100)	NS
Non- *Bt* cotton	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	
Major *Bt* hybrids					
RCH 773 BG II	19.00 (38.00)	15.00 (30.00)	17.00 (34.00)	17.00 (34.00)	NS
RCH 776 BG II	18.00 (36.00)	14.00 (28.00)	15.00 (30.00)	15.67 (31.30)	
US 51 BG II	7.00(14.00)	7.00 (14.00)	6.00 (12.00)	6.67 (13.30)	
Ankur 3028 BG II	4.00 (8.00)	8.00 (16.00)	8.00 (16.00)	6.67 (13.30)	
US 81 BGII	2.00 (4.00)	6.00(12.00)	4.00 (8.00)	4.00 (8.00)	

Figures without parentheses are numbers of farmers and figures in parentheses are data in percentage; Chi-square test with Cramer's *V* to indicate the strength of association; NS = non-significant

Table 2. Percentage of farmers having knowledge of cotton pests and targeting for control (n=150)

Scientific name	Local Name	Name of district								Tests	
		Hisar		Sirsa		Bhiwani		Mean			
		Pest	Target	Pest	Target	Pest	Target	Pest	Target	Pest	Target
Sucking pests											
Thrips tabaci	Churda	88.00	68.00	90.00	76.00	72.00	46.00	83.30	63.33	0.22*	0.26**
Bemisia tabaci	Safedmakkhi	100	100	100	100	100	100	100	100	NS	NS
Amrasca biguttula biguttula	Tela	100	100	100	100	100	100	100	100	NS	NS
Phenacoccus solenopsis	Milibug	78.00	0.00	88.00	0.00	84.00	0.00	83.30	0.00	NS	NS
Aphis gossypii	Chepa	84.00	4.00	86.00	6.00	74.00	2.00	81.30	4.00	NS	NS
Oxycarenus hyalinipennis		100	0.00	100	0.00	100	0.00	100	0.00	NS	NS
Dysdercus cingulatus	Laldi	100	0.00	100	0.00	100	0.00	100	0.00	NS	NS
Bollworms											
Helicoverpa armigera	Hari sundi	42.00	0.00	56.00	0.00	38.00	0.00	45.30	0.00	NS	NS
Pectinophora gossypiella	Gualbi sundi	12.00	0.00	28.00	0.00	8.00	0.00	16.00	0.00	0.24*	NS
Earias vittella & *E. insulana*	Kanto wali sundi	60.00	0.00	64.00	0.00	42.00	0.00	55.30	0.00	NS	NS
Spodoptera litura	Tambacu keeda	88.00	12.00	92.00	16.00	68.00	8.00	82.70	32.00	0.28**	NS

Chi-square test with Cramer's V to indicate the strength of association. NS = non-significant,* significant at 5% level, ** significant at 1% level

Based on mean value, the incidence, severity and yield losses were negligible due to bollworms. Among defoliators, the incidence, severity and yield losses due to tobacco caterpillar were rated from low to medium by the majority of farmers. There is a significant association between farmers estimating incidence and severity of leafhopper but yield losses are non-significantly associated with districts. In case of bollworms, the incidence, severity and yield losses, all are

Table 3. Percentage of farmers estimating incidence, severity and yield loss of major cotton insect pests in selected districts (n=150)

Insect	Categor	Incidence					Severity					Yield loss				
		Hisar	Sirsa	Bhiwani	Mean	Tests	Hisar	Sirsa	Bhiwani	Mean	Tests	Hisar	Sirsa	Bhiwani	Mean	Tests
Sucking pests																
Thrips tabaci	Low	64.00	52.00	70.00	62.00	NS	68.00	60.00	74.00	67.33	NS	94.00	86.00	90.00	90.00	NS
	Medium	36.00	48.00	30.00	38.00		32.00	40.00	26.00	32.67		6.00	14.00	10.00	10.00	
	High	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Bemisia tabaci	Low	0.00	0.00	0.00	0.00	NS	0.00	0.00	0.00	0.00	NS	0.00	0.00	0.00	0.00	NS
	Medium	0.00	0.00	0.00	0.00		14.00	16.00	10.00	13.33		18.00	10.00	24.00	17.30	
	High	100	100	100	100		86.00	84.00	90.00	86.67		82.00	90.00	76.00	82.70	
Amrasca biguttula biguttula	Low	0.00	0.00	0.00	0.00	0.25**	0.00	0.00	0.00	0.00	0.24**	0.00	0.00	0.00	0.00	NS
	Medium	24.00	10.00	36.00	23.33		28.00	12.00	38.00	26.00		38.00	26.00	42.00	35.33	
	High	76.00	90.00	64.00	76.67		72.00	88.00	62.00	74.00		62.00	74.00	58.00	64.67	
Phenacoccus solenopsis	Low	94.00	90.00	92.00	92.00	NS	98.00	94.00	96.00	96.00	NS	98.0	96.00	98.00	97.30	NS
	Medium	6.00	10.00	8.00	8.00		2.00	6.00	4.00	4.00		2.00	4.00	2.00	2.70	
	High	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Aphis gossypii	Low	94.00	96.00	92.00	94.00	NS	96.00	98.00	94.00	96.00	NS	98.00	100	96.00	98.00	NS
	Medium	6.00	4.00	8.00	6.00		4.00	2.00	6.00	4.00		2.00	0.00	4.00	2.00	
	High	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Oxycarenus hyalinipennis	Low	14.00	16.00	20.00	16.70	NS	30.00	32.00	34.00	32.00	NS	92.00	96.00	90.00	92.70	NS
	Medium	86.00	84.00	80.00	83.30		70.00	68.00	66.00	68.00		8.00	4.00	10.00	7.30	
	High	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Dysdercus cingulatus	Low	88.00	82.00	90.00	86.70	NS	90.00	86.00	92.00	90.00	NS	92.00	94.00	94.00	93.30	NS
	Medium	12.00	18.00	10.00	13.30		10.00	14.00	8.00	10.00		8.00	6.00	6.00	6.70	
	High	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Bollworms																
Old world bollworm (*Helicoverpa armigera*)	Nil	94.00	96.00	94.00	94.70	NS	94.00	96.00	94.00	94.70	NS	90.00	94.00	92.00	92.00	NS
	Low	6.00	4.00	6.00	5.30		6.00	4.00	6.00	5.30		10.00	6.00	8.00	8.00	
	Medium	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Pink bollworm (*Pectinophora gossypiella*	Nil	98.00	96.00	94.00	96.00	NS	98.00	96.00	94.00	96.00	NS	94.00	96.00	98.00	96.00	NS
	Low	2.00	4.00	6.00	4.00		2.00	4.00	6.00	4.00		6.00	4.00	2.00	4.00	
	Medium	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Spotted and spiny bollworm (*Earias vittella* & *E. insulana*)	Nil	92.00	90.00	86.00	89.30	NS	92.00	90.00	86.00	89.30	NS	84.00	76.00	88.00	82.70	NS
	Low	8.00	10.00	14.00	10.70		8.00	10.00	14.00	10.70		16.00	24.00	12.00	17.30	
	Medium	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	
Defoliator																
Tobacco caterpillar (*Spodoptera litura*)	Low	72.00	52.00	70.00	64.70	0.19*	80.00	58.00	74.00	70.67	0.20*	64.00	54.00	78.00	65.30	0.20*
	Medium	28.00	48.00	30.00	35.30		20.00	42.00	26.00	29.33		36.00	46.00	22.00	34.70	
	High	0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00		0.00	0.00	0.00	0.00	

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant,* significant at 5% level, ** significant at 1% level

non-significantly associated with districts. The incidence, severity and yield losses due to tobacco caterpillar among defoliators were dependent of districts with association values of V= 0.19, 0.20 and 0.20, respectively.

Pests incidence during different cotton crop growth stages: There is less variation in farmers estimating pest incidence during different crop durations in all three districts (Table 4). The highest pest incidence was at 61-90 days after sowing (DAS) by cent per cent farmers followed by 70.0, 18.0 and 9.3% farmers at 91-120, >120 and 31-60 DAS, respectively and minimum pest incidence (6.0%) was estimated by farmers at <30 DAS. This may be due to the prevailing high temperature, high humidity and lush growth of plants in July-August (61-90 DAS) providing conducive environment for higher pest incidence. Statistical analysis of data revealed that estimation of pest incidence at different crop growth stages is independent of the district.

Use of insecticides at different crop durations: On the mean basis, the cent per cent farmers targeted the pests for their control at 61-90 DAS as maximum pest incidence was estimated at this crop duration followed by 61.3, 13.3 and 8.0% farmers at 91-120, >120 and 31-60 DAS, respectively (Table 5). None of the farmers in three districts used insecticides when the crop is <30 days old as very low pest incidence was recorded at this stage. The data revealed that use of insecticides by farmers against target pests at different crop duration is non-significantly associated with districts.

Components of insect pest management: The knowledge of the sampled respondents about the various components of pest management ranged from 8.6 to 52.6% (Table 6). Among the various components of plant protection, about 52.6% of respondents read the label and leaflets before applying the insecticides. Majority of the farmers (32.6%) used the seed supplied by agricultural input dealers which is claimed to be already treated with insecticides. Only 23.3% of the farmers knew about the economic thresholds of insects. The knowledge of yellow sticky trap, use of stickers and bioagents of cotton insect pests were found to be 28.0, 18.0 and 8.67%. Relatively a few farmers (8.6%) were familiar with bioagents in *Bt* cotton. Yang et al (2005) reported that farmers have moderate level of awareness about natural enemies in their *Bt* cotton field farmers. Statistical analysis of data revealed that farmers' knowledge about various components of pest management is independent of the district.

Information source for inputs in cotton cultivation: Most of the farmers (44.6%) were dependent on agricultural input dealers for information on inputs in cotton cultivation followed by the recommendations from CCS HAU/CICR-RS, and relatives/neighbours *i.e.*, 34.6 and 32.0% farmers, respectively. It is followed by farmers using information disseminated through TV/ social media, farmers' group, newspaper and radio, respectively. A few farmers were also depending on self-observations (4.6%) and farmers' field group (3.3%) as an information source for input in cotton cultivation. However, statistical analysis revealed that it was non-significantly associated with districts (Table 7). The

Table 4. Percentage of farmers estimating pests incidence during different crop duration in selected districts (n=150)

Crop duration	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
< 30 DAS	8.00	6.00	4.00	6.00	NS
31-60 DAS	10.00	10.00	8.00	9.33	NS
61-90 DAS	100	100	100	100	NS
91-120 DAS	70.00	68.00	72.00	70.00	NS
>120 DAS	18.00	20.00	16.00	18.00	NS

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant

Table 5. Percentage of farmers using insecticides against target pests at different crop durations in selected districts (n=150)

Crop duration	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
< 30 DAS	0.00	0.00	0.00	0.00	NS
31-60 DAS	8.00	10.00	6.00	8.00	NS
61-90 DAS	100	100	100	100	NS
91-120 DAS	60.00	68.00	56.00	61.33	NS
>120 DAS	14.00	14.00	12.00	13.33	NS

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant insecticide usage seems to be highly influenced by agriculture input dealers. The main reason for this dependence appeared to be that most of farmers depend on the dealers for credit. Rani and Selvaraj (2009) also reported that primary source of information for input in cotton cultivation is the local input dealer.

Constraints faced by cotton growers in insect pest management: The major constraints faced by farmers in insect pest management were the high cost of insecticides, weather problems and lack of knowledge about bioagent which are represented by 85.3, 83.3 and 80.0% farmers, respectively (Table 8). Yadav and Goel (2019) reported that the major problem in implementing the plant protection measures was the high cost of pesticides (99.0%), adulteration (82.0%), lack of knowledge about correct dose (54.5%) and non-availability of pesticides (38.0%).The next major constraint was the insubstantial control or poor efficacy of insecticides, which was reported by 73.3% farmers. It

Table 6. Percentage of farmers having knowledge about various components of pest management (n=150)

Particulars	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
Economic threshold level	24.00	30.00	16.00	23.33	NS
Labels on pesticide containers	52.00	62.00	44.00	52.67	NS
Use of stickers	16.00	26.00	12.00	18.00	NS
Bioagents	8.00	12.00	6.00	8.67	NS
Seed treatment	32.00	38.00	28.00	32.67	NS
Yellow sticky trap	30.00	36.00	18.00	28.00	NS

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant

Table 7. Percentage of farmers getting the information for inputs in cotton cultivation (n=150)

Source of information	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
Radio	10.00	8.00	8.00	8.66	NS
TV/Social media	12.00	12.00	10.00	11.33	NS
Newspaper	8.00	10.00	8.00	8.66	NS
CCS HAU/CICR-RS	38.00	34.00	32.00	34.66	NS
Relative or neighbor	32.00	30.00	34.00	32.00	NS
Agriculture input dealer	44.00	50.00	40.00	44.67	NS
Farmers' group	12.00	10.00	10.00	10.67	NS
Farmers' field school	4.00	4.00	2.00	3.33	NS
Self-observation	4.00	4.00	6.00	4.67	NS

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant

Table 8. Percentage of farmers indicating different constraints faced by cotton growers in insect pest management (n=150)

Constraints	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
High cost of pesticides	92.00	88.00	76.00	85.33	NS
Non-availability of good seeds	52.00	42.00	64.00	52.67	NS
Weather problems	100.00	82.00	68.00	83.33	0.35**
Non-availability of labour	66.00	72.00	52.00	63.33	NS
Insubstantial control	74.00	78.00	68.00	73.33	NS
Lack of knowledge about traps	48.00	32.00	72.00	50.70	0.33**
Non-availability of chemicals	52.00	40.00	66.00	52.67	0.21*
Lack of knowledge about bioagents	88.00	62.00	90.00	80.00	0.32**
Non-availability of sprayer	0.00	0.00	32.00	10.07	0.49**

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant,* significant at 5% level, ** significant at 1% level

could be due to various reasons like use of spurious insecticides or inappropriate selection of insecticides against target pests, improper application methodology, poor conditions of spraying equipments, unfavourable weather condition, *etc*. Kumar (2014) revealed that the most important constraints faced by the farmers in *Bt* cotton production were non-availability of labour, unfavourable weather conditions, inadequate insect pest control, high cost of pesticides and fertilizers, and non-availability of inputs *etc.*, which also corroborate the present findings. The other constraints faced by cotton growers were the non-availability of labour (63.3%), non-availability of good seed (52.6%) and the non-availability of insecticides on time (52.6%) and lack of knowledge about traps (50.7%). Mohanasunderm (2015) also reported that among the various problems faced by *Bt* cotton farmers in cotton cultivation, the shortage of labour at the time of cultivation and harvesting is the major constraint in cotton production. On mean basis, about 10.0% of farmers responded to non-availability of sprayers among selected districts. Statistical analysis of data revealed that the constraints faced by cotton growers in insect pest management namely, non-availability of sprayers, weather problems, lack of knowledge about traps, lack of knowledge about bioagents and non-availability of insecticides are significantly associated with districts.

Insecticide use pattern in cotton: The neonicotinoid compounds were the most commonly used insecticides by the majority of cotton growers for control of sucking pests (Table 9). Among neonicotinoids, the leading insecticides were thiamethoxam and imidacloprid which were used against sucking pests of cotton by 81.3 and 78.0% farmers, respectively. The next leading insecticide was emamectin benzoate by 40.0% farmers mainly for thrips management in cotton followed by fipronil, dimethoate and monocrotophos. Cypermethrin, acepahte, and ethion were used by 8.0, 7.3 and 6.0% farmers, respectively. Neem based insecticides and buprofezin were found to be used by 8.0 and 6.0% farmers, respectively. Some novel insecticides were also reported to be used by farmers which include flonicamid, diafenthiuron, spiromesifen and dinotefuran. Spinetoram was found to be used by 2.7% farmers in Hisar and Sirsa districts only, while none of the farmer reported its use in Bhiwani district. The use of majority of insecticides in cotton against target pest is independent upon district except monocrotophos, emamectin benzoate, fipronil and flonicamid which are significantly associated with the district.

Table 9. Percentage of farmers using different insecticides in cotton in selected districts (n=150)

Insecticide	Name of district				Tests
	Hisar	Sirsa	Bhiwani	Mean	
Organophosphates					
1. Monocrotophos	14.00	12.00	30.00	18.67	0.21*
2. Dimethoate	16.00	18.00	24.00	19.33	NS
3. Acephate	6.00	10.00	6.00	7.33	NS
Synthetic pyrethroids					
1. Cypermethrin	8.00	12.00	4.00	8.00	NS
2. Ethion	4.00	12.00	2.00	6.00	NS
Neonicotinoids					
1. Imidacloprid	76.00	82.00	76.00	78.00	NS
2. Thiamethoxam	80.00	86.00	78.00	81.33	NS
3. Dinotefuron	6.00	8.00	2.00	5.33	NS
Other groups					
1.Emamectin benzoate	40.00	60.00	20.00	40.00	0.33**
2. Fipronil	20.00	46.00	6.00	24.00	0.39**
3. Flonicamid	8.00	20.00	4.00	10.67	0.22*
4. Azadirachtin	10.00	10.00	4.00	8.00	NS
5. Diafenthiuron	6.00	10.00	4.00	6.67	NS
6. Buprofezin	6.00	8.00	4.00	6.00	NS
7. Spiromesifen	6.00	8.00	2.00	5.33	NS
8. Spinetoram	4.00	4.00	0.00	2.70	NS

Chi-square test with Cramer's *V* to indicate the strength of association. NS = non-significant,* significant at 5% level, ** significant at 1% level

In present investigation, the use of insecticides was high, probably because farmers assume that the only solution to pest problems is to use insecticides. Similar findings have been reported in many developing countries where growing dependence on synthetic insecticides/non-IPM for the control of crop pests is alarming (Kumela et al 2018, Zhang et al 2018, Ochilo et al 2018, January et al 2018). Present results of group-wise stratification of insecticides usage in Haryana are in line with the findings of Dhawan et al (2011).

CONCLUSION

It can be inferred that among sucking pests, whitefly, *B. tabaci*, leafhopper, *A. biguttula biguttula* and up to some extent thrips, *Thrips tabaci* are considered important pests by the farmers in *Bt* cotton hybrids grown in Haryana in kharif 2019 and farmers are using conventional as well as newer molecules for the management of these insect pests. The insecticide usage pattern varied greatly between three selected districts indicating no definite insecticide usage pattern among the major cotton-growing districts of Haryana. Efforts are needed to educate the farmers about the identification of pests and natural enemies, as well as the establishment of economic thresholds for pests and adopting suitable control measures of pest management.

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Effect of Green Waste Compost on Growth and Root Morphology of Ornamental Shrubs

Samil Kamboj, R.K. Dubey*, Manisha Dubey[1], Sandeep Sharma[1] and Ravi Deepika

Department of Floriculture and Landscaping, [1] Department of Vegetable Science
Punjab Agricultural University Ludhiana-141 004, India
**E-mail: rkduey.flori@pau.edu*

Abstract: The present investigations on effect of green waste compost on growth and root morphology of the present investigations on ornamental shrubs were undertaken at, Punjab Agricultural University, Ludhiana, during March 2021-February 2022. One year old three ornamental shrubs (*Murraya paniculata, Hibiscus rosa-sinensis* and *Tabernaemontana coronaria*) were transplanted in polybags of size 9×7 inches, filled with green waste compost (GWC) and soil in different proportions during second week of March 2021. The maximum plant height (57.33 cm), root collar diameter (10.00 mm), number of primary branches per plant (3.30), shoot dry weight (23.70 g), primary root diameter (12.55 mm) and plant nitrogen percentage (3.73%) were observed in *Hibiscus rosa-sinensis* in GWC 50% + soil 50%. Maximum root dry weight (13.95 g) and primary root length (11.61 cm) were observed in *Murraya paniculata* in GWC 50% + soil 50%. Plants growing in media (GWC 100%) were somewhat stunted and chlorotic for several weeks after transplanting probably due to higher levels of EC (4.15 dS/m) and pH (7.62). The growing media composition was observed to be an ideal in GWC 50% + Soil 50% i.e. pH (7.22), EC (1.64 dS/m), OC (2.39%), available nitrogen (0.55%), available phosphorus (0.22%), available K (0.47%), total porosity (38.50%) and water holding capacity (123.67%) for nursery raising of ornamental shrubs. Based on growth performance evaluated, the sequence for growth parameters is *Hibiscus rosa-sinensis*>*Murraya paniculata*>*Tabernaemontana coronaria*. GWC 50% + soil 50% proved superior growing medium for growth of three ornamental shrubs.

With the growth of environmental concern in the world, nurserymen are also looking for sustainable substitutes of media for growing plants. The green waste compost being easily available and economical is one substitute that can be used. The green waste compost mainly consist of compost made from garden litter, fresh leaves, dead leaves, pruned parts of young shrubs and trees, bark, wood etc. It contains low level of micro pollutants due to its origin from plant source, therefore hold better environmental feasibility and further can be used in organic farming (Bustamante et al 2016). Where chemical fertilizer release nutrient immediately and get depleted, green waste releases nutrients slowly in the media hence preferred as long-term nutrient source. The reason is presence of high number of lignocellulosic compounds (75%) which take more time to decompose by aerobic microbes (Gabhane et al 2012). A considerable amount of phosphorus and potassium in green waste compost stimulate better plant and root growth. Manipulation of highly degraded urban soils by green waste compost is beneficial as it improves physicochemical properties of soil together with enhancing microbial processes. Green waste compost maintains pH by improving buffering capacity of soil, enhance nutrient retention as well as decrease fertilizer requirements. Presence of humic acid and micronutrients also act as growth promoting factors for the plants. Well decomposed green waste reduce soil borne pathogen.

The present study is centralized on effect of green waste compost on the growth habit of three shrubs namely *Murraya paniculata* (Kamini), *Hibiscus rosa-sinensis* (China rose) and *Tabernaemontana coronaria* (Double chandni). These shrubs are highly used in large landscape gardening and backyard gardens in India in various forms as to line streets, for shade purpose, in topiary, as a specimen plant, as natural screen as hedge or along a pillar or wall and as a pot plant. *Hibiscus rosa-sinensis* (China rose) has white to pink, red colour flowers belonging to Malvaceae family. *Murraya paniculata* (Kamini) is an evergreen fragrant white flowering shrub belonging to Rutaceae family. It produces flower from April to August and grown in tropical as well as subtropical countries. Similarly, *Tabernaemontana coronaria*is an evergreen shrub of Apocynaceae family. It produces double white flowers throughout the year. This shrub has thick foliage and is used as a specimen shrub or also as a shrubbery border. It is branched and bush shrub with origin from India, China and Thailand. Its leaves are oval with wavy margins. The objective was to standardize the optimum proportions of green waste compost for nursery raising of flowering shrubs and to evaluate the growth performance of nursery of flowering shrubs raised in green waste compost.

MATERIAL AND METHODS

The present experiment was carried out during period of March 2021-February 2022 at Punjab Agricultural University, Ludhiana. In experiment treatments combination were used three shrubs species (*Hibiscus rosa-sinensis, Murraya paniculate* and *Tabernaemontana coronaria*) and five different media compositions (green waste compost 100%, green waste compost 75% + soil 25%, green waste compost 50% + soil 50%, green waste compost 25% + soil 75% and FYM 50% + soil 50% as control). The experiment was conducted by following factorial completely randomized design. The height of plant was recorded at bi-monthly intervals. Root collar diameter was measured at soil level with the help of digital Vernier caliper at bi-monthly intervals. The number of primary branches per plant was manually counted at bi-monthly intervals. Shoot and root dry weight was recorded after uprooting the plant, shoot was separated from the roots and then sun dried for 2-3 days before being dried in the oven. After sun drying samples were placed in oven at 60°C for 48 hours for drying to achieve a constant weight. The length of primary root was recorded after uprooting the plants at the end of experiment. Diameter of primary root collar was measured after uprooting the plants at the end of experiment. All the media were analyzed for their chemical characteristics. Different physical and chemical properties of growing media were determined to identify the factors that affect the growth of ornamental shrubs. The air-dried media samples were used for specific determination of maximum water holding capacity, bulk density (g cm^{-3}), nitrogen (Subbiah and Asija1956), phosphorus was measured using Olsen method and potassium was measured using flame photometer given by Jackson (1967). The maximum water holding capacity (WHC) was determined with the help of Keen's box (steel box of 5.0 cm I.D.) and 4 cm height with perforated bottom and a filter paper disc fixed with a steel ring at the bottom end) for the soil samples.

RESULTS AND DISCUSSION

Plant height (cm): The plant height varied statistically significantly in 3 shrub species (Table 2). The interaction between GWC treatments × months × species was also significant. With respect to the treatments maximum plant height was in T_3 (GWC 50% + Soil 50%) in *H. rosa-sinensis* (65.49 cm), *T. coronaria* (56.38 cm) and *M. paniculata* (54.30 cm) as compared to other treatments. The three different shrub species were statistically significant irrespective of treatments and months. The height maximum was in *H. rosa-sinensis* (57.33 cm), whereas minimum in *M. paniculata* (47.90 cm)which was at par with *T. coronaria* (50.28 cm). Among the three shrub species, maximum increment in plant height was observed in *H. rosa-sinensis* (92.58%) followed by *T. coronaria* and *M. paniculate* in T_3 (GWC 50% + Soil 50%) as compared to other treatments from period of April to February. Irrespective of GWC treatments maximum increment in plant height was in *T. coronaria* (24.34%) followed by *H. rosa-sinensis*, *M. paniculate* from June to August i.e. rainy season as compared to other months, irrespective of GWC treatments. Prasad et al (2011) reported that the growth rate of plants was poor with the use of 100% composted green waste. Better aggregate stability of the media amended with compost also enhanced flower quality in Chrysanthemum (Singh et al 2016). Additionally, there was reduction of vegetative development when more than 50 percent green waste compost amendment as in *Gloxinia sylvatica*, *Justicia carnea*, *Lysmachia congestiflora, Pelargonium*, *Petunia* and *Salvia* due to higher pH of growing media leading to immobilization of various plant nutrients and higher heat generated via compost decomposition in the media mixture (Wilson et al 2002, Thangam et al 2009, Dubey et al 2013).

Number of primary branches per plant: The interaction between number of primary branches, months and shrub species was statistically significant. The mean maximum number of primary branches was in *H. rosa-sinensis* (3.30) whereas minimum in *M. paniculata* (1.45) which was at par with the *T. coronaria* (1.85). Different treatments were statistically significant, maximum number of primary

Table 1. Characteristics of different growing media

Treatments	pH	EC	N (%)	P (%)	K (%)	Bulk density (g/cm^3)	Maximum water holding capacity (%)
T1-GWC 100%	7.62	4.15	0.64	0.27	0.55	0.97	166.70
T2-GWC 75% + Soil 25%	7.32	3.86	0.58	0.23	0.50	1.05	151.38
T3-GWC 50% + Soil 50%	7.22	1.64	0.55	0.22	0.47	1.13	123.67
T4-GWC 25% + Soil 75%	7.13	1.35	0.29	0.15	0.39	1.27	112.78
T5-FYM 50% + Soil 50% (Control)	7.76	2.76	0.38	0.16	0.41	1.19	92.42

branches in *H. rosa-sinensis* (3.63) and *M. paniculata* (2.16) in T_3 (GWC 50% + soil 50%) as compared to other treatments. In *M. paniculata* number of primary branches was maximum in T_3 i.e., 2.16 which was at par with the T_4 (GWC 25% + soil 75%) and T_1 (GWC 100%). In *T. coronaria* T_3 showed maximum number of primary branches (1.96) which was at par with T_2 (GWC 75% + soil 25%), T_5 (control) and T_4 whereas minimum number of primary branches (1.57) was observed in T_1. *H. rosa-sinensis* showed mean maximum number of primary branches (3.63) in T_3 which was at par with T_2 (GWC 75% + soil 25%) and T_5 (FYM 50% + soil 50%) and significantly different from other treatments and minimum number of primary branches (2.79) was in T_1. Early flowering and a greater number of flowers with longer blooming period observed in Kalanchoe (*Kalanchoe blossfeldiana*) when treated with paddy straw compost with burnt rice husk (Kaur et al 2015). Mugnai et al (2007) observed that the plant height was reduced with increased CGW concentration treatments, resulting in lesser and thinner branches. The plants cultivated in T 75 and T 100 exhibited low quality and growth.

Shoot dry weight (g): The interaction between different levels of green waste compost treatments and shrub species was significant. Among the shrub species with respect to treatments mean maximum shoot dry weight was in *H. rosa-sinensis* (23.70 g) in T_3 and mean minimum shoot dry weight was in *T. coronaria* (12.00 g) in T_1 which was at par with *T. coronaria* in T_5 (12.69 g), T_4 (12.83 g) and T_2 (13.00 g). Irrespective of different treatments shoot dry weight was statistically significant, mean maximum shoot dry weight was in *H. rosa-sinensis* (19.88 g) followed by *M. paniculate* (17.39 g) and *T. coronaria* (13.30 g). Among the different treatments results was also statistically significant. Mean maximum (20.43g) and mean minimum (14.61g) shoot dry weight irrespective of three different shrub species was in T_3 and T_1 respectively. Similar results were reported by Gong et al (2018) in calendula and geranium where shoots dry weight was significantly greater in medium amended with green waste vermicompost as compared to unamended media. Papafotiou et al (2005) reported that when peat was substituted organic waste compost (OWC) by 50% led to increase in fresh weight of leaves in *Codiaeum variegatum*.

Root collar diameter (mm): Root collar diameter increases with the increase in growth of the shrubs. It has positive correlation with the growth of the plant. The interaction between GWC treatments × months × species was statistically significant (Table 3). The different treatments were statistically significant. The mean maximum root collar diameter was in *H. rosa-sinensis* (11.55 mm) followed by *T. coronaria* (9.81 mm) and *M. paniculata* (7.12 mm) in T_3 as compared to other

Table 2. Effect of green waste compost based growing media on plant height (cm) of different shrub species

Species	Treatment/ Month	April	June	August	October	December	February	Mean	Mean (species)
Hibiscus rosa-sinensis	T_1	34.89	40.77	47.41	52.78	57.28	60.89	49.00^C	57.33^A
	T_2	41.22	47.83	58.33	65.15	71.00	74.11	59.61^{AB}	
	T_3	43.44	50.99	63.77	72.22	78.88	83.66	65.49^A	
	T_4	39.00	45.70	55.11	62.33	66.11	67.33	55.93^B	
	T_5 (Control)	40.33	46.50	56.11	61.11	66.44	69.22	56.62^B	
Mean		39.78^D	46.36^D	56.15^C	62.72^{BC}	67.94^{AB}	71.04^A		
Murraya paniculata	T_1	32.78	36.66	43.88	47.66	51.22	53.77	44.33^C	47.90^B
	T_2	34.78	38.44	46.33	49.89	54.11	57.55	46.85^{BC}	
	T_3	36.67	43.15	54.26	59.56	64.11	68.04	54.30^A	
	T_4	35.11	39.11	45.00	48.77	51.44	55.66	45.85^{BC}	
	T_5 (Control)	34.89	40.11	48.41	52.31	55.55	57.89	48.19^B	
Mean		34.85^F	39.49^E	47.58^D	51.64^C	55.29^B	58.58^A		
Tabernaemontana coronaria	T_1	29.77	35.44	40.77	46.78	49.00	52.77	42.42^D	50.28^B
	T_2	38.89	44.89	55.16	59.77	61.33	64.66	54.12^{AB}	
	T_3	36.67	44.61	57.83	62.66	66.18	70.33	56.38^A	
	T_4	34.00	40.72	49.11	51.40	52.22	55.22	47.11^{CD}	
	T_5 (Control)	37.55	45.11	53.00	55.77	57.00	59.66	51.35^{BC}	
Mean		35.38^D	41.15^C	51.17^B	55.27^{AB}	57.14^A	60.52^A		

See Table 1 for details

Table 3. Effect of green waste compost based growing media on root collar diameter (mm) of different shrub species

Species	Treatment/ Month	April	June	August	October	December	February	Mean	Mean (species)
Hibiscus rosa-sinensis	T_1	7.73	8.50	9.21	10.10	10.56	11.33	9.57^{BC}	10.00^{A}
	T_2	7.68	8.76	9.76	10.57	11.17	12.60	10.09^{B}	
	T_3	8.67	10.32	10.84	11.96	13.14	14.35	11.55^{A}	
	T_4	7.33	8.14	9.19	9.30	9.66	10.07	8.95^{C}	
	T_5 (Control)	7.85	8.75	9.84	10.35	10.43	11.89	9.85^{B}	
Mean		7.85^{E}	8.89^{D}	9.77^{CD}	10.46^{BC}	10.99^{B}	12.05^{A}		
Murraya paniculata	T_1	2.94	3.99	4.93	5.79	6.66	7.13	5.24^{D}	5.95^{B}
	T_2	3.43	4.13	5.09	5.95	6.60	7.59	5.46^{CD}	
	T_3	3.70	4.92	6.75	8.34	8.84	10.18	7.12^{A}	
	T_4	3.39	4.69	5.94	7.07	7.56	7.97	6.10^{B}	
	T_5 (Control)	3.00	4.04	5.67	6.34	7.24	8.71	5.83^{BC}	
Mean		3.29^{F}	4.36^{E}	5.68^{D}	6.70^{C}	7.38^{B}	8.32^{A}		
Tabernaemontana coronaria	T_1	5.37	6.07	6.80	7.33	7.48	8.41	6.91^{D}	8.32^{C}
	T_2	6.63	7.68	8.47	9.39	9.90	10.25	8.72^{B}	
	T_3	7.50	8.65	9.55	10.29	10.80	12.07	9.81^{A}	
	T_4	6.47	7.61	7.98	8.59	8.92	9.52	8.18^{BC}	
	T_5 (Control)	5.94	7.21	7.84	8.22	8.66	9.98	7.98^{C}	
Mean		6.38^{E}	7.44^{D}	8.13^{CD}	8.76^{BC}	9.15^{B}	10.05^{A}		

See Table 1 for details

Table 4. Effect of green waste compost based growing media on number of primary branches per plant of different shrub species

Species	Treatment/ Month	April	June	August	October	December	February	Mean	Mean (species)
Hibiscus rosa-sinensis	T_1	1.77	2.00	2.44	2.99	3.55	4.00	2.79^{C}	3.30^{A}
	T_2	2.00	2.33	3.77	3.66	4.22	5.11	3.52^{AB}	
	T_3	1.66	2.22	3.77	4.00	4.77	5.33	3.63^{A}	
	T_4	1.99	2.55	2.99	2.99	3.78	4.00	3.05^{BC}	
	T_5 (Control)	2.44	2.55	3.44	3.66	4.33	4.77	3.53^{AB}	
Mean		1.97^{C}	2.33^{C}	3.28^{B}	3.46^{B}	4.13^{A}	4.64^{A}		
Murraya paniculata	T_1	0.89	1.11	1.77	1.77	1.89	2.33	1.63^{AB}	1.45^{B}
	T_2	0.22	0.77	0.88	0.88	1.11	1.22	0.85^{C}	
	T_3	0.11	0.11	2.89	3.11	3.33	3.44	2.16^{A}	
	T_4	0.55	0.44	2.11	2.11	2.11	2.66	1.66^{A}	
	T_5 (Control)	0.22	0.22	1.00	1.11	1.55	1.66	0.96^{BC}	
Mean		0.40^{B}	0.53^{B}	1.73^{A}	1.80^{A}	2.00^{A}	2.26^{A}		
Tabernaemontana coronaria	T_1	1.33	1.33	1.33	1.55	1.78	2.11	1.57^{B}	1.85^{B}
	T_2	1.66	1.77	2.00	2.00	2.00	2.33	1.96^{A}	
	T_3	1.66	1.77	1.66	1.88	2.00	2.11	1.85^{AB}	
	T_4	1.77	1.77	1.89	1.89	2.00	2.11	1.90^{A}	
	T_5 (Control)	1.77	1.89	2.00	2.00	2.00	2.11	1.96^{A}	
Mean		1.64^{B}	1.71^{B}	1.78^{B}	1.86^{AB}	1.95^{AB}	2.15^{A}		

See Table 1 for details

treatments. Among the 3 shrub species, mean root collar diameter was maximum in *H. rosa-sinensis* (10.00 mm) which was at par with *T. coronaria* (8.32 mm) and minimum in *M. paniculata* (5.95 mm) irrespective of different treatments. Gong et al (2018) also reported significantly higher stem diameter of calendula and geranium amended with green waste vermicompost as compared to unamended media. Beldaet al (2013) reported increase in growth parameters in viola and calendula seedlings grown in media amended with 25 or 50% vermicompost as compared to control.

Root dry weight (g): The interaction between different levels of green waste compost treatments and shrub species was significant. Among the shrub species with respect to treatments mean maximum root dry weight was in *M. paniculata* (13.95 g) which was at par with the *H. rosa-sinensis* (13.89 g) in T_3 and minimum root dry weight was observed in *T. coronaria* (9.50 g) inT_1which was at par with *M. paniculata* (9.67 g) in T_1 and *T. coronaria* (10.00 g) in T_4. Among the different treatments results were statistically significant. Mean maximum (13.48 g) and mean minimum (10.12 g) root dry weight irrespective of three different shrub species was in $T_3$and T_1, respectively. Irrespective of different treatments, root dry weight of three shrub species statistically significant, mean maximum root dry weight was observed in *H. rosa-sinensis* (11.89 g) followed by *M. paniculata* (11.69 g) and *T. coronaria* (10.87 g). Gong et al (2018) reported

Table 5. Effect of Green waste compost based growing media on shoot dry weight (g) of different shrub species

Treatments	Species			
	Hibiscus rosa-sinensis	*Murraya paniculata*	*Tabernaemontana coronaria*	Mean
T_1	17.67^{f}	14.17^{h}	12.00^{i}	14.61^{D}
T_2	19.67^{cd}	15.00^{gh}	13.00^{i}	15.89^{C}
T_3	23.70^{a}	21.59^{b}	16.00^{g}	20.43^{A}
T_4	20.07^{c}	17.43^{f}	12.83^{i}	16.77^{B}
T_5 (Control)	18.32^{ef}	18.78^{de}	12.69^{i}	16.59^{B}
Mean	19.88^{A}	17.39^{B}	13.30^{C}	

See Table 1 for details

Table 6. Effect of green waste compost based growing media on root dry weight (g) of different shrub species

Treatments	Species			
	Hibiscus rosa-sinensis	*Murraya paniculata*	*Tabernaemontana coronaria*	Mean
T_1	11.19^{d}	9.67^{fg}	9.50^{g}	10.12^{D}
T_2	12.17^{bc}	12.33^{b}	11.00^{de}	11.83^{B}
T_3	13.89^{a}	13.95^{a}	12.61^{b}	13.48^{A}
T_4	10.33^{ef}	11.08^{de}	10.00^{fg}	10.47^{D}
T_5 (Control)	11.50^{cd}	11.44^{cd}	11.24^{d}	11.39^{C}
Mean	11.89^{A}	11.69^{A}	10.87^{B}	

See Table 1 for details

Table 7. Effect of green waste compost based growing media on primary root length (cm) of different shrub species

Treatments	Species			
	Hibiscus rosa-sinensis	*Murraya paniculata*	*Tabernaemontana coronaria*	Mean
T_1	8.77^{ef}	9.22^{de}	9.22^{de}	9.07^{D}
T_2	10.27^{bc}	9.61^{cd}	9.22^{de}	9.70^{B}
T_3	11.55^{a}	11.61^{a}	10.94^{ab}	11.36^{A}
T_4	9.44^{de}	9.94^{cd}	8.38^{f}	9.25^{CD}
T_5 (Control)	9.33^{de}	9.88^{cd}	9.33^{de}	9.51^{BC}
Mean	9.87^{A}	10.05^{A}	9.42^{B}	

See Table 1 for details

significantly greater root dry weight of calendula and geranium amended with green waste vermicompost as compared to unamended media.

Primary root length (cm): Among the different shrub species with respect to treatments mean maximum primary root length was in *M. paniculata*(11.61cm) which was at par with *H. rosa-sinensis* and *T. coronaria* in T_3 (GWC 50% + soil 50%), whereas mean minimum was in *T. coronaria* in T_4. which was at par with *H. rosa-sinensis* in T_1. Shrub species irrespective of different green waste compost media treatments were statistically significant, mean maximum primary root length was in *M. paniculata* (10.05 cm) which was at par with *H. rosa-sinensis* and mean minimum primary root length was in *T. coronaria* (9.42cm). The treatments were also statistically significant, irrespective of three ornamental shrub species, mean maximum (11.36 cm) and mean minimum (9.07 cm) primary root length was observed in T_3 and T_1 respectively. De Falco et al (2021) reported that with the addition of green compost at the lowest dosages (C50-C25) to the growth substrate showed significantly increase in main root length in chard, sorrel and radish. The beneficial impact on the roots may be suitable at the nursery level to prepare seedlings for transplanting with established root systems that can effectively recover from transplant shock.

Primary root diameter (mm): The interaction between different levels of green waste compost treatments and shrub species shows significant results. Among the different shrub species mean maximum primary root diameter was in *H. rosa-sinensis* (12.55 mm), whereas mean minimum was in *M. paniculate* (8.15mm) which was at par with *M. paniculate* (8.31 mm and 8.72 mmi in T_5 and T_4 respectively.) Irrespective of three ornamental shrub species results were statistically significant, mean maximum (10.75 mm) and mean minimum (9.25 mm) primary root diameter in T_3 and T_1respectively. Amongst the shrub species irrespective of different green waste compost media treatments, mean maximum (11.35 mm) and mean minimum (8.76 mm) primary root diameter was in *H. rosa-sinensis* and *M. paniculate ,* respectively.

Survival percentage: The interaction of different GWC treatments × shrubs species was non-significant (Table 9). Among the different shrub species irrespective of treatments, *H. rosa-sinensis* showed mean highest survival percentage of. 88.00 % which was at par with *T. coronaria* and *M. paniculata.* Survival percentage showed significant results among different GWC treatments irrespective of shrub species. Plants treated with T_3 showed highest survival percentage which was at par with T_2, T_4 and T_5. Prasad et al (2001) also reported the rate of plants was poor with the use of 100% composted green waste.

Table 8. Effect of green waste compost based growing media on primary root diameter (mm) of different shrub species

Treatments	Species			
	Hibiscus rosa-sinensis	*Murraya paniculata*	*Tabernaemontana coronaria*	Mean
T_1	10.38^{c}	8.15^{f}	9.24^{de}	9.25^{C}
T_2	10.49^{c}	9.40^{de}	9.33^{de}	9.74^{B}
T_3	12.55^{a}	9.24^{de}	10.48^{c}	10.75^{A}
T_4	11.52^{b}	8.72^{ef}	9.86^{cd}	10.03^{B}
T_5 (Control)	11.82^{b}	8.31^{f}	9.22^{de}	9.78^{B}
Mean	11.35^{A}	8.76^{C}	9.63^{B}	

See Table 1 for details

Table 9. Effect of green waste compost based growing media on survival percentage of different shrub species

Treatments	Species			
	Hibiscus rosa-sinensis	*Murraya paniculata*	*Tabernaemontana coronaria*	Mean
T_1	73.33^{a}	73.33^{a}	73.33^{a}	73.33^{B}
T_2	93.33^{a}	86.67^{a}	80.00^{a}	86.67^{AB}
T_3	93.33^{a}	93.00^{a}	93.33^{a}	93.77^{A}
T_4	86.67^{a}	93.33^{a}	93.33^{a}	91.11^{A}
T_5 (Control)	93.33^{a}	86.67^{a}	86.67^{a}	88.89^{A}
Mean	88.00^{A}	86.67^{A}	85.33^{A}	

See Table 1 for details

CONCLUSION

Among various green waste compost based growing media used for nursery raising of flowering shrubs, GWC 50% + soil 50% proved superior growing medium for growth of three ornamental shrubs than other media. Based on growth performance evaluated, the sequence for growth parameters is *Hibiscus rosa-sinensis*>*Murraya paniculata*>*Tabernaemontana coronaria*. The present study concluded that green waste compost up to level of 50% in combination with soil can be used for nursery growing of ornamental shrubs.

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Performance Evaluation of Different Furrow Openers for Sustainable Tillage: A Review

Dilwar Singh Parihar, Baldev Dogra[1] and Mahesh Kumar Narang

Department of Farm Machinery and Power Engineering, Punjab Agricultural University Ludhiana-141 004, India
[1]College of Agricultural Engineering and Technology, Choudhary Charan Singh Haryana Agricultural University Hisar-125 004, India
E-mail: dilwar-fmpe@pau.edu

Abstract: Controlled soil disturbance, low draft and vertical force requirements are among the primary characteristics of a practical and efficient furrow opener. The furrow openers should also be capable of maintaining acceptable surface residue retention, precise and uniform seed placement, and regular inter-plant spacing when integrated into a conservation seeding system. The objective of this paper is to study and review various furrow openers based on their performance in terms of soil disturbance, draft requirement, seeding performance, seed emergence, and residue handling ability. The furrow openers that were reviewed included single disc and double disc type, hoe, shoe, shovel, runner, and inverted T type furrow openers. The double disc type is the most effective in terms of soil disturbance, while the single disc type requires the lowest draft force. The hoe and shoe type furrow openers are the most accurate in seed placement, while the double disc and inverted t type furrow openers resulted in the highest seed emergence rates. The inverted T, shovel and runner type furrow openers are effective in residue handling, while the hoe, shoe and double disc type furrow openers struggles in the heavy residue fields. The selection of furrow opener should depend on specific farming needs.

Keywords: Conservation tillage, Draft requirement, Residue handling, Seeding performance

The purpose of tillage is to transform soil physical, structural, and ecological properties so that a healthier crop can be produced (Mohanty et al 2007). However, tillage weaken soil structure and consequently decrease soil water holding ability and disturb soil biology, having an adverse impact on the nutrient stock and storage ability of soil (Lal 2004, Farooq et al 2011, Devine et al 2014). The reduced tillage system, has been advertised as a low-budget, energy-efficient method of increasing crop yields (Bianchini and Magalhães 2008, Farooq and Nawaz 2014). There are different types of tillage practices available, including no-tillage, zero-tillage, and conservation tillage. In practice, still, there is high attention in the no tillage system. There are a few advantages to no-till seeding, including the reduction in field passes, the reduction in soil compaction, labor saving, time reduction, and reduced fuel consumption (Tebrugge and Bohrnsen 2000, Chen et al 2004, Sarauskis et al 2009). Zero tillage is economically greater, and additional grain yield was noted under zero tillage as compared to traditional farming techniques (Nagarajan et al 2002). There has been a rise in yield by 5 percent to 10 percent with zero- tillage technology and saving in sowing time by up to 70 percent as well as 60 percent savings in operating costs (Rautaray 2004). The purpose of conservation tillage is to ensure that the soil surface is covered with at least 30% crop residue after seeding. As a result of this, water and wind erosion will be prevented, as well as significant water loss from naked soil surfaces (ASABE Standards 2013). In comparison to bare and fallow soil, this process reduces erosion by 50% (Karayel 2009).

It is important to manage crop residue well because residue interferes with sowing operations, especially in rice fields, which is a serious limitation to conservation tillage (Carter 1994). Due to the variation of soil texture, weather circumstances, and soil properties, paddy soils have a compound nature in term of soil failure and draft requirement (Tagar et al 2014). It is therefore vital to use adequate machinery, as well as manage residues effectively, in order to ensure precise sowing operations. Residue management as well as mechanical factors which affect seed germination and emergence are seed damage while metering; uniformity in sowing and placement of seed; and fertilizer mixing with seed while placing in the furrow. The furrow opener is a most important element of a seeding system because it loosens the soil and opens a furrow to create finest seed zone conditions for plant. The work of furrow opener is to precisely place the seed and fertilizer simultaneously in the prepared seed bed and create optimum condition for seed germination. Seed emergence and crop yield have been used to evaluate furrow opener performance by several researchers. The furrow opener should be designed in such a way that it can perform the desired task precisely with minimum power requirement, particularly in no till paddy residue condition (Murray et al 2006) In tillage systems, there are several types of furrow openers, including hoes, chisels, and discs. These openers have their advantages and disadvantages in terms of soil disturbance, seeding performance, field condition and draft requirements (Chaudhuri 2001). Furrow openers should create a neat groove in the wet soil zone with least soil disturbance to escape mixing the top dry soil with the underlying moist soil at seed level. Degree of seed bed preparation can be determined by the size of soil aggregates in the seed bed preparation operation. The disc furrow openers have lesser soil disturbance as compare to the hoe type furrow openers (Parent et al 1993, Janelle et al 1995). These are typically adopted for effective seed placement as well as straw cutting to prevent the loose straw dragging and clogging with at the time of sowing. Throughout, several furrow openers double disc furrow performed adequately (Baker et al 1996). Furrow opener geometry, soil and straw conditions, directly affect the straw cutting performance of disc type furrow openers. Paddy soil requires a more draft force for tillage (Karayel and Sarauskis 2011) and loose paddy straw is pushed into soil without cutting, producing a hair pinning effect. Hence, augmentation of furrow opener performance in paddy filed under direct drilling conditions is still an important task.

Functional requirements of furrow openers: The functional requirements (Fig. 1) of a furrow opener are to:

- Open the furrow with desired depth.
- Maintain the uniformity while making furrow.
- Furrow opening with least disturbance in the soil.
- Avoid over compacting the side of furrow.
- Ensure that soil does not flow back into the furrow before seeding.
- Promote enough soil back in the furrow for seed coverage.

There are several type of furrow openers are used, in this article some major type of openers by different researchers are discussed. Some important parameters viz. soil disturbance, residue handling/cutting ability, draft requirement, seeding performance (depth, speed, and variations), plant emergence are considered for performance of furrow opener. Broadly, furrow openers can be divided into two categories tine and disc type furrow openers.

Classification of tine furrow openers: Tine furrow openers are simple in construction and most popular in conventional seeding system. In the conservation tillage system, narrow pointed type furrow openers are used for sowing wheat in paddy stubble condition, generally they are known as Hoe type furrow openers (Barr 2018). Godwin and O'Dogherty (2007) define narrow tine tools as those that operate at depths between 1 and 6 times their widths (i.e. depth/width ratio = 1:6). Now a day's various narrow furrow openers are being used for sowing such as knife point, Inverted T furrow opener and spear point furrow opener shown in Figure 2 (Murray et al 2006, Desbiolles and Leonard 2008).

Several other types furrow opener used in conservation tillage seeding systems contain duck foot and curved chisel (Murray et al 2006, Hasimu and Chen 2014). Generally, the no tillage furrow opener designed for low or minimum soil disturbance while sowing. Narrow tine openers are considered for conservation tillage system because they create less soil disturbance and have a lower draft force than other tine openers (Solhjou et al 2012).

Tine Furrow Openers

Hoe type: There are several types of hoe type furrow openers, which include tines or chisels shaped to penetrate the soil vertically in the soil. A tube attached to the hollow tine generally has an open back end, where seed is conveyed. A pointed hoe digs furrow according to the depth setting of the furrow opener. It lifts and pushes the top soil towards the sides and forms a V-shaped groove. This type of furrow opener performs fine under extensive range of soil conditions but not in residue fields. Advantages of hoe type opener are they penetrate in the soil with less vertical load, low cost, easy maintenance. And lastly, they do not form mark on the surfaces at the sides of furrows. Some disadvantages of less residue cutting ability, obstruct with large stone and higher soil movement depending upon the shape of furrow opener. Baker (1976) explored the hoe, triple disc and chisel-type furrow openers in soil bin having sandy loam soil under no tillage system. In the hoe type furrow opener, 27 % wheat seedling emerged with four-time lesser vertical force requirement as compare to disc type furrow openers. The seeding performance of hoe type of openers were at par in term of seeding depth because hoe type openers place the seed at desired depth. However, shovel and shoe type opener do not place the seed at required depth. Sandy clay loam and loamy sand soil are best suited for the hoe type furrow openers for attaining better performance for separation of seed and fertilizer (Chaudhuri 2001). It is common for hoe furrow openers to move out of the soil depth (Altuntas et al 2006).

Shoe type: This kind of furrow opener delivers seeds and fertilizer concurrently in distinct bands at the desired depth. Its boot is protected by metal covering to avoid obstruction. Fertilizer is commonly placed in a band at the side of the seeds at identical depth. The furrow opener forms a narrow channel in the soil. The length of the shoe aids in pressing the bottom of the furrow. Study specifies that although the shoe-type openers had superior compaction of the furrow bottom and not as much of variation in depth (Chaudhuri 2001). The shoe-type furrow opener, with either a single or twin boot, is used for sowing in heavy and medium soils, where seeds are placed at 20 to 70 millimeters deep. However, this type of opener has the tendency to sink the soil depth (Altuntas et al 2006).

Shovel type: The shovel type opener is a tapered pointed furrower. The principal edge of the opener is a sharp-pointed. The opener is mounted on the standard with the help of bolts for easy replacement. At the back of the boot one or two tubes attached for seed and fertilizer distribution. In comparison to hoe or shoe type furrow openers, shovel type furrow openers have a more versatile function. Shovel type opener are easier to fabricate as compared to disc type openers (Altuntas et al 2006). These type furrow openers are the widely used in seed drills for trashy, stony, and light to medium soils shovel-type openers are used. Commonly there are three shovels used i.e. reversible, single point shovel and spear point shovel. In stony and root infested fields, shovel type openers are recommended. It is easy in assembly, inexpensive and easily repairable. In a study it was observed that the shovel type furrow opener attains low draft requirement and less soil penetration force for highest seed emergence rate (Altuntas et al 2006). Different types of furrow openers are given in the Figure 3.

Runner type: The runner type opener is used for crop which are sown at shallow depth such as maize. A backward sword shaped blade with sharp edge penetrate in the soil and form a furrow with minimum soil disturbance. It operates in fine prepared seed bed and used typically for shallow sowing crops. It compacts the soil in the furrow bottom because of its length. Abernathy and Porterfield (1969) evaluated the different sized runner-type furrow openers, for compaction analysis in sandy soil. They concluded that the runner type furrow openers does not compact the sandy soils. Also,

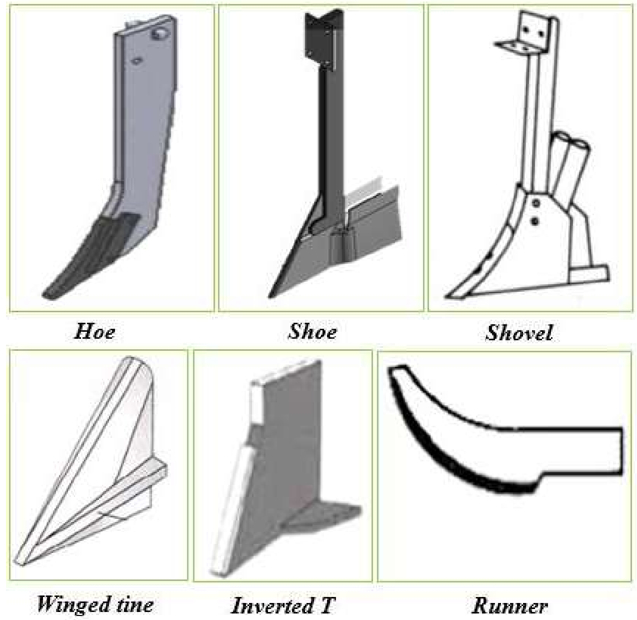
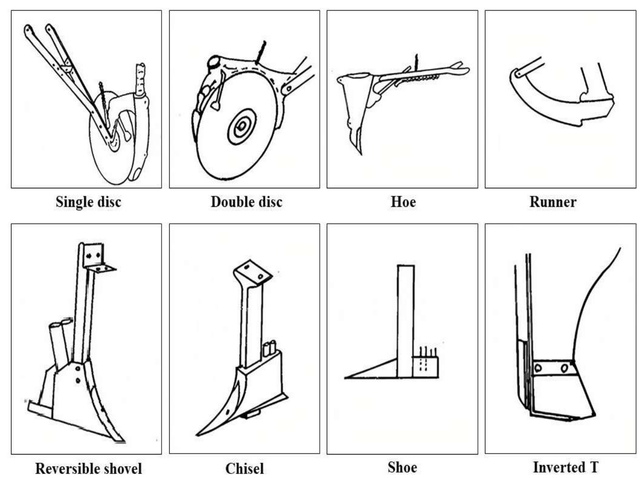


Fig. 2. Different types of tine furrow openers



Source: Chaudhuri 2001

Fig. 3. Different type of furrow openers

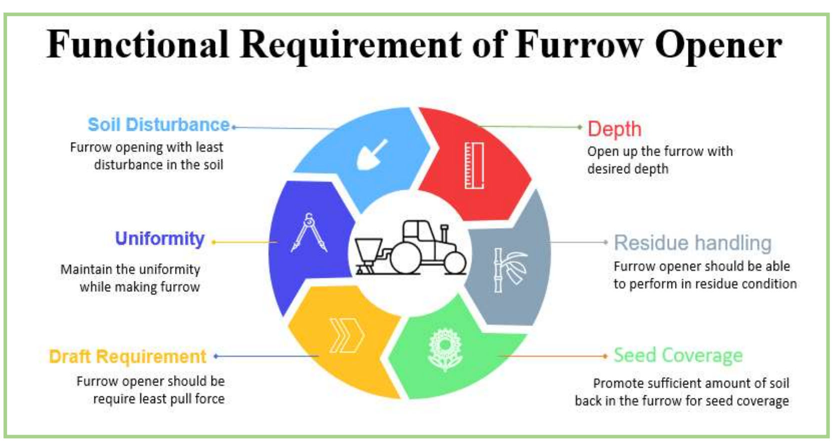


Fig. 1. Functional requirement of furrow openers

runner type furrow openers are not recommended in fields having less moisture content because it hampers in making good seed to soil contact. For shallow seeding depth, runner-type furrow opener could be best suited because it creates less soil disturbance. Moreover, the soil backfill in the runner type furrow openers is less due to shallow operating depth.

Inverted-T furrow opener: An inverted T furrow opener basically looks like a knife-type furrow opener with plane wings fixed to both sides and trailing from the bottom (Murray et al 2006). The Inverted-T furrow openers are recommended for direct sowing under upland soil conditions. The inverted -T makes a fine cut, therefore not cause too much soil disturbance, and establish good plant emergence under no-tillage conditions. The wings of the furrow openers creates an inverted t shaped profile in the soil. The purpose of making inverted t shape is to create sub surface disturbance to maintain favorable condition for the seed germination and growth within the furrow. As a result, moisture is conserved within the seed zone by keeping moister and deeper soil undisturbed. The wheat emergence was found better in comparison with other types of furrow openers. Additionally, Du et al (2004) found that inverted T openers produced the highest percentage of the emergence of sorghum compared with a winged point, narrow point, and triple disc openers. A total of 90% of the plants emerged in a single day in the Inverted-T-shaped slots, after which the rest emerged one to two days later. Furthermore, Baker (1976) found that an inverted T opener had the lowest vertical force (penetration resistance) in sandy loam soil at a depth of 38 mm, followed by a hoe and triple disc openers. It is possible to reduce moisture loss in the seed zone by inverting the T opener so that the seeds are shattered subsurface and high humidity chambers can be created for seed storage also, they have good residue handling capability (Aikins et al 2019).

Winged tine openers: Inverted T openers have wings on both sides and without wing narrow pointed openers can also be fixed with wings (Hasimu and Chen 2014). In winged type opener can be modified with various position of wings in horizontal and vertical positions and several lift heights and widths (McKyes 1985). If the lift height of the wing is increased the soil movement by the furrow opener also increases. As compare to same sized knife furrow opener lateral soil movement by winged furrow opener in much significant and the backfill by this furrow opener is also less in same working conditions (Hasimu and Chen 2014). The main advantage of winged furrow openers is it can effectively handle the residue as compare to without winged furrow opener (Aikins et al 2019). The draft requirement by the winged furrow openers is more than the hoe furrow opener. The higher draft requirement may be due to increase in surface are by wings in the furrow opener (McKyes 1985).

Disc type furrow openers: Disc type furrow openers are available as single, double, and triple disc types. Even though flat, plain, and notched disc, curved disc, waved and ripple types can be used individually as a furrow opener, they are pulled at an angle with the direction of motion to cut and shift the soil for furrow making. To achieve minimum soil disturbance and higher speed in seeding operation single and double disc type furrow openers are preferred (Ashworth et al 2010). Disc type furrow openers are usually demand higher vertical load for penetrating in the soil and maintaining uniformity in the depth of sowing (Murray et al 2006). Although, it was found that the disc openers has lower soil disruption and depth variation which results in good plant emergence and better crop establishment.

Single disc: Generally single disc type openers have a large diameter up to 2 feet. It may be plane disc, notched, waved, ripped, or curved disc for residue cutting and making furrow by soil movement (Fig. 4). These types of furrow openers cut a furrow in the topsoil and drive the furrow portion to the side, in that way disturbing to the topsoil. Single disc furrow openers are extensively used in the cereal crop sowing. Single disc type furrower may be mounted as aligned, sole angle or multiple angle type. By disc and tilt angle both width and depth of furrow can be managed. For sowing in the trashy or mulched field disc, type furrower are performed well.

Double disc: Disc type furrow opener create furrow with the least disturbance but also provide better residue handling than the tine type furrow openers (Yang et al 2016). This allows them to operate in heavy straw load condition with minimum depth variation. However, it requires enough soil strength to cut the straw efficiently. In loose soil condition straw not cut properly hence choking or bulldozing may be occurred during the operation (Ashworth et al 2010). When double disc type furrow opener penetrates in the soil the cutting edge of the disc cut and displace the top soil and form a V shaped furrow. The seed tube is placed in such a position that the seed is delivered at the place where the trailing edge of the disc can bury the seed in the furrow bottom. Generally, for speedy operation in the trashy land, double disc type furrow opener provide efficient performance. Disk-type furrow openers can be operated under various soil conditions.

Since Disc type furrow openers can work in dense soil conditions adequately hence have need of big and strong frame, therefore it makes them costlier. Ahmad et al (2017) investigated that double disc furrow openers require more pull force than single disc furrow opener, when the greater operating depth is required. Double disc furrow opener can also be consisting of two plane, notched, waved, or ripped

Table 1. Research findings on furrow openers and performance parameters

Reference	Furrow openers	Operating speed (kmph)	Findings
Soil disturbance			
Brandelero et al 2015	Double disc	3.4-9.2	Soil disturbance area increased up to 23.7 % which leads to poor seed germination rate and badly affect the soybean yield by reducing up to 20 %.
Godara et al 2015	Shovel and Shoe	2-4.5	Soil disturbance was more for shovel type furrow opener with comparison to shoe type furrow opener because of wider design of shovel type furrow opener.
Hashimu and chen 2014	Hoe, Winged hoe, and Spoon type	2.7-8.1	Considering both soil stepping and draft force requirement, the hoe opener showed better performance than the winged hoe and spoon openers.
Francetto et al 2016	Hoe and Disc		Hoe type furrow opener has more disturb area of soil as compare to disc type furrow openers.
Seeding performance			
Burce et al 2013	Hoe, Tine, and Double Disc	4-8	Hoe furrow opener showed no significant difference in furrow shape even in hard soils with great soil penetration, and the variation of the lateral and vertical seed distributions was reasonable.
Karad and Gaikwad 2018	Shovel and Disc	2-4.5	The use of disc furrow opener achieves the uniform depth as well as uniform width of cut and hence there is uniform growth of plants and ultimately there is increment in farmer's yield.
Altikat et al 2012	Hoe, Disc and Wing hoe	2.7-8.1	Hoe type furrow openers had the lowest coefficient of variation (17.04%) followed by winged hoe type seeder (24.13%) and disc type seeder (21.02%).
Draft requirement			
Altuntas et al 2006	Hoe shoe and shovel	2-4.5	Lowest soil penetration resistance, draft and tuber emergence was found in shovel type furrow opener
Karad and Gaikwad 2018	Shovel and Disc	2-4.5	Shovel type furrow opener produce greater soil disturbance with ultimately requires unnecessarily more draft force for pulling of tractor
McLaughlin et al 2019	Hoe and triple disc	2.7-5.4	The main difference between the two opener types was the approximately 200 N per opener higher draft for the hoe opener
Ahmad et al 2017	Various disc type furrow openers (single disc; tooth-type; notched-type; double disc)	7-10	The draft and vertical forces for double disc and toothed-type single disc furrow openers were the highest and lowest, respectively for all operating depths and speeds
Seed emergence			
Altikat and Celik 2012	hoe, disc, and wing hoe type openers	2.7-8.1	The highest emergence percentage (77.13%) was obtained with hoe type furrow openers followed by the disc and the winged hoe type openers (73.72% and 67.34%, respectively). Hoe-type furrow opener provided better sowing performance and seed emergence in comparison to the no-till seeders with disc- and wing hoe type furrow openers.
Doan et al. 2005	Disc hoe type	2.7-5.4	The results showed that the disc opener produced a faster emergence than the hoe opener. Disc opener showed an average of 36% faster emergence rate than hoe opener in canola crop.
Ahmad et al 2017	Various disc type furrow openers (single disc; tooth-type; notched-type; double disc)	7-10	Notched-type and smooth-type single disc furrow openers pushed the straw into the paddy field (straw hair-pinning), which might reduce crop emergence due to decreased soil-seed contact.
Residue handling			
Altikat and Celik 2012	hoe, disc, and wing hoe type openers		The higher the stubble the larger the coefficient of variation of sowing depth. Coefficient of variation of 18.72% at 12-cm stubble height increased up to 19.24% at 24-cm stubble height. However, variation coefficient of sowing depth was 17.15% under the standing stubble conditions and 19.14% under the flat stubble condition.
Ahmad et al 2017	Various disc type furrow openers (single disc; tooth-type; notched-type; double disc)	2.7-8.1	Double disc and smooth-type single disc furrow openers had the highest and lowest straw-cutting efficiencies, respectively

type discs (Fig. 5). Ahmad et al (2017) evaluated different cutting edges like smooth, toothed, notched disc for draft force requirement and straw cutting efficiency. Smooth disc type furrow opener observed more draft as compare to other type. However, straw cutting efficiency was highest in toothed type disc. The use of disc openers is not suitable for working in wet soil conditions due to the excessive accumulation of soil, which can disrupt their functionality. Disc openers pushes the top layer of the soil in the furrow bottom (Chaudhuri 2001, Desbiolles 2006, Yang et al 2016). Due to various herbicide and low moisture of top soil into the seed zone delay the seed germination and emergence of plant. Moreover, their various rotating parts and complex geometry make them costlier than other furrow openers (Chaudhuri 2001, Murray et al 2006).The efficiency of double disc opener depends upon various factors such as horizontal and vertical force, straw cutting ability, straw load, type of coulter, operating speed, and strength of soil (Kushwaha et al 1986).

Furrow opener performance indicators: The soil disturbance may be prime factor for assessing the performance of furrow opener. Straw cutting ability and draft requirement are other serval factor that can be considered while evaluation of furrow opener (Vamerali et al 2006). These performance measures subsequently power the seedling emergence rate, crop growth and biomass/grain yield (Chaudhuri 2001). Some are the major performance characteristics are discussed in this article are given below

- Draft requirement for furrow opener
- Soil disturbance in furrow opening
- Accuracy and uniformity of furrow depth and seed placement
- Seed emergence rate
- Ensure seed delivery, spacing, and seed-soil contact are not interfered with by residues;
- Facilitate good spread and optimum separation of seeds and fertilizer (Conte et al 2011, Francetto et al 2016).

Soil disturbance: Soil disturbance contains soil loosening and movement of soil triggered by a furrow opening device. When a furrow opener penetrates in the soil, soil particle moves from one place to another with the help of soil cutting tool, predominantly in the vertical track (Barr et al 2019). Also soil drives in all three dimension in the furrow (i.e. Forward, Lateral and Vertical) (Conte et al 2011). Soil disturbance triggered by the opener should be the least possible, as it is accountable for extreme soil water losses and serious weed problems. It also affects the seed and fertilizer scattering pattern undesirably. Soil disruption increased with increasing operational forward speed of seed drill (Godara et al 2015). Darmora and Pandey (1995) and Conte et al (2011) acknowledged the necessity for the measurement of individual soil disturbance limits including furrow cross-sectional area, draft force requirement and effective depth to develop a soil disturbance performance index. In general, sandy clay loam soil has low disturbance as compare to loamy sand soil with the same size of furrow opener.

Seeding performance: The furrow opener is a critical component of the seeding system, as it creates the furrow in which the seed is placed. The type of furrow opener used can have a significant impact on seeding performance, including seed placement accuracy, seed-to-soil contact, and soil moisture retention. One key factor that affects seeding performance is the depth and shape of the furrow created by the opener. Furrow openers that create a uniform and consistent furrow depth can improve seed placement accuracy

Table 2. Effect of furrow openers on the various parameter

Furrow opener	Functional requirement				
	Soil disturbance	Draft requirement	Residue handling	Seeding performance	Seed emergence
Hoe type		Low (FAO 2015)	Poor (FAO 2014)	Good (Darmora and Pandey 1995)	
Shoe	Low (Chaudhuri 2001)			Good (Altuntas et al 2006)	
Shovel		Low (Altuntas et al 2006).			High (Altuntas et al 2006)
Runner	Low			Poor (Abernathy and Porterfield 1969)	
Inverted T	Low (Baker 1976)		Moderate (Baker 1976; Aikins et al 2019)		High Du et al. (2004)
Winged tyne	High (Hasimu and Chen 2014)		Good (Aikins et al 2019)	Moderate (Hasimu and Chen 2014)	
Disc type	Low (Yang et al 2016)	Low (Tajudin and Balasubramanium 1995)	Good (Zhang et al 2016)		High (Munir et al 2012)

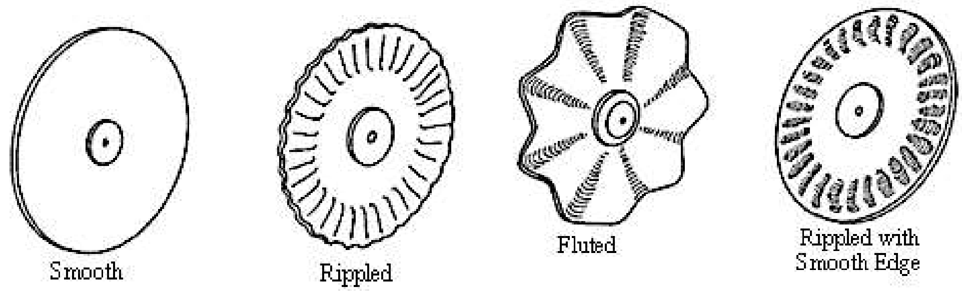


Fig. 4. Different type of disc used in disc furrow openers

Source: Francetto et al 2016

Fig. 5. Disc type furrow opener

and ensure consistent seed-to-soil contact. In addition, furrow openers that create a V-shaped furrow with sharp edges can help to retain soil moisture around the seed, which can promote germination and seedling emergence. A furrow opener's seeding performance can be evaluated by measuring the depth of seeding, the uniformity of seeding depth, and early crop growth. This includes crop emergence rate and plant density per unit area. Furrow opener type, soil condition, weather condition and residue type affect the seeding performance. Research has shown that different types of furrow openers can have varying effects on seeding performance. For example, a disc furrow opener may provide better seed-to-soil contact than a sweep or knife furrow opener, but may also create more soil disturbance. Similarly, a double-disk furrow opener may provide better soil moisture retention than a single-disk opener, but may also require more draft force. A furrow opener must be able to create and place the seed and fertilizer in furrows accurately with minimum depth variation. Attaining this leads to uniform seed emergence and batter crop growth, which ultimately provide higher yield (Aikins and Afuakwa 2008).

Draft requirement: The draft requirement of a furrow opener is an important consideration when selecting the appropriate tool for planting. Different types of furrow openers can have varying draft requirements, depending on factors such as the shape and size of the opener and the soil type. In general, furrow openers with a larger surface area or more aggressive design will have a higher draft requirement than those with a smaller surface area or less aggressive design. Draft is vital aspect that affect the seeding in the residue field. Low draft requirements by furrow opener leads to low fuel consumption and enable farmers to use smaller tractors. In developing contries it is desirable to reduce operating cost and use smaller tractor and machineries on the farm (Collins and Fowler 1996, Yao et al 2009). Draft requirement of furrow openers depend on the size of the wing of furrow opener, shape of furrow opener, depth of operation, residue condition, moisture content etc. The soil cutting theory also stated that the tool having wider cutting width need higher draft force as compare to narrow cutting edge (McKyes 1985). Darmora and Pandey (1995) evaluated seven different furrow openers, they also observed that draft force is related to opener width. Collins and Fowler, 1996 stated that for a 10 mm increase in seeding depth, the draft force increases by 20%. Altuntas et al 2006 evaluated the performance of different furrow openers for draft requirement and several other parameters, the outcome indicate that soil penetration resistance increase with forward speed of furrow opener. Also the shape of furrow opener influences the draft requirement.

Seed emergence: Furrow opener can significantly affect seed emergence, as different types of furrow openers can create furrows at varying depths and with different levels of soil compaction and disturbance. The depth of the furrow is particularly important, as shallow furrows may not provide enough soil cover for the seed, while deep furrows may make it difficult for the seed to emerge from the soil. Soil compaction around the furrow can also impede seed emergence, particularly in heavy soils or soils with high clay content. In addition, the placement of the seed in the furrow is critical, as some furrow openers may place the seed too close to the surface or too deep in the soil. To ensure optimal seed emergence and crop establishment, it is essential to select a furrow opener that is appropriate for the specific planting situation, considering factors such as soil type, crop type, and planting conditions. Seed emergence directly affects the plant population and ultimately the yield of the crop. Seed emergence depends upon the type of furrow openers, placement depth, seeding environment, moisture content, soil type and residue density (Altuntas et al 2006). In a study of the effects of soil compaction, depth variations, soil disturbances, and soil moisture content on emergence rate, it was determined that the soil moisture content within the furrow opener groove was critical (Siemens and Wilkins 2006). Choudhary et al 1985 investigated the effect of seeding performance on crop establishment in paddy harvested fields. Depth variation in sandy soil was 31% higher than in silty loam soil. The emergence percentage decreased with the increase in the stubble height (Altikat et al 2012).

Residue condition: The residue handling capacity varies according to type of furrow openers. The objective of residue handling is to confirm that the surface residue cover is minimum disturbed while avoiding the residues from interfering with the seed drill (Aikins et al 2019). The residue management of a furrow opener can be observed by the capacity to stop residue burial (hair pinning) with the soil into the furrow. That can considerably affect the germination and seed emergence rate. A furrow opener must be able to adequately handle crop residue during the seeding pass. Siemens and Wilkins et al (2006) conducted field studies to evaluated different furrow openers in the paddy stubble field for sowing of wheat. The residual moisture was present in the field for initial growth of the seed. To achieve this, deep furrow openers had the ability to place the seed in to moist condition and this result in better emergence even fluctuation in depth.

Comparison of disc and tine furrow openers: When compared with disc openers, tine furrow openers require less weight to penetrate hard soil and better adapt to these conditions. Generally, tine openers can create deeper furrows because of their improved penetration ability. Moreover, a tine opener causes more soil disturbance, especially at high speeds, so they cannot be operated at high forward speeds. Munir et al (2012) revealed that disk-type furrow opener has higher seedling emergence rate index (ERI) and grain yield as compare to the tine furrow opener. Particularly, in paddy stubble field it can be operated at higher speeds which creates good seed-soil contact and best soil conditions for water and nutrient availability to the crop. They are more likely to accumulate residue during sowing operations which leads to clumping and seeder blockage, ultimately which may affect seedling emergence (Altikat and Çelik 2012). The draft force required for disc furrow is comparatively less than the conventional tine furrow opener.

CONCLUSION

Research has shown that furrow openers that create a uniform and consistent furrow depth, provide adequate seed-to-soil contact, and minimize soil disturbance can improve seed emergence rates and ultimately crop yield. Disc type furrow openers causes greater soil disturbance due to there rolling and cutting ability at higher speeds. Moreover the disc openers causes less draft and better residue handling capacity due to sharp edges. However, the tine furrow openers has better penetration ability thus, work more consistently in hard soils. Also tine openers are limited due to clumping and blockage, hence not recommended for more than 3.5 kmph operating speed. Modification in the operating condition could help to achieve better performance with the tine openers such as increasing tine rake angle, tine width, and operating depth. Concave cutting edge on tine openers reduces soil disturbance and improve residue handling. For better results, it is preferable to operate a tine opener above its critical depth in order to prevent smearing and the exponential increase in draft. Compared to wingless openers, wing tine openers cause greater soil disturbance, but handle residue better. Through subsurface shattering and the creation of high humidity chambers dedicated to seeds, inverted T openers can help reduce moisture loss in the seed zone. Overall, the choice of furrow opener should be based on the specific planting conditions, considering factors such as soil type, residue management, crop type, and planting conditions. By selecting the appropriate furrow opener, farmers can optimize seeding performance, improve crop productivity, and ultimately achieve greater profitability.

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Laboratory Assessment of Tractor Operated Fertilizer Broadcaster Based on Operational Parameters

Azadwinder Singh, Anoop Kumar Dixit, Shiva Bhambota[1], Apoorv Prakash* and Gursahib Singh Manes

Department of Farm Machinery and Power Engineering
College of Agricultural Engineering and Technology, Punjab Agricultural University-141 004, India
[1]Krishi Vigyan Kendra Fatehgarh Sahib-140 406, India
**E-mail: apoorvprakash@pau.edu*

Abstract: The production of the major crops increased many folds from last two decades. This tremendous increase in agriculture is attributed to factors such as high yield varieties of seed, more area allotted to the various crops, good quality of fertilizers and improved methods of cultivation etc. The consumption of fertilizers has also increased many folds. To meet this high demand of fertilizers, centrifugal spreaders become into existence and due to low cost of operation and simplicity of use, these centrifugal spreaders are very popular. Therefore, a setup was made to evaluate the performance of the machine in Indian conditions. The maximum uniformity of spread was at 0^0 angle of fins. The coefficient of variation with apparent swath width was varied from 32.18% to 92.92%. With overlapping, coefficient of variation was 7.05 at notch setting 4 (opening area 9.40cm^2), disc speed of 400 rpm and fin angle of -10^0. At setting N2S2A2, the effective swath width was 6.75 m with 5.4 m left turn and 8.1 m right turn. The fertilizer application rate was 1500.42 kg h^{-1} corresponding to this setting.

Keywords: Angle of vanes, Centrifugal spreader, Orifice opening area, Skewness of spread

The tremendous increase in agriculture is attributed to factors such as high yield varieties of seed, more area allotted to the various crops, good quality of fertilizers and improved methods of cultivation. Solid chemical fertilizers are one of important sources for plant nutrition during its growing period, and works to improve the properties of soil (soil structure and the acidity degree)(Neina et al 2019). India is the second largest consumer of fertilizers in the world after China. In India, it is increased from 12.14 Mt in 1990-91 to 50 Mt in 2019-20 (Anonymous 2022). Increased inputs have played a pivoted role in augmenting production in India. Punjab, a leading agriculture state alone consumed 1.8 Mt of fertilizer during the year 2018-19 (Singh 2018). Efficient application with correct type and recommended dose of chemical fertilizer is important for achieving profitable yields (Singh 2018). The hand broadcasting is the most common method used for distribution of fertilizers. Besides being laborious and time consuming, its effectiveness depends on the skill and judgment of the farmer (Przywara et al 2020). Manually operated fertilizer spreaders can be used for applying recommended dose of fertilizer at different notch settings. Farmers of the region although shown interest in manual operated type fertilizer spreaders resulting in manufacturing of these spreaders. The popularity of this type of machine is due to its design characteristics, ease of handling and cleaning, economy and relatively small size for a given width of spread (Rahman et al 2018). However, has the disadvantage of low field capacity, producing an uneven distribution pattern and uneven application can lead to over dosage, causing material wastage, burnt crop and under application can leads to nutrient deficient plants and limited growth. The weight of the machine may cause stress on the neck of the operator. The mechanical broadcasters are better for fertilizer application but their performance depends on their design parameters as well as operational parameters (Villette et al 2012). The most famous mechanical broadcasters are the centrifugal distribution machine, and it was due to its low cost, low power requirement, simplicity of mechanical design and ease of maintenance.

There are many physical parameters which affect the performance of centrifugal broadcasters and every crop required different amount of fertilizer. Mass flow and rotational speed affect fertilizer centrifugal spreading is examined by Villette et al (2012) through potential interpretation in terms of the amount of fertilizer per vane. Kweon et al (2009) proposed a method which employed control of the drop location of fertilizer particles on the spinner disc to optimize the spread pattern uniformity. Research also been conducted on the simulation of particle motion to generate spread patterns for distributor worldwide. A tractor operated fertilizer broadcaster is commercially available and is being sold by manufacturers in the state to spread fertilizer with great ease and efficiency. There is no scientific data available regarding the performance of the machine. Hence, this study was carried out to evaluate tractor operated fertilizer broadcaster in Indian conditions and carry out necessary modifications based on the evaluation results.

MATERIAL AND METHODS

Description of tractor-operated fertilizer broadcaster: The tractor operated fertilizer broadcaster is a conical shaped broadcaster operated by the PTO (Fig. 1). The fertilizer-spreading disc consists of four metal vanes on its periphery for spreading the fertilizer (Table 1). Provision for adjustment of feed rate is present. In addition, slitter leavers have been provided to facilitate scattering at left or right side.

Experimental design and layout: Laboratory experiments were performed with Di-ammonium phosphate (DAP) as fertilizer to evaluate the selected broadcaster at Department of Farm Machinery & Power Engineering, PAU, Ludhiana to avoid any external disturbing factors such as wind speed. The broadcaster was attached with 36.76 kW tractors (Massey Ferguson 5245DI). A setup of 44 collecting channels (915 X 30 X 16.25 cm each) made up of iron bars and plastic sheets were fabricated and laid on the flat surface in a straight line perpendicular to the direction of fertilizer broadcaster (Fig. 2). The entire setup was able to cover the entire range of distributed fertilizer. The amount of fertilizer distributed from all the 44 channels was collected independently in separate plastic bags. Thereafter, the bags were weighed to analyse the effect of operational parameters on swath width and distribution pattern. Various parameters, such as notch setting, disc speed, angle of vanes, and type of fertilizer are known to affect the performance of broadcaster (Sharabasy and Afify 2007). In the study, 81 experiments in randomized block design encompassing 27 treatments were performed with three replications (9 treatments for each), comprising different combinations of independent parameters (disc speed, angle of vanes, and application rate). The data obtained for each dependent parameter (apparent swath width, fertilizer rate, and skewness of spread) with replications was analyzed using the statistical analysis software SAS 9.3 version.

Notch setting: Amount of fertilizer distribution was maintained through the metering device of the broadcaster. The application rate of granules was maintained by adjusting the size of an orifice to deliver the required amount of fertilizer. At level 1, the orifice was the smallest, which expelled the least amount of fertilizer. At level 8, the orifice area was maximum, which allowing the application of maximum amount of fertilizer to the field. The broadcaster was evaluated at three different notch settings (2, 4 and 6) based on recommended amount of fertilizer for crops and on preliminary trials.

Table 1. Brief specifications of fertilizer broadcaster

Parameters	Specification
Type of machine	PTO driven, mounted type
Source of power (hp)	Tractor, 35
Capacity (l)	361
Height (mm)	1130
Spreading width (m)	12-14
No. of vanes	4
Transmission ratio	1:1
Feed rate adjustments	8
Weight of the machine (kg)	63

Fig. 1. Stationary view of the fertilizer broadcaster

Fig. 2. Setup for evaluation of fertilizer broadcaster

Disc speed: Fertilizer broadcasting pattern and swath width of distribution varies according to the revolution speed of disc (rpm). Three levels of disc speed (300, 400 and 500 rpm) were selected by controlling the PTO rpm.

Angle of vanes: There are four C-shaped vanes (18 cm each) fastened to the circular disc of the broadcaster for providing the even distribution of fertilizer. Circular disc of this broadcaster equipped with five adjustments for angles of vanes, which allow the distribution pattern of spreading to be shifted towards right or left as per requirements. The ranges of vanes angle provided in the disc are +20°, +10°, 0°, -10° and -20°. Three different levels of vane angles (-10°, 0°, +10°) were selected.

Measurement of Parameters

Apparent swath width: Fertilizer broadcasters spread granular fertilizers and seed by centrifugal action. These fertilizers are spread to the certain width which is considered as apparent swath width.

Fertilizer rate (kgh^{-1}): The amount of fertilizer distributed by the broadcaster per unit time is called fertilizer application rate. It represents the quantity of fertilizer that flows out from the fertilizer broadcaster to the field.

$$\text{Fertilizer rate, (kg h}^{-1}) = \frac{W_t \times 60}{1000 \times T}$$

where, W_t= total fertilizer collected from channels in each treatment (g)

T = time taken in each treatment (min)

Skewness of spread:

$$\text{Skewness of spread} = \frac{\mu - v}{\sigma}$$

where, μ= mean of collected data

v = medium of collected data

σ = standard deviation

Evaluation for overlapping and without overlapping: The pattern acquire by fertilizer broadcasting was skewed. To get an even distribution, overlapping was done from each side of the pattern. A computer program was developed in PHP language. It has been found that there was a decrease in coefficient of variation after overlapping the swaths to certain spacing. To reduce the coefficient of variation three adjacent swaths were overlapped by computer program. Overlapping was done on both the side of pattern as left on left spacing and right on right spacing. After overlapping, the best result with maximum uniformity pattern was selected and corresponding spacing of maximum uniformity pattern was obtained from the computer program. It was observed that there was different spacing for left turn and right turn for maximum uniformity of spread.

RESULTS AND DISCUSSION

Effect of operational parameters on fertilizer rate: The fertilizer application rate displayed by the broadcaster increased with an increase in the level of notch setting (n = 2, 4, 6) at all disc speeds (300, 400 and 500 rpm) and angles of vanes (-10°, 0°, +10°). The mean values of fertilizer application rate were 91.01 kgh^{-1}, 595.50 kg h^{-1} and 1869.4 kgh^{-1} at notch setting levels 2, 4, and 6, respectively (Fig. 3). This increase was observed due to the increase in the size of orifice of the hopper with higher notch settings, leading to expulsion of higher amounts of fertilizer from the hopper to the circular disc. A similar trend of change in fertilizer application rate was recorded with varying disc speed of the broadcaster. As the disc speed was increased from 300 rpm to 400 rpm, the fertilizer application rate was increase from 851.90 kgh^{-1}to 966 kgh^{-1}. However, further increase in disc speed to 500 rpm lead to a decline in the fertilizer application rate to 738.02 kg h^{-1} (Fig. 3). This altering trend in fertilizer rate associated with disc speed maybe due to a change in the nature of vibrations. Sharabasy and Afify (2007) reported similar observation, stating that initial increase in disc speed led to a corresponding increase in the fertilizer application rate due to increase in vibration amplitude of the machine. Subsequently, fertilizer rate decreased as the amplitude decreased with further increase in disc speed.

The effects of notch setting and disc speed on fertilizer rate were statistically significant. However, no significant effect of angle of vanes was observed on fertilizer rate. The interactions between independent parameters, i.e. notch setting and disc speed, notch setting and angle of vanes, disc speed and angle of vanes were found significant (Fig. 4). Fertilizer rate were 856.68 kg h^{-1}, 848.82 kg h^{-1} and 850.41 kg h^{-1} at angle of fins -10°, 0° and +10° respectively.

Effect of operational parameters on apparent swath width: The apparent swath width exhibited variation with the notch setting of the fertilizer broadcaster (Fig. 5). As the notch setting level was increased from n=2 to n=4, the apparent

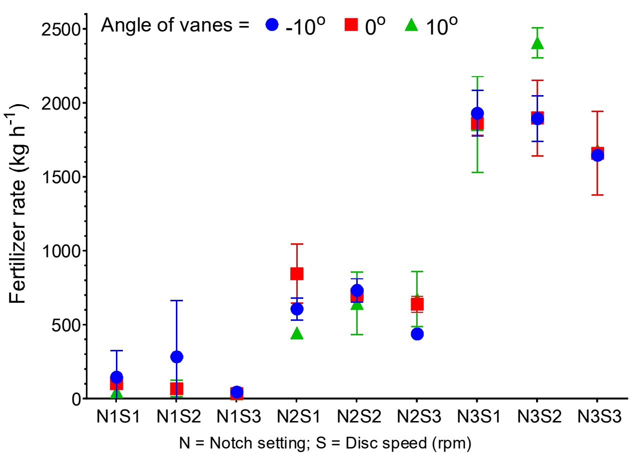
Table 2. Details of various independent parameters and their levels

Parameters	Levels	Remarks	Dependent parameters
Notch setting	3	To vary the application rate	• Apparent swath width (m)
Disc speed (rpm)	3	To vary the spread	• Fertilizer rate (kg/h)
Angle of vanes	3	To change the direction of flow	• Skewness of spread
Type of fertilizer	1	DAP	

swath width of distribution increased from 12.47 to 12.99 m, which further increased to 13.18 m at the notch setting level n = 6. This variation in apparent swath width is likely due to the increase in the amount of material present on the circular disc due to centrifugal action. Similar observations have been reported by Coetzee and Lombard (2011), wherein increase in fertilizer rate led to a concomitant increase in the apparent swath width. The overall trend of apparent swath width of the distributed fertilizer displayed a consistent increase with increase in the disc speed of the broadcaster. The mean of apparent swath width values recorded at various notch setting levels and angles of vanes were 12.01 m, 13.17 m, and 13.46 m at disc speeds of 300 rpm, 400 rpm and 500 rpm, respectively (Fig. 5). The observed increase in the apparent swath width may be explained by the increase in the angular speed of the disc, which provides higher terminal velocity to the fertilizer particles enabling them to travel a longer distance. The effect of disc speed on apparent swath width was more pronounced than that of the notch setting. mean values of swath width obtained from various experiments were 12.74 m, 12.93 m, and 12.96 m when the angles of vanes were-10°, 0°, +10°, respectively (Fig. 5).

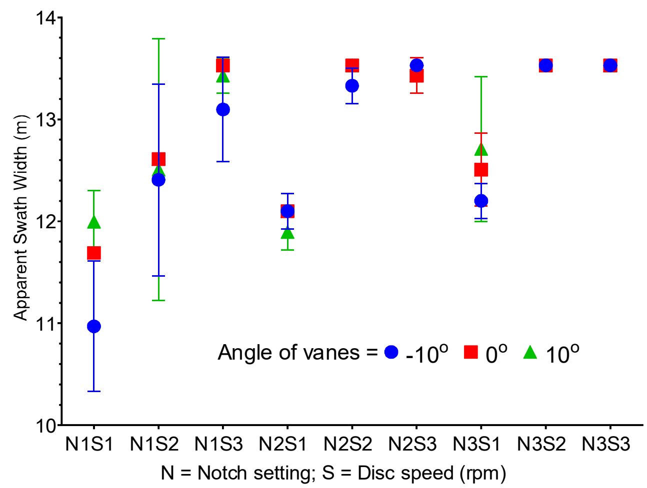
The effect of notch setting and disc speed on swath width were statistically significant, whereas that of the angle of vanes was not significant (Fig. 6). The interactions between disc speed and notch setting and that between notch setting and angle of vanes were also statistically significant. In contrast, the cumulative effect of disc speed and angle of vanes was non-significant. The combined interaction of these three parameters was also non-significant.

Effect of operational parameters on skewness of spread: The skewness of spread of the broadcaster varied with the



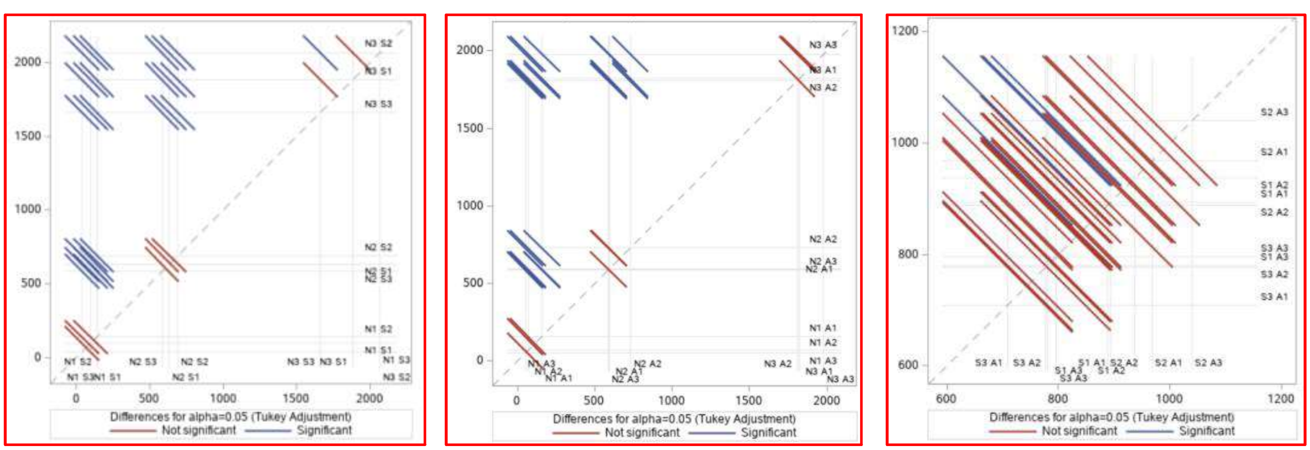
Error bars represent standard deviation about the mean in all graphs

Fig. 3. Effect of operational parameters on fertilizer rate (kg h^{-1})



Error bars represent standard deviation about the mean in all graphs

Fig. 5. Effect of operational parameters on apparent swath width (m)



a) Notch setting & Disc speed b) Notch setting and angle of vanes (c) Disc speed and angle of vanes

Fig. 4. Diffogram for the interaction of operational parameters on fertilizer rate (kg h^{-1})

level of notch setting (Fig. 7). As the notch settings were increased from n=2 to n=4, and thereafter to n=6, the mean skewness of spread values of the fertilizer broadcast were 0.29, 0.23, and 0.16, respectively. This decrease in the skewness of spread was noted irrespective of the angle of vanes and disc speed, and may be explained by the increased orifice size at every level, which enables a discrete flow, whereas a large expulsion area facilitates a uniform flow of the fertilizer. The skewness of spread of the tractor-operated fertilizer broadcaster also changes with the disc speed. The observed mean skewness of spread was 0.35, 0.11 and 0.22 at disc speeds of 300 rpm, 400 rpm, and 500 rpm, respectively (Fig. 7). However, the lowest coefficient of skewness of spread was at 400 rpm. Similar results have been reported by Sharabasy and Afify (2007) states that optimum value of disc speed at which the least value of skewness of spread was observed; and further increase or decrease in disc speed would distort the spread pattern of fertilizer. A slight variation was observed in the skewness of spread when the angle of vanes was altered. The observed mean skewness of spread was 0.23, 0.21, and 0.24 at angles of vanes -10°, 0° and +10°, respectively (Fig. 8). The most optimal distribution was at a 0° angle of fins. Similar has been observed by Yildirim (2006).

The effects of notch setting and disc speed on skewness of spread were significant, whereas that of angle of fins was non-significant (Fig. 8). The interactions between disc speed and notch setting, and angle of fins, and disc speed and angle of fins were also statistically significant. In addition, the cumulative interaction of the three parameters was significant. Kweon et al (2007) reported that increase in disc speed with higher quantity of fertilizer leads to reduced oscillation, since the particles are subjected to an increased force against the sidewalls of the vanes.

Coefficient of variation without overlapping: The least coefficient of variation without overlapping of swaths was 30.04% at N2S3A3 among all the combination of independent parameters (Fig. 9). The apparent swath width corresponding to this was13.53 m with fertilizer rate of 473.62 kg h^{-1}. At N1 notch setting (n = 2), the minimum value of coefficient of variation was 40.01% which comes at disc speed of 500 rpm and 0°angle of fins. The apparent swath width corresponding to variation was 13.53 m and fertilizer rate was 33.48 kg h^{-1}. At notch setting N3 (n=6), the minimum value of coefficient of variation was found to be 32.18% with disc speed of 500 rpm and +10° of angle of fins. The apparent swath width was calculated 13.53 with fertilizer rate of 1680 kg h^{-1}.

Coefficient of variation with overlapping: At notch setting 4 i.e., N2, the minimum value of coefficient of variation was

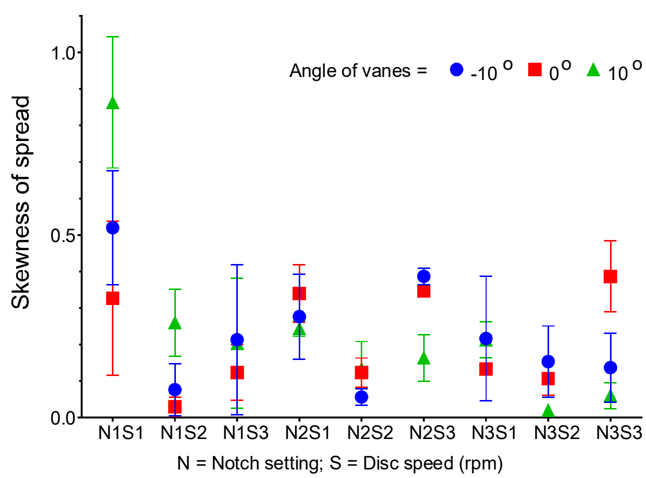
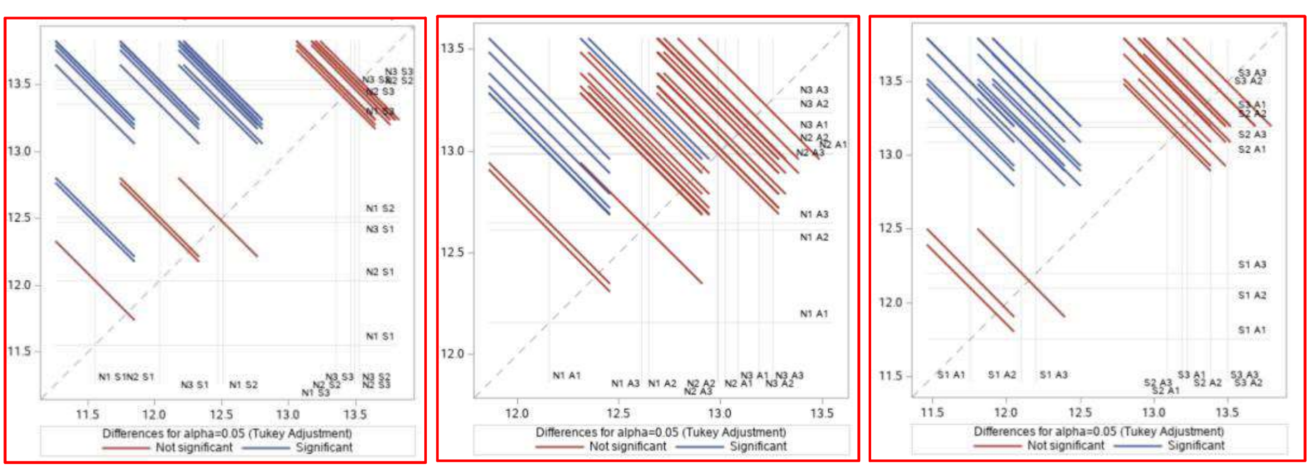
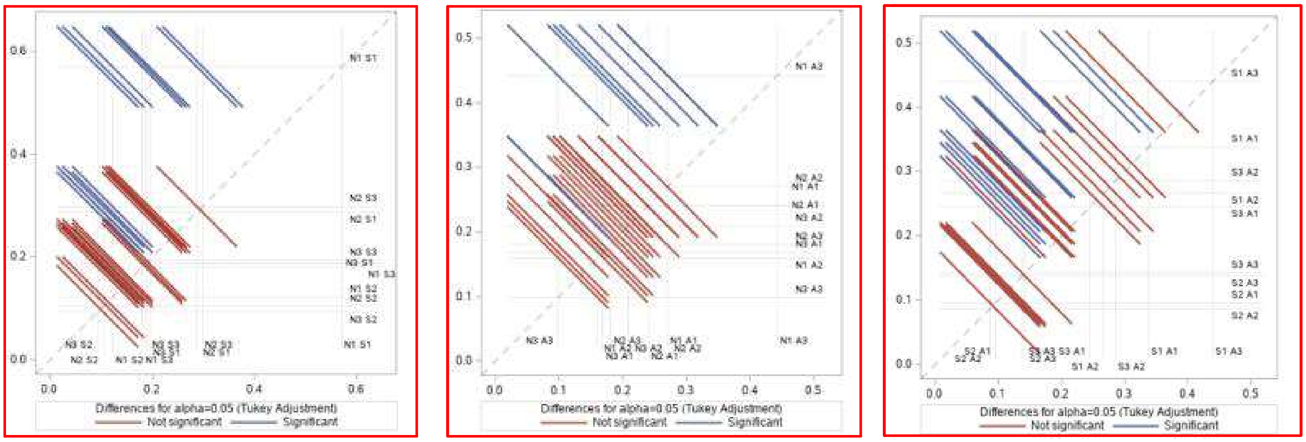


Fig. 7. Effect of operational parameters on skewness of spread



a) Notch setting & Disc speed b) Notch setting and angle of vanes (c) Disc speed and angle of vanes

Fig. 6. Diffogram for the interaction of operational parameters on apparent swath width



a) Notch setting & Disc speed b) Notch setting and angle of vanes (c) Disc speed and angle of vanes

Fig. 8. Diffogram for interaction of operational parameters on skewness of spread

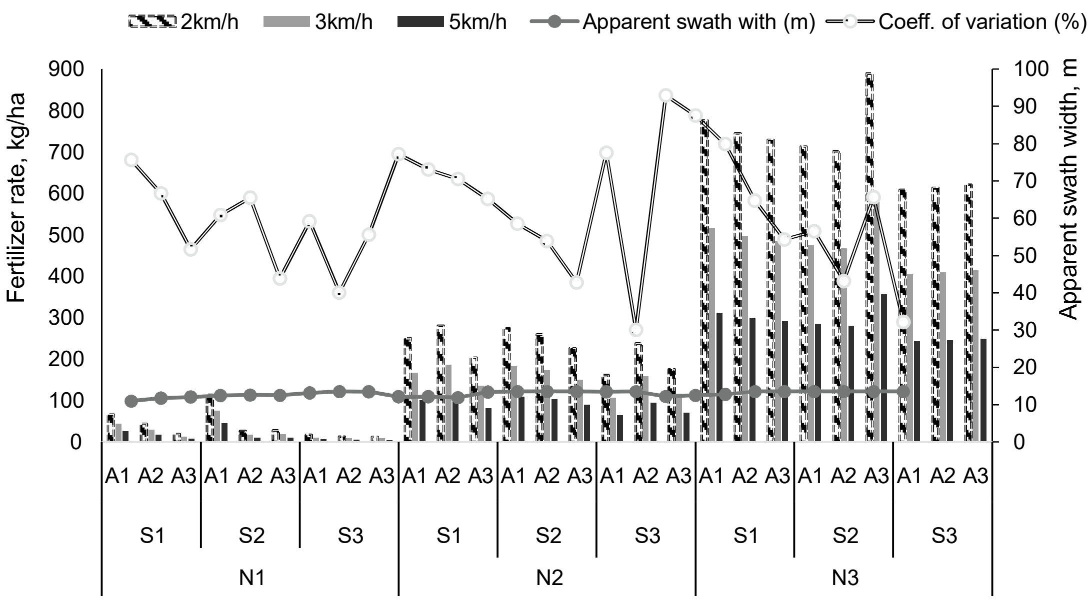


Fig. 9. Outcomes at different operational combinations without overlapping

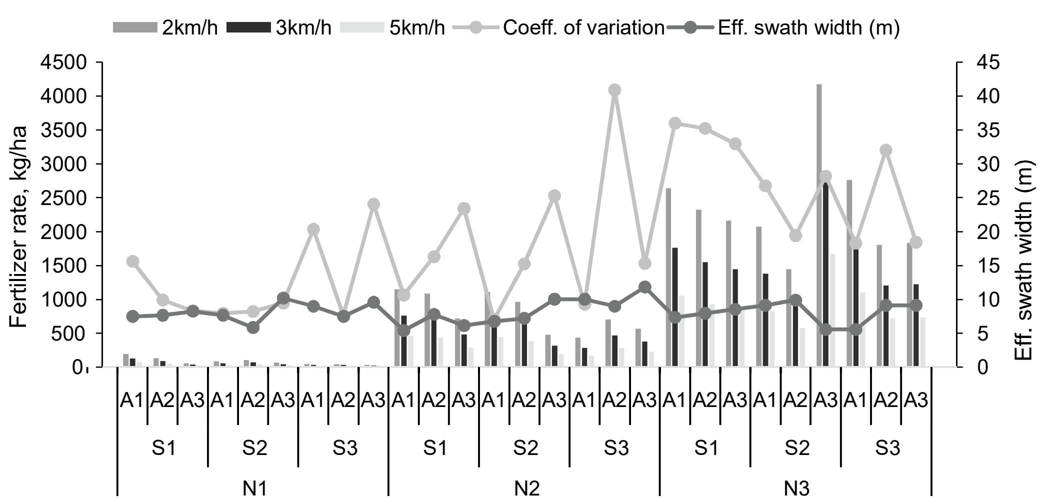


Fig. 10. Outcomes at different operational combinations with overlapping

7.05 at disc speed 400 rpm with -10° angle of vanes (Fig. 10). At notch setting 6 i.e., N3, the minimum coefficient of variation was 18.30 at disc speed 500 rpm with -10° angle of vanes. Overall, the minimum coefficient of variation was 7.05% at N2S2A1 after overlapping the swath width. At this setting, the fertilizer application rate was 25.01 kg/min and effective swath width was 6.75 m.

CONCLUSIONS

The fertilizer rate and apparent swath width increased with increase in level of notch setting (orifice opening area). The effect of notch setting and disc speed was statistically significant on fertilizer application rate. The skewness of variation decreased with increase in opening area of orifice. The maximum uniformity of spread was found at 0° angle of vanes. The coefficient of variation with apparent swath width varied from 32.18 to 92.92%. The best operational parameters from the study were notch setting 4 (opening area 9.40 cm^2), disc speed of 400 rpm and fin angle of -10^0.

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Histopathological Alterations in Major Organs of Freshwater Ornamental Goldfish, *Carassius auratus* (L.), Variety Shubunkin Reared in Inland Saline Water

Deepa Bhatt, Vaneet Inder Kaur*, Meera D. Ansal, Shanthanagouda A. Holeyappa[1] and Neelam Bansal[2]

Department of Aquaculture, College of Fisheries,[1]Department of Aquatic Environment, College of Fisheries
[1]Department of Veterinary Anatomy, College of Veterinary Science
Guru Angad Dev Veterinary Animal Science University, Ludhiana-141 004, India
**E-mail: vaneetinderkaur@gadvasu.in*

Abstract: The present study was carried out in glass aquaria (50 liter) for 120 days to assess histopathological alterations in major organs of freshwater ornamental goldfish, *Carassius auratus* (L.), variety Shubunkin reared in inland saline water in different salinity levels (2 to 10 ppt/‰) prepared from stock inland saline water (12 ‰). After proper conditioning, fishes were acclimatized (gradual increase in salinity @ 1 ‰ at 1-hr interval) to different salinity levels. After 4 months of salinity exposure in terms of histopathological alterations revealed deviation from normal responses/alterations beyond 4 ‰. The study presented alterations in gills, kidney and liver, which increased gradually from 2‰ up to 10‰. Although fish was capable of adapting and growing up to 6 ‰ in inland saline water, however, <4 ‰ can be considered safe with respect to overall performance of fish especially histopathological alterations. The results of present study suggest <4 ‰ as safe level to rear gold fish in inland saline water.

Keywords: Gold fish, Histopathological alterations, Inland saline water, Salinity stress

Among various abiotic factors, salinity is one of the critical parameters for overall well-being of freshwater fishes as it determines the level of osmoregulatory stress. Preliminary effect of salinity on fish physiology impacts the osmoregulation process where the ionic concentration of the body is maintained through intake or loss of ions by major organs such as gills, kidney and intestine (Al-Hilali and Al-Khshali 2016). The saline water tolerance in freshwater fishes vary between and within the species (Islam et al 2014). As freshwater fishes are stenohaline, growth decreases with the increase in salinity leading to homeostasis imbalance (Enayati et al 2013). Several reports are available on the rearing of freshwater ornamental fishes like gold fish, crucian carp and molly in natural or artificial saline water (Vasagam et al 2005, Schofield et al 2006, Küçük 2013). Schofield et al (2006) reported that goldfish can thrive in low saline environments (<10 ‰) for longer duration, and at higher salinities for short period. Their salinity tolerance is similar to that of *Cyprinus carpio*, but is higher and lower than that of *Hypophthalmicthys molitrix* and *Tilapia zillii*, respectively (Wang et al 1997). Due to its hardy nature, its culture in brackish water and marine environment is being taken up.

Hitherto, no report on the effects of Inland Saline Water (ISW) on histopathological studies of freshwater ornamental fish is available. Hence, optimizing the rearing technology of freshwater ornamental fish, goldfish *C. auratus* L. in North-Western states of India (inland saline or water-logged areas) is imperative. Further, it is also vital to study the stress response at different salinity levels in order to determine the optimal salinity tolerance for sustained culture practices which shall in turn, enhance the socio-economic status of the farmers as well. Moreover, the adaptability and tolerance of this species to physico-chemical changes in water vary greatly due to the differences in ionic composition of ISW to sea water (Dhawan et al 2010). With these limitations and opportunities, the present research was designed to study the adaptability of goldfish in ISW with special reference to histopathological responses.

MATERIAL AND METHODS

Experimental designs: A four-month experiment was carried out in glass aquaria (50 L capacity) at Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana to assess the histopathological alterations in gills, liver and kidney of *C. auratus* (L.) at different salinity levels of ISW (12‰) collected from village Shajrana, district Fazilka (Punjab). The experimental fish (Shubunkin) were procured from local market, and then conditioned for 15 days in indoor conditions. After proper conditioning, fishes (average length and weight - 8.04-8.25 cm and 6.30-6.80 g) were acclimatized gradually by increasing the salinity @ 1‰ per hour to 5 salinity levels (0, 2, 4, 6, 8 and 10‰) and were distributed (10 fishes per replicate) randomly in control and experimental salinity treatments (triplicates). All the experimental aquaria were provided with continuous oxygen supply and fishes were fed ad-libitum twice a day throughout the experimental period. Water quality parameters were analyzed with respect to temperature, pH, salinity, dissolved oxygen (DO), electrical conductivity (EC), total alkalinity (TA), total hardness (TH) and ammoniacal nitrogen (NH_3-N).

Histopathological studies: After completion of the experiment, histopathological studies were carried out from the control and salinity treatments. The targeted tissue (gills, kidney and liver) samples were fixed in 10% neutral buffered formalin (NBF; Cat #501128, Sigma Aldrich). They were washed in running tap water followed by alcohol dehydration, clearing with xylene and then embedding in paraffin wax. Thin sections (4 µm) were cut and mounted on gelatinized slides using a rotary microtome (Leica RM2125 RT, Germany). After this, the sections were mounted on microscope slides (Super Frost, #G10106, Abdos, India) and then dried at 33°C overnight. After observing the sections under a microscope (LEICA DM3000 LED), the good quality sections having no nicks were chosen for hematoxylin and eosin staining (Shanthanagouda et al 2014). The slides were mounted using Dibutylphthalate Polystyrene Xylene (DPX) mount (product code-23140, Molychem., Mumbai) and were examined. These slides were photographed under a light microscope (Nikon 80i) and captured using digital camera.

RESULTS AND DISCUSSION

During the experimental period, range of the water quality parameters *viz.* water temperature pH, DO, EC ,TA ,TH, NH_3-N was 14.2 - 30.5 °C, 7.14 - 8.99, 5.50 - 9.52 mg l^{-1}, 0.64 - 18.91 mS cm^{-1}, 232 - 336 $CaCO_3$ mg l^{-1}, 305 - 2930 $CaCO_3$ mg l^{-1} and 0.010 - 0.298 mgl^{-1}. During the present study, fishes were under chronic salinity stress for 120 days, resulting in prominent histopathological alterations in the major organs (gills, liver and kidney). Such alterations were occuring even at the lowest salinity treatment 2‰, which became severe with the increase in salinity to the extent of physiological breakdown (8‰ and 10‰).

The photomicrographs for the histopathology of gills showed remarkable changes with lamellar bending, thinning of inter lamellar region (Fig. 1 B, C, D), fused secondary lamellae, talengiactasia, hyperplasia, blood congestion and complete degeneration of secondary lamellae in highest salinities (Fig. 1 E, F). The alterations implicate the defense mechanisms (Fernandes and Mazon 2003; Akaishi et al 2004) and these alterations not only result in reduced secretary and excretory functions of the gills (Tilak et al 2006), but possibly lead to destruction of gill structure resulting in asphyxia (only 60 % fish survival in present study at highest salinity of 10‰). Further, the severity of gill alterations also indicated that the fishes were approaching tolerance limit to salinity resulting in osmoregulatory failure (Lawson and Alake 2011) causing mortality (6-10 ‰ in the present study).

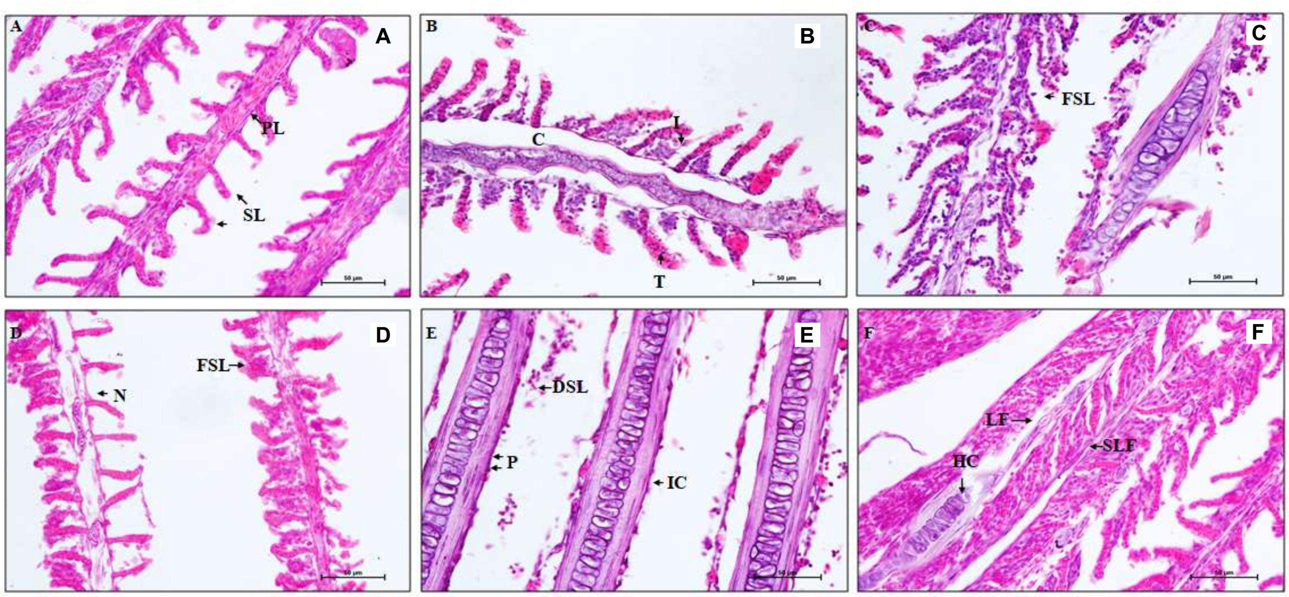

Fig. 1. Representative pictures of histomorphology of the gills of gold fish exposed to different salinity conditions for 120 days. A) 0‰ (freshwater) Primary lamellae (PL), Secondary lamellae (SL); B) 2 ‰ Infilteration (I), Telangiectasia (T), Chondrocytes (C); C) 4 ‰ Fused secondary lamellae (FSL); D) 6 ‰ Fused secondary lamellae (FSL), Necrosis (N); E) 8 ‰ Degenerated secondary lamellae (DSL), Increased Chondrocytes (IC), Perichondrium (P) and F) 10 ‰ Lamellar fusion (LF), Hyaline cartilage (HC), Secondary lamellar fusion (SLF). H & E stain. Scale bar= 50 µm

The photomicrographs for the kidney structures in control (Fig. 2 A) showed normal proximal and distal convoluted tubule structures, while the major histological alterations in kidney during increasing salinity conditions were observed in terms of degeneration of glomerular and distal convoluted tubules (Fig. 2 B, C). These alterations became severe

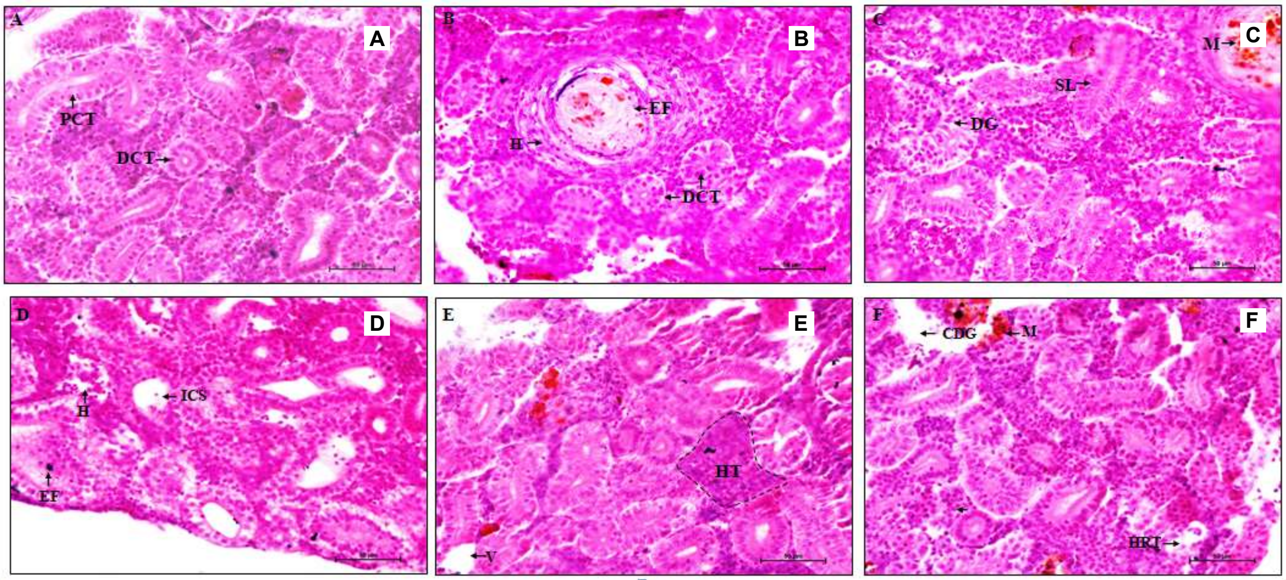

Fig. 2. Representative pictures of histomorphology of the kidney of gold fish exposed to different salinity conditions for 120 days. A) 0‰ (freshwater), Proximal convoluted tubule (PCT), Distal convoluted tubule (DCT); B) 2 ‰ Edmatous fluid (EF), Hyperplasia of tubules (H), Distal convoluted tubule (DCT); C) 4 ‰ Degeneration of glomerulus (DG), Shrunken lumen (SL), Melanomacrophage (M); D) 6 ‰, Edmatous fluid (EF), Haemorrhages (H), Increased capsular space filled with fluid (ICS); E) 8 ‰ Hematopoetic tissue (HT), Vacuolization (V) and F) 10 ‰ Complete degeneration of glomerulus and emptying of Bowman's capsule (CDG), Hypertrophy in renal tubules (HRT), Melanomacrophage (M). H & E stain. Scale bar= 50 µm

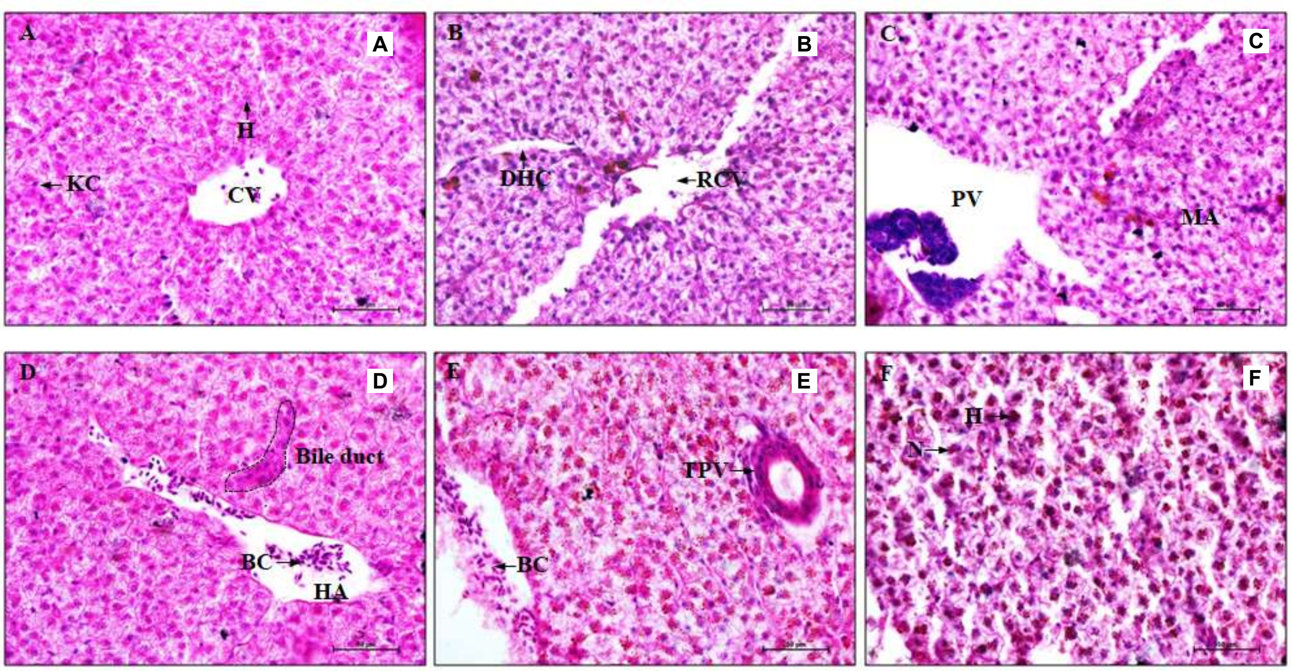

Fig. 3. Representative pictures of histomorphology of the liver in gold fish exposed to different salinity conditions for 120 days. A) 0‰ (freshwater) Hepatocytes (H), Central vein (CV), Kupffer cells (KC); B) 2 ‰ Rupturing of central vein (RCV), Disorganization of hepatic chords (DHC); C) 4 ‰ Melanomacrophage aggregates (MA), portal vein (PV); D) 6 ‰ Blood congestion (BC), Hepatic artery (HA); E) 8 ‰ Thinning of portal vein (TPV), Blood congestion (BC); F) 10 ‰ Haemorrhages (H), Necrosis (N). H & E Stain. Scale bar= 50 µm

leading to complete degeneration of glomerulus, emptying of Bowman's capsule (Fig. 2D, E), hypertrophy of renal tubules, haemorrhages and mononuclear cellular infiltration in higher salinity treatments (Fig. 2 F). Similar alterations were observed by Raskovic et al (2013) in common carp exposed to water having high pH and low DO (stream water), but were comparatively milder (Silva and Martinez 2007). Additionally, results in terms of shrinking and degeneration of glomerulus along with increased amount of edematous fluid were in accordance with the observations of Abdelhamid and El-Ayouty (1991) w.r.t. pollutant exposure/water quality alterations. In liver, the photomicrographs of control fish showed normal Kupffer cells, hepatocytes containing homogenous cytoplasm with a centrally placed nucleus and central vein (Fig. 3 A). Ruptured central vein and disorganization of hepatic chords were observed in 2‰ (Fig. 3 B). Further, there was an increase in the density of melanomacrophage aggregates in the parenchymal tissue of liver (Fig. 3 C) which indicated degenerative and necrotic processes (Pacheco and Santos 2002). In 6 and 8‰ hepatic artery, blood congestion and thinning of portal vein were observed (Fig. 3D, E). Hence, haemorrhages and necrosis at 10‰ can be explained as inhibition of DNA synthesis required for the growth and maturation of liver under abnormal conditions (Sanad et al 1997).

The most of the previous studies w.r.t. histological alterations in different organs of fishes were carried out to observe the effect of chemical, xenobiotics and pollutants or variations in water quality (especially temperature) (Velmurugan et al 2009, Hadi and Alwan 2012, Banaee et al 2013, Sharma and Tamot 2013, Drishya et al 2016, Sultana et al 2016). There is a lack of information regarding the effect of salinity on freshwater fish with special reference to histological alterations. Hence, the present study can be considered as the baseline for conduction of elaborative studies in near future.

CONCLUSION

The freshwater ornamental Shubunkin gold fish, *Carassius auratus* (L.) reared under different salinity regimes (0-10‰) depicted histopathological alterations w.r.t. gills, kidney and liver. The alterations started gradually from 2 ‰ onwards which became more severe at 10‰. Although fish was capable of adapting and growing under salinity conditions up to 6 ‰ in inland saline water, however, <4 ‰ salinity can be considered safe w.r.t. overall performance of the fish. Hence, freshwater ornamental Shubunkin gold fish, *Carassius auratus* (L.) can be reared for longer periods in inland saline water and it is further recommended to conduct field trials for better understanding of the effect of salinity under dynamic environmental conditions.

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Commensurable Analysis of Scientific Communications Published in Reviews in Aquaculture Applying Scientometric Analysis

Vaneet Inder Kaur, Nirmal Singh, H.S. Banga, Tarvinder Singh[1] and Gurpreet Singh[2]

Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana-141 004, India
[1]Indian Institute of Technology, Ropar-140 001, India
[2]Goswami Ganesh Dutt Sanatan Dharama College, Chandigarh-160 030, India
E-mail: vinnygulati@gmail.com

Abstract: This study presents a longitudinal and visualization mapping of scientific communications published in *Reviews in Aquaculture* during the period 2011-2020, applying scientometric approaches to depict the scientific contributions, collaboration trends, and research hotspots in the subject of aquaculture. Metadata of 412 articles published in *Reviews in Aquaculture* was retrieved and downloaded from Scopus database. The network graphs were visualized using '*VOSviewer*' and '*Gephi*' software. The chronological growth of scientific communications published in the journal, most productive authors, institutions, and countries vis-à-vis collaboration trends amongst them were scrutinized. The subject facets engulfed by the journal were identified based on co-occurrence of keywords. The findings would be useful for strengthening collaborative research as well as to pay required attention towards the slenderly explored sub-domains in aquaculture.

Keywords: Scientometrics, Aquaculture, Bibliometrics, Collaboration, Visualization

Despite scalable progress in society, hunger and malnutrition remain the major global issues in contemporary times. As per statistics of the Food and Agriculture Organization (FAO) of the United Nations (UN) for the year 2018, more than 11% of the world population suffers from poverty and lack access to quality food necessary for good human health (Kwasek et al 2020). Fish forms a valuable source of nutrients with special reference to protein in the human diet and it has been recorded that global consumption of aquatic food (excluding algae) has increased at an average annual rate of 3.0% from 1961 to 2019, a rate almost twice that of annual world population growth (1.6%) for the same period, with annual per capita consumption reaching to 20.5 kg in 2019 (FAO 2022). The global fish production was 177.8 million metric tonnes (mmt), including 90.3mmt from capture and 87.5mmt from aquaculture sector, and is expected to cross 200 mmt by 2030 (FAO 2022).

The contribution of aquaculture to the global production of aquatic animals reached a record 49.2% in 2020 along with considerable change in utilization and processing of fisheries and aquaculture production in the past decades. In 2020, 89% (157 mmt) of world production (excluding algae) was used for direct human consumption, compared to 67% in 1960. The reminder (over 20 mmt) was used for non-food purposes, mainly fishmeal and fish oil. Among these two major by-products, fishmeal is considered as highly nutritious for all the major livestock animals including farmed fish and fish oil represents the richest available source of long-chain polyunsaturated fatty acids (PUFAs), with a wide range of critical functions for human health (The Fish Site 2021). In addition to food value of fish and its by-products, fishing and aquaculture provide ample employment and livelihood to a large population viz. in 2020, about 58.5 million people were engaged in capture fisheries (38.03 million) and aquaculture (20.47 million) sector (FAO 2022).

After China, India is the second largest aquaculture producer in the world, with total fish and shell fish production of about 16.248 million metric tons (mmt) during 2021-22 with inland sector share of 12.121 mmt (74.60%) with export earnings of approximately Rs. 57,586.48 crores. Further, within inland fisheries, a major shift from capture fisheries to aquaculture has been witnessed in the last 3 decades and contribution of freshwater aquaculture has increased from 34% in mid 1980s to 78% in recent years. During 2020-21, fisheries contributed 1.10 % to National Gross Value Added (GVA) and 6.72% to the Agricultural GVA (Handbook on Fisheries Statistics 2022). Hence, identification of the prevalent scenario of scientific developments in aquaculture could be pivotal to further strengthen the occupation for uplifting it to a key role player in ensuring food security.

Nowadays, analysis of new trends and topics in various disciplines has attracted considerable attention in the academic and research spheres. Bibliometrics/ scientometrics has emerged as one of the widely used quantitative methods for understanding the changing landscape of concerned disciplines. Since journals are the primary sources of information carrying first-hand accounts of the research in the respective subject(s), scholars have analyzed the articles published therein for presenting the evolution and structure of the concerned subject domains. *Reviews in Aquaculture* is a pioneer journal enlisted amongst the high-impact journals, publishing reviews on developments in aquaculture techniques, policies, and planning. It takes in its ambit peer-reviewed review articles on the diverse aspects of aquaculture encompassing production and market trends, practices and technological developments, aquaculture-environment interactions, species in aquaculture; biology and culture of pivotal and emerging species, artificial propagation of species, feeds and feeding, genetics and aquaculture; health management in aquaculture; policy developments, product quality, and traceability and socio-economics aspects of aquaculture (https://onlinelibrary.wiley.com/journal/17535131). Scientometric analysis of *Reviews in Aquaculture* can help the academicians, researchers, and scholars to understand the strengths and gaps in aquaculture for determining their future course of action for boosting up the profession by paying attention to the research hotspots and for taking measures to fill the gaps, if any.

A few attempts have been made globally to study trends in fisheries and aquaculture applying scientometric analysis. Gasol and Durate (2000) conducted a comparative analysis in aquatic microbial ecology and referred to the analyses of general underlying trends to be useful in formulation of predictions to provide new avenues for research. To examine the impact of humans on fish habitats and the aquatic ecosystem from 1946 to 2014, Tao et al (2016) applied bibliometric analysis on the productive capacity of fish habitats (PCFH) and found that the research on PCFH is becoming a notable area of interest with research emphasis on fish-habitat relationship and fish production, aquaculture techniques and fishery products, habitat conservation and fishery management, and climate change. Qian et al (2018) conducted citation analysis on the *Journal of Fishery Sciences of China* during 2013-2016. Distribution analysis of keywords in the text brought forward that the keyword 'growth', 'gene cloning' and '*Litopenaeus vannamei'* had the most frequent appearances. During the bibliometric analysis of relevant research trends based on academic articles about the aquatic microbial community published during 1991 to 2018, Du et al (2020) found a strong correlation amongst the keywords 'bacteria', 'Denaturing Gradient Gel Electrophoresis', '16S rRNA', 'pyrosequencing' and 'sediment'.

However, none of the studies cited above applied network visualization for better revelations of hidden intricacies amongst the nodes (keywords, institutions, and countries) based on their co-occurrences. The exploration of authorship, institutional and geographical collaborations, identification of sub-domains of aquaculture dealt in by *Reviews in Aquaculture* journal based on co-occurrence of keywords, collaboration trends amongst authors, institutions, and countries using network visualization makes this study a first of its kind in the subject of aquaculture. This study aims to obtain the following objectives:

- To identify predominant authors, institutions, and countries contributing in *Reviews in Aquaculture* and their collaboration status through visualization maps
- To examine predominant subjects of aquaculture research published in *Reviews in Aquaculture* w. r. to author keywords

MATERIAL AND METHODS

Datasets: The bibliographical data about the articles published in Reviews in Aquaculture journal were accessed from *Scopus* for the period 2011-2020. The bibliographical parameters encompassing authors, title, year, source title, volume, issue number, page count, citation data, affiliations and author keywords were included while downloading data. A total of 412 records were downloaded in tab-delimited text (CSV) format and were used for scientometric analysis.

Subject analysis: The subject terms representing theme of records enlisted under the heading 'author keywords' field in the Scopus data were used for subject analysis (n=1490 terms) having one or more occurrences in records retrieved). Further, the singular and plural terms were standardized to avoid duplicity of appearance of terms.

Institutional productivity-cum-collaboration analysis: It was observed that in the original data file retrieved from *Scopus*, the institutional names were not listed uniformly viz. sometimes full name of an institution was given, at other instances only abbreviations were available. Moreover, sometimes name of the department(s)/college(s)/ section(s)/laboratory was also prefixed to the name of the institution. Hence, hurdles were faced in analyzing institutional contributions and collaborations. To overcome this problem, institutional names were examined individually and were standardized manually to bring uniformity for analyzing these through visualization software. The institution(s) with multiple campuses within a country were treated as a single institution(s) for analyzing institutional contributions and collaborations.

Data visualization: The scientometric aspects of records under study viz. prolific authors, institutions and countries, and collaborations amongst them vis-à-vis keywords based subject inferences were mapped using network visualization software '*VOSviewer*' (https://www.vosviewer.com/). Based on the co-occurrence of authors, institutions and countries within same records, respective clusters were developed using cluster schema of *VOSviewer*, each cluster representing inter-related nodes and each node in a network falling under one cluster only. The network visualizations developed have nodes and edges, nodes representing the variable being explored viz. authors, institutions, countries and subject terms and edges representing the links between nodes establishing their inter-connectivity. The thickness of edges indicates the strength of collaboration amongst authors, institutions, countries and co-occurrence of keywords and size of the nodes represents the number of records to which an author, institution and country has contributed and the frequency of occurrence of keywords in articles representing theme of the article(s).

RESULTS AND DISCUSSION

Chronological trends of publications in reviews in aquaculture: The number of records published in *Reviews in Aquaculture* from 2011 to 2020 witnessed significant growth, especially after 2017 (Table 1). The number of records published from 2011 to 2017 witnessed a nominal growth. Thereafter, an exponential growth was observed in publications in the journal, the year 2020 witnessing publishing of maximum number of records. On the other hand, advocating that the scientific communications are supposed to get more citations with the passage of time, the average citations earned by scientific communications were found to be higher in records published during the early years of decade. Year 2013 witnessed maximum number of citations viz. 65.95 citations per article on an average. The up-rise in number of articles and vice-versa for average citations are graphically represented in Figure 1.

Table 1. Number of records and their citation pattern

Year	No. of records	Total citations	Average citations
2011	11	511	46.45
2012	17	609	35.82
2013	22	1451	65.95
2014	18	609	33.83
2015	17	491	28.88
2016	19	615	32.37
2017	23	544	23.65
2018	56	1520	27.14
2019	75	960	12.80
2020	154	718	4.66

Most productive authors: All except 5.09% articles (21 single-author publications of a total of 412) have been the outcome of collaborative authorship. More than two-third of the records (69.42%; 286/412) have been an outcome of collaborative efforts of 2 to 5 authors and 21.60% (89/412) scientific communications were authored by 6 to 10 co-authors, each. A total of 16 articles have been authored in collaboration by more than 10 authors including an article having 28 co-authors in total. A total of 30 authors having contributed to at least four articles were identified and visualized using network visualization software (Fig. 2). As there are little chances that authors may have same surname and first name amongst identified 30 prolific authors, the authorship collaboration map has been developed based on the data processed using *VOSviewer* without any further cross examination of author names. Fourteen clusters, of which seven comprise of two to five authors were developed using cluster schema of *VOSviewer* based on co-occurrences of author names in records. The size of nodes represents the contribution of respective authors in terms of number of articles and edges show the strength of links between authors. Four clusters have 4-5 authors, and 3 have collaborations amongst two authors each. Six scientists from

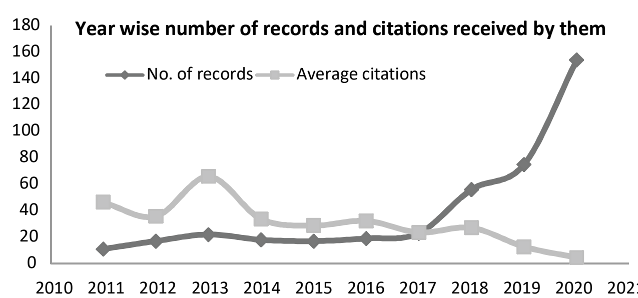
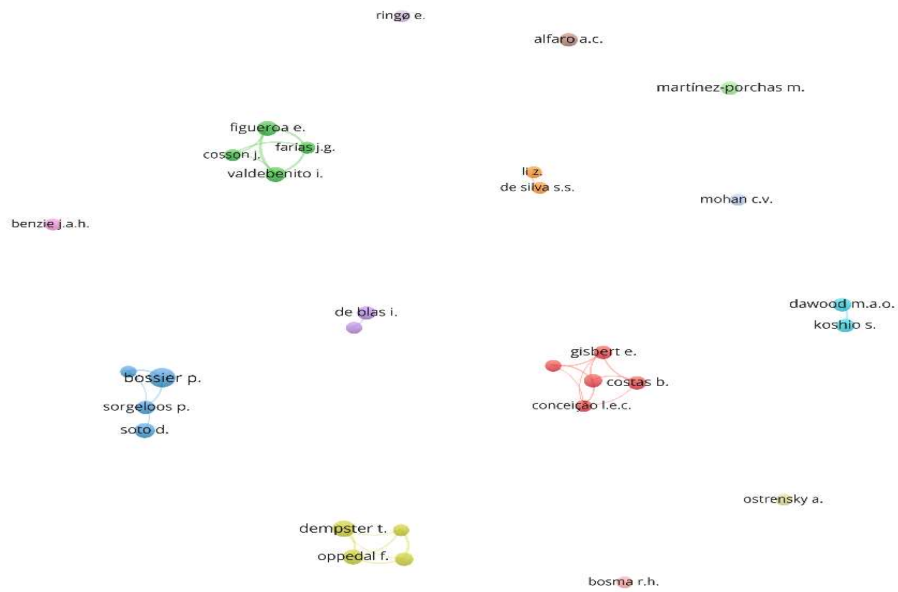


Fig. 1. Chronological growth of publications

Table 2. Top contributor institutions

Name of institution	Links	Total link strength	Documents
Ghent University, Belgium	8	14	18
Wageningen University and Research, Wageningen, Netherlands	9	13	17
Institute of Marine Research, Norway	9	14	15
University of South Bohemia, Czech Republic	1	2	12
University of Stirling, Stirling, United Kingdom	6	8	11



Cluster 1: Boglione C. (D-5; L-0; LS-0; C-869), Conceição L.E.C. (D-4; L-4; LS-5; C-425), Costas B. (D-5; L-3; LS-5; C-1165), Gisbert E. (D-5; L-4; LS-7; C-910) and Yúfera M. (D-5; L-4; LS-8; C-518)

Cluster 2: Cosson J. (D-4; L-3; LS-6; C-681), Farãas J.G. (D-4; L-3; LS-10; C-838), Figueroa E. (D-6; L-3; LS-12; C-1078) and Valdebenito I. (D-6; L-3; LS-12; C-1078)

Cluster 3: Bossier P. (D-9; L-2; LS-5; C-2149), Defoirdt T. (D-4; L-2; LS-4; C-399), Sorgeloos P. (D-5; L-3; LS-4; C-841) and Soto D. (D-6; L-1; LS-1; C-894)

Cluster 4: Dempster T. (D-7; L-3; LS-6; C-3066), Kristiansen T.S. (D-5; L-3; LS-7; C-1640), Oppedal F. (D-6; L-3; LS-8; C-2953) and Stien L.H. (D-4; L-3; LS-7; C-1369)

Cluster 5: Cabanillas-Ramos J. (D-4; L-1; LS-2; C-481) and De Blas I. (D-5; L-1; LS-2; C-1404)

Cluster 6: Dawood M.A.O. (D-5; L-1; LS-4; C-3537) and Koshio S. (D-5; L-1; LS-4; C-1040)

Cluster 7: De Silva S.S. (D-4; L-1; LS-2; C-428) and Li Z. (D-4; L-1; LS-2; C-2303)

Clusters 8-14: Alfaro A.C. (D-5; L-0; LS-0; C-869), Benzie J.A.H. (D-4; L-0; LS-0; C-832), Bosma R.H. (D-4; L-0; LS-0; C-482), Martãnez-Porchas M. (D-5; L-0; LS-0; C-1136), Mohan C.V. (D-4; L-0; LS-0; C-896), Ostrensky A. (D-4; L-0; LS-0; C-884) and Ringã¸ E. (D-4; L-0; LS-0; C-543)

Fig. 2. Most productive authors (D-number of articles, L-number of links, LS-Total Link Strength, C-Citations)

Table 3. Top contributing scientists

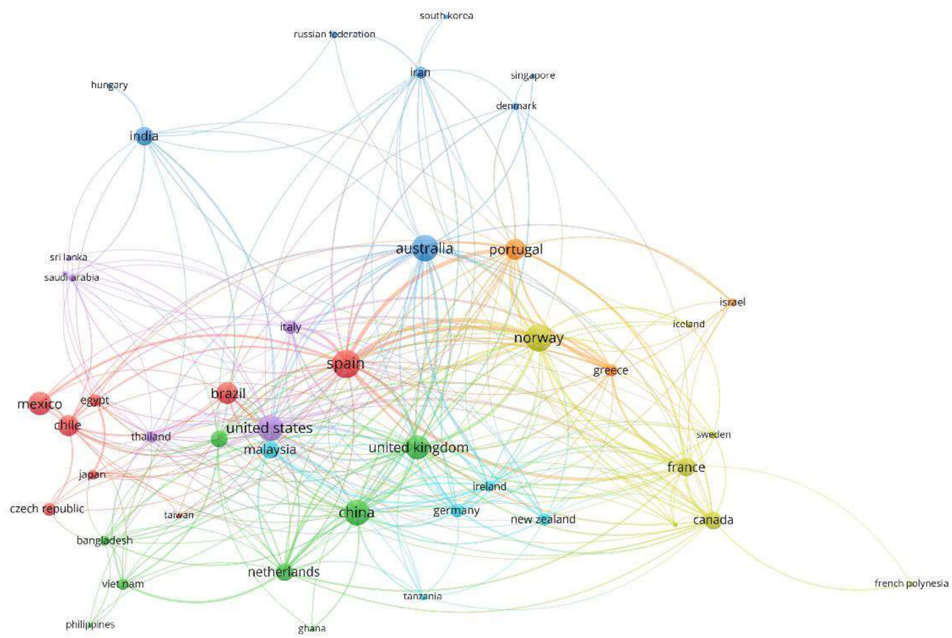
Name of scientist	Institute	Area of expertise/ specialization
Professor (Dr.) Peter Bossier	Director, Laboratory of Aquaculture and Artemia Reference Center, Ghent University, Belgium	Bioscience Engineer - Microbial community management, host-microbial interactions, and genetics
Dr. Tim Dempster	School of Bio-Sciences, University of Melbourne, Parksville, Australia	Ecological Research - Fishing, aquaculture and other anthropogenic practices
Dr. Elias Gustavo Figueroa	School of Aquaculture, Catholic University of Temuco, Temuco, Chile	Biotechnology- Optimization of Reproduction in Chilean Aquaculture
Dr. Oppedal F	Institute of Marine Research, Matre Research Station, Matredal, Norway	Aquaculture Management – Salmon Welfare
Dr. Doris Soto	Interdisciplinary Center for Aquaculture Research (INCAR), Universidad de Concepcion Chile, Concepcion, Chile	Aquaculture, Environmental Impact, and Aquatic Biodiversity
Dr. Valdebenito	School of Aquaculture, Catholic University of Temuco, Temuco, Chile	Reproductive factors in fish of aquaculture interest

Most Influential Authors in terms of Citations– Dawood M.A.O.,Dempster T, Oppedal F., Li Z., Bossier P., Kristiansen T.S., De Blas I., Stien L.H., Costas B. and MartÃnez-porchas M.

4 countries had contributed highest number of articles, whereas ten scientists found to be the most influential authors in terms of citations earned by their articles published in Review in Aquaculture (Table 3).

Major contributing countries: Authors from a total of 62 countries contributed to 412 records. Twenty three nation(s) from the Europe continent collectively contributed to the highest number of records (n=324), followed by contribution of 174 records from 19 Asian nations and 102 records from 4 North America countries. Five South American countries contributed to 66 records. Two Australasia countries added to 57 records and 9 African nations contributed to 26 records (Fig. 3). Two countries (Poland and Nigeria) of the total 62 countries having contributed records to *Reviews in Aquaculture* did not have any international collaboration. Twenty two nation(s) from the Europe, 19 Asian nations, 4 North America countries, 5 South American countries, 2 Australian countries and 8 African nations worked in international collaboration contributing articles for *Reviews in Aquaculture*, depicting the joint efforts at global level to boost aquaculture sector. The network visualization map of the countries having contributed at least three articles published in *Reviews in Aquaculture* was developed. Out of the resulting 46 countries, Poland did not have any article in international collaboration. Hence, it was excluded from network visualization. The top 45 countries with at least three articles published in the journal under study are shown in Figure 3.

Institutional productivity: Authors from 627 institutions/ organizations contributed to the total of 412 records. Thirty nine institutions contributed to 5 or more articles each, of which 6 institutions did not have inter-institutional



Cluster 1	Brazil (D-32; LS-21; C-6732), Chile (D-29; LS-27; C-5477), Czech Republic (D-13; LS-7; C-3166), Egypt (D-12; LS-13; C-6465), Japan (D-8; LS-13; C-2027), Mexico (D-36; LS-13; C-8465), Spain (D-49; LS-100; C-9982) and Taiwan (D-3; LS-5; C-764)
Cluster 2	Bangladesh (D-8; LS-19; C-2480), Belgium (D-21; LS-40; C-4025), China (D-43; LS-57; C-23546), Ghana (D-3; LS-4; C-496), Netherlands (D-23; LS-48; C-3884), Philippines (D-3; LS-4; C-1430), United Kingdom (D-37; LS-75; C-8762) and Viet Nam (D-10; LS-19; C-1390)
Cluster 3	Australia (D-45; LS-62; C-18365), Denmark (D-5; LS-11; C-1085), Hungary (D-3; LS-2; C-344), India (D-25; LS-18; C-7332), Iran (D-12; LS-20; C-2913), Russian Federation (D-5; LS-4; C-3363), Singapore (D-3; LS-3; C-734) and South Korea (D-3; LS-2; C-505)
Cluster 4	Canada (D-22; LS-53; C-5712), France (D-24; LS-67; C-6150), French Polynesia (D-3; LS-3; C-1201), Iceland (D-4; LS-10; C-1094), Monaco (D-3; LS-10; C-752), Norway (D-46; LS-106; C-12400) and Sweden (D-5; LS-17; C-1120)
Cluster 5	Colombia (D-3; LS-8; C-1232), Italy (D-15; LS-52; C-3699), Saudi Arabia (D-5; LS-13; C-1567), Sri Lanka (D-3; LS-8; C-690), Thailand (D-10; LS-30; C-2056) and United States (D-43; LS-70; C-11094)
Cluster 6	Germany (D-14; LS-46; C-3715), Ireland (D-9; LS-33; C-2868), Malaysia (D-23; LS-35; C-6815), New Zealand (D-12; LS-20; C-2923) and Tanzania (D-4; LS-10; C-1142)
Cluster 7	Greece (D-12; LS-46; C-2140); Israel (D-6; LS-10; C-4053) and Portugal (D-30; LS-66; C-5469)

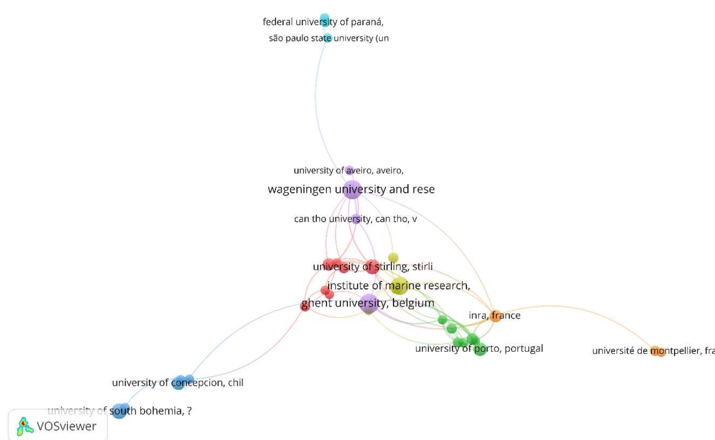
Fig. 3. Most productive countries (D-number of articles, L-number of links, LS-Total Link Strength, C-Citations)

collaboration. Hence, collaboration visualization map of 34 institutions having worked in collaboration with one or more institutions (that contributed to >5 records each) is represented by Figure 4. Scientometric data (number of articles, number of links, link strength) of the top 5 institutions (10 records in collaboration) is also tabulated (Table 2). It has been observed that the academic and research institutions from European countries have contributed predominantly towards the *Reviews in Aquaculture*, having presence in 6 of the total 7 cluster. Cluster 1 represents the collaboration amongst Australia, European and Asian countries. Cluster 2 and 3 depicts the collaborative efforts amongst European nations.

Subject analysis based on keywords: The top 45 keywords (out of the total 1490) having incurred in 5 or more articles were considered for cluster analysis (Fig. 5). A total of eight clusters were identified by clustering schema of *VOSviewer* software. The nodes in figure represent the keywords/ subject terms and edges reveal the relation between different keywords representing subject(s). Subject analysis reveals that the focus of most of the institutes is on sustainable development of fisheries and aquaculture through feed and health management strategies through omics approach. Further, the key word based analysis clearly indicated that intensification of aquaculture practices; environmental impact and resilient measures thereof are also being considered. Cluster analysis indicated that research emphasis is also given on major aquaculture fish (Tilapia and Salmon) and shellfish (Vannamei) species of high economic value at global level.

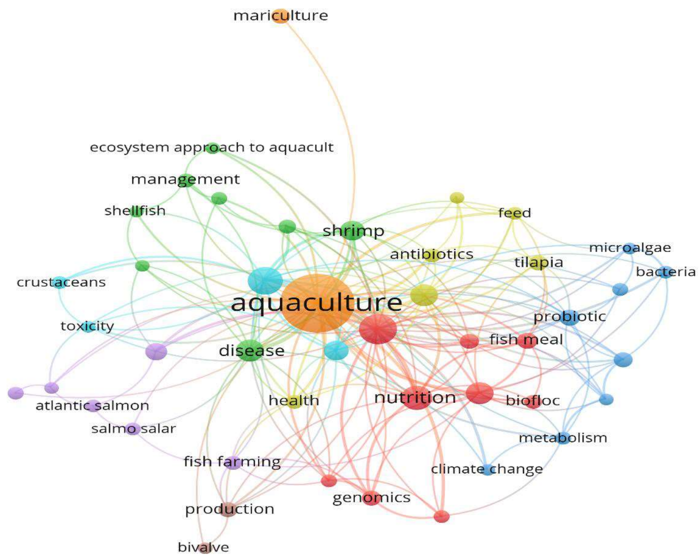
CONCLUSIONS

The study presented trends and hotspots in aquaculture based on scientometric analysis and network visualization of metadata of 412 records published in *Reviews in Aquaculture* retrieved from Scopus database. The distribution of number of records published in *Reviews in Aquaculture* has been found uneven. From the year 2018 onwards, the publication trends of the journal witnessed exponential growth. This could be attributed to the recognition of growing significance of aquaculture globally as correspondingly the scientific outcome on the subject is also growing. *Reviews in Aquaculture* has witnessed contribution of articles from around the globe. A strong international collaboration trend



Cluster 1	CSIRO, Australia; Deakin University, Australia; Shanghai Ocean University, Shanghai, China; Universiti Putra Malaysia, Malaysia; University College Cork, Cork, Ireland; University of Stirling, Stirling, United Kingdom and Worldfish, Penang, Malaysia
Cluster 2	Instituto de Ciencias Marinas de Andalucía, Cadiz, Spain; IRTA, Sant Carles de la Rápita, Spain; University of Algarve, Faro, Portugal; University of Bergen, Bergen, Norway; University of Crete, Greece; University of Porto, Portugal and University of the Algarve, Portugal
Cluster 3	Interdisciplinary Center for Aquaculture Research, Chile; University of Chile, Chile; University of Concepcion, Chile; University of La Frontera, Temuco, Chile and University of South Bohemia, Czech Republic
Cluster 4	Institute of Marine Research, Norway; Norwegian Institute of Food, Fisheries and Aquaculture Research, Norway; University of Melbourne, Australia and University of Tasmania, Australia
Cluster 5	Can Tho University, Can Tho, Viet Nam; Ghent University, Belgium; University of Aveiro, Aveiro, Portugal and Wageningen University and Research, Wageningen, Netherlands
Cluster 6	Federal University of Parana, Brazil; Shantou University, Shantou, China and São Paulo State University (UNESP), Jaboticabal, Brazil
Cluster 7	INRA, France; Université De Montpellier, France and Université Laval, Québec, Canada

Fig. 4. Collaboration map of most productive institutions (n=33) having inter-institutional collaboration and scientometric details of institutions (n=5) that contributed 10 records



Cluster 1	Aquafeed; Biofloc; Fish meal; Genomics; Growth; Metabolomics; Nutrition; Stress and sustainability
Cluster 2	Biodiversity; Disease; Ecosystem approach to aquaculture; *Litopenaeus vannamei*; Management; Meta-analysis; Shellfish and shrimp
Cluster 3	Bacteria; Bioremediation; Climate change; Disease resistance; Immunostimulant; Metabolism; Microalgae and Probiotic
Cluster 4	Antibiotics; Feed; Health; Immunity; Oxidative stress and Tilapia
Cluster 5	Atlantic salmon; Environmental impact; Fish farming; Gene expression; Salmo salar; Spermatozoa
Cluster 6	Crustaceans; Environment; Fish and Toxicity
Cluster 7	Aquaculture and Mariculture
Cluster 8	Bivalve and Production

Fig. 5. Keyword visualization map (O-Occurrences, L-Links, LS-Total Link Strength)

was observed amongst nations as except Poland and Nigeria, all other 60 countries have international collaborations for one or more articles. European nations have contributed maximum number of records and consequently, institutions representing Europe emerged as predominant contributors of articles to *Reviews in Aquaculture*. Moreover, the top five countries having contributed highest number of articles also falls under the European Union with the focus on aquaculture practices with management strategies *vis a vis* environmental impacts. Diverse facets of the subject aquaculture are being explored globally for strengthening of the sector in view of its growing and widening significance towards attainment of sustainable food security.

AUTHOR CONTRIBUTION

Vaneet Inder Kaur - Manuscript writing and critical inputs w.r.to subject matter related discussion; Nirmal Singh - Conceived the idea, manuscript writing and data analysis with software; Harmanjit Singh Banga - Manuscript writing and suggestions for data analysis; Tarvinder Singh Handa - Data retrieval from Scopus and preparation of data for analysis; Gurpreet Singh - Manuscript writing w.r.t. scientometric aspects.

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Synergistic Effect of Prebiotic with Gut Isolated Probiotic Bacteria on Survival, Growth and Carcass Composition of *Labeo rohita*

Prem Kumar, Vaneet I. Kaur[1], Anuj Tyagi[1] and Sachin Khairnar[1]

Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu, Jammu-180 009, India
[1]College of Fisheries, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana -141 004, India
E-mail: fish9897@gmail.com

***Abstract*:** Feeding trial (120 days) was conducted to evaluate the synergistic effects of dietary supplementation of prebiotic and probiotic bacteria (*Lactobacillus plantarum* FLB1), isolated from the intestine of Indian Major Carp (*Labeo rohita*) on the survival, growth and carcass composition of *L. rohita* fingerlings. Six experimental diets were formulated containing basal diet as control (T1) and prebiotics, i.e. β-glucan (BG) and Mannan oligosaccharide (MOS) were added in basal diet (T1) at 2 different levels (T2 @ 0.2% BG + 10^8 *L. plantarum*, T3 @ 0.5% BG + 10^8*L. plantarum*, T4 @ 0.2% MOS+10^8*L. plantarum*, T5 @ 0.5% MOS + 10^8*L. plantarum*, and T6 @ 10^8*L. plantarum)* and fed to the fish for a period of 120 days @ 5% body weight in two parts, daily. Hematological parameters were observed at 30, 60 and 120 days intervals while growth parameters at 30, 60, 90 and 120 days intervals. Total body length (cm) and fish body weight (g) were significantly higher in T3. Average hemoglobin content, total erythrocyte count, total leukocyte count, hematocrit and erythrocyte sedimentation rate of fishes fed with T3 diet were significantly improved. Mean corpuscular volume, corpuscular hemoglobin and corpuscular hemoglobin content revealed positive effect in pre- and pro-biotic fed fishes. There was significant improvement in the carcass composition in T3 in terms of highest flesh protein as compared to other treatments and control.

***Keywords*:** Prebiotics, Probiotics, β-glucan (BG), Mannan oligosaccharide (MOS), Synergistic effect

In India, inland fish production has crossed 16.24 million tons during the year 2021 contributing over 76% to the total fish production (pib.gov.in 2023). However, disease outbreak is a major hurdle in the intensification and harnessing of full potential in carp farming. Overuse of antibiotics and therapeutics is leading to negative impacts like development of antimicrobial and drug resistance, accumulation of chemical residues in tissue and reduced consumer preferences. As a therapeutic measure in aquaculture (tanks, ponds or cages), antibiotics (mixed with specially formulated medicated feed) are orally administered to fishes for shorter duration. However, fishes do not effectively metabolize those antibiotics and therefore, a large portion (75%) is released back into the environment in the form of feces (Burridge et al 2010). Further, the quality and quantity of antibiotics and other related compounds used in aquaculture differed significantly between countries. Hence, the rise in bacterial antibiotic resistance and residues have become a global concern which calls for the need to develop alternative therapies for controlling bacterial pathogens in animal production, especially in aquaculture. Although vaccines are being developed and marketed to address this problem, however, these cannot be a universal disease control measure in aquaculture (Lara-Flores 2011).

Among alternative therapies, a variety of useful feed additives like medicinal herbs, immunostimulants, probiotics and prebiotics having beneficial effects on the host are being used in aquaculture. Certain non-digestible carbohydrates *viz.* polydextrose, lactosucrose, resistant inulin and oligofructose, transgalacto oligosaccharides (TOS), isomalto-oligosaccharides (IMO), palatinos, xylooligosaccharides (XOS), mannan oligosaccharides (MOS), lactose, hemicellulose, soybean oligosaccharides, glucooligosaccharides (GOS), gluconic acid and β-glucan display authentic prebiotic properties (Verkhnyatskaya et al 2019). The oligosaccharides prebiotics are reported to reduce β-glucuronidase and nitroreductase activities resulting in the enhancement of immunity, modulation of mucin production and expression of immune regulatory genes (Arturo et al 2010). This study was carried out to investigate the effect of different doses of prebiotic compounds combined with fish gut probiotic on rohu, *Labeo rohita*.

MATERIAL AND METHODS

Preparation and maintenance of experimental tanks: The study was conducted in outdoor cemented tanks ($20m^2$) where 3-5cm soil bottom layering was done to stimulate natural conditions and liming @ 300kg/ha for disinfection and as per requirement (pH balance) throughout the experimental period. Water exchange (1/4th of water) was performed with freshwater once a week.

Procurement, conditioning and stocking of experimental fish: Fingerlings of rohu (720 nos.) distributed randomly in all experimental tanks (40/tank) in triplicates. Proximate analysis of the feed ingredients, formulated feeds and fish flesh, water quality parameters, microbiological, hematological, and biochemical parameters studies were carried out. Prebiotics β-glucan (BG) and Mannan oligosaccharide (MOS) were added in basal diet (Table 1). The bacterial count of *L. plantarum* mixed in feed adjusted through spectrophotometer was @ 10^8 cfu/g. To achieve accurate final concentrations in the diet, the bacterial suspension was gradually added to feed pellets with a hand sprayer for uniform distribution in the laminar airflow chamber under sterilized conditions. The fishes were fed with experimental diet for a period of 120 days @ 5% body weight daily in two parts.

Water quality parameters: Water samples were collected fortnightly in the morning hours for analyzing the physico-chemical parameters (APHA 2013) *viz*. temperature, pH, dissolve oxygen (DO), total alkalinity (TA), total hardness (TH), orthophosphate (PO_4^{-2}) and ammoniacal nitrogen (NH_3-N).

Blood collection: Prior to collection of blood, fishes were anesthetized with clove oil @ 30-50 mg/l (1-part clove oil and 9-parts 94% ethanol) (Hajek et al 2006) and blood were collected via caudal vein puncture and then pooled from a random sample consisting of five fish from each replicate. Hematological parameters were observed at 30, 60 and 120 days intervals while growth parameters at 30, 60, 90 and 120 days. Blood (heparinized 150 IU/ml) collected from each group were analyzed for the hematological parameters *viz.* hemoglobin (Hb), total erythrocyte count (TEC), total leukocyte count (TLC), hematocrit (Ht) and erythrocyte sedimentation rate (ESR). The mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin content (MCHC) were calculated by the method of Mukherjee (1988). Hemoglobin (Hb) concentration was estimated by acid haematin method (Sahli 1962).

Growth of fish: The growth of fish was assessed in terms of total body length and weight at monthly intervals during 120 days of experimental period. At the end of the experiment, growth (length and weight) parameters in terms of total length gain (TLG), percent total length gain (%TLG), net weight gain (NWG), percent net weight gain (%NWG), specific growth rate (SGR), growth index (GI), condition factor (K) and feeding efficiency in terms of protein efficiency ratio (PER), feed conversion ratio (FCR) and feed conversion efficiency (FCE) for each treatment was calculated.

Flesh composition: Carcass composition was assessed at the end of experiment. Flesh quality (% wet weight basis) in terms of total proteins, total lipids, total carbohydrates, ash content and moisture were estimated for each treatment and control.

Statistical analysis: The differences between parameters were analyzed by using SPSS software 16 version.

RESULTS AND DISCUSSION

Physico-chemical parameters of water: Physico-chemical parameters were under optimal range (Boyd 1988) with insignificant differences among the treatments (Table 2).

Growth of fish: Survival rate was 100% in all the treatments along with control. The total body length gain (cm) was significantly higher in T3 (9.95) followed by T5 with non-significant differences (Table 3). The body length gain was significantly low in T1 (7.43). The total body net weight gain (g) was maximum in T3 (59.73) with % net weight gain of 581.86 followed by T5 and minimum in T1 (33.34). The specific growth rate was also significantly higher in T3 (1.60) followed by T5 (1.50) and minimum in T1 (1.19) (Table 4). The growth Index depicted the same pattern with highest value in T3 followed by T5.

In all the treatments, 100 % survival rate at the end of experiment indicated no harmful effect of prebiotics (β-glucan and MOS) and probiotic (*L. Plantarum*) supplemented diets. The length and growth parameters revealed improved fish growth performances in synbiotic and probiotic incorporated diets (T2-T6), with significant improvement in T3 (0.5% β-glucan + 10^8 of *L. plantarum/* g feed*)* and T5 (0.5% MOS + 10^8 LAB) as compared to that of control. No information is available on synergistic effect of β-glucan and *L. plantarum* or MOS and *L. plantarum* on rohu, however, Giri

Table 1. Details of treatment for experiment

Treatments					
Control diet	Basal diet supplemented with gut probiotic bacterial (GPB) culture				
T_1	T_2	T_3	T_4	T_5	T_6
Basal diet*	Basal diet + 0.2% BG + 10^8 *L. plantarum*	Basal diet + 0.5% BG + 10^8 *L. plantarum*	Basal diet + 0.2% MOS+10^8*L. plantarum*	Basal diet + 0.5% MOS+10^8*L. plantarum*	Basal diet + 10^8 *L. plantarum*

*Rice bran[1] (49%) + Mustard meal[1] (49%) + Vit-Min. mixture (1.5 %) + Salt (0.5%)
[1]De-oiled rice bran, β-glucan (BG) and MOS (Mannan Oligosaccharide)

et al (2013) reported higher specific growth rate and feed utilization efficiency in the juveniles of *L. rohita* when fed with *L. plantarum* only. The positive interactive effects of heat-killed *L. plantarum* (HK-LP) and β-glucan (BG) on body weight gain, specific growth rate, feed intake and protein efficiency ratio was reported by Dawood et al (2015) in juvenile red sea bream, *Pagrus major*. Probably, β-glucan was degraded in the digestive gland by glucanases to produce energy, hence, permitting the use of more proteins for growth. Mishra et al (2006 a,b) reported increased SGR and reduced FCR in *L. rohita* when β-glucan was supplemented in diets as immunostimulant or through direct injection.

Feeding efficiency: Feeding efficiency of prebiotic and probiotic supplemented feed was observed in terms of FCR, FCE and PER (Table 5). FCR was significantly lower in T3

Table 2. Mean physico-chemical parameters of water in different treatments

Parameters	Treatments					
	T1	T2	T3	T4	T5	T6
Temperature (^{o}C)	30.64±0.31	30.78±0.32	30.79±0.32	30.81±0.33	30.73±0.31	30.70±0.32
pH	7.73±0.04	7.74±0.03	7.80±0.03	7.76±0.03	7.78±0.03	7.71±0.03
DO (mg/l)	6.47±0.04	6.50±0.03	6.46 ±0.06	6.44 ±0.06	6.55±0.04	6.40±0.05
TA ($CaCO_3$ mg/l)	154.52±1.73	161.63±2.05	156.96±2.10	160.26±2.34	160.07±1.86	164.33±1.87
TH ($CaCO_3$ mg/l)	245.04±2.75	241.96 ±3.03	243.15±2.59	242.30±2.71	239.19±3.04	243.48±2.36
NH_3-N (mg/l)	0.04±0.002	0.04±0.002	0.04±0.002	0.04±0.003	0.04±0.002	0.04±0.002
Orthophosphate (mg/l)	0.07±0.002	0.07±0.001	0.08±0.002	0.07±0.002	0.08±0.001	0.07±0.001
NO_3^-N (mg/l)	0.30±0.01	0.32±0.01	0.31±0.01	0.32±0.01	0.30±0.01	0.29±0.01

See Table 1 for treatment details. Values are Mean ± S.E. (p≤0.05)
DO – Dissolved oxygen, TA – Total Alkalinity, TH – Total hardness

Table 3. Length parameters of rohu, *Labeo rohita* (Ham.) during the experimental period

Month	Days	Treatments					
		T1	T2	T3	T4	T5	T6
May	0	8.70±0.15	9.00±0.06	8.78±0.10	8.80±0.06	8.67±0.11	8.79±0.14
	30	9.87±0.16	10.73±0.38	10.63 ±0.45	10.76±0.49	9.61 ±0.22	10.91±0.15
June	60	11.37±0.07	12.35±0.27	11.94±0.22	12.25±0.33	11.39±0.32	12.64±0.43
July	90	13.23±0.15	14.12±0.19	14.96±0.34	13.91±0.11	14.29±0.06	14.45±0.20
August	120	16.13±0.15	17.25±0.41	18.73±0.12	17.04±0.15	18.21±0.12	17.01±0.15
Total length gain		7.43±0.20	8.25±0.43	9.95±0.06	8.24±0.12	9.54±0.04	8.22±0.18
Length gain %		85.55±3.49	91.67±5.12	113.44±3.42	93.64±1.38	110.02±1.39	93.64±3.23

See Table 1 for treatment details

Table 4. Weight parameters of rohu, *Labeo rohita* (Ham.)in different treatments during the experimental period

Month	Days	Treatments					
		T1	T2	T3	T4	T5	T6
May	0	10.50±0.15	10.47±0.18	10.27±0.07	10.38±0.22	10.47±0.18	10.33±0.18
	30	16.23±0.29	19.63±0.75	26.53±0.64	19.51±0.54	21.73±0.48	19.10±0.53
June	60	24.30±0.23	28.53±0.65	35.55±1.86	28.63±0.77	31.53±0.58	28.37±0.74
July	90	35.43±0.61	41.00±0.40	46.03±1.23	40.94±0.64	42.27±0.24	39.98±0.23
August	120	43.84±1.97	56.60±4.20	70.00±0.51	52.16±0.52	63.05±1.32	50.51±0.11
Net weight gain		33.34±2.05	46.14±4.16	59.73±0.53	41.78±0.54	52.58±1.27	40.18±0.16
% NWG		317.96±22.38	440.74±39.31	581.86±7.35	402.96±11.48	502.60±13.29	389.11±8.01
Specific growth rate		1.19±0.05	1.40±0.06	1.60±0.01	1.35±0.02	1.50±0.02	1.32±0.01
Growth index		2.18±0.22	3.41±0.39	4.82±0.07	3.03±0.11	4.03±0.13	2.89±0.08

See Table 1 for treatment details

(2.97) followed by T5 whereas, FCE and PER was significantly higher in T3 (0.34, 1.32) and T5 (0.33.1.29) as compared to other treatments and control. Similar result was observed in previous works in FCR in all pre- and pro-biotic supplemented feeds. For instance, the extracellular enzymes might be activated by pre- and pro-biotic supplementation resulting in high feed utilization and improved growth performance. Cultured species are primarily affected by beneficial bacteria through the enhancement of host nutrition due to the stimulation of digestive enzymes resulting in a higher growth and feed efficiency ratio (Suzer et al 2008). Furthermore, the presence of beneficial bacterial cells in the intestine improves microbial balance, which in turn improves nutrient absorption and utilization Lara-Flores 2003). Moreover, the prebiotics addition in fish feed along with probiotics improve feed digestion and availability of nutrients (Gibson et al 2004) leading to improvement in overall fish nutrition and feed efficiency. Khattab et al (2004) revealed positive effect of biogen (synbiotic) @ 0.1% in terms of best growth performance and feed efficiency (reduced FCR) in *O. niloticus*.

Hemato-immunological parameters: The hemato-immunological parameters of fish including Hb, TEC, TLC, Ht or Packed Cell Volume (PCV), MCV, MCH, MCHC and ESR were analyzed at 30, 60 and 120 day interval. Significant difference was observed in hemoglobin content of all the treatments between 30 days and 60days interval except T1 where no significant difference in Hb was observed up to 120days. Elevated hemoglobin content at 60 days in T2, T3, T4, T5 and T6 reduced after 120 days but remained higher in comparison to 30 days. T3 had the significantly higher hemoglobin content at 60 days interval within itself and in comparison, to other treatments (Table 6). At 60 days interval, RBC count was also observed significantly higher in T3 (2.17) followed by T5 and lowest in T1 (1.82). The WBC were significantly higher in all the pre and probiotics supplemented treatments (T2-T6) as compared to control T1. The WBC count was observed highest in T3 (6.01) followed by T5 (5.32), T2, T4, T6 and T1 after 30 days supplemented feeding. Post 60 and 120 days of feeding, WBC reduced significantly in T3 (5.22) and T5 (5.01). Nayak et al (2007) reported higher leucocyte count in rohu, treated with probiotic bacterium *B. subtilis*. Misra et al (2006) revealed significantly increased leucocyte count after 28 day and highest on 42^{nd} day with reduction on 56^{th} day, when BG was added @ 100 to 500 mg/kg. Hassan et al (2014) also recorded highest levels of Hb, Ht, TEC and TLC in *T. niloticus* when fed with *B. licheniformis* ($4.8x10^5$cfu/g) in combination with 5g/kg yeast extract.

The results with respect to hematology in the present study clearly indicated the improvement of health status of fish in T3 (*L. plantarum* @ 10^8 cfu/g + 0.5% β glucan) followed by T5 (*L. plantarum* @ 10^8 cfu/g + 0.5% β glucan) and T6 (*L. plantarum* @ 10^8 cfu/g) in terms of Hb, TEC, PCV and ESR at day 60 whereas TLC also followed the same pattern, but maximum value was at day 30. MCV, MCH and MCHC followed the same pattern and showed variations in accordance to Hb, Ht and TEC. In teleost, probiotic can positively stimulate several immune-hematological parameters, including mononuclear phagocytic cells (monocytes and macrophages), polymorphonuclear leukocytes (neutrophils) and natural killer cells (Balcazar 2003). The results of the present study depicted positive effect of supplementation of probiotic and prebiotic on fish health in terms of higher hemoglobulin along with improved TEC, TLC and Ht. The improvement in hematological parameters is due to the ability of the probiotics to stimulate blood formation (Renuka et al 2014). Further, because of the complex structure of ß glucan, have superior ability to activate the immune response and act as biological response modifiers (Miura *et al* 1996). Larger molecular weight glucans

Table 5. Changes in total feed consumption (g) of *Labeo rohita* (Ham.) in different treatments during the experimental period

Month	Days	Treatments					
		T1	T2	T3	T4	T5	T6
May	30	15.75±0.23	15.70±0.26	15.40±0.10	15.57±0.33	15.70±0.26	15.50±0.26
June	60	24.35±0.44	29.45±1.13	39.80±0.96	29.27±0.81	32.60±0.66	28.65±0.79
July	90	36.45±0.35	42.80±0.97	53.33±2.79	42.95±1.15	47.30±0.87	42.55±1.11
August	120	53.15±0.92	61.50±0.60	69.05±1.85	61.41±0.96	63.40±0.36	59.97±0.35
Total feed		129.70±0.64	149.45±0.63	177.58±2.17	149.20±2.68	159.00±1.96	146.67±1.25
Feed conversion ratio		3.92±0.28	3.29±0.30	2.97±0.06	3.57±0.02	3.03±0.09	3.65±0.04
Feed conversion efficiency		0.26±0.02	0.31±0.03	0.34±0.01	0.28±00	0.33±0.01	0.27±0.003
Protein efficiency ratio		1.01±0.06	1.21±0.11	1.32±0.05	1.10±0.01	1.29±0.03	1.07±0.01

See Table 1 for treatment details

(yeast ß glucan) activate leukocyte, stimulating phagocytic, cytotoxic, antimicrobial activities and production of reactive oxygen species. Studies have shown that insoluble ß glucan of larger molecular weight have greater biological activity than that of its soluble and low molecular counterparts (Ooi and Liu 2000).

Flesh Composition

Total protein: The protein content was significantly higher in all the pre- and pro-biotics supplemented treatments (T2-T6) as compared to T1 (12.58). Among ß glucan + probiotics treatments (T2-T3), the total protein content was significantly higher in T3 (14.90) whereas in MOS + probiotics treatment (T4-T5), the difference for total protein content were insignificant. The results revealed that supplementation of pre and/or probiotic (T2-T6) in feed improved the flesh quality in terms of total protein content (Table 7).

Total lipid: Total mean lipid content in fish flesh ranged from 1.80 to 1.87 g/ 100 g (on wet wt. basis) in treatments T1, T2,

Table 6. Comparative hematological parameters of rohu, *L. rohita* (Ham.) at different time interval in different treatments

Parameters	Days	Treatments					
		T1	T2	T3	T4	T5	T6
Hb (g %)	30	4.23±0.00	5.06±0.04	5.41±0.0	4.91±0.02	5.20±0.00	4.94±0.03
	60	4.33±0.03	5.53± 0.03	6.44±0.07	5.36± 0.03	6.21±0.06	5.20± 0.05
	120	4.32± 0.04	5.22±0.05	5.73±0.09	5.01±0.04	5.25±0.05	4.96±0.03
RBC (x10^6/mm^3/l)	30	1.71±0.02	1.75±0.01	1.75±0.03	1.82±0.01	1.86±0.02	1.87±0.03
	60	1.82±0.01	2.06±0.01	2.17±0.02	2.07±0.02	2.11±0.02	2.02±0.01
	120	1.79±0.04	1.96±0.05	2.08±0.04	1.91±0.03	2.04±0.00	1.90±0.03
WBC (x10^3/mm^3/l)	30	3.87±0.02	4.84±0.02	6.01±0.08	4.55±0.05	5.32±0.07	4.48±0.05
	60	3.95±0.08	4.65±0.04	5.22±0.06	4.35±0.05	5.01±0.08	4.29±0.02
	120	3.83±0.04	4.35±0.06	4.96±0.12	4.16±0.05	4.67±0.02	4.07±0.02
PCV (%)	30	21.93±0.08	23.63±0.31	24.56±0.06	22.96±0.08	23.83±0.03	23.33±0.12
	60	23.50±0.20	26.43±0.14	27.76±0.14	26.46±0.23	26.76±0.38	25.93±0.03
	120	23.47± 0.12	25.10±0.15	26.45±0.25	25.05±0.28	25.43±0.46	24.84±0.05
MCV (μm^3)	30	126.84±1.62	134.55±2.16	139.95±2.69	126.21±1.50	127.95±1.69	124.42±2.43
	60	128.90± 1.67	127.93±1.77	127.79± 1.37	127.71± 2.57	126.70± 2.54	128.39± 0.59
	120	130.79±2.48	128.03±3.32	126.85± 1.52	131.23± 1.92	124.69± 2.62	130.36±2.35
MCH (g %)	30	24.46±0.39	28.81±0.44	30.84±0.65	26.98±0.09	27.91±0.33	26.35±0.35
	60	23.77± 0.27	26.77±0.30	29.66±0.64	25.88±0.15	29.42±0.51	25.74±0.43
	120	24.08±0.83	26.66±0.82	27.51±0.96	26.28± 0.45	25.73±0.18	26.06±0.48
MCHC (g %)	30	19.28±0.07	21.42±0.41	22.03±0.04	21.38±0.20	21.82±0.03	21.19±0.24
	60	18.44±0.30	20.93±0.30	23.21±0.38	20.28±0.29	23.22±0.12	20.05±0.24
	120	18.40±0.28	20.82±0.12	21.67±0.49	20.03± 0.35	20.66±0.52	19.99±0.16
ESR	30	2.36±0.005	2.31±0.003	2.27±0.005	2.32±0.003	2.30±0.008	2.29±0.003
	60	2.35±0.00	2.18±0.01	2.06±0.03	2.20± 0.00	2.07± 0.01	2.21± 0.01
	120	2.82± 0.01	2.27± 0.01	2.21±0.01	2.28±0.01	2.28±0.01	2.30±0.00

See Table 1 for treatment details

Table 7. Flesh composition (% wet weight basis) of *L. rohita* (Ham.) in different treatment at completion of experiment

Parameters (%)	Treatments					
	T1	T2	T3	T4	T5	T6
Total proteins	12.58±0.11	14.07±0.09	14.90±0.12	14.03±0.07	14.37±0.13	14.04±0.12
Total lipid	1.80±0.03	1.87±0.09	1.87±0.09	1.77±0.12	1.80±0.10	1.85±0.12
Total carbohydrate	3.76±0.03	3.37±0.13	3.50±0.15	3.43±0.13	3.43±0.12	3.03±0.05
Ash	1.84±0.03	1.79±0.01	1.77±0.02	1.71±0.04	1.70±0.02	1.69±0.01
Moisture	79.92±0.17	79.16±0.022	78.00±0.10	79.13±0.07	78.17±0.05	79.19±0.24

See Table 1 for treatment details

T3, T4, T5, and T6 (Table 7) and the differences among treatments were insignificant (T6=T5=T4=T3=T2=T1). The results indicated that probiotic supplementation did not affect the flesh total lipid content significantly.

Total carbohydrate: In different treatments, the total carbohydrate content in fish flesh was significantly higher in T1 (3.76g/100 gm) with lowest in T6 (3.03g/100 gm). The results revealed significant decrease in total carbohydrate content of flesh with pre and/or probiotic supplements.

Ash content: Maximum ash content was observed in T1 (1.84g/100gm) and lowest in T6 (1.69/100gm) with significant difference. The result indicated that pre and/or probiotic supplementation in fish feed resulted in decrease in ash content of fish flesh.

Moisture content: In different treatments, moisture in fish flesh was highest in T1 (79.92) and lowest in T3 (78.00) and the differences among treatments were significant (Table 7). Among ß Glucan + probiotics treatments (T2-T3), the total moisture content was significantly low in T3 (78.0), whereas in MOS + probiotics treatment (T4-T5), the mean total moisture content was significantly low in T5 (78.17).

Flesh quality in terms of total protein and fat revealed improved body composition of rohu in the present study. Toutou et al (2016) also reported improved biochemical composition in terms of enhanced protein and fat content of grass carp when fed with either probiotics alone or in combination with prebiotic in the form of commercial synbiotic (Microban aqua). According to Abdel Tawwab et al (2010), yeast supplementation improved the protein content of flesh with no significant differences in lipid content of *O. niloticus*. These findings are also in agreement with present study in terms of improved flesh quality with probiotic with β-glucan/MOS (extracted from yeast) supplemented diets.

CONCLUSION

The diet having only probiotic (*L. plantarum@* 10^8) revealed higher net profit (6.91%) in comparison to control on the basis of the overall fish growth performance, health status with special reference to hematological parameters and flesh composition, *L. plantarum* @ 10^8 cfu/g + 0.5 % ß-glucan can be given as booster diet to fingerlings *L. rohia* for 60 days during the growth period along with routine feeding. Hence, dietary supplementation of prebiotic β-glucan along with *L. plantarum* @ 10^8cfu/g in diet is effective in enhancing the immunity and overall health status of *L. rohita*.

AUTHOR'S CONTRIBUTIONS

Prem Kumar-Conducted the experiment and analysis work w.r.to all parameters and manuscript writing; Vaneet Inder Kaur- Conceived the idea, planning and execution of experimental study and critical inputs in manuscript; Anuj Tyagi-Helped in isolation of probiotic bacteria and feed formulation; Sachin O. Khairnar- helped in analysis work pertaining to hemato-immunological parameters and flesh composition.

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Diversity of Mollusks and Herpetofauna along River Beas in Punjab, India

B.K. Babbar, Tejdeep Kaur, Shammi Kapoor[1] and Priya Katyal[1]

Department of Zoology, [1]Department of Microbiology,
College of Basic Sciences & Humanities, PAU, Ludhiana-141 004, India
E-mail: bhupinder@pau.edu

Abstract: The diversity of herpetofauna and mollusks along the river bank can act as bioindicator of the ecological environment. The faunal diversity was studied from 2019 to 2021 along the bank of River Beas during the spring, summer, and autumn seasons. In 2019, four mollusks, and six reptilian species were recorded along the river bank. However, in 2020, six mollusks species and two amphibians were observed. Mollusks of eight different species, Indian bullfrog, and freshwater turtles were observed in 2021. An increase in the number of mollusk species along River Beas from 2019 to 2021 indicated an improvement in the quality of water. Two amphibian species namely *Hoplobatrachus tigerinus* and *Rana limnocharis* were predominant in the study area. These were not seemed to be affected by changes occurring along the river because of the presence of alternate habitats in the form of large tracts of land under paddy cultivation in the study area. Large groups of turtles near the river bank in 2021 indicated an improvement in water quality.

Keywords: River Beas, Mollusks, Herpetofaunal diversity, Bioindicators

For in situ conservation of biological diversity, Punjab has created Protected Area Network (PAN) to accord protection to its wildlife and associated habitat. The state has a PAN consisting of 13 wildlife sanctuaries, five wildlife Conservation Reserves, four Community Reserves, and six Ramsar Sites. River Beas has been notified as Conservation Reserve by the Government of Punjab, Dept. of Forests and Wildlife Preservation (Forest Branch) in 2017 and the Ramsar site in 2019 (Anonymous 2019). The River Beas rises from the Himalayas in Himachal Pradesh and flows 185 km across Punjab State from Talwara, Hoshiarpur, and merges with Sutlej River at Harike, Tarntaran, Punjab. The Beas Conservation Reserve hosts the only population of Indus River dolphins *(Platanista gangetica* minor) in India. Ninety-four gharials were also introduced near the village Gagdewal in Beas. The smooth-coated otter (*Lutrogale perspicillata*) was also reported in River Beas along with freshwater turtles like the Indian softshell turtle, Indian flapshell turtle, narrow-headed softshell turtle, spotted pond turtle, crowned river turtle and brown roofed turtle, and more than 90 fish species. A wide variety of avian species (about 500 species) were also reported (Kanwar et al 2013, Kanwar 2019, Kanwar and Lomis 2020, RIS 2020). Regular monitoring of faunal diversity is essential especially when anthropogenic activities disturb the ecosystem. In May 2018, the accidental release of molasses in River Beas from a sugar mill near Gurdaspur District severely impacted the aquatic life and the ecological environment of the river. The diversity of herpetofauna and mollusks along the river bank can act as an index of bio-indicator of the ecological environment. They also form an important link in the food chain. Keeping this in view, surveys were conducted from 2018-19 to 2020-21 along river Beas to study herpatofauna and molluskan diversity.

MATERIAL AND METHODS

This study was conducted from 2019 to 2021 during the autumn and spring seasons. Faunal diversity at different sites along river Beas bank in village Dhilwan, Amritsar district, villages Chambha and Harike, Tarntaran district and villages Alampur and Bhait, Gurdaspur district were recorded (Plate 1). Both live and dead mollusk species were collected by hand picking method along the bank of river Beas as well as in paddy crop fields, brought to the laboratory, washed, and then identified (Patil et al 2011). Amphibian and reptiles were also photographed and identified (Ali et al 2016, Ali et al 2017). A Cannon Powershot camera was used for the photography of animals along the river bank.

RESULTS AND DISCUSSION

In 2019, four mollusks i.e. *Indoplanorbis exustus, Gabbia orcula, Lymnaea, Corbicula,* and one amphibian *Hoplobatrachus tigerinus* were observed along the river bank in Dhilwan, and six reptilian species *Varanus varius, Craspedocephalus gramineus, Eutropis macularia, Gerarda prevostiana, Fowlea piscator and* rat snake *were* observed in

Talwara, and Harike, near the river bank (Plates 2 and 3). However, in 2020, two new molluskan species i.e. *Bellamya bengalensis*, and *Cryptozona semirogata,* were observed near the river bank in Chamba and Harike, and one new amphibians, *Rana limnocharis* was observed in the village Dhilwan (Plates 2 and 3). In 2021, two more mollusks i.e. *Gyraulus, and Bithynia tentaculata* were observed in paddy crop fields and near the river bank in Bhait, Alampur, Dhilwan, Chamba, and Harike. Indian bullfrog and groups of freshwater turtles were also observed near the river bank in village Harike during this year (Plates 2 and 3). Classification of mollusks, reptiles, and amphibians spotted along River Beas from 2019 to 2021 is given in Tables 1-2.

Mollusks were more in areas where the water current was

Plate 1. GPS Map showing selected villages

Table 1. Classification of mollusk spotted along river Beas from 2019 to 2021

Kingdom	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia
Phylum	Mollusca	Mollusca	Mollusca	Mollusca	Mollusca	Mollusca	Mollusca	Mollusca
Class	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Bivalvia
Order	Basommato-phora	Littornimorpha	Basommato-phora	Architaenioglossa	Basommato-phora	Littorni-morpha	Stylommatophora	Venerida
Family	Planorbidae	Bithyniidae	Lymnaeidae	Viviparidae	Planorbidae	Bithy-niidae	Ariophantidae	Cyrenidae
Genus	*Indoplanorbis*	*Gabbia*	*Lymnaea*	*Ballamya*	*Gyraulus*	Bithynia	*Cryptozona*	*Corbicula*
Species	*Exustus*	*Orcula*		*Bengalensis*			*Semirugata*	
Binomial name	*Indoplanorbis exustus* (Deshayes 1834)	*Gabbia orcula* (Frauenfeld 1862)	*Lymnaea* (Lamarck 1799)	*Ballamya bengalensis* (Lamarck 1822)			*Cryptozona semirugata* (Beck 1837)	*Corbicula* (Muller 1774)
Common name	Freshwater snail	Freshwater snail	Freshwater snail	Freshwater snail	Freshwater snail	Freshwater snail	Land snail	Freshwater clam
Already reported	In Pothwar, Pakistan Chandigarh (Afshan et al 2013, Maansi et al 2021)		In Madhya Pardesh, Assam, Jammu, Pothwar, Pakistan, (Garg et al 2009, Roy and Gupta 2010, Sharma and Chowdhary 2011, Afshan et al 2013)	In Gujarat, West Bengal, Narmada River, Assam, Rajasthan (Bhalodia et al 2001, Roy and Nandi 2008, Pir et al 2010, Roy and Gupta 2010, Vyas et al 2012, Sharma et al 2013)	In Pothwar, Pakistan, Chandigarh (Afshan et al 2013, Maansi et al 2021)	In Pothwar, Pakistan, Chandigarh (Afshan et al 2013, Maansi et al 2021)		


Indoplanorbis

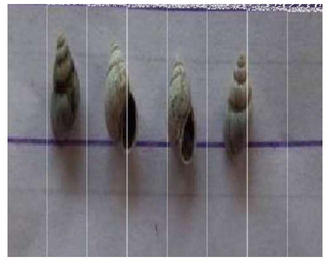

Gabbia orula


Lymnaea


Corbicula


Cryptozona


Bellamya


Bithynia


Gyraulus

Plate 2. Different mollusk species observed along River Beas


Fowlea piscator (Asiatic Water Snake)


Varanus varius (Monitor Lizard)


Craspedocephalus gramineus
(Indian green pit viper)


Eutropis macularia (Bronze grass skink)


Gerarda prevostiana (Glossy marsh snake)


Hoplobatrachus tigerinus (Indian bull frog)




Fresh Water Turtles and their foot marks along River Beas Bank

Plate 3. Reptiles and Amphibians observed along River Beas

Table 2. Classification of Amphibians and Reptiles spotted along river Beas from 2019 to 2021

Kingdom	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia	Animalia
Phylum	Chordata	Chordata	Chordata	Chordata	Chordata	Chordata	Chordata	Chordata	Chordata
Class	Amphibia	Amphibia	Reptilia	Reptilia	Reptilia	Reptilia	Reptilia	Reptilia	Reptilia
Order	Anura	Anura	Squamata	Squamata	Squamata	Squamata	Squamata	Squamata	Testudines
Family	Dicroglossidae	Dicroglossidae	Varanidae	Viperidae	Scincidae	Homalopsidae	Colubridae	Colubridae	
Genus	*Hoplobatrachus*	*Rana*	*Varanus*	*Craspedocephalus*	*Eutropis*	*Gerarda*		*Fowlea*	
Species	*H. tigerinus* *(*Daudin 1803)	*R. limnocharis* *(*Gravenhorst 1829)	*V. varius* (Merrem 1820)	*C. gramineus* (Shaw 1802)	*E.macularia* (Blyth 1853)	*G. prevostiana* (Gray 1849)		*F. piscator* (Schneider 1799)	
Common name	*Indian bull frog*	Frog	Monitor lizard	Indian green pit viper	Bronze grass skink	Glossy marsh snake	Rat snake	Asiatic water snake	Fresh water turtle
Already reported	In Kasur, Pakistan, Kalbagh Game Reserve, Indus River, Pakistan (Ali et al 2016, 17)	In Ropar Wetland (Ladhar 2000)				West Coast, Maharashtra (Prabhakar et al 2020)		West Coast, Maharashtra (Prabhakar et al 2020)	Kalbagh Game Reserve, Indus River, Pakistan (Ali et al 2017)

slow. These animals depend upon this habitat for feeding and breeding. Species diversity recorded along River Beas indicated good water quality to support biodiversity. Freshwater mollusks (mussels and snails), like other macro-invertebrates, are vital components of any aquatic ecosystem. Their sensitivity to the habitat's conditions allows them to act as biological indicators of the health of the ecosystem. Out of the various mollusks recorded during the present study *Indoplanorbis exustus, Gabbia orcula, Lymnaea, Corbicula* are the most resistant species. These species were reported to thrive well in polluted waters and during dry phases (Gnatyshyna et al 2011, Sharma et al 2013, Guo and Feng 2018, Kavitha et al 2018). Gupta et al (2015) reported the maximum variety and number of gastropods in upstreams and the lowest variety and number in downstream near wastewater discharge areas. An increase in the number of molluskan species along River Beas from 2019 to 2021 indicated an improvement in the quality of water.

Maansi et al (2021) studied the diversity of molluskan fauna from freshwater bodies of Chandigarh and River Ghagar. They reported minimum molluskan diversity in polluted water bodies (Ghaggar River, N-Choe, and Dhanas Lake) and maximum molluskan diversity in Sukhna Lake, Chandigarh. Singla et al (2017) reported the presence of five species of snails, *Indoplanorbis exustus*, *Radix luteola*, *Melanoides tuberculata*, *Bithynia tentaculata*. *Kashmiriensis,* and *Cryptozona bistrialis* belonging to families, Planorbidae, Lymnaeidae, Thiaridae, Bithyniidae, and Ariopahntidae in paddy crop fields in Punjab State. Out of these 7 species, 3 species were also recorded along river Beas during the present study. *Lymnaea and Gyraulus* species were also reported in the Ropar wetland, Punjab (Brraich and Saima 2018). Jamwal et al (2017) reported three mollusk species (*Digoniostoma pulchella, Bellamya bengalensis* and *Lymnaea acuminata*) in River Beas near Indore, district Kangra, Himachal Pradesh.

Punjab biodiversity board had earlier reported 85 mollusk, 35 reptiles, and 15 amphibian species in Punjab. Rais et al (2012) reported 35 species of herpetofauna, amphibians (5 species), and reptiles (30 species) in selected areas of North Punjab, Pakistan. Out of these, three reptiles, a monitor lizard, Asiatic water snake, and fresh water turtles, and two amphibians, *Hoplobatrachus tigrinus, and Rana limnocharis* were recorded along River Beas during the present study. Ali et al (2016) reported three amphibian species *Bufo stomaticus, Hoplobatrachus tigrinus,* and *Euphlyctis cyanophlyctus,* and 4 reptilian species *Lissemys puntata, Varanus bengalensis, Xenochrophis piscator,* and *Kachuga smithi* from water catchment area in Kasur dist., Punjab. *Xenochrophis piscator* (Asiatic water snake), and *Nilssonia gangetica* (Indian softshell turtles) were also reported earlier in River Beas. The River Beas was also earlier reported to support seven species of freshwater turtles (Kanwar et al 2013).

CONCLUSIONS

An increase in the number of molluskan species along River Beas from 2019 to 2021 indicated improvement in the quality of water. Two amphibian species were predominant in the study area. These were not seemed to be affected by changes occurring along the river because of the presence of alternate habitats in the form of large tracts of land under paddy cultivation in the study area. Large groups of turtles near River Bank in 2021 also indicated an improvement in water quality. The increase in the area under agriculture along the river bank and human activities disturb their habitat. Therefore, there is a need to conserve these areas well for long-term sustainability of animal diversity.

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Mammalian Diversity Recorded along River Beas in Punjab, India

Tejdeep Kaur Kler, B.K. Babbar, Sachin Kumar, Shammi Kapoor[1] and Priya Katyal[1]

Department of Zoology, [1]Department of Microbiology,
College of Basic Sciences & Humanities, Punjab Agricultural University, Ludhiana-141 004, India
E-mail: tejdeepkler@pau.edu

Abstract: Present communication pertains to survey on mammalian diversity along the river Beas from 2018-19 to 2020-21 during autumn and spring seasons. Mammalian diversity has been influenced by habitat loss because of cultivated areas along the rivers and other anthropogenic activities. Twelve species of mammals have been identified comprising of one vulnerable and one near threatened species along the river Beas. There is need to restore, rejuvenate and protect wild habitats along the course of river Beas.

Keywords: River Beas, Mammalian diversity, Punjab

Large mammals are often described keystone species that maintain stability of ecosystem and have impact on biodiversity. Rapid agricultural expansion and economics growth has been instrumental in changing habitats, distribution pattern, range and diversity of mammals in India (Das et al 2006). In Punjab, there is intensive agriculture and substantial increase in area under crops in last five decades (Anonymous 2022). Habitat fragmentation and habitat loss has affected distribution of animal fauna in Punjab State (Dinesh et al 2021, Kumar et al 2021, Sethy et al 2021). The Beas River is a river in north India. It rises in the Himalayas in central Himachal Pradesh, India, and flows for some 470 kilometers before merging with the Sutlej River at Harike Wildlife Sanctuary, situated south of Amritsar in Punjab. The river flow through extensively cultivated landscape. Other than crop cultivation, the vegetation composition along the river banks comprises of different types of grasses, shrubs and indigenous trees. The habitat heterogeneity and a complex land-water association make a floodplain able to maintain disproportionally high biodiversity. In spite of its vital importance, the information on the mammalian biodiversity along river bank is incomplete. Keeping this in view, surveys were conducted from 2018-19 to 2020-21 to study mammalian diversity along River Beas.

MATERIAL AND METHODS

This study was conducted from 2018-19 to 2020-21 during autumn and spring seasons. Mammalian diversity was studied at different sites along River Beas bank at Dhilwan (31.5100°N, 75.3354°E) of district Kapurthala; Goindwal Sahib (31.3638°N, 75.1370°E), Chamba kalan (31.2083°N, 75.0093°E) and Harike (31.1689°N, 74.9428°E) of district Tarntaran; Talwara (31.9311° N, 75.8941°E) and Tanda (31.6648° N, 75.6303° E) of district Hoshiarpur; Alampur (31.4963° N, 75.5306°E) and Bhet (31.7689°N, 75.5322°E) of district Gurdaspur. Mammalian species seen were photographed and identified (Atwal et al 1984). Rodent species were identified on the basis of characteristic burrow entrances; all the burrow entrances were closed in the evening and next day reopened and active burrows were counted. Number of active rodent burrows gives a rough estimate of rodent population (Singla *et* al 2022). Transect sampling method was used relying on visual counts for survey of large mammals (Buckland et al 2015). Mammalian species were identified as per the "Book of Indian Animals" by Prater (1971).

RESULTS AND DISCUSSION

During the study period in 2018-19, different species of rodents, wild boar, blue bull, black naped hare, Indian grey mongoose and palm squirrel were recorded. Stray animals like cows and dogs were spotted at locations during most of the observations at Talwara, Dhilwan, Goindwal Sahib and Harike. Survey of wheat crop after sowing during this period on the bank of River Beas in villages of district Tarntaran revealed the presence of three rodent species i.e., *Bandicota bengalensis, Mus booduga and Rattus meltada.* Number of burrows ranged from 19.15 to 28.73/ha indicating high rodent population density. At pre-harvest stage, survey of wheat crop fields in districts Gurdaspur, Hoshiarpur, Amritsar and Tarntaran near riverbank revealed the presence of four rodent species i.e., *B. bengalensis, Tatera. indica, M. booduga and R. meltada.* However, at this stage, rodent population density was medium with number of rodent burrows ranging from 2.29 to 8.90/ha. Survey in paddy crop fields revealed very low rodent infestation and number of burrows were only 1.89/ha. Foot prints and damage of Blue Bull, Wild Boar and Indian Hare were also noted in villages of district Gurdaspur, Amritsar and Hoshiarpur near river Beas. In 2019-20, four rodent species, blue bull and wild Boar were recorded near riverbank at different locations surveyed. In addition, Rhesus Macaque, Common Indian mongoose, Indian hare, feral horse, palm squirrel were also recorded. In 2020-21, survey of rodent diversity and damage in paddy fields near riverbank and along riverbank indicated presence of three rodent pest species. Population density was low to high rodent infestation in these fields. Number of burrows were less in monsoon season. Wild boar was also recorded in villages near river bank of district Gurdaspur, Tarntaran and Amritsar. However, Blue Bull was seen only near river bank at Dhilwan. In addition, Indian jackal and feral horses were also recorded (Table 1). During present study, survey of wheat and paddy crop fields near River Beas in villages of district Gurdaspur, Hoshiarpur, Amritsar and Tarntaran revealed medium to high population density of different rodent species like *B. bengalensis, T. indica, M. booduga and R. meltada* in wheat and low population density in paddy crop fields. Singla and Babbar (2010) also reported four rodent species with high rodent infestation in wheat crop fields in comparison to paddy in Punjab. Blue bull was also located along River Beas in villages of district Gurdaspur, Amritsar and Hoshiarpur. Earlier studies also reported that blue bull prefers to live near water areas and in the habitat dominated by plants like *Sisso* (*Dalbergia sisso*), saguwan (*Tectona grandis*), eucalyptus, poplar, *Jamun* (*Eugenia jambolana*) (Aryal 2007). Indian jackal and feral horses were also recorded during the survey.

Mudappa and Choudhury (2016) had assessed status of Indian Grey Mongoose and added to IUCN list of Least Concern. Present investigation has confirmed that the sightings of Indian Grey Mongoose are becoming rare and rarer as per author s surveys carried out in last two and half decades. The population is declining at regional level in intensive agricultural scenario of Punjab State. Rhesus macaque was noted in groups of 2-5 individuals per sighting at Talwara and mostly in spring season. Farmers were also enquired about human-macaque conflict in crop fields which they replied in negative. Anand et al (2021) observed high feeding intensity of rhesus macaque on cultivated crops in human modified landscapes in North India. It was further mentioned to be positively correlated to the presence of deciduous forest stands in vicinity of crop fields.

Numerous hares (3 to 5 in numbers) were sighted in river side grass patches and sometimes in fallow fields along the River Beas at locations namely Goindwal Sahib in district Tarntaran, Talwara and Tanda falling in district Hoshiarpur. In the surveyed areas, natural enemies of hare like Mongoose and stray dogs were at Goindwal Sahib and Talwara.

Table 1. List of mammals recorded along River Beas

Species	No. of individuals per sighting	Locations	IUCN status	IWPA status schedule
Palm Squirrel *Funambulus pennanti* (Wroughton 1905)	2-5	Goindwal Sahib, Chamba kalan, Harike, Bhet, Talwara, Tanda	Least concern	IV
Soft Furred field Rat *Rattus meltada* (Gray 1837)	2-14 burrows/acre	Dhilwan, Talwara, Tanda	Least concern	IV
Indian Mole Rat *Bandicota bengalensis* (Gray 1835)	7-29 burrows/acre	Dhilwan, Goindwal Sahib, Chamba kalan, Harike	Least concern	IV
Indian Gerbil *Tatera indica* (Hardwicke 1807)	7-17 burrows/acre	Dhilwan, Bhet, Talwara, Tanda	Least concern	V
Common Indian Field Mouse *Mus booduga* (Gray 1837)	4-11 burrows/acre	Goindwal Sahib, Dhilwan, Bhet	Least concern	IV
Golden Jackal *Canis aureus* (Linnaeus 1758)	1-2	Chamba kalan, Harike	Least concern	II
Indian Grey Mongoose *Herpestes edwardsii* (E. GeoffroySaint-Hilaire 1818)	1-2	Goindwal Sahib, Chamba kalan, Harike, Bhet, Talwara	Least concern	II
Black Naped Hare *Lepus nigricollis* (F. Cuvier 1823)	3-5	Goindwal Sahib, Dhilwan, Tanda	Least concern	IV
Wild Boar *Sus scrofa* (Linnaeus 1758)	2	Dhilwan, Bhet, Tanda	Least concern	III
Rhesus Macaque *Macaca radiate* (E. Geoffroy 1812)	2-5	Talwara	Vulnerable	II
Blue Bull *Boselaphus tragocamelus* (Pallas 1766)	4-10	Dhilwan	Least concern	III
Feral Horse *Equus* sp. Linnaeus 1758	4-6	Talwara, Harike	Near threatened	I

Predator prey conflict between Indian Hare and Indian Mongoose was observed on one occasion at Talwara, where their chase in wild vegetation ended in hare disappearing in some burrow or natural pit. Golden Jackal was recorded moving adjacent to tall wild grasses along banks of River Beas in locations at village Chambakalan and at Harike falling in district Tarntaran. Jackal was noted solitary on two occasions in severe summer days in June month near the water line in riverine habitat. It was quick to disappear in more than 6 feet tall grasses on slightest disturbance. Farmers working in adjoining fields have narrated that they often hear howling of Jackals near wild vegetation edges for food searches. Srinivas and Jhala (2021) had mentioned that three species of wild canis occur in India which include two species of wolves namely Indian wolf (*Canis lupus pallipes*), Himalayan wolf (*Canis himalayensis*) and one species of Golden Jackal (*Canis aureus*). In India, Golden Jackal is protected under Schedule II (part II) of the Indian Wildlife (Protection) Act, 1972. The act provides complete protection for Schedule I and Schedule II species and heavy penalties are prescribed for any offence. As per IUCN list, grey wolves and golden jackals are considered as least concern (Boitani et al 2020, Hoffmann et al 2020). Workers stated that wild Canis species are getting threatened by increasing populations of stray dogs. Competition for food resources, shelter and anthropogenic threats might pose threat to wolf and jackal populations and irreversibly impact their survival (Jhala and Giles 1991, Chawla et al 2020). Feral Horse *Equus* sp. were observed in group of 4 to 6 in riparian zone along the River Beas at Talwara. One group was at Harike and these feral horses were noted grazing in river side area in proximity to small pools and islands. More than two groups were recorded at Talwara in summer season, there occurrence near river water pools and riparian zone seemed to be because of water requirement, grasses and isolated/undisturbed habitats. Changmail et al (2021) reported feral horse distributed in grasslands along riverine habitats and in wetlands dominated by different grass species in Dibru Saikhowa National Park in Northeast India.

Small populations of wild Boar inhabiting uncultivated areas along the river Beas at Dhilwan, Bhet and Tanda. There were incidences of wild boar damage to tuberous crops, sugarcane and cereal crops in study areas. Yasmita-Ulman et al (2020), observed that negative human-wildlife conflict interactions in agroforestry systems originates mainly from intolerance of land owners to crop depredations by small mammals like rodents and Indian hare. Diversity of mammalian species in India and their populations are under threat due to anthropogenic activities and human settlements. Intensive agriculture and urbanization are proven threats to habitats of smaller mammals like civets, mongoose, jungle cats and wild boars (Michael and Joonu 2021, Singh et al 2021). Bhat and Bhat (2022) had reported 18 species of mammalian comprising two vulnerable, and three near-threatened species from Ghatiga on Bustard Sanctuary, Madhya Pradesh.

Twelve species of mammals were recorded inhabiting wild vegetation blocks/patches for sheltering/hiding and foraging purposes along River Beas. There are needed urgent and timely interventions to strengthen already existing wild vegetation blocks/small forests for conservation of mammalian diversity in River Beas Conservation Reserve.

CONCLUSION

Habitat niches inhabited by mammalian species along River Beas deserve attention of conservationists and ecologists for their restoration and rejuvenation involving all stakeholders.

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AUTHOR CONTRIBUTION

Dr Tejdeep Kaur Kler was Principal Investigator of River Beas Project under whose mandate work was done. Dr Tejdeep Kaur Kler and Dr B K Babbar carried out surveys, compiled data and wrote manuscript. Sachin Kumar involved in surveys and recording data. Dr Shammi Kapoor was convener of River Beas Project under which study was conducted and helped in experimental designing. Dr Priya Katyal was involved in field surveys and data compilation.

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Occurrence of Amphibians and Reptiles at Village Ponds in Ludhiana, Punjab: Diversity, Threats and Conservation Prospects

Gurkirat Singh Sekhon, Randeep Kaur Aulakh and Tejdeep Kaur Kler
Department of Zoology, Punjab Agricultural University, Ludhiana-141 004, India
E-mail: gurkiratsekhon@gmail.com

Abstract: Amphibians and reptiles, collectively referred as herpetofauna are integral constituents of animal diversity associated with village pond habitats in agro ecosystems. Present investigation was carried out with objective to assess diversity of amphibians and reptiles in relation to water quality parameters by selecting two ponds in villages namely Jhamat and Malakpur; third selected pond was in Punjab Agricultural University (PAU) campus falling under district Ludhiana from March 2019 to February 2020. Line/point transect methods and visual encounter surveys were followed to record diversity of amphibians and reptiles. Overall, two species belonging to class Amphibia i.e. *Euphlyctis cyanophlyctis* (Schneider, 1799) and *Duttaphrynus stomaticus* (Lutken, 1864) and four species of class Reptilia i.e *Lygosoma punctate* (Gmelin, 1799), *Ptyas mucosa* (Linnaeus, 1758), *Bungarus caeruleus* (Schneider, 1801) and *Varanus bengalensis* (Daudin, 1802) were recorded at studied ponds. Class Reptilia was indicated positive correlation with Class Amphibia. Both Amphibia and reptilia had also shown positive relation with water temperature, ambient temperature, pond area and fallow land area; whereas values of pH, BOD and DO were found to be negatively related. Land use changes due to agricultural intensification and anthropogenic factors have been altering water quality parameters thereby threating herpetofauna inhabiting freshwater pond habitats in villages. Urgent and focused conservation interventions are required to preserve village pond habitats accompanied with steps to conserve animal groups like herpetofauna in agriculture dominated Punjab State.

Keywords: Amphibians, Ecological indicators, Reptiles, Village ponds

Large portion of world's animal diversity is associated with the agro ecosystems. Agricultural landscape constitutes crop fields, pond habitats, human settlements, allied sectors like dairy farms and barren lands. Two-thirds of entire terrestrial ecosystems are composed of human modified ecosystems like agricultural landscape, human habitations and agroforestry plantations. Freshwater bodies like village bonds are characteristic features of agro ecosystems. Ponds are the abode of diverse types of animal communities which are inter related and inter dependent (Albero et al 2021, Lewis-Phillips et al 2020). Amphibians and reptiles are cold blooded, habitat specific and sensitive to environmental changes; they are considered as indicators of healthy ecosystems. Amphibians and reptiles are collectively referred to as herpetofauna constituting prey and predators components in intricate food webs existing particularly in terrestrial and specifically in agricultural ecosystems (Yadav et al 2022). Vertebrate predator populations like herpetofauna have potential as biological control agents of lower vertebrates like rodents in agro ecosystems (Twining et al 2022).

In India, a total of 421 species of amphibian under 64 genera and 16 families have been reported; 73.6% amphibian species are endemic most of these fall under rare or threatened species categories. 43 species belonging to 15 genera and 06 families have been represented in agro-ecosystems (Dinesh et al 2019). There are about 571 species of reptiles are reported in India which belong to three orders of reptiles: order Crocodylia having 3 species of crocodiles, 34 species of turtles and tortoises belonging to order Testudines and order Squamata having 231 of lizards and 303 of snakes (Aengals et al 2018). Sethy et al (2021) stated 64 species of reptiles (39 genera and 10 families) have been represented in agro-ecosystem; out of these 15 species of reptiles are endemic.

As per IUCN list of threatened species, 41% of amphibian species and 21% species of reptiles are threatened with extinctions globally (https://www.iucnredlist.org/). Wide spread declines in diversity of herpetofauna have been reported which have potential of serious repercussions on food chains via trophic guilds in agricultural habitats. Numerous workers have reported negative effects of agricultural intensification on animal communities like amphibians, reptiles and water dependent birds in ponds (Kaur et al 2018, Sidhu et al 2021). Systematic and detailed field studies are lacking on documentation of herpetofauna from agro-ecological zones of India (Dinesh et al 2021, Sethy et al 2021). Herpetofauna inhabiting village pond habitats in agricultural areas have been less studied particularly in India specifically in agrarian state like Punjab. Inventories of Herpetofauna of pond habitats both at local and regional scales is still unexplored which is a hurdle in formulating conservation strategies. Therefore, the present study was planned to assess diversity of amphibians and reptiles in relation to water quality parameters by selecting two ponds in villages of Ludhiana district and one pond in PAU Campus, Ludhiana.

MATERIAL AND METHODS

The present study was carried out in three selected ponds (A, B, C) in villages namely Jhamat, Malakpur and sewage treatment pond of Punjab Agricultural University (PAU) campus (latitude 30° 54' 3.4740"N and longitude 75° 51' 26.1972"E) in Ludhiana district from March 2019 to February 2020. Selected habitats of ponds were surveyed on fortnightly basis following Line/Point count transect methods (Verner 1985). Pond habitat characteristics Pond A (area 1.01 ha) was located in the village surrounded by residential houses. Pond B (1.21 ha) near outskirts of the village having residential houses on one side of the pond and agricultural fields on other sides. Pond C (1.61 ha) was manmade pond unit and consisted of 4 tanks having walled boundaries formed for sewage treatment plant. Pond C was located near the botanical garden and surrounded by crop fields of PAU campus. All the studied ponds except pond C were natural ponds which remained filled throughout the year and were dependent on rainfall. Pond C remained filled with treated sewage water. Field observations on amphibians and reptiles were carried out through visual encounter survey. Identification of species was made by noting down morphological characteristics which were compared with identification keys given by Daniel (2002) in "The Book of Indian Reptiles and Amphibians" and standard reference book entitled "Snakes of India: Field Guide" authored by Whitaker and Captain (2004).

The analysis of the physico-chemical water quality parameters like water temperature, pH, biological oxygen demand (BOD) and dissolved oxygen (DO) were carried out. Temperature was recorded from Indian meteorological department website. Water temperature was recorded by using Mercury glass thermometer. pH was determined by using portable digital pH meter for the water samples. BOD and DO was analyzed at as per reference of APHA (2012). Correlation analysis was carried between Class amphibia and reptilia with physico-chemical characteristics of water and areas of selected ponds. Google maps were taken from www.google maps and land use maps were prepared by Punjab Remote Sensing Centre, Ludhiana.

RESULTS AND DISCUSSION

Two species belonging to class Amphibia i.e *Euphlyctis cyanophlyctis* (Schneider 1799) (Indian skipper frog or skittering frog) and *Duttaphrynus stomaticus* (Lutken 1864) (Indian marbled toad or Punjab toad); four species of class Reptilia i.e *Lygosoma punctate* (Gmelin 1799) (common dotted garden skink or punctuate supple skink), *Ptyas mucosa* (Linnaeus 1758), (Indian rat snake or oriental rat snake), *Bungarus caeruleus* (Schneider 1801) (Indian krait or blue krait) and *Varanus bengalensis* (Daudin 1802) (Bengal monitor or common Indian monitor) were recorded (Table1). Indian skipper frogs were often found along the edges of ponds A and B and most prevalent during the monsoon season followed by summer season. Their population was variable at both ponds A and B; highest numbers were recorded during the monsoon season and lowest during summer season. Indian marbled toad was observed in pond edges, under leaves and undergrowth at the pond B. Population number followed the same trend as that of Indian skipper frog. Herpetofauna was not found in winter months because of hibernation.

Field investigations of natural populations of amphibians have revealed correlations between population declines and vicinity to agricultural areas (Davidson et al 2002, Houlahan and Findlay 2003). In present study, amphibian species were not observed at Pond C which seemed to be because of sewage treated water and surrounded by crop fields. Water draining from intensive agricultural lands also consists of pesticides/herbicides which affect non-target organisms in the water bodied by modifying the structure and functionality of freshwater ecosystems (Vera et al 2009). At Pond A, only one species of frog namely *E. cyanophlyctis* was noted; it was having walled banks and surrounded by human habitation; anthropogenic impact might be the cause behind less diversity of amphibian taxa. One species each of frog and toad was recorded at pond B; it seemed to be due to the habitat features like large size of water body and less human disturbance was complemented by wide array of wild vegetation. Agriculture and human settlements have been proved as having negative effects on diversity of herpetofauna worldwide (Thompson et al 2016). *L. punctuate* and *V. bengalensis* were recorded at pond A. They were mostly observed hidden beneath logs, rocks, or among piles of leaves and twigs. Their population showed seasonal variation at pond A being highest during the monsoon season. At pond B, *L .punctate* and *P. mucosa* were recorded in undergrowth, dung piles and in wild vegetation along the edges. Their population showed seasonal variation which was highest during the monsoon season. The sightings of *P. mucosa* was always solitary in nature. *B.caeruleus* was observed only at the pond C along the short wild grass during the monsoon season.

Correlation analysis of water quality parameters with diversity of amphibians and reptiles was carried out. The average ambient temperature and water temperature were strongly related to the diversity Class Amphibia at Pond A and B (Table 2). It may be inferred that increase in said parameters seem to have positive influence on the diversity of Class Amphibia. Strong positive relationship of ambient temperature and water temperature was in Class Reptilia at all studied ponds. Amphibians have been recognized indicator taxa to evaluate habitat quality and environmental stresses. pH indicted inverse relationship with Class Reptilia at selected ponds. BOD was insignificant for Class Reptilia at pond A. DO have negative correlation with Class Reptilia which could be due to species specific foraging niches as herpetofauna were having adaptations for both aquatic and terrestrial existence. Class Amphibia and Class Reptilia had positive relation with water temperature, ambient temperature, pond area and fallow land area; pH, BOD and DO was found negatively related to species richness (Fig. 1). In the current study, two species of amphibians in selected ponds are indicative of quality of fresh water habitat and healthy food webs. Four species of reptiles belonging to families namely Scincidae (1 species), Culobridae (1 species), Elapidae (1 species) and Varanidae (1 species) were documented during the present survey. All the four reptilian species observed had habitat preference of vegetation/rocks/bricks/piles of leaves which were present around the banks of ponds. The occurrence of these species seemed to be because of availability of prey, shelter and undisturbed habitat in the present study. Rapid population declines of reptiles has evoked a concern worldwide to understand the ecological roles (Lips et al 2006, Connelly et al 2011, Bohm et al 2013). Current literature has pointed out data deficient representation of reptiles in worldwide research which has been accentuated by spatial bias in monitoring work (Piccolo et al 2020).

The diversity of the herpetofauna around village ponds requires urgent attention for their habitat preservation and should include assessment of extent of anthropogenic pressures at local levels in Punjab State. Different studies have pointed out that significant stressors affecting animal diversity in freshwater ponds are overexploitation of resources, water pollution, rate of water flow, degradation of habitat and invasion by alien species (Noges et al 2015, Soni et al 2019). Regional studies have pointed that pond management efforts are vital as water quality parameters of

Table 1. Different species of class Amphibia and Reptilia observed at selected ponds

Name of species	Summer			Monsoon			Winter		
	Pond A	Pond B	Pond C	Pond A	Pond B	Pond C	Pond A	Pond B	Pond C
Euphlyctis cyanophlyctis	+	+	-	+	+	-	-	-	-
Duttaphrynus stomaticus	-	+	-	-	+	-	-	-	-
Lygosoma punctata	+	+	-	+	+	-	-	-	-
Ptyas mucosa	-	+	-	-	+	-	-	-	-
Bungarus caeruleus	-	-	-	-	-	+	-	-	-
Varanus bengalensis	+	-	-	-	-	-	-	-	-

(+) Observed, (-) not observed

Table 2. Seasonal variation in water temperature, pH, BOD and DO in selected ponds

Location	Season	Water temperature (^oC)	pH	BOD (mg/l)	DO (mg/l)
Pond A	Summer	22.85	7.9	2.9	4.9
	Monsoon	24.35	7.5	0.4	5.2
	Winter	14.37	8.1	2.4	6.4
Pond B	Summer	22.7	7.4	2.8	4.8
	Monsoon	24.45	7.2	2	6.0
	Winter	14.12	7.9	2.6	6.8
Pond C	Summer	24.1	7.7	3.8	4.6
	Monsoon	25.7	7.4	1.2	5.2
	Winter	18.02	9	3.2	6.0

freshwater ponds are declining (Das and Dey 2020, Nandal et al 2020). Jackson et al (2016) observed that multiple factors tend to act antagonistically, and therefore their cumulative mean effect is less than the sum of their single mean effects in freshwater ecosystems. Assessment of faunal diversity like amphibians, reptiles and birds around fresh water lake might be used as an indicator of health of environment and provide baseline data for conservation of freshwater habitats (Jagatheeswari 2016, Sugumaran and Duraimurugan 2019). The significant contribution to regional biodiversity can be made by the irrigation ponds which can potentially sustain high taxonomic richness (Declerck et al 2006, Kawano et al 2006, Chester and Robson 2013, Kim et al 2016). With the worldwide recognition for the significant role of freshwater ecosystems like ponds, lakes and wetlands; current emphasis is to conserve these water bodies both for sustenance of overall diversity and ecosystem services (Jenkins et al 2015, Johnson et al 2016, Kaur et al 2020). At present, information on effects of agricultural intensification and its practices on occurrence,

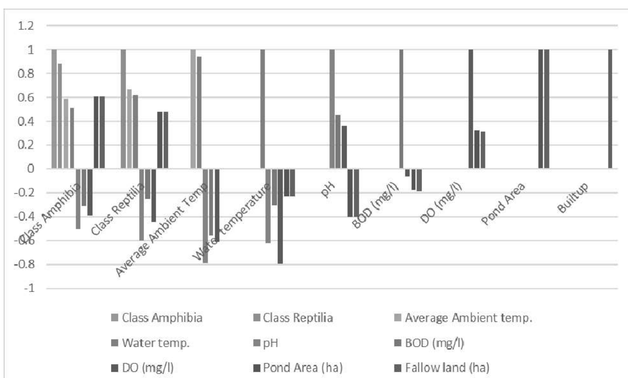

Fig. 1. Overall correlation analysis of ambient temperature, water quality parameters and habitat types with animal diversity at selected ponds

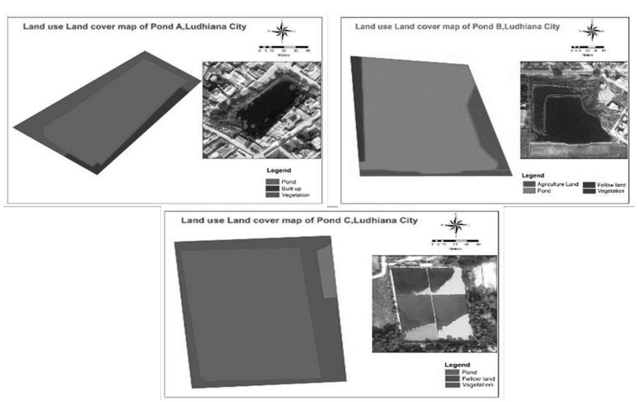

Fig. 2. Land use land cover map of Pond A, B and C in Ludhiana

diversity and populations of amphibians and reptiles at village ponds is limited and not well documented. To build up authentic data base, there is need of extensive and systematic field projects to record and identify herpetofauna in agro ecosystem. So that in future, conservation programmes may be formulated, implemented and become part of comprehensive in-vitro biodiversity preservation efforts.

CONCLUSION

Species richness of herpetofauna at selected ponds was location specific dependent on habitat features, anthropogenic stresses and water quality. Current findings indicate the need of well laid environment impact assessment programmes to recommend urgent interventions for freshwater pond habitat maintenance resulting in preservation of native herpetofauna in Punjab State.

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Study of Factors Influencing Fish Consumption Pattern in Punjab (India)

Rajinder Kaur, Grishma Tewari[1], Sarbjeet Kaur[1]* and Jitendra Kumar[2]

Krishi Vigyan Kendra, Barnala-148107, India
[1]Department of Fisheries Resource Management, College of Fisheries, GADVASU, Ludhiana-141004, India
[2]Fisheries Department, Government of Uttar Pradesh-226 007, India
**E-mail: dr.sarbjeet4@gmail.com*

Abstract: The present study aims to assess the fish consumption patterns and consumer preferences towards fish and fish-products in Punjab. The study was carried out in Punjab through a structured questionnaire to collect information about fish consumption patterns during 2021-22. A Google form created in English and Punjabi was circulated through e-mails, WhatsApp groups and various other social media platforms. The, total of 636 responses were recorded from rural (53%) and urban areas (47%) which revealed that 42.8% respondents were fish eaters while rest 57.2% as non-fish eaters. The major reason behind less consumption of fish and fish-products in Punjab was due to religious constraints (62.9 % of the non-fish eaters), followed by fishy flavour and odour (15.7%) and presence of spines (1.1%). Moreover, people have good knowledge about the facts of quality, spoilage, health benefits of fish but only 26.8% of fish eaters consider nutritional value of fish as selection criteria. Therefore, the findings of the present study signify the need to promote consumption of cheaper and easily digestible protein rich fish meat as well as to create awareness about nutritional benefits of eating fish in Punjab.

Keywords: Constraints, Fish consumption, Health benefits, Pattern, Punjab

Fisheries is the fastest growing animal food sector and it bridges the gap between the immensely increasing human population and nutritional security. Significant benefaction of fisheries sector is much clear and efficient towards the global hunger fight, achieve food security, and enhance nutrition as visualized in UN 2030 Agenda for Sustainable Development Goals **(**Bennet et al 2018**)**. Fish is considered as the cheapest source of animal protein and along with that, fish flesh commonly known as white meat have edible taste and flavour. While considering the nutritive value, fish is relatively superior than other animal protein consumed by human beings, in respect to its protein content/quality, easy digestibility, omega-3 fatty acids, minerals and vitamins. Therefore, it is advocated that fish should be part of human diet for various health benefits such as for better eyesight, controlling cardiovascular diseases as well as healthy skin and hair.

Worldwide, consumption of aquatic foods has reached about 20.2 kg per capita in 2020-more than double of the average consumption as compared to that of 1960s (9.9 kg per capita). Rising incomes and urbanization, betterment in the post-harvest techniques and modifications in the dietary habits have resulted in the projection of consumption of aquatic food by 15% increase to supply on average 21.4 kg per capita by 2030 (FAO 2022). Fish consumption preferences vary with topographical conditions, region, community etc. In India, Tripura ranks first in consuming the largest number of fish per capita (29 kg) annually (Deb 2021) whereas only 0.4 kg per capita in Punjab, despite of contributing a huge amount in the fish production (The Tribune 2022a). In Punjab, area of 43,691 acres is under fish farming, producing approximately 1,89,647 tonnes of fish but annual per capita fish consumption is very low, against the national per capita fish consumption (9 kg) (The Tribune 2022b). Keeping the above facts in view, an attempt was made to assess the factors influencing fish consumption patterns in Punjab.

MATERIAL AND METHODS

The study was carried out in Punjab (India) through a structured questionnaire to collect information about fish consumption patterns in the state from 2021-22. A Google form was created in English as well as Punjabi and circulated through e-mails, WhatsApp groups and various other social media platforms. The questionnaire included the respondents' personal details and different parameters of fish consumption pattern. For non-fish eaters, the form ends with reasons for not consuming fish and fish products whereas for fish-eaters, it continues further with different parameters including selection criteria, frequency to consume fish, facts about the health benefits of eating fish and others. The total sample size for consumer survey was 636, including the respondents from urban and rural areas. The collected information was compiled in the form of tabulating frequencies, percentages, and graphs (pie and bar graphs) for efficient representation and interpretation of data.

RESULTS AND DISCUSSION

An online survey was conducted by using structured questionnaire to assess the fish consumption patterns as well as the factors influencing fish consumption in Punjab. Total 636 responses were recorded from both rural and urban areas with a contribution of 53 % and 47 %, respectively, covering 22 districts of Punjab with maximum responses recorded from Ludhiana (30.5%). Among the total respondents, 56.9 % were male whereas 43.1 % were female. Basic information of respondents was also collected with respect to different age groups, education, occupation, family size and income status (Table 1). Out of total 636 responses, 42.8% were fish eaters whereas 57.2% do not like to consume fish which might be attributed to the consumer preference for chicken, mutton, pork etc. by meat eaters in Punjab (less than 40%) and majority of the fish consumers have the preference of spineless fish flesh. From the survey, it was observed that people from Punjab prefer to eat spineless fishes (41.2%) like - *Wallago attu* (Malhi), *Sperata seenghala* (Seenghara) *Pangasius* (Pangas) etc., in the form of fresh fish (33.8%), fish products (20.2%) and both (46%). They usually prefer to eat fish-products also like fried fish/curry (80.9%), followed by fish cutlets, fish fingers, fish balls, fish pickle, and fish sausage. The frequency of eating fish/fish-products is comparatively lower in Punjab as majority of the respondents include the fish/fish products in their diet once in a month (fresh fish - 55.5% whereas canned, dried, and frozen fish – 60.3%). The lower frequency of eating fish in Punjab, is mainly due to religious constraints (62.9 % of the non-fish eaters), followed by fishy flavour and odour (15.7%), presence of intramuscular spines (1.1%), higher price (0.8%), less availability in the area (0.6%). The 76.6% of the total respondents became fish eaters from vegetarian which could be attributed to their educational status and knowledge about the nutritional benefits of eating fish/fish products. The reviewing the present results indicated people used to gain the knowledge about the benefits of eating fish through various sources like fisheries education (31.3%), book/newspaper (23.5%), social media (23.2%), radio/TV & others (22.1).

Fish consumption patterns revealed that people of Punjab usually prefer to eat fresh fish and select on the basis of parameters like nutritional value (60.7%), taste (37.5%), availability in local area (1.5%), and cheaper price (0.4%). Normally buy the fish from local market (75.4%), super market (19.9%) and fish landing centres (4.8%) and prefer to prepare fish/fish-products at home (62.5%). Whereas they prefer to consume fish-products also and used to select fish-products over other animal meat products due to quality and taste (54.4%) followed by nutritional value (26.8%), availability in local area (15.1%) and affordable price (3.7%). People from Punjab are fond of eating spineless fishes (cat fishes like Malhi, Sanghara and Pangas) and do not prefer to eat carps due to presence of intramuscular spines. But carp culture is the backbone of aquaculture in the Punjab, producing a huge quantity of fish (1.51 lakh tonne during 2019-20), which is widely preferred by the migrants especially from Bihar, Uttar Pradesh, Andhra Pradesh, West Bengal, Madhya Pradesh, and others, as a constant food whereas only 25 % of native population (Punjabi) consumes fish, preferably as snacks **(**IWP 2016**)**. The consumers (Punjabi population) attitude towards fish and fish-products were assessed and was observed that fish is easily available (109%), comparatively cheaper source of animal protein than others (79%) and good for human health (148%) (Fig. 1). The 62.9 % of the non-fish consumer respondents do not consume fish due to religious constraints. Fish consumption preferences vary with topographical conditions, region, community etc. Sabat et al (2008) reported that major problems in fish consumption were varying supply chain, less availability of fresh fish, high price, and presence of bones in

Table 1. Basic information about the respondents (N = 636)

Age group	18-30 years	56.3%
	30-50 years	34.1%
	50-70 years	8.5%
	>70 years	1.1%
Education status	Primary	4.6%
	High school	22.8%
	Graduation	44.3%
	Post-graduation	28.3%
Occupation	Fish farmer	3.8%
	Animal husbandry and agriculture	3 % and 10%
	Govt. and private job	14% and 14.2%
	Self-employed	14.9%
	Unemployed	13.8%
	Students	26.3%
Family size (No.)	<4	26.3%
	4-8	69.3%
	>8	4.4%
Income /month	<Rs. 25,000/-	25.3%
	Rs. 25,000-50,000/-	32.1%
	Rs. 50,000-1,00,000/-	26.9%
	>Rs. 1,00,000/-	15.7%

fish meat in the northern India. The lack of awareness, lesser preference of fish meat over other animal protein and undeveloped taste of fish were the major constraints behind the lesser consumption of fish and its value-added products. Bhuyan et al (2017) revealed that among various reasons behind high consumption of fish in Assam were palatability of fish, higher nutritional content, and availability at cheaper price than other types of animal protein. Sanjeev et al (2021) reported that in Kerala, rate of consuming fish is very high in comparison to the national average whereas the fish consumption rate was far below than the Kerala figures in the tribes of Wayanad district, Kerala. Affordable price, local availability of preferred fish species were the major factors influencing the good procure amount and higher consumption of fish in the tribe. Majagi and Somashekar (2020) observed that income plays an important role with respect to preference of fish species (price of fish) to buy for consumption. Rahman and Islam (2020) reported that lower fish consumption is majorly due to lower income levels, followed by high prices and lack of knowledge about nutritional value of fish among people of Bangladesh. Budhathoki et al (2022) observed that knowledge and attitude towards the quality and safe consumption of aquatic food is as major factor, whereas price plays a central role.

Therefore, based on results, culture, and production of spineless fish like Pangas should be promoted in Punjab, as 90% of the aquaculture production is dependent on carp culture which is less preferred for consumption in the state,

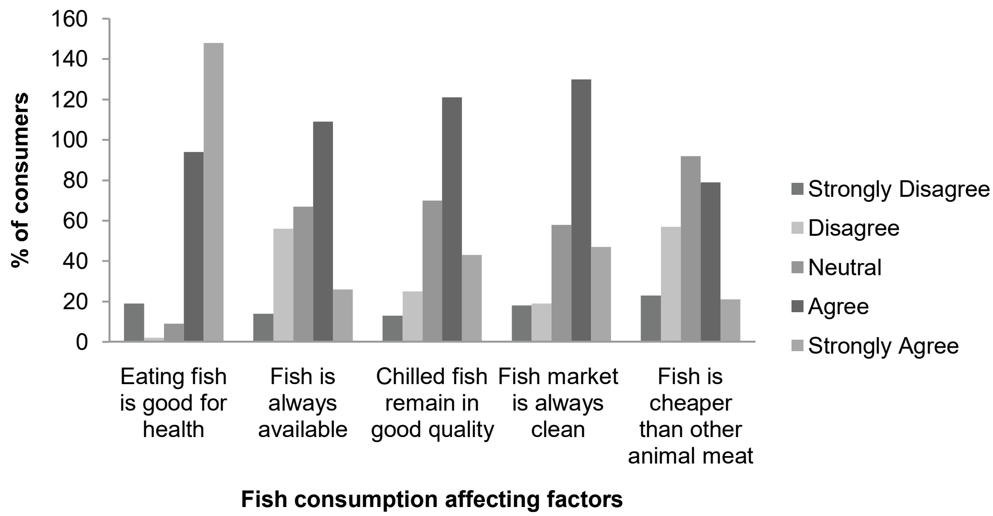

Fig. 1. Consumer's attitude towards eating fish and fish-products

Table 2. Major parameters showing fish consumption patterns in the Punjab

Parameter	Percent response
Preference of fish or fish products over other types of animal meat	Due to nutritional value (60.7%) Due to taste (37.5%)
Frequency of eating fresh or frozen fish	Once in a month (55.5% and 60.3%, respectively)
Frequency of eating fish products	
Form of fish consumption	Both fresh fish and fish products (46.0%)
Preferred fish-product	Fried fish (80.9%)
Criteria for selecting fish product	Quality and Taste (54.4%)
Preferred fish species	Cat fishes (41.2%)
Source of buying fish	Local market (75.4%) Super market (19.9%) Fish lending centres (4.8%)
Fish and fish-product prepared by consumers at home	Yes (62.5%) No (37.5%)
Any problem encountered with fish consumption	No (80.9%) Yes (9.2%) Not sure (9.9%)

Table 3. Knowledge of consumers about quality and nutritional value of fish

Parameter	Percent responses
Parameter to decide quality of fresh fish	• Smell (28.3%) • Skin colour (22.8%) • Skin texture (15.1%) • Gill colour (8.5%) • All of the above (60.7%) • Don't know (11.4%)
Fish as a good source of Omega-3	• Yes (91.2%) • No (2.2%) • Not Sure (6.6%)
Benefits of eating fish for human health	• Yes (86.4%) • Not sure (13.6%)
Reason for spoilage of fish	• Temperature (12.1%) • Moisture (1.5%) • Microorganisms (18.4%) • All of the above (57.7%) • Don't know (10.3%)

whereas Pangas is imported from other states (Andhra Pradesh and West Bengal) for sale in the Punjab.

CONCLUSIONS

The present study assessed the factors influencing fish consumption patterns as well as consumer preferences towards fish and fish products in Punjab. The people of Punjab prefer spineless fish and fish-products depending upon their taste and quality. Lower frequency of fish consumption is recorded which is mainly due to religious constraints. The study indicated lower per capita fish consumption which further provokes the need to create awareness about the nutritional benefits of consuming fish and to promote fish consumption.

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Screening of Seaweeds for Antibacterial and Antifungal Activities

Siddhnath and Sarbjeet Kaur[1]

Department of Fish Processing Technology, College of Fisheries,[1] Department of Fisheries Resource Management
College of Fisheries, GADVASU, Ludhiana-141 004, India
E-mail: kumarsiddhnath@gmail.com

***Abstract*:** Commonly occurring seaweeds, *Caulerpa scapelliforms, Ulva lactuca*, *Padina tetrastromatica, Steochspermum marginatum* and *Acanthophora spicifera* collected from the Tuticorin coast, Tamil Nadu (India) were evaluated for antibacterial and antifungal activity by agar diffusion method. Four different solvents *viz.* petroleum ether, benzene, chloroform, and methanol wereused for extraction. By using commercial medicine, Amikacin-AK30-30mcg/disc, Cephalexin-CN30-30mcg/disc, Ciprofloxacin-CIP5-5mcg/disc and Fluconazole-FLC25-25mcg/disc as control and zones of inhibition were compared. The seaweed extracted using petroleum ether as a solvent showed the best antifungal activity and was more effective than commercial antibiotic, fluconazole. None of the extracts were active against *Staphylococcus aureus*, a gram-positive bacterial pathogen. The methanol extract was active against *Salmonella typhi, Pseudomonas aeruginosa, Proteus vulgaris,* and *Escherichia coli* except for *Caulerpa scapelliforms* and *Ulva lactuca*. All the four extracts of *Acanthophora spicifera* showed antibacterial activity.

***Keywords*:** Seaweeds, Antibacterial, Antifungal, Solvents

The marine environment has a rich biological and chemical diversity (Dayanidhi et al 2021). This diversity of marine organisms and habitat offers a wide variety of natural marine compounds. Several oceanic compounds show pharmacological activities and are helpful in the invention and discovery of novel bioactive compounds, primarily for deadly diseases like Cancer, Acquired immunodeficiency syndrome (AIDS) and Arthritis (Samuel et al 2011, Pham-Huy and Huy 2022). Many other compounds have been developed as analgesics or to treat inflammation (Giriwono et al 2019). Seaweed (macroalgae), a marine organismis widely distributed in the coastal regions of all the continents. Several scientists have worked on the seaweed extracts, exhibiting antibacterial and antifungal activity (Hellio et al 2000, Lima-Filho et al 2002, Taskin et al 2007, Seenivasan et al 2010). Moreover, crude extracts of Indian seaweeds were found to be active only against Gram-positive bacteria whereas, majority of the pathogenic bacteria are gram-negative. Therefore, keeping this in view, the present work was designed to study the antibacterial and antifungal activity of four different solvents extracts of five seaweeds collected from Tuticorin coast, Tamil Nadu.

MATERIAL AND METHODS

Collection and preparation of selected seaweeds: Fresh seaweed samples of *Caulerpa scalpelliformis, Ulva lactuca, Padina tetrastromatica, Stoechospermum marginatum* and *Acanthophora spicifera* were handpicked from the intertidal zone of Hare Island, Tuticorin (TN) located at 8.8°N, 78.3°E The collected seaweed samples were cleaned with seawater and freshwater, dried in the shade, and powdered to extract antimicrobial compounds.

Preparation of organic seaweed extracts: The organic seaweeds extracts were prepared by following the method of El Shafay et al (2016). Powdered seaweed sample (5g) were soakedin 50 mL of four different solvents (analytical grade) i.e. petroleum ether, benzene, chloroform and methanol for three days. Then soaked samples were filtered and concentrated in a rotary evaporator at 35^{0}C. The residual water was then removed with the help of vacuum pump (Agilent vacuum pump PVL401), followed by suspension of weighted crude extract in dimethyl sulfoxide (DMSO) to the final concentration of50 mg/mL and stored in a refrigerator.

Bioassay: For bioassay, fungal pathogen *Candida albicans* ATCC 90028 and five gram-negative bacterial pathogens namely *Salmonella typhi* ATCC 35640, *Pseudomonas aeruginosa* ATCC27853, *Proteus vulgaris* ATCC 33420, *Klebsiella pneumoniae* ATCC 700603, *Escherichia coli* ATCC 25922 and one pathogenic gram-positive bacteria namely *Staphylococcus aureus* ATCC 25923 were used. A loop full of the microorganism was inoculated in nutrient broth from the 24 hours incubated nutrient agar slant of each test organism at pH-7.4 to activate the bacterial strains used as test organisms. The broths were kept for incubation at 37°C for 24 hours so that the microorganism can grow till the log phase. The nutrient broth was maintained as control without inoculating the test organisms. These bacteria were obtained from Hi-Media Ltd., Mumbai. Amikacin-AK30-30mcg/disc, Cephalexin-CN30-30mcg/disc, Ciprofloxacin-CIP5-5mcg/disc and Fluconazole-FLC25-25mcg/disc were usedas control medicine. The bioassay was carried out under a sterile condition in a clinical laboratoryby following the agar diffusion method as described by Perez et al (1990). Inoculums in the exponential phase of growth, equivalent to a 0.5 McFarland standard were swabbed on to the surface of Muller Hinton agar media. Different crude extract (2 mL) was transferred to a sterile 6 mm Whatman No.1. Filter paper disc (E-760) and after drying it was placed in the seeded agar plate, along with the control discs which was prepared with the solvents alone. The inhibition zone was observed after 24 hours of incubation at 37°C.

RESULTS AND DISCUSSION

Among five different seaweeds (*Caulerpa scalpelliformis, Ulva lactuca, Padina tetrastromatica, Stoechospermum marginatum* and *Acanthophora spicifera*) collected, the extracts of all seaweeds except *Caulerpa scalpelliformis* showed specific activity against the pathogens (bacteria i.e. *Salmonella typhi, Pseudomonas aeruginosa, Proteus vulgaris, Klebsiella pneumoniae* and *Escherichia coli* and fungi i.e. *Candida albicans)* (Table 1, 2). The present study showed that petroleum ether extract of *Ulva lactuca*is active against fungal pathogen, *Candida albicans,* in accordance with the findings of Oranday et al (2004). Further, the petroleum ether extract of *Caulerpa scalpelliformis*, *Ulva lactuca,* and *Padina tetrastromatica*; the benzene extract of *Stoechospermum marginatum* were active against fungal pathogen *Candida albicans*. The different extracts of the red alga, *Acanthophora spicifera* were not found active against fungal pathogen, *Candida albicans*. The zone of inhibition indicates that extracts of all the seaweeds exhibit antifungal activities except *Acanthophora spicifera* which are quite comparable with the commercial antifungal agent i.e. fluconazole (Table 2). The petroleum ether extract of *Caulerpa scalpelliformis* was more effective (32mm) than the commercial antifungal agent fluconazole (27mm). Similarly, Kaur et al (2016) observed *Inula racemosa* root extract were active against phyto-pathogenic fungi (*Dreschlera oryzae*). Tuney et al (2006) found that the methanol extract of *Ulva rigida* collected from the coast of Urla (Izmir, Turkey) hadno antifungal activity against *Candida albicans*. Karthick et al (2014) reported that *Caulerpa scalpelliformis* contains

Table 1. Antibacterial activity of sea weeds extracts from Hare Island of Tuticorin coast

Name of the bacterial pathogens	Control medicine			*Caulerpa scalpelliformis*				*Ulva lactuca*				*Acanthophora spicifera*				*Padina tetrastromatica*				*Stoechospermum marginatum*			
	AK30	CN30	CIP5	PE	BZ	CF	MN	PE	BZ	CF	MN	PE	BZ	CF	MN	PE	BZ	CF	MN	PE	BZ	CF	MN
Salmonella typhi ATCC 35640	16	20	21	-	-	-	-	-	-	-	-	15	-	-	-	-	-	-	-	-	-	-	16
Pseudomonas aeruginosa ATCC 27853	14	15	14	-	-	-	-	-	-	-	-	-	-	-	14	17	-	-	14	-	14	-	-
Proteus vulgaris ATCC 33420	16	20	21	-	-	-	-	16	-	-	-	-	-	-	-	-	-	-	15	-	-	-	-
Klebsiella pneumoniae ATCC700603	26	29	30	-	-	-	-	-	-	23	-	-	20	24	-	-	-	-	-	-	-	-	-
Escherichia coli ATCC 25922	16	20	22	-	-	-	-	-	-	-	-	-	-	-	16	-	-	-	-	-	-	-	16

*PE-Petroleum ether; BZ-Benzene; CF-Chloroform; MN-Methanol; FLC25-Fluconazole; AK30-Amikacin; CN30-Cephalaxine; CIP5-Ciprofloxacin
*Zone in mm indicates the distance from the border of the disc to the edge of the clear zone.
(-): hyphen indicates no clear zone has been observed

Table 2. Antifungal activities of sea weeds extracts from Hare Island of Tuticorin coast

	Control medicine	*Caulerpa scalpelliformis*				*Ulva lactuca*				*Acanthophora spicifera*				*Padina tetrastromatica*				*Stoechospermum marginatum*			
	FLC25	PE	BZ	CF	MN	PE	BZ	CF	MN	PE	BZ	CF	MN	PE	BZ	CF	MN	PE	BZ	CF	MN
Candida albicans ATCC90028	27	32	-	-	-	24	-	-	-	-	-	-	-	22	-	-	-	-	22	-	-

See Table 1 for details

tannin, flavonoids, glycosides, phenols, saponins, and terpenoids. This indicated that in the particular case that petroleum ether could be able to successfully extract antifungal compounds in an effective concentration.

The petroleum ether extracts of *Acanthophora spicifera*was sensitive against Gram-negative bacterial pathogen *Salmonella typhi* but comparatively lower than the commercial antibiotic agents i.e. Amikacin, Cephalexin and Ciprofloxacin. The methanol extract of *Stoechospermum marginatum* was active against Gram-negative bacterial pathogen *Salmonella typhi.* The methanol extract of a *Padina tetrastromatica* and benzene extract of *Stoechospermum marginatum* showed similar activities like Ciprofloxacin and Amikacin against Gram-negative bacterial pathogen *Pseudomonas aeruginosa*. However, petroleum ether extracts of *Padina tetrastromatica* was more effective (17 mm) than commercial antibiotics. The methanol extract of *Ulva lactuca* showed antibacterial activity against Gram-positive bacterium *Staphylococcus aureus* and Gram-negative bacterium *Escherichia coli* (Selvi et al 2001, Oranday et al 2004). The methanol extract of *Ulva lactuca* did not show such antibacterial activity against all these pathogenic bacteria whereas petroleum ether extract of *Ulva lactuca* was active against Gram-negative bacterial pathogen *Proteus vulgaris*. This could be attributed to the presence of lipophilic and phenolic compounds, especially steroids fatty acids, in *Ulva lactuca* organic extract towards antimicrobial activity (El-Baky et al 2008). The zone of inhibition (16 mm) indicates that the methanol extract of *Padina tetrastromatica* was similarly active against *Escherichia coli* as commercial antibiotic i.e. Amikacin. The chloroform extract of *Ulva lactuca* and *Acanthophora spicifera* and the benzene extract of *Acanthophora spicifera* showed lesser activity than needed against Gram-negative bacterial pathogen *Klebsiella pneumoniae*. The methanol extract of *Stoechospermum marginatum* and *Acanthophora spicifera* were active against Gram-negative bacterial pathogen *Escherichia coli*. All the extracts studied were inactive against the Gram-positive bacterial pathogen *Staphylococcus aureus* which might be attributed to the inhibited entry of antimicrobial agents into cell due to the presence of high percentage (90-95%) peptidoglycan, lipopolysaccharides and phospholipids in the bacterial cell wall. Marke et al (2022) also tested *Tinospora cordifolia* (natural herbal shrub) extracts for assessing antibacterial activity and suggested that phytochemical compounds present in this natural herbal shrub can be utilized for formulation of drugs against some gastrointestinal pathogens like *Staphylococcus aureus, Escherichia coli, Vibrio cholera, Salmonella typhi* and *Shigella*.

During the present study, different extracts of *Caulerpa scalpelliformis* were inactive against all the studied pathogenic bacteria whereas Selvi et al (2001) reported that the methanol extract of *Caulerpa scalpelliformis* showed antibacterial activity against Gram-positive bacterium *Staphylococcus aureus* and Gram-negative bacterium *Salmonella typhi, Pseudomonas aeruginosa, Klebsiella pneumoniae* and *Escherichia coli*. Non-conformance of the findings might be due to different natural factors such as environmental conditions (light, temperature or salinity), reproductive state, age of the seaweed, geographic location and seasonality (Perez et al 2016). Hence, information made during the present study revealed that seaweed extracts with different solvents exhibit antibacterial and antifungal properties which could be further utilized forproduction of organic pharmaceutical products. Moreover, this created a quest also for the discovery of antibiotic compounds of natural origin.

CONCLUSION

Seaweeds have huge potential for discovery of compounds with various bioactivities. Seaweeds collected from the coastal areas of Tuticorin exhibit antibacterial and antifungal properties. The test microorganism showed resistance towards seaweed extracts prepared with different organic solvents, which is indicating the availability of biologically active compounds at altered concentrations in seaweeds. The study is signifying the need to explore the possibilities of having bioactive compounds in seaweeds with pharmaceutical importance.

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Utilization of Resources by Integrating Fish and Poultry Farming- A Case Study

R. Kaur and P.S. Tanwar

Krishi Vigyan Kendra, Barnala-148 107, India
Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana-141 004, Punjab
E-mail: rajindergoraya7@gmail.com

Abstract: The integration of aquaculture with livestock or crop farming provides quality protein food, resource utilisation, recycling of farm waste, employment generation and economic development. On farm waste recycling, an important component of integrated fish farming is highly advantageous to the farmers as it improves the economy of production and decrease the adverse environmental impact of farming. Aiming to evaluate the utility and efficacy of integrated fish and poultry farming for self-employment and nutritional security purpose, the integrated model of Fish cum Poultry Farming in 1 hectare (ha) fish-pond with composite carp culture and rearing 1000 White Leghorn poultry birds in a shed constructed (24 x 36 ft) over the fish pond was studied in 2022. From the conducted study, it was assessed that farmer is earning a net profit of approximately 6.78 lakh rupees annually from 1 ha of integrated fish and poultry farming. Hence, the results prove the efficacy of integrated fish cum poultry farming as a profitable venture for the farmers and in addition, could address the issues of sustainability and livelihood security effectively.

Keywords: Environment friendly, Fish, Integrated farming, Poultry, Profitability

Aquaculture is a farming system which can go a long way in augmenting animal protein production and generating gainful employment. The modern fish culture technologies have immense potential for increasing fish production, but they push up the prime cost due to higher input cost, intensive use of protein rich quality feed and mineral fertilizers etc (Bhagawati and Tamuli 2020). Substituting the tradition farming practice with integrated farming system can be a profitable approach that involves crop cultivation, dairy, poultry, fishery, vegetable and fruit production etc. together for higher efficiency. Integration of different enterprises/crop by utilizing farm's available resources is one of the best multidisciplinary approaches to reduce the input cost and enhancing the farmers' income (Singh et al 2020). For the betterment and upliftment of farming community, Government of India is campaigning the flagship programme for “doubling farmer's income (DFI) by 2022” and in this regards, many interventions have been suggested including IFS for ensuring higher farm productivity and profitability for sustainable economic growth for farming communities in India (DFI 2017). Out of all the integrated farming systems, fish cum poultry is the most trending one (Sharma et al 2016). Fish cum livestock farming is considered as an excellent innovation for judicious recycling of organic waste and optimum production of high-class protein at low cost (Ayyappan et al 2011). This is also one of the best methods of both waste disposal and waste utilization (Bhagawati and Tamuli 2020). Integrated farming has immense potential to emerge out as an effective tool for the improvement of rural economy due to low investment and high profitability (Nanda and Bandopadhyay 2011).

In aquaculture, formulated feed costs about 60-70 % of the total production cost and the use of animal manure can considerably reduce the operational costs and make it possible for low to medium income fish farmers to profitably engage in the enterprise (Nath et al 2020). Direct use of livestock wastes is one of the most widespread and conventionally accepted forms of integrated fish farming. This practice increases the efficiency of both livestock and fish culture through the profitable utilization of animal and feed waste products. About 80 per cent of the chicken dropping represents undigested food stuffs due to very short digestive tract of chicken Sharma et al 2016). Under the fish cum poultry integrated farming system, nutrients from poultry waste gets recycled in the pond that allows for escalation of production and income while reducing the effluents along with the dumping of the wastes would have bad impact on the environment (Singh et al 2014, Misra et al 2016). Hence, in this study, the economics of integrated fish cum poultry farming system was assessed.

MATERIAL AND METHODS

Farm location: The study was conducted in village Alkara (30.5003° N, 75.2960° E) of district Barnala (Punjab), on integrated fish cum poultry farming model (Fig. 2). This case study involves comprehensive assessment of a social microscope comprising person, group, social institution, district, or community (Burgess 1993, Young 1996). Primary data for the study was collected from the farmer through interview and farm visit.

Pond management: In the fish-pond, 6 species of carps including IMCs and CMCs (*Catla, Labeo rohita, Cirrhinus mrigala, Hypophthalmichthys molitrix, Ctenopharyngodon idella, Cyprinus carpio*) were stocked with 10,500 individuals of size ranging from 50-100g. Fish was stocked in the ratio of 3:4:3 (surface feeder : column feeder : bottom feeder) @ catla (2000), silver carp (1150); rohu (2500), grass carp (1700); mrigala (2000), common carp (1150) following the composite carp culture practice for aquaculture. For maintaining water quality, a four paddle-wheel aerator is installed in the pond for aeration, operated daily at the dawn or as per necessity. Addition of fresh water, pH evaluation (using digital pH meter) are the routine practices over there. Pond water is used by the farmer for irrigating fruits and vegetables grown at the pond dyke. To prevent the entry of piscivorous birds, nylon thread is used to cover the pond. A mannequin is installed at the pond dyke for prevention of the same. Application of potassium permanganate (bimonthly) and CIFAX (thrice a year during Feb/March, June/July and October/November) is done to reduce the chances of disease occurrence in fish pond.

Livestock Rearing

Housing of birds: Fully vaccinated 1000 White Leghorn birds is reared in a concrete poultry shed constructed over the pond with concrete pillars. The poultry birds are kept under intensive system (Fig. 1) and are confined entirely to the house and 100-110 g of feed per bird is provided daily. The poultry eggs are collected twice a day.

Horticulture: For utilising the area of pond dyke, he planted seasonal vegetables such as carrot, radish, turnip, spinach, capsicum, green pepper, bottle gourd etc. for household consumption. Fruit like Guava, Kinnow, Indian blackberry

Fig. 1. Poultry shed constructed over fish pond

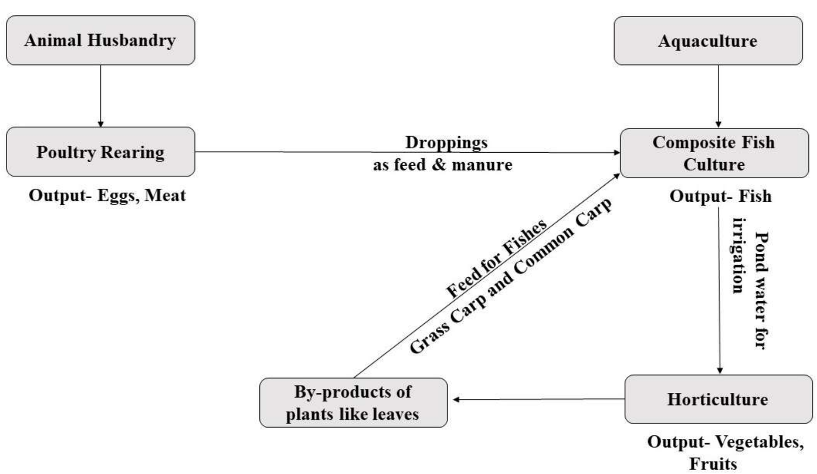

Fig. 2. Flow diagram of the studied integrated farming system model

(Jamun) are also grown near the pond dykes. They are irrigated with pond water and are organically grown without the use of additional fertilizers.

RESULTS AND DISCUSSION

Economic analysis of integrated fish cum poultry farming was made to evaluate the sustainability of the existing integrated farm (includes year-round variable cost and net profit). With absolute dependency on poultry droppings, the total fish production from one ha fish pond was 6.3 tonnes (average production of 2.52 tonnes in 0.4 ha). The percentage return on total expense was 49.67 % in the present study and the average annual net income was Rs. 6.78 lakh rupees with an average of Rs. 56,500.00 per month (Table 1). Robiul et al (2009) also observed higher growth rate and fish production in integration with poultry farming. Bhuiyal et al (2014) documented that the integrated farming system has the potential to improve the efficiency of small and marginal farmers for enhancing their production and income. Sharma et al (2016) conducted a study to evaluate the performance of fish cum poultry cum horticulture integrated farms in comparison to non-integrated fish farms in district Udham Singh Nagar, Uttarakhand. The overall results of the study revealed that the integrated pond management with fish, poultry and horticulture was an excellent approach for sustainable production, income generation and employment opportunity of the resource poor rural households. Misra et al (2019) assessed the utility of integrated fish cum poultry farming system in Arunachal Pradesh for self-employment and observed high cost-benefit

Table 1. Economics of fish cum poultry integrated farming model (One ha)

Parameter	Unit amount	Cost (in Rs.)
1. Expenditure		
A. Fixed Cost		
Pond excavation		3,00,000.00
Poultry shed		80,000.00
Total fixed cost		3,80,000.00
B. Variable cost		
II. Fish pond		
Fish fingerling	12000	36,000.00
Supplementary feed	-	-
Medicines, electricity and miscellaneous	50,000	50,000.00
Labour	Rs. 12,000 per month (Full time)	1,44,000.00
Sub total (I)		2,30,000.00
III. Poultry unit		
Poultry birds	1000 (Rs. 300/bird)	3,00,000.00
Feed	1 Quintal per day	7,40,000.00
Sub total (II)		10,40,000.00
Total variable cost (I+II)		12,70,000.00
C. Total cost		
Variable cost		12,70,000.00
Depreciation on fixed capital @10% per year		38,000.00
Interest on fixed capital @15% per year		57,000.00
Grand total		13,65,000.00
IV. Gross income		
Fish harvesting	6.3 tonnes (Rs. 110 per Kg)	6,93,000.00
Eggs	Rs. 180 per tray	13,00,000.00
Meat	Rs. 50 per bird	50,000.00
Total Income		20,43,000.00
V. Net income (Gross income – Total cost)		6,78,000.00
Average income per month		Rs. 56,500.00

ratio in the integrated system as compared to traditional farmer's practice. The study suggested that integrated fish and poultry farming system is a viable option to increase the income of small and marginal farmers.

CONCLUSION

Case study shows the success of fish cum poultry Integrated Farming System (IFS) model of the farmer with zero waste generation. IFS model is a profitable venture for the farmers to earn more profit in comparison to traditional farming. It can also be easily adopted by young people for income and self-employment generation.

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Length-Weight Relationship and Condition Factor of *Mastacembelus armatus* (Lacepède 1800) from Burhi Gandak River, North Bihar, India

Sahil, H.S. Mogalekar*, Sanjay, Jham Lal and Dheeraj Kashyap

College of Fisheries, Dr. Rajendra Prasad Central Agricultural University, Muzaffarpur-843 121, India
E-mail: frmcof14@gmail.com

Abstract: The study has been conducted to investigate the length-weight relationship and Fulton's condition factor (Kn) of the freshwater spiny eel, *Mastacembalus armatus* (Lacepede 1800) from the river Burhi Gandak. A total 300 specimens of *M. armatus* were collected during July 2019 and June 2020. The observed length of *M. armatus* was ranged from 6 cm to 58.4 cm and weight from 3 to 654g. The species showed negative allometric growth pattern as depicted by the estimated b value (growth coefficient) i.e., 2.52 and the Fulton's condition factor (Kn) as 0.29±0.15. Highly significant results were reported for all LWR regression parameters. These findings will serve as a foundation for the sustainable conservation and management of the fishery resources in Burhi Gandak River, North Bihar, India.

Keywords: Burhi Gandak, Length-weight relationship, Condition factor, *Mastacembelus armatus,* Bihar

The representative fishes of family Mastacembelidae is named spiny eels due to the presence of spines running along the back of the fish. *Mastacembelus armatus* is found in India, Pakistan, Bangladesh, Sri Lanka, Myanmar, Nepal, Thailand, Indonesia, Malaysia and southern China (Froese and Pauly 2023). Moreover, this species is one of the potential candidates in ornamental fisheries of Bihar (Raut et al 2020). Bihar is an entirely landlocked state in a subtropical region of the India, being in 7th place with a contribution of 6.41 lakh tonnes and estimated human consumption of 8.82 capita/kg. Burhi Gandak, a tributary of the Ganga river has given rise to enormous resources like Mauns, Chaurs and Pokhar in the northern part of Bihar. The river Burhi Gandak is originated from central Himalaya with its basin stretching over Bihar state, has vast underutilized fisheries resources that produce an enormous diversity of species (Singh et al 2018). The icthyofaunal diversity of the river Burhi Gandak is dominated by the order Cypriniformes (Sahil 2020). *M. armatus* fetches higher market (Rs. 300/ Kg) as food fishes in local markets of North Bihar especially when sold alive. The length-weight relationships (LWR) of fish are significant in fisheries and fish biology (Mir et al 2012). Including other morphometric traits, the LWR can be used to distinguish taxonomic groups, and to depict other life events like metamorphosis, growth, and the beginning of maturity changes over time (Thomas et al 2003). LWR can also be used to set yield equations, which can be used to calculate the number of fish landed and compare the population across time and space. Fisheries researchers can apply the LWR parameters (a and b), to estimate a fish's weight from its length, to compute condition indices, to compare the life histories and morphologies of populations from various region (Sani et al 2010), and to investigate ontogeny allometric changes. In studies on fisheries and fish biology, Fulton's condition factor (K) is frequently utilized. In order to describe the "condition" of a certain fish, this component is derived using the relationship between a fish's weight and length (Froese 2006). The condition of sexual development, the availability of food sources, the age, and the sex of some species are all indicated by a fish's K values. However, no work has been reported so far from this region, henceforth, the present investigation has been taken up to provide baseline data for further studies, management and conservation of *M. armatus*.

MATERIAL AND METHODS

The present investigation was conducted from July 2019 to June 2020 in Burhi Gandak river to study the length weight relationship of *M. armatus*. Fish samples were randomly collected representing various classes of lengths on monthly intervals from Pilkhi Ghat (26°02'20.7"N 85°33'30.8"E), Dholi (26°00'08.0"N 85°36'23.0"E) and Pusa (25°59'48.7"N 85°39'39.0"E) sampling points in the Muzaffarpur and Samastipur using traditional gears like bamboo made traps, Ghanas and nets with floats and sinkers attached for the gear stability. The collected fishes were cleaned with water and morphometric and meristic characters were observed for the identification by using the standard taxonomical keys (Talwar and Jhingran 1991; Jayaram 1981). Length and weight measurements were taken to the nearest 1.0 mm and 0.01g using verniar caliper and weighing balance respectively for analysis. The taxonomic identification of collected specimens were done in fish biology laboratory of College of Fisheries, Dholi. The length-weight relationship was estimated using formula $W=aL^b$ by Le Cren (1951). Logarithmically: LogW = log a+b log L, where L is the total length of mussels, W is the total body weight of mussels, "a" and "b" is the intercept of regression curve and regression coefficient respectively (Froese 2006). Fish condition, fatness, or well-being are compared using the condition factor on the grounds that heavier fish of a given length are in better condition (Mir et al 2012). Using Fulton's formula, the coefficient of condition K was determined (Fulton's 1904).

$$K=100 \times W/L^3$$

Where, K=condition factor, W= mean weight of fish (gm), L= mean length of fish (mm)

RESULTS AND DISCUSSION

The length of *M. armatus* ranged from 6 cm to maximum being 58.4 cm whereas weight ranged from 3g to 654g. The estimated b value for the *M. armatus* is 2.52 which lies within the expected range of 2.5 to 3.5. Hossain et al (2006) observed that *M. aculeatus* collected from the Mathabhanga River in southwest Bangladesh had a negative allometric growth trend, which is consistent with the findings of our study. However, also claimed that *Macrongathus pancalus* from the Mathabhanga River in southwest Bangladesh had experienced positive allometric growth. This contradicts the findings of this study and could be explained by seasonal variation, the number of specimens observed, variations in the observed length ranges, habitat, and the level of fishing pressure. Abujam and Biswas (2016) observed seasonal variations in *M. pancalus* positive allometric growth (b>3) and negative allometric growth (b<3) in upper Assam, India. The majority of fishes have b values between 2.7 and 3.3. (Abdallah 2002). During the present study b values are consistent with those of other investigations (Sarkar et al 2008; Mir et al 2012). Sani et al (2010) examined LWR for a variety of freshwater species from the Gomti and Betwa rivers in Uttar Pradesh, including *Gudusia. chapra, Wallago attu, Sperata. aor, Sperata. seenghala, Clupea. gurua,* Mastacembelus *armatus, and Puntius soph*ore and observed similar trend. Khan et al (2012) revealed the isometric growth 2.5-3.5 of *Heteropneustis fossilis* and *Channa marulius* from the Ganga river basin. The length weight relationship as represented indicates negative allometric growth pattern for this species, lower value (<3) signifies as the length of the fish grows the body becomes slimmer (Fig. 1). Based on their genetic makeup, the actual length-weight connection differs from species to species.

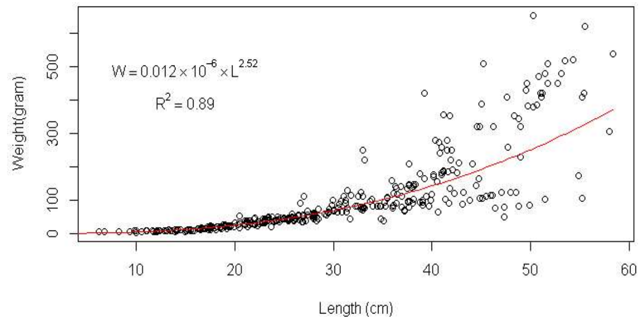

Fig. 1. Length weight relationship of *Mastacembelus armatus*

The mean value of fulton condition factor (Kn) is 0.29±0.15 while the mean modified condition factor (K) was 1.38 which may be attributed to different environmental conditions of river. In all size groups of males and females, the values of Kn fluctuate. In the current study's analysis of the length-weight connection and relative condition factor in *M. armatus*, the growth rate is quite excellent.

CONCLUSION

The research provides crucial data on length-weight relationships of Mastacembelus armatus, aiding sustainable fishery management and biodiversity conservation in Indian rivers. Additionally, it offers baseline information for an online fish database to support the conservation of lesser-known freshwater eels in North India.

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Development of Handloom Union Fabric from Ecofriendly Sisal Fibre and Characterization

Bhawna Soun, Harminder Kaur Saini and K. Brar
Department of Apparel and Textile Science, College of Community Science
PAU, Ludhiana-141 004, India
E-mail: bhawnasoun94@gmail.com

Abstract: Sisal, an environmentally friendly fibre, is one of the most commonly used natural fibre because it is cheaper, poses no health hazards, and provides a solution to environmental pollution. The present study investigates the development of eco-friendly handloom union fabric developed from sisal/cotton blended yarns and the effect of weave structure on the physio-mechanical properties of sisal/cotton union fabrics. Three different weaves, viz., plain, basket, and horizontal zig-zag twill, were prepared in handloom by using the 2/20 count cotton yarn as warp and the 12s count sisal/cotton (35/65) blended yarn as weft. The results inferred that handloom fabrics are more eco-friendly than power loom fabrics. Among all the three weaves the mechanical properties of the basket weave were found to be superior as compared to the plain weave and basket weaves. Basketweave had maximum bending length in the weft direction, which means it is stiffer than the other two weaves.

Keywords: Physio-mechanical, Union fabrics, Ecofriendly and horizontal zig-zag twill

Sisal fiber is one of the most widely used natural fibers and is very easy to grow. It has rapid renewal cycles and grows unchecked in the field and railroad track hedges. It occupies the 6th place among plant fibres, which represent 2% of the world's production of plant fibres (Nayak et al 2011). In India estimated area of sisal cultivation is about 50 thousand hectares, as compared to 352 lakh hectares in world area of cultivation (Basu et al 2012). It was introduced in India during the 15th century by the Portuguese. Indian species of Agave/rambansare *A. mexicana, A. veracruz, A.cantala, A. sisalana* and *A.americana*. In India, sisal is grown primarily in Andhra Pradesh, Bihar, Orissa, Karnataka, Maharashtra, Chhattisgarh, Uttrakhand and West Bengal (Shroff and Karolia 2015). Sisal fibers are cheaper, pose no health hazards, and also provide a solution to environmental pollution (Zwane et al. 2019). These versatile plants can survive in various climatic regions that include semi-arid, arid and humid regions, and all types of soil with minimal cost of cultivation.

Sisal plantation is carried out for utilizing the soil which is neither good for agriculture nor tree plantation as it does not produce any pesticide load to the environment. It is not infested by disease and insect pest; and therefore, it helps in reducing soil erosion through its extensive root system and contributes positively to watershed management. It has several distinguishing characteristics which makes sisal a 'specialty crop' for conservation agriculture (Sarkar and Jha 2017). Unlike other fibres, sisal is not a seasonal crop. It is drought resistant and can grow in a variety of soils. Currently sisal is found on embankments, bunds and roadsides, serving the purpose of soil conservation and protection as hedge plantation (Alapati and Shaik 2017).The most elegant and dignified fabric is woven fabric, which is the most adaptable due to the way it is composed of yarns in the shape of thick or thin sheets. It is possible to create a variety of designs, including plain, twill, basket weaves and variations like herringbone, diamond, horizontal zig-zag twill etc., due to the variation in interlacement. The physio-comfort characteristics of woven materials are somewhat impacted by these weave differences (Baruah et.al 2022). Different types of yarns are used in both the warp and weft directions of the fabric to create union textiles. Union fabric exhibits excellent strength, enhanced crease resistance, greater moisture absorption, high luster, and other desirable qualities. In order to lower the expensive price of textiles as well as the weight of the fabric, a variety of union fabrics may be made by combining several types of yarns, such as silk with cotton, ramie, rayon, polyester, sisal acrylic (Nayak et al 2009). Sisal has historically been the most used material for agricultural twine due to its strength, longevity, and versatility, stretchability, a fondness for specific dye substances, and saltwater resistance. Sisal fibre will be useful in the growing market for eco-friendly textiles. However, because it is a harsh fibre, blending it with other fibres is necessary to provide the finished fabric more increased qualities like comfort, flexibility, drapability (Agrawal et al 2018). Cotton fibre was used for blending in order to fulfil these increased qualities. The handloom sector benefits from flexibility in small production volumes, innovation-friendliness, minimal investment, labour-intensive production, and market-requirement adaptability. Despite these benefits, the mechanisation, modernization, and sophistication of the textile industry have made the handloom sector a sunset industry. With the exception of India, Sri Lanka, Bangladesh, Thailand, and Cambodia, handloom textiles have lost their appeal and market over the past century.

MATERIAL AND METHODS

Location: The study was conducted in the department of Apparel and textile Science, PAU, Ludhiana and the handloom weaving was done at Naresh Handloom, VPO Jeevanpur, Rahon Road, Ludhiana Punjab.

Procurement of raw material: The sisal fibre used for the present study was procured from Girish Grah Udhyog Kotdwar, Uttrakhand and cotton fibre and yarns were procured from the Synthetic & Art Silk Mills' Research Association (SASMIRA) Mumbai. Blended yarn of sisal/cotton 35/65 ratio in 12s count was developed in ring spinning system at Central Institute for Research on Cotton Technology (CIRCOT), Mumbai.

Determination of fabric properties: Developed fabrics were studied for physical and mechanical properties to analyze their suitability for product development. The physical properties studied were fabric weight, fabric count and fabric thickness. The mechanical properties studied were tensile strength and bending length.

GSM (IS: 1964: 1970): The fabric's GSM is determined as the sample's weight in one g/m² of length. To maintain moisture equilibrium, the samples were conditioned at standard atmospheric conditions for 24 hours before being weighed. The specimen's GSM value was derived using the testing method IS: 1964:1970. The fabric sample were spread out on a flat surface, and using a template, square swatches of 25x25 cm were marked. Swatches were then cut and condition. As per Nadiger and Subramanium (2001), the reliability of the weight of the standard swatches was considered to be 0.5 g accurate. The GSM of the fabric was determined.

25cm × 25cm- "a' gram, GSM= 16 x "a' grams/meter square

Fabric Count (Numerical expression): When the fabric was completely free of wrinkles, the number of ends (warp) and picks (weft) per unit area were used to describe the fabric count in woven fabrics. The fabric sample's count was determined using a pick glass using BS Method 2862:1957. With the use of a magnifying counting device, the warp and weft yarn present in a square inch of fabric were counted randomly at various places along the length and width of the fabric sample. The average end and pick values per inch were then calculated.

Fabric thickness (IS: 7702-1985): The thickness of a textile fabric is determined by a precise measurement of the distance between two plane parallel plates when these are separated by the cloth, a known arbitrary pressure between the plates being applied and maintained. One of the plates was considered as pressure foot and the other as the anvil. The principle of the measurement of the fabric thickness is expressed in test method IS: 7702- 1985. The measurement was made at five different portions of the fabric. The thickness was calculated by taking the mean of the five measurements.

Tensile Strength (IS -1969 -1985): Tensile strength is the fabric's ability to withstand a force load which is usually represented in kilogram's or pounds. The tensile property of the fabric was recorded through computer using *Universal Strength Tester' according to IS test method 1969-1985. Samples of the size 325x60 mm were cut from warp and weft direction of the fabrics. The experiment was conducted under standard atmospheric conditions of 65±2 percent relative humidity (RH) and temperature of 27±2°C. The breaking strength of the test specimen was determined using the machine's indicators. The tensile property of all samples was thus determined in this manner.

Bending length (ASTM – D1388-64): The stiffness tester was used to determine the bending height of fabric. Cantilever test method was used in which the fabric specimen is allowed to bend under its own weight as the length, of the overhanging portion of the specimen, is gradually increased. The free length which bends under its own weight sufficiently to make its leading edge intersect a plane of 41.5degree inclination is taken as the measure of stiffness of the fabric. The fabric specimen was cut with the help of acrylic specimen preparing template (150 x 25 mm). Transferred the specimen on to the platform of the instrument. Note the reading on the scale confirming to the reference mark on the platform which was the bending length of the fabrics in cms. Similarly test at least five test specimens each for warp way and weft way.

Constructional details of developed handloom union fabrics: Three weaves were constructed in handloom i.e., HP= Handloom plain weave fabric, HB= Handloom Basket weave fabric and HT=Handloom Twill weave fabric using single ply sisal/cotton(35/65) blended yarn of 12s count in weft and cotton yarn of 2/20 count in warp direction. Thread counts in plain weave were 41 as ends per inch (EPI) and 34 as picks per inch(PPI), basket weave (EPI=63, PPI 43) and in twill weave, (EPI=42, PPI=36) in handloom prepared samples (Sample HP, HT and HT) are shown in Table 2.

RESULTS AND DISCUSSION

Analysis of physio-mechanical properties of handloom union fabrics: Physical properties of union fabrics encompass the features that provides basic texture, hand feel and dimension to the fabric. The physical and mechanical properties of developed handloom union fabrics in different weaves i.e., plain, basket and twill are shown in following figures.

Fabric weight: Figure 1 elucidates the average weight and weight/unit area of the different handloom weaves. HP (Plain weave) fabric has minimum weight of 172.7 g/m^2 and it was maximum for HB (handloom basket weave) i.e., 259g/m^2 respectively. The weight of basket weave fabric is more due to two or more warp and filling yarns that are used to create basket weave.

Elongation at break: The elongation at break is measurement which shows how much a material can be stretched as a percentage of its original dimension before its break. The plain weave had maximum elongation in warp direction i.e, 30.47 percent followed by basket (26.83 %) and twill weave (17.63%) respectively, whereas basket weave had highest elongation (31.43%) in weft direction followed by plain (16.47%) and twill weave (7.6%) (Fig. 2). Twill weave had minimum elongation both in warp and weft direction. This means a higher percentage of elongation indicates a better-quality material when combined with good tensile strength.

Breaking strength (N): Basket weave fabric had maximum breaking strength values 538than plain 441.33and twill weave 498in warp wise direction (Fig. 3). For weft direction, basket weave exhibited highest strength (340±4) as compared to plain and twill weave. So, it can be concluded that basket weave had more breaking strength than plain weave and twill weave.

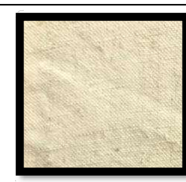
Fabric Stiffness (cm): Stiffness is an important characteristic of a fabric. Stiffness is measured by bending length of the fabric. Bending length is the length of fabric that will bend under its own weight to a definite extent. Bending length determines the draping quality of a fabric. Depicting bending length for the fabrics which evaluates the stiffness of the fabric as bending length is directly proportional to stiffness (Fig. 4). It was seen that HB had maximum stiffness 3.55in warp and 4.65in weft direction whereas it was minimum in warp direction for handloom plain weave fabric.

Fabric thickness (mm): It is the distance between the top and bottom surface of the fabric. Figure 5 reveal the thickness of the union fabrics, where basket weave union fabric had maximum thickness i.e., 1.216 followed by twill weave 0.81 and plain weave 0.80 respectively. Thickness is

Table 1. Sisal fibre used for the study

Sisal specie	Local name	Common name	Scientific name	Family
Agave americana	Pita fibre, Rambans	Century plant, Maguey or American aloe	*Agave americana*	Agavaceae

Table 2. Developed Sisal union fabrics (Cotton × Sisal / Cotton) in different weaves on handloom

Code assigned	Composition / EPI and PPI	Type of weave	Images
HP	Cotton (2/20) × Sisal/ Cotton (12s) EPI=41, PPI=34	Plain	
HB	Cotton (2/20) × Sisal/ Cotton (12s) EPI=63, PPI=43	Basket	
HT	Cotton (2/20) × Sisal/ Cotton (12s) EPI=42, PPI=36	Horizontal zig zag twill	

directly associated with the fabric comfort. The basket weave is thicker as compared to plain and will weave. Thus, it can be used to prepare any upholstery products where heavy fabric is required.

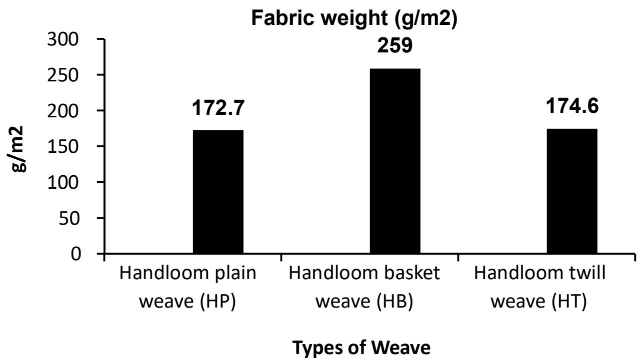


Fig. 1. Fabric weight of different weaves

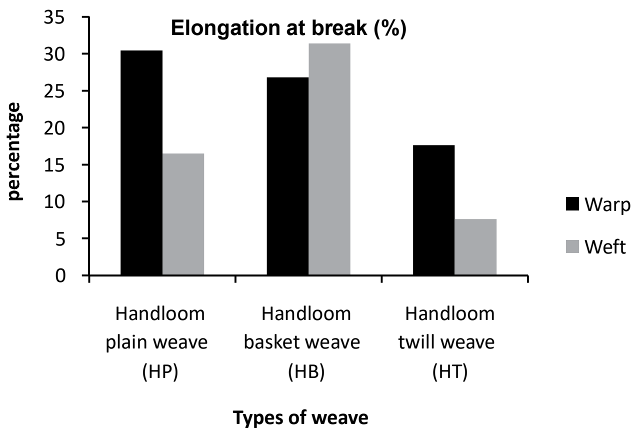


Fig. 2. Elongation at break of different weaves

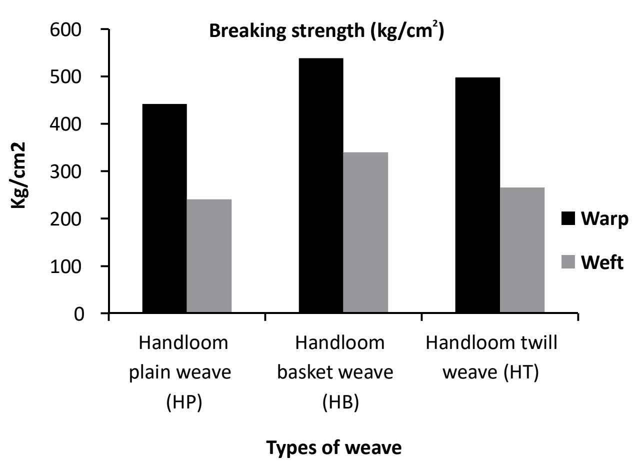
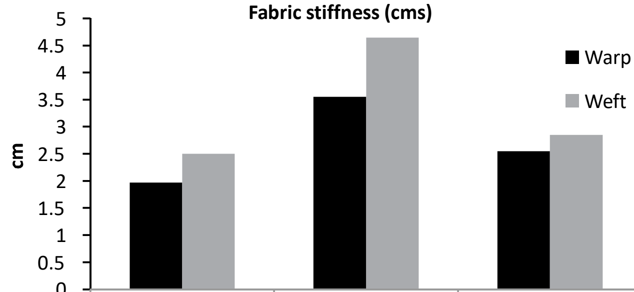


Fig. 3. Breaking strength of different weave



	Handloom plain weave (HP)	Handloom basket weave (HB)	Handloom twill weave (HT)
Warp	1.97	3.55	2.55
Weft	2.5	4.65	2.85

Fig. 4. Fabric stiffness of different weave

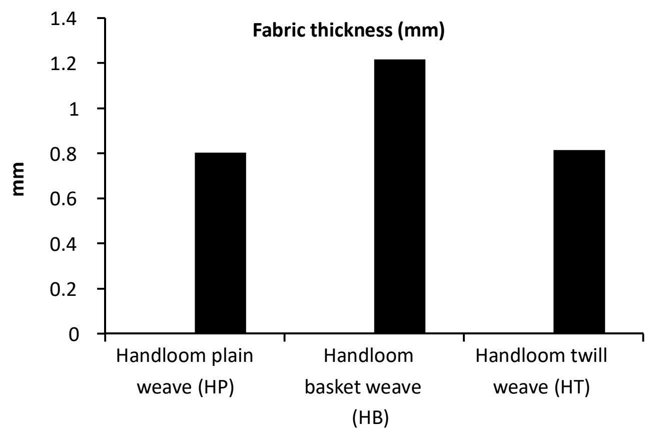


Fig. 5. Fabric thickness of different weaves

CONCLUSION

HP (handloom Plain weave) fabric has minimum weight of 172.7 g/m^2 whereas HB (handloom basket weave) has maximum weight i.e., 259g/m^2 respectively. Basket weave fabric had maximum breaking strength values 538N than plain 441N and twill weave 498N in warp wise direction. For weft direction, basket weave exhibited highest strength 340N as compared to plain and twill weave. So, it can be concluded that basket weave had more breaking strength than plain weave and twill weave both in warp and weft direction. Basket weave had maximum bending length 3.55cm in warp and 4.65cm in weft direction whereas it was minimum in warp direction for handloom plain weave fabric. It was inferred that basket weave was thicker as compared to plain and twill weave. Thus, it can be used to prepare any apparel/home textile products where heavy fabric is required. Twill fabric can be used for preparing waist coat, skirt, trousers, etc. The greater bending length along the weft direction of the fabric tells us that the fabric is stiffer in the weft direction than in the warp direction. This can be due to the result of fabric density which is more in the weft direction of the fabric than in the warp direction and the composition of weft yarn. Plain weave and twill weave had minimum bending length which means they are more comfortable than basket weave and can be used for apparel application.

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Utilization *of Heliconia rostrata* (Lobster claw) Fibre for Developing Eco-friendly Home Textile Products

O.R. Devi and H.K. Saini

Department of Apparel and Textile Science, Punjab Agricultural University, Ludhiana-141 004, India
E-mail: oinam.roselyn@gmail.com

***Abstract*:** '*Heliconia rostrata*', an ornamental plant was treated in aqueous medium for 45 days to extract the fibre. The functional groups of the fibre were studied using FTIR spectroscopy for better understanding of their behaviour. Various physical characterizations like fibre length, fibre diameter, bundle strength, moisture content and fibre yield percentage were performed on *H. rostrata* fibre. The TGA analysis proved that the *H. rostrata* fibre was thermally stable at a higher temperature range of 250-300°C. Due to the larger diameter and coarser nature of fibre, *H. rostrata* fibre could not use to develop yarns. Therefore, textile products were developed directly by inserting 4 to 5 fibres in weft direction and different count of cotton and jute yarns were used as warp yarns, thus eliminating yarn preparatory process. Total six eco-friendly home textile products were developed directly from fibres on handloom using plain weave and cost of each product was calculated. The results indicate that the price of developed eco-friendly home textiles was high as compared to products which are commercially available in the market. However, the costs can be brought down by scaling up the production. Therefore, developing eco-friendly home textile products directly from fibres will create a variety in fabric types and cater to the requirements of the present fashion world.

Keywords: Ornamental plant, *H. rostrata*, Retting, Properties, Eco-friendly

In the time of globalization and industrialization, the major concern is not only development but also environmental pollution which is also being taken into consideration. Nowadays, natural fibres have earned a great deal of attention because of use in industrial applications. Natural fibres have been used for the preparations of sacks, hessian bags etc. since very early times but synthetic fibres largely replace them starting in the middle of the 20th century as they have number of properties. These synthetic fibres provide a number of benefits, including better mechanical property. However, the concern of the society with a sustainable development promoted a comeback to lignocellulosic materials, and today natural fibres are replacing synthetic ones, such as glass fibres in the automotive industry (Naik et al 2022). Lignocellulosic fibres are highly appealing option from economic and ecological perspective because they are non- toxic, typically inexpensive, have a low density, and are less abrasive to molds and processing machinery equipment. Besides they consume less energy to be produced as they are sustainable materials which are available in nature and are biodegradable and eliminate carbon emissions together (Girijappa et al 2019). Many common lignocellulosic fibres such as jute (*Corchorus capsularis*), sisal (*Agave sisalana*), and flax (*Linumu sitatissimum*), used in several commercial products. Several other less common fibres also have a great potential to be used as reinforcement in polymer matrix composites. These less common fibres are not yet largely exploited due to being restricted to a certain ecosystem and/or region or simply because they are only obtained as a by-product of other harvests (Navarro et al 2013).

Heliconias (Heliconia spp.) belong to the family Heliconiaceae and are members of the diverse taxonomic group Zingiberales. Quinaya and Almeida (2019) observed fibres extracted from the stem of *H. rostrata* plant have microstructural characteristics and thermal properties similar to several other commonly used lignocellulosic fibres, such as coir (*Cocos nucifera*). From the thermo gravimetric analysis, the onset of the thermal degradation of *H. rostrata* fibre, is higher than 230°C. Therefore, this fibre could be used with several common thermoplastic matrices in the manufacturing of composites, because processing temperatures will be lower than the beginning of the thermal degradation of the fibres. Therefore, in this work the physio-chemical properties of fibre obtained from the stalk of *H. rostrata* ornamental plant was determined to explore the use of this fibre for developing eco-friendly home textile products.

MATERIAL AND METHODS

Fibres extraction and processing: The *H. rostrata* fibre used in the experiments was extracted in Thoubal district, Manipur, India (Fig.1). The fibre was extracted for different retting days, i.e., 14, 21, 30, 45 and 60 days. These time durations were used to optimize the days for water retting.

Visual and tactile observations were used to evaluate each retted fibre sample. Fibre strength was checked manually by hand. The fibre with better strength was selected and time taken for retting of selected fibre samples were considered as optimum time for water retting.

Optimization of bleaching concentration by whiteness index: The optimization of bleaching agent was done at different bleaching concentration 1.0, 2.0, 3.0, 4.0 and 5.0% for 45 min at 70°C, measured weight loss and whiteness index were calculated with the help of Colorflex. Fibre that showed minimum weight loss with good whiteness index was considered as optimum bleaching concentration.

Sample preparation & conditioning: The retted dried samples were oven dried for 24 hours at 105°C before milling and conditioned at 65 ± 2% relative humidity and 27 ± 2°C temperature for 24 hrs to ensure environmental equilibrium moisture content, prior to testing. (NIS: 43:1980) and (ASTM: D1776-79).

Fibre morphology: Longitudinal and cross-sectional surface of *H. rostrata* fibre images were recorded using Field Emission Scanning Electron Microscope (FESEM) Model number JSM 5400, JEOL ltd. Japan.

Fourier transform infrared spectroscopy (FTIR) analysis: The Fourier transform infrared (FTIR) spectra of fibre sample was recorded in the region of 400-4000 cm^{-1} at a resolution of 0.5 cm^{-1} on Perkin-Elmer Spectrum 2 equipped with an attenuated total reflection (ATR) attachment.

Thermo -gravimetric (TGA) analysis: TGA analysis was done with Perkin Elmer TGA 4000 and degradation of the sample with respect to temperature was recorded. The analysis was done by heating the fibre from 30 to 500°C with a rate of 20°C/ min and nitrogen as purge gas.

Fibre diameter: Diameter of 200 *H. rostrata* fibres were measured using projection microscope (magnification 10x) supplied by Leica Microsystems.

Bundle strength: For the determination of bundle strength, pressley strength test method (IS 3675:66) was used and small tuft of 80 to 120 fibres was taken for measurement. About 10 reading of sample were tested and analyzed.

Fibre moisture content: The moisture content of fibers was expressed in percentage on oven dry basis.

$$\text{Moisture content } \% = \frac{(\text{a - b})}{\text{a}} \times 100$$

Where, a = original mass in g of the test specimen, and b = oven dry mass in g of the test specimen

Fibre yield percentage: The yield percentage of the fibre was calculated.

$$\text{Yield } \% = \frac{\text{Weight of the fibres}}{\text{Weight of the plant material}} \times 100$$

Development of eco-friendly home textile products from *H. rostrata* fibre: To develop the fabric from *H. rostrata* fibres, 4 to 5 fibres were directly inserted as weft and different yarn counts (cotton and jute) were used as warp using plain weave on handloom.

Out of these developed fabrics, total six eco-friendly home textiles were prepared and costing of each product was calculated. The cost price of each textile product was calculated by considering the cost of weaving, designing, cost of raw material used and the labour involved in making it. The selling price of each product was calculated by adding 30 per cent profit to the cost price of each product as it considered as genuine profit margin and consumer acceptance.

RESULTS AND DISCUSSION

Optimization of days for water retting: The optimization of time duration for water retting was done to obtain the good quality fibre, for which the stems of *H.rostrata* were kept in water for 14, 21, 30, 45 and 60 days (Table 1). Fibres looked fresh and difficult to separate the fibre from the bark till 14, 21 and 30 days of retting Good quality shiny with good strength fibre was achieved during 45 days of retting. However, with increase in the retting time from 45 to 60 days, there was slightly loss in strength of the fibre and fibre was disintegrated on rubbing. Therefore, 45 days was considered as optimum time duration for extraction of *H. rostrata* fibre from stem.

Optimization of bleaching concentration by whiteness index (WI): The increase in bleaching concentration from 1.0 to 5.0 %, loss in fibre weight from 4.9 to 9.78% and whiteness index was increased from 48.23 to 63.84 (Table 2). Weight loss was due to removal of unwanted impurities from the

Table 1. Optimization of retting days of *H. rostrata* plant (Water retting)

Retting time (Days)	Observation recorded
14	Barks look fresh, difficult to separate the fibre
21	Barks are not fully retted
30	Fibres are not fully separated
45	Fully retted fibre with better strength
60	Fibres disintegrated on rubbing

Table 2. Optimization of bleaching concentration by whiteness index

Bleaching concentration (%) (Hydrogen hypochlorite)	Weight loss (%)	Whiteness index value (WI)
1.0	4.93	48.23
2.0	6.01	57.88
3.0	7.10	59.62
4.0	7.20	60.12
5.0	9.78	63.84

fibre. The fibre "*H. rostrata"* showed minimum weight loss in 1.0% bleaching concentration which is 4.93% but WI value was minimum (48.23). Thus, the 2.0% hydrogen hypochlorite bleach concentration is suitable for *H. rostrata* fibre, which also maintains its weight and improves whiteness as well.

Fibre morphology: The wavy strands on the surface are present throughout the length of the fibre and have rough surface in the longitudinal view (Fig. 2a). These also have large oval shaped pores in between them (Fig. 2b). In the cross sectional view, it looks like a greyish matter with the left border being whitish. There are also longitudinal strands in the background towards the left and the right corners (Fig. 2c). The shape of the cross-section of *H. rostrata* fibre is irregular and cannot be defined of having any particular shape.

Fourier transform infrared spectroscopy (FTIR) analysis: FTIR spectrum of the *H. rostrata* fibre showed well defined broad peaks at 3333, 1424, 1027 and 554 cm^{-1}. The broad peak observed corresponds to the stretching vibration mode of intra and intermolecular hydroxyl (-OH) bond of cellulose. The peak 1424 cm^{-1} proved the presence of alcohols, carboxylic acids, esters, ethers and aliphatic compounds. The strong and broad bond at 1027 cm^{-1} is for C-O of which indicates presence of cellulose in the fibre. The strong bond at 554 cm^{-1} was assigned to C-X of which confirms the presence of bromo-alkanes in the fibres.

Thermo -gravimetric (TGA) analysis: The fibre mass decreased slightly in low temperatures (25-150°C) due to the loss of moisture content (Fig. 4). The hemicellulose degradation process was between the temperature 200-300°C and the decomposition of cellulose and lignin was between the temperatures of 300-400°C. There was

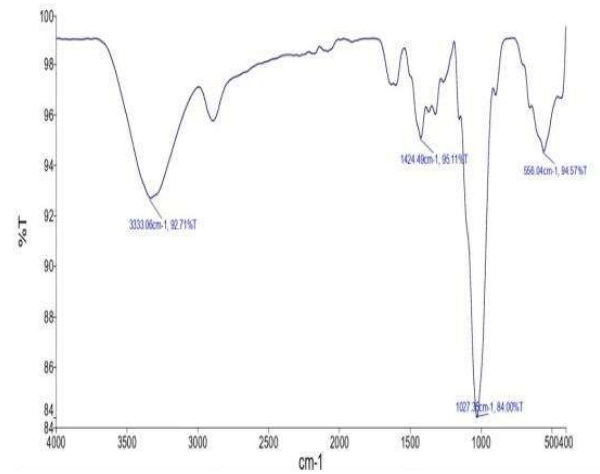


Fig. 3. FTIR analysis of *H. rostrata* fibre

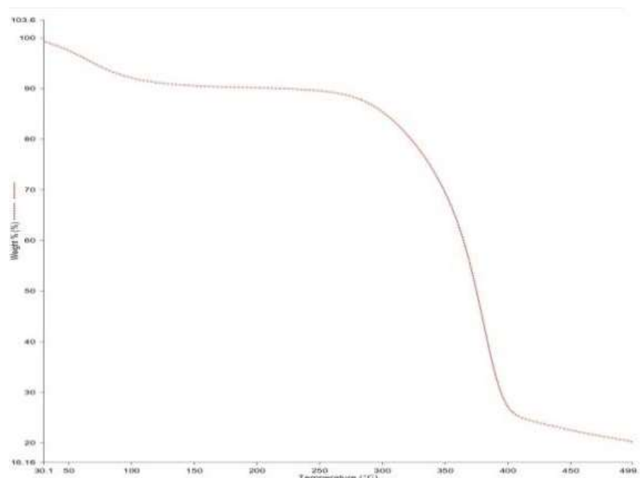


Fig. 4. Thermo-gravimetric analysis of *H. rostrata* fibre

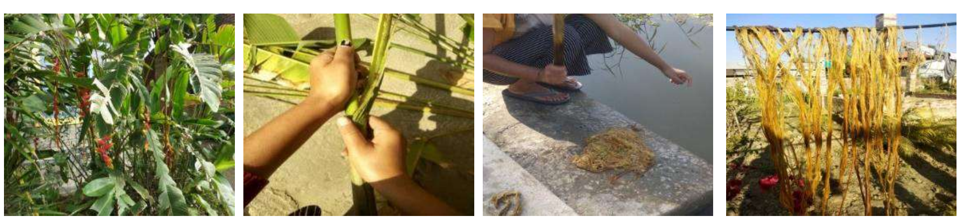
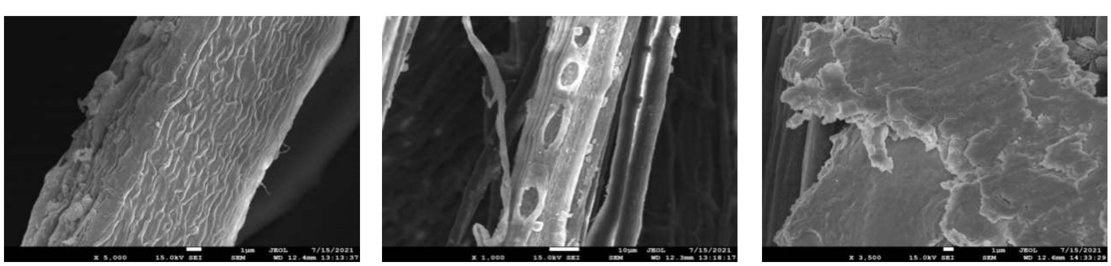


Fig. 1. Extraction of fibre from *H. rostrata* plant



(a) (b) (c)

Fig. 2. (a-b) Longitudinal and (c) cross sectional view of *H. rostrata* fibre

significant drop in weight (mass of the fibre) between 300 and 400°C, proving that the cellulose had completely broken down due to the breakdown of its molecular structures. Most of the cellulose's structure is crystalline, making it strong and resistant to hydrolysis. Strong intramolecular and intermolecular hydrogen bonds in this crystalline structure require higher energies to be broken down. It was observed that at high temperature 400-500°C, pectin and wax present in fiber were degraded. These indicate that thermal decomposition of the fibre is not a single step process but involves the initial degradation of primary products followed by the cellulose and secondary products. Thus, from the thermal analysis it can be summarized that *H. rostrata* fibre is thermally stable up to a range of 250-300°C.

Physical properties analysis of *H. rostrata* fibre: *H. rostrata* fibre showed larger diameter (35.71µm), exhibited very coarse nature (Table 3). This coarse nature of *H. rostrata* fibre opened a new arena of application. The coarser fibre had high importance in the end-use specific applications like floor covering, carpet backs, mattress and other non-apparel purposes. Fibres are typically used in groupings rather than separately, such as in yarns or fabrics. Thus, during the tensile break of yarns or fabrics, bundles or groups of fibres are involved. Additionally, there is a correlation between spinning efficiency and bundle strength that is at least as strong as the correlation between spinning efficiency and intrinsic strength as measured by testing individual fibres. Compared to testing individual fibre, testing bundles of fibres takes less time and exerts less force. These factors have made determining the breaking strength of fibre bundles more significant than determining the strength of a single fibre. The fibre bundle strength and length of *H. rostrata* fibre were 55.43 cm and 29.77(g/tex). The important consideration when using natural fibre as reinforcing materials is moisture content, which affects natural fibre's dimensional stability, electrical resistivity, tensile strength, porosity, and swelling behaviour in composite materials (Razali et al 2015). The moisture content of *H. rostrata* fibre possessed lower moisture content (8.35 %). Usually, between 4.5 and 7.5 per cent of the weight of the plant is composed of fibre. The soil's fertility, the variety sowed, the plants' fibre content, crop management, harvesting stages, season, and the prevalence of disease and pests all have impact on yield of fibres (Fatma and Jahan 2017). Therefore, *H. rostrata* fibre have fibre yield percentage of 5.25%.

Table 3. Physical properties analysis of *H. rostrata* fibre

Physical parameters	*H. rostrata* fibre
Fiber diameter (µm)	35.71
Fibre length (cm)	55.43
Bundle strength (g/tex)	29.77
Moisture content (%)	8.35
Fibre yield (%)	5.25

Development of fabrics and eco-friendly home textile products from *H. rostrata* fibre: Different home textile products are prepared from the *H. rostrata* fibre. Firstly, the fabric was developed on handloom in plain weave by directly inserting the fibre as weft yarn and one or two ply cotton and jute yarns as warp yarns. After weaving, the fabrics were cut, stitched, decorated and finished to develop total six eco-friendly textile products.

Details of developed eco-friendly home textile products from *H. rostrata* fibre: A thin table runner (Fig. 5a) of size 36" x 14" was prepared by inserting *H. rostrata* fibres as weft with one ply cotton yarns of 20Ne yarn count as warp. Raw edges of runner were finished by knotting. Another table runner of size 36"x 14" was prepared by inserting the fibre in weft direction and 1.5Ne yarn count cotton in warp direction (Fig. 5b). Net embroidered lace was used for decoration and two laces have been used in the centre of the table runner facing each other. A table mat (Fig 5c) of size 19"x 14" was prepared by using 2 ply cotton of 1.5Ne yarns as warp. Net embroidered lace was used for decoration. One pair of round placemats of diameter of 12" were developed by inserting the fibres of 1.5 yarn count cotton as warp. For finishing, the raw edges were

Table 4. Calculation of cost for prepared eco-friendly home textiles

Product name	Raw material cost (in ₹)					Selling price (₹)
	Weaving cost (a)	Finishing (Lining, zipper and embellishments) (b)	Labour cost (c)	Cost (a-c)	Profit margin (30%)	
Thin table runner	1800	-	200	2180	660	2840
Thick table runner	1800	100	250	2050	620	2650
Table mat	900	60	200	1160	350	1500
Handbag	900	240	300	1440	430	1900
Round placemat	900	60	200	1160	350	1500
File cover	500	90	250	840	252	1100

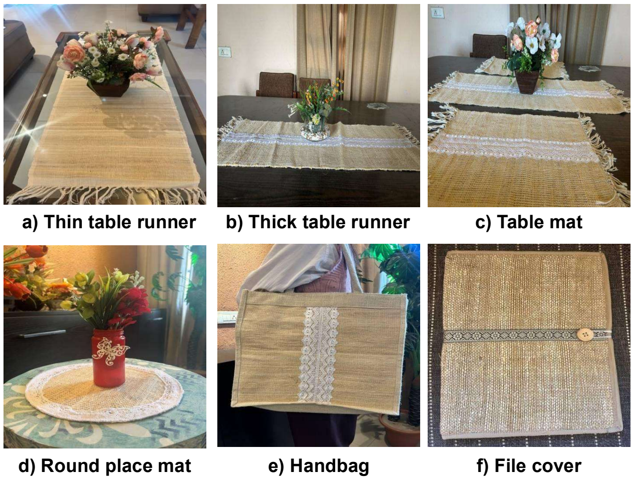


Fig. 5. Eco-friendly home textile products developed from *H. rostrata* fibre

stitched with pico machine and lace has been attached in the sides of the placemats (Fig. 5d). Hand bag was prepared by inserting the fibre on the one ply cotton of 20 Ne yarn count as warp yarns. White colour jute fabric was used on the side, up and bottom of the bag. Cotton rope was used as handle. And for decoration, embroidered net lace was placed in the centre in both sides of the bag (Fig. 5e). Lastly, file cover of size 9.5”x 12” was prepared on 2 ply jute yarn of diameter 2 mm as warp yarn. For decoration, border laces of black colour were paste in the centre of the file cover. Non -woven fabrics were used as lining material for file cover. Wooden button and elastic strings were used to fold file cover (Fig. 5f).

Economics of friendly home textile products: The total cost of thin table runner was ₹ 2180.00 and a profit margin of 30 per cent has been added to the cost price to calculate quoted price. Thus, the selling price was ₹ 2840.00. For thick table runner, the total cost was ₹ 2050.00 and a profit charges was ₹620.00, thus selling price of product was ₹2650.00. In case of Table mat, the total cost was ₹1160.00 and the selling price was ₹ 1500. Data in Table 4 also indicated that for a handbag, the total selling cost of handbag was ₹1900.00 which includes total cost of ₹1440.00 and profit charges of ₹430.00. The selling price of round placemats was ₹1500.00 in pairs which comprises of total cost of ₹1160.00 and profit charges of ₹350.00. Lastly, the selling price of file cover was ₹1100.00 which covers total cost of ₹ 840.00 and profit charges of ₹ 250.00.

Therefore, the price of the developed eco-friendly home textile products was quiet high as compared to products which are commercially available in the market. The reason is due to the higher fibre extraction cost. The weaving cost was also high because the fabric was developed directly from the fibres and weavers inserted 4 to 5 fibres at one time as weft. Among the developed textile products, the cost of thick table runner was high as compared to other products. However, the costs can be brought down by scaling up the production.

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CONCLUSION

The present study includes extraction of fibre from *Heliconia rostrata* stem, analysis of fibre properties and development of textile products. The suitable time of retting for *H. rostrata* fibres was 45 days. Fibres have rough surfaces and have oval shaped pores in longitudinal surfaces which highlight possibility useful in acoustic and insulation application. The fibre bundle strength, fibre length, fibre yield percentage and fibre diameter of *H. rostrata* fibre were 55.43 cm, 29.77 (g/tex), 5.25% and 35.71 µm, respectively. Due to the larger diameter and coarser nature of fibre, *H. rostrata* fibre could not be possible to develop yarns. Therefore, textile products were developed directly by inserting the 4 to 5 fibres in weft direction and different count of cotton and jute yarns were used as warp yarns, thus eliminating yarn preparatory process.

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Effect of Pre-treatment on Aggregation, Biochemical Quality and Membrane Clarification of Pineapple Juice

Samreen, Ch V.V. Satyanarayana[1], L. Edukondalu[2], Vimala Beera[3] and V Srinivasa Rao[4]

Department of Food Process Engineering, College of Food Science & Technology
PJTS Agricultural University, Rudrur-503 188, India
[1]Department of Food Process Engineering, Dr. N.T.R College of Food Science & Technology, ANGR Agricultural University, Bapatla-522 101, India
[2]Department of Food Process Engineering, College of Food Science & Technology, ANGR Agricultural University, Pulivendula-516 390, India
[3]Department of Food Safety and Quality Assurance, Dr. N.T.R College of Food Science & Technology, ANGR Agricultural University, Bapatla-522 101, India
[4]Department of Statistics and Mathematics, Agricultural College, ANGR Agricultural University, Bapatla-522 101, India
E-mail: samreen.sam26@gmail.com

Abstract: Pineapple (*Ananas comosus* L., Merril) is the most popular tropical non-citrus fruits, mainly because of their attractive aroma, refreshing flavour and Brix/acid ratio. The research was carried out on the physicochemical analysis and membrane clarification of pineapple juice after pretreatment. Pretreatment of pineapple juice was performed using egg albumin with different concentrations and observed that 2 g/L concentration gave effective removal of colloidal substances of pineapple juices. The biochemical analysis of pineapple juice after pretreatment revealed TSS, colour intensity, browning index, turbidity, titrable acidity, pH, viscosity, total antioxidant activity, total phenolic content, total anthocyanin content, reducing sugars, non-reducing sugars, total sugars and colour were 13.255%, 8.254, 6.024, 0.782, 3.357%, 4.813, 2.33cP, 20.202 mg/g, 60.231 (mg of GAE/g of dry material), 0.7744mg/100mL, 7.605%, 1.951%, 9.556% and 8.984, respectively. There were significant differences among all the treatments of pineapple juices. There is a reduction in most of the biochemical constituents due to pretreatment. It is due to aggregation of these components and retention on the membrane. Permeate flux generally declined with time for both MF and UF. However, increase in permeate flux was achieved with increase in TMPs and feed flow rates. The permeate flux was high during MF of pineapple juice than UF. The decrease in pore size and MWCO also decreased the permeate flux. In MF and UF of pineapple juice, the initial fluxes were high but gradually decreased.

Keywords: Membrane clarification, Microfiltration, Ultrafiltration, Egg albumin, Permeate flux, MWCO, Pore size

Pineapple (*Ananas comosus* L., Merril) is the tropical non-citrus fruit, mainly because of its attractive aroma, refreshing flavour and Brix/acid ratio. This juice have been used in fruit based beverages individually, in the form of mixture or combined with other fruit juices. As an ingredient, the concentrated juice from pineapple blends well with other aromas of fruits resulting in a pleasant product with a competitive market price. Pineapple juice is a popular product because of nutritional compounds for human health identified as phytochemicals, such as vitamin C, carotenoid, flavanoid and phenolic compounds (Laorko et al 2010). Due to these characteristics and increasing public awareness about nutritional food, the demand for the pineapple fruit has significantly increased in the last years. Consequently, many industries producing pineapple fruit juice as well as pharmaceutical companies extracting health beneficial compounds from the fruits have been developed. There is a worldwide increasing tendency for the consumption of tropical fruits, juices and fruit drinks due to the interest in ready to consume healthy products. Fruit juices are liquid foods that provide vitamins, sugars, mineral compounds and water. Consumers have individual preferences for specific appearance, consistency and flavor characteristics. Traditional methods of processing fruits limit the possibility to retain freshness as much as possible and its health-beneficial compounds. Similarly, the concentration of fruit juices by thermal evaporation results in color degradation and reduction of most thermally sensitive compounds. Phytochemicals in pineapple juice are reduced during a conventional heating and often leads to detrimental change in the sensorial and nutritional quality. Membrane technology is an alternative to produce a juice with good nutritional characteristics as it does not destroy the vitamins and other nutrients. It is also an alternative because of its operational advantages such as mild temperature, ease of scale-up and simplicity.

Introduction of membrane processing enables production of additive-free juices with high quality and natural fresh like taste. Juice clarification, stabilization, depectinization and concentration are typical steps in which membrane processes such as microfiltration (MF), ultrafiltration (UF), nanofiltration (NF) and reverse osmosis (RO) can be potentially utilized. Clarification based on membrane processes, particularly UF and MF, have replaced conventional clarification, resulting in elimination of chemical clarifying agents and simplified process for continuous production. Purpose of the membrane processing is to remove suspended solids as well as haze-inducing and turbidity causing substances to obtain a clear juice after storage. Pineapple juice in its original state have a turbid appearance that makes it hard to preserve during the storage. Since the main problem with juices is stability, there is a need for research to solve this problem besides preservation of color. According to the literature, the present methods used for polyphenol elimination to produce stable juice involve liquid extraction with organic solvents. However, these methods require high temperature to increase the extraction rate and yield, but, may denature the polyphenols leading to undesirable byproducts. Therefore, MF and UF as modern methods are used to reduce juice turbidity. MF and UF are non- thermal and low cost separation technologies for juices emerging in recent years. UF and MF have been applied in vegetal juices, pulps and wine industries, reducing many steps of the conventional clarification. Also, pectinolytic enzymes can be reduced and sometimes can even be eliminated. Keeping in view of the above points, a study was undertaken on membrane processing of pineapple juice after pretreatment with egg albumin. The study also constituted the analysis of physicochemical characteristics of juice and establishing operational parameters to achieve high permeate flux.

MATERIAL AND METHODS

Pineapple (cv. *Simhachalam)* variety was obtained from local market, Bapatla, Guntur dist. Andhra Pradesh and properly sorted to discard fruits of mechanical damage while transportation. Pineapple fruits were properly peeled, cut into slices and used for extraction of juice.

Pre-treatment on aggregation and clarification of pineapple juice: The pretreatment was performed using a fining agent called egg albumin. The juice was subjected to four concentration levels *i.e.,* 0.25, 0.5, 1 and 2 g/L and effect of pretreatment was analysed (Table 1). After the collection of juice, the egg albumin powder was added and mixed thoroughly. The juice samples were muslin cloth filtered and centrifuged at 4000 rpm (2147 g) for 5 min (Domingues et al 2011). The supernatant was used for biochemical quality analysis to determine the effect of pretreatment. The concentration of egg albumin which resulted in better clarification was determined by biochemical quality analysis. This concentration was subsequently used for pretreatment of pineapple juice in all the experiments. The pretreatment was performed to remove the colloidal substances present in the juices. Colloids can decrease the permeate flux during filtration of the juice due to presence of pectinases, cellulase, hemicellulase, xylanase, carbohydrase, glucanase or arabinose. Removal of aggregates of these species via pretreatment may increase the permeate flux due to the reduction in the size of the particles and the subsequent decrease in viscosity (Valero et al 2014). The results of the biochemical analysis were expressed statistically with SPSS software.

The pretreated pineapple juice with egg albumin was subjected to physicochemical analysis. Total soluble solids (TSS) of juice were measured by Refractometer (ATAGO make, range 58-90%) and expressed in terms of % Brix. The pH measurement was performed using a digital pH meter (Systronics digital pH meter 355). The colour intensity was measured using a Systronics PC based Double Beam Spectrophotometer at absorbance of 510 nm. Similarly, Browning index was expressed as the ratio of 420 nm to 520 nm using Systronics PC based Double Beam Spectrophotometer (Valero et al 2014). The turbidity and color was also measured using Systronics PC based Double Beam Spectrophotometer at absorbance of 700 nm and 420 nm respectively. The turbidity values of both juices were measured according to the procedure given by Valero et al (2014). Titratable acidity of both juices are determined by the procedure of AOAC (2005). Titratable acidity is expressed as the amount of free acid mainly as anhydrous citric acid present in fruit, conveniently in g acid per 100 g or 100 ml.

$$\%\ \text{acidity} = \frac{a \times b \times c \times d \times 100}{e \times w \times 1000} \qquad (1)$$

where, a = titre value (volume of 0.1N NaOH)

b = Normality of the alkali (0.1N), c = volume made up, d = equivalent weight of the acid, w = weight or volume of sample taken (g or ml) , e = aliquot

Viscosity of the fruit juice was determined by using Digital Viscometer (Brookfield, Model: DV1MLV). Lane and Eynon method (Ranganna, 1986) was used for determination of total, reducing and non-reducing sugars.

$$\text{Reducing sugars}\% = \frac{(\text{Factor}\ (0.052) \times \text{dilution} \times 100)}{(\text{titre} \times \text{wt. of sample})} \qquad (2)$$

$$\text{Total sugars}\% = \frac{(\text{Factor}\ (0.052) \times \text{dilution} \times 100)}{(\text{titre} \times \text{wt. of sample})} \qquad (3)$$

Non-reducing sugars % = Total sugars-reducing sugars (Saeed and Iftikhar et al 2002, Ahmmed et al 2015)

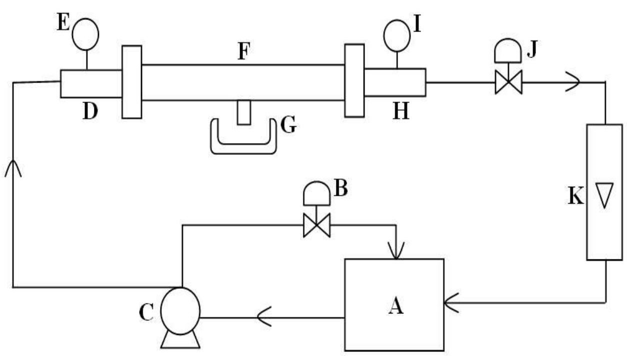
The antioxidant assay was estimated by ferric reducing antioxidant power method using ascorbic acid as standard and total Phenolic content by Folin Ciocalteu's method using gallic acid as standard (Kametkar et al 2014). The total anthocyanin content was determined by the procedure given by Raj et al (2011). Anthocyanins are water soluble phenolic glycosides belonged to flavonoid pigments having C_{15} skeleton of flavones as basic structural unit.

$$\text{Total O.D./100ml} = \frac{\text{O.D.} \times \text{Volume made up} \times 100}{\text{ml of juice taken}} \quad (4)$$

$$\text{Total anthocyanin (mg/100 ml)} = \frac{\text{Total O.D./100 ml}}{87.3} \quad (5)$$

Membrane clarification of pineapple juice: Membrane clarification (MF and UF) of pineapple juice after pretreatment was carried out at Dr. N.T.R. College of Agricultural Engineering, Bapatla in hollow fibre membrane module setup (Model: HFM – 01). The term membrane processing in this thesis is essentially clarification of juices using membranes.

Hollow fibre membrane module setup: The schematic of hollow fiber membrane set up is shown in Figure 1 and Plate 1. The heart of the set up is the hollow fiber module (F). The feed is drawn by the booster pump (C) and fed to the module by 6 mm polyurethane tube via a Perspex flange. Two pressure gauges in the range of 0 to 60 psi (4.1364 bar) are attached to the upstream and downstream of the module. A3/4 inch needle valve (J) of stainless steel has been fitted in the retentate line after the module. This valve is used for fine tuning of pressure and flow rate through the module. A rotameter (K) of range 0 to 50 L/h is attached to the retentate line and the retentate stream is recycled back to the feed tank (A). A bypass line is connected from the pump to the feed tank and a1/2inch stainless steel needle valve (B) is attached to the bypass line. The permeate flows through a 5 mm polyurethane pipe into permeate collector (G). By controlling the bypass valve (B) and retentate valve (J), one can control the flow rate and the transmembrane pressure drop across the module, independently. The transmembrane pressure



A : Feed tank, B : Bypass valve, C : Booster pump, D : Short piece, E : Upstream pressure gauge (0 – 4.21 kg/cm^2 (60 psi)), F : Hollow fibre module, G : Permeate collector, H : Short piece, I : Downstream pressure gauge (0 – 4.21 kg/cm^2(60 psi)), J : Pressure valve (Needle type), K : Rotameter (0 – 50 Lph)

Fig. 1. Schematic diagram of the hollow fibre membrane module setup

Plate 1. Hollow fibre membrane setup

Table 1. Independent and dependent variables for pretreatment on aggregation and clarification of pineapple juice

Independent variables	Dependent variables
Concentrations of egg albumin: 0.25, 0.5, 1 and 2 g/L	Total soluble solids (TSS), pH, Turbidity, Viscosity, Titratable acidity, Colour, Colour intensity, Browning index, Total antioxidant activity (TAA), Total phenolic content (TPC), Total anthocyanin content (TAC), Reducing and non-reducing sugars, Total sugars

drop is the arithmetic average of the readings in the pressure gauges E and I. The physical dimension of the setup is 70 mm in length, 48 mm in width and 65 mm in height. The weight of the setup is approximately 10 kg. One power point of domestic line 220 V is required to run the pump.

Membrane processing of pineapple juice was carried out in the membrane module setup with different hollow fibre cartridges. The container was filled with 250 mL of juice. The operation was done in total recycle mode. The suction, retentate, by-pass lines were kept in feed solution and continuous operation was carried out. The permeate was collected at permeate line separately. The technical specifications of MF and UF membrane were given in Table 2 and 3, respectively. Further, pure water flux data was collected both for MF and UF membranes using distilled water. After each run, the set up was flushed with distilled water and then cleaned with 0.1 N hydrochloric acid (HCl) for 30 mins in total recycle mode according to the washing protocol given by the manufacturer. After thorough washing, the permeability of the cartridges was analysed to measure the change in permeability of the hollow fibres. All the experiments were conducted in triplicate at room temperatures (30±2°C). After every experiment, the membranes were cleaned properly and stored in the 1% formalin solution for future use.

The permeate flux was calculated as

$$J^* = \left(\frac{1}{A}\right) \times \left(\frac{dv}{dt}\right) \qquad (6)$$

Where, J*= Permeate flux (L/h m^2), A= Area of the membrane (m^2), dv= Volume of flow rate (L), dt= Time of flow rate (h)

The permeate collected was stored in glass bottles. The experiments were performed according to the different conditions laid down in the Table 3 and analyzed to obtain high permeate flux.

RESULTS AND DISCUSSIONS

In pineapple juice after the pre-treatment, the 2 g/L concentration level recorded highest removal of colloidal substances. There was a decrease in all the constituents of the juice but not high to lose the essential components of juice. The decrease in complex colloidal components might increase in permeate flux of the juice decreasing the fouling. There was decrease in TSS of pineapple juice. The 2 g/L concentration recorded TSS of 13.255 % for pineapple juice. Similar results were recorded by Valero et al (2014). The decrease of colour intensity was observed for pineapple juice. The colour intensity for pineapple juice was 8.254 at 2 g/L concentration. As the concentration level of egg albumin

Table 2. Technical specifications of MF and UF membranes

Membrane material	Pore size (µm)	Water permeability as claimed by the manufacturer (m / Pa.s)
MF membranes		
Poly acrylonitrile (PAN) (Make : M/s Technoquips seperations equipments Pvt. Ltd., Kharagpur)	0.2	53.4×10^{-11}
Poly acrylonitrile (PAN) (Make : M/s Technoquips seperations equipments Pvt. Ltd., Kharagpur)	0.1	44.5×10^{-11}
UF membranes		
Poly Sulphone (PS) (Make : M/s. Technoquips Seperations Equipments Pvt Ltd., Kharagpur)	120	29.7×10^{-11}
Poly Sulphone (PS) (Make : M/s. Technoquips Seperations Equipments Pvt Ltd., Kharagpur)	70	24.3×10^{-11}
Poly Sulphone (PS) (Make : M/s. Technoquips Seperations Equipments Pvt Ltd., Kharagpur)	44	20.5×10^{-11}
Poly Sulphone (PS) (Make : M/s. Technoquips Seperations Equipments Pvt Ltd., Kharagpur)	10	13.4×10^{-11}

Table 3. Operating variables for microfiltration and ultrafiltration of pineapple juice

Operating variables	
Membrane poresizes	MF - 0.1 and 0.2 µm UF – 120, 70, 44 and 10 kDa
Transmembrane pressures (TMP):	0.3447 bar (5 psi), 0.6894 bar (10 psi), 1.0342 bar (15 psi) and 1.3789 bar (20 psi)
Crossflow velocities/ Feed flow rates	0.024 m/s (20 Lph), 0.037 m/s (30 Lph) and 0.049 m/s (40 Lph)

increased, the clarity in the juice increased, but there was significant decrease in colour intensity of pineapple juice. This was in accordance with Mirsaeedghazi et al (2010). There was significant decrease in all the biochemical attributes as the concentration level of egg albumin increased which enhanced the filtration process while membrane processing by high permeate flux. Valero et al (2014) recorded decrease in pineapple juice upon using egg albumin as pre-treatment. BI values were high prior to the pre-treatment and in non- clarified juice. Similarly, Alper et al (2005) also observed decrease in browning degree for non-clarified juice. The decrease of 49.1% was obtained with gelatine and bentonite combined conventional fining. Browning index being 6.024 for pineapple juice in 2 g/L concentration level of egg albumin. The turbidity values decreased as the concentration level increased. The highest clarity was obtained with 2 g/L egg albumin. The pineapple juice recorded high mean values of turbidity because of fibrous and pectinaeceous present in fruit pulp. Similarly turbidity was also decreased with addition of PVPP and bentonite and increased the clarity of the juices (Vardin and Fenercioglu 2003, Valero et al 2014). The data pertaining to pineapple juice of all the biochemical attributes were also tabulated (Table 4).

Titrable acidity (TA) values decreased as the concentration of egg albumin increased. The pH values increased for pineapple juice. The mean values for TA of pineapple juice was 3.357 expressed as citric acid. Similarly, the mean pH value for pineapple juice was 4.813. The best results were for 2 g/L concentration level of egg albumin. However, there was significant difference among all the concentration levels of egg albumin. Similar, trend was observed by Alighourchi et al (2009). Initially the concentration level 0.25 and 0.5 g/L did not show any significant difference in the values (Molina et al 2009). Viscosity of the pineapple juice decreased with increase of concentration level of egg albumin. The mean value of viscosity of pineapple juice at 2 g/L concentration level was 2.33 cP. There was a significant decrease in viscosity in both the juices at different concentration levels of egg albumin. The antioxidant activity decreased as the concentration level of egg albumin increased. The highest decrease was observed in 2 g/L egg albumin for pineapple juice as 20.202mg/g. The centrifugation and clarification process might aid to the decrease in the antioxidant activity of pineapple juice. Similar results were recorded by Vegara et al (2013) in terms of pomegranate juice. The results pertaining to antioxidant activity obtained in this study were not in accordance to Valero et al (2014) where the antioxidant activity increased upon increase of concentration level of egg albumin. Total phenolic content of pineapple juice also significantly decreased with increase of concentration level of egg albumin. Valero et al (2014) and Pinelo et al (2010) also observed decrease of total phenolic content with 2 g/L egg albumin. The decrease of phenolic content might be due to the changes in the different clarification techniques with egg albumin.

Total anthocyanin content decreased as the

Table 4. Biochemical characteristics of pineapple juice after pretreatment with different concentrations of egg albumin (Mean± SD)

Property	Concentration of egg albumin (g/L)			
	0.25	0.5	1	2
Total soluble solids (%)	15.224±0.010	14.926±0.010	14.325±0.050	13.255±0.050
Colour intensity	8.850±0.050	8.644±0.010	8.454±0.010	8.254±0.010
Browning index	6.433±0.010	6.321±0.050	6.284±0.050	6.024±0.050
Turbidity	0.824±0.010	0.801±0.010	0.791±0.010	0.782±0.050
Titratable acidity (%)	4.204±0.010	3.957±0.010	3.723±0.010	3.357±0.050
pH	3.86±0.010	4.136±0.015	4.57±0.010	4.813±0.050
Viscosity (cP)	5.213±0.057	4.77±0.017	3.55±0.010	2.33±0.01
Total antioxidant activity (mg/g)	22.553±0.040	21.852±0.010	21.198±0.057	20.202±0.058
Total phenolic content (mg of GAE/g of dry material)	81.256±0.010	73.422±0.020	67.584±0.020	60.231±0.050
Total anthocyanin content (mg/100 mL)	0.812±0.050	0.8103±0.010	0.7924±0.010	0.7744±0.030
Reducing sugars (%)	9.325±0.020	9.273±0.050	8.873±0.010	7.605±0.010
Non - reducing sugars (%)	1.82±0.02	1.685±0.010	1.352±0.010	1.951±0.010
Total sugars (%)	11.145±0.050	10.958±0.050	10.225±0.010	9.556±0.010
Colour	9.114±0.02	9.105±0.05	8.996±0.01	8.984±0.02

concentration of egg albumin in clarification increased. There was no significant difference for pineapple juice at 0.25 and 0.5 g/L of egg albumin. The anthocyanin significantly decreased for 1 and 2 g/L egg albumin. Anthocyanin compounds are labile and undergo degradative reactions. There are many variations in the stability of the structure of anthocyanins (Wrolstad 2000, Delgado and Paredes 2002). Anthocyanins remain stable in dried form than in state of high water activity (Wrolstad et al 2005). Reducing sugars significantly decreased for pineapple juice. The highest decrease was at 2 g/L concentration level egg albumin. The mean value of decrease obtained for pineapple juice was 7.605%. Similarly, the decrease was obtained for total sugars and non-reducing sugars. Total sugars significantly decreased for pineapple juice as the concentration level increased. The decrease of total sugars was achieved due to conversion into simpler sugars while hydrolysis in juice.

The colour values significantly decreased for pineapple juice. Anthocyanins are phenolic compounds responsible for colour in juice. As, the phenolic content of the juice was in decreasing trend, the anthocyanins also decreased in juices. This might be the reason for the decrease of colour in juice as anthocyanins decreased. The loss of anthocyanin pigments occurred because of addition of egg albumin as it is sequestering agent it removes the colloidal substances in flocs which might have reduced the pigments in turn aided to decrease in colour in pineapple juice. Vardin and Fenercioglu (2003) recorded the same decrease with the use of gelatine addition and natural sedimentation. The 2 g/L egg albumin was the best concentration level as flocculating agent to remove the colloidal substances and was utilized as pre-treatment prior to membrane processing of pineapple juice to increase permeate flux and reduce fouling.

Membrane processing of pineapple juice and establishing operational parameters to achieve high permeate flux: In membrane processing, both Microfiltration (MF) and Ultrafiltration (UF) hollow fibre cartridges were used for clarification of pineapple juice. The different combinations of membrane pore sizes, transmembrane pressures and flow rates were given in Table 3.

I.MF of pineapple juice: Pineapple juice after pre-treatment with egg albumin was subjected to membrane processing using microfiltration with membrane pore sizes of 0.2 and 0.1 μm. The permeate flux declined with time on both the membranes (Table 5 and 6). The permeate flux of pineapple juice was low because the juice was thick with colloidal pectinaceous substances. The pineapple juice highest flux was 121.191 L/m^2 h for 0.2 μm pore size membrane because of its larger pore size. As pineapple juice is a colloidal solution of highly complex sugars, filtration of juice would have formed a secondary layer of colloids on the membrane surface due to concentration polarization (Blatt et al 1970). The decline in permeate flux was perhaps due to the formation of secondary layer on the membrane surface. The decline in permeate flux during MF was also recorded earlier (Chilukuri et al 2001, Bottino et al 2002, Cassano et al 2003, Onsekizoglu et al 2010). The permeate flux increased as the transmembrane pressures and flow rates increased. The permeate flux was observed to decrease gradually and reach a steady state flux for both the membrane pore sizes. The flux decreases sharply initially due to membrane fouling and gradually thereafter and finally attains a steady state value. Similar results were obtained by Rai et al (2006) for mosambi juice and by Karmakar et al (2017) for coconut water. The difference in steady state values for both the pore sizes was marginal. Similar results were observed during MF of Tomato juice (Bottino et al 2002). It was also observed that as the processing time increased there was a decline in permeate flux probably because of deposition of colloidal substances while clarification on membrane surface. These colloidal substances resist flow of permeate which leads to fouling. Fouling may have occurred due to pore narrowing by smaller particles that may have accumulated on the pore walls (Chilukuri et al 2001) or by pore plugging. High amount of permeate flux was obtained for 0.2 μm pore size probably because of its larger pore size. High fluxes were recorded due to pretreatment with egg albumin at 2 g/L concentration as large flocs of colloidal substances were removed.

UF of pineapple juice: Ultrafiltration (UF) of pineapple juice was carried out with four different hollow fibre membranes with different molecular weight cut off (MWCO) *i.e.,* 120, 70, 44 and 10 kDa, transmembrane pressures 0.3447 bar (5 psi), 0.6894 bar (10 psi), 1.0342 bar (15 psi) and 1.3789 bar (20 psi) and three flow rates 20, 30, 40 Lph. After pre-treatment with egg albumin at 2 g/L concentration, pineapple juice was subjected to membrane processing. It was observed through the biochemical quality analysis that large colloidal substances present in the juice was removed in the form large flocs as egg albumin is a good flocculating agent. The permeability of UF membranes was determined using distilled water at different pressures prior to the ultrafiltration of pineapple juice.

120 kDa MWCO membrane: Membrane processing of pineapple juice was performed with 120 kDa MWCO membrane. The pineapple juice pre-treated with egg albumin was fed to the equipment for membrane processing. The permeate flux was observed to decrease as it was observed for filtration for all other membrane pore sizes (Table 7). The increase of flux with increase in flow rate was observed in the filtration process of pineapple juice. There was decrease in

Table 5. Variation of permeate flux at different transmembrane pressures and feed flow rates in MF of pineapple juice using 0.2 μm pore size membrane (Mean± SD)

Time (min)	Permeate flux (L/m² h)											
	TMP 1.37890 bar (20 psi)			TMP 1.0342 bar (15 psi)			TMP 0.6894 bar (10 psi)			TMP 0.3447 bar (5 psi)		
	20 Lph, 20 psi	30 Lph, 20 psi	40 Lph, 20 psi	20 Lph, 15 psi	30 Lph, 15 psi	40 Lph, 15 psi	20 Lph, 10 psi	30 Lph, 10 psi	40 Lph, 10 psi	20 Lph, 5 psi	30 Lph, 5 psi	40 Lph, 5 psi
0	0	0	0	0	0	0	0	0	0	0	0	0
10	98.227±1.87	105.535±1.65	121.191±9.86	87.326±5.22	98.218±2.54	115.758±8.57	85.452±4.16	98.458±7.80	108.041±9.59	84.261±5.64	91.606±2.00	96.458±5.80
20	93.864±1.21	104.009±1.79	112.25±2.37	89.921±1.27	93.686±1.85	103.675±2.37	80.489±1.33	82.505±1.91	91.755±2.37	73.525±1.39	80.123±1.96	82.238±2.37
30	84.709±1.33	96.653±1.91	108.325±2.48	81.518±1.33	84.376±1.91	96.32±2.48	78.517±1.39	73.419±1.96	86.724±2.42	69.489±1.33	73.452±1.91	77.917±2.42
40	79.982±1.04	86.201±1.44	97.573±2.54	78.688±1.44	79.415±2.02	85.701±2.31	71.942±1.56	67.323±2.14	76.089±2.54	65.193±1.44	67.523±1.79	72.625±2.31
50	62.352±0.87	81.779±1.45	87.25±2.60	64.582±1.44	73.006±2.02	81.119±2.60	52.595±1.44	56.291±2.02	70.661±2.71	52.528±1.56	56.391±1.85	69.343±2.42
60	61.185±1.15	77.889±1.73	82.272±2.31	47.701±1.15	66.741±1.73	77.555±2.31	44.561±1.15	49.224±1.73	64.2152.31	44.561±1.15	49.224±1.73	59.548±2.31
70	41.635±1.21	44.095±1.79	66.375±2.37	37.129±1.27	41.148±1.85	61.23±2.37	30.96±1.33	44.775±1.91	51.089±2.37	27.653±1.39	28.879±1.96	44.509±2.37
80	33.689±1.27	37.581±1.85	56.480±2.42	33.488±1.27	36.795±1.85	52.407±2.42	27.720±1.27	37.949±1.85	39.588±2.42	26.091±1.27	14.385±1.85	37.616±2.42
90	30.817±1.33	38.344±1.91	52.374±2.48	33.454±1.33	38.344±1.91	44.189±2.48	26.057±1.33	33.199±1.91	34.881±2.48	13.905±1.33	15.811±1.91	30.294±2.48
100	22.658±1.39	35.138±1.96	35.834±2.54	24.945±1.39	31.022±1.96	34.805±2.54	20.637±1.39	25.455±1.96	25.973±2.54	10.658±1.39	11.359±1.96	23.400±2.54
110	20.152±1.21	31.432±1.79	31.975±2.54	21.445±1.21	23.079±1.79	28.217±2.37	19.184±1.21	23.834±1.79	25.928±2.37	9.523±1.21	10.629±1.79	16.984±2.37
120	19.846±1.27	29.358±1.85	30.327±2.42	19.119±1.27	21.398±1.85	24.284±2.42	16.922±1.27	23.038±1.85	20.722±2.42	8.825±1.27	10.161±1.85	14.035±2.42

Table 6. Variation of permeate flux at different transmembrane pressures and feed flow rates in MF of pineapple juice using 0.1 μm pore size (Mean± SD)

Time (min)	Permeate flux (L/m² h)											
	TMP 1.37890 bar (20 psi)			TMP 1.0342 bar (15 psi)			TMP 0.6894 bar (10 psi)			TMP 0.3447 bar (5 psi)		
	20 Lph, 20 psi	30 Lph, 20 psi	40 Lph, 20 psi	20 Lph, 15 psi	30 Lph, 15 psi	40 Lph, 15 psi	20 Lph, 10 psi	30 Lph, 10 psi	40 Lph, 10 psi	20 Lph, 5 psi	30 Lph, 5 psi	40 Lph, 5 psi
0	0	0	0	0	0	0	0	0	0	0	0	0
10	89.979±1.21	97.419±1.79	110.106±2.37	84.743±1.27	89.612±1.85	98.316±2.37	73.785±1.33	87.024±1.91	89.312±2.37	73.752±1.39	77.322±1.96	80.094±2.37
20	83.234±1.39	91.388±1.85	107.434±2.54	79.749±1.44	82.867±2.02	93.252±2.60	67.689±1.50	76.356±2.08	82.500±2.66	67.656±1.56	74.189±2.14	76.893±2.71
30	78.122±1.33	86.125±1.91	104.037±2.48	69.976±1.33	77.789±1.91	89.889±2.48	56.658±1.39	71.095±1.96	77.489±2.42	56.691±1.33	62.114±1.91	69.343±2.42
40	71.529±0.87	75.615±1.45	93.252±2.60	62.621±1.44	70.862±2.02	77.389±2.60	49.391±1.44	64.381±2.02	70.462±2.71	49.324±1.56	57.566±1.85	62.054±2.42
50	48.920±1.04	71.291±1.44	81.709±2.54	45.900±1.44	68.718±2.02	73.697±2.31	33.399±1.56	51.223±2.14	68.418±2.54	47.690±1.44	54.395±1.79	58.991±2.31
60	44.428±1.21	57.529±1.79	77.951±2.37	41.482±1.27	50.382±1.85	59.515±2.37	31.689±1.33	39.631±1.91	50.082±2.37	44.824±1.39	41.080±1.96	39.364±2.37
70	37.981±1.15	47.156±1.73	69.709±2.31	37.195±1.15	44.622±1.73	48.186±2.31	25.058±1.15	36.301±1.73	45.400±2.31	36.634±1.15	38.218±1.73	38.911±2.31
80	36.748±1.33	37.024±1.91	61.163±2.48	31.388±1.33	36.415±1.91	39.726±2.48	21.658±1.33	33.414±1.91	36.082±2.48	24.721±1.33	30.535±1.91	33.080±2.48
90	31.732±1.27	34.648±1.85	43.918±2.42	23.379±1.27	30.875±1.85	34.314±2.42	21.389±1.27	28.259±1.85	30.541±2.42	19.303±1.27	23.795±1.85	27.926±2.42
100	23.545±1.21	26.191±1.79	38.278±2.54	21.425±1.21	24.651±1.79	29.717±2.37	19.422±1.21	24.379±1.79	26.716±2.37	11.792±1.21	19.379±1.79	24.701±2.37
110	19.345±1.39	24.531±1.96	30.990±2.54	17.621±1.39	22.012±1.96	25.201±2.54	14.424±1.39	21.282±1.96	20.528±2.54	9.756±1.39	14.622±1.96	18.899±2.54
120	19.379±1.27	21.807±1.85	28.865±2.42	17.133±1.27	19.068±1.85	22.352±2.42	13.488±1.27	15.394±1.85	18.314±2.42	7.722±1.27	11.720±1.85	14.081±2.42

permeate flux with increase in time (Bottino et al 2002). Though the constant TMPs were tend to be maintained there was decrease in permeate flux with time, this might be because of fouling as the pineapple juice is higher in its viscosity. Similar results were expressed by Bottino et al (2002) that poor pulp fluidity and high viscosity, the membrane pores clog which in turn increased the pressures and further channel clogging caused the decline in permeate flux.

70 kDa MWCO membrane: The permeate flux declined gradually for all the experiments at different TMPs and flow rates (Table 8). The initial increase in permeate flux was increased with increase in flow rates and TMPs, and then declined. The highest permeate flux obtained at 40 Lph flow rate, 20 psi (1.3789 bar) TMP was 97.244 L/m^2 h and then the lowest flux obtained was 5.512 L/m^2 h at 20 Lph flow rate, 5 psi (0.3447 bar) TMP.

The decline in permeate flux might have occurred because of concentration polarization on the membrane surface. The concentration polarization might have occurred because of some colloids which could not be removed in the pretreatment process of egg albumin. These colloids might adhere to the membrane surface causing plugging of pores. This in turn could have reduced permeate flux of the juice. The fouling might be predominant because of the tighter membrane pore size where easy clogging of membrane would have taken place. Initially high amount of flux rates were achieved because of the high TMPs and flow rates but gradually the permeate flux decreased. The increased flux rates initially might also because clean membrane in which the pores were unclogged.

44 kDa MWCO membrane: The pineapple juice clarification was also performed with 44 kDa MWCO membrane at different trans membrane pressures and flow rates. The membrane permeability of the 44 kDa membrane was verified with different TMPs. The permeate flux declined gradually for all the experiments at different TMPs and flow rates (Table 9). The initial increase in permeate flux was increased with increase in flow rates and TMPs and then declined. The highest permeate flux for 44 kDa MWCO was obtained at 40 Lph flow rate, 20 psi TMP was 90.284 L/m^2 h and then the lowest flux was 4.488 m^2 h at 20 Lph flow rate, 5 psi TMP. Similarly, the highest permeate flux for 10 kDa MWCO membrane was 86.921 L/m^{2}h and the lowest was 3.392 m^{2}h. The decrease in permeate flux was probably due to concentration polarization by sediments of pineapple which could not be removed while aggregation process with egg albumin.

10 kDa MWCO membrane: The membrane permeability of the 10 kDa MWCO membrane was determined as the

Table 7. Variation of permeate flux at different transmembrane pressures and feed flow rates in UF of pineapple juice using 120 kDa MWCO membrane (Mean± SD)

Time (min)	Permeate flux (L/m^2 h)											
	TMP 1.37890 bar (20 psi)			TMP 1.0342 bar (15 psi)			TMP 0.6894 bar (10 psi)			TMP 0.3447 bar (5 psi)		
	20 Lph, 20 psi	30 Lph, 20 psi	40 Lph, 20 psi	20 Lph, 15 psi	30 Lph, 15 psi	40 Lph, 15 psi	20 Lph, 10 psi	30 Lph, 10 psi	40 Lph, 10 psi	20 Lph, 5 psi	30 Lph, 5 psi	40 Lph, 5 psi
0	0	0	0	0	0	0	0	0	0	0	0	0
10	81.519±1.39	91.388±1.85	108.720±2.54	73.768±1.44	81.152±2.02	93.038±2.60	56.591±1.50	71.028±2.08	80.785±2.66	56.558±1.56	61.981±2.14	73.035±2.71
20	73.901±1.21	86.192±1.79	104.533±2.37	71.510±1.27	73.535±1.85	88.241±2.37	49.457±1.33	64.448±1.91	73.235±2.37	49.424±1.39	57.500±1.96	70.877±2.37
30	70.190±1.33	75.355±1.91	99.750±2.48	63.974±1.33	69.857±1.91	83.672±2.48	47.852±1.39	60.477±1.96	69.557±2.42	51.447±1.33	54.328±1.91	63.512±2.42
40	50.206±1.04	71.291±1.44	93.286±2.54	41.382±1.44	64.431±2.02	76.012±2.31	31.556±1.56	53.946±2.14	64.131±2.54	41.380±1.44	47.490±1.79	60.277±2.31
50	44.628±0.87	57.722±1.45	87.679±2.60	37.029±1.44	44.455±2.02	67.956±2.60	24.891±1.44	51.289±2.02	63.174±2.71	38.318±1.56	44.557±1.85	53.779±2.42
60	37.981±1.15	47.156±1.73	67.480±2.31	31.488±1.15	44.875±1.73	49.473±2.31	21.758±1.15	38.016±1.73	44.542±2.31	27.090±1.15	30.635±1.73	37.682±2.31
70	41.960±1.21	37.091±1.79	62.087±2.37	23.379±1.27	41.593±1.85	43.223±2.37	21.355±1.33	29.984±1.91	41.293±2.37	24.062±1.39	24.355±1.96	29.717±2.37
80	31.732±1.27	33.447±1.85	50.907±2.42	21.392±1.27	24.617±1.85	37.273±2.42	19.389±1.27	24.345±1.85	33.114±2.42	11.759±1.27	19.345±1.85	22.181±2.42
90	23.749±1.33	27.468±1.91	43.585±2.48	17.654±1.33	22.045±1.91	33.166±2.48	14.457±1.33	21.315±1.91	25.320±2.48	9.789±1.33	14.655±1.91	20.004±2.48
100	21.425±1.27	26.348±1.85	34.614±2.42	17.133±1.27	19.068±1.85	25.611±2.42	13.488±1.27	17.174±1.85	23.038±2.42	7.722±1.27	14.081±1.85	15.321±2.42
110	17.515±1.21	19.121±1.79	29.660±2.54	14.625±1.21	17.332±1.79	21.785±2.37	13.156±1.21	15.530±1.79	17.498±2.37	6.752±1.21	13.529±1.79	12.353±2.37
120	17.314±1.39	19.035±1.96	21.187±2.54	14.054±1.39	16.755±1.96	17.592±2.54	12.055±1.39	13.539±1.96	14.352±2.54	6.312±1.39	7.323±1.96	13.012±2.54

Table 8. Variation of permeate flux at different transmembrane pressures and feed flow rates in UF of pineapple juice using 70 kDa MWCO membrane (Mean± SD)

Time (min)	Permeate flux (L/m² h)											
	TMP 1.37890 bar (20 psi)			TMP 1.0342 bar (15 psi)			TMP 0.6894 bar (10 psi)			TMP 0.3447 bar (5 psi)		
	20 Lph, 20 psi	30 Lph, 20 psi	40 Lph, 20 psi	20 Lph, 15 psi	30 Lph, 15 psi	40 Lph, 15 psi	20 Lph, 10 psi	30 Lph, 10 psi	40 Lph, 10 psi	20 Lph, 5 psi	30 Lph, 5 psi	40 Lph, 5 psi
0	0	0	0	0	0	0	0	0	0	0	0	0
10	77.331±1.21	75.422±1.79	97.244±2.37	69.413±1.27	72.677±1.85	86.097±2.37	47.885±1.33	51.356±1.91	76.665±2.37	47.723±1.39	54.295±1.96	71.520±2.37
20	65.012±1.39	71.057±1.85	92.857±2.54	61.819±1.44	41.048±2.02	80.176±2.60	31.589±1.50	67.570±2.08	67.923±2.66	41.313±1.56	44.391±2.14	67.204±2.71
30	44.361±1.33	57.462±1.91	82.172±2.48	37.881±1.33	32.052±1.91	72.310±2.48	26.652±1.39	64.036±1.96	65.913±2.42	38.451±1.33	39.125±1.91	63.769±2.42
40	38.148±0.87	56.612±1.45	76.531±2.60	36.291±1.44	30.988±2.02	64.312±2.60	21.592±1.44	59.286±2.02	61.459±2.71	24.588±1.56	30.568±1.85	59.053±2.42
50	37.524±1.04	47.323±1.44	70.991±2.54	32.632±1.44	22.945±2.02	60.277±2.31	21.222±1.56	51.073±2.14	55.985±2.54	24.029±1.44	29.041±1.79	50.973±2.31
60	31.765±1.21	40.555±1.79	57.800±2.37	22.783±1.27	21.058±1.85	46.438±2.37	19.355±1.33	24.312±1.91	40.221±2.37	14.955±1.39	19.279±1.96	35.720±2.37
70	23.849±1.15	27.568±1.73	50.545±2.31	20.889±1.15	17.421±1.73	35.753±2.31	14.557±1.15	21.415±1.73	41.541±2.31	11.825±1.15	9.556±1.73	29.107±2.31
80	21.391±1.33	26.315±1.91	42.084±2.48	16.088±1.33	16.766±1.91	27.936±2.48	13.454±1.33	17.141±1.91	29.436±2.48	7.688±1.33	14.048±1.91	24.506±2.48
90	17.482±1.27	19.087±1.85	34.872±2.42	15.821±1.27	14.258±1.85	30.456±2.42	13.123±1.27	15.496±1.85	27.326±2.42	6.719±1.27	13.495±1.85	21.066±2.42
100	17.381±1.27	19.101±1.85	32.042±2.42	14.493±1.27	13.787±1.85	22.181±2.42	12.122±1.27	14.418±1.85	20.809±2.42	6.379±1.27	13.078±1.85	13.863±2.42
110	15.415±1.39	16.430±1.96	26.402±2.54	12.424±1.39	13.761±1.96	17.827±2.54	9.658±1.39	11.723±1.96	16.541±2.54	5.524±1.39	10.083±1.96	11.396±2.54
120	14.925±1.21	16.089±1.79	19.327±2.54	12.414±1.21	14.523±1.79	16.574±2.37	9.412±1.21	10.425±1.79	15.353±2.37	5.512±1.21	6.684±1.79	9.738±2.37

Table 9. Variation of permeate flux at different transmembrane pressures and feed flow rates in UF of pineapple juice using 44 kDa MWCO membrane (Mean± SD)

Time (min)	Permeate flux (L/m² h)											
	TMP 1.37890 bar (20 psi)			TMP 1.0342 bar (15 psi)			TMP 0.6894 bar (10 psi)			TMP 0.3447 bar (5 psi)		
	20 Lph, 20 psi	30 Lph, 20 psi	40 Lph, 20 psi	20 Lph, 15 psi	30 Lph, 15 psi	40 Lph, 15 psi	20 Lph, 10 psi	30 Lph, 10 psi	40 Lph, 10 psi	20 Lph, 5 psi	30 Lph, 5 psi	40 Lph, 5 psi
0	0	0	0	0	0	0	0	0	0	0	0	0
10	67.424±1.39	74.385±1.85	90.284±2.54	59.391±1.44	67.291±2.02	83.391±2.60	55.367±1.50	63.025±2.08	73.497±2.66	49.212±1.56	59.351±2.14	69.176±2.71
20	37.424±1.21	64.136±1.79	86.097±2.37	32.732±1.27	23.045±1.85	73.663±2.37	21.658±1.33	66.641±1.91	68.947±2.37	24.062±1.39	28.709±1.96	66.375±2.37
30	31.698±1.33	47.056±1.91	84.744±2.48	22.749±1.33	21.025±1.91	69.309±2.48	21.322±1.39	59.748±1.96	63.769±2.42	14.988±1.33	19.312±1.91	59.481±2.42
40	23.916±1.04	27.735±1.44	71.634±2.54	20.723±1.44	17.254±2.02	64.264±2.31	19.222±1.56	41.331±2.14	61.987±2.54	9.723±1.44	11.459±1.79	57.833±2.31
50	21.658±0.87	26.575±1.45	68.385±2.60	16.022±1.44	16.699±2.02	61.740±2.60	14.391±1.44	21.248±2.02	55.885±2.71	7.555±1.56	14.081±1.85	38.902±2.42
60	17.548±1.15	19.154±1.73	60.406±2.31	15.887±1.15	14.325±1.73	54.489±2.31	13.554±1.15	17.241±1.73	44.542±2.31	6.785±1.15	13.562±1.73	32.537±2.31
70	17.414±1.21	19.135±1.79	56.728±2.37	14.493±1.27	13.787±1.85	50.297±2.37	13.089±1.33	15.463±1.91	34.862±2.37	6.312±1.39	13.012±1.96	27.788±2.367
80	15.482±1.27	16.496±1.85	37.830±2.42	12.491±1.27	13.827±1.85	31.184±2.42	12.122±1.27	14.418±1.85	29.255±2.42	5.591±1.27	15.226±1.85	22.181±2.42
90	14.858±1.33	16.022±1.91	35.138±2.48	12.347±1.33	13.456±1.91	23.648±2.48	9.691±1.33	11.756±1.91	20.218±2.48	5.445±1.33	10.116±1.91	19.123±2.48
100	12.054±1.39	14.155±1.96	30.903±2.54	11.522±1.39	12.992±1.96	17.827±2.54	9.312±1.39	11.325±1.96	16.326±2.54	5.094±1.39	9.193±1.96	10.538±2.54
110	11.856±1.21	12.821±1.79	24.429±2.54	10.864±1.21	12.822±1.79	16.512±2.37	7.954±1.21	9.521±1.79	13.811±2.37	5.052±1.21	7.182±1.79	8.748±2.367
120	11.491±1.27	12.159±1.85	15.964±2.42	9.521±1.27	12.047±1.85	14.320±2.42	7.521±1.27	8.366±1.85	11.055±2.42	4.488±1.27	4.936±1.85	7.904±2.42

Table 10. Variation of permeate flux at different transmembrane pressures and feed flow rates in UF of pineapple juice using 10 kDa MWCO membrane (Mean± SD)

Time (min)	Permeate flux (L/m^2 h)											
	TMP 1.37890 bar (20 psi)			TMP 1.0342 bar (15 psi)			TMP 0.6894 bar (10 psi)			TMP 0.3447 bar (5 psi)		
	20 Lph, 20 psi	30 Lph, 20 psi	40 Lph, 20 psi	20 Lph, 15 psi	30 Lph, 15 psi	40 Lph, 15 psi	20 Lph, 10 psi	30 Lph, 10 psi	40 Lph, 10 psi	20 Lph, 5 psi	30 Lph, 5 psi	40 Lph, 5 psi
0	0	0	0	0	0	0	0	0	0	0	0	0
10	55.501±1.27	64.288±1.79	86.921±2.42	49.885±1.33	59.135±1.91	77.060±2.42	47.424±1.39	57.521±1.96	71.915±2.42	44.295±1.44	54.057±2.02	67.166±2.48
20	23.716±1.39	27.501±1.85	77.851±2.54	20.723±1.44	17.254±2.02	73.744±2.60	19.255±1.50	41.364±2.08	66.637±2.66	9.656±1.56	21.331±2.14	61.887±2.71
30	21.391±1.33	26.315±1.91	71.453±2.48	16.088±1.33	16.766±1.91	65.879±2.48	14.424±1.39	21.282±1.96	62.483±2.42	7.688±1.33	14.048±1.91	55.408±2.42
40	17.715±0.87	19.314±1.45	64.955±2.60	15.721±1.44	14.158±2.02	59.724±2.60	13.388±1.44	17.074±2.02	53.527±2.71	6.552±1.56	13.495±1.85	47.734±2.42
50	17.514±1.04	19.335±1.44	61.559±2.54	14.393±1.44	13.687±2.02	50.545±2.31	12.956±1.56	15.330±2.14	45.695±2.54	6.279±1.44	13.112±1.79	42.398±2.31
60	15.515±1.21	16.530±1.79	48.796±2.37	12.491±1.27	13.827±1.85	41.936±2.37	12.088±1.33	14.385±1.91	36.363±2.37	5.524±1.39	12.419±1.96	33.147±2.37
70	14.958±1.15	16.122±1.73	40.683±2.31	12.447±1.15	13.556±1.73	35.967±2.31	9.791±1.15	11.856±1.73	32.966±2.31	5.545±1.15	10.216±1.73	28.036±2.31
80	12.087±1.33	14.188±1.91	33.938±2.48	11.555±1.33	13.025±1.91	31.151±2.48	9.345±1.33	11.358±1.91	28.579±2.48	5.127±1.33	9.226±1.91	22.362±2.48
90	11.823±1.27	12.787±1.85	29.469±2.42	10.831±1.27	12.788±1.85	28.183±2.42	7.921±1.27	9.487±1.85	24.325±2.42	5.019±1.27	7.148±1.85	16.864±2.42
100	11.524±1.21	12.081±1.79	24.901±2.54	9.554±1.21	12.193±1.79	24.358±2.37	7.554±1.21	9.090±1.79	20.928±2.37	4.521±1.21	6.515±1.79	15.867±2.37
110	10.435±1.39	11.425±1.96	20.828±2.54	8.756±1.39	9.435±1.96	16.669±2.54	7.056±1.39	7.492±1.96	14.826±2.54	3.424±1.39	5.608±1.96	9.681±2.54
120	10.382±1.27	11.347±1.85	15.964±2.42	8.415±1.27	9.148±1.85	12.320±2.42	6.532±1.27	7.058±1.85	9.319±2.42	3.392±1.27	5.058±1.85	6.317±2.42

membrane was prior used for the membrane filtration of other juice which might clog the pores of membrane. The permeate flux declined gradually for all the experiments at different TMPs and flow rates (Table 10). The initial increase in permeate flux was increased with increase in flow rates and TMPs and then declined. The highest permeate flux recorded for 10 kDa MWCO membrane was obtained at 40 Lph flow rate, 20 psi (1.3789 bar) TMP as 86.921 L/m^2h and the lowest recorded was at 20 Lph flow rate, 5 psi (0.3447 bar) TMP as 3.392 L/m^2h.

In all the experiments with 70, 44 and 10 kDa MWCO membrane the permeate flux decline was observed. The decline in permeate flux was observed with decrease in membrane pore size. This was observed because of tighter membrane whose pore size was less which could prevent high amounts of permeate to flow through them and moreover the pore narrowing was predominant factor caused by fouling. Mohammadi et al (2002) defined fouling as existence and growth of microorganisms and irreversible collection of materials on the membrane surface which results in a flux decline. The viscosity of pineapple juice can also be considered as one of the constraints for the declining permeate flux. The permeate flux observed initially high because of the removal of pectinolytic substances due to aggregation with egg albumin which was achieved at 2 g/L concentration. Tapre and Jain (2014) observed that pectin enzymes were used in apple juice preparation to facilitate pressing or juice extraction and to aid in the separation of a flocculant precipitate by sedimentation, filtration or centrifugation. The permeate flux gradually decreased though constant TMP and flow rate were maintained because of formation of gel layer on the membrane surface. The gel layer could not be flushed by frequent cleaning in between intervals because the pineapple juice was thicker and high in viscosity.

CONCLUSIONS

Pineapple fruits of high grade variety were selected, cleaned, and juice was extracted. Pretreatment of pineapple juice was performed using egg albumin at different concentrations and observed that 2 g/L concentration gave good results in removal of colloidal substances of both juices. The optimum concentration of flocculant *i.e.,* 2 g/L of egg albumin effectively remove the colloids. Biochemical analysis of pineapple juice after pretreatment with optimum egg albumin concentration revealed the mean values of TSS, colour intensity, browning index, turbidity, titrable acidity, pH, viscosity, total antioxidant activity, total phenolic content, total anthocyanin content, reducing sugars, non-reducing sugars, total sugars and colour. The characteristics were analyzed in triplicate and found that they were in permissible limits. Permeate flux declined with time in both MF and UF. However, increase in permeate flux was achieved with increase in TMPs and feed flow rates. The permeate flux was high for MF than UF. The decrease in pore size also decreased the steady state permeate flux.

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Comparison of Bioactive Compounds and Antioxidant Potential of Different Cereal Grasses for Potential Therapeutic Use

D Bernice Ekhe, Renuka Aggarwal, Kiran Bains and Amandeep Singh Sidhu

Department of Food and Nutrition, Organic Farming
Punjab Agricultural University, Ludhiana-141 004, India
E-mail: bernicedecember@gmail.com

Abstract: Wheat grass is presumed to contain bioactive compounds and antioxidant activity and has gained popularity as a health drink. The present study was undertaken to explore the possibility of using other cereal grasses in place of wheat as affected by variety and growth length. The bioactive compounds total phenol content (TPC) and total flavonoid content (TFC) increased significantly with the increase in length of the grass in wheat varieties but decreased in rice and barley grasses. Maximum TPC and TFC was observed in the barley grass and rice grass at 6- and 4-inches length, respectively. Wheat grass showed maximum content at 15 and 20 cm inches. Wheatgrass of variety PBW 550 at length of 20 cm showed the highest antioxidant power as evidenced by FRAP (ferric reducing antioxidant power) and ABTS (2,2-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid) assay. Rice variety PR 126 at 10 cm and barley variety PL 807 at 15 cm exhibited maximum scavenging activity comparable to wheat grass. Wheat grass is the best cereal grass in terms of bioactive and antioxidant potential. There is a significant variation in these components of cereal grasses that change with increase in length of the grass. These should be harvested at the growth length when the bioactive compounds and antioxidant activities are optimum. Rice and barley grass also exhibit notable antioxidant potentials, which could be considered as a substitute for wheatgrass. The different varieties of these cereal grasses contain non-significantly different bioactive compounds. Hence, any variety which is available at a particular location can be cultivated.

Keywords: Wheat grass, Rice grass, Barley grass, Bioactive compounds, Free radical scavenging activity

The cereal grasses have gained wide attention as a novel and convincing source of natural antioxidants. The young leaves obtained from cereal grasses of the *Poaceae* or *Gramineae* family and are presumed to contain maximum bioactive and freely available compounds such as amino acids, vitamins, and trace elements (Pająk et al 2014). Major cereal grass that has gained popularity today is wheatgrass. Germination and sprouting increase antioxidants, nutrients and phenolic compounds contents of cereal grasses (Fortună et al 2018). The wheat grains sprouted for 6-10 days are considered as 'wheatgrass' (Akbas et al 2017, Benincasa et al 2019). Wheatgrass has been considered as storehouse of nutrients, containing considerable amounts of vitamins, minerals, amino acids, active enzymes antioxidants and chlorophylls (Ahmed et al 2021, Kaur et al 2021). The wheatgrass juice have antioxidant, antimutagenic, antimicrobial, antiallergenic, nephron-protective, anti-inflammatory and diuretic properties (Özköse et al 2016, Hebbani et al 2020, Thakur et al 2020, Choi et al 2021). The evidence also suggests the protective effect of wheatgrass juice against chemotherapy-induced damage (Avisar et al 2020a, 2020b). Its gluten-free properties has been found to be beneficial for those with gluten allergy (Qamar et al 2019). The young grass of the rice plant (*Oryza sativa)* has been introduced in recent times to substitute the use of wheatgrass, mainly in high rice production areas especially Asian countries where rice acts as the major food source of energy (Chomchan et al 2016). Khanthapok et al (2015) reported that rice grass juice possesses DNA protective and antioxidant properties including vitamins, minerals and phytonutrients. Wangchareon and Phimphilai (2016) observed that 6-20 days old rice grass has a high level of antioxidant compounds and contain higher levels of polyphenols while containing comparable antioxidant activities to wheatgrass juice. The use of barley (*Hordeum vulgare* L.) as a cereal grain date back to ancient times. Barley grass is the young leaves of the barley plant. Barley grass juice contain vitamins C and B like wheatgrass juice in addition to minerals like calcium, copper and manganese (Niroula et al 2019). The use of natural substances from plants as a functional component in beverages is on the rise resulting in functional beverages taking the leading spot among functional foods (Wangchareon and Phimphilai 2016). Wheatgrass has been blended with other sources like wheatgrass juice blended with pomegranate (Kashudhan et al 2016) and with kombucha (Sun et al 2015) which showed higher and more stable antioxidant activity suggesting that these could be used as novel beverages with effective, healthy and functional therapeutic properties (Sun et al 2015, Kashudhan et al 2016).

With the increase in the possibilities of wheatgrass to be used in the food industry, exploration to include other cereal grasses, importance to determine the appropriate variety and harvesting stage is needed. This study aimed to investigate the effect of different cereal grasses harvested at varied lengths on the bioactive compounds and antioxidant potential to determine the optimum harvesting stage for their potential therapeutic use.

MATERIAL AND METHODS

Plant materials, cultivation and harvesting: Three varieties each of wheat (Unnat PBW 550, PBW 343, PBW 550), rice (PR 121, PR 114, PR 126) and barley (PL 807, PL 89 and DWRB 123) were procured from the Department of Plant Breeding and Genetics and School of Organic Farming, Punjab Agricultural University, Ludhiana, India. The seeds were soaked for 10 hours and germinated for 12 hours, then washed under tap water and broadcasted in plastic trays using the same soil for each of the varieties with no fertilizer application. The trays were kept indoor at room temperature 20-22°C. The seeds were watered 3-4 times at an interval of 4 hours daily. The grasses were cut with scissors 1cm above the soil when they reached the desired length i.e., 10 cm, 15 cm and 20 cm. The different cereal grasses attained the desired length at different durations of days. Wheat varieties attained the length of 10 cm by 5-6 days, 15 cm by 7-8 days and 20 cm by 10-11 days. Rice varieties attained 10 cm by 8-9 days, 15cm by 10-11 days and 20 cm by 15-16 days. Whereas, barley varieties attained 10 cm by 6-7 days, 15 cm by 8-9 days and 20 cm by 10-11 days.

Total phenol and total flavonoid content: TPC was determined using Folin Ciocalteu method (Singleton et al 1999) and expressed as GAE (Gallic acid equivalent)/100g. TFC was estimated using the method of (Zhishen et al 1999) and expressed as mg QE (Quercetin equivalent)/100g.

Antioxidant potential of cereal grasses: Antioxidant potential of cereal grasses was estimated by FRAP, ABTS and DPPH (2,2-diphenyl-1-picrylhydrazyl) assay. FRAP assay of the extract (Tadhani et al 2007). The radical scavenging assay was determined by ABTS and DPPH assay following the protocol of (Re et al 1999) and (Brand-Williams et al 1995), respectively. The results were expressed as mg ferrous sulphate equivalent per 100 g (mg $FeSO_4$ equivalent/100g) for FRAP and % RSA (radical scavenging activity) for ABTS and DPPH assay.

Statistical analysis: The mean and standard deviation were computed using Microsoft Excel (2010) Statistical Analysis Tool Pack. Two-way Analysis of Variance was done using SAS (Statistical Analysis System, version 9.3 for windows) Institute Inc. A p-value of less than 0.05 was considered statistically significant in all cases.

RESULTS AND DISCUSSIONS

The TPC and TFC of different cereal grasses has been shown in Table 1. There was no significant difference in TPC and TFC among the wheat varieties with mean values ranging from 110.41-117.38 mg GAE/100g and 90.76-101.25 mg QE/100g fresh weight respectively (Table 1). Among the rice varieties a higher TPC and TFC was observed in variety PR 126 with a mean value of 56.78 mg GAE/100g and 81.99 mg QE/100g fresh weight respectively. Barley varieties showed significant difference in both TPC, with variety DWRB 123 exhibiting higher mean value of 91.05 mg GAE/100g fresh weight and TFC with variety PL 891 showing higher mean value of 72.63mg QE/100g fresh weight.

Table 1. Total phenol content (mg GAE/100g) and total flavonoid content (mg QE/100g) of different cereal grasses

Crop/variety	TPC	TFC
Wheat		
Unnat PBW 550	114.11^{a}	101.25^{ab}
PBW 343	110.41^{a}	90.76^{b}
PBW 550	117.38^{a}	95.78^{b}
Rice		
PR 121	48.87^{a}	72.83^{a}
PR 114	45.67^{a}	65.64^{b}
PR 126	56.78^{b}	81.99^{c}
Barley		
PL 807	71.61^{a}	69.20^{ac}
PL 891	88.97^{b}	72.63^{b}
DWRB 123	91.05^{c}	67.79^{c}

Values in columns followed by different superscripts differ significantly ($p \leq 0.05$)

Table 2. Total phenol content (mg GAE/100g) and total flavonoid content (mg QE/100g) of different cereal grasses harvested at different growth lengths

Crop/Length	10 cm	15 cm	20 cm
Total phenol content			
Wheat	106.16^{a}	113.46^{a}	122.28^{b}
Rice	63.94^{a}	57.10^{b}	30.28^{c}
Barley	77.48^{ac}	94.58^{b}	74.87^{c}
Total flavonoid content			
Wheat	70.36^{a}	97.97^{b}	119.47^{c}
Rice	95.18^{a}	71.28^{b}	54.00^{c}
Barley	65.30^{ac}	75.91^{b}	66.28^{c}

Values in columns followed by different superscripts differ significantly ($p \leq 0.05$)

The maximum TPC (122.28 mg GAE/100g) and TFC (119.47 mg QE/ 100g) , was observed in 20 cm in wheat grass. In rice grass, 10 cm length grass exhibited maximum TPC (63.94 mg GAE/100g) and TFC (95.18 mg QE/100g) . Among the barley varieties maximum TPC was (94.58 mg GAE/100g) and was 75.91 mg /100g) was obtained at 15 cm.

The varieties of cereal grasses used in the study showed significant difference in their FRAP activity (Fig. 1) Among the wheat varieties the highest FRAP activity (198.47 mg $FeSO_4$ /100g) was exhibited by variety PBW 550. PR 126 showed highest value of 134.70 mg $FeSO_4$ /100g among the rice varieties while among the barley varieties the highest value (64.05 mg $FeSO_4$ /100g) was observed in PL 807. When the ABTS and DPPH scavenging activity among wheat varieties was considered, no significant difference was observed (Fig. 2 and 3). The rice variety PL 126 exhibited higher ABTS activity of 19.87% while no significant difference in DPPH activity In barley variety PL 807 exhibited higher ABTS activity of 16.06 % RSA while not showing significant difference in DPPH activity. Considering the effect of length of cereal grasses on the antioxidant activity, wheatgrass showed highest FRAP activity of (186.87 mg $FeSO_4$ /100g) at 20 cm while lowest activity (163.48 mg $FeSO_4$ /100g) was observed at 10 cm. FRAP activity was maximum (140.86 mg $FeSO_4$ /100g) at 10 cm in rice grass, while barley grass did not show significant difference when growth length was considered. Similarly, the highest ABTS value was observed at 20 cm in wheatgrass with an activity of 52.09 % RSA. The rice grass did not show significant difference. In barley grass however, higher ABTS activity was observed at 15 cm with activity of 18.30% RSA. No significant difference in DPPH scavenging activity in wheat and rice grass when length was considered whereas, activity at 20 cm was found to be lower in barley grass.

The cereal grasses showed significant difference in bioactive compounds and antioxidant potential. The bioactive compounds, in wheat varieties obtained in the present study are similar to that reported in previous studies (Qamar et al 2019, Devi et al 2020) and the bioactive compounds increased with the increase in length of the grass with optimum bioactive compounds observed at 20 cm which concurs with previous study (Savsatli 2020, Devi et al 2020). Among the rice varieties higher bioactive compounds was observed in 10 cm length grass . Rattanapon et al(2017) also recorded that TPC in rice grass decreased as the seedling time increased.

Antioxidant potential (DPPH) in present study among wheat and barley grasses was lower than that reported by Qamar et al (2019)being 99.29 and 51.69% for DPPH radical scavenging activity in barley and wheatgrass respectively which could be due to varietal differences and the growing condition. The antioxidant activities determined by FRAP was maximum at 10 cm in the rice grass, while ABTS and DPPH scavenging activity did not show significant difference till 20 cm which was similar to results obtained by (Rattanapon et al 2017) where antioxidant activity determined as ABTS and FRAP was higher in rice aged 7 days compared with 14 and 21 days whereas, the DPPH radical scavenging activity was higher on day 14 than on day 7 and 21. The higher content of antioxidant activities in early stages could be attributed to a high oxygen demand during early germination. TPC increased during seedling growth, which results in a significant increase in antioxidant potential

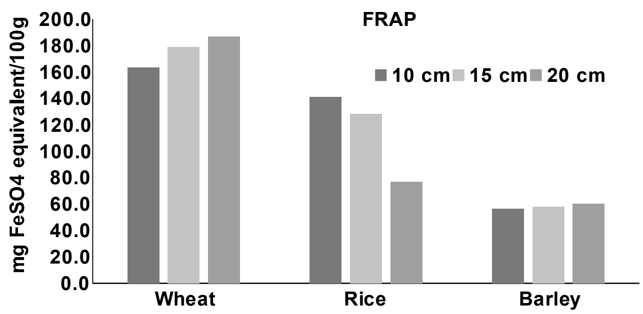

Fig. 1. FRAP-reducing activity of cereal grasses harvested at different growth lengths

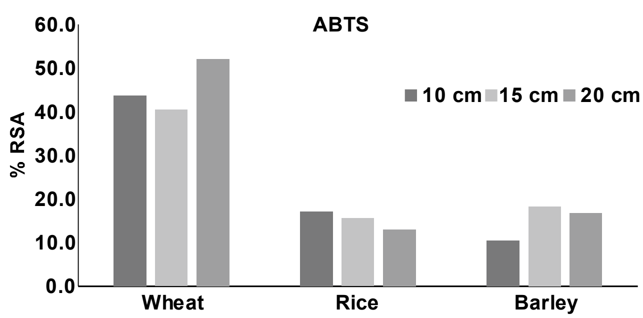

Fig. 2. ABTS- free radical scavenging activity of cereal grasses harvested at different growth lengths

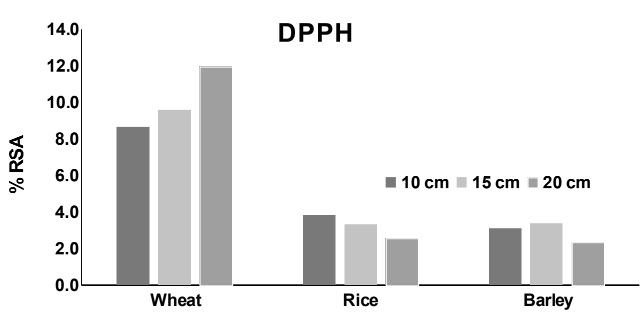

Fig. 3. DPPH- free radical scavenging activity of cereal grasses harvested at different growth lengths

as reported by Zhang et al (2015). Previous studies on wheatgrass by earlier scientist reported correlation between antioxidant activity and bioactive compounds which was also observed in the present study(Shakya et al 2014, Savsatli 2020, Kaur et al 2021).

CONCLUSIONS

The work done in this paper indicates that among the analysed cereal grasses of wheat, rice and barley, significant difference was observed in the bioactive compounds and antioxidant activity of different varieties at different lengths. Wheatgrass had maximum bioactive compounds and exhibited maximum antioxidant activity followed by rice and barley grasses. In addition, the study suggests that different grasses should be harvested at the growth length when the bioactive compounds and antioxidant activities are optimum. Moreover, as grasses of rice and barley varieties also exhibit notable antioxidant potentials, they could be considered as a substitute for wheatgrass to meet the need of the population for health enhancing therapeutic products.

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Community Structure of Macrobenthic Fauna in Achenkovil River, Southern- Western Ghats, Kerala, India

S. Sujitha and R. Sreejai
Department of Zoology, St. Stephen'S College, Pathanapuram
University of Kerala-689 695, India
E-mail: sujithashylesh7020@gmail.com

Abstract: A study on the water quality parameters and community structure of the macrobenthic fauna was carried out in the Achenkovil river basin, Kerala. Nine sampling sites were selected for the study and the macrobenthic fauna was collected using Van Veen Grab (0.025m2). Both the water and sediment samples were collected bimonthly and seasonally. Fourteen water quality parameters were analyzed to monitor the influence of water quality on the community structure of macrobenthos. The maximum mean value for DO and silicate was in station 1 (S1). The water temperature, BOD, turbidity, conductivity, TDS, salinity, alkalinity, hardness, pH, nitrate, and phosphate have their maximum value in the downstream stretches of the river. A total of 3563 macrobenthic individuals belonging to 8 orders, 32 families, 32 genera, and 32 species were collected and identified. Among the macrobenthic community, the order Ephemeroptera showed the maximum abundance (860 Ind/m^2) and minimum for Zygoptera (63 Ind/m^2). Station S1, part of a pristine forest region, is characterized by rich benthic diversity and abundance. The pollution-sensitive taxa such as Ephemeroptera, Plecoptera, and Trichoptera were the dominant members of the community structure in S1. Their presence is an indication of good water quality. The midstream and downstream segments of the river are facing severe anthropogenic stress. An increase in the abundance of pollution-tolerant organisms such as Chironomids in the midstream and downstream segments of the river is an indication of deteriorating water quality. The diversity, distribution, and abundance of the macrobenthic community were highly influenced by the variations in water quality due to various natural as well as anthropogenic impacts.

Keywords: Biogeochemical cycles, Diversity, Freshwater, Invertebrates, Species richness

Benthos plays a vital role in the functioning of an ecosystem (Iyagbaye et al 2017). They serve as the food source for most aquatic organisms. Freshwater sediments serve as the home for a diverse group of benthic invertebrates with rich diversity and abundance but their distribution is uneven which creates sampling difficulty. They decompose complex organic matter into simple absorbable forms, oxygenate the underlying sediments, and thus play an efficient role in biogeochemical cycles (Basu et al 2018). Interruptions among the complex sediment-dwelling benthos and associated food web sometimes cause a sudden change in the equilibrium setup of the environment (Poikane et al 2016). Thus, the species richness, diversity, abundance, distribution, and functional importance of benthic invertebrates remain unnoticed until unexcepted changes occur in the ecosystem. Anthropogenic activities play a negative role in the species richness of benthic macroinvertebrates (Mola and Gawad 2014). The structural assemblage of the macrobenthic community is very complex and includes a variety of organisms from microbes to phytobenthos and zoobenthos and covers different levels of the food web (Idowu and Funso 2019). The study on the macrobenthic community structure reveals differences in species composition, abundance, biomass, and distributional patterns in various aquatic ecosystems (Zabby and Hart 2006). Knowledge about diversity, abundance, richness, evenness, and community structure are important parameters to determine the natural or anthropogenic changes in the water body concerning time (Jun et al 2016). The macrobenthic fauna shows uneven distributional patterns in riverine ecosystems (Basu et al 2018). The physicochemical parameters influence the structural assemblage of the macrobenthic community (Zabby and Hart 2006). As they are slow-moving, they tend to remain in their original habitat with great acclimation potential (Sandin 2000). They can withstand changes in water quality and a high amount of pollution. High loads of pollution in the water body cause an increase in the number of tolerant species, thus increasing the abundance of particular species and decreasing the diversity and species richness. They serve as an efficient tool to evaluate water quality and are commonly used in biomonitoring programs. Thus, they are considered good bio-indicators for the environmental changes in any aquatic ecosystem. Abdel and Gawad (2019) observed that macrobenthic invertebrates are the most ideal indicators for biomonitoring and provide an ecological outline of the present status of the river. Similar studies on the influence of water quality parameters and the community structure of macrobenthos were carried out by several workers (Mophin-Kani and Murugesan (2014), Nautiyal et al 2017, Basu et al 2018, Kamal et al 2021, Mishra et al 2022, Priyanka and Prasad (2022), and Sekhar (2022)). No one has yet attempted to study the macrobenthic fauna of the Achenkovil river. The present study was carried out in the Achenkovil river to ascertain the composition and structural diversity of macrobenthic fauna, the environmental factors and anthropogenic impacts responsible for the community patterns.

MATERIAL AND METHODS

Study area: This study includes the Achenkovil river, Southern Western Ghats, Kerala, India. The river drains through highly varied geological formulations and covers the highland, midland and lowland physiographic provinces of the state. About 60% of the highland is occupied by dense forest, 5% by degraded forest and 10% is agricultural land. Nearly 40% of the Midland region is under double-crop paddy cultivation. The lowland region is a narrow strip of land along the West Coast and is occupied by 80% agricultural land (mixed agricultural/horticultural plantation) and 10% under double crop paddy cultivation. The rest of the area is occupied by water bodies. The study area experiences a tropical climate with three distinct seasons-premonsoon (February- May), monsoon (June-September) and postmonsoon (October- January). Floods are common in the midland and lowland regions during the monsoon months. In non-monsoon months, as the freshwater flow decreases, salinity intrusion occurs in the lowland tracks of the river making the river water saline. This adversely affects the biotic community and creates a lot of technical problems in this region.

Sampling sites: Samples were collected bimonthly and seasonally in premonsoon (February-May), monsoon (June-September), and postmonsoon seasons (October- January), early in the morning hours (06.00 -11.30 h) throughout the study period (2018-2020). The entire river body is divided into three segments- upstream with 9° 07' 39.53' N and 77° 07' 58.56' E with an elevation of 870 ft a.m.s.l, midstream- 9° 13' 59.37' N and 76° 40' 38.4' E with an elevation of 66 ft a.m.s.l, and downstream with 9° 19' 29.07' N and 76° 26' 54.31' E with an elevation of 6 ft a.m.s.l- with three stations in each segment of the river (Total 9 sampling sites along the entire stretch of the river) (Fig. 1).

Identification of macrobenthic fauna: Macrobenthic fauna was collected using Van Veen grab (0.025 m^2). Triplicate samples were taken for precision. The grab samples collected were sieved through a series of mesh sieves-3000 µm (3mm), 2000 µm (2 mm), 1000 µm (1mm), and 500 µm (0.5 mm) mesh and the sediments retained in the 0.5 mm sieve was washed, and carefully transferred to a white plastic tray and was sorted out. All the collected samples were preserved in 4% formalin for subsequent analysis. In the laboratory, the preserved sample was examined using a stereomicroscope (Magnus MSZ- BI LED) and identified using standard taxonomic literature- Young and Yule (2004), Dudgeon (1999), Thorp and Covich's Freshwater Invertebrates (2015), Merrit and Cummins (1996).

Physicochemical analysis: Water temperature (°C) was measured *in situ* by using a Mercury thermometer (with ± 0.1°C accuracies). The samples for DO and BOD were fixed with alkaline potassium iodide and manganous sulphate at the site itself. The water samples were then carried immediately to the laboratory for further analysis. The water samples were collected using clean polyethylene bottles and carried immediately to the laboratory for further analysis. DO (mg/l), BOD (mg/l), pH, turbidity (NTU), conductivity (µS/cm), salinity (ppt.), alkalinity (mg/l), hardness (mg/l), TDS (mg/l), phosphate (mg/l), silicate (mg/l) and Nitrate (mg/l) was carried out using standard references (APHA 2017).

Statistical analysis: Multivariate statistical analysis such as PCA was employed to study the variation in environmental parameters (PCA) and the relationship between macrobenthic fauna and environmental parameters were carried out using CCA. Community structure was assessed using biodiversity indices and Engelmann's scale. Biodiversity indexes and CCA were carried out using PAST (Version 4.09) software. PCA was carried out using SPSS (Version 22).

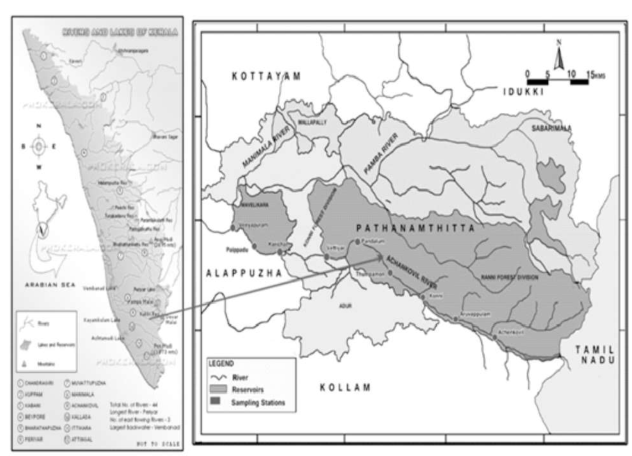

Fig. 1. Map showing the study sites (Stations S1 to S9) in the Achenkovil River basin, Kerala. The upstream sites include Achenkovil (S1), Aruvappulam (S2), and Konni (S3); Midstream includes Thumpamon (S4), Pandalam (S5), and Vettiyar(S6) and the Downstream includes Karichal (S7), Payippad (S8), and Veeyapuram (S9)

RESULTS AND DISCUSSION

Environmental variables: The variation in the environmental parameters was analyzed using the multivariate statistical technique (PCA) (Table 1 and 2). The PCA showed four principal components, which explained 77.98% of the total variance. PC1 explained 27.14% of the total variance and had a significant contribution from TDS, conductivity, salinity, and hardness with a strong positive loading value of >0.75. PC2 accounted for 20.28% of the total variance and has a strong positive correlation with BOD, DO, water temperature, and silicate. PC3 accounted for 16.53% of the total variance and had a strong positive correlation with depth, turbidity, phosphate, and nitrate. PC4 accounted for 14.04% of the total variance and had a strong positive correlation with pH and alkalinity (Table 3). Absolute loading value >0.75 is of strong significance and these parameters can be used to monitor the variations in water quality (Liu et al 2003).

Temperature is an important factor that plays a major role in the physical, chemical, and biological characteristics of water. The water temperature in the present study varied from 21.80°C to 30.20°C with the maximum mean value noted in S9 which may be due to high solar radiation, lack of

Table 3. Shows the variation in the environmental parameters analyzed using principal component analysis

Parameters	Components			
	1	2	3	4
Water temp.	0.179	0.756	0.217	0.376
Depth	-0.362	0.399	0.621	0.079
DO	-0.419	-0.76	0.283	0.076
BOD	-0.09	0.822	-0.094	0.209
pH	0.284	0.135	-0.102	0.778
Turbidity	0.271	-0.09	0.626	0.5
Conductivity	0.812	0.246	-0.05	0.372
Salinity	0.848	0.323	0.124	0.252
Alkalinity	0.265	0.259	0.052	0.763
Hardness	0.858	0.138	0.038	0.294
TDS	0.87	0.043	0.232	0.148
Phosphate	0.466	-0.07	0.751	-0.09
Silicate	-0.371	-0.74	0.053	-0.05
Nitrate	0.027	-0.18	0.865	-0.09
Eigen values	3.799	2.839	2.314	1.965
% of variance	27.14	20.28	16.53	14.04
Cumulative %	27.14	47.41	63.94	77.98

Table 1. Water quality parameters of Achenkovil river

Stations	Water temp. (°C)	Depth (m)	DO (mg/l)	BOD (mg/l)	Turbidity (NTU)	TDS (mg/l)	Conductivity (µS/cm)
S1	23.91 ±2.06	0.90±0.19	6.77±0.97	1.39±0.79	4.83±4.79	143.54±111.5	74.95±14.45
S2	25.26±1.55	1.32±0.81	6.48±0.97	1.65±0.67	5.04±2.99	118.38±43.80	64.13±13.24
S3	25.33±1.21	2.43±0.23	5.90±1.09	2.10±0.76	5.48±2.17	107.90±37.58	59.57±12.80
S4	26.23±1.10	3.49±0.20	6.16±0.92	2.20±0.64	5.52±2.71	104.81±41.70	66.89±14.29
S5	26.92±1.00	4.11±0.21	6.26±0.95	2.93±1.60	6.69±1.80	108.13±39.56	72.54±21.03
S6	26.99±0.79	3.19±0.35	5.90±1.07	2.90±1.64	6.96±2.25	126.78±40.50	75.94±19.08
S7	27.25±0.72	3.85±0.35	5.54±0.87	2.61±0.81	7.93±2.35	128.70±41.94	82.78±11.74
S8	28.10±0.99	3.15±0.22	5.25±1.13	2.94±0.60	8.58±2.79	250.59±127.7	173.60±167.13
S9	28.65±1.27	2.08±0.24	4.95±0.94	2.87±0.63	8.75±2.50	283.98±132.04	183.07±172.09

Table 2. Water quality parameters of Achenkovil river

Stations	Salinity (ppt.)	pH	Hardness (mg/l)	Alkalinity (mg/l)	Nitrate (mg/l)	Phosphate (mg/l)	Silicate (mg/l)
S1	0	6.85±0.30	14.90±3.75	10.67±2.39	0.74±0.16	0.48±0.17	3.52±0.54
S2	0	6.74±0.19	15.40±3.01	10.93±2.42	0.92±0.16	0.49±0.14	2.73±0.54
S3	0	6.81±0.12	14.49±5.30	12.46±3.22	0.87±0.23	0.58±0.15	2.89±0.60
S4	0	6.78±0.18	17.34±3.46	11.86±2.87	0.92±0.22	0.66±0.17	3.12±0.74
S5	0	6.87±0.22	14.07±3.02	11.81±3.34	0.94±0.18	0.55±0.19	3.07±0.65
S6	0	6.87±0.36	14.15±2.50	12.40±2.50	0.99±0.21	0.60±0.20	2.46±0.34
S7	0.01±0.01	6.81±0.39	13.14±2.11	11.81±2.24	0.88±0.21	0.56±0.18	2.06±0.53
S8	0.26±0.16	6.94±0.35	30.50±23.09	14.37±3.56	1.01±0.17	0.80±0.24	2.05±0.45
S9	0.27±0.17	7.13±0.24	32.97±23.49	14.68±2.97	1.08±0.20	1.08±0.17	2.11±0.66

canopy cover, low rainfall, low water levels, and clear skies (Abilash and Mahadevaswamy 2021). The minimum mean water temperature was in S1, the headwater station has a thick canopy cover that prevents the direct heating of the surface water. The depth is in the range of 0.65m to 4.39m with maximum depth from S5. Sand mining by the locals residing near the river banks for house constructions may be a reason for an increase in depth. A high temperature causes a decrease in the DO level (in S9) which is a natural phenomenon, since warmer water was more easily saturated with oxygen and thus holds less DO (Yang et al 2021). The high DO in S1 may be due to water turbulence (Kannel et al 2007) resulting from the rugged topography of the river basin. The increase in pH value in S9 may be due to high photosynthetic activity (Craft et al 2018). The pH value shows a clear trend toward alkalinity which may be due to anthropogenic impacts, wastewater discharge and agricultural activities (Azouzi et al 2017). Heavy rain in the monsoon season causes surface runoff accompanied by sand, silt, clay, organic matter, etc. may be the reason for high turbidity in S9 during the monsoon season (Sanalkumar et al 2014). The mean turbidity was greater than the BIS permissible limit (5 NTU). An increase in turbidity is considered a limiting factor in the biological productivity of aquatic ecosystems (Mahajan and Billore 2014). Agricultural runoff resulting from heavy rainfall may be the reason for an increase in the value of nitrate and phosphate during the monsoon season (Varol et al 2012). The input of more silicious sediments along with surface runoff may be the reason for the high silicate content in the water body (Jaji et al 2007). The high BOD value noted in S8 may be due to low rainfall, low water flow, and high temperature (Girija et al 2007). The conductivity and salinity were maximum in S9. The intrusion of saline water from Kayamkulam lake into S8 and S9 during the premonsoon season may be the reason for high salinity and a corresponding increase in conductivity in the water body.

Macrobenthic community: The composition and distribution of macrobenthic fauna in the present study include a total of 3563 individuals belonging to 8 orders, 32 families, 32 genera, and 32 species (Table 4). The 8 orders include Ephemeroptera, Plecoptera, Zygoptera, Anisoptera, Coleoptera, Diptera, Hemiptera, and Trichoptera. The species composition of different orders of macrobenthic fauna revealed that the largest group was Ephimeroptera comprising 8 species, followed by 7 species of Coleoptera, 4 species each for Diptera, Hemiptera, Anisoptera. The order Ephemeroptera accounted for 24% of the total macrobenthic fauna and was the most dominant, diverse, and abundant group. This order was represented by 8 families. The second largest group was Coleoptera with 7 families, followed by Anisoptera, Diptera and Hemiptera with 4 families each, Trichoptera with 3 families and Plecoptera and Zygoptera with a single family. The least represented order was Zygoptera, which accounted for only 2% of the total macrobenthos studied (Fig. 2).

The abundance, relative abundance and dominance status of macrobenthic fauna studied in the Achenkovil river basin were calculated using Engelmann's scale (Table 4). The species *Notophlebia jobi, Caenis* sp.*, Baetis* sp. of order Ephemeroptera*, Neoperla* sp.*,* of order Plecoptera, *Chironomous* sp.*, Atherix* sp.*,* of order Diptera, *Micronecta* sp., of order Hemiptera, *Stylogomphus* sp.*,* of order Anisoptera *Eubrinax* sp.*, Cylloepus* sp.*, Hydrophilus* sp.*,* of order Coleoptera*, Economous* sp.*, Chimarra* sp.*, Hydropsyche* sp. of order Trichoptera (Relative abundance (RA%) range 3.2 to 10%) are the most abundant and sub-dominant species. *Dudgeodes* sp., *Sparsorythus gracillis, Afronurus kumbakkaraiensis, Torleya nepalica and Aethephemera nadiinae* of order Ephemeroptera, *Tabanus* sp. of order Diptera, *Microvelia douglasi, Lethocerus indicus, Nepa* sp. of order Hemiptera*, Euphae* sp. of order Zygoptera, *Anax* sp., *Corydalus* sp., *Crocothemis* sp. of order Anisoptera, *Agabus* sp. of order Coleoptera (RA % range from 1.1 to 3.1%) are the recedent species, *Tipula* sp.of order Diptera, *Rhyssemus* sp., *Hydrocanthus* sp.and *Hydrena* sp. of order Coleoptera (RA% less than 1%) are reported as sub-recedent species.

Canonical correspondence analysis: The relationship between the macrobenthic fauna and environmental variables (Fig. 3) was depicted using multivariate statistical analysis (CCA). The first canonical axis explained over 47.85% (Eigenvalue, 0.122) and the second 31.72% (Eigenvalue, 0.081) of the variation in the macrobenthic fauna data set. The Monte Carlo permutation test performed on the first two axes showed no significant differences. The CCA reveals that water quality parameters such as depth, water temperature, BOD, turbidity, TDS, conductivity, salinity, pH, hardness, alkalinity, nitrate, and phosphate show a positive correlation with macrobenthic groups such as Diptera and Anisoptera. The DO and silicate show a negative correlation with Ephemeroptera, Plecoptera, Hemiptera, Zygoptera, Coleoptera and Trichoptera in the first canonical axis. The second canonical axis revealed that water temperature, BOD, turbidity, TDS, conductivity, salinity, pH, hardness, alkalinity, nitrate, and phosphate shows a positive correlation with macrobenthic groups such as Plecoptera, Diptera, Hemiptera, Zygoptera, Coleoptera, Trichoptera whereas depth, DO and silicate shows a negative correlation with Ephemeroptera and Anisoptera. The studied physicochemical parameters have a strong influence on the community structure of macrobenthic fauna. From the CCA ordination plot, it is clear that the macrobenthos shows spatial variation concerning their environmental requirements. Dipterans commonly chironomids were one of the dominant taxa in natural (Copatti et al 2013) or non-natural environments (Hepp et al. 2010). Most of the members of EPT and Coleoptera are commonly known for their pollution-sensitive nature. They are absent from highly disturbed habitats.

The station-wise analysis reveals that the total number of taxa ranged from 4 (S9) to 32 (S1), individuals from 37 (S9) to 1461 (S1) Ind/m^2, dominance ranged from 0.043 (S3) to 0.661 (S9), Simpson from 0.339 (S9) to 0.957 (S3), Shannon from 0.73 (S9) to 3.28 (S3), Evenness from 0.44 (S8) to 0.86 (S3) and Margalef from 0.83 (S9) to 5.17 (S3) (Table 5). This study gave a reference state of the structural composition of the macrobenthic fauna of the Achenkovil river basin. Aquatic insects form the major group in the community structure of macrobenthos studied, and this is similar to the observations made by (Arimoro et al 2015). Abhilash and Mahadevaswamy (2021) observed that aquatic insects are generally considered the dominant macro-invertebrates in freshwater ecosystems. Spatio-temporal variations have a

Table 4. Relative abundance and dominance status of macrobenthic fauna in the Achenkovil river basin

Order	Family	Genus/Species	Abundance	Relative abundance	Status
Ephemeroptera	Leptophlebidae	*Notophlebia jobi*	171	4.80	Sub-dominant
	Caenidae	*Caenis* sp.	169	4.74	Sub-dominant
	Teloganodidae	*Dudgeodes* sp.	87	2.44	Recedent
	Baetidae	*Baetis* sp.	153	4.29	Sub-dominant
	Tricorythidae	*Sparsorythus gracillis*	74	2.08	Recedent
	Heptageniidae	*Afronurus kumbakkaraiensis*	94	2.64	Recedent
	Ephemerellidae	*Torleya nepalica*	65	1.82	Recedent
	Ephemeridae	*Aethephemera nadiinae*	47	1.32	Recedent
Plecoptera	Perlidae	*Neoperla*	175	4.91	Sub-dominant
Diptera	Chironomidae	*Chironomus* sp.	275	7.72	Sub-dominant
	Athericidae	*Atherix* sp.	125	3.51	Sub-dominant
	Tipulidae	*Tipula* sp.	34	0.95	Sub-recedent
	Tabanidae	*Tabanus* sp.	39	1.09	Recedent
Hemiptera	Notonectidae	*Micronecta* sp.	114	3.20	Sub-dominant
	Vellidae	*Microvelia douglasi*	87	2.44	Recedent
	Belostomatidae	*Lethocerus indicus*	98	2.75	Recedent
	Nepidae	*Nepa* sp.	75	2.10	Recedent
Zygoptera	Euphaeidae	*Euphae* sp.	63	1.77	Recedent
Anisoptera	Gomphidae	*Stylogomphus* sp.	290	8.14	Sub-dominant
	Aeshnidae	*Anax* sp.	78	2.19	Recedent
	Corydalidae	*Corydalus* sp.	39	1.09	Recedent
	Libellulidae	*Crocothemis* sp.	80	2.25	Recedent
Coleoptera	Dytiscidae	*Agabus* sp.	41	1.15	Recedent
	Psephenidae	*Eubrinax* sp.	183	5.14	Sub-dominant
	Elmidae	*Cylloepus* sp.	160	4.49	Sub-dominant
	Scarabaeidae	*Rhyssemus* sp.	20	0.56	Sub-recedent
	Hydraenidae	*Hydrena* sp.	16	0.45	Sub-recedent
	Hydrophilidae	*Hydrophilus* sp.	120	3.37	Sub-dominant
	Noteridae	*Hydrocanthus* sp.	26	0.73	Sub-Recedent
Trichoptera	Economidae	*Economous* sp.	142	3.99	Sub-dominant
	Philopotamidae	*Chimarra* sp.	175	4.91	Sub-dominant
	Hydropsychidae	*Hydropsyche* sp.	248	6.96	Sub-dominant

Table 5. Spatial variation of biodiversity indices in the Achenkovil river basin

Stations	Dominance _D	Simpson _1-D	Shannon _H	Evenness _e^H/S	Margalef
s1	0.045	0.955	3.23	0.79	4.254
s2	0.056	0.944	3.124	0.784	4.541
s3	0.043	0.957	3.288	0.864	5.179
s4	0.065	0.935	3.01	0.7	4.328
s5	0.118	0.882	2.704	0.622	4.122
s6	0.085	0.915	2.719	0.722	3.961
s7	0.094	0.906	2.551	0.713	3.416
s8	0.471	0.529	1.269	0.445	1.789
s9	0.661	0.339	0.73	0.519	0.831

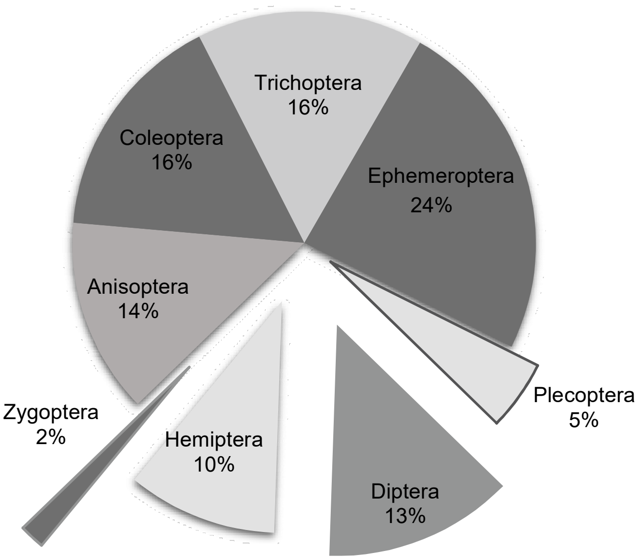

Fig. 2. Relative abundance of major orders of macrobenthic fauna studied

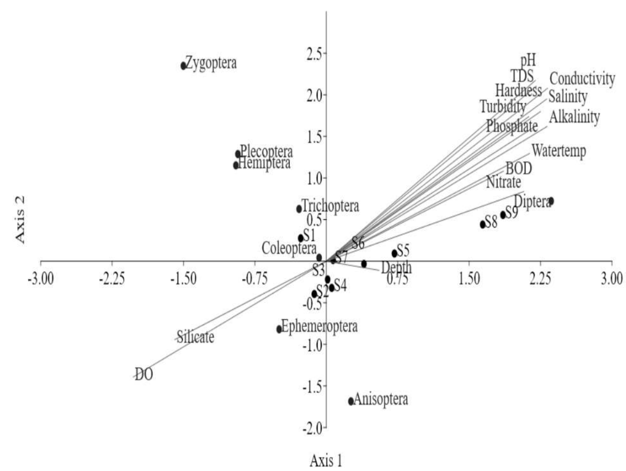

Fig. 3. Relationship between the macrobenthic fauna and the environmental parameters

strong influence on the community structure of macrobenthic fauna (Sasikala et al 2017). The structure and composition of biotic community change with the physicochemical and hydrobiological characteristics of the environment which is often reflected in the distribution, diversity and abundance pattern of species (Abhilash and Mahadevaswamy 2021). The Ephemeroptera had the highest number of species represented by 8 genera, accounting for 24% of the total macrobenthic fauna studied (Fig. 4). Leptophlebidae (19.88%) and Caenidae (19.65%) were the most abundant family among Ephemeroptera. The second largest group was Coleoptera (16%) with 7 families, followed by Trichoptera (16%), Anisoptera (14%), Diptera (13%), Hemiptera (10%), Coleopterans are abundantly seen in sites with good vegetation as it provides food and breeding places. They can also tolerate moderate levels of pollution (Popoola et al 2019). Chironomus are pollution tolerant and they dominated in sites with high turbidity, TDS, conductivity, and even low DO values. It can withstand hypoxic conditions (Popoola et al 2019). An increase in the number of Chironomus species is also an indication of anthropogenic stress . and are commonly used as bioindicators in water quality monitoring programs (Al- Shami et al 2010). The diversity and abundance of odonates in an area depend on the habitat heterogeneity formed due to the complexity of vegetation, the nature of the substrate, and physicochemical characteristics (Wijesooriya et al 2022). The Ephemeroptera shows higher species richness and abundance at the reference site and a reduction in species number and diversity towards the midstream and downstream stretch of the river. The abundance of some families of Ephemeroptera like Heptagenidae, Tricorythidae, Leptophlebiidae, Teloganodidae, etc indicates good water quality and the undisturbed forest habitat along the river banks and within the catchment in the reference site. *Baetis* and *Caenis* were present in the downstream segment of the river, as they can tolerate moderate levels of pollution. The decrease in the macrofaunal composition towards the midstream and downstream stretch of the river is an indication of pollution load and the corresponding deterioration of the water quality (Kumar et al 2012). Plecoptera and Trichoptera are sensitive to water quality degradation and occur only in clean and well-oxygenated water (Priyanka and Prasad 2014). They are abundant in the reference site (S1). Specific families within the Ephemeroptera, Plecoptera and Trichoptera (EPT taxa) help to monitor various types of disturbance in the water body (Abhijna et al 2013)). The spatial variation in the water quality status can be revealed from the values recorded for species diversity, richness, dominance and evenness indices. The highest values for Simpson, Shannon, Evenness and Margalef were noted in S3 and Dominance in S9. High dominance in S9 may be due to the disappearance of more sensitive taxa replaced with more tolerant species like Chironomous, thus reducing species richness and diversity (Copatti et al 2013). The highest diversity of macrobenthic fauna in the reference site may be due to the thick canopy cover that lowers the atmospheric and water temperature and provides diverse habitats for a variety of macrobenthic fauna leading to increased diversity. The Shannon index value for stations 5 to 9 and the Margalef index value for stations 8 and 9 were below three, which is an indication of the polluted water body. Similar reports were given by Kabir and Offioong (2016), in the Alaro stream, Ibadan. The low relative abundance of pollution-sensitive organisms, in the midstream and downstream segments, indicates that the river Achenkovil is already stressed across its reaches. However, water quality was more impacted during the rainy season, due to surface runoff. The CCA revealed that most taxa were sensitive to environmental changes. More sensitive macrobenthic fauna was in the upstream stations than in the midstream and downstream stations, as these species are favored by more DO levels and lower levels of conductivity, nitrate and phosphate. It is clear from the CCA plot that most of the Hemipterans are associated with high water temperature and less dissolved oxygen, indicating their less dependency on oxygen. Moreover, the Hemipterans possess additional respiratory structures such as plastron, siphon, etc. that help us to use atmospheric oxygen (Abhilash and Mahadevaswamy 2021).

CONCLUSION

The diversity, distribution pattern and abundance of macrobenthic fauna are highly influenced by environmental variables. Some species of macrobenthic fauna are pollution sensitive and some are pollution tolerant. So, their presence or absence can be used to predict water quality. To conclude, macrobenthic fauna has the potential to act as biological indicators of pollution status. Thus, keeping in mind the importance of the study, steps should be taken for the maintenance and conservation of freshwater ecosystems.

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Diseases Infesting Forage Alfalfa (*Medicago sativa* L.) and Management

Hana'a Burezq

Desert Agriculture and Ecosystems Program, Environment and Life Sciences Research Centre
Kuwait Institute for Scientific Research, PO Box 24885 Safat 13109, Kuwait
E-mail: mdavidson@kisr.edu.kw

Abstract: Alfalfa or lucerne, *Medicago sativa,* is one of the important legumes grown as a fodder crop providing feed hay with valuable nutrients. It is the principal source of fibre and protein in animal diets, thus plays an important role in the dairy industry. Alfalfa can significantly accumulate nitrogen by its deep rooting system and fix atmospheric nitrogen by biological fixation. Alfalfa plant stand covers the soil to minimize erosion by water or wind. Also grown as a rotational crop, Alfalfa provides the soil with nutrients and organic matter and the foliage acts as soil manure. Alfalfa is produced in several countries such as the USA, Australia, Europe and the Middle East. Pests, including insects, diseases, and nematodes, affect alfalfa hay productivity, considerably reducing yield, plant stand life, and fodder quality. Alfalfa is subjected to several infectious diseases causing severe menace, that can limit forage production, decline the fodder quality and restrict stand persistence. Many times, they cause sudden stand loss, delayed regrowth after cutting, ultimately stunted alfalfa stand. Managing the diseases is an important aspect of economical alfalfa production. A thorough understanding of the disease status, their seasonal incidence, damage symptoms and management strategies is an important tool in effective alfalfa fodder production. The current review highlights the different types of diseases caused by diverse pathogens infesting alfalfa crop, the symptoms of their infestation and managing the diseases organically in an environmentally safe manner.

Keywords: Alfalfa, Lucerne, Legume, Fodder, Disease, Pathogen

Alfalfa, *Medicago sativa,* is herbaceous perennial legume belonging to the family Fabaceae, which is primarily cultivated as a forage crop to provide high quality forage, which can be grazed by animals or harvested as hay or silage to be used as an animal feed (Paolo et al 2015). It is a plant that has been grown as feed for livestock for hundreds of years. It is treasured for its maximum content of nutrients *viz.,* proteins, vitamins and minerals. It is highly valued as a legume fodder, where millions of acres are devoted to it (Graham et al 1972). Alfalfa is widely planted over 32 million hectares due to its high nutritive and biomass producing values (Heuzé 2016). Alfalfa has several other benefits apart from forage purposes. Several pharmaceutical industries especially homeopathic pharmacy utilizes it for producing a nutritional tonic. The higher content of protein (60.5 %), minerals, enzymes, vitamins etc. excel the utility of Alfalfa to use as a tonic (Anonymous 1962), which is of increasing demand for its added nutrition, which enhances appetite and digestion resulting in improved mental and physical vigour (Boericke 1927). Alfalfa is an abundant source of Vitamin A and E and rich in Vitamin C (1.78 mg/g), but as the foliage dries vitamin content is lost. Amylase, peroxidase, lipase, and pectinase are notable enzymes present in Alfalfa (Uphof 1968). Alfalfa sprouts are used in salad making, shoots are utilized as leafy vegetable and dehydrated Alfalfa is articulated as a tablet for dietary supplement (Capneura 2008).

Soil fertility conservation by enhancing organic matter content to improve soil fertility has gained attention in soil research these days (Kusvuran et al 2014). Alfalfa is recognized as soil building legume crop, as it is able to significantly accumulate nitrogen by its deep rooting system and biologically fix atmospheric nitrogen (Jarvis 2005). It effectively uses excess water by its high-water consumption capacity with its deep roots. Alfalfa in crop rotation or in a mixed cropping apart from enriching the soil with organic matter, additionally aid in physical and chemical fortification of the soil (Kusurvan et al 2014). The soil aeration and drainage are enhanced due to penetration of alfalfa roots in soil, thereby improving the symbiotic fixation of nitrogen and activity of free-living nitrogen fixers in the soil. Alfalfa, greatly contributes in achievement of sustainable agriculture, as it is cut for foliage, the root system dies, providing organic matter to soil which is degraded by soil micro-organisms and foliage regrows and root develops again (Vasileva et al 2015). Alfalfa removes more N, P, and K on a per acre basis than any other major crop. For example, a six-ton hay yield of Alfalfa removes about 350 lbs of nitrogen (N), 40 lbs of phosphorous

(P) and 340 lbs of potassium (K) (Hanson et al 2013).

Alfalfa pests significantly reduce forage productivity, quality and stand life, minimizing the economic outcomes in alfalfa production. Sufficient awareness of the factors that encourage the build-up of the pest and the skill to diagnose the signs and indications of insects, pathogens and nematodes that infest Alfalfa is a significant tool in successful alfalfa production.

Diseases cause major yield reduction in Alfalfa and reduce the feeding value of the foliage. Several pathogens were reported to infest Alfalfa the severity of which differs seasonally and geographically. Pathogens that cause fungal diseases include virus, fungi, bacteria and nematodes. For a plant to be infested by a disease, the variety must be susceptible to pathogen and the environmental conditions, temperature and humidity favor disease development. The major and minor diseases that infest Alfalfa are detailed as follows:

Major Diseases of Alfalfa

Seedling or damping off (*Pythium ultimum, Pythium irregulare, Pythium violae, Phtopthora megasperma, Rhizoctonia solani*): Damping off is an ailment of seedling, where seeds get infested and die before germination or the emerging seedling will be stunted or collapse or dyeing of seedlings dye shortly after emergence. The damaged seedling before germination is discoloured and soft, and necrotic lesions occur after germination. The lesions after germination girdle the root and stem leading to plant death (Laurine et al 2017). The girdled plants subjected to root tip necrosis may be stunted and yellowish in colour. In older seedlings, near the soil surface a discolored constricted area is seen. The extent of discolouration is dependent on the age of seedling and duration of environmental conditions that favours the progress of the disease. The disease-causing genus, *Pythium* and *Phythopthora* commonly occur in most soil conditions as the fungi is transported by water, infested soil particles and carry over by infected plants. Damping off in Alfalfa leads to *Pythium* sp. that occurs under cool soil temperature. The infestation due to *R. solani* is often influenced by soil organic matter content of the soil enriched by earlier crops, with the infestation increases with the level of organic matter content (Bucciarelli et al 2018). After emergence the seedlings show water-soaked appearance and no proper demarcation between healthy and infested tissues could be noticed. Typical root-tip necrosis as well as inhibition of lateral root formation is caused by infestation of *P. violae. Rhizoctonia solani* causes pre-emergent death of seedlings as well as post emergent necrosis of stem near the soil surface (Lamichhane 2017). *Phythopthora megasperma* is yet another soil borne pathogen, distressing poorly drained soils.

Seedling blight and seed rot (*Pythium, Phythopthora, Rhizoctonia, Aphanomyces, Fusarium* etc): The disease is favoured by several soil pathogens belonging to the genera *Pythium*, *Phythopthora*, *Rhizoctonia*, *Aphanomyces*, *Fusarium* etc. Cool and wet soil conditions will boost the infection. Seeds being destroyed or killed before germination is the major symptom of the disease, mainly caused by *Pythium* sp. The different plant parts display water-soaked lesions initially and collapse and die later (Al-Askar et al 2013). *Aphanomyces* can cause the cotyledons to turn to purple before the seedling dies and will not allow the seedling to die.

***Phythopthora* root rot (*Phythopthora megasperma* f.sp. *medicaginis*):** The soil borne fungus kind of microbe survives well in excess soil moisture with the hard spores in soil or infected plant debris, transported by irrigation water. Wet and cold weather along with heavy, poorly drained soil are favourable for *Phythopthora* development. The disease will be more abundant in water logged areas with excessive moisture and soil compaction increase the root rot problem and cause severe seedling loss (Linda et al 2002). The affected plants wilt and turn yellowish or reddish brown in high temperature. The roots develop numerous dark lesions on the taproot and ultimately the roots will rot. The root cortex shows yellowish discolouration. The affected plants display reddish discolouration which resembles phosphorus deficiency. A thin stand of alfalfa crop will be noticed in severely infested region. Regrowth of foliage will be very slow after harvest.

***Stagonospora* crown and root rot (*Stagnospora meliloti*):** The disease infests crown and root of Alfalfa influenced by cold weather. The spores of the pathogen that occurs as black pyncnidia is transported by irrigation water and spread as the water splashes to injected leaves, stem, plant debris etc. The fungus finds its path of entry into the plant through crown and progress its growth towards taproot slowly The disease will sustain in plant and will take around one to two years to spoil the plant; ultimately the plant will lose its vigour and growth. The crown will be infected anytime but leaves and stem after spring rain. The bark tissues of the infected roots and crowns being rough and cracked are the main symptoms. Red streaks occur in xylem in the middle of the root, down to the rotted portion of the crown. The affected crown tissues will be firm and dry or will lead secondary organisms to invade the tissue (Frate and Davis 2007). The infection causes irregular light shaded lesions with diffuse margins.

***Pythium* root rot (*Pythium ultimum; Pythium irregulare*):** The soil borne fungal pathogens, *Pythium ultimum* and *Pythium irregulare*, endure in soil and debris for several years as oospores). They affect alfalfa seeds causing seed blight or rot and cause water soaking and eventually the roots and stem will be killed as they emerge. The pathogen can cause damage to mature plants by destroying the fine roots without any prominent symptom, but a measurable yield loss. *Pythium* and *Phytopthora,* causing damping off and root rots infest in acidic, poorly drained and soils of high organic matter content. *Phytopthora medicaginis* can affect alfalfa plants in all stages of development and is the common pathogen causing root rot in Alfalfa (Deborah and Laurine 2015). The infection due to *Phythopthora medicaginis* displays stunted growth, reddish purple colour change of leaves and ultimately wilted plants. The infected plants form brown lesions, which turn black with the centre of roots become yellow. The infested damped off tap roots results in pencil tip like appearance. Affected taproots are damped off below the crown region giving a pencil-point look, which can be easily uprooted from the soil in a single pull. The infested seedlings die rapidly making it difficult to distinguish between death due to *Phytopthora* caused by *Pythium*. In general, the whole tap root system of *Phytopthora* infected plants will be rotted and collapse, but only few roots of *Pythium* infested plants rots (Gray 2007).

***Fusarium* seed rot (*Fusarium* sp.):** Several *Fusarium* species cause several types of infections in Alfalfa such as seedling blight, root and crown rot of Alfalfa. *Fusarium* root rot is a chronic condition causing the plant to slowly decline. They spoil the fine roots of plants without causing perceptible decay, but with quantifiable yield reduction. The noticeable symptom is poor and stunted growth of plant above ground level. The roots have brownish or reddish-brown lesions that spoil the roots. *Fusarium* rot infection together with crown rot kills the plant (Ellis et al. 2013).

***Rhizoctonia* root rot or canker (*Rhizoctonia solani*):** *Rhizoctonia* root canker occurs in high temperature conditions causing serious seedling damping off globally. New stands are planted when temperature is ideal for disease development. Only few strains of fungus cause root canker. Occurrence of prominent elliptical lesions in roots at points where lateral roots emerge is a distinctive symptom of *Rhizoctonia* infestation. During winter season, when the fungus is inactive the lesions turn black and stay stable. If the infection is severe, the roots will be girdled leading to death of the plant. New roots will sprout in cold weather if the infestation is not severe.

Complex crown rot: It is a disease caused by the blend of several pathogens such as *Fusarium*, *Pythium, Rhizoctonia*, *Phoma* and *Stagonospora*. Infections in crown occurs, if the crown is injured by causes such as mechanical damage, insect injury, nematode infestation, frost etc., which permits access of pathogen to the crown. The appearance of brown necrotic dead tissues in crown region is a typical symptom, which leads to stunting of plant and yield reduction. Avoiding mechanical injury to the crown area hinders entry of the pathogen to the crown region (William 2016). Planting resistant varieties is yet another tactic.

Bacterial wilt (*Clavibacter michiganense* sub sp. *insidosum*): The bacterium penetrates taproots through wounds developed by insects like Clover root curculio. The disease appears in young plants of three years old. The infested plants are usually scattered and not clustered. The affected plants appear stunted with very slow regrowth after cutting. The plant parts turn to yellowish colour initiating from leaves. The infested roots display yellowish discolouration of the root vascular tissue. The internal root tissues show yellow compared to white in healthy tap roots. Planting resistant cultivars is the management strategy.

Fusarium wilt (*Fusarium oxysporum f. sp. medicaginis*): The disease is highly influenced by high soil temperature and warm climates. The damage can occur in all kinds of soil, but severe infestation is noticed in the soil with nematode activity. The foremost visual symptom of *Fusarium* wilt is shoot wilting, followed by stem blanching that leads to a reddish tint in the foliage (Peterson et al 2017). The disease leads to reddish brown discolouration of plants, especially in the inner parts of roots, that proliferates as the plant ages. *Fusarium* wilt causes dark discolouration, in contrast bacterial wilt shows yellowish brown discolouration (Antonopoulos and Elen 2008). Planting root knot nematode resistant variety will counterpart *Fusarium* wilt resistance, which lessens acquaintance of Alfalfa to pathogen by nematode feeding on the roots.

***Verticillium* Wilt (*Verticillium albo-atrum*):** *Verticillium* Wilt is highly favoured by warm temperature and seriously infest susceptible cultivars, reducing yield up to 50 per cent in the second year of production. The symptoms of wilt include yellowing of leaf tips in a V-shaped pattern, upward rolling of the edges of apical leaflets. As the symptoms progress, leaves turn reddish in shade and become desiccated, defoliated with an intact petiole left over (Göre et al 2011). The infested stem remain green until the leaves are dead. The intact xylem tissues of roots turn brown. The fungi can be carried through alfalfa seed and survives in alfalfa hay and in animal manure. The fungus enters alfalfa roots directly by roots and spread occurs through infection of stem cuts. Care should be taken to prevent importation of infected seed or plant materials. The harvesting equipment's should be cleaned when moving from infested fields into new fields. The infested field should be harvested separately to avoid spreading of pathogen. Care should be taken with irrigation water from infested fields as pathogen spores can be carried in water.

Anthracnose (*Colletotrichum trifolli*): Anthracnose commonly occurs in established alfalfa stands. The development of disease pathogen is maximum in summer and the pathogen persists in debris and crown. Rainfall and irrigation water disperse spores, which is the inoculum source into plant parts such as stem and petioles. Spores also disperse through seeds contaminated in threshing process. Crown rot is the important phase of anthracnose disease which can be diagnosed by bluish black v shaped rot in the crown, which further infests stem and leaves (Harrison and Dixon 1993). The stem acquainted with the crowns turn bleached white and the crowns die and the leaves do not drop from the stem. Irregularly shaped large oval shaped blackened lesions with black borders are also produced due to anthracnose. Acervuli, black fruiting bodies will develop in the lesions, which enlarge, coalesce and wither the stem resulting in a typical 'shepherd crook' symptom on top of the stem.

Minor diseases: Apart from the major diseases, several other diseases are causes by numerous pathogens in alfalfa crop occurs on a regular basis, but most of them are not considered a major cause that limits yield and crop productivity. However, sometimes, they become a serious problem that affect the quality of alfalfa hay. The following are the list of few of those pathogens with minor importance in Alfalfa:

Alfalfa dwarf/Pierces disease (*Xylella fastidiousa*): The sharp shooter, *Homalodisca vitripennis* act as vector of the bacterium, *Xylella fastidiousa*, that cause alfalfa dwarf disease. The green and red headed sharp shooters are responsible for this disease in Alfalfa. The bacterium also causes Pierces' disease of grapes and almond leaf scotch. As the insects acquire the bacterium by feeding on infested plants, they spread the infestation by subsequent feeding of the non- infested alfalfa plants (Blua et al 1999). The disease is of minor importance in Alfalfa. The blue green sharp shooter transmits the disease from Alfalfa to grapes. The primary symptom is stunted growth after first cutting, which may not be apparent for certain period after initial infestation. The infested plants have smaller, darker, distorted, mottled or yellow-coloured leaflets. The tap roots sows yellowish wood with fine dark streaks on a diagonal cut. The yellowing occurs as a ring under the bark with no yellow or brown shades as in bacterial wilt infected by *Clavibacter insodiosum*. Dwarf disease slowly worsens and kill the plant in a year (Costa et al 1999). The green and red headed sharp shooters require grasses like bermuda grass, water grass etc. and hence preventing the growth of grassy weeds help to overcome the disease.

Alfalfa mosaic virus (AMV) and cucumber mosaic virus (CMV): These viruses are not economically important, but feed on Alfalfa and transmit the virus to other economically important food crops and causes severe yield losses. Tomato and potato are such infested crop being transmitted from Alfalfa. Cucumber mosaic virus acquires the virus from the infected plants and spread to beans plant. Alfalfa mosaic virus cause yellow mottling and streaking on leaves, but cucumber mosaic virus shows no specific symptoms (Abdalla et al 2012).

***Aphanomyces* root rot (*Aphanomyces eutriches*):** *Aphanomyces* root rot (APR) is a serious disease-causing severe yield loss in seeded and established alfalfa stands. The disease is caused by soil borne water mold, *Aphanomyces eutriches*. The disease occurs in alfalfa fields with poor drainage and fields with heavier compacted soils receiving excessive water. The disease occurs with variations in crops like soyabean, snapbean, fababean red kidney bean, pea, red clover etc (Wakelin et al. 2002). *A. euteiches* produce microscopic, long-lived resting spores in the roots of alfalfa plants and they remain dormant in soil for up to 10 years. If the spores come across a susceptible crop, oospores can germinate and directly infect plants or under wetter conditions produce several microscopic spores that can subsequently infest plants.

Crown wart (*Physoderma alfalfa*): *Physoderma alfalfa* is a soil borne fungus that will endure in soil forever as resting sporangia, that release zoospores under favourable conditions that penetrate crown buds. The disease is prevalent in wet, waterlogged conditions. The disease cause large knobby swellings on crown and roots and the plant shows stunted growth and often dies maintaining the soil with good drainage, avoiding excessive irrigation and avoid planting Alfalfa in fields with prior history aids to overcome the pests.

Common leaf spot (*Pseudopziza medicaginis*): The disease is caused by the fungus, *Pseudopziza medicaginis* hibernates in plant debris in the soil surface, and get discharged in air. The foliar disease proliferates under cool and wet conditions. The typical symptoms are occurrence of tiny, circular black spots on leaves which turn yellowish and drop. In circumstances of cold weather, apothecia, the fruiting bodies of the pathogen are visible as circular raised bodies in the surface of spots. Though the disease does not destroy the plant but decreases vigour, hay quality and yield (Naseri and Marefat 2008). Severe yield loss is noticed. Early harvesting is recommended as the disease progress over time.

Downy mildew (*Peronospora trifoliorum*): The disease is a cold loving foliar disease, shows up when the temperature is severely cold and humidity is extremely high for the pathogen to produce spores and infest the plant (Liatukas 2014). The abaxial side of the leaves show lighter shade and adaxial side yellowish in colour. The infected areas show

Fig. 1. Damping off

Fig .1. Seedling blight

Fig. 2. Seed rot

Fig. 3. *Phytopthora* root rot

Fig. 4. *Stagonospora* crown and root rot

Fig. 5. *Pythium* root rot

Fig. 6. *Fusarium* seed rot

bluish gray mycelial mats and spores under microscopic view. Spore production is more pronounced in the dawn when high humidity is experienced (Mario and Lira 2018). The entire buds and leaves of the plant will be prone to attack resulting in distortion and yellowing of leaves leading to leaf drop and loss in plant vigour, which ultimately show reduction in yield and quality. Early harvest prevents yield losses.

Lepto leaf spot (*Leptospaerulina briosiana*): It is a fungal disease associated with leaf spots and dead areas surrounded by brown margins giving an 'eyespot'. The disease is said to be seed borne, but transmits by rain and wind. The spots later coalesce and fall out. The fungus builds up on leaves and stem and the spores spread to healthy plant (Feng et al 2016). Early harvest and crop rotation avoiding Alfalfa for more than a year will aid to overcome the disease.

Rust (*Uromyces straiatus*): The disease cause uredial lesions and chlorotic spots and reduce the market value of the foliage. The leafy spurge (*Euphorbia esula*) is an

Fig. 7. *Rhizoctonia* root canker

Fig. 8. Complex crown rot

Fig. 9. Bacterial wilt

Fig. 10. *Fusarium* wilt

Fig. 11. *Verticillium* wilt

Fig. 12. Anthranose

important weed in alfalfa ecosystem and a carrier of rust urediospores. The pycnial and aerial stage occur on spurge and the cycling or uredial stage and overwintering or telial stage occurs in Alfalfa. Therefore, alfalfa rust is a possible candidate for biological control of leafy spurge (Statler et al 1987). Severity of the disease varies from season to season as influenced by temperature and rainfall. Nitrogen deficiency also promote the disease. Destroying the alternate host, leafy spurge from alfalfa weed is the best management option.

Sclerotinia stem and crown rot or white mold (*Sclerotinia trifoliorum*): Two species of *Sclerotinia* causes stem and crown rot, both exposing similar symptoms and need similar environmental conditions for infection. *Sclerotinia sclerotiorum* has a wider range and *Sclerotinia trifolium* infests legumes. The disease is easily identified by white, cottony, mycelial growth on crowns and stems (Batura 2013). The first sign of the disease is wilting of stem. White moist mycelium can be found on stem in soil adjacent to infectious plants. The disease is a cold weather disease proliferate in wet and foggy winters which high humidity is favourable for the disease (Aurelija et al 2012). Weeds such as chick weed favours the disease by prolonging moist conditions in the canopy. Early planting will aid to escape the disease.

Spring black stem (*Phomo medicaginis*): The cool season foliar disease, produces brownish black fruiting bodies called pycnidia on typical stem and leaf lesions. Spores are released from pycnidia on dead stems during wet weather. The symptoms include development of small black to brown spots on underside of leaves, petioles and stems. Irregularly or triangularly shaped lesions coalesce and become light brown (Akamatsu 2008). The infested leaves turn yellow and wither before falling. The tender shoots will be girdled and killed. Most damage occurs before cutting. Management strategies include early harvesting and planting resistant varieties (Castell-Miller and Zeyen 2007). Using pathogen free seeds helps to minimize the infestation as the pathogen is seed borne. Crop rotation can eliminate accumulation of inoculum in the field.

***Stemphylium* leaf *spot* (*Stempylium botryosum*):** It is a cool-season foliar disease favoured by cold temperature and moist weather. The disease is prevalent in first and second cuttings as defoliation occurs under heavy disease pressure. An irregularly shaped lesion with dark border, with the spot increase in size as the damage extends is the typical symptom. It is not considered as an economic important disease as other leafspot infesting Alfalfa (Pablo 2005). No specific recommended management strategy, but early harvest aids to avoid infestation.

Summer black stem and leaf spot (*Cercospora medicaginsis*)*:* The typical symptom of the disease is the plant forms dense canopy. As the plant is infested brown areas of spots with waxy margins arise, the spot appears gray or silvery as spores are produced on the spot and a diffuse yellow colour surrounds the spots. The disease is favoured by high temperature and humidity, early harvest before extensive defoliation minimizes yield loss (Naceur et al 2020).

Management of Alfalfa diseases

Major Diseases

Damping off: Usage of high-quality seeds under conditions of environment that favour rapid germination and seedling growth minimizes the inflectional chance. Excess irrigation to the seedling should be avoided. Planting of seedlings in compacted or poorly-drained soils should be avoided. Fast germinating cultivars planted in well-drained soil under ideal temperature and pH aid to overcome pathogen infestation, while in moist soil conditions, the pathogen destroy the seedlings (Laurine and Deborah 2015). Crop rotation is not recommended for *Pythium* rot because of its broad host range.

Seedling blight and seed rot: The seedling planted in a well-prepared seed bed not being cold or wet minimize the infestation. A natural product, hydroquinone, obtained from the leaves, bark and fruits of ericaceous shrubs such as cowberry, blueberry etc. inhibits the seed born fungi (El-Wakil and El-Metwally 2000) and sodium metabisulfite is an antiseptic and antimicrobial agent (Ash and Ash 2009). Treating alfalfa seedlings with 10mM hydroquinone and 12mM sodium metabisulfide minimizes seedling mortality and increases their survival rate by enriching photosynthetic pigments in plant parts thereby enhancing growth parameters (Al-Askar et al 2013).

***Phythopthora* root rot:** Selection of planting area with internal drainage without a history of *Phythopthora* root rot is the basic strategy in management of the disease. Planting resistant cultivars also help in root rot prevention. Improved soil moisture drainage is a pre-requisite in management. Avoid water logging in the field for more than two days. Rotation of Alfalfa with non-host crops of the disease will favour to avoid infection. Proper fertilization of the crop to promote luxurious growth will aid to resist infestation.

***Stagonospora* crown and root rot**: The disease is more severe during prolonged period of warm, moist weather; therefore, provision of optimum growing condition is an important strategy to avoid *Stagonospora* infection. Rotating with non-host crops for more than a year will eliminate the inoculum from the field. Resistant cultivars are not yet developed, but germplasm with moderate resistance has been released. Planting in firm seed bed with adjusted soil pH and nutrient level with proper soil drainage favours rapid emergence of seedlings and vigorous growth (Deborah and Laurine 2015).

Pythium root rot*:* Management root rot of Alfalfa caused by *Pythium* and *Phythopthora* entails a many-sided tactic. Planting in field with good drainage and no root rot history avoids infection. Planting resistant cultivars for *Pythium* and *Phytopthora* will reduce the pest issue. Seed treatment with high quality seeds treated with fungicides controls seedling blight and early root rots. Rotating crops with non-host crops will aid for dismantling of inoculum stagnation in the soil (Bodah 2017). Regular fertilization of the crop for vigorous crop growth will aid to resist infestation.

***Fusarium* seed rot:** The sole management tactics of this infection is maintaining favourable growing condition. The natural inhibitors of hydroquinone and sodium metabisulphite are promising antifungal agents against these fungi (Al-Askar et al 2013).

***Rhizoctonia* root rot or canker:** Planting in well-drained soils, prepared by tillage, fertility management by supplementing nutrients and weed management aid in managing root canker or rot due to *Fusarium* sp. Planting of seedling in recommended seed rate to avoid overcrowding minimizes the disease pressure. Crop rotation helps to break the lifecycle of pathogen and improve soil fertility. Plant growth promoting bacteria such as *Bacillus pumilus* and *Pseudomonas putida* along with antagonistic fungi such as *Aspergillus awamori, Aspergillus niger* and *Trichoderma harzianum* were beneficial in management of *Fusarium* root rot in Pea plants (Foroud et al 2014).

Complex crown rot: Avoiding mechanical injury to the crown area hinders entry of the pathogen to the crown region (William 2016). Timely irrigation to avoid excess moisture helps to overcome the disease (Bowden 2014).

Bacterial wilt: Planting resistant cultivars is the management strategy. The cultivars Ranger, Agate, Iroquis, Oneida, 120, 532, Lahontan, Orestan, Ladak, Vernal, Caliverdi carry resistance genes against the pathogen. Resistant varieties have carbohydrate storage reserve to give vigor to plant. Maintaining good stand vigor by maintaining potassium level and general fertility helps to overcome the pathogen. Harvesting young stands before old ones and destroying debris from the field also aid in managing the pathogen. The plant stand should not be mowed when wet. Rotating non-host crops for 2-3 years removes the source the infestation entirely from the field (Stuteville and Erwin 1990).

***Verticillium* wilt:** Care should be taken with irrigation water from infested fields as pathogen spores can be carried in water. An irrigation management strategy with the available water maintained at 20 percent led to significant reduction in severity of the disease and extent of xylem colonization by the pathogen (Cabral and Marouelli 2020). The mycoparasite, *Hypocrea rufa*, the antagonist, *Trichoderma hamatum*, and the pathogen *Zygorhynchus moelleri* are efficient natural enemies against the pests (Acharya et al 1995). A meroditerpenoid metabolite isolated from brown algae *C. tamariscifolia* and characterized as methoxybifurcarenone possess antifungal agent against *Verticillium albo-atrum* (Bennamara et al 1999).

Anthracnose: Crop rotation with non-host crops other than Alfalfa and clover for at least two years will eliminate the source of inoculum to the field. In infested fields alfalfa can be harvested before losses become severe.

Minor Diseases

Alfalfa dwarf/Pierces disease: The bacterial pathogen is transmitted by xylem-feeding insects, sharpshooters and spittle bugs, but green and red headed sharp shooters spread the infestation. The green and red headed sharp shooters require grasses like bermuda grass, water grass etc. and hence preventing the growth of grassy weeds help to overcome the disease. Minimizing the attractiveness of sharpshooters by preventing weed growth manages the disease (Civerolo 2001). Hotwater treatment of alfalfa seedlings at 50^{O}C for 45 mins. sanitizes the plant materials against *phytoplasma* the causal agent of Pierces disease, without affecting seedling survival and development (EFSA 2015).

Alfalfa Mosaic Virus (AMV) and Cucumber Mosaic Virus (CMV): Alfalfa mosaic virus can be managed by controlling the vector, aphids by foliar spray of insecticides. Resistant cultivars to alfalfa mosaic virus provide a management tool. Regular roughing of plants manages weeds, which will avoid aphid attraction to alfalfa field (Olawale 2020). Foliar spray of melatonin (MTL) and Salicylic Acid (SA) (100 µM) act as eco-friendly antiviral compounds that causes significant increase in morphological characters such as shoot and root length, number of leaves, leaf area and biomass, chlorophyll and carotenoid compounds and antioxidant compounds, whereas reduces oxidative damages caused by the virus, by reduction in hydrogen peroxide, superoxide anions, hydroxyl radicals and malondialdehyde (Ahmed et al 2021).

***Aphanomyces root rot* (*Aphanomyces eutriches*):** The most effective means of managing the root rot is use of resistant cultivars. Maintaining proper drainage in the field is the significant management strategy. Avoiding stagnant water in the field prevent development of oospores. Minimizing soil compaction can help to overcome wet soil conditions. Increased concentration of calcium in soil is found to lower the severity of the incidence of the pathogen. Crop rotation is not an effective management strategy as oospores of *A. euteiches* will survive in the soil for longer duration. Cover crops such as crucifers and green manure crops such as oats were found to lower the incidence of *A. eutriches*. Seed treatments aid to overcome the disease only until seedling emergence. Several fungicides, soil fumigants and biological control options is effective against the pathogen. The active ingredients of the dinitroaniline herbicides, Pendimethalin (Prowel®) and trifluralin (Treflan®) were detrimental to *A. eutriches* (Hughes and Craig 2007). The beneficial bacterium, *Bacillus velezensis* UCMB5113 and beneficial earthworm, *Lumbricus terrestris* positively enhance the health and growth of Alfalfa with taller plants and higher biomass and thereby enhance plant growth and biological control of root rot. Earthworm considerably reduce disease infestation as they consume the pathogen (Jan et al 2020)

Crown wart*:* Maintaining the soil with good drainage, avoiding excessive irrigation and avoid planting Alfalfa in fields with prior history aids to overcome the pests. Resistant cultivars are suggested to minimize infestation (Samac 2014). Soil should be levelled and tilled deeply to reduce compaction. Seedlings should be planted on beds to alleviate disease severity. Reduce the length and time of flood irrigation to avoid infestation (UK CAB International 1987)

Common leaf spot: Severe yield loss is noticed. Early harvesting is recommended as the disease progress over time. Though the disease wont severely infests the plant, it reduces vigour, hay quality and yield. Resistant cultivars are an option. Crop rotation reduce inoculum in the field. The fungicides, Raclostronin@6–9 fl oz/acre and Azoxystrobin@6–15.5 fl oz/ acre offers a solution (Qin et al 2016).

Downy mildew*:* Early harvest prevents yield losses as the inoculum as well as susceptible host tissues of downy mildew were removed, as they are short lived and will be dead allowing new susceptible tissues to be grown. Early cutting minimizes humidity among crop canopy reducing risk of infestation (Skinner and Stuteville 2015). Seed treatment with a systemic fungicide, metalaxyl and mefenoxam protects seedlings from downy mildew infestation. Foliar spray of fungicides Ridomil (metalaxyl) and Curathane (cymoxanil) significantly reduce the infestation. The Ridomil applied alfalfa fields showed 29.94 per cent greater yield than control and Curathene applied fields show 28.37 percent greater yield (Mario and Lira 2018).

Lepto leaf spot: Early harvest can prevent leaf loss and accumulation of spores. Crop rotation avoiding Alfalfa for more than a year will aid to overcome the disease. Choosing well-adapted high yielding alfalfa varieties and using certified high yielding seeds aid in healthy crop stand. Plant in warm, well-drained and well-prepared seed bed with balanced nutrients with adequate amount of phosphate and potash. Cut heavily infested strands in the mid to late bud stage for high yield, minimal leaf loss etc. Cut as the foliage is dry to avoid the spread of pathogen. Minimize weed and insect infestation (Jim 2011).

Rust (*Uromyces straiatus*): Destroying the alternate host, leafy spurge from alfalfa weed is the best management option. Cultivars resistant to alfalfa diseases are recommended. Early cutting of alfalfa foliage is suggested when the disease is severe. Foliar fungicidal spray of copper compounds such as Champ Formula, Kocide 2000, Nordox 75 WG, Nordox 75 WG are recommended (Statler et al 2017).

***Sclerotinia* stem and crown rot or white mold:** Early planting will aid to escape the disease. Management of the disease is based on adjusting planting date by spring which allows the plant to develop resistance to the disease. Deep ploughing bury the sclerotia in the soil and reduce disease incidence. Planting the seedlings in fields with no history of the disease and rotating with non-host crops for 2 to 3 years will aid in minimizing the infestation (Malvick 2001)

Spring black stem: Using pathogen free seeds helps to minimize the infestation as the pathogen is seed borne. Crop rotation can eliminate accumulation of inoculum in the field. Planting moderately resistant varieties can significantly reduce losses (Stephen 2011).

Stemphylium leaf spot: Early harvest aids to avoid infestation. Any systemic fungicide at 0.6 kg rate applied at 50% bloom; and repeated at 7-10 days later for maximum of three applications reduce the infestation (Pearse 2006).

Summer black stem and leaf spot: Adjusting the cutting schedule is the most practical and economical means of management. Any copper hydroxide fungicide can be applied 10-14 days or earlier to harvesting can reduce the loss (Stephen 2011).

CONCLUSION

Alfalfa is an important crop suitable for integrated pest management. Alfalfa can endure pest injuries to certain degree and not lose significant quantity or yield. The ecosystem of Alfalfa is tremendously complex with manifold interactions of crop, pest and the natural enemies occurring at various levels. Alfalfa is chiefly managed in farms with the growers taking several pest control decisions. A bulk quantity of alfalfa cultivated all around the globe is consumed in dairies where forage quality, timing of cutting etc gains more importance. Integrated pest management requires awareness to take decisions based on extensive knowledge base of the insect pests, diseases, nematodes, weeds and wildlife in alfalfa ecosystem. IPM decision makers should be proficient not only in farm sciences, but should have an exhaustive knowledge and understanding of the ecological moralities that administer the fluctuations in pest and natural enemy species that impose considerable dynamics in the economics of alfalfa production and farm utilization as fodder.

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Structure and Distribution of Fish Community in Middle of Hamrin Dam Lake

Raaed Sami Attee, Luay Mohmmed Abbas[1*] and Maen Faleh Mahdi[2]

Department of Animal Production, Agriculture of College, University of Diyala, Iraq
[1]Chief Scientific Researcher, Livestock and Fisheries Center, Agricultural Research Department
The Ministry of Science and Technology, Iraq
[2]Director of Agriculture in Diyala governorate, Ministry of Agriculture, Iraq
**E-mail: mnfalh726@gmail.com*

Abstract: The nature of structure and distribution of the fish community in the middle of Hamrin Dam lake was studied for the period from July 2020 to April 2021. The lowest temperature was 12.6 m in February and the highest temperature was 34.3^{o}C in August 2020. Furthermore, the lake water was characterized as freshwater, and the lowest salinity was 0.16 g/l during February and the highest 0.38 g/l in September. The levels of dissolved oxygen were high, with highest concentration of 12.5 mg/L in December 2020, while the lowest was 7.4 mg/L in July 2020. A total of 5762 with a total weight of 979.4 kg were collected, where these fishes were represented by 23 species of fish belong to six families. The dominant commercial fishes 13 species were recorded with a total weight of 703.0 kg formed 71.8% of the total fish catches. However, the rest was non-commercial species (10 species) with total weights of 276.4 kg, which represented 28.2% of total fish catches. The fish *Planiliza abu* ranked the first in terms of numbers with 11.7%, followed by the common carp *Cyprinus carpio* by 10.8%, then *Mesopotamichthys sharpeyi* by 8.4%, and the *Luciobarbus xanthopterus* by 8.3%. In terms of weights, the S*ilurus triostegus* was ranked the first with 23.7%, followed by *Luciobarbus xanthopterus* by 18.6%, and *Cyprinus carpio* fish by 10.9% ranked third. The *Cyprinion macrostomus* recorded and the lowest numbers by 0.1% and the lowest weights 0.01% of total fish catches. The study indicate that the waters of the Hamrin Dam lake are freshwater, good airing, and there are abundant numbers and weights of commercial fish.

Keywords: Fish community, Hamrin Dam lake

The fish has high percentage of animal protein estimated at 15-20% (Fagbenro et al 2005) and is nutritionally balanced because it contains a percentage of fats and essential amino acids such as methionine, lysine, vitamins, and an important source of minerals such as calcium, iron, iodine, zinc, and phosphorous and unsaturated fatty acids (Farhan et al 2015). Fish are one of the largest groups within the animal kingdom, as this group includes about 1000 economic species that are used in food production (Yesser et al 2013). Water temperature affects the physiology and behavior of aquatic organisms and is considered one of the important environmental factors determining its abundance and diversity, especially in fish, as well as affects the physical, chemical, and life characteristics. Temperature controls the metabolic rate and growth rate, which is the main factor behind all aspects of life and its conditions (Silvano et al 2000). Hamrin Lake is one of the largest and most important freshwater lakes in Iraq, Determine the types of fish in the lake and the amount of monthly and total catch for each type of fish, determine the prevalent, seasonal, and rare species, come up with a database on the preparation, lengths and weights of different fish and determine the means of fishing used.

MATERIAL AND METHODS

Hamrin Dam Lake is in the Diyala Governorate in eastern Iraq, 120 km northeast of Baghdad. The lake can accommodate two billion and 400 million cubic meters. The lake belongs to the Hamrin Dam, which is located on the Alwand River in Diyala Governorate. The main sources of water revenue for the reservoir are the Diyala River, the Alwand River, and the seasonal natural valleys.

Sampies collection: Water and fish samples were collected from Hamrin Dam lake once a month for a period starting from July 2020 to April 2021 during the morning hours at the middle of each month, where specific physical and chemical properties of the water were measured (30 cm depth from the surface layer of the lake) (Table 1). The air and water temperature were measured using a simple mercury thermometer. Both pH and the electrical conductivity of water was measured using pH type 9811 HI, and Ec -TDS Meter. Conversely, the concentration of water salinity was measured using the following equation:

Salinity (g/L) = Electrical conductivity (µS/cm x 0.00064)

The Secchi disk was used to measure transparency, and the dissolved oxygen concentration was measured using an oxygen meter. Various types and sizes of nets were used for fishing, including the gill net, with dimensions of 4 x 100 m , the mesh size ranged between 2.5 to 10 cm) and cast net with a diameter of 2 m and the mesh size 1.5 cm and 2.5 cm. Fishing operations were carried out by 3 throws for 6 hours (at the rate of throwing nets 2 times for every two hours) for 8 km. The Seine net was used by two Seine for an hour and 15 minutes for each Seine, as they were 250 meters in length and 6 meters in height at the middle, and 4 meters at the ends. A fishing boat with a length of 4.5 m, a width of 1.80 m, and a motor of 40 horsepower was used, considering the stability of the same fishing effort (numerically or by weight) in the fishing area during the study period, which was expressed as kg/hour. The total length (TL) were measured Numerical abundance and weight were calculated using percentages of number or weight per month and season.

RESULTS AND DISCUSSION

The highest water temperature was in August, 34.3°C and the lowest water temperature in February (12.6°C) for the year 2021. The current study results agreed with most of the previous studies conducted on the Tigris River that the highest water temperature in the river is during the hot months and the lowest degree is during the colder months (Al-Sultany 2014). Mahmoud et al (2018) mentioned that the climate has a significant impact on the water temperature in addition to the current speed, as the temperature increases in stagnant places due to the long period of exposure to sunlight, unlike running water. Temperature is an environmental factor that interacted with the growth, reproduction, nutrition, presence, and abundance of species (Taher et al 2011). Similarly, water temperature ranges ranging from 11-30°C were recorded in Lake Habbaniyah (Al-Rudaini et al (1999). Seasonal changes in water temperature are considered suitable for fish living and were within the appropriate ranges that fish can tolerate. Likewise, it is consistent with many previous local studies of inland water bodies (Al-Tamimi 2002, Salman 2013). The pH was lowest in July 7.4, while the highest 8.9 in January 2021. The current results indicate that the monthly changes in the pH values of the Lake Hamrin waters were close to the results of many previous local studies that indicated the light or semi-neutral basicity of inland waters (Al-Rudaini 2010, Salman 2013). Most studies indicated that the Tigris River waters are neutral, tending slightly to alkaline, where the slight fluctuation in the pH degree is related to the concentration of gases dissolved in the water such as carbon dioxide, hydrogen sulfide, and ammonia, which usually ranges in most natural waters between 4 to 9 (Al-Tamimi 2000). The pH in the current study slight changes t in the different months, were within the appropriate and harmless limits for fish, which ranged from 6.5 to 8.5. Salinity concentration ranged between 0.16 g / l in February 2021 to 0.38 g / l in September 2020, The fluctuation of water salinity concentration in Hamrin Dam lake in indicate the season of cold and warm months and its rise in the season of hot months is due to the variation in water temperatures in this season. In addition, rising water levels are caused by the rains in the winter season and the beginning of the spring season, causing water salinity to be diluted. Either, the high temperature in the summer season and an increase in the rate of water evaporation as well as an increase in agricultural and industrial activities whose waste flows into the rivers feeding the lake itself, leads to increase in its concentration. Salman (2013) recorded the lowest value of salinity in February 0.15 and the highest in August 0.33 in Lake Dukan Dam and these results are consistent with the

Table 1. Physical and chemical properties of water in Hamrin Dam lake

Properties/Month	Water temperature (°C)	pH	Salinity concentration (g/L)	Transparency (cm)	Dissolved oxygen concentration (mg/L)
July 2020	33.5	7.4	0.36	49.2	7.4
August	34.3	8.2	0.35	52.0	7.7
September	32.2	8.2	0.38	54.3	7.5
October	28.0	8.2	0.26	42.0	8.4
November	27.0	8.1	0.24	38.3	9.5
December	25.5	7.8	0.21	36.2	12.5
January 2021	14.4	8.9	0.19	32.8	12.0
February	12.6	7.8	0.16	42.3	11.2
March	26.0	7.9	0.17	37.4	9.4
April	29.0	7.9	0.20	34.2	8.1
Range	12.6-34.3	7.4-8.9	0.16-0.38	32.8-54.3	7.4-12.5
Average	26.3±2.3	8.1±0.1	0.26±0.25	2.4±41.9	9.4±0.6

current study. On the other hand, the monthly and locational changes of water transparency values for Hamrin Dam Lake throughout the study period showed that the highest value of water transparency was 54.3 cm in September 2020 and the lowest of 32.8 cm in January 2021. The low values of water transparency are due to the lake effect, consequently, the suspended clay material increases, and the lake level varies from the rise and decrease in the amount of water in it during the seasons of the year, in addition to the temperature. Water transparency is related to the density of suspended and dissolved substances, the movement of the water column, light intensity, and the prevailing weather condition (Hadjmttsis et al 2006). The highest value of dissolved oxygen was recorded in December, which was 12.5 mg/l, while was 7.4 mg/l in July. The 5762 fish were caught with a total weight of 979.4 kg, and 23 species of fish were recorded belonging to 6 families. Cyprinidae family formed the largest proportion with 16 species by 76.92% of the total catch. The Cichlidae and Bagridae family recorded 2 species, while the families of Mugilidae, Siluridae, and Mestacembelidae represented one species (Table 2). The current study agreed with that different local environments dominated by the Cyprinidae family (Wahab 2013).

The *Planiliza abu* fish dominated numerically with 11.7% of the total fish catches, followed by *Cyprinus carpio* by 10.8%, and *Barbu sharpeyiy* 8.4%, and the *Luciobarbus xanthopterus* (8.3%) (Table 3). The *Silurus triostegus*, dominated by weight with 23.7%, followed by the *Luciobarbus xanthopterus* fish by 18.6%, then *Cyprinus carpio* by 10.9%, and Aspius vorax 10.5% of the total fish catches.

Some local studies dealt with the types of fish caught according to their numbers and weights for different water bodies, in which the *Planiliza abu* and *Carassius auratus* exceeded in terms of number by 39.6 and 24.7%, respectively. The weight dominance of common carp and goldfish was 43.2%and 23.5%, respectively (Al-Radini et al 1999). Fish are caught in different quantities and seasons of the year and from one site to another, and this is due to several factors, the most important of which are the different fishing methods used locally, the time and place of fishing, and the different environments. The monthly changes of the types, numbers, and weights of fish caught in Hamrin Dam lake throughout the study period given in Table 4. Thus, the types of fish fluctuated in the fishing samples throughout the study period, where 15 species of fish were recorded as the lowest number in September of 2020, while the number of fish increased to 20 species in April of 2021. The lowest number of fish caught was 490 fish by 8.5%, in February 2021, and the highest number of fish caught was 644, representing 11.2% of the total catch. The monthly weight of the fish caught recorded the lowest total weight of 87.6 kg in August 2020, which represented 8.9% of the total fish catch. The fish weight returned to 114.9 kg in April of 2021 and was 11.7% of the total catch. The current results indicate that there are clear changes and fluctuations in the numbers and weights of fish caught throughout the study period. Al-Rudaini et al (2001) in the Haditha Dam Lake and Al-Rudaini (2009) in the Radwaniya Lake, recorded superiority in the number of fish caught for the hot months, especially July and August, respectively, in terms of the number of species, numbers of fish and their weight. The results explained the presence of all types of resident fish that were dominant in the monthly fishing samples for the season of high-water temperature and increased feeding activity. The number of fish caught is usually high in the warm months in Iraqi rivers.

The total quantity of fish caught was distributed among commercial species 13 species, and it was characterized by

Table 2. Types of fish caught in Hamrin Dam lake

Local name of the species	Scientific name	Family
Common carp*	*Cyprinus carpio*	Cyprinidae
Bunni*	*Mesopotamichys sharpeyi*	
Yellowfin barbell*	*Luciobarbus xanthopterus*	
Shabout*	*Arabibarbus grypus*	
Nabbash	*Luciobarbus barbulus*	
Goldfish*	*Carassius auratus*	
Himri*	*Carasiobarbus luteus*	
Leuciscus vorax*	*Leuciscus vorax*	
Tigris bleak	*Alburnus caetuleus*	
Sellal	*Chalcalburuns sellal*	
Grass carp*	*Ctenopharyngodon idella*	
Common bleak	*Alburnus mossulensis*	
Berzem*	*Luciobarburs kersin*	
kangal fish	*Cyprinion macrostomus*	
Bizz*	*Luciobarburs esocinus*	
Menon	*Capoeta damascina*	
The blue tilapia*	*Oreochromis aureus*	Cichlidae
The Nile tilapia*	*Oreochromis niloticus*	
Tigris Mystus	*Mystus pelusius*	Bagridae
Stinging catfish	*Heteropneustes fossilis*	
Abu mullet*	*Planiliza abu*	Mugilidae
Tire-track eel	*Mastacembelus mastacembelus*	Mastacemblidae
Cat fish	*Silurus triostegus*	Siluridae

*Commercial

dominance in terms of the number of fish (Table 5). Among non-commercial fish 4560 fish recorded with a total weight of 703.0 kg, and among non-commercial fish, 10 species were recorded total 1202 fish with a total weight of 276.4 kg. The lowest number of commercial fish out of the total catches was 398 fish in February 2021, and the lowest weight was 61.2 kg in July 2020. However, the same fish species formed the highest number of 498 fish in December, and the highest total

Table 3. Types of fish caught according to their numbers and weight in Hamrin Dam Lake

Species	Total number	Percent	Total weight (kg)	Percent	Total length ranges (cm)	Total weight range (gm)
*Planiliza abu**	672	11.7	35.9	3.7	8.7-26.2	16.2-160.1
*Cyprinus carpio**	623	10.8	106.1	10.9	9.3 -43.2	12.1665.0
*Mesopotamichys sharpeyi**	483	8.4	36.9	3.8	7.8 -37.6	13.1-410.0
*Luciobarbus xanthopterus**	476	8.3	182.6	18.6	9.3 -56.3	14.2-2350
*Oreochromis aureus**	472	8.2	18.1	1.8	7.4-23.5	8.1-167
*Arabibarbus grypus**	463	8.0	54.2	5.5	11.2-46.7	22-486
*Carassius auratus**	462	8	51.1	5.2	9.1-37.1	12.3-395.6
*Carasiobarbus luteus**	361	6.2	89.9	9.2	6.1-38.6	5.5-1401
*Leuciscus vorax**	359	6.2	102.9	10.5	9.4-58.6	22.1-1664
Heteropneustes fossilis	248	4.3	11.2	1.1	7.2-30.1	11.2-138
Silurus triostegus	239	4.2	231.9	23.7	15.3-71.2	155-3250
Mystus pelusius	237	4.1	7.7	0.8	7.3-23.2	11-81.5
Luciobarbus barbulus	184	3.2	6.1	0.6	7.4-29.3	11.2-186
*Oreochromis niloticus**	158	2.7	10.6	1.1	7.3-23.3	11.2-157
Alburnus caetuleus	133	2.3	6.1	0.6	7.2-26.1	11.3-147
Chalcalburuns sellal	129	2.2	4.3	0.4	6.6-22.2	8-62
Capoeta damascina	23	0.4	5.7	0.6	10.5-30.4	97.6-328
*Ctenopharyngodon idella**	20	0.3	13.4	1.4	25.7-56.3	142-9101
*Luciobarburs kersin**	9	0.1	1.2	0.1	14.3-35.1	53-346
Mastacembelus mastacembelus	6	0.1	3.1	0.3	38.3-61.2	163-606
Alburnus mossulensis	2	0.1	0.16	0.02	18.3-19.2	83.1-86.4
*Luciobarburs esocinus**	2	0.1	0.14	0.01	19-23.1	53.1-87.6
Cyprinion macrostomus	1	0.1	0.12	0.01	18.8	122
	5762		979.42			

*Commercial

Table 4. Monthly changes of fish species, total numbers, and total weights caught in Hamrin Dam lake

Month	Number of fish species	Total number of fish	%	Total weight of the fish	%
July 2020	17	553	9.6	88.8	9.1
August	18	592	10.2	87.6	8.9
September	15	592	10.3	100.5	10.3
October	16	578	10.0	96.8	9.9
November	17	591	10.3	97.4	9.9
December	18	644	11.2	89.6	9.1
January 2021	16	550	9.5	105.7	10.8
February	16	490	8.5	103.1	10.5
March	18	540	9.4	95.1	9.7
April	20	632	11.0	114.9	11.7
Total	23	5762		979.42	

Table 5. Number and weight of commercial and non-commercial fish caught in Hamrin Dam lake

Month	Commercial fish		Non-commercial fish	
	Number	Weight (kg)	Number	Weight (kg)
July 2020	428	61.2	125	27.6
August	448	63.0	144	24.6
September	463	64.8	129	35.7
October	466	65.5	112	31.3
November	493	76.3	98	21.1
December	498	71.1	146	18.3
January 2021	458	82.1	92	23.6
February	398	71.8	92	31.3
March	418	62.6	122	32.5
April	490	84.5	142	30.4
Total	4560	703.0	1202	276.4

Table 6. Monthly changes, commercial and non-commercial catch rates per unit for fish caught in Hamrin Dam lake

Month	Total catch (kg/h)	Commercial catch (kg/h)	Non-commercial catch (kg/h)
July 2020	14.8	10.2	4.6
August	14.6	10.5	4.1
September	16.8	10.8	6.0
October	16.1	10.9	5.2
November	16.2	12.7	3.5
December	14.9	11.9	3.0
January 2021	17.6	13.7	3.9
February	17.2	12.0	5.2
March	15.9	10.4	5.5
April	19.2	14.1	5.1
Total	163.3	117.2	46.1

weight was 84.5 kg in of April of 2021. In non-commercial fish recorded the 92 lowest number of fish with weight of18.3 kg in December. The highest numbers and weights, amounting to 146 fish in December, with a total weight of 35.7 kg in September. The species, numbers, and weights of fish in water bodies depend on the environment, productivity, and food of fish.

The monthly changes in the values and rates of total, commercial and non-commercial catch in the unit effort of fish caught in Hamrin Dam lake are given in Table 6. The lowest total catch was 14.6 kg/hour (August 2020), 10.2 kg/hour in July 2020 and 3.0 kg/hour of non-commercial fishing December 2020. Though, the amount of fishing increased in the subsequent months and recorded the highest amount of total fishing 19.2 kg / hour and for commercial fishing was 14.1 kg/hour in April and for non-commercial fishing was 6.0 kg/hour in September 2020. Catch per unit Effort (CPUE) expresses the number (individuals) or weight of fish (kg) caught during the unit of time/hour. It includes the number of fishermen, the number of boats, the number of fishing days, the types of nets and the number of their throws, as well as the lunar phase, which is very important during night fishing operations (Siddig et al 2013). Current results showed that the total and commercial fish catches took a fluctuating pattern during the study period, as the number of fish caught increased in the cold months, and the amount of catch reached its peak with the decrease in water temperature with the beginning of the spring months and the winter months Al-Rudaini et al (2001) confirmed the high number of fish caught during the warm months in some inland water bodies, as fluctuation in the catch per unit effort was observed throughout the different months.

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