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PGR NOTE

Evaluation and Validation of Bold Seeded Accession in Ricebean [*Vigna umbellata* (Thunb.) Ohwi and Ohashi]

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A total of 2,202 accessions of ricebean have been conserved in the National Genebank of India at Indian Council of Agricultural Research-National Bureau of Plant Genetic Resources (ICAR-NBPGR), New Delhi. During routine seed monitoring for seed viability, one unique accession IC009634 was observed with very bold seeds, 100-seed weight of 37.44g. The bold seeded expression was validated under field conditions at two locations viz; ICAR-NBPGR, Regional Station, Shimla and ICAR-NBPGR, Regional Station, Shillong. This accession has the potential to be utilized in the *Vigna* improvement programme.

Key Words: Bold seeded, Germplasm, Ricebean, Sikkim, *Vigna*

Ricebean [*Vigna umbellata* (Thunb.) Ohwi and Ohashi], is a multipurpose legume of Fabaceae family mainly grown in *Kharif* season. It originated in Myanmar–Thailand and India (Tian *et al*., 2013). It is widely grown in Nepal, Bhutan, and North-East India, Myanmar, Southern China, Northern Thailand, Laos, Vietnam, Indonesia and East Timor (Tomooka, 2009). In parts of South-East Asia, ricebean exists as a complex with its wild form from which it is not taxonomically distinguished and with which it can form fertile hybrids (Seehalak *et al.,* 2006). In the North-eastern hills of India, ricebean is used as a pulse, vegetable, and fodder crop. The crop is mainly cultivated under shifting cropping systems and is particularly important for ethnic groups in these areas. Ricebean is well suited to outcrossing, based on its prominent yellow flowers raised above the leaf canopy. In the tropics, ricebean is perennial based on its very thick stem and roots (Tomooka *et al.,* 1997).

This minor pulse has been little studied, but its prolific growth and abundant pods suggest a high yield potential as a vegetable (green pods), grain, and forage crop (Smartt, 1991). Most of the research work on ricebean has focused on its high level of resistance to the major storage pest bruchid beetles (*Callosobruchus* spp.) (Kashiwaba *et al.,* 2003; Somta *et al.,* 2006). Arora *et al.* (1980) collected and evaluated 300 rice bean accessions from eastern and North-eastern India and reported that this crop is free from diseases such as yellow mosaic virus, *Cercospora*, and bacterial leaf spot (*Xanthomonas* spp.). Other species of this genus suffer greatly. Pandiyan *et al.* (2008) also reported the highest level of mungbean yellow mosaic virus resistance in ricebean among different species of *Vigna.*

A total of 2,202 accessions of ricebean collected across the geographical areas of India, has been conserved in the National Genebank of India at Indian Council of Agricultural Research-National Bureau of Plant Genetic Resources (ICAR-NBPGR), New Delhi. During routine seed monitoring for seed viability in 2020, one unique accession IC009634 (collected from Sikkim) was observed for the first time with very bold seeds, weighing 37.44 g for 100-seed weight.

To date, we have not found any report in literature with such a stable genotype with high average 100 seed weight in any cultivated ricebean genotype. Hence, to validate the bold seededness IC009634 was grown at two locations viz; ICAR-NBPGR, Regional Station, Shimla (31º5'N and 77º5'E with 1,924 amsl) and ICAR-NBPGR, Regional Station Shillong (25º41'N long 91º55'E with 100 amsl during *Kharif* 2020. As ricebean is temperate crop, both these environments were well-suited for getting the desirable expression. At

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Fig. 1. a) Seed weight of IC009634; and popular reference rice bean varieties PRR1 (b), PRR2 (c) and VRB3 (d)

both locations, this bold seeded accession (IC009634) along with three checks namely, PRR-1, PRR-2 and VRB-3 were characterized for ten quantitative traits such as, days to germination, days to 50% flowering, leaflet length (cm), leaflet width (cm), no. of branches/ plant, plant height (cm), pod length (cm), seed length (cm), seed width (cm) and 100 seed weight (g); seven qualitative traits viz., flower colour, leaflet shape, pod colour, pod curvature, seed shapes, seeds colour and seeds lustre using a minimal descriptor.

Accession IC009634 grows vigorously with dark green broad leaves. The leaflet length (14.36 cm) and width (10.73 cm) of IC009634 were higher than checks. The average mean days to 50% flowering was 65 days. Accessions from Taiwan were reported with 54-78 days to 50% flowering (Tian *et al.,* 2013). Plant height ranged between 163-210 cm with an average of 191 cm, which is also very high compared to the checks, where it was 45.33 cm, 61.66 cm and 76.00 cm in PRR-1, PRR-2 and VRB-3, respectively. The number of branches per plant was recorded from 5 to 6. The average pod length was 12.83 cm. Seed length and seed width was almost double as compared to check varieties. Seed length was 11.31 cm in IC009634, and in PRR-1, PRR-2 and VRB-3 it was 6.17 cm, 6.49 cm and 6.82 cm, respectively. Similarly, the average seed width of IC009634 was 6.90 cm and 3.83 cm, 3.31 cm and 3.66 cm in PRR-1, PRR-2 and VRB-3, respectively. Average 100-seeds weight was 38.05 g which is significantly higher as compared with checks, PRR-1 (6.18 g), PRR-2 (5.89 g) and VRB-3 (5.94 g). In earlier studies, the highest 100-seed weight 29.86 g reported in the landrace Rhidi Kemagh (IC423239) collected from Zoehnoboto District of Nagaland (Pattanayak *et al.,* 2018). Studies with the Indian (Sharma *et al*., 1995; Singh *et al*., 1998; Mishra *et al*., 2008; Gupta *et al*., 2009) and international (Tian *et al.,* 2013) collections of ricebean reported high variations in 100 seed weight, seed yield, number of branches per plant, pod length and days to maturity. In Japan and Korea, small (<9 g), non-branching and early flowering accessions were prominent. In China, accessions with slightly larger seed (up to 10 g) and branching growth habit were found from Yunnan province, southern China. Bold seeded accessions (>21 g) were found in Nepal (Tian *et al.,* 2013). In ricebean, 100-seed weight exhibited high heritability and high genetic advance (Gupta *et al.,* 2009). In IC009634, the flower colour was yellow and the leaflet shape was ovate. Pods were semi-curved with brown at maturity. Seeds were cylindrical in shape, light brown and shiny.

The present study reports a novel and stable genotype with high 100-seed weight. Therefore, the identified accession has tremendous potential as a promising donor for ricebean breeding programs.

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RESEARCH ARTICLE

Decline in Occurrence and Distribution of *Sesamum prostratum*, a Crop Wild Relative of Sesame, along the Eastern Coastal Region of India

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An exploration was undertaken for collection of the narrow-endemic wild relative of sesame, *Sesamum prostratum* Retz., a potential Critically Endangered (CR) species, specific to the coastal habitats, in the strands between 150-300 m from the sea shore of the east coast. Herbarium specimens, the authentic source for occurrence and distribution, for *S. prostratum* at various herbaria (collected during 1802-1988) are available. A trip was undertaken in Dec. 2020 along the east coast region of Tamil Nadu, between the latitudes 10.29 N to 13.56 N, that covered all previously recorded collection sites. Unfortunately, in 71% of the sites surveyed, *S. prostratum* was absent. All the sites with *S. prostratum* were found to be undisturbed or with minimal human interference. Our preliminary assessment as per the IUCN guidelines suggests for CR category. In sites around the Chennai sea shore, we found *Canavalia rosea*, with *Ipomoea pes-caprae*, while the endemic *S. prostratum* was absent. The loss could be due to two reasons, habitat-based-probable spread of introduced species such as *Ipomoea pes-caprae*, protection afforded to *Spinifex littoreus* (besides severe urbanization, cyclonic storms, and tsunami,where the habitat itself is fragmented and prone to disasters) and the non-competitive nature of the species. Strategies for threat assessment (especially post-disasters) with an action plan, is discussed that could prevent extinction risk, especially of endemic.

Key Words: Climate change, Conservation strategies, Critically endangered, Narrow-endemic, Tsunami.

Introduction

The genus *Sesamum* L. belongs to the family Pedaliaceae, and it includes an important oilseed crop, *S. indicum*, native to India (Bedigian, 2015). The genus *Sesamum*, comprising seven sections viz., *Aptera*, *Ceratotheca*, *Chamaesesamum*, *Dicerocaryum*, *Josephinia*, *Sesamopteris*, and *Sesamum*, occurs mainly in Africa, Australia, India, Malaysia, the Philippines and Indonesia, with 31 species (Ihlenfeldt, 1988; Namiki, 1995; Pradheep *et al*., 2021).

Two of the seven sections of *Sesamum*, namely *Sesamum* and *Chamaesesamum*, are native to the Indian subcontinent (Bedigian, 2015; Pradheep *et al*., 2021). The wild species of the cultivated sesame (sections, *Aptera*, *Chamaesesamum*, *Sesamopteris*, *Sesamum*) is represented in the National Genebank (NGB) of ICAR-NBPGR by a few hundred accessions. However, from the section *Chamaesesamum*, only *S. laciniatum* is collected and conserved (with *S. laciniatum* synonymized to *S. prostratum*, please refer Bedigian, 2015).The habitat for these two taxa are distinct; *S. prostratum*, is a species of the coastal areas of the east coast (Rao, 1971; Rao and Sastry, 1974), and is distributed along the coastal regions of Tamil Nadu, Puducherry and Andhra Pradesh; whereas *S. laciniatum* is documented to occur in hard laterite soil and distributed in the Malabar region and Deccan hills of Peninsular India (Pradheep *et al*., 2021).Our recent studies had shown that the morphological characters of lamina, calyx, stem, and seed are very distinct and worth considering as distinct taxa (Pradheep *et al*., 2021). Furthermore, *S. prostratum* is endemic to certain pockets of India and Sri Lanka (Kumar *et al*., 2019). In Sri Lanka, it is categorized as a critically endangered (CR) species (Wadugodapitiya *et al*., 2013; Weerakoon *et al*., 2020). We undertook an exploration targeted to collect *S. prostratum* material for its conservation, and

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to map its current distribution pattern, from the eastern coastal areas of Tamil Nadu.

Materials and Methods

A preliminary survey of herbarium collections of *Sesamum* deposited at the Botanical Survey of India (BSI), Coimbatore (MH, the Madras Herbarium) and BSI, Howrah (CAL, the Calcutta Herbarium), and online herbaria was undertaken. The herbarium study provided clues to the possible areas for survey (Table 1) and key morphological features to distinguish *S. prostratum* and *S. laciniatum*. Additionally, habitat features associated with the presence of *S. prostratum*, were noted from earlier reports (Rao, 1971; Matthew, 1981; Irwin *et al*., 2015; Dhaarani *et al*., 2018; IL&FS, 2019).

On the basis of habitat, collection records and phenology of *S. prostratum*, the exploration trip was organized during December 2020. We had started our trip from the southern side Kodiyakkarai (Nagappattinam District) and moved along the northern side in the East Coast till Pulicat Lake (Thiruvallur District), between the latitudes 10.29 N to 13.56 N (Fig. 1). During our trip, we had primarily focused to the narrow strip of coastal tracts adjacent to the sea (up to 700 m from seashore, Rao, 1971), covering nearly 50 km (Fig. 1). Germplasm was collected following standard operating procedures (ICAR-NBPGR, 2016).

A preliminary assessment of the threat status of *S. prostratum* in India was made as per the latest IUCN guidelines (IUCN, 2022). In brief, this species is mainly evaluated to identify the degree of threat of extinction, and to categorize appropriately among the threatened categories, Critically Endangered (CR), Endangered (EN), and Vulnerable (V). IUCN guidelines provide five criteria, A) population size reduction, B) geographic range size, and fragmentation, C) small and declining population, D) very small population, and E) quantitative analysis of extinction risk. In geographic range, the area of occupancy (AOO) has not been calculated as per

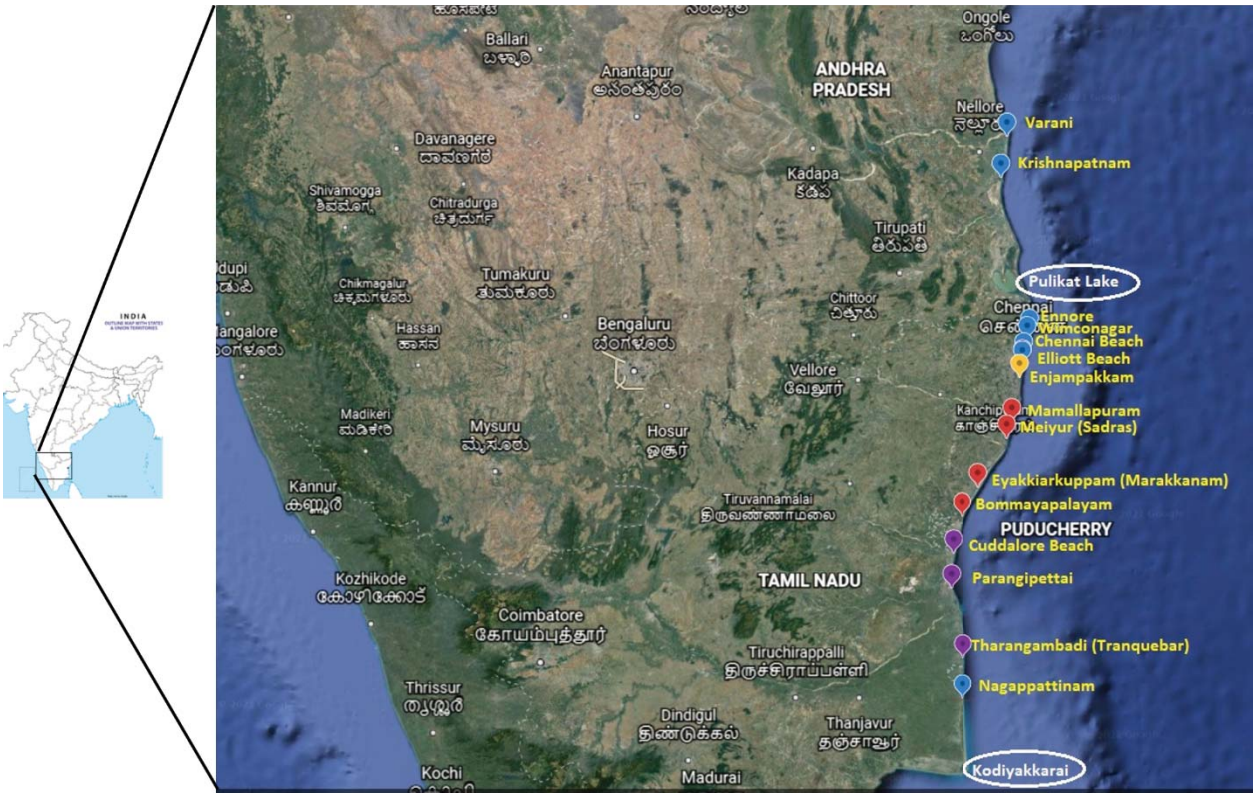

Fig. 1. Areas explored for *Sesamum prostratum* (Kodiyakkarai to Pulicat Lake, cumulative coastal walk of 50 kms at the pinned spots). Blue pinned (Nagappattinam, Elliott beach, Chennai beach, Wimco Nagar, Ennore, *Varani* and *Krishnapatnam*, italicized places not covered during this trip) and violet pinned (Tharangambadi, parangipettai and Cudddalore beach) sites are based on herbarium (#1-14 of Table 1) and literature information (#15-18 of Table 1), respectively. Red pinned (Bommayapalayam, Marakkanam, Meiyur and Mamallapuram) sites are the collection sites and yellow pinned (Enjampakkam) site, indicate only one plant seen in the spot.

the recommended 2 × 2 km grid size, since this is an endemic species exhibiting a linear habitat. The species' biological characteristic features influence the correction factor towards 1, and each site never exceeded the 150 × 500 m area, hence we were unable to quantify the AOO.

Results and Discussion

Augmenting the diversity in crop wild relatives (CWR) is a priority in recent years (Joseph-John and Pradheep, 2018). There are 18 herbarium specimens of *S. prostratum* at the MH, the CAL and other online herbaria (Table 1); in addition, there were few specimens from the inland sites, which were observed to be misidentifications of *S. laciniatum* (Pradheep *et al*., 2021). The earliest herbarium specimen of *S. prostratum* was the one collected in 1802 AD (from India) and the complete list of authentic collections sites for *S. prostratum* over a period between 1802 and 1988 are provided in Table 1 and Fig. 1. Notably, all were documented prior to the 2004 tsunami. The area along the East Coast of Tamil Nadu region, and the collection spots of *S. prostratum*, are provided in Fig. 1. At rare occasions, we observed the presence of a few plants of *S. prostratum* within 50 m from the seashore in Bommayapalayam site and up to 700 m from the seashore in Marakkanam site.

Unnoticed decline in the population of Sesamum prostratum distribution

The prominent sites from where *S. prostratum* had been collected in the past were covered in the current trip (Fig. 1), except for Krishnapatnam, and Varani [from Andhra Pradesh] (Table 1; Fig. 1). However, *S. prostratum* was located only at five locations *viz*., Bommayapalayam (700 m from Auroville Beach), Eyakkiarkuppam (Marakkanam), Meiyur (Sadras), Mamallapuram (earlier Mahaballipuram), and Enjampakkam. Interestingly most of the sites were non-overlapping, with reference to the previous specimen reports, except the last two of the five sites where *S. prostratum* is located during the trip. Seed samples were collected from these five locations except Enjampakkam, where only one plant

Table 1. List of collection spots for *S. prostratum*, as per the herbarium specimens studied at the MH, the CAL, and the other online herbaria.

S. No.	Collector No. / Ref	Place	Year	Herbarium
A. Based on specimens:				
1.	Klein	India	1802	B-W 11614-01 0
2.	Anon.		1818	K001123830
3.	Herb. Hookerianum	Chennai	1825	K00884563
4.	Anon.		1826	K001123829
5.	Anon.		1859	CAL334676
6.	G Bidies.n.	Ennore, Chennai	1879	MH00133781
7.	JS Gamble 12781	Varani, Nellore dt., Andhra Pradesh	1883	CAL, MH
8.	JS Gamble 17140	Near Chennai	1885	CAL, MH
9.	Anon.	Nagappattinam	1886	MH
10.	G Watt 12920	Mahaballipuram (Mamallapuram)	1899	CAL
11.	CA Barber 31	Elliot Beach, Chennai	1899	MH
12.	CEC Fischer 4118	Krishnapatnam, Nellore dt.	1917	CAL
13.	G Davidse & DB Sumithaarachchi 8964	Panama, Sri Lanka	1974	CAL
14.	D Narasimhan 685	ITC Compound, Near Wimco Nagar Railway Station, Chennai	1984	MH
15.	N Parthasarathy & K Ravikumar 85500	Enjampakkam, Chennai	1987	MH
16.	R Rajan 89743	Auroville Beach	1988	MH
17.	Anon.	Chennai Beach		MH00133784
18.	Anon.	Chennai Beach		MH00133782
B. Based on literature studies:				
19.	Matthew, 1981*	Cuddalore beach		-
20.	Irwin et al., 2015*	Adayar Theosophical Society		-
21.	Dhaarani et al., 2018*	Tharangambadi		-
22.	IL&FS, 2019*	Parangipettai		-

* Marked ones are as per the literature information. For details, please refer to the citation list.

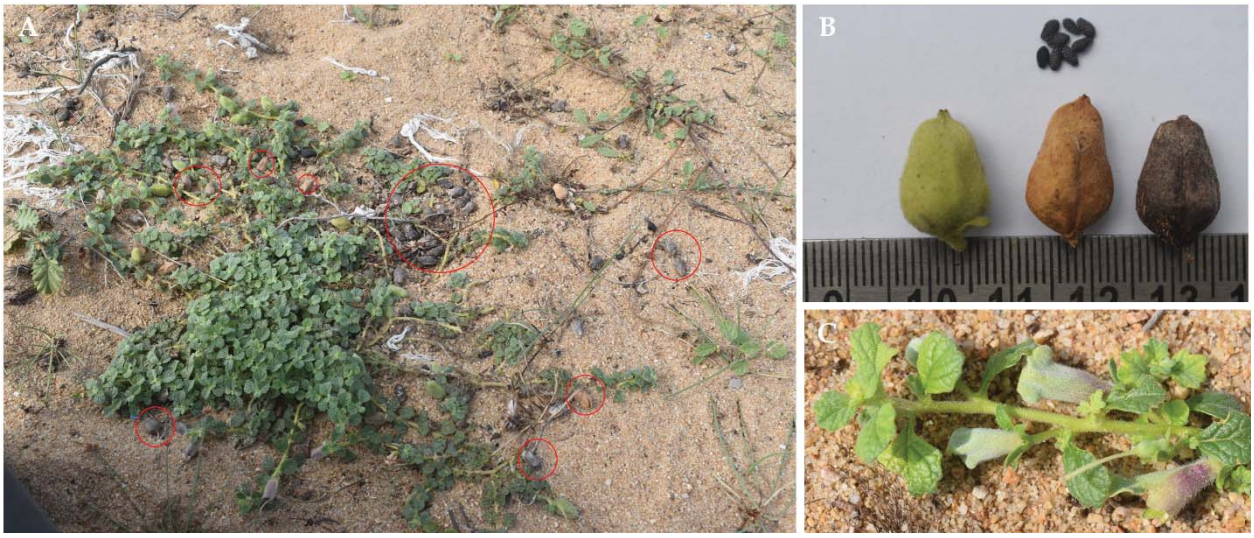

Fig. 2. A: Presence of previous years' capsules (marked as red rings in the figure) lying underneath / adjacent to the plant. B (from L to R): Simultaneous collection of current year maturing capsules (green), matured capsules of previous year (brown), matured capsules relatively older than the previous year' ones (blackish brown). C: Phyllody infection in *S. prostratum*.

was spotted, possibly due to severe urbanization and now being within the city limit. Also, of the four sites, where seeds were collected, only at Bommayapalayam and Eyakkiarkuppam sites, could a large population be located with roughly 1000 plants as a single population. Furthermore, although some reports underscore the potentiality of *S. prostratum* for resistance to phyllody (Thangavelu, 1989; Subramanian, 2003), we noticed phyllody infection in *S. prostratum* plants (upto 5%) at this site (Fig. 2). Detailed resistance reaction studies, in future, in these population would potentially help identify the resistant or tolerant genotypes that could be of potential use in introgression of genes from *S. prostratum* to sesame cultivars to overcome phyllody related yield losses. These plants exhibit a prostrate habit, there would be possibility of additional or newer insect groups responsible for the phyllody infection on the spreading-type plants, that needs detailed research. At the other two sites, Meiyur and Mamallapuram, were represented with a population of less than 100 plants in each. This indicates that *S. prostratum* is absent in roughly 71% of the sites known for previous occurrence. All the sites of location of *S. prostratum* were observed to be in isolated pockets, clearly defining the population, that are undisturbed or with minimal human interference. An independent in-depth study on the phenotypic variability for the various biometric traits, and molecular diversity assessment at intra- and inter-population level is required to understand its variability and diversity patterns. In most of the sites visited (Fig. 1), two species, *Ipomoea pes-caprae* (L.) R.Br. and *Spinifex littoreus* Merr., were dominant. However, a wild relative of jack and sword bean, *Canavalia rosea*, was present in some pockets of seashore around Chennai along with *I. pes-caprae*. Hence, a significant decline in the areas of occurrence is indicated and size of the population of *S. prostratum*, a CR species of Sri Lanka (and probably for India too), and calls for the immediate need to implement conservation strategies for such species that are at risk of severe loss of diversity and even extinction.

Preliminary threat assessment of S. prostratum as per IUCN guidelines

S. prostratum, an endemic species of India and Sri Lanka (Kumar *et al*., 2019 and citations therein; Pradheep *et al*., 2021), is categorized by IUCN as 'CR' in Sri Lanka (Wadugodapitiya *et al*., 2013); along Indian coastal areas too it appears to be under high risk. Based on the IUCN guidelines (IUCN, 2022), we have preliminarily assessed the threat status of *S. prostratum* (Table 2). The annotation as per the IUCN guidelines for *S. prostratum* is: A2ace; B1ab (i,iii)-B2ab (ii,iii); D1 (Table 2). After assessment with maximal number of threat criteria possible, we suggest that the threat assessment level for *S. prostratum* in India falls in critically endangered (CR) category. Being endemic to Indian sub-continent (India and Sri Lanka), and IUCN had already categorized this species as 'CR' in Sri Lanka, our observations and assessment (in line with IUCN guidelines except for AOO) suggest *S. prostratum* for a 'CR' in India

Table 2. Preliminary threat assessment of the *Sesamum prostratum* in India as per the latest IUCN guidelines (IUCN, 2022).

S. No	Criteria	Category			Remarks
		Critically Endangered (CR)	Endangered (EN)	Vulnerable (VU)	
A	Population size reduction (Population with reference to the number of sites)	≥80%	≥50% **A2ace (70.59%)**	≥30%	As per the 22 sites mentioned in the table 1, 8 sites were excluded (2-Andhra Pradesh, 1-SriLanka, 3-unknown, 1-documented location as India, 1- two were from Chennai Beach and probably same location), and the 3 new sites identified were included. Making it to a total of 17 sites (22-8+3), of which in 5 sites it is spotted. Percentage conversion (5/17) makes it to 70.59%.
B	Geographic range* Extent of occurrence (EOO) Area of occupancy (AOO)	B1: <100 km^2 B2: <10 km^2 **B1ab (i, iii) (<55 km^2)** **B2ab (ii, iii) (5 km^2)**	B1: <5000 km^2 B2: <500 km^2	B1: <20000 km^2 B2: <2000 km^2	**B1:** from the first spot, Bommayapalayam, to the last spot, Mamallapuram, it is ca.90 km in length. *S. prostratum* is spotted only within 150 m width (150-300 m from seashore) calculating to 13.5 km^2.Even if this is present in the northern most point (ca. 280 km from Mamallapuram), Varani-Andhra Pradesh, would add 42 km^2 area. Hence, it is roughly55 km^2. **B2:** Total area of each site never exceeded 150 × 500 m, and being 'linear' habitat (coastline) with biological characteristic influence on correction factor nears 1 for this endemic species, unit value of 1 km^2per site was used.
C	Small and declining population size Number of mature individuals (data not available for C1 and C2)	**<250** Categorized based on number of individuals in maximum number of sites with 1, 150-200, and 150-200 individuals.	**<2500**	**<10000**	Three of the five sites spotted were noted with less than 250 mature individuals. One site, Enjampakkam, was identified with only one mature individual; two sites, Mamallapuram and Meiyur were with around 150-200 mature individuals.
D	Very small population or very restricted distribution	**D1 (<50)** Number of mature individuals (**one only**) in smallest population	**<250**	**<1000**	In the smallest population site, only one mature individual has been spotted and in other two sites, Mamallapuram and Meiyur, very restricted distribution pattern was observed.
E	Quantitative analysis of extinction risk	(Data not available)			

***:** The AOO was not calculated as per the recommended 2×2 km grid size (IUCN, 2022) for the four reasons, 1) the total area per site not exceeded 150 × 500 m, 2) linear habitat, 3) biological characteristic features' influence on correction factor approaches 1, and 4) the species being endemic to Indian Sub-continent (India and Sri Lanka).

too. Hence, its high time to act and make appropriate conservation efforts by drafting key strategies and work plan to conserve this species both *in situ* and *ex situ*, to ensure it is free of extinction risk.

The reasons for the reduced number and size of populations at different sites could be due to habit and other morphological features of the species besides climate change: 1) niche-specificity of the species (absent in moist coastal habitat, tsunami affecting beaches), 2) prostrate habit with stem not rooting at nodes, 3) hard capsules, not fully dehiscent and therefore retaining nearly half the seeds in the capsule itself, 4) no seed dispersal through birds or animals, as older capsules of previous years' were found beneath/adjacent to the plant (Fig. 2), 5) around 5-20 per cent germination in *ex situ* conditions may also indicate its non-competitiveness in natural conditions, 6) habitat-based changes in the fragile ecosystem may include severe urbanization around Chennai, cyclonic storms (Kathiresan and Rajendran, 2005a, b; Sandilyan and Kathiresan, 2012; Malik *et al*., 2019). Additionally, industrialization events like thermal power plants (eg. Parangipettai) and ports (eg. Ennore) could be the potential factors for the decline in *S. prostratum* from those sites, where the sample specimens were preserved earlier (IL&FS, 2019; Table 1).

Fig. 3. A: Single plant view of *S. prostratum* from *ex-situ* conservation (stubble transplantation) at Vriddhachalam, of samples collected from Bommayapalayam site; B: flowers; and C: developing capsules from the *ex situ* conserved plant.

Conservation strategies and way forward

Impact assessment of tsunami on biodiversity for territories that are located near the tsunami epicentre, Andaman and Nicobar Islands of India (Porwal *et al*., 2012) and other parts in South-East Asia (Fernando *et al*., 2006; Suppasri *et al*., 2015) are well documented, especially for the mangrove ecosystems (Ayyappan *et al*., 2016). Availability of such assessment reports for coastal strand ecosystems too at state biodiversity level or the appropriate national or global Red List authorities would be of a help in drafting targeted action plan with strategies to include all species (flora or fauna), especially the endemic plants (Mounce *et al*., 2018).

***In situ* conservation measures:** Expanding the protected area is an important key to lock the biodiversity loss and is one of the strategic plans of Aichi Biodiversity Target 11, to conserve biodiversity (Spiliopoulou *et al*., 2021; CBD, 2012). Special need-based conservation drive especially for the non-competitive species is a real concern that requires multiple strategies and action plan. Certain non-competitive species like *S. prostratum* are very niche-specific (Rao and Sastry, 1974), especially in dry coastal strands (if wet, other competitive species dominate). In collaboration with National Biodiversity Authority (NBA), appropriate measures are being taken to earmark the Bommayapalayam site (11.99 N; 79.85 E) as a biodiversity heritage site for *in situ* conservation. This site has been selected for the *in situ* conservation, for the following reasons: 1) largest population among the sites documented with *S. prostratum*; 2) the location being accessible to public, would create a general awareness on the importance of conservation of biodiversity and nurture citizen scientists; 3) its continuity with Auroville beach areas, an important tourist site and prone to trampling damage if proper conservation measures are not taken to protect it; and 4) the site is near academic institutions, and easily accessible for frequent monitoring.

***Ex situ* conservation measures:**The TNAU-Regional Research Station, Vriddhachalam, collected *S.prostratum* accessions from Bommayapalayam and Marakkanam of Villupuram district (Fig. 3), were collected as stubbles along with coastal saline soil in a polythene bag, and with sufficient quantity of sea water to maintain the plant health till transplantation.These were planted in a concrete ring (2.5′ diameter and 1.5′ height) filled with well decomposed farmyard manure (FYM) and coastal saline soil (as cover soil) and irrigated with coastal saline water for initial 2-3days of plant establishment. From 4th day onwards, normally water was provided through drip irrigation.The matured and dried capsules were allowed to perpetuate in the cement rings itself for further successful establishment and maintenance.

Seeds of two *S. prostratum* accessions were soaked in water for 12h followed by 0.3% KNO_3 treatment for 30 min and then sown in the cement rings filled with well decomposed FYM, and drip irrigated for 4-5 h every day. These new plants were established, and flowering and fruiting were initiated during December 2021. Seeds of *S. prostratum* collected from these sites during this collection trip were conserved for the *ex situ* conservation, at the NGB of ICAR-NBPGR, bearing the accession numbers, IC0641138, IC0641140, and IC0641141.

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RESEARCH ARTICLE

Towards Strengthening the National Herbarium of Cultivated Plants with Rice Landrace Diversity

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Landrace diversity in rice was enriched at the National Herbarium of Cultivated Plants (NHCP), ICAR-NBPGR, New Delhi with addition of over 1,000 herbarium specimens. The samples were selected from material grown for characterization under an ICAR-DBT project on "Mainstreaming rice landraces diversity in varietal development through genome-wide association studies: a model for large-scale utilization of gene bank collections of rice". The paper discusses the role of NHCP in preserving landraces of rice and follow same model to other crop landraces. Addition of 17 important landraces with their characters and associated passport information data would help the users working in different fields.

Introduction

A herbarium provides a traditional classical approach for use of crop genepool diversity, variation in species, particularly in cultivated plant taxa (Harlan and de Wet, 1971; Funk, 2003). The genetic diversity stored in the form of herbarium specimens with all available data serves as an important resource of additional data for research and breeding.

Large-scale expeditions undertaken in the 19th century lead to the scientific advancement of biogeography, plant evolution, phylogenetic, systematic and biodiversity, ecology, etc. Plant explorers invariably use herbarium specimens collected from diverse habitats to synthesize information to plan and maximize their search to tap genetic diversity across phyto-geographic regions (Funk, 2018). Herbarium studies offer a quick approach to familiarize users with the species in general and landraces in particular of the target area. Using the characters of landrace and associated data, facilitate to examine the differences and their variation pattern under one roof (Diane *et al*., 2010; Bhaskar *et al.,* 2016; Pandey *et al.,* 2016). Moreover, the herbarium also serve as base material for various studies, besides preserving the plant specimen passport information (locality, habit, habitat, flowering, and fruiting time, etc.), facilitates researchers in confirming and determining the identity of new/doubtful material.

Many of the landraces and primitive cultivars have already vanished and some are on the verge of it due to the high yielding modern cultivars. The remaining ones are deteriorating gradually due to natural hybridization during seed multiplication, natural selection or genetic drift and unsuitable growing conditions. In recent times impetus on landraces conservation is mainly due to threat and vulnerablity. Unlike the wild taxa, location of crop landraces for collection remains confined due to their agro-ecological need, specificity and selection by the farmers who have played a pivotal role in age-old conservation.

The National Herbarium of Cultivated Plants (code-NHCP) at ICAR-National Bureau of Plant Genetic Resources, New Delhi, India holds significant collections of cultivated taxa and wild relatives/weedy relatives of both native and exotic origin, and taxa of potential value identified under plant genetic resources (PGR) programme (Pandey *et al*., 2015; Pandey *et al*., 2020). The NHCP is listed in the Index Herbarium which is a global directory of public herbaria in different regions (Holmgren and Holmgren, 1998; http://sciweb. nybg.org/science2/IndexHerbarium.asp). It occupies an important place among the 25 major Indian herbaria mainly dealing with plant genetic resources (Singh, 2010; Nayar *et al*., 2014). NHCP differs in its mandate from the general herbaria across the country in representing wide range

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of variability in crop plants depicted as cultivars, primitive types/ landraces, wild/semi-domesticated forms and crop wild relatives (CWR)/ weedy types and also the minor economic species collected from different agro-ecological regions of India under various PGR programmes including the introduced material under various research programme and vouchers of research material deposited.

Among the major crops of Indian origin, the cultivated rice (*Oryza sativa* L.) are also rich in the genetic diversity including the wild progenitors (Patra *et al*., 2016). In first effort of its own kind, a systematic approach towards collections of rice landraces from neglected regions, across eco-geographical regions, tribal/north-eastern region has been attempted to enrich the repository of NHCP. The germplasm of rice retrieved and purified from the National Genebank, was raised in the experimental area at Indian Agricultural Research Institute (IARI), New Delhi, India. This paper mainly aims to highlight the role of NHCP in general and landraces of rice in particular in complementing the holdings of plant genetic resources and their documentation. The objectives of this work are to introduce the readers about the role of NHCP in preserving of landraces of various crops, including rice; and follow same model to other crop landraces.

Materials and Methods

A total of 10,086 rice landraces collected from different rice growing areas of India out of 1,15,152 (as on December 2021) accessions of rice landraces collected from different agro-ecogeographical regions of the country and conserved in the National Gene Bank, ICAR-NBPGR, New Delhi formed the basis of this study (http://www.nbpgr.ernet.in/Research_Projects/Base_Collection_in_NGB.aspx). The germplasm of rice landraces was characterized during *kharif* 2020-21 at IARI fields, New Delhi under the ICAR-DBT project on "Mainstreaming rice landraces diversity in varietal development through genome-wide association studies: a model for large-scale utilization of gene bank collections of rice".

Of 10,086 rice landraces, more than 1,000 samples were selected based on uniqueness of morphological characters, geographic representation, and other unique traits. Three specimens of each accession were selected and processed as per herbarium standard method (Jain and Rao, 1977). For processing and drying of the specimens no preservatives were used except sprinkling with the naphthalene powder to keep specimens under insect-free conditions. The label data were drawn from the original databank 'as it is'. Besides a subset of over 50 mature spikes were added in spike collection of NHCP.

For landrace nomenclature, the authors have relied on data especially for the name(s) of the different landraces stored under the National Genebank database. Validation in some cases was possible through literature but in others, names could not be checked and therefore treated 'as it is'.

Result and Discussion

The NHCP has 25,283 herbarium specimen's representative of 267 families, 1,546 genera and 4,378 species (as on March 31, 2021) of important taxa of plant genetic resource (PGR) (Pandey *et al*., 2021). In the past an effort has been laid for collection and preservation of landraces from the Indian gene center. ICAR-NBPGR has undertaken studies on plant systematics through field and herbarium resources to extract useful information on various eco-geography of crop genepools. In the national perspective, among the existing herbaria which cater to the diversity specific collections of crops plants, and potential taxa of PGR value, the 'National Herbarium of Cultivated Plants' at ICAR-NBPGR, New Delhi with its ten regional stations serves for the benefit of users. The herbarium of the M.S. Swaminathan Research Foundation (MSSRF) at Chennai, India at its Centre for Sustainable Agricultural and Rural Development lays emphasis on similar taxa represented from the Eastern and Western Ghats respectively (Arora, 1994).

Assembling distinct rice landrace diversity

In the past, rice collections were added in the NHCP through specimens/seeds collected from various sources, explorations undertaken in different agro-ecological zones of India, material introduced from abroad under various research/breeding/selection programmes and vouchers deposited of the systematic studies on crop-groups (Pandey *et al.,* 2014, Pandey *et al.,* 2016; Pandey, 2019; Pandey and Pradheep, 2019). Under various PGR activities collections especially the landrace diversity was made from the North-Eastern region, West Bengal and Uttar Pradesh, India (Semwal *et al*., 2014). Earlier holdings of 250 rice landraces were assembled through efforts by the ICAR-NBPGR mainly represented from Bihar, Karnataka, Kerala and Odisha and the machine vision project "Use of Machine Vision for Distinguishing

Among Crop Varieties" at IARI and NATP project. Among the recent landrace collections native rice varieties identified for mother trial seed multiplication (2018-19) in project site of Assam were added through Assam Agriculture University and Foundation for Development Integration, Guwahati, Assam.

Several collections of rice landraces of India were maintained in the Indian Museum (CNH; earlier known as Bengal Economic Museum), Kolkata, West Bengal. A catalogue on races of rice in India compiled before the Bengal Economic Museum became part of the Indian Museum, Kolkata, West Bengal through the province all its districts (The Agricultural Ledger, 1910) provide details on the rice races grown during that time. However, presently the collection of cultivated plants in general and landraces in particular are least represented in global as well as the national herbarium collections. At national level, NHCP is one among the herbaria focusing on cultivated plants study.

Distribution of Landraces

The rice landrace germplasm collections conserved in NGB from different states of the country are denoted by a generic name '*dhan*' in majority of cases but also have a specific name used by traditional farmers. One such example is landrace 'Govindbhog' showing variation in panicle characters. These landraces, are confined to West Bengal, Madhya Pradesh, Chhattisgarh, Uttar Pradesh, and Odisha. Some more examples of similar types are 'Kala nunia', 'Kala namak' that are reported from wider localities.

Among the 1,000 indigenous rice landraces, Odisha and Chhattisgarh were the major contributors of representation followed by Madhya Pradesh, Karnataka and Kerala. Other states were less represented in the collection (Fig. 1). The list of landraces name and affiliation of state of origin and cultivation are provided in Annexure 1. A total of 99 exotic landraces are represented from 22 countries; the top listed were Bangladesh, followed by Philippines, China and others (Fig. 2).

These landraces under cultivation are represented from diverse regions of the country and are routinely collected and conserved in the national collection (Joseph and Abdul, 1998; Mark, 2014; Ghosh *et al*., 2019; Rana *et al*., 2009). Apart from the morphological diversity, they showed diverse potential to adapt to the climatic changes and allelic variation for resistance to biotic and abiotic stresses (Hyles e*t al.*, 2020). For example, *boro* rice of Assam is known for suitability in water logged area having stagnant water or flood conditions. The diversity was noted for colour and size of kernals, aroma, growing season (*Sali, bora, aus* in North Eastern Region; *kuruvai, samba* in southern region), the inflorescence type, or the spikelet arrangement, etc.

Some of the selected landraces included in NHCP are discussed below (arranged alphabetically):

Ambemohar: small grain aromatic rice is popularly grown in Maharashtra but also popular in other parts of the country. It is also known for its excellent flavour among the non-basmati rices.

Badshabhog: this landrace is primarily with the state of Chhattisgarh and is characterized by highly aromatic small grain. It finds its origin during Mughal regime and the word 'bhog' is the offering to 'Hindu Gods'. However, this landrace is also grown in West Bengal, Odisha, Madhya Pradesh, Uttar Pradesh, Bihar, Jharkhand, Assam and Maharashtra.

Basmati: is a speciality group of rice known world over for its long grain quality and excellent aroma. It fetches premium export value in the international market. Majority of the produce comes from the Indo-Gangetic plains or the Tarai region of the country.

*Chennallu***:** is one among various landraces that have been used in India for their medicinal purpose. It grown by 'Mavilan' tribe of Kerala who administer popped rice soaked in water to feed to diarrhea patients.

Chinikapoor: is a long slender grain type of landrace from Uttar Pradesh which is classified under aromatic group. It is also known under cultivation in Maharashtra, West Bengal and Chhattisgarh.

Eravapandy: a traditional rice landrace from Kerala is being used in the international and national breeding programmes. It is known as a source of gall midge resistance.

Gobindbhog: the traditional rice landrace with aroma is used during special occasions and offered to the God Gobind (Lord Krishna) and hence the name 'Gobindbhog'. It is a fine grain aromatic rice which has got GI tag from West Bengal. This is also grown in the neighbouring states, Uttar Pradesh, Odisha, Chhattisgarh and Madhya Pradesh. In present landrace holdings a lot of variation among the morphological traits was recorded. This traditional/cultural practices of aromatic

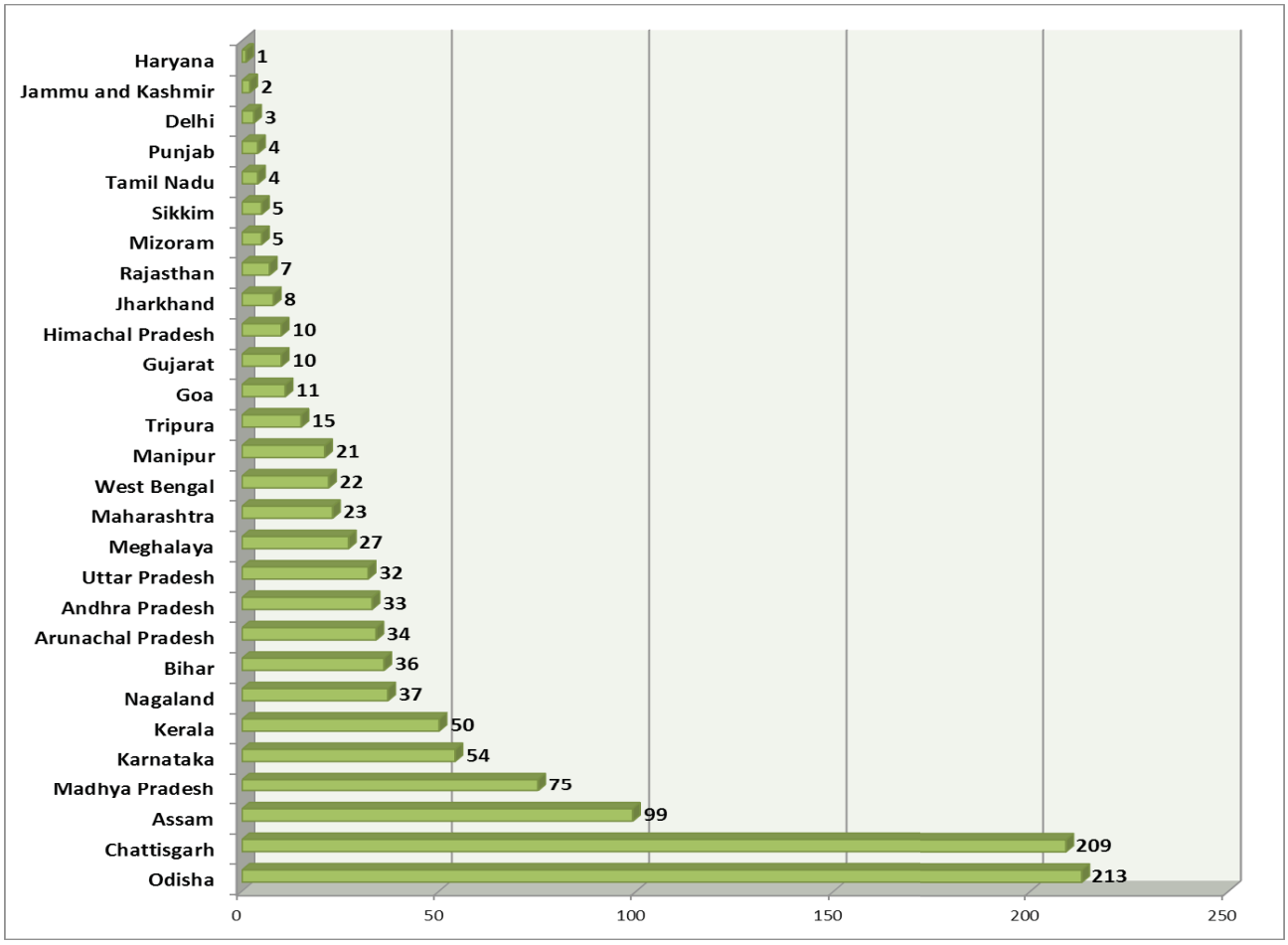

Fig. 1. Herbarium specimens of rice landraces at NHCP represented from different states of India (bar represents total landraces from that state)

rice being offered to god must have also been practiced for other landraces of different region and with the same terminology 'Gobindbhog'.

Hathia panjara: it is a traditional landrace from Chhattisgarh which has characteristic double spikelets and also lodging resistance. The landrace has an average taste and medium market value but yields high in conditions when no commercial fertilizer is used.

Ishwarakora: finds its place in the pedigree of the 77 CVRC released rice varieties in the country. This landrace is known for tolerance for both abiotic to biotic stresses.

Jal dooba: is a traditional rice variety from Odisha suitable for sub-mergence tolerance. It is very late duration variety with strong culm suitable for cultivation under lowland conditions.

Kala joha: cultivar has unique aroma, super-fine kernel, good cooking qualities, antioxidant properties and excellent palatability. It is grown mainly in Assam and other North-eastern states. It has comparable aroma and quality as that of other scented rices of India, except the elongation ratio. The Johah as GI status given to 43 known varieties of the Joha rice which are grown in the region.

Lalnakanda: a sticky and aromatic rice from Odisha and Tarai Himalayan region which is popular for preparation of soups. It finds a better price as compared to the other common landraces in the region/country.

Mohan bhog: it is one of the non-basmati farmer's varieties from Madhya Pradesh. This landrace is grown traditionally for small grain and high aroma occupies sizable acreage due to aroma, fineness and tolerant to biotic and abiotic stress.

Pokkali: it is a unique saline tolerant rice variety that is cultivated in an organic condition in the water-logged coastal regions of Alappuzha, Thrissur and Ernakulam

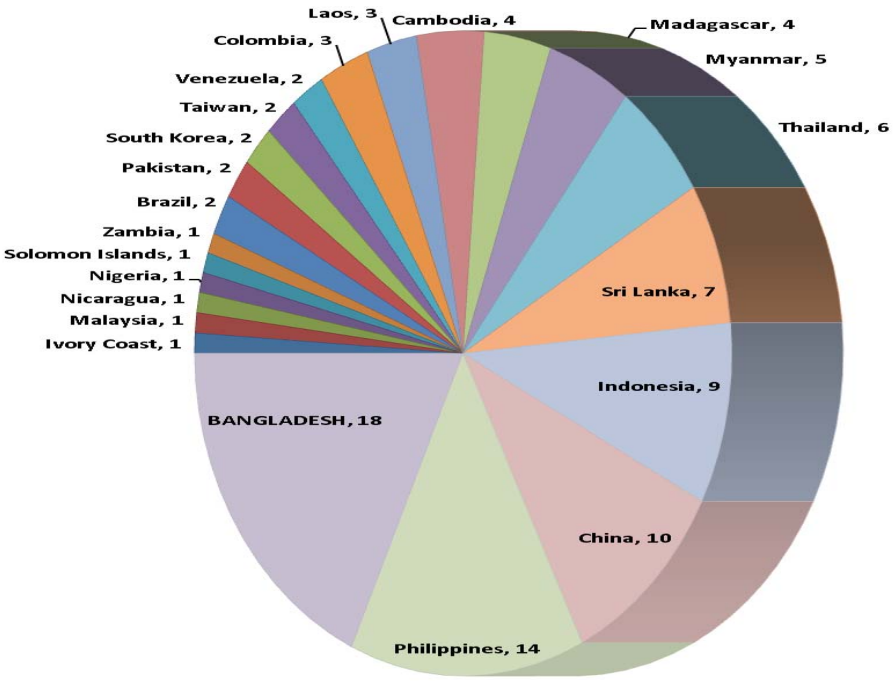

Fig. 2. Herbarium specimens of rice landraces at NHCP from different countries (no. represents landraces from the country)

districts. The organically-grown Pokkali is famed for its peculiar taste and its high protein content. It is one of the important landraces from which the major 'Saltol locus' was derived and used in breeding programme. The 'saline tolerance' trait was derived from other important landraces, viz. *Getu*, *Nona bokra* and others.

Potti akkullu: it is mostly grown in coastal districts specifically for the straw used to thatch the huts and as a feed the cattle. It has compatible degree of resistance to foot-rot disease.

Thavalakannan: is a landrace from Kerala, finds its use in pedigree of 19 CVRC rice varieties. This is one of the popular varieties of Palakkadan Matta (bold red rice with a unique taste); drought resistant, flood tolerant known for resistance to rice tungro virus (RTV) and BPH, also it is more nutritious, as good source of iron, zinc and vitamin B6. It can thrive under low input costs.

Tulapunji: is a landrace from West Bengal which has an excellent aroma with medium-long grain.

Herbarium specimens of landraces depicted flag leaf and part of stem, panicle and kernel at mid maturity with husk colour fully developed and in most cases kernel with awns. Since the kernels are the most distinguishing traits in a landrace, herbarium sheet with panicle and seed stored in pouches are attached on the sheet. The data recorded in the herbarium label provided characters of kernel, awans-colour, length, size; flag leaf-size, and angle; stem ribs and hairyness, stiffness, hollowness, etc. Some characters like stem type, lower leaf size, resistant traits and tiller numbers, aroma was noted in the label information (Fig. 3).

NHCP intends to facilitate landraces of crops for PGR studies

The NHCP intends to facilitate users for study on landraces and knowledge upgradation on the following:

- Support information diverse ecological of collecting sites, range of ecological amplitude and locating ecotypic variation.
- Charting distribution of crop landraces and its mapping for *ex-situ* representation
- Assessing gaps in the collection and conservation, prioritizing areas for future collecting, especially rare and localized types.
- Providing information on ethnic reference, preferences of users, agro-ecology
- Potential, physiological or stress traits (water logged, salinity tolerance, medicinal value, etc.).

Fig. 3. Herbarium specimens of rice landraces: (1-2) with important notes on label; (3-4) landrace 'Govindbhog' showing variation in panicle characters

- Source of material availability for molecular studies

Advantages of representing diversity in herbarium

Due to quick adoption of high-yielding rice varieties during last 4-5 decades, landraces have been restricted to localized cultivations (Deb, 2005). For plant genetic resources study, depiction of rice landraces diversity, in the form of genebank conservation and preserving in herbarium were found benificial, as evaluated below:

- Characters of full panicle available over separated seeds conserved in the genebank
- Character assessment of landrace identity based on morphological characters such as flag leaf, stem strength, kernel size, etc.
- Available seed/ kernel/husk as complementary seed collections with herbarium specimens
- Easy access of large landrace samples at a time for comparative study
- Comparitive analysis of wider landrace diversity
- Samples available for other studies (morphological, biochemical, molecular)
- Exchange/access to digitized images for larger users without legal hassles
- Easy demonstration and teaching of landrace diversity

Conclusions

National Herbarium of Cultivated Plants (NHCP) at ICAR-NBPGR since 1980's after its functioning has done a commendable work in field of taxonomy and systematics of taxa of PGR relevance. Co-ordinated efforts at the national level need to be streamlined to evaluate priorities of crops of PGR relevance to gear up work on the management of genetic resources on the one hand and for basic research on cultivated plant taxonomy (which is more intricate but needs better deal), particularly when India happens to be a centre of diversity for many crop plants.

At national level, link with other herbaria working on PGR needs to be established. With the existing infrastructure for cultivated taxa, the NHCP take responsibility to priorities adding more crop landraces in phased manner to facilitate greater success of national programme.

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ANNEXURE 1

Check-list of landraces* in NHCP

S. No.	State	Landraces (in alphabetical order)*
1	Andhra Pradesh	*Akashmelo, Baiyagunda, Basmati, Battagunda, Chokmanthu, Danwar, Dasara mathi, Davedya, Deshi, Kasturi, Deshi kuseri, Dhouri, Dodgui, Dukke, Errabadal, Gadakhunta, Hathi panjar, Jajin, Jira, Kakdiha, Kanhai, Karidhan, Karendole, Kasam Chudi, Kolhapur Scented, Mettavari, Milagrosa, Nandi, Potti Akkulu, Raipuri hagurma, Sukla Gurmatia, Swarrna, Yerrasannadhanyam*
2	Arunachal Pradesh	*Amkel Ahu, Ampakhi, Amyong Ahu, Grondla,Jajoni bardhan, Jakar, Khamti Sali, Mugme, Serpum, Sukarakaya, Thio Sali, Tinin, Tuying Sali, Thayang, Upma*
3	Assam	*Aanjan, Abrini mi mittem, Ahima Fhutki Sali, Aihati, Bamak, Bardhan Sali, Boga balam, Bogabora, Biriajam, Borh, Pual Sali, Chokuwa, Chit Muchhi, Dangor bayahunda, Darmali, Daria, Darma, Daula, Dhipa bhajana, Dhubuli, Dudha dhan, Ful badam, Hampori Chokura, Heera jyota, Jahari, Jikal, Julami, Kala joha, Ketchadi, Kharka Jaha, Khokhadi, Koimurali, Kothiya, Laicha, Lashur khudwani, Lawadaba, Losmon bhug, Mairen, Menaka, Narayanpuria, Paita bora, Raimukhi sali, Raksail, Ramsali, Rangun Sali, Reia dhan, Santisaha, Sadian Pakhi, Shaitya Sali, Sikkim Basmati, Silguri, Solpuna, Sonajul, Sonamoti, Solpuna, Sonaril, Sorujahingia dhan, Sunasur lahi sali, Tapar, Tulsi joha, Tumai*
4	Bihar	*Abonisail, Banglasafri, Barogawa, Bhadia dhan, Budha dhan, Chaurai, Dahia, Daria, Darnia, Deobhog Gamiri, Garemalet, Givalpor, Gola dhan, Jaisinga, Jhara, Jhulanwa, Kajaroo, Kalikamod, Kariokanha, Koya, Luchai, Lahama, Lahana, Manipur, O Muslim Dhan, Ratho munda, Rajal dhan, Rasbhog, Sagarbhog, Samer dhan, Sarkarma, Tehri sukla, Tikar*
5	Chhattisgarh	*Aardhana luchai, Aelmidi, Ajankati, Alsengha Goda, Bachcha kalam, Bagabardhana, Bahush bod, Bakoi, Bamia, Barangi, Baranmburia Barondao, Basangi, Basua bhog, Bega Hudi, Bhainsamundaria, Bhatha bhulau, Bhata Gadakhunta, Bhatta Gadakuta, Bhata Gurmatia, Bhata kabari, Bhata kaliyari, Bhata swarna, Bhatama karam, Bhejari Deshi, Birla Bhanga, Black pattu, Bodikaluni, Bodama Raipur, Bolandabazar, Budha Koila, Budma Wadlu, Bhudkud, Chah, Changpalngat, Chaurala, Chhote Baikoni, Chhote Sathka, Chingar Chopa, Chinsura, Chiraiphola, Chmuchiggum, Dahi Gurmatia, Dai, Damar, Dapa, Dengi chudi, Dhaiya Dhan, Dhumniya, Dodga, Dol, Diwadi, Dudhnag, Dudhiya Potiya, Durdulanga, Fangsin, Gahi, Gajara, Gampuriya, Gandhak, Gathu dhan, Gatta, Ghathul, Gitti, Gobindbhog Goda, Gurmatia Naguir, Hansir, Hardi Ganth, Haruna luchia, Harrakanth, Hiran Pongari, Kajla bunde, Kakadisar, Kakamaranga, Kalamali, Kalapana, Kalchi, Kalkh, Kali Mai, Kandjhiya, Kankariya, Kankari phooli, Karanga dhan, Kariya benikath, Karni, Kathir, Kekering, Khargilas, Khariko john, Kari Koliyari, Khairka khuchi, Khera Basang, Kher Khouli, Khuti, Kkiyaketiki, Kodesarlu, Koharin, Kolyar, Kordur Kati, Kosari, Kunkhari, Kurlukh, Kurma, Jangal Jihati, Jal, Jaleminda, Jhilli Maniphool, Jhilliparag, Jholar, Jiladar, Jira chudi, Jouchang, Juagonar Sali, Jugul banku, Lagin Chudi, Lakshmi belag, Lalat, Lali Majhoi, Laludhan, Lallu Sagar, Latiya Gurmatia, Lati luchai, Laza, Lim Chudi, Loharsidhi, Luhera, Mahsuri dwarf, Malchi, Malko, Malrani kajar, Mandri, Mandariya, Maradmalkhan, Mati Tenga, Matko, Mauha, Mekarajhundi, Mogenga, Moirang, Mote, Moti dhan, Muhar, Makhanguda, Mukhara, Nagu barhi, Namba, Narangi, Nawa Singul, Nijo, Nimdhan, Nirgubi, Niwadi, Orayeeboot, Padari, Pahuni, Pandrin, Parbat kala, Parau Gurmatia, Patelpachisi, Pinwari Luchai, Pipariya, Pita luchai, Pode, Pohasal, Pora, Poran Sona, Punai Solto, Safed Deshi, Saina Goda, Sam Pasan, Sadka, Sathaka, Satlamara, Shadhar Champa, Shahnagar, Sindur Chinga, Soap, Sona Kathi, Sukharuwala, Sukla Pora, Surhi, Surmati, Raigadhiya Gurmatia, Raishri, Rajhusa, Rani kajal, Ruiphool, Taitungal, Thumsi, Tjumaki, Tolbal, Turiya, Umariya, Uswa, Vijaya mahsuri, Vilayat Chudi, Viranjphool, Wanga Walk*
6	Delhi	*Basmatibahar, Punjabi*
7	Goa	*Banakumar, Barik Katsal, Dhamna Pinda, Dhavopandyo, Kalomanik, Kempadu, Middlekendal, Nanya, Sambarsal, Surja Mukhi, Zadi Dangar*
8	Gujarat	*Baresal Madheli, Chandanchur dhan, Fulako, Karikaman, Karnatara, Khadrhiyalal Kimari, Kodakuri, Lal Goda, Local*
9	Haryana	*Tiu*
10	Himachal Pradesh	*Chandan Kath, Dhonal, Jaldhepa, Jaran, Jiridhan, Kuruvai, Rudha, Tiu, Zag*
11	Jammu and Kashmir	*Biyee, Govindbhog*
12	Jharkhand	*Alubilu, Brusabkisan Hathi Panjara,Kanamati, Mangur Mudi, Mansoori Dhan, Ranikajal Dhan, Tenkarola*
13	Karnataka	*Anilam Anil, Arrampottan, Bangara kolee, Bangara Sanna-3, Banka, Bebbana, Beliki, Boo jaddu, Chandravali, Chippige, Dugga bhalha, G 1, Gamnada Batta, Gidhan Pakhi, GK-5, GK-7, GK-9, Hompal Gidda, Honasu, Intan Gidda, Jeervel, Jeerugodu, Kadulile, Kaggali kecrona, Kamdari, Kanakunja, Kappu batta, Kari Alshi, Kari kandaka, Kariga Javele, Karidadi gossi, Kari Swarna, Kavekantak, Kempu doddisal, Kochuthonnuran, Krishna Leela, Kurud, Manjupani, Mavaokar, Misse batta, Moranda, Mugad Suganda, Nawali, Nazar Bat, Neergula batta, Neermullare, Pokkali, Puhkutt Kodi-2, Punkutt kodi-1, Putta batta, Rahodaya, Sanna batta-2, Sanna mallige-2, Somsali.*
14	Kerala	*Ambala dhan, Ambe, Ariyan, Athiyan Kootumundon, Buh, Choman, Chenkayama, Chethuvadi, Chopru Ekyu, Clamme, Cult. Kurathache, Cutt. Kunnurai, Dilbaxa, Eravapandy, Farm Chitteri, Guria karma, Harikat dhan, Jaya, Karaga, Kattasemba, Kegie, Kempathi bhatta, Kottar Samba, Kozhppulli Pokkali, Lavdhan, Luchyee, Madhu, Mangari, Maji Ranga Bardhan Sali, Maraninboota, Methi Mahipal, Munnamvila, Navara, Nahazing, Nouva, Onavatan, Palguna, Panancheri, Pandakari, Phote local, Rajakayama, Sakthi, Thaichundan, Thavalakanan, Thekkancheera, Thotta kaima, Tulsiphwi, Veluthadichan*

S. No.	State	Landraces (in alphabetical order)*
15	Madhya Pradesh	*Aama Ghul, Ajan Piwari, Anakodan, Angian, Auria Buta, Badhumani, Badsa bhog, Bag Moonch, Baka, Bakawand, Balsen, Batri, Binjo, Bohita, Budali, Budali Banko, Bega hudi, chhote Haslo, Chiraigudi,Chhui Khadan, Chhura,Chhoti Luchai, Desa, Dhan Deshi, Dhawra Basant, Dhodki bhath, Deshi lal mota, Gajudhai, Gajraj, Halikilal Luchai, Jaimahakali, Kadamphool, Kallu Kamod, Kardi, Karmi, Kelakhamish, Kosam Khadi Badi, Kota Deshi, Koram, Kubri Mohar, Kusiyari, Kuwalari, Laloo, Lal Bahari, Lal Baurash, Lalmati, Lalkotam, Luchai Safed, Luji, Lunagi, Matraj, Motisar, Mudri Gurmatia, Mazhla Gurmatia, Nadawar, Nago, Nagesar, Nariayal Jhopa, Ragunath, Ramkaroni, Ranikaja, Ratad deshi, Rated, Saitu Gurmatia, Sikiya, Sironj, Surmalia, Sisath, Viranch*
16	Maharashtra	*Ayakhu Ketezuwa, Buh, Dodaki, Dhanesal, Gharkhat, Halvaziniya, Himasal, Iswra kora, Lavesal, Lavha mugad, Masins rice, Marisal, Palashkeda barik, Pandharisal, Phougak, Picharde, Shennel, Sonephala, Sonebhat, Takebhat, Taothali, Vargol*
17	Manipur	*Akhi joha, Changmansan, Chalau, Changadi, Darria, Hmgmihsiangmatsn, Makodo, Mandu, Maora, Minil Bija Bini, Napchong, Phoungang Angan phou, Phonlem, Phazai, Sonajuli Sali,Taothabi Angouba, Tong Khollen bhupal, Tumai, Tumai Angouba*
18	Meghalaya	*Gelon, Dalleymarshi, EB:17, Lowguti, Kbarim, Mekatchu, Mimitim Michibol, Mima Mitambing, Narajib, Sona Juli Sali, Siltukri Sali, Tharrangsing*
19	Mizoram	*Karusi, Malapotipatnai, Sukulemba, Tzulu Narila*
20	Nagaland	*Asin Mai, Athikan, Chali, Changphai, Chusuro Mono, Eravapandy, Hai Dhorom Dhan, Konra, Laloo, Longi, Moyou, Ncpateni, Nangcha Tsuk, Nziera, Origasho, Rajpateni, Rugopet, Ribolu, Ropu, Saheb juha, Shimoi, Sitabanwas, Soloh Kabu, Sonporo Ekyov, Sungro Ekyu, Suli Tsuk, Suli Tsuk Tsokunkvu, Surudaka dhan, Tahzyah, Teheang,Thiethieru, Tikapateni, Tipfu Shye, Tzu Mabok/Tzumasu, Yamkok*
21	Odisha	*Alsanga, Anaikomban, Arkal bhat, Bachakalam, Badhimami, Basgati, Basan, Baskbanda, Basmati Aman, Bayalachampa, Benigiri, Bhondu, Bhogi, Bhojni, Bhutah Gay, Biranchi pool, Bodi, Bodki, Bogihali, Changang, Chikoo, Chheligudia, Chenga, Chinai, Chinikapoor, Chingfore Chokua, Chirainokhi, Chotasail, Chilakat, Chilhar, Chinai,Chirhola, Chittal chini, Chittikannerulu, Chhotobelki, Cuttack chandi, Daanr, Dakhuri khuji, Dalbadal, Daliya, Dalipohala, Danger, Dasahar Amathi, Dasarakanta, Dasara Mathi, Deepak, Dekaradokar, Deulbhog, Dhal champa, Dhalakiri, Dhamsi, Dhaneberwa local, Dhara Dhuta, Dharial, Dudhasai, Dhaud, Dhikash, Dhobjira, Dhola Champa, Dhenkisali, Dhula kakara, Edolia, Gandhak, Gayasu, Gobindbhog. Gudumani,Haldi jhota, Hangnyang, Hardighati, Haribhog, Harratkhat, Himalaya, Horujhangia, Jabra Mardan, Jaga Balia, Jaladhuti, Jaldoba, Jaradola, Jhati, Jorkusumalu, Kalchi, Kaliasia, Kamal, Kamod, Kanai bashicn, Kanchi, Kanchi Ratna Chudi, Kanthabako, Kanthamala, Karangaguda, Karprui dhan, Kartik pateni, Kaldeyakhuji, Keppu, Khaojee, Kuchi Siliguti, Kurippala, Kuruma, Kusumakunda, Laktimachi, Lalkanda dhan, Langphou Phougang, Latika, Latamoul, Lera, Loudubi, Madhi, Madmalen, Mahamya, Mainaguri, Mahulbahal, Matarmala, Matchkati, Mehapal, Mohilkuchi, Motakhatia, Motara, Mukusuala, Muskhbadji, Mypali, Nagana, Naherkeli, Nai badhai, Naikani, Nalikalam dhan, Nanimundi, Nathmohan, Nazate Khudwani, Nazusamba, Nimekanta, Oabu Jiuku, Pahad jhili, Pakhuda Chhada, Panama komba, Pandavi, Papasaphuli, Parijathiki, Pavurturma, Poibasangi, Puage, Rajmlli, Rajuhendi, Ramaboiti, Ranga Lachai, Ranganath, Rangobhonda, Rangabankoi, Rangoon Samba, Rangakalama, Rangsuri, Rangisali, Raskola, Ratna, Ratan Chudi, Ratna Pokhiai, Ratanpanjar, Red Sirumani, Runiakali, Saanra, Sadadumracn, Sakar Mator, Salakana, Sanakumaguntha, Sareada, Sajani, Salekdam, Septidhan, Shalaidhonti, Sharad Chadi, Sornavari, Singha, Singhujnupa, Siulipana, Sthani Halva, Suka, Sukal Mundi, Sukarkaya, Sukasari, Suna Khadika, Sunamuibarapanua, Sundarbhajna, Sundarsali, Surda, Swarna, Tamthomamdinh, Taramukhi, Tella balchilu, Tetilia, Tinikalam, Thapulin khudwani, Ujog, Upusali, Useli*
22	Punjab	*Basmati Bahar, Basmati Kamon, Chalu Jeera, Jhetinbari*
23	Rajasthan	*Duggabhalha, Katghora Mansara, Scgera, Suthar*
24	Sikkim	*Dalleymarshi, Darash dhan, Krishna bhog Khavarisal, Salalai,*
25	Tamil Nadu	*Murungukaran*
26	Tripura	*Bedi, Chennallu, Dup Galong,Gangaorasad, Lal Koram,Lanka Pora, Murali early, Panikelash, Jamrai, Dharial, Galra, Sinduri, Latakalasuna Mukhi Asus, Sayari,*
27	Uttar Pradesh	*Badukaibatiyari, Beniga, Cheenajohan, Daftol, Dakhinkalma, Dalbadal, Dhaniya, Dudha kalama, Faram Chudi, Goviindbhog, Jarhen, Julysal, Kabrabadam, Kapoor Kanti, Local dhan,Lurca, Luhera, Mwarhi, Pyapachini, Red Ratha, Sarfed nakanda, Sarankrahm, Sondhi, Sullenpur, Thakur bhog, Timili*
28	West Bengal	*Dudhkanthi, Durar phool, Gangia, Govind bhog, Hijli, Kakro, Kala chudi, Kookley, Lalnkanda, Malgoindi, Mansara dhan, Mariach bhog, Marich mukhi, Patnai Sagar, Rani dhan, Ratan pakhia, Sadabochi, Simlaya, Suna phulia, Tulai panji*

*: name as stored in NGB database

ANNEXURE 2

Some of the selected landraces included in NHCP and their passport data

S. No.	IC No	Vernacular Name	Coll Date	Coll. no./other ID	Locality	Latitude/Longitude	
1	IC323554	Ambemohar	3/1/2000	Ambemohar	Maharashtra	NA	NA
2	IC99994	BadshahBhog	10/21/1985	P-111	Dehradun, Uttarakhand	30.3165	78.0322
3	IC323620	Basmati	3/1/2000	NA	Punjab	NA	NA
4	IC461262	Chennallu	NA	AC-3765	NA	NA	NA
5	IC300525	ChiniKapoor	3/1/2000	C:30I KH-99-1482	Raipur, Chhattisgarh	21.2514	81.6296
6	IC598101	Eravapandy	5/19/2012	RJR-681	Adilabad, Telangana	19.7327	78.6427
7	IC124946	GobindBhog	3/28/1974	CGR:5363	Bastar, Chhattisgarh	19.2073	81.9339
8	IC342613	Hathipanjara	1/31/2002	VKG21/6	Khunti, Jharkhand	23.0833	85.2833
9	IC460410	IswaraKora	NA	NCS976/ AC-17976	Maharashtra	NA	NA
10	IC514380	JALDOBA	4/12/2005	IRGC-45863	NA	NA	NA
11	IC323725	Kalajoha	3/1/2000	KalaJoha	Assam	NA	NA
12	IC462111	Lal Nakanda	7/4/2006	JBS-884 /AC-20907	Koraput, Odisha	18.8561	82.7347
13	IC115058	Mohanbhog	11/22/1977	CGR:8509	Mandla, Madhya Pradesh	22.5979	80.3714
14	IC324584	Pokkali	10/28/1990	M-14/90/ NIC 4909	Ernakulam, Kerala	10.0833	76.5483
15	IC323750	PottiAkkullu	3/1/2000	MTU-1	Andhra Pradesh	NA	NA
16	IC324567	Thavalakannan	1/1/1989	V.4449	Kerala	NA	NA
17	IC594017	Tulaipanji	12/31/2011	DP/HNS-2006	South 24 Parganas, West Bengal	22.1894	88.8143

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RESEARCH ARTICLE

Inheritance of Resistance to Downy Mildew [*Pseudoperono sporacubensis* (Berk. and Curt.) Rostovzev.] in Ridge Gourd [*Luffa acutangula* (Roxb.) L.]

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Ridge gourd [*Luffa acutangula* (Roxb.) L.] is one of the important cucurbit vegetable cultivated widely in India and other Asian countries. Downy mildew caused by *Pseudoperono sporacubensis* (Berk. and Curt.) Rostovzev severely affects the ridge gourd production during rainy season. Therefore, in the current study 12 genotypes including some promising inbred lines were screened for downy mildew resistance which resulted in the selection of three moderately resistant advanced selections *viz*., IIHR-17-2-1 (PDI -16.25), IIHR-7-5-1 (PDI -21.40) and IIHR-17-1-7-3 (PDI -21.60) with low AUDPC values (467.59-634.26).Genetics of downy mildew resistance in two different F_2 populations derived from IIHR-52-1-30 × IIHR-17-1-7-3 and IIHR-23-8-10 × IIHR-7-5-1 respectively revealed that two major genes are interactive in dominant suppression epistasis and complementary epistasis. These results will aid ridge gourd breeder to develop a strategic downy mildew disease resistant breeding program.

Key Words: Downy mildew screening, Inheritance, Resistance, Ridge gourd

Introduction

Ridge gourd [*Luffa acutangula* (L.)Roxb.] is one of the important cucurbits grown as a cash crop in many tropical and sub-tropical regions of the India and abroad. It is enriched with vital elements such as vitamin C, zinc, iron, riboflavin, magnesium, thiamine and dietary fibre and therefore, contribute to human nutrition. It is traditionally described as a medicinal plant which used to cure a number of ailments. In India, it is commercially grown in Andhra Pradesh, Tamil Nadu, Karnataka, Gujarat, Maharashtra, Assam and West Bengal during spring-summer and rainy season and provides a livelihood for resource-poor farmers.

However, the rainy season crop is found to be severely affected by downy mildew disease caused by *Pseudoperono sporacubensis* (Berk. and Curt.) Rostovzev. It is one of the important foliar disease, causing significant yield losses in region with high humidity and rainfall. The pathogen survives on live plants for reproduction and sporangia produced are dispersed through air, rain splash or physically through equipments (Lange *et al*., 1989). Survival of sporangia is highly depended on the prevailing environmental conditions (Thomas,1996). Since the identification of *P. cubensis* in Cuba by Berkeley and Curtis (1868), this pathogen has been reported from 70 countries worldwide infecting more than 20 genera of cucurbits (Lebeda and Urban, 2007). Downy mildew management relies on aggressive spray programme in conjunction with resistant varieties and cultural techniques to reduce the losses. However, efficacy of fungicides control has been diminished with increasing insensitivity of *Ps. cubensis* population towards it (Reuveni *et al*., 1980; Thomas and Jourdain, 1992; Heaney *et al*., 2000). Resistant varieties, an integral component of disease management programme, are economically viable and environmentally safe solution to manage this devastating disease. Host resistance has been identified in cucumber (Barnes and Epps, 1954; Wehner and Shetty, 1997; Shetty *et al*., 2002) and melon (Lebeda and Widrlechner, 2003; Taler *et al*., 2004). However, there is no report on downy mildew disease resistant sources and inheritance pattern in ridge gourd. Efforts are underway to identify ridge gourd resistance source(s) against downy mildew disease at ICAR-Indian Institute of Horticultural Research (IIHR), Bengaluru since 2010. The present research work was taken up to confirm the downy mildew resistance

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in advanced inbred lines of ridge gourdand further studying its inheritance so that resistance loci could be incorporated into a commercial ridge gourd cultivar.

Materials and Methods

Natural Screening and Disease Assessment

Ridge gourd germplasm screening trials for downy mildew screening was initiated at ICAR-IIHR, Bengaluru since 2010. Resistant selections were advanced through selfing for 3-4 generations. Finally, four advanced promising inbred selections (IIHR-17-1-7-4, IIHR-7-5-1, IIHR-17-2-1, IIHR-17-1-7-3), two advanced breeding lines (IIHR-53-1-3, IIHR-6-1-1), two released varieties (Arka Sujat, Arka Sumeet), two popular hybrids (Naga, NS-03) along with two highly susceptible checks (IIHR-52-1-30, IIHR-23-8-10) were screened against downy mildew resistance during *kharif* season. All these genotypes were true-to-type with highly uniform plant type due to continuous selfing for 3-4 generations.

Screening experiment was conducted in randomized block design with three replications (six plants per replication) by repeating susceptible checks between the test rows and around the field to facilitate the disease infection. Each genotype was sown in a single row-to-row and plant-to-plant spacing of 150 cm and 50 cm respectively. Recommended cultural practices except the application of fungicides were followed throughout the growing period. Plants were scored based on the 0-9-point interaction phenotype scale as suggested by Jenkins and Wehner (1983) where 0=No disease, 1=Few small leaf lesions, 2=Few lesions on few leaves with no stem lesions, 3=Few lesions on few leaves or with superficial stem lesions, 4= Few well-formed leaf lesions or superficial stem lesions, 5=Few well-formed leaf lesions or enlarging stem lesions, 6=Many large leaf lesions or deep stem lesions with abundant sporulation or plant more than 50% defoliated, 7= Many large coalescing leaf or stem lesions, over 75% of plant area affected or defoliated, 8=Plants largely defoliated, leaf or stem with abundant sporulating lesions and 9= Plants dead. Downy mildew incidence was recorded after appearance of several disease symptoms on the susceptible checks (45 days after sowing) and continued throughout the growing period at 30 days interval. Total three scoring were used to calculate percent disease index (PDI) as per the formula followed by Jamadar and Desai (9) and Santhosh (17). Average PDI was used to calculate apparent infection rate (r) and area under and disease progress curve (AUDPC) for each genotype (Vander Plank, 1963; Wilcoxen *et al.* (1975).

Artificial Screening and Disease Assessment

Artificial screening by leaf disc assay method of the four advanced selections along with a susceptible check (IIHR-52-1-30) was conducted during *kharif* as per method described earlier (Salati *et al*., 2010). Seven leaf discs per plant (three plants per genotype) placed abaxially onto 0.4 per cent agar medium in petri-plates were inoculated with three droplets (10 µl) of inoculum suspension and incubated for 5 days at 20 ºC temperature, 80 per cent relative humidity and photoperiod of 16 h. One petri-plate with seven leaf discs without inoculation were kept as control. Petri dishes were stored under dark condition for sporulation at 20 ºC. Test plants were scored for disease development after 7 days post inoculation (dpi) using a binocular magnifier. Plants were rated on 0-4 phenotypic scale where 0= no sporulation; 1= light sporulation (difficult to see with naked eye) 2 = sporulation area inferior to the diameter of the deposited inoculum droplet; 3 = sporulation area corresponding to the diameter of the deposited inoculum droplet; 4 = sporulation area superior to the diameter of the deposited inoculum droplet. Percent disease index (PDI) was calculated as per the formula followed by Salati *et al*. (2010).

Inheritance of Downy Mildew Resistance

This experiment was conducted in two different crosses. In the first cross, highly susceptible ridge gourd advanced line 'IIHR-52-1-30' (female parent) was crossed with the promising line IIHR-17-1-7-3 (male parent) whereas in the second cross, highly susceptible line IIHR-23-8-10 (female parents) was crossed with the advanced inbred lines IIHR-7-5-1 (male parent) to get two F_1 populations. The F_1 plants were selfed to get F_2 seeds and back crosses (BC1; F_1 × susceptible parent, BC_2; F_1 × resistant parent) were also performed to evaluate the various inheritance patterns determined from analyzing the F_2 segregation data. In the field, 30 plants of susceptible parent (IIHR-52-1-30), 30 plants of resistant parent (IIHR-17-1-7-3), 30 F_1 (IIHR-52-1-30 × IIHR-17-1-7-3) plants, 178 F_2 individuals, 29 susceptible back cross progenies (F1 × IIHR-52-1-30) and 30 resistant back cross progenies (F1 × IIHR-17-1-7-3) were evaluated against the downy mildew disease under high disease pressure conditions during *kharif* season.

Another F_2 population with 45 plants obtained by crossing IIHR-23-8-10 × IIHR-7-5-1 was also evaluated to study the genotypic effect, if any, in the inheritance pattern of downy mildew resistance in ridge gourd. Twenty-nine plants of susceptible parent (IIHR-23-8-10), 30 plants of resistant parent (IIHR-7-5-1), 7 F_1 (IIHR-23-8-10 × IIHR-7-5-1) plants, 45 F_2 individuals, 13 susceptible back cross progenies (F_1 × IIHR-23-8-10) and 30 resistant back cross progenies (F_1 × IIHR-7-5-1) were evaluated against the downy mildew disease under similar epiphytotic conditions.

Plants were rated on 0-9 phenotypic scale as suggested by Jenkins and Wehner (1983). PDI was calculated as per the formula followed by Jamadar and Desai (1999) and Santhosh (2011). Mean PDI data was used for the calculations.

PDI	Reaction categories
1-10	Resistant (R)
11-25	Moderately resistant (MR)
26-50	Moderately susceptible (MS)
51-75	Susceptible (S)
>75	Highly susceptible (HS)

Results and Discussion

The reaction of test genotypes of ridge gourd to downy mildew disease under field and laboratory conditions is summarized in Table 1 and 2. Three advanced lines have showed promising response towards downy mildew resistance as there were mild symptoms of downy mildew throughout growing period. The susceptible lines (IIHR-52-1-30 and IIHR-23-8-10) were highly infected with symptoms of leaf lesions, stem lesions, intensive defoliation with abundant sporulation. The disease development was very slow and delayed in four advanced inbred selections when compared with highly susceptible lines (Fig. 1). Among twelve genotypes at advanced stage of screening, three genotypes viz., IIHR-7-5-1, IIHR-17-2-1, IIHR-17-1-7-3 (PDI= 11.00-25.00) (Table 1, Fig.1) were found to be moderately resistant, IIHR-17-1-7-4 and Arka Sumeet were found to be moderately susceptible (PDI=26.00-50.00). Rest of the selections and commercial varieties and hybrids were found to be susceptible and mildew growth was observed on leaves (Table 1).

AUDPC values widely varied from 467.59 (IIHR-17-2-1) to 2819.00 (IIHR-23-8-10) under different screening conditions (Table 1). Three advanced selections with lower PDI also had lower AUDPC values which were ranging between 629.00-810.00 (Table 1). Most susceptible genotypes showed higher PDI and AUDPC values which indicated the significance of disease parameters used in the study. Apparent infection rate of advanced selections was slightly higher because of some infection in the later stages of the crop growth (105 DAS) (Table 1). PDI values of test genotypes in natural and controlled screening were positively co-related and correlation coefficient values under natural epiphytotic condition was 0.97 (Table 2, Fig. 2, 3). These moderately resistant sources against downy mildew along with alreadyidentified sources in melon (Goswami *et al*., 2011; Lebeda and Widrlechner, 2003; Taler *et al*., 2004) and cucumber (Shetty *et al*., 2002; Wehner and Shetty, 1997) remain useful in integrated disease management programme. These identified lines of ridge gourd can

Table 1. PDI, AUDPC and apparent rate of infection against downy mildew in ridge gourd advanced inbred selections under field conditions

S. No.	Genotype	PDI			Mean PDI	Disease Reaction	AUDPC	Apparent rate of infection (r)
		45 DAS	75 DAS	105 DAS				
1	IIHR-17-1-7-4	0.62	20.74	65.92	29.09	MS	810.19	0.15
2	IIHR-7-5-1	1.85	19.75	42.59	21.40	MR	629.63	0.11
3	IIHR-17-2-1	0.00	13.58	35.18	16.25	MR	467.59	0.14
4	IIHR-17-1-7-3	0.62	19.75	44.44	21.60	MR	634.26	0.14
5	IIHR-52-1-30	77.16	99.38	100.00	92.18	HS	2819.44	0.01
6	IIHR-23-8-10	76.67	98.89	100.00	91.85	HS	2808.33	0.01
7	IIHR-6-1-1	69.14	91.36	99.26	86.58	HS	2633.33	0.01
8	IIHR-53-1-3	52.22	74.44	96.67	74.44	S	2233.33	0.02
9	ArkaSumeet	24.20	46.42	64.07	44.90	MS	1358.33	0.03
10	ArkaSujat	61.11	83.33	93.83	79.42	HS	2412.04	0.01
11	Naga	55.56	77.78	96.91	76.75	HS	2310.19	0.02
12	NS-03	35.19	57.41	77.78	56.79	S	1708.33	0.03

PDI-Per cent disease index; AUDPC-Area under disease progress curve; DAS-Days after sowing; r-mean apparent rate of infection; MR-Moderately resistant; MS-Moderately susceptible; HS-Highly susceptible; S-Susceptible

Fig. 1. ***Field screening*****: IIHR-7-5-1, moderately resistant inbred selection (*left*) and highly susceptible inbred selection, IIHR-52-1-30 (*right*) against downy mildew in ridge gourd**

Fig. 2. ***Artificial screening*** **(left control, right inoculated leaf discs in each photo): IIHR-7-5-1, moderately resistant inbred selection (*left*) and highly susceptible inbred selection, IIHR-52-1-30 (*right*) against downy mildew in ridge gourd**

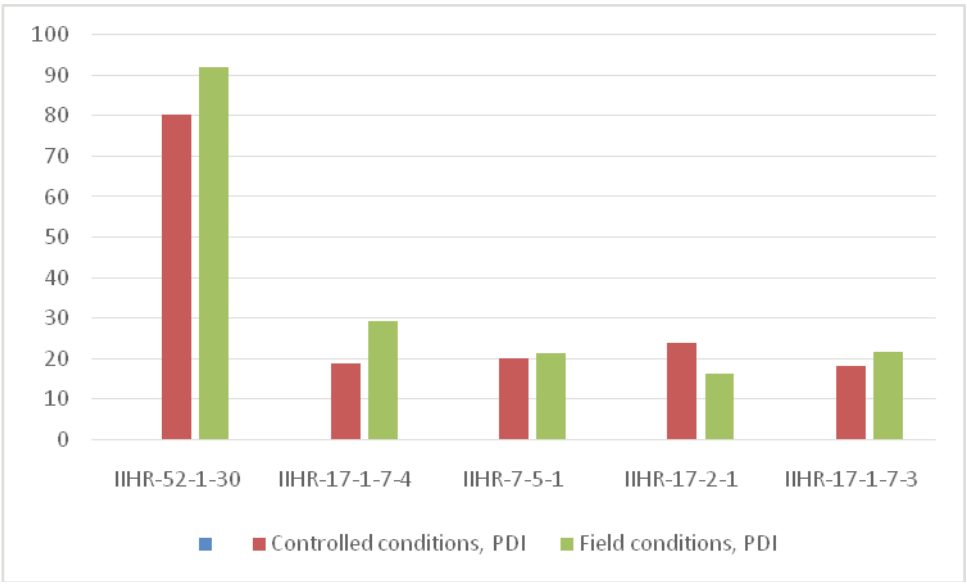

Fig. 3. Comparison of inbred selections PDI under controlled conditions and field conditions

Table 2. Disease response of selected ridge gourd advanced selections along with susceptible checks under field and laboratory (Controlled) conditions

Genotype	Controlled conditions		Field conditions
	Sporulation Rank	PDI	Mean PDI
IIHR-52-1-30	3.21	80.16	92.18
IIHR-17-1-7-4	0.75	18.65	29.09
IIHR-7-5-1	0.80	20.11	21.40
IIHR-17-2-1	0.95	23.81	16.25
IIHR-17-1-7-3	0.72	17.99	21.60
Correlation coefficient			0.977**

** Significant at 1% level
PDI-Per cent disease index

be advanced further for improvement in downy mildew resistance.

The segregation in F_2 and backcross progeny of the two crosses was subjected to chi-square analysis for assessing the goodness of fit to various classical Mendelian ratios (Table 3). Data represented in Table 3 showed that all the plants of inbred lines used as female parent, i.e. IIHR-52-1-30 were susceptible while those of the male parent IIHR-17-1-7-3 were resistant to downy mildew disease. The F_1 plants showed susceptible reaction against downy mildew infection. The results may indicate the dominance of susceptibility over resistance. The number of segregants into resistant and susceptible classes in the F_2 generation was 33 and 145 respectively. Out of the various Mendelian ratios tested, the chi-square values were significant for all ratios except 3 (resistant): 13 (susceptible) with a chi square value of 0.005 and probability of 0.90-0.95 which suggested that two pairs of genes were responsible for resistance to downy mildew disease in IIHR-17-1-7-3. In the test cross with resistant parent, the segregation ratio had the best fit with 1:1 (χ^2= 0.533; P = 0.40 – 0.50). However, as in the test cross with susceptible parent, the segregation ratio had goodness of fit with 1:0 (χ2 = 0.00; p = 1.00) *i.e*., all the plants were susceptible. These results showed that the parents were different in pairs of epistatic interaction and expressed dominant and recessive interaction. Thus, two pairs of genes of dominant and recessive interaction governed the inheritance of downy mildew disease in ridge gourd. Similarly, El-Hafez *et al*. (1990) mentioned that resistance to downy mildew in cucumber plants is controlled by two pairs of dominant and recessive interaction genes (13 susceptible: 3 resistant). Badr and Mohamed (1998) also reported similar results for downy mildew resistance in cucumber. However, so far, reports on genetic analysis of downy mildew resistance in ridge gourd are not available.

In the second cross IIHR-23-8-10 × IIHR-7-5-1, the number of segregants into resistant and susceptible classes in the F_2 generation was 26 and 19 respectively. Out of the various mendelian ratios tested, the chi square values were significant for all ratios except 9:7 (resistant: susceptible) *(*χ^2 = 0.0.043; P = 0.80 – 0.90). In the test cross with susceptible parent, the segregation ratio had the best fit with 1:3 (χ^2= 0.026; P = 0.80 – 0.90). While in the test cross with resistant parent all the plants were resistant, hence the segregation ratio had goodness of fit with 1:0 (χ2 = 0.00; p = 1.00). The available evidence supported the presence of complementary gene action for the downy mildew resistance. This segregation pattern was not reported by any workers for downy mildew resistance in cucurbits. However multiple genes for downy mildew resistance have been reported in melon and cucumber (Epinat, 1994; Ren *et al*., 2009; Criswell

Table 3. Estimates of chi square values and their probability for classical Mendelian ratios for downy mildew resistance in the F_2 and test cross population of ridge gourd

Cross		Generations	Number of Plants			Genetic ratio	Chi square value	P value
			Resistant (R)	Susceptible (S)	Total	R:S		
I.	IIHR-52-1-30 x IIHR-17-1-7-4 (S × R)	P1	0	30	30	-	-	-
		P2	25	5	30	-	-	-
		F1	0	30	30	-	-	-
		F2	33	145	178	3:13	0.005	0.942
		BC1	0	29	29	0:1	-	-
		BC2	17	13	30	1:1	0.533	0.465
II.	IIHR-23-8-10 x IIHR-7-5-1 (S × R)	P1	0	29	29	-	-	-
		P2	30	0	30	-	-	-
		F1	7	0	7	-	-	-
		F2	26	19	45	9:7	0.043	0.836
		BC1	3	10	13	1:3	0.026	0.873
		BC2	28	2	30	1:0	-	-

et al., 2011; Zhang *et. al*., 2013; Yoshioka *et. al*., 2014; Wang *et. al*., 2016).

The difference in gene interaction in the two crosses studied,may be attributed to the presence of set of non-allelic genes in male and female parents. Therefore, it has been concluded that the complicated nature of digenic dominant interaction restricts the understanding on nature of inheritance and steps forwards to improve the crop to sustain downy mildew resistance. Therefore because of the complex nature of inheritance, the breeding programshould be accompanying with crossing followed by recurrent selection which may be effective to get desirable recombinants with downy mildew resistance.

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RESEARCH ARTICLE

Potential of Gynoecious Line in Generating Superior Heterotic Hybrids in Bitter Gourd (*Momordica charantia* L.)

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An experiment was designed to find out the extent of heterosis in bitter gourd using gynoecious line, KAU-MCGy-101 and three monoecious lines *viz*., Preethi, Priyanka, and MC133. Hybrids were generated by both direct and reciprocal crosses. All the hybrids exhibited earliness in terms of days to first female flower opening, node bearing first female flower, and days to first harvest. The hybrid KAU-MCGy-101 × Priyanka exhibited significantly higher heterosis with regard to heterobeltiosis (-16.03 %) and standard heterosis (-18.63 %) for days to first female flower opening. All the six hybrids manifested significant standard heterosis for sex ratio in the desirable negative direction, ranged from 44.25 to -55.52 %. The highest yield was recorded in the hybrid KAU-MCGy-101 × Priyanka (22.88 t/ha) and was followed by KAU-MCGy-101 × Preethi (20.42 t/ha) and KAU-MCGy-101 × MC 133 (20.05 t/ha). Heterobeltiosis for yield (t/ha) varied from 14.72 to 57.85 % and the standard heterosis ranged between 37.59 to 89.31 %.The best performing hybrids were KAU-MCGy-101 × Preethi, KAU-MCGy-101 × Priyanka, and KAU-MCGy-101 × MC 133 with respect toearliness, number of fruits per plant, and yield. Thus, the superiority of hybrids having gynoecious line as a maternal parent is prominent in the present study.

Key Words: Gynoecious hybrids, Heterobeltiosis, *Momordica charantia*, Standard heterosis, Yield

Introduction

Bitter gourd (*Momordica charantia* L., 2n=22) is an economically important cucurbitaceous vegetable known for its immense medicinal properties. This vegetable with a bitter taste is popular and grown extensively in India, China, Japan, South East Asia, Tropical Africa and South America. Bitter gourd shows variation in fruit size, color, surface texture, and edible maturity throughout the cultivated area (Robinson and Decker-Walters, 1999). A region-specific variation in consumer preference is observed for fruit color, shape, and size (Dey *et al*., 2008).

Bitter gourd is predominantly a monoecious crop with cross-pollination as a rule. Hence heterosis is well exploited for early harvest, higher yield and other agronomic traits (Alhariri *et al.*, 2018). The development of hybrids in the bitter gourd is labor-intensive because of hand pollination. But the reports on gynoecious bitter gourd lines give scope for less expensive hybrid seed production (Behera, 2004; Dey *et al.*, 2010). Gynoecy is a condition where all the flowering nodes produce only female flowers (Airina *et al*., 2013). Hence open pollinated seeds in the gynoecious parent will be F_1 hybrids and hybrid seed production becomes more economical by reducing the cost of male flower pinching and hand pollination (Behera *et al*., 2009). Inheritance of gynoecy has been well documented in cucumber and is commercially employed for hybrid seed production (Kumar and Singh, 2004). In bitter gourd, gynoecism is under the control of a single recessive gene (Ram *et al*., 2006). Though the utilization of gynoecious lines for crop improvement programs is limited in bitter gourd, it is having immense potential (Ram *et al*., 2002). The succeeding generations using gynoecious line as a maternal parent exhibited a very high percentage of pistillate flowers with high yield potential both in cucumber as well as bitter gourd. (Dey *et al*., 2010; Shukla *et al*., 2014; Kumari *et al*., 2021). A highly stable

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bitter gourd gynoecious line, KAU-MCGy-101 was identified from Kerala Agricultural University (Minnu Ann, 2019) which can be efficiently utilized for crop improvement programs. Scientific literature pertaining to the use of gynoecious lines for heterosis breeding in bitter gourd are very scarce (Behera *et al*., 2009; Dey*et al*., 2012; Alhariri *et al*., 2018). Hence, the present experiment was formulated to investigate the scope of heterosis breeding exploiting gynoecious line in bitter gourd.

Materials and Methods

Development of F_1 hybrids

The work was undertaken in the experimental field of the Department of Vegetable Science, College of Agriculture (COA), Vellanikkara, Kerala Agricultural University (KAU), Thrissur, during the period of 2019-2020. Gynoecious line KAU-MCGy-101, identified from the Department of Vegetable Science, COA, KAU, and three monoecious lines *viz*., Preethi, Priyanka, and MC 133 were used for the hybridization. Preethi and Priyanka are the two promising varieties with high yield and light green fruits, which were released from KAU. MC 133 is a high yielding inbred line with white fruits. Ten plants per genotype were maintained in the crossing block, following recommended cultivation practices (KAU, 2016). Six hybrids were developed by crossing KAU-MCGy-101 with Preethi, Priyanka, and MC 133. KAU-MCGy-101 was used as both male and female parents. Hermaphrodite flowers induced in KAU-MCGy-101 by spraying 200 ppm silver thiosulphate solution after the first female flower emergence (Minnu Ann, 2019), which were used as a pollen source in the crosses involving KAU-MCGy-101 as a male parent.

Evaluation of parents and F_1 hybrids

The six hybrids along with the four parents were evaluated in Randomized Block Design with three replications. Preethi was considered as the standard variety. Observations such as days to first female flower opening, node bearing first female flower, sex ratio, number of seeds, number of harvests, fruit length (cm), fruit girth (cm), fruit weight (g), days to the first harvest, number of fruits per plant and yield (t/ha) were taken from five randomly selected plants from each replication. Analysis of variance was done for all characters using GRAPES **(**Gopinath *et al*., 2020). Genetic parameters *viz*., heterobeltiosis (HB%) and standard heterosis (SH%) were calculated as the deviation of the mean performance of hybrids from their better parent and standard variety as proposed by Briggle (1963) and Hayes *et al*. (1965).

Results and Discussion

Mean performance

Significant differences were observed among the parental lines and their hybrids for all the characters interpreted. The range of mean values of different traits observed in the parents and hybrids are depicted in Table 1.Considerable variations were observed among the parents and hybrids for the traits determining the earliness of a variety/hybrid. Comparing with the monoecious parents, the gynoecious parent, KAU-MCGy-101 exhibited earliness in terms of minimum days to first female flower opening (32.28), lowest node number bearing first female flower (13.89) and minimum days to first harvest (52.61). Among the monoecious parents, Priyanka was found superior for the traits attributing earliness such as, minimum days to first female flower opening (34.67), lowest node number bearing first female flower (19.67) while, the minimum days to first harvest was observed in Preethi (61.44). the earliness of the gynoecious bitter gourd lines in comparison to the monoecious lines was also reported by Behera *et al*. (2009) and Dey *et al*. (2010). Among the hybrid combinations KAU-MCGy-101 × Priyanka was found to be earlier than both the gynoecious and monoecious parents, followed by KAU-MCGy-101 × MC 133 and KAU-MCGy-101 × Preethi. Yield (parents=14.49 to 12.08 t/ha and hybrids=22.88 to 16.63 t/ha) and yield attributing traits like sex ratio (parents=0.00 to 21.56 and hybrids=8.18 to 10.25), fruit weight (parents= 105.40 to 163.33 g and hybrids=118.07 to 172.47 g), fruit length (parents=16.65 to 22.01cm and hybrids=15.82 to 21.84 cm), fruit girth (parents=14.99 to 18.59 cm and hybrids= 15.94 to 17.27 cm), number of harvests (parents= 6.17 to 9.83 and hybrids= 9.00 to 12.00) and number of fruits per plant (parents= 40.17 to 57.33 and hybrids= 44.67 to 69.67) also showed ample variations among the parents and their hybrids. Among the parents used for hybridization, the lowest sex ratio was found in the gynoecious line KAU-MCGy-101 (0.00) whereas; the highest was in Priyanka (21.56). As regard to sex ratio (male:female), the crosses MC 133 × KAU-MCGy-101 and Priyanka × KAU-MCGy-101 registered the lowest (8.18) and highest (10.25) values, respectively. Among the hybrids, minimum days to the first harvest were registered in the hybrid KAU-MCGy-

Table 1. Mean performance of parents and hybrids for earliness and yield characters

	Days to first female flower opening	Node bearing first female flower	Sex ratio (Male: Female)	Fruit weight (g)	Fruit length (cm)	Fruit girth (cm)	Number of seeds	Days to first harvest	Number of harvests	Number of fruits per plant	Yield (t/ ha)
Preethi	35.78	20.56	18.39	133.67	18.01	17.33	22.20	61.44	6.17	40.17	12.08
Priyanka	34.67	19.67	21.56	163.33	22.01	18.59	29.07	65.33	7.50	42.50	12.93
MC 133	35.67	19.78	13.13	153.40	20.43	15.45	33.87	61.67	8.67	44.50	13.10
KAU-MCGy-101	32.28	13.89	0.00	105.40	16.65	14.99	19.13	52.61	9.83	57.33	14.49
Preethi × KAU-MCGy-101	31.78	14.44	8.62	118.07	15.82	17.27	15.73	58.44	11.33	62.17	17.64
Priyanka × KAU-MCGy-101	32.67	14.95	10.25	137.00	17.01	16.11	16.67	59.67	9.33	46.17	16.63
MC 133 × KAU-MCGy-101	31.33	15.56	8.18	163.07	19.26	16.68	33.53	57.67	9.00	44.67	18.13
KAU-MCGy-101 × Preethi	31.89	16.89	9.57	165.13	21.84	16.25	21.80	57.89	12.00	61.17	20.42
KAU-MCGy-101 × Priyanka	29.11	12.89	8.94	172.47	20.93	17.15	32.87	56.44	11.33	69.67	22.88
KAU-MCGy-101 × MC 133	31.78	15.89	9.94	153.20	19.19	15.94	34.47	57.44	10.33	63.33	20.05
CD (0.05 %)	3.45	3.04	3.345	22.74	2.71	0.98	3.78	5.44	3.06	13.05	2.478
CV (%)	6.15	10.78	17.96	9.05	8.25	3.45	8.50	5.39	18.67	14.31	8.58
SEd	1.64	1.45	1.592	10.82	1.29	0.47	1.80	2.59	1.46	6.21	1.179

101 × Priyanka (56.44) followed by KAU-MCGy-101 × MC 133 (57.44). This is comparatively very early than monoecious parents which require a period of 61.44 to 65.33 days for the first harvest. The general approach of selecting parental lines based on mean performance does not give a valid result (Kumar *et al*., 2017). Therefore, we have determined the heterotic potential for all the traits under study.

Heterosis

The extent of the heterotic response of the F_1 hybrids largely depends on the breeding value and genetic diversity of the parents included in the cross (Geleta and Labuschagne, 2004). From the mean performance, it is clear that a wide variation exists for the earliness and yield attributing traits of gynoecious and monoecious parents. For most of these traits, the hybrids registered markedly significant heterosis. The characters like days to first female flower opening and node bearing first female flower are considered as good indices of earliness. All the hybrids manifested heterosis for these characters in the desirable negative direction (Table 2). The cross KAU-MCGy-101 × Priyanka exhibited highly significant heterosis of -16.03 and -18.63 percentage in terms of heterobeltiosis and standard heterosis for days to first female flower opening. Crosses *viz*., Preethi × KAU-MCGy-101, MC 133 × KAU-MCGy-101, KAU-MCGy-101 × Preethi and KAU-MCGy-101 × MC 133 also registered significant negative heterosis for days to first female flower opening over better parent and standard variety. Among the six hybrids, the lowest node number (12.89) bearing the first female flower appeared in the cross KAU-MCGy-101 × Priyanka, which showed statistically superior heterosis of -34.46 per cent over its better parent and -37.30 per cent over the standard variety. Significant negative heterosis was observed in all other hybrids for the same trait. There are reports of hybrids involving gynoecious line in bitter gourd and cucumber exhibiting heterosis in a negative direction for days to first female flower opening and node bearing first female flower (Dey *et al*., 2012; Airina *et al*., 2013; Jat *et al*., 2015; Alhariri *et al*., 2018).

Similar to other cucurbits, the ratio between male and female flowers is the crucial determining factor for high yield in bitter gourd. A lower sex ratio is always desirable for high yield and productivity (Thangamani and Pugalendhi, 2013; Alhariri *et al*., 2018). Hence, heterosis in the negative direction is desirable for this trait. All

the crosses registered significant heterosis over standard variety (Table 2), which was mainly due to the high mean value of the standard variety for sex ratio (18.39). With respect to heterobeltiosis, the superior hybrid was KAU-MCGy-101 × Priyanka (-58.56 %) followed by Preethi × KAU-MCGy-101 (-53.13 %). It is important to note that these hybrids having a gynoecious parent are exhibiting earliness along with a lower sex ratio (male:female). The results of the present investigations are in accordance with the findings of Dey *et al*. (2012). Khan and Behera (2011) also reported a lower sex ratio (male:female) in gynoecious × monoecious hybrids, compared to the monoecious × monoecious hybrids of bitter gourd. Heterosis in the desirable negative direction in terms of heterobeltiosis and standard heterosis was found in all the hybrids for days to first harvest (Fig. 1). A highly significant heterobeltiosis of -13.61 per cent was observed in the hybrid KAU-MCGy-101 × Priyanka. Days to the first harvest is a critical observation as it directly contributes towards the earliness.

In the case of fruit weight, the highest significant heterobeltiosis of 23.54 per cent was expressed by the hybrid KAU-MCGy-101 × Preethi, while the hybrid KAU-MCGy-101 × Priyanka showed 29.03 per cent in terms of standard heterosis (Fig. 1). KAU-MCGy-101 × Preethi and KAU-MCGy-101 × Priyanka were the best performing hybrids with respect to fruit length (Table 2). Meanwhile, most of the hybrids showed negative heterosis for the fruit girth. In this case it is discernable that, most of the combinations with gynoecious line as one parent yielded long and slender fruits compared to the monoecious parents. Generally, long slender fruits in bitter gourd are more preferred in the market. Hence these hybrids are in accordance with the consumer predilection. On contrary, Dey *et al*. (2010) reported significant positive heterosis for fruit girth in gynoecious × monoecious hybrids of bitter gourd. The deviation could be on account of the variation in genotypes used in hybrid combinations. Regarding the number of seeds, significant heterosis was observed for the hybrids KAU-MCGy-101 × Priyanka and KAU-MCGy-101 × MC 133 (Table 2). The hybrid KAU-MCGy-101 × Priyanka manifested 13.07 per cent heterosis over better parent and 48.05 per cent heterosis over the standard variety. Comparatively a higher heterosis of 55.26 per cent over standard variety was obtained in the hybrid KAU-MCGy-101 × MC 133. For all the hybrids standard heterosis was found to be significant for the number of harvests ranging from 45.95 to 94.59 per cent.

The number of fruits per plant is one of the most important traits, which directly contributes towards yield (Alhariri *et al*., 2018). The magnitude of heterosis for

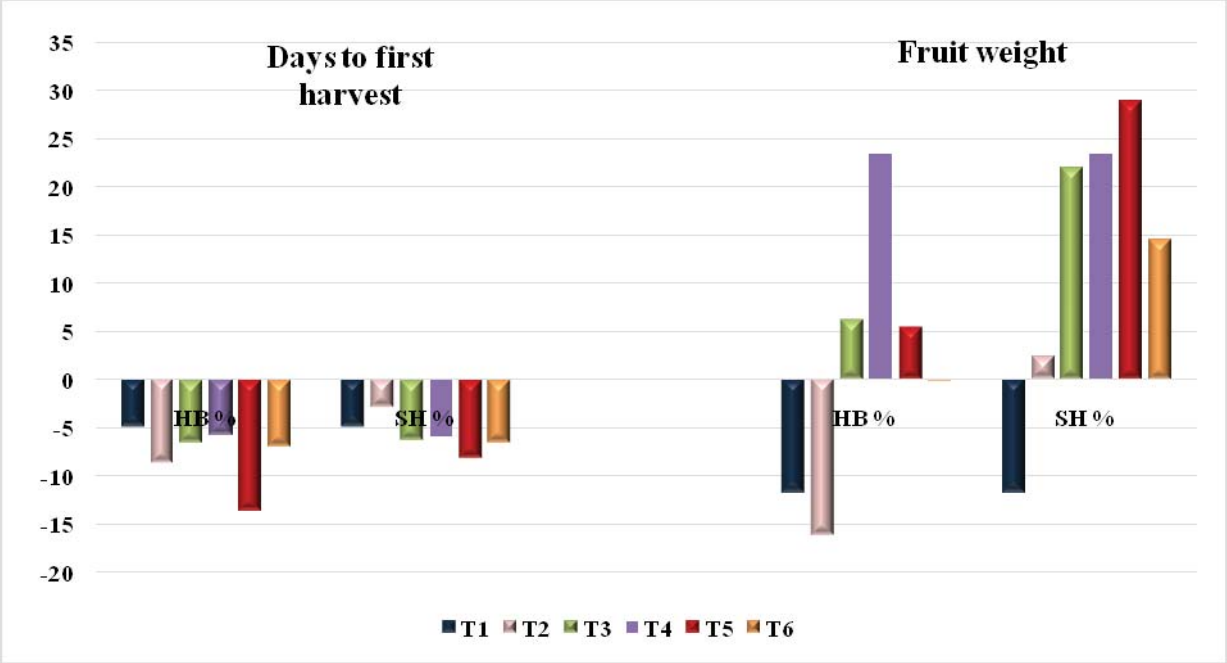

Fig. 1. Heterobeltiosis (HB %) and standard heterosis (SH %) percentage for days to first harvest and fruit weight

(T1- Preethi × KAU-MCGy-101, T2- Priyanka × KAU-MCGy-101, T3- MC 133 × KAU-MCGy-101, T4- KAU-MCGy-101 × Preethi, T5- KAU-MCGy-101 × Priyanka, T6-KAU-MCGy-101 × MC 133)

Table 2. Heterosis for different characters in F_1 hybrids of bitter gourd

	Days to first female flower opening		Node bearing first female flower		Sex ratio (Male:Female)		Number of seeds		Number of harvests		Fruit length (cm)		Fruit girth (cm)	
	HB %	SH %	HB %	SH %	HB%	SH %	HB %	SH %	HB %	SH %	HB %	SH %	HB%	SH %
Preethi × KAU-MCGy-101	-11.18**	-11.18**	-29.73**	-29.73**	-53.13**	-53.13**	-29.13**	-29.13**	15.25	83.78**	-12.16	-12.16	-0.31	-0.31
Priyanka × KAU-MCGy-101	-5.77	-8.70	-23.98**	-27.27**	-52.47**	-44.25**	-42.66**	-24.92**	-5.08	51.35**	-22.72**	-5.53	-13.37**	-7.04**
MC 133 × KAU-MCGy-101	-12.15**	-12.42**	-21.35**	-24.32**	-37.69**	-55.52**	-0.98	51.05**	-8.47	45.95**	-5.73	6.96	7.94**	-3.73
KAU-MCGy-101 × Preethi	-10.87**	-10.87**	-17.84**	-17.84**	-47.95**	-47.95**	-1.80	-1.80	22.03**	94.59**	21.27**	21.29**	-6.19**	-6.19**
KAU-MCGy-101 × Priyanka	-16.03**	-18.63**	-34.46**	-37.30**	-58.56**	-51.40**	13.07**	48.05**	15.25	83.78**	-4.91	16.23**	-7.74**	-1.00
KAU-MCGy-101 × MC 133	-10.90**	-11.18**	-19.66**	-22.70**	-24.27	-45.94**	1.77	55.26**	5.08	67.57**	-6.07	6.57	3.15	-8.00**

**Significant at 5% level, Fisher's t test.

the number of fruits per plant varied from11.20 to 73.44 per cent over standard variety (Fig. 2). Considering the number of fruits per plant, the most promising hybrids were KAU-MCGy-101 × Preethi, KAU-MCGy-101 × Priyanka, and KAU-MCGy-101 × MC 133. The present study observed highly significant heterosis for yield for all the hybrids. The highest (22.88 t/ha) and lowest yield (16.63 t/ha) were recorded in the hybrid KAU-MCGy-101 × Priyanka and Priyanka × KAU-MCGy-101, respectively. Heterobeltiosis for yield was ranged from 14.72 to 57.85 per cent and the standard heterosis ranged from 37.59 to 89.31 per cent (Fig. 2). Significant heterosis for yield is the result of the interaction of simultaneous increase in the expression of heterosis for yield attributing traits (Grafius, 1959). Three cross combinations *viz*., KAU-MCGy-101 × Preethi, KAU-MCGy-101 × Priyanka, and KAU-MCGy-101 × MC 133 with very high heterosis for yield over better parent as well as over standard variety may be considered outstanding for exploitation through heterosis breeding (Fig. 3). The mean values of earliness and yield attributing traits of these hybrids were desirably higher than those of the parents (Fig. 4).

Utilization of gynoecious lines as female parent ensures better success in heterosis breeding of bitter gourd. It is conspicuous that hybrids developed with gynoecious parent were characterized with high heterosis for earliness attributing traits like days to first female flower opening, node bearing first female flower, and days to first harvest over the monoecious parents and standard variety. The traits such as lower sex ratio and a greater number of fruits per plant contributed to the realization of higher yield in these hybrids.The

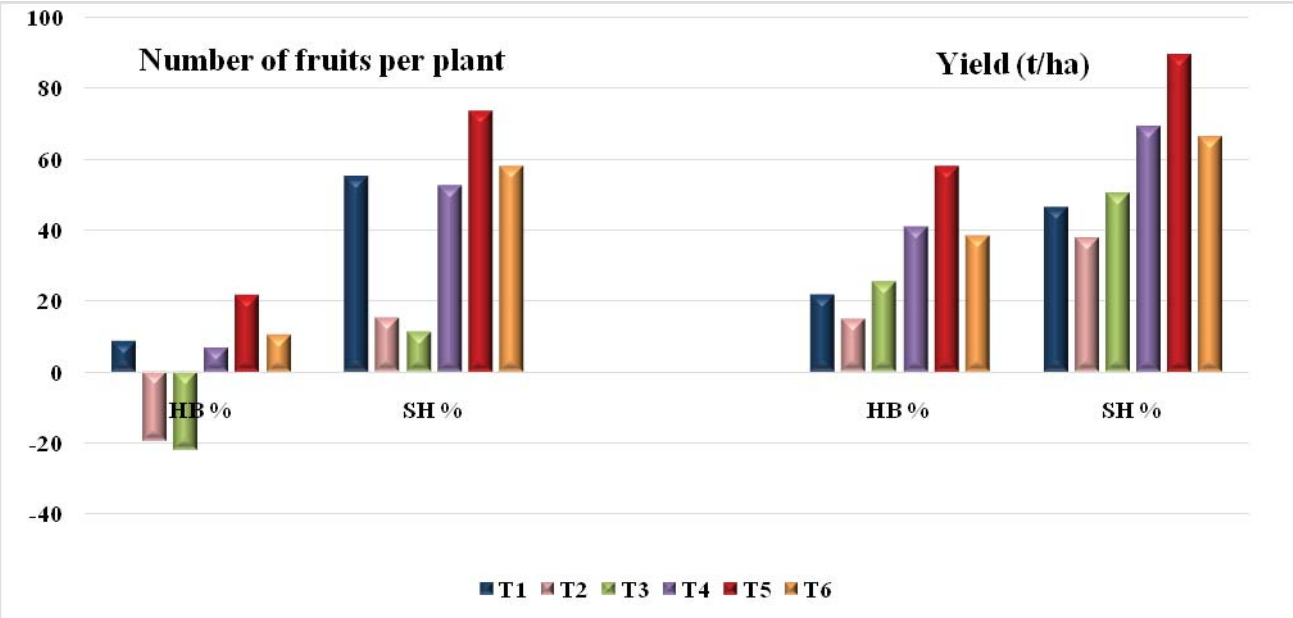

Fig. 2. Heterobeltiosis (HB %) and standard heterosis (SH %) percentage for number of fruits per plant and yield (t/ha) (T1- Preethi × KAU-MCGy-101, T2- Priyanka × KAU-MCGy-101, T3- MC 133 × KAU-MCGy-101, T4- KAU-MCGy-101 × Preethi, T5- KAU-MCGy-101 × Priyanka, T6-KAU-MCGy-101 × MC 133)

Fig. 3. High yielding hybrids KAU-MCGy-101 X Preethi (A), KAU-MCGy-101 × Priyanka (B) and KAU-MCGy-101 × MC 133 (C)

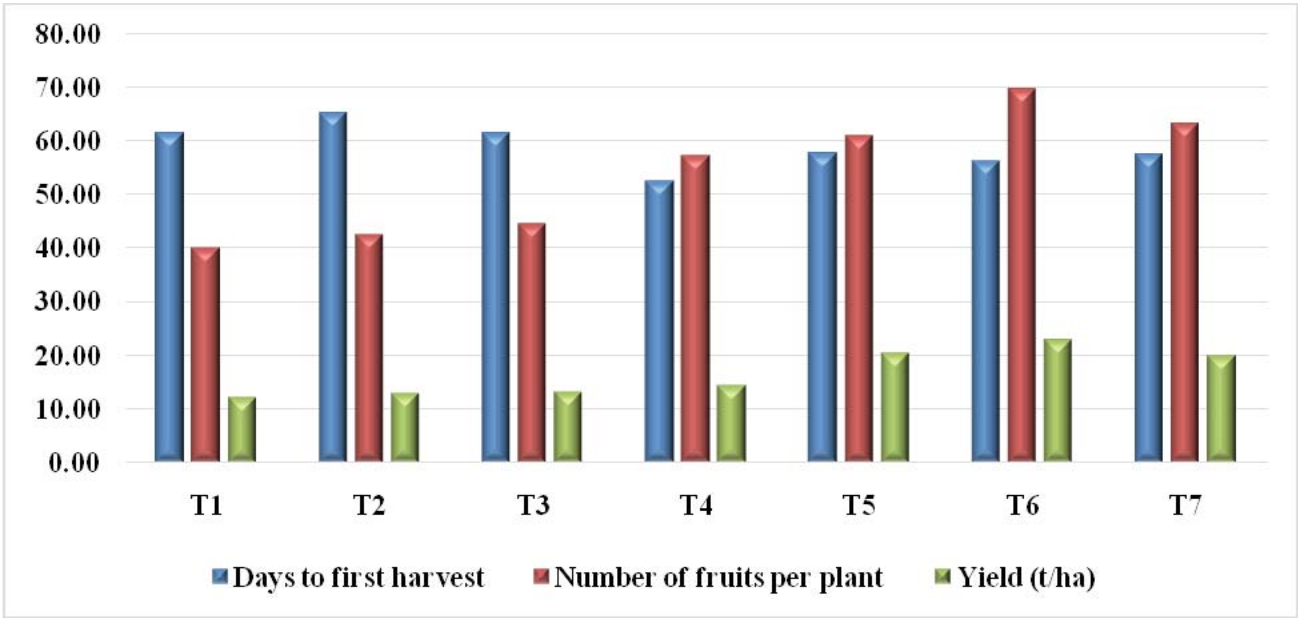

Fig. 4. Comparison between earliness and yield contributing traits in parents and superior hybrids.
T1-Preethi, T2-Priyanka, T3-MC133, T4- KAU-MCGy-101 (Parents), T5- KAU-MCGy-101 × Preethi, T6- KAU-MCGy-101 × Priyanka, T7- KAU-MCGy-101 × MC 133 (Superior hybrids)

superiority of hybrids having gynoecious line as a maternal parent is prominent in the present study. A similar trend was inferred from earlier authors, when a different gynoecious line was employed in the heterosis breeding program (Behera *et al*., 2009; Dey *et al*., 2012; Alhariri *et al*., 2018). Hence the gynoecious line, KAU-MCGy-101 used in this study is proven as a promising parent in hybridization to develop early and high yielding hybrids in bitter gourd.

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RESEARCH ARTICLE

Morphological and Anatomical Characterization of Dewlap in Polycross Progenies of *Saccharum robustum* Brandes & Jesw. Ex Grassl.

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Interspecific hybridization is the backbone of sugarcane improvement and the most widely used species have been *Saccharum officinarum* and *S. spontaneum.* Contribution of the other wild species *S. robustum* to the modern sugarcane varieties has been negligible due to the poor agronomic and quality traits coupled with susceptibility to diseases like mosaic and smut. The utilization of red-fleshed *S. robustum* gained a certain level of recognition due to its high polyphenol and anti-oxidant content with potential health benefits associated with it. The current study was undertaken to ascertain the morphological and anatomical variability in dewlap within a set of 20 progenies of red-fleshed *S. robustum.* The red-fleshed *S. robustum* have dark purple coloured dewlap which was inherited in their progenies with a little variation to its colour. However, the progenies showed wide variation in dewlap shape, size, cell composition and arrangements. The organization of vascular bundles, distribution of silica cells, cork cells, and hairs varied between the progenies. The correlation of anatomical traits with yield and quality traits showed that abaxial hairiness on dewlap was positively correlated with pol% but long cells on epidermis were negatively correlated with cane thickness, single cane weight and cane length. This paper discusses the variation for morphological and anatomical traits of dewlap and its usefulness in the crop improvement programsof sugarcane.

Key Words: Anatomy, Dewlap, Characterization, Sugarcane, Wild species

Introduction

Leaf blade and leaf sheath are two characteristics distinct parts of a sugarcane leaf. The blade and the sheath are separated by leaf joints. The leaf joint has two wedge-shaped structures known as a dewlap which provide mobility for leaf orientation. Predominantly there are three basic shapes of dewlap comprising rectangular, deltoid and ligular. The well-defined, easily observable and stable characteristics of the dewlap have enabled a range of systematic studies in sugarcane (Artschwager, 1951). However, the structure and inheritance of dewlap have not been researched so far. *Saccharum robustum* Brandes and Jeswiet ex Grassl, a wild species, is known to have exhibiting wide adaptability to various abiotic stresses, (drought and salinity) and a few clones for biotic stresses (Vasantha *et al.,* 2017, Viswanathan *et al*., 2017). Rakkiyappan *et al.,* (2012) demonstrated high antioxidants content in the red-fleshed *S. robustum,* which enhanced its further utilization in breeding programmes. The current study was undertaken to investigate the morphological and anatomical variability in dewlap within a set of inter-specific progenies of *Saccharum.* The study further sought to establish the anatomical difference between the various parts of the leaf *ie.,* leaf lamina, midrib, dewlap and leaf sheath.

Materials and Methods

The present investigation was carried out at ICAR-Sugarcane Breeding Institute Research Centre, Kannur (11º53'2708.7"N, 75º22'2728.4"E). Twenty progenies from two polycrosses of red-fleshed *S. robustum* clones (NG 77-84 and NG 77-76) acquired through the pollen grains from 16 different clones of *S. officinarum* (Chandran *et al.,* 2020) were used as the experimental material. These progenies were planted in a plot size of 10 ft × 2 rows with a spacing of 90 cm between the rows and 10 cm between the plants in three replications. Data were recorded on agro-morphological and quality traits at 11th month after planting. Both the colour and shape of the dewlap were recorded on first fully expanded leaf and an RHS (Royal Horticultural Society) colour chart was used for recording colour. For measuring the surface area, the dewlap region of the fully opened leaves (N+1) from actively growing sugarcane (7 month old

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crop) were used. The leaf was sampled and two pairs of dewlap was dissected out by cutting through the visible margin using a sharp razor blade from two different shoots of same age. The excised dewlap was placed on a graph paper and area calculated by counting the squares occupied by the dewlap. Hair groups 51, 51a, 52, 53, 55 58 and 58a and 65 (Artschwager,1951) were observed and scored "1" for presence and "0" for absence. For the morphological and anatomical description, the terminology and descriptor states were adopted from Artschwager (1951). The epidermis was separated by digesting tissue with nitric acid with a pinch of potassium chlorate. For cross-sectional anatomy of leaf parts, free hand sections were obtained and stained with safranine and mounted in 1% glycerin. The anatomical structures were observed under a compound microscope (Zeiss Primostar, Carl Ziessmicroscopy Pvt. Ltd). Measurements of anatomical traits were taken at 20X magnification using ocular micrometer and the data on epidermal anatomy and yield and quality traits were analyzed using the statistical software package SPSS 16.

Results and Discussion

I. Morphology of Dewlap

Dewlaps are different in different varieties of sugarcane and its structure is used for taxonomic classification. In the present work colour of the dewlap was not much varied between the progenies. But the size of the dewlap and the distribution of hairs showed wide variation.

Colour

Seventeen progenies had dark purple (Colour code-183A) dewlap similar to the maternal parent, while 3 progenies were with purple (Colour code -183C) dewlap (Table 1. The commercial hybrid used for comparison had purplish green dewlap. Artschwager (1951) opined that the colour of dewlap may vary considerably between varieties and even along one stalk the color may changes as they mature. Such development was observed in our studies as well. Among the progenies studied, colour of the dewlap did not show much variation. Majority of the population (85%) had same colour to the dewlap of that of female parent and only three progenies showed less intensified colour for the dewlap showing poor segregation for this trait.

Shape

Nine different types of dewlaps were observed in progenies based on its shape (Fig. 1).

The dewlap surface is slightly ruffled and the resulting softer structure often causes older dewlaps to break and tear (Artschwager, 1951). Artschwager

Table 1. Shape and colour of the dewlap in 20 progenies and two parental clones and a control

S.No	Clone	Female parent	Shape of dewlap	RHS colour code	Colour
1	GUK 14-7	NG 77-84	Tall deltoid subcrescent	183 A	Dark Purple
2	GUK 14-16	NG 77-84	Squarish sub crescent	183 A	Dark Purple
3	GUK 14-30	NG 77-84	Ligulate subcrescent	183 A	Dark Purple
4	GUK 14-33	NG 77-84	Descending narrow deltoid crescent	183 A	Dark Purple
5	GUK 14-41	NG 77-84	Ascending medium-tall ligulate	183 A	Dark Purple
6	GUK 14-48	NG 77-84	Ascending narrow ligulate	183 A	Dark Purple
7	GUK 14-69	NG 77-84	Tall deltoid subcrescent	183 A	Dark Purple
8	GUK 14-129	NG 77-84	Ligulate subcrescent	183 A	Dark Purple
9	GUK 14-130	NG 77-84	Equilateral deltoid	183 A	Dark Purple
10	GUK 14-675	NG 77-84	Squarish sub crescent	183 C	Purple
11	GUK 14-722	NG 77-84	Tall and short squarish-subcrescent	183 A	Dark Purple
12	GUK 14-732	NG 77-84	Squarish sub crescent	183 A	Dark Purple
13	GUK 14-734	NG 77-84	Ascending narrow ligulate	183 A	Dark Purple
14	GUK 14-745	NG 77-84	Ascending medium-tall ligulate	183 C	Purple
15	GUK 14-754	NG 77-84	Squarish sub crescent	183 A	Dark Purple
16	GUK 14-755	NG 77-84	Ascending medium-tall ligulate	183 A	Dark Purple
17	GUK 14-804	NG 77-76	Squarish sub crescent	183 A	Dark Purple
18	GUK 14-829	NG 77-76	Tall and short deltoid-crescent	183 C	purple
19	GUK 14-836	NG 77-76	Tall and short deltoid-crescent	183 A	Dark Purple
20	GUK 14-864	NG 77-76	Tall and short deltoid-crescent	183 A	Dark Purple
21	NG 77-76	fp	Ligulate subcrescent	183 A	Dark Purple
22	NG 77-84	fp	Ligulate subcrescent	183 A	Dark Purple
23	Co 86032	Control	Equilateral deltoid	137C	Purple green

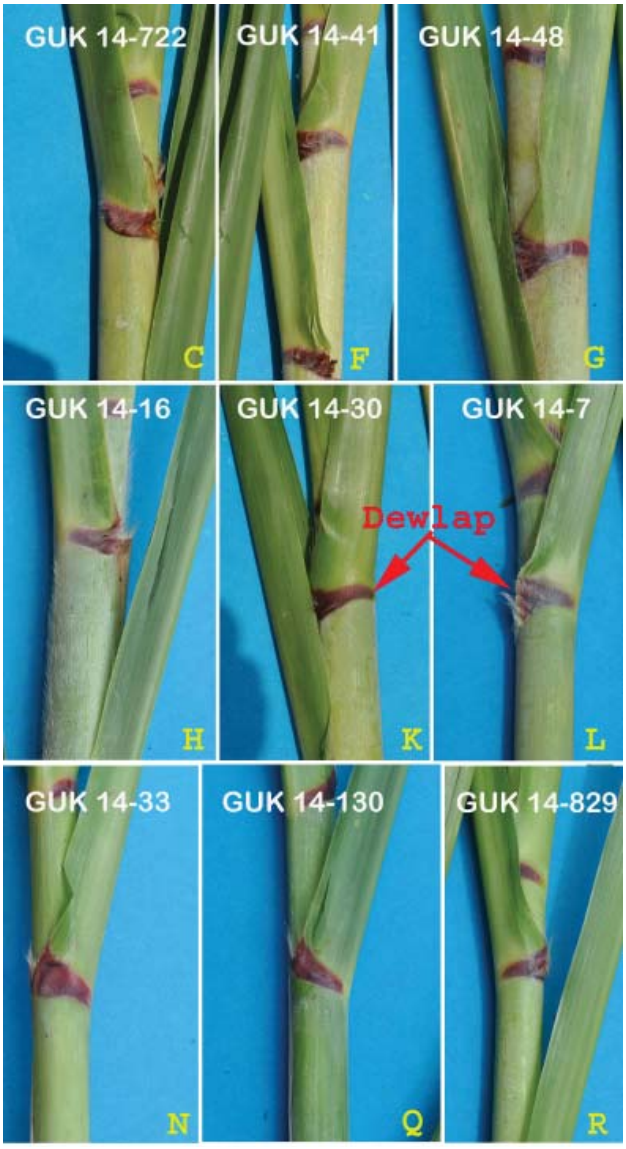

Fig. 1. Shapes of Dewlap
(C = Tall and short squarish-subcrescent, F = Ascending medium-tall ligulate, G = Ascending narrow ligulate, H = Squarish-subcrescent, K= Ligulate-subcrescent, L = Tall deltoid-subcrescent, N = Descending narrow deltoid-crescent, Q = Equilateral deltoid, R = Tall and short deltoid-crescent)

(1940) and Glyn (2004) recognized three main types of dewlaps. The rectangular or squarish type, the deltoid or triangular type, and the ligular type. All the three main types were observed in the progenies studied (Table 1). In addition, there were many intermediary forms. Nine types of dewlap were observed of the 20 types described in sugarcane by earlier workers. The result indicated that good segregation occurred for dewlap shape and other anatomical characters.

Size of Dewlap

In the check variety Co 86032 the dewlap area was 60.5 ± 5.3 mm² (Table 1). High variation was observed for surface area of progenies ranging from 25.8 ± 2.5 mm^2 (GUK 14-864) to 233 ± 8.8 mm^2 (GUK 14-734). The area of dewlaps in the female parents was 108.5 ± 10.4 mm² and 110.5 ± 10.3 mm² respectively for NG 77-76 and NG 77-84

Pubescence on Dewlap

The occurrence/distribution of hairs was on both adaxial and abaxial side of the dewlap.

Abaxial surface (Outer surface): Two types of hairs were present at abaxial surface of the dewlap viz., 58 and 58a (Table 2). 58 groups of hairs are short, felt like and often hidden under a layer of wax.

Most of the progenies had the hair group 58, except GUK 14-836 and GUK 14-864. 50% of the progenies had the hair group 58a. 10 progenies had both 58 and 58a hair groups, and ten had either 58 or 58a hair groups. There was no clones are free of both hair groups. The female parent NG 77-76 and the cultivated variety Co 86032 had both the hair groups 58 and 58a. Whereas, the female parent NG 77-84 had only 58 hair group.

Adaxial surface (Inner surface): The adaxial surface of dewlap had short felt like hairs, which is covered by long silky lashes towards the outer edge. The margin of the leaf just above the dewlap had shorter hairs that change into spine in the higher parts. Inner hairs are mainly five groups viz., 51, 52, 55, 63 and 65. The hair group 52 were short hairs usually cover the entire surface of dewlap.

Hair groups 51 and 51a were present in all 23 genotypes. Hair groups 52 was present in almost all genotypes except in female parent NG 77-84, and in the progenies GUK 14-129, GUK 14-745, GUK 14-754, and GUK 14-864. Hair group 65 present in five progenies GUK 14-16, GUK 14-30, GUK 14-41, GUK 14-48, GUK 14-755 and in the female parent, NG 77-84. In twelve progenies only 51, 51a, and 52 hair groups were present. Hair group 53 was observed only in two progenies (GUK 14-41 and GUK 14-48) and hair group 65 in three progenies (GUK 14-16, GUK 14-30 and GUK 14-755). There was no progenies with all the five group of hairs, but three main groups were present in five of them, viz., GUK 14-16, GUK 14-30, GUK 14-41, GUK 14-48, and GUK 14-755. Hair group

Table 2. Surface area and anatomical features on adaxial and abaxial epidermis of dewlap

S.No	Clone	Surface area (mm^2)	Adaxial surface				Abaxial surface				
			NLC	No. of cork cell	S-deposit	Hairiness	NLC	No.of silica cell	No.of Cork cell	S-deposit	Hairiness
1	GUK 14-7	162.0±11.3	400.0±9.0	0.0	1	3	79.7±2.9	32.0±5.1	26.0±6.2	3	1
2	GUK 14-16	227.5±9.4	325.0±6.2	0.0	0	0	109.0±8.5	47.7±7.0	24.0±4.0	2	1
3	GUK 14-30	172.3±6.1	134.7±9.0	0.0	1	3	80.0±7.9	55.7±5.2	49.3±4.6	0	0
4	GUK 14-33	177.5±6.7	227.0±9.4	0.0	1	3	123.0±5.0	11.3±2.5	100.0±9.9	0	2
5	GUK 14-41	135.5±8.9	271.7±5.0	0.0	1	3	136.0±3.3	118.3±9.8	150.7±9.7	0	1
6	GUK 14-48	106.2±10.4	97.0±5.0	0.0	1	3	127.3±5.6	105.7±4.5	93.0±2.4	0	2
7	GUK 14-69	160.8±11.1	80.0±2.0	0.0	1	3	115.0±5.0	91.7±3.1	70.7±4.9	1	3
8	GUK 14-129	164.7±11.4	218.0±2.0	0.0	0	3	232.7±4.1	4.0±0.3	30.7±6.6	2	1
9	GUK 14-130	115.7±10.1	139.7±7.8	5±0.5	1	3	108.7±2.5	107.0±3.7	44.0±5.9	2	1
10	GUK 14-675	88.3±10.0	179.0±7.0	0.0	1	2	92.5±7.5	75.7±6.6	70.7±8.4	1	2
11	GUK 14-722	63.8±4.9	195.0±5.0	0.0	1	3	60.7±5.2	24.0±3.7	4.0±1.6	1	3
12	GUK 14-732	76.3±5.6	66.0±2.0	0.0	3	3	53.0±6.2	14.7±3.4	20.7±4.1	1	2
13	GUK 14-734	233.0±8.8	226.7±5.0	0.0	1	1	67.0±5.0	61.0±8.8	16.3±7.6	3	3
14	GUK 14-745	152.0±8.3	230.0±7.5	18.0±2.5	1	1	135.3±5.2	62.7±5.0	76.3±2.6	0	1
15	GUK 14-754	115.8±6.5	248.5±6.5	0.0	1	2	118.0±5.9	80.7±0.9	38.0±9.1	1	3
16	GUK 14-755	175.8±8.8	188.0±8.0	0.0	1	2	118.7±8.2	56.0±6.7	42.3±4.8	2	1
17	GUK 14-804	89.8±9.3	208.0±8.0	5.5±1.0	1	3	140.0±5.0	52.0±4.9	78.0±5.9	2	1
18	GUK 14-829	76.8±6.2	198.7±7.7	12.5±3.0	1	2	151.3±8.4	138.7±9.3	90.7±8.1	1	1
19	GUK 14-836	29.0±2.9	265.3±7.5	0.0	1	2	117.5±2.5	71.0±2.9	64.3±5.4	1	1
20	GUK 14-864	25.8±2.5	262.0±9.9	0.0	1	2	154.7±6.2	108.0±8.6	37.7±6.1	1	0
21	NG 77-76	108.5±10.4	200.0.±8.2	0.0	1	3	150.0±5.0	146.7±4.8	72.0±2.8	3	0
22	NG 77-84	110.5±10.3	169.3±7.4	15.0±3.0	1	3	64.5±0.5	57.0±4.6	72.0±8.6	1	2
23	Co 86032	60.5±5.3	106.0±0.5	0.0	1	3	36.0±7.1	29.0±3.6	26.3±8.5	1	3

NLC= Number of long cells; S-deposit= Silica deposit (Graded 0 to 3 Scale) ; Hairiness=Graded 0 to 3 scale

51 and 51a were present in five progenies viz., GUK 14-129, GUK 14-745, GUK 14-754,and GUK 14-864 and in the cultivated variety Co 86032 but other hair groups were totally absent. Between the female parents difference was observed in the hair group distribution, in NG 77-76, hair groups 51, 51a and 52 were present, while in NG 77-84 hair groups are 51, 51a, and 65 were present.

The pubescence on dewlap of sugarcane was described by Artschwager (1951). Hair group 51,52,55,63 and 65 predominately occurs at inner side (adaxial) in sugarcane. All these groups were observed in the progenies. Hair group 52 that present mainly at inner dewlap surface which was sparse to dense, 51 and 51a are short hairs to long hairs occurs marginally or occasionally entire. Another hair group 65 was found in juxtaposition with the ligule which forms a file of long or medium long hairs at the base of the ligule extending between leaf edge and midrib.

The dewlaps are devoid of clear venation as generally observed in sheath and blade, the dorsal hairs appear uniformly scattered but rarely found distributed in a banded pattern. When there is only sparse distribution they are mainly confined to the marginal zone, but rarely they are dense near the midrib. The hairs on outer surface are short and appressed but rarely more prominent. When semi-long hairs of group 58 are observed on the outer dewlap surface, there will be more pronounced, and long hairs on the inner surface as well (Artschwager, 1951). The second type of hair group is 58a, which are long and reported for the first time by Artschwager (1948) in clones of *S. spontaneum* and *S. robustum* and subsequently among varieties of noble canes. These hair group was also available in 12 out of 20 progenies an in one of the female parents in the present study. The hairs are long and mostly marginal and because of its marginal implantation, the genotype with this hair group had the best advantage protecting the growing point (Artschwager, 1951).

Hair group 51 are the longest and most conspicuous and over up the inner surface of the dewlap. The length of it gradually decreases towards the midrib region. Hair group 52 was also described by Artschwager (1951) for the first time, which are medium long and starting from

the base to higher up in the dewlap. All the progenies, the parents and the check varieties had these hair group indicating its universal presence in most of the genotypes. Hair group 65 forms a single row of hairs not growing the above the ligule height but extended between leaf edges. The hair group 55 was found behind the midrib but rarely across the midrib (Artschwager, 1951). Our studies showed that the hair group65 was present only in three progenies and the hair group 55 only in two progenies indicating their rare occurrence.

II. Leaf Anatomy

The anatomical features of lamina, midrib, dewlap and leaf sheath showed considerable difference between them, though they are part of same organ, the leaf. The intensity of trichomes also varies between lamina and leaf sheath and dewlap.

CS of lamina

In lamina, the epidermis was not continuous, but interrupted by stomata. The distance between the vascular bundle was relatively smaller than leaf sheath and lamina (Fig. 2). The vascular bundles are of three kinds, they are large, medium, and small as reported earlier (Joarder *et al*., 2010). The small bundles are situated near the lower epidermis, while the large and medium ones were found in the center of the leaf blade. The small and medium sized bundles occur between the large bundles and alternate with one another. Sugarcane is a C4 plant which follows the Kranz anatomy having photosynthetically active chlorenchymatous bundle sheath. The vascular bundle consists of phloem and xylem. The xylem of the large bundle is fully developed and consists of two large meta xylem vessels and a number of small elements. In the smaller bundles the xylem is greatly reduced in size.

In the midrib region thick layer of parenchyma tissue was found between the fibro-vascular bundles and the upper epidermis; hence, all vascular bundles in this part of the blade were restricted to the lower side of bundles, is reinforced by a thick solid layer of sclerenchyma. The

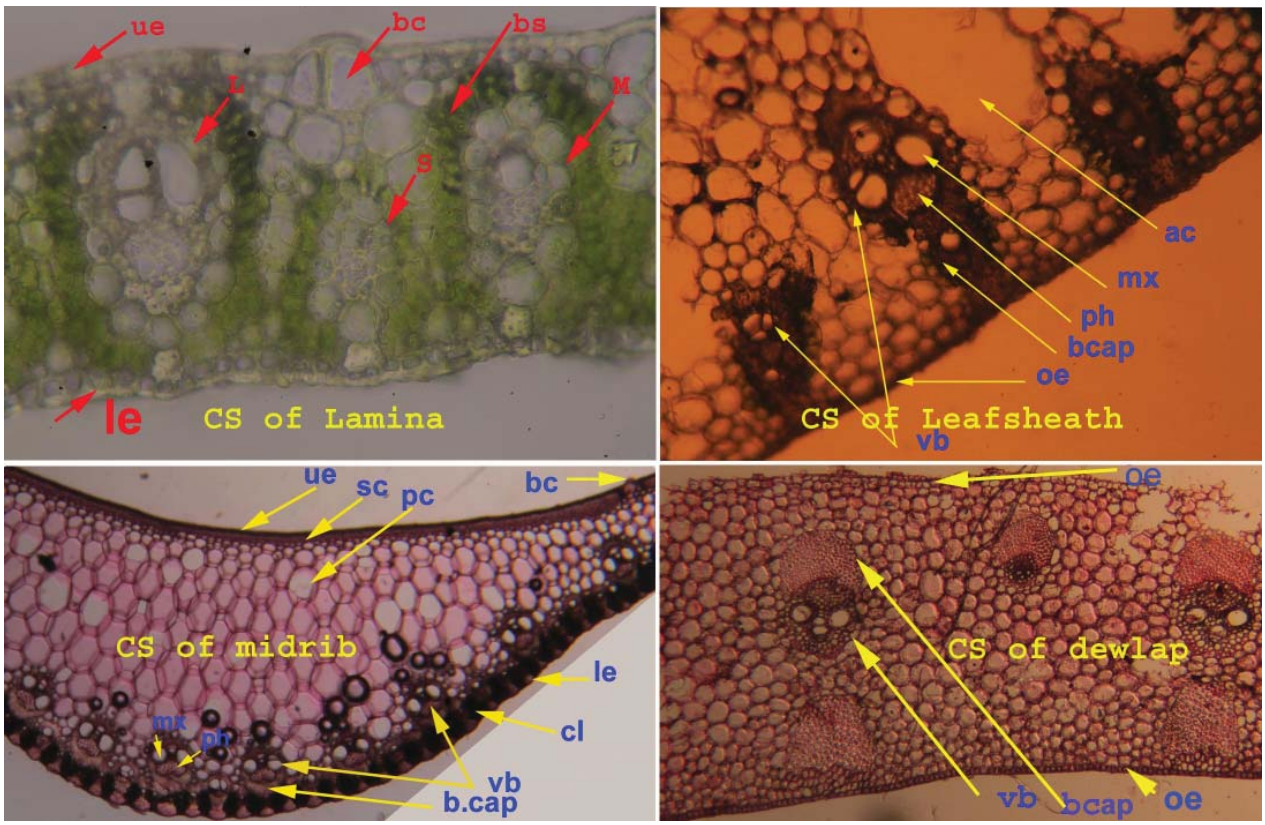

Fig. 2. CS of leaf at lamina, dewlap, mid rib and leaf sheath
(ue = Upper epidermis, L = Large vascular bundle, M = Medium size vascular bundle, S = Small vascular bundle, bc = Bundle cap, bs = Bundle sheath, ac = Aerenchyma, mx = Metaxylem, ph= Phloem, oe=Outer epidermis, vb= Vascular bundle, e=Epidermis, sc=Sclerenchyma, pc = Parenchyma, cl = Chlorenchyma, le = Lower epidermis)

layer of parenchyma which is found between the bundles and the upper epidermis does not contain chlorophyll and the white color of the upper side of the midrib was attributed to it, but greener at the lower side as a result of the presence of chlorophyll-bearing cell adjacent to the lower epidermis.

CS of Leaf Sheath

A cross section at about half way the length of the sheath shows radial rows of fibro vascular bundles. Unlike the mid rib the largest bundles are located at about equal distance both from the upper and lower epidermis. Towards the outer epidermis (morphologically the lower surface) the bundles gradually became smaller. The number of vascular bundles in a one radial row varies from 2 to 4. In the parenchymatous tissues, in which the bundles are embedded, showed large cavities alternating with the radial rows of bundles. In some cases several vascular bundles are often found united into one large composite bundle in such cases the composite bundle is surrounded by a sclerenchymatous layer of cells which extends from the radial vascular bundle row towards the upper epidermis.

CS of Dewlap

As in other portion of the leaf the epidermis is single layered but epidermal cells are not interrupted by stomata. Vascular bundle of leaf blade, leaf sheath, and dewlap are different in their arrangements. In leaf sheath a large bundle constitute of two single bundles, where towards the dewlap one of the bundle is gradually disappears. In dewlap mature vascular bundle have bundle cap which is collenchymatous. Vascular bundles are of three types large, medium, and small which is alternates with one another. And the parenchyma cells, seen in between vascular bundles. Vascular bundles are seen towards the abaxial (outer) surface of epidermis. Bundle cap present only opposite to the large vascular bundle. Large vascular bundles consist of large xylem vessels. In dewlap chlorophyll pigments present in epidermal layer, bundle sheath, and in the upper layer parenchyma cells but often masked by the anthocyanin pigment which give dark purple color to the dewlap region in the progenies.

In sugarcane most of anatomical the features of taxonomic value are associated with the leaf blade (Metcalfe 1960) and hence detailed anatomical studies have been reported initially in leaf blade and subsequently the studies were extended to understand the detailed vasculature of the leaf and its parts (Artschwager 1925, 1940; Van Dillewijn, 1952, Martin, 1961). The present study on cross section of lamina, midrib, dewlap and sheath conforms with the previous reports on the differences between the four parts of the leaf on vascular tissues, mechanical tissues and in their arrangements. Dewlap was characterized by the absence of stomata and collenchymatous bundle sheath bundle caps are seen only for larger vascular bundles. Cobert and Evert (1982) also reported the collenchymatous bundle cap in dewlap of sugarcane. The collenchymatous bundle caps substantiate the flexibility (Artschwager 1951) offered to the dewlap for orientation of the lamina during the early stages of leaf development. The dewlap was also reported to be cracked, disfigured or withered while leaf attaining maturity and the same was observed in the present studies.

At midrib region the size of the bundles gradually decreases from the inner band (next to the inner epidermis) to the outer one (Van Dallewijn, 1952). Isaac (1939) observed that varieties which are resistant to the top-borer, *Scirpophaga nivella* F., are characterized by strong and hard midrib, whereas those which are susceptible to top-borer possessed weak midribs. *S. robustum* in general and more particularly the red fleshed *S. robustum* belongs to the forma *sanguine* had broad leaf and thick midrib and so as the progenies. But further studies may be required to understand the extent of the sclerenchymatous sub-epiderma layer in the midrib region and the top borer screening in these progenies to confirm the results. Rao (1947) by studying the anatomy of some fifty varieties of sugarcane was able to confirm the observation of Isaac (1939). Cobert and Evert (1982) found that the leaf sheath has a lesser number of vascular strands compared to lamina as a result of the fusion of vascular strands at the leaf joint. Towards the dewlap, the sheath is narrower and has more thickness and the air cavities gradually reduce and finally disappear. There are single vascular bundles that are generally larger and arranged alternately with the composite vascular bundles.The cross-section of the lamina and leaf sheath showed difference in the number of vascular bundles and fusion of more than one vasculature in leaf sheath region. However, earlier studies (Colbert and Evert,1982) showed that, the cross-sectional areas are rather increasing to both sieve tubes and tracheal elements while it continues to leaf sheath. Hence, the reduction in number of the vascular bundles does not affect the translocation of water and food between the

leaf parts. The inner epidermis consists of cells of various sizes indicating the divisional or cambial activity at the dewlap region though cambial activity in mature leaves was not a regular feature of monocot leaves. This is in conformation with the earlier report on the anatomy of leaves by Moreland (1942) showing cambial activity in leaves of monocots and later by Artschwager (1951). He also observedthe development of secondary meristem as an extension of cambium from one vascular bundle to another and opined that monocotyledonous plants share similarities to dicots in these aspects.

Epidermal anatomy of the dewlap

Epidermis comprised of hexagonal cells not interrupted by stomata. In addition to the normal long cells, silica cells, various forms of silica depositions, cork cells and dried cells constitute the epidermis. Trichomes also observed on epidermal cells.

Abaxial (outer) epidermis: Dried cell is absent in abaxial (outer) surface of epidermis (Fig. 3). Stomata was generally absent but a few stomata were observed in GUK 14-130. Number of normal cells is varying in different genotypes, the highest number of cells per unit area was observed in GUK 14-129 (232.7±4.1), and the lowest in cultivated variety Co 86032 (36.0±7.1, Table 2). The female parents NG 77-76 have 150±5.0 and NG 77-84 had 64.5 ±0.5 normal cells. Silica cell was present profusely on abaxial surface. Highest number of silica cell was found in female parent NG 77-76 (146.7±7.8) and the lowest number in check variety Co 86032 (4±0.3) The female parent NG 77-84 had 57 silica cells per unit area. Cork cells were present on abaxial surface of all genotypes. The female parents NG 77-76 and NG 776-84 had 72±2.8 and 72±8.6 cork cells respectively. Highest number of cork cell (150.7±9.7) was observed in GUK 14-41 and lowest number (4±1.6) in GUK 14-722. The cultivated variety Co 86032 had 26.3±8.5 cork cells.

Silica deposition was high (scale=3) in female parent NG 77-76 and low (scale=1) in NG 77-84and cultivated variety Co 86032. Medium deposition (scale=2) of silica were found in five progenies and low (scale =1) in eight progenies. In progenies viz., GUK 14-745, GUK 14-30, GUK 14-33, GUK 14-41 and GUK 14-48 the silica deposit was completely absent (Scale=0). Apart from NG 77-76, high silica deposits were found in GUK 14-7, and GUK 14-734. Number of hairs were less on abaxial surface compared to that of adaxial surface. Hairs were completely absent on abaxial surface of GUK 14-30, GUK 14-864 and the female parent NG 77-76. In ten clones, hairs were low (scale=1), five clone has medium hairs (scale=2), and another five clones had profuse hairs (scale 3). NG 77-84 female parent had medium hairs and the cultivated variety had profuse hairs on abaxial surface of the dewlap.

Adaxial (inner) epidermis: Silica cells and stomata were completely absent on the adaxial (inner) surface (Fig. 4) however, silica depositions of various shapes were present. Low distribution (scale 1) of silica deposition was found in for most of the genotypes, except GUK 14-732 where it was very high (scale 3). Silica deposition was completely absent in GUK 14-16 and GUK 14-129. The highest number (400±9.0) of long cell was found

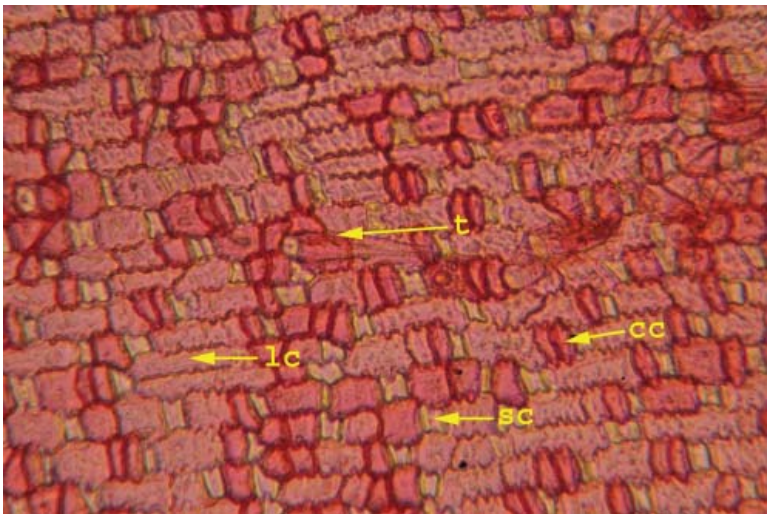

Fig. 3. Abaxial epidermis of GUK 14-48
(t= Tannin cells, lc=Long ells, cc = Cork cells, sc= Silica cells)

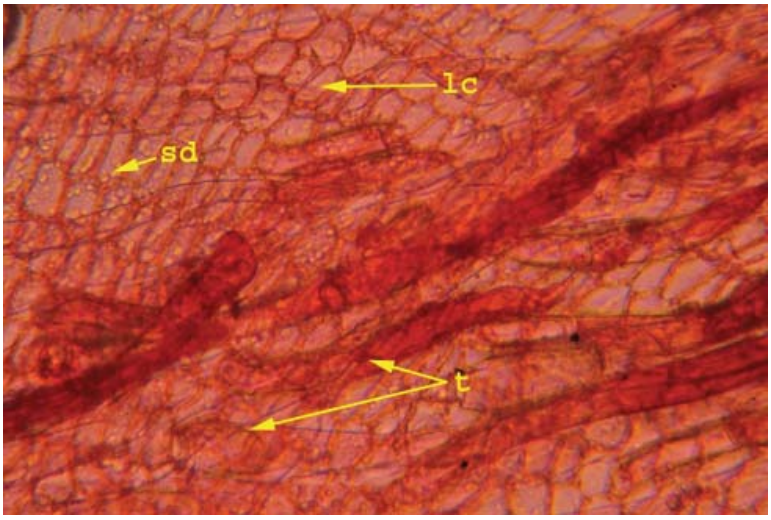

Fig. 4. Adaxial epidermis of GUK 14-48
(lc=Long cells, sd= Silica deposits, t= Trichomes)

in progeny GUK 4-7 and the lowest (66±2.0) in GUK 14-732. Long cells in female parent NG 77-76 was 200±8.2 and NG 77-84 was 169.3±7.4, and in cultivated variety Co 86032 had 106±0.5. Cork cell only present in five progenies, they are GUK 14-84, GUK 14-130, GUK 14-745, GUK 14-804, and GUK 14-829.

Distribution of hairs on the adaxial (inner surface) surface of the dewlap also showed wide variation between progenies. Hairs were totally absent in GUK 14-16 where as 11 progenies, cultivated variety (Co 86032) and both female parents had a dense distribution of hairs (scale =3, Table 2). Six progenies had profuse hairs (scale =2) and two progenies had a sparse distribution of hairs (scale =1). In addition to these cells, some of the progenies (GUK 14-16, GUK 14-41, GUK 14-130, and GUK 14-864) showed brown coloured cells which appears to be ruptured and dried cells.

The distribution of silica cells, cork cells, hairs, and other deposition vary between the genotypes. Silica cell provides mechanical stability to the tissues, it protect against fungi, insects and other herbivores. It also help to facilitate light interception, drought resistance and problems related to nutritional disorders including excess availability (Motomura *et al*., 2006). The progenies with high silica cells viz., GUK 14-41, GUK14-48, GUK 14-130. GUK 14-829, GUK 14-86 are the potential genotypes for evaluating against different stresses.

III. Correlation studies

Two-tailed correlation analysis based on Pearson coefficient was done for 18 characters. A significant positive correlation was observed for germination with number of millable canes (0.731**), HR brix at Middle of the cane (0.430*, Yield (-0.484*) but negatively correlated with cane thickness (-0.479*) and single cane weight (-0.505*). A similar positive correlation was also found between NMC and other quality traits (Table 3). This confirms the earlier result from the evaluation of a large number of progenies at the ground nursery level (Chandran *et al*., 2020) with unreplicated data. HR brix at bottom was positively correlated with HR brix at Middle (0.933**), bottom and top (0.891**) and at Brix at 11th month (0.797**) and Pol % (0.786**) at 11th month. The number of long cells on the epidermis per unit area was negatively correlated (-0.590*)with cane thickness, single cane weight (-0.448*)and cane length (-0.454*). Hairs on the adaxial surface of dewlapwere negatively correlated (0.419*) with adaxial long cells. Another interesting correlation observed was between abaxial hairiness on dewlap with pol% (0.434*). Abaxial hairiness was negatively correlated with adaxial hairs (-0.505*). Abaxial silica cell was positively correlated with abaxial cork cells (0.503*)as in most of the cases, the cork cells and silica cells were observed in pairs. The significant positive correlation of abaxial hairiness on dew lap with pol%, negative correlation with number of long cells on epidermis with cane thickness and single cane weight and cane length can be used for selecting the progenies for further crop improvement programme of interspecific crosses.

Sugarcane leaf composed of leaf blade or lamina and leaf sheath. At the junction of the lamina and leaf

Table 3. Correlation of agronomical, quality and anatomical traits of 20 progenies, female parents and check variety

	30d	NMC	Ctk	HRBB	HRBM	HRBT	Ext	Scwt	CL	Brix	Pol	Yld	ADNC	ADH	ABNC	ABS	ABC	ABH
30d	1.000																	
NMC	**0.731****	1.000																
Ctk	**-0.479***	**-0.535****	1.000															
HRBB	0.318	**0.523**	-0.272	1.000														
HRBM	**0.430***	**0.557****	-0.198	**0.933****	1.000													
HRBT	0.406	**0.517***	-0.173	**0.891****	**0.978****	1.000												
Ext	0.121	0.090	**0.475***	-0.011	0.219	0.318	1.000											
Scwt	**-0.505***	**-0.627****	**0.829****	**-0.458***	-0.396	-0.351	0.370	1.000										
CL	-0.198	-0.047	0.081	-0.159	-0.162	-0.088	0.150	0.009	1.000									
Brix	0.308	**0.420**	-0.028	0.797	**0.877****	**0.870****	0.297	-0.270	-0.052	1.000								
Pol	0.279	**0.422**	-0.031	**0.786***	**0.854****	**0.852****	0.301	-0.282	-0.059	**0.987****	1.000							
Yld	**0.484***	**0.694****	0.070	0.272	0.379	0.359	**0.528****	0.061	-0.034	0.342	0.339	1.000						
ADNC	0.403	0.369	**-0.590****	0.228	0.191	0.156	-0.203	**-0.448***	**-0.454***	-0.098	-0.077	0.109	1.000					
ADH	-0.382	-0.326	0.106	-0.233	-0.180	-0.131	-0.005	0.270	0.020	-0.123	-0.122	-0.226	**-0.419***	1.000				
ABNC	0.321	0.087	-0.242	-0.139	-0.169	-0.142	-0.120	-0.210	-0.014	-0.254	-0.290	-0.098	0.240	-0.039	1.000			
ABS	0.146	-0.141	0.069	-0.038	-0.099	-0.169	-0.177	0.064	-0.150	-0.165	-0.179	-0.119	-0.043	-0.060	0.291	1.000		
ABC	0.202	-0.033	-0.025	-0.292	-0.252	-0.247	0.027	0.163	-0.371	-0.329	-0.312	0.007	-0.026	0.210	0.359	**0.503***	1.000	
ABH	0.053	0.237	0.168	0.129	0.134	0.115	0.189	0.024	0.033	0.404	**0.434***	0.333	-0.329	0.042	**-0.505***	-0.323	-0.228	1

* significant at 5% level ** Significant at 1% level

(30d- Germination at 30 days; NMC- Number of millable canes; Ctk- Cane diameter; HRBB- HR brix at Bottom of the cane ; HRBM – HR brix at Middle of the cane; HRBT- Hr brix at top of the Cane; Ext-Juice extraction (%); Scwt- Single cane weight; CL- Cane length; Brix- Hydrometer Brix at 11^{th} month; Pol- Pol %; Yld- cane yield/plot(kg); ADNC- Adaxial normal cells; ADH- Adaxial hairs; ABNC- Abaxial normal cells; ABS-Abaxial silica cells; ABC, Abaxial cork cells; ABH- Abaxial hairs)

sheath the appendages like ligule towards the inner side and ligular process or auricle on either side and the dew lap were present. Trichomes also present at various intensities on leaf sheath, lamina and on the appendages (Artschwager, 1951). The dewlap or the leaf joint is a peculiar structure found in some the graminaceous members including sugarcane. Dewlap is located just above the ligule at the joint of leaf blade with leaf sheath. It is also referred as leaf joint/ leaf triangle. The tips or inner margins of the two blade joint hinge areas almost meet on the back surface of the midrib. The dewlaps form a hinge of theblade joint and this enablesome kind of mobility to the leaf blade and in turn, help the plant to orient the leaves. The red-fleshed *S. robustum* had a distinct dark purple coloured dewlap which is inherited to their progenies without much variation in colour that provides the advantage in identifying the interspecific hybrids. But varied for shape and size of the dewlap and for different composition of the epidermal tissues. Though the leaf sheath, dewlap and lamina constitute same organ, anatomically they are distinguishable in the size and arrangement of vascular bundles. The variation was due to its function such as for supporting provided with more bundle cap cells in leaf sheath and dewlap compared to lamina. The flexibility and strength was offered due to collenchymatous bundle cap in dewlap, sclerenchymatous bundle cap for leaf sheath and lamina and in the later with only minimal bundle cap cells. The distribution vascular bundle also varied between these regions, the vascular bundles often composite ones which are more widely placed in leaf sheath alternating with air cavities. In dewlap the vascular bundles are closer but without air cavities in between and in leaf blade it was closely arranged alternating one large vascular bundle with two smaller ones. The present study revealed the detailed structure and anatomical difference between lamina, midrib, dewlap and leaf sheath and the epidermal tissue composition of dewlap in twenty interspecific progenies developed from polycrosses on red-fleshed *S. robustum* and its usefulness in the crop improvement programme of sugarcane.

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RESEARCH ARTICLE

Machine Learning Algorithms for Protein Physicochemical Component Prediction Using Near Infrared Spectroscopy in Chickpea Germplasm

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Prediction of physicochemical components of chickpea flour using near infrared spectroscopy requires discovering exact wavelength regions that provide the most useful data before preprocessing. This study used six essential machine learning techniques to develop models for predicting proteinphysicochemical component in chickpea: Linear Regression (LR), Artificial Neural Network (ANN), Partial Least Squares Regression (PLSR), Random Forest (RF), Support Vector Regression (SVR) and Decision Tree Regression (DTR). Performance measurements such as Root Mean Square Error and Karl Pearson's Correlation Coefficient and Coefficient of Determination were used to validate the models. RF and ANN models showed significant improvement over all other models in terms of accuracy.

Key Words: Artificial Neural Network, Chickpea, Machine learning, Near infrared spectroscopy, Random Forest, Spectroscopy

Introduction

Near infrared spectroscopy (NIS) is an efficient method for identifying and analyzing many components in a sample (Acquah *et al.,* 2016) and can be an excellent predictive germplasm evaluation procedure. Combining bands of numerous hydrogen-containing groups in moisture, protein, fat, and carbohydrate, and the vibrational information in these organic molecules are used to assess the chemical composition of samples (Batten and Berardo*,* 1998). Several techniques have been developed to extract quantitative information from Near Infra-Red (NIR) spectra using wavelengths. Some of the most used calibration methods for NIR spectroscopy are Principal Component Regression (PCR), Linear Regression (LR) and Partial Least Squares Regression (PLSR). On the other hand, Non-linear algorithms such as Artificial Neural Network (ANN), Support Vector Regression (SVR), Decision Tree Regression (DTR) and Random Forest (RF) models are not as extensively used, but they may deliver superior results when the spectral data and the quantitative value of interest have a non-linear connection (Pasquini, 2003). Such supervised predictive modelling techniques use known data to develop models capable of predicting values for future events. Selecting the most effective predictive modelling technique at the start saves considerable time.

Chickpea is rich in protein and evaluating the physicochemical components of protein in the chickpea germplasm is fundamental to identifying superior genotypes and use them further in breeding programmes. Here, we report a comparative study of six essential machine learning techniques to develop models for predicting concentrations of protein physicochemical components in chickpea germplasm with an objective to identify the best model to facilitate improved use of NIS in biochemical assay of germplasm.

Materials and Methods

Plant sample and spectral data: A random set of 237 chickpea germplasm accessions were obtained from the National Genebank, ICAR-National Bureau of Plant Genetic Resources, New Delhi. Chickpea seeds were homogenized in a Foss Cyclotec mill and the flour was transferred to a circular cuvette. Spectra in the wavelength range 400-2498 nm were captured, with 2 nm spacing, using a Foss NIRS 6500 cuvette spinning model. The instrument was calibrated against white mica each time the sample was scanned. The average spectrum was

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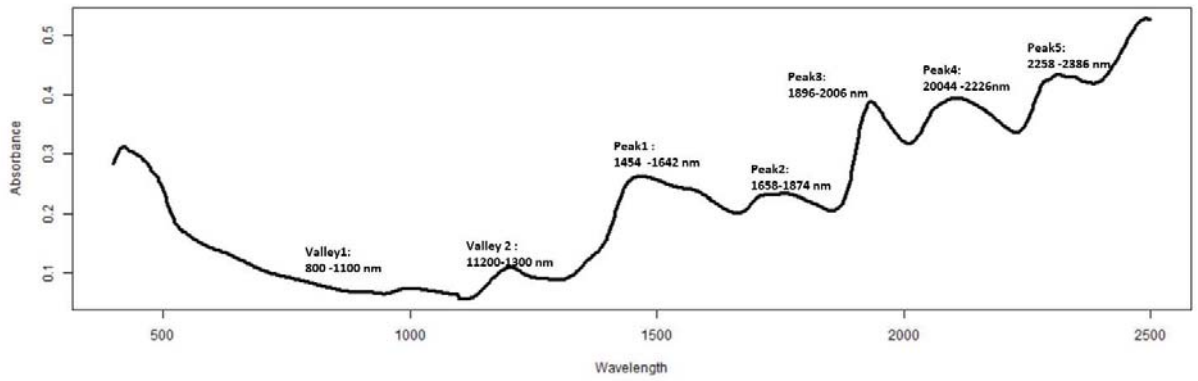

Fig. 1. Average spectrum of chickpea

recorded after scanning the material 32 times (Fig. 1). The concentration of protein physicochemical in chickpea seedsamples was measured in the chemical laboratory, which served as reference data for training and measuring the performance of prediction models.

Development of machine learning models: Machine learning techniques LR, ANN, PLSR, RF, SVR, and DTR were used to develop near infrared spectroscopy prediction models utilizing preprocessed spectra. The Comprehensive R Archive Network (CRAN) provided all model development packages. All models would be iterated 5000 times. The 237 samples were randomly separated into two groups: 75% (176 samples) as a training data set and 25% (61 samples) as a testing data set. All spectra were scaled so that the resulting model could be interpreted in terms of variance around the mean. To identify the best suited combination for all six machine learning methods, 27, 776 combinations were used. The preprocessed combination with the highest *r* and R^2 and the lowest RMSE value was chosen as the best. To evaluate the efficacy of the regression model, the RMSE, r, and R2 between measured and predicted concentration levels of protein physicochemical component in chickpea crop were calculated. The ideal model for each component was chosen based on the lowest RMSE value of prediction, highest *r*, and highest R^2 between measured and predicted values.

Table 1. Wavelength range for preprocessing

S.No.	Wavelength Range	No. of Wavelengths	File Name	Wavelength Characteristic
1	800-1100 nm	300	P6	N-H2nd overtone O-H2nd overtone C-H3rd overtone
2	1100-1300 nm	200	P7	C-H2nd overtone O-Hcombinations
3	1404-1642 nm	232	P1	C-H combinations, O-H, N-H, and 1st overtone
4	1658-1874 nm	216	P2	C-H and 1stovertone
5	1896-2006 nm	110	P3	O-H, N-H combinations
6	2004-2226 nm	222	P5	C-H, O-H, N-H combinations
7	2258-2386 nm	128	P4	C-H combinations

Results and Discussion

Different algorithms produced optimum prediction for protein with different RMSE for specific wavelength range at different *r* and R^2 values (Table 1 and Table 2). For instance, the RF algorithm produced the best prediction in a wavelength range of 1404-1642 nm with an RMSE of 0.09, *r* of 1.00, and R^2 of 0.87. On the other hand, the ANN algorithm predicted at 2258-2386

Table 2. Preprocessing methods applied on spectra with performance measures

Algorithm	RMSE	r	R^2	Moving Average[1]	Binning	Derivative	SG Polynomial	SG Window	Scatter Correction
RF	0. 09	1. 00	0. 87	0	0	0	0	0	SNV
SVR	0. 08	0. 95	0. 86	6	0	0	0	0	SNV-Detrend, MSC, SNV
ANN	0. 08	0. 94	0. 86	12	8	3	4	7	SNV-Detrend, SNV, MSC
DTR	0. 08	0. 93	0. 85	2	2	0	0	0	MSC, SNV
LR	0. 08	0. 93	0. 85	8	2	1	2	3	MSC, SNV
PLSR	0. 11	0. 84	0. 70	0	0	0	0	0	SNV-Detrend, MSC, SNV

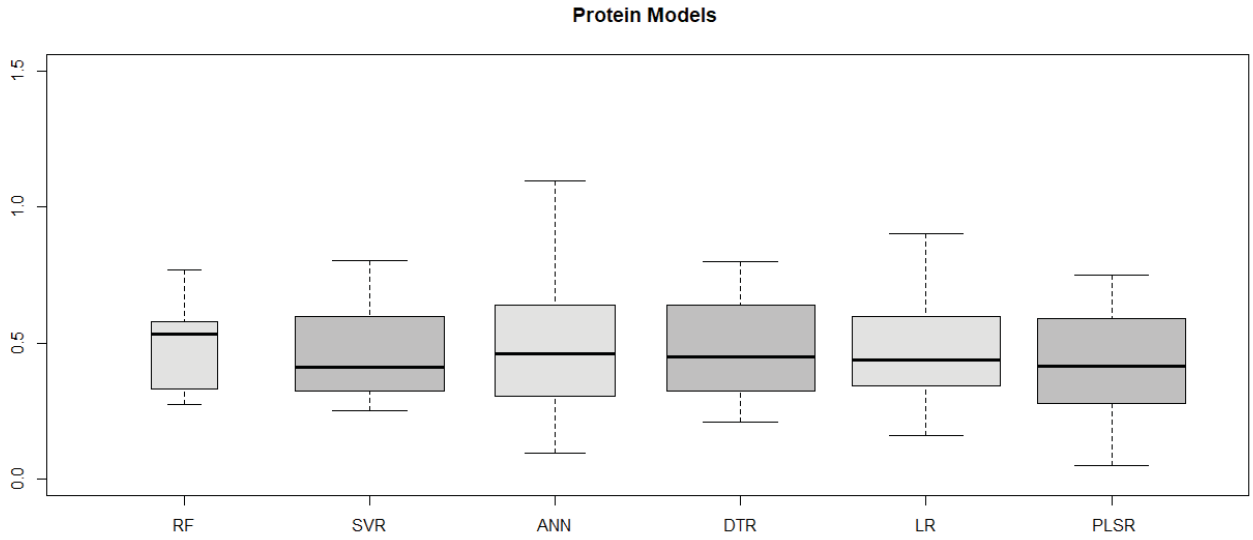

Fig. 2. Boxplots for six machine learning models for protein physicochemical component of chickpea

nm wavelength with an RMSE of 0.08, *r* of 0.95, and R^2 of 0.86.

Boxplots showed no outliers in the datasets, and the median value of all component models was observed to be close to 0.5. Compared to other models, ANN model for protein had maximum range, indicating that ANN models have maximum variability. Data variability of the DTR and SVR models were similar with practically equal coverage areas. The RF model had the least data variability with a negatively skewed median line, whereas the LR model was found to be positively skewed, whereas the PLSR model was symmetric.

Preprocessing is challenging to judge prior to model validation. All preprocessing techniques aim to minimize unmodeled variability in the data to improve the feature sought in the spectra (Rinnan *et al*., 2009). It has a linear relationship with a response variable such as a constituent. This is possible with the correct preprocessing technique, but there is always the risk of applying the wrong kind or over-processing, which will result in the loss of essential data. In the present study, experiments with 27, 776 preprocessing combinations were planned to achieve an appropriate preprocessing approach. They were put to the test, and the optimum preprocessing combination that gave the best prediction value from a model was identified.

It was discovered that RF performed best in the wavelength range 1404-1642 nm. Comparing all six models reveals that the RF models outperform all others. The RF technique is ensemble-based enhancing the model accuracy. Due to the randomness, the RF algorithm examines all variables at each node outperforms all other machine learning algorithms in processing speed and resistance to over-fitting noise. RF reduces the significant variance of a flexible model like a decision tree by combining numerous trees into one ensemble model. RF is less computationally intensive and does not require a graphics processor to complete training. A closer look at the data showed that the ANN model could also predict the physicochemical component of proteins within 500 iterations. It is worth noting that the tests were carried out under the laboratory condition and the models' dependability can only be proven once they've been applied to real-world procedures.

Conclusion

NIR, as a non-destructive technique, requires no or minimal sample preparation. Its use to find concentrations of physicochemical component provides excellent predictive methodology for germplasm evaluation. Results of the present study and application of machine learning algorithms is expected to scale up predicting physicochemical components in chickpea as well as other leguminous crops.

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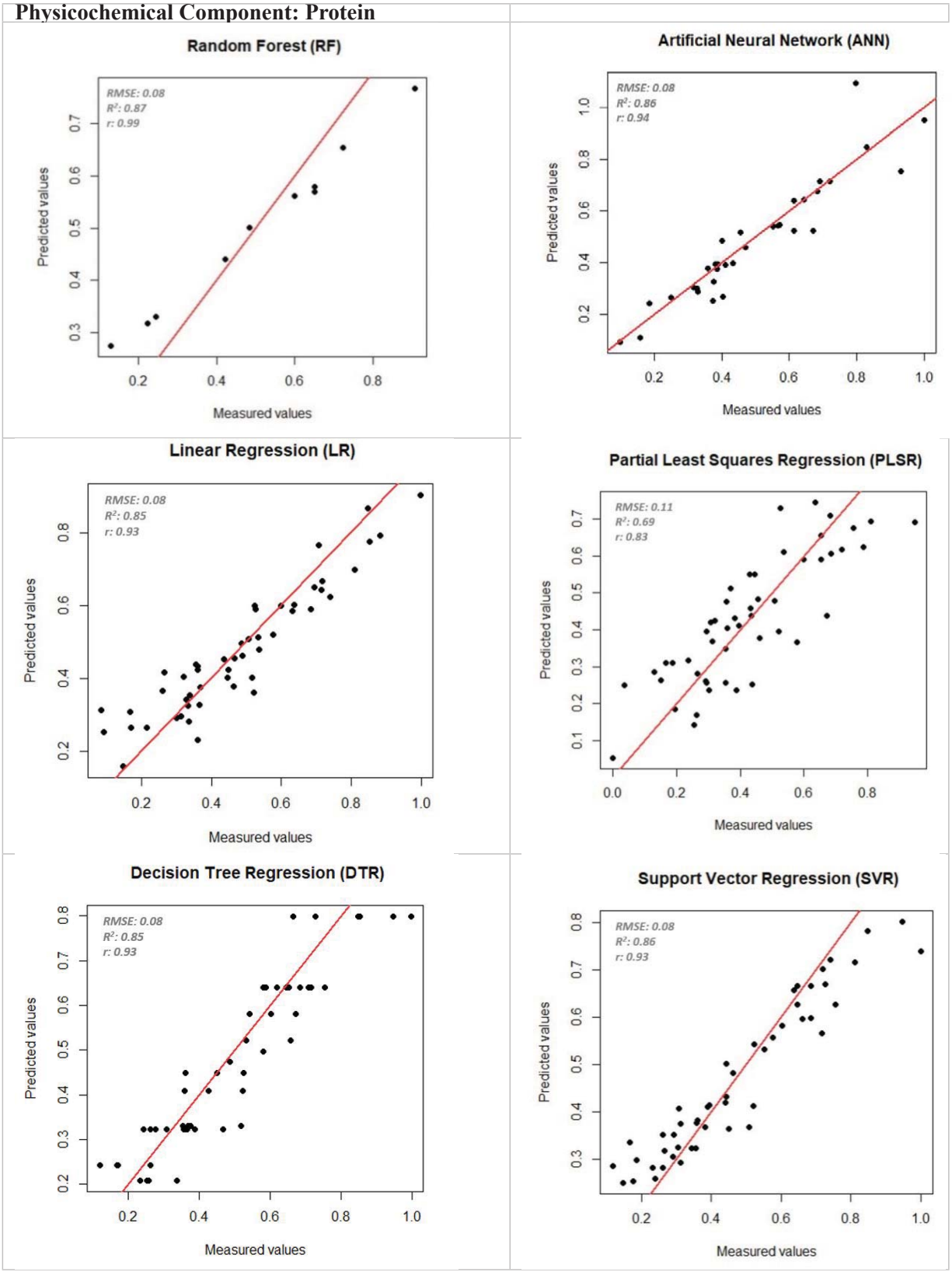

Fig. 3. Correlation between the measured and predicted value of protein physicochemical component of chickpea using all six prediction models. RMSE: root mean square error, R^2: coefficient of determination, *r*: correlation coefficient.

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RESEARCH ARTICLE

Gene Action Studies in Gynoecious Cucumber (*Cucumis sativum* L.) Lines under Mid Hill Conditions of Western Himalayas

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Genetic improvement of crop plants is a continuous process and plant breeders continuously strive for developing new varieties, which are high yielding and superior to the existing ones. The success of a breeding programme lies in the choice of appropriate parents and the breeding method followed. Therefore, the experiment was carried out involving gynoecious lines in cucumber at Palampur and Bajaura, to identify suitable parents and gather information on gene action. The line × tester analysis revealed significant differences due to lines, testers and line × tester interaction at both the locations for most of the traits, indicating appreciable diversity in the experimental material. Estimates of general combining ability effects, necessitates inclusion of lines G-3, G-1, Plp-Gy-1 and EC-5082 and the testers K-pap, Sel-75-2-10, K-90 and KL-1 for making crosses which was corroborated by the superiority of their cross combinations. The estimates of GCA and SCA variances pointed out that for majority of traits, non-additive gene action was in appreciable magnitude suggesting the exploitation of hybrid vigour in cucumber.

Key Words: Breeding, Cucumber, Gene action, Lines, Testers

Introduction

Cucumber (*Cucumis sativus* L.) is member of Cucurbitaceae family, which comprises 117 genera and 825 species in warmer parts of the world (Gopalakrishnan, 2007). Cucumber is a thermophillic and frost-susceptible crop, growing best at temperature above 20 ºC. Cucumber is thought to have originated in India (Harlan, 1975) because of the fact that *Cucumis sativus* var *hardwickii*, progenitor of cultivated cucumber is found in the Himalayan foothills of India. Today, cucumber is grown throughout the world in large commercial farms, glasshouses and small gardens.Its fruits are eaten at immature stage as refreshing salad vegetable and are said to have cooling effect, prevent constipation and are useful to jaundice patients. Globally, it is cultivated in 2,271,260 hectares areawith an annual production of 83,753,861 tonnes (Anonymous, 2017). Compared to this, the corresponding figures for India are 25,676 hectares and 1.61 lakh tons (Anonymous, 2017). Cucumber is both a leading commercial crop and popular home garden vegetable in low and mid hills of Himachal Pradesh and the crop brings lucrative returns to the hill farmers during July to October, when it is not produced in the adjoining plains.

After the first report of hybrid vigour in cucumber (Hayes and Jones, 1916), a large number of hybrids have been developed and almost ninety per cent of the total cucumber area planted is covered by hybrids in developed countries like USA, France, Germany, Netherlands, Russia and Japan. The development of hybrid cultivar became easy after gynoecious sex expression was obtained from a Korean cultivar. Gynoecious allele is dominant and cucumber hybrids involving gynoecious parent will bear high proportion of female flowers, resulting in earliness and good yield.The first gynoecious hybrid cultivar, 'Saprtan Dawn' was introduced in 1962 by CE Peterson.

Despite being home of cucumber, the work on breeding and improvement of cucumber has been rather limited in India. At national level, F_1 hybrid 'Pusa Sanyog' has been released by IARI, Katrain (Gill *et al*., 1973) by crossing gynoecious line, isolated from a Japanese variety 'Kaga Aomoga Fushinavi' with 'Green Long' of Naples, an Italian variety, which outyielded

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the recommended variety by 128.78 per cent. But this hybrid is confined to cooler and sub-tropical conditions. Dr. YS Parmar University of Horticulture and Forestry has also developed two hybrids KH-1 and KH-2 but under high rainfall areasthey perform poorly.

Development of new strains superior to the existing ones, with respect to yield and other desirable traits is one of the primary objectives of vegetable breeding. Higher productivity is the need of the hour, and can be met by adopting heterosis breeding. For exploitation of heterosis, choice of suitable parents is of utmost importance. The study of general combining ability (GCA) of parents and specific combining ability (SCA) of crosses provides information for selecting suitable parents and cross combinations, respectively.Of the different biometrical approaches now available to determine the genetic information from the performance of hybrids and to identify appropriate cross-combinations, the "Line × Tester" mating design as proposed by Kempthorne (1957) gives comparable estimate of the genetic make-up of genotypes.

For breeders, agro-ecological diversity of environments represents a double-edged sword. This diversity complicates breeding and testing of improved genotypes with adequate adaptation, but it also permits identification of extreme environmental conditions that guarantee selection pressure from important stresses (Ramagosa and Fox, 1993). Since the quantitative characters are considerably influenced by the environment, a study under different locations/ environments is likely to bring out genotype-environment interactions for precise estimates of the genetic variation and prediction of genetic advance under selection. Thus, the present investigation was undertaken to estimate the nature of combining ability and type of gene action with respect to yield and yield attributing traits in cucumber.

Materials and Methods

Description of locations

The present investigations were carried out at the Experimental Farms of the Department of Vegetable Science and Floriculture, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur and Hill Agricultural Research and Extension Centre, Bajaura, Kullu during *Kharif*, 2009.

The Palampur Experimental Farm is located at an elevation of about 1290.8 m above mean sea level with 32°8' North latitude and 76°3' East longitude, representing mid hill zone of Himachal Pradesh and has a sub-temperate climate with high rainfall (2,500 mm). The soil of this zone is silty clay loam with acidic reaction. The Bajaura Experimental Farm is situated at 31°8' North latitude and 77° East longitude at an elevation of 1,090 m above mean sea level. Bajaura falls under mid-hill, sub-humid zone (Zone-II) of the stateand is endowed with mild summer and cool winter with low monsoon rains. The soil of this location is sandy loam with high water-table.

Breeding material

The experimental material comprised of F_1 populations of 55 crosses, five gynoecious lines, 11 testers and two standard checks (Table 1). All the lines used as female parents were crossed to each of the testers by hand pollination in a line × tester model at Palampur. Simultaneously, parents were also maintained by selfing.

Experimental layout and cultural practices

The F_1 population of 55 crosses, 16 parents and two standard checks were grown in a completely randomized Block Design with three replications during summer-rainy seasons of 2009 in each environment. Seeds were sown in poly bags (size 6" × 3") on March 14 at Palampur and March 19, at Bajaura inside a poly-house and transplanted at 2-4 true leaf stage in the field on April 22, at Palampur and May 1 at Bajaura at an inter and intra-row spacing of 1.5 m and 0.5 m, respectively. The vines were staked within fortnight after transplanting. Recommended cultural operations were followed as per the package of practices for raising a healthy crop.

Data collection and statistical analysis

The agronomical and morphological traits investigated included: days to first female flower appearance, nodal position of first female flower, days taken to first picking, fruit length (cm), fruit diameter (cm), average fruit weight (g), number of marketable fruits per vine, marketable yield per vine (kg), harvest duration, number of primary branches per plant, vine length (m) and total soluble solids (%). Observations were recorded from five competitive plants in each entry and replication for the horticultural traits.The data recorded on 55 crosses along with 16 parents and two standard checks were subjected to analysis of variance, as per the model suggested by Panse and Sukhatme (1967).The variation among the hybrids was partitioned further into sources

Table 1. Germplasm of cucumber and their sources of procurement

S. No.	Genotype	Location
a) Lines		
1.	EC-5082	Regional Research Station, Indian Agriculture Research Institute, Katrain
2.	Plp-Gy-1 (Plp)	Department of Vegetable Science and Floriculture, CSK HPKV, Palampur
3.	G-1	Department of Vegetable Crops, Dr. YS Parmar University of Horticulture and Forestry, Nauni, Solan
4.	G-3	-do-
5.	PCUCP-4	GB Pant University of Agriculture and Technology, Pantnagar, Uttrakhand
b) Testers		
1.	KL-1	Department of Vegetable Science and Floriculture, CSK HPKV, Palampur
2.	Khira Paprola (K-Pap)	-do-
3.	Japanese Long Green (JLG)	Regional Research Station, Indian Agricultural Institute, Katrain
4.	Poinsette	National Seeds Corporation, New Delhi
5.	DPC-1	Department of Vegetable Science and Floriculture, CSK HPKV, Palampur
6.	EC-173934	-do-
7.	Summer Green (SG)	-do-
8.	K-90	Department of Vegetable Crops, Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan
9.	Sel-75-2-10	Department of Vegetable Science and Floriculture, CSK HPKV, Palampur
10.	K-75	Department of Vegetable Crops, Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan
11.	KL-3	Department of Vegetable Science and Floriculture, CSK HPKV, Palampur
c) Checks		
1.	Solan Khira Hybrid-1	Department of Vegetable Crops, UHF, Solan
2.	Pusa Sanyog	Regional Research Station, IARI, Katrain

attributed to general combining ability (GCA) and specific combining ability (SCA) components in accordance with the procedure suggested by Kempthorne (1957). The per cent contribution of lines, testers and their interactions were calculated as per statistical procedures suggested by Singh and Chaudhary (1977).

Results and Discussion

Studies on combining ability

The combining ability studies evaluate the parental lines on the basis of their general combining ability (GCA) effects and the performance of these parents in specific cross combinations (SCA). General combining ability effects, being related to additive genetic effects, represent the fixable components of genetic variance and are used to classify the parents for the breeding behaviour in hybrid combinations, whereas, specific combining ability effects are related to non-fixable component of genetic variance (Hayman, 1960, Sprague, 1966).

Analysis of variance for combining ability

The analysis of variance for combining ability at pooled over environments (Table 2) revealed significant differences among hybrids for all the traits studied. Significant differences for mean sum of squares due to lines were observed for all the traits, when tested against mean sum of squares due to error. The mean sum of squares due to lines × environment and tester × environment were significant for all the traits, except nodal position of first female flower, fruit girth, number of primary branches and vine length in pooled analysis. Line × tester × environment interactions were found significant for all the traits against mean sum of squares due to error except fruit girth. The significance of mean squares due to lines × environment, testers × environment and (L vs T) × environment interactions for majority of the traits suggested that both parents as well as their interaction variances were influenced by the environment as also reported by Sudhakhar *et al.* (2005) and Sharma *et al*. (2006) with their set of breeding material and environment.

General combining ability (GCA) effects

The mean sum of squares due to lines and testers were significant for all the characters, hence GCA effects have been estimated for all the traits exhibiting significant mean sum of squares. The parents with general combining ability effects for different traits in pooled over environments have been presented in Table 3 and 4.

The results with regard to all the 16 parents for combining ability have been found to be variable as no single parent has exhibited significant GCA effects for all the traits. A perusal of GCA effects for earliness (days

Table 2. Analysis of variance for combining ability (pooled over environments) in cucumber

S. No.	Sources of variation	df	Days to first female flower appearance	Nodal position of first female flower	Days taken to first picking	Fruit length (cm)	Fruit girth (cm)	Average fruit weight (g)	Marketable fruits per vine	Marketable yield per vine (kg)	Harvest duration	Number of primary branches	Vine length (m)	Total soluble solids (%)
1	Lines	4	30.739@**	1.705@**	12.675**	76.792@**	2.477@**	17509.700@**	15.123**	0.388**	91.159@**	0.787@**	2.920@**	0.875**
2	Testers	10	6.801**	0.401**	17.936**	87.221@**	2.804@**	4605.387**	22.592**	1.596**	1.482**	0.177**	0.990@**	0.246**
3	Line vs Tester	40	7.392**	0.831**	19.240**	18.573**	0.672**	3866.856**	11.216**	1.181**	24.737**	0.360**	0.334**	0.613**
4	Line ×Loc	4	32.099**	0.111	47.434**	5.421**	0.001	841.758**	1.187**	0.015	141.465**	0.001	0.002	0.630**
5	Tester ×Loc	10	2.740**	0.106	5.758**	3.678**	0.014	822.366**	1.166**	0.195**	32.140**	0.002	0.001	0.001
6	L vs T ×Loc	40	4.726**	0.285**	5.389**	3.158**	0.043	688.951**	1.964**	0.175**	17.730**	0.241**	0.058**	0.144**
	Error	216	0.233	0.060	0.343	0.356	0.020	12.115	0.042	0.002	0.407	0.093	0.011	0.030

* Significant at 5% level of significance when tested against MSS due to error
** Significant at 1% level of significance when tested against MSS due to error
@Significant at 5% level of significance when tested against MSS due to line x teste

Table 3. Estimates of general combining ability (GCA) effects of lines in F_1 generation at pooled over environments in cucumber

S.No.	Lines / Traits	EC-5082	Plp-Gy-1	G-1	G-3	PCUCP-4	SE (gi)±	SE (gi-gj) ±	CD(5%)
1	Days to first female flower appearance	-0.033	0.183**	-0.997**	-0.151*	0.998**	0.061	0.086	0.120
2	Nodal position of first female flower	0.184**	-0.044	-0.085**	0.004	-0.060**	0.026	0.037	0.051
3	Days taken to first picking	0.029	0.005	-0.377**	-0.227**	0.570**	0.072	0.101	0.140
4	Fruit length (cm)	1.043**	0.471**	0.570**	-1.190**	-0.893**	0.079	0.111	0.154
5	Fruit girth (cm)	-0.299**	0.156**	-0.030	0.064**	0.109**	0.019	0.026	0.037
6	Average fruit weight (g)	3.065**	-1.117**	-3.989**	4.177**	-2.136**	0.423	0.598	0.829
7	Marketable fruits per vine	0.047*	-0.015	0.591**	-0.027	-0.596**	0.023	0.033	0.046
8	Marketable yield per vine (kg)	0.040**	-0.007	0.054**	0.048**	-0.132**	0.004	0.006	0.008
9	Harvest duration	0.662**	0.094	-1.010**	2.224**	-1.969**	0.069	0.098	0.136
10	Number of primary branches	-0.042	0.047	0.128**	-0.081**	-0.052	0.031	0.044	0.060
11	Vine length (m)	0.040**	0.031**	-0.022**	-0.018**	-0.031**	0.006	0.008	0.011
12	Total soluble solids (%)	0.019	0.067**	-0.025	0.025	-0.086**	0.017	0.024	0.034

* Significant at 5% level of significance
** Significant at 1% level of significance

Table 4. Estimates of general combining ability (GCA) effects of Testers in F_1 generation at pooled over environments in cucumber

S. No.	Lines / Traits	KL-1	K-PAP	JLG	Poinsette	DPC-1	EC-173934	SG	K-90	Sel-75-2-10	K-75	KL-3	SE (gj)+	SE (gi-gj) +	CD (5%)
1	Days to first female flower appearance	0.282**	-0.898**	-0.598**	-0.272**	0.486**	0.265**	-0.234**	0.248**	-0.656**	0.309**	1.069**	0.091	0.128	0.178
2	Nodal position of first female flower	0.075	-0.143**	0.091*	-0.091*	0.000	-0.059	0.129**	0.091*	-0.004	-0.020	-0.070	0.039	0.055	0.076
3	Days taken to first picking	1.098**	-1.705**	-0.585**	-0.338**	-0.182	-0.164	0.141	0.448**	-0.687**	0.695**	1.279**	0.106	0.150	0.208
4	Fruit length (cm)	2.237**	-0.041	3.423**	-0.872**	-1.345**	-1.020**	-0.443**	-0.715**	-0.825**	-0.732**	0.334**	0.117	0.165	0.229
5	Fruit girth (cm)	-0.273**	0.236**	-0.464**	-0.065*	0.043	0.223**	-0.204**	0.092**	0.102**	-0.253**	0.563**	0.028	0.039	0.054
6	Average fruit weight (g)	4.422**	12.094**	-15.205**	-12.151**	-4.695**	4.290**	5.258**	15.007**	-8.427**	-12.198**	11.604**	0.627	0.887	1.230
7	Marketable fruits per vine	0.137**	1.650**	-0.057	-1.358**	0.355**	0.188**	0.045	-0.427**	1.189**	0.237**	-1.960**	0.035	0.049	0.068
8	Marketable yield per vine (kg)	0.106**	0.442**	-0.175**	-0.389**	0.008	0.092**	0.086**	0.080**	0.147**	-0.108**	-0.289**	0.006	0.009	0.012
9	Harvest duration	0.806**	2.776**	-1.467**	-3.811**	0.355**	0.378**	0.095	0.661**	0.759**	-2.563**	2.011**	0.103**	0.145	0.201
10	Number of primary branches	0.019	0.045	0.022	-0.234**	-0.051	0.061	-0.004	0.043	0.089	0.023	-0.012	0.046	0.065	0.090
11	Vine length (m)	0.042**	-0.071**	0.114**	-0.215**	0.010	-0.047**	-0.055**	0.084**	0.053**	0.039**	0.047**	0.008	0.012	0.016
12	Total soluble solids (%)	-0.021	-0.065*	-0.060*	0.061*	-0.022	0.066**	0.027	0.118**	0.184**	-0.141**	-0.146**	0.025	0.036	0.050

* Significant at 5% level of significance
** Significant at 1% level of significance

to first female flower appearance, nodal position of first female flower and days taken to first picking) revealed that G-1 and G-3 among lines and K-pap, JLG and Poinsette among testers with significant negative GCA effects were the best combiners across environments.

G-1, G-3 and EC-5082 among lines and K-pap, Sel-75-2-10, KL-1, EC-173934, SG and K-90 among testers exhibited the highest positive GCA effects for marketable yield per vine across the environments. Of these lines and testers, G-3, EC-5082, K-90, Sel-75-2-10, K-pap, SG, KL-1 and EC-173934 were also found to be best combiners for average fruit weight, marketable fruits per vine and fruit size (fruit length and fruit girth), thereby suggesting close association between GCA of the lines and testers for fruit yield with fruit number, fruit weight, and fruit size.

For plant growth characters, G-1 and Plp-Gy-1 among lines and KL-1, JLG, K-90 and Sel-75-2-10 among testers for number of primary branches and vine length were good combiners. For total soluble solids, Plp-Gy-1, K-90 and Sel-75-2-10 were observed good general combiners over the environments. Different parents expressing high desirable GCA in respect of yield and component traits have been reported by different workers by using different genetic materials and locations (Singh and Sharma 2006, Tiwari and Singh 2016 and Malav *et al*. 2018).

Additive parental effects as measured by GCA effects are of practical use to the breeders since non-allelic interactions are unpredictable. On the basis of present investigations for GCA effects, it may be concluded that the parents *viz.,* EC-5082, G-1, G-3, KL-1, K-Pap, Summer Green, K-90 and Sel-75-2-10 are good general combiners for yield and its component traits and may be utilized in hybridization programmes for getting transgressive segregants.

Specific combining ability (SCA) effects

Variances due to line × tester × location interaction was non-significant for fruit girth in pooled analysis. Consequently, SCA effects of the traits showing non-significant variances were not estimated. The specific combining ability (SCA) effects estimated for different traits have been presented in Table 5. No single cross could reveal significant SCA for all the traits. Majority of the cross combinations exhibiting desirable SCA effects, had at least one of the parents as good or average general combiner. Similar views have also been expressed by earlier researchers (Singh and Sharma 2006, Munshi *et al.,* 2006 and Yadav*et al.* 2007).

The cross combinations Plp × K-Pap, G-3 × Poinsette and G-1 × K-75 can be exploited to isolate transgressive segregants in early generations as they involve both parents with high GCA effects for earliness and marketable yield per vine, respectively. Similarly, in other cross combinations involving one good and other poor or average combiner may give desirable transgressive segregants in the later generations if the additive effect of one parent and complementary epistatic effects (if present in the cross) act in same direction and maximize the desirable plant attributes as reported by Sharma (1999).

In few cross combinations *viz.* PCUCP-4 × KL-3 (earliness), PCUCP-4 × JLG (marketable yield per vine) and PCUCP-4 × K-Pap (total soluble solids), although significant SCA effects were observed but these hybrids had both the parents as poor general combiners. This might be due to parental lines used in the present study had origin from the diverse genetic background and hence exhibited high SCA effects. These observations corroborate the views of Krishna Prasad and Singh (1994) who opined that, it is not necessary that parents having higher estimates of GCA effects would also give higher estimates of SCA effects, usually the highest estimates of SCA effects are obtained from crosses involving diverse parents. Both parents with high GCA effects when crossed had probably low magnitude of non-additive gene effects resulting in small degree of SCA effects. Therefore, recurrent selection for specific combining ability could be followed in the segregating generations, on the assumption that an important part of heterosis results from the non-linear interaction of genes at different loci from interaction between alleles at the same locus or from both causes in combination.

On the basis of present study for GCA and SCA effects, it may be concluded that cross combinations Plp-Gy-1 × K-pap, G-1 × K-pap, Plp-Gy-1 × K-90, G-1 × K-90 and G-3 × Sel-75-2-10 came out to be the best specific combiners for yield and yield contributing traits. Similar findings for identification of superior parental lines, tester and hybrids based on GCA and SCA effects for fruit yield and morphological characters in cucumber were reported by Kumar *et al.* (2013), Golabadi *et al.* (2015) and Tak *et al*. (2017).

Table 5. Estimates of specific combining ability (SCA) effects of crosses in F_1 generation in cucumber (pooled over environments)

S. No.	Traits Crosses	Days to first female flower appearance	Nodal position of first female flower	Days taken to first picking	Fruit length (cm)	Fruit girth (cm)	Average fruit weight (g)	Marketable fruits per vine	Marketable yield per vine (kg)	Harvest duration	Number of primary branches	Vine length (m)	Total soluble solids (%)
1	EC-5082 × KL-1	0.700**	-0.159	0.260	0.684**	-	-58.490**	-0.263**	-0.750**	-7.774**	0.154	0.028	-0.250**
2	EC-5082 × K-pap	-0.286	-0.418**	-1.128**	-1.709**	-	-1.293	-0.944**	-0.184**	-1.187**	-0.320**	-0.024	0.010
3	EC-5082 × JLG	1.226**	-0.071	2.415**	-3.483**	-	-23.385**	-1.639**	-0.552**	-4.306**	0.069	-0.050**	-0.011
4	EC-5082 × Poinsette	-1.387**	0.138	0.255	-0.529*	-	-23.665**	-1.004**	-0.431**	-4.462**	0.025	0.127**	0.051
5	EC-5082 × DPC-1	1.566**	0.272**	2.037**	-1.102**	-	22.841**	-0.201**	0.241**	-1.267**	-0.057	-0.063**	0.074
6	EC-5082 × EC-173934	-0.757**	-0.073	-1.307**	0.936**	-	5.511**	2.001**	0.470**	1.822**	0.405**	0.104**	0.086
7	EC-5082 × SG	-0.784**	-0.119	-1.504**	-1.925**	-	10.478**	0.936**	0.315**	0.632**	0.286**	0.011	-0.065
8	EC-5082 × K-90	-0.435*	0.078	-0.461	2.554**	-	15.992**	0.484**	0.178**	4.233**	-0.119	-0.022	0.145*
9	EC-5082 × Sel-75-2-10	-1.257**	0.258**	-2.286**	1.541**	-	18.645**	-0.758**	0.095**	2.302**	0.002	-0.107**	0.078
10	EC-5082 × K-75	0.452*	0.055	0.582*	1.194**	-	7.229**	1.569**	0.420**	7.290**	-0.348**	0.098**	-0.197**
11	EC-5082 × KL-3	0.963**	0.039	1.138**	1.839**	-	26.137**	-0.182*	0.197**	2.717**	-0.097	-0.101**	0.079
12	Plp × KL-1	-0.016	-0.378**	-0.716**	0.427	-	6.215**	0.058	0.146**	4.322**	0.175	-0.028	0.125*
13	Plp × K-pap	-0.835**	-0.160	-0.774**	0.496	-	24.179**	0.755**	0.475**	0.394	0.208*	0.052**	-0.051
14	Plp × JLG	0.727**	0.135	0.693**	2.862**	-	-18.218**	0.881**	-0.067**	2.984**	0.119	0.052**	-0.201**
15	Plp × Poinsette	0.799**	0.249**	0.863**	-0.718**	-	31.166**	1.406**	0.612**	4.106**	0.318**	-0.014	0.103
16	Plp × DPC-1	-0.386	0.032	0.493*	0.295	-	42.723**	0.415**	0.553**	3.717**	0.143	0.056**	-0.097
17	Plp × EC-173934	-0.186	0.211*	0.272	-0.826**	-	4.410**	-0.819**	-0.126**	-0.195	0.158	-0.027	0.023
18	Plp × SG	-0.416*	0.029	0.245	0.624*	-	-4.443**	-1.434**	-0.366**	2.200**	-0.328**	0.031	0.046
19	Plp × K-90	1.768**	0.032	1.354**	-1.192**	-	-23.818**	-1.333**	-0.525**	-7.921**	-0.208*	-0.018	0.030
20	Plp × Sel-75-2-10	0.560**	0.037	1.245**	-0.187	-	-28.350**	-1.209**	-0.602**	-6.074**	-0.420**	-0.157**	-0.004
21	PlP × K-75	-2.012**	-0.200*	-3.671**	-0.695**	-	-21.596**	-0.079	-0.254**	-3.317**	0.063	-0.053**	-0.095
22	Plp × KL-3	-0.002	0.012	-0.005	-1.086**	-	-12.266**	1.360**	0.152**	-0.216	-0.228*	0.106**	0.120*
23	G-1 × KL-1	-0.670**	-0.175*	-0.442	1.669**	-	18.930**	0.056	0.237**	3.093**	0.194	-0.158**	0.060
24	G-1 × K-pap	-0.336	-0.124	-1.472**	0.430	-	20.739**	-0.081	0.134**	0.128	0.137	0.145**	-0.029
25	G-1 × JLG	0.711**	-0.102	1.265**	-0.517*	-	-19.626**	-0.498**	-0.337**	-5.135**	-0.100	0.005	0.099
26	G-1 × Poinsette	0.384	-0.171*	0.161	-0.652*	-	1.710	-1.908**	-0.327**	-2.790**	-0.261*	-0.045*	-0.105
27	G-1 × DPC-1	-0.791**	-0.025	-1.331**	0.164	-	-11.978**	1.159**	0.085**	-0.264	-0.085	0.064**	0.028
28	G-1 × EC-173934	0.008	0.151	0.667**	-0.050	-	20.876**	-1.430**	-0.052**	2.521**	-0.339**	-0.064**	-0.110
29	G-1 × SG	0.347	-0.015	-0.818**	0.482	-	4.140**	0.921**	0.264**	0.248	0.092	-0.006	0.046
30	G-1 × K-90	0.086	-0.103	0.541*	-1.063**	-	9.642**	0.537**	0.276**	2.738**	0.228*	-0.075**	-0.012
31	G-1 × Sel-75-2-10	1.102**	0.334**	2.010**	-1.349**	-	-24.920**	-0.535**	-0.438**	0.390	-0.167	0.133**	0.001
32	G-1 × K-75	-0.553**	0.098	-0.457	0.007	-	-0.993	2.994**	0.559**	2.128**	0.399**	0.040*	0.287**
33	G-1 × KL-3	-0.289	0.133	-0.123	0.877**	-	-18.520**	-1.214**	-0.400**	-3.057**	-0.097	-0.040*	-0.265**
34	G-3 × KL-1	-0.183	0.074	0.369	-1.121**	-	16.727**	0.364**	0.264**	0.362	0.044	0.067**	0.077
35	G-3 × K-pap	0.748**	0.512**	0.766**	0.057	-	-28.724**	0.473**	-0.225**	1.722**	0.135	-0.075**	-0.229**
36	G-3 × JLG	-1.302**	0.249**	-1.642**	-0.724**	-	19.047**	-0.708**	0.062**	0.632**	-0.467**	0.000	0.186**
37	G-3 × Poinsette	-1.545**	-0.265**	-2.047**	1.092**	-	5.480**	0.392**	0.117**	3.476**	-0.244*	-0.080**	-0.139*
38	G-3 × DPC-1	-0.775**	0.007	-1.428**	0.846**	-	-31.834**	0.937**	-0.217**	3.810**	0.181	-0.075**	-0.006
39	G-3 × EC-173934	0.196	-0.291**	-1.223**	-0.165	-	-24.215**	-1.466**	-0.562**	-8.462**	-0.264**	-0.068**	-0.110
40	G-3 × SG	0.501*	0.521**	1.531**	-0.825**	-	15.235**	0.307**	0.228**	3.321**	-0.083	0.001	0.049
41	G-3 × K-90	-0.789**	-0.441**	-1.862**	0.275	-	12.082**	1.359**	0.454**	3.754**	0.157	0.071**	0.038
42	G-3 × Sel-75-2-10	0.066	-0.347**	-0.473*	0.416	-	13.535**	1.424**	0.473**	0.282	0.341**	-0.019	0.001
43	G-3 × K-75	1.971**	-0.051	3.615**	0.897**	-	8.440**	-2.640**	-0.447**	-6.772**	0.057	0.076**	0.097
44	G-3 × KL-3	1.113**	0.032	2.393**	-0.748**	-	-5.773**	-0.443**	-0.147**	-2.125**	0.142	0.104**	0.035
45	PCUCP-4 × KL-1	0.169	0.639**	0.528*	-1.658**	-	16.619**	-0.214**	0.103**	-0.003	-0.568**	0.091**	-0.012
46	PCUCP-4 × K-pap	0.710**	0.190*	2.608**	0.726**	-	-14.901**	-0.203**	-0.201**	-1.057**	-0.160	-0.098**	0.299**
47	PCUCP-4 × JLG	-1.361**	-0.210*	-2.731**	1.862**	-	42.182**	1.964**	0.894**	5.825**	0.380**	-0.006	-0.073
48	PCUCP-4 × Poinsette	1.750**	0.048	0.768**	0.807**	-	-14.691**	1.114**	0.029*	-0.331	0.162	0.013	0.089
49	PCUCP-4 × DPC-1	0.386	-0.286**	0.228	-0.203	-	-21.751**	-2.311**	-0.663**	-5.997**	-0.181	0.018	0.001
50	PCUCP-4 × EC-173934	0.739**	0.003	1.592**	0.104	-	-6.582**	1.715**	0.270**	4.314**	0.041	0.055**	0.111
51	PCUCP-4 × SG	0.352	-0.415**	0.547*	1.644**	-	-25.410**	-0.731**	-0.441**	-6.401**	0.033	-0.037*	-0.076
52	PCUCP-4 × K-90	-0.630**	0.434**	0.428	-0.575*	-	-13.898**	-1.048**	-0.383**	-2.803**	-0.058	0.044*	-0.201**
53	PCUCP-4 × Sel-75-2-10	-0.472*	-0.283**	-0.496*	-0.422	-	21.090**	1.078**	0.473**	3.100**	0.243*	0.150**	-0.077
54	PCUCP-4 × K-75	0.143	0.097	-0.069	-1.404**	-	6.919**	-1.844**	-0.279**	0.671**	-0.171	-0.161**	-0.092
55	PCUCP-4 × KL-3	-1.786**	-0.216*	-3.403**	-0.882**	-	10.422**	0.479**	0.199**	2.681**	0.280**	-0.069**	0.030
	SE (Sij)±	0.203	0.087	0.237	0.261	-	1.403	0.078	0.014	0.230	0.102	0.018	0.057
	SE(sij-skl)±	0.287	0.123	0.336	0.369	-	1.984	0.110	0.020	0.325	0.145	0.026	0.081
	CD (5%)	0.397	0.170	0.465	0.511	-	2.749	0.152	0.027	0.450	0.201	0.036	0.112

* Significant at 5% level of significance
** Significant at 1% level of significance

Gene action

After the identification of appropriate parents and potential crosses, the next important step in a dynamic breeding programme is with respect to adoption of suitable breeding methodology for the purposeful management of generated variability which largely depends upon the type of gene action in the population for the traits under genetic improvement (Sprague 1966). The nature of gene action has been inferred from the estimates of GCA and SCA variances, which are presented in Table 6 for pooled analysis.

A perusal of the values indicated that the estimates of σ^2_{sca} were higher as compared to σ^2_{gca} (average) for all the traits studied, similarly reported by Pradhan *et al*. (2016), Bhutia *et al*. (2017) and Naik *et al.* (2018). It indicates predominant role of non-additive gene action, which means hybrid vigour could better be exploited for these traits. The results of analysis of variance for combining ability were also confirmed from the study of additive (σ^2A) and dominant (σ^2D) components of variance. In all the traits studied, where SCA variances were higher than GCA values, dominant components of variance (σ^2D) were also higher than the additive components (σ^2A) indicating the role of non-additive gene action. For fruit length, though SCA variances were higher but the value of σ^2D was low. This might be attributed to the fact that statistically GCA variance is the additive portion of the variability, but it also includes additive × additive and higher order of epistatic interaction.The results of present study are in accordance with the earlier researchers for the traits related to marketable yield and fruit size (Singh and Sharma, 2006, Munshi *et al*., 2006 and Yadav *et al*., 2007).

The results obtained in this study lead to the conclusion that the hybrids were less stable over the environments as compared to the lines and testers. Conclusively, it may be stated that non-additive gene action governs the traits studied and thus, hybrid vigour could better be exploited for these traits.

Proportional contribution of lines, testers and their interactions

In pooled analysis, the per cent contribution of line × tester interactions were found to be greater than the individual contribution of lines and testers for all the traits except fruit length, fruit girth, vine length and total soluble solids where per cent contribution of testers was greater than individual contribution of lines and line × tester interactions.

Table 6.Estimates of genetic components of variance and proportional (%) contribution of lines, testers and their interactions in cucumber (pooled over environments)

Components S. Traits No.		σ^2 GCA (Average)	σ^2 GCA × Env.	σ^2 SCA	σ^2 SCA × Env.	σ^2 A	σ^2 D	Heritability (%) (Narrow sense)	Genetic Advance 5%	% Contribution of Lines (%)	Testers (%)	Interaction (%)
1	Days to first female flower appearance	0.317	0.70	1.090	1.50	1.268	2.179	27.790	0.750	26.548	20.105	53.347
2	Nodal position of first female flower	0.001	0.03	0.071	0.07	0.002	0.142	2.770	0.020	12.744	9.443	77.814
3	Days taken to first picking	0.051	1.29	2.930	1.68	0.206	5.860	5.420	0.190	4.403	24.661	70.937
4	Fruit length (cm)	1.123	0.10	1.827	0.94	4.493	3.653	54.110	2.270	18.404	48.634	32.961
5	Fruit girth (cm)	0.038	0.09	0.084	0.10	0.151	0.169	45.890	0.380	15.683	45.907	38.409
6	Average fruit weight (g)	27.371	14.00	581.863	228.11	109.483	1163.726	11.390	5.330	1.789	19.974	78.237
7	Marketable fruits per vine	0.197	0.07	1.954	0.64	0.787	3.909	13.920	0.420	5.662	37.286	57.053
8	Marketable yield per vine (kg)	0.004	0.01	0.197	0.06	0.016	0.394	4.330	0.040	2.456	24.455	73.089
9	Harvest duration	0.522	4.23	19.152	5.69	2.089	38.303	2.830	0.260	10.618	17.323	72.059
10	Number of primary branches	0.002	0.01	0.063	0.05	0.007	0.127	7.870	0.040	8.907	10.191	80.902
11	Vine length (m)	0.002	0.01	0.008	0.02	0.010	0.016	22.630	0.100	5.992	53.235	40.773
12	Total soluble solids (%)	0.003	0.01	0.016	0.04	0.013	0.032	3.510	0.020	10.234	69.426	52.982

After having an insight into the GCA and SCA effects and variances as well as additive (σ^2A) and dominant (σ^2D) components of variance, it may be worthwhile to effect improvement in cucumber by developing superior open-pollinated varieties through selection in segregating population for the traits associated with earliness, fruit length and fruit girth. Alternatively, exploitation of hybrid vigour or reciprocal recurrent selection, which capitalizes on both additive and non-additive variances, might be more effective for marketable fruits per vine, marketable yield per vine, average fruit weight, number of primary branches, harvest duration and vine length, which had either high or equal dominant (σ^2D) components of variance to that of additive (σ^2A) components.The hybrids in cucumber are likely to dominate on account of the gynoecious lines and the relatively ease in producing hybrid seed commercially. The inclusion of gynoecious lines in heterosis and gene action study may lead to conclusion quite contrary to the ones obtained with mostly monoecious lines.

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Conflict of Interest

The authors declared that they have no conflict of interest.

Contribution of authors

MS performed hybridization programme, field evaluation and statistical analysis. AV contributed in statistical analysis and manuscript writing. YS participated in all steps of evaluation and data compiling.

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RESEARCH ARTICLE

Studies on Diversity of Lemon (*Citrus limon* (L.) Burm.) based on Quantitative Traits under West Bengal Conditions

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A survey was done in 12 districts of West Bengal to identify the elite lemon genotypes resulted in the identification of 52 accessions which were further characterized by using 22 quantitative characters for descriptive analysis, hierarchical cluster analysis, discriminant analysis, correlation co-efficient analysis and principal Component analysis and biplot. Wide variation was observed in 12 quantitative characters*i.e.*fruit weight, fruit rind thickness, juice weight, juice volume, juice percentage, number of seeds per fruit, seed weight, seed length, seed width, TSS: Acid ratio, total sugar and non reducing sugars led to, hierarchical cluster analysis these collections into 20 clusters. Major characters responsible for such clustering by Canonical discriminant analysis were fruit length, juice volume, juice percentage, seed length and TSS: Acid. Principal component analysis (PCA) explained seven components with cumulative variance of 77.432%. However, biplot analysis revealed genotypes present in different quadrant of scoring plot had higher values of quantitative characters remained in corresponding quadrant of loading plot. From this study it is concluded that the variability found in lemon genotypes can be exploited for the selection of elite material for further conservation, detailed evaluation and utilization in the crop improvement programme.

Key Words: Clustering, Dendrogram, Lemon, Principal component analysis

Introduction

In India citrus fruit ranks second in area after mango and third in production after banana and mango, and accounts for 15.41% of the total fruit area and 12.89% of the total fruit production. Lime and lemon being the third most important fruits of citrus, contribute 28.51% share of citrus fruit area and 25.1% of citrus fruit production in India (Saxena, 2018). Lemon fruits have high medicinal and nutritive values as excellent source of vitamin C, beta-carotene and thiamine. Lemon is not usually taken as fresh fruit. The juice is used for making squash refreshing drink and in many culinary purposes, as a garnish of fish and meat. Fruits are used for making pickles and lemon oil. It is also used as stain remover and as a bleaching agent. The health benefits of lemon include treatment of throat infections, indigestion, constipation, dental problems fever, internal bleeding, rheumatism, burns, obesity, respiratory disorders, cholera and high blood pressure,beneficial for hair and skin care. It is very well known for its therapeutic property since generations, lemon helps to strengthen immune system, cleanse stomach, and it is considered as a blood purifier (Organic Facts, 2015).

Therefore, lemon has a great prospect for commercial exploitation despite of its tremendous potentiality; it still has not gained the importance in India. It is mostly grown in homestead and kitchen gardens and very few varieties have been developed which are also not wellaccepted/ adapted throughout India. The diverse eco-geographical distribution in India and the occurrence of spontaneous mutation and natural hybridization have given rise to a wide range of variability in citrus (Dubey *et al*., 2016).

Lemon is heterozygous in nature and thus exhibits a wider variability in seedling population. Information on genetic diversity and phylogeny of cultivars can improve the efficiency of germplasm characterization and its use in breeding programs (Gulsen and Roose, 2001). Importance of clonal selections in crop improvement is well recognized by earlier workers (Badge and Patil,

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1989; Badiyala *et al*., 1992). In West Bengal, diversity of a large sample of lemon genotypes from a wide range of geographic locations has not been reported earlier. Hence, the present investigation of diversity was carried out to know the variability and heterogeneity among different lime collections and locate the elite genotypes possessing desirable fruit characteristics.

Materials and Methods

Fifty two genotypes of lemon were selected by thorough survey and first hand information from growers covering 12 districts of West Bengal during 2015-17. The different collections were named based on code used for different districts (first two letters) and the first letter 'L' of lemon. Thus, different genotypes were named as BNL (collected from Bankura), BRL (Bardhaman), BIL (Birbhum), CBL (Cooch Behar), HGL (Hooghly), HRL (Howrah), NAL (Nadia), PNL (North 24 parganas), PML (Pascim Medinipur), PRL (Purba Medinipur), PUL (Purulia) and PSL (South 24 Parganas). Characterization of selected lemon was performed using 22 quantitative characters chosen from 'Citrus Descriptor' (IPGRI, 1999).Twenty fully matured and healthy fruits from each genotype were collected randomly from different directions of the canopy and brought to laboratory of Fruit Science Department of Bidhan Chandra Krishi Viswavidyalaya for recording quantitative observation. Electronic (digital) balance was used for recording fruit and seed weight. Fruit size, fruit rind thickness, vesicle length and seed size were measured by slide calliperse. Total soluble solids content of fruits was determined with the help of a digital refractometer and calibrated in ºbrix at 20 ^{o}C. Titratable acidity, total sugar, reducing sugar and non-reducing sugars are estimated by following the methods as described in A.O.A.C. (1984). Ascorbic acid was estimated by the method as described by Ranganna (2000).

Statistical analysis was done for descriptive, hierarchical cluster, discriminant, principal component and biplot analysis. Descriptive statistics used the data to provide descriptions of the population. Hierarchical cluster analysis following single linkage (nearest neighborhood) method, where distance matrix was Euclidian, was attempted to identify relatively homogeneous groups of varieties. Cluster members were further subjected to canonical discriminant analysis for multiple group problems to find out characters responsible for such clustering. Principal component Analysis and Biplot analysis were done to clarify the relation between genotypes and variables

Results and Discussion

Descriptive analysis of 52 genotypes (Table 1) indicated higher co-efficient of variation (>20) for 12 quantitative

Table 1. Variability study of different quantitative characters of lemon

Characters	Minimum	Maximum	Mean	Std. Deviation	CV (%)
Fruit weight (g)	40.5	170.00	100.15	32.53	32.48
Fruit diameter (mm)	30.50	71.82	50.95	9.37	18.39
Fruit length (mm)	41.63	107.31	73.64	13.94	18.93
Oil glands (/cm2)	32.00	86.00	66.11	11.49	17.38
Rind thickness (mm)	1.45	7.16	3.32	1.33	40.02
Number of segments	7.66	14.00	10.93	1.12	10.28
Vesicle length	3.37	17.22	12.11	2.11	17.44
Juice weight (g)	21.5	64.50	33.72	8.72	25.88
Juice volume (ml)	20.00	65.00	33.26	8.85	26.62
Juice percentage	21.17	58.33	35.14	8.70	24.78
Number of seeds/ fruit	0.00	67.00	27.84	21.58	77.51
Seed weight (g)	0.00	0.35	0.08	0.06	77.34
Seed length (mm)	0.00	12.42	7.08	3.92	55.31
Seed width (mm)	0.00	6.50	3.69	2.09	56.78
Acidity (%)	4.36	7.40	5.73	0.85	14.82
pH	1.74	2.90	2.07	0.26	12.82
TSS (°brix)	4.8	8.40	6.54	0.77	11.84
TSS: Acid	0.78	1.94	1.17	0.25	21.34
Ascorbic acid (mg/100 ml juice)	32.00	55.00	40.83	5.68	13.92
Reducing Sugars (%)	1.00	1.53	1.19	0.12	10.55
Total Sugars (%)	1.58	3.33	2.21	0.52	23.85
Non Reducing Sugars (%)	0.23	2.10	0.96	0.47	49.49

characters like fruit weight, fruit rind thickness, juice weight, juice volume, juice percentage, number of seeds per fruit, seed weight, seed length, seed width, TSS: acid, total sugars and non reducing sugars. Among 12 quantitative characters the co-efficient of variation was remarkably high (>50) in seed characters like number of seeds per fruit (77.51), seed weight (77.34), seed length (55.31) and seed width (56.78). The coefficient of variation was also high in rind thickness (40.02) and non-reducing sugar (49.59). Higher co-efficient of variation revealed higher variability which indicated that superior clones can be identified from the existing variation. Dubey *et al.* (2016) also found higher coefficient of variation in traits like fruit weight, fruit length, juice volume and seed content.

Wide range of values were recorded in few important physical characters of fruits (Table 1) *viz.* fruit weight (40.5 – 170 g), fruit length (41.63 – 107.31 mm), fruit diameter (30.50 – 71.82 mm), rind thickness (1.45 – 7.16 mm), segment number (7.66-14.00), vesicle length (3.37 – 17.22 mm), seed number (0.00 – 67.00) and seed weight (0.00 – 0.35 g). Wide variation of physical characters in lemon fruit was also noticed by Govind and Singh (2002), Singh *et al*. (2009), Singh and Kaur (2009) and Archan *et al.* (2013). Fallahi *et al*. (1990) obtained lesser fruit weight (97.0 to 106.5 g) and rind thickness (3.5 to 4.1 mm) as compared to values obtained in present studies. In contrast, higher range was obtained by Rana *et al*. (2003) in fruit weight (125-525g), fruit length (9.00-13.50 cm), fruit diameter (5.27-9.68 cm) with lesser number of segments (8-12) and number of seeds (0-40). The value was moderate for fruit weight (34.0 - 157.5 g), fruit length (4.08 to 6.87 cm), fruit breadth (4.04 to 5.90 cm), peel thickness (1.9 to 4.1 mm) in 'Baramasi lemon' germplasm at Punjab conditions (Jawandha *et al.,*2012). Akhter *et al*. (2013) also observed variation in fruit weight (28.98-92.53 g), fruit length (4.77-8.03 cm), fruit width (3.23-5.73 cm), seed weight (0.81-2.00 g) and peel thickness (0.18-0.33 cm).

In the present investigation, the variation was quite high in juice volume (20.00 – 65.00 ml), juice weight (21.5-64.50) and juice percentage (21.17-58.33). Fallahi *et al*. (1990) obtained comparatively lesser juice volume (39.1 to 44.4 ml) and juice per cent (40.5 to 43.4) in eight lemon cultivars at Arizona conditions. Earlier findings revealed that the percentage of lemon juice was 16.00 to 51.20 (Rana *et al*., 2003), 49 to 57 in Kachai lemon (Singh *et al*., 2006), 29.7 to 54.6 at Punjab condition (Jawandha *et al.,* 2012) and 37.68 to 41.23 at mid hills of Meghalaya (Mukhim *et al*., 2015).

Prominent variations in bio-chemical characters of fruits like TSS (4.8 – 8.40 °brix), titratable acidity (4.36 – 7.40 %), TSS/acid ratio (0.78-1.94), pH (1.74 – 2.90), ascorbic acid (32 – 55 mg/100 ml juice), reducing sugars (1.00-1.53 %), total sugars (1.58-3.33%) and non reducing sugars (0.23-2.10) were obtained in present study among different lemon collections. Wide variations of chemical composition of fruits were also obtained by different workers (Tisserat *et al.,* 1998; Govind and Singh, 2002; Singh *et al*., 2009; Singh and Kaur, 2009 and Archan *et al.,* 2013). Earlier findings exhibited that the TSS and titratable acid content were 7.26 to 7.66 ^{o}brix and 4.8 to 5.4 per cent, respectively, in the eight lemon cultivars (Fallahi *et al*., 1990) and 7.10-9.50 ^{o}brix and 4.90-5.90 per cent, respectively, in different lemons genotypes (Rana *et al*., 2003). In 'Baramasi lemon' germplasm at Punjab conditions the TSS content ranged from 7.0 to 8.8 °brix, acidity from 4.7 to 7.3 per cent and vitamin C from 25.28 to 92.91 mg/100 ml fruit juice (Jawandha *et al.,* 2012). A moderate range was noticed by Akhter *et al*. (2013) in five lemon germplasm for pH (2.12-2.15), TSS (4.87-6.03 °brix) and vitamin-C (16.61-46.22 mg/100 ml) content. In mid hills of Meghalaya, TSS (≥6.3°Brix), titratable acidity (4.18 to 4.35%), ascorbic acid (≥32.41 mg/100 g) and TSS: acidity (≥1.51) were also good in fruits of Assam lemon (Mukhim *et al*., 2015). It is clear that the variation of biochemical composition noted in the present studies was more or less similar as earlier findings.

Hierarchical cluster analysis grouped 52 lemon collections into 20 clusters considering 22 quantitative characters (Table 2 and Fig 1). Major characters responsible for such clustering by Canonical discriminant analysis were fruit length, juice volume, juice percentage, seed length, TSS: Acid (Table 3). It was found that members of a particular cluster consisted of genotypes from various sampling areas and thus indicated that phenotypic variability was not influenced by their habitat or other environmental factors. Cluster analysis is able to identify physico-chemical variability among different clusters. The variation among clusters might be due to heterozygosity, seedling population and nucella rembryony. Earlier finding of Zandkarimi *et al*. (2011) expressed 5 main clusters from 19 genotypes of lime

Table 2. Clusters of lemon genotypes based on quantitative characters using single linkage clustering methods on squared Euclidean distance matrix

Cluster Number	Cluster Members
1.	BNL 1
2.	BNL 2, BNL 3, BNL 4, BRL 2, BIL 1, HGL 4, HGL 5, NAL 2, NAL 3, NAL 6, PNL 2, PNL 3, PNL 4, PNL 5, PNL 6, PNL 7, PML 1, PML 3, PUL 1, PSL 1, PSL 3, PSL 4, PSL 5
3.	BNL 5, NAL 7, NAL 9
4.	BRL 1
5.	BIL 2
6.	CBL 1
7.	CBL 2
8.	HGL 1
9.	HGL 2
10.	HGL 3
11.	HGL 6
12.	HGL 7, HRL 2, PML 2, PRL 1, PRL 2, PRL 3, PRL 4, PRL 5
13.	NAL 1
14.	NAL 4
15.	NAL 5
16.	NAL 8
17.	PNL 1
18.	PUL 2
19.	PUL 3
20.	PSL 2

Table 3. Canonical Discriminant Function Coefficient based on quantitative characters of lemon

Variable coefficients	Function				
	1	2	3	4	5
Fruit length	-0.046	-0.104	0.098	-0.035	0.072
Juice volume	0.127	0.256	-0.066	-0.022	-0.002
Juice percentage	0.031	0.002	0.170	0.099	0.043
Seed length	0.844	-0.167	0.010	0.034	-0.014
TSS: Acid	-2.160	0.745	-4.292	3.036	2.781
(Constant)	-5.349	-0.533	-6.078	-3.978	-9.906
Eigen value	18.629a	4.316a	3.644a	2.108a	1.347a
% of Variance	62.0	14.4	12.1	7.0	4.5
Cumulative %	62.0	76.4	88.5	95.5	100.0
Canonical Correlation	0.974	0.901	0.886	0.824	0.758
Unstandardized coefficients					

and lemon. Similarly only 5 clusters were obtained by Shrestha *et al.* (2012) and 4 clusters by Kumar *et al.* (2013) from lime diversity. The more number of clusters in the present study might be due to more collection of lemon genotypes from different agro-climatic zones.

Principal component analysis explained 7 components using 22 quantitative characters with cumulative variance of 77.432 per cent with reference to eigen value more than 1. Zandkarimi *et al*. (2011) obtained 7 main factors using 31 characters with 85.99% of the total variance where as Dubey *et al.* (2016) obtained 4 components using 11 physico-chemical characters. In the present studies, out of 22 characters fruit weight, fruit diameter, fruit length, rind thickness, juice weight, juice volume, juice percentage, number of seed, seed width as observed in the first two components of PCA contributed more to the total variation. Higher contribution of fruit weight, fruit length and fruit width as revealed from PCA of pummelo genotypes was also obtained by Singh *et al.* (2015).

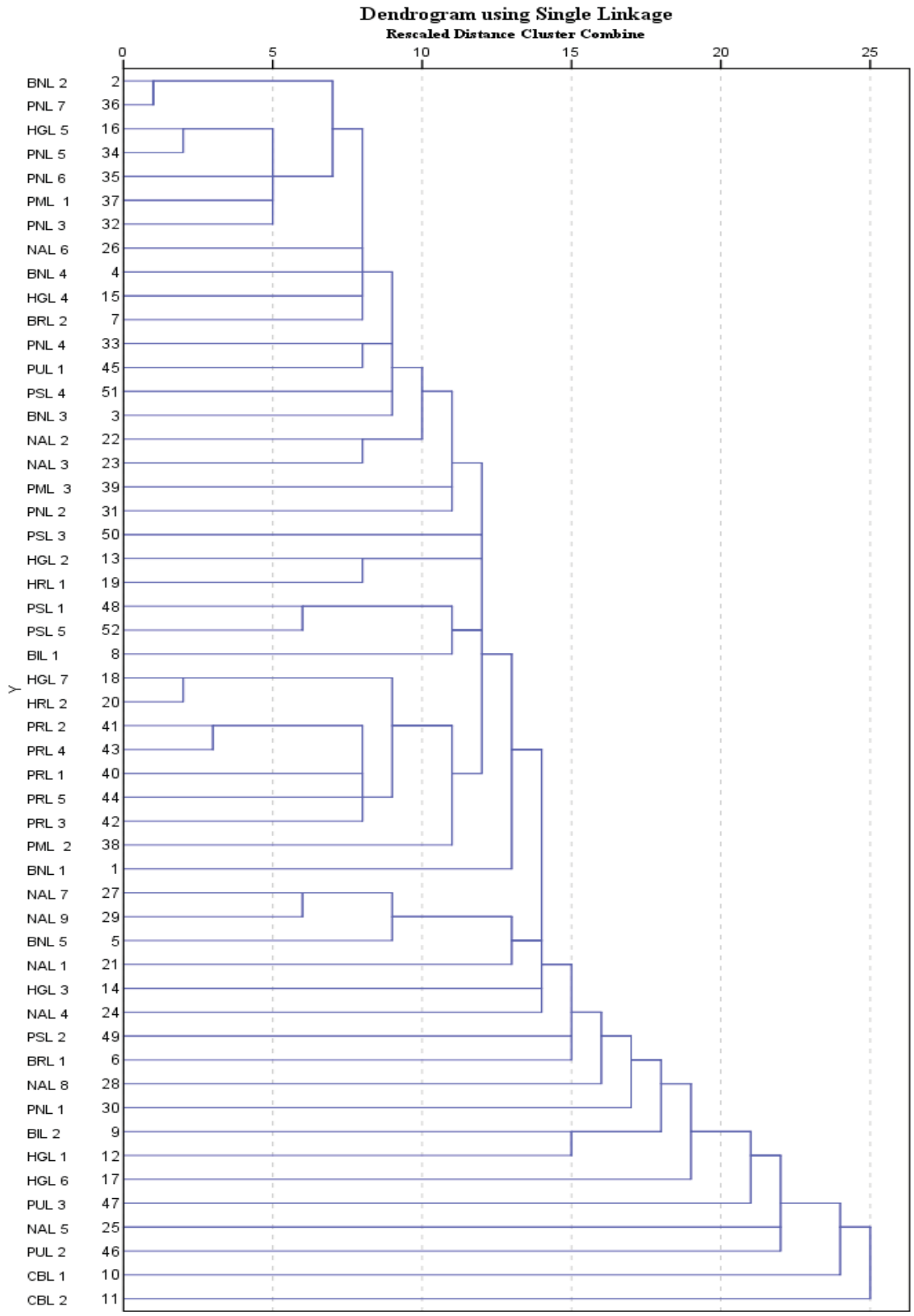

Fig. 1. Dendrogram (by single linkage) of different lemon genotypes using quantitative characters

Table 4. Component matrix resulted by PCA for quantitative characters of lemon

Variables	Components matrix						
	F1	F2	F3	F4	F5	F6	F7
Fruit weight	**0.810**	0.447	0.071	0.067	0.133	-0.002	0.087
Fruit diameter	**0.835**	-0.008	0.172	0.197	-0.048	0.144	-0.197
Fruit length	**0.677**	0.488	-0.168	-0.120	-0.005	-0.064	0.168
Oil glands/cm^2	0.018	**0.602**	-0.174	0.149	0.479	-0.251	-0.039
Rind thickness	**0.518**	-0.130	-0.025	0.008	-0.505	0.297	-0.289
Number of segments	0.391	-0.103	-0.269	**0.464**	0.286	0.349	-0.298
Vesicle length	0.262	0.424	0.261	-0.228	0.365	**0.464**	0.034
Juice weight	**0.749**	0.443	-0.108	-0.185	-0.148	-0.268	-0.123
Juice volume	**0.753**	0.424	-0.149	-0.164	-0.142	-0.260	-0.131
Juice percentage	**-0.532**	-0.150	-0.164	-0.274	-0.302	-0.224	-0.373
Number of Seed	**0.678**	-0.475	0.185	0.109	-0.043	0.040	-0.237
Seed weight	0.405	-0.288	**0.501**	0.138	-0.075	-0.088	0.416
Seed length	0.546	**-0.574**	0.188	0.189	0.093	-0.191	0.188
Seed width	**0.553**	-0.373	0.332	0.226	-0.178	-0.191	0.141
Acidity	-0.279	0.241	**0.759**	0.076	0.085	-0.002	-0.100
pH	0.128	-0.010	-0.254	-0.512	-0.181	0.093	**0.574**
TSS	0.125	-0.383	-0.184	0.145	**0.624**	-0.297	-0.097
TSS: acid	0.288	-0.468	**-0.669**	-0.003	0.337	-0.179	0.097
Ascorbic acid	0.028	-0.111	-0.284	0.119	0.210	**0.599**	0.190
Reducing Sugars	0.206	-0.034	**-0.584**	0.219	-0.511	0.097	0.066
Total Sugars	-0.262	0.321	-0.1624	**0.811**	-0.249	-0.058	0.158
Non Reducing Sugars	-0.355	0.353	-0.0104	**0.772**	-0.146	-0.102	0.145
Eigen value	5.378	2.854	2.302	2.208	1.836	1.300	1.154
Variability (%)	24.447	12.976	10.465	10.040	8.346	5.909	5.246
Cumulative %	24.447	37.424	47.889	57.930	66.276	72.185	77.432

Biplot analysis (Fig. 2 and Fig.3) revealed that genotypes remained in the 1st quadrant of scoring plot (HGL 6, PSL 5, HGL 1, BIL 2, NAL 5, PSL 1, HGL 3, BRL 1 etc.) had obviously higher values of characters loaded in 1st quadrant of loading plot (oil glands/cm^2, vesicle length, fruit length, juice weight, juice volume, fruit weight). Again, 2nd quadrant revealed that genotypes confined in this quadrant (CBL 1, PRL 2, PRL 3, HRL 2, NAL 8, PML 2, HGL 7, PNL 5 etc.) possessed higher values of 3 characters (total sugars, non reducing sugars, acidity). Similarly genotypes (PML 3, PUL 2, HGL 4, PNL 1, NAL 9, NAL 1, BRL 2, BNL 4, PSL 2 etc.) remained in the 3rd quadrant of biplot, contained higher juice percentage and genotypes of 4th quadrant of biplot (NAL 1, NAL 3, PNL 4, HGL 2, BNL 3, PSL 4, PUL 3, BNL 5 etc.) showed higher reducing sugars, pH, ascorbic acid, rind thickness, fruit diameter, number of segments, seed weight, seed width, number of seed, seed length, TSS, TSS: Acid.

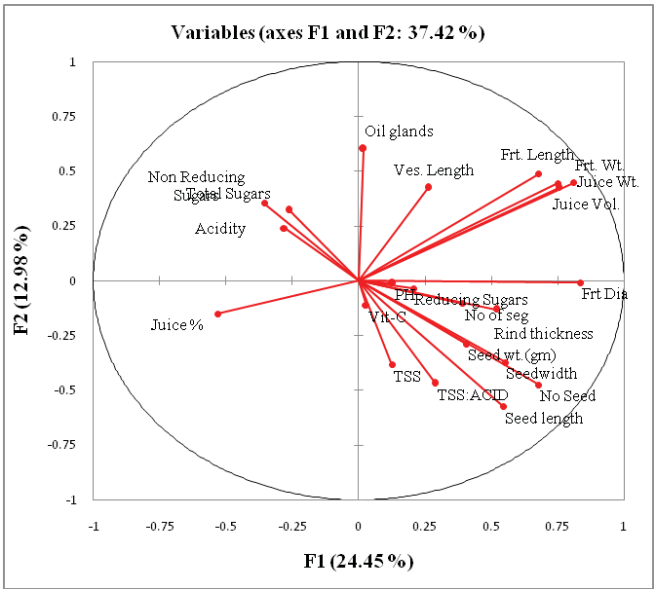

Fig. 2. Loading biplot of PCA (F1 Vs F2) for quantitative characters of lemon

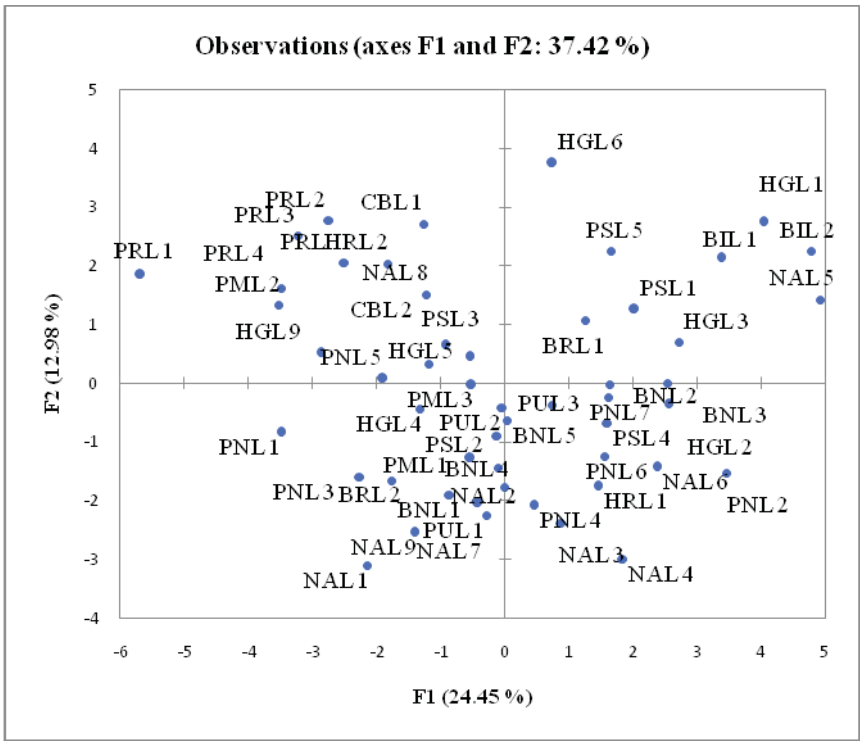

Fig. 3. Scoring biplot of PCA (F1 Vs F2) for lemon genotypes based on quantitative characters

Conclusion

From the above results, it is concluded that there is a profound diversity among acid lime collections and few genotypes may be exploited for various attributes based on consumers acceptance. Few of the genotypes viz., PNL1, PNL3, BRL2, PML2, HGL9, NAL4, HGL1, BIL2, NAL5, PUL3, BNL5, PNL7 etc.may be exploited as breeding material for development of improved varieties in lemon.

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RESEARCH ARTICLE

Elucidating the Response of Rice Genotypes to Iron Toxicity through Hydroponics

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Iron (Fe) toxicity is a major abiotic stress that impedes rice cultivation in many lowland environments worldwide. Although several cultural and management practices are advocated to address this problem, the best and most economic approach to combat this issue is the use of tolerant cultivars. The present study was aimed at elucidating the response of rice genotypes, including local landraces and released varieties to iron toxicity by screening seedling growth in a hydroponic solution containing varying levels of iron. The increasing concentration of iron had significant effect on all characters studied and the severity of symptoms. Significant difference was also observed for concentration × variety interaction for all the characters studied except root and shoot length of the seedlings. Increase in root number was observed at 600 ppm, indicating the repair mechanism of the plants against edaphic stress beyond the threshold level. Varsha, a mid-early, high yielding red-kernelled rice variety and Chuvannamodan, an indigenous landrace of Kerala, showed tolerance to high concentration of iron (> 600ppm), among the ten genotypes screened. It can be inferred that evaluation of genotypes at 600 to 800ppm concentration of iron in a hydroponic solution is a quick and efficient methodology to delineate the tolerance of rice genotypes to iron toxicity.

Key Words: Abiotic stress, Hydroponics, Iron toxicity, *Oryza sativa*, Rice

Introduction

Rice (*Oryza sativa* L.) cultivation is facing a multitude of problems including abiotic as well as biotic stresses. In recent times, abiotic stresses affecting the crop growth are on the rise owing to the changing climatic conditions, the consequences on productivity being either direct or indirect. Abiotic stresses include drought, waterlogging/ flood, salinity, nutrient deficiency/toxicity etc.. Iron (Fe) toxicity is a major stress to rice cultivation in many lowland environments worldwide (Asch *et al*., 2005). It often occurs in rice grown in submerged paddy fields with low pH, leading to dramatic increase in ferrous ion concentration, disrupting cell homeostasis and impairing growth and yield (Aung and Masuda, 2020). The typical symptoms are generally manifested as tiny brown spots starting from the tips and spreading towards the base of the lower leaves (Doberman and Fairhurst, 2000). Subsequently, the whole leaf turns yellowish or orange to brown. Growth and tillering are greatly affected and the root system is coarse, scanty and dark brown. The yield is reportedly reduced by 12-100%, depending on the severity of toxicity and the tolerance of the rice cultivars (Benckiser *et al*., 1982; Audebert and Sahrawat, 2000).

Many cultural practices are adopted to ameliorate iron toxic soil conditions. It includes trenching around the fields, application of dolomite, lime and chalk, and recurrent draining-off of accumulated irrigation water after submersion etc.. Rice plants have developed physiological avoidance and/or tolerance mechanisms to survive under Fe-toxic conditions (Nozoe *et al*., 2008). Molecular studies have shown that there are four defence mechanisms adopted by the rice plants to mitigate the iron toxicity. They include either iron exclusion by suppressing the genes involved in Fe uptake and translocation, or by retaining the excess Fe in the root system itself rather than by translocation to shoots or by compartmentalisation of Fe in the shoot by regulating the vascular Fe transport or by detoxifying reactive oxygen species produced in the plant system in response to Fe toxicity (Aung and Masuda, 2020). It has been reported that an iron tolerant variety absorbs

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less Fe or transports less from roots to leaves, indicating the presence of physiological avoidance mechanisms (Tadano, 1975; Audebert and Sahrawat, 2000). However, this problem can be best addressed by identifying and developing tolerant cultivars. Wide inter-varietal variability of iron toxicity tolerance in rice have been reported by Gunawardena *et al*. (1982) and Mohanty and Panda (1991). Thus, exploitation of this variability in iron toxicity reaction among genotypes while combating the stress is realised to be the sustainable and cheap alternative. Heritability of this trait has been detailed by Abifarin (1985) and Wu *et al*. (1997). The circumstances thus warrant, adoption of tolerant genotypes adapted to the specific growing environment, to avoid yield losses. Three approaches were in vogue to screen rice varieties for iron toxicity tolerance viz., field trials, pot trials and hydroponics trials. Shimuzu *et al*. (2005) advocated the use of mass screening using culture solution as the most effective method to identify iron toxicity tolerant cultivars as this allows stress conditions to be efficient, easily controlled and reproducible.

In India, iron toxicity is reported especially in Kerala, Orissa, West Bengal and Andaman Islands (Ponnamperuma, 1978; Sahrawat and Singh, 1998 and Mandal *et al*., 2004). Kerala has agro-ecological zones with wide variability in rice genetic resources and kernelled bold rice varieties over white kernelled ones by the local population (Suma *et al*., 2018). Hence, breeding for rice varieties tolerant to stress conditions like Fe toxicity require resistant genes from native landraces. There are reports that most of the modern semi-dwarf, high-yielding rice cultivars were sensitive to iron toxicity (Wade *et al.*, 1999). Studies by Benckiser *et al*. (1984) and Onaga *et al*. (2013) demonstrated that some traditional cultivars have better tolerance to iron toxicity. Therefore, the present study was aimed at elucidating the response of rice genotypes to iron toxicity under varying levels of iron stress in order to deduce the threshold toxicity level of iron that can help differentiate between tolerant and susceptible genotypes. Further, the attempt could resize the number of accessions to be screened for tolerance in elaborate field experiments to a more manageable level.

Materials and Methods

Ten rice genotypes procured from the germplasm collections maintained at the College of Agriculture, Kerala Agricultural University, Thrissur, Kerala, India constituted the experimental material. Thekkencheera and Chuvannamodan were the landraces and Ptb 40 (Matta Triveni), Ptb 49 (Kairali), Ptb 56 (Varsha), Ptb 60 (Vaisakh), Mo 16 (Uma), Mahsuri, and CR 1009 (Ponmani), were the high yielding varieties. Jarwa, a variety from Andaman & Nicobar Islands was also included as a check to elucidate the response. The experiment was laid out in factorial design with the treatments arranged in a completely randomized fashion with two replications, the 10 genotypes and 5 levels of iron. Varying levels of Fe concentrations (0, 200, 400, 600 and 800 ppm) were imposed. Six days old germinated seeds were sown in holes of a polystyrene plate covered at the bottom with nylon net (Fig. 1). The polystyrene plate was floated on a plastic tray filled with normal-strength Yoshida's solution (Yoshida *et al*., 1976), 10 l in each tray, including 1.78 mM silicon (Si) at pH 5.0. The seedlings were exposed to varying iron concentrations by the addition of different concentrations of ferrous Fe with 0.09 mM Fe-EDTA, following the procedure recommended by Shimuzu *et al*. (2005).

The culture solution was renewed weekly, and adjusted to pH 5.0 twice a day with 1N NaOH/HCl. The seedlings were grown until 15 days (3-4 leaf stage), and Fe toxicity responses were scored by subjective visual assessment of bronzing symptoms on developed leaves by following symptom scoring system adopted by Shimuzu *et al*. (2005), modified as shown in Table 1. Subsequently, the plants were harvested for measuring growth attributes like root length (cm), shoot length (cm), number of roots, iron reversibly adsorbed on root zone (%), root dry weight (g) and shoot dry weight (g). Shoot and root length were measured from the base of the culm to the longest leaf and from culm base to longest root, respectively. The collected plant roots

Table 1. Bronzing score classified into nine ranks according to inspection of leaf blades

Score	Leaf order			
	1st	2nd	3rd	4th
1	N	N	N	N
2	T	N	N	N
3	T	T	N	N
4	P	T	N	N
5	P	T	T	N
6	P	P	T	N
7	D	P	P/T	T/N
8	D	D	P/T	T
9	D	D	D/P	No Leaf/GS

N: normal; T: discoloration of leaf tip; P: partly discoloured; D: rolled or dead leaf; GS: Growth stunted

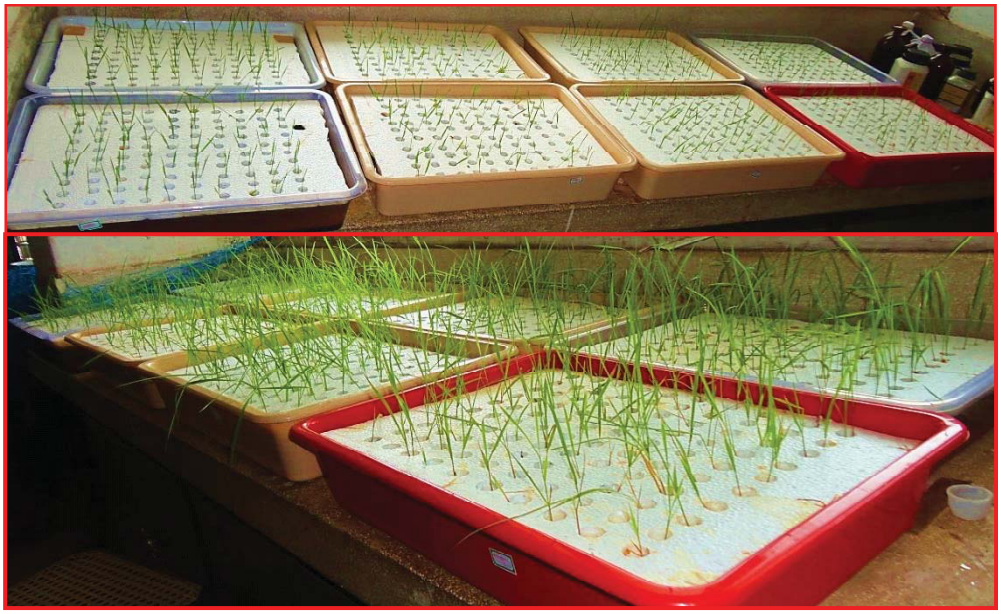

Fig. 1. Seedlings grown in polystyrene plates under different Fe concentrations

were washed thoroughly with deionized water without dislodging the iron plaques on the root surface. The roots were then immersed in 25 ml 0.01M calcium chloride solution for 5 min to release the adsorbed iron. Calcium chloride solution containing iron was treated with concentrated hydrochloric acid to dissolve the ferric iron and 5 ml of this solution was made up to 50 ml and the Fe content was estimated using Atomic Absorption Spectrophotometer (Model: Analyst-400 Perkin-Elmer). The iron adsorbed on the roots was correlated with the performance of different varieties under varying concentrations of iron. Further, root and shoot samples were wrapped in Aluminium foil separately and oven dried at 80 ^{o}C for 48 h for measuring shoot and root dry weight separately. Statistical analysis was done using SAS (9.3 version).

Results and Discussion

Analysis of variance indicated that the increasing Fe concentration had significant effect on all characters studied (Table 2). Significant difference was also observed for concentration × variety interaction for all the characters except root and shoot length. Grouping of treatments based on the significant difference of mean values of the characters studied (Table 3) indicated that, irrespective of the genotypes there was significant reduction in root and shoot length beyond 400 ppm. It was earlier reported that excess iron can lead to reduction in shoot length, which can be a useful characteristic for screening of tolerant genotypes (Bresolin *et al*., 2019). Ferrous toxicity inhibits cell division and elongation of the primary roots and subsequently the growth of lateral roots (Li *et al*., 2016). However, there was an increase in root number at 400 ppm, indicating the triggering of inherent defence mechanism in plants against a stress beyond a threshold level. Shoot length difference was not significant beyond 600 and 800 ppm concentrations, indicating that 600 ppm is the threshold level above which the effect of toxicity was more pronounced. Most of the varieties studied exhibited an increase in growth parameters up to 200 ppm revealing the beneficial effect of iron availability and uptake by the plants up to this concentration. This is consistent with the earlier reports that, adequate Fe concentration in the plant tissue is

Table.2. Analysis of Variance for effect of varying Fe concentrations on growth attributes of ten selected rice varieties

		Mean square values					
	Df	Root length (cm)	Shoot length (cm)	No. of roots	Fe adsorbed on roots (mgL^{-1})	Root dry weight (g)	Shoot dry weight (g)
Concentration	4	11.715**	102.356**	10.465**	39.199**	.00128**	.00195**
Variety	9	8.286**	65.467**	5.690**	2.113**	.00229**	.00144**
Concentration × variety interaction	36	0.454	3.014	1.4816**	0.833**	.000017**	.000095**

**significantly different at 1% level

Table 3. Grouping of treatments based on the significant difference of mean values of the characters studied

Treatment	Root length (cm)		Shoot length (cm)		No.of roots		Fe adsorbed on roots (mgL^{-1})		Root dry weight (g)		Shoot dry weight (g)	
	Mean	Letter group	Mean	Letter group	Mean	Letter group	Mean	Letter group	Mean	Letter group	Mean	Letter group
Control	6.069	AB	18.35	A	5.7	B	0.57	E	0.0805	C	0.0795	A
200ppm	6.579	A	18.32	A	5.55	BC	1.51	D	0.09	B	0.067	B
400ppm	6.196	A	17.60	A	6.60	A	2.76	C	0.103	A	0.0635	B
600ppm	5.439	B	14.59	B	5.15	C	3.593	B	0.091	B	0.0575	C
800ppm	4.626	C	13.53	B	4.65	D	3.868	A	0.09	B	0.054	C

in the range of 70-300 mg kg^{-1} (Wells *et al*., 1993). Iron deficiency or toxicity occurs at concentrations below or above this sufficiency range (Fageria *et al*., 1981). Changes in shoot length, root length and nutrient accumulation in tissues during early developmental stages have been reported to constitute an objective form of evaluation that can be used in conjunction with bronzing scores (Bresolin *et al*., 2019).

The severity of symptoms increased linearly with the increase in Fe concentration (Fig. 2). However, the varieties expressed varying degrees of tolerance to different Fe concentrations, providing scope for breeding for iron toxicity tolerance. Bronzing score did not increase in variety Kairali from 200 to 600 ppm concentration. Variety Varsha expressed the lowest bronzing score (4.00) at 800 ppm. Maximum scoring was exhibited by the variety Mahsuri.

Varietal Response to Fe Toxicity

Root and shoot elongation was affected significantly beyond 400 ppm, except in Kairali and Vaishak, which exhibited more root elongation at 200 ppm (Table 4a). All the growth attributes were severely affected at 800 ppm. Chuvannamodan, an upland landrace showed increased root length and Varsha, showed an increase in shoot length at 600 ppm than the immediate lower concentration (400 ppm). Kuraev (1966) reported that the initial toxic effect of high iron inhibits root development, and this was more pronounced at higher iron concentrations (200 ppm), which may have been due to possible toxicity mechanisms such as the iron-induced production of superoxide (O_2^-). However, Kairali, Jarwa, Vaishak,

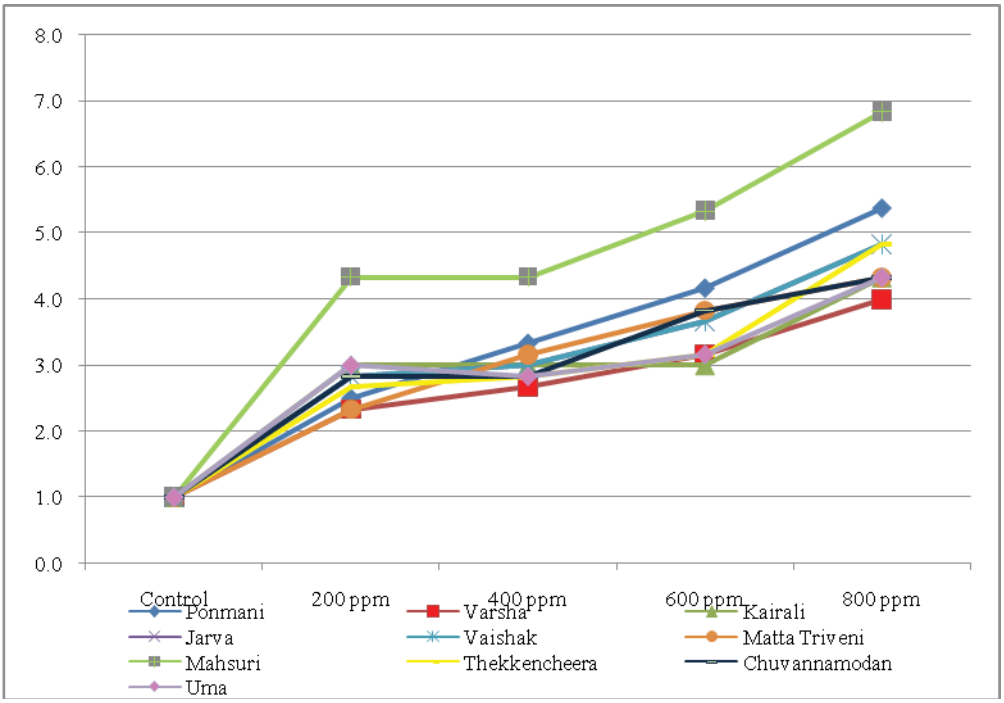

Fig. 2. Bronzing score for ten rice cultivars at varying Fe concentrations

Table 4a. Effect of different iron concentration on root length, shoot length and number of roots in 10 varieties of rice

Varieties	Root Length (cm)					Shoot Length (cm)					Number of roots				
	Control	200 ppm	400 ppm	600 ppm	800 ppm	Control	200 ppm	400 ppm	600 ppm	800 ppm	Control	200 ppm	400 ppm	600 ppm	800 ppm
Ponmani	4.33	5.07	4.67	4.45	3.75	15.83	14.95	15.67	12.33	11.91	4.50	7.00	5.50	5.00	5.00
Varsha	6.66	6.80	6.00	5.50	4.91	22.08	21.83	24.16	19.03	17.25	6.50	5.50	6.00	5.50	5.50
Kairali	6.42	6.38	6.66	5.33	5.00	19.17	18.41	17.10	14.08	13.98	6.50	5.50	7.50	7.00	5.00
Jarwa	4.58	6.18	5.97	5.17	4.60	16.58	19.52	20.00	15.91	16.42	4.00	4.00	7.00	4.00	4.00
Vaishak	5.50	5.75	5.93	4.25	3.50	16.92	15.20	13.93	12.17	11.66	4.50	4.50	6.50	5.00	3.00
MT	7.55	7.45	6.93	5.83	4.92	19.90	19.10	18.45	15.58	14.25	7.50	6.50	7.00	4.00	4.00
Mahsuri	6.28	7.15	5.90	5.67	5.33	16.68	16.06	13.93	11.73	10.83	6.00	4.50	4.50	3.50	3.50
Thekkencheera	5.25	5.78	5.16	4.37	3.67	18.27	22.65	18.35	15.35	14.17	6.00	6.00	7.50	6.50	5.50
Chuvannamodan	7.57	7.85	7.83	8.62	5.80	22.62	21.20	21.20	16.93	13.85	5.50	4.50	7.50	5.00	5.00
Uma	6.57	7.40	6.93	5.23	4.80	15.52	14.33	13.22	12.78	10.95	6.00	7.50	7.00	6.00	6.00

Table 4b.Effect of different iron concentration on iron adsorbed on roots , dryweight of roots and dry weight of shoots in 10 varieties of rice

Varieties	Iron adsorbed on roots (mg L^{-1})					Dry weight of roots (g)					Dry weight of shoots (g)				
	Control	200 ppm	400 ppm	600 ppm	800 ppm	Control	200 ppm	400 ppm	600 ppm	800 ppm	Control	200 ppm	400 ppm	600 ppm	800 ppm
Ponmani	0.979	1.699	2.089	3.185	3.397	0.07	0.09	0.09	0.09	0.09	0.09	0.08	0.07	0.07	0.05
Varsha	0.906	1.942	2.144	3.508	3.626	0.10	0.10	0.12	0.10	0.10	0.11	0.08	0.09	0.07	0.07
Kairali	0.140	1.469	4.098	2.392	2.573	0.09	0.09	0.12	0.11	0.11	0.09	0.08	0.06	0.06	0.06
Jarwa	0.816	1.697	2.753	3.181	3.370	0.08	0.08	0.09	0.09	0.08	0.07	0.07	0.06	0.05	0.05
Vaishak	0.718	1.219	1.661	2.843	3.222	0.08	0.11	0.12	0.09	0.09	0.07	0.06	0.06	0.06	0.06
MT	0.206	1.499	4.062	4.296	4.537	0.09	0.10	0.12	0.11	0.11	0.10	0.09	0.08	0.06	0.06
Mahsuri	0.470	1.498	3.216	4.074	4.372	0.07	0.06	0.06	0.06	0.06	0.06	0.03	0.06	0.04	0.03
Thekkencheera	0.881	1.395	3.268	5.817	5.968	0.08	0.08	0.10	0.09	0.09	0.08	0.07	0.05	0.06	0.06
Chuvannamodan	0.515	1.718	1.986	3.334	3.755	0.08	0.10	0.14	0.10	0.10	0.09	0.09	0.08	0.07	0.07
Uma	0.059	0.930	2.289	3.306	3.859	0.11	0.12	0.11	0.11	0.11	0.08	0.06	0.07	0.06	0.06

Thekkencheera and Chuvannamodan exhibited increased root number at 400ppm, a mechanism to overcome the toxicity. These results corroborated with the results of Reddy *et al*. (2019) that higher values of number of fresh roots, iron adsorbed on root surface and shoot weight are mechanisms observed in rice plants to overcome iron toxicity. Onyango *et al.* (2019) also reported that the number of new lateral roots, under both moderate and severe stress levels, increased in stressed plants of all the varieties compared with control plants. Root architectural traits like formation of an aerenchyma and a large number of lateral fine roots, facilitate the diffusion of oxygen into the rhizosphere, thereby increasing the redox potential above the threshold for Fe oxidation (Wu *et al.,* 2014). Fe exclusion, a root-based tolerant mechanism through inhibition of root-Fe uptake, is achieved by forming Fe plaques on the root surface due to Fe^{3+} precipitation, Fe^{3+} in turn formed by rhizospheric oxidation of Fe^{2+} (Becker and Asch, 2005). In the present study also, the excluded Fe estimated as Fe reversibly adsorbed on roots have increased significantly above 400 ppm. Matta Triveni (MT) showed higher magnitude of Fe exclusion at 400 ppm (4.06 mg/g) and beyond. It is reported that iron uptake and transport related genes such as *OsIRT1*, *OsIRT2*, *OsYSL2*, *OsYSL15*, and *OsNRAMP1* are highly suppressed in roots under Fe toxic conditions (Quinet *et al*., 2012; Finatto *et al*., 2015; Aung *et al.*, 2018). The growth of the plants was affected severely as indicated by the reduction in root and shoot dry weight. The trend of reduction in shoot dry weight with increasing Fe toxicity also supports the results of Nugraha *et al*. (2016). The variety Mahsuri exhibited least tolerance though this variety was acclaimed to be an Fe toxicity tolerant variety (Nugraha *et al*., 2016). This variety recorded a decrease in root weight with slight increase in iron concentration (200 ppm) and least value for both dry root and shoot weight at 800 ppm. In a similar study using four levels of lime and three levels of fertilizer using three rice varieties at RARS, Kumarakom, Kerala, it was reported that integration of genetic tolerance and nutrition management could reduce the intensity of iron toxicity in acid sulfate soils (Thampatti *et al*. 2005).

Varsha, a high yielding variety and Chuvannamodan, an upland landrace of Kerala exhibited comparative tolerance to iron toxicity at higher concentrations (600 and 800 ppm) over all other varieties. This is evident from the high values for length of shoot and root and dry weight of root and shoot. High amount of adsorbed iron on root surface (5.96 mg/L) in variety Thekkencheera at higher Fe concentration reveals that this variety has a higher capability of iron exclusion mechanism to combat iron toxicity stress. This may be due to increased adaptability of the variety indigenous to the problematic zone. However, this requires further confirmation. In addition, significant tolerance level was expressed by the varieties Uma, Mattatriveni and Kairali.

Conclusion

The major problem in field screening large numbers of genotypes for tolerance to Fe-toxic conditions is to provide sufficiently homogenous and elevated Fe levels in the soil, to elucidate comparable stress levels to all materials. Screening genotypes in hydroponics can help negate this problem. Evaluation of genotypes at 600 to 800ppm concentration of iron would help identifying the difference in varietal reaction to iron at toxic levels. The varieties differed in their response to varying concentrations of Fe. Varsha and Chuvannamodan showed tolerance to high concentration of iron (> 600ppm) among the ten varieties studied. In a previous study conducted by Suma *et al*. (2018), the high yielding variety Varsha had registered high value for elongation ratio and volume expansion ratios of grains on cooking in comparison to other landraces and varieties of rice. The iron absorbed by the plant parts *viz*., roots and older leaves and molecular studiesare needed to understand the mechanism of defence adopted by individual varieties to combat iron toxicity stress. A thorough evaluation of germplasm field testing in hotspot areas after an initial screening through hydroponics will help elucidate the genotypes with high tolerance to this abiotic stress.

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RESEARCH ARTICLE

Orchidopedia App –A Tool for Exploration and Collection of Orchid Species

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Identification is a major bottleneck during germplasm exploration and collection of wild species of orchids. An application named "*Orchidopedia*" is developed on the basis of database of wild orchids. This app is available offline and works independently. It has vast information for about 172 species belonging to 56 genera of wild orchid species. Orchidopedia is an android based mobile application that focuses on orchid species of the North-Eastern states of India. The app is user friendly and can be freely downloaded from Google Play Store (https://play.google.com/store/apps/details?id=nrco.orchidopedia&hl=en) and also from the website of ICAR-National Research Centre for Orchids, Pakyong, Sikkim for researchers, botanists, orchid growers/lovers, stakeholders and entrepreneurs. The application is built on minimum sdk version 4.1 and target sdk version 10.0 android smartphones.

Key Words: Android Application, Android studio and Orchids, Software development kit, SQLite DB

Introduction

Orchids are the most fascinating flowering plants on the earth and represent incredible range of diversity in shape, size, colour, structure and appearance (Singh *et al.*, 2019a). Orchidaceae is the second largest family of flowering plants, represented by 22,500 species in 800 genera distributed throughout the world (Mabberley, 2008; Singh *et al.,* 2019a; Pamarthi *et al*., 2019). It is estimated that about 1263 species (155 genera) of orchids are found in India, with the Himalayas as their primary home and others scattered in Eastern and Western Ghats; among the 1,263 species, nearly 311 are endemic species (Singh *et al.,* 2019a). The distribution of orchids species in different regions of India is as follows *viz.,* North-Western Himalayas (200 species), North-Eastern India (800 species), the Western Ghats (300 species). Due to its peculiar gradient and varied climatic conditions, North-Eastern India harbours the largest group of temperate, sub-tropical orchids. The species diversity is highest in Arunachal Pradesh, with 612 species, and Sikkim has the richest and most diversified with 560 species of orchids (Singh *et al*., 2019a).

Some of the orchids are used as medicine, food and integral part of socio-cultural events (Medhi and Chakrabarti, 2009 and Meitei et *al*., 2019). Orchids are also used for like preparation of traditional artefacts made from dried leaves of *Cymbidium* in Sikkim (Singh *et al.,* 2019b). The wild species of orchids were used as progenitors in breeding programmes and intra and inter-sectional compatibility found at the species level for development in modern hybrids (Devadas *et al.,* 2016). Pamarthi *et al.* (2019) reported 90 wild species of orchids having potential breeding value and utilized in the development of hybrids or improved lines. The indigenous orchid species *viz*., *C. eburneum*, *C. erythraeum*, *C. hookerianum*, *C. iridiodes, Paphiopedilum druryi, Vanda coerulea* and *V. tessellata* are widely utilized in different crop improvement programmes. Some of the native species of orchids have a significant role in developing the particular trait or character modern hybrids or varieties like *Cymbidium iridiodes,* a native scented species used as a male parent in developing two scented lines (Pamarthi *et al.,* 2019). Devadas *et al.* (2019) attempted to develop the new hybrid having potential ornamental value by using native *Phaius* species (*P. flavus*x *P. tankervilleae*).

With the availability of significant diversity in the morphology of orchid species, identification of wild

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species is a major challenge during germplasm expedition. Tomar *et al.* (2019) developed the "Orchid farming" android application recently for the cultivation of five genera of orchids which gives benefit to orchid growers and farmers. Enormous information about orchids are compiled in a single android application compatible with all smartphones. In continuation, the ICAR-National Research Centre for Orchids, Pakyong, Sikkim has developed an Android-based mobile application named 'Orchidopedia' which provides the quality and collective information on every aspect of orchid species according to their genera. The android app can be downloaded from the Google Play Store that contains 172 species (56 genera) of orchids of the Himalayan region. The work is initiated after the compilation of information of important orchid species.

Further, the application data is updated as per current research data procuring. This android application provides details about classification, distribution, morphological description of orchids. The present generation is well versed with the usage of Android smartphone for their standard requirement. This application support target SDK version Android 4.1 (API level 16) to Android 10.0 (API level 30) mobile phones, which cover almost 96.4% of Android smartphones being used in our country (Tomar *et al.,* 2019). The details of the Orchidiopedia application are given in Fig. 1. Mobile applications allow companies to transform every sector, and now it is marching towards the agricultural industry. Android are less costly than IoS, and a wide range of android smartphones are available in the market for users. Because of the merits of the android, this simple, offline and effective application is developed to transfer scientific information to the botanists, researchers, students and farmers.

Material and Methods

The android mobile application "Orchidopedia" was developed at the ICAR-NRC for Orchids, Pakyong, Sikkim, during 2019. It was built for encapsulating 172 species belonging to 56 genera (Table 1) and their respective species in one hand held device.The floristic nomenclature consultation of each species was confirmed from the online databases, namely, Govaerts (2012), Tropicos (2018), IPNI (2018), eFloras (2018), Plantlist (2019) and POWO (2020). Each species is provided with classification, habitat, distribution, morphological description, key characters, flowering

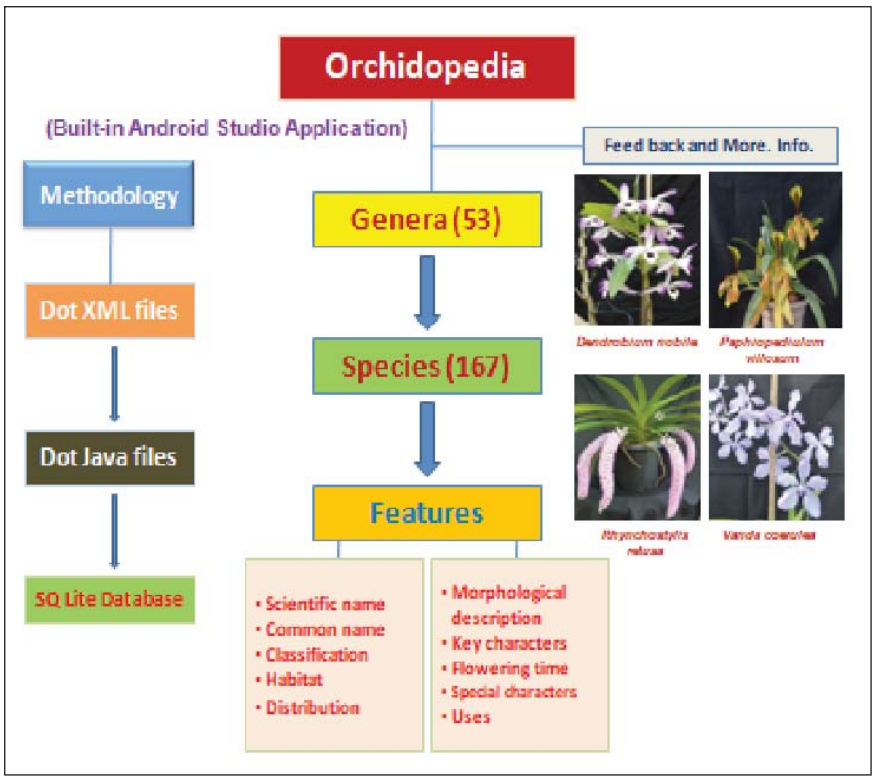

Fig. 1. Outline of the Flow Chart of 'Orchidopedia'

Table.1. List of Orchid species (Genus-Species) used for developing "*Orchidiopedia*" mobile application

S. No.	Genus	Number of species	Details of Species
1	*Acampe*	2	*A. praemorsa, A. rigida*
2	*Acanthephippium*	1	*A. sylhetense*
3	*Acrochaene*	1	*A. punctata*
4	*Aerides*	4	*A. crispa, A. multiflorum, A. odoratum, A. rosea*
5	*Agrostophyllum*	1	*A. brevipes*
6	*Anthogonium*	1	*A. gracile*
7	*Arachnis*	1	*A. labrosa*
8	*Arundina*	1	*A. graminifolia*
9	*Ascocentrum*	2	*A. ampullaceum, A. aurantiacum*
10	*Bulbophyllum*	11	*B. affine, B. careyanum, B. cauliflorum, B. crassipes, B. guttulatum, B. gymnopus, B. hirtum, B. leopardinum, B. odoratissimum, B. viridiflorum, B. wallichii*
11	*Calanthe*	5	*C. biloba, C. chloroleuca, C. herbacea, C. plantaginea, C. sylvatica*
12	*Cattleya*	1	*C. bowringiana*
13	*Ceratostylis*	1	*C. subulata*
14	*Chiloschista*	1	*C. parishii*
15	*Cleisostoma*	1	*C. linearilobatum*
16	*Coelogyne*	5	*C. corymbosa, C. cristata, C. fuscescens, C. nitida, C. Punctulata*
17	*Conchidium*	1	*C. muscicola*
18	*Cymbidium*	8	*C. aloifolium, C. dayanum, C. devonianum, C. eburneum, C. elegans, C. ensifolium, C. hookerianum, C. tigrinum*
19	*Dendrobium*	47	*D. aduncum, D. amoenum, D. anceps, D. aphyllum, D. aqueum, D. bensoniae, D. bicameratum, D. capillipes, D. cathcartii, D. chrysanthum, D. chrysotoxum, D. crepidatum, D. densiflorum, D. denudans, D. devonianum, D. eriiflorum, D. falconeri, D. farmeri, D. fimbriatum, D. formosum, D. gibsonii, D. heterocarpum, D. hookerianum, D. kingianum, D. lindleyi, D. lituiflorum, D. loddigesii, D. longicornu, D. macrostachyum, D. infundibulum, D. jenkinsii, D. moschatum, D. nanum, D. nobile, D. ochreatum, D. parishii, D. pendulum, D. porphyrochilum, D. praecinctum, D. primulinum, D. rotundatum, D. ruckeri, D. stuposum, D. terminale, D. thyrsiflorum, D. transparens, D. williamsonii*
20	*Diplocentrum*	1	*D. recurvum*
21	*Diplomeris*	1	*D. hirsuta*
22	*Epidendrum*	3	*E. ellipticum, E. radicans, E. xanthinum*
23	*Eria*	8	*E. coronaria, E. ferruginea, E. globulifera, E. javanica, E. lasiopetala, E. porteri, E. tomentosa, E. vittata*
24	*Eulophia*	1	*E. speciosa*
25	*Gastrochilus*	2	*G. acutifolius, G. dasypogon*
26	*Geodorum*	1	*G. densiflorum*
27	*Goodyera*	1	*G. procera*
28	*Herpysma*	1	*H. longicaulis*
29	*Hygrochilus*	1	*H. parishii*
30	*Liparis*	3	*L. manni, L. plantaginae, L.viridiflora*
31	*Lycaste*	1	*L. cruenta*
32	*Micropera*	2	*M. obtusa, M. pallida*
33	*Neogyna*	1	*N. gardneriana*
34	*Ornithochilus*	1	*O. difformis*
35	*Panisea*	2	*P. demissa, P. uniflora*
36	*Paphiopedilum*	6	*P. fairrieanum, P. hirsutissimum, P. insigne, P. spicerianum, P. venustum, P. villosum*
37	*Papilionanthe*	2	*P. teres, P. uniflora*
38	*Pelatantheria*	1	*P. insectifera*
39	*Phaius*	3	*P. flavus, P. mishmensis, P. tankervilleae*
40	*Phalaenopsis*	4	*P. deliciosa subsp. hookeriana, P. lobbii, P. mannii, P. taenialis*
41	*Pholidota*	3	*P. articulata, P. imbricata, P. Rubra*
42	*Phreatia*	1	*P. elegans*
43	*Pinalia*	2	*P. amica, P. Pumila*
44	*Pleione*	2	*P. maculata, P. praecox*
45	*Renanthera*	1	*R. imschootiana*
46	*Rhynchostylis*	1	*R. retusa*
47	*Satyrium*	1	*S. nepalense*
48	*Schoenorchis*	1	*S. gemmata*
49	*Smitinandia*	1	*S. micrantha*
50	*Sunipia*	4	*S. bicolor, S. cirrhata, S. intermedia, S. scariosa*
51	*Thrixspermum*	1	*T. musciflorum*
52	*Thunia*	3	*T. alba, T. alba var. bracteata, T. marshalliana*
53	*Uncifera*	1	*U. obtusifolia*
54	*Vanda*	7	*V. alpina, V. coerulea, V. cristata, V. pumila, V. stangeana, V. tessellata, V. testacea*
55	*Vanilla*	1	*V. planifolia*
56	*Zygopetalum*	1	*Z. maculatum*

time, special characters and uses. The original data for each of the sections were sourced from institute reports like annual reports (ICAR-NRCO, 2017; 2018; 2019) and technical bulletins and arranged as per the application's data structure. There are different slabs in each genus that provide information about the genus and their related species. Users can also identify Orchid's species by the high-resolution images and while there is an option of selecting genus and species in the application. The application"Orchidopedia" is built-in Android Studio applicationan Open Source software. Java Core Library provided most functions of the application and the Gradle-advanced build toolkit, to automate and manage the build process leads to the define flexible custom build configurations (Developers Android, 2020). For the creation of this application, two file formats were used. Firstly dot XML files that give design support of application and secondly, dot JAVA files that provide backend programming support. Default permissions, namely *"android.permission.INTERNET"* and *"android.permission.SEND_SMS"* had enabled in the AndroidManifest.xml file of the application, which users need to allow these permissions for using *Feedback* options. Due to the installation of SQLite Database in user's smartphone the application fetches or retrieves massive data of orchid in offline mode also (Fig. 2). This is the inbuilt software of the android operating system which is compatible with the application and store information that the application needs to display to users. It is just like a small pack of databases connected with application. When the application is installed from Google Play Store, it gets attached and provides offline service to the users. In SQLite Database, data are sub-divided into attributes which helps to split according to the user's need. As per user reliability, the hide-unhide feature is enabled because of the enormous data displayed on single screen users who can easily select information according to their needs. The application size is not more than 18 MB which will work on any device without acquiring much space.

Results and Discussion

The genus *Dendrobium* represented the highest number of species (48), followed by *Bulbophyllum* (11), *Cymbidium* (8), *Eria* (8), *Paphiopedilum* (7), *Pinalia* (7), *Vanda* (7), *Pholidota* (6), *Coelogyne* (5), *Calanthe* (5), *Aerides* (4), *Liparis* (4), *Papilonanthe* (4), *Phalaenopsis* (4), *Pleione* (4), *Sunipia* (4), *Liparis* (3), *Micropera* (3), *Phaius* (3), *Thunia* (3), *Gastrochilus* (2), *Panisea* (2), *Phreatia* (2), *Acampe* (2), *Ascocentrum* (2); and 28 genera represented single species. Each species contain ten specific features viz. taxonomical classification, habitat, distribution, flowering time, morphological description and economic importance. Each genus and species are arranged in alphabetical order so that the viewer can quickly check the species level information. Taxonomical classification of each species arranged from class to species level. Status of IUCN of each species is also provided to know the distribution of frequency level of each species. Morphological descriptions and key characters were given to understand the characters of the species. High resolution images also provided to each species for identification. Flowering time, habitat and distribution gives valuable information for explorers and collectors. This application also provides the economic usage of orchids to the users.

How to Use?

"Orchidopedia" app allows users to know information as per requirement in simple and straightforward ways and screenshots (Steps-6), which provide an example of functionality.

- Start the application after downloading from Google Play Store. In Fig. 3 splash screen displayed that will take time to load the SQLite Data with the application.
- As shown in Fig. 4, after full loading of application, a screen occurs that contains *Genera*, *More Information* and *Feedback* option.
- While clicking on the genera option, 53 genera unhide systematically, and user can select any genus as per their choice. For instance, in Fig. 5, we decided on *Dendrobium*.
- Each genus contains the name of its related single or multiple species. As shown in Fig. 5, *Dendrobium* genera listed out various species by their scientific name.
- While selecting *Dendrobium nobile* in Fig. 5, details of related species occurred with subheadings, for instance, common name, botanical name, image, tribe-sub tribe, morphological information, flowering time, etc.
- Users can easily hide/un-hide data as per their need. This functionality gives vital help while accessing particular species detail.
- *Feedback* includes email service and messaging

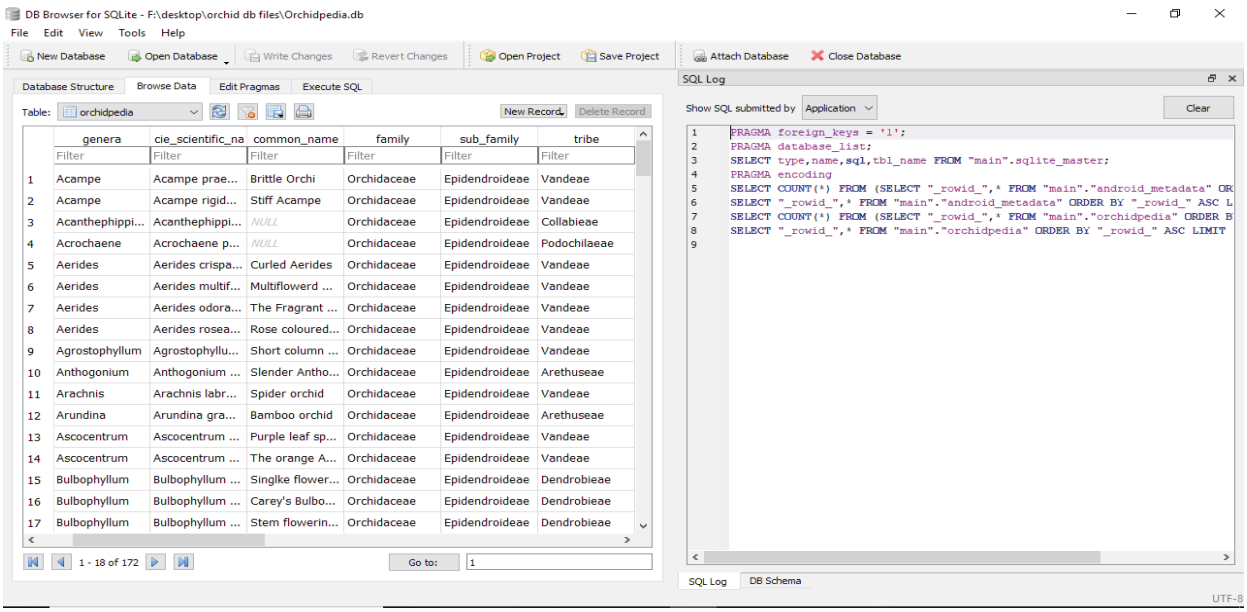

Fig. 2. In-built SQLite Database in browser

Fig. 3. Splash screen and home screen

Fig. 4. List of genera unhide after clicking genera option tool bar and select any species

Fig. 5. Information of selected species

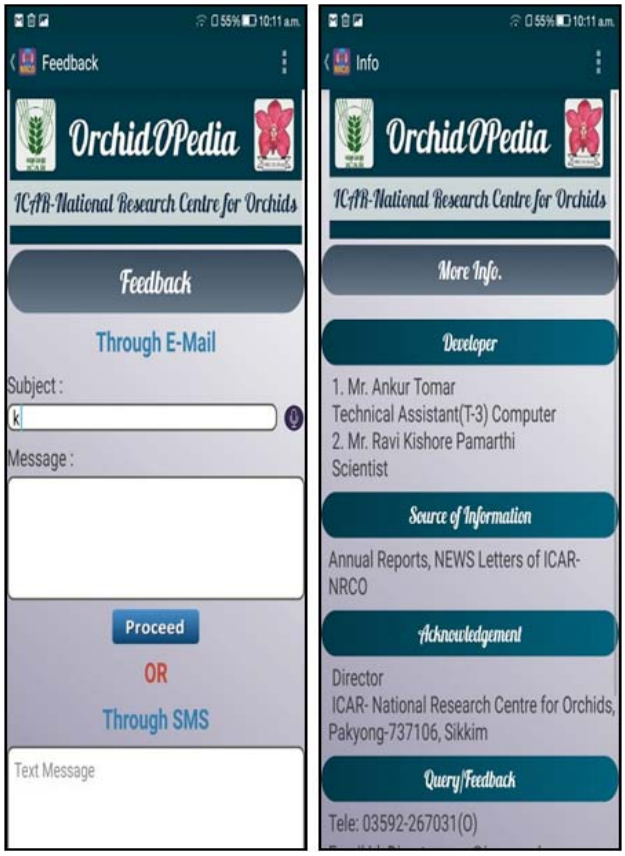

Fig. 6. Feedback form and more information

service that helps orchid growers communicate with us in respective areas (Fig. 6).

- *More Info* & *About Us* gives detail about the institute and their developer info which will assist the userin performing the desired work smoothly (Fig. 6).
- The *menu* option is also attached with the application that helps users can go directly to the main menu from any stage.

Conclusion

In India, most of the orchids are distributed in North-East India and some of the native genera like *Cymbidium, Paphiopedilum, Vanda, Arachnis* and *Dendrobium* are cultivated on a large scale for cut flower production. The *Cymbidium* is grown in NEH Region, mainly in Sikkim, Darjeeling hills, Arunachal Pradesh, Mizoram and Assam. By using this android application, explorers, collectors, orchid growers, students, researchers, botanists, horticulturists, and farmers across North-East India will surely identify wild species of orchids. This mobile app will be helpful in searching of information about wild species. Publication of applications under Google Play store available in public domain including the non-governmental organization, consumers and policymakers. It is a standalone application and freely available and is now in the english language option due to the management of vast orchid data. However, the option "*language selection*" is included as per users' requirement and their responses. Feedback and suggestions about the features and functionality of application have also provided in app. Orchid lovers have already downloaded the application in India and also in other countries. So far, 900+ users have downloaded this application from google playstore [Fig. 7] (Google Play Console, 2021).

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Fig. 7. Information of number of Users who downloaded the application

Conflict of Interest: The authors declare that they have no conflict of interest.

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RESEARCH ARTICLE

Development of Genic Simple Sequence Repeat Markers as Novel Genomic Resources in Dolichos Bean (*Lablab purpureus* L.)

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Dolichos bean (*Lablab purpureus* L.) is an old cultivated pulse of Africa, Asia and Australia. In spite of its multiple uses, there are limited genomic resources available, which could facilitate genetic diversity studies and DNA fingerprinting. Here, we report the extraction of SSR markers from the Expressed Sequence Tags (ESTs) of dolichos bean. From a total 1129 unique ESTs of dolichos bean available in public domain, a total of 83 SSRs were identified. Among these EST-SSRs, tri-nucleotide repeats were most abundant (37) followed by tetra-nucleotide repeats (18) and di-nucleotide repeats (15). Functionality of the novel SSRs were validated by successful PCR amplification in nine varieties of dolichos bean using ten SSR markers. The novel EST-SSR markers are expected to be helpful in genetic diversity studies as well as for varietal DNA fingerprinting in this important legume.

Key Words: Dolichos bean, Expressed sequence tags (ESTs), Polymorphic Simple sequence repeats (SSRs)

Introduction

The Dolichos bean (*Lablab purpureus* L.) commonly called as hyacinth bean, field bean, Indian bean, Egyptian kidney bean is an old domesticated pulse and widely cultivated throughout the tropical regions in Africa, Asia and Australia. It is a self-pollinated, bushy semi-erect plant with 2n=22 chromosomes and belongs to Fabaceae family (Ramesh and Byregowda, 2016). While dolichos is used in human diet and animal forage as vegetable, green pods and pulses, it also has utility in intercropping, weed suppressor, and soil erosion retardant. Dolichos, being tolerant to drought and salinity, is grown in a wide variety of climates and soil types. Despite all these advantages, dolichos remained underutilized in terms of cultivation and very limited research was performed for generating genomic resources, diversity analysis and genetic improvement (Ramesh and Byregowda, 2016; Keerthi *et al*., 2018).

Availability of genomics resources in crops helps accelerate studies on genetic diversity, population structure, marker trait association, Quantitative Trait Loci (QTL) identification, gene mapping, comparative genomics and marker assisted breeding which ultimately contribute to genetic improvement and varietal development (Keerthi *et al*., 2018; Kumari *et al*., 2019). At present Simple Sequence Repeat (SSR) markers also known as microsatellite markers are the preferred choice of markers. SSR markers are widely distributed throughout genome, highly variable, co-dominant, multi-allelic and particularly informative (Kumari *et al*., 2019; Wang *et al*., 2011; Desai *et al*., 2021). Expressed Sequence Tag (EST) derived simple sequence repeat markers (EST-SSRs), SSR repeats found in coding sequences, may be generated more rapidly at low cost using the publicly available sequence databases.

There are limited reports on development of SSRs in dolichos bean and most of the markers used are transferred from other species. Zhang *et al*. (2013) has used EST based approach for SSR identification in dolichos bean. However, subsequently, there has been a substantial increase in the submission of dolichos EST sequences in the public sequence databases, which could be used for generation of EST-SSRs. Here we report the development and validation of novel EST-SSRs from dolichos sequences.

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Materials and Methods

Mining of EST-SSRs

A total of 1,486 ESTs (excluding the previously utilized ESTs) of dolichos bean were downloaded from dbEST database of NCBI. Duplicates were removed using *EGassembler* (Masoudi *et al*., 2006). 1129 unique ESTs were subjected to SSR mining and primer designing using *WebSat* software (Martins *et al*., 2009). The search criteria restricted dinucleotide repeats with at least six repeats and a minimum of five repeats for trinucleotide, tetranucleotide, pentanucleotide, and hexanucleotide repeats. Primers were designed *in silico* using *WebSat* software. Out of 75 primers that were designed, 15 primer pairs were synthesized for laboratory validation.

SSR Marker Analysis

Seeds of nine dolichos varieties (Table 1) were obtained from National Genebank, ICAR-NBPGR, New Delhi. DNA extraction and PCR assay were carried out as per standard practice (Saroha *et al* 2022). 15 primers were checked for amplification in three varieties— Arka Jay, Phule Gauri and ArkaAmogh. Ten primer pairs, producing expected amplicons, were then employed in marker analysis of nine released varieties (Table 1). Each amplicon was scored based on molecular weight by using *Alpha View* software. Data were analysed for polymorphic information content (PIC), Shannon's Information Index, observed and expected heterozygosity using *GenAlEx* v6.5 software (Peakell *et al*., 2012).

Results and Discussion

Identification of EST-SSRs from Dolichos Bean

Out of 1,486 downloaded ESTs from NCBI database, total of 1,129 sequence were filtered after removing the duplicates and used in final analysis. All these unique SSR were searched for presence of different SSRs. Total of 83 SSRs from unique 1129 ESTs were identified. Among these SSRs, tri-nucleotide repeats (37) were predominant followed by tetra-nucleotide repeats (18). This was unlike previous reports in Mung bean, Pigeonpea, Cranberry, black alder Maqui Black and pepper, where dinucleotide SSRs were predominant (Tangphatsornruang *et al*., 2009; Dutta *et al*., 2011; Zhu *et al*., 2012; Anupama *et al*., 2015; Bastías*et al*., 2016; Kumari *et al*., 2019). Dinucleotide SSRs were next abundant (15), followed by pentanucleotide repeats (8) and hexanucleotide repeats (5). Among the tri-nucleotide SSRs, the dominant repeat motif was TGA/TCA (21.6%) where as in di-nucleotide repeats, GA distribution was dominant (21.7%).

Table 1. List of released dolichos varieties used for screening and validation of EST-SSR

Accession No.	Name of the variety
IC0393738	Arka Jay
IC0395441	Phule Gauri
IC0588958	Arka Amongh
IC0584609	IIVR SEM-8, Kashi Haritima
IC0588959	Arka Soumya
IC0588960	Arka Sambhram
IC0589768	JIB (P) 04-14
IC0594178	Tirupati Field Bean-2 (TFB-5)
IC0618490	Chhattisgarh Sem-1 (Indira Sem-1)

For the first time, Zhang *et al*. (2013) had reported 22 EST-SSR in dolichos bean. However, since then, the number of ESTs reported in dolichos bean has increased very significantly. The present study, adds as many as 83 EST-SSRs by utilizing publicly available data to generate genomic resources in dolichos. Out of 83 EST-SSR loci, primers could be designed for 75 SSRs (***Supp Table 1***). BLASTx analysis of 75 SSR loci against *Arabidopsis* genome identified known functions for as many as 39 EST-SSR loci extracted in the present study (Table 2). Important hits comprised diverse gene families such as NAD(P)-binding Rossmann-fold superfamily, homeobox-leucine zipper protein family, ChaC-like family, ribosomal protein S12/S23 family, cupredoxin superfamily protein, etc. associated with important biological process such as senescence, late embryogenesis, somatic embryogenesis, transport protein subunit, Syntaxin of plants, etc.

Validation of EST-SSRs and Diversity Analysis

Functionality of the loci and designed primer-pairs was tested for a subset of 15 EST-SSRs. Ten primer pairs yielded amplification of expected size in three dolichos varieties (Fig. 1A). These 10 SSRs were then employed to carry out marker analysis in nine varieties of *dolichos lablab* (Table 1) to ascertain the utility of these markers for DNA profiling either for genetic diversity studies or for cultivar identification.Three SSR loci—LpSSR1, LpSSR41 and LpSSR43—were found to be polymorphic (Fig. 1B) in nine varieties. Ability of these primers to be used for fingerprinting was estimated by the polymorphic information content (0 to 0.49), Shannon index (0 to 0.687), effective number of alleles (1.0 to 1.976) and expected heterozygosity (0 to 0.494). The

Table 2. Annotation of ESTs harbouring SSRs with reference to *Arabidopsis thaliana*

E-SSR	ESTS ID	Annotation
Lpssr_1	JZ151299.1	Syntaxin of plants
Lpssr_2	JZ151202.1	Single hybrid motif superfamily protein
Lpssr_4	JZ150215.1	NAD (P)-binding Rossmann-fold superfamily protein
Lpssr_5	JZ151139.1	Senescence-associated gene
Lpssr_6	JZ151402.1	Hydroxyproline-rich glycoprotein family protein
Lpssr_10	JZ151044.1	ChaC-like family protein
Lpssr_15	JZ150703.1	Senescence-associated family protein
Lpssr_16	JZ150206.1	Ribosomal protein S25 family protein
Lpssr_17	JZ151108.1	MLP-like protein 43
Lpssr_20	JZ150081.1	Late embryogenesis abundant proteins
Lpssr_24	JZ150859.1	Calcium-binding EF-hand family protein
Lpssr_26	JZ151018.1	Putative thioredoxin
Lpssr_30	JZ151051.1	FKBP-like peptidyl-prolyl cis-trans isomerase family protein
Lpssr_32	JZ151302.1	Somatic embryogenesis receptor-like kinase-like protein
Lpssr_33	JZ150096.1	TIFY domain/Divergent CCT motif family protein
Lpssr_36	JZ151158.1	Chalcone-flavanone isomerase family protein
Lpssr_37	JZ150717.1	Dormancy/auxin associated family protein
Lpssr_38	JZ150951.1	SCAMP1
Lpssr_39	JZ150769.1	SR1
Lpssr_40	JZ151306.1	Alcohol dehydrogenase
Lpssr_41	JZ151154.1	KH domain-containing protein / zinc finger (CCCH type) family
Lpssr_42	JZ151346.1	DHHC-type zinc finger family protein
Lpssr_43	JZ151332.1	Protein N-terminal asparagine amidohydrolase
Lpssr_44	JZ151201.1	Metacaspase 3
Lpssr_45	JZ150363.1	Cupredoxin superfamily protein
Lpssr_47	JZ150148.1	ELIP2
Lpssr_48	JZ151311.1	Single hybrid motif superfamily protein
Lpssr_52	JZ150251.1	Beta-6 tubulin
Lpssr_54	JZ151385.1	Cotton fiber protein
Lpssr_57	JZ150828.1	Ribosomal protein S12/S23 family protein
Lpssr_58	JZ151062.1	cotton fiber protein
Lpssr_63	JZ151251.1	Homeobox-leucine zipper protein family
Lpssr_65	JZ150240.1	PyrD
Lpssr_66	JZ150070.1	Major latex-like protein
Lpssr_69	JZ150331.1	Bifunctional inhibitor/lipid-transfer protein/seed storage 2S albumin superfamily protein
Lpssr_70	JZ150836.1	Hypothetical protein AT1G63310
Lpssr_71	JZ151305.1	Mediator of RNA polymerase II transcription subunit 19a-like protein
Lpssr_73	JZ150142.1	Putative transport protein subunit
Lpssr_74	JZ150963.1	Taximin

low polymorphism observed in the dolichos EST-SSRs could be due to the conserved nature of the genic regions, which are known to be relatively less polymorphic than the intergenic SSRs. Similar observations have been reported in dolichos (Zhang *et al*., 2013) and in other species (Kalia *et al*., 2011; Preeti *et al*., 2020). Additionally, low polymorphism could also be due to the limited number of genotypes used in present study; use of larger panel of varieties and diverse germplasm accessions could possibly unravel the polymorphism at the identified SSR loci.

Conclusion

Dolichos bean has remained as an under-studied species in addition to being an underutilized legume. Availability of genomic resources is lacking in this important legume species and as a result genetic studies have been carried out with inadequate number of markers. EST-SSRs reported in this study, are expected to be valuable additional genomic resource in dolichos bean that can facilitate downstream applications such as diversity analysis and cultivar identification.

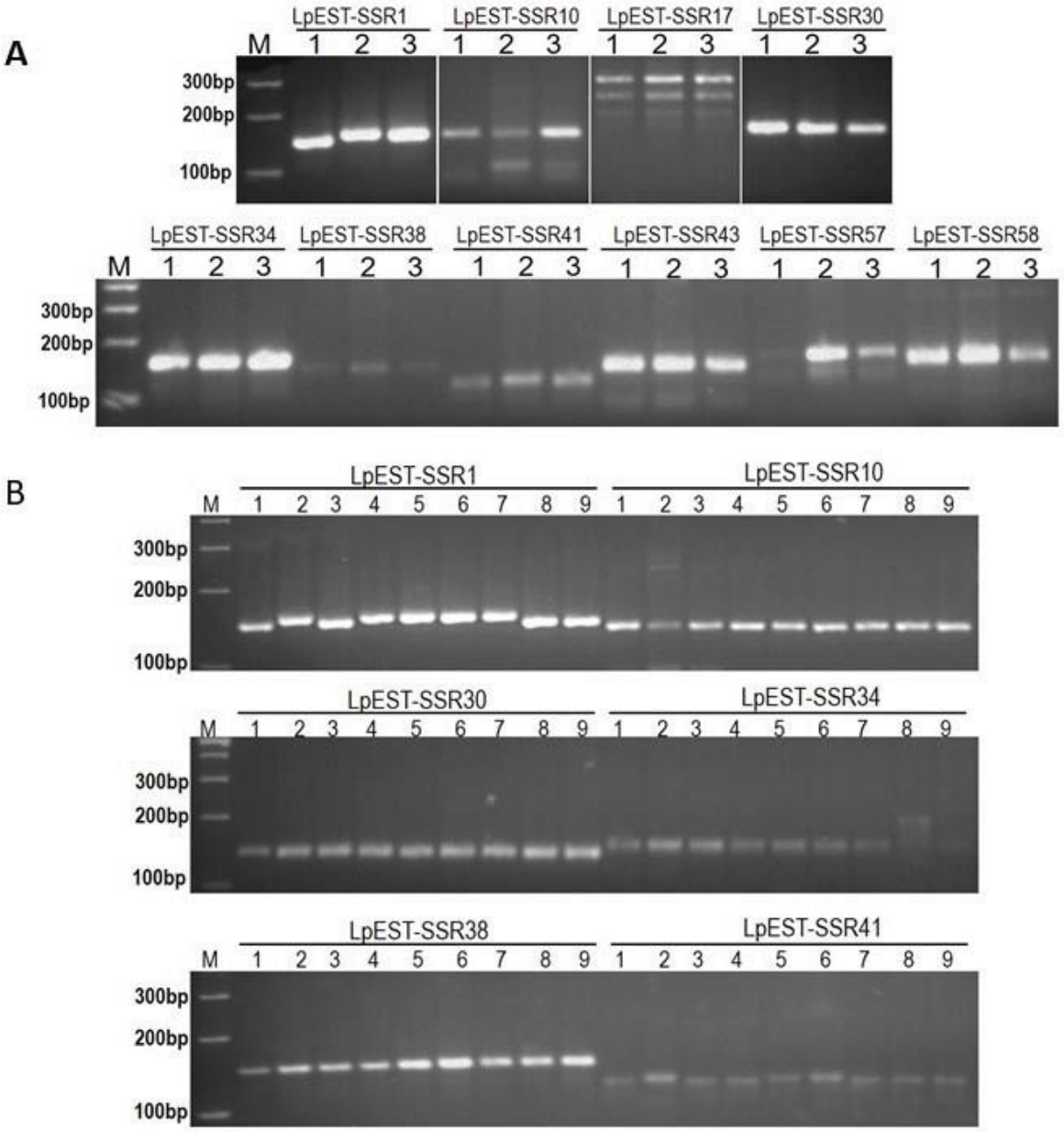

Fig. 1. Validation of EST-SSR markers on dolichos bean varieties. A. Screening of SSR EST-SSR primers for PCR amplification of three varieties. B. Validation of EST-SSR markers on nine released dolichos varieties

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*Supplementary Table or Figure mentioned in the article are available in the online version.

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Supplementary Table 1. Details of EST-SSR markers and respective primers for PCR amplification.

Primer_ID	SSR motif	Forward Primer (5'-3')	Reverse Primer (5'-3')	Annealing Temperature (ºC)	Product size (bp)
Lpssr_1	ATT	CATAGTTCTCCCCATAGTCCT	AACATTATACACACGCATGAA	54	151
Lpssr_2	TC	AGAGAAGAGAGAGATGGCTTG	GAATTCCCATCAACTTTTACC	55	150
Lpssr_3	AGAGA	GGAGAGGAAAAGAGTTGAAGT	AGAAAAGGGTCTGTGAGAGAG	55	129
Lpssr_4	TTC	AGTAGTGGGAGAAAGAGGATG	CGTAGCTTTATTAGCATCACC	54	165
Lpssr_5	TTA	TTGTATTACGGTATCGAGGTT	TGAAGAACTCGGACTTGATAA	54	150
Lpssr_6	ATCA	CGCAAGGTGTAGTAAGAGGTA	GCATCAATTTCCTTAACATGA	55	154
Lpssr_7	TGG	GAACAGTTTTGGGAATTTGA	ACTCTCTCTCCGACGAAAAT	55	184
Lpssr_8	ATGCA	TGAGAACTTGTGCCTTTTTAG	CATACAGATTCTTCTGCTACACA	55	123
Lpssr_9	TATAT	TAGCATACCCTATTGGAAAAA	AACATTTAAACAACCATCACG	55	149
Lpssr_10	AGA	CATCCAAGAAACAAGAAAAAG	GAGAAGGGAGAGAAGAGAAGA	54	147
Lpssr_11	TCA	CAGAACCTATTGCTTTGGAC	GCTTAAAAGTGGGGATACCT	54	151
Lpssr_12	TCTCT	GAGCAGAGAGAAAGAGAGGAG	AACCATGATTTATTGGGATCT	55	166
Lpssr_13	AAG	AATTTCCTTCCACCTTCAAT	GGAGAGAGGGAAATGACG	55	167
Lpssr_14	GCAT	TCATACTTGACAAAGTTGCAG	TTCCCATTAAGAAAAAGAAGG	55	150
Lpssr_15	AAT	GATTCTTGCACTTTAGGGTTT	TCTATTAATTCCCAGATCACG	54	151
Lpssr_16	CTT	GGAGTGATGAGCTTGTATTTG	GCCATTACGGCCTAGTTAC	54	149
Lpssr_17	TTG	CTGCGTGTACTTTATCGTTCT	TTCACATAGAAGAGCACACCT	55	139
Lpssr_18	TCGTTG	CTGCGTGTACTTTATCGTTCT	TTCACATAGAAGAGCACACCT	54	139
Lpssr_19	TTAT	TATTGGACCTTAAATGCACAC	ATTTGTGGCGCTATATAAACA	55	149
Lpssr_20	TCA	GCCCTGAAGTTCATATCTCTT	CAAGAAGAAGAAGGAAGATGG	55	141
Lpssr_21	CTT	TTTCTTCTTCATTCCTCATCA	GGGGAACTGGCTACTCTATTA	54	151
Lpssr_22	GTTT	GAGTGAGAGCCATGTGTTAAG	ATCCAAAGCCATCATTATTCT	55	158
Lpssr_23	CTTT	TAAGAATAATGATGGCTTTGG	ATTTCTCTTGCTCTTGGTCTT	54	144
Lpssr_24	CAA	GGGATTATAACCTCCTCCTTC	TTTGAACACTTGTTGAACCTC	55	145
Lpssr_25	CTG	AAAGCATGTAGTTGAAAATGG	GTGTTGTTAAAGGAGGGTCTT	54	172
Lpssr_26	TTC	CTGATTTAATGGGATTTTCCT	GGAGAATCAGAAAGGGAGATA	54	157
Lpssr_27	GA	AAGCTGCAATAATCAAGTGAG	TTCATGTAGCACACTTCACAC	55	141
Lpssr_28	CAG	ACCATGAGGAGAAAGAAGAAC	CTCACTTGATTATTGCAGCTT	54	156
Lpssr_29	TGA	GTGCTATTGAGGGGACTAGAT	TATGCTCCAATAAAACGCTAT	54	152
Lpssr_30	GGGAAG	AGTTACGGGGAAAAGACTCTA	TTACGTAAGTGCAAGTACCAAG	54	147
Lpssr_31	ATCAA	GGGGGCAAAGAATATATTAAG	AAGAGTGGGTCACAGAAATG	54	143
Lpssr_32	ATG	TAGTTACGGGGGATCTAGC	TGCTCTACCTCCTCATCAATA	54	145
Lpssr_33	AT	TTTCTGAAGCTAACCAATCTG	GTGGCCTTCATACGAATTTAT	55	155
Lpssr_34	AAT	TTTCTGAAGCTAACCAATCTG	GTGGCCTTCATACGAATTTAT	55	155
Lpssr_35	ATAC	TTTCTGAAGCTAACCAATCTG	GTGGCCTTCATACGAATTTAT	55	155
Lpssr_36	TGA	GATGTATACGCATTTGGTGTT	GAAGCCTAACTGTCATGGATA	54	154
Lpssr_37	CCT	ACTACTCCTCCGGTATCTCC	GGAAAACCCGACTATATCTCA	55	156
Lpssr_38	ATTT	ATTATAAGCCACACTCAGCAA	CCATGCTCTCAGCATATACAC	56	146
Lpssr_39	CCG	AAAGGGTAGAGAAAATTCACG	TTGTCAAATAGGTTTGAATGG	55	155
Lpssr_40	AGGT	GTGAAGATTGTGGAGCTCTG	AACTCCTAATTAAAAGGCAACTT	56	157
Lpssr_41	TAT	CCATCTTGTATTAGCCAGGTC	TACGGCAAAACTATAACATGG	56	122
Lpssr_42	AT	AGATGAGGATATCCGAAGATG	TGAGAGTCTCTGCATGTCTTT	56	146
Lpssr_43	TCGT	AGCATCAGCTTGTTCTCAAC	GTTGGTGAGGATGAATGAGTA	56	145
Lpssr_44	ATA	TAGCAAGAACTTGAATTGAGC	AGGTGACTACACTGAAAGCAA	55	151
Lpssr_45	AGATT	GGATTGAGAAAATAGGCAAAT	AACAACCAAAAGAAGCCTAAC	55	138
Lpssr_46	TAAT	GCCTTTGTGTGTGTCTTTAAT	GATCAATGTCAACAGGTTTGT	55	155
Lpssr_47	AGG	GAATTTCGTGCTCACCTTAG	CAGGAGAAGCTTCACTCTCAG	57	148
Lpssr_48	TC	AGTTACGGGAGAGAGAGAAGA	CGAATCAGCATACTTCAAATC	55	139
Lpssr_49	GA	GGAAGAGAGTTCCCTTTTGTA	GGATCATATTGGATCGTTTTT	55	152
Lpssr_50	ACAA	GCCATCCTGTTTCACTTATTT	AAGAAGAAAGGGAAGATGAGA	55	140
Lpssr_51	TTTTA	ATTTGCTCTAGGTCCATTTGT	GTGCTTTCTGTTGAGGTTTTA	55	144
Lpssr_52	ACT	GCCATTACGGCCTAGTTAC	TGTGAAGGATTTCTCTCATTT	54	151

Primer_ID	SSR motif	Forward Primer (5'-3')	Reverse Primer (5'-3')	Annealing Temperature (ºC)	Product size (bp)
Lpssr_53	AAAT	ACATTCAACGTTTTTACGACA	ATGGCATAATATTTGCTTGTG	56	174
Lpssr_54	GA	GTTTTGATGCCATTCTCAAT	CGTGGTATTTATTGCATGATT	55	142
Lpssr_55	AATT	CTCAAATAAAATGGCAAGCTA	GGGAGGAGCATAGTAGGTAGA	55	129
Lpssr_56	TAAA	ACCTAGTGTTTCATTAGTTTTGA	GAAGGATGTTTAATGTTAGGAAA	53	148
Lpssr_57	TGGAAC	GGAGTCCATATTTGGATTTTT	TAGCAAGTCCACACATGATTT	54	159
Lpssr_58	CT	CAAAACACATTCCAATCATTC	AATTTCCATTTCCTCACTCTC	54	148
Lpssr_59	TCTT	GAATCACACCAAGTGTTTGTT	GAATAATGAGCAATAGTGTTGG	54	139
Lpssr_60	TGA	ACACGATTTTCCAACTTAACA	CTAAAGCAGTTTTCTGAACGA	55	180
Lpssr_61	TCA	TTGCAATCTTCTCCTTATTTG	CACCAGATTGATAACGATGAT	55	150
Lpssr_62	TCA	TCTTCTCCTTATTTGCTGTTG	AACATCACCAGATTGATAACG	55	149
Lpssr_63	ATC	CACCTCGTTCTAGGGTTATCT	CAGACAAACCCAATGTTAAAG	55	151
Lpssr_64	TGT	GCATCCCCTTTGATCTATATT	AAACAAACCAAAGAAATCTCC	54	153
Lpssr_65	GA	TCGTATCTGTGTTCGTTTGAT	AGTTACGGGGATGTGTAGTTC	55	149
Lpssr_66	AT	CAGAATTTAGGGAGGTTCTCT	TACGGGGGATACAATATTAGC	55	161
Lpssr_67	CAA	TTCACATAGAAGAGCACACCT	CTGCGTGTACTTTATCGTTCT	55	138
Lpssr_68	ACGACA	TTCACATAGAAGAGCACACCT	CTGCGTGTACTTTATCGTTCT	55	138
Lpssr_69	CA	GACAACACACACAACACACAT	TGAACAAAAACCAATAACACC	55	141
Lpssr_70	ATC	AGAGAAACAAACTTCCAAACC	ACAGATGTGGTTCTATTCTCG	54	149
Lpssr_71	TCT	GTTCCAGAACTGAACCAAAG	AATTAGAATTGACCCGTGAAG	54	145
Lpssr_72	AGAA	AGAAGAGAATTTTGGGTTTTG	CCTCACAAGAACAGGTAAAGA	54	162
Lpssr_73	CCA	CAACAAAGGATATGAAACCAA	GTTAACCTACGTGGAAGGAAG	55	154
Lpssr_74	AG	AAAACACAGTCCAATTTGAAG	AGGAGAGAAAACCAATGAAAG	54	151
Lpssr_75	TAAA	TATATGCTTAAAGGGGAGAGC	CCTGTGAAATTTAAAGCAAAC	54	173

EST-SSR markers shown in bold letters were synthesised and screened against dolichos varieties for PCR amplification. ESTS-SSR markers in bold and underlined have been used for validation in dolichos bean varieties for studying polymorphism and diversity.

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REVIEW ARTICLE

Survey, Collection and Preliminary Observation of Culinary Melon Germplasm from Southern India

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Culinary melon **(***Cucumis melo* spp. *agrestis* var. *acidulus***)** from southern India belongs to the family Cucurbitaceae and commonly known as 'Mangalore melon' or 'sambar cucumber' mainly used for culinary purpose. It is a common and popular vegetable found in almost every home in coastal districts of southern India. This crop has a special feature that the fruits can be stored up to 8-10 months without losing their freshness. Collection and conservation of the germplasm is first and foremost activity in the crop improvement programme. Sixty eight accessions of culinary melon were collected from farmer's fields and vegetable markets of southern Indian states namely Karnataka (31), Kerala (15), Telangana (1), Andhra Pradesh (16) and Tamil Nadu (5). The collected accessions had variability in fruit shape, fruit weight, fruit colour, flesh thickness, seed size, fruit yield, storage life, and other traits. Majority of the farmers cultivated the crop in small areas of paddy fallow land. Fruits may be stored for many weeks by hanging them from the roof ceiling, firmly bound by thin coconut fiber ropes/cut drip wire**.** The seeds of collected germplasm accessions are conserved at the College of Horticulture, Sirsi and will be further evaluated for their growth and yield parameters.

Key Words: Accessions, Collection, Culinary melon, Southern India, Germplasm, Survey

Introduction

Culinary melon **(***Cucumis melo* spp. *agrestis* var. *acidulus***)** is an important under-exploited vegetable of the family Cucurbitaceae and commonly known as 'Mangaluru melon','sambar cucumber', 'moggekayi' and so on. The fruit looks like a cucumber, has unique taste, flavor and aroma when cooked. Unlike the dessert melon, culinary melon is used in preparation of 'sambhar' (vegetable stew with lentils), 'dosa' (southern India rice bread), curries and 'chutneys' (Shruti *et al.,* 2016). It is an important vegetable in most of the homes of coastal districts of South India. It is an ideal summer vegetable crop in the fallow lands of paddy and also grown in *Kharif* mainly for fresh vegetable. South Indian culinary melon may be stored for 8-10 months without losing freshness. It produces small to large sized fruits with smooth tender skin, white flesh usually with high to medium acidity, little sweetness and odour (Swamy 2017). However, cultivation of culinary melon is restricted to few coastal districts of South India because of poor agronomic performance, blonde taste and less awareness about the crop to the rest of the world.

There is need for sustained effort to maintain and facilitate access to culinary melon / southern Indian melon germplasm. This is especially important for melon group and its wild relatives of the more diverse and less intensively cultivated culinary melon. With this background, survey and collection of culinary melon landraces / accessions from different Southern Indian states were collected and preliminary data were recorded.

Methodology

The survey and collection of culinary melon landraces/ accessions were initiated during April to August 2018 targeting five southern Indian states, namely, Karnataka, Kerala, Andhra Pradesh, Tamil Nadu and Telangana. Primary information of crop and growing region were collected from the State Agriculture and Horticulture Departments, ICAR-NBPGR Regional Stations, Krishi

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Vigyana Kendras, State Agricultural Universities and ICAR Institutes. Datasheet for collection of primary data was designed and used (Form 1). Random sampling technique was used for collecting accessions from growing regions of culinary melon. The areas of survey and collection are presented in Figure 1. The information collected from farmers was documented and 12 fruit component traits for 53 collected accessions namely shape, color, length (cm), breadth (cm), flesh thickness (cm), weight (kg) seed cavity length and width (cm), 100 seed-weight (g), seed per fruit, seed length and diameter (mm) were recorded in the laboratory to study the variability in the fruit collection. The mean and range were computed for comparing the traits among the collected accessions.

Results and Discussion

A total of 68 landraces of culinary melon, including two wild melons were collected from farmer's fields, vegetable markets of Kerala, Karnataka Andhra Pradesh and Tamil Nadu as well as from Kerala Agricultural University, Thrissur, Kerala and YSR Horticultural University, Venkataramagudem, Andhra Pradesh (Table1).

Survey and collection of germplasm

State-wise details are as follows:

Kerala

Surveyed 18 farmers' fields in the district of Thrissur, Mallapuram, Kannur, Wayanad, Kasaragod and Trivandrum. A total of 13 fruits were collected from Chevvoor, Vylantur, Pilicode, Adur, Kumbla, Ambalavayal, Edacheri, Muttippalam, Peravoor and Puttur (Kottikal), besides two seed samples from Kerala Agriculture University, Velanikkara, Thrissur. The fruits collected from Thrissur district are elongated and yellow incolor without prominent stripes. The fruits collected from Mallapurum district are small, round, yellow in color, called as 'Kanivellari'. In the hilly region (Wayanad) striped type fruits are predominantly found. The shelf -life of the fruit ranged from 3-6 months and mainly used for culinary purpose (Mr. Ashraf from Kannur, Personal communication). Farmers observations on shelf-life is supported by the study of Koli and Murthy (2013).

Karnataka

The traditional belt of culinary melon is Coastal and Malnad regions of Karnataka. During May (9-14) 2018 Sirsi, Yallapura, Siddapur, Kumta and Honnavartaluks of Uttar Kannada; Sagar, Hosnagara, Thirthalli, Soraba taluks of Shivammoga; Sringeri, Kadur taluks of Chickmagalore; Belthangadi, Ujire, Sulya, Mangalore

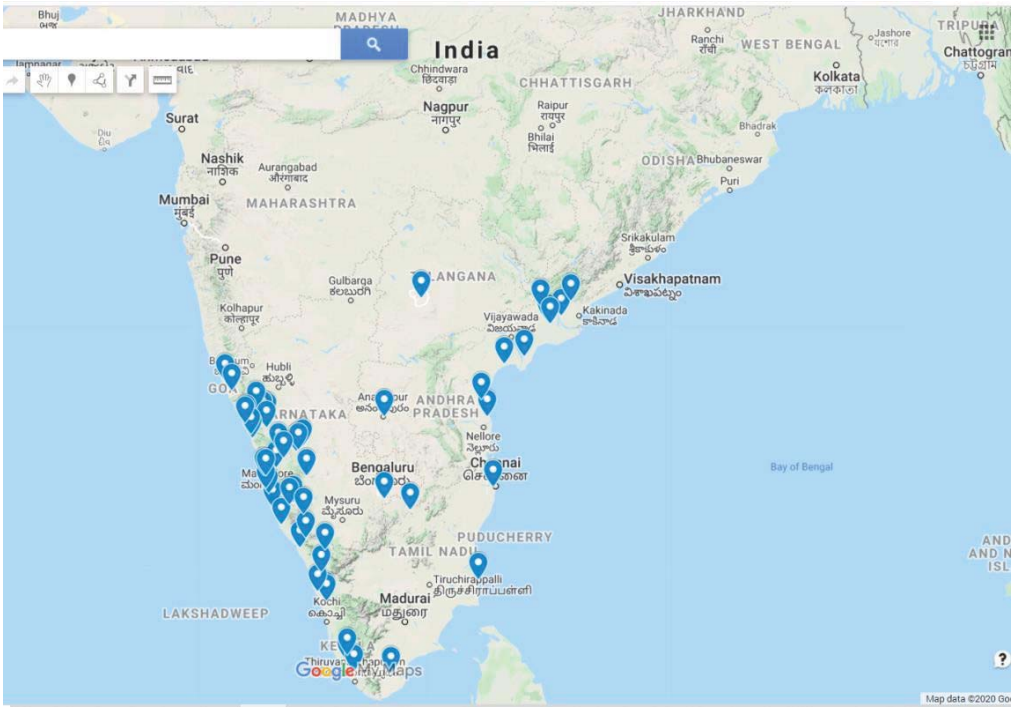

Fig. 1. Map showing survey and collection area of culinary melon landraces from southern India

Table 1. Landraces/ accessions of culinary melon/Mangalore melon collected from southern India (68)

Accession No	Botanical Name	Place of Collection	State	Latitude	Longitude	**Remarks**
MS1	*C. melo* var. *acidulus*	Chevvoor, Thrissur	Kerala	1 º46''N	76 º21''E	Fruits are ovate in shape, with green to yellow color rind, and white flesh
MS2	*C. melo* var *acidulus*	Chevvoor, Thrissur	Kerala	1 º46''N	76 º21''E	Elongated yellow colored fruit, Fully ripe fruit had good flavor
MS3	*C. melo* var *acidulus*	Vylantur Thrissur	Kerala	1 º61''N	76 ºº8''E	Elongated green to yellow colored fruits on maturity, looks like cucumber
MS4	*C. melo* var *acidulus*	Chevvoor, Thrissur	Kerala	9 º51''N	76 º76''E	Elongated green to yellow colored fruits on maturity, good juice content
MS5	*C. melo* spp. *agrestis*	Velanikkara, Thrissur	Kerala	1 º55'' N	76 º27'' E	Commonly known as senat seed, small gourd, and wild musk melon. Striped fruits, small to medium size>10 fruits/vine small seeded
MS6	*C. melo* spp. *agrestis*	Velanikkara, Trissur	Kerala	1 º55'' N	76 º27'' E	Commonly known as senat seed, small gourd, wild musk melon. Striped fruits, small to medium size >10 fruits/vine
MS-7	*C. melo* var. *acidulus*	Pilicode, Kasaragod	Kerala	12 º2 º''N	75 º11''E	Patchy striped, ovate shaped fruits
MS8	*C. melo* var. *acidulus*	Adur, Wayanad	Kerala	11 º59''N	76 º21''E	Ovate shape, green yellow stripe fruits with good flesh content
MS9	*C. melo* var. *acidulus*	Ambalavayal, Wayanad	Kerala	11 º63''N	76 º28''E	Oblate shape, green yellow stripe with medium fruit size
MS10	*C. melo* var. *acidulus*	Edacheri, Kozhikode	Kerala	11 º67''N	75 º61''E	Oblate shape, less prominent stripes, orange color on maturity
MS11	*C. melo* var. *acidulus*	Kozhikode	Kerala	11 º68''N	75 º61''E	Oblate shape, green with yellow stripes, matures at 55 days after sowing
MS12	*C. melo* var. *acidulus*	Kumbla, Kasaragod	Kerala	12 º5 º''N	74 º98''E	Ovate shaped, greenish yellow prominent stripes with average fruit weight of1.5-2 kg
MS13	*C. melo* var. *acidulus*	Peravoor, Kannur	Kerala	11 º89''N	75 º73''E	Oblate shaped, greenish yellow prominent stripes on maturity
MS14	*C. melo* var. *acidulus*	Puttur (Kottakal) Malappuram	Kerala	11 ºº2''N	76 ºº4''E	Round shaped, yellow color, small fruit
MS15	*C. melo* var. *acidulus*	Muttippalam, Mallapuram	Kerala	11 ºº4''N	76 ºº3''E	Round, golden yellow color, medium sized fruits, little sourish flesh
MS16	*C. melo* var. *acidulus*	Haleangadi, Mangaluru	Karnataka	13 ºº4''N	74 º79''E	Oblate, prominent green yellow stripes, medium sized fruits
MS17	*C. melo* var. *acidulus*	Kuluvelu, Mangaluru	Karnataka	13 ºº4''N	74 º8 º''E	Pyriform shaped, green yellow striped fruits
MS18	*C. melo* var. *acidulus*	Pavanji, Mangaluru	Karnataka	13 ºº2''N	74 º79''E	Seed samples were collected
MS19	*C. melo* var. *acidulus*	Olabailu, Udupi	Karnataka	12 º69''N	75 º6 º''E	Ovate, patchy green yellow striped fruit with good flesh content
MS20	*C. melo* var. *acidulus*	Konkarni, Udupi	Karnataka	12 º67''N	75 º6 º''E	Dark green color oblate shaped fruit with good flesh content
MS21	*C. melo* var. *acidulus*	Bapnalli, Sirsi	Karnataka	14 º68''N	74 º84''E	Green color, oblate fruit good crunchy flesh
MS22	*C. melo* var. *acidulus*	Manbhagi Sirsi	Karnataka	14 º74''N	74 º78''E	Oblate shape, green yellow striped fruit
MS23	*C. melo* var. *acidulus*	Manbhagi, Sirsi	Karnataka	14 º74''N	74 º78''E	White color, ovate, medium sized fruit
MS24	*C. melo* var. *acidulus*	Salkani, Sirsi	Karnataka	14 º66''N	74 º71''E	Oblong shaped, striped fruit
MS25	*C. melo* var. *acidulus*	Manchikere,Yellapur	Karnataka	14 º85''N	74 º82''E	Medium sized, ovate, green fruit
MS26	*C. melo* var. *acidulus*	Kadabala, Sirsi	Karnataka	14 º94''N	74 º81''E	White color, oblong fruit
MS27	*C. melo* var. *acidulus*	Kadabala, Sirsi	Karnataka	14 º94''N	74 º81''E	Light green, oblate shaped fruit
MS28	*C. melo* var. *acidulus*	Kadabala, Sirsi	Karnataka	14 º94''N	74 º81''E	Oblate shaped, green, prominent ribs on skin. Good flesh content
MS29	*C. melo* var. *acidulus*	Andur, Sirsi	Karnataka	14 º71''N	74 º9 º''E	Ovate, medium sized, green, yellow striped fruit
MS30	*C. melo* var. *acidulus*	Vajralli, Yellapur	Karnataka	14 º88''N	74 º57''E	>10 month shelf life, striped ,oblate fruit, young fruits bitter in taste
MS31	*C. melo* var. *acidulus*	Karingolli, Hosnagar Shimoga	Karnataka	13 º91''N	75 ºº6''E	Oblate shape, patchy green with yellow stripes
MS32	*C. melo* var. *acidulus*	Karingolli, Hosnagar Shimoga	Karnataka	13 º91''N	75 ºº6''E	Oblate shape, patchy green with yellow stripes with good flesh content

Accession No	Botanical Name	Place of Collection	State	Latitude	Longitude	**Remarks**
MS33	*C. melo* var. *acidulus*	Shantaveri, Thirthahalli, Shimoga	Karnataka	13 º75''N	75 º19''E	Oblate shape, patchy green with yellow stripes, medium sized fruit
MS34	*C. melo* var. *acidulus*	Araga, Thirthahalli, Shimoga	Karnataka	13 º73''N	75 º2 º''E	Oblate shape, patchy green with yellow stripes
MS35	*C. melo* var. *acidulus*	Baarkodi, Sringeri, Chickmagalore	Karnataka	13 º41''N	75 º18''E	Oblate shape, patchy green with yellow stripes
MS36	*C. melo* var. *acidulus*	Kimnajji, Belthangadi, Dakshina Kannada	Karnataka	12 º92''N	75 3 º''E	Oblate shape, orange color fruits on maturity, early type
MS37	*C. melo* var. *acidulus*	Koyyur, Belthangadi, Dakshina Kannada	Karnataka	12 º9 º''N	75 3 º''E	Elliptical, patchy green with yellow stripes on maturity
MS38	*C. melo* var. *acidulus*	Bellare, Sulya, Dakshina Kannada	Karnataka	12 º66''N	75 º36''E	Oblate, green yellow striped fruit
MS39	*C. melo* var. *acidulus*	Medanaadu, Kodagu	Karnataka	12 º41''N	75 º66''E	Oblate shaped, green with scattered yellow patches
MS41	*C. melo* var. *acidulus*	Hosakoppa, Sidhapur, Uttara Kannada	Karnataka	14 º73''N	74 º98''E	Oblate medium sized orange color on maturity fruits
MS42	*C. melo* var. *acidulus*	Harsikatta, Sidhapur, Uttara Kannada	Karnataka	14 º72''N	74 º98''E	Green color oblate shaped big size fruits with good flesh content
MS43	*C. melo* var. *acidulus*	Gokarna, Kumta, Uttara Kannada	Karnataka	14 º7 º''N	74 º32''E	Elongated green yellow Scattered stripes
MS44	*C. melo* var. *acidulus*	Hegde, Kumta, Uttara Kannada	Karnataka	14 º55''N	74 º31''E	Oblate shaped, patchy green with yellow stripes on maturity
MS45	*C. melo* var. *acidulus*	Honnavar, Uttara Kannada	Karnataka	14 º23''N	74 º5 º''E	Oblate yellow color with less prominent stripes fruits
MS46	*C. melo* var. *acidulus*	Honnavar, Uttara Kannada	Karnataka	14 º27''N	74 º44''E	Pyriform, green with yellow stripes on maturity
MS48	*C. melo* var. *acidulus*	Medipetnum, Hyderabad	Telangana	13 º24''N	77 º7 º''E	Round, orange color small size fruit
MS49	*C. melo* var. *acidulus*	Avupadu, West Godavari	Andhra Pradesh	16 º81''N	81 º53''E	Round, orange color small fruit
MS50	*C. melo* var. *acidulus*	Avupadu West Godavari	Andhra Pradesh	17 º41''N	81 º28''E	Seed samples collected
MS51	*C. melo* var. *acidulus*	Prakasaraopalem, West Godavari	Andhra Pradesh	16 º94''N	81 º4 º''E	Seed samples collected
MS52	*C. melo* var. *acidulus*	Venkataramagudem, West Godavari	Andhra Pradesh	17 º31''N	81 º18''E	Round, golden yellow color
MS53	*C. melo* var. *acidulus*	Nerakoduru, Guntur,	Andhra Pradesh	15 º91''N	8 º47''E	Seed samples collected
MS54	*C. melo* var. *acidulus*	Nerakoduru, Guntur,	Andhra Pradesh	15 º91''N	8 º47''E	Seed samples collected
MS56	*C. melo* var. *acidulus*	Rajendra Nagar, Hyderabad	Andhra Pradesh	17 º32''N	78 º4 º''E	Seed samples collected
MS57	*C. melo* var. *acidulus*	Rajendra Nagar, Hyderabad	Andhra Pradesh	17 º32''N	78 º4 º''E	Round, orange color small fruit
MS59	*C. melo* var. *acidulus*	Nagupalli, Khammam	Andhra Pradesh	17 º66''N	82 º99''E	Seed samples collected
MS60	*C. melo* var. *acidulus*	Aswaraopet, Khanmam	Andhra Pradesh	17 º24''N	81 º33''E	Seed samples collected
MS63	*C. melo* var. *acidulus*	Venkataramagudem, West Godavari	Andhra Pradesh	17 º31''N	81 º18''E	Seed samples collected
MS64	*C. melo* var. *acidulus*	Podalakur, Nellur	Andhra Pradesh	14 º38''N	79o 73''E	Seed samples collected
MS65	*C. melo* var. *acidulus*	Naredumilli, East Godhavari	Andhra Pradesh	17 º16''N	82 ºº6''E	Seed samples collected
MS66	*C. melo* var. *acidulus*	Darsi, Prakasam	Andhra Pradesh	15 º77''N	79 º68''E	Seed samples collected
MS68	*C. melo* var. *acidulus*	Mirzapuram, Krishna	Andhra Pradesh	16 º94''N	81 º4 º''E	Round yellow color fruit with little sour taste flesh
MS69	*C. melo* var. *acidulus*	Prakasaraopalem, West Godavari	Andhra Pradesh	16 º9 º''N	81 º42''E	Seed samples collected
MS70	*C. melo* var. *acidulus*	Avapadu, West Godavari	Andhra Pradesh	13 ºº7''N	8 º2 º''E	Seed samples collected
MS71	*C. melo* var. *acidulus*	Chennai, Market	Tamil Nadu	8 º71''N	77 º75''E	Round orange color small fruit
MS72	*C. melo* var. *acidulus*	Tirunelvelli	Tamil Nadu	11 º94''N	79 º81''E	Globular shaped yellow color small fruit
MS73	*C. melo* var. *acidulus*	Kariakal	Tamil Nadu	15 º91''N	8 º47''E	Oblate golden yellow color fruit
MS74	*C. melo* var. *acidulus*	Krishnagiri	Tamil Nadu	12 º51''N	78 º22''E	Elliptical Stripe fruit, Medium size fruit
MS75	*C. melo* var. *acidulus*	Krishnagiri	Tamil Nadu	12 º51''N	78 º22''E	Pyriform shaped striped fruit
MS79	*C. melo var. acidulus*	Salkani, Sirsi	Karnataka	14 º66''N	74 º71''E	Seed samples collected

taluks of Dakshina Kannada; Bramhavartaluk of Udupi; and Madikeri taluk of Kodagu were surveyed. A total of 30 fruits were collected during survey. The fruits from coastal Karnataka were ovate to oblate in shape and patchy green color rind with white prominent stripe. However, green color fruits were observed in Malnad regions especially Sirsi, Siddhapur and Yellapur taluks of Uttara Kannada. Most of the farmers of Uttara Kannada grow the culinary melon organically and able to store fruits from *Kharif* to next summer season. Fruits can be stored for many weeks by hanging them from the ceiling, firmly bound by thin banana fiber ropes/cut drip wire (Dattatreya Hegde-personal communication).

Andhra Pradesh/Telangana

Telangana and Andhra Pradesh areas were surveyed during April 27-29, 2018 and seed samples of 13 accessions were collected from YSR Horticulture University as well as from vegetable growers and four fruit selfless from Budhuvelle (Medipetnum market), Hyderabad, Venkataramdugem, Bapatla, West Godavari districts of Andhra Pradesh. The fruits are small, round in shape with scattered patches on skin and turns yellow color on maturity.

Tamil Nadu

Accessions from Tamil Nadu are similar to Kerala types and cultivated in limited area (Dr. John Joseph-personnel communication). Hossur, Krishnagiri, Dharmapuri, Salem, Dindugul, Palakad (Kerala State), Tirunelveli, Chennai market and Coimbatore districts of Tamil Nadu were surveyed during July 10-14, 2018 and five fruits collected.

According to the information collected from scientists of Tamil Nadu Agricultural University, Coimbatore and Centre of Excellence for Vegetables at Reddiarchatram, Tamil Nadu crop cultivation area is decreasing in Tamil Nadu and grown in some parts of Tirunelveli and Madurai regions. Farmers are mainly growing pumpkin for culinary purpose rather than culinary melon in northern parts of Tamil Nadu. However, during British rule in India culinary melon (Madras cucumber) was introduced to India's east coast (Madras province) in 1805. Madras province as defined under British law, which extended to present-day Tamil Nadu, Andhra Pradesh, parts of Karnataka and Kerala (Vidya, 2012). Madras was the main shipping centre for Kerala and Tamil Nadu. Fruit produced in Kerala was shipped through Madras as a result, 'Vellari' crop in Kerala is also known by Tamilians as 'Madras cucumber' (personnel communication)

Variability collected

Out of 68 accessions collected from the five Southern Indian states, 51 fruit samples were subjected for post-harvest observations on 12 fruit component traits to understand the extent of variability in the collection. Data is presented in Table 2 and variability of fruit size, shape and color is presented in Figure 2. The analysis of data revealed that significant variation exists among the collected accessions for fruit traits. The accessions had bigger size fruits and weight ranged from 220-3,900 g with mean of 1559 g. The accessions collected from Kadabala Sirsi (MS-26, MS-27, MS-28) had maximum fruit weight. The application of organic fertilizer (panchamrutha) at flowering stage and sufficient irrigation increases fruit size as informed by Kadabala farmer Mr. SN Bhat. The fruits length and breadth are associated with flesh thickness, seed cavity length and width. Fruits component variability was higher in Karnataka accessions namely MS-21, MS-22, MS-26, MS-27, MS-28, MS-30 and MS-79 compared to accessions from other states.

The flesh thickness of collected accessions ranged from 1.1 to 5.5 cm with a mean of 3.27 cm. The Karnataka state collected accessions viz., MS-21 had maximum flesh thickness (5.50 cm) followed by MS-30 (5.25 cm) and MS-28 (5.00 cm) which is important component for culinary purpose. The maximum length of the fruit (35.50 cm) was observed in accession MS-2 collected from Thrissur district of Kerala. High variability was observed for seed characters namely seed length, seed diameter, seed number and 100-seed weight. The seed length varied from 6.92 to 9.97 mm with mean of 8.64 mm similarly seed diameter (1.94-4.15 mm), seeds/fruit (180-604) and 100-seed weight (1.66–3.68g). The seeds of *agrestis* and Andhra Pradesh collections are small (less than 6 mm seed length) compared to others. Dhillon *et al.* (2012), Fergany *et al.* (2011), Manohar and Murthy (2012) also reported < 9 mm seed length in collected culinary melon landraces.

The collected fruits had wide variability in shape and color. Among the 51 collected accessions, fruit shape varied from ovate (18), oblate (17), elongated (4), elliptical (6) and round (6). The maximum number of ovate and oblong fruits were collected from Karnataka had higher flesh thickness compared to elongated,

Table 2. Variability in mean values of fruit components among the south Indian collected accessions

Accessions No	Fruit length (cm)	Fruit breadth (cm)	Fruit weight (g)	Flesh thickness (cm)	Seed cavity length (cm)	Seed cavity width (cm)	Seeds/ fruits	Seed length (mm)	Seed diameter (mm)	100 seed weight (g)
MS 1	35.50	8.25	1430	2.75	29.00	1.75	400.00	9.17	3.42	2.90
MS 2	23.00	7.75	680	1.70	17.00	4.00	482.00	7.90	3.01	1.90
MS 3	17.00	7.50	500	1.80	11.50	4.00	300.00	7.80	3.12	2.10
MS 4	32.50	13.00	2850	4.00	27.40	5.30	525.00	8.20	3.10	1.75
MS 7	16.70	11.00	1100	2.50	14.00	7.50	324.00	7.80	3.60	2.10
MS 8	10.00	7.00	220	1.10	7.50	4.00	258.00	8.90	2.01	1.68
MS 9	26.50	11.50	1800	4.50	14.00	4.00	400.00	8.62	3.40	2.25
MS 10	24.00	13.50	2300	3.50	19.00	4.50	286.00	7.92	3.60	2.10
MS 11	19.00	11.50	2300	3.50	15.00	5.50	352.00	8.25	3.58	2.60
MS 12	22.00	13.00	1700	3.20	15.50	5.00	305.00	9.20	2.44	2.80
MS 13	19.00	10.50	1300	3.50	14.50	4.50	238.00	8.50	3.20	1.95
MS 14	9.20	9.00	490	1.70	7.00	4.50	321.00	6.92	3.50	1.68
MS 15	10.20	10.50	520	2.50	7.50	6.00	352.00	7.00	3.12	1.86
MS 16	11.50	8.00	260	2.00	9.50	4.00	278.00	9.37	4.08	2.59
MS 17	13.00	8.00	490	2.30	8.50	3.50	180.00	9.83	4.15	2.55
MS 19	17.50	10.50	820	3.00	10.00	4.50	255.00	8.72	3.99	2.71
MS 20	24.00	15.00	2100	4.30	14.00	6.50	515.00	8.72	3.30	1.95
MS 21	28.00	15.50	3450	5.50	21.50	5.60	289.00	8.32	2.80	2.20
MS 22	19.30	12.50	3850	3.50	12.50	5.50	272.00	8.81	3.80	1.89
MS 23	21.00	16.00	2270	4.75	16.00	5.00	375.00	8.71	3.40	1.80
MS 26	29.00	18.00	3900	4.65	13.00	4.00	295.00	9.66	3.92	3.36
MS 27	26.00	17.50	3750	4.70	19.50	6.00	604.00	9.19	3.98	3.15
MS 28	27.00	18.50	3900	5.00	20.50	7.00	598.00	8.95	4.04	2.96
MS 29	18.50	14.00	1650	3.60	11.00	6.50	565.00	8.10	2.45	2.38
MS 30	17.00	14.00	1550	5.25	12.00	5.50	300.00	9.03	3.77	2.97
MS 31	22.00	16.00	2400	4.50	12.00	5.00	445.00	9.33	3.88	3.68
MS 32	23.50	11.80	1900	4.25	12.00	4.50	483.00	8.84	3.55	1.98
MS 33	23.00	12.00	1400	3.50	18.00	5.00	498.00	9.55	3.94	2.85
MS 34	23.00	10.30	1410	3.10	16.00	4.30	483.00	9.33	3.88	3.68
MS 35	26.30	12.60	2150	3.50	17.00	5.80	398.00	8.84	3.55	1.98
MS 36	24.00	10.50	1150	3.00	16.00	4.50	520.00	9.55	3.94	2.85
MS 37	22.50	11.50	1500	2.50	13.00	7.00	361.00	9.97	3.29	2.79
MS 38	23.00	11.20	1600	3.50	15.00	5.00	258.00	7.89	1.94	2.37
MS 39	21.00	11.80	1300	3.20	13.50	5.20	380.00	9.90	3.97	3.06
MS 40	22.00	11.20	1300	2.50	16.00	6.00	353.00	9.51	4.03	2.85
MS 41	24.50	12.50	1900	3.50	17.50	5.50	238.00	9.23	3.33	2.84
MS 42	23.50	12.00	1500	4.00	15.50	4.50	511.00	9.46	3.80	2.89
MS 43	27.50	13.00	2150	3.80	17.00	5.00	500.00	7.17	2.95	1.66
MS 44	26.50	11.80	1900	3.70	19.50	5.50	307.00	8.56	3.50	2.31
MS 45	19.50	12.50	1500	3.50	13.00	4.70	494.00	8.53	3.39	2.16
MS 46	15.00	13.00	1250	3.50	11.00	5.20	232.00	8.55	3.53	2.21
MS 47	15.00	10.00	750	2.70	10.50	4.50	319.00	8.35	3.62	3.50
MS 57	16.50	10.70	800	3.50	10.50	4.00	300.00	9.80	3.86	2.80
MS 65	12.50	7.30	330	1.50	9.00	4.50	542.00	6.93	3.37	2.63
MS 68	16.70	10.00	850	3.00	11.00	5.50	421.00	7.93	3.90	1.76
MS 71	11.00	9.50	700	2.50	8.00	5.50	321.00	7.81	3.73	1.77
MS 72	19.00	10.00	900	3.10	15.50	4.00	465.00	7.32	3.77	2.17
MS 73	16.00	9.50	750	2.20	11.50	4.70	382.00	9.64	4.12	2.66
MS 74	14.50	11.00	800	3.00	10.50	4.50	347.00	9.30	3.80	2.23
MS 75	17.00	8.00	700	2.00	13.00	5.00	320.00	7.20	2.70	2.10
MS 79	20.00	10.50	1400	3.00	12.50	5.50	412.00	8.61	3.82	2.99
Mean	20.42	11.60	1558.55	3.27	14.32	4.99	379.58	8.64	3.49	2.45
Minimum	9.20	7.00	220.00	1.10	7.00	1.75	180.00	6.92	1.94	1.66
Maximum	35.50	18.50	3900.00	5.50	29.00	7.50	604.00	9.97	4.15	3.68

Fig. 2. Culinary melon variability collected from southern Indian states

FORM1

Data Sheet for collection of Primary Data/Information on Mangalore Southekayi Landraces/ Local Varieties

1. Name of Collecting Institute :
2. Date of Collecting:
3. Collecting Number:
4. Name & Address of Farmer:
5. Age of farmer:
6. Educational level: Illiterate/Read only/Read & write/Primary school/Middleschool/High school/PUC/Diploma/Degree/PG
7. Status of Sample: Traditional cultivar/Landrace/Variety
8. Local/Vernacular Name(s) :
9. Type of sample collected: Fruits/Seeds
10. Extent of Areas (in ha or Number of plants):
11. Growing condition :Wetland/Dryland
12. Grown as: Monocrop/Intercrop
    If intercrop, specify the crops:
13. Cultural practices adopted by the farmer:
14. Fruit shape:
15: Fruit weight:
16. Skin/Rind colour/pattern:
17. Number of fruits per vine :
18. Days to fruit harvest :
19. Other crops grown:
20. Driving force to take up this crop:
21. USP (Unique selling preposition) of this crop such as taste, shape, colour :
22. Additional Notes :
    Parts used-Culinary uses-Shelf life-Storage method-Source of seeds-Reaction to pest and diseases-Marketing-Income-
    Is it a profitable crop as mono crop or supports the livelihood?

elliptical and round shape fruits. Similarly, fruit color ranged from dark green to white, yellow to golden yellow to orange, green and white to yellow stripes. The green color fruits didn't change its color and had longer shelf life (>8 months) compared to other fruit types. All types of fruits showed more than six month shelf-life, indicating promising genetic source for enhancing the shelf-life of other melon group vegetables (Manohar and Murthy, 2012). The preliminary observations revealed that the accessions collected from Karnataka namely MS 21, MS 27, MS 28, MS 30, MS 34, MS 36, MS 38, MS 39, MS 42 were oblate shaped and medium to bigger sized fruits with higher shelf-life. These accessions had detailed evaluation along with collected germplasm of culinary melon from southern India in augmented design.

Conclusion

Collection, conservation and utilization of desirable germplasm in crop improvement program is a continuous process. The survey and collection generate precious genetic resources of culinary melon from southern India which can be utilized for genetic improvement of culinary melon as well as other melon group vegetables for various biotic and a biotic resistance breeding. Evaluation of collected germplasm will open various ways for future research programs. The shelf-life of culinary melon can be exploited through introgression breeding for enhancing storage life of musk melon and other melon group of vegetables in near future.

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SHORT COMMUNICATION

Effect of Different Pollen Sources on Fruit Set and Maturity of Exotic Pear (*Pyrus communis* L.) cvs. Carmen and Abate Fetel under Kashmir Conditions

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The cultivated pear (*Pyrus communis* L.) is an economically important fruit crop often showing gametophytic self-incompatibility. Therefore, this species needs to be pollinated by cross-compatible cultivars that bloom in the same time. Selection of appropriate pollinizers for pear cultivars is thus very important to produce commercial yield. The present study was undertaken on two exotic pear cultivars *viz*., Carmen and Abate Fetel used as maternal parents with seven pollinizer cultivars *viz.,* William Bartlett, Fertility, Clapp's Favorite, Chinese Sandy Pear, Max Red Bartlett, Kings Pear and Beurre d'Amanalis to assess the pollination behaviour, compatibility and effect of these pollen sources on fruit maturity. The treatments included self, open and cross pollination and the design of experiment was Randomized Complete Block Design (RCBD) with three replications per treatment. Among all the studied cultivars Chinese Sandy Pear was earliest (18^{th}-19^{th} March) in bud burst whereas, Abate Fetel was late in all the floral phenology parameters.Maximum flowering duration (17 days) was recorded in William Bartlett and Fertility whereas minimum (12 days) in Chinese Sandy Pear and Kings Pear.Highest compatibility in terms of fruit set (74.00%) was recorded with pollinizer "Fertility" followed by 72.83% with "William Bartlett" and 72.67% with "Max Red Bartlett. Likewise, highest mean fruit retention percentage (46.42%) was recorded with pollen source "Fertility" followed by 46.21% and 46.11% with "William Bartlett" and "Max Red Bartlett", respectively. Maximum days (127.10 days) from full bloom required to reach the harvestable stage were observed with pollinizer William Bartlett and minimum (123.49 days) with Clapp's Favourite. The pollinizer cultivars Fertility, William Bartlett and Max Red Bartlett proved to be best pollinizers for cvs. Carmen and Abate Fetel in terms of flowering duration, bloom synchronization and fruit set under Kashmir conditions.

Key Words: Fruit set, Flower phenology, Maturity, Pear, Pollination, Pollen compatibility

Introduction

Pear (*Pyrus communis* L.) cultivars are generally considered as self-unfruitful and do not set fruit by their own pollen due to the self-incompatibility. Self-fertilization in pear is prevented by a gametophytic self-incompatibility system (De-Nettancourt, 2001) and most of the pear cultivars are self-incompatible (Stern *et al.*, 2004). This universal phenomenon of incompatibility averts the process of self-pollination. The transportation of pollen from flowers of one variety to those of another is probably the most critical single process in the series of events leading to the production of a good quality fruit. Pollination is the sexual portion of a tree's life cycle and involves the integration of several biological and physical factors comprising compatibility of different varieties, coincident blossoming periods, plenty of pollinators and suitable weather conditions. Absence of any of these factors may affect the crop yield and quality. Indeed, pollination management should be regarded as a production factor in its own right for the pear crop as it can affect the agronomic and economic yields and thereby many components such as fruit set, fruit quality (e.g. size, shape, colour and storability) and seed content. For effective cross pollination it is very important that the cultivars produce sufficient quantity of viable, compatible pollen and bloom approximately at the same time and the compatible pollinizers must be planted in the right proportion (Ershadi *et al*., 2010). Recently, two coloured pear cultivars Carmen and Abate Fetel were introduced by Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shalimar campus and under Kashmir conditions both

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these cultivars produce abundant bloom, however, bloom synchronization of these cultivars with the cultivars already grown in valley is not known. This necessitates the study on floral phenology and compatibility of different pollinizer cultivars with these varieties to ensure effective cross pollination with the ultimate aim of enhanced fruit set and yield in pear.

Materials and Methods

Healthy pear plants of uniform and full bearing age (6 years) and size growing under high density plantation were selected and tagged for the present study. The experiment was conducted at Experimental Farm of Division of Fruit Science, Sher-e-Kashmir University of Agricultural Science & Technology of Kashmir, Shalimar, Srinagar, Jammu and Kashmir during 2017-2018. The orchard soil was moderately deep with medium fertility status. Maternal parents *viz,* Carmen (M_1) and Abate Fetel (M_2) were crossed at balloon stage with male parents viz.,William Bartlett (P_1), Fertility (P_2), Clapp's Favourite (P_3), Chinese Sandy Pear (P_4), Max Red Bartlett (P_5), Kings Pear (P_6), Beurred'Amanalis (P_7) besides being self pollinated (P_8) and open pollinated (P_9). The female plants were planted at a spacing of 3m x 3m square system of planting on Quince C rootstock subjected to uniform cultural practices during the study. Pollination was carried out using three different modes *viz.,* self-pollination (bagging was done on unopened flowers and left as such for natural self-pollination), open pollination (three branches on all sides of every tagged tree were left unbagged for open pollination) and hand pollination (the emasculated flowers, covered with bags were pollinated 24 h after emasculation with the pollen of the parent as per crossing plan). Both maternal cultivars were pollinated with different pollen sources constituting the treatment combinations. The design of experiment was RCBD with nine treatments and three replications comprising of 54 cross combinations, number of flowers studied per replication were 25 and a total of 1,350 flowers were crossed.

Observations were recorded on date of swollen bud, date of bud burst, date of green cluster, date of balloon stage, date of initial bloom (10 %), date of full bloom (80 %) and date of complete petal fall. Flowering duration was calculated as the days from initial bloom to complete petal fall. The fruit set (%) was worked out after 21 days of pollination by dividing total number of fruit lets produced to the total number of flowers pollinated and multiplied by 100.

Fruit retention was calculated by dividing the number of fruits harvested to the number of fruits set after 21 days of pollination multiplied by 100. Per cent fruit drop was worked out by subtracting per cent fruit retention from 100 and average was worked out. The date of harvesting was recorded when fruits were harvested after attaining proper size and colour and converted into days which was counted from full bloom.The data recorded were subjected to statistical analysis as per the method of Snedecor and Cochran (1994). The significant difference of the means was tested at 5 % level.

Result and Discussion

Floral phenology

Floral phenology of different varieties involved in the crossing program is presented in Fig. 1. Considerable variations were exhibited by the varieties in attaining the different phenological stages from swollen bud stage to complete petal fall.Earliest swollen bud stage was observed in Clapp's Favourite and Chinese Sandy Pear (13^{th}-14^{th} March) followed by Kings Pear (14^{th}-15^{th} March) whereas Abate Fetel (21^{st}-22^{nd} March) was late in reaching swollen bud stage. Minimum number of days were taken by Chinese Sandy Pear to reach the bud burst stage (18^{th}-19^{th} March) and green cluster stage (21^{st}-22^{nd} March) closely followed by Clapp's Favourite and Kings Pear *i.e.* 19^{th}-20^{th} March and 23^{rd}-24^{th} March in bud burst and green cluster stage. Abate Fetel was late in the commencement of bud burst and green cluster stages *i.e.* 25^{th}-26^{th} March and 28^{th}-29^{th} March, respectively. Chinese Sandy Pear exhibited ballon stage earliest (25^{th}-26^{th} March) which was almost statistically at par with Kings Pear (26^{th}-27^{th} March), however, Abate Fetel variety was late to reach balloon stage (1^{st}-2^{nd} April) (Fig. 1). Earliest initial bloom was observed in Chinese Sandy Pear (29^{th}-30^{th} March) and Kings Pear (30^{th}-31^{st} March) the later variety was earliest to reach full bloom (2^{nd}-3^{rd} April) closely followed by Chinese Sandy Pear (3^{rd}-4^{th} April) whereas, late initial bloom (7^{th}-8^{th} April) and full bloom (11^{th}-12^{th} April) were recorded in Abate Fetel. Chinese Sandy Pear and Kings Pear were earliest in commencement of petal fall (10^{th}-11^{th} April) whereas, late complete petal fall was observed in Abate Fetel (20^{th}-21^{st} April) (Fig. 1). The differences in flower bud development period is due to varietal character which appears to be a principle factor in controlling flower bud development (Anand, 2003).

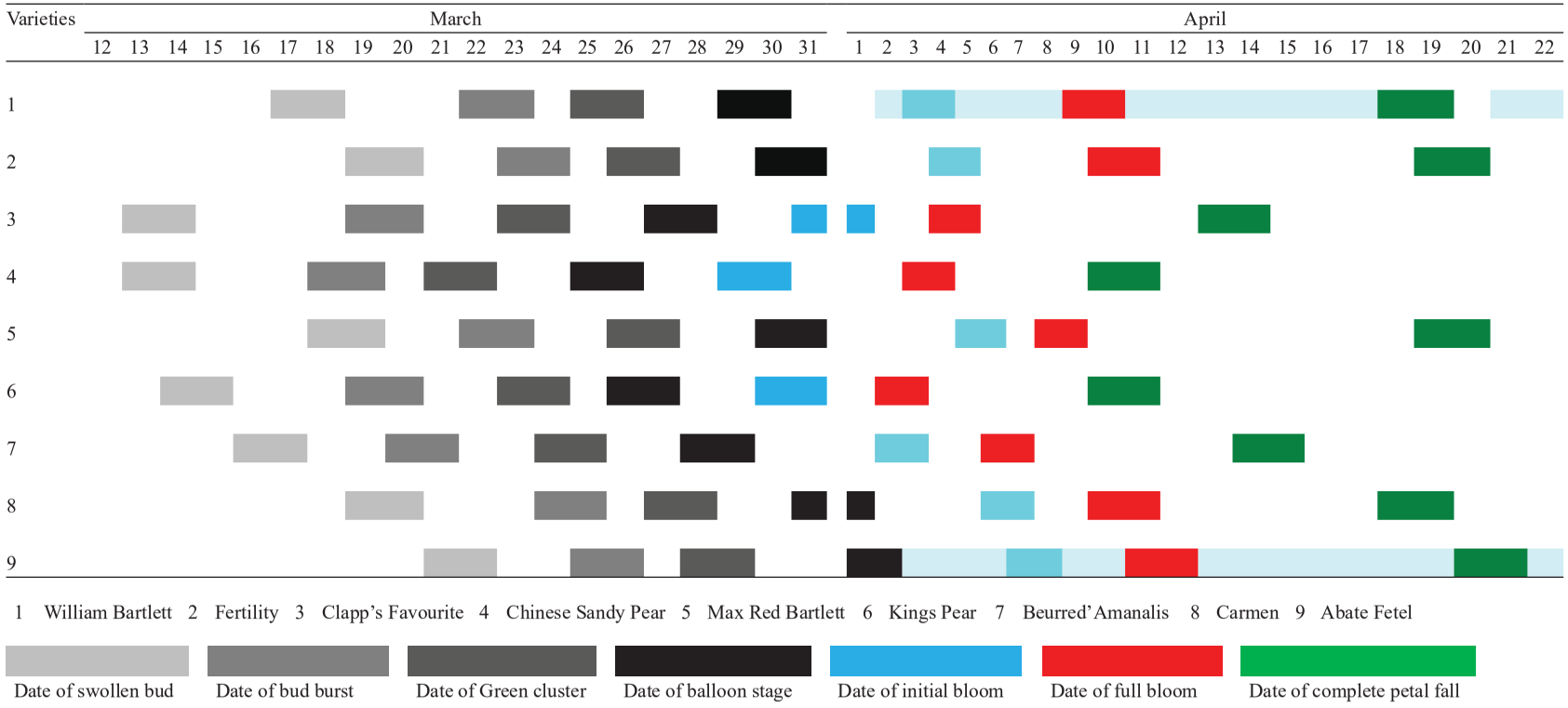

Fig. 1. Floral phenology of pear cultivars involved in crossing plan

The complex mechanisms of chilling requirements and subsequent heat unit accumulation, may affect flowering date and duration of anthesis differently in different cultivars (Malgarejo, 1996). Arzani (2004) supported this concept and stated that different genotypes of Asian pear showed different flowering times and periods. Besides environmental factors like temperature, rainfall and relative humidity may directly or indirectly, singly or collectively play an important role during flower bud development period. Dhillon and Gill (2013) also reported the affect of climatic conditions especially temperature on flowering of hard pear (*Pyrus pyrifolia*).

Figure 2 depicts that maximum flowering duration (17 days) was recorded in William Bartlett and Fertility followed by Max Red Bartlett (16 days) whereas minimum (12 days) duration of flowering was recorded in Chinese Sandy Pear and Kings Pear.Flowering duration a highly variable character primarily is regarded as a varietal character, but temperature has a great effect on duration of flowering. The time of flowering in pear is influenced by chilling requirement for breaking the rest period and heat requirement to develop flower buds to bloom.Variation in duration of flowering between different cultivars may be attributed to differential development of floral parts in various cultivars which is greatly attributed to their genetic difference. Similar variations in flowering duration of different genotypes of pear were also reported by Aulakh *et al.* (1981) who stated that the duration of flowering varied from 21 days in Baggugosha to 29 days in Smith. Dhillon and Gill (2013) also reported flowering duration in hard pear ranging between 14 to21 days in the first year and 9 to11 days in the following year.

Fruit set and retention

Perusal of data presented in Table 1 reveals that maximum mean fruit set (74.00%) was recorded with pollinizer cultivar Fertility followed by William Bartlett (72.83 %) and Max Red Bartlett (72.67 %), while under open pollination mean fruit set of 68.66 % was recorded. Under self-pollination and with the pollinizer cultivar Beurre de Amanalis, zero per cent fruit set was recorded. Interaction combination of 'Carmen × William Bartlett'(81.33 %) recorded the maximum fruit set percentage followed by 'Carmen × Max Red Bartlett' (80.00%) and 'Carmen × Fertility' (78.67%) which was higher than open pollination (74.66 %) whereas, minimum fruit set (54.67%) was recorded in 'Abate Fetel × Kings Pear' combination. It is evident from the Table 1 that pollen source proved a major factor in retaining fruits and number of fruits harvested in both the exotic cultivars. Maximum fruit retention (46.42 %) was recorded with pollen source Fertility followed by William Bartlett (46.21 %) and Max Red Bartlett (46.11 %) and minimum fruit retention percentage (29.80 %) was observed with Kings Pear irrespective of cultivars which was even lower than that obtained from open pollination (38.56%). Maximum fruit retention was obtained in the cross combinations of Carmen × William Bartlett (50.79 %) followed by Carmen × Max Red Bartlett (48.24 %)

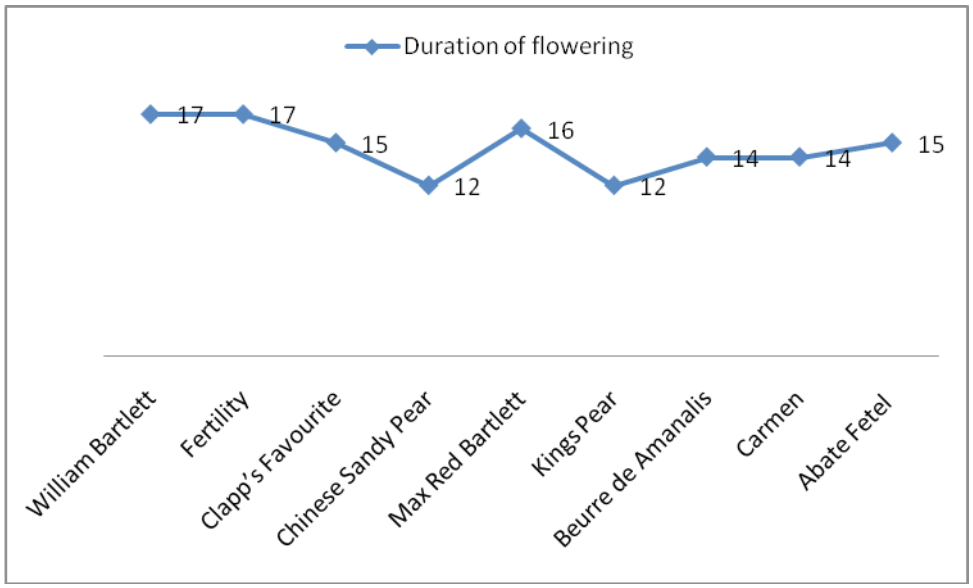

Fig. 2. Flowering duration of different pear cultivars involved in crossing plan

Table 1. Effect of pollen source on fruit set and fruit retention in Carmen and Abate Fetelpears

Maternal parents Pollen source	Fruit set (%)			Fruit retention (%)		
	Carmen	Abate Fetel	Mean	Carmen	Abate Fetel	Mean
William Bartlett	81.33 (64.40)	64.33 (53.12)	72.83 (58.76)	50.79	41.64	46.21
Fertility	78.67 (62.48)	69.33 (56.36)	74.00 (59.42)	47.54	45.31	46.42
Clapp's Favourite	66.67 (54.72)	60.00 (50.76)	63.33 (52.74)	35.90	33.43	34.66
Chinese Sandy Pear	65.33 (53.91)	56.33 (49.20)	60.83 (51.55)	34.72	30.31	32.51
Max Red Bartlett	80.00 (63.48)	65.33 (53.91)	72.67 (58.69)	48.24	43.99	46.11
Kings Pear	57.33 (49.20)	54.67 (47.66)	56.00 (48.43)	30.31	29.30	29.80
Beurred' Amanalis	0.00 (0.01)	0.00 (0.01)	0.00 (0.01)	0.00	0.00	0.00
Self-pollination	0.00 (0.01)	0.00 (0.01)	0.00 (0.01)	0.00	0.00	0.00
Open Pollination	74.66 (59.78)	62.67 (52.32)	68.66 (59.78)	41.02	36.11	38.56
Mean	56.00 (45.33)	48.03 (40.37)		32.06	28.89	
$CD_{0.05}$						
Maternal parent		2.11			0.60	
Pollen source		0.98			1.13	
M × P		2.98			1.60	

Values in the parenthesis are arc sine transformed values

and Carmen × Fertility (47.54 %) whereas, minimum fruit retention was recorded in Abate Fetel × Kings Pear (29.30 %). Under open pollination Carmen and Abate Fetel recorded 41.02 per cent and 36.11 per cent fruit retention, respectively. Conducive climatic conditions during flowering play a vital role in achieving successful pollination and fruit set. Bee activity which is necessary for fruit set may be affected by unfavorable weather conditions during blooming under temperate conditions. Tatari *et al.* (2017) and Cerovic *et al.* (2020) also worked on fruit set and fruit retention using different pollinizers in pear. Difference in fruit set within the same variety using different pollinizers is attributed to degree of compatibility within combinations; the higher compatibility resulted in higher fruit set (Tatari *et al.*, 2017). Bashir *et al.* (2010) also reported differences in fruit retention in apple with different pollinizers and varieties. In addition to the genetic variances, there could be numerous factors responsible for differential fruit set and fruit retention between the varieties. These factors include temperature and other climatic conditions, effective pollination period of varieties, stigmatic receptiveness, ovule longevity, pollen germination, ploidy level of cultivars, post blossom temperatures, time of hand pollination, skill of emasculation and fertilization process (Verma, 1997).

Furthermore, in case of self-pollination in both the cultivars *viz.,* Carmen and Abate Fetel, no fruit set was observed which proves full self-incompatibility. Earlier reports in different pear varieties (Gent Drouard and Fertility) also recorded full self-incompatibility (Qadir, 2007). No fruit set recorded in cvs. Carmen and Abate Fetel under self-pollination and hand cross pollination with Beurred' Amanalis could be due to self-incompatibilityand cross incompatibility. Growth of pollen tube is essential for its entry into the viable ovules for the process of fertilization needed for sufficient fruit set (Sanzol and Herrero, 2001).No fruit set in both the cvs. Carmen and Abate Fetel by pollen source Beurred' Amanalis may be attributed to its triploid nature resulting in production of sterile pollen.

Minimum fruit drop was recorded with pollinizer cultivars Fertility (53.77 %), William Bartlett (53.78%) and Max Red Bartlett (53.88%) being statistically at par with each other (Table 2). Maximum fruit drop (70.19 %) was observed with pollinizer cultivar Kings Pear which was statistically different from other pollinizer cultivars

followed by Chinese Sandy Pear (67.48 %) and Clapp's Favourite (54.33 %). Under open pollination the fruit drop of 61.43 per cent was registered. Among different cross combinations minimum fruit drop was registered in Carmen × Max Red Bartlett (51.75 %) which was statistically at par with Carmen × Fertility (52.45 %) whereas maximum fruit drop was recorded in Abate Fetel × Kings Pear (70.70 %) which was statistically at par with Carmen × Kings Pear (69.68 %) and Abate Fetel × Chinese Sandy Pear (69.68 %). In the earlier reports, Pandit (2014) in apple and Singh *et al.* (2004) in pear reported differences in fruit drop using different pollinizers. Fruit bearing species containing greater than one seed (apple, pear and quince) ideally drop fruits which possess lesser number of seeds. These fruits cannot tolerate the harsh environmental conditions i.e. drought, reduced fertility etc. and hence, more susceptible to fruit drop (Racsko *et al.*, 2007).

The analyzed data regarding fruit maturity as influenced by different pollen source presented in Table 2 reveal that pollen source had a direct effect on number of days required by a variety to reach its harvestable stage. Carmen was earliest in maturation of fruits which took significantly lesser number of days (108.07) after full bloom to reach the harvest maturity compared to Abate Fetel which took 142.42 days after full bloom to reach maturity. Maximum days (127.10) after full bloom required to reach the harvestable stage were observed with pollinizer cultivar William Bartlett followed by Max Red Bartlett (126.88 days) whereas minimum days to harvest were registered in Clapp's Favourite (123.49 days) as pollen source .Maximum numbers of days (145.00) to maturity were recorded in Abate Fetel under open pollination and minimum number of days (105.21) were taken by Carmen under open pollination to reach maturity. Among cross combinations the maximum number of days (143.55) were taken by Abate Fetel × William Bartlett and minimum number of days (106.66) were recorded in Carmen × Clapp's Favourite combination to attain the harvest maturity. The remarkable difference between cultivars in days taken to maturity may be due to their distinct genetic makeup and intrinsicpaternal character. Present results are in agreement with the results of Pandit (2014) who reported difference in fruit maturity of apple using different pollinizers. Moreover, the variation among the different treatment combinations is due to metaxenic effect of pollen as it is clearly evident from the data that some pollinizers hastened the maturity while others delayed it (Ghnaim and Al-Muhtaseb, 2006).

Conclusion

For getting higher yields, high fruit set is required which is achieved by managing successful pollination and fertilization, at the right time. In the present study both pear cultivars Carmen and Abate Fetel exhibited gametophytic self-incompatibility, however, cross compatibility with different pollinizer cultivars was evidenced by fruit set. All the pollinizer cultivars used in present study were found to be the effective for pollination of both the cultivars (Carmen and Abate Fetel) except Beurre de Amanalis which showed cross

Table 2. Effect of pollen source on fruit drop and days to fruit maturity in Carmen and Abate Fetel pears

Maternal parents Pollen source	Fruit drop (%)			Days to fruit maturity (%)		
	Carmen	Abate Fetel	Mean	Carmen	Abate Fetel	Mean
William Bartlett	49.20	58.36	53.78	110.66	143.55	127.10
Fertility	52.45	54.69	53.57	107.21	141.00	124.10
Clapp's Favourite	64.09	66.57	65.33	106.66	140.33	123.49
Chinese Sandy Pear	65.28	69.68	67.48	107.66	141.33	124.49
Max Red Bartlett	51.75	56.01	53.88	110.66	143.10	126.88
Kings Pear	69.68	70.70	70.19	108.44	142.66	125.55
Beurred' Amanalis	0.00	0.00	0.00	0.00	0.00	0.00
Self pollination	0.00	0.00	0.00	0.00	0.00	0.00
Open Pollination	58.97	63.89	61.43	105.21	145.00	125.10
Mean	45.71	48.87		108.07	142.42	
$CD_{0.05}$						
Maternal parent		0.54			0.40	
Pollen source		1.01			0.71	
M × P		1.40			1.00	

incompatibility with both the cultivars due to triploid genotypic constitution nature (pollen sterility). However, best pollinizer cultivars in terms of flowering duration andcompatibility were Fertility, William Bartlett and Max Red Bartlett. These pollinizer cultivars proved efficient for both the cultivars under study as the blooming period of these synchronizes with the blooming period of the cultivars under study, which is a prerequisite for effective pollination and also improved fruit set and retention and ultimately fruit yield.

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SHORT COMMUNICATION

Preliminary Evaluation of Some Cape Gooseberry (*Physalis peruviana* L.) Genotypes under Jammu Plains

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The present investigation aimed to assess 10 genotypes of cape gooseberry at SKUAST Jammu. A evaluation was conducted using a randomized complete block design in three replications. The significant amount of variation was observed among the genotypes for all the studied traits. High magnitude of genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) values were observed for resistance against fruit cracking, acidity, pectin, total sugar, number of days to fruit set, fruit length, fruit breadth, yield per plant which indicates the presence of high genetic variation. The PCV was estimated to be higher than the GCV for all the observed traits. However, close estimates of GCV and PCV revealed that genetic variance played a major role in the phenotypic expression of most of the traits. As a result phenotypic selection is both effective and desirable. The magnitude of heritability ranged from 35.47% to 95.88% indicating the presence of additive gene action and the need for population improvement through selection. Considerable amount of genetic variability among the genotypes indicating greater potential of the genotypes for their exploitation to improve yield and its component traits. The genotypes have great potential for further improvement.

Key Words: Cape gooseberry, GCV, Genetic diversity, Heritability, PCV

Introduction

Cape gooseberry (*Physalis peruviana* L.) is a herbaceous under-exploited, exotic fruit crop grown for edible fruits. The genus *Physalis* belonging to the family Solanaceae is among the largest genera in subfamily Solanoideae with about 100 species. Cape gooseberry has entered the small fruits ranking and has also shown great promise for the national and international markets, with a high value as a fresh fruit with a unique flavour that appeals to the consumers for this berry (Rodrigues *et al*., 2014). It is also known as Jam fruit due to high pectin content, flavour, aroma, colour as well as nutritional importance and various health benefits (Puente *et al*., 2011). In India, it is grown successfully in some states like Madhya Pradesh, Uttar Pradesh, Haryana, Punjab, Nilgiri hills, West Bengal, and in some other parts of country. There is good scope and potential of this nutritive annual berry to be grown under the subtropical conditions of Jammu plains. This berry is gaining special attention particularly due to its high productivity, availability in lean period, wider adaptability, quick growing in nature, non-perennial occupation of land and luscious fruit with pleasant acidic taste. Introduction and evaluation is one of the important methods for bringing improvement in any fruit crop and for the selection of parents in a viable hybridization programme. The degree of genetic diversity influences the planning and execution of any breeding effort aimed at improving quantitative characters. As a result, plant breeding success is entirely dependent on the presence of genetic variability in desired traits and plant breeder selection skills (Tiwari *et al*., 2018). Hence, the knowledge of genetic variability, genetic advance and heritability are the key foundations for the improvement of the traits. Therefore, the present investigation was carried out to study the genetic variability, genetic advance and heritability in different genotypes of cape gooseberry under Jammu plains.

Materials and Methods

The present investigation was conducted at Research Farm, Division of Fruit Science, Faculty of Agriculture, Sher-e-Kashmir University of Agricultural Sciences and Technology, Jammu during the year 2020-2021. The experimental site is located at an elevation of 260 m above mean sea level in subtropical zone at 32º39᾽ N latitude

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and 74º48' E longitude. The climate of the experimental site is subtropical with hot and dry summer, hot and humid rainy season and cold winter months. The mean annual maximum temperature during summer rises up to 43^0C and minimum temperature during winter falls up to 2.4 ºC. Annual precipitation in this area is about 1,000-1,200 mm mostly during July to October (about 70 per cent). The experiment was laid down in Randomized Block Design with three replications. The experimental material consists of ten genotypes of cape gooseberry (*Physalis peruviana* L.) viz. CITH CGB Sel-1, CITH CGB Sel-2, CITH CGB Sel-3, CITH CGB Sel-4, CITH CGB Sel-5, CITH CGB Sel-6, CITH CGB Sel-7, CITH CGB Sel-8, CITH CGB Sel-9 and CITH CGB Sel-10 exotic material procured by ICAR-Central Institute of Temperate Horticulture (CITH) Srinagar. The seeds sown in the month of June in well prepared and raised seed beds and partially covered with paddy straw. The beds were irrigated on alternate days. When the plants attained the height of about 20 cm, the transplanting of seedlings was done in well prepared seed beds in each replication. Plant to plant and row-row spacing was 1m×1m. The transplanting was done on rainy season on 25th July, 2020. A total of 22 traits were analyzed. Five plants were randomly selected and tagged in each replication of the treatment for data collection. The data obtained were statistically analyzed using software Windostat ver. 9.3.

Genotypic and Phenotypic Coefficient of Variation

According to the formula given by Burton and Devane (1952) the genotypic and phenotypic coefficient of variation were calculated:

$$GCV\ (\%) = \frac{(\text{Genotypic variance})^{1/2}}{X} \times 100$$

$$PCV\ (\%) = \frac{(\text{Phenotypic variance})^{1/2}}{X} \times 100$$

GCV = Genotypic coefficient of variation

PCV = Phenotypic coefficient of variation

X = Mean of the character

Heritability

Heritability in broader sense (h^2b) defined as the proportion of the genotypic variance to the phenotypic variance. It was calculated using the following formula given by Allard (1960).

$$h^2b = \frac{\sigma^2g}{\sigma^2p} \times 100$$

σ^2g = genotypic variance

σ^2p = phenotypic variance

Genetic Advance (GA)

Genetic Advance was calculated by following formula given by Miller *et al*. (1958).

$GA = K\ \sigma p\ h^2$

K = Constant (standard selection differential) having value of 2.06 at 5% selection intensity

σp = Phenotypic standard deviation

h^2 = Heritability estimates

3.9.5 Genetic advance (per cent of mean)

$$\text{Genetic advance (\% of mean)} = \frac{\text{G.A.}}{X} \times 100$$

GA = Genetic advance

X = Mean of the character

Results and Discussion

The extent of variability among the different parameters in 10 genotypes of cape gooseberry are presented in Table 1. The results revealed that highest genotypic variance was observed for date of harvest (200.91) followed by date of initial fruit set (92.58), date of initiation of flowering (78.26), plant height (46.15), ascorbic acid (11.17), leaf area (10.94), total sugar (3.86), TSS (1.18), reducing sugar (1.06), non reducing sugar (0.94), fruit volume (0.94), shoot number (0.74), resistance to viral infection (0.44), fruit weight (0.38), resistance to fruit cracking (0.27), fruit length (0.12), acidity (0.05), pectin (0.04), carotenoids (0.01), yield (0.01) and the lowest was observed for stem thickness (0.003). The phenotypic variance was also highest for date of harvest (273.42) followed by date of initial fruit set (122.83), date of initiation of flowering (94.28), plant height (81.28), leaf area (30.84), ascorbic acid (14.73), total sugar (4.23), fruit volume (1.85), TSS (1.65), shoot number (1.45), reducing sugar (1.12), non reducing sugar (0.98), fruit weight (0.95), resistance to viral infection (0.48), resistance to fruit cracking (0.28), fruit breadth (0.24), fruit length (0.18), acidity (0.05), pectin (0.04), carotenoids (0.02), yield (0.01) and the lowest was recorded in stem thickness (0.01). Genotypic and Phenotypic coefficient of variability were higher in case of resistance against fruit cracking (39.40 and 39.77 respectively) followed by titratable acidity (33.50 and 34.31), non reducing sugar (24.66 and 25.18), reducing

Table 1. Descriptive statistics for extent of variability in traits among different genotypes of Cape Gooseberry (*Physalis peruviana* L.)

Parameters	Range			Coefficient of variation (%)		Heritability (h^2 %)	Genetic Advance	Genetic Advance (% of Mean)
	Minimum	Maximum	Mean value	GCV	PCV			
Stem thickness (cm)	2.95	3.15	3.03	1.78	2.91	37.31	0.07	2.24
Shoot number	13.61	16.07	14.77	5.82	8.17	50.75	1.26	8.54
Plant height (cm)	90.33	114.66	98.51	6.90	9.15	56.78	10.55	10.71
Leaf area (cm^2)	51.40	62.02	57.32	5.77	9.69	35.47	4.06	7.08
Time to initiate flowering (days)	31.69	62.20	54.24	16.31	17.90	83.00	16.60	30.61
Time of fruit set (days)	38.48	72.07	62.32	15.44	17.78	75.38	17.21	27.61
Time of harvesting (days)	86.50	134.28	119.08	11.90	13.89	73.48	25.03	21.02
Fruit weight (g)	11.10	13.37	12.29	5.34	7.94	40.27	0.81	6.59
Fruit length (cm)	2.00	3.04	2.42	14.60	17.57	69.10	0.61	25.01
Fruit breadth (cm)	2.31	3.47	2.82	15.21	17.52	75.39	0.77	27.20
Fruit volume (cc)	11.15	14.71	12.53	7.73	10.86	50.72	1.42	11.35
Yield (kg/plant)	0.88	1.26	1.11	8.87	12.55	49.99	0.14	12.92
TSS (º Brix)	9.48	12.61	11.15	9.74	11.54	71.27	1.89	16.94
Acidity (%)	0.41	1.10	0.71	33.50	34.31	95.37	0.48	67.40
Carotenoids (mg/100g)	1.13	1.60	1.39	9.81	12.33	63.32	0.22	16.09
Pectin (%	0.53	1.02	0.83	24.46	25.52	91.84	0.40	48.28
Ascorbic acid (mg/100g)	22.41	31.12	26.40	12.66	14.54	75.81	5.99	22.70
Reducing sugar (%)	2.68	5.48	4.22	24.47	25.13	94.85	2.07	49.10
Non reducing sugar (%)	2.55	5.27	3.94	24.66	25.18	95.88	1.96	49.74
Total sugar (%)	5.22	10.75	8.15	24.10	25.26	91.07	3.86	47.39
Resistance to viral infection (%)	0	3.70	2.94	22.55	23.79	89.86	1.29	44.03
Resistance to cracking (%)	0	2.00	1.33	39.40	39.77	98.18	1.07	80.43

sugar (24.47 and 25.13), pectin (24.46 and 25.52), total sugar (24.10 and 25.26), resistance to viral infection (22.55 and 23.79), date of initiation of flowering (16.31 and 17.90), date of initial fruit set (15.44 and 17.78), fruit breadth (15.21 and 17.52), fruit length (14.60 and 17.57), ascorbic acid (12.66 and 14.54), date of harvest (11.90 and 13.89), carotenoids (9.81 and 12.33), TSS (9.74 and 11.54), yield (8.87 and 12.55), fruit volume (7.73 and 10.86), plant height (6.90 and 9.15), shoot number (5.82 and 8.17), leaf area (5.77 and 9.69) whereas, the minimum GCV and PCV was observed in stem thickness (1.78 and 2.91). Similar results were also obtained by Prajapati *et al*. (2015) in tomato. Kerketta and Bahadur (2019) also reported that highest magnitude of GCV and PCV in tomato was observed in acidity (28.21 and 42.89 respectively) followed by TLCV incidence (28.04 and 40.51, respectively). The PCV was higher than GCV for all the traits. Results showed that there is a narrow difference between genotypic and PCV for traits such as stem thickness, date of initiation of flowering, date of harvest, TSS, titratable acidity, pectin, ascorbic acid, reducing sugar, non reducing sugar and total sugar which indicates that environment has less influence on expression of these traits. Hence, it can be concluded that genotypic variability had more contribution towards total variance indicating the good scope for crop improvement and selection among the genotypes. Higher GCV and PCV was observed for traits like shoot number, plant height, leaf area, fruit weight, fruit length, fruit breadth, fruit volume, yield indicating the higher magnitude of variability among these parameters. Robinson (1966) classified the estimate of heritability into three categories i.e. low (5-10%), medium (10-30%) and high (30% and above). In the present investigation all the traits showed high estimate of heritability which ranged from 35.47 per cent to 98.17 %. The high magnitude of heritability estimate in broad sense is useful in selection of superior genotypes but heritability combined with genetic advance are more effective for selection of best genotype than the heritability values alone. The traits of resistance to fruit cracking, titratable acidity, pectin, reducing sugar, non reducing sugar, total sugar and resistance against viral infection showed high heritability along with high genetic advance as percent of mean indicating that these traits are controlled by additive gene action which is a very important tool for selection and crop improvement while the traits including date of initiation of flowering, date of initial fruit set, date of harvest, yield, fruit length,

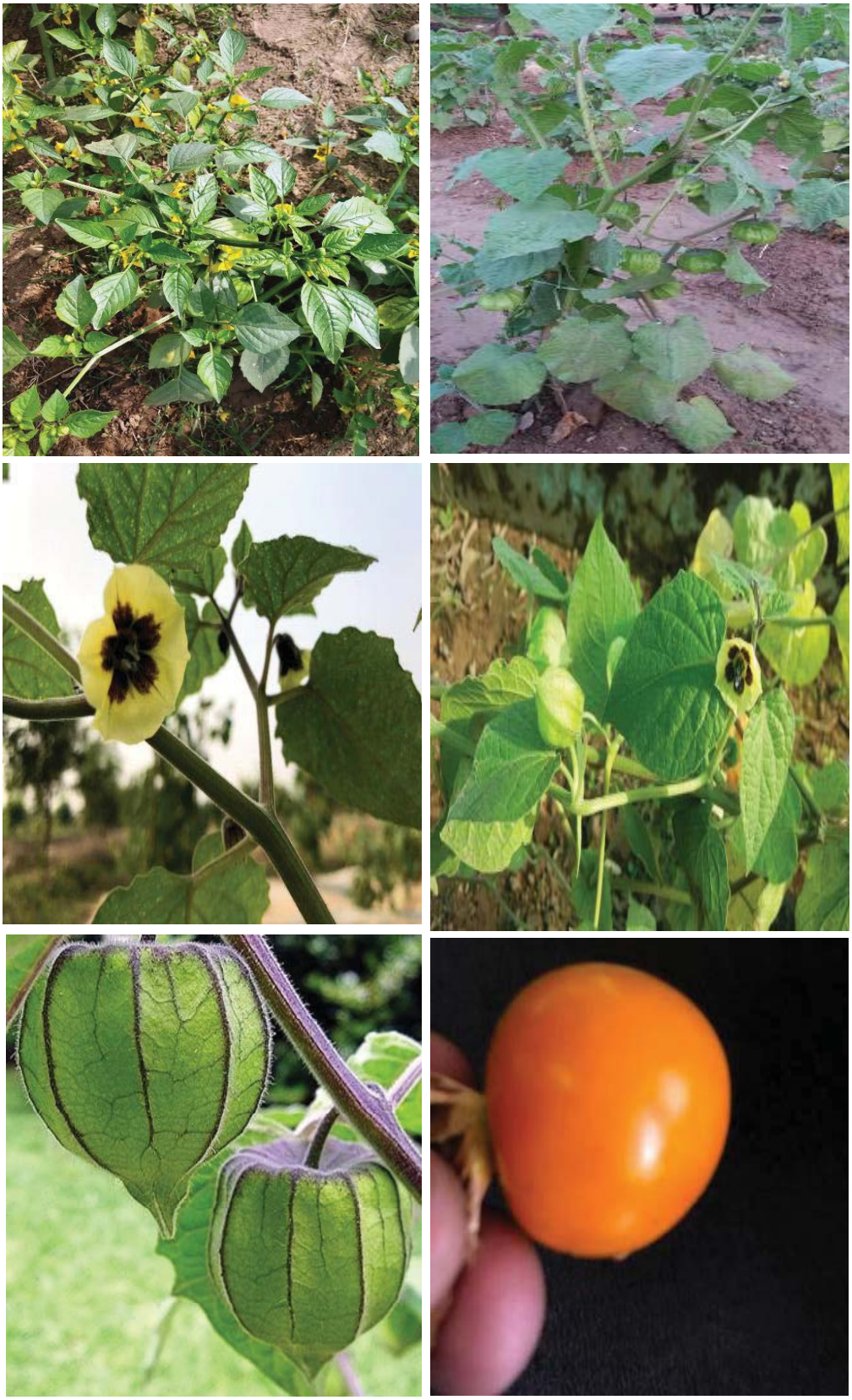

Fig. 1. Vegetative, flowering and fruiting in CITH CGB Sel-10 genotype

fruit breadth and fruit volume showed high heritability along with moderate genetic advance as percent of mean which indicates the presence of additive and non additive action of the genes and the phenotypic expression might be largely affected by the non additive genes. Heritability values were higher than those of genetic advance for all traits which indicated that they were least influenced by environment changes and showed that genotypes were true representative of their genotypes and selection based on phenotypic performance would be reliable. Similar results were also reported by Meena *et al*. (2018) in tomato. In conclusion it is evident that considerable genotypic variation among the genotypes indicating greater potentiality for their exploitation to improve yield and its component traits. There was a good scope for selection also. The overall performance in relation to fruit yield and weight was best in CITH CGB Sel-9 and CITH CGB Sel-10 genotypes. However further work is warranted in this regard.

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Plant Germplasm Registration Notice*

The Plant Germplasm Registration Committee of ICAR in its XXXXIIInd meeting held on December 21st, 2020 at the National Bureau of Plant Genetic Resources, New Delhi approved the registration of following 39 germplasm lines out of 43 proposals considered. The information on registered germplasm is published with the purpose to disseminate the information to respective breeders for utilization of these genetic stocks in their crop improvement programmes. Upon request, the developer(s)/author(s) is/are obliged to distribute the material for crop improvement programme of National Agricultural Research System.

1. SC-11/SP70/TI-26/SB-8 (IC0635696; INGR20079), a Rice (*Oryza sativa*) Germplasm with Higher Culm Strength in Elite Genetic Background of Samba Mahsuri

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Strong culm is one of most important agronomical trait and directly involved in lodging resistance. Lodging resistance is a complex trait, determined by culm diameter, strength and elasticity of culm. To obtain novel genetic sources for strong culm, 10,500 EMS induced mutant population was developed in the genetic background of Samba Mahsuri in collaboration with CSIR-CCMB. The mutant, TI-26 (SP-70/SB-8/SC-11) had strong culm which can be used as donor material for rice improvement. The selection of strong culm mutant SC-11 was done in M2 generation after screening every season, selected single strong culm plant and advanced to M8 through panicle to row method.

Morpho-agronomic characteristics: The mutant, TI-26 (SP-70/SB-8/SC-11) was evaluated for their agro-morphological variations at Indian Institute of Rice Research (ICAR-IIRR); ICRISAT, Hyderabad and Kakanoor village, Ranga Reddy district during years 2013-2016. The important agro morphological traits are given in the table.

Agro morphological traits

Characteristics	Description
Plant ht (cm)	105
No of tillers	18
No of panicles	18
Panicle length (cm)	22
Grains per panicle	220
Yield / plant (g)	21.2
Days of flowering (50%)	100
Grain type	Medium bold

Associated characters and cultivation practices Phenotyping for Strong Culm

TI-26 (SP-70/SB-8/SC-11) was selected during the years of 2012-2016 by estimating the culm strength and diameter using prostate tester. SC-11 has more culm diameter and culm strength (11.5 mm, 31.5 Nu/m^2) than the wild type (5.1 mm, 25 Nu/m^2). Anatomical characters studied under scanning electron microscope (SEM) revealed an increase in the thickness of lignified epi/sub epidermal and lignified parenchymatous tissue layers and a decrease in the distance of inter vascular bundles which explain the strong culm nature of the mutant. Strong culm mutant also showed higher photosynthetic rate (9.8 µ mol CO_2 m^2S^{-1}) as compared to wild type (3.5 µ mol CO_2 m^2S^{-1}). Whereas, transcription rate of the mutant was lower (0.40 µ mol H_2O m^2 S^{-1}) as compared to wild type (1.80 µ mol H_2O m^2 S^{-1}). (IIRR newsletter, 2016; Gopi *et al*., 2019 presented poster at NCIPBB conference).

[*]*Compiled and edited by: Anjali Kak and Veena Gupta, Division of Germplasm Conservation, ICAR-National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110012*

Table 1. Summary of the molecular analysis using SSR markers

Designation	Markers per chromosome												Total	% of genomic similarity
	1	2	3	4	5	6	7	8	9	10	11	12		
TI-26	13	11	10	11	10	12	11	10	11	10	11	10	130#	97% genomic similarity with wild type
	13	11	10	11	8	10	11	10	11	10	11	10	126*	

is the total number of markers tested
* is the total number of monomorphic markers with wild type (BPT 5204)
Highlighted chromosomes has polymorphic regions

Genotyping: To know the allelic status of reported strong culm genes (*SCM2* and *SCM3*), PCR based allele mining was performed and determined that this mutant has novel allele than the reported alleles (Gopi *et al.,* 2019a (poster at NCIPBB conference)). For the estimation of genomic similarity with wild type a total of 130 SSR markers (selected based on a ~1 marker per 5Mb) which were spread uniformly across the twelve linkage groups were used. The results indicated that TI-26 (SP-70/ SB- 8/SC-11) has 97% of genomic similarity with wild type (BPT 5204) (Table 1). To identify mutated loci for conferring the culm strength, MutMap (NGS based approach) analysis was performed in strong culm and weak culm bulks of F2 population derived from BPT 5204 × TI-26. The sequence reads of strong culm bulk was compared with the wild type as well as reads of weak culm bulk was compared with the strong culm mutant. Analysis of SNP index plots (prepared based on the comparison of strong culm bulk with wild type) indicated two peak regions on chromosome 5 (27.7Mb-29.2Mb) & chromosome 6 (6.2Mb-10.8Mb), the peak at chromosome 6 matched with the QTLs (qSC-2 & qSC3) identified using F2 mapping population derived from TI-26 × TN1 (diverse parent) (Annexure B; Figure 3). Validation of identified SNPs, thereby identification of candidate gene(s) having correlation to the culm strength is in the progress (Gopi *et al*., 2019 (poster presented at NCIPBB conference). This identified mutant (SC-11) having strong culm is a novel genetic resource. Since, it shows strong culm nature which was deciphered through physical analysis, anatomical features and molecular characterization. Thus, SC-11 can be used as a donor in rice improvement programmes for imparting strong culm trait.

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Gopi P, GS Laha, B Vishalakshi, K Divya, L Swathi, PV Srividya, B Suneel, RM Sundaram, HK Patel, RV Sonti and MS Madhav (2019b) Identification of novel genetic resources for sheath blight tolerance from mutant lines of samba mahsuri presented poster at NCIPBB conference during November 08-09, 2019 at Hyderabad, India.

2. ShB-1/SB-5 (IC0635695; INGR20080), a Rice (*Oryza sativa*) Germplasm Highly Tolerant to Sheath Blight. Medium Slender Grain Type in Genetic Background of Samba Mahsuri.

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Sheath blight is a major biotic stress in rice. There are no absolute resistance sources are available in the germplasm. To obtain novel resistant/tolerant sources for sheath blight, 10,500 EMS induced mutant population was developed in the genetic background of Samba Mahsuri in collaboration of ICAR-IIRR and CSIR-CCMB. The selection of sheath blight tolerant mutant (ShB-1/SB-5) was done at M_2 generation after screening multiple seasons, selected single tolerant plant is advanced to M_8 through panicle to row method.

Morpho-agronomic characteristics: The mutant ShB-1 was evaluated for the agro-morphological variations at Indian Institute of Rice Research Farm (ICAR-IIRR) during 2016 (Gopi *et al*., 2017a & b). All the cooking and eating quality characteristics were similar to that of Samba Mahsuri (wild type). The important agro morphological traits were given below

Characteristics	Description
Plant ht (cm)	85
No of tillers	13
No of panicles	13
Panicle length (cm)	16.2
Grains per panicle	210
Yield / plant (g)	16.2
Days of flowering (50%)	108
Grain type	Medium slender

Associated characters and cultivation practices

Phenotyping for Sheath blight tolerance: ShB-1 mutant was screened for sheath blight tolerance under field and laboratory conditions (detached leaf method) during the years of 2013-2015. The mutant ShB-1 showed mean score of '0' for sheath blight tolerance whereas Samba Mahsuri (wild type), showed mean score of 9.0 (Gopi *et al*., 2017a). ShB-1 also screened with 10 most virulent and diverse *R. solani* isolates (TN14-1, RNR 13-F, TTB-1, WGL-12-1, Gosaba-1, Kaual, PNT, Jamalpur-Bangar and Imphal-1) collected from various hotspot regions of India and showed resistance with a mean score of 2.84 and whereas wild type, TN1 (susceptible check) and Tetep (resistant check) showed mean score of 9.0, 9.0 and 4.45, respectively. Further, this mutant line screened at four hotspot locations (Kaul, Pantnagar, Chinsura and Monkompu) of India, which revealed mean score of 3.0. The identified mutant line can be served as best genetic resource in rice breeding programme as there are no reports on existence of absolute resistance genetic material in rice.

Genotyping: For the estimation of genomic similarity with wild type, a total of 130 SSR markers, which spread uniformly across the genome revealed 97% genomic similarity with wild type. Through MutMap analysis mutated locus on chromosome-1 was identified.

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3. GW 2012-475 (IC0633421; INGR20081), an Early Maturity and High Yielding Wheat (*Triticum aestivum*) Germplasm

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Wheat is the second most important crop after rice both in terms of area and production in India. India produced a record 101.20 million tonnes of wheat from29.55 million hectares with a productivity of 34.24 q/ha in the year 2018-19 (ICAR- IIWBR, 2019). Wheat, a temperate crop, prefers a cooler climate for growth and reproduction. High temperatures during crop growth and grain filling stages are a major concern to its production. South Asia, comprising of India, Nepal, Pakistan and Bangladesh is one of the major wheat producing and consuming area in the world. This region suffers significant losses each year due to high temperature stress (Kumar *et al*., 2013). Earliness or early maturity is an adaptation strategy where early heading lines complete the initial seed setting and grain filling under favorable temperatures and avoid the late incidence of heat stress. Earliness has been suggested as a good approach for wheat breeding in the eastern Gangetic plains that suffers from high temperature stress during grain filling (Joshi *et al*., 2007). Improving the genetic adaptation of wheat cultivars to heat stress is an important objective in breeding programme. Looking to this need breeding work for early maturing and high grain yield was started at Wheat Research Station, Vijapur. Considering this aspect, cross was made using diverse genotypes (MACS 2496/CMH83.2578//GW 496/ WH 147//GW 496) and generation advancement was made using pedigree selection method (Allard, 1960). This genotype has been developed at Wheat Research

Station, SD Agricultural University at Vijapur for early maturity.

Morpho-agronomic characteristics: GW 2012-475 has semi-erect growth habit with medium broad leaves with 60 days to complete heading stage. During the period of testing, it was tested at national level under short duration screening nursery (SDSN) during the 2015-16 to 2017-18 (ICAR-IIWBR. 2019) in the central zone, in which GW 2012-475 found to be superior with 5.8 and 5.8 per cent higher grain yield over the years and locations to the check variety Sonalika, and WR 544, respectively.

Table 1. Performance of GW 2012-475 genotype during 2015-16 to 2017-18

Trait	Year	Genotype	Checks		
		GW 2012-475	Sonalika	HD 2932	WR 544
Mean Yield (g/plot)	2015-16	486	442	406	436
	2016-17	465	485	520	465
	2017-18	518	461	517	488
	Average	490	463	481	463
Heading days	2015-16	61	59	62	55
	2016-17	59	60	62	58
	2017-18	61	58	62	60
	Average	60	59	62	58
Maturity days	2015-16	109	110	109	105
	2016-17	109	112	112	107
	2017-18	113	112	114	111
	Average	110	111	112	108
Grains/spike	2015-16	44	34	40	35
	2016-17	39	42	45	44
	2017-18	52	43	49	49
	Average	45	40	45	43
1000 gr.wt (g.)	2015-16	43	44	42	41
	2016-17	44	46	43	43
	2017-18	40	42	38	38
	Average	42	44	41	41

Associated characters and cultivation practices: GW 2012-475 has medium broad leaves and flag leaf attitude is semi erect type, where as grains are bold, amber colour and oblong shape. Ear is having parallel shape and strong waxiness on ear and peduncle. It matures within 110 days; the agronomic practices for cultivation are to be followed as per late sown irrigated condition. This genotype is adapted to central zone.

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4. GW 2010-321 (IC0633420; INGR20082), an Early Maturing and High Yielding Wheat (*Triticum aestivum*) Germplasm

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Wheat is the second most important crop after rice both in terms of area and production in India. India produced a record 101.20 million tonnes of wheat from 29.55 million hectares with a productivity of 34.24 q/ha in the year 2018-19 (ICAR-IIWBR, 2019). Wheat, a temperate crop, prefers a cooler climate for growth and reproduction. High temperatures during crop growth and grain filling stages are a major concern to its production. South Asia, comprising of India, Nepal, Pakistan and Bangladesh is one of the major wheat producing and consuming area in the world. This region suffers significant losses each year due to high temperature stress (Kumar *et al*., 2013). Earliness or early maturity is an adaptation strategy where early heading lines complete the initial seed setting and grain filling under favorable temperatures and avoid the late incidence of heat stress. Earliness has been suggested as a good approach for wheat breeding in the eastern Gangetic plains that suffers from high temperature stress during grain filling (Joshi *et al*., 2007). Improving the genetic adaptation of wheat cultivars to heat stress is an important objective in breeding programme. Looking to this need breeding work for early maturing and high grain yield started at Wheat Research Station, Vijapur. Considering this aspect, cross was made using diverse genotypes (GW 366/HW 1042//KAUZ*2//TC*6/RL 6081/3/KAUZ) and generation advancement was made using pedigree selection method (Allard, 1960). This genotype has been developed at Wheat Research Station, S.D. Agricultural University at Vijapur for early maturity.

Morpho-agronomic characteristics: GW 2010-321 has erect growth habit and it's mature within 108 days. During the period of evaluation, it was tested at national level under short duration screening nursery (SDSN) during the 2015-16 to 2017-18 (ICAR-IIWBR, 2019) in the North eastern plain zone (NEPZ), in which GW 2010-321 found to be superior with 30.8, 26.5, and 16.1 per cent higher grain yield over the years and locations to the check variety Sonalika, DBW 14 and WR 544 respectively.

Table 1. Performance of GW 2010-321 genotype during 2015-16 to 2017-18

Trait	Year	Genotype	Checks		
		GW 2010-321	Sonalika	DBW 14	WR 544
Mean Yield (g/plot)	2015-16	290	215	254	247
	2016-17	531	434	455	504
	2017-18	541	393	368	422
	Average	454	347	359	391
	% increase over check	-	30.8	26.5	16.1
Heading days	2015-16	67	66	66	63
	2016-17	69	66	65	63
	2017-18	70	67	70	66
	Average	69	66	67	64
Maturity days	2015-16	104	102	105	104
	2016-17	108	105	105	104
	2017-18	112	108	109	107
	Average	108	105	106	105
Grains/spike	2015-16	51	43	47	44
	2016-17	53	42	41	42
	2017-18	40	40	42	44
	Average	48	42	43	43
1000 gr. wt (g.)	2015-16	35	37	38	37
	2016-17	37	41	38	40
	2017-18	36	38	37	38
	Average	36	39	38	38

Associated characters and cultivation practices: GW 2010-321 has medium broad leaves and flag leaf attitude is semi erect type, whereas grains are medium, amber colour and oblong shape. Ear is having tapering shape and strong waxiness on peduncle. This genotype posses 48 grains per spike which is 10.41% higher over best checks DBW 14 and WR 544. It matures within 108; the agronomic practices for cultivation are to be followed as per late sown irrigated condition. This genotype is adapted to North eastern plain zone (NEPZ).

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5. UPB 1065 (IC0635429; INGR20083), a Barley (*Hordeum vulgare*) Germplasm with Low Beta Glucan Content (<3.5%) – Malt Quality Trait. High Filtration Rate and Kolbach Index. High Yield and Resistance to Yellow Rust.

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Barley is an important crop for the production of malt products such as beer, health drinks etc. β-glucans content is an indicator of malt modification and quality in barley. Beta glucans (β-D-(1-3), (1-4) glucans) are major constituents of barley endosperm walls (Palmer, 1989). In the process of barley malt production and beer brewing, the incomplete degradation of endosperm cell wall causes excessive wort beta glucan (WBG), which would influence the expansion of hydrolase and protease into the malt cells and decrease the extract content in the wort. Excessive residual β-glucan in the malt will lead to an increase of (VIS) viscosity, which is not conducive to the filtration of wort and beer, and results in reduced malt quality. In addition to viscosity, β-glucans has a relationship with other malt quality traits such as speed of filtration and Kolbach index and may affect extract value. Low level of glucans upto 3.5 is regarded as a good malting trait (Kumar *et al*., 2017). Since, there is scarcity of genotypes with low levels of β-glucans, thus the genotype UPB 1065 with a β-glucans content <3.5 can serve as an important source for malt barley breeding programme for incorporating low β-glucans, which is required for good malting quality. UPB 1065 has been developed through selection breeding method from ICARDA material at G.B. Pant University of Agriculture and Technology, Pantnagar, Uttarakhand in 2016-17.

Morpho-agronomic characteristics: UPB 1065 is a six rowed hulled barley genotype with a range of 58-89 days to heading, 118-133 days to maturity, 85-113 plant height, 71-125 tiller per meter, yellow grain color and thousand grain weight of 35-48 gm. It has been tested in AICRP Co-ordinated Trials (IVT-MB-TS, NWPZ) during 2016-17 and recorded an average yield of 46.0 quintals per hectare. Malt quality tests of IVT malt barley trial in North Western Plains (IVT-MB-TS, NWPZ) showed that UPB 1065 has low β-glucan content i.e. 3.3% as compared to checks during 2016-17. The genotype was also included in National Barley Genetic stock nursery (NBGSN, 2017-18) as a source for low beta glucan. It was further tested at IIWBR, Karnal in 2018-19 and revealed low β-glucan content i.e. 3.5%.

Associated characters and cultivation practices: In addition to β-glucan content, UPB 1065 possess other malt quality traits like, high filtration rate and kolbach index, which affects extract value. Generally, good malt quality lines have poor resistance to yellow rust, a serious disease of barley. High degree of resistance to yellow rust was observed in UPB 1065 in National Barley Disease Screening Nursery, 2016-17. Combination of good malt quality characters with high level of resistance

is an additional advantage for the breeding programme for developing good malt varieties.

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6. EP 87 (IC343586) (IC0343586; INGR20084), a Drought Tolerant Sorghum (*Sorghum bicolor*) Germplasm

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Sorghum [*Sorghum bicolor* (L.) Moench] is one of the important dry-land crops of semiarid tropics It is mainly grown in the drought prone areas to meet the food and fodder security of the region especially Maharashtra, Karnataka, Andhra Pradesh and Telangana where occurrence of drought is very common. Grain and fodder of post-rainy harvest fetches more prices because of good quality grain and fodder (Seetharama *et al*., 2010). Drought is one of the most damaging abiotic stresses affecting agriculture (Boyer, 1982). If it occurs at flowering, or in the grain filling stages, it may lead to reduction in yield, or crop failure. Drought response in sorghum has been characterized at both pre- and post-flowering stages (Talwar *et al*., 2010).

Experiments were conducted by Samdur *et al.* (2020) for two consecutive years in same field using split plot design with three replications during post-rainy seasons 2015–2016 and 2016–2017at Centre on Rabi sorghum (ICAR-Indian Institute of Millets Research), Solapur, Maharashtra. The main plots consisted of three levels of moisture regimes (three environments), namely (1) drought stress environment at GS1 stage (vegetative phase 20–35 days after sowing) without irrigation after, (2) drought stress environment at GS2 stage (pre-anthesis 40–55 days after sowing) without irrigation after and (3) well-watered (non-stress) environment, where irrigation was given as per need of crop. The sub plots consisted of 42 and 25genotypes grown during post-rainy seasons 2015–2016 and 2016– 2017, respectively, including four check varieties (Phule Anuradha, Phule Suchitra, CSV26 and M35-1). These four varieties were usually used in the yield evaluation trials of All-India Coordinated Research Project on Sorghum as drought-tolerant and high yielding genotypes. The result shows two genotypes CSV 26 and EP 87 (IC 343586) were found to be moderately tolerant under GS1 and GS2 environments and showed moderate adaptability for stover yield. All these genotypes identified using WGMI for drought tolerance will be utilized in future breeding program. In an earlier experiment by Talwar *et al*., (2010) during three rabi seasons of 2007-08, 2008-09 and 2009-10, the sorghum germplasm EP 87 (IS 343586) has been identified as new sources having improved post-flowering drought tolerance based on the yield components and green leave area retention at the physiological maturity. The rabi sorghum germplasm EP 87 (IC 343586) has consistently proved as a drought tolerant germplasm in five years of experiment. It can be used as a parent in the breeding programme to develop drought tolerant variety.

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7. SPV 2389/IIMR-SC-1542 (IC0485033; INGR20085), a Sorghum (*Sorghum bicolor*) Germplasm with Low HCN Content, High Protein Yield. High Seed Yielding Single-Cut Forage Genotype (Dual-Purpose Type).

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India faces a net deficit of 61.1 per cent green fodder, 21.9 per cent dry crop residues and 64 per cent feeds (Sunil Kumar *et al*., 2012).Forage sorghum has significant role in fulfilling the demand of green and dry fodder in the country. Both quantity and quality of the fodder is equally important in forage sorghum breeding. The quality of forage sorghum is determined by *in vitro* dry matter digestibility, protein content, protein yield and less HCN (Aruna *et al*., 2015). Hydrogen cyandide (HCN) is toxic to animals and can cause mild to severe reactions. High protein yield is preferred as it helps in improving the body weight of the animals. Low seed yield especially in forage lines is a drawback for farmers who take up seed production. Therefore high seed yielding forage lines are preferred. Genotypes which produces more green fodder at 50% flowering (Per day productivity) is economic trait among farmers.

SPV 2389 was developed at ICAR-Indian Institute of Millets Research, Hyderabad by crossing NSSV 13 × CSV 15 and subsequent selection was made through pedigree method till F6 generation. NSSV 13 is a sweet sorghum variety which has high juiciness content in its stalks and has high TSS%. CSV 15 is a dual purpose sorghum variety which has high grain yield as well as stover yields. The entry was tested for two years 2015 and 2016 under AICRP trials in 8-10 locations.

Morpho-agronomic characteristics: SPV 2389 takes 68 to 70 days for 50% flowering and has a plant height of 220-240 cms and 10- 12 leaves/plant. The proposed genotype SPV 2389 has low HCN content (95 ppm) very less than the safe limit (200 ppm). SPV 2389 has high protein yield with mean protein yield of 11.11q/ha over the better check CSV 21F (10.20 q/ha). SPV 2389 has highest seed yielding ability with mean seed yield of 2026 kg/ha as against the better check CSV 21F (1376 kg/ha). SPV 2389 has high per day productivity for green fodder yield with mean per day productivity of 6.8 q/ha/day over the better check CSV 21 F (6.4q/ha/day).

Associated characters and cultivation practices: The high brix/total soluble sugars recorded in SPV 2389 is also an important quality attribute which imparts to the palatability of the fodder and the line can serve as base material in forage breeding programs. The line also showed comparative resistance to foliar diseases (leaf blight, anthracnose and Zonate leaf spot and downey mildew) tested under AICSIP 2015. The proposed line can be important base material source while improving sorghum for fodder yield and quality for the Northern and southern forage growing states of the country.

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8. SPV 2315 (IC0635700; INGR20086), a Sorghum (*Sorghum bicolor*) Germplasm Resistance to Foliar Diseases (Anthracnose, Zonate leaf spot, Leaf Blight and Grey Leaf Spot). High Per Day Productivity for Green Fodder. High Seed Yield.

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In India, forage sorghum contributes to about 60-70% of the total green forage supply especially during Kharif and this can be enhanced by breeding cultivars with improved fodder yield. Foliar diseases (Anthracnose, leaf blight, Zonate leaf spot, grey leaf spot) are important biotic constraints to grain and forage sorghum production. Foliar diseases affect both the yield and quality of the fodder fed to animals. Foliar diseases can best be managed by host plant resistance.

The proposed single-cut forage sorghum genotype SPV 2315 was developed from double cross involving (CSV 20 × Pant Chari 5) × (CSV 20 × PVK 809)-4-2-2-2-1-2. CSV 20 and PVK 809 are dual purpose sorghum varieties while Pant Chari 5 is a forage sorghum variety) and subsequent selection through pedigree method till F_8 generation (Station code: DSR SC-40- 2). SPV 2315 was tested under All India Coordinated Research Trial for two years viz., *kharif* 2015 and *kharif* 2016.

Morpho-agronomic characteristics: The proposed genotype recorded multiple resistance to foliar diseases (Anthracnose, Zonate leaf spot, Leaf blight and Grey leaf spot). The entry showed 17.3, 15.59, 29.5, 57.3 per cent mean improvement over better check for Anthracnose, Zonate leaf spot, Leaf Blight and Grey leaf spot respectively. SPV 2315 also recorded high per day productivity for green fodder yield (6.38 q/ha/day) over better check CSV 21F (5.88 q/ha/day) and also recorded high seed yield 1690 kg/ha over the better check CSV 21F (1620 kg/ha).

Associated characters and cultivation practices SPV 2315 recorded 75 to 80 days for days to 50% flowering with a plant height of 230-250 cms having around 11-14 leaves/plant with leaf length of about 75 cm and leaf width of 7 cm. SPV 2315 showed average fodder yield of 540 q/ha (green) and 162 q/ha (dry).

Mean performance of SPV 2315 for foliar diseases, per day productivity for green fodder yield and seed yield.

Year of testing	No of test locations	SPV 2315	Latest varietal check CSV30F	CD@5%	%increase Overcheck
Anthracnose					
2014	4	4.8	6.3	4.9	23.8
2015	3	4.3	4.7	1.1	8.5
Mean		4.55	5.5		17.3
Zonate leaf spot					
2014	3	4.2	5.9	4.7	28.8
2015	1	5	5	0.7	0.0
Mean		4.6	5.45		15.6
Leaf Blight					
2014	3	2.8	4.1	2.3	31.7
2015	1	2.7	3.7	1.1	27.0
Mean		2.75	3.9		29.5
Grey leaf spot					
2014	1	1	5.7	1.7	82.5
2015	1	2.5	2.5	0.7	0.0
Mean		1.75	4.1		57.3
Per day productivity for green fodder yield					
2014	11	6.79	5.99	0.64	13.4
2015	10	5.97	5.69	0.6	4.9
Mean		6.38	5.84		9.2
Seed yield					
2015	11	1690	1060	980	59.4
Mean		1690	1060	980	59.4

*For disease resistance, scores recorded 1-9, values closer to 1 indicate resistance and closer to 9 indicate susceptible.

9. SPV-2296 (DSR 1145) (IC0635025; INGR20087), a Sorghum (*Sorghum bicolor*) Germplasm Tolerant to Shoot Fly and Downy Mildew. High Protein Content (12.2%) and High Grain Yield with Higher Nutrient-Use Efficiency.

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Breeding programmes aimed at development of high yielding genotypes endowed with resistance/tolerance to shoot fly and downy mildew have made significant progress. SPV 2296 is a *kharif* grain sorghum genotype with high grain yield and moderate tolerance to shoot fly and downy mildew, having higher protein content in the grains. It is a pedigree selection from the cross (RS 29 × NR 486) × NR 486. It has semi-loose symmetric panicles and medium bold elliptic seeds. In yield evaluation trials under AICRP-Sorghum, SPV 2296 recorded grain yield (2980 kg/ha) superiority of about 10% over the best performing grain yield check CSV 20 (2710 kg/ha) in Zone I (Tamil Nadu, Telangana, Andhra Pradesh, North Gujarat and Rajasthan) of sorghum growing area. At all India level SPV 2296 (3449 kg/ha) was marginally superior to the best check, CSV 20 (3397 kg/ha) in terms grain yield. It took 70-75 days to flower and 105-110 days to mature, and had an average plant height of 200-230 cm. It had medium bold seeds with average seed size of 2.45g/100 seeds. SPV 2296 has higher nutrient-use efficiency and can yield higher under good management. At 50% of RDF, SPV 2296 was 3% superior for grain yield over check CSV 20, while at 125% RDF it was superior by 2.3%. The loss in grain yield due to reduction in fertilizer dose by 50% was less (–7.6%) in SPV 2296 compared to CSV 20 (–13.1%), while the gain in grain yield due to higher dose of fertilizer was more in SPV 2296 (9.3%) compared to CSV 20 (3.7%).

For shootfly SPV 2296 recorded 4% less incidence of dead hearts under artificial infestation compared to CSV 20 (50.2%) and 28% lesser compared to susceptible check (DJ 6514). For stem borer it exhibited resistance on a par to the checks. SPV 2296 exhibited higher resistance to downy mildew (6.4%) compared to check (13.3%) and susceptible check (25.8%). It was on a par with the checks for resistance to other major diseases like grain mould (4.9 and 5.2 field grade and threshed grade, respectively, compared to 4.6 and 5.1 in check), anthracnose, leaf blight and zonate leaf spot at all India level.

The grains of SPV 2296 contain high protein content (12.19%) compared to check variety CSV 20 (10.97%) while for starch and fat contents, it (64.6% and 3.16%, respectively) was on a par with check (64.6% and 3.12%, respectively). For stover quality parameters the variety was on a par with check.

The breeding line SPV 2296 with high grain yield and medium bold seeds has exhibited good combining ability in subsequent crossing programmes. The high yield coupled with moderate resistance to major pest like shootfly and disease like downy mildew makes SPV 2296 an ideal base material for further sorghum improvement and consolidation of gains already achieved through recombination breeding.

10. EC718515 (EC718515; INGR20088), a Wild Lentil (*Lens orientalis*) Germplasm Resistant to Rust (*Uromyces fabae*) and Powdery Mildew (*Erysiphe trifolii*).

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The wild lentil accession EC718515 (ILWL230) belongs to *Lens orientalis* was identified after preliminary characterization and evaluation of 405 global wild annual lentil collections. The genotype was screened against rust (*Uromyces fabae* (Grev.) Fuckel) and powdery mildew (*Erysiphe trifolii*) under hot spot natural field condition at CSKHPKV Research and Extension centre Dhaulakuan Himachal Pradesh during 2012-2013 (Singh *et al.,* 2014). The genotype was further validated for confirming stable resistance against rust and powdery mildew during 2014-2015, 2015-2016 and 2016-2017(Singh *et al.,* 2020). Based on the disease score, an accession EC718515 (ILWL203) has been reported resistant against the rust and powdery mildew diseases.

Morpho-agronomic characteristics: Besides possessing resistance against rust and powdery mildew, an accession EC718515 (ILWL230) was also reported promising for other agro-morphological traits viz; number of pods plant^{-1} (141) and number of seeds plant^{-1}(160). As far as distinct morphological traits are concerned, the following qualitative features were also reported using lentil descriptor states jointly developed by IBPGR/ ICARDA (Table 1).

The above mentioned important characters have their special significant value for enhancing genetic gains of cultivated varieties, which need to be considered while planning future lentil genetic improvement programme for introgressing resistance against rust and powdery mildew as well as agronomic improvement of cultivated gene pool.

Table 1. Descriptor and descriptor state of EC718515 (ILWL230) for important qualitative characters

Descriptor	Descriptor state
Seedling stem pigmentation	Absent
Leaf pubescence	Slight
Tendril length	Rudimentary
Leaflet size	Large
Cotyledon colour	None
No. of pods/ plant	141

References

IBPGR/ICARDA (1993) Lentil descriptors: IBPGR Secretariat, Rome, Italy pp 15.

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11. EC718266 (EC718266; INGR20089), a Wild Lentil (*Lens nigricans*) Germplasm Resistant against Rust (*Uromyces fabae* (Grev.) Fuckel)

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The wild lentil accession EC718266 (IG136639) belongs to *Lens nigricans* was identified after preliminary characterization and evaluation of 405 global wild annual lentil collections. The genotype was screened against rust (*Uromyces fabae* (Grev.) Fuckel) disease under hot spot natural field condition at CSKHPKV Research and Extension centre Dhaulakuan Himachal Pradesh during 2012-2013 (Singh *et al.,* 2014). The genotype was further validated for confirming stable resistance during 2014-2015, 2015-2016 and 2016-2017(Singh *et al.,* 2020). Based on the disease score, an accession EC718266 (IG136639) has been reported resistant against the rust.

Morpho-agronomic characteristics: Besides possessing resistance against rust disease, an accession EC718266 (IG136639) was also reported promising for other agro-morphological traits viz; number of pods plant^{-1} (27) and number of seeds plant^{-1}(51). As far as distinct morphological traits are concerned, the following qualitative features were also reported using lentil descriptor states jointly developed by IBPGR/ICARDA (Table 1).

The above mentioned important characters have their special significant value for enhancing genetic gains of cultivated varieties, which need to be considered while planning future lentil genetic improvement programme for introgressing resistance against rust disease.

Table 1. Descriptor and descriptor state of EC718266 (IG136639) for important qualitative characters

Descriptor	Descriptor state
Seedling stem pigmentation	Absent
Leaf pubescence	Slight
Tendril length	Rudimentary
Leaflet size	Large
Cotyledon colour	None
No. of pods/ plant	27

References

IBPGR/ICARDA (1993) Lentil descriptors: IBPGR Secretariat, Rome, Italy pp 15.

Singh M, IS Bisht, S Kumar, M Dutta, KC Bansal, A Sarker, A Amri, SK Agrawal and SK Datta (2014) Global wild annual *Lens* collection: a potential resource for lentil genetic base broadening and yield enhancement. *PLoS ONE*, **9(9)**:e107781. doi:10.1371/journal.pone.0107781.

Singh M, **S** Kumar, AK Basandrai, D Basandrai, DR Saxena, A Sarker and K Singh (2020). Evaluation and identification of wild lentil accessions for enhancing genetic gain of cultivated varieties. *PLOS ONE* **15(3)**: e0229554.

12. EC271515 (EC271515; INGR20090), a French Bean (*Phaseolus vulgaris*) Germplasm Resistant toWhite Mold Disease (*Sclerotinia sclerotiorum*)

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The common bean (*Phaseolus vulgaris* L.) germplasm accession EC271515 was introduced from CIAT Columbia and screened against white mold (*Sclerotinia sclerotiorum*) under controlled environment at CSKHPKV Palampur. The germplasm was screened using various inoculation procedures and modified straw test method was found most promising. The genetic materials comprising of a panel of 516 common bean accessions were screened in controlled conditions which resulted into the identification of an accession EC271515 found resistant against white mold disease and subsequently validated thrice to confirm the stable resistance against the pathogen (Chauhan *et al.,* 2020).

Table 1. Descriptor and descriptor state of EC271515 for important qualitative characters

Descriptor	Descriptor state
Plant growth habit	Semi pole
Leaflet shape	Ovate lanceolate
Flower colour	Light pink
Pod shape	Round
Seed colour	Maroon
Pod pubescence	Absent
Pod colour	Green
No. of pods plant^{-1}	08
No. of seeds pod^{-1}	06

The above mentioned important agro-morphological characters have their significant value for enhancing genetic gains, which need to be considered while planning future common bean genetic improvement programme for introgressing resistance against white mold.

Morpho-agronomic characteristics: Besides possessing resistance against white mold, an accession EC271515 was also reported promising for other agro-morphological traits viz; number of pods plant^{-1} (08) and number of seeds pod^{-1} (6). As far as distinct morphological traits are concerned, the following qualitative characters were also reported using common bean descriptor states jointly developed by IBPGR/NBPGR (Table 1).

References

Chauhan S, S Katoch, SK Sharma, PN Sharma, JC Rana, K Singh and M Singh (2020) Screening and identification of resistant sources against *Sclerotinia sclerotiorum* causing white mold disease. *Crop Sci.* **60**: 1986–1996.

IBPGR/NBPGR (1993) Common bean descriptors: IBPGR Secretariat, Rome, Italy pp 15.

13. IC278744 (IC0278744; INGR20091), a French Bean (*Phaseolus vulgaris*) Germplasm Resistant toWhite Mold Disease (*Sclerotinia sclerotiorum*)

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The common bean (*Phaseolus vulgaris* L.) germplasm accession IC278744 was collected from Chhogtali Sirmour Himachal Pradesh and screened against white mold (*Sclerotinia sclerotiorum*) under controlled

environment at CSKHPKV Palampur. The germplasm was screened using various inoculation methods and modified straw test method was found most appropriate. The genetic materials comprising of a panel of 516 common bean accessions were screened in controlled conditions which resulted into the identification of an accession IC278744 found resistant against white mold disease and subsequently validated thrice to confirm the stable resistance against the pathogen (Chauhan *et al.,* 2020).

Morpho-agronomic characteristics: Besides possessing resistance against white mold, an accession IC278744 was also reported promising for other agro-morphological traits viz; number of pods plant^{-1} (10) and number of seeds pod^{-1} (6). As far as distinct morphological traits are concerned, the following qualitative characters were also reported using common bean descriptor states jointly developed by IBPGR/NBPGR (Table 1).

The above mentioned important agro-morphological characters have their significant value for enhancing genetic gains, which need to be considered while planning future common bean genetic improvement programme for introgressing resistance against white mold.

Table 1. Descriptor and descriptor state of IC278744 for important qualitative characters

Descriptor	Descriptor state
Plant growth habit	Pole type
Leaflet shape	Ovate
Flower colour	White
Pod shape	Slightly curved
Seed colour	Maroon
Pod pubescence	Intermediate
Pod colour	Green

References

Chauhan S, S Katoch, SK Sharma, PN Sharma, JC Rana, K Singh and M Singh (2020). Screening and identification of resistant sources against *Sclerotinia sclerotiorum* causing white mold disease. *Crop Sci.* **60**: 1986–1996.

IBPGR/NBPGR (1993) Common bean descriptors: IBPGR Secretariat, Rome, Italy pp 15.

14. NIPB-1 & NIPB 1B (IC0637026 & IC0637027; INGR20092), a Cytoplasmic Male Sterile Line of Cauliflower (*Brassica oleracea* var. *botrytis*) with Compact Creamy White Curd. Strongly Waxy with Bluish Green Broad Leaves.

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Higher yield, greater uniformity and quality of F1 hybrids call for heterosis breeding in cauliflower. Attempts therefore have long been made to generate CMS lines in *B. oleracea* by means of different cytoplasmic sources for hybrid production (Shiga and Baba, 1973; Renard *et al*., 1992; Zhu and Wei, 2009 and Kamiński, Dyki and Stępowska, 2012). However, not all have been successful mainly due to poor stability of CMS and/or adverse effect on agronomic traits. At NIPB, *Erucastrum canariense* credited male sterility was successfully transferred from *B. napus* background to cauliflower to yield a CMS cauliflower line called 'NIPB-1'. In 2010, cauliflower variety Pusa Meghna (CC – 2n=2x=18) as male parent was crossed with CMS- (*E. caneriense*) *B. napus (*AACC – 2n=4x=38) to obtain F1 plants. In subsequent backcrosses, cauliflower (Pusa Meghna) was used as recurrent male parent to recover the cauliflower genotype with male sterile flowers. Crosses at initial stages could not be obtained at field level due to embryo degeneration occurring within 12-14 days of hand pollination. Therefore, embryo rescue technique/ ovary culture has to be resorted every time after some days of pollination to recover the succeeding backcross generations. Pistils harvested at 14 days after pollination (14 DAP) for embryo rescue gave the best results (Table 1). At every cycle of crossing some crosses were always left in the field till maturity to see the possibility of any natural seed setting. Eventually, at BC4 stage some seeds were formed on hybrids plants. Plants obtained from such seeds were very healthy that formed perfect cauliflower curd. Subsequent advancement of generations never required embryo rescue.

Morpho-agronomic characteristics: NIPB-1 has strongly waxy bluish green leaves, light yellow sterile flowers, anthers are short with absence of pollen grains, nectaries are good making it attracted to bees, an

Table 1. Summary of results of embryo rescue towards the recovery of interspecific hybrids between *B. napus* × cauliflower

Cross	Time of sampling of ovaries	No. of ovaries cultured	No. of surviving ovaries	No. of ovules obtained and cultured	No. of plants recovered
	5 DAP	60	0	0	0
CMS (*E. canariense*) *B. napus* × *B. oleracea*	9 DAP	60	28	23	2
	14 DAP	60	32	47	5

advantage for hybrid seed production. It belongs to mid early maturity group which bears compact big circular curd not covered with young leaves and mostly shows anthocyanin pigmentation just before its opening to flower. Though, Pusa Meghna was the recurrent parent in synthesis of NIPB-1, it is different for some of the morphological features such as curd size, anthocyanin pigmentation and leaf size and colour which may be attributed to *caneriense* cytoplasm or effect of residual *Brassica napus* linked genes.

Associated characters and cultivation practices: NIPB-1 is a cauliflower CMS line which can be used as such as a valuable CMS female parent for production of hybrid variety or can be used as a source of male sterility for integration in other female combiners. Some incidences of cabbage butterfly larvae were seen during its early growth which was secured by insecticidal spray. No major incidences of other diseases and insects were noticed during the course of development of this material. It grows well under standard cultural practices normally followed for cauliflower farming.

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15. DRMR 2300 (NPJ-149) (IC0609646; INGR20093), an Indian Mustard (*Brassica juncea*) Germplasm with High Temperature Tolerance at Seedling Stage.

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DRMR 2300 (IC609646; NPJ149) is an advanced breeding line of Indian mustard (*B. juncea*) procured from IARI, New Delhi. DRMR 2300 was tested for physio-biochemical parameters (RWC, Chlorophyll stability index (CSI) during 2016-17 at DRMR, Bharatpur. The genotype DRMR 2300 was superior over checks (BPR-543-2, Urvashi) in terms of percent increase of RWC (Bud stage, 0.49%) and CSI (pod stage, 3.08%) under stress condition. DRMR 2300 showed seedling mortality ≤20% and DW/10 seedlings ≥ 4g under field conditions at DRMR, Bharatpur during 2017-18.

During 2018-19 DRMR 2300 was evaluated for high temperature tolerance at seedling stage under field conditions at 3 locations (Dholi, Kanpur and Ludhiana) along with checks. Average seedling mortality (%) of DRMR 2300 over three locations was 18.2% and DW/10 seedlings over three locations was 7.0 g (Table 1). Genotype DRMR 2300 showed average SPAD value of 41.4 and RWC 83.3% over 3 locations (Dholi, Kanpur and Ludhiana) which is higher than checks at seedling stage.

During 2018-19 proposed germplasm DRMR 2300 was evaluated for high temperature tolerance at seedling stage under field conditions at 3 locations (Dholi, Kanpur, Ludhiana) along with checks under plant physiological traits (AICRP-RM, 2019, page PHY 1). Seedling mortality ≤20% and DW/10 seedlings ≥ 40mg rated genotypes tolerant under field conditions.

Table 1. Pooled data of DRMR 2300 over three locations (Dholi, Kanpur, Ludhiana) for high temperature tolerance at seedling stage under AICRP-RM 2018-19 under field trials

Genotypes	Seedling mortality (%)	DW/10 seedlings (g)	SPAD	RWC (%)
DRMR 2300	18.2	7.0	41.4	83.3
RGN 73 (ZC)	34.2	6.3	36.4	72.5
PM 25 (NC)	19.8	5.5	40.1	72.2
Kranti (NC)	27.4	5.1	36.8	72.0
RH 749 (ZC)	23.2	5.7	37.5	66.9

Average seedling mortality (%) over three locations was 18.2% and DW/10 seedlings over three locations was 7.0g (Table 3). Genotype DRMR 2300 showed average SPAD value of 41.4 and RWC 83.3%over3 locations (Dholi, Kanpur and Ludhiana) which is higher than checks (AICRP-RM, 2019, page PHY 5, Table 4) at seedling stage.

References

DRMR Annual Report, 2016-17, page 29

DRMR Annual Report, 2018-19, page 46.

Annual Progress Report, AICRP-RM, 2019, page PHY 1,4-5

Table 3. Screening of genotypes from different agro-climatic zones for high temperature at seedling stage under field conditions

Genotypes	Seedling mortality (%) (<20%)				DW/10 seedlings (g) (≥ 4g)			
	Dholi	Kanpur	Ludhiana	Average	Dholi	Kanpur	Ludhiana	Average
DRMR 2300	19.9	16.5	18.2	18.2	9.2	4.5	7.2	7.0
RGN 73 (ZC)	34.9	42.8	25.0	34.2	8.9	3.5	6.6	6.3
Pusa Mustard 25 (NC)	27.6	15.8	16.0	19.8	5.8	4.7	6.0	5.5
JD 6 (ZC)	34.8	50.2	14.4	33.1	3.8	2.8	4.9	3.8
Kranti (NC)	32.5	28.8	20.8	27.4	4.6	3.4	7.3	5.1
RH 749 (ZC)	40.4	14.6	14.5	23.2	4.2	4.6	8.3	5.7
Mean	31.3	35.2	18.1		5.5	3.5	6.6	
CD (p=0.05)	3.12	4.7	4.2		1.04	0.50	1.33	

(AICRP-RM, 2019 Page: PHY 4)

Table 4. Effect of high temperature at seedling stage on SPAD and RWC under field conditions

Genotypes	SPAD				RWC(%)			
	Dholi	Kanpur	Ludhiana	Average	Dholi	Kanpur	Ludhiana	Average
DRMR 2300	46.5	41.5	36.3	41.4	89.9	77.5	82.6	83.3
Pusa Mustard 25 (NC)	42.3	41.9	36.2	40.1	78.8	76.5	61.3	72.2
Pusa Mustard 30 (LR)	35.3	33.3	34.9	34.5	76.3	73.6	49.2	66.4
JD 6 (ZC)	37.3	36.6	34.4	36.1	78.5	67.7	52.3	66.2
Kranti (NC)	35.1	39.2	36.2	36.8	76.7	72.4	66.9	72.0
RH 749 (ZC)	34.8	42.0	35.6	37.5	67.3	76.7	56.8	66.9
Mean	38.6	37.6	35.6		77.9	73.1	68.7	
CD (p=0.05)	1.17	2.8	0.73		1.61	1.8	9.5	

(AICRP-RM, 2019 Page: PHY 5)

Morphological Characters of DRMR2300

	Value
Plant Height (cm)	195.0
Days to maturity	138.0
Primary branches/plant (no.)	8.0
Secondary branches/plant (no.)	14.0
Main shoot length (cm)	75.0
Siliquae/plant (no.)	307.0
Siliqua length (cm)	4.3
Seeds/siliqua (no.)	16.0
1000 seed weight (g)	6.2
Oil content (%)	41.90
Seed yield/plant (g)	20.1

16. ICIRG226-29-2-2 (IC0636678; INGR20094), a Castor (*Ricinus communis*) Germplasm with High Ricinoelic Acid. Early Maturity.

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Castor (*Ricinus communis* L.) oil demand is increasing globally because of presence of 85-88% ricinoelic acid (RA) in oil which is the only natural source of RA. Any increase in RA content beyond the existing level (85-88%) would increase the value of oil and benefit the industry and farmers. So far no high ricinoleic type (>88% ricinoleic acid) castor cultivars are available in the world as there were no donors for high RA content for developing the cultivars. Therefore, the ICAR-Indian Institute of Oilseeds Research (ICAR-IIOR), Hyderabad, has developed the first high ricinoleic type inbred line *viz.,* ICIRG226-29-2-2 from germplasm. It was evaluated in randomized block design with two replications at multilocations for two years.

Morpho-agronomic characteristics: Ricinoleic acid content in ICIRG226-29-2-2 was 91.5% (Table 1) and oil content was 45%. It matured in 106 days and gave 17% higher seed yield (2438 kg/ha) than the early check, DCS-9 (2075 kg/ha) in multilocation trial (Table 2). It has red stem with bloom on it and medium size spiny capsules and seeds.

Associated character and utility: Low node number (11) and short plant height (77 cm) are the other traits of ICIRG226-29-2-2. This inbred line would serve as a unique souse of high RA content coupled with early maturity and high yield in castor breeding for developing early maturing high ricinoleic type-high yielding hybrids and varieties. ICIRG226-29-2-2 would also serve as base material for studying genetics and inheritance of high RA content trait, and for developing/identifying genes/ markers for high RA content for intensifying molecular breeding research in castor.

Table 1. Ricinoleic acid content in ICIRG226-29-2-2

Inbred line	Ricinoleic acidc ontent (%)							Mean over years
	Irrigated		Rainfed			Overall mean		
	S.K.Nagar		Palem	ICAR-IIOR				
	2018	2019	2019	2018	2019	2018	2019	
ICIRG226-29-2-2	90	91	92	92	92	91	92	91.5
DCS-9 (early check)	88	89	89	88	89	88	89	88.5
GC3 (check)	88	86	88	-	88	88	87	87.5
CV (%)	1.3	0.9	0.2	0.5	0.8			
CD (P=0.05)	1.002	NS	0.8	1.3	NS			

Table 2. Mean phenological traits and seed yield of ICIRG226-29-2-2

Inbred line	Days to 50% flowering		Mean	Days to maturity		Mean	Seed yield (kg/ha)		Mean
	2018	2019		2018	2019		2018	2019	
ICIRG226-29-2-2	44	42	43	114	98	106	3196	1679	2438
DCS-9 (early check)	45	45	45	114	104	109	2603	1547	2075
GC3 (check)	45	51	48	114	109	112	4501	2195	3348

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17. CSIR-IHBT-ST-03 (IC0635703; INGR20095), a Tetraploid Stevia (*Stevia rebaudiana*) Germplasm with Large Leaf Size

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Stevia rebaudiana (Bertoni) is a herbaceous perennial plant of the Asteraceae family and native to Paraguay (South America). The chromosome count of stevia is 2 n=22 (diploid) and genome size (2C value) is estimated to be 2.72 pg or 2660 Mbp (Yadav *et al.*, 2014) The genus *Stevia cav.* consists of approximately 230 species of herbaceous, shrub and sub-shrub plants. It has been recognized worldwide for its excellent sweetening property. Leaves of stevia produce diterpene glycosides (stevioside and rebaudiosides), which are non-toxic, high-potency sweeteners (300 times sweeter than sucrose) and substitutes for sugar (Megeji *et al.*, 2005, Rajasekaran *et al*., 2007) in different drinks, beverages and bakery products (Abou-Arab *et al.*, 2010). Manipulation of ploidy is a valuable tool which has been recognized in plant breeding programmes to improve agronomic traits, particularly biomass yield. For polyploidization in plants, colchicine is being widely used since its first report having this property (Blakeslee and Avery 1937). Colchicine treatment disrupts the polymerization of microtubules, and hence, interrupts spindle-fiber development during cell division (Bartels and Hilton 1973).

Leaves being the economic part in stevia, where steviolglycoside synthesis takes place, improvement in leaf characteristics will have direct influence on leaf biomass yield. With an aim to increase leaf size resulting into high leaf biomass yield, tetraploid stevia genotype CSIR-IHBT-ST-03 (C-7-3-4) has been developed through colchicine treatment of stevia seeds (Fig. 1). Polyploidy status (tetraploid) was analyzed and confirmed through flow cytometry and cytology of root tips (Yadav *et al*., 2013). The genotype CSIR-IHBT-ST-03 was evaluated for morphological as well as biochemical traits along with control. Tetraploid stevia (CSIR-IHBT-ST-03) is having large leaf size (max. leaf length and width: 11.34 cm × 7.80 cm respectively) as compared to diploid control (5.75 cm × 3.45). The autotetraploids in stevia had significantly increased leaf size (Yadav *et al*., 2013). The genotype CSIR-IHBT-ST-03 was developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The genetic stock CSIR-IHBT-ST-03 has increased leaf size, internode length, stem thickness, stevioside and rebaudioside-content and has potential to be utilized as low calorie natural sweetener which is a substitute for sugar or zero calorie synthetic sweeteners.

Associated characters and cultivation practices: *Stevia rebaudiana* (Bertoni) is a herbaceous perennial plant of the Asteraceae family, native to Paraguay (South America). Propagation of stevia is through seeds as well as clonal (Yadav *et al.*, 2010). Leaves of stevia is the economic part which produces diterpene glycosides (stevioside and rebaudiosides), which are high-potency sweeteners (300 times sweeter than sucrose) and substitutes for sugar or synthetic sweeteners.

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18. CSIR-IHBT-VJ-05 (IC0630604;INGR20096), a Tagar IndianValerian (*Valeriana jatamansi*) Germplasm with High Fresh Root Biomass Yield of 2.71 kg/plot (6 sqm). Essential oil content: 0.31%.

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Valeriana jatamansi Jones is popularly known as Indian valerian, Mushkbala (Kashmiri/ Hindi), Sugandhwala or Tagar (Sanskrit) and is a native plant of Himalayan origin. *Valeriana jatamansi*, a perennial medicinal herb, is now endangered and at the edge of becoming extinct in India (Mahajan and Pal, 2016). It is therefore a pressing need to conserve and maintain this species in their natural habitat. The species is extensively utilized for its roots and rhizomes which contain essential oil. In order to improve productivity and essential oil content of the roots, a breeding programme has been undertaken to identify promising selection for commercial cultivation.

Nine accessions of Indian valerian were initially shortlisted based on root biomass accumulation and essential oil content over two years. The progeny plants of these nine accessions were evaluated in multi-location trials for root biomass and essential oil content at four locations in mid- and high-altitude regions over a period of two years along with check variety 'Himbala'. Overall, CSIR-IHBT-VJ-05 performed better than check at all the four locations with root biomass yield of 2.71 kg/plot (6 sqm) and essential oil content of 0.31%. The genotype CSIR-IHBT-VJ-05 was developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The characteristic features of CSIR-IHBT-VJ-05 are plant height of about 45 cm, large leaf size with serrated margins and pointed apex. The selection CSIR-IHBT-VJ-05 is vigorous in growth and has a potential to be utilized as aromatic plant on commercial basis.

Associated characters and cultivation practices: *Valeriana jatamansi*, a perennial medicinal herb of the Valerianaceae family, is widely found in the temperate zone of the western Himalaya at an altitude of 1300–3300 m amsl. In India, the species is now endangered and at the edge of becoming extinct (Nayar and Sastry, 1998) due to over-exploitation from its natural habitat to meet the burgeoning demand.

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19. IIHR-CA-13 (IC0618244; INGR20097), a Bramhi/Indian Birthwort (*Centella asiatica*) Germplasm with Higher Asiaticoside Content (3.73%). Higher Total Triterpene Content (7.67%). Higher Dry Biomass Content (2276 kg/ha).

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Indian Pennywort (*Centella asiatica*) belonging to the family Apiaceae is one of the important medicinal herb that is used as a memory booster. Triterpenoids like asiaticoside, madecassoside, asiatic acid and madecassic acid contribute to the medicinal properties. IIHR-CA-13 genetic stock is a clonal selection from the germplasm collected from Shimoga, Karnataka. The genotype has high asiaticoside content (3.73%) and higher tri- terpenoid content (7.67 %). This line is particularly good for pharmaceutical industries for extraction of triterpenoids. It was developed at ICAR-Indian Institute of Horticultural Research (ICAR-IIHR), Bengaluru.

Morpho-agronomic characteristics: IIHR-CA-13 is characterized by erect growth habit and medium sized leaves. Important traits are mentioned as below based on 3 years pooled data.

Associated characters and cultivation practices: IIHR-CA-13 is moderately tolerant to leaf spot caused by *Cercospora centellae*. The crop requires damp, moist and shady habitats for its growth. Clayey soils with good moisture along with organic matter suits very well for the crop. The plant is propagated through stem cuttings comprising of rooted node with few leaves @ 1,10,000 cuttings/ha. The cuttings are planted at a spacing of 30 × 30 cm in the main field preferably during June-July. Organic manuring should be done with 20t/ha FYM per year.

Traits	IIHR CA-13	Vallabh medha (check)	CD (5%)	CV
Asiaticoside (%)	3.73	1.94	0.42	13.27
Total Triterpenes (%)	7.67	5.00	0.27	7.19
Dry biomass yield (kg/ha/year)	2276	1421.67	4.00	6.16
Plant Height (cm)	8.52	6.71	1.72	11.95
No. of primary branch per plant	5.20	7.27	2.08	7.94
No. of Nodes per plant	8.00	8.40	2.27	9.08
No of Leaves per plant	29.00	33	7.88	6.98
Leaf Length (cm)	2.59	2.45	0.25	5.52
Leaf Width (cm)	4.03	3.73	0.72	9.79
Internodal length (cm)	6.89	7	0.48	4.06
Rosette diameter (cm)	13.04	12.13	1.01	5.06
Petiole length (cm)	9.63	7.37	1.29	9.47
Swollen petiole base (mm)	4.00	4.50	1.17	14.81
petiole thickness (mm)	1.83	1.93	0.61	17.44
Length between leaf base (cm)	1.53	0.97	0.36	15.33
Stem thickness (mm)	1.25	1.25	0.11	4.41

References

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20. IIHR-CA-1 (IC0618233; INGR20098), a Bramhi/Indian Birthwort (*Centella asiatica*) Germplasm with Higher Fresh Biomass Yield of 15t/ha/year. Higher Total Carotenoid (32.33 mg/100g) and Iron (149.5 ppm) Content, Broad Sized Leaves with Long Petiole.

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Indian Pennywort (*Centella asiatica*) belonging to the family Apiaceae is one of the important medicinal herb that is used as a memory booster. Triterpenoids like asiaticoside, madecassoside, asiatic acid and madecassic acid contribute to the medicinal properties. It is also used as a green leafy vegetable because of high vitamin and mineral content. IIHR-CA-1 genetic stock is a clonal selection from the germplasm collected from Pune, Maharashtra. The genotype has very broad leaves with long petiole (12.17 cm) and high fresh biomass (15038 kg/ha/year) and dry biomass yield (2506.33 kg/ha/year). This line is good for both medicinal and vegetable purpose as it has broad leaves, more biomass yield and high carotenoid and iron content. It is developed at ICAR-Indian Institute of Horticultural Research (ICAR-IIHR), Bengaluru.

Morpho-agronomic characteristics: It is characterized by erect growth habit and very broad sized leaves, long

petiole, easy to harvest and can be harvested at every 30 days interval. Important traits are mentioned as below based on 3 years pooled data.

Associated characters and cultivation practices: IIHR-CA-1 is moderately tolerant to leaf spot caused by *Cercospora centellae*. The crop requires damp, moist and shady habitats for its growth. Clayey soils with good moisture along with organic matter suits very well for the crop. The plant is propagated through stem cuttings comprising of rooted node with few leaves @ 1,10,000 cuttings/ha. The cuttings are planted at a spacing of 30 × 30cm in the main field preferably during June-July. Organic manuring should be done with 20t/ha FYM per year.

Traits	IIHR CA-1	Vallabh Medha (check)	CD (5%)	CV
Fresh biomass yield (kg/ha/year)	15038	8530	9.80	6.61
Dry biomass yield (kg/ha/year)	2506.33	1421.67	4.00	6.16
Total carotenoids (mg/100g fresh weight)	32.33	26.58	-	-
Iron (ppm)	149.5	128	-	-
Asiaticoside (%)	1.207	1.94	0.42	13.27
Total Triterpenes (%)	4.19	5.00	0.27	7.19
Plant Height (cm)	10.92	6.71	1.72	11.95
No. of primary branch per plant	5.73	7.27	2.08	7.94
No. of Nodes per plant	6.07	8.40	2.27	9.08
No of Leaves per plant	25	33	7.88	6.98
Leaf Length (cm)	3.79	2.45	0.25	5.52
Leaf Width (cm)	5.54	3.73	0.72	9.79
Intermodal length (cm)	10.03	7	0.48	4.06
Rosette diameter (cm)	19.8	12.13	1.01	5.06
Petiole length (cm)	12.17	7.37	1.29	9.47
Swollen petiole base (mm)	9.00	4.50	1.17	14.81
petiole thickness (mm)	2.83	1.93	0.61	17.44
Length between leaf base (cm)	3.07	0.97	0.36	15.33
Stem thickness (mm)	2.45	1.25	0.11	4.41

References

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21. PR-9 (IC0636677; INGR20099), a Dwarf Tuberose Selection (*Polianthes tuberosa*) with Average Plant Height 48.49 cm. Short and Straight Spikes Suitable for Pot Culture, Vertical Panel and other Purposes

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Tuberose (*Polianthes tuberosa* L.) is one of the important fragrant bulbous ornamental cultivated for loose & cut flower, extraction of essential oil, etc. It belongs to the family Amaryllidaceae. It has gained popularity on account of ease in cultivation, fragrance, keeping quality, economic returns, wide adaptability to varied climate and soil (Safeena *et al*., 2015). The tuberose variety PR-9 was developed at ICAR-Directorate of Floricultural Research, Pune, Maharashtra (latitude of 18.5204° N, longitude of 73.8567° E and altitude of 560 MSL on the western margin of the Deccan plateau). It is a selection among the Open Pollinated Population of the cv. 'Phule Rajani'. The plants as well as spikes are very short unlike the parent Phule Rajani. It was evaluated for three years (2017-18 to 2019-20) along with parent as check.

Table 1. Performance of the Identified Variety 'PR-9' over Parent' Phule Rajani' During 2017-18 to 2019-20.

Character	2017-18		2018-19		2019-20		Average	
	PR-9	Phule Rajani	PR-9	Phule Rajani	PR-9	Phule Rajani	PR-9	Phule Rajani
Plant height (cm)	39.6	78.8	54.3	90.6	51.58	94	48.49	87.80
Spike length (cm)	32.2	67.6	44	80.4	46	82.5	40.73	76.83
Rachis length (cm)	22.2	37	31	43.6	24.52	42	25.91	40.87
Number of leaves	10.8	12	9.7	13.4	8.8	13	9.77	12.80
Days to flowering	98	90	NA	NA	NA	NA	98.00	90.00
Number of florets per spike	39.2	27.2	46.3	38.6	46	37.5	43.83	34.43
Floret diameter (cm)	3.6	3.8	4.2	5	4.78	4.8	4.19	4.53
Floret length (cm)	4	6	5	6.9	6.08	6.2	5.03	6.37
Stem thickness (cm)	1.3	1.2	1.5	1.1	1.3	1.3	1.37	1.20
Spike longevity (in field days)	7	6	8	8.5	8	7.5	7.67	7.33
Number of bulbs per clump	10	12	13	15	12	13	11.67	13.33
Vase life of spike in tap water (days)	6	6	7	7	7	7	6.67	6.67
No of spikes per clump	5	5.2	5.5	4.8	6	6.5	5.50	5.50
Plants pread (cm)	51	78.4	54	98.4	58.5	86	54.50	87.60

Morpho-agronomic characteristics: It is very dwarf (average plant height 48.49 cm) with short and straight spikes. The florets are with light pink tinge (63B, Red-Purple Group) at bud stage, turning greenish white (NN 155B, White Group) at opening/ anthesis. Petals show a distinguished spot/mark in the outer end. Stems are sturdy with thickness more than the parent variety (1.37 cm against 1.20 cm). Average rachis length is 25.91 cm with 43.83 numbers of florets per spike. Each bulb produces on an average5.50 spikes per clump per year (Table.1).

Associated characters and cultivation practices: The variety PR-9 produces dwarf plants which are well suited to pot-culture, for vertical gardening (vertical panels) and best suited for urban dwellers (for terrace/roof gardening). Flowering commences 98 days after planting and continues almost through the year. However, during winter months (Nov- Jan) growth gets slowed down. The florets are arranged compactly on short rachis giving an impression of hyacinth spike. Also, the floret opening is proper till the end of spike. Sweet fragrance of the florets is an additional attraction. Spikes can be used for vase decoration also (with a shelf-life of 6.67 days in vases). Bulbs can be lifted after 2-3 years depending on the growth & flowering, purpose and climate. On an average 11.67 bulbs are produced from each clump per year. It is propagated by bulbs and can be planted during March-June. The optimum growing temperature requirement is 25-30 °C. It can be grown on a wide range of soil. However, it prefers deep friable soil rich in organic content having good water holding capacity and pH around 6.5-7.5. NPK @ 300:250:300 kg/ha along with FYM (20t/ha) is recommended for good growth and flowering.

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22. CSIR-IHBT-Gr-11-6 (IC0630601; INGR20100), a Gerbera (*Gerbera jasmeonii*) Germplasm with Double Flower Shape

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Gerbera (*Gerbera jamesonii* Bolus ex Hook. f.), family Asteraceae, is one of the important cut-flowers used for cut-flower, aesthetic decoration, bouquet making and has high demand in domestic and export markets (Singh *et al*., 2017). Gerbera holds the fifth position among the top ten traded flowers in the world and have a market

of €140 million. In 2017, a total of 1074 million gerbera cut-flowers sold worldwide (Holland, 2018). In India, the production of gerbera was estimated to be 5020 tonnes of loose flowers and 20.53 lakh number of cut flowers from an area 1150 ha (NHB 2018). It is widely grown in Karnataka, Maharashtra, Chhattisgarh and North Eastern states. It is a dwarf perennial herbaceous plant, growing in clumps with solitary flower heads on long slender stems which grow well above the foliage. In general, gerbera is grown in protected condition to meet the quality standards of international and domestic market.

In India, exotic cultivars of gerbera are predominantly grown on commercial scale. Indigenous cultivars may play an important role to combat import of gerbera cultivars and save our valuable foreign exchange in terms of royalty which we pay to foreign breeder companies. With an objective to develop unique selections of gerbera, hybridization program was undertaken involving elite and diverse parental lines differing for flower color (Singh *et al*., 2013), shapes and size at CSIR-Institute of Himalayan Bioresource Technology (CSIR-IHBT), Palampur, Himachal Pradesh, India (Singh *et al*., 2009 & 2011). CSIR-IHBT has developed a gerbera selection (CSIR-IHBT-Gr-11-6) through breeding program. The hybrid F_1 genotypes of gerbera were developed through controlled crossing program to obtain mature seeds from different cross combinations. The seeds were initially cultured on MS basal medium for development of micro-shoots and re-cultured on MS media supplemented with different doses of growth regulators (BAP, IBA, NAA), to achieve shoot proliferation. Highest number of micro-shoots, number of leaves and leaf length were observed in MS medium supplemented with 1 mg/L BAP + 0.03mg/L IBA + 0.025 mg/L NAA. For *in vitro* rooting, half strength MS medium supplemented with 0.4 mg/L IBA was found best. Rooted plantlets were successfully hardened in trays filled with moist sand and transferred to sleeves for cultivation in soil.

The hybrid F_1 genotypes were morphologically characterized under field conditions with respect to floral traits and evaluated for agronomic performance over a period of four years. Based on mean performance of hybrid gerbera genotypes compared to respective parents, gerbera genotype CSIR-IHBT-Gr-11-6 was found promising having double flower shape of mini size (flower diameter of 9.65 cm) and is light yellow in color. The genotype CSIR-IHBT-Gr-11-6 has been developed at CSIR-IHBT, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: CSIR-IHBT-Gr-11-6 was found promising having double flower shape of mini size (flower diameter of 9.65 cm) and is light yellow in colour.

Associated characters and cultivation practices: Gerbera is commercially gerbera is grown in polyhouse or shade house. Day temperature of 22^{o}-25^{o}C and night temperature of 12-16^{o}C is ideal for growth and flower production. (Aswath *et al.*, 2015).

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23. CSIR-IHBT-Gr-Y-1 (IC0630600; INGR20101), a Gerbera (*Gerbera jasmeonii*) Germplasm with Double Flower Shape. Standard Size (>10 cm).

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Gerbera (*Gerbera jamesonii* Bolus ex. Hooker F.) belongs to family Asteraceae and is one of the important cut-flowers grown for domestic and export markets. It ranks fifth among the top ten cut flowers in the global market. Apart from its use in beds, borders, pots and rock gardens, it also has considerable export potential (Tija, 2001). It is a perennial plant which can be grown under wide range of climatic conditions. Based on its floral biology, gerbera is an out-cross in breeding behaviour and clonal propagation through tissue culture has resulted in development of large number of gerbera cultivars. New variations in gerbera can be developed through hybridization program involving diverse parents which will widen the range of floral variations and facilitate selection of desirable genotypes that can be clonally multiplied through vegetative propagation. With an objective to develop unique selections of gerbera, hybridization program was undertaken involving elite and diverse parental lines differing for flower color (Singh *et al*., 2013), shapes and size at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh, India (*Singh et al*., 2009 & 2011).

Hybrid F_1 genotypes of gerbera were developed through controlled crossing program and mature seeds obtained from different cross combinations were used for the establishment of *in vitro* gerbera cultures. Seeds were cultured on MS basal medium and the developing micro-shoots from seeds were further cultured on MS media supplemented with different doses of growth regulators to achieve shoot proliferation. Of the different media, highest number of micro-shoots, number of leaves and leaf length were observed in MS medium supplemented with 1 mg/L BAP + 0.03mg/L IBA + 0.025 mg/L NAA which gave best proliferation among the gerbera genotypes. Half strength MS medium supplemented with 0.4 mg/L IBA was found best for *in vitro* rooting. Rooted plantlets were successfully hardened in trays filled with moist sand and transferred to sleeves for cultivation in soil. The hybrid F_1 genotypes were morphologically characterized under field conditions with respect to floral traits and evaluated for agronomic performance over a period of four years. Based on mean performance of hybrid gerbera genotypes compared to respective parents, gerbera genotype CSIR-IHBT-Gr-Y-1 was found promising having double flower shape of standard size (flower diameter of 10.84 cm) and is yellow in color. The genotype CSIR-IHBT-Gr-Y-1 has been developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: CSIR-IHBT-Gr-Y-1 was found promising having double flower shape of standard size (flower diameter of 10.84 cm) and is yellow in color.

Associated characters and cultivation practices: Gerbera is commercially gerbera is grown in polyhouse or shade house. Day temperature of 22^{o}-25^{o}C and night temperature of 12-16^{o}C is ideal for growth and flower production. (Aswath *et al*., 2015)

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24. IIHRGO-1 (IC0632739; INGR20102), a Gerbera (*Gerbera jasmeonii*) Germplasm for Flower Colour and Flower Form: Bright Red (RHS colour: 40A, Red Group) and Double Type Flowers. Ability to Grow under Open Field Conditions.

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Gerbera, family *Asteraceae*, is one of the important cut-flowers grown for domestic and export markets. In India, Gerbera was grown in 1180 ha with production of 212 lakhs of cut flowers, amounting to the fourth most important cut-flower. Highest production of Gerbera comes from Uttarakhand with 7.80 (000' MT), while share of Karnataka is 6.2 (000' MT) (Anon., 2017-18). In India, all the gerbera varieties are imported and suitable for growing inside the polyhouse. However, to reduce the cost of cultivation, the genotype for growing outside is required. The Gerbera germplasm IIHRGO-1 was developed from the cross between IIHR99-5 × Savana followed by selection at F_1 stage, at ICAR-Indian Institute of Horticultural Research, Bengaluru, Karnataka (13° 58' N Latitude, 78°E Longitude and at an altitude of 890 meter above mean sea level), India. The germplasm IIHRGO-1 was evaluated for flower quality traits, reaction to insect-pests and diseases under open grown condition in Randomized Complete Block Design, during 2016-17 to 2018-19.The germplasm IIHRGO-1 is unique for flowers with bright red flower colour (RHS colour: 40A, Red Group), double type flowers and its ability to grow under open field conditions. It is suitable for bedding, cut flower and flower arrangement.

Morpho-agronomic characteristics: The Gerbera germplasm IIHRGO-1 flowers is having bright red colour (RHS colour: 40A, Red Group) and double type flowers. On an average, it has recorded flower stalk length (41.90 cm), stalk diameter (0.52 cm), flower diameter (8.51 cm), vase life (8.08 days) and number of flowers/plant/month (4.03) (Table 1).

Associated characters and cultivation practices: This hybrid selection possess moderate resistance to thrips and leaf miner. It has leaf thrips damaged (7.8%) during summer under Good Agricultural Practices (GAP), bud borer flowers damaged (4.89%) and leaf spot leaf damaged (5.12%) (Table 2).

Table 1. Evaluation of genetic stock IIHRGO-1 with Arka Krishika (check) for flower quality and flower yield traits (pooled data of three years)

Genotype	Stalk length (cm)	Stalk diameter (cm)	Flower diameter (cm)	Vase life (days)	Number of flowers/plant/month
IIHRGO-1	41.90	0.52	8.51	8.08	4.03
Arka Krishika	39.09	0.60	8.11	7.94	4.09
C.D. at 5%	NS	0.07	NS	NS	NS

Table 2. Flower quality traits, disease and insect-pests reaction of genetic stock IIHRGO-1 and Arka Krishika (check)

Trait	IIHRGO-1	Arka Krishika
RHS Colour Chart	Red Group, 40A	Yellow group, 10A
Flower form	Double	Semi-double
Thrips (% leaf damaged) during summer under GAP	7.8	6.9
Bud borer (% flowers damaged)	4.89	5.10
Leaf spot (% leaf damaged)	5.12	5.65

Gerbera is generally grown under polyhouse with shade net, however, IIHRGO-1 is highly suitable for open grown conditions. It grows best in well drained loamy soil, rich in organic matter, having adequate moisture holding capacity with soil pH (5.5-6.5) and EC less than 1 dS/cm^2. The suckers with 4-5 leaves to be planted on raised beds at a spacing of 60 cm between rows and 30 cm between plants accommodating 9 plants/m^2. The water requirement during the peak summer is 4-6 litres/m^2/day and 2 to 3 litres/m^2/day during the winter.

During bed preparation, a basal dose of FYM @ 20 kg/m^2, and first three months of planting apply 10:15:20 g NPK/m^2 and 15:10:30g NPK/month/m^2 from fourth month onwards (when flowering starts) in two splits at 15 days interval is good for establishment. It is multiplied through suckers and tissue culture.

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25. CSIR-IHBT-TM-09 (IC0630603; INGR20103), a Marigold (*Tagetes minuta*) Germplasm with High biomass yield 58.11kg/plot (24sqm). Essential Oil Content 0.343%.

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Wild marigold (*Tagetes minuta*; family Asteraceae) is an aromatic herb, commercially cultivated for its essential oil present in aerial parts of the plant. Volatile oil of wild marigold is used in perfumery and as a flavor component in food products, and have suppressive biological activity against different pathogens and insects (Vasudevan *et al*., 1997). Due to high demand of its essential oil, there has been increasing interest in the cultivation of this plant for commercial production (Singh *et al*., 2003). The (Z)-β-ocimene (52.01 %) content was highest in inflorescence while dihydrotagetone (84.85 %) content was highest in foliage (Kumar *et al*., 2020). In this context, with an aim of varietal development, selective breeding of wild marigold was done using progeny selection approach. Based on biomass production, nine breeding lines were selected for further evaluation in multi-location trials at four locations over two years. 'Him Gold' variety of wild marigold was used as the check variety. CSIR-IHBT-TM-09 performed well at all the locations with biomass yield of 58.11 kg/ plot (24 sqm) and essential oil content of 0.343% (3.43 g/ kg). The genotype CSIR-IHBT-TM-09 was developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The characteristic features of CSIR-IHBT-TM-09 are plant height of more than two meters, secondary branches more than 40 and large leaf size (leaf length more than 11 cm and width more than 5 cm) and has potential to be utilized as aromatic plant on commercial basis.

Associated characters and cultivation practices: Wild marigold is an annual crop suitable for cultivation in the plain and hilly areas, as a monocrop or intercrop. In India, wild marigold is found naturally in the western Himalayas between altitudes range of 1000–2500 m.

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26. CSIR-IHBT-TM-03 (IC0630602; INGR20104), aMarigold (*Tagetes minuta*) Germplasm for High Essential Oil Content: 0.375% (3.75g/kg).

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Wild marigold (*Tagetes minuta*; family Asteraceae) is an aromatic herb, commercially cultivated for its essential oil present in aerial parts of the plant. Volatile oil of wild marigold is used in perfumery and as a flavor component in food products, and have suppressive biological activity against different pathogens and insects (Vasudevan *et al*., 1997). Due to high demand of its essential oil, there has been increasing interest in the cultivation of this plant for commercial production (Singh *et al*., 2003). The (Z)-β-ocimene (52.01 %) content was highest in inflorescence while dihydrotagetone (84.85 %) content was highest in foliage (Kumar *et al*., 2020). Nine breeding lines were evaluated in multi-location trials at four locations over two years for essential oil content. 'Him Gold' variety of wild marigold was used as the check variety. CSIR-IHBT-TM-03 performed better than check at all the

locations with essential oil content of 0.375% (3.75 g/kg). The genotype CSIR-IHBT-TM-03 was developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The characteristic features of CSIR-IHBT-TM-03 are plant height of about two meters, high number of secondary branches (40). It has potential to be utilized as aromatic plant on commercial basis.

Associated characters and cultivation practices: Wild marigold is an annual crop suitable for cultivation in the plain and hilly areas, as a monocrop or intercrop. In India, wild marigold is found naturally in the western Himalayas between altitudes range of 1000–2500 m.

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27. CSIR-IHBT-RD-04 (IC0635435; INGR20105), a Damask Rose (*Rosa damascena*) Germplasm for High Flower Yield 4.92 kg/plot (12sqm).

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Damask rose (*Rosa damascena*) is an important essential oil bearing plant which has high demand globally in manufacture of perfumes, colognes and cosmetics. It originated in Damascus region of Asia minor and occupies one of the most important position as an aromatic plant for the extraction of essential oil. It is cultivated in Bulgaria, France, Italy, Turkey, Iran, Morocco and U.S.A for the production of attar (otto) of rose or oil of roses and is suitable for cultivation under sub-tropical and temperate conditions. It belongs to Rosaceae family and is an erect, perennial, hermaphrodite shrub possessing multiple green prickly stems up to 1−2 m in height, compound leaves with oval serrated leaflets. Flowering occurs during onset of summer season and continues for 30-35 days. The flowers are renowned for their fine fragrance, and are commercially harvested for rose oil (either "rose otto" or "rose absolute") used in perfumery and to make rose water and "rose concrete".

With an objective to improve productivity of damask rose through breeding, four clones of damask rose (*Rosa damascena*) along with check varieties 'Jwala' and 'Himroz' were evaluated for flower yield (kg) and essential oil content (mg/kg) at four locations in plains, mid- and high-altitude regions over a period of two years. Flower yield in CSIR-IHBT-RD-04 was 4.92 kg/plot (12 sqm plot) which was 22.6% higher than check variety Jwala. The genotype CSIR-IHBT-RD-04 was developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The characteristic features of CSIR-IHBT-RD-04 are plant height of nearly two meters when left unpruned. The plants are vigorous in growth and flower in the month of April and May. The essential oil content was observed to be *at par* with check varieties.

Associated characters and cultivation practices: Damask rose is suitable for cultivation in sub-tropical northern plains, mid hills and mild temperate regions (Kumar *et al*., 2013). The plants are short, compact, bushy and flower in March-April under sub-tropical conditions. It flowers for 25-30 days in a year.

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28. CSIR-IHBT-CH-14-1 (IC0635436; INGR20106), a Chrysanthemum (*Dendranthema grandiflora*) Germplasm with Yellow Flower Colour. Double Flower Shape (8.36cm diamter). Spray type.

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Chrysanthemum is one of the important ornamental cut flower plants globally and is ranked second among the top ten cut flowers sold in the global market. It highly attractive and charming flowering plant and commercially grown for cut flowers, garden display and pot culture (Kameswari *et al.,* 2014; Joshi *et al*., 2010; Jamal Uddin *et al*., 2015).Floriculture is a fast emerging in India due to its varied agro-climatic conditions (Suvija *et al*., 2016). Chrysanthemum belongs to the oldest ornamental plants of humanity and is still improved and cultivated (Halmagyi *et al*., 2004). It belongs to the family Asteraceae and is well adapted to different agro-climatic conditions. Chrysanthemum is a highly heterozygous plant that shows inbreeding depression and self-incompatibility; as a result, conventional crossbreeding is a powerful method for developing modern chrysanthemum cultivars (Su *et al*., 2019). To create unique flower types in Chrysanthemum F_1 hybrids were developed through a controlled crossing program. The F_1 chrysanthemum genotypes were morphologically characterized under field conditions with respect to floral traits and evaluated for agronomic performance. The selected F_1 hybrids of Chrysanthemum were evaluated for morphological and floral traits under protected cultivation over a period of four years from 2015 to 2018. Based on mean performance of these selections, the genotype CSIR-IHBT-CH-14-1 was found promising having yellow (9A, yellow group) coloured double flower shape, large size flowers 8.36 cm flower head diameter, suitable as cut flower chrysanthemum. The genotype CSIR-IHBT-CH-14-1 was F_1 selection from a cross between Yellow puma × White star, developed at CSIR - Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The genotype CSIR-IHBT-CH-14-1 has large flower head diameter of 8.36 cm, number of flowers /plant (54.76) and plant height is 72.28cm. It is suitable for spray and cut flower. It has a potential for commercial utilization as cut flower.

Associated characters and cultivation practices: Chrysanthemum is commercially propagated through terminal cuttings and suckers. It grows well in sandy loam soil rich in organic matter and nutrients with pH of 6.5 to 7.2.

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29. CSIR-IHBT-CH-14-2 (IC0635437; INGR20107), a Chrysanthemum (*Dendranthema grandiflora*) Germplasm with Brick Red Flower Colour with Bicoloured Florets (Yellow Colour on Floret Tips). Spray Type.

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Chrysanthemum is one of the most important ornamental cut flower plants and is ranked second among top ten cut flowers sold in the global market. It is commercially grown for cut flowers, garden display and pot culture (Kameswari *et al.,* 2014; Joshi *et al.,* 2010; Jamal Uddin *et al*., 2015). It belongs to the family Asteraceae and is well adapted to different agro-climatic conditions (Suvija *et al.,* 2016).Chrysanthemum is a highly heterozygous plant that shows inbreeding depression and self-incompatibility; as a result, conventional crossbreeding is a powerful method for developing modern chrysanthemum cultivars (Su *et al*., 2019). With an objective to develop unique flower types in chrysanthemum F_1 hybrids were developed through different cross combinations in controlled crossing program. The hybrid F_1 genotypes were morphologically characterized under field conditions with respect to floral traits and evaluated for agronomic performance. The selected F_1 hybrids of chrysanthemum were evaluated for morphological and floral traits under protected cultivation over a period of four years from 2015 to 2018. Based on mean performance, the genotype CSIR-IHBT-CH-14-2 was found promising having brick red (185A, greyed purple) flowers with bicoloured florets (yellow colour on floret tip 5A, yellow group) spray chrysanthemum. The genotype CSIR-IHBT-CH-14-2 was F_1 selection from Yellow Puma × Shyamal, developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The genotype CSIR-IHBT-CH-14-2 has flower head diameter of 6.19 cm, number of flowers /plant 48.56 and plant height is 63.46 cm. It is spray type chrysanthemum.

Associated characters and cultivation practices: Chrysanthemum is commercially propagated through terminal cuttings and suckers. It grows well in sandy loam soil rich in organic matter and nutrients with pH of 6.5 to 7.2.

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30. CSIR-IHBT-CH-14-4 (IC0635438; INGR20108), a Chrysanthemum (*Dendranthema grandiflora*) Germplasm for Pink Flower Colour. Flower Diameter 9.94cm. Plant Height 100.97cm.

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Chrysanthemum is one of the most important ornamental cut flower plants and is ranked second among top ten cut flowers sold in the global market. It is commercially grown for cut flowers, garden display and pot culture (Kameswari *et al.,* 2014; Joshi *et al*., 2010; Jamal Uddin *et al*., 2015). It belongs to the family Asteraceae and is well adapted to different agro-climatic conditions (Suvija *et al.,* 2016). Chrysanthemum is a highly heterozygous plant that shows inbreeding depression and self-incompatibility; as a result, conventional crossbreeding is a powerful method for developing modern chrysanthemum cultivars (Su *et al*., 2019). With an objective to develop unique flower types in chrysanthemum F_1 hybrids were developed through different cross combinations in controlled crossing program. The hybrid F_1 genotypes were morphologically characterized under field conditions with respect to floral traits and evaluated for agronomic performance. The selected F_1 hybrids of chrysanthemum were evaluated for morphological and floral traits under protected cultivation over a period of four years from 2015 to 2018. Based on mean performance, the genotype CSIR-IHBT-CH-14-4 was found unique having pink (61A, red purple group) flowers, double korean flower shape, flower diameter 9.94 cm, plant height 100.97 cm. The genotype CSIR-IHBT-CH-14-4 was F1 selection from Yellow Puma × Shyamal, developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The genotype CSIR-IHBT-CH-14-4 has flower head diameter of 9.94 cm, number of flowers /plant 46.12 and plant height is 100.97cm. It has a potential for commercial utilization as cut flower.

Associated characters and cultivation practices: Chrysanthemum is commercially propagated through terminal cuttings and suckers. It grows well in sandy loam soil rich in organic matter and nutrients with pH of 6.5 to 7.2.

References

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31. CSIR-IHBT-CH-14-8 (IC0635439; INGR20109), a Chrysanthemum (*Dendranthema grandiflora*) Germplasm for Dark Pink Flower colour. Spatulate (Fluted) Florets. Flower diameter 7.99cm.

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Chrysanthemum is one of the important ornamental cut flower plants globally and is ranked second among the top ten cut flowers sold in the global market. It highly attractive and charming flowering plant and commercially grown for cut flowers, garden display and pot culture (Kameswari *et al.,* 2014; Joshi *et al.,* 2010; Jamal Uddin *et al.,* 2015). Floriculture is a fast emerging in India due to its varied agro-climatic conditions (Suvija *et al.,* 2016). Chrysanthemum belongs to the oldest ornamental plants of humanity and is still improved and cultivated (Halmagyi *et al.,* 2004). It belongs to the family Asteraceae and is well adapted to different agro-climatic conditions. Chrysanthemum is a highly heterozygous plant that shows inbreeding depression and self-incompatibility; as a result, conventional crossbreeding is a powerful method for developing modern chrysanthemum cultivars (Su *et al*., 2019). To create unique flower types in Chrysanthemum F_1 hybrids were developed through a controlled crossing program. The F_1 chrysanthemum genotypes were morphologically characterized under field conditions with respect to floral traits and evaluated for agronomic performance. The selected F_1 hybrids of Chrysanthemum were evaluated for morphological and floral traits under protected cultivation over a period of four years from 2015 to 2018. Based on mean performance of these F_1 hybrids, the genotype CSIR-IHBT-CH-14-8 was found unique having dark pink flowers (59A, red purple group) with quilled/spatulate florets, flower diameter 7.99 cm, spray chrysanthemum. The genotype CSIR-IHBT-CH-14-8 was F_1 selection from Purnima × Shyamal, developed at CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh (Latitude: 32.0934° N, Longitude: 76.5439° E and at an altitude of 1300m amsl).

Morpho-agronomic characteristics: The genotype CSIR-IHBT-CH-14-8 has flower head diameter of 7.99 cm, number of flowers /plant 32.98 and plant height is 75.20cm. It is spray type chrysanthemum and has a potential for commercial utilization as cut flower as well as garden purpose.

Associated characters and cultivation practices: Chrysanthemum is commercially propagated through terminal cuttings and suckers. It grows well in sandy loam soil rich in organic matter and nutrients with pH of 6.5 to 7.2.

References

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32. AS 04-1687 (IC0636675; INGR20110), a Sugarcane (*Saccharum officinarum*) Germplasm with Drought and Water Logging Tolerance.

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Development and identification of climate resilient crop varieties with enhanced tolerance to heat, drought, salinity, excess water/ flooding, chilling are essential in order to sustain and improve crop yields to manage the challenges of climate change (Maheshwari *et al*., 2015). Introgression of abiotic tolerant genes into climate resilient varieties depends upon the availability of potential sources from wild relatives and their exploitation. *Saccharum spontaneum* has been utilized in sugarcane breeding since 1912 due to its wider adaptability under biotic and abiotic conditions. The proposed genetic stock AS 04-1687 having drought and waterlogging tolerance is an interspecific hybrid derived from a cross between sugarcane commercial cultivar (BO 102) and the wild species *S. spontaneum* (IND 84-337) having the cytotype 2n=56 which was not used in the breeding programme earlier. The hybrid AS 04-1687 is characteristic with erect habit, purple coloured cylindrical internode and leaf sheath which is tight and glabrous with brown coloured dewlop and deltoid auricle.

Evaluation for drought and water logging tolerance: Twenty-seven ISH/IGH hybrids with diverse genetic base were tested for tolerance to drought at four AICRP(S) centres and water logging at 3 centres located in both tropical (Padegaon and Anakapalle/ Kolhapur and Vuyyuru) and subtropical (Karnal and Faridkot/ Pusa) regions along with 3 standards for each centre in replicated trials (Alpha design) during 2016-17 (Plant crop) and 2017-18 (Plant and ratoon crops). Drought was imposed by withdrawing irrigation between 60 and 150 days after planting and harvesting in plant and ratoon crops respectively. Water logging was imposed under either natural water logging or water stagnation (minimum 15 cm) condition for 150-210 days after planting/harvesting. Data obtained at harvest from all the centres under both normal and drought/waterlogging conditions in two plant and one ratoon crops for the important traits viz., cane yield (t/ha), CCS yield (t/ha), juice sucrose %, single cane weight (kg), cane diameter, tillers mortality %, relative water content after drought and number of millable canes (‘000/ha) were considered for pooled analysis and percent changes due to drought and waterlogging were estimated.

Performance of the proposed clone AS 04-1687 under drought conditions: Among the entries evaluated, the clone AS 04-1687 exhibited the best performance with less than 20% reduction for cane yield t/ha (18.5%), CCS t/ha (10.08%) and NMC (‘000/ha) (13.01%) under drought condition while in the checks, reductions were 29.77%, 29.73% and 17.57% respectively for these three characters. The traits viz., cane diameter and relative water content after drought had shown least impact due to drought condition with 1.01% and 2.11% reductions respectively. In the checks 3.58% reduction for cane diameter and 4.31% reduction for relative water content after drought were observed. Tillers mortality % was less in AS 04-1687% with 30.09% while it was higher with 39.84 in the checks (Table 1). Among the entries

Table 1. Performance of the proposed clone AS 04-1687 under drought conditions

Characters	AS 04 -1687			Checks***		
	Normal	Drought	% change	Normal	Drought	% change
Cane yield t/ha	116.15	94.66	-18.51	94.0	66.01	-29.77
CCS t/ha	8.79	7.9	-10.08	12.15	8.53	-29.73
NMC* (000’/ha)	173.12	150.61	-13.01	83.8	69.07	-17.57
Cane Diameter (at 360 days)	1.97	1.95	-1.01	2.65	2.54	-3.58
Tillers mortality %	33.45	30.09	-10.07	39.97	39.84	-0.32
RWC** after drought	78.43	76.77	-2.11	83.66	80.05	-4.31

* Number of millable canes (Thousands/ha)**RWC Relative water content
***Mean of 9 checks viz., Co 92005, CoM 0265, Co 86032, CoV 94101, CoV 92102, CoV 09356, BO 91, BO 154 and BO 145
(Source: PICI – AICRP(S) annual report for the year 2016-17 and 2017-18)

tested, the clone AS 04-1687 was identified as a best clone for drought tolerance.

Performance of the proposed clone AS 04-1687 under waterlogging conditions: Pooled analysis indicated that the proposed clone AS 04-1687 exhibited low percent reduction for many traits after the waterlogging period. The clone showed 2.01%, 8.26 % and 5.20% reduction for CCS t/ha, single cane weight (kg) and NMC (000'/ha) respectively while the checks showed 19.79%, 20.04% and 10.05% reduction respectively for these traits (Table 2). Cane yield also exhibited less reduction (16.42%). Sucrose % was unaffected in AS 04-1687 while the checks recorded 5.24 % reduction. Considering the superior performance for the above traits, AS 04-1687 was considered as a better waterlogging tolerant clone.

The interspecific genetic stock AS 04-1687 developed through hybridization between the subtropical commercial variety- BO 102 and *S. spontaneum* (IND 84-337) is identified as a superior genetic stock (IC0636675:INGR20110) as it is tolerant to both drought and waterlogging conditions. The clone has broad genetic base as it was developed from the new cytotype 2n=56 of *Saccharum spontaneum* as the present day varieties are the derivatives of the cytotype 2n=64 and 2n=112. This clone can be effectively utilized in sugarcane breeding to develop climate resilient varieties to withstand drought as well as water logging conditions.

Reference

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Table 2. Performance of the proposed clone AS 04-1687 under waterlogging condition

Characters	AS 04- 1687			Checks**		
	Normal	Water logging	% change	Normal	Water logging	% change
Cane yield t/ha	90.86	75.94	-16.42	74.23	61.26	-17.47
CCS t/ha	8.14	7.98	-2.01	10.51	8.43	-19.79
Sucrose % at harvest	12.91	13.02	0.85	18.69	17.71	-5.24
Single Cane Weight (kg)	0.73	0.67	-8.26	1.05	0.84	-20.04
NMC (000'/ha)*	108.84	103.18	-5.20	67.96	61.13	-10.05

* Number of millable canes (Thousands/ha)
** Mean of 12 checks viz., Co 86032, CoM 0265, CoM 88121, 83 R 23, CoA 06231, CoA 92081, Co 0238, BO 91, CoJ 88 (two locations), Co 98014 (two locations)
(Source: PICI – AICRP(S) annual report for the year 2016-17 and 2017-18)

33. BM 1010-168 (IC0636674; INGR20111), a Sugarcane (*Saccharum* sp.) Germplasm Tolerant to Drought. High Relative Water Content under Drought.

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Drought is the important yield limiting factor in sugarcane which is recurrent now-a-days hence development of drought tolerant sugarcane varieties in the climate change scenario is assuming more importance. INGR 20111, a genetic stock possessing drought tolerance and high relative water content under drought was the selection from Co 98010 × (Co 1148 x SES 404) at ICAR-Sugarcane Breeding Institute, Coimbatore. It was evaluated for drought tolerance in two plant and one ratoon crops under AICRP(S) programme in four locations viz., Padegaon (Peninsular Zone), Anakapalle (East Coast Zone), Faridkot (North West Zone) and Karnal (North West Zone). Drought was imposed by withdrawing irrigation between 60 and 150 days and data were compared with the normal crop raised under recommended irrigation.

INGR 20111 recorded 73.28 t/ha of cane yield under drought condition and showed 21.19 % reduction compared to normal condition while the checks recorded higher 29.78 % reduction (Table 1). Relative water content was unaffected due to imposition of drought in INGR 20111, but the 4.31 % reduction was observed with the checks. The clone showed less reduction for cane yield, CCS t/ha, single cane weight, number of

millable canes and relative water content after drought period hence can be used as a potential parent for the developing drought tolerant varieties. The genetic stock has long and erect canes with long internodes.

SES 404, a new *S. spontaneum* species clone was not exploited in sugarcane varietal development programme so far hence utilization of the proposed genetic stock with SES 404 in its pedigree as parent in hybridization will help in broadening the genetic base of the new sugarcane varieties. This genetic stock was resistant and moderately resistant to red rot for cf671 and cf94012 pathotypes respectively.

Table 1. Performance of INGR 20111 under drought and normal conditions

Characters	BM 1010-168			Checks**		
	Normal	Drought	% change	Normal	Drought	% change
Cane yield t/ha	92.99	73.28	-21.19	94.00	66.01	-29.78
CCS t/ha	9.49	7.64	-19.47	12.15	8.53	-29.79
Single Cane Weight (kg)	0.83	0.74	-10.46	1.14	0.99	-13.12
NMC (000'/ha)*	115.58	96.13	-16.83	83.80	69.07	-17.58
Relative Water Content after drought	80.30	80.31	0.01	83.66	80.05	-4.31

* Number of millable canes (Thousands/ha)

** Mean of 12 checks viz., Co 86032, CoM 0265, CoM 88121, 83 R 23, CoA 06231, CoA 92081, Co 0238, CoPb 91, CoJ 88 (two locations), Co 98014 (two locations)

Reference

Annual reports (2016-17) and (2017-18) Crop Improvement – AICRP (Sugarcane).

34. SBIEC 14006 (IC0636673; INGR20112), a Wild Sugarcane (*Erianthus arundinaceus*) Germplasm for High Harvestable Biomass. High Fibre Content.

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Biomass based industries like power generation, paper production and particle board manufacturers require quality and uninterrupted supply of biomass as raw materials. INGR 20112 was first generation selection from the open pollinated fluff of *E. arundinaceus* clone IK 76-75 at ICAR-Sugarcane Breeding Institute, Coimbatore. It recorded the highest mean harvestable biomass of 265.28 t/ha across three plant and one ratoon crops with average fibre content of 27.54% (Table). It also recorded 2.18 cm cane diameter and 1.24 kg of single cane weight. The cane grows up to 4 -5 m tall

Table. Performance of SBIEC 14006 for harvestable biomass yield (t/ha) and fibre % in cane under four environments

Clones	HBM t/ha					Fibre %				
	2013-14 (P)	2014-15 (P)	2015-16 (P)*	2016-17 (R)**	Mean	2013-14	2014-15	2015-16 (P)	2016-17 (R)	Mean
SBIEC 14001	132.12	145.23	182.00	191.23	162.64	24.23	25.21	26.11	26.48	25.51
SBIEC 14002	186.92	200.00	234.22	206.75	206.97	20.41	20.48	21.32	22.20	21.10
SBIEC 14003	158.08	159.14	212.15	226.78	189.04	21.98	20.27	20.18	21.11	20.88
SBIEC 14004	136.15	161.34	185.21	186.27	167.24	25.24	22.04	25.00	23.13	23.85
SBIEC 14005	143.08	158.12	162.16	194.25	164.40	26.50	23.38	25.88	23.28	24.76
SBIEC 14006	233.65	249.29	282.83	295.34	265.28	27.24	26.99	28.22	27.70	27.54
INGR 12017	203.00	217.12	227.77	228.98	219.22	21.17	20.79	21.58	20.05	20.90
CD	24.04	28.16	23.08	17.37		2.86	1.83	1.09	1.22	
CV	12.23	15.86	17.95	14.66		5.97	7.26	10.03	12.52	

* Plant crop ** Ratoon crop

in 12 months. The leaf sheath is tightly attached to the cane hence available up to harvest without wasting the biomass. Tall and non-lodging nature of canes makes the genetic stock amenable for mechanical harvesting. It can be ratooned for at least 7-8 years hence no need for replanting every year which brings down the cost of cultivation in ratoon crop. This is as an ideal Type II energy cane due to more biomass yield per unit area and requires low input and low nutrient requirements and incurs less production cost.

Reference

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35. Pune Selection-2 (IC0637024; INGR20113), a Papaya (*Carica papaya*) GermplasmTolerant to Papaya Ringspot Virus and Yellow Flesh

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Pune Selection-2 (PS-2) is a near homozygous, yellow (mango-yellow) fleshed, dioecious papaya (*Carica papaya* L.) line. The line is consistently showing field tolerance to *Papaya Ring Spot Virus* strain papaya (PRSV-P) with appreciable yield (Sharma and Tripathi, 2019). The parental material was a segregating local collection of papaya named 'Madhubala'. PS-2 was selected from the segregating population (Datar *et al*., 2013). Since then, it is developed into a line by sib-mating and selection at IARI, Regional Station, Pune. The PRSV tolerant PS-2 line has good potential in resistance breeding and as target variety for vegetable purpose specifically in Northern States of India (Sharma *et al.,* 2017; Mahapatro *et al*., 2019).

Morpho-agronomic characteristics: Mean height of the plant is 164 cm, mean stem girth is 40 cm, leaf shape is palmate type and petiole colour is green. Petal colour is cream yellow. Average fruiting height is 91 cm. Average length of the fruiting column is 45cm. Mean fruit weight is 1455g. Average yield per plant is 25.98 kg. Shape of the fruit is oblong. Average thickness of flesh is 3.22 cm with yellow colour and average TSS value of 8.16 ºBrix. Average intensity of PRSV intensity of Red Lady was 20.48 which were four times higher than Pune Selection-2 (5.88). The line performed better than both the checks (papaya cv. Red Lady and Pusa Nanha) under severe PRSV-P pressure.

Associated characters and cultivation practices: PS-2 exhibits remarkable field tolerance to PRSV-P. It shows late and mild PRSV infection. Growth, fruiting characters and incidence of PRSV-P in PS-2 along with two checks of papaya, namely, Red Lady and Pusa Nanha are given in Table 1.

Under Pune conditions, it is recommended to plant seedlings having six to eight leaves in spring season (February-March) since the virus transmitting aphid-vector population is minimal from February to June. Seedlings should be raised in insect-proof polyhouse. Being a dioecious line, two seedlings per hill should be planted to maintain higher ratio of productive female plants. Per plant space required is 4.3 to 4.4 square meters that can be achieved by maintaining row to row and plant to plant distance of 2.4m × 1.8 m respectively or by square plantation of 2.1 m × 2.1 m. About 2,300 plants can be accommodated per hectare under both spacing. One square foot FYM, 2 kg neem seed cake and 1 kg sterameal should be applied per hill before plantation. Inorganic fertilizers N:P:K at the rate of 300:300:300 g/plant should be applied in four split doses at alternate month. Foliar application of a balance mix of all micronutrients (2g/L) at alternate month along with additional spray of boron (2g/L) at the time of fruit setting, and calcium (2g/L) before fruit ripening is recommended.

Papaya is one of the cite-worthy fruit crops of the tropical region with major commercial importance owing to its rich nutritive and medicinal value. However, its true potential has remained under-exploited due to inadequate quality planting materials of right varieties, high pre- and post-harvest losses. This crop is ought to be popularized amongst Indian farmers as a nutrition-rich source for poor; and moreover, the PRSV tolerant

Table 1. Comparison of growth, fruiting characters, yield and PRSV reaction of Pune Selection-2 with local checks (Red Lady and Pusa Nanha

Variety	PS-2				Red Lady				Pusa Nanha			
Characters	2016-17	2017-18	2018-19	Avg.	2016-17	2017-18	2018-19	Avg.	2016-17	2017-18	2018-19	Avg.
Plant Height (cm)	167	174	151	164	148	175	147	157	103	106	91	100
Stem girth (cm)	37	45	38	40	35	46	36	39	29	32	27	29
Fruiting Height (cm)	107	82	83	91	86	90	75	84	45	55	45	48
Fruiting Column Length (cm)	27	64	44	45	31	54	51	45	36	41	39	39
Yield (kg/plant)	19	36.44	22.5	25.98	15.47	19.13	17	17.20	19	13.06	16.69	16.25
Average fruit weight (g)	1520	1470	1375	1455	1289	1677	1087	1351	1215	712	879	935
Flesh Thickness (cm)	4	2.67	3	3.22	2.6	2.83	2.5	2.64	2.88	2.67	2.83	2.79
Flesh Colour	Yellow				Red				Yellow			
TSS (ºBrix)	8	7.83	8.67	8.167	9.8	9.17	8	8.99	8.71	8.17	10	8.96
PRSV Intensity (%)	5.82	0	11.83	5.88	25.58	6.45	29.41	20.48	18.06	3.45	4.76	8.76

PS lines (Mahapatro *et al*., 2019), are advocated as vegetable also.

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36. Pune Selection-5 (IC0637025; INGR20114), a Papaya (*Carica papaya*) Germplasm Tolerant to Papaya Ring Spot Virus and Yellow Flesh.

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Pune Selection-5 (PS-5) is a near homozygous, yellow fleshed, dioecious papaya (*Carica papaya* L.) line. The line is consistently showing field tolerance to *Papaya Ring Spot Virus* strain papaya (PRSV-P) with good yield (Sharma and Tripathi, 2019). The parental material was a segregating local collection of papaya named 'Madhubala'. PS-5 was selected from the segregating population (Datar *et al*., 2013). Since then, it is developed into a line by sib-mating and selection at IARI, Regional Station and Pune. The PRSV tolerant PS-5 line has good potential in resistance breeding and as target variety for vegetable purpose in Northern States of India (Sharma *et al.,* 2017 b).

Morpho-agronomic characteristics: Mean height of the plant is 141 cm, mean stem girth is 31 cm, leaf shape is palmate type and petiole colour is green. Petal colour is yellow. Average fruiting height is 77cm. Average length of the fruiting column is 49 cm. Mean fruit weight is 1280 g. Average yield per plant is 23.63 kg. Shape of the fruit is oblong. Average thickness of flesh is 3.31 cm with yellow colour and average TSS value of 8.11 ºBrix. Average intensity of PRSV infection was 4.34%. The line performed better than both the checks (papaya cv. Red Lady and Pusa Nanha) under severe PRSV-P pressure.

Associated characters and cultivation practices: PS-5 is having field tolerance to PRSV-P. It shows late and mild PRSV infection. Growth, fruiting characters and incidence of PRSV-P in PS-5 along with two checks of papaya, namely, Red Lady and Pusa Nanha are given in Table 1. Under Pune conditions, it is recommended to plant seedlings having six to eight leaves in spring season (February-March) since the virus transmitting aphid-vector population is minimal from February to June. Seedlings should be raised in insect-proof polyhouse. Being a dioecious line, two seedlings per hill should be

Table 1. Comparison of growth, fruiting characters, yield and PRSV reaction of Pune Selection-5 with local checks (Red Lady and Pusa Nanha)

Variety	PS-5				Red Lady				Pusa Nanha			
Characters	2016-17	2017-18	2018-19	Avg.	2016-17	2017-18	2018-19	Ave.	2016-17	2017-18	2018-19	Avg.
Plant Height (cm)	154	139	130	141	148	175	147	157	103	106	91	100
Stem girth (cm)	30	35	29	31	35	46	36	39	29	32	27	29
Fruiting Height (cm)	91	70	71	77	86	90	75	84	45	55	45	48
Fruiting Column Length (cm)	35	59	52	49	31	54	51	45	36	41	39	39
Yield (kg/Plant)	15.73	31.01	24.16	23.63	15.47	19.13	17	17.20	19	13.06	16.69	16.25
Average fruit weight (g)	1092	1267	1481	1280	1289	1677	1087	1351	1215	712	879	935
Flesh Thickness (cm)	4	2.67	3.25	3.31	2.6	2.83	2.5	2.64	2.88	2.67	2.83	2.79
Flesh Colour	Yellow				Red				Yellow			
TSS (ºBrix)	8	7.67	8.67	8.11	9.8	9.17	8	8.99	8.71	8.17	10	8.96
PRSV Intensity (%)	8.33	0	4.68	4.34	25.58	6.45	29.41	20.48	18.06	3.45	4.76	8.76

planted to maintain higher ratio of productive female plants. Per plant space required is 4.3 to 4.4 square meters that can be achieved by maintaining row to row and plant to plant distance of 2.4m × 1.8 m respectively or by square plantation of 2.1 m × 2.1 m. About 2,300 plants can be accommodated per hectare under both spacing. One square foot FYM, 2 kg neem seed cake and 1 kg sterameal should be applied per hill before plantation. Inorganic fertilizers N:P:K at the rate of 300:300:300g/plant should be applied in four split doses at alternate month. Foliar application of a balance mix of all micronutrients (2g/L) at alternate month along with additional spray of boron (2g/L) at the time of fruit setting, and calcium (2g/L) before fruit ripening is recommended.

Papaya is one of the cite-worthy fruit crops of the tropical region with major commercial importance owing to its rich nutritive and medicinal value. However, its true potential has remained under-exploited due to inadequate quality planting materials of right varieties, high pre- and post-harvest losses. This crop is ought to be popularized amongst Indian farmers as a nutrition-rich source for poor; and moreover, the PRSV tolerant PS lines (Mahapatro *et al*., 2019), are advocated as vegetable also.

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37. IPC 126A & IPC 126B (IC635036 &IC635037; INGR20115), a Dark purple (Black) Tropical Carrot (*Daucus carota*) CMS Line.

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Black carrot is rich in anthocaynins, which are powerful antioxidants and anticancer bioactive phytochemicals. For developing black coloured anthocyanin rich hybrids in carrot, CMS line is essential. Multinutrient rich multicoloured carrot hybrids will have potential for fresh market as well as industry. For this, a natural petaloid (anthers turn to petal like structures) CMS system found in an already developed variety 'Pusa Rudhira' (IPC-122) at Division of Vegetable Science, ICAR-IARI, New Delhi (Kalia *et al*., 2019), which was used as a source

material for introgression of CMS system into IPC 126 (Pusa Asita) and the maintainer was also searched in this background and CMS line 'IPC 126A' was established which produces black roots with self core colour like that of maintainer.

Morpho-agronomic characteristics of CMS line IPC 126A: The CMS line IPC 126A is the first CMS line of tropical black carrot developed indigenously. It is suitable for sowing during main season (*i.e.* October to January). For fresh consumption; 90 days old roots are ready to harvest (Table 1). Leaves are purple green and leaf petiole colour is purple. It produces dark purple or black colour, long roots with self-core character. The average root length was found to be 24.0 cm, root diameter 30.8 cm, core diameter 11.54cm and root weight 95 g. The height of flowering plants was 126.5 cm with profuse flowering umbels and seed setting. The floral traits *i.e.* petal size, petal colour, petaloid (anther converted petals) colour and size and nectaries showed normal development. The marketable root yield at fresh consumption stage was recorded to be 14.6 t/ha which was lower than its maintainer line IPC 126B (17.8t/ha) at harvest (Table 1). The processing industry requirement is bulky roots (120-130 days) which yield around 22.2 t/ha for IPC 126A and 30.1 t/ha for IPC 126B lines. Unlike European type, IPC 126A and IPC 126B does not require vernalization and produce seeds in plains during winter season. The bolting (elongation of flower stalk) occurs during February-March month and produce abundant seeds which are harvested during April-May months in plains of India. The CMS line showed potential in hybrid breeding of black and multicoloured multinutrient rich carrot hybrids, which can be used in commercial hybrid breeding programme.

IPC 126B: male fertile maintainer line of CMS line IPC 126A: The 'IPC 126B' or IPC 126 has been released as Pusa Asita, an open-pollinated variety of black colour carrot for main season cultivation in Delhi region. It is a variety of tropical type black carrot and suitable for sowing during September to October months in plains

Table 1. Important horticultural traits and seed yield of CMS line IPC 126A line as compared to the fertile maintainer line IPC 126B

Traits	IPC 126A	IPC 126B
Maturity traits		
Growing season	Main season	Main season
Sowing period	September – October	September – October
Harvesting for fresh consumption	December – January	December – January
Harvesting for processing*	January – February	January – 1st Fortnight February
Plant and root traits		
Root skin colour	Dark purple	Dark purple
Root core colour	Dark purple (self-core)	Dark purple (self-core)
Plant height (cm)	68.4	87.5
Gross plant weight (cm)	185	215.0
Root length (cm)	24.0	25.6
Root diameter (mm)	30.8	37.4
Core diameter (mm)	11.54	10.8
Root weight for fresh consumption (g)	95.0	115.0
Root yield for fresh consumption (t/ha)	14.6	17.8
Root weight (g)	195.0	200.7
Root yield for processing purple (t/ha)	22.2	30.1
Floral traits		
Petal colour	Light purple	Light purple
Petal length (cm)	1.5	1.11
Petaloid colour	Light purple	-
Petaloid length (cm)	1.30	-
Petaloid width (cm)	0.47	-
Style length (cm)	1.57	1.16
Petaloid shape	Spoon	-
Nectary development	Prominent	Prominent
Observation on natural seed setting	Abundant	Abundant

*Require more time for bulkiness and colour accumulation.

of north India. It matures in 90 days after sowing during December to January months. Plants vigorous, erect, medium to large in spread and leaves are purple green with purple petiole. It produces dark purple or black roots with self core colour (Table 1).

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38. IPC 98A & IPC 98B (IC0598343 & IC0637028; INGR20116), a Red Colour Tropical Carrot CMS Line (*Daucus carota*)

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Carrot (*Daucus carota* L.; 2n= 2x=18) is an important root vegetable crop being grown worldwide. Tropical red colour carrot is rich in lycopene, a powerful anticancer antioxidant. In India, carrot is grown on 86,200 ha area and production is 1,379,031 MT with productivity of 15.9 t/ha (NHB Database 2017). Hybrids are superior for uniformity, higher yield, earliness and have higher recovery of marketable roots per unit area. But, lack of cytoplasmic male sterile (CMS) lines in tropical types having red colour was major reason which hampered the progress of hybrid breeding in India. Fortunately, a 'petaloid' type sterile cytoplasm was found as a natural mutant in tropical red carrot genotype 'IPC-122' at Division of Vegetable Science, ICAR-IARI, New Delhi which was tapped and introgressed into different elite genotypes. It was introgressed into 'IPC 98', an elite genotype of tropical red carrot by backcross breeding (Kalia *et al.,* 2019) and used in development of only tropical carrot hybrid 'Pusa Vasuda' as female parent. This CMS line is expected to play a key role in establishment of indigenous hybrid seed industry of tropical carrot.

Morpho-agronomic characteristics of CMS line IPC 98A: The 'IPC 98A'is the first CMS line of tropical carrot having red coloured self-core roots developed indigenously. It is suitable for sowing during main crop season in north Indian plains (*i.e.* September to October) and roots gets ready for harvest in 90-100 days (Table 1). The height of flowering plants was 100.5 cm with profuse flowering and seed setting. The floral traits *i.e.* petal size, petal colour, petaloid (anther converted petals) colour, size and nectaries showed normal development. It produces red colour, long roots with self-core character.

Table 1. Important horticultural traits and seed yield of CMS line IPC 98A line as compared to the fertile maintainer line IPC 98B

Traits	IPC 98A	IPC 98B
Maturity traits		
Growing season	Main season	Main season
Sowing period	September – October	September – October
Harvesting period	December – January	December – January
Plant and root traits		
Plant height (cm)	87.4	89.1
Gross plant weight (cm)	185.0	254.7
Root colour	Red	Red
Root core colour	Red	Red
Root length (cm)	24.4	24.8
Root diameter (mm)	31.0	38.5
Core diameter (mm)	9.4	11.2
Root weight (g)	151.0	164.5
Root yield (t/ha)	30.0	29.25
Floral traits		
Petal colour	White	Green
Petal length (cm)	1.49	1.01
Petaloid colour	Light purple	-
Petaloid length (cm)	1.86	-
Style length (cm)	0.94	0.61
Petaloid width (cm)	0.90	-
Petaloid shape	Trident	-
Nectary development	Prominent	Prominent
Honey bee visit	Abundant	Abundant

Average root length of IPC 98A was found to be 24.4 cm, root diameter 31.0 mm, small core diameter 9.4 mm and root weight 151.0 g (Fig. 3A-B). The marketable root yield was recorded to be 30.0 t/ha which was at par with its maintainer line IPC 98B (29.25t/ha) (Table 1). The bolting (elongation of flower stalk) occurs during February–March months and produce seeds profusely which are harvested during April-May months in plains. This CMS line has been used in breeding 'Pusa Vasuda'

red root hybrid for the main season. The IPC 98A performed well in different hybrid combinations showing potential, thereby, in commercial hybrid breeding of indigenous tropical hybrids.

IPC 98B: male fertile maintainer line of CMS line IPC 98A: The 'IPC 98B' is an elite genotype of main season tropical carrot. It is suitable for sowing in September to October in plains of north India. It matures during December to January months in 90-100 days after sowing. Plants are medium, vigorous, semi-erect, medium in spread and leaves are green. It produces red root with self-core colour (Table 1).

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39. YF 5-2-7 (IC-633085) (IC0633085; INGR20117), a Watermelon (*Citrullus lanatus*) Germplasm with Saffron Coloured Flesh and High Carotenoid Content. Non-Lobed (Entire) Leaves

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Watermelon [*Citrulluslanatus* (Thunb.) Mansf.] is an important crop grown in different parts of country. It is one of the most widely cultivated crops in the world and its global consumption is greater than that of any other cucurbit. Presently the red fleshed varieties are widely cultivated in India. The major nutritional components in watermelon consist of carbohydrates (6.4 g/ 100g), vitamin A (590 IU) and lycopene (4100 µg/ 100g) in red flesh varieties. Presently the red fleshed varieties are widely cultivated in India which contain low amount of carotenoid content. Now a day's people are very conscious to health issues and there is demand of varieties rich in carotenoids. Therefore, breeding for specific flesh coloured varieties having high nutritive value are often a challenge to attract consumers.

Watermelon showed a wide range of genetic variability in quantitative and qualitative traits (Choudhary *et al*., 2016). Watermelon being highly cross pollinated crop possess varying flesh colour *viz*., red, white, yellow and saffron having different profile of nutrients. Keeping in view, identified and homogenized a saffron flesh coloured genotype of watermelon (YF 5-2-7) having high carotenoid content. YF 5-2-7 is high in carotenoid content (7.10-9.18 µg/ g FW) in comparison to popular red fleshed varieties which have 3.92-4.14 µg /g FW carotenoid content. It is characterized by non-lobed (entire) leaves, round fruits having dark green rind with very narrow stripes, saffron flesh and blackish brown seeds. YF 5-2-7 produced round fruits weighing 2.5-3 kg, rind thickness (1.0-1.3 cm), TSS (10-11%) and bear 3-4 fruits/plant. Fruits ready for harvesting in 80-85 days after sowing. The analysis of carotenoid content was done using the biochemical method as suggested by Hartmut and Alan, 1983.

Table 1. Salient characteristics of YF 5-2-7 (IC-0633085)

Trait	Description
Days to first fruit harvest after sowing	80-85 days
Number of fruit/ plant	3-4
Fruit weight	2.5-3.0 kg
Fruit diameter	14.8-18.0 cm
Rind thickness	1.0-1.3 cm
TSS	10.0-11.0%
Carotenoid content	7.10-9.18 µg/ g FW
Sex form	Monoecious
Leaf shape	Non-lobed (Entire)
Fruit shape	Round
Rind colour	Dark green with very narrow stripes
Flesh colour	Saffron

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Indian Journal of Plant Genetic Resources (IJPGR) is the official publication of the Indian Society of Plant Genetic Resources. Aim of the Journal is to disseminate knowledge on plant genetic resources (PGR) research and application. Being the only journal in the area of PGR, the journal aims to provide a forum for discussion and debate on current issues of PGR. For publication in the journal, the authors must be a member of the Society. IJPGR publishes full-length papers or short communications of original scientific research in the field of plant genetic resources. Review articles (with prior consent or invitation only) summarizing the existing state of knowledge in topics related to plant genetic resources will also be published.

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- Status reports on ecology, conservation, traditional knowledge and use of PGR

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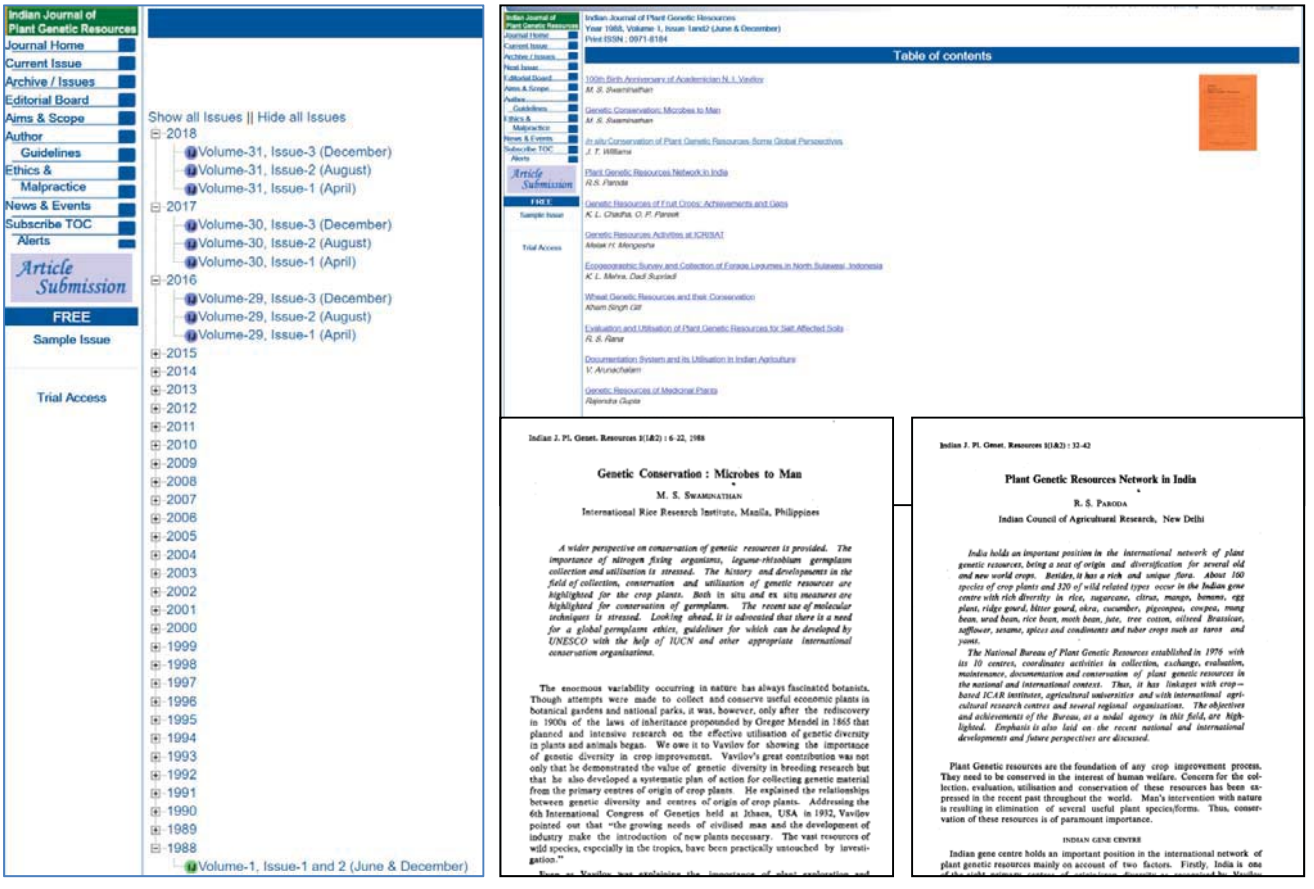

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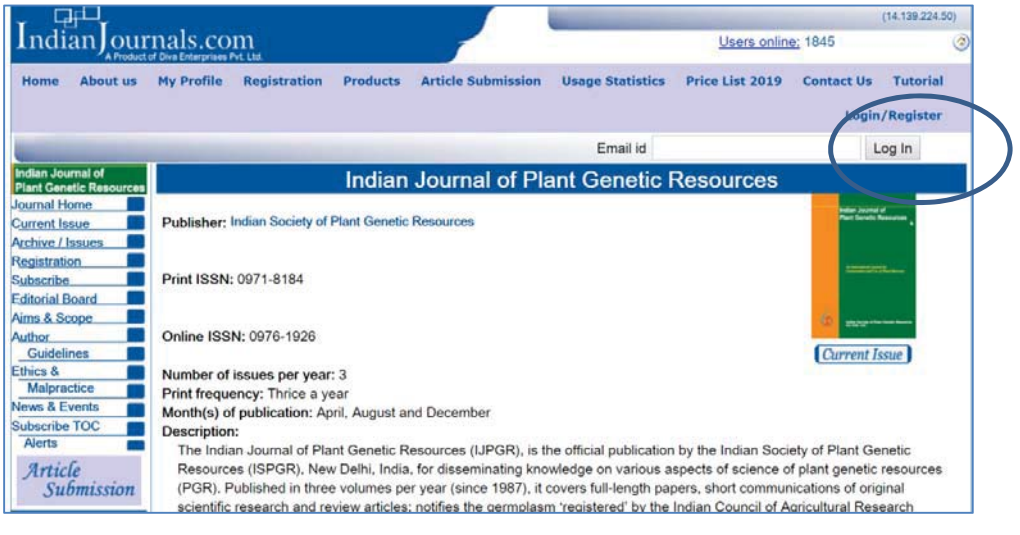

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