

Conservation and Molecular Characterization of *Morinda* sp.: an Emerging Economical bio-sorbent

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Morinda genus belongs to family Rubiaceae and is distributed throughout the tropics. *Morinda* has been derived from two Latin words *Morus* (Mulberry) and *indicus* (Indian). Eighty species of *Morinda* are known to occur in tropical regions (Mabberley, 1997). Following eight species are found in India:

- *Morinda citrifolia* L.
- *Morinda pubescens* J.E. Smith syn. *Morinda tinctoria* Roxb. syn. *Morinda tomentosa* Heyne ex Roth.
- *Morinda angustifolia* Roxb.
- *Morinda persicaefolia* Ham.
- *Morinda villosa* Hook.f.
- *Morinda umbellata*
- *Morinda trimera*
- *Morinda elliptica*

Morinda citrifolia and *Morinda tomentosa* (Fig. 1) are the two species on which molecular characterization and conservation work has been carried out at National Bureau of Plant Genetic Resources (NBPGR), New Delhi, India. Importance of these two species, its conservation and molecular characterization status is discussed here.

MORINDA TOMENTOSA HEYNE EX ROTH. SYN. MORINDA TINCTORIA ROXB.

Morinda tomentosa is a syn. of *Morinda pubescens* J.E. Smith. Another syn. is *Morinda tinctoria* Roxb. It is an evergreen underutilized fruit tree. It is also known as Aal or Nunaa. In India it is distributed in Kerala, Tamil Nadu, Gujarat, Rajasthan, Madhya Pradesh, Maharashtra. It has broad range of therapeutic and nutritional value. It is broadly used for making morindine dye for the dyeing of silk, cotton and wool. *Morinda tomentosa* is also being traditionally used by livestock owners in semiarid belt and tribal in north east and north of Gujarat, India for feeding cattles and buffaloes and improves milk yield (Rangnekar, 1991). Its fruits are edible and its wood is used for making plates, dishes and implements in India (Jukema et al., 1991). Its usage as quality wood was also reported (Jain and Singh, 1999). Another very important upcoming usage of *Morinda tinctoria* is as an environmentally safe bio-sorbent. A number of pollutants like dyes, ammonia, nitrite etc.

are being released from various sources in to water bodies and are posing health hazards and their economical removal is very much needed. Few studies pertaining to the use of *Morinda tinctoria* as bio-sorbent are cited here. Dyes draining in to water bodies from various industrial units like textile, food, paper, cosmetics and plastics etc. if untreated cause environmental pollution. There is a need to remove these toxic dyes from waste water using environmentally safe, cost effective adsorbents. There are reports available where *Morinda tinctoria* seeds have been used for removing reactive dyes (such as reactive red 198 dye) from waste water and was found to be one among the efficient adsorbents (Vijayalakshmi et al. 2013). There is another report in which leaves, stems or barks of *Morinda tinctoria* have been used for nitrite (produced due to incomplete oxidation of nitrogenous organic matter and is also used as a meat preservative in western countries) removal present in waste water (Suneetha and Ravindhranath, 2012a). Similarly ammonia pollution which is caused due to discharges from industrial, municipal and agricultural sources is a concern of environmental researchers and ashes of leaves of *Morinda tinctoria* have shown affinity towards ammonia (Suneetha and Ravindhranath, 2012b).

IMPORTANCE OF MORINDA CITRIFOLIA L.

Morinda citrifolia L. is an evergreen small fruit tree and is commonly known as Noni or Indian Mulberry. India and South East Asia is believed to be the centre of its origin (Mortan, 1992). In India Noni is distributed in Kerala, Tamil Nadu, Maharashtra, Goa, Andaman and Nicobar Islands, Lakshadweep. It is the most popular plant used for medicinal and nutritional benefits (Blanco et al, 2006). All plant parts (roots, stems, bark, leaves, flowers, and fruits) are used to prepare products ranging from health drinks to cosmetics, however its fruit is regarded as the most valuable plant part. The roots are used to produce a yellow or red dye for dyeing clothes and mats, and the fruits are eaten for health and nutrition.

Noni fruit juice has anti-microbial, anticancer, anti-inflammatory, and antioxidant effects. The fruit juice of *M. citrifolia* L. is in high demand in medicine for various illnesses, such as arthritis, diabetes, high blood pressure, muscle aches and pains, menstrual difficulties, headaches, heart disease, Acquired Immune Deficiency Syndrome (AIDS), cancer, gastric ulcers, sprains, mental depression, senility, poor digestion, atherosclerosis, blood vessel problems and drug addiction. Approximately 160 phytochemical compounds have been reported from *Morinda*. Following are the major constituents responsible for various biological effects viz. phenolic compounds like anthraquinones (such as damnacanthal, morindone, morindin), aucubin, asperuloside, coumarin (scopoletin); organic acids like caproic and caprylic acids and alkaloids like proxeronine. Other important compounds identified are vitamins (ascorbic acid and provitamin A), amino acids such as aspartic acid, and minerals.

Prof. P. I. Peter is the first person in India to show the importance of Noni and Noni based products to the world. He established World Noni Research Foundation (WNRF), Chennai and Indian Noni Cultivation Council. He is the Chairman of Noni Biotech, Chennai, India and encouraged cultivation of Noni at large scale through ‘Contract Farming’. This single crop oriented organization is basically involved in research, development and product development on Noni. Need based projects are outsourced from all over India including ICAR institutions, State Agricultural Universities, Private Universities, research organizations and the work is being done in different areas viz. Noni crop improvement and management, plant protection, Pharmacology, Clinical science and Food science.

In the United States alone, a number of patents have been registered by the US Patent and Trademark Office since 1976 (USPTO, 2005). European Union has also accepted Noni juice as a novel food (European Commission, Scientific Committee for Food, 2002). Various Noni products are available in the market in the form of juice, capsules and powders and Noni tea for health and wellness of mankind.

STATUS OF *MORINDA* SPECIES CONSERVED AT NBPGR

A total of 153 accessions of *Morinda* have been conserved at National Genebank, NBPGR. These accessions belong to five species and have been collected from different parts of India and details are given in Table 1.

MOLECULAR CHARACTERIZATION OF *MORINDA CITRIFOLIA* AND *MORINDA TOMENTOSA*

Neutral (RAPD: Random Amplified Polymorphic DNA; ISSR: Inter Simple Sequence Repeats) and gene based (SCoT: Start Codon Targeted) molecular markers were used for diversity analyses and species differentiation in *Morinda citrifolia* and *Morinda tomentosa* collected from Gujarat, Rajasthan, Madhya Pradesh, Tamil Nadu and Kerala regions of India. The representative gel profiles showing level of polymorphism in *Morinda citrifolia* and *Morinda tomentosa* based on ISSR, RAPD and SCoT markers are shown in Fig. 2. Summary of the markers used is detailed in Table 2. When all the three marker systems were compared SCoT markers showed higher level of polymorphism. Molecular marker based data can be used for designing strategies for exploration, conservation and utilization of *Morinda* to further enrich the variability and increase the utility of these species. Molecular characterization work was accomplished by funding provided by WNRF, Chennai and using the facilities available at NBPGR.

Morinda citrifolia and *Morinda tomentosa* are the two important underutilized fruit plants of immense medicinal and nutritional value. And the role of *Morinda tinctoria* as an emerging cost

effective bio-sorbent for waste water treatment is also noteworthy. Collection, conservation and molecular characterization of *Morinda* sp. will certainly aid its utilization for present and for posterity.

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Table 1 Status of *Morinda* species available at NBPGR

Name of species	Total Number of Accessions and Place of collection
<i>Morinda citrifolia</i>	117 (Kerala, Tamil Nadu, Maharastra, Goa, Andaman and Nicobar Islands, Lakshadweep)
<i>Morinda tomentosa</i>	32 (Gujarat)
<i>Morinda tinctoria</i>	2 (Andhra Pradesh, Andaman and Nicobar Islands)
<i>Morinda angustifolia</i>	1(Andhra Pradesh)
<i>Morinda trimera</i>	1 (Andaman and Nicobar Islands)

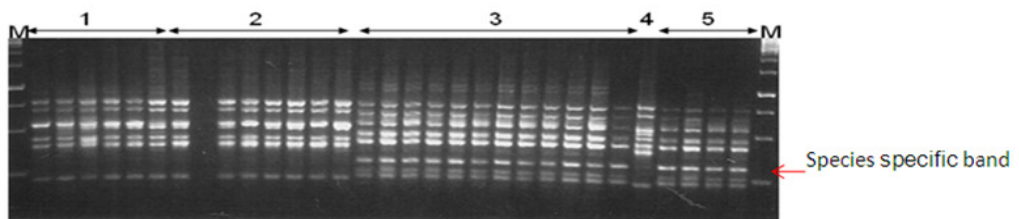
Table 2 Molecular markers used for genetic variation studies in *Morinda* sp.

Name	N	Area of collection	Markers	Genetic variation
<i>Morinda tomentosa</i>	31	Gujarat	79 RAPD	%P=92.4%; J=0.65; h=0.32
<i>Morinda tomentosa</i>	31	Gujarat	191 ISSR	%P=76.4%; J=0.61
<i>Morinda tomentosa</i>	20	Madhya Pradesh	97 RAPD	%P=57.73%; h=0.14
<i>Morinda tomentosa</i>	20	Madhya Pradesh	70 ISSR	%P=62.85%; h=0.20
<i>Morinda tomentosa</i>	20	Madhya Pradesh	131 SCoT	%P=70.23%; h=0.20
<i>Morinda tomentosa</i> <i>Morinda citrifolia</i>	25	Kerala, Gujarat, Rajasthan, Tamil Nadu	177 SCoT	%P=89.27%;J=0.51; h=0.36 (Including both the species) %P=59.2%; h=0.13 (<i>M. tomentosa</i>) %P=23.5%; h=0.04 (<i>M. citrifolia</i>)
			176 ISSR	%P=90.40%; J=0.55

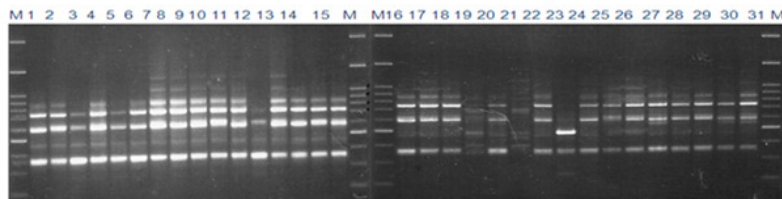
N= No. of accessions; J=Jaccard's similarity coefficient; h=Nei gene diversity; %P=% Polymorphism



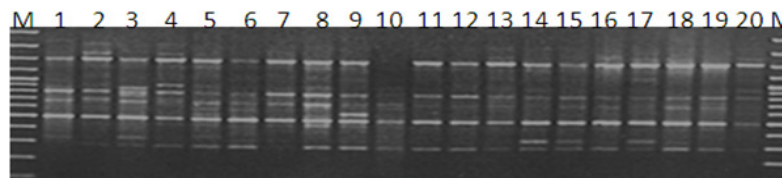
Fig 1 *Morinda tomentosa* and *Morinda citrifolia* tree, fruits and seeds



Polymorphism in *Morinda tomentosa* (1: Gujarat; 2: Rajasthan; 4: Tamil Nadu) and *Morinda citrifolia* (3: Kerala; 5: Rajasthan) with ISSR primer VHV(GT)₇; M: 1Kb molecular weight marker



Polymorphism in 31 *Morinda tomentosa* genotypes collected from Gujarat with RAPD primer OPE07



Polymorphism in 20 genotypes of *Morinda tomentosa* collected from Madhya Pradesh with SCoT 50 marker

Fig 2 Gel profiles showing level of polymorphism in *Morinda citrifolia* and *Morinda tomentosa* based on ISSR, RAPD and SCoT markers