Genetic Diversity in Azadirachta indica as Revealed by AFLP Markers

Fruiting Years	Average Fruit Weight (g)		Pulp Stone ratio		TSS (%)		Acidity (%)		Phenolics (mg/g)	
	ABA	KA	ABA	KA	ABA	KA	ABA	KA	ABA	KA
1998	28.5	12.4	18.5	10.5	14.0	12.7	0.8	1.3	2.2	4.4
1999	30.2	13.0	19.2	10.8	14.2	12.4	0.8	1.2	2.0	4.9
2000	29.6	13.2	18.8	11.2	14.0	12.2	8.0	1.3	2.1	4.6
Mean	29.4	12.8	18.5	10.8	14.0	12.4	0.8	1.3	2.1	4.6

Table 1. Physico-chemical characterstics of Alu Bokhara Amritsari (ABA) and Kala Amritsari (KA)

Table 2. Flowering, maturity and yield attributes of Alu Bokhara Amritsari (ABA) and Kala Amritsari (KA)

Fruiting	Flowerin	Date of full bloom		Yield kg/tree		Time of maturity		
Years	ABA	KA	ABA March	KA February	ABA	KA	ABA	KA
1998	24.2 - 14.3	7.2 – 24.2	5	14	3	5	28.5 - 4.6	22.5 - 27.5
1999	25.2 - 13.3	6.2 - 22.2	6	12	7	11	29.5 - 5.6	21.5 - 25.5
2000	22.2 - 15.3	9.2 - 23.2	4	12	16	18	30.5 - 7.6	23.5 - 27.5

uniform sweetness, high TSS acid ratio coupled with better shelf life, the variety deserves to be exploited in the Indian subtropics as a potential fruit for export.

References

- Bal JS, V Jit and SS Virdi (1996) Genetic diversity of plum in the Punjab plains. In Abstracts 2nd Crop Science Congress, New Delhi, 17-24 Nov., 279p.
- Chopra SK, SS Misra, VP Bhutani and A Kashyap (1986) Studies on maturity indices in relation to the length of storage in Santa Rosa plum. (*Prunus salicina* Lindl.) In: Advances in Research on Temperate Fruits, pp 319-326.
- Dhatt AS, YR Chanana, GS Nijjar, PPS Minhas, AS Dhillon and DK Uppal (1991) FLA 1-2, A new variety of plum. J. Res. PAU 28: 152.

- Dhatt AS, SS Gill and MP Singh (1982) Performance of plum (*P. salicina* Lindl.) cultivars under sub tropical conditions. *J. Res. PAU* 19: 165-167.
- Karkara BK, DD Uppal and KK Jindal (1993) Improvement of stone fruits and nuts. In: KL Chadha and OP Pareek (ed.) Advances in Horticulture Vol. 1 pp 431-44.
- Rangana S (1979) Manual of Analysis of Fruit and Vegetable Products Tata Mcgraw Hill Publicity Company Ltd., New Delhi, pp 69-73.
- Uppal DK and K Kumar (1992) Achievement and strategies of Temperature fruit breeding. Proceedings of Golden Jubilee National Seminar on Emerging Trends in Temperature Fruit Production in India, National Horticulture Board Technical Communications, pp 19-31.

Genetic Diversity in Azadirachta indica as Revealed by AFLP Markers

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Key Words: Neem, AFLP, Genetic diversity

Neem (Azadirachta indica A. Juss) is a notable multipurpose tree species that grows naturally in the Thailand, Indian subcontinent and Myanmar. Of late, Neem has gained tremendous significance worldwide because of its spectacular therapeutic and bioactive properties. Traditionally in India neem has a special cultural significance and its medicinal properties are mentioned in the ancient scriptures. The important active ingredients - azadirachtin and salanin are derived from the seeds. Because of the growing interest in this important tree species the International Neem network has established an International Neem provenance trial in 16 countries aiming at its genetic improvement (Anonymous, 1997). The information on the extent and organization of genetic diversity of the species could be useful for effective improvement program and germplasm conservation. A large number of methodologies exist for assessment of genetic diversity in the plant species. DNA based markers provide useful information regarding genetic diversity and relationship between accessions as these remain unaffected by the environmental factors. Amplified fragment length polymorphism (AFLP) has been widely used for discrimination of closely related cultivars in different crop species and also to detect genetic variability in the germplasm of a crop species. In the present study, 14 lines of neem have been subjected to AFLP analysis to detect genetic variability.

Seed of 14 lines of neem were collected from Punjab, Haryana and Rajasthan in (Table 1). These seeds were grown in the greenhouse of National Bureau of Plant Genetic Resources (NBPGR), New Delhi. Total genomic DNA was isolated from the leaves of 8-week-old seedlings using the method of Dellaporta et al. (1983). DNA concentration was quantified by fluorometric method and samples were diluted to a final concentration of 100 ng/ μ l. AFLP kit was procured from Perkin Elmer Applied Biosystems and the reactions were carried out according to the protocol provided by the manufacturer. The data obtained in the form of peaks during analysis was converted to numerical values using Genotyper (version 2.5). AFLP bands ranging in size from 50 to 500 bp were scored for presence (1) and absence (0). The pair-wise genetic similarity between the samples was estimated according to the Jaccard's coefficient.

Tree No.	Tehsil	District	State
N5	Jahajpur	Bhilwara	Rajasthan
N9	Ladpura	Kota	Rajasthan
N27	Bikaner (CSWRI)	Bikaner	Rajasthan
N36	Ganganagar	Ganganagar	Rajasthan
N37	Abohar	Firozpur	Punjab
N40	Malot	Mukatsar	Punjab
N45	Bhatinda	Bhatinda	Punjab
N48	Ludhiana (PAU)	Ludhiana	Punjab
N52	Pehwa	Kurukshetra	Haryana
N56	Kaithal	Kaithal	Haryana
N59	Barwala	Hisar	Haryana
N60	HAU Hisar	Hisar	Haryana
N62	Hisar (cantt.)	Hisar	Haryana
N66	Bawal (workshop)	Rewari	Haryana

 Table 1: Details of Neem lines used in the present study. Tree number represents number given to each line

The stastical analysis was carried using NTSYS Software (version 1.70). A dendrogram was constructed by employing UPGMA and principal co-ordinate analysis was also performed.

The value of similarity for all the 14 lines ranged from 0.012 to 0.78. The two neem samples N48 and N56, collected from Ludhiana (Punjab) and Kaithal (Haryana), respectively, displayed greater genetic similarity with a similarity coefficient of 0.78. The resultant phenogram (Fig. 1) grouped all the 14 lines broadly into three subgroups. The samples from Ladpura (Rajasthan) and HAU (Hissar) form one cluster sample from Bhatinda (Punjab) and Barwala (Haryana) forms the second cluster and rest fall in the third cluster. The sample from Hissar Cantt, N62, was more distinct and it does not form cluster with any sample. The same is supported further by the principal component analysis (Fig. 2).



Fig. 1. Phenogram generated using UPGMA cluster analysis, showing relationship between 14 lines of neem using AFLP Indian J. Plant Genet. Resour. 14: 237-239 (2001)



Fig. 2. Principal component analysis based on AFLP study of 14 neem lines with 523 amplicons

The present study revealed that there is large genetic diversity between the samples collected from different part of the western states of India, which is corroborated by studies of Singh *et al.* (1999). Future explorations can be carried out in these states for germplasm collections. To understand more about the extent and organization of genetic diversity of this species more exhaustive study by using more number of primers and marker systems is required.

References

- Anonymous (1997) Report of the workshop of the International Neem Network. Rangoon, Myanmar, 20 p.
- Dellaporta SL, J Wood and JB Hick (1983) A Plant DNA Minipreparation: Version II. *Plant Mol. Biol. Rep.* 1: 19-21.
- Singh A, MS Negi, J Rajgopal, S Bhatia, UK Tomar, PS Srivastava and M Lakshmikumaran (1999) Assessment of genetic diversity in Azadirachta indica using AFLP markers. Theo. Appl. Genet. 99: 272-279.

"Kairali" - A Mosaic Resistant Vegetable Cowpea

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Vegetable cowpea is cultivated on commercial scale in Kerala. Incidence of mosaic diseases caused by viruses is a major handicap in its cultivation and the disease is very severe in summer season and is quiet difficult to control. Cultivating resistant varieties is the most economical and practical method to tackle this disease. Immunity/resistance to mosaic disease have been reported in grain cowpea by Khatri and Singh (1974), Patel *et al.* (1982) and Doraiswamy *et al.* (1983). Cowpea mosaic virus symptoms, transmission and host range

studies were reported by Sreelekha (1987) from Kerala. However, no cultivars of vegetable cowpea grown in Kerala are found resistant to mosaic disease. Hence, a breeding programme was initiated to evolve mosaic resistant vegetable cowpea with good yield and pod characters suited to large scale cultivation in Kerala.

The technical programme consisted of collection of germplasm with special reference to mosaic resistant accessions and local cultivated types, cataloguing the germplasm for mosaic incidence, growth and yield attributes to locate resistance, sap inoculation of viruses to confirm the resistance in the resistant accessions

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