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Genetic Diversity and Population Structure analysis of Cucumis species (*Cucumis sativus* L. and *Cucumis melo*) revealed by SSR Markers

Rekha Bheemappa Chittapur* , Backchand Tembhurne, Raghavendra Gunnaiah, Prabhuling Guranna, Kisan Jadhav, Namita Raut, Satish D

INTRODUCTION & AIM

- Cucurbitacae is the diverse family distributed through out the world. The genus Cucumis contains more than 30 species, including cucumber which is an economically important crop and grown widely around the world
- *Cucumis melo* and *Cucumis sativus* are widely cultivated species
- Sativus types are commonly known as cucumbers, and *melo* types as culinary melons.
- While some of the melons(*Cucumis melo)* known as cucumbers or southekai, or yare southe in regional languages as fruits are similar to *Cucumis sativus* are being cultivated in several districts of Karnataka.
- These melons represent a diverse botanical group of cucurbits consumed as salad or as a culinary melons used for pickling, curry preparation or consumed as a raw similar to *sativus* types
- These local cultivars are tolerant to biotic and abiotic factors. Most studies are reported in commercial cultivars of sativus types, and very few are reported in diverse botanical groups of melons
- In Karnataka, botanical groups of melons have not been exploited in crop improvement programs despite having several desirable traits
- The assessment of genetic diversity and population structure of collections is a prerequisite for conservation and their utilization for future crop improvement programmes
- To study the genetic diversity and structure within and between the two species SSR markers have been used to characterize accessions and define the structure of germplasm collections

METHOD

Collection of Germplasm: The experimental material for the present investigation consisted of 150 diverse germplasm comprising *C. melo* and *C. sativus* types collected from different parts of the country. A total of 116 collections belonged to *C. melo*, the local cultivars representing different regions of Karnataka *viz*; Bagalkot, Bijapur, Badami, Hasan, and Belgaum districts. 34 were of *C. sativus* representing NBPGR collections, commercial cultivars types and two were IARI released varieties.

Diversity for fruit traits in *Cucumis melo*

- **Design**: Augmented Block design **Season** : *Kharif* 2020
- **Phenotyping**: Growth yield and fruit quality traits were recorded.
- **Genotyping by SSR markers**: 150 cucumis germplasm were fingerprinted with 47 highly polymorphic Simple Sequence Repeat (SSR) markers which were evenly distributed in the genome.
- **Statistical analysis**: Molecular diversity analysis was by software GENALEX 6.5 and Darwin. The neighbour-joining method was used to construct Tree topologies with the DARWIN software package at 10000 bootstrap values.
- **Population Structure analysis**: Statistical tool STRUCTURE 2.0, the admixture model was used with a burn in period of 150000 with 30000 Markov Chain Monte Carlo repetitions an admixture model following Hardy-Weinberg equilibrium and correlated allele frequencies as well as independent loci for each run. Ten independent runs were performed for each simulated value of K, ranging from 2 to 10.

FUTURE WORK

- These local cultivars of melons have potential applications in developing improved varieties.
- The existing diverse lines of the melon subpopulation can be utilized for exploitation of heterosis by undertaking crosses between the genetically diverse populations.
- Diversity study will be useful in evolutionary and conservation biology.

RESULTS & DISCUSSION

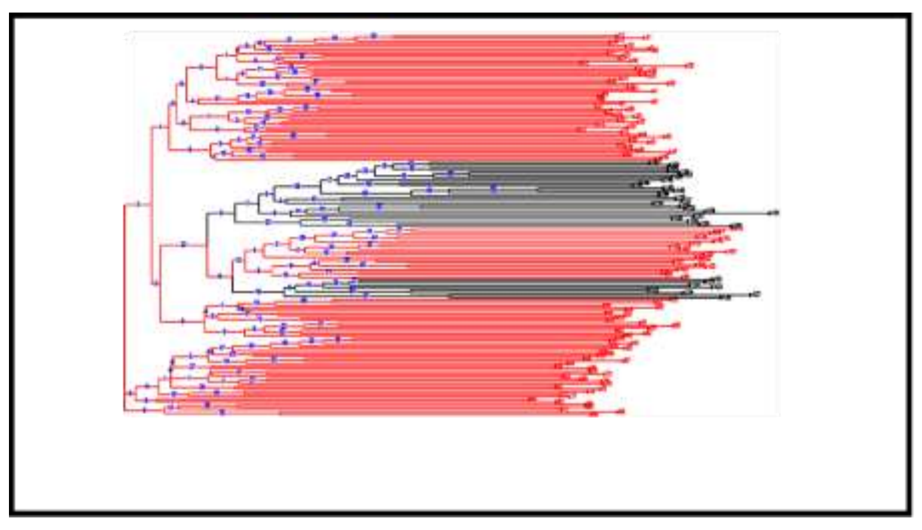

Fig 1.Unweighted neighborhood joining tree representing phylogenetic tree

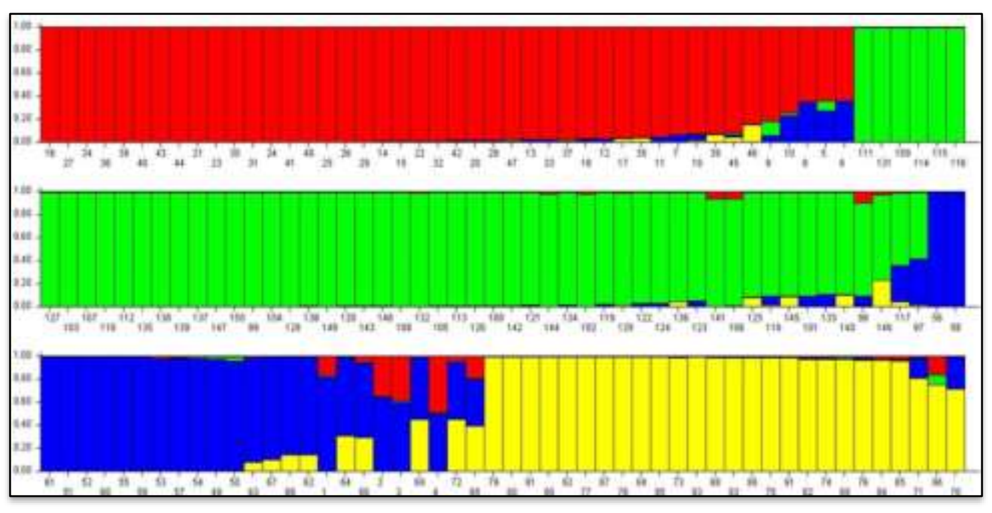

Fig 3.Q plot of Population structure of 150 cucumber genotypes (k=4) allowing admixtures using STRUCTURE 2.3.4

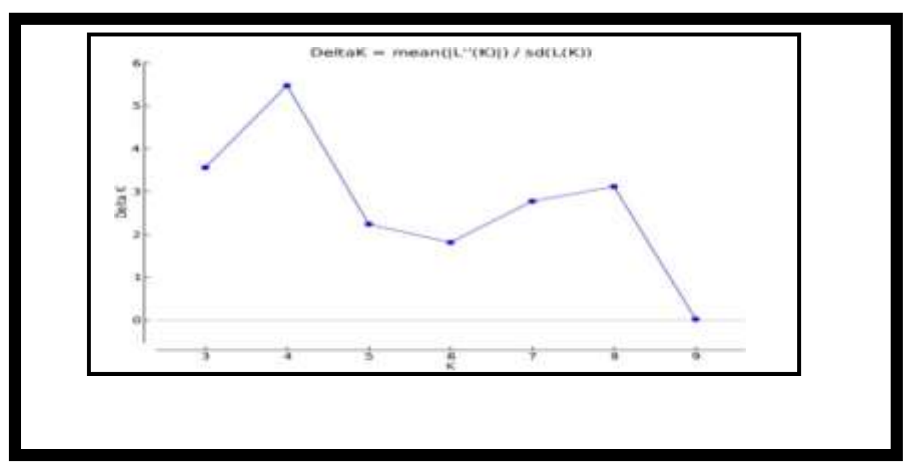

Fig 2.Delta K values for different numbers of populations assumed (K) in the STRUCURE analysis xtracted by STRUCTURE HARVESTOR

Table 1.Analysis of molecular variance (AMOVA) among and within the 4 populations

Source	df	SS	MS	Est.Var.	%
Among Populations	3	614.396	204.799	2.36	12
Among Individuals	146	4857.75	33.272	15.581	78
Within Individuals	150	316.5	2.11	2.11	10
Total	299	5788.65		20.052	100

F-Statistics	Value	P(rand >= data)
Fst	0.118	0.001
Fis	0.881	0.001
Fit	0.895	0.001

- A total of 602 alleles were identified through gel electrophoresis with a mean PIC value of 0.8.
- The Unrooted Neighbor-joining phylogenetic tree depicted clusters based on a dissimilarity matrix including germplasm of both species (Fig 1)
- Delta K value indicated presence of four populations (Fig 2) such that all *sativus* types with some melons were grouped in pop II (green colour), while all melons were grouped as three distinct populations pop 1 (Red), Pop III (Blue) and Pop IV (Yellow)(Fig 3) indicating the high amount of heterogeneity in the base population of melons while least in *sativus* types
- The AMOVA result revealed the percent of genetic variation among and within *Cucumis* species is 78 percent, among populations is 12 percent and within individuals is 10 percent(Fig 4)
- Allelic diversity is high in population II (Fig 5)
- Highest Fis value of 0.881 indicates the maximum extent of genetic similarity among the collections

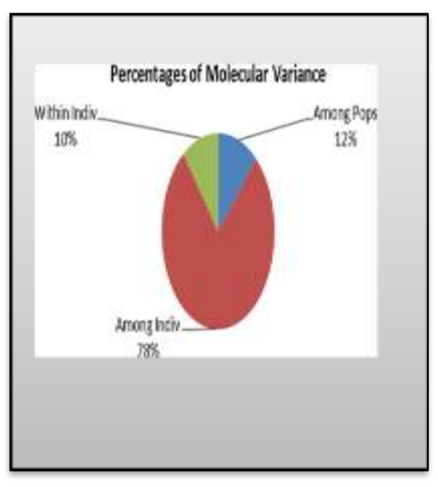

Fig 4..Analysis of molecular variance among the 4 populations of Cucumber genotypes

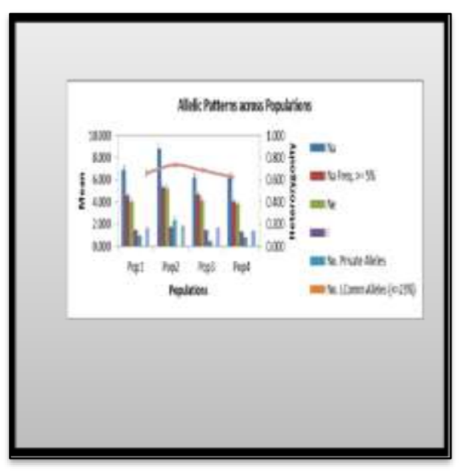

Fig 5.Distribution of markers, allelic patterns

- Fst (0.118) revealed less diversity among the populations.
- Highest Nei's Genetic identity was between pop 4 and pop 3 (0.639) indicating gene flow during evolution and maximum genetic distance was between pop 1 and pop 2 (0.734)(Table 1)

CONCLUSION

- Based on SSR data, population structure analysis revealed presence of four distinct populations, largely corresponding to species differentiation
- Eventhough both the species are different, the extent of genetic similarity I among the germplasm was high indicating the gene flow between the species during the course of evolution.
- Formation of three sub populations within *cucumis melo* confirmed the wide diversity existing within the species. While formation of single cluster of all sativus types indicated less diversity in *sativus* compared to melons. .
- Admixtures between the populations are evidence of hybridization and migration during evolution of species. The present findings of genetic structure of cucumis species revealed the existence of wide diversity in melons that could aid in exploitation for the development of improved varieties and species conservation.

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