Dissection of genetic diversity present in eggplant populations using simple sequence repeat markers

Mahammed Faizan*, B. N. Harish Babu‡, B. Fakrudin§, D. Lakshmana¶, M. Rakshith*†

*Department of Genetics and Plant Breeding, College of Horticulture, Madigere, University of Agricultural and Horticultural Sciences, Shimoga-577210, Karnataka, India; ‡Zonal Agricultural and Horticultural Research Station, Hiriyur, University of Agricultural and Horticultural Sciences, Shimoga-577210, Karnataka, India; ¶Department of Biotechnology and Crop improvement, PG centre, GKVK, Bangalore University of Horticultural Sciences, Bagalkot, Karnataka-587104, India

ABSTRACT

Eggplant (Solanum melongena L.) is the third most important solanaceous vegetable and most diversified within species spread across the world-geographical area. A study was conducted to assess the genetic diversity among the selected fifty-four eggplant genotypes (sub-categorized into five sub-population) using twenty-three SSR markers. The Analysis of Molecular Variance among the five sub-population of eggplant revealed the existence of 90.67% variation within populations and 9.34% variation among populations. The SSR markers analysis revealed important locus-wise information like mean Observed-Heterozygosity (0.216), mean Expected-Heterozygosity (0.496), Shannon’s Information Index (0.879), mean number of different alleles (3.209), mean number of effective alleles (2.535), Fixation-Index (0.649). Further, Phylogenetic-analysis clearly categorize genetically distinct individuals in which the most diversified clusters was cluster-1 (C1) out of total of five clusters and especially, wild cultivars were grouped into cluster-5 (C5). The obtained results can be used in eggplant breeding and germplasm conservation in a resourceful manner.

KEYWORDS: DNA, Genetic diversity, SSR markers, AMOVA, GenAlEx, PCA

INTRODUCTION

Eggplant (Solanum melongena L.), is one of the important solanaceous vegetables grown in tropical and subtropical regions of the world including India. It is a warm season herbaceous perennial but grown as an annual crop for commercial purposes and is popularly called as Brinjal in India. Eggplant is rich in carbohydrates, proteins, fat, dietary fibres, vitamins and minerals properties (Somawathi et al., 2014).

The assessment of genetic features is a vital process for breeders to produce a new genetic line or to improve an existing one further. India as being major diversity hub for eggplant the occurrence of variation for various traits is high. Since there is a need to evaluate and characterize the eggplant basic material which is distributed in multiple areas. Earlier, genetic evaluation was mainly based on morphological, (Faizan et al., 2021a) physiological (Faizan et al., 2021b) and biochemical traits (isozymes and chromatography) (Weijun, 1992; Isshiki & Fujieda 1994). Presently, molecular markers have massive and latent to discover genetic diversity by identifying polymorphisms. Molecular diversity analysis is one of the powerful tools for genome determination, genotype recognition, and studying the evolution pattern of crop plants and utilizing it in further breeding programmes. Several works related to molecular genetic diversity analysis in eggplant have been reported using different types of molecular markers viz., RAPD (Hu & Quiros, 1991; Tiwari et al., 2009; Ali et al., 2011), AFLP (Liao et al., 2009), SCARs (Liao et al., 2009), ISSR (Tiwari et al., 2009) and SSR (Jellan et al., 2016; Mikaela et al., 2017).

During recent eras, SSR molecular markers are becoming more popular because of their co-dominant inheritance, high abundance, enormous extent of allelic diversity, ease of assessing SSR size variation through PCR and high reproducibility (Stagel et al., 2008). The development of SSR markers derived from the SSR-enriched genomic library of eggplant was reported by Nunome et al. (2003, 2009).
The goal of the current investigation was to characterize eggplant genotypes and to study the relationship between the genotypes which fall under different subgroups using SSR molecular marker. Valuation of genetic diversity is imperative for breeding purposes, and the exploitation of molecular markers helps fast-track the evaluation progression.

**MATERIAL AND METHODS**

**Plant Material**

Around fifty-four eggplant genotypes were obtained from different sources and categorised into different sub-groups based on their type or kind viz: a) local cultivars (9 genotypes), b) commercial hybrids (2 genotypes), c) released varieties (15 genotypes), d) advanced breeding lines (24 genotypes), and e) wild relatives/related species of eggplant (4 species) (Table 1).

**Molecular Characterization**

**DNA extraction**

DNA samples from fresh fully opened leaf tissue of each genotype were extracted using the CTAB method (Doyle & Doyle, 1987). DNA quality and quantity were assessed on a 0.8 % agarose gel stained with ethidium bromide and also by using a NanoDrop® ND-1000 spectrophotometer respectively.

**SSR Analysis**

Twenty-three SSRs markers were selected to evaluate genetic diversity present among eggplant germplasm. The selection of eggplant SSRs was based on their high polymorphism information content and the quality scores reported by Nunome et al. (2009), Jellan et al. (2016) and Mikaela et al. (2017) (Table 2).

The polymerase chain reaction (PCR) mixture confined with >80 ng DNA, 5 pmol of each primer, PCR master mix Ampliqon® and nuclease free water, total PCR mixture composed of 10 µl.

PCR amplification was achieved using the Eppendorf® PCR System. The amplification conditions involved an initial step of 3 min at 94°C, followed by 35 cycles of 1 min at 94°C, 1 min at 55-69°C and 1 min at 72°C, final extension at 72°C for 10 min and final withhold temperature with 4°C. The obtained PCR products were gel electrophorized using 2.5% agarose gel and the PCR profile image was captured using a gel documentation system (Syngene Pvt. Ltd., USA).

The scoring of the PCR profile was done by using the software GeneTool (Syngene Pvt. Ltd., USA). A 100 bp ladder was used as a standard molecular weight size marker for each gel alongside the DNA samples. The bands were scored based on the weightage of DNA fragments. The analysis was repeated at least twice to confirm the reproducibility of the results and the variation for each band was observed around 5 to 10 bp.

**Table 1: List of genotypes used in the present investigation**

| Name of the genotype | Sub-group | Sources of Collection |
|----------------------|-----------|----------------------|
| PUSA UPKAR           | c         | IIVR, Varanasi,      |
| ARIKA KRANTI         | c         | Uttar Pradesh        |
| BHAGYAMATI           | c         |                      |
| PUSA ANKUR           | c         |                      |
| PUSA BINDU           | c         |                      |
| PUNJAB SADABAHAR     | c         |                      |
| ARUNA                | c         |                      |
| SHOBHNA              | c         |                      |
| SWARNA MANJARI       | c         |                      |
| CH-215               | d         |                      |
| JAWAHAR BRINJAL-8    | c         |                      |
| JAWAHAR BRINJAL-69   | c         |                      |
| R-2580               | d         | V.R.S. Kalyanpur,    |
| R-2594               | d         | Uttar Pradesh        |
| R-2591               | d         |                      |
| MALAPUR LOCAL        | d         |                      |
| L-2232               | d         |                      |
| L-3272               | d         |                      |
| R-2581               | d         |                      |
| L-2230               | d         |                      |
| L-3268               | d         |                      |
| M4                   | d         | COH, Muddigere       |
| M21                  | d         |                      |
| M23                  | d         |                      |
| M17                  | d         |                      |
| MATTIGULLA           | a         |                      |
| RAMDURGA             | a         |                      |
| MELAVANKI            | a         |                      |
| M6                   | d         |                      |
| M19                  | d         |                      |
| VERY GREEN LONG      | a         | ZRS, Chianky,        |
| I1HR-322             | d         | Palamu, Jharkhand    |
| PANT SAMRAT          | c         |                      |
| I1HR-7               | d         |                      |
| LONG GREEN           | a         |                      |
| SWARNA PRATIBHA      | c         |                      |
| SWARNA MANI          | c         |                      |
| EARLY ROUND MARKET   | a         | Local Collection     |
| RAMPUR LOCAL         | a         |                      |
| HEBBAL GULLA         | a         |                      |
| ROUND GREEN          | a         |                      |
| IC354140             | d         |                     |
| IC90785              | d         |                    |
| IC99676- LONG        | d         |                    |
| IC99676- ROUND       | d         |                    |
| IC90691              | d         |                    |
| IC354597-ROUND       | d         |                    |
| SUVARNA GP098        | b         | Suvarna Seeds PVT. LTD. |
| VIJAYA ARBH98        | b         | Vijaya Seeds PVT. LTD. |
| CO-2                 | c         | TNAU, Coimbatore, Tamil Nadu |
| Solanum macrocarpon  | e         | COH, Bangalore       |
| Solanum indicum      | e         |                      |
| Solanum torvum       | e         |                      |
| Solanum mammosum     | e         |                      |

COH- College of Horticulture, VRS- Vegetable Research Station, ZRS- Zonal Research Station, IIVR- Indian Institute of Vegetable Research, NBPGR- National Bureau of Plant Genetic Resources, TNAU- Tamil Nadu Agricultural University

**Molecular Analysis**

The molecular analysis was done using GenAlEx V.6.0 software for PCA derivation and parameter calculation of the number...
of different alleles (Na), number of effective alleles (Ne), Shannon’s Information Index (I), Observed Heterozygosity (Ho), Expected Heterozygosity (He), number of migrants (Nm), Polymorphic Information Content (PIC) value, Fixation Index (F) and F-statistics. The analysis of molecular variance was determined from Arlequin V.3.0 (Excoffier & Lischer, 2010). A Neighbour-Joining (NJ) tree (Figure 3) was constructed across the samples to analyze the genetic relationship among the individuals and populations using MEGA V.5.0 software.

RESULTS

Analysis of Molecular Variance (AMOVA)

Analysis of Molecular Variance (AMOVA) with a fixation index value of 0.649 specified significant differences among the individuals used in the present study. The per cent variation found among the sub-populations was 9.34 and within sub-populations was 90.66 per cent (Table 3).

Statistics of SSR Genetic Marker

Around twenty-three SSR markers were used for genotyping in which twenty-two were polymorphic with mean value of 0.858 ranged from 0.0 for marker SM104 to 0.968 for EM147. The number of different alleles (Na) among the SSR loci ranged between 1.00 and 7.44 (SM114) with mean of 3.209. The effective number of alleles (Ne) was high about 5.643 (SM114) with the highest Shannon’s Information Index (I) of 1.825 (SM114). The Observed Heterozygosity (Ho) ranged from 0.00 to 0.992 (SM128) and whereas the Expected Heterozygosity (He) was about 0.00 to 0.818 (SM114). The fixation index for
twenty-three SSR was ranged from -0.362 (SM128) to 1.000 (Table 4).

In the sub-population, POP4 recorded the highest number of distinct alleles (4.17) followed by POP3 (3.91) and POP1 (3.61) while the minimum number of distinct alleles was recorded in POP2 (1.826). Ne value was maximum for the sub-population, POP4 (3.02) and the POP2 chronicled minimal value of 1.78. The Expected heterozygosity (He) values for sub-populations ranged between 0.332 (POP2) and 0.577 (POP3 and POP4). Whereas, observed heterozygosity (Ho) values for sub-populations ranged between 0.185 (POP5) and 0.238 (POP3). The percentage polymorphism observed maximum for POP1 and POP3 (95.65%) and its least value was observed in the case of POP2 (60.87%) with a mean percentage polymorphism of 85.22 (Table 5).

F Co-efficient and Genetic Distance

The major components of F-statistics are $F_{IS}$, $F_{IT}$, and $F_{ST}$; where, ‘I’ represent individuals, ‘S’ represents sub-population and ‘T’ represents the total population. The $F_{IS}$ mean value across all the loci and populations was 0.684. In the present study, the mean $F_{IT}$ value observed was 0.721. However, the observed $F_{ST}$ mean value was 0.238. Among the pair-wise population $F_{ST}$ the values ranged from a minimum of 0.021 between POP1 and POP3 and a maximum of 0.348 between POP2 and POP5. The pair-wise $F_{ST}$ values indicated the presence of significant genetic differentiation among the sub-populations of eggplant working collection (Table 6). Considering the SSR markers, the $F_{ST}$ values ranged between 0.00 (SM104) and 0.473 (SM127) indicating the presence of significant genetic differentiation among the accessions of eggplant for individual SSR loci studied (Table 4). Pair-wise genetic distance across sub-populations ranged from 0.053 (POP1 and POP3) to 1.03 (POP2 and POP5) (Table 7).

Principal Coordinate Analysis

The Principal Coordinate Analysis (PCA) was done to understand the relationships between the sampled accessions based on genetic distance. The relative position of the fifty-four genotypes is illustrated in Figure 2. However, there was no distinct categorization of eggplant genotypes were observed except the genotypes of POP5 (Wild/related species of eggplant) are situated in quadrant C. Whereas, in Figure 1 it was observed that the sub-population POP5...
Pop1- Local cultivars, Pop2- Hybrids, Pop3- Released varieties, Pop4- Advanced breeding lines and Pop5- Wild/related species of eggplant

Figure 3: Neighbor joining (NJ) tree constructed using MEGA V.7.0 based on pair-wise genetic distance across five sub-populations of eggplant.

(wild/related species of eggplant) while the lower right quadrant consisted of no sub-populations. The upper left quadrant consisted of sub-population POP2 (Hybrids) and lower left quadrant entailed sub-populations POP3 (Registered varieties), POP1 (Local cultivars) and POP4 (advanced breeding lines).

Phylogenetic Analysis

The phylogenetic analysis for 54 accessions of eggplant across five sub-populations resulted in the formation of five major clusters (Figure 4). The accessions from POP4, POP3, POP1 sub-populations are dispersed across all major clusters (all 5 major clusters), possibly due to a high rate of gene flow or genetic drift. Cluster 1 included the individuals of POP1, POP2, POP3 and POP4. The second cluster consisted of individuals of POP1, POP3 and POP4 whereas, cluster 3 entailed POP1, POP3 and POP4. Cluster 4 comprised of POP1, POP3 and POP4 though cluster 5 entailed of POP3, POP4 and POP5. The individual accessions of POP5 which consisted of wild species of eggplant have formed a separate sub cluster under the major cluster 5.

DISCUSSION

Eggplant is an important Solanaceous vegetable which has luxurious genetic diversity for various traits including drought tolerance (Jellan et al., 2016). This was manifested from the results of genetic diversity analysis of 54 accessions of eggplant in the present study.

Analysis of Molecular Variance (AMOVA) with a mean fixation index value of 0.649 for eggplant indicated significant differences among the individuals, among the sub-populations and also indicates the presence of gene flow among the sub-populations. The AMOVA fallouts show a very high level of intra-population diversity and low nevertheless significant genetic differentiation in interpopulation diversity of eggplant in the present study. Similarly, Gramazio et al. (2017) also reported that, the AMOVA showed significant genetic discrepancy among forty-eight eggplant accessions in the genetic diversity analysis of eggplant.

Phylogenetic Analysis

The phylogenetic analysis for 54 accessions of eggplant across five sub-populations resulted in the formation of five major clusters (Figure 4). The accessions from POP4, POP3, POP1 sub-populations are dispersed across all major clusters (all 5 major clusters), possibly due to a high rate of gene flow or genetic drift. Cluster 1 included the individuals of POP1, POP2, POP3 and POP4. The second cluster consisted of individuals of POP1, POP3 and POP4 whereas, cluster 3 entailed POP1, POP3 and POP4. Cluster 4 comprised of POP1, POP3 and POP4 though cluster 5 entailed of POP3, POP4 and POP5. The individual accessions of POP5 which consisted of wild species of eggplant have formed a separate sub cluster under the major cluster 5.

DISCUSSION

Eggplant is an important Solanaceous vegetable which has luxurious genetic diversity for various traits including drought tolerance (Jellan et al., 2016). This was manifested from the results of genetic diversity analysis of 54 accessions of eggplant in the present study.

Analysis of Molecular Variance (AMOVA) with a mean fixation index value of 0.649 for eggplant indicated significant differences among the individuals, among the sub-populations and also indicates the presence of gene flow among the sub-populations. The AMOVA fallouts show a very high level of intra-population diversity and low nevertheless significant genetic differentiation in interpopulation diversity of eggplant in the present study. Similarly, Gramazio et al. (2017) also reported that, the AMOVA showed significant genetic discrepancy among forty-eight eggplant accessions in the genetic diversity analysis of eggplant.

The mean Nm value (1.026) indicated an overall number of migrants and possible divergence among the microsatellite loci used in the working collection of the eggplant. The maximum expected and observed heterozygosity (He) was 0.496. The mean heterozygosity values for sub-populations ranged between 0.332 (POP2) to 0.558 (POP1). Similar findings were reported by Vilanova et al. (2014) in 30 eggplant accessions using 19 SSRs.
Faizan et al.

**G1-Suvarna Gp098, G2-Pusa Upkar, G3-Vijaya Arbh98, G4-Arka Kranti, G5-Bhagyamati, G6-IIHR322, G7-Rampur Local, G8-Pusa Ankur, G9-R2580, G10-R2594, G11-R-2591, G12-Ic354140, G13-Early Round Market, G14-M4, G15-M21, G16-M23, G17-M17, G18-Pusa Bindu, G19-Mattigulla, G20-Ramdurga, G21-Malapur Local, G22-Pant Samrat, G23-Ic90785, G24-Ic99676 Long, G25-IIHR7, G26-Punjab Sadabahar, G27-L2232, G28-L3272, G29-IC99676 Round, G30-Aruna, G31-Shobha, G32-Long Green, G33-Swarna Manjari, G34-Melavanki, G35-Hebbal Gulla, G36-R-2581, G37-Swarna Pratibha, G38-Round Green, G39-L2230, G40-L3268, G41-M6, G42-M19, G43-CH215, G44-Jawahar Brinjal-8, G45-Swarna Mani, G46-IC90691, G47-Jawahar Brinjal-69, G48-Very Green Long, G49-IC354597 Round, G50-CO 2, G51-Solanum macrocarpon, G52-Solanum indicum, G53-Solanum torvum and G54-Solanum mammosum. G- Genotypes; L- Endogenous control 100bp Ladder**

**Figure 5:** SSR banding pattern of 54 accessions of eggplant obtained by different microsatellite markers.
Table 4: Locus-wise information on the number of different alleles (Na), number of effective alleles (Ne), Shannon’s Information Index (I), Observed Heterozygosity (Ho), Expected Heterozygosity (He), number of migrants (Nm), Polymorphic Information Content (PIC) value, Fixation Index (F) and F-statistics across populations of eggplant

| Marker Name | Na | Ne | I | Ho | He | F | PIC value |Nm | FIS | FST |
|------------|----|----|---|----|----|---|-----------|---|-----|-----|
| SM104      | 1.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.000 | 0.00 | 0.00 | 0.00 |
| SM107      | 3.80 | 0.97 | 3.36 | 0.68 | 1.06 | 0.31 | 0.058 | 0.149 | 1.00 | 0.00 |
| SM114      | 7.40 | 0.92 | 5.64 | 0.42 | 1.82 | 0.11 | 0.818 | 0.017 | 0.209 | 0.035 |
| SM116      | 5.00 | 0.83 | 4.18 | 0.60 | 1.43 | 0.19 | 0.728 | 0.058 | 0.353 | 0.170 |
| SM117      | 2.40 | 0.40 | 1.90 | 0.28 | 0.67 | 0.12 | 0.410 | 0.111 | 1.00 | 0.00 |
| SM119      | 2.40 | 0.51 | 1.99 | 0.37 | 0.68 | 0.21 | 0.414 | 0.120 | 1.00 | 0.00 |
| SM120      | 1.80 | 0.20 | 1.71 | 0.19 | 0.52 | 0.13 | 0.373 | 0.096 | 1.00 | 0.00 |
| SM126      | 5.40 | 1.03 | 3.65 | 0.40 | 1.43 | 0.15 | 0.712 | 0.032 | 0.340 | 0.076 |
| SM127      | 2.40 | 0.40 | 1.59 | 0.19 | 0.50 | 0.14 | 0.301 | 0.097 | 1.00 | 0.00 |
| SM128      | 4.80 | 0.49 | 3.88 | 0.328 | 1.42 | 0.097 | 0.992 | 0.008 | 0.733 | 0.028 |
| S.Em. ±   | 6.96 | 0.23 | 0.28 | 0.601 | 0.049 | 0.412 | 0.647 | 1.000 | 0.000 | 0.000 |

Table 5: Population-wise information on the number of alleles (N), number of different alleles (Na), number of effective alleles (Ne), Shannon’s Information Index (I), Observed Heterozygosity (Ho), Expected Heterozygosity (He), Fixation Index (F) and percentage polymorphism (%) observed across 23 microsatellite loci

| Populations | N | Na | Ne | I | Ho | He | F | P (%) |
|-------------|---|----|----|---|----|----|---|-------|
| Pop1        | 9.00 | 3.60 | 2.75 | 1.02 | 0.21 | 0.55 | 0.709 | 95.65 |
| S.E.m ±     | 0.00 | 0.34 | 0.26 | 0.097 | 0.086 | 0.042 | 0.115 | -     |
| Pop2        | 2.00 | 1.82 | 1.78 | 0.497 | 0.217 | 0.332 | 0.419 | 60.87 |
| S.E.m ±     | 0.00 | 0.174 | 0.162 | 0.092 | 0.088 | 0.058 | 0.171 | -     |
| Pop3        | 14.95 | 3.91 | 2.82 | 1.052 | 0.238 | 0.577 | 0.661 | 95.65 |
| S.E.m ±     | 0.043 | 0.421 | 0.267 | 0.095 | 0.087 | 0.038 | 0.119 | -     |
| Pop4        | 23.95 | 4.17 | 4.024 | 1.096 | 0.226 | 0.577 | 0.692 | 95.65 |
| S.E.m ±     | 0.043 | 0.420 | 0.312 | 0.107 | 0.087 | 0.046 | 0.111 | -     |
| Pop5        | 3.957 | 2.522 | 2.289 | 0.732 | 0.185 | 0.438 | 0.686 | 78.26 |
| S.E.m ±     | 0.043 | 0.314 | 0.275 | 0.111 | 0.076 | 0.056 | 0.109 | -     |
| GRAND Mean  | 10.774 | 3.209 | 2.535 | 0.879 | 0.216 | 0.496 | 0.649 | 85.22 |
| S.E.m ±     | 0.747 | 0.174 | 0.122 | 0.049 | 0.037 | 0.023 | 0.055 | 6.96 |

Percent polymorphism observed across the 23 SSR markers for five sub-populations represents the amount of diversity present in intra-population and inter-population. The PIC values obtained in the present study were ranged from 0.0 for marker SM104 to 0.968 for EM147, with a mean PIC value of 0.85 (Figure 5). The PIC value has shown to be influenced by the occurrence of variants per locus as well as relative distribution of the alleles (Botstein et al., 1980). The maximum range of PIC value indicates that the markers were quite informative showing high replicability and reliability, covering the entire genome, evenly distributed on chromosomes and having high potentiality for multiplexing and high throughput genotyping. Similarly, mean PIC values of 0.574 and 0.50 were obtained by Hurtado et al. (2012) and Vilanova et al. (2014) while assessing 52 and 19 accessions, respectively.

Table 7: Pair-wise genetic distance for five sub-populations of eggplant germplasm

| POP1 | POP2 | POP3 | POP4 | POP5 |
|------|------|------|------|------|
| 0.000 |      |      |      |      |
| 0.509 | 0.000 |      |      |      |
| 0.593 | 0.398 | 0.000 |      |      |
| 0.077 | 0.525 | 0.077 | 0.000 |      |
| 0.640 | 1.030 | 0.597 | 0.731 | 0.000 |

The significant mean values, FIS (0.684), FST (0.721) and FST (0.238) for five sub-populations of eggplant indicated the level of clustering across the sub-populations which, clearly revealed the existence of distinct genetic clustering at each hierarchy of the population i.e., individual, sub-population and total population. In quantitative genetics, F-statistics describe the

expansive and organisation of genetic diversity was indicated by allelic frequency (Na, Nm, Ne, He and Ho) found in eggplant (Tumblen et al., 2011; Ge et al., 2013; Vilanova et al., 2014).
statistically expected level of heterozygosity in a population; more specifically the expected degree of a reduction in heterozygosity when compared to Hardy–Weinberg expectation.

Pair-wise genetic distance across sub-populations indicated the presence of significant genetic differentiation among the populations than among the individuals within a population which may have occurred due to genetic contamination of subpopulations. The genetic distance of eggplant population was illuminated by Behera et al. (2005) and Gramazio et al. (2017) where, genetic distance between sub-populations more than pair-wise FST value indicated different populations of eggplant being more genetically diverse than individuals within a population.

In eggplant genetic diversity analysis, PCA uses an orthogonal transformation to convert a set of observations of possibly correlated variables into a set of values of linearly uncorrelated variables (principal components). The Principal Coordinate Analysis obtained in the present study separated the five sub-populations of eggplant as well as 54 individuals of five sub-populations which is possibly due to high degree of diversity between different populations than within a particular population (Figure 2) as that of Peakall and Smouse, 2012. Similar results for PCA analysis were obtained by Hurtado et al. (2012) in which, eggplant accessions of the different origins were admixed from China and Sri Lanka were mostly distributed in different areas of the plot (Hurtado et al., 2012).

Phylogenetic analysis clearly distinguishes a genetically distinct individual or a population from the rest (Yang et al., 2015). In Solanum species, analysis of the phylogeny of populations is an important step in understanding the relationships among the populations and their evolution.

Based on the results of Neighbour joining tree method, two major clusters on the basis of genetic similarity were formed for five eggplant sub-populations wherein, Cluster 1 includes POP1, POP4, POP2 and POP3. On the other point, the cluster C2 entailed of sub-population POP5 which, show clear dissimilarity between the sub-populations of two clusters. Further, the accessions from POP4, POP3, POP1 sub-populations are dispersed across all major clusters (Figure 4), possibly due to high rate of gene flow or genetic drift. The individual accessions of POP5 which consisted of wild species of eggplant have formed a separate sub cluster under the major Cluster 5. The admixture of different individuals of five sub-populations of eggplant was possibly due to drastic effect of genetic flow or genetic drift. Similarly, these types of genetically admixed clusters were obtained by Demir et al. (2010) and Jellan et al. (2016) during the phylogenetic assessment of twenty eggplant genotypes each.

The high degree of genetic variation within populations could be due to high rate of gene flow. Further, the contribution of allelic variation to the difference between populations is less compared to their contribution to the variation within population. Eggplant is modified with different style of flower that leads to crosspollination in nature which expectedly; may result in genetic contamination of eggplant accessions.

CONCLUSION

The eggplant or brinjal which is a most acceptable and commercially grown solanaceous vegetable after tomato, potato and chilli. Due to high abiotic as well as biotic stress incidence, the productivity of crop decreasing day by day. In order to enhance the yield potentiality of crop breeder needs to study the presence of genetic variation and diversity in a gene pool. There is a need of broaden the genetic base for development of adversely aclimatised eggplant hybrids for Indian seed markets. The obtained analytical results can be utilised for grouping of individual based on genetic similarities and dissimilarities. Such information aids in the selection of diverse parents for obtaining superior allele combinations in the hybrid or varietal development programmes.

Disclosure Statement

No potential conflict of interest was reported by the authors.

REFERENCES

Ali, Z., Xu, Z. L., Zhang, D. Y., He, X. L., Bahadur, S., & Yi, J. X. (2011). Molecular diversity analysis of eggplant (Solanum melongena) genetic resources. Genetics and Molecular Research, 10(2), 1141–1155. https://doi.org/10.4238/vol10-2gmnr1279
Behera, T. K., Sharma, P., Singh, B. K., Kumar, G., Kumar, R., Mohapatra, T., & Singh, N. K. (2005). Assessment of genetic diversity and species relationships in eggplant (Solanum melongena L.) using STMS markers. Scientia Horticulturae, 107(4), 352–357. https://doi.org/10.1016/j.scienta.2005.11.004
Bostein, D., White, R. L., Skolnick, M., & Davis, R. W. (1980). Construction of a genetic linkage map in man using restriction fragment length polymorphisms. American Journal of Human Genetics, 32(3), 314–331.
Demir, K., Bakir, M., Sarikamis, G., & Acunalp, S. (2010). Genetic diversity of eggplant (Solanum melongena) germplasm from Turkey assessed by SSR and RAPD markers. Genetics and Molecular Research, 9(3), 1568–1576. https://doi.org/10.4238/vol9-3gmnr878
Excoffier, L., & Lischer, H. E. (2010). Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. Molecular Ecology Resources, 10(3), 564-567. https://doi.org/10.1111/j.1755-0998.2010.02847.x
Faizan, M., Harish Babu, B. N., Lakshmana, D., Ganapathi, M., & Rakshit, M. (2021a). Investigation on response of growth and yield characters of eggplant over moistures stress and dissection of genetic parameters. International Journal of Chemical Studies, 9(6), 08-14.
Faizan, M., Harish Babu, B. N., Lakshmana, D., Ganapathi, M., & Rakshit, M. (2021b). Physiological and root growth response of eggplant genotypes upon drought stress and assessment of genetic parameters at different developmental stage. International Journal of Ecology and Environmental Sciences, 3(4), 22-33. https://doi.org/10.5281/zenodo.5588965
Ge, H., Liu, Y., Jiang, M., Zhang, J., Han, H., & Chen, H. (2013). Analysis of genetic diversity and structure of eggplant populations (Solanum melongena L.) in China using simple sequence repeat markers. Scientia Horticulturae, 162, 71–75. https://doi.org/10.1016/j.scienta.2013.08.004
Gramazio, P., Prohens, J., Borràs, D., Plazas, M., Herrera, F. J., & Vilanova, S. (2017). Comparison of transcriptome-derived simple sequence repeat (SSR) and single nucleotide polymorphism (SNP) markers for genetic fingerprinting, diversity evaluation, and establishment of relationships in eggplants. Euphytica, 212, 264. https://doi.org/10.1007/s10681-017-2057-3
Hu, J., & Quiros, C. F. (1991). Identification of broccoli and cauliflower cultivars with RAPD markers. Plant Cell Reports, 10, 505-511. https://doi.org/10.1007/BF00234583
Hurtado, M., Vilanova, S., Plazas, M., Gramazio, P., Fonseka, H. H., Fonseka,
Faizan et al.  

R., & Prohen J. (2012). Diversity and relationships of eggplants from three geographically distant secondary centres of diversity. *PLoS ONE*, 7(7), 1-15. https://doi.org/10.1371/journal.pone.0041748

Isshiki, S. O., & Fujieda, K. (1994). Phylogeny of eggplant and related *Solanum* species constructed by allozyme variation. *Scientia Horticulturae*, 59(3-4), 171-176. https://doi.org/10.1016/0304-4238(94)90010-8

Jellan, R. R., Saracanlao, A. O., Canama, S. J. B., Manaday, R. G. M., & Evelyn, F. D. (2016). SSR-based genetic relationship in eggplant (*Solanum melongena* L) genotypes with varying morphological response to drought. *Philippine Journal of Crop Science*, 41(3), 57-64.

Liao, Y., Sun, B., Sun, G., Liu, H., Li, Z., Li, Z., Wang, G. & Chen, R. (2009). AFLP and SCAR markers associated with peel color in eggplant (*Solanum melongena*). *Agricultural Sciences in China*, 8(12), 1466-1474. https://doi.org/10.1016/S1671-2927(08)60360-0

Mikaela, A. B., Ocampo, E. T. M., Canama, A. O., & Delfin, E. F. (2017). Hybridity testing of eggplant F1 progenies derived from parents with varying response to drought using SSR markers. *Philippine Journal of Science*, 146(3), 277-286.

Miyatake, K., Shinmura, Y., Matsunaga, H., Fukuoka, H., & Saito, T. (2019). Construction of a core collection of eggplant (*Solanum melongena* L.) based on genome-wide SNP and SSR genotypes. *Breeding Science*, 69(3), 498-502. https://doi.org/10.1270/jsbsb.18202

Nunome, T., Negoro, S., Kono, I., Kanamori, H., Miyatake, K., Yamaguchi, H., Ohyama, A., & Fukuoka, H. (2009). Development of SSR markers derived from SSR-enriched genomic library of eggplant (*Solanum melongena* L.). *Theoretical and Applied Genetics*, 119, 1143-1153. https://doi.org/10.1007/s00122-009-1116-0

Nunome, T., Suwabe, K., Iketani, H., Hirai, M., & Wricke, G. (2003). Identification and characterization of microsatellites in eggplant. *Plant Breeding*, 122(3), 256-262. https://doi.org/10.1046/j.1439-0523.2003.00916.x

Peakall, R., & Smouse P. E. (2012). GenAlEx 6.5: genetic analysis in Excel. Population genetic software for teaching and research: an update. *Bioinformatics*, 28(19), 2537-2539. https://doi.org/10.1093/bioinformatics/bts460

Somawathi, K. M., Rizliya, V., Wijesinghe, D. G. N. G., & Madhujith, W. M. T. (2014). Antioxidant Activity and Total Phenolic Content of Different Skin Coloured Brinjal (*Solanum melongena*). *Tropical Agricultural Research*, 26(1), 152-161. http://doi.org/10.4038/tar.v26i1.8060

Stagel, A., Portis, E., Toppino, L., Rotino, G. L., & Lanteri, S. (2008). Gene-based microsatellite development for mapping and phylogeny studies in eggplant. *BMC Genomics*, 9, 357. https://doi.org/10.1186/1471-2164-9-357

Tiwari, S. K., Karhilaoo, J. L., Harneed, N., & Gaikwad, A. B. (2009). Molecular characterization of brinjal (*Solanum melongena* L.) cultivars using RAPD and ISSR Markers. *Journal of Plant Biochemistry and Biotechnology*, 18, 189-195. https://doi.org/10.1007/BF03263318

Tumbilcan, Y., Frary, A., Daunay, M. C., & Doğanlar S. (2011). Application of EST-SSRs to examine genetic diversity in eggplant and its close relatives. *Turkish Journal of Biology*, 35(2), 125-136. https://doi.org/10.3906/biy-0906-57

Vilanova, S., Hurtado, M., Cardona, A., Plazas, M., Gramazio, P., Herrera, F. J., Andújar, I., & Prohens, J. (2014). Genetic diversity and relationships in local varieties of eggplant from different cultivar groups as assessed by genomic SSR markers. *Notulae botanicae Horti Agrobotanici Cluj-Napoca*, 42(1), 59-65. https://doi.org/10.15835/nbha4219414

Weijun. (1992). Inheritance of isozymes and morphological characters in the brinjal eggplant. *Acta Genetica Sinica*, 19, 423-429.

Yang, L., Wen, C., Zhao, H., Liu, Q., Yang, J., Liu, L., & Wang, Y. (2015). Development of polymorphicgenic SSR markers by transcriptome sequencing in the welsh onion (*Allium fistulosum* L.). *Applied Sciences*, 5(4), 1050-1063. https://doi.org/10.3390/app5041060