Assessment of genetic divergence using Mahalanobis $D^2$ and principle component analysis in strawberry (Fragaria x ananassa Duch.)

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Abstract

Studies were carried out to assess the genetic divergence among 30 genotypes of strawberry using Mahalanobis $D^2$ and principle component analysis of 19 quantitative characters. Thirty genotypes of strawberry were grouped into six clusters. Maximum (9) genotypes were included in cluster I and minimum (2) in cluster VI. Maximum inter cluster distance (380.23) was observed between cluster I and VI and minimum (205.34) between the cluster V and cluster VI. Principle component analysis showed more than 85.59 per cent of variability for quantitative character in different genotypes. The genotypes of cluster II were observed with days to 50% flowering, days to first harvest, number of fruit per plant, fruit weight, fruit yield per plant, fruit diameter and genotypes of cluster VI for highest mean for petiole length, number of runners per plant, days to fruit set from flowering, pH of juice and ascorbic acid. Ascorbic acid, fruit length, fruit weight and plant height were observed significant variable components and Missionarry, Lucunde, Wild Local (IC-319130) and Chandler were found with maximum value corresponding to these four variables. Selecting genotypes from divergent clusters and utilizing them in hybridization programme is likely to produce desirable recombinants, and may lead to improvement in strawberry for yield and its contributing traits.

Keywords: Strawberry (Fragaria x ananassa), genetic divergence, quantitative characters, principle component analysis

Introduction

The modern cultivated strawberry (Fragaria x ananassa Duchesne) is one of the most delicious, refreshing and nutritious among soft fruits of the world. It is amongst the few fruits crop which gives quicker and very high returns per unit area on the capital investment. Fragaria species belongs to family Rosaceae with a basic chromosome number of X=7, the cultivated strawberry Fragaria x ananassa Duch. has chromosome number (2n) of 56. It is a monoecious octoploid hybrid of two largely dioecious octoploid species Fragaria chiloensis Duch. and Fragaria virginiana Duch. had taken place spontaneously in Europe (Larson, 1994) [6]. In seventeenth century when female plants of Fragaria chiloensis Duch. of Chilean origin were grown in proximity to male Fragaria plants of North American origin (Galletta et. al. 1990) [4]. Since that time, extensive hybridization between parent species and their descendants have occurred, making Fragaria x ananassa Duch. a highly variable and heterozygous species, which was first developed in France in seventeenth century. Strawberry is a herbaceous perennial and short day plant requires different culture than tree fruits. It has short stem known as crown. The crown produces leaves at very close interval along stem axis and flowers at terminal position on stem axis. The edible portion of strawberry includes the ripened receptacle and achenes (true fruits and seed). Commercial strawberry cultivation in the USA has been started in the beginning of the 18th century and within 25 years it gained significant momentum. Most of the European countries had started growing strawberries commercially since 18th century and today nearly 60% of the world’s production is only produced from Europe. In India, Strawberries were first introduced by the NBPGR Regional Research Station, Shimla (Himachal Pradesh) in the early sixties (Sharma and Singh, 1999; Sharma, 2000) [9, 8]. Due to the constant efforts of strawberry breeders, the world wide interest for strawberry cultivation has boosted its production tremendously, which has resulted in widespread popularity of strawberry in the last Six decades.
Multivariate analysis such as D² cluster and principle component analysis have been proved to be useful tools in selecting genotypes for improvement. Mahalanobis D² analysis has been successfully used in measuring the variability. Principle component analysis is useful device for representing a set of variable by a much smaller set of composite variable that account for much of the variance among the set of original variable. It allows visualization of the differences among the individuals, identification of possible groups and relationships among individual and variables. An understanding of nature and magnitude of variability among the strawberry genotypes is a pre-requisite for its improvement. Precise information on the nature and degree of variability helps the plant breeder in choosing the diverse parents for purposeful hybridization. The success of any breeding programme in general and improvement of specific trait through selection in particular, totally depends upon the genetic variability present in the available germplasm of a particular crop (Parmar et al. 2013) [10]. The progress of breeding is, however, conditioned by the magnitude, nature and interrelationship of genotypic and non-genotypic variation. Among the quantitative characters, yield is a complex character, which is dependent on a number of yield contributing characters (Savitha, 2008) [7]. Therefore, the present study was carried out the thirty genotypes of strawberry to understand the genetic diversity its genetic improvement by D² and principal component analysis.

Materials and Methods

The experiment was carried out at the Horticulture Research Farm, Department of Horticulture, Allahabad School of Agriculture, Sam Higginbottom Institute of Agriculture, Technology and Sciences, Allahabad, in the year in 2015-2016. The experiment was conducted in Randomized Block Design having thirty genotypes in three replications. The thirty genotypes collected from NBPGR, Regional Station, Bhowali, Nainital, Uttarakhand. The allocation of treatments of the individual plots using random number in each replications with spacing 45 x 45 cm plant to plant and row to row respectively. Five plants from each replication were taken for recording observation on 19 traits viz. Plant height, Plant spread, Number of leaves per plant, Petiole length, Number of runners per plant, Leaf Area Index, Days taken to first flowering, Days taken to 50% flowering, Days to fruit set from flowering, Days to first harvest, Number of fruits per plant, Fruit weight, Fruit yield per plant, Fruit diameter, Fruit length, Total Soluble Solids, pH of juice, Acidity, Ascorbic acid.

Results and Discussion

Data on the variability parameters is presented in (Table 1) and the variability of each trait was expressed by standard deviation and the coefficient of variation. The lowest values of standard deviation were recorded in acidity and TSS while it was highest for fruit yield per plant. The coefficient of variation was lowest for ascorbic acid followed by leaf area index while highest coefficient of variation was found for fruit yield per plant.

On the basis of D² analysis 30 genotypes were grouped into six clusters (presented in Table 2). Maximum numbers of genotypes 9 were grouped into cluster I (Wild Local IC-319130, Sheet Master, Sweet Heart (C1), Sweet Heart, Missionary, Mastodan, CH-iii-23, Selva, CH-ii-14) followed by cluster III having 6 genotypes (Lesson, Swiss-2, Jucunda, Julikot Local, North West, Mecharanches), cluster V having 5 genotypes (Haward-17, Dil Pasand, N. R. Round Head, Wild Local IC-319133, Wild IC-319128), cluster II having 4 genotypes (Lucunde, CH-10, Tioga, Cavaliar), cluster IV having 4 genotypes (Swiss, Addie, CH-iii-40, Belruby), cluster VI having 2 genotypes (Chandler, Red Ground).

Table 1: variability of quantitative traits in 30 strawberry genotypes

| Variables                  | Minimum | Maximum | Mean | Standard deviation | CV% |
|----------------------------|---------|---------|------|--------------------|-----|
| Plant height (cm)          | 10.08   | 27.5    | 17.74| 0.46               | 4.45|
| Plant spread (cm)          | 17.38   | 44.89   | 34.73| 0.34               | 1.69|
| No. of leaves per plant    | 18      | 83.75   | 38.65| 0.71               | 3.18|
| Petiole length (cm)        | 7.25    | 20.25   | 13.5 | 0.29               | 3.77|
| No. of runners per plant   | 1.57    | 114.17  | 25.53| 0.8                | 5.42|
| Leaf Area Index (cm²)      | 54.44   | 81.35   | 66.03| 0.27               | 0.7 |
| Days taken to first flowering | 31.38 | 83.37   | 52.2 | 0.54               | 1.79|
| Days to 50% flowering      | 42.72   | 90.63   | 69.47| 0.54               | 3.55|
| Days to first set from flowering | 3.11 | 7.19    | 4.35 | 0.13               | 3.2 |
| Days to first Harvest      | 47.18   | 100.21  | 71.42| 0.83               | 2.02|
| No. of fruits per plant    | 8.09    | 27.68   | 16.12| 0.26               | 2.83|
| Fruit weight (g)           | 1.83    | 23.52   | 8.69 | 0.34               | 4.72|
| Fruit yield per plant (g)  | 28.11   | 580.58  | 156.53| 7.8                | 6.83|
| Fruit diameter (mm)        | 10.82   | 45.76   | 24.28| 0.65               | 4.64|
| Fruit Length (mm)          | 10.35   | 50.8    | 35.48| 0.84               | 4.12|
| TSS (%Brix)                | 6.1     | 12.32   | 9.13 | 0.1                | 1.81|
| pH of juice (pH)           | 3.28    | 5.92    | 4.39 | 0.09               | 3.75|
| Acidity (%)                | 0.11    | 0.19    | 0.16 | 0.01               | 0.64|
| Ascorbic Acid (mg/100g of pulp) | 48.45 | 50.01   | 49.36| 0.09               | 0.32|

Table 2: Thirty Genotypes of Strawberry grouped into 6 clusters

| Clusters | No. of genotypes included | Genotypes                                |
|----------|---------------------------|------------------------------------------|
| I        | 9                         | Wild Local (IC-319130), Sheet Master, Sweet Heart (C1), Sweet Heart, Missionary, Mastodan, CH-iii-23, Selva, CH-ii-14 |
| II       | 4                         | Lucunde, CH-10, Tioga, Cavaliar          |
| III      | 6                         | Lesson, Swiss-2, Jucunda, Julikot Local, North West, Mecharanches |
| IV       | 4                         | Swiss, Addie, CH-iii-40, Belruby         |
| V        | 5                         | Haward-17, Dil Pasand, N.R. Round Head, Wild Local (IC-319133), Wild (IC-319128) |
| VI       | 2                         | Chandler, Red Ground                     |
Maximum inter cluster distance was observed between cluster I and cluster VI (380.23) (Table 3), which was followed by cluster IV and cluster VI (379.64), cluster III and cluster VI (266.50), cluster II and cluster VI (227.25), cluster V and cluster VI (205.54), cluster I and cluster V (124.68), cluster II and cluster V (108.73). The highest intra cluster distance was recorded for cluster V (41.06), followed by cluster IV (40.04), cluster II (30.99), whereas cluster III (29.15), cluster I (28.08) showed minimum intra cluster distance.

Mean performance of individual cluster for different characters are presented in (Table 4) which showed that cluster II is most suitable to select better genotypes for earliest days to 50% flowering (49.535 days), earliest days to first harvest (59.360 days), number of fruits per plant (21.797), fruit weight (12.372 g), fruit yield per plant (260.847 g), fruit diameter (27.310 mm), whereas cluster VI is suitable for petiole length (15.50 cm), number of runners per plant (114.167), earliest days to fruit set from flowering (3.403 days), pH of juice (4.713) and ascorbic acid (49.490 mg). Moreover, cluster I was recorded for leaf area index (66.740), earliest days to first flowering (39.219 days), fruit length (39.418 mm), TSS (9.448 °Brix). Plant height (21.391 cm), plant spread (39.853 cm) and acidity (0.170%) were recorded in cluster IV.

Table 3: Mahalanobis average intra and inter-cluster distance (D²) of 30 strawberry genotypes

| Cluster | I     | II    | III   | IV    | V     | VI    |
|---------|-------|-------|-------|-------|-------|-------|
|         | 28.08 | 30.99 | 29.15 | 40.04 | 41.06 | 0.00  |

Table 4: Cluster mean for nineteen yield and fruit contributing characters in Strawberry genotypes

| Cluster | Plant Height (cm) | Petiole Length (cm) | Days to Flowering | Days to First Harvest | No. of Fruit Per Plant | Fruit Yield (g) | Fruit Diameter (mm) | TSS (g/l) | pH of Juice | Acidity | Ascorbic Acid (mg/100 ml) |
|---------|------------------|--------------------|------------------|----------------------|-------------------------|-----------------|-------------------|-----------|-------------|---------|--------------------------|
| I       | 18.843           | 14.609             | 39.219           | 9.323                | 34.728                  | 174.320         | 26.468            | 39.418    | 9.448       | 0.162   | 49.485                   |
| II      | 16.792           | 11.200             | 41.513           | 9.360                | 49.535                  | 177.872         | 26.047            | 35.227    | 9.230       | 0.147   | 49.148                   |
| III     | 15.608           | 9.781              | 51.315           | 8.07                 | 49.490                  | 185.748         | 29.079            | 30.655    | 8.503       | 0.163   | 49.231                   |
| IV      | 21.391           | 31.499             | 8.295            | 108.72               | 49.254                  | 149.034         | 25.191            | 30.522    | 8.923       | 0.170   | 49.219                   |
| V       | 12.667           | 31.499             | 124.68           | 169.17               | 49.311                  | 219.317         | 35.240            | 3.796     | 0.142       | 49.388 |
| VI      | 17.917           | 150.00             | 77.715           | 154.87               | 49.445                  | 237.107         | 30.522            | 0.181     | 0.135       | 49.490 |
| Mean    | 17.737           | 13.499             | 69.413           | 71.420               | 49.351                  | 237.107         | 30.522            | 0.181     | 0.135       | 49.490 |

Table 5: Principal components analysis traits of common strawberry collection

| Traits | PCA 1 | PCA 2 | PCA 3 | PCA 4 | PCA 5 |
|--------|-------|-------|-------|-------|-------|
| Plant Height | 0.198 | 0.156 | 0.149 | 0.167 | 0.186 |
| Plant Spread | 0.293 | 0.217 | 0.262 | 0.128 | 0.354 |
| No. of leaves Per Plant | 0.215 | 0.212 | 0.122 | 0.242 | 0.351 |
| Petiole length | -0.279 | -0.248 | -0.034 | -0.057 | 0.264 |
| No. of Runners Per plant | -0.204 | -0.254 | 0.172 | 0.447 | 0.071 |
| Leaf Area Index | 0.179 | 0.303 | 0.115 | 0.099 | 0.073 |
| Days Taken to First Flowering | -0.262 | 0.312 | 0.115 | 0.099 | 0.073 |
| Days Taken to 50% Flowering | -0.152 | 0.391 | 0.095 | 0.082 | 0.288 |
| Days to Fruit Set From Flowering | -0.215 | 0.206 | 0.350 | 0.047 | 0.160 |
| Days to First Harvest | -0.187 | 0.255 | 0.371 | 0.071 | 0.359 |
| No. of Fruits Per Plant | 0.205 | -0.199 | 0.315 | 0.097 | 0.080 |
| Fruit Weight | 0.115 | 0.035 | 0.558 | 0.097 | 0.059 |
| Fruit Yield Per Plant | 0.249 | -0.019 | -0.216 | 0.343 | 0.040 |
| Fruit Diameter | 0.286 | -0.197 | 0.069 | -0.417 | 0.147 |
| Fruit Length | 0.137 | 0.411 | -0.038 | -0.206 | 0.078 |
| pH of Juice | 0.277 | -0.256 | 0.075 | 0.109 | -0.118 |
| Acidity | 0.228 | 0.132 | 0.383 | -0.200 | -0.059 |
| Ascorbic Acid | 0.327 | 0.098 | -0.138 | 0.050 | 0.092 |
| Eigene Value (Root) | 8.07 | 3.60 | 2.39 | 1.27 | 0.93 |
| % Var. Exp. | 42.47 | 18.94 | 12.57 | 6.70 | 4.90 |
| Cum. Var. Exp. | 42.47 | 61.41 | 73.98 | 80.68 | 85.59 |

To assess overall diversity patterns among the genotypes, the principle component analysis were conducted (table 5), Principle component analysis revealed that five components PCA 1, PCA 2, PCA 3, PCA 4, and PCA 5 with Eigen values 8.07, 3.60, 2.39, 1.27 and 0.93 respectively, have accounted for 85.59% of the total variation. The first two principle component analysis PCA 1 and PCA 2 with a proportion of 42.47% and 18.94%, respectively contributed more total variation. Characters with the largest absolute values closer to unity with in the first principle component influence the clustering more than those with lower absolute values closer to zero. Therefore, in this study, differentiation of the genotypes into different cluster was because of a cumulative effect of a number of characters rather than the contribution of few characters. Characters having relatively higher value in the first principle component (PCA 1) were ascorbic acid, pH of juice, TSS, days to fruit set, petiole length, leaf area index, leaf weight, plant height, fruit diameter, number of runners per plant, fruit yield per plant.
plant spread, fruit diameter, petiole length, pH of juice, fruit yield per plant had more contribution to total diversity and they were responsible for the differentiation of the five clusters. The second principle component (PCA 2), which accounted 18.94% of the total variation contributed from fruit length, days to first harvest, days taken to 50% flowering, days taken to first flowering and leaf area index. Characters like fruit weight, number of fruit per plant and acidity were the characters contributed to the third principal component (PCA 3). Similarly plant height, number of runners per plant and number of leaves per plant were the characters contributed to the fourth principal component (PCA 4). Leaf area index and plant height were contributed to fifth principal component (PCA 5).

Higher standard deviation and coefficient variation for yield show high degree of diversity among the evaluated genotypes. The higher inter-cluster distance than average intra-cluster distance, confirms wide genetic diversity among the genotypes of different groups than those the same cluster. This finding was similar with the findings of Uddin and Mitra (1994) \[11\]. They obtained higher inert cluster distance in multivariate in the sesame. The maximum inter cluster distance between cluster I and cluster VI, and cluster IV suggested that if they were chosen for hybridization program to improve the vigour and yield (Table 3), may give broad spectrum of variability in segregating generations. This result supports the findings of Ahmed et al. (2002) \[1\]. Divergence were given greater emphasis for deciding the type of cluster for the purpose of further selection and the choice of parents for hybridization (Jaadeb and Samal, 1991) \[5\]. Higher inter and intra-cluster distances indicate higher genetic variability among accessions between and cluster respectively. The minimum inter and intra-cluster distances indicate the close genetic relationship among the accessions of two clusters and within the clusters. Accessions among the cluster separated by high D\(^2\) value could be used in hybridization for the obtaining wide spectrum of variations among the segregates (Chahal and Gosal, 2002) \[3\].

It is revealed that crosses should be made between accessions belonging to the distance clusters for high heterotic response. The relative discriminating power of PCA was revealed by the Eigen values which was high in PCA 1 (8.07) and lower in PCA 5 (0.93) (Table 5). From this study, a combination of PCA and genetic divergence will produce better results when considering genetic variability among the strawberry. This agrees with the finding of Aremu et al. (2007) \[2\]. However, these analysis are very useful for the collection, management and use in future breeding programs.

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