RESEARCH ARTICLE

Genetic Divergence Studies in Horse gram (Macrotyloma uniflorum Lam.Verdc) for Quantitative Traits using Mahalanobis D² Analysis

Geetha K* and Divya S
Regional Research Station, Paiyur, Tamil Nadu Agricultural University, Krishnagiri -635 112, Tamil Nadu, India

ABSTRACT

The present study aimed to estimate the nature and magnitude of genetic divergence in 60 horse gram genotypes for ten characters using Mahalanobis D² analysis. The genotypes were grouped into thirteen clusters. Concerning intracluster distance, the genotypes falling within cluster III (31.51) has maximum divergence. Maximum inter-cluster distance was observed between cluster VIII and XIII (184.05), followed by cluster XI and XIII (170.83). Cluster VIII exhibited a higher mean for hundred seed weight, grain yield per plot and Bhusha yld/plot and cluster VI contained genotypes with the higher mean value for pod length and the number of seeds per pod. Among the yield contributing characters, the traits viz., grain yield per plot (28.30%) followed by the number of pods per plant (26.21 %) and the number of branches per plant (18.36%) contributes to major genetic divergence. The results indicated that from cluster IV the genotype HA 871-5-67/2 was selected for plant height, from cluster II the genotype Morappur-1 was chosen for the number of branches per plant, from cluster XII the accession HG 102 was selected for the trait number of pods per plant, from cluster VI the genotype T 45 was chosen for pod length and the number of seeds per pod. From cluster VIII the genotypes 12EB and IC 9606 were nominated for hundred seed weight, grain yield per plot and Bhusa yield per plot.

Keywords: Horse gram; D² analysis; Cluster

INTRODUCTION

Horse gram [Macrotyloma uniflorum (Lam.) Verdc.] generally known as poor man’s pulse crop belongs to the family fabaceae, widely known for its hardiness, adaptable to adverse climatic conditions even unsuitable for other crops. The bhusa of horse gram after removal of pod is used as feed for horses; hence it obtained its name as horse gram. The crop is dispersed all over the tropics and reports have mentioned that the crop has been cultivated in the regions of Sudan, Ethiopia, Zaire, Kenya, Tanzania, South Africa, and Angola. According to Vavilov, India is the primary center of origin for horse gram because the horse gram has been cultivated in peninsular India since pre-historic times. The crop is native to India and distributed all over the country from Uttarkhand in North to Tamil Nadu in south and Gujarat in the west to Bengal in East. It is majorly grown in the states of Karnataka, Andhra Pradesh, Tamil Nadu, Madhya Pradesh, and Maharashtra. The total area under horse gram in India is 4.58 lakh hectares and the production is about 2.97 lakh tonnes with the productivity of 648 kg/ha. In Tamil Nadu, it is grown in an area of 0.79 lakh hectares and with annual grain production of about 0.61 lakh tonnes with a productivity of 776 kg per hectare (http://www.indiastat.com). Horse gram is a cleistogamous self-fertile crop with chromosome number of 2n = 20,22, 24. Horse gram is an annual herb with semi-erect growth habitat growing from 60 to 120 cm height. Leaves are trifoliate with the length of 2 to 5 cm. Ovate lanceolate stipules and short, bisexual, bracteate, pedicellate, zygomorphic, and complete peduncles are found in horse gram. The flowers of horse gram have downy teeth, lanceolate calyx, light yellow corolla, five petals, diadelphous (9+1) stamens with alternately arranged short filaments, and have long anthers. The gynoecium is with superior ovary. Style file from the terminal, curved, stigma capitate, and hairy. Pods are linear with nearly 5-7 seeds. Seeds are black, light red, brown in colour of 3 to 6 mm long and skinny testa with small hilum are present. Horse gram is a short day and day-neutral crop that requires nearly 120-180 days to mature from the date of sowing. It has a rich source of protein,
minerals, and vitamins. On comparing the nutrient content of horse gram with pigeon pea and chickpea, horse gram is having nearly 23 % protein and higher lysine content. Besides these benefits, horse gram is also cultivated as a green manure crop to enrich soil fertility through fixing the biological nitrogen. Till now most of the varieties were developed through single plant selection from locals. A great need has emerged to improve the quality and yield of the crop through breeding approaches. Being an underutilized and unexplored legume crop, knowledge of germplasm diversity and genetic relationships in the available germplasm accessions is indispensable in the breeding programme for effective selection of superior diverge genotypes to be used in hybridization program that produces higher heterotic effects. The widely used technique to measure genetic diversity is Mahalanobis D² (Murthy and Arunachalam, 1966 & Dasgupta and Singh, 2003). Hence, the present investigation has been carried out to understand the nature and magnitude of genetic diversity in the existing sixty horse gram germplasm using Mahalanobis D².

**MATERIAL AND METHODS**

In the present investigation, sixty horse gram accessions were used for genetic diversity study of quantitative traits and were raised in Regional Research Station, Paiyur during *Rabi 2020* in Randomized Block Design with three replications (Table 1.) Each genotype was raised in three rows of 30 ×10 cm spacing between rows and plants. The observations were recorded on five randomly selected plants per replication for each genotype. All the suggested packages of practices were carried out in time to raise a good crop. Biometrical observations were recorded on ten quantitative traits as follows:

**Days required for 50% flowering:** The number of days required for opening of floral buds from the date of sowing in 50% of plants was recorded.

**Days for maturity:** The number of days required for the maturity of pods in plants was noted.

**No. of pods per plant:** Total number of pods on each selected plant was counted and the average was noted.

**Plant height:** The height of each randomly selected plant was measured before harvesting from the soil level of the plant to the apex by using thread and scale.

**No.of primary branches/plant:** It was counted at maturity from randomly selected plants, and the average values were recorded.

**Pod length:** The length of each pod was measured by keeping the pod on a scale and average values were recorded.

**Table 1. List of selected horse gram genotypes used for Diversity Analysis**

| S. No. | Horse gram genotypes       |
|--------|---------------------------|
| 1.     | HYD 90-20 kr              |
| 2.     | 10 EB                     |
| 3.     | HPK4                      |
| 4.     | Shivalikundam              |
| 5.     | HG66K-20                  |
| 6.     | KPT20 kr                  |
| 7.     | Pattambi                  |
| 8.     | Marugapuri                |
| 9.     | IC9628                    |
| 10.    | Mathipallil               |
| 11.    | HG20(IC8619)              |
| 12.    | Kambainallur              |
| 13.    | HG92-5-67/26              |
| 14.    | IND-1                     |
| 15.    | Vitalur                   |
| 16.    | Paichal                   |
| 17.    | Tricky Buff               |
| 18.    | Karungallur               |
| 19.    | Hebbal                    |
| 20.    | Poonamalli- 3             |
| 21.    | Poonamalli- 4             |
| 22.    | Ambasamudram              |
| 23.    | Tenkasi                   |
| 24.    | DPI 1332                  |
| 25.    | KPM 6                     |
| 26.    | NA2                       |
| 27.    | IC9626                    |
| 28.    | Mecheri                   |
| 29.    | Chinnathadagam            |
| 30.    | Paiyur                    |
| 31.    | Cuddalore                 |
| 32.    | DPI 1240                  |
| 33.    | DPI 1241                  |
| 34.    | KPT                       |
| 35.    | Chettipalayam             |
| 36.    | VLM-9                     |
| 37.    | Kollapatti,               |
| 38.    | Madam Block               |
| 39.    | Paiyur 2                  |
| 40.    | 13 EB                     |
| 41.    | VZM-1                     |
| 42.    | Bangalore 208             |
| 43.    | Bangalore 302             |
| 44.    | APLS 888                  |
| 45.    | HPK-2                     |
| 46.    | VLM Buff                  |
| 47.    | PLS-9                     |
| 48.    | KK 20 kr                  |
| 49.    | Cholachi                  |
| 50.    | T 45                      |
| 51.    | Bangalore 96              |
| 52.    | 12 EB                     |
| 53.    | IC 9606                   |
| 54.    | KK 30 kr                  |
| 55.    | 14 EC                     |
| 56.    | Tenkasi buff              |
| 57.    | HA 871-5-67/2             |
| 58.    | Morappur-1                |
| 59.    | HG 102                    |
| 60.    | VLM – 1                   |
100 seed weight: The weight of 100 well-filled seeds was determined on fine chemical balance and an average value was noted.

Grain yield per plant: The total pods on each plant at maturity were harvested separately and the seeds were taken out. The weight of total seeds per plant was recorded and average values were considered.

Bhusa yield per plant: After harvest of five randomly selected plants, dry weight was determined and expressed in grams per plant.

Statistical analysis was performed by using TNAU STAT.

RESULTS AND DISCUSSION

The analysis of variance imparted significant differences for most of the characters studied. Tocher's method was used for clustering of horse gram accessions. The sixty horse gram accessions were grouped into thirteen clusters using Mahalanobis $D^2$ analysis (Table 2.). Cluster I comprises of 39 genotypes forming the major cluster, followed by clusters III, IV and IX (3 genotypes), clusters II, V and VIII (2 genotypes), and clusters VI, VII, X, XI, XII & XIII (1 genotype). Clusters VI, VII, X, XI, XII & XIII had only one genotype which represents the uniqueness of the genotypes falling in the respective clusters.

Table 2. Clustering of sixty germplasm accessions of horse gram

| Clusters | Number of genotypes | Genotypes |
|----------|---------------------|-----------|
| I        | 39                  | HYD 90-20 kr, 10 EB, HPK4, Srivaikundam, HG6K-20, KPT20 kr, Pattambi, Marugapuri, IC9628, Mathipalli, HG20 (IC8619), Kambainallur, HG92-5-67/26, IND-1, Vitalur, Paichal, Trichy Buff, Karungallur, Hebbal, Poonamalli-3, Poonamalli-4, Ambasamudram, Tenkasi, DPI 1332, KPM 6, NA2, IC9626, Mecheri, Chinnathadagam, Pailur 1, Cuddalore, DPI 1240, DPI 1241, KPT, Chettipalayam, VLM-9, Kollapatti, Madam Block, Pailur 2 |
| II       | 2                   | 13 EB, VZM-1 |
| III      | 3                   | Bangalore 208, Bangalore 302, APLS 888 |
| IV       | 3                   | HPK-2, VLM Buff, PLS-9 |
| V        | 2                   | KK 20 kr, Cholachi |
| VI       | 1                   | T 45 |
| VII      | 1                   | Bangalore 96 |
| VIII     | 2                   | 12 EB, IC 9606 |
| IX       | 3                   | KK 30 kr, 14 EC, Tenkasi buff |
| X        | 1                   | HA 871-5-67/2 |
| XI       | 1                   | Morappur-1 |
| XII      | 1                   | HG 102 |
| XIII     | 1                   | VLM-1 |

Durga et al., 2014 have carried out the diversity study using Mahalanobis $D^2$. For the study, they have taken the twenty-three horse gram germplasm accessions. Totally six clusters were obtained and the cluster strength ranges from single genotype to 14 genotypes. Cluster I has 14 genotypes, cluster II has five genotypes, and the other clusters have only one genotype. The inter-cluster distance was found high in between cluster IV and V. Among the six clusters, cluster V recorded higher mean for leaf width, number of primary branches per plant, secondary branches per plant, seed yield, test weight and seedling length, and the intracluster distance ranges from 0 to 15.17. Cluster I had a higher intracluster distance. Clustering of horse genotypes into different clusters was also reported by Sunil et al., 2009 and Vishwanatha et al., 2016.

The inter and intracluster distances are presented in Table 3. Maximum inter-cluster distance was observed between cluster VIII and XIII (184.05) followed by cluster XI & XIII (170.83) and cluster XII & IV (143.39) and minimum inter-cluster distance was observed between cluster II and cluster VI (22.53), which indicated the lowest degree of divergence. The parents should be carefully chosen from two clusters having broader inter-cluster distance. Hence, in the present investigation, the parents can be chosen from clusters VIII and XIII in order to create variability in the progenies.

Maximum intracluster distance was observed for cluster III (31.51) followed by cluster IV (28.24, cluster VII (23.66), cluster I (23.07), cluster V (22.21),cluster IX (17.29) and cluster II (14), whereas the clusters VI, VII, X, XI, XII, and XIII recorded the distance of 0 which represents the presence of single genotype in the respective clusters. Comparing the distance of intracluster with inter clusters, the distance of intracluuster was very low indicating the homogenous and heterogenous nature of genotypes within and between clusters.

A wide range of variations was observed for cluster mean analysis for all the characters undertaken in the study (Figure 1.). For days to fifty percent flowering the cluster mean ranges from 50 days to 66.67 days. Cluster X exhibited a lower mean value for days to fifty percent flowering and for days to maturity, the cluster mean ranges from 93.17 days to 112.33 days. Cluster II contained genotypes with the lower mean value for days to maturity. Therefore, the genotypes in Cluster X and II were early in flowering. The cluster mean value of plant height ranges from 23.37 cm to 53.33 cm. Cluster X showed greater mean value for plant height. Regarding the number of branches per plant the cluster mean value ranged from 2.53 to 8.70. cluster XI contained genotypes with higher mean value for number of branches per plant. For the
trait number of pods per plant, the cluster mean value ranges from 25.40 to 52.83. Cluster XII exhibited a greater mean value for number of pods per plant. For pod length, the cluster mean value ranges from 4.23 cm to 5.27 cm. Cluster VI contained genotypes with a higher mean value for pod length and for number of seeds per pod the cluster mean value ranges from 5.17 to 5.80. Cluster VI exhibited greater mean for number of seeds per pod. With regard to the character hundred seed weight, the mean value ranges from 3.05 g to 3.56 g. In this, cluster VIII contained genotypes with higher mean value for hundred seed weight. For grain yield per plot, the cluster mean value ranges from 50.33 g to 293.83 g and the cluster VIII records higher mean value. For Bhusa yield per plot, the mean value ranges from 153.33 g to 434.67 g. Cluster VIII showed a higher mean value.

Figure 1. Cluster mean values of 13 clusters for ten different quantitative characters in horse gram and their contribution to total divergence
Among the thirteen clusters, cluster VIII exhibited a higher mean for hundred seed weight, grain yield per plot and Bhusa yield/plot, and cluster VI contained genotypes with the higher mean value for pod length and the number of seeds per pod.

The selection and choice of parents primarily rely on the contribution of characters towards divergence. Almost all the clusters were highly discrete from each other with respect to all the characters (Figure 1.). The highest contribution in the manifestation of genetic divergence was exhibited by grain yield per plot (28.30%) followed by the number of pods per plant (26.21 %) and number of branches per plant (18.36%) and moderately for Bhusa yield per plot (g) (11.35). The lowest contribution was reported for the number of seeds per pod (0.62) followed by pod length (0.96), days to maturity (1.24), hundred seed weight (1.36), days to fifty percent flowering (5.42), plant height (6.16). Greater heterotic effects can be obtained by using divergent parents for crossing rather than using closely related ones.

Table 3. Average intra and inter-cluster D2 values for thirteen clusters in sixty horse gram accessions

| Clusters | I   | II  | III | IV  | V   | VI  | VII | VIII | IX  | X   | XI  | XII | XIII |
|----------|-----|-----|-----|-----|-----|-----|-----|------|-----|-----|-----|-----|------|
| I        | 23.07 | 34.47 | 35.80 | 43.00 | 32.13 | 32.90 | 35.44 | 58.37 | 51.82 | 58.85 | 67.90 | 69.49 | 70.03 |
| II       | 14.00 | 47.69 | 65.62 | 51.03 | 22.53 | 40.53 | 117.62 | 80.87 | 79.18 | 92.42 | 56.48 | 33.46 |
| III      | 31.52 | 83.06 | 33.57 | 49.09 | 45.12 | 63.10 | 61.24 | 45.39 | 78.91 | 39.70 | 83.32 |
| IV       | 28.24 | 46.05 | 53.09 | 62.07 | 59.61 | 67.61 | 101.25 | 58.75 | 143.39 | 114.25 |
| V        | 22.21 | 35.08 | 55.87 | 44.63 | 60.36 | 61.77 | 25.72 | 61.79 | 106.63 |
| VI       | 0     | 53.18 | 102.48 | 99.44 | 112.30 | 44.79 | 74.53 | 57.36 |
| VII      | 0     | 74.97 | 89.97 | 63.42 | 107.80 | 104.50 | 83.64 |
| VIII     | 23.66 | 51.87 | 66.85 | 72.34 | 127.46 | 184.04 |
| IX       | 17.29 | 37.84 | 116.90 | 81.31 | 102.68 |
| X        | 0     | 131.21 | 56.14 | 112.01 |
| XI       | 0    | 114.98 | 170.83 |
| XII      | 0    | 84.73 |
| XIII     | 0    |  

*Bold diagonal values depicts the intra cluster distance while the other values show the inter cluster distances.

CONCLUSION

Based on inter-cluster distance, clusters VIII and XIII were found as most divergent clusters and the genotypes viz., 12 EB, IC 9606 from cluster VIII and the genotype VLM-1 from cluster VIII of these divergent clusters can be used for future improvement in heterosis in yield targeted traits with the creation of wider variability. With regard to the percentage of contribution, the highest contribution for genetic divergence was exhibited by grain yield per plot (28.30%), followed by a number of pods per plant (26.21 %). The trait contributed to maximum towards divergence needs to be given more importance for choosing further selection and choice of parents for hybridization. Based on cluster mean performance, the parents were selected from cluster X (HA 871-5-67/2) for days to fifty percent flowering, cluster VII (Bangalore 96) for days to maturity, cluster IV (HA 871-5-67/2) for plant height, cluster II (Morappur-1) for number of branches per plant, cluster XII (HG 102) for number of pods per plant, cluster VI (T 45) for pod length and number

Figure 1. Cluster mean values of 13 clusters for ten different quantitative characters in horse gram and their contribution to total divergence
of seeds per pod, cluster VIII (12EB, IC 9606) for hundred seed weight, grain yield per plot and Bhusa yield per plot(g).

REFERENCES

Dasgupta, T, and M. Singh. 2003. ICPN. ICRISAT, 10: 38-41.

Durga, K. K. 2012. Variability and divergence in horse gram (Dolichos uniflorus). Journal of Arid Land., 4(1): 71-76.

Murty, B. R. and V. Arunachalam. 1966. The nature of divergence in relation to breeding systems in some crop plants. Indian J Genet Plant Breed., 26: 188-198.

Rao, C.R. 1952. Advanced Statistical Method in Biometrical Research. John Wiley and Sons. New York.

Sunil, N., Sivaraj, N., Pandravada, S. R., Kamala, V., Reddy, P. R. and K. S. Varaprasad. 2009. Genetic and geographical divergence in horse gram germplasm from Andhra Pradesh, India. Plant Genet. Resour., 7(1): 84-87.

Viswanatha, K. P., Yogesh, L.N. and K. Amitha. 2016. Mophological diversity study in horse gram (Macrotyloma uniflorum (Lam.) Verdc) based on Principal Component Analysis (PCA). Electron. J. Plant Breed., 7(3): 767-770.