

Delineating the Genetic Diversity in Oat Genotypes through Multivariate Analysis for Utilization in Breeding Program

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Abstract

Oat (*Avena sativa* L.) is an important multi-purpose crop, cultivated for fodder, feed and grain purpose. Earlier oat was used for forage purpose but now with increasing health related issues as well as food security under changing climatic conditions; this crop has been emerged as sustainable dual purpose crop. Oat has emerged as a beneficial grain cereal for human consumption. Generally diverse individuals are likely to produce more heterotic effects during the crossing programme and desirable segregants are also produced. Therefore, in this present research, a total of 56 genotypes were evaluated for sixteen yield and yield contributing traits. K-means clustering and principal component analysis was done using R studio software. From clustering the genotypes were grouped in 4 cluster. Out of which cluster 1 and cluster 3 were most diverse. Highest cluster mean value for maximum traits was observed for cluster 3 as well. Principal component analysis showed that PF-1 and PF-2 was regarded as most important for yield factors. It was seen that PF-1 was loaded on seed yield, axis length and days to 50% flowering while PF-2 on green fodder yield, dry matter yield and plant height. Biplot depicted that variation in traits dry matter yield, green fodder yield, days to 50% flowering, seed yield and plant height was contributed by both principal component. Genotypes selected from diverse clusters can be incorporated in hybridization crop improvement programme.

Key words : Cluster analysis, oat, PCA analysis, grain, fodder.

Oat belongs to family poaceae, is an important crop used for grain, fodder and feed. Therefore, it is regarded as one of the greatest dual-purpose cereal crops, fitting well into the diets of both humans and cattle. The naturally allopolyploid farmed oat *Avena sativa* L. ($2n=6x=42$), as well as wild weedy hexaploid species such as *Avena sterilis* and *Avena fatua*, have developed through multiple cycles of interspecific hybridization and polyploidization, integrating three separate diploid genomes (Yan *et al.*, 2016). Oat is utilized for human and livestock consumption because to its high lipid, protein and mineral content. For an effective cattle sector to fulfil the demands of an

expanding population, more nutritional and high producing fodder kinds are necessary. As a result, fodder cultivars must produce large volumes of highly digestible green fodder and have excellent regeneration potential after cuttings (Stevens *et al.*, 2004). Oat forage can be sown as intercropping with a legume such as vetch, pea, berseem (Ross *et al.*, 2004; Undersander, 2003). Oat grain is rich in seed crude protein, antioxidants, soluble protein and beta-glucan. Beta-glucan is soluble fibre which helps in digestion improvement, reduces heart attack risk, treatment against asthma, obesity and cholesterol.

The heritable variation within and across populations of a species is depicted by genetic variety, also known as genetic divergence or genetic distance. A more encompassing definition is "any quantitative measure of genetic

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difference determined between individuals, groups, or species, whether at the sequence level or at the allele frequency level" (Beaumont *et al.*, 1998). Diversity analysis aid in the designing of breeding strategies to assemble available variety, in addition to offering a prognostic evaluation of genetic diversity within a species. In breeding, an accurate and exact assessment of the levels and patterns of genetic variation is beneficial. Assessment is one of the several genetic techniques being investigated for oat improvement. Because the diversity in agronomically important traits will be exploited in a crop development effort, a large collection of resources is necessary (Jaipal and Shekhawat, 2016).

PCA is a multivariate statistical approach for condensing data with many associated variables into a smaller number of new variables. It aids in the selection of the most useful major cause of divergence in the first component, and summarises that these are the core elements of plant architecture that require further attention. Keeping this in view, the present paper includes multivariate analysis using K means clustering and principal component analysis to identify most diverge analysis, understanding genetic closeness between 56 oat genotypes based on 16 yield and yield contributing traits. The paper also focuses on identification of most important characters responsible for variation.

Materials and Methods

The field experiment was performed at the Forage Research Area of the Department of Genetics and Plant Breeding, Chaudhary Charan Singh Haryana Agricultural University, Hisar (Haryana) during the *Rabi* season of 2017-18. A total of fifty six oat genotypes, including two checks (KENT and UPO212) were investigated in RCBD design (Table 1). In the present research, sixteen morphological traits i.e. Plant height (cm), Number of days to

maturity, Days to 50% flowering, Flag leaf length (cm), Leaf length (cm), Leaf width (cm), Leaf stem ratio, Internode length (cm), Number of tillers plant⁻¹, Peduncle length (cm), Axis length (cm), Number of spikelets plant⁻¹, Seed yield plant⁻¹ (g), 100 Seed weight (g), Green fodder plant⁻¹ (g), Dry fodder plant⁻¹ (g) were studied. Statistical multivariate analysis was done using R studio software. The cluster membership was analysed based on k-means clustering. The cluster plot and graphs were drawn using R studio as well.

Table 1. List of fifty six different genotypes of oat used in the experiment

Name of genotype	Source	Name of genotype	Source
HFO809	CCSHAU, Hisar	HFO615	CCSHAU, Hisar
HFO810	CCSHAU, Hisar	HFO619	CCSHAU, Hisar
HFO815	CCSHAU, Hisar	HFO525	CCSHAU, Hisar
HFO816	CCSHAU, Hisar	HFO523	CCSHAU, Hisar
HFO832	CCSHAU, Hisar	HFO505	CCSHAU, Hisar
HFO852	CCSHAU, Hisar	HFO502	CCSHAU, Hisar
HFO862	CCSHAU, Hisar	HFO498	CCSHAU, Hisar
HFO867	CCSHAU, Hisar	HFO488	CCSHAU, Hisar
HFO874	CCSHAU, Hisar	HFO430	CCSHAU, Hisar
HFO876	CCSHAU, Hisar	HFO424	CCSHAU, Hisar
HFO878	CCSHAU, Hisar	HFO419	CCSHAU, Hisar
HFO879	CCSHAU, Hisar	HFO414	CCSHAU, Hisar
HFO883	CCSHAU, Hisar	HFO409	CCSHAU, Hisar
HFO885	CCSHAU, Hisar	OS346	CCSHAU, Hisar
HFO704	CCSHAU, Hisar	HF0503	CCSHAU, Hisar
HFO706	CCSHAU, Hisar	OS403	CCSHAU, Hisar
HFO707	CCSHAU, Hisar	OS377	CCSHAU, Hisar
HFO601	CCSHAU, Hisar	HFO267	CCSHAU, Hisar
HFO602	CCSHAU, Hisar	OS305	CCSHAU, Hisar
HFO603	CCSHAU, Hisar	HFO103	CCSHAU, Hisar
KENT	AUSTRALIA	DUNAV	BULGARIA
UPO212	GBPUAT, Pantnagar	JO-1	JNKVV, Jabalpur
HFO604	CCSHAU, Hisar	RO-19	CCSHAU, Hisar
HFO605	CCSHAU, Hisar	KALOGEN	BULGARIA
HFO607	CCSHAU, Hisar	ALGERIAN	ALGERIA
HFO610	CCSHAU, Hisar	PLP-1	CSKHPAU, Palampur
HFO611	CCSHAU, Hisar	HFO872	CCSHAU, Hisar
HFO614	CCSHAU, Hisar	HFO864	CCSHAU, Hisar

Results

Using K-means cluster analysis, the diverse 56 oat genotypes were grouped into 4 clusters. Cluster 3 had maximum number of genotypes i.e. 17 followed by cluster 2 with 15 genotypes (Table 2). Cluster 1 and 4 had 10 and 14 genotypes respectively. The cluster membership plot as shown in Fig. 1 depicts the grouping pattern. Divergence between different genotypes can be seen from the plot along with genotypes occupying space of more than one cluster. The highest inter cluster distance (Table 3) was seen between cluster 1 and cluster 3 (6.66), followed by cluster 1 and cluster 2 (5.94). The highest intra-cluster distance was observed for cluster 3 (5.11) followed by cluster 4 (4.86). The highest mean value (Table 4) for traits was seen in cluster 3 i.e. number of spikelets plant⁻¹ (66.32), seed yield plant⁻¹ (g) (23.62), 100 seed weight (g) (3.29), axis length (cm) (34.34), internode length (cm) (24.4), flag leaf length (cm) (28.07), leaf length (cm) (43.82), leaf stem ratio (0.38) and peduncle length (cm) (43.31). The highest mean for number of tillers plant⁻¹ and plant height (cm) was seen in cluster 2 i.e. 8.16 and 107.35 respectively. The highest

cluster mean for trait green fodder per plant (g) and dry fodder plant⁻¹ (g) was also seen in cluster 2 i.e. 180.58 and 63.29 respectively. Cluster 1 showed highest mean for number of days to

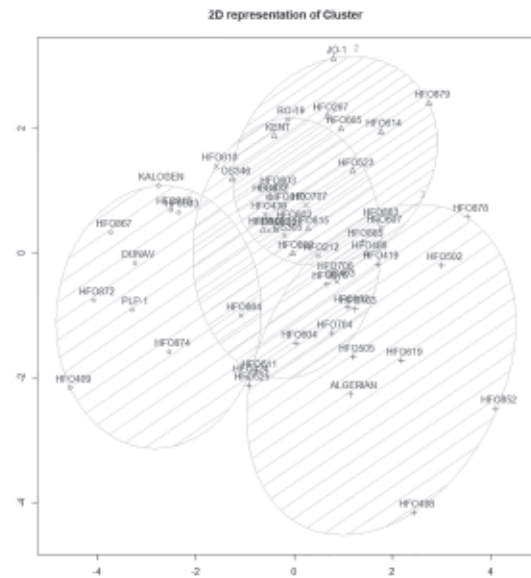


Fig. 1. K-means clustering plot. Analysis divided 56 oat genotypes into 4 clusters based on 16 yield and yield contributing traits.

Table 2. Grouping of 56 oat genotypes into four cluster based on 16 yield contributing traits

Cluster	Genotypes grouped under respective cluster
C1	HFO816, HFO874, HFO867, HFO409, HFO424, DUNAV, KALOGEN, PLP-1, HFO872, HFO503
C2	HFO809, HFO815, HFO879, HFO602, KENT, HFO605, HFO614, HFO615, HFO523, HFO488, HFO414, OS346, OS377, HFO267, JO-1
C3	HFO852, HFO862, HFO876, HFO878, HFO706, HFO604, HFO619, HFO525, HFO498, HFO502, HFO505, HFO883, HFO885, HFO704, HFO419, ALGERIAN, HFO103
C4	HFO832, HFO810, HFO707, HFO601, HFO603, HFO212, HFO607, HFO610, HFO611, HFO430, OS403, OS305, RO-19, HFO864

[TP- Number of tillers plant⁻¹, NOS- Number of spikelets panicle⁻¹, SY- Seed yield (g plant⁻¹), GFY- Green fodder yield (g plant⁻¹), DMY- Dry matter yield (g plant⁻¹), AL- Axis length (cm), SI- 100-seed weight (g), DF- Days to 50% flowering, PH-Plant height (cm), DM- Days to maturity, FLL- Flag leaf length (cm), LL- Leaf length (cm), LW- Leaf width (cm), LS ratio- Leaf: Stem ratio, IL- Internode length (cm), PL- Peduncle length (cm)]

Table 3. Inter and intra cluster distances between clusters

	C1	C2	C3	C4
C1	4.83			
C2	5.94	4.43		
C3	6.66	5.43	5.11	
C4	5.84	5.26	5.64	4.86

Table 4. Cluster mean of 16 yield contributing traits in each of four clusters

Cluster mean	TP	NOS	SY	GFY	DMY	AL	SI	IL
1	5.69	50.31	14.82	127.6	40.43	25.82	2.34	21.64
2	8.16	53.4	18.61	180.58	63.29	32.93	2.41	22.9
3	7.66	66.32	23.62	134.76	48.9	34.34	3.29	24.4
4	6.33	63.57	17.11	151.12	56.43	33.3	2.95	23.97
	PH	DM	FLL	LL	LW	LS ratio	PL	DF
1	95.51	119.2	22.38	35.77	2.12	0.34	31.65	98.27
2	107.35	115.2	27.8	41.42	2.06	0.32	38.36	94.62
3	101.76	114.65	28.07	43.82	2.28	0.38	43.31	91.53
4	104.54	120.91	26.29	43.74	2.45	0.31	38.78	98.88

[TP- Number of tillers plant⁻¹, NOS- Number of spikelets panicle⁻¹, SY-Seed yield (g plant⁻¹), GFY-Green fodder yield (g plant⁻¹), DMY- Dry matter yield (g plant⁻¹), AL- Axis length (cm), SI-100-seed weight (g), DF- Days to 50% flowering, PH- Plant height (cm), DM- Days to maturity, FLL- Flag leaf length (cm), LL- Leaf length (cm), LW- Leaf width (cm), LS ratio- Leaf: Stem ratio, IL- Internode length (cm), PL- Peduncle length (cm)]

Table 5. Principal components for 16 yield contributing traits and factor variable relations

Parameters	Principal component						
	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6	PC 7
Standard Deviation	1.85	1.41	1.26	1.14	1.1	1.03	1.02
Eigen values	3.42	1.99	1.59	1.31	1.21	1.06	1.04
Proportion of Variance	0.214	0.124	0.100	0.082	0.075	0.066	0.065
Cumulative Proportion (%)	21.38	33.81	43.77	51.93	59.46	66.09	72.59
Traits	Factor loadings						
TP	-0.28	0.05	-0.15	0.41	-0.07	0.45	-0.16
NOS	-0.24	-0.31	0.28	-0.25	0.04	-0.13	0.00
SY	-0.37	-0.19	0.14	0.04	-0.04	0.11	-0.30
GFY	-0.18	0.47	0.07	0.11	0.09	0.24	-0.17
DMY	-0.20	0.47	-0.03	-0.27	0.04	-0.04	0.06
AL	-0.33	0.09	-0.04	0.38	-0.06	-0.46	0.13
SI	-0.21	-0.23	0.29	-0.13	-0.26	-0.17	-0.52
DF	0.32	0.16	0.44	0.16	-0.03	0.20	0.16
PH	-0.27	0.42	0.09	-0.39	0.04	-0.02	0.00
DM	0.15	0.15	0.47	-0.09	-0.51	-0.08	0.22
FLL	-0.29	-0.10	-0.11	-0.07	-0.01	0.16	0.33
LL	-0.26	0.06	0.31	0.24	-0.24	0.25	0.09
LW	-0.13	-0.16	0.39	0.31	0.46	-0.09	0.32
LS ratio	-0.17	-0.30	-0.04	-0.38	-0.05	0.47	0.37
IL	-0.22	0.07	0.17	-0.14	0.45	-0.18	0.07
PL	-0.24	0.00	-0.28	0.09	-0.42	-0.28	0.37

[TP- Number of tillers plant⁻¹, NOS- Number of spikelets panicle⁻¹, SY-Seed yield (g plant⁻¹), GFY-Green fodder yield (g plant⁻¹), DMY- Dry matter yield (g plant⁻¹), AL- Axis length (cm), SI-100-seed weight (g), DF- Days to 50% flowering, PH- Plant height (cm), DM- Days to maturity, FLL- Flag leaf length (cm), LL- Leaf length (cm), LW- Leaf width (cm), LS ratio- Leaf: Stem ratio, IL- Internode length (cm), PL- Peduncle length (cm)]

maturity (119.2) while cluster 4 showed highest mean for leaf width (cm) (2.45) and days to 50% flowering (98.88).

The principal component analysis resulted that seven principal components had eigen values more than one and overall explained 72.59% of the total included variability. Principal components with eigen values higher than one were selected for interpretation (Kaiser, 1958). The first principal component explained 21.4 % of the total variation. The second, third, fourth, fifth, sixth and seventh principal components explained 12.4, 10, 8.2, 7.5, 6.6 and 6.5% of the total variability, respectively. Data presented in Table 5 clearly indicated that PF-1 was loaded on seed yield, axis length and days to 50% flowering. Likewise, PF-2 showed a strong and positive factor with the traits green fodder yield, dry matter yield and plant height. The PF-3 was loaded with days to maturity, days to 50% flowering and leaf width. The PF-4 showed the high loading value for number of tillers per plant, plant height, L:S ratio and axis length. PF-5 confirms the deviation recorded by days to maturity, leaf width and internode length. PF-6 showed the strong and positive factor with number of tillers plant⁻¹, axis length and L: S ratio. Lastly, PF-7 was loaded on 100 seed weight, L:S ratio and peduncle length.

Discussion

Knowledge of the genetic behaviour of existing oat species and cultivars is essential for making use of the genetic variety found in oat genotypes and for developing improved oat cultivars. Individuals that are more varied are more likely to cause heterotic effects throughout the crossover programme, as well as attractive segregants. Since the cluster 1 and 3 showed the highest inter cluster distance, good performing genotypes can be selected as parents in hybridization crop improvement programmes from these groups as these will be most diverge.

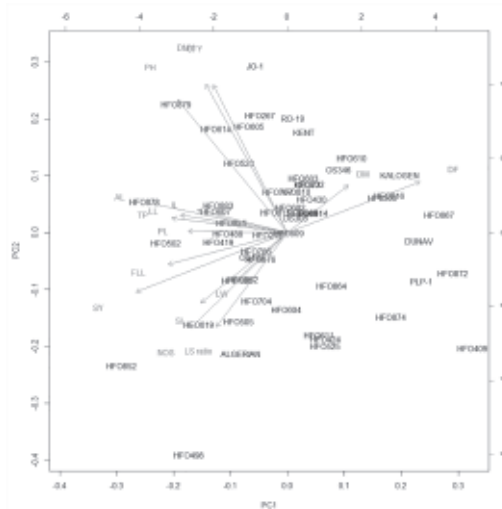


Fig. 2. PCA biplot using R studio software.

[TP- Number of tillers plant⁻¹, NOS- Number of spikelets panicle⁻¹, SY- Seed yield (g plant⁻¹), GFY- Green fodder yield (g plant⁻¹), DMY- Dry matter yield (g plant⁻¹), AL- Axis length (cm), SI- 100-seed weight (g), DF- Days to 50% flowering, PH-Plant height (cm), DM- Days to maturity, FLL- Flag leaf length (cm), LL- Leaf length (cm), LW- Leaf width (cm), LS ratio- Leaf: Stem ratio, IL- Internode length (cm), PL- Peduncle length (cm)]

Interestingly, its cluster 3 only that showed highest cluster mean for maximum number of traits under study. This makes it suitable for selection of genotypes from this cluster.

Through the PCA analysis, among the seven principal factors, PF-1, PF-2 regarded as green fodder yield factors and seed yield factors. Singh *et al.* (2018), Poonia *et al.* (2017), Kumar *et al.* (2006), Yadav *et al.* (2011) and Ahmed *et al.* (2011) also observed similar results of multivariate analysis to the present study. Biplot (Fig. 2) is made by combining score plot and loading plot. The more parallel a variable vector is to a principal component axis, the larger it contributes only to that PC. The longer the

vector, the more variation can be explained by both principal components (dry matter yield, green fodder yield, days to 50% flowering, seed yield and plant height).

Conclusion

Cluster 1 and cluster 3 showed maximum inter-cluster distance showing potential selection of diverse genotypes for oat hybridization programmes. Cluster 3 showed maximum traits that had highest cluster mean. Principal component analysis reduced the 16 traits into 7 principal component out of which PF1 and PF 2 was obtained as factor of interest.

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