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Pitout JDD, Church DL, Gregson DB, Chow BL, McCracken M, Mulvey M, Laupland KB (2007). Molecular epidemiology of CTXM-producing *Escherichia coli* in the Calgary Health Region: emergence of CTX-M-15-producing isolates. *Antimicrob. Agents Chemother.* 51: 1281-1286.

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ARTICLES

Multivariate Analysis Of Genetic Divergence Among Ethiopian Mustard (*Brassica Carinata* A. Braun) Landraces In Ethiopia

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Full Length Research Paper

Multivariate analysis of genetic divergence among Ethiopian mustard (*Brassica Carinata* A. Braun) landraces in Ethiopia

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Characterization of existing genetic variability is a prerequisite for further crop improvement activity. This study was designed to assess genetic variability among randomly selected Ethiopian mustard (*Brassica Carinata* A. Braun) genotypes from different agro ecology of Ethiopia. The present study was undertaken to determine nature of association of agronomic traits of 36 Ethiopian mustard (*Brassica carinata*) genotypes at Adet Agricultural Research Center, Ethiopia. The experiment was laid out in simple lattice design with two replications. Cluster analysis revealed that the 36 genotypes were grouped in 14 distinct clusters. The maximum average intra cluster D^2 was obtained in cluster VI ($D^2=492.03$), whereas the lowest D^2 was recorded in cluster XII ($D^2=159.94$), which shows the presence of less genetic variability or diversity within these clusters; principal component analysis revealed that ten PCs (PC1 - PC10), which are extracted from the original data and having latent roots greater than one, accounted for just about 90.5% of the total variation.

Key words: Cluster analysis, Ethiopian mustard, genetic divergence, principal component analysis.

INTRODUCTION

Ethiopian mustard (*Brassica carinata* A. Braun) is one of the most economically important crop in Ethiopia. *Brassica carinata*, commonly known as Ethiopian mustard, arose naturally from a cross between *Brassica nigra* and *Brassica oleracea* in the horn of Africa (Nigussie and Becker, 1999). The species is only found under cultivation, mainly in Ethiopia and surrounding countries (Hanelt, 1986; Tsunoda, 1980). It is cultivated as leaf vegetable and oilseed crop next from noug (*Guizotia abyssinica* Casa) and Linseed (*Linum*

ustatimum L) in the country. The oil content of mustard varies, ranging from 38-45% depending on the variety. Apart from vegetable and oil, it is also used as raw materials in industries, where its oil is indeed of immense importance in: leather tanning, manufacture of varnishes, diesel fuel, soap and lamps (Nigussie and Becker, 1999; Tesfaye et al., 2014a).

In Ethiopia, although reliable statistical information on the distribution and production of mustard is lacking, the crop has been cultivated widely in many areas of the

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country with low amount of yield (Tesfaye et al., 2014b). This might be due to the fact that mustard has been widely neglected by research and development programs (Misteru and Yared, 2013) and its genetic resources are being eroded by physical and bio-physical factors (Tewodros et al., 2013). As a result, the country frequently faces a considerable amount of genetic erosion for the last decades (Adefris, 2005; Tewodros and Getachew, 2013). Therefore, collection and evaluation of Ethiopian mustard genotypes is the best means of obtaining genetic variability for further improvement of this crop (Nigussie and Becker, 2002; Adefris and Becker, 2006).

Genetic divergence is the statistical distance between the genotypes. It is determined by using cluster analysis, which assigns genotypes into different groups (Singh and Chaudhary, 1999). Crossing of genotypes belonging to the same cluster would not be expected to yield desirable recombinants. Consequently, a crossing program might be formulated in such a way that parents belong to different clusters. The more diverse the parents, within overall limits of fitness, the greater are the chances of obtaining higher heterotic expression among F_1 's and broad spectrum of variability in segregating populations (Norden, 1980; Rao et al., 1981).

The Euclidean genetic distances are the square roots of the sum of square of the distance between the multidimensional space values of the variables for any two genotypes. This had been used to classify pair of genotypes into different groups and one of the most important biometrical techniques for estimating genetic distance present in a population (Jerry, 2001).

The use of D^2 statistic (Mahalanobis, 1936) is one of the most important biometrical techniques for estimating genetic divergences present in a population. It is defined as the statistical distance between two points and takes into account the covariance and correlations among the variables (Sharma, 1996). The D^2 values represent the index of genetic divergence among the genotypes both at intra-cluster and inter-cluster levels. Genetic architecture of a population is generally believed to be the result of breeding system, gene flow within and between population, isolation mechanism and prolonged selection by various natural and artificial forces (Chandel and Gosal, 2002).

In central and northern parts of the country, there is a huge amount of Ethiopian mustard genotypes distributed across diverse agro-geographical areas that have not been properly evaluated before (Nigussie and Becker, 2002) and their attribute remains unknown by breeders (Yared et al., 2012). So, detailed descriptions of genotypes based on agronomical characters have tremendous impact on the conservation and genetic diversity of the crop. The present study, therefore, intended to evaluate the multivariate analysis of genetic divergence among Ethiopian mustard (*Brassica carinata*) landraces using agronomic traits. So far, to characterize

and cluster within collected genotypes for further breeding works.

MATERIALS AND METHODS

Description of the experimental site

The field experiment was conducted at Adet Agricultural Research Center which is located at a latitude of 11°16'N and longitude 37°29'E at an altitude of 2240 meter above sea level (m.a.s.l.). The area receives mean annual rainfall of 1230 mm with maximum and minimum temperature of 26.1 and of 13.50°C, respectively. The soil type is sandy loam with pH 5.90.

Experimental materials and procedures

A total of 36 genotypes of Ethiopian mustard were used in the study. The genotypes were collected by Institute of Biodiversity and Conservation (IBC) from diverse agro-ecological areas of northern Ethiopia with an altitude range of 1600- 2700 m.a.s.l, representing one of the major mustard production areas in the country. The genotypes and area of collection are described in Table 1.

The experiment was laid as 6 x 6 simple lattice designs using 5 m x 1.8 m plots with two replications. Single row plots, with each row 5 m long and spacing between plots, rows and replications were 0.6, 0.3 and 2 m, respectively. The rates of fertilizer application was 40.3 and 150 kg/ha Urea and DAP, respectively. Fertilizer were applied only at sowing and the seed rate was 10 kg/ha. Other cultural practices were followed as recommended for the area (Nigussie and Becker, 2002).

Data collection

The following data was recorded from the central four rows.

1. Days to flowering (DF): It was recorded as number of days from planting to a stage when 50% of the plants in a plot produced flower.
2. Days to maturity (DM): The number of days from the date of sowing to a stage when 90% of plants have reached their physiological maturity.
3. Biomass (BM/P): The total above ground biological yield in grams obtained from each plot at harvest.
4. Harvest index (HI/P): The fraction of dry seed in the above ground biological yield on a plot basis.
5. Thousand Seed weight (TSW): The weight in grams of 500 seeds sampled from each plot and multiplied by two.
6. Seed yield (SY/P): Seed yield per plot was measured in grams after moisture of the seed is adjusted to 7%.
7. Oil content (OC): The proportion of oil in the seed to the total oven dried seed weight as measured by Nuclear Magnetic Resonance Spectrometer (NMRS).
8. Oil yield (OY/P): The amount of oil in grams obtained by multiplying seed yield per plot by corresponding oil percentage.

The data for the following characters were recorded from ten randomly taken plants each experimental plot and the average were considered per plant basis.

1. Primary branches per plant (PB/PL): The average number of primary branches per plant.
2. Secondary branches per plant (SB/PL): The average number of secondary branches formed on primary branches per plant.
3. Number of pods per plant (PD/PL): The average number of pods counted from the same sample plants.
4. Siliqua (Pod) Length (SL): The main Siliqua from the ten sampled

Table 1. List of genotypes considered in the study and their origin.

Code	Acc. No.	Area of collection	Altitude (m)	Code	Acc. No.	Area of collection	Altitude (m)	Code	Acc. No.	Area of collection	Altitude (m)
1	PGRC/E 20052	Shewa/AdisAlem	2540	13	PGRC/E208558	*	*	25	PGRC/E 21001	Shewa/Jibat	2350
2	"20059	Shewa/Chaliya	1630	14	"208559	*	*	26	"21057	Gojjam	*
3	"20068	Shewa/Ambo	2010	15	"208560	*	*	27	"21069	Bale	2450
4	"20080	*	*	16	"208565	*	*	28	"21162	Bedele	1920
5	"20163	East Tigray	2300	17	"208570	*	*	29	"21163	Wellega/Jima Arjo	1820
6	"20168	Gondar	2400	18	"208571	*	*	30	"21266	Wollo/Borena	2570
7	"20169	*	*	19	"208572	*	*	31	"21278	Welo/Desezuriya	*
8	"208507	*	*	20	"208576	*	*	32	"21369	Jimma	1720
9	"208524	*	*	21	"208584	*	*	33	"213168	Kefa	*
10	"208528	*	*	22	"208585	Shewa/Boset	1600	34	YD	Released in 1986	
11	"208545	*	*	23	"208594	Hararghe	1750	35	Holetta-1	Released in 2005	
12	"208551	*	*	24	"208961	E. Wellega	2700	36	Local check	®	2240

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plants were measured in cm and averaged to represent the pod length.

5. Number of seeds per pod (SD/PD): The average number of seeds per pod obtained from two randomly sampled pods of each of the 10 randomly taken plants.

6. Plant height (PH): The height of plants in each plot measured in centimeters from the ground surface to the top of the main stem at maturity.

Statistical analysis

Multivariate analyses of cluster and principal component analyses of genotypic values were computed using the procedures CLUSTER (ward's minimum) and PRINCOMP, respectively using SAS software version 9.00 (SAS, 2001). The genotypic values were determined by the method described by Zhu (1996) but considering the interaction component as nil (Falconer, 1981). genetic distance between clusters was calculated using the generalized Mahalanbis D^2 statistic using the equation: $D^2_{ij} = (X_i - X_j) S^{-1} (X_i - X_j)$

Where, D^2_{ij} is the square distance between any two genotypes i and j , X_i and X_j is the vectors for the values for genotype i^{th} and j^{th} genotypes, and S^{-1} is the inverse of pooled variance covariance matrix.

RESULTS AND DISCUSSION

Analysis of variance

The analysis of variance for characters showed significant differences between genotypes (Table 2). The analyzed data indicated the existence of variability within the collected genotypes for selection from genotypes and the genetic improvement of this crop. Among 16 characters, seven (that is, days to maturity, grain filling period, secondary branches per plant, harvest index, seed yield per plot, seed yield per hectare and oil content) showed highly significant ($p < 0.01$) differences among the tested genotypes.

Genetic divergence

The interest of breeders in the use of measurements of genetic diversity dissimilarity as parameters of the indication of parental lines to be

used in crosses is based on the biometric relationship between the heterosis manifested in hybrids and the divergence in the gene frequencies of parents (Falconer, 1981). More efforts have been devoted to the study of genetic divergence after proof was obtained for the existence of significant correlation between parental diversity and hybrid performance in different crops (Smith and Smith, 1987).

Cluster mean analysis

Thirty six Ethiopian mustard genotypes were grouped into XIV clusters (Table 3). The mean value of the 16 quantitative characters in each cluster was presented. The number of genotypes per cluster varied from one to eight genotypes in cluster. Clusters II and VI were the largest cluster comprising of eight genotypes, followed by Cluster IV that contained five genotypes. Two genotypes were grouped in cluster III, VII, VIII, XIV and one

Table 2. The mean squares, error and CV (%) for the 16 characters studied.

Character	Replication (df=1)	Genotypes (35)	Error (71)	CV (%)
DF	1449.01	246.83*	139.41	17.50
MD	58.68	259.38**	78.34	6.19
GFP	924.50	600.88**	135.19	15.37
PH	3068.06	8443.00*	114.54	6.29
PBP	0.01	4.68*	2.33	11.41
SBP	4.01	485.21**	34.04	15.16
LP	2.72	0.40NS	0.29	13.89
NPP	11138.24	26.44NS	4826.97	26.44
NSP	9.78	4.41NS	4.28	15.87
BM(gm)	2.14	2.213*	1.18	24.98
BMh	5932098.80	6146428.60	3266225.70	24.98
HI	94955.69	43406.92**	29184.58	29.72
TSW	0.34	0.35NS	0.29	13.49
SY	58319.07	157404.78**	101594.99	15.46
SYh	58319.07	437235.51**	101594.99	15.46
OC	2.77	6.75**	1.96	3.42
OY	1122.03	496.24*	230.82	16.69

*, ** = significant at the 0.05 and 0.01 probability levels respectively; ns = not statistically significant; Df = degrees of freedom; CV (%) = coefficient of variation; DF = days to flowering; DM = days to maturity; GFP = Grain filling period; PH = Plant height; PBP = number of primary branches per plant; SBP = Number of secondary branches per plant; LP = Length of pod; NPP = Number of pods per plant; NSP = Number of seeds per pod; BM = Biomass per plot; BMh = Biomass per hectare (kg); SY (g) = seed yield per plot; SYh = Seed yield per hectare; HI = Harvest index per plot; TSW = Thousand seed weight; OC = Oil content; OY = Oil yield per plot.

Table 3. Mean values of XIV clusters for 16 characters of the 36 genotypes.

Character	Cluster													
	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV
DF	67.8	64.0	67.3	68.3	55.0	67.0	76.0	63.5	68.5	57.7	68.4	72.7	67.0	68.0
DM	158.8	138.8	145.3	138.8	136.5	139.5	133.3	137.0	140.3	148.5	147.0	149.2	138.0	137.8
GFP	91.0	74.8	78.0	70.5	81.5	72.5	57.3	73.5	71.8	90.8	78.6	76.5	71.0	69.8
PH	169.8	149.5	179.0	177.3	164.0	168.2	165.3	178.5	173.3	175.7	171.9	169.3	156.0	167.3
NPP	203.5	169.8	109.0	97.0	75.0	160.5	102.8	344.0	155.0	204.3	131.3	111.3	80.5	101.3

Table 3. Contd.

NSP	14.3	12.3	12.1	13.0	14.7	13.8	12.0	10.0	13.8	12.9	12.8	14.7	13.1	12.5
PBP	13.1	13.6	13.9	14.6	14.3	12.4	13.2	12.5	13.5	14.3	13.2	13.5	14.1	12.1
SBP	34.5	39.5	30.5	24.3	35.5	37.8	55.0	32.5	26.8	46.5	48.7	29.3	25.5	47.5
LP	4.5	5.3	4.1	4.5	3.5	4.5	4.3	4.5	4.5	4.2	4.6	4.0	4.0	4.5
TSW	3.9	4.4	4.0	4.2	3.7	3.7	4.3	4.1	3.9	3.9	4.1	4.3	4.3	3.6
HI	538.5	342.3	466.3	670.8	1389.9	357.9	558.1	531.9	301.2	578.7	748.0	443.3	271.6	709.7
BM	3660.0	3090.0	3427.5	2550.0	1260.0	2364.0	2240.0	1890.0	2850.0	2920.0	1956.0	2750.0	3690.0	1800.0
SY	1606.3	1002.1	1332.0	1388.2	1315.5	855.3	1224.5	937.3	767.0	1620.7	1507.5	1174.8	1003.5	1277.5
SYh	2677.2	1670.2	2220.0	2313.6	2192.5	1425.4	2040.8	1562.1	1278.3	2701.2	2512.4	1957.9	1672.4	2129.2
OC	39.3	42.0	42.0	40.7	42.1	40.8	41.1	42.1	42.0	40.1	40.0	40.3	41.6	42.2
OY	110.8	69.1	94.5	94.6	104.8	81.5	84.3	72.1	65.0	108.8	105.0	84.8	83.1	97.4

DF = Days to flowering; DM = Days to maturity; GFP = Grain filling period; PH = Plant height; PBP = Number of primary branches per plant; SBP = Number of secondary branches per plant; LP = Length of pod; NPP = Number of pods per plant; NSP = Number of seeds per pod; BM = Biomass per plot; SY(gm) = Seed yield per plot; SYh = Seed yield per hectare; HI = Harvest index per plot; TSW = Thousand seed weight; OC = Oil content and OY = Oil yield per plot.

genotype (singleton) in each in cluster I, V, IX, X, XI, XII and XIII.

Cluster I consisted of one genotype having the characteristics such as late maturing (159 days), less oil content (39.3%), higher number of pod per plant (203.5), relatively high seed yield per plot (2677.2 g). Cluster II consisted of eight genotypes including the local check that comprised one released variety (Yellow Dodolla), which are characterized by the following features: heavy 1000-seed weight (4.4 g), short plant height (149.5 cm), very long pod length (5.3 cm) and relatively low seed yield per plot (1002 g). Cluster III consisted of two genotypes and also the largest plant height (179 cm) and relatively low number of seed per pod (12.1) characterized in this cluster. The cluster had intermediate characters in other agronomic traits. Four genotypes were grouped in cluster IV. High number of primary branches per plant (14.6) and low number of secondary branches per plant (24.3) were recorded.

Cluster V had one genotype. It was found with

high number of seeds per pod (14.7), heavy harvest index (1389.9), short pod length (3.5cm), poor biomass (1260gm), early maturity (136.5 days) and relatively higher oil content (42.1%).

Cluster VI consisted eight genotypes including one released variety (Holetta-1) that are characterized by relatively light 1000-seed weight (3.7 g) except and low seed yield per plot (855.3 gm).

Only two genotypes each were grouped in clusters VII and VIII. The former had late flowering (76.0), but early maturing (133) short time for grain filling period (57.3), relatively low number of seed per pod (12) and highest number of secondary branch per plant (55).

Cluster VIII had only two genotypes. High number of pod per plant (344) could characterize this cluster and lowest number of seed per pod (10) and relatively low oil yield (72.1).

Cluster IX, X, XI, XII and XIII had one genotype in each cluster that was the lowest mean seed yield per plot (767 gm) and oil yield per plot (65) in

cluster IX. The lowest number of secondary branches/plant (25.5), lowest harvest index (271.6) and the highest value of biomass were recorded in cluster XIII. In addition, cluster XIV had two genotype that could be described; only highest oil content (42) and light 1000-seed weight (3.6 g).

Divergent genotypes could have good breeding values and hybridization among members of the distant clusters is suggested to get desirable progenies. The selection of parents should also consider the special advantage of each cluster and each genotype within a cluster depending on specific objective of hybridization. Thus, the crosses involving cluster I, V, VII and XIV with any other cluster except cluster II, VIII and IX are suggested to produce high magnitude of heterosis or desirable transgressive segregants, which might be successful in the breeding of Ethiopian mustard genotypes. Quantification and classification of genetic diversity among genotypes is essential for parental selection in

Table 4. Average intra- (bold face) and inter-cluster divergence D² value in 36 Ethiopian mustard genotypes.

Cluster	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV
I	281.46	1321.64	594.32	1200.84	2611.85	1961.34	1606.48	2201.40	1838.31	741.89	1729.21	1245.13	1209.23	1977.13
II		225.82	738.83	984.67	2197.28	780.64	980.61	1234.14	518.81	1238.75	1555.24	492.73	611.09	1446.26
III			433.55	907.75	2356.65	1415.83	1209.89	1735.71	1253.05	771.30	1536.84	743.79	718.50	1649.19
IV				399.81	1484.03	1099.93	459.89	1133.37	1299.50	602.12	643.89	514.15	1420.89	781.61
V					240.21	1758.46	1298.54	1321.84	2204.20	1944.94	1019.51	1786.96	2743.04	872.27
VI						492.03	757.95	560.63	518.96	1604.63	1388.23	737.97	1362.14	1058.05
VII							314.41	703.01	1110.48	1033.81	649.01	532.92	1539.77	477.19
VIII								321.44	1058.44	1687.97	1151.50	1007.55	1842.40	732.85
IX									490.42	1685.52	1753.01	812.96	961.24	1502.90
X										402.80	1006.01	899.22	1464.26	1314.54
XI											212.48	1068.93	2048.72	476.06
XII												159.94	1012.56	1007.07
XIII													346.28	2012.24
XIV														161.97

DF = Days to flowering; DM = days to maturity; GFP = grain filling period; PH = plant height; PBP = number of primary branches per plant; SBP = Number of secondary branches per plant; LP = length of pod; NPP = number of pods per plant; NSP = number of seeds per pod; BM = biomass per plot; SY(gm) = Seed yield per plot; SYh = Seed yield per hectare; HI = harvest index per plot; TSW = Thousand seed weight; OC = oil content; OY = oil yield per plot.

breeding programs. Knowledge of the naturally occurring diversity in a population helps to identify diverse groups of genotypes that can be used for hybridization program.

Estimation of intra and inter cluster square distances (D²)

The average intra and inter cluster D² values are presented in Table 4. Maximum average of intra cluster D² was obtained in cluster VI (D²=492.03) followed by cluster IX (D²=490.42) and cluster III (D²=433.55), whereas the lowest D² was recorded in cluster XII (D²=159.94), which shows the presence of less genetic variability or diversity within these clusters.

The highest average inter cluster D² was recorded between clusters I and V (D²=2611.85)

followed by cluster III and cluster V (D²=2356.65) and cluster V and cluster IX (D² = 2204.20) which had shown these clusters were genetically more divergent from each other than any other clusters in this study. In line with Ghaderi et al. (1984), increasing parental distance implies a great number of contrasting alleles at the desired loci, and then to the extent that these loci recombine in the F₂ and F₃ generation following a cross of distantly related parents, the greater will be the opportunities for the effective selection for yield factors.

Minimum inter cluster distance was observed between cluster IV and cluster VII (D²=459.89) signifying that genotypes in these clusters were not genetically diverse or there were little genetic diversity with between these clusters. This signifies that, crossing of genotypes from these two clusters might not give higher heterotic value in our

breeding program in the subsequent generation in the F₁ and not a wide range of variability observed in the segregating F₂ population.

The maximum genetic recombination is expected from the hybridization of the parents selected from divergent cluster groups. In the present case, therefore, maximum recombination and segregation of the progenies is expected from crosses involving parents selected from cluster V and cluster III and cluster V, followed by cluster V and cluster IX, however, the breeder must specify his objectives in order to make best use of the characters where the characters are divergent.

Principal component analysis

Principal component analysis (PCA) is one of the multivariate statistical techniques, which is a

Table 5. Eigenvectors, total variance, cumulative variance and eigenvalues of the first ten principal components (PCs) for 16 characters of 36 genotypes.

Character	Eigen vectors									
	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10
Days to 50% flowering	-0.19	-0.05	0.50	0.10	0.26	0.03	-0.43	0.22	0.01	0.07
Days to maturity	0.07	0.14	-0.27	0.18	0.14	0.14	-0.30	0.29	-0.21	0.17
Grain filling period	0.19	0.30	-0.55	0.05	-0.10	0.07	0.13	0.03	-0.15	0.06
Plant height (cm)	0.04	0.07	0.0001	-0.03	0.65	-0.24	0.27	0.33	-0.25	0.19
Pods/plant	-0.07	0.20	-0.09	0.38	0.38	0.16	-0.08	-0.15	0.17	0.73
seeds/pod	-0.08	0.09	-0.17	-0.40	-0.03	0.32	-0.49	0.27	0.24	0.11
10 branches/plant	-0.03	0.15	0.04	-0.45	0.40	0.17	0.13	-0.53	0.00	0.18
20 branches/plant	0.15	-0.20	0.19	0.38	-0.12	0.24	0.02	-0.01	-0.52	0.14
Length of pod	-0.09	0.05	0.00	0.48	0.07	0.27	0.26	-0.01	0.60	0.50
1000 Seed weight (gm)	-0.03	-0.03	0.13	-0.11	-0.02	0.77	0.18	-0.03	-0.24	-0.09
Harvest index	-0.21	0.49	0.26	-0.04	-0.16	0.06	0.03	0.02	0.13	0.02
Biomass (gm/plot)	-0.21	0.49	0.26	-0.04	-0.16	-0.07	0.13	-0.02	-0.06	0.02
Seed yield(gm/plot)	0.45	0.20	0.24	-0.06	0.01	0.04	0.04	0.11	0.09	-0.06
Seed yield/ha (Kg/ha)	0.45	0.20	0.24	-0.06	0.01	0.04	0.04	0.11	0.09	-0.06
Oil Content (%)	-0.20	-0.13	0.00	-0.22	-0.06	0.12	0.47	0.60	0.13	-0.26
Oil yield per plot (OY/P):	0.42	0.11	0.16	-0.02	-0.20	-0.04	-0.02	0.01	0.19	-0.08
Eigenvalue	3.60	2.65	1.83	1.52	1.25	1.16	1.07	0.86	0.78	0.67
Difference	0.96	0.82	0.31	0.27	0.09	0.09	0.22	0.08	0.11	0.03
% of total variance	21.2	15.57	10.75	8.91	7.34	6.82	6.31	5.04	4.59	3.97
% of cumulative variance	21.2	36.77	47.52	56.43	63.78	70.6	76.91	81.95	86.54	90.51

powerful tool for investigating and summarizing underlying trends in complex data structures (Legendre and Legendre, 1998). When the PCA was run on correlations, one rule of thumb is to retain those factors whose eigen values are greater than one. The sum of the eigen values is usually equal to the number of variables. The coefficients defining the first ten principal components of these data are given in Table 5. The coefficients are scaled to present correlations between observed variables and derived components.

The analysis revealed that the first ten principal components (PC1 - PC10), scores for each individual might act as an adequate summary of the original 16 variables in any further analysis of the data having latent roots greater than one accounting nearly 90.5% of the total variation among the 36 Ethiopian Mustard genotypes studied.

These principal components might be used to summarize the original 16 variables in any further analysis of the data. Out of the total principal components retained PC1, PC2 and PC3 with values 21.2, 15.57 and 10.75%, respectively of the total variance. The five two principal components PC1, PC2, PC3, PC4 and PC5 with values of 21.2, 36.77, 47.52, 56.43 and 63.78% respectively contributed more to the cumulative variation.

According to Chahal and Gosal (2002), characters with largest absolute values closer to unity with in the first principal 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 to the small contribution of each character ($\pm 0.0001-0.7256$). The positive and negative loading shows the presence of positive and negative correlation trends between the components and the variables.

Conclusion

The analysis of variance showed the presence of highly significant differences among the tested genotypes for day of maturity, grain filling period, secondary branches per plant, harvest index, seed yield per plot and oil content showed highly significant ($p < 0.01$) difference among the tested genotypes. Likewise, length of pod and number of seed per pod showed non-significance among tested genotypes. Similarly day of flowering, plant height, primary branches per plant, biomass per plot, oil yield per plot showed significance difference at $p < 0.05$ among tested genotypes.

Genetic distance is very important for hybridization program to get better yield and best recombinant parents. Therefore, relative squared distance was estimated in this study. Thus based on these values (D^2) between any genotypes the 36 Ethiopian mustard genotypes were

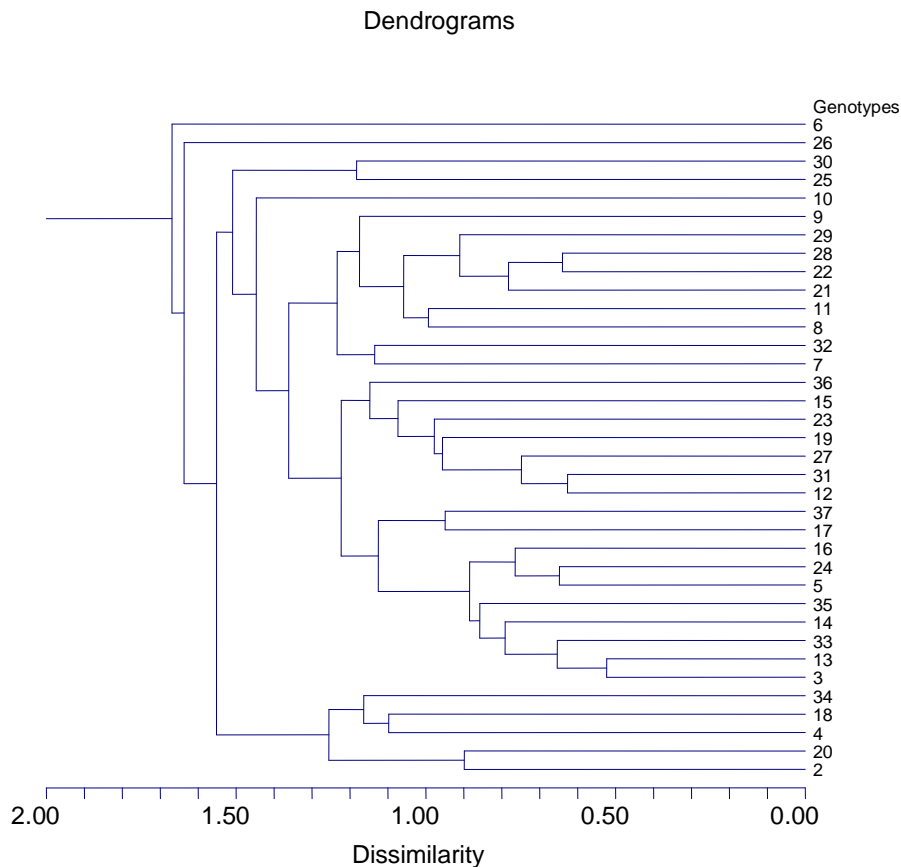


Figure 1. Dendrograms showing the clusters of 36 Ethiopian mustard genotypes.

grouped by fourteen (14) distinct clusters. Accordingly for the result the number of genotypes per cluster varied from one to eight genotypes in cluster. Cluster II and VI were the largest cluster comprising of eight genotypes followed by Cluster IV containing five genotypes. Two genotypes were grouped in Cluster III, VII, VIII, XIV and one genotype (singleton) in each in Cluster I, V, IX, X, XI, XII and XIII. These clusters were also called singletons even though they consist of only one genotype (Figure 1).

The maximum average intra cluster D^2 was obtained in cluster VI ($D^2=492.03$) followed by cluster IX ($D^2=490.42$) and cluster III ($D^2=433.55$) whereas the lowest D^2 was recorded in cluster XII ($D^2=159.94$), which shows the presence of less genetic variability or diversity within these clusters. The highest average inter cluster D^2 was recorded between cluster I and cluster V ($D^2=2611.85$) followed by cluster III and cluster V ($D^2=2356.65$) and cluster V and cluster IX ($D^2= 2204.20$) which had shown these clusters were genetically more divergent from each other than any other clusters in this study. The minimum inter cluster distance was observed between cluster IV and cluster VII ($D^2=459.89$) signifying that genotypes in these clusters were not genetically diverse or there were little genetic diversity between these clusters. The total principal components retained PC1, PC2, PC3, PC4,

PC5, PC6, PC7, PC8, PC9 and PC10 with values 21.2, 15.57, 10.75, 8.91, 7.34, 6.82, 6.31, 5.04, 4.59 and 3.97% respectively of the total variance. The five two principal components PC1, PC2, PC3, PC4 and PC5 with values of 21.2, 36.77, 47.52, 56.43 and 63.78% respectively contributed more to the total variation.

Conflict of Interests

The author(s) have not declared any conflict of interest.

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Full Length Research Paper

Genetic polymorphism of blood potassium in goat belonging to the different breeds in Mongolia

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In goats belonging to the different breeds and sub breed, the genetic polymorphism at the determinant locus of blood potassium was revealed by flame spectrophotometer method. The kalemic systems in those breeds were characterized by a polymorphism of middle level due to the existence of the two phenotypes and of three genotypes. The polymorphic character of this system is given by the distributional discontinuity of potassium ions in whole blood; the discontinuous space range were 10-34 m eq/L in the Mongolian native, 0.38-20.3 m eq/L in the Govigurbansaihan and 10.27-15.8 m eq/L in the AltainUlaan breeds. The animals with potassium ion concentration below the discontinuity space are of LK type (with low potassium) and those with ionic concentration above the discontinuity space are of HK type (with high potassium). The blood potassium level is determined by two alleles; K^L and K^h , being in incomplete dominance relationship; the allele K^L , responsible for low potassium, is dominant compared to its recessive K^h allele which causes high levels of blood potassium. These two alleles at the Ks locus, located on an autosomal chromosome, determine three genotypes; dominant homozygote ($K^L K^L$), heterozygote ($K^L K^h$), and recessive homozygote ($K^h K^h$). In the Mongolian native breed, the allele K^h was less frequent (20%) than its dominant K^L (80%), in the Govi Gurban Saihan breed, and the frequency of the alleles were also 5 and 95%, respectively. The phenotype LK (80-100%) achieved a much higher frequency than the phenotype HK (5-20%) in those breeds. Consequently, the recessive homozygosis and heterozygosis recorded an equal frequency (50%, 50%) in the Mongolian native breed, and the frequency of recessive homozygosis were slightly higher than heterozygosis (66%>34%) in the Govi Gurban Saihan.

Key words: Blood potassium, genetic polymorphism, adaptation, goat.

INTRODUCTION

The existence of two distinct levels of blood potassium ion concentrations is due to some biophysical and

biochemical features of the Na/K-ATPase activity in the membranes of the two types of red cells; enzyme that uses

energy derived from ATP hydrolysis to maintain intracellular potassium ions and expel sodium ions. This phenomenon is possible because the ATPase enzyme is intimately involved in sodium-potassium pump mechanism from the level of cell membrane (Tucker, 1971).

The goats represent the second species after sheep, on the extent of potassium polymorphism investigation. In comparison with other species in which, in most breeds, the phenotype LK is predominant (Evans and King, 1955; Evans and Phillipson, 1957; Evans, 1954a; Taneja and Ghosh, 1965), in the Mongolian native, Govi Gurban Saihan breeds and Altai ulaan sub breed goats, the phenotype LK is widespread, so the results are not similar to some other studies in goat breeds (Hrinca, 2012; Hrinca and Vicovan, 1986). Although, this characteristic is also common on the phylogenetic scale of species. In the other species of domestic animals, this research field is almost nonexistent, and only a few summary reports have been recorded, such as for cow (Ellory and Tucker, 1970; Janchiv and Merkurieva, 1978; Komatsa et al., 2004), buffalo (Sengupta, 1974), yak (Janchiv and Merkurieva, 1978; Kamenek, 1977) and zebu (Evans, 1963) among mammals and in birds within the palmiped family. This is because in these animals the kalemic polymorphism is very less obvious. The variability of potassium concentration in erythrocytes and whole blood, depending on species, breed, individual, age, sex, physiological status, etc., was frequently reported in domestic animals by clinicians and physiologists, without specifying the limits of normal and pathological ones.

Many studies have noted distributional discontinuity of potassium in the blood of animals, which has suggested that this chemical element presents polymorphism having genetic determinism (Ellory and Tucker, 1970; Erkoc et al., 1987; Evans and Phillipson, 1957; Evans, 1954b; Moradi Shahrabak et al., 2011; Mostaghni, 2004; Kamenek, 1977; Hrinca, 2012; Sengupta, 1974; Tucker 1971). The present paper proposed to study the goats' adaption ability in Mongolian dry harsh environment through revealing the genetic structure at the determination locus of blood potassium level in whole blood.

MATERIALS AND METHODS

The existence of polymorphous character of blood potassium in goats was investigated on a random population of the different breeds and sub breed from the distinctive regions in Mongolia. The blood samples were taken from animals by jugular venipuncture directly in tubes with heparin as an anticoagulant. The determination of genetic variants of blood potassium was made using the flame spectrophotometer of atomic absorption system (Shimadzu). For K determination, whole blood was diluted by distilled water in a

ratio 1:500.

Analysis of variance and correlations of blood potassium polymorphism between breeds were calculated by Data analysis in Excel program.

The detection of potassium phenotypes

The identification of the blood potassium types was made depending on the cationic concentrations of potassium in whole blood of goats. The polymorphic character of blood potassium in goats is given by the discontinuous variability of its concentration distribution. The animals with ionic concentration of potassium below the discontinuity space are of LK type (with low potassium) and those with ionic concentration above the discontinuity space are of HK type (with high potassium).

The allelic phenotypic and genotypic frequencies (f) of potassium system were calculated according to incomplete dominance phenomenon by which the kalemic system is inherited.

RESULTS

Each group consisted of 20 goats from Mongolian native, GoviGurbanSaihan andAltainulaanbreeds. The whole blood potassium concentrations ranged per group from 10-34, 0.38-20.3 and 10.27-15.8 m eq/L, respectively. The concentration distributions of blood potassium in goat populations are presented as a curve.

The curve shows that the goats could be divided into two subpopulations via LK having 0.38-18 m eq/L of K^+ and HK having 18.83 -34.09 eq/L of K^+ with mean of 6.193 ± 1.2 m eq/L of K^+ in the Govi-gurbansai Khan breed, 15.97 ± 1.12 m eq/L of K^+ in the Mongolian native and 13.56 ± 0.4 12 m eq/L of K^+ in the Altai ulaan breed goats (Figure 1).

The goat breeds are characterized by the predominance of group with phenotype LK (80-100%); the group with phenotype HK have a moderate representation (5-20%) (Figure 2). Their gene frequency obtained in the populations are given in Table 1. In those breeds, the two potassium alleles have a very unbalanced distributed. The recessive allele K^h having a very little spreading (5-20%) in comparison with its dominant K^L which has a high frequency (30-100%). As a result of this fact, the distributions of potassium genotypes are not also very uniform. Therefore, on the total populations too, the homozygosis and heterozygosis for both types were 50% in the Mongolian native breed; the homozygote (66%) was much higher compared to the heterozygote (34%) in the Govi Gurban Sai Khan breed.

DISCUSSION

The polymorphism of erythrocyte potassium was for the first time detected in sheep by Evans and King (1955).

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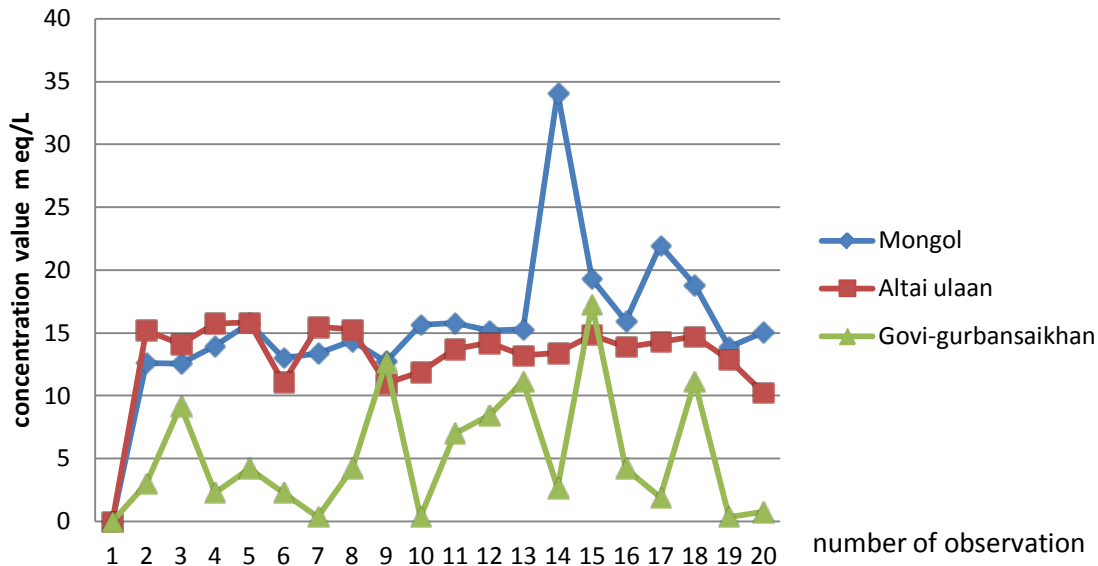


Figure 1. Concentration distributions of blood potassium in goats.

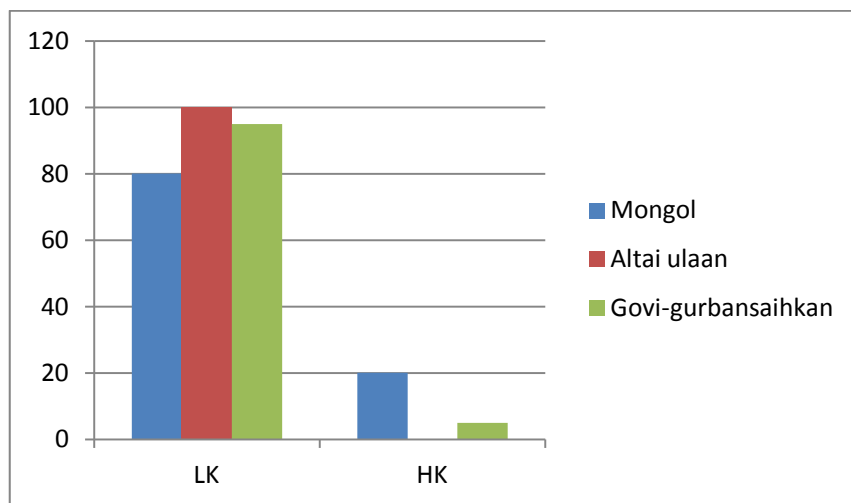


Figure 2. Phenotypic structure at the locus K in the different breeds.

Table 1. Determination gene and genotypes of blood potassium polymorphism.

Breeds of goat	Number of observation	Gene frequency (%)		
		K ^L K ^L	K ^L K ^h	K ^h K ^h
Mongol	20	30	50	20
Govi-gurbansaikhan	20	61	34	5
Altaiulaan	20	100	-	-

The existences of polymorphism for potassium in red blood cells or whole blood were confirmed in goat (Evans and Phillipson, 1957; Gurcan et al., 2011; Khan and

Taneja, 1983; Komatsa et al., 2004; Haba et al., 1991; Panon et al., 1987; Zhang, 2007). Furthermore, several studies have reported, as well as in sheep, some

associations between the biochemical polymorphism of potassium; other blood electrolytes are involved in adaptation process (Moradi Shahrababak et al., 2011; Hrinca and Vicovan, 1986; Taneja and Ghosh, 1965). Potassium polymorphism in goats can be used as a selection tool for the genetic improvement of this species if the studies concerning the association correlation of genetic structure of blood potassium with the production and reproductive traits, with the health status of individuals or with the resistance of animal body to environmental or adaptation factors require such approaches.

Conclusions

For the kalemic system the Mongolian native and Govigurbansaihan breeds were characterized by middle polymorphism due to the existence of two phenotypes (LK and HK) and of three genotypes ($K^L K^L K^L K^h$ and $K^h K^h$).

In the Mongolian and Govi Gurban Saihan breeds the allele K^L is more common; phenotype HK recorded a less frequency than LK phenotype; genotypically, there were less frequency of recessive homozygosis ($K^h K^h$), a middle incidence of heterozygosis ($K^L K^h$) and high presence of dominant homozygosis ($K^L K^L$).

The summed homozygosis of both types (dominant +recessive) were equal with the heterozygosis in the Mongol native breed.

Therefore, it can be considered that Mongolian native and Govi Gurvan Saihan goats have a good ability in adaptation.

Conflict of Interests

The author(s) have not declared any conflict of interest.

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