

MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF LOCAL COMMON BEAN (*PHASEOLUS VULGARIS* L.) GENOTYPES

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Abstract. This study was conducted for morphological and molecular characterization of 33 common bean genotypes collected from Ordu province in Turkey. Genetic relationships among the local common bean genotypes were also identified. In presented genotypes, 3 phenological and 22 morphological characteristics were investigated. The first flower and the first beans were formed 43.42 and 48.55 days after sowing time respectively. The first fresh bean harvest was realized 67.85 days after the sowing time. Pod lengths and pod widths of the genotypes varied between 10.93 and 23.23 cm (average 14.76 cm) and 9.39-22.73 mm (average 15.22 mm) respectively. The first 3 dimensions (PC1, PC2 and PC3) of principal component analysis on morphological data explained 72.27% of total variation. Seed main color, seed secondary color, pod color, pod cross-section and stringiness were prominent characteristics in screening of common bean genotypes. Of the SSR primers, SSR-IAC116 yielded the greatest PIC value (0.82) and it was followed by BMD-45-AIA (0.77) and PV ag004 (0.72) primers. The primers had a mean number of alleles per locus of 2.55. The BM210 had the greatest number of polymorphic alleles per locus (6 alleles). Cluster analysis, composed from molecular data, revealed 3 main groups. The genotypes G14 and G17 were placed alone in the first and second groups and the rest were clustered in the third group. The similarity index values among 33 local common bean genotypes varied between 0.34 and 0.97. The genotypes G04 and G22 were identified as the closest genotypes with a genetic similarity coefficient of 0.97.

Keywords: *genetic resources, diversity, SSR, PCA, polymorphism*

Introduction

Common bean (*Phaseolus vulgaris* L.) is an annual herbaceous vegetable and is a member of *Phaseolus* genus of legumes (*Fabaceae*) family. About 90% of culture common beans is constituted by *Phaseolus vulgaris* L. species of *Phaseolus* genus. There are two gene centers of beans as of Central America (Mesoamerica) and South America (Andean) (Gepts, 2008). There is no precise information about the entry time of the beans to Turkey. However, it is thought that beans entered from Europe to Anatolia 250-300 years ago (Eşiyok, 2012). Bean is produced as the main crop in several regions of Turkey and cultivated as the second crop especially in coastal regions. Ordu province is located in Central and Eastern Black Sea regions and fresh green beans are produced over 478 ha land area with an annual production of 2.870 tons (TÜİK, 2017). Agricultural cultivation and biodiversity have been tightly associated for thousands years and it is still an ongoing process (Lockwood, 1999; Norris, 2008). Turkey with current geographical position and ecological conditions is an origin and diversity center of several plants. Such a diversity resulted from the history since Turkey is among the oldest civilizations. However, natural resource diversity and richness of Turkey is continuously decreasing because of some factors such as rapid

increase in population, environmental pollution, widespread of newly released cultivars and natural disasters (Özgen et al., 1995). From such threatening factors especially high cultivar dynamism in vegetable crops have led farmers to leave local cultivars and use hybrid cultivars instead (Bellon et al., 2015). To encourage the farmers for agro-ecologic, organic and sustainable farming is the most preservative approach for such a rich biodiversity (Altieri, 1999; Finegan and Nasi, 2004; Crowder et al., 2010). Plant genetic resources are composed of local genotypes, their wild relatives, old cultivars and lines with fully-identified genetic characteristics. Local genotypes (landraces) are populations generated through natural or artificial selections for years. Just as commercial cultivars, these populations are stable, high-yield and uniform populations with low adaptation capacities for the regions other than their origin (Zeven, 1998). Besides, biotic and abiotic stress tolerance and some nutritional quality traits of local genotypes are indispensable for plant breeders (Dwivedi et al., 2016). Therefore, preservation and appraisal of local genotypes are the issues of top priority (Negri et al., 2009). Genetic richness has a significant place in plant breeding. Thus, morphologic characterization and DNA-based definition of landraces or populations and put forth of relativity degrees are significant issues in breeding studies. Due to landraces or local genotypes have a large genetic bases, such as resistance to pests and diseases and carrying genes related to several quality attributes, they constitute a significant gene source for preservation of the potential of the population they belonged to. Some genotypes even have significant quality attributes desired by the consumers and such consumers generally pay more for these products (Negri and Tosti, 2002; Galvan et al., 2006). Although common bean is originated from Central America, it is quite well-adapted to ecological conditions of Black Sea region. The region has a quite large genetic variation since local farmers have been mixed-sowing the genotypes they used for years. Morphological differences are used to put forth such a broad diversity. Morphological characterization in essence is an evolutionary method basically focusing on traditional identification of evolutionary and pedigree relations. Using morphological measurements in diversity assessment for phenotypic and agronomic traits such as flower color, growth habit, yield potential, plant height, stress tolerance etc., have limitations, because such morphological traits are governed quantitatively and under environmental influence (Skroch and Nienhuis, 1995). However, the genetic analyses carried out with DNA markers are independent from environment effect. Such analyses are also more informative and quite available for characterization of genetic materials. For genetic characterization of common bean genotypes, restriction fragment length polymorphisms (RFLP) (Velasquez and Gepts, 1994), random amplified polymorphic DNA (RAPD) (Mavromatis et al., 2010; Bukhari et al., 2015), retrotransposon-based interprimer binding sites (iPBSs) (Nemli et al., 2015) and simple sequence repeated (SSR) (Yu et al., 2000; Gaitán-Solís et al., 2002; Blair et al., 2011; Khaidizar et al., 2012) marker techniques were used. Simple sequence repeated markers, also called as microsatellite, are commonly encountered in several loci of a genome (Tautz and Renz, 1984) and these markers were successfully used in genetic characterizations of the common beans (Gomez et al., 2005; Blair et al., 2006; Buso et al., 2006; Sarikamis et al., 2009; Khaidizar et al., 2012; Ulukapı and Onus, 2013). This study was carried out to determine the genetic diversity between 33 local common bean genotypes collected from Ordu province by morphological and molecular characterization. Research findings were expected to have great contributions for definition of a common bean

genetic pool of Ordu province, for preservation of local common bean genotypes and for further breeding studies to be carried out on common beans.

Materials and methods

As the material of the study, 33 bean genotypes collected from different towns of Ordu province in Turkey were used (Fig. 1; Table 1).

The seeds were sown in 14-liter pots containing 3:1 (v/v) peat:perlite mixture. Each pot had 3 plants, and 6 plants were used for observations for each genotype.

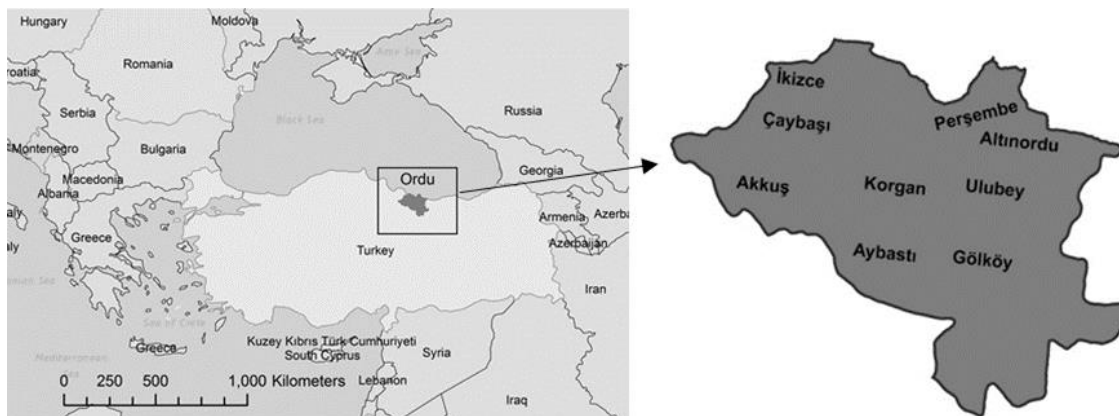


Figure 1. Location of the common bean genotypes sampling

Table 1. Coordinates of collected common bean genotypes

Genotype	Location	Latitude (North)	Longitude (East)	Altitude (m)	Genotype	Location	Latitude (North)	Longitude (East)	Altitude (m)
G01	Altınordu	40.945°	37.916°	89	G18	Çaybaşı	41.029°	37.083°	523
G02	Altınordu	40.945°	37.929°	50	G19	Çaybaşı	41.017°	37.080°	509
G03	Altınordu	40.948°	37.917°	85	G20	Akkuş	40.792°	37.007°	1243
G04	Altınordu	40.937°	37.895°	68	G21	Akkuş	40.793°	37.020°	1262
G05	Altınordu	40.968°	37.858°	160	G22	Akkuş	40.804°	37.037°	1223
G06	Altınordu	40.965°	37.869°	106	G23	Akkuş	40.713°	37.036°	951
G07	Altınordu	40.957°	37.885°	83	G24	Akkuş	40.709°	37.000°	1045
G08	Altınordu	40.943°	37.882°	134	G25	Akkuş	40.882°	37.066°	1122
G09	Perşembe	41.055°	37.777°	13	G26	İkizce	41.057°	37.086°	141
G10	Perşembe	41.116°	37.759°	25	G27	Aybastı	40.669°	37.423°	978
G11	Perşembe	41.033°	37.788°	250	G28	Aybastı	40.671°	37.413°	749
G12	Ulubey	40.887°	37.785°	447	G29	Aybastı	40.672°	37.397°	698
G13	Altınordu	40.968°	37.959°	49	G30	Aybastı	40.680°	37.428°	911
G14	Altınordu	40.961°	37.967°	159	G31	Aybastı	40.686°	37.446°	1048
G15	Gök köy	40.749°	37.676°	796	G32	Aybastı	40.714°	37.411°	775
G16	Korgan	40.826°	37.335°	678	G33	Aybastı	40.696°	37.399°	802
G17	Çaybaşı	41.034°	37.104°	289					

Morphological observations

Local common bean genotypes were evaluated for the time elapsed from sowing time to the first bloom (Flo, day), the first pod formation time (PF, day), the first fresh bean harvest time (GPH, day), pod length (PL, cm), pod width (PW, mm), leaf length (LL, cm), leaf width (LW, cm), terminal leaflet length (TLL, cm), terminal leaflet width (TLW), leaf color (LC), seed length (SL, mm), seed width (SWi, mm), 100 seed weight (SWe, g), pod cross-section (PCS), pod roughness (PR), stringiness (S), shape of distal part (beak, B), pod color (PC), pod secondary color (PSC), pod curvature level (PCL), seed clarity (SC), main seed color (MSC), secondary seed color (SSC) and distribution of secondary color on the seed (DSSC) characteristics in accordance with the standards specified in Genchev and Kiryakov (2005).

Molecular characterization

The BM146, BM210, Bmd-45-AIA, Bmd-8, DROUGH1, PH10B11, PH7B3, PV aaat001, PV ag004, PV at007, PV at008, PV atcc001, PV atcc003, PV atct001, PV gaaat001, SSR-IAC26, SSR-IAC63 and SSR-IAC116 primers selected among the ones used by Ulukapı and Onus (2013) were used for molecular characterization of present common bean genotypes. DNA isolation was performed by using CTAB (cetyl trimethyl ammonium bromide) method in accordance with the protocol of Haymes (1996). DNA concentrations were diluted with TE solution (10 mM Tris, 1 mM EDTA, pH 8.0) to 5 ng/μl. PCR reactions were performed in LongGene A300 Fast thermal cycler in a total volume of 15 μl generated through addition of 7.5 μl PCR Master Mix (Dreamtaq Green Master Mix), 1 μl forward primer (10 pmol), 1 μl reverse primer (10 pmol), 2.5 μl ddH₂O and 3 μl DNA. PCR amplifications were performed in accordance with the protocol of Khaidizar et al., (2012). Amplification conditions were set as; pre-denaturation at 94 °C for 3 min, 2 cycles at 94 °C for 30 s, at 37 °C for 60 s, at 72 °C for 2 min, 2 cycles at 94 °C for 30 s, at 50 °C for 60 s, at 72 °C for 2 min, 41 cycles at 93 °C for 30 s, at 50 °C for 60 s, at 72 °C for 2 min and finally at 72 °C for 5 min. Resultant SSR-PCR products were run in 3% agarose (Fisher BioReagents) gel containing 1x TAE (Tris-Acetic acid-EDTA) solution in SCIE-PLAS (Hu20) electrophoresis unit. Electrophoresis process was performed at 90 Watt and 300 mA current rates for 4 hours. Agarose gel was stained with ethidium bromide (10 mg/ml) for 20 min and imaged under UV transilluminator (Syngene-Ingenius). Bands in resultant gel images were scored as 1 (present) and 0 (absent) and binary data matrices were generated for data analyses.

Statistical analysis

Descriptive statistics for morphological data were calculated with SPSS v.22.0 statistical software, principal component analysis for qualitative phenotypic data was performed with the aid of Past3 software. Polymorphism information content (PIC) values of SSR primers were calculated with the aid of the following modified by Anderson et al. (1993):

$$PIC_i = 1 - \sum_{j=1}^n P_{ij}^2$$

where p_{ij} is the frequency of the j^{th} allele for marker i and summation extends over n alleles. Molecular data were analyzed via Dice module of NTSYSpc v.2.02 (Rolf, 2000) software. Cluster analysis was performed by using UPGMA (un-weighted pair group method with arithmetic mean) (Sneath and Sokal, 1973) and a correlation matrix was generated then.

Results and discussion

Morphological characterization of bean genotypes

With regard to type of growth, 93.93% of the genotypes were climbing type (31 genotypes) and 6.07% showed dwarf growing habit (G09, G24). Sözen et al. (2014) carried out a study with 85 bean genotypes and identified 12 genotypes (14.1%) as dwarf, 42 genotypes (49.4%) as semi-dwarf and 31 genotypes (36.5%) as climbing type. Descriptive statistics for quantitative pod and leaf characteristics and phenological observations of the local bean genotypes are provided in *Table 2*. Mean flowering time of the local genotypes apart from the sowing time was 43.42 days. The earliest flowering was observed in G09, G15, G21, G26 and G27 genotypes with 41 days and the latest flowering was observed in G07 with 55 days. The earliest genotype formed the first pod in 46 days and the latest genotypes formed the first pod in 59 days. Mean the first pod formation time was 48.55 days. The first fresh pod harvest was performed in 58 days in early genotypes (G01, G24, G26, G27 and G32) and in 85 days in late genotype (G07). Pod lengths varied between 10.93 and 23.23 cm. The shortest pod was observed in G09 genotype with 10.93 cm and the longest pod was observed in G33 genotype with 23.23 cm. Pod widths varied between 9.39 and 22.73 mm. The lowest pod width value was obtained in G14 genotype with 9.39 cm and the largest pod was obtained in G25 genotype with 22.73 cm as was seen in *Table 2*. Variation coefficients for tip leaf length, tip leaf width, side leaf length and side leaf width were respectively calculated as 15.67, 13.93, 14.49 and 13.43%. Kar et al. (2005) assessed earliness, yield and quality attributes of 4 determinate and 5 pole-type bean cultivars under unheated greenhouse conditions. Researchers reported the earliest flowering time as 58 days for determinate cultivars and 59 days for pole-type cultivars and reported the first pod harvest times as between 63 and 68 days. Erdiñ et al. (2013) reported the earliest flowering time as 42 days, the latest flowering time as 77 days, the first pod harvest time as 68 days and the latest pod harvest time as 127 days in different common bean genotypes. In another genetic characterization study carried out with 300 common bean genotypes in Honduras, the first flowering times were reported as between 31 and 37 days (Meza et al., 2013). Akbulut et al. (2013) carried out a study with 12 bean genotypes grown in Burdur province and reported the first pod formation time as between 46 and 68 days. In similar previous studies carried out with common beans in Turkey, pod lengths were reported as between 7.48 and 13.8 cm and pod widths as between 7 and 25 mm (Düzdemir, 1998; Madakbaş et al., 2004).

Seed characteristics evaluations of local bean genotypes are provided in *Table 3*. 100 seed weights varied between 29.67 and 66.40 g. The lowest seed weight value was observed in G14 and the highest seed weight value was obtained from G06 genotype. The greatest variation was observed in seed thickness (16.91%) and it was followed by seed weight (14.85), seed length (9.56%) and seed width (7.62%). Piergiovanni et al. (2006) reported that seed variation of the common bean germplasm in Abruzzo and Lazio (Italy) was low and 100-seed weight varied between 32.9 and 91.4 g.

Table 2. Descriptive statistics data obtained from some plant growing and leaf characteristics

	Flo (day)	PF (day)	GPH (day)	PL (cm)	PW (mm)	TLL (cm)	TLW (cm)	LL (cm)	LW (cm)
Mean	43.42	48.55	67.85	14.76	15.22	14.88	10.89	14.05	10.24
Minimum	41.00	46.00	58.00	10.93	9.39	10.71	8.53	10.91	8.04
Maximum	55.00	59.00	85.00	23.23	22.73	23.77	17.75	21.48	16.25
Std. dev.	2.93	2.59	6.01	2.88	2.98	2.33	1.52	2.04	1.37
CV %	6.74	5.33	8.85	19.54	19.55	15.67	13.93	14.49	13.43

Std. dev.: standard deviation; CV: coefficient of variation

Table 3. Descriptive statistics data obtained from the seed characteristics

	100 SWe (g)	SL (mm)	SWi (mm)	ST (mm)
Mean	49.07	14.09	8.21	6.52
Minimum	29.67	11.70	6.22	5.04
Maximum	66.40	16.28	9.30	10.42
Std. dev.	2.19	1.34	0.62	1.10
CV %	14.88	9.56	7.62	16.91

Std. dev.: standard deviation; CV: coefficient of variation

Principal component analysis (PCA)

Results of principal component analysis for 12 qualitative characteristics of bean genotypes are provided in Table 4. PC 1 (41.97%), PC 2 (21.37%) and PC 3 (8.93%) axes explained 72.27% of total variation among the genotypes. On PC 1 axis, seed main color with a variation coefficient of 0.95 was the primary attribute, the most distinctively indicating the variation among the genotypes. On PC 2 axis, pod secondary color (0.80) and seed secondary color (0.75) were the most distinctive characteristics. On PC 3 axis, pod cross-section (0.82) and stringiness (0.61) were the most distinctive characteristics effecting the variation among the genotypes. Lima et al. (2012) reported that first two principle components explained about 34% of the total variation on common bean genotypes. Meza et al. (2013) indicated that first tree principle components explained 34.18% of total variation and reported the first flowering time, ripened pod color and pod harvest time as the most distinctive characteristics.

Molecular characterization

The 18 SSR markers used in molecular characterizations generated a total of 63 alleles and 46 of them (73%) were polymorphic among the common bean genotypes. Number of alleles per locus varied between 2 and 6 (average 2.55 alleles per locus) (Fig. 2). While PBM210, PV aaat001 and PV ag004 SSR primers yielded the greatest number of alleles per locus, the greatest number of polymorphic alleles per locus (6 alleles) was observed in BM210 primer. Polymorphism information content (PIC) of the primers varied between 0.06 and 0.82 (Fig. 3). The greatest PIC value (0.82) was obtained from SSR-IAC116 primer.

Table 4. Eigenvectors of the first three dimensions of PCA

Characters	PC 1	PC 2	PC 3
Pod cross section	-0.06	0.11	0.82
Pod roughness	-0.32	0.57	0.10
Stringiness	0.22	0.25	0.61
Shape of distal part (beak)	-0.03	-0.26	-0.38
Pod color	-0.31	-0.02	0.22
Pod secondary color	-0.34	0.80	-0.03
Pod degree of curvature	0.26	-0.11	0.49
Seed clarity in pods	-0.27	0.27	0.06
Leaf color	-0.27	0.06	0.23
Seed main color	0.95	0.30	-0.03
Seed secondary color	-0.55	0.75	-0.08
Distribution of seed secondary color	-0.57	0.54	-0.11
Individual variance %	41.97	21.37	8.93
Cumulative variance (PC1 + PC2 + PC3): 72.27%			

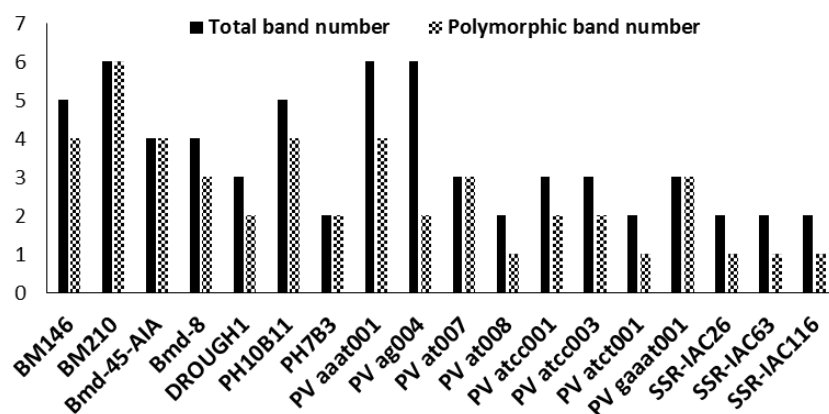


Figure 2. Total number and polymorphic band numbers revealed by SSR primers in common bean genotypes

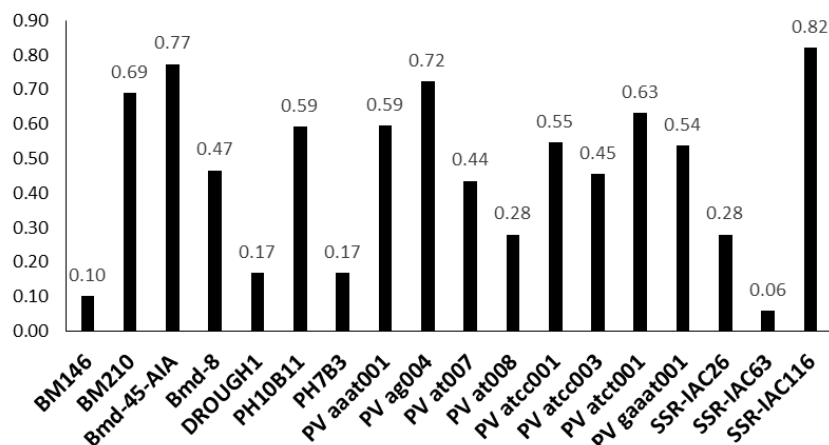


Figure 3. Polymorphism information content of SSR primers in common bean genotypes

Lioli et al. (2005) carried out a diversification study with 33 local common bean genotypes using 14 SSR primers and reported number of alleles per locus as between 2 and 11. Sarıkamış et al. (2009) used 12 SSR primers in a genetic diversity study for 30 common bean genotypes and identified 10 SSR primers as polymorphic. Researchers obtained 45 polymorphic alleles from 10 SSR primers and reported number of alleles per locus as between 2 and 10. Khaidizar et al. (2012) identified bean genotypes collected from Northern Anatolia with the aid of 30 SSR primers and obtained 72 alleles from these SSR primers. Ulukapı and Onus (2013) indicated 73% polymorphism ratio for 22 SSR primers and reported PIC values as between 0.047 and 0.373. Buah et al. (2017) used 6 SSR primers for bean genotypes and obtained 41 alleles (average 7.8 alleles per locus). De Luca et al. (2018) used microsatellite markers for Italian local bean genotypes and reported PIC values as between 0.315 and 0.928.

Genetic relationships between local common bean genotypes

Results obtained from PCR reactions revealed that similarity index among bean genotypes varied between 0.34 and 0.97 (Table 5). A dendrogram was generated with SSR primers (Fig. 4) and bean genotypes were separated into 3 main groups in this dendrogram.

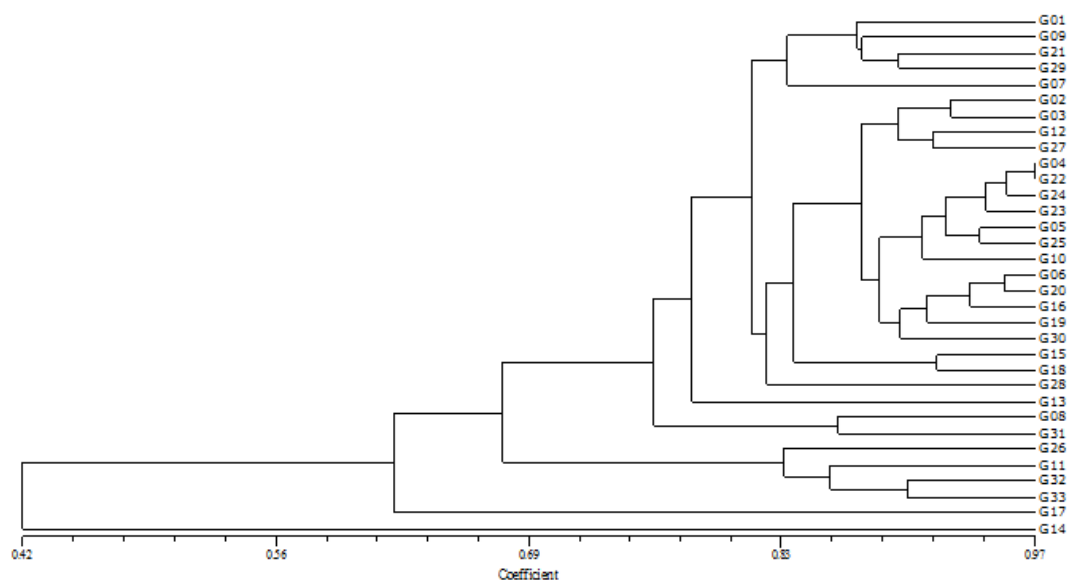


Figure 4. UPGMA dendrogram based on SSR markers

The genotype G14 collected from Altınordu town of Ordu and the genotype G17 collected from Çaybaşı town of Ordu were alone placed in the first and second group of the dendrogram. The difference of G14 and G17 genotypes were also quite distinctive in principal coordinate scatter plot (Fig. 5). All the other genotypes were clustered in the third group. The genotypes G04 and G22 collected from Altınordu and Akkuş towns of Ordu province were the closest genotypes with a genetic similarity index of 0.97. Cluster analysis did not yield town-based geographical separation of the genotypes. Such a case was because local bean producers generally produce their own seeds and

seeds of a locality are served to different local markets. Khaidizar et al. (2012) reported genetic similarity coefficient between the bean genotypes collected from Erzurum and Bayburt provinces as between 0.211 and 0.796 and gathered bean genotypes under two main groups. Ulukapı and Onus (2013) used SSR primers for characterization of 39 bean genotypes and reported genetic similarity coefficients as between 0.52 and 0.98. Researchers gathered bean genotypes under two main groups of a dendrogram. Bukhari et al. (2015) carried out a characterization study with 45 bean genotypes and separated bean genotypes into 7 main groups and reported genetic similarity coefficients as between 0.56 and 0.92.

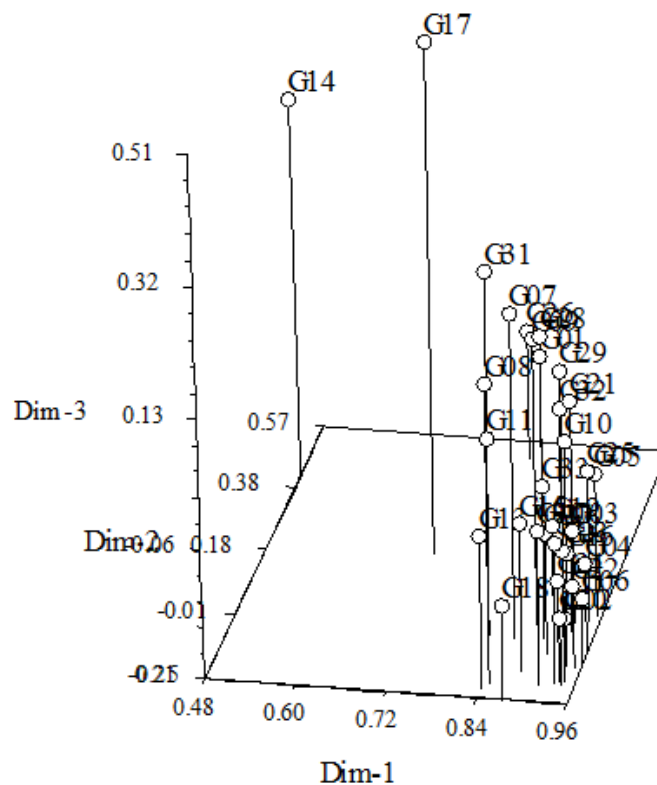


Figure 5. Scatterplot illustration drawn using SSR data

Conclusion

Genetic variations in a gene pool constitute basic sources for breeders. Hereditary characteristics of the materials in this genetic pool should be identified with proper methods and genetic relations among them should be put forth. Such efforts play a significant role in meeting farmer needs. Present local common bean genotypes grown in Central Black Sea region exhibited differences in seed color, pod color and pod stringiness. Molecular characterizations by SSR markers used in this study did not revealed distinctive groups or geographical separations with regard to morphological characteristics. Bean genotypes of Ordu province had a genetic similarity of between 34 and 97%. Such a broad genetic variation may have significant contributions in developing new common bean cultivars.

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Table 5. Genetic similarity matrix between 33 common bean local accessions based on SSR markers

	G01	G02	G03	G04	G05	G06	G07	G08	G09	G10	G11	G12	G13	G14	G15	G16	G17	G18	G19	G20	G21	G22	G23	G24	G25	G26	G27	G28	G29	G30	G31	G32	G33
G01	1.00																																
G02	0.82	1.00																															
G03	0.80	0.93	1.00																														
G04	0.77	0.90	0.91	1.00																													
G05	0.82	0.88	0.90	0.96	1.00																												
G06	0.83	0.93	0.91	0.94	0.93	1.00																											
G07	0.85	0.75	0.79	0.77	0.81	0.79	1.00																										
G08	0.78	0.75	0.76	0.77	0.75	0.84	0.81	1.00																									
G09	0.87	0.79	0.75	0.81	0.84	0.80	0.87	0.72	1.00																								
G10	0.82	0.83	0.87	0.94	0.92	0.90	0.81	0.81	0.86	1.00																							
G11	0.62	0.66	0.69	0.64	0.68	0.67	0.59	0.54	0.57	0.59	1.00																						
G12	0.86	0.92	0.91	0.88	0.86	0.91	0.82	0.79	0.81	0.87	0.57	1.00																					
G13	0.73	0.79	0.81	0.81	0.76	0.84	0.72	0.71	0.72	0.79	0.49	0.86	1.00																				
G14	0.43	0.41	0.42	0.40	0.45	0.37	0.34	0.36	0.37	0.40	0.43	0.44	0.40	1.00																			
G15	0.75	0.85	0.80	0.87	0.85	0.80	0.74	0.74	0.77	0.84	0.58	0.86	0.70	0.43	1.00																		
G16	0.82	0.87	0.92	0.92	0.90	0.95	0.85	0.81	0.81	0.92	0.67	0.90	0.85	0.35	0.81	1.00																	
G17	0.67	0.57	0.67	0.64	0.69	0.58	0.73	0.62	0.70	0.68	0.48	0.61	0.59	0.44	0.58	0.64	1.00																
G18	0.68	0.83	0.77	0.88	0.83	0.84	0.67	0.75	0.70	0.81	0.54	0.83	0.74	0.41	0.92	0.81	0.49	1.00															
G19	0.80	0.84	0.85	0.89	0.87	0.92	0.83	0.86	0.75	0.85	0.68	0.85	0.75	0.36	0.85	0.92	0.59	0.86	1.00														
G20	0.82	0.91	0.90	0.93	0.88	0.96	0.78	0.82	0.77	0.86	0.61	0.92	0.85	0.36	0.85	0.92	0.57	0.89	0.90	1.00													
G21	0.86	0.83	0.84	0.88	0.92	0.88	0.82	0.75	0.88	0.90	0.61	0.87	0.75	0.44	0.79	0.87	0.70	0.80	0.81	0.86	1.00												
G22	0.81	0.93	0.88	0.97	0.93	0.94	0.73	0.77	0.83	0.91	0.60	0.88	0.81	0.40	0.87	0.89	0.60	0.88	0.86	0.93	0.88	1.00											
G23	0.78	0.88	0.86	0.96	0.91	0.92	0.74	0.81	0.80	0.94	0.60	0.89	0.82	0.43	0.88	0.89	0.60	0.89	0.85	0.91	0.86	0.96	1.00										
G24	0.80	0.93	0.85	0.94	0.90	0.91	0.72	0.76	0.82	0.87	0.55	0.91	0.79	0.42	0.89	0.87	0.58	0.90	0.85	0.93	0.88	0.97	0.92	1.00									
G25	0.79	0.88	0.93	0.96	0.94	0.90	0.78	0.79	0.81	0.89	0.68	0.86	0.76	0.45	0.85	0.87	0.69	0.83	0.87	0.88	0.86	0.93	0.91	0.90	1.00								
G26	0.69	0.69	0.72	0.74	0.77	0.70	0.60	0.56	0.68	0.68	0.83	0.64	0.53	0.53	0.63	0.67	0.60	0.60	0.68	0.68	0.75	0.70	0.67	0.69	0.77	1.00							
G27	0.87	0.87	0.89	0.86	0.87	0.89	0.79	0.75	0.79	0.85	0.63	0.92	0.85	0.46	0.79	0.86	0.56	0.79	0.79	0.87	0.85	0.86	0.89	0.82	0.84	0.64	1.00						
G28	0.81	0.75	0.80	0.87	0.85	0.83	0.77	0.82	0.80	0.89	0.58	0.79	0.73	0.43	0.78	0.82	0.71	0.79	0.84	0.82	0.83	0.84	0.85	0.83	0.89	0.73	0.76	1.00					
G29	0.89	0.79	0.80	0.87	0.89	0.87	0.81	0.78	0.87	0.89	0.58	0.86	0.77	0.49	0.78	0.86	0.67	0.79	0.84	0.85	0.90	0.87	0.88	0.87	0.85	0.73	0.87	0.85	1.00				
G30	0.84	0.87	0.82	0.86	0.84	0.92	0.83	0.84	0.82	0.84	0.57	0.92	0.79	0.37	0.80	0.88	0.56	0.81	0.89	0.90	0.81	0.86	0.87	0.89	0.84	0.64	0.82	0.84	0.87	1.00			
G31	0.68	0.70	0.72	0.76	0.74	0.75	0.71	0.86	0.71	0.80	0.55	0.71	0.65	0.45	0.77	0.72	0.70	0.78	0.82	0.74	0.75	0.76	0.81	0.75	0.78	0.63	0.65	0.81	0.77	0.75	1.00		
G32	0.72	0.78	0.75	0.79	0.82	0.79	0.64	0.61	0.74	0.74	0.85	0.71	0.62	0.50	0.67	0.71	0.61	0.67	0.71	0.74	0.80	0.78	0.76	0.75	0.79	0.87	0.71	0.72	0.75	0.71	0.67	1.00	
G33	0.72	0.80	0.77	0.76	0.79	0.78	0.71	0.61	0.74	0.71	0.87	0.71	0.66	0.38	0.67	0.77	0.58	0.61	0.74	0.73	0.74	0.75	0.70	0.71	0.76	0.80	0.71	0.66	0.69	0.71	0.60	0.90	1.00

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