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PHENOTYPIC AND MOLECULAR GENETIC DIVERSITYAMONG SOME TURKISH BEAN GENOTYPES

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ABSTRACT

The genetic relationships among 96common Turkish bean (*Phaseolus vulgaris* L.) genotypes were investigated based on72 phenotypic characters and molecular ISSR and RAPD markers. Main seed color and predominant secondary seed color were determined as the primary characteristics to distinguish the bean accessions. The mentioned traits, among 72 individual phenotypic traits, explained 58.46 % of phenotypic variation in the first three axes of the principal component analysis (PCA) beside flower duration, plant height, and a number of nodes on the stem. The 21 ISSR primers and 8 RAPD primers having clear and readable band data were also employed, 358 and 116 polymorphic bands were obtained from them, respectively. Polymorphism information content value for ISSR varied between 0.15 and 0.50, while that for RAPD was from 0.31 to 0.48. Among the examined genotypes, molecular genetic relationship determined based on dendrogram obtained by Jaccard distance matrix. Based on the results, the 52% and 48% of the bean genotypes were categorized as Mesoamerican and Andean originated genotypes. In addition, genetic variation values were determined by using Nei and Shannon coefficients and they were highly variable among bean genotypes.

Keywords: Common bean, genetic diversity, molecular markers, phenotypic characters.

INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is originated from Latin America and has been cultivated for 7000 years (Salk *et al.* 2008). It has two gene pools as Mesoamerican and Andean (Duran *et al.* 2005; Galvan *et al.*2006; Blair*et al.* 2006; Kwak and Gepts 2009; Angioi *et al.* 2010). It has been grown in almost all parts of Turkey for 300 years (Salk *et al.* 2008).

China ranks first in the production of fresh broad beans, and Turkey's production with 632.301 tons meets 2.96 % of world fresh broad bean production (FAO, 2013). There has been a large variability in common bean having a widespread distribution in Turkey (Erdinc *et al.* 2013a).Determination of genetic diversity among local bean populations is of great importance especially in local breeding programs aiming cultivar improvement and germplasm management (Jose *et al.* 2009). The phenotypic traits (Duran *et al.* 2005; Galvan *et al.* 2006; Chiorato *et al.* 2007) and molecular markers (Blair *et al.* 2005; Paneda *et al.* 2005; Gonçalves-Vidigal *et al.* 2007; Zhang *et al.* 2008; Jose *et al.* 2009; Galvan *et al.* 2010) could be used in bean genetic diversity studies. It has been reported that RAPD and ISSR markers are frequently used to determine the genetic diversity in common bean as well as other crops (Tiwari *et al.* 2005; Sicard *et al.* 2005; Marotti *et al.* 2006; Sensoy *et al.*2007; Tertivanidis *et al.* 2008; Turkmen *et al.* 2012; Erdinc *et al.* 2013b).

In Turkey, characterization studies in bean were done with biochemical markers or phenotypic traits (Balkaya, 1999; Balkaya and Yanmaz, 2002; Madakbas and Ergin, 2011; Madakbas *et al.* 2012), but molecular markers are rarely used for instance in Erciş-Gevaş district of Van province (Sarikamis *et al.* 2009), Northeast Anatolia (Khaidizar *et al.* 2012), Eskişehir (Ceylan *et al.* 2014) and Kırşehir (Madakbas *et al.* 2016). This study aimed to determine the genetic diversity among some Turkish bean genotypes using both phenotypic characters and molecular markers. We hope that the data introduced here will improve the efficiency of bean selection and improvement programs.

MATERIALS AND METHODS

Plant material: Ninety-five common bean (*Phaseolus vulgaris* L.) and one runner bean (*P. coccineus* L., as a check/outgroup genotype) accessions collected from different regions of Turkey were used in this study (Table 1).

Phenotypic characters: Data on 72 morphological traits were recorded according to UPOV (2005) protocol, and the technical instructions of the Turkish Ministry of Agriculture. Ninety-six bean genotypes were grown in an open field in Gevas-Van Province of Turkey. Three

replicates of 10 plants (1 m x 0.2 m spacing) per accession were grown in a completely randomized design. Total seventy-two phenotypic characters were investigated. Quantitative characters were transformed into 3-5 distinct classes.

DNA extraction: DNA extraction was done according to a modified CTAB procedure (Doyle and Doyle, 1987). Biotech UV 1101 photometer was employed for DNA quantification.

ISSR and RAPD analyses: The 21ISSR primers and 8 RAPD primers (Table 2) were employed (Tiwari *et al.* 2005; Marotti *et al.* 2006; Martins *et al.* 2006). PCR reaction mixture had: 30 ng DNA, 1.5 mM $MgCl_2$, 0.2 µM Primer, 0.2 mM dNTP, 1X PCR buffer (100 mM Tris-HCl, pH 8.8, 50 mM KCl), 1 unit of Taq DNA polymerase (Promega, USA) in a total volume of 20 µL. The ISSR PCR reactions modified from Marotti *et al.* (2006) were carried out as follow: denatured (3 min at 94 °C), then (45 cycles of 60 sec at 94°C, 60 sec at 38.0-60.6°C, and 2 min at 72°C) and a final extension (at 72°C for 7 min). The RAPD PCR reactions were modified from Galvan *et al.* (2006): denatured at 94 ºC (1 min), then (45 cycles: at 94°C (45 sec), at 36°C (60 sec), at 72°C (1 min)) and a final extension at 72°C (7 min). The amplified products were electrophoresed on 1.5 %agarose gel in 1X TAE buffer at 90 V for 2 h and then visualized.

Data analysis: Jaccard distance matrix was employed to explore the molecular genetic variation among the studied beans. In order to determine similarity coefficients, the polymorphic ISSR and RAPD bands were scored as 1 (presence) or 0 (absence) (Tiwari *et al.* 2005; Martins *et al.* 2006). For each primer, the mean polymorphism information content (PIC) calculated according to following formula: $PIC= (1-pi^2)/n$, where pi is the frequency of presence (1) for each band, and n is the number of bands of primer (Weir, 1990). Jaccard dissimilarity coefficients were calculated from the matrix obtained from combined of ISSR and RAPD molecular data. The R computer statistical software was used to get dendrogram via Ward's method (Ward, 1963) for hierarchical cluster analysis. The molecular genetic variation (i.e., Nei's gene diversity (Nei, 1973), Shannon's information index (Shannon and Weaver, 1949) and percentage of polymorphic loci) among the studied beans that were divided into groups based on their origins, growth habit, and seed color were calculated with a computer program POPGENE (Yeh *et al.* 1997).

The principal component analysis (PCA) based on 72 morphological traits was performed to identify the patterns of variation within the sets of common bean accessions using the PRINCOM procedure implemented in SAS (SAS Institute 2015).

RESULTS AND DISCUSSION

Phenotypic Traits: In the present study, phenotypic characterization showed a wide variation in seed characteristics. It was determined that main seed color of 42 accessions was white, while there was also different main seed colors such as 12cream, 25 brown, 4 red, and 13 black. Moreover, it was found that secondary seed colors were white (3 genotypes), brown (12 genotypes), red (9 genotypes), purple (6 genotypes), and black (1 genotype). For seed size, the seeds were divided into very small (8.33%), small (14.58%), medium (34.38%), large (29.17 %) and very large (13.54%) groups. Based on the 100-seed weight, it is assumed that the genotypes heavier than 40g were Andean (52%), and the genotypes lighter than 40g were Mesoamerican (48%) (Singh, 2001). Indeed, Angioi *et al.* (2010) studied the bean genotypes of the Balkans and Southeast Europe where Turkey is located and confirmed that there were also Andean (55%) and Mesoamerican (45%) bean genotypes.

The degree of genetic relationship among 96 Turkish bean genotypes was determined using phenotypic and molecular markers. Stoilova *et al.* (2005) emphasized the importance of phenotypic variation in the future bean breeding programs, and phenotypic traits could be useful in discrimination in bean genotypes. Some traits highlight the distinction between bean genotypes in phenotypic characterization because more variation causing significant differences were seen in them. In another study, it was indicated that some morphological characters such as the shape and the color of the seed had a high degree of heritability (De La Cruz *et al.* 2005).

Principal Component Analysis: PCA was applied to evaluate and elucidate the variation among bean genotypes using 72 morphological traits. Using PCA based on the correlation matrix, eigenvalues, percentages of variation, and load coefficients of the first three components were calculated for all studied morphological traits. It was found that the first three principal components accounted for 27.55, 19.36, and 11.56% of the variations, respectively, and the cumulative proportion of the variation approached 58.46 % of the total variance (Table 3) that was higher than those of Morojele and Mbewe, 2015 (54.57%) and Chiorato *et. al.,* 2005 (40.66%),but lower than those of Molosiwa *et al.* 2014 (77.12%), Grahic *et al.* 2013 (67.45%), Vidak *et al.* 2015 (82.16%), and Sofi *et al.* 2014 (60.71%) who analyzed common bean accessions with fewer traits.

The first three principal components were very significant, and they were plotted graphically to demonstrate the relationship among 96 common bean accessions (Fig. 1). The first PCA demonstrated a high impact; accounting for 27.55% of the total variability. The traits contributing to this high variation were: predominant secondary seed color and distribution of

secondary color. Among those components, predominant secondary seed color was the most discriminative in common bean. The second PCA was responsible for 19.36%of the total variability. Phenotypic traits contributing to this PCA group were: main seed color, plant height, and a number of nodes on the stem. In this PCA group, main seed color accounted 65.30% of variations. According to these results, although it was determined that seed traits were more effective in the distinction of common bean genotypes in the first component of PCA, some plant traits, as well as seed characteristics were weighted the second component. The third PCA component was related to variation in flower duration.

Allele diversity and genetic relationship by molecular methods: Total 474 polymorphic bands, 358 from ISSR primers and 116 from RAPD primers, were obtained (Table 2).The average polymorphic band number and the average polymorphism in ISSR were 17.04 and 95.83%, respectively. These values in RAPD were 14.50 and 97.60%, respectively. Genetic relationships in the studied beans were compared with their Warn clustering (Figure2).

Galvan *et al.* (2003), studied the genetic relationships among 10 commercial bean cultivars with ISSR and RAPD markers and found out that 33 out of 75 bands were polymorphic from 9 ISSR primers, and 17 out of 27 bands were polymorphic from 4 RAPD primers. Marotti*et al.* (2006) revealed the genetic relationships among 20 bean genotypes with 8 ISSR primers, 7 semi-random primers and 6 RAPD primers, and determined that 110 out of 130 bands were polymorphic in ISSR method, and 46 out of 67 bands were polymorphic in RAPD method.

Higher molecular genetic diversity index values were obtained from ISSR markers than those of RAPD markers. Moreover, the origin and the number of genotypes are important in polymorphism; therefore, we observed more polymorphism in much and more divergent genotypes. ISSR and RAPD methods have had abundant and sufficient polymorphic bands in bean molecular characterization similar to the earlier studies (Galvan *et al.* 2003; Tiwari *et al.* 2005; Martins *et al.* 2006; Marotti *et al.* 2006). However, Sarikamıs *et al.* (2009) investigated the genetic relationships among 28 bean genotypes in Van province of Turkey with 12 SSR primers and found only 45 polymorphic bands which were too low.

According to genetic distance matrix (unpresented data), the most dissimilar bean genotypes were G54 (Andean, mottled seeded, pole), G37 (Andean, mottled seeded, pole), and G155 (Andean, black seeded, pole), while the least distinct ones were G33 (Mesoamerican, mottled seeded, pole), G20 (Mesoamerican, white seeded, pole), and G14 (Mesoamerican, white seeded, pole) (Figure 2). The lowest genetic distance was observed among the pair of accessions G131-G155 (0.2946), G57-G62 (0.3007) and G8-G37 (0.3083) while the highest was noted in G33-G112 (0.8432), G33-G79 (0.8187) and G33-G82 (0.8176) based on combined ISSR and RAPD analysis. The hierarchical cluster analysis separated the 96 common bean accessions into four different groups. The number of common bean accessions in a split that colored blue, red, black and green were 24, 19, 22, and 31, respectively. Each group was further divided into different subgroups (Figure 2).

Molecular genetic variation: The genetic diversity values among the genotypes in ISSR method were H=0.16 and I=0.28; in RAPD method, these were H=0.16 and I=0.26; and in the combined ISSR and RAPD methods, these were H=0.16 and I= 0.27. The genetic variation among genotypes of bean genotypes was also determined based on seed color, growth habit, and origins (Table 4). On the studied Turkish bean genotypes, there was relatively higher variation in Mesoamerican genotypes than Andean ones; there was relatively higher diversity in pole genotypes than bush ones; there was relatively higher variation in white-seed genotypes than the colored and mottled ones.

The bean genotypes were divided into two main groups based on their origins, as 50 Mesoamerican and 46 Andean genotypes (Figure 1). Genetic variation criteria, according to the origin of bean genotypes, showed that they were relatively different. The genetic variation in Andean bean genotypes was found to be relatively higher than that of Mesoamerican. In addition, the rate of polymorphism was higher in Andean genotypes than that of Mesoamerican. Moreover, for origins in bean genotypes, ISSR markers data gave a relatively higher rate of genetic variation and polymorphism values compared to RAPD markers (Table 4).

Chiorato *et al.* (2007) stated that there was relatively higher variation in Mesoamerican genotypes compared to Andean ones, which is in line with our findings. However, Zhang *et al.*(2008) determined higher variation in Andean bean genotypes. Dela Cruz *et al.* (2005) obtained genetic diversity values among the wild bean genotypes collected from different regions of Mexico as H=0.14 and I=0.29. De Meaux *et al.* (2003) determined the genetic diversity index in beans similar to the present study; however, some other researchers found relatively higher variation in their bean genotypes (Blair *et al.* 2007; Zhang *et al.*2008).

The studied beans were separated into two groups based on their growth habit, as 65 pole and 31 bush ones. Bush genotypes were approximately half of pole genotypes in number, but they had similar genetic variation value as pole ones. Similarly, for growth habit

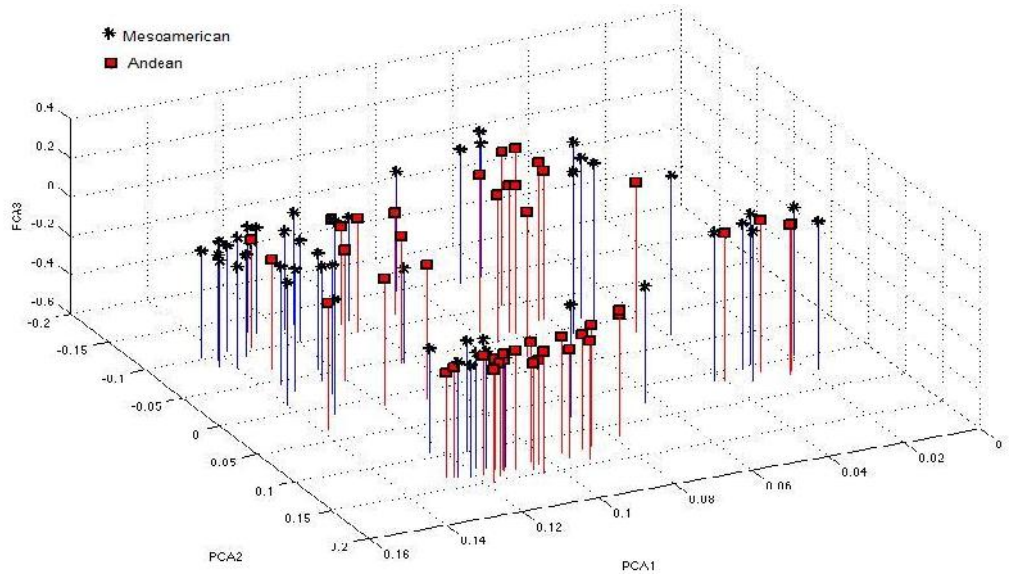

Figure 1. The first three principle components principle component analysis (PCA) plot conducted using 72 morphological characters.

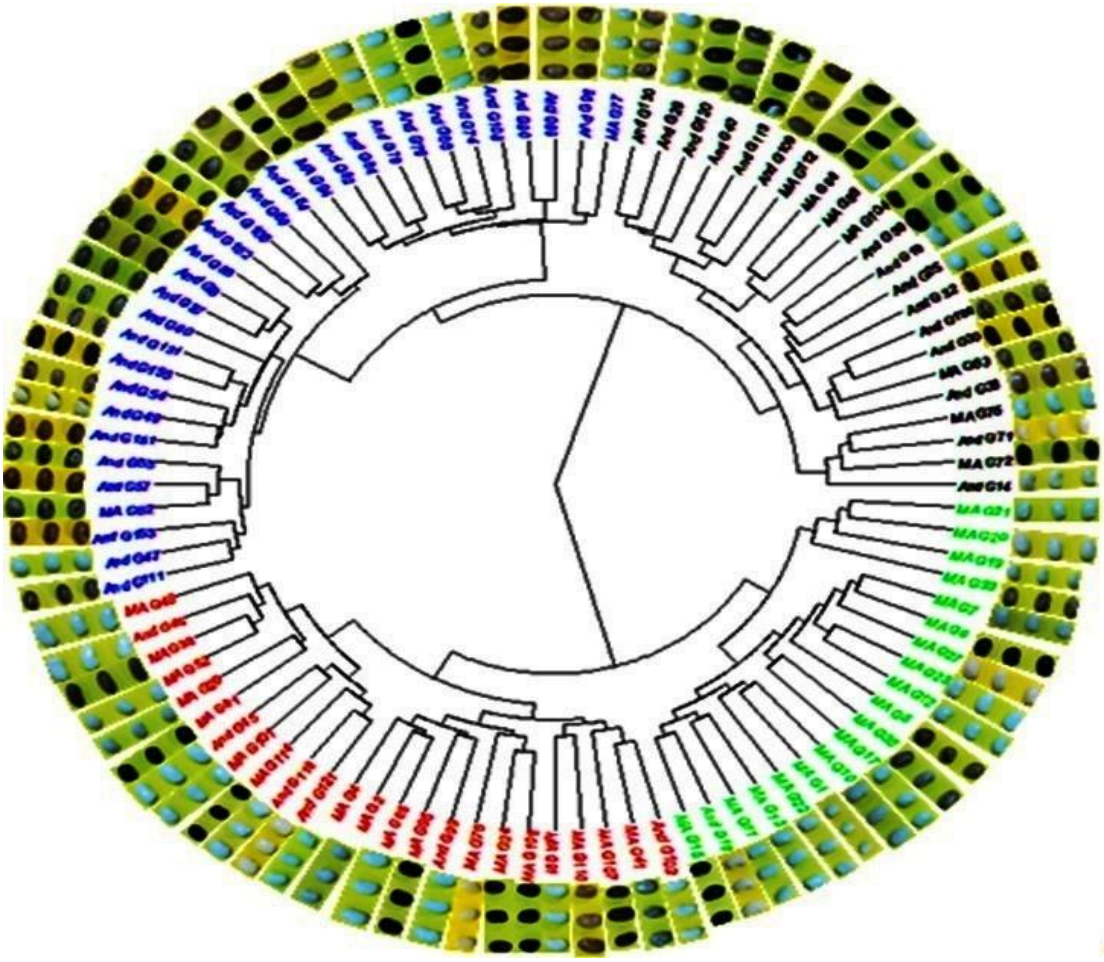

Figure 2. Associations among some Turkish bean genotypes revealed by hierarchical clustering analysis on the basis of the combined ISSR and RAPD Jaccard distance values

Table 1. Some distinguished traits of the common bean accessions used in the present study.

Acc. #		Grow Habit	Seed* color	100 seed weight (g)	Acc. #		Grow Habit	Seed color	100 seed weight (g)	Acc. #		Grow Habit	Seed color	100 seed weight (g)
G1	TR 38468(Tekirdağ)	Pole	W	36.00	G38	Sarıkız Fasulyesi	Pole	Br	33.35	G89	Yer Hammadisi- Kulcalı	Bush	BrR	40.38
G2	TR 66339(Isparta)	Pole	W	25.10	G39	Siyah Fasulye	Pole	B	38.73	G90	Sarıkız	Bush	Br	41.30
G4	TR 57755 (Kayseri)	Pole	W	35.15	G40	Ayşe Kadın	Pole	BDB	55.10	G91	Passport-I	Bush	W	38.21
G5	TR 62580 (Balıkesir)	Pole	BDB	27.27	G41	Barbunya	Pole	CV	35.80	G94	Trabzon-Zaga	Bush	B	34.51
G6	TR 39451 (Hatay)	Pole	B	31.13	G45	Tufanbeyli	Pole	W	42.60	G95	F1 101-948	Bush	B	18.73
G7	TR 35426 (Konya)	Pole	W	29.97	G46	Sırık Ayşe Kulcalı	Pole	W	54.65	G96	393Y-151-1/6	Pole	BrW	40.35
G8	TR 43862 (Balıkesir)	Pole	BDB	50.30	G47	Nohut Fasulye	Pole	W	48.15	G98	142-9 759 2-4	Bush	B	33.20
G10	TR 66751 (Bartın)	Pole	W	31.75	G48	Ayşe Beyaz	Pole	W	35.30	G99	137-15-2 1233	Pole	W	41.31
G11	TR 61436 (Bursa)	Pole	W	33.10	G49	Börülce Abant	Pole	WBV	43.00	G100	445 1/6 177	Pole	BrR	59.83
G12	TR 53827 (Kırklareli)	Pole	W	30.55	G52	Adıyaman-II	Pole	W	36.25	G101	143-6 A.B. 1244-1245	Pole	W	36.80
G13	TR 38059 (Kütahya)	Pole	W	26.80	G54	Gülnar-I	Pole	CR	40.90	G102	142-5 A.B. 1242-1243	Pole	W	42.23
G14	TR 66342 (Afyon)	Pole	W	40.73	G55	Gülnar-II (Barbunya)	Pole	CV	55.05	G104	559 2/1 667	Bush	Br	18.61
G15	TR 37990 (Amasya)	Pole	B	31.05	G57	Gülnar-III	Pole	BDB	46.90	G105	F2 23 960	Pole	BDB	44.80
G16	TR 50771 (Denizli)	Pole	W	45.90	G58	Gülnar-IV	Pole	CR	36.00	G106	F1 103 950	Bush	B	18.10
G17	TR 62848 (Isparta)	Pole	W	26.75	G60	Gülnar-VI	Pole	BV	41.47	G107	F2 25 961	Bush	Br	27.78
G18	TR 53662 (Çanakkale)	Pole	W	45.00	G62	Gülnar-VII	Pole	CR	33.53	G108	425 1/6 Y 164	Pole	CR	42.35
G19	TR 68587 (Eskişehir)	Pole	W	15.83	G63	Gülnar-VIII	Pole	CR	31.15	G109	ST43 1071	Pole	BDB	52.50
G20	TR 57749 (Kayseri)	Pole	W	29.80	G65	Fas-Agadir-Suk (Pazar-II)	Pole	W	37.05	G110	385 1-6,1-5 147	Pole	Br	25.90
G21	TR 49668 (Burdur)	Pole	W	26.86	G66	Fransa-Gandiyam	Pole	B	44.80	G111	3-2 1003	Pole	BDB	52.60
G22	TR 28035 (Denizli)	Pole	W	20.55	G71	TR 50763 (Denizli)	Bush	W	47.20	G112	F2-7 957	Bush	B	35.33
G23	TR 43477 (İstanbul)	Pole	W	33.05	G72	TR 42164 (Hatay)	Bush	B	33.06	G113	400-1/6 154	Pole	RW	43.48
G25	TR 51364 (Bolu)	Pole	W	44.50	G74	Yunus-90	Bush	W	48.70	G114	F1 104 951	Bush	B	17.13
G27	TR 31516 (Amasya)	Pole	W	27.30	G75	Nassau	Bush	W	31.02	G118	ER-128	Pole	W	40.13
G28	TR 28018 (Aydın)	Pole	W	37.83	G76	Tender	Bush	W	26.60	G120	Nadide	Bush	Br	39.63
G29	Şehirali	Pole	W	18.83	G77	Atlanta	Bush	W	26.70	G121	*P. coccineus*	Pole	W	88.73
G30	4F-89	Pole	R	44.20	G78	Yalova-17	Bush	W	35.56	G130	Önceler 98	Bush	CR	47.00
G32	Barbunya Kınalı	Pole	BrB	62.68	G79	Yalova-5	Bush	W	48.28	G131	Horoz fasulye	Pole	CR	50.60
G33	9 no'lu hat (America)	Pole	BDB	31.56	G81	5 no'lu hat (America)	Bush	B	16.93	G151	YYU-6	Pole	BrV	54.97
G34	10 no'lu hat (America)	Pole	R	30.15	G82	6 no'lu hat (America)	Bush	LR	46.57	G152	YYU-8	Pole	BrV	53.50
G35	11 no'lu hat (America)	Pole	BDB	34.93	G84	Kıbrıs Amerikan	Bush	LBr	41.50	G153	YYU-14	Pole	BDB	40.40
G36	Ayşe Kadın	Pole	BrW	50.10	G85	Barbunya	Bush	CR	60.00	G154	YYU-29	Pole	Br	39.65
G37	Kılçıksız Boncuk Ayşe	Pole	BDB	58.60	G88	İspanya	Bush	W	17.79	G155	YYU-41	Pole	B	52.60

*B: Black, BDB: Brown-Dark Brown, Br: Brown, BrB: Brown-Black, BrR: Brown-Red, BrV: Brown-Violet, BrW: Brown-White, CR: Cream-Red, CV: Cream-Violet, LR: Light red, LBr: Light Brown, R: Red, RW: Red-White, W: White, WBrV: White-Brown-Viole

Table 2. ISSR and RAPD primers used in this study.

ISSR	Sequence 5´-3´	Base number	Annealing temperature (^{0}C)	PIC value	Total/polymorphic band number
LOL-7	$(GA)_6CC$	14	44.0	0.15	11/9
LOL-8	$(GT)_6CC$	14	44.0	0.28	20/20
LOL-10	$(GAG)_3GC$	11	38.0	0.44	9/9
LOL-12	$(GTG)_3GC$	11	38.0	0.20	14/13
3F	$(GA)_8YT$	18	52.6	0.27	31/31
4F	$(GA)_8YC$	18	54.8	0.38	21/21
5F	$(GA)_8YG$	18	54.8	0.24	28/28
8F	$(AC)_8YA$	18	52.6	0.32	19/19
Sola-2	DD-$(CGA)_5$	17	56.8	0.40	12/10
Sola-3	DBH-$(CGA)_5$	18	59.0	0.44	19/19
Sola-4	VHV-$(GT)_7G$	18	55.2	0.32	20/20
Sola-5	$DBD(AC)_7$	17	51.1	0.50	11/10
Sola-6	BDB-$(CAC)_5$	18	59.7	0.34	18/15
ISSR-3	ACTGACTGACTGACTG	16	49.2	0.26	23/22
ISSR-5	VHVCTCTCTCTCTCTCTCT	19	55.9	0.42	21/21
ISSR-6	VDVGTGTGTGTGTGTGTGT	19	55.9	0.36	18/18
812	GAGAGAGAGAGAGAGAA	17	50.4	0.34	19/19
826	ACACACACACACACACC	17	52.8	0.44	14/14
889	AGTCGTAGTACACACACACACAC	23	60.6	0.35	16/15
PHV6	CCACTCTCTCTCTCTCTCT	19	56.7	0.44	11/10
PHV7	GTGGTGTGTGTGTGTGTGT	19	56.7	0.39	15/15
Total					370/358
RAPD	**Sequence 5´-3´**				
OPA-04	AGG ACT GCT C			0.36	15/14
OPA-15	ACG GAA GCC C			0.38	12/12
OPA-17	GAG CCC GAC T			0.48	8/7
OPB-05	TGC GCC CTT C			0.38	7/7
OPE-07	AGA TGC AGC C			0.46	14/14
FAG R5	GGC TGC GAC A			0.44	22/22
OPE-14	TGC GGC TGA G			0.31	18/18
OPF-01	ACG GAT CCT G			0.31	22/22
Total					118/116

Table 3. Principal component analysis (PCA) of characters associated with common bean accessions based on phenotypic characters.

	PC axis				
	1	2	3	4	5
Eigen values	17.59	12.36	7.38	4.76	2.29
Explained proportion of variation (%)	27.55	19.36	11.56	7.45	3.58
Cumulative proportion of variation (%)	27.55	46.91	58.46	65.91	69.49
Character	**Eigen vectors**				
Emergence time	0.01	-0.04	-0.19	0.08	-0.46
Start of climbing	0.06	0.19	0.08	-0.26	-0.02
Speed of climbing	0.18	0.17	0.09	0.12	-0.03
Strength of climbing	0.18	0.18	0.09	0.12	-0.02
Growth habit	0.13	0.17	0.08	0.00	-0.03
Flower duration	0.12	0.14	0.20	-0.08	-0.13
Plant architecture	0.13	0.18	0.09	-0.02	0.02
Compatibility to machine harvesting	0.06	0.09	0.03	-0.02	0.00

Plant height	0.25	0.34	0.19	0.23	0.15
Number of nodes on the stem	0.22	0.26	0.15	0.08	0.03
Harvest time of fresh bean	0.02	0.05	0.09	-0.01	-0.11
Anthocyanin coloration of hypocotyl	0.02	-0.04	0.07	0.01	-0.01
İntensity of leaf color	0.01	0.04	-0.01	0.03	-0.01
Leaf rugosity	0.00	0.05	0.04	0.05	0.04
Size of terminal leaflet	0.07	0.04	0.05	0.05	0.08
Shape of terminal leaflet	-0.01	0.03	0.03	0.03	-0.02
Shape of terminal leaflet apex	0.00	-0.03	-0.02	-0.02	-0.01
Presence of leaf at phys. maturity.	0.06	0.02	0.04	0.04	-0.11
Size of bract	-0.03	-0.02	0.13	-0.02	0.25
Color of bract	0.00	-0.02	0.05	-0.05	0.00
Shape of bract	-0.02	0.02	0.01	-0.04	0.06
Color of standard	0.07	-0.15	0.07	-0.01	0.05
Color of wing	0.06	-0.13	0.07	0.00	0.03
Wing opening status	0.08	-0.01	0.04	0.02	-0.14
Flower bud length	-0.01	0.01	-0.04	0.06	0.10
Flower bud width	0.02	0.02	-0.03	0.10	-0.05
Peduncle length	0.01	-0.08	-0.14	0.15	-0.02
Style raise	0.03	0.02	0.02	0.04	-0.05
Num. of nodes where the first flower	0.16	0.02	0.12	0.15	-0.23
Number of flower buds per bunch	-0.08	-0.03	-0.06	0.13	0.12
Number of pods per bunch	-0.08	-0.03	0.00	0.06	0.08
Status of pod on the plant (in bush types)	-0.12	-0.17	-0.07	0.06	0.01
First pod height (in bush types)	-0.16	-0.18	-0.15	0.12	0.05
Pod ground color	0.00	0.01	-0.01	-0.01	0.00
İntensity of pod ground color	-0.06	0.03	-0.04	-0.01	0.03
Pod secondary color	0.06	-0.03	-0.03	-0.05	0.05
Hue of pod secondary color	0.07	-0.03	-0.04	-0.07	0.04
Density of flecks of secondary color	0.10	-0.04	-0.06	-0.08	0.10
Brittleness	0.01	-0.01	0.01	0.00	-0.01
Stringiness of ventral suture	-0.08	0.04	0.09	-0.24	0.22
İmmature seed color	0.01	-0.03	-0.02	0.03	-0.08
Pod length	0.05	0.04	-0.01	0.22	0.21
Pod width	0.09	-0.01	-0.07	0.20	-0.07
Pod transversal width	-0.01	-0.05	-0.08	0.17	0.01
Length of pod beak	-0.02	0.03	-0.18	0.05	0.09
Curvature of pod beak	0.00	0.01	0.00	-0.06	0.11
Texture of pod surface	0.02	0.02	-0.02	0.04	0.03
Degree of pod curvature	-0.01	0.04	0.01	0.04	0.04
Shape of pod curvature	-0.02	0.02	-0.02	0.05	0.01
Apparent state of seed on pod	-0.03	0.00	0.02	-0.06	-0.02
Constrictions on pod	-0.02	-0.04	0.06	-0.07	-0.01
Seed number of per fresh pod	0.02	0.05	0.17	-0.02	0.19
Pod tip shape	0.00	0.00	0.00	0.00	-0.01
Tip shape the direction of pod	-0.06	0.02	0.10	-0.14	0.27
Shape of pod cross section	-0.04	-0.06	-0.06	0.06	-0.07
Seed size	0.15	0.01	-0.15	0.31	0.05
One hundred seed weight	0.09	-0.02	-0.12	0.21	0.06
Seed length	0.03	0.02	-0.16	0.27	0.26
Seed width	0.10	-0.03	-0.10	0.15	-0.16
Seed height	0.09	0.01	-0.08	0.12	-0.04
Seed shape	-0.08	0.01	-0.10	0.22	0.26
Degree of curvature (kidney shaped only)	-0.06	0.03	-0.08	0.13	0.16
Width of seed cross section	0.04	-0.02	-0.04	0.04	-0.13
Shape of median seed cross-section	0.07	0.02	-0.02	0.02	-0.05

Number of seed colors	0.10	-0.03	-0.06	-0.05	-0.03
Main seed color	0.24	-0.65	0.56	0.19	0.04
Predominant secondary seed color	0.65	-0.21	-0.42	-0.35	0.21
Distribution of secondary color	0.23	-0.06	-0.11	-0.09	-0.11
Veining	-0.01	0.07	-0.01	0.03	-0.03
Color of hilum ring	0.08	-0.04	-0.05	-0.02	-0.01
Seed brightness	0.01	0.01	0.02	0.01	-0.05

in bean genotypes, ISSR markers data gave a relatively higher rate of genetic variation and polymorphism compared to RAPD markers. The bean genotypes were divided into three groups based on their seed color, as 41 white-seeded ones, 25 other colored-seed ones, and 30 mottled-seed ones. It was found out that the white-colored seed bean genotypes had the highest genetic variation values. The rate of polymorphism was the highest in white-seed genotypes. For seed color in bean genotypes, ISSR markers data generally gave a relatively higher rate of genetic variation and polymorphism compared to RAPD markers (Table 4).

Table 4. The genetic variation values in bean genotypes for origins, growth habit, and seed color as measured by ISSR and RAPD, and combined ISSR-RAPD marker data.

	N[a]	ISSR			RAPD			ISSR-RAPD		
		H[b]	I[c]	Pol. %[d]	H[b]	I[c]	Pol. %[d]	H[b]	I[c]	Pol. %[d]
All genotypes	96	0.1647	0.2751	100	0.1598	0.2621	100	0.1635	0.2719	100
Origins										
Mesoamerican	50	0.1498	0.2475	84.08	0.1401	0.2313	85.34	0.1474	0.2436	84.39
Andean	46	0.1669	0.2790	93.58	0.1672	0.2705	93.10	0.1670	0.2769	93.46
Growth habit										
Pole	65	0.1612	0.2682	94.47	0.1512	0.2456	87.07	0.1588	0.2626	93.04
Bush	31	0.1607	0.2651	82.12	0.1595	0.2343	82.76	0.1604	0.2245	82.28
Seed color										
White	41	0.1688	0.2826	94.41	0.1639	0.2664	89.66	0.1676	0.2783	93.25
Colored	25	0.1524	0.2492	73.18	0.1397	0.2228	62.93	0.1493	0.2427	70.68
Mottled	30	0.1439	0.2359	74.30	0.1426	0.2332	75.86	0.1436	0.2353	74.68

[a]N= Number of genotypes in each population; [b]H= Nei's gene diversity;
[c]I= Shannon's information index; [d]: Percentage of polymorphic loci.

Conclusions: In Turkey, one of the major bean producer countries, landraces have been adapted to various ecological conditions of these regions, which have brought a natural selection and higher genetic diversity. Although Turkey is not the origin of common bean, it contains a rich genetic variation. The gene pool in the studied bean genotypes in Turkey was divided into Andean (52%) and Mesoamerican (48%) ones. Moreover, the rich genetic diversities were found to be among the populations generated by different phenotypic characteristics. The highest genetic diversity was observed in white-seeded bean genotypes, followed by other single colored ones having similar but a little bit higher genetic diversity than the mottled ones. One of the most striking results is that the most reliable results can be received with the use of a combination of phenotypic and molecular findings. In the light of the obtained data, it is thought that this studied bean gene pool will be an important genetic resource in breeding studies and will provide the convenience of germplasm management.

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