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Genetic diversity and population structure of common bean (*Phaseolus vulgaris* L.) accessions through retrotransposon-based interprimer binding sites (iPBSs) markers

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Abstract: The common bean (*Phaseolus vulgaris* L.) is an important crop throughout the world and is highly recommended for many developing countries due to its nutritional components. A retrotransposon-based DNA marker system, interprimer binding sites (iPBSs), developed from reverse transcriptase primer binding sites, was used to determine genetic diversity in common bean accessions. The genetic polymorphism and effectiveness of the marker system were evaluated. A total of 180 polymorphic bands were detected using 47 iPBS primers among 67 common bean accessions, with an average of 4 polymorphic fragments per primer. The genetic similarity between accessions was calculated using the software NTSYS-pc and ranged from 0.09 to 0.99. The average polymorphism information content value for the iPBS markers was 0.73. A model-based clustering method classified the common bean accessions into 4 populations using STRUCTURE software. The results indicated that the iPBS marker method can successfully determine the genetic diversity level in common bean accessions.

Key words: Common bean, retrotransposon, iPBS, population structure, genetic diversity

1. Introduction

The common bean (*Phaseolus vulgaris* L.) is one of the most ancient legumes and one of the five cultivated species of the genus *Phaseolus* (Broughton et al., 2003). The common bean originated in Latin America and has become a major food for human consumption (Broughton et al., 2003), providing vital nutrients such as proteins, vitamins, and minerals in diets in many developing countries, especially in Africa (Broughton et al., 2003). Globally, the annual production of green and dry beans is 17 million tons (FAO, 2010) and is almost twice that of the second most important legume, chickpea (*Cicer arietinum* L.) (Gepts et al., 2008). The common bean, a self-pollinated crop, is a true diploid (n = 11) with a small genome of 588 megabase pairs (Arumuganthan and Earle, 1991).

Transposable elements (TEs) are discrete regions of DNA that can move within genomes (Baranek et al., 2012). TEs are important for phylogenetic analysis because they can change their genomic location, creating genomic diversity (Baranek et al., 2012). Recent studies have shown that, depending on its function, a TE in a gene may be conserved in different plant species, subspecies, and cultivars (Xu and Ramakrishna, 2008). TEs can be classified into two major groups according to their mode of transposition: DNA transposons (group I), which replicate directly via a DNA intermediate, and retrotransposons (group II), which replicate through an RNA intermediary (Casacuberta and Santiago, 2003). Retrotransposons play important roles in plant genomes according to genome size (increasing genome size), structure, evolution, variable copy number, and random distribution (Kumar and Bennetzen, 1999). Furthermore, retrotransposons are the most abundant and widely distributed mobile genetic element in eukaryotic genomes and show polymorphism within and between species (Kumar and Bennetzen, 1999). Retrotransposons comprise 35% of the common bean genome (Schmutz et al., 2014). Retrotransposons can be divided into two primary groups according to the presence or absence of a long terminal repeat (LTR): LTR and non-LTR retrotransposons. LTR retrotransposons predominate in plant genomes and can be used as molecular markers due to their ubiquitous distribution, abundant copy number, high heterogeneity, and random nature of insertional polymorphisms resulting from different retrotransposon

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insertion mechanisms (Shen et al., 2011; Guo et al., 2014c). In addition, LTR retrotransposons contain highly conserved regions for primer design in the development of retrotransposon-based markers (Andeden et al., 2013). Another important feature of LTR retrotransposons is their stability over millions of years due to the nature of their insertion (Sanz et al., 2007). Several studies report that retrotransposons are a model marker method by which to characterize accessions in yeast (Bleykasten-Grosshans and Neuveglise, 2011), plants (Baranek et al., 2012), and animals (Brandt et al., 2005). Shen et al. (2011) showed that retrotransposon-based retrotransposon microsatellite amplified polymorphism (REMAP) and interretrotransposon amplified polymorphism (IRAP) markers constitute a simple technique with high reliability for the study of genetic diversity and relationships among Japanese apricot varieties. Furthermore, some studies report that retrotransposon-based markers, such as sequence-specific amplified polymorphism (SSAP) and IRAP markers, have a higher discriminatory power than standard DNA markers, such as amplified fragment length polymorphism (AFLP) and simple sequence repeat (SSR) markers, in genetic diversity analyses (Breto et al., 2001; Sensi et al., 2003; Labra et al., 2004; Kalendar et al., 2011)

Recently, a relatively new universal retrotransposon-based marker system for DNA fingerprinting, interprimer binding sites (iPBSs), was used by Kalendar et al. (2010). iPBSs play an important role in the formation of many important traits of plants (Chen and Liu, 2014). iPBS primers are designed to correspond to the conserved parts of primer binding site sequences among different LTR retrotransposon families (Monden et al., 2014). The iPBS marker method has several advantages compared with other retrotransposon markers: iPBSs can discriminate among genotypes without prior sequence knowledge and are highly reproducible due to their primer length and the high stringency achieved by the annealing temperature (Guo et al., 2014b). Indeed, this marker system has been used successfully for several genetic diversity studies in plants, such as apricot (Baranek et al., 2012), *Cicer* species (Andeden et al., 2013), and *Vitis vinifera* (Guo et al., 2014a, 2014b). Chen and Liu (2014) also reported that iPBS and start codon-targeted polymorphism markers are effective marker systems for discriminating genotypes in *Myrica rubra.*

The aim of the present study was to evaluate the population structure and genetic diversity of common bean accessions collected from seven countries using iPBS markers.

2. Materials and methods

2.1. Plant material and extraction of DNA

Sixty-seven accessions of the common bean collected from different locations in Turkey, as well as from other countries, including Bulgaria, the Netherlands, the USA, India, England, and Germany, were analyzed (Table 1). Seeds were germinated and 15-day-old young leaves from seedlings were randomly harvested for each accession, placed in an Eppendorf tube, and stored at −80 °C until use. The young leaf tissue was ground to a fine powder in 2-mL Eppendorf tubes containing 3–4 stainless steel balls (4 mm in diameter) using a TissueLyser (Technogen Co., Turkey). The powder was subjected to DNA extraction. Total genomic DNA was isolated according to the protocol of Saghai-Maroof et al. (1984), with some modifications. The DNA concentration was measured using a Qubit 2.0 Fluorometer (Thermo Fisher Scientific Inc., USA), and the quality of the isolated DNA was checked by electrophoresis through a 0.8% agarose gel. The final DNA concentration was adjusted to 30 ng/µL for iPBS analysis and the diluted DNA was stored at −20 °C for PCR analysis.

2.2. iPBS analysis

The 47 iPBS primers that were obtained from Kalendar et al. (2010) and used for genotyping are listed in Table 2. The iPBS method was conducted essentially as reported by Kalendar et al. (2010), with some modifications. PCR amplifications with these primers were performed in a 20-µL reaction mixture containing 1 unit of Taq DNA polymerase (Applied Biological Materials Inc., Canada), 20 mM $MgCl_2$ (Applied Biological Materials Inc.), 10 mM of each dNTP (Thermo Fisher Scientific Inc.), PCR buffer (Applied Biological Materials Inc.), 1 mM primer for 12–13-nt primers or 0.6 mM for 18-nt primers, and 30 ng genomic DNA. A Peltier thermal cycler (DNA Engine DYAD; Bio-Rad, USA) was programmed as follows: one cycle at 94 °C for 30 s, followed by 35 cycles of 94 °C for 25 s, 45–65 °C for 45 s (for details see Table 2), and 72 °C for 1 min, with a final extension step of 72 °C for 5 min. The thermal cycler was programmed to hold the product at 4 °C. After amplification, the PCR products and a 100-bp DNA ladder (Thermo Fisher Scientific Inc.) were separated by electrophoresis through 3% agarose gels in 1X TBE buffer for approximately 2 h at 120 V. The gels were photographed using a G-box SYNGENE gel documentation system.

2.3. Genetic diversity and population structure

Only reproducible and clearly amplified bands were scored for the construction of a data matrix. Amplification products from the 67 common bean accessions were classified as present (1) or absent (0) for the iPBS marker method described for retrotransposon-based markers by Baránek et al. (2012) and Kalendar et al. (2010). The polymorphic information content (PIC) was calculated from the iPBS data using the equation $PIC = 1 - \Sigma p_i^2$, where p_i is the proportion of the *i*th population (Anderson et al., 1993). The genetic similarity among the 67 common bean accessions was illustrated by a Jaccard similarity

Table 1. List of *P. vulgaris* accessions grouped by variety name, code number, and location.

Code number (#)	Location	Name of the variety	Code number (#)	Location	Name of the variety
1	Gölcük/Turkey	Surmeli barbunya	35	Tekirdağ/Turkey	Mora
2	Bozdağ/Turkey	Alacalı barbunya	36	Isparta/Turkey	Arba
3	Gölcük/Turkey	Şeker barbun	37	Isparta/Turkey	Taze fasulye
4	Gölcük/Turkey	Alacalı Ayşe	38	Karadeniz/Turkey	Alman ayşe
5	Gölcük/Turkey	Ege barbunya	39	Turkey	Alman sarıkız
6	Gölcük/Turkey	Elindar	40	Sarıkız/Turkey	Meksika fasulyesi
7	Bozdağ/Turkey	Yerli barbunya	41	Turkey	Kuru fasulye 13
8	Gölcük/Turkey	Ayşe kadın	42	Turkey	Volare
9	Bozdağ/Turkey	Kula barbunya	43	Turkey	Mergseed
10	Kırklareli/Turkey	Ak	44	Bulgaria	Helda
11	Kırklareli/Turkey	Melka	45	Turkey	Emergo155
12	Bandırma/Turkey	Boncuk	46	Germany	Purple teepe 141
13	Bandırma/Turkey	Sarıkız fasulye	47	Germany	Akkiz
14	Bandırma/Turkey	Ayşe kadın	48	Turkey	Roma 2
15	Bandırma/Turkey	Sarı şeker	49	Turkey	Roma 42
16	Bandırma/Turkey	Kaynarca	50	Turkey	Admires 3060
17	Gölcük/Turkey	Kuru fasulye	51	Netherlands	Kuzga
18	Gölcük/Turkey	Beyon	52	India	Dolic hos
19	Kırklareli/Turkey	Horoz	53	India	Flora
20	Kırklareli/Turkey	Manda Fasulye	54	USA	Lima
21	Bandırma/Turkey	Boncuk Ayşe	55	England	Maxi
22	Bandırma/Turkey	Hatay Oturak	56	England	Cobra
23	Bandırma/Turkey	Gino	57	England	Algarve
24	Kırklareli/Turkey	Sarıkız	58	Turkey	Magnum
25	Yalova5/Turkey	Yalova 5	59	Bursa/Turkey	Alman ayşe 5
26	Yalova17/Turkey	Yalova 17	60	Netherlands	Limka
27	Gino/Turkey	Gino 10	61	Netherlands	No:209
28	Sarıkız/Turkey	Dilme sarıkız	62	USA	Maxi bell
29	Selçuk/Turkey	Taze	63	USA	Provider
30	Tokat/Turkey	Günlük	64	USA	E-Z Pick
31	Özayşe/Turkey	Özayşe	65	USA	Fortex
32	Tire/Turkey	Piyazlık	66	Turkey	Arya
33	Antalya/Turkey	Horan	67	Turkey	Meluk
34	Karadeniz/Turkey	Ispir			

Table 2. Interprimer binding site primer sequences for genetic diversity analysis among common bean accessions.

Primer no.	iPBS primer name	Primer sequence	Length of primers (nt)	Annealing temperature °C
1	2074	GCTCTGATACCA	12	50
2	2075	CTCATGATGCCA	12	51
3	2076	GCTCCGATGCCA	12	51
4	2077	CTCACGATGCCA	12	46
5	2375	TCGCATCAACCA	12	50
6	2376	TAGATGGCACCA	12	50
7	2379	TCCAGAGATCCA	12	46
8	2380	CAACCTGATCCA	12	46
9	2381	GTCCATCTTCCA	12	48
10	2383	GCATGGCCTCCA	12	46
11	2384	GTAATGGGTCCA	12	45
12	2387	GCGCAATACCCA	12	46
13	2388	TTGGAAGACCCA	12	46
14	2389	ACATCCTTCCCA	12	46
15	2391	ATCTGTCAGCCA	12	48
16	2392	TAGATGGTGCCA	12	48
17	2393	TACGGTACGCCA	12	48
18	2394	GAGCCTAGGCCA	12	48
19	2270	ACCTGGCGTGCCA	13	48
20	2272	GGCTCAGATGCCA	13	51
21	2277	GGCGATGATACCA	13	45
22	2217	ACTTGGATGTCGATACCA	18	46
23	2218	CTCCAGCTCCGATTACCA	18	58
24	2219	GAACTTATGCCGATACCA	18	55
25	2220	ACCTGGCTCATGATGCCA	18	52
26	2221	ACCTAGCTCACGATGCCA	18	56
27	2222	ACTTGGATGCCGATACCA	18	58
28	2226	CGGTGACCTTTGATACCA	18	50
29	2228	CATTGGCTCTTGATACCA	18	60
30	2230	TCTAGGCGTCTGATACCA	18	51
31	2232	AGAGAGGCTCGGATACCA	18	56
32	2237	CCCCTACCTGGCGTGCCA	18	46
33	2238	ACCTAGCTCATGATGCCA	18	60
34	2239	ACCTAGGCTCGGATGCCA	18	58
35	2243	AGTCAGGCTCTGTTACCA	18	55
36	2244	GGAAGGCTCTGATTACCA	18	53
37	2246	ACTAGGCTCTGTATACCA	18	55
38	2249	AACCGACCTCTGATACCA	18	54
39	2251	GAACAGGCGATGATACCA	18	56
40	2252	TCATGGCTCATGATACCA	18	54
41	2253	TCGAGGCTCTAGATACCA	18	56
42	2255	GCGTGTGCTCTCATACCA	18	46
43	2395	TCCCCAGCGGAGTCGCCA	18	56
44	2398	GAACCCTTGCCGATACCA	18	56
45	2401	AGTTAAGCTTTGATACCA	18	52
46	2402	TCTAAGCTCTTGATACCA	18	52
47	2415	CATCGTAGGTGGGCGCCA	18	46

*Primers were used as in Kalendar et al. (2010).

matrix, which was constructed with the software NTSYS-pc (Numerical Taxonomy and Multiware Analysis System, version 2.0) (Rohlf, 1988).

The genetic structures of the accessions analyzed were also investigated using a model-based clustering algorithm (STRUCTURE v.2.2) that genetically separates groups according to allele frequencies (Pritchard et al., 2000). Ten runs of STRUCTURE were performed by setting the number of populations (K) from 2 to 10, except for K = 1, for which only one run was performed. Each run consisted of a burn-in period of 100,000 steps followed by 100,000 Monte Carlo Markov chain replicates, assuming an admixture model and correlated allele frequencies. The probability of the best fit into each number of assumed clusters (K) was estimated by an ad hoc statistic, ΔK, based on the rate of change in the log probability of data between consecutive K values according to Evanno et al. (2005) using STRUCTURE HARVESTER (Earl and vonHoldt, 2011).

3. Results and discussion

The iPBS technique as a universal marker is based on the single primer amplified region principle because it uses a single primer as a forward and reverse, similar to the random amplified polymorphic DNA or inter-SSR techniques (Mehmood et al., 2013). The iPBS method has been applied to various species for identifying genetic relationships (Smykal et al., 2011; Baranek et al., 2012; Gailite and Rungis, 2012; Andeden et al., 2013; Mehmood et al., 2013; Chen and Liu, 2014; Guo et al., 2014b). These markers were found to amplify efficiently for genetic diversity analyses (Baranek et al., 2012; Andeden et al., 2013) because the primers used for PCR can anneal to genomes that contain diverse LTR sequences (Kalendar et al., 2010). Nonetheless, iPBS markers have not been studied in the common bean until recently. We also calculated certain parameters such as PIC to determine the effectiveness of the discriminatory power of this marker system.

3.1. Analysis of polymorphic iPBS bands

A total of 83 iPBS primers were screened. Among these primers, 47 iPBS primers (56%) (21 12–13-nt long and 26 18-nt long) generated clear and scorable polymorphic bands in our PCR analysis (Table 2). Similar results were observed in the literature (Kalendar et al., 2010; Smykal et al., 2011; Baranek et al., 2012; Andeden et al., 2013; Mehmood et al., 2013; Chen and Liu, 2014; Guo et al., 2014b). Genetic diversity studies with iPBS markers showed that the marker system revealed an enormous number of polymorphisms in several studies. For instance, Chen and Liu (2014) used the same 18-nt primers (2220, 2243, 2244, 2238, 2249, 2237, 2253, 2255, and 2217) that we used in the present study, and these primers successfully amplified the target region. However, the remaining 36 iPBS primers did not result in any amplification in the common bean genome because the regions targeted by them are not conserved. Similarly, Guo et al. (2014b) screened 41 iPBS primers but were able to select only 15 primers with stable and clear bands, with the remainder not showing any amplification. These findings demonstrate that these iPBS primers are not conserved among common bean, grape, and several species used in Kalendar et al. (2010).

The 47 iPBS primers amplified a total of 180 polymorphic bands in the common bean genome (Table 3). Similar results about the number of bands were observed in the literature; for instance, in apricot cultivars (Baránek et al., 2012), *Cicer* species (Andeden et al, 2013), *Myrica rubra* (Chen and Liu, 2014), and grape varieties (Guo et al., 2014b). These results show that the efficiency of iPBS markers for the common bean is more conserved compared to grape (Guo et al., 2014b), *Cicer* species (Andeden et al., 2013), *Myrica rubra* (Chen and Liu, 2014), and apricot (Baranek et al., 2012). These results also confirmed that the iPBS markers utilized generated a large number of bands and the copy number was very high in the genomes studied. The sizes of the bands amplified using different primers were within the range of 100 to 2000 bp, consistent with the results reported by Chen et al. (2014) and Mehmood et al. (2013). Figure 1 shows the representative amplification results for the primer iPBS 2394.

The number of polymorphic bands per locus varied from 1 (iPBS 2375 and iPBS 2277) to 12 (iPBS 2394) using the 47 iPBS primers (21 12–13-nt iPBS primers and 26 18-nt iPBS primers) (Table 3). When comparing the characteristics of the primers, the mean number of polymorphic bands generated by the 18-nt primers was 4, whereas the mean number of polymorphic bands generated by the 12–13-nt primers was 3.5. These results showed that 18-nt primers can identify a higher degree of polymorphism than the 12–13-nt primers, which is in agreement with the results reported by Kalendar et al. (2010). Similarly, Baránek et al. (2012) used sixteen 12–13-nt primers and six 18-nt primers among apricot cultivars and reported that the average number of products generated was 6, though the average number of products generated by 18-nt primers was 5. Guo et al. (2014b) also reported that 18-nt iPBS primers were more efficient than 12–13-nt. Furthermore, based on multiple patterns of retrotransposon conservation, these genetic diversity studies show the presence of duplicate/multiple copies in the genome.

PIC is an important index that scores the usefulness of the efficiency of polymorphic loci and determines the discriminating power of a primer among accessions (Guo and Elston, 1999). PIC values varied from 0.94 for primer

Table 3. Genetic diversity parameters for 47 iPBS markers evaluated in 67 common bean accessions.

Primer no.	iPBS primer name	Number of polymorphic bands	PIC	Primer no.	iPBS primer name	Number of polymorphic bands	PIC
1	2074	2	0.55	25	2220	2	0.63
2	2075	5	0.82	26	2221	4	0.81
3	2076	4	0.78	27	2222	5	0.84
4	2077	2	0.62	28	2226	5	0.82
5	2375	1	0.03	29	2228	2	0.54
6	2376	4	0.77	30	2230	3	0.72
7	2379	3	0.73	31	2232	6	0.87
8	2380	6	0.85	32	2237	2	0.54
9	2381	2	0.54	33	2238	6	0.86
10	2383	3	0.69	34	2239	6	0.86
11	2384	2	0.55	35	2243	3	0.82
12	2387	3	0.73	36	2244	3	0.75
13	2388	3	0.76	37	2246	3	0.76
14	2389	4	0.77	38	2249	3	0.74
15	2391	4	0.77	39	2251	3	0.78
16	2392	3	0.7	40	2252	8	0.9
17	2393	4	0.77	41	2253	7	0.89
18	2394	12	0.94	42	2255	4	0.79
19	2270	5	0.81	43	2395	8	0.91
20	2272	2	0.68	44	2398	6	0.87
21	2277	1	0.43	45	2401	3	0.75
22	2217	3	0.73	46	2402	2	0.63
23	2218	2	0.62	47	2415	2	0.56
24	2219	4	0.81			Total 180	Mean 0.73

iPBS 2394 to 0.03 for primer iPBS 2375, with a mean of 0.73 (Table 3). This result showed that iPBS 2394 was the most informative and efficient primer for discriminating among common bean accessions. The average PIC values (0.73) in this study were higher than those reported by Mehmood et al. (2013) (0.24) and Guo et al. (2014b) (0.44) for iPBS markers in different accessions. Therefore, the iPBS primers used in this study of the common bean were more suitable than in guava (0.24) and grape (0.44) accessions. However, the mean PIC value is similar, which was in accordance with a similar study on *Cicer* species (Andeden et al., 2013). These studies show that various genotypes can be successfully discriminated by iPBS markers. Furthermore, the PIC values were higher than the values found in previous common bean studies using different marker systems (Guo et al., 2014b). The average PIC values (0.73) in this study were higher than those reported in common bean accessions by Nemli et al. (2014a) using plant peroxidase (POX) (0.40) and AFLP (0.51) markers (2014b), indicating that iPBS markers can reveal more polymorphism for the evaluation of genetic variation among common bean accessions. The high PIC value in this study may be due to large variation in the number of loci observed for iPBSs compared to POX (Nemli et al., 2014a) and AFLP (Nemli et al., 2014b) sequences. The PIC value of iPBS primers was higher than for other retrotransposon-based markers for distinguishing several accessions (Shen et al., 2011; Castro et al., 2012; Žiarovská et al., 2012; Andeden et al., 2013), showing that iPBS markers are informative and very reliable for use in genetic diversity analysis compared to other retrotransposon-based markers such as IRAP, REMAP, and SSAP.

3.2. Genetic diversity among common bean accessions

The genetic similarities, also referred to as gene differences, of 67 common bean accessions were constructed using a 0/1 data matrix with NTSYS-pc version 2.1 software based on 180 observed polymorphic bands (Guo et al., 2014a; Sharma and Nandineni, 2014). The genetic similarity values ranged from 0.09 to 0.99, and the mean genetic similarity value of 0.54 indicated that a wide range of polymorphism exists among these common bean accessions. The maximum genetic similarity value derived between #42 (Turkey) and #15 (Turkey) accessions (0.99) indicated that these are the two most closely related accessions; the minimum genetic similarity value derived between #52 (India) and #9 (Turkey) accessions (0.09) indicated that these accessions are highly distinct from each other. These results were similar to the findings of the

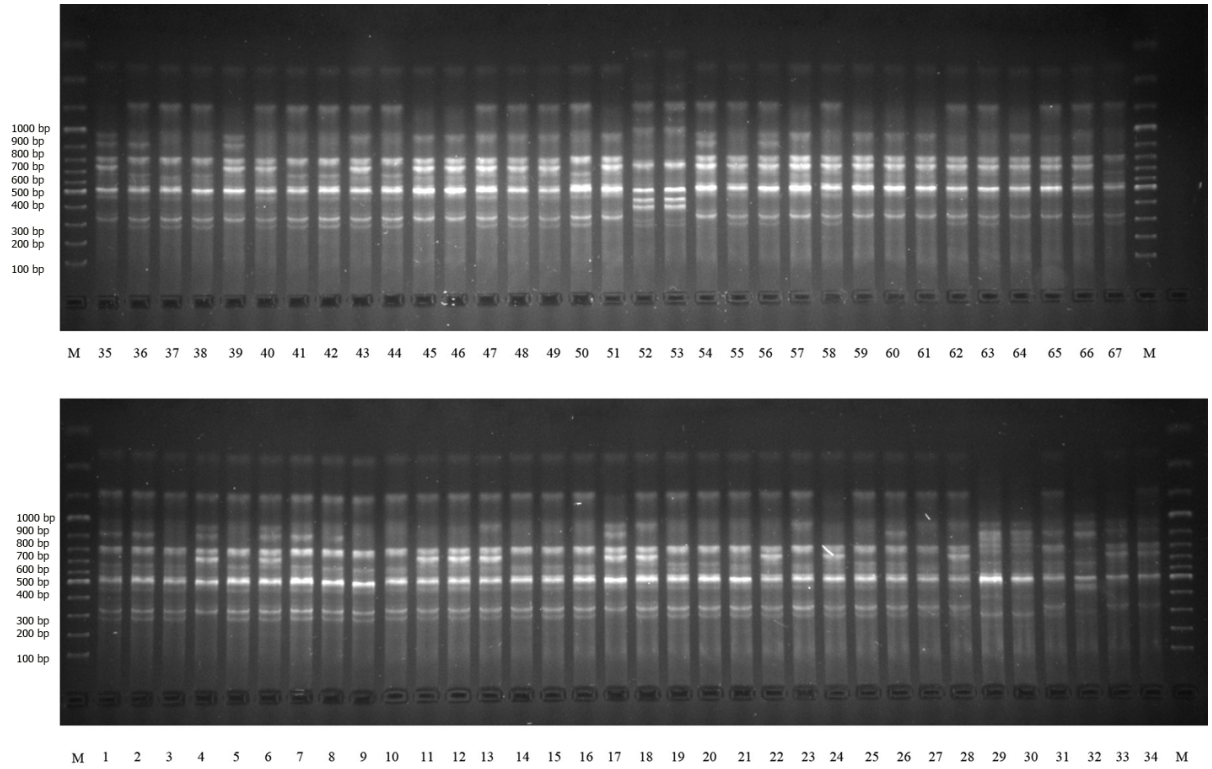

Figure 1. iPBS markers profiles of the 67 common bean accessions using primer 2394. M indicates the molecular weight standard (100–1000 bp ladder; Thermo Fisher Scientific Co.).

POX gene-based marker study conducted by Nemli et al. (2014a). iPBS markers were also used in *Myrica rubra* to detect genetic variation (Chen and Liu, 2014) that ranged between 0.55 and 0.94.

3.3. Population structure among common bean accessions

Population structure means a nonrandom distribution of the genetic diversity, which changes over time in species between groups (Hamrick and Loveless, 1989). Based on the STRUCTURE software (Figure 2), clustering analyses of the 180 polymorphic loci of the common bean accessions revealed a significant genetic diversity among the accessions. After running the K-value from 2 to 10, the highest peak was detected at K = 4, according to the ΔK criteria of Evanno et al. (2005). This implied that the 4 groups (POP I, POP II, POP III, and POP IV) were most reasonable for explaining the 67 common bean accessions (Figure 2). POP I consisted of 15 accessions [red colors: 7 accessions from Turkey (#17, 33, 34, 35, 39, 45, 59), 3 from the Netherlands (#51, 60, 61), 3 from the USA (#54, 64, 65), and 1 each from Germany (#46) and England (#57)]. Three accessions from Turkey (#29, 30, and 32) belonged to POP II. The other 47 accessions, mainly collected in Turkey (88%), were placed in POP III (blue colors: 41 from Turkey, 2 from England, 2 from the USA, 1 from Bulgaria, and 1 from Germany). The two Indian accessions were well separated in STRUCTURE (POP IV; #52 and 53) (Figure 2). This study supports the previous results of common bean genetic diversity analysis based on POX polymorphism that Indian accessions are clustered into one group (Nemli et al., 2014a). This result was related to the geographical distribution of each accession, which is a major factor in deciding the genetic diversity of varieties (Zecca et al., 2012). In general, the accessions from Turkey tended to cluster together within POPIII with the accessions from abroad. The common bean is a predominantly self-pollinated species, which limits gene flow and growth habit; compared to outcrossing species, genetic differentiation is expected to be derived more from geographical origin (Ferreira et al., 2007; Josias et al., 2010). These results were in accordance with an earlier study on common bean accessions in which genotypes from different countries classified in the same group had a similar geographical origin and similar crossing history using POX gene-based markers (Nemli et al., 2014a). Since Turkey is not a gene center for the common bean,

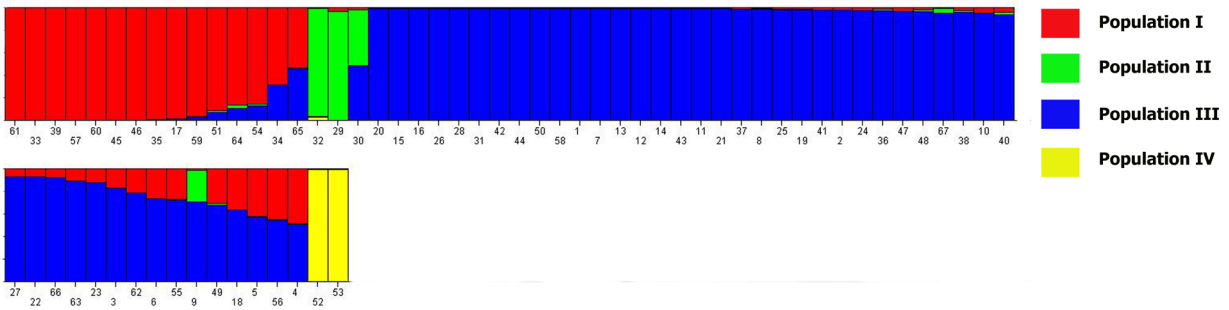

Figure 2. Population structure of 67 common bean accessions. Each population is represented by a different color (red, green, blue, yellow).

this may be due to the complex history of common bean domestication, which involves large-scale movement around the world. Therefore, it was found that Turkish accessions have been introduced from other countries. These results showed the potentiality of intraregional germplasm exchange among accessions.

3.4. Conclusion

We successfully characterized the population structure of 67 common bean accessions that were mainly collected in Turkey using iPBS markers and model-based clustering. A large degree of genetic diversity was detected among the accessions using these iPBS markers. To the best of our knowledge, this is the first study to detect genetic diversity among common bean accessions using iPBS markers. The results from the study showed that iPBS markers can be used in the assessment of genetic diversity and molecular biology analysis at the intraspecific level with great advantages, such as being robust and highly informative. The study also confirms that the iPBS marker method is an efficient method of phylogenetic diversity analysis, variety identification, and germplasm conservation. This method also could be used in plant breeding studies via map-based cloning in the common bean for future study.

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