Turkish Journal of Agriculture and Forestry

http://journals.tubitak.gov.tr/agriculture/

Research Article

Turk J Agric For
(2019) 43: 69-79
© TÜBİTAK
doi:10.3906/tar-1804-95

Characterization of quince (*Cydonia oblonga* Mill.) accessions by simple sequence repeat markers

Murat GÜNEY[1,*], Salih KAFKAS[2], Aysen KOÇ[1], Servet ARAS[1], Hakan KELEŞ[1], Harun KARCI[2]

[1]Department of Horticulture, Faculty of Agriculture, Yozgat Bozok University, Yozgat, Turkey

[2]Department of Horticulture, Faculty of Agriculture, Çukurova University, Adana, Turkey

Received: 20.04.2018 • **Accepted/Published Online:** 15.10.2018 • **Final Version:** 06.02.2019

Abstract: In Turkey, quince (*Cydonia oblonga* Mill.) is found as both a wild and a cultivated tree species. The Karanlıkdere Valley is a pit area between Şefaatli and Yerköy districts in Yozgat Province that originates from the River Delice. Members of the Yozgat Karanlıkdere Valley quince population, containing 17 quince accessions and 15 commercial quince cultivars, were DNA fingerprinted using 30 simple sequence repeat (SSR) primers to identify the genetic relationships among them. A total of 111 alleles were detected for all 32 accessions, and the number of alleles revealed by SSR analysis ranged from 2 to 10 alleles per locus with a mean value of 3.70 alleles per locus. The Ms06g03 primer gave the highest number of alleles (*Na* = 10). Polymorphism information content (*PIC*) values ranged from 0.20 (CH05e04) to 0.78 (Ms06g03) with a mean *PIC* value of 0.45. Structure analysis and unweighted pair group method with arithmetic average (UPGMA) clustering of the accessions depicted three major clusters, where several pairs of accessions could not be separated. This study indicated that the SSR markers could be utilized as a reliable tool for the determination of genetic variations and relationships of quince accessions. Furthermore, the results of this study will be useful for starting a cross-breeding cultivar program for quince.

Key words: Quince, *Cydonia oblonga*, simple sequence repeat, structure analysis

1. Introduction

Quince (*Cydonia oblonga* Mill.) is a pome fruit, like apple and pear, that belongs to the family Rosaceae. It originated in northern Iran, the Hazar Sea, South Caucasus, Khurasan, and Anatolia. Its fruit is mainly used by the food industry to produce jam, jelly, and marmalade. Quince is a good source of minerals, vitamins (especially vitamin C), and sugars (Bucsek et al., 1996) as well as flavonoid compounds, such as quercetin, rutin, and kaempferol (Silva et al., 2002, 2005). The total global production of quince has reached about 677,949 metric tons, and Turkey is one of the main producer countries, along with Uzbekistan and China, and is responsible for about 19% of the total global production (Faostat, 2016).

In Turkey, most of the quince cultivars originated from seedlings accidentally grown in backyards or at the borders of orchards (Özbek, 1978). There are still many natural quince accessions in different regions of Turkey that are valuable genetic resources for quince breeding. All quince cultivars and accessions belong to one species in the genus *Cydonia*. Identification and characterization of quince accessions morphologically are considerably difficult due to high similarities in the tree structure and fruit traits of quince plants (Yamamoto et al., 2004). Identification of relationships based on morphological characteristics has been widely used in many species including walnut (Keles et al., 2014), cherry (Rakonjac et al., 2010), and olive (Cantini et al., 1999). However, morphological characterization does not perfectly reveal the relationship due to the influence of environmental factors and low heritability (Cadee, 2000; Bucheyeki et al., 2009). DNA-based molecular markers have been effective tools to characterize plant materials for the last several decades (Lacis et al., 2009). Simple sequence repeats (SSRs) are a marker of choice due to their codominant nature, abundance in the genome, suitability for automation, high polymorphism, and repeatability (Kacem et al., 2017). One of the main advantages of SSR markers is their transferability between closely related species (Schlotterer and Tautz, 1992). The use of SSR markers for molecular characterization is well proven in different species, such as apple (Gasi et al., 2016), apricot (Hormaza, 2002), peach (Bouhadida et al., 2007), pear (Fan et al., 2013), pistachio (Zaloglu et al., 2015), and walnut (Topcu et al., 2015). Therefore, the use of SSR markers to determine the relationships among quince accessions can be highly reliable.

* Correspondence: murat.guney@bozok.edu.tr

This work is licensed under a Creative Commons Attribution 4.0 International License.

Information on genetic diversity among quince accessions is a prominent prerequisite for future breeding studies and therefore it is necessary to study genetic relationships among them. Here we report the use of SSRs for molecular characterization of the collection of quince accessions that originated in the Delice River and were maintained in Sefaatli and Yerkoy districts of Yozgat Province in Turkey; these accessions include local accessions and commercial cultivars. The main objectives were to determine the genetic relationships among the quince accessions with the expectation that the results may improve the knowledgebase on the level of diversity of the regional collection of quince and will be useful for the conservation and management of these genetic resources.

2. Materials and methods

2.1. Plant material

A total of 32 quince accessions, 17 from Yozgat Province Karanlıkdere Valley (six accessions from Yerkoy District and eleven accessions from Sefaatli District) and 15 commercial quince cultivars collected from different geographical origins in Turkey, were used in the current study (Table 1).

2.2. DNA extraction

Total genomic DNA was extracted from fresh leaves by the CTAB method described by Doyle and Doyle (1987) with minor modifications (Kafkas et al., 2006). The DNA yield was assessed by Qubit Fluorometer (Invitrogen) based on the manufacturer's instructions. Isolated DNA was subsequently diluted to 10 ng/µL for SSR-PCR reactions and stored at –20 °C.

2.3. SSR-PCR reactions

One hundred SSR primers derived from apple and pear were tested for amplification and polymorphism in eight quince accessions. Finally, 30 SSR primers were selected for characterization of the 32 quince accessions (Table 2).

All SSR-PCR reactions were carried out based on a three-primer strategy according to Scheulke (2000) with minor modifications. The reactions were done in a total volume of 12.5 µL containing 10 ng of DNA; 75 mM Tris–HCl (pH 8.8); 20 mM $(NH_4)_2SO_4$; 2.0 mM $MgCl_2$; 0.01% Tween 20; 200 µM of each dNTP; 10 nM M13 tailed forward primer at the 5′ end; 200 nM reverse primer; 200 nM universal M13 tail primer (5′-TGTAAAACGACGGCCAGT-3′) labeled with FAM, VIC, NED, or PET dye; and 0.6 U of Taq DNA polymerase. PCR amplifications were done in two consecutive steps. The first step involved initial denaturation at 94 °C for 3 min, followed by 28 cycles at 94 °C for 30 s, 58 °C for 45 s, and 72 °C for 60 s. The second step included 10 cycles at 94 °C for 30 s, 52 °C for 45 s, and 72 °C for 60 s, and a final extension at 72 °C for 5 min. When the PCRs were completed, the reactions were subjected to denaturation for capillary electrophoresis in an ABI 3130xl genetic analyzer (Applied Biosystems Inc.

Table 1. The origins of quince accessions and commercial quince cultivars used in this study, (*) Refers to cultivar.

Accession/Cultivar	Location	Accession/Cultivar	Location
*Bencikli	Yalova	Yerkoy2	Yerkoy/Yozgat
*Altın	Yalova	Yerkoy3	Yerkoy/Yozgat
*Ekmek1	Yalova	Yerkoy4	Yerkoy/Yozgat
*Beyazayva	Kocaeli	Yerkoy5	Yerkoy/Yozgat
*Limon	Yalova	YerkoySinasi	Yerkoy/Yozgat
*Sekergevrek	Yalova	Sefaatli1	Sefaatli/Yozgat
*Demir1	Kocaeli	Sefaatli2	Sefaatli/Yozgat
*Bardak	Bursa	Sefaatli3	Sefaatli/Yozgat
*SapancaEsme	Sakarya	Sefaatli4	Sefaatli/Yozgat
*Ekmek2	Bursa	Sefaatli5	Sefaatli/Yozgat
*Viranyadevi	Yalova	Sefaatli6	Sefaatli/Yozgat
*Tekkes	Yalova	Sefaatli7	Sefaatli/Yozgat
*Havan	Yalova	Sefaatli8	Sefaatli/Yozgat
*Gordes	Yalova	Sefaatli9	Sefaatli/Yozgat
*Esme1	Sakarya	Sefaatli10	Sefaatli/Yozgat
Yerkoy1	Yerkoy/Yozgat	Sefaatli11	Sefaatli/Yozgat

Table 2. Summary of SSR primers developed from apple and pear tested in quince accessions, (*) SSRs isolated from pear.

No.	Origin and reference	Acronyms	No. of tested primer	No. of nonamplified primer	Monomorphic	Polymorphic	Transferability rate (%)	Polymorphic rate (%)
1	Guilford et al. (1997)	NZ	4	-	-	4	100	100
2	Hokanson et al. (1998)	GD	3	1	2	-	66.7	-
3	Oddou et al. (2001)	MSS	1	-	1	-	100	-
4	Liebhard et al. (2002)	CH, MS	34	7	18	9	79.4	26.5
5	Yamamoto et al. (2002a)	KA	1	-	1	-	100	-
6	Yamamoto et al. (2002b)	*NH	7	-	4	3	100	42.9
7	Hemmat et al. (2003)	GD	2	-	2	-	100	0
8	Silfverberg-Dilworth et al. (2006)	Hi	19	3	12	4	84.2	21.1
9	Fernandez et al. (2006)	EMPc	6	-	4	2	100	33.3
10	Inoue et al. (2007)	IPPN	1	-	1	-	100	-
11	Khan et al. (2007)	CH	1	-	1	-	100	-
12	Han et al. (2009)	BAC	3	-	1	2	100	66.7
13	Han et al. (2011)	CN, CTG	17	3	8	6	82.4	35.3
14	Moriya et al. (2012)	MEST	1	-	1	-	100	-
	Total		100	14	56	30	86	30

(ABI), Foster City, CA, USA) using a 36-cm capillary array with POP7 as the matrix (ABI). Denaturation of the samples was done by mixing 0.5 µL (in 6-FAM and VIC labeled primers) or 1.0 µL (in NED and PET labeled primers) of the amplified product with 0.3 µL of the size standard and 9.7 µL of Hi-Di formamide. The fragments were resolved using the ABI data collection software 3.0; SSR fragment analysis was performed using GeneMapper 4.0 (ABI).

2.4. Data analysis

After capillary electrophoresis of the SSR, the number of alleles per locus (*Na*), effective number of alleles (*Ne*), expected heterozygosity (*He*), and observed heterozygosity (*Ho*) were calculated using the program GenAlEx version 6.5 (Peakall and Smouse, 2012). Polymorphism information content (*PIC*) of each locus was calculated using the software PowerMarker version 3.25 (Liu and Muse, 2005). A dendrogram was obtained using the band similarity coefficient in NTSYSpc v2.21c (Rohlf, 2009) software by an unweighted pair-group method with arithmetic averages (UPGMA). Identification of population structure and admixed individuals was performed using the model-based software STRUCTURE 2.3.4 (Pritchard et al., 2000). In this model, a number of populations (K) are assumed to be present, each of which is characterized by a set of allele frequencies at each locus. Individuals in the sample are assigned individually to populations (clusters) or jointly to more populations if their accessions indicate that they are admixed. Ln P (D) values (logarithm probability for each K) were used for determination of delta K (ΔK), which, in turn, determines probable population number. Delta K is a term that is calculated by the change in the ratio of logarithm probability (ΔK = 2 to 10). The highest value of K in delta K provides information about probable population number.

3. Results

A total of 100 SSR primers were screened to find polymorphic SSRs in quince from which 30 SSR primer pairs generated scorable and polymorphic bands. These were then used for fingerprinting of 17 accessions and 15 commercial quince cultivars. The electropherogram of the CH05c06 locus is given in Figure 1.

3.1. Polymorphism levels of SSR loci

Of the 100 SSR primers screened here, 14 failed during amplification and 56 were monomorphic, while the 30 remaining SSR primers generated polymorphic alleles when tested in the eight quince cultivars and accessions. These 30 primers were then used for characterization of quince accessions (Table 2). The transferability rate of the 100 SSR loci was 86%, while the polymorphism rate was 34.9% in the amplified loci.

A total of 111 alleles were detected for all 32 accessions, and the number of alleles ranged from 2 to 10 alleles per locus (Table 3) with a mean value of 3.70. The Ms06g03 locus produced the highest number of alleles (*Na* = 10). The number of effective alleles (*Ne*) ranged from 1.29 (CH05e04) to 5.16 (Ms06g03) with a mean of 2.35. The observed heterozygosity (*Ho*) varied from 0.00 to 1.00 with a mean of 0.58. Observed heterozygosity (*Ho*) was

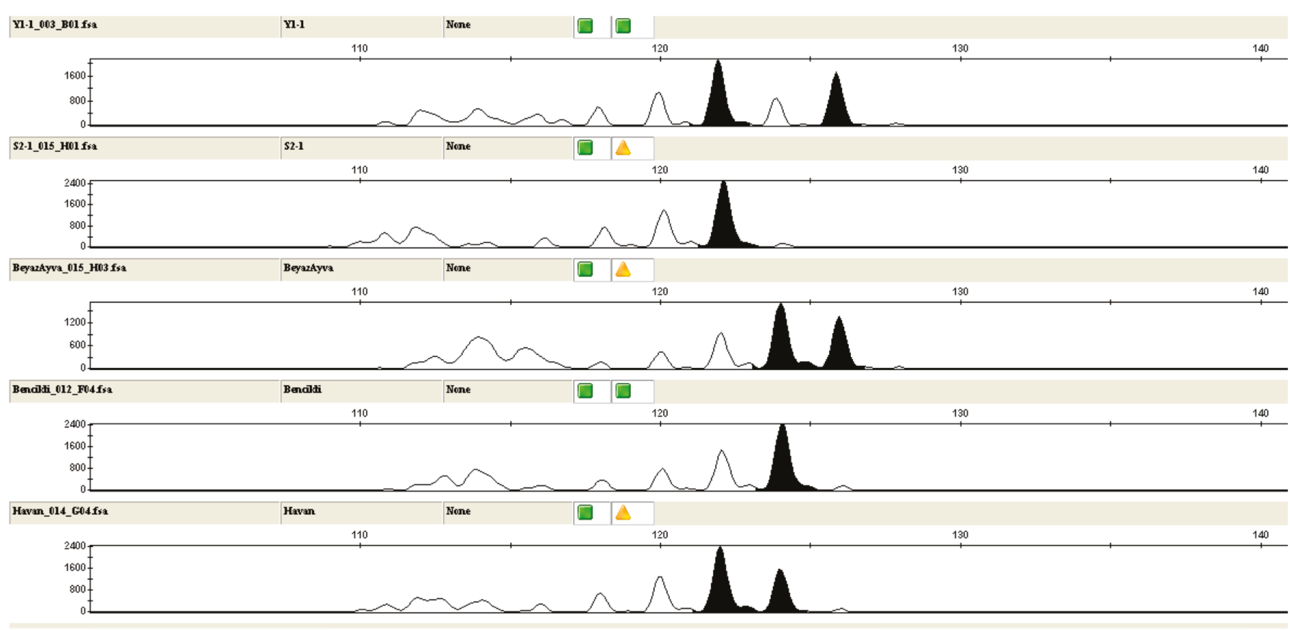

Figure 1. Electropherogram of CH05c06 SSR locus in five *C. oblanga* accessions.

Table 3. Allele range, number of alleles (*Na*), number of effective alleles (*Ne*), observed heterozygosity (*Ho*), expected heterozygosity (*He*), and polymorphism information content (*PIC*) values based on 30 SSR primers developed from *C. oblanga* accessions, (*) SSRs isolated from pear.

No.	SSR loci	Allele ranges	*Na*	*Ne*	*Ho*	*He*	*PIC*
1	Hi02d04	257–269	5	3.80	1.00	0.74	0.70
2	EMPC117	104–115	4	2.87	0.97	0.65	0.59
3	*NH007b	137–152	4	2.56	0.47	0.61	0.53
4	*NH001c	214–223	3	1.94	0.17	0.49	0.38
5	Ms06g03	170–190	10	5.16	0.97	0.81	0.78
6	CH02c02a	147–161	5	4.15	1.00	0.76	0.72
7	CN884916	295–299	2	1.84	0.65	0.46	0.35
8	Hi03e03	208–212	2	1.40	0.22	0.28	0.24
9	CN893899	115–124	3	2.68	0.97	0.63	0.55
10	Nz28f4	111–126	4	3.90	1.00	0.74	0.70
11	CH04g09	177–203	6	1.44	0.28	0.31	0.29
12	CTG1068442	247–252	3	2.04	0.69	0.51	0.39
13	CH01d01	200–211	3	1.68	0.55	0.41	0.34
14	CH05e04	175–183	2	1.29	0.19	0.22	0.20
15	Hi15a13	234–248	2	1.93	0.74	0.48	0.37
16	CH02g01	221–233	3	1.96	0.39	0.49	0.42
17	CH03d02	254–266	2	1.52	0.44	0.34	0.28
18	BACSSR3	103–114	4	2.94	0.97	0.66	0.60
19	CN872071	170–184	3	1.99	0.69	0.50	0.44
20	Nz26c6	208–227	6	2.81	0.41	0.64	0.61
21	*NH011b	208–227	5	1.98	0.48	0.49	0.47
22	CH05d11	186–194	3	1.45	0.38	0.31	0.27
23	CN918070	274–288	4	3.10	0.55	0.68	0.62
24	CTG1072881	147–194	5	2.93	0.97	0.66	0.59
25	Hi08e06	212–250	6	1.31	0.26	0.24	0.23
26	EMPC11	155–161	3	2.95	0.84	0.66	0.59
27	Nz04f3	123–125	2	1.82	0.31	0.45	0.35
28	Nz02b1	288–303	2	1.52	0.38	0.34	0.28
29	CH05c06	122–126	3	1.79	0.41	0.44	0.40
30	BACSSR4	303–305	2	1.69	0.00	0.41	0.32
	Total	-	111	-	-	-	-
	Min	-	2	1.29	0.00	0.22	0.20
	Mean	-	3.70	2.35	0.58	0.51	0.45
	Max	-	10	5.16	1.00	0.81	0.78

highest for the Ms06g03, CH02c02a, Hi02d04, and Nz28f4 loci. The average value of expected heterozygosity (*He*) was 0.51 with the highest value (0.81) observed for the Ms06g03 locus. *PIC* ranged from 0.20 (CH05e04) to 0.78 (Ms06g03) with an average value of 0.45 (Table 3).

3.2. Genetic relationships among quince accessions

The dendrogram generated from the UPGMA algorithm is depicted in Figure 2. Genetic similarity coefficients ranged from 0.52 to 1.00. The UPGMA clustering pattern grouped all the accessions into three major clusters (Figure

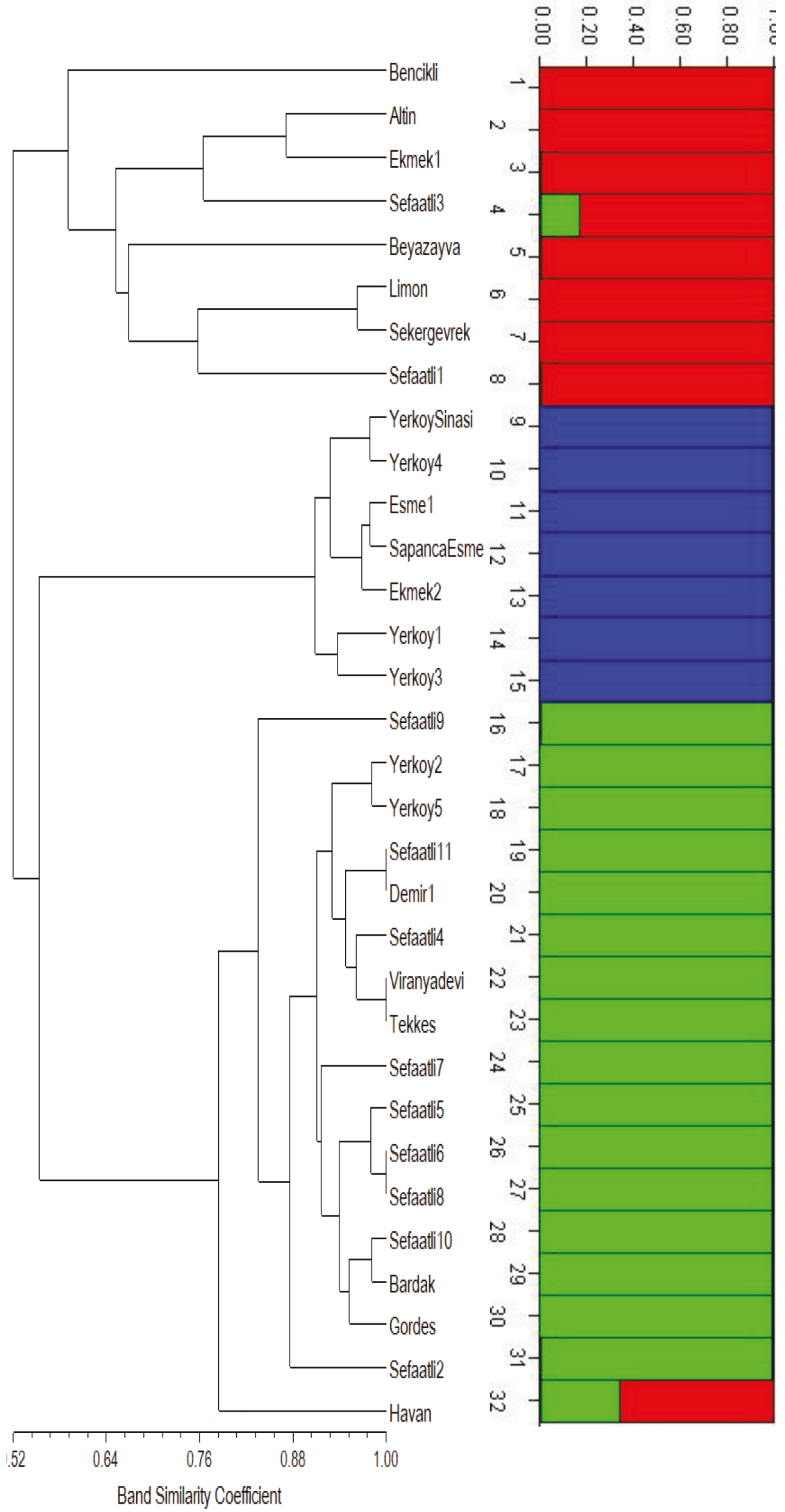

Figure 2. UPGMA and STRUCTURE (ΔK = 3) analysis of 32 *C. oblanga* accessions.

2). In Cluster-I, the highest genetic similarity coefficient (0.96) was obtained between the Limon and Sekergevrek commercial quince cultivars. The lowest genetic similarity coefficient (0.60) was obtained between the Bencikli and Limon commercial quince cultivars. In Cluster-II, Esme-1 and YerkoySinasi were closely related accessions to SapancaEsme and Yerkoy-4, respectively, with 0.98 genetic similarity coefficients. The lowest genetic similarity coefficient (0.88) was obtained between SapancaEsme and Ekmek-2.

Cluster-III was mainly divided into two subclusters: the first subcluster included accessions from the Sefaatli location along with Gordes and Bardak, while the second subcluster contained accessions from both locations together with Tekkes, Demir-1, and Viranyadevi. The Sefaatli-2, Sefaatli-9, and Havan accessions were in the outgroup of cluster-III. Several pairs of accessions could not be separated in cluster-III, including Demir-1 with Sefaatli-11, Viranyadevi with Tekkes, and Sefaatli-6 with Sefaatli-8. The lowest genetic similarity coefficient (0.71) was obtained between Havan and Sefaatli-9.

3.3. Structure analysis

The structural genetic analysis was performed in 32 quince accessions using 111 SSR primers by the programs STRUCTURE and STRUCTURE HARVESTER. The highest value of ΔK was found to be 3 (Figure 3), which corresponded to the most probable number of populations in the study. Thus, all accessions were divided into three main clusters similar to the UPGMA analysis results (Figure 2). At ΔK = 3, accessions present in Cluster-I, Cluster-II, and Cluster-III possessed mutual alleles within their clusters. Furthermore, at ΔK = 3, the 4th and 32nd accessions had mutual alleles within their own clusters and outside of them as well (except Cluster-III) (Figures 2 and 4). The dendrograms of these accessions' relationships were very similar to the structural genetic analysis (Figure 2).

4. Discussion

4.1. SSR polymorphism

Studies on molecular characterization of quince accessions are limited in the literature (Sanchez, 1988; Yamamoto et al., 2004; Dumanoglu et al., 2009; Bayazit et al., 2011; Azad et al., 2013; Yuksel et al., 2013; Topcu et al., 2015; Pınar et al., 2016). The SSR technique has several advantages, such as high polymorphism, codominance, and reproducibility. In the present study, we present a few polymorphic SSR markers for quince, which were developed from apple and pear (Table 3). Transferability of the tested SSR markers was relatively high. The transferability of apple SSRs across the family Rosaceae was studied by Gasic et al. (2009) and ranged from 25% in apricot to 59% in pear. Yamamoto et al. (2004) tested 118 SSRs from pear and apple and 77 of these (65%) were transferable to quince. The transferability ratio was higher in our study than in the above studies. The rate of polymorphism of transferable SSRs in quince has not been observed to be high in different studies. For

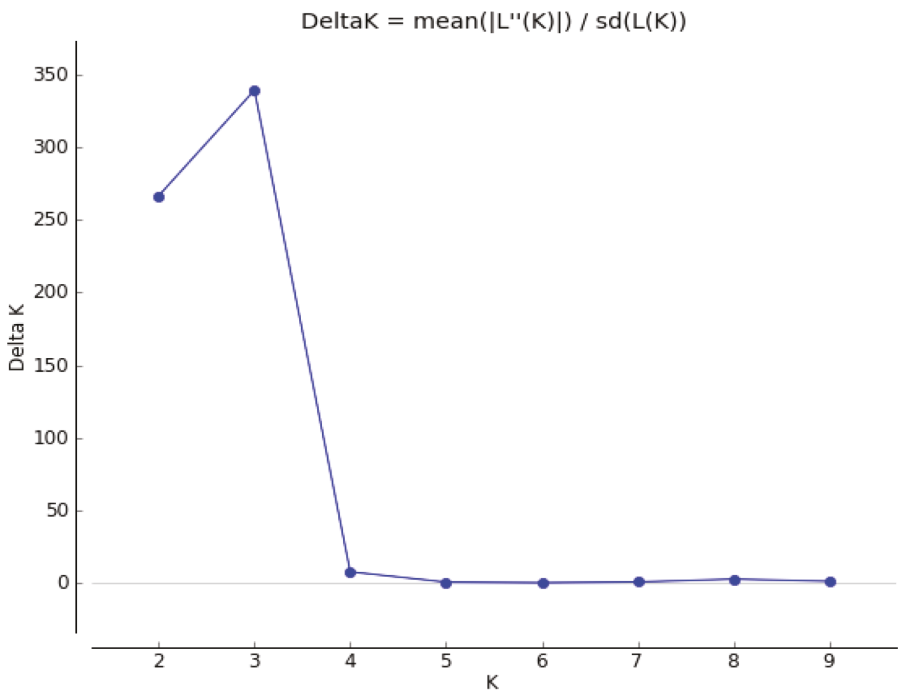

Figure 3. Value of ΔK estimated for the structure analysis of 32 *C. oblanga* accessions (ΔK = 3).

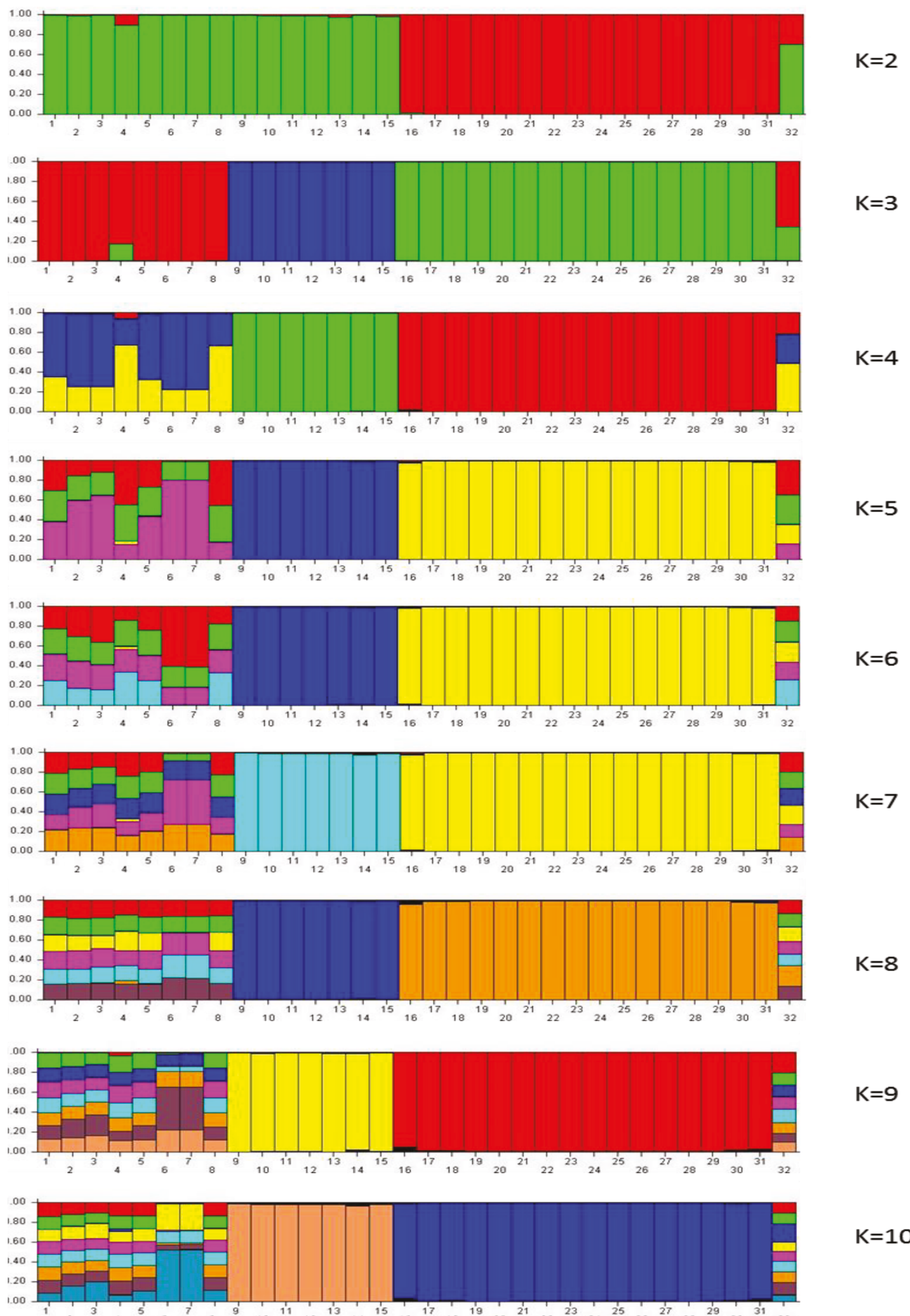

Figure 4. Population structure of 32 *C. oblanga* accessions estimated from 30 SSRs using STRUCTURE (ΔK = 2 to ΔK = 10).

example, it was reported to be 28.5% by Yamamoto et al. (2004) and we calculated it to be 30% in the present study.

Several studies revealed that SSR loci derived from apple and pear can be used to perform molecular fingerprinting of species in the family Rosaceae, such as *Cydonia oblonga* Mill. For example, Yamamoto et al. (2004) obtained a total of 122 alleles using 39 SSR primers by characterizing 20 quince genotypes. Dumanoglu et al. (2009) detected 20 alleles with a mean value of 2.85 from seven SSR primers by fingerprinting six quince genotypes. Yuksel et al. (2013) tested eight SSR primers for 15 quince genotypes and obtained 44 alleles with a mean value of 5.5. Moreover, in an experiment by Azad et al. (2013), 13 SSR primers were tested for 40 quince genotypes and 73 alleles were detected with a mean value of 5.4. In the present study, 111 alleles with an average value of 3.7 alleles per primer were detected by characterizing 32 quince accessions, including three SSRs derived from pear and 27 SSRs from apple.

In the current study, the *PIC* values were between 0.20 and 0.78 with an average value of 0.45, while Azad et al. (2013) reported a *PIC* value of 0.76. In our study, *He* and *Ho* were 0.51 and 0.58, respectively, while Azad et al. (2013) reported them to be 0.69 and 0.78, respectively; Dumanoglu et al. (2009) reported values of 0.51 and 0.65, and Yuksel et al. (2013) reported 0.62 and 0.77, respectively.

4.2. Genetic relationships among quince accessions

Structural genetic analysis based on SSR and UPGMA clustering produced similar results on genetic relationships of quince accessions. Although all the accessions were collected from different geographical locations as different accessions, the analyses demonstrated that several of them may be clones of known cultivars or other accessions. For example, it is possible that Sefaatli-11 is a clone produced from the Demir-1 cultivar, while Sefaatli-6 and Sefaatli-8 are the same clones. Although there was no polymorphism between Viranyadevi and Tekkes in the present study, they were found to be genetically distant cultivars in previous studies (Yuksel et al., 2013; Topcu et al., 2015; Pinar et al., 2016). These results provide proof for the elimination of mislabeled or misgrafted accessions in the germplasm. It is also possible that the SSRs used in this study were not sufficient to distinguish between the three pairs of accessions here.

The Yerkoy-2 accession was close to Yerkoy-5, and the Sefaatli-10 cultivar was close to Bardak, with both pairs having a 0.98 genetic similarity index (Table 1; Figure 2). Furthermore, the Sefaatli-1 and Sefaatli-3 accessions were found to be closely related to six commercial cultivars (Bencikli, Altın, Ekmek1, Beyazayva, Limon, and Sekergevrek) in cluster-I.

4.3. Conclusions

The SSR primers developed from apple and pear were found to be highly transferable to quince. However, the level of polymorphism was somewhat lower than expected. Therefore, it is necessary to screen a large number of SSR primers from apple and pear or to develop novel SSRs from quince to have highly polymorphic SSRs for further genetic studies in quince. Such SSRs are still a marker of choice in germplasm characterization, parental identification, and population genetics due to their codominant nature and reproducibility. Turkey has quince varieties that are rich in genetic resources, and thus germplasm characterization is an essential requirement for effective cross-breeding programs for this species in the future.

Acknowledgment

The authors thank the Yozgat Bozok University Scientific Research Projects Unit (Project No. 6602c-ZF/17-98) for its financial support.

References

Azad MK, Nasiri J, Abdollahi H (2013). Genetic diversity of selected Iranian quinces using SSRs from apples and pears. Bioch Genet 5: 426-442.

Bayazit S, Imrak B, Küden A, Güngör MK (2011). RAPD analysis of genetic relatedness among selected quince (*Cydonia oblonga* Mill.) accessions from different parts of Turkey. Hort Sci 38: 134-141.

Bouhadida M, Casas AM, Moreno MA, Gogorcena Y (2007). Molecular characterization of Miraflores peach variety and relatives using SSRs. Sci Hortic 111: 140-145.

Bucheyeki TL, Gwanama C, Mgonja M, Chisi M, Folkertsma R, Mutegi R (2009). Genetic variability characterisation of Tanzania sorghum landraces based on simple sequence repeats (SSRs) molecular and morphological markers. Afr Crop Sci J 17: 71-86.

Bucsek MJ, Nyeki J, Szabo Z, Kadar A (1996). Quantitation of mineral elements of different fruit pollen grains. Mikrochim Acta 13: 333-338.

Cadee N (2000). Genetic and environmental effects on morphology and fluctuating asymmetry in nestling barn swallows. J Evol Biol 13: 359-370.

Cantini C, Cimato A, Sani G (1999). Morphological evaluation of olive germplasm present in Tuscany region. Euphytica 109: 173-181.

Doyle JJ, Doyle JL (1987). A rapid isolation procedure for small quantities of fresh leaf tissue. Phytochem Bull 19: 11-15.

Dumanoğlu H, Güneş NT, Aygün A, San B, Akpınar AE, Bakır M (2009). Analysis of clonal variations in cultivated quince (*Cydonia oblonga* 'Kalecik') based on fruit characteristics and SSR markers. New Zeal J Crop Hort 37: 113-120.

Fan L, Zhang M, Liu Q, Li L, Song Y, Wang L, Zhang S, Wu J (2013). Transferability of newly developed pear SSR markers to other Rosaceae species. Plant Mol Biol Rep 31: 1271-1282.

Faostat (2016). Agriculture data [online]. Accessed October 2017. http://faostat.fao.org.

Fernandez-Fernandez F, Harvey NG, James CM (2006). Isolation and characterization of polymorphic microsatellite markers from European pear (*Pyrus communis* L). Mol Ecol Notes 6: 1039-1041.

Gasi F, Kanlic K, Stroil BK, Pojskic N, Asdal A, Rasmussen M, Kaiser C, Meland M (2016). Redundancies and genetic structure among *ex situ* apple collections in Norway examined with microsatellite markers. HortScience 51: 1458-1462.

Gasic K, Han Y, Kertbundit S, Shulaev V, Iezzoni AF, Stover EW, Bell RL, Wisniewski ME, Korban SS (2009). Characteristics and transferability of new apple EST-derived SSRs to other Rosaceae species. Mol Breed. 23: 397-411.

Guilford P, Prakash S, Zhu JM, Rikkerink E, Gardiner S, Bassett H, Forster R (1997). Microsatellites in *Malus* × *domestica* (apple): abundance, polymorphism and cultivar identification. Theor Appl Genet 94: 249-254.

Han Y, Chagne D, Gasic K, Rikkerink EHA, Beever JE, Gardiner SE, Korban SS (2009). BAC-end sequence-based SNPs and Bin mapping for rapid integration of physical and genetic maps in apple. Genomics 93: 282-288.

Han Y, Zheng D, Vimolmangkang S, Khan MA, Beever JE, Korban SS (2011). Integration of physical and genetic maps in apple confirms whole-genome and segmental duplications in the apple genome. J Exp Bot 62: 5117-5130.

Hemmat M, Weeden NF, Brown SK (2003). Mapping and evaluation of *Malus* × *domestica* microsatellites in apple and pear. J Am Soc Hortic Sci 128: 515-520.

Hokanson SC, Szewc-McFadden AK, Lamboy WF, McFerson JK (1998). Microsatellite (SSR) markers reveal genetic identities, genetic diversity and relationships in a *Malus* × *domestica* Borkh. core subset collection. Theor Appl Genet 97: 671-683.

Hormaza JI (2002). Molecular characterization and similarity relationships among apricot (*Prunus armeniaca* L.) genotypes using simple sequence repeats. Theor Appl Genet 104: 321-328.

Inoue E, Matsuki Y, Anzai H, Evans K (2007). Isolation and characterization of microsatellite markers in Japanese pear (*Pyrus pyrifolia* Nakai). Mol Ecol Notes 7: 445-447.

Kacem NS, Muhovski Y, Djekoun A, Watillon B (2017). Molecular characterization of genetic variation in somaclones of durum wheat (*Triticum durum* Desf) using SSR markers. Eur Sci J 13: 426-437.

Kafkas S, Ozkan H, Ak BE, Acar I, Atli HS, Koyuncu S (2006). Detecting DNA polymorphism and genetic diversity in a wide pistachio germplasm: comparison of AFLP, ISSR and RAPD markers. J Am Soc Hortic Sci 131: 522-529.

Keles H, Akca Y, Ercisli S (2014). Selection of promising walnut genotypes (*Juglans regia* L.) from inner Anatolia. Acta Sci Pol Hortorum Cultus 13: 167-173.

Khan MA, Durel CE, Duffy B, Drouet D, Kellerhals M, Gessler C, Patocchi A (2007). Development of molecular markers linked to the 'Fiesta' linkage group 7 major QTL for fire blight resistance and their application for marker assisted selection. Genome 50: 568-577.

Lacis G, Rashal I, Ruisa S, Trajkovski V, Iezzoni AF (2009). Assessment of genetic diversity of Latvian and Swedish sweet cherry (*Prunus avium* L.) genetic resources collections by using SSR (microsatellite) markers. Sci Hortic 121: 451-457.

Liebhard R, Gianfranceschi L, Koller B, Ryder CD, Tarchini R, Van de Weg E, Gessler C (2002). Development and characterisation of 140 new microsatellites in apple (*Malus* × *domestica* Borkh.). Mol Breed 10: 217-241.

Liu K, Muse SV (2005). PowerMarker: integrated analysis environment for genetic marker data. Bioinformatics 21: 2128-2129.

Moriya S, Iwanami H, Kotoda N, Haji T, Okada K, Terakami S, Mimida N, Yamamoto T, Abe K (2012). Aligned genetic linkage maps of apple rootstock cultivar 'JM7' and *Malus sieboldii* 'Sanashi 63' constructed with novel EST-SSRs. Tree Genet Genomes 8: 709-723.

Oddou-Muratorio S, Aligon, C, Decroocq S, Plomion C, Lamant T, Mush-Demesure B (2001). Microsatellite primers for *Sorbus torminalis* and related species. Mol Ecol Notes 1: 297-299.

Özbek S (1978). Özel Meyvecilik. Çukurova Üniversitesi, Ziraat Fakültesi Yayın No: 128, 485 s.

Peakall R, Smouse PE (2012). GenAlEx 6.5: genetic analysis in Excel. Population genetic software for teaching and research - an update. Bioinformatics 28: 2537-2539.

Pınar H, Kaymak S, Özogun S, Uzun A, Unlu M, Bircan M, Ercişli S, Orhan E (2016). Morphological and molecular characterization of major quince cultivars from Turkey. Not Bot Horti Agrobo 44: 72-76.

Pritchard JK, Stephens M, Donnelly P (2000). Inference of population structure using multilocus genotype data. Genetics 155: 945-59.

Rakonjac V, Akšić MF, Nikolić D, Milatović D, Čolić S (2010). Morphological characterization of 'Oblačinska'sour cherry by multivariate analysis. Sci Hortic 125: 679-684.

Rohlf FJ (2009). NTSYSpc: Numerical Taxonomy System, ver.2.21c. Setauket, NY, USA: Exeter Publishing.

Sanchez EE, Menendez RA, Daley LS, Boone RB, Jahn OL, Lombard PB (1988). Characterization of quince (*Cydonia*) cultivars using polyacrylamide gel electrophoresis. J Environ Hortic 6: 53-59.

Scheulke M (2000). An economic method for the fluorescent labeling of PCR fragments. Nat Biotechnol 18: 233-234.

Schlotterer C, Tautz D (1992). Slippage synthesis of simple sequence DNA. Nucleic Acids Res 20: 211-215.

Silfverberg-Dilworth E, Matasci CL, Van de Weg WE, Van Kaauwen MPW, Walser M, Kodde LP, Soglio V, Gianfranceschi L, Durel CE, Costa F et al. (2006). Microsatellite markers spanning the apple (*Malus x domestica* Borkh.) genome. Tree Genet Genomes 2: 202-224.

Silva BM, Andrade PB, Ferreres F, Domingues AL, Seabra RM (2002). Phenolic profile of quince fruit (*Cydonia oblonga* Mill.) pulp and peel. J Agric Food Chem 50: 4615-4618.

Silva BM, Andrade PB, Martins RC, Valentao P, Ferreres F, Seabra RM, Ferreira MA (2005). Quince (*Cydonia oblonga* Mill.) fruit characterization using principal component analysis. J Agric Food Chem 53: 111-122.

Topçu H, Kafkas S, Doğan A, Akcay ME, Ercişli S (2015). Turkey genetic relatedness among quince (*Cydonia oblonga* Miller) accessions from Turkey using amplified fragment length polymorphisms. J Appl Bot Food Qual 88: 197-201.

Yamamoto T, Kimura T, Sawamura Y, Manabe T, Kotobuki K, Hayashi T, Ban Y, Matsuta N (2002a). Simple sequence repeats for genetic analysis in pear. Euphytica 124: 129-137.

Yamamoto T, Kimura T, Shoda M, Ban Y, Hayashi T, Matsuta N (2002b). Development of microsatellite markers in the Japanese pear (*Pyrus pyrifolia* Nakai). Mol Ecol Notes 2: 14-16.

Yamamoto T, Kimura TJ, Soejima T, Sanada T, Ban Y, Hayashi T (2004). Identification of quince varieties using SSR marker developed from pear and apple. Breed Sci 54: 239-244.

Yüksel C, Mutaf F, Demirtaş I, Öztürk G, Ergül A (2013). Characterization of Anatolian traditional quince cultivars, based on microsatellite markers. Genet Mol Res 12: 5880-5888.

Zaloğlu S, Kafkas S, Doğan Y, Güney M (2015). Development and characterization of SSR markers from pistachio (*Pistacia vera* L.) and their transferability to eight *Pistacia* species. Sci Hortic 189: 94-103.