



Genetic diversity analysis of apricot cultivars grown in China based on SSR markers

Ming Li¹, Pinguang Zheng², Biyong Ni², Xia Hu¹, Xingjun Miao³ and Zhong Zhao³

¹ Forestry College, Fujian Agriculture and Forestry University, Fuzhou, China

² Fuzhou Botanical Garden, Fuzhou, China

³ Key Laboratory of Environment and Ecology in Western China of Ministry of Education, College of Forestry, Northwest A&F University, Yangling, China

Summary

The simple sequence repeat (SSR) markers were used to investigate the genetic diversity and relationships of 76 accessions of the main cultivated apricot cultivars in China. The 10 SSR markers revealed that the observed number of alleles (n_a) per locus was 4.6, the number of effective alleles (n_e) was 3.46, and a higher value of genetic diversity parameters ($H_e = 0.65$, $I = 1.26$) was maintained at the species level. The genetic diversity in the Chinese group and Central Asian group was high, whereas the genetic diversity in the European group was possible low. UPGMA dendrogram and genetic structure analysis determined seven major clusters and grouped the cultivars in agreement with their geographic and species origins. The European group showed a very close genetic relationship with the North American subgroup and a distant genetic relationship with the Chinese and Central groups. The Irano-Caucasian group had a close genetic relationship with the apricot germplasms originated in Xinjiang, China. Additionally, the Chinese group and Central Asian group shared a few of similar genetic structure. The kernel-using apricot had a close genetic relationship with *Prunus sibirica* and was most likely an interspecific hybrid of *P. sibirica* and *Prunus armeniaca*.

Keywords

apricot, genetic diversity, genetic relationship, genetic structure, polymorphic, *Prunus armeniaca*

Introduction

Genetic diversity is the core of biological diversity. The presence of plant genetic diversity is essential to ensure its long-term adaptation to the changing ecological environment (Sreekanth et al., 2012). Abundant plant genetic diversity can provide a wide range of genetic backgrounds for crop genetics and breeding research (Li et al., 2014). To date, artificial selection and genetic drift have caused the loss of a large amount of genetic diversity in crops, thereby reducing the germplasm potential of the crops in the modern agriculture system. This problem has received increasing attention (Laidò et al., 2013). The assessment of the genetic diversity of a crop germplasm is very important for the protection of endangered resources and the efficient use of developable resources (Ouborg et al., 2006).

The apricot is a plant of *Rosaceae* family, genus *Prunus* L.,

Significance of this study

What is already known on this subject?

- The existing gene pool of the cultivated apricot in China is rich, but some local cultivars are facing serious genetic erosion, which means that accurate description and identification of apricot cultivars is necessary for the protection and utilization.

What are the new findings?

- The 10 SSR markers revealed that apricot cultivars grown in China maintained a relatively high level of genetic diversity ($H_e = 0.65$, $I = 1.26$). The genetic diversity in the Chinese group and the Central Asian group was high, whereas the genetic diversity in the European group was possible low. UPGMA dendrogram and genetic structure analysis determined seven major clusters and grouped the cultivars in agreement with their geographic and species origins.

What is the expected impact on horticulture?

- We expected to provide a reference for the major traits, genetic diversity, and genetic relationships of cultivated apricot varieties in China, and enhance the protection and utilization of resources and breeding.

subgenus *Prunophora* Focke, and section *Armeniaca*. It is an economically important stone fruit that is widely distributed from the temperate to subtropical regions of all continents (Decroocq et al., 2003). Section *Armeniaca* includes 10 species, 4 of which are commonly recognized [*Prunus armeniaca* L., *Prunus mandshurica* (Maxim.) Koehne, *Prunus sibirica* L. and *Prunus mume* (Sieb.) Sieb. & Zucc.] (Zhebentyayeva et al., 2003). Mainly cultivated apricots worldwide belong to only one species (*Prunus armeniaca* L.) (Hormaza et al., 2007). Kostina classified apricot germplasm into four main eco-geographical groups according to their morphological and physiological characteristics in different geographical regions: 1) the Central Asian group; 2) the Irano-Caucasian group; 3) the European group; and 4) the Dzhungar-Zailij group, which was closely linked to the wild Tien-Shan apricot (Kostina, 1964). The classification of Kostina was improved by Kryukova with the addition of the Chinese group and the inclusion of the Dzhungar-Zailij group in the Central Asian group (Kryukova, 1989).

The history of apricot cultivation in China stretches over 2,000 years, and the main grown cultivars are from the Chinese and Central Asian groups (Wang et al., 2011). Accord-

ing to the morphology, cultivation and molecular marker data, China and Central Asia (from the Tianshan Mountains to Kashmir) are considered the two primary centers of the origin of cultivated apricots worldwide; Near Eastern region is regarded as the secondary gene center of origin (Hormaza et al., 2007). The Chinese group is the oldest and most diverse group in the world, and more than 2,000 out of the over 3,000 apricot cultivars worldwide are distributed in China (Zhebentyayeva et al., 2012). Based on molecular data, Zhebentyayeva et al. (2003, 2008) indicated that the apricot varieties cultivated in China belonged to the interspecific hybrid of the northeast apricot, Siberia apricot or plum with the common apricot. The Central Asian group is one of the oldest groups and shows a high level of diversity because of the seed propagation and abundant wildlife resources (He et al., 2007). Recently, some hybrids of the cultivated apricot varieties derived from European and North America with the plumcot were also introduced to China for cultivation (Zhang et al., 2014). The existing gene pool of the cultivated apricot in China is rich, but some local cultivars are being replaced with varieties with higher profits and better market demand, whereas some rare apricot varieties with local cultivation are facing serious genetic erosion (Zhang et al., 2008). The accurate description and identification of apricot cultivars is necessary for the breeding and commercialization of valuable cultivated apricot varieties. Due to the limitations of the environmental conditions for growth, the existing gene pool of some cultivated apricot varieties also limits their introduction into new areas (Krichen et al., 2010). Generally, these problems have led to an inevitable need for genetic solutions and the utilization of genetic diversity and genetic relationship analyses for the protection and utilization of apricot cultivars (Yilmaz et al., 2012).

The combination of necessary and less laborious morphological evaluations with the analysis of molecular markers leads to more reliable conclusions for the assessment of genetic diversity (Fu, 2015). To this end, several molecular techniques that are not affected by environmental changes, including inter-simple sequence repeats (ISSRs), sequence-related amplified polymorphisms (SRAPs), amplified fragment length polymorphisms (AFLPs) and simple sequence repeats (SSRs), have been used to describe the diversity and genetic characterization of apricot germplasms worldwide (Bourguiba et al., 2012; Hagen et al., 2002; Li et al., 2014; Romero et al., 2003; Yilmaz et al., 2012). Due to the advantages of high reliability and polymorphisms as well

as co-dominance, SSR markers have been widely applied to study the genetic diversity and inter-species genetic relationships of apricots in China, Turkey, Morocco and other different eco-geography groups (Bourguiba et al., 2012; Wang et al., 2011; Yilmaz et al., 2012; Decroocq et al., 2016). In the present study, we used SSR markers to investigate the genetic diversity and genetic relationships of 76 accessions of the main cultivated apricot varieties in China. The objectives of this study are as follows: 1) to assess the genetic diversity of the main cultivated apricot varieties in China; 2) to analyze the genetic relationships among the main cultivated apricot varieties in China; and 3) to estimate the extent of variation in apricot germplasms between and within ecogeographical groups and subgroups.

Materials and methods

Plant materials

The experimental materials were 76 accessions of the main cultivated apricot varieties in China, including 32 varieties in the Chinese group, 20 varieties in the Central Asian group represented by the Sinkiang apricot, 9 cultivars in the European group (including 6 North American subgroup cultivars), 2 cultivars in the Irano-Caucasian group, 5 kernel-using apricot varieties and 8 plumcot hybrids. The names, origins and main characteristics of the varieties are shown in Table 1. The leaves were collected from 76 apricot accessions cultivated from the apricot genebank of the Weihe River Experimental Station at Northwest A&F University, China, to extract genomic DNA using the UNI-Q-10 Plant Genomic DNA Prep Kit (Sangon, Shanghai, China). The DNA quality and concentration were determined using the Epoch™ microplate spectrophotometer (BioTek, Winooski, VT, USA) and diluted to a working concentration of 30 ng μL^{-1} .

DNA amplification

The extracted apricot genomic DNA was amplified by PCR using 10 SSR markers; the primer sequences and sources are listed in Table 2. The ABI-Veriti 96-well gradient PCR machine (Applied Biosystems, Foster City, CA, USA) was used for the DNA amplification reaction. The PCR amplification was performed in a 15 μL reaction system containing 30 ng of genomic DNA, 0.25 mM primers, 3 mM MgCl_2 , 0.4 mM dNTPs and 1 U of Taq DNA polymerase (Takara). The PCR amplification program was as follows: initial denaturation at 95°C for 3 min; 35 cycles of denaturation at 94°C for 30 s, an-

TABLE 2. List of SSR markers and genetic diversity parameters for the accessions studied.

| Primer | Reference | SSR motive | Annealing temp. (°C) | n_a | n_e | Ho | He | <i>I</i> |
|-------------|--------------------------|---------------------------------------|----------------------|-------|-------|------|------|----------|
| AMPA101 | Hagen et al., 2004 | (TC) ₁₁ (AC) ₁₂ | 56 | 5 | 4.63 | 0.79 | 0.79 | 1.56 |
| AMPA119 | Hagen et al., 2004 | (TA) ₉ | 57 | 7 | 5.45 | 0.66 | 0.82 | 1.77 |
| BPPCT039 | Dirlewanger et al., 2002 | (GA) ₂₀ | 55 | 5 | 3.95 | 0.86 | 0.75 | 1.46 |
| pchgms3 | Sosinski et al., 2000 | (CT) ₁₉ | 57 | 5 | 4.51 | 0.78 | 0.78 | 1.56 |
| pchgms5 | Sosinski et al., 2000 | (CA) ₉ (TA) ₈ | 51 | 2 | 1.33 | 0.26 | 0.25 | 0.41 |
| ssrPaCITA23 | Lopes et al., 2002 | (AC) ₂ (AG) ₁₈ | 51 | 7 | 3.53 | 0.79 | 0.72 | 1.52 |
| UDAp-414 | Messina et al., 2004 | (AG) ₂₁ | 56 | 4 | 3.65 | 0.82 | 0.73 | 1.33 |
| UDAp-415 | Messina et al., 2004 | (GA) ₂₁ | 56 | 4 | 3.56 | 0.54 | 0.72 | 1.33 |
| UDAp-420 | Messina et al., 2004 | (CT) ₂₀ | 56 | 5 | 4.15 | 0.86 | 0.76 | 1.51 |
| UDP96-001 | Cipriani et al., 1999 | (CA) ₁₇ | 57 | 2 | 1.31 | 0.20 | 0.24 | 0.40 |

n_a is the observed number of alleles, n_e is the effective number of alleles, Ho is the observed heterozygosity, He is the expected heterozygosity, *I* is the Shannon's information index.

TABLE 1. Apricot cultivars evaluated in this study with their origins and key botanical traits.

| Group | Cultivar | Origin | Major traits |
|--------------------|------------------------|---------------------|--|
| Chinese group | Yuxing | Anhui, China | High fruit quality (attractiveness, size), flower and fruit drop easily |
| | Luotuo Huang | Beijing, China | Combining very early ripening with superior fruit quality (size, firmness, sugar) |
| | Shuangrenxing | Gansu, China | Very large fruit size, good postharvest characteristics |
| | Tangwangchuan dajixing | Gansu, China | Large fruit size, high fruit quality (attractiveness, succulent, fragrant, firmness) |
| | Chuanzhongxing | Hebei, China | High productivity, flesh is firm, high cold and salt resistance |
| | Gongfoxing | Hebei, China | Large fruit size, well-adapted, high fruit quality (size, sugar, succulent) |
| | Jiguangxing | Hebei, China | Early ripening, flesh is firm, suitable for flesh processing |
| | Miantaoxing | Hebei, China | Small fruit size, less juice content, suitable for dried apricot processing |
| | Qihong | Hebei, China | Late fruit maturity |
| | Zihexing | Hebei, China | Very early ripening, high resistance for bleeding disease and bacterial shot-hole |
| | Shanxing | Hebei, China | <i>P. sibirica</i> , rootstock cultivar, high cold and drought resistance, barren soil tolerance |
| | Jidanxing | Henan, China | Strong tree vigor, high productivity |
| | Bayuehong | Liaoning, China | Very late ripening, high fruit quality (attractiveness, size, sugar) |
| | Meinong | Liaoning, China | Purple spots on flesh, sweet-sour juice |
| | Caopeixing | Shaanxi, China | Cold resistance, medium ripening, relatively high productivity |
| | Damingxing | Shaanxi, China | Large fruit size, medium ripening, sweet-sour flesh |
| | Jingyangdayinxing | Shaanxi, China | Strong tree vigor, high fruit quality (size, sugar, succulent, fragrant) |
| | Yinxiangbai | Shaanxi, China | Strong tree vigor, high productivity, high fruit quality, white-green flesh |
| | Daguoxing | Shandong, China | High productivity, drought resistance, barren soil tolerance, fruit crack easily |
| | Laixijinxing | Shandong, China | High productivity, early ripening, cold resistance, high quality (size, sugar, fragrant) |
| | Shiguan 1 | Shandong, China | Early ripening, fruit crack and cold resistant, high quality (attractiveness, fragrant) |
| | Xinshiji | Shandong, China | Early ripening, high fruit quality (size, attractiveness, fragrant, firmness) |
| | Shajinhong | Shanxi, China | Late fruit maturity, well-adapted, high fruit quality (attractiveness, firmness) |
| | Duanwuxing | Shanxi, China | Early ripening, flesh fragrant, high fruit quality (sugar, succulent) |
| | RV | North China | High productivity, cold and drought resistance |
| | Tw | North China | High productivity, large size |
| | Zhongqiumei | North China | Late ripening, high fruit quality (attractiveness, sugar, fragrant) |
| | Huaguanxing | North China | High cold & drought resistance, well-adapted, large petals |
| | Xiaoyuehong | North China | High productivity, high fruit quality (sugar, succulent), purple spots on flesh |
| | Shijilong | North China | Large fruit size, sweet-sour flesh |
| | Ribendajixing | Japan | Late ripening, large fruit size, drought resistance, low productivity |
| | Ribenhongxing | Japan | Large fruit size, low productivity, medium ripening, |
| Central Asia group | Dongxing | Aksu, Sinkiang | Very late ripening, high productivity, cold and drought resistance |
| | Cuheyixing | Karghalik, Sinkiang | High fruit quality (attractiveness, firmness, fragrant), flesh for dried apricot |
| | Xiheyixing | Karghalik, Sinkiang | High fruit quality (attractiveness, firmness, fragrant), sweet-sour flesh, flesh for dried apricot |
| | Zaoshuheyixing | Karghalik, Sinkiang | Early ripening, flesh for dried apricot |
| | Akeyageleke | Khotan, Sinkiang | High productivity, late ripening, high fruit quality (sugar, succulent, fragrant) |
| | Anjianghuanna | Khotan, Sinkiang | High fruit quality (sugar, firmness, fragrant) |
| | Huanna | Khotan, Sinkiang | Strong tree vigor, high fruit quality (sugar, succulent) |
| | Huanghongyike | Khotan, Sinkiang | High fruit quality (attractiveness, firmness, sugar, fragrant) |
| | Kaerhuanna | Khotan, Sinkiang | High productivity, large fruit size, high firmness, good processing characteristics |
| | Katakehuanna | Khotan, Sinkiang | High fruit quality (attractiveness, sugar, firmness, fragrant) |
| | Kuikipiman | Khotan, Sinkiang | High productivity, well-adapted, high fruit quality (attractiveness, firmness, fragrant) |

TABLE 1. Continued.

| Group | Cultivar | Origin | Major traits |
|----------------------------|--------------------|---|--|
| Central Asia group | Pinaizi | Khotan, Sinkiang | High productivity, cold resistance, high fruit quality (size, succulent, fragrant) |
| | Kuerletuoyong | Korla, Sinkiang | High productivity, high fruit quality (attractiveness, firmness, sugar, fragrant) |
| | Aketuoyong | Kuqa, Sinkiang | High productivity, flesh is firm |
| | Kuchetuoyong | Kuqa, Sinkiang | Small fruit size, high fruit quality (attractiveness, fragrant) |
| | Baxing | Southern Sinkiang | Flesh for dried apricot |
| | Kezimayisang | Southern Sinkiang | Early ripening, high productivity, high sugar |
| | Tedazaoshuxing | Southern Sinkiang | Early ripening, large fruit size |
| | Saimaiti Zixing | Yingjisha, Sinkiang Tokkuztara, Sinkiang | High productivity, well-adapted, flesh for dried apricot <i>P. dasycarpa</i> , dark purple fruit |
| Europe group | Jintaiyang | Unknown, Europe | Early ripening, large fruit size, high fruit quality (attractiveness, sugar, succulent, fragrant) |
| | Tyrinthos | Italy | High and stable productivity, cold resistance |
| | Tianren | Unknown, Europe | High fruit quality (attractiveness, sugar, fragrant), sweet kernel |
| | Aozhoutianxing | Australia | High productivity, sweet-sour flesh, well-adapted |
| North America sub-group | Dapengwang | America | Very large fruit size, high productivity, storage stability |
| | Manaoxing | America | High and stable productivity, early ripening, good postharvest characters |
| | Katy | America | Large fruit size, high productivity, sweet-sour flesh, beautiful appearance |
| | Gold rich | America | High productivity, sour flesh, storage stability, suitable for processing |
| | Zhengkui Jinya | America Armenia | High productivity, cold resistance, strong tree vigor High productivity, large fruit size, cold resistance, high fruit quality (sugar, succulent, fragrant) |
| Iran-Caucasian group | Sulian No.4 | Armenia | Large fruit size, cold resistance, sweet and juicy flesh |
| | Baiyubian | Beijing | <i>P. armeniaca</i> × <i>P. sibirica</i> , sweet kernel apricot, well-adapted, high cold and drought resistance |
| Kernel-using apricot group | Chaoren | Hebei | <i>P. armeniaca</i> × <i>P. sibirica</i> , sweet kernel apricot, very large kernel, high cold and drought resistance |
| | Guoren | Hebei | <i>P. armeniaca</i> × <i>P. sibirica</i> , sweet kernel apricot, high productivity, high pest and disease resistance |
| | Youren | Hebei | <i>P. armeniaca</i> × <i>P. sibirica</i> , sweet kernel apricot, high oil content in kernel |
| | You1 | Hebei | <i>P. armeniaca</i> × <i>P. sibirica</i> , sweet kernel apricot, very large kernel, barren soil tolerance |
| | Xingmei | Shanxi | <i>P. simonii</i> , very large fruit size, high fruit quality (attractiveness, firmness, fragrant) |
| Plumcot group | Weidi | America | <i>P. simonii</i> , early ripening, lilac peel with red spots, high fruit quality (sugar, succulent, fragrant) |
| | Weihou | America | <i>P. simonii</i> , high productivity, atropurpureus peel, high fruit quality (sugar, succulent, fragrant) |
| | Weiwang | America | <i>P. simonii</i> , fuchsia peel, high fruit quality (size, attractiveness, sugar, succulent, fragrant) |
| | Weixing | America | <i>P. simonii</i> , early ripening, yellowish red peel |
| | Konglongdan | America | <i>P. simonii</i> , very large fruit size |
| | Fengweihuanghou | America | <i>P. simonii</i> , yellowish peel and flesh, high fruit quality (sugar, succulent, fragrant) |
| | Fengweimeigui | America | <i>P. simonii</i> , early ripening, violet black peel, high fruit quality (firmness, sugar, succulent, fragrant) |

nealing at 51–57°C (depending on the primers) for 30 s, and extension at 72°C for 60 s; and a final extension at 72°C for 5 min. The PCR products were separated by electrophoresis on a 6% denaturing polyacrylamide gel, and the fragments were visualized by silver staining. The genotypes were visually assessed on the fluorescent plate.

Data analysis

For each SSR locus, the allelic composition and the number of total alleles were determined for each accession. The genetic diversity parameters including the observed number of alleles (n_a), the effective number of alleles (n_e), the observed heterozygosity (H_o), the expected heterozygosity (H_e), the Shannon's information index (I), the coefficient of gene differentiation (F_{st}) and the gene flow (N_m), were statistically analyzed using the POPGENE 1.32 (Nei, 1973; Shannon, 2001; Yeh et al., 1997). Additionally, the unweighted pair-group method with arithmetic means (UPGMA) hierarchical clustering tree was constructed based on Nei's unbiased genetic distances of different apricot cultivar groups (Nei, 1973; Yeh et al., 1997). The evolutionary tree among the germplasms of the 76 cultivated apricot accessions was constructed using the GenAlEx 6.5 software based on the Jaccard genetic similarity coefficient (Peakall and Smouse, 2012).

The genetic structure was analyzed using the STRUCTURE 2.3.4 software based on Bayesian clustering (Pritchard et al., 2000). To identify the true number of populations (K) covering the majority of the structures in the data, we used a burn-in period of 50,000 Markov Chain Monte Carlo iterations and 100,000 runs, with an admixture model following the Hardy-Weinberg equilibrium; we correlated the allele frequencies and independent loci for each run. The true number of populations (K value) in the classification was set as 1–20, and each K value was independently run 7 times. Subsequently, a $Pr(X|K)$ index with respect to each K value was used to calculate ΔK using the formula described by Evanno (Evanno et al., 2005). The optimal K value depends on the first peak of $\Delta K = |L''(K)|/s[Pr(x|k)]$, where $|L''(K)|$ denotes the absolute value of the second order rate of change of $Pr(X|K)$ and $s[Pr(x|k)]$ denotes the standard deviation of $Pr(X|K)$ (Laidò et al. 2013).

Results

Genetic diversity analysis

A total of 46 alleles were amplified from the genomic DNA of 76 apricot germplasm accessions, with 3.6068 effective alleles obtained per locus (Table 2). The highest number of alleles and number of effective alleles were observed with marker AMPA119 (7 and 5.45, respectively),

which indicated that the genetic diversity parameters were the highest ($H_e = 0.82$, $I = 1.77$), followed by marker AMPA101. The lowest number of alleles was observed with the primers pchgms5 and UDP96-001, and the number of effective alleles was different – 1.33 and 1.31, respectively. Similarly, the expected heterozygosity (H_e) and the Shannon information index (I) obtained with the pchgms5 and UDP96-001 markers were low, and all other markers showed a higher genetic diversity index. Table 3 shows the genetic diversity of the apricot populations cultivated in different geographical ecological groups. The genetic diversity of the Chinese group was the highest ($H_e = 0.67$, $I = 1.27$), followed by the Central Asian group and the plumcot group, whereas the genetic diversity of the Iran-Caucasian group was the lowest ($H_e = 0.62$, $I = 0.73$). At the species level, the genetic diversity of the population parameters of apricot cultivation had greatly increased, with an average observed heterozygosity (H_o) of 0.64, expected heterozygosity (H_e) of 0.65, and Shannon's information index (I) of 1.26. So, the high genetic diversity was detected in the apricot population currently cultivated in China.

Genetic relationships among cultivars

The ΔK value corresponding to each K value was calculated, and the scatter plot was prepared (Figure S1). The results showed that the maximum ΔK value appeared at $K = 5$, indicating that these 76 apricot germplasm accessions could be divided into five subpopulations based on the genetic population structure (Figure 1). These five major subpopulations consisted of the Chinese group, the Central Asian group, the European group, the kernel-using apricot group and the plumcot group. The UPGMA cluster analysis showed that the 76 apricot germplasm accessions could be divided into seven clusters based on the Jaccard genetic similarity coefficient. Among them, the 31 common apricot varieties in the Chinese group and one variety (Xiheyixing) in the Central Asian group formed cluster I. Cluster II consisted of 18 apricot varieties in the Central Asian group and 2 apricot varieties in the Irano-Caucasian group. Cluster III consisted of 9 apricot varieties in the European group, including the North American subgroup. Cluster IV consisted of 5 varieties in the kernel-using apricot group and one apricot germplasm (Shanxing) in the Chinese group. Cluster V consisted of 7 species in the plumcot group. 'Xingmei' alone formed cluster VI, and 'Zixing' alone was in cluster VII.

Genetic relationships among cultivar groups

For the analysis of the genetic relationships among the 7 apricot clusters, the UPGMA phylogenetic tree was constructed based on Nei's unbiased genetic distance using

TABLE 3. Genetic diversity parameters revealed by SSR markers for the apricot accessions.

| Population | Sample size | n_a | n_e | H_o | H_e | I |
|----------------------------|-------------|-------|-------|-------|-------|------|
| Chinese group | 32 | 4.5 | 3.53 | 0.64 | 0.67 | 1.27 |
| Central Asian group | 20 | 4.4 | 3.05 | 0.61 | 0.62 | 1.16 |
| European group | 3 | 2.9 | 2.43 | 0.77 | 0.65 | 0.92 |
| North American subgroup | 6 | 3.5 | 2.73 | 0.68 | 0.64 | 1.05 |
| Iran-Caucasian group | 2 | 2.3 | 2.13 | 0.60 | 0.62 | 0.73 |
| Kernel-using apricot group | 5 | 3.3 | 2.60 | 0.68 | 0.61 | 0.97 |
| Plumcot group | 8 | 4.0 | 3.21 | 0.76 | 0.64 | 1.15 |
| At species level | 63 | 4.6 | 3.46 | 0.64 | 0.65 | 1.26 |

n_a is the observed number of alleles, n_e is the effective number of alleles, H_o is the observed heterozygosity, H_e is the expected heterozygosity, I is the Shannon's information index.

TABLE S1. Summary of Chi-square tests for Hardy-Weinberg equilibrium (HWE).

| | Chinese group | | Central Asian group | | European group | | North American subgroup | | Iran-Caucasian group | | Kernel-using apricot group | | Plumcot group | |
|-----------|---------------|---------|---------------------|--------|----------------|-------|-------------------------|--------|----------------------|--------|----------------------------|-------|---------------|-------|
| | Chi-square | P | Chi-square | P | Chi-square | P | Chi-square | P | Chi-square | P | Chi-square | P | Chi-square | P |
| AMPA101 | 10.001 | 0.440 | 6.779 | 0.746 | 1.339 | 0.720 | 3.289 | 0.772 | 1.000 | 0.801 | 1.600 | 0.659 | 7.833 | 0.645 |
| AMPA119 | 45.867 | 0.162 | 17.432 | 0.065 | 0.750 | 0.861 | 20.889 | 0.022* | 4.000 | 0.046* | 10.300 | 0.113 | 14.500 | 0.151 |
| BPPCT039 | 14.310 | 0.159 | 3.072 | 0.980 | 3.250 | 0.777 | 1.150 | 0.765 | 1.622 | 0.654 | 4.000 | 0.261 | 2.438 | 0.875 |
| Pchgms5 | 0.752 | 0.386 | 1.301 | 0.254 | 0.333 | 0.564 | 0.417 | 0.519 | - | - | - | - | 0.077 | 0.782 |
| UDAP414 | 5.439 | 0.489 | 4.321 | 0.633 | 2.000 | 0.572 | 3.961 | 0.682 | 1.000 | 0.801 | 5.200 | 0.518 | 3.400 | 0.757 |
| UDAP415 | 19.890 | 0.003** | 8.876 | 0.181 | 0.000 | 1.000 | 3.808 | 0.703 | 1.622 | 0.203 | 3.250 | 0.777 | 3.042 | 0.804 |
| UDAP420 | 36.999 | 0.000** | 21.224 | 0.020* | 2.000 | 0.572 | 5.000 | 0.544 | 2.222 | 0.528 | 2.400 | 0.494 | 3.400 | 0.334 |
| UDP96-001 | 2.671 | 0.102 | 0.027 | 0.869 | 0.000 | 1.000 | 0.111 | 0.739 | - | - | 0.143 | 0.705 | - | - |
| PACITA23 | 9.851 | 0.829 | 10.270 | 0.975 | 0.333 | 0.954 | 1.071 | 0.784 | 1.000 | 0.317 | 2.875 | 0.824 | 15.333 | 0.806 |
| Pchgms3 | 7.389 | 0.688 | 8.053 | 0.624 | 5.333 | 0.502 | 7.933 | 0.243 | 4.000 | 0.046* | 13.000 | 0.224 | 16.000 | 0.100 |

P probability; - Monomorphic locus; * P < 0.05; ** P < 0.01.

the POPGENE 1.32 software (Figure 2). The results showed that the genetic distance was closest between the North American subgroup and the European group. The genetic distance between the Central Asian group and the Irano-Caucasian group was also close, and these two groups were genetically close to the Chinese group. However, a large distance existed between the kernel-using apricot group and the plumcot group.

Discussion

Genetic diversity analysis

The results obtained with the SSR markers showed that the main apricot germplasms cultivated in China had high genetic diversity indices ($H_e = 0.66$, $I = 1.29$). The study of Zhebentyayeva et al. (2003) on the genetic diversity of the main cultivated apricots in the European group from different geographic sources showed that the expected heterozygosity was 0.645, which was similar with the apricots cultivated in China in the present study. Bourguiba et al. (2012) investigated the genetic diversity of the apricots cultivated in Algeria, Morocco and Tunisia and showed an expected heterozygosity of 0.593, which was also lower than that of the apricots cultivated in China. Zhang et al. (2014), using SSR markers, analyzed 94 apricot cultivars in China, and showed a high level of genetic diversity with the average expected heterozygosity of 0.792, which was higher than the result of our study. Wang et al. (2011) analyzed the genetic diversity of 150 core samples of the Chinese apricot germplasm and showed a higher expected heterozygosity (0.731) than the result of our study, which might be related to the number of samples.

As an original center of apricot cultivation, propagation and domestication with long-term seed propagation and plenty of wildlife resources, China and Central Asia have the oldest apricot germplasm resources with the richest diversity (Zhebentyayeva et al., 2012). In this study, the apricot germplasms in the Chinese and Central Asian groups showed higher genetic diversity parameters than the other ecological groups. The European group including the North American subgroup was considered the youngest group in origin and showed the lowest genetic diversity parameters (Romero et al., 2003; Zhebentyayeva et al., 2012). The 5 kernel-using apricot varieties in this study were mostly breeding hybrids from the same or close parents, whose genetic bases were narrow, and they showed low genetic diversity (Ai et al., 2011). As a hybrid crossing different varieties, the plumcot has the genetic backgrounds of both the apricot and plum and thus demonstrates a higher genetic diversity (Liu et al., 2007). The genetic diversity of the Iran-Caucasian group was low, possibly because there were only two accessions, and the small sample size affected the genetic diversity.

Genetic relationships among cultivars

Analysis of the genetic structures and genetic relationships among varieties is necessary for seed breeding hybrids (Zhang et al., 2014). The high genetic diversity of the apricot population cultivated in China provides a rich germplasm potential to improve genetic breeding. The 76 apricot germplasm accessions were basically distinguished according to their eco-geographical origins and phylogroups. Romero et al. (2003) analyzed 40 apricot accessions using SSR markers and showed that the accessions were distinguished according to their ecological and geographical origin. The study of Zhang et al. (2014) showed that the application of SSR mark-

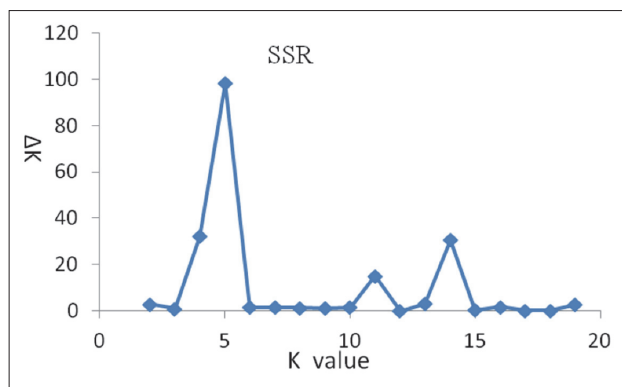


FIGURE S1. ΔK values for different numbers of populations assumed (K) in the STRUCTURE analysis.

ers could distinguish the germplasms of kernel-using apricot, Sinkiang apricot and foreign apricot from the germplasm of Chinese apricot.

In this study, with the exception of the ‘Shanxing’ belonging to *P. sibirica*, the 31 apricot cultivars in the Chinese group, including ‘Ribenxing’ and ‘Ribendajixing’ that originated in Japan, were grouped into cluster I. With the exception of ‘Zixing’ and ‘Xiheyixing’, the apricot germplasms with Central Asian and Irano-Caucasian origins were grouped into cluster II. Many studies showed that the cultivated apricot varieties in the Irano-Caucasian

group were derived from the propagation and integration of the germplasms in the Chinese group and the Central Asian group (Bourguiba et al., 2010; Yilmaz et al., 2012). These germplasms were genetically modified in the Iranian plateau, adapted to the local ecological environment, and propagated in the Irano-Caucasian area (Zhebentyayeva et al., 2012). Thus, the genetic relationship of the apricot varieties in the Irano-Caucasian and Central Asian groups was the closest, and these varieties were grouped into the same cluster (cluster II). Previous studies suggested that the apricot germplasm cultivated in North America originated in European, with a close genetic relationship between them (Hagen et al., 2002; Romero et al., 2003; Zhebentyayeva et al., 2003). In this study, the germplasms of the apricot cultivated in European and North America were grouped into cluster III, with the similar genetic background.

The kernel-using apricot is the apricot germplasm hybrid from *P. armeniaca* and *P. sibirica*, which were native to China and showed different genetic background from the apricot germplasms of the other groups (Liu et al., 2012). The kernel-using apricot has characteristics of fruit and seeds with a flat shape, less juice and less fruit, a sweet and very large kernel, high productivity, and cold and drought resistance (Ai et al., 2014). The traits of the kernel-using apricot differ greatly from those of *P. armeniaca* but are partially similar to *P. sibirica*, with unique alleles that are different from common apricot varieties in China (Liu et al., 2010). Ai et al. (2011)

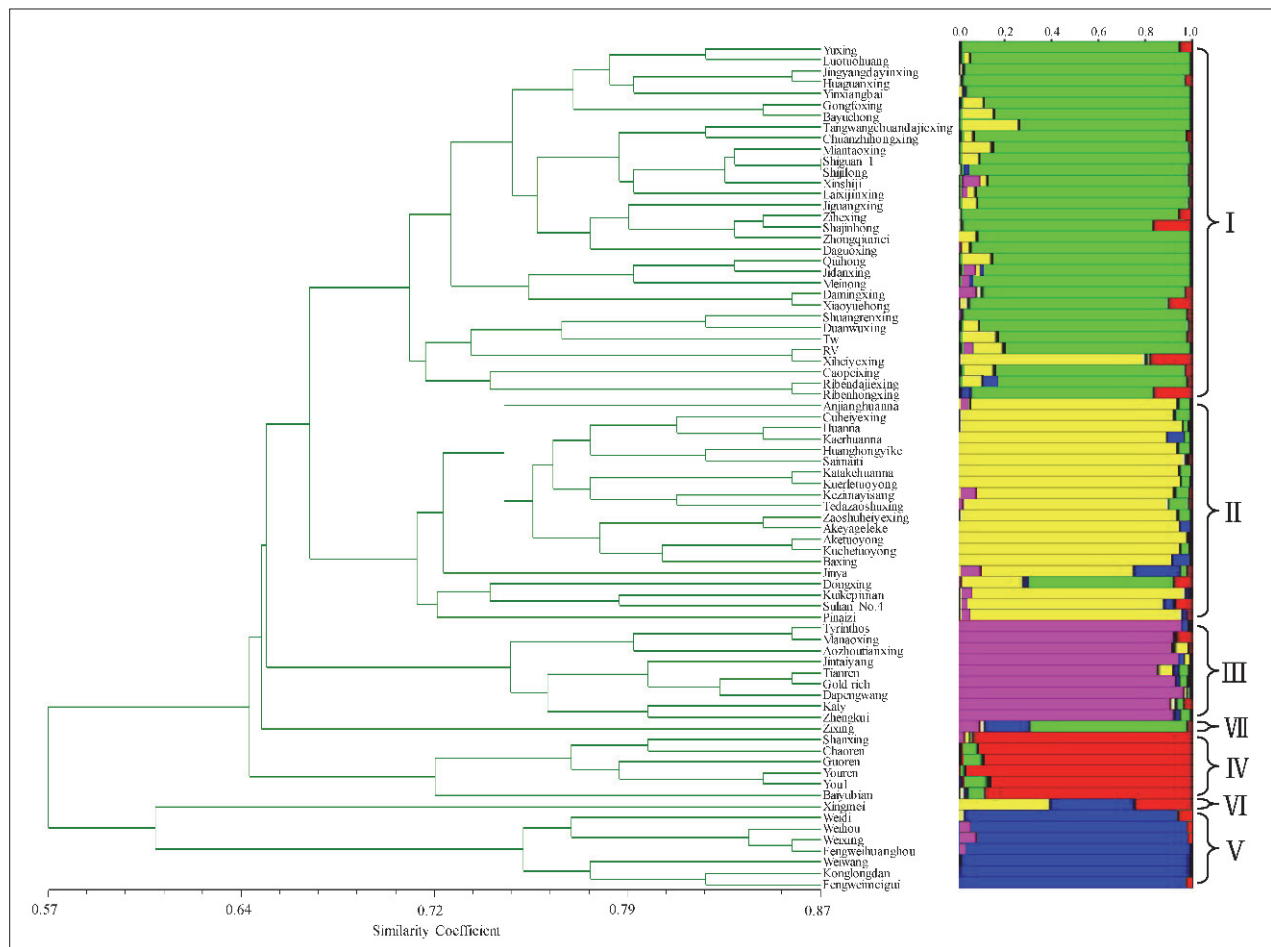


FIGURE 1. Clustering of 76 apricot cultivars in China revealed by SSR markers. Left: the UPGMA phylogenetic tree based on the Jaccard genetic similarity coefficient among the varieties obtained with SSR markers; Right: the speculated genetic structure of the clusters. Different colors represent the probability for each sample belonging to the subset when $K = 5$, and each box represents a sample.

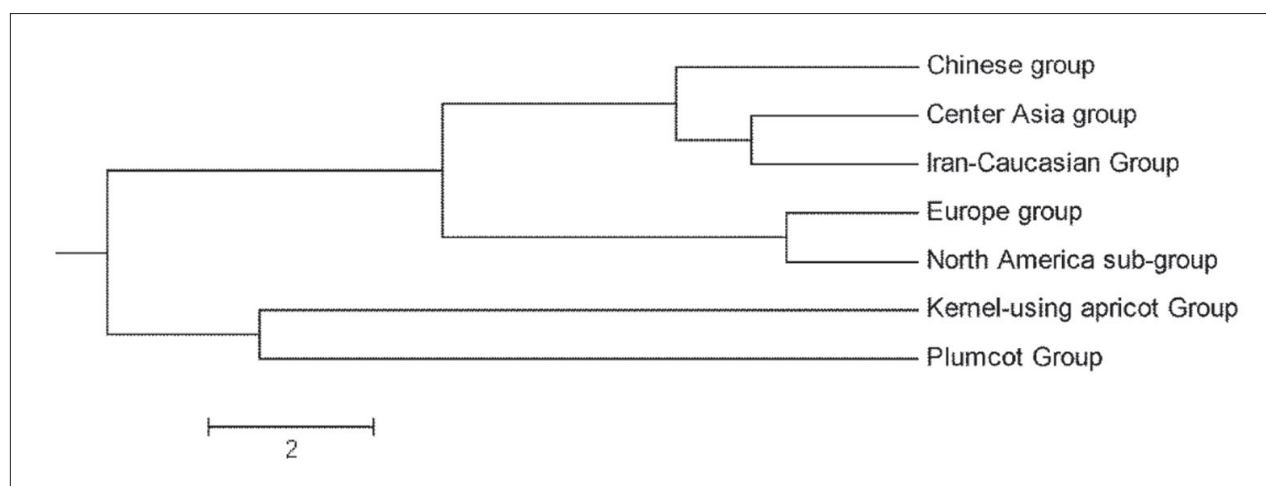


FIGURE 2. UPGMA dendrogram for the cultivar groups included in this study generated by Nei's unbiased genetic distance based on SSR markers.

suggested that the kernel-using apricot had a closer genetic relationship with *P. armeniaca* and a large genetic distance from *P. sibirica* based on SRAP markers. In this study, the kernel-using apricot had a similar genetic background to the 'Shanxing' variety belonging to *P. sibirica*. We support that the kernel-using apricot is most likely a new type of natural hybridization or introgression with the common apricot and Siberian apricot and should be classified as an independent species or subspecies (Zhang et al., 2013).

In this study, 8 plumcot varieties showed the closest genetic relationships, including 7 plumcot species from America grouped in cluster V and 'Xingmei' (a species of Chinese plumcot) that was alone in cluster VI. According to Hagen et al. (2002), 'Zixing' belongs to the plum hybrid, whereas traditional studies suggested that 'Zixing' belongs to a hybrid of *P. cerasifera* × *P. armeniaca* (Kostina, 1964; Soldatov and Petr, 2007). 'Zixing' was alone in cluster VII, while a portion of its genetic background was more similar with common apricot than plumcot; thus, we recommend 'Zixing' is most likely a special species as the natural hybrid of common apricot and plum.

Genetic relationships among cultivar groups

In this study, the genetic distances of the Central Asian group from the Irano-Caucasian group and the Chinese group were small, whereas its genetic distance from the European group was large. Zhebentyayeva et al. (2003) indicated that the genetic identity of the Chinese group with the European group was low, whereas its genetic identities with the Irano-Caucasian group and the Fergana subgroup were high; the Fergana subgroup belonged to the Central Asian group. Geographically, the expected heterozygosity of the apricot gradually decreased in the chain China – Central Asia – Middle Europe, and the genetic distance of the apricot germplasm between Middle Europe and China was large (Pedryc et al., 2009). Thus, the genetic resources derived from the Chinese and Central Asian groups may have been introduced into the Irano-Caucasian group during the long propagation process. In this study, the apricot germplasms from Europe and North America showed the closest genetic distance, supporting the hypothesis that the apricot germplasm in the North American subgroup is from the propagation of the European group (Romero et al., 2003; Zhebentyayeva et al., 2003). Decroocq et al. (2016) used SSR markers to reveal the genetic relationship among different apricot eco-geography

groups and clustered the European apricot group and Irano-Caucasian apricot group together by structure analysis. There are only two individuals of Irano-Caucasian group in this study, and they showed the closer genetic relationship with Central Asia group apricots. We believed more extensive samples and effective method could give us more clear results. Hagen investigated the genetic diversity of different types of apricot germplasms, and the constructed phylogenetic tree showed that *P. mume* and *P. dasycarpa* belonged to a unique branch, which was distinguished from the common apricot groups (Hagen et al., 2002). In this study, although the hybrid type of the plumcot group and the kernel-using apricot group were hybrids of the germplasms from the common apricot and other plums or apricots, their genetic distances from the other common apricot groups were still large.

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Addresses of authors:

Ming Li¹, Pinguang Zheng², Biyong Ni², Xia Hu^{1,*},
Xingjun Miao³ and Zhong Zhao³

¹ Forestry College, Fujian Agriculture and Forestry
University, Fuzhou, China

² Fuzhou Botanical Garden, Fuzhou, China

³ Key Laboratory of Environment and Ecology in Western
China of Ministry of Education, College of Forestry,
Northwest A&F University, Yangling, China

* Corresponding author; E-mail: lake-autumn@163.com