



Full Length Article

Identification of Quantitative Trait Loci Associated with Fruit Quality Traits in Pear

Long Wang, Su-Ke Wang, Yan-Li Su, Yun Chen, Lei Wang, Xiu-Gen Li* and Jian Yang*

Zhengzhou Fruit Research Institute, Chinese Academy of Agricultural Sciences, Zhengzhou 450009, China

*For correspondence: zgspear@163.com; yangjian@caas.cn

Received 01 August 2019; Accepted 07 October 2019; Published 16 January 2020

Abstract

Genetic map based quantitative trait loci (QTL) analysis is the main method for pear fruit traits research. To accelerate molecular breeding of fruit traits in pear, we analyzed QTL of ten pear fruit traits based on genetic map constructed in 2017. A total of 90 fruiting-age F₁ seedlings derived from *Pyrus communis* (L.) ‘Red Clapp’s Favorite’ × *Pyrus pyrifolia* Nakai ‘Mansoo’ were used for QTL analysis. Candidate genes were screened in the regions of the pear genome sequence corresponding to the QTL. The study identified 56, 421, 139, 64, 59, 37 and 66 candidate genes related to single fruit weight, skin color, fruit core size, fruit diameter, soluble solid content, flesh firmness, and fruit length, respectively, from the pear genome database. Based on the high-density genetic map, ten QTL for the ten traits were identified. The QTL were located on linkage group 11 (LG 11), LG 3, LG 5 and LG 12, respectively. Genetic map based QTL analysis is the primary method for pear fruit traits research. Candidate genes were screened, the fruit traits of F₁ population were measured and ten QTL for the 10 traits were detected. © 2020 Friends Science Publishers

Keywords: Candidate gene; Molecular marker; Phenotypic traits; QTL

Introduction

Phenotypic selection is the main method used for traditional pear breeding. It cannot be carried out until the individual seedlings have completed the long juvenile phase necessary before the plants can fruit. During this period, a large area of cultivated land will be occupied by those individual seedlings exhibiting undesirable fruit traits. In addition, quantitative fruit traits are markedly affected by environmental conditions. Such observations contribute to the problems associated with phenotypic selection. Genotypic selection could identify the seedlings with the desirable traits based on the relationship between genotype and phenotype. Genotypic selection is carried out through the use of molecular markers, linked to specific traits, and mainly involving quantitative trait locus (QTL) identification. Thus, QTL identification of fruit quality traits is an important aspect of molecular pear breeding.

QTL for apple agronomic traits, such as disease resistance, insect resistance and dwarfing traits, and fruit quality traits have been mapped onto the apple genome, including resistance to fire blight (*Erwinia amylovora*) (Nybom *et al.* 2012), powdery mildew (*Podosphaera leucotricha*) (Jensen *et al.* 2014), apple scab (*Venturia inaequalis*) (Soriano *et al.* 2014; Franceschi *et al.* 2016), woolly apple aphid (*Eriosoma lanigerum*) (Jensen *et al.*

2014) and rosy apple aphid (*Dysaphis plantaginea*) (Pagliarani *et al.* 2016), as well as fruit quality traits such as fruit firmness (Chagné *et al.* 2014), and titratable acidity (Xu *et al.* 2012). Sun *et al.* (2015) detected 12 apple QTL related to fruit firmness, fruit weight, FA, and sugar content, which were mapped to LG 7.

QTL location with respect to pear agronomic traits has been mainly focused on resistance to diseases such as fire blight, pear scab (*Venturia pyrina*), and black spot (*Fabraea maculata*) (Iketani *et al.* 2001; Dondini *et al.* 2005; Pierantoni *et al.* 2007; Terakami *et al.* 2007; Won *et al.* 2014; Perchepied *et al.* 2015; Terakami *et al.* 2016). The study of fruit quality trait QTL in pear has not been as detailed as in apple because of the relative lack of research on the former crop. Iketani *et al.* (2001) constructed a genetic map of 82 F₁ individuals, using QTL identified by random amplified polymorphic DNA markers, and identified resistance alleles for pear scab and susceptibility alleles for black spot in different linkage groups in ‘Kinchaku’. The research into QTL analysis of pear fruit traits started in 2014, including traits such as fruit firmness, fruit color, fruit ripening date, fruit friction discoloration, and juice content (Wu *et al.* 2014; Yamamoto *et al.* 2014). Yao *et al.* (2017) and Xue *et al.* (2017) identified a red skin color QTL on LG 5. To date, QTL research into pear fruit traits has been insufficient to develop sufficient molecular

markers to improve selection efficiency in pear breeding. To achieve this purpose, it still need to identify new QTL of pear fruit traits by using different hybrid crosses among different pear cultivars.

The aim of the present study was to evaluate the ten main fruit quality traits (single fruit weight, fruit diameter, fruit length, fruit core size, flesh firmness, soluble solid content, skin color, fruit glucose content, fruit sorbitol content, and fruit malic acid content) and to identify the associated QTL that hadn't been reported before, in a population of 90 F₁ individuals from the 'Red Clapp's Favorite' (*Pyrus communis* L.) × 'Mansoo' (*Pyrus pyrifolia*) cross. The QTL associated with the ten fruit traits were identified based on a genetic map constructed in 2017 (Wang *et al.* 2017). The study offers some important insights into map-based cloning of pear genes and marker-assisted breeding for improved fruit quality of pear in the future.

Materials and Methods

Plant material

An F₁ population, consisting of 90 fruiting individuals derived from the cross between 'Red Clapp's Favorite' (*P. communis*) and 'Mansoo' (*P. pyrifolia*), was used for QTL identification. 'Red Clapp's Favorite' was the maternal parent and is a bud mutant of the European pear 'Clapp's Favorite', selected in the US; 'Mansoo' was the male parent and was bred in the Republic of Korea. Hybridization was carried out in 2010, and F₁ plants were grown at the Zhengzhou Fruit Research Institute, Chinese Academy of Agricultural Sciences in Henan Province, China (34° N, 113° E).

Candidate gene searching *in silico*

The regions associated with the QTL on the physical map were identified by mapping the correlative markers. The genes within the QTL regions, together with the functional annotation information, were available on the *P. × bretschneideri* genome website (<http://peargenome.njau.edu.cn>). The candidate genes associated with a specific pear fruit trait were predicted based on their biological functions.

Pear fruit quality evaluation

The fruit traits single fruit weight, fruit diameter, fruit length, fruit core size, flesh firmness, soluble solid content, skin color, fruit glucose content, fruit sorbitol content, and fruit malic acid content were measured in all F₁ individuals in 2016, and the skewness and kurtosis values for the ten fruit traits were analyzed by S.P.S.S. (IBM, Armonk, NY, USA).

Five ripe fruits were randomly collected from each F₁ individual. Fruit length and fruit diameter were measured using Vernier calipers, single fruit weight was determined as

the average weight of the five fruits, flesh firmness was determined using the fruit hardness tester GY-4 (TOP Instrument Company, Zhejiang, China), soluble solid content was determined using a PAL-1 refractometer (ATAGO, Tokyo, Japan), and skin color was classified into one of six levels according to the coloration (red) area on the surface of the ripe fruit (Table 1).

Fruit glucose content, fruit sorbitol content, and fruit malic acid content were determined using the liquid chromatography-tandem mass spectrometry (LC-MS/MS) method (Yuan *et al.* 2017). Methanol, acetonitrile, and ethanol were purchased from Merck (Darmstadt, Germany). Standards were purchased from BioBioPha (Yunnan, China) and Sigma-Aldrich (Saint Louis, MO, USA), and were dissolved in methanol and stored at -20°C for LC-MS/MS analysis. Cryopreserved fruit samples were ground at 30 Hz for 1 min with an electric grinder (MM400, Retsch, Haan, Germany). A sub-sample (100 mg) of powder was extracted overnight at 4°C with 1.0 mL 80% (v/v) methanol, and vortexed three times to achieve full extraction. After centrifugation at 12,000 g for 15 min, the supernatant was decanted from the pellet and dried under nitrogen at 35°C, then reconstituted with 100 μL 30% (v/v) aqueous methanol and vortexed to ensure that it had dissolved fully. The solution was centrifuged at 12,000 g for 15 min, after which the supernatant was decanted away and stored in a vial at 4°C for LC-MS/MS analysis. Instrumental systems for LC-ESI MS/MS analysis included ultra-performance liquid chromatography and tandem mass spectrometry (MS/MS) (Applied Biosystems 4500 QTRAP, <http://www.appliedbiosystems.com.cn/>).

The chromatography conditions included: 1) column: Waters (Milford, MA, USA), ACQUITY UPLC HSS T3, C18, 1.8 μm, 2.1 mm×100 mm; 2) mobile phase: the polar phase was ultrapure water containing 0.1% (v/v) formic acid and the non-polar phase was acetonitrile containing 0.1% (v/v) formic acid; 3) elution gradient: water: acetonitrile 95:5 (v/v) for 0 min, 5:95 (v/v) for 11.0 min, 5:95 (v/v) for 12.0 min, 95:5 (v/v) for 12.1 min, and 95:5 (v/v) for 15.0 min; 4) flow rate: 0.4 mL/min; 5) column temperature: 40°C; 6) injection volume: 5 μL. Eluate was analyzed by mass spectrometry.

QTL analysis of pear fruit traits

MapQTL5.0 (Ooijen 2004) was used to analyze QTL. Interval mapping (IM) was used to detect the QTL for the ten fruit traits. The threshold value was determined for each trait by the permutation test with 1000 replications. The significant log odds score (LOD) threshold was calculated by the permutation test to be 3.5 with the confidence interval ($P < 0.05$), the LOD threshold of 2.5 (Wu *et al.* 2014) was used to identify potential QTL, and the region with the highest QTL score was considered to be the QTL site. MapChart 2.2 software (Voorrips 2002). was used to draw the map.

Table 1: The classified levels of skin color

Score	Coloration (red) area (%)
0	0 (Skin color was green or russet)
1	< 20
2	21-40
3	41-60
4	61-80
5	81-100

Results

Candidate genes involved in pear fruit quality traits

After aligning the QTL regions with the physical positions on the pear genome pseudo-chromosomes *in silico*, seven QTL within a region < 5 cM and with the highest LOD score and the highest variance accounted for of a specific trait, had their corresponding positions identified to permit direct searches for candidate genes. A total of 56 genes linked to the QTL associated with single fruit weight were obtained from the pear genome database, and we also identified 421, 139, 64, 59, 37, and 66 genes related to skin color, fruit core size, fruit diameter, soluble solid content, flesh firmness, and fruit length, respectively, from the pear genome database (Table 4).

Pear fruit quality traits

The fruit traits of the individual F_1 seedlings were measured, namely fruit length, fruit diameter, fruit core size, single fruit weight, flesh firmness, soluble solid content, skin color, fruit glucose content, fruit sorbitol content and fruit malic acid content (Table 2; Fig. 1a–1j). The distribution of single fruit weight was 57.5–569.48 g (mean: 249.43 g), fruit diameter was 46.8–103.62 mm (mean: 77.33 mm), fruit length was 51.21–110.97 mm (mean: 74.89 mm), fruit core size was 18.27–54.16 mm (mean: 34.22 mm), and flesh firmness was 9.1–70.4 N/cm² (mean: 39.85 N/cm²). Soluble solid content ranged from 5.1 to 15.5 Brix% (mean: 10.38 Brix%), while the skin color score ranged from 1 to 5, the distribution of fruit glucose content was 0.84–5.26 µg/g (mean: 2.99 µg/g), and fruit sorbitol content and fruit malic acid content were 0.6–7.07 µg/g (mean: 3.35 µg/g) and 0.18–1.02 µg/g (mean: 0.42 µg/g), respectively. In addition, statistical analysis showed that, for each trait, the absolute values of kurtosis and skewness were both less than 2, and the distribution was unimodal (Table 2), indicating that each of the traits was normally distributed (Fig. 1), and consistent with the typical distribution of quantitative character inheritance controlled by polygenes.

QTL analysis

Ten external and internal fruit quality traits were selected for QTL analysis. Single fruit weight, fruit length, fruit diameter, and skin color affect fruit external quality, while

fruit core size affects the proportion of the fruit which is edible, and flesh firmness, soluble solid content, fruit glucose content, fruit sorbitol content, and fruit malic acid content affect mouth sensation, including sweetness, acidity, and texture. Because the ten traits were normally distributed, QTL analysis was conducted by interval mapping, using MapQTL 5.0 software. Ten significant QTL were detected on LG 3 (n=2), LG 5 (1), LG 11 (6), and LG 12 (1). The QTL for single fruit weight, fruit diameter, fruit length, soluble solid content, fruit glucose content, and fruit malic acid content were all located on LG 11, while the QTL for flesh firmness and fruit sorbitol content were both mapped onto LG 3, and the QTL for SC was localized to LG 5, while the QTL for FCS was located on LG 12 (Fig. 2). The maximum LOD score, the peak position on the map, the interval between the marker and the peak position, the nearest marker, and the percentage of the variance in the trait explained by the marker for each QTL are all shown in Table 3.

Discussion

QTL Mapping in pear is difficult and has rarely been used for pear fruit trait research, because pear is a highly heterozygous self-incompatible plant, with a long juvenile phase (Wu *et al.* 2013). With the development of sequencing technology, high-density linkage maps have started to be used for QTL detection in pear. One of the QTL for single fruit weight was found on LG BYH8 (Zhang *et al.* 2012), which may correspond to LG 11 in the pear reference map (Yamamoto *et al.* 2007). In the current study, the QTL for single fruit weight was detected on LG 11 (Fig. 2), which is in accordance with the result of Yamamoto *et al.* (2007). Moreover, the QTL for fruit diameter and fruit length were also detected on LG 11 (Fig. 2) and were located in almost the same region as that for single fruit weight, suggesting that the three traits might be controlled by the same QTL. It would be a valuable approach for making a comparison of fruit-related QTL between apple and pear because of their collinearity for all LGs (Pierantoni *et al.* 2004; Yamamoto *et al.* 2007). Several QTL related to fruit traits had been reported in apple (Liebhard *et al.* 2003; Kenis *et al.* 2008; Longhi *et al.* 2012; Kunihisa *et al.* 2014). Potts *et al.* (2014) found QTL for single fruit weight, fruit length and fruit diameter on both LG 3 and LG 5 of apple (with similar results in both years of the study); it would be interesting to determine whether the QTL correspond to those identified in our study because of the homologous regions of chromosome 3 and chromosome 11 (Wu *et al.* 2014). Chang *et al.* (2014) also detected two QTL related to single fruit weight on LGs 11 and 5 in apple. All the above findings for apple and pear indicated that the QTL for single fruit weight, fruit length, and fruit diameter were most possibly located on LG 11 and were common to apple and pear.

The identification of candidate genes based on QTL has become a widespread practice in current breeding

Table 2: Statistics of fruit traits in F₁ population

Trait	Mean	SD	Variation	Kurtosis	Skewness	Minimum	Maximum	Range
Fruit length (mm)	74.89	11.674	0.17	-0.347	0.225	51.21	110.97	59.76
Fruit diameter (mm)	77.33	10.375	0.15	-0.316	0.084	46.80	103.62	56.82
Fruit core size (mm)	34.22	5.448	0.19	0.517	0.135	18.27	54.16	35.89
Single fruit weight (g)	249.43	99.88	0.43	-0.063	0.726	57.50	569.48	511.98
Flesh firmness (N/cm ²)	39.85	16.675	0.35	0.109	-0.169	9.10	70.40	61.20
Soluble solid content (%)	10.38	2.661	0.16	1.744	-0.083	5.10	15.50	10.40
Skin colour	0.75	1.039	1.23	-0.665	0.509	0.00	4.00	3.00
Fruit glucose content (μg/g)	2.99	0.899	0.30	-0.062	0.204	0.84	5.26	4.42
Fruit sorbitol content (μg/g)	3.35	1.119	0.33	0.762	0.367	0.60	7.07	6.47
Fruit malic acid content (μg/g)	0.42	0.18	0.43	1.603	1.302	0.18	1.02	0.80

Table 3: QTLs analysis of ten fruit quality traits using interval mapping

Fruit Quality Trait	LOD	Peak position (cM)	Linkage group	Nearest Marker	Distance (cM)	% Exp
Single fruit weight	7.03	65.95	11	Marker360922	1	28.6
Fruit diameter	5.6	67.50	11	Marker356146	0	23.1
		68.12		Marker356147		
		68.75		NAU _{py} 24v		
Fruit length	5.92	65.95	11	Marker360922	1	25
Fruit core size	3.2	141.59	12	Marker76172	0	14.1
Skin colour	16.83	31.88	5	Marker30575	0	56.2
Flesh firmness	3.95	102.64	3	Marker186745	0	17.3
Soluble solid content	6.47	94.48	11	Marker227159	1.55	27.1
Fruit glucose content	5.36	28.64	11	Marker137219	1	27.6
Fruit malic acid content	3.03	5.21	11	Marker155408	1	15.2
Fruit sorbitol content	5.41	102.64	3	Marker186745	0	25.1

Note: % Exp = % of the variance explained by the marker

Table 4: The geneid associated with fruit traits

Fruit traits	The geneid associated with fruit traits								
Single fruit weight	Pbr008131.1	Pbr029588.1	Pbr038262.1	Pbr029567.1	Pbr029576.1	Pbr038287.1	Pbr038257.1	Pbr029582.1	Pbr028416.1
	Pbr017924.1	Pbr030387.1	Pbr038263.1	Pbr029568.1	Pbr029577.1	Pbr038288.1	Pbr038258.1	Pbr029583.1	Pbr029395.1
Skin colour	Pbr017926.1	Pbr030393.1	Pbr038264.1	Pbr029569.1	Pbr029578.1	Pbr038289.1	Pbr038259.2	Pbr029584.1	Pbr029562.1
	Pbr000399.1	Pbr000453.1	Pbr000448.1	Pbr000446.1	Pbr000574.1	Pbr042074.1	Pbr002528.1	Pbr002535.1	Pbr000447.1
	Pbr000405.1	Pbr000572.1	Pbr000472.1	Pbr000536.1	Pbr000503.1	Pbr042080.1	Pbr002481.1	Pbr002548.1	Pbr000573.1
Fruit core size	Pbr000404.1	Pbr000568.1	Pbr000481.1	Pbr000571.1	Pbr000473.1	Pbr042076.1	Pbr002492.1	Pbr002544.1	Pbr000520.1
	Pbr000255.1	Pbr000327.1	Pbr000283.1	Pbr000296.1	Pbr000265.1	Pbr000281.1	Pbr014618.1	Pbr014626.1	Pbr000309.1
	Pbr000300.1	Pbr000332.1	Pbr000302.1	Pbr000259.1	Pbr000292.1	Pbr000267.1	Pbr014634.1	Pbr014607.1	Pbr000303.1
	Pbr000289.1	Pbr000301.1	Pbr000276.2	Pbr000321.4	Pbr000320.1	Pbr000310.1	Pbr014637.1	Pbr014600.1	Pbr000263.1
Flesh firmness	Pbr000318.1	Pbr000272.1	Pbr000294.2	Pbr000278.1	Pbr000322.1	Pbr000257.1	Pbr014636.1	Pbr014602.1	Pbr000270.1
	Pbr032925.1	Pbr032941.1	Pbr032949.1	Pbr032954.4	Pbr032912.1	Pbr032940.1	Pbr032922.1	Pbr032910.1	Pbr032923.1
	Pbr017419.1	Pbr039775.1	Pbr039785.1	Pbr039788.1	Pbr039794.1	Pbr039792.1	Pbr039798.1	Pbr039786.1	Pbr039776.1
	Pbr039707.1	Pbr039777.1	Pbr039774.1	Pbr039781.1	Pbr039803.1	Pbr039799.1	Pbr039801.1	Pbr039806.1	Pbr039790.1

Note: Data were not all shown

programs (Yang *et al.* 2012; Chagné *et al.* 2014; Bastiaanse *et al.* 2016). The candidate gene linked to a specific trait could be used in marker-assisted selection. For example, *PGI* and *ACO1* were associated with the QTL for fruit firmness located on LG 10 in apple (Longhi *et al.* 2012; Chagné *et al.* 2014). Another gene, *LARI*, expressed in apple fruit, was linked to QTL for polyphenolic composition mapping to LG 16 (Chagné *et al.* 2012). In the current study, candidate genes were selected in QTL regions associated with seven fruit traits. It would be interesting in the future to study these candidate genes in breeding projects. The red color of pear fruit is particularly appealing to consumers and the market. The determination of color in fruit is due mostly to the anthocyanin group of flavonoids, which are controlled by both genetic and environmental conditions. Previous studies reported that the anthocyanin

biosynthetic pathway might be regulated by the MYB transcription factor family, as MYB1 and MYB10 were believed to be important transcription factors for red pigmentation in the skin and flesh in apple, respectively (Takos *et al.* 2006; Espley *et al.* 2007). In the present study, we detected one QTL for skin color on LG 5 (Fig. 2). Dondini *et al.* (2008) found one QTL for skin color on LG 4, using the F₁ population from a cross between the two pear cultivars ‘Abbé Fétel’ and ‘Max Red Bartlett’, while Wu *et al.* (2014) detected four QTL for fruit skin color on LG 4 (n=1), LG 13 (1), and LG 16 (2) of pear. Yao *et al.* (2017) also found a skin color QTL on LG 5 of Chinese pear, and screened candidate genes surrounding the LOD peak region (scaffold97.0, 741019bp; scaffold97.0 502339bp), identifying 84 genes in total, from which the *PyMYB114* gene was selected to be responsible for the red skin color

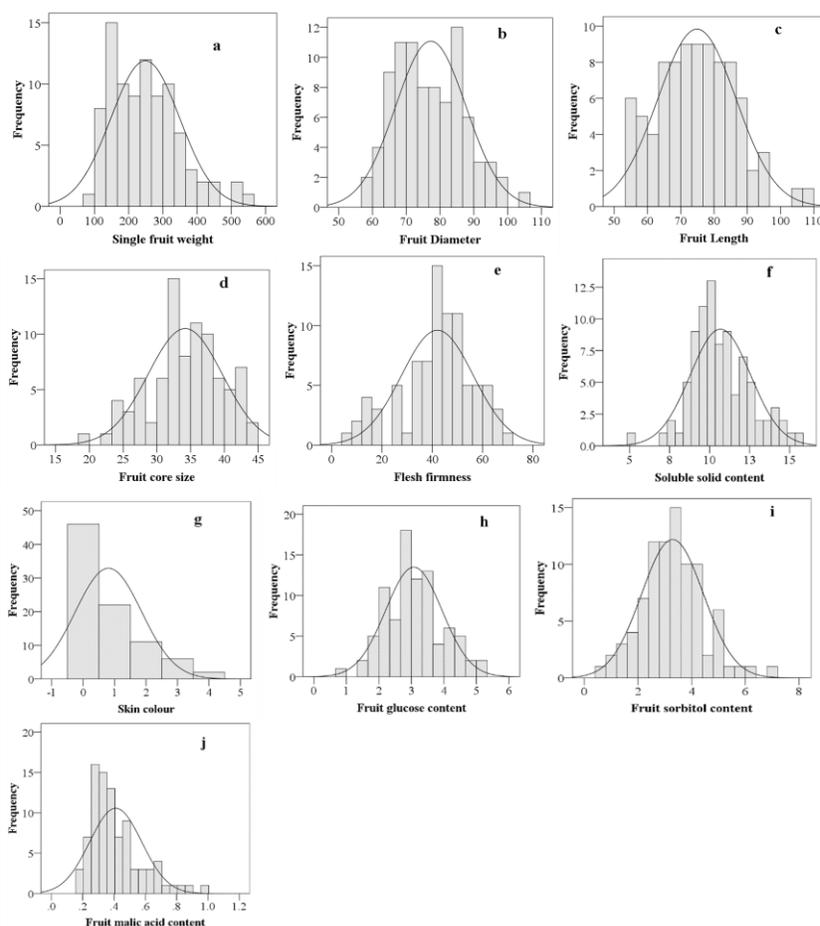


Fig. 1: Distribution of fruit traits in the F₁ population. **Note:** Frequency is shown on the y-ordinate. **a:** distribution of single fruit weight; **b:** distribution of fruit diameter; **c:** distribution of fruit length; **d:** distribution of fruit core size; **e:** distribution of flesh firmness; **f:** distribution of soluble solid content; **g:** distribution of skin color; **h:** distribution of fruit glucose content; **i:** distribution of fruit sorbitol content; **j:** distribution of fruit malic acid content

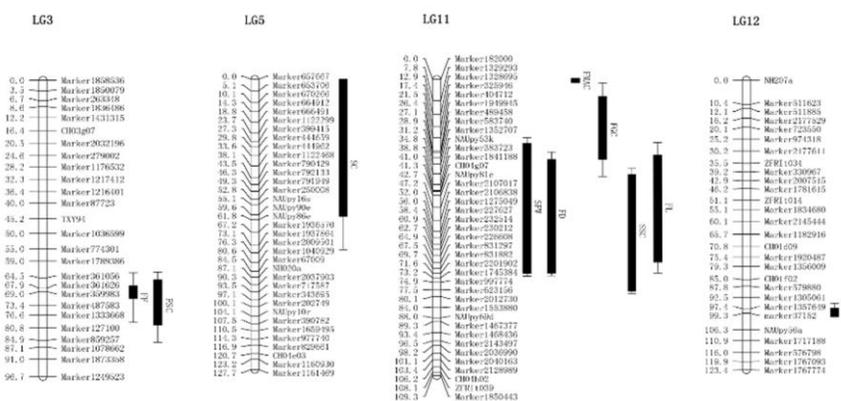


Fig. 2: Significant QTL for ten fruit traits identified on the genetic linkage map of 'Red Clapp's Favorite' x 'Mansoo'. **Note:** The marker and genetic position being shown on the left side and the distribution of the LOD value being shown on the right. The solid bars indicate the QTL identified on the linkage groups with the 99% confidence interval, while the thin lines represent the 95% confidence interval for eight fruit traits, with the exception of FMAC and FCS (FMAC confidence interval was set at the 96.5% and 93.6% levels, while those of FCS were set at the 94.1% and 90.0% levels). Abbreviations: single fruit weight (SF), fruit diameter (FD), fruit length (FL), fruit core size (FCZ), flesh firmness (FF), soluble solid content (SSC), skin color (SC), fruit glucose content (FGC), fruit sorbitol content (FSC) and fruit malic acid content (FMAC)

trait. Xue *et al.* (2017) also reported a skin color QTL on LG 5, using Asian pear populations. In order to determine whether the skin color QTL reported by Xue *et al.* (2017), Yao *et al.* (2017), and ourselves were in the same region, we analyzed the three QTL regions of the corresponding physical locations. We found that all three skin color QTL contained the scaffold97.0 sequence, suggesting that the three skin color QTL might be the same gene.

Several QTL for sugar and organic acid content have been reported in apple and pear. Ma *et al.* (2016) discovered two QTL for fruit malic acid content on LGs 8 and 16, using 191 F₁ individuals. Furthermore, two QTL for fruit glucose content and fruit sorbitol content were detected on one region of LG 3, while one further QTL for fruit glucose content was identified on LG 4 in apple (Ma *et al.* 2016). In the current study, the QTL for fruit glucose content and fruit malic acid content were both located on LG 11, while one QTL related to fruit sorbitol content was identified on LG 3 (Fig. 2). It has been reported that some homologous regions exist between chromosome 3 and chromosome 11 in apple and pear (Wu *et al.* 2013), suggesting that the QTL associated with fruit glucose content in the current study may be the same gene as that reported by Ma *et al.* (2016).

Two QTL for flesh firmness were detected on LG 4 in the Japanese pear F₁ population 'Akiakari'×'Taihaku' (Yamamoto *et al.* 2014), while two pear QTL for flesh firmness were located at the top of LG 3 (Saeed *et al.* 2014). In this study, we also identified one QTL related to flesh firmness, albeit at the bottom of LG 3 (Fig. 2); it would be of interest to determine whether the QTL we detected was the same QTL as that reported by the other groups. In addition, one QTL for soluble solid content was located on LG 11 in the current study (Fig. 2), while the other groups reported QTL for this trait on LGs 5, 10, 14 (Wu *et al.* 2014), 2, 16 (Saeed *et al.* 2014), 4 and 8 (Yamamoto *et al.*, 2014). Soluble solid content QTL were also detected on LGs 2, 3, 6, 8, 9, 10, 12, 13, 14, 15 and 16 in the map of apple (Liebhard *et al.* 2003; Kenis *et al.* 2008; Potts *et al.* 2014; Kuniyama *et al.* 2014; Guan *et al.* 2015). Interestingly, published studies on either apple or pear found that no soluble solid content QTL was located on LG 11, which conflicts with the finding of the current study. Possible reasons for this discrepancy might be due to: different cultivars were included in the different studies; the soluble solid content trait was strongly affected by the environment; and soluble solid content was a complex trait, involving various monosaccharides, organic acids, amino acids, polyphenols, soluble pectin, etc.

Conclusion

Ten QTL associations with ten fruit quality traits were detected. And the study obtained 56, 421, 139, 64, 59, 37 and 66 candidate genes related to single fruit weight, skin color, fruit core size, fruit diameter, soluble solid content, fruit firmness and fruit length from the pear genome database, respectively.

Acknowledgements

This work was supported by the Agricultural Science and Technology Innovation Program (CAAS-ASTIP), the Earmarked Fund for China Agriculture Research System (CARS-28) and the Central Public-interest Scientific Institution Basal Research Fund (CPSI-BRF).

References

- Bastiaanse H, HCM Bassett, C Kirk, SE Gardiner, C Deng, R Groenworld, D Chagne, VGM Bus (2016). Scab resistance in 'Geneva' apple is conditioned by a resistance gene cluster with complex genetic control. *Mol Plant Pathol* 17:1–14
- Chagné D, D Dayatilake, R Diack, M Oliver, HS Ireland, A Watson, SE Gardiner, JW Johnston, RJ Schaffer, S Tustin (2014). Genetic and environmental control of fruit maturation, dry matter and firmness in apple (*Malus × domestica* Borkh.). *Hortic Res* 1:1–12
- Chagné D, C Krieger, M Rassam, M Sullivan, J Fraser, CM Andre, M Pindo, M Troglio, SE Gardiner, RA Henry-Kirk, AC Allan, T Mcghee, WA Laing (2012). QTL and candidate gene mapping for polyphenolic composition in apple fruit. *BMC Plant Biol* 12:12
- Chang YS, R Sun, HH Sun, YB Zhao, YP Han, DM Chen, Y Wang, XZ Zhang, ZH Han (2014). Mapping of quantitative trait loci corroborates independent genetic control of apple size and shape. *Sci Hortic* 174:126–132
- Dondini L, L Pierantoni, V Ancarani, MD'Angelo, KH Cho, IS Shin, S Musacchi, SJ Kang, S Sansavini (2008). The inheritance of the red colour character in European pear (*Pyrus communis*) and its map position in the mutated cultivar 'Max Red Bartlett'. *Plant Breed* 127:524–526
- Dondini L, L Pierantoni, F Gaiotti, R Chioldini, S Tartarini, C Bazzi, S Sansavini (2005). Identifying QTLs for fire-blight resistance via a European pear (*Pyrus communis* L.) genetic linkage map. *Mol Breed* 14:407–418
- Espley RV, RP Hellens, J Putterill, DE Stevenson, S Kutty-Amma, AC Allan (2007). Red colouration in apple fruit is due to the activity of the MYB transcription factor, MdMYB10. *Plant J*, 49:414–427
- Franceschi PD, V Cova, S Tartarini, L Dondini (2016). Characterization of a new apple S-RNase allele and its linkage with the Rvi5 gene for scab resistance. *Mol Breed* 36:1–11
- Guan Y, C Peace, D Rudell, S Verma, K Evans (2015). QTLs detected for individual sugars and soluble solids content in apple. *Mol Breed* 35:1–13
- Iketani H, K Abe, T Yamamoto, K Kotobuki, Y Sato, T Saito, O Terai, N Matsuta, T Hayashi (2001). Mapping of Disease-Related Genes in Japanese Pear using a Molecular Linkage Map with RAPD Markers. *Breed Sci* 51:179–184
- Jensen PJ, G Fazio, N Altman, C Praul, TW Mcnellis (2014). Mapping in an apple (*Malus × domestica*) F₁ segregating population based on physical clustering of differentially expressed genes. *BMC Genomics* 15:261
- Kenis K, J Keulemans, MW Davey (2008). Identification and stability of QTLs for fruit quality traits in apple. *Tree Genet Genom* 4:647–661
- Kuniyama M, S Moriya, K Abe, K Okada, T Haji, T Hayashi, H Kim, C Nishitani, S Terakami, T Yamamoto (2014). Identification of QTLs for fruit quality traits in Japanese apples: QTLs for early ripening are tightly related to preharvest fruit drop. *Breed Sci* 64:240–251
- Liebhard R, M Kellerhals, W Pfammatter, M Jertmini, C Gessler (2003). Mapping quantitative physiological traits in apple (*Malus × domestica* Borkh.). *Plant Mol Biol* 52:511–526
- Longhi S, MT Hamblin, L Trainotti, CP Peace, R Velasco, F Costa (2013). A candidate gene based approach validates Md-PG1 as the main responsible for a QTL impacting fruit texture in apple (*Malus × domestica* Borkh.). *BMC Plant Biol* 13:37
- Longhi S, M Moretto, R Viola, R Velasco, F Costa (2012). Comprehensive QTL mapping survey dissects the complex fruit texture physiology in apple (*Malus × domestica* Borkh.). *J Exp Bot* 63:1107–1121

- Ma B, S Zhao, BH Wu, DM Wang, Q Peng, A Owiti, T Fang, L Liao, C Ogutu, SS Korban, SH Li, YP Han (2016). Construction of a high density linkage map and its application in the identification of QTLs for soluble sugar and organic acid components in apple. *Tree Genet Genomics* 12:1–10
- Nybohm H, A Mikiciński, L Garkava-Gustavsson, J Schic, M Lewandowski, P Sobiczewski (2012). Assessment of fire blight tolerance in apple based on plant inoculations with *Erwinia amylovora* and DNA markers. *Trees* 26:199–213
- Ooijen JM (2004). *MapQTL® 5. Software for the Mapping of Quantitative Trait Loci in Experimental Populations*. Kyazma BV, Wageningen, Netherlands
- Pagliarani G, E Dapena, M Miñarro, C Denancé, Y Lespinasse, E Rat-Morris, M Troglio, CE Durel, S Tartarini (2016). Fine mapping of the rosy apple aphid resistance locus Dp-fl on linkage group 8 of the apple cultivar 'Florina'. *Tree Genet Genom* 12:1–12
- Perchepped L, D Leforestier, E Ravon, P Guérif, C Denance, M Tellier, S Terakami, T Yamamoto, M Chevalier, Y Lespinasse, CE Durel (2015). Genetic mapping and pyramiding of two new pear scab resistance QTLs. *Mol Breed* 35:197
- Pierantoni L, L Dondini, KH Cho, IS Shin, F Gennari, R Chiodini, S Tartarini, SJ Kang, S Sansavini (2007). Pear scab resistance QTLs via a European pear (*Pyrus communis*) linkage map. *Tree Genet Genomics* 3:311–317
- Pierantoni L, KH Cho, IS Shin, R Chiodini, S Tartarini, L Dondini, SJ Kang, S Sansavini (2004). Characterisation and transferability of apple SSRs to two European pear F1 populations. *Theor Appl Genet* 109:1519–1524
- Potts SM, MA Khan, Y Han, MM Kushad, SS Korban (2014). Identification of Quantitative Trait Loci (QTLs) for Fruit Quality Traits in Apple. *Plant Mol Biol Rep* 32:109–116
- Saeed M, L Brewer, J Johnston, TK Mcghee, SE Gardiner, JA Heyes, D Chagné (2014). Genetic, metabolite and developmental determinism of fruit friction discolouration in pear. *BMC Plant Biol* 14:241
- Soriano JM, M Madduri, JG Schaart, A Burgh, MPW Kaauwen, BITZ Lidija, R Groenwold, R Velasco, EV Weg, H Schouten (2014). Fine mapping of the gene Rvi18 (V25) for broad-spectrum resistance to apple scab, and development of a linked SSR marker suitable for marker-assisted breeding. *Mol Breed* 34:2021–2032
- Sun R, Y Chang, F Yang, Y Wang, H Li, Y Zhao, D Chen, T Wu, X Zhang, Z Han (2015). A dense SNP genetic map constructed using restriction site-associated DNA sequencing enables detection of QTLs controlling apple fruit quality. *BMC Genom* 16:747
- Takos AM, FW Jaffé, SR Jacob, J Bogs, SP Robinson, AR Walker (2006). Light-induced expression of a MYB gene regulates anthocyanin biosynthesis in red apples. *Plant Physiol* 142:1–17
- Terakami S, S Moriya, Y Adachi, M Kuniyama, C Nishitani, T Saito, K Abe, T Yamamoto (2016). Fine mapping of the gene for susceptibility to black spot disease in Japanese pear (*Pyrus pyrifolia* Nakai). *Breed Sci* 66:271–280
- Terakami S, Y Adachi, H Iketani, Y Sato, Y Sawamura, N Takada, C Nishitani, T Yamamoto (2007). Genetic mapping of genes for susceptibility to black spot disease in Japanese pears. *Genome* 50:735–741
- Voorrips RE (2002) MapChart: software for the graphical presentation of linkage maps and QTLs. *J Hered* 93:77–78
- Wang L, XG Li, L Wang, HB Xue, J Wu, H Yin, SL Zhang (2017). Construction of a high-density genetic linkage map in pear (*Pyrus communis* × *Pyrus pyrifolia* Nakai) using SSRs and SNPs developed by SLAF-seq. *Sci Hortic* 218:198–204
- Won K, H Bastiaanse, YK Kim, JH Song, SS Kang, HC Lee, KH Cho, LR Brewer, G Singla, SE Gardiner, D Chagne, V Bus (2014). Genetic mapping of polygenic scab (*Venturia pirina*) resistance in an interspecific pear family. *Mol Breed* 34:2179–2189
- Wu J, LT Li, M Li, MA Khan, XG Li, H Chen, H Yin, SL Zhang (2014). High-density genetic linkage map construction and identification of fruit-related QTLs in pear using SNP and SSR markers. *J Exp Bot* 65:5771–5781
- Wu J, ZW Wang, ZB Shi, S Zhang, R Ming, SL Zhu, MA Khan, ST Tao, SS Korban, H Wang, NJ Chen, T Nishio, X Xu, L Cong, KJ Qi, XS Huang, YT Wang, X Zhao, JY Wu, C Deng, CY Gao, WL Zhou, H Yin, GH Qin, YH Sha, Y Tao, H Chen, YN Yang, Y Song, DL Zhan, J Wang, LT Li, MS Dai, C Gu, YZ Wang, DH Shi, XW Wang, HP Zhang, L Zeng, DM Zheng, CL Wang, MS Chen, GB Wang, L Xie, V Sovero, SF Sha, WJ Huang, SJ Zhang, MY Zhang, JM Sun, LL Xu, Y Li, X Liu, QS Li, JH Shen, JY Wang, RE Paull, JL Bennetzen, J Wang, SL Zhang (2013). The genome of the pear (*Pyrus bretschneideri* Rehd.). *Genome Res* 23:396–408
- Xu K, A Wang, S Brown (2012). Genetic characterization of the Ma locus with pH and titratable acidity in apple. *Mol Breed* 30:899–912
- Xue H, T Shi, F Wang, H Zhou, J Yang, L Wang, S Wang, Y Su, Z Zhang, Y Qiao, X Li (2017). Interval mapping for red/green skin colour in Asian pears using a modified QTL-seq method. *Hortic Res* 4:1–10
- Yamamoto T, S Terakami, N Takada, S Nishio, N Onoue, C Nishitani, M Kuniyama, E Inoue, H Iwata, T Hayashi, A Itai, T Saito (2014). Identification of QTLs controlling harvest time and fruit skin colour in Japanese pear (*Pyrus pyrifolia* Nakai). *Breed Sci* 64:351–361
- Yamamoto T, T Kimura, S Terakami, C Nishitani, Y Sawamura, T Saito, K Kotobuki, T Hayashi (2007). Integrated reference genetic linkage maps of pear based on SSR and AFLP markers. *Breed Sci* 57:321–329
- Yang B, L Dougherty, M Li, G Fazio, L Cheng, K Xu (2012). A natural mutation-led truncation in one of the two aluminum-activated malate transporter-like genes at the Ma locus is associated with low fruit acidity in apple. *Mol Genet Genom* 287:663–678
- Yao GF, ML Ming, AC Allan, C Gu, LT Li, X Wu, RZ Wang, YJ Chang, KJ Qi, SL Zhang, J Wu (2017). Map-based cloning of the pear gene MYB114 identifies an interaction with other transcription factors to coordinately regulate fruit anthocyanin biosynthesis. *Plant J* 92:437–451
- Yuan YF, YP Zhao, JL Yang, YM Jiang, F Lu, YX Jia, B Yang (2017). Metabolomic analyses of banana during postharvest senescence by 1H-high resolution-NMR. *Food Chem* 218:406–412
- Zhang RP, J Wu, XG Li, MA Khan, H Chen, SS Korban, SL Zhang (2012). An AFLP, SRAP, and SSR genetic linkage map and identification of QTLs for fruit traits in pear (*Pyrus L.*). *Plant Mol Biol Rep* 31:678–687