Analysis of Genetic Similarity in Selected Peach Cultivars and Their Hybrids by Microsatellite DNA Markers

Jau-Yeuh Wang, Chia-Wei Song, Min-Huai Liou, and Shyi-Kuan Ou

Division of Horticulture, Agricultural Research Institute, Taichung 413, Taiwan

Keywords: Peach, Microsatellite, Genetic similarity, Fruit characteristics ABSTRACT

The typical fruit characteristics of 34 selected peach cultivars and their hybrid seedlings were conducted for genetic similarity and identification by microsatellite markers. Twelve polymorphic microsatellite locus were screened from 17 primer pairs, and certainly obtained 3~9 alleles in each microsatellite. An AC/GT repeats (UPD-96-005) could be used to distinguish stony hard cultivar of 'Ying Ge Tao' and 'Ba Yue Tao'. Furthermore, 34 genotypes were assessed genetic similarity by total 87 microsatellite markers and cluster analysis of UPGMA that divided into two main groups. The first group contained 16 genotypes of 'Ying Ge Tao' and its hybrid progenies, and the second group was composed by 18 genotypes of 'Premier' and its hybrid progenies. The dendrogram showed the progenies would be grouping with parent and accompanied with the level of TTS (total soluble solid) and acidity separately. The results reveal that microsatellite DNA markers are useful in cultivars identification and allow to early selection of the progenies with superior fruit characteristics.

INTRODUCTION

Peach (*Prunus persica* (L.) Batsch) is one of important economic crops in Taiwan. According the Yearly report of Taiwan's Agriculture in 2004, the growing area was 2,942 ha and year's output value was about 0.88 billion NT dollars. Since 1950, high-chill cultivars were introduced and planted at more than 1500 m above sea level; while the cultural practices caused soil erosion and water contamination in higher mountains. A breeding program had been undertaken by Agricultural Research Institute from 1980 to solve the problems. The breeding objectives were to obtain high quality and low chilling cultivars. Nowadays, there are three new cultivars had been released for low land cultivation. The breeding of improved woody perennial fruit is long-term and expensive. Themarker-assisted selection (MAS) offers the possibility of saving the time and expense for plantation and evaluation in the field. The purpose of this study is to establish the microatellite markers as one of the useful MAS tools in our peach breeding program.

MATERIALS AND METHODS

- Plant Materials and DNA Extraction: 34 genotypes from breeding program, including 3 parents of 'Premier', 'Ying Ge Tao' and 5-40N. Genomic DNA was isolation from leaf samples of each genotype and following by the DNeasy® Plant Kit (Qiagen).
- PCR Amplification and Microsatellite Analysis: 17 flanking microsatellite sequences had been used. (Cipriani et al., 1999). PCR was performed in a volume of $25 \,\mu$ l containing 1.25 u Taq polymerase (Roche-FastStart), 1xPCR buffer, 2.0 mM MgCl₂, 200 μ M of each dNTP, 0.2 μ M of each primer, and 50 ng template DNA. Reactions were performed on a Gene Amp 9700® by an initial denaturation for 5 min at 95°C followed by 35 cycles of 45 s at 94°C, 45 s at 56°C ~58°C, 2 min at 72°C; and a final extension of 8 min at 72°C. Products were separated on a 3% Metaphor agarose gel and stained to check and determine approximately size of amplification fragments.
- Genetic Similarity Analysis and Genotype Identification: Polymorphic loci were recorded and genetic similarity was measured by program of GelComparII version 3.5 (Applied Maths, 2003). The similarity coefficients were calculated according to Dice's, and dendrogram produced by clustering the data with UPGMA.

RESULTS AND DISCUSSION

S	imilarity coefficient (%)	Line	Remark (Combinati	on	
£7	* * * * * * * * * * * *					•Dendrogram from microsatellite data grouped genotypes according to putative pedigrees
		E16-7	sweet	lgxPr		
		D16-6 E16-14	sweet	lgxPr lgxPr		and fruit characteristics such as the level of TTS, acidity and skin pubescence separately.
		D16-7	sweet	lgxPr		
		B3-2-63	sweet	lgxPr		This technology can be applying to early selection of progenies with superior fruit
		B3-2-64	sweet	IgxPr		
		E16-16	sour	lgxPr	1	characteristics (Fig. 1).
		E16-28	sour	lgxPr		
		E16-2 E16-15	sour	lgxPr		•Dinucleotide repeats are most abounding in plant and AG/CT or AC/GT repeats are more
		Ying Ge Tao (E)	sour.early flower	igxer		
		Ying Ge Tao	sour			enriching. 12 microsatellites revealed polymorphism, and several markers presented a
		Ba Yue Tao	sour		2	
		Nan Shan Taian	Tao sweet,big,fibrous		3	high discrimination for cultivars identification (Fig. 2, 3).
		swatow	bitter,dwarf,sour		1	ingli discrimination for cultivars identification (Fig. 2, 5).
		Ku Tau	bitter,sour		4	
		23-26	very sweet			
		B4-2-49	very sweet	23-26 23-26xPr		
		E131-9	very sweet	C1-57-2		
		B4-2-20	very sweet	Pr	5	
		Spring Honey	sweet		۲.	
		C1-57-3	very sweet	23-26xPr		
		premier	sweet			
	-	C1-74-26	very sweet	TuTu	i -	
		C1-46-1	sweet	Prx5-40N Prx5-40N		
		C1-46-4	sweet	Prx5-40N Prx5-40N	6	
	4	C1-46-7	sweet	Prx5-40N		
		C1-46-11	sweet	Prx5-40N	1	
		E16-48	sour	IgxPr	1	
		E16-51	sour	lgxPr	7	
L		5-40N	sour		8	
		9-17N	sour			
	1 0 1 0 0 1		0			Fig. 2 Amplification profile in 24 constructs of Fig. 2 Amplification profile in 24 constructs of
Fig. 1 Dendrogram of 34 genotypes of peach based						Fig. 2 Amplification profile in 34 genotypes ofFig. 3 Amplification profile in 34 genotypes of
on microsatellitea and cluster analysis of UPGMA.						peach by microsatellite primer of UDP96-005. peach by microsatellite primer of UDP96-015.
on incrosatentea and cluster analysis of UPGNIA.					•	peace by merosatemic primer of OD190-000. peace by merosatemic primer of OD190-015.



行政院農業委員會農業試驗所 Agricultural Research Institute, COA