

Effectiveness of Molecular Markers for Detecting the Non-astringent Trait of Chinese Persimmon and a Trial to Identify the Gene Conferring Chinese PCNA Trait in 'Luo Tian Tian Shi'

Keizo Yonemori¹, Soichiro Nishiyama², Hisayo Yamane² and Akihiko Sato³

(¹) Ryukoku University, Otsu, Japan; (²) Kyoto University, Kyoto, Japan; (³) NIFTS, Higashi Hiroshima, Japan

INTRODUCTION

CHINESE-PCNA trait in 'Luo Tian Tian Shi'

- **Pollination-constant and non-astringent (PCNA)** type fruits stably loss their astringency on the tree.
 >> **one of the most important trait in persimmon breeding**
- PCNA-trait in Chinese 'Luo Tian Tian Shi' is conferred by single **dominant locus (CPCNA/cpcna)**.
 >> **regarded as a promising germplasm**

- We developed **7 molecular markers** by AFLP analysis. These markers were co-segregated with *CPCNA*, and we constructed genetic map at *CPCNA* locus.

>> **practical application of RO-2 marker in breeding programs**
 (Ikegami et al. 2011, JJSHS)

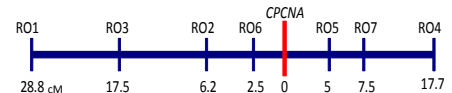
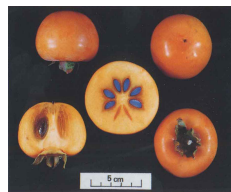


Fig. 1 Genetic map of the CPCNA region
 Mapping Population : L-O ('Luo Tian Tian Shi' X Japanese PCNA cultivar 'Okugosho' | 81 F₁ individuals)

EXP. 1

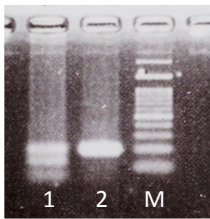
Validity of the RO-6 marker in two segregated F₁

F₁ population

- **L-Y population** | 109 individuals ... 'Luo Tian Tian Shi' X 'Yotsumizo'
- **L-I population** | 74 individuals ... 'Luo Tian Tian Shi' X 'Iwasedo'



- **Phenotyping** | soluble phenol content & tannin cell size (2004-2008)



- **Genotyping** | RO-6 marker (CAPS) which is the closest to *CPCNA*

Fig. 2 Detection of RO6 genotype with the CAPS marker
 1 : PCNA individual 2 : non-PCNA individual

Results

Table 1 Segregation of the phenotype and the genotype of RO-6 marker in two F₁ population

F ₁ Population	Phenotype	RO-6		Fitness ratio
		Present	Absent	
L-Y	PCNA	54	1	0.991
	non-PCNA	0	54	
L-I	PCNA	30	0	1.000
	non-PCNA	0	34	

EXP. 2

Comparative genomics using *Diospyros lotus*

- Almost all cultivated *D. kaki* is hexaploid (2n = 6x = 90) and has highly heterozygotes in alleles
 >> much complexities of allele segregation
 >> **chromosome walking using a close diploid relative *D. lotus***

Results

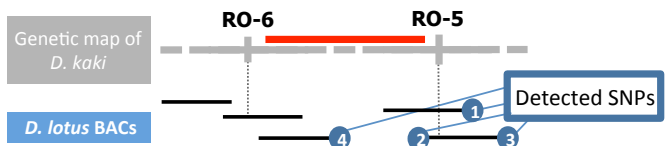


Fig. 3 Map construction at the CPCNA region
 SNPs is detected in *D. lotus* F₁ population for constructing *D. lotus* genetic map

Table 2 Detection of the SNPs markers in F₁ population of *D. lotus*

individual	genotype			
	RO5	RO6	SNP1	SNP4
Male	-	-	-	-
Female	+	+	+	+
KK2 1	-	-	-	-
KK2 2	+	+	+	-
KK2 3	-	-	-	+
KK2 4	-	-	-	+
KK2 5	-	-	-	-
KK2 6	-	-	-	+
KK2 7	+	ND	+	ND
KK2 8	+	ND	+	ND

No co-segregation between RO5 and RO6

>> no linkage in *D. lotus*??
 >> **We need more individuals for constructing genetic map in *D. lotus***

CONCLUSIONS

- RO-6 marker is highly co-segregated with *CPCNA* in L-Y and L-I population.
- We constructed *D. lotus* BAC contig at *CPCNA* region.
 >> **Enabling to construct the genetic map using SNPs on the BAC sequence?**