

Development of SSR markers to study diversity in the genus *Paeoniae*

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작약 유전다양성 검증을 위한 SSR 마커 개발 및 집단구조 분석

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Objectives

The purpose of our study was to develop and characterize novel polymorphic microsatellite markers for assessing genetic diversity, understanding population structure, genotype identification and molecular breeding in *Paeonia*.

Materials and Methods

○ Materials

The accessions of *Paeonialactiflora* PALL were obtained from National Institute of Horticultural & Herbal Science, Rural Development Administration (RDA).

○ Methods

- Construction of SSR - enrich library and characteristics of an enriched library (ABI 3730xl DNA sequencer), primer design and marker development (ARGOS 1.46). SSR markers were checked by agarose and acrylamide gel and polymorphic markers were selected. All the accessions were amplified by these polymorphic markers and genotyped by ABI 3500 with a BigDye terminator kit (Applied Biosystems).

- Basic statistics, including total number of alleles, allele frequency, accession- specific alleles, major allele frequency (M_{AF}), and polymorphic information content (PIC), were calculated from shared allele frequencies using PowerMarker V3.23. Genetic distance between each pair of accessions using the genetic analysis package POPGENE version 1.31. The UPGMA algorithm was used to construct an unrooted phylogram from a distance matrix using MEGA4 software.

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Results

In this study, 14 novel polymorphic microsatellite markers were developed and characterized through construction of an SSR-enriched library from genomic DNA of *Paeonia lactiflora* PALL. In total, 57 alleles across 85 *Paeonia* accessions were detected, with an average of 4.1 alleles per locus. Values of major allele frequency (M_{AF}), genetic diversity (GD) and polymorphism information content (PIC) ranged from 0.39 to 0.88 (mean = 0.619), from 0.21 to 0.71 (mean = 0.50) and from 0.19 to 0.67 (mean = 0.43), respectively. The mean genetic distance is 0.5559, indicating that there was a wide variation among the *Paeonia* accessions.

Table 1. General characteristics of the 14 new polymorphic markers developed for *Paeonia*.

Primer name	NA ^a	M_{AF} ^b	GD ^c	PIC ^e
knu14	7.0	0.39	0.71	0.66
knu21	2.0	0.88	0.21	0.19
knu32	3.0	0.80	0.34	0.31
knu35	7.0	0.62	0.56	0.52
knu39	6.0	0.41	0.68	0.62
knu73	3.0	0.59	0.52	0.43
knu78	6.0	0.47	0.71	0.67
knu94	2.0	0.77	0.35	0.29
knu114	3.0	0.54	0.51	0.39
knu115	3.0	0.56	0.50	0.38
knu116	4.0	0.55	0.57	0.49
knu124	3.0	0.78	0.36	0.32
knu136	5.0	0.48	0.58	0.49
knu154	3.0	0.72	0.41	0.33
Mean	4.1	0.61	0.50	0.43

^a: Number of alleles, ^b: Major allele frequency, ^c: Gene diversity, ^d: Polymorphism information content

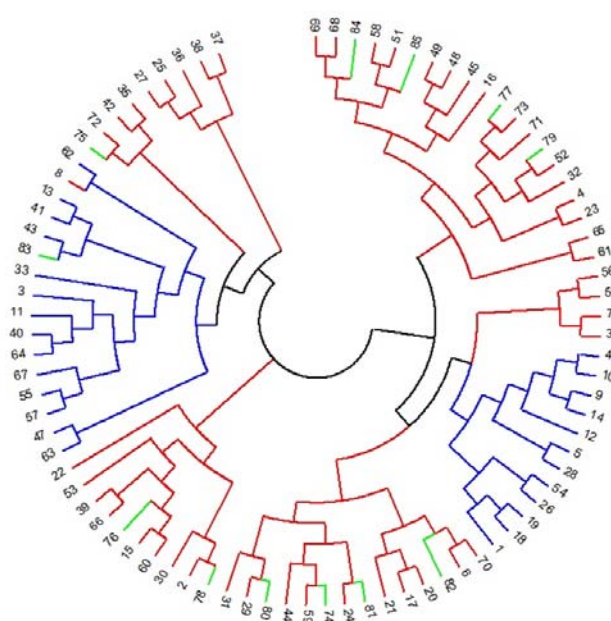


Fig. 1. UPGMA dendrogram showing genetic relationships among 85 genotypes collected from various parts of Korea.

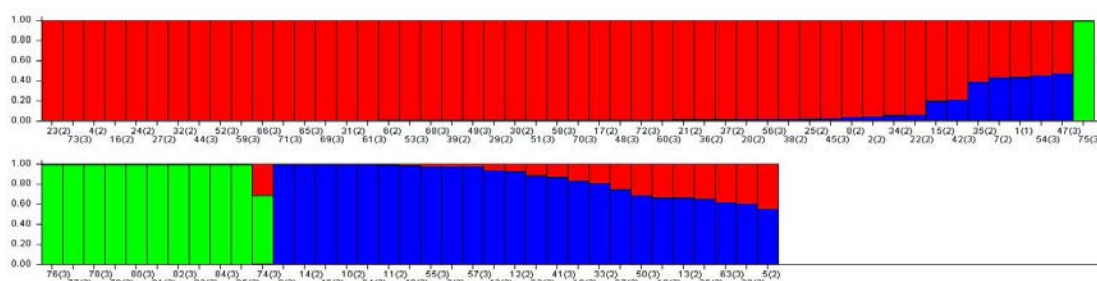


Fig. 2. Assignment of 85 genotypes used in this study to three genetic groups using STRUCTURE software (Pritchard et al. 2000).