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Sequence comparisons among 5' flanking regions with cloning of *SP11/SCR* genes in *Brassica rapa* L.

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Objectives

Here we tried to the cloning of 12 alleles of *SP11* from *Brassica rapa* by using the PCR method, and comparison of the deduced nucleotide sequences.

Materials and Methods

1. Material

Plant nine(Class I) and three(Class II) inbred lines of *Brassica rapa* were used as PCR cloning.

2. Methods

PCR cloning, Sequencing

Results and Discussion

SP11/SCR have been identified in and have definitively established that *SP11/SCR* is the sole male determinant of SI in the genus *Brassica*. It was revealed by deletion analyses of the *S⁰-SP11* promoter-gus fusions that the region around -192 bp contains the element(s) required for GUS expression in the tapetum, and that the region between -124 and -143 represents the minimal promoter region for pollen expression. In this paper, we extend the analysis of the 5'-flanking regions of *SP11/SCR* genes by comparing the genomic sequences of the genera *Brassica* and *Raphanus*. Twenty-two inbred were used as plant materials. The 5'-flanking regions of *SP11/SCR* genes amplified with a set of primers, *SP11*-PF designed based on the 5'-flanking region of *S⁰* and *SP11*-PR2 and *SP11*-PR3 designed based on the conserved signal peptide coding region of four alleles of *SP11* and sequenced.

Alignment of these sequences are highly conserved (53-96% identity) in the 5'-flanking region of *SP11/SCR* genes. Similarities between the nucleotide sequences from the *B. rapa* and *B. oleracea* lines ranged from 76% to 96%, not significantly higher than the similarities of *R. sativus* lines (from 60% to 91%). In addition, the position and sequence of the putative TATA box were conserved in *B. rapa*, *B. oleracea* and *R. sativus* lines. From database searches in the 5'-flanking region, these repeats and a putative pollen-specific sequences were not also found in the 5'-flanking region of *B. oleracea* and *R. sativus* lines. Moreover other common repeat or palindromic sequence was not found in these sequences.

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