(05-1-81)

Characterization of eight *Rumex* species by FISH (fluorescence *in situ* hybridization) and 5S rDNA spacer sequences

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Objectives

We applied the FISH technique to the genus *Rumex*, in order to analyze rDNA sites on the chromosomes. We also attempted to determine the inter-specific relationships among species, using 5S rDNA spacer sequences.

Materials and Methods

1. Material

Plant - Eight *Rumex* species (*R. crispus, R. japonicus, R. longifolius, R. nipponicus, R. obtusifolius, R. maritinus, R. acetocella* and *R. acetosa*) were collected from natural Korean populations, and were grown in a plant growth facility at Chungnam National

2. Methods

The 45S rDNA repeat units were labeled with biotin-16-dUTP via nick translation. The 5S rDNA probe was obtained by PCR using the total cucumber genomic DNA as template DNA, and labeled with digoxigenin-11-dUTP. The 5S sequences were determined using the automatic DNA sequencer (ABI377)

Results and Discussion

Using both molecular and cytogenetic methods, we have examined and characterized eight *Rumex* species distributed throughout the Korean peninsula. The somatic metaphase chromosomes in the *Lapathum* group were 2n=6x=60 in three of the species (*R. crispus, R. japonicus* and *R. longifolius*), 2n=5x=50 in one species (*R. nipponicus*), and 2n=4x=40 in two of the species (*R. obtusifolius* and *R. maritinus*). In the two dioecious groups, the chromosome numbers were 2n=6x=42 (female) and 43 (male) in *R. acetocella*, and 2n=2x=14 (female) and 15 (male) in *R. acetosa*. Using the FISH method, we physically mapped the 5S and 45S rDNA genes on the chromosomes of all the studied species. All of the species, except *R. japonicus*, possessed four 45S rDNA sites, whereas the 5S rDNA gene sites were variable, ranging between 2 and 8. Polymorphic 5S rDNA sites at the same ploidy level were found among species in the subgenus *Lapathum* group. Nucleotide sequence analysis revealed that the 5S rDNA spacer also varied between the species, ranging between 308 and 315 bp, except for each 120 bp coding region. FISH and 5S rDNA spacer sequencing were successfully applied to the identification and characterization of the eight *Rumex* species examined in this study.

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