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Modified random amplification microsatellite polymorphism (mRAMP)

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Simple sequence repeats (SSR) are widely distributed in eukaryotic genome and serve as an informative source of genetic marker for molecular mapping and differentiation of genetic diversity. Though SSR markers are highly variable and codominant, it is hard to develop SSR markers between plant lines of close genetic background. To overcome this problem, we developed an alternative marker system, named modified random amplification microsatellite polymorphism (mRAMP), This modified RAMP uses 18 mers of SSR with 2 selective nucleotides at the 3' end (not at the 5' end as in RAMP) as a forward direction primer and commercial arbitrary random primers as a reverse direction primer to generate PCR bands in semi-random manner. mRAMP was run under the same PCR reaction conditions as in RAMP. mRAMP showed different amplification patterns in PCR bands compared to those of RAMP and allowed to thoroughly differentiate the sequence discrepancy of the two nucleotides adjacent to the 3' end of the same SSR motif for specific primers. Sequence analysis of several PCR bands of mRAMP revealed that they all contained the same SSR motif corresponding to the target sequence of specific primers. The number of PCR bands by mRAMP was dependent upon the combinations among specific and arbitrary primers. For any 2-nucleotide specific primer AC, the most frequent SSR motif in the pepper EST data base (http://genepool.kribb.re.kr/), generated more PCR bands than other rare motifs such as AAG or AATG. Similar pattern was found in UBC603 out of 20 arbitrary primers. When mRAMP markers were placed on the pepper map of the intraspecific cross between *Capsicum annuum* 'CM334' and 'Chilsungcho', all linked markers were found scattered on the whole linkage groups without skewness. mRAMP was proved to be a useful marker system for generating large number of dominant markers for pepper lines with close genetic backgrounds.

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