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Marker-Assisted Selection of Disease and Pest Resistant Mungbean Lines Using Cel-I Genotyping

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Abstract

Mungbean (*Vigna radiata*) is cultivated on about 6 million hectares worldwide. Short duration varieties are highly attractive as a rotation crop in cereal farming systems in South and Southeast Asia. Mungbean is consumed as grains or sprouts, and the green pods are eaten as a vegetable; the grains are processed into a variety of products such as noodles, sweets or drinks. Major constraints for mungbean yield and profitability are diseases caused by Mungbean yellow mosaic virus (MYMV) and damage from bruchid beetles (*Callosobruchus* sp.), a storage pest. Quantitative trait loci analyses in populations segregating for Mungbean yellow mosaic virus and bruchid resistance have led to the identification of markers linked with resistance genes. Single nucleotide polymorphic (SNP) markers mapping to chromosomes 3, 4 and 5 showed strong association with bruchid resistance, while markers on chromosomes 2, 5, 7, 9 and 10 were found associated with MYMV resistance. Selection based on these markers facilitates breeding of disease- and pest-resistant varieties. However, genotyping SNP markers in the absence of specialised equipment requires their conversion into polymerase chain reaction (PCR)-based markers. Often, SNP markers are converted to cleaved amplified polymorphic markers (CAPS) by including a restriction enzyme digestion step specific for the SNP base. Alternatively, tetra markers can be developed that use two allele-specific primers in addition to locus-specific PCR primers. Efficient conversion of SNP markers to PCR markers depends on the sequence context; not all SNPs can be successfully transformed into CAPS or tetra markers. To overcome this limitation, we have adopted a CEL-I nuclease-based SNP genotyping method, which detects SNPs independent of the sequence context at low cost. CEL-I nuclease can be easily purified from fresh celery. It cuts the DNA double strand at single-stranded mismatch sites and is used in mutation detection. We have shown that this nuclease can also be applied for SNP detection. The method has been successfully tested for genotyping mungbean breeding lines for presence and absence of MYMV and bruchid resistance alleles. The technique requires only very simple equipment and therefore is well suited for marker-assisted selection in breeding programs that lack access to sophisticated laboratories.

Keywords: Marker-assistant selection, mungbean, SNP genotyping

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