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Identification of SNPs associated with *loci* for disease resistance response in the soybean genome

Pinheiro, M[1,2]; Barros, BA[1]; Pereira, RM[1,3]; Piovezani, AR[1,4], Camargo, PO[1]; Brito Júnior, SL[1,5] e Abdelnoor, RV[1]

[1]Empresa Brasileira de Pesquisa Agropecuária /Embrapa Soja, Londrina, PR, 86001-970.
[2]Centro de Ciências Biológicas, Univ. Est. do Norte do Paraná, PR, 86360-000.
[3]Fac. Ciências Biológicas e Ambientais, Univ. Fed. da Grande Dourados, MS, 79804-970.
[4]Ins. de Matemática e Estatística/Interunidades de Bioinformática, Univ. de São Paulo, SP, 05508-090.
[5]Dep. de Biologia Geral, Univ. Est. de Londrina, PR, 86051-990.

E-mail: maisa@cnpso.embrapa.br

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Soybean is an important crop worldwide in terms of economic and nutritional value. However, soybean production is threatened by pathogen attacks. The genetic basis of plant disease resistance has been subject to intense research. In recent years, SNP markers have gained much interest in the scientific and breeding community. However, to date few examples exist of SNP haplotypes being exploited to tag disease-resistance *loci* in plants. The main goal of this study was to identify SNPs markers associated with genes related to defense response in the soybean genome. Eight soybean genotypes, including disease resistant sources, were chosen for SNP discovery: Conquista, PI459025, PI230970, PI561356, Pintado, PI595099, BRS282, and BRS 231. Furthermore, related sequences from the Williams 82 cultivar were downloaded from soybean genome reference and included into analysis as a standard genotype. A total of 2,723 sequences were selected from GenBank by a keyword search focused on the following words: disease resistance gene, disease resistance plant, resistance gene analogs), coiled-coil (CC), leucine-rich repeat (LRR), leucine zipper (LZ), nucleotide binding site (NBS). All sequences were blasted against soybean genome and those which showed a single hit were used to design 38 primer sets to produce amplicons of approximately 250-600 bp in length. Genomic DNA was isolated from fresh leaf tissue of eight soybean genotypes using modified CTAB method. PCR fragments of interest were excised and extracted from the agarose gel and cloned into a pGEM-TEasy vector for sequencing. To detect SNPs among eight soybean genotypes, ABI trace files were aligned, and the mutations were identified using Sequencher 4.1.4 software. Out of 38 primer sets designed, 31 (81,58%) amplified an expected PCR product and the analysis of 16 primer sets were completed. High quality sequence data for all soybean genome were obtained. Of the 47 single-based changes identified, 51% of the SNPs were of the C↔T (G↔A) variety (i.e., transitions), whereas transversions accounted for 34%. The remaining variation (15%) is represented by Indels. In the future, the association between putative SNPs and disease resistance response should be validated using mapping populations.