

DEVELOPMENT AND CHARACTERIZATION OF SIMPLE SEQUENCE REPEAT MARKERS FOR *Phaseolus vulgaris*. ¹Buso GSC, ¹Amaral ZP, ¹Brondani RV, ¹Moretzsohn MC, ²Brondani C, ³Ferreira ME. ¹Laboratório de Genética de Plantas - EMBRAPA-Recursos Genéticos e Biotecnologia, ²Embrapa - Arroz e Feijão, ³Embrapa - Recursos Genéticos e Biotecnologia e Universidade Católica de Brasília. buso@cenargen.embrapa.br

Beans are important protein source for South American and African populations. However, the bean production has stunted in the last thirty years and one of the possible reasons for this is the lack of variability in the commercial cultivars. The EMBRAPA-Arroz e Feijão has maintained a collection of bean varieties and their wild relatives and there is the necessity to develop new methods for genetic studies of beans, aiming to have genetic diversity and population genetics estimates and genetic mapping. Microsatellite or simple sequence repeats (SSR) markers are efficient for detailed genetic analysis. They are PCR based markers, codominants and multiallelic. A battery of SSR markers has been developed as part of a bean genetic analysis and genetic breeding programs. The genomic DNA was digested with the restriction enzyme Tsp509 I. Fragments between 300 and 800 bp were recovered onto a nylon membrane. An enrichment procedure was used where the fragments that had SSRs were selected by hybridization to biotinylated oligonucleotides bound to magnetic beads. This fraction of selected fragments were used for development of an AG/TC enriched genomic library. This library was screened for clones that contained SSRs by colony hybridization with probes (AG)₁₃. The positive clones were additionally screened by anchor-PCR and further sequenced. The SSRs flanking sequences were used for primer design. From 286 SSR positive colonies screened by hybridisation, in 100 the presence of AG repetitions was confirmed by anchor-PCR. A sub-sample of 91 clones was sequenced and from those, 28 showed sufficient adjacent DNA sequence for SSR primer design. From those, 25 were designed and the conditions for amplification of these primers were tested in a representative (84 accessions) sample of the bean germplasm collection. Six of these primers did not amplify any fragment and 8 were monomorphic for the sample analysed. The other 11 loci amplified 3 to 10 alleles per locus. Gene diversity varied from 0.21 to 0.83. This is a good indicative of the high level of SSR loci polymorphism in an autogamous species such as *Phaseolus vulgaris* and of the SSR primers potential for genetic studies of wild and cultivated *Phaseolus*. Órgão Financiador : EMBRAPA