RESEARCH PAPER

Asian Journal of Bio Science, Vol. 4 Issue 2 : 249-253 (October, 2009 to March, 2010)

Molecular characterization of elite cotton cultivars using ISSR markers

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(Accepted : August, 2009)

Genetic variability and relationship between varieties are of great importance for cotton breeding. ISSR marker systems were used for identification and genetic diversity analysis of elite *G. hirsutum*, *G. arboreum* and introgressed lines. 12 cotton genotypes were subjected to ISSR analysis using 55 ISSR primers. PCR products were subjected to agarose gel electrophoresis and the banding patterns were compared among 12 elite cotton varieties of diploid, tetraploid and introgressed cotton. Out of 55 ISSR primers tested, 15 were scorable, producing 101 marker bands with 83 being polymorphic. The primer IS-08 generated the greatest number of polymorphic markers. The ISSR markers were found to be reproducible and polymorphic. A dendrogram constructed from ISSR data classified 12 cotton genotypes into two major clusters, one containing six genotypes belonging to *G. hirsutum* cultivars and the other contained 4 genotypes belonging to *G. arboreum* cultivars. Two introgressed cultivars PAIG-8/1 and PAIG-27 showed highest level of genetic similarity with G. arboretum varieties. ISSR technique was thus found to be efficient method for detecting DNA polymorphism useful for DNA fingerprinting and genetic diversity analysis in cotton.

Key words : Genetic diversity, Molecular markers, ISSR, Cotto

INTRODUCTION

Cotton 'the white gold' is the world's leading natural fiber crop and it is the corner stone of textile industries world wide. The cultivated cotton include *Gossypium arboreum* (L) and *Gossypium herbacium* (L), Old World species, both diploid species with an AA genome native to southern Asia, Africa and two allotetraploid species *Gossypium barbadense* (L) and *Gossypium hirsutum* (L),New World species with AD genome from Central, North and South America. Although small gains in yield and fiber quality continue to be made by conventional breeding programs, genetic improvement of agronomic traits is beginning to plateau as a result of an increasing narrow germplasm base for selection. Genetic diversity is desirable for long term crop improvement and reduction of vulnerability to important crop pests. Genetic diversity resulting from interspecific introgression can be evaluated with morphological characteristics, seed proteins, isozymes and DNA markers. To have reliable estimates of genetic relationship, a large number of polymorphic markers are required. This limits the use of morphological characteristics and isozymes, which are few, or lack adequate levels of polymorphism in *Gossypium* spp. Therefore, there is a need to study polymorphism at the DNA level which can be indicative of genetic diversity in cotton. DNA markers have proven to be valuable in crop breeding especially in studies of genetic diversity and in cultivar identification.

Polymerase chain reaction (PCR) based molecular markers, e.g. ISSR, RAPD, SSR, STS, AFLP etc. are useful for various applications in the plant breeding. Inter Simple Sequence Repeats (ISSR) are arbitrary markers in which only one primer is used. The ISSR technique involves amplification of a DNA segment present at an amplifiable distance between two identical microsatellite repeat regions oriented in opposite strands. This technique uses microsatellites, usually 16 to 25 bp long, as primers in the single primer PCR reaction targeting multiple genomic loci to amplify mainly the inter SSR sequences of different size (Reddy *et al.*, 2002). The primers used can be repeated of di, tri, tetra or penta nucleotides anchored with one or two base sequences at 3'or 5' end (Zietkiewich *et al.*, 1994). Unanchored primers also can be used (Gupta *et al.*, 1994). ISSR are reproducible markers with 92-95 per cent efficiency (Reddy *et al.*, 2002). The present molecular analysis was carried out to analyze genetic relationship and genetic diversity of the cultivars.

MATERIALS AND METHODS

Plant material and DNA extraction:

The list of elite cotton cultivars used in the present study is as below.

Elite G. hirsutum cultivars.

1. PH-93 2. PH-325 3. PH-348 4. NH-452 5. NH-