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DEVELOPMENT OF HIGH THROUGHPUT DNA MARKER SYSTEMS IN COTTON BASED ON GENE AND REGULATORY SEQUENCES

Yingzhi Lu, Mingxiong Pang, Sarah Higbie, Doug Hinchliffe and Chen Niu
Youlu Yuan and Shuxun Yu
Cotton Research Institute
Chinese Academy of Agricultural Sciences
Anyang, Henan, China
Jinfa Zhang
Las Cruces, NM

Abstract

Molecular markers such as RFLP, RAPD, AFLP, SSR, and SNP have been developed and used in numerous plants including cotton (*Gossypium hirsutum* L.) for fingerprinting, linkage map construction, gene/QTL mapping, and genetic diversity studies. However, many of the marker systems have some limitations in terms of polymorphism levels, high throughput-ness, and anonymousness. Markers such as cDNA-RFLP, EST-SSR, and SNPs are gene sequence specific, but the level of DNA polymorphism within certain plant species including cotton is low, limiting their utilization in the development of genome-wide linkage maps and gene discovery. Our current research focuses on the development of new DNA marker systems using array-based hybridization and PCR-based techniques. High throughput marker systems were developed based on conserved DNA sequences for promoters, cis-acting elements, disease resistant gene analogous, microRNA genes, untranslated regions, and introns. To profile start codon regions, an ATG-anchored AFLP analysis was devised and proved to be reliable. Oligonucleotide arrays, initially designed for expression analysis, were also successfully used to detect allelic variation via direct hybridization of labeled genomic DNA. Single base changes (SNP) or insertion/deletion (Indel) within oligonucleotide region in an array produce differential hybridization signals, called single-feature polymorphism (SFP). The array-based marker technology is very powerful in that numerous markers can be assayed at once that can provide genome-wide coverage. Compared with the traditional molecular markers, the modified and new marker systems are highly cost effective and gene-regions or regulatory regions targeted. They can better resolve genetic relationships between genotypes, rendering a higher discriminatory power.