

Molecular Diversity and Polymorphism Information Content of Selected *Gossypium hirsutum* Accessions

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RESEARCH PROBLEM

The narrow genetic base of cultivated cotton germplasm is hindering the cotton productivity worldwide. The objective of the present study is to evaluate the genetic diversity within *Gossypium hirsutum* accessions using simple sequence repeat (SSR) markers.

BACKGROUND INFORMATION

Effective use of *Gossypium hirsutum* L. lines in cotton genetic improvement programs depends on the extent of genetic variation for desirable alleles and the accurate characterization of the variability among germplasm accessions. Marker assisted selection has provided the potential for efficient development of disease and pest resistant plants.

Association mapping is used to identify chromosomal regions containing disease-susceptibility loci or loci involved in other phenotypic traits of interest like fiber quality. It has been advocated as the method of choice for mapping complex-trait loci. Such studies are very limited in cotton and therefore are important for cotton breeding. *Gossypium hirsutum* accessions with resistance to reniform nematodes from the USDA collection were evaluated and genotyped with SSR markers.

RESEARCH DESCRIPTION

Both genetic diversity and association mapping is based on the strength of association between the genetic marker and phenotype. For the current study, we have used 96 accessions, screened for partial reniform nematode resistance using chromosome specific primers sets. These accessions are from the USDA collection.

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Genomic DNA was obtained from the greenhouse grown plants using DNeasy plant mini kits. Polymerase chain reaction (PCR) was performed and polymorphisms at each locus were assessed by electrophoresis of the PCR products in a vertical gel system on a polyacrylamide gel. The profiles produced by SSR markers were scored manually: each allele was scored as present (1) or absent (0) for each SSR locus (Table 1).

Genetic diversity was calculated at each locus for allelic Polymorphism Information Content (PIC), with program CERVUS version 2.0 based on allelic frequencies among all 96 genotypes analyzed. The PIC values for each SSR were estimated by determining the frequency of alleles per locus using the following formula:

$$PIC = 1 - \sum x_i^2$$

where x_i is the relative frequency of the i th allele of the SSR loci. Markers were classified as informative when PIC was ≥ 0.5 .

For association analysis, the Excel spreadsheet was run through softwares STRUCTURE and TASSEL. The program STRUCTURE implements a model-based clustering method for inferring population structure using genotypic data consisting of unlinked markers. TASSEL (Trait Analysis by aSSociation, Evolution and Linkage) uses most advanced statistical methods to maximize statistical power for finding a Quantitative Trait Locus (QTL).

RESULTS AND DISCUSSION

For the genetic diversity, the primer sets yielded 177 alleles of which 173 were polymorphic and were amplified by 48 SSR primers. The mean number of alleles per locus was 3.40 (StDev 0.995), but the number varied from 1 to 4. The PIC values ranged from 0.00 to 0.95 and the relation with the number of alleles is shown in Fig. 1. Seventy-two percent of markers used had PIC values of 0.50 or greater. In our study, the majority (80%) of the informative SSRs contained at least 10 repeats. Although contradictory references also exist (e.g., Struss and Plieske 1998), a similar positive relationship between the number of tandem repeats and the level of polymorphism also was observed in tomato (Smulders et al., 1997) and maize (Vigouroux et al., 2002).

CONCLUSION

Analysis of genetic distance and population structure provided evidence of no significant population structure in the *G. hirsutum* accessions. The results provide preliminary insight into the SSR informativeness of the cotton genome and are very useful as a framework for future studies in cotton that will accelerate

development of superior cotton cultivars. These tests between SSR markers using their PIC values suggests that the majority of the informative SSRs were present between these accessions as their PIC values were 0.5.

These results provide preliminary insight into the cotton genome and are very useful as a framework for future 'association studies' in cotton that will accelerate development of superior cotton cultivars through the AMAS program. These tests between 52 markers using a general linear methodology suggest that a significant association between these accessions does not exist. A more detailed study of the population structure must be done in order to find more associations among the accessions.

LITERATURE CITED

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Table 1. Scoring for the presence of nematodes.

Accessions	Primer 0827	Primer 0834	Primer 0946	Primer 1047	Primer 1064
TX1	1	1	1	0	1
TX5	1	1	1	1	1
TX9	0	1	1	1	1
TX10	0	1	1	1	1
TX11	0	1	1	1	1
TX16	1	1	1	1	1
TX17	1	1	1	1	1
TX18	0	1	1	1	1
TX19	1	0	1	0	1