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IMPROVEMENT OF 'DES 56' THROUGH BACKCROSS INTROGRESSION FROM A WILD DAY-NEUTRAL FLOWERING CONVERTED ACCESSION

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Abstract

Plant breeders rely on genetic variation. It is a necessary tool that allows them to continually improve crop cultivars. Currently, there is little diversity among cultivated cottons. Similar pedigrees have lead to stagnation in yield increases and improved fiber traits. One possible solution to this problem is the introduction of new genes from wild accessions from Mexico that have been converted to flower in the United States. In order for U.S. cotton breeders to efficiently utilize these accessions the wild plants should be categorized for their novel genes through backcross introgression into elite lines and genetic marker analysis. This would categorize the beneficial traits that are easily transferred to elite lines, making breeders more willing to start introducing new genetics into the U.S. cotton gene pool. In this research project simple sequence repeat (SSR) markers were used to categorize the introgression of traits from a converted MDN 63 into the elite DES 56.

Introduction

Genetic diversity is an essential tool for plant breeders. The lack of dramatic yield increases among cotton cultivars in recent years may indicate a need for increased diversity among cotton cultivars. There is a lack of exotic germplasm and overall similarity of modern cotton cultivar pedigrees that calls attention to itself (Liu et al., 2000). There is a need for the exploration of exotic lines that may possess valuable traits that could be introgressed into cultivated lines.

Converted day-neutral Gossypium hirsutum L. race stocks exist that may hold the genes necessary for the further improvement of today's cotton cultivars. Though these lines are available for direct use by breeding programs (McCarty, et al., 1996), further characterization would make them more useful for breeders. It must be determined if genetic variations exist between the converted stocks and modern cultivars and if those variations can be incorporated into the modern lines with positive results in yield or fiber properties. Molecular markers can be used to identify polymorphism between converted stocks and modern cultivars and further to follow the introgression of desired sequences into the modern lines.

Simple Sequence Repeat (SSR) markers were used here to identify polymorphism between DES 56, elite genotype, and MDN 63, wild, converted genotype, and a BC_1F_1 population resulting from their combination.

Materials and Methods

SSR Marker Analysis

Twenty-one SSR primer pairs were selected to analyze DES 56, MDN 63, and the BC_1F_1 progeny population (Table 2). The progeny population consisted of 44 plants that were transplanted to the greenhouse after the 2001 season. Both parents and the progeny were sampled for DNA extraction. The DNA was resuspended in TE buffer and quantified fluorometrically. The DNA was diluted and stored at 4˚C. The amplification profile was that used by Liu (2000):

- initial denaturation at 94˚C for 12 min.
- 40 cycles: 93˚C for 15
  55˚C for 30 s
  72˚C for 1 min
- 1 min. at 72˚C followed the 40th cycle

All primers were tested on the parental DNA to determine if polymorphism existed. Primers that appeared to show polymorphism were then used to analyze the backcross progeny population.

BC_2F_1 Yield Comparison

Plot size for this part of the study was single rows, 15 feet long at 36 in. row spacing. Seed was planted from the BC_1F_1 plants that were transplanted to the greenhouse after the 2001 season. Production practices as those recommended by the University of Georgia Cooperative Extension were followed. The BC_1F_1 rows were compared to the parental rows and analyzed using ANOVA. The top ten yielding progeny advance to the BC_2F_2 in 2003 (Table 1).

Results and Discussion

SSR Marker Analysis

- Primers 1, 5, 9, 10, 12-15, and 18-21 appeared to be polymorphic for DES 56 and MDN 63 (Table 2) (Figure 1).
- Primer 9 was used to analyze the backcross progeny for polymorphism with the parental lines (Figure 2).
- 53% of the progeny population was similar to DES 56 (one band)
- 47% of the progeny population was heterozygous (two bands)
- This experiment was highly exploratory in nature. Further study must be performed to evaluate the other primers and the backcross progeny.
- Further exploration of these SSR primers and the backcross populations' yield data and fiber traits may lead to correlations that will allow breeders to more easily select plants that contain exotic alleles and desirable yield and fiber characteristics.

BC_2F_1 Yield Comparison

ANOVA results indicated that there was not a significant difference in yield between any of the BC_2F_1 progeny and DES 56 and MDN 63. It will be important to evaluate the backcross progeny at various locations to more accurately categorize the introgression of traits from the wild accessions. The lint fraction and fiber trait data from this experiment is yet to be analyzed. There was a wide segregation among the BC_1F_1 population for lint fraction and fiber trait data in 2001, implying that there was some genetic variation imparted by the exotic donor parent (May, 2001).

References

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Table 1. Top yielding BC,F, progeny row seedcotton yields.

ENTRY	REP 1	REP 2	MEAN	RANK	SSR
	SEEDCOTTON, LBS/PLOT				
1	4.2	3.8	4.0	1	H
9	3.7	4.2	4.0	1	DES56
46	3.9	3.7	3.8	2	H
48	3.1	4.2	3.7	3	DES56
30	3.6	3.6	3.6	4	DES56
43	3.8	3.4	3.6	4	H
16	4.0	2.9	3.5	5	unknown
35	3.9	3.0	3.5	5	H
42	2.9	4.0	3.5	5	DES56
20	4.0	2.8	3.4	6	DES56
MDN63	3.5	1.7	2.6	13	MDN63
DES56	1.2	1.7	1.5	20	DES56
	LSD0.10		NS		

Table 2. Primers used for amplification of simple sequence repeat (SSR) loci in DES 56, MDN 63, and BC_1F_1 population.

SSR	Flanking Primer Sequence (5'-3')	Allele Size Range	Chromosome Location
BNL3563	AAGCATAAACTTGACACAAGCCAATGGGCAAGAAAAGGGAAC	228-276	10*Lo*
BNL1053	AGGGTCTGTCATGGTTGGAGCATGCATGCGTACGTGTGTA	176-194	3
BNL1064	TTTGCGGGTAATCCTATTGCTGTCTATGGGACATTTCGCA	140-160	6*sh*
BNL3065	CAAACGGGAGACCAAAAAAACGAACTGGCGAGTTAGTGCT	178-214	16
BNL2960	TAAGCTCTGGAGGCCAAAAACCATTTCAATTTCAAGCATACG	150-224	10*Lo*
BNL1679	AATTGAGTGATGCTAGCATTTCAGCAAAGGGATTTGCTGGCAGTA	164-190	12
BNL169	TCACAAATAAAAGTGAAATTGCGGGCTGGTGACCATAAAAGGA	196-214	20*sh*
BNL3482	ATTTGCCCCAGGTTTTTTTTGCAACACCTTTTCCTCCCTA	138-146	26*Lo*
BNL3255	GACAGTCAAACAGAACAGATATGCTTACACGACTTGTTCCCACG	210-232	5*sh*
CML60	GAAGATTCCATCTGCAGACCCAGCCAACAAAACCATAAACATGAACTC	113-125	7*Lo*
BNL3441	CGTCATAAACCGTGCTTGTGGGCCACTTTAAGGCTGTCAC	201-217	3*sh*
BNL256	TTTTGCTCCATTTTTTTGCCTTTATTAATTTCGTTTAGCTTCCG	188-218	10*Lo*
BNL3792	TTCGAGATCCCCTGTTCTGACATATTCCAGTCAAACCAAACG	216-236	20*Lo*
BNL3895	CGCTCTTGGTCATGGATTTTGCCAAGCTCACTGGAAGAAC	176-200	10*Lo*
BNL3955	AGAGATGCAATGGGATCGACATGTGATAATGCGGGGAATG	161-207	17*Lo*
BNL3103	ACTTTGAGATATTGTTATTCTACCCGTCGAACAATTACGAATCAAATG	188-220	25*sh*
BNL3995	ATATTTTATTCTTTTAATAGCTTTATTCCCTTGGAAAAACCCATGGTGAT	182-198	5*sh*
BNL1440(2)	CCGAAATATACTTGTCATCTAAACGCCCCCGGACTAATTTTTCAA	240-244	25*Lo*
		258-270	6*sh*
BNL2590	GAAAAACCAAAAAGGAAAATCGCTCCCTCTCTCTAACCGGCT	186-206	9*Lo*
BNL2544	GCCGAAACTAAAACGTCCAATCCTTACTCACTAAGCAGCCG	206-222	18*Sh*
BNL3479	AGTGGGTTGGACTTTCATGCCACGGGCTTTTTTTTTTTCA	148-256	18*Lo*

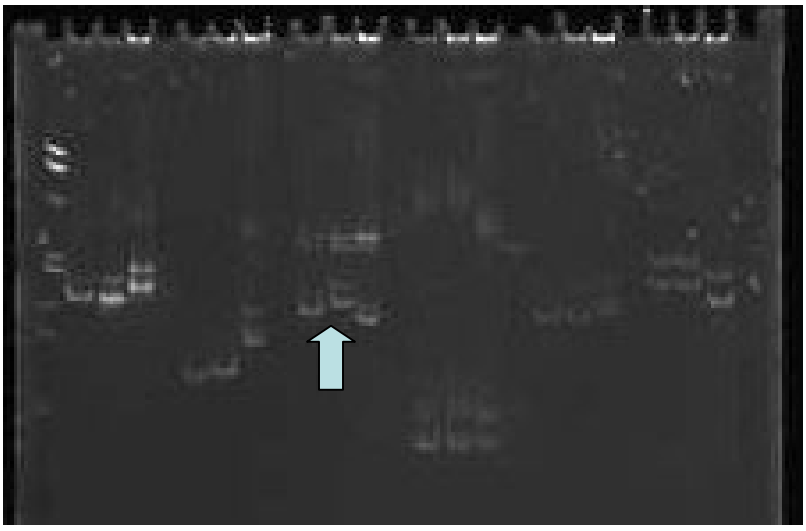

Figure 1. 12% polyacrylamide gel showing polymorphism between DES 56 and MDN 63. The arrow indicates primer 9.

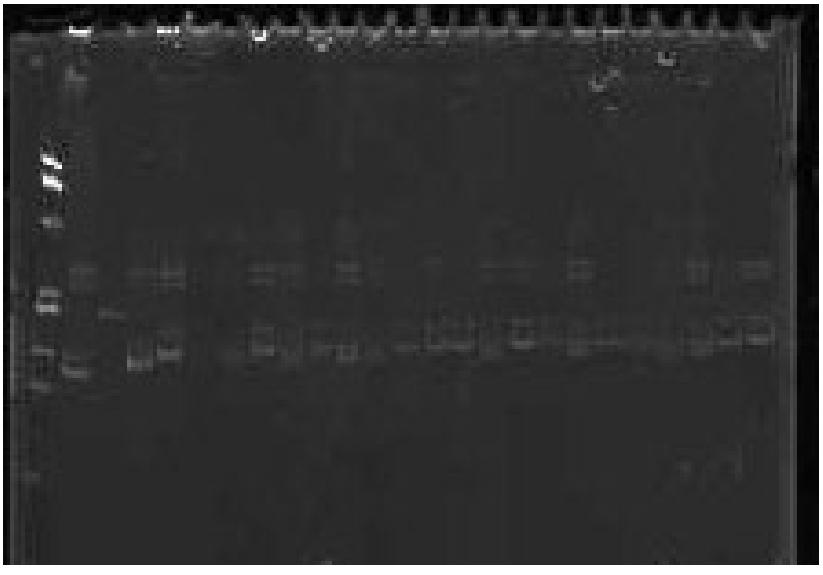

Figure 2. PCR products from primer 9 amplified with the backcross progeny population DNA.