

TWENTY-FIVE YEARS OF INTROGRESSION BREEDING THROUGH INTERSPECIFIC HYBRIDIZATION BETWEEN *GOSSYPIMUM HIRSUTUM* AND *GOSSYPIMUM BARBADENSE*

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Abstract

Transferring desirable traits from Pima to Upland cotton has been a long lasting goal for many cotton geneticists and breeders since the discovery of the Mendelian laws in the turn of the 20th century. However, success in developing commercial cotton cultivars from the interspecific breeding is limited. This presentation will summarize my 25 years of experience in interspecific breeding between Upland and Pima cotton. In the mid-1980's, I employed interspecific crossing between Upland and Pima to introduce desirable genes for resistance to spider mites and Verticillium wilt from Pima to Upland cotton. Then, breeding objectives were expanded to include fiber quality, sub-okra leaf type and heterosis. This resulted in the development of high-yielding breeding lines with sub-okra leaf or cleistogamous flowers, and lines with high fiber quality. Since the early 2000's, molecular markers and gene expression profiling have been used to identify quantitative trait loci or differential expressed genes in Pima cotton or introgressed population. In the meanwhile, new introgressed lines with higher yield potential and/or better fiber quality than Upland cotton parents have been developed, indicating a simultaneous introduction of desirable genes for yield and fiber quality into Upland cotton. Tolerance to drought and salt stresses, and Verticillium wilt has also been evaluated in these introgressed lines. The presentation presented examples on single gene trait transfer, discovery of new traits due to transgressive segregations, and breeding for single multi-gene traits and multiple multi-gene traits including lint yield and fiber quality.

Introduction

Increasing lint yield and enhancing fiber quality has been a most important breeding goal for cotton improvement. Upland cotton, *Gossypium hirsutum*, is known for its high yield potential and wide adaptations, while long extra staple cotton, known as Pima cotton (*G. barbadense*) in the U.S., has superior fiber quality, but only grown in semi-arid areas in the world. Even though both are cultivated tetraploid species originated from the same ancestor 1-2 million years ago, tremendous genetic variation has accumulated during evolution and artificial selection since domestication. This has resulted in reproductive barrier, indicated by hybrid breakdown in F2 and advanced interspecific generations. Transferring desirable traits from Pima to Upland cotton has been a long lasting goal for many cotton geneticists and breeders since the discovery of the Mendelian laws in the turn of the 20th century. However, success in developing commercial cotton cultivars from the interspecific breeding is limited.

In 1986, I started interspecific breeding between Upland and Pima in China to transfer spider mite resistance, Verticillium wilt tolerance and sub-okra leaf type from Pima into Upland cotton. I made many Upland x Pima hybrids to study heterosis, combining abilities and quantitative genetics in the interspecific populations. After I saw numerous unexpected segregations including new traits such as cleistogamous flowers, I started to make selections for these traits and for yield and fiber quality. After 10 years of breeding between Upland and Pima, I came to the United States in 1995 and it has been a rewarding experience. One of the major research areas has been on Upland x Pima. We have had more technologies at our disposal including DNA markers, gene mapping, functional genomics and molecular breeding. It has been another 15 years. We can overcome the geographic distance and physical isolation, as it has been the case in the human history. Can we overcome the genetic distance and reproductive isolation between plant species, i.e., the two tetraploid cotton species?

The objective of the presentation was to provide some examples on single gene trait transfer, discovery of new traits due to transgressive segregations, and breeding for single multi-gene traits and multiple multi-gene traits.

Genetic Differences between Upland and Pima Cotton

The two cultivated species have numerous contrasting traits including morphological traits such as leaf shape, flower and pollen color, petal spot and seed fuzz, disease and insect resistance such as Verticillium wilt and spider mites, and agronomic traits such as lint yield, fiber length, strength and fineness (Table 1). However, when compared at the DNA level and gene expression level, the differences between Upland and Pima cotton are even

higher. For example, when compared Pima Giza 75 (which is an Egyptian cotton) with Upland SG 747 which is a mid-South Upland cotton cultivar, almost 6,000 of 24,000 genes were differentially expressed in developing fibers 10 days after flowering, which accounted for 24% of the total genes on the Affymetrix GeneChip. The question is: are some of these genes related to cotton yield and fiber quality? Can we transfer them from Pima to Upland to make Upland even higher yield with better fiber quality?

Table 1. Phenotypic comparisons between Upland and Pima cotton

Trait	Upland	Pima	Type of inheritance
Leaf shape	normal	sub-okra	Qualitative
Flower color	cream	yellow	Qualitative
Pollen color	cream	yellow	Qualitative
Petal spot	no	yes	Qualitative
Seed fuzz	yes	no	Qualitative
Verticillium wilt	susceptible	tolerant	Both
Spider mite	susceptible	tolerant	Both
Plant height	short	tall	Quantitative
Lint yield	high	low	Quantitative
Boll size	large	small	Quantitative
Lint percentage	high	low	Quantitative
Fiber length	shorter	long	Quantitative
Fiber strength	weak	strong	Quantitative
Fiber fineness	coarse	fine	Quantitative

Transfer of Single Gene Traits

I began Upland x Pima breeding by transferring the Pima sub-okra leaf type to Upland through backcrossing. One line, 73001 turned out to perform very well, in comparison to the recurrent parent and also a commercial cultivar at that time. It had more than 10% higher lint yield and 5% higher lint percentage and similar fiber quality (Table 2).

Table 2. Yield potential of an Upland cotton line with sub-okra leaf type, 3 locations, 1991-1992

Line	Lint yield	Lint	Length	Micronaire	Strength
	LB/A	%	inch	unit	g/tex
73001 (sub-okra)	1127.9	43.1	1.11	4.7	22.7
E-jian 1 (normal)	1029.9	40.9	1.11	4.6	22.6
LSD	*	*	ns	ns	ns

*- significant at the P=0.05 level; ns- not significant.

There were some interesting new traits segregated out from the interspecific hybrid populations. One of them is cleistogamous flowers, i.e., closed flowers (Fig. 1). When cotton flowers are not open when blooming, it not only prevents out-crossing due to insect pollinators, it also prevents rains from getting inside the flowers. So cotton plants can still set bolls when it rains during flowering season, which is very common in many areas. One line did very well. It had 20-40% higher yield than its recurrent parent and also a commercial cultivar, with extremely higher boll load and high lint percentage. But seed size and boll size were smaller. Thus, the above results demonstrate that it is possible to increase Upland cotton yield by introducing one gene from Pima cotton, through the change of a morphological trait.



Fig. 1. An Upland cotton line with cleistogamous flowers selected from Upland x Pima cotton.

Many other new traits can also be isolated in Upland x Pima hybrid populations, such as natural defoliation type (Fig. 2) and stay-green type (Fig. 3). These two traits may have some use in cotton production.



Fig. 2. A natural defoliation Upland cotton selected from Upland x Pima cotton.



Fig. 3. A stay-green Upland cotton type selected from Upland x Pima cotton.

Transfer of Single Multi-Gene Trait

Can we improve quantitative traits such as yield and fiber quality by transferring multiple genes from Pima to Upland cotton? Our results showed that in a cross between Acala and Pima and through pedigree selections, Acala cotton yield can be significantly increased, by almost 30% based on replicated advanced yield tests in 2009 and 2010 (Table 3). Acala cotton fiber quality can be also maintained.

Table 3. High-yielding lines selected in Acala x Pima from pedigree selection, Las Cruces, NM, 2009-2010

Line	Lint yield	Lint yield	Lint	Length	Micronaire	Uniformity	Strength
	LB/A	CK %	%	inch	unit	%	g/tex
Year 2009							
08N1562	1148.3	139.8	39.7	1.24	4.10	83.13	35.27
08N1564	1214.7	147.9	38.4	1.20	4.35	83.97	33.43
1517-99 (P1, CK)	821.6	100.0	37.5	1.22	4.42	84.20	34.30
Pima (P2)	975.3	118.7	38.7	1.36	4.37	86.23	42.13
Year 2010							
08N1562	1467.3	128.8	44.3	1.12	4.95	81.70	30.03
08N1564	1207.4	106.0	44.6	1.11	5.01	83.03	30.07
1517-08 (CK)	1138.9	100.0	41.3	1.13	4.90	82	32.53
Pima (P2)	na	na	na	na	na	na	na

P1- Parent 1; P2- Parent 2; CK- check; na- not available.

What about fiber quality? Our experience indicates that it is extremely difficult to transfer Pima fiber quality to Acala cotton without sacrificing cotton yield. Transferring single Pima fiber trait such as fiber length or micronaire is much easier. But for the transfer of Pima fiber strength, it is very difficult (Table 4).

Table 4. High fiber quality lines selected in Acala x Pima from pedigree selection, Las Cruces, NM, 2009-2010

Line	Lint	Length	Micronaire	Uniformity	Strength
	%	inch	unit	%	g/tex
08N1223-3	36.80	1.35	4.36	81.6	34.65
08N1110-2	25.35	1.35	3.90	85.1	38.80
08N1099-1	34.28	1.34	3.19	81.3	34.80
08N1120-1	35.69	1.34	3.96	83.9	39.65
08N1288-3	40.05	1.32	4.23	85.8	41.40
08N1868-2	38.61	1.30	4.09	86.1	42.55
08N1066-1	30.86	1.24	4.07	81.8	38.80
08N1046-1	23.71	1.37	4.82	86.9	43.20
Pima S-7 (P2)	37-40	1.32-1.44	3.8-4.6	84-87	40-45

P2- Parent 2.

Transfer of Multiple Single Multi-Gene Traits

Simultaneously transferring the three Pima fiber quality traits (length, strength and fineness) to Acala while maintaining Acala cotton yield is extremely difficult if not impossible. For example, one breeding line, 08N1046-1, reached the Pima fiber quality, but its lint percentage and yield were very low (Table 4).

Breeding between short or medium staple Upland and Pima would be more difficult because they are more different than the differences between Acala and Pima cotton. We resorted to use backcrossing instead of pedigree selections. We have developed and tested many backcross inbred lines (BIL) in Las Cruces, New Mexico and Maricopa, Arizona when Dr. Richard Percy was with USDA-ARS there. As shown in Table 5, some of the BIL lines did very well in the two locations in three years. Some had even higher yield than SG 747, the recurrent parent, but also with improved fiber quality in fiber length, strength and micronaire than the recurrent Upland parent. The results have demonstrated that Upland cotton yield and fiber quality can be improved at the same time by interspecific breeding between Upland and Pima.

Table 5. Field performance of selected backcross inbred lines, AZ and NM, 2005-2007

Line	Lint yield	Length	Strength	Micronaire
	%	inch	g/tex	unit
NMHT-73	100.10	1.17	30.23	5.30
NMHT-103	100.34	1.14	29.40	5.01
NMHT-132	100.60	1.18	30.82	5.23
NMHT-09	102.98	1.18	30.10	4.95
NMHT-87	106.27	1.20	29.98	4.87
NMHT-35	108.89	1.20	30.28	4.43
Pima (P2)	58.11	1.38	39.58	4.25
SG 747 (P1 & RP)	100	1.13	26.58	5.47

P1- Parent 1; RP- recurrent parent.

Summary

Based on my 25 years of experience in breeding between Upland and Pima, we can transfer single gene traits from Pima to Upland cotton to improve cotton yield. We also can select new traits which may have potentials in enhancing yield. A multi-gene trait such as fiber length or micronaire can also be successfully transferred from Pima to Upland cotton. But transferring Pima fiber strength is very difficult and transferring Pima fiber length, strength and fineness without yield penalty is almost impossible.

However, Upland cotton yield and fiber quality can be improved at the same time by breeding between Upland and Pima. The success in introgression breeding is a numbers game in that it depends on the population size, time one can invest and persistency, selection pressure and also how lucky one is.

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