

**GENETIC MANIPULATION OF COTTON
LEAF STOMATAL DENSITY TO ENHANCE
DROUGHT TOLERANCE**

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Introduction

Unique upland cotton germplasm is under development that shows out-standing tolerance to abiotic stresses. Several advanced generation pro-geny lines which resulted from interspecific crosses of selected individual plants of original upland (Deltapine 90) acala type cotton and extra long staple pima (Pima S-6) cultivars have exhibited transgressive traits of agronomic value. Foremost among the characteristics for which selection was undertaken were drought and heat stress tolerance. Initial early generation selections were based upon superior germination and stand establishment with little regard for lint yield or quality beyond reproduction of the line. Only the best half of the selections were advanced in each of the first two years of the tests. Promising crosses were made and their progeny were included in later evaluations, as reported in previous Beltwide Proceedings. (McDaniel and Dobrenz, 1994; McDaniel, 1997).

A three year parallel study with paired progeny tested at near optimal and sub-optimal irrigation levels was then conducted which revealed that some strains maintained relatively low season long leaf temperatures. Progeny of these strains were increased, bulked and yield tested at several Arizona locations. These selections, tested as Mac95 and Mac13 performed as well as extant commercial cultivars judged by seed cotton yield and fiber length and strength. A reselection of Mac33 and Mac37, stress tolerant high fiber quality sister lines of Mac13, proved to contain a proportion of individuals that exhibited excellent stress tolerance. When observed closely, a number of these progeny were found to have significantly fewer stomata on the upper leaf surface than the normal range for upland cottons. Data of leaf surface stomatal densities of several selected individual plants from this population, compared with control plants, are illustrated in Table One. A single relatively unproductive plant was identified which had no observable stomata on the upper leaf surface; unfortunately this plant was lost. Third generation selfed progeny of several of these low upper leaf stomata number individuals were field tested this summer with fiber yield and quality data presently being analyzed. A winter greenhouse seed increase of the best of these lines in under way, with full scale field comparisons under imposed water stress planned for the 2000 season.

Synopsis of Previous Research

The genetic alteration of the number and proportion of stomata between upper and lower leaf surfaces of the cotton plant offers a unique opportunity to enhance the ability of the plant to withstand drought stress, especially under conditions of low humidity combined with high temperatures and high irradiance. Several authors have alluded to the potential utility of stomatal characteristics for breeding (Jones, 1987; Lu et al., 1998; and Willmer and Fricker, 1996). Because of the ability of the plant to maintain a boundary layer of moisture beneath the leaf, except under very windy conditions, the leaf that has fewer upper surface stomata should be better able to control transpirational water loss under abiotic stress conditions, while still maintaining adequate levels of transpirational cooling and uptake of atmospheric CO₂ for effective rates of photosynthetic metabolism. (Foster and Smith, 1986; Meinzer and Grantz, 1991; Mott et al., 1982).

Field studies which I conducted under irrigated agricultural conditions on University of Arizona Experimental Stations in southern Arizona indicate that this is so. Cotton selections with a proportion of individuals which displayed altered stomatal densities withstood water and high temperature stresses quite well. When parallel evaluations were conducted on plant materials of identical descent under near optimal and sub-optimal drip irrigation, the progenitors of the low upper leaf surface stomatal number lines out-performed the control cultivars under the sub-optimal irrigations. They set fewer bolls under near-optimal irrigations, as they were still growing vegetatively and were late in the transition to a fruiting cycle.

Under furrow irrigation, where a substantial drought stress may occur in the days just before subsequent mid-summer peak bloom irrigations, the low stomatal density lines did not exhibit the signs of stress seen in adjacent rows of control plants. Although these progenitor lines were not selected for lint yield per se, all materials in my breeding program have been rigorously selected for good seedling vigor and for excellent fiber quality characteristics. Present efforts include the evaluation of comparative lint yields of individual low upper leaf surface stomata number selections currently being increased for field testing. Other structural and physiological leaf alterations which may accompany the manipulation of stomatal densities are also being investigated, as are environmental and temporal effects.

Summary

Several plant selections have been made based upon individual progenitor plants which exhibited dramatically reduced upper leaf surface stomatal densities and altered upper to lower leaf surface stomatal ratios. Those plants

appeared normal with regard to other morphological characteristics, and closely resembled their acala type ancestors, with a growth habit similar to Deltapine acala 90, cream pollen and white petals, and four or five locule bolls. Preliminary HVI fiber tests indicated that fiber length of bolls from these plants generally ranged from 28 to 37 mm, with fiber strengths of 28 to 38 grams per tex, micronaire between 3.8 and 5.0 and uniformity (UI) of 83 to 87. Only individual plant and small hand harvested experimental block lint yield data have been taken to date. A sizable increase of individuals verified as expressing the low stomatal number trait is underway in my winter greenhouse, with the anticipation of replicated yield tests under desert agricultural conditions this coming season to verify the level of abiotic stress tolerance which they exhibit.

Acknowledgments

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Table 1: Mean stomatal densities of upper and lower young leaf surfaces of individual cotton plants grown at the Marana Agricultural Center, 1997

Plant I.D.	PROGENY			UPLAND CONTROLS			
	Stomata/mm ²			Stomata/mm ²			
	Up. Surf.	Low. Surf.	Ratio L/U	Plant I.D.	Up. Surf.	Low. Surf.	Ratio L/U
M33-4	68	149	2.2	M10-1	159	313	2.0
M33-9	16	215	13.4	M10-3	177	369	2.1
M37-2	28	160	5.7	M11-1	149	333	2.2
M37-17	54	209	3.9	M11-2	177	289	1.6
MEAN	41.5	183.2	6.3	MEAN	165.5	326	2.0

Data represent the means of observations of three individual leaves from each plant. Leaves were harvested, transported to the laboratory on ice and stomatal impressions were made within as short a time as practical. An adjacent plot of Mac95, the original selection made from an interspecific cross, served as an upland type control. Lines 33 and 37 represent later generation progeny selections drawn from the same genetic background as Mac95.