Heat Tolerance of Cotton Genotypes Assessed by Electrolyte Leakage, Chlorophyll Fluorescence Analysis and Performance in the Field in Pakistan

J. Gorham[1], J. Bridges[1], and M.N.A. Malik[3]
[1]Centre for Arid Zone Studies, Univ. of Wales, Bangor, LL57 2UW, U.K.; [2]Central Cotton Research Institute, Multan, Pakistan.

ABSTRACT

Germination and emergence of 117 cotton accessions were tested in the laboratory at 43$_o$C and 30$_o$C. The absolute heat tolerance of detached leaves of the 117 accessions was tested by chlorophyll fluorescence analysis (CFA) and by electrolyte leakage tests. The heat tolerance of whole plants was examined in a large, modified growth chamber. The relationships between temperature, net photosynthesis, stomatal conductance, transpiration and transpirational leaf cooling were examined using an automated CIRAS infra-red gas analyzer to measure gas exchange parameters while applying defined profiles of increasing leaf temperatures. An early-sowing field experiment at Multan examined the responses of 16 cotton lines to early heat stress. CIM443, CIM 438, BH89, GH8, NIAB78 SLS1 and 1269/93 were more heat-tolerant than Acala SJ2 at the germination stage. The chlorophyll fluorescence assay (CFA) revealed that CIM448 and CIM435 performed better than CIM443, but SLS1 performed badly in CFAs. Of the wild species, G. sturtianum was least tolerant, while Thespesia populnea was the most heat-tolerant wild relative. The hexaploid G411 (G. hirsutum cv. NC8 x G. australe) was the most tolerant interspecific hybrid. Electrolyte leakage tests largely confirmed these results. CIM448 and NIAB 313 were less leaky than Acala SJ2, while DNH40 was susceptible to heat damage. Only the electrolyte leakage test gave data which correlated well with yields in the field experiment.

Introduction

The main cotton-growing belt in the Pakistan experiences extreme summer temperatures that may exceed 50$_o$ C. Figure 1 Shows weather data for Multan in 1995, when daily maximum temperatures in excess of 45$_o$ C were experienced on several occasions between May and July. Cotton in this area is mainly irrigated, so leaf temperatures are always several degrees below air temperatures. Nevertheless, such high temperatures affect yields, particularly of cotton sown in April (Taha *et al.*, 1981). Indeed, early sowing provides a useful screening technique for heat tolerant cotton in this area. These trials seek to establish the usefulness of physiological tests (chlorophyll fluorescence analysis and electrolyte leakage) for heat tolerance, and compare the data with those from early-sowing experiments in the field.

Material and Methods

Early sowing experiment. Sixteen cultivars were sown in April 1997 at the Central Cotton Research Institute (CCRI), Multan, Pakistan. This resulted in the early fruiting phases coinciding with the hottest period of the season with subsequent heat damage to the crop. A number of parameters were recorded, including anther dehiscence, node number, boll set etc.

Electrolyte leakage test. Samples of leaves were taken from both the early-sowing field experiments, and from the same cultivars growing in greenhouses at the University of Wales, Bangor. Leaf disks were cut and washed in deionized water before being placed in suitable containers with fresh deionized water. Some replicates were kept at room temperature, while others were subjected to 50° C in a water bath for 1 hour. Electrical conductivity of the bathing medium was measured, and the samples were then autoclaved to obtain total electrolyte leakage. Percent injury was calculated as 100 * (treated-control)/(total-control).

Chlorophyll fluorescence analysis. Detached leaves from plants growing in Bangor were subjected to chlorophyll fluorescence analysis as described by Sethar *et al.* (1995) and Akhtar *et al.* (1996). For in vivo measurements, dark-adaptation clips were used with an OS100 modulated fluorometer.

Photosynthetic gas exchange. Plants were grown in a greenhouse in Bangor until required. Measurements were made in a Fisons 2340 growth cabinet converted to act as a ‘glove box’, with most of the equipment outside the chamber. Gas exchange was measured with an automated broad leaf chamber and a CIRAS-1 infrared gas analyzer, with only the leaf end of the chamber inserted into the growth cabinet. Leaf temperatures were controlled by either the cabinet, or by the automated leaf chamber.

Results

Germination and emergence of 117 cotton accessions were tested in the laboratory at 43ºC and 30ºC. CIM-443, CIM-438, BH-89, GH-8, NIAB-78, SLS-1 and

Acala 1269/93 were more heat-tolerant than the check variety, Acala SJ2, at the germination stage. Ashraf *et al.* (1994) also reported that NIAB-78 was heat-tolerant.

Comparing electrolyte leakage performed at Multan (Figure 2) with CFA of plants grown in Bangor (Figure 3) it can be seen that the former was more closely related to seed cotton yields in the field in an early-planting experiment. DNH-40 was the most heat-sensitive cultivar, with CIM-443 the most tolerant in terms of seed cotton yield.

CFA, which gives an indication of cellular heat tolerance, suggested that CIM448 and CIM435 were more tolerant in vitro than CIM443, but SLS1 performed badly in CFAs in Bangor. Of the wild species, *Gossypium sturtianum* was least tolerant (Figure 4), while *Thespesia populnea* was the most heat-tolerant wild relative (data not shown). The hexaploid G411 (*G. hirsutum* cv. NC8 x G. *australe*) was the most tolerant interspecific hybrid.

CFA and the electolyte leakage test measure different physiological processes, and it is therefore not surprising that the two tests do not always give the same rank order for different varieties. Figure 5 shows that there is greater variation for electrolyte leakage than for chlorophyll fluorescence, but also that the most ‘leaky’ varieties (here CIM-443 and SLS-171) have the lowest Fv/Fm ratios after heat treatment.

Several electrolyte leakage tests in Bangor confirmed the heat tolerance of *Thespesia populnea* and the sensitivity of *Gossypium sturtianum*. In electrolyte leakage tests on plants grown in Bangor, Carolina Queen, CIM-1100, CIM-448, GH-8 and NIAB-313 were less leaky than CIM-435, BH-89 or NIAB-78 while DNH-40, CIM-443 and SLS-171 were susceptible to heat damage (Figure 6). Two *Gossypium barbadense* varieties, Pima-S6 and Dandra, were among the least leaky *Gossypiums*.

Figure 7 shows that the ranking of varieties in the two tests is dependent on environment, i.e. the two sets of data are not closely related. The Multan data shown here was obtained 85 days after planting. Data from 70 days after planting gave a slightly different rank order and was not so closely correlated with seed cotton yield (see Figure 1).

As leaf temperature increases, net photosynthesis rates fall, but stomatal conductance and transpiration rates are maintained. On-going research is examining varietal differences in transpirational cooling of leaves.

Conclusions

Chlorophyll fluorescence analysis and electrolyte leakage provide information about different aspects of cellular heat tolerance of cotton leaves, and gas exchange studies, in the field or in defined conditions, examine transpirational cooling effects. However, only whole plant experiments integrate all aspects of the effects of high temperatures on growth and yield of cotton.

Acknowledgements

This document is an output from a project funded in part by the Department for International Development Plant Sciences Research Programme, managed by the Centre for Arid Zone Studies, University of Wales, Bangor, for the benefit of developing countries. The views expressed are not necessarily those of DFID.

References

Akhtar, L.H., J. Gorham, M.A. Mirza and A.L. Sheikh. (1996): Heat stress tolerance in the wild species of the genus Gossypium measured by chlorophyll fluorescence. The Pakistan Cottons 40:49-58.

Ashraf, M., M.M. Saeed and M.J. Qureshi. (1994): Tolerance to high temperature in cotton (*Gossypium hirsutum* L.) at initial growth stages. Environ Exp Bot; 34:275-83.

Sethar M.A., D.L. Laidman, J.M. Wilson and R.B. Mirbahar. (1995) Hardening potential in cotton (*Gossypium hirsutum* L.) at high temperature stress. Pakistan Journal of Botany; 27:165-8.

Taha M.A., M.N.A. Malik, F.I. Chaudhry and M.I. Makhdum. (1981): Heat-induced sterility in cotton sown during early April in West Punjab. Exp. Agric. 17:189-94.

Figure 1. Meteorological data, Multan, 1995.

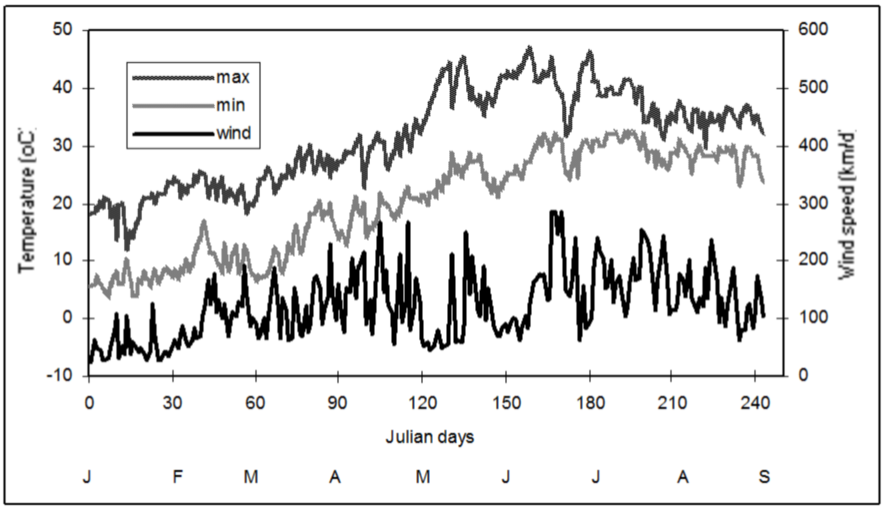

Figure 2. Electrolyte leakage (16 varieties, Multan, 1 hour at 50oC) vs seed cotton yield (April sowing).

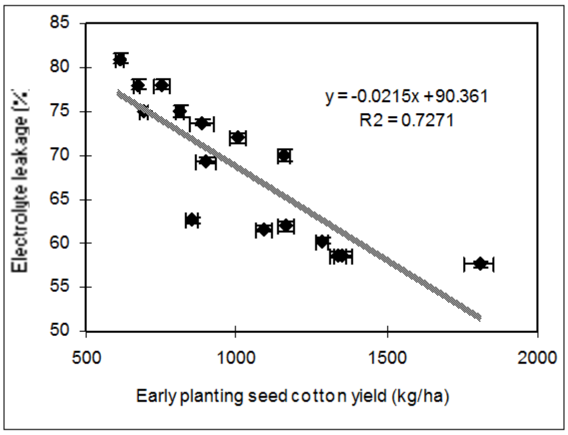

Figure 3. Fv/Fm ratios (Bangor, after 3 hours at 45oC as % of controls) vs seed cotton yields (April sowing).

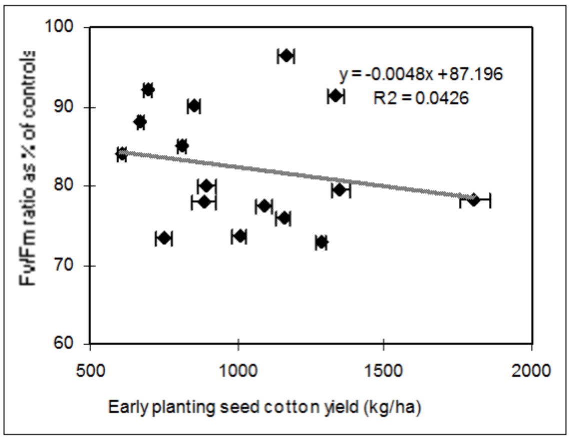

Figure 4. Decrease in Fv/Fm ratios at 46oC.

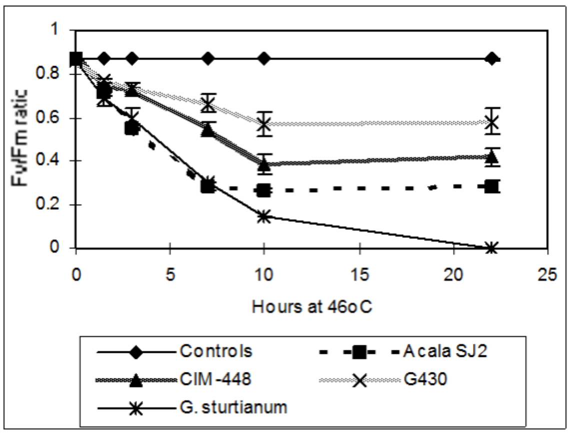

Figure 6. Changes in net photosynthesis (Pn) and stomatal conductance (gs) with increasing leaf temperature.

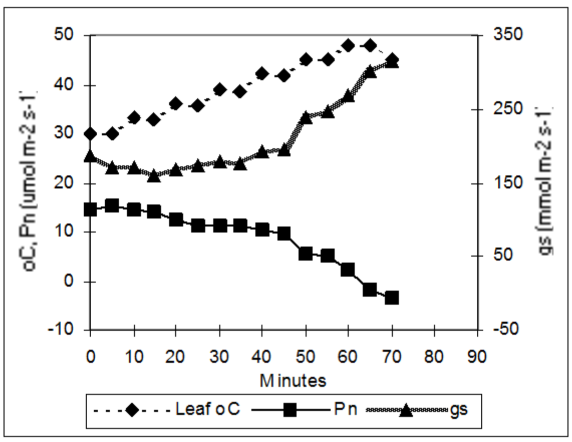

Figure 5. Electrolyte leakage (Bangor).