GENETIC VARIATION IN COTTON FOR TOLERANCE TO WATERLOGGED CONDITIONS

Warren Conaty[1], Greg Constable[2] and Daniel Tan[1]
[1] Faculty of Agriculture, Food and Natural Resources, The University of Sydney, Sydney, NSW 2006,
[2] CSIRO Division of Plant Industry, Australian Cotton Research Institute, Narrabri, NSW 2390.

Introduction

Waterlogging is a world-wide phenomenon that affects both the distribution of plants in their natural ecosystems as well as crop yield in agricultural regions. Waterlogging can severely affect cotton growing in the heavy clays (vertosols) in Australia. The annual production losses due to waterlogging in Australia is $A180 million (Price 1993). Cotton production in Australia is:

- is focused in regions with heavy clay soils with inherent low drainage rates;
- is characterised by field with low gradients;
- is almost exclusively furrow irrigated; and

As cotton is grown in summer dominant rainfall regions, which have a high probability of intense summer storms, there is a high likelihood of disruption of irrigation schedules and the occurrence of waterlogged conditions. Waterlogging can result in yield reductions of up to 10% (Bange *et al.* 2004).

Waterlogging occurs in saturated soils, when the air filled porosity (AFP) falls below 10% (Hodgson 1982). In a well drained soil, the air filled porosity usually ranges from 10 to 40% of the total soil volume. However, waterlogging reduces these gas-filled pores, substantially diminishing the supply of oxygen to the roots resulting in hypoxia. This occurs due to the prevention of the replacement of oxygen consumed in the soil.

Waterlogging has a wide range of effects on cotton including decreased root and shoot biomass as well as an increase in root and leaf senescence (Bange *et al.* 2004; Hodgson and Chan 1982). Waterlogging also affects nutrient availability, uptake, translocation and leaf nutrient concentration (Hocking *et al.* 1985). Furthermore, hypoxia results in decreased stomatal conductance and leaf water potential, and hence, reduced photosynthesis.

This study evaluates the physiological responses of fourteen cotton genotypes under waterlogged and non-waterlogged irrigated conditions in relation to leaf nutrient level, leaf colour, leaf photosynthesis, plant morphology and final yield.

Methodology

Thirteen different upland cotton (*Gossypium hirsutum*) and one pima cotton genotype (*Gossypium barbadense*) from a range of natural environments were subjected to waterlogged and non-waterlogged irrigated (control) conditions at the ACRI, Narrabri (see Table 1). Control plots were irrigated based on Best Management Practices. Waterlogged plots were subjected to optimal irrigation schedules plus 48 to 72 hours of additional irrigation. The physiological responses of these cotton genotypes were assessed in relation to leaf nutrient status, leaf colour, leaf photosynthetic rate, plant morphology and crop yield. More regular measurements were undertaken on four cotton genotypes (Georgia King, LA 887, Sicot 71 and Sicot 73) with variable responses to waterlogged conditions.

Table 1. Cotton genotypes, origin and environmental conditions used in this study

Variety	Origin	Environment	Soil Type
CIM 443	Pakistan	Dry	Clay
Coker 315	USA- Texas	Cool	Clay
Gohar 87	India	Dry	Clay
Sicot 71	Australia- CSIRO	Dry	Clay
Sicot 73	Australia- CSIRO	Dry	Clay
Sicot 80	Australia- CSIRO	Dry	Clay
Codetec 401	Brazil	Wet	Loam
Deltapine 16	USA- Mississippi	Humid	Silt
Deltapine 90	USA- Arizona	Hot	Loam
Georgia King	USA- Georgia	Humid	Silt
LA 887	USA- Louisiana	Humid	Silt
McNair 1032	USA- South Carolina	Humid	Silt
PD93057	USA- South Carolina	Humid	Silt
Pima A-8	USA- Arizona	Hot	Loam

Leaf Nutrient Status

The first fully expanded leaf was collected at regular intervals, dried and analysed via Inductively Coupled Plasma Mass Spectrometry (ICP-MS) for phosphorus (P), potassium (K), iron (Fe), manganese (Mn), magnesium (Mg), calcium (Ca), copper (Cu), zinc (Zn), boron (B), aluminium (Al) and sodium (Na).

Leaf Colour

Leaf colour was measured using the Minolta Spad-502.

Leaf Photosynthetic Rate

Leaf photosynthetic rates were measured 24 hours before waterlogging and then again 24 hours after the irrigation event for the control and during waterlogging.

Boll Retention and Yield

Boll retention and yield (kg of seed/ha) were measured.

Results

Leaf Nutrient Status

Leaf blade concentrations of phosphorus varied between genotypes ($P<0.001$). Phosphorus concentrations decreased due to waterlogging except for the genotypes Sicot 80, PD93057, Sicot 73 and Sicot 71.

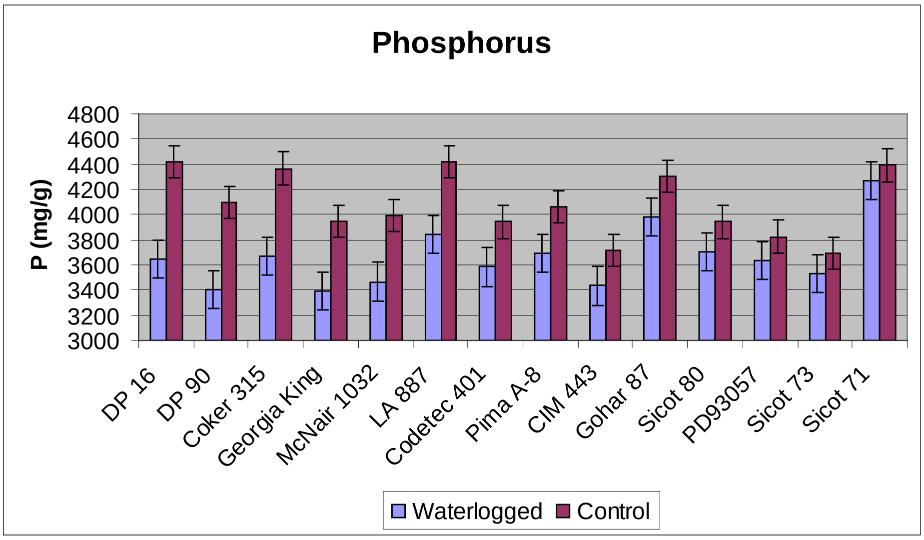

Figure 1. Phosphorus concentrations (mg/kg) in leaves of irrigated and waterlogged genotypes.

Leaf blade concentrations of potassium varied between genotypes (P<0.001). Potassium concentrations decreased due to waterlogging except for Coker315, Pima A-8, PD93057, Codetec 401, McNair 1032, Sicot 71, Gohar 87 and Sicot 73.

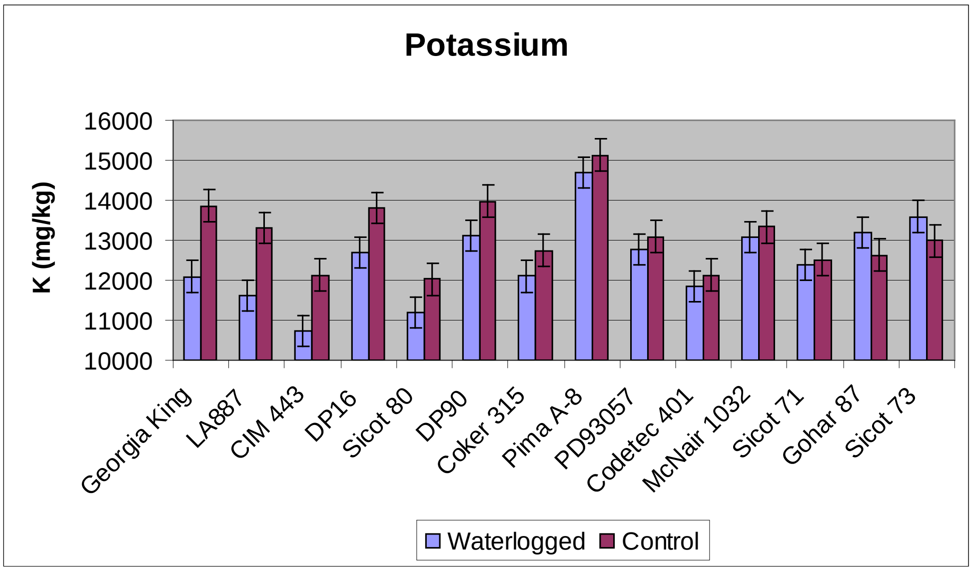

Figure 2. Potassium concentrations (mg/kg) in leaves of irrigated and waterlogged genotypes.

Leaf iron concentration increased in all genotypes due to waterlogging (Figure 3). Genotypes displayed different concentrations of iron (P<0.001). However, no genotypes studied showed similar iron leaf blade concentrations in control and waterlogged plots. Symptoms of iron chlorosis (yellowing cf. Figure 6) is simply not due to the iron concentration in the leaf. However, this anomaly is due to the fact that waterlogging reduces the available forms of iron and increases unavailable iron concentrations.

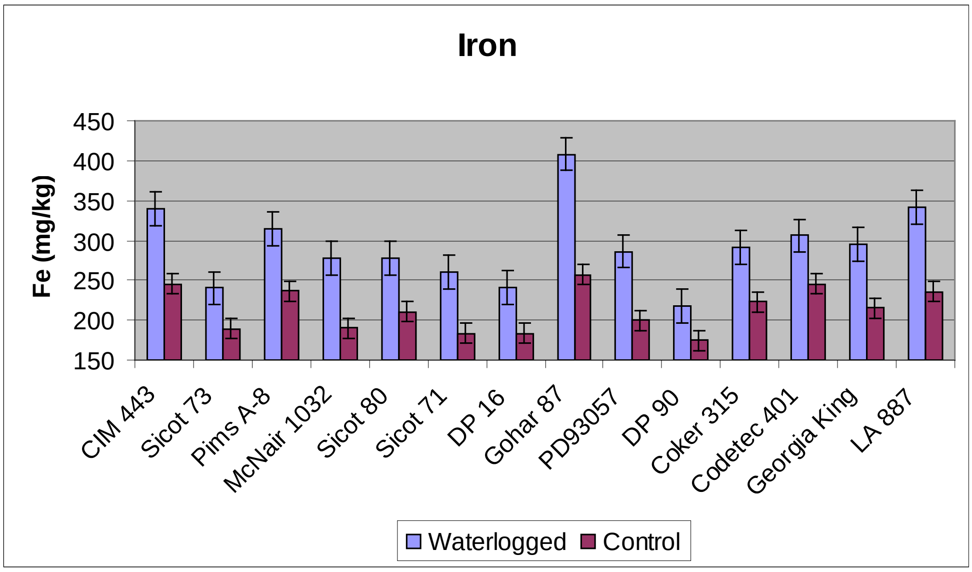

Figure 3. Iron concentrations (mg/kg) in leaves of irrigated and waterlogged genotypes.

Photosynthetic Rate

Pre-waterlogging, photosynthetic rates were higher in waterlogged treatments (P=0.007) due to higher soil moisture availability (data not shown) (Figure 4). However, similar effects were observed between the cultivars (P=0.652) in the same treatment.

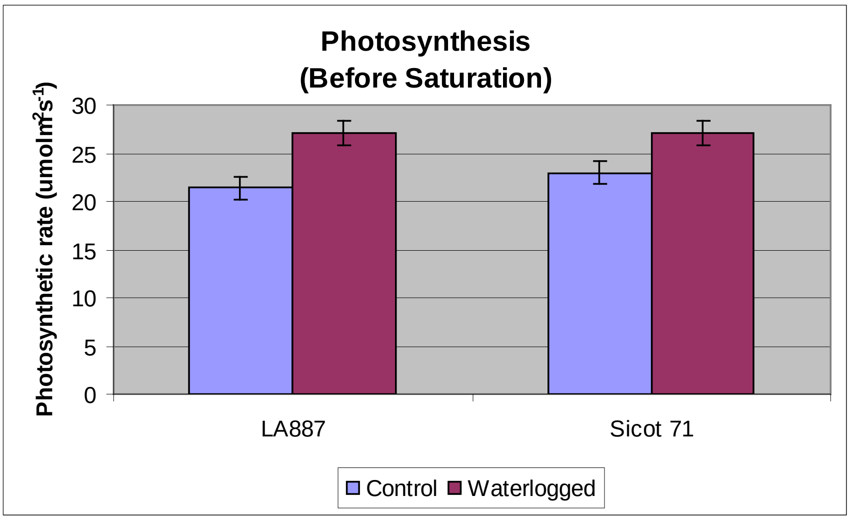

Figure 4. Leaf photosynthetic rates of genotypes LA 887 and Sicot 71 before irrigation.

Following saturation, photosynthetic rate was higher (P=0.018) in the two cultivars LA 887 and Sicot 71 in the control compared to the waterlogging treatment (Figure 5). However, Sicot 71 did not show statistically different photosynthetic rates across treatments (P=0.693).

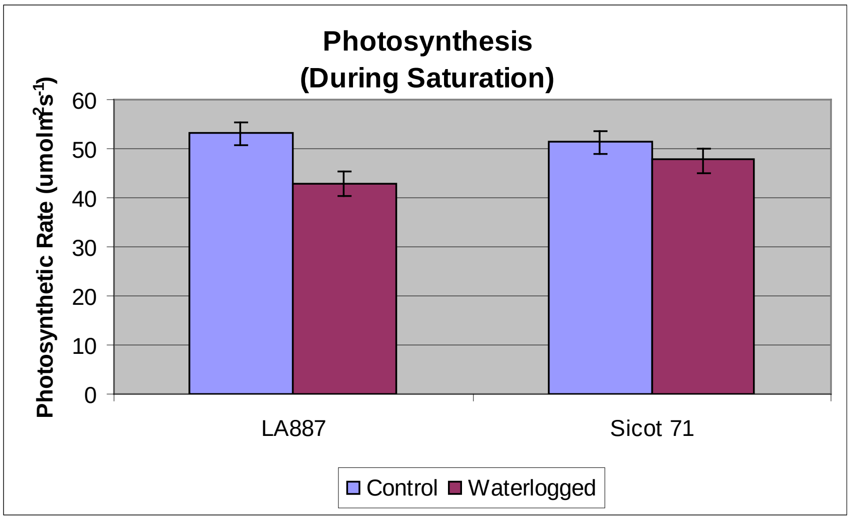

Figure 5. Leaf photosynthetic rates of the genotypes LA 887 and Sicot 71 post irrigation (control) and during waterlogging.

Leaf Colour

Leaf colour was strongly affected by waterlogging, resulting in yellowing of leaves (P<0.001). However some genotypes were not as affected by waterlogging. The genotypes that had no difference in leaf colour in response to waterlogging were Sicot 71, Sicot 73, Coker 315, Sicot 80 and Pima A-8.

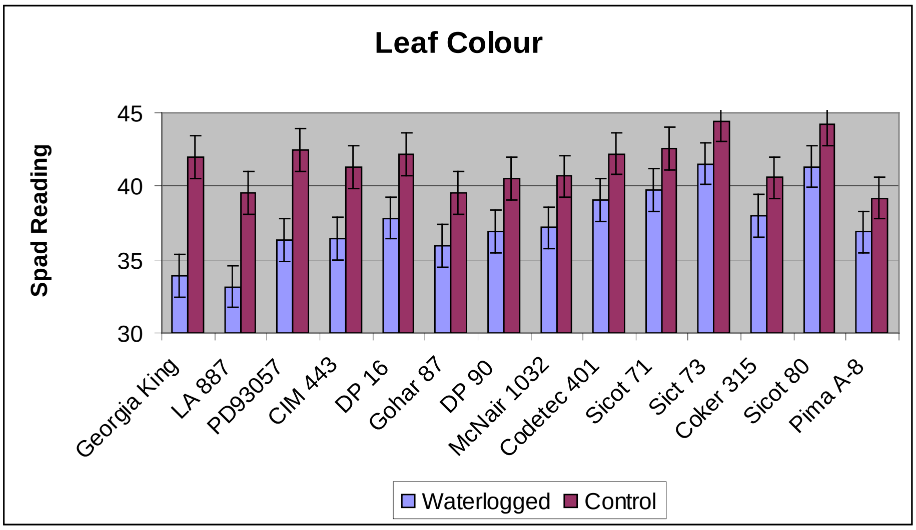

Figure 6. Relative leaf colour for waterlogged and control irrigated genotypes.

Boll Retention and Yield

There was a higher (*P*=0.007) boll retention rate in the control compared to the waterlogging treatment in all cultivars except Sicot 7 (Figure 7). Yield results (kg seed/ha) are shown in Figure 8.

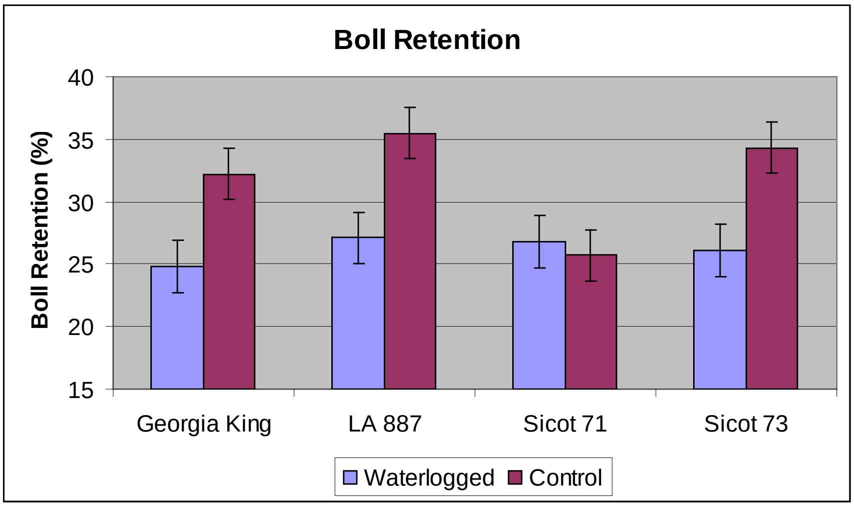

Figure 7. Boll retention rates for Georgia King, LA 887, Sicot 71 and Sicot 73 under waterlogged and control irrigated conditions.

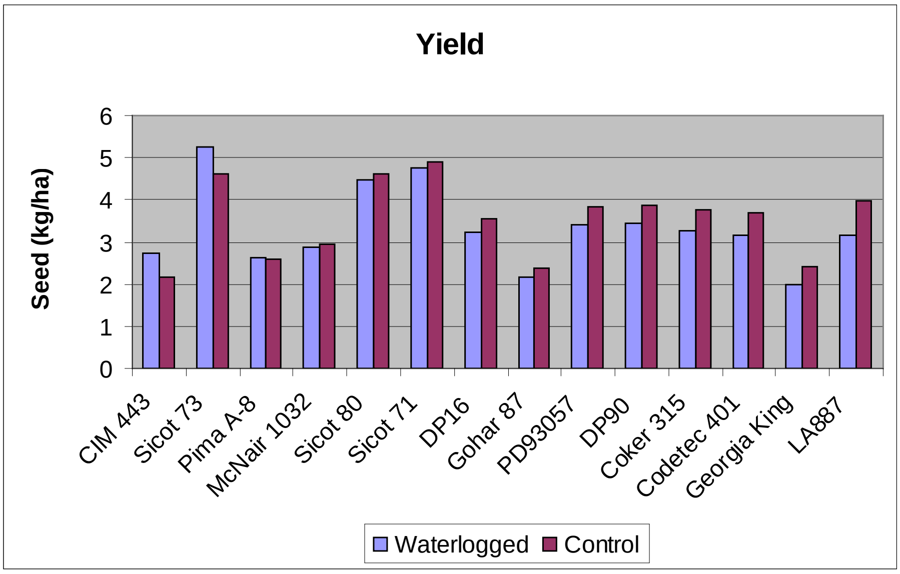

Figure 8. Yield (kg seed/ha) for waterlogged and control irrigated genotypes (Note: 1kg seed/ha is approximately equal to 40kg ginned lint/ha).

Discussion

Leaf blade nutrient concentration

Waterlogging reduced both phosphorus and potassium leaf blade concentrations. This can be attributed to inhibited uptake under anaerobic conditions (Trought and Drew 1980). These deficiencies in waterlogged plants also occur due to the inhibition of ion uptake and the translocation of nutrients from older leaves to supply the nutrients to the younger leaves (Drew 1983). At present, there have been no studies that investigate whether waterlogging impacts on the population of active arbuscular mycorrhizal fungi (AMF). Such a study is necessary in order to determine if waterlogging adversely affects AMF populations, and thus, the potential phosphorus uptake.

Waterlogging increased the iron concentration in all genotypes. Numerous studies on cotton and other field crops have shown an increase in iron concentration following waterlogging (Hodgson 1990). This increase may be due to the effect of increased iron availability in the soil. When a soil is inundated, ferric iron (Fe^{3+}) is reduced to the more soluble ferrous (Fe^{2+}) ions, increasing soil iron availability. This chemical reaction is characterised by the transformation of the brown colour in an iron-rich soil to shades of grey as large amounts of iron ions are brought into solution (Ponnamperuma 1984). This effectively increases the availability of these elements, which can reflect an increase in total iron concentration of waterlogged plants.

However, acute deficiencies of iron can still occur in waterlogged cotton. This is because iron nutrition should reflect the active chemical form of iron (Fe^{2+}), rather than total iron content. Therefore total iron concentrations can be misleading (Figure 3) as it is the active form of iron, Fe^{2+} that is critical for plant nutrition and chlorophyll production (Hodgson 1990). The inherent problem in iron nutrition is that the determination of the Fe^{2+} content cannot be achieved by a commercial laboratory, and requires the use of fresh leaves, analysed a few hours after sampling.

An iron deficient plant will show signs of interveinal yellowing. The veins usually remain green, but if the deficiency is extreme the leaf can become totally chlorotic and turn white. Ironically, the total iron concentration in chlorotic leaves is often similar to, or higher than in green leaves. This is because the majority of this iron is unavailable for use by plants as it is in the inactive Fe^{3+} form (Hodgson 1990). Iron deficiencies occur in cotton because as the soil becomes increasingly waterlogged, the diffusion of CO_2 gas out of the soil is reduced, creating a build up of CO_2 in the soil. This results in the formation of bicarbonate ions, increasing both the soil pH and the alkalinity of the leaf tissue. Under these conditions, available ferrous iron (Fe^{2+}) is reduced to inactive forms of iron (predominantly ferric or Fe^{3+}) (Hodgson 1990). For each unit increase in leaf pH, the availability of iron is decreased approximately 100-fold (Hodgson 1990). Therefore, waterlogging results in the reduction of available iron which the plant must convert into ferrous iron before it can be used. This process is costly to a plant already suffering from reduced metabolism associated with hypoxia, resulting in iron deficiencies.

Foliar application of iron (II) sulphate on cotton can help ameliorate the effects of iron chlorosis, returning cotton foliage to its normal colour. In addition, foliar sprays of iron chelates applied to crops before irrigation and subsequent waterlogging events can help prevent iron deficiencies. Despite the risk of foliar burn, a single spray on cotton at Narrabri increased lint yields by 6%, and two foliar sprays increased lint yield by 11% (Hodgson 1990).

Photosynthetic Rate and Leaf Colour

Photosynthetic rates in response to flooding were reduced by 19% in the susceptible genotype (LA 887) and 7% in the more tolerant genotype (Sicot 71). Similar results were observed by Meyer *et al.* (1987) which saw photosynthetic rates in cotton decreased by 16% following seven days of waterlogging. Flooding and subsequent waterlogging of the soil is usually followed by a rapid reduction in the rate of photosynthesis in waterlogging susceptible plants (Kozlowski and Pallardy 1984). Photosynthesis of waterlogged plants is reduced initially by stomatal closure and later by a reduction in photosynthetic capacity (Kozlowski and Pallardy 1984). Plants under waterlogging stress have reduced stomatal aperture, and transpiration rates were reduced. This helps prevent waterlogging-induced wilting but also reduced photosynthetic rate.

Chlorosis of leaves, resulting from a reduction in the level of chlorophyll in the leaf, is often one of the early symptoms of waterlogging. Leaf yellowing was observed in all genotypes to varying levels of significance. As chlorophyll is the light capturing pigment in plants, a reduction in its level in the leaf will reduce photosynthetic rate. Decreased chlorophyll content due to waterlogging has been reported in numerous *Brassica* species (Ashraf and Mehmood 1990), barley (Pang *et al.* 2004), maize (Bragina *et al.* 2004), field pea (Ladygin 2004) and okra (*Hibiscus* *esculantus*) (Ashraf and Arfan 2005). The inhibition of chlorophyll production in leaves is at least partly responsible for a reduction in photosynthetic rates (Bragina *et al.* 2004). The importance of chlorophyll content to photosynthesis is emphasised by a parallel decline in both chlorophyll and photosynthetic rate (Kozlowski and Pallardy 1984).

Boll Retention and Yield

Waterlogged conditions affect almost every aspect of shoot behaviour and thus final yield of the crop. The effects of waterlogging on the shoot are due to modifications in internal flow of water, hormones, toxins, inorganic nutrients and photosynthates from the root to the shoot. Waterlogging reduced boll retention in the genotypes Georgia King by 23%, LA 887 by 24% and Sicot 73 by 24%. No difference in boll retention was observed in Sicot 71 as a result of waterlogging. These results are parallel with those observed by Bange *et al.* (2004). According to Bange *et al.* (2004) dry matter accumulation was reduced by 32% due to a 35% reduction in radiation use efficiency. The reduction in dry matter (32%) resulted in a decreased yield, achieved principally through decreased boll retention, and hence, reduced boll number.

These reductions in boll retention rates due to waterlogging reduced crop yields. Waterlogging substantially reduced the yield (kg/ha seed) of waterlogging sensitive varieties LA 887 by 21%, Georgia King by 17%. In contrast, yields of genotypes with more tolerance to waterlogged conditions were only reduced by 4% for Sicot 71 and 3.4% for Sicot 80. These differences in yield are probably due to boll retention rates.

Conclusion

Waterlogging reduces vegetative growth, dry matter accumulation and subsequently, crop yield. The physiological consequences of waterlogged conditions include reduced photosynthetic rate, reduced water and nutrient uptake, and altered shoot and root hormonal status.

There is some genetic tolerance to waterlogged conditions when comparing the physiological response of the more susceptible varieties (LA 887 and Georgia King) with varieties with increased tolerance (Sicot 71, Sicot 73 and Sicot 80). Until this study, there have been a limited number of studies investigating the genetic variation in the response of cotton genotypes to waterlogging stress. The knowledge obtained from further studies into these areas may be utilised in agronomic field management practices and conventional and transgenic breeding programs. Such manipulation of genes controlling these morphological adaptations has the potential to enable sensitive plant species to endure longer periods of waterlogging before cellular functions cease due to a lack of energy.

However, it is important that correct agronomic management and best management practices are still observed in relation to waterlogging. Improvements to field gradients through laser levelling, shortening of furrow lengths and the implementation of two metre beds are some methods to reduce of the extent of waterlogging stress in furrow irrigation. Furrow irrigation efficiency can be increased through the use of larger siphons and the broadening of tail drains. This will reduce the inundation period and therefore, the potential period of waterlogging stress.

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