PHYSIOLOGICAL APPLICATIONS FOR DETERMINING WATER USE EFFICIENCY AMONG COTTON GENOTYPES

A Dissertation

by

JOSHUA BRIAN BYNUM

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

May 2008

Major Subject: Agronomy

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Approved by:

Chair of Committee,	J. Tom Cothren
Committee Members,	Robert G. Lemon
	Leonardo Lombardini
	Maria Balota
Head of Department,	David Baltensperger

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ABSTRACT

Physiological Applications for Determining Water Use Efficiency

Among Cotton Genotypes. (May 2008)

Joshua Brian Bynum, B.S. West Texas A&M University; M.S., Texas A&M University

Chair of Advisory Committee: Dr. J. Tom Cothren

Drought stress can substantially alter plant metabolism by decreasing plant growth and photosynthesis. The lack of rapid and reliable screening criteria and measurement techniques for determining water use efficiency (WUE) of crop plants has greatly restricted progress in this critical area of crop improvement. In grain sorghum (Sorghum bicolor L.), WUE was associated with the transpiration ratio [CO2 assimilation (A) / transpiration rate (E), A:E] from leaf gas exchange measurements. Research is needed to identify drought effects on plant productivity and to exploit the use of this knowledge in breeding and agronomic efforts. Therefore, the objectives of this study were to determine if differences in A:E and other physiological parameters existed between two selected cotton (*Gossypium hirsutum* L.) genotypes and to evaluate the response of cotton genotypes experiencing water stress at two different growth stages on biomass production and yield.

Two experiments were conducted using two cotton genotypes differing in drought tolerance. Each experiment was repeated three times in a randomized complete block design with six replications. In Experiment I, the water stress treatment was induced by withholding water when the plants reached the 4-node growth stage. The

water stress treatment in Experiment II was imposed at early bloom. Gas exchange and chlorophyll fluorescence measurements were collected during dry-down and recovery periods to determine water stress effects on plant physiology. Biomass was partitioned following the recovery period, to examine phenotypic responses of plants exposed to water stress.

The results of these experiments indicate that A:E is significantly increased as leaf water potential (ψ_L) decreases with no differences in A:E between the two genotypes. Gas exchange measurements showed significant decreases with declining ψ_L and significant increases upon re-watering; yet, no differences were observed between the two genotypes. Chlorophyll fluorescence was not different between genotypes in either light- or dark-adapted leaves. In Experiment I TAM 89E-51 had a significantly greater seedcotton yield; however, in Experiment II TAMCOT 22 had the greater yield. These experiments suggest that the effects of water stress on cotton are a function of the intensity of the stress and the growth stage in which the stress is experienced.

DEDICATION

This dissertation is dedicated to my beautiful wife and best friend, Katie, whose love, encouragement, support, friendship, and sacrifice made it possible for me to complete this degree.

ACKNOWLEDGEMENTS

I would first like to thank God, through whom all things are made possible, for blessing me with such a rich life and the ability to complete this task.

I would like to recognize and extend appreciation to the following members of my graduate committee for their support and mentorship: Dr. Tom Cothren, committee chair; Dr. Robert Lemon, member; Dr. Leonardo Lombardini, member; and Dr. Maria Balota, member. Their assistance, encouragement, and support are greatly appreciated.

Recognition is given to my co-workers and peers: Vladimir da Costa and Danny Fromme. I would also like to thank the following student workers for countless hours spent providing help with this project: Charles Swanson, Joerdan Kennedy, Ellen Batchelder, Nicola Cottee, Matt Nors, and Charles (Jr.) Carden.

I would like to extend special gratitude to Justin Scheiner for his assistance and friendship not only on this project but throughout his stay in College Station.

Special appreciation is given to my committee chair, advisor, mentor, and confidant, Dr. Tom Cothren, for his advice, example, support, and generosity. Dr. Cothren has allowed me to pursue what has become the greatest opportunity of my professional life. I am forever indebted to Dr. Cothren for his teachings, mentorship, and friendship.

Special appreciation is also extended to Dr. Robert Lemon for his advice, mentorship and support over the last six years. Dr. Lemon has made me a better researcher through his example and mentorship.

Thanks is also extended to my beautiful wife Katie, and loving family Kevin, Ronda, Tim, Charlotte, Rex, Brenda, Dustin, Lynsie, Marlie, and Brady, for their unconditional and continuous support, encouragement, and love.

Difficult tasks in life can be made much easier by the people around you who offer their friendship, love, support, advice, and encouragement. To those mentioned on this page, “thanks”, and I am forever grateful.

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CHAPTER I

INTRODUCTION

Drought is considered a predominant factor for restricting crop yields in agriculture. An understanding of the response of plants to water deficit is important in efforts to model cotton growth, estimate irrigation needs, and breed drought-resistant cultivars (Pace et al., 1999). The ability of breeders to select more water use efficient cultivars has the potential to seriously impact the economic return for producers.

Cotton (*Gossypium hirsutum* L.) is considered one of the most drought-tolerant field crops grown in the United States. However, the Federal Crop Insurance Corporation rates drought the greatest cause of disasters experienced by cotton producers. While considered to be drought-tolerant relative to other crop species, cotton responds well to supplemental water by producing yields proportional to rainfall or supplemental irrigation. Reports suggest that irrigation may enhance cotton yields by 224 to 448 kg ha^{-1} over dryland. Of the 5.6 million hectares of cotton planted in 2005 in the United States, ~70% were under dryland production. With the large hectarage of cotton under dryland or limited irrigation practices, the utility of water use efficient cultivars would convey a tremendous advantage to producers and the market. Cotton cultivars that can endure and/or recover from drought are needed to minimize yield loss in dryland areas and to reduce the water needs of irrigated production.

The lack of simple, rapid, and reliable screening criteria and measurement

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This dissertation follows the style and format of Crop Science.

techniques for water use efficiency (WUE) of crop plants has greatly restricted progress in this critical area of crop improvement (Hall et al., 1990). In grain sorghum (*Sorghum bicolor* L.), WUE was associated with the transpiration ratio [CO_2 assimilation (A) / transpiration rate (E), A:E] from leaf gas exchange measurements (Peng and Krieg, 1992). A better understanding of how drought stress affects physiological parameters and overall plant growth is fundamental. Research is needed to identify how plant productivity is affected by drought and to exploit this knowledge in breeding and agronomic efforts to improve yield under drought stress and WUE.

LITERATURE REVIEW

Drought Stress and Plant Productivity

Drought stress can substantially alter plant metabolism by decreasing plant growth and photosynthesis, thus having a profound affect on agriculture (Tezara et al., 1999). The nature and extent to which drought stress affects plants is a function of the intensity and duration of the stress, as well as of the genetically-determined capacity of species to cope with the environment (Chaves, 1991).

Drought stress is purported to impact a number of plant processes and elicit many detrimental responses on plant productivity. Drought stress is reported to reduce leaf expansion (Hsiao, 1973; Masle and Passioura, 1987), reduce the number of leaves on sympodial branches (Krieg and Sung, 1986), decrease leaf elongation rate (Cutler and Rains, 1977), lower dry matter accumulation and water use efficiency (Quisenberry et al., 1981), and to decrease plant height, leaf area, and total nodes (Pace et al., 1999). Many of these responses, such as reduced growth and decreased size of leaf canopy due

to stress-induced senescence, are of potential value for plant survival and adaptation to drought (Chaves, 1991).

Productivity of plants is greatly depressed by water deficiency (Austin et al., 1986). However, the effect of water stress on the mechanisms of CO_2 assimilation (Boyer, 1971; Boyer, 1976; Boyer and Younis, 1983; Lawlor, 1983; Longstreth et al., 1980; Sharkey and Seemann, 1989) has not been analyzed in sufficient detail, in genotypes differing in assimilation capacity, to show how the photosynthetic mechanism may be modified to improve assimilation under water stress (Fischer et al., 1981; Lawlor, 1983; Mahon and Hobbs, 1981; Sharkey and Seemann, 1989; Von Caemmerer and Farquhar, 1981). Currently, there is no single measure of plant water status that can be correlated with all the numerous effects of water stress (Kramer, 1988).

Leaf Gas Exchange

Photosynthesis is an essential process to maintain plant growth and development. It is well known that photosynthetic systems in higher plants are sensitive to drought stress (Falk et al., 1996). The rate of photosynthesis, in combination with the leaf area, determines plant productivity (Austin et al., 1986; Gillford and Evans, 1981; Gutteridge and Keys, 1985; Lawlor et al., 1989). Therefore, the functional state of photosynthesis has been considered an ideal physiological activity to monitor the health and vitality of plants (Clark et al., 2000). It is generally assumed that drought-induced decreases in photosynthesis are due primarily to stomatal closure, which decreases CO_2 availability in the mesophyll, rather than to the direct effect on the capacity of the photosynthetic apparatus (Genty et al., 1987; Cornic 1994). Although stomatal regulation of leaf gas

exchange under drought conditions has been well documented (Tenhunen et al., 1987), inactivation of photosynthetic activity due to non-stomatal effects has also been reported (Boyer et al., 1987). Lawlor (1995) presented strong evidence that water stress affects mesophyll metabolism and reduces photosynthetic capacity. This occurred as a consequence of decreased ribulose bisphosphate (RuBP) synthesis (Gimenez et al., 1992), decreased rubisco activity and reductions in carboxylation efficiency (Martin and Ruiz-Torres, 1992), or both (Plaut and Federman 1991; Faver et al., 1996). Tezara and Lawlor (1995) reported that stomatal control of photosynthetic rate becomes progressively less effective as water stress intensifies. It is well established that the rate of photosynthesis is depressed at moderate leaf water deficits (Lange et al., 1971). Several reports have documented the remarkable resistance of the photosynthetic apparatus to dehydration, which allows for the maintenance of full photosynthetic capacity by the leaves, thus permitting a rapid recovery of the plant after rehydration (Kaiser, 1982, 1987; Cornic et al., 1989). Despite great progress in understanding the effects of water stress on photosynthesis, there is still no unified concept of the events that reduce photosynthetic efficiency (Lawlor, 1995).

Chlorophyll Fluorescence

A review of the literature overwhelmingly reveals in excess of 3500 papers on chlorophyll fluorescence, of which ~20% have implications for agricultural issues. Chlorophyll fluorescence has evolved as a very useful and informative indicator for photosynthetic electron transport in intact leaves, algae, and isolated chloroplasts (Briantais et al. 1986; Renger and Schreiber 1986; Schreiber and Bilger 1987, 1992;

Krause and Weis 1991; Karukstis 1991). Assessing the health or integrity of the internal apparatus driving the photosynthetic process within a leaf (i.e., the thylakoid membrane) using chlorophyll fluorescence provides a rapid and accurate technique of detecting and quantifying plant tolerance to stress (Glynn et al., 2002).

Light energy absorbed by chlorophyll molecules in a leaf can undergo one of three fates: it can be used to drive photosynthesis (photochemistry), excess energy can be dissipated as heat or it can be re-emitted as light (chlorophyll fluorescence). The above three processes occur in competition, such that any increase in the efficiency of one will result in a decrease in the yield of the other two. Therefore, by measuring the yield of chlorophyll fluorescence, information about changes in the efficiency of photochemistry and heat dissipation are gained (Maxwell and Johnson, 2000). For reasons so far unknown, at room temperature the variable fluorescence originates almost exclusively from photosystem II (PSII) (Schreiber et. al., 1986). Hence, fluorescence changes reflect primarily the state of PSII and the operating efficiency of PSII is related to A (Genty et al., 1989, 1990). The PSII operating efficiency is determined by the ability to drive photochemistry rather than non-photochemical processes. This relationship between the PSII operating efficiency and A in leaves allows fluorescence to be used to detect differences in the response of plants to environmental challenges and, consequently, to screen for tolerance to environmental stresses such as drought. Since photosynthetic traits are closely correlated with the rate of carbon exchange, chlorophyll fluorescence parameters allow us to estimate the influence of environmental

stress on plant growth and yield (Araus et al., 1998; Guo and Li 2000; Fracheboud et al., 2004).

OBJECTIVES

The objectives of this study are to (i) determine if differences in A:E and physiological parameters, such as gas exchange rate and chlorophyll fluorescence, exist between two selected cotton genotypes and (ii) evaluate the response of two different cotton genotypes experiencing water stress at two different growth stages on biomass production and yield.

CHAPTER II

METHODS AND MATERIALS

PLANT MATERIAL AND TREATMENTS

Two experiments were conducted using two cotton (*Gossypium hirsutum* L.) genotypes differing in drought tolerance chosen from the Texas A&M University Cotton Improvement Laboratory. Selection was based on previous drought tolerance experiments (unpublished data). Plants of the more tolerant cultivar, TAMCOT 22, and the less tolerant experimental line, TAM 89E-51, were grown in plastic pots (31 cm in diameter and 31 cm in height) under glasshouse conditions at the Plant Growth Facility of the Institute of Plant Genomics and Biotechnology at Texas A&M University. Each pot was filled with 12 kg of sterilized potting mix. The potting mixture contained peat moss, vermiculite, perlite, and dolomitic lime stone. Nutrient requirements were supplied weekly using a 20-20-20 complete fertilizer. Air temperature during the day and night was maintained at 28 °C with a photoperiod of 14L/10D h. Each experiment was repeated three times and all were designed as a randomized complete block with six replications. An additional set of six replicates were potted for biomass partitioning. Leaf water potential (ψ_L) was maintained at values > -1.3 MPa by frequent watering except when stress treatments were imposed. The gradual water stress was monitored daily using a pressure chamber to measure ψ_L on the third fully expanded leaf from the plant apex. When ψ_L values reached ~3.5 MPa, pots were rewatered and recovery measurements were collected. In Experiment I, the water stress treatment, induced by withholding water, occurred when the plants reached the 4-node growth stage.

Experiment II was conducted similarly to Experiment I; however, in Experiment II the water stress treatment was initiated at first flower.

LEAF GAS EXCHANGE MEASUREMENTS

Gas exchange measurements were collected from the third fully expanded leaf from the plant apex using a portable LI-6400 (LI-COR Inc., USA). CO_2 assimilation rate (A) [μmol (CO_2) m^{-2} s^{-1}], stomatal conductance (g_s) [mol (H_2O) m^{-2} s^{-1}], intercellular CO_2 concentration (C_i) [μmol (CO_2) mol^{-1} (air)], and transpiration rate (E) [mol (H_2O) m^{-2} s^{-1}] were calculated as described in the LI-6400 users manual (LI-COR Biosciences, 2002). The relative humidity of the leaf chamber (6 cm^2) was set at 50% during measurements. The CO_2 flux was regulated to an inside leaf chamber concentration of 370 μmol (CO_2) mol^{-1}. A light response curve of control (well-watered) plants was conducted to determine the light saturation level of each genotype (Fig. 1). The light response curve indicated that both genotypes were light-saturated at a photosynthetic photon flux density (PPFD) of 1200 μmol (photons) m^{-2} s^{-1}. Therefore, a light source (6400-02B LED; LI-COR Inc., USA) was used to maintain PPFD at 1200 μmol (photons) m^{-2} s^{-1} during gas exchange measurements. Measurements were collected in two-day intervals from 1000 to 1300 h, similarly in both experiments.

CHLOROPHYLL FLUORESCENCE MEASUREMENTS

Light- and dark-adapted leaves were used to measure chlorophyll fluorescence with a PAM-2100 portable fluorometer (Heinz Walz GmbH, Germany). Dark adaptation was obtained by attaching light-exclusion clips to the leaf surface *in situ* for 30 minutes prior to each reading. Minimal fluorescence (F_o) and the maximum quantum yield of

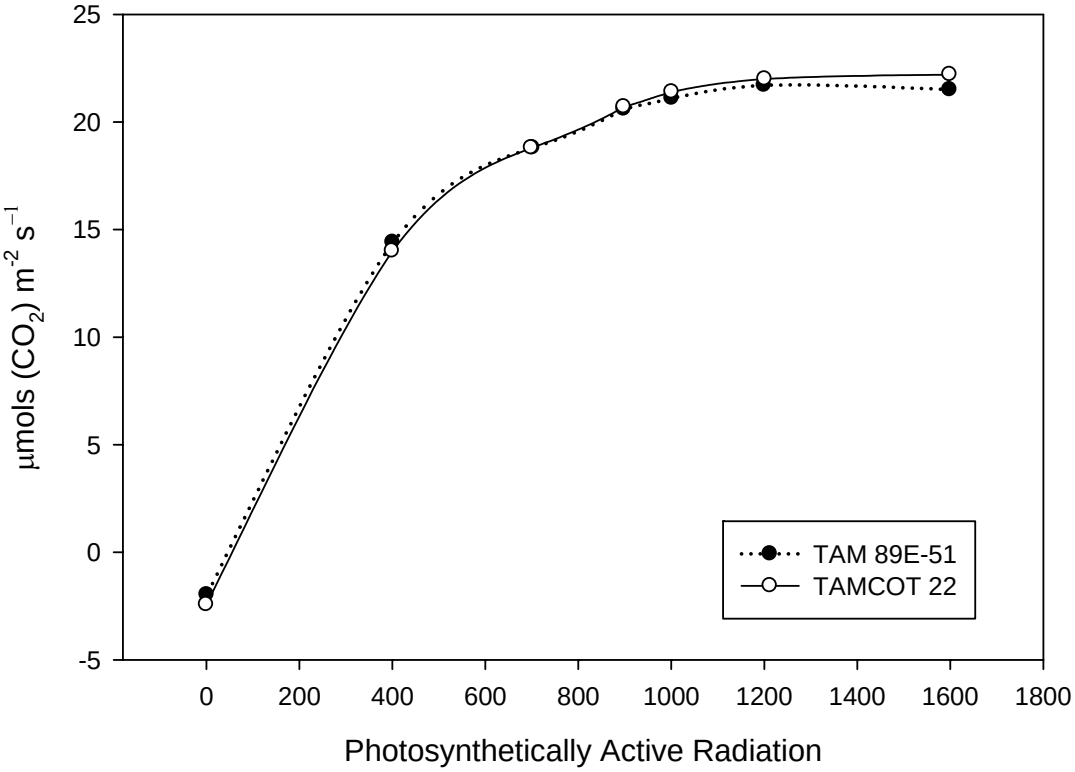

Fig. 1. Light response curves for two cotton genotypes (TAM 89E-51 and TAMCOT 22). CO_2 assimilation rate (µmols m^{-2} s^{-1}) values are the mean of six replications.

PS II (F_v/F_m) are two parameters collected that were of interest. The quantum yield of PS II was determined by means of the ratio of variable (F_v) to maximum chlorophyll fluorescence (F_m), which is defined as the efficiency of excitation capture by open PS II centers (Araus et al., 1998).

BIOMASS PRODUCTION AND SEEDCOTTON YIELD

Ten days after the recovery period of each experiment, six randomly selected water stressed and control plants were harvested for shoot material. Measurements included total leaf area and weight of stems, leaves, and fruit. Dry mass was determined following drying at 75 °C for 72 h.

In both experiments an additional six water stressed and control plants per genotype were grown to maturity. At full maturity, these plants were harvested for seedcotton and the sum weight of all bolls on each plant was calculated as seedcotton yield.

STATISTICAL ANALYSIS

The statistical analysis system computer software was used for analysis of all data (SAS®; version 8.02). Data were combined over experiments where permissible. All means were tested with analysis of variance using the General Linear Model (GLM), and separated using Fisher's Protected Least Significant Difference (LSD) at a significance of 10% (SAS, 1999-2001). Pearson's correlation coefficients were determined for gas exchange measurements using PROC CORR in SAS. All graphical displays of data in this document were produced with SigmaPlot® 2008 (version 10.0) software.

CHAPTER III

RESULTS AND DISCUSSION

EXPERIMENT I

Significant correlations were present between all parameters measured using the LI-COR 6400 ($P < 0.001$) (Table 1). The greatest correlation existed between E and g_s (r = .8691). The highest correlation with A was E (r = 0.852) followed by g_s (r = 0.734) and C_i (r = 0.663).

Leaf Gas Exchange Measurements

Pettigrew and Meredith (1994) reported differences between well-watered genotypes for A. Our data also showed differences for A between the two selected genotypes when well-watered. Therefore, data are represented as a percentage of the respective well-watered control. In both genotypes A significantly declined with $\psi_L \cong$ -1.3 MPa ($P < 0.001$) (Fig. 2). No differences were observed between genotypes for A at any ψ_L ($P = 0.517$). At a ψ_L = -3.3 MPa, both genotypes showed photosynthetic rates that were less than 30% of their respective well-watered control. However, A significantly increased in both genotypes upon re-watering ($P < 0.001$) (Fig. 3). The genotypes responded similarly to re-watering and showed an increase in A above 80% of the well-watered control by -2.5 MPa, and 100% by -1.7 MPa.

Stomatal conductance was significantly reduced almost immediately after water was withheld in both genotypes ($P < 0.001$) (Fig. 4). TAMCOT 22 dropped to less than 20% of its well-watered control at ψ_L = -1.6 MPa, whereas TAM 89E-51 fell below 20% at -2.0 MPa. However, there were no significant differences ($P = 0.972$) between the two

Table 1. Pearson correlation coefficients (r) for LI-6400 gas exchange variables using two cotton genotypes (TAM 89E-51 and TAMCOT 22) in the greenhouse, N=630.

	Stomatal Conductance	Internal CO_2 Concentration	Transpiration Rate
Net Photosynthetic Rate	0.7344***	0.6633***	0.8520***
Stomatal Conductance	----------	.7344***	0.8691***
Internal CO_2 Concentration	----------	----------	0.6990***

*** Significant at the .001 probability level.

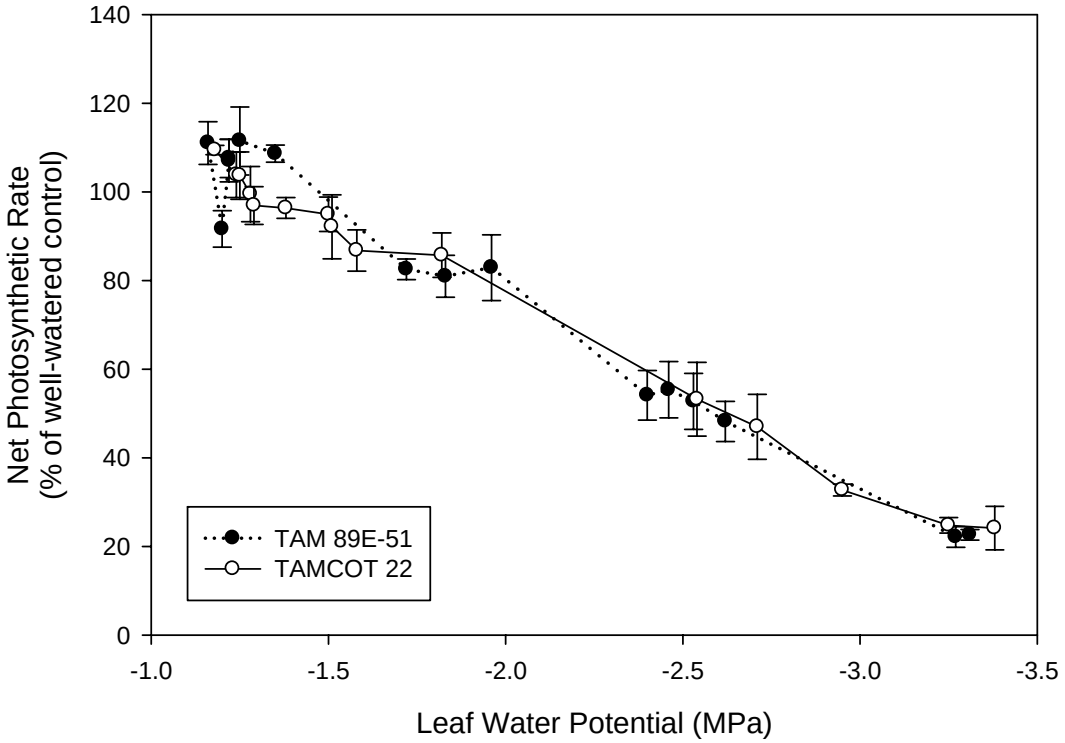

Fig. 2. Net photosynthetic rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a dry-down period (water withheld for 14 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

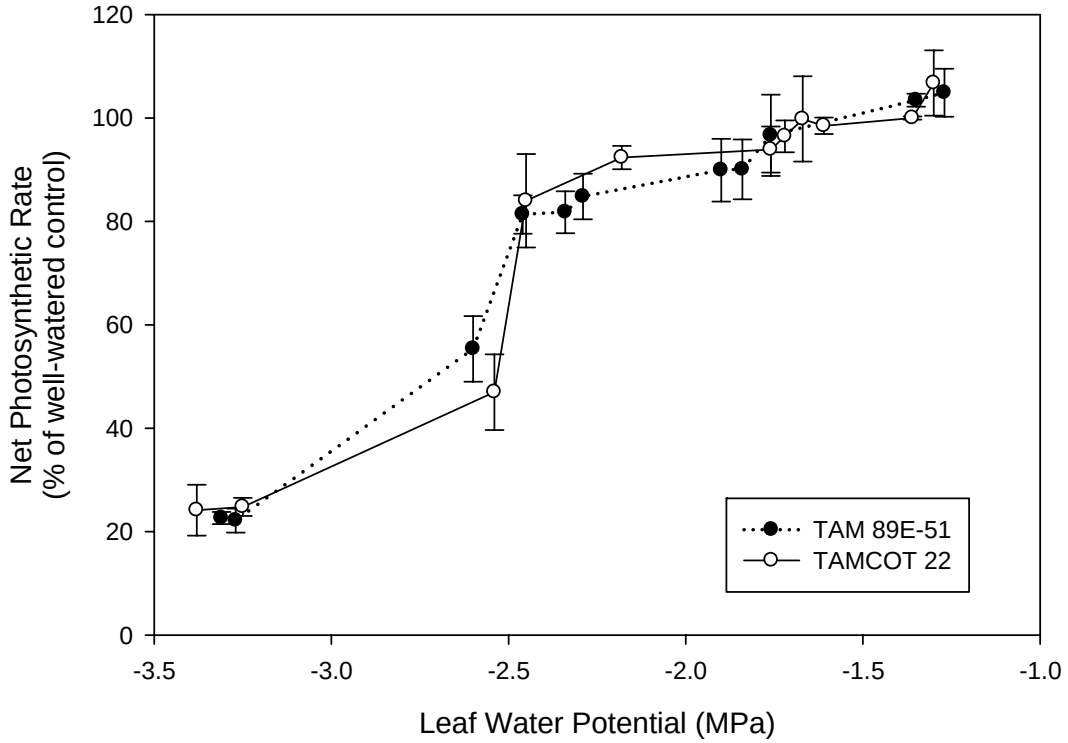

Fig. 3. Net photosynthetic rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a recovery period (sufficient water for 10 d) that began after water stress at node 4. Values are the mean ± SE of the mean for six replications.

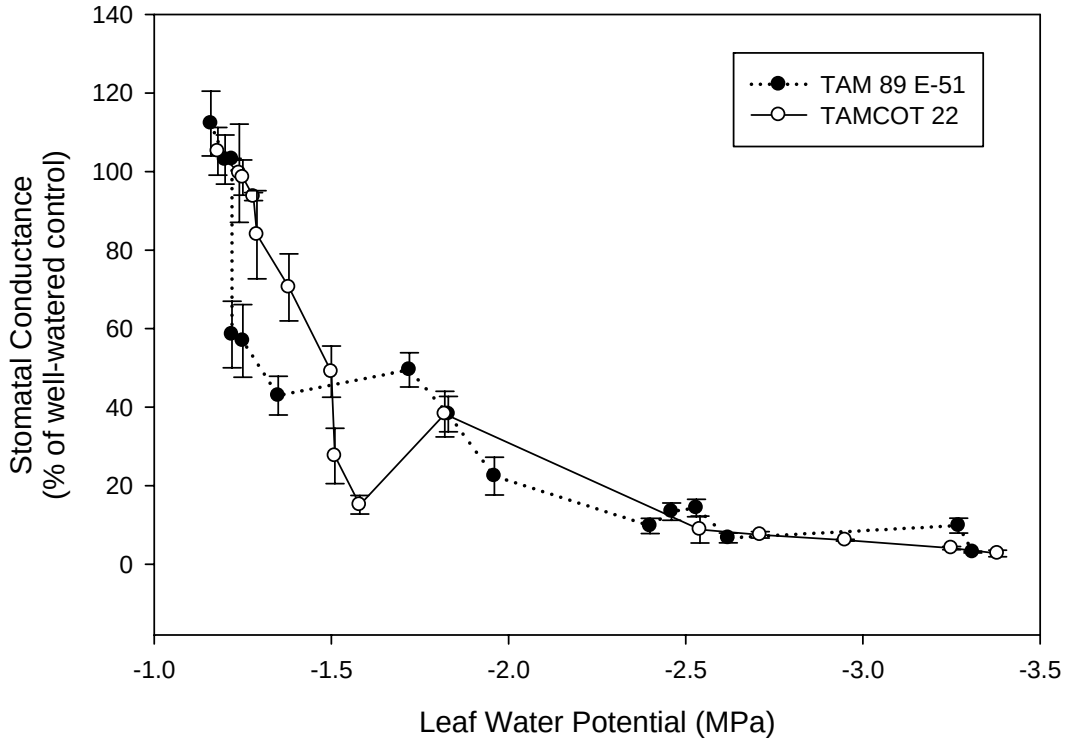

Fig. 4. Stomatal conductance (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a dry-down period (water withheld for 14 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

genotypes. Upon re-watering, neither genotype reached g_s levels above 20% of the well-watered control plants until ψ_L were >-2.5 (Fig. 5). Once ψ_L had reached -1.3 MPa, TAMCOT 22 plants showed 100% recovery compared to the well-watered control; however, even at ψ_L indicating rehydration, the TAM 89E-51 plants did not reached the g_s levels of the well-watered control plants.

Affirming the high correlation between g_s and E shown in Table 1, the significant decrease in E due to decreasing ψ_L mirrored that of g_s ($P < 0.001$) (Fig. 6). During the recovery period, both genotypes significantly increased to levels similar to the well-watered controls at a ψ_L near -1.4 MPa ($P < 0.001$) (Fig. 7).

Transpiration ratio (calculated as A:E) significantly increased in both genotypes as ψ_L decreased ($P < 0.001$) (Fig. 8). In both genotypes, A:E was in excess of 5 μmol (CO_2) mol^{-1} (H_2O) at ψ_L > -2.5 MPa; however, no differences were observed between the two genotypes.

Chlorophyll Fluorescence

The steady state value of fluorescence (F_t) did not significantly differ across changes in leaf water potential ($P = 0.990$) (Fig. 9). The F_t values for TAM 89E-51 were more erratic across a range of ψ_L levels than for TAMCOT 22. Values of F_t for TAM 89E-51 ranged from 85% of the well-watered control at -1.2 MPa to 125% at -3.4 MPa.

Quantum yield values for TAMCOT 22 ranged from 92% to 108% of the well-watered control across the ψ_L levels examined (Fig. 10). The array of quantum yield values for TAM 89E-51 was more broad than TAMCOT 22 and ranged from 86% to 115% of the well-watered control. No significant differences were identified for

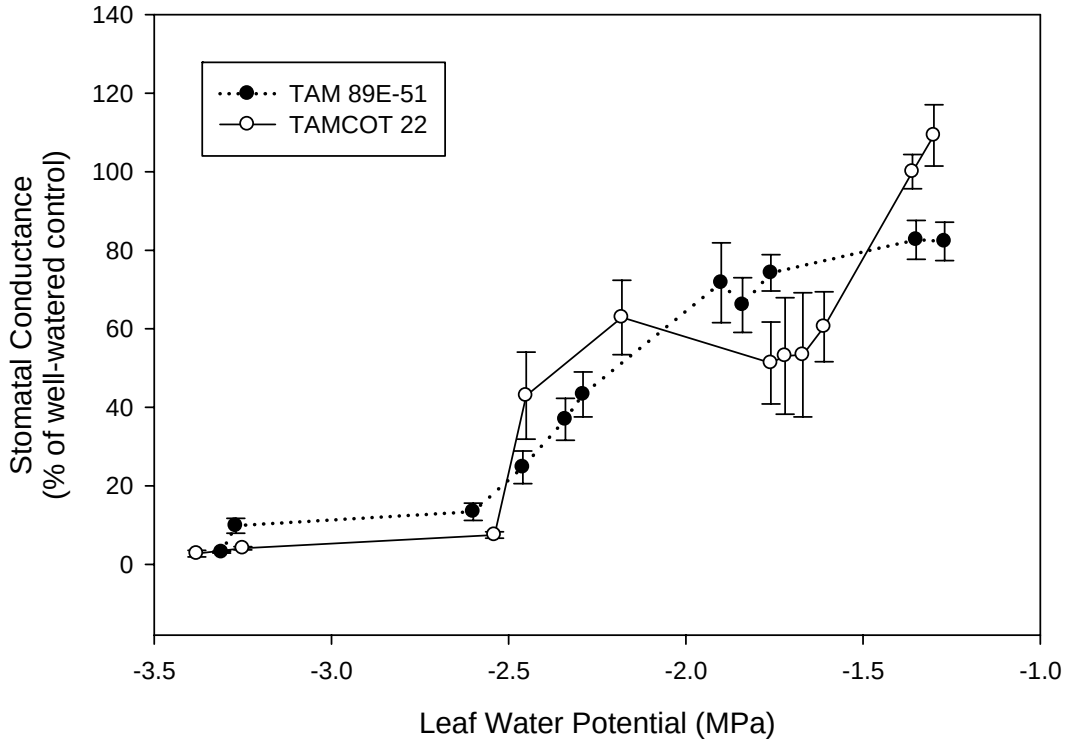

Fig. 5. Stomatal conductance (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a recovery period (sufficient water for 10 d) that began after water stress at node 4. Values are the mean ± SE of the mean for six replications.

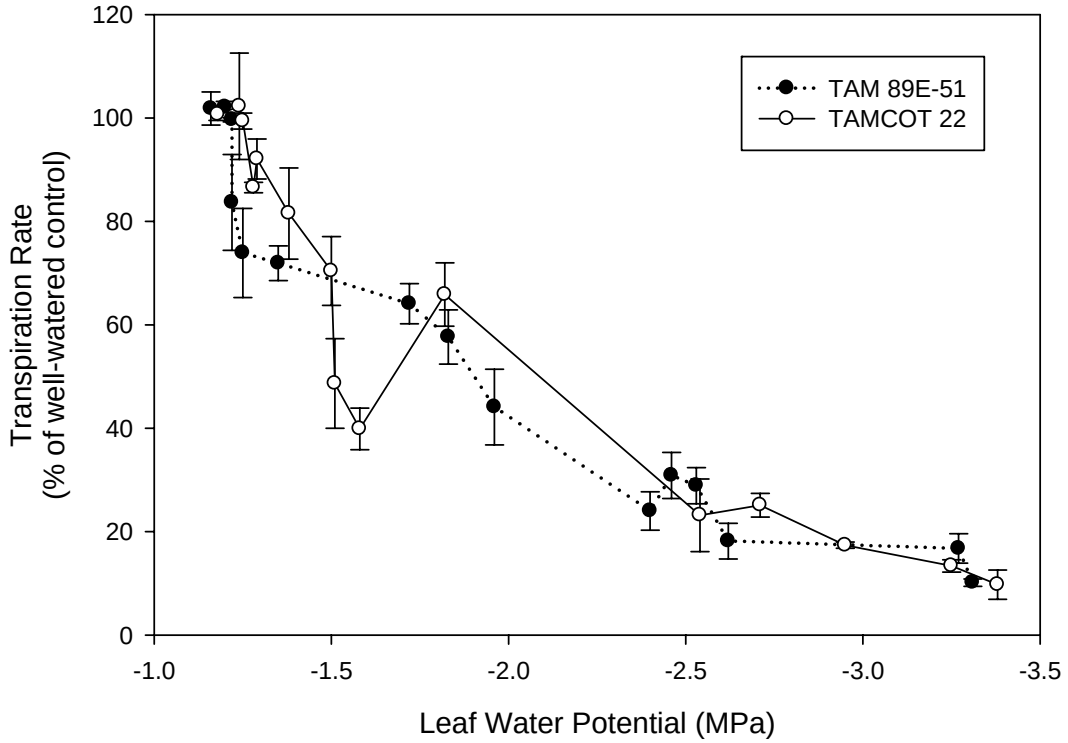

Fig. 6. Transpiration rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a dry-down period (water withheld for 14 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

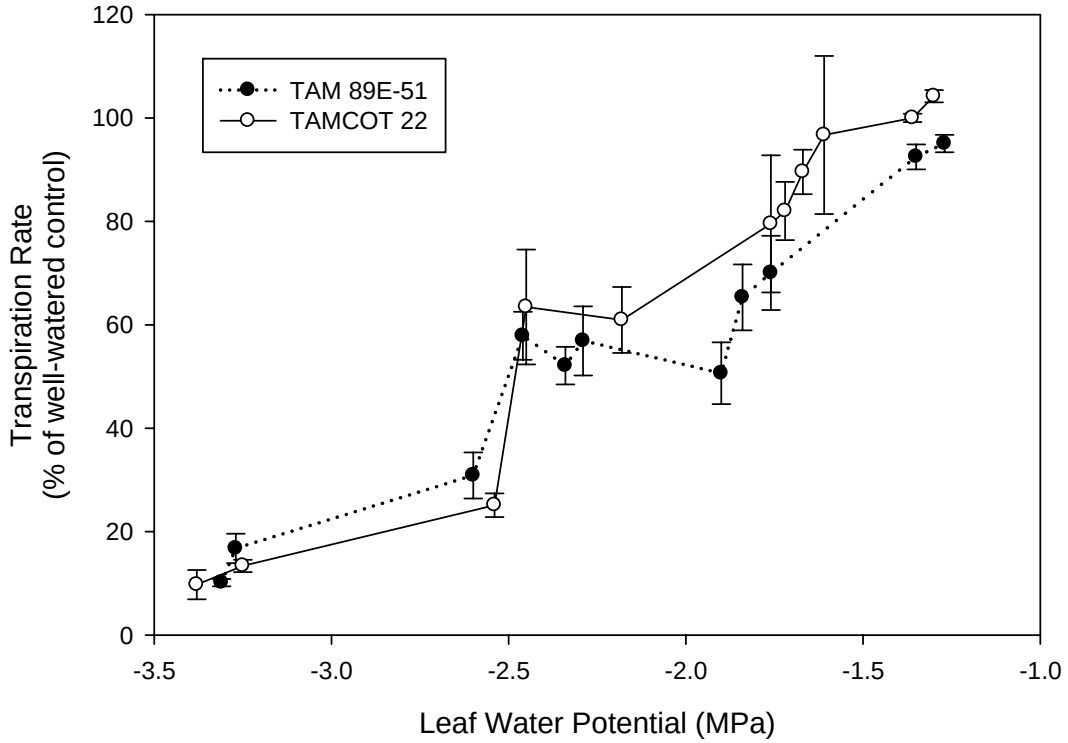

Fig. 7. Transpiration rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a recovery period (sufficient water for 10 d) that began after water stress at node 4. Values are the mean ± SE of the mean for six replications.

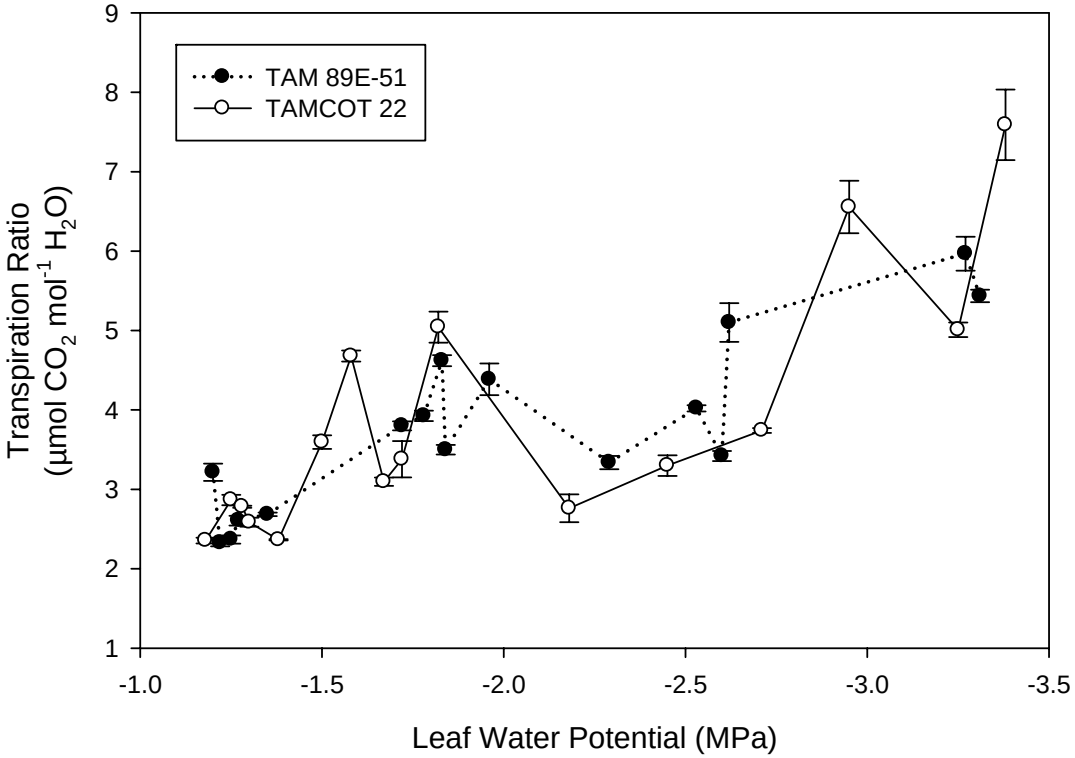

Fig. 8. Transpiration ratio (A:E) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

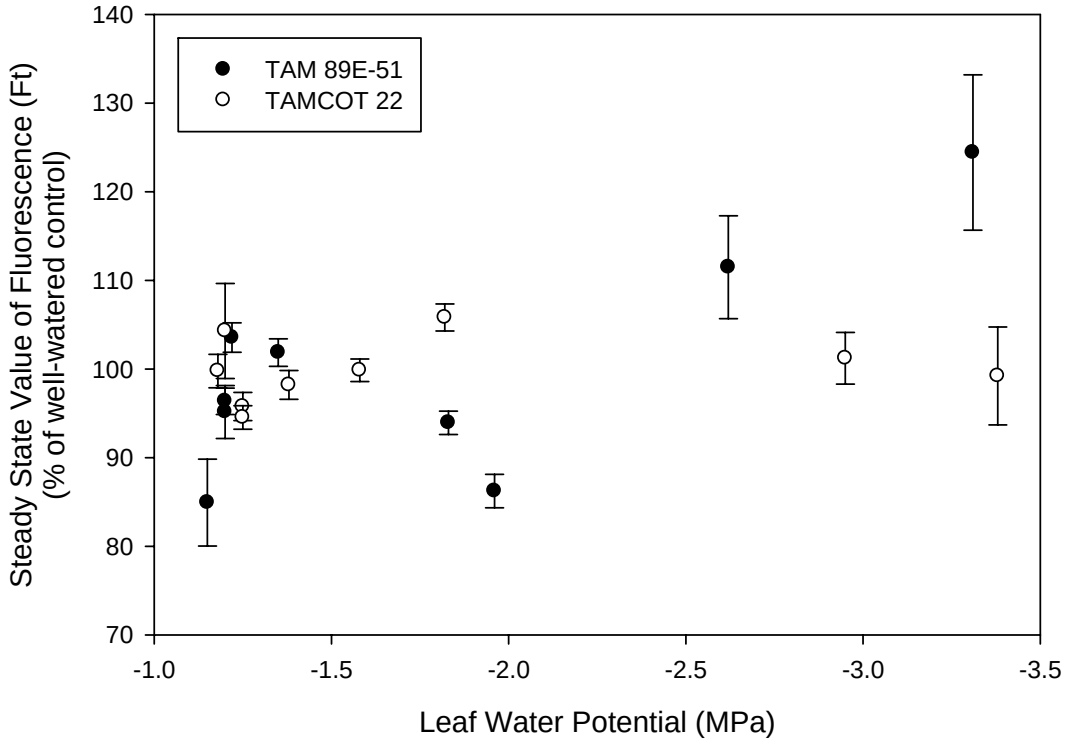

Fig. 9. Steady state value of fluorescence (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

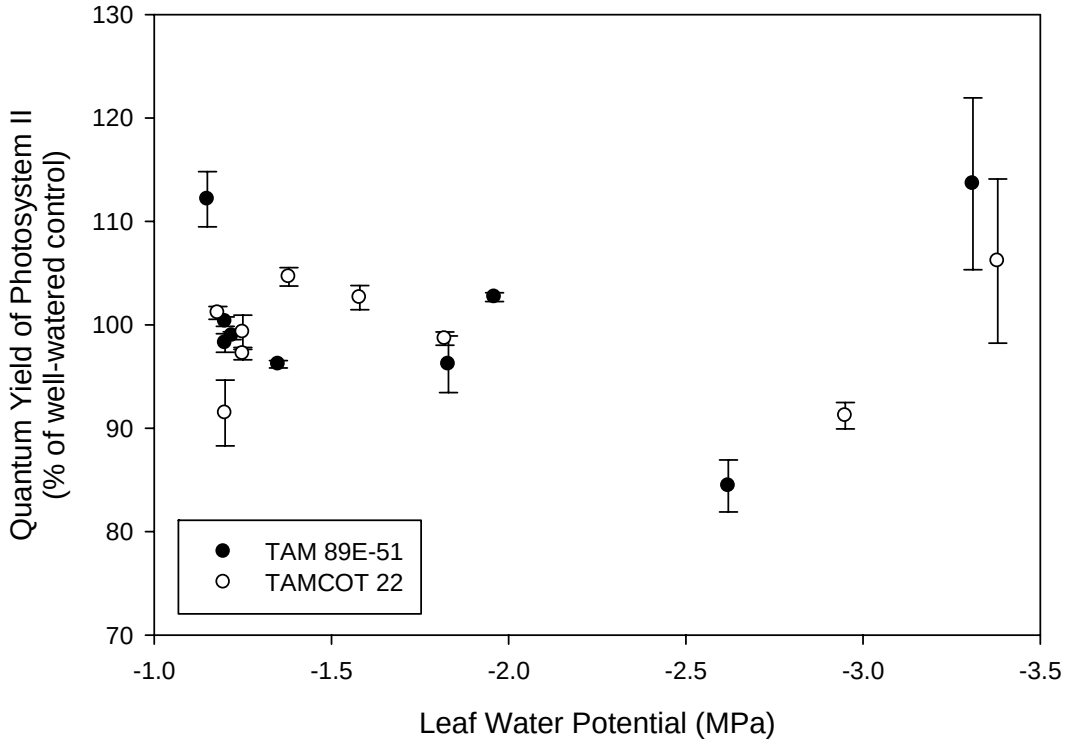

Fig. 10. Quantum yield of photosystem II (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

quantum yield as ψ_L decreased ($P = 0.991$).

No pattern of consistency could be drawn from measuring photochemical quenching (qP) (Fig. 11) or non-photochemical quenching (qNP) (Fig. 12). Significance values for both qP ($P = 0.998$) and qN ($P = 0.997$) were highly insignificant. Decreasing ψ_L appeared to have no response on these two parameters.

Figs. 13 through 15 display the results of dark-adapted fluorescence parameters. No differences were noted between genotypes for any of the dark-adapted parameters (F_o, $P = 0.710$; $F_v/F_m = 0.738$; $F_m = 0.905$).

Biomass Production and Seedcotton Yield

No statistical differences were observed between the two genotypes for leaf area or leaf dry weight (Table 2). No differences were noted between the two genotypes for stem dry weight; however, as a percent of the well-watered control plants, significant differences were seen for stem weight in favor of TAMCOT 22 ($P < 0.073$). TAM 89E-51 had a statistically greater fruit dry weight ($P < 0.005$), and a significantly greater percentage of fruit weight relative to the well-watered control ($P < 0.022$). Significant differences were observed for absolute seedcotton yield between the two genotypes in favor of TAM 89E-51 ($P < 0.029$). TAM 89E-51 had a statistically higher percentage of seedcotton yield relative to the well-watered control than did TAMCOT 22 ($P < 0.017$).

EXPERIMENT II

Leaf Gas Exchange Measurements

Both the dry-down period (Fig. 16) and recovery period (Fig. 17) indicated similar responses ($P = 0.920$ and $P = 0.908$, respectively) between the two genotypes

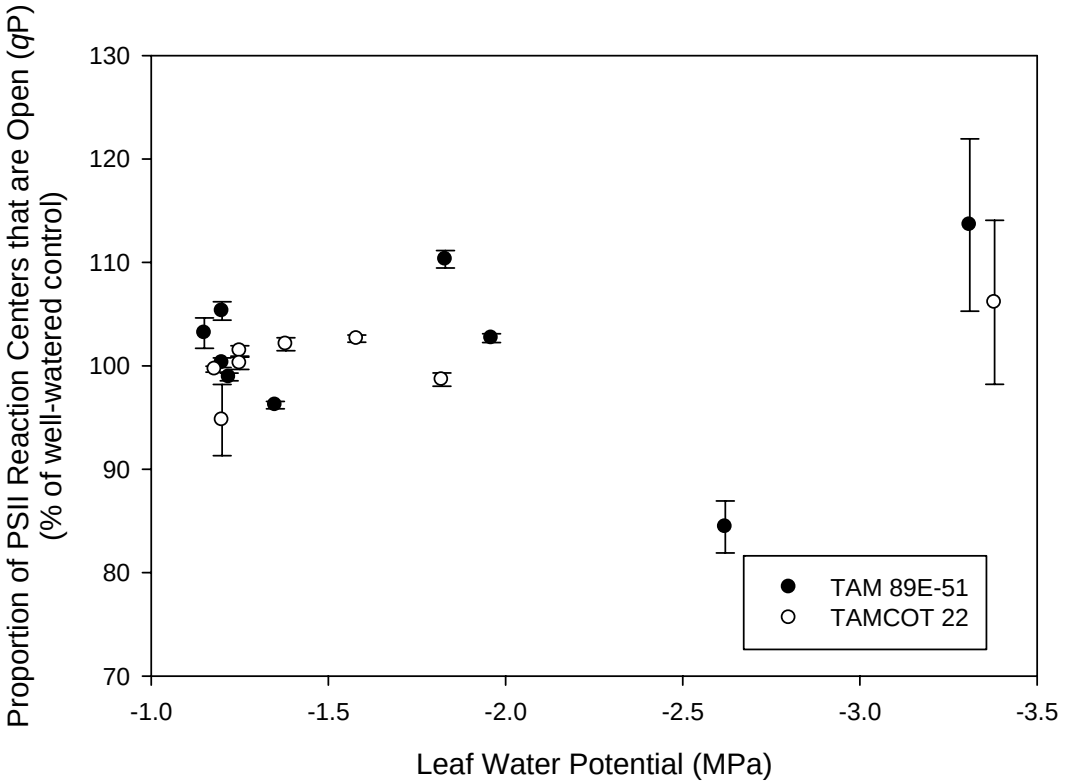

Fig. 11. Proportion of photosystem II reaction centers that are open (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

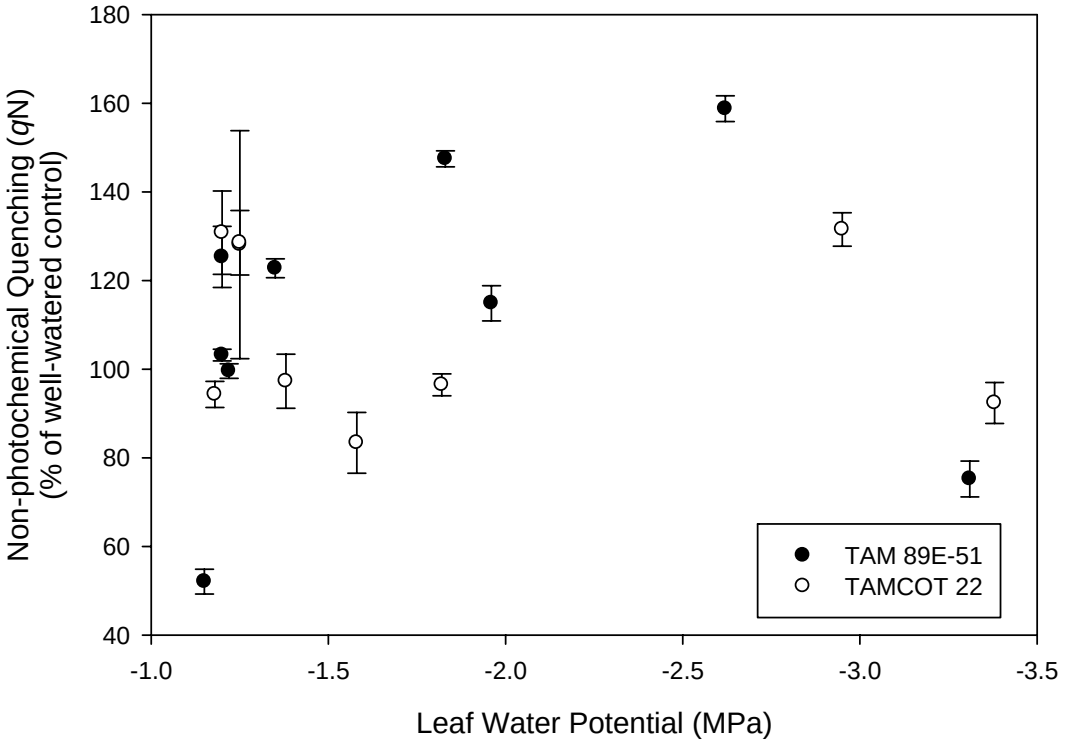

Fig. 12. Non-photochemical quenching (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

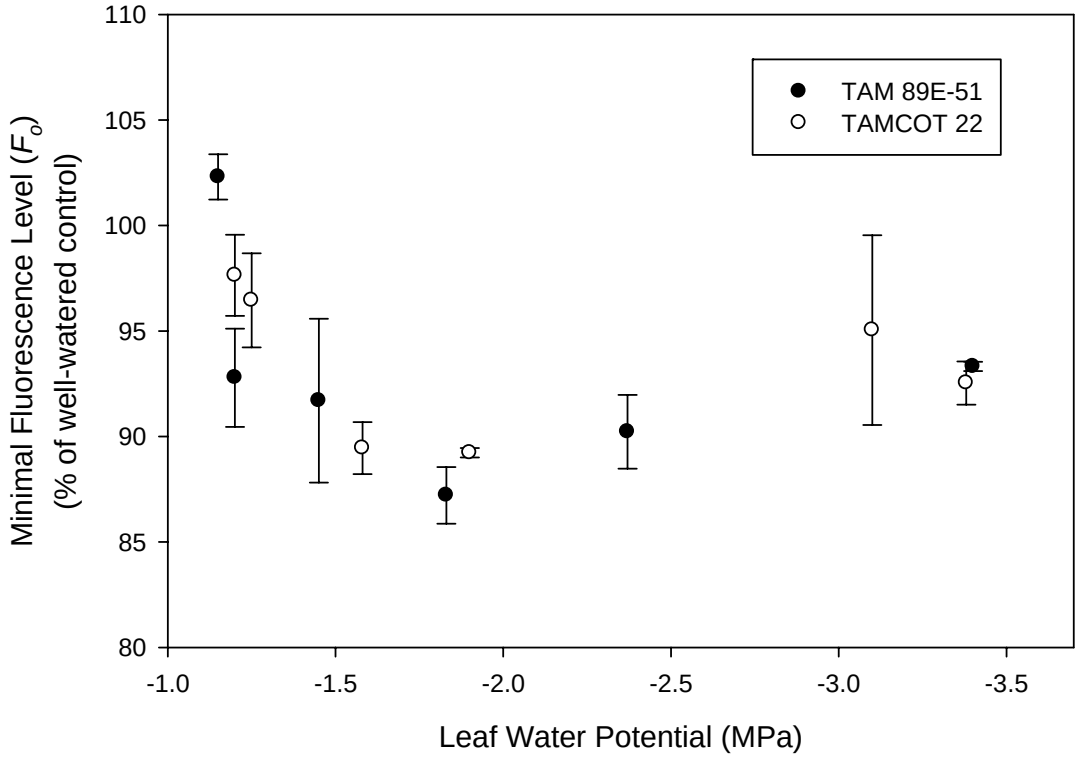

Fig. 13. Minimal fluorescence level (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

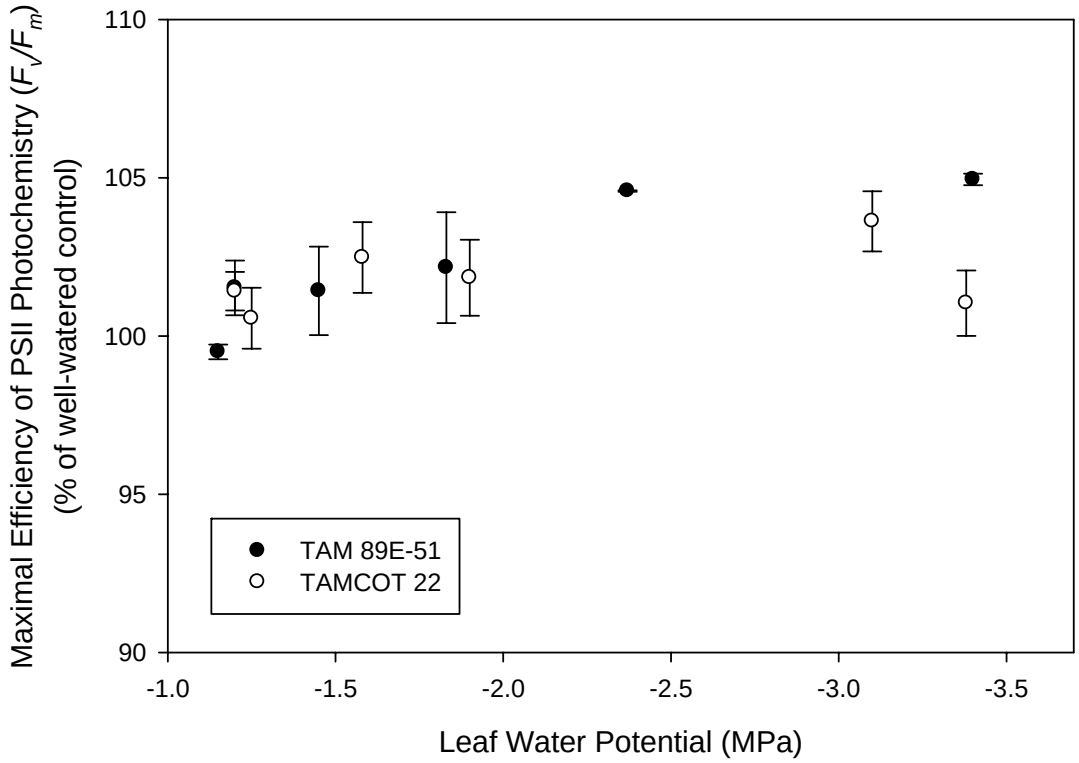

Fig. 14. Maximal efficiency of photosystem II photochemistry (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

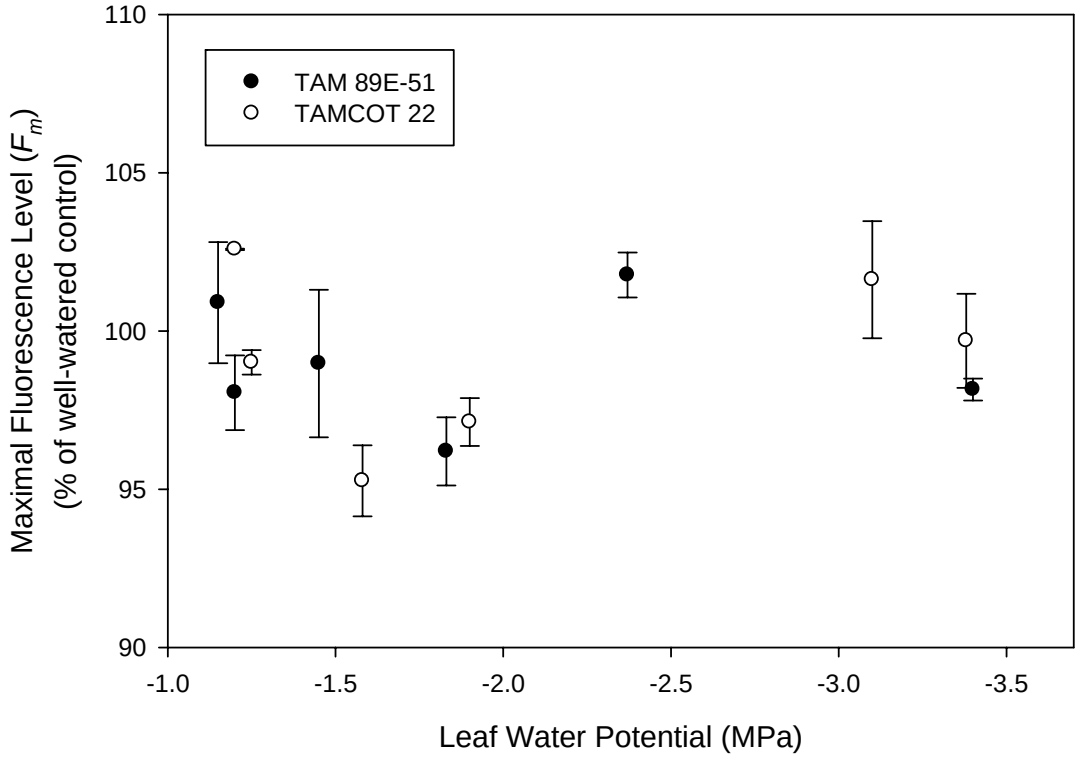

Fig. 15. Maximal fluorescence level (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 14 d) and recovery (sufficient water for 10 d) that began at node 4. Values are the mean ± SE of the mean for six replications.

Table 2. Pairwise comparisons for Experiment I (water-stressed at node 4) between two cotton genotypes (TAM 89E-51 and TAMCOT 22) for biomass partitioning components (dry weights) and seedcotton. Values represent the mean of six replications.

Experiment 1										
	Leaf Area		Leaf		Stem		Fruit		Seedcotton	
	cm^2	% of control	grams	% of control	grams	% of control	grams	% of control	grams	% of control
TAM 89E-51	5626.0	51.0	37.2	33.6	16.6	27.9	4.8	69.5	161.9	99.6
TAMCOT 22	5883.7	52.6	42.2	40.9	16.1	33.7	1.4	28.0	148.4	90.4
p-value†	.4444	.5880	.2766	.1050	.7138	*.0730*	*.0055*	*.0220*	*.0291*	*.0169*

† Significance level of test. Values in italics are significant at $P \leq 0.10$.

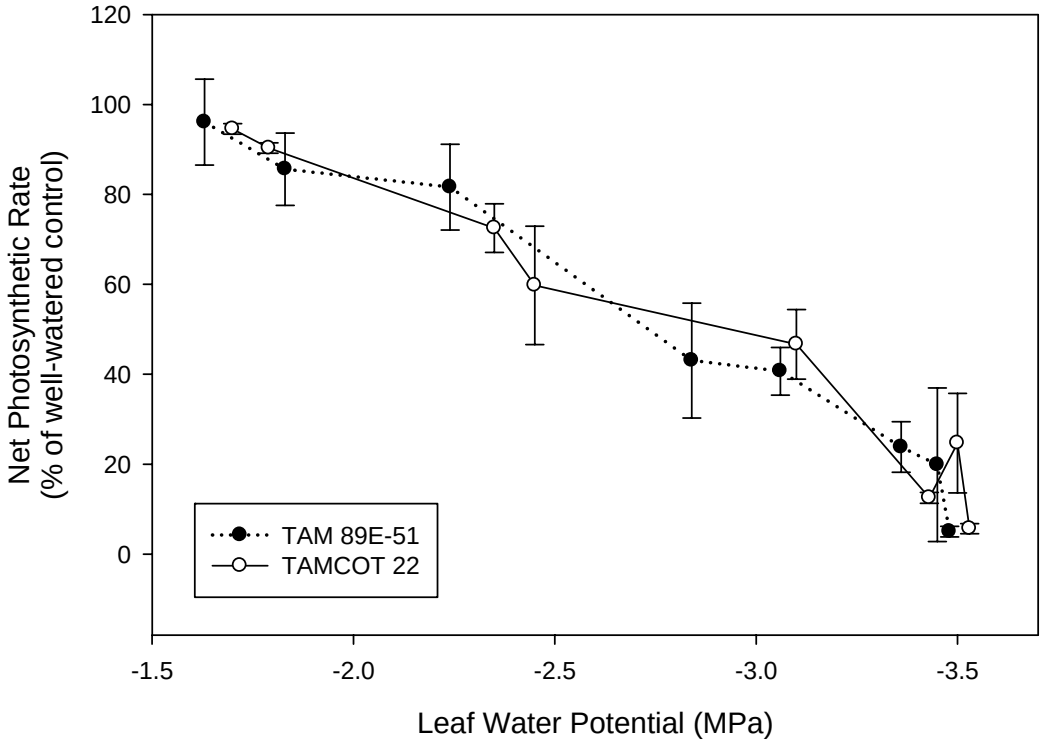

Fig. 16. Net photosynthetic rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a dry-down period (water withheld for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

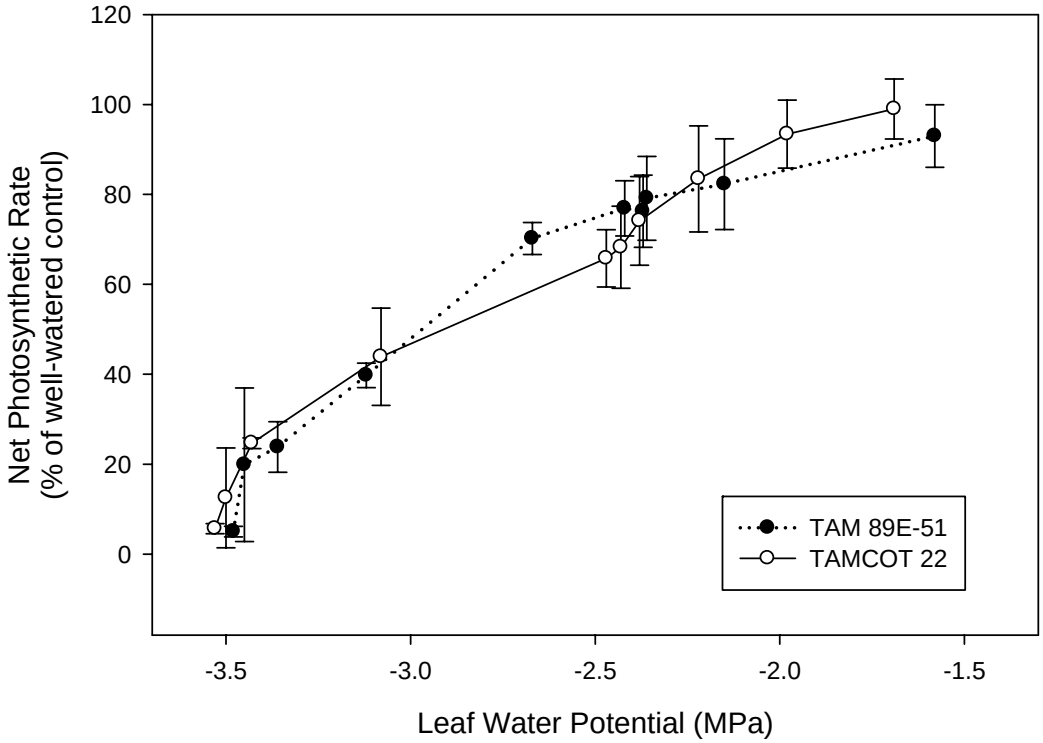

Fig. 17. Net photosynthetic rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a recovery period (sufficient water for 10 d) that began after water stress at early bloom. Values are the mean ± SE of the mean for six replications.

with change in ψ_L for A. CO_2 assimilation rate for both genotypes fell below 20% of the well-watered control plants at -3.4 MPa during the dry-down period. Each genotype reached full recovery relative to the well-watered control plants near -1.6 MPa.

Following the onset of water withholding, g_s of both genotypes was significantly reduced to levels lower than 20% of the well-watered control near -2.5 MPa ($P < 0.001$) (Fig. 18). Upon re-watering, g_s of each genotype was similar up to -2.2 MPa ($P = 0.193$) (Fig. 19). At ψ_L > -2.2 MPa during the recovery period, TAMCOT 22 continued to steadily increase g_s to levels surpassing the well-watered control. However, at ψ_L > -2.2 MPa, TAM 89E-51 began to level off reaching only 78% of the well-watered control. As in Experiment I, E mirrored the response of g_s as the ψ_L decreased in both genotypes (Fig. 20). During the recovery period, the response of the genotypes was almost parallel for E, and both genotypes recovered to levels near 100% of the well-watered controls by -1.6 MPa (Fig. 21).

The A:E in Experiment II was similar to Experiment I (Fig. 22). As leaf water potential decreased, A:E significantly ($P < 0.001$) increased in both genotypes. The A:E at ψ_L near -3.5 MPa was higher for both genotypes in Experiment II; however, there were no significant differences between the genotypes for A:E ($P = 0.480$).

Chlorophyll Fluorescence

Although no significant differences were observed, F_t increased with decreasing ψ_L for TAM 89E-51 ($P = 0.785$) (Fig. 23). However, the response, in terms of F_t, to changes in ψ_L had no effect on F_t for TAMCOT 22.

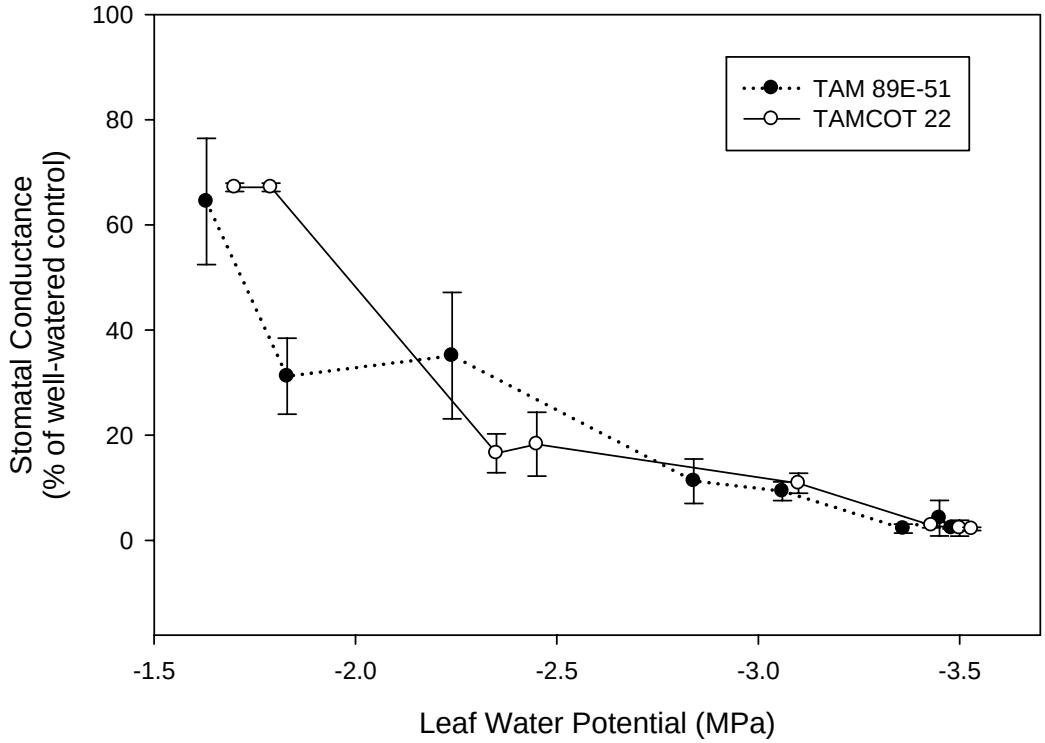

Fig. 18. Stomatal conductance (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a dry-down period (water withheld for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

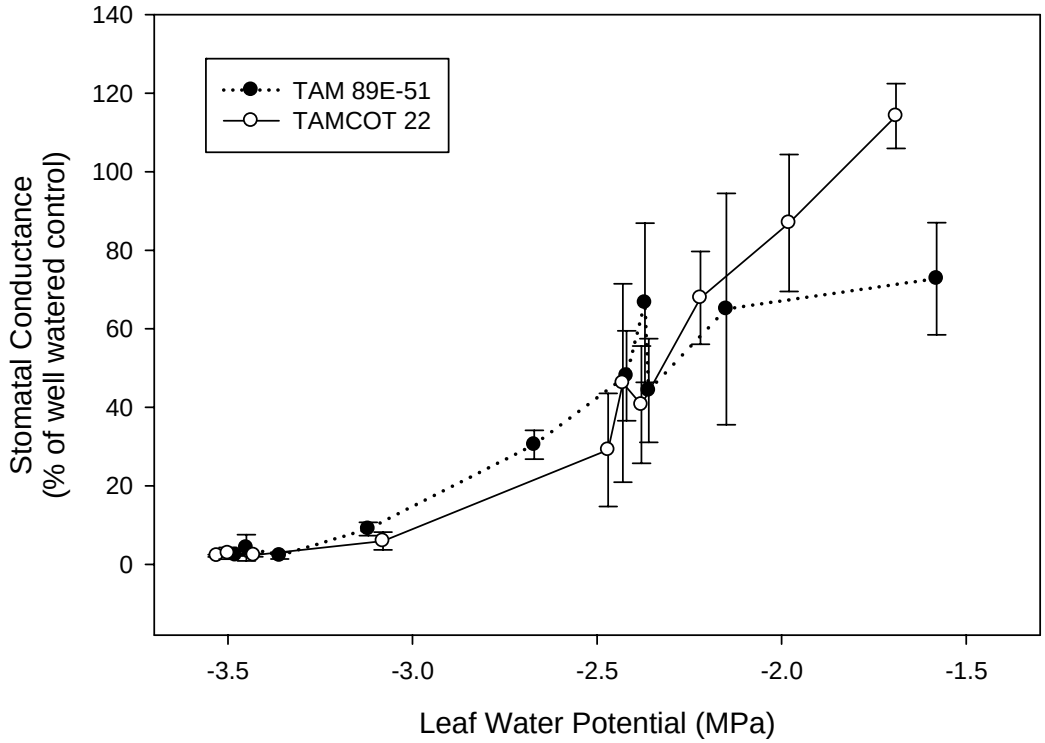

Fig. 19. Stomatal conductance (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a recovery period (sufficient water for 10 d) that began after water stress at early bloom. Values are the mean ± SE of the mean for six replications.

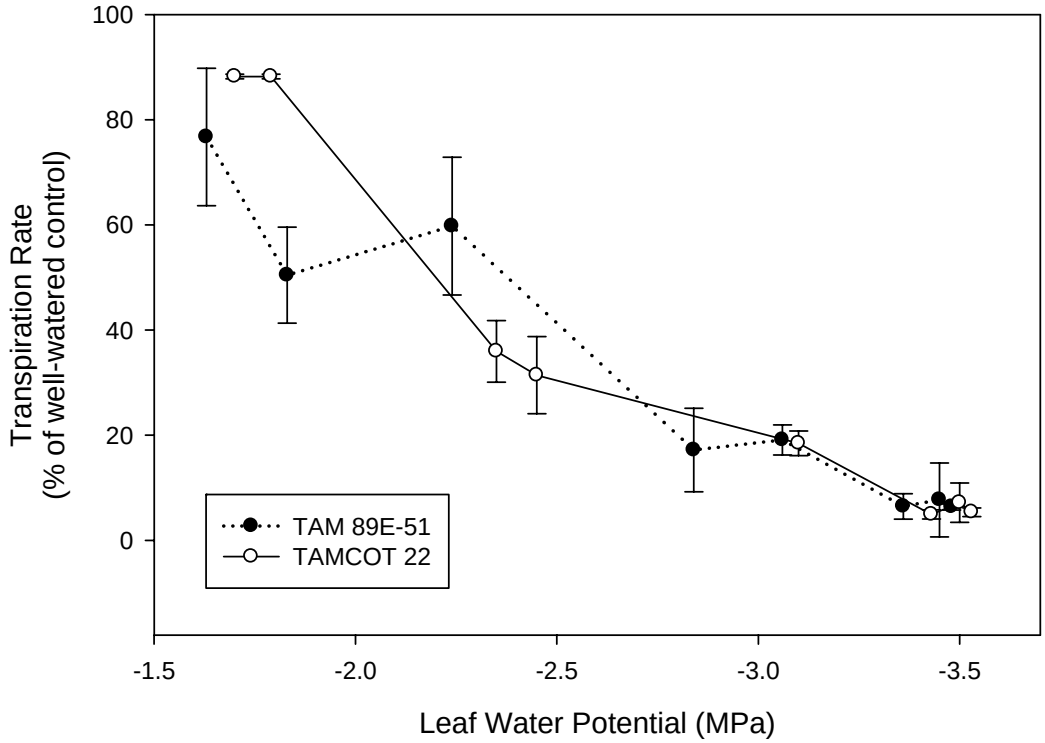

Fig. 20. Transpiration rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a dry-down period (water withheld for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

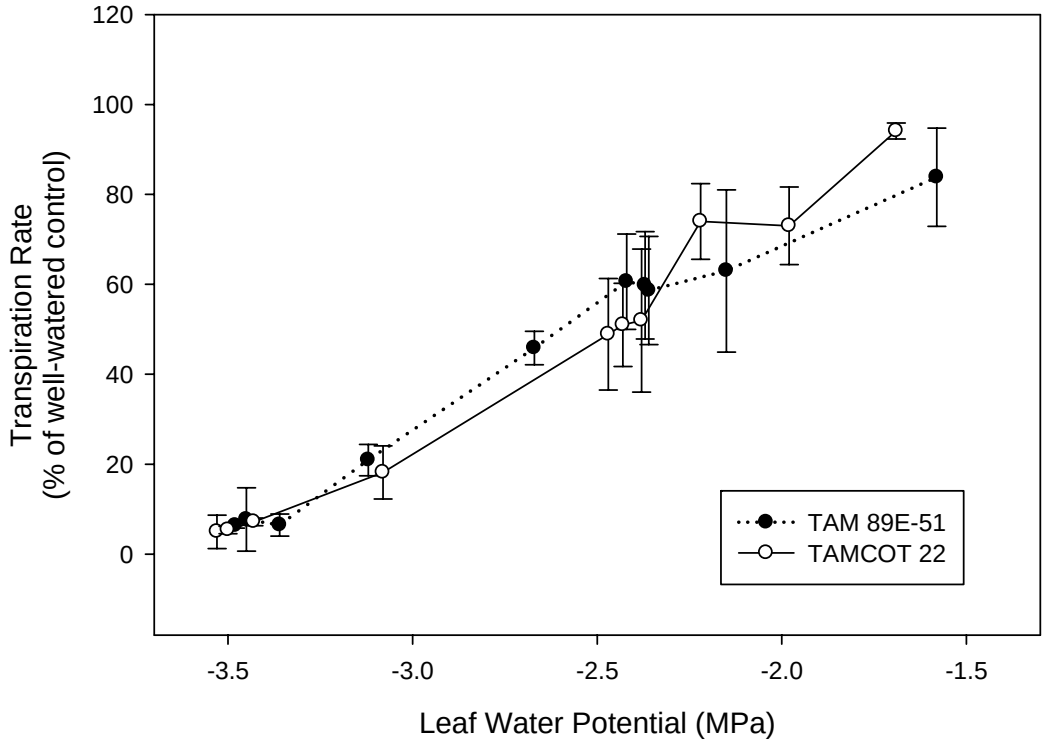

Fig. 21. Transpiration rate (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during a recovery period (sufficient water for 10 d) that began after water stress at early bloom. Values are the mean ± SE of the mean for six replications.

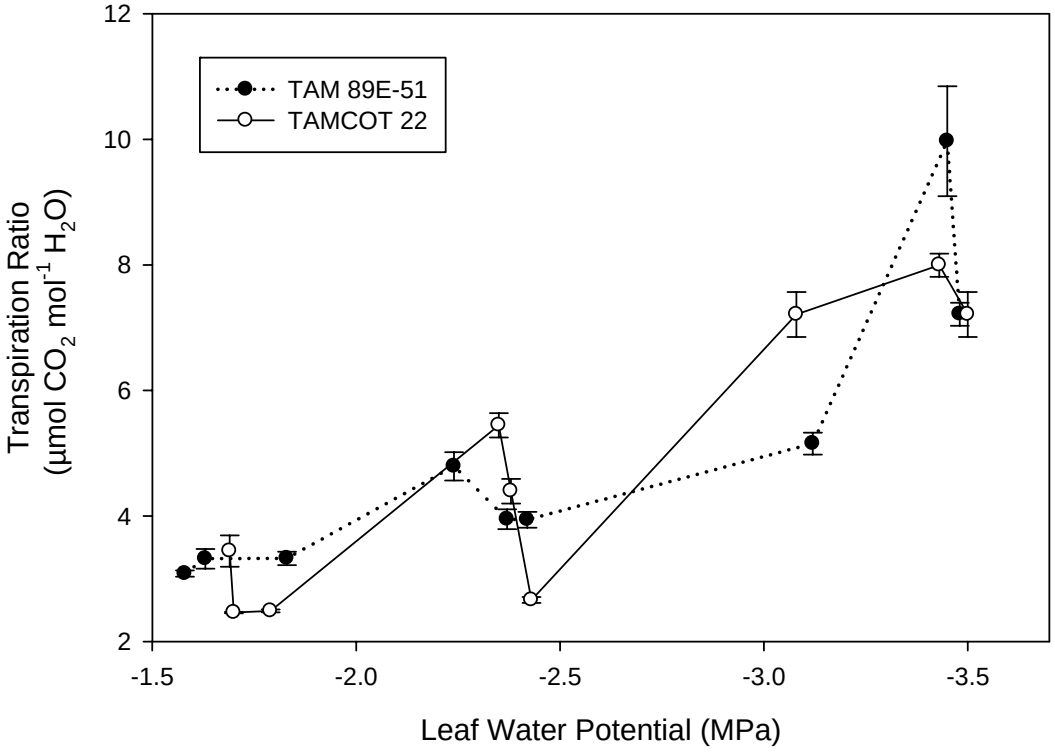

Fig. 22. Transpiration ratio (A:E) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

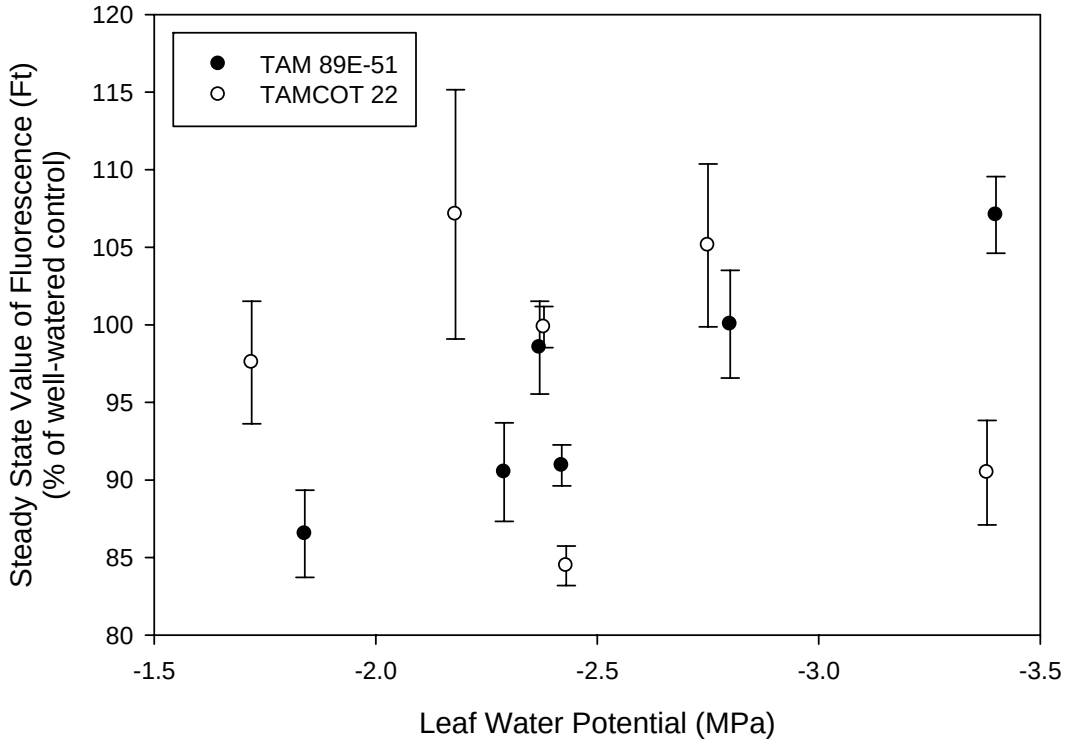

Fig. 23. Steady state value of fluorescence (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

No significant effect was observed for quantum yield as ψ_L decreased for TAMCOT 22 ($P = 0.913$) (Fig. 24). Quantum yield values ranged from 97% to 125% relative to the well-watered control plants for TAM 89E-51, and tended to decrease as ψ_L decreased. In Fig. 25, the proportion of PS II reaction centers that were open (qP) at varying ψ_L mirrored the results of quantum yield.

Despite large variation in the data with no significant differences ($P = 0.132$), both genotypes showed an increase in non-photochemical quenching (qN; Fig. 26). Similar to the results of Experiment I, there were no significant differences for any of the dark-adapted parameters (F_o, $P = 0.996$; F_v/F_m, $P = 0.986$; F_m, $P = 0.984$) (Figs. 27 - 29).

Biomass Production and Seedcotton Yield

No significant differences were observed for leaf area between the two genotypes (Table 3). No differences between the two genotypes for leaf dry weight were reported; however, leaf weight expressed as a percentage of the well-watered control plants, was significantly ($P < 0.054$) greater in TAMCOT 22 than in TAM 89E-51. Stem weight, as a percentage of the well-watered control, was statistically ($P < 0.077$) greater in TAM 89E-51. The absolute fruit weight was significantly ($P < 0.001$) greater for TAM 89E-51; however, relative to the well-watered control plants, TAMCOT 22 had a significantly ($P < 0.001$) greater fruit weight. In contrast to Experiment I, absolute yield was statistically ($P < 0.016$) different in favor of TAMCOT 22. In relation to the well-watered control plants, TAMCOT 22 had a significantly ($P < 0.042$) greater yield than TAM 89E-51.

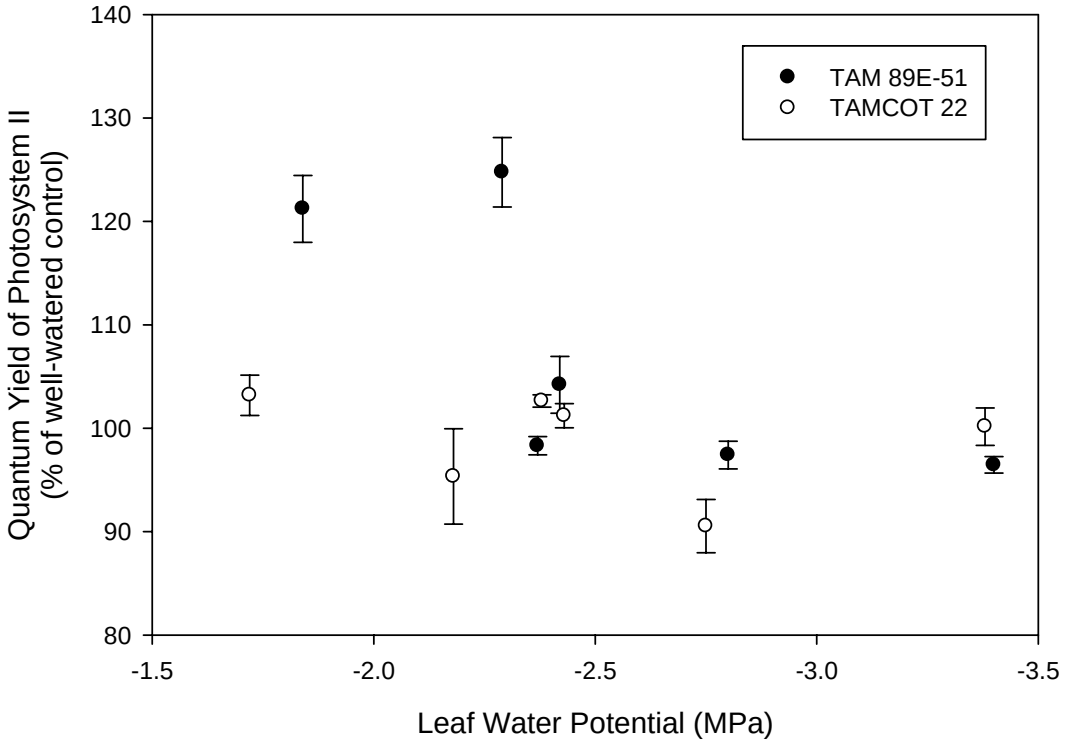

Fig. 24. Quantum yield of photosystem II (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

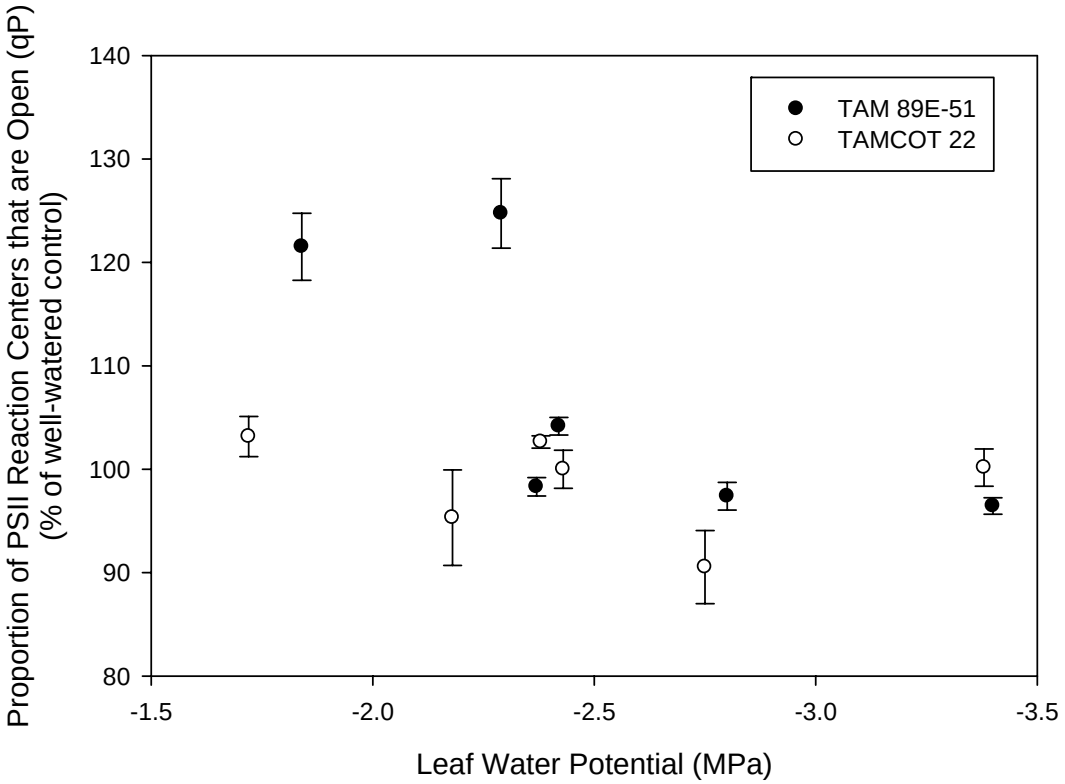

Fig. 25. Proportion of photosystem II reaction centers that are open (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

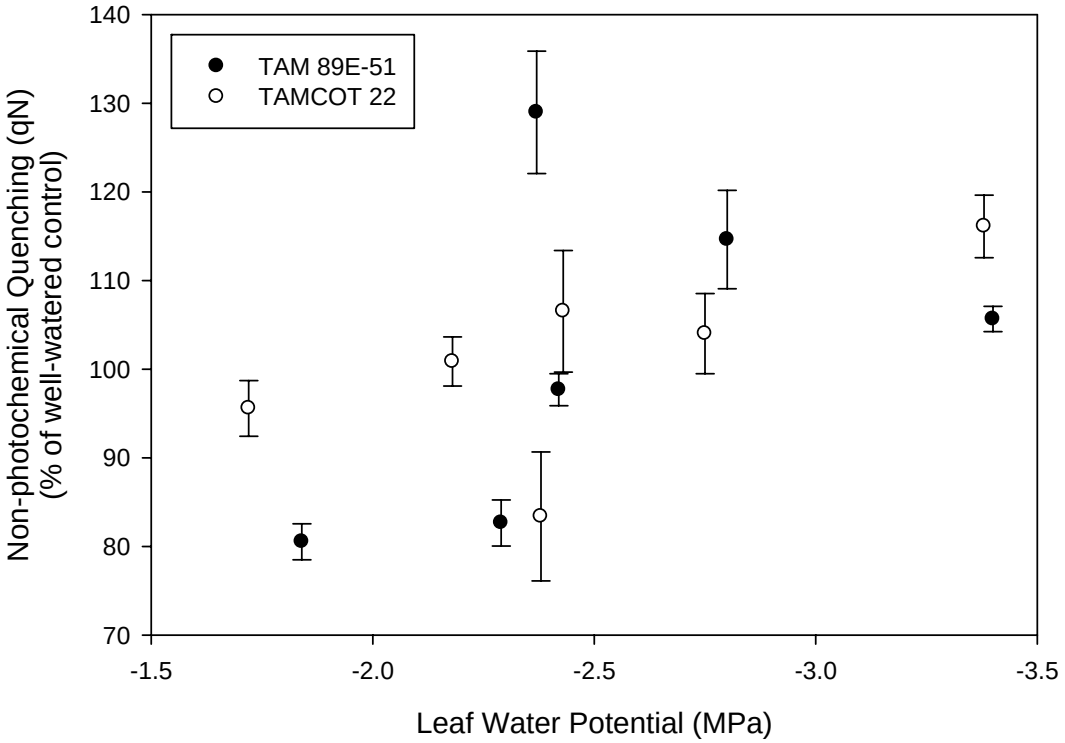

Fig. 26. Non-photochemical quenching (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

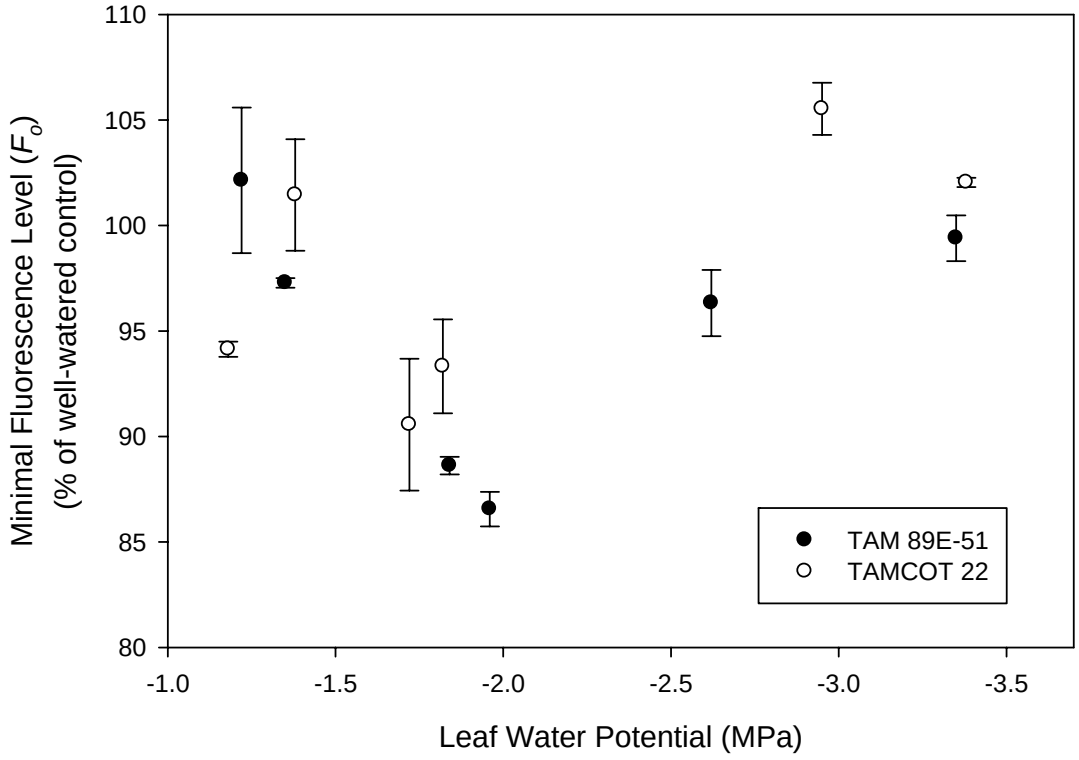

Fig. 27. Minimal fluorescence level (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

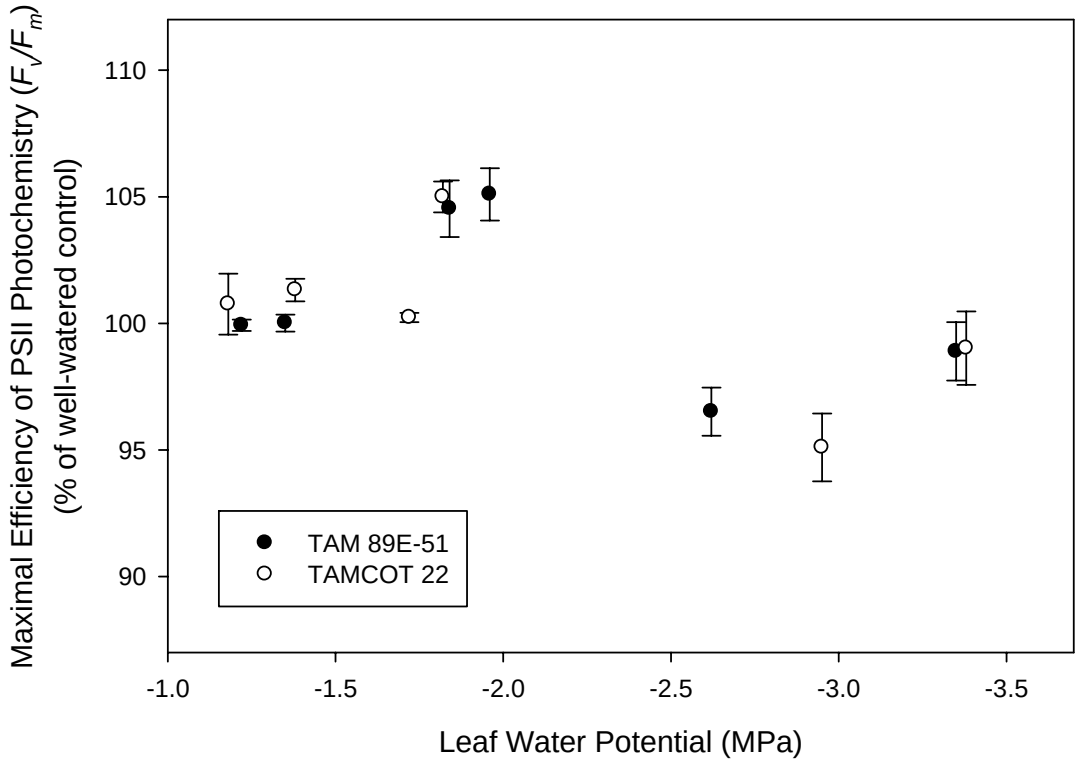

Fig. 28. Maximal efficiency of photosystem II photochemistry (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

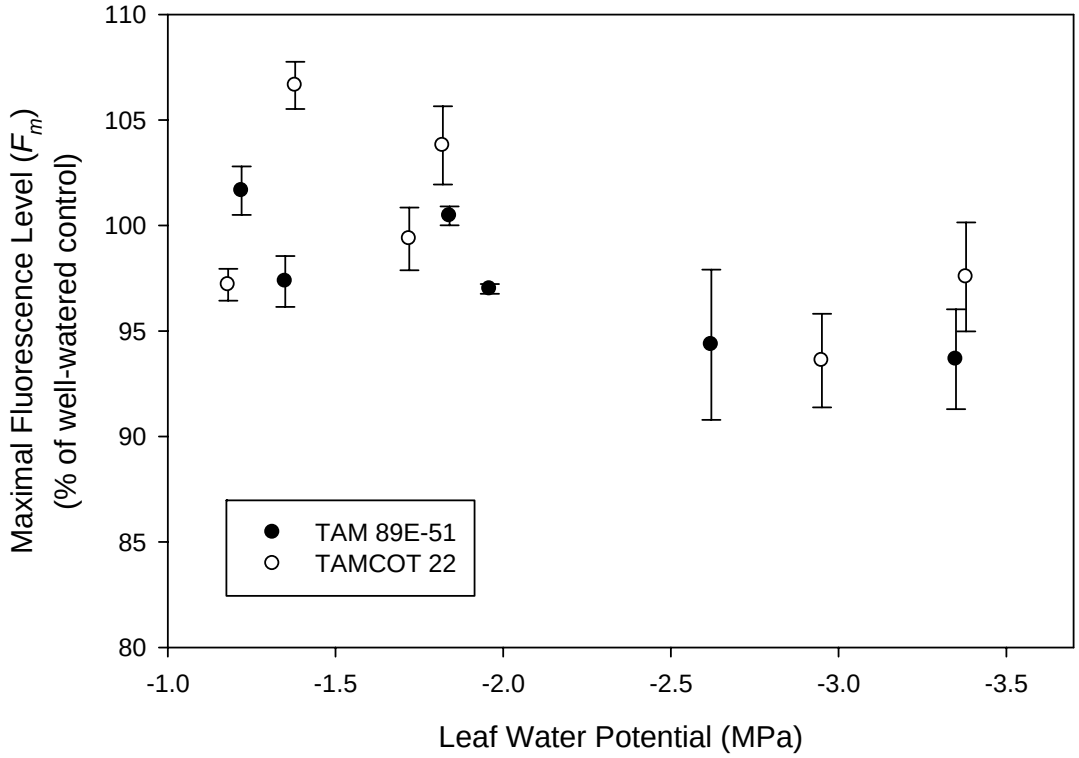

Fig. 29. Maximal fluorescence level (% of well-watered control) of two cotton genotypes (TAM 89E-51 and TAMCOT 22) during periods of dry-down (water withheld for 10 d) and recovery (sufficient water for 10 d) that began at early bloom. Values are the mean ± SE of the mean for six replications.

Table 3. Pairwise comparisons for Experiment II (water-stressed at early bloom) between two cotton genotypes (TAM 89E-51 and TAMCOT 22) for biomass partitioning components (dry weights) and seedcotton. Values represent the mean of six replications.

	Experiment 1I									
	Leaf Area		Leaf		Stem		Fruit		Seedcotton	
	cm^2	% of control	grams	% of control	grams	% of control	grams	% of control	grams	% of control
TAM 89E-51	12944	82.2	51.4	67.6	61.3	84.5	53.1	44.0	128.2	78.9
TAMCOT 22	12261	91.7	56.1	81.7	66.5	74.1	46.9	49.5	148.6	89.5
p-value†	.4521	.1202	.3309	*.0540*	.2123	*.0776*	*.0009*	*.0012*	*.0161*	*.0420*

† Significance level of test. Values in italics are significant at $P \leq 0.10$.

CHAPTER IV

DISCUSSION

LEAF GAS EXCHANGE MEASUREMENTS

The greatest correlation among gas exchange parameters existed between E and g_s. These results were similar to reports by Peng and Krieg (1992) who found high correlation (r = 0.97) between E and g_s in grain sorghum (*Sorghum bicolor* L.). The highest correlation with A was E, followed by g_s. The values were consistent with those reported by Peng and Krieg (1992) with correlation values of 0.98 and 0.81 for E and g_s, respectively, with A.

Pettigrew and Meredith (1994) noted significant differences between well-watered cotton genotypes for A. Our data indicated that measurements of A between the well-watered controls of the two genotypes selected for the present experiments were not constant over time. However, the absolute values of A for each genotype were very consistent with the ranges of A reported by Pettigrew and Meredith (1994). Absolute values of A and g_s were also consistent with those of Lu et al. (1997) in cotton.

In the present study, the response of both cotton genotypes to decreasing ψ_L concur with Brestic et al. (1995) who reported decreases in A and g_s with water stress up to 10 d after withholding water in French beans (*Phaseolus vulgaris* L.). Escalona et al. (1999) also noted a 40% reduction in A by water stress in field grown grapevines (*Vitis vinifera* L.), and Lu and Zhang (1998) found decreases in A and g_s with increasing water stress in wheat (*Triticum aestivum* L.). Our data indicated that both genotypes reached levels near 100% of the well-watered controls for A and g_s after re-watering. This

suggests minimal damage to the photosynthetic mechanism by water-stress in our experiments. Previous studies report that A is impaired mainly by g_s at moderate leaf water deficits, and only when drought was prolonged to the point of leaf dehydration, or if other stresses (heat, high light) are imposed, will electron transfer mechanisms be affected (Boyer and Bowen, 1970; Chaves, 1991; Cornic et al., 1992). Quick et al. (1992) suggested that water stress inhibits A primarily by stomatal closure. Giardi et al. (1996) agreed with the previous reports that only intense, long-term water stress can result in destruction to photochemistry mechanisms.

Peng and Krieg (1992) reported a significant correlation ($P = 0.01$) between WUE and A:E ($r^2 = 0.97$) in grain sorghum. Heitholt (1989) also reported that WUE and A:E were strongly correlated ($r^2 = 0.76$) across water-stress treatments in winter wheat. Therefore, WUE in the present study was evaluated using A:E. Our data indicated increases in A:E for both genotypes as ψ_L decreased. This finding is consistent with Lui et al. (2005) who also reported an increase in WUE of potato (*Solanum tuberosum* L.) under progressive soil drying. The authors attribute this response to A being less sensitive to water stress than g_s.

CHLOROPHYLL FLUORESCENCE MEASUREMENTS

Discrepancies in the literature on the effects of water stress on chlorophyll fluorescence parameters were due to environmental conditions. The bulk of the literature reporting significant changes in dark-adapted fluorescence parameters (i.e., F_v/F_m) under water stress were conducted under field conditions (Angelopoulos et al., 1996). In terms of dark-adapted fluorescence parameters, the results of the present experiments are very

similar to previous studies conducted under controlled environments. Lu and Zhang (1998) showed no effects on the PS II photochemistry of dark-adapted leaves, including F_v/F_m, during water stress of wheat plants. In a similar study, Pankovic et al. (1999) showed no differences in F_v/F_m among water-stressed sunflower (*Helianthus annuus* L.) plants when compared to a well-watered control.

Regarding the light-adapted fluorescence parameters (F_t, ϕ_{II}, qP, qNP), previous literature suggested that the response of plants to water stress may be species-dependent. Escalona et al. (1999) showed a 59% decrease in quantum yield of water-stressed, field-grown grapevines; however, Brestic et al. (1995) reported that water stress had no impact on the quantum yield of French bean (*Phaseolus vulgaris* L.). Our data are consistent with those of Brestic et al. (1995), with no statistical differences seen between the two genotypes or as ψ_L decreased for quantum yield, qP and qNP.

BIOMASS PRODUCTION AND SEEDCOTTON YIELD

The results of these experiments were similar to results of previous literature in terms of biomass partitioning and seedcotton yield. Pace et al. (1999) noted a 32% decrease in stem weight, up to 63% decrease in leaf area, and a 35% decrease in leaf weight of cotton plants under water stress. Pankovic et al. (1999) reported a decrease in total number of leaves of sunflower plants under water stress. Peng and Krieg (1992) reported no significant differences among water use of grain sorghum genotypes, and suggested that the primary determinant of WUE was leaf area, which reflects biomass production. No differences were seen in the present study for leaf area or leaf weight in Experiment I; however, in Experiment II TAMCOT 22 had a significantly greater leaf

weight as a percentage of the well-watered control than TAM 89E-51. This increase in leaf weight may partially explain the increase in seedcotton yield exhibited by TAMCOT 22 in Experiment II.

CHAPTER V

CONCLUSIONS

According to data generated in these experiments, single-leaf measurements of A, E, and g_s, did not adequately explain differences in WUE between the two genotypes. Light- and dark-adapted chlorophyll fluorescence parameters failed to show separation between the two genotypes, and could indicate that water stress alone does not lead to photooxidation and photoinhibition.

Biomass partitioning, taken after the recovery period, explained differences in phenotypic responses between the genotypes following a water stress event. Differences were noted for stem weight and fruit weight in both experiments. In Experiment I (water stress at node 4), TAM 89E-51 had a significantly greater yield than TAMCOT 22; however, in Experiment II (water stress at early bloom) the statistically greater yield was in favor of TAMCOT 22. Since the genotypes did not respond the same to water stress when the stress event occurred at the two different growth stages, this indicates that genotypes can respond differently to water stress depending on their maturity.

Results from these experiments suggest that the effects of water stress on cotton are a function of the intensity of the stress, as well as the growth stage in which the stress is experienced. Based on data presented in these experiments, distinguishing WUE between cotton genotypes should not be based on how the genotype responds during a water stress event at a single growth stage. Rather, the assessment should be based on the severity and duration of the water stress event over several growth stages.

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APPENDIX A

EXPERIMENT I – GAS EXCHANGE PARAMETERS

Date	Genotype	Net Photosynthetic Rate μmol (CO_2) m^{-2} s^{-1}	Stomatal Conductance mol (H_2O) m^{-2} s^{-1}	Transpiration Rate mol (H_2O) m^{-2} s^{-1}
09/06/07	TAM 89E-51	23.45	0.4345	5.69
09/08/07	TAM 89E-51	23.17	0.4447	5.75
09/13/07	TAM 89E-51	11.80	0.0913	1.97
09/15/07	TAM 89E-51	12.09	0.1089	2.32
09/18/07	TAM 89E-51	17.50	0.2370	3.61
09/20/07	TAM 89E-51	19.62	0.4057	4.17
09/22/07	TAM 89E-51	22.57	0.6727	7.61
02/23/08	TAM 89E-51	21.35	0.4054	6.99
02/26/08	TAM 89E-51	18.86	0.1828	5.02
02/28/08	TAM 89E-51	19.85	0.2168	4.36
03/02/08	TAM 89E-51	12.17	0.1050	3.03
03/05/08	TAM 89E-51	5.16	0.0277	0.93
03/07/08	TAM 89E-51	19.08	0.2498	5.65
03/09/08	TAM 89E-51	22.60	0.3368	5.87
03/12/08	TAM 89E-51	24.33	0.8170	9.38
04/25/08	TAM 89E-51	19.41	0.8653	8.38
04/30/08	TAM 89E-51	21.06	0.9410	8.88
05/02/08	TAM 89E-51	23.90	0.9420	8.90
05/07/08	TAM 89E-51	17.94	0.2050	4.36
05/09/08	TAM 89E-51	10.07	0.0611	1.60
05/11/08	TAM 89E-51	4.71	0.0285	0.87

Date	Genotype	Net Photosynthetic Rate $\mu mol\ (CO_2)\ m^{-2}\ s^{-1}$	Stomatal Conductance $mol\ (H_2O)\ m^{-2}\ s^{-1}$	Transpiration Rate $mol\ (H_2O)\ m^{-2}\ s^{-1}$
05/14/08	TAM 89E-51	20.20	0.3126	6.18
05/16/08	TAM 89E-51	18.41	0.2257	5.34
09/06/07	TAMCOT 22	22.32	0.9265	7.47
09/08/07	TAMCOT 22	24.05	1.1550	8.47
09/13/07	TAMCOT 22	19.17	0.1872	3.53
09/15/07	TAMCOT 22	10.89	0.1009	2.23
09/18/07	TAMCOT 22	21.37	0.6510	5.92
09/20/07	TAMCOT 22	20.75	0.6600	5.39
09/22/07	TAMCOT 22	22.10	1.2375	8.85
02/23/08	TAMCOT 22	21.15	0.5175	7.59
02/26/08	TAMCOT 22	22.18	0.2523	6.34
02/28/08	TAMCOT 22	21.10	0.2588	4.58
03/02/08	TAMCOT 22	7.88	0.0645	2.10
03/05/08	TAMCOT 22	6.26	0.0363	1.26
03/07/08	TAMCOT 22	21.81	0.4985	7.21
03/09/08	TAMCOT 22	26.11	0.7386	8.54
03/12/08	TAMCOT 22	25.53	1.0838	9.87
04/25/08	TAMCOT 22	20.71	1.0182	8.78
04/30/08	TAMCOT 22	27.90	1.0320	10.01
05/02/08	TAMCOT 22	20.00	0.9850	8.45
05/07/08	TAMCOT 22	18.94	0.2212	3.94
05/09/08	TAMCOT 22	11.43	0.0884	2.01
05/11/08	TAMCOT 22	4.74	0.0272	0.73
05/14/08	TAMCOT 22	21.44	0.6052	8.41
05/16/08	TAMCOT 22	20.72	0.5308	6.91

APPENDIX B

EXPERIMENT I – CHLOROPHYLL FLUORESCENCE MEASUREMENTS

Date	Genotype	F_t	ϕ_{II}	qP	qN	F_o	F_v/F_m	F_m
09/06/07	TAM 89E-51	0.344	0.635	0.783	0.325	0.378	0.756	1.548
09/08/07	TAM 89E-51	0.408	0.647	0.87	0.32	0.346	0.758	1.43
09/13/07	TAM 89E-51	0.408	0.6	0.805	0.408	0.267	0.788	1.263
09/15/07	TAM 89E-51	0.335	0.678	0.732	0.343	0.286	0.781	1.309
09/18/07	TAM 89E-51	0.358	0.674	0.784	0.477	0.26	0.793	1.254
09/20/07	TAM 89E-51	0.339	0.67	0.631	0.196	0.256	0.795	1.248
09/22/07	TAM 89E-51	0.351	0.668	0.682	0.312	0.315	0.742	1.222
02/23/08	TAM 89E-51	0.361	0.667	0.699	0.233	0.32	0.75	1.278
02/26/08	TAM 89E-51	0.355	0.652	0.855	0.391	0.251	0.798	1.243
02/28/08	TAM 89E-51	0.600	0.536	0.9	0.43	0.253	0.782	1.156
03/02/08	TAM 89E-51	0.386	0.616	0.869	0.278	0.263	0.773	1.158
03/05/08	TAM 89E-51	0.365	0.586	0.647	0.369	0.249	0.766	1.065
03/07/08	TAM 89E-51	0.591	0.491	0.875	0.324	0.274	0.784	1.266
03/09/08	TAM 89E-51	0.391	0.641	0.879	0.271	0.321	0.756	1.316

Date	Genotype	F_t	ϕ_{II}	qP	qN	F_o	F_v/F_m	F_m
03/12/08	TAM 89E-51	0.491	0.744	0.739	0.193	0.259	0.759	1.075
04/25/08	TAM 89E-51	0.464	0.728	0.733	0.185	0.318	0.734	1.195
04/30/08	TAM 89E-51	0.465	0.730	0.712	0.328	0.263	0.789	1.243
05/02/08	TAM 89E-51	0.483	0.746	0.744	0.251	0.391	0.821	2.181
05/07/08	TAM 89E-51	0.470	0.734	0.728	0.333	0.409	0.830	2.401
05/09/08	TAM 89E-51	0.459	0.732	0.730	0.326	0.315	0.742	1.222
05/11/08	TAM 89E-51	0.493	0.738	0.746	0.258	0.356	0.795	1.848
05/14/08	TAM 89E-51	0.385	0.521	0.733	0.311	0.320	0.750	1.278
05/16/08	TAM 89E-51	0.444	0.580	0.805	0.408	0.425	0.825	2.425
09/06/07	TAMCOT 22	0.383	0.544	0.853	0.329	0.276	0.754	1.123
09/08/07	TAMCOT 22	0.560	0.511	0.870	0.320	0.325	0.779	1.472
09/13/07	TAMCOT 22	0.485	0.509	0.842	0.188	0.301	0.759	1.251
09/15/07	TAMCOT 22	0.496	0.467	0.818	0.251	0.267	0.768	1.153
09/18/07	TAMCOT 22	0.359	0.483	0.875	0.272	0.238	0.790	1.130
09/20/07	TAMCOT 22	0.329	0.510	0.796	0.098	0.246	0.786	1.149
09/22/07	TAMCOT 22	0.365	0.583	0.745	0.330	0.260	0.796	1.275
02/23/08	TAMCOT 22	0.489	0.601	0.777	0.220	0.278	0.797	1.364
02/26/08	TAMCOT 22	0.355	0.624	0.783	0.325	0.250	0.787	1.171

Date	Genotype	F_t	ϕ_{II}	qP	qN	F_o	F_v/F_m	F_m
02/28/08	TAMCOT 22	0.359	0.446	0.787	0.330	0.410	0.836	2.507
03/02/08	TAMCOT 22	0.456	0.747	0.865	0.223	0.445	0.823	2.510
03/05/08	TAMCOT 22	0.475	0.748	0.865	0.197	0.251	0.798	1.243
03/07/08	TAMCOT 22	0.446	0.738	0.784	0.477	0.453	0.817	2.478
03/09/08	TAMCOT 22	0.644	0.687	0.796	0.098	0.433	0.818	2.373

APPENDIX C

EXPERIMENT II – GAS EXCHANGE PARAMETERS

Date	Genotype	Net Photosynthetic Rate $\mu mol\ (CO_2)\ m^{-2}\ s^{-1}$	Stomatal Conductance $mol\ (H_2O)\ m^{-2}\ s^{-1}$	Transpiration Rate $mol\ (H_2O)\ m^{-2}\ s^{-1}$
09/28/07	TAM 89E-51	19.80	0.6726	8.43
10/02/07	TAM 89E-51	13.47	0.0284	0.86
10/04/07	TAM 89E-51	6.12	0.0123	0.12
10/06/07	TAM 89E-51	13.90	0.2050	3.86
10/09/07	TAM 89E-51	12.33	0.4373	5.31
10/11/07	TAM 89E-51	20.10	0.6480	8.04
03/21/08	TAM 89E-51	16.86	0.3270	5.25
03/23/08	TAM 89E-51	16.52	0.1816	3.73
03/26/08	TAM 89E-51	5.42	0.0179	0.64
03/28/08	TAM 89E-51	10.27	0.0811	2.06
03/30/08	TAM 89E-51	15.60	0.2507	4.16
04/02/08	TAM 89E-51	15.54	0.3153	4.29
05/07/08	TAM 89E-51	20.56	0.6000	7.00
05/09/08	TAM 89E-51	13.70	0.1360	3.24
05/11/08	TAM 89E-51	1.14	0.0167	0.49
05/14/08	TAM 89E-51	7.81	0.0393	1.00
05/17/08	TAM 89E-51	18.50	0.8370	7.44
05/23/08	TAM 89E-51	20.80	0.5040	7.27
05/25/08	TAM 89E-51	21.20	0.5150	6.58
09/28/07	TAMCOT 22	20.10	0.7450	8.90
10/02/07	TAMCOT 22	12.51	0.0223	0.44

Date	Genotype	Net Photosynthetic Rate $\mu mol\ (CO_2)\ m^{-2}\ s^{-1}$	Stomatal Conductance $mol\ (H_2O)\ m^{-2}\ s^{-1}$	Transpiration Rate $mol\ (H_2O)\ m^{-2}\ s^{-1}$
10/04/07	TAMCOT 22	6.32	0.0123	0.15
10/06/07	TAMCOT 22	14.90	0.3029	4.62
10/09/07	TAMCOT 22	16.77	0.5055	6.58
10/11/07	TAMCOT 22	21.77	0.8477	8.50
03/21/08	TAMCOT 22	21.54	0.8226	8.74
03/23/08	TAMCOT 22	15.88	0.1474	3.07
03/26/08	TAMCOT 22	8.85	0.0225	1.37
03/28/08	TAMCOT 22	8.75	0.0628	1.42
03/30/08	TAMCOT 22	15.44	0.3984	5.88
04/02/08	TAMCOT 22	16.08	0.2924	4.12
05/07/08	TAMCOT 22	20.50	0.8226	8.74
05/09/08	TAMCOT 22	16.90	0.2330	4.10
05/11/08	TAMCOT 22	1.29	0.0154	0.42
05/14/08	TAMCOT 22	9.38	0.0505	1.31
05/17/08	TAMCOT 22	13.20	0.0971	2.27
05/25/08	TAMCOT 22	22.57	0.8082	7.38

APPENDIX D

EXPERIMENT II – CHLOROPHYLL FLUORESCENCE MEASUREMENTS

Date	Genotype	F_t	ϕ_{II}	qP	qN	F_o	F_v/F_m	F_m
09/28/07	TAM 89E-51	0.430	0.737	0.910	0.205	0.400	0.839	2.478
10/02/07	TAM 89E-51	0.435	0.729	0.920	0.260	0.390	0.835	2.358
10/04/07	TAM 89E-51	0.425	0.737	0.953	0.344	0.416	0.819	2.301
10/06/07	TAM 89E-51	0.421	0.733	0.937	0.359	0.405	0.824	2.299
10/09/07	TAM 89E-51	0.455	0.720	0.932	0.380	0.440	0.825	2.513
10/11/07	TAM 89E-51	0.650	0.570	0.570	0.410	0.456	0.822	2.558
03/21/08	TAM 89E-51	0.510	0.655	0.655	0.422	0.426	0.825	2.431
03/23/08	TAM 89E-51	0.418	0.758	0.991	0.365	0.451	0.824	2.558
03/26/08	TAM 89E-51	0.425	0.741	0.945	0.353	0.459	0.821	2.558
03/28/08	TAM 89E-51	0.410	0.744	0.943	0.385	0.461	0.820	2.558
03/30/08	TAM 89E-51	0.510	0.655	0.655	0.422	0.428	0.830	2.519
04/02/08	TAM 89E-51	0.429	0.746	0.940	0.309	0.426	0.833	2.553
05/07/08	TAM 89E-51	0.421	0.752	0.938	0.316	0.481	0.754	1.956
05/09/08	TAM 89E-51	0.410	0.760	0.909	0.236	0.419	0.792	2.016

Date	Genotype	F_t	ϕ_{II}	qP	qN	F_o	F_v/F_m	F_m
05/11/08	TAM 89E-51	0.458	0.746	0.945	0.321	0.328	0.819	1.810
05/14/08	TAM 89E-51	0.436	0.710	0.957	0.310	0.334	0.816	1.815
05/17/08	TAM 89E-51	0.403	0.773	0.982	0.282	0.491	0.783	2.266
05/23/08	TAM 89E-51	0.434	0.763	0.939	0.231	0.384	0.815	2.069
05/25/08	TAM 89E-51	0.400	0.750	0.965	0.404	0.379	0.799	1.885
09/28/07	TAMCOT 22	0.358	0.756	0.956	0.361	0.408	0.796	2.000
10/02/07	TAMCOT 22	0.429	0.748	0.904	0.332	0.345	0.809	1.808
10/04/07	TAMCOT 22	0.431	0.750	0.949	0.274	0.416	0.788	1.963
10/06/07	TAMCOT 22	0.456	0.736	0.980	0.462	0.419	0.787	1.969
10/09/07	TAMCOT 22	0.445	0.739	0.925	0.385	0.368	0.815	1.981
10/11/07	TAMCOT 22	0.570	0.560	0.560	0.494	0.374	0.810	1.969
03/21/08	TAMCOT 22	0.875	0.354	0.354	0.471	0.344	0.828	1.998
03/23/08	TAMCOT 22	0.650	0.570	0.570	0.410	0.331	0.823	1.866
03/26/08	TAMCOT 22	0.510	0.655	0.655	0.422	0.346	0.823	1.953
03/28/08	TAMCOT 22	0.469	0.650	0.650	0.477	0.338	0.819	1.861
03/30/08	TAMCOT 22	0.765	0.288	0.287	0.581	0.345	0.820	1.916
04/02/08	TAMCOT 22	0.483	0.519	0.519	0.608	0.330	0.823	1.866
05/07/08	TAMCOT 22	0.570	0.637	0.637	0.386	0.415	0.783	1.913

Date	Genotype	F_t	ϕ_{II}	qP	qN	F_o	F_v/F_m	F_m
05/09/08	TAMCOT 22	0.431	0.756	0.949	0.274	0.339	0.803	1.720
05/11/08	TAMCOT 22	0.431	0.706	0.706	0.426	0.328	0.820	1.821
05/14/08	TAMCOT 22	0.519	0.621	0.621	0.465	0.318	0.812	1.690
05/17/08	TAMCOT 22	0.508	0.644	0.644	0.442	0.315	0.819	1.736
05/25/08	TAMCOT 22	0.505	0.622	0.622	0.478	0.345	0.809	1.808

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