

AGRONOMY & SOILS

Response of Four Cotton Genotypes to N Fertilization for Root Hydraulic Conductance and Lint Yield

Philip J. Bauer*, William T. Pettigrew, and B. Todd Campbell

ABSTRACT

In controlled environments, hydraulic conductance of cotton (*Gossypium hirsutum* L.) roots is affected by nitrate supply. Limited information is available on the influence of nitrogen (N) fertilizer application on cotton root hydraulic conductance under field conditions. The objective of this study was to determine if applied N influenced root hydraulic conductance and lint yield of four diverse genotypes under field conditions. Studies were conducted in 2009 and 2010 at Florence, SC and Stoneville MS. Treatments were two applied N fertilizer rates (0 and 112 kg N ha⁻¹) and four genotypes (AGC 85, PD 2, Siokra L23, and Tamcot 22). Root hydraulic conductance was measured twice each year at Florence and once each year at Stoneville. Nitrate-N (NO₃-N) was determined in the stems of the plants on which root hydraulic conductance was measured. Cotton yield was measured at the end of the season. Stem NO₃-N concentration was higher in the N-fertilized cotton plant stems than in the stems of the unfertilized plants at all measurement times. Nitrogen fertilizer increased yield by 47% at Florence and by 23% at Stoneville. An N rate X genotype interaction occurred for lint yield at Stoneville. Nitrogen fertilization significantly increased lint yield for AGC 85, Siokra L23, and Tamcot 22 but not for PD 2. No significant interaction between N rates and genotypes occurred at Florence. All four genotypes had similar increases in boll weight and decreases in lint percent with N fertilization, suggesting that the differential yield increase among genotypes with N was due to boll number and not yield components. No differences occurred between N rates or among genotypes for root hydraulic conductance.

Nitrogen-deficient cotton exhibits many of the same symptoms that are attributed to water deficit stress (Hodges and Constable, 2010). These include reduced growth and leaf area expansion (Wullshleger and Oosterhuis, 1990). The reductions in growth may be due to reduced hydraulic conductance. Root hydraulic conductance is a measure of the ability of roots to conduct water. Root hydraulic conductance of cotton grown in controlled environments has been reported to be lower when some nutrient anions are deficient in the rooting media, including NO₃-N (Radin and Boyer, 1982; Radin and Matthews, 1989; Clarkson et al., 2000). The influence of NO₃-N on root hydraulic conductance may be due to changes in the number or the activity of water channel proteins (aquaporins) in the membranes of root cells (Clarkson et al., 2000). In corn (*Zea mays* L.) roots, nitrate deficiency reduced hydraulic conductance through the activity but not the number of aquaporin proteins (Gorska et al., 2008). A limited amount of research has been conducted on N deficiency effects on cotton hydraulic conductance in the field. In Arizona, whole plant hydraulic conductance of field-grown cotton was not affected by N deficiency (Radin et al., 1991).

Available data suggests little variability among genotypes adapted to the Mid-South for root hydraulic conductance (Pettigrew et al., 2009; Pettigrew and Meredith, 2012). Data is not available regarding whether genotypes respond differently to applied N for root hydraulic conductance.

Although there have been numerous studies investigating fertilizer N applications, reducing excessive reactive N in the environment has spurred renewed interest in increasing N use efficiency. Recent studies on improving N management in cotton include using remote sensing for site-specific application (e.g. Rutto et al., 2013) and managing N rates in crop rotations (Boquet et al., 2009; Hutmacher et al., 2004). Rochester (2011) found in high yielding environments optimal N uptake occurs when yield is 12.5 kg of lint for each kg of N.

A better understanding of the effect of N on root hydraulic conductance under field conditions may help in achieving higher N use efficiency through

P.J. Bauer* and B.T. Campbell, USDA-ARS, 2611 West Lucas Street, Florence, SC 29501; and W.T. Pettigrew, USDA-ARS, Crop Production Systems Research Unit, P.O. Box 350, Stoneville, MS 38776

*Corresponding author: phil.bauer@ars.usda.gov

better N management and through identifying potentially exploitable genetic variability. We conducted a two-year study to assess genotypic responses to N for root hydraulic conductance under field conditions. Our objective was to determine if N fertilization and genotype influence cotton root hydraulic conductance and lint yield.

MATERIALS AND METHODS

Field experiments were conducted in 2009 and 2010 at Florence, SC and Stoneville, MS. The experiment at Florence was conducted on a soil mapped as Goldsboro loamy sand (fine-loamy, siliceous, subactive, thermic Aquic Paleudult) soil. At Stoneville, the experiment was conducted on a soil mapped as Bosket fine sandy loam (fine-loamy, mixed, thermic Mollic Hapludalf). At both locations in both years, treatments were N rate (0 and 112 kg N ha⁻¹) and genotype (AGC 85, PD 2, Siokra L23, and Tamcot 22). The experimental design was a randomized complete block and there were five replicates of each treatment combination at each location both years.

The four genotypes used in this experiment originated in Arizona (AGC 85, Percy et al., 2006), South Carolina (PD 2, Culp et al., 1985), Texas (Tamcot 22, Thaxton et al. 2005), and Australia (Siokra L23, Reid, 1992). AGC 85 is a germplasm line with tolerance to heat. Siokra L23 is an okra-leaf type cotton with putative tolerance to soil water deficit stress. PD 2 is a short-season maturity cultivar with excellent fiber properties. Tamcot 22 is a mid-season maturity cultivar with high yield potential and excellent fiber quality.

Planting occurred on 4 May 2009 and 18 May 2010 at Florence. Planting dates at Stoneville were 27 April 2009 and 21 April 2010. Seeding rate was approximately 10 seeds m⁻¹ of row. Plots at both Florence and Stoneville were two rows that were 1-m wide and 10.7 m long. Each two-row plot was bordered on each side by a genotype that was common to all plots at each location in each year. Plots were managed using Clemson University or Mississippi State University recommendations for soil fertility (except N), insect management, and defoliation. Weeds were controlled using a combination of pre- and post-plant herbicides and hand-weeding.

Application of N differed between the two locations. At Stoneville, 112 kg N ha⁻¹ (as urea-ammonium nitrate solution) was applied preplant and incorporated into the soil of the designated plots. At Florence, the N (as dry fertilizer NH₄NO₃) was

knifed into the 112 kg N ha⁻¹ plots shortly after the first flower buds were detected in June each year. The N fertilizer was placed approximately 15-cm to the side of each row and 10-cm deep.

Root hydraulic conductance was determined twice each year in Florence and once each year in Stoneville. Data were collected when plants had no visual symptoms of water deficit stress. At both locations, Dynamax HPFM high-pressure flow meters (Dynamax, Houston, TX) were used to measure conductance. The first measurements in Florence were made at about one week after the N fertilizer was applied. This measurement time was selected to evaluate whether the N application affected root hydraulic conductance before the N had a substantial influence on plant size. Measurements were made over two to three days centered at 51 days after planting (DAP) in 2009 and at 49 DAP in 2010. The second measurements were made at 122 DAP in 2009 and 103 DAP 2010. Measurements in Stoneville were made at 57 DAP in 2009 and 77 DAP in 2010. For the measurements, two (early measurements at Florence and at Stoneville) or one (late measurements at Florence) representative plant(s) were chosen in each plot and hydraulic conductance of the roots was measured as described by Pettigrew et al. (2009).

Measuring soil NO₃-N concentrations near all root surfaces that contribute to total root hydraulic conductance was not feasible, so we measured NO₃-N in the stems of the plants to assess uptake of that nutrient at the time of the hydraulic conductance measurements. Branches and leaves were removed from the stems and a 30-cm stem section was collected. Stems were dried at 60 °C for three days and ground. Anions were extracted from the ground stem material with water. Concentration of NO₃-N in the extracts was measured by chemically suppressed ion chromatography (IC) using a Dionex 2000 Ion Chromatograph (Dionex Corporation, Bannockburn, IL) (ASTM Standard D4327-11).

At the end of each season at both locations, 50 bolls (25 bolls in Florence in 2010) from each plot were handpicked for lint percent determinations. At Florence, first sympodial position bolls in the middle of the canopy were picked from the entire plot length. At Stoneville, one plant within the plot was selected and all the bolls on that plant were picked before moving to an adjacent plant and picking all its bolls. This continued until 50 bolls were harvested. Seed cotton from the bolls was ginned on laboratory gins. The two rows of each plot were then harvested with two-row cotton pickers equipped with on-board

weighing systems. Lint yield was calculated by multiplying the plot seed cotton weight from the weighing system by lint percent.

Analysis of variance was conducted on all data over years but by location because of the different timings of the N application using the GLIMMIX procedure in SAS. Years, N rates, and genotypes were considered fixed effects and replicates were considered random. Means were separated using the pdiff procedure when sources of variation were significant ($P \leq 0.05$).

RESULTS AND DISCUSSION

At both sampling times in Florence and at Stoneville, cotton grown with applied N fertilizer had higher stem $\text{NO}_3\text{-N}$ concentration than the cotton grown without N fertilizer. Averaged over years and genotypes, stem $\text{NO}_3\text{-N}$ concentrations for the unfertilized and fertilized cotton were 0.69 and 2.12 g kg^{-1} during the squaring period at Florence, 0.01 and 0.15 g kg^{-1} during the boll fill period at Florence, and 1.17 and 3.21 g kg^{-1} at Stoneville (all $P \leq 0.05$ for F values from ANOVA). Differences among genotypes occurred for stem $\text{NO}_3\text{-N}$ concentration. Averaged over years and N rates at Florence, Tamcot 22 ranked highest (but not significantly different from only Siokra L23) for average $\text{NO}_3\text{-N}$ concentration among the four genotypes at the squaring period but ranked lowest (but not significantly different from AGC 85) during the boll fill period (data not shown). At Stoneville, Siokra L23, Tamcot 22, and AGC 85 all had greater stem $\text{NO}_3\text{-N}$ concentration than PD 2 (data not shown). The lack of consistency among genotypes across locations and sampling times is similar to the petiole $\text{NO}_3\text{-N}$ concentration ($\text{NO}_3\text{-N}$ concentration of the petiole of the uppermost fully expanded leaf) results of Sunderman et al. (1979)

who compared two genotypes with five fertilizer N rates and found an inconsistent genotype response.

The response to applied N for lint yield was greater at Florence than at Stoneville. Nitrogen fertilization resulted in a 47% yield increase at Florence but only a 23% increase at Stoneville. Averaged over N rates at Florence, AGC85 and Tamcot 22 had higher yield than the other two genotypes. A genotype X N rate interaction occurred for lint yield at Stoneville (Table 1). At that location, N fertilization significantly increased yield of AGC 85, Siokra L23, and Tamcot 22 but not PD 2 (Table 1). PD 2 is an early maturing cultivar, which may suggest limited yield potential, but our observations in Florence indicate that Tamcot 22 appears to mature even earlier. Although the N rate X genotype interaction was not significant at Florence, PD 2 exhibited a smaller response to N (246 kg ha^{-1}) than did the other three genotypes (approximately 400 kg ha^{-1}).

Boll size was lower at Stoneville than at Florence (Table 2) because of the way seed cotton samples were collected. Only mid-canopy first position bolls were sampled at Florence whereas all bolls in the canopy were sampled at Stoneville. Even though sampling methods differed, results from the two locations for yield components were similar. At both locations, N fertilization increased boll weight (Table 2) but decreased lint percent (Table 3). Others (Boman et al., 1997; Fritschi et al., 2003) have found decreasing lint percent with increasing N application rate. Genotypes differed for both boll size (Tables 2) and lint percent (Tables 3). Ranking of the genotypes for these yield component traits were identical at the two locations. PD 2 had the smallest boll size and lowest lint percent of the four genotypes. The N X genotype interaction that occurred for lint yield at Stoneville did not occur for boll size or lint percent, indicating that the difference in genotype response to N rate for yield was likely due to boll number.

Table 1. Effect of N fertilization rate and genotype on lint yield. Data are averaged over years at both locations.

Genotype	Florence			Stoneville		
	N Rate (kg ha^{-1})					
	0	112	Mean	0	112	Mean
	----- kg ha^{-1} -----					
AGC85	987	1380	1184a	970de	1277a	1123a
PD 2	699	945	822b	875ef	950def	912b
Siokra L23	644	1066	855b	1013cd	1205ab	1109a
Tamcot 22	822	1248	1035a	841f	1126bc	984b
Mean	788b	1160a		925a	1139b	

†Means within a column or row followed by the same letter are not different ($P < 0.05$)

Table 2. Effect of N fertilization rate and genotype on individual boll weight. Data are averaged over both years at both locations.

Genotype	Florence			Stoneville		
	N Rate (kg ha ⁻¹)					
	0	112	Mean	0	112	Mean
----- gms -----						
AGC85	5.3	5.9	5.6a	3.9	4.5	4.2a
PD 2	4.8	5.2	5.0c	3.3	3.9	3.6c
Siokra L23	5.0	5.5	5.3b	3.5	4.1	3.8b
Tamcot 22	5.3	6.0	5.6a	3.7	4.3	4.0a
Mean	5.1b	5.6a		3.6a	4.2a	

†Means within a column or row followed by the same letter are not different (P<0.05).

Table 3. Effect of N fertilization rate and genotype on lint percent. Data are averaged over years at both locations.

Genotype	Florence			Stoneville		
	N Rate (kg ha ⁻¹)					
	0	112	Mean	0	112	Mean
----- % -----						
AGC85	40.2	39.7	39.9b	39.8	38.1	39.0b
PD 2	38.4	37.5	38.0c	37.0	35.3	36.2c
Siokra L23	41.5	41.1	41.3a	42.0	40.3	41.1a
Tamcot 22	41.4	41.2	41.3a	41.2	40.3	40.8a
Mean	40.4a	39.9b		40.0a	38.5b	

†Means within a column or row followed by the same letter are not different (P<0.05).

Nitrogen level and genotype means for root hydraulic conductance are shown in Table 4. Larger root systems have higher hydraulic conductance (Tyree et al., 1985), which likely explains the higher root hydraulic conductance during the boll fill period at Florence than during the squaring period. Although the cotton plants fertilized with N had higher stem NO₃-N concentration and yield than the unfertilized plants, N fertilization had no significant influence on root hydraulic conductance at the times measurements were made in this study (Table 4). Radin et al. (1991) also found that N deficiency did not reduce hydraulic conductance of field-grown cotton in Arizona. Additional studies appear warranted to determine why the N response on root hydraulic conductance found in controlled environments is less apparent under field conditions in order to better explain N effects on growth and development in the field.

No differences among the four genotypes occurred for root hydraulic conductance (Table 4) and the N level X genotype interaction was not significant at either sampling time in Florence or in Stoneville. The lack of difference among genotypes for root hydraulic conductance in this study supports the limited amount of previous research on cotton genotype evaluations.

Although Pettigrew et al. (2009) reported root hydraulic conductance differed among four commercial cultivars, a subsequent study found no differences among ten cultivars (Pettigrew and Meredith, 2012). More extensive searches of cotton germplasm for both root and whole plant hydraulic conductance appear warranted. In soybean (*Glycine max* L.), genotypes have been found that can provide tolerance to drought periods via differences in leaf hydraulic conductance (Sloane et al., 1990; Sinclair et al., 2008).

Table 4. Effect of N fertilization rate and genotype on root hydraulic conductance. Data are averaged over both years. No significant (P<0.05) differences between N rates or among genotypes occurred.

N Rate (kg ha ⁻¹)	Florence		Stoneville
	Squaring	Boll Fill	
	----- kg s ⁻¹ MPa ⁻¹ X 10 ⁻⁵ -----		
0	3.0	5.9	7.3
112	2.2	6.5	7.3
Genotype			
AGC85	3.2	6.6	7.1
PD 2	2.7	6.6	7.8
Siokra L23	2.5	5.8	7.5
Tamcot 22	2.0	5.9	6.9

DISCLAIMER

Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

REFERENCES

- American Society for Testing and Materials. Test method for anions in water by chemically suppressed ion chromatography. ASTM Standard D4327-11. Annual Book of Standards, Vol. 11.01.
- Boman, R.K., W.R. Raun, R.L. Westerman, and J.C. Banks. 1997. Long-term nitrogen fertilization in short-season cotton: interpretation of agronomic characteristics using stability analysis. *J. Prod. Agric.* 10:7:70-75.
- Boquet, D.J., B.S. Tubana, H.J. Mascagni, Jr., M. Holman, and S. Hague. 2009. Cotton yield responses to fertilizer nitrogen rates in a cotton-corn rotation. *Agron. J.* 101:400-407.
- Clarkson, D.T., M. Carvajal, T.I. Henzler, R.N. Waterhouse, A.J. Smyth, D.T. Cooke, and E. Steudle. 2000. Root hydraulic conductance: diurnal aquaporin expression and the effects of nutrient stress. *J. Exp. Bot.* 51:61-70.
- Culp, T.W., R.F. Moore, and J.B. Pitner. 1985. Registration of PD-2 cotton. *Crop Sci.* 25:198-199.
- Fritschi, F.B., B.A. Roberts, R.L. Travis, D.W. Rains, and R.B. Hutmacher. 2003. Response of irrigated acala and pima cotton to nitrogen fertilization: growth, dry matter partitioning, and yield. *Agron. J.* 133-146.
- Gorska, A., A. Zwieniecka, N.M. Holbrook, and M.A. Zwieniecki. 2008. Nitrate induction of root hydraulic conductivity in maize is not correlated with aquaporin expression. *Planta.* 228:989-998.
- Hodges, S.C., and G. Constable. 2010. Plant responses to mineral deficiencies and toxicities, pp. 142-161. In J. McD. Stewart, D.M. Oosterhuis, J.J. Heitholt, and J.R. Mauney (eds) *Physiology of Cotton*. Springer, New York, New York.
- Hutmacher, R.B., R.L. Travis, D.W. Rains, R.N. Vargas, B.A. Roberts, B.L. Weir, S.D. Wright, D.S. Munk, B.H. Marsh, M.P. Keeley, F.B. Fritschi, D.J. Munier, R.L. Nichols, and R. Delgado. 2004. Response of recent acala cotton varieties to variable nitrogen rates in the San Joaquin Valley of California. *Agron. J.* 96:48-62.
- Percy, R.G., O.L. May, M. Ulloa, and R.G. Cantrell. 2006. Registration of AGC85, AGC208, and AGC375 upland cotton lines. *Crop Sci.* 46:1828-1829.
- Pettigrew, W.T., and W.R. Meredith. 2012. Genotypic variation in physiological strategies for attaining cotton lint yield production. *J. Cotton Sci.* 16:179-189.
- Pettigrew, W.T., W.T. Molin, and S.R. Stetina. 2009. Impact of varying planting dates and tillage systems on cotton growth and lint yield production. *Agron. J.* 101:1131-1138.
- Radin, J.W., and J.S. Boyer. 1982. Control of leaf expansion by nitrogen nutrition in sunflower plants. Role of hydraulic conductivity and turgor. *Plant Physiol.* 69:771-775.
- Radin, J.W., and M.A. Matthews. 1989. Water transport properties of cortical cells in roots of nitrogen- and phosphorus-deficient cotton seedlings. *Plant Physiol.* 89:264-268.
- Radin, J.W., J.R. Mauney, and P.C. Kerridge. 1991. Effects of nitrogen fertility on water potential of irrigated cotton. *Agron. J.* 83:739-743.
- Reid, P.E. 1992. Siokra L23. *Plant Var. J.* 5:2.
- Rochester, I.J. 2011. Assessing internal crop nitrogen use efficiency in high-yielding irrigated cotton. *Nutr. Cycl. Agroecosyst.* 90:147-156.
- Rutto, E., B.D. Arnall, J.L. May, K. Butchee, and W.R. Raun. 2013. Ability of cotton (*Gossypium hirsutum* L.) to recover from early season nitrogen deficiency. *Journal of Cotton Sci.* 17:70-79.
- Sinclair, T.R., M.A. Zwieniecki, and N.M. Holbrook. 2008. Low leaf hydraulic conductance associated with drought tolerance in soybean. *Physiologia Plantarum.* 132:446-451.
- Sloane, R.J., R.P. Patterson, and T.E. Carter. 1990. Field drought tolerance of a soybean plant introduction. *Crop Sci.* 30:118-123.
- Sunderman, H.D., A.B. Onken, and L.R. Hossner. 1979. Nitrate concentration of cotton petioles as influenced by cultivar, row spacing, and N application rate. *Agron. J.* 71:737-737.
- Thaxton, P.M., C.W. Smith, and R. Cantrell. 2005. Registration of 'Tamcot 22' high-yielding upland cotton cultivar. *Crop Sci.* 45:1165-1166.
- Tyree, M.T., S. Patino, J. Bennink, and J. Alexander. 1995. Dynamic measurements of root hydraulic conductance using a high-pressure flowmeter in the laboratory and field. *J. Exp. Botany.* 46:83-94.
- Wullschlegel, S.D., and D.M. Oosterhuis. 1990. Canopy development and photosynthesis as influenced by nitrogen nutrition. *J. Plant Nut.* 13:1141-1154.