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Intermittent Shade Effect on Gas Exchange of Cotton Leaves in the Humid Southeastern USA

Philip J. Bauer,* E. John Sadler, and James R. Frederick

ABSTRACT

Convective cumulus clouds intermittently shade growing plants on most days during the summer months in the southeastern USA. Previous research indicates a significant delay in the recovery of stomatal conductance (g_s) of cotton (*Gossypium hirsutum* L.) leaves following a period of shade. Our objective was to determine the effect of shade on leaf net CO_2 exchange rate (CER) and g_s of three cotton cultivars. We monitored CER and g_s of greenhouse- and field-grown cotton before, during, and after shading plants for up to 9 min at photosynthetically active radiation (PAR) levels of <300 μmol quanta m^{-2} s^{-1}. The three cultivars had the same gas exchange response to shade. With the imposition of a 6-min shade on greenhouse-grown plants at 7 weeks after planting (WAP), g_s of uppermost fully expanded leaves fell 43%. Two weeks later, a 6-min shade reduced g_s of uppermost fully expanded leaves by 97%. Under field conditions, a 9-min shade reduced g_s by 35% early in the cutout period (cessation of vegetative growth) in 1992 and 42% late in the cutout period in 1994. Under both greenhouse and field growing conditions, the low PAR levels with shade reduced CER to near 0 μmol CO_2 m^{-2} s^{-1}. Recovery of CER to preshade levels after the shade was removed coincided with the rate of recovery of g_s. In the greenhouse-grown plants, recovery of CER to preshade levels following 6 min of shade did not occur until 7 min (at 7 WAP) and 10 min (at 9 WAP) after the shade was removed. Field-grown leaves needed only 4 min to recover to preshade levels of CER and g_s following 9 min of shade. The results suggest that, following a brief shade period, field-grown cotton leaves reacclimate within 4 min, while leaves on greenhouse-grown plants may take longer.

SCATTERED CLOUDS cause highly variable irradiance levels on typical summer days in the humid southeastern USA. Shading due to clouds can quickly and substantially affect crop water status (Sojka et al., 1984) and energy balance components (Evans and Sadler, 1987). Less is known about the effect that intermittent shading has on leaf gas exchange. Portable gas exchange measuring devices are often used in field research to describe treatment effects on plant health and productivity, and collection of these data can be frustrated by overpassing clouds. A better understanding of the influence of variable irradiance on leaf gas exchange will improve the quality of the leaf gas exchange data that are collected in the region.

Reduced light intensity affects both CER and g_s. Immediately upon shading, CER is limited by the lack of PAR. Shading also causes stomates to close. In field experiments in Israel, Petersen et al. (1991) found that after 5 min of shade (PAR = 120 μmol quanta m^{-2} s^{-1}), leaf abaxial g_s of 'Acala SJ-2' cotton fell to about 30% of preshade levels. The rate of stomatal opening following the total 15-min shade period was slower than the rate of stomatal closure, with abaxial g_s still being less than 70% of preshade levels 25 min after the shade was removed.

With genotypes adapted to the Southeastern region, our casual observations suggested that gas exchange recovery of cotton leaves following a brief shade is not as prolonged as reported for the more arid conditions in Israel. Since limited information is available on recovery of CER and g_s following a brief shade event, our objective was to measure the effect of intermittent low light irradiance on leaf gas exchange of cotton grown in the humid southeastern USA. A second objective was to compare cultivars of diverse origin for their transient leaf gas exchange response to shade.

MATERIALS AND METHODS

Data were collected from greenhouse-grown plants in 1992 and from field-grown plants in 1992, 1993, and 1994 at Clemson University's Pee Dee Research and Education Center in Florence, SC. In the greenhouse experiment and the field experiments of 1992 and 1993, the cotton cultivars Deltapine Acala 90 and Pee Dee 3 were used because of their diversity of origin (Deltapine Acala 90 is of western USA origin, and Pee Dee 3 is of eastern USA origin). Both were widely grown throughout the southeastern USA Cotton Belt when we began these experiments. In 1994, Acala SJ-2 was evaluated with the other two cultivars.

Crop Culture

The greenhouse-grown plants were seeded into 6.5-L pots containing a commercial potting medium on 25 Feb. 1992. Four seeds were planted in each pot, and emerged seedlings were thinned to provide two uniform seedlings per pot at about 2 WAP. Plants were watered as needed throughout the experiment. Once a week, a commercial fertilizer that contained N, P, K, and micronutrients was applied with the irrigation water. Plants were grown without supplemental lighting. Midday photosynthetic photon flux density at the top of the plants was approximately 1600 μmol quanta m^{-2} s^{-1}. Experimental design was randomized complete block with four replicates. A replicate of each cultivar consisted of five pots (10 plants).

In the field experiments, experimental design in 1992 and 1993 was randomized complete block with three replicates. Plot size was four rows, each 0.97 m wide by 8.85 m long. Experimental design was randomized complete block with two replicates in 1994. Plot size in 1994 was four rows, each 0.97 by 14.75 m. In all three years, row orientation was east–west. Preplant tillage consisted of disking and harrowing each year. Plots were in-row subsoiled and then seeded in mid-May each year. Nitrogen (78.4 kg ha^{-1}), S (11.2 kg ha^{-1}), and B (0.56 kg ha^{-1}) were applied annually. Lime, P, K, and Mn were

P.J. Bauer and E.J. Sadler, USDA-ARS Coastal Plains Soil, Water, and Plant Res. Ctr., 2611 W. Lucas St., Florence, SC 29501-1241; J.R. Frederick, Dep. of Agronomy, Clemson Univ., Pee Dee Res. and Educ. Ctr., Florence, SC 29501. Contribution of the USDA-ARS and the South Carolina Agric. Exp. Stn. Received 22 Sept. 1995. *Corresponding author (bauer@florence.ars.usda.gov).

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Abbreviations: CER, leaf net CO_2 exchange rate; EDT, Eastern Daylight Time; g_s, stomatal conductance; PAR, photosynthetically active radiation; WAP, weeks after planting.

applied based on soil tests to meet Clemson University Cooperative Extension Service recommendations. Weeds were controlled with a combination of pre- and postemergence herbicides, mechanical cultivation, and handweeding. For insect pest control, each year a systemic insecticide was applied in-furrow at planting and organophosphate and pyrethroid insecticides were applied throughout each season as pest populations warranted. All pesticides were applied at label-recommended rates. Plots were irrigated (furrow, 1992 and 1993; overhead sprinkler, 1994) when visual symptoms of water deficit stress were first noticed.

Data Collection and Analysis

A shading apparatus was constructed of two layers of commercially available greenhouse black shadecloth. The frame of the apparatus was 2.6 m long and 1.77 m wide. To reduce the influence of sun angle and to improve uniformity of shading, the legs of the frame were 0.76 m long on the side placed towards the sun and 2.06 m long on the side away from the sun. When the gas exchange measurements were taken, the shading apparatus was placed over the plants from the south.

For the greenhouse-grown plants, leaf gas exchange measurements were taken on 9 and 23 April (7 and 9 WAP). All pots were watered the evening before the measurements and were placed outside before 0800 h (EDT) the next morning. The pots were placed in rows oriented in an east–west direction. Leaf gas exchange data from the field-grown plants were collected on 10 and 11 Aug. 1992 (during the cutout period; i.e., when there is no vegetative growth), 27 Aug. and 1 Sept. 1993, and 26 Aug. 1994 (at the end of the cutout period, just before first boll opening). All leaf gas exchange measurements were collected when skies were cloud-free between 1030 and 1500 h (EDT). Two LI-COR 6200 photosynthesis systems (Li-Cor,[1] Lincoln, NE) with 1-L chambers were used to measure leaf gas exchange on uppermost fully expanded leaves. For the greenhouse-grown plants, each instrument was assigned two replicates, and both instruments were used to collect data from two replicates at the same time (one leaf per replicate at each measurement time). In the field, duplicate samples were collected at each measurement time by simultaneously using both instruments within each replicate. For both the field- and greenhouse-grown plants, the sampling procedure consisted of taking a preshade measurement and then placing the shading apparatus over the plants and immediately taking another measurement. Measurements of the shaded leaves were collected at 3-min intervals for 6 min (greenhouse experiment; field experiment in 1993) or 9 min (field experiments in 1992 and 1994). After the last measurement under the shade, the apparatus was removed and another measurement was immediately taken. Thereafter, measurements were made at 3-min intervals for up to 12 min. Gas exchange was not measured twice on the same leaf. Quantum sensors mounted on the chambers measured PAR. Under the shading apparatus, PAR was always <300 μmol quanta m^{-2} s^{-1}. Light intensity for the unshaded measurements was approximately 2000 μmol quanta m^{-2} s^{-1}.

The leaf gas exchange data were subjected to analysis of variance on each date that measurements were collected. Single degree-of-freedom contrasts were calculated to compare preshade CER and g_s with shaded and post-shade rates. Regression equations were calculated to relate CER and g_s for all post-shade measurements.

[1] Mention of a trade name is for information only. It does not constitute a guarantee by the USDA or Clemson University, nor does it constitute an endorsement to the exclusion of other products that may also be suitable.

RESULTS AND DISCUSSION

The cultivars had similar CER and g_s responses to shade (Fig. 1 and 2). The F-values for the cultivar × measurement time interactions for CER and g_s were all nonsignificant (P = 0.10). These results are similar to those of Pasternak and Wilson (1973), who reported the leaf gas exchange response to shade to be similar among four sorghum [*Sorghum bicolor* (L.) Moench] cultivars.

The CER response to shade for uppermost fully expanded leaves of the greenhouse-grown plants at 7 and 9 WAP is shown in Fig. 1. Preshade CER was about 18 μmol m^{-2} s^{-1} on both measurement dates. With shading, CER fell rapidly to 0 μmol m^{-2} s^{-1} or below (Fig. 1). Photosynthetic rates returned to preshade levels at 7 and 10 min post shade for the 7- and 9-wk-old plants, respectively. Stomatal closure was greater when measurements were collected at 9 WAP than at 7 WAP (Fig. 3). At 7 WAP, g_s levels fell from 0.235 to 0.135 mol H_2O m^{-2} s^{-1} with the imposition of shade. At 9 WAP, preshade g_s (0.265 mol H_2O m^{-2} s^{-1}) was similar to the preshade levels when the plants were younger (i.e., at 7 WAP), but g_s fell to 0.040 mol H_2O m^{-2} s^{-1} by the end of the 6-min shade period.

Photosynthesis and leaf g_s were higher in the field-grown than in the greenhouse-grown plants (Fig. 2 and 3), as has been previously reported (Radin, 1992). As

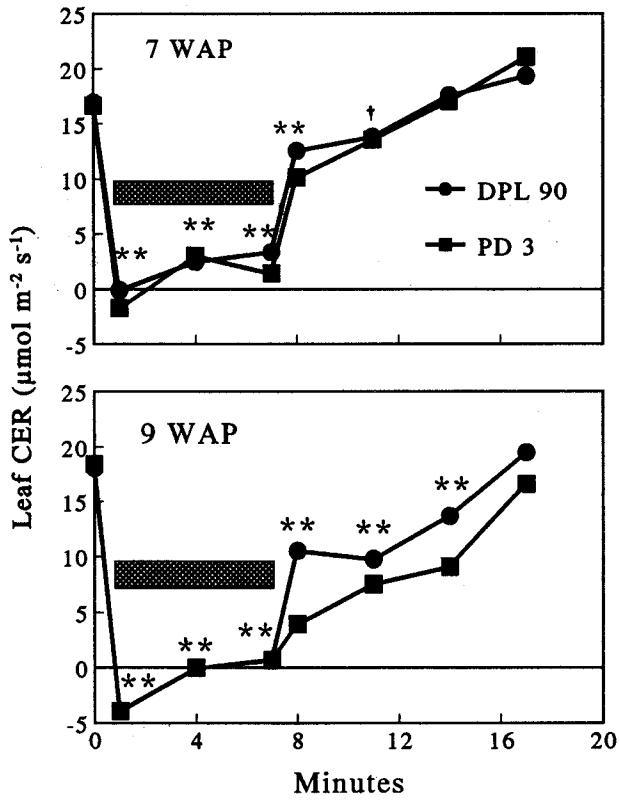

Fig. 1. The effect of a 6-min shade period on uppermost fully expanded leaf net CO_2 exchange rate (CER) of two greenhouse-grown cotton cultivars at 7 (top) and 9 (bottom) weeks after planting (WAP). The symbols † and ** over the markers indicate that CER (averaged over both cultivars) was lower than the preshade measurement at P = 0.10 and P = 0.01, respectively. Hatched bars indicate the shade period.

with the greenhouse-grown plants, the rate of recovery of CER following the shade period in the field coincided with the rate of stomatal reopening. Leaf CER immediately following the shade period was lower than preshade rates, but subsequent CER measurements were the same as those taken before shade was imposed (Fig. 2). In 1993, the shade period was shorter (6 min in 1993; 9 min in 1992 and 1994). The trend for CER recovery from shade was similar to the other two years (data not shown), but the measurement immediately after shade was removed was not different ($P < 0.05$) from the preshade measurement. Leaf g_s was less ($P < 0.05$) than the preshade levels in both 1992 and 1994 immediately after the shade was removed (Fig. 3). Similar to CER, g_s after the initial post-shade measurement was not different from the preshade g_s.

The relationship between leaf CER and g_s is shown in Fig. 4 for all post-shade measurements in Fig. 1, 2, and 3. Following the shade period, leaf CER was proportional to g_s in the uppermost fully expanded leaves of the greenhouse-grown plants (Fig. 4). For uppermost fully expanded leaves of the field-grown plants, the relationship between CER and g_s was less defined, because g_s of those leaves was higher and both leaf CER and g_s returned to preshade levels more rapidly in the field experiments. Regression equations for the relationship between CER and g_s in the different environments of this study are: greenhouse leaf CER = $0.93 + 87.52(g_s) - 80.35(g_s)^2$, $R^2 = 0.94$; field leaf CER in 1992 = $12.92 + 18.11(g_s) - 4.08(g_s)^2$, $R^2 = 0.66$; field leaf CER in 1994 = $7.46 + 14.87(g_s) - 3.04(g_s)^2$, $R^2 = 0.63$.

Lower leaf CER at a similar g_s for the field-grown cotton in 1994 compared with 1992 (Fig. 4) indicates that the mesophyll was less able to fix carbon at the 1994 measurement date than at the 1992 measurement date. This is probably because the leaves in 1994 (when measurements were made in late August) were older than the leaves in 1992 (measurements made in mid-August). Wullschleger and Oosterhuis (1990) reported that, for cotton leaves, CER declined almost linearly as leaf age advanced from about 20 to 60 d from when they unfolded. Even though maximum CER differed, g_s was similar in 1992 and 1994 (Fig. 3 and 4), and the recovery response following shade was the same in each year (Fig. 3).

The recovery of cotton leaf g_s in this study following a shade period was more rapid than for cotton grown in Israel (Petersen et al., 1991). This could have been due to growing conditions or to the method of measuring g_s. We measured gas exchange of both leaf surfaces simultaneously and did not separate abaxial and adaxial surfaces. Petersen et al. (1991), on the other hand, measured only abaxial cotton leaf surfaces. Sharpe (1973)

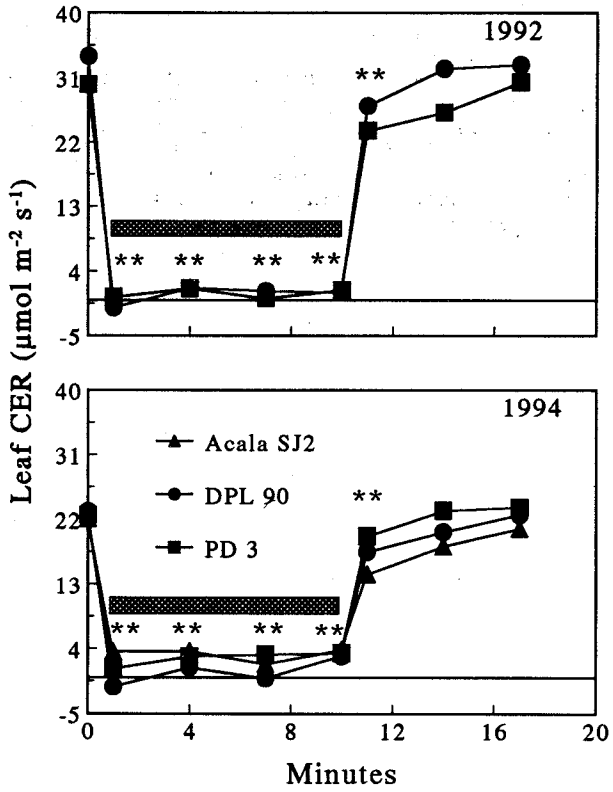

Fig. 2. The effect of a 9-min shade period on uppermost fully expanded leaf net CO_2 exchange rate (CER) of two cotton cultivars in August 1992 (top) and three cotton cultivars in August 1994 (bottom) that were grown under field conditions. Hatched bars indicate the shade period. The symbol ** over markers indicates that CER (averaged over cultivars) was lower than the preshade measurement at P = 0.01.

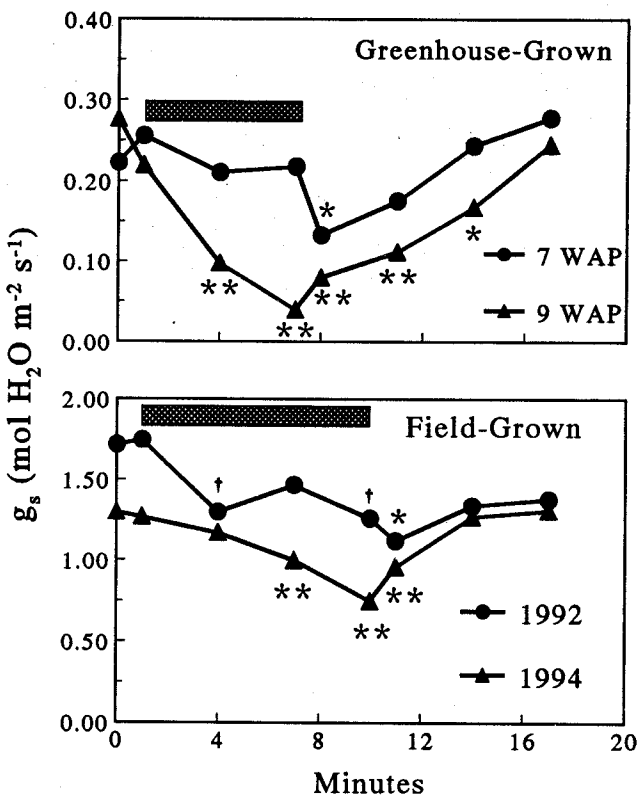

Fig. 3. The effect of shade on stomatal conductance (g_s) of uppermost fully expanded leaves on greenhouse-grown (top) and field-grown (bottom) cotton plants. Greenhouse-grown plants were measured at 7 and 9 weeks after planting (WAP). Field measurements were taken in August of each year. Hatched bars indicate the shade period (6 min, greenhouse; 9 min, field). The symbols †, *, and ** over or under a marker indicate that g_s was lower than the preshade measurement at P = 0.10, P = 0.05, and P = 0.01, respectively.

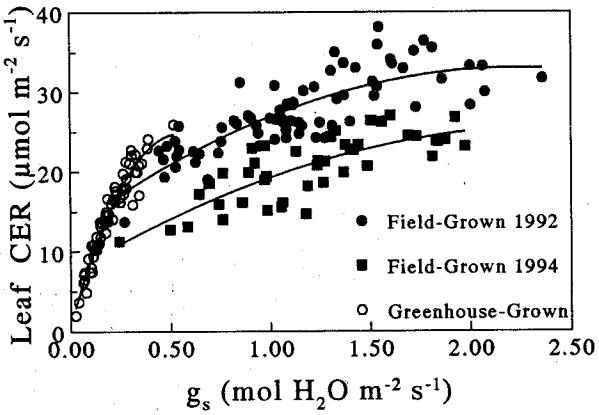

Fig. 4. Relationship between stomatal conductance (g_s) and leaf net CO_2 exchange rate (CER) after shading for uppermost fully expanded leaves of greenhouse-grown (all measurements from 7 and 9 wk after planting) and field-grown cotton plants in mid-August 1992 and late August 1994.

showed that cotton adaxial stomates are more sensitive than abaxial stomates to diurnal changes in light intensity. Kanemasu and Tanner (1969) reported a similar finding for snap bean (*Phaseolis vulgaris* L.). Our results indicate that cotton leaves in the humid Southeast return to preshade gas exchange levels rapidly after shading. This finding is supported by Garcia et al. (1990), who found that it took only 4 min for canopy CER of soybean [*Glycine max* (L.) Merr.] to attain preshade levels following a shade period of more than 10 min at PAR < 500 μmol quanta m^{-2} s^{-1}.

Obviously, the amount of reduction in light intensity and the length of time the leaves are shaded will determine the degree of stomatal closure and the amount of time needed for g_s to return to preshade levels. For our measurements, we chose a light intensity (<300 μmol quanta m^{-2} s^{-1}) lower than that found under most isolated clouds, but similar in shade duration (9 min) to that a researcher may experience while taking gas exchange measurements in the field. We conclude that following a brief shade, field-grown cotton leaves need only a few minutes (about 4 min) to readjust to preshade levels before midday gas exchange data collection can be resumed in the humid southeastern USA; greenhouse-grown plants may take longer to recover.

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