2001

Light-saturated CO2 assimilation rates of corn and velvetleaf in response to leaf nitrogen and development stage

John L. Lindquist

University of Nebraska-Lincoln, jlindquist1@unl.edu

Light-saturated CO$_2$ assimilation rates of corn and velvetleaf in response to leaf nitrogen and development stage

John L. Lindquist
Corresponding author. Department of Agronomy, University of Nebraska, Lincoln NE 68583-0817; jlindquist1@unl.edu

Single-leaf CO$_2$ assimilation rate under saturating light (C$_A$) varies as a function of leaf nitrogen content per unit leaf area (N$_L$). Measured C$_A$ for many crop species also declines with developmental stage. Because these relationships may have strong implications for crop–weed competition, a field experiment was conducted to quantify corn and velvetleaf C$_A$ as influenced by leaf N$_L$ and stage of development. Crop and weed C$_A$ were measured on the most recent fully expanded leaves of plants grown in four nitrogen (N) application treatments. Both corn and velvetleaf C$_A$ increased with increasing N$_L$, up to about 1.5 g N m^{-2}. Corn and velvetleaf N$_L$ values were rarely less than 0.75 g N m^{-2}, indicating that both species may restrict leaf growth in order to maintain constant N$_L$ under conditions of limited soil N. Corn C$_A$ declined to half its maximum by physiological maturity, whereas velvetleaf C$_A$ only declined 18%. Although velvetleaf C$_A$ was considerably lower than that of corn, the difference decreased as the growing season progressed. Because corn leaf production is complete by anthesis and velvetleaf continues vegetative production throughout its life cycle, velvetleaf will produce relatively greater quantities of biomass late in the season, which may increase competition for light.

Nomenclature: Velvetleaf, *Abutilon theophrasti* Medik. ABUTH; corn, 'Pioneer 3489 and 33A14', *Zea mays* L.

Key words: Photosynthesis, rubisco, competition.

A relationship between crop productivity and nitrogen (N) supply is commonly used for fertilizer management schedules (Evans 1987). Because 50 to 80% of the N in plant leaves is found in photosynthetic proteins (Hikosaka and Terashima 1995; Makino and Osmond 1991; Sage and Pearcy 1987), a strong correlation between light-saturated CO$_2$ assimilation rate (C$_A$, µmol CO$_2$ m^{-2} s^{-1}) and N content per unit leaf area (N$_L$, g N m^{-2}) is commonly observed (Ampong-Nyarko and DeDatta 1993; Hilbert et al. 1991; Hunt et al. 1985; Marshall and Vos 1991; Muchow 1990; Sage and Pearcy 1987; Sage et al. 1990; Sinclair and Horie 1989). Increased leaf N concentration in response to N supply will, therefore, promote greater photosynthetic rates (Hasegawa and Horie 1996) and plant productivity. This relationship greatly benefits crop productivity, but also has strong implications regarding crop–weed competition relationships under conditions of limited N supply.

Sinclair and Horie (1989) reviewed a number of studies that showed a high correlation between C$_A$ of crop leaves and their N$_L$. They proposed that the relationship could be quantified using the equation:

$$C_A = A_m((2/(1 + exp(-a(N_L - b)))) - 1) \qquad [1]$$

where A$_m$ represents the asymptote of C$_A$ at high N$_L$, *b* represents the N$_L$ below which C$_A$ becomes negative, and *a* is a species-specific shape coefficient. Sinclair and Horie (1989) quantified this relationship for corn using data from Wong et al. (1985) and from their own experiment, in which C$_A$ was measured on solution-culture or pot-grown corn during early developmental stages. Lindquist and Mortensen (1999) quantified the relationship between C$_A$ and

N$_L$ for field-grown corn and velvetleaf during corn-grain fill and obtained a smaller estimate of A$_m$ than that reported by Sinclair and Horie (1989).

Leaf N content typically declines during later stages of plant development (Lindquist and Mortensen 1999). Effects of this decline on C$_A$ must be accounted for within our plant growth models to accurately predict dry matter production (Peng et al. 1995), leaf area development, and crop–weed competition. Hasegawa and Horie (1996) suggested that an ontogenetic decline in C$_A$ occurs as a consequence of reduced N$_L$ as well as factors such as the quantity and activation state of carboxylating enzymes, nonstructural carbohydrate concentration, and sensitivity of stomatal function. If the latter factors affect C$_A$ independent of N$_L$, then a decline in C$_A$ at later development stages may be expected, regardless of N$_L$. Therefore, the smaller estimate of corn A$_m$ observed by Lindquist and Mortensen (1999) may be the result of these ontogenetic changes in the overall physiological efficiency within the leaves of these species. Hasegawa and Horie (1996) proposed a relationship similar to that found in Equation [1] but allowed A$_m$ to vary with development stage (DVS) of the crop:

$$A_m = A_{max} - c(DVS)$$

where A$_{max}$ is the absolute maximum observable C$_A$ and *c* defines the rate at which A$_m$ declines with development stage. The objectives of this research were to quantify field-grown corn and velvetleaf CO$_2$ assimilation rate under full-sun conditions as influenced by leaf N and stage of development.

Materials and Methods

Field experiments were conducted under rain-fed conditions in 1998 and 1999 at the University of Nebraska Agricultural Research and Development Center near Mead, NE. Soil at the site was a Sharpsburg silty clay loam (fine, smectitic, mesic, Typic Argiudoll) with 3.5% organic matter. The field was previously cropped with corn and disked in the spring followed by field cultivation for seedbed preparation. The experiment was designed as a randomized complete block with four replications; treatments included four N application rates (0, 45, 90, and 180 kg N ha^{-1}) and corn (Pioneer 3489 and 33A14 in 1998 and 1999, respectively) or velvetleaf grown in monoculture or in mixture. Corn and velvetleaf were in rows spaced 0.76 m apart, the corn at five plants m^{-1} row (65,790 plants ha^{-1}) and velvetleaf at 2 or 10 plants m^{-1} row. Corn was seeded on May 11, 1998, and May 3, 1999, with a conventional planter.[1] Velvetleaf seed was obtained from the University of Nebraska Agricultural Research farm near Havelock, NE, the previous fall. Untreated seeds (100 times the desired density) were sown directly into all corn rows with a push planter immediately after planting the crop. Crop and weeds emerged simultaneously on May 17, 1998, and May 16, 1999. Each experimental unit was 5 by 13 m, and N was applied as NH_4NO_3 granules using a calibrated wheel-driven fertilizer spreader on May 13, 1998, and May 3, 1999. Velvetleaf treatment densities were established by hand thinning beginning on June 5, 1998, and May 25, 1999. Weeds other than velvetleaf were removed by hand as needed and by interrow cultivation on June 25, 1998, and June 15, 1999. Corn began anthesis on July 10, 1998, and July 19, 1999, and maturity was reached on September 8 in both years. The zero N treatments were approximately 3 d later in their development than all other treatments.

Instantaneous light-saturated leaf CO_2 assimilation rate (C_A) was measured on individual leaves of corn and velvetleaf under full-sun conditions (> 1800 µmol m^{-2} s^{-2}) using a portable photosynthesis system[2] at least seven times in each year (36, 44, 50, 58, 63, 78, and 92 d after planting [DAPßqb; in 1998 and 45, 53, 59, 65, 87, 103, 108, and 120 DAP in 1999). Because only 20 to 25 measurements of each species were possible at each sampling date, it was not possible to sample one leaf of each species from each subplot. Therefore, measurements were taken on randomly selected, recently fully expanded and fully illuminated leaves in as many treatments as possible within two blocks of the experiment at each sampling date. A 1.0-L chamber with restrictors to maintain constant leaf area was clamped across the entire width of a corn leaf approximately 20 cm from the leaf tip, where the midrib was not large enough to cause chamber leakage. For velvetleaf, the chamber was clamped across a portion of the leaf or the entire leaf, depending on leaf size. All measurements were taken between 10:00 A.M. and 2:00 P.M. central U.S. standard time. Following each measurement, the leaf was clipped to include approximately 30 cm of the distal portion of the corn leaf, or the entire velvetleaf lamina. Each leaf was measured for area, dried, weighed, and N concentration ([N], g kg^{-1}) was determined. Leaf N_L (g N m^{-2}) was obtained from [N] and specific leaf area (cm^2 g^{-1}).

Daily weather data were obtained through the High Plains Climate Center from an automated weather station

TABLE 1. Average daily air temperature (degrees C) and precipitation (mm) in 1998 and 1999, and the 30-yr normal. Data were obtained from the High Plains Climate Center; the normals reported were for Ashland, NE.

Month	Air temperature			Precipitation		
	1998	1999	Normal	1998	1999	Normal
		C			cm	
April	10.6	10.6	10.7	4.7	13.3	6.7
May	18.8	16.1	16.5	9.6	14.0	10.8
June	21.0	20.8	21.9	15.1	12.1	9.8
July	25.0	26.2	24.9	9.3	5.3	7.8
August	23.9	22.3	23.4	14.8	7.5	9.8
September	21.7	17.3	18.3	2.1	6.7	8.2

approximately 0.5 km from the experimental field (Table 1). Thermal units accumulated from emergence were calculated using a base temperature of 10 C for both species. Thermal units to anthesis and maturity were 583 and 1,419 in 1998, respectively, and 716 and 1,432 in 1999, respectively. Development stage at time of measurement was quantified using a dimensionless scale: 0 (emergence), 1.0 (corn anthesis or velvetleaf first flower), and 2.0 (corn physiological maturity or complete velvetleaf leaf dry down). Intermediate stages were determined from the number of thermal units accumulated between emergence and the sampling date (Lindquist and Mortensen 1999). The relationship between leaf appearance and thermal units accumulated from emergence is approximately linear during vegetative corn growth (Muchow and Carberry 1989). Therefore, vegetative growth stage can be inferred directly from DVS and thermal units to anthesis, when corn had an average of 19 leaves.

Analysis of variance (ANOVA) (PROC GLM[3]) was conducted to evaluate the effect of N rate and mixture treatment on C_A, N_L, and [N]. Because not all treatments could be measured at every sampling time, this analysis was conducted with all sampling dates pooled to provide sufficient degrees of freedom. Nonlinear regression analysis (PROC NLIN[3]) was conducted to quantify the relationship between C_A and N_L and in relation to DVS. Measured C_A was regressed on N_L and DVS using Equations 1 and 2. Dummy variables were initially included in the model (Swinton et al. 1992) to evaluate whether parameter estimates varied between years. If the approximate 95% confidence interval of a parameter estimate included zero, the null hypothesis that the parameter did not differ from zero was accepted and the parameter was removed from the analysis.

Results and Discussion

Average daily air temperature and precipitation were close to the 30-yr norm during the growing season in both 1998 and 1999 (Table 1). Optimal temperature for corn CO_2 assimilation rate ranges between 28 and 40 C (de Wit et al. 1978). Mean temperatures during measurements were below 38 C for all but one sampling time in 1999 (July 29, DVS = 1.26), when mean temperature during sampling was 42 C. Therefore, measurements made at that sampling time were not used in the statistical analyses, but are plotted in Figure 1.

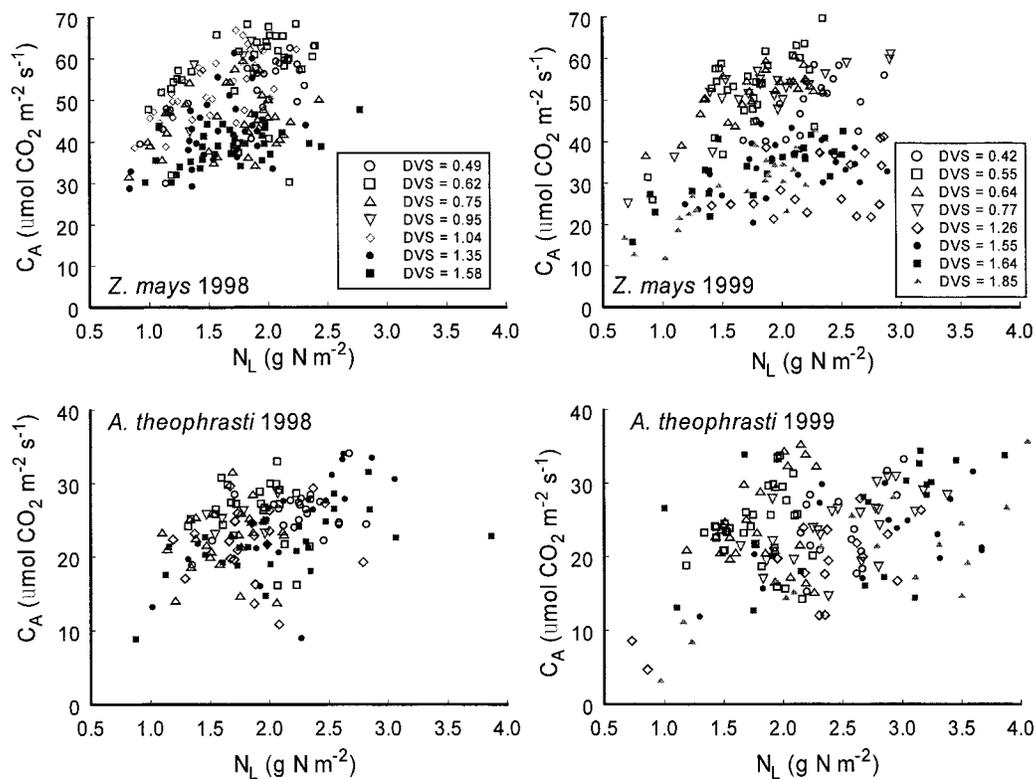Year and treatment did not interact to affect mean corn and velvetleaf C_A, N_L, and [N] when pooled across all sam-

FIGURE 1. Light-saturated CO_2 assimilation rate (C_A) in corn (*Zea mays* L.) and velvetleaf (*Abutilon theophrasti*) as a function of leaf nitrogen content (N_L) and development stage (DVS) in 1998 and 1999. Development stage at the time of sampling was identical between corn and velvetleaf in each year, using a dimensionless scale based on accumulated thermal units.

pling times (results not shown). Values also did not differ between years for either species. Corn C_A was constant across all treatments where N was applied (47.3 μmol CO_2 m^{-2} s^{-1}) but was lower when no N was applied (41.3 μmol CO_2 m^{-2} s^{-1}). Corn N_L and [N] increased with each increase in N application. Crop–weed mixture treatments did not affect corn C_A, N_L, or [N]. Mean velvetleaf C_A did not vary across any treatment (23.4 μmol CO_2 m^{-2} s^{-1}), based on ANOVA. Velvetleaf N_L and [N] increased with increasing N application but were lower (0.048 g N g^{-1} biomass and 2.0 g N m^{-2} leaf) in mixture compared to similar values in monoculture (0.050 g N g^{-1} biomass and 2.3 g N m^{-2} leaf). Similar response of both N_L and [N] across treatments indicated that neither species adjusted specific leaf area to compensate for changes in available N in leaves. Interference between corn and velvetleaf did not influence corn C_A, N_L,

or [N] but reduced N_L and [N] in velvetleaf, indicating that corn may be a better competitor for soil N. However, lack of an interaction between N application and mixture treatments on velvetleaf N_L or [N] indicates that the effect was constant across N application treatments. In other words, ability of corn to compete for soil N did not increase or decrease with varying N supply.

Estimates of the four parameters in Equations 1 and 2 did not vary between years for either corn or velvetleaf (results not shown), so data were pooled across years for subsequent analyses. However, for completeness of presentation, Figure 1 shows results separately for each year. Corn C_A varied as a function of both N_L and DVS (Figure 1, Table 2). The absolute maximum CO_2 assimilation rate (A_{max}) of corn was 63.8 μmol CO_2 m^{-2} s^{-1} (Table 2), which is somewhat greater than the 51, 52, and 57 μmol CO_2 m^{-2} s^{-1}

TABLE 2. Parameter estimates (± standard error), coefficient of determination (r^2) values, residual mean square error (RMSE) values, and P values for the relationship between full-sun CO_2 assimilation (C_A, μmol CO_2 m^{-2} s^{-1}) and leaf nitrogen (N_L, g N m^{-2}) in corn and velvetleaf, as affected by development stage in 1998 and 1999.[a,b]

	Parameter estimate						
Species	a g^{-1} m^2	b g N m^{-2}	A_{max} μmol CO_2 m^{-2} s^{-1}	c μmol CO_2 m^{-2} s^{-1} DVS^{-1}	r^2	RMSE	n	P value
Corn	3.68 (0.67)	0.45 (0.09)	63.8 (1.19)	15.54 (0.96)	0.51	66.4	373	< 0.0001
Velvetleaf	1.45 (0.26)	NS	29.5 (1.43)	2.80 (0.82)	0.06	28.6	299	< 0.0001

[a] The relationship was described by the equation $C_A = A_m((2/(1 + exp(-a(N_L - b)))) - 1)$, where $A_m = A_{max} - c(DVS)$. A_m represents the asymptote of C_A at high N_L, b represents the N_L below which C_A becomes negative, and a is a species-specific shape coefficient; A_{max} is the absolute maximum observable C_A, and c defines the rate at which A_m declines with development stage. NS indicates that the parameter did not differ from zero at the 95% confidence level.

[b] Data were pooled over years.

values reported by Louwerse et al. (1990), Muchow and Sinclair (1994), and Sinclair and Horie (1989), respectively. The maximum decreased at a rate of 15.5 μmol CO_2 m^{-2} s^{-1} per DVS, so that A_m at high N_L was estimated at 40.6 μmol CO_2 m^{-2} s^{-1} (DVS = 1.5), which was consistent with the value of 40.5 ± 1.3 μmol CO_2 m^{-2} s^{-1} reported by Lindquist and Mortensen (1999). Estimates of parameters a and b of Equation 1 were within the range of those reported by Muchow and Sinclair (1994) and Sinclair and Horie (1989).

Corn N_L ranged from 0.6 to 2.8 g N m^{-2} (Figure 1), which contrasted greatly with the results reported by Sinclair and Horie (1989), in whose study N_L ranged between 0.25 and 1.0 g N m^{-2}. Corn leaf [N] ranged between 0.02 and 0.05 g N g^{-1} biomass, which was consistent with whole-plant leaf [N] reported by Lindquist and Mortensen (1999). The discrepancy between the N_L values reported here and those of Sinclair and Horie (1989) may have resulted if there were large differences in the quantity of N available for uptake in these studies or if the growth chamber–grown corn reported by Sinclair and Horie (1989) was sufficiently stressed to reduce capacity for N uptake. It is likely that the field soil in this study supplied greater quantities of N through mineralization than did the N-limited solution and sand culture reported by Sinclair and Horie (1989). However, corn also may be more efficient at N acquisition under field conditions than it is when roots are restricted in pots within a controlled-environment chamber. In either case, because corn N_L in this experiment was rarely less than 1.0 g N m^{-2}, corn may restrict leaf growth in order to maintain constant N_L under conditions of limited soil N supply.

Velvetleaf C_A varied as a function of both N_L and DVS (Table 2, Figure 1). The estimate of velvetleaf A_{max} was 29.5 μmol CO_2 m^{-2} s^{-1} (Table 2), which was only slightly greater than the value of 24.6 ± 0.9 μmol CO_2 m^{-2} s^{-1} reported by Lindquist and Mortensen (1999). The maximum C_A declined at a rate of 2.8 μmol CO_2 m^{-2} s^{-1} per DVS, a much smaller decline than occurred in corn. Whereas the fit of Equations 1 and 2 was significant at P < 0.001, the r^2 of the regression was small because the magnitude of the response across N_L and DVS was not great. Velvetleaf C_A reached a maximum C_A at approximately 1.5 g N m^{-2}, and less than 15% of the leaves sampled had N_L less than 1.5 g N m^{-2}. Moreover, N_L values greater than 3.5 g N m^{-2} were commonly observed (Figure 1). Therefore, velvetleaf may be more prone to luxury consumption of N than corn. The small decline in C_A across DVS may be the result of high leaf turnover, which would allow the plant to maintain relatively high quantities and activity of carboxylating and other enzymes critical for photosynthesis in new leaves while recycling N and carbohydrates from old leaves as they senesce. Although the velvetleaf CO_2 assimilation rate was considerably lower than that of corn, it maintained a relatively constant C_A over time, whereas corn C_A decreased much more rapidly. Therefore, velvetleaf productivity remained much more constant as the growing season progressed, and the greater late-season growth of leaves may have conferred greater competition for light.

The estimate of b did not differ from zero for velvetleaf, indicating that this species may be capable of CO_2 assimilation at N_L levels approaching zero. However, N_L levels below 0.75 g N m^{-2} were not observed in the field, regardless of N supply. As with corn, this result indicated that velvetleaf may restrict leaf growth in order to maintain constant N_L under limited N supply. The effect of limiting N supply may be greater in velvetleaf than corn leaves because velvetleaf has greater leaf [N] throughout the growing season (Lindquist and Mortensen 1999). Moreover, because corn produces all of its leaf area by anthesis, whereas velvetleaf continues leaf production throughout its life cycle, the effects of soil N supply on total leaf area production may be expected to be greater for velvetleaf than for corn.

Sources of Materials

Acknowledgments

Literature Cited

Ampong-Nyarko, K. and S. K. De Datta. 1993. Effects of light and nitrogen and their interaction on the dynamics of rice-weed competition. Weed Res. 33:1–8.

de Wit, C. T., J. Goudriaan, H. H. van Laar, F.W.T. Penning de Vries, R. Rabbinge, H. van Keulen, L. Sibma, and C. de Jonge. 1978. Simulation of assimilation, respiration and transpiration of crops. Pudoc, Wageningen, The Netherlands: Simulation Monographs. 141 pp.

Evans, C. E. 1987. Soil test calibration. Pages 23–29 in J. R. Brown, ed. Soil testing: correlation, calibration, and interpretation. Special Publication 21. Soil Science Society of America, Madison, WI.

Hasegawa, T. and T. Horie. 1996. Leaf nitrogen, plant age and crop dry matter production in rice. Field Crops Res. 47:107–116.

Hikosaka, K. and I. Terashima. 1995. A model of the acclimation of photosynthesis in the leaves of C3 plants to sun and shade with respect to nitrogen use. Plant Cell Environ. 18:605–618.

Hilbert, D. W., A. Larigauderie, and J. F. Reynolds. 1991. The influence of carbon dioxide and daily photon flux density on optimal leaf nitrogen concentration and root:shoot ratio. Ann. Bot. 68:365–376.

Hunt, E. R., Jr., J. A. Weber, and D. M. Gates. 1985. Effects of nitrate application on *Amaranthus powelii* Wats. I. changes in photosynthesis, growth rates, and leaf area. Plant Physiol. 79:609–613.

Lindquist, J. L. and D. A. Mortensen. 1999. Ecophysiological characteristics of four maize hybrids and *Abutilon theophrasti*. Weed Res. 39:271–285.

Louwerse, W., L. Sibma, and J. van Kleef. 1990. Crop photosynthesis, respiration, and dry matter production in maize. Can. J. Plant Sci. 74:479–484.

Makino, A. and B. Osmond. 1991. Effects of nitrogen nutrition on nitrogen partitioning between chloroplasts and mitochondria in pea and wheat. Plant Physiol. 96:355–362.

Marshall, B. and J. Vos. 1991. The relation between the nitrogen concentration and photosynthetic capacity of potato (*Solanum tuberosum* L.) leaves. Ann. Bot. 68:33–39.

Muchow, R. C. 1990. Effects of leaf nitrogen and water regime on the photosynthetic capacity of kenaf (*Hibiscus cannabinus* L.) under field conditions. Aust. J. Agric. Res. 41:845–852.

Muchow, R. C. and P. S. Carberry. 1989. Environmental control of phenology and leaf growth in a tropically-adapted maize. Field Crops Res. 20:221–236.

Muchow, R. C. and T. R. Sinclair. 1994. Nitrogen response of leaf photosynthesis and canopy radiation use efficiency in field-grown maize and sorghum. Crop Sci. 34:721–727.

Peng, S., K. G. Cassman, and M. J. Kropff. 1995. Relationship between

leaf photosynthesis and nitrogen content of field grown rice in tropics. Crop Sci. 35:1627–1630.

Sage, R. F. and R. W. Pearcy. 1987. The nitrogen use efficiency of C3 and C4 plants. II. Leaf nitrogen effects on the gas exchange characteristics of *Chenopodium album* L. and *Amaranthus retroflexus* L. Plant Physiol. 84:958–963.

Sage, R. F., T. D. Sharkey, and R. W. Pearcy. 1990. The effect of leaf nitrogen and temperature on the CO_2 response of photosynthesis in the C3 dicot *Chenopodium album* L. Aust. J. Plant Physiol. 17:135–148.

Sinclair, T. R. and T. Horie. 1989. Leaf nitrogen, photosynthesis, and crop radiation use efficiency: a review. Crop Sci. 29:90–98.

Swinton, S. M., D. D. Buhler, F. Forcella, J. L. Gunsolus, and R. P. King. 1994. Estimation of crop yield loss due to interference by multiple weed species. Weed Sci. 42:103–109.

Wong, S., I. R. Cowan, and G. D. Farquhar. 1985. Leaf conductance in relation to rate of CO_2 assimilation. I. Influence of nitrogen nutrition, phosphorous nutrition, photon flux density, and ambient partial pressure of CO_2 during ontogeny. Plant Physiol. 78:821–825.