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Leaf area relationships to plant vegetative characteristics in cotton (*Gossypium hirsutum* L.) grown in a temperate sub-humid environment

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Abstract

Measurement or estimation of leaf area is essential for understanding crop responses to experimental treatments. The objective of this study was to develop regression models for estimating leaf area of field-grown cotton (*Gossypium hirsutum* L.) from measurements of leaf dry weight (LDW), vegetative components (stems and leaves) dry weight (VDW) and plant height (PH). Three cotton cultivars (Deltapine 25, Sahel and Siokra 324) with different leaf morphologies were grown under varying growth conditions created by four different planting dates in a temperate sub-humid environment (Gorgan, Iran). Leaf area, LDW, VDW and PH were measured at one month after emergence, squaring, flowering, bolling, boll opening and second harvest. Data set for validation was collected during growing season of 2003 in different experiments. Measured leaf area ranged from 170 to 8167 cm^2 $plant^{-1}$. Different regression models were examined for describing leaf area relationships to LDW, VDW and PH. It was found that the power function gives the best fit in terms of R^2 and root mean square of error (RMSE). Cultivar differences were not significant and a general equation was adequate for all the three cultivars. LDW and VDW provided good estimation of leaf area. However, PH was not a good predictor of leaf area. It was concluded that cotton leaf area can be estimated or simulated as a function of LDW or VDW with reasonable accuracy.

***Keywords*:** Leaf area index; Power function; Leaf dry weight; Plant height; Cotton cultivars.

Introduction

Leaf area is a determinant factor in radiation interception, photosynthesis, biomass accumulation, transpiration and energy transfer by crop canopies. It is also important with respect to crop-weed competition and soil erosion (Jonckheere et al., 2004). Therefore, leaf area is measured in many different studies and its accurate measurement is necessary for understanding crop responses to experimental treatments.

Measurement of leaf area in crops like cotton with various types of leaf area meters is difficult, labor-intensive and costly because there is much variation in number, size and shape of leaves (Reddy et al., 1989). On the other hand, measuring instruments are very expensive and often not available in developing countries and remote research stations. When available, these instruments are prone to large errors as a result of incorrect use that may lead to inconclusive results (Daughtry and Hollinger, 1984; de Jesus et al., 2001). These problems have been recognized by many researchers, who have developed less expensive and/or alternative, indirect methods (e.g., Johnson, 1967; Wendt, 1967; Ma et al., 1992). Indirect methods are based on the assumption that mass and size dimensions of different plant parts are allometric (Gardner et al., 1985). These indirect methods may increase precision of leaf area determination where sample or leaf size are difficult to handle (Ma et al., 1992) and can reduce the overall sampling effort necessary to estimate leaf area (Lieth et al., 1986).

Indirect methods of measuring leaf area can be classified as non-destructive and destructive methods. In non-destructive methods, leaf area is usually estimated by measuring the number, width or length of plant parts or whole plant, e.g., leaf width, length and number, branch length and number, and plant height. These measurements can be undertaken without cutting the plants. Non-destructive methods have been successfully applied for various crops such as cotton and castor (Wendt, 1967), sorghum (Shih et al., 1981), soybean (Lieth et al., 1986), pearl millet (Pyne et al., 1991), maize (Stewart and Dwyer, 1999) and sunflower (Bange et al., 2000). In some cases, these methods (except for plant height) are also time consuming and labor-intensive because they include many measurements.

Indirect, destructive methods estimate leaf area as a function of dry weight of plant parts or total above ground dry weight (Jonckheere et al., 2004). These measurements need cutting the plants. It has been reported that leaf and/or total dry weight have a close relationship to leaf area in wheat (Aase, 1978), barley (Romas et al., 1983), alfalfa (Sharrett and Baker, 1985), soybean (Lieth et al., 1986), pearl millet (Pyne et al., 1991), peanut (Ma et al., 1992) and several grasses (Retta et al., 2000). In cotton, Rhoads and Bloodworth (1964) and Johnson (1967) used specific leaf area (SLA, ratio of leaf area to leaf dry weight) to estimate leaf area. Reddy et al. (1989) stated that the accuracy of this method depends on the accuracy of the SLA determinations. SLA depends on temperature (Acock et al., 1979), solar radiation (Reddy et al., 1989) and carbon dioxide concentration (Lieth et al., 1986).

The objective of this study was to develop relationships between plant leaf area and plant vegetative characteristics in cotton cultivars grown under a wide range of planting dates. Plant vegetative characteristics that were used were leaf dry weight, dry weight of vegetative components (leaves and stems) and plant height.

Materials and Methods

Field experiment

A field experiment was conducted in 2000 at Hashem-Abad Research Station of Cotton Research Institute, Gorgan (latitude 36.85 ^{o}N, longitude 54.27 ^{o}E and 13 m asl), Iran. The site has a silty clay loam soil with a mean annual precipitation of 607 mm, mean solar radiation of 15.7 MJ m^{-2} d^{-1}, mean maximum temperature of 22.7 ^{o}C and mean minimum temperature of 12.6 ^{o}C. The climate is temperate sub-humid.

Three cultivars (Deltapine 25, Sahel and Siokra 324) differing in leaf morphologies were sown in a randomized complete block design with split-plot arrangement and four replications. Main plots were planting dates and subplots (5 × 11 m) were three cultivars. Sahel and Deltapine 25 have normal leaf type but Siokra 324 is an introduced cultivar from Australia and has okra leaf type. The okra leaf type (compared to normal leaf type) is characterized by cleft leaves and a relatively small leaf area per leaf. The four planting dates were 24 April, 9 and 25 May and 9 June 2000. The wide range of planting dates was selected to create different growth conditions. Plant population density was approximately 6.25 plants m^{-2}.

Average topsoil (0-30 cm depth) organic C, P and K were 1.4%, 23.2 ppm and 470 ppm, respectively. Fertilizer was applied at the time of planting at a rate of 46 kg N ha^{-1} and 96 kg P_2O_5 ha^{-1}. Irrigation was performed when required to avoid any water deficit based on soil moisture measurements. Trifluralin (2.5 L ha^{-1}) was used as pre-plant treatment for weed control. During growing season weeds were hand-controlled. Intensive insect control practices were employed so that pest effect was minimal.

Destructive plant samples were taken from each plot at one month after emergence, squaring, flowering, bolling, boll opening and second harvest. Emergence, squaring, flowering, bolling, boll opening and second harvest occurred at 9-13, 44-59, 63-75, 71-80, 112-119 and 166-212 days after planting, respectively, depending to planting date. Plant height (PH), leaf area (LA), leaf dry weight (LDW) and vegetative components (leaves + stems) dry weight (VDW) were obtained from a randomly chosen sample of three plants. Leaf area was measured with an area meter (Delta-T devices, Cambridge, England) and leaf and vegetative dry weight were determined after oven drying for 72 h at 72 ^{o}C. Dead and senesced leaves were excluded from measurements.

Data analysis

Different regression models were examined for describing leaf area relationships to LDW, VDW and PH. These models were selected from published works (Lieth et al., 1986; Payne et al., 1991; Ma et al., 1992) or determined after evaluating scatter plots of the data (Daniel and Wood, 1980; Draper and Smith, 1981; Montgomery and Peck, 1992). The best model was selected based on coefficient of determination (R^2) and root mean square of error (RMSE). Regression analysis showed that the power function ($Y = a X^{b}$) provides the best fit. The power function was fitted to data after logarithmic transformation [$\ln(Y)=\ln(a) + b \ln(X)$]. RMSE of estimation was calculated as:

$$RMSE = [\Sigma(P-O)^2/(n-1)]^{0.5}$$

where P is the predicted leaf area, O is the measured leaf area and n is the number of observation. Regression analysis was carried out using the Statistical Analysis System (SAS Institute, 1989). The power function first fitted to the data of each cultivar at each sampling time (developmental stage). Because there was no significant difference between developmental stages with respect to the coefficients of the power function, data of all the samplings were pooled and used in regression analysis, except for LA-VDW and LA-PH relationships at boll opening and second harvest (see sections 3.2 and 3.3). Therefore, we had data sets containing 96 (6 samplings × 4 replicates × 4 planting dates) or 64 observations (4 samplings × 4 replicates × 4 planting dates) for each cultivar.

Model evaluation

To test that how the models work on independent data sets (from the data used for model development), LA, LDW, VDW and PH data were gathered during growing season of 2003 at Hashem-Abad Research Station of Cotton Research Institute, Gorgan, Iran. These data were measured on cv. Sahel and B557 at different growth stages on various experiments including mixed cropping, irrigation methods, lysimeter, sowing date, foliar fertilization and weed control, and seed crops. Sampling procedure was the same as described above for year 2000 experiment. Leaf area predicted using the different models were plotted versus measured ones for validating the models. RMSE of prediction was also calculated.

Results

Relationship of leaf area to leaf dry weight

Leaf area ranged from 170 to 8167 cm^2 per plant across cultivars, corresponding to 1 to 49 g per plant leaf dry weight (Figure 1). The power function described well relationship between leaf area and leaf dry weight (Table 1; Figure 1). RMSE values were between 438 cm^2 $plant^{-1}$ for Siokra and 485 cm^2 $plant^{-1}$ for Delatpine, which were 15 to 16% of their corresponding means of leaf area (Table 1). R^2 values were high (0.98) and the same for all the three cultivars. There was no significant difference between cultivars based on confidence intervals for the coefficients of the power equation (Table 1). Therefore, one general equation ($LA = 125.3\ LDW^{1.078}$; $R^2 = 0.98$; RMSE = 465 cm^2 $plant^{-1}$) can be used for the three cultivars instead of individual equations.

Table 1. Parameter estimates for the power function ($Y=aX^b$) describing the relationship of leaf area to leaf dry weight. R^2 and RMSE values are also included.

Cultivar	a	b	R^2	RMSE
Deltapine 25	133.0±11.2	1.064±0.016	0.98	485
Sahel	128.0±11.0	1.076±0.015	0.98	470
Siokra 324	116.3±11.2	1.090±0.017	0.98	438
Pooled data	125.3±10.7	1.078±0.010	0.98	465

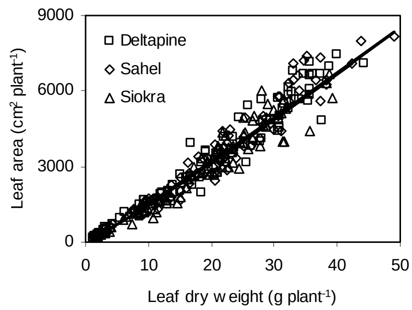


Figure 1. Relationship of leaf area to leaf dry weight described by the power function.

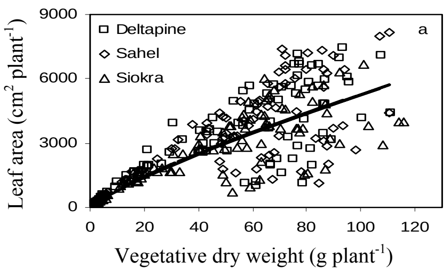
Relationship of leaf area to vegetative dry weight

Vegetative dry weight varied from 2 to 116 g plant^{-1} across cultivars (Figure 2). Deltapine and Sahel had slightly greater vegetative dry weight than Siokra at each sampling (data not shown). Figure 2 shows leaf area plotted as a function of vegetative dry weight. The variation of data around the regression line highly increased when vegetative dry weight was greater than 30 g plant^{-1} or leaf area greater than 2000 cm^2 plant^{-1} as a result of senescence of lower leaves of the canopy (Fig. 2a). The data are more scattered than that of leaf area-LDW relationship. R^2 values were greater than 0.83 for all cultivars, but RMSE values ranged from 1036 to 1361 cm^2 plant^{-1} (39 to 42% of the means), indicating that the relationships are not appropriate (Table 2).

When the data of the last two samplings (boll opening and second harvest) were discarded a good relationship was detected between leaf area and vegetative dry weight (Table 2; Fig. 2b). The relationship accounted for 98-99% of the variation in leaf area. RMSE values were 544 cm^2 plant^{-1} for Deltapine, 505 cm^2 plant^{-1} for Sahel and 437 cm^2 plant^{-1} for Siokra. There was no significant difference between cultivars for the coefficients of the power function (Table 2). As to LDW relationship, therefore, one general equation was adequate for all the three cultivars (LA = 99.8 VDW $^{0.935}$; R^2 = 0.98; RMSE = 489 cm^2 plant^{-1}), which is valid up to bolling stage.

Table 2. Parameter estimates for the power function ($Y=aX^b$) describing the relationship of leaf area to vegetative dry weight for whole growing season or till bolling stage. R^2 and RMSE values are also included.

Cultivar	a	b	R^2	RMSE
Whole growing season				
Deltapine 25	138.7±13.8	0.796±0.036	0.83	1294
Sahel	129.8±13.8	0.819±0.035	0.85	1361
Siokra 324	115.8±13.5	0.809±0.034	0.85	1036
Pooled data	127.3±12.0	0.809±0.020	0.84	1238
Up to bolling stage				
Deltapine 25	109.0±11.5	0.924±0.018	0.98	544
Sahel	101.4±11.2	0.946±0.015	0.99	505
Siokra 324	91.2±11.5	0.929±0.017	0.98	437
Pooled data	99.8±11.0	0.935±0.011	0.98	489



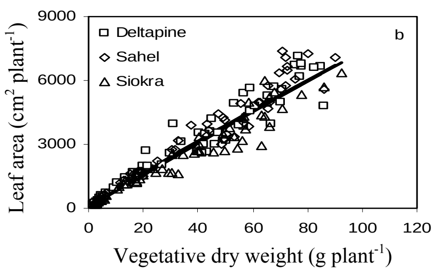


Figure. 2. Relationship of leaf area to vegetative dry weight described by the power function. (b) is based on data till bolling stage.

Relationship of leaf area to plant height

Plant height ranged from 17 to 122 cm (Figure 3). Large variation in leaf area was observed when plant height was greater than 70 cm, again due to senescence of lower leaves of the canopy (Fig. 3a). Although R^2 values were greater than 0.75, RMSE values were between 1249 and 1690 cm^2 $plant^{-1}$ (47 to 52% of the means)(Table 3). Therefore, these relationships are not enough precise for use.

Similar to vegetative dry weight, when the data of the last two samplings, i.e., boll opening and second harvest, were deleted a relatively good relationship was emerged (Table 3; Figure 3b). R^2 values increased to greater than 0.94 and RMSE values decreased to 24 to 26% of the means (Table 3). Differences between cultivars with respect to the coefficients of the power equation were not significant and one equation was adequate for all the three cultivars ($LA = 0.76\ PH^{1.925}$; $R^2 = 0.93$; RMSE = 692 cm^2 $plant^{-1}$).

Table 3. Parameter estimates for the power function ($Y=aX^b$) describing the relationship of leaf area to plant height for whole growing season or till bolling stage. R^2 and RMSE values are also included.

Cultivar	a	b	R^2	RMSE
Whole growing season				
Deltapine 25	3.15±1.48	1.563±0.093	0.75	1573
Sahel	2.44±1.47	1.618±0.090	0.77	1690
Siokra 324	1.80±1.47	1.637±0.089	0.78	1249
Pooled data	2.48±1.26	1.598±0.053	0.76	1516
Up to bolling stage				
Deltapine 25	0.94±1.27	1.902±0.059	0.94	679
Sahel	0.68±1.24	1.975±0.054	0.96	737
Siokra 324	0.60±1.24	1.933±0.053	0.96	618
Pooled data	0.76±1.16	1.925±0.037	0.93	692

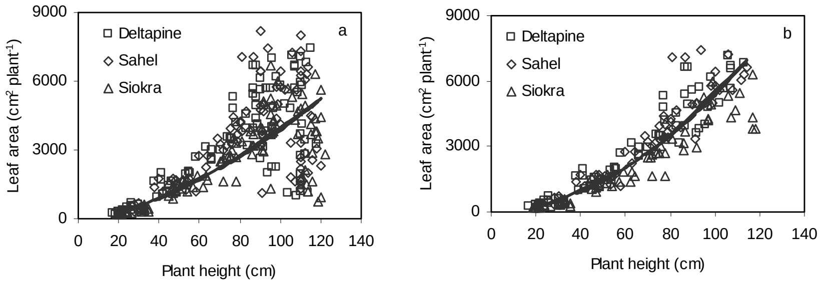


Figure 3. Relationship of leaf area to plant height described by the power function. (b) is based on data till bolling stage.

Model evaluation

Predicted values of leaf area from LDW, VDW and PH by the general regression models are presented in Fig. 4 versus the measured leaf area in various experiments. For independent data, measured leaf area varied between 73 and 5148 cm^2 plant $^{-1}$ with a mean of 2389 cm^2 plant $^{-1}$. Results showed that leaf area-LDW and leaf area-VDW models provided the reasonably good estimates of leaf area. RMSE of estimation was 503 cm^2 plant $^{-1}$ (21% of the measured mean) for leaf area-LDW and 401 cm^2 plant $^{-1}$ (17% of the measured mean) for leaf area-VDW relationship. Therefore, the relationships of leaf area to LDW and VDW, described by power functions, appear to be little affected by different experimental conditions. Leaf area-LDW tended to have slightly higher variability compared to leaf area-VDW (21% versus 17%) probably due to changes in specific leaf weight between seasons. However, leaf area-PH relationship showed a poor performance with a RMSE of 1517 cm^2 plant $^{-1}$ (63% of measured mean). Thus, based on independent data set, PH is not recommended for leaf area prediction.

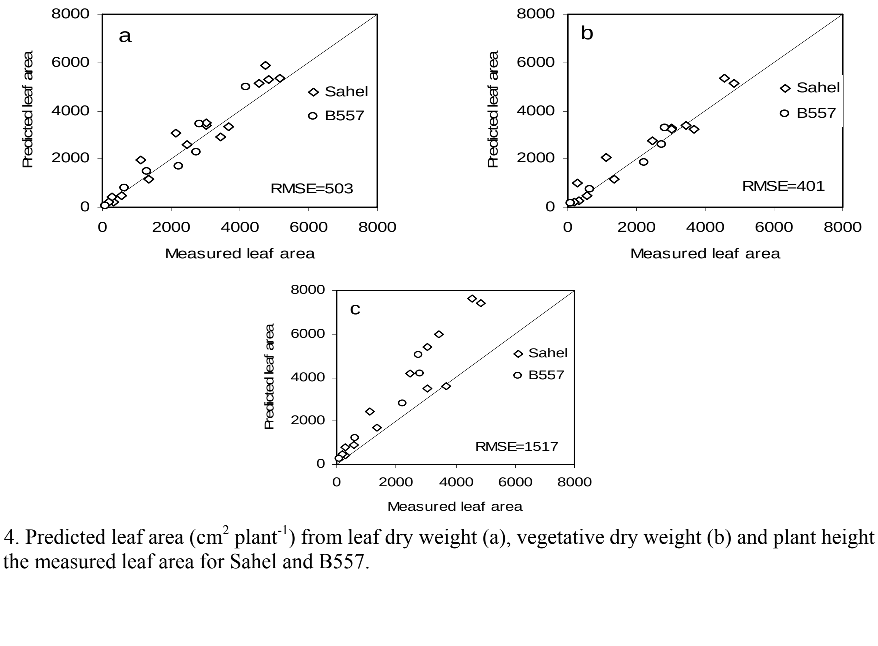


Figure 4. Predicted leaf area (cm^2 plant^{-1}) from leaf dry weight (a), vegetative dry weight (b) and plant height (c) versus the measured leaf area for Sahel and B557.

Discussion

We found that leaf area of cotton cultivars can be estimated using power functions from LDW and VDW. Cultivar differences were not significant and a generalized equation was adequate for all cultivars (Deltapine 25, Sahel, Siokra 324 and B557) used in this study with different leaf morphologies. Leaf area can be estimated from LDW with the function, $LA = 125.3\ LDW^{1.078}$ or from VDW with the function, $LA = 125.3\ VDW^{0.935}$.

The close relationship between leaf area and LDW found in this study is in agreement with those reported by Sharret and Baker (1985) in alfalfa, Pyne et al. (1991) in pearl millet, which showed that the power equation best described the relationship between leaf area and leaf dry weight. Awal et al. (2004) in oil palm found that leaf dry weight was strongly correlated with leaf area in both linear and non-linear regression. Some variability around the regression line may be due to environmental effects on SLA. When SLA values were calculated and examined, it was found that 49% of the variation in SLA is accounted for by mean air temperature between two samplings (Figure 5). This is in agreement with findings of Acock (1980) and Acock et al. (1979) that SLA increases with increase in temperature. Relationship between SLA and radiation was not considerable in the present study (data not shown), but Reddy et al. (1989) reported that 93% of variation in SLA is accounted for by radiation and SLA decreases with increase in radiation. SLA is also affected by growth stage and leaf maturity (Jonckheere et al., 2004). However, Reddy et al. (1989) reported that the variations in SLA with growth stage and leaf maturity were small to insignificant when variation due to light flux density was removed.

The relationship between VDW and leaf area found in this study is in agreement with findings of others; Sharrett and Baker (1985) in alfalfa, Lieth et al. (1986) in soybean and Ma et al. (1992) in peanut reported that leaf area can be successfully estimated from vegetative dry weight using a power function. However, Romas et al. (1983) and Retta et al. (2000) found that a simple, linear regression model between leaf area and vegetative dry weight is adequate to estimate leaf area.

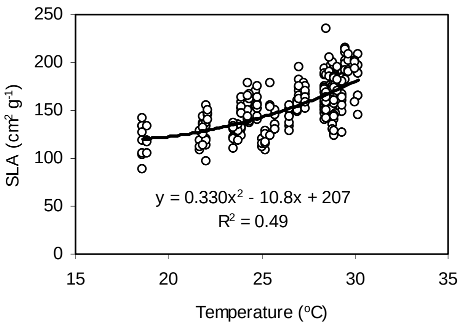


Figure 5. Relationship between specific leaf area (SLA) and mean air temperature. Data are pooled across cultivars.

PH did not provide reasonable estimates of leaf area and was not recommended for use. However, Lieth et al. (1986) found a power relationship between leaf area and plant height with R^2 of 0.97 and coefficient of variation of 24% similar to that we found for cotton.

Inappropriateness of PH compared to LDW and VDW is expectable, because PH responds to environmental factors differently.

In both LA-LDW and LA-VDW relationships, it seems that the variation around the regression line has increased with increasing LDW, VDW or leaf area (Figure 1 and Figure 2); this is because the variance of a variable tends to increase in proportion to the mean of the variable as noted by Ma et al. (1992).

The relationships of leaf area to LDW and VDW were established under different growth conditions in a temperate sub-humid environment created by choosing a wide range of planting dates and evaluated under various experimental conditions (treatments). The relationships between leaf area and LDW and VDW can also be used in simulation models of cotton. The applicability of the equations obtained in this study to other cultivars and/or environments should be tested.

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