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Research Article
Linear Model in the Estimate the Sunn (*Crotalaria juncea* L.) Leaf Area

[1]Matheus Langhi Alves, [1]Mariana Conceição de Souza, [1]Gustavo Henrique Gottardo Löff, [1]Rodrigo Araújo Marques, [1]Paulo Eduardo Teodoro, [1]Caio Cezar Guedes Corrêa, [2]Carlos Antonio da Silva Junior and [1]Francisco Eduardo Torres

[1]State University of Mato Grosso do Sul (UEMS), Aquidauana, Mato Grosso do Sul, 87020-900, Brazil
[2]Department of Forest Engineering, State University of Mato Grosso (UNEMAT), Alta Floresta 78580-000, Mato Grosso, Brazil

Abstract
Models to estimate leaf area according to the linear dimensions of leaves have been developed in several green manure crops. The aim of this work was to model and identify the best models for estimating the leaf area determined by digital photos in sun. The trial was installed in the experimental area of the State University of Mato Grosso do Sul-Unit University Aquidauana (UEMS/UUA), Brazil. During the growth and development of plants, randomly 200 expanded leaves of the lower, middle and superior portions were collected in order to obtain the plant canopy representation. These leaves were separated into two samples; the first with 150 leaves was used to generate mathematical models for measuring the leaf area and the second with 50 leaves was used to validate the generated equations and identify the best accuracy. At each collected leaf was measured the length and width. Subsequently, it was calculated the product of length×width. Then, the real leaf area for all 200 leaves was determined by digital photos. The linear model based on the product between length×width is the most appropriate to estimate sunn leaf area, because it had the highest Pearson linear correlation coefficient, lower mean square error and lower root mean square error.

Key words: Digital photo, leaf dimension, linear model, correlation, kurtosis

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Corresponding Author: Carlos Antonio da Silva Junior, Department of Forest Engineering, State University of Mato Grosso (UNEMAT), Alta Floresta 78580-000, Mato Grosso, Brazil

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Sunn (*Crotalaria juncea* L.) is a specie originated in India, with wide adaptation to tropical regions. The plants are shrubby, with upright and determined growth (Calegari *et al*., 1993). It is widely used as a green manure because of its nutrient cycling capacity and green mass production (Teodoro *et al*., 2015). In this sense, Torres *et al*. (2014) mentioned that sunn may lay between 150-165 kg ha^{-1} $year^{-1}$ of nitrogen in the soil, producing 10-15 t of dry matter, corresponding to 41 and 217 kg ha^{-1} of P_2O_5 and K_2O, respectively.

Leaf area quantification of a green manure species is important because it is related to the ability of solar radiation absorption, with transpiration and the fresh and dry mass production. Adami *et al*. (2008) proposed an alternative method using digital photos to estimate leaf area in intact and damaged soybean leaflets with advantages of lower cost and similar accuracy to the standard method, which uses the LI-COR equipment.

Therefore, it is important estimating the leaf area through indirect and more financially economical methods, as by using previously developed mathematical equations for each species. Mathematical equations are usually considered accurate, easy to use and enable quick implementation of the measures in the field, allowing successive assessments over the growth of particular species (Blanco and Folegatti, 2005).

For generating mathematical equations, models that describe the relationship between the linear dimensions of leaves (length, width or length×width product) and its total area are used. The M models to estimate leaf area according to the linear dimensions of leaves have been developed in several green manure crops such as jack bean (*Canavalia ensiformis*, Toebe *et al*., 2012), gray velvet bean (*Stizolobium cinereum*, Filho *et al*., 2012a) and radish (*Raphanus sativus*, Filho *et al*., 2012b).

Thus, the aim of this study was to model and identify the best models for estimating the leaf area determined by digital photos in sunn.

MATERIALS AND METHODS

The trial was installed in the experimental area of the State University of Mato Grosso do Sul-Unit University Aquidauana (UEMS/UUA), in the Municipality of Aquidauana (MS), located in the Brazilian Cerrado (or Savanna), comprising the coordinates 20°27'S and 55°40'W, with an average elevation of 170 m.

The soil was classified as Ultisol sandy loam texture, with the following chemical characteristics in the layer 0-0.20 m: pH (H_2O) = 6.2, Al exchangeable ($cmol_c$ dm^{-3}) = 0.0, Ca+Mg ($cmol_c$ dm^{-3}) = 4.31, P (mg dm^{-3}) = 41.3, K ($cmol_c$ dm^{-3}) = 0.2, organic matter (g dm^{-3}) = 19.7, V (%) = 45.0, m (%) = 0.0, sum of bases ($cmol_c$ dm^{-3}) = 2.3 and Cation Exchange Capacity (CEC) ($cmol_c$ dm^{-3}) = 5.1. The climate of the region according to the classification described by Köppen-Geiger is Aw (Savanna Tropical) with average annual rainfall of 1200 mm and maximum and minimum temperatures of 33 and 19°C, respectively.

Sunn seeds (cultivar, varieties or lines should be described!!) were sown manually on April 16, 2014 in experimental areas with 50 m^2. The spacing used was 0.45 m between rows at a density of 15 plants per meter linear. There was no base fertilization and coverage were performed. During the growth and development of plants, randomly 200 expanded leaves of the lower, middle and superior portions were collected in order to obtain the plant canopy representation. These leaves were separated into two samples: the first with 150 leaves was used to generate mathematical models for measuring the leaf area and the second with 50 leaves was used to validate the generated equations and identify the best accuracy.

At each collected leaf was measured the Length (L) and Width (W) with a millimeter ruler. Subsequently, it was calculated the product of length×width (L×W). Then, we determined the real leaf area for all 200 leaves. For this, each leaf was placed under a white sheet of paper and, subsequently, photographed with digital camera Sony, model DSC-W110®, disposed on a base perpendicular to 30 cm distance from the leaf using seven megapixels resolution. These 200 pictures (images) were individually processed with the Sigma Scan Pro v.5.0® software (Jandel Scientific, 1991) for determining the leaf area of each leaf using the digital photos method (Y).

With the division of the dataset in two samples (the first consisting of 150 leaves for generation and the second consisting of 50 leaves for validation of models) and with validation statistics of the following described models, it is possible infer about the quality of setting and, additionally, to evaluate if the models adequately estimate, overestimate or underestimate the leaf area.

For the data of L, W and L×W from first sample (150 leaves) the following parameters were calculated: maximum and minimum value, average, coefficient of variation (CV), kurtosis, asymmetry and after normality was verified through Kolmogorov-Smirnov test (p-value).

Subsequently, with 150 leaves the leaf area was modeled by digital photos (Y) according to the L or the W and/or the L×W, using the models: linear (Y = ax+b), quadratic ($Y = ax^2+bx+c$) and potency ($Y= ax^b$), totaling nine equations (three models x three independent variables).

Validation of the nine leaf area estimation models was based on 50 values estimated by the generated model ($\hat{Y}_i$) and the 100 values observed (Y_i) in the second sample. Then, we calculated the Pearson linear correlation coefficients (r) between $\hat{Y}_i$ and Y_i, Mean Square Error (MSE) and Root Mean Square Error (RMSE) through, respectively, the expressões:

$$\text{MSE} = \frac{\sum_{1=1}^{n}(\hat{Y}_i - Y_i)^2}{n}$$

$$\text{RMSE} = \sqrt{\frac{\sum_{1=1}^{n}(\hat{Y}_i - Y_i)^2}{n}}$$

where, $\hat{Y}_i$ are the estimated values of leaf area, Yi are the observed values of leaf area using the digital photos method, $\bar{Y}$ is the average of the observed values and n is the number of leaves (n = 50). The best models that estimate the sunn leaf area were those with r near to 1, MSE and RMSE near to 0.

RESULTS AND DISCUSSION

The wide variability of the leaves size (Fig. 1), observed by the amplitude of Length (L), of Width (W), Length×Width (L×W) and leaf area (Y), combined with to the high magnitude of the coefficient of variation (CV) (Table 1), it is important for the generation of models, because it allows its use in small, medium and large leaves. Therefore, this wide data set of Length (L), Width (W), Length×Width (L×W) and leaf area (Y) determined by digital photos (150 leaves) is suitable for the proposed study.

The measures of central tendency, variability, asymmetry, kurtosis and Lilliefors's test, in relation to characters L, W, L×W and Y in sunn, from 150 plants evaluated, data showed considerable distance to the distribution normal (p>0.05) (Table 1). However, according to the central limit theorem, even if the basic population is not normal, the distribution of the sample average will be approximately normal for samples greater than 30 observations (Fonseca and Martins, 1995; Bussab and Morettin, 2004). Given these considerations, in relation to normality can be inferred that the data from these traits provide credibility to proposed study.

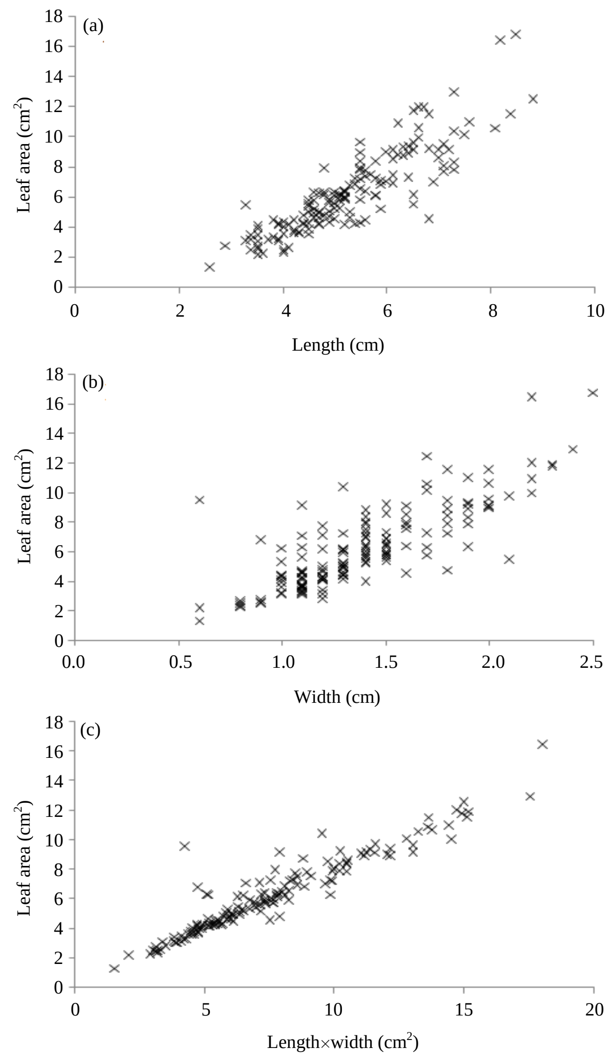

Fig. 1(a-c): Relationship between the Length (L), Width (W), L×W and leaf area measured on 150 leaves of sunn (*Crotalaria juncea* L.)

Table 1: Descriptive statistics for the variables leaf length (L), width (W), L×W and leaf area (Y) estimated at 150 leaves of sunn (*Crotalaria juncea* L.) grown in cerrado/pantanal ecotone

Statistic	Length (L, cm)	Width (W, cm)	L×W (cm^2)	Y-leaf area (cm^2)
Minimum	2.60	0.60	1.56	1.32
Mean	5.30	1.41	7.74	6.35
Maximum	8.80	2.50	21.25	16.77
CV (%)	23.70	27.26	45.42	43.95
Kurtosis[1]	2.80	3.02	4.14*	4.30*
Asymmetry[2]	0.40*	0.54*	1.00*	0.98*
p-value[3]	0.06	0.01	0.01	0.01

[1]Kurtosis statistically different from three by t-test at 5% probability, [2]Asymmetry statistically different from one by t-test test at 5% probability, [3]p-value of Kolmogorov-Smirnov normality test, CV: Coefficient of variation

The dispersion diagrams between the variables L, W, L×W and Y, shows patterns in the data that suggest the

Table 2: Models for determination of leaf area obtained by digital photos (Y), using the Length (L), Width (W) and the product of Length×Width (L×W) of leaves as independent variables (x) and the coefficient of determination (r^2) for each model, based on 150 leaves of sunn (*Crotalaria juncea* L.) grown in cerrado/pantanal ecotone

Dependent variable (x)	Model	Equation	r^2
	Linear	$\hat{Y} = 1.939x-3.933$	0.760
Length (L)	Quadratic	$\hat{Y} = 0.091x^2+0.921x-1.26$	0.765
	Potency	$\hat{Y} = 0.385x^{1,650}$	0.783
	Linear	$\hat{Y} = 5.807x-1.845$	0.640
Width (W)	Quadratic	$\hat{Y} = 1.990x^2-0.214x+2.395$	0.659
	Potency	$\hat{Y} = 3.893x^{1,281}$	0.630
	Linear	$\hat{Y} = 0.755x+0.501$	0.905
L×W	Quadratic	$\hat{Y} = 0.0001x^2+0.740x+0.560$	0.905
	Potency	$\hat{Y} = 0.928x^{0,938}$	0.906

Table 3: Validation of models based on the pearson linear correlation coefficient (r), Mean Square Error (MSE) and Root Mean Square Error (RMSE) based on the observed (Y_i) and estimated ($\hat{Y}_i$) leaf area of 50 leaves of sunn (*Crotalaria juncea* L.) grown in cerrado/pantanal ecotone

Dependent variable (x)	Model	R	MSE	RMSE
	Linear	0.9022*	2.6442	1.6261
Length (L)	Quadratic	0.9054*	2.5182	1.5869
	Potency	0.9056*	2.7058	1.6449
	Linear	0.8544*	3.2966	1.8156
Width (W)	Quadratic	0.8869*	2.7175	1.6485
	Potency	0.8645*	3.5416	1.8819
	Linear	0.9875*	0.4089	0.6394
L×W	Quadratic	0.9875*	0.4857	0.6969
	Potency	0.9873*	0.4508	0.6714

*Significant at 1% probability by the t-test with n-2 degrees of freedom

adjustment of nonlinear and linear models (Fig. 1). It can verify in Table 2 that the mathematical models adjusted from L×W had the highest coefficients of determination ($R^2 \geq 0.905$). Similar results were obtained for the cabbage (*Brassica oleracea*, Marcolini *et al*., 2005), cowpea (*Vigna unguiculata*, Lima *et al*., 2008), crambe (*Crambe abyssinica*, Toebe *et al*., 2010), potato at 50 days after emergence (*Solanum tuberosum*, Busato *et al*., 2010), forage turnip (*Raphanus sativus*, Filho *et al*., 2012b), jack bean (*Canavalia ensiformis*, Toebe *et al*., 2012), pigeonpea (*Cajanus cajan*, Filho *et al*., 2015a) and canola (*Brassica napus*, Filho *et al*., 2015b).

Table 3 shows the linear model ($\hat{Y} = 0.755x+0.501$) with based on L×W product presented higher Pearson correlation coefficient (r = 0.9875), lower mean square error (MSE = 0.4089) and lower root mean square error (RMSE = 0.6394), which provides greater efficiency in its use to estimate sunn leaf area. Similar results was verified by Marcolini *et al*. (2005), Lima *et al*. (2008), Toebe *et al*. (2010, 2012), Busato *et al*. (2010) and Filho *et al*. (2012a, b, 2015a, b).

However, if the researcher need more quickly in estimating the leaf area in sunn, can be used models generated from the measurement of L. In addition, for measuring only L, the researcher saves labor and resources.

CONCLUSION

Linear model based on the product between length×width is the most suitable for estimating the sunn leaf area, because it had the highest Pearson linear correlation coefficient, lower mean square error and lower root mean square error.

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