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Leaf area estimation of cassava from linear dimensions

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ABSTRACT

The objective of this study was to determine predictor models of leaf area of cassava from linear leaf measurements. The experiment was carried out in greenhouse in the municipality of Botucatu, São Paulo state, Brazil. The stem cuttings with 5-7 nodes of the cultivar IAC 576-70 were planted in boxes filled with about 320 liters of soil, keeping soil moisture at field capacity, monitored by puncturing tensiometers. At 80 days after planting, 140 leaves were randomly collected from the top, middle third and base of cassava plants. We evaluated the length and width of the central lobe of leaves, number of lobes and leaf area. The measurements of leaf areas were correlated with the length and width of the central lobe and the number of lobes of the leaves, and adjusted to polynomial and multiple regression models. The linear function that used the length of the central lobe $LA = -69.91114 + 15.06462L$ and linear multiple functions $LA = -69.9188 + 15.5102L + 0.0197726K - 0.0768998J$ or $LA = -69.9346 + 15.0106L + 0.188931K - 0.0264323H$ are suitable models to estimate leaf area of cassava cultivar IAC 576-70.

Key words: *Manihot esculenta* Crantz, leaf biometrics, statistical models, multiple regression.

INTRODUCTION

In Brazil, cassava (*Manihot esculenta* Crantz) is grown throughout the country with estimated production of 23.1 million tons in 2015. The southeast region contributed with 2.4 million tons, and the state of São Paulo stood out with a production equivalent to 54% of the total produced at this region (IBGE 2015). The IAC 576-70 is the main "sweet" cultivar planted in the State of São Paulo, occupying around 90% of the area, with high yields and excellent culinary qualities (EMBRAPA 2003).

Cassava is a perennial, shrubby plant with a tuberous root (Lorenzi et al. 2012). It has simple leaves inserted on the stem on disposal alternating-spiral, lobed and long petiolate (Conceição 1987). The leaves are photosynthetic organs responsible for the absorption of light energy, which is used to coordinate chemical reactions that are vital to plants.

Commonly, the leaf area is considered an important parameter for plant growth, because it is directly related to physiological processes such as photosynthesis, respiration and transpiration (Silva et al. 2008b). Leaf area can be estimated by destructive and non-destructive, direct or indirect methods (Marshall 1968).

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Destructive methods are those that require the removal of leaves from the plant at the time of evaluation, while nondestructive ones do not require the removal of the leaves, thus preserving the integrity of the plant. Direct methods are based on measurements made directly on the leaves, while indirect ones consist of known correlations between a measurable biometric variable and the real leaf area (Flumignan et al. 2008).

Among the commonly used methods, the non-destructive indirect one can be an alternative for estimating leaf area based on the regression relationship, using dimensions of the leaf such as length and width. This method has been used in various crops such as faba bean (Peksen 2007), potato (Silva et al. 2008a), cotton (Fideles Filho et al. 2010), sunflower (Aquino et al. 2011) and soybean (Richter 2014), allowing the continuity of the assessments in the same plant during its development cycle. In addition, it is a fast, accurate (Bianco et al. 2008) and low-cost technique.

Regression analysis is a statistical method, which uses the relationship between two or more variables, in such way that a variable can be estimated from another or other variables (Neter et al. 2005). Simple linear regression examines the relationship between two variables; however, a model with more than two regressor variables is called multiple regressions (Downing and Clark 2011). One of the methods to study models with more than two variables is known as best subsets regression, which provides the best-fitting regression models utilizing specified predictors.

In the literature, there are some indicators that are used to test the difference between the models and, thus the most appropriate one is chosen, such as the coefficient of determination (R^2), the adjusted coefficient of determination (R^2_{ajusted}), Mallows statistic (*Cp*), among others (Montgomery and Runger 2012). Floriano et al. (2006), with the objective of developing and selecting models to describe the growth in height of trees, used the coefficient of determination (R^2) and the *Cp* Mallows statistic, among other criteria for comparison of the models.

The use of mathematic models for estimating leaf area in cassava crops, based on leaf dimensions, has already been demonstrated by Schons et al. (2009), who evidenced the importance of this technique. However, there are no studies in the literature on mathematical functions that allow this measurement for the cultivar IAC 576-70. Thus, the objective of this study was to determine the most appropriate model to estimate leaf area of cassava IAC 567-70 using measures of length, width and numbers of lobes of the leaf.

MATERIALS AND METHODS

The experiment was conducted in a greenhouse in the municipality of Botucatu, state of São Paulo, Brazil (22º51' S; 48º26' W; altitude of 840 m). Before installation of the experiment, an analysis was carried out of the chemical and physical characteristics of the soil at 0-20 cm depth and of the soil water retention curve, to define the amount of water to be applied. The cultivar used was cassava IAC 576-70, whose shoot has green-purplish sprouts, green young stem, green-purple petiole, obovate leaf lobe and smooth and average height of the first ramification (Lorenzi 2012).

Planting was carried out in November 2014 with stem cuttings removed from the middle third of healthy plants, sectioned at right angles with the aid of a machete, producing stem cuttings with 0.20 m in length containing at least 5 to 7 nodes. A stem cutting was distributed in each of the 48 boxes with 320 liters of soil classified as medium texture Neosol (EMBRAPA 2006). They were covered with a soil layer of approximately 0.05 m. According to the chemical analysis of the soil, there was no need for fertilization at planting. After 40 days, topdressing fertilizing with urea (45% N) was performed at the rate of 40 kg of nitrogen per hectare, following the instructions of Lorenzi (1996) for the cassava crop.

The control of water in the soil was performed by puncture tensiometers – one was installed in each box at 0.20 m depth. The plants were inspected and irrigated daily, keeping soil moisture at field capacity.

At 80 days after planting, 140 leaves were randomly collected from the top, the middle third and base of the plant, considering the photosynthetically active and undamaged leaves. The leaves were packed in plastic bags and taken to the laboratory for measurement of length (L) and width of the central lobe (W) with the aid of a millimeter ruler (Fig. 1), followed by count of the number of lobes (N) and determination of leaf area (LA). The LAs were determined by using the leaf area integrator, model Li-3100C (Li-Cor, Lincoln, NE, USA).

Prior to the evaluation of the models, Pearson correlation coefficients (r) were estimated; they represent intensity of co-variation between the variables (Steell et al. 1997). The models of leaf area according to dimensions of the leaves were estimated from polynomial and multiple regression, considering LA as the dependent variable and L, W and N as independent variables. The leaf area was also related to the products of the variables: L x W (K), L x N (J), W x N (T) and L x W x N (H). We used linear regression and polynomial (quadratic) methods, as well as the multiple linear regression method (best subsets regression).

In the choice of linear and quadratic regressions, values of the coefficient of determination (R^2) and of the S (standard error) were analyzed. In the multiple linear regression, Mallows' Cp (Cp) was considered together with R^2, R^2_{ajusted} and the S model, which was the best model that showed the

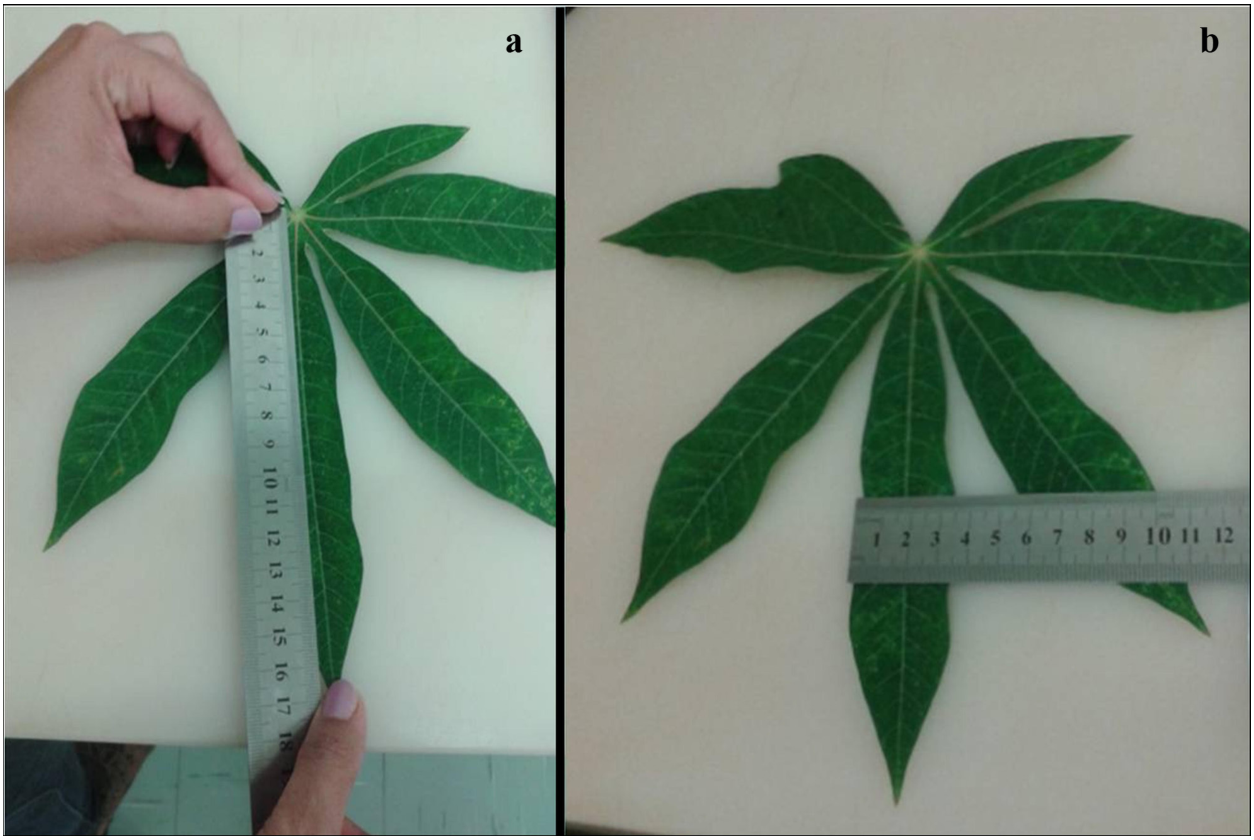

Figure 1 - Demonstration measurement length **(a)** and width **(b)** of the central lobe in cassava leaves IAC 576-70.

highest values of R^2 and R^2_{ajusted} with smaller values of S and Cp close to the number of predictors defined in the model.

Mallows' Cp (Equation 1) was used as an aid in choosing among multiple regression models. This method compares the precision and bias of the full model:

$$Cp=\frac{SQR}{QMR}-(n-2p) \qquad (1)$$

Where,

SQR is the sum of the squares of the residual error;

QMR is the mean square residual error;

n is the number of observations;

p is the number of predictors.

A model that features too many predictors can be relatively imprecise, while one with too few can produce biased estimates. Thus, the Mallow's Cp value closest to the number of predictors indicates that the model is relatively precise and has little variance in the estimation of regression coefficients.

Pearson's correlation analysis at 5% probability, polynomial regression and multiple regression were performed by using the software Minitab 16.

RESULTS AND DISCUSSION

It was found that the values of leaf area (LA) of cassava, IAC 576-70, ranged from 32.45 to 203.77 cm^2, averaging 119.63 cm^2. The length of the central lobe (L) of the leaves varied from 5.50 to 18.00 cm, with average values of 12.58 cm, while the width of the central lobe (W) of the leaves ranged from 1.10 to 4.30 cm, with mean values of 2.61 cm. The leaves presented (N) about 5 to 8 lobes (Table I).

The analysis of the independent variables studied showed values of standard deviation less than ± 2.85, indicating that the data present little dispersion compared with their average. However, the dependent variable showed a high standard deviation value (± 44.64), which may be explained by the difference in size of leaves collected throughout the plant (top, middle third and base) (Table I). Similar results to those obtained in this study were found by Silva et al. (2015), when estimating the leaf area of mangoes with non-destructive methods. According to Cock et al. (1979), Irikura et al. (1979), Tan and Cock (1979), the size of cassava leaves varies with the stage of plant development and changes in the branching pattern. However, factors such as solar radiation, temperature and water can also influence the leaf area of plants.

According to Ferreira (2000), Pearson's correlation coefficient (r) indicates the strength of the relationship between two variables and may range from -1 to +1. The negative or positive sign determines the direction of the correlation and the value suggests the degree of relationship between variables. The values -1 and +1 indicate the maximum correlation, and 0 means that there is no correlation between the variables. Pearson's correlation coefficients (r) between the leaf area and the variables L, K, J and H showed positive values of r ranging from 0.634 to 0.957; therefore, it is a significant correlation (Table II). However, the variable N showed a negative value of r (-0.008); thus, the correlation is not significant (p= 0.921).

The polynomial regression functions were carried from the variables that showed significant correlation. The adjustments obtained with the product of independent variables presented R2 below of 0.64 and standard error mean of 31.44 (Table III). These results disagree with those obtained by Busato et al. (2010), when estimating the leaf area of potatoes, and by Lima et al. (2008), who, when studying cowpea plants, observed that the highest coefficient of determination was in the function that used the product of the length and width of the leaves. The adjustment with the variable L had the lowest standard error (S= 12.62)

TABLE I
Minimum and maximum values, standard deviation and averages of leaf area (LA), length (L) of the central lobe, width (W) of the central lobe and the number (N) of lobes in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil.

Values	Dependent variable	Independent variables		
	LA (cm^2)	L (cm)	W (cm)	N
Minimum	32.45	5.50	1.10	5.00
Maximum	203.77	18.00	4.30	8.00
Standard deviation	± 44.64	± 2.85	± 0.71	± 0.91
Average	119.63	12.58	2.61	6.46

TABLE II
Correlation between the leaf area (LA) and length (L) of the central lobe width (W) of the central lobe, the number (N) of lobes and product length and width (K), length and number of lobules (J), width and number of lobes (T), length, width and number of lobes (H) in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil.

Dependent variable	Independent Variables	r	*p*	Significance
LA	L	0.957	0.001	*
LA	W	0.085	0.315	ns
LA	N	-0.008	0.921	ns
LA	K	0.634	0.000	*
LA	J	0.797	0.000	*
LA	T	0.095	0.260	ns
LA	H	0.670	0.000	*

r: Pearson's correlation coefficients;
p: level of significance;
ns: not significant;
* significant at 5% probability.

and highest coefficient of determination (R2= 0.92), indicating be the most appropriate linear model to estimate the leaf area of cassava (Table III).

According to the linear regression of leaf area in function of the central lobe length, there was less dispersion of data compared with the straight obtained, with $LA= -69.91114 + 15.06462L$ being the function that represents the leaf area in a satisfactory manner (Fig. 2). Partelli et al. (2006) also found that the length of the central veins is an appropriate parameter to estimate the leaf area of coffee plants grown in full sun with different ages. Schons et al. (2009), working with arrangements of cassava and maize plants, used the length of the major lobe of cassava leaves to estimate the leaf area.

Multiple regression analysis applied by the best subsets regression method consists of selecting the best combinations of independent variables from a set that possibly includes all the important variables. Thus, the study related to leaf area with L, K, J, H, generated regression models of best adjustments with the specified predictors. The most appropriate models were those which used three predictors in the function, because it showed R2 values equal to 0.917, adjusted R2 of 0.915, standard error of 13.0 and Mallow's Cp equal to the number of predictors (Table IV), suggesting that the functions $LA= -69.9188 + 15.5102L + 0.0197726K – 0.0768998J$ and $LA= -69.9346 + 15.0106L + 0.188931K – 0.0264323H$ are relatively accurate

TABLE III
Polynomial models for estimate of the leaf area as a function of length (L) of the central lobe, the length and width product (K), length and number of lobes (J), length, width and number of lobes (H), in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil.

Variables	S	Function	R^2
L	12.62	$LA= -69. 91114 + 15.06462L$	0.92
K	34.26	$LA= 2.88266 + 4.88301K - 0.0360K^2$	0.41
J	27.08	$LA= -12.778 + 1.62925J$	0.63
H	32.97	$LA= -4.70241 + 0.79114H - 8.42567x10^{-4}H^2$	0.46

S: standard error of the mean;
R^2: coefficient of determination.

and impartial. Zenginbal et al. (2006), researching a model to estimate leaf area in the tea culture in different cultivars used the multiple regression analysis, obtaining an equation with 99% accuracy.

The dispersion of the data of the measured leaf area and the leaf area estimated of the three main models discussed in this work are showed in the Figs. 3, 4 and 5. The methods used for estimating the leaf area showed reduced variability of the data, since they were close to the line corresponding to the 1:1 ratio. It was found that the sum of errors between the measured and estimated leaf area was 0.006, when using the linear function for L (Fig. 3), 0.059 to function with multiple variables L, K and J (Fig. 4), and 0.093 utilizing the variables L, K and H (Fig. 5), which proves the similarity of behavior between models.

The results suggest the use of any of the three functions to estimate the leaf area of cassava cultivar IAC 576-70, because of the precision found in these models. Considering that the measurements of the three variables (central lobe length, central lobe width and number of lobes) may require more time to obtain data and determine the leaf area, it is suggested that the linear function should be used for the length of the central lobe. However, it is believed that the use of three variables in the model can estimate the leaf area in cassava plants more precisely, when used in future experiments.

CONCLUSIONS

The leaf area of IAC 576-70 cassava is estimated by non-destructive methods, from the linear and linear multiple regression analysis.

The regression functions LA= -69.91114 + 15.0646L, LA= -69.9188 + 15.5102L + 0.0197726K – 0.0768998J, and LA= -69.9346 + 15.0106L + 0.188931K – 0.0264323H estimate accurately the leaf area of cassava, cultivar IAC 576-70, but the multiple linear regression model may be most appropriate since it uses a higher number of predictor variables.

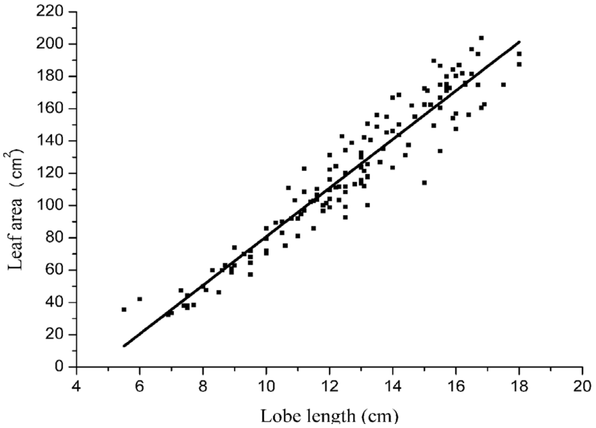

Figure 2 - Estimate of the leaf area (LA) in function of length (L) of the central lobe using the fit linear LA = -69.91114 + 15.06462L in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil.

TABLE IV

Application of the best subsets regression method between the leaf area (LA) and length (L) of the central lobe, the length and width product (K) length and number of lobes (J), length, width and number lobes (H) in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil.

Dependent variable	Independent variables	R^2	R^2_{ajusted}	S	Mallows' Cp
LA	L	0.916	0.916	13.0	-0.2
LA	J	0.634	0.632	27.1	464.6
LA	L, J	0.917	0.916	13.0	1.0
LA	L, K	0.917	0.915	13.0	1.5
LA	L, K, H	0.917	0.915	13.0	3.0
LA	L, K, J	0.917	0.915	13.0	3.0
LA	L, K, J, H	0.917	0.914	13.1	5.0

R^2: coefficient of determination;
S: standard error of the mean.

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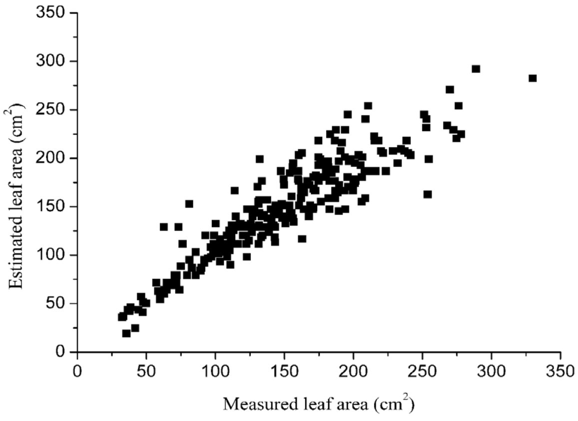

Figure 3 - Dispersion of data between the measured leaf area (cm^2) and the estimated leaf area (cm^2) using the function LA = -69.91114 + 15.06462L in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil. (LA = leaf area; L = lobe length).

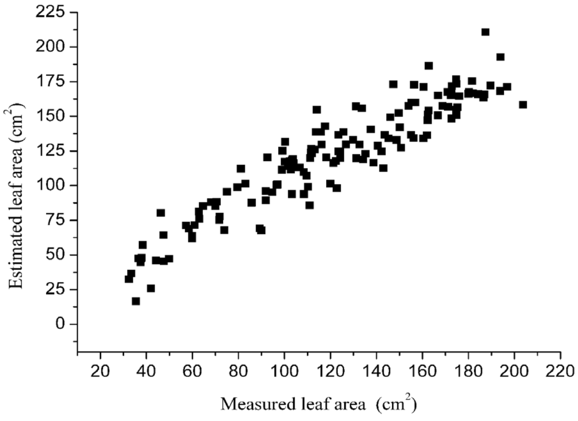

Figure 4 - Dispersion of data between the measured leaf area (cm^2) and the estimated leaf area (cm^2) using the function LA = -69.9188 + 15.5102L + 0.0197726K - 0.0768998J in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil. (LA = leaf area; L = lobe length; K = lobe length x lobe width; J = lobe length x number of lobes).

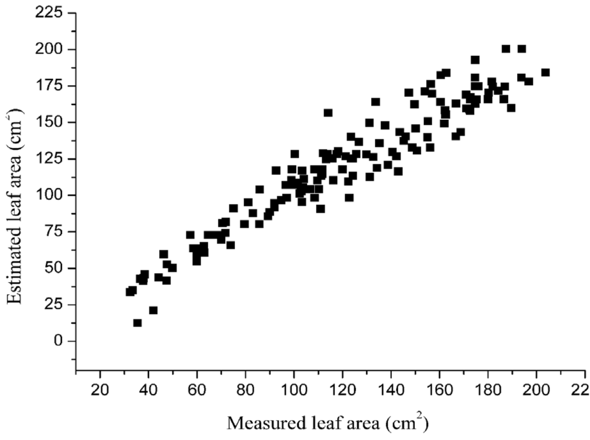

Figure 5 - Dispersion of data between the measured leaf area (cm^2) and the estimated leaf area (cm^2) using the function LA = -69.9346 + 15.0106L + 0.188931K - 0.0264323H in cassava leaves, cultivar IAC 576-70, at 80 days after planting in the region of Botucatu, SP, Brazil. (LA = leaf area; L = lobe length; K = lobe length x lobe width; H = lobe length x lobe width x number of lobes).

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