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Estimation of *Thunbergia grandiflora* leaf area from allometric models

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Abstract

Sky vine (*Thunbergia grandiflora* Roxb) is a vine with important structural components for forest environments. Studies on growth and development are necessary, because of the environmental and economic importance. The leaf area determination is essential for ecophysiological studies to understand the relationship of the plant with the environment. The objective of this work was to estimate an allometric equation to estimate the leaf area of *T. grandiflora* from linear dimensions. 200 leaves of different shapes and sizes were collected from adult plants and the length (L), width (W), the product between length and width (LW), and real leaf area (LA) were measured. The linear regression, linear without intercept, quadratic, cubic, power, and exponential models were used to estimate the equations. The criteria for determining the best model were higher determination coefficient (R^2), Willmott's agreement index (d), lower Akaike information criterion (AIC), the root of the mean error square (RMSE), and BIAS index closer to zero. The leaf area of *T. grandiflora* can be estimated satisfactorily by the equation $\hat{y} = 0.58*LW$.

Keywords: leaf dimension, non-destructive method, vines, sky vine

Introduction

Sky vine (*Thunbergia grandiflora* Roxb), is a perennial, semi-woody, robust and very vigorous vine. It is also known as blue thunbergia or Bengal clock vine. It belongs to the Acanthaceae family and is native to India, southern China and Myanmar (Meyer & Lavergne, 2004; Sartin et al. 2014). Its leaves are arranged oppositely and emerge from hairy stems, also known as pubescent petioles, with an oval or triangular shape, slightly jagged, dark green and shiny, 8-16 cm long (Caedo et al. 2014). This species is suitable for tropical or subtropical climates, with exposure to full sun to ensure its maximum development. Because of this, we use this species in covering pergolas, walls, and extensive fences, but also because of tolerating excessive sun radiation, and by it has high adherence and fast growth, characterizing it as a very rustic and vigorous species. Despite the preference for high temperatures, it tolerates mild temperatures as in some areas of southern Brazil, requiring little maintenance and regular pruning (Sartin et al. 2014).

Because it is a very vigorous and fast-growing species, its leaf density has great potential for application in ornamental gardens (Sartin et al., 2014). Also, it is a species that has attracted attention because it also has medicinal purposes helping to treat diseases such as hemorrhages ear diseases, stomach complaints, tonic for wounds (Mathew & Thomas, 2019; Oliveira et al. 2019). Because of the environmental and economic importance that this species presents and the lack of work on it, further studies are needed that seeks to evaluate its growth, development, and reproduction. Among these studies, the leaf area is important to determine the growth (Fascella et al., 2013; Abreu et al., 2015) and productivity (Walia & Kumar et al., 2017; Liu et al., 2017) of crops. This variable is directly related to the amount of light energy intercepted and the conversion of chemical energy in

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photosynthesis (Sala et al., 2015).

The leaf area can be measured using direct or indirect methods and, also, destructive or non-destructive methods (Santos et al., 2014; Keramatlou et al., 2015). The direct and destructive method is simple and precise, but requires more time and labor, as well as causing the destruction of leaves (Schmildt et al., 2014; Salazar et al., 2018). When measured by the direct and non-destructive method, electronic equipment is used to deduce a non-destructive reading of the plant, however, this equipment has a higher cost. The indirect and non-destructive method, on the other hand, allows successive assessments of the same plant quickly and accurately, using allometric equations from the length and width of the leaves (Ribeiro et al., 2020; Pinheiro et al., 2020). The objective of this work was to estimate an allometric equation to estimate the leaf area of *Thunbergia grandiflora* from linear dimensions.

Material and Methods

The research was carried out at the Unidade de Ensino, Pesquisa e Extensão (UEPE), Floricultura-Belvedere, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil. 200 leaves were collected from adult plants with different shapes and sizes. The leaves chosen were healthy, with no symptoms of attack by pests and diseases, or abiotic factors (Figure 1).

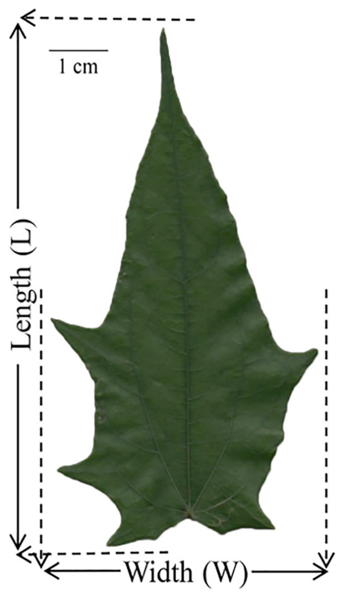

Figure 1. Maximum length (L) and width (W) of leaf of *Thunbergia grandiflora* used to estimate leaf area.

The maximum length (L) and maximum width (W) were measured from images scanned on a digital flatbed scanner (Epson Scan I365), with a known scale. The real leaf area (LA) was measured using the ImageJ® software (Ribeiro et al., 2018).

From the data of length (L), width (W), product (LW) and real leaf area (LA), a descriptive analysis was performed to obtain the maximum and minimum, mean, median, total amplitude, variance, standard deviation, standard error, coefficient of variation and coefficients of asymmetry and kurtosis.

Regression analyzes were performed to choose the equation that satisfactorily estimates the leaf area of *T. grandiflora*, using the following statistical models: linear, linear without intercept (0.0), quadratic, cubic, power and exponential. The Y value was estimated for X, whose values were those represented by length (L), width (W), or product (LxW). The best equations were chosen based on the highest determination coefficient (R^2) and Willmott's agreement index (d) (Willmott et al., 1981), lowest Akaike information criterion (AIC) (Akaike, 1974) and root of the mean square of error (RMSE) (Janssen & Heuberger, 1995), and BIAS index closer to zero (Leite & Andrade, 2002). The statistical analyzes were obtained with the software R (R Core Team, 2020).

Results and Discussion

The variation in the leaf length (L) was from 5.077 to 19.407 cm, with an average of 12.799 cm and 14.330 cm in amplitude (Table 1). The leaf width averaged 6.232, with values ranging from 2.447 to 10.868 cm and 8.421 cm in amplitude. The product between length and width (L.W) ranged from 12.423 to 206.286 cm^2, with an average of 83.532 and 193.863 cm in amplitude. The real leaf area (LA) had an average of 49.138 cm^2, ranging from 9.965 to 115.804 cm^2 and 105.839 in amplitude. Leaf variations are common in plants and when it comes to plants with a climbing habit, where there is a need for a tutor to support themselves, greater variations can occur because the leaves can move according to the need of the plant, such as the case of blue vine (Fiorello et al., 2020).

The leaf area varied from 9,965 cm^2 to 115,804 cm^2, with a higher percentage of leaves (37%) in the range of 30.1 cm^2 to 50.0 cm^2 (Table 2). Such an event is common in healthy plants and positive for this work because, due to its diversified leaf sizes, the analyzes will be with satisfactory accuracy and good data distribution (Shi et al. 2017).

The scatter plots between the pairs of variables L, W, and LA showed different relationships between them, suggesting adjustments of linear and non-linear models (Figure 2).

Table 1. Minimum, maximum, mean, median, standard deviation, standard error, and coefficient of variation (CV), asymmetry and kurtosis for length (L), width (W), length by width (L.W) and leaf area (LA) of *Thunbergia grandiflora* leaves.

Descriptive Statistic	L (cm)	W (cm)	L.W (cm²)	L.L (cm²)	W.W (cm²)	LA (cm²)
Minimum	5.077	2.447	12.423	25.776	5.988	9.965
Maximum	19.407	10.868	206.286	376.632	118.113	115.804
Mean	12.799	6.232	83.532	170.121	42.314	49.138
Total amplitude	14.330	8.421	193.863	350.856	112.125	105.839
Median	12.889	6.149	80.516	166.114	37.805	46.821
Variance	6.346	3.499	1491.483	4267.470	607.114	449.067
Standard deviation	2.519	1.871	38.620	65.326	24.640	21.191
Standard error	0.178	0.132	2.731	4.619	1.742	1.498
CV (%)	19.68	30.02	46.23	38.40	58.23	43.13
Asymmetry[a]	0.042	0.282	0.666	0.570	0.881	0.578
Kurtosis + 3[b]	2.898	2.495	2.887	2.894	2.969	2.936
Lilliefors	0.267[ns]	0.203[ns]	< 0.001**	0.012*	< 0.001**	0.002*

[a] Asymmetry differs from zero by t test at 5% probability; [b] Kurtosis differs from three by the t test at 5% probability; [ns] Not significant; * Significant at 5% probability; ** Significant at 1% probability.

Table 2. Percentage distribution of the real leaf area (LA) *Thunbergia grandiflora* leaves in relation to different size ranges.

LA (cm²)	(%)
[0 - 30.0]	19
[30.1 - 50.0]	37
[50.1 - 70.0]	27
[70.1 - 90.0]	14
[90.1 - 120.0]	3

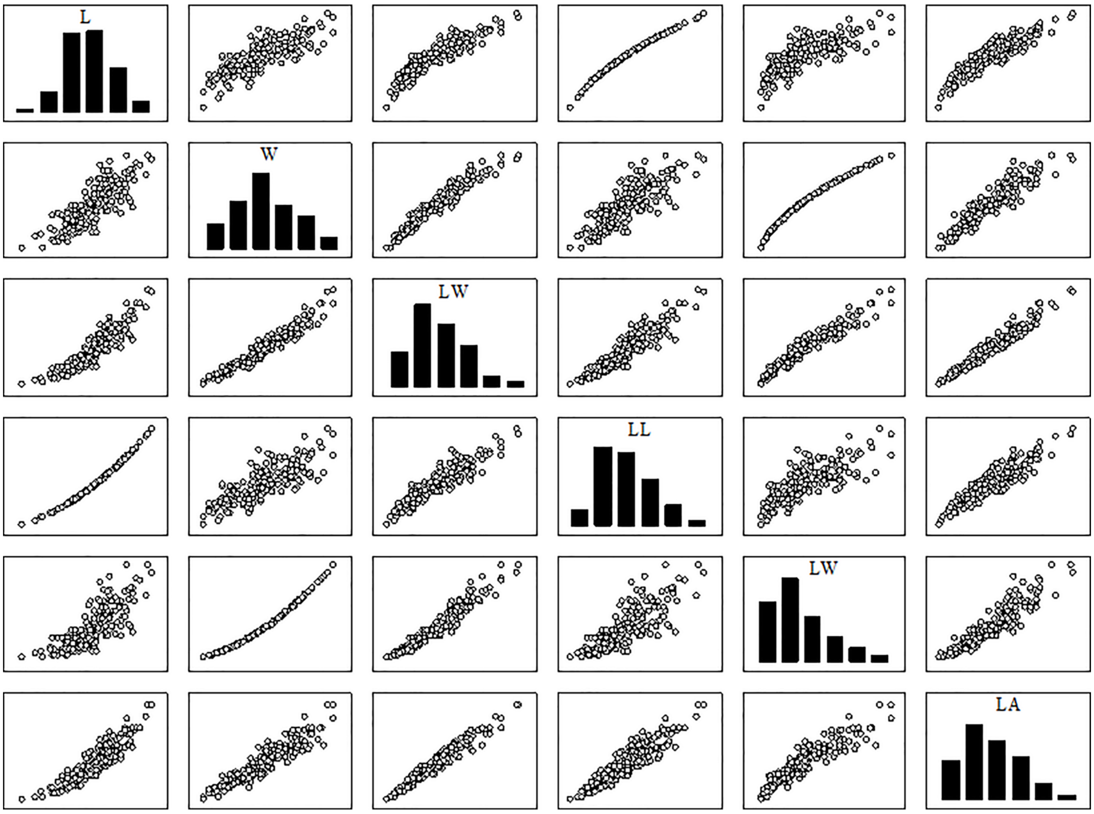

Figure 2. Histograms between leaf length (L), leaf width (W), product of length and width (LW) and leaf area (LA) of *Thunbergia grandiflora* leaves.

The linear models had R^2 ranging from 0.829 to 0.991, quadratic 0.843 to 0.949, cubic 0.842 to 0.948, potential 0.844 to 0.948 and finally the exponentials with lower R^2 ranging from 0.801 to 0.877 (Table 3). All models estimated the leaf area of *T. grandiflora* with determination coefficients (R^2) above 0.801. This indicates that 80.1% of the variation observed in the leaf area of this species is explained by the proposed model using linear dimensions of the leaf. Such variations were also observed in *Triticum aestivum* L. (Apolo-Apolo et al., 2020) and *Vitis vinifera* L. (Teobaldelli et al., 2020).

Table 3. Estimated equations, estimated standard error, coefficients of determination and Akaike information criterion as a function of linear measurements of *Thunbergia grandiflora* leaves.

Model	x	r	R^2	AIC	RMSE	MAE	*d*	Equation
Linear	L	0.911	0.829	1440.09	8.726	7.186	0.951	$\hat{y} = -48.92 + 7.66*L$
Linear	W	0.930	0.865	1393.24	7.761	5.935	0.963	$\hat{y} = -16.53 + 10.54*W$
Linear	L.W	0.974	0.948	1203.53	4.830	3.755	0.986	$\hat{y} = -4.51 + 0.53*LW$
Linear (0.0)	L.W	0.974	0.991	1199.48	2.367	1.865	0.987	$\hat{y} = 0.58*LW$
Linear	L.L	0.919	0.843	1422.64	8.353	6.748	0.956	$\hat{y} = -1.55 + 0.30*LL$
Linear	W.W	0.924	0.853	1409.82	8.090	6.232	0.959	$\hat{y} = 15.51 + 0.79*WW$
Quadratic	L	0.842	0.919	1424.63	8.353	6.748	0.956	$\hat{y} = -2.50 + 0.15*L + 0.29*L^2$
Quadratic	W	0.931	0.866	1392.74	7.713	5.841	0.963	$\hat{y} = -8.40 + 7.80*W + 0.21*W^2$
Quadratic	L.W	0.974	0.949	1202.96	4.781	3.719	0.987	$\hat{y} = 1.75 + 0.60*LW - 0.0003*LW^2$
Quadratic	L.L	0.919	0.842	1424.60	8.352	6.746	0.956	$\hat{y} = -2.23 + 0.31*LL + 0.00002*LL^2$
Quadratic	W.W	0.931	0.865	1393.13	7.721	5.824	0.963	$\hat{y} = 8.60 + 1.14*WW - 0.003*WW^2$
Cubic	L	0.919	0.842	1426.38	8.348	6.725	0.956	$\hat{y} = 11.79 - 3.55*L + 0.60*L^2 - 0.008*L^3$
Cubic	W	0.931	0.865	1394.21	7.703	5.814	0.963	$\hat{y} = 1.45 + 2.59*W + 1.06*W^2 - 0.04*W^3$
Cubic	L.W	0.949	0.948	1230.04	4.782	3.726	0.973	$\hat{y} = 0.03 + 0.67*LW - 0.001*LW^2 + 0.000002*LW^3$
Cubic	L.L	0.919	0.842	1426.50	8.350	6.760	0.956	$\hat{y} = -4.09 + 0.34*LL - 0.0002*LL^2 + 0.0000003*LL^3$
Cubic	W.W	0.932	0.866	1393.03	7.680	5.822	0.963	$\hat{y} = 5.17 + 1.42*WW - 0.009*WW^2 + 0.00003*WW^3$
Potency	L	0.919	0.844	1422.78	8.356	6.752	0.956	$\hat{y} = 0.25 * L$^2.05
Potency	W	0.931	0.867	1390.89	7.716	5.838	0.963	$\hat{y} = 4.09 * W$^1.34
Potency	L.W	0.974	0.948	1201.45	4.775	3.722	0.987	$\hat{y} = 0.91 * LW$^0.90
Potency	L.L	0.919	0.844	1422.78	8.356	6.752	0.956	$\hat{y} = 0.25 * LL$^1.03
Potency	W.W	0.931	0.867	1390.89	7.716	5.838	0.963	$\hat{y} = 4.09 * WW$^0.67
Exponential	L	0.912	0.832	1438.43	8.690	7.126	0.951	$\hat{y} = 7.07 * 1.16$^L
Exponential	W	0.918	0.842	1426.44	8.433	6.612	0.954	$\hat{y} = 13.99 * 1.21$^W
Exponential	L.W	0.918	0.842	1362.22	8.433	6.612	0.954	$\hat{y} = 22.60 * 1.01$^LW
Exponential	L.L	0.942	0.887	1472.93	7.182	5.870	0.967	$\hat{y} = 19.95 * 1.00$^LL
Exponential	W.W	0.895	0.801	1489.57	9.472	7.814	0.939	$\hat{y} = 27.58 * 1.01$^WW

x: linear dimensions; r: Pearson's linear correlation coefficient; R^2: coefficient of determination; AIC: Akaike information criterion; RMSE: root of the mean square error; MAE: mean absolute error; d: Willmott's agreement index.

The linear model without intercept, using the product between length and width (L.W) obtained the highest R^2 (0.991) and d (0.987), in addition to the lowest RMSE (2,367) and AIC (1199.48) (Table 3). Thus, the equation $\hat{y} = 0.58*LW$ generated from this model is the most suitable for estimating the area in a satisfactory way for *T. grandiflora*, as it has the best adjustments. Similar results were obtained in studies with *Tectona grandis* (Silva et al., 2020), *Theobroma cacao* (Schmidt et al., 2017) and *Mesosphaerum suaveolens* (Ribeiro et al., 2020).

Scatter diagrams between length (L) (cm), width (W) (cm), product (LxW) (cm^2) and leaf area (LA) (cm^2) to indicate patterns that fit linear and non-linear models were made (Figure 3A and B). Blue vine, like other vines, move around in their tutors to find ways to increase their survival, one of the possibilities is the search for light through their leaves, that is, increase their leaf area avoiding shading and maximizing the interception of light consequently photosynthesis (Ferrero et al., 2019).

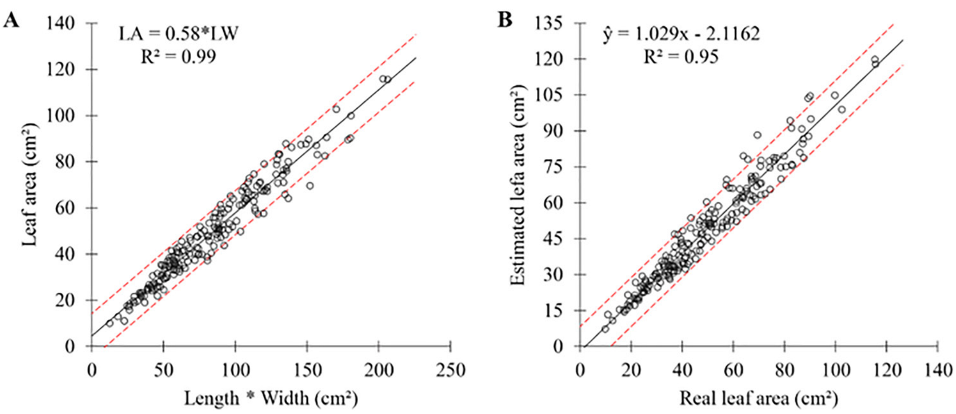

Figure 3. Relationship between real leaf area and length x width (A) and relationship between estimated leaf area and real leaf area (B).

The leaf area of a plant acts as an important control over photosynthesis, respiration, transpiration and other physiological attributes related to the diverse processes of the ecosystem (Wales et al., 2020). The leaf area is a characteristic of the structure of vegetation and has become an important parameter in ecological, agronomic, landscape and also in modeling studies (Zhao et al., 2021).

Conclusion

The leaf area of *Thunbergia grandiflora* can be estimated satisfactorily by a non-destructive method that uses linear measurements of the leaves. Many models presented values adjusted to the parameters, but the most adequate to estimate the leaf area of *T. grandiflora* was $\hat{y} = 0.58 * LW$.

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