Practical methods for non-destructive measurement of tree leaf area

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Abstract In an agroforestry context, the knowledge of leaf area is an important parameter to take into consideration because tree foliage shades the intercrop. Single leaf measurement (for example leaf length and width) is a widely used method to estimate leaf area in a rapid non-destructive way. In this study, the objectives were to estimate the leaf area of different leaf sizes and shapes for Acacia mangium Willd. (Mimosaceae), Tectona grandis L. (Verbenaceae), Hevea brasiliensis Muell.-Arg. (Euphorbiaceae), and Swietenia macrophylla King. (Meliaceae) Indonesian agroforestry trees. In order to characterise leaf area in field conditions a digital photograph method was used. Leaf length was measured in order to build relationships between leaf length versus actual leaf area. Additional measurements obtained from the digital pictures such as lamina width, number of leaflets, leaflet length and width were

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recorded in order to test the benefit regarding leaf area estimation based only on the leaf length model. The combination of these different linear leaf measurements led to various degrees of precision of leaf area estimation. The different models can then be chosen according to a compromise between the accuracy of leaf area estimation and the time allocated to field measurements.

Keywords Leaf area estimation · Models · Leaf length · Leaf width · Digital pictures · Leaflet

Introduction

In agroforestry systems light interception by the tree canopy is one of the major biophysical parameters to take into account in order to assess crop yield. Light distribution between the tree and the intercrop can be estimated thanks to radiative balance models based on a description of the trees. According to the degree of precision expected, the trees can be represented by simple three-dimensional volumic shapes (Courbaud et al. [2003](#page-9-0)), or in more detail with realistic architectural virtual plants (Godin et al. [1999](#page-9-0)). In this latter case the leaves have to be described with the maximum of accuracy by combining spatial location, Euler angles (azimuth, elevation and roll), leaf dimension and leaf area (Willaume et al. [2004](#page-9-0)). The geometrical structure of the leaves and total leaf area are the two main parameters which affect the capacity of the canopy to intercept light (Ross [1981](#page-9-0); Sinoquet and Andrieu [1993\)](#page-9-0). In this kind of study the foliage has to be described at the leaf scale. Different common direct methods are available to measure leaf area for any single leaf: different types of laser planimeters (area meter), scanner methods (Caldas et al. [1992\)](#page-9-0), gravimetric methods (Ross et al. [2000](#page-9-0)), projected area (Westoby and Wright [2003](#page-9-0)), integrating sphere (Serrano et al. [1997\)](#page-9-0), or with a fix camera and image analysis software (Granier et al. [2002\)](#page-9-0).

However, measurement of leaf area by direct methods for a whole plant or part of the plant is timeconsuming and involves a large amount of labour. Thus, many equations have been built in order to estimate leaf area by indirect methods. Montgomery [\(1911](#page-9-0)) first suggested that the area of single leaves can be calculated from linear measurements such as leaf length (L) and width (W) of each lamina. Single leaf area (LA) can be estimated using a general relationship $LA = b L W$ where b is an empirical leaf shape coefficient which differs among species, cultivar (Stewart and Dwyer [1999](#page-9-0); Bange et al. [2000](#page-9-0); Tivet et al. [2001](#page-9-0); Bhatt and Chandra [2003](#page-9-0); Pinto et al. [2004\)](#page-9-0) or crop development stage (Bange et al. [2000](#page-9-0)).

In most tropical regions, various tree species have been associated with crops, constituting 2-strata agroforestry systems. This offers the opportunity to ensure a dual function, both economic (to increase the farmers' income) and environmental (to avoid degradation of the natural environment occurring under pure-crop based land use systems). In Indonesia, farmers often associate crops with different tree

species such as Hevea brasiliensis (rubber tree) well known for latex production, Tectona grandis (teak) and Swietenia macrophylla (mahogany) planted for their wood quality and Acacia mangium frequently used for soil fertility improvement. Among these four species, only H. brasiliensis leaves have been studied for leaf area assessment relations by Lim and Narayanan ([1975\)](#page-9-0). In this kind of agroforestry system, a non-destructive method is essential as the trees are an important income for the farmers and generally cannot be felled for scientific studies. According to leaf shape and size, direct leaf area determination in the field is often difficult, and it is generally unrealistic to use sophisticated leaf area measurement devices. A possibility is to take a picture of the leaf directly with a digital camera. It is then possible to measure leaf length. It is easier and less time consuming to measure leaf length than both leaf length and leaf width (Kandiannan et al. [2002](#page-9-0)), particularly for compound leaves.

The objectives of this study were (i) to investigate whether leaf area determination with a digital camera and image analysis could give a good estimation of the actual leaf area for leaves of different sizes and shapes (simple and compound), (ii) to develop models relating leaf length and leaf area for further use in agroforestry systems in Indonesia, and (iii) to test complementary measurements from the digital pictures (such as leaf width, number of leaflets, leaflet length and leaflet width) and to estimate the benefit in leaf area estimation compared to a model that uses only the leaf length as input.

Species	Number of Type of trees	growth	Structure	Leaf shape	Phyllotaxy	Total height (cm)	Basal diameter (cm)
Acacia mangium (Mimosaceae)	10	Continuous	Branch	Simple (phyllode)	Spiral $(3/8)$	534.4 ± 32.2 a	8.36 ± 0.52 a
Tectona grandis (Verbenaceae)	15	Rhythmic (2 Gu)	Monocaule	Simple	Opposite- decussate	390.6 ± 9.86 b	6.04 ± 0.19 b
Hevea brasiliensis (Euphorbiaceae)	15	Rhythmic (6 Gu)		Monocaule Compound (3) leaflets)	Spiral $(5/13)$	237.1 ± 6.98 c	2.52 ± 0.06 d
Swietenia macrophylla 15 (Meliaceae)		Rhythmic (8 Gu)	Monocaule	Compound $(4-12)$ leaflets)	Spiral $(3/8)$	204.5 ± 5.06 d 3.37 ± 0.15 c	

Table 1 Description of the four species concerning the type of growth, trunk structure, leaf shape, phyllotaxy, total height (mean in cm \pm s.d.), basal diameter (mean in cm \pm s.d.)

a, b, c and d: comparison using the Mann-Whitney-Wilcoxon test; in the event of a change of letter between two lines, the difference is significant at the 95% threshold. Gu = Growth unit

Materials and methods

Plant material

Four tree species with different growth modalities, leaf shapes and sizes (Table [1](#page-1-0) and Fig. 1) were studied in Indonesian agroforestry systems. Acacia

Fig. 1 Pictures of the different leaf sizes and shapes for average leaves of (A) Acacia mangium, (B) , Tectona grandis (C) Hevea brasiliensis and (D) Swietenia macrophylla

mangium Willd. (Mimosaceae) with simple leaves, Hevea brasiliensis Muell.-Arg. (Euphorbiaceae) with trifoliate leaves and Swietenia macrophylla King. (Meliaceae) with compound leaves composed of 4–12 leaflets were studied in a site in Pakuan Ratu $(04^{\circ}32'$ S, $104^{\circ}56'$ E), situated in north Lampung area in the southern part of Sumatra Island, Indonesia. Tectona grandis L. (Verbenaceae) with big simple leaves was studied near Malang $(08^{\circ}10' S,$ $112^{\circ}20'$ E), situated in the eastern part of Java Island, Indonesia. All studied trees were 2 years old.

Sampling and measurements

The leaves were selected in a way to have all the sizes ranging from the smallest to the biggest ones. A total of 512 leaves were sampled on 10 A. mangium trees, 159 leaves on 15 T. grandis trees, 213 on 15 H. brasiliensis trees and 242 on 15 S. macrophylla trees. Leaf length (L) was measured to the nearest millimetre from the leaf tip or from the rachis tip (for S. macrophylla) to the base of the leaf blade (the point of attachment of the petiole on the stem). Numerical photographs were taken, with a Nikon Coolpix 4500 digital camera, for all the selected leaves. The zoom lens was placed in the middle of its course to avoid as far as possible image distortion. High quality JPEG pictures were taken at 1 m distance from the leaves. We used a homogeneous white background for easier analysis. A ruler was placed near the leaves to establish the scale. One to several leaves per picture, according to the leaf size, were memorized. The area of each individual leaf (in square centimetres) was then determined by image analysis by full pixel region (Optimas V 6.5, Media Cybernetics, Sylver Spring, MD, USA). Leaf length measured on Optimas software was also recorded to determine the robustness of the numerical photograph method in comparison to the measured leaf length. In addition, complementary leaf measurements were recorded directly on the Optimas software such as the lamina width for simple leaves of A. *mangium* and T. grandis. For S. macrophylla compound leaves, the number of leaflets was noted and the leaflet length and width were measured for the biggest leaflet of each leaf. Concerning H. brasiliensis, leaflet length and width were measured for the terminal leaflet.

Analysis method

A linear model was first established for Optimas leaf length (OL) versus directly measured leaf length (L): $OL = f (L)$. The relationship between the Optimas determined leaf area (OLA) and leaf length (L) was established using the following non-linear weighted regression model adjusted with maximum likelihood estimations (assuming normality) (Proc NLP, SAS software):

 $ELA = \alpha(L)^{\beta} + \varepsilon$

Where ELA is the estimated leaf area $\text{(cm}^2\text{), } \alpha$ and β are the estimated parameters, L is the leaf length (cm) and ε corresponds to the error term (ε = N (0, σ) $(L)^k$). The models that predict the leaf area (ELA) were evaluated using the linear model $ELA = f$ (OLA). Linear and non-linear models were fitted using the procedures REG and NLP of SAS software. The agreement between $OL = f (L)$ and $ELA = f$ (OLA) were evaluated using the coefficient of determination (R^2) , the modelling efficiency index (EI) (Mayer and Butler [1993\)](#page-9-0), the root mean square errors (RMSE) and the F-statistical test for bias, slope $= 1$ and intercept $= 0$. Several models were then tested to study the gain obtained by using additional variables for leaf area estimation. Only the more accurate ones are proposed in this paper. These models were fitted with the maximum likelihood, and

model comparison was based upon \mathbb{R}^2 , EI, RMSE and the F-statistical test.

Results

Leaf length estimation

The leaves of the four species had a significantly different lamina length (Table 2). The smallest leaves were A. mangium phyllodes with an average of 16.25 ± 6.46 cm and the biggest ones were T. grandis leaves with 39.94 ± 21.87 cm. S. *macrophylla* and *H*. brasiliensis leaf length were intermediate with respectively 25.59 ± 11.05 cm and 36.47 ± 14.08 cm.

The relationships between the measured leaf length and Optimas leaf length were satisfactory for all the four species. The coefficient of determination was highly significant with values above 0.98 (Table 2) but the simultaneous F-test for bias, slope $= 1$ and intercept $= 0$ was significant for all the species ($P < 0.001$). The slope was significantly lower than 1 for A. mangium and T. grandis. The intercept was significantly different from 0 for A. mangium, H. brasiliensis and S. macrophylla. However, the mean absolute error was quite low and the modelling efficiency index was close to the maximum value 1 and similar to R^2 respectively for the four species. The RMSE values ranged from 0.57 cm to 1.1 cm (Table 2).

Leaf length	Acacia	Hevea	Mahogany	Teak	
Number of observations	512	213	242	159	
Mean \pm s.d. (L)	16.25 ± 6.46	36.47 ± 14.08	25.59 ± 11.05	39.94 ± 21.87	
Mean (OL)	16.18	36.10	25.26	38.42	
Mean errors	-1.82	-0.24	0.03	-0.007	
Mean absolute errors (MAE)	0.49	0.66	0.44	0.83	
Modelling efficiency index (EI)	0.98	0.99	0.99	0.99	
RMSE	0.67	0.84	0.57	1.11	
Linear regression (F-statistics)					
R^2	0.98	0.99	0.99	0.99	
Slope	$10.14**$	3.36 ns	0.18 ns	85.89***	
Intercept	$4.49*$	$6.67*$	$11.55***$	0.03 ns	
Bias	$7.62***$	73.03***	29.15***	192.5***	

Table 2 Statistical summary of leaf length determined by Optimas versus measured leaf length in centimetres

Units: means and RMSE are given in cm, EI and R^2 are dimensionless. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

s.d. = standard deviation

Estimation of leaf area from leaf length

The leaf area was, as leaf length, significantly different for all the four species (Table [3\)](#page-5-0). A. mangium showed the smallest leaf area with an average of $65.21 \pm 35.20 \text{ cm}^2$ compared to 909.61 \pm 747.34 cm² for *T. grandis* leaves. Contrary to leaf length, S. macrophylla leaf area was higher with 380.31 \pm 255.89 cm² compared to *H. brasilien*sis leaf area of 226.37 ± 118.93 cm².

Figure [2](#page-6-0) presents the relationship between leaf length (L) and Optimas leaf area (OLA) with the fitted curves (ELA) and the corresponding 95% confidence intervals. The regressions (Table [3](#page-5-0), bold characters) were highly significant in all cases $(P < 0.001)$. The confidence intervals of estimates show that the heteroscedasticity was satisfactorily taken into consideration.

The measured (OLA) versus simulated leaf area (ELA) scatterplots (Fig. [3](#page-6-0) and Table [3](#page-5-0)) showed a non-significant bias for S. macrophylla leaf area estimation based on leaf length. The F-statistical test was significant for the three others species. The coefficients of determination were quite high for the four species and ranged from 0.84 to 0.97 (Table [3](#page-5-0)). The modelling efficiency index was similar to the R^2 value, except for a slight difference for S. macrophy*lla*, showing no major deviation from the line $y = x$. Relative to the leaf area the RMSE was very high for S. macrophylla.

Estimation of leaf area from other measured variables

For A. mangium and T. grandis simple leaves, taking into account the lamina width in addition to the length (see additional models in Table [3\)](#page-5-0) improved the leaf area estimation (Fig. [4](#page-7-0)). The simultaneous F-statistical test for bias, slope $= 1$ and intercept $= 0$ was non significant for the two species (Table [3](#page-5-0)). The coefficient of determination was highly significant with values higher than 0.99. The modelling efficiency index was close to the maximum value 1. When compared to the average value of leaf area the RMSE was low. Compared with the simple leaf length model f(L), RMSE for the leaf length and width model f(LW) decreased by 61% and 66% for A. mangium and T. grandis respectively. In the case of H. brasiliensis and S. macrophylla compound leaves,

respectively five and four relations were retained according to different variables taken into account (see Table [3](#page-5-0) for the fitted equations). For H. brasiliensis, taking into account additional variables such as leaflet length and width improved the leaf area estimation (Table [3](#page-5-0)). For all the 5 relations, the F-statistical test was no longer significant. The coefficient of determination ranged from 0.92 to 0.94 (Table [3](#page-5-0) and Fig. [4\)](#page-7-0) and the modelling efficiency index was of the same order. When compared with the average value of leaf area the RMSE was quite low. Compared to the simple leaf length model f(L) the benefit ranged from 20% to 32%. For S. macrophylla, leaf area estimation was also improved by additional measurements such as leaflet length and width and number of leaflets (Table [3\)](#page-5-0). The F-statistical test was no longer significant when number of leaflets (nbl) was introduced but was still significant for the f(Llw) model ($P < 0.05$). In all cases the coefficient of determination was significant and ranged from 0.86 to 0.96 (Table [3](#page-5-0) and Fig. [4](#page-7-0)). The modelling efficiency index ranged from 0.83 to 0.96. The RMSE was improved when compared with the mean leaf area. Compared with the simple leaf length model f(L) the benefit based on the RMSE ranged from 5% to 54%.

Discussion

Single leaf area estimation based on leaf length

The results from this study show a good correlation between measured leaf length and estimated leaf length on Optimas software. The RMSE to mean leaf length ratio show a very slight bias with an error of $2-4\%$ of the measured length. For T. grandis leaves, the bias, more important for the biggest leaves, can be due to the crooked shape of the lamina. However, these results suggest that a digital picture of the leaf, taken from selected leaves in the field, is a satisfactory method to determine the leaf area, as nondestructive leaf area determination is of great interest to avoid plant degradation. In practice, in the field, pictures of A. mangium single flat leaves can be realised directly on the tree with a scale stick on the lamina, while for compound and non-flat leaves pictures have to be realised on cut leaves.

Table 3 Statistical summary for the different validation models Table 3 Statistical summary for the different validation models

Units: means and RMSE are given in cm², EI and R² are dimensionless. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Units: means and RMSE are given in cm², EI and R² are dimensionless. * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$

Fig. 2 Leaf area related to measured leaf length (L) for A. mangium, T. grandis, H. brasiliensis and S. macrophylla. The 95% confidence interval of individuals is given by the dashed lines. The solid line is the non-linear regression line computed with the corresponding bold equations in Table [3](#page-5-0)

Leaf area (cm²)

Leaf area (cm²)

Measured (OLA) Estimated (ELA)

Measured (OLA) Estimated (ELA)

Leaf area (cm²)

Leaf area (cm²)

Leaf area can be estimated via simple leaf measurements. According to the present results, the tested models led to different leaf area estimation accuracies. Considering only the leaf length model, both for simple leaves (A. mangium and T. grandis) and compound leaves (H. brasiliensis), the estimation of leaf area showed a significant bias regarding the F-statistical test. Only S. macrophylla showed a

non-significant bias, but this can be explained by a great dispersion of observations.

 $\mathbf 0$

 $0.918x + 16.139$ $R^2 = 0.8832$

0 200 400 600

ELA (cm²)

0

200

400

OLA (cm²)

 OLA (cm²)

600

0

40

80

OLA (cm²)

OLA (cm²)

120

160

700

OLA (cm²)

 OLA (cm²)

Improvement of single leaf area estimation

In the case of simple leaves, taking into account leaf length plus leaf width in the model improved up to 60% the precision of leaf area estimation, and the

 $.0128x - 4.5689$ $R^2 = 0.8485$

0 700 1400

ELA (cm²)

Fig. 4 Estimated leaf area (ELA) compared to Optimas determined leaf area (OLA) from the different models described in Table [3](#page-5-0) for A. mangium, T. grandis, H. brasiliensis and S. macrophylla. The solid line is the linear regression line and the dashed line is $y = x$

bias was eliminated. The RMSE to leaf area mean ratio decreased under 5% for both species. Many other studies have established a successful method of non-destructive estimation of leaf area by measuring linear dimensions such as both length and width (Stewart and Dwyer [1999;](#page-9-0) Bange et al. [2000](#page-9-0); Tivet et al. [2001](#page-9-0); Bianco et al. [2003](#page-9-0); Pinto et al. [2004](#page-9-0)). Other leaf area estimation studies focused on leaf width measurement only on Kiwifruit (Greer [2000](#page-9-0); Seleznyova and Greer [2001](#page-9-0)) or leaf length measurement only on Pepper (Kandiannan et al. [2002](#page-9-0)) or on apple (Willaume et al. [2004\)](#page-9-0). In many cases, leaf length and width were measured for simple leaves. From these different studies on leaf area estimation none had tested the performance of one variable compared with the other.

For compound leaf species such as H. brasiliensis and S. macrophylla, estimation of the leaf area from leaf length only was less satisfactory in comparison with the other models. The RMSE to leaf area mean ratio was 19% for H. brasiliensis and 26% for S. macrophylla. The dispersion of errors can result from the different shapes and sizes of the leaflets. Taking into account additional variables such as number of leaflets, leaflet length and leaflet width improved in different manners the leaf area estimation. The addition of only one variable eliminated the bias revealed by the F-statistical test. In the case of S. macrophylla, the f(Llw) model did not suppress the bias even though the R^2 , EI and RMSE were improved in comparison with the average leaf area. Taking into account the number of leaflets with leaf length did not add much precision to the estimation of leaf area, in comparison with the leaf length model f(L), with only 5% of benefit, but it eliminated the bias. For this species, the addition of the number of leaflets in the different models always eliminated the bias.

In our case, only one leaflet was measured, the terminal one for H. brasiliensis and the biggest one for S. macrophylla, for the different models studied. Former studies on trifoliate leaves such as Glicine max (L.) Merr. (Wiersma and Bailey [1975](#page-9-0)) and Hevea brasiliensis Müll. Arg. (Lim and Narayanan [1975\)](#page-9-0) considered all the leaflets for each single leaf. In the case of the rubber tree the authors considered the trifoliate leaf as 3 simple leaves and the measurements were done at the leaflet level in terms of lamina length and width. In their study the standard error of estimation for the area-length relationship ranged between 6.7% and 12.7% , and for the area-width relationship between 7.4% and 16.5% for different clones and leaflet positions (Lim and Narayanan [1975](#page-9-0)). Considering both length and width in their models, these authors improved the leaflet area estimation with an average error estimation of 4.3%. Our results can not be directly compared with these because in our study the total leaf area was taken into account and not each leaflet. Indeed, there is a significant difference of size between the terminal leaflet and the two lateral ones. However, comparison with A. mangium and T. grandis simple leaves show a good agreement with the H. brasiliensis leaflets studied by Lim and Narayanan [\(1975\)](#page-9-0). Measurement of each leaflet length and leaflet width represents the maximum of accuracy in leaf area estimation (Lim and Narayanan [1975;](#page-9-0) Wiersma and Bailey [1975](#page-9-0)). But this involves additional time constraints to obtain measurements of all the lateral leaflets. Wiersma and Bailey ([1975\)](#page-9-0) suggested that estimation of leaf area by using lw products or leaflet length (l^2) or leaflet width (w^2) of only the terminal leaflet provides a useful and rapid estimation of the leaf area. Lim and Narayanan [\(1975](#page-9-0)) estimated the entire rubber leaf area by measurement of the length and width of the terminal leaflet and one lateral leaflet. In our study, the best fitted model was when the total leaf length and the terminal leaflet length and width were taken into account. In the case of another trifoliate leaf species, Phaseolus vulgaris L., Bhatt and Chandra ([2003\)](#page-9-0) estimated the entire leaf area by measuring leaf length and maximum width without taking into account the leaflet level. Among their different models tested L^2 , W^2 , $L + W$ and LW, the best correlation existed with $L + W$ and LW. From a t-test analysis their $L + W$ model was most accurate to estimate leaf area. In our case, the additive models, not shown here (except with the leaflet number), were not as accurate as the multiplicative models.

The present study proposes a series of relations concerning simple and compound leaves for 2-yearold trees of four species with different types of leaves. The different models studied are only applicable to trees, but not to seedlings, as the leaf shape of seedlings can be very different from trees. Indeed, A. mangium seedlings are characterised by compound leaves and S. macrophylla by simple leaves. For

T. grandis the leaves are simple but with a different shape (from obovate to deltoid). And for H. brasiliensis the leaves of seedlings have the same shape but are smaller. This study offers the possibility to select either one of the different models according to the accuracy of leaf area estimation required and the time allocated to field measurements. In this way, estimation of single leaf area can be done by easy measurements performed directly on the tree.

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