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Leaf area estimation by linear regression models in Cassava (Morphotype: Philippine)

M.R. Karim, M.S.A. Fakir, M.G. Mostafa and A.K.M.A. Prodhan
Department Crop Botany, Bangladesh Agricultural University, Mymensingh-2202
E-mail: fakirmsa@yahoo.com

Abstract: Regression models of estimating leaf area based on linear measurements of length and breadth of leaf-lobes were investigated in Philippine Morphotype of Cassava (*Manihot esculenta*), at Mymensingh (24°75´N, 90°50´E). The length (L) and breadth (B), fresh (LF_{wt}) and dry weights (LD_{wt}) of one hundred leaves were used for leaf area (LA) prediction Models. Fifteen regression models were estimated for predicting LA. Only four models, appeared proper, were LA = -9.96 + 12.128^{**} L (Model # 1), LA = 1.933 + 0.907^{**} L×B (Model # 7), LA = 7.732 + $60.76^{**}LF_{wt}$ (Model # 10) and LA = 10.41 + $175.48^{**}LD_{wt}$ (Model # 13). These regression models showed linear relationships when actual leaf area plotted against predicted leaf area of another one hundred leaves from different samples and that this confirmed accuracy of the developed models. Moreover, models selection indices had high predictive ability (high R^2) with minimum error (low mean square error and percentage deviation). The selected models appeared accurate, rapid although unsophisticated and can be used for estimation of LA in both destructive (Models # 10 and 13) and non-destructive (Models # 1 and 7) means in Philippine Morphotype of Cassava.

Key words: *Manihot esculenta*, leaf area prediction, nondestructive, destructive, regression.

Introduction

Cassava (*Manihot esculenta* Crantz) is a perennial shrub and is valued for it's under ground starchy edible tuberous root. Cassava roots are consumed as staple in Africa and used as raw material in garments, bakery, pharmaceutical and feed industry (Islam *et al.*, 2007a). At harvest, the plant height ranges from 100 to 200 cm with 2 to 3 branches bearing 300 to 500 simple leaves. The large simple, dark green, reddish veined leaves are palmately divided into about 3 to 7 leaf-lobes, each with 3 to 14 cm long and 6 to10 cm wide. The number of tubers varies from 3 to 11 per plant with length of the tuber varies from 20 to 50 cm, and that of weight of the tuber 35 to 500g (Cock *et al*., 1979; Islam *et al.*, 2007b; Islam *et al.*, 2008). Leaf is the primary source of food production apparatus in Cassava. Leaf size and weight directly affect the growth of storage root (Boerboom, 1978; Islam *et al.*, 2007). Continuous leaf shed is accompanied by emergence of new leaf in Cassava. Hence the dynamics of leaf production in Cassava is an important factor. Hence leaf area, leaf number, leaf abscission are the key factor to Cassava yield improvement (Karim, 2004).

The measurement of leaf area and leaf production of a plant is an important index of growth and development and is commonly employed in Agronomical and Physiological studies. Cassava yield or storage tuber (sink) yield depend on foliage structure (sink). In Cassava plant assimilates is translocated from source to sink. Both destructive and non-destructive methods have been used to predict leaf area in a range of tree crops and non-tree crops. Some examples of non-destructive methods are used in Cassava (Connor and Cock, 1981; Yao *et al.*, 1988; Alves and Setter, 2000 and Karim, 2004); Sour orange (Ramkhelawan and Brathwaite,1990); Black pepper (Kandiannan *et al*., 2002); Soybean (Chen and Welch, 2002); and destructive methods are used in *Typha* (Bianco *et al*., 2003); Waterhyacinth (Marchi and Pitelli, 2003); Chayote (Meitei and Singh, 2002) and faba bean (Erkut, 2007). Both the destructive and non-destructive methods have advantages and disadvantages. Non destructive estimation is simpler, can be employed in the field without using any sophisticated machine and without removing the leaves. The destructive methods may be laborious, time consuming but may be rapid and advantageous in case of bulk LA estimation. There is scanty information on leaf area estimation of Cassava at abroad (Connor and Cock, 1981; Alves and Setter, 2000) and only one report in Bangladesh (Karim, 2004). The current research was conducted to develop and test regression models that would enable the leaf area to be predicted from simple, linear, destructive and non-destructive measurements.

Materials and Methods

Crop establishment: The experiment was conducted at the field laboratory of the Department of Crop Botany, Bangladesh Agricultural University, Mymensingh (24°75′N latitude and 90°50′E longitude) between May 2003 and November 2004. The soil was silty loam at a mean elevation of 18m above the sea level belonging to the Sonatola soil series of non calcareous dark gray flood plain soil under the old Brahmaputra Flood Plain Agro-ecological zone-9. The soil was silty loam having a total nitrogen 0.10%, organic matter 1.35%, available phosphorus 18.5 ppm, potassium 0.28 ppm, sulphur 18 ppm, pH 6.8. The CEC and fertility status of the soil low and medium, respectively.

The experimental area was fertilized with cowdung (16 t ha^{-1}), Urea (83kg ha^{-1}), TSP (62 kg ha^{-1}) and MP (62 kg ha^{-1}) at the time of final land preparation. The Morphotype of Cassava was Phillippine. The size of unit plot was 4.5 m^2 (3m × 1.5m) with plot to plot and block to block distances were 0.5m and 1.0m, respectively. Plant spacing was 1m × 0.75m. The plots were raised up to 15 cm from the soil surface. Healthy and uniform size (about 12 cm with 6 nodes) 8 months old stem cuttings of Cassava, collected from previous expt., were planted with an angle of 45° from ground, placing two-third of the cutting in the soil. The distance between two stalks was 75 cm and 1 stem cutting was planted in each hole. The cuttings were treated with the fungicides (Bavistin, @5g per Liter) before planting. Cutting was watered after planting, and watering continued for several days until their establishment. Other cultural practices were carried out when needed.

Crop sampling and data collection: Fifteen plants from three replications were randomly selected. Length (L) and breadth (B), area (LA), fresh (LFwt) and dry weights (LDwt) were recorded after oven drying (80°C±2) samples until constant weight. Length and width (cm) of the all lobes were measured and averaged out (Fig.1 A, B). Leaf area (cm^2) was measured by a leaf area meter (Model # L1-3000, Licor, USA) for calculating and predicting leaf area from L and B of leaf-lobes and linear regression Models were developed (LA = a+b×L), (LA = a+b×B) and (LA = a+bL×B). Models were also developed for calculating and predicting LA from LFwt and LDwt (g) using regression Models (LA = a+b×LFwt and LA = a+b×LDwt). One hundred leaves were randomly sampled for developing models and another separate one hundred leaves were used for prediction.

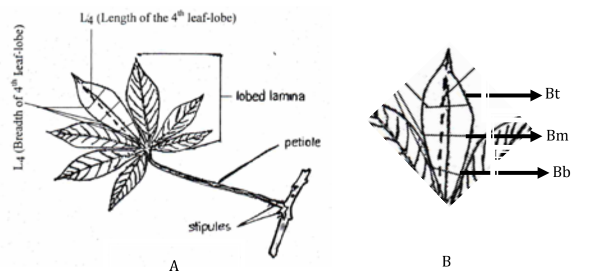

Fig.1. Diagram of a Cassava leaf lamina (A) showing two linear measurements taken in non-destructive method, a single leaf-lob (B) showing breadth at different position

L = Mean leaf-lobe length. Henceforth, it will be designated only leaf length (L), and L = (L_1 + L_2 + L_3 ---- L_n)/n, where: L_1 is the length of the 1st leaf-lobe of the leaf L_2 is the length of the 2nd leaf-lobe of the leaf L_n is the length of the L_nth leaf-lobe of the leaf n is the number of leaf-lobes of the leaf. B = Mean leaf breadth, Where: B = (B_1 + B_2 + B_3 --- B_n)/n, where, B_1 is the leaf breadth, {(B_1 = B_{b1} + B_{m1} + B_{t1})/n} of the 1st leaf-lobe, where, B_{b1} is the breadth of the 1st leaf-lobe at base from the tip, B_{m1} is the breadth of the 1st leaf-lobe at middle from the tip, B_{t1} is the breadth of the 1st leaf-lobe at top, and similarly, B_2 is the breadth of the 2nd leaf, B_n is the breadth of the L_nth leaf. For breadth measurement, each leaf-lobe was devided into three equal portions, breadth at base (B_b), middle (B_m) and top (B_t) (Fig.1 A, B).

Statistical analysis: The data were compiled and analysed following the analysis of variance (ANOVA) technique and a correlation study between the related data was also carried out with a computer package programme SPSS 10 for Windows. Regression analysis was also carried out.

Results

Variation in leaf size and weight: There were variations in length (L) and breadth (B), fresh (LF_{wt}) and dry weights (LD_{wt}) of Cassava leaf (Table 1). Leaf length varied between 3.1 and 11.2 cm with mean L and standard deviation (Std) were 6 cm and 1.40, respectively. The variation in B was 6.3 to 15.4 cm and that of L x B was 30.1 to 164.8 cm^2. Variation of LF_{wt} ranged 0.4 to 2.4 g and that of LD_{wt} 0.2 to 0.8 g .

Correlation of leaf area with leaf size and weight: The correlation coefficients (r) of L, B with actual leaf area (LA) are presented in the Table 2. The squared values of L, B, and LxB were also correlated with LA (Table 2). Natural logarithms of L, B, LFwt, LDwt, were also correlated with log LA. The correlation coefficients of L, B, LxB, LF_{wt} LD_{wt} L^2, B^2, $(LxB)^2$, $LFwt^2$ and $LDwt^2$ with LA ranged 0.631 to 0.987 and appeared highly significant ($P \le 0.01$). The value of r ranged 0.869 to 0.952 when the variables L, B, LxB, LFwt, LDwt, L^2, $B^{2,}$ $(LxB)^2$, $LFwt^2$ and $LDwt^2$ were correlated with LA. The value of r was 0.649 to 0.987 when Log L, Log B, Log (LxB), Log LF_{Wt}, and Log LDwt were correlated with Log LA. The relationship of variables L^2, B, B^2 and LD_{wt} with LA and that of Log B with log LA showed lower correlation, varied from 0.631 to 0.868 (Table 2). High correlation existed between L and B (r = 0.97**, data not shown) and between LF_{wt} and LD_{wt} (r = 0.98**, data not shown).

The standard error of the regression coefficient (r), coefficient of determination (R^2) and mean square error (MSE) for 15 models are presented in the Table 3. For each of these models the regression coefficients (b) of the independent variable are all significantly different from zero (b#0). Equations 7, 9, 10 and 12 had the overall higher predictive ability indicated by high R^2 values (R^2=0.974, 0.961, 0.906 and 0.914, respectively). Equations 1, 3, 8, 13, and 15, also showed good predictive potential (R^2 = 0.750, 0.714, 0.885, 0.809, and 0.791, respectively).

Table 1. Variation of leaf length (L) and width (B), and leaf fresh (LF_{wt}) and dry weights (LD_{wt}) in Cassava† (Morphotype : Philippine)

Parameter	Leaf dimension and weight				
	L (cm)	B (cm)	L×B (cm^2)	LF_{wt} (g)	LD_{wt} (g)
Minimum	3.1	6.3	30.1	0.4	0.2
Maximum	11.2	15.4	164.8	2.4	0.8
Average	6.0	11.1	66.6	0.9	0.3
Standard deviation	1.40	1.90	22.80	0.30	0.10

†: One hundred randomly selected leaves were sampled

In order to test and compare the accuracy of the regression models, a further 100 leaves of the same Morphotype of Cassava were randomly selected from different plants and similar linear measurements were recorded again. The regression models (Table 3) used to predict LA following total deviation and unexplained variations were compared (Table 4). The percentage of deviation then computed for each Model (Table 5). The regression models that allow non-destructive measurement (Models # 1, 3, 7 and 8) showed small percentage of deviation and mean square error (3.49 and 108.28, 1.18 and 0.30, 3.50 and 11.31, 0.88

and 52.854, for % deviation and MSE, respectively) (Tables 4 and 5).

Table 2. Correlation coefficient of leaf area (LA) with leaf size (leaf length, L; leaf breadth, B) and weight (leaf fresh weight, LF_{wt}; leaf dry weight, LD_{wt}) in Cassava (Morphotype: Philippine)

Independent	Dependent variable	
	LA	Log LA
L	0.869**	-
L^2	0.868**	-
Log L	-	0.845**
B	0.637**	-
B^2	0.631**	-
Log B	-	0.649**
L×B	0.987**	-
$(L\times B)^2$	0.938**	-
Log (L×B)	-	0.987**
LF_{wt}	0.952**	-
$LFwt^2$	0.882**	-
Log LF_{wt}	-	0.956
LD_{wt}	0.899**	-
LD_{wt2}	0.841**	-
Log LD_{wt}	-	0.893**

Two asterisks denotes significant at 1% level of probability after t-test.

Models # 7, 8 and 9 were two dimensional and among them Model # 7 had high predictive ability (R^2 = 0.974) (Table 3). Again, the regression model following destructive measurement (Models # 10, 13 and I5) showed also smaller percentage of deviation (3.28, 3.18, and 2.00, respectively) and Models # 12 and 15 showed lower mean square error (0.009 and 0.002 respectively) (Tables 3 and 5).

Models # 12 and 15 were the equations of LF_{wt} and LD_{wt} and their R^2 values were smaller (0.914 and 0.791, respectively). The relationship between the predicted LA and observed LA for 100 leaves was plotted using Models # 1, 7, 10 and 13 (Fig.2). The points lying near the straight line, represented predicted LA ≈ observed LA. The plot for Models # 1 and 7 were unbiased in predicting LA. The plot for Models # 10 and 13 showed that a fair estimate can also be obtained. Colinearity existed between L and B (r = 0.97**, data not shown) and LF $_{wt}$ and LD_{wt} (r =0.98**, data not shown) in both the methods. Models # 1 and 10 were simple and rapid for LA estimation.

Discussion

Estimation of leaf area has been an important index of `source' size. Measurement of leaf size, area and weight are often employed in the Agronomical and Physiological experiments to understand the relationship of source with sink (yield). Conventionally, leaf area (LA) is measured /estimated by electronic devices *viz*., by leaf area meter of different models. In such method, leaves are usually harvested destructively. Moreover, leaf area meter is a sophisticated instrument, may not, be available, in a particular place. Other traditional methods are (i) graph paper tracing method where leaves are placed and traced on printed graph paper and LA is estimated from graph paper area (ii) K-value method where leaf area is obtained by multiplying leaf length (L) with leaf breadth (B) and a constant `K'. The K is determined dividing actual leaf area (LA) by apparent leaf area (LxB). i.e. K = (LA/Lx B) in approximately rectangular/square leaf (Alam *et al.*, 2006).

Table 3. Standard error (SE) of the regression coefficient (b), coefficient of' determination (R^2) and mean square error (MSE) of the regression models derived for predicting the leaf area (LA) in Cassava (Morphotype: Philippine)

Model Number	Regression Model	SE of B	R^2	MSE
	Models with leaf length (L) alone			
1	LA = -9.96+12.128 L	0.698	0.755	108.280
2	LA= 27.77+0.915 L^2	0.053	0.754	108.770
3	Log LA=2.187+1.079 LogL	0.069	0.714	0.030
	Models with leaf breadth (B) alone			
4	LA= -16.783+7.146 B	0.873	0.406	262.720
5	LA= 23.169+0.311 B^2	0.039	0.398	266.104
6	Log LA= 1.131+1.233 Log B	0.146	0.420	0.062
	Models using both L and B			
7	LA = 1. 933+0.907 (LxB)	0.015	0.974	11.310
8	LA= 36.406+0.0052 $(LxB)^2$	0.0001	0.885	52.854
9	Log LA= 0.029+0.965 log L+ 0.986 log B	0.021,0.031	0.961	0.002
	Models with leaf fresh weight (LFwt) alone			
10	LA= 7.732+60.761 LFwt	1.981	0.906	41.731
11	LA=39.996+24.226 $LFwt^2$	1.311	0.777	98.572
12	Log LA=4.228+0.888 Log LFwt	0.027	0.914	0.009
	Models with leaf dry weight (LDwt) alone			
13	LA= 10.41+175.48 LDwt	8.614	0.809	84.470
14	LA= 41.494+210.72 LDw^{t2}	13.671	0.708	129.11
15	Log LA = 5.21+0.888 Log LDwt	0.040	0.791	0.022

All logarithms are to the base e, Selected Models are bold ones

All these methods are destructive and/or laborious and time consuming. Non-destructive and unsophisticated estimation of LA from simple linear measurements of leaf size, mid-rib length (L) and width (B) have been used by researchers (Ramkhelawan and Brathwaite, 1990,1992; Lu *et al.*, 2004). Further LA estimation in large number of plants would be highly advantageous for weight measurements. Estimation of LA from destructive measurement *viz*., fresh and dry weights of leaf are also used and these provide, in many cases, a better estimate as indicated from high R^2 value (Pariari *et al.*, 2002; Lu *et al.*, 2004). Sometime, simple transformation of linear measurement of leaf size, squared value of L and B, natural log transformation of L, B and leaf' weight improves the degree of estimation (Ramkhelawan and Brathwaite, 1990, 1992; Pariari *et al.,* 2002; Lu *et al.,* 2004).

Table 4. Comparison of accuracy of regression Models derived for predicting the leaf area (LA) in Cassava (Morphotype: Philippine)

Model Number	Regression Model	∑(Total deviations)[a]	∑(Total deviations)[2b]	Unexplained variations[c]
	Models with leaf length (L) alone			
1	LA = -9.96+12.128 L	-244.84	24175.81	24.5
2	LA= 27.77+0.915 L^2	-530.54	42716.85	24.5
3	Log LA= 2.187+1.079 LogL	177.97	1302.10	18.6
	Models with leaf breadth (B) alone	-126.34	22702.57	59.4
4	LA= -16.783+7.146 B			
5	LA= 23.169+0.311 B^2	-618.80	47449.32	60.2
6	Log LA= 1.131+1.233 Log B	-96.20	22154.16	58
	Models using both L and B			
7	LA = 1. 933+0.907 (LxB)	249.49	16730.86	2.6
8	LA= 36.406+0.0052 $(LxB)^2$	62.17	15671.41	12.0
9	Log LA= 0.029+0.965 log L+ 0.986 log B	852.00	63893.56	3.9
	Models with leaf fresh weight (LFwt) alone			
10	LA= 7.732+60.761 LFwt	-230.37	25099.66	9.4
11	LA=39.996+24.226 $LFwt^2$	-466.36	28048.70	22.3
12	Log LA=4.228+0.888 Log LFwt	-133.02	24503.44	8.6
	Models with leaf dry weight (LDwt) alone			
13	LA= 10.41+175.48 LDwt	-230.37	25099.66	19.1
14	LA= 41.494+210.72 $LDwt^2$	533.87	29617.44	29.2
15	Log LA = 5.21+0.888 Log LDwt	140.72	29587.02	20.9

All logarithms are to the base e, Selected Models are bold ones; **a**∑ (Total deviations = ∑(Actual leaf area - predicted leaf area), **b**∑ (Total deviations)^2 = E(Actual leaf area - predicted leaf area) ^{2} **,c** Unexplained variation = 100 (1-R^2) where R^2 is the coefficient of determination of the regression Model

Table 5. Comparison of the percentage of deviation of regression Models for predicting leaf area (LA) in Cassava (Morphotype: Philippine)

Model Number	Regression Model	Total leaf area measured (cm^2)	Total leaf area predicted (cm^2)	Percentage variations[a]
	Models with leaf length (L) alone			
1	LA = -9.96+12.128 L	7007.96	7252.80	3.49
2	LA= 27.77+0.915 L^2	7007.96	7538.500	7.03
3	Log LA= 2.187+1.079 Log L	7007.96	7092.33	1.18
	Models with leaf breadth (B) alone			
4	LA= -16.783+7.146 B	700.96	7134.30	1.80
5	LA= 23.169+0.311 B^2	7007.96	7626.76	8.80
6	Log LA= 1.131+1.233 Log B	7007.96	7104.76	1.38
	Models using both L and B			
7	LA = 1. 933+0.907 (LxB)	7007.96	6758.47	3.50
8	LA= 36.406+0.0052 $(LxB)^2$	7007.96	6945.79	0.88
9	Log LA= 0.029+0.965 log L+ 0.986 log B	7007.96	6156.00	3.91
	Models with leaf fresh weight (LFwt) alone			
10	LA= 7.732+60.761 LFwt	7007.96	7238.33	3.28
11	LA=39.996+24.226 LFwt2Log	7007.96	7474.32	6.65
12	Log LA=4.228+0.888 Log LFwt	7007.96	6156.01	12.15
	Models with leaf dry weight (LDwt) alone			
13	LA= 10.41+175.48 LDwt	7007.96	7238.33	3.18
14	LA= 41.494+210.72 $LDwt^2$	7007.96	7541.83	7.60
15	Log LA = 5.21+0.888 Log LDwt	7007.96	7148.67	2.00

All logarithms are to the base e, Selected Models are bold ones; [a] Percentage of deviation = (Total actual leaf area - total predicted area) x 100/total actual leaf

High R^2, low SEM and small percentage of deviation are considered for selection indices in regression models of estimation of any LA measurement. In the present study for regression models # 1, 7, 10, and 13 had high R^2, low SEM and minimum percentage of deviation (0.755, 108.28 and 3.49; 0.974, 11.31 and 3.50; 0.906, 41.73 and 3.28; and 8.614, 84.47 and 3.18 , respectively) (Tables 2-5). These models were tested for accuracy by plotting actual LA versus estimated (predicted) LA using regression models and results showed that there was a high co-linearity (Fig. 2). Therefore, these, models could be used to estimate LA. Of the four Models # (1, 7, 10 and 13) used to estimate LA in Philippine Morphotype of Cassava, Models # 1 and 7 were non-destructive and unsophisticated, and Models # 10 and 13 were destructive ones (Tables 2- 5). Such estimation of LA from leaf length and width, fresh and dry weights of leaf were also used by Ramkhelawan and Brathwaite (1990) in sour orange; Paiari *et al*. (2002) in pointed gourd; Son *et al.* (2002) in tomato and Williams and Martinson in grape (2003).

In Philippine Morphotype of Cassava all the variables showed significant correlation coefficients when correlated with their respective dependent variables. The value of r was between 0.031and 0.987 (Table 1). Regression Models # 1, 7, 10 and 13 showed good predictive ability, lower mean square error and small percentage of deviation. These Models were accurate, rapid and unsophisticated and, therefore, can be used for estimation of leaf area in Philippine Morphotype of Cassava.

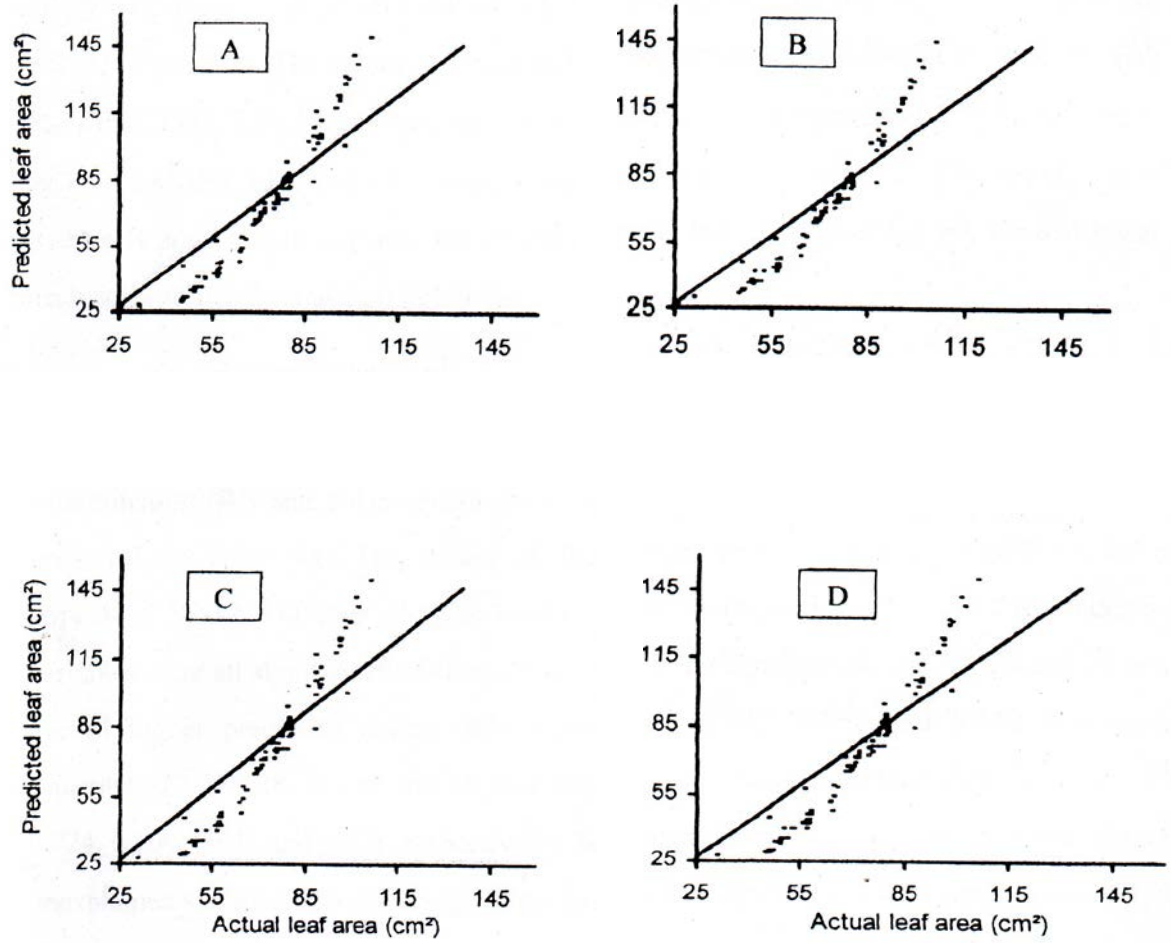

Fig. 2. Comparison of leaf area (LA) estimated from the four regression models with actual leaf area of 100 leaves in Cassava (Morphotype: Philippine) in which (A) the predicted leaf area (LA= -9.96+12.128 L) was derived from the model # 1, (B) the predicted leaf area (LA= 1.933+0.909 LxB) was derived from the model # 7, (C) the predicted leaf area (LA= 7.732+60.761L.F_{wt}) was derived from the model # 10, and (D) the predicted leaf area (LA= 10.41+175.48L.D_w) was derived from the model # 13

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