Determination of a Mathematical Model to Estimate the Area and Dry Weight of the Leaf Limbo of *Prunus persica* cv. Jarillo

Determinación de un Modelo Matemático para la Estimación del Área Foliar y Peso Seco del Limbo de *Prunus persica* cv. Jarillo

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Abstract. *A study was conducted to determine the variables that estimated the leaf limbo area and the leaf limbo dry weight of peach* ***Prunus persica*** *(L.) Batsch cv. Jarillo. Fifty leaves, aged 2.5 months, were selected and measured: leaf limbo length and width, petiole length, leaf length, petiole diameter, leaf limbo fresh weight, petiole fresh weight, leaf fresh weight, leaf limbo dry weight, petiole dry weight, leaf dry weight, length/width limbo, petiole length/limbo length and leaf limbo area. The results allowed to obtain regression equations for estimating the leaf area and the limbo dry weight. Using the lineal models LA =* $\beta_1 + \beta_2$ *(LLL x LLW) and LA=* $\beta_1 + \beta_2$*LLL +* β_3*LLW a leaf area equation was determined. Alternative models to calculate limbo dry weight were evaluated LLDW = -*$\beta_1 + \beta_2$ *LLFW and LLDW= -*$\beta_1 + \beta_2$*LLL +* β_3*PL. The best equations found with an* R^2 *of 0.99 were LA = 1.572 + 0.65169(LLL x LLW), LA=-23.106+2.8064LLW + 3.6761LLL and LLDW = -0.002+0.401(LLFW).*

Key words: Fruits, regression models, growth and development, statistical methods.

Resumen. *Se realizó un estudio para determinar las variables que estimaran el área del limbo foliar y el peso seco del limbo de durazno* ***Prunus persica*** *(L.) Batsch cv. Jarillo. Se seleccionaron cincuenta hojas con 2,5 meses de edad, fueron medidos: ancho del limbo, longitud del limbo, longitud del peciolo, longitud hoja, diámetro peciolo, peso fresco del limbo, peso fresco del peciolo, peso fresco de la hoja, peso seco del limbo, peso seco peciolo, peso seco de la hoja, longitud /ancho limbo, longitud del peciolo/longitud del limbo, área foliar del limbo. Los resultados alcanzados permitieron obtener ecuaciones de regresión para estimar el área foliar del limbo y el peso seco del limbo. Se halló una ecuación para la determinación del área foliar del limbo con los modelos lineales LA =* $\beta_1 + \beta_2$ *(LLL x LLW) y LA=* $\beta_1 + \beta_2$*LLL +* β_3*LLW. También se evaluaron modelos alternativas para calcular el peso seco del limbo, LLDW = -*$\beta_1 + \beta_2$*LLFW y LLDW= -* $\beta_1 + \beta_2$*LLL +* β_3*PL. Las mejores ecuaciones encontradas con un* R^2 *del 0,99 fueron LA = 1,572 + 0,65169(LLL x LLW), LA=-23,106+2,8064LLW + 3,6761LLL y LLDW = -0,002+0,401(LLFW).*

Palabras clave: Frutales, modelos de regresión, crecimiento y desarrollo, métodos estadísticos.

Mathematical models are a simplified representation of real systems allowing one to understand, explain, estimate or predict the reality (Heiner, 2007; Thornley, 1976, cited by Curiel *et al.*, 2007). In this regard, linear and multiple regression methods and experimental variables data associated with plant organs, such as leaf area and dry weight among others, enables the construction of mathematical models to estimate the plants photosynthetic capacity. Studies on the methods of estimating the leaf limbo area are in most cases costly, time consuming and sometimes it may cause severe plant damage. Casierra *et al.* (2008) claims that the leaves length and width are reliable values and when applied in regression equations it accurately reflects the leaf limbo area. It is, therefore, important to establish mathematical models based on simple measures that can estimate the area and dry weight of the leaf limbo without damaging plant material.

There is a close relationship between the leaf limbo area and its linear parameters (e.g. length and width), and such relationship can be described by regression equations (Simón and Trujillo, 1990; Venturieri, 1996; Murillo *et al.*, 2004; Muñoz *et al.*, 2008; Espitia *et al.*, 2006; Ruiz *et al.*, 2007; Galindo and Clavijo, 2007; Cardona *et al.*, 2009; Ruiz *et al.*, 2010; Wang and Zhang, 2012). Factors that can predict the production of dry weight, such as the amount of chlorophyll, temperature and photoperiod, have been directly related to the leaf area (Campostrini and Yamanishi 2001; Birch *et al.*, 2003).

Several authors have shown that the variation of the leaf area is the most significant factor in the variation

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of dry weight accumulated by plants (Gutiérrez and Lavín, 2000; Astegiano *et al*., 2001; Disegna *et al*., 2005; Muñoz *et al*., 2008). It has also been shown that the plants photosynthesis capacity is directly related to the limbo surface expressed as leaf area index (Kozlowski *et al*., 1991; Calderón *et al*., 2009). In the case of peach [*Prunus persica* (L) Batsch] its photoassimiliate production is mainly in leaves and its size corresponds to the leaf area (Marquínez and Corchuelo, 1998). The leaf area index (LAI) is one of the most commonly used parameters for the structural analysis of crop canopy and it relates the plants leaf area with the section of ground area (Arias *et al*., 2007). Full grown peach crop intercepts 95% of incident light with different LAI values. The LAI of this specific percentage is known as critical leaf area index and is determined by leaf area per plant (Montaldi, 1995; De la Casa *et al*., 2007; Acosta *et al*., 2008; Tinoco *et al*., 2008). Therefore, the aim of this study was to generate an empirical mathematical model applying multiple linear regression techniques, in order to estimate the leaf limbo area and leaf limbo dry weight of peach plants [*P. persica* (L.) Batsch cv. Jarillo].

MATERIALS AND METHODS

Characteristics of the experimental farm. The geo-referenced position of the peach trees (*P. persica* (L) Batsch cv. Jarillo) and their crop age (four to six years) were considered for the selection of the Las Delicias farm, located in the Province of Pamplona, Colombia (Chíchira). Fifty sample leaves, aged 2.5 months of five random trees, were selected and their characteristics are given in Table 1.

Table 1. Characteristics of the experimental farm in the province of Pamplona, Colombia.

Province	Village	Farm	Height above sea level (m)	Crop age (years)	Sowing distance (m)
Pamplona, North of Santander	Chíchira	Las Delicias	2170	5	9 X 9

Estimated leaf area using non-destructive method. The leaf limbo length (LLL) and the leaf limbo width (LLW) of fifty photosynthetically active and mature peach leaves were determined according to the reported procedure (Kumar, 2009). The product of the combination of LLL x LLW was the area of a rectangle that has as its base the leaf limbo length (LLL) and as its height the leaf limbo width (LLW).

The leaf limbo area was measured using an electronic scanner (Area Meter® AM300). Statistical analysis was performed with the aid of the statistical software package R and a scatter diagram was generated according to Murillo *et al*. (2004) and Ruiz *et al*. (2010) in *Vigna unguiculata.* The X-axis represents the product (length x width) of the leaf limbo area and the Y-axis the leaf limbo area. In order to estimate the leaf area the models used by Bianco *et al*. (2008a), Bianco *et al*. (2008b), Burgos *et al*. (2010) and Muñoz (1987) were considered:

$$LA = \beta_1 + \beta_2 (LLL \times LLW)$$

Where:

LA= Estimated leaf limbo area.
β_1 = Intersection.
β_2 = Increase in leaf limbo area when the combination of LLL x LLW is incremented by one unit
LLL x LLW = Length x width of the leaf limbo.

An alternative model by Ruiz *et al*., 2007 was also considered:

$$LA= \beta 1 + \beta 2LLL + \beta 3LLW$$

LA = Estimated leaf limbo area.
β_1 = Intersection.
β_2= Increase in leaf area when LLL is incremented by one unit.
β_3 = Increase in leaf area when LLW is incremented by one unit.

Estimated dry weight using non-destructive method. The dry weight of the fifty leaves was measured using a forced air circulation oven and an electronic scale.

A total of fourteen variables were used according to the methodology applied by Venturieri in 1996 in order to obtain an estimated dry weight model:

LLW = Leaf limbo width (cm); LLL = Leaf limbo length (cm); PL = Petiole length (cm), LL = Leaf length (LLL + PL) (cm); PD = Petiole diameter (cm); LLFW = Leaf limbo fresh weight (g); PFW = Petiole fresh weight (g); LFW = Leaf fresh weight (LFW + PFW) (g); LLDW = Leaf limbo dry weight (g); PDW = Petiole dry weight (g); LDW = Leaf dry weight (LLDW+ PDW) (g); LLL/LLW = Leaf limbo length/width (cm); PL/LLL = Petiole length/ Leaf limbo length (cm); LA = Leaf limbo area (cm^2).

Statistical analysis was performed with the aid of the statistical software package R. Correlation, simple and multiple linear regression analysis and Stepwise was used according to Bendel and Afifi (1977).

RESULTS AND DISCUSSION

Regression methods to determine leaf area of plants are an economic and useful tool in agronomic and plant physiology research (Casierra *et al*., 2008; Fallovo *et al*., 2008). It is also used to calculate the source-sink strength of leaf area and dry weight (Marquínez y Corchuelo, 1998). As were reported for many species, such as *P. persica* (Demirsoy *et al*., 2004), *Pisum sativum* (Galindo y Clavijo, 2007 y Ruiz *et al*., 2007), *Malus domestica* (Curiel *et al*., 2007) and *Zea mays* (Sezer *et al*., 2009), estimating leaf area from equations using leaf area measurement is a reliable and nondestructive method for accurately assessing leaf area. Therefore, in accordance with previous studies, the most appropriate mathematical approach to develop regression estimators and equations by using measured leaf parameters such as length and width were used.

As a result, the present study indicates that the leaf limbo length and width of *P. persica* (L.) Batsch cv. Jarillo presented the lowest coefficient variation values (<20%), considered low in agreement with previous findings of Zamora (1989). The regression equations generated from the leaf limbo and leaf limbo dry weight were obtained using descriptive statistics: minimum value, 25th percentile, mean and median, standard deviation, coefficient of variation, 75th percentile and maximum value (Table 2). It is important to highlight that the analyses of the fourteen variables using the Pearson coefficient (Table 3) showed that there was close relationship between them, thus allowing the selection of the independent variables more accurately: leaf limbo area and dry weight (Sokal y Rohlf, 1981).

Table 2. Descriptive statistics of the variables used to estimate the models in peach trees in the province of Pamplona, Colombia.

Statistics	Leaf limbo area (cm^2)	Leaf limbo width (cm)	Leaf limbo length (cm)	Petiole Length (cm)	Leaf Length (cm)	Petiole Diameter (cm)	Leaf limbo fresh weight (g)	Petiole fresh weight (g)	Leaf fresh weight (g)	Leaf limbo dry weight (g)	Petiole dry weight (g)	Leaf dry weight (g)
Minimum value	13.44	2.49	7.92	0.7	8.62	0.1	0.23	0.01	0.240	0.090	0.0	0.09
Percentile 25	28.32	3.54	11.17	1.1	12.27	0.1	0.54	0.01	0.54	0.21	0.0	0.21
Media	35.23	3.90	12.89	1.2	14.09	0.16	0.71	0.03	0.73	0.28	0.01	0.29
Median	37.09	4.07	13.22	1.2	14.42	0.2	0.71	0.03	0.740	0.28	0.01	0.29
Standard deviation	10.91	0.60	2.51	0.19	2.69	0.05	0.26	0.02	0.27	0.1	0.01	0.11
Variance coefficient (%)	30.96	15.33	19.47	15.92	19.11	30.27	36.36	57.5	37.05	36.75	61.7	37.42
Percentile 75	44.19	4.38	15.01	1.3	16.3	0.2	0.89	0.04	0.930	0.350	0.01	0.36
Maximum value	52.01	4.88	17.09	1.6	18.69	0.2	1.21	0.06	1.27	0.48	0.02	0.5

A Stepwise regression procedure was used to calculate the leaf limbo area and dry weight of *P. persica* (L.) Batsch cv. Jarillo and the independent variables are given in Tables 2 and 3. The statistical results of the thirteen estimation models obtained from such procedure are shown in Tables 4, 5, 6 and 7. It should be noted that the R^2 in all models were equal to 0.99 (P<0.01) and the inclusion of a large number of independent variables did not improve the model´s adjustment, as were reported in other species such as *Xanthosoma sagittifolium* (Simón y Trujillo, 1990). Therefore, from a statistical perspective any of the applied methods can be used to estimate leaf limbo area and dry weight of *P. persica* (L.) Batsch cv. Jarillo. Nevertheless, from a practical point of view, the ones that take into account the leaf´s limbo length and width are considered optimal. It is also important to emphasize that the models obtained from the present study confirms the assumptions of simple and multiple regressions methods such as normality errors, non-autocorrelated errors and homogeneity of variance, similar to those in *Carica papaya* (Cardona *et al*., 2009).

Table 3. Pearson correlation coefficient between variables to estimate the models in peach trees in the province of Pamplona, Colombia, Colombia.

	[1]LLW	[2]LLL	[3]PL	[4]LL	[5]PD	[6]LLFW	[7]PFW	[8]LFW	[9]LLDW	[10]PDW	[11]LDW	[12]LLL/LLW	[13]PL/LLL	[14]LA
LLW	---	0.98 [15]**	0.92**	0.98**	0.83**	0.96**	0.86**	0.96**	0.97**	0.89**	0.96**	0.94**	0.81**	0.98**
LLL		----	0.96**	0.99**	0.84**	0.99**	0.93**	0.99**	0.99**	0.95**	0.99**	0.95**	0.88**	0.99**
PL			----	0.96**	0.78**	0.97**	0.94**	0.97**	0.97**	0.94**	0.97**	0.96**	0.92**	0.95**
LL				-----	0.84**	0.99**	0.94**	0.99**	0.99**	0.95**	0.99**	0.95**	0.88**	0.99**
PD					----	0.79**	0.79**	0.79**	0.81**	0.83**	0.81**	0.73**	0.63**	0.86**
LLFW						------	0.95**	0.99**	0.99**	0.96**	0.99**	0.95**	0.92**	0.99**
PFW							------	0.96**	0.95**	0.99**	0.95**	0.87**	0.93**	0.92**
LFW								-----	0.99**	0.96**	0.99**	0.95**	0.93**	0.98**
LLDW									-----	0.96**	0.99**	0.95**	0.92**	0.99**
PDW										------	0.97**	0.90**	0.92**	0.94**
LDW											-------	0.95**	0.92**	0.99**
LLL/LLW												-------	0.88**	0.94**
PL/ LLL													-------	0.86**
LA														------

[1] Leaf limbo width; [2]Leaf limbo lenght; [3]Petiole length; [4] Leaf length; [5] Petiole diameter;[6] Leaf limbo fresh weight; [7] Petiole fresh weight; [8] Leaf fresh weight; [9] Leaf limbo dry weight; [10] Petiole dry weight; [11] Leaf dry weight; [12] Length/width leaf limbo; [13] Petiole length/leaf limbo length; [14] Leaf limbo area; [15] Highly significant.

Table 4. Leaf limbo area regressions in peach tress in the province of Pamplona, Colombia.

General equations	**1**	**2**	**3**	**4**
	[1]LA= 123.18098 [2]LLDW	LA= 1.04219 [3]LLL +77.92906 LLDW	LA= 1.58128LLL +87.77627LLDW -107.63413 x [4]PL/LLL	LA=1.36943LONL+13.14513 [5]PD+ 84.57134 [6]PDW -91.10593 PL/LLL
R^2 of model	0.99	0.99	0.99	0.99
Significance of model	[7]* *	* *	* *	* *
Significance value of estimators β_1, β_2 and β_3	**	** and **	**, ** and**	**, ** and **

[1] Leaf limbo area; [2] Leaf limbo dry weight; [3] Leaf limbo length; [4] Petiole length; [5] Petiole diameter; [6] Petiole dry weight; [7] Highly significant.

Table 5. Leaf limbo area regressions in peach tress in the province of Pamplona, Colombia.

General equations	**5**	**6**
	[1]LA=-23.106+2.8064 [2]LLW + 3.6761 [3]LLL	LA = 1.572 + 0.65169 (LLL x LLW)
R^2 of model	0.99	0.99
Significance of model	[4]* *	* *
Significance value of estimators β_1, β_2 and β_3	[5]***, ** and ***	** and **

[1] Leaf limbo area; [2] Leaf limbo width; [3] Leaf limbo length; [4] Highly significant; [5] Very highly significant

Table 6. Leaf limbo dry weight regression in peach tress in the province of Pamplona, Colombia.

General equations	1	2	3	4
	[1] LLDW = -0.002+0.401 [2]LLFW	LLDW = 0.06043 [3]PD + 0.38424 LLFW	LLDW= -0.01137 [4]PL +0.10195 PD –0.39320 LLFW	LLDW=0.00160 [5]LLW -0.01585PL+0.34439LLFW
R^2 of model	0.99	0.99	0.99	0.99
Significance of model	[6]* *	* *	* *	* *
Significance value of estimators β_1, β_2 and β_3	** and **	** and **	** , ** and **	**, ** and **

[1]Leaf limbo dry weight; [2] Leaf limbo fresh weight; [3] Petiole diameter; [4] Petiole length; [5] Leaf limbo width; [6] Highly significant.

Table 7. Leaf limbo dry weight regression in peach tress in the province of Pamplona, Colombia.

General equations	5	6	7
	[1]LLDW=0.00144 [2]LLW +0.30513 [3]LLFW+1.68597x [4]PDW	LLDW=0,00120 LLW +1.04704 LLFW-0.72331 [5]LFW +3.44587 PDW	LLDW = - 0.276802 + 0.03163 [6]LLL + 0.124152 [7]PL
R^2 of model	0.99	0.99	0.99
Significance of model	[8]* *	* *	* *
Significance value of estimators β_1, β_2, β_3 and β_4	**, ** and **	**, **, ** and **	** and **

[1] Leaf limbo dry weight; [2] Leaf limbo width; [3] Leaf limbo fresh weight; [4] Petiole dry weight; [5] Leaf fresh weight; [6] Leaf limbo length; [7] Petiole length; [8] Highly significant

In order to determine a mathematical model to predict nondestructive leaf limbo area of *P. persica* cv. Jarillo, six equations were generated and evaluated (Tables 4 and 5). The first regression model analyzed, when using the product of the leaf´s limbo length and width (*LLL x LLW*) with a proportional coefficient (β_2), as found in other species such as *Persea bombycina* (Chattopadhyay *et al*., 2011), *Mangifera indica* (Ghoreishi *et al*., 2012) and *Jatropha curcas* (Pompelli *et al*., 2012), was $LA = \beta_1 + \beta_2\ (LLL \times LLW)$. The second multiple lineal regression model used to estimate the leaf limbo area with two proportional coefficient (β_2) and (β_3); also found in *Vitis vinifera* (Legorburo *et al*., 2007); *Theobroma grandiflorum* (Venturieri, 1996), *S. rebaudiana* (Espitia *et al*., 2006), *Ocimum basilicum* (Ruiz *et al*., 2007), *P. sativum* (Galindo and Clavijo, 2007), *Merremia cissoides* (De Carvalho *et al*., 2011) and *Triticum aestivum* (Cogliatti *et al*., 2010), was $LA = \beta_1 + \beta_2\ LLL + \beta_3 LLW$.

In this regard, when evaluating the results of the above equations, the 0.99 coefficient of determination obtained in *P. persica* (L.) Batsch cv. Jarillo and in other varieties of *P. persica* (Demirsoy *et al*., 2004) suggests that 99 of the observed points is due to a high level of statistical significance ($P<0.01$). These findings are similar to those of *Nicotiana tabacum* (Bozhinova, 2006), *Actinidia deliciosa* (Mendoza *et al*., 2007), *Psidium guajava* (Singh, 2007), *Sida rhombifolia* y *Sida cordifolia* (Bianco *et al*., 2008a), *Ageratum conyzoides* (Bianco *et al*., 2008b) and *Xanthium strumarium* (Bianco *et al*., 2010). Therefore, the results reported in the present study, together with previous reports in other species such as *O. basilicum* (Ruiz *et al*., 2007), have shown that the two best equations to calculate the leaf limbo area of *P. persica* (L.) Batsch cv. Jarillo with an R^2 of 0.99 were: $LA = 1.572 + 0.65169\ (LLL \times LLW)$ (Figure 1) and $LA=-23.106+2.8064LLW + 3.6761LLL$ (Figure 2). This confirms that leaf length and width measurements are the most frequently dimensions used to estimate leaf limbo area as it can be easily used in the field due to its simplicity and accuracy (Mendoza *et al*., 2007).

As mentioned previously Stepwise analysis regression was also performed to predict dry weight of *P. persica* (L.) Batsch cv. Jarillo leaves and seven equations were generated and evaluated (Tables 6 and 7). Results showed that the best equation to calculate dry weight with a 0.99 R^2 was $LLDW = -0.002+0.401(LLFW)$ (Figure 3). These findings confirm those of *O. basilicum* (Ruiz *et al*., 2007).

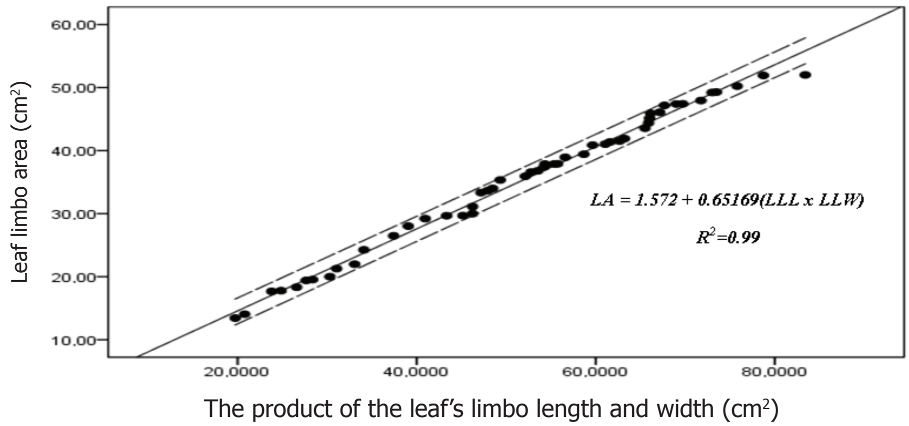

Figure 1. Linear regression with 95 % prediction limits of the relations between Leaf limbo area (*LA*) determined with the product of the leaf´s limbo length and width (*LLL x LLW*) in peach trees in the province of Pamplona, Colombia.

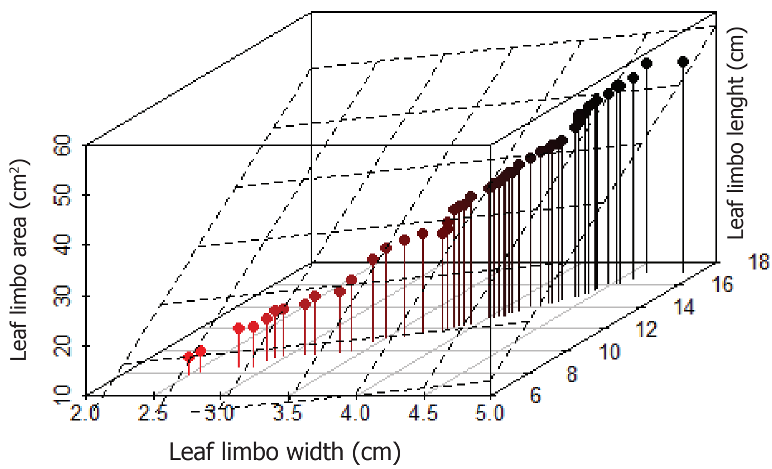

Figure 2. Multiple linear regression *LA=-23.106+2.8064LLW + 3.6761 LLL*, relationship between leaf limbo area (*LA*) with leaf limbo width (*LLW*) and leaf limbo length (*LLL*) in peach trees in the province of Pamplona, Colombia.

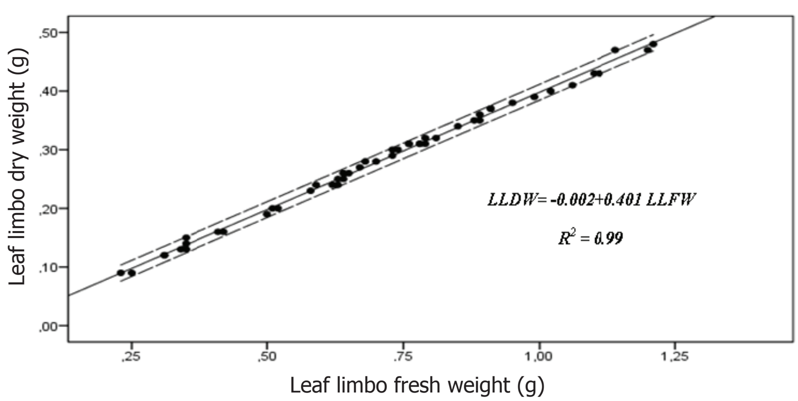

Figure 3. Linear regression with 95 % prediction limits of the relations between Leaf limbo dry weights (*LLDW*) determined with the leaf limbo fresh weight (*LLFW*) in peach trees in the province of Pamplona, Colombia.

To summarize we can conclude that the present study validated the models and methodology (Demirsoy *et al*., 2004) used to estimate the leaf area and dry weight of *P. persica* (L.) Batsch cv. Jarillo. It is important to highlight that these models meet statistical parameters and are adjusted to the Colombian tropical conditions.

CONCLUSIONS

The models analyzed in this study can be used to estimate the leaf area and dry weight of peach trees.

The most practical options to estimate the limbo leaf area of peach plants, *P. persicae* (L.) Batsch cv. Jarillo were: a) *LA =1.572 + 0.65169(LLL x LLW)* and b) *LA = -23.106+2.8064 LLW + 3.6761LLL* and for the leaf limbo dry weight: *LLDW= -0.002+0.401(LLFW).*

The simple lineal and multiple regressions showed that the chosen variables to calculate the models are linearly related to the leaf limbo area.

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