# Leaf biomass and leaf area equations for three planted trees in Iran

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#### ABSTRACT

Leaf area (LA) and leaf biomass (LB) are important variables for most physiological, horticultural and agronomic studies involving plant growth, development rate, radiation use efficiency, and water or nutrient use. Measuring these variables need destructive and aggressive sampling. Fortunately, evolving allometric equations can help for low cost and non-destructive estimation of such variables. The aims of this study are Estimate, compare and develop allometric models of LA and LB per tree and per stand for Alnus subcordata (AS), Populus deltoides (PD) and Taxodium distichum (TD) plantations. We selected 12 sample trees in each stand. Leaf Fresh weight of randomly selected branches was weighted in the field. Branchlevel LA was modeled as a function of branch diameter ( $R^2 > 0.8$ ) and total fresh weight of LB has been calculated for each sampled tree. For each species, 100 leaves from all canopy directions of trees were randomly selected and transported to the laboratory. At the lab, leaf area has been measured using leaf area meter. Allometric equations were derived using regression analysis. For all species, derived equations showed high accuracy (R<sup>2</sup> ranged from 0.837 to 0.947). However, with respect to mean square error, power regression equations

(individual leaf area =  $a(L \times W)b$  and LA or LB = a DBH<sup>b</sup>) are best models to estimate Individual Leaf Area, LA, and LB of AS, PD, and PD. The highest LAI was in the order of 16.9 > 5.5 > 4.5 for AS, PD, and TD, respectively.

**Keywords:** Non-destructive sampling; Regression analysis; Broad-leaved; Needle-leaved.

#### **1. INTRODUCTION**

Forests play a major role in the flow energy and material exchange between the land and atmosphere [1]. Leaf surface, as the main exchange surface, is a key important indicator in biological studies [2]. Leaf surface area and leaf weights are two important factors that affect many tree and stand-level processes and functions (including photosynthesis, gas exchange, growth, development rate, radiation use efficiency, water and nutrient use, nutrient cycling stand productivity and canopy dynamics [3, 4-6].

Leaf Area Index (LAI) is one of the most important parameters for analyzing the structure of the canopy; because it explains the response of plants to the environmental condition of sites, whether these changes are natural or anthropogenic [7, 8]. LAI is defined as the total area of leaves per unit area of land, leaf area (LA) is defined as the one-sided projected surface area [3, 4, 9], and leaf biomass (LB) is the total dry weight of leaves of a tree. LA and LB estimation methods are divided into two direct and indirect categories [7]. In the direct method, the leaves surface and dry weight are measured directly. The major limitations of these methods are being costly and destructive. The fact that makes indirect methods more popular among scientists.

Indirect method models the relationship of these attributes to those which are readily measurable. This approach models LA or LB based on one or more easily measured dimensions of the tree, which makes the method cheap, rapid, reliable and non-destructive [10]. There is a general relationship between the amount of LA, diameter at breast height (DBH), height (H), and biomass production. DBH is the most commonly used variable for modeling LA and LB [11], while some researchers reported that LB and LA estimation has been significantly improved by adding variable relating to the crown structure to the models [12].

Adl [13] estimated LB and LAI of Quercus brantii and Pistacia mutica in Zagros forests. LB for Quercus brantii and Pistacia mutica were 1317.3 and 57.2 (kg ha<sup>-1</sup>), respectively and mean LAI was 1.20. Babaei Kafaki et al. [14] estimated LAI and LB (3.33 and 1864 kg ha<sup>-1</sup>, respectively) for Quercus macranthera coppice stand at the northeast of Khalkhal in Iran. Pourhashemi et al. [15] estimated LB and LAI (37 kg and 3.7, respectively) for Celtis Caucasica using direct method (leaf gathering from the crown) in the oldest urban forests of Sanandaj city in Iran. Also, the compound variable,  $DBH^2 \times H$ , was the most efficient factor to estimate the LB ( $R^2=0.69$ ), but in LAI equation, the root of DBH was the best variable ( $R^2=0.72$ ). Kumar Sarker et al. [16] developed and compared 16 different allometric equations based tree DBH and H for predicting LA and LB of Artocarpus chaplasha in Bangladesh. Their result showed that the models based on only single predictor of tree DBH have more statistical accuracy among all models. Rance et al. [17] reported that the LA and LB are related to stem cross-sectional area. However studies about LA and LB are scarce, and model quantification of LA and LB for fast-growing trees is even rarer. The aims of this study are

Estimate, compare and develop allometric models of LB and LA per tree and per stand for three native and introduced tree species in northern Iran.

#### 2. MATERIAL AND METHODS

#### 2.1. Site description

The study site is located on the southern coast of the Caspian Sea, 10 Km from Amol city, north of Iran 36°35'N", 52°10'E", 5 m above sea level. Rainfall with wetter months occurs between September and March, and a dry season occurs from April to August. The climate is temperate, with a mean annual temperature of 16.9 °C and a mean annual precipitation of 823.5 mm. The soil of plantations is poor drainage and has a silty loam texture with pH 7.6-8.1.

#### 2.2. Stand description

The study site was established in 1992 with the aim of wood production using three rapid growth species including alder (*Alnus subcordata* C.A. Mey) (AS), eastern cottonwood (*Populus deltoides* Bartr. Ex Marsh) (PD) and bald cypress (*Taxodium distichum* L.C. Rich) (TD). The site where previously covered by natural stands dominated by *Carpinus betulus* and *Parrotia perssica* [10]. Tree spacing in all stands is  $4 \times 4$  m. No thinning operations were made in these plantations.

#### 2.3. Sampling design

36 sampling plots with dimensions of  $16 \times 16$  m were selected randomly in each of the three stands [18]. In each plot, the DBH of individual trees was measured with calipers, the crown width (CW) measured with a tape, height (H) and the crown height (CH) was measured with a Haglöf-VERTEX IV clinometer. The average values of tree DBH, H, CH, and CW of stands are summarized in Table 1.

The diameter range of all trees of each species was divided into 12-diameter size classes (class size was 3 cm for all species). From every size class, one representative tree was selected and destructively sampled. In total, 36 trees were sampled; 12 trees per species. The trees were felled down with a chainsaw. Prior to branch removal, the diameter of each branch was measured and five representative branches from the smallest to the largest branch throughout the crown were sampled. All branches were then clipped from the tree, and the leaves were collected in order to obtain total fresh weight at the site using a hanging scale to the nearest 0.1 kg. Three samples of 200 g of leafs were collected from each tree and taken to the laboratory in sealed plastic bags, dried at 70 °C until achieving a stable weight. Additionally, 100 leaves per species were collected in randomly and were transferred to the laboratory and individual LA, leaf length (LL) and leaf width (LW) were measured with a leaf area meter (type CI-202, USA; see Table 2). Subsequently, the leaves were dried in an oven at 65 °C until they reached a constant dry weight and then weighed on a scale with a precision of three digits. All measurements were made in late of the growing season.

**Table 1.** Summary characteristic of Stands, Diameter at Breast Height (DBH), Height (H), Crown Height (CH), Crown Width (CW).

Species	Variable	Mean	Standard Deviation	Minimum	Maximum	Skewness	Kurtosis
	DBH (Cm)	31.32	7.70	15.92	50.96	0.39	-0.32
AS	H (m)	21.99	4.40	10.10	31.80	-0.16	-0.35
$n/ha = 459 \pm 51$	CH (m)	8.91	2.70	2.90	16.40	0.37	-0.28
	CW (m)	4.02	1.39	1.20	7.35	0.21	-0.65
	DBH (Cm)	27.43	6.36	12.42	43.95	0.00	-0.25
PD	H (m)	29.96	4.96	16.90	44.20	-0.21	0.34
$n/ha = 557 \pm 38$	CH (m)	11.54	3.41	4.00	20.90	0.14	-0.35
	CW (m)	3.145	1.22	1.00	5.850	0.15	-0.79
	DBH (Cm)	28.45	6.86	11.27	47.05	0.01	-0.43
TD	H (m)	17.74	2.67	9.80	24.10	-0.33	0.48
$n/ha = 557 \pm 48$	CH (m)	6.67	1.95	1.10	14.90	0.24	1.58
	CW (m)	4.25	0.96	1.48	6.40	-0.24	0.13

**Table 2.** Means  $\pm$  standard error (SE), minimum (min) and maximum (max) values for the leaf length (L), width (W), leaf area (LA) and length (L)  $\times$  width (W) of the AS, PD, and TD.

Spacios	L (Cm)			W (Cm)			LA (Cm <sup>2</sup> )		
Species	Mean±SE	Min	Max	Mean±SE	Min	Max	Mean±SE	Min	Max
AS	9.0±0.3	2.3	19.9	7.8±0.2	4.5	12.3	54.9±3	12.7	188.9
PD	10.6±0.2	4.2	15.4	10.4±0.2	4.6	13.8	73.8±3	12.8	130.7
TD	4.1±0.1	2.1	7.9	1.9±0.1	0.7	3.6	3.3±0.2	0.5	8.9

To estimate LA and LB per hectare, we first calculated the LA ( $m^2$ ) and LB (Kg) by applying the corresponding adjusted allometric equation to all the trees in the plot area. Considering a sampling plot with an area of 256  $m^2$ , the LA and LB per hectare was obtained for every species.

#### 2.4. Statistical analysis

DBH of the harvested trees was used to prepare a simple linear and power function fitted to the data using least square regression. The model with the best goodness of fit was selected based on by SE (standard error) and adjusted  $R^2$  (coefficient of multiple determination). For comparing LB and LA of different plantations, one-way ANOVA was applied. The ANOVA was followed by a Dunnet-test to separate the species and the different leaf characteristics. All differences were considered significant at P<0.05. All statistical analysis were conducted on SPSS software, ver. 19.0 (SPSS Inc., Chicago, II., USA).

#### **3. RESULTS AND DISCUSSION**

#### 3.1. Predict leaf area

The regression analysis showed that most of the variation in LA values was explained by "length  $\times$  width" as the predictor variable (Table 3). The best fitting equations (showing the highest coefficient of determination, and the lowest standard error) were obtained for the individual leaf area by power model within all planted trees.

Figure 1 presents the relationships between LA (cm<sup>2</sup>), L×W and distribution of the residuals for each species. Visually, there is a good correlation between predictors and dependent variable for all species. For all species, based on the distribution of residuals, heteroscedasticity of residuals is obvious.

Estimating LA through measuring leaf

dimensions has been an interesting subject for many researchers. Most of these researchers used leaf length, leaf width, or combinations of these variables as predictors in allometric models [19, 20]. In some scientific work, it is not possible to measure these variables destructively, because the research needs to be continued while the plant is performing its functions. In this study, we used leaf length and leaf width combinations to establish allometric models for predicting leaves variables. Results from the present study were in accordance with some of the previous studies (including ref. [13, 19, 22]) on establishing reliable equations for predicting LA through measuring leaf dimensions. Some researches, as well as our research, showed that LA and LB of leaves can be predicted using leaves dimensions with high accuracy.

## **3.2.** Allometric equations for tree leaf area and LB

The average values for LA and LB of AS, PD, and TD trees are summarized in Table 4. The LA was 312.3  $m^2$  for AS, 85.3  $m^2$  for TD and 46.8  $m^2$  for PD. In addition, The LB was 30.89, 13.94, and 5.09 Kg for AS, PD, and TD, respectively (Table 4).

**Table 3.** Fitted coefficient (b) and constant (a) values of the models used to estimate the individual leaf area (LA in  $cm^2$ ) of single leaves from the length (L) and width (W) measurements. A coefficient of determination ( $R^2$ ), mean square errors (MSE), and F of the various models are also given. L and W are in cm.

Species	Model	а	b	$\mathbf{R}^2$	MSE	<b>F</b> <sub>calc</sub>
AS	$LA=a(L\times W)+b$	0.746	-0.319	0.977	4.289	4071.25***
AS	$LA = a(L \times W)^b$	0.778	0.988	0.982	3.117	5253.99***
PD	$LA=a(L\times W)+b$	0.638	1.715	0.975	4.052	3891.84***
PD	$LA = a(L \times W)^b$	0.636	1.005	0.972	3.873	3435.64***
TD	$LA=a(L\times W)+b$	0.329	0.661	0.876	0.560	686.50***
	$LA = a(L \times W)^b$	0.561	0.855	0.923	0.243	1153.89***

Table 4. Means ± standard error (SE), minimum (min) and maximum (max) values for LA and LB of the AS, PD, and TD.

Species	]	$LA(m^2)$		LB (Kg)			
Species	Mean±SE	Min	Max	Mean±SE	Min	Max	
AS	312.3+21	51.5	750	30.89+2.1	5.19	75.71	
PD	46.8+3.4	4.6	97.3	5.09+0.31	0.52	11.15	
TD	85.3+7.1	19.5	263.3	13.94+1.2	2.19	46.31	

Table 5 summarizes the results of modeling LA and LB based on power equations. For all species and variables, the models were highly significant (*P-value* < 0.001). Coefficients of determination showed a strong relationship between tree DBH and LB (ranges from 0.837 to 0.947) for all species. Figure 2 presents the relation ships between individual leaf area (cm<sup>2</sup>), L×W (Cm), and distribution of the residuals for each species.

Based on multiple means comparison results, there were significant differences between LAI of the different plantation. Mean LAI of AS, PD and TD are 16.9 > 5.5 > and 4.5, respectively (Table 6). LB was also different significantly between species and for AS, PD, and TD and was 12032.5 > 2490.6 > and 6504.6 Kg ha<sup>-1</sup>, respectively.

**Table 5.** Fitted coefficient (a and b) of the models used to estimate LA (m<sup>2</sup>) and LB (kg) of a single tree from DBH (Cm). Coefficient of determination ( $\mathbb{R}^2$ ), mean square errors (MSE), and F ( $F_{calc}$ ) of the various models are also given.

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Variable	а	b	$\mathbf{R}^2$	MSE	<b>F</b> <sub>calc</sub>
LA	0.029	2.610	0.947	0.223	161.00***
LB	0.003	2.610	0.947	0.223	161.00***
LA	0.008	2.509	0.905	0.344	85.76***
LB	0.001	2.509	0.905	0.344	85.76***
LA	0.157	1.798	0.849	0.347	50.48***
LB	0.017	1.943	0.837	0.406	46.33***
	LA LB LA LB LA	LA 0.029   LB 0.003   LA 0.008   LB 0.001   LA 0.157	LA 0.029 2.610   LB 0.003 2.610   LA 0.008 2.509   LB 0.001 2.509   LB 0.157 1.798	LA0.0292.6100.947LB0.0032.6100.947LA0.0082.5090.905LB0.0012.5090.905LA0.1571.7980.849	LA0.0292.6100.9470.223LB0.0032.6100.9470.223LA0.0082.5090.9050.344LB0.0012.5090.9050.344LA0.1571.7980.8490.347

Table 6. Means  $\pm$  Standard error (SE) values for LAI and LB of the AS, PD, and TD.

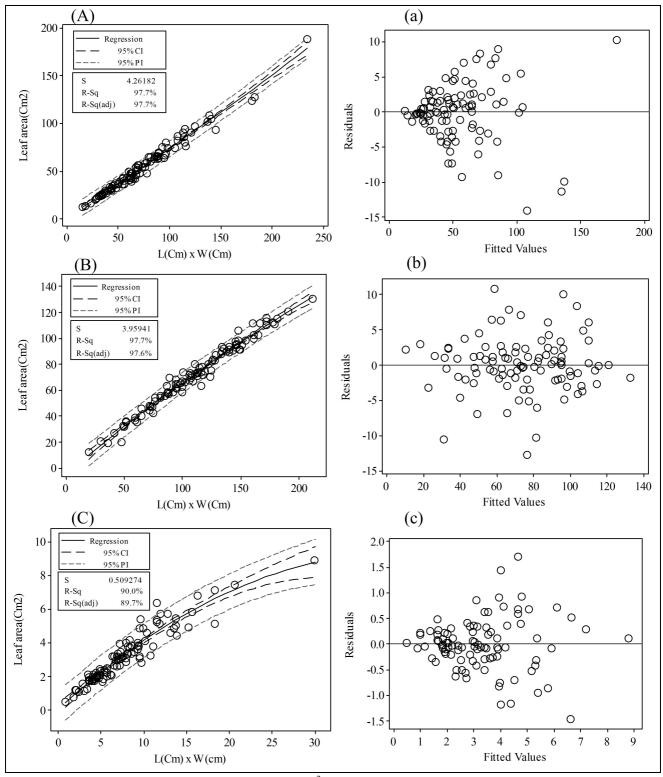
Species	LAI	LB (Kg/ha)
AS	16.95+7.65 a	12032.55±2235.84 a
PD	5.52+2.64 b	2490.62±201.02 b
TD	4.46+2.33 b	6506.6±686.7 c

The lowercase letter shows a significant difference at 0.05 significant level.

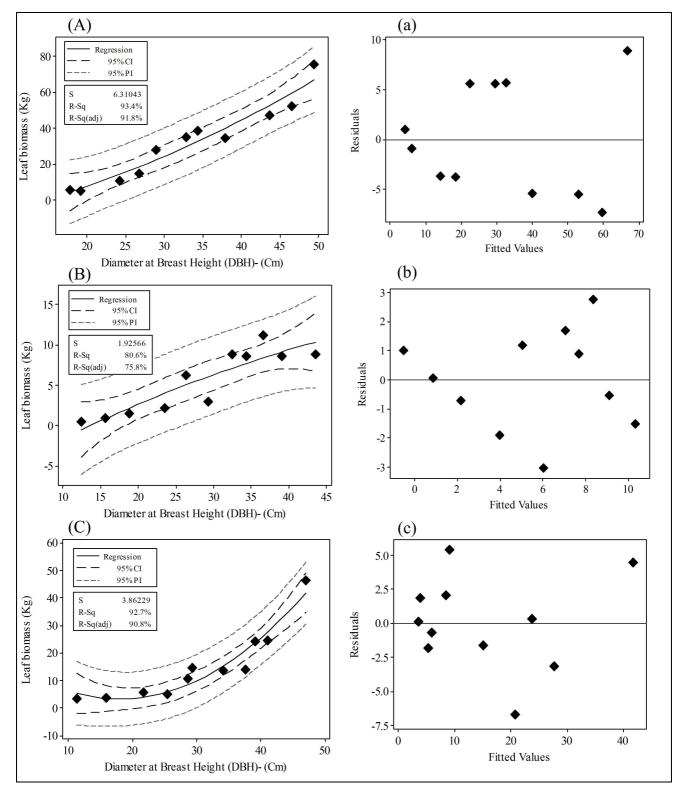
It has been proved that using allometric equation is a better alternative for estimating those variables which need to be destructively measured; because this method is not only environmentally friendly but also cost and time effective [23]. The results of this study showed that LA and LB can be estimated using easily measured tree variables such as diameter at the breast; all equations were highly significant ( $R^2 = 0.837 - 0.947$ ). The proposed models accounted for more than 80% of the variation based on DBH in the LA and LB models (Table 4 and 5) and provided a sound, nondestructive means to predict these canopy properties in fast growing trees. Therefore, a regression model can be a good alternative method for determining LA instead of devices of LA meter which is consistent with the findings of Calvo-Alvarado et al. [24], and Pokorný and Tomášková [25]. Socha and Wezyk [26] found that the diameter at breast height (DBH), explain more than 80% of the variation in LA and LB of Scots pine (*P. sylvestris* L.). Based on finding that reported by Grace et al. [27] and Vertessy et al. [28] DBH could explain 91% of the variation in LA of *Acacia koa* and *Eucalyptus regnans*, respectively.

Unfortunately, we could not compare our results with other similar works in similar forests because allometric equations on canopy properties are unavailable in northern Iran. However, further development of these equations through destructive methods and increased sample sizes would facilitate the development of regional estimates of LA and LB. Finally, the results of this study could serve as a basis for more precise quantification of fast growing tree physiological and environmental processes in the plantation of northern Iran, mostly for the areas being planted by AS, PD, and TD. The founding of

this study is important for the ecological purpose (including transpiration rate, biomass estimation, light interception, and carbon storage). In addition, this study is important for tree growth model of these fast growing species.



**Figure 1.** Relationship between individual leaf area (cm<sup>2</sup>), L(Cm) ×W(Cm), and L×W residuals against individual leaf area (A, B, and C are *A. subcordata*, *P. deltoides*, and *T. distichum*, respectively) (n = 100).



**Figure 2.** Relationship and residuals between DBH (X axis) and total tree LB in Kg for 12 harvested trees (Y axis) (A, B, and C are *A. subcordata*, *P. deltoides*, and *T. distichum*, respectively).

#### 4. CONCLUSIONS

This study is one of the few reports on the allometric relationship for estimate individual leaf area from length  $\times$  width (L×W) and estimate

LA and LB of plantation trees in Iran. Through regression analysis, it was found that there was a strong power relationship (coefficient of determination > 0.8) between LA, LB, and DBH within each of the planted trees, with a level of significant relationship. The analysis of mean square error between in linear and power models in three planted species show that the power regression equation (individual leaf area =  $a(L \times W)b$  - LA or LB = a DBH<sup>b</sup>) can best estimate of Individual Leaf Area, LA and LB for *Alnus subcordata* and *Populus deltoides*, and well for *Taxodium distichum*.

#### **AUTHORS' CONTRIBUTION**

JE: Field works and collecting the data, the laboratory analysis, running the data analysis, and writing the paper; HS: Designing the experiment, supervising the work, and writing the paper; SMH: Designing the experiment and supervising the work; BN: Supervising the field works and collecting the data. The final manuscript has been read and approved by all authors.

#### TRANSPARENCY DECLARATION

The authors declare no conflicts of interest.

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