Relation between leaf area and dimensions of selected medicinal plants

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ABSTRACT: In this research, leaf area prediction models were developed for some leaf-used medicinal plants namely *Calamintha nepeta, Datura stromonium, Melissa officinalis, Mentha piperita, Nerium oleander, Origanum onites* and *Urtica dioica* growing wild in Black Sea region of Turkey. Lamina width, length and leaf area were measured non-destructively to develop the models. The actual leaf areas of the plants were measured by PLACOM Digital Planimeter, and multiple regression analysis with Excel 7.0 computer package program was performed for the plants separately. The produced leaf area prediction models in the present study were formulized as $LA = (a) + (b_1 \times L) + [(b_2 \times (L \times W)] + (b_3 \times L^2) + (b_4 \times W^2) + [b_5 \times (L \times W^2)] + [b_6 \times (L^2 \times W)] + [b_7 \times (L^2 \times W^2)]$ where LA is leaf area, W is leaf width, L is leaf length and $a, b_1, b_2, b_3, b_4, b_5, b_6$, and b_7 are coefficients. R^2 values for medicinal plants tested varied with species from 0.82 in *Origanum onites* to 0.98 in *Urtica dioica*. All R^2 values and standard errors were found to be significant at the $P < 0.001$ level.

Keywords: modelling; leaf area; medicinal plants

Medicinal plants have been harvested from the wild since ancient times (Dhillion, Ampornpan 2000; Dhillion et al. 2002). Today, over 60% of the world's population, 80% in developing countries depend directly on plants for their medical purposes. Traditional medicine is still recognized as the primary health care system in many rural communities because of its effectiveness, lack of modern medical alternatives, and cultural preferences (Manandhar 1998; Tabuti et al. 2003).

Biologically active components of the plants evaluated here, namely hyoscyamine in *Datura stromonium* L. (Patterson, O'Hagan 2002), essential oil in *Calamintha nepeta* L. (Pooter et al. 1986), *Melissa officinalis* L. (Sadraei et al. 2003), *Mentha piperita* L. (Mucciarelli et al. 2001) and *Origanum onites* L. (Baydar et al. 2004), oleandrin in *Nerium oleander* L. (Afaq et al. 2004), vitamins, minerals and amino acids in *Urtica dioica* L. (Aksu, Kaya 2004) are found to be concentrated in leaves. Therefore leaves are the most important organs of those plants.

Leaf area is routinely measured in experiments concerning interesting crops where some physiological phenomena such as light, photosynthesis, respiration, plant water consumption and transpiration are being studied (Rieger, Duemmel 1992; Horsley, Gottschalk 1993; Gottschalk 1994; Kersteins, Hawes 1994; Picchioni, Weinbaum 1995; Uzun 1996; Centritto et al. 2000). In addition, leaf number and area of a plant are important in terms of cultural practices such as training, pruning, irrigation, fertilization etc. The leaf area estimation models that aim to predict leaf area non-destructively can provide researches with many advantages in agricultural experiments. Moreover, these kinds of models enable researchers to carry out leaf area measurements on the same plants over the course of a study, resulting in reduced experimental variability (Gamiely et al. 1991; NeSmith 1991, 1992). Leaf area can be determined by using expensive instruments and/or predictive models. Recently, new instruments, tools and machines such as hand scanners and laser optic apparatuses have been developed for leaf and fruit measurements. These are very expensive and complex devices for both basic and simple studies. Furthermore, non-destructive estimation of leaf area saves time as compared with geometric measurements (Robbins, Pharr 1987). For this reason, several leaf area prediction models were produced for some plant species in previous studies. But, to the authors' knowledge, there are no published reports related to leaf area prediction model for any medicinal plant. Due to the lack of such information, in the present study, we aimed to develop reliable equations that allow for the non-destructive estimation of leaf area through linear measurements of the aforesaid plant species.

MATERIAL AND METHODS

Brief description of plant materials

Calamintha nepeta (*Lamiaceae*) is a perennial herb with showy flowers, ovate leaves in outline with the apex usually sharp-pointed and a distinctive minty odour. Primarily a weed of pastures, fields, and noncrop areas. *Datura stromonium* (*Solanaceae*) is an annual, growing up to a height of 1.5 m and has trumpet shaped lavender flowers that release a fragrant smell. The leaves are deep green, large and oval with wavy margins. Plant develops large, prickly seed pods at each stem division. *Melissa officinalis* (*Lamiaceae*) is a perennial plant growing wild in fields and gardens and along road sides. It has an upright stem that grows as high as 1.5 m. Light green toothed ovate leaves grow in opposite pairs at each joint. The flowers may range in colour from pale yellow to rose coloured or blue white. When bruised, the whole plant smells like lemon. *Mentha piperita* (*Lamiaceae*) is a perennial herb up to 1 m high with underground runners. It has green stems, leaves and reddish-violet flowers. Leaves are shortly but distinctly stalked, 6 cm or more in length, a 2 to 4 cm broad and their margins finely toothed. *Nerium oleander* (*Apocynaceae*) is a shrub or small tree. It is native to the Mediterranean area and grows to about 9 m, with alternate, very straight, dark green leaves. Its flowers may be white, yellow, red, pink, or intermediate colours. Its fruit is a brown, podlike structure with many small seeds. *Urtica dioica* (*Urticaceae*) is a well-known perennial herb and highly successful 'weed' species. The oval-shaped leaves are easily recognized; they have deeply serrated edges and bear stinging hairs. Small greenish flowers in axillary clusters bloom in summer. *Origanum onites* (*Lamiaceae*) is an upright perennial herb native to Europe and naturalized in the Middle East. It can reach heights of 1.5 m. It has square, red stems, elliptical leaves, and clusters of deep pink flowers. The leaves are opposite, petiolate, about 2–3 cm long, nearly entire hairy beneath.

All plants described above are growing wild in Black Sea region of Turkey and were identified by Dr. Hasan Korkmaz, Department of Biology, University of 19 Mayis, Samsun, Turkey.

Experimental procedures

Leaf samples were randomly taken from wild populations of the medicinal plants tested between March and August 2004 at a five time intervals. In this period, 30 leaves were collected for each plant within the first three day of April, May, June, July and August to catch the different phases of leaf morphogenesis. Thus, 150 leaf samples for each plant and a total of 1,050 leaves for seven plants were processed at the same day as they were collected in the following manner. First, they were placed on the photocopier desktop by holding flat and secure and copied on A3 sheet (at 1:1 ratio). Second, a Placom Digital Planimeter (Sokkisha Planimeter Inc., Model KP-90) was used to measure actual leaf area of the copy. Selection of leaf dimensions for measurement was governed by variation in leaf characteristics (e.g., size, shape, and symmetry) and practical constraints (e.g., ease and accuracy of measurements under field conditions). Given these concerns, we chosed maximum leaf width (W) and length (L) to correlate with leaf area. Leaf width (cm) was measured from tip to tip at the widest part of the lamina and leaf length (cm) was measured from lamina tip to the point of petiole intersection along the midrib (Fig. 1). The leaf positions were selected with regard to points that could be easily identified and used to facilitate the measurement of leaf length and width.

Model constructions

Multiple regression analysis of the data was performed for each plant separately. A search for the best model for predicting leaf area was conducted with various subsets of the independent variables, namely, length (L), width (W), ($L^2 \times W$), ($L \times W$), ($L^2 \times W^2$), ($L \times W^2$) and (W^2). The best estimating equations for the leaf area (LA) of the plants tested were determined with the Excel 7.0 and formulized as $LA = (a) + (b_1 \times L) + [(b_2 \times (L \times W)] + (b_3 \times L^2) + (b_4 \times W^2) + [b_5 \times (L \times W^2)] + [b_6 \times (L^2 \times W)] + [b_7 \times (L^2 \times W^2)]$ where LA is leaf area, W is leaf width, L is leaf length and a, b_1, b_2, b_3, b_4, b_5, b_6, and b_7 are coefficients of the produced equation. Multiple regression analysis was carried out until the least sum of square was obtained.

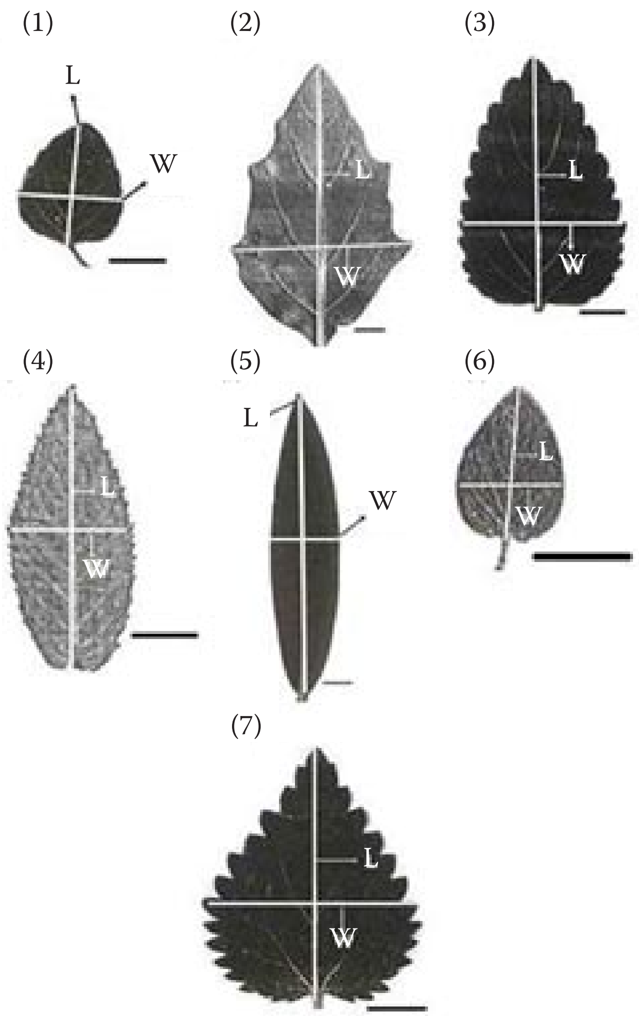


Fig. 1. Leaf diagram of *Calamintha nepeta* (1), *Datura stromonium* (2), *Melissa officinalis* (3), *Mentha piperita* (4), *Nerium oleander* (5), *Origanum onites* (6), and *Urtica dioica* (7), showing the position of leaf length (L) and leaf width (W). Bars seen at the lower right corner of each figure represent 1 cm

RESULTS

Multiple regression analysis used for determination of the best fitting equation for estimation of leaf area in the medicinal plants evaluated here showed that most of the variation in leaf area values was explained by the basic parameters (length and width). The variation explained by both parameters was 91% for *Calamintha nepeta* (SD = 0.593), 96% for *Datura stromonium* (SD = 12.300), 97% for *Melissa officinalis* (SD = 6.667), 93% for *Mentha piperita* (SD = 1.811), 98% for *Nerium oleander* (SD = 9.285), 82% for *Origanum onites* (SD = 0.328) and 98% for *Urtica dioica* (SD = 7.871). The produced leaf area prediction models in the present study were $LA = (a) + [b_2 \times (L \times W)] + [b_6 \times (L^2 \times W)]$ for *Calamintha nepeta*, $LA = (a) + (b_1 \times L) + (b_3 \times L^2) + [b_6 \times (L^2 \times W)] + [b_5 \times (L \times W^2)]$ for *Datura stromonium*, $LA = (a) + [b_5 \times (L \times W^2)] + [b_7 \times (L^2 \times W^2)]$ for *Melissa officinalis*, $LA = (a) + (b_4 \times W^2) + [b_6 \times (L^2 \times W)] + [b_2 \times (L \times W)]$ for *Mentha piperita*, $LA = (a) + [b_5 \times (L \times W^2)] + [b_6 \times (L^2 \times W)] + [b_7 \times (L^2 \times W^2)]$ for *Nerium oleander*, $LA = (a) + [b_6 \times (L^2 \times W)] + [b_5 \times (L \times W^2)]$ for *Origanum onites* and $LA = (a) + [b_6 \times (L^2 \times W)] + [b_5 \times (L \times W^2)] + [b_2 \times (L \times W)] + [b_7 \times (L^2 \times W^2)]$ for *Urtica dioica* (Table 1).

DISCUSSION

Investigations which correlate leaf length and width with leaf area are most common (Smith, Kliewer 1984; Elsner, Jubb 1988), but some studies also include petiole length (Manivel, Weaver 1974) and leaf weight (Montero et al. 2000). But, regression equations that incorporate leaf length and/or width are generally chosen for their simplicity and accuracy, and because these measurements are non-destructive. Thus, many studies have been carried out to estimate leaf area by linear measurements of leaf width and length in the following plants: soybean (Lieth et al. 1986), cucumbers (Robbins, Pharr 1987), orange (Arias et al. 1989; Ramkhelawan, Brathwaite 1992), French bean (Rai et al. 1990), coconut (Mathes et al. 1990), banana (Potdar, Pawar 1995), grape (Uzun, Çelik 1999), miscanthus (Vargas et al. 2002), broad

Table 1. The Equation of leaf area $LA = (a) + (b_1 \times L) + [b_2 \times (L \times W)] + (b_3 \times L^2) + (b_4 \times W^2) + [b_5 \times (L \times W^2)] + [b_6 \times (L^2 \times W)] + [b_7 \times (L^2 \times W^2)]$ for the medicinal plants tested

Plants		a	b_1	b_2	b_3	b_4	b_5	b_6	b_7	R^2
Calamintha nepeta	LA	−0.23554		1.067838				−0.1526		0.91
	SE	0.088***		0.105***				0.039***		
Datura stromonium	LA	19.39368	−8.55345		1.5604		0.3830632	−0.3006		0.96
	SE	4.46***	1.87***		0.25***		0.04***	0.04***		
Melissa officinalis	LA	3.233871					0.20786		−0.01064	0.97
	SE	0.27***					0.012***		0.001***	
Mentha piperita	LA	0.102834		0.887465		−0.24204		−0.0112		0.93
	SE	0.23***		0.039***		0.102***		0.0005***		
Nerium oleander	LA	6.24034					0.153101	0.034572	−0.00821	0.98
	SE	1.47***					0.022***	0.004***	0.001***	
Origanum onites	LA	0.676645					−0.044446	0.300381		0.82
	SE	0.035***					0.002***	0.011***		
Urtica dioica	LA	−1.1554		0.9781			−0.05403	−0.04145	0.006555	0.98
	SE	0.16***		0.011***			0.002***	0.002***	0.0005***	

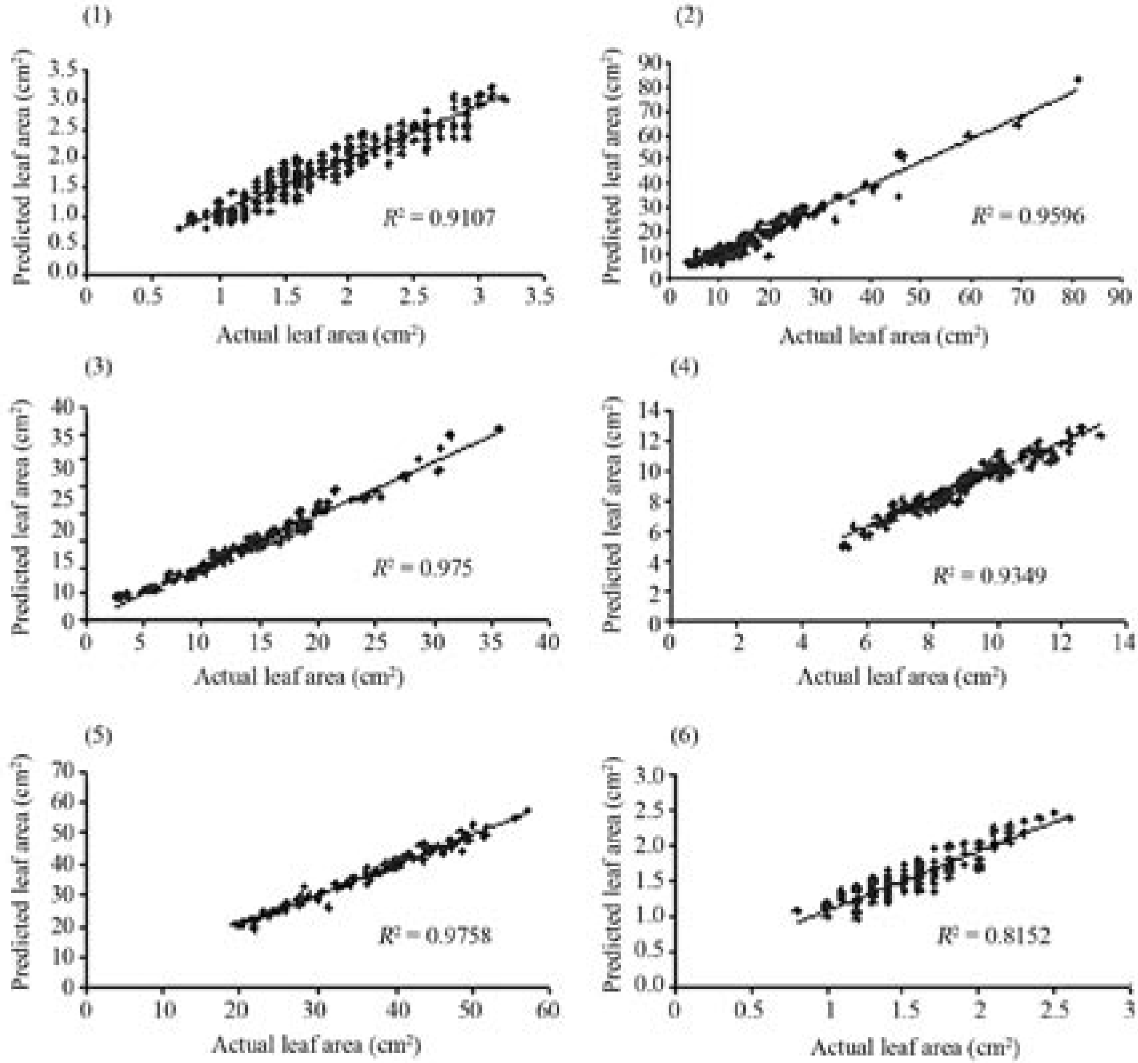
LA – leaf area, L – leaf length, W – leaf width, SE – standard error, a, b_1, b_2, b_3, b_4, b_5, b_6 and b_7 are coefficients of produced equation, ***R^2 and all SE values are significant at $P < 0.001$

bean (Odabaş 2003), cherry (Demirsoy, Demirsoy 2003) strawberry (Demirsoy et al. 2004a), peach (Demirsoy et al. 2004b) and summer snowflake (Çırak et al. 2004). The same authors found that there were close relationships between leaf area value, leaf length and leaf width for these plants (R^2 = 0.94 for soybean, R^2 = 0.76 to 0.99 for cucumber, R^2 = 0.89 to 0.93 for orange, R^2 = 0.99 for French bean, R^2 = 0.95 to 0.98 for coconut, R^2 = 0.96 for banana, R^2 = 0.98 for grapes, R^2 = 0.91 for miscanthus, R^2 = 0.99 for broad bean, R^2 = 0.95 for cherry, R^2 = 0.99 for strawberry, R^2 = 0.99 for peach and 0.97 for summer snowflake). We found that there was a very close relationship between actual and predicted leaf area for all medicinal plant tested (Fig. 2).

Our results were consistent with those of other studies mentioned above that used linear measurements of leaves from different plants for estimating leaf area. Coefficients of determination were generally high ($R^2 > 0.95$) for the best-fit models in the current and previous studies. It is interesting to note that R^2 values for medicinal plants tested varied with species greatly from 0.82 in *Origanum onites* to 0.98 in *Urtica dioica*. But, the difference is not surprising given the evident differences in size and shape of leaves among the species (Fig. 1). Apart from different species, regression coefficients of leaf area estimation can even be different between the cultivars of the same species. For example, using the grapevine cultivars Niagara and DeChaunac, Willams and Martinson (2003) found that the product of maximum leaf length and width was most highly correlated with leaf area, but R^2 values of cultivars were different (0.99 for Niagara and 0.96 for Dechaunac).

CONCLUSIONS

Here, we developed the models for predicting leaf area for the medicinal plants namely *Calamintha nepeta, Datura stromonium, Melissa officinalis, Mentha piperita, Nerium oleander, Origanum onites* and *Urtica dioica* which are economically important in Turkey and over the world. As the understanding of plant growth and development has been increasing, such mathematical models as this



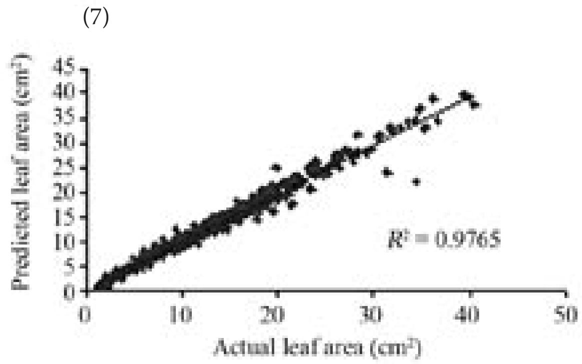


Fig. 2. Relationship between actual leaf area (cm²) and predicted leaf area (cm²) for *Calamintha nepeta* (1), *Datura stromonium* (2), *Melissa officinalis* (3), *Mentha piperita* (4), *Nerium oleander* (5), *Origanum onites* (6) and *Urtica dioica* (7)

shown in Table 1 will be very useful tools for prediction of leaf area for many plants without using of expensive devices. Because maximum leaf width and length are dimensions that can be easily measured in the field, use of these equations would enable researchers to make non-destructive measurements or repeated measurements on the same leaves. Such equations would also allow researchers to estimate leaf area in relation to factors like crop load, drought stress, and insect damage. Therefore, the models produced in the present study can be used safely by medicinal plant researchers for the species used in this research. On the other hand, different models can be developed by researches studying medicinal plants different from those used in the present study.

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Vztah plochy listů a rozměrů vybraných léčivých rostlin

ABSTRAKT: V rámci výzkumu byly vyvinuty predikční modely některých rostlin, jejichž listy se používají k léčivým účelům. Jsou to *Calamintha nepeta*, *Datura stromonium*, *Melissa officinalis*, *Mentha piperita*, *Nerium oleander*, *Origanum onites* a *Urtica dioica*, rostoucí divoce v oblasti černomořského pobřeží Turecka. Pro vypracování modelů byly měřeny šířka, délka a plocha listů a lístků nedestruktivní metodou. Plocha listů rostlin byla měřena digitálním planimetrem PLACOM, pro jednotlivé rostliny byla provedena několikanásobná regresní analýza pomocí programu Excel 7.0. Predikční modely plochy listů získané v rámci výzkumu jsou vyjádřeny vzorcem $LA = (a) + (b_1 \times L) + [b_2 \times (L \times W)] + (b_3 \times L^2) + (b_4 \times W^2) + [b_5 \times (L \times W^2)] + [b_6 \times (L^2 \times W)] + b_7 \times (L^2 \times W^2)]$, kde LA je plocha listu, W je šířka listu, L je délka listu a a, b_1, b_2, b_3, b_4, b_5, b_6 a b_7 jsou koeficienty. Hodnoty R^2 pro zkoumané léčivé rostliny se v závislosti na druhu pohybovaly od 0,82 u *Origanum onites* do 0,98 u *Urtica dioica*. Všechny hodnoty R^2 a standardní chyby byly shledány jako významné při úrovni $P < 0{,}001$.

Klíčová slova: modelování; plocha listu; léčivé rostliny

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