Leaf allometry and prediction of specific leaf area (SLA) in a sugar beet (*Beta vulgaris* L.) cultivar

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Abstract

Sugar beet cv. Rizor was grown for five growing seasons (2002–2006) in field conditions in Thessaly, central Greece. A total of 55 samplings took place during the growing seasons and allometric growth of the leaves was monitored. Highly significant ($p<0.001$) quadratic relationships were found between individual leaf mass (LM), individual leaf area (LA), aboveground dry biomass (ADB), and leaf area index (LAI). Only the LM-LA relationship ($LA = 43.444\ LM^2 - 10.693\ LM + 118.34$) showed a relatively high r^2 (0.63) and thus could be used for prediction of LA. Specific leaf area (SLA) was significantly related with leaf water content (LWC) ($SLA = 26\,279\ LWC^2 - 44\,498\ LWC + 18\,951$, $r^2 = 0.91$, $p<0.001$) and thus LWC could be a good indirect predictor of SLA in this cultivar.

Additional key words: leaf area; leaf mass; non-destructive methods; specific leaf mass.

Introduction

Field physiological studies demand reliable, rapid, and easily applied determinations. Destructive methods should be substituted by non-destructive ones. Physiological parameters such as leaf area index (LAI, the ratio of LA in m^2 per m^2 ground) and percentage of crop cover are now easily determined (Bouman 1992, Bouman *et al.* 1992, Röver and Koch 1995, van Henten and Bontsema 1995) and used in field research and crop growth models (Yin *et al.* 2000, Launay and Guérif 2003) since they are related with yield formation. However, investment in technologically perfect equipment usually demands a high initial cost, which could be restrictive for the widespread application of instrumental determinations.

The classical indirect methods for leaf area (LA) estimation using leaf dimension measurements (leaf length and width) have been reviewed by Květ and Marshall (1971); this is why we cite only methods not included in this paper. The possible equations have been developed for beans (Bhatt and Chanda 2003), faba bean (Peksen 2007), flax (Kurt *et al*. 2005), groundnut (Ma *et al.* 1992, Kathirvelan and Kalaiselvan 2007), maize (Stewart and Dwyer 1999), potato (Zrůst *et al.* 1974), pearl millet (Payne *et al.* 1991), rice (Bhan and Pande 1966, Palaniswamy and Gomez 1974), sorghum (Shih *et al.* 1981), sugar beet (Tsialtas and Maslaris 2005, 2008), safflower (Çamaş *et al.* 2005), soybean (Wiersma and Bailey 1975), sunflower (Schneiter 1978, Bange *et al.* 2000, Rouphael *et al.* 2007), taro (Lu *et al.* 2004), and white clover (Gamper 2005).

Although using simple equations is an easy, indirect way to estimate LA of individual leaves, this is not applicable to large numbers of leaves, *i.e.* when LAI should be determined. LAI is related with photon energy interception but its instrumental determination demands a high cost investment. For this reason, total leaf area (TLA) prediction models based on individual leaf or total aboveground biomass have been developed for many crops, *e.g.* alfalfa (Sharratt and Baker 1986), barley (Ramos *et al.* 1983), cotton (Rhoads and Bloodworth 1964, Akram-Ghaderi and Soltani 2007), groundnut (Ma *et al.* 1992), pearl millet (Payne *et al.* 1991), potato (Zrůst *et al.* 1974), sweet sorghum (Shih *et al.* 1981), and winter

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Abbreviations: ADB – aboveground dry biomass; LA – individual leaf area; LAI – leaf area index; LDM – leaf dry mass; LM – individual leaf dry mass; LWC – leaf water content; RGR – relative growth rate; SE – standard error; SLA – specific leaf area; TLA – total leaf area; TLM – total leaf mass.
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wheat (Aase 1978). TLA is related with leaf dry mass (LDM) *via* specific leaf area (SLA, the LA per LDM) by a function like TLA = LDM × SLA.

SLA is of high significance for plant physiological research since it is related with leaf morphology, leaf life span, and relative growth rate (RGR). It is considered as the best predictor of resource use strategy (Castro-Díez *et al.* 2000, Wright and Westoby 2001, Vendramini *et al.* 2002).

The aim of this work was to explore any significant and meaningful relationship between LA, individual LM, aboveground dry biomass (ADB), LAI, SLA, and leaf water content (LWC) in sugar beet cv. Rizor grown for five years in field conditions and sampled 9 to 12 times during the growing season.

Materials and methods

A long-term experiment, studying physiology and yield formation in sugar beet under the semi-arid conditions of central Greece, was established at Nikaia (39º33'N, 22º27'E, 98 m a.s.l.), Thessaly Plain. From 2002 to 2006, seeds of sugar beet cv. Rizor (*SESVANDERHAVE* NV/SA, Tienen, Belgium) were mechanically drilled (between 18 and 24 March) in eight rows (8-m long) per plot, at 50 cm apart, and at 15 cm spacing in the row. Seasonal changes of mean monthly temperature and rainfall are given in Table 1. The soil was inorganic, clayey (clay content > 45 %) with pH≈8.0. Adequate inorganic fertilization was applied as both basal (110 kg N ha^{-1}, 90 kg P ha^{-1}, and 265 kg K ha^{-1}) and top-dressing (40 kg N ha^{-1}). When necessary, protection was taken against cercospora leaf spot, powdery mildew, weeds, and insects by chemical sprayings. Supplemental irrigation was provided according to the needs of the crop and the availability of irrigation water.

Table 1. Mean monthly temperature [ºC] and monthly rainfall [mm] during the growing season (March to October) for the five years of experimentation (2002–2006). Means ± SE.

Month	Temperature	Rainfall [mm]
March	9.50±0.67	38.50±7.33
April	13.20±0.41	34.80±9.35
May	19.00±0.61	25.90±11.20
June	24.20±0.40	29.10±17.01
July	26.30±0.35	27.00±9.21
August	25.80±0.33	10.00±3.82
September	20.70±0.37	61.40±18.89
October	16.20±0.39	71.00±11.72
Mean or total	19.40±2.20	297.70±7.00

Each growing season, successive samplings took place from early June to early or mid-November in a Randomized Complete Block design experiment with six replications. On each sampling occasion, three rows, 7 m long (10.5 m^2), were harvested by hand and fresh root and aboveground biomass were weighed. In each plot, two healthy, full developed and typical of the plot sugar beets selected and topped. Tops were oven dried at 75 ºC till constant mass and water content of the aboveground biomass was calculated by comparing fresh and dry masses. Also, three upper, healthy, and fully-expanded sunlit leaves were collected from each plot, sealed in a plastic bag, and put in a portable refrigerator. The samples were transferred to the Physiology Laboratory of Larissa factory, *Hellenic Sugar Industry SA*, for determinations. LA was measured using the *WinDias* image analysis system (*Delta-T Devices*, Cambridge, UK) and after drying at 75 °C for 48 h, specific leaf area (SLA) was estimated as the ratio of LA to LM. Leaf water content (LWC) calculated by comparing fresh and dry masses. On each sampling date, 18 leaves were measured. LAI was determined non-destructively using the *SunScan* canopy analysis system (*Delta-T Devices*, Cambridge, UK) between the 3rd and 4th rows in each plot. Due to the high repeatability, two measurements were taken in each plot and the mean was estimated. In case of deviated measurements, a third LAI determination was conducted. In 2002, leaf trait determinations began in mid-July due to technical problems. A total of 55 samplings took place during the five growing seasons (2002–2006).

Figures were displayed using the *Excel 98 software (MS Office, Microsoft)* and the significance of correlations was determined by *SPSS 14* (*SPSS*).

Results and discussion

In this five-year experimentation, a large data set of physiological parameters (LA, LM, LWC, SLA, ADB, LAI) determined by both destructive and non-destructive methods was gathered. We aimed to explore any significant and meaningful relationship between them and to examine the putative indirect estimation of difficultly determined parameters by the easiest ones. The use of indirect methods is based on the assumption that mass and size of different plant parts are allometric.

Recently, Tsialtas and Maslaris (2005, 2008) proposed simple models for the non-destructive LA estimation in cv. Rizor and in sugar beet cultivars with different leaf morphology. In sugar beet cv. Rizor, non-destructive LAI estimation based on LA measurements is feasible (Tsialtas and Maslaris 2007). This work confirmed a previous finding since a highly significant, quadratic function between LAsand LAI was evident (LAI = $0.00002\ LA^2 + 0.0157\ LA - 0.1141$, $r^2 = 0.74$, $p<0.001$, $n = 55$).

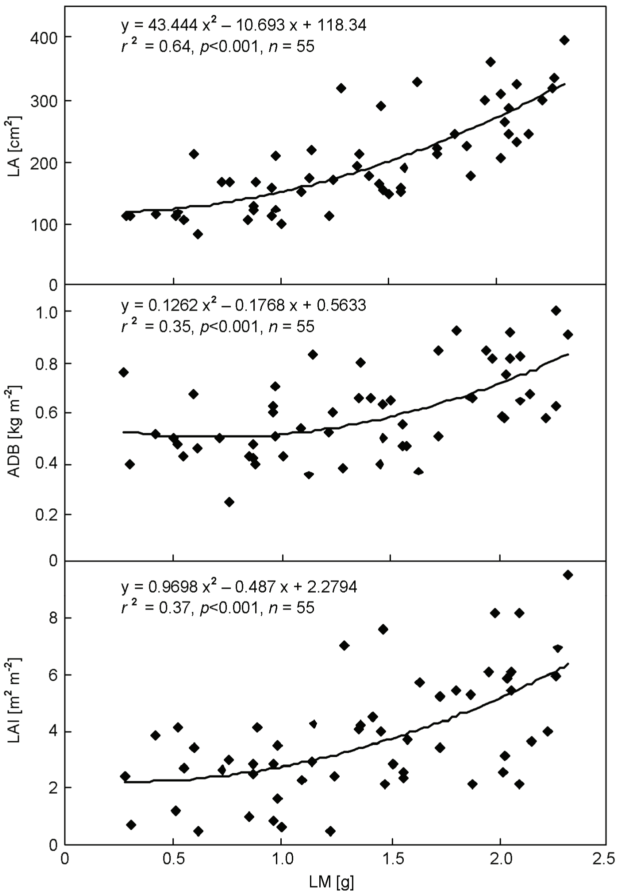

Fig. 1. Best-fitted curves of the relationships between LM and LA, ADB, or LAI.

LM was significantly related with LA, ADB, and LAI (Fig. 1). Although the three quadratic functions were significant, only the LM-LA relationship had a relatively high r^2. The other two relationships showed low r^2 ($\leq$0.37) and wide data scattering and thus could not be used for reliable LAI or ADB predictions (Fig. 1). High, linear predictability of LA by LM was previously reported for pearl millet (Payne *et al.* 1991) for which LA was also related with TLA. On the individual plant or area level, a close relationship between TLA and TLM was found for many species such as alfalfa (Sharratt and Baker 1986), barley (Ramos *et al.* 1983), cotton (Akram-Ghaderi and Soltani 2007), grasses (Retta *et al.* 2000), pearl millet (Payne *et al.* 1991), and winter wheat (Aase 1978). Close relationships between TLA and ADB were also reported (Sharratt and Baker 1986, Akram-Ghaderi and Soltani 2007) but in some cases (cereals) they showed a high predictability till a specific growth stage (Aase 1978, Ramos *et al.* 1983) after which biotic or abiotic effects on leaf growth and development biased the relationship. In sugar beet, aboveground biomass consists of leaves and petioles; mass partitioning between these two components during the growing season probably affects negatively the LM-ADB and LM-LAI relationships resulting in low r^2. Partitioning of photo-assimilates between aboveground constituents is possibly responsible for the high scattering of data in the LA-ADB and ADB-LAI relationships (Fig. 2). Both relationships were highly significant (p<0.001) but with low r^2($\leq$0.35) and practically with no use for indirect estimation of allometric components of sugar beet growth in the field.

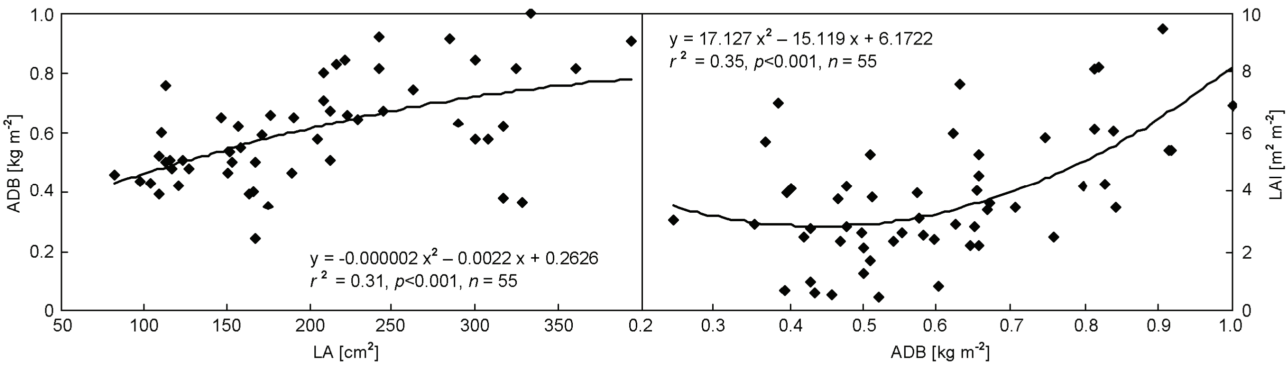

Fig. 2. Quadratic functions of the best-fitted LA-ADB and ADB-LAI relationships.

SLA is related with leaf morphology, water economy, CO_2 assimilation rate, and leaf life span (Virgona *et al.* 1990, Castro-Díez *et al.* 2000, Wright and Westoby 2001). In sugar beet, SLA depended on water availability, but no growth stage effect on SLA was found by Rinaldi (2003). For other plant species, there were reports that age or abiotic factors (irradiance, temperature) affect SLA (Barnes *et al.* 1969, Reddy *et al.* 1989, Lee and Heuvelink 2003, Akram-Ghaderi and Soltani 2007). These findings are consistent with our results (unpublished data). In our work, SLA was significantly (p<0.001) related with LM and LWC with quadratic functions showing moderate (0.64) or high (0.91) r^2, respectively (Fig. 3). Previous works predicted SLA based on abiotic factor measurements such as temperature or irradiance (Lee and Heuvelink 2003). We propose SLA estimation in cv. Rizor by measuring LWC and adapting results in a quadratic function (SLA = 26 279 LWC^2 – 44 498 LWC + 18 951, r^2 = 0.91, p<0.001, n = 55). We estimated LWC by drying leaf

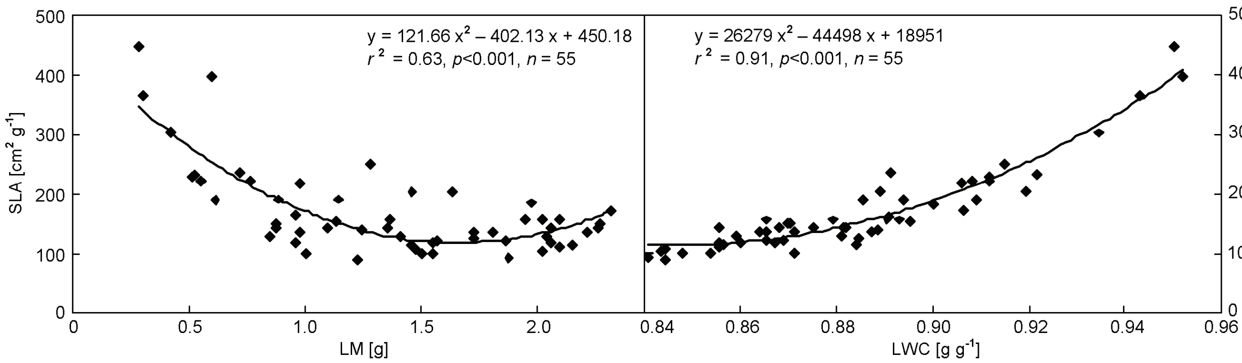

Fig. 3. Best-fitted curves of the LM-SLA and LWC-SLA relationships.

samples according to the conventional way which is time demanding. However, rapid and reliable estimations of LWC could be obtained by drying leaves in a microwave oven (Diprose 2001) and thus the disadvantage of time demand for LWC estimation is easily overcome.

References

Aase, J.K.: Relationship between leaf area and dry matter in winter wheat. – Agron. J. **70**: 563-565, 1978.

Akram-Ghaderi, F., Soltani, A.: Leaf area relationships to plant vegetative characteristics in cotton (*Gossypium hirsutum* L.) grown in a temperate sub-humid environment. – Int. J. Plant Prod. **1**: 63-71, 2007.

Bange, M.P., Hammer, G.L., Milroy, S.P., Rickert, K.G.: Improving estimates of individual leaf area of sunflower. – Agron. J. **92**: 761-765, 2000.

Barnes, D.K., Pearce, R.B., Carlson, G.E., Hart, R.H., Hanson, C.H.: Specific leaf weight differences in alfalfa associated with variety and plant age. – Crop Sci. **9**: 421- 423, 1969.

Bhan, V.M., Pande, H.K.: Measurement of leaf area of rice. – Agron. J. **58**: 454, 1966.

Bhatt, M., Chanda, S.V.: Prediction of leaf area in *Phaseolus vulgaris* by non-destructive method. – Bulg. J. Plant Physiol. **29**: 96-100, 2003.

Bouman, B.A.M.: Accuracy of estimating the leaf area index from vegetation indices derived from crop reflectance characteristics, a simulation study. – Int. J. remote Sens. **13**: 3069-3084, 1992.

Bouman, B.A.M., van Kasteren, H.W.J., Uenk, D.: Standard relations to estimate ground cover and *LAI* of agricultural crops from reflectance measurements. – Eur. J. Agron. **1**: 249-262, 1992.

Çamaş, N., Ayan, A.K., Esendal, E.: Leaf area prediction model for safflower (*Carthamus tinctorius* L.). – Pakistan J. biol. Sci. **8**: 1541-1543, 2005.

Castro-Díez, P., Puyravaud, J.P., Cornelissen, J.H.C.: Leaf structure and anatomy as related to leaf mass per area variation in seedlings of a wide range of woody plant species and types. – Oecologia **124**: 476-486, 2000.

Diprose, M.F.: Some considerations when using a microwave oven as a laboratory research tool. – Plant Soil **229**: 271-280, 2001.

Gamper, H.: Nondestructive estimates of leaf area in white clover using predictive formulae: The contribution of genotype identity to trifoliate leaf area. – Crop Sci. **45**: 2552-2556, 2005.

Hoffmann, C.M., Blomberg, M.: Estimation of Leaf Area Index of *Beta vulgaris* L. based on optical remote sensing data. – J. Agron. Crop Sci. **190**: 197-204, 2004.

Kathirvelan, P., Kalaiselvan, P.: Groundnut (*Arachis hypogea* L.) leaf area estimation using allometric model. – Res. J. agr. biol. Sci. **3**: 59-61, 2007.

Kurt, O., Uysal, H., Uzun, S.: Non-destructive area estimation of flax (*Linum usitatissimum* L.). – Pakistan J. Bot. **37**: 837-841, 2005.

Květ, J., Marshall, J.K.: Assessment of leaf area and other assimilating plant surfaces. – In: Šesták, Z., Čatský, J., Jarvis, P.G. (ed.): Plant Photosynthetic Production. Manual of Methods. Pp. 517-555. Dr W. Junk N.V. Publ., The Hague 1971.

Launay, M., Guérif, M.: Ability for a model to predict crop production variability at the regional scale: an evaluation for sugar beet. – Agronomie **23**: 135-146, 2003.

Lee, J.H., Heuvelink, E.: Simulation of leaf area development based on dry matter partitioning and specific leaf area for cut chrysanthemum. – Ann. Bot. **91**: 319-327, 2003.

Lu, H.-Y., Lu, C.-T., Wei, M.-L., Chan, L.-F.: Comparison of different models for nondestructive leaf area estimation in taro. – Agron. J. **96**: 448-453, 2004.

Ma, L., Gardner, F.P., Selamat, A.: Estimation of leaf area from leaf and total mass measurements in peanut. – Crop Sci. **32**: 467-471, 1992.

Palaniswamy, K.M., Gomez, K.A.: Length-width method for estimating leaf area of rice. – Agron. J. **66**: 430-433, 1974.

Payne, W.A., Wendt, C.W., Hossner, L.R., Gates, C.E.: Estimating pearl millet leaf area and specific leaf area. – Agron. J. **83**: 937-941, 1991.

Peksen, E.: Non-destructive leaf area estimation model for faba bean (*Vicia faba* L.). – Sci. Hort. **113**: 322-328, 2007.

Ramos, J.M., del Moral, L.F.G., Recalde, L.: Dry matter and leaf area relationships in winter barley. – Agron. J. **75**: 308-310, 1983.

Reddy, V.R., Acock, B., Baker, D.N., Acock, M.: Seasonal leaf area-leaf weight relationships in the cotton canopy. – Agron. J. **81**: 1-4, 1989.

Retta, A., Armbrust, D.V., Hagen, L.J., Skidmore, E.L.: Leaf and stem area relationships to masses and their height distributions in native grasses. – Agron. J. **92**: 225-230, 2000.

Rhoads, F.M., Bloodworth, M.E.: Area measurement of cotton leaves by a dry-weight method. – Agron. J. **56**: 520-522, 1964.

Rinaldi, M.: Variation of specific leaf area for sugar beet depending on sowing date and irrigation. – Ital. J. Agron. **7**: 23-32, 2003.

Rouphael, Y., Colla, G., Fanasca, S., Karam, F.: Leaf area estimation of sunflower leaves from simple linear measurements. – Photosynthetica **45**: 306-308, 2007.

Röver, A., Koch, H.-J.: Indirect determination of leaf area index of sugar beet canopies in comparison to direct measurement. – J. Agron. Crop Sci. **174**: 189-195, 1995.

Schneiter, A.A.: Non-destructive leaf area estimation in sunflower. – Agron. J. **70**: 141-142, 1978.

Sharratt, B.S., Baker, D.G.: Alfalfa leaf area as a function of dry matter. – Crop Sci. **26**: 1040-1043, 1986.

Shih, S.F., Gascho, G.J., Rahi, G.S.: Modelling biomass production of sweet sorghum. – Agron. J. **73**: 1027-1032, 1981.

Stewart, D.W., Dwyer, L.M.: Mathematical characterization of leaf shape and area of maize hybrids. – Crop Sci. **39**: 422-427, 1999.

Tsialtas, J.T., Maslaris, N.: Leaf area estimation in a sugar beet cultivar by linear models. – Photosynthetica **43**: 477-479, 2005.

Tsialtas, J.T., Maslaris, N.: Leaf shape and its relationship with Leaf Area Index in a sugar beet (*Beta vulgaris* L.) cultivar. – Photosynthetica **45**: 527-532, 2007.

Tsialtas, J.T., Maslaris, N.: Leaf area prediction model for sugar beet (*Beta vulgaris* L.) cultivars. – Photosynthetica **46**: 291-293, 2008.

van Henten, E.J., Bontsema, J.: Non-destructive crop measurements by image processing for crop growth control. – J. agr. eng. Res. **61**: 97-105, 1995.

Vendramini, F., Díaz, S., Gurvich, D.E., Wilson, P.J., Thompson, K., Hodgson, J.G.: Leaf traits as indicators of resource-use strategy in floras with succulent species. – New Phytol. **154**: 147-157, 2002.

Virgona, J.M., Hubick, K.T., Rawson, H.M., Farquhar, G.D., Downes, R.W.: Genotypic variation in transpiration efficiency, carbon isotope discrimination and carbon allocation during early growth in sunflower. – Aust. J. Plant Physiol. **17**: 207-214, 1990.

Wiersma, J.V., Bailey, T.B.: Estimation of leaflet, trifoliate, and total leaf areas of soybeans. – Agron. J. **67**: 26-30, 1975.

Wright, I.J., Westoby, M.: Understanding seedling growth relationships through specific leaf area and leaf nitrogen concentration: generalizations across growth forms and growth irradiance. – Oecologia **127**: 21-29, 2001.

Yin, X., Schapendonk, A.H.C.M., Kropff, M.J., van Oijen, M., Bindraban, P.S.: A generic equation for nitrogen-limited Leaf Area Index and its application in crop growth models for predicting leaf senescence. – Ann. Bot. **85**: 579-585, 2000.

Zrůst, J., Partyková, E., Nečas, J.: Relationships of leaf area to leaf weight and length in potato plants. – Photosynthetica **8**: 118-124, 1974.