

An Estimation of Individual Leaf Area in Cabbage and Broccoli Using Non-destructive Methods

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ABSTRACT

Accurate and nondestructive methods to determine individual leaf areas of plants are useful tools in physiological and agronomic research. A determination of the individual leaf area (LA) of such species in Brassicaceae family as red cabbage (*Brassica oleracea* var. *cappitata* L. f. Rabra), cabbage (*Brassica oleracea* var. *cappitata* L. F. Alba) and broccoli (*Brassica oleracea* var. *Italica* L.) involves measurements of leaf parameters including leaf length (L) and width (W), or some combination of these parameters. This research was carried out during 2008 (on eight cabbage, six broccoli, and three red cabbage genotypes) under open field conditions, to see whether an equation could be developed to estimate leaf area of cabbage and broccoli across their genotypes. Regression analysis of LA vs. L and W revealed several equations that could be used for estimating the area of individual cabbage and broccoli leaves. A linear equation having W as the independent variable provided the most accurate estimate of red cabbage as well as ordinary cabbage LA. The linear equation ($LA = a + bW^2$) exhibited a high accuracy and precision in estimating red cabbage and non-red cabbage LA. For broccoli a linear equation having LW as the independent variable provided the most accurate estimate of LA, but required twice the time needed for leaf area measurement.

Keywords: Brassicaceae, Correlation, Leaf length, Leaf width, Planimeter, Regression.

INTRODUCTION

Plant leaf area is an important determinant of light interception and consequently of transpiration, photosynthesis as well as plant productivity (Goudriaan and Van Laar, 1994; Demirsoy *et al.*, 2007). Plant physiologists and agronomists have demonstrated the importance of this parameter in estimating crop growth, development rate, yield potential, radiation use efficiency, and water as well as nutrient take up (Bhatt and Chanda, 2003; Olivera and Santos, 1995; Williams, 1987; Williams and Martinson, 2003).

Leaf area can be measured by destructive or nondestructive measurement methods. Many methods have been so far devised to facilitate the measurement of leaf area.

However, these methods, including those of tracing, blueprinting, photographing, or using a conventional planimeter, require the excision of leaves from the plants. It is therefore not possible to make successive measurements of the same leaf. Plant canopy is also damaged, which might create problems to other measurements or experiments. Leaf area can be measured quickly, accurately, and nondestructively using a portable scanning planimeter (Daughtry, 1990; Demirsoy, 2009), but it is suitable only for small plants with few leaves (Nyakwende *et al.*, 1997).

An alternative method to measure leaf area is using image analysis with image measurement and analysis software. The capture of image by digital camera is rapid, and the analysis using proper software is accurate (Bignami and Rossini, 1996), but

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the processing is time consuming, and the facilities are generally expensive. Therefore, an inexpensive, rapid, reliable, and nondestructive method for measuring leaf area is sought by the agronomists.

If the mathematical relationships between leaf area and one or more dimensions of the leaf (length and width) could be clarified, a method using just linear measurements to estimate leaf area would be more advantageous than many of the methods mentioned above (Beerling and Fry, 1990; Villegas *et al.*, 1981). Various combinations of measurements and various equations relating length and width to area have been developed for several such horticultural crops as cucumber (Cho *et al.*, 2007; Robbins and Pharr, 1987), pepper (De Swart *et al.*, 2004), grape (Montero *et al.*, 2000; Williams and Martinson, 2003), kiwifruit (Mendoza de Gyves *et al.*, 2007), rabbiteye blueberry (NeSmith, 1991), muskmelon (Panta and NeSmith, 1995), faba bean (Peksen, 2007), zucchini squash (Rouphael *et al.*, 2006), tomato (Schwarz and Kläring, 2001), chestnut (Serdar and Demirsoy, 2006), peach (Demirsoy *et al.*, 2004), cherry (Demirsoy and Demirsoy, 2003 a; b), strawberry (Demirsoy *et al.*, 2005) while information on an estimation of leaf area in cabbages and broccoli is still lacking.

Therefore, the aim of this study was to develop an equation for leaf area prediction from linear measurements of leaf length and width in cabbages and broccoli.

MATERIALS AND METHODS

The experiment was conducted from 3 April 2008 to 9 July 2008 on farmland belonging to Guilan University, Rasht, Iran (37°16'N, 51°3'E). The cabbages (red and non-red cabbage) as well as broccoli hybrids used in this research are listed in Table 1. The soil was a sandy loam, pH 6.8, containing total N (2.8%), total C (1.2%), a C/N ratio of 2.33 along with 15, 65, 110 mg kg⁻¹ of Ca, P, and K, respectively, and finally an EC of 0.08 ds cm⁻¹. The land was prepared through plowing and disking and then formed into beds prior to transplanting of the experimental plants. Beds were 1.2 m wide on 1.5 m centers. Granular fertilizer, 120N-50P-200K kg ha⁻¹, according to results based on soil tests was applied by broadcast to beds and worked into the soil before transplanting. Transplants were prepared in cold frame and hardened by reducing irrigation and temperature before being transplanted. Transplanting took place at 50×60 cm distance between plants and

Table 1. Genotypes, cultivars, and companies from which the plants had been taken.

| Genotype | Cultivar | Company |
|-------------|-------------------------|------------|
| Red cabbage | Hyb. Red Rookie | Sakata |
| Red cabbage | Hyb Red Dynasty | Asgrow |
| Red cabbage | Hyb Schwarzkopf 2 | Kiepenkerl |
| Cabbage | Hyb. Royal Vantage | Sakata |
| Cabbage | Hyb. Nozomi | Sakata |
| Cabbage | Hyb. Blue Dynasty | Asgrow |
| Cabbage | Hyb. Zennith | Asgrow |
| Cabbage | Hyb. Platinum Dynasty | Asgrow |
| Cabbage | Hyb. Green Challenger | Asgrow |
| Cabbage | Hyb. Green Voyager | Asgrow |
| Cabbage | Hyb. Brunswijker | Kiepenkerl |
| Broccoli | Hyb. Packman | Petoseed |
| Broccoli | Hyb. Captain | Petoseed |
| Broccoli | Hyb. Liberty | Petoseed |
| Broccoli | Hyb. Heritage | Petoseed |
| Broccoli | Hyb. Tradition | Petoseed |
| Broccoli | Hyb. Calabrese natalino | Kiepenkerl |

Table 2. Correlation between red cabbage, cabbage, broccoli and total samples' LA and their measured linear parameters.

| | <i>L</i> (cm) | <i>W</i> (cm) | <i>L</i> ² (cm ²) | <i>W</i> ² (cm ²) | <i>LW</i> (cm ²) |
|---------------------------------------|------------------|------------------|---|---|---------------------------------|
| Red cabbage LA (cm ²) | 0.60** | 0.96*** | 0.61** | 0.96*** | 0.87*** |
| Cabbage LA (cm ²) | 0.40* | 0.53** | 0.30* | 0.48** | 0.47** |
| Broccoli LA (cm ²) | 0.89*** | 0.92*** | 0.89*** | 0.93*** | 0.97*** |
| Total plants LA (cm ²) | 0.94*** | 0.97*** | 0.94*** | 0.98*** | 0.97*** |

*, ** and ***: Significant at $P \leq 0.05$, $P \leq 0.01$ and $P \leq 0.001$ respectively.

within plant rows respectively. Irrigation was applied as necessary.

For leaf area estimation 18 leaves which varied in size from large to small (in each cultivar) were randomly selected from different levels of the canopy in 30 plants, 30 days following their simultaneous transplanting. Leaves of cabbage and broccoli cultivars were measured for their area (LA), length (*L*) and width (*W*). Immediately after cutting, leaves were placed in plastic bags and transported to the laboratory. Leaf *L* was measured from lamina tip to the point of intersection of the lamina and the petiole, along the midrib of the lamina, while leaf *W* was measured from end-to-end between the widest lobes of the lamina perpendicular to the lamina mid-rib. Values of *L* and *W* were recorded to the nearest 0.01 mm. The area of each leaf (LA) was measured using a planimeter (A. OTT Kempten, Germany, Bayern). The dependent variable (LA) was regressed with different independent variables, including *L*, *W*, *L*², *W*², and the product *L*×*W*. The values of the coefficients (*b*), constants (*a*), *R* square (*R*²) and Root of Mean Square Error (RMSE) were also recorded (Table 3).

RESULTS AND DISCUSSION

Correlation analysis demonstrated strong relationships ($P < 0.001$) between leaf area

(LA) as the dependent variable and maximum leaf width (*W*), the square of width (*W*²) and product of length times width (*LW*) for red cabbage, broccoli and the rest of total measured leaves (Table 2). For cabbage, a strong correlation ($P < 0.01$) was obtained between leaf area (LA) and maximum leaf width (*W*), the square of width (*W*²) and product of length times width (*LW*) (Table 2). This is in agreement with previous studies (Mendoza de Gyves *et al.*, 2007; Peksen, 2007; Rivera *et al.*, 2007; Rouphael *et al.*, 2006; 2007) on nondestructive equation development for predicting leaf area using simple linear measurements. This study demonstrated that equations with a mere measurement of *W* were more acceptable for estimating leaf area for red cabbages, cabbage and broccoli. Equation $LA = a + b LW$ that required two measurements per leaf could estimate leaf areas of cabbages and broccoli, accurately, but took double the time taken for leaf measurements. The linear equation having *W* and *W*² as independent variables exhibited a high accuracy and precision in estimating individual leaf areas. This equation was preferred because of its simplicity and convenience, as it only involves one variable. As stated by Robbins and Pharr (1987), equation selection requires a balance between predictive qualities of the equation and the economy of including the least

**Table 3.** Regression equations between leaf area (cm²) and L (cm), W (cm), L² (cm²), W² (cm²) and LW(cm²) in red cabbage, cabbage and broccoli, as well as total leaves.

| Genotype | Regression equation | R ² | RMSE |
|-------------|----------------------------------|----------------|--------|
| Red cabbage | LA= -5.8927+1522 L | 0.3652 | 95.43 |
| Red cabbage | LA= -338.86+3.28 W | 0.915 | 34.91 |
| Red cabbage | LA= 208.42+0.0026 L ² | 0.375 | 94.683 |
| Red cabbage | LA= 54.078+0.0067 W ² | 0.916 | 34.721 |
| Red cabbage | LA= 74.026+0.0053 LW | 0.7526 | 59.575 |
| Cabbage | LA= 2.709+0.6956 L | 0.1602 | 37.564 |
| Cabbage | LA= 5.5981+0.8961 W | 0.2771 | 34.85 |
| Cabbage | LA= 36.997+0.0034 L ² | 0.1492 | 37.808 |
| Cabbage | LA= 43.43+0.0049 W ² | 0.2373 | 35.797 |
| Cabbage | LA= 34.514+0.0048 LW | 0.2240 | 34.85 |
| Broccoli | LA= -34.63+1.5861 L | 0.7981 | 5.706 |
| Broccoli | LA= -34.63+1.1698 W | 0.8441 | 5.0131 |
| Broccoli | LA= 15.419+0.003 L ² | 0.7944 | 5.7577 |
| Broccoli | LA= 3.2834+0.0088 W ² | 0.8691 | 4.5952 |
| Broccoli | LA= 5.9737+0.0058 LW | 0.9501 | 2.836 |
| Total | LA= -124.08+1.8971 L | 0.8866 | 59.29 |
| Total | LA= -104.28+2.2706 W | 0.9470 | 40.539 |
| Total | LA= 20.467+0.0047 L ² | 0.8746 | 62.345 |
| Total | LA= 21.72+0.0073 W ² | 0.9687 | 31.172 |
| Total | LA= 17.651+0.006 LW | 0.9492 | 39.706 |

number of variables necessary to predict leaf area.

It is resulted equations $LA= -338.88+3.2859 W$, $LA= 5.5981+0.8961 W$, and $LA= 3.2834 + 0.0088 W^2$ can be easily employed to estimate accurately and in large numbers, the leaf area of red cabbage, cabbage, broccoli plants respectively in many experimental comparisons without the need for any expensive instruments, such as a leaf area planimeter or digital camera equipped with image measurement software. Equation $LA= 21.72+0.0073 W^2$ could be properly employed to estimate leaf areas (in all leaves) that must be evaluated in research works.

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محاسبه سطح برگ کلم پیچ و کلم بروکلی با استفاده از روشهای اندازه‌گیریهای غیر تخریبی

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چکیده

محاسبه دقیق و غیر مخرب سطح برگ گیاهان ابزاری مفید در مطالعات فیزیولوژیکی و کشاورزی است. تعیین سطح برگ گیاهان خانواده کلمها مانند کلم پیچ قرمز، کلم پیچ سبز و کلم بروکلی شامل اندازه‌گیری پارامترهای طول، عرض، و یا برخی ترکیبات این پارامترها می‌باشد. این تحقیق در سال ۱۳۸۷ (روی ۸ ژنوتیپ کلم پیچ سبز، ۶ ژنوتیپ کلم بروکلی و ۳ ژنوتیپ کلم پیچ قرمز) در شرایط کشت مزرعه‌ای به منظور تعیین بهترین معادله رگرسیونی برای هر یک اجرا شده است. امکان محاسبه سطح برگ با شاخصهایی همانند طول برگ، عرض برگ و مربع آنها و همچنین حاصلضرب آنها اندازه‌گیری‌هایی بر ۶ رقم کلم پیچ و ۴ رقم بروکلی انجام شد. معادلات رگرسیونی منجر به ایجاد چندین معادله مفید برای برآورد سطح برگ کلم پیچ و بروکلی شد. معادله خطی که در آن عرض برگ متغیر وابسته بود منجر به بیشترین دقت در برآورد کلم پیچ قرمز و کلم پیچ سبز شد. معادله $LA = a + bW^2$ بیشترین دقت و درستی را در محاسبه سطح برگ کلم پیچ قرمز و کلم پیچ سبز داشت. برای بروکلی معادله‌ای که حاصلضرب طول در عرض را به عنوان متغیر وابسته داشت بیشترین دقت را در مورد سطح برگ فراهم نمود اما دو برابر زمان برای اندازه‌گیری سطح برگ مورد نیاز بود.