

Short communication:

Leaf chlorophyll content of ornamental plants: A choice of destructive or non-destructive measurement

(Kandungan klorofil dalam daun tanaman hiasan: Pilihan cara pengukuran yang memusnahkan ataupun tak memusnahkan)

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Key words: chlorophyll, non-destructive sampling, ornamental crops, SPAD-502

Abstract

Most leaf chlorophyll determination requires collection and chemical extraction of tissue samples. This destructive method may not be suitable for the time series experiments and studies involving gas exchanges where the *in situ* measurements of total leaf chlorophyll are needed. Thus, in this study, a portable chlorophyll meter was used to compare the accuracy and validity of chlorophyll determination techniques. Leaf tissues of seven ornamental species were analysed for both methods. Strong correlation values were found between the acetone-extractable method and the portable chlorophyll meter readings. Regression equations were plotted and in most cases the correlations were more than 0.70. These regression equations therefore can be used to give convenient, fast and accurate values to the users.

Abstrak

Kebanyakan pengukuran kandungan klorofil daun dijalankan dengan mengambil sampel tisu dan menentukan kandungannya secara kimia. Cara memusnahkan ini kurang sesuai bagi ujikaji yang melibatkan faktor masa dan kajian yang melibatkan pertukaran gas yang memerlukan jumlah kandungan klorofil diukur secara *in situ*. Oleh yang demikian, meter pengukur klorofil mudah alih digunakan dalam kajian ini untuk membandingkan kejituan dan kesahihan kedua-dua teknik penentuan klorofil. Tisu daun tujuh jenis tanaman hiasan telah dianalisis dengan kedua-dua cara. Terdapat nilai korelasi yang tinggi antara cara pengekstratan menggunakan aseton dengan penggunaan meter mudah alih. Kebanyakan nilai korelasi melebihi 0.70. Persamaan regresi boleh digunakan untuk memberi nilai yang mudah, cepat dan tepat kepada pengguna.

Introduction

Chlorophyll has been known to be one of the most vital pigments involved in the photosynthetic activity of most green plants (Heath 1969). It is critical not only in the

physiology but also in crop productivity and plant economy (Salisbury and Ross 1992). It is therefore not surprising that many plant physiologists have described a strong positive correlation of photosynthetic

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activities to the concentration of leaf chlorophyll (Schaper and Chacko 1991; Salisbury and Ross 1992). As a result, a direct relationship was also reported for the chlorophyll content and the increased nutrient uptake, especially nitrogen and magnesium (Piekielek and Fox 1992; Schepers et al. 1992; Reeves et al 1993), and the adaptation to mild environmental stresses, such as salinity and drought (Hall and Rao 1987; Salisbury and Ross 1992).

In many cases, destructive chlorophyll determinations are done in most plants. The use of chemical extraction either by acetone or ethanol has dominated the protocol. Such a method not only requires substantial planting materials but may also produce a misleading result. The result could be misinterpreted if the experiment involved a time sequence study such as monitoring the leaf chlorophyll content over a period of time. Therefore, the readings have to be taken on the same samples through out the experiment. In this case, non-destructive sampling will be needed. Moreover, chemical extraction may also cause pigment degradation because of exposure to light and explosion by ethanol. Alternatively, destructive sampling can be avoided by using SPAD-502, a portable meter that determines the relative amount of chlorophyll by measuring the transmittance of the leaf in two wavelength reactions, i.e. in the red and near-infrared regions. The meter is also handy when *in situ* quantification of total leaf chlorophyll is needed (Schaper and Chacko 1991). However, such tool only gives relative values. As a result, it has to be calibrated or translated to the amount of leaf chlorophyll content for every crop prior to its use (Schaper and Chacko, 1991). In this study, the chlorophyll contents of 11 ornamental plants comprising indoor and outdoor plants were determined using the destructive and non-destructive methods.

The regression equations and their *r* values obtained are highlighted for the use of future researchers.

Materials and methods

This study was carried out from May 1996 to December 1997. Determination of leaf chlorophyll content was done using both destructive and non-destructive methods. The readings for leaf chlorophyll measurement were taken from nine outdoor ornamental plants, namely *Cassia siamea*, *Cananga odorata*, *Saraca thaipingensis*, *Mimusop elengi*, *Pometia pinnata*, *Melia azederach*, *Gardenia carinata*, *Diospyrus discolor* and *Melaleuca leucadendron*. Two indoor ornamental plants, *Ficus benjamina* and *Ophiophogon jaburan*, were also included in this study. These plant species were planted under natural habitat in Serdang for the outdoor ornamental crops and as potted plants for the indoor ornamental plants.

Leaves for the non-destructive measurement were sampled using SPAD-502 meter (Minolta, Japan). Later, a minimum of nine 4 cm² leaf samples were taken to the laboratory and kept in a vial together with 80% (v/v) acetone. These samples were kept in the dark for 36 h at room temperature following the method as suggested by Coombs et al. (1986). After 36 h, the absorptions at 664 and 647 nm were measured using spectrophotometer (Model Shimazu, Japan). These wavelengths were chosen as they are close to the absorption peaks in 80% (v/v) acetone of chlorophyll *a* and *b* (Coombs et al. 1986). An average of 9–20 leaves of different age groups were sampled randomly.

The absorption values were later used for the calculation of total chlorophyll based on the equation :

$$\text{Total chlorophyll} = 7.93(A_{664}) + 19.53(A_{647})$$

A regression analysis between chlorophyll concentration and SPAD values, and its correlation coefficient for each species were then determined using *Excel* softwares. The relationships between the destructive measurement for each species (acetone extraction) and the non-destructive measurements (SPAD values) were plotted for four ornamental species. The plants

selected were *F. benjamina* and *O. jaburan* which represented indoor ornamental plants and *D. discolor* and *M. leucadendron* the outdoor ornamental plants.

Results and discussion

In this study, a strong correlation between the non-destructive, SPAD values and total

chlorophyll extracted using acetone was observed. For the indoor ornamental plants, *F. benjamina* showed a correlation coefficient of 0.97 and *O. jaburan* showed a coefficient of 0.87 (Figure 1). Similarly, *r* values of 0.96 and 0.87 were obtained for *D. discolor* and *M. leucadendron* respectively (Figure 2).

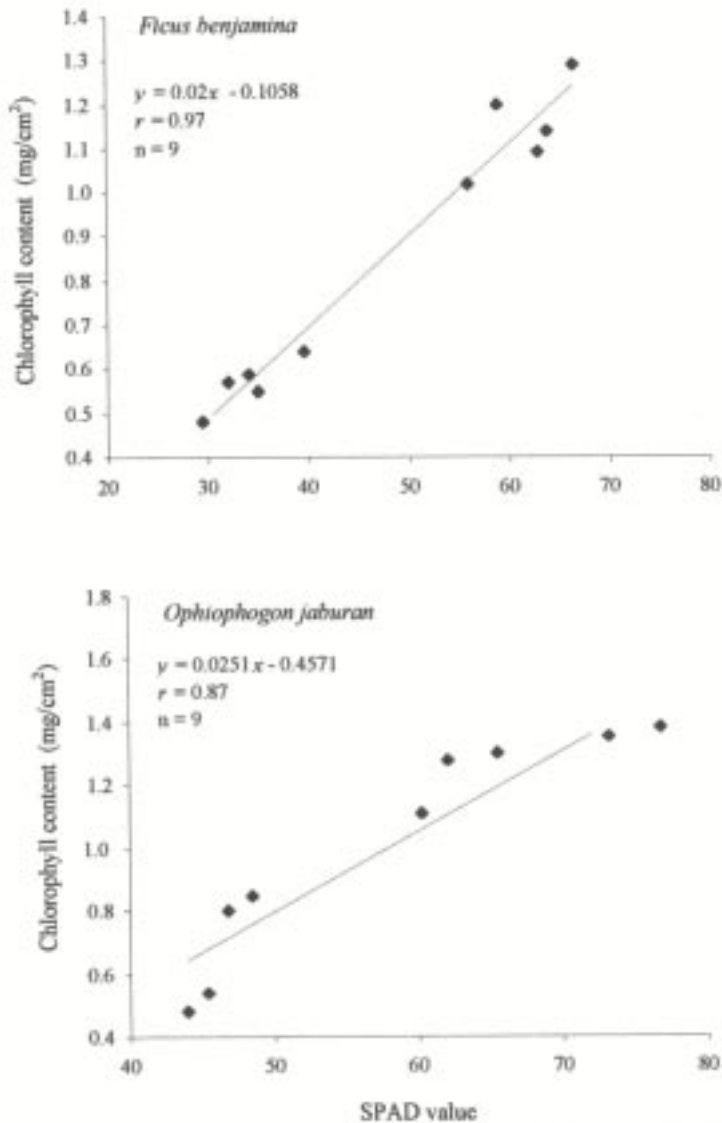


Figure 1. A comparison of the destructive sampling of chlorophyll extraction versus the non-destructive sampling for two indoor ornamental plants

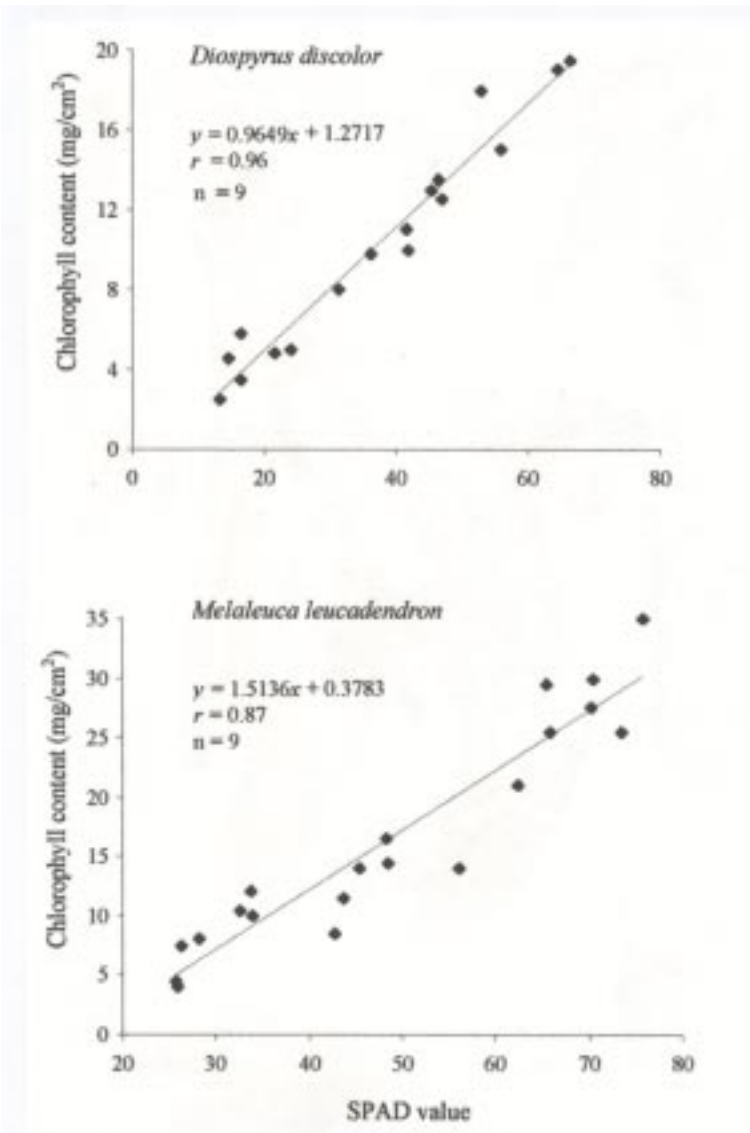


Figure 2. A comparison of the destructive sampling of chlorophyll extraction versus the non-destructive sampling for two outdoor ornamental plants

The regression equations and their correlation coefficient values for other ornamental plants in this study are presented in Table 1. In general, all species consistently showed a strong correlation between the destructive and the non-destructive methods. The values of the correlation coefficient ranged from 0.70 for *Mimusop elengi* to 0.87 for *Saraca thaipingensis*.

The study indicated that there is strong correlation between the values of chlorophyll contents obtained by the destructive and the non-destructive methods. The regression line shown in this study described a linear relationship for the two measurement techniques. The correlation coefficients for these methods were also shown. A similar trend was also observed for maize (Piekielek

Table 1. Relationship between chlorophyll content obtained by non-destructive and destructive sampling methods for seven ornamental species

Crop	Equation	<i>r</i>
<i>Cassia siamea</i>	$y = 0.91x + 3.53$	0.82
<i>Cananga odorata</i>	$y = 0.67x + 7.76$	0.81
<i>Saraca thaipingensis</i>	$y = 0.68x + 3.77$	0.87
<i>Mimusop elengi</i>	$y = 0.47x + 5.61$	0.70
<i>Pometia pinnata</i>	$y = 1.19x + 5.07$	0.82
<i>Melia azederach</i>	$y = 0.44x + 6.21$	0.80
<i>Gardenia carinata</i>	$y = 0.35x + 8.12$	0.75

Note: *y* is chlorophyll content (mg/cm²)
x is SPAD value

and Fox 1992), wheat (Reeves et al. 1993) and tree species (Schaper and Chacko 1991).

Overall, results shown that these equations could be used to calculate the total chlorophyll content as the minimum coefficient values was 0.70. It is hopeful that researchers interested in the development of tropical ornamental plants could use the study as the basis for chlorophyll content. Moreover, studies on chlorophyll content can also be done to describe certain leaf nutritional status such as nitrogen and magnesium as described by Schepers et al. (1992).

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