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Evaluation chlorophyll contents assessment on three indoor ornamental plants with plant growth regulators

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ABSTRACT

The Minolta SPAD 502 meter is a hand-held light meter used to measure the relative greenness of leaves in a rapid manner. (SPAD is an acronym for Soil Plant Analysis Development.) SPAD 502 determines the relative amount of chlorophyll present by measuring the transmittance of the leaf in two wave bands (600-700 and 400-500 nm). Portable, non-destructive chlorophyll meters could be a valuable tool for forest managers and researchers. The effect of gibberellic acid and benzyladenine on Ficus benjamina, Schefflera arboricola and Dizigotheeca elegantissima was evaluated at pot cultivation conditions. This study was performed in three factorial test based on completely randomized design and 4 repeation with 9 treatments. The objective of this study was to evaluate the ability of a estimate chlorophyll (CHL) of foliar application with gibberellic acid (GA_3) and benzyladenine (BA) at 0, 100 and 200 mg.L^{-1} levels 60, 120 and 180 day after spray on chlorophyll contents. The obtained results show, highest chlorophyll content of Ficus benjamina, Schefflera arboricola and Dizigotheeca elegantissima flower was achieved for 200 mg.L^{-1} GA_3 and 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} for 180 DAP which showed significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the growth regulator up to 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} BA significantly increased chlorophyll content compared to control treatment. Compared to other concentrations, control had the lowest chlorophyll content ($p<0.05$). Results of the investigation showed that application of plant growth regulators in higher concentrations had positive effects on leaf chlorophyll content of Ficus benjamina, Schefflera arboricola and Dizigotheeca elegantissima foliage plants.

Keywords: Benzyladenine, Chlorophyll Contents, *Dizigotheeca elegantissima*, *Ficus benjamina*, Gibberellic Acid, *Schefflera arboricola*.

INTRODUCTION

Rapid, non-destructive estimation of total chlorophyll and nitrogen content is a potentially important application for both forest managers and researchers. Significant correlations between total foliar extractable chlorophyll and chlorophyll content index values (CCI) obtained with portable chlorophyll meters have been reported for a number of agricultural species, including cabbage, cotton, and pea [14], sorghum and pigeonpea [34] muskmelon [1], corn [8], and several fruit tree species [22, 24]. As up to 75% of nitrogen in mesophyll cells can be located in the chloroplast [16], similar relationships have been observed between CCI and foliar or whole plant in agricultural species, including corn [3], cotton [33] and grasses [19]. Significant correlations have been reported for CCI and

total chlorophyll in paper birch (*Betula papyrifera Marsh*.) [18], red maple (*Acer rubrum* L.) [25] andapple (Malus domesticaBorkh.) [4]. Cate and Perkins [5] reported significant correlations between CCI and total chlorophyll in sugar maple (*Acer saccharum* Marsh.) leaves collected during the fall coloration period. These limitations notwithstanding, hand-held chlorophyll meters can still be effective tools for evaluating forest tree species. After establishing a general correlative relationship for a particular species, it is possible to use a chlorophyll meter in applications for which precise values are not necessary. For example, rapid assessment of relative chlorophyll and/or nitrogen content in sugar maple canopies would be of particular utility for managers or researchers investigating sugar maple decline, the symptoms of which include foliar nutrient deficiencies and reduced chlorophyll content [13]. The accuracy of a handheld chlorophyll meter in predicting chlorophyll and nitrogen content in growing-season sugar maple leaves has not been previously reported. collected data of twelve plant species and they proved significant relationships between SPAD readings and total extractable chlorophyll from the leaves. The chlorophyll meter readings depend on the age and genotype of corn. Chlorophyll meters have been used to quantify plant greenness for about fifteen years. Minolta SPAD chlorophyll meter can detect the N deficiency but it does not replace other traditional means to adjust fertilizer N rates [2, 30, 31].

In this study, the effect of municipal solid waste compost and gibberellin levels were assessed on thequality of tulip plants. For most traits, MSWC 10% leaded to good results. MSWC 30% delayed flowering time about 7 days. The most flowers and bulbs number were obtained in GA3100 mg.l^{-1} MSWC 10 and 20% in combination with peatmoss had the highest leaves chlorophyll content. It seems that soil salinity caused by high percentages of municipal solid waste compost that is a limiting factor for tulip growth [17].

The main objective of the present work was to study the effects of different plant growth regulators, gibberellic acid and benzyladenine on the chlorophyll contents of *Ficus benjamina*, *Schefflera arboricola* and *Dizigotheeca elegantissima* foliage plants.

MATERIALS AND METHODS

In 2013 year, *Ficus benjamina*, *Schefflera arboricola* and *Dizigotheeca elegantissima* plants were cultivated at the experimental farm of University Azad Jiroft. Three Test factorial methods in completely randomized design test with 4 repeats and 9 treatments were used for this expriment. Uniform plants size of 18-20 cm were selected, then transferred to greenhouse and were planted in pots with capacity of 20 kg soil. Greenhouse temperature was 22ºC to 28ºC during night and day, respectively. Plants, based on field water capacity, were uniformly irrigated.

The present work was conducted during the successive seasons of 2013 year at greenhouse of national research centre (research and production station). Plastic pots of 30 cm in diameter were used for cultivation *Ficus benjamina*, *Schefflera arboricola* and *Dizigotheeca elegantissima* pants in which that were filled with media containing a mixture of sand, rice husk, leaf composts and peat as 1:1:1:1 [v/v] during growth. The plants were fertilized with 3% liquid fertilizer in some doses after 4, 6 and 8 weeks from transplanting. This study was performed in three factorial test based on completely randomized design and 4 repeation with 9 treatments. Application of benzyladenine [0, 100 and 200 mg L^{-1}] and gibberellic acid [0, 100 and 200 mg L^{-1}] in which each contained 10 ml [0.l'%] Tween-20 surfactant. For each pot was used 40 cc of solution at each stage (three stages) with 15 days intervals [6].

Estimation of Chlorophyll Contents

Chlorophyll meter device works in such a way that there is an illuminating in the first part for production of red and infrared light and then there is a receptor to absorption the light passed through the leaf sample. This receptor changes the past light to analogue electrical signals. These signals are relayed by an amplifier and then changed to digital signals by a convertor. The signals are then interpreted by a microprocessor and digital numbers are presented in monitor and are saved automatically in memory. It should be noted that SPAD number is not a determination of chlorophyll rate; rather it is an estimation of chlorophyll concentration. This number has a high correlation with leaf chlorophyll rate. At first, for device calibration turn on the device and then read the number without put the leaf in the leaf chamber once and then read the numbers for three places of each leaf and then estimate the mean of these three numbers by pressing the average button. It should be noticed that the estimation should not be performed on leaf veins. SPAD number has the high correlation with leaf chlorophyll and nitrogen [10].

RESULTS AND DISCUSSION

Chlorophyll meter shows the relative density of leaf chlorophyll based on the rate of light passed through leaf in two wave lengths that their absorption is different by chlorophyll. The highest rate of chlorophyll absorption is in two blue and red wave lengths and the lowest rate is in green wave length and at infrared wave length chlorophyll doesn't have any absorption. Therefore, principle of this device is based on difference between red light illuminated to leaf and infrared light passed through leaf. Because with increasing in leaf chlorophyll the absorption rate of red light will be increased. So, the selected wave lengths for this device are red wave length (that has the highest absorption and is not affected by carotene) and infrared wave length (that has low absorption). Mean comparison using duncan method revealed that the highest chlorophyll content of *ficus Benjamina* flower was achieved by 200 $mg.L^{-1}$ GA_3 + 200 $mg.L^{-1}$ BA, followed by 100 $mg.L^{-1}$ BA 60 DAP which showed or non significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the growth regulator up to 200 $mg.L^{-1}$ GA_3 + 200 $mg.L^{-1}$ BA significantly increased chlorophyll content compared to control treatment. Compared to other concentrations, control and 100 $mg.L^{-1}$ GA_3 had the lowest chlorophyll content ($p<0.05$). The highest chlorophyll content of *Ficus benjamina* flower was achieved by 200 $mg.L^{-1}$ GA_3 + 200 $mg.L^{-1}$ BA, followed by 100 $mg.L^{-1}$ GA_3 + 200 $mg.L^{-1}$ BA for 120 DAP. Which showed significant difference from control treatment in either of the levels ($p<0.05$). Compared to other concentrations, control and 100 $mg.L^{-1}$ BA had the lowest chlorophyll content ($p<0.05$). Mean comparison using duncan method revealed that the highest chlorophyll content of *Ficus benjamina* flower was achieved by 200 $mg.L^{-1}$ GA_3, followed by 100 $mg.L^{-1}$ BA 180 DAP which showed significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the growth regulator up to 200 $mg.L^{-1}$ significantly increased chlorophyll content compared to control treatment with mean 25.49%. Compared to other concentrations, control and 100 $mg.L^{-1}$ BA had the lowest chlorophyll content ($p<0.05$). Results of the investigation showed that application of plant growth regulators in higher concentrations had positive effects on leaf chlorophyll content of *ficus Benjamina* foliage plants. They reported a better or equal prediction for the greenness index than the SPAD measurements. On the other hand, Kawashima and Nakatani [11] reported an index using only the red and blue color bands [(red - blue)/(red + blue)] as the most applicable function to estimate the chlorophyll content on plants. These results are in line with the results of the current study. Output from the Minolta SPAD-502 chlorophyll meter is a func tion of leaf transmittance in the red and near infrared portion of the electromagnetic spectrum (650 and 940 nm). Since the SPAD-502 meter underestimates chlorophyll content in water-stressed environments, subse quent discussion of chlorophyll content will be confined to the results of the direct measurement of chlorophyll by extraction. Increased chlorophyll content of leaves of GA_3 treated plants may be indication of increased rate of photosynthesis [12] and decreased chlorophyll degradation and increased chlorophyll synthesis and the increase in total chlorophyll content can also be attributed to involvement of growth regulators in promoting the synthesis of chlorophyll as well as development of chloroplast [9]. These results were in accordance with Kanjilal and Singh [12] who explained that application gibberlic acid increased chlorophyll content in chamomile. Similarly Singh and Hippalgaonkar [26] explained application of kinetin increased chlorophyll content.

Table 1- Effect of GA_3 and BA on Chlorophyll Contents of *Ficus benjamina* Plant. 60, 120 and 180 Day After Spray

GA_3	BA	Leaf Chlorophyll Index (SPAD)		
		60	**120**	**180 days**
0	0	12.58b	24.53d	51.1c
	100	18.57ab	27.08cd	54.09bc
	200	16.49b	28.32bcd	57.93abc
100	0	12.78b	32.62bc	60.69ab
	100	13.48b	27.74bcd	57.05abc
	200	13.75b	34.53b	62.49a
200	0	16.97b	33.98bc	64.68a
	100	13.66b	33.87bc	59.77ab
	200	23.66a	43.09a	57.99abc

Means followed by same letter are not significantly different at P< 0.05 probability using Duncan's test.

Mean comparison using duncan method revealed that the highest chlorophyll content of *Schefflera arboricola* flower was achieved by 200 $mg.L^{-1}$ GA_3 + 100 $mg.L^{-1}$, 100 $mg.L^{-1}$ GA_3 + 200 $mg.L^{-1}$ BA with 60 DAP which showed significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the

growth regulator up to 200 mg.L^{-1} GA_3+ 200 mg.L^{-1} BA significantly increased chlorophyll content compared to control treatment. Compared to other concentrations, control and 100 mg.L^{-1} GA_3 had the lowest chlorophyll content ($p<0.05$). Consist with findings by other workers [14, 20, 21] who reported that SPAD-502 meters give differing prediction responses for different plant species, the calibration lines from this investigation were species specific. Thus any attempt to draw calibration models necessitate individual regression model be developed for each particular species and cultivar. Varying leaf thickness effects the precision of SPAD-502 meter readings [29], thus increasing variability.

Table 2- Effect of GA_3 and BA on Chlorophyll Contents of *Schefflera arboricola* Plant. 60, 120 and 180 Day After Spray

GA_3	BA	Leaf Chlorophyll Index (SPAD)		
		60	**120**	**180 days**
0	0	4.62abc	41.64c	46.07c
	100	17.3d	45.95bc	49.78bc
	200	24.35abc	47.64bc	53.04abc
100	0	27.35ab	46.33bc	53.48abc
	100	23.6bc	48.22bc	54.03ab
	200	28.45a	51.14ab	57.08ab
200	0	20.97cd	56.49a	50.95bc
	100	28.55a	56.63a	52.58abc
	200	27.95ab	56.48a	59.99a

Means followed by same letter are not significantly different at P< 0.05 probability using Duncan's test.

The highest chlorophyll content of *Schefflera arboricola* flower was achieved by 200 mg.L^{-1} GA_3 + 100 mg.L^{-1} BA, 200 mg.L^{-1} GA_3 and 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} BA for 120 DAP. Compared to other concentrations, control treatment had the lowest chlorophyll content ($p<0.05$).

Mean comparison using duncan method revealed that the highest chlorophyll content of *Schefflera arboricola* flower was achieved by 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} BA and 100 mg.L^{-1} GA_3 + 100 mg.L^{-1} BA, 180 DAP which showed significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the growth regulator up to 200 mg.L^{-1} significantly increased chlorophyll content compared to control treatment with mean 28.06%. Compared to other concentrations, control and 100 mg.L^{-1} GA_3 had the lowest chlorophyll content ($p<0.05$). Results of the investigation showed that application of plant growth regulators in higher concentrations had positive effects on leaf chlorophyll content of *Schefflera arboricola* foliage plants. Mean comparison using duncan method revealed that the highest chlorophyll content of *Dizigotheeca elegantissima* flower was achieved by 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} for 60 DAP which showed significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the growth regulator up to 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} BA significantly increased chlorophyll content compared to control treatment. Compared to other concentrations, control and 100 and 200 mg.L^{-1} BA had the lowest chlorophyll content ($p<0.05$). The SPAD-502 meter provides a quick, nondestructive measure of the relative greenness of leaves at a specific moment of time. Chlorophyll concentration in leaves are thus well correlated with meter readings for Benjamin fig and cottonwood. The genetic load of hybrids evaluated in our trial is mostly subtropical whereas Waskom et al. [32] tested temperate climate materials. Diversity in genetic background may interfere on N metabolism, causing variations in leaf chlorophyll content [20].

The highest chlorophyll content of *Dizigotheeca elegantissima* flower was achieved by 100 mg.L^{-1} GA_3 + 200 mg.L^{-1} BA for 120 DAP. Compared to other concentrations, control treatment had the lowest chlorophyll content ($p<0.05$). Mean comparison using duncan method revealed that the highest chlorophyll content of *Dizigotheeca elegantissima* flower was achieved by 200 mg.L^{-1} GA_3 + 200 mg.L^{-1} BA and 200 mg.L^{-1} GA_3, for 180 DAP which showed significant difference from control treatment in either of the levels ($p<0.05$). Increasing concentration of the growth regulator up to 200 mg.L^{-1} significantly increased chlorophyll content compared to control treatment with mean 45.90%. Compared to other concentrations, control and 100 mg.L^{-1} BA had the lowest chlorophyll content ($p<0.05$). Results of the investigation showed that application of plant growth regulators in higher concentrations had positive effects on leaf chlorophyll content of *Schefflera arboricola* foliage plants. The point at which leaf chlorophyll reaches its maximum, has been termed "photosynthetic maturity" and dose not necessarily correspond to maximum leaf size. the increase of SPAD readings with plant growth has already been documented by [7, 20, 27].

Under high irradiance, chloroplast rearrangement within leaf cells can ingluence the amount of light absorbed by the leaf [28] and hence, chl meter readings.

Table 3- Effect of GA_3 and BA on Chlorophyll Contents of *Dizigotheeca elegantissima* Plant. 60, 120 and 180 Day After Spray

GA_3	BA	Leaf Chlorophyll Index (SPAD)		
		60	**120**	**180 days**
0	0	8.35d	12.22e	30.67d
	100	9.03cd	13.36de	33.95cd
	200	9.03cd	14.86bcde	38.66abc
100	0	10.21cd	14.33bcde	34.95cd
	100	11.76cd	17.9ab	36.37bcd
	200	16.76a	19.75a	39.3abc
200	0	16.81a	14.02cde	42.65ab
	100	16.28ab	16.12bcd	39.27abc
	200	17.03a	17.6abc	44.75a

Means followed by same letter are not significantly different at P< 0.05 probability using Duncan's test.

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