COMPARISON OF GREEN AND VARIEGATED FOLIAGE PLANT SPECIES BASED ON CHLOROPHYLL FLUORESCENCE PARAMETERS UNDER DIFFERENT LIGHT INTENSITIES

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Abstract

To compare green and variegated foliage plants namely; Hedera helix, Ardisia pusilla and Scindapsus aureus were treated under three different light intensities (2, 10 and 20 µmol m⁻² s⁻¹) using florescent lights for three months, and photosynthetic characteristics and chlorophyll content were measured. IMAGING-PAM chlorophyll fluorescence system was used to determine photosynthetic characteristics. The results showed that Fv/Fm (maximal PS-II quantum yield) and Y (II) (effective PS-II quantum yield) values were lower in variegated species compared to those in green species at 2 µmol·m 2 s⁻¹ light intensity. Although, Fv/Fm and Y (II) values in variegated S. aureus was lower than variegated H. helix, but variegated S. aureus survived and Hedera helix plants died after low light treatment. In contrast, Fv/Fm and Y (II) values were increased in green and variegated species with increasing the light intensity, except Fv/Fm value for variegated S. *aureus* which was slightly decreased at 20 μ mol m² s⁻¹ light intensity. The ETR (electron transport rate) value was also lower in both green and variegated species at 2 µmol m² s⁻¹ intensity. ETR values saturated in green and variegated plants with increasing the light intensity. Higher level of chlorophyll was obtained in green species compared to variegated species for both low and high light intensity treatments. Chlorophyll content in green species decreased at 20 µmol·m⁻²·s⁻ light intensity, whereas in variegated species, chlorophyll content increased with increasing the light intensities. The results of chlorophyll fluorescence parameters showed that especially low light intensity created stress condition in variegated foliage plants compared to green plants, and affected the normal growth and development.

Key words: Green and variegated leaf, Foliage plants, Chlorophyll fluorescence, Light intensity.

Introduction

Foliage plants (Hedera helix, Ardisia pusilla and Scindapsus aureus) are often used as house plants because of their attractive foliage and their ability to survive and grow under limited indoor light (Chen and Henny, 2008; Kim et al., 2012). Foliage plants have widely diverse forms, patterns of foliar variegation, and colors. Based on their appearances, foliage plants can be simply categorized into three groups; green-leaf, variegated-leaf, and flowering foliage plants (Chen et al., 2005). Among the characteristics of foliage plants, variegation is an important trait, which is popular and provides unique visual appearance and aesthetic variation in interior design, making it one of the considerations of consumer's preferences in purchasing decisions (Chen et al., 2004). Leaf variegation can be appeared by differential gene expression, leaf blisters, virus, or genetic mosaics such as chimeras (Marcotrigiano, 1997), and variegation appearance can be altered by environmental factors, particularly light intensity (Tilney-Bessett, 1986). However, it is established that foliage plants can effectively improve the indoor air quality by reducing many components including volatile organic compounds (Wolverton & Wolverton, 1993; Kim et al., 2010).

Environmental stress factors, such as light, temperature, chilling, heavy metals and drought may produce deleterious effects on photosynthesis and consequently damage higher plant or algal growth. The most limiting factor for plant growth is reduced light intensity (Manaker, 1997). Although the light intensity of outdoor is generally higher than 1000 μ mol·m⁻²·s⁻¹ at sunny days, typical indoor light intensity is less than 40 μ mol·m⁻²·s⁻¹ (Manaker, 1997). Because plants growth depends on light (i.e., photosynthesis), plants require a particular light environment for proper growth and development (Maloof *et al.*, 2001). Moreover, to sustain higher photosynthetic capacity or survival, plants modify their morphology, physiology, and biomass allocation at different light conditions (Sims & Pearoy, 1992; Feng *et al.*, 2004). However, different plant species respond differently to light intensity.

During the last years, the measurement of chlorophyll fluorescence has been proved to be a quantitative, powerful tool, which is widely used to examine the photosynthetic parameters in algae and plants. Measurements are both non-destructive and non-invasive, and applications range from a method of rapid identification of injuries in leaves without visual symptoms that permit to assess the photosynthetic performance in vivo (Baker, 2008; Krause and Weis, 1991; Woo et al., 2008). Fluorescence imaging reveals a wide range of internal leaves characteristics, including spatial variations due to differences in physiology, development, nutritional state, pigment distribution, morphology, and optical properties. It has been showed that measurement of variable chlorophyll fluorescence from intact plants offers several values that are very useful for understanding the functioning of specific process of photosynthesis. If photosynthesis or related or physiological processes are altered due to biotic or abiotic stresses, the yield and kinetics fluorescence will be changed significantly. IMAGING-PAM method employs a combination of chlorophyll and saturation type of light, which enables the analysis of chlorophyll fluorescence. Several studies have been reported on measurement of chlorophyll fluorescence characteristics focused on the effects of environmental stress factors such as light intensity, temperature, drought, and chilling in various plants (Li *et al.*, 2008; Islam *et al.*, 2011; Zhang *et al.*, 2011; Fu *et al.*, 2012; Hogewoning *et al.*, 2012).

The objective of this study was to compare green and variegated foliage plant species (*H. helix, A. pusilla* and *S. aureus*) depending on the chlorophyll fluorescence parameters, and chlorophyll content under different light intensities. In this paper we show that variegated foliage plant species were more susceptible at low light intensity compared to the green plant species, which may help the house plant growers to maximize the light condition for normal growth and development of foliage plants in the indoor environment for reducing the pollutants.

Materials and Methods

Plant materials: Two-years-old English Ivy (Hedera helix), Small Coralberry (Ardisia pusilla) and Golden Phothos (Scindapsus aureus) foliage plant species were obtained from a commercial nursery at Suwon, South Korea. Both green and variegated leaves of each plant species were used as plant materials in this study. The plants were transplanted in 19-cm diameter plastic pots containing a uniform growing medium comprised of Mix-4 (Sun Grow Horticulture, Bellevue, WA), bark-humus (Biocom. Co., Seoul, Korea), and sand at 5:1:1, v/v/v. Mix # 4 contained Canadian sphagnum peat-moss (55% to 65% by volume), perlite, dolomitic lime, gypsum, and a wetting agent. The potted plants were grown in a shaded greenhouse for two weeks after transplanting. Temperature of the shaded greenhouse ranged from 24 to 26°C. Then all plants were acclimated at indoor environment with a light intensity of 20 μ mol·m⁻²·s⁻¹ for two weeks and the room temperature was 23 ± 2^{00} C (Kim et al., 2010). After acclimation, the plants were treated under three different light intensities (2, 10 and 20 μ mol·m⁻²·s⁻¹) in the different chambers for three months using florescent lights with a temperature of $24 \pm 1^{\circ}$ C; the photoperiod of each chamber was 12/12-h (day/night) for all species. The plants were watered every 3 day with the excess water allowed to drain. Morphological responses of the plants on different light intensity treatment were observed and photograph was taken after three months.

Measurement of chlorophyll fluorescence: The chlorophyll fluorescence of green and variegated leaves of foliage plant species (*H. helix, A. pusilla* and *S. aureus*) was determined to compare the photosynthetic performance. Chlorophyll fluorescence was measured using IMAGING-PAM (Heinz Walz GmbH, Effeltrich, Germany) chlorophyll fluorescence system as described by Siebke & Weis, 1995 and Rascher *et al.*, 2001. All chlorophyll fluorescence kinetics parameters were measured on the adaxial side of the same leaf after the leaves had been dark adapted for 10 min (Schreiber *et al.*, 1986).

The dark fluorescence yield (F_0) was determined after dark adaptation using F₀, F_m-key. After dark adaptation, normally all PS-II reaction centers open with a weak non-actinic modulated light pulse (0.1 µmol·m $^{2}\cdot s^{-1}$) at a low frequency (1 Hz). The maximal fluorescence yield (F_m) was determined after dark adaptation, and the $F_{\rm m}$ value was assessed at the plateau level reached during application of a saturation pulse at 20-s intervals, with all PS-II reaction centers close. Measurement of F_m involves averaging of three images recording. The maximum fluorescence yield (F_m') was observed in illuminated samples, which normally lowered with respect to $F_{\rm m}$ by non-photochemical quenching. The variable fluorescence (F_v) was calculated as $F_v = (F_m - F_0)$. Maximal PS-II quantum yield (F_v/F_m) was calculated after dark adaptation according to the equation: $F_v/F_m = (F_m - F_0)/F_m$. The effective PS-II quantum yield, Y (II) was calculated according to Genty et al. (1989) formula as: Y (II) = $(F_{\rm m}'-F)/F_{\rm m}')$ by the Imaging-Win software (Walz), represented the PS-II quantum efficiency.

After kinetic induction fluorescence, rapid light curve (RLC) measurements were obtained through the application of a series of 20-s light exposure with increasing photosythetically active radiation (PAR) values (50, 100, 150, 200, 250, and 300 μ mol·m⁻²·s⁻¹). Each light increment was followed by a saturating pulse for assessment of fluorescence parameters. The apparent electron transport rate (ETR) was calculated by the equation:

ETR= 0.5 X Y (II) X PAR X 0.84 μ mol·m⁻²·s⁻¹

where 0.5 is a multiplication factor because transport of a single electron requires the absorption of 2 quanta, and 0.84 is the specific fraction of incident quanta absorbed by leaf (Ralph *et al.*, 2005). Photosynthetically active radiation (PAR) is the display of light intensity in μ mol·m⁻²·s⁻¹, which is corresponds to the intensity of actinic light as well as also by the intensity and frequency of the measuring light, as defined by the intrinsic PAR-list. Simultaneously, ETR values were obtained automatically using the Imaging-Win software.

Chlorophyll content measurement: the For relationship of chlorophyll content and fluorescence parameters in foliage plant species, chlorophyll content was measured after three months of treatment with different light intensity (2, 10 and 20 μ mol·m⁻²·s⁻¹). The portable chlorophyll meter (SPAD-502, Minolta, Japan) was used to measure the chlorophyll content (Netto et al., 2005). Both green and variegated leaves of the foliage plant species (H. helix, A. pusilla and S. aureus) were selected randomly for each treatment to measure chlorophyll content of third leaf from the top, and three readings was measured for each third leaf, avoiding main veins during measurement. Three replications were used for each treatment and mean values were expressed for chlorophyll level.

Results

Green and variegated foliage plant species (*H. helix*, *A. pusilla* and *S. aureus*) were grown in separate chambers and treated with three different light intensities (2, 10 and 20 μ mol·m⁻²·s⁻¹) for three months. The morphological responses of the foliage plants on different light intensity treatments are shown in Fig. 1. Variegated plant species showed more susceptibility compared to green species at low light intensity treatment, and the variegated Ivy (*H. helix*) plants almost died after three months at 2 μ mol·m⁻²·s⁻¹ light intensity treatment. However, there was a common problem of leaf abscission associated with the growth and it was used under low light conditions in this study, which is similar to the responses of *Ficus benjamina* L. to low light level (Chen *et al.*, 2001; Li *et al.*, 2009).

To evaluate the different functional levels of photosynthesis, Chlorophyll fluorescence parameters were measured using both green and variegated leaves. The dynamics of F_0 , F_m , F_m' and Fv/Fm were measured under three different light intensity (2, 10 and 20 µmol·m $^{2} \cdot s^{-1}$) treatments. The results showed that Fv/Fm (maximal) PS-II quantum yield) value was lower in all variegated species compared to green species at low light intensity (Fig. 2). The Y (II) (effective PS-II quantum yield) value was also decreased in all variegated species compared to green species at low light (2 μ mol·m⁻²·s⁻¹) intensity (Fig. 3). Although Fv/Fm and Y(II) values in variegated S. aureus was little lower compared to variegated Ivy (H. *helix*) plants, but variegated S. aureus plants survived, and variegated H. helix plants died at 2 μ mol·m⁻²·s⁻¹ light intensity treatment after three months. These results indicating that variegated H. helix plants are more susceptible to low light intensity, and variegated S. aureus plants have tolerance to low light, genetic factors may responsible for this phenomenon along with the light

factor. On the other hand, Fv/Fm and Y (II) values were increased for both green and variegated species with increasing the light intensity, and highest values were observed at 20 µmol·m⁻²·s⁻¹ light intensity treatment, except the Fv/Fm value for variegated *S. aureus* that was slightly decreased at 20 µmol·m⁻²·s⁻¹ light intensity.

The results of electron transport rate (ETR) showed that ETR value was lower in both green and variegated plant species at low light (2 µmol·m⁻²·s⁻¹) intensity (Figs. 4, 5, and 6). The ETR value was increased with increasing the light intensity in both green and variegated S. aureus, variegated *H. helix* and green *A. pusilla*, whereas the ETR value for green *H. helix* and variegated *A. pusilla* that was slightly decreased at 20 µmol·m⁻²·s⁻¹ light intensity. The saturation points based on the ETR values were higher in the variegated species compared to green species. It may be due to less green area present in the variegated foliage leaves compared to green leaves. The ETR values in green H. helix and variegated A. pusilla may be saturated at 10 μ mol·m⁻²·s⁻¹ light intensity, as it is well known that different plant species respond differently to the light intensity. Photoinhibition is manifested by a decrease of the photosynthetic electron transport rate (ETR) (Critchley, 1981; Ogren and Oquist, 1984a), and it has been suggested that this reduction is caused by an increased radiationless decay at the reaction centers of PS-II (Powles & Bjorkman, 1982).

Chlorophyll content in both green and variegated leaves of foliage plant species was measured. The total chlorophyll content was higher in all green species compared to variegated species in both low and high light intensity treatments (Table 1). Chlorophyll content was reduced in both green and variegated species at low light intensity (2 μ mol·m⁻²·s⁻¹). Chlorophyll content in green plant species was slightly decreased at 20 μ mol·m⁻²·s⁻¹ light intensity, whereas in variegated species chlorophyll content gradually increased with increasing the light intensity.



Fig.1. Morphological response of green and variegated foliage plants (*Hedera helix*, *Ardisia pusilla* and *Scindapsus aureus*) under three different light intensity (2, 10 and $20 \,\mu$ mol·m⁻²·s⁻¹) treatments after three months.



Fig. 2. Dynamics of *Fv/Fm* (maximal PS-II quantum yield) in the dark-adapted leaf samples of green and variegated *Hedera helix*, *Ardisia pusilla* and *Scindapsus aureus* under three different light intensity (2, 10 and 20 μ mol·m⁻²·s⁻¹) treatments after three months. Data are the mean of three replications.





Fig. 3. Dynamics of Y (II) (effective PS-II quantum yield) in the dark-adapted leaf samples of green and variegated *Hedera helix*, *Ardisia pusilla* and *Scindapsus aureus* under three different light intensity (2, 10 and 20 μ mol·m⁻²·s⁻¹) treatments after three months. Data are the mean of three replications.



Fig. 4. Light-response curve of ETR (apparent electron transport rate) in the dark-adapted leaf samples of green and variegated *Hedera helix* under different light intensity treatments (2, 10 and 20 μ mol.m⁻²·s⁻¹) after three months. Rapid light curve (RLC) measurements were obtained through the application of a series of 20-s light exposures with increasing photosynthetically active radiation (PAR) values (0, 50, 100. 150, 200, 250, and 300 μ mol·m⁻²·s⁻¹). Data are the mean of three replications.

Fig. 5. Light-response curve of ETR (apparent electron transport rate) in the dark-adapted leaf samples of green and variegated *Ardisia pusilla* under different light intensity treatments (2, 10 and 20 μ mol·m⁻²·s⁻¹) after three months. Rapid light curve (RLC) measurements were obtained through the application of a series of 20-s light exposures with increasing photosynthetically active radiation (PAR) values (0, 50, 100. 150, 200, 250, and 300 μ mol·m⁻²·s⁻¹). Data are the mean of three replications.



Fig. 6. Light-response curve of ETR (apparent electron transport rate) in the dark-adapted leaf samples of green and variegated *Scindapsus aureus* under different light intensity treatments (2, 10 and 20 μ mol·m⁻²·s⁻¹) after three months. Rapid light curve (RLC) measurements were obtained through the application of a series of 20-s light exposures with increasing photosynthetically active radiation (PAR) values (0, 50, 100. 150, 200, 250, and 300 μ mol·m⁻²·s⁻¹). Data are the mean of three replications.

Discussion

Chlorophyll fluorescence provides insights into a plant's ability to tolerate environmental stresses damage and extent to which those stresses damage the photosynthetic apparatus (Maxwell & Johnson, 2000). Therefore, fluorescence parameters are excellent tools for studying the stress-induced changes in PS-II (Naumann et al., 2008) that are believe to play a key role in the response of photosynthesis to environmental stresses (Zribi et al., 2009). Fv/Fm, as one of the most important chlorophyll fluorescence parameters, reflects the maximal photochemical efficiency of the active center of PS-II in the dark. The results of the present study showed that Fv/Fm and Y (II) values were decreased in all foliage plant species, especially in variegated plant species at low light intensity (2 µmol·m $^{2} \cdot s^{-1}$) treatment, which indicate the occurrence of photoinhibitory damage in response to light stress. Moreover, the results of Fv/Fm, Y (II), and ETR are mostly supported with the chlorophyll content results in this study under different light intensity treatments.

Extremely strong light often decreases chlorophyll content in plant leaves because of the inhibition of chloroplast formation. In contrast, under weak light, the number of chloroplasts per unit leaf area in plant leaves drops, but the larger chloroplasts results in increased chlorophyll content in plant leaves (Fu et al., 2012). Exposure to a high light intensity induced chlorophyll loss, led to a marked decline in the photochemical efficiency of PS-II and caused considerable degradation of Rubisco in both susceptible and tolerant varieties of Sorghum bicolor L. (Jagtap et al., 1998). In this study, we show chlorophyll content declined in both green and variegated species at low light intensity (2 μ mol·m⁻²·s⁻¹), and higher chlorophyll content was obtained in all green species at 10 μ mol·m⁻²·s⁻¹ light intensity, and then chlorophyll content slightly decreased. On the other hand, for variegated species higher level of chlorophyll was found at 20 μ mol·m⁻²·s⁻¹ light intensity treatment.

Li et al. (2008) reported the significant decrease in Fv/Fm and Y (II) values under drought stress condition in cucumber seedlings. Zhang et al. (2011) showed the low values of the chlorophyll fluorescence parameters $(F_{\rm m}, Fv \text{ and } Fv/Fm)$ in un-shaded oriental lily (Lilium auratum L.) plants compared to shaded plants, which led to reduced productivity of net photosynthetic rate. They also suggested that high light intensity damaged the photosystem-II, leading to photoinhibition. Photoinhibition is accompanied by slower rates of electron transport, a lower quantum yield of photosynthesis and diminished plant growth. Greater Fv/Fm value results in higher light utilization efficiency and stronger ability of plants to adapt to low-light conditions. Under normal physiological conditions, the Fv/Fm values of the vast majority of C_3 plants are between 0.8-0.84 ranges (Fu et al., 2012). It is well known that when the Fv/Fm value of a plant is below 0.8–0.84 range, the plant is exposed to some environmental stresses, such as drought (Yang et al., 2004; Lu et al., 2004) photo-oxidative stress (Shao et al., 2013), low light (Chen et al., 2001; Li et al., 2009) or high temperature (Fracheboud et al., 1999; Wang et al., 2004) or light stress (Groom and Baker, 1992; Li et al., 2002; Chen et al., 2006). However, it has been reported that different plant species respond differently to the light intensity. In this study, we showed that in 2 μ mol.m⁻²·s¹ light intensity treatment, the *F*v/*F*m values was lower than 0.7 in all variegated species, whereas the Fv/Fm values was higher than 0.7 in two green species except S. aureus. In contrast, at 10 µmol.m⁻²s⁻ ¹light intensity treatment, the *Fv/Fm* values was around 0.75 in both green and variegated H. helix and A. pusilla species, but the Fv/Fm value was about 0.8 in green S. aureus and below 0.7 in variegated S. aureus. To best of our knowledge, there is no available report on comparative study of chlorophyll fluorescence parameters in green and variegated foliage plants under different light intensity. Further studies are necessary to fix up the appropriate light intensity to be used for each foliage plant species for growing in the indoor environment.

Light intensities (µmol·m ⁻² ·s ⁻¹)	Type of leaves	Chlorophyll content (SPAD)		
		Hedera helix	Ardisia pusilla	Scindapsus aureus
2	Green	31.63 ± 0.20	50.62 ± 3.42	34.01 ± 1.08
	Variegated	21.41 ± 1.17	33.00 ± 1.04	28.04 ± 2.15
10	Green	32.07 ± 0.62	58.92 ± 1.52	36.91 ± 0.53
	Variegated	23.02 ± 0.35	44.84 ± 0.48	30.42 ± 0.89
	Green	31.59 ± 0.84	53.21 ± 1.07	36.47 ± 0.56
20	Variegated	24.13 ± 0.27	46.15 ± 0.57	31.08 ± 0.93

Table 1. Measurement of chlorophyll content in leaves of green and variegated foliage plant species under different light intensity treatments after three months (data are the mean of three replications).

Conclusion

This result showed that normal growth and photosynthetic performances were affected in green and variegated foliage plant species by low light intensity treatment. Chlorophyll content, Fv/Fm, Y (II) and ETR values were declined with light stress aggravated, and variegated foliage plant species were more susceptible compared to the green plant species at low light intensity. This meant that light stress decreased maximal photosynthetic electron transport potential and subsequently decreased the capacity of preventing light induced damages. It is possible that light stress may regulate the photosynthesis capacity or linear electron transport rate in leaves of foliage plants. Based on the chlorophyll fluorescence parameters, the present finding demonstrated that low light intensity created more stress condition in variegated foliage plant species compared to the green plant species, which may help to the consumers standardizing the light condition for foliage plants to be grown in the indoor environment for reducing the airborne pollutants.

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