JPPE Check for updates

An Analysis of the Growth and Photosynthetic Responses of Potted *Veronica pusanensis* Y.N.Lee according to the Shading Levels

Se Hoon Park1[†], Jae Hwan Lee^{1,2[†]}, and Sang Yong Nam^{3,4*}

¹Graduate Student, Department of Environmental Horticulture, Sahmyook University, Seoul 01795, Republic of Korea
 ²Researcher, Natural Science Research Institute, Sahmyook University, Seoul 01795, Republic of Korea
 ³Professor, Department of Environmental Horticulture, Sahmyook University, Seoul 01795, Republic of Korea
 ⁴Director, Natural Science Research Institute, Sahmyook University, Seoul 01795, Republic of Korea

ABSTRACT

Background and objective: *Veronica pusanensis*, an endemic species of South Korea belonging to the Plantaginaceae family, is found in Busan, South Korea. Due to its high ornamental value, it is expected to be utilized as a flower crop. However, currently it is an endangered species with its habitats being destroyed and reduced.

Methods: Therefore, in this study, we analyzed the growth and photosynthetic responses of potted plants of *V. pusanensis* under different shading levels to enable mass production. Polyethylene (PE) shading films were selected as the shading material, and shading levels were designed at 0, 35, 45, 60, 75, and 99%, respectively.

Results: Results showed that shoot height, shoot width, ground cover, leaf length, leaf width, shoot fresh and dry weight, chlorophyll content (SPAD units), and chlorophyll fluorescence parameters F_v/F_m and Pl_{ABS} were highest under the 35% shading level. This indicates that it is relatively more desirable to grow *V. pusanensis* in shade culture compared to under direct sunlight. Meanwhile, root fresh weight and dry weight were highest under the 0% shading level.

Conclusion: Therefore, it is recommended to grow *V. pusanensis* under direct sunlight to significantly increase root biomass when the purpose is to facilitate rootage when transplanting plants for habitat restoration. On the other hand, for the cultivation of *V. pusanensis* as an ornamental flower crop, it is recommended to grow the plants under the 35% shading level to significantly increase plant sizes and maintain ensure the proper functioning of photosynthetic responses in photosystem II (PSII).

Keywords: chlorophyll fluorescence, CIELAB, phenotypic plasticity, Pseudolysimachion pusanensis, SPAD units

Introduction

Veronica was previously known to belong to the Scrophulariaceae family, but now belongs to the Plantaginaceae family (Olmstead, 2002). *Veronica* is distributed throughout the Northern Hemisphere and the Australasian region, and various species are discovered also in Western Asia and New Zealand (Albach et al., 2004). Approximately 450 species of *Veronica* are distributed worldwide (Albach et al., 2008), and they grow naturally in broad and diverse regions from coastal to alpine areas due to their excellent ecological adaptability (Albach et al., 2005). *Veronica* plants live in diverse environments such as terrestrial, semi-aquatic, and aquatic depending on the species, showing excellent adaptability (Salehi et al., 2019). In terms of medicinal use, extracts of *Veronica* plants are known

^{*}Corresponding author: Sang Yong Nam, namsy@syu.ac.kr, 10 https://orcid.org/0000-0002-4351-447X



© 2023 by the Society for People, Plants, and Environment. This is a Peer-Reviewed Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/4.0/) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

[†]These authors contributed equally to this work.

This paper was supported by the Sahmyook University Research Fund in 2022.

Received: April 17, 2023, Revised: May 16, 2023, Accepted: May 23, 2023

First author: Se Hoon Park, pseh1005@naver.com, D https://orcid.org/0009-0004-4247-3145

Jae Hwan Lee, dlwoghks1236@naver.com, 💿 https://orcid.org/0000-0002-4621-5942

to be effective in eliminating free radicals (Harput et al., 2011), have anti-inflammatory effects, and show cytotoxic activity in cancer cells such as KB epidermoid carcinoma and B16 melanoma (Harput et al., 2002). Moreover, *Veronica* plants also showed an effect in treating rheumatism (Beara et al., 2015), lung and respiratory diseases (Grundemann et al., 2013), and antithrombosis and wounds (Küpeli et al., 2005), thereby expected to be used effectively as a medicinal crop.

V. pusanensis is a rare plant found in Busan, South Korea and is an endangered species that needs protection because its habitats are decreasing and it is damaged by poaching (Shin et al., 2012). Since V. pusanensis grows as a creeping habit and tends to cover the ground, it is expected to be used as a ground cover plant in coastal areas or gardens (Song et al., 2020a), and it can also be supplied as flower crops if they can be mass-produced since it has excellent ornamental value. Various physiological studies have been conducted in the past, such as a cuttage study using auxin in V. pusanensis (Kim et al., 2021), an investigation on salinity tolerance (Kwon et al., 2022), a study on light intensity treatment (Song et al., 2020a), a physiological exploration on the growth and flowering response according to photoperiod and temperature treatment (Song et al., 2020b), and a study on sprouting response according to temperature treatment (Song et al., 2019). However, there is insufficient technology for mass production and protection of the plant of V. pusanensis from abiotic stress during the cultivation period. As previously mentioned, V. pusanensis is an endangered species with a small wild population and rapid habitat destruction. Thus, by establishing the cultivation technology for mass production, it would be possible to help restore the habitats and also supply this species as a flower crop.

Potted cultivation has a relatively small soil volume and soil moisture content due to the limited pot capacity and thus has high requirements for management compared to open-field cultivation (Lee and Nam, 2022b). However, unlike open-field cultivation, potted cultivation allows plant movement and facilitates the control of plant growth. Commercial plastic pots are typically brown or black, and these colors can significantly increase soil temperature during intense summer sunlight, leading to moisture loss and causing complex abiotic stress for plants (Lee and Nam, 2022b). Therefore, it is necessary to come up with methods to protect plans such as shading.

To induce the stable growth of plants, newly planted or cutting plants must not be exposed to direct sunlight, and the plant should be protected from rapid environmental changes by using shading materials such as shading nets or shading films (Fowler and Chaffee, 2010; Semchenko et al., 2012). Plants grown under adequate shading levels are protected from rapid temperature rise, photodamage, and lack of soil moisture, and can be managed in a relatively stable environment (Nam et al., 2022). Moreover, since they are relatively low-maintenance and have a long irrigation cycle, they also help reduce labor during cultivation (Nam et al., 2022). Depending on the plant species, the plant cells might be damaged when exposed to strong direct sunlight, which causes the chloroplasts on the cell surface to move to the side walls, and this is referred to as chloroplast avoidance movement (Kasahara et al., 2002). Chlorophyll fluorescence analysis is one of the methods to identify plant stress levels and the inactivity of the reaction center of photosystem II (PSII), which can be done in the non-destructive investigation (Lechaudel et al., 2010; Lee et al., 2022b; Shim and Jeon, 2022). Chlorophyll fluorescence analysis that had been applied in various studies in the past had its usefulness proved by being used in assessing the photosynthetic efficiency and stress of plants through various fluorescence parameters such as F_v/F_m, Φ_{Do} , ABS/RC, DI_o/RC, and PI_{ABS} (An et al., 2022; Choi et al., 2022; Kwon et al., 2021; Lee et al., 2022b; Oh and Lee, 2022; Yang et al., 2022).

Accordingly, this study established the fundamental data on the growth and photosynthetic responses of *V. pusanensis* affected by shading levels to contribute to the mass production of *V. pusanensis*.

Research Methods

Plant material

As the plant material, we selected *Veronica pusanensis* which is an endemic species of South Korea and has high ornamental value. We selected plants around the shoot height of 3 cm and shoot width of 5 cm and applied them to the study.

Experimental environment and shading levels

The study was conducted for a total of 5 weeks from May 9 to June 14, 2022, at the experimental greenhouse $(37^{\circ}38'40" \text{ N } 127^{\circ}06'25" \text{ E})$ in the Department of Environmental Horticulture, Sahmyook University. Plants were planted in round plastic pots with a diameter and height of 11 × 10.5 cm, at the center of the pot. Here, we used horticultural substrates containing fertilizers (Hanareumsangto, Shinsung Mineral, South Korea). We designed six shading levels: no shade (i.e., direct sunlight) (0%), greenhouse glass (35%), greenhouse glass and 1 layer of clear polyethylene (PE) shading film (45%), greenhouse glass and 1 layer of white PE shading film (60%), greenhouse glass and 2 layers of white PE shading film (75%), and greenhouse glass and 1 layer of black PE shading film (99%). For the photo-

 Table 1. Shading levels of photosynthetic photon flux density

 (PPFD) in this study

_				
	Standard shading	Relative shading	g PPFD	
	levels (%)	levels (%)	$(\mu mol m^{-2} s^{-1})$	
	0	0	1783.2 ± 193.2^{z}	
	35	36.1	1140.9 ± 143.4	
	45	44.2	996.1 ± 121.6	
	60	60.8	699.8 ± 94.8	
	75	74.1	460.6 ± 58.0	
	99	99.1	16.3 ± 4.9	

^zMean \pm standard deviation

synthetic photon flux density (PPFD) of each shading level, we calculated the mean and standard deviation of the measurements taken at 1 p.m. once a week on sunny days using a portable spectral radiometer (SpectraPen mini, Photon Systems Instruments, Czech Republic) (Table 1).

The plants of all treatments including the control (0% shading level) were arranged on beds that are 1 m high, and the average temperature in the greenhouse during the study was $20.9 \pm 2.1^{\circ}$ C, and the average temperature of the outdoor area where the control was placed was $20.1 \pm 2.0^{\circ}$ C (Fig. 1A). Here, the relative humidity in the greenhouse was $58.5 \pm 9.5\%$, and the relative humidity of the outdoor area was $60.0 \pm 9.6\%$ (Fig. 1B). The average cloud cover was 4.6 ± 2.2 okta, and this was evaluated by visual observation, which is the same as the measurement method of Korea Meteorological Administration, at 1 p.m. every day (Fig. 1C). The plants were watered 3 times a week. In this study, watering continued until gravitational water drainage occurred.

Measurement methods for growth and leaf color parameters

We measured shoot height, shoot width, root length, shoot fresh and dry weight, root fresh and dry weight, ground cover, leaf length, leaf width, and leaf color of CIELAB L^* , a^* , and b^* values. Here, we measured shoot height from the soil surface in the pot up to the point where the plant is the tallest and shoot width by measuring the broadest area of the plant seen from above. For root length, we measured the longest of the plant roots. Secondary anal-



Fig. 1. Temperature, relative humidity, and cloud cover in this study. Cloud cover index, 0 okta: sky clear; 1-2 okta: few clouds; 3-4 okta: scattered; 5-7 okta: broken; 8 okta: overcast; 9 okta: sky obscured.

ysis was conducted for ground cover by squaring the shoot width of the plant width. Fresh weight was measured after washing the plant and naturally drying it for 12 hours in an enclosed space, and dry weight was measured after hot air drying of the plant for 24 hours using a hot air dryer (HK-DO135F, HANKUK S&I, South Korea). CIELAB values $(L^*, a^*, and b^*)$ were measured by setting the spectrophotometer (CM-2600d, Konica Minolta, Japan) to CIELAB D65/10° with reference to the leaf color measurement method Lee et al. (2022a), after which we collected data including specular components. For RHS values, we compared the results of each of L^* , a^* , and b^* with the Royal Horticultural Society (RHS) colour charts edition V and evaluated the leaf colors by selecting 2 approximate RHS values for each treatment. In addition, for leaf colors, we converted the means of CIELAB L^* , a^* , and b^* to converted colors using Converting Colors designed by Zettl (2023).

Measurement methods for chlorophyll fluorescence

We analyzed the chlorophyll content and photosynthetic state of V. pusanensis affected by shading levels. Here, we measured the chlorophyll content using a portable chlorophyll meter (SPAD-502, Konica Minolta, Japan) and estimated the chlorophyll content relative to the same area for each treatment using SPAD units. We measured chlorophyll fluorescence responses using a portable chlorophyll fluorometer (FluorPen FP 110/D, Photon Systems Instruments, Czech Republic) by measuring the part where the leaf veins do not pass among the central part of the fully developed leaf. Here, before measuring the chlorophyll fluorescence, the plant was dark-adapted for approximately 15 minutes using dark-adaptation leaf clips according to the manufacturer's guidelines (PSI, 2023), and we measured 5 times each in 3 replications on June 14, 2022, which is the end date of the study. We selected and applied five chlorophyll fluorescence parameters that can be used as plant stress indicators. We used the following equations to calculate F_v/F_m (1) representing the maximum quantum yield of photosystem II (PSII), Φ_{Do} (2) representing the probability that absorbed photons will be dissipated, ABS/RC (3) representing absorption flux per reaction center, DI₀/RC (4) representing dissipated energy flux per reaction center, and PI_{ABS} (5) that represents the performance index of absorption basis (PSI, 2023; Stirbet and Govindjee, 2011).

$$F_v / F_m = (F_m - F_o) / F_m$$
 (1)

$$\Phi_{Do} = 1 - \Phi_{P_o} = F_o / F_m \tag{2}$$

$$ABS/RC = M_o \cdot (1/V_j) \cdot (1/\Phi_{P_o})$$
(3)

$$DI_o / RC = (ABS / RC) - (TR_o / RC)$$
(4)

$$PI_{ABS} = (RC/ABS) \cdot [\Phi_P / (1 - \Phi_P)] \cdot [\Psi_o / (1 - \Psi_o)] \quad (5)$$

Statistical analysis

Analysis of variance (ANOVA) was conducted using SAS 9.4 (SAS Institute, USA) to analyze the results of the study. To compare the means, statistical analysis was conducted using Duncan's multiple range test at p < .05. The study was in a completely randomized design, with 5 plants assigned to each treatment in 3 replications.

Results and Discussion

Analysis of plant sizes, biomass, and leaf color

Shading levels had diverse effects on the plant sizes of *Veronica pusanensis* (Fig. 2). The results showed that shoot height of *V. pusanensis* was highest at 14.78 cm under the 35% shading level (Fig. 3A). *Sedum zokuriense* showed the highest shoot height under the 65% shading level (Lee et al., 2021b), whereas *Sarcandra glabra* showed the highest shoot height under the 50% shading level and *Ardisia crenata* under the 35% shading level (Bae et al., 2016), indicating that the preferred shading level (Bae et al., 2016), indicating that the preferred shading level (s, indicating that shade culture is better than cultivation under direct sunlight in order to significantly increase the plant shoot sizes (Fig. 3B). Root length was longest at 21.87 cm under the 60% shading level (Fig. 3C).



Fig. 2. The plant shape of Veronica pusanensis grown under shading levels for 5 weeks.



Fig. 3. Plant sizes of *V. pusanensis* as affected by shading levels for 5 weeks. Vertical bars indicate the standard error, and asterisks (***) indicate significant at $\rho \langle .001$. Different lowercase letters indicate significant differences at $\rho \langle .05$ based on Duncan's multiple range test (DMRT).

A previous study reported that root length of *V. nakaiana* and *V. pyrethrina* was in inverse proportion to the increase of shading levels (Kwon et al., 2020). On the contrary, in this study, root length increased along with shading levels from 0% to 60%, showing conflicting results from a previous study, but the results from 75% to 99% shading levels were similar to a previous study. Like the results of shoot width, ground cover was high at 868.1 and 861.8 cm² under the 35 and 45% shading levels (Fig. 3D). Leaf

length was highest at 3.57 cm under the 35% shading level (Fig. 3E). Meanwhile, leaf width was highest at 3.46 cm under the 35% shading level, which was similar to the results of shoot height, shoot width, and leaf length (Fig. 3F). A previous study showed similar results as this study with *Litsea japonica* showing the highest leaf length and leaf width under the 35% shading level, but *Osmanthus fragrans* var. *aurantiacus* and *Pittosporum tobira* showed the highest leaf length and leaf width under the 50% shad-

ing level (Choi et al., 2012a), indicating that the responses to leaf growth according to shading levels vary among species. Phenotypic plasticity is the ability of a plant to change its morphological characteristics so that it can adapt to the changing growth environment (Bradshaw, 1965; DeWitt et al., 1998; Sultan, 1987). Among phenotypic plasticity, shade-induced phenotype induces the plant to show the morphological characteristics suitable to shade (Weijschede et al., 2006). In a shade-induced phenotype, we can expect to see an increase in the plant's shoot height, shoot width or ground cover, and leaf sizes (Lee and Nam, 2022b), and this change is expected to help buffer the negative impact on growth due to the reduced amount of light received in shade.

Shoot fresh weight was highest at 18.51 g under the 35% shading level (Fig. 4A). Meanwhile, *V. rotunda* var. *subintegra* showed the highest shoot fresh weight when grown under the 55% shading level in the seedling process (Lee et al., 2020b). Root fresh weight was highest at 3.53 g under the 0% shading level (Fig. 4B). Shoot dry weight was highest at 2.58 g under the 35% shading level (Fig.

4C). Similarly, stem dry weight of Illicium anisatum was highest under the 35% shading level (Choi et al., 2012b). Root dry weight was highest at 0.87 g under the 0% shading level (Fig. 4D). Both root fresh weight and dry weight were highest under the 0% shading level, which was contrary to the result that root length was highest under the 60% shading level. This may be because factors such as carbohydrate content and thickness in the main root increased while the length of the side roots relatively decreased as there was a greater amount of light received. In addition, stimulation by water stress may have also caused an increase in the root fresh weight and dry weight. In previous studies, root fresh weight and dry weight of V. nakaiana and V. pyrethrina (Kwon et al., 2020) and V. rotunda var. subintegra were highest under the 0% shading level (Lee et al., 2020a), and root dry weight of Distvlium racemosum, Neolitsea aciculata, and Stauntonia hexaphylla was also highest under the 0% shading level, showing consistent results with this study (Choi et al., 2012b). In summary of the results of this study and previous studies, shading was better than direct sunlight to



Fig. 4. Shoot and root weight of *V. pusanensis* as affected by shading levels for 5 weeks. Vertical bars indicate the standard error, and asterisks (***) indicate significant at $p \leq .001$. Different lowercase letters indicate significant differences at $p \leq .05$ based on DMRT.

significantly increase shoot biomass. On the other hand, to significantly increase root biomass, it was relatively more beneficial to grow under direct sunlight. In addition, in the plant sizes and biomass analysis, the control (0% shading level) was placed outside the greenhouse and shading film due to limitations in the study, and thus it could have been affected by additional abiotic stress due to air temperature and soil temperature rise, humidity change, and damage caused by rainfall. In particular, as shown in the results, there may have been a negative effect on shoot growth, which is something to refer to.

CIELAB is the color space made in 1976 by the International Commission on Illumination (Commission Internationale de l'Eclairage; CIE) in France (Lee et al., 2022c). CIELAB was developed in 1976 to make up for the deficiencies of Hunter Lab developed in 1958, and unlike Hunter Lab indicated in L, a, and b, it is indicated in L^* , a^* , and b^* that include asterisk (Lee and Nam, 2022a). In this study, the leaf color analysis of V. pusanensis using CIELAB showed different results by shading level (Table 2). CIELAB L^* , which is the color space coordinate representing lightness, was highest at 46.90 under the 0% shading level. Meanwhile, CIELAB a^* that represents green-red opponent colors showed the same significance level under the 35-60% shading levels. CIELAB b^* that represents blue-vellow opponent colors was highest at 24.64 under the 0% shading level. b^* tended to be relatively high in the treatment where growth parameters such as shoot height,

shoot width, fresh weight, and dry weight were low. The results of previous studies showed some similarities with this study, proving that the growth rates of Orostachys japonica and O. boehmeri (Lee et al., 2022c), and Delosperma cooperi (Lee et al., 2022d) had a negative correlation with b^* . Meanwhile, L^* and b^* showed a positive correlation with each other (Kim et al., 2022; Lee et al., 2022c; 2022d). Similarly, this study also showed that L^* and b^* increased together under the 0% shading level, implying that it would be possible to use L^* and b^* as stress indicators for the leaf color of some plants including V. pusanensis. Royal Horticultural Society (RHS) values were 146B and 147B under the 0% shading level, showing that the color was relatively closer to yellow. On the other hand, the values were 137A and N137D under the 35-60% shading levels, showing that the leaf color was relatively green.

As a result, assuming that increased plant sizes and dark green leaf color contribute positively to the purchasing power of consumers when supplying *V. pusanensis* as an ornamental flower crop, it is recommended to grow *V. pusanensis* under the 35% shading level in order to significantly increase plant sizes and maintain dark green leaf color.

Analysis of chlorophyll content and fluorescence

Chlorophyll content (SPAD units) was highest in the order of 35 to 45% shading levels at 50.94 and 50.55, re-

Shading levels (%)		CIELAB values		DUC l ^Z	Converted color ^y
	L^*	<i>a</i> *	b^*	- KHS values	(color chip)
0	46.90 a ^x	-10.49 c	24.64 a	146B, 147B	
35	38.91 c	-8.51 a	16.98 c	137A, N137D	
45	39.73 c	-8.39 a	17.21 c	137A, N137D	
60	38.89 c	-8.35 a	17.43 c	137A, N137D	
75	41.74 b	-9.11 b	21.89 b	146A, 148A	
99	42.85 b	-9.52 b	21.02 b	146A, 148A	
Significance ^w	***	***	***		

Table 2. Leaf color reading values of CIELAB values (L^* , a^* , b^*), RHS values, and converted color of *Veronica pusanensis* as affected by shading levels for 5 weeks

^zRoyal Horticultural Society (RHS) colour charts edition V values.

^yColors converted using CIELAB L^* , a^* , and b^* values.

^xMeans separation within columns by Duncan's multiple range test at p < .05.

^{w***}: significant at p < .001.



Fig. 5. Cholophyll content (SPAD units) and photosynthetic responses of *V. pusanensis* as affected by shading levels for 5 weeks. Vertical bars indicate the standard error, and asterisks (***) indicate significant at $p \langle .001$. Different lowercase letters indicate significant differences at $p \langle .05$ based on DMRT.

spectively, showing the same results as shoot width and ground cover (Fig. 5A). This is consistent with the results of previous studies that, when growing Hylotelephium telephium (Nam et al., 2022), Hoya carnosa and Spathiphyllum wallisii (Lee et al., 2021a), and S. zokuriense (Lee et al., 2021b) grown under conditions of optimal shading levels and light intensity, an increase in chlorophyll content was observed concomitant with high shoot width. Meanwhile, V. rotunda var. subintegra showed relatively higher chlorophyll content under the 55-75% shading levels compared to the 0% shading level (Kwon et al., 2020). In summary of the results above, some plants including V. pusanensis seem to have homeostasis to maintain a constant total amount of carbon dioxide assimilation by maximizing plant sizes and chlorophyll density under low light intensity. F_v/F_m, which represents maximum quantum yield of photosystem II (PSII), was highest at 0.85 under the 35% shading level, and lowest at 0.81 under the 99% shading level (Fig. 5B). F_v/F_m , which is a parameter that represents a maximum quantum yield of PSII, is known to range from 0.78 to

0.84 in higher plants species that are not stressed (Björkman and Demmig, 1987; Butler and Kitajima, 1975; Genty et al., 1989; Govindjee, 1995; Govindjee, 2004; Paillotin, 1976; Yoo et al., 2012). F_v/F_m was slightly higher than the normal range under the 35% shading level, whereas it was within the normal range in other treatments. In particular, the maximum quantum yield was significantly low under the 99% shading level where the light intensity was extremely low, but it did not have a negative effect so much as to significantly deteriorate the function of the reaction center of PSII. As a result, plant sizes or biomass of V. pusanensis was relatively low under the 99% shading level, but shade tolerance was excellent, and growth rates are expected to be quickly recovered by moving the plant to an environment with high light intensity. Φ_{D0} that represents the probability that the absorbed photon will be dissipated was highest at 0.18 under the 99% shading level (Fig. 5C). ABS/RC that represents absorption flux per reaction center was highest at 1.99 under the 99% shading level (Fig. 5D). ABS/RC, one of chlorophyll fluorescence

parameters, represents the state of the reaction center that is inactivated as the number of reaction centers in reduction state increases in PSII (Spoustová et al., 2013). Meanwhile, DI₀/RC that represents dissipated energy flux per reaction center was highest at 0.38 under the 99% shading level, indicating that the energy use efficiency has decreased (Fig. 5E). PIABS is a performance index with an absorption basis (Srivastava et al., 1999). This index represents the degree of photosynthetic activity and is a set of light energy absorbing capacity, electron transfer efficiency, and electron fixation efficiency of PSII (Thach et al., 2007). PIABS represents the overall vitality of the photosynthetic apparatus (Strasser et al., 2000; Živčák et al., 2008), and it is used as an index that represents the soundness of plants (Oukarroum et al., 2007). PIABS was highest at 8.13 under the 35% shading level (Fig. 5F). This result was consistent with the results of chlorophyll content, shoot fresh weight, and dry weight, indicating that higher PIABS leads to higher chlorophyll density and more carbon dioxide assimilation.

In conclusion, as a result of comprehensively evaluating the plant sizes, biomass, leaf color, and photosynthetic responses analysis of *V. pusanensis* affected by shading levels, we discovered that shading levels significantly increase the size of *V. pusanensis* and maintain dark green leaf color. It is recommended to cultivate the plant under the 35% shading level to maintain the activity of PSII in the optimal state.

Conclusion

Veronica pusanensis is an endemic species belonging to the Plantaginaceae family and found in Busan, South Korea. *V. pusanensis* has high ornamental value and can be used as a flower crop, but it is currently an endangered species with its habitats being destroyed and reduced. Accordingly, this study analyzed the growth and photosynthetic responses of potted *V. pusanensis* according to shading levels for mass production of *V. pusanensis*. We selected polyethylene (PE) shading films as shading materials and designed 6 shading levels such as 0, 35, 45, 60, 75, and 99%, respectively. The results showed that shoot height, shoot width, ground cover, leaf length, leaf width, shoot fresh weight and dry weight, chlorophyll content (SPAD units), and chlorophyll fluorescence parameters F_v/F_m , PI_{ABS} were highest under the 35% shading level, indicating that it is relatively more desirable to grow *V*. *pusanensis* in shade culture compared to under direct sunlight. Meanwhile, root fresh weight and dry weight was highest under the 0% shading level, indicating that it is recommended to grow under direct sunlight to significantly increase root biomass when the purpose is to facilitate root-age when transplanting plants for habitat restoration. To cultivate *V. pusanensis* as an ornamental flower crop, it is recommended to grow under the 35% shading level to significantly increase plant sizes and maintain ensure the proper functioning of photosynthetic responses of photosystem II (PSII).

References

- Albach, D.C., H.M. Meudt, and B. Oxelman. 2005. Piecing together the "new" Plantaginaceae. America Journal of Botany 92(2):297-315. https://doi.org/10.3732/ajb.92.2. 297
- Albach, D.C., M.M. Martinez-Ortega, L. Delgado, H. Weiss-Schneeweiss, F. Özgökce, and M.A. Fischer. 2008. Chromosome numbers in Veroniceae (Plantaginaceae): review and several new counts. Annals of the Missouri Botanical Garden 95(4):543-566. https://doi.org/10.3417 /2006094
- Albach, D.C., M.M. Martinez-Ortega, M.A. Fischer, and M.W. Chase. 2004. A new classification of the tribe Veroniceae—problems and a possible solution. Taxon 53(2):429-452. https://doi.org/10.2307/4135620
- An, H.R., S.Y. Lee, P.M. Park, and Y.J. Kim. 2022. Effects of substrate water contents on post-shipping performance of *Phalaenopsis*. Horticultural Science and Technology 40(1):52-62. https://doi.org/10.7235/HORT.20220006
- Bae, E.J., E.J. Jin, J.H. Bae, K.S. Lee, and S.M. Choi. 2016. Growth and physiological characteristics of *Sarcandra glabra* and *Ardisia crenata* under different light intensity. Journal of Korean Society for People, Plants, and Environment 19(2):85-93. https://doi.org/10. 11628/ksppe.2016.19.2.85

Beara, I., J. Živković, M. Lesjak, J. Ristić, K. Šavikin,

Z. Maksimović, and T. Janković. 2015. Phenolic profile and anti-inflammatory activity of three *Veronica* species. Industrial Crops and Products 63:276-280. https://doi.org/ 10.1016/j.indcrop.2014.09.034

- Björkman, O. and B. Demmig. 1987. Photon yield of O₂ evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. Planta 170:489-504. https://doi.org/10.1007/BF00402983
- Bradshaw, A.D. 1965. Evolutionary significance of phenotypic plasticity in plants. Advances in Genetics 13:115-155. https://doi.org/10.1016/S0065-2660(08)60048-6
- Butler, W.L. and M. Kitajima. 1975. Fluorescence quenching in photosystem II of chloroplasts. Biochimica et Biophysica Acta 376(1):116-125. https://doi.org/10.1016/0005-2728 (75)90210-8
- Choi, D.S., T.K.L. Nguyen, and M.M. Oh. 2022. Growth and biochemical responses of kale to supplementary irradiation with different peak wavelengths of UV-A light-emitting diodes. Horticulture, Environment, and Biotechnology 63(1):65-76. https://doi.org/10.1007/s13 580-021-00377-4
- Choi, S.M., H.C. Shin, K.S. Lee, E.G. Bae, K.O. Choi, and K.Y. Huh. 2012a. Effects of shading rates on growth characteristics and photosynthesis in four broad-leaved evergreen trees. Journal of Korean Society for People, Plants, and Environment 15(2):99-106.
- Choi, S.M., H.C. Shin, K.Y. Huh, and H.J. Jung. 2012b. Seedling quality of broad-leaved evergreen trees with different shading levels. Journal of Korean Society for People, Plants, and Environment 15(4):265-271.
- DeWitt, T.J., A. Sih, and D.S. Wilson. 1998. Costs and limits of phenotypic plasticity. Trends in Ecology and Evolution 13(2):77-81. https://doi.org/10.1016/S0169-5 347(97)01274-3
- Fowler, J., and L. Chaffee. 2010. General information on propagation by stem cuttings. University of California Davis, California, USA.
- Genty, B., J.M. Briantais, and N.R. Baker. 1989. The relationship between the quantum yield of photosynthetic electron transport and quenching of chlorophyll fluorescence. Biochimica et Biophysica Acta (BBA) - General Subjects 990(1):87-92. https://doi.org/10.1016/S0304-41 65(89)80016-9
- Govindjee, G. 1995. Sixty-three years since Kautsky:

chlorophyll *a* fluorescence. Australian Journal of Plant Physiology 22(2):131-160.

- Govindjee, G. 2004. Chlorophyll *a* fluorescence: a bit of basics and history. in: G.C. Papageorgiou, Govindjee (Eds.). Chlorophyll *a* fluorescence: a signature of photosynthesis. Advances in Photosynthesis and Respiration (pp. 19:1-41). Dordrecht, The Netherlands: Springer.
- Grundemann, C., M. Garcia-Kaufer, B. Sauer, E. Stangenberg, M. Könczöl, I. Merfort, M. Zehl, and R. Huber. 2013. Traditionally used *Veronica officinalis* inhibits proinflammatory mediators via the NF-κB signalling pathway in a human lung cell line. Journal of Ethnopharmacology 145(1):118-126. https://doi.org/10.1016/j.jep.2012.10.039
- Harput, U.S., I. Saracoglu, M. Inoue, and Y. Ogihara. 2002. Anti-inflammatory and cytotoxic activities of five *Veronica* species. Biological and Pharmaceutical Bulletin 25(4): 483-486. https://doi.org/10.1248/bpb.25.483
- Harput, U.S., Y. Genc, N. Khan, and I. Saracoglu. 2011. Radical scavenging effects of different *Veronica* species. Records of Natural Products 5(2):100-107.
- Kasahara, M., T. Kagawa, K. Oikawa, N. Suetsugu, M. Miyao, and M. Wada. 2002. Chloroplast avoidance movement reduces photodamage in plants. Nature 420(6917): 829-832. https://doi.org/10.1038/nature01213
- Kim, H.J., J.H. Lee, J.H. Lee, M.S. Ko, and S.Y. Nam. 2022. A study on indoor cultivation of *Petrosedum rupestre* and *P. rupestre* cv. Angelina using commercial white T5 LEDs. Journal of Agricultural, Life and Environmental Sciences 34(3):354-367. https://doi.org/10.22 698/jales.20220035
- Kim, S.H., J.H. Kim, H.J. Oh, S.Y. Kim, and G.U. Suh. 2021. Vegetative propagation of *Veronica dahurica* and *Veronica pusanensis* by stem cuttings with auxins. Rhizosphere 17:100315. https://doi.org/10.1016/j.rhisp h.2021.100315
- Küpeli, E., U.S. Harput, M. Varel, E. Yesilada, and I. Saracoglu. 2005. Bioassay-guided isolation of iridoid glucosides with antinociceptive and anti-inflammatory activities from *Veronica anagallis-aquatica* L. Journal of Ethnopharmacology 102(2):170-176. https://doi.org/ 10.1016/j.jep.2005.05.042
- Kwon, H.H., H.J. Oh, J.H. Kim, and S.Y. Kim. 2021. Development of raising seedling technology for *Veronica pyrethrina* Nakai using plug trays. Journal of People,

Plants, and Environment 24(5):499-507. https://doi.org/ 10.11628/ksppe.2021.24.5.499

- Kwon, H.H., H.J. Oh, W. Cho, Y.H. Kwon, S.H. Yang, and S.Y. Kim. 2022. Growth and physiological responses of *Pseudolysimachion pusanensis* (Y. N. Lee) Y. N. Lee to NaCl treatment. Journal of People, Plants, and Environment 25(2):133-141. https://doi.org/10.11628/ksppe. 2022.25.2.133
- Kwon, Y.H., H.H. Kwon, M. Gil, M.J. Jeong, S.Y. Kim, and Y.H. Rhie. 2020. Growth of *Veronica nakaiana* and *Veronica pyrethrina* under different shading levels. Flower Research Journal 28(4):331-339. https://doi.org/ 10.11623/frj.2020.28.4.12
- Lechaudel, M., L. Urban, and J. Joas. 2010. Chlorophyll fluorescence, a nondestructive method to assess maturity of mango fruits (cv. 'Cogshall') without growth conditions bias. Journal of Agricultural and Food Chemistry 58(13):7532-7538. https://doi.org/10.1021/jf101216t
- Lee, J.H., H.B. Kim, and S.Y. Nam. 2022a. Evaluation of the growth and leaf color of indoor foliage plants under high temperature and continuous lighting conditions at different light intensity. Journal of Agricultural, Life and Environmental Sciences 34(1):26-36. https://doi.org/ 10.22698/jales.20220004
- Lee, J.H., H.J. Yoo, and S.Y. Nam. 2022b. Chlorophyll fluorescence response of indoor foliage plants as affected by light intensity levels under high temperature and continuous lighting conditions. Journal of Agriculture and Life Science 56(1):19-26 https://doi.org/10.14397/jals.2 022.56.1.19
- Lee, J.H., R.A.M. Cabahug, N.H. You, and S.Y. Nam. 2021a. Chlorophyll fluorescence and growth evaluation of ornamental foliage plants in response to light intensity levels under continuous lighting conditions. Flower Research Journal 29(3):153-164. https://doi.org/10.11623/frj.202 1.29.3.05
- Lee, J.H. and S.Y. Nam. 2022a. Analysis of growth and leaf color changes of *Sedum album* cv. Athoum according to the spectral power distribution of several white LEDs. Flower Research Journal 30(4):184-193. https://doi.org/ 10.11623/frj.2022.30.4.03
- Lee, J.H. and S.Y. Nam. 2022b. Effects of shading treatment on the growth and leaf color quality of potted *Phedimus takesimensis* cv. Atlantis. Journal of Agricultural, Life

and Environmental Sciences 34(3):413-424. https://doi. org/10.22698/jales.20220040

- Lee, J.H., S.Y. Soh, H.J. Kim, and S.Y. Nam. 2022c. Effects of LED light quality on the growth and leaf color of *Orostachys japonica* and *O. boehmeri*. Journal of Bio-Environment Control 31(2):104-113. https://doi.org/10.1 2791/KSBEC.2022.31.2.104
- Lee, J.H., S.Y. Soh, and S.Y. Nam. 2022d. Growth evaluation of potted *Delosperma cooperi* (Hook. f.) L. Bolus to shading levels, potting media, and fertilization rates. Flower Research Journal 30(1):1-9. https://doi.org/10.11 623/frj.2022.30.1.01
- Lee, J.H., Y.S. Lim, and S.Y. Nam. 2021b. Optimization of shading levels, potting media, and fertilization rates on the vegetative growth of *Sedum zokuriense* Nakai. Flower Research Journal 29(4):239-246. https://doi.org/ 10.11623/frj.2021.29.4.04
- Lee, S.I., S.H. Yeon, J.S. Cho, and C.H. Lee. 2020a. Growth assessment of the potted cultivation of *Veronica rotunda* var. *subintegra* (Nakai) T. Yamaz. Flower Research Journal 28(1):14-20. https://doi.org/10.11623/frj.2020.2 8.1.03
- Lee, S.I., S.H. Yeon, J.S. Cho, M.J. Jeong, and C.H. Lee. 2020b. Optimization of cultivation conditions on effective seedlings of *Veronica rotunda* var. *subintegra* (Nakai) T. Yamaz. Protected Horticulture and Plant Factory 29(2):181-188. https://doi.org/10.12791/KSBE C.2020.29.2.181
- Nam, J.W., J.H. Lee, J.G. Lee, S.Y. Hwang, and S.Y. Nam. 2022. Characteristics of growth and leaf color of *Hylotelephium telephium* cv. Lajos and *H. sieboldii* cv. Mediovariegatum as affected by shading levels. Flower Research Journal 30(4):172-183. https://doi.org/10.1162 3/frj.2022.30.4.02
- Oh, S.I. and A.K. Lee. 2022. Effect of rooting promoter treatments on cutting and growth of *Sedum takesimense*. Horticultural Science and Technology 40(1):12-20. https://doi.org/10.7235/HORT.20220002
- Olmstead, R.G. 2002. Whatever happened to Scrophulariaceae. Fremontia 30:13-22.
- Oukarroum, A., S.E. Madidi, G. Schansker, and R.J. Strasser. 2007. Probing the responses of barley cultivars (*Hordeum vulgare* L.) by chlorophyll a fluorescence OLKJIP under drought stress and re-watering. Environmental

and Experimental Botany 60(3):438-446. https://doi.org/ 10.1016/j.envexpbot.2007.01.002

- Paillotin, G. 1976. Movement of excitations in the photosynthetic domains of photosystem II. Journal of Theoretical Biology 58(1):237-252. https://doi.org/10.1016/0022-51 93(76)90150-8
- Photon Systems Instruments (PSI) 2023, April 17. FluorPen FP 110 PAR-FluorPen FP 110 Monitoring Pen MP 100. FluorPen & PAR FluorPen, Photon Systems Instruments website. Retrieved from https://handheld.psi.cz/docume nts/FluorPen_Monitoring_Manual_02_2021.pdf
- Salehi, B., M. Shivaprasad Shetty, N.V. Anil Kumar, J. Živković, D. Calina, A. Oana Docea, S. Emamzadeh-Yazdi, C. Sibel Kılıç, T. Goloshvili, S. Nicola, G. Pignata, F. Sharopov, M.D.M. Contreras, W.C. Cho, N. Natália, and J. Sharifi-Rad. 2019. *Veronica* plants—drifting from farm to traditional healing, food application, and phytopharmacology. Molecules 24(13):2454. https://doi.org/ 10.3390/molecules24132454
- Semchenko, M., M. Lepik, L. Götzenberger, and K. Zobel. 2012. Positive effect of shade on plant growth: amelioration of stress or active regulation of growth rate?. Journal of Ecology 100(2):459-466 https://doi.org/10.1111/j.136 5-2745.2011.01936.x
- Shim, D. and S.H. Jeon. 2022. Appropriate amount of nitrogen fertilizer for shading cultivation of tea tree seedlings. Horticultural Science and Technology 40(6):643-653. https://doi.org/10.7235/HORT.20220058
- Shin, H.T., M.H. Yi, J.S. Shin, B.C. Lee, and J.W. Yoon. 2012. Distribution of rare plants-Ulsan, Busan, Yangsan. Journal of Korean Nature 5(2):145-153. https://doi.org/ 10.7229/jkn.2012.5.2.145
- Song, S.J., M.J. Jeong, S.Y. Kim, and S.Y. Lee. 2020a. Phenotypic plasticity and ornamental quality of four Korean native *Veronica* taxa following different light intensity treatment. Flower Research Journal 28(3):123-131. https://doi.org/10.11623/frj.2020.28.3.03
- Song, S.J., M.J. Jeong, and S.Y. Lee. 2020b. Ecophysiology of growth and flowering of four Korean native *Veronica* taxa in response to the photoperiod and cold treatment. In III International Symposium on Germplasm of Ornamentals 1291 (pp. 205-214). https://doi.org/10.17660/Ac taHortic.2020.1291.26
- Song, S.J., U.S. Shin, H.J. Oh, S.Y. Kim, and S.Y. Lee.

2019. Seed germination responses and interspecific variations to different incubation temperatures in eight *Veronica* species native to Korea. Horticultural Science and Technology 37(1):20-31. https://doi.org/10.12972/kj hst.20190003

- Spoustová, P., H. Synková, R. Valcke, and N. Čeřovská. 2013. Chlorophyll *a* fluorescence as a tool for a study of the potato virus Y effects on photosynthesis of nontransgenic and transgenic Pssu-ipt tobacco. Photosynthetica 51(2):191-201. https://doi.org/10.1007/s11099-013-0023-4
- Srivastava, A., R.J. Strasser, and G. Govindjee. 1999. Greening of peas: parallel measurements of 77 K emission spectra, OJIP chlorophyll *a* fluorescence, period four oscillation of the initial fluorescence level, delayed light emission, and P700. Photosynthetica 36(13):365 https://doi.org/10.1023/A:1007199408689
- Stirbet, A. and G. Gavindjee. 2011. On the relation between the Kautsky effect (chlorophyll *a* fluorescence induction) and photosystem II: basics and applications of the OJIP fluorescence transient. Journal of Photochemistry and Photobiology B: Biology 104(1-2):236-257. https://doi. org/10.1016/j.jphotobiol.2010.12.010
- Strasser, R.J., A. Srivastava, and M. Tsimilli-Michael. 2000. The fluorescence transient as a tool to characterize and screen photosynthetic samples, pp. 445-483. In: M. Yunus, U. Pathre, and P. Mohanty (eds.). Probing photosynthesis: Mechanisms, regulation and adaptation (pp. 445-483). London, UK: Taylor & Francis.
- Sultan, S.E. 1987. Evolutionary implications of phenotypic plasticity in plants. Evolutionary Biology 21:127-178. https://doi.org/10.1007/978-1-4615-6986-2_7
- Thach, L.B., A. Shapcott, S. Schmidt, and C. Critchley. 2007. The OJIP fast fluorescence rise characterizes *Graptophyllum* species and their stress response. Photosynthesis Research 94:423-436. https://doi.org/10.1007/s11120-00 7-9207-8
- Weijschede, J., J. Martínkova, H. De Kroon, and H. Huber. 2006. Shade avoidance in *Trifolium repens*: costs and benefits of plasticity in petiole length and leaf size. New Phytologist 172(4):655-666. https://doi.org/10.1111/j.14 69-8137.2006.01885.x
- Yang, H.R., Y.J. Park, M.J. Kim, J.Y. Yeon, and W.S. Kim. 2022. Growth responses of Korean endemic *Hosta minor* under sub-optimal artificial lighting. Horticultural

Science and Technology 40(3):286-295. https://doi.org/ 10.7235/HORT.20220027

Yoo, S.Y., K.C. Eom, S.H. Park, and T.W. Kim. 2012. Possibility of drought stress indexing by chlorophyll fluorescence imaging technique in red pepper (*Capsicum annuum* L.). Korean Journal of Soil Science and Fertilizer 45(5):676-682. https://doi.org/10.7745/KJSSF.2012.45. 5.676

- Zettl, A. 2023, April 12. Converting Colors. Converting Colors website. Retrieved from https://convertingcolors.com
- Živčák, M., M. Brestič, K. Olšovská, and P. Slamka. 2008. Performance index as a sensitive indicator of water stress in *Triticum aestivum* L. Plant, Soil and Environment 54(4):133-139. https://doi.org/10.17221/392-PSE