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Effect of Wavelength of Light on Floral Bud
Differentiation of Chrysanthemum

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**Effect of Wavelength of Light on Floral Bud Differentiation of
Chrysanthemum**

(キク花芽分化に対する光波長影響の研究)

2013

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INTRODUCTION

Chrysanthemum (*Chrysanthemum morifolium* Ramat.), rose, carnation and gladiolus are the four main cut flowers in the world. Chrysanthemum is the most popular one of the main cut flowers with great production. It is used for decorating in funeral, Buddhism and for flower-arranging or ornament in our daily life. In Japan, it is ranked at the first position in the quantity of the shipment from the grower to the flower market, occupied more than 33% of total cut flowers quantity (data was from MAFF of Japan in 2011), and consumed about 20 billion plants per year.

Chrysanthemums originated in East Asia and have been cultivated for more than 2500 years. The chromosome is 18 (2n) in original wild type chrysanthemum, and there are also many polyploid species. The popular species (*chrysanthemum morifolium* Ramat.) is a polyploid (2n=54).

As a short day plant (SDP), wild type of chrysanthemum responds to short day length and floral bud differentiation appear from late August to early September, then flowers in autumn. For year round production, growers have been using techniques such as shading in summer or night lighting from autumn to spring to regulate its flowering. Lighting in the night will cause inhibition for flowering, and it is called night break (NB) effect. Growers are used to applying incandescent (INC) lamps as the NB light source for flowering regulation in SDPs production, for good inhibition effect of INC. From 1950's, INC lamps have been applied for NB treatment in chrysanthemum production because of its low price. However, INC lamps have very low electrical to light energy transformation efficiency and increase the cost in production. Recently, because the price of chrysanthemum is dropping down in the market in these years, it is necessary to reduce the production cost. On the other hand, in the situation of preventing global

warming and in order to save energy, the Japanese government has decided to halt the manufacture and selling of INC lamps. Therefore, it is necessary to find a new light source that can be used in agriculture as an alternative to INC lamps.

Recently, light sources like fluorescent lamp or LED lamp have been trying using for light cultural in chrysanthemum. Since the fluorescent lamp is easy to drop down the illumination and easy to be damaged, it has become expected that LED lamps, which are characterized by high phototransformation efficiency and low power consumption, will serve as an alternative to INC lamps. Because plants can perceive wide wavelength of light in nature and LED is a monochromatic lamp, the effect of LED lamp applying in the growth of chrysanthemum is unclear. Since there is scarcely applied instance in using LED lamp in light culture of chrysanthemum, it is necessary to verify its effect on chrysanthemum.

Although chrysanthemum is a SDP, since the breed improvement activities have been carried out during centuries, many mutants have been improved and some of them have lost their response to photoperiod then can flower regardless of the photoperiod. From 1988 (Kawada and Funakoshi), chrysanthemum is classified into the four flowering types, summer flowering, summer-to-autumn flowering, autumn flowering and winter flowering type. Table 0-1 shows the natural flowering season and development characteristic of chrysanthemum flowering types. Summer flowering type of chrysanthemum is very difficult to regulate its flowering and autumn flowering type of chrysanthemum is precious few suitable cultivars for shading cultivation. However, summer-to-autumn flowering type of chrysanthemum has longer critical day length than autumn flowering type. Therefore, it is possible for applying lighting in the night and also possible for applying shading cultivation in the long day condition to regulate its flowering. In chrysanthemum production, it is established the method of alternate application the cultivations of summer-to-autumn flowering type and autumn flowering type for

chrysanthemum year round production. Growers harvest summer-autumn flowering type of chrysanthemum from Jul. to Sep. and autumn flowering type of chrysanthemum from Nov. to Jun. in the next year. Now summer-to-autumn flowering type cultivar 'Iwa no hakusen' and autumn flowering type cultivar 'Jimba' or 'Seiko no makoto' are the popular cultivars that are applied in chrysanthemum year round production in Japan (Ohishi, K. 2011).

In NB regulating flowering, different flowering type chrysanthemum may have different response in NB light qualities. For the investigation of the NB effect being regulated by light, it is necessary to focus on the light qualities and the photoreceptors of plants.

The light environment, such as light intensity, light quality and photoperiod, plays an important role in plants and affects their photomorphogenic development, which includes seed germination, shoot architecture and flowering (Quail, 2002; Nagatani, 2004; Takano et al., 2005; Rockwell et al., 2006; Takano et al., 2009). Photoperiod has a marked influence on reproductive growth and regulates the flowering of plants. Flowering plants can be classified into three groups depending on their responses to the photoperiod: long-day plants (LDPs), SDPs, and day-neutral plants (Thomas and Vince-Prue, 1997). The Light signal is perceived by the photoreceptors of plants, such as phytochromes, cryptochromes and phototropins, and the day-length is measured by the circadian clock (Srikanth and Schmid, 2011). Phytochromes are mainly photoreceptors of red (R) and far-red (FR) light and are encoded by three genes *PHYA*–*PHYC* in rice (*Oryza sativa*), an SDP (Takano et al., 2005).

Borthwick (1952) showed that far red (FR) light irradiation, which followed by red (R) light, inhibited lettuce germination that induced by R light irradiation. This study suggested that the pigment that induced antergic activities that respond to R and FR light. Followed studies proved that phytochromes have the maximum absorption spectra at 668nm and 730nm, and have two forms of P_{fr} and P_r that response to R light and FR light respectively (Kelly and

Lagarias, 1985; Lagarias et al., 1987) (Fig. 0-1). P_{fr} or P_r form could change to each other by irradiating by R or FR light and kept a balance in photoreversibility.

Phytochrome A (phyA) mediates FR light (Mockler et al., 2003), while phytochrome B (phyB) mediates R light and inhibits flowering in rice (Ishikawa et al., 2009). Plants regulate their flowering by transducing the light signal into the circadian clock that controls the CONSTANS (CO) protein, a promoter activating the expression of *FLOWERING LOCUS T (FT)* gene, which encodes a florigen under inductive conditions (Yanovsky and Kay, 2002; Kobayashi et al. 1999; Kardailsky et al. 1999; Corbesier et al. 2007).

NB treatment inhibits flowering in rice, but the effect is reversed in the *phyB* mutant (Ishikawa et al., 2005). Thus, phytochromes are involved in the NB effect on flowering. When phytochromes perceive different wavelengths of light, plants reveal distinct physiological responses. Therefore light qualities of NB also have different effects on flowering (Kadman-Zahavi and Ephrat, 1972). Recently, it has become expected that LED lamps, which are characterized by high phototransformation efficiency and low power consumption, will serve as an alternative to INC lamps. Since LED is a monochromatic lamp, the different light wavelengths of LED lamps have different effects on the flowering of chrysanthemums.

Ishikura et al. (2009) reported that NB by emission of R light from an LED showed a similar effect with the use of an INC lamp on chrysanthemum flowering inhibition. Ohishi et al. (2010) showed that NB using a 630 nm LED had the highest NB effect. However, NB using LED lamps of other wavelength regions had rarely been attempted on chrysanthemums. There are very few reports on the effect of NB treatment by LED light of the same wavelength for different flowering types of chrysanthemum, for which the flowering season were different, such as summer-to-autumn flowering type and autumn flowering type chrysanthemums. Generally, FR light can reverse the effect of R light (Reid et al., 1967), but INC lamps which

contain both R and FR light, had marked NB effects on many flowering types of chrysanthemum. Therefore, it was necessary to verify the effect of R + FR light from LED lamps. Since there are many effects that are unclear, there is limited application of LED lamp in chrysanthemum production. In this study, we studied the NB effect of LED lamps on chrysanthemum flowering by different wavelengths.

Light intensities were also influenced on the effect which caused by light. In inhibitory flowering, low light intensities, such as lower than 30 lux of incandescent, could not inhibited the flowering of chrysanthemum (Yasuda, 1961). For growers applying LED in production, it is also necessary to understand that lowest light intensity which can inhibit floral bud differentiation. So that growers can manage to set up the height of LED lamp and make sure the lamp numbers. Therefore, the lowest of LED light intensities need to be verified.

As a cut-flower, chrysanthemum is needed to be grown to about 70 cm in shoot length at the time of harvesting. Shoot length elongation is very important parameter for chrysanthemum in production. Therefore, it is also necessary to verify the effect on plant height and shoot elongation by the LED NB light culture.

Light quality and photoperiod regulate many aspects of plant growth and development, including stem elongation and flowering. Shoot elongation is affected by light wavelength; blue and far-red (FR) light have a positive effect (Hirai et al., 2006; Shimizu et al., 2008; Arai and Ohishi, 2010), whereas red (R) light has an inhibitory effect (Reid et al., 2002). It is also to be thought that the phytochromes regulate the shoot elongation. It is necessary to investigate shoot elongation responses to different light qualities during NB treatment in chrysanthemums, as well as the relationship between different LED wavelengths and shoot elongation.

In this study, we studied several wavelengths of LED lamps and light intensities of NB effect on inhibitory flowering and effect of shoots and leaves growing. It was showed obvious

differences on floral bud differentiation and shoot elongation by different light qualities from the results. The results of this study would have some directions and references for the applying LED lamp in chrysanthemum production.

Table 0-1 Flowering season of different flowering groups of chrysanthemum and flowering related developmental characteristic^z

Cultivar flowering groups	Flowering season in the natural condition	Rosette ^y characteristic	Juvenile ^x characteristic	Photosensitivity		
				Critical day length ^w	Proper critical day length ^v	Weeks to flowering ^u
Summer flowering type	early flowering	very weak	very weak	-	-	-
	normal flowering	weak	weak	-	-	-
	late flowering	weak	weak	-	-	-
Summer-to-autumn flowering type	early flowering	-	strong	more than 17 h	13 ~ 14 h	7 ~ 8
	normal flowering	-	normal ~ strong	17 h	13 ~ 14 h	7 ~ 8
	late flowering	-	normal ~ strong	16 h	12 ~ 13 h	7 ~ 9
Autumn flowering type	early flowering	-	-	14 ~ 15 h	12 h	8 ~ 10
	normal flowering	-	-	13 h	12 h	9 ~ 10
	late flowering	-	-	12 h	-	11 ~ 12
Winter flowering type	Dec. and after Dec.	-	-	-	-	13 ~ 15

^z Modified from Newly Organized Agricultural Encyclopedia (Ishihara, et al., 2004).

^y In the low temperature of winter, the shoot elongation has been terminated and leaves are circularly arranged. '-' means no weak or strong characteristic.

^x After entering rosette stage, it is difficult to be induced floral bud differentiation.

^w Day length which is longer than critical day length will not flower. '-' means without critical day length.

^v Day length which is longer than critical day length will obviously delay flowering. '-' means not obviously.

^u The number of weeks to flowering in short day condition.

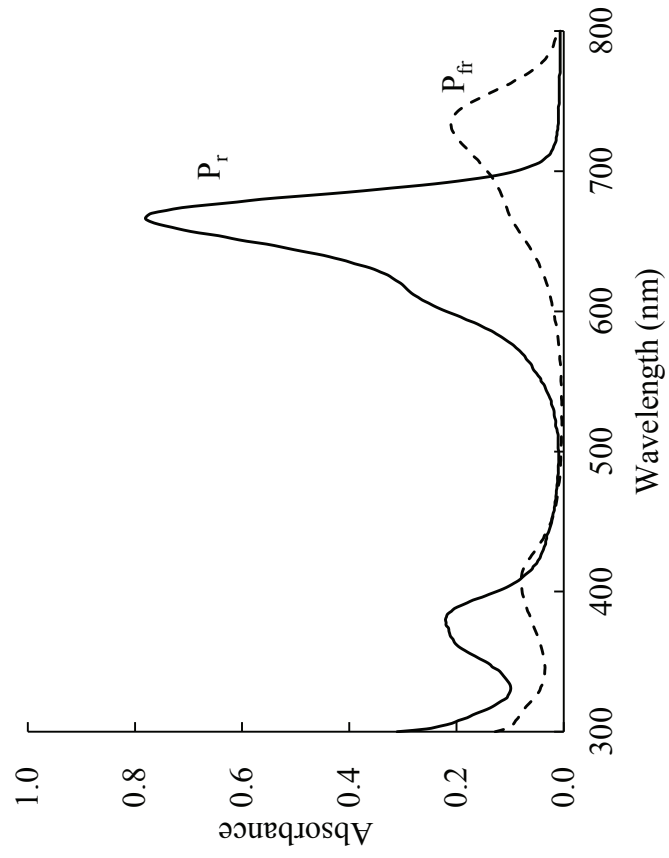


Fig.0-1 Pytochrome absorbance spectra from 300 nm to 800 nm.

Part 1 Night-break Effect of LED Light with Different Wavelengths on Floral Bud Differentiation of *Chrysanthemum morifolium* Ramat ‘Jimba’ and ‘Iwa no hakusen’

In order to take place of the INC lamps, it is necessary to verify the flowering inhibitory effect of LED lamps in chrysanthemum light culture. Although chrysanthemum is a SDP, it has several flowering types that flower from April to February in the next year. In Japan, autumn-flowering type and summer-to-autumn flowering type chrysanthemum are usually applied in chrysanthemum year around production. Therefore, in this study, we applied the cultivars of the two flowering type for the LED light quality NB experiment.

1. Materials and methods

1.1 Plant materials

Two cultivars of chrysanthemum were used as plant materials. One was the autumn-flowering type, ‘Jimba,’ which flowered in October under natural conditions in Japan. The other was the summer-to-autumn flowering type, ‘Iwa no hakusen,’ which flowered in June under natural condition of Japan. The two cultivars were representative chrysanthemum cultivars in Japan.

1.2 Light sources

Fluorescent lamps (40W, FLR40SW/M; NEC Inc., Japan; PPF of $144 \mu\text{mol m}^{-2}\cdot\text{s}^{-1}$ at the plant medium surface) were used as light source in the day time. NB light sources were incandescent lamp (abbr. to INC; 100V 40W; Ashahi electric Co. Ltd, Japan) and four kinds of LED lamps (12V; Shibasaki Inc., Japan) with peak emission wavelengths of 630 nm, 660 nm, 690 nm, and 735 nm. INC lamp was 67 mm in length and 35 mm in sphere diameter (Fig. 1-1A). LED lamp was a stick shape, which size was 21.8 cm \times 2.4 cm, with four tiny light-emitting

diodes in it. The LED pattern was contained 22 LED lamps that were installed uniformly on the ceiling of the growth chamber, so that the LED lighting could irradiate all the plants equably (Fig. 1-1B).

Light intensity was measured using LI-1800 (LI-COR Inc., Lincoln, NE, USA) light spectrophotometer at the surface of the plant medium, 40 cm bellowing the lamp. And eight positions of that surface were measured for calculating the average of the light intensity (Fig. 1-2). Light intensities of LED lamps were shown as photon flux per unit wavelength at the peak emission in the spectrum (Table 1-1) and photon flux density (PFD) was calculated (Table 1-2).

1.3 Methods

Shoot cuttings were reserved in the dark place at 5°C during the transportation. Before rooting, the bottoms of shoot cuttings were soaked in the water for about 1 h for absorbing enough water. OXYBERON (Bayercropscience Co., Ltd, Japan) was used as the plant growth regulator for rooting. The 10 cm long of shoot cuttings were rooted in a planting case (size of 48cm × 34cm × 10cm) which filled with the BM2 plant medium (Berger Peat Moss Ltd., Canada). The BM2 plant medium was about 9 cm deep in the planting case and root cuttings were rooted about 1 cm deep into the medium. The planting case was divided into two parts where shoot cuttings of the cultivars were planted. Every planting case was a treatment plot and contained 50 plants of each cultivar. After being rooted, the whole planting box was cover with a plastic film to keep the moisture for better rooting effect in the first week. From the second week, the plastic film was taken off.

Planting cases were put into growth chamber with a constant temperature of 23°C after rooted and plants had been growing inside for 6 weeks. Lighting in the day time had been given fluorescent lamp irradiation for 12 h (9:00~21:00) a day, a SD condition. NB treatments lighting had been given irradiation in the middle of the night time for 6 h (23:00~5:00) a day.

Six treatment plots had been given six different NB treatments for 6 weeks. Treatments of LED-630, LED-660, LED-690, LED-735, LED-630 + 735 and INC was irradiated by LED lamp which peak emissions at 630 nm, 660nm, 690nm, 735nm and 630nm + 735nm and by INC lamp respectively. The treatment LED-630 + 735 was composed of 22 LED lamps divided equally between 630 nm and 735 nm emissions. The control comprised plants that grew in a 12-h photoperiod (short day, SD) condition without NB treatment. Three growth chambers were used in this study. Each growth chamber was divided into two parts, an upper half and a lower half, which served as the NB treatment plots. NB lighting was blocked between the two halves of each growth chamber, and it did not interfere with the other half. Every treatment was repeated 3 times in this study.

For water and fertilizer management, the planting cases were put into the sink and plants was given 0.05% nutrient solution (N:P₂O₅:K₂O = 6.5:6:19, Hyponex Co., Ltd, Japan) every week. From the 3th week, because of the increasing transpiration, plants were given one more time of water irrigation a week.

1.3 Data collection and analysis

Five plants of each cultivar were sampled in every treatment each week. Their shoot length, leaf number, and floral bud differentiation (FBD) were measured or observed under the microscope. The floral differentiation level was classified into 7 stages (Fig. 1-3) and the index of FBD was calculated by the formula as below:

$$\text{Index of FBD} = \sum_{k=1}^n a_k \frac{1}{n \times 3.0} \quad (1)$$

(n = number of the samples; a_k = status of floral bud stage)

Experimental data were analyzed by ANOVA and Tukey's multiple comparison test ($P < 0.05$) or student's *t* test ($P < 0.05$). We assumed that the flowers differentiated when the index of FBD

reached 2.0; there were four samples with a status of 0.5 and one sample with a status of 1.0.

The P_{fr}/P_{total} of the phytochromes was estimated by using the formula of Sager et al. (1988):

$$P_{fr}/P_{total} = \left(\sum_{300nm}^{800nm} N_{\lambda} \delta_{r\lambda} \right) \left(\sum_{300nm}^{800nm} N_{\lambda} \delta_{r\lambda} + \sum_{300nm}^{800nm} N_{\lambda} \delta_{fr\lambda} \right)^{-1} \quad (2)$$

[P_{total} = total content of phytochrome molecule, including the P_{fr} and P_r form of phytochrome,

P_{fr} = content of P_{fr} (FR light-absorbing) form of phytochrome molecule,

N = the photon flux ($\text{mol m}^{-2} \cdot \text{s}^{-1}$, measured by LI-1800),

Λ = wavelength from 300 nm to 800 nm (interval of 2 nm),

δ_r = the photochemical cross section of P_r ($\text{m}^2 \text{mol}^{-1}$),

δ_{fr} = the photochemical cross section of P_{fr} ($\text{m}^2 \text{mol}^{-1}$)]

Data of red and far-red absorption state δ_r and δ_{fr} from 300 to 800 nm were also used from Sager et al. (1988).

2. Results

2.1 FBD in the two cultivars

In 'Jimba,' the FBD was observed in the SD treatment and the index of FBD reached 1.0 in the sixth week (Fig. 1-4). In LED-735, it was also observed FBD which began at the third week, and the index of FBD reached 0.98 in the sixth week. In other treatments, completely inhibited FBD was observed in the sixth week, and the index of FBD were 0.022, 0.047, 0, 0.025, 0.041 in treatments of LED-630, LED-660, LED-690, LED-660 + 735 and INC respectively (Fig. 1-4). Regardless of the different light sources, the FBD was completely inhibited in those treatments. The results in treatments of SD and LED-735 had no significant difference, and their results were significant higher than those of the other treatments.

In 'Iwa no hakusen,' similar results were observed as those in 'Jimba,' as FBD was also observed and the index of FBD reached 0.86 in the sixth week under the SD treatment (Fig.

1-4). However, the results differed from 'Jimba' in LED-630 and LED-660; FBD was observed and indices reached 0.72 and 0.76, respectively, in the sixth week. The FBD of 'Iwa no hakusen' was not inhibited in either of the treatments. In LED-735, it did not inhibit the FBD and a delayed effect was observed with an index of 0.27 in the sixth week. The index of FBD in the other treatments, LED-690, LED-660 + 735, and INC, were 0.09, 0.09, and 0.17 respectively, which showed a strongly inhibitory effect on FBD. The results in treatments of SD, LED-630 and LED-660 had no significant difference between any of them, but all of them had significant difference to those of the other treatments.

In cultivars difference, although 'Iwa no hakusen' was observed its FBD in SD condition, the index of FBD was little lower than that of 'Jimba' and the two results had significant difference. This might be the reason of juvenile characteristic of 'Iwa no hakusen.' In LED-735, 'Iwa no hakusen' was observed an inhibitory effect on FBD and the index of FBD had significant difference to that of 'Jimba.' In treatments of LED-630 and LED-660, the results had significant difference between the two cultivars. In LED-690 and INC, although the FBD was inhibited in both cultivars, it had significant difference between the two cultivars, because of the not completely inhibitory results in those treatments of 'Iwa no hakusen' (Table 1-4).

As a results, 'Jimba' and 'Iwa no hakusen' were different in FBD using LED-735 and LED-630 or LED-660 NB irradiation.

The light spectrum of LED-630 and LED-660 were distributed in the R light range of 600 nm ~700 nm. Generally, NB by R light has the strongest effect on inhibition of flowering in SDPs (Thomas and Vince-Prue, 1997). In 'Jimba,' both the LED-630 and LED-660 treatments inhibited its flowering perfectly. However, the R light treatments did not inhibit FBD in 'Iwa no hakusen,' suggesting that 'Iwa no hakusen' differed from 'Jimba' in R light NB response.

Although, total photon flux of LED-660 was the lowest and that of LED-630 was the

highest (Table 1-1), the results of both treatments were similar in ‘Jimba’ or in ‘Iwa no hakusen.’ Moreover, the results of both cultivars were different in the same total photon flux of LED-630 or LED-735. We considered that the difference of cultivars was contributed more than total photon flux of LEDs to the different results of both cultivars. Therefore, the results were less affected by total photon flux of LEDs in this study, but light qualities.

2.1.1 The relation between PFD of R light and index of FBD

Fig. 1-5A showed the relation between PFD of R light ratio and index of FBD. PFD of R light in every NB light sources was calculated from 600 nm to 699 nm. In ‘Jimba,’ in low PFD of treatments, such as LED-735 and LED-690 or LED-660 + 735, there were high index of FBD and low index of FBD results. In high PFD of treatment LED-630, there was low index of FBD result; while in low PFD of treatment LED-660, there was also low index of FBD result. In ‘Iwa no hakusen,’ in low PFD of treatments, such as LED-660 or LED-690, there were high index of FBD and low index of FBD results. While in high PFD of treatment LED-630, there was high index of FBD result.

From the results, index of FBD did not change according to the PFD of R light.

2.1.2 The relation between PFD of FR light and index of FBD

Fig. 1-5B showed the relation between PFD of FR light and index of FBD. PFD of FR light was calculated from 700 nm to 799 nm. In ‘Jimba,’ in low PFD of FR light treatments, such as LED-630 and LED-660, there were inhibitory results in FBD. In high PFD of FR light treatment INC, there was a low index of FBD result; while in LED-735, there was a high index of FBD result. In ‘Iwa no hakusen,’ in low PFD of FR light treatments of LED-630 and LED-660, there were high index of FBD results. While in high PFD of INC, there was a similar

result to the low PFD treatments of LED-660 + 735 and LED-690.

From the results, index of FBD did not change according to the PFD of FR light.

2.1.3 The relation between R/FR light ratio and index of FBD

Fig. 1-6A showed the relation between R/FR light ratio and index of FBD. In ‘Jimba,’ in low R/FR light ratio treatments, except for LED-735, there was showed inhibitory results with low index of FBD. While in high R/FR light ratio of LED-630, there was also a low index of FBD. In ‘Iwa no hakusen,’ in low R/FR light ratio treatments, such as INC, there was a low index of FBD. In high R/FR light ratio of LED-630, there was a high index of FBD.

In these results, R/FR light ratio was not related to the index of FBD in ‘Jimba.’ In high R/FR light ratio could not inhibit FBD in ‘Iwa no hakusen.’

2.1.4 The relation between internode length and index of FBD

Fig. 1-6B showed the relation between internode length and index of FBD. The internode length was in the scale from 1.0 cm to 1.56 cm in treatments. In the similar internode length scale of 1.2 cm to 1.4 cm, there were high index of FBD result in LED-735 and low index of FBD results in other treatments in ‘Jimba.’ Similarly, in ‘Iwa no hakusen,’ in the similar internode length scale of 1.0cm to 1.2cm, there were high index of FBD results in treatments of LED-660 and LED-690.

In these results, internode length was not related to index of FBD.

2.1.5 The relation between P_{fr}/P_{total} ratio and index of FBD

Fig. 1-7 showed the relation between P_{fr}/P_{total} ratio and index of FBD. In ‘Jimba,’ P_{fr}/P_{total} ratio at 0.47 was showed an inhibitory result in LED-660 + 735. When P_{fr}/P_{total} was higher than 0.47, there were inhibitory results in other treatments. When P_{fr}/P_{total} was lower than 0.47, there was high index of FBD result in LED-735. However, in ‘Iwa no hakusen,’ when P_{fr}/P_{total} ratio was higher than 0.47, there were low index of FBD results in treatments of LED-660 + 735,

LED-690 and INC, and also there were high index of FBD results in treatments of LED-630 and LED-660.

These results showed that it could inhibit the FBD when P_{fr}/P_{total} ratio was higher than 0.47 in 'Jimba'. But P_{fr}/P_{total} ratio was not related to index of FBD in 'Iwa no hakusen.'

2.2 Plant height and leaf number in the two cultivars

2.2.1 Plant height

In 'Jimba,' plant heights six weeks after NB treatment in SD, LED-630, LED-660, LED-690, LED-735, LED-660 + 735, and INC were 23.5, 23.1, 24.9, 21.9, 36.3, 31.1, and 36.9 cm respectively (Fig. 1-8). The plant heights in LED-735, LED-660 + 735 and INC were significantly longer than other treatments.

In 'Iwa no hakusen,' plant heights six weeks after NB treatment in SD, LED-630, LED-660, LED-690, LED-735, LED-660 + 735, and INC were 29.0, 25.9, 25.8, 26.7, 35.8, 34.5, and 36.8 cm respectively (Fig. 1-8). Similar to 'Jimba,' plant heights in treatments of LED-735, LED-660 + 735 and INC were significant longer than those in other treatments.

In cultivars difference, plant height of 'Iwa no hakusen' was significant longer than 'Jimba' in SD, LED-630, LED-690 and LED-660 + 735, and had no difference in LED-660, LED-735 and INC (Table 1-5).

2.2.2 Leaf number

In 'Jimba,' leaf numbers in LED-690 and SD were 20.9 and 24.9 in treatments at the sixth week. And it was the least and the most result in all the treatments. Leaf numbers were 23.2, 25.1, 27.5, 24.5 and 24.7 in treatments of LED-630, LED-660, LED-735, LED-660 + 735 and INC respectively (Fig. 1-9).

In 'Iwa no hakusen,' leaf numbers in LED-690 and LED-735 were 24.9 and 29.5 in

treatments at the sixth week. And it was the least and the most result in all the treatments. Leaf numbers were 28.5, 26.2, 27.5, 27.5, and 27.2 in other treatments of SD, LED-630, LED-660, LED-660 + 735, and INC respectively (Fig. 1-9). There was no relationship between leaf number and light quality of NB treatments.

In cultivars difference, except for LED-735, there was significant difference in leaf number in all the treatments of the two cultivars (Table 1-5). It was suggested that 'Iwa no hakusen' had more leaf number than 'Jimba.'

2.2.3 Internode length

Internode length was calculated by using the data of plant height and leaf number.

In 'Jimba,' internode length was from 1.0 to 1.5 cm in treatments at the six week (Fig. 1-10). Although the data was different in the 0 week, it was regulated by light quality treatments for sixth weeks. The shortest internode length was in treatment SD, LED-630 and LED-660, and had no significant difference to that of LED-690. Treatment of LED-735 and LED-660 + 735 was 1.3 cm and significant longer than SD, LED-630, LED-660 and LED-690. The longest internode length was in treatment INC and had significant difference to those in all the other treatments.

In 'Iwa no hakusen,' there was no difference in the 0 week, and it was regulated by light quality treatments for sixth weeks. Internode length was from 1.0 to 1.4 cm at the sixth week in treatments (Fig. 1-10). The shortest internode length was in treatments of LED-660 and LED-690, and had no difference to that in treatments of SD and LED-690. The longest internode length was in treatment INC, and it had no difference to that in treatments of LED-735 and LED-660 + 735. The internode length in treatments of INC, LED-735 and LED-660 + 735 had significant difference to that in other treatments. The results were showed a similar tendency to that in 'Jimba.'

In cultivars difference, except for INC, there was no difference in internode length in all the treatments of the two cultivars (Table 1-5).

2.2.4 Shoot elongation

Shoot elongation was calculated the difference of the plant height in the sixth week and in the 0 week. In 'Jimba,' shoot elongation in treatments of SD, LED-630, LED-660 and LED-690 were 13.63, 13.24, 14.62, 12.07 cm respectively, and there was no difference among them (Fig. 1-11). Shoot length in treatments of LED-735, LED-660 + 735, and INC were 26.59, 21.25, and 26.63cm respectively, and was significant longer than that in the other treatments from the fourth week to the sixth week. There was no difference in shoot elongation between the treatments of INC and LED-735, and both of them were significant longer than that in LED-660 + 735.

In 'Iwa no hakusen,' shoot elongation in treatments of SD, LED-630, LED-660, and LED-690 were 18.82, 16.21, 15.41, and 17.01 cm respectively, and there was no difference between any of them (Fig. 1-11). Shoot elongation in treatments of LED-735, LED-660 + 735, and INC was 25.92, 24.37, and 26.84 cm respectively, and was significant longer than those in the other treatments in the sixth week. The longest shoot elongation was in treatment INC and had no difference to that in LED-735. Shoot elongation in both of the treatments was significant longer than that in LED-660 + 735. The results were showed a similar tendency to that in 'Jimba.'

In cultivars difference, there was significant difference in treatments of SD, LED-630, LED-690, and LED-660 + 735 and no difference in treatments of LED-660, LED-735, and INC in elongation (Table 1-3).

3. Discussion

3.1 Inhibitory effect on FBD by different light quality of NB treatments

Although the flowering season of the two cultivars was different, they flowered in an inductive SD condition. The results were suggested that 'Iwa no hakusen' was basically a SDP and also could also flower in autumn. It was also suggested that different wavelength of NB had different inhibitory effect on flowering and the results were also different in cultivars. In 'Jimba,' except for LED lamp peak emission at 735 nm, all the wavelength NB treatments had good effect on inhibition flowering. In 'Iwa no hakusen,' INC had good NB effect and LED lamp peak emission at 690 nm or 660 nm + 735 nm had similar inhibitory effect to INC. LED lamp peak emission at 630 nm and 660 nm could not inhibited FBD and had a similar effect as SD treatment. LED lamp wavelength at 735 nm was showed a delay effect on FBD, different to that in 'Jimba.'

3.1.1 R light LED NB effect on FBD

The light spectrum of LED-630 and LED-660 were distributed in the R light range of 600 nm ~700 nm. NB by R light has the strongest effect on inhibition of flowering in SDPs (Thomas et al., 1997). And R light peak emission at 630 nm of LED lamp has the strongest effect on flowering inhibition of chrysanthemum (Ohishi et al., 2010).

In 'Jimba,' both the two treatments inhibited its flowering perfectly. Except for LED-735, other NB treatment of lamps were also contained R light. So that, PFD of R light might have relate to index of FBD. Fig. 1-5A was showed the relation between PFD of R light in every treatment and index of FBD. In low R light PFD of treatment LED-690 and LED-735, there were different results in index of FBD. And in high and low R light PFD treatments, such as LED-630 and LED-690, there were similar results of index of FBD.

Because R light treatments could not inhibit FBD of ‘Iwa no hakusen,’ it suggested that ‘Iwa no hakusen’ was different to ‘Jimba’ in R light NB response. In Fig. 1-5A, in similar low R light PFD of treatments of LED-690 and LED-660, there were different results in index of FBD. And in different R light of PFD treatments of LED-630 and LED-660, there were similar results in index of FBD.

It suggested that PFD of R light had no relation to index of FBD.

3.1.2 FR light LED NB effect on FBD

The light spectrum of LED-735 was distributed in the FR light range, over 700 nm. LED-735 was used for FR light treatment. NB by FR light irradiation induced flowering (Ishiguri and Oda, 1972). And because of floral bud was observed in LED-735 in ‘Jimba,’ it was supposed that PFD of FR light might relate to the index of FBD in ‘Jimba.’ In Fig. 1-5B, PFD of FR light in INC was higher than that in LED-735, however the FBD was inhibited in INC, and it was observed in LED-735. PFD of FR light in other treatments was from 5 to 0 $\mu\text{mol m}^{-2} \cdot \text{s}^{-1}$, but the results were all similar to low index of FBD. It was suggested that PFD of FR light was not related to index of FBD in ‘Jimba.’

Because of the FBD in LED-735, acting similarly to that in SD condition, it suggested that it had no NB effect on ‘Jimba’ in the NB treatment of FR light. In another study, we carried out a 9-h day elongation using 735 nm LED lamp irradiation in ‘Jimba’ after 15 h of fluorescent lamp irradiation. After 6 weeks of treatment, inhibition in flowering was observed, which was similar to that of growing in long day (LD) condition (data not shown). It has been suggested that FR light irradiation at night could not reverse the inhibition of flowering in ‘Jimba’ grown in the LD condition. In SDP rice, NB by FR light failed to suppress *Hd3a* expression and showed no difference to the flowering in the SD treatment. This result suggests that FR

light NB had no effect on the flowering time (Ishikawa et al., 2009). Therefore, the NB by FR light treatment had no NB effect on ‘Jimba,’ and FR light treatment showed similar effects as the dark treatment.

On the other hand, the FR light NB showed a serious delay in FBD in ‘Iwa no hakusen.’ This suggests that FR light of 735 nm LED NB had an inhibitory effect on the flowering of ‘Iwa no hakusen’ (Fig. 1-5B). Therefore, it was assumed that photon flux density (PFD) of FR light would relate to the index of FBD in ‘Iwa no hakusen.’ In low PFD of FR treatments LED-690 and LED-630, there were different results in index of FBD. While in high PFD of FR treatment INC and LED-735, there were similar results of lower index of FBD. Fig. 1-5B indicates that no relation was found between PFD of FR light and index of FBD of ‘Iwa no hakusen.’ The floral bud inhibitory effect of FR light by PFD could not be explained in this way.

3.1.3 The relation between R/FR light ratio and index of FBD

Generally, R/FR ratio was used as a parameter of plant photomorphogenesis. Treatments of LED-690, LED-660 + 735 and INC were contained R and FR light. In chrysanthemum production, INC lighting contained wide wavelength and was a good light source for inhibition of flowering. LED lamps of 690 nm and 660 nm + 735 nm had a similar inhibitory effect to INC on both of the two cultivars (Fig. 1-4A, B).

In Fig. 1-6, in high and low R/FR ratio treatments of LED-630 and LED-690, there were similar results of low index of FBD. While in similar R/FR ratio treatments of LED-690 and LED-735, there were different results in index of FBD in ‘Jimba.’ In ‘Iwa no hakusen,’ in high R/FR ratio of LED-630 and low R/FR ratio treatments of LED-660, there were similar results of high index of FBD. There was no relationship between R/FR ratio and FBD in both cultivars

Although NB by FR light can overcome the R light NB effect on inhibition of flowering in

R and FR light alternating irradiation experiment (Reid et al., 1967), FR light could not totally cancel the R light NB effect when R and FR irradiated at the same time, because of the R light continuing irradiation until the end of NB treatment. Therefore, the R + FR NB inhibition of FBD of 'Jimba' was because of the R light inhibitory effect. In 'Iwa no hakusen,' although the FBD was observed in treatment LED-660, treatment LED-660 + 735 inhibited it. It suggested that NB by R + FR could inhibition FBD in 'Iwa no hakusen,' but the reason was unclear yet. As a result, combination R + FR light of NB had good inhibitory effect on both of chrysanthemum cultivars.

3.1.4 The relation between internode length and index of FBD

The shoot length was elongated before flowering in many sun plants. Chrysanthemum was also a kind of plant which was elongated its shoot length before flowering. Therefore, it was supposed that internode length had related to the index of FBD. But in Fig. 1-6B, there was no relation between internode length and index of FBD in both cultivars. Therefore, shoot elongation had no relation to index of FBD in this study.

3.1.5 The relation between P_{fr}/P_{total} percentage and index of FBD

Light regulated flowering by adjusting the activity of phytochromes. There were two light absorbing forms of phytochromes, biologically active P_{fr} and inactive P_r , which have two maximum absorption spectra at 660 nm and 735 nm respectively. Therefore, it was supposed that P_{fr}/P_{total} percentage had related to the index of FBD. In Fig. 1-7, in high P_{fr}/P_{total} percentage treatments, there were showed low index of FBD. And in low P_{fr}/P_{total} percentage treatment LED-735, there was showed high index of FBD in 'Jimba.' While in 'Iwa no hakusen,' in high P_{fr}/P_{total} percentage treatments of LED-630 and LED-660, there were results of high index of

FBD. Although index of FBD was inhibited when P_{fr}/P_{total} percentage was higher than 0.47 in ‘Jimba,’ it was different in ‘Iwa no hakusen.’

It was suggested that P_{fr}/P_{total} percentage could not explain the index of FBD in both cultivars.

3.1.6 PhyA and phyB effect on FBD

Phytochrome A (phyA) and Phytochrome B (phyB) were the different function of phytochromes that mainly mediated FR and R light.

PhyB was involved in mediating R light NB inhibiting flowering. In rice, phyB mutants had lost the NB effect and flowering was similar to that in the wild type in SD treatment (Ishikawa et al., 2005). PhyB can be induced physiologically active P_{fr} form in continued R light because of photoreversibility. In the night time, the P_{fr} form of PhyB had been photoconverted to the inactive P_r form gradually; and since there was no R light, it was called dark reversion (Medzihradzky et al., 2013). As the P_{fr} form is the physiologically active form of phyB, the P_{fr} form of phyB must be active in the ‘Jimba’ exposed to the R light NB, which photoconverted the P_r form of phyB to the P_{fr} form at night time and thus inhibited FBD.

PhyA also had been photoconverted to P_{fr} form (max 85%) in the R light. Those P_{fr} forms of phyA were imported to the nucleus and repressed *PHYA* mRNA transcription (Quail, 1994). Therefore, the content of PhyA protein declined in the cytoplasm under R light. In the dark, after the P_{fr} dropped below the critical level, high transcriptional activity resumed, and phyA was synthesized in P_r form that accumulated in the cytoplasm. Therefore, in the SD treatment, phyA protein decreased during the daytime and increased at night in *Arabidopsis* (Mockler et al., 2003). Similarly, under FR light, only approximately 1% of phyA was converted to the P_{fr} form and phyA accumulated in the cytoplasm, with approximately 99% in the P_r form (Quail, 1994). Therefore, similar changes took place in phyA between FR light of NB and in the night

time of SD in ‘Jimba.’ The FBD results were similar to those in LED-735 and SD treatments of ‘Jimba.’

In chrysanthemum breeding, breeders have continually selected the mutants from the original chrysanthemum and improved them. Some mutations have lost their response to photoperiod and can flower regardless of the photoperiod. ‘Iwa no hakusen’ is the one of improved chrysanthemum cultivars from a mutant that can flower in June. The mutation in ‘Iwa no hakusen’ might be the factor responsible for the difference between ‘Iwa no hakusen’ and ‘Jimba’ in terms of the light quality NB response to flowering.

3.2 Effect on plant growth by different light quality of NB treatments

The plant heights at LED-735, LED-660 + 735 and INC were significantly longer than that in other treatments (Fig. 1-8), and the elongation of these shoots at six weeks was about 25 cm, although those at other treatments were around 13 cm in ‘Jimba’ and 15 cm in ‘Iwa no hakusen’ (Fig. 1-11). Although there was no significant difference in leaf number among the treatments except for LED-690 and LED-735 (Fig. 1-9), the internode lengths calculated by plant height and leaf number in LED-735, LED-660 + 735 and in INC were significantly longer than those in other treatments (Fig. 1-10)

From the results, shoot elongation under irradiation by LED-735, LED-660 + 735 and INC enhanced internode elongation but not node number, as there was no difference in leaf number but there was a significant difference in plant height. Internode elongation is controlled by endogenous gibberellins (GA) (Grete et al., 1998), and Reid et al. (2002) reported that GA biosynthesis is inhibited by R light, and this response differs between in species and developmental stages. In our study, internode length and shoot elongation under the LED-630 and LED-660 treatments showed no difference from that under the SD treatment (Fig. 1-10, Fig.

1-11); therefore, we conclude that R light did not have an inhibitory effect on GA biosynthesis in chrysanthemums in this study.

As shown in Fig. 1-11, the LED-735, LED-660 + 735, and INC treatments promoted internode elongation and had the wavelength 735 nm (FR light) in common. Rajapakse et al. (1993) reported that FR light enhanced stem elongation in chrysanthemums, and Hisamatsu et al. (2005) suggested that in *Arabidopsis* shoot elongation is caused by GA biosynthesis induced by FR light. Therefore, we suggest that the internode elongation in 'Jimba' and 'Iwa no hakusen' in the present study was caused by FR-induced GA biosynthesis.

Regarding varietal differences, although the 'Iwa no hakusen' plants were taller and had more leaves than the 'Jimba' plants (Table 1-5), the effect of NB treatment on both varieties was similar. Interestingly, Fukui et al. (2010) found that FBD differed between 'Jimba' and 'Iwa no hakusen,' depending on the wavelength of light.

3.2.1 The relation between internode length and PFD of FR light

INC and LED-660 + 735 contained R light and FR light (Fig. 1-2). As mentioned above, since R light had no contribution to internode elongation, it was considered that the effect in these treatments was demonstrated by GA biosynthesis induced by FR light only, as well as LED-735. PFD at the range of 700 nm to 799 nm had significantly close relationship with internode length (Fig. 1-12A, B). Therefore, internode elongation may be affected by intensity of only FR light irradiation.

3.2.2 The relation between internode length and R/FR ratio

Previous studies on the relationship between shoot elongation and light have demonstrated that the R/FR ratio is related to shoot elongation, and a low R/FR ratio promotes shoot

elongation (Pierik et al., 2004; Franklin and Whitelam, 2005; Lorrain et al., 2008). In the present study, we found no significant relationship between the R/FR ratio and internode length (Fig. 1-13A, B).

3.2.3 The relation between internode length and phytochromes

Phytochromes play an important role in plant photomorphogenic responses (Whitelam and Halliday, 2007) and the P_{fr} and P_r forms of phytochrome are photoreversible when responding to R and FR light (Borthwick et al., 1952). The ratio of P_{fr}/P_{total} can be estimated from the spectral photon flux distribution of irradiation, and phytochrome photoconversion (Sager et al., 1988; Hanyu et al., 1996). It was showed the relationship between internode length and P_{fr}/P_{total} or P_r/P_{total} in different NB treatments (Fig. 1-14A, B). In ‘Jimba,’ there was no relationship between internode length and P_{fr} or P_{fr}/P_{total} , and neither P_r nor P_{fr}/P_{total} promoted internode elongation.

Recently studies, have found that phyA and phyB have different functions. Reid et al. (2002) and Foo et al. (2006) reported that phyA-mediated FR light responses regulate GA synthesis in plants, and consequently, affect stem elongation. Moreover, the transduction of light signals via phytochromes (phyA or phyB) regulates the CONSTANS (CO) protein, which may activate the *FLOWERING LOCUS T (FT)* gene, which controls flowering (Kardailsky et al., 1999; Kobayashi et al., 1999; Corbesier et al., 2007). ‘Iwa no hakusen’ has a normal signal transduction of phytochromes with respect to stem elongation, but this function varies with respect to floral bud differentiation. On the basis of the results of the present study, we are of the opinion that phyA was induced by FR light, that in turn enhanced GA biosynthesis, resulting in greater shoot elongation.

Total conclusion

R light NB showed marked flowering inhibition effect on the autumn flowering type of chrysanthemum 'Jimba,' but not in the summer-to-autumn flowering type of chrysanthemum 'Iwa no hakusen.' This suggests that a treatment of only R light could not inhibit flowering in all the popular chrysanthemum cultivars. A combination of R and FR light NB of LED lamps (660 nm + 735 nm) showed a better inhibitory effect than R light or FR light irradiation alone on both the flowering types chrysanthemum cultivars. Thus, a combination R and FR light of LED lamps was a potential light source for inhibiting flowering in chrysanthemum.

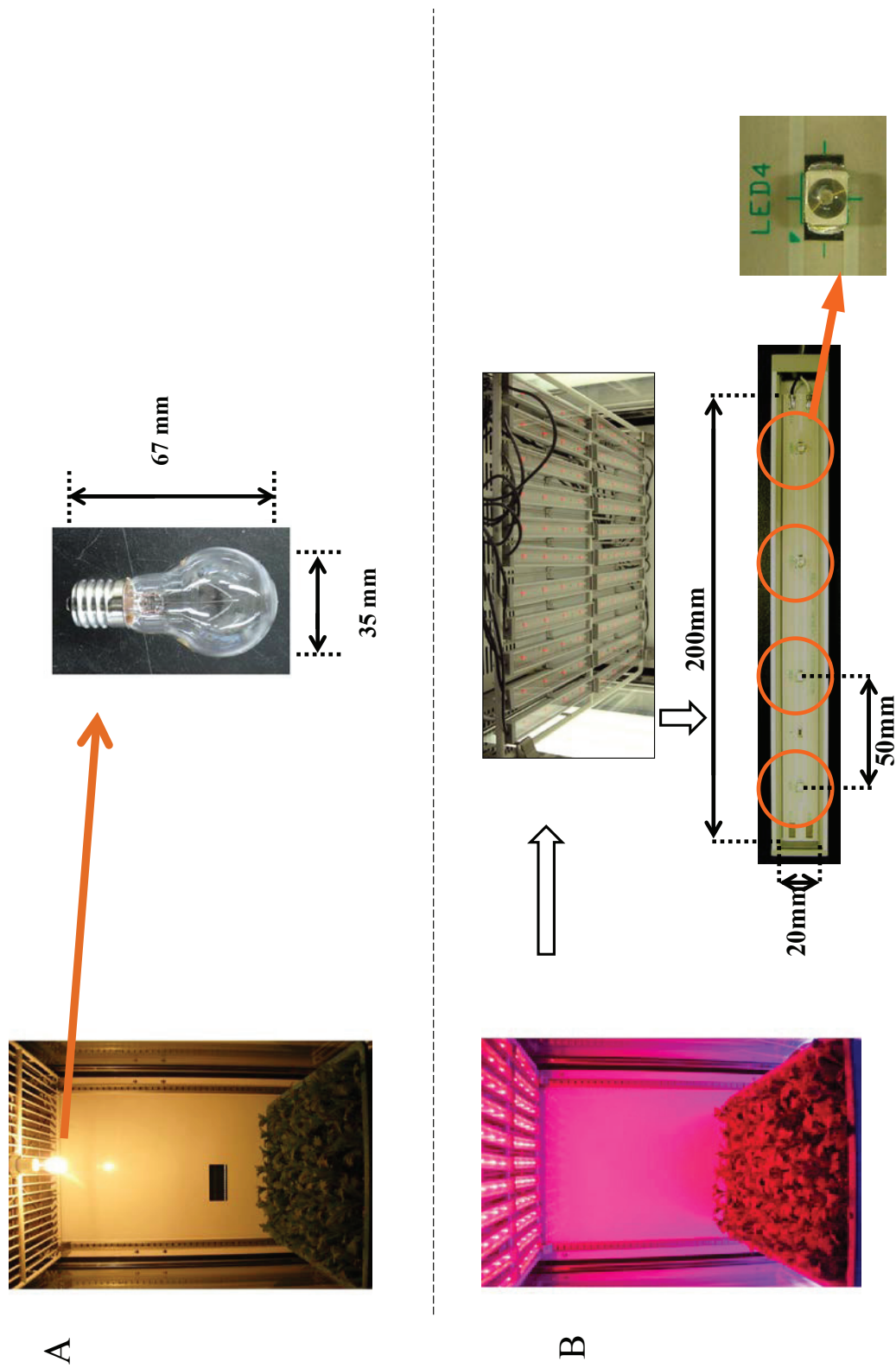


Fig. 1-1 Chrysanthemum plants had been growing in the plant growth chamber for 6 week. The INC lamp was install in the central of the frame which was hanging 40 cm over the plant medium surface (A). NB treatment of LED pattern was contained 22 LED lamps that were installed at the same height of the INC frame (B).

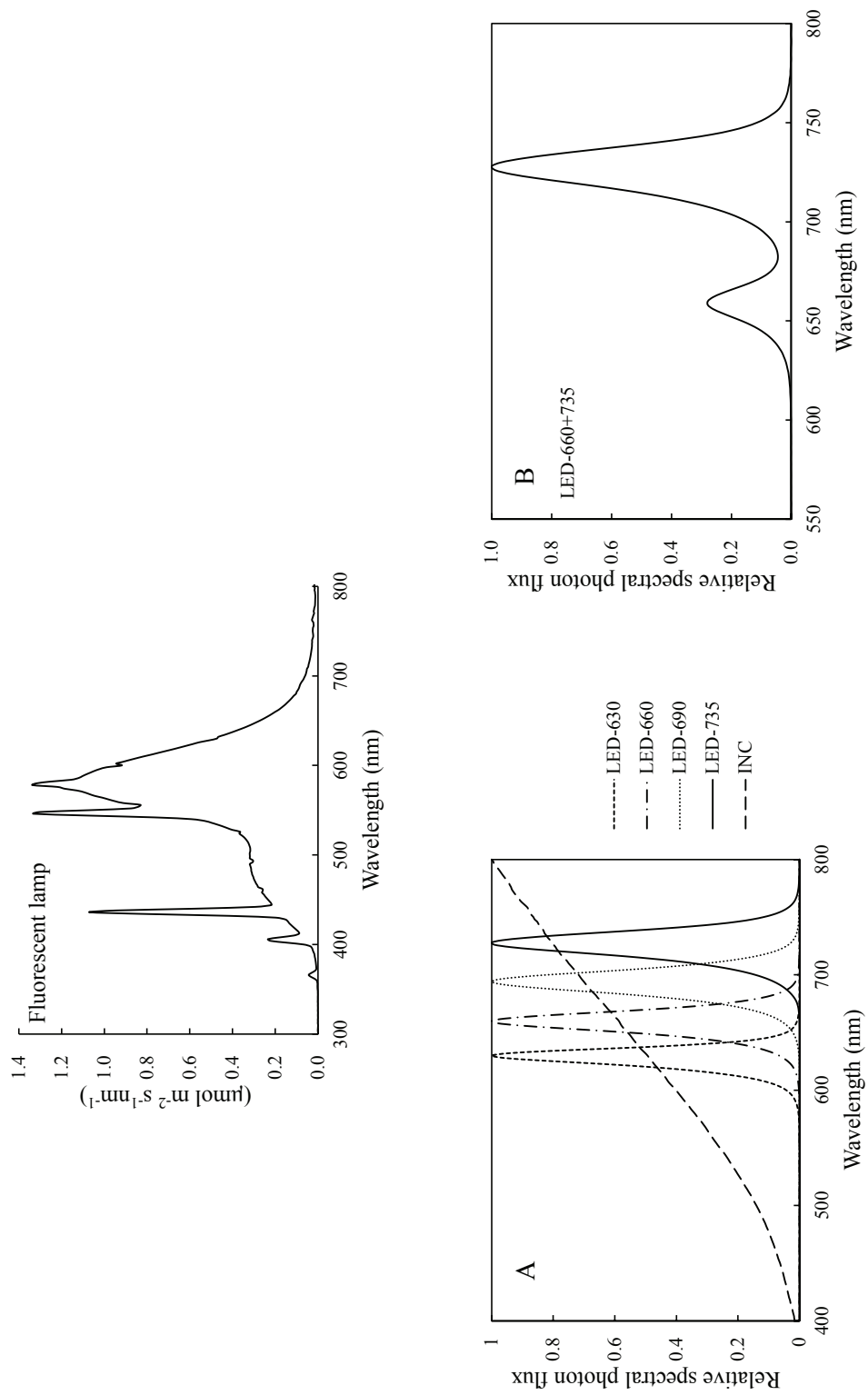


Fig. 1-2 The light spectral of fluorescent lamp and NB treatment of lamps. Relative spectral photon flux distributions of LED-630, LED-660, LED-690, LED-735, INC (A) and LED-660 + 735 (B). Spectral photon flux distribution values were calculated relative to the maximum value (set to 1.0).

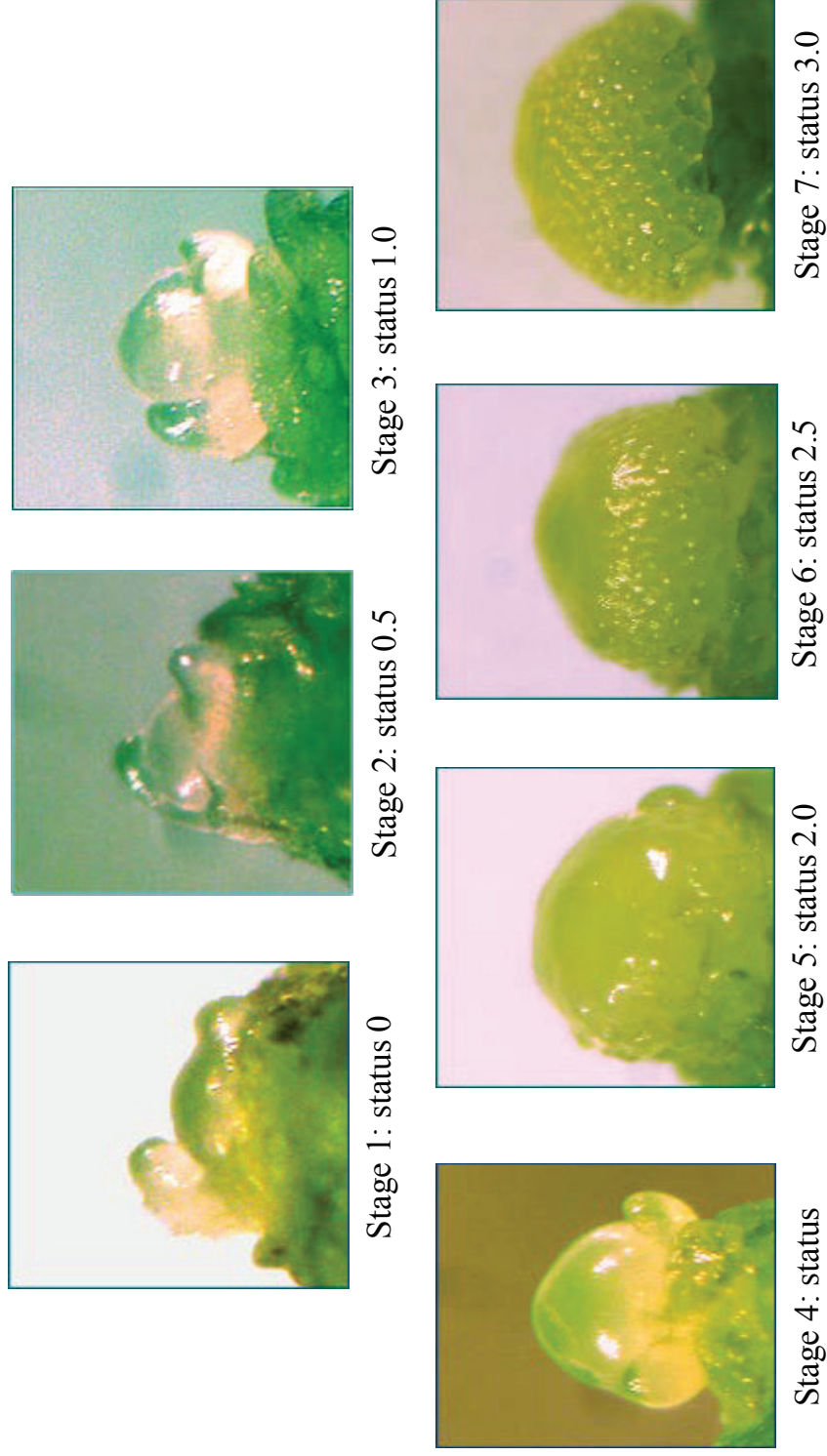


Fig. 1-3 The status in different developmental stages of floral bud of Chrysanthemum.
 Stage 1: no floral bud differentiation; Stage 2: swollen shoot tip; Stage 3: earlier stage of bracts of involucre forming;
 Stage 4: later stage of bracts of involucre forming; Stage 5: earlier stage of floret primordium forming;
 Stage 6: medium stage of floret primordium forming; Stage 7: later stage of floret primordium forming.

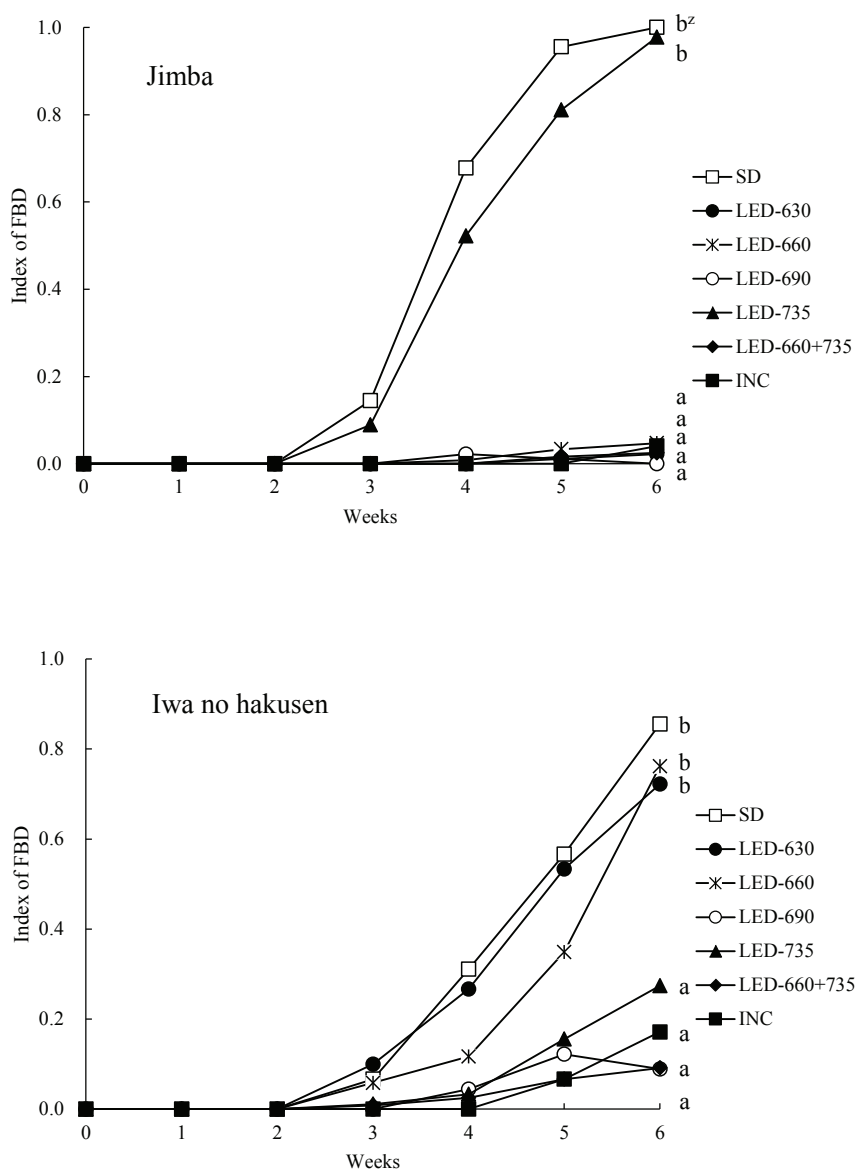


Fig. 1-4 The index of FBD of 'Jimba' and 'Iwa no hakusen' from 0 week to the sixth week after NB treatments.

^z: Difference letters indicate significant difference in index FBD at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

Table 1-1. Wavelengths of peak emission or light intensities of LED patterns and total photon flux of light sources

Wavelength of peak emission	Light intensity (photon flux at wavelength of peak emission)		Total photon flux ^z
	$\mu\text{mol m}^{-2}\text{s}^{-1}\text{nm}^{-1}$	$\mu\text{mol m}^{-2}\text{s}^{-1}\text{nm}^{-1}$	
LED-630	630 nm	0.766	7.87
LED-660	660 nm	0.147	1.83
LED-690	694 nm	0.268	3.78
LED-735	728 nm	0.457	6.60
LED-660 + 735	660 nm, 728 nm	0.059 _(660 nm) , 0.211 _(728 nm)	7.50
INC	—	—	16.10

^z: Total photon flux was calculated from 400 nm to 800 nm.

Table 1-2 Cultivars difference between 'Jimba' and 'Iwa no hakusen' in index of FBD at the sixth week

Treatments	Index of FBD		Cultivars difference
	Jimba	Iwa no hakusen	
SD	1.000	0.856	**z
LED-630	0.022	0.722	*
LED-660	0.047	0.762	*
LED-690	0.000	0.089	*
LED-735	0.978	0.274	*
LED-660 + 735	0.025	0.092	NS
INC	0.041	0.171	*

^z: Asterisk (*) indicate significant difference at $P < 0.05$ by Student's *t* test.

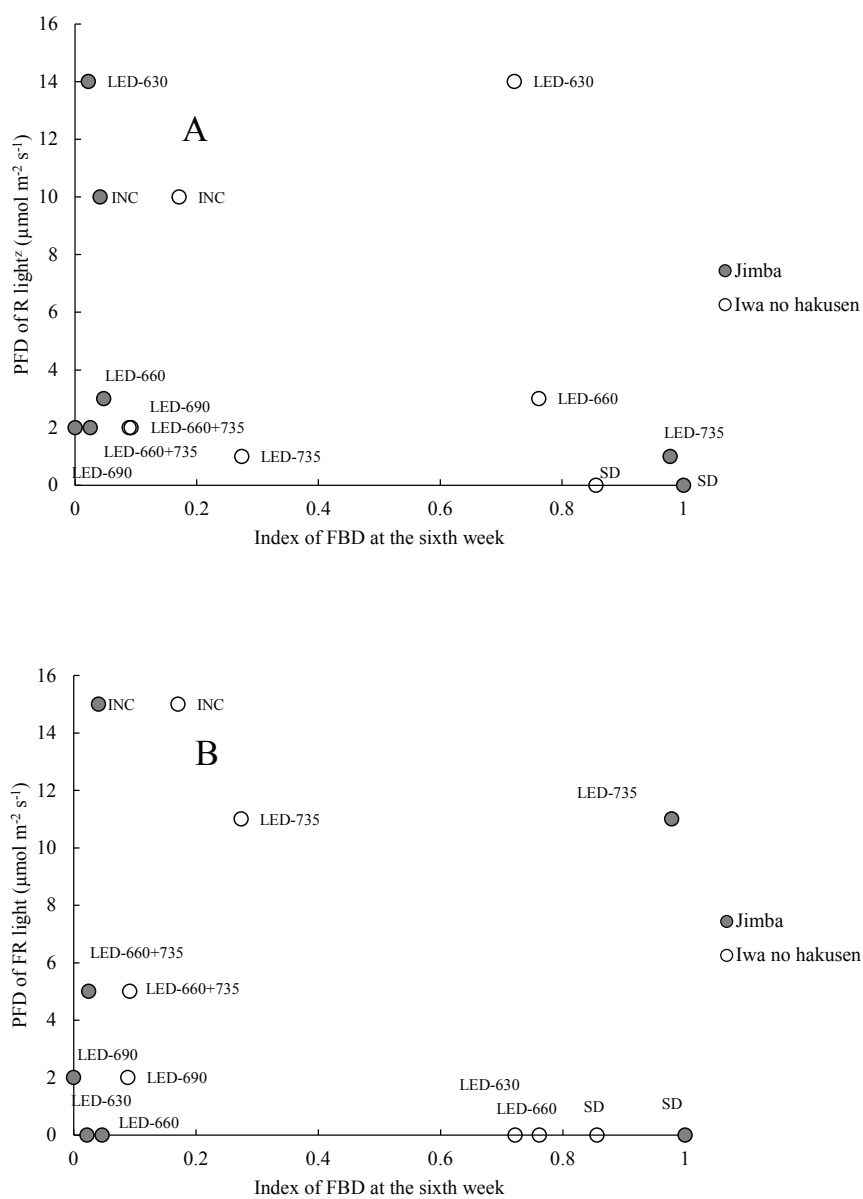


Fig. 1-5 The relation between index of FBD and PFD of R light(A) or PFD of R light (B) after treatment in the sixth week.

^z: PFD of R light in every NB light source was calculated from 600 nm to 699 nm and PFD of FR light was from 700 nm to 799 nm.

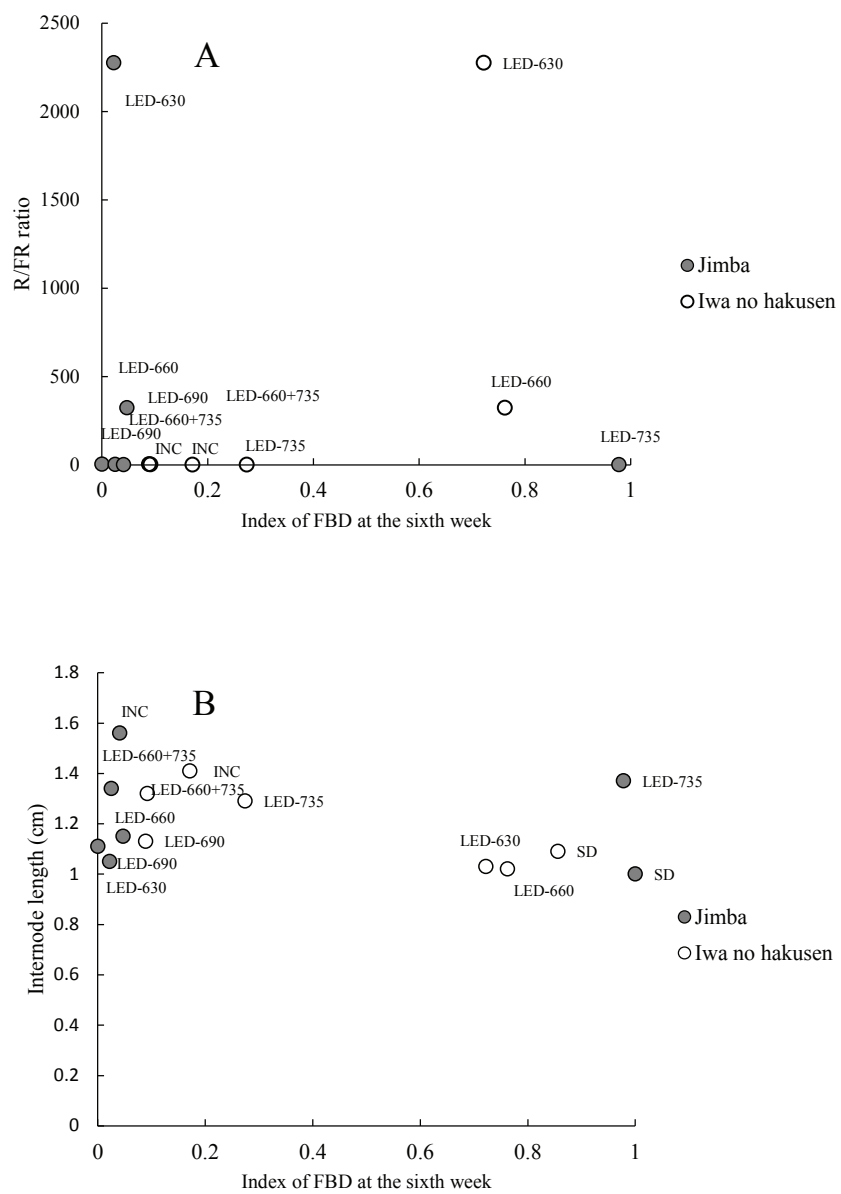


Fig. 1-6 The relation between index of FBD and R/FR ratio (A) or internode length (B) after treatment at the sixth week.

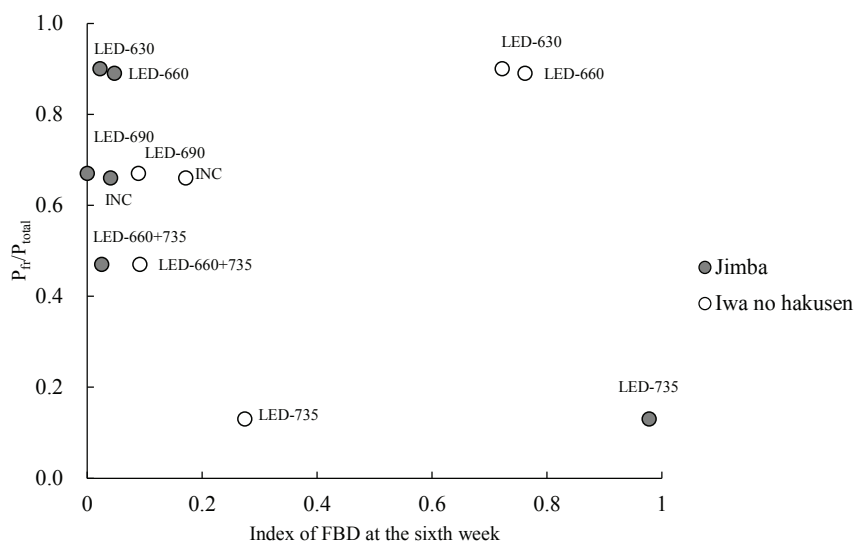


Fig. 1-7 The relation between index of FBD and P_{fr}/P_{total} at the sixth week.

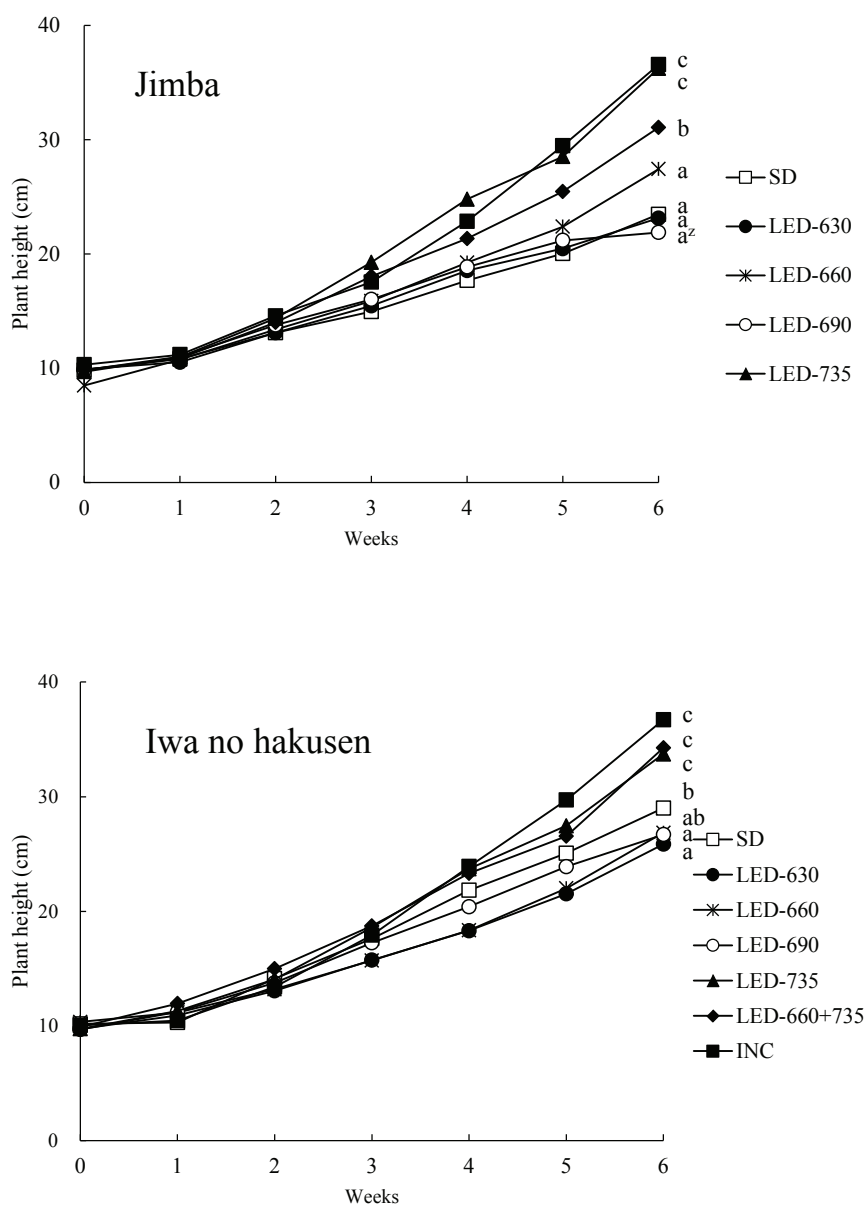


Fig. 1-8 The shoot length of 'Jimba' and 'Iwa no hakusen' from 0 week to the sixth week after NB treatments.

^z: Difference letters indicate significant difference in shoot length at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

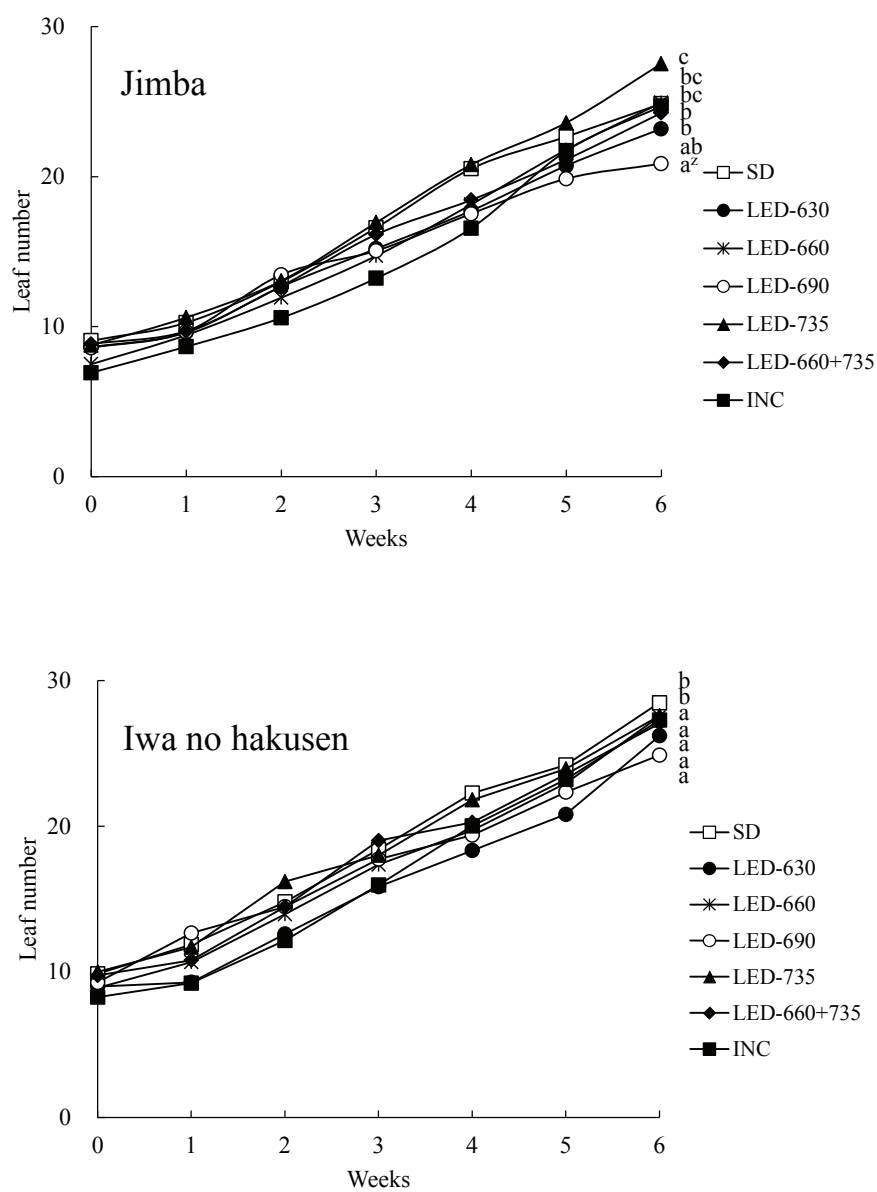


Fig. 1-9 The leaf number of 'Jimba' and 'Iwa no hakusen' from 0 week to the sixth week after NB treatments.

^z: Difference letters indicate significant difference in leaf number at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

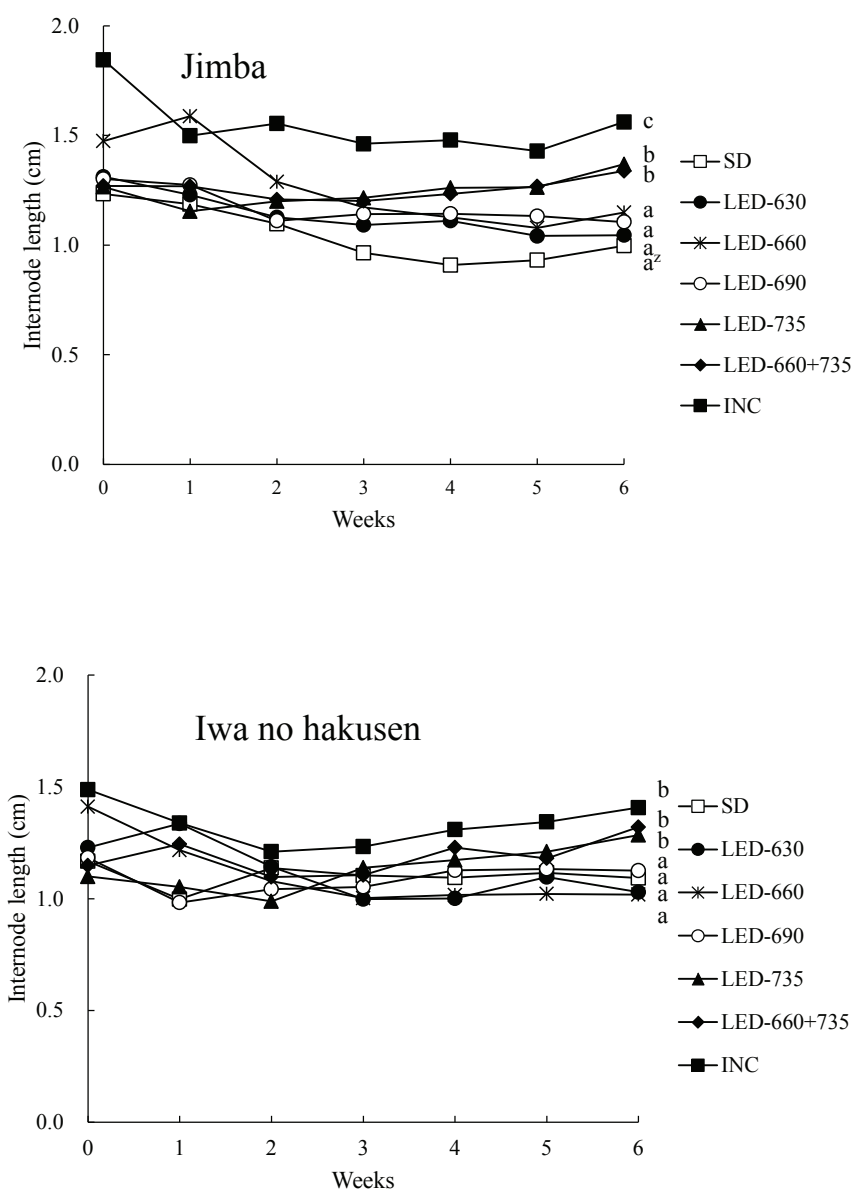


Fig. 1-10 The internode length of 'Jimba' and 'Iwa no hakusen' from 0 week to the sixth week after NB treatments.

^z: Difference letters indicate significant difference in internode length at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

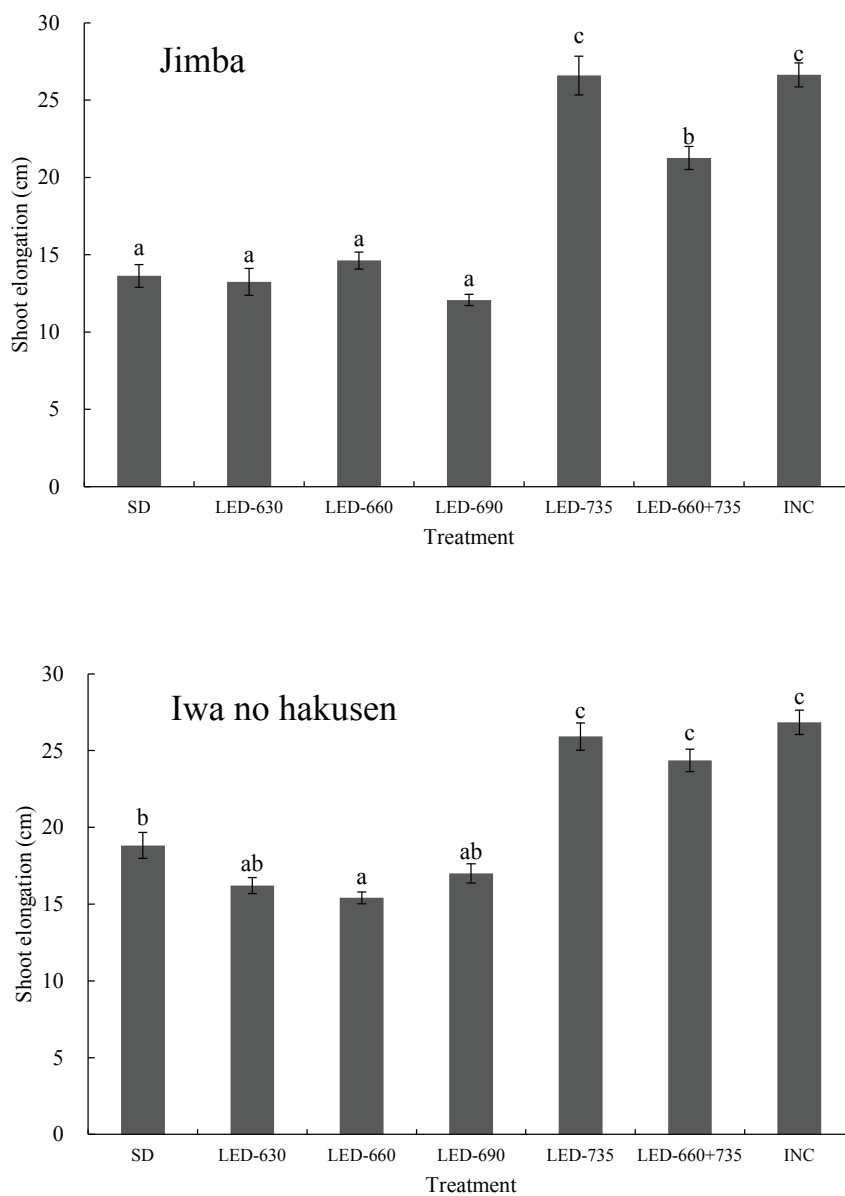


Fig. 1-11 The shoot elongation of 'Jimba' and 'Iwa no hakusen' in the sixth week after treatment.

^z: Difference letters indicate significant difference in elongation at $P < 0.05$ by Tukey's multiple comparison test.

Table 1-3 The cultivar difference in plant height, leaf number, internode length and shoot elongation in the sixth week after treatment.

	Treatment		Cultivar		Cultivars difference	Treatment	Cultivar		Cultivars difference
	Jimba	Iwa no hakusen	Jimba	Iwa no hakusen			Jimba	Iwa no hakusen	
Plant height (cm)	SD	23.5	29.0	29.0	* ^z	SD	1.0	1.1	NS
	LED-630	23.1	25.9	25.9	*	LED-630	1.0	1.0	NS
	LED-660	24.9	25.8	25.8	NS	LED-660	1.0	1.0	NS
	LED-690	21.9	26.7	26.7	*	LED-690	1.1	1.1	NS
	LED-735	36.3	35.8	35.8	NS	LED-735	1.3	1.2	NS
	LED-660 + 735	31.1	34.5	34.5	*	LED-660 + 735	1.3	1.3	NS
	INC	36.9	36.8	36.8	NS	INC	1.5	1.4	*
Leaf number	SD	24.9	28.5	28.5	*	SD	13.6	18.8	*
	LED-630	23.2	26.2	26.2	*	LED-630	13.2	16.2	*
	LED-660	25.1	27.5	27.5	*	LED-660	14.6	15.4	NS
	LED-690	20.9	24.9	24.9	*	LED-690	12.1	17.0	*
	LED-735	27.5	29.5	29.5	NS	LED-735	26.6	25.9	NS
	LED-660 + 735	24.5	27.5	27.5	*	LED-660 + 735	21.3	24.4	*
	INC	24.7	27.2	27.2	*	INC	26.6	26.8	NS

^z: Asterisk (*) indicate significant difference at $P < 0.05$ by Student's t test.

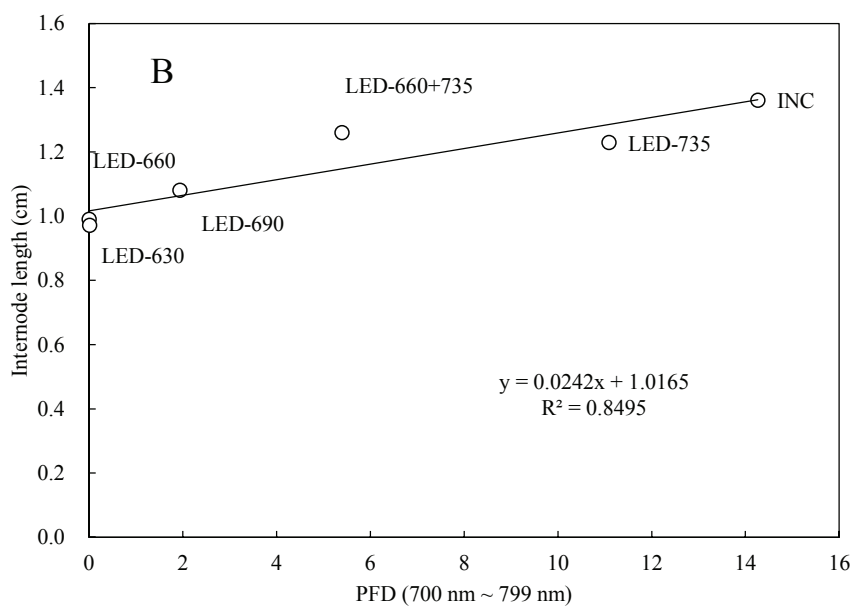
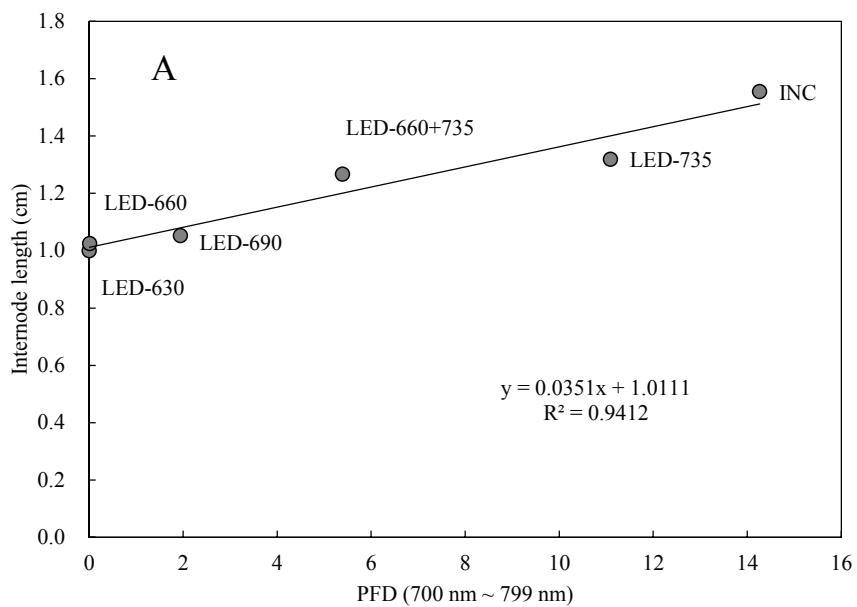


Fig. 1-12 The relation between internode length and PFD at the range of 700 nm to 799 nm in 'Jimba' (A) and in 'Iwa no hakusen' (B).

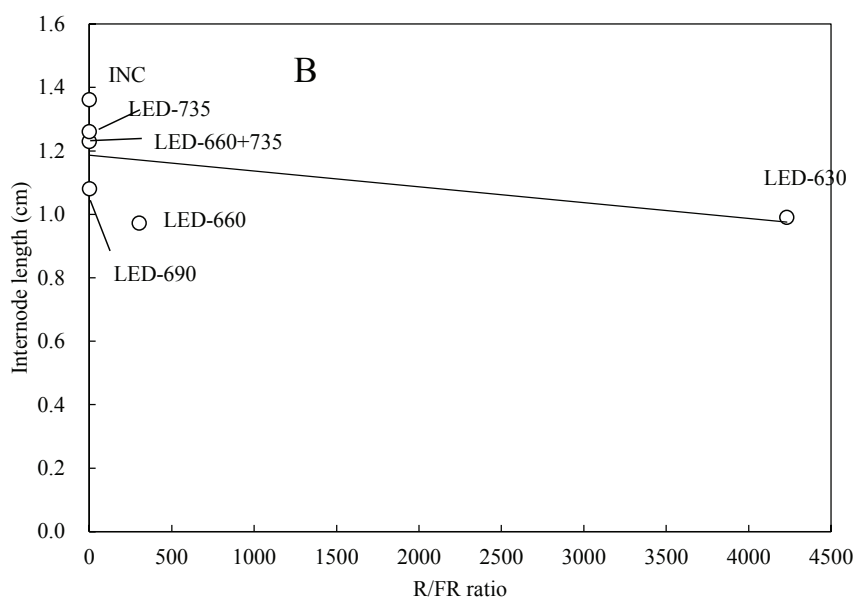
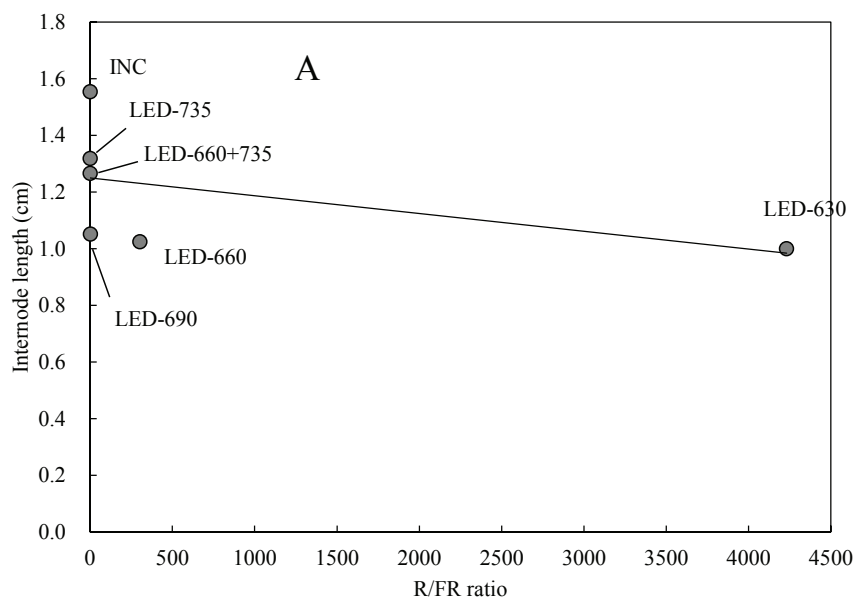


Fig. 1-13 The relation between internode length and R/FR ratio in 'Jimba' (A) or 'Iwa no hakusen' (B).

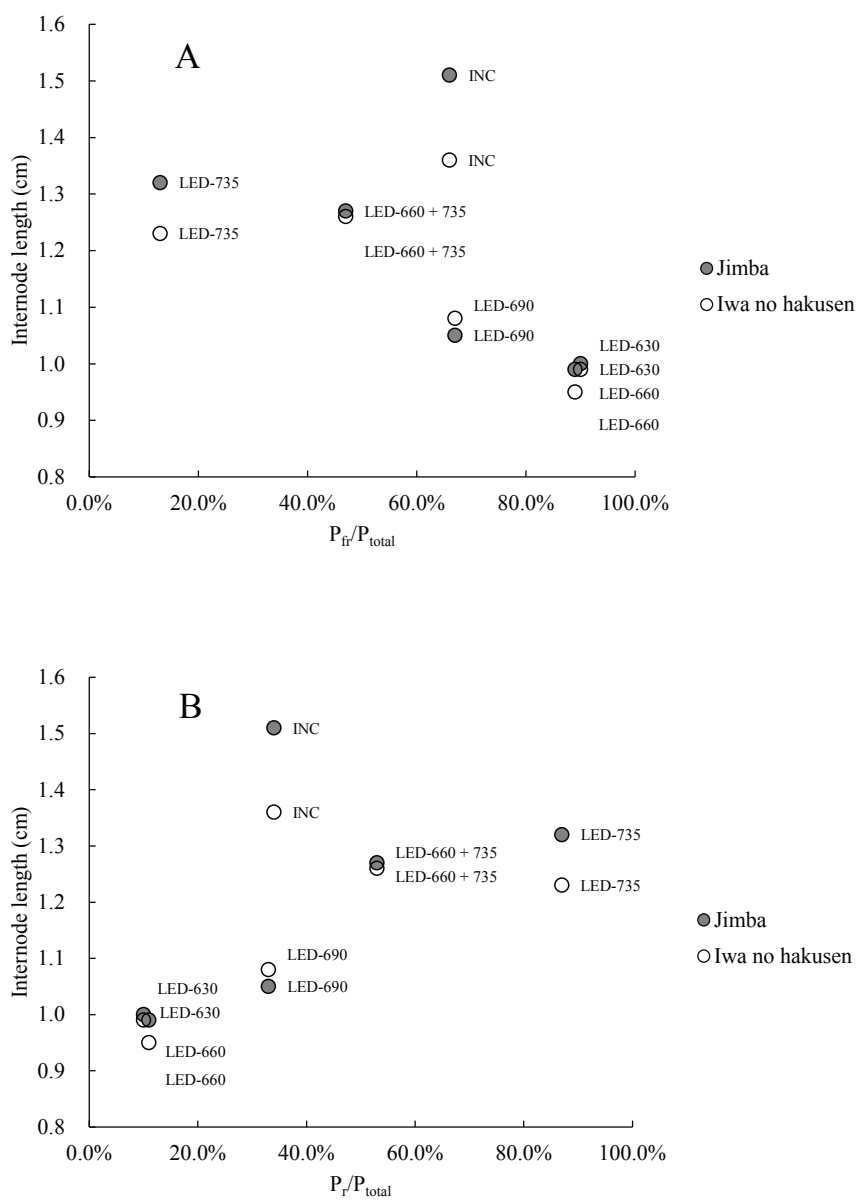


Fig.1-14 The relation between internode length to P_{fir}/P_{total} (A) or P_r/P_{total} (B).

Part 2 FBD Responds to Light Qualities of LED Night Break Lighting in Various Chrysanthemum Cultivars

In previous study, R light of LED peak emission at 630 nm and 660 nm had good effect on inhibiting FBD in autumn flowering type of chrysanthemum cultivar ‘Jimba’, but not in summer-to-autumn flowering type of chrysanthemum cultivar ‘Iwa no hakusen’. And FR light of LED peak emission at 735 nm had different inhibitory effect on FBD in both of the two cultivars. It was supposed that the difference occurred in different flowering type of chrysanthemum. It was also considered that same flowering type of chrysanthemum had same responses to light qualities NB in FBD. For better application LED lamp in chrysanthemum light culture, it was necessary to make it clear on the NB effect of LED light qualities in more chrysanthemum cultivars.

Moreover, NB light intensity was not the same in the previous study of ‘Jimba’ and ‘Iwa no hakusen’. For better NB effect comparison, the LED patterns were adjusted to a similar intensity level in this study.

1. Materials and methods

1.1 *Plant materials*

Twelve chrysanthemum cultivars were used as plant materials. Those cultivars included summer-to-autumn flowering type, autumn flowering type and late-autumn flowering type chrysanthemum and were very popular cultivars in Japan. The flowering season of those cultivars were from late June to December (Table 2-1).

1.2 *Light sources*

Light sources were the same as that was used in Part 1. The light intensity of LED-660 was

the lowest in the 5 different wavelengths of LED lamp, and its light peak emission was $0.138 \mu\text{mol m}^{-2} \cdot \text{s}^{-1} \cdot \text{nm}^{-1}$. For keeping all the treatments in a similar light intensity level, all the NB LED lamps were adjusted into the level of peak emission between 0.06 and $0.10 \mu\text{mol m}^{-2} \cdot \text{s}^{-1} \cdot \text{nm}^{-1}$. For decreasing the LED light intensity, number of the LED sticks was decreased and LED stick was banded up by aluminum foil for blinding part of LED light in some treatments. LED-630 nm LED was decreased to 8 sticks and others were 11 sticks (Fig. 2-1). For adjusting the INC lamp, electric transformer was used and the light intensity was adjusted to $0.1 \mu\text{mol m}^{-2} \cdot \text{s}^{-1} \cdot \text{nm}^{-1}$ at 700 nm.

Light intensity was measured by LI-1800 (LI-COR Inc., Lincoln, NE, USA) light spectrophotometer at the position of 40 cm bellowing the lamps, a distance from the lamp to the surface of planting medium. The light spectrum was showed in Fig. 2-2. And PFD of R and FR light of every treatment was showed in Table 2-2.

1.3 Methods

Shoot cutting was rooted in the plastic case with the size of 32cm x 25cm x 7cm. Two cultivars of plants were rooted in two plastic cases and were carried out the same NB treatments at the same time. The management of growing and NB treatment were the same as Part 1.

1.3 Data collection and analysis

The sampling, items measurement and FBD observation were as same as Part 1. Experiment data were analysed by ANOVA and Tukey's multiple comparison test ($P < 0.05$).

2. Results

2.1 FBD of cultivars by NB treatments

FBD was observed in some of cultivars by treatment LED-690 and LED-735, but some cultivars were not. According the FBD being observed or not by LED-735, we classified the 12

cultivars into two groups, group A and group B.

2.1.1 Group A

There were 9 cultivars that were belonged to group A. They were ‘Sei aegean’, ‘Sei no nami’, ‘Sei elsa’, ‘Remidas’, ‘Sei no makura’, ‘Seiko koumyou’, ‘Seiko no makoto’, ‘Jimba’ and ‘Sei yukino’.

Group A was normal flowering type of chrysanthemum. The cultivars classified into this group can be inhibited their FBD by R light treatments, but not by treatment LED-690 or LED-735. In this group, floral bud began to be observed at the 3rd or the 4th week, and the florets were differentiated at the 6th week by treatment LED-690 or LED-735. Cultivar which flowers in September or October in natural condition belonged to this type. On the other hand, although ‘Sei aegean’ flowers in July in natural condition, it belonged to this type.

(1) Sei aegean

The FBD of ‘Sei aegean’ after treatments was showed in Fig. 2-3. In LED-735, FBD was observed at the 3rd week and the index of FBD reached to 1.0 at the 6th week. In LED-690, FBD was observed at the 4th week and the index of FBD reached to 0.633 at the 6th week. And it was showed inhibitory effect on FBD in treatments of LED-630, LED-660, LED-660+735, and INC, with indices of FBD 0.067, 0.233, 0.067, and 0.033 in the 6th week respectively.

(2) Sei no nami

The FBD of ‘Sei no nami’ after treatments was showed in Fig. 2-3. In LED-735, FBD was observed at the 4th week and the index of FBD reached to 0.867 at the 6th week. In LED-690, FBD was observed at the 5th week and the index of FBD reached to 0.766 at the 6th week. Other treatments were showed perfect inhibitory effect on FBD and the indices of FBD were all 0.0 in the 6th week.

(3) Sei elsa

The FBD of 'Sei elsa' after treatments was showed in Fig. 2-3. In LED-735, FBD was observed at the 3rd week and the index of FBD reached to 1.0 at the 6th week. In LED-690, FBD was observed at the 3rd week and the index of FBD reached to 0.9 at the 6th week. Other treatments of LED-630, LED-660, LED-660+735, and INC were showed inhibitory effect on FBD and the index of FBD reached to 0.067, 0.0, 0.0, and 0.0 respectively in the 6th week.

(4) Remidas

The FBD of 'Remidas' after treatments was showed in Fig. 2-3. In LED-735, FBD was observed at the 3rd week and the index of FBD reached to 1.0 at the 6th week. In LED-690, FBD was observed at the 3rd week and the index of FBD reached to 0.967 at the 6th week. Other treatments of LED-630, LED-660, LED-660+735, and INC were showed perfect inhibitory effect on FBD and the indices of FBD were 0.1, 0.0, 0.0, and 0.0 respectively in the 6th week.

(5) Sei no makura

The FBD of 'Sei no makura' after treatments was showed in Fig. 2-4. In LED-735, FBD was observed at the 3rd week and the index of FBD reached to 1.0 at the 6th week. In LED-690, FBD was observed at the 4th week and the index of FBD reached to 0.8 at the 6th week. Other treatments were showed perfect inhibitory effect on FBD and the indices of FBD were all 0.0 in the 6th week.

(6) Sei koumyou

The FBD of 'Sei koumyou' after treatments was showed in Fig. 2-4. In LED-735, FBD was observed at the 4th week and the index of FBD reached to 0.967 at the 6th week. In LED-690, FBD was observed at the 4th week and the index of FBD reached to 0.9 at the 6th week. Treatments of LED-630 and LED-660 were showed inhibitory effect on FBD and the indices of FBD reached to 0.267 and 0.233 respectively. Treatments of INC and LED-660+735

were showed perfect inhibitory effect on FBD and the indices of FBD were 0.0 in the 6th week.

(7) Seiko no makoto

The FBD of ‘Seiko no makoto’ after treatments was showed in Fig. 2-4. In LED-735, FBD was observed at the 4th week and the index of FBD reached to 0.8 at the 6th week. In LED-690, FBD was observed at the 4th week and the index of FBD reached to 0.467 at the 6th week. LED-660+735 was showed an inhibitory effect on FBD and the index of FBD reached to 0.3 in the 6th week. Other treatments of LED-630, LED-660, and INC were showed perfect inhibitory effect on FBD and the indices of FBD were 0.0, 0.0, and 0.03 respectively in the 6th week.

(8) Jimba

The FBD of ‘Jimba’ after treatments was showed in Fig. 2-4. In LED-735, FBD was observed at the 3rd week and the index of FBD reached to 0.983 at the 6th week. In LED-690, FBD was observed at the 3rd week and the index of FBD reached to 0.8 at the 6th week. Other treatments of LED-630, LED-660, LED-660+735, and INC were showed perfect inhibitory effect on FBD and the indices of FBD were 0.0, 0.017, 0.0, and 0.0 respectively in the 6th week.

(9) Sei yukino

The FBD of ‘Sei yukino’ after treatments was showed in Fig. 2-4. In LED-735, FBD was observed at the 4th week and the index of FBD reached to 0.333 at the 6th week. Other treatments of LED-630, LED-660, LED-690, LED-660+735, and INC were showed inhibitory effect on FBD and indices of FBD were 0.0, 0.0, 0.133, 0.033, and 0.0 respectively in the 6th week.

2.1.2 Group B

Group B was NB lighting sensitive type of chrysanthemum. FBD was inhibited seriously by any NB light quality in this group. Two summer-to-autumn flowering type cultivars and one

late-autumn flowering type of chrysanthemum cultivar were belonged to group B. They were ‘Cent west’, ‘Sei opti’, and ‘Sei tsudoï’.

(1) Cent west

The FBD of ‘Cent west’ after treatments was showed in Fig. 2-4. The FBD was inhibited by any treatments. And the indices of FBD in treatments of LED-630, LED-660, LED-690, LED-735, LED-660+735, and INC were 0.1, 0.033, 0.033, 0.0, 0.1, and 0.0 respectively.

(2) Sei opti

The FBD of ‘Opti’ after treatments was showed in Fig. 2-4. The FBD was inhibited in all treatments. And the indices of FBD in treatments of LED-630, LED-660, LED-690, LED-735, LED-660+735, and INC were 0.033, 0.033, 0.1, 0.033, 0.033, and 0.033 respectively.

(3) Sei tsudoï

The FBD of ‘Sei tsudoï’ after treatments was showed in Fig. 2-4. The FBD was inhibited in all treatments. The index of FBD was 0.033 in LED-735 and it was all 0 in the other treatments.

2.2 Plant growth items in various cultivars after NB treatments

The plant growths items were included plant height, leaf number, internode length and shoot elongation that were measured every week. All the data were showed in order from the maximum to the minimum after 6 weeks of treatments.

(1) Cent west

The results of plant heights were 42.9, 42.3, 42.2, 41.3, 38.0, and 34.4 cm in treatments of LED-660+735, LED-735, LED-690, LED-INC, LED-630, and LED-660 respectively at 6th week. Except for LED-660, plant height was no difference in all the treatments (Fig. 2-6). Leaf numbers were 34.0, 32.0, 31.4, 29.0, 28.0, and 26.2 in treatments of LED-630, LED-660+735, INC, LED-690, LED-660, and LED-735 respectively at 6th week (Fig. 2-8). The results of

internode length were 1.61, 1.46, 1.34, 1.31, 1.22, and 1.12 cm in treatments of LED-735, LED-690, LED-660+735, INC, LED-660, and LED-630 respectively. Except for LED-690, internode length in LED-735 was and significant longer than the other treatments (Fig. 2-10). The results of shoot elongation were 35.2, 34.5, 34.4, 33.5, 30.3, and 26.6 cm in treatments of LED-660+735, LED-735, LED-690, INC, LED-630, and LED-660 respectively. Shoot elongation was no difference in all the treatments except for LED-630 (Fig. 2-12).

The top three results of plant height and internode length were in the three treatments of LED-660+735, LED-735, and LED-690. And shoot elongation was in LED-660+735, INC and LED-735.

(2) Sei Aegean

The data of plant heights were 35.1, 30.4, 30.2, 25.3, 23.4, and 22.7 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively at 6th week. Plant height in LED-735, LED-660+735, and INC was significant longer than those in other treatments (Fig. 2-6). Leaf numbers were 27.0, 25.4, 25.4, 24.0, 22.8, and 21.8 in treatments of INC, LED-735, LED-660+735, LED-690, LED-660, and LED-630 respectively at 6th week. Leaf number in INC was the most and significant more than those in LED-630, LED-660, and LED-690 (Fig. 2-8). The data of internode length were 1.30, 1.20, 1.19, 1.08, 1.06, and 0.99 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively. Internode length in INC was significant longer than those in LED-630, LED-660, and LED-690 (Fig. 2-10). The data of shoot elongation were 35.1, 30.4, 30.2, 25.3, 23.4, and 22.7 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively. Shoot elongation in INC was the longest and significant longer than those in other treatments (Fig. 2-12).

The top three results of plant height, internode length and elongation were in the treatments

of INC, LED-735, and LED-660+735.

(3) Sei opti

The results of plant heights were 34.0, 30.8, 30.7, 26.6, 21.2, and 20.3 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively at 6th week. Plant heights in LED-735, LED-660+735, and INC were significant longer than those in other treatments (Fig. 2-6). Leaf numbers were 26, 24.6, 24.4, 24.2, 21.8, and 21.4 in treatments of INC, LED-690, LED-735, LED-660+735, LED-660, and LED-630 respectively at 6th week. Leaf number in INC was significant more than those in LED-630 and LED-660 (Fig. 2-8). The results of internode length were 1.30, 1.27, 1.26, 1.08, 0.99, and 0.93 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results in LED-735, LED-660+735, and INC were significant longer than those in other treatments (Fig. 2-10). The results of elongation were 34.0, 30.8, 30.7, 26.6, 21.2, and 20.3 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively. The result in INC was significant longer than those in LED-630, LED-660, and LED-690 (Fig. 2-12).

The top three results of plant height, internode length and elongation were in the treatments of INC, LED-735, and LED-660+735.

(4) Sei no nami

The data of plant heights were 26.2, 24.2, 23.6, 21.1, 20.3, and 18.2 cm in treatments of LED-735, INC, LED-690, LED-660+735, LED-630, and LED-660 respectively at 6th week. The longest plant height was in LED-690 and significant longer than those in LED-630, LED-660, and LED-660+735 (Fig. 2-6). Leaf numbers were 26.6, 26.0, 24.6, 24.6, 23.4, and 22.6 in treatments of LED-735, LED-690, INC, LED-660+735, LED-630, and LED-660 respectively at 6th week. Except for LED-660, in leaf numbers were no difference in all the treatments (Fig. 2-8). The data of internode length were 0.99, 0.95, 0.93, 0.87, 0.86, and 0.81 cm in treatments of

INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results of internode length in LED-735, INC, and LED-690 were significant longer than those in other treatments (Fig. 2-10). The data of elongation were 26.2, 24.2, 23.6, 21.1, 20.3, and 18.2 cm in treatments of LED-735, INC, LED-690, LED-660+735, LED-630, and LED-660 respectively. The results of elongation in LED-735 and INC were significant longer than those in other treatments (Fig. 2-12).

The top three results of plant height, internode length and elongation were in the treatments of LED-735, INC, and LED-690.

(5) Sei elsa

The results of plant height were 29.8, 28.6, 27.5, 26.7, 23.1, and 21.3 cm in treatments of LED-735, LED-690, LED-660+735, INC, LED-630, and LED-660 respectively at 6th week. The results in LED-735, LED-690, and LED-660+735 were significant longer than those in LED-630 and LED-660 (Fig. 2-6). Leaf numbers were 26.4, 24.6, 23.8, 23.0, 22.2 and 21.4 in treatments of LED-690, LED-735, INC, LED-660+735, LED-660 and LED-630 respectively at 6th week. Except for LED-630 and LED-690, leaf number was no difference in treatments (Fig. 2-8). The results of internode length were 1.21, 1.20, 1.12, 1.09, 1.08, and 0.96 cm in treatments of LED-735, LED-660+735, INC, LED-690, LED-630, and LED-660 respectively. Except for LED-660, internode length was no difference in treatments (Fig. 2-10). The results of shoot elongation were 20.7, 19.6, 18.5, 17.7, 14.1, and 12.2 cm in treatments of LED-735, LED-690, LED-660+735, INC, LED-630, and LED-660 respectively. Shoot elongation in LED-630 and LED-660 was significant shorter than those in other treatments (Fig. 2-12).

The top three results of plant height and elongation were in the treatments of LED-735, LED-690, and LED-660+735. The top three results of internode length were in the treatments of LED-735, LED-660+735, and INC.

(6) Remidas

The data of plant heights were 30.1, 29.6, 29.0, 28.1, 21.7, and 20.6 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively at 6th week. The results in LED-630 and LED-660 were significant shorter than those in other treatments (Fig. 2-6). Leaf numbers were 22.8, 20.8, 19.2, 18.6, 17.8, and 17.8 in treatments of LED-690, LED-735, INC, LED-660+735, LED-630, and LED-660 respectively at 6th week. Also Leaf numbers in LED-630 and LED-660 were significant less than those in other treatments (Fig. 2-8). The data of internode length were 1.57, 1.56, 1.42, 1.23, 1.21, and 1.15 cm in treatments of INC, LED-660+735, LED-735, LED-690, LED-630, and LED-660 respectively. The results in LED-735, LED-660+735 and INC were significant longer than those in other treatments (Fig. 2-10). The data of elongation were 19.2, 18.7, 18.1, 17.2, 10.8, and 9.7 cm in treatments of INC, LED-735, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results of LED-630 and LED-660 were significant less than those in other treatments (Fig. 2-12).

The top three results of plant height, internode length and elongation were in the treatments of INC, LED-735, and LED-660+735.

(7) Sei no makura

The data of plant heights were 27.0, 25.6, 24.7, 23.8, 23.0, and 23.0 cm in treatments of LED-735, INC, LED-690, LED-660+735, LED-630, and LED-660 respectively at 6th week and were no difference in all treatments (Fig. 2-7). Leaf numbers were 22.6, 20.0, 19.8, 19.6, 19.6, and 19.2 in treatments of LED-735, LED-660, LED-630, LED-690, LED-660+735, and INC respectively at 6th week and were also no difference in all treatments (Fig. 2-9). The data of internode length were 1.33, 1.26, 1.21, 1.20, 1.17, and 1.16 cm in treatments of INC, LED-690, LED-735, LED-660+735, LED-630, and LED-660 respectively. Except for LED-630 and LED-660, the results of internode length were no difference in other treatments (Fig. 2-11). The

data of elongation were 27.0, 25.6, 24.7, 23.8, 23.0, and 23.0 cm in treatments of LED-735, INC, LED-690, LED-660+735, LED-630, and LED-660 respectively and were no difference in all treatments (Fig. 2-13).

The top three results of plant height and elongation were in the treatments of INC, LED-735, and LED-690. In internode length, the top three results were in treatments of INC, LED-690, and LED-660+735.

(8) Sei koumyou

The data of plant height were 25.8, 24.3, 23.4, 20.9, 17.3, and 16.7 cm in treatments of INC, LED-660+735, LED-735, LED-690, LED-630, and LED-660 respectively at 6th week. The results of plant height in LED-735, LED-660+735, and INC were significant longer than those in other treatments (Fig. 2-7). Leaf numbers were 23.4, 23.0, 22.4, 22.4, 18.4, and 18.0 in treatments of LED-690, LED-735, INC, LED-660+735, LED-630, and LED-660 respectively at 6th week. Except for LED-630 and LED-660, leaf numbers were no difference in treatments (Fig. 2-9). The data of internode length were 1.15, 1.09, 1.02, 0.95, 0.94, and 0.89 cm in treatments of INC, LED-660+735, LED-735, LED-630, LED-660, and LED-690 respectively. The result in INC was significant longer than those in other treatments (Fig. 2-11). The data of elongation were 16.1, 14.6, 13.8, 11.2, 7.6, and 7.1 cm in treatments of INC, LED-660+735, LED-735, LED-690, LED-630, and LED-660 respectively. The results in LED-735, LED-660+735 and INC were significant longer than those in LED-630 and LED-660 (Fig. 2-13).

The top three results of plant height, internode length and elongation were in the treatments of INC, LED-660+735, and LED-735.

(9) Seiko no makoto

The data of plant height were 25.7, 25.2, 22.7, 22.5, 21.4, and 20.1 cm in treatments of LED-735, INC, LED-660+735, LED-690, LED-630, and LED-660 respectively at 6th week. The

results of plant height in LED-735 and INC were significant longer than those in LED-630, LED-660, and LED-690 (Fig. 2-7). Leaf numbers were 24.6, 24.4, 24.0, 23.0, 20.6, and 20.2 in treatments of LED-630, LED-690, LED-735, LED-660, INC, and LED-660+735 respectively at 6th week. The results of leaf number in LED-630, LED-690, and LED-735 were significant more than those in LED-660+735 and INC (Fig. 2-9). The data of internode length were 1.22, 1.13, 1.07, 0.92, 0.91, and 0.87 cm in treatments of INC, LED-660+735, LED-735, LED-690, LED-660, and LED-630 respectively. The results in LED-735, LED-660+735 and INC were significant longer than those in other treatments (Fig. 2-11). The data of elongation were 16.2, 15.8, 13.3, 13.1, 12.0 and 11.5 cm in treatments of LED-735, INC, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results in LED-735 and INC were significant longer than those in LED-630 and LED-660 (Fig. 2-13).

The top three results of plant height, internode length and elongation were in the treatments of LED-735, INC, and LED-660+735.

(10) Jimba

The data of plant heights were 28.5, 27.3, 26.6, 24.7, 23.0, and 22.9 cm in treatments of INC, LED-660+735, LED-735, LED-690, LED-660, and LED-630 respectively at 6th week. The result in INC was significant longer than those in LED-630, LED-660, and LED-690 (Fig. 2-7). Leaf numbers were 27.4, 25.9, 25.9, 25.5, 25.8, and 23.5 in treatments of LED-690, LED-660+735, LED-660, LED-630, INC, and LED-735 respectively at 6th week. Except for LED-735, Leaf numbers were no difference in treatments (Fig. 2-9). The data of internode length were 1.13, 1.10, 1.05, 0.90, 0.90, and 0.89 cm in treatments of LED-735, INC, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results in LED-735, LED-660+735 and INC were significant longer than those in other treatments (Fig. 2-11). The data of elongation were 19.3, 16.5, 16.4, 13.6, 12.9, and 12.4 cm in treatments of INC, LED-735,

LED-660+735, LED-690, LED-630, and LED-660 respectively. The result in INC was significant longer than those in LED-630, LED-660, and LED-690 (Fig. 2-13).

The top three results of plant height, internode length and elongation were in the three treatments of INC, LED-735, and LED-660+735.

(11) Sei yukino

The results of plant heights were 33.8, 29.2, 29.1, 27.4, 27.0, and 26.6 cm in treatments of LED-735, INC, LED-660+735, LED-690, LED-630, and LED-660 respectively at 6th week. The results in LED-735, LED-660+735 and INC were significant longer than those in LED-630 and LED-660 (Fig. 2-7). Leaf numbers were 30.0, 29.6, 28.6, 28.4, 26.0, and 25.8 in treatments of LED-660, LED-690, LED-735, LED-630, INC, and LED-660+735 respectively at 6th week (Fig. 2-9). The results of internode length were 1.18, 1.13, 1.13, 0.94, 0.93, and 0.89 cm in treatments of LED-660+735, LED-735, INC, LED-630, LED-690, and LED-660 respectively. The results of LED-735, LED-660+735, and INC were significant longer than those in other treatments (Fig. 2-11). The data of elongation were 25.1, 20.6, 20.5, 18.8, 18.3, and 18.0 cm in treatments of LED-735, INC, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results of LED-735, LED-660+735, and INC were significant longer than those in LED-630 and LED-660 (Fig. 2-13).

The top three results of plant height, internode length and elongation were in the three treatments of LED-735, INC, and LED-660+735.

(12) Sei tsudoii

The data of plant heights were 20.9, 19.6, 18.3, 18.3, 18.2, and 17.5 cm in treatments of LED-735, LED-660+735, INC, LED-690, LED-630, and LED-660 respectively at 6th week. The result in LED-735 was significant longer than those in LED-630 and LED-660 (Fig. 2-7). Leaf numbers were 26.2, 26.2, 24.8, 23.0, 21.8, and 20.4 in treatments of LED-630, LED-660,

LED-690, LED-660+735, LED-735, and INC respectively at 6th week. Except for INC, leaf numbers were no difference in treatments (Fig. 2-9). The data of internode length were 0.96, 0.90, 0.86, 0.74, 0.70, and 0.67 cm in treatments of LED-735, INC, LED-660+735, LED-690, LED-630, and LED-660 respectively. The results in LED-735, LED-660+735 and INC were significant longer than those in other treatments (Fig. 2-11). The data of elongation were 13.3, 12.0, 10.8, 10.8, 10.7, and 10.0 cm in treatments of LED-735, LED-660+735, INC, LED-690, LED-630, and LED-660 respectively. Except for LED-660+735, the result in LED-735 was significant longer than those in other treatments (Fig. 2-13).

The top three results of plant height, internode length and elongation were in the three treatments of INC, LED-735, and LED-660+735.

3. Discussion

3.1 FBD

Chrysanthemum cultivars group A included all the autumn flowering type chrysanthemum cultivars, which flower in October in the natural condition in Japan, summer-to-autumn flowering type chrysanthemum cultivars of ‘Sei aegean’ and ‘Sei no nami’, which flower in July and September respectively in the natural condition in Japan, and late autumn flowering type chrysanthemum cultivar ‘Sei yukino’, which flowers in November in the natural condition in Japan. The entire autumn flowering type chrysanthemum cultivars were inhibited their FBD by R light NB or R light contained NB, and had no respond to the NB treatment of LED-735. As discussion in Part 1 of ‘Jimba’, autumn flowering type chrysanthemum could not sense the FR light NB in flowering responding. FR light could not inhibit the flowering in autumn flowering type chrysanthemum. FR light was just like the dark treatment to autumn flowering type chrysanthemum, as same as without NB.

Although ‘Sei aegean’ and ‘Sei no nami’ were summer-to-autumn flowering type chrysanthemum, they belonged to this group. Except for LED-735, FBD was inhibited by NB treatments, as same as those results in autumn flowering type chrysanthemum. It was suggested that some cultivars of summer-autumn flowering type had similar light quality responding to autumn flowering type of chrysanthemum in flowering.

Late-autumn flowering type of ‘Sei yukino’, which flowering season was November, was also belong to group A. In treatment LED-735, the index of FBD was lower than those of other cultivars in group A, being showed an inhibitory result in FBD. Since its critical day length was about 11 h, 12 h of day time length in this study was a little bit long for FBD of ‘Sei yukino’. In LED-735, the FBD result might be related to the inhibitory effect by the day length to ‘Sei yukino’.

FBD was observed in treatment LED-690 in all the cultivars of group A. Except for ‘Sei yukino’, the index of FBD was very high in LED-690, just little lower than those in LED-735. Ohishi (2011) reported that LED-630 nm light had most effect on inhibition of chrysanthemum flowering. Wavelength of LED-690 nm was far from LED-630 nm in spectrum, so that it was indicated that LED-690 nm light had weaker flowering inhibitory ability than LED-630 nm. Furthermore, the light intensity of LED-690 was lower than that in Part 1. FBD of ‘Jimba’ was inhibited by LED-690 in study Part 1, but not in this study. It was suggested that LED-690 nm LED in this light intensity was not capable of inhibiting the FBD of group A in this study.

Group B included 3 cultivars of summer-to-autumn flowering type chrysanthemum ‘Cent west’, ‘Sei opti’, and late-autumn flowering flowering type chrysanthemum ‘Sei tsudo’. In summer-to-autumn flowering type of ‘Cent west’, ‘Sei opti’, FBD were inhibited by LED-735, different to ‘Sei aegean’ and ‘Sei no nami’ in group A. On the other hand, R light and other NB treatments were also inhibited FBD of the two cultivars. The results indicated that the FBD

responding to FR light NB was different in the group of summer-to-autumn flowering type chrysanthemum. Therefore, it was suggested that the light regulating flowering related internal factor in summer-to-autumn flowering type of chrysanthemum were more complicated than that in autumn flowering type of chrysanthemum. For understanding the internal difference between the two groups, more researches were needed to be done in molecule study in the future.

In same light intensity of treatment LED-690, the FBD was inhibited in ‘Cent west’ and ‘Sei opti’, but not in the cultivars of group A. It was suggested that ‘Cent west’ and ‘Sei opti’ were more sensitive to light intensity and quality than other cultivars.

Late-autumn flowering type of ‘Sei tsudoï’ was belonged to group B. Similar to the ‘Sei yukino’ in group A, 12 h of day length might have already inhibited the FBD of ‘Sei tsudoï’. Compared to ‘Sei yukino’, ‘Sei tsudoï’ flowered in December, a shorter day length than that of ‘Sei yukino’. It was suggested that critical day length of ‘Sei tsudoï’ was shorter than ‘Sei yukino’. So that, the inhibitory effect on the FBD of ‘Sei tsudoï’ by 12 h of day length was more than that in ‘Sei yukino’.

R light of LED-630 and LED-660 could not inhibit the FBD of summer-to-autumn flowering type of chrysanthemum cultivar ‘Iwa no hakusen’ in Part 1 and high index of FBD was observed. However the similar results were not found in other summer-to-autumn flowering type cultivars in this study. Although it was showed index of FBD higher than 0.2 in summer-autumn flowering type of ‘Sei aegean’ and autumn flowering type of ‘Seiko koumyou’ in the treatment of LED-630 or LED-660, it was suggested that ‘Sei aegean’ was little instability and ‘Seiko koumyou’ was less sensitive under the treatment of R light NB.

3.2 Plant height

As a cut-flower, plant height of chrysanthemum was an important index in production.

After six weeks of treatment, there were six cultivars, which were ‘Sei aegean’, ‘Sei opti’, ‘Sei no nami’, ‘Remidas’, ‘Seiko koumyou’ and ‘Jimba’, which had the highest result in treatment INC (Fig. 4). And there were six cultivars, which were ‘Cent west’, ‘Sei elsa’, ‘Sei no makura’, ‘Seiko no makoto’, ‘Sei yukino’ and ‘Sei tsudoï’, which had the highest result in treatment LED-735. Plant height results in treatment LED-660 + 735 were no difference to those in INC or LED-735, except that in ‘Seiko no makoto’. On the other hand, the shortest plant height results were in the treatments of LED-660 or LED-630.

Treatments of INC, LED-735, and LED-660 + 735 were far red (FR) light contained treatments. FR light promotes plant shoot growth in many plants (Hirai et al., 2006; Arai and Ohishi, 2010). Therefore those FR light contained treatments regulated the significant higher plant height than those in without FR light treatments, LED-660 or LED-630.

In every treatment, ‘Cent west’ had the longest plant height and was significant different to the cultivars (Table 2-3). In R light NB treatment of LED-630 and LED-660, plant height of ‘Seiko koumyou’ had the shortest and was significant shorter than most of other cultivars. In FR light contained treatments, plant height of ‘Sei tsudoï’ became the shortest and significant shorter than many cultivars. The plant height of ‘Sei tsudoï’ did not elongate much in FR light contained treatments. It was suggested that FR light NB had better promotive effect on shoot growth in ‘Seiko koumyou’ than in ‘Sei tsudoï’. Careless the cultivars, it was promoted shoot growth in FR light NB more than in R light NB. Compared to the R light contained treatments, all the cultivars were in the similar more elongation tendency in FR light contained treatments.

3.3 Leaf number

The maximum leaf number were 34.0 (LED-630), 27.0 (INC), 26.0 (INC), 26.6 (LED-735), 26.4 (LED-690), 22.8 (LED-690), 22.6 (LED-735), 23.4 (LED-690), 24.6 (LED-630), 27.4

(LED-690), 30.0 (LED-660), and 26.2 (LED-630) in cultivars of ‘Cent west’, ‘Cei aegean’, ‘Sei opti’, ‘Sei no nami’, ‘Sei elsa’, ‘Remidas’, ‘Sei no makura’, ‘Seiko koumyou’, ‘Seiko no makoto’, ‘Jimba’, ‘Sei yukino’, and ‘Sei tsudoï’ respectively (Table 2-4). And the minimum leaf number were 26.2 (LED-735), 21.8 (LED-630), 21.4 (LED-630), 22.6 (LED-660), 21.4 (LED-630), 17.8 (LED-660), 19.2 (INC), 18.0 (LED-660), 20.2 (LED-660 + 735), 22.8 (LED-735), 25.8 (LED-660 + 735), and 20.4 (INC) respectively. From the results, we could not find any correlation between leaf number and NB light qualities.

In ‘Sei no makura’, leaf number was no difference between any NB treatments (Fig. 2-9). In cultivars of ‘Cent west’, ‘Sei opti’, ‘Sei no nami’, ‘Sei elsa’, and ‘Jimba’, there were no significant difference between any treatments, except the maximum and minimum leaf number results. Therefore, we concluded that there was no effect on leaf number by NB light qualities. The results were similar to the Part 1.

3.4 Internode length

There were six cultivars, which were ‘Cei aegean’ (1.30 cm), ‘Sei opti’ (1.30 cm), ‘Remidas’ (1.57 cm), ‘Sei no makura’ (1.33 cm), ‘Seiko koumyou’ (1.15 cm), and ‘Seiko no makoto’ (1.22 cm), which had the longest internode length in treatment INC (Table 2-5). And there were six cultivars, which were ‘Cent west’ (1.61 cm), ‘Sei no nami’ (0.99 cm), ‘Sei elsa’ (1.21 cm), ‘Jimba’ (1.19 cm), ‘Sei yukino’ (1.18 cm), and ‘Sei tsudoï’ (0.96 cm), which had the longest internode length in treatment LED-735. Except for LED-690 in ‘Seiko koumyou’, the shortest internode length results were all in treatments LED-630 or LED-660.

From the results, similar to the plant height, the longest internode length results were all in FR light contained treatments of INC or LED-735. Although there was no longest internode length in treatment LED-660 + 735, there was no significant difference between internode

length in treatment LED-660 + 735 and INC or LED-735, except for in 'Sei no nami'. While there were the shortest results in those without FR contained treatments, LED-630 or LED-660. As mentioned above, because NB light qualities had no effect on leaf number, the plant height was contributed by internode length. We concluded that FR light contained treatments promoted plant height by promoting internode length.

The study were supported the results of FR light increasing in internode elongation in Part 1 and had similar results to many sun plants (Erik et al., 2001).

3.5 Shoot elongation

Cultivar 'Cent west' had the longest shoot elongation in all treatments and was significant different to that in other cultivars. 'Seiko koumyou' has the shortest shoot elongation in LED-630 and LED-660, but in FR light contained treatments it was little longer than 'Sei tsudoï' (Table 2-6). Although the results were affected by light quality, it was also decided by the characteristic of cultivars themselves. For example, long shoot length of cultivar 'Cent west' was still longer after FR irradiation, while short length of cultivar 'Sei tsudoï' was still shorter than others.

Because phytochromes had maximum absorption spectra at 660 nm and 740 nm (Sager J.C. et al., 1988), wavelength at 660 nm and 735 nm in this study had the maximum effect of R and FR light on shoot elongation. For comparing the shoot elongation effect by FR light and by R light in cultivars, we calculated the results by using the method of subtracting plant height of LED-660 by that of LED-735 (Fig. 2-14).

In Fig. 2-14, cultivar 'Sei opti' was more than 10 cm and was the longest in cultivars. Although 'Cent west' had the longest shoot elongation, the 735 nm of FR light had more contributive elongation in 'Sei opti' than in 'Cent west'. It was suggested that the elongation of

'Sei opti' was easier affect by 735 nm of FR light than that of 'Cent west'. Except 'Cent west', cultivars of 'Iwa no hakusen', 'Sei nami', 'Sei elsa' and 'Remidas' were around 8 cm and they were also sensitive to elongation induced by LED-735 nm of FR. Cultivars of 'makura', 'makoto', 'Jimba' and 'tsudo' were less than 5 cm and it was suggested that they were less sensitive to elongation induced by 735 nm of FR.

Total conclusion

R light NB treatment inhibited FBD in all the cultivars in this study. In summer-to-autumn flowering type of chrysanthemum, we could not find cultivar like 'Iwa no hakusen', which was lost NB effect by R light. FR light could not inhibit all the cultivars of autumn flowering type and some cultivars of summer-to-autumn flowering type of chrysanthemum. And many cultivars were less sensitive to LED-690 in inhibitory of FBD.

It was not like our expectation that it could not inhibit FBD in all the cultivars of summer-to-autumn flowering type of chrysanthemum by R light NB. Summer-to-autumn flowering type was more complex than autumn flowering type chrysanthemum in flowering responding to light quality of NB. It was confirmed that combination of R and FR light LED treatment of LED-660 + 735 had a good inhibitory on FBD in all the cultivars. Thus, a combination of R and FR light of LED lamps was a potential light source for inhibiting flowering in chrysanthemums.

Table 2-1 Twelve chrysanthemum cultivars were used in the study, including summer-to-autumn flowering, autumn flowering, and late-autumn flowering types.

Cultivar flowering group	Cultivar			Critical day length	Natural flowering time
	Name	Flower color	Flower form type		
Summer-to-autumn flowering type	Cent west				Jun.
	Sei aegean	white	spray chrysanthemum	16~24 h	Jul.
	Sei opti	yellow	spray chrysanthemum		Jul.
	Sei no nami	white	standard chrysanthemum	Sep.	
Autumn flowering type	Sei elsa	white	standard chrysanthemum		Oct.
	Remidas	yellow	spray chrysanthemum		Oct.
	Sei no makura	yellow	standard chrysanthemum	12~15 h	Oct.
	Seiko koumyou	yellow	standard chrysanthemum		Oct.
	Seiko no makoto	white	standard chrysanthemum		Oct.
	Jimba	white	standard chrysanthemum		Oct.
Late-autumn flowering type	Sei yukino	white	spray chrysanthemum		Nov.
	Sei tsudoji	yellow	spray chrysanthemum	~11 h	Dec.

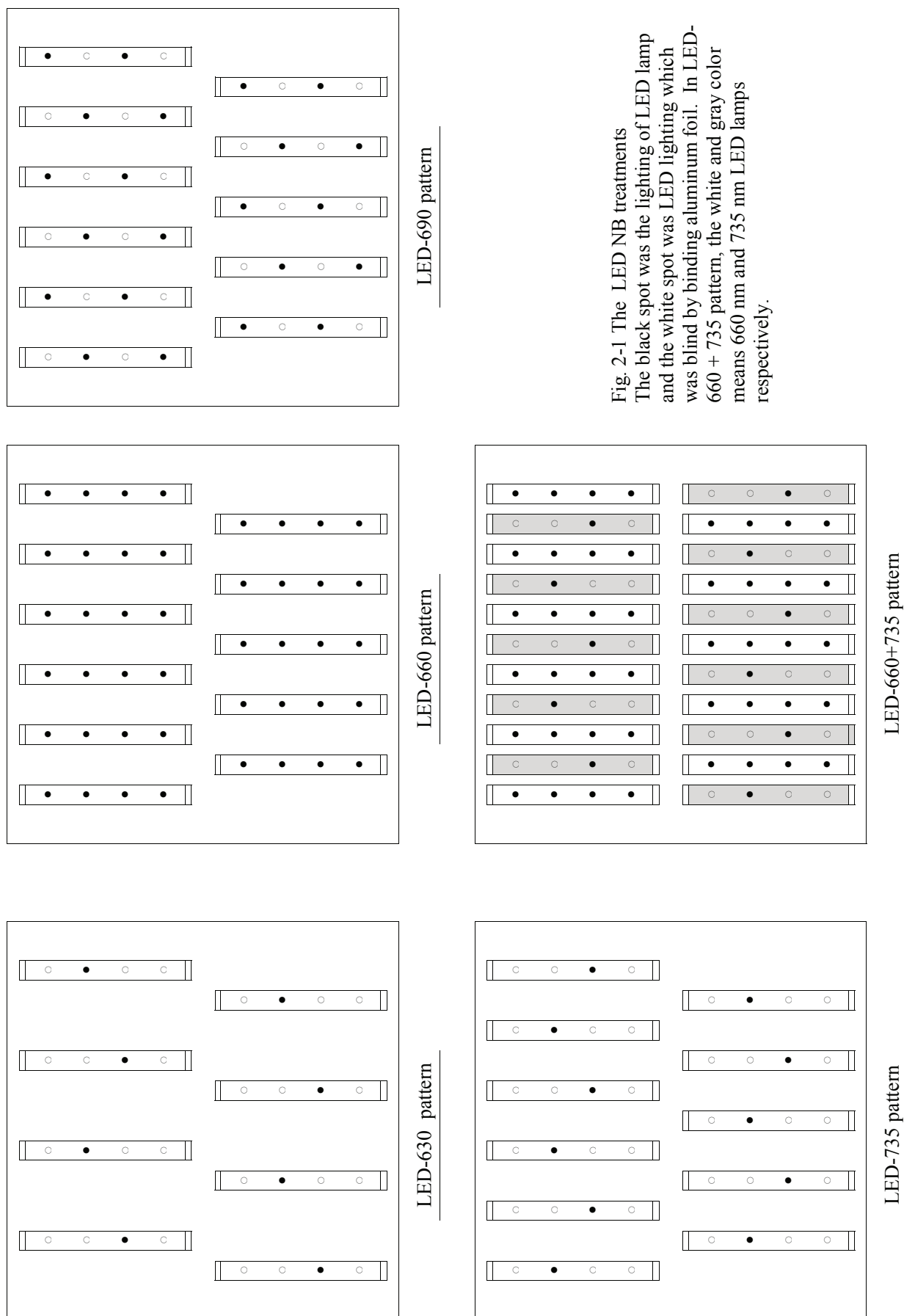


Fig. 2-1 The LED NB treatments
 The black spot was the lighting of LED lamp
 and the white spot was LED lighting which
 was blind by binding aluminum foil. In LED-
 660 + 735 pattern, the white and gray color
 means 660 nm and 735 nm LED lamps
 respectively.

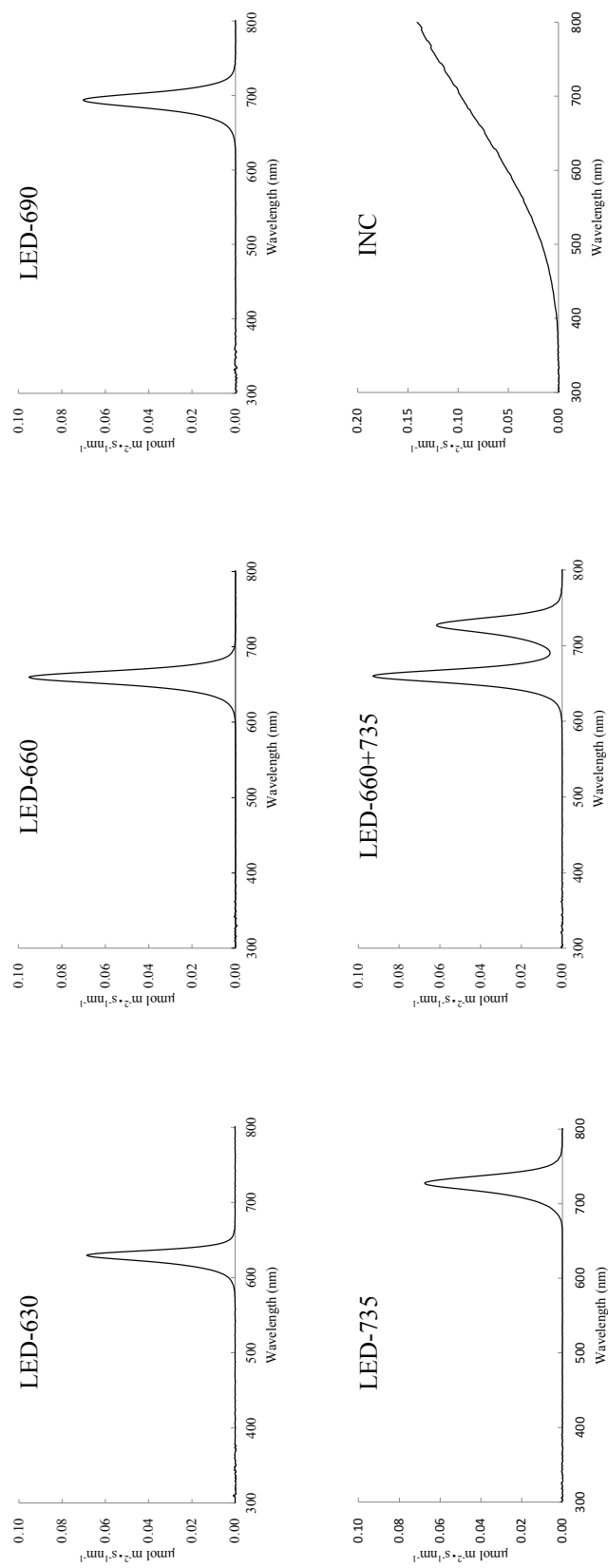


Fig. 2-2 Light spectrum of six NB light treatments. LED light intensity was adjusted to the level between 0.06 and 0.10 $\mu\text{mol m}^{-2} \cdot \text{s}^{-1} \cdot \text{nm}^{-1}$ at wavelength of peak emission. The light intensity of INC lamp was adjusted to 0.10 $\mu\text{mol m}^{-2} \cdot \text{s}^{-1} \cdot \text{nm}^{-1}$ at 700 nm.

Table 2-2 The PFD and R/FR ratio of all the light sources.

	LED-630	LED-660	LED-690	LED-735	LED-660+735	INC
PFD of R light ($\mu\text{mol m}^{-2}\text{s}^{-1}$) ^z	15.497	2.369	1.429	0.090	2.401	7.392
PFD of FR light ($\mu\text{mol m}^{-2}\text{s}^{-1}$) ^y	0.007	0.006	0.487	1.854	1.692	11.821
R/FR	2259.999	388.185	2.934	0.049	1.419	0.625

^z: PFD of R light was calculated from 600 nm to 699 nm.

^y: PFD of FR light was calculated from 700 nm to 799 nm.

Group A

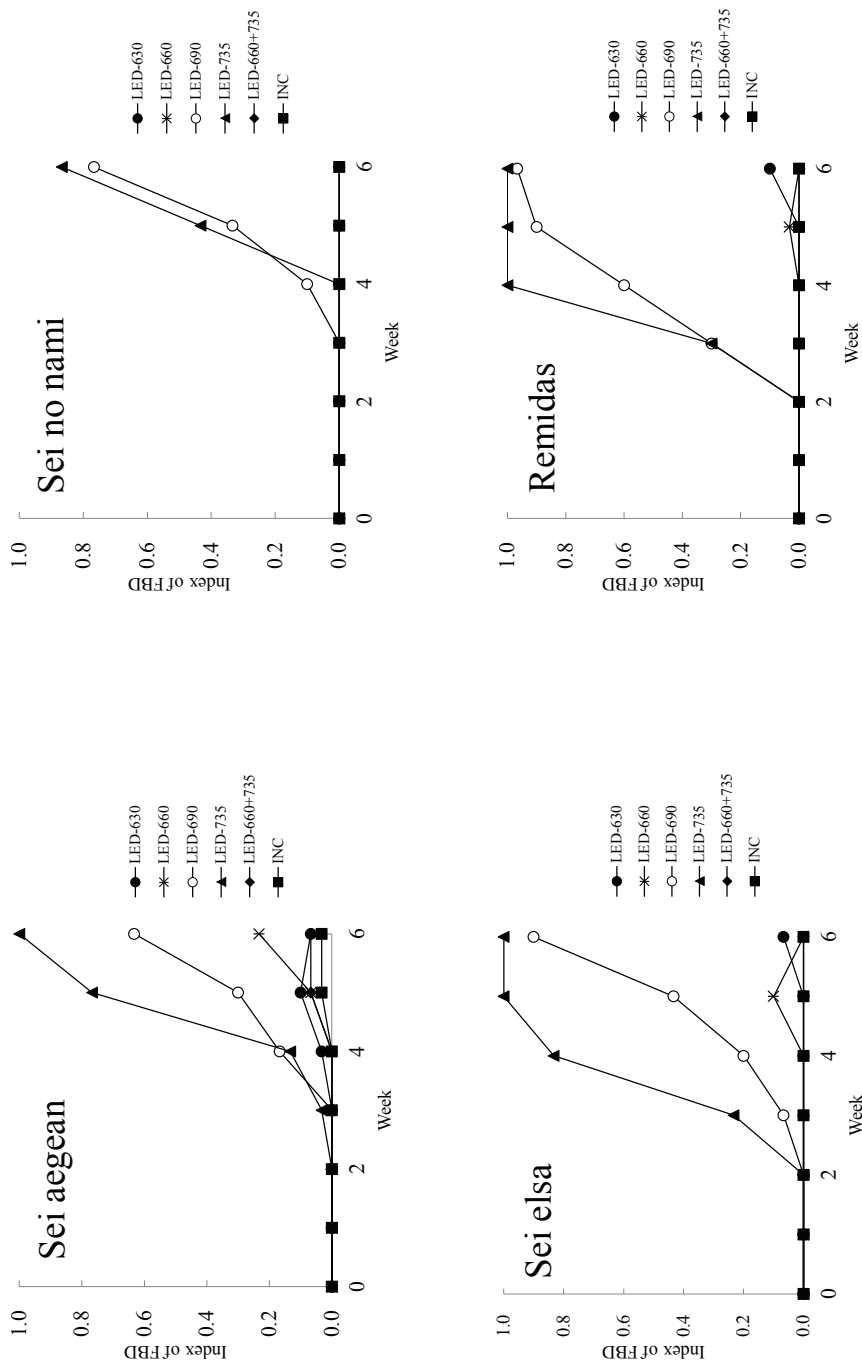


Fig. 2-3 The index of FBD of cultivars in group A from 0 week to the sixth week after NB treatments.

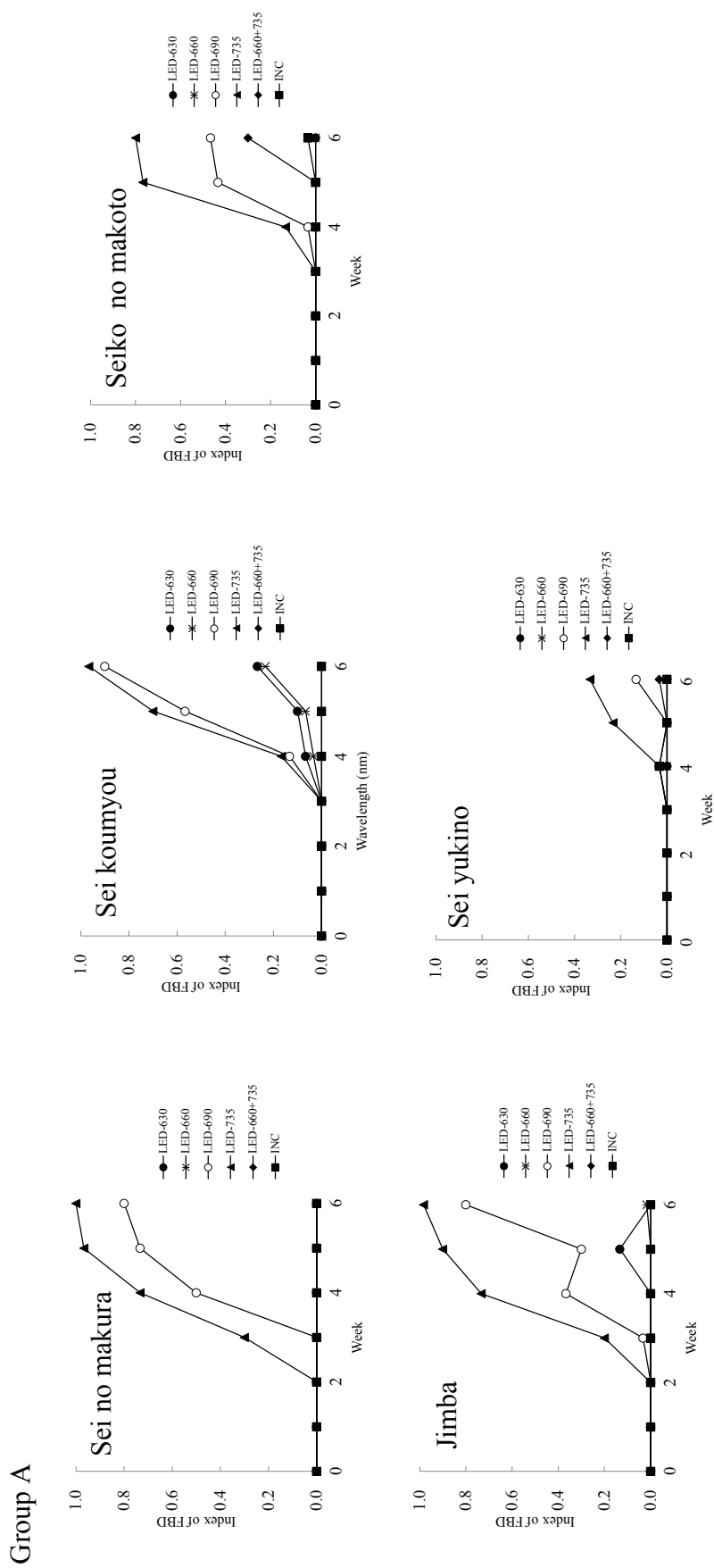


Fig. 2-4 The index of FBD of cultivars in group A from 0 week to the sixth week after NB treatments.

Group B

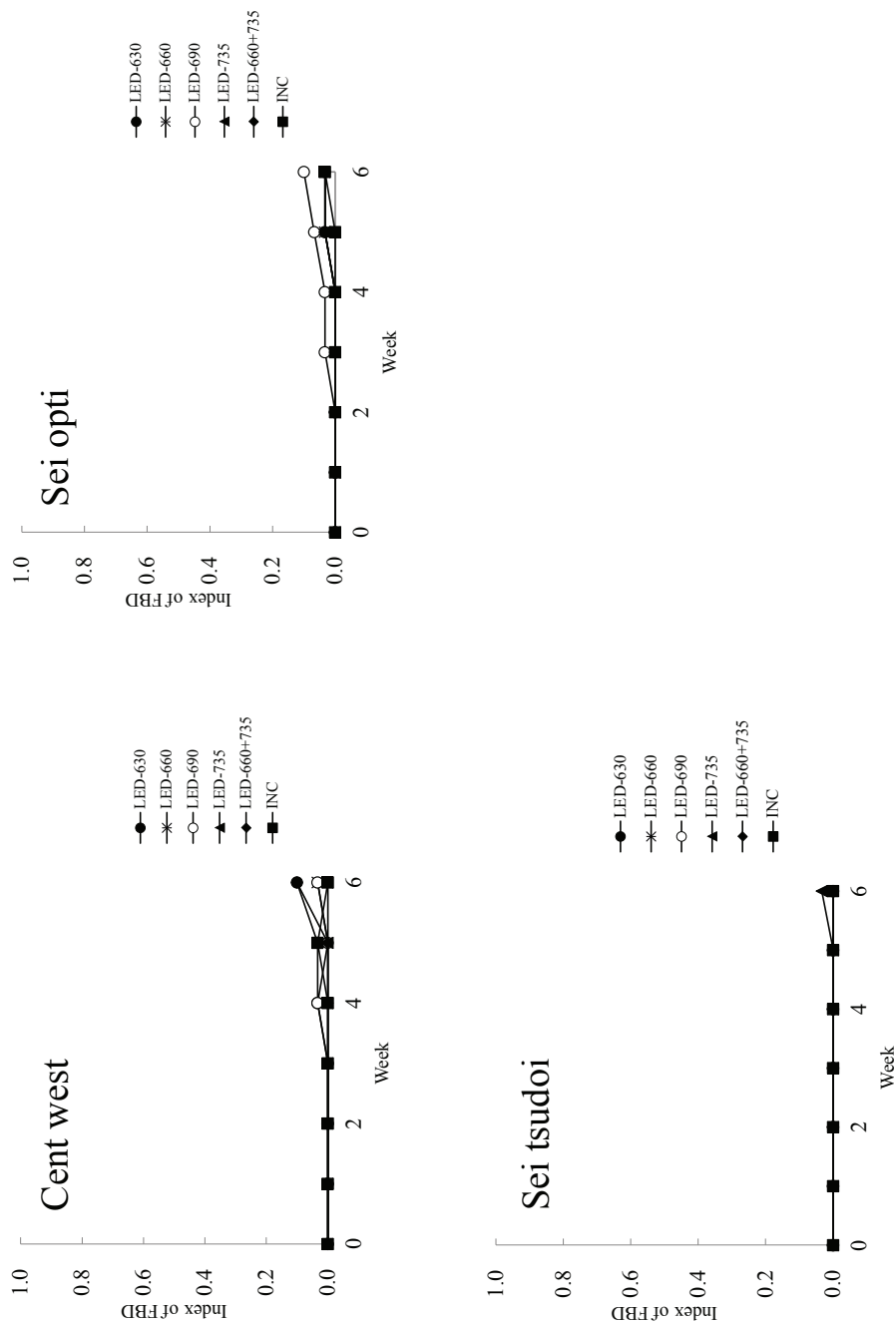


Fig. 2-5 The index of FBD of cultivars in group B from 0 week to the sixth week after NB treatments.

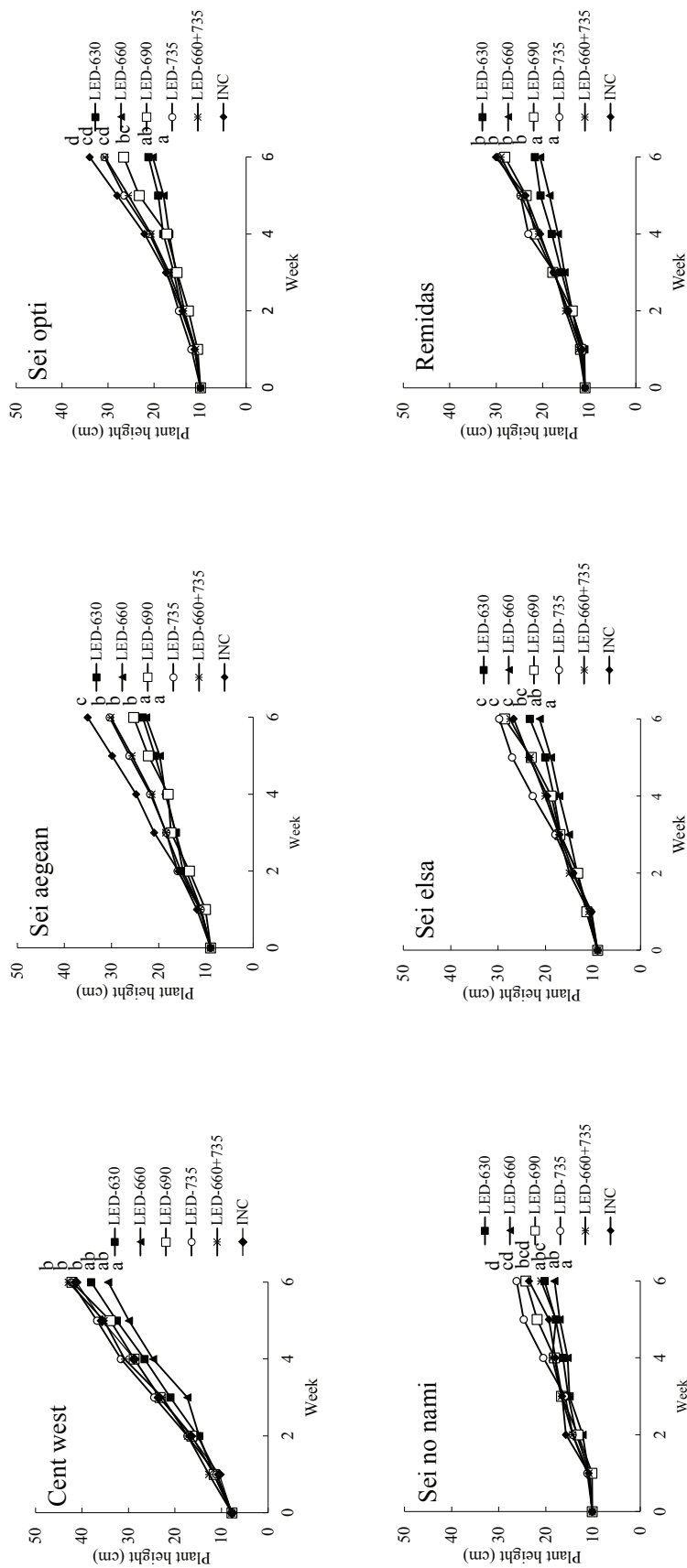


Fig. 2-6 The plant height of cultivars from 0 week to the sixth week after NB treatments.

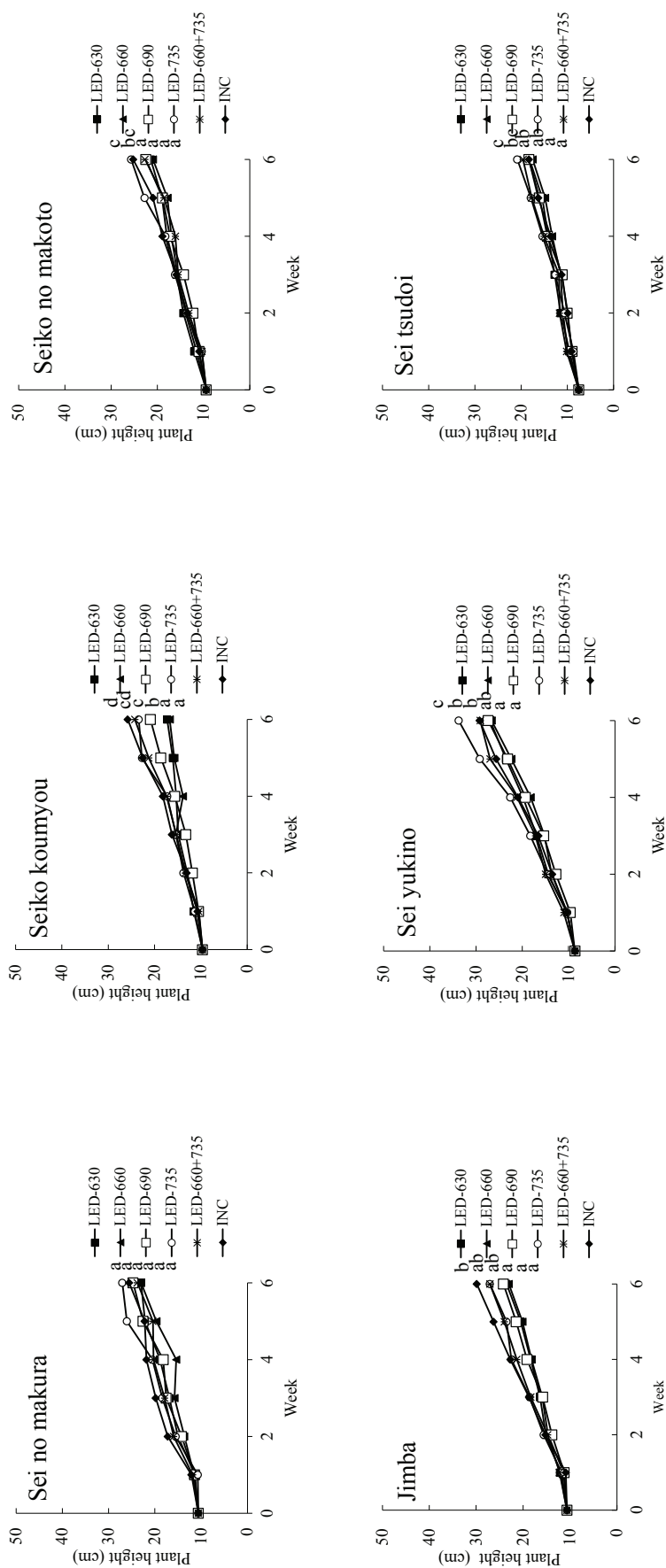


Fig. 2-7 The shoot length of cultivars from 0 week to the sixth week after NB treatments.

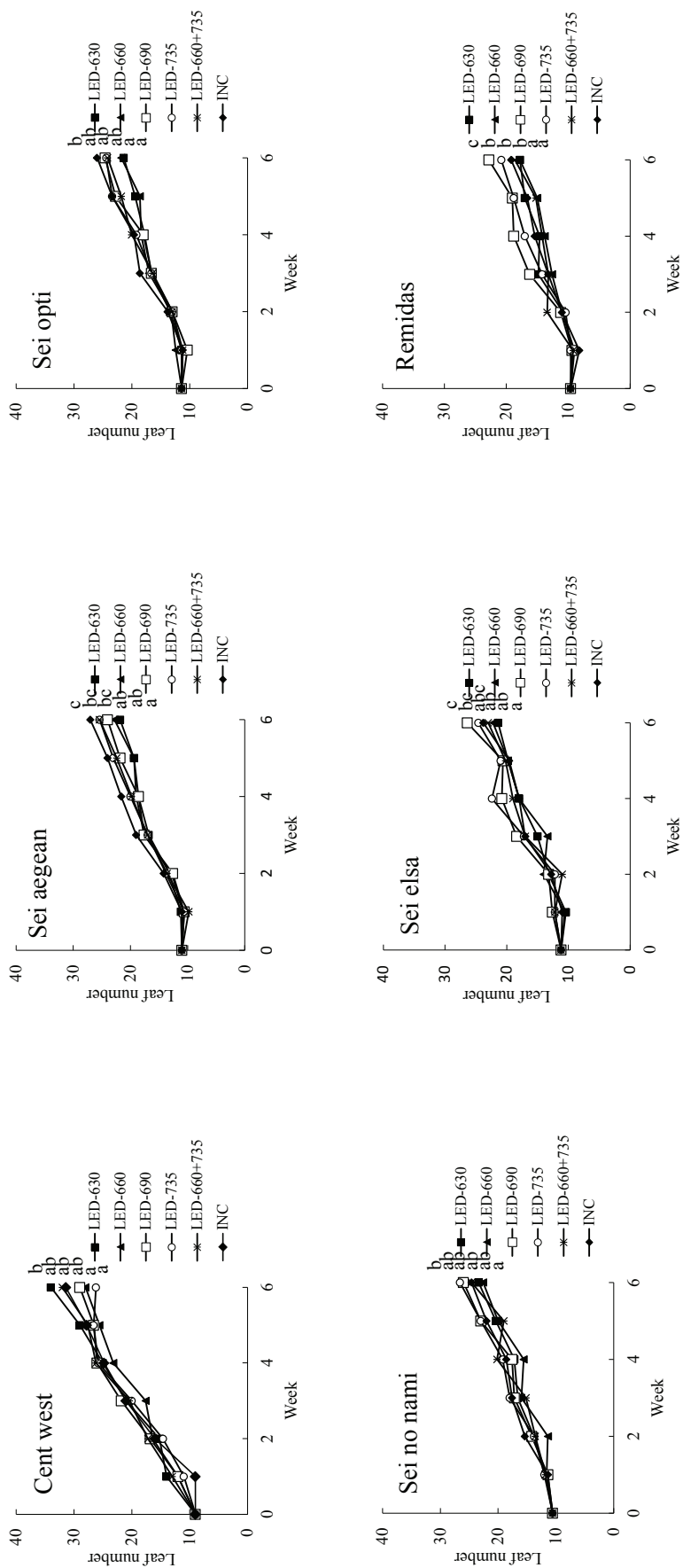


Fig. 2-8 The leaf number of cultivars from 0 week to the sixth week after NB treatments.

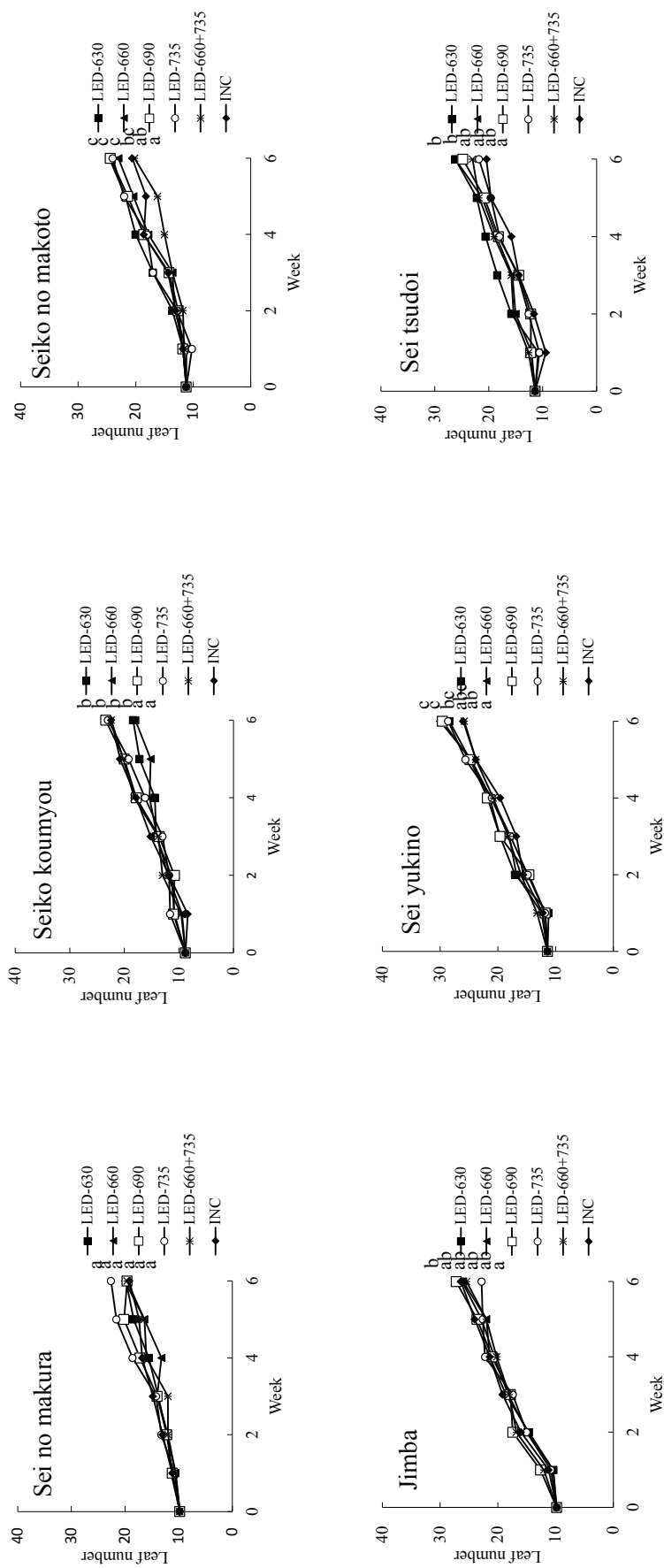


Fig. 2-9 The leaf number of cultivars from 0 week to the sixth week after NB treatments.

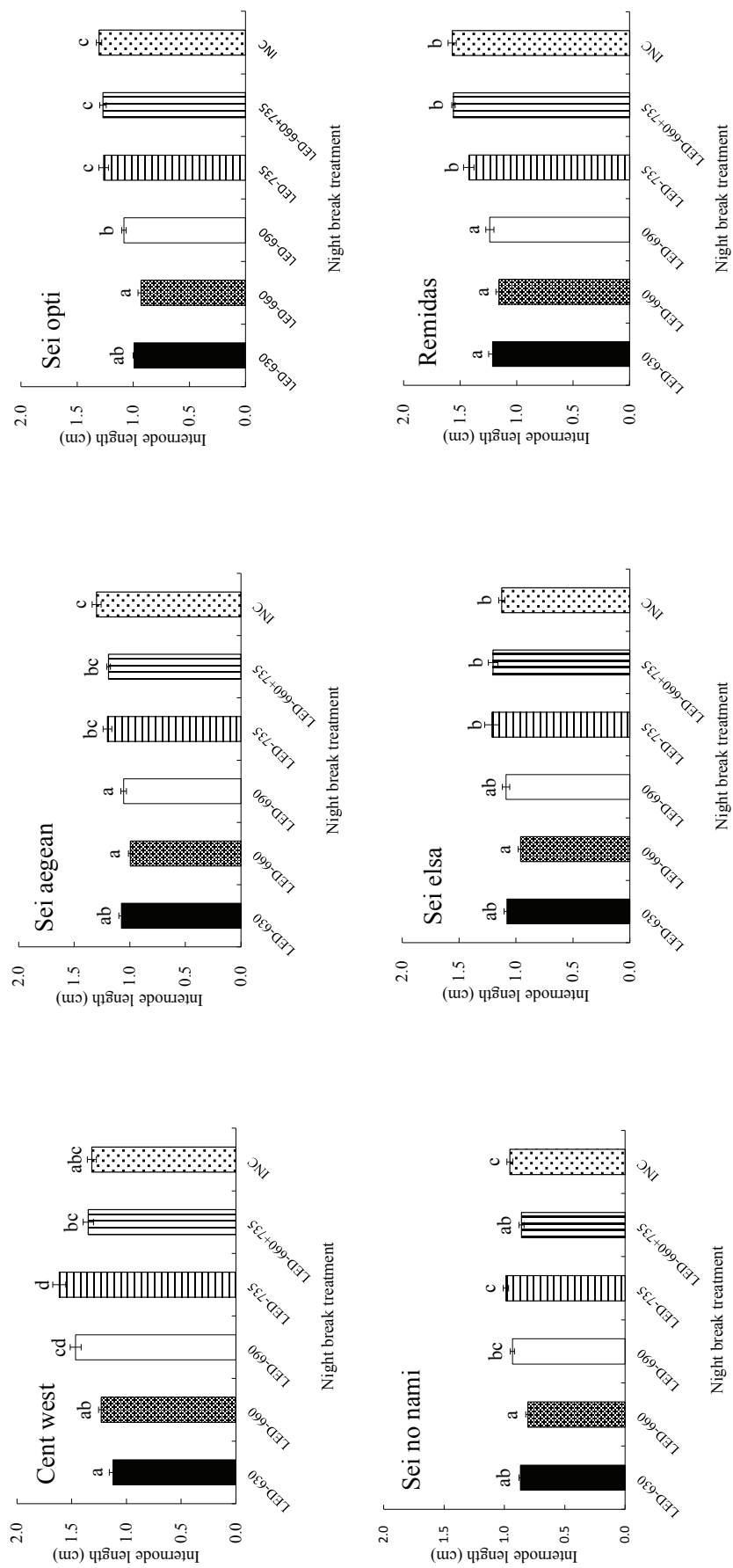


Fig. 2-10 The internode length of cultivars in the sixth week after NB treatments.

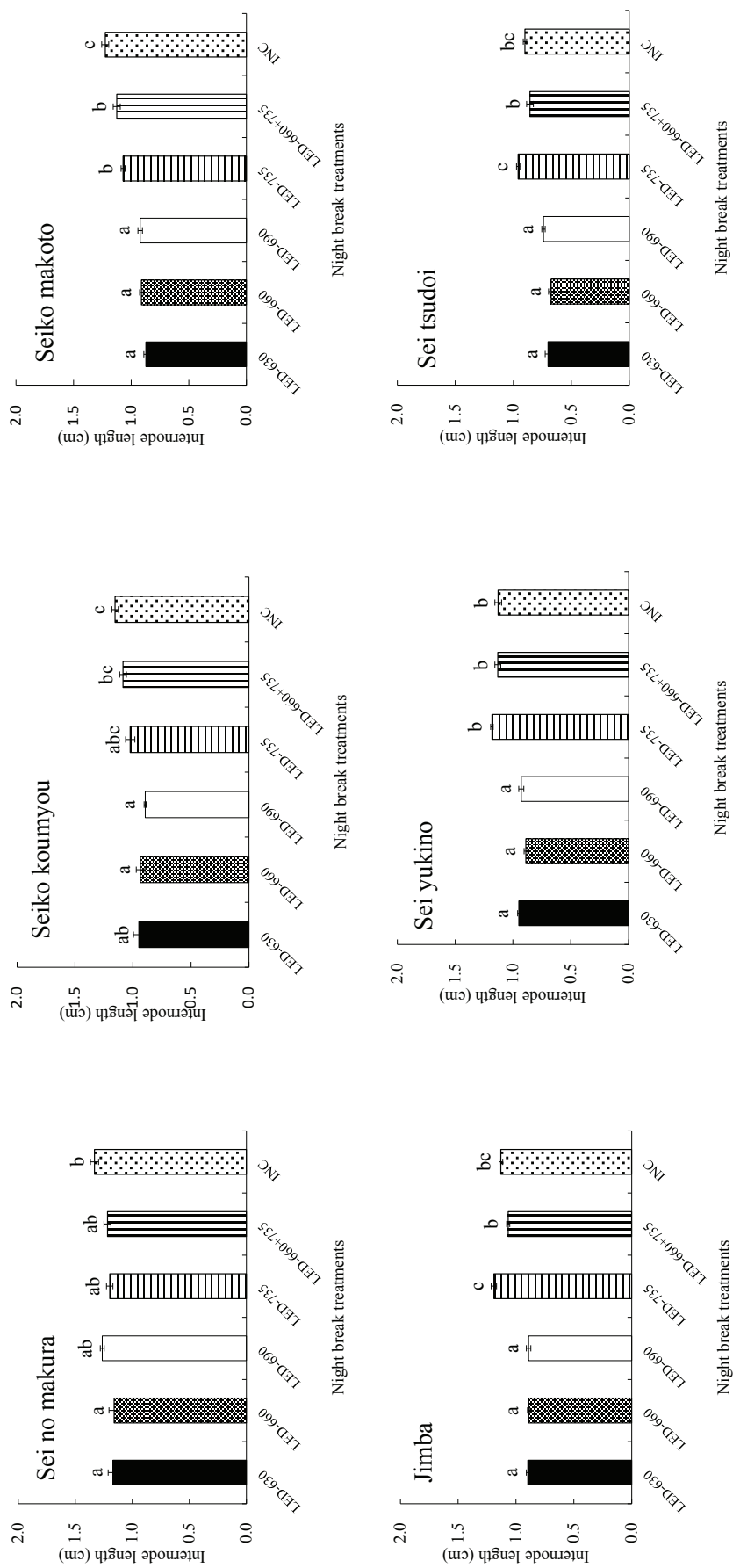


Fig. 2-11 The internode length of cultivars in the sixth week after NB treatments.

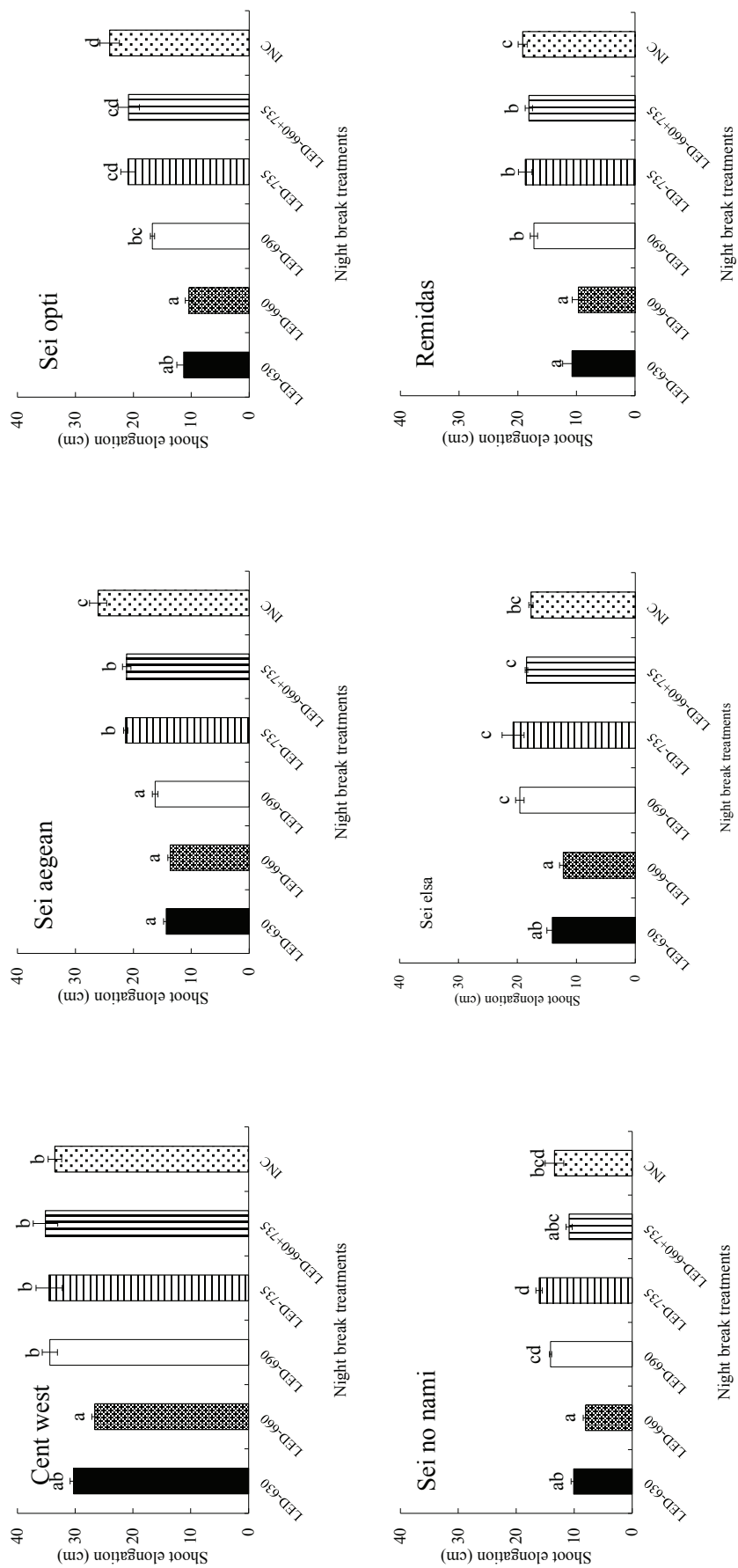


Fig. 2-12 The shoot elongation of cultivars in the sixth week after NB treatments.

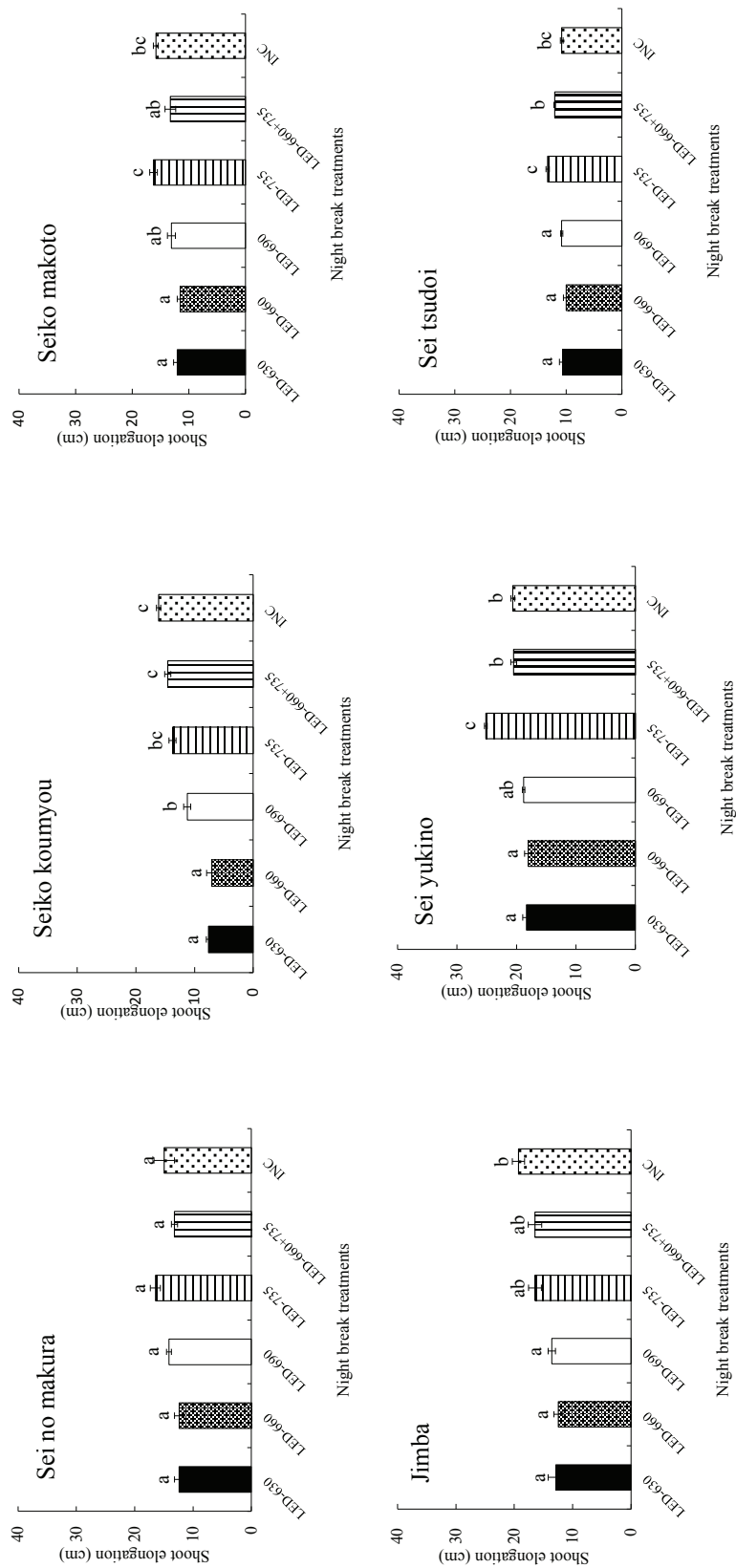


Fig. 2-13 The shoot elongation of cultivars in the sixth week after NB treatments.

Table 2-3. Plant height (cm) of cultivars difference after NB treatments at the 6th week.

Cultivars	Treatments							
	LED-630	LED-660	LED-690	LED-735	LED-660+735	INC		
Cent west	38.04	e 34.36	f 42.16	g 42.3	e 42.92	e 41.28	e	g
Iwa no hakusen	23.6	cd 22.66	d 24.88	cde 31.22	cd 29.56	d 31.84	d	def
Sei aegean	23.4	cd 22.66	d 25.28	de 30.38	cd 30.2	d 35.12	d	f
Sei opti	21.22	bc 20.28	bcd 26.6	def 30.82	cd 30.72	d 33.98	d	ef
Sei no nami	20.28	bc 18.2	abc 24.24	cd 26.22	abc 21.06	a 23.58	a	ab
Sei elsa	23.12	cd 21.26	cd 28.64	f 29.78	cd 27.5	bcd 26.74	bcd	bcd
Remidas	21.68	bc 20.56	bcd 28.14	f 29.62	cd 29	cd 30.06	cd	cdef
Sei no makura	22.98	cd 22.98	d 24.72	cde 27.04	bc 23.8	ab 25.56	ab	bc
Seiko koumyou	17.3	a 16.74	a 20.92	ab 23.44	ab 24.26	abc 25.82	abc	bc
Seiko no makoto	21.44	bc 20.94	cd 22.5	bc 25.66	abc 22.68	ab 25.22	ab	bc
Jimba	23.54	cd 23.08	d 24.2	cd 27.1	bc 27.08	bcd 29.9	bcd	cdef
Sei yukino	26.96	d 26.64	e 27.42	ef 33.76	d 29.1	d 29.24	d	cde
Sei tsudoi	18.2	b 17.52	ab 18.32	a 20.86	a 19.58	a 18.34	a	a

Table 2-4. Leaf number of cultivars difference after NB treatments at the 6th week.

Cultivars	Treatments											
	LED-630	LED-660	LED-690	LED-735	LED-660+735	INC						
Cent west	34	f	28	f	29	ef	26.2	cdef	32	d	31.4	f
Iwa no hakusen	23.4	bcde	23	de	27	def	28.2	ef	26	c	26.4	d
Sei aegean	21.8	bcd	22.8	de	24	bcd	25.4	bcdef	25.4	c	27	e
Sei opti	21.4	bcd	21.8	abcd	24.6	bcd	24.4	abcd	24.2	bc	26	de
Sei no nami	23.4	bcde	22.6	cde	26	bcde	26.6	def	24.6	bc	24.6	cde
Sei elsa	21.4	bcd	22.2	bcde	26.4	cdef	24.6	abcde	23	abc	23.8	bcde
Remidas	17.8	a	17.8	a	22.8	ab	20.8	a	18.6	a	19.2	a
Sei no makura	19.8	abc	20	abc	19.6	a	22.6	abc	19.6	ab	19.2	a
Seiko koumyou	18.4	ab	18	ab	23.4	bc	23	abcd	22.4	abc	22.4	abcd
Seiko no makoto	24.6	cde	23	de	24.4	bcd	24	abcd	20.2	ab	20.6	abc
Jimba	26.2	de	26	def	27.2	def	22.8	abc	25.4	c	26.4	de
Sei yukino	28.4	e	30	f	29.6	f	28.6	f	25.8	c	26	de
Sei tsudoi	26.2	de	26.2	ef	24.8	bcd	21.8	ab	23	abc	20.4	ab

Table 2-5. Internode length (cm) of cultivars difference after NB treatments at the 6th week.

Cultivars	Treatments							
	LED-630	LED-660	LED-690	LED-735	LED-660+735	INC		
Cent west	1.12	1.23	1.46	1.61	1.35	1.32		
Iwa no hakusen	1.01	0.99	0.92	1.11	1.14	1.21		
Sei aegean	1.08	0.99	1.06	1.20	1.19	1.30		
Sei opti	0.99	0.93	1.08	1.26	1.27	1.30		
Sei no nami	0.87	0.81	0.93	0.99	0.86	0.95		
Sei elsa	1.08	0.96	1.09	1.21	1.20	1.12		
Remidas	1.21	1.16	1.24	1.42	1.56	1.57		
Sei no makura	1.17	1.16	1.26	1.20	1.22	1.33		
Seiko koumyou	0.95	0.94	0.89	1.02	1.09	1.15		
Seiko no makoto	0.87	0.91	0.92	1.07	1.13	1.22		
Jimba	0.90	0.89	0.89	1.19	1.07	1.13		
Sei yukino	0.95	0.89	0.93	1.18	1.13	1.13		
Sei tsudoji	0.70	0.67	0.74	0.96	0.86	0.90		

Table 2-6. Shoot elongation (cm) of cultivars difference after NB treatments at the 6th week.

Cultivars	Treatments											
	LED-630	LED-660	LED-690	LED-735	LED-660+735	INC	INC	INC				
Cent west	30.28	d	26.6	f	34.4	i	34.54	f	35.16	f	33.52	h
Iwa no hakusen	14.2	bc	13.26	d	15.48	cdef	21.82	de	20.16	e	22.44	efg
Sei aegean	14.36	bc	13.62	d	16.24	defg	21.34	cde	21.16	e	26.08	g
Sei opti	11.32	ab	10.38	abcd	16.7	efgh	20.92	cde	20.82	e	24.08	fg
Sei no nami	10.1	ab	8.02	ab	14.06	bcde	16.04	abc	10.88	a	13.4	ab
Sei elsa	14.08	bc	12.22	bcd	19.6	h	20.74	bcd	18.46	de	17.7	bcde
Remidas	10.78	ab	9.66	abc	17.24	fgh	18.72	abcd	18.1	cde	19.16	cdef
Sei no makura	12.38	ab	12.38	cd	14.12	bcde	16.44	abcd	13.2	ab	14.96	abc
Seiko koumyou	7.62	a	7.06	a	11.24	ab	13.76	ab	14.58	abcd	16.14	abcd
Seiko no makoto	12.02	ab	11.52	bcd	13.08	abc	16.24	abcd	13.26	abc	15.8	abcd
Jimba	12.9	b	12.44	cd	13.56	abcd	16.46	abcd	16.44	bcde	19.26	cdef
Sei yukino	18.34	c	18.02	e	18.8	gh	25.14	e	20.48	e	20.62	defg
Sei tsudoi	10.66	ab	9.98	abcd	10.78	a	13.32	a	12.04	ab	10.8	a

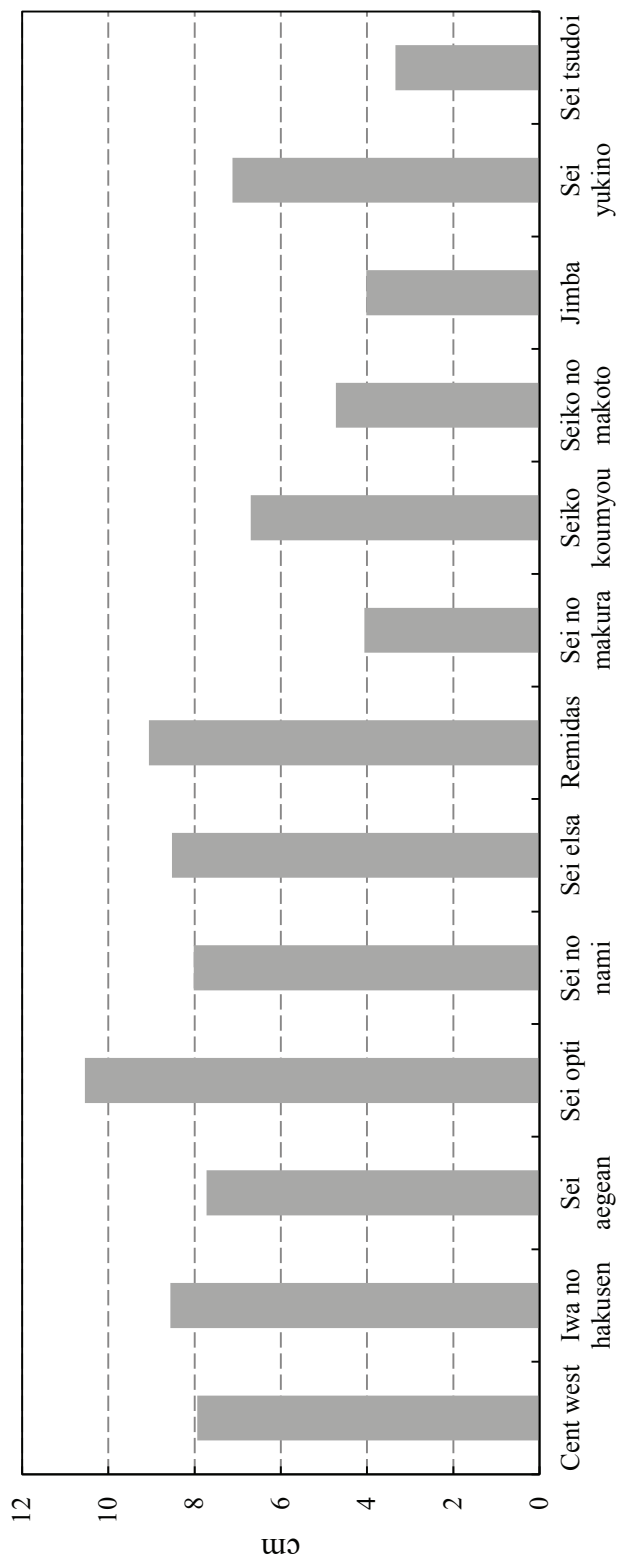


Fig. 2-14 The elongation difference between in LED-735 and in LED-660 of all cultivars. For comparing the shoot elongation effect by FR light and by R light in cultivars, we calculated the results by using the method of subtracting plant height in LED-660 by that in LED-735

Part 3 Night-break Effect of the Lowest Intensities of LED on Floral Bud Differentiation of *Chrysanthemum morifolium* Ramat ‘Jimba’ and ‘Iwa no hakusen’

It was since 1950s that had been applying incandescent (INC) lamp night lighting in chrysanthemum production in Japan. Because of its high electronic consumption and low energy conversion, INC was taken place of by new light source consequentially. Currently, the popularization of LED lamp for the production of chrysanthemum is not prevalent because the effective wavelength and light intensity for inhibition of flowering is still not obvious. For effectively applying LED lamp in chrysanthemum light culture, it is needed to understand that the lowest LED light intensities for inhibition flowering on chrysanthemum. In Part 1, FBD of chrysanthemum ‘Jimba’ was inhibited by LED NB treatments which light peak emission were at 630 nm, 660 nm and 690 nm. In this study, we researched the lowest light intensity at 630 nm and 690 nm that could inhibit the flowering of chrysanthemum to establish a method for effective using of LED lamp.

Experiment 1 Night-break Effect of the Lowest Light Intensity of LED Peak Emission at 630 nm or 690 nm on Floral Bud Differentiation of *Chrysanthemum morifolium* Ramat ‘Jimba’

1. Materials and methods

1.1 Plant materials

Autumn flowering of chrysanthemum cultivar ‘Jimba’ was used as plant material in this study. ‘Jimba’ is a short day plant and flowers in autumn in natural environment. It was a very popular as a main cultivar in chrysanthemum year round production.

1.2 Night break light sources

LED-630 and LED-690 nm (Shibasaki Inc., Japan) were used as the light sources in this study. Light intensities were measured by PS-100 (Apogee Instruments, Inc., USA) 15 cm bellowing the LED lamps. The light intensity was measured at fifty sites of that surface and average was calculated in every treatments.

There were 4 NB treatments of LED-630 which peak emission were 0.050, 0.083, 0.101 and 0.150 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ respectively (Fig. 3-1). And those 4 treatments were abbreviated to 630_{p0.050}, 630_{p0.083}, 630_{p0.101} and 630_{p0.150} respectively. There were also 4 NB treatments of LED-690 which peak emission were 0.160, 0.260, 0.350 and 0.460 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ respectively (Fig. 3-2). And those 4 treatments were abbreviated to 690_{p0.160}, 690_{p0.260}, 690_{p0.350} and 690_{p0.460} respectively. The distribution of light intensities was showed in Fig. 3-3 and Fig. 3-4.

1.3 Methods

Shoot cuttings 'Jimba' were planted in the plastic case (size of 50 cm x 35 cm x 7.5 cm) which was filled with planting material BM2 (Berger peat moss Ltd., Canada). Plants had been growing in the growth chamber with constant temperature at 23 °C and 12 h lighting (08:00~20:00) with white fluorescent lamp (40W FLR40SW/M; NEC Inc., Japan) for 6 weeks. The NB treatments were carried out in the night time (23:00~05:00) every day. Every NB treatment contained 50 plants. The management of fertilizing and irrigation was as same as Part 1.

1.2 Data collection and analysis

The data collection and analysis was as same as Part 1.

2. Results

2.1 FBD

In LED-630 treatments, FBD was not inhibited at treatments of $630_{p0.050}$ and $630_{p0.083}$ and the indexes of FBD were 0.8 and 0.4 in the 6th week. FBD was delayed at $630_{p0.100}$ and inhibited obviously at $630_{p0.150}$. Their indexes of FBD were 0.280 and 0.030 in the 6th week respectively. The index of FBD was significant lower in $630_{p0.150}$ than that in $630_{p0.050}$ and $630_{p0.083}$ (Fig. 3-5).

In LED-690 treatments, indexes of FBD were 0.52 and 0.34 at treatments of $690_{p0.160}$ and $690_{p0.260}$ respectively in the 6th week. The FBD was delayed in $690_{p0.350}$ and inhibited in $690_{p0.460}$. Their index of FBD was 0.200 and 0.067 in 6th week respectively. The index of FBD was significant lower in $690_{p0.460}$ than that in $690_{p0.160}$ and $690_{p0.260}$ (Fig. 3-5).

2.2 Plant height

In LED-630 treatments, plant heights were 19.51, 21.71, 18.58 and 16.78cm in $630_{p0.050}$, $630_{p0.083}$, $630_{p0.101}$ and $630_{p0.150}$ respectively. It was the significant higher in $630_{p0.083}$ than in $630_{p0.150}$ (Fig.3-6).

In LED-690- treatments, plant heights were 19.14, 19.92, 19.97 and 20.85 cm in $690_{p0.160}$, $690_{p0.260}$, $690_{p0.350}$ and $690_{p0.460}$ respectively. It was the significant highest in $690_{p0.460}$ than in $690_{p0.160}$ (Fig.3-6).

2.3 Leaf number

In LED-630 treatments, leaf numbers were 26.39, 28.15, 27.13 and 25.60 in $630_{p0.050}$, $630_{p0.083}$, $630_{p0.101}$ and $630_{p0.150}$ respectively. It was significant more in $630_{p0.083}$ than in $630_{p0.150}$.

In LED-690 treatments, leaf numbers were 22.87, 23.63, 24.62 and 24.85 in $690_{p0.160}$, $690_{p0.260}$, $690_{p0.350}$ and $690_{p0.460}$ respectively. It was significant more in $690_{p0.460}$ than that in $690_{p0.160}$ (Fig.3-7).

2.4 Internode length

In LED-630 treatments, internode lengths were 0.74, 0.77, 0.69 and 0.66 cm in $630_{p0.050}$, $630_{p0.083}$, $630_{p0.101}$ and $630_{p0.150}$ respectively. It was significant longer in $630_{p0.083}$ than that in $630_{p0.150}$ (Fig.3-8).

In LED-690 treatments, internode lengths were 0.84, 0.84, 0.81 and 0.84 cm in $690_{p0.160}$, $690_{p0.260}$, $690_{p0.350}$ and $690_{p0.460}$ respectively. There was no difference among these treatments (Fig.3-8).

2.5 Shoot elongation

In LED-630 treatments, shoot elongations were 12.81, 15.67, 11.87 and 10.51 cm in $630_{p0.050}$, $630_{p0.083}$, $630_{p0.101}$ and $630_{p0.150}$ respectively. It was the significant longest in $630_{p0.083}$ than that in other treatments. It was shortest in $630_{p0.150}$ and significant shorter than that in $630_{p0.050}$ and $630_{p0.083}$.

In LED-690 treatments, shoot elongations were 18.90, 19.89, 19.95 and 20.72 cm in $690_{p0.160}$, $690_{p0.260}$, $690_{p0.350}$ and $690_{p0.460}$ respectively. It was the longest in $690_{p0.460}$ and significant longer than that in $690_{p0.160}$.

3. Discussion

In light intensities of LED-630 and LED-690, only $630_{p0.150}$ and $690_{p0.460}$ could inhibit FBD of 'Jimba'. It was showed the relation between FBD and light intensity of LED-630 or LED-690 (Fig. 3-10). The inhibition level of FBD had depended on night break light intensity. High light intensity had higher inhibition ability on FBD than low light intensity. For realizing index of FBD less than 0.2, at least it was needed above $0.150 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ of LED-630 and above $0.460 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ of LED-690. LED light of 630 nm had more effective inhibition of

flowering than that of 690 nm with small amount of electric energy.

In ordinary chrysanthemum production, it was needed 50 lux of INC NB lighting for inhibition flowering (Kimura, 1974). Ishikura (2009) reported that R light LED could inhibition flowering of autumn-flowering type chrysanthemum at the light intensity 1/2 of the INC lamp. In this study, we reported that light intensity (photon flux irradiance) of LED-630 peak emission at $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ and LED-690 peak emission at $0.46\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ could inhibit flowering of autumn-flowering type chrysanthemum.

In treatments of LED-630, it was showed that plant height high was higher in low light intensity treatment than that of in high light intensity treatment, except for 630_{p0.083}. The results were opposite in treatments of LED-690 and plant height was higher in high light intensity treatments than that of in low light intensity treatments. It was showed the relation between the plant height and LED light intensity in Fig. 3-11. The plant height was slightly reduced according the increased light intensity of LED-630. But it did not change according the increased light intensity of LED-690.

Although there was significant difference in leaf number, the results were very close among the treatments. It was showed the relation between the leaf number and LED light intensity in Fig. 3-12. The leaf number was slightly increased according the increased light intensity of LED-630 and slightly decreased according the increased light intensity of LED-690. But the relativity of leaf number and light intensity was very low. It was suggested that leaf number was not related to the light intensity.

The relation between internode length and light intensity was showed in Fig. 3-13. Internode length was showed decreased tendency according the increased light intensity of LED-630 and increased tendency according the increased light intensity of LED-690.

The relation between elongation and light intensity was showed in Fig. 3-14. Shoot

elongation was showed decreased tendency according the increased light intensity of LED-630 and no tendency according the increased light intensity of LED-690.

Because of no relation between the leaf number and light intensity, effect of the light intensity on shoot growth was the shoot elongation which was calculated by plant height and leaf number. It was suggested that R light of LED-630 NB lighting had little negative effect on internode length and shoot elongation. Although LED-690 of NB lighting had little positive effect on internode length, it had no relation in shoot elongation. It was suggested that the effect on shoot growth by LED-690 of NB was very weak.

Experiment 2 Night-break Effect of the Lowest Light Intensity of LED Mixed Red and Far-red Light on Inhibition of *Chrysanthemum* ‘Iwa no hakusen’

In Part 1, R light of LED NB could not inhibit the FBD in ‘Iwa no hakusen’, but combination of R and FR light of LED NB inhibited it. In experiment 1 of Part 3, it was confirmed that photon flux irradiance peak over $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ could inhibit FBD of ‘Jimba’ in R light of LED-630 nm. In this experiment, we studied the FR light intensity of NB effect on ‘Iwa no hakusen’ by applying LED-735 irradiating with R light of LED-630 together.

1 Materials and methods

1.1 Plant materials

Chrysanthemum morifolium ‘Iwa no hakusen’ was used as the plant material. It was classified summer-to-autumn-flowering type, and flowered at the end of June in natural condition and was a popular cultivar in Japan.

1.2 Night break light sources

Two kinds of LED lamp with different wavelength (Shibasaki Inc., Japan) had been used. One was R light LED lamp with peak emission at 630 nm, and other was FR light LED lamp with peak emission at 735 nm. R light intensity of LED-630 which peak emission were at $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$, and FR light intensity of LED-735 which peak emission were at 0.05, 0.10, 0.15 and $0.20 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. Thus, four kinds of treatments combined R light and different intensity of FR light were used for NB treatment; 0.15+0.05 (LED-630 peak emission at $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ + LED-735 peak emission at $0.05 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$), 0.15+0.10, 0.15+0.15 and 0.15+0.20. The distance between LED lamps to the top of the plants was kept in 15 cm constant according to plant growth. All the light intensities were measured by PS-100 (Apogee Instruments, Inc., USA) 15 cm bellowing the LED lamps (Fig. 3-15, Table 3-1).

1.3 Methods

Shoot cuttings were planted in the plastic case (size of 50 cm x 35 cm x 7.5 cm) which was filled with planting material BM2 (Berger peat moss Ltd., Canada). Plants had been growing in the growth chamber with constant temperature at 23 °C and 12 h lighting (08:00~20:00) with white fluorescent lamp (40W FLR40SW/M; NEC Inc., Japan) for 6 weeks. The NB treatments were carried out in the night time (23:00~05:00) every day. Every NB treatment contained 50 plants. Fertilizer solution (N:P₂O₅:K₂O=6.5:6:19, Hyponex Co., Ltd, Japan) diluted at 0.05% was supplied to plants every week. Five plants from each treatment were chose every week and measured plant height and number of leaves, and FBD was investigated by microscope.

1.4 Data collection and analysis

The data collection and analysis was as same as Part 1.

2. Results

The results were showed that FBD of 'Iwa no hakusen' could not be inhibited by the treatments of 0.15+0.05 and 0.15+0.10, and index of FBD reached to 0.63 and 0.51 respectively in the 6th week (Fig. 3-16). Treatments of 0.15+0.15 and 0.15+0.20 were showed low FBD index, which were 0.26 and 0.17 respectively. FBD index at 0.15+0.05 and 0.15+0.10 were significant higher than those at 0.15+0.15 and 0.15+0.20.

The results of shoot length were 29.8, 31.8, 31.1 and 30 cm in the 6th week respectively (Fig. 3-17) and there was no significant difference in shoot length. Leaf number were 28.6, 30.0, 28.1 and 27.3 in the 6th week respectively (Fig. 3-18), and leaf number in 0.15+0.10 has significant difference to that in 0.15+0.20.

Internode lengths were the 1.11, 1.12, 1.07 and 1.03 cm in the 6th week respectively. Internode length in treatment 0.15+0.05 was significant shortest than that in other treatments (Fig. 3-19). Shoot elongations were 23.32, 24.41, 25.19 and 23.24 cm in the 6th week respectively. There was no significant difference among these treatments (Fig. 3-20).

3. Discussion

According to stronger intensity of FR light, index of FBD of 'Iwa no hakusen' was decreasing at treatment from 0.15+0.05 to 0.15+0.20. As R light intensity (630 nm) was constant at $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. It was showed an inhibitory effect on the FBD on 'Iwa no hakusen' by the adding FR light. In part 1, FR light had no effect on FBD in 'Jimba', and Sumitomo et al. (2012) also showed that FR light had no effect on flowering in 'Reagan'. 'Jimba' and 'Reagan' belonged to autumn-flowering type chrysanthemum, while 'Iwa no hakusen' belonged to summer-to-autumn-flowering type chrysanthemum. It was indicated that FR light NB might have different effect on different flowering type of chrysanthemum. The difference might occur in flowering related interior genes and proteins that responded to light

signal and it was still unclear.

FR light promoted shoot elongation (Rajapakse et al., 1993). In this experiment, although the internode length was significant longer in treatment 0.15+0.20 than that in 0.15+0.05, the data were very close. And there was no significant difference in shoot elongation among four treatments of light intensity of FR light. It was suggested that FR light intensity of peak emission at $0.05 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ of 735 nm had the same elongation effect of intensity of $0.20 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. The difference of FR light intensity between 0.05 and $0.20 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ was not enough to cause significant difference of shoot elongation in 'Iwa no hakusen'. Although leaf number in treatment 0.15+0.10 was significant more than that in treatment 0.15+0.20, it was not consider that there was regularity among these treatments.

In this experiment, i FBD on cultivar 'Iwa no hakusen' was inhibited by NB of R+FR light. Hakuzan and Nagayoshi (2013) also showed a similar result in 'Iwa no hakusen'. Although FBD could not be inhibited by single R light on 'Iwa no hakusen', R light combined with FR light inhibiting FBD. The inhibition effect of using R+FR light was better than those of using R or FR light only. FR light intensity which combined with R light with $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ had enough inhibition effect of FBD of 'Iwa no hakusen' at higher than over $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. In practical using LED lamp, growers should manage the installation of the number of LED lamps and the height to the top of chrysanthemum plants for keeping these R and FR light intensity.

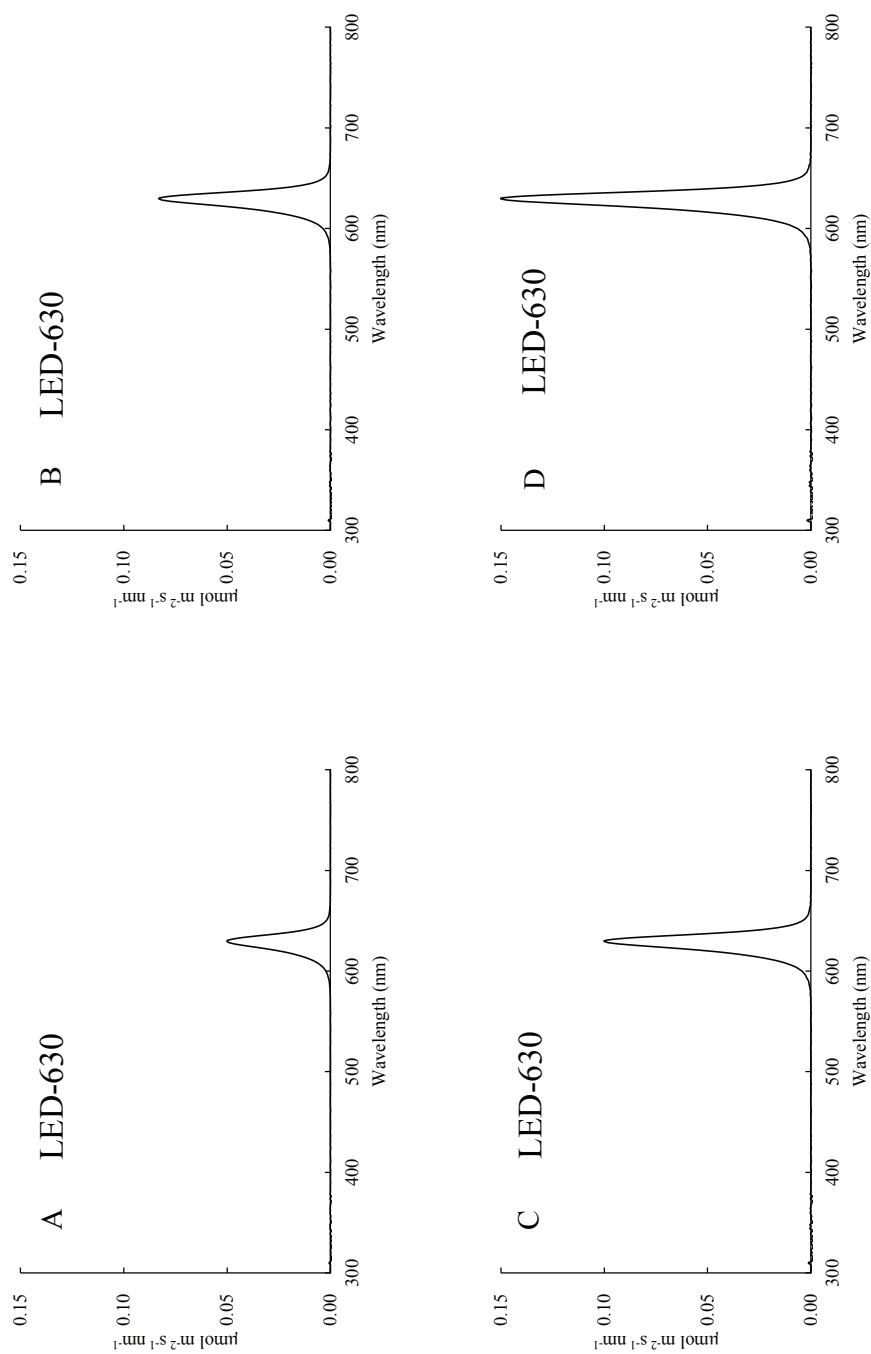


Fig. 3-1 Light intensity of LED-630. Light intensities of four NB treatments in peak emission were 0.050, 0.083, 0.100 and 0.150 $\mu\text{mol m}^{-2} \text{s}^{-1} \text{nm}^{-1}$ from A to D respectively.

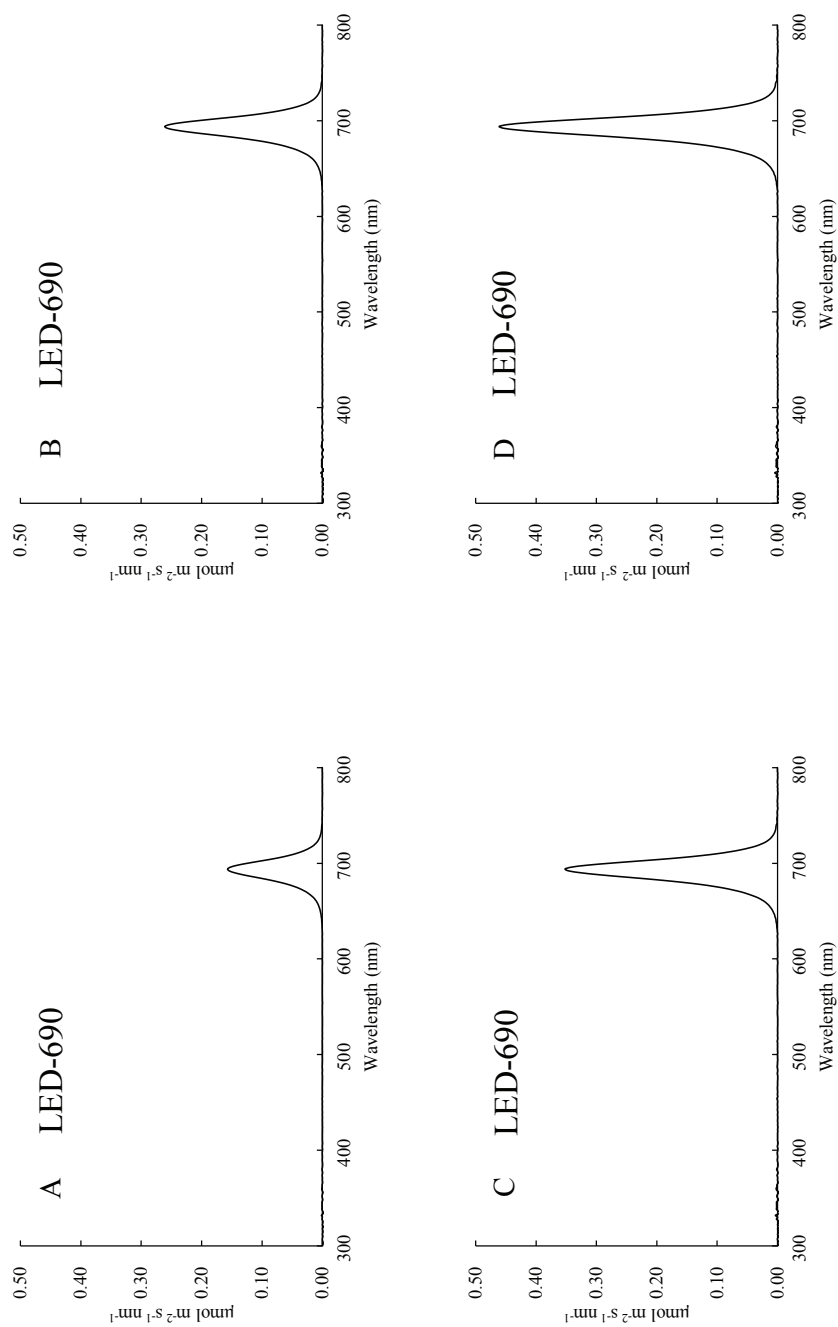


Fig. 3-2 Light intensity of 690 nm LED lamp. Light intensities of four NB treatments in peak emission were 0.157, 0.261, 0.352 and 0.461 $\mu\text{mol m}^{-2} \text{s}^{-1} \text{nm}^{-1}$ from A to D respectively

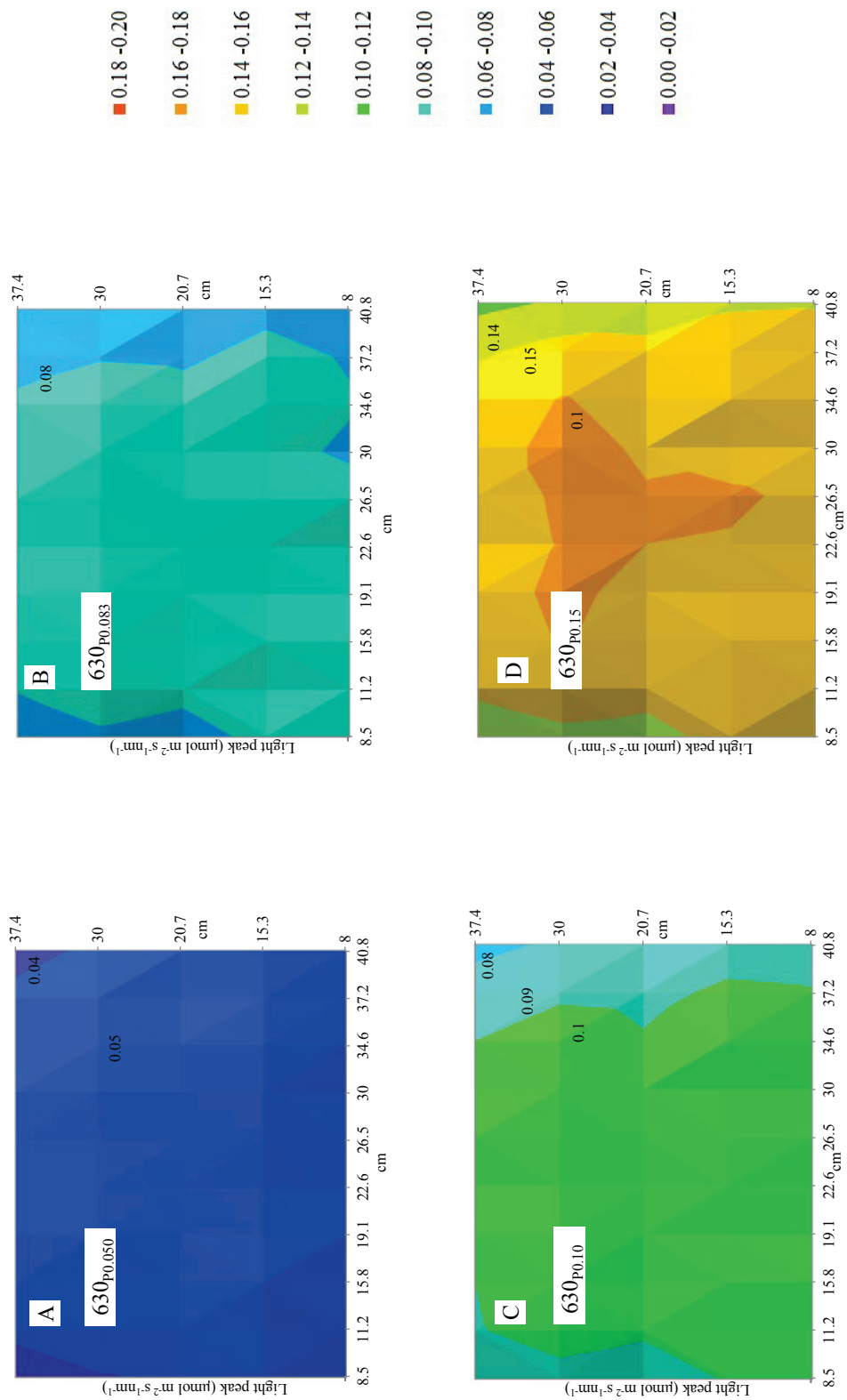


Fig. 3-3 Light intensity distribution in peak emission at LED-630 treatments were measured in plant growth chamber 15 cm below the LED pattern. Light intensity distribution of treatments of $630_{p0.050}$, $630_{p0.083}$, $630_{p0.10}$ and $630_{p0.150}$ were showed from A to D respectively.

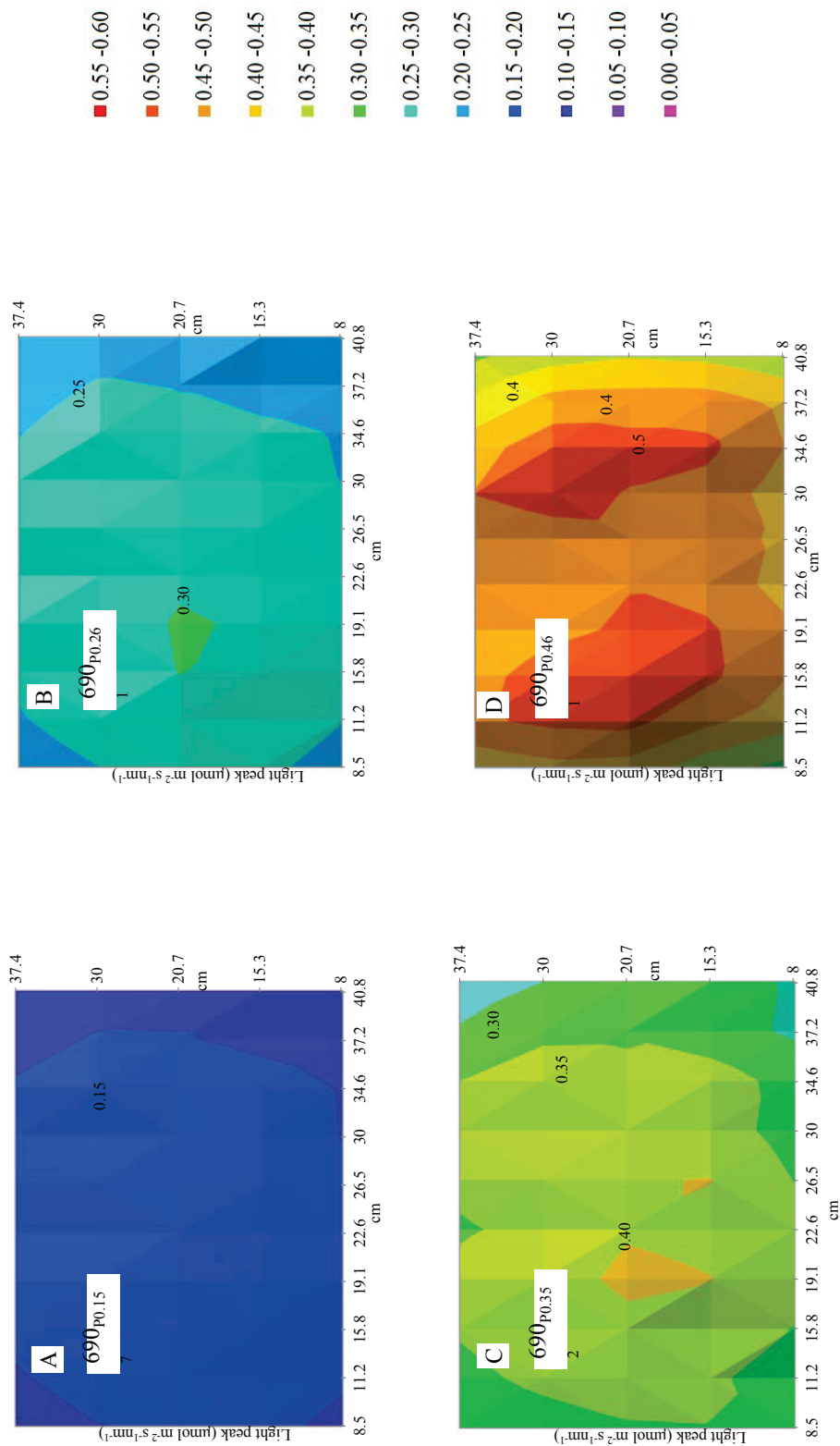


Fig. 3-4 Light intensity distribution in peak emission at 690 nm were measured in plant growth chamber 15 cm below the LED pattern. Light intensity distribution of treatments of 690_{P0.15}, 690_{P0.26}, 690_{P0.35}, and 690_{P0.46} were showed from A to D respectively.

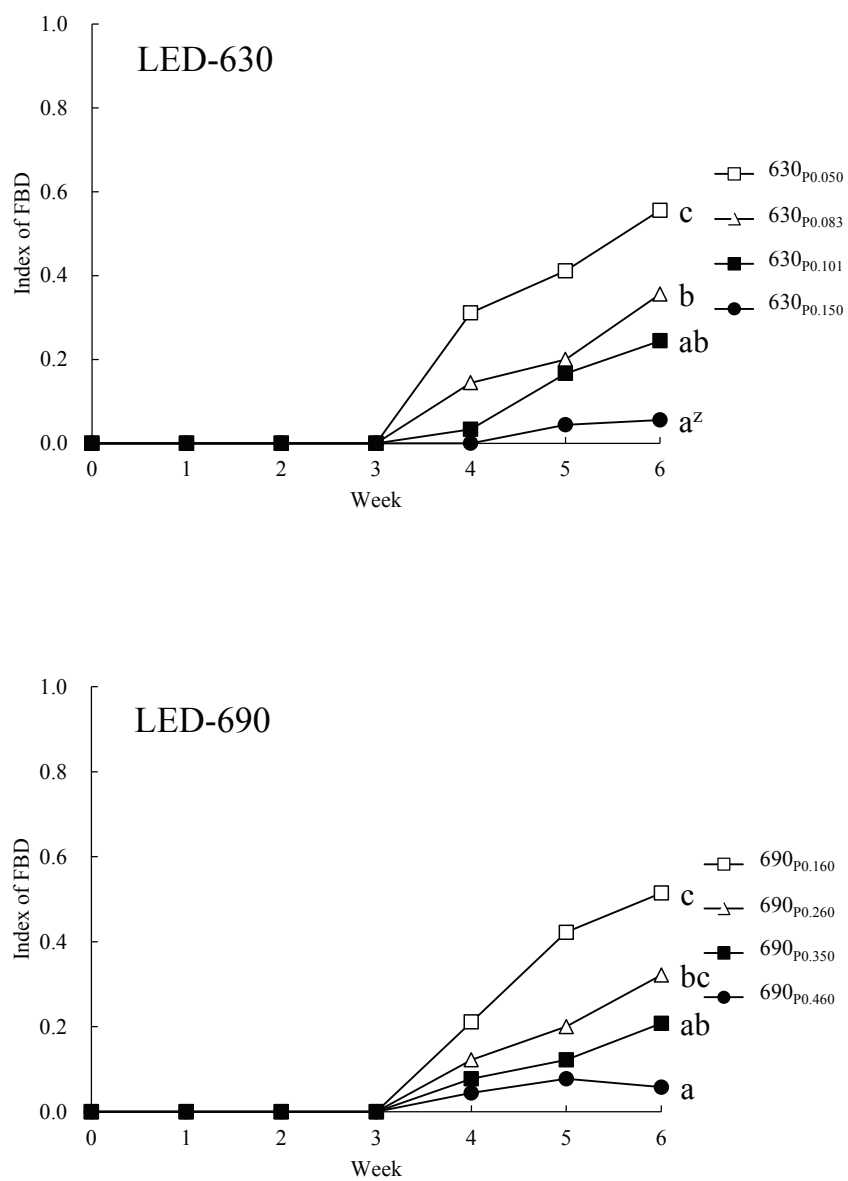


Fig. 3-5 The index of FBD of 'Jimba' from 0 week to the sixth week after LED-630 and LED-690 light intensity NB treatments.

^z: Difference letters indicate significant difference in index of FBD at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

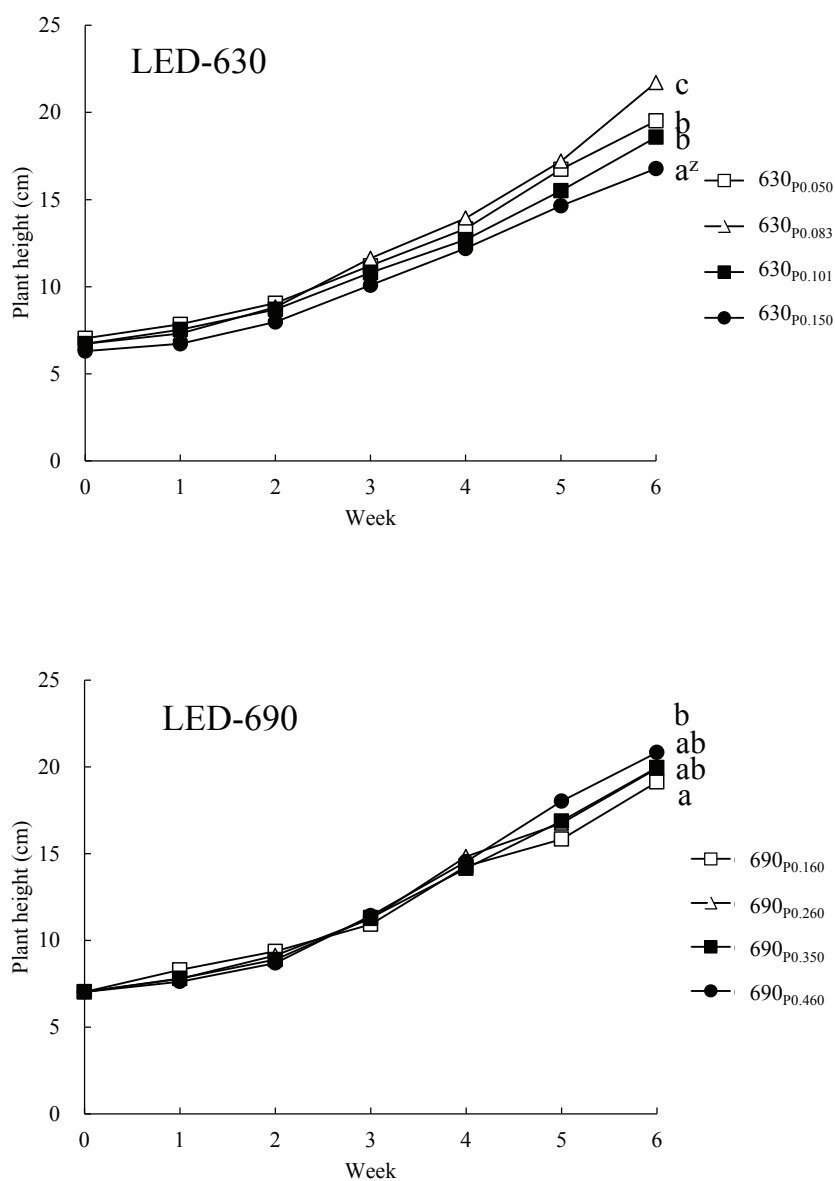


Fig. 3-6 The shoot length of 'Jimba' from 0 week to the sixth week after LED-630 and LED-690 light intensity NB treatments.

^z: Difference letters indicate significant difference in plant height at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

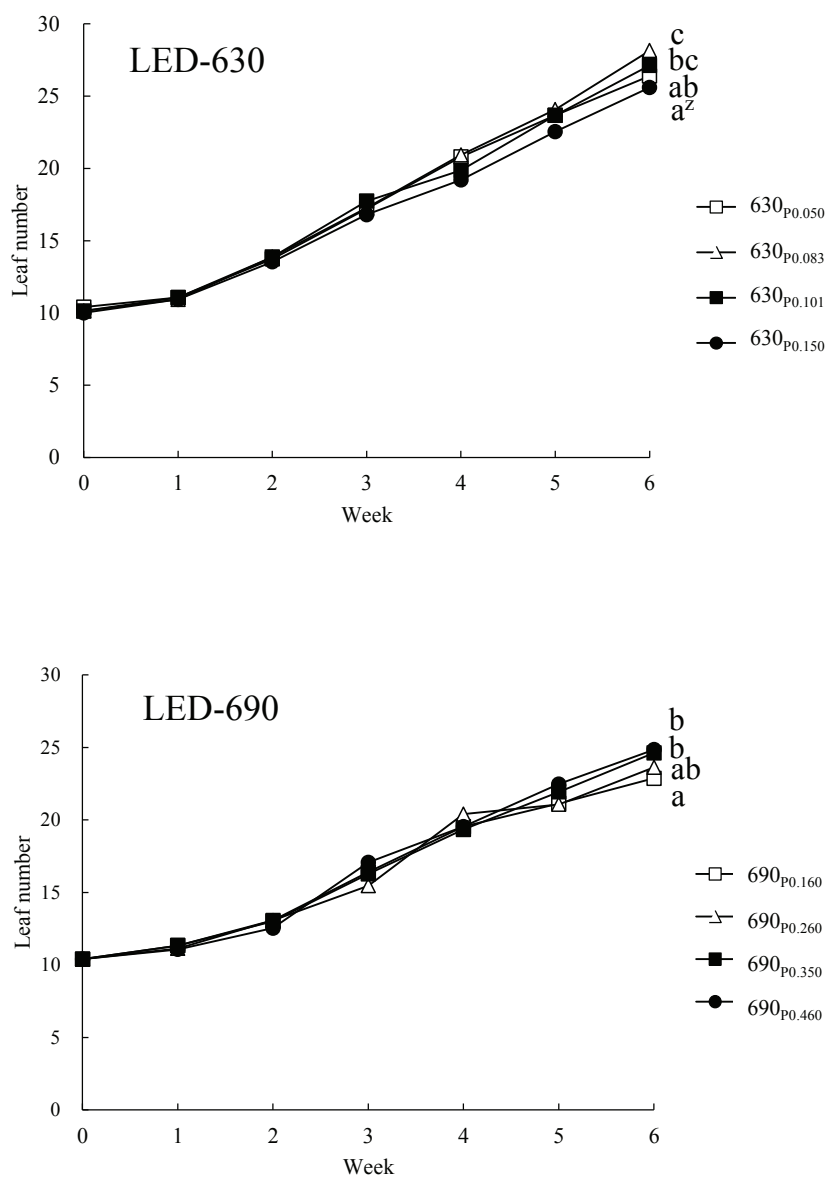


Fig. 3-7 The leaf number of 'Jimba' from 0 week to the sixth week after LED-630 and LED-690 light intensity NB treatments.

^z: Difference letters indicate significant difference in leaf number at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

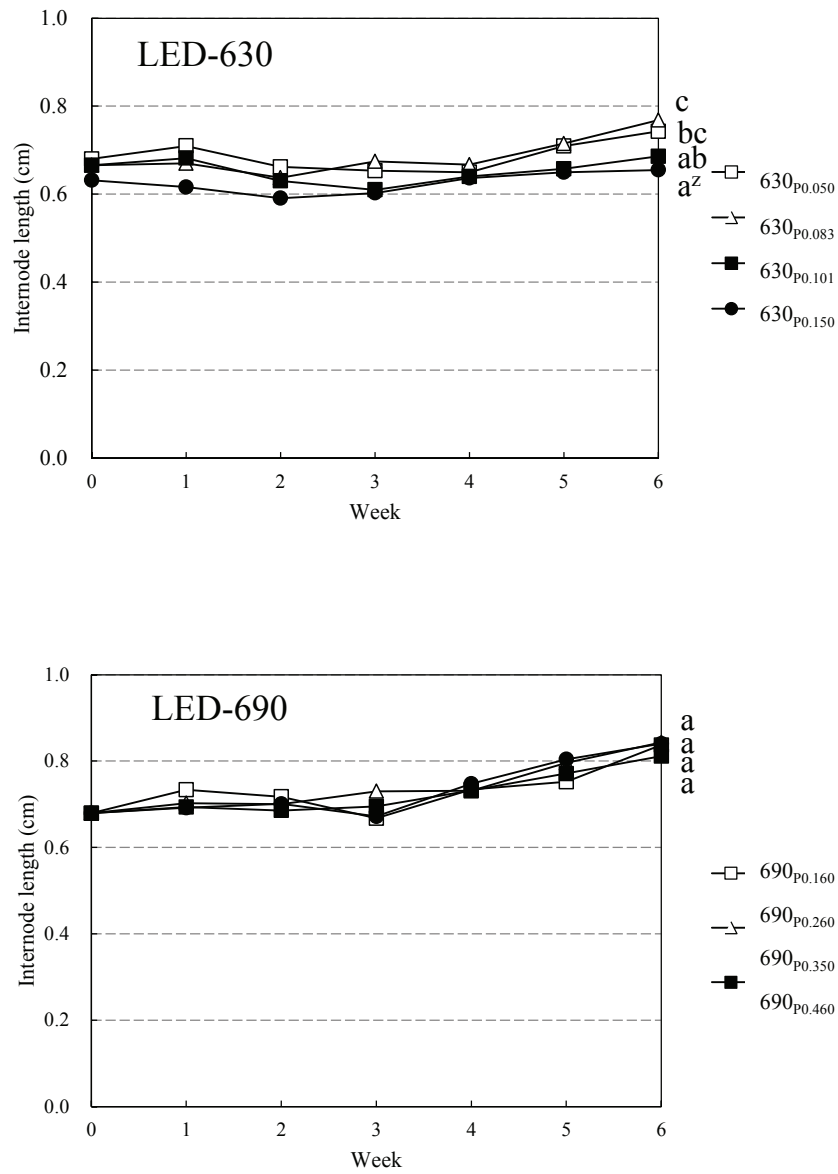


Fig. 3-8 The internode length of 'Jimba' from 0 week to the sixth week after LED-630 and LED-690 light intensity NB treatments.

^z: Difference letters indicate significant difference in internode length at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

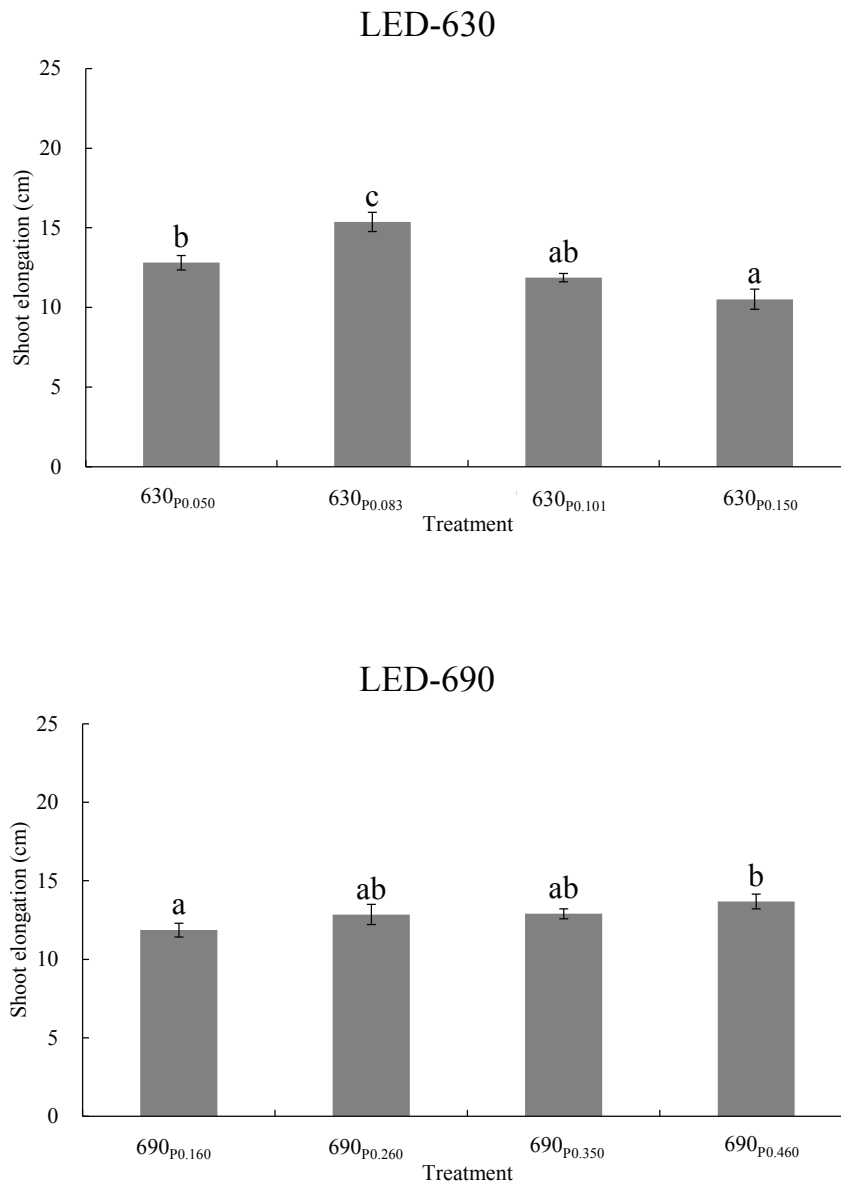


Fig. 3-9 The shoot elongation of 'Jimba' in the sixth week after LED-630 and LED-690 light intensity NB treatments.

^z: Difference letters indicate significant difference in shoot elongation at the sixth week at $P < 0.05$ by Tukey's multiple comparison test.

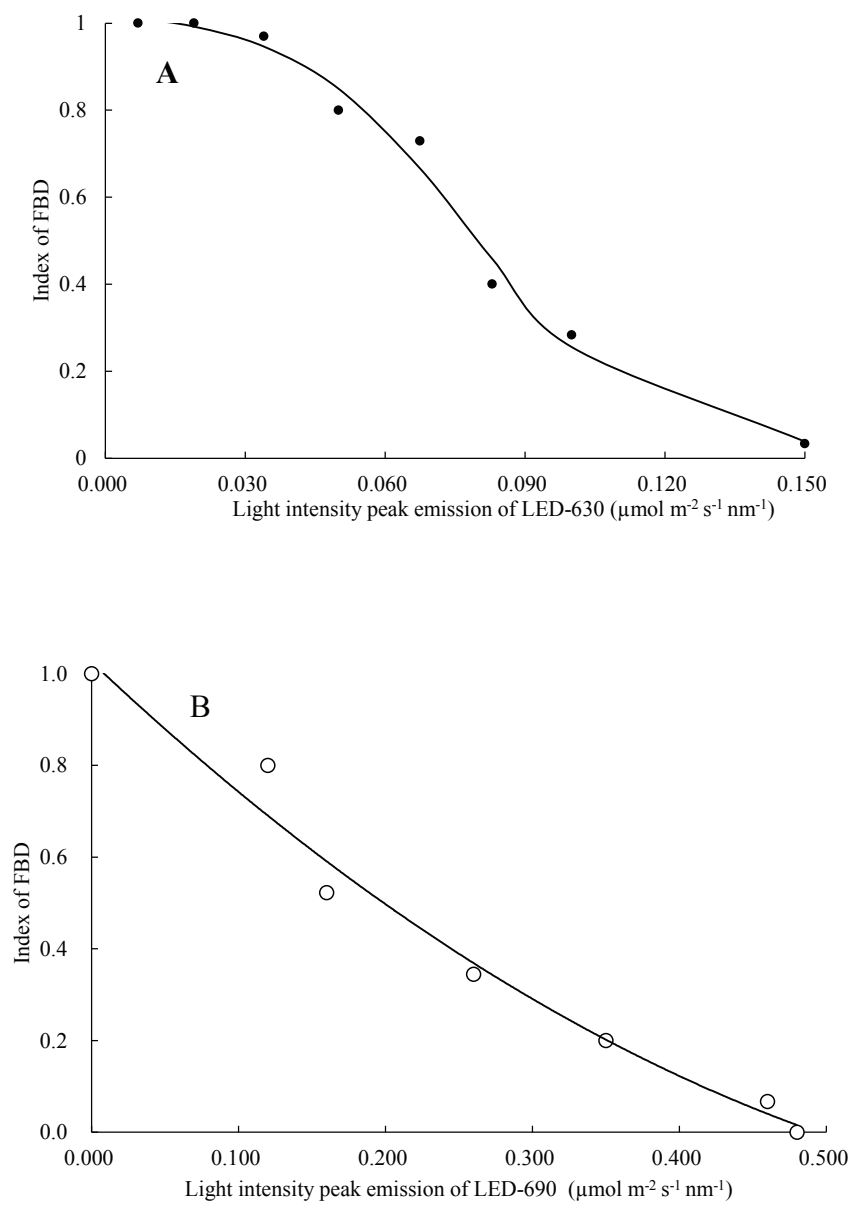


Fig. 3-10 The relation between index of FBD and LED-630 light intensity (A) or LED-690 light intensity (B).

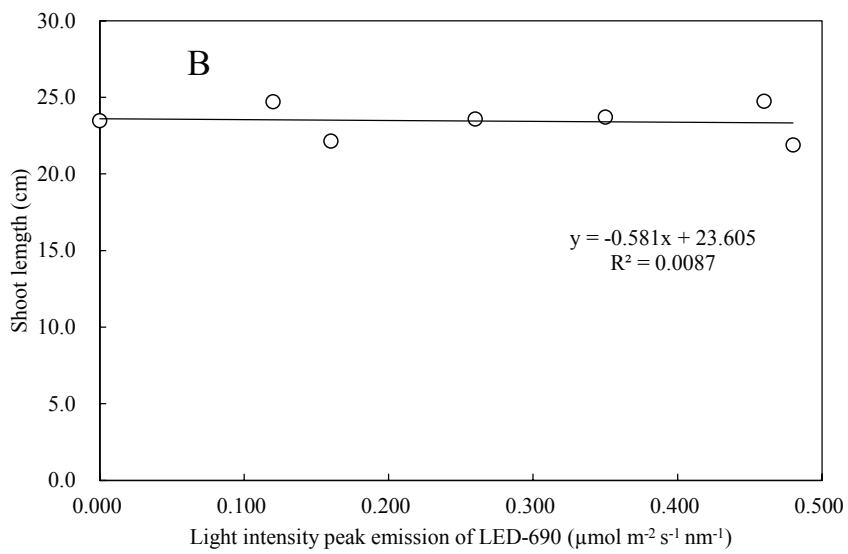
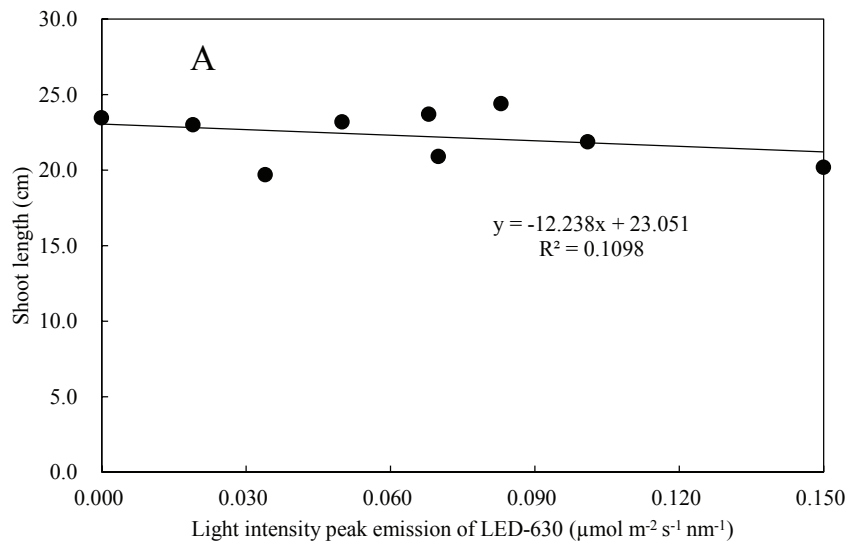


Fig. 3-11 The relation between shoot length and LED-630 light intensity (A) or LED-690 light intensity (B).

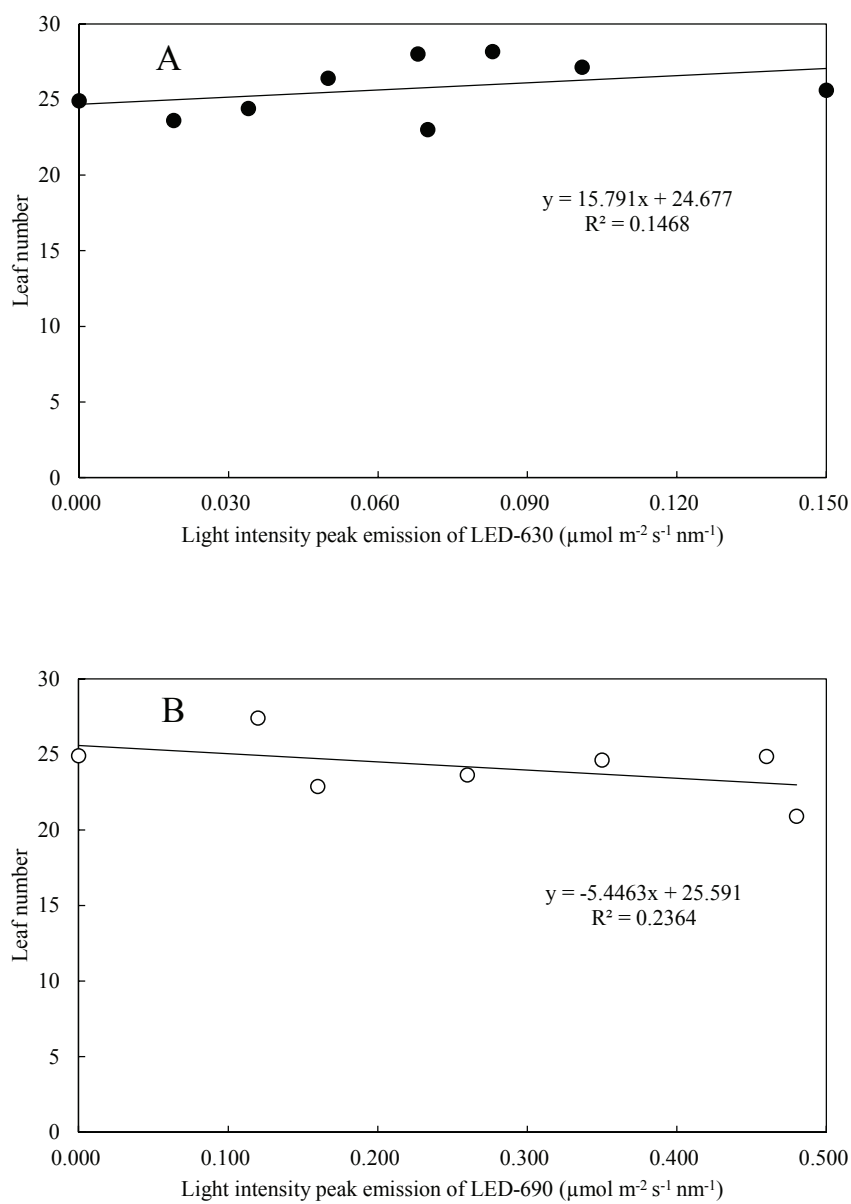


Fig. 3-12 The relation between leaf number and LED-630 light intensity (A) or LED-690 nm light intensity (B).

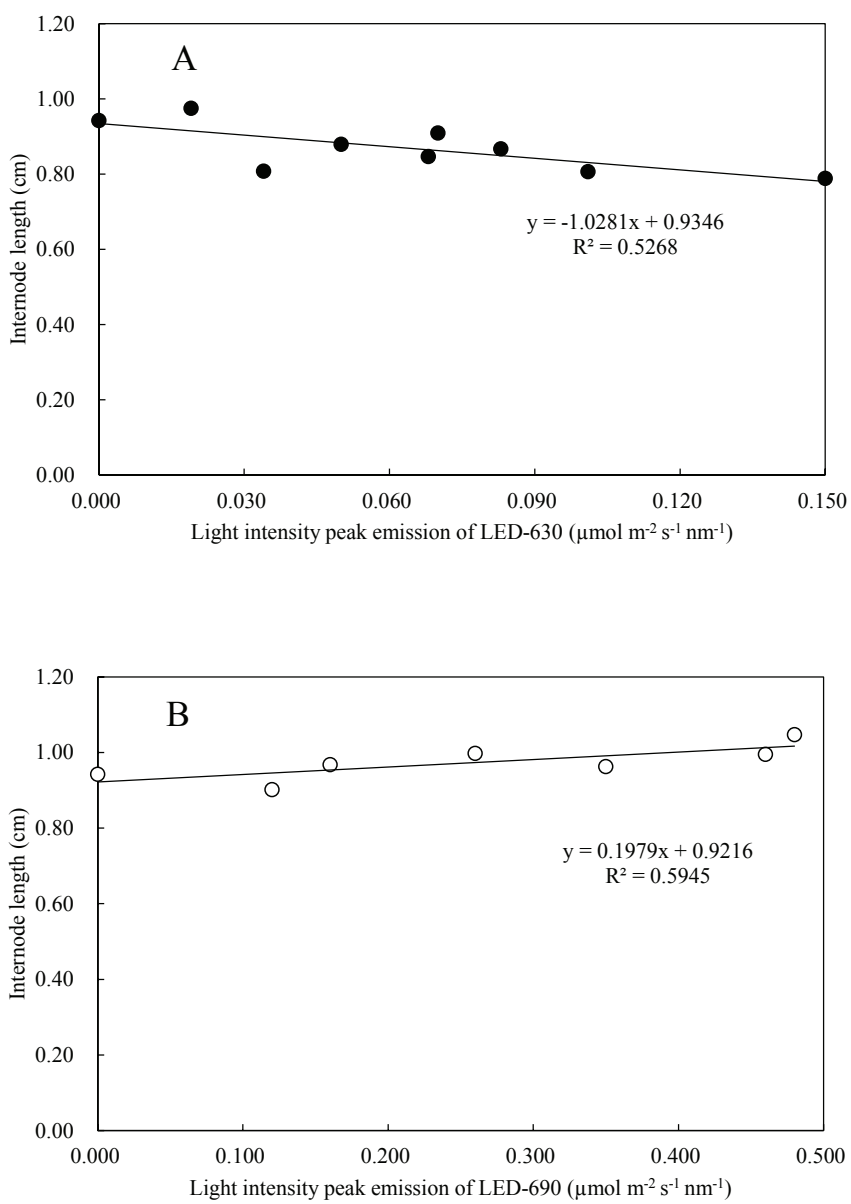


Fig. 3-13 The relation between internode length and LED-630 light intensity (A) or LED-690 light intensity (B).

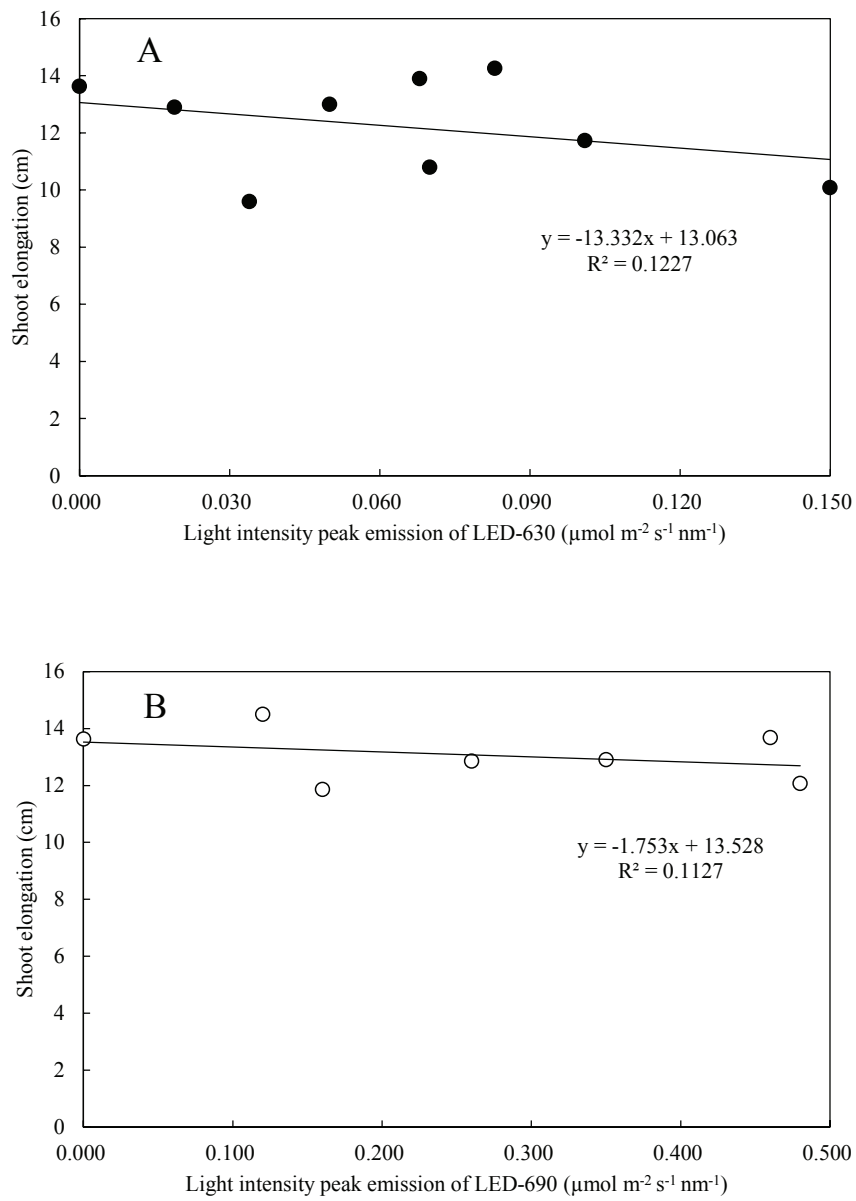
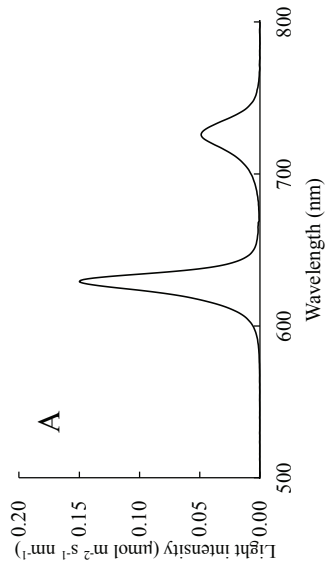
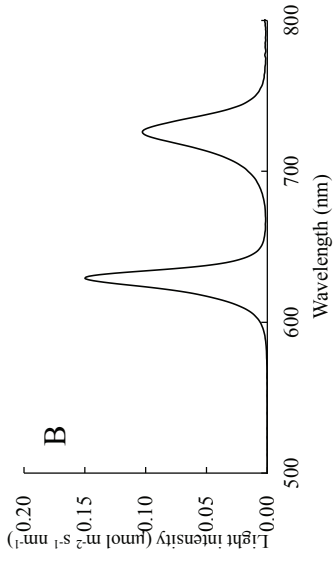


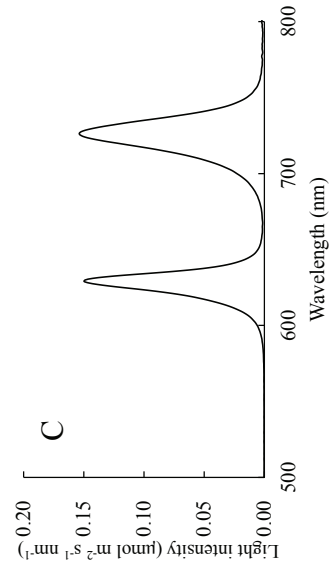
Fig. 3-14 The relation between shoot elongation and LED-630 light intensity (A) or LED-690 light intensity (B).



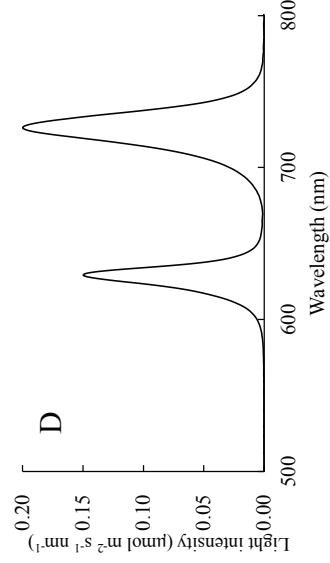
Treatment 0.15 + 0.05



Treatment 0.15 + 0.10



Treatment 0.15 + 0.15



Treatment 0.15 + 0.20

Intensity of combination of LED-630 and LED-735. Light intensities of four NB treatments which light peak emission were 0.15 + 0.05, 0.15 + 0.10, 0.15 + 0.15, and 0.15 + 0.20 $\mu\text{mol m}^{-2} \cdot \text{s}^{-1} \cdot \text{nm}^{-1}$ from A to D respectively.

Table 3-1. R and FR light photon flux density in treatments

Treatment (R+FR)	Photon flux density ^z ($\mu\text{mol m}^{-2} \text{s}^{-1}$)		
	R	FR	Total
0.15 + 0.05	2.82	1.30	4.12
0.15 + 0.10	2.90	2.79	5.69
0.15 + 0.15	2.97	4.15	7.12
0.15 + 0.20	3.01	5.30	8.31

^z: Photon flux density of R light was calculated from 600 nm to 699 nm and FR light was from 700 nm to 799 nm.

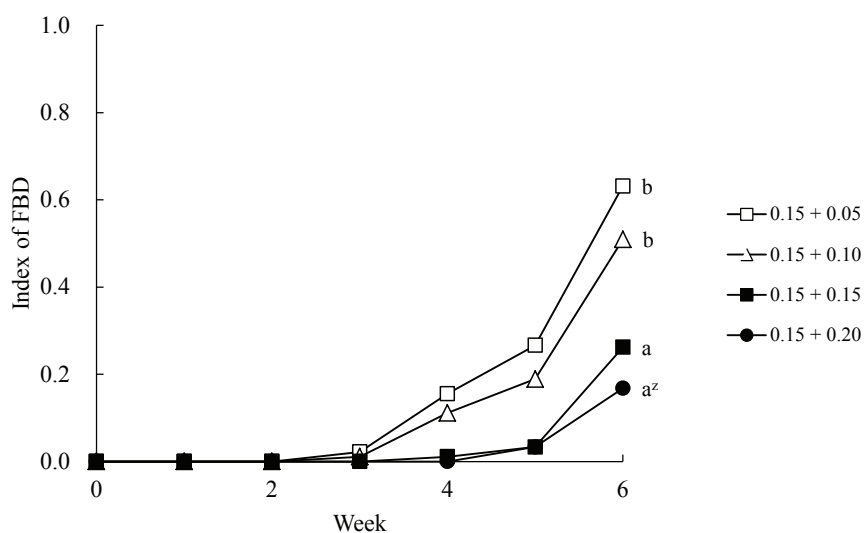


Fig. 3-16. The index of FBD of 'Iwa no hakusen' from 0 week to the sixth week after LED-630 + 735 light of NB treatments.

^z: Different letters indicate significant difference at $P < 0.05$ by Tukey's multiple comparison test.

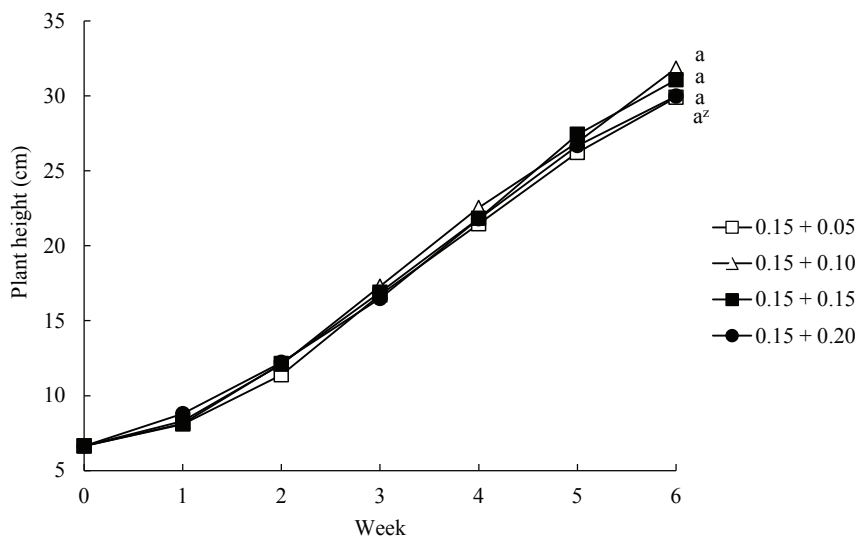


Fig. 3-17. The shoot length of 'Iwa no hakusen' from 0 week to the sixth week after LED-630 + 735 light of NB treatments.
^z: Different letters indicate significant difference at $P < 0.05$ by Tukey's multiple comparison test.

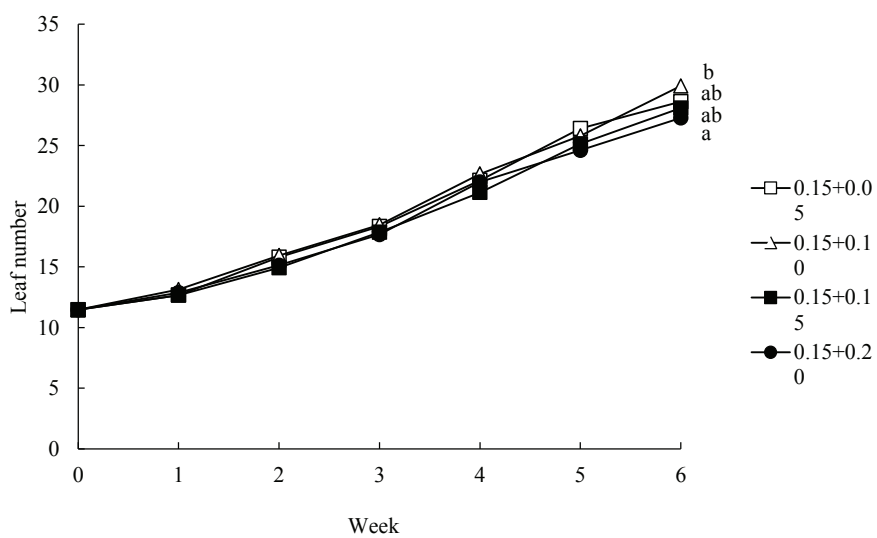


Fig. 3-18. The leaf number of 'Iwa no hakusen' in the sixth week after LED-630 + 735 light of NB treatments.
^z: Different letters indicate significant difference at $P < 0.05$ by Tukey's multiple comparison test.

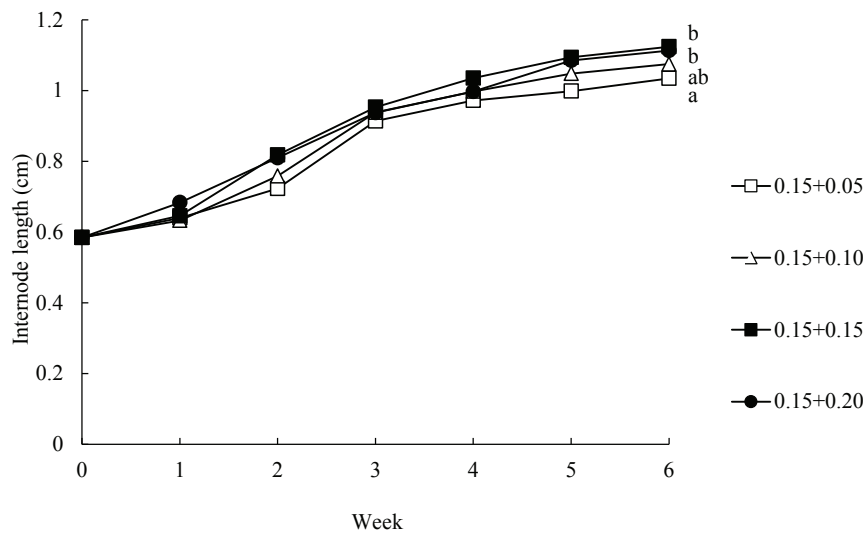


Fig. 3-19. The internode length of 'Iwa no hakusen' from 0 week to the sixth week after LED-630 + 735 light of NB treatments.

^z: Different letters indicate significant difference at $P < 0.05$ by Tukey's multiple comparison test.

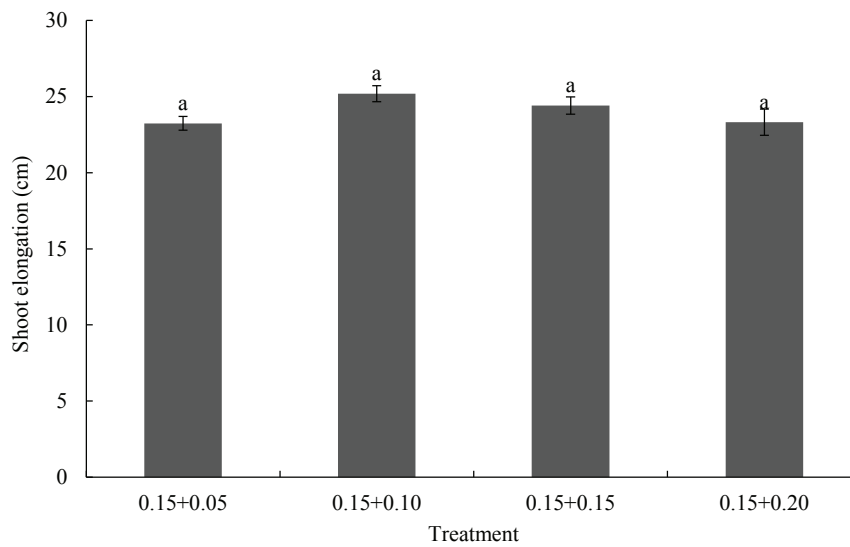


Fig. 3-20 The shoot elongation of 'Iwa no hakusen' in the sixth week after LED-630 + 735 light of NB treatments.

^z: Different letters indicate significant difference at $P < 0.05$ by Tukey's multiple comparison test.

Total Discussion and Conclusion

Applying lighting in the night for chrysanthemum light culture had been carried out since 1950s in Japan. It had been applying incandescent (INC) lamp lighting in the short-day length season for inhibiting flowering of chrysanthemum and applying shading in the long-day season for promoting flowering of chrysanthemum production. Therefore, it was possible for the growers to regulate the flowering time of chrysanthemum for year around production. In light culture of chrysanthemum, INC lamp spent a lot of electric power in energy consumption and increases the cost in production. In the situation of preventing global warming and for saving energy, it is needed to search new light source and take the place of the INC lamp and to reduce the cost in chrysanthemum production. New light source of LED had been developed in agriculture. Because plants can perceive wide wavelength of light in nature and LED is the lamp that emits single-wavelength, the effect of LED lamp applying in the growing of chrysanthemum is unclear.

In our Part 1 study, we had test the night break (NB) effect of INC lamp and LED lamp which peak emission were 630, 660, 690, 735 and 660+735 nm. The results were showed R light contained of LED-630 and LED-660, LED-690, LED-660+735 and INC had perfect inhibitory effect on FBD of autumn-flowering type 'Jimba', but FR light of LED-735 did not. The results were as similar as the report of R light had good inhibitory effect on short-day plant (Reid et al., 1967). In summer-autumn flowering type of 'Iwa no hakusen', R light of LED-630 and LED-660 could not inhibit its FBD, FR light of LED-735 delay its FBD, and LED-690, LED-660+735 and INC could inhibit its FBD very well. NB of R and FR light had different effect on 'Jimba' and 'Iwa no hakusen', but R and FR contained light sources of LED-690, LED-660+735 and INC had good inhibitory effect on both of the two cultivars.

For inhibitory of autumn-flowering type of 'Jimba', R light of NB inhibited its FBD was because the R light NB induced phyB. PhyB was attributed for NB inhibiting flowering in SDPs (Mockler et al., 2003). But it was different in 'Iwa no hakusen' in NB treatments of LED-630 and LED-660. It was suggested that 'Iwa no hakusen' had the different reaction in phyB responding to R light NB in flowering.

In the investigation of plant growth, it was found that FR light contained treatments of LED-735, LED-660+735 and INC had significant promotion effect on plant height growing, but not on leaf number. The promotion effect was not related to cultivars, but irradiance of FR light (Fig.1-12). Because there was no effect on leaf number or node number, the promotion of plant height was attributed to internode length elongation which was induced by FR light. It was suggested that this effect was because of the biosynthesis of GA which was induced by FR light (Hisamatsu et al., 2005)

In 'Jimba' and 'Iwa no hakusen', NB of R or FR light had different effect. This difference was supposed to be classified by the difference of flowering group of chrysanthemum. We had test the NB effect of LED on 12 cultivars. These cultivars were included summer-to-autumn flowering, autumn-flowering and late-autumn flowering type of chrysanthemum.

The results showed that 12 cultivars were classified into 2 groups by flowering or not in treatment of LED-735. [Group A] Normal type: The cultivars classified into this group cannot be inhibited their FBD by LED-690 and LED-735. This group was included all the autumn-flowering type cultivars and some of summer-to-autumn flowering type of cultivars. [Group B] Light sensitive type: FBD of which cultivars were classified into this group were inhibited or delayed by any light quality. This group was included autumn-flowering type cultivars and late-autumn flowering type of cultivar.

In Group A, all the autumn-flowering type cultivars had the same response to R or FR light

NB as the normal short-day plants. Because of some summer-to-autumn flowering type in this group, it was suggested that some of summer-to-autumn flowering type of cultivars had the same response to R and FR light NB as autumn-flowering type cultivars. And they were different to those summer-to-autumn flowering type of cultivars in Group B. It was suggested that the light regulating flowering related internal factor in summer-to-autumn flowering type of chrysanthemum were more complicated than that in autumn-flowering type chrysanthemum. The difference of FBD responding to R or FR light of NB was not simply classified by the flowering type of chrysanthemum.

LED-690 could not inhibit FBD in Group A. It was thought that light intensity of LED-690 was too low to inhibit it. And it was suggested that the NB of LED-690 had weaker inhibition than R light. Although late-autumn-flowering type of 'Sei tsudoï' was belonged to Group B, 12 h of day length might have already inhibited its FBD in this study.

Same as the study in Part 1, combination of R and FR LED light of NB had good inhibitory effect in all of the cultivars, similar to that in INC. And FR light contained treatments promoted internode elongation according to the cultivars.

Light intensity was also affected the inhibitory effect. We had the study of LED light intensity of NB on 'Jimba'. It was showed the relation between FBD and light intensity of LED-630 or LED-690 (Fig. 3-10). The inhibition level of FBD had depended on NB light intensity. For realizing index of FBD less than 0.2, at least it was needed above $0.150 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ of LED-630 and above $0.460 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ of LED-690. LED light of 630 nm had more effective inhibition of flowering than that of 690 nm with small amount of electric energy. In 'Iwa no hakusen', we carried out the combination R and FR LED light intensity of NB study. FR light of LED-735 intensity which combined with R light of 630 nm with $0.15 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ had enough inhibition effect of FBD of 'Iwa no hakusen' at higher than over

$0.15\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. In practical using LED lamp, growers should manage the installation of the number of LED lamps and the height to the top of chrysanthemum plants for keeping these R and FR light intensity.

As a conclusion, R light of LED had most inhibitory on autumn-flowering type of 'Jimba', such as LED-630 or LED-660. Combination of R and FR LED light not only had good inhibitory effect on 'Jimba', but also in summer-autumn-flowering type of 'Iwa no hakusen'. It could inhibit FBD in 'Jimba' when light intensity of peak emission was higher than $0.150\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ in LED 630 nm, and it could inhibit that in 'Iwa no hakusen' when light intensity of 735 nm was higher than $0.150\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ combining 630 nm lighting at $0.150\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. Because 'Jimba' and 'Iwa no hakusen' were representative chrysanthemum cultivars in Japan, autumn-flowering and summer-to-autumn flowering type of cultivars had the similar NB responses to 'Jimba' and 'Iwa no hakusen'. Therefore, the results of this study should have reference to chrysanthemum LED light culture. Growers could manage the light quality of LED and the installation of the number of LED lamps and the height to the top of chrysanthemum plants in production.

SUMMARY

Chrysanthemum (*Chrysanthemum morifolium* Ramat.) is one of the most important cut flowers and has the highest consumption in the world. Currently, for inhibiting its early flowering and for increasing its shoot length in autumn or in winter, growers have been applying night break lighting using incandescent (INC) lamp. Now for saving energy, Japanese government has decided to cancel the manufacture and selling of INC lamp. Recently, LED lamp has been developed to a highly effective, low electric consumption and long duration of new light source in agriculture. As LED a monochromatic lamp, different light wavelength of LED lamp has different effect on the flowering of chrysanthemum.

We studied the NB effect on chrysanthemum by applying INC lamp and LED lamp peak emission at 630, 660, 690, 735 and 660+735 nm. The results were showed that red (R) light of LED-630 and LED-660 had perfect inhibition on floral bud differentiation (FBD) in autumn flowering type cultivar of 'Jimba', but could not inhibit that in summer-to-autumn flowering type cultivar of 'Iwa no hakusen'. Far-red (FR) light treatment of LED-735 had no effect on inhibition of floral bud differentiation in 'Jimba', but delay that in 'Iwa no hakusen'. Treatments of LED-690 and INC lamp inhibited FBD in 'Jimba' and strongly delayed that in 'Iwa no hakusen'. Treatment of combination of R light LED-660 and FR light LED-735 inhibited FBD in both of the two cultivars.

LED-735, LED-660+735 and INC lamp, which were contained FR-light, enhanced shoot elongation on both 'Jimba' and 'Iwa no hakusen' and had significant difference to that in control (short day condition), LED-630 and LED-660 treatments. Photon flux density (PFD) at the range of 700 nm to 799 nm had significantly close relationship with internode length. It was considered that the internode elongation was brought about by GA biosynthesis induced by FR-light. There was no relationship between internode length and P_{fr}/P_{total} or R/FR ratio.

It was supposed that the difference of FBD in 'Jimba' and 'Iwa no hakusen' responding to R or FR light of NB was classified by the flowering type of chrysanthemum. And for validating the LED NB effect on more chrysanthemum cultivars, 12 cultivars were investigated in the next study.

The results showed that 12 cultivars were classified into 2 groups by flowering or not in treatment of LED-735. [Group A] Normal type: The cultivars classified into this group cannot be inhibited their FBD by LED-690 and LED-735. They were 'Sei aegean', 'Sei no nami', 'Sei elsa', 'Remidas', 'Sei no makura', 'Seiko koumyou', 'Seiko no makoto', 'Jimba' and 'Sei yukino'. In this group, the florets were differentiated at the 6th week. All cultivars in which flowering season was September or October belonged to this type. On the other hand, although flowering season of 'Sei aegean' was July, it belonged to this type. [Group B] Light sensitive type: FBD of which cultivars were classified into this group were inhibited or delayed by any light quality. The flowering season of these cultivars belonging to this group was divided into June or July: 'Cent west' and 'Sei opti', and November: 'Sei tsudo'. As 'Sei tsudo' belonged to late 'autumn flowering type chrysanthemum', 12h of light irradiation by white fluorescent lamp might inhibit FBD. On the other hand, 'Cent west' and 'Sei opti' belonged to early 'summer-to-autumn-flowering type chrysanthemum', and FBD of these cultivars were also inhibited by irradiation of LED-690 or LED-735. The difference of FBD responding to R or FR light of NB was not simply classified by the flowering type of chrysanthemum. It was confirmed that combination of R and FR LED light of NB had good inhibitory effect on FBD in all of the cultivars in the study.

Currently, the popularization of LED lamp for the production of chrysanthemum is not prevalent because the effective wavelength and light intensity for inhibition of flowering is still not obvious. We researched the lowest light intensity peak emission at 630 nm and 690 nm that

could inhibit the flowering of 'Jimba' to establish a method for effective using of LED lamp.

In LED-630, FBD of 'Jimba' was not inhibited at 0.050 and 0.083 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. It was delayed by 0.100 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ and inhibited obviously by 0.150 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. In LED-690, light intensities of 0.160 and 0.260 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ could not inhibit FBD. The FBD was delayed by 0.350 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ and was inhibited by 0.460 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. For realizing FBD index less than 0.2, at least it was needed above 0.350 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ at LED-690 and above 0.100 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ at LED-630. LED-630 had more effective inhibition of flowering than that of 690 nm with small amount of electric energy.

Next, it was investigated the lowest intensity of FR light under combination of R and FR light of LED-630+735 for inhibiting FBD in 'Iwa no hakusen'. The results showed that FBD of 'Iwa no hakusen' could not be inhibited by the treatments of 0.15+0.05 (LED-630 + LED-735) and 0.15+0.10, and was seriously delayed by treatment of 0.15+0.15, inhibited by treatment of 0.15+0.20. There was no significant difference in shoot length. And it was not consider that leaf number had regularity among these treatments.

The inhibition effect of using R+FR light was better than those of using R or FR light only. FR light intensity which combined with R light with 0.15 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ had enough inhibition effect of FBD of 'Iwa no hakusen' at higher than over 0.15 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$. In practical using LED lamp, growers should manage the installation of the number of LED lamps and the height to the top of chrysanthemum plants for keeping these R and FR light intensity.

摘要

キク (*Chrysanthemum morifolium* Ramat.) は世界的に生産量の多い重要な花きである。キクは周年栽培を行うにあたり、日照時間が短い秋から冬にかけて夜間に白熱電球を点灯して暗期中断を行うことで花芽分化を抑制し、切り花として十分な茎長が行われている。しかし、白熱電球は地球温暖化および省エネルギー対策の観点から製造販売の中止が決定している。その代替として注目されるLEDは低消費電力で長寿命である一方、単波長の光源であるため、波長によるキクの成長に及ぼす影響が白熱電球と異なることが知られている。

本研究では、630、660、690 および 735 nm の 4 種類の波長の LED を暗期中断の光源として用いてキクの花芽分化に及ぼす影響を調査した。その結果、赤色光である 630 および 660 nm の LED は、秋ギク‘神馬’に対して完全な花芽分化の抑制効果を見せたが、夏秋ギク‘岩の白扇’では花芽分化の抑制効果が見られなかった。遠赤色光である 735 nm の LED は、‘神馬’の花芽分化を抑制しなかったが、‘岩の白扇’では花芽形成の遅延が確認された。白熱電球と 690 nm の LED では、‘神馬’の花芽分化の抑制と、‘岩の白扇’の花芽分化の大幅な遅延が見られた。また、660 nm と 735 nm の LED の混合照射では、両品種の花芽分化を抑制した。

同時にシュート伸長の効果も測定した。735 nm の LED、660 と 735 nm の LED の混合、および白熱電球では、両品種において対照区である短日処理と比較して大幅なシュート伸長の促進効果が確認できた。また、700~799 nm にかけての光量子束密度と節間長には強い正の相関が見られた。これらのシュート伸長は、遠赤色光により誘導されたジベレリンにより節間伸長したことによるものと考えられる。なお、シュート伸長の程度と P_{fr} 型/ P_r 型の量比もしくは R/FR 比との間には相関は見られなかった。

‘神馬’と‘岩の白扇’とで各種光源による暗期中断に対して異なる応答を見せるのは、これらの品種特性が異なることと関連していると考えられる。そこで、開花期が異なる 12 品種に対しても同様の実験を行った。

実験に用いた 12 品種は、735 nm の LED での暗期中断により開花したものとしなかったものに分類できた。[A]通常型:690 および 735 nm の LED 照射では花芽分化は抑制されなかった。通常型には‘セイエーグ’、‘精の波’、‘セイエルザ’、‘レミダス’、‘精の枕’、‘精興光明’、‘精の誠’、‘神馬’‘精ゆきの’が分類された。[B]光感受型:どの光源による暗期中断であっても花芽分化の抑制や遅延が見られた。光感応型には‘セントウエスト’、‘セイオプティ’および‘精つどい’が含まれた。なお、赤色光と遠赤色光の LED を組み合わせた場合は、どの品種に対しても花芽分化の抑制効果が見られた。

現在、キク生産において LED は普及していない。これは、開花抑制に適した波長と光量は不明であるためである。本研究では、630 および 690 nm の LED での暗期中断が秋ギクの花芽分化を抑制することを確認できたことから、‘神馬’に対し様々な光量の 630 および 690 nm の LED を照射し、花芽分化を抑制しうる最小の光量を調査した。630 nm の LED においては、0.050 および 0.083 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ では花芽分化は抑制されなかったが、0.100 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ では花芽分化が遅延し、0.150 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ になると完全に花芽分化が抑制された。これらは花芽分化の抑制の程度は暗期中断の光源の光量に依存することを示していた。

次に 690 nm の LED においては、0.160 および 0.260 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ では花芽分化の抑制には不十分であったが、0.35 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ では花芽分化が遅延し、0.460 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ になると花芽分化の抑制が見られた。

以上のことより、‘神馬’の花芽分化を抑制するには、690 nm の LED では 0.35 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ 以上、630 nm の LED では 0.100 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ 以上の光量が必要であり、630 nm の LED がより効率が良かった。ただし、花芽分化を完全に抑えるには、630 nm の LED で 0.150 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ 以上の光量が望ましい。

‘岩の白扇’を用いて赤色光である 630 nm と遠赤色光である 735 nm の LED の混合照射における、花芽分化抑制に必要な 735 nm の LED の最小光量を調査した。630 nm の LED の

光量は $0.150 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ で統一した。その結果、735 nmのLEDの光量が0.05および0.10 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ では花芽分化は抑制されなかったが、0.15 および 0.20 $\mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ では花芽形成率が低下した。従って、‘岩の白扇’に対して遠赤色光を $0.150 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ の赤色光と混用した場合には、遠赤色光の光量が $0.150 \mu\text{molm}^{-2}\text{s}^{-1}\text{nm}^{-1}$ 以上の時に十分な花芽分化抑制効果が得られた。

以上の結果より、実際にキクの栽培に利用する際は、植物体の上部において上記の光量が保てるようなLEDの個数や設置位置を調節する必要がある。

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