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Effects of Different Light Sources on Shoot to Root Ratio and Intumescence Incidence in Tomato Seedlings Grown in a Commercial Closed Seedling Production System

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ABSTRACT

Intumescence is a physiological disorder in which cell walls are disrupted. Intumescence causes leaf deformation, browning and defoliation, and commonly affects tomato plants. This is a limitation in seedling production. The causes of intumescence are not known; however, ultraviolet (UV) deficiency is considered as one of the causes of intumescence. Other possible causes include high humidity and rapid changes in the water environment. In this study, tomato seedlings were grown in a commercially-available closed seedling production system under either newly installed LEDs with a low UV light intensity or conventional fluorescent lamps sources. The relationship between the shoot/root (S/R) ratio and incidence of intumescence was evaluated. In addition, the effects of different light sources were evaluated on the xylem pressure potential in the seedlings under rapidly fluctuating water conditions. As a result, the S/R ratio of seedlings grown under LEDs, with low UV intensity, was greater than that of seedlings grown under fluorescent lamps. There was a positive correlation between the S/R ratio and the incidence of intumescence of four types of seedlings with different combinations of light sources and varieties. Xylem pressure potential decreased more in seedlings grown under LEDs, compared to seedlings grown under fluorescent lamps and dry conditions, but increased significantly when irrigated. These results suggest that reducing the occurrence of intumescence in seedling production systems can be possible by a light source that can help seedlings grow with small S/R ratios and reduce fluctuations in the water condition.

Introduction

A closed seedling production system that can control the cultivation environment (internal temperature, day length, and irrigation) is usually used for growing vegetable seedlings because of its ability to establish stability (Chun and Kozai, 2001; Ohyama et al., 2003). Artificial light sources are used in closed-type seedling production systems and, recently, LEDs have replaced fluorescent lamps as an artificial source of light. However, it has been reported that different light sources have various effects on plants (Ohashi-Kaneko 2007; Jishi et al. 2016; Hernández 2016; Masuda et al. 2021; Moosavi-Nezhad et al. 2021). Furthermore, the incidence of intumescence reportedly increases in tomatoes when LEDs are used as a light source (Misu et al., 2018). Intumescence is a physiological disorder in which

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the cell wall is destroyed by turgor pressure and the leaves of tomatoes are deformed (Lang et al., 1983 Craver et al., 2014; Wu et al., 2017). Intumescence causes not only leaf deformation but also leaf browning and defoliation, even as the symptoms advance (Miyama and Yasui, 2021). Seedlings from intumescent plants cannot be used, which is a limitation in seedling production. Although the causes of intumescence have not been identified, UV deficiency is considered to be one of the causes (Lang and Tibbitts, 1983; Craver et al., 2014; Kubota et al., 2017). The cuticular layer of tomato leaves becomes thinner by UV deficiency, which promotes the occurrence of intumescence (Suzuki et al., 2020). It is also speculated that genes related to intumescence induced by UV irradiation are suppressed under UV deficiency conditions (Wu et al., 2017). However, it seems that the phyto-physiological mechanism whereby intumescence increases under UV deficiency conditions has not yet been clarified. Other possible causes include high humidity (Lang and Tibbitts, 1983; Misu et al., 2018) and rapid changes in water condition (Miyama and Yasui, 2021). The extent of intumescence varies among varieties (Ozawa et al., 2018; Miyama and Yasui, 2021); the damage caused by intumescence is more severe in varieties with a higher shoot to root (S/R) ratio.

The S/R ratio of tomato seedlings differs with the wavelength of the irradiating light source; the stronger the UV light, the smaller the S/R ratio (Bekhzod et al., 2014). Therefore, differences in UV light intensities of light sources may affect the S/R ratio of tomato seedlings, and the differences in the S/R ratio could affect the degree of intumescence. However, none of the previous studies have reported the effects of different light sources used in a closed seedling production system on the tomato S/R ratio and intumescence.

Bottom-fed irrigation is commonly used in closed seedling production systems. The soil moisture content and relative humidity tend to increase rapidly in such closed systems (Misu et al., 2018). This is considered to be a condition where intumescence is likely to occur.

The cause of this type of intumescence is unknown; however, in seedling production, intumescence should be avoided. Therefore, it is necessary to identify the cause of intumescence when using LEDs as an artificial light source for closed seedling production systems and to establish a solution. In this study, the S/R ratio was specified, and the effects of different light sources were evaluated on the occurrence of intumescence in closed seedling production systems.

Materials and Methods

Seedling production systems with different light sources

A commercially available closed-type seedling production system ("Nae-Terace"; Mitsubishi Chemical Aqua Solutions Co., Ltd., Tokyo, Japan) was equipped with four shelves. Two types of light sources, LEDs and conventional fluorescent lamps, were installed on two shelves each, in a unified closed-type seedling production system. Photon flux density of the tested light sources were based on wavelength and measured on the cultivation shelf of a commercially available closed-type seedling production system (Fig. 1), with a newly installed light source as LEDs (Fig. 1A) and with previously installed light sources as fluorescent lamps (Fig. 1B). The photon flux density was set at 300–800 nm on the growing surface, with 251 µmol cm^{-2} s^{-1} under LEDs, and with 284 µmol cm^{-2} s^{-1} under fluorescent lamps. Furthermore, the photon flux density at 300–400 nm on the growing surface was 0.09 µmol cm^{-2} s^{-1} under LEDs and 2.60 µmol cm^{-2} s^{-1} under fluorescent lamps.

Specimen species and seedling growth method

'Momotaro Peace' (Takii Seed Co., Ltd., Kyoto, Japan) is usually characterized by a high S/R ratio and a high incidence of intumescence. Meanwhile, 'Reiyo' (Sakata Seed Co., Ltd., Kanagawa, Japan) usually has a low S/R ratio and a low incidence of intumescence (Miyama and Yasui, 2021). These cultivars were used for the purpose of this experiment.

The seeds were sown in 128-well cell trays filled with medium (Na Tera; Mitsubishi Chemical Aqua Solutions Co., Ltd., Tokyo, Japan) which contained peat moss, vermiculite, and pearlite. After sowing, the seedlings were grown in a closed seedling production system after 2 days of incubation in the dark at 30 °C. The closed seedling production system was set at a light/dark photoperiod of 16/8 h (light period: 8:00–0:00 h; dark period: 0:00–8:00 h) and with a carbon dioxide concentration of 1000 ppm, as a conventional concentration in this system. (Misu et al., 2018).

The temperature was maintained at 30 °C/30 °C for 3 days, 30 °C/25 °C for 1 day, and 28 °C/23 °C thereafter. The irrigation start time was set to the beginning of the light period and the irrigation duration took 10 min. Otsuka-A formula (OAT Agrio Co., Ltd., Tokyo, Japan; NH_4–N = 23 ppm; NO_3–N = 233 ppm; P_2O_5 = 120 ppm;
K_2O = 405 ppm; CaO = 230 ppm; MgO = 60 ppm; MnO = 1.5 ppm; B_2O_3 = 1.5 ppm; Fe = 2.7 ppm; Cu = 0.03 ppm;

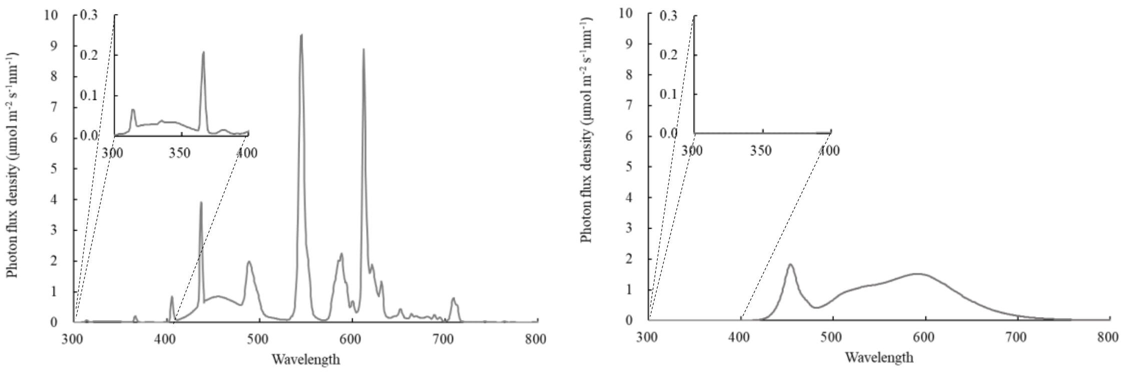

Fig. 1. Photon flux density of the tested light source based on the wavelength, measured on the cultivation shelf of a commercially available closed-type seedling production system with (A) newly installed light source: LEDs and (B) previously installed light sources: fluorescent lamps.

Zn = 0.09 ppm; Mo = 0.03 ppm) was used as the culture medium, and the electrical conductivity (EC) was set to 1.2 dS/m. The temperature and relative humidity in the closed seedling production system were measured using a thermometer and humidity meter (RTR-507; TandD Co., Ltd, Nagano, Japan), respectively.

S/R ratio survey

Seedlings were grown in a closed seedling production system and were sampled 12 days after sowing, and the S/R ratio was determined thereafter. Sampling was performed for each light source and variety. The shoots and roots of the sampled tomato seedlings were dried at 60 °C and weighed. The S/R ratio was calculated as dry matter weight. Shoot weight was defined as the above-ground weight of plants.

Screening for intumescence incidence

The number of intumescent plants in the closed seedling production system was examined 16 days after sowing. The incidence rate of intumescence plants was calculated. Three to eight plants were screened in four replicates for each light source and each variety.

Xylem pressure potential of the tomato seedlings grown under fluctuating water conditions

Xylem pressure potential was measured using a pressure chamber (600D; PMS Instrument Co., Ltd., Oregon, USA). As test samples, fourteen-day-old seedlings were grown in a closed seedling production system under LEDs where fluorescent lamps were used. At this time, intumescence was observed in ‘Momotaro Peace’ but not in ‘Reiyo’, under both LED and fluorescent lamp illumination. Therefore, xylem pressure potential was calculated only for ‘Reiyo’. The seedlings were placed in a controlled environment chamber (LPH-411PFQDT-SP; Nippon Medical & Chemical Instruments Co., Ltd, Tokyo, Japan) in light and at 28 °C, with a relative humidity of 50%. After 12 h of no irrigation, the relative humidity changed to 90% and bottom feeding was initiated. Xylem pressure potential was measured at 0, 12, and 14 h (i.e. 2 h after changing the environmental conditions in the controlled environment chamber) after the seedlings were placed in a controlled environment chamber, with four to six seedlings sampled for each condition.

Statistical analysis

A t-test was performed using the Excel (Microsoft Corporation, USA) add-in software Statcel2 (OMS Publishing Co., Ltd, Tokyo, Japan). Twelve days after sowing, differences in the dry matter weights of shoots and roots, as well as the S/R ratios, were tested between seedlings of ‘Momotaro Peace’ and ‘Reiyo’ via a t-test (at alpha levels of 1% and 5%). Differences in the incidence of intumescence between ‘Momotaro Peace’ and ‘Reiyo’ seedlings were evaluated 16 days after sowing, according to a t-test (at alpha levels of 1% and 5%). Differences in xylem pressure potential among light sources were tested using t-tests.

Results and discussion

Effect of different light sources on the S/R ratio and intumescence incidence in tomato seedlings

The relative humidity in the closed seedling production system increased rapidly to 90% during 10 min of irrigation at the beginning of the light period. Then, it was maintained at 40%–60% (Fig. 2).

Table 1. Effect of different light sources in a closed seedling production system on shoot and root dry matter weight and shoot/root (S/R) ratio of 'Momotaro Peace' and 'Reiyo' seedlings, 12 days after sowing

Light source	Shoot DW(mg)		Root DW(mg)		S/R ratio	
	'Momotaro Peace'	'Reiyo'	'Momotaro Peace'	'Reiyo'	'Momotaro Peace'	'Reiyo'
LEDs	38.44	60.00	1.26	2.59	30.59	23.19
Fluorescent lamps	56.44	73.27	2.99	5.05	18.89	14.50
t-test	*	ns	**	**	**	**

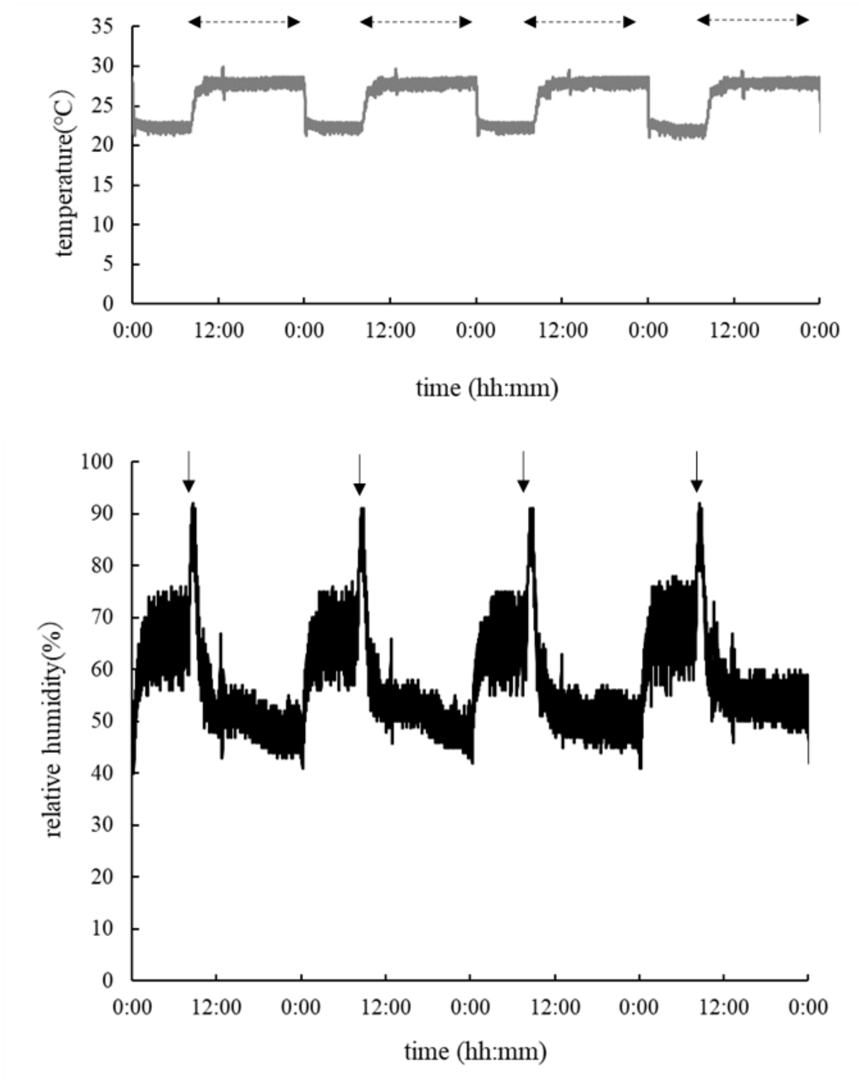

Fig. 2. Changes in temperature and relative humidity of the closed seedling production system from day 13 to day 16 after sowing; light period (dashed line): 8:00–0:00 h, dark period: 0:00–8:00 h; irrigation time 8:00–8:10 h (↓ indicates the irrigation start time).

Based on the t-test, * indicates a significant difference at the 5% level, and ** indicates a significant difference at the 1% level. The S/R ratio was calculated after arcsine transformation. The incidence of intumescence was higher in seedlings grown under LEDs than in those grown under fluorescent lamps in both 'Momotaro Peace' and 'Reiyo', 16 days after sowing (Table 2). The incidence of intumescence was higher in 'Momotaro Peace' than in 'Reiyo', irrespective of the light source. The incidence of intumescence was 100% for 'Momotaro Peace' when the seedlings were grown under LEDs. Keiyo seedlings showed no intumescence when grown under fluorescent lamps (Table 2). A positive correlation was observed between S/R ratios (of both cultivars) and the incidence of intumescence, 16 days after sowing (r = 0.9764; Fig. 3) under both light sources.

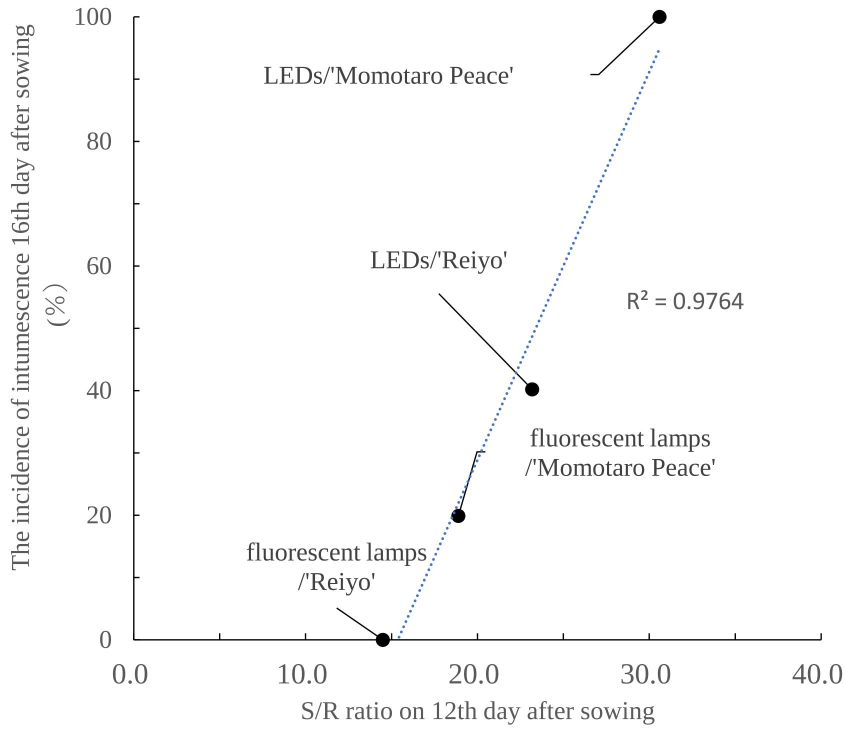

Fig. 3. Effects of the shoot/root (S/R) ratio on day 12, after sowing the tomato seedlings of different varieties on the incidence of intumescence under different light sources

Table 2. Effects of different light sources in a closed seedling production system on the incidence of intumescence in 'Momotaro Peace' and 'Reiyo' seedlings, 16 days after sowing

	Incidence of intumescence (%)	
Light source	'Momotaro Peace'	'Reiyo'
LEDs	100.0	40.2
fluorescent lamps	19.9	0.0
t-test	**	*

Based on the t-test, * indicates a significant difference at the 5% level, and ** indicates a significant difference at the 1% level. Calculated after *arcsine* transformation, from three to eight plants in four replicates for each light source and each variety.

Changes in xylem pressure potential under changing water conditions of two varieties of seedlings grown under different light sources

Fourteen days after sowing, 'Reiyo' seedlings without intumescence were grown for 12 h in a closed seedling production system equipped with LED under conditions of 50% relative humidity and no irrigation. At this time, the xylem pressure potential of seedlings grown under LEDs was significantly lower than that of seedlings grown under fluorescent lamps (Fig. 4). When the relative humidity was increased from 50% to 90%, the xylem pressure potential of seedlings grown under LEDs and fluorescent lamps increased to the same level as that of seedlings grown under non-irrigated and bottom-fed conditions. The increase in xylem pressure potential during the change from dry to wet conditions was greater in seedlings grown under LEDs than in seedlings grown under fluorescent lamps (Fig. 4).

Discussion

The S/R ratio and the incidence of intumescence among the two tested cultivars were higher in 'Momotaro Peace' than in 'Reiyo' (Table 1 and 2), which confirmed previous results (Miyama and Yasui, 2021). The effect of different light sources in a closed seedling production system on the S/R ratio of seedlings was examined, and showed that the S/R ratio of seedlings grown under LEDs was higher than that of seedlings grown under fluorescent lamps on both varieties. It has been reported that both UV and visible light irradiation reduces the S/R ratio of tomato plants (Bekhzod et al., 2014). As shown in Fig.1, the UV intensity of the LEDs in this study was extremely lower than that of the fluorescent lamps. Therefore, the higher S/R ratio observed under LEDs in the present study could be attributed to fewer roots due to lower intensity of UV light. Both varieties showed a higher incidence of intumescence under LEDs than under fluorescent lamps. In this regard, it has been reported that lower UV intensities result in a higher incidence of intumescence (Lang and Tibbitts, 1983; Craver et al. 2014; Kubota et al. 2017; Wu et al. 2017; Suzuki et al. 2020).

Therefore, the low UV intensity conditions under LEDs in this study may have increased the incidence of intumescence. Furthermore, the xylem pressure potential of seedlings grown under LEDs was lower than that of seedlings grown under fluorescent lamps after 12 h of non-irrigation (50% relative humidity) and increased more significantly when the conditions were rapidly changed to irrigation at 90% relative humidity (Fig. 4). There was a positive correlation between the S/R ratio and the incidence of intumescence in four seedlings with different combinations of light source and varieties (Fig. 3).

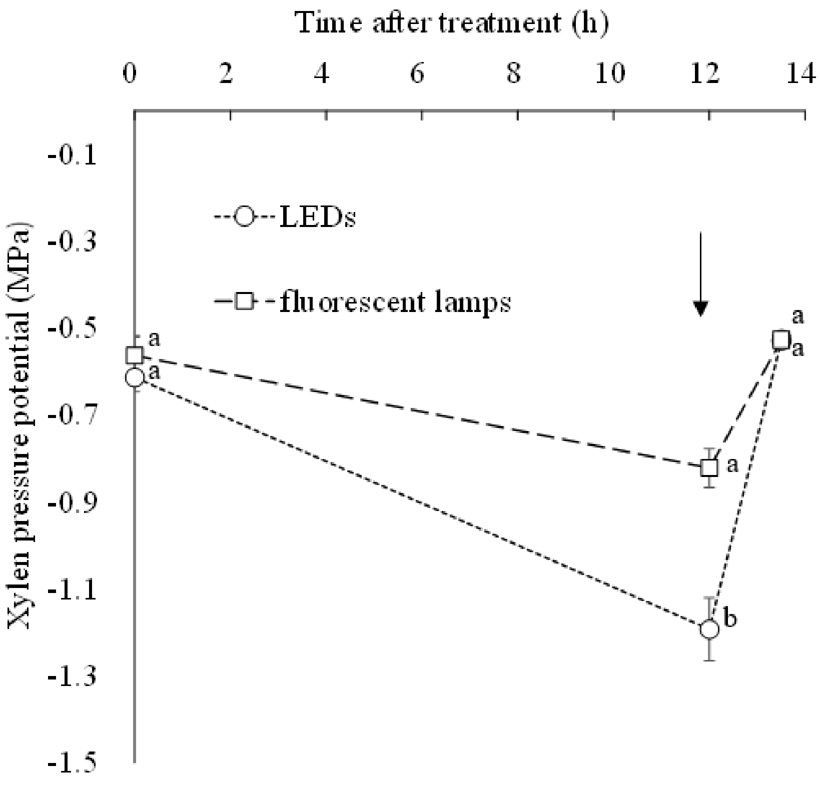

Fig. 4. Changes in the xylem pressure potential of tomato seedlings grown in a closed-type seedling production system. Data pertain to 'Reiyo' seedlings on day 14 after sowing. Different letters indicate significant difference at the 5% level according to the t-test results. Vertical bars indicate the standard errors (n = 4–6). Temperature: 28 °C; relative humidity: 50%, with no irrigation until 12 h. At 12 h (↓), the relative humidity changed to 90%, as a result of bottom-fed irrigation in a chamber with a controlled environment.

Miyama and Yasui (2021) found that intumescence was more severe in varieties with higher S/R ratios. The S/R ratio and the incidence of intumescence were positively correlated. The higher the S/R ratio, the lower the value of xylem pressure potential during the drying process, and the greater the increase in xylem pressure potential when changing to wetter conditions. Assuming the same water uptake per root weight, the higher the root weight, the faster the water uptake rate, and assuming the same transpiration per shoot weight, the lower the shoot weight, the slower the transpiration rate. Therefore, it can be inferred that as the S/R ratio increases, the balance between the water absorption rate and transpiration rate is easily disrupted and that the amount of change in xylem pressure potential increases. Thus, the turgor pressure is thought to destroy the cell wall and cause intumescence.

The commercially-closed seedling production system in this study showed a rapid increase in relative humidity with an increase in soil water content during irrigation. This suggested that rapid changes in the water environment were likely to occur within the closed seedling production system.

The above results indicated that seedlings grown under low UV intensity LEDs in a closed seedling production system had a higher S/R ratio than those grown under fluorescent lamps. It was considered that intumescence would be more likely to occur due to an enhanced range of fluctuation in the xylem pressure potential and, consequently, changes in turgor pressure in a closed seedling production system, where rapid changes from dry to wet conditions are repeated.

Conclusion

In this research, an important finding was that inducing the growth of seedlings with small S/R ratios can suppress the occurrence of intumescence. In addition, it would be necessary to take measures to reduce the fluctuation range of the water environment in the closed seedling production system by installing humidity control devices on the seedlings, as another strategy to suppress the occurrence of intumescence.

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Conflicts of interest

The authors declare that there are no conflicts of interest.

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