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Effective scion propagation using light-emitting diode irradiation for nursery stock production in Japanese persimmon

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ABSTRACT

The objectives of this study were to determine the developmental responses of Japanese persimmon seedlings (*Diospyros kaki* Thunb.) to variations in photoperiod and light conditions, and to develop an effective method for scion propagation using light-emitting diode (LED) irradiation for nursery stock production. Shoot length and number of nodes increased after an irradiation treatment of 20 h/day compared with that of 12 h/day. Shoot length under red LED light was significantly greater than under blue LED light in the 20 h/day treatment. The number of nodes also increased under red LED light compared to blue LED light. The number of leaf primordia was significantly larger in grafts in late July compared to late June. The survival rate of scions and the shoot lengths were greater for grafts in late July compared to late June. The number of buds in grafts in late July was significantly higher than in late June. Based on these results, we propose that twice grafts per year, consisting of a bark graft in February and a green graft in July, is a better alternative to the conventional once graft in Spring because of the higher survival rate of scions and their accumulation of larger numbers of buds. Our alternative method achieves up to a 6-fold improvement in scion propagation rate compared to the once graft approach.

1. Introduction

Japanese persimmon (*Diospyros kaki* Thunb.) is believed to have originated in East Asia (Yakushiji and Nakatsuka, 2007) and is a deciduous fruit crop that is now cultivated worldwide, particularly in Brazil, China, Israel, Italy, Japan, Spain, South Korea, and New Zealand (FAOSTAT, 2015). Japanese persimmon is one of the principal fruit crops in Japan, ranking fourth in total fruit production (MAFF, 2015). Since 1959, new cultivars have been released in Japan, especially pollination constant non-astringent (PCNA)-type cultivars that have large-sized fruit and a bright skin color, and are highly palatable (Sato and Yamada, 2016). Economically, the development and expansion of PCNA-type persimmon cultivars has been required to increase the agricultural competitiveness of Japan. However, low efficiency of the scion propagation is a serious problem for the nursey stock production. Therefore, it needs to establish an efficient method of propagation of the scions to promote rapid expansion of the excellent new cultivars.

Propagation of fruit crops is performed asexually using a technique consisting of cutting, layering, grafting, budding, and tissue culture (micropropagation) to produce genetic clones identical to the parent trees. In persimmon, cutting and layering are in general technically difficult (Tetsumura et al., 2001). Moreover, application of the various micropropagation techniques (Cooper and Cohen, 1985; Fukui et al., 1989; Sugiura et al., 1986; Yokoyama and Takeuchi, 1976, 1981, 1988) to commercial nursery stock production is limited by varietal differences in the rooting of microcuttings (Tetsumura et al., 2002). Grafting using a dormant scion is frequently used for propagating persimmon; however, the efficiency of stock multiplication is low because of a once-per-year grafting schedule and the moderate growth rate under open culture. Similar to apple (Sanada et al., 1983), the effective persimmon stock production will be achieved by successive grafts per year together with the scion multiplication.

Factors, such as temperature, water, nutrition, and light, which affect plant growth, also influence the success of propagation of nursery stock (Lavender, 1984). Of these factors, the quality and intensity of light can have a significant effect on plant growth, development, and biomass production (McDonald, 2003). As natural sunlight is not sufficient for horticultural industry purposes, artificial lighting is commonly used to improve the production and quality of plants in the greenhouse (Olle and Viršilė, 2013; Ooi et al., 2016). In plant growth chambers, illumination is obtained from conventional light sources, such as cool-white fluorescent light, high pressure sodium lights, and incandescent lights. However, these light sources have some limitations due to their short lifetime, high consumption of electricity, and heat emission. Recently, use of light emitting diodes (LEDs) for plant growth in controlled environments has emerged as an attractive, low-cost

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alternative method for producing vegetables and flowers (Arsovski et al., 2012; Bantis et al., 2016; Chen et al., 2014; Hirai et al., 2006; Lin et al., 2013). With respect to woody plants, shoot elongation and/or leaf expansion are significantly enhanced by LED irradiation in wild cherry (Astolfi et al., 2012), grape (Kubota and Uehara, 2000), and Japanese pear (Ito et al., 2014). Growth and development responses to a light spectrum depend on the plant species and/or cultivar (Astolfi et al., 2012; Lin et al., 2013; Olle and Viršilė, 2013). In lettuce (Chen et al., 2014), sweet pepper (Masuda et al., 2004), and grape (Kubota and Uehara, 2000; Heo et al.,2006), the red LED (640–690 nm) wavelength promotes shoot growth, and eggplant and sunflower (Hirai et al., 2006) are promoted with the blue LED. However, little information is available for developmental responses to variations in photoperiod and light conditions in persimmon. The objectives of this research were to (1) determine the effects of variation in photoperiod and LED wavelength on the growth characteristics of persimmon seedlings, and (2) to establish an effective method of scion propagation using LED irradiation for nursery stock production.

2. Materials and methods

2.1. Experiment 1: effect of photoperiod duration and use of three-wavelength fluorescent light on plant growth

In a preliminary experiment, we examined the effects of varying the period of irradiation with three wavelengths of fluorescent light: blue, 400–500 nm; green, 500–600 nm; and red, 600–700 nm. Growth of persimmon plantlets in this experiment was measured in two successive years (2011 and 2012). In 2011, seedlings of the cultivar ‘Atago’ were used. Seeds were sowed in a Jiffy-7® peat pot (Jiffy International AS, Kristiansand, Norway); after germination, the seedlings were grown in hydroponic culture for 21–28 d in a controlled environment room at a constant 25 °C and 60% relative humidity, with a 12 h photoperiod using cool white fluorescent lamps. Uniformly sized seedlings were then individually transplanted to a 0.3 L polypot filled with culture soil (sandy loam/peat moss/vermiculite = 7:2:1) fertilized with 0.03 g N/ 0.03 g P_2O_5/0.03 g K_2O. After 36 days, ten seedlings per cultivar and treatment were transferred to an environmentally controlled growth chambers and exposed to five different photoperiods, namely 12 h/day, 14 h/day, 16 h/day, 18 h/day, or 20 h/day, using a three-wavelength fluorescent lamp (FL40SSEX-D/37-SHG, NEC Co., Tokyo, Japan). Environmental conditions in the growth chambers were set at 25 °C/ 20 °C (temperatures with a 12 h cycle) and 60% relative humidity; photosynthetic photon flux density was maintained at around 100 μmol $m^{-2}s^{-1}$ for all treatments at plant level. The pots were watered daily using automated micro-sprinklers. Shoot length and number of nodes were determined every 30 days. In 2012, seedlings of the cultivars ‘Atago’, ‘Fuyu’, and ‘Triumph’ were used. Sowing, potting, and other experimental conditions were identical to the 2011 experiment, except that the irradiation periods were either 12 h/day or 20 h/day. Again, shoot length and number of nodes were determined at approximately 30-day intervals.

2.2. Experiment 2: effect of different LED wavelengths on plant growth

Seedlings of the cultivars ‘Atago’ and ‘Fuyu’ were used in 2012. Sowing and potting conditions were as described above in *Experiment 1*. Ten seedlings per cultivar and treatment were transferred to a growth chamber (LPH-220S equipped with a 3 LH-75DPS, NK system, Osaka, Japan) with blue (445 nm), red (660 nm), or mixed wavelength light (blue, 445 nm; green, 520 nm; red, 660 nm; ratio of wavelengths = 1 : 1 :1) (Fig. 1). A photoperiod of 20 h/day or 12 h/day was initiated at 59 d and 182 d after sowing, respectively. Environmental conditions in the growth chambers were set at 25 °C/20 °C (temperatures with a 12 h cycle) and 60% relative humidity; photosynthetic photon flux density was maintained at around 100 μmol $m^{-2}s^{-1}$ for the two treatments at plant level. Shoot length, number of nodes, internode length, stem diameter, number of leaf primordia, leaf thickness, leaf area, and degree of leaf curl were determined at 107 days for the 20 h/day treatment and at 122 days for the 12 h/day treatment. Scale leaves of the axillary buds were decorticated under a stereoscopic microscope, and the number of leaf primordia was determined.

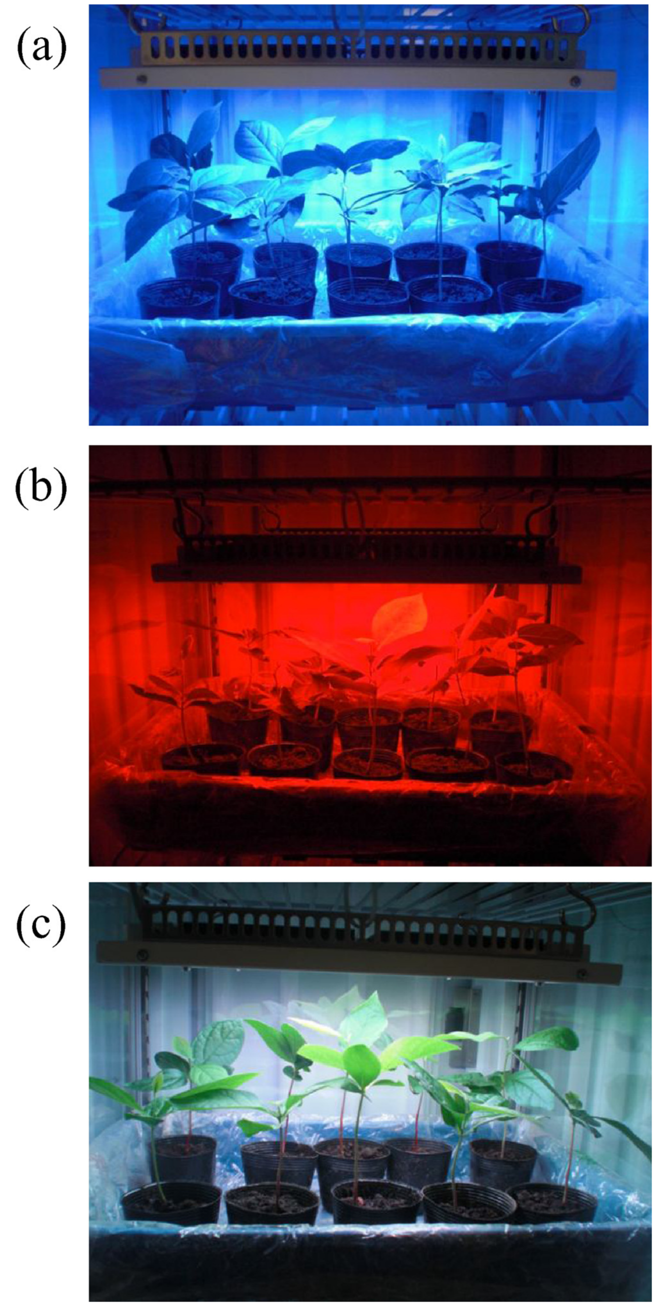

Fig. 1. Growth of Japanese persimmon seedlings under three different light treatments from light emitting diodes (LEDs): (a) blue, 445 nm; (b) red, 660 nm; (c) mixed (blue, 445 nm/ green, 552 nm/ red, 660 nm). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

2.3. Experiment 3: effects of time of grafting on the production of scions

‘Akiou’ was used as the scion for nursery stock production in an experiment using twice grafts per year (Fig. 2). Bark grafting was performed on 27 February 2012 on a two-year-old ‘Fuyu’ rootstock in a controlled glasshouse maintained at a minimum temperature of 18 °C.