

Plant Morphogenesis under Different Light Intensity

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Abstract. The innovative LED light source (Sun Box) with irradiation spectrum close to the sun spectrum in the wavelength range 440-660 nm was used in experiment for study the influence of light intensity (75, 135, 230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$) on the growth and development of plants. Standard fluorescent lighting was used as a control. The experiments were carried out on plantlets of *Stevia rebaudiana* and *Solanum tuberosum*, cvs. Snegir, Rozhdestvenskiy and Kamchatskii) *in vitro*. The illumination intensity of 75 and 230 $\mu\text{mol/s}\cdot\text{m}^2$ promoted development of *S. rebaudiana* plantlets with optimal values of morphometric parameters and well developed roots, which is important for plantlet adaptation to soil conditions. For *S. tuberosum* plantlets (Snegir and Rozhdestvenskiy cultivars), radiation intensity of 135 $\mu\text{mol/s}\cdot\text{m}^2$ was optimal for micropropagation. The illumination intensity of 230 $\mu\text{mol/s}\cdot\text{m}^2$ led to a formation of plantlets with the largest total fresh mass among experimental groups. Sun Box light with intensity of 75 $\mu\text{mol/s}\cdot\text{m}^2$ could be applied for micropropagation of these cultivars: plantlets were the highest with the largest internodes number. Thus, the plant response to different light intensity was species-specific, and – in case of potato plantlets – cultivar-specific. The use of artificial light sources with distinct PPFD level could be preferable for *S. tuberosum* and *S. rebaudiana* plantlet micropropagation *in vitro*, as it could shorten the cultivation time, accelerate cultivation time, and reduce the cost of electricity.

Introduction

Sunlight is a significant factor for plant life; it is absorbed by chlorophyll and used by the plant to construct the primary organic matter [1]. The light factor, along with the temperature factor, is one of the main exogenous modulators of the electrical plant activity [2, 3]. The connection of light with the structure of plants was revealed as early as the beginning of the last century [4], revealing its influence on morphometric indices. Light is characterized by several parameters: spectrum, intensity, etc. Each parameter has a directional effect, determining the features of morphogenesis. The absence of some wavelengths, performing various regulatory functions, in the irradiation spectrum of visible light, limits the realization of the photosynthetic potential of plants. Therefore, for the best plant growth, development, and maximum harvest the presence of all visible spectrum areas (with the predominance of red, green, blue and violet, and small amounts of ultraviolet and infrared wavelengths) is needed, when using artificial light sources as an alternative to sunlight. [5]. Light intensity also affects plant development.

Knowledge about the light influence on the development of the morpho-physiological plant parameters is necessary for modeling of growing conditions. It is particularly important in case of species significant for humans, such as potato (*Solanum tuberosum* Bullfinch) - a valuable agricultural species taking a third place in the world among the plants used for food. Another important species is *Stevia rebaudiana*, a medicinal plant producing natural sugars stevioside and rebaudioside A. Potentially, the plant can be used in the treatment of type 2 diabetes [6].

To date, valuable data on *in vitro* potatoe cultivation in different light conditions have been accumulated [5, 7-11]. With respect to stevia, studies on the effect of photoperiod [12, 13] and spectral composition of light on plant growth [14], synthesis and accumulation of steviosides were conducted. These studies were carried out with both intact plants [15-17] and plants *in vitro* [18].

The purpose of this work was to study the effect of different light intensities (75, 135, 230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$) on growth and development of *Stevia rebaudiana* and *Solanum tuberosum*, cvs. Snegir, Rozhdestvenskiy and Kamchatskii *in vitro* in order to select optimal light conditions for accelerated plant micropropagation.

Experimental

Investigation of the light intensity effect on plant growth was carried out in the Federal Scientific Center of the East Asia Terrestrial Biodiversity (FSCATB), Far Eastern Branch of the Russian Academy of Sciences (Vladivostok) in 2015-2018. For the experiments, plantlets of *Stevia rebaudiana* and non-viral potatoe plantlets (cv. Snegir, Rozhdestvenskiy and Kamchatskii early ripening variety) were used. The microcloning method was used for plant propagation. *Stevia* plantlets were obtained from seeds and placed in the collection of the FSCATB. The explants were placed in test tubes with agarized Murashige and Skoog nutrient medium [19] with some modifications. Plantlets were cultivated in controlled light-cultural conditions at 25 °C, 70% air humidity and 16 hours photoperiod.

For the experiment, a novel LED light source (Sun Box) was designed and manufactured in the Center of Laser Technologies of the Institute of Automation and Control Processes of the Far Eastern Branch of the Russian Academy of Sciences (IALS RAS, the Far Eastern Branch of the Russian Academy of Sciences). The emission spectrum of the Sun Box was close to the solar radiation spectrum in the wavelength range 440-660 nm and (Fig. 1). Four values of Sun Box light intensity, 75, 135, 230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$, were tested. The irradiation level was measured with the spectrophotometer (TKA, St. Petersburg). Fluorescent lamps with irradiation intensity of 49 $\mu\text{mol/s}\cdot\text{m}^2$ were used as control illumination. The experiment was carried out for 5 to 7 weeks for potato and stevia, respectively. Morphometric parameters such as plant height, length of internodes, leaf length and width, fresh root mass and green mass (shoots with leaves) were measured. The results were represented as mean with standard error of mean, and were processed using "Statistica" package version 10.0, the graphs were built in MS Excel 2010.

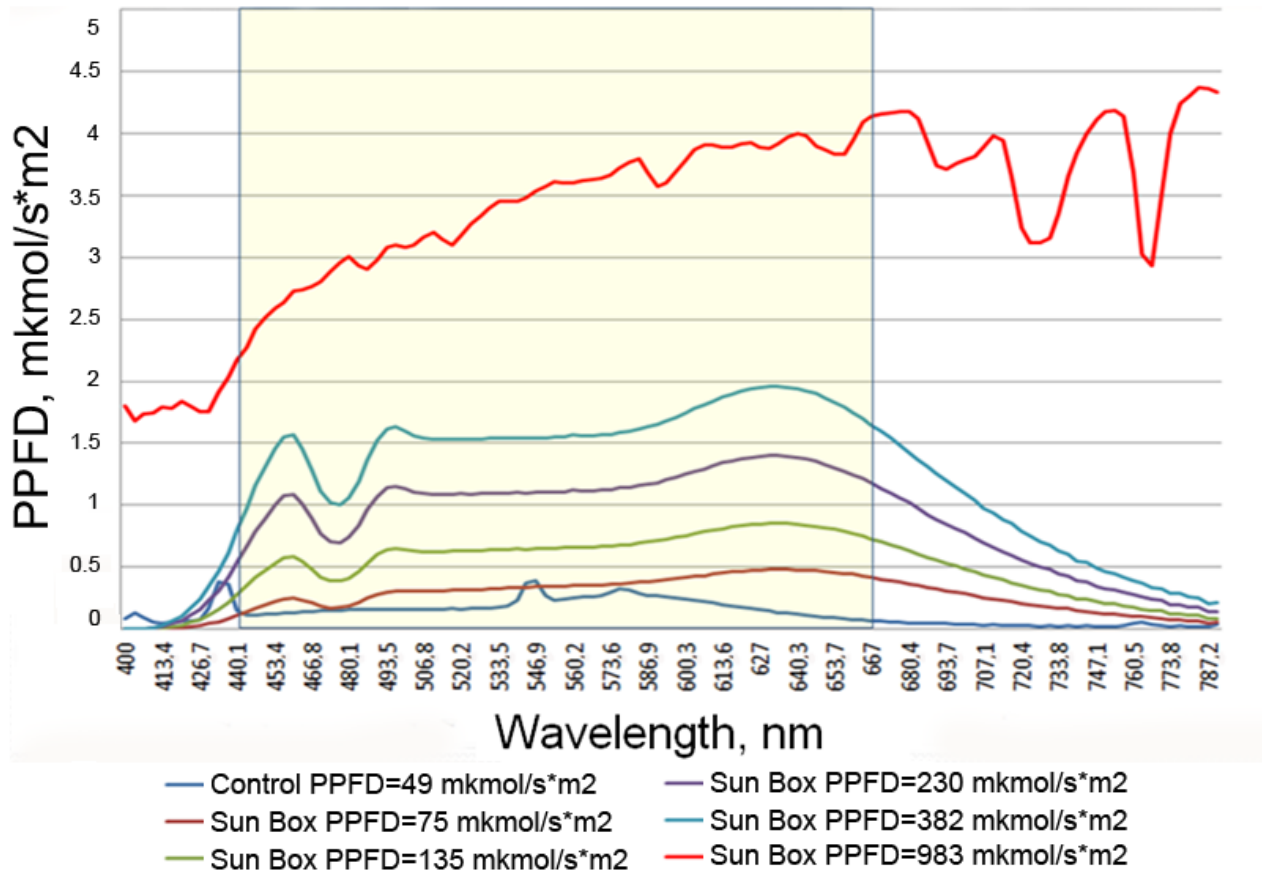


Fig. 1. Artificial LED Sun Box light spectra in the wavelength range 440-660 nm (highlighted in yellow) with different intensity. Control – luminescent lamps OSRAM L 36W/765. The solar spectrum measured in a summer cloudy day in Vladivostok is given for comparison [20].

Results

Cultivation of the *S. rebaudiana* plantlets at intensity level of $75 \mu\text{mol}/\text{s}\cdot\text{m}^2$ provided the development of plantlets with reliably higher levels of morphometric parameters, in comparison with control and other experimental groups (Fig. 2). Parameter values of plantlets cultivated at $135 \mu\text{mol}/\text{s}\cdot\text{m}^2$ did not differ from that of control plantlets, except for fresh green mass, which was significantly higher than in control. Similar data were obtained for plantlets at $230 \mu\text{mol}/\text{s}\cdot\text{m}^2$, but biomass accumulation in such plantlets was maximal, comparing with plantlets of other experimental groups and control. Light intensity level of $382 \mu\text{mol}/\text{s}\cdot\text{m}^2$ inhibited plant development, though caused intensive root formation.

Morphometric parameters of *S. tuberosum* plantlets (cvs. Snegir and Rozhdestvenskiy) at light intensity of $75 \mu\text{mol}/\text{s}\cdot\text{m}^2$ were equal to those of control plantlets. PPFD level of $135 \mu\text{mol}/\text{s}\cdot\text{m}^2$ provided the maximal values of height, leaf quantity and length, and fresh root mass; and total fresh mass values were 1.5 times higher than in control (Fig. 2). Plantlets at light intensity $230 \mu\text{mol}/\text{s}\cdot\text{m}^2$ and $382 \mu\text{mol}/\text{s}\cdot\text{m}^2$ were characterized with some lowering of growth parameters, comparing with control group, though the difference was statistically insignificant.

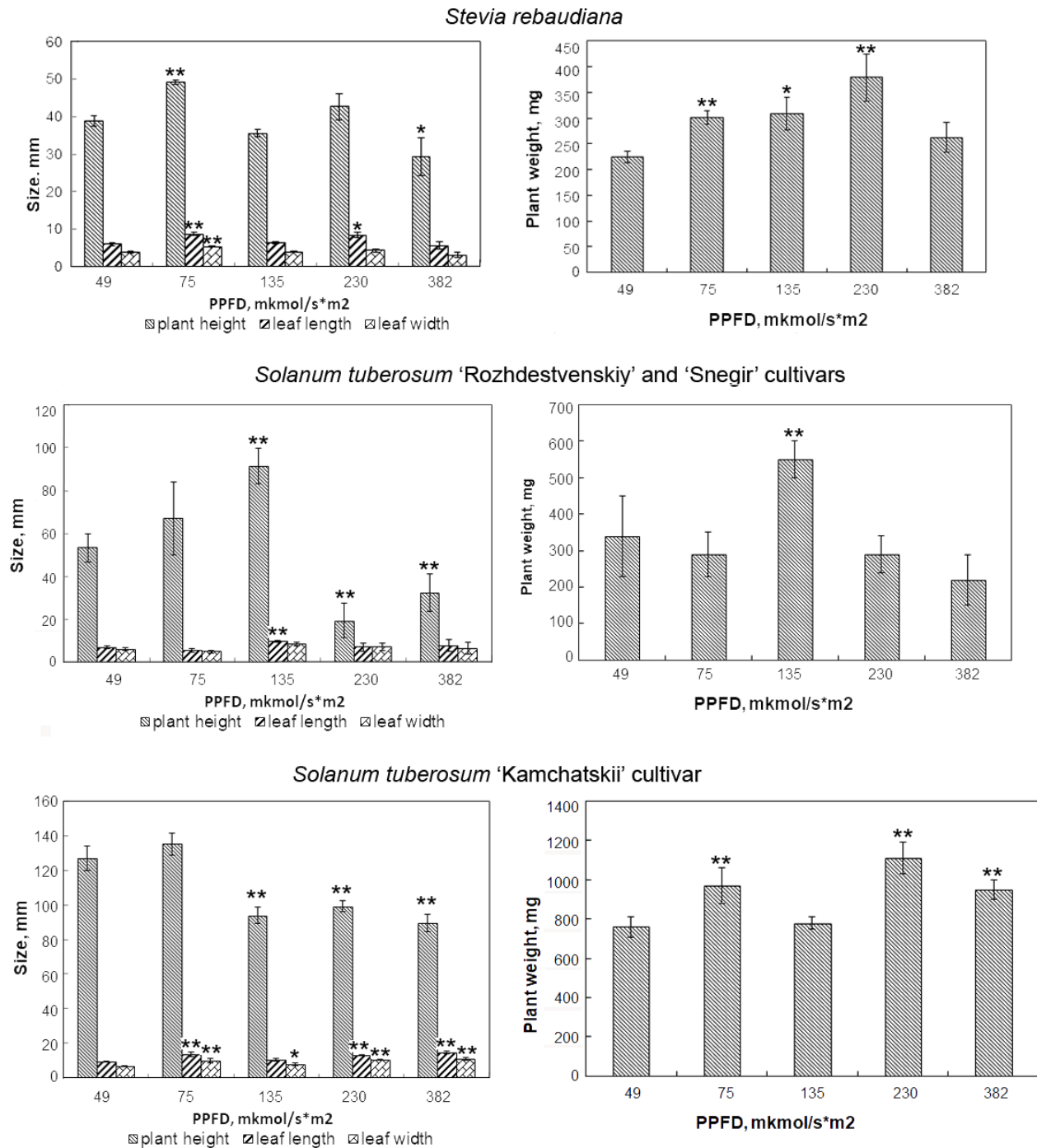


Fig. 2. Height of plantlets, leaf length and width, total fresh mass of *Stevia rebaudiana* and *Solanum tuberosum* (cvs. Snegir, Rozhdestvenskiy and Kamchatskii) plantlets under different Sun Box light intensities. * $P < 0.05$; ** $P < 0.01$, versus values of the control.

The results obtained for *S. tuberosum* (cv. Kamchatskii) showed that height of plantlets under PPFD values 135, 230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$ was significantly lower than in control group. The length of internodes of plantlets under 230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$ was reliably less than in control. Interestingly, substantially greater leaves length and width were observed for plantlets in all experimental groups, comparing with the control. Root fresh weight was substantially higher for plantlets in all experimental groups, in comparison with control, and was the highest for plantlets under 230 $\mu\text{mol/s}\cdot\text{m}^2$. The total fresh weight of plantlets grown at high light intensities (230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$) significantly exceeded the weight of the control plantlets by 1.5 and 1.25 times, respectively (Fig. 2).

Summary

Different LED Sun Box light intensities (75, 135, 230 and 382 $\mu\text{mol/s}\cdot\text{m}^2$) affected values of morphometric parameters of stevia and potato plantlets cultivated *in vitro*. In case of *S. rebaudiana*, the illumination intensity of 75 and 230 $\mu\text{mol/s}\cdot\text{m}^2$ promoted development of plantlets with optimal values of morphometric parameters and well developed roots, which could possibly adapt to soil conditions more effectively.

For *S. tuberosum* plantlets (cvs Snegir and Rozhdestvenskiy), radiation intensity of 135 $\mu\text{mol/s}\cdot\text{m}^2$ was optimal. The increase of root mass and intensive development of the aboveground parts was observed.

At the same time, cultivation of potato plantlets of cv Kamchatskii at intensity of 230 $\mu\text{mol/s}\cdot\text{m}^2$ led to formation of plantlets with the largest total mass among experimental groups. Plantlets grown at 75 $\mu\text{mol/s}\cdot\text{m}^2$ displayed characteristics favorable for micropropagation: had the highest height values and internodes number. For conditions of long-term cultivation in collection the intensity of 135 $\mu\text{mol/s}\cdot\text{m}^2$ was the most acceptable for potatoes of cv Kamchatskii, since the fitness of plantlets cultivated under such conditions was close to the control plantlets.

Thus, the application of artificial LED light source (Sun Box) seems promising for cultivation of stevia and potato plantlets *in vitro*. The plant response to different light intensity was species-specific, and, in case of potato plantlets, cultivar-specific. Varying the intensity parameters in the range of 60-100 $\mu\text{mol/s}\cdot\text{m}^2$ can have a positive or negative influence on the growth and development of individual organs and the whole plantlet. At the same time, cultivation of plantlets under the investigated light regimes, depending on the purpose of the experiment, could significantly reduce the labor costs of micropropagation, as well as reduce the cost of electricity.

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