Study of the Photosensitizer Methylene Blue in Plants Grown in Indoor Growth Chambers

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In the quest to improve agricultural quality and productivity, artificial light has been used in the form of photonic supplementation (photobiomodulation), biostimulating plants and influencing their development for optimal quality in hydroponic cultivation in agriculture. Understanding new forms of integrated management, as well as bioecology in hydroponic cultivation supplemented with optimized photobiomodulation, remains a significant challenge for the scientific community. Overcoming these challenges is essential to address the current expansion of agricultural sector activity and the risks related to emerging contaminants, among which pharmaceutical actives are included. The photosensitizing drug methylene blue (MB) is used in the treatment of infections by inactivating microorganisms and in photodynamic therapy processes. The objective of this study is to evaluate the effects of continuous exposure to residual concentrations of MB in indoor plant growth systems that rely on hydroponics supplemented with light. Butterhead lettuce seeds (*Lactuca sativa*) were germinated for 24 h in phenolic foam moistened with water at 23°C. After germination, the phenolic foam was transferred to individual baskets in an indoor cultivation chamber (LED Keisue Hydroponic Plants Growing Vertical Farm KES 2.0). The system included continuous fertigation in the floating mode of hydroponic nutrient solution (PM3 and PM4), supplemented with iron. Humidity and temperature control were set at 75% and 18°C, respectively. Photonic supplementation was carried out with light-emitting diodes (LEDs) in the proportion of 4:1:1:1 for red, green, blue, and white wavelengths, respectively, with a total photosynthetic photon flux density (PPFD) of 100 µmol/m^2/s. The photoperiod was 18:6 hours of light and dark, respectively. The seedlings were kept in two groups: control ($n = 10$) and MB ($n = 15$). The seedlings were cultivated for 34 days, continuously exposed to MB at a dose of 0.1-10 mg/ml, dissolved in the nutrient solution. Estimates of MB concentration in the nutrient solution were obtained by UV-Vis absorbance scanning. Chlorophyll analyses were estimated based on the SPAD value (five measurements per leaf, SPAD 502 Plus, Spectrum Technologies Inc., USA) and emission spectra of fluorescence measured *in vivo* using a portable fiber optic-based spectrofluorometer (MM Optics, Brazil). The former depends on light transmission after exposing the leaf to LEDs emitting light in the red and infrared regions. *In vivo* fluorescence involved excitation at 408 and 532 nm and capturing the emission spectra. Preliminary results show that plants grown under MB exposure had SPAD values of 20.2+/-3.4, significantly lower compared to the control, which had 29.2+/-3.0, respectively, indicating a loss of vigor. Fluorescence showed suppression at the evaluated FS concentrations, suppressing the emission of chlorophyll a. The plant's photosynthetic apparatus may have been altered by MB residue in the hydroponic fluid, affecting plant development and reducing *in vivo* chlorophyll a fluorescence. However, new studies are still needed to understand in more detail the process of MB incorporation in plants supplemented with artificial light.