



ARTEMISININ CONTENT IN *in vitro*-CULTURED PLANTS OF *Artemisia annua* L. DEVELOPED UNDER DIFFERENT LIGHT SPECTRA QUALITY.

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Abstract: Purpose of study: Artemisinin (ART) is the central compound in the most effective antimalarial treatment, however it is found in low concentrations in *Artemisia annua* [1]. Abiotic factors, as light, can influence the biosynthesis and optimize its production [2]. The aim of this study is to investigate the production of ART of *in vitro*-cultured *A. annua* developed under different light quality. Methods: CPQBA-1 seeds of *A. annua*, provided by Dr. Pedro Melillo (UNICAMP), were sown into a sterile medium. Plantlets were micropropagated in MS medium [3] without growth regulators, in dark (D – negative control), under white fluorescent light (W – positive control - 20 $\mu\text{mol}/\text{m}^2/\text{s}$) or different LED light conditions (10 $\mu\text{mol}/\text{m}^2/\text{s}$): yellow (Y), blue (B), green (G), red (R) by 16 hours of light, 25°C. 60 days plantlets without roots were dried in ventilated oven (40°C). 40mg of dried parts were extracted with 10mL of ethanol using sonication (45min). Extracts were evaporated than solubilized in 1mL of methanol. The procedure was performed in triplicate. LC-MS experiments were performed using a Flexar (PerkinElmer) system using C18 column (PerkinElmer, 150 \times 4.6mm, 3 μm) interfaced to an ESI and SQ 300 MS quadrupole. MS parameters were 12L.min⁻¹ (300°C) of drying gas, 80psi for nebulization. Mobile phase was water (A) and acetonitrile (B), both with 0.1% of formic acid, starting with 55% B (9min), increased to 100% B (1min), maintained 100% B (2min), decreased to 55% B (1min) and kept at 55% B (2min). Flow rate was 1.2mL.min⁻¹ of which 45% were sent to MS. Injection volume was 20 μL . SIM was used at positive mode at 283.1m/z for ART [M+H]⁺. A five points calibration curve (1.5 to 80 $\mu\text{g}\cdot\text{mL}^{-1}$) of ART (Sigma Aldrich) was obtained. Statistical analyses were performed on Statistica 7.0 software using ANOVA with Tukey post-hoc. Results: The results are shown in Figure 1. All treated groups produced less ART than W (p<0,0001) and more than D (p<0,0001). Among the treated groups, the plants grown in Y and in G do not showed statistical difference (p=0,9130); plants grown in B showed the higher production of ART (p<0,0001) and those grown in R showed the lower production (p<0,0006). Conclusions: The results suggest that ART can be enhanced by culturing the plants *in vitro* under a combination of W and B [4].

References:

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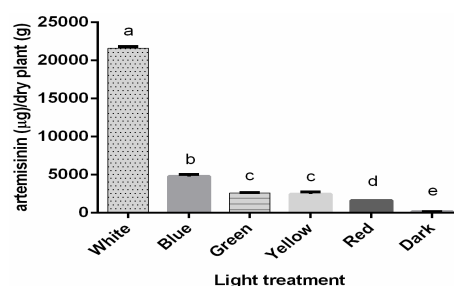


Figure 1 – Artemisinin content (µg) per gram of dry aerial parts for each light treatment (p<0,05).