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ACCLIMATIZATION OF MICROPROPAGATED Stevia rebaudiana (Bert.) PLANTLETS AND COMPARISON OF THEIR GROWTH AND STEVIOSIDE CONTENT WITH CONVENTIONALLY PROPAGATED PLANTS

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Abstract

Stevia rebaudiana (Family Asteraceae) is commercially cultivated to obtain non caloric sweet Steviol glycosides from leaves. Micropropagation through *in vitro* culture can be used to overcome limitations in conventional propagation methods. However micropropagated plantlets need gradual exposure to the natural environment for successful establishment in soil. Therefore a protocol was developed to determine the best acclimatization procedure. Micropropagated plantlets were initially transferred into coir pellets, then after four weeks they were transferred into different potting mixtures. Acclimatized and conventionally propagated plants with more or less similar morphology (approximately 10 cm) were planted in the same potting mixture and the growth parameters (shoot morphology and pigment contents) were compared. After four weeks on coir pellets, 76.0 % of micropropagated plantlets survived. The plantlets grown in sand: compost: coir dust (1:1:1) potting mixture showed the highest percentage of survival (90.0%) after four weeks. After one month of growth, conventionally propagated plants showed non significantly higher mean shoot length (14.70 ± 2.73 cm), mean number of leaves (17.30 ± 2.81) , total chlorophyll $(31.47 \pm 0.07 \text{ mg L})$ and total carotenoid $(5.10 \pm 0.09 \text{ mg L})$ than acclimatized plants. However, after three months of growth, acclimatized plants showed higher growth $(26.4 \pm 1.10 \text{ cm of mean shoot length}, 63.0 \pm 36.26 \text{ of mean number of leaves})$ and significantly higher pigment contents (42.25 \pm 0.22 mg L¹ of total chlorophyll and 7.15 \pm 0.05 mg L¹ of total carotenoid) than the conventionally propagated plants. After three months of growth Stevioside contents in both plant types were compared. According to HPLC analysis, Stevioside contents in acclimatized micropropagated plants (11.46 \pm 0.03 mg mL¹) was higher than the conventionally propagated plants $(10.13 \pm 0.03 \text{ mg mL}^{-1})$. Results suggests that this acclimatization procedure can be used to obtain genetically uniform better quality micropropagated S. rebaudiana plantlets for commercial scale plantations.

Keywords: Stevia rebaudiana, micropropagation, acclimatization, Stevioside, HPLC