6th World Congress on Stevia

Stevia Tasteful 2014

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Stevia Tasteful 2014: The Subtle Balance

Science, Formulation & Exhibition

June 19 - 20, 2014 Berlin, Germany

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IN VITRO CULTURE AND ACLIMATION PROCESS OF *STEVIA REBAUDIANA*

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Stevia rebaudiana (Bert.) Bertoni (Asteraceae) leaves are a natural source of steviol glycosides, which can be used for sweetening and flavouring foods and beverages.

The recent interest of farmers on this crop has increased the demand for propagation material. The farmers can use seedlings, cuttings or, alternatively, wealthy plants obtained from in vitro propagation (Madan et al., 2010). This work reports the results of a simple, fast and non-expensive method developed for micro-propagation, rooting and acclimation of Stevia.

Objectives: development of a method to micro-propagate, rooting and acclimate Stevia.

Methodology: plant material was collected from a commercial clone growing in field. Plant material was sterilized by stirring for 7 minutes in a solution of chlorine 5% plus 10 drops of tween 80 per 100 mL of chlorine solution. Thereafter, the explants were washed in sterilized water and moved to a solution of ethanol 70%, kept there for 5 minutes, washed again and inoculated in two different culture media (medium A – MS without hormones and 20 g/L of sucrose; and medium B – MS with 0.5 mg/L of kinetin and 20 g/L of sucrose). The multiplication rate was determined for each subculture of 2 months in the two media tested. As complementary data, the fresh weights of 10% of the micro-propagated plants per subculture were determined. The rate of spontaneous rooting was also determined, and trials of plant rooting performed through auxin shocks using 2mg/ mL of IBA (Indole-3-butyric acid)(Abdullateef and Osman,2012), a solution to dip the basal part of 100 Stevia plants for 30 seconds. Thereafter, the plants were transferred to the MS media without hormones but containing activated coal. The time that the plants take to get a minimum of roots allowing them to acclimate to the soil substrate was recorded. The acclimatization in soil substrate was performed in a greenhouse under a misting irrigation system working for 10 seconds each 20 minutes. The acclimatization rate was determined during two weeks.

Results: In culture media A and B the monthly multiplication rates were 383.2 and 306.9%, respectively. The mean plant fresh weights per subculture were 1.1 and 0.9 g, respectively for A and B subculture media. In medium A, the rate of spontaneous rooting, after 4 months in culture, was 10.5%, and in B medium was less than 4%. Rooting induced process was evaluated each week after hormone shock. In the first week, 30% of plants developed roots, having increased this number to 70% in the second week. No significant differences were found in rooting of plants coming from the different initial media growth. The acclimatization rate was 100% after 2 weeks in the greenhouse.

Conclusion: Using the medium A (the best suited for multiplication and spontaneous rooting) it was possible to obtain more than 50 times the initial number of plants in excellent developing conditions to be transferred to the field.

References: Abdullateef R, Osman M, (2012) International Journal of Biology, Vol. 4, No. 1; pp 146-153
Madan S., et al (2010), Indian Journal of Natural Products and Resources, Vol1 (3), pp 267-286
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