In vitro SHOOT MULTIPLICATION AND ROOTING OF VANILLA (Vanilla fragrans)

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This study was conducted in tissue culture laboratory at Export Agriculture Research Station in Matale from 28th April to 31st October with an objective to investigate a suitable micropropagation protocol for *in vitro* shoot multiplication and rooting in *Vanilla fragrans* (Salisb) Ames. A two factor factorial experiment was arranged in Complete Randomized Design consisted with eight treatments having three replicates for each.

Single nodal segments of 1.5 – 2 cm in length excised from *in vitro* grown plantlets were used as explants. The explants were cultured in Murashige and Skoog (MS) basal medium and Nitsch and Nitsch (N69) basal medium supplemented with four concentrations of 6-benzyladenine (0.0, 0.5, 1.0 and 2.0 mg/l) for shoot multiplication. Percentage of live and contaminations, number of shoots, shoot length, number of leaves and overall appearance were recorded in two weeks interval for a period of 12 weeks. A higher survival rate (95.6%) was observed in all treatments giving a very low percentage of contaminations (1.9%). Murashige and Skoog medium with 0.0, 0.5, 1.0 and 2.0 mg/l BA gave 1, 7.7, 6 and 5.4 shoots per explant respectively while Nitsch and Nitsch medium with 0.0, 0.5, 1.0 and 2.0 mg/l BA produced 1, 3.4, 6 and 5.3 shoots per explant. The highest shoot multiplication was recorded from the explants grown in MS medium with 0.5 mg/l BA (7.7 shoots).

There was no treatment effect on shoot length, number of leaves and the overall appearance. Then two forms of the Murashige and Skoog medium (solid and agitated liquid media) with 0.5 mg/l BA was compared for shoot multiplication from nodal explants. The Percentage of live and contaminations, number of shoots, shoot length, number of leaves and overall appearance were recorded in two weeks interval for a period of 12 weeks. A higher percentage (80%) of live cultures and lower percentage (20%) of contaminations were observed in all treatments. Results indicated the highest shoot multiplication in solid MS medium (6.7 shoots per explant) than the liquid medium (1 shoot per explant). The highest overall appearance was also observed in the soil medium (7.4) over the liquid medium (3). But the status of the medium was not effective for the shoot length and the number of leaves.

The micro shoots of 2 cm in length were transferred to Murashige and Skoog medium with combination of two level of indole-3-butyric acid (0.0 and 0.2 mg/l) and two levels of activated charcoal (0 and 2 g/l). Number of roots, root length and shoot length were

recorded after six weeks of culturing. Indole-3-butyric acid and activated charcoal had no effect on the number of roots and shoot length. But the highest root length (3.6 cm) was observed when activated charcoal was incorporated in the medium than without activated charcoal (2.6 cm).

Based on the result of the study the suitable protocol for vanilla shoot multiplication and rooting is solid MS medium supplemented with 0.5 mg/l BA for shoot multiplication and MS medium with 2 g/l activated charcoal for rooting.

Key words: *Vanilla fragrans* (Salisb) Ames, Benzyladenine, Indole butric acid, Shoot multiplication, Rooting, protocol