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In vitro multiple shoot and root induction of *Zeuxine flava* (Sandaraja)

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Zeuxine flava (Sandaraja) is an important medicinal plant in Ayurvedic medicine system of Sri Lanka. It is being used for treatment of snakebites. This plant is listed under threatened category in 1999 List of threatened flora and fauna (Red list). Vegetative propagation methods have limited success and pod development frequency is very low. However seed germination under natural conditions have

never been observed. Therefore it is important to develop a micro propagation protocol for mass propagation of this important medicinal plant.

Apical meristems and nodal segment explants were tested for induction of shoots *in vitro*. Explants were surface sterilized using 10% Clorox (Sodium hypochlorite 5.25% v/v) and 70% alcohol each followed by three successive washings in sterile distilled water. Three different basal media – WPM, MS and KC were used with different concentrations of BAP, IAA and 2,4-D. WPM containing 2.25 mg/L BAP and 1.0 mg/L 2,4-D induced shoot buds from both axillary and apical buds after six weeks of incubation under 16h and 8hr dark photoperiod at 25 ± 1^0C. None of the other tested media induced buds from explants. When concentrations of the basal medium were lowered to 1/2, 1/5, and 1/10, time taken for bud induction was reduced.

Elongated shoots were transferred into MS medium-supplemented 0.1 mg/L NAA, 1.0 mg/L BAP and 1.0 mg/L kinetin. Within four weeks, axillary buds of *in vitro* produced shoots started to initiate.
Explants, which were maintained in, shoot induction medium, after 8 subcultures were able to produce root nodules as well as hairy roots without multiple shoots. When elongated shoots were transferred into MS medium supplemented with 0.1 mg/L NAA, 1.0 mg/L BAP and 1.0 mg/L kinetin root nodules were formed. However when elongated shoots were transferred into shoot induction medium during 4 weeks of incubation they were able to produce root nodules.

According to the results obtained 1/10 WPM medium supplemented with 2.25 mg/L BAP and 1.0 mg/L 2,4-D was the best medium for bud induction. However MS medium supplemented with 0.1 mg/L NAA, 1.0 mg/L BAP and 1.0 mg/L kinetin was the most suitable medium for multiple shoot induction. 1/10 WPM medium supplemented with 2.25 mg/L BAP and 1.0 mg/L 2,4-D found to be the best medium for root induction

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