

In Vitro Propagation of Vanilla Planifolia Using Nodal Explants and Varied Concentrations of Naphthaleneacetic acid (NAA) and 6-Benzylaminopurine (BAP).

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Abstract : Background: Vanilla planifolia is the only edible fruit of the orchid family (Orchidaceae) among the over 35,000 Orchidaceae species found worldwide. In Ghana, Vanilla was discovered in the wild, but it is underutilized for commercial production, most likely due to a lack of knowledge on the best NAA and BAP combinations for in vitro propagation to promote successfully regenerated plant acclimatization. The growing interest and global demand for elite Vanilla planifolia plants and natural vanilla flavour emphasize the need for an effective industrial-scale micropropagation protocol. Tissue culture systems are increasingly used to grow disease-free plants and reliable in vitro methods can also produce plantlets with typically modest proliferation rates. This study sought to develop an efficient protocol for in vitro propagation of vanilla using nodal explants by testing different concentrations of NAA and BAP, for the proliferation of the entire plant. Methods: Nodal explants with dormant axillary buds were obtained from year-old laboratory-grown Vanilla planifolia plants. MS media was prepared with a nutrient stock solution (containing macronutrients, micronutrients, iron solution and vitamins) and semi-solidified using phytigel. It was supplemented with different concentrations of NAA and BAP to induce multiple shoots and roots (0.5mg/L BAP with NAA at 0, 0.5, 1, 1.5, 2.0mg/L and vice-versa). The explants were sterilized, cultured in labelled test tubes and incubated at $26^{\circ}\text{C} \pm 2^{\circ}\text{C}$ with 16/8 hours light/dark cycle. Data on shoot and root growth, leaf number, node number, and survival percentage were collected over three consecutive two-week periods. The data were square root transformed and subjected to ANOVA and LSD at a 5% significance level using the R statistical package. Results: Shoots emerged at 8 days and roots at 12 days after inoculation with 94% survival rate. It was discovered that for the NAA treatments, MS media supplemented with 2.00 mg/l NAA resulted in the highest shoot length (10.45cm), maximum root number (1.51), maximum shoot number (1.47) and the highest number of leaves (1.29). MS medium containing 1.00 mg/l NAA produced the highest number of nodes (1.62) and root length (14.27cm). Also, a similar growth pattern for the BAP treatments was observed. MS medium supplemented with 1.50 mg/l BAP resulted in the highest shoot length (14.98 cm), the highest number of nodes (4.60), the highest number of leaves (1.75) and the maximum shoot number (1.57). MS medium containing 0.50 mg/l BAP and 1.0 mg/l BAP generated a maximum root number (1.44) and the highest root length (13.25cm), respectively. However, the best concentration combination for maximizing shoot and root was media containing 1.5mg/l BAP combined with 0.5mg/l NAA, and 1.0mg/l NAA combined with 0.5mg/l of BAP respectively. These concentrations were optimum for in vitro growth and production of Vanilla planifolia. Significance: This study presents a standardized protocol for labs to produce clean vanilla plantlets, enhancing cultivation in Ghana and beyond. It provides insights into Vanilla planifolia's growth patterns and hormone responses, aiding future research and cultivation.

Keywords : Vanilla planifolia, In vitro propagation, plant hormones, MS media

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