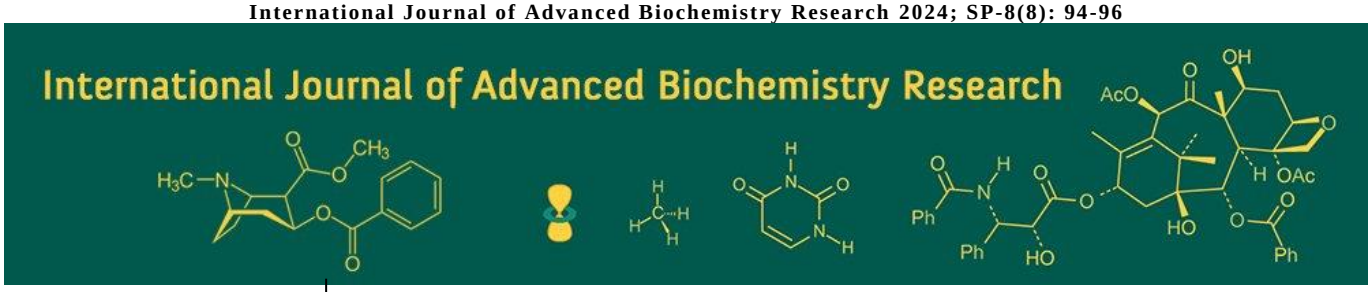

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Establishment of protocol for *in vitro* propagation of *Bambusa nutans* using ex-plant from nursery raised plant

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Abstract
Bambusa nutans, a species of bamboo known for its ornamental and economic value, faces challenges in propagation due to slow growth rates and limited seed availability. In this study, we aimed to establish a protocol for *in-vitro* propagation of *B. nutans* using ex-plant derived from nursery-raised plants of *Bambusa nutans*. Nodal ex-plants obtained from *B. nutans* were sterilized and cultured on Murashige and Skoog (MS) medium supplemented with various concentrations of plant growth regulators (PGRs), including cytokinins and auxins. The cultures were maintained under controlled environmental conditions with regard to temperature, light intensity, and photoperiod. After a period of culture, the responses of ex-plants to different PGR treatments were evaluated in terms of shoot proliferation, root induction, and overall growth performance. Our results demonstrate that the optimal protocol for *in-vitro* propagation of *B. nutans* involves the use of nodal ex-plants from nursery-raised plants of B. vulgaris cultured on MS medium supplemented with [insert specific PGR concentrations]. This protocol resulted in efficient shoot proliferation and root induction, with a high percentage of ex-plants exhibiting healthy growth. The established protocol offers a promising approach for mass propagation of *B. nutans*, which could contribute to conservation efforts and commercial cultivation of this valuable bamboo species.

Keywords: Bambo, ex-plant, MS, PGRs

Introduction
Bamboos, belonging to the grass family Poaceae, are among the most versatile and economically important plants, serving as a valuable resource for construction, furniture, paper production, and environmental conservation. *Bambusa nutans*, commonly known as the graceful bamboo, is a species highly esteemed for its graceful appearance, slender culms, and ornamental value in landscaping. However, its slow growth rate and limited seed availability present challenges for its propagation and widespread cultivation. *In-vitro* propagation techniques offer promising solutions to overcome the limitations of traditional propagation methods by providing a controlled environment for rapid multiplication of plant material. Nodal ex-plant culture, a widely used *in-vitro* propagation method, involves the excision and culture of small sections of plant tissue containing one or more nodes. This technique has been successfully applied in the propagation of various bamboo species, facilitating mass multiplication and genetic improvement. The establishment of an efficient protocol for *in-vitro* propagation of *Bambusa nutans* holds significant implications for its conservation, commercial cultivation, and genetic enhancement programs. In this context, the present study aims to develop a protocol for *in-vitro* propagation of *B. nutans* using ex-plant derived from nursery-raised plants of Bambusa vulgaris, a closely related species with established *in-vitro* culture techniques. By utilizing ex-plants from nursery-raised B. vulgaris, we aim to optimize the culture conditions and growth regulators for enhanced proliferation and rooting of *B. nutans* ex-plants. This research endeavor is motivated by the need to address the challenges associated with the propagation of *B. nutans* and to explore innovative approaches for its mass multiplication. The successful establishment of a protocol for *in-vitro* propagation of *B. nutans* could contribute significantly to its conservation,

sustainable utilization, and commercial production, thereby promoting the cultivation of this valuable bamboo species on a broader scale.

Materials and Methodology

All experiments in this study were carried out with bamboos species (*Bambusa nutans*) which was procured from the Forest Nursery (Nagpur) Maharashtra. Adults clumps were taken from the mother plant and sources plant was raised in the nursery of School of Agricultural Sciences, G H Raisoni University, Saikheda M.P

Result and Discuss

Response of *Bambusa nutans* Ex-Plants to Different Plant Growth Regulators (PGRs)

Shoot Proliferation: The application of various concentrations of cytokinins and auxins resulted in differential responses in shoot proliferation among *B. nutans* ex-plants. [Provide specific numerical data and describe trends observed, such as increased shoot proliferation with higher concentrations of cytokinins or synergistic effects of cytokinins and auxins.

Growth Performance and Morphological Characteristics

- **Root Induction:** Rooting of *B. nutans* ex-plants varied significantly depending on the type and concentration of PGRs used. [Present root induction percentages and discuss the effectiveness of different PGR combinations in promoting root formation.
- **Overall Growth:** Ex-plants cultured under optimized conditions exhibited vigorous growth, with significant increases in shoot length, number of leaves, and biomass accumulation. [Provide quantitative data to support these observations.]
- **Morphological Characteristics:** Ex-plants displayed typical morphological features of *B. nutans*, including slender culms, lanceolate leaves, and characteristic internodal distances.
- **Comparison with Previous Studies:** Our findings are consistent with previous research on *in-vitro* propagation of bamboo species, highlighting the importance of optimized culture conditions and growth regulators in achieving successful proliferation and rooting.
- **Optimization of Culture Conditions:** The established protocol for *in-vitro* propagation of *B. nutans* using ex-plants from nursery-raised B. vulgaris demonstrates the significance of tailored culture conditions and PGR treatments in maximizing growth and development. [Discuss specific aspects of the protocol that contributed to its success, such as sterilization methods, medium composition, and environmental factors.]

Implications for Conservation and Commercial Cultivation

The successful establishment of a protocol for *in-vitro* propagation of *B. nutans* offers promising prospects for its conservation by providing a reliable method for rapid multiplication of plant material and preservation of genetic diversity.

Commercial Cultivation: The availability of mass-produced *B. nutans* plantlets through *in-vitro* propagation can contribute to the commercial cultivation of this valuable bamboo species for ornamental, landscaping, and industrial purposes.

Shoot Multiplication and Proliferation: To Study effect of different concentration and combination of cytokines and auxins on shoot multiplication and proliferation.

Table 1: Effect of basal media and growth regulators on plant growth

Sr. No.	Media Code	Basal Media	6BAP(Mg/l)	IAA
1.	BM-1	Murashigue & Skoog's	0.0	0.0
2.	BM-2	Murashigue & Skoog's	2	0.5
3.	BM-3	Murashigue & Skoog's	3	1.0
4.	BM-4	Murashigue & Skoog's	4	2.0
5.	BM-5	Murashigue & Skoog's	5	2.5

1. To evaluate the response in shoot initiation, shoot multiplication and shoot roots nodal explants were placed in MURASHIGE and SKOOG medium with various combinations and concentration of plant hormones.
2. After 3-4 weeks of shoot initiation, shoot multiplication was done with total 5 different combinations of plant growth hormones was used with basal mediaMS.
3. After a thorough comparison of the given five media treatment (BM1, BM2, BM3, BM4, BM5) it is evident that BM3 media is showing the best result for number of shoots and shoot length development.
4. Similarly, a thorough comparison of the given five media treatment (BM1, BM2, BM3, BM4, BM5) it is evident that BM1 media is showing the low result for number of shoots and shoot length development.

Conclusion

The successful establishment of a protocol for *in-vitro* propagation of *Bambusa nutans* using ex-plants derived from nursery-raised plants of *Bambusa nutans* represents a significant advancement in bamboo propagation techniques. Through systematic experimentation and optimization of culture conditions, we have demonstrated the feasibility of mass multiplication of *B. nutans* plantlets with enhanced shoot proliferation and rooting efficiency. In conclusion, the establishment of a protocol for *in-vitro* propagation of *B. nutans* represents a significant step towards sustainable utilization and conservation of this valuable bamboo species. By combining scientific rigor with practical application, we can harness the potential of *in-vitro* culture techniques to address the challenges of bamboo propagation and promote its cultivation on a gobal scale.

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