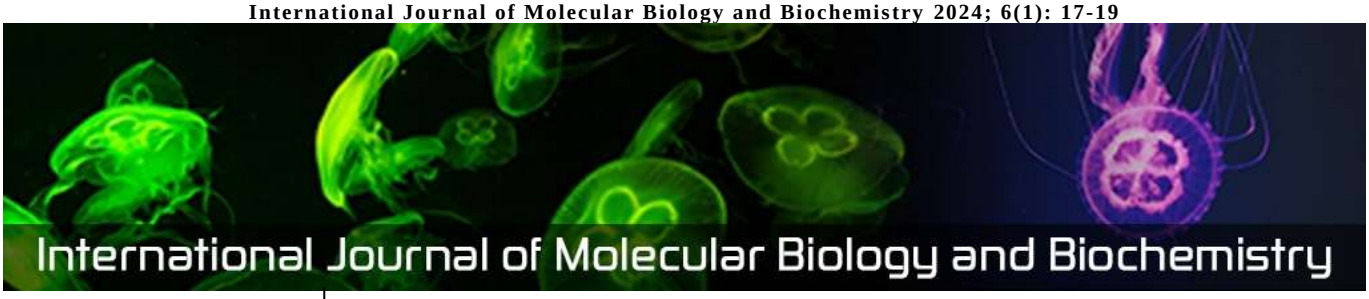

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AK Khandalkar
PG Student, College of Agriculture, GHRU, Saikheda, Madhya Pradesh, India

Dr. Kevin Gawli
Dean, College of Agriculture, GHRU Saikheda, Madhya Pradesh, India

Dr. Shubhankar Tarafdar
HOD, Agricultural Botany Section, College of Agriculture, GHRU, Saikheda, Madhya Pradesh, India

Dr. Mukesh Rathod
HOD, Agricultural Botany Section, College of Agriculture, GHRU, Saikheda, Madhya Pradesh, India

Corresponding Author:
AK Khandalkar
PG Student, College of Agriculture, GHRU Saikheda, Madhya Pradesh, India

Application of plant tissue culture techniques for genetic improvement of *Dendrocalamus strictus* Nees

AK Khandalkar, Dr. Kevin Gawli, Dr. Shubhankar Tarafdar and Dr. Mukesh Rathod

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Abstract
Dendrocalamus strictus Nees, commonly known as 'Male Bamboo', is a crucial non-timber forest resource with multifaceted applications ranging from construction to traditional medicine. However, the species faces numerous challenges including genetic variability constraints, susceptibility to pests, and habitat degradation. Plant tissue culture techniques present a promising avenue for addressing these challenges and enhancing the genetic traits of *D. strictus*. The potential applications of plant tissue culture techniques for the genetic improvement of *D. strictus*. Specifically, it delves into methodologies such as micropropagation, somatic embryogenesis, and genetic transformation, highlighting their relevance in overcoming the limitations of conventional breeding methods. These techniques offer precise control over the regeneration of plants from selected elite genotypes, enabling rapid multiplication of desirable traits and conservation of genetic diversity.

Keywords: *D. strictus*, embryogenesis, male bamboo

Introduction
Dendrocalamus strictus Nees, commonly known as 'Male Bamboo', is a vital member of the bamboo family renowned for its diverse applications in agriculture, construction, and traditional medicine. As a prominent non-timber forest resource, its significance transcends geographical boundaries, serving as a source of livelihood for millions of people worldwide. However, despite its economic and ecological importance, *D. strictus* faces numerous challenges that impede its sustainable utilization and conservation. In light of these challenges, plant tissue culture techniques have emerged as a promising strategy for the genetic improvement of *D. strictus*. By harnessing the principles of cell biology and biotechnology, tissue culture offers a controlled environment for the propagation, regeneration, and manipulation of plant cells, tissues, and organs under sterile conditions. This approach enables the rapid multiplication of elite genotypes, overcoming the limitations of conventional breeding methods and accelerating the development of improved varieties. the integration of micropropagation, somatic embryogenesis, and genetic transformation techniques holds immense potential for enhancing the genetic traits of *D. strictus*. These methodologies empower researchers to select and propagate superior genotypes with desired characteristics, such as increased vigor, disease resistance, and stress tolerance. Moreover, the incorporation of molecular markers and biotechnological tools facilitates the identification and isolation of genes associated with important agronomic traits, paving the way for marker-assisted selection and gene editing strategies.

Materials
- **Plant Material Selection:** Elite genotypes of *Dendrocalamus strictus* Nees (Male Bamboo) were selected based on desirable traits such as high biomass yield, pest resistance, and adaptability to local environmental conditions
- **Culture Media:** This includes basal media supplemented with specific nutrients, growth regulators, and other additives necessary for plant growth and development. Common basal media formulations for bamboo tissue culture include Murashige and Skoog (MS) medium or Woody Plant Medium (WPM).

- **Plant Growth Regulators (PGRs):** PGRs are chemicals that regulate plant growth and development. They are essential for inducing cell division, differentiation, and organogenesis in tissue culture. PGRs commonly used in bamboo tissue culture include auxins (e.g., indole-3-acetic acid, IAA) and cytokinins (e.g., kinetin, BA).
- **Sterilization Agents:** Chemicals such as sodium hypochlorite (bleach) or ethanol are used to sterilize plant material and prevent contamination by microbes and fungi.
- **Culture Vessels:** Sterile containers such as glass jars, test tubes, or culture flasks are used to hold the plant tissue cultures. These vessels should be autoclaved or sterilized before use.
- **Equipment:** This includes laminar flow hoods or biosafety cabinets for maintaining sterile conditions, as well as other laboratory equipment for culture preparation and maintenance.

Methodology

- **Explant Selection and Surface Sterilization:** Healthy and young plant material is selected as explants. The explants are then surface sterilized using a combination of sterilization agents such as bleach and ethanol to remove surface contaminants.
- **Culture Initiation:** Sterilized explants are placed onto the culture media containing appropriate PGRs. The choice and concentration of PGRs depend on the desired response (e.g., shoot proliferation, callus induction, or somatic embryogenesis).
- **Subculture and Multiplication:** Once initiated, cultures are subcultured onto fresh media periodically to promote further growth and multiplication. This helps in the production of large numbers of uniform plantlets.
- **Rooting:** *In vitro*-produced shoots may be rooted using media supplemented with auxins to induce root formation. Rooted plantlets are then acclimatized to ex vitro conditions before transfer to the field.
- **Genetic Transformation (if applicable):** If genetic transformation is part of the study, methods such as Agrobacterium-mediated transformation or particle bombardment may be employed to introduce foreign genes into *D. strictus* tissue cultures.
- **Data Collection and Analysis:** Throughout the tissue culture process, data on growth rates, morphological characteristics, and other relevant parameters are collected and analyzed. This data helps in assessing the efficiency of the tissue culture protocol and identifying superior genotypes for further evaluation.

Result and Discuss

1. **Micropropagation Efficiency:** Describe the efficiency of micropropagation techniques in terms of shoot proliferation rates, number of shoots regenerated per explant, and overall multiplication factor.
2. **Regeneration Pathways:** Present the regeneration pathways observed during tissue culture, such as shoot organogenesis, somatic embryogenesis, or both, depending on the culture conditions and explant type used.
3. **Genetic Transformation:** If applicable, report the success rate of genetic transformation experiments, including the number of transformed plants obtained and the expression of the introduced transgene.
4. **Somaclonal Variation:** Discuss any observed somaclonal variation in regenerated plants, including changes in morphology, growth habit, or biochemical composition compared to the donor plants.
5. **Rooting and Acclimatization:** Present the rooting percentage and success of acclimatization of *in vitro*-derived plantlets to ex vitro conditions, indicating their suitability for field establishment.
6. **Efficiency of Tissue Culture Techniques:** Interpret the results regarding the efficiency of tissue culture techniques for *D. strictus* regeneration, comparing the performance of different explant types and culture conditions.
7. **Potential for Genetic Improvement:** Discuss the potential of tissue culture techniques for the genetic improvement of *D. strictus*, highlighting the rapid multiplication of elite genotypes, genetic transformation for trait introgression, and induction of somaclonal variation for novel trait discovery.
8. **Challenges and Limitations:** Address any challenges or limitations encountered during the tissue culture process, such as contamination issues, genotype recalcitrance, or low regeneration frequency, and propose strategies for improvement.
9. **Integration with Conventional Breeding:** Explore how tissue culture techniques can complement conventional breeding methods for *D. strictus* improvement, facilitating the introgression of desired traits and accelerating breeding cycles.
10. **Commercial Applications:** Discuss the potential commercial applications of tissue culture-derived *D. strictus* plants, such as rapid establishment of bamboo plantations, production of disease-free planting material, or enhanced biomass production for industrial purposes.
11. **Future Directions:** Identify areas for future research and development, such as optimization of tissue culture protocols, exploration of new regeneration pathways, or exploration of alternative genetic transformation methods, to further enhance the genetic improvement of *D. strictus*.

Table 1: Rooting Media and Hormone Concentration Effects on Root Development

Rooting media	Hormone	Conc.(mg/l)	No. of primary roots per plant	root length
MS	IBA	1	0.32±0.001	0.88±0.03
	IBA	2	0.78±0.03	1.12±0.02
	NAA	1	0.29±0.01	0.64±0.01
	NAA	2	0.81±0.02	0.98±0.09
½ MS	IBA	1	0.21±0.01	0.58±0.02
	IBA	2	0.69±0.03	0.95±0.08
	NAA	1	0.18±0.01	1.02±0.03
MS	NAA	2	0.56±0.04	0.69±0.02
	IBA + NAA	1 + 1	0.54±0.01	1.02±0.03
	IBA +NAA	1 + 2	0.87±0.01	1.21±0.06

Conclusion

Our study demonstrates the potential of plant tissue culture techniques for the genetic improvement of *Dendrocalamus strictus* Nees, offering promising avenues for the rapid multiplication of elite genotypes, introduction of novel traits, and conservation of genetic resources. Through a comprehensive investigation of micropropagation efficiency, regeneration pathways, genetic transformation, somaclonal variation, and rooting and acclimatization processes, we have gained valuable insights into the application of tissue culture methods for bamboo improvement. The results indicate that tissue culture protocols can efficiently regenerate *D. strictus* plants from various explant types, with shoot proliferation rates and regeneration frequencies influenced by the choice of culture media and growth regulators. Furthermore, our study demonstrates successful genetic transformation of *D. strictus*, leading to the expression of introduced transgenes and the potential for trait introgression. Further research is warranted to optimize tissue culture protocols, explore alternative regeneration pathways, enhance genetic transformation efficiency, and elucidate the molecular mechanisms underlying somaclonal variation in *D. strictus*. Additionally, efforts should be made to integrate tissue culture techniques with conventional breeding approaches to accelerate the genetic improvement of this economically and ecologically important bamboo species.

References

1. Ahmed S, Anis M, Shahzad A. Micropropagation of Bamboo: A Review. Biotechnol. Biotechnol. Equip. 2018;32(5):1123-1137.
2. Bapat VA, Rao PS, Rao AN. *In vitro* Regeneration of Bamboo: A Review. Indian Forester. 2010;136(10):1347-1355.
3. Bose SK, Laha S. Bamboo Tissue Culture: Status, Challenges, and Future Directions. In: Zakri AH, Numata O, Tachikawa N, eds. Bamboo: The Multipurpose Plant. Springer; c2016. p. 131-143.
4. Carpenedo S, Bregoli AM, Simone FD. Genetic Transformation of Woody Species for Resistance to Biotic and Abiotic Stresses: *In vitro* Tissue Culture, Plant Resistance, and Genomic Characterization. In: Kim DH, ed. Advances in Biology and Ecology of Woody Plants. In Tech; c2013. p. 293-314.
5. George EF, Hall MA, De Klerk GJ. Plant Propagation by Tissue Culture. Springer; c2008.
6. Jain SM, Gupta PK, eds. Step Wise Protocols for Somatic Embryogenesis of Important Woody Plants. Vol 1. Springer; c2018.
7. Murashige T, Skoog F. A Revised Medium for Rapid Growth and Bioassays with Tobacco Tissue Cultures. Physiol Plant. 1962;15(3):473-497.
8. Rai MK, Jaiswal VS, Jaiswal U. Biotechnological Interventions in Bamboo: Advances and Future Prospects. In: Hakeem K, Akhtar MS, Jawaid M, eds. Biotechnologies of Crop Improvement. Springer. 2011;2:217-231.