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Patterns of adventitious root induction during different seasons in some bamboo species

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Abstract: The influence of seasons and auxin /non-auxin growth regulators on adventitious root formation was examined to evolve cloning procedures for *Bambusa multiplex, B. tulda, B. vulgaris* and *Dendrocalamus membranaceus*. During three growing seasons (winter, summer and rainy), single-node culm cuttings were prepared from mature culms and treated for 24 h with 2 mM indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), naphthalene acetic acid (NAA), boric acid and water (control) separately. The treated cuttings were horizontally placed, covered completely with sand and maintained for 2 months in low-cost mist chamber. The best rooting occurred in summer season, which was enhanced by 56.5 per cent and 19.4 per cent over that in winter and rainy seasons, respectively. In general, the potential of different bamboo species for adventitious rhizogenesis was found to be in the order: *B. vulgaris* > *B. multiplex* > *D. membranaceus* > *B. tulda*. The treatment with boric acid, NAA and IBA resulted in significantly increased overall adventitious rooting than water treated control but at the individual species level, growth regulators (IBA and NAA) significantly enhanced adventitious rhizogenesis only in *B. tulda.*

Key words: Adventitious rhizogenesis, *Bambusa tulda, Bambusa multiplex,* boric acid, growth regulators.

INTRODUCTION

Vegetative propagation of bamboos involves various procedures, most of which aim at transforming the innumerable buds present at every node into planting material (Banik, 1980). Among these procedures, propagation through adventitious rhizogenesis of culm/branch cuttings is a viable option having the advantage of obtaining enormous number of cuttings from a clump and low costs of transport, handling and labour (Dransfield and Widjaja, 1995).

The method of rooting of cuttings has been adopted for propagation of bamboos since long (Pathak, 1899; Dabral, 1950; McClure, 1966). However, bamboo species exhibit significant variation in the capability of adventitious rhizogenesis, some rooting with

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ease while others posing severe limitations. Further, overriding influence of planting season and species-specific differential response to various growth regulators have also been encountered. Hence, the present study was carried out to investigate adventitious root formation as influenced by season and also to find out the effect of auxin/non-auxin growth regulators on four important bamboo species *viz.*, *Bambusa multiplex* (Laur.) Raeush. ex Schult. & Schult. F., *B. tulda* Roxb., *B. vulgaris* Schrader ex Wendl. and *Dendrocalamus membranaceus* Munro, so as to evolve cloning procedures.

D. membranaceus, *B. multiplex* and *B. tulda* exhibit long flowering cycles of 20, 30 and 25-40 years, respectively. Flowering is uncommon in *B. vulgaris* and when a clump flowers, it produces a large number of spikelets but no seeds (Naithani and Sas Biswas, 1992; Dransfield and Widjaja, 1995). Hence, efficient clonal procedures need to be evolved for rapid and mass multiplication of these important bamboo species.

MATERIALS AND METHODS

During three growing seasons (winter, summer and rainy) of 2002, single-node culm cuttings of all the bamboo species were prepared from mature culms of second year's growth collected from a 5-year-old plantation. The nodal segments were surface disinfected for 5 min with 0.25 per cent (w/v) aqueous mercuric chloride and subsequently washed with sterilized water. Sixty sterilized single-node segments, about 10-15 cm long, were immersed for 24 h in water (control) or in 2 mM aqueous solution of indole-3-acetic acid (IAA), indole-3-butyric acid (IBA), naphthalene acetic acid (NAA) or boric acid. Each treatment consisted of three replicates each of 20 single-node culm cuttings. The treated cuttings were horizontally placed and covered completely with sand (10 cm deep) in beds of a low-cost mist chamber maintained at 70 ± 5 per cent RH and 30 ± 2ºC.

After two months, the cuttings were scored for adventitious rooting percentage and number of roots per cutting. The data obtained were subjected to statistical analysis, employing analysis of variance (ANOVA), '*F*'-test for significance at $P = 0.05$ and computing LSD values to separate means in different groups using statistical software SX (Version 2.0, NH Analytical Software, 1987).

RESULTS

Significant influence of the planting season on adventitious rhizogenesis was recorded. Best rooting occurred in the summer season, which was enhanced by 56.5 per cent and 19.4 per cent over that in winter and rainy seasons, respectively (Fig. 1). Among the growth regulators, treatment with boric acid, NAA and IBA resulted in significantly superior adventitious rooting than water treated control (Fig. 2). Overall, the potential of different bamboo species for adventitious rhizogenesis was found to be in the

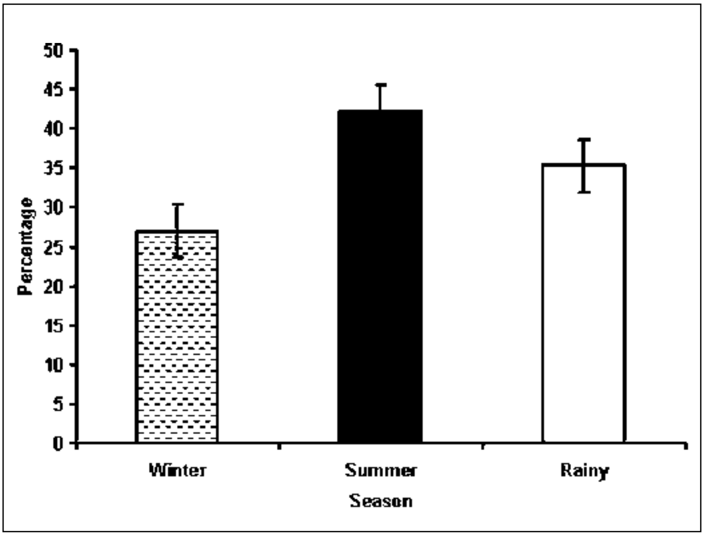

Figure 1. Percentage of adventitious rooting in bamboo species in three planting seasons.

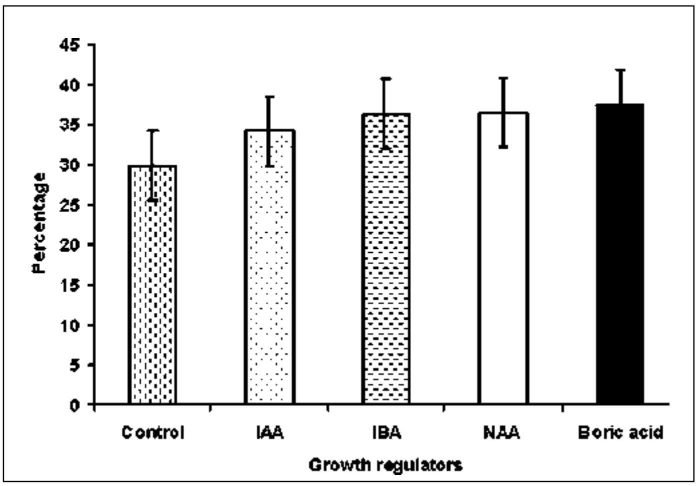

Figure 2. Influence of growth regulators on percentage of adventitious rooting in bamboo species.

order: *B. vulgaris* > *B. multiplex* > *D. membranaceus* > *B. tulda* (Fig. 3).

Interaction of species and seasons was recorded to be significant for adventitious rooting. In general, summer season proved to be better for rooting (Table 1). Interaction between growth regulators and seasons was significant only in the case of *B. tulda.* In *B. multiplex,* cuttings treated with boric acid in rainy season gave the maximum rooting (70.2%). Best treatment-season combinations for *B. vulgaris* and *D. membranaceus* were IAA - summer season and IBA - rainy season, respectively (Table 2).

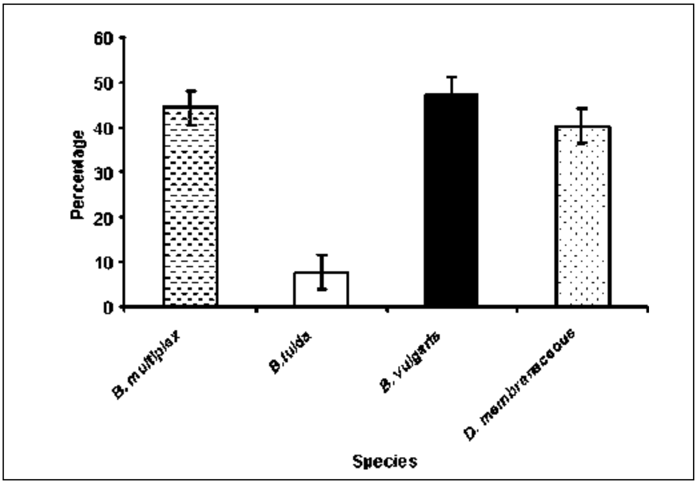

Figure 3. Percentage of adventitious rooting in bamboo species.

Table 1. Percentage of adventitious rooting and root number per cutting in single-node culm cuttings of bamboo species during different seasons (data are mean of three replicates)

Species	Season	Rooting (%)	Root number
B. multiplex	Winter	35.1	3.0
	Summer	56.4	5.0
	Rainy	52.1	4.5
B. tulda	Winter	0	0
	Summer	15.3	2.9
	Rainy	0	0
B. vulgaris	Winter	42.6	3.3
	Summer	62.5	5.6
	Rainy	56.7	4.9
D. membranaceus	Winter	23.6	2.8
	Summer	50.4	4.0
	Rainy	52.8	4.3
$LSD_{0.01}$		15.33	NS

DISCUSSION

Adventitious rhizogenesis of cuttings primarily depends on environment or internal factors, and/or the interaction of the two. Season has a guiding influence on the physiological state of the parent plant and therefore plays an important role in induction and growth of adventitious roots in the detached cuttings. The importance of the physiological status of the bamboo plants for subsequent adventitious rhizogenesis in culm cuttings has been pointed out (Gupta and Pattanath, 1976; Dai, 1981; Agnihotri, 1998). The onset of summer results in emergence of new leaves, side-branches,

Table 2. The interaction of season and growth regulators on adventitious rhizogenesis in single-node culm cuttings of bamboo species (data are mean of three replicates)

Season	Treatment	Rooting (%)			
		B. multiplex	*B. tulda*	*B. vulgaris*	*D. membranaceus*
Summer	Control	51.1	2.6	50.0	38.1
	IAA	55.1	25.7	64.3	48.3
	IBA	58.4	6.7	53.8	56.8
	NAA	58.6	25.7	58.9	57.5
	Boric acid	58.6	8.5	61.4	56.3
Rainy	Control	36.1	0	53.8	43.3
	IAA	51.6	0	50.3	43.3
	IBA	53.4	0	50.1	58.4
	NAA	55.1	0	59.8	56.7
	Boric acid	70.2	0	49.9	53.4
Winter	Control	41.7	0	36.1	13.6
	IAA	21.9	0	39.9	19.4
	IBA	43.3	0	41.2	40.0
	NAA	39.7	0	29.7	28.0
	Boric acid	43.4	0	48.8	19.4
	$LSD_{0.05}$	NS	5.87	NS	NS

resumption of active extension (inter-nodal) growth and upward mobilization of stored photosynthates from the underground rhizome and this coincides with good rooting (Agnihotri and Ansari, 2000; Sanjay Singh *et al.*,2002, 2004). In the present investigation, occurrence of rhizogenesis in the summer season only in *B. tulda* and significantly superior root induction in other species from summer (April) to rainy (July) season conforms to the above views. The acquisition of sensitivity by cells/ tissues towards phytohormones for differentiation and organization into a specific organ depends upon several external and internal factors, including environmental conditions like temperature and humidity (Trewavas, 1991). Seemingly, environmental conditions facilitated the development of sensitivity towards applied growth regulators to trigger the process of adventitious root formation in these bamboo species. Nevertheless, the results revealed the variation in species in their intrinsic capability for adventitious rhizogenesis.

The extent and seasonal variation in the induction of adventitious roots in these bamboo species revealed three patterns - (a) rooting recalcitrant, with extreme season-specificity as in *B. tulda*, (b) rooting amenable, with season-specificity as in *D. membranaceus* and (c) rooting amenable with no season-specificity as in *B. multiplex* and *B. vulgaris.* As a whole, in terms of the ease of rooting of cuttings these may be placed as c > b > a. This differential behaviour pertaining to adventitious rhizogenesis should be considered for optimal production of clonal plantlets through rooted cuttings in these bamboos.

Exogenous application of growth regulators, mostly auxins, has been reported to positively influence induction and growth of adventitious roots in culm cuttings of bamboos (Suzuki and Ordinaro, 1977; Uchimara, 1978; Seethalakshmi *et al.,* 1989; Surendran *et al.,*1989; Agnihotri and Ansari, 2000; Sanjay Singh *et al.,* 2002). In the present study, the treatment with boric acid, NAA and IBA resulted in significantly superior overall adventitious rooting than water treated control. However, at the individual species level, growth regulators significantly enhanced adventitious rhizogenesis only in *B. tulda* where IBA and NAA proved significantly superior (Fig. 2, Table 2). Low levels of auxin often result in failure of adventitious rooting (Cooper, 1935; Smith and Wareing, 1972). Exogenous application of auxin becomes effective if their endogenous level is low, for example, due to inactive growth phase or less accumulation in distal plant parts/nodal segments. This may be a reason for enhancement of rooting by exogenous auxin application in *B. tulda,* which is known to be a very reluctant-to-root species (McClure and Kennard, 1955). Regarding promotive influence of boric acid as evident in the present investigation, boron has been implicated in adventitious rhizogenesis but it induces root growth and development rather than initiation (Hamberg, 1951; Gorter, 1958; Josten and Kutschera, 1999). In cuttings of mung bean, Middleton *et al.* (1978) reported that rooting was initiated by auxin but pre-primordial growth was dependent on the presence of boron. It is believed that boron influences rooting by regulating endogenous auxin levels through enhancement of IAA-oxidase activity and mobilization of oxygen-rich citric and isocitric acids into the rooting tissues (Jarvis *et al.,*1983; Jarvis, 1986). Therefore, further studies on the role of boric acid individually and in combination with various auxins should be carried out to economize propagation procedures.

In summation, bamboo species exhibit differential adventitious rhizogenesis behaviour in culm cuttings *vis-à-vis* seasons, which can be exploited in efficient mass clonal multiplication of these species. Exogenous application of 2 mM boric acid, IBA and NAA can be employed for further enhancement of adventitious root induction.

REFERENCES

Agnihotri, K. 1998. Physiomorphological Changes Associated with Adventitious Root Formation in Culm Branch Cuttings of Some Bamboos of Central India. Ph.D. Thesis, FRI Deemed University, Dehra Dun.

Agnihotri, K. and Ansari, S.A. 2000. Adventitious rhizogenesis in relation to seasonal variation, size of culm branch cuttings and IAA treatment in Bamboos. *Indian Forester* 126: 971-984.

Banik, R.L. 1980. Propagation of bamboo by clonal methods and by seeds. In: G, Lessard and A. Chouinard (Eds.). Bamboo Research in Asia. IDRC, Canada: 139-150.

Cooper, W.C. 1935. Hormones in relation to root formation on stem cuttings. *Plant Physiol*. 10: 789-794

Dabral, S.N. 1950. A preliminary note on propagation of Bamboos from culm segments. *Indian Forester* 76: 313-314.

Dai, O.H. 1981. Raising plants of bushy bamboos from branched culms with notched internodes. *Forest Science and Technology* 1: 3-6.

Dransfield, S. and Widjaja, E.A. 1995. Plant Resources of South-East Asia, Bamboos. Backuys Publishers, Leiden: 39-40.

Gorter, C.J. 1958. Synergism of indole-3-acetic acid in the root production of *Phaseolus* cuttings. *Physiol. Plant* 11: 1-9.

Gupta, B.N. and Pattanath, P.G. 1976. Variation in stored nutrients in culms of *Dendrocalamus strictus* and their effect on rooting of culm cuttings as influenced by their method of planting. *Indian Forester* 102: 235-241.

Hamberg, T. 1951. Rooting experiments with hypocotyls of *Phaseolus vulgaris* L.. *Physiol. Plant* 4: 358-369.

Jarvis, B.C. 1986. Endogenous control of adventitious rooting in non-woody cuttings. In: M.B. Jackson (Ed.), New Root Formation in Plants and Cuttings. Martinus Nijhoff Publishers, Dordrecht : 191-222.

Jarvis, B.C., Ali, A.H.N. and Saheed, A.I. 1983. Auxins and boron in relation to the rooting response and ageing of mung bean cuttings. *New Phytology* 97: 197-204.

Josten, P. and Kutschera ,U. 1999. The micronutrient boron causes the development of adventitious roots in sunflower cuttings. *Ann. Bot.* 84: 337-342.

McClure, F.A. 1966. The Bamboos, A Fresh Perspective. Harvard University Press, Cambridge, Massachusetts, USA: 347p.

McClure, F.A. and Kennard, W.C. 1955. Propagation of bamboo by whole culm cuttings. *Proceedings American Society Horticultural Sciences* 65: 283-288.

Middleton, W., Jarvis B.C. and Booth, A. 1978. The boron requirement for root development in stem cuttings of *Phaseolus aureus* Roxb. *New Phytology* 81: 287-297.

Naithani, H.B. and Sas Biswas 1992. Gregarious flowering of *Dendrocalamus membranaceus. Indian Forester* 118: 300-301.

Pathak,S.L. 1899. The propagation of common male bamboo by cuttings in the Pinjaur-Pateala forest nurseries. *Indian Forester* 25: 307-308.

Sanjay Singh, Ansari, S.A. and Kumar, P. 2002. Clonal propagation of *Bambusa nutans* through culm and culm-branch cuttings. *Indian Forester* 128: 35-40.

Sanjay Singh, Kumar, P. and Ansari, S.A. 2004. A simple method for large-scale propagation of *Dendrocalamus asper. Scientia Horticulturae* 100: 251-255.

Seethalakshmi, K.K., Venkatesh, C.S. and Surendran, T. 1989. Vegetative propagation of bamboos using growth promoting substances-1. *Bambusa balcooa* Roxb. *Indian Journal of Forestry* 6: 98-103.

Smith, D.R. and Wareing, P.F. 1972. The rooting of actively growing and dormant leafy cuttings in relation of the endogenous hormone levels and photoperiod. *New Phytology* 71: 483-500.

Surendran, T., Seethalakshmi, K.K. and Somen, C.K. 1989. Vegetative propagation of *Bambusa arundinacea* and *Dendrocalamus strictus* by culm cuttings. *Malaysian Forester* 49: 432-456.

Suzuki, T. and Ordinaro, F.F. 1977. Some Aspects and Problems of Bamboo Forestry and Utilization in the Philippines. Asia Forest Industries, College, Laguna, Philippines.

Trewavas, A.J. 1991. How do plant growth substances work? II. *Plant Cell Environment* 14: 1-12.

Uchimara, E. 1978. Ecological studies on the cultivation of bamboo forest in Philippines. *Bulletin of Forestry and Forest Products Research Institute* 301: 79-118.