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Effect of Rooting Hormone on Macro-Proliferation of Different Bamboo Species

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ABSTRACT: Bamboo is an ecologically and economically important resource with significant potential to support livelihoods, enhance carbon sequestration, and promote sustainable land management. However, large-scale cultivation is constrained by irregular flowering, limited seed production because most of the species do not produce seed every year, and short seed viability, which limit natural regeneration. Therefore, the development of efficient vegetative propagation techniques is essential to ensure a continuous supply of quality planting material. Therefore, the present study was conducted from February to June 2024 at the Bamboo Resource Centre, College of Forestry, Navsari Agricultural University, Navsari, Gujarat, to evaluate the effect of different auxin concentrations on macro-proliferation of different bamboo species. The experiment was laid out in a factorial completely randomized design using one-year-old seedlings of *Bambusa nutans*, *Bambusa tulda* and *Bambusa polymorpha*, subjected to seven auxin treatments including control, IBA (indole-3-butyric acid) (100 and 200 ppm), NAA (naphthalene acetic acid) (100 and 200 ppm) and their combinations. Observations on sprouting, growth, rooting, survival and multiplication rate were recorded at 120 days after treatment. Results revealed that *Bambusa nutans* exhibited superior growth performance and multiplication rate, whereas *Bambusa polymorpha* performed better in root development and survival. The results revealed that across treatments, G_1 (control) consistently recorded significantly higher values for all parameters. Among the auxin-treated propagules, G_2 (IBA 100 ppm) showed comparatively better performance and remained closer to the control. However, higher concentrations of auxins and their combinations, particularly G_4 (IBA 100 ppm + NAA 200 ppm), G_5 (IBA 200 ppm + NAA 100 ppm), G_3 (IBA 200 ppm), and G_7 (NAA 200 ppm), resulted in a significant reduction in all parameters. Overall, almost all parameters responded positively to the control, while the application of auxins adversely affected the performance of macro-proliferated propagules. The findings demonstrate that exogenous auxin application is not required for macro-proliferation of bamboo under nursery conditions, providing a cost-effective and ecologically sustainable approach for large-scale bamboo propagation and ecological restoration.

Keywords: *Bambusa* spp., Growth and root parameters, Macro-proliferation, Rooting hormones (Auxins).

1. INTRODUCTION

Bamboo, a perennial woody grass belonging to the subfamily *Bambusoideae* under the family *Poaceae,* is one of the most versatile and economically important plant groups in tropical and subtropical regions. Globally, bamboo comprises about 1662 species distributed across 121 genera (Canavan et al., 2017). Owing to its wide range of uses and importance in the rural economy, it is often referred to as "Green Gold", "Poor Man's Timber" and "Cradle to Coffin

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Plant". Bamboo serves as a valuable source of raw material for construction, handicrafts, furniture, paper, and bioenergy, while also playing an important ecological role in soil conservation and carbon sequestration.

The natural distribution of bamboo extends across Asia, Africa and the Americas while Europe has no native bamboo species. Bamboo occupies 1.50 lakh sq. km, representing 17.71% of India's total forest area (ISFR, 2021). India is home to 148 bamboo species, including indigenous and exotic species of bamboo from 29 genera. Bamboos are found in almost all the states of the country, from the tropical to the temperate regions, and the alluvial plains to the high mountains; the only exception where they do not occur naturally is Kashmir. Its occurrence is largely influenced by climatic and edaphic factors such as rainfall, temperature, soil type and altitude. Bamboo is distributed from 47° S to 50° 30′ N latitude and from sea level to about 4300 m altitude (Ahmad et al., 2021). The deciduous and semi-evergreen areas of northeastern India, as well as the tropical moist deciduous forests of northern and southern India, have the highest number of species. Nearly 90 bamboo species are found in India, out of which 41 are indigenous to the hilly areas of northeastern India (Sharma & Nirmala, 2015).

Bamboo is a multifunctional, fast-growing plant on Earth. Bamboo has played a crucial role in the daily lives of millions of people in tropical countries, providing environmental, social, and economic benefits (Ahmad et al., 2021). Bamboo is an essential part of existence, not only in Indian culture but also across Southeast Asian cultures. This group of species is most closely connected to the cornerstone of rural life and culture in our nation due to the variety of functions they serve. Due to its lignified culms, which can serve as a substitute for lumber in many ways, as well as its rapid growth, complex rhizome system, and sustainability, it has evolved into a plant with conservation value that can help to lessen the effects of climate change. Not just pandas, but many other creatures depend on bamboo as a food resource. Like rice, maize, wheat, and sugar cane, bamboo is a vital grass closely tied to human livelihoods. It satisfies needs for shelter, food, paper, and more; bamboo is known as "the plant of a thousand uses" for a reason.

In recent years, the increasing demand for bamboo planting material for plantation forestry, agroforestry, and commercial cultivation has underscored the need for efficient mass-multiplication techniques. Bamboo can be propagated either by seed or by vegetative methods. However, seed propagation is unreliable because most bamboo species flower gregariously only once in their lifetime after long intervals of 12 to 120 years and produce seeds with short viability. As a result, an assured seed supply is rarely available for nursery production. In addition, the monocarpic nature of bamboo and the difficulty of improvement through hybridization make large-scale seed propagation highly uncertain.

The habit of unpredictable gregarious flowering and seeding, followed by death, makes seed propagation of bamboo uncertain, as seeds have short viability. Improving through hybridization is very difficult. To achieve these objectives, large-scale plantation and farming, along with a vegetative propagation technique using PGRs, will offer great potential for the production of high-quality planting material (Ilorkar et al., 2021).

Vegetative propagation of bamboo can be achieved through the branch-cutting method, single-node (bud) cutting method, culm-cutting (flute) method, layering method, and rhizome (offshoot) planting method, all of which are commonly employed (Hamilton et al., 2022). However, these methods often require a large number of explants and usually produce a limited number of propagules. Further, when propagules are obtained from vegetative parts of unknown age, there is a possibility that such planting stock may flower earlier in the field, which can adversely affect plantation performance. Therefore, the production of planting material of known physiological age is highly desirable.

Further, the age of bamboo plants is not known in vegetative saplings until the explants are taken from the mother plant of known age. Using bamboo planting material of unknown age is a big risk, as such a plantation may flower and dry up in the field if it reaches its physiological age. Therefore, nowadays farmers are demanding known-age plants, which is only possible through seedling propagation. However, the bamboos are monocarpic in nature and produce seeds only once in their lifetime; therefore, there is no assured seed supply for the nursery.

Macro-proliferation of bamboo seedlings offers an effective means of producing large numbers of plants from seedling-origin stock of known physiological age. Bamboo seedlings have the capacity to proliferate, and this potential of bamboo can be used for mass propagation through the macro-proliferation method. In this technique, seedlings raised for about six months to one year produce multiple shoots from rhizomes, which are subsequently separated and replanted as independent propagules (Kumar, 1991). The process can be repeated over successive cycles to obtain a large number of planting stocks suitable for field planting.

Since rooting and survival of separated propagules are important for the success of this method, the use of plant growth regulators has often been considered beneficial. Previous studies have reported the positive role of auxins in vegetative propagation of bamboo under nursery conditions (Ilorkar et al., 2021; Venkatesh & Kariappa, 2021). Despite the reported usefulness of plant growth regulators in vegetative propagation, their role in macro-proliferation of bamboo seedlings remains inconsistent and species-specific. Therefore, the present study aims to evaluate whether auxin application is essential for improving macro-proliferation efficiency in bamboo species.

However, the response to exogenous auxins may differ among bamboo species depending upon their genetic constitution and endogenous hormonal balance. Therefore, it is necessary to evaluate the role of rooting hormones in macro-proliferation of important bamboo species under nursery conditions. In this context, the present study was undertaken to assess the effects of different auxin concentrations on the growth, rooting, survival, and multiplication of Bambusa nutans, Bambusa tulda, and Bambusa polymorpha, with a view to developing a suitable protocol for large-scale propagation.

2. MATERIALS AND METHODS

2.1. Experiment Site

The present study was carried out from February to June 2024 at the Bamboo Resource Centre, College of Forestry, Navsari Agricultural University (NAU), Navsari, Gujarat, India. Navsari is situated in the coastal region of South Gujarat at 20°57′ N latitude and 72°54′ E longitude, at an altitude of 9 m above mean sea level. The region falls under the South Gujarat Heavy Rainfall Zone (AES-III). It is characterised by a tropical, humid climate with hot summers, moderately cool winters, and a warm, humid monsoon. The experiment was carried out under nursery conditions.

2.2. Experimental Methods, Treatments and Procedures

2.2.1. Experimental design and treatments

The experiment was conducted under nursery conditions using a factorial completely randomized design (FCRD) in which one-year-old seedlings of three bamboo species, viz., *Bambusa nutans* (B_1), *Bambusa tulda* (B_2) and *Bambusa polymorpha* (B_3) were transplanted into polybags of size 6 × 8 inches and treated with seven different levels of auxin concentrations, namely G_1: control, G_2: IBA 100 ppm, G_3: IBA 200 ppm, G_4: IBA 100 ppm + NAA 200 ppm, G_5: IBA 200 ppm + NAA 100 ppm, G_6: NAA 100 ppm and G_7: NAA 200 ppm. The combination of three bamboo species and seven auxin treatments resulted in twenty-one treatment combinations. Each treatment combination was replicated three times, with fifteen propagules maintained in each replication. Thus, each treatment combination consisted of a total of forty-five propagules (fifteen propagules × three replications). Overall, 945 propagules (21 treatment combinations × 45 propagules) were used in the experiment. The treatment combinations were designated as B_1G_1 to B_3G_7, where B_1, B_2, and B_3 represented the three bamboo species, and G_1 to G_7 represented the seven auxin treatments.

2.2.2. Plant material and preparation of growing media

One-year-old, well-managed seedlings of *Bambusa nutans*, *Bambusa tulda*, and *Bambusa polymorpha*, with multiple culms (three or more) and well-developed roots and rhizomes, were selected for macro-proliferation. Prior to initiation, nursery beds were cleaned, and weeds were removed. The growing medium was prepared using clay soil (54.3% clay) and sand in a 2:1 (v/v) ratio and filled into polybags of size 6 × 8 inches. The polybags were arranged in nursery beds and watered regularly to maintain adequate moisture.

2.2.3. Preparation and application of auxins (PGR)

Auxin solutions of IBA and NAA at concentrations of 100 ppm and 200 ppm were prepared by dissolving 100 mg and 200 mg of IBA and NAA, respectively, in a small quantity of NaOH solution, then diluting with distilled water to a final volume of 1000 ml. Separate stock solutions of IBA and NAA were prepared and mixed in appropriate proportions to obtain the desired treatment combinations. Seven treatment levels were used in the study: G_1 (Control, without auxin treatment), G_2 (IBA 100 ppm), G_3 (IBA 200 ppm), G_4 (IBA 100 ppm + NAA 200 ppm), G_5 (IBA 200 ppm + NAA 100 ppm), G_6 (NAA 100 ppm), and G_7 (NAA 200 ppm). The propagules of each bamboo species were treated with the corresponding auxin solutions according to the assigned treatment combinations, then planted in polybags under nursery conditions.

2.2.4. Macro-proliferation procedure

Macro-proliferation of bamboo seedlings was carried out following the standard procedure described by Kumar (1991), with slight modifications, including the application of rooting hormones to enhance the growth, rooting, and survival of propagules. Seedlings were carefully removed from polybags, and soil adhering to roots and rhizomes was washed off. Each plant was dissected at the rhizome neck using sharp secateurs to obtain propagules containing the rhizome, roots, and shoot. Approximately 75 per cent of the shoot portion was trimmed to reduce transpiration loss. To minimise mortality, division was preferably performed in the evening or under relatively cool conditions.

The separated propagules were dipped in a fungicide solution of carbendazim at the rate of 1 g L^{-1} for 15–20 minutes to prevent fungal infection. Subsequently, 50 per cent of the length of the basal portion of each propagule was treated with the respective auxin solution for 15 minutes.

2.2.5. Transplanting and maintenance

After treatment, propagules were transplanted into polybags containing prepared growing medium. The planted propagules were maintained under partial shade (50%) in a low-cost polytunnel covered with a 50% green shade net. The shade net reduced incident sunlight by approximately 50% of the incident sunlight, thereby maintaining favourable humidity and temperature conditions for sprouting and establishment of the propagules. Irrigation was applied as required during the initial stage and subsequently at an interval of two days. Routine nursery operations, such as weeding and cleaning, were carried out regularly. After one month, plants were gradually exposed to full sunlight.

2.3. Observations Recorded

Observations were recorded at 120 days after initiation of the experiment on days to first sprouting, number of sprouts, basal diameter, number of leaves per plant, shoot height, root length, number of rootlets, number of rhizomes, survival percentage and multiplication rate.

2.4. Statistical Analysis

The data were analysed using analysis of variance appropriate for a factorial completely randomised design. The standard error of the mean (SEm±) and critical difference (CD) at a 5% level of significance were calculated to compare treatment means following the method described by Panse & Sukhatme (1985).

3. RESULTS AND DISCUSSION

The results are presented under different categories to facilitate a clear understanding of the effects of auxin treatments and their interactions on macro-proliferation performance of three bamboo species. The results of the present investigation of auxin concentrations and their interactions revealed significant variation among bamboo species, providing clear insights into the physiological and propagation responses under macro-proliferation.

3.1. Growth Parameters

Days to the first sprout varied significantly among species, treatments, and their interactions. Among the species, earlier initiation of sprouting was recorded in B_1: *Bambusa nutans* (9.21 days), whereas sprouting was delayed in B_3: *Bambusa polymorpha* (22.73 days). Among auxin treatments, the earliest sprouting was recorded in G_1: control (10.37 days), followed by G_2: IBA 100 ppm, whereas higher concentrations, particularly G_7: NAA 200 ppm, resulted in delayed sprouting (20.41 days). This indicates that exogenous auxin application beyond optimum levels suppressed bud activation. The interaction effect was also significant, and the earliest sprouting was recorded in B_1G_1 (4.29 days; Table 1), indicating a better response of B_1 (*Bambusa nutans*) under untreated conditions, while higher auxin concentrations delayed sprout initiation.

The number of sprouts per propagule differed significantly among species. B_1: *Bambusa nutans* recorded the highest number of sprouts (1.79), whereas B_2: *Bambusa tulda* recorded the lowest (0.85). Among treatments, G_1: control (2.08) showed a significantly higher number of sprouts, while the minimum was recorded in G_7: NAA 200 ppm (0.76). The interaction effect indicated that B_1G_1 recorded the highest number of sprouts (2.56), further indicating that untreated propagules of B_1: *Bambusa nutans* performed best during macro-proliferation (Table 1).

The reduction in sprout production at higher auxin concentrations and in auxin combinations may be attributed to the inhibitory effects of supra-optimal hormone levels on bud differentiation and shoot initiation. Similar findings were also reported by Singh et al.(2002) in bamboo propagation studies.

Basal diameter also varied significantly among species. B_2: *Bambusa tulda* recorded the maximum basal diameter (2.86 mm), whereas B_3: *Bambusa polymorpha* recorded the minimum (1.89 mm). Among treatments, G_1: control recorded the highest basal diameter (3.12 mm), whereas the lowest was observed under G_7: NAA 200 ppm (1.66 mm). The interaction effect showed maximum basal diameter in B_2G_1 (4.09 mm), indicating better growth performance of B_2: *Bambusa tulda* under untreated conditions (Table 1).

The number of leaves per propagule was significantly higher in B_1: *Bambusa nutans* (9.67), whereas B_2: *Bambusa*

Table 1. Effect of different concentration of auxins on macro-proliferation of different *Bambusa* species for growth parameters

Treatment	Days taken for the first				Number of sprouts				Basal diameter (cm)				Number of leaves per rooted				Shoot height (cm)			
	B1	**B2**	**B3**	**Mean**	**B1**	**B2**	**B3**	**Mean**	**B1**	**B2**	**B3**	**Mean**	**B1**	**B2**	**B3**	**Mean**	**B1**	**B2**	**B3**	**Mean**
G1 - Control	4.29	9.71	17.11	10.37	2.56	1.67	2.02	2.08	2.82	4.09	2.44	3.12	14.82	6.80	10.96	10.86	48.62	46.11	35.93	43.55
G2 – IBA 100 ppm	6.20	10.80	19.26	12.09	2.22	0.91	1.93	1.69	2.42	3.99	1.87	2.76	13.22	4.11	10.87	9.40	37.55	41.17	34.72	37.81
G3 – IBA – 200 ppm	12.00	18.40	25.98	18.79	1.13	0.56	1.20	0.96	1.70	1.83	1.70	1.75	6.18	2.93	6.44	5.18	15.95	17.83	27.97	20.58
G4 – IBA 100 ppm + NAA 200 ppm	8.27	14.65	20.55	14.49	2.11	0.80	1.44	1.45	2.31	2.85	1.86	2.34	10.44	3.18	8.71	7.44	32.52	33.67	33.19	33.13
G5 - IBA 200 ppm + NAA 100 ppm	9.89	15.98	23.69	16.52	2.04	0.78	1.44	1.42	1.64	2.77	1.83	2.08	9.64	3.16	7.22	6.67	25.63	33.23	32.81	30.56
G6 – NAA 100 ppm	11.78	17.80	24.14	17.91	1.69	0.69	1.40	1.26	1.89	2.76	1.82	2.15	9.16	3.00	7.00	6.39	24.55	25.28	28.09	25.97
G7 – NAA 200 ppm	12.02	20.80	28.40	20.41	0.76	0.51	1.02	0.76	1.54	1.75	1.68	1.66	4.20	2.18	5.09	3.82	15.54	17.31	26.64	19.83
Mean	9.21	15.45	22.73		1.79	0.85	1.50		2.05	2.86	1.89		9.67	3.62	8.04		28.62	30.66	31.34	
	B	G	Interaction B × G		B	G	Interaction B × G		B	G	Interaction B × G		B	G	Interaction B × G		B	G	Interaction B × G	
S. Em. ±	0.23	0.34	0.60		0.03	0.05	0.09		0.03	0.04	0.07		0.17	0.25	0.44		0.39	0.59	1.03	
C.D. at 5 %	0.64	0.98	1.70		0.09	0.14	0.25		0.08	0.12	0.21		0.47	0.72	1.25		1.11	1.69	2.93	
C. V. %		6.53				11.05				5.56				10.71				5.89		

B1 - Bambusa nutans, B2 - Bambusa tulda and B3 – Bambusa polymorpha *tulda* recorded the lowest (3.62). Higher leaf production in Bambusa nutans is directly associated with its higher sprouting ability and faster establishment. Among treatments, G_1: control (10.86) recorded the highest number of leaves, while the lowest was observed in G_7: NAA 200 ppm (3.82). The interaction effect indicated that B_1G_1 recorded the maximum number of leaves (14.82), suggesting that untreated propagules of B_1: *Bambusa nutans* exhibited superior vegetative growth under nursery conditions (Table 1).

Shoot height varied significantly among species and treatments. The maximum shoot height was recorded in B_3: *Bambusa polymorpha* (31.34 cm), whereas the minimum was recorded in B_1: *Bambusa nutans* (28.62 cm). Among treatments, G_1: control recorded the maximum shoot height (43.55 cm), whereas the minimum was observed under G_7: NAA 200 ppm (19.83 cm). The interaction effect showed maximum shoot height in B_1G_1 (48.62 cm), indicating that untreated propagules exhibited better establishment and subsequent shoot growth compared with auxin-treated propagules (Table 1).

These results are in close proximity to the earlier reports by Saharia and Sen (1990) and Agnihotri and Ansari (2000), who observed that the exogenous application of various growth regulators, mostly auxins, had been reported to positively influence growth induction in culm cuttings of bamboos. However, in the present study, untreated propagules performed better indicating that endogenous hormonal balance in seedling-derived propagules is sufficient for sprouting and growth. Similar observations were also reported by Singh et al.(2002) in bamboo cuttings.

The superior performance observed under control conditions may be attributed to adequate endogenous auxin levels present in seedling-derived propagules, which regulate cell division and elongation without external supplementation. The application of higher concentrations of exogenous auxins might have disrupted the internal hormonal balance, leading to inhibitory effects on sprouting and growth. Similar inhibitory effects of excessive auxin concentrations have been reported in vegetative propagation of woody species, where supra-optimal hormone levels adversely affect physiological processes.

3.2. Root Parameters

Root characteristics differed significantly among species and treatments. B_3: *Bambusa polymorpha* recorded the highest root length (18.51 cm), whereas the lowest was observed in B_1: *Bambusa nutans* (16.21 cm). Among treatments, G_1: control recorded significantly higher root length (22.52 cm), while the lowest was recorded under G_7: NAA 200 ppm (12.73 cm). The interaction effect showed maximum root length in B_2G_1 (22.99 cm) (Table 2).

The number of rootlets was also significantly influenced by species and treatments. B_3: *Bambusa polymorpha* recorded the highest number of rootlets (21.42), whereas the lowest was observed in B_1: *Bambusa nutans* (11.90). Among treatments, G_1: control recorded the maximum number of rootlets (19.22), whereas the minimum was recorded under G_7: NAA 200 ppm (12.27). The interaction effect showed the maximum number of rootlets in B_3G_1 (25.00) and the minimum in B_1G_7 (7.00) (Table 2).

The number of rhizomes was significantly higher in B_3: *Bambusa polymorpha* (2.34), whereas the lowest was recorded in B_2: *Bambusa tulda* (1.44). Among treatments, G_1: control recorded the maximum number of rhizomes (2.69), while the minimum was observed under G_7: NAA 200 ppm (1.22). The interaction effect indicated the maximum number of rhizomes in B_1G_1 (3.27) and the minimum in B_2G_7 (0.80) (Table 2). These results are in close conformity with earlier findings of Seethalakshmi et al. (1983) and Nain et al. (2007), who reported variation in rooting behaviour among bamboo species. Similar observations were also reported by Razvi et al. (2015) in Dendrocalamus giganteus and by Rana et al. (1987) and Nautiyal et al. (1991) in other tree species.

The variation in rooting response among species and treatments may be linked to differences in endogenous hormone levels, carbohydrate reserves and genetic constitution. The superior rooting under control conditions suggests that seedling-origin propagules possess sufficient physiological maturity and hormonal balance to initiate root formation. Excessive auxin concentrations may have led to callus formation or inhibited root elongation, thereby reducing overall rooting efficiency.

3.3. Survival Percentage

Species and auxin treatments significantly influenced survival percentage. Among species, B_3: *Bambusa polymorpha* recorded the highest survival (45.71%), whereas B_1: *Bambusa nutans* recorded the lowest (31.43%). Among treatments, G_1: control recorded the maximum survival (59.26%), while the lowest was observed under G_7: NAA 200 ppm (23.70%). The interaction effect indicated that B_2G_1 had the highest survival (73.33%), whereas the

Table 2. Effect of different concentrations of auxins on macroproliferation of different *Bambusa* species for root parameters, survival percentage and multiplication rate

Treatment	Days taken for the first				Number of sprouts				Basal diameter (cm)				Number of leaves per rooted				Shoot height (cm)			
	B1	B2	B3	Mean	B1	B2	B3	Mean	B1	B2	B3	Mean	B1	B2	B3	Mean	B1	B2	B3	Mean
G1 - Control	22.89	22.99	21.69	22.52	14.47	18.20	25.00	19.22	3.27	2.00	2.80	2.69	48.89	73.33	55.56	59.26	2.34	1.72	2.22	2.09
G2 – IBA 100 ppm	21.60	22.47	19.95	21.34	11.00	16.80	22.60	16.80	2.80	1.70	2.47	2.32	42.22	55.55	53.33	50.37	1.89	1.68	1.96	1.84
G3 – IBA – 200 ppm	13.00	14.07	17.12	14.73	12.13	12.67	19.40	14.73	1.20	1.13	2.20	1.51	15.56	24.44	40.00	26.67	1.39	1.36	1.36	1.37
G4 – IBA 100 ppm + NAA 200 ppm	19.38	18.86	19.21	19.15	13.53	15.07	22.27	16.96	2.80	1.60	2.40	2.27	40.00	51.11	44.44	45.19	1.88	1.60	1.60	1.69
G5 - IBA 200 ppm + NAA 100 ppm	13.63	18.86	18.80	17.10	12.93	13.40	21.73	16.02	2.20	1.53	2.33	2.02	31.11	42.22	44.44	39.26	1.68	1.57	1.43	1.56
G6 – NAA 100 ppm	13.14	16.53	18.34	16.00	12.27	13.20	19.60	15.02	1.53	1.33	2.27	1.71	26.66	39.90	42.22	36.26	1.63	1.39	1.43	1.48
G7 – NAA 200 ppm	9.86	13.86	14.47	12.73	7.00	10.47	19.33	12.27	0.93	0.80	1.93	1.22	15.56	15.56	40.00	23.70	1.31	1.30	1.22	1.28
Mean	16.21	18.23	18.51		11.90	14.26	21.42		2.10	1.44	2.34		31.43	43.16	45.71		1.73	1.52	1.60	
	B	G	Interaction B × G		B	G	Interaction B × G		B	G	Interaction B × G		B	G	Interaction B × G		B	G	Interaction B × G	
S. Em. ±	0.20	0.31	0.54		0.24	0.37	0.64		0.04	0.07	0.12		1.04	1.59	2.75		0.02	0.04	0.06	
C.D. at 5 %	0.58	0.89	1.54		0.69	1.05	1.81		0.13	0.19	0.33		2.96	4.52	7.84		0.07	0.10	0.18	
C. V. %		5.30				6.94				10.31				11.86				6.63		

B1 - Bambusa nutans, B2 - Bambusa tulda and B3 – Bambusa polymorpha lowest survival (15.56%) was observed in B_1G_3, B_1G_7, and B_2G_7. The higher survival under G_1: control may be attributed to better root development and absence of hormonal stress (Table 2).

Reduced survival at higher auxin concentrations may be due to inhibitory effects of excessive hormone application. Similar observations were reported by Tomar et al. (2011), who found that untreated propagules showed better survival under nursery conditions.

3.4. Multiplication Rate

The multiplication rate differed significantly among species and treatments. B_1: *Bambusa nutans* recorded the highest multiplication rate (1.73), whereas B_2: *Bambusa tulda* recorded the lowest (1.52). Among treatments, G_1: control recorded the highest multiplication rate (2.09), while the lowest was observed under G_7: NAA 200 ppm (1.28). The interaction effect showed the maximum multiplication rate in B_1G_1 (2.34) and the minimum in B_3G_7 (1.22) (Table 2).

The higher multiplication rate in B_1: *Bambusa nutans* may be attributed to better sprouting ability and rhizome development. The superior performance under G_1: control indicates that macro-proliferation efficiency is largely governed by the inherent regenerative capacity of the species rather than exogenous hormone application. These findings are consistent with Banik (1995), who reported that bamboo seedlings can be effectively multiplied through rhizome division without the need of growth regulators.

Unlike several earlier reports that advocate the use of auxins for vegetative propagation, the present study clearly demonstrates that exogenous auxin application is not only unnecessary but may even be detrimental to macro-proliferation of seedling-derived bamboo propagules under nursery conditions.

4. CONCLUSION

The present investigation demonstrated that macro-proliferation is an efficient technique for large-scale multiplication of bamboo using seedling-derived planting material of known physiological age. Significant variation was observed among the three bamboo species evaluated. *Bambusa nutans* exhibited superior sprouting, vegetative growth and multiplication rate. Whereas *Bambusa polymorpha* showed better rooting performance and survival percentage. *Bambusa tulda* displayed intermediate performance for most of the parameters. Among the auxin treatments, the control (without auxin application) consistently recorded the highest values for sprouting, growth, rooting, survival and multiplication rate. Although IBA at 100 ppm performed relatively better than the other auxin treatments, none of the auxin-treated propagules surpassed the control. In contrast, higher concentrations of IBA and NAA, particularly NAA at 200 ppm, adversely affected most growth and rooting parameters, resulting in reduced survival and multiplication rates. The superior performance of untreated propagules may be attributed to the adequate regenerative potential and physiological vigour of seedling-derived rhizome segments. The findings indicate that exogenous auxin application is not necessary for successful macro-proliferation of *Bambusa nutans*, *Bambusa tulda* and *Bambusa polymorpha* under nursery conditions. Therefore, macro-proliferation without auxin treatment can be recommended as a simple, cost-effective, and environmentally sustainable approach for the large-scale production of high-quality bamboo planting material for plantation development, agroforestry systems, and ecological restoration programmes.

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CRediT authorship contribution statement

Agravat Shivaniben: Field experimentation, Data collection, Writing – original draft. **Jayesh Pathak:** Conceptualisation, Experiment supervision, writing, reviewing and editing. **S. A. Huse:** Reviewing and editing. **M. B. Tandel:** Reviewing and editing. **Jayendra Chavda:** Field assistance and data collection.

Conflict of interest

The authors declare that they have no known competing financial or personal relationships that could have influenced the work reported in this paper

Declaration of generative AI and AI-assisted technologies in the writing process

No AI tools have been used in the writing process. Basic checks of grammar, spelling and punctuation need no declaration.

Data availability statement

Data generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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