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Biochemical and physiological effects of propagule type and auxin concentration on adventitious root formation in novel Jasmine genotypes

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Abstract

Jasmine is primarily propagated through asexual methods, and bio-stimulants can improve soil fertility around plant roots. Some jasmine genotypes are mainly propagated through cuttings; however, the rooting process is slow and inconsistent. Specific jasmine genotypes are primarily propagated through cuttings, but the rooting process is slow and uneven. This study evaluated the regeneration potential of stem cuttings by investigating the effects of auxins and various propagule types on adventitious root formation, along with associated biochemical changes. A field experiment conducted by the Department of Floriculture and Landscaping at TNAU, Coimbatore, Tamil Nadu, from December 2023 to June 2024, during the winter monsoon, investigated the rooting of stem cuttings in three *Jasminum* genotypes using auxin. Stem cuttings of *J. sambac* Double Flower type (DF), *J. grandiflorum* White Flower type (WF) and a new cultivar of *J. multiflorum* CO.1 Winter Jasmine (CO.1 WJ) were treated with three Indole-3-Butyric Acid (IBA) concentrations (0.5, 1, 1.5 g L^{-1}) and distilled water as control. Utilizing a Factorial Completely Randomized Design for terminal and semi-hardwood cuttings, the study found that rooting hormone significantly enhanced root formation and stem and shoot growth. Semi-hardwood cuttings of *J. sambac* (DF) treated with 1g L^{-1} IBA had the highest rooting rate (88.60%), number of roots (9.34), root length (12.74 cm), shoot length (10.26 cm) and number of leaves (6.60). The highest rooting rate of 48.15% was obtained from the terminal cuttings of *J. grandiflorum* (WF) that had been treated with 1g L^{-1} IBA, in addition to other parameters such as number of roots (13.52), root length (12.03 cm), shoot length (14.30 cm), and number of leaves (13.48). On the other hand, *J. multiflorum* (CO.1 WJ) recorded the highest rooting rate of 72.23%, root number of 16.52, root length of 17.22 cm, shoot length of 14.14 cm and number of leaves, which was pegged at 31.70 when terminal cuttings were treated with 1g L^{-1} IBA. The current findings indicate that the application of auxins is crucial for promoting early root initiation and achieving higher rooting success, making it advantageous for vegetative propagation.

Keywords

jasmine; vegetative propagation; growth regulator; IBA; rooting

Introduction

Jasmine is one of the oldest flowers grown in India. Its worldwide popularity

stems from its inherent beauty and fragrance. It has medicinal uses and is used to manufacture aromatic soaps, perfumes and cosmetics (1). There are only three commonly grown *Jasminum* species among the many existing ones, namely *J. sambac, J. grandiflorum and J. auriculatum* (2). The following are some more or less known *Jasminum* species with suitable economic features according to the initial study done at Tamil Nadu Agricultural University that are *J. calophyllum*, *J. nitidum*, *J. rigidum*, *J. flexile* and *J. multiflorum*. These plants have beautiful blooms that are ideal for marketing loose-cut flowers and garden design. In addition, they can blossom nearly all year round (3)

Rooting hormones increase success rates by speeding up root development (4). The studies on the external application of several plant growth regulators (PGRs) and the analysis of intrinsic phytohormones have indicated their significant role in flower development (5-7).

Auxin, either endogenous or exogenous, enhances the rooting of cuttings by affecting differentiated root cells or cell arrangements that are recognizable parts of an individual system, according to whether they come from stems or roots.

Chrysanthemum cuttings treated with a concentration of 0.4 g L^{-1} exhibited superior rooting performance compared to other treatments. This optimal concentration resulted in a higher percentage of successful rooting, demonstrating its effectiveness in promoting root development in chrysanthemum propagation (8). Various levels of IBA (Indole-3-butyric acid) were tested on a selective carnation variety. The results indicated that biostimulants helped speed regeneration, shoot elongation, and root formation. These substances proved to be helpful in the overall propagation and establishment of carnation plants as well (9).

Furthermore, rapid initiation and easy formation of roots through plant growth regulators lead to speedy regeneration of plants, including their shoots. Earlier studies on the propagation of Jasmine have indicated varying responses of species and genotypes (10-12). This study aimed to develop effective propagation protocols for three common *Jasminum* genotypes, which have been identified as relatively difficult to root.

Materials and Methods

Experimental design

The study was carried out from December 2023 - June 2024 in the Department of Floriculture and Landscaping, TNAU, Coimbatore, to identify the ideal vegetative propagule and optimize growth regulator requirements feasible for three novel *Jasminum* genotypes. The genotypes included a double flower type of *J. sambac* (DF), a white flower bud type of *J. grandiflorum* (WF), and a new cultivar CO.1 Winter Jasmine of *J. multiflorum* (CO.1 WJ) evolved at TNAU. The double flower type genotype of *J. sambac* (DF) is high-yielding with bolder buds. The white flower bud type of *J. grandiflorum* (WF) is unique in Jasmine since the commercial varieties available in the market produce pink-tinged buds. CO.1 Winter Jasmine of *J. multiflorum* is a winter-season flowering type producing a high yield. These three genotypes have been shown to possess higher consumer and market preferences. However, bottlenecks have been observed in large-scale propagation of these novel types owing to the poor rhizogenic potential of stem cuttings of these genotypes.

Stem cuttings were taken from healthy two-year-old mother plants of the specified genotypes in the Jasmine Germplasm at TNAU, Coimbatore. The experiment used a 2 x 3 Factorial Completely Randomized Design (FCRD) with two main factors: type of cuttings and concentration of IBA. The first factor had two treatments, terminal cutting (C_1) and semi-hardwood cutting (C_2), while the second factor included four treatments: three IBA concentrations (0.5 g L^{-1} [G_1], 1 g L^{-1} [G_2] and 1.5 g L^{-1} [G_3]) and distilled water (G_4). Each treatment had three replications, with 15 cuttings per replication. The rooting process lasted 30 days, from February 20 to March 22, 2024, at 32°C/14°C Day/night temperatures and 88% relative humidity. IBA was dissolved in 1N NaOH and diluted to 0, 8, or 16 g L^{-1} with distilled water, with the solutions adjusted to pH 7.00 using 1N HCl. The base of each cutting was immersed in one of these solutions for 30 seconds before being planted in the rooting substrate.

Collection and preparation of plant materials

Two cuttings were used: (i) semi-hardwood cuttings from two-year-old mother plants with immature growth and 3-4 nodes without leaves, and (ii) mature terminal cuttings of 10-15 cm length with 8-10 leaves. Leaves on the cuttings stimulate root initiation by translocating carbohydrates to the stem base, aiding root growth. Young leaves and buds supply auxin, which moves basipetal to promote root growth. An inclined cut was made at the bottom, and a transverse cut was made at the top of each cutting. The basal end (2.5-3.0 cm) was dipped in 0.5% copper oxychloride solution for 10 minutes and then treated with a growth regulator solution. In polyethene bags (10 cm in diameter and 15 cm tall), the treated cuttings were planted vertically with a rooting substrate made up of equal portions of peat and perlite before being placed in a mist bed that maintained an approximately 85% relative humidity with 30±2°C temperature for at least three months. They observed days taken to sprout, buds sprouted, number of roots per cutting, root length, shoot length, number of leaves and rooting percentage.

Physiological and biochemical changes

Total phenol content (mg g^{-1})

The Folin-Ciocalteu method was employed to determine total phenolic content calorimetrically (13). About 250 mg of fresh leaves were placed in test tubes, and 5 ml of 80 % ethanol was added, followed by heating in a water bath at 80°C for 10 minutes before cooling down. After that, the sample was crushed using 80% ethanol and centrifuged at 5000 rpm for ten minutes. The supernatant was pipetted out and 1 ml was mixed with 1 ml of Folin-Ciocalteu phenol reagent and 2 ml of sodium carbonate. The mixture

was heated for 5-10 minutes in a water bath, and absorbance was measured using a UV-current spectrophotometer at 660 nm. The internal standard used was catechol. The fixed wavelength of 660 nm was selected based on previous studies, where it was identified as the maximum absorbance (λ max) for the Folin-Ciocalteu reaction (14).

Soluble protein (mg g^{-1})

Increasing soluble protein content increases photosynthetic efficiency, resulting in better plant growth and higher yields. The soluble protein content in leaf samples was determined using a specific method and expressed in mg g-1 fresh weight units. Leaf samples were collected during the early morning hours to ensure consistency and accuracy, typically between 6 AM and 9 AM (15). This is normally when photosynthesis begins, and soluble proteins are generally synthesized. Collecting samples during this time can provide insight into maximum protein activity levels, which can be immediately weighed after cutting them into pieces (1-2 cm). Using a pestle and mortar, the samples were then macerated with 10 ml of phosphate buffer (0.01M, pH 7.0). The extract was centrifuged at 10,000 rpm at 4°C for 20 minutes, and then 0.1 ml of the supernatant was mixed with 5 ml of a dye mixture. After 15 minutes, the colour intensity was measured at an optical density of 595 nm (15). For the preparation of the dye mixture, 100 mg of Coomassie Brilliant Blue (G250) was dissolved in 50 ml of 95% ethanol (molecular biology grade) or for higher purity and 100 ml of Orthophosphoric acid (Reagent grade (AR) is higher to avoid contamination. This solution was made up to 200 ml using distilled water. One ml of the dye solution was diluted with 4 ml of distilled water for sample analysis.

IAA oxidase activity µg of unoxidized auxin g^{-1} h^{-1})

The IAA oxidase activity was determined using a spectrophotometric method, with IAA (Indole-3-acetic acid) as the standard for measurement and comparison (16). The Garden Weber reagent was added to the aliquot extracted using phosphate buffer. The development of a pink colour was then measured at 540 nm. The IAA activity was expressed in µg of unoxidized IAA g^{-1} h^{-1}.

Statistical analysis

To make inferences, the calculated F values were compared with tabulated F values at a 5% significance level (P = 0.05). A factorial completely randomized design (FCRD) was used to analyze data collected in triplicate for different shoot and root parameters. Statistical analysis was performed using "TNAUSTAT," while the statistical program Star 2.0.1 was used for principal component analysis and basic descriptive statistics. The breeding tool GRAPES 1.1.0 was employed for correlation studies (17).

Results and Discussion

Shoot and Root Parameters

Days taken for sprouting and number of buds sprouted

The type of cuttings significantly affected the sprouting duration and number of sprouts per cutting. In *J. sambac* (DF), semi-hardwood cuttings sprouted the quickest, taking 13.13 days and yielded the highest number of sprouts per cutting (1.992), while terminal cuttings took 13.97 days and produced fewer sprouts (1.34). For *J. grandiflorum* (WF), terminal cuttings sprouted earlier (12.66 days) and had more sprouts per cutting (1.908) compared to semi-hardwood cuttings (13.086 days and 1.308 sprouts). In *J. multiflorum* (CO.1 WJ), terminal cuttings had the shortest sprouting time (13.21 days) and the highest number of sprouts per cutting (3.19), whereas semi-hardwood cuttings took 15.41 days and produced fewer sprouts (2.27).

Growth regulators and their interactions greatly influenced sprouting time and the number of sprouts per cutting. Semi-hardwood cuttings sprouted faster and produced more sprouts, likely due to stored carbohydrates aiding bud development. In vegetative propagation, food reserves in cuttings are crucial for early leaf bud development and subsequent root formation, which supports nutrient and water absorption. (Table 1) displays how various cuttings and growth regulators affected sprouting days and bud numbers in three *Jasminum* genotypes. These results align with similar research on other plant species, including studies on Jasmine (18), Bougainvillea (19) and (20), *Stevia rebaudiana* (19), *Cestrum nocturnum* (21), *Thuja compacta* (22) and carnation (9).

Number of leaves per cutting

A significant difference was observed in the number of leaves per cutting among different cuttings. In *J. sambac* (DF), semi-hardwood cuttings (C_2) exhibited the maximum number of leaves (4.75), while terminal cuttings (C_1) had the minimum (3.90). The interaction effect showed an increased number of leaves (6.60) in semi-hardwood cuttings treated with IBA 1g L^{-1} (C_2G_2), followed by 5.47 leaves in cuttings treated with IBA 1g L^{-1} (G_3). In *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ), terminal cuttings (C_1) had the maximum number of leaves (12.26 and 25.05, respectively), while semi-hardwood cuttings (C_2) had the minimum (9.33 and 14.78, respectively). The interaction effect showed an increased number of leaves in terminal cuttings (13.48) of *J. grandiflorum* (WF) and (31.70) J. *multiflorum* (CO.1 WJ).

The increase in leaf number of terminal cuttings can be attributed to greater height of the plant and a more significant number of branches. This results in enhanced root and shoot growth due to exogenous auxin-promoting callus formation, cell enlargement and protein production. However, the unavailability of callus and root initiation in hardwood cuttings led to dry twigs, while growth regulators improved nutrient uptake and leaf production (23). (Table 2) displays the effect of various types of cuttings with different regulators on the number of leaves per cutting in three new *Jasminum* genotypes. Similar results were observed in *Jasminum arborescence* and Bougainvillea glabra (24, 25).

Table 1. Effect of Different Types of Cuttings and Growth Regulators on Days Taken for Sprouting and Number of Buds Sprouted in three novel *Jasminum* Geno-

Parameters	Days taken for sprouting									Number of buds sprouted								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)			*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings → / Growth reg dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	13.33	12.53	12.84	12.45	12.36	12.40	12.88	16.66	14.77	1.33	2.03	1.68	1.70	1.33	1.51	3.20	2.82	3.01
G_2	13.40	12.72	13.06	12.11	12.44	12.27	12.77	14.44	13.60	1.42	2.36	1.89	2.36	1.47	1.91	3.36	2.34	2.85
G_3	13.56	11.94	12.75	11.44	13.03	12.23	12.22	14.89	13.55	1.47	2.34	1.91	2.34	1.42	1.88	3.31	2.36	2.83
G_4	15.41	15.48	15.48	14.66	14.50	14.58	14.99	15.67	15.33	1.13	1.22	1.17	1.22	1.13	1.17	2.92	1.58	2.25
Mean	13.97	13.13		12.66	13.08		13.22	15.41		1.34	1.99		1.90	1.30		3.19	2.27	
	S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)	
Cutting (C)	0.117	0.251		0.196	0.418		0.314	0.672		0.106	0.226		0.108	0.232		0.09	0.191	
Growth regulator (G)	0.166	0.355		0.277	0.519		0.444	0.950		0.150	0.320		0.153	0.328		0.127	0.271	
Interaction (C×G)	0.23	0.501		0.391	0.836		0.628	1.343		0.235	0.453		0.217	0.464		0.179	0.383	

Shoot length (cm)

In the case of *J. sambac* (DF), it was semi-hardwood cuttings (C_2) showed the greatest shoot length (8.62 cm), while terminal cuttings (C_1) recorded least length (7.13 cm). When considering a combination of cutting and auxin treatments, there were significant increases in the shoot lengths over that seen in control plants. The semi-hardwood cuttings with IBA 1g L^{-1} treated had a maximum shoot length (C_2G_2) of 10.26 cm, followed by IBA 1.5 g L^{-1} (9.93 cm) (C_2G_3). As for *J. grandiflorum* (WF) as well as *J. multiflorum* (CO.1 WJ), terminal cuttings recorded the highest growth rates (10.24 cm and 10.45 cm, respectively).

Cuttings and auxin treatments significantly resulted in longer shoot lengths than the control. When IBA at 1 g L^{-1} was utilized on terminal cuttings, a maximum of 14.30 cm in *J. grandiflorum* (White Flower Bud type) and 14.14 cm in *J. multiflorum* (CO.1 WJ) were recorded for both plants' shoots. Different cuttings and growth regulators stimulated shoot length in three *Jasminum* genotypes, as shown in (Table 2) below. This increase in shoot length in terminal cuttings may be due to increased root growth and a more significant number of roots per cutting, which enhance their water and nutrient absorption capacities. IBA treatment boosts cell division, lengthening and protein production, resulting in flourishing vegetative development. Identification of various plants having similar properties was done in *Bellis perennis* and *Tecoma stans* (26, 27).

Root length (cm)

Generally, an increased number of roots per cutting resulted in a shorter average root length. In *J. sambac* (DF), semi-hardwood cuttings (C_2) had the longest root length (10.313 cm), whereas terminal cuttings (C_1) had the shortest (9.225 cm). Among the growth regulator treatments, cuttings treated with IBA 1g L^{-1} (G_2) exhibited the longest root length (12.747 cm), while the control group (G_4) had the shortest (7.128 cm). The longest roots were found in semi-hardwood cuttings treated with IBA 1g L^{-1} (C_2G_2), followed by those treated with IBA 1.5 g L^{-1} (C_2G_3). In *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ),

Table 2. Effect of different types of cuttings and Growth regulators on Days taken for sprouting and number of buds sprouted in three novel *Jasminum* genotypes

Parameters	Number of leaves									Shoot length (cm)								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)			*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings → / Growth red dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	3.36	3.36	3.47	13.07	11.56	12.31	24.70	21.59	23.15	6.76	9.04	7.90	9.92	5.77	7.85	10.91	6.81	8.86
G_2	5.47	6.60	6.03	13.48	11.90	12.69	31.70	23.11	27.40	8.25	10.26	9.25	14.30	6.81	10.56	14.14	6.82	10.46
G_3	4.36	5.40	4.89	13.08	12.05	12.56	28.51	23.74	26.13	7.60	9.93	8.76	12.45	6.79	9.62	12.22	7.37	9.79
G_4	2.40	3.39	2.90	9.44	9.33	9.33	15.30	14.78	15.04	5.94	5.23	5.58	4.30	4.13	4.22	4.54	4.69	4.61
Mean	3.90	4.75		12.26	11.18		25.05	20.80		7.13	8.62		10.24	5.87		10.45	6.42	
	S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)	
Cutting (C)	0.067	0.143		0.064	0.195		0.089	0.191		0.048	0.102		0.022	0.047		0.034	0.072	
Growth regulator (G)	0.095	0.202		0.091	0.275		0.126	0.27		0.067	0.144		0.031	0.067		0.048	0.102	
Interaction (C×G)	0.134	0.286		0.129	0.390		0.178	0.381		0.095	0.204		0.044	0.094		0.068	0.145	

terminal cuttings (C_1) had the longest roots (10.13 cm and 15.14 cm, respectively), while semi-hardwood cuttings (C_2) had the shortest (5.64 cm and 8.149 cm respectively). The longest roots were observed in terminal cuttings treated with IBA 1g L^{-1} ppm (C_1G_2), followed by those treated with IBA 1.5 g L^{-1} (C_1G_3). The length (cm) of roots concerning three novel *Jasminum* genotypes is demonstrated through the impact of various cuttings and growth regulators (Table 3).

The increased root length can be attributed to early callus formation, cell differentiation, enhanced cell elongation and vascular tissue differentiation, all promoting root growth. Adventitious root formation (ARF) is a complex developmental process involving various biochemical, physiological and histological events during root primordia's induction, initiation, development and elongation (Fig.1).

The observed increase in root length compared to the control group might be attributed to the augmentation of carbohydrate hydrolysis, accumulation of metabolites, and cell division produced by auxin (28). These results are consistent with previous studies conducted on *Jasminum sambac*, *Grewia optiva* and *Robinia pseudoacacia*, *Bougainvillea glabra*, and *Dendranthema grandiflora* cv. Snowball, *Cestrum nocturnum*, *Tagetes erecta* marigold and carnation (8, 9, 21, 25, 29-32).

Number of roots per cutting

In *J. sambac* (DF), semi-hardwood cuttings (C_2) produced the highest number of roots per cutting (7.449) compared to terminal cuttings (C_1), which showed a minimum of 4.143 roots per cutting. Among the auxin treatments, cuttings treated with IBA at 1g L^{-1} (G_2) resulted in the maximum number of roots (9.34). The interaction effect of cutting type and growth regulator concentration was significant, with the maximum number of roots per cutting (19.35) observed in semi-hardwood cuttings treated with IBA at 1g L^{-1} (C_2G_2). However, there was less root formation in terminal cuttings (4.097) treated with IBA at 500 ppm (C_1G_1). In the control treatment, the maximum number of roots formed in semi-hardwood cuttings (C_2G_4) was 7.44. In *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ), terminal cuttings (C_1) had an increased number of roots (10.214 and 16.523, respectively), while semi-hardwood cuttings (C_2) showed a minimum of 7.025 and 7.452 roots respectively. The effect of different types of cuttings and growth regulators on number of roots per cutting in three novel *Jasminum* genotypes is shown in (Table 3).

The fewer roots found on some cuttings could result from the fact that initially, more vegetative shoots may develop, leading to lower callus formation and root initiation. Additionally, rooting could be induced by the increased velocity of moving sugars to the base of cuttings, making IBA application necessary to promote translocation indirectly (33). This agrees with what was observed in *Cestrum nocturnum* (21), *Hibiscus rosa-sinensis* (34), *Bougainvillea glabra* (35) and *Duranta repens* (27)**.** Many herbaceous perennial crops typically grow an increasing number of roots with auxin application (36). Similar results have been found in Chrysanthemum, Mexican Snow Ball and Night Queen (21, 37, 38).

Rooting percentage

The various cuttings had statistically different survival rates. According to the results obtained from *J. sambac* (DF), 80.97% (C_2) was observed in semi-hardwood cuttings, while 73.54% (C_1) was recorded for terminal cuttings. Furthermore, among all plant growth regulator treatments used, IBA at a concentration of 1000 ppm showed 88.6% (G_2) as the highest survival rate, while the least survival

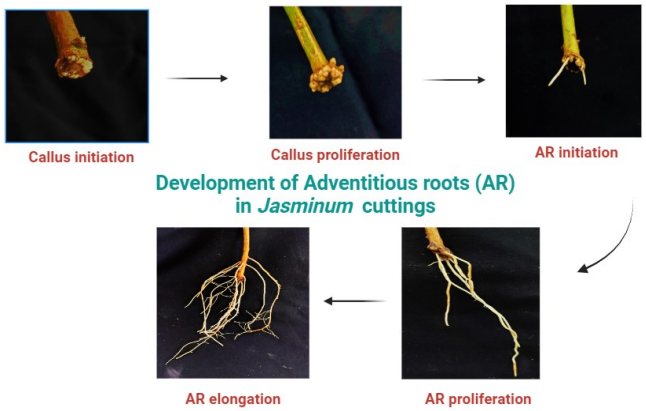


Fig. 1. Development of adventitious roots (AR) in *Jasminum* cuttings

Table 3. Effect of different types of cuttings and Growth regulators on root length and number of roots per cutting in three novel *Jasminum* genotypes

Parameters	Root length (cm)									No of roots per cutting								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)			*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings →																		
Growth reg dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	9.74	10.03	9.88	11.34	4.93	8.138	16.14	8.14	12.14	4.09	6.70	5.40	10.23	6.51	8.37	14.23	9.89	12.06
G_2	10.22	12.74	11.48	12.03	8.14	10.08	17.22	9.18	13.20	5.09	9.34	7.21	13.52	7.51	10.51	16.52	8.84	12.68
G_3	9.91	11.24	10.57	11.94	6.24	9.09	17.06	10.14	13.60	4.24	8.34	6.29	11.66	8.34	10.00	15.66	7.34	11.50
G_4	7.02	7.23	7.12	5.22	3.26	4.24	10.15	5.12	7.63	3.13	5.41	4.27	5.44	5.73	5.588	4.44	3.77	4.08
Mean	9.22	10.31		10.13	5.64		15.14	8.14		4.13	7.44		10.21	7.02		10.21	7.45	
	S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)		S.E(d) ±	CD(5%)	
Cutting (C)	0.009	0.019		0.030	0.030		0.010	0.020		0.040	0.085		0.089	0.191		0.128	0.275	
Growth regulator (G)	0.012	0.026		0.092	0.043		0.013	0.029		0.560	0.120		0.126	0.27		0.182	0.388	
Interaction (C×G)	0.017	0.017		0.129	0.061		0.019	0.041		0.079	0.169		0.178	0.381		0.257	0.549	

was noted in one from the control group at 69.26% (G_4). The interaction effect between types of cutting and growth regulators established that 1g L^{-1} IBA applied on semi-hardwood cuttings resulted in a higher survival percentage of 88.6% (C_2G_2) than those treated with IBA at a concentration of 1.5 g L^{-1} which resulted to 83.31% (C_2G_3).

In *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ), terminal cuttings showed the maximum rooting percentage of 41.44% and 72.23%, respectively, while semi -hardwood cuttings had a minimum of 33.34% and 35.47% respectively. Among the plant growth regulator treatments, IBA 1g L^{-1} showed the maximum survival percentage of 48.15% in *J. grandiflorum* (WF) and 72.23 % in *J. multiflorum* (CO.1 WJ), while the control showed a minimum of 29.30% and 29.26%, respectively. The interaction effect between the type of cutting and concentration of growth regulator indicated that terminal cuttings treated with IBA 1g L^{-1} recorded an increased rooting percentage of 48.15% in *J. grandiflorum* (WF) and 72.23% in *J. multiflorum* (CO.1 WJ), followed by 46.15% and 70.26% in treated terminal cuttings (C_1). The effect of different types of cuttings and growth regulators on rooting percentage in three novel *Jasminum* genotypes is shown in (Table 4). The terminal and semi-hardwood cuttings showed a rise in rooting percentage, which can be linked to the standard root and shoot parameter growth, while hardwood cuttings were minimal for them. Similar findings have been reported in *Lonicera japonica* and *Rosa damascena* (28, 39).

PCA for morphological parameters

Principal Component Analysis (PCA) reduces complex datasets to uncorrelated principal components, capturing the most variance in the data (40). This technique highlights critical factors influencing rooting and reproduction, simplifying the analysis of physiological, morphological and environmental conditions affecting *Jasminum* propagation through stem cuttings.

Eigenvalues, % of variation and % contribution of each variable

In the study, eigenvalues greater than 1 were observed in the principal component (PC1) for all novel three *Jasminum* genotypes: *J. sambac* (DF) with 5.099 (84.977% of total divergence), *J. grandiflorum* (WF) with 5.403 (90.043% of total divergence) and *J. multiflorum* (CO.1 WJ) with 5.19 (86.502% of total divergence) (Table 5). The

Table 4. Effect of different types of cuttings and Growth regulators on rooting percentage (%) in three novel *Jasminum* genotypes

Parameters	Rooting percentage (%)								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings → / Growth reg dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	72.40	82.71	77.55	42.16	35.37	38.76	71.40	36.71	54.05
G_2	78.23	88.60	83.41	48.15	36.31	42.33	72.23	38.60	55.41
G_3	76.26	83.31	79.78	46.15	32.37	39.26	70.26	37.31	53.78
G_4	67.29	69.26	68.26	29.30	29.32	29.31	44.62	29.26	36.94
Mean	73.54	80.97		41.44	33.34		64.63	35.47	
	S.E(d) ±		CD(5%)	S.E(d) ±		CD(5%)	S.E(d) ±		CD(5%)
Cutting (C)	0.128		0.060	0.124		0.058	0.246		0.115
Growth regulator (G)	0.181		0.085	0.175		0.082	0.347		0.162
Interaction (C×G)	0.257		0.120	0.248		0.116	0.491		0.230

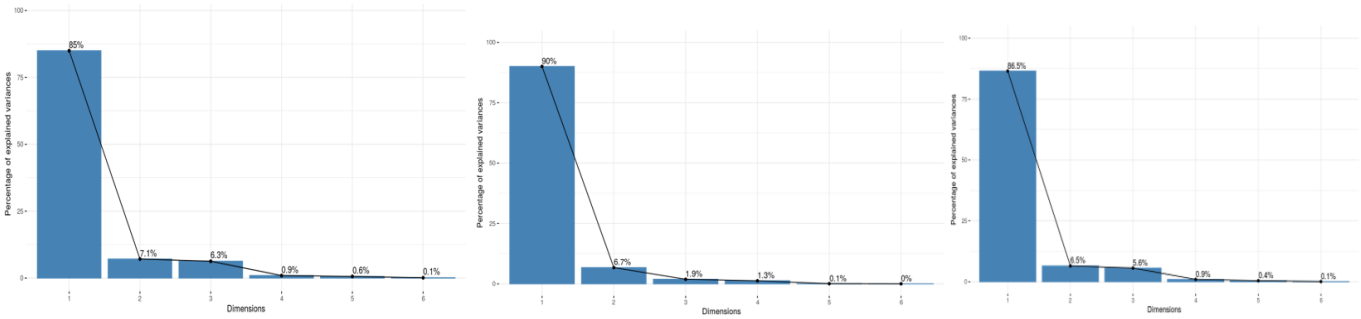
Table 5. Eigenvalues of Novel Three *Jasminum* genotypes

Principal Component	Eigenvalue	% of variance	Cumulative % of variance
	***J. sambac* (DF)**		
PC1	5.099	84.977	84.977
PC2	0.427	7.113	92.091
PC3	0.377	6.278	98.369
PC4	0.056	0.928	99.297
PC5	0.036	0.592	99.889
PC6	0.007	0.111	100
	***J. grandiflorum* (WF)**		
PC1	5.403	90.043	90.043
PC2	0.403	6.719	96.762
PC3	0.113	1.88	98.642
PC4	0.076	1.266	99.908
PC5	0.004	0.068	99.976
PC6	0.001	0.024	100
	***J. multiflorum* (CO.1 WJ)**		
PC1	5.19	86.502	86.502
PC2	0.389	6.49	92.992
PC3	0.337	5.611	98.603
PC4	0.056	0.93	99.533
PC5	0.024	0.403	99.936
PC6	0.004	0.064	100

Note: Days taken for Sprouting (**DS**), Number of Buds Sprouted (**NS**), Number of Leaves (**NL**), Number of Roots (**NR**), Root Length (**RL**) and Shoot Length

percentage of variation in relation to each principal component could be demonstrated by a scree plot obtained by a graph between eigenvalues and principal component numbers (Fig. 2).

The graph shows that in *J. sambac* (DF), the first principal component (PC1) had an eigenvalue of 5.099 (84.977%). In *J. grandiflorum* (WF), PC1 had an eigenvalue of 5.403 (90.043%). In *J. multiflorum* (CO.1 WJ), PC1 had an eigenvalue of 5.19 (86.502%). Eigenvalues gradually decreased with increasing principal components. The maximum contribution to the variance was due to PC1, followed by PC2. The PC1 showed the maximum contribution of variables on principal components with parameters *viz*., such as days taken for sprouting (DS), number of buds sprouted (NS), number of leaves (NL), number of roots (NR), root length (RL) and shoot length



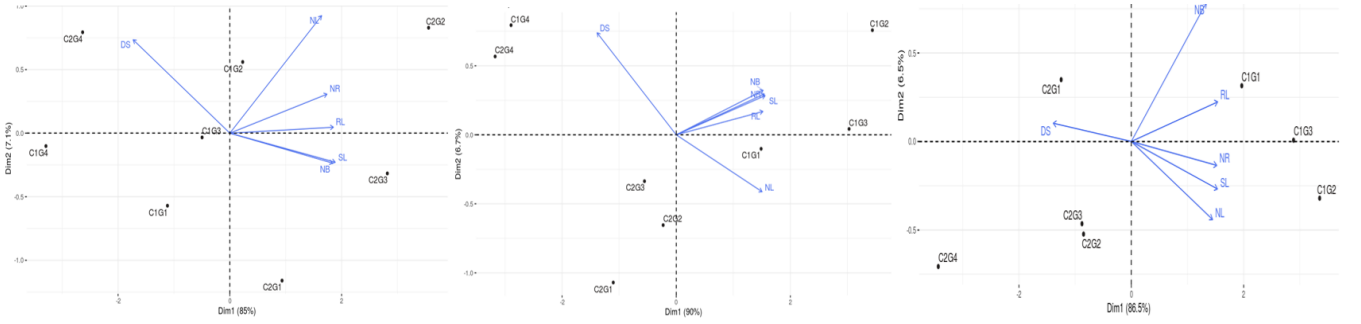
J. sambac (DF) *J. grandiflorum* (WF) *J. multiflorum* (CO.1 WJ)

Fig. 2. Scree plot of variables

Table 6. Percent Contribution of variables on principal components of novel three *Jasminum* genotypes

Variables	PC1	PC2	PC3	PC4	PC5	PC6
			***J. sambac* (DF)**			
DS	15.703	33.815	10.037	26.243	26.243	0.005
NS	17.699	3.472	20.415	2.103	2.103	53.525
NL	14.135	53.417	12.124	13.923	13.923	5.88
NR	15.828	5.883	42.04	1.031	1.031	21.388
RL	17.994	0.137	14.981	56.695	56.695	0.545
SL	18.64	3.276	0.403	0.006	0.006	18.656
			***J. grandiflorum* (WF)**			
DS	14.116	53.748	13.955	5.989	7.618	4.573
NS	17.017	10.187	29.708	6.019	36.34	0.729
NL	16.613	16.73	16.679	20.089	19.727	10.162
NR	17.507	8.597	0.013	24.707	1.729	47.447
RL	16.938	2.813	37.711	40.797	0.341	1.401
SL	17.809	7.925	1.935	2.399	34.245	35.687
			***J. multiflorum* (CO.1 WJ)**			
DS	15.022	1.099	63.251	0.64	9.715	10.273
NS	13.975	64.167	6.813	2.299	0.52	12.227
NL	16.302	20.177	14.4929	46.484	2.521	0.023
NR	18.012	1.887	8.495	39.948	27.986	3.673
RL	18.381	5.201	6.108	0.835	10.138	59.336
SL	18.308	7.469	0.841	9.794	49.12	14.469

Note: Days taken for Sprouting (**DS**), Number of Buds Sprouted (**NS**), Number of Leaves (**NL**), Number of Roots (**NR**), Root Length (**RL**) and Shoot Length (**SL**).



J. sambac (DF) *J. grandiflorum* (WF) *J. multiflorum* (CO.1 WJ)

Fig. 3. Contribution of variables on principal component

(SL) as given in the (Table 6) and (Fig. 3).

The correlation between variables and the principal component of commonly three *Jasminum* genotypes are represented in (Table 7) and (Fig. 4). These findings align with the principal component analyses of key traits in Mysuru Jasmine (41).

Biochemical changes

Total phenol content

A significant increase in phenol content was observed across all treatments (Table 8). In *J. sambac* (DF), semi-hardwood cuttings with IBA 1000 ppm showed the highest phenol content (2.673 mg g^{-1}), while the control had the lowest. For *J. grandiflorum* (WF), terminal cuttings with IBA 1000 ppm had the highest phenol content (3.157 mg g^{-1}); the lowest was in the control. In *J. multiflorum* (CO.1 WJ), terminal cuttings with IBA 1000 ppm also had the highest phenol content (2.673 mg g^{-1}), while the control had the lowest.

In *J. sambac* (DF), IBA-treated semi-hardwood cuttings exhibited the highest leaf phenol content. Conversely, IBA-treated terminal cuttings showed the highest phenol levels in *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ). This phenol increase may result from elevated H_2O_2 production due to enhanced respiration or activation of pathways like the hexose monophosphate shunt and acetate pathways, which release conjugated phenols via hydrolytic enzymes. Additionally, phenol accumulation could stem from sugar depletion used in their biosynthesis. Similar findings of increased phenolic content with IBA treatment have been observed in *Olea europaea* (34) and *Polianthes tuberosa*

Table 7. Correlation between variables and PCs of novel three *Jasminum* genotypes

Variables	PC1	PC2	PC3	PC4	PC5	PC6
			J. sambac (DF)			
DS	15.703	33.815	10.037	26.243	26.243	0.005
NS	17.699	3.472	20.415	2.103	2.103	53.525
NL	14.135	53.417	12.124	13.923	13.923	5.88
NR	15.828	5.883	42.04	1.031	1.031	21.388
RL	17.994	0.137	14.981	56.695	56.695	0.545
SL	18.64	3.276	0.403	0.006	0.006	18.656
			***J. grandiflorum* (WF)**			
DS	14.116	53.748	13.955	5.989	7.618	4.573
NS	17.017	10.187	29.708	6.019	36.34	0.729
NL	16.613	16.73	16.679	20.089	19.727	10.162
NR	17.507	8.597	0.013	24.707	1.729	47.447
RL	16.938	2.813	37.711	40.797	0.341	1.401
SL	17.809	7.925	1.935	2.399	34.245	35.687
			***J. multiflorum* (CO.1 WJ)**			
DS	15.022	1.099	63.251	0.64	9.715	10.273
NS	13.975	64.167	6.813	2.299	0.52	12.227
NL	16.302	20.177	14.4929	46.484	2.521	0.023
NR	18.012	1.887	8.495	39.948	27.986	3.673
RL	18.381	5.201	6.108	0.835	10.138	59.336
SL	18.308	7.469	0.841	9.794	49.12	14.469

Note: Days taken for sprouting (**DS**), Number of Buds Sprouted (**NS**), Number of Leaves (**NL**), Number of Roots (**NR**), Root Length (**RL**) and Shoot Length

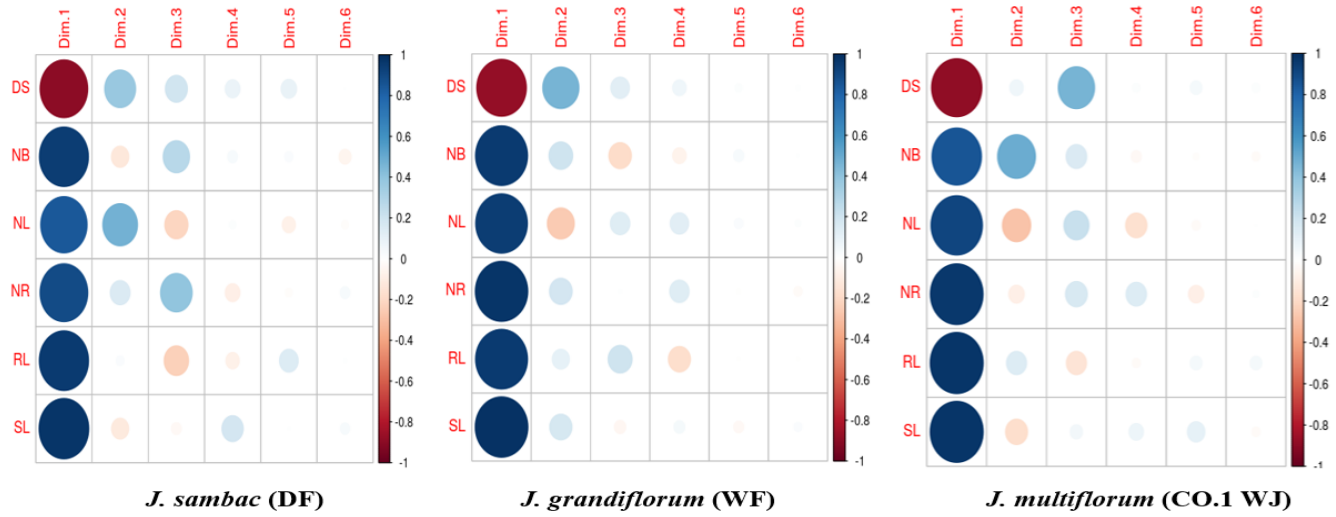


Fig. 4. Correlation between variables and principal component

Table 8. Effect of different types of cutting and growth regulators on phenol content (mg g^{-1}) for novel three *Jasminum* genotypes

Parameters	Phenol content (mg g^{-1})								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings → / Growth reg dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	2.06	2.47	**2.26**	3.03	2.47	**2.75**	2.47	2.06	**2.26**
G_2	2.15	2.67	**2.41**	3.15	2.67	**2.91**	2.67	2.15	**2.41**
G_3	2.17	2.48	**2.32**	3.08	2.48	**2.78**	2.48	2.17	**2.32**
G_4	1.06	1.05	**1.05**	2.06	2.05	**2.055**	1.05	1.06	**1.05**
Mean	**1.86**	**2.16**		**2.83**	**2.41**		2.16	**1.86**	
	S.E(d) ±		CD(P=0.05)	S.E(d) ±		CD(P=0.05)	S.E(d) ±		CD(P=0.05)
Cutting (C)	0.035		0.076	0.035		0.074	0.035		0.076
Growth regulator (G)	0.050		0.107	0.049		0.104	0.050		0.107
Interaction (C×G)	0.071		0.151	0.069		0.148	0.071		0.151

(42).

Soluble protein

The soluble protein content varied significantly across treatments in different jasmine genotypes (Table 9). In *J. sambac* (DF), semi-hardwood cuttings with IBA 1000 ppm had the highest protein content (13.410 mg g^{-1}), while terminal cuttings and the control had the lowest. For *J. grandiflorum* (WF), terminal cuttings with IBA 1000 ppm showed the highest protein content (13.423 mg g^{-1}), with semi-hardwood cuttings and the control being lower. Similarly, in *J. multiflorum* (CO.1 WJ), terminal cuttings treated with IBA 1000 ppm recorded the highest protein content (13.553 mg g^{-1}), while the control had the lowest.

In *J. sambac* (DF), IBA-treated semi-hardwood cuttings had the highest soluble protein content in leaves. In contrast, IBA-treated terminal cuttings exhibited the highest levels in *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ). Plant growth elements enhance the source-sink relationship, boosting the translocation of photoassimilates and supporting organ development. This aligns with the findings that auxin treatment regulates starch and protein content (43). Similar increases in soluble protein due to IBA treatment have been observed in *Berberis thunbergii* (44) and *Pleurotus sajorcaju* (45).

IAA oxidase activity

In *J. sambac* (DF), terminal cuttings had the highest IAA oxidase activity, while semi-hardwood cuttings with IBA 1000 ppm showed the lowest. For *J. grandiflorum* (WF), terminal cuttings with IBA 1000 ppm had the lowest activity, and semi-hardwood cuttings had the highest. In *J. multiflorum* (CO.1 WJ), terminal cuttings with IBA 1000 ppm exhibited the lowest IAA oxidase activity, while semi-hardwood cuttings had the highest (Table 10).

IAA oxidase is responsible for auxin degradation through oxidation, so increased enzyme activity reduces auxin levels and slows plant growth. In this study, *J. sambac* (DF) semi-hardwood cuttings treated with IBA showed the lowest IAA oxidase activity. Similarly, terminal cuttings of *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ) with IBA also had minimal IAA oxidase activity. Proper nutrition and optimal conditions are crucial for maintaining healthy plant growth and efficient physiological processes (46- 48).

Table 9. Effect of different types of cutting and growth regulators on Soluble protein (mg g^{-1}) for novel three *Jasminum* genotypes

Parameters	Soluble Protein (mg g^{-1})								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings → / Growth reg dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	12.05	13.02	**12.54**	13.02	11.05	**12.04**	13.25	12.22	**12.74**
G_2	12.22	13.41	**12.81**	13.42	12.17	**12.79**	13.55	12.25	**12.90**
G_3	12.27	13.35	**12.81**	13.31	12.12	**12.71**	13.35	12.22	**12.79**
G_4	10.07	10.22	**10.14**	10.12	10.05	**10.08**	10.32	10.04	**10.18**
Mean	**11.65**	**12.50**		**12.47**	**11.35**		**12.62**	**11.68**	
	S.E(d) ±		CD(P=0.05)	S.E(d) ±		CD(P=0.05)	S.E(d) ±		CD(P=0.05)
Cutting (C)	0.020		0.042	0.016		0.034	0.029		0.062
Growth regulator (G)	0.028		0.059	0.023		0.049	0.041		0.088
Interaction (C×G)	0.039		0.083	0.032		0.069	0.058		0.124

Table 10. Effect of different types of cutting and growth regulators on IAA oxidase (µg of unoxidized auxin g^{-1} h^{-1}) for novel three *Jasminum* genotypes

Parameters	IAA oxidase (µg of unoxidized auxin g^{-1} h^{-1})								
Genotypes	*J. sambac* (DF)			*J. grandiflorum* (WF)			*J. multiflorum* (CO.1 WJ)		
Cuttings → / Growth reg dosage ↓	C_1	C_2	Mean	C_1	C_2	Mean	C_1	C_2	Mean
G_1	15.90	14.94	**12.74**	11.46	12.23	**11.84**	10.26	12.51	**11.84**
G_2	15.91	12.91	**12.79**	11.07	13.41	**12.24**	9.72	11.43	**10.82**
G_3	15.57	13.84	**12.90**	12.15	13.77	**12.96**	10.22	11.73	**10.73**
G_4	16.74	16.57	**10.18**	14.19	14.38	**14.28**	11.59	14.72	**13.15**
Mean	**16.74**	**14.52**		**12.22**	**13.45**		**10.45**	**12.60**	
	S.E(d) ±	**CD(P=0.05)**		**S.E(d) ±**	**CD(P=0.05)**		**S.E(d) ±**	**CD(P=0.05)**	
Cutting (C)	0.012	0.027		0.078	0.166		0.036	0.078	
Growth regulator (G)	0.018	0.038		0.110	0.234		0.051	0.110	
Interaction (C×G)	0.025	0.053		0.155	0.332		0.073	0.155	

Conclusion

This 2-year study found that different dosages of IBA and different types of cuttings significantly influenced the properties of different *Jasminum* genotypes. It was determined that *J. sambac* (DF) had a more significant rooting proportion for semi-hardwood cuttings since it had superior root and shoot growth than terminal cuttings. On the other hand, terminal cuttings in *J. grandiflorum* (WF) and *J. multiflorum* (CO.1 WJ) had a higher survival rate accompanied by better root and shoot development than semi-hardwood ones. Among various concentrations of Indole Butyric Acid (IBA), 1g L^{-1} was found to be most effective in enhancing the rooting of *J. sambac* (DF) semi-hardwood cuttings as well as terminal ones obtained from *J. grandiflorum* (WF) and *J. multiflorum* (CO. WJ). This concentration promoted the rapid multiplication of these species.

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Authors' contributions

KR carried out the experiment, took observations and analyzed the data. GM guided the research by formulating the research concept, helped secure funds, and approved the final manuscript. CR reviewed the manuscript and helped in procuring research grants. VK contributed by imposing the experiment and helped edit, summarise, and revise the manuscript. SRC helped summarize and revise the manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interests to declare.

Ethical issues: None.

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

During the preparation of this work, the author(s) did not used AI tools and the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

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