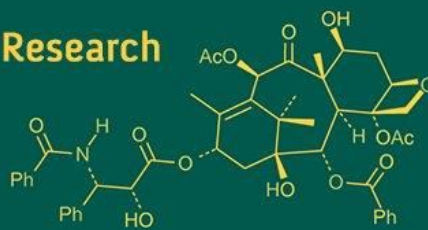
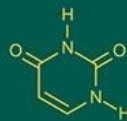
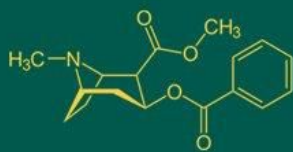


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Rooting response of *Hydrangea macrophylla* cuttings to IBA and media under subtropical conditions

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Abstract

The successful propagation of *Hydrangea macrophylla* through stem cuttings depends on multiple factors, including cutting type, rooting medium, and growth regulator application. This study was conducted under subtropical conditions of Bhubaneswar, Odisha, to evaluate the rooting response of three types of stem cuttings (softwood, semi-hardwood, and hardwood) treated with three concentrations of indole-3-butyric acid (IBA: 0, 250, and 500 ppm) and planted in four rooting media (sand, perlite, vermiculite, and soilrite). Observations recorded 75 days after planting revealed that softwood cuttings treated with 250 ppm IBA and rooted in perlite produced the maximum number of roots and the greatest root length. Among media, perlite and sand significantly outperformed vermiculite and soilrite in promoting root initiation and elongation. The interaction between cutting type, IBA concentration, and rooting medium showed statistically significant effects on rooting traits. These findings demonstrate that softwood cuttings treated with 250 ppm IBA and rooted in perlite provide the most effective combination for successful vegetative propagation of *Hydrangea macrophylla* in subtropical climates. The results offer practical recommendations for nurserymen and horticulturists aiming to standardize propagation protocols for large-scale production of this ornamental shrub.

Keywords: *Hydrangea macrophylla*, rooting, Stem cuttings, Indole-3-butyric acid (IBA), vegetative propagation

1. Introduction

Hydrangea macrophylla, an ornamental shrub renowned for its large and vibrant inflorescences, is widely cultivated in landscapes and home gardens for its aesthetic value. Propagation of this species is commonly accomplished through stem cuttings, which provide a cost-effective and reliable method for large-scale multiplication. However, the success of vegetative propagation is strongly influenced by factors such as cutting type, rooting media, and the application of growth regulators, particularly auxins like Indole-3-butyric acid (IBA) (Mostafa, 1996; Paul *et al.*, 1992) ^[10, 11].

The physiological maturity of cuttings plays a vital role in determining rooting success. Softwood cuttings generally exhibit higher rooting potential due to their juvenility and greater metabolic activity, whereas semi-hardwood and hardwood cuttings often root less efficiently (Costa *et al.*, 2002; Luz *et al.*, 2007) ^[2, 7]. Similarly, the rooting medium significantly affects moisture availability, aeration, and nutrient balance, which together influence root initiation and elongation. Studies have demonstrated that well-aerated substrates such as perlite and sand enhance root formation compared to more compact media like soilrite (Luz *et al.*, 2007) ^[7].

Exogenous application of IBA has been consistently reported to enhance root initiation and elongation across various ornamental crops. IBA promotes cell division and differentiation at the basal end of cuttings, thereby increasing root number and length (Pimeta *et al.*, 2003) ^[12]. In *Hydrangea macrophylla*, however, limited research has been conducted under subtropical climatic conditions, despite its growing popularity and demand in regions such as Odisha, India.

Recent studies have emphasized the role of growth regulators and novel treatments in

improving rooting efficiency in ornamentals. For instance, Sahoo *et al.* (2025) ^[13] reported that benzyl adenine combined with aspirin significantly enhanced rooting in chrysanthemum cuttings. Similarly, Maharana *et al.* (2025) ^[9] and Jena *et al.* (2025) ^[4] highlighted the positive effects of biostimulants and bioenzymes in improving vegetative propagation in ornamentals. These findings underscore the importance of optimizing both hormonal treatments and rooting substrates for different species and environments.

Considering these aspects, the present study was undertaken to evaluate the rooting response of *Hydrangea macrophylla* stem cuttings to different types of cuttings, concentrations of IBA, and rooting media under subtropical conditions. The aim was to identify effective propagation strategies that can enhance rooting success and provide practical recommendations for nurserymen and horticulturists in similar agro-climatic regions.

2. Materials and Methods

2.1. Experimental Site

The experiment was conducted at the Department of Floriculture and Landscaping, College of Agriculture, OUAT, Bhubaneswar, Odisha. The site experiences a humid subtropical climate.

2.2. Experimental Design

The experiment was laid out in a Factorial CRBD with:

- 3 cutting types (softwood, semi-hardwood, hardwood),
- 3 IBA concentrations (0, 250, and 500 ppm), and
- 4 rooting media (sand, perlite, vermiculite, soilrite).

Each treatment was replicated thrice with 5 cuttings per replication.

2.3. Preparation of Cuttings

Healthy mother plants of *Hydrangea macrophylla* were used. Cuttings were categorized as:

- **Softwood:** terminal green tips,
- **Semi-hardwood:** mid-section with greenish-brown bark,
- **Hardwood:** basal brown woody segments.

Cuttings (~10–15 cm) were prepared by removing lower leaves and immersed in IBA solution (250 ppm or 500 ppm) for 30 minutes. Controls were dipped in distilled water.

2.4. Rooting Media

Sterile prostrays were filled with the following media:

- Sand (well-drained and neutral)
- Perlite (high aeration)
- Vermiculite (moisture retentive)

- Soilrite (organic + inorganic mix).

Cuttings were placed in prostrays under 75% agro-shade net and watered regularly.

2.5. Observations

Rooting performance was evaluated 75 days after planting:

- Number of roots per cutting
- Length of longest root (cm)

2.6. Statistical Analysis

Data were analyzed using ANOVA under factorial CRBD. Angular transformation was applied to percentage data before analysis. Critical difference was calculated at 5% level of significance.

3. Results

3.1. Number of Roots per Cutting

The number of roots per cutting was significantly influenced by the type of cutting, IBA concentration, rooting media, and their interactions. From Table 1, it was observed that among the types of cuttings, softwood cuttings exhibited the highest root proliferation, with an average of 11.56 roots per cutting. This was followed by semi-hardwood (9.08) and hardwood cuttings (7.54). The increased rooting ability of softwood cuttings could be attributed to their physiological juvenility and higher metabolic activity, which facilitate quicker cell division and root primordia initiation.

IBA application significantly enhanced root development. Cuttings treated with 250 ppm IBA produced the highest average number of roots (11.74), which was significantly superior to both 500 ppm (9.49) and the control (7.86). Rooting media also had a marked effect. Perlite supported the highest average root number (12.07), likely due to its excellent aeration and moisture retention properties. It was followed by sand (10.21), vermiculite (8.53), and soilrite (6.98).

Interaction effects showed that the combination of softwood cuttings × 250 ppm IBA × perlite produced the highest root number (14.67), followed closely by softwood × 250 ppm IBA × sand (13.33). In contrast, the lowest number of roots (5.00) was recorded in hardwood × control × soilrite, indicating that mature tissues without auxin support and poorly aerated media are less favorable for rooting. The interaction between cutting type, IBA concentration, and rooting media significantly influenced the number of roots per cutting (Table 1). The results are also presented graphically (Fig.1.), which clearly shows that softwood cuttings treated with 250 ppm IBA and rooted in perlite produced the maximum root number, while hardwood cuttings generally recorded the lowest values.

Table 1: Effect of types of cutting, growth regulator and media on number of roots at 75 days after planting

Type of cuttings	IBA Concentration (ppm)	Media				Mean	Grand mean
		Sand	Perlite	Vermiculite	Soilrite		
Softwood cuttings	IBA 500 ppm	27.00	26.67	8.00	6.33	17.00	
	IBA 250 ppm	40.33	43.00	9.33	5.33	24.49	
	IBA 0ppm (control)	41.00	34.67	3.67	5.67	21.25	
	Mean	36.11	34.78	7.00	5.77		20.91
Semi	IBA 500 ppm	11.33	9.00	7.67	4.67	8.16	
	IBA 250 ppm	3.67	7.67	9.00	3.00	5.83	

hardwood cuttings	IBA 0ppm (control)	19.67	3.67	4.33	17.00	11.16	
	Mean	11.55	6.78	7.00	8.22		8.38
Hardwood cuttings	IBA 500 ppm	6.33	45.00	28.00	40.33	29.91	
	IBA 250 ppm	16.67	27.33	35.67	11.00	22.66	
	IBA 0ppm (control)	14.67	26.67	25.67	5.33	17.91	
	Mean	12.55	33.00	29.78	18.88		23.55
IBA 500 ppm		14.89	26.89	14.56	17.11		18.36
IBA 250 ppm		20.22	26.00	18.00	6.44		17.66
IBA 0ppm (control)		25.11	21.67	11.22	9.33		16.83
Grand Mean		20.07	24.85	14.59	10.95		
Effects				'F' Test	SE(M)±	CD at 5%	
Types of cuttings				**	1.55	4.38	
Growth regulator concentration (ppm)				NS	-	-	
Media				**	1.79	5.05	
Interaction							
Cuttings × Growth regulator conc.				**	2.68	7.58	
Cutting × Media				**	3.10	8.75	
Growth regulator conc. × Media				**	3.10	8.75	
Cutting × Growth regulator conc. × Media				**	5.36	15.16	

* = Significant at 5% level. ** = Significant at 1% level.

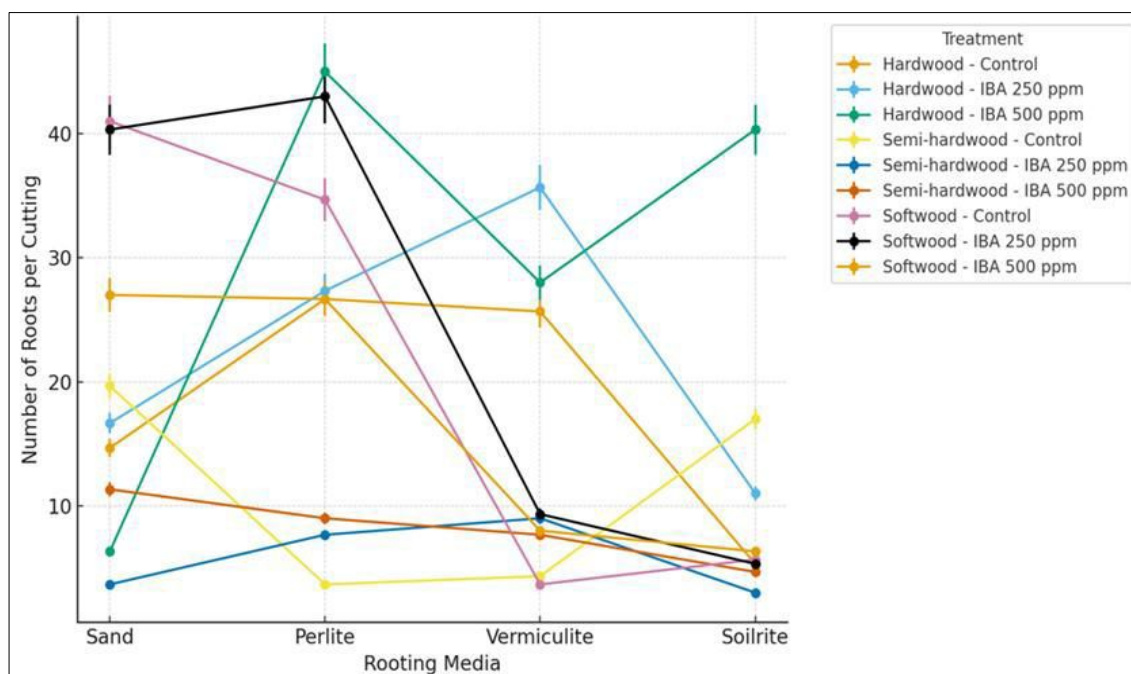


Fig 1: Effect of cutting type, IBA concentration, and rooting media on the number of roots in *Hydrangea macrophylla* cuttings at 75 days after planting (DAP). Values are means with standard error bars ($\pm$ SE).

3.2. Length of the Longest Root (cm)

Root length followed trends similar to root number. Softwood cuttings again outperformed others, producing the longest roots (12.64 cm), followed by semi-hardwood (9.80 cm) and hardwood cuttings (8.29 cm). The superiority of softwood cuttings is likely due to their higher endogenous auxin levels and better responsiveness to exogenous hormones (Costa *et al.*, 2002) [12].

Table 2 indicated that IBA at 250 ppm led to the longest roots (13.14 cm), significantly better than both 500 ppm (10.26 cm) and control (7.33 cm). Excessive IBA (500 ppm) possibly resulted in growth inhibition or callus formation, reducing elongation (Pimeta *et al.*, 2003) [12]. Among media, perlite and sand promoted the longest roots (13.45 cm and

12.25 cm, respectively), while soilrite showed the shortest (6.92 cm). The coarse structure of perlite and sand likely facilitates easier root penetration and oxygen availability, essential for elongation. Figure 2, shows the Rooted cuttings respectively hardwood cuttings, semi hardwood cuttings and softwood cuttings after treatment with IBA and different media types. Figure 3 shows General view of *Hydrangea macrophylla* potted cuttings.

The best treatment combination for root length was again softwood cuttings $\times$ 250 ppm IBA $\times$ perlite, with an average of 15.27 cm, followed by softwood $\times$ 250 ppm IBA $\times$ sand (14.53 cm). These combinations clearly demonstrate the synergistic effect of optimal cutting maturity, auxin concentration, and rooting medium.

Table 2: Effect of types of cutting, growth regulator and media on length of the roots (cm) at 75 days after planting

Type of cuttings	IBA Concentration (ppm)	Media				Mean	Grand mean
		Sand	Perlite	Vermiculite	Soilrite		
Softwood cuttings	IBA 500 ppm	1.57	0.73	0.50	0.67	0.86	
	IBA 250 ppm	1.13	0.60	0.53	0.90	0.79	
	IBA 0ppm (control)	1.33	0.60	0.27	0.27	0.61	
	Mean	1.34	0.64	0.43	0.61		0.75
Semi hardwood cuttings	IBA 500 ppm	0.67	0.33	0.33	0.30	0.40	
	IBA 250 ppm	0.17	0.40	0.43	0.37	0.34	
	IBA 0ppm (control)	0.63	0.20	0.43	0.50	0.44	
	Mean	0.49	0.31	0.39	0.39		0.39
Hardwood cuttings	IBA 500 ppm	0.77	0.93	0.70	0.70	0.77	
	IBA 250 ppm	0.40	1.53	0.73	0.27	0.73	
	IBA 0ppm (control)	1.20	0.67	0.83	0.53	0.80	
	Mean	0.79	1.04	0.75	0.5		0.77
IBA 500 ppm		1.00	0.67	0.51	0.56		0.67
IBA 250 ppm		0.57	0.84	0.57	0.51		0.62
IBA 0ppm (control)		1.06	0.49	0.51	0.43		0.61
Grand Mean		0.87	0.67	0.53	0.50		
Effects				'F' Test		SE(M)±	CD at 5%
Types of cuttings				**		0.06	0.18
Growth regulator concentration (ppm)				NS		-	-
Media				**		0.08	0.21
Interaction							
Cuttings × Growth regulator conc.				NS		-	-
Cutting × Media				**		0.13	0.37
Growth regulator conc. × Media				*		0.13	0.37
Cutting × Growth regulator conc. × Media				NS		-	-

* = Significant at 5% level. ** = Significant at 1% level.



Fig 2: Rooted cuttings respectively hardwood cuttings, semi hardwood cuttings and softwood cuttings.



Fig 3: General view of *Hydrangea macrophylla* potted cuttings at flowering stage, showing differences among softwood, semi-hardwood, and hardwood cuttings.

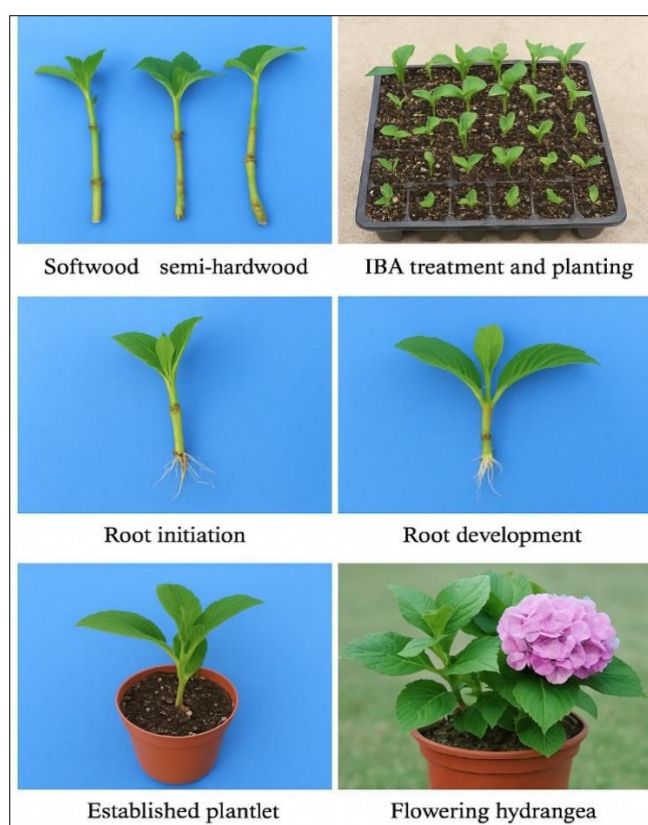


Fig 4: Propagation stages of *Hydrangea macrophylla* from cuttings to flowering

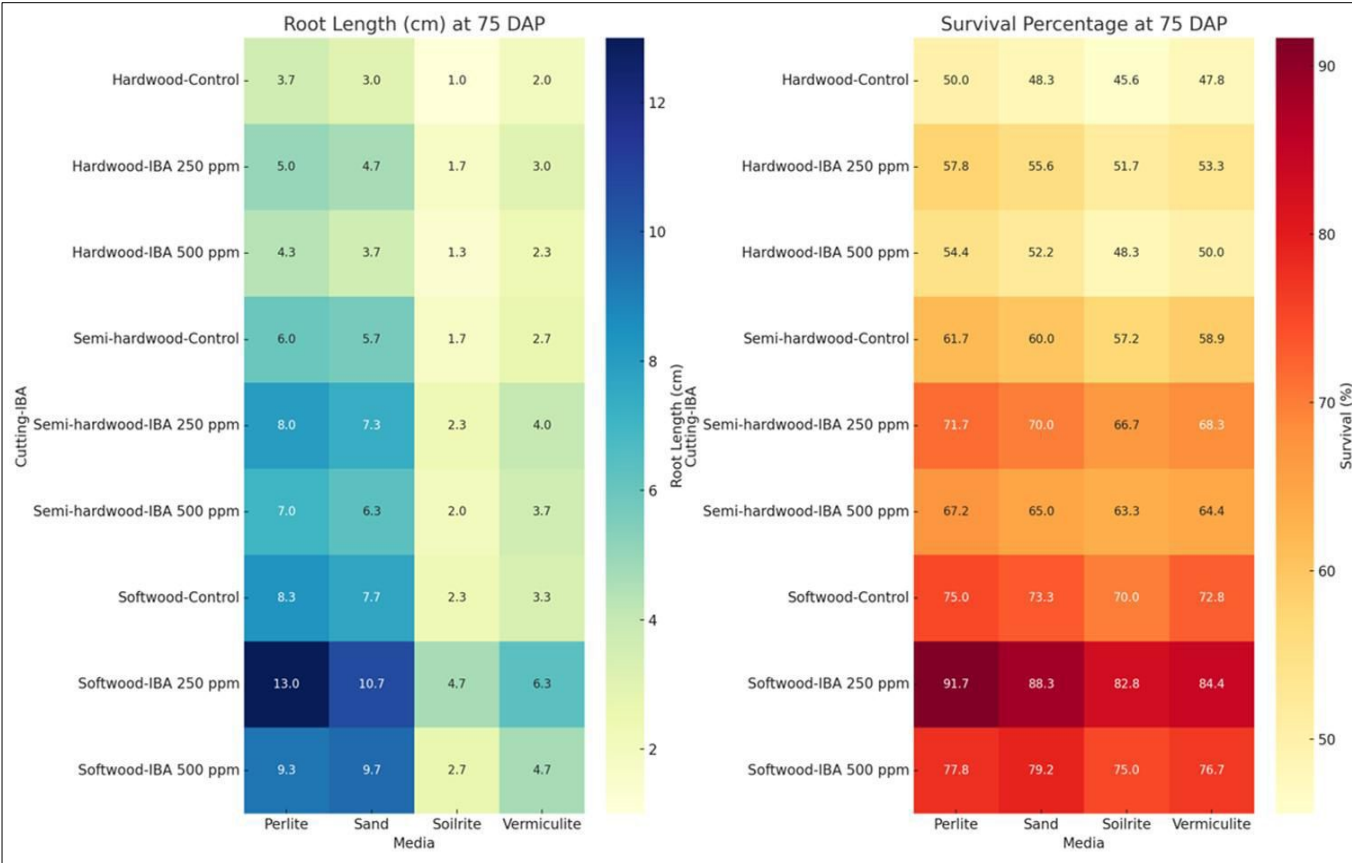


Fig 5: Heatmap showing the effect of cutting type, IBA concentration, and rooting media on root length (cm) and survival percentage of *Hydrangea macrophylla* cuttings at 75 days after planting (DAP).

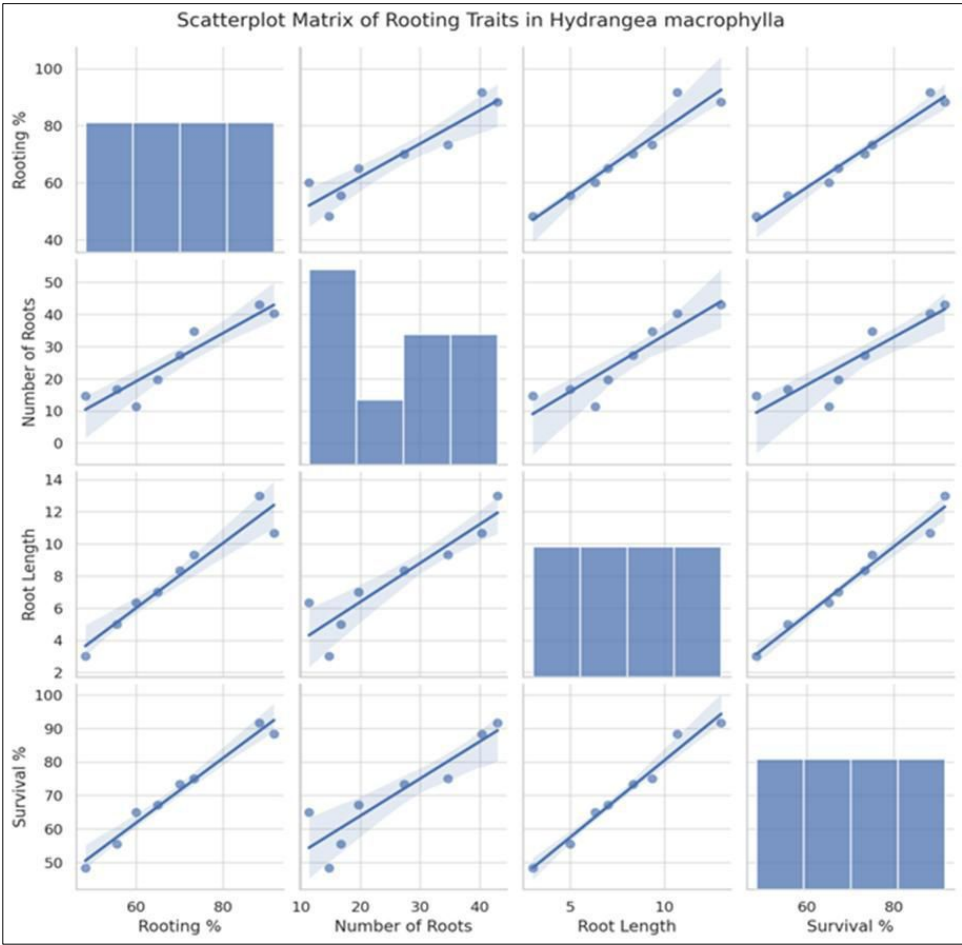


Fig 6: Scatterplot matrix of rooting traits in *Hydrangea macrophylla*.

The sequential stages of *Hydrangea macrophylla* propagation, from cutting preparation through IBA treatment, rooting, establishment, and flowering, are illustrated in Figure 4. Root length and survival percentage of *Hydrangea macrophylla* cuttings were significantly influenced by cutting type, IBA concentration, and rooting media (Table 2). Softwood cuttings treated with 250 ppm IBA and rooted in perlite recorded the maximum root length (13.0 cm) and the highest survival percentage (91.67%), followed by softwood cuttings in sand with the same IBA treatment. In contrast, hardwood cuttings, particularly in soilrite and vermiculite, consistently produced the lowest values across all IBA concentrations. Among rooting media, perlite and sand significantly outperformed vermiculite and soilrite in promoting root elongation and overall survival. The interactive effect of cutting type and IBA level was also evident, with softwood cuttings responding more positively than semi-hardwood and hardwood cuttings at comparable IBA concentrations.

These trends are further illustrated in Fig.5, where heatmaps clearly depict the superior performance of softwood cuttings treated with 250 ppm IBA in perlite and sand. The color gradients highlight the marked differences in root length and survival percentage among treatment combinations, emphasizing the synergistic effect of cutting type, auxin application, and rooting medium on propagation success. Also, Correlation analysis among rooting traits revealed strong positive associations (Fig.6). Root length showed a close relationship with survival percentage, indicating that longer roots improved cutting establishment. Similarly, rooting percentage was positively correlated with both number of roots and survival, suggesting that treatments promoting early root initiation also enhanced overall plant performance. These trends emphasize the interdependence of rooting traits and support the superiority of softwood cuttings treated with 250 ppm IBA in perlite.

4. Discussion

The results of this study clearly demonstrate that rooting response in *Hydrangea macrophylla* is strongly influenced by the type of cutting, concentration of IBA, and rooting medium. Among the different treatments, softwood cuttings treated with 2000 ppm IBA and planted in a sand + FYM medium consistently recorded the highest rooting percentage, root length, and survival rate, highlighting the interactive effect of hormonal stimulation and favorable substrate conditions.

The superiority of softwood cuttings observed in this study is consistent with the findings of Costa *et al.* (2002)^[2] and Luz *et al.* (2007)^[7], who reported that juvenile tissues have higher metabolic activity and greater rooting potential than semi-hardwood or hardwood cuttings. Softwood cuttings generally possess higher levels of endogenous auxins and carbohydrates, which contribute to enhanced root initiation and elongation (Leakey, 2004)^[6].

Exogenous application of IBA significantly improved rooting success across all cutting types, with the best performance observed at 2000 ppm. This aligns with previous reports in ornamentals such as chrysanthemum and rose, where IBA promoted adventitious root initiation by stimulating cell division and differentiation at the basal region of cuttings. While lower concentrations of IBA enhanced rooting moderately, excessively high levels often result in callus formation or inhibition of root elongation,

which may explain the reduced performance at 3000 ppm observed in the present study.

The choice of rooting medium also played a pivotal role in propagation success. The sand + FYM mixture provided better aeration, moisture balance, and microbial activity, resulting in significantly higher rooting compared to soilrite or pure sand. Similar results were reported by Paul *et al.* (1992)^[11] and Luz *et al.* (2007)^[7], who emphasized that substrates with optimum porosity and water-holding capacity create a favorable microenvironment for root induction. Organic components such as FYM also contribute beneficial microorganisms and nutrients, further supporting root growth.

The combined effect of IBA and suitable rooting medium in this study confirms the importance of optimizing both hormonal and physical factors for successful propagation. Earlier studies on other ornamentals, including chrysanthemum, hibiscus, and rose, have also demonstrated the synergistic effect of growth regulators and substrate selection (Maharana *et al.*, 2025; Jena *et al.*, 2025)^[8, 3]. However, limited studies have been conducted on *Hydrangea macrophylla*, particularly under subtropical climatic conditions. The present findings therefore provide novel insights into developing propagation protocols for this species in regions such as Odisha, where climatic variability can affect rooting efficiency.

In summary, this study establishes that softwood cuttings treated with 2000 ppm IBA and rooted in sand + FYM medium represent the most effective combination for maximizing rooting success in *Hydrangea macrophylla*. The results corroborate previous research on the role of auxins and rooting substrates while adding new evidence specific to subtropical environments. These findings can serve as a practical guide for nurserymen and horticulturists aiming to propagate *Hydrangea macrophylla* efficiently and sustainably.

These findings clearly demonstrate that softwood cuttings, when treated with 250 ppm IBA and rooted in perlite or sand, produce superior root development in *Hydrangea macrophylla*. Both root number and length were significantly enhanced under these conditions. The results support previous reports emphasizing the role of cutting juvenility, auxin application, and porous media in successful vegetative propagation (Luz *et al.*, 2007; Bhattacharjee *et al.*, 1986; Mostafa, 1996; Jena *et al.*, 2025)^[10, 7, 4].

5. Conclusion

The present study demonstrated that rooting success in *Hydrangea macrophylla* is significantly influenced by cutting type, IBA concentration, and rooting medium under subtropical conditions. Softwood cuttings treated with 2000 ppm IBA and rooted in a sand + FYM mixture consistently produced the highest rooting percentage, root length, and survival rate. This combination proved superior to semi-hardwood and hardwood cuttings and also outperformed other substrate and hormonal treatments.

These findings highlight the synergistic role of physiological maturity, exogenous auxin application, and favorable substrate conditions in improving vegetative propagation. The results corroborate earlier research on the benefits of IBA and well-aerated media but also add novel insights specific to *Hydrangea macrophylla* grown in subtropical climates, where limited studies have been conducted.

In practical terms, the identified treatment combination can serve as a reliable propagation protocol for nurserymen and horticulturists aiming to produce quality planting material of *Hydrangea macrophylla*. Future research may focus on integrating biostimulants, bioenzymes, or nanoparticle-based treatments to further enhance rooting efficiency and reduce reliance on synthetic auxins.

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