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Cloche duration and auxin-based regulators using standardised indole-3-acetic acid for growth dynamics of rose (*Rosa chinensis*) cuttings

Dita Anjelita^a, Indra Purnama^{b,c,*}, Seprita Lidar^a, Anisa Mutamima^d, Yuliana Susanti^b

^aDepartment of Agrotechnology, Universitas Lancang Kuning, Indonesia ^bGraduate School of Agricultural Sciences, Universitas Lancang Kuning, Indonesia ^cCenter for Sustainable Tropical Agricultural Research, Universitas Lancang Kuning, Indonesia ^dDepartment of Chemical Engineering, Universitas Riau, Indonesia

Abstract

Roses (*Rosa chinensis*) are widely favoured ornamental plants due to their captivating blooms and vibrant colours. Propagation through stem cuttings allows for the replication of superior traits from parent plants but often faces challenges due to suboptimal environmental conditions. This study aimed to evaluate the effects of cloche duration and types of auxin-based plant growth regulators (PGRs) on the growth dynamics of rose stem cuttings. A factorial completely randomised design (CRD) was employed with two factors: cloche duration (P) and auxin-based PGRs (A), each with three replications. Cloche duration (P) included: P1: 2 weeks, P2: 4 weeks, and P3: 6 weeks, while auxin-based PGRs (A) included: A1: No PGR, A2: commercial PGR containing 6 mg naphthaleneacetic acid (NAA)/10 mL, A3: mung bean sprouts extract containing 6 mg indole-3-acetic acid (IAA)/10 mL, and A4: garlic extract containing 6 mg IAA/10 mL. The experiment involved 36 plots, each with three plants, totalling 108 plants. Results indicated that the interaction between cloche duration and auxin-based PGRs had no significant effect on the observed parameters, which included bud sprouting time, survival rate, plant height, number of branches, number of flowers, and root volume. However, the individual factors - cloche duration and auxin-based PGRs showed significant effects on several of these parameters. A cloche duration of 6 weeks (P3) significantly improved survival rate and root volume, while a duration of 2 weeks (P1) promoted plant height. Among the PGR treatments, commercial PGR (A2) significantly enhanced plant height, whereas garlic extract (A4) increased branch numbers. These findings highlight the potential of optimizing cloche duration and natural auxin-based treatments to improve propagation efficiency in roses.

Keywords: auxin-based treatments, cloche application, natural plant growth enhancers, root and shoot development, stem cutting propagation

1 Introduction

Roses (*Rosa chinensis*) are among the most iconic ornamental plants, cherished globally for their captivating blooms, vibrant colours, and cultural symbolism in various traditions (Hibrand Saint-Oyant *et al.*, 2018). As a highdemand crop in the floriculture industry, roses are extensively propagated to meet market needs. Stem cutting is a widely used propagation method as it preserves the superior genetic traits of parent plants. However, successful propagation of rose cuttings depends on multiple factors, including environmental conditions, physiological readiness of the cuttings, and the application of growth stimulants (Ibrahim *et al.*, 2017; Morales-Orellana *et al.*, 2022; Otiende *et al.*, 2021).

Despite its advantages, propagation through stem cuttings often faces significant challenges, such as delayed bud sprouting, low survival rates, poor root development, and stunted growth. Additionally, achieving optimal shoot growth, branch formation, and flower production is often

^{*} Corresponding author: indra.purnama@unilak.ac.id

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hindered by suboptimal environmental conditions, including fluctuations in humidity and temperature (Braun & Wyse, 2019; Shin *et al.*, 2023;). To address these issues, protective cloches, or plant covers, are used to create a controlled microenvironment, enhancing rooting and growth dynamics. Cloches help to maintain consistent humidity and temperature levels, reducing water loss from cuttings and improving survival rates (Pandey *et al.*, 2023). For example, Rinanto *et al.* (2022) reported that cloche application significantly improved rooting percentage and shoot development in Rosa sp. cuttings. However, determining the optimal cloche duration remains crucial, as excessive humidity may lead to fungal infections, while insufficient coverage may fail to protect cuttings from environmental stress (Hernandez & Martinez, 2018).

In addition to environmental management, the use of plant growth regulators (PGRs) is integral to promoting root formation, shoot elongation, and overall plant development. Auxins, as a class of PGRs, are particularly effective in stimulating cell division and elongation, thereby enhancing rooting success (Mutryarny *et al.*, 2022; Traversari *et al.*, 2022). Synthetic auxins, such as naphthaleneacetic acid (NAA) and indole-3-butyric acid (IBA), have shown significant efficacy in promoting rooting and growth of cuttings in two rose cultivars (Susaj *et al.*, 2012).

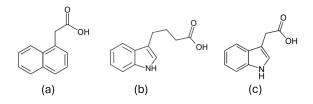


Fig. 1: Structural representation of synthetic: (a) naphthaleneacetic acid (NAA), (b) indole-3-butyric acid (IBA), and natural auxins: (c) indole-3-acetic acid (IAA).

However, growing concerns over the environmental impact and cost of synthetic PGRs have led to increased interest in natural auxin sources, such as garlic (*Allium sativum*) and mung bean sprouts (*Vigna radiata*), which contain measurable levels of indole-3-acetic acid (IAA) (Atif *et al.*, 2021; Windarto *et al.*, 2023). Garlic have been reported to contain approximately 1.47 ng IAA/g, along with other phytohormones such as gibberellins and abscisic acids, contributing to their effectiveness as plant growth stimulants (Atif *et al.*, 2021). Another study indicated that IAA levels in mung bean sprouts could reach 27.3 %, depending on extraction methods and growth conditions (Cai *et al.*, 2023). Previous studies have demonstrated the effectiveness of natural auxins in enhancing root volume, shoot growth, and branch formation in ornamental plants (Abdel-Rahman *et al.*, 2020). Fig. 1 illustrates the chemical structures of synthetic auxins, including (a) NAA and (b) IBA, alongside the natural auxin (c) IAA.

While the effects of cloche application and auxin treatments on plant propagation are well-documented, there remains a lack of comprehensive studies evaluating their combined impact on rose stem cuttings. Furthermore, limited research exists on the comparative effectiveness of synthetic versus natural auxins, particularly in promoting parameters such as survival rate, bud sprouting time, root volume, and branch or flower production. In addition, the optimal cloche duration to maximise growth and survival across these parameters in *R. chinensis* cuttings remains unclear.

This study aimed to investigate the combined effects of cloche duration and auxin-based treatments on the propagation of *R. chinensis* stem cuttings. Specifically, their impact on bud sprouting time, survival rate, plant height, flower and branch production, and root volume, was evaluated with the aim of identifying the most effective cloche duration and auxin type – synthetic or natura 1 – for optimising rose propagation.

2 Materials and methods

2.1 Study design and experimental setup

The experiment was conducted to evaluate the effects of cloche duration and auxin-based treatments on the propagation of *R. chinensis* stem cuttings. A factorial completely randomised design (CRD) was used, consisting of two factors: cloche duration and auxin-based treatments. Cloche duration (P) included three levels: 2 weeks (P1), 4 weeks (P2), and 6 weeks (P3). Auxin-based treatments (A) consisted of four levels: no PGR (A1), a commercial PGR (Growtone 3.75 SP, Deltagro Inc., Indonesia) containing 6 mg NAA/10 mL (A2), mung bean sprout extract containing 6 mg IAA/10 mL (A3), and garlic extract containing 6 mg IAA/10 mL (A4). The experimental setup, including the treatment application process and the cloche duration arrangement, is illustrated in Fig. 2 to provide a clearer visualisation of the methodology.



Fig. 2: Experimental setup illustrating auxin-based treatments and cloche duration for R. chinensis stem cuttings.

There was no negative control for cloche duration, as preliminary observations indicated that *R. chinensis* stem cuttings could not survive without cloche protection. Each treatment combination was replicated three times, resulting in 36 experimental units. Each unit contained three cuttings for a total of 108 plants.

2.2 Environmental conditions

The experiment was conducted in an open-field with an average temperature of 32.8 ± 2 °C, relative humidity between 70% and 84%, and a natural photoperiod of approximately 12 h per day. The conditions were monitored using a digital thermo-hygrometer, ensuring consistency throughout the study.

2.3 Preparation of plant materials

Healthy parent plants of *R. chinensis*, aged 6 months, were selected as the source of stem cuttings. These plants were not registered commercial cultivars, but represent typical local types widely cultivated in home gardens in the region. The cuttings were prepared to a uniform length of 10 cm with 2 nodes. Before planting, the basal ends of the cuttings were treated by immersing them in their respective 10 mL PGR solutions for 30 min to ensure absorption.

2.4 Cloche application

Cloches were constructed using transparent polyethylene sheets supported by wire frames. After planting, the cuttings were immediately covered with cloches, which were removed based on the assigned durations: 2 weeks (P1), 4 weeks (P2), or 6 weeks (P3). Cloches provided a stable environment with high humidity and moderate temperature, which are crucial for successful rooting (Jha *et al.*, 2019; Pandey *et al.*, 2023). The methodology for cloche construction and application followed the guidelines described by Rinanto *et al.* (2022) with some modifications.

2.5 Preparation of auxin-based treatments

The auxin-based treatments in this study included both synthetic and natural sources. The commercial PGR contained 3 % NAA and was recommended to be used at a dose of 20 g L⁻¹, a concentration that has been widely utilised in previous studies for optimising root initiation and shoot elongation in vegetative propagation (Nafery *et al.*, 2022). When prepared according to this recommendation, the resulting solution contains 6 mg of NAA per 10 mL. Natural auxin-based treatments were prepared using mung bean sprouts (*Vigna radiata*) and garlic cloves (*Allium sativum*). Preliminary steps involved blending 100 g of each material without the addition of distilled water. The resulting volumes of extract were measured to determine the extraction efficiency, where mung bean sprouts produced 32.31 mL of extract, while garlic cloves produced 22.98 mL.

The concentration of IAA in each extract was quantified using high-performance liquid chromatography (HPLC). The HPLC analysis confirmed IAA concentrations of 20.52 mg L⁻¹ for mung bean sprout extract and 22.37 mg L⁻¹ for garlic extract. Using these values, the amount of raw material required to produce an extract equivalent to 6 mg IAA per 10 mL was calculated. The calculations determined that 905 g of mung bean sprouts and 1167 g of garlic cloves were required to prepare the respective extracts at the desired concentration.

For the preparation of natural auxin treatments, 905 g of mung bean sprouts and 1167 g of garlic cloves were separately blended without the addition of water. The resulting extracts were filtered to remove solids and standardised to ensure a uniform auxin concentration of 6 mg IAA per 10 mL. Unlike conventional methods, which typically rely on the percentage yield of extracts (Abbasifar *et al.*, 2020; Aryan *et al.*, 2023; Dewi & Sabhara, 2022; Rashedy, 2022; Windarto *et al.*, 2023;), this approach was specifically developed to match the natural auxin content with the synthetic auxin concentration. By standardising the primary bioactive component, such as natural auxins, with synthetic auxins, this method enables more accurate and meaningful comparisons.

2.6 Planting and maintenance

Cuttings were planted in polybags filled with a mixture of sandy loam soil and organic compost (2:1 ratio). Watering was performed daily to maintain consistent soil moisture, and weeds were manually removed as necessary. Fertilisation was carried out using NPK 15-30-15 fertiliser (Agrimore-P, Agricon Inc., Indonesia) at a dose of 2 gL^{-1} of water. Fertiliser application began at 45 days after planting and was repeated weekly throughout the study. Pest and disease control were conducted using both insecticides and fungicides. For pest control, lambda-cyhalothrin (Matador[®] 2.5EC, Syngenta, Switzerland) was applied at a dose of 2 mL L^{-1} of water. Fungicide applications were performed using mancozeb (Dithane® M-45, Corteva Agriscience, USA) at a dose of 3 gL^{-1} of water. The first fungicide application was carried out by spraying the growing medium before planting, while subsequent applications were conducted biweekly after cloche removal by spraying the entire plant. All pesticide applications followed recommended dosages to minimise the risk of pesticide residues on rose tissues and prevent environmental contamination. This approach is in line with previous findings by Malhat *et al.* (2023) and Purnama *et al.* (2025), which highlight the importance of adhering to label-recommended doses to ensure both crop safety and ecological sustainability.

2.7 Data collection

Six parameters were recorded to assess the performance of stem cuttings. Time to bud sprouting was recorded as the number of days from planting until the first visible sprout appeared. Survival rate was calculated as the percentage of viable cuttings out of the total number planted. Plant height was measured at 90 days after planting, from the soil surface to the tip of the tallest shoot, using a standard measuring tape. The number of flowers and branches produced per cutting was manually counted. Root volume was measured using the water displacement method after cleaning the roots to remove soil debris, following the methodology outlined by Hosseini *et al.* (2017).

2.8 Statistical analysis

All data were analysed using analysis of variance (AN-OVA) to evaluate the effects of cloche duration, auxin-based treatments, and their interactions. Significant differences among treatment means were identified using Duncan's Multiple Range Test (DMRT) at a 5 % significance level. Statistical analyses were performed using SPSS soft

3 Results

3.1 Bud sprouting time

Bud sprouting time, defined as the number of days from planting until the first visible bud appeared, is a critical indicator of the initial establishment of *R. chinensis* cuttings. Although the interaction between cloche duration and auxin

Table 1: Bud sprouting time (days) of R. chinensis stem cuttings under different cloche durations and auxin-based treatments (n = 3).

Cloche duration	A1	A2	A3	A4	Mean
P1 (2 weeks)	19.67	14.67	15.00	22.83	18.04
P2 (4 weeks)	12.67	18.67	20.17	18.17	17.41
P3 (6 weeks)	18.00	22.50	16.67	10.33	16.87
Mean	16.78	18.61	17.28	17.11	

A1: no plant growth regulators; A2: growtone; A3: mung bean sprout extract; A4: garlic extract.

treatments was not statistically significant, a slight trend was observed where longer cloche durations generally supported faster bud sprouting when combined with natural auxins (Table 1).

3.2 Survival rate

Cloche duration had a significant impact on the survival rate of R. chinensis cuttings, while auxin-based treatments and their interaction with cloche duration did not show statistically significant differences. The survival rate ranged widely across treatments, reflecting the combined influence of environmental variability and experimental conditions. The 6-week cloche duration (P3) produced the highest survival rate, averaging 80.56%, significantly higher than the 2-week (P1) and 4-week (P2) durations, which averaged 36.10% and 41.66%, respectively (see Table 2). The extended cloche duration effectively provided a more stable microenvironment under the open-field conditions, characterised by an average temperature of 32.8 ± 2 °C and relative humidity of 70-84 %. These environmental factors are known to exacerbate water loss and stress in unprotected cuttings, particularly in shorter cloche durations (P1 and P2). Despite the lack of a statistically significant interaction, a

Table 2: Survival rate (%) of R. chinensis stem cuttings under different cloche durations and auxin-based treatments (n = 3).

Cloche duration	A1	A2	A3	A4	Mean
P1 (2 weeks)	33.33	33.33	33.33	44.44	36.10 ^a
P2 (4 weeks)	44.44	44.44	33.33	44.44	41.66 ^a
P3 (6 weeks)	66.67	88.89	66.67	100.00	80.56^{b}
Mean	48.14	55.56	44.44	62.97	

A1: no plant growth regulators; A2: growtone; A3: mung bean sprout extract; A4: garlic extract. Same letters in the same column indicate no significant differences at the 5 % level.

noticeable trend emerged where extended cloche durations paired with garlic extract (A4) tended to yield higher survival rates, as shown in Table 2.

3.3 Plant height and number of branches

Plant height and the number of branches are key indicators of vegetative growth in *R. chinensis* cuttings. These parameters reflect the interplay between cloche duration and auxin-based treatments in regulating both vertical and lateral growth. While cloche duration significantly influenced plant height, the number of branches was primarily affected by auxin treatments. Interestingly, their interaction did not produce statistically significant effects, suggesting independent roles for these factors in shaping growth outcomes (Tables 3 and 4). While the interaction between cloche duration and auxin treatments was not statistically significant, observable trends suggest that a 2-week cloche duration (P1) generally supported greater plant height, particularly when combined with synthetic auxin (A2, 37.67 cm) and garlic extract (A4,

Table 3: *Plant height (cm) of* R. chinensis *stem cuttings under different cloche durations and auxin-based treatments (n = 3).*

Cloche duration	A1	A2	A3	A4	Mean
P1 (2 weeks)	30.50	37.67	26.00	35.33	32.38 ^b
P2 (4 weeks)	25.67	30.33	18.17	27.17	25.33 ^a
P3 (6 weeks)	32.00	26.67	27.50	28.67	28.70 ^{ab}
Mean	29.39	31.56	23.89	30.39	

A1: no plant growth regulators; A2: growtone; A3: mung bean sprout extract; A4: garlic extract. Same letters in the same column indicate no significant differences at the 5 % level.

Table 4: *Number of branches of* R. chinensis *stem cuttings under different cloche durations and auxin-based treatments* (n = 3).

Cloche duration	A1	A2	A3	A4	Mean
P1 (2 weeks)	7.33	6.67	6.67	8.50	7.29
P2 (4 weeks)	7.00	6.00	6.67	7.50	6.79
P3 (6 weeks)	6.83	6.67	6.00	7.00	6.62
Mean (A)	7.06	6.44	6.44	7.67	

A1: no plant growth regulators; A2: growtone; A3: mung bean sprout extract; A4: garlic extract.

35.33 cm). This may indicate that earlier exposure to external conditions promotes shoot elongation through adaptive responses. In terms of branching, garlic extract (A4) yielded the highest number of branches overall (7.67), with the most notable effects under shorter cloche durations (P1A4 and P2A4). These patterns imply that garlic extract effectively stimulates lateral bud activation, but its benefits may be reduced under prolonged cloche conditions due to delayed acclimatisation.

3.4 Number of flower

Flower production is a crucial parameter for evaluating the ornamental value and reproductive success of R. chinensis cuttings. In this study, the number of flowers was influenced significantly by auxin-based treatments, while cloche duration showed no significant effect. This suggests that hormonal regulation played a more dominant role in enhancing flowering compared to environmental modifications (see Table 5). Although no significant differences were observed among cloche durations, certain trends were evident. The 6-week cloche duration (P3) produced the highest average number of flowers (7.00 flowers per cutting), slightly surpassing the 4-week (P2) and 2-week (P1) durations, which averaged 6.75 and 6.54 flowers, respectively. Cuttings treated with garlic extract (A4) exhibited earlier floral emergence compared to other treatments, although this parameter was not systematically recorded. The earlier flowering trend in garlic extract-treated plants suggests a potential role of

Table 5: *Number of flowers of* R. chinensis *stem cuttings under different cloche durations and auxin-based treatments* (n = 3).

Cloche duration	A1	A2	A3	A4	Mean
P1 (2 weeks)	6.00	6.50	6.33	7.33	6.54
P2 (4 weeks)	6.50	7.00	5.33	8.17	6.75
P3 (6 weeks)	6.67	5.67	8.00	7.67	7.00
Mean	6.39	6.39	6.56	7.72	

A1: no plant growth regulators; A2: growtone; A3: mung bean sprout extract; A4: garlic extract.

natural auxins in accelerating the transition from vegetative to reproductive growth.

Table 6: Root volume (mL) of R. chinensis stem cuttings under different cloche durations and auxin-based treatment (n = 3).

Cloche duration	A1	A2	A3	A4	Mean
P1 (2 weeks)	2.00	2.00	2.17	2.50	2.16 ^{ab}
P2 (4 weeks)	2.00	1.00	1.00	2.17	1.67 ^a
P3 (6 weeks)	2.09	2.67	2.50	4.10	2.86^{b}
Mean	2.02	1.89	2.06	2.94	

A1: no plant growth regulators; A2: growtone; A3: mung bean sprout extract; A4: garlic extract. Same letters in the same column indicate no significant differences at the 5 % level.

3.5 Root volume

A well-developed root system is fundamental for the successful establishment of *R. chinensis* cuttings, as it supports water and nutrient absorption critical for sustained growth. The study revealed that cloche duration played a significant role in determining root volume, while the effects of auxinbased treatments were less pronounced. No significant interaction between cloche duration and auxin treatments was observed (Table 6). Despite the absence of a statistically significant interaction, the combination of a 6-week cloche duration with garlic extract (A4) showed a trend toward increased root volume. This may be attributed to the prolonged high-humidity conditions enhancing auxin uptake efficiency and promoting root system expansion. Further studies could explore the underlying physiological adaptations that contribute to these patterns.

4 Discussion

4.1 Bud sprouting time

The shortest bud sprouting time was recorded under the 6week cloche duration (P3, 16.87 days), while the longest was observed in the 2-week treatment (P1, 18.04 days). These results indicate that extended cloche use plays a key role in creating a favourable microenvironment for early shoot emergence, especially in open-field settings where temperature and humidity fluctuate considerably. Such protective conditions likely reduce water loss and physiological stress, thus accelerating bud development. This finding is consistent with Rinanto *et al.* (2022), who reported the effectiveness of cloches in stabilising microclimates and supporting vegetative growth in ornamental species.

The open-field conditions in this study, with average temperatures of 32.8 ± 2 °C and relative humidity ranging from 70–84 %, imposed environmental challenges on unprotected cuttings. Longer cloche coverage likely buffered the cuttings from harsh conditions such as desiccating wind and direct sunlight, promoting hormonal activity that facilitates sprouting.

In terms of auxin treatments, cuttings without PGRs (A1) sprouted fastest (16.78 days), followed by garlic extract (A4, 17.11 days) and mung bean sprout extract (A3, 17.28 days). The slowest sprouting was observed in cuttings treated with synthetic auxin (Growtone, A2, 18.61 days), which may have delayed shoot initiation by promoting early root formation. This reflects the distinct physiological responses triggered by synthetic versus natural auxins. Natural extracts, which contain IAA and other bioactive compounds, may stimulate coordinated root-shoot growth and improve adaptation to fluctuating conditions (Mandal *et al.*, 2023; Sani & Yong, 2021).

These findings reinforce the importance of combining prolonged cloche duration with natural auxin treatments to enhance propagation success under open-environment conditions. Promoting early bud emergence using low-input, ecofriendly strategies such as cloches and plant-derived PGRs offers a promising approach for sustainable rose cultivation in tropical climates.

4.2 Survival rate

The 6-week cloche duration (P3) significantly improved cutting survival by reducing environmental stress through a more stable, humid microenvironment. This protection from high temperatures and desiccating winds likely facilitated physiological processes critical for plant establishment. Similar findings were reported by Martins *et al.* (2015), who emphasised the importance of maintaining consistent environmental conditions to ensure successful propagation in outdoor settings.

While differences among auxin treatments were not statistically significant, trends suggest that garlic extract (A4) resulted in the highest survival rate (62.97%), followed by Growtone (A2, 55.56%). The control (A1) and mung bean sprout extract (A3) showed lower survival rates, at 48.14% and 44.44 % respectively. The superior performance of garlic extract may be linked to its bioactive compounds that support stress tolerance and root formation under fluctuating conditions (Cheng *et al.*, 2016; Hayat *et al.*, 2018).

The performance of natural auxins (A3 and A4), particularly under prolonged cloche duration, indicates their potential as viable alternatives to synthetic auxins in open-field propagation. Although mung bean extract (A3) was less effective than garlic extract, the difference may reflect variability in auxin content or extraction methods. The highest survival rate (100%) was achieved in the P3A4 treatment, demonstrating the synergistic effect of environmental buffering through cloche application and the physiological support of natural auxins. These results support the integration of prolonged cloche use and natural plant growth regulators to enhance the resilience and survival of cuttings under challenging field conditions.

4.3 Plant height and number of branches

The 2-week cloche duration (P1) resulted in the tallest plants (32.38 cm) and the highest number of branches (7.29), suggesting that early exposure to environmental cues may trigger adaptive responses such as shoot elongation and lateral bud activation. These findings are consistent with Sánchez-Blanco *et al.* (2019), who reported that moderate environmental stress can stimulate both vertical and lateral growth in ornamental species. Conversely, the 4-week cloche (P2) produced the shortest plants (25.33 cm) and fewer branches (6.79), possibly due to delayed acclimatisation. The 6-week treatment (P3) showed intermediate values, which supports the idea that prolonged humidity stabilisation leads to more controlled, yet slower, growth (Rinanto *et al.*, 2022).

Auxin treatments had a significant effect on both parameters. Growtone (A2) promoted the greatest height (31.56 cm), consistent with its known role in enhancing cell elongation and division (Abdel-Rahman et al., 2020; Susaj et al., 2012; Yusnita et al., 2017). Garlic extract (A4) followed closely (30.39 cm), confirming its potential as a natural auxin source. Mung bean extract (A3) produced the shortest plants (23.89 cm), likely due to lower IAA content. For branching, garlic extract (A4) was the most effective (7.67 branches), attributed to its sulfur-based compounds and auxin-like activity (Cheng et al., 2016). Interestingly, the control (A1) also showed relatively high branching (7.06), indicating that R. chinensis may naturally exhibit strong lateral growth under suitable field conditions. Both A2 and A3 resulted in fewer branches (6.44), suggesting a physiological trade-off between vertical and lateral development.

Notably, the combination of P1 with A2 produced the tallest plants, while P1 with A4 yielded the highest number of branches. These synergistic effects indicate that moderate cloche duration, when paired with either synthetic or natural auxins, can support both vegetative parameters effectively under open-field propagation. This interaction reflects the complementary roles of environmental modulation and growth regulators in plant morphogenesis. The data reinforce the potential of garlic extract as a natural auxin for improving structural growth traits, particularly when paired with shorter cloche durations that promote early acclimation.

4.4 Number of flower

Prolonged cloche duration contributed to a stable microenvironment conducive to flowering by maintaining consistent humidity and temperature. However, the absence of significant differences among cloche durations suggests that hormonal regulation may have had a stronger influence on flower development. Rinanto *et al.* (2022) previously reported that while cloches stabilise reproductive growth, their effect varies depending on species and environmental context. Among the auxin treatments, garlic extract (A4) pro-



Fig. 3: Visual comparison of R. chinensis flowers under different treatment combinations: P2A2 (left) and P3A4 (right).

duced the highest average flower number (7.72 per cutting), indicating its potential as a natural auxin enriched with compounds that enhance nutrient uptake, cell division, and floral initiation (Cheng *et al.*, 2016; Hayat *et al.*, 2018). Mung bean sprout extract (A3) yielded moderate results (6.56), while both the control (A1) and Growtone (A2) averaged 6.39 flowers. This similar performance between A1 and A2 suggests that Growtone may prioritise vegetative over reproductive growth, as noted by Jhonson *et al.* (2023).

The highest flower count (8.17) was observed in the P2A4 treatment (garlic extract with 4-week cloche duration), suggesting a synergistic effect between moderate environmental

stability and auxin-driven hormonal regulation. Renau-Morata *et al.* (2021) and Gomes & Scortecci (2021) support the idea that natural auxins influence gene expression related to flowering, enabling plants to respond adaptively to environmental cues. These effects are attributed to the activation of transcription factors involved in floral transition and plasticity, even at low auxin concentrations.

Visual comparisons (Figure 3) between P2A2 and P3A4 revealed more abundant and larger flowers in the P3A4 group. While garlic extract promoted higher flower quantity, minor variations in colour and size uniformity were noted compared to synthetic auxins, suggesting potential trade-offs between floral yield and quality. Such differences are consistent with the findings of Sun *et al.* (2021), Pan *et al.* (2022), and Wang *et al.* (2022), which link auxin distribution and gene regulation to petal size, expansion, pigmentation, and anthocyanin biosynthesis.

Garlic extract also appeared to accelerate flowering, as floral emergence occurred earlier than in other treatments, although this was not quantitatively measured. IAA within garlic is likely involved in this effect through its regulatory role in floral induction. Past studies show that auxin's influence on flowering is context-dependent, varying with concentration, species, and environmental conditions (Ionescu *et al.*, 2017). For instance, auxin promotes flowering in *Arabidopsis thaliana* via ARF1/ARF2, but can delay it in shortday species like *Xanthium*. Environmental variables such as heat and nitrogen availability also modulate auxin-related flowering responses, as seen in *Lactuca sativa* and *Brassica napus* (Hao *et al.*, 2022; Li *et al.*, 2022).

Taken together, these results underscore the prominent role of garlic extract in promoting floral induction in *R. chinensis*, both in terms of quantity and timing. Its combination with moderate cloche duration presents a promising strategy for optimising reproductive success in open-field propagation systems.

4.5 Root volume

The greatest root volume was achieved under the 6-week cloche duration (P3, 2.86 mL), indicating that extended protection enhances root development by maintaining humidity and reducing exposure to environmental stress. This supports previous findings by Rinanto *et al.* (2022), who noted that prolonged cloche use stabilises microclimates conducive to root system establishment. In contrast, the 4-week duration (P2) produced the lowest root volume (1.67 mL), likely due to insufficient environmental buffering. The 2-week duration (P1, 2.16 mL) supported early rooting but may have limited sustained development due to early exposure.

Garlic extract (A4) led to the highest root volume (2.94 mL), suggesting that its bioactive compounds, including sulphur metabolites and IAA, promote cellular expansion and root elongation (Cheng *et al.*, 2016; Hayat *et al.*, 2018). Mung bean sprout extract (A3) showed comparable performance (2.06 mL), while the control (A1, 2.02 mL) and synthetic auxin (A2, 1.89 mL) yielded lower values. This modest performance of Growtone may reflect a greater emphasis on shoot elongation rather than root enhancement, as previously reported by Traversari *et al.* (2022).

The highest root volume (4.10 mL) occurred in the P3A4 combination, confirming the synergistic effect of prolonged cloche protection and the physiological activity of garlic extract. These findings highlight the potential of integrating environmental buffering and natural auxin supplementation to maximise rooting success.

Beyond efficacy, natural auxins offer significant ecological benefits. Synthetic auxins such as NAA and IBA are chemically produced and may leave residues that exceed safety thresholds, contributing to environmental contamination (Le *et al.*, 2020; Abbasifar *et al.*, 2020). In contrast, plant-derived auxins are biodegradable and pose minimal risk to soil and water quality (Rashedy, 2022). Their use aligns with sustainable agriculture practices by reducing chemical input and supporting organic propagation systems.

Economically, natural auxins can be extracted from lowcost, renewable resources, such as agricultural byproducts, offering a cost-effective alternative for small-scale and commercial producers (Aryan *et al.*, 2023). This approach also aligns with circular bioeconomy principles by reducing waste and dependence on synthetic inputs (European Commission, 2017). While promising, the application of natural auxins warrants further optimisation. Future research should focus on improving extraction efficiency, ensuring formulation stability, and evaluating their performance under diverse field conditions. A deeper understanding of their physiological interaction with environmental factors could inform broader adoption in sustainable *R. chinensis* propagation.

5 Conclusions

This study highlights the importance of cloche duration and auxin-based treatments in optimizing the propagation success of *R. chinensis* stem cuttings. Prolonged cloche durations, particularly 6 weeks (P3), significantly improved survival rates, root volume, and overall plant establishment by maintaining a stable microenvironment. Among the auxin treatments, garlic extract (A4) demonstrated superior performance, confirming its potential as a sustainable alternative to synthetic PGRs. Although no significant interaction effects were observed, trends suggest that prolonged cloche durations combined with bioactive-rich natural auxins can enhance propagation outcomes. A key contribution of this study is the standardisation of natural IAA concentration (6 mg per 10 mL or 600 ppm) to ensure comparability with synthetic PGRs. This approach addresses the inconsistency in previous studies that relied on crude extracts without precise auxin quantification. Future research should explore additional natural auxin sources, such as coconut water, seaweed extract, or fermented plant extracts, while maintaining standardised IAA concentrations to enable direct comparisons. Additionally, validating these findings across different climates and plant species will further support the broader application of natural auxins in sustainable horticultural practices.

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Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

Author contributions

Dita Anjelita: methodology, investigation, data curation, writing – original draft preparation. Indra Purnama: conceptualization, supervision, funding acquisition, writing – review & editing, project administration. Seprita Lidar: supervision, validation, formal analysis, data curation. Anisa Mutamima: resources, visualization, writing – original draft preparation, data curation. Yuliana Susanti: formal analysis, methodology, resources, project administration.

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