Introduction

Apocynum lancifolium Russanov is a perennial herb and a subspecies of A. venetum belonging to the Apocynaceae family. Known as a halotolerant plant, A. lancifolium thrives in various environments, including coastal or reclaimed lands, deserts, and salt marshes (Son et al., 2011). In South Korea, it naturally grows along the western and southern coasts, and in the Gyeonggi-do region. Moreover, occasional inland distribution has been observed in Danyang-gun, Chungcheongbuk-do (Kim et al., 2021a). A. lancifolium is renowned for its medicinal properties, which include cardioprotective, antihypertensive, hepatoprotective, and antidepressant functions. Ongoing research has focused on its potential as a natural cosmetic ingredient in the aesthetics industry (Walternberger et al., 2016; Fan et al.,
Effect of Propagation Media and Plant Growth Regulators on Adventitious Rooting of Softwood Cuttings of *Apocynum lancifolium* Russanov

1999; Zheng et al., 2012). The Korea Forest Service currently designates *A. lancifolium* as an endangered species, thus, underscoring the importance of conserving this native plant of South Korea.

According to the initial report of the National Garden Promotion Plan by the Korea Forest Service (2016), the horticulture industry has steadily expanded, both domestically and internationally. Recently, South Korea has seen increased interest in horticulture and garden construction. With the implementation of the Nagoya Protocol, there is a growing need to protect biosovereignty that can reduce the reliance on foreign bioresources and mitigate royalty payments for imported horticultural species. Oh et al. (2021) highlighted that out of 4,000 native plants in South Korea, approximately only 600 exhibit significant development potential, with few currently in use. To realize the full potential of native plants, there must be sufficient production capacity to meet the increasing demands, thereby necessitating the development of advanced propagation techniques for mass production.

Among various methods employed for plant reproduction, cutting propagation promotes mass propagation without introducing genetic mutations. Thus, this method can be readily employed at farms or seed nurseries to accelerate the development of new cultivars (Oh et al., 2021). Several factors influence the rooting rate and root development during cutting propagation, including plant growth regulators (PGRs), timing of cuttings, propagation media, and humidity (Kim and Kim, 2012). While Kim et al. (2021) examined the effects of NaCl on seed germination in *A. lancifolium* during propagation, the cutting propagation method for this species has not yet been investigated. Research has shown that the seed germination rate of *A. lancifolium* is generally high; however, its mass production is challenging due to the relatively low survival rate of germinated seedlings. Additionally, as germination rates decline with reduced vitality during seed storage, limitations exist in terms of reproduction. Therefore, the status of seed germination for *A. lancifolium* suggests the need for more systematic and efficient research. Accordingly, this study aimed to investigate the impact of propagation media and PGRs (rooting agents) on the rooting of *A. lancifolium*, to establish a system for the vegetative reproduction of this native plant species.

### Research Methods

#### Scion collection and greenhouse conditions

To prepare scion samples, softwood from 2-year-old *A. lancifolium* plants was collected from the Sejong National Arboretum greenhouse in May 2023. To prevent water loss, the collected softwood was soaked in water for subsequent experiments, and to minimize transpiration, one of the two leaves on each scion was removed (Fig. 1). Scion lengths were standardized to 6-7 cm (2-3 nodes), and the base was cut diagonally. Scions were then buried in a propagation box (52 cm × 36.5 cm × 9 cm) to a depth of approximately 3 cm. The cutting propagation experiments were conducted in a solar-heated greenhouse equipped with an automatic fog sprayer to maintain constant air humidity. All scions were drip-irrigated once per day and the amount of water was approximately 1,000 ml per propagation box. A shading net is installed in the greenhouse to avoid direct sunlight falling on scions and maintain an average natural light intensity of approximately 115 μmol/s/m². Mean relative humidity and temperature in the greenhouse were maintained at 75 ± 5% and 25 ± 5°C, respectively.

#### Effects of propagation media on rooting

To assess the impact of propagation media on *A. lancifolium* rooting, the central portion of the scion was used, excluding the apical and basal regions of the stem. The propagation media included Kanuma soil as volcanic ash soil, commercial potting mixes, perlite, and peat moss (Table 1). Each medium was filled in a propagation box, and the cutting propagation was replicated four times, with 15 samples per treatment. The boxes were placed on their respective plates, and after six weeks, growth characteristics, including cutting survival rate, rooting rate, shoot height, the number of leaves, root length, the number of roots, dry weight of shoots, and dry weight of roots, were examined.

#### Effects of PGRs on rooting

The middle section of the scion, excluding the apical and basal areas of the stem, was used to investigate the
effects of PGRs on *A. lancifolium* rooting. The basal part of the scion was immersed in IBA (indole-3-butyric acid, Duchefa, Haarlem, The Netherlands) at varying concentrations (100, 500, and 1,000 mg · L$^{-1}$) for 30 min.
Additionally, a 1:1 (v/v) solution of Rootone (0.4% 1-naphthalacemide, Biosciencekorea, Seoul, Korea) diluted in water was applied for the same 30-min immersion period. Cutting propagation was conducted across five treatments, including a control treatment using Kanuma soil, with four replicates and 15 samples per treatment. The propagation boxes were positioned on their respective plates, and after six weeks, growth characteristics were evaluated. Experimental conditions and growth parameters were consistent with those of the propagation media experiment.

Statistical analysis

Data from the experiments were analyzed using the R program (version 4.2.1). Analysis of variance was performed, and Tukey’s honestly significant difference test was applied to assess the statistical significance of mean values among different treatments for comparison. Percentage data were arcsine-transformed for analysis (Anderson and McLean, 1974).

Results and Discussion

Effects of propagation media on rooting

The results of the analysis on the effects of propagation media on *A. lancifolium* scions are presented in Fig. 2 and Table 2. The cutting survival rate of *A. lancifolium* scions was the highest when Kanuma soil was used as the propagation medium, followed by perlite, commercial potting mixes, and peat moss (81.7 ± 5.0%, 66.7 ± 2.7%, 40.0 ± 7.2%, and 35.0 ± 6.3%, respectively). Kanuma soil also exhibited a significantly high rooting rate and root length (81.7 ± 5.0% and 36.2 ± 2.5 mm, respectively). Contrastingly, peat moss showed a 0% rooting rate. The highest shoot height (69.1 ± 14.3) was observed for commercial potting mixes, followed by Kanuma soil (41.1 ± 5.1), perlite (29.9 ± 2.0), and peat moss (14.4 ± 1.8). Water-holding capacity and permeability should be properly balanced in the soil during the plant growth period. The propagation media has been well known to affect anchoring the cutting in place, holding water for the cutting, supplying sufficient aeration for adventitious rooting (Hartmann et al. 2011). According to Erstad and Gislerod (1994), water in excess prevents proper aeration. Given our results and previous studies, we speculate that peat moss contains high water-holding capacity, which had a negative effect on the rooting of *A. lancifolium*. The most suitable conditions for plant growth

![Fig. 2. Effects of propagation media on rooting. (A) Kanuma soil. (B) commercial potting mixes. (C) perlite, and (D) peat moss. Bar = 10 cm.](image-url)
are 20-30% air volume with a porosity of 85% or more (Gruda and Schnitzler, 2004). When mixing peat moss and perlite, the air porosity was higher than 100% when mixing peat moss and perlite (Verdonck and Demeyer, 2004). In addition, the importance of changes in physical properties depending on the components of the media is recognized and research on this has been conducted consistently. Lee et al. (2009) reported that water-holding capacity was the most critical factor for the rooting of Hydrangea serrata for. acuminata, while Lee and Lee (2007) found that a high proportion of peat moss was advantageous for the rooting of highbush blueberries. In this study, although Kanuma soil showed a positive effect on the root induction and development, a combination of commercial potting mixes and Kanuma soil in an appropriate ratio is recommended for considering overall plant growth of A. lancifolium. The ideal mixture of media varies according to the plant species; nonetheless, further research is needed to explore the effects of physicochemical properties of propagation media.

Effects of PGRs on rooting

Among the known PGRs, auxin is known to facilitate adventitious rooting (Steffens and Rasmussen, 2016) and is widely used for the formation of adventitious roots in cutting propagation (Kochhar et al., 2008). Auxin is a tryptophan-derived hormone involved in promoting growth and development in most plants; additionally, it plays a role in plant gravitropism and phototropism (Woodward and Bartel, 2005; Kepinski and Leyser, 2005). These diverse effects across all plants arise from controlling cell division, elongation, and specific differentiation steps (Davies, 2004). Particularly, IBA is the most effective PGR in the formation of adventitious roots (Pop et al., 2011). Fig. 3 and Table 3 present the results of analyzing the effects of PGRs on A. lancifolium scions. The Rootone-treated treatment, control treatment, and 100 mg · L⁻¹ IBA treatment exhibited significantly higher cutting survival rates than the 500 mg · L⁻¹ and 1,000 mg · L⁻¹ IBA treatments. A similar pattern was found for rooting rates, with that for the Rootone-treated treatment reaching 86.67%. However, all treatments showed similar root lengths. Excessive auxin treatment can negatively affect root growth by increasing the production of ethylene (Burg and Burg, 1966). In this study, although there was no significant difference, lower cutting survival rates and rooting rates were observed in the 500 mg · L⁻¹ IBA treatment, which could partly be attributed to ethylene production. Conversely, the 1,000 mg · L⁻¹ IBA treatment showed the highest root number (9.33 ± 1.0). This indicated that treatment with 1,000 mg · L⁻¹ IBA led to an abundance of short roots (data not shown). The 1,000 mg · L⁻¹ IBA treatment also displayed the highest shoot height and the number of leaves, although these differences were not statistically significant. Further, auxin treatment did not seem to significantly affect the shoots of A. lancifolium scions.

### Table 2. Effect of rooting media on the rooting and shooting of Apocynum lancifolium at 6 weeks after cuttings

<table>
<thead>
<tr>
<th>Rooting media</th>
<th>Survival rate (%)</th>
<th>Shoot height (mm)</th>
<th>The number of leaves</th>
<th>Rooting rate (%)</th>
<th>Root length (mm)</th>
<th>The number of roots</th>
<th>Dry weight (mg)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Commercial potting mixes</td>
<td>40.0 ± 7.2 b</td>
<td>69.1 ± 14.3 a</td>
<td>15.2 ± 1.1 a</td>
<td>38.3 ± 6.9 b</td>
<td>26.9 ± 2.3 b</td>
<td>4.7 ± 0.6 a</td>
<td>0.0819 ± 0.0098 a</td>
<td>0.0054 ± 0.0008 a</td>
</tr>
<tr>
<td>Peat moss</td>
<td>35.0 ± 6.3 b</td>
<td>14.4 ± 1.8 b</td>
<td>4.2 ± 0.8 c</td>
<td>0.0 ± 0.0 c</td>
<td>0.0 ± 0.0 b</td>
<td>0.0 ± 0.0 b</td>
<td>0.0045 ± 0.0012 b</td>
<td>0.0000 ± 0.0000 b</td>
</tr>
<tr>
<td>Perlite</td>
<td>66.7 ± 2.7 a</td>
<td>29.9 ± 2.0 b</td>
<td>9.4 ± 0.5 b</td>
<td>53.3 ± 2.7 b</td>
<td>19.8 ± 2.0 b</td>
<td>3.9 ± 0.1 a</td>
<td>0.0128 ± 0.0016 a</td>
<td>0.0032 ± 0.0010 ab</td>
</tr>
<tr>
<td>Kanuma soil</td>
<td>81.7 ± 5.0 a</td>
<td>41.1 ± 5.1 ab</td>
<td>10.5 ± 0.7 b</td>
<td>81.7 ± 5.0 a</td>
<td>36.2 ± 2.5 a</td>
<td>4.6 ± 0.3 a</td>
<td>0.0271 ± 0.0029 b</td>
<td>0.0051 ± 0.0010 a</td>
</tr>
<tr>
<td>P values</td>
<td>0.0001</td>
<td>0.0021</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0001</td>
<td>0.0018</td>
</tr>
</tbody>
</table>

*Different letters within columns indicate significant differences at $p \leq .05$ using Tukey’s HSD test for multiple variances (± SE).
Effect of Propagation Media and Plant Growth Regulators on Adventitious Rooting of Softwood Cuttings of *Apocynum lancifolium* Russanov

### Table 3. Effect of auxin treatments on the rooting and shooting of *Apocynum lancifolium* at 6 weeks after cuttings

<table>
<thead>
<tr>
<th>Rooting media</th>
<th>Survival rate (%)</th>
<th>Shoot height (mm)</th>
<th>The number of leaves</th>
<th>Rooting rate (%)</th>
<th>Root length (mm)</th>
<th>The number of roots</th>
<th>Dry weight (mg)</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>81.7 ± 5.0 a</td>
<td>41.1 ± 5.1</td>
<td>10.5 ± 0.7</td>
<td>81.7 ± 5.0 a</td>
<td>36.2 ± 2.5</td>
<td>4.6 ± 0.3 b</td>
<td>0.0271 ± 0.0029</td>
<td>0.0005 ± 0.0010</td>
</tr>
<tr>
<td>IBA 100</td>
<td>76.7 ± 4.3 a</td>
<td>40.1 ± 4.7</td>
<td>10.6 ± 0.8</td>
<td>76.7 ± 4.3 a</td>
<td>34.4 ± 1.7</td>
<td>5.9 ± 0.4 b</td>
<td>0.0257 ± 0.0057</td>
<td>0.0091 ± 0.0047</td>
</tr>
<tr>
<td>IBA 500</td>
<td>40.0 ± 4.7 b</td>
<td>43.6 ± 3.7</td>
<td>10.8 ± 0.5</td>
<td>40.0 ± 4.7 b</td>
<td>23.8 ± 2.0</td>
<td>6.0 ± 0.7 b</td>
<td>0.0164 ± 0.0014</td>
<td>0.0037 ± 0.0005</td>
</tr>
<tr>
<td>IBA 1000</td>
<td>31.7 ± 1.7 b</td>
<td>53.1 ± 3.2</td>
<td>11.6 ± 0.3</td>
<td>31.7 ± 1.7 b</td>
<td>32.1 ± 7.9</td>
<td>9.3 ± 1.0 a</td>
<td>0.0215 ± 0.0010</td>
<td>0.0158 ± 0.0066</td>
</tr>
<tr>
<td>Rootonez</td>
<td>91.7 ± 6.3 a</td>
<td>35.7 ± 2.6</td>
<td>9.8 ± 0.5</td>
<td>86.7 ± 6.1 a</td>
<td>37.1 ± 2.6</td>
<td>4.9 ± 0.7 b</td>
<td>0.0204 ± 0.0019</td>
<td>0.0040 ± 0.0007</td>
</tr>
<tr>
<td>P values</td>
<td>0.0001</td>
<td>0.0744</td>
<td>0.3130</td>
<td>0.0001</td>
<td>0.1960</td>
<td>0.0010</td>
<td>0.1540 ± 0.1540</td>
<td></td>
</tr>
</tbody>
</table>

*1-naphthylacetamide 0.4% powder.

zDifferent letters within columns indicate significant differences at *p* ≤ 0.05 using Tukey's HSD test for multiple variances (± standard error).

Fig. 3. Effects of plant growth regulators on rooting. (A) no treatment, (B) 100 mg L⁻¹ IBA, (C) 500 mg L⁻¹ IBA, (D) 1,000 mg L⁻¹ IBA, (E) a 1:1 (v/v) solution of rootone (0.4% 1-naphthylacetamide) diluted in water.
Conclusion

To collect practical data on the cutting propagation of *A. lancifolium*, the effects of propagation media and PGRs on the scions were investigated. The cutting survival rate and rooting rate of *A. lancifolium* scions were the highest (both 81.7%) when Kanuma soil was used as the propagation medium. Contrastingly, the peat moss treatment showed a 0% rooting rate, indicating a lack of adventitious roots induction. The shoot height, the number of leaves, and dry weights of shoots and roots were the highest in the commercial potting mixes. When the cutting survival rate and rooting rate of scions were analyzed for different PGRs, the Rootone-treated treatment showed 10% higher survival rate and 5% rooting rate than the control treatment even though there was no statistical difference. Further, the treatment with high-concentration IBA increased the formation of roots but negatively affected the cutting survival rate and rooting rate. Overall, the results of this study are expected to be useful in establishing a mass production system for *A. lancifolium* via cutting propagation and in developing a method for advancing cultivar breeding.

References


