Systemic transport of green-synthesized silver nanoparticles mediated by *Fusarium solani* as potential resin inducers in *Gyrinops versteegii* (Gilg.) Domke

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Abstract

Agarwood is an aromatic resin formed through the induction of physical stress or microbial infection. Current induction methods still face limitations, including slow processes, low efficiency, and potential tissue damage, so innovative approaches are needed. Nanotechnology offers a promising solution, and silver nanoparticles (AgNPs) are known to enhance plant physiological responses; however, their application in agarwood has not yet been investigated. This study analyzed the physicochemical properties of AgNPs green-synthesized by *Fusarium solani*, their distribution and translocation in agarwood stems, and their effects on interxylary phloem structure. The resulting AgNPs were stable, with zeta potential values ranging from -33.17 to -38.33 mV. The AgNPs underwent internalization, distribution, and systemic translocation, with Ag accumulation increasing as concentration and exposure time increased. The translocation patterns at high and very low concentrations were not markedly different. Anatomical observations showed that the interxylary phloem in AgNP-treated stems was more distinct and more abundant than in the control, indicating activation of defense mechanisms.

Keyword: agarwood; silver nanoparticle; green synthesis; nanoparticle translocation

Introduction

Agarwood resin forms in response to physical stress or microbial infection[1], but current induction methods are slow, damaging to tissue, and inefficient.[2] Therefore innovative approaches are needed. Nanotechnology is a promising option because nanomaterials combine nanoscale size with high reactivity[3] and nanoparticles can be produced simply and sustainably through green synthesis.[4] Fusarium solani, a natural endophyte of agarwood involved in resin formation, can also serve as a green synthesis agent.[6] However, research on agarwood induction has largely focused on biological and chemical agents, and studies on nanoparticles remain limited. Understanding how nanoparticles interact with plant tissues is therefore necessary before their effects can be evaluated. Distribution and translocation analysis can reveal these interactions. Silver (Ag) does not naturally occur in agarwood, making it easy to trace, and it can induce stress through the reactive oxygen species (ROS) pathway,[5] resembling the mechanism of agarwood formation. AgNPs can thus serve as both a translocation marker and an agarwood inducer.

Research Method

Fusarium solani was cultured for 7 days in the dark at 110 rpm, after which the mycelia were filtered, washed, and incubated in sterile deionized water under the same conditions to obtain a cell-free filtrate. $AgNO_3$ was then added to the filtrate and incubated for 5 days in the dark, with a color change from yellow to brown indicating successful synthesis.

The resulting AgNPs were diluted to four concentrations (100%, 50%, 25%, and 5%) and characterized using a Zetasizer Nano-ZS. Subsequently, each concentration was injected into the stem of agarwood, and samples were collected from four points (A, B, C, and D) at 6, 12, 18, and 24 h after injection.

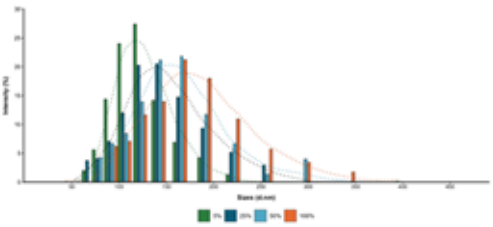


Picture 1. Schematic illustration of the stem injection and sampling design for AgNP application in G. versteegii seedlings.

The samples were then dried and analyzed using X-ray fluorescence (XRF) to detect Ag. In addition, transverse sections of the stems were observed under a light microscope using neutral red staining.

Discussion & Result

The hydrodynamic size of AgNPs varied with concentration. At high concentrations, frequent particle collisions lead to small aggregates and transient clusters.[11] Upon dilution, collision frequency decreases, allowing some aggregates to disperse.[11]

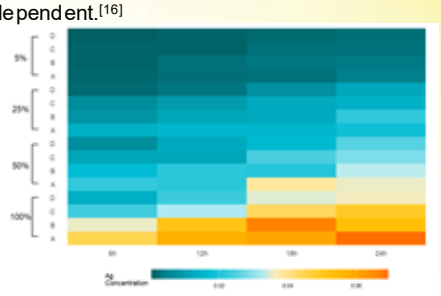


Picture 2. Hydrodynamic size distribution of green-synthesized AgNPs produced by F. solani, as determined by DLS.

An increase in pH occurs because the reduction of Ag^+ becomes more intense,[12] and at alkaline pH, the zeta potential decreases (becomes more negative).[13] A zeta potential below -30 mV indicates good colloidal stability,[14] as stronger electrostatic repulsion reduces the risk of aggregation.[15]

AgNP Code	Conc. (ug/mL)	pH	Size (nm)	*Zeta potential* (mV)
5%	5,4	6,33	171,65 ± 7,55^{d}	-33,17 ± 1,82^{a}
25%	27	6,58	199,67 ± 3,46^{c}	-34,87 ± 1,45^{ab}
50%	54	7,08	219,24 ± 7,19^{b}	-37,13 ± 1,36^{b}
100%	108	7,21	244,53 ± 5,72^{a}	-38,33 ± 1,3^{b}

At 6-12 h, blue coloration dominated, indicating that Ag had only just begun to enter and move through the tissue, so its concentration was still low. By 18-24 h, changes became apparent at the 50% and 100% concentrations, consistent with the uptake and translocation of AgNPs in plant tissues being dose- and time-dependent.[16]

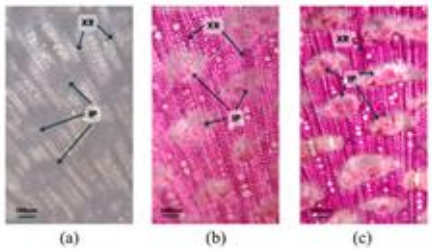


Picture 3. Heatmap of the effect of AgNP exposure duration on the concentration of AgNPs detected in agarwood stems

The translocation factor (TF) represents the ratio of Ag at points B-D relative to point A (the injection site). Ag largely retained at the injection site. The spatial decline in AgNP distribution reflects the limited xylem flow rate and interactions with plant tissues.[18]



Picture 4. Heatmap of the translocation factor (TF) of AgNPs in agarwood stems



The interxylary phloem (IP) in the treated stems differed from that of the control, appearing more abundant and with more distinct boundaries. IP and xylem rays (XR) serve as the structural foundation for agarwood resin formation.[20]

Conclusion

Green-synthesized AgNPs were stable and underwent uptake, distribution, and translocation in agarwood stems. Ag accumulation increased with AgNP concentration and exposure time, whereas translocation increased only with exposure time. AgNP treatment also produced more abundant, more distinct interxylary phloem than the control, indicating defense system activation.

Reference