



## Making *Paulownia elongata*'s Genetic Transformation and Plant Regeneration as Effective as Possible

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### ABSTRACT

Four different types of explants from *Paulownia elongata* were used to assess the impact of plant tissue culture media with different concentrations of growth regulators and carbon sources on organogenesis and plant regeneration. When full leaf or half leaf explants with attached petioles were cultivated on either MS or B5 medium with 25 M thidiazuron, 10 M indole acetic acid, and 30 g L<sup>-1</sup> maltose as a carbon source, the best response to adventitious shoot proliferation was seen among the several treatments used. On a modified MS medium containing 5 M indole butyric acid, proliferated branches were rooted within five days, and plants were effectively acclimated and hardened for greenhouse transfer. Protocols for genetic transformation were improved by using the neomycin phosphotransferase II and green fluorescent protein genes. Using media that was tailored for shoot growth and rooted, *Agrobacterium tumefaciens* mediated transformation of half leaves with petiole explants yielded transgenic plants. PCR and RT-PCR were used to find the GFP transgene's insertion and expression

**Keywords:** *Paulownia*, Plant, Regeneration, Plant extract, Plants

### INTRODUCTION

The fast-growing *Paulownia* genus includes nine species and several natural hybrids of trees are indigenous to China and its neighboring nations. Trees can be cultivated for a variety of purposes, including the production of animal feed, biochar, and bio composite materials. They are also grown for the production of timber, musical instruments, and agroforestry programs. According to research on *Paulownia* species, the tree could be fully utilised as a valuable feedstock for the production of bioenergy. *Paulownia* leaves make an excellent feed crop for tiny ruminants because they are high in proteins and other nutrients. *Paulownia* is a popular phytoremediation agent because of its quick growth, which enables it to remove heavy metals, trace elements, and swine feces from contaminated soils quickly. *Paulownia* species with nectariferous flowers are a plentiful source of honey. Long-term, systematic research on three *Paulownia* species disprove the idea that *P. tomentosa* is invasive, despite the fact that the plant had enjoyed this reputation. The need for planting material for paulownias forced the creation of in vitro regeneration techniques. In response to various combinations of cytokinin and auxin treatments, *Paulownia* plant regeneration was accomplished through somatic embryogenesis or organogenesis employing shoot tip, petiole, stem, leaf, cotyledon, and hypocotyl explants. However, organogenesis based shoot proliferation rates were only 1 shoot to 5 shoots per explant, which was noticeably low. This shows the urgent need for explant selection and medium composition optimization to enhance *Paulownia* regeneration and enable the commercial production of attractive clones as well as serve as target tissues for the insertion of genes and the creation of transgenic plants with value-added features. *Paulownia elongata* transgenic plant attempts have not been very effective due to the plant's poor regeneration after genetic modification. In *P. elongata* and *P. fortunei* explants, agrobacterium-

mediated transformation produced transgenic callus, hairy roots, or shoots with kanamycin resistance at a very low frequency. In earlier studies, various *Agrobacterium tumefaciens* strains were used to evaluate transformation and transgenic plant recovery. The strains were not neutralised, however, and as a result cultures either produced galls or hairy roots, and no transgenic plants were collected. *Agrobacterium* mediated transformation was used in other research, although the amount of recovered transgenic shoots was minimal. Additionally, efforts have been made to enhance biolistic production procedures for transgenic plants in *P. elongata*. After successful transformation, transgenic *Paulownia kawakamii* plants displaying a MADS box transcription factor involved in the regulation of shoot morphogenesis were described. The adventitious shoot induction and proliferation response of *Paulownia elongata* were examined in the current study utilising different explants, basal salts, carbon sources, and doses of growth regulators at various levels.

### DESCRIPTION

Histology and scanning electron microscopy was used to record the shoot induction and proliferation responses. Additionally, a genetic transformation procedure was created using effective regeneration techniques that were developed as a result of these investigations. Using the *Agrobacterium tumefaciens* strain Eha105, transgenic plants expressing the GFP reporter gene and the selectable marker *NPTII* gene were created. The study shows how a highly effective regeneration process was created and used to create transgenic *P. elongata* trees. Sterilization and germination of seeds *P. elongata* certified seeds were purchased from Kisden LLC in Parrottsville, Tennessee, and kept at 4°C until usage. By soaking seeds in distilled water with a few drops of 20 Fungigone for one hour, seeds were allowed to germinate. The seeds were then three times thoroughly rinsed in sterile distilled water and given a 1-minute soak in 70% ethanol. Seeds treated with ethanol were moved to a laminar. Initiation of and growth of new shoots. The licorice roots were gathered in the Gazipur neighborhood of Dhaka, Bangladesh. Licorice was examined to make sure it was clean and free of contaminants before extraction. Then, a tiny piece of licorice was oven dried at 45°C and submerged in water for 24 hours at a ratio of 1:10. The licorice extract was then three times filtered through nylon mesh. On either MS or B5 medium with 25 M TDZ, shoot proliferation was seen in early experiments on *Paulownia* organogenesis. The combination of different carbon sources and IAA concentrations with MS or B5 medium containing 25 M TDZ was explored in order to further increase the shoot proliferation rates. No adventitious shoot induction was seen among the different explants incubated on SIM treatments in the control medium, which lacked growth regulators. Finally, a 50:50 mixture of the produced PVA solution and licorice solution was added. For one hour, a magnetic stirrer was utilised to create a homogenous, uniform solution. The prepared solution is then injected into the electrospinning machine's pump using a syringe. PVA and licorice extract were used in the electrospinning machine to create the nano-fibrous membrane under optimal processing conditions. We present the creation of a highly effective technique for *P. elongata* plant regeneration. The best response to shoot proliferation was seen in whole leaves and half leaves with petioles among the several explants studied. Specific cells in explant tissues undergo differentiation during morphogenesis to become meristemoids, which later develop into shoots and roots. From the cut ends of the cells, the induction of meristemoids and subsequent shoot production.

### CONCLUSION

At last it is concluded that *Paulownia* species have a quick growth rate and offer numerous financial and environmental advantages. Rapid spread of superior clonal material will benefit from an effective *in vitro* technique. The current work clarifies the selection of explants, media, plant growth regulators, carbon sources, and their focus on priming and multiplication stages for *P. elongata* micro propagation. Organogenesis-derived plants demonstrated brisk growth. Additionally, the regeneration.