

Comparing diploid and modified putative polyploid lines of trees *Agathis robusta*, *Elaeocarpus grandis*, and *Paulownia tomentosa*



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Background

Polyploidy or genome doubling is a natural force in plant evolution and can enhance plant growth and resilience. In forest trees, technical difficulties have prevented polyploidisation as a routine breeding tool, but new technology has now been developed for producing putative polyploid lines.

We examined unmodified (diploid) progenitors and modified (putative polyploid) lines of tree species *Agathis robusta* (Araucariaceae), *Elaeocarpus grandis* (Elaeocarpaceae), and *Paulownia tomentosa* (Scrophulariaceae).

Aims and Objectives

1. Verify ploidy levels of clones lines.
2. Verify genome stability of putative polyploids.
3. Quantify morphology, physiology, and growth of putative polyploids.

Materials and Methods

Diploid parents and modified clones of tree species *A. robusta*, *E. grandis*, and *P. tomentosa* about 10-15 cm in height were grown for three to six months in a naturally lit glasshouse. A combination of flow cytometry and chromosome counts was used to verify ploidy of clones compared to diploid lines. A total genomic marker, Amplified Fragment-Length Polymorphism (AFLP) verified the genome stability. A range of techniques were used to compare the foliar traits and growth of polyploid clone lines with diploid parents. Only, *A. robusta* and *P. tomentosa* clone lines were available for foliar and growth traits.

Results

Nuclear DNA content and genome stability

	Genome content (linear fluorescence)	Dissimilarity to other clone lines of same species (%)	Genome stability within clone lines (%)
<i>A. robusta</i>			
Parent	NA	4	99
Clone 1	NA	7	98
Clone 2	NA	9	99
Clone 3	NA	9	99
<i>E. grandis</i>			
Parent	110	1	100
Clone 1	140	4	100
Clone 2	135	4	100
<i>P. tomentosa</i>			
Parent	100	12	99
Clone 3	135	17	96
Clone 7	75	16	96
Clone 15	100	15	98

Table 1
Nuclear DNA content obtained from flow cytometry and genome stability within clone lines and genomic re-organization of clones determined from AFLP profiles for *A. robusta*, *E. grandis* and *P. tomentosa*. NA = data not available.

Leaf morphology

Modified clones of *A. robusta* and *P. tomentosa* had larger leaf size and greater leaf elongation rate compared to their parents.

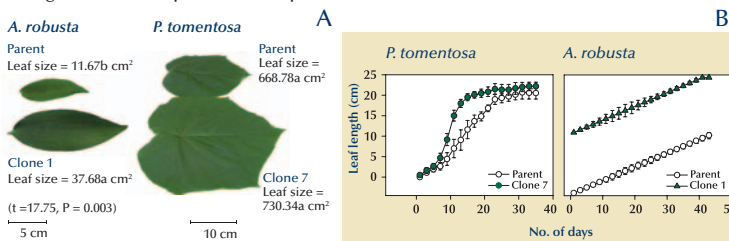


Figure 2
Differences in leaf size (A) and leaf elongation rate (B) for parents and clones of *A. robusta* and *P. tomentosa* after 3 months and 6 months growth in the glasshouse, respectively.

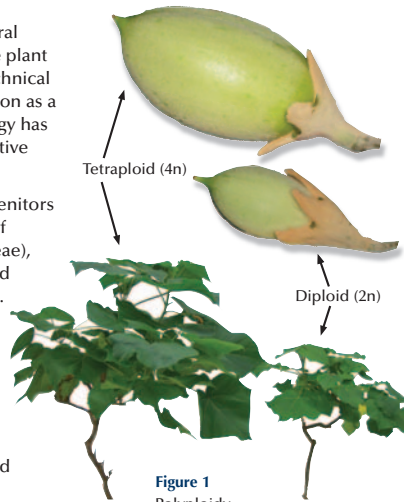


Figure 1
Polyploidy

Leaf anatomy

Modified clones of *A. robusta* and *P. tomentosa* exhibited thicker leaf blades, palisade mesophyll and epidermis, and larger stomatal aperture lengths than their parents.

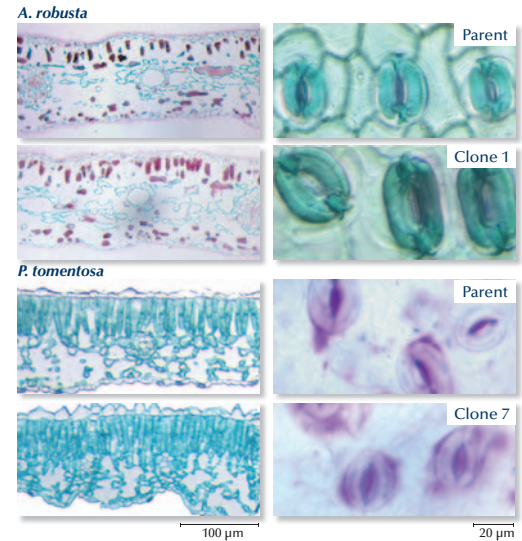


Figure 3
Differences in leaf blade thickness and stomatal size of parent and clone lines of *A. robusta* and *P. tomentosa* species.

Leaf physiology

Agathis robusta and *P. tomentosa* clone lines had an increased photosynthetic rate and different carboxylation and electron transport rates than parental lines.

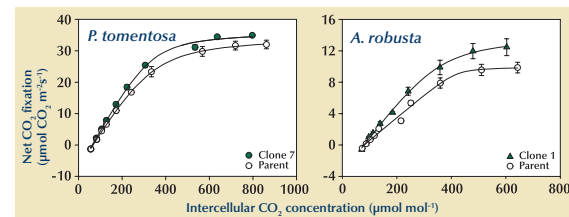


Figure 4
Differences in CO₂ response curves between parent and clone lines of *A. robusta* and *P. tomentosa* species.

Plant growth

Agathis robusta and *P. tomentosa* modified clone lines had increased growth in plant height and shoot biomass compared to diploid parental lines after 3 months and 6 months growth in the glasshouse, respectively.

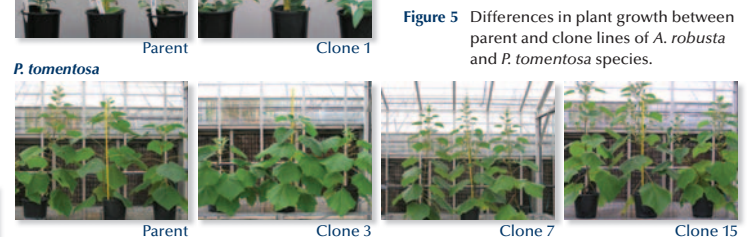


Figure 5
Differences in plant growth between parent and clone lines of *A. robusta* and *P. tomentosa* species.

Conclusions

Newly generated clone lines have undergone a successful stable polyploidisation process.

Properties associated with selected polyploid clones included increased nuclear DNA content, larger and thicker leaves with greater stomatal aperture lengths, increased photosynthetic rates and shoot biomass than the parent plants.

Acknowledgements

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