

Subin Hinjan 2010: Transformation of Antisense *LIM* Gene into *Eucalyptus camaldulensis* Dehnh. Doctor of Philosophy (Agricultural Biotechnology), Major Field: Agricultural Biotechnology, Interdisciplinary Graduate Program.
Thesis Advisor: Assistant Professor Sermsiri Chanprame, Ph.D. 120 pages.

The LIM proteins are transcription factors that involved in lignin biosynthesis pathway. In this research, we investigated the function of *LIM* gene in eucalyptus by antisense transformation. The partial *LIM* gene was cloned from *Eucalyptus camaldulensis* cDNA and constructed into a binary vector, pCAMBIA1301. The recombinant plasmid was then transformed into *Agrobacterium tumefaciens* strain EHA105 for the genetic transformation of *in vitro* elite clone of *E. camaldulensis*. The putative transgenic lines were selected based on their hygromycin resistance character and the existence of antisense *LIM* was confirmed by PCR analysis and Southern blot hybridization.

Eight transgenic lines were transplanted in a bio-safety greenhouse. It was found that survival rate and growth of the transgenic lines were higher than the wild type. The expression of *LIM* and other genes in lignin biosynthesis pathway were measured by real-time PCR. The expression of the *LIM*, *C3H* and *C4H* were reduced in all transgenic eucalyptus lines, especially in *LIM* gene which was more than 99.8 % reduced when compare to the wild type. In contrast, the *PAL*, *CCR* and *CAD* genes expression were increased in transgenic lines. The hot water extraction substrates were lower, while wood density and pentosan content were higher than those of the wild type. One transgenic line has much thicker cell wall than the wild type. The result implies that the LIM protein may play roles as transcription factor in eucalyptus lignin biosynthesis. It may also involve in the synthesis of pentosan, cell wall and hot water extraction substrates.

Student's signature

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