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## Association Genetics of the *Cinnamyl Alcohol Dehydrogenase (CAD)* and *Cinnamate 4-hydroxylase (C4H)* Genes with Basic Wood Density in *Neolamarckia cadamba*

<sup>1</sup>B.L. Tchin, <sup>1</sup>W.S. Ho, <sup>2</sup>S.L. Pang and <sup>1</sup>J. Ismail

<sup>1</sup>Forest Genomics and Informatics Laboratory, Department of Molecular Biology, Faculty of Resource Science and Technology, Universiti Malaysia Sarawak, 94300, Kota Samarahan, Sarawak, Malaysia

<sup>2</sup>Applied Forest Science and Industry Development, Sarawak Forestry Corporation, 93250 Kuching, Sarawak, Malaysia

**Abstract:** Association genetics study is a powerful approach to detect the potential genetic variants (i.e., SNPs) underlying the common and complex adaptive traits. Once the quantitative trait nucleotides are identified, such powerful approach provides significant advantages to the forest industry. Hence, attempts were made to discover SNPs from *Neolamarckia cadamba* partial *C4H* (3,538 bp) and *CAD* (2,354 bp) DNA sequences and further associate those SNPs with basic wood density. Overlapping primers were designed in flanking the partial *C4H* and *CAD* DNA from 12 *N. cadamba* trees. The amplified DNA fragments were sequenced and the basic wood density measurements were determined for each tree. The sequence variation analyses revealed that there were 60 and 32 SNPs detected in the partial *C4H* and *CAD* DNA sequences, respectively. Those SNPs were distributed throughout the exon, intron, 5'-UTR and 3'-UTR regions. The total nucleotide diversities were  $\pi = 0.00302$  and  $\theta_w = 0.00412$ . The synonymous mutations ( $\pi = 0.00983$ ;  $\theta_w = 0.01210$ ) were more common than nonsynonymous mutations ( $\pi = 0.00045$ ;  $\theta_w = 0.00089$ ) for both *C4H* and *CAD* genes. LD declined linearly over short distance at the loci examined. Association genetics study also revealed that 4 and 6 SNPs from *C4H* and *CAD* genes, respectively were in significant associations with basic wood density of *N. cadamba* ( $p < 0.05$ ). The genetic variation identified by the SNP markers, once validated, will facilitate the selection of *N. cadamba* parental lines or seedlings with optimal quality through Gene-assisted Selection (GAS) approach.

**Key words:** *Neolamarckia cadamba*, *cinnamate 4-hydroxylase*, *cinnamyl alcohol dehydrogenase*, single nucleotide polymorphism, association genetics

### INTRODUCTION

*Neolamarckia cadamba*, or locally known as Kelampayan is one of the Lesser-known Commercial Timbers (LKCT) which possess various benefits for wood-based industry. It is used for making picture frame, moulding, skirting, wooden sandals, disposable chopstick, general utility furniture, veneer, plywood as well as pulp and paper (Lim *et al.*, 2005). The leaves, roots and bark of Kelampayan also have been reported to have high pharmacological values (Joker, 2000; Patel and Kumar, 2008; Acharyya *et al.*, 2010). To date, studies on Kelampayan at molecular level are still limited. Recently, a Kelampayan tree transcriptome database (NcdbEST) had been developed (Ho *et al.*, 2010). It provides useful genomics information and resources for researchers to deeply explore the genomics basic of the Kelampayan.

Association genetics or mapping study is an alternative approach that enables researcher to use modern genomic approaches to identify marker-trait associations at higher resolution compared to the widely used family-based Quantitative Trait Locus (QTL) mapping. It has emerged as a new tool in dissecting the genetic basis of complex traits such as physical and chemical wood property traits variation down to sequence level by harnessing the genetic variation at the population level. In just a few years, association genetics study has been broadly embraced in forest tree species, such as *Pinus*, *Pseudotsuga*, *Populus* and *Eucalyptus* (Neale and Kremer, 2011). Association genetics study is a powerful approach in identifying genes or loci that contribute to variation in complex traits (Gonzalez-Martinez *et al.*, 2006; Hall *et al.*, 2010). It has long been argued that the phenotypic variations among