

Expression of phenylalanine ammonia-lyase and lignin content in lignin-contrasting sugarcane varieties

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Lignin is a limiting factor to use sugarcane bagasse for bioethanol production. Aiming to identifying the possible genes coding for enzymes of lignin biosynthesis in sugarcane, an *in silico* study was conducted in the SUCEST database. Sequence baits for each enzyme were obtained from other plant genomes (*Sorghum bicolor*, *Oryza sativa*, *Zea mays* and *Arabidopsis thaliana*) and a comparative analysis was made with the SUCEST databank through the construction of phylogenetic trees. This process allowed us to identify the most probable orthologous sequences of the SAS for each one of the enzymes (4 *PAL*, 2 *OMT*, 4 *HCT*, 1 *F5H*, 2 *CCR*, 4 *CCoAOMT*, 4 *CAD*, 4 *4CL*, 5 *C4H*, 1 *C3H*, 2 *AldOMT*). Preliminary expression analyses of the 4 orthologous of *PAL*, by semiquantitative RT-PCR, were carried out using two varieties of sugarcane contrasting for lignin content, IAC04-683 (low) and IAC04-589 (high), using different stages of internode development (immature, intermediate and mature). Although the lignin content was low in immature internodes, the overall expression of *PAL2*, *PAL3*, and *PAL4* was high in these tissues. *PAL2* and *PAL4* also kept a high expression level in intermediate internodes, however, they exhibited low expression level in mature internodes, what can be correlated with low lignification activity in these mature tissues. *PAL1* showed a constitutive expression in different tissues and varieties. Analyses of lignin content were realized by Klason and thioglycolic acid methods. In the Klason insoluble fraction lignin accumulated in intermediate and mature internodes, mainly in the IAC04-589 variety. On the other hand the content of the Klason soluble fraction was similar throughout the stem in both varieties. Thioglycolic analyses showed similar results with those obtained in the insoluble fraction. Analyses of all identified genes are being conducted by qPCR, coupled by lignin analysis by mass spectrometry.

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