

Molecular Characterization of A Cellobiohydrolase Gene Family in the Fungus *Pleurotus ostreatus*

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Cellulose is the most abundant biological polymer on Earth. Its chemical composition consists of D-glucose units linked by β -1,4- glycosidic bonds forming linear polymeric chains with a reducing and a non-reducing end. Cellulose chains may either adhere to each other, via hydrophobic and van der Waals interactions, forming crystalline structures or remain more loosely packaged (amorphous cellulose). Consequently, the physical structure and morphology of native cellulose is complex and not uniform.

Biological degradation of cellulose depends on the action of three types of enzymes: endoglucanases (E.C.3.2.1.4), cellobiohydrolases (E.C.3.2.1.91) and β -glucosidases (E.C.3.2.1.21). All them hydrolyse β -1,4-glycosidic bonds but they differ on the substrate specificity. Endoglucanases hydrolyse the amorphous regions of the cellulose fibers generating new reducing and non-reducing ends, cellobiohydrolases attack the molecule ends yielding cellobiose units, and β -glucosidases hydrolyse cellobiose molecules yielding glucose.

Cellobiohydrolases can be classified into two groups: type I (CBHI) and type II (CBHII), each having opposite chain-end specificities. CBHI prefer the reducing ends while CBHII act at non-reducing ends.

By the screening of a genomic library from the basidiomycete *Pleurotus ostreatus* var. *florida*, we have isolated five *cbhI* genes, named *cbhI1*, *cbhI2*, *cbhI3*, *cbhI4* and *cbhI5*, proving the occurrence of a multigenic family coding for this enzymatic activity. Using this sequences as probe, it has been possible to know the conditions in which are expressed those genes. This has allowed the synthesis of the each gene cDNA and, by comparison of this sequence with the corresponding genomic sequence, the characterization of their structure.

On the other hand, using the RFLP technique and a progeny of 80 monokaryons derived from the dikaryon N001, the five genes have been mapped on the linkage map of *P. ostreatus* var. *florida* mapping the *cbhI1* to the chromosome IV and the others to the chromosome VI.