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"Lytic Polysaccharide MonoOxygenase" (LPMO) enzymes are widespread enzymes with a copper atom in the active site. In the natural environment, they stimulate the conversion of biopolymers (for example, cellulose or chitin) [1]. For several years now, IFPEN and INRAE have been working on gaining a better understanding of these enzymes [2, 3] and also on the cellulases produced by the T. reesei fungus, with a view to their use for bio-based industrial processes.

Fungi possessing up to 60 coding genes for AA9 LPMOs

A PhD thesis [4] led by the two institutes and supported by the ANR SNOEBORD project studied a sub-family of LPMOs possessing **atypical and previously unknown properties** [5]. Some polypores, fungi that decay wood (white rot), **can possess up to 60 coding genes**¹ for LPMOs belonging to the AA9 LPMO family². However, the functional relevance of such redundancy has yet to be

discovered. Previous comparative transcriptomic studies of six Polypore fungi cultivated on cellulosic substrates demonstrated the overexpression of numerous coding genes for AA9, including some with a C-terminal domain³ of unknown function "X282" (figure 1).

¹ Gene whose translation leads to the formation of a protein

² AA9: Enzymes classified as having an Auxiliary Activity (AA) family 9 according to reference nomenclature

³ Enzymes are amino acid chains linked by a peptide bond. The enzyme's first amino acid thus possesses a free amino called N-terminal and the last a free carboxylic acid function, called C-terminal.



Figure 1 : Three-dimensional structural model of the PcoAA9 LPMO with its X282 domain (cyan) and 9-residue motif (orange)

A sub-family of AA9 LPMO has been shown to have strange phosphate affinity

On the basis of structural predictions and phylogenetic analyses⁴, we selected and characterized six AA9-X282 LPMOs with different domains and atypical characteristics. Unexpectedly, having examined a broad range of conditions, these AA9-X282s only demonstrated weak cellulose bonding properties and little or no cellulolytic oxidative activity. **The proteomic analysis then revealed the presence of multiple phosphorylated residues on the surface of these AA9-X282 LPMOs**, including a conserved residue near the copper site. More in-depth analyses relating to a glycine-rich C-terminal extension with 9 residues suggested it may possess phosphate binding properties.

⁴ A phylogenetic analysis is used to compare the DNA sequence of genes from the same family

LPMO AA9 enzymes with more than one trick up their sleeve

The results of this work partially call into question the involvement of AA9 LPMO enzymes in the degradation of plant cell walls, and open up new avenues to explain the divergence in function of

certain LPMO enzyme members of the AA9 fungal family.

References

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