

UTILIZING A LYTIC POLYSACCHARIDE MONOOXYGENASE FROM *FUSARIUM OXYSPORUM* FOR THE ADVANCED PRODUCTION OF NANOCELLULOSE FROM WHEAT STRAW

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ABSTRACT

The focus of this study is on the characterization of different lytic polysaccharide monooxygenases (LPMOs) belonging to the AA9 family of CAZy database (<http://www.cazy.org/>), aiming to explore their potential in enhancing the production of bio-based materials. A recently identified and characterized AA9 LPMO from *Fusarium oxysporum* (FoLPMO9) is investigated, with an emphasis on harnessing its capabilities for the isolation of nanocellulose, a promising green biomaterial with a great potential to replace plastics¹. FoLPMO9 was expressed heterologously in *Pichia pastoris* under the methanol-induced AOX1 promoter, followed by biochemical and functional characterization. Notably, FoLPMO9 exhibited versatile regioselectivity, demonstrating C1, C4, and C1/C4 oxidative cleavage patterns on cellulose substrates. At the next step, an enzymatic process for the isolation of nanocellulose from the solid pulp of OxiOrganosolv pretreated wheat straw biomass² was introduced. The process entailed a multi-step enzymatic treatment utilizing hemicellulases and cellulases³, succeeded by the oxidative cleavage mediated by LPMOs. Apart from FoLPMO9, other LPMOs with different stereoselectivity, namely TthLPMO9G² and MtLPMO9H⁴, were tested. Moreover, a post-treatment step with each AA9 LPMO was also conducted in order to assess their varied effects on the final nanocellulose product. The efficacy of this approach was validated through advanced nanocellulose characterization techniques. The findings underscore the significance of employing AA9 LPMOs for the enzyme-mediated isolation, as well as the functionalization of nanocellulose, providing a sustainable and efficient approach for the production of bio-based, high-added-value products from lignocellulose.

KEYWORDS: Nanocellulose, Lytic Polysaccharide Monooxygenases, *Fusarium oxysporum*, Wheat Straw

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