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Novel enzymatic tools for lignin degradation

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PURPOSE OF THE ABSTRACT

In Nature, the white-rot basidiomycetes are the most extensively studied natural lignin-degrading microorganisms [1]. The ligninolytic activity of these fungi has been mainly associated with the production of extracellular, non-specific and oxidative ligninolytic enzymes: laccases and high-redox peroxidases [2]. On the other hand, Peroxygenases are largely unexplored and may provide a rich source of new ligninolytic and auxiliary enzymes, given the breadth of different kind of enzymes that belongs to this class, and pathways that functionalise and/or degrade aromatic compounds [3]. Moreover, peroxygenases emerged as a promising group of enzymes that could replace P450s in many reactions; indeed, peroxygenases could step in instead of P450s for biocatalytic transformations and thus enable scale-up of such processes [4]. The utilization of P450s is often complicated due to multi-component systems. Even in the case of BM3-like P450s, the catalytically self-sufficient flavocytochrome fusion proteins, there is still a dependence on NAD(P)H and significant uncoupling rate, which further leads to the enzyme inactivation [5].

This work delves into the substrate scope exploration of the novel bacterial peroxygenases obtained through genome mining. A panel of novel enzymes is efficiently produced in E. coli with good yield (50-300 mg/L). Through screening a set of various substrates and employing available structural data and computational tools the aim is to provide kinetic, stability, spectroscopic, electrochemical, and structural fingerprints of new and engineered ligninolytic enzymes endowed with peroxygenase activities. The identification of the mechanistic and molecular features of wild-type and mutated enzymes will contribute to the construction of a unique toolbox of new ligninolytic enzymes as well as experimental tools for their characterization, which will have a key impact on their further biotechnological exploitation.

FIGURES

FIGURE 1

FIGURE 2

KEYWORDS

Biocatalysis | Green chemistry | Novel enzymes | Protein engineering

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