FUNCTIONAL CHARACTERISATION OF RECOMBINANT PSEUDOMONAS AERUGINOSA LIPOXYGENASE

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In the industrial sector, there is an increasing reliance on biocatalysis, an approach that uses enzymes as catalysts, to enhance sustainability and process efficiency. Biocatalysis offers a greener alternative to traditional chemical methods by enabling highly selective reactions to be carried out under mild conditions, thereby reducing energy consumption and environmental impact. One particularly promising application of biocatalysis is the transformation of fatty acids into valuable compounds, including biofuels, polyols, and flavour compounds. In this context, lipoxygenases (LOX) have been identified as key enzymes in the peroxidation of polyunsaturated fatty acids. While most research has focused on human and plant LOX, the potential of bacterial LOX, which was only recently discovered, remains largely unexplored [1].

Among bacterial enzymes, *Pseudomonas aeruginosa* lipoxygenase (PaLOX) represents a promising candidate due to its reported stability and ability to oxidise a broad range of fatty acid substrates [2]. To better evaluate its suitability for biocatalytic applications, this study focused on the biochemical and functional characterisation of recombinant PaLOX. The *palox* gene was expressed in the yeast *Pichia pastoris*, enabling extracellular production of the recombinant enzyme. PaLOX was purified by immobilised-metal affinity chromatography. The effects of temperature, pH, and selected inhibitors on enzyme activity and stability were investigated. Enzymatic activity was measured using a spectrophotometric lipoxygenase activity assay. Further details of the study will be presented during the poster session.

[1] R. Chrisnasari, M. Hennebelle, J.-P. Vincken, W. J. H. Van Berkel, and T. A. Ewing, “Bacterial lipoxygenases: Biochemical characteristics, molecular structure and potential applications,” Biotechnology Advances, vol. 61, p. 108046, Oct. 2022, doi: 10.1016/j.biotechadv.2022.108046.

[2] C. Hashem, H. Stolterfoht, C. Rinnofner, S. Steinberger, M. Winkler, and H. Pichler, “Secretion of Pseudomonas aeruginosa Lipoxygenase by Pichia pastoris upon Glycerol Feed,” Biotechnology Journal, vol. 15, no. 11, p. e2000089, Aug. 2020, doi: 10.1002/biot.202000089.