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Pyranose 2-oxidase as a Biocatalyst for Synthesis of Rare Sugars

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Abstract:

Pyranose 2-oxidase catalyzes the oxidation of aldopyranoses by using molecular oxygen as an electron acceptor to yield the corresponding keto-aldoses and hydrogen peroxide. This enzyme belongs to the Glucose-Methanol-Choline (GMC) oxidoreductase superfamily and contains flavin adenine dinucleotide (FAD) as a cofactor. P2O catalyzes regiospecific oxidation at the C2 position. Mechanistic and structural investigation of P2O has been carried out extensively. Site-directed mutagenesis, kinetic isotope effects, pH-dependent and transient kinetics studies have shown that P2O follows a hydride transfer mechanism. P2O has been proposed to be a key biocatalyst in the biotransformation of carbohydrates. P2O can potentially convert sugars to provide a pool of keto-sugar intermediates for synthesis of rare sugars, fine chemicals and drugs. Therefore, this research use enzyme engineering approach to improve the performance of P2O in biocatalysis. Site-directed mutagenesis of active site residues that are crucial for sugar binding but not involved in sugar oxidation will be carried out. To examine substrate specificity, common sugars will be tested if they are substrates for P2O wild-type and variants. Variants with good sugar oxidation activity when compared to the wild-type enzyme will be further studied to identify the effect of mutation on flavin reduction. Product identifications will also be carried out to identify keto-sugar products from wild-type or variants reaction. Knowledge obtained from these studies should be useful for industrial applications to produce high value sugars.

Dr. Thanyaporn Wongnate is a faculty member at the school of Biomolecular Science and Engineering, Vidyasirimedhi Institute of Science and Technology (VISTEC), Thailand. She obtained her Ph.D from Department of Biochemistry, Mahidol University in 2011. Her Ph.D. thesis investigated functional roles of key catalytic residues of the enzyme pyranose 2-oxidase using various techniques. Dr. Wongnate did post-doctoral research training at Department of Biological Chemistry, University of Michigan. Her postdoctoral research investigated the reaction mechanism of methyl-coenzyme M reductase, the enzyme that is responsible for biological methane production and anaerobic methane oxidation. Her research theme focuses on studying the

processes that are important in the global carbon cycle, bioenergy, and biorefinery.