Functional expression of fungal peroxidase (*Coprinus cinereus* peroxidase) in *Pichia pastoris* and characterization

Dong Seok Yun^{1,3}, Bo Young Lee¹, Hyun Seong Jeong¹, Yong Hwan Kim², Bong Keun Song¹, Beom Soo Kim³

¹Korea Research Institute of Chemical Technology ²Dept. of Chemical Eng. Kwangwoon University ³Dept. of Chemical Eng. Chungbuk National University

Peroxidase has many applications such as polymerization of phenolic compounds and waste water treatment. A cDNA encoding for a peroxidase was isolated from the fungus *Coprinus cinereus* by RT-PCR. It contains an open reading frame of 1092 bps. The deduced mature protein consisted of 344 amino acids and was preceded by a signal peptide of 20 amino acids. The cDNA was cloned into the $PIC_Z \alpha$ A(wCiP) and expressed in the *Pichia pastoris* X-33. Peroxidase-secreting transformants were selected by their ability to oxidize the substrate 2'2-azinobis-3-ethylbenzthiaoline-6-sufonic acid (ABTS).

While wCiP activity of P. pastoris grown in flask was only 120U/mL, in bioreactor on the optimized condition, extracelluar wCiP activity was a maximum of 1151U/mL. And the Rz value of purified wCiP using SEC was measured 0.843, which was comparative with that of CiP from wild *Coprinus cinereus* fungi.