

## Cell surface display of lipase on the *Pseudomonas putida* and its biocatalytic applications to enantioselective resolution

Seung Hwan LEE<sup>1</sup>, and Sang Yup LEE<sup>2\*</sup>

<sup>1</sup>KAIST, Dept of Chemical & Biomolecular Engineering,  
BioProcess Engineering Research Center,

<sup>2</sup>KAIST, Dept of Chemical & Biomolecular Engineering,  
BioProcess Engineering Research Center,

Department of BioSystems and Bioinformatics Research Center

Here we will present the development of a new cell surface display system in *Pseudomonas putida* KT2442 using OprF, an outer membrane protein of *P. aeruginosa*, as an anchoring motif by C-terminal deletion-fusion strategy. Expression of fusion protein on the cell surface was confirmed by western blot analysis, immunofluorescence microscopy, FACS analysis and measurement of whole cell lipase activity. The whole cell lipase activity of recombinant *P. putida* T2442 was 5 times higher than that of recombinant *Escherichia coli* displaying lipase in the same manner. The optimal conditions for surface displayed lipase were 47°C and pH 9.0, and the whole cell lipase activity was maintained over 90% of the initial activity in organic solvents at 47°C during a week.

As a biocatalytic application, enantioselective resolution of 1- phenyl ethanol was carried out in hexane. (R)-phenyl ethyl acetate was successfully produced with the conversion of 41.9% and the enantiomeric excess of greater than 99% in 36 h reaction. These results suggest that OprF anchor can be used for the efficient display of proteins on the *P. putida* KT2442 and consequently for various biocatalytic applications.

This work was supported by MOCIE grants from the Intelligence Bioinformatics and Application Center (TGW10011093) at the KRIBB