

## Effect of carbon sources on whole-cell styrene monooxygenase reaction with the styrene isomerase deletion mutant of *Pseudomonas putida*

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### Abstract

The chiral (*S*)-styrene oxide is an important chemical intermediate for synthesis of several drugs and functional food ingredients.<sup>1)</sup> The reaction of styrene into enantio-pure (*S*)-styrene oxide was studied with the styrene isomerase deletion mutant of *Pseudomonas putida* SN1 (*styC*-) as a whole-cell styrene monooxygenase. Since the reaction is dependent on regeneration of endogenous NADH, the effect of various carbon sources including glucose, citrate and formate on the rate and yield of the epoxidation reaction was investigated. The experiment was conducted in batch mode under the conditions that neither styrene nor O<sub>2</sub> was limiting the reaction rate. The epoxidation reaction rate with different carbon sources decreased in the order; citrate > glucose > formate > no carbon added. For each mole of styrene an equimolar (*S*)-styrene oxide was produced regardless of the carbon source used, but oxygen requirement (YO<sub>2</sub>, mol O<sub>2</sub>/mol styrene oxide) was significantly varied as follows; 1.69 with citrate, 2.13 with glucose, 3.36 without added carbon source, and 3.49 with formate. This result indicates that citrate is the most efficient carbon source in terms of reaction rate and O<sub>2</sub> consumption. However, material balance calculation suggested that, for all the exogenous carbon sources used, a large proportion of NADH produced and O<sub>2</sub> consumed were wasted without being used for the SMO reaction. Since O<sub>2</sub> transfer is the most important factor usually limiting the volumetric productivity of oxygenase reaction, metabolic engineering to increase YO<sub>2</sub> will be crucial for the further development of whole-cell SMO biocatalyst.

### **Reference**

1. Panke S, de Lorenzo V, Kaiser A, Witholt B, Wubbolts MG., 1999. Engineering of a stable whole-cell biocatalyst capable of (S)-styrene oxide formation for continuous two-liquid-phase applications. *Appl. Environ. Microbiol.* 65(12), 5619-2.