

Electronic Supplementary Information

Efficient Asymmetric Biosynthesis of (R)-(-)-epinephrine in the hydrophilic ionic liquid-containing systems

Zhiqun Xu,¹ Qiao Wu¹, Meixia Yang,¹ Shuai Wang,¹ Zhenshou Wang,¹ and Xiaoping Xu ^{*1}

¹Key Laboratory of Biopharmaceutical, College of Chemistry, Fuzhou University, Fuzhou, 350108, P.R.China

(E-mail: Xiaoping Xu: xu@fzu.edu.cn)

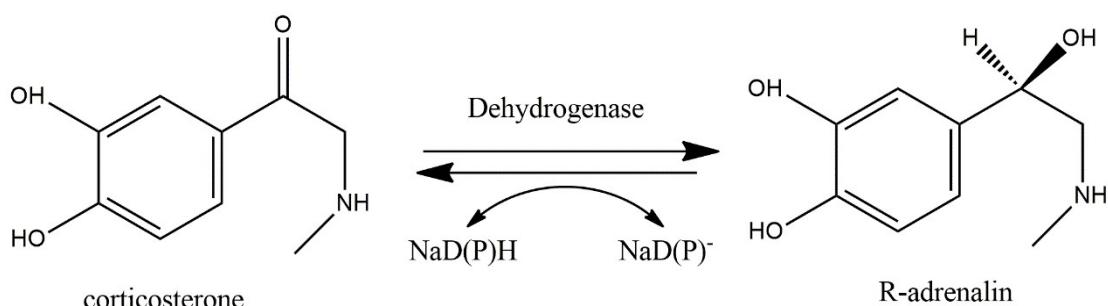


Fig. S1 Asymmetric bio-catalysis of R-adrenalin from corticosterone

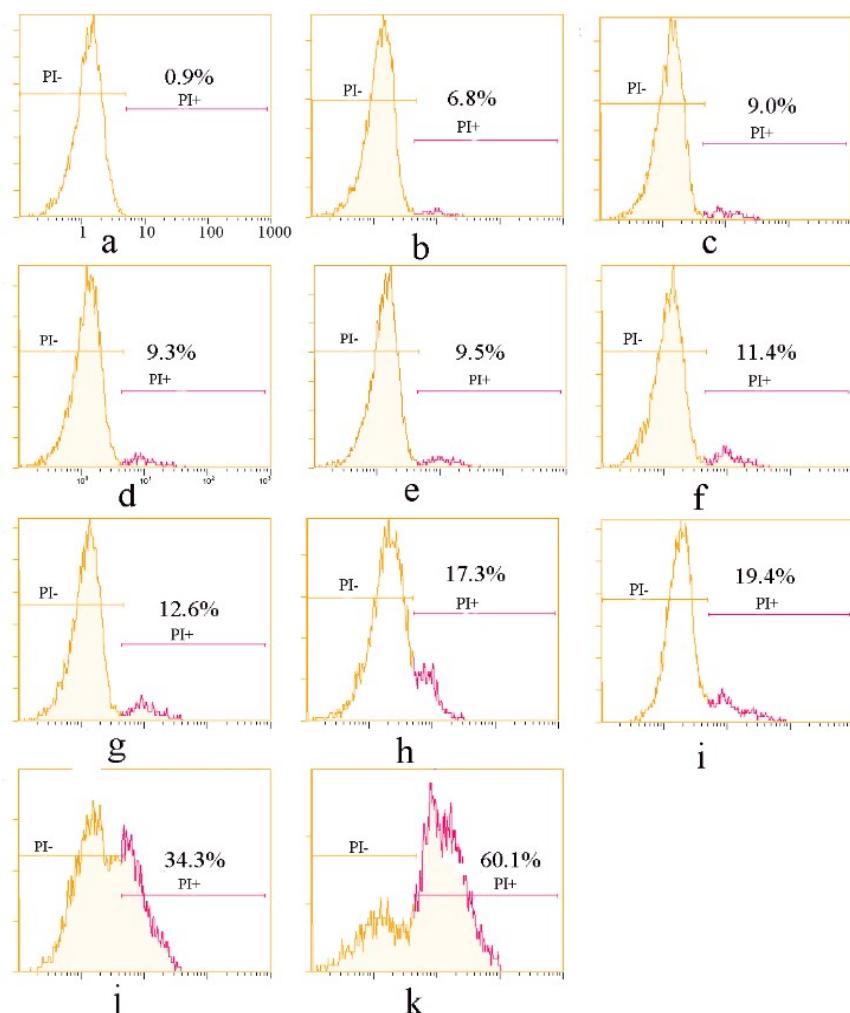


Fig. S2 Membrane integrity of *Acinetobacter* sp. UN-16 cells measured by FCM after being incubated in various hydrophilic ionic liquids *Abscissa—fluorescence intensity ; ordinate—the number of cells PI⁺—the number of dyed cells, PI[□]—the number of no dyed cells, a-k: H₂O, [HOOCEMIM]NO₃, [HOEMIM]NO₃, [EMIM]NO₃, [EMMIM]NO₃, [HOEMIM]Cl, [EMIM]Cl, [BMIM]Cl, [C₇MIM]Cl, [EMIM]BF₄, [HOEMIM]OTF.

Experimental

Asymmetric Biosynthesis of (R)-(-)-epinephrine in the hydrophilic ionic liquids

The experiment was carried out in 11 Erlenmeyer flasks(50ml) containing 20 ml of fermentation medium, each flask was inoculated with 200 μ l seed culture solution. After being incubated for 18 hours (34°C, 200 r/min, pH7.0), a fixed amount of adrenalone (12 mmol/L) was inoculated each flask. One of flasks was inoculated with 1mL water as a contrast group, the rest of flasks were inoculated with 1mL ionic liquid ([EMIM]Cl, [BMIM]Cl, [C₇MIM]Cl, [EMIM]NO₃, [EMIM]BF₄, [HOEMIM]Cl, [HOEMIM]NO₃, [EMMIM]NO₃, [HOOCEMIM]NO₃ and [HOEMIM]OTF) respectively. Then incubated for 22 hours(34°C, pH7.0, 200 r/min).

Measurement of the effects on the biological catalysis by *Acinetobacter* sp.UN-16 cells

The metabolic activity retention(%) was defined as the ratio of the consumed glucose amount by the cells pretreated in various ionic liquids to that pretreated in aqueous solution(as the control, was taken as 100%). The metabolic activity retention of *Acinetobacter* sp.UN-16 cells was assayed after 22h cultivating to various ionic liquid systems. After the cells were separated from the reaction medium, they were transferred to glucose solution (10 mL, 10 g/L), and then incubated at 34°C and 200 r/min for 4 h. The glucose concentration in the medium was determined with DNS method. In recent years, High Performance Liquid Chromatography (HPLC) and High Performance Capillary Electrophoresis(HPCE) are the most practical, representative technology in the field of bioinstrumentation. Here, the yields of the (R)-(-)-epinephrine were analyzed by HPLC, and the HPCE method was used for the distinguishing between (R)-(-)-epinephrine and (S)-(-)-epinephrine(the e.e. value was defined as the content of (R)-(-)-epinephrine).

Measurement of the effects on the *Acinetobacter* sp.UN-16 cells

The cell-free supernatants containing released intracellular components(primarily proteins and nucleic acids) were separated by centrifuging and diluted for measurement of OD. The OD₂₆₀ and OD₂₈₀ of the samples were determined using ultraviolet spectrophotometer from the samples at 0h and 22h. The OD₂₆₀ and OD₂₈₀ values at 22h were revised by deducting the corresponding 0 h absorbance values, and these corrected values for the 22 h samples are a measure of the release of intracellular components into the medium during incubation with ionic liquids. For the *Acinetobacter* sp.UN-16 cells, the flow cytometry was used to detect the cell membrane integrity after being dyed with propidium iodide(1 mg/mL) at 4°C in the dark. The fluorescent signal was measured at 660 nm, and the percentage of damaged cells can be represented by the PI dyed cells dividing the total number of cells.