

Supporting information

for
**Study on the kinetics of Homogeneous Enzyme reactions
confined in a Micro/Nanofluidics device**

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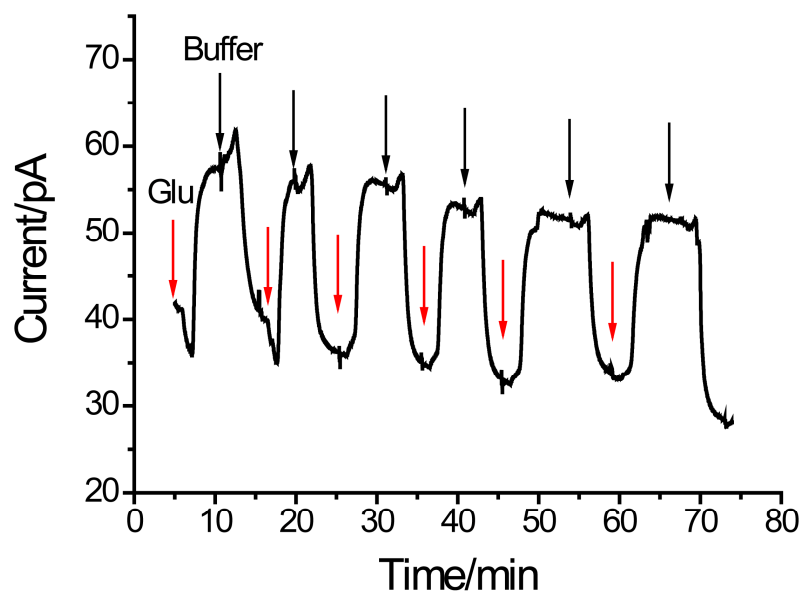


Fig. S 1. Current responses upon six times introduction of a 0.5 mM glucose solution through the concentrated enzymes (the glucose and buffer introduction points are indicated with arrows).

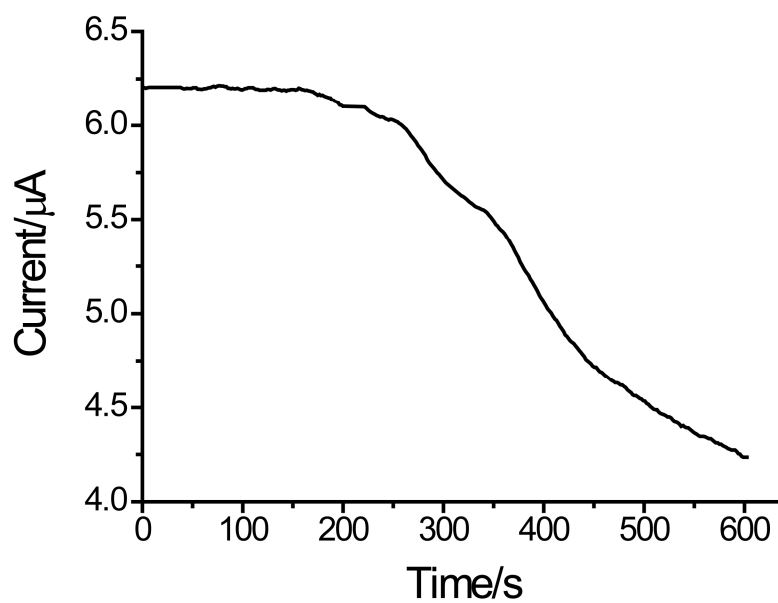


Fig. S 2. The change of separation current of high voltage power supply with the enzyme enrichment time.

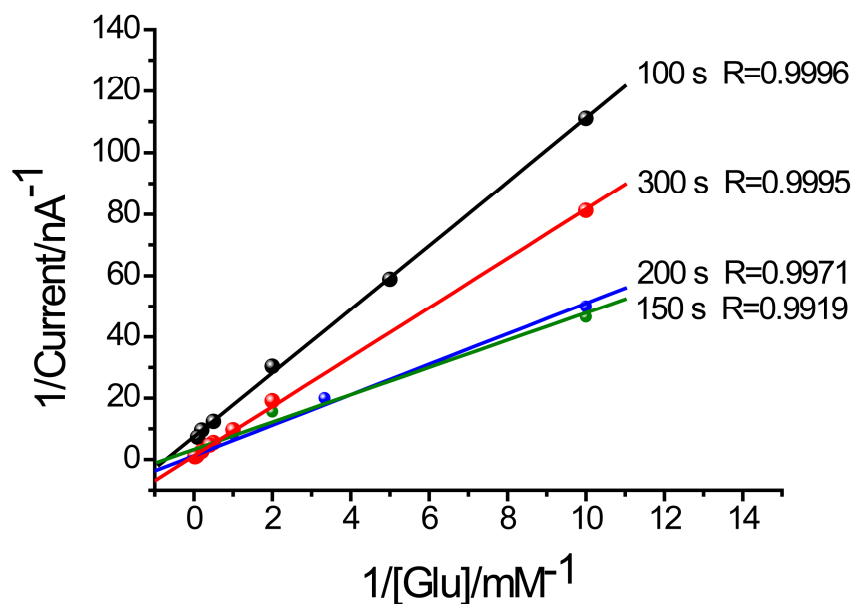


Fig. S 3. Double reciprocal plots of GOx activity versus glucose concentration for different enzyme concentration time.

Note S 1. Why a reduced fluidic transport velocity would result in reduced substrate concentration near the enzymes?

As shown by our results in Figure 8, the electrochemical response of hydrogen peroxide generated from the homogeneous enzymatic reaction increased with the enzyme enrichment time in the studied enrichment time region. This could help us to prove the reliability of our statement.

In addition, when the enrichment time was 300 s, the liquid flow rate was about 0.019 cm s⁻¹ as listed in Table 1. Assumed the enriched enzyme occupied 100 μm of the channel (estimated from our experimental results in Fig. 3A), it would take about 0.5 s for the glucose solution to flow through the enriched enzymes. If we used the reaction constant k for soluble GOx ($k=14 \pm 0.3 \text{ s}^{-1}$) which has been report previously⁴⁹, the glucose concentration c_t after passing the enriched enzyme region was equal to $9.12 \times 10^{-4} c_0$ (c_0 is the original concentration of glucose) for 0.5 s reaction time (t) according to equation (S1). Thus, we can conclude that for the enriched enzyme quantity studied, the substrate concentration does drop significantly after the substrate flows through the enzymes. This again confirms the statement made in our manuscript.

$$\ln \frac{c_0}{c_t} = kt \quad (\text{S } 1)$$