Supporting Information

Mechanism of an Enzymatic Reaction-Induced SERS Transformation for the Study of Enzyme-Molecule Interfacial Interactions

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Figure S1

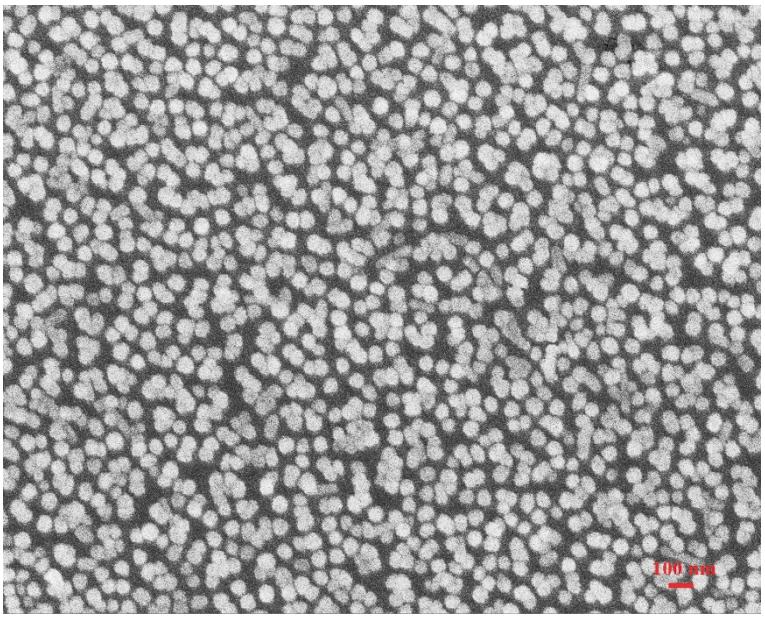

Figure S1. SEM image of the silver nanoparticle-decorated substrate. The surface of the glass slide was covered with 70 nm silver nanoparticles. The scale bar is 100 nm.

Study of the enzymatic reaction kinetics

Herein, the Michaelis-Menten model was used to account for the observed dependence of the SERS intensity on the glucose concentration. The model assumes the following reaction mechanism:[1-5]

$$E + S \rightleftarrows ES \rightarrow E + P$$

where E, S, ES and P are the enzyme, substrate (in this case, glucose), enzyme-substrate complex and product, respectively. The mechanism assumes that a quasi-equilibrium is established between the enzyme-substrate complex and the free enzyme and free complex, as implied by the double arrow. Subsequently, the enzyme-substrate complex concentration can be approximated by the following equation:

$$\frac{[E][S]}{[ES]} = K_M \qquad (1)$$

Note that in the equation $[E]=[E]_o-[ES]$, in which $[E]_o$ is the initial enzyme concentration, one can substitute for [E] in Eq. 1 to derive the following equation:

$$[ES] = \frac{[E]_o[S]}{K_M + [S]} \qquad (2)$$

The quantity proportional to the enzyme complex concentration [ES] was calculated using the decrease in the intensity of the 998 cm^{-1} band as a function of glucose concentration. During the reaction, it could be easily concluded from Fig. S3 that the glucose concentration [*S*] is inversely proportional to the SERS intensity of the band at 998 cm^{-1} (I_c). Therefore, [*S*] is proportional to I_o-I_c. Therefore, we obtain the

relationship $[ES] \propto \frac{I_o - I_c}{I_o}$, in which I_o is the SERS intensity of the 998 cm^{-1} band at zero glucose concentration and I_c is the SERS intensity of that band at concentration c. Accordingly, Eq. (2) can be rewritten as:

$$\frac{I_o - I_c}{I_o} = \frac{\sigma[E]_o[S]}{K_M + [S]} \qquad (3)$$

in which σ is a constant of proportionality (with dimensions M^{-1}). Fitting Eq. (3) to the data given in Fig. S3 produced the fit from which the value K_M = 8.2 x 10^{-6} M was derived.

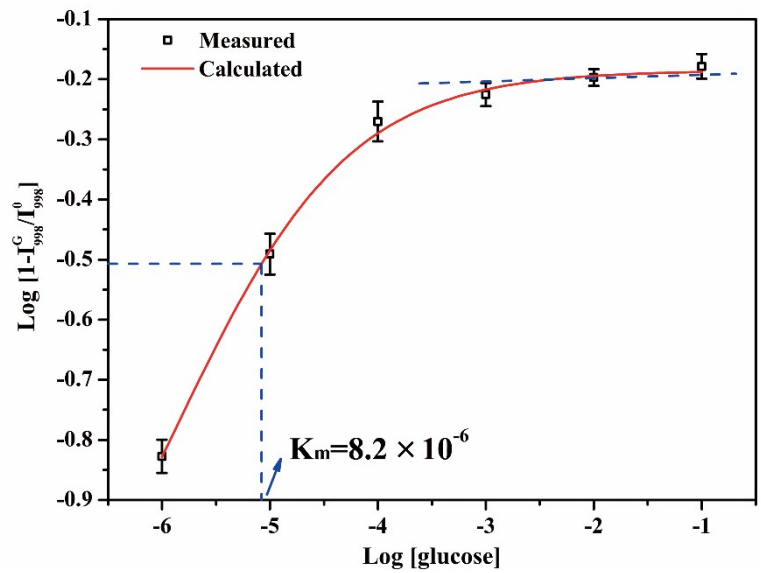

Figure S2. Study of the enzymatic reaction kinetics using the Michaelis-Menten model

Figure S3

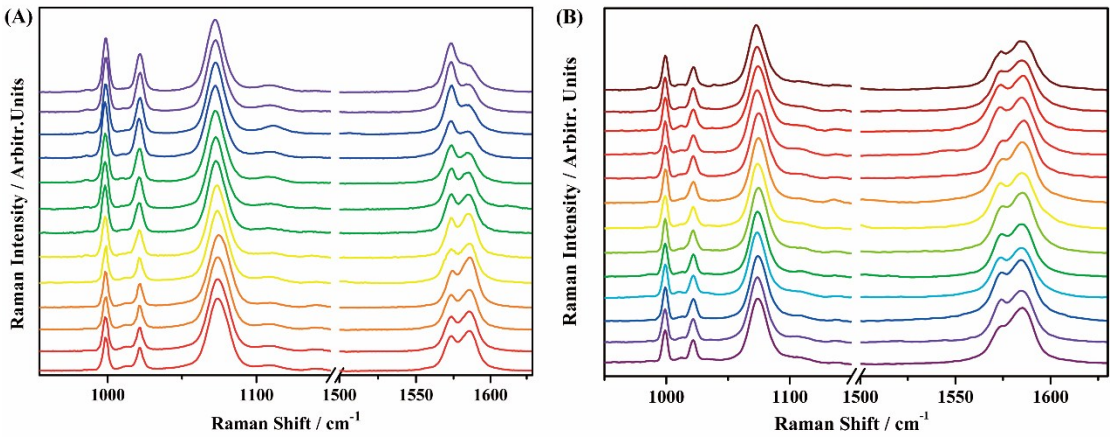

Figure S3. Normalized SERS spectra of MBA anchored to AgNP-coated glass slides upon exposure to a 10^{-6} M glucose solution with (B) and without (A) a deoxygenation process with increasing immersion time.

Figure S4

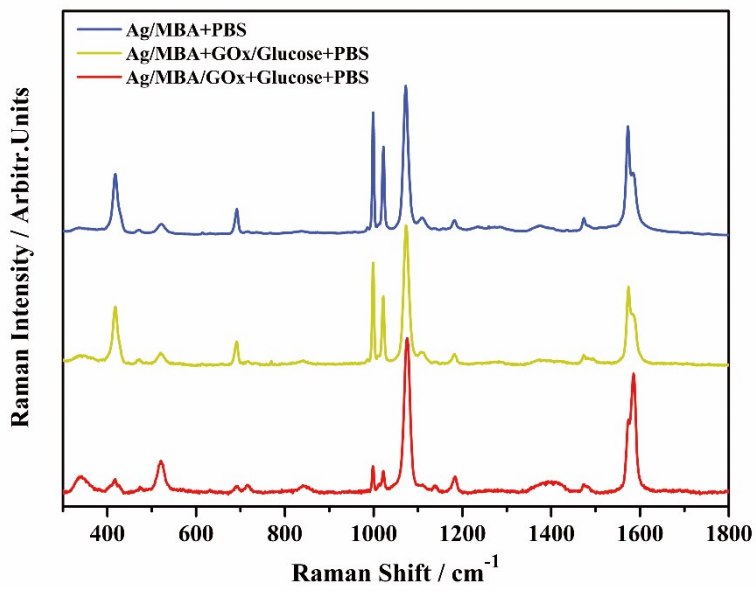

Figure S4. The SERS spectra of Ag/MBA with (a) immersion of the Ag/MBA complex in a mixed solution of Gox and glucose and (b) immersion of the Ag/MBA/Gox complex in a glucose solution (c).

Figure S5

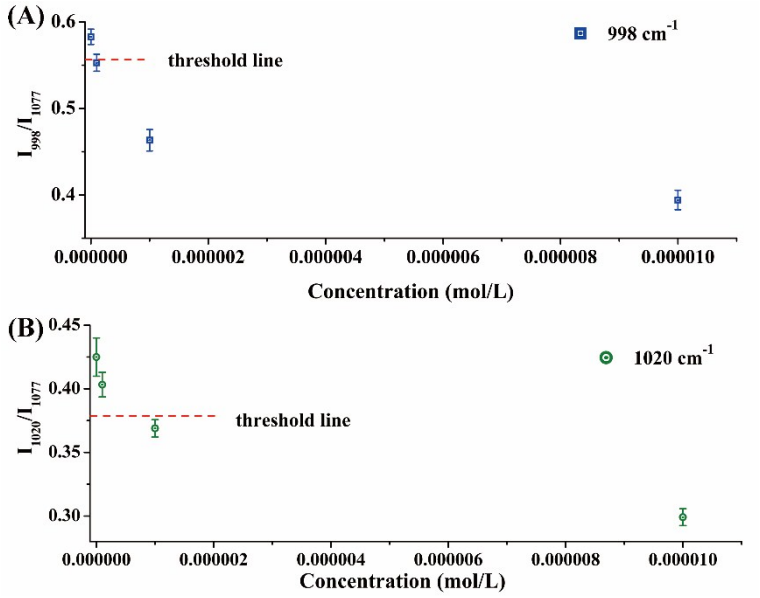

Figure S5. The SERS relative intensity ratio of the bands I_{998}/I_{1077} (A) and I_{1020}/I_{1077} (B) as a function of the glucose concentration, where the threshold line indicates the position 3 standard deviations from the value for the blank sample.

References

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