## **Supporting Information**

## In situ assembly, regeneration and plasmonic immunosensing of a Au nanorod monolayer in a closed-surface flow channel

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Figure S1. An SEM image of the AuNRs used in this work.

Before collecting the SEM image, the AuNRs were chemically adsorbed onto a silicon wafer and washed with piranha solution to clean the surface and to introduce a dense layer of negative charges at the wafer surface.



Figure S2. Photographs of the darkfield microscope system (a) and the microfluidic system (b) used in this study



Figure S3 The AuNR retention with increasing regeneration time.

Conditions for AuNR generation: AuNR suspension concentration is  $0.033 \times C_0$  (C<sub>0</sub> is the original AuNR concentration in solution (as synthesized)); CTAB concentration is  $2.7 \times 10^{-3}$  M; the incubation time is 5 min.

Conditions for regeneration: A solution containing 10 times diluted aqua regia (concentrated HNO<sub>3</sub>: concentrated HCl:  $H_2O = 1$ : 3: 36 (v/v)) was injected into the flow cell and incubated for 1 min, followed by distilled water to rinse, and subsequently 2.0 M NaOH solution (incubate for 10 min) to reactivate the flow channel. The error bars represent standard deviations of 3 replicates.



Figure S4. The density of the immobilized AuNRs on the flow channel (evaluated as particle number per cm<sup>2</sup>) as a function of (a) CTAB concentration, (b) AuNR suspension concentration and (c) incubation time. Experimental conditions: (a) AuNR suspension concentration =  $0.033 \times C_0$  (C<sub>0</sub> = original AuNR concentration in solution (as synthesized)) and the incubation time = 5 min; (b) CTAB concentration =  $2.7 \times 10^{-3}$  M and the incubation time = 5 min; (c) CTAB concentration =  $2.7 \times 10^{-3}$  M and the AuNR concentration =  $0.033 \times C_0$ . The error bars represent standard deviations of five replicates.



Figure S5. The density controlled assembly of the AuNR monolayer inside the closed-surface, microfluidic channel with a flow channel of 45  $\mu$ m (w) × 15  $\mu$ m (h). (a) A blank (before AuNR fabrication); (b-d) a AuNR monolayer fabricated by the injection of a 50 times diluted AuNR solution (in deionized water) into the flow channel with an incubation time of 10 s (b), 1 min (c) and 30 min (d). The spots in red or orange represent the immobilized AuNRs (The scattering spots of AuNRs are not clear primarily due to the high background noise arising from the rough glass surface). The same microfluidic chip was used for images (a-d). The scale bar is 10  $\mu$ m.

