

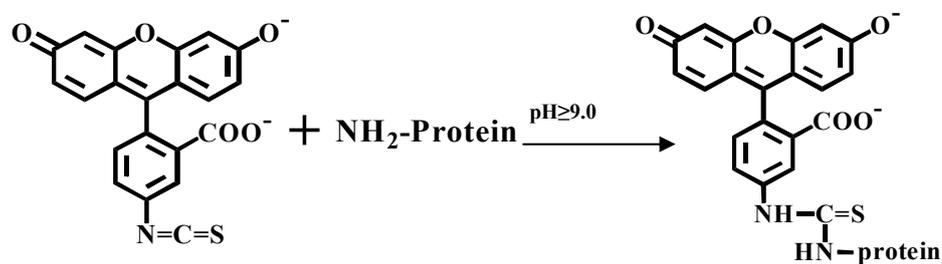
## Supporting information.

### Rapid protein concentration, efficient fluorescence labeling and purification on a micro/nanofluidics chip

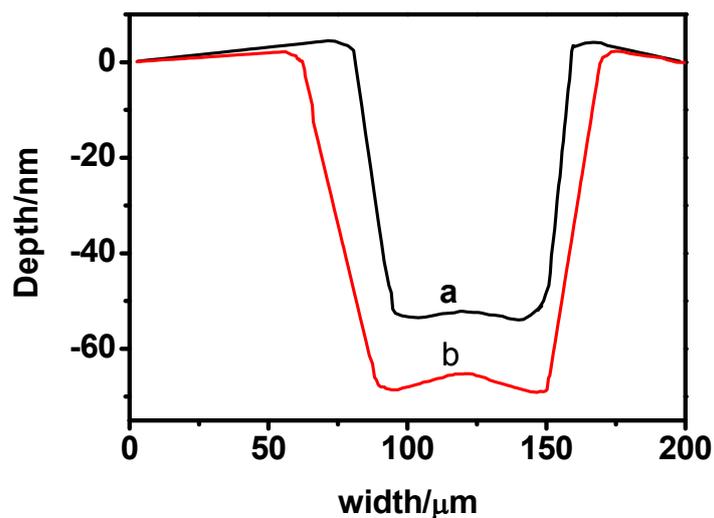
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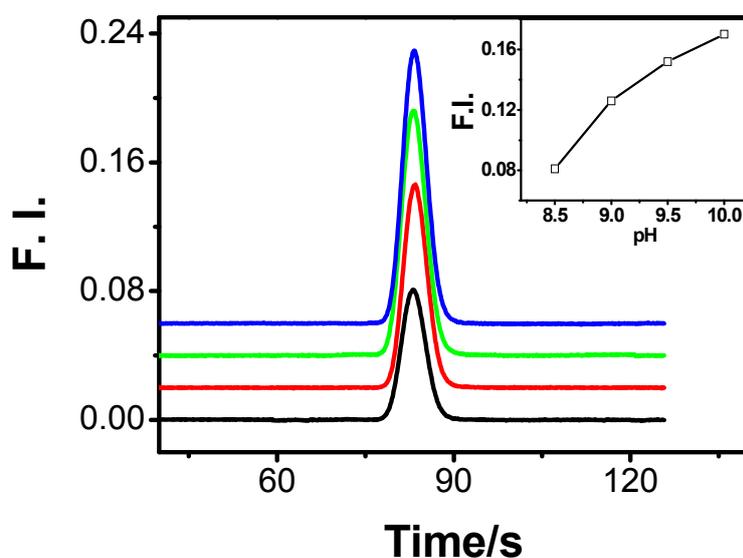
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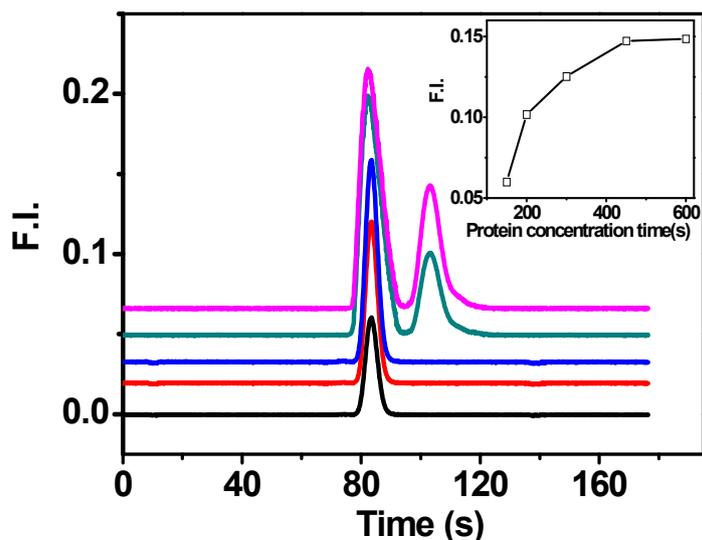
**Scheme S1.** Reaction scheme of FITC with amino residues of protein side chains. Left: FITC; green fluorescence. Right: FITC-protein conjugate; green fluorescence.



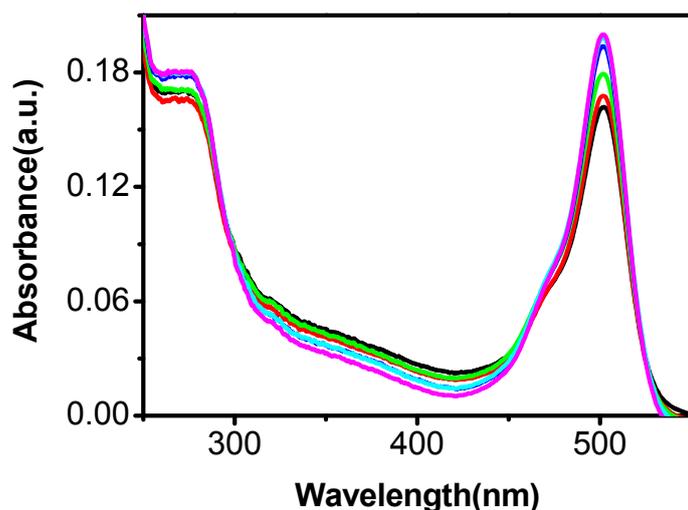
**Figure S1.** Depth profiles of the fabricated nanochannels on PC surfaces after 60-min (a) and 80-min (b) UV-irradiation through a photomask.



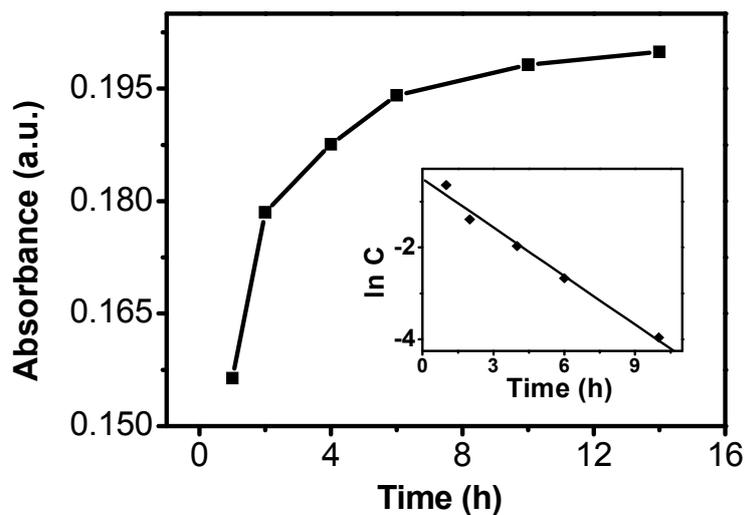
**Figure S2.** Micro/nanofluidics chip electropherogram of labeling product FITC-BSA after 300 s concentration, 600 s FITC flowing and 90 s purification processes in solutions with pH value of 8.5, 9.0, 9.5 and 10.0 (from bottom to top). The inset shows the influence of solution pH on labeling reaction efficiency.



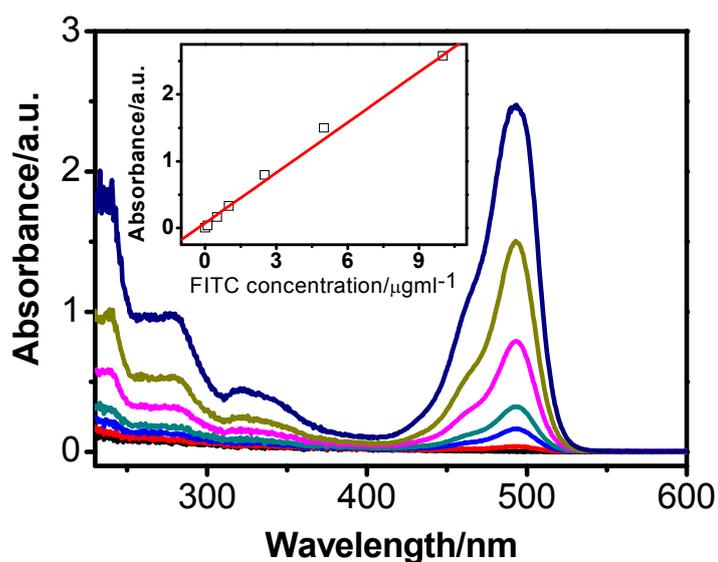
**Figure S3.** Typical electropherograms of the purified protein with different BSA concentration time. Concentration time (from bottom to top): 150, 200, 300, 450, and 600 s. The FITC flowing time: 600 s. The FITC-BSA peak appears at 83 s, and FITC at 103 s. The inset shows the plot of FITC-BSA fluorescence intensity versus protein concentration time. Reaction conditions: 100  $\mu\text{g/ml}$  BSA (1.5 nM), 6  $\mu\text{g/ml}$  FITC solution (15 nM) in 20 mM BCB solution (pH 9.0). The separation voltage was 1000 V.



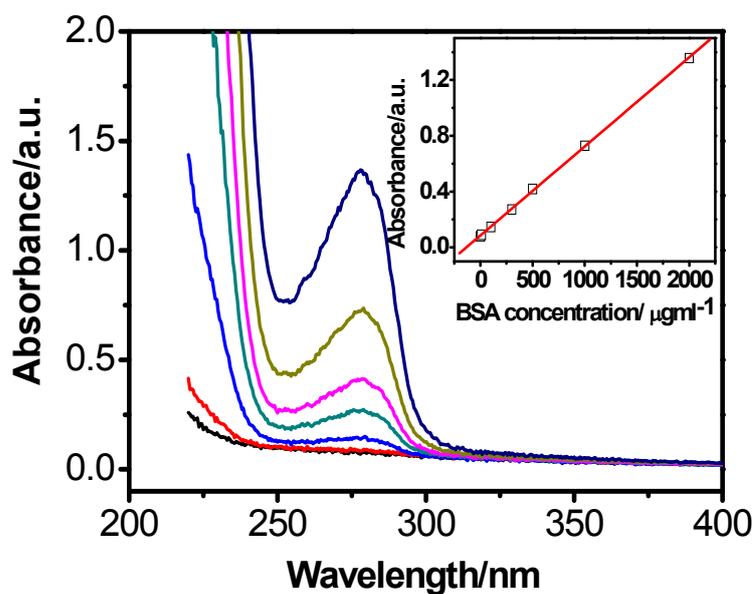
**Figure S4.** The reaction kinetics of BSA labeled with FITC in bath system. From bottom to top are 1 h, 2 h, 4 h, 6 h, 10 h, 14 h reaction time in the dark. Other reaction conditions: pH 9.0, temperature 25  $^{\circ}\text{C}$ , BSA concentration 100  $\mu\text{g/ml}$ .



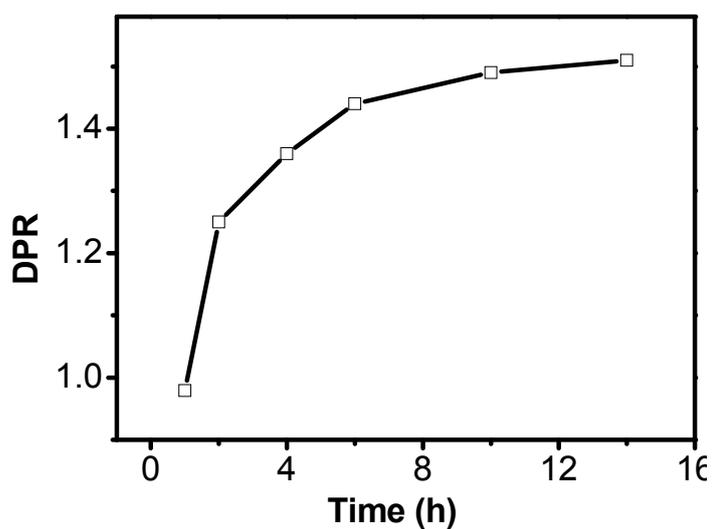
**Figure S5.** UV-vis absorbance intensity of the purified FITC-BSA product obtained from different labeling reaction time in bath system in the dark. The reaction was performed in a pH 9.0 buffer solution at 25°C. BSA concentration: 100  $\mu\text{g/ml}$ . The inset shows the plot of the natural Logarithm of protein (BSA) concentration (lnC) as a function of reaction time (t).



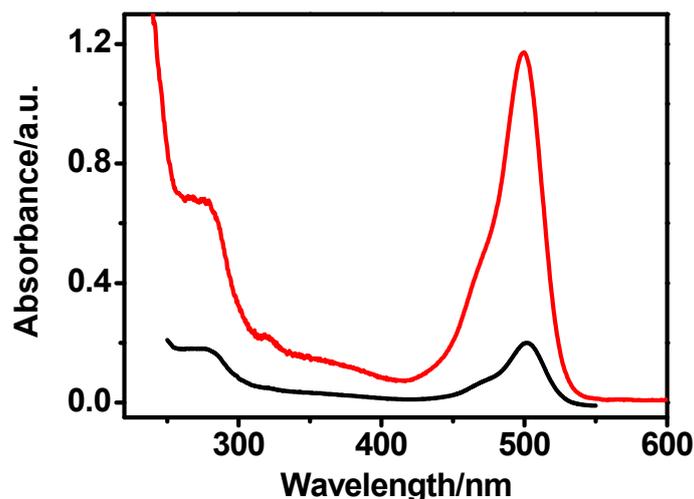
**Figure S6.** UV-vis spectra of FITC solutions with different concentration (from bottom to top: 0, 0.01, 0.05, 1, 2.5, 5, 10  $\mu\text{g/ml}$ ). The inset displays the calibration curve of FITC.



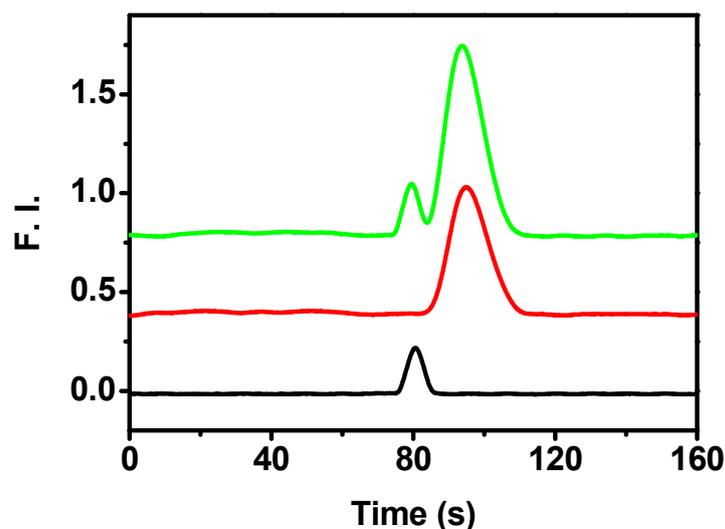
**Figure S7.** UV-vis spectra of BSA solutions with different concentration (from bottom to top: 0, 10, 100, 300, 500, 1000, 2000 $\mu\text{g/ml}$ ). The inset shows the calibration plot of BSA.



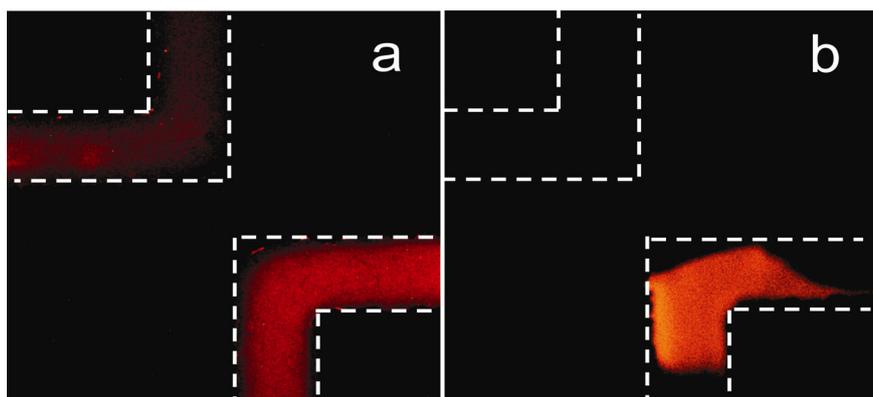
**Figure S8.** Plot of DPR values of FITC-BSA versus reaction time. Data were from Figure S4.



**Figure S9.** UV-vis absorbance spectra of the reaction product FITC-BSA synthesized from conventional bath system (black curve) and our micro/nanofluidics device (red curve).



**Figure S10.** Electropherograms of IgG labeled with FITC on the micro/nanofluidics chip. Curves from bottom to top correspond to the purified product FITC-IgG, FITC, and unpurified reaction product (including FITC-IgG and unreacted FITC), respectively. Reaction conditions: 239  $\mu\text{g/ml}$  IgG, 12  $\mu\text{g/ml}$  FITC in 20 mM BCB solution (pH 9.0). The IgG concentration time: 300 s at 400 V, the reaction time at room temperature (FITC flowing time): 600 s, followed by purification time of 90 s. Separation voltage: 1000 V.



**Figure S11.** The photo images showing purification of the formed RBITC-BSA conjugates on the micro/nanofluidic device. Images were taken after buffer instead of RBITC solution was electrokinetically driven through the nanochannel for 30 s (a) and 90 s (b). Separation voltage: 1000 V applied at reservoirs 3 (anode) and 6 (cathode) as indicated in Figure 1A. Dotted lines indicate the outlines of the microchannels.