## Enhanced biosensing of tumor necrosis factor-alpha based on aptamer-functionalized surface plasmon resonance substrate and Goos-Hänchen shift

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## Supplementary Figures

- Fig. S1 Validation of binding between BSA and aptamer by isothermal titration calorimetry.
- Fig. S2 Sensorgrams of GH-aptasensing of BSA.
- Fig. S3 Sensorgrams of GH-aptasensing of TNF- $\alpha$ .
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**Fig. S1** Validation of binding between BSA and aptamer by isothermal titration calorimetry. Representative ITC data for BSA-specific aptamer (6.5  $\mu$ M) and BSA (100  $\mu$ M) at 25 °C. Top panel: the raw baseline-smoothed ITC data plotted as heat flow versus time, and the bottom panel shows the integrated and concentration-normalized heat for each injection versus the BSA-specific aptamer molar ratio with a nonlinear analysis of the data with the independent binding model gives the best-fit values.



**Fig. S2** Sensorgrams of aptamer-based GH-sensing of BSA. BSA-specific aptamer functionalization and blocking with thiol-PEG molecules for sensing of (**a**) 10 aM to 1  $\mu$ M BSA concentrations and (**b**) 1 aM to 1 nM BSA. (**c**) Time-course of sequential injection of titrated BSA solutions from 10 aM to 1  $\mu$ M (excluding intermediate running buffer washes). The data points within the 0-1 minute range are based on the average of 50 measurements, while the rest are derived from the average of 100 measurements. (**d**) Time-course of serial-diluted BSA injections from 1 aM to 1 nM (excluding intermediate running buffer washes). Averages for data points within the 0-1 minute timeframe are based on 50 measurements, while those beyond 1 minute are derived from an average of 100 measurements.



**Fig. S3** Sensorgrams of aptamer-based GH-sensing of TNF- $\alpha$ . (a) TNF- $\alpha$ -specific aptamer functionalization and PEG-blocking. (b) Sequential GH-aptasensing of various TNF- $\alpha$  concentrations (1 fM, 1 pM, 1 nM, and 1  $\mu$ M). The data points in the 0-1 minute range are averages from 50 measurements, whereas the remaining points are averages from 100 measurements.



**Fig. S4** Sensorgrams of TNF- $\alpha$  GH-immunosensing. (a) Antibody-functionalization and PEG-blocking of the Ausensing surface. (b) Sequential injections of TNF- $\alpha$  and binding to antibody on Au surface after establishing the running buffer baseline. Data are depicted as averages in (b); for data points in the 0-1 minute segment, they are calculated from 50 measurements, while for the remaining data points they are derived from 100 measurements.