Electronic Supplementary Information

A fluorescent molecularly imprinted device for on-line analysing AFP in human serum

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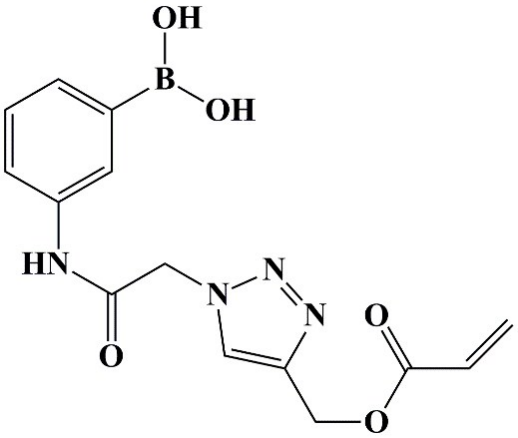

Fig. S1 Molecular structure of APBA-PA.

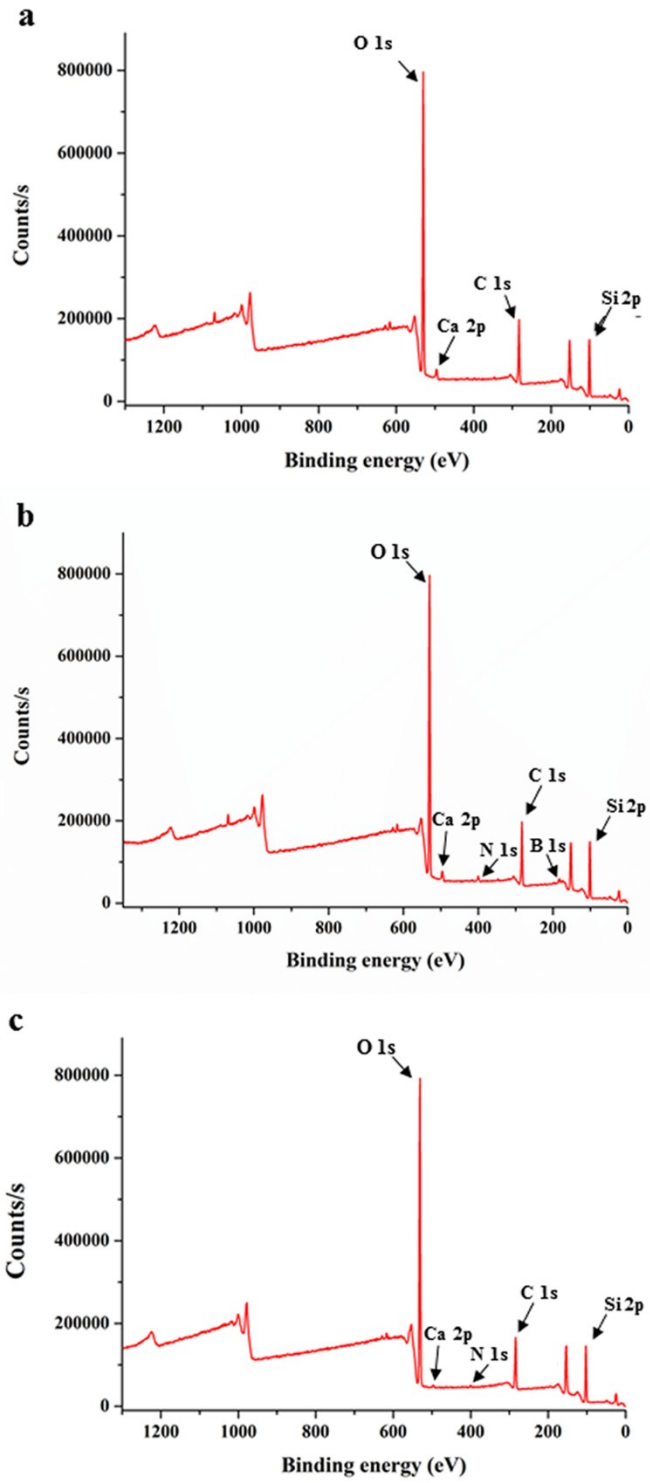

Fig. S2 XPS spectra of (a) the original microscope slide, (b) ABPA-PA modified microscope slide and (c) the fluorescent molecularly imprinted device.

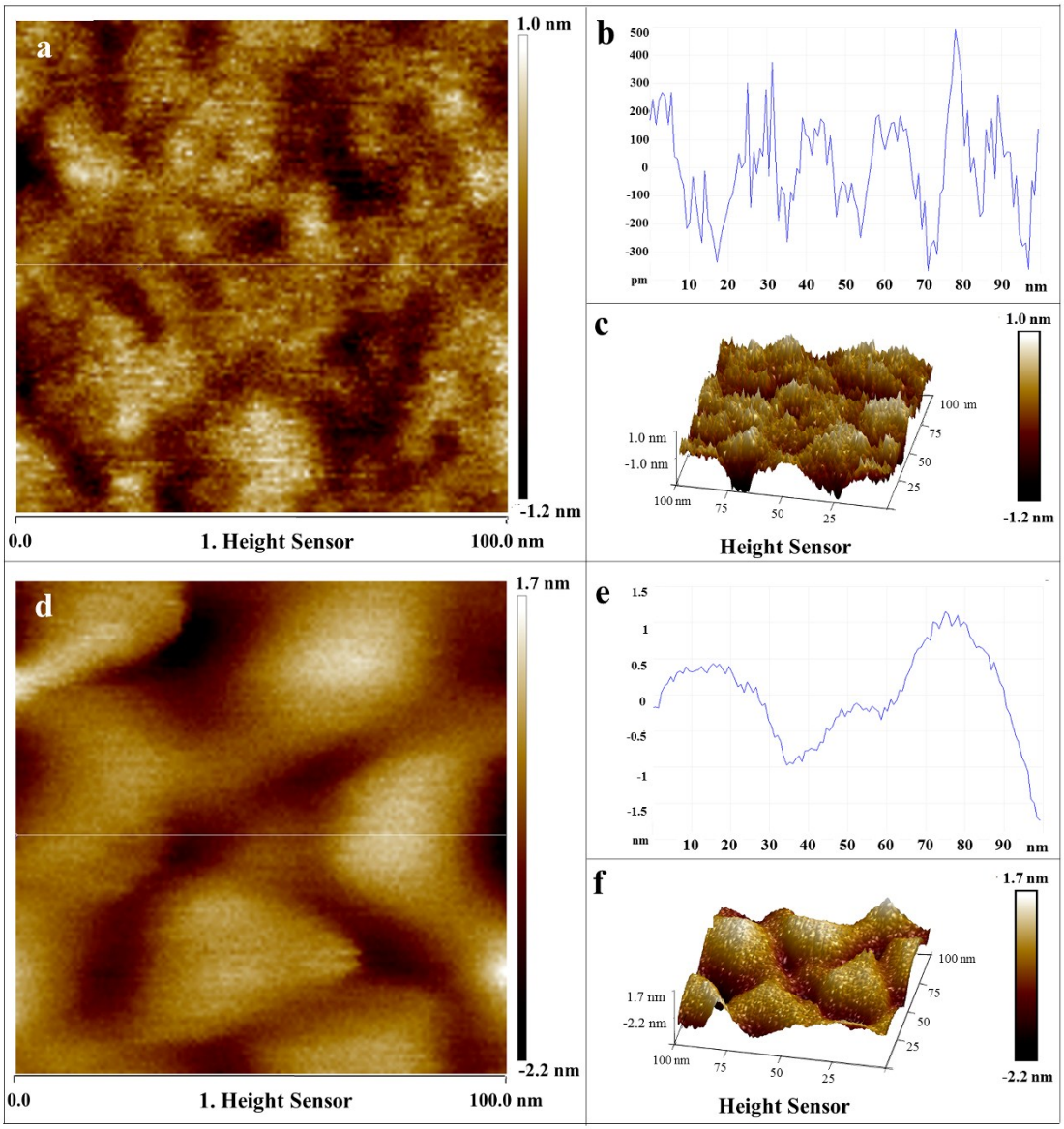

Fig.S3 (a) AFM image of the carbon-carbon bonds functionalized microscope slide; (b) information of the tangential line in Fig.S3a; (c) 3D morphology of the carbon-carbon bonds functionalized microscope slide; (d) AFM image of the APBA-PA modified microscope slide; (e) information of the tangential line in Fig.S3d; (f) 3D morphology of the APBA-PA modified microscope slide.

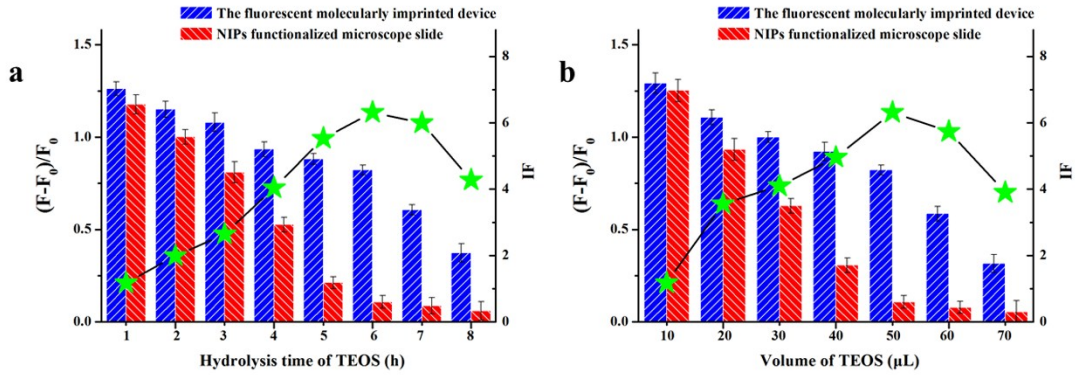

Fig. S4 Effects of imprinting conditions

Optimization of the imprinting conditions

The imprinting conditions, including the hydrolysis time and the volume of TEOS, were optimized to prepare the AFP analysis device with best performance by simultaneously considering the fluorescence response ability and the imprinting factor. As can be seen from Fig. S4, the fluorescence response ability of the device decreased with the increasing hydrolysis time of TEOS, due to the prepared SiO_2 occupied the binging sites of the APBA-PA for the template. However, bared APBA-PA modified microscope slide showed limited imprinting factor for its lack of imprinted cavities. When the AFP-MIPs gradually produced in this process, the size-matching effect of the imprinted cavities endow the device with appropriate selectivity. Furthermore, the imprinting factor started to reduce when the hydrolysis time was longer than 6 h, which could be attributed to the damage of the excessive SiO_2 to the imprinted cavities. Thus, 6 h was chosen to the optimal hydrolysis time of TEOS.

The volume of TEOS decided the concentration of the solution, thus it was also be studied in this work. As Fig. S4b showed, the fluorescence response ability of the device decreased with the increasing volume of TEOS from 10 μL to 50 μL, while the imprinting factor increased in this process. When the volume of TEOS was greater than 50 μL, both the fluorescence response ability and the imprinting factor decreased seriously, thus 50 μL was chosen to be the optimal volume of TEOS.

To sum up, the optimal imprinting conditions were found to be 30 mL PB solution (20 mM, pH 7.4) containing 50 μL of TEOS with a hydrolysis time of 6 h.

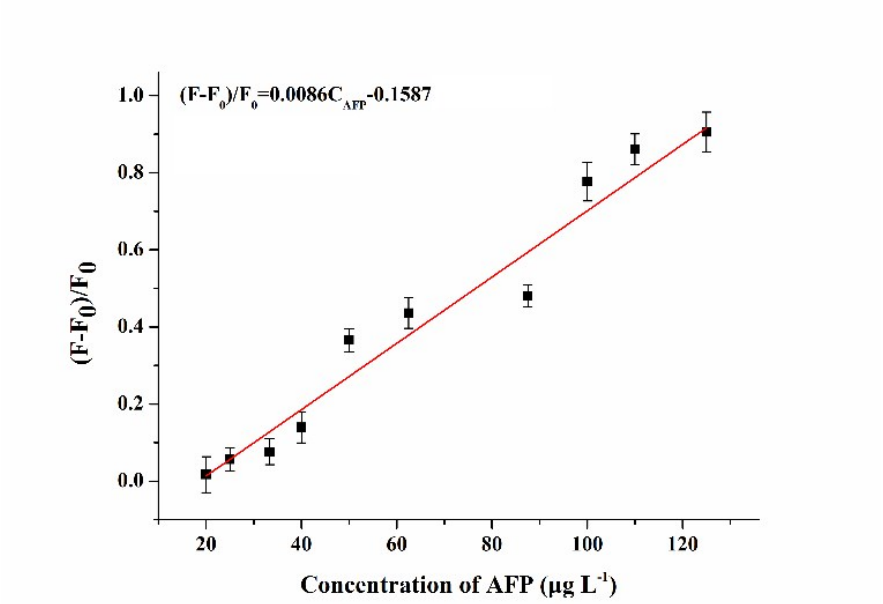

Fig. S5 Fluorescence responses of the fluorescent molecularly imprinted device with increasing concentrations of AFP.

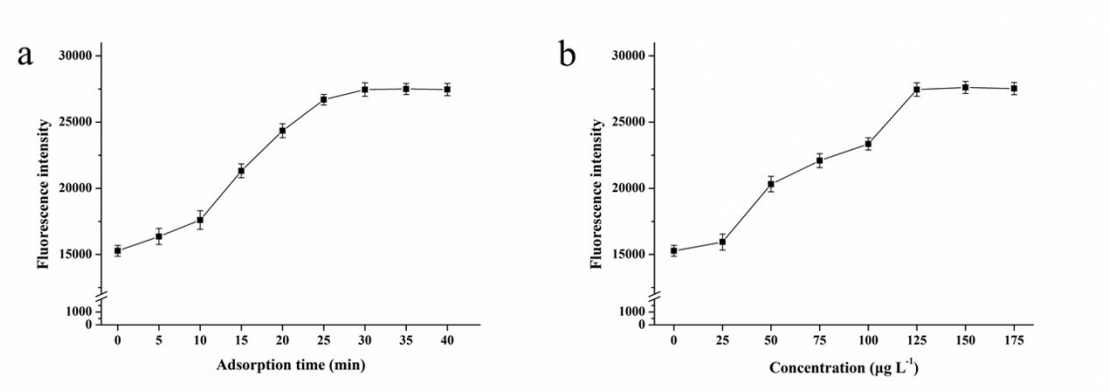

Fig. S6 Adsorption performance of the fluorescent molecularly imprinted device towards AFP (a) adsorption isotherms; (b) rebinding rate

Adsorption efficiency study of the device

The adsorption efficiency of MMINs was comprehensive evaluated by investigating the adsorption capacity and analysing the rate of adsorption equilibrium. Fig. S6 exhibited the adsorption isotherms of the device at different equilibrium concentrations. It is obviously that the fluorescence intensity of the device increased along with the increasing of AFP concentration until they reached the saturation level at 125 μg L^{-1}. Thus, 125 μg L^{-1} was identified as the experimental concentration of AFP in the following experiments.

To gain further understand on the adsorption performance and binding mechanism of the device towards AFP, the rate of adsorption equilibrium was studied. As Fig. S6b showed, 125 μg L^{-1} AFP binding in all case reached the adsorption plateau within 30 min, thus 30 min was chosen to be the equilibrium time of adsorption of the device towards AFP in the following experiments.

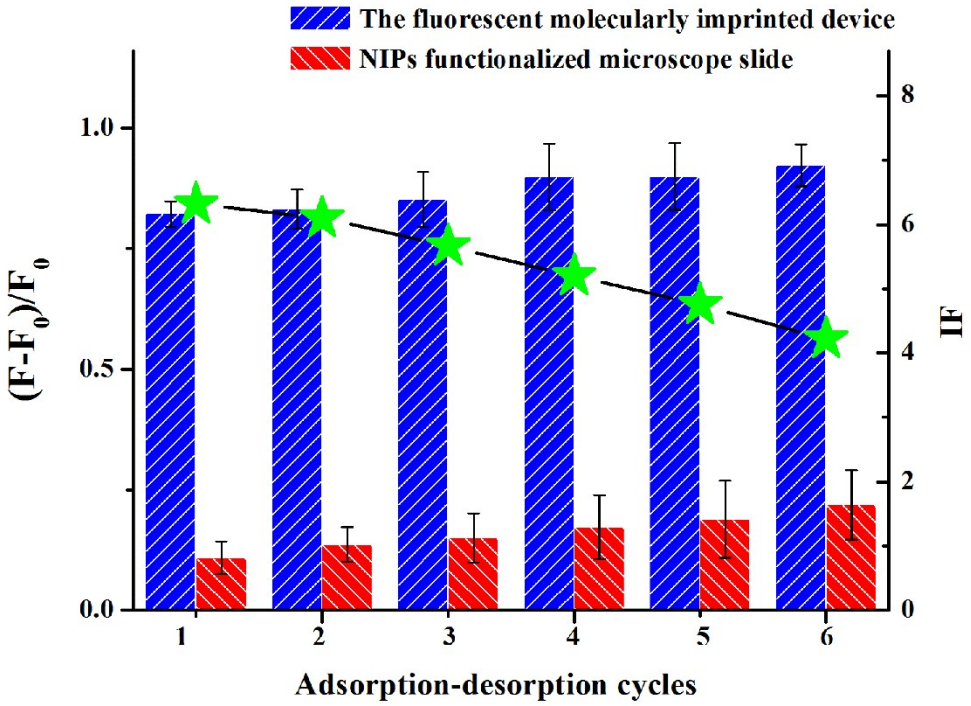

Fig. S7 Reusability studies of the fluorescent molecularly imprinted device and the NIPs functionalized microscope slide.

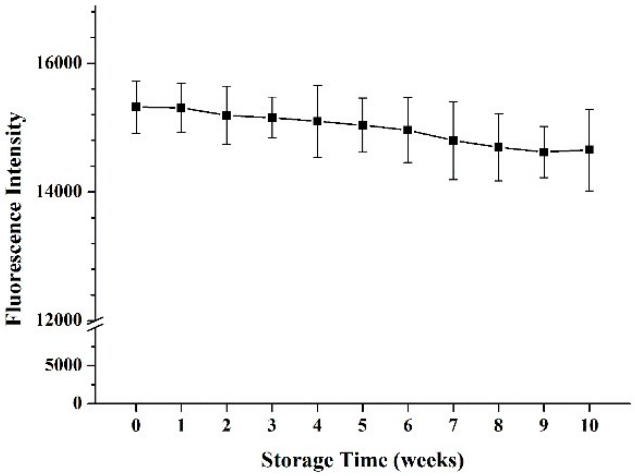

Fig. S8 Storage stability of the fluorescent molecularly imprinted device.

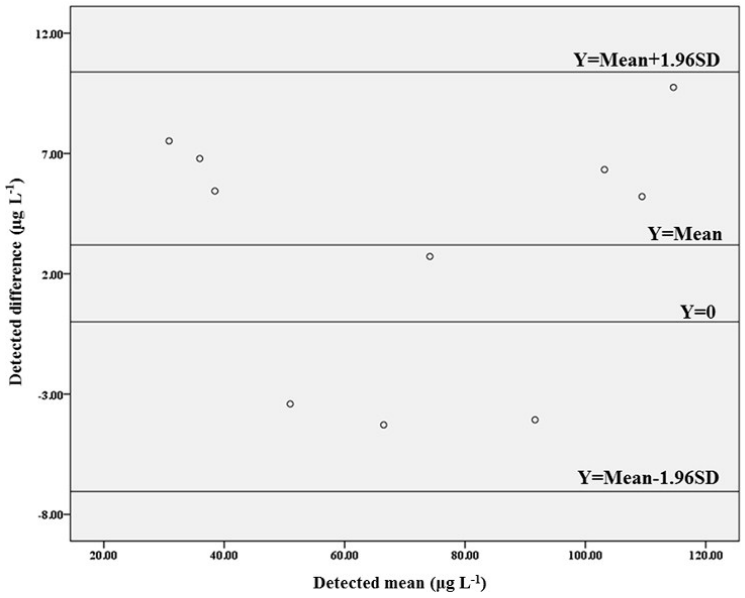

Fig. S9. Bland-Altman plots for detecting AFP concentration by ELISA and this method.

Table S1 Accuracy of the method for spiked at different levels (*n*=5).

NO.	Add (μg L^{-1})	Found (μg L^{-1})	RSD (%)	Recovery rate (%)
1	50.0	44.0	9.0	88.1
2	80.0	71.7	7.7	89.7
3	120.0	102.6	11.0	85.5

Table S2. The information of the tested proteins

	Molecular Weight (kDa)	Isoelectric point	Size (nm^3)	Ref.
AFP	69	4.75	5×5×5	1
HRP	40	7.2	4.0×6.7×11.7	2
OVA	45	4.7	7.0×4.5×5.0	2
IgG	150	8.0	14×9×4	3
BSA	66	4.8	14×4×4	4
HSA	67	4.8	12×2.7×2.7	5

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