

Supplementary material

Capillary electrophoresis-laser-induced fluorescence (CE-LIF)-based immunoassay for quantifying antibodies against cyclic citrullinated peptides

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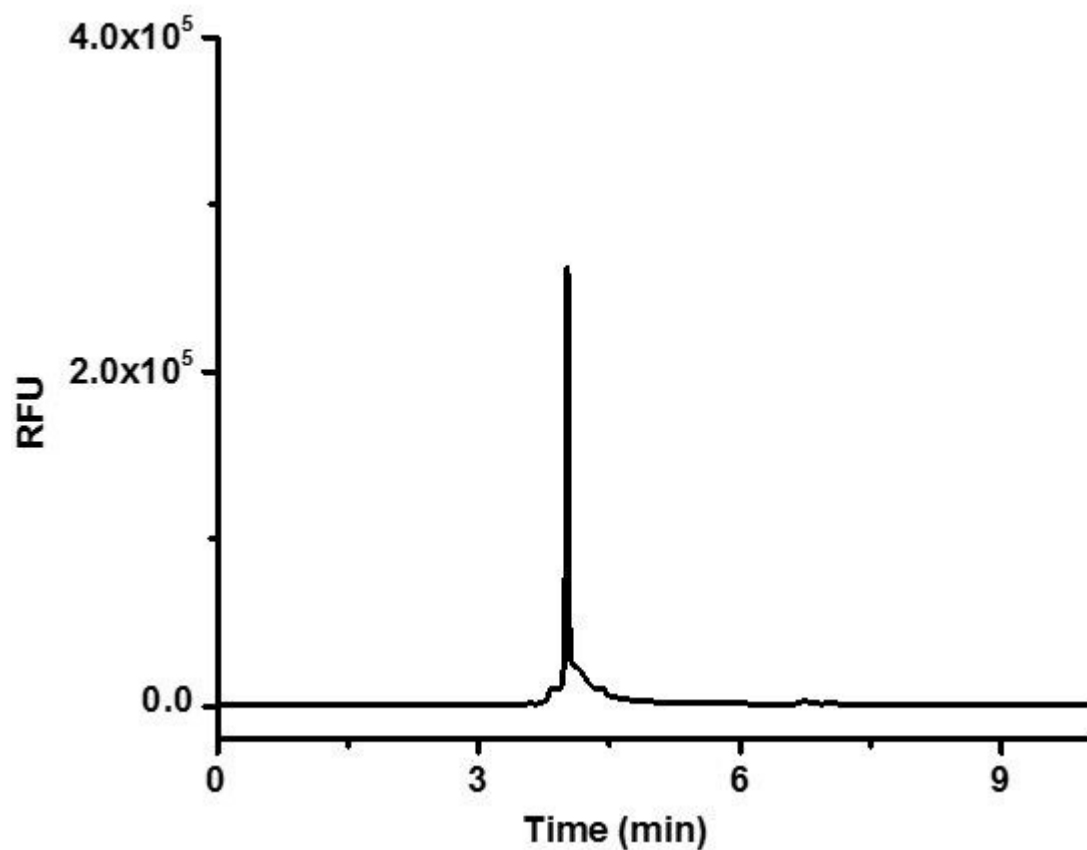
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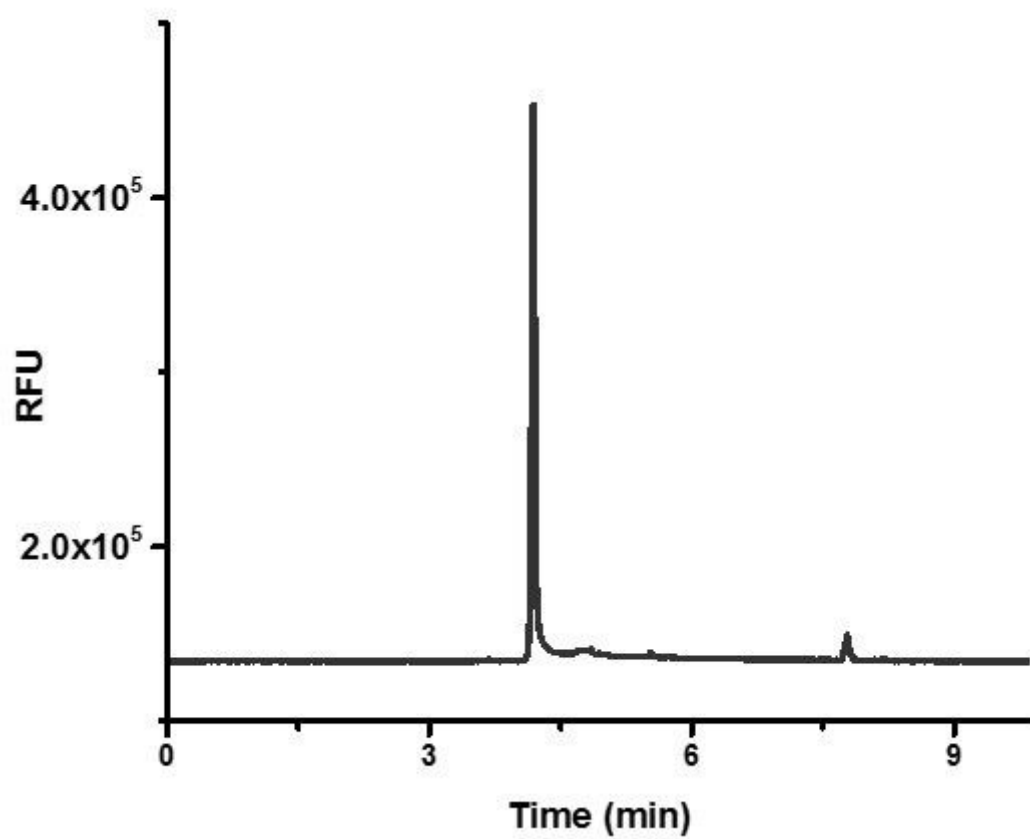
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Fig S1. Electropherograms with different buffers and pH (a). F-CCP using 50 mM CAPS pH 9.3, (b) F-CCP using 50 mM Sodium borate pH 9.3 , (c) F-CCP using 50 mM sodium borate pH 10.0, (d) F-CCP using 100 mM sodium borate pH 9.3, (e) F-CCP using 25 mM sodium borate pH 9.3

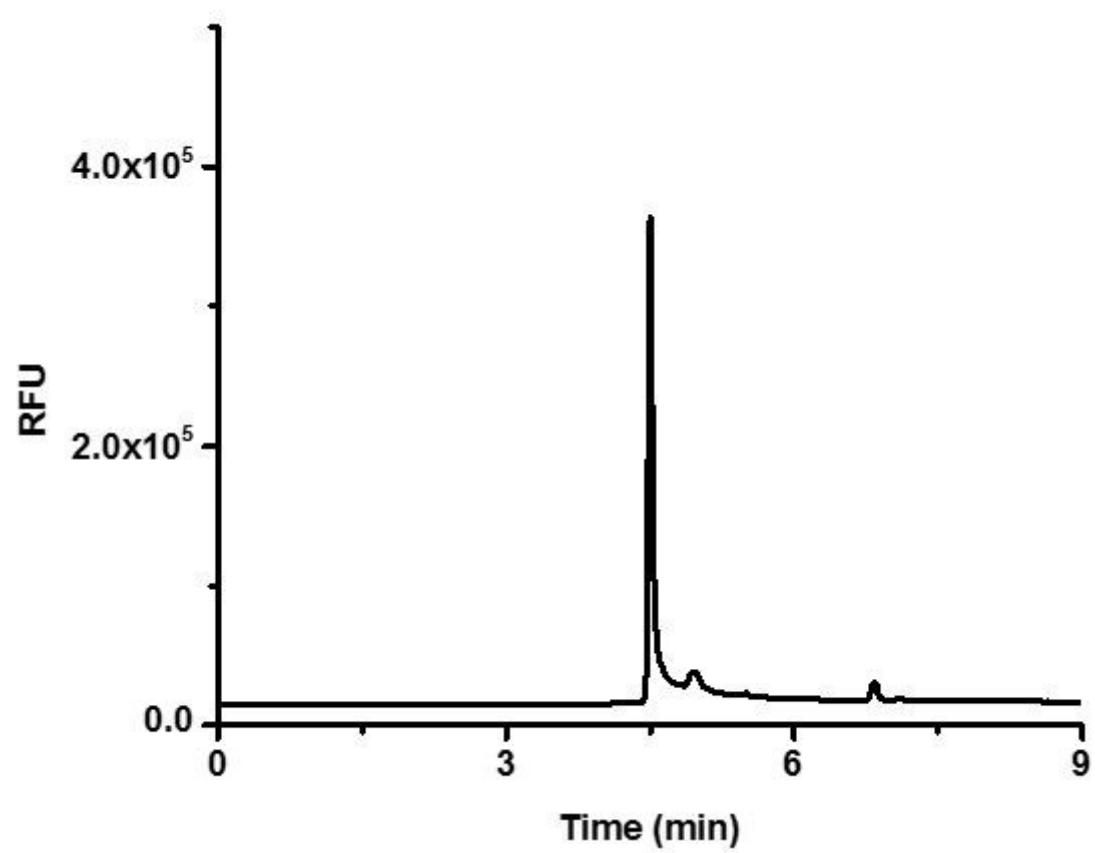
(a)



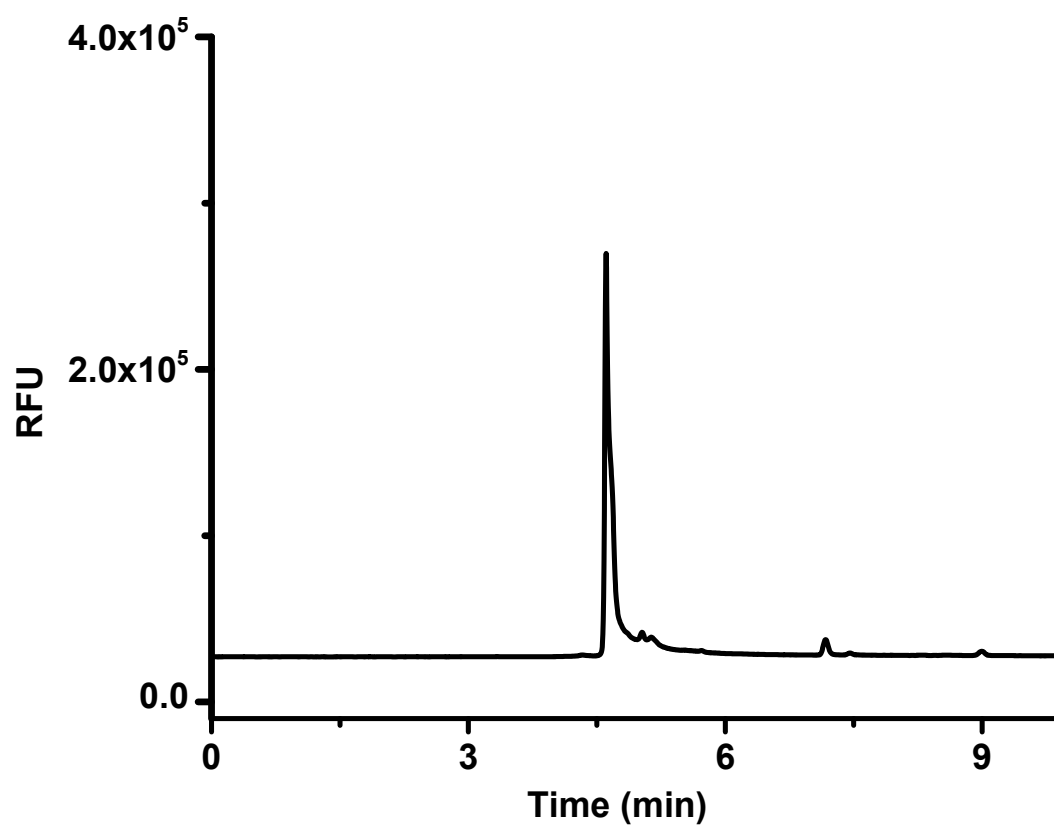
(b)



(c)



(d)



(e)

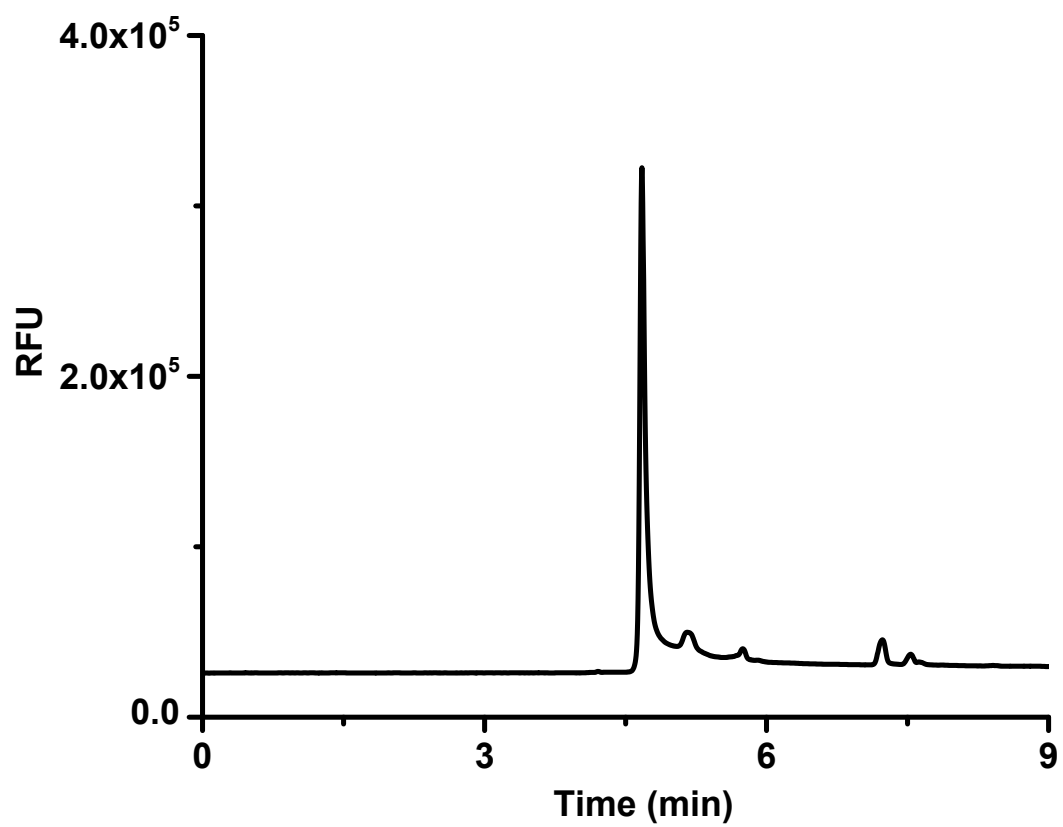


Fig S2. The peak profiles at LOD of F-CCP11A (4 ng/mL) and F-CCP (2 ng/mL)

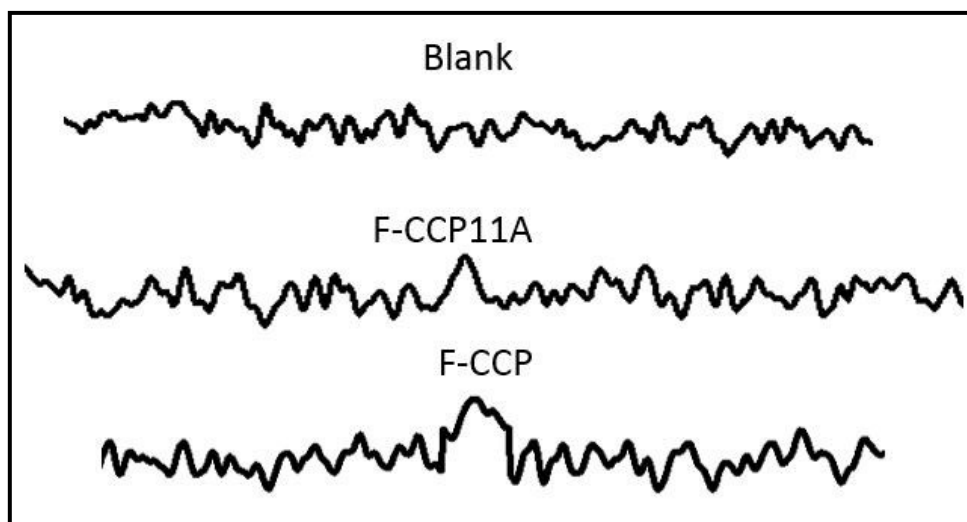
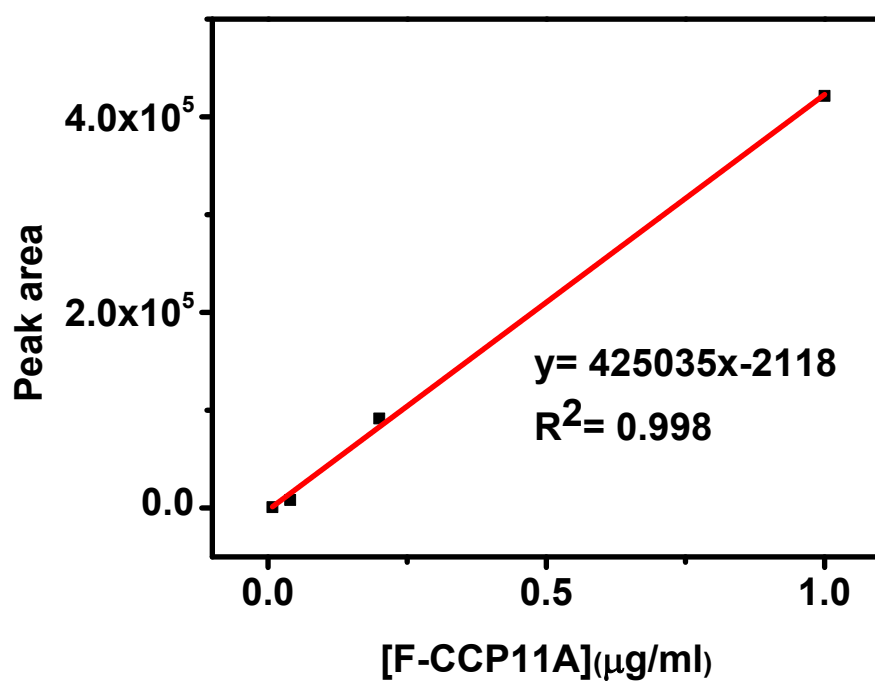


Fig S3. Standard curves plotted between peak area vs. concentration of a range from 0.008 – 1.0 $\mu\text{g/mL}$ (a) F-CCP11A and (b) F-CCP.

(a)



(b)

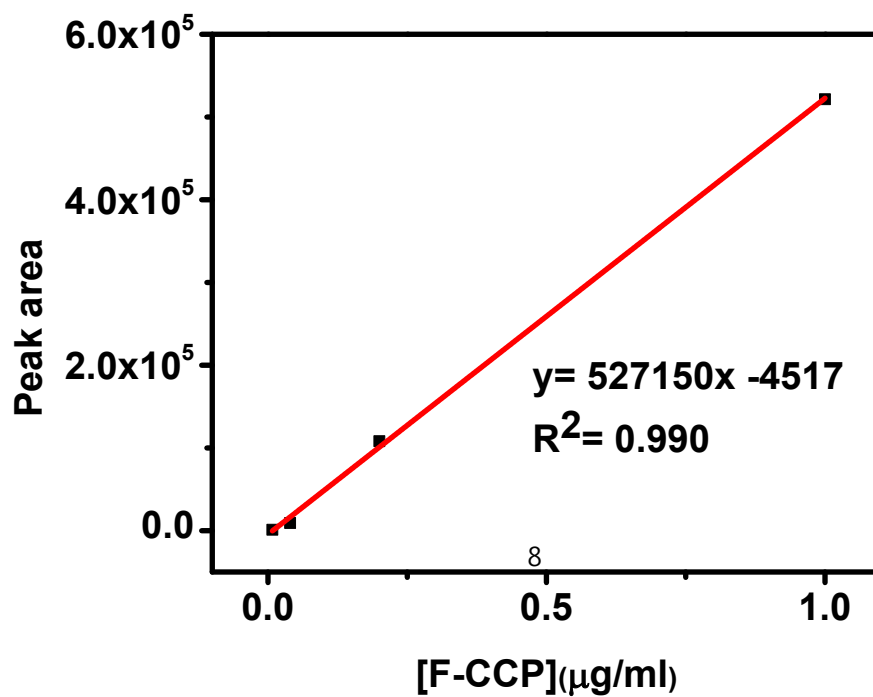
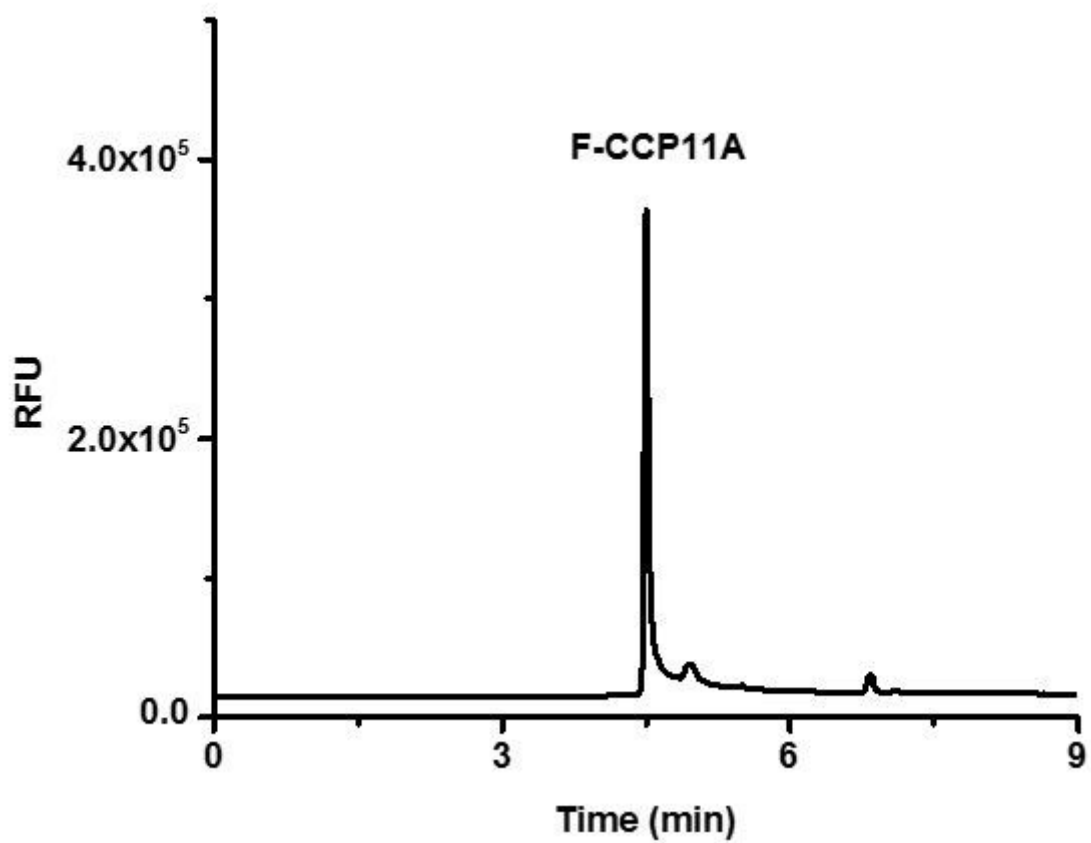


Fig S4. Electropherograms of immuno-reaction at 4°C, overnight. No immuno peaks were observed.

(a) F-CCP11A, (b) F-CCP

(a)



(b)

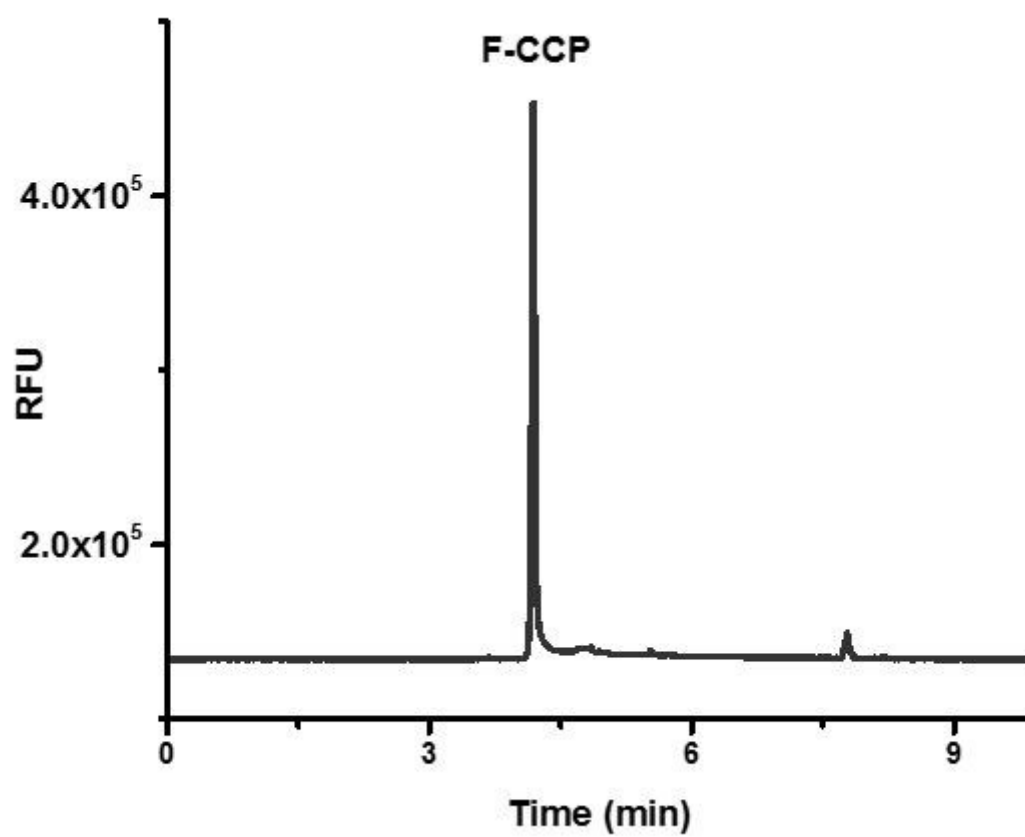


Fig S5. Electropherograms after immunoreaction at 37°C between CCP peptides and anti-CCP antibodies. A range of antibody concentrations varied from 0.25-16 µg/mL was treated with 0.5 µg/ml CCP peptides, incubated at 37°C, 12 h. (a) anti-CCP was diluted in PBS, (b) CCP peptides were treated with human IgG serum without the presence of anti-CCP, (c) anti-CCP was diluted in FBS.

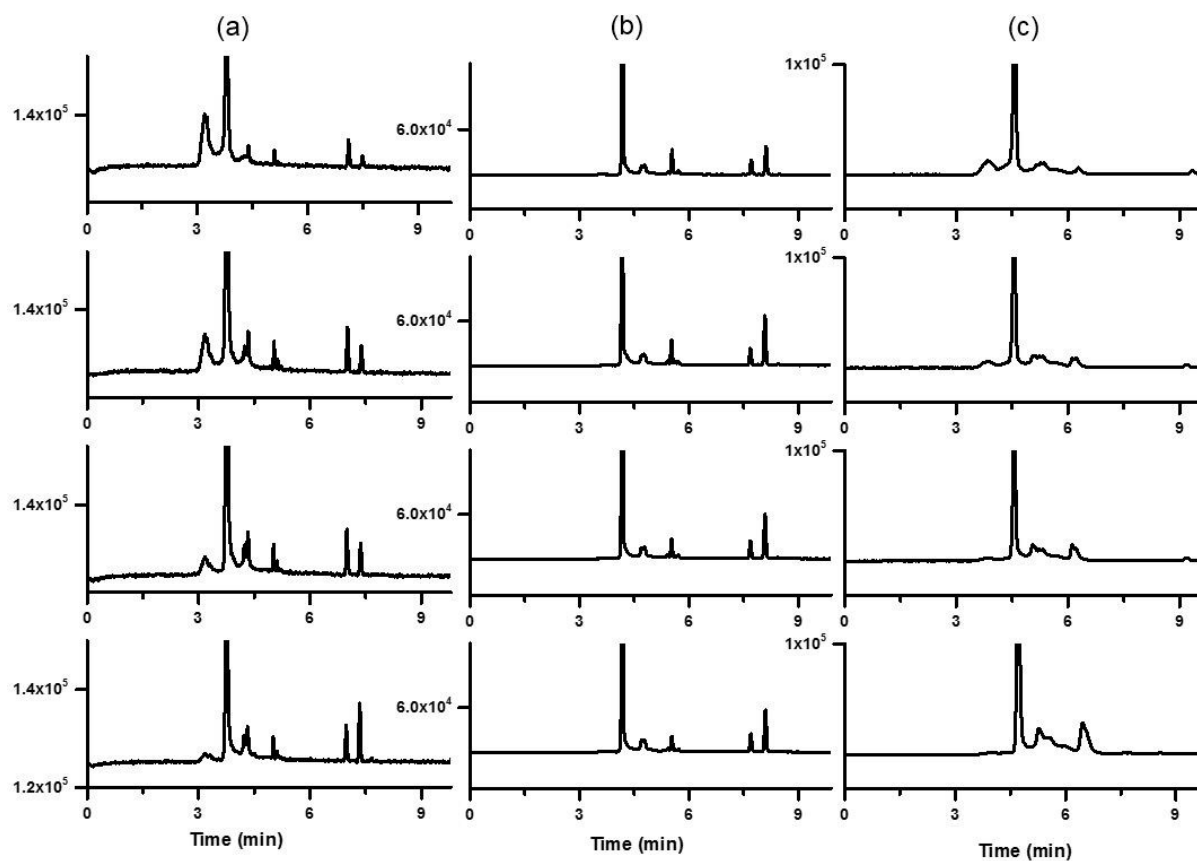
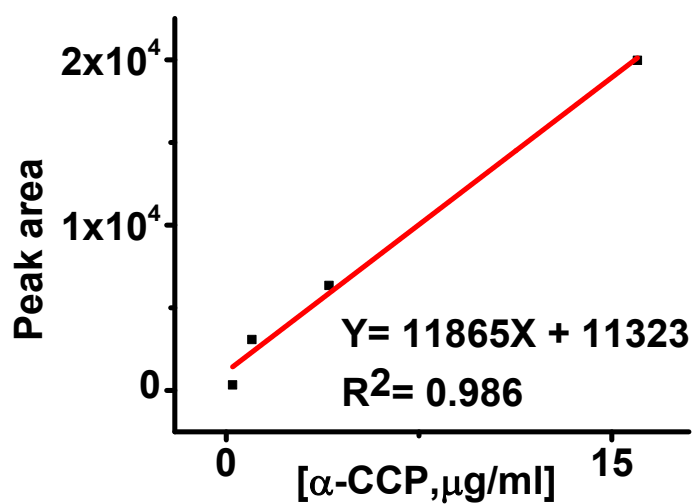


Fig S6. Standard curves of peak area vs. concentration for affinity binding between (a) F-CCP11A and anti-CCP and (b) F-CCP and anti-CCP immunocomplex.

(a)



(b)

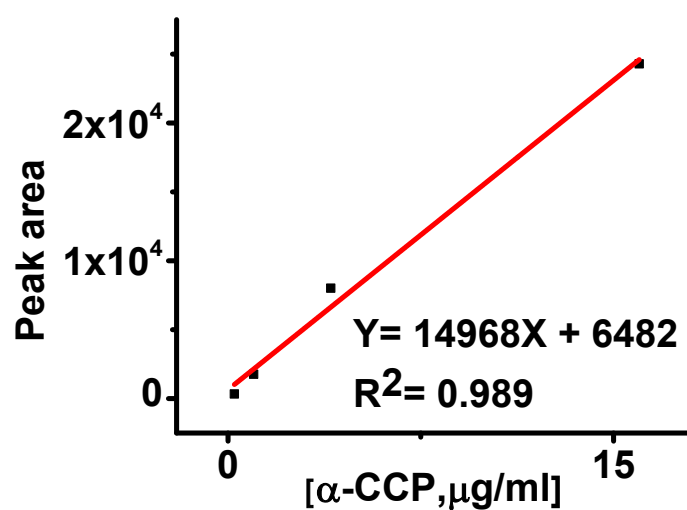


Table 1S

Interday precision	QC sample (µg/mL)	Reproducibility (RSD, %)	Accuracy (%)
F-CCP11A	0.5	2.8 ± 1.2	95 ± 1.7
	0.1	2.5 ± 1.0	104 ± 2.0
F-CCP	0.5	4.6 ± 1.5	104 ± 1.7
	0.1	3.1 ± 0.5	107 ± 1.5

Intraday precision	QC sample (µg/mL)	Reproducibility (RSD, %)	Accuracy (%)
F-CCP11A	0.5	4.0 ± 1.7	105 ± 3.5
	0.1	3.4 ± 1.8	88 ± 2.1
F-CCP	0.5	4.9 ± 1.1	99 ± 5.6
	0.1	3.8 ± 2.2	101 ± 4.6