Supporting Information

An integrated rotational paper-based ELISA platform for multiplex inflammatory biomarker analysis in whole blood with a DIY centrifuge

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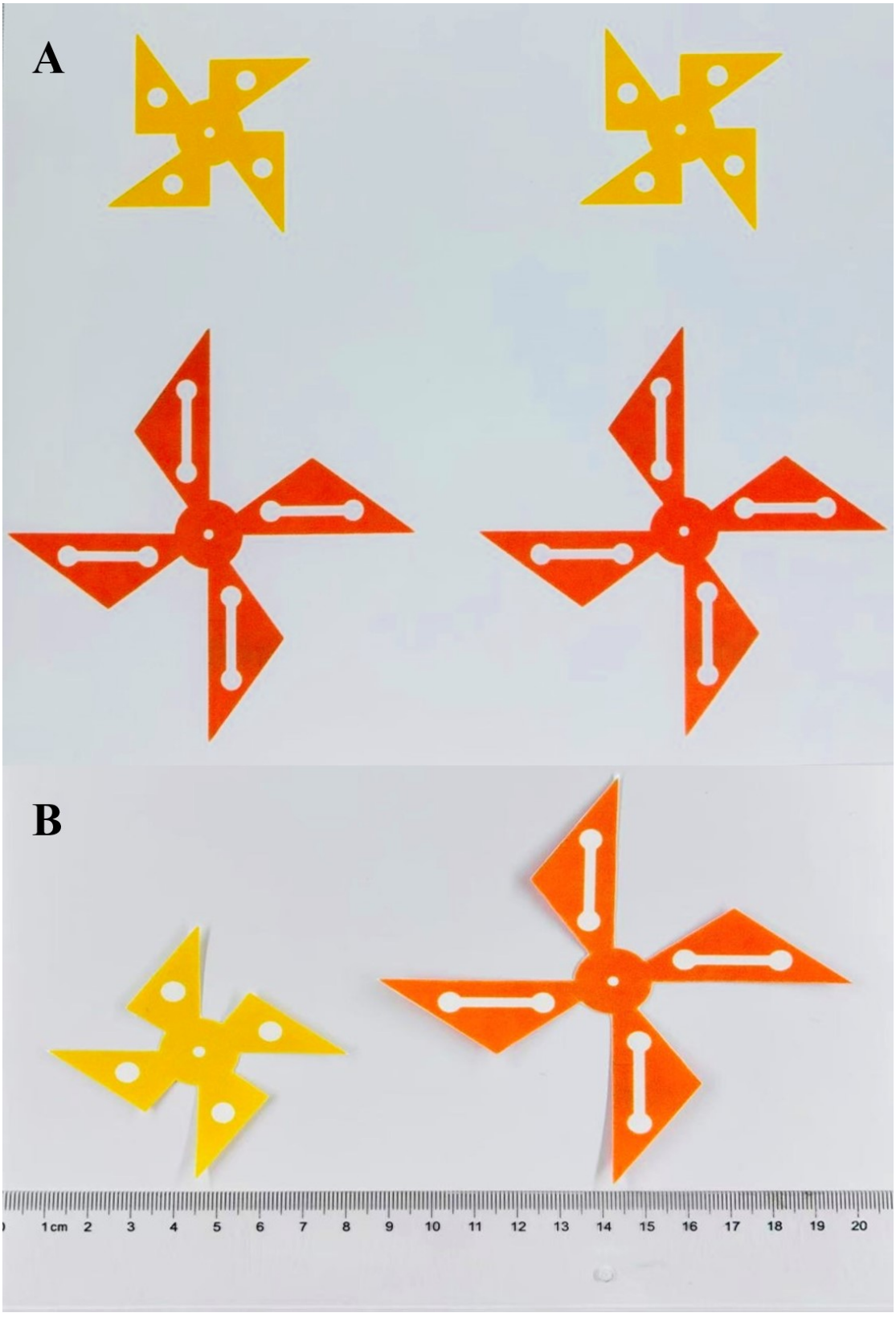

Figure S1. Photograph of paper-based components of the μPADs. (A) Photograph of paper-based microfluidic device prepared using one sheet of chromatography paper (A4, 210 mm × 297 mm). (B) The two component layers are defined as the reaction disc (light yellow color,

left, 55 mm in diameter) and the washing disc (light orange color, right, 110 mm in diameter).

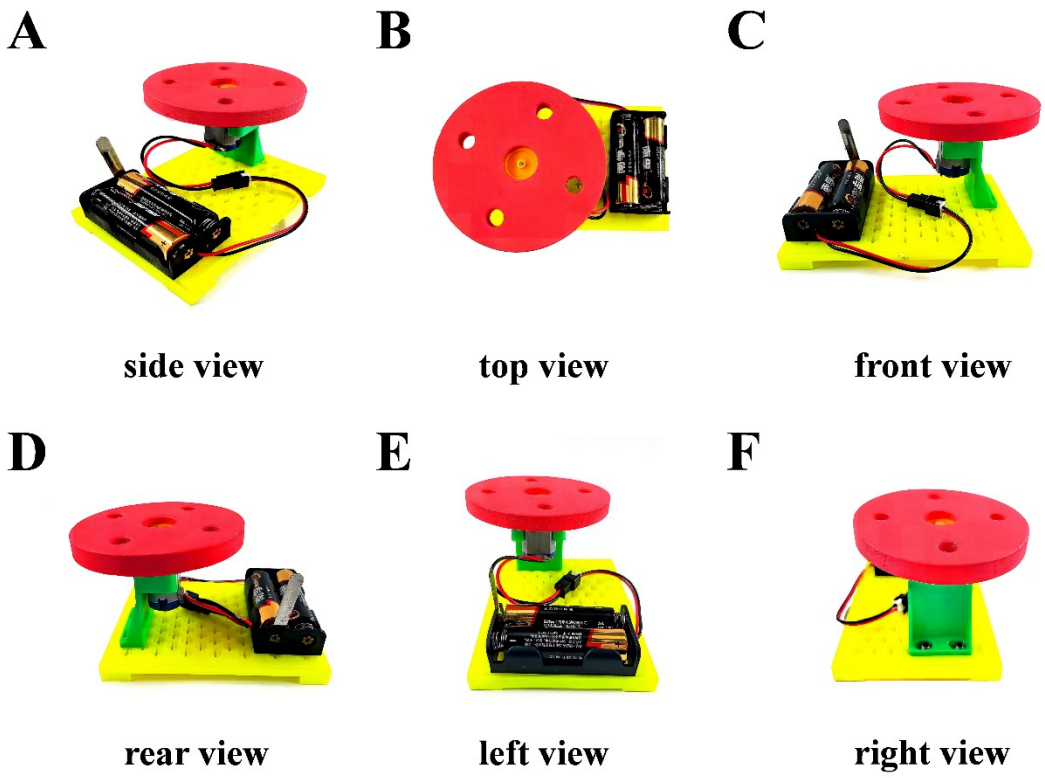

Figure S2. Schematic diagram of a DIY centrifuge assembly. (A) Side view of DIY centrifuge. (B) Top view of DIY centrifuge. (C) Front view of DIY centrifuge. (D) Rear view of DIY centrifuge. (E) Left View of DIY centrifuge. (F) Right view of DIY centrifuge.

Figure S3. Photograph of centrifuge assembly components. (A) Photographs of all centrifugation device components: DIY centrifuge, medical tape, two 1.5V alkaline batteries in series, double-sided tape, a hot glue gun, two capillary tubes, and a circular sheet of plain paper. (B) A top view of the constructed centrifugal device.

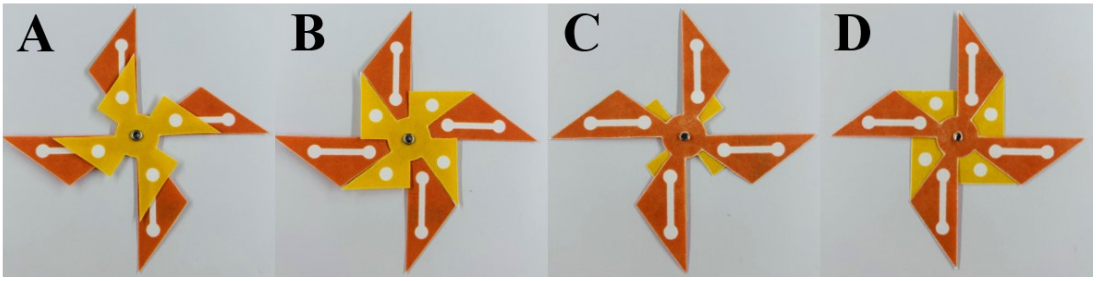

Figure S4. Photograph of the paper-based chip rotational device in different processes. (A) Front when washing. (B) Front side during incubation. (C) Back when washing. (D) Back side during incubation.

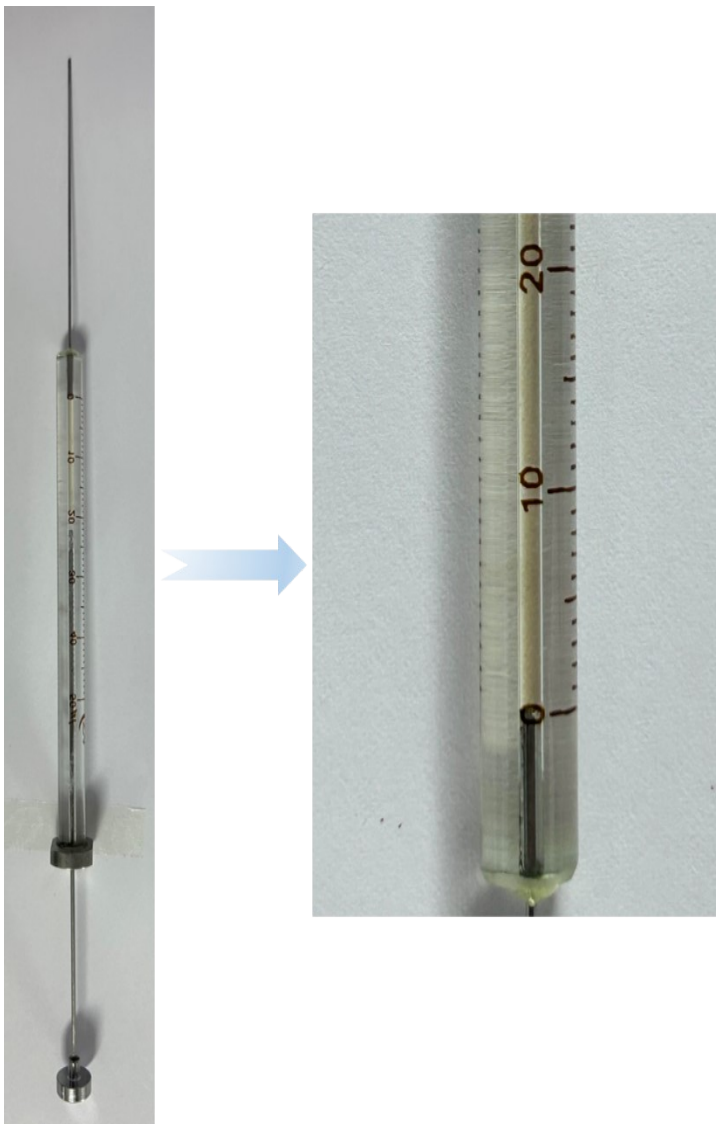

Figure S5. The picture of the serum sample drawn from one capillary tube by a 50 μL syringe after DIY centrifugation.

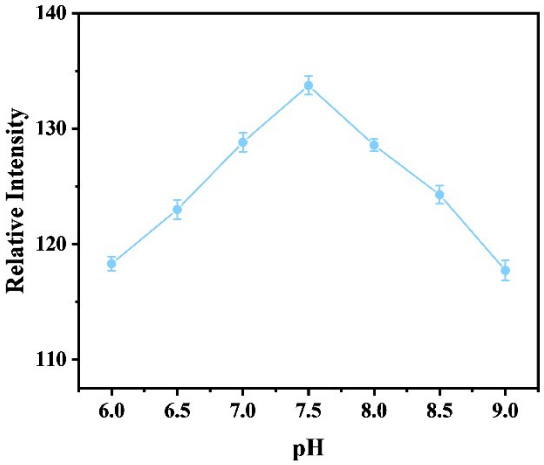

Figure S6. Variation of signal with different pH value from 6.0, 6.5, 7.0, 7.5, 8.0, 8.5, to 9.0, respectively and with CRP as 10.0 ng/mL.

Table S1 Analytical results and spiked CRP with 500-fold dilution with PBS buffer (pH=7.4) and its recovery of in actual serum samples (n=3)

Sample	Spiked concentration (μg/mL)	Diluted concentration (500-fold, ng/mL)	Measured concentration (ng/mL)	Recovery (%)
CRP	1.5	3.0	3.11±0.15	103.55
	3.0	6.0	6.25±0.30	104.12
	5.0	10.0	9.92±0.35	99.22
	7.5	15.0	14.63±1.09	97.56
	10.0	20.0	20.46±1.21	102.32
	12.5	25.0	24.45±0.58	97.78

Table S2 Comparison of this method with reported methods (CRP)

Detection method	Detection limit (ng/mL)	Linear response (ng/mL)	Reference
Fluorescence immunoassay	0.76	1-1000	[1]
A nanobody-based double antibody sandwich ELISA kit	1	6-200	[2]
Electrochemical immunoassay	1.5	10-150000	[3]
Electrochemiluminescence Immunoassay	130	300-160000	[4]
Surface plasmon resonance	9	6-7000	[5]
Surface Enhanced Raman Scattering	1.09	1.56-25	[6]
This work	0.659	3-25	

Table S3 Comparison of this method with reported methods (PCT)

Detection method	Detection limit (ng/mL)	Linear response (ng/mL)	Reference
Fluorescence immunoassay	0.032	0.05-50	[7]
Surface Enhanced Raman Scattering	0.042	0-20	[8]
Electrochemical immunoassay	0.7	1-10	[9]
Lateral Flow Immunoassay	0.1	0.2-300	[10]
Chemiluminescence immunoassay	0.037	0-100	[11]
This work	0.015	0.01-5	

Notes and references

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