Electronic Supplementary Material (ESI) for Lab on a Chip

3D Origami-based Multifunction-integrated Immunodevice: Low-cost and Multiplexed Sandwich Chemiluminescence Immunoassay on Microfluidic Paper-based Analytical Device

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Supplemental Information

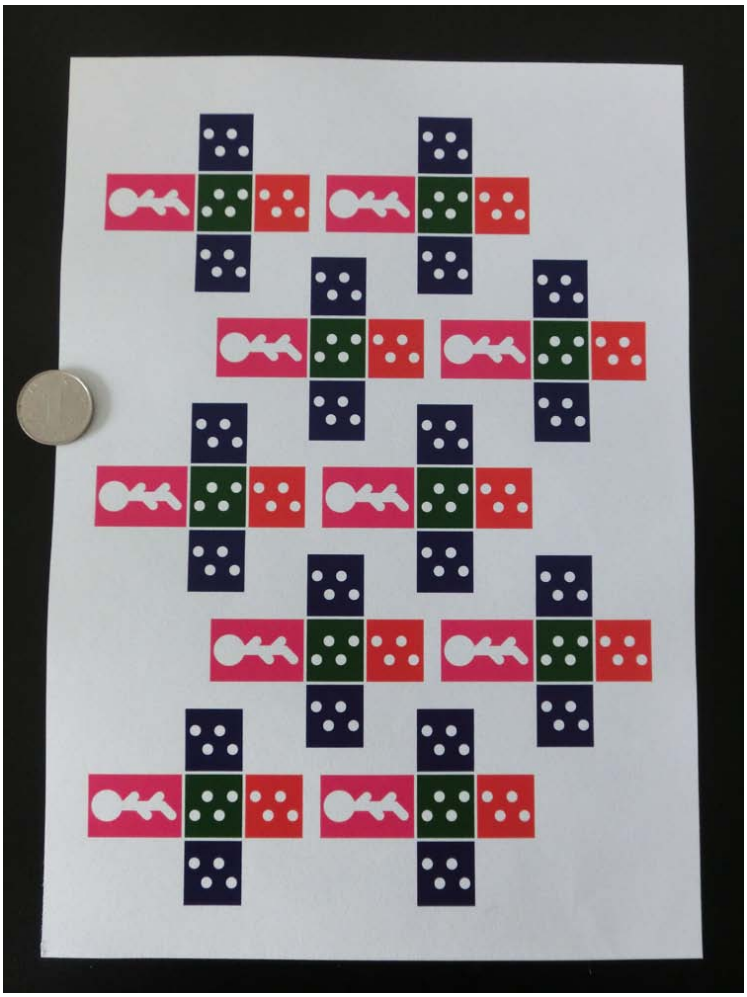

Figure S1. Wax-printed 3D origami-based devices on a paper sheet (A4) before baking.

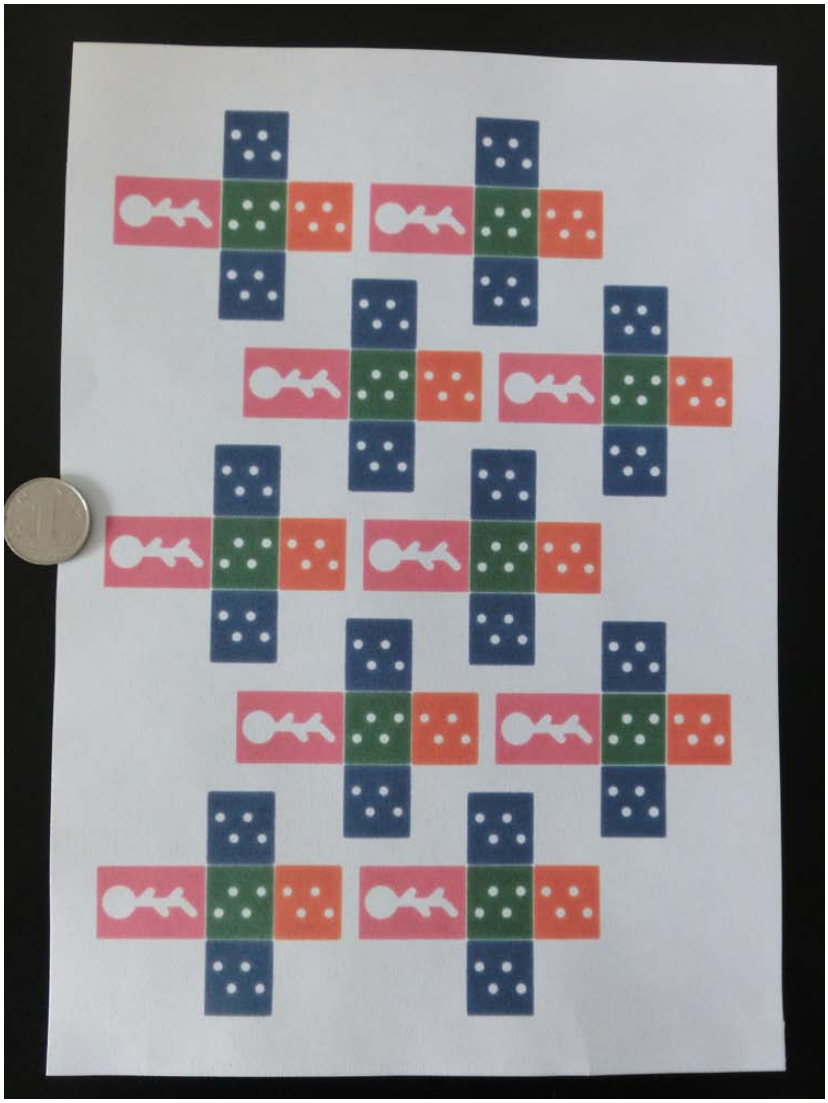

Figure S2. Wax-printed 3D origami-based devices on a paper sheet (A4) after baking.

Synthesis of AgNPs-luminol

The AgNPs-luminol were prepared by the luminol reduction method reported by He [1] with slight modification. Briefly, 10 mL of 5 mM $AgNO_3$ aqueous solution were added to the mixture solution containing 50 mL absolute ethanol and 25 mL ultrapure water with vigorous stirring at room temperature, followed by quick addition of 2.5 mL 0.01 luminol (0.1 M NaOH) to the mixture solution. And the solution was continuously stirred for 2 h, during which time a color change from colorless to primrose yellow to deep yellow, indicating the formation of silver nanoparticles. The size of the as-prepared AgNPs was 20 ± 1.2 nm in diameter.

Preparation of AgNPs-luminol-antibody conjugate

The AgNPs-luminol labeled Ab_2 was prepared according to the following procedure, as reported previously with some modifications [2]. Typically, 23 μg Ab_2 dissolved in 0.1 M PBS was added to 1 mL AgNPs-luminol (pH was adjusted to 6.5 using 0.1 M NaOH), followed by incubation at room temperature for 1 h. After that, 5% BSA solution was added to a final concentration of 1% with stirring for 5 min. The conjugate was centrifuged at 12000 rpm for 45 min (CR22GIII, HITACHI), and the soft sediment was resuspended in 1.0 mL PBS containing 1% BSA (PBS-BSA, pH 7.4). Then the centrifugation was carried out again, and the precipitates were resuspended in 1.0 mL PBS-BSA (pH 7.4). The obtained AgNPs-luminol labeled Ab_2 can be used directly or stored at 4 ℃ for months.

Reference

1 Y. He, D. Liu, X. He and H. Cui, *Chem. Commun.*, 2011, **47**, 10692.

2 J. F. Bao and J. G. Shen, *Experimental technology of immunology*, Zhejiang University Press, 2006, p. 38.