# **Electronic Supplementary Information (ESI)**

### Simultaneous electrokinetic stacking and separation of anionic

## and cationic species on paper fluidic channel

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### **Effect of EOF**



**Figure S1** The influence of the direction of EOF on the enrichment effect. (A) It shows the enrichment process diagram of PAD that treated with PEI. (B) It shows enrichment process diagram in which the PAD has not been subjected to any treatment. The voltage is 300 V, the background electrolyte is 100 mM  $NH_4Ac + 0.4\%$  HEC, and the arrow indicates the direction of the EOF.

#### **Paper Material**

We explored the effects of different paper materials on enrichment. Fig. S2 shows the enrichment process of quantitative filter paper and glass fiber paper. The

quantitative filter paper and the glass fiber paper were treated in the same process, and the direction of EOF was reversed by PEI. Both paper substrate gave positive result in term of the simultaneous stacking as shown in Fig. S2(A). However, the stacking bands of Brilliant blue and Rhodamine 6G on the filter paper channel are less integrate than that on the glass fiber paper. We also explored the cellulose acetate membrane, which has a small sample load and poor water absorption, and the Joule heating effect is obvious when 300 V is applied, causing the channel evaporate quickly, resulting in an open circuit (not shown). Therefore, glass fiber paper was preferred as the material for demonstration of this work in the main manuscript.



Figure S2 Different paper materials vs enrichment effects. (A) It is the enrichment process diagram of the quantitative filter paper at different times (B) It is the enrichment effect diagram of the glass fiber paper at different times. The voltage is 300 V, BGE is 100 mM NH<sub>4</sub>Ac + 0.4% HEC, and the analytes in the anode and cathode reservoirs are mixed solutions of Rhodamine 6G and Brilliant blue of  $5 \times 10^{-6}$  mg/mL.

#### Stacking of small and macro charged species

The 200  $\mu$ L mixed solution of 2 mM Cu<sup>2+</sup> and 5×10<sup>-7</sup> g/mL KNO<sub>2</sub> was added to the anode and cathode reservoirs, and the voltage of 300 V was applied. After enrichment for 100 s, disconnect the power supply and add Griess reagent at the cathode end of the paper-based fluidic channel.

The 200  $\mu$ L mixed solution of cytochrome C and phycocyanin at a concentration of 0.1 mg/mL was added to the anode and cathode reservoirs.

**Formula Derivation** 

$$J = \frac{I}{S} \tag{1}$$

$$R = \rho \frac{L}{S} \tag{2}$$

$$U = IR \tag{3}$$

$$U = I\rho \frac{L}{S} \tag{4}$$

$$E = \frac{U}{L} \tag{5}$$

$$E = \rho \frac{I}{S} = \rho J \tag{6}$$

$$\sigma = \frac{1}{\rho} \tag{7}$$

$$E = \frac{J}{\sigma} \tag{8}$$

Eqn (1) is the equation of definition of current density, J is current density, I is current, and S is cross section area. Eqn (2) is the law of resistance, R is the resistance,  $\rho$  is the resistivity, L is the length. Eqn (3) is obtained by the Ohm's law, U is the voltage. Substituting eqn (2) into eqn (3) gets eqn (4). Eqn (5) is a formula for calculating the electric field strength, E is the electric field strength. Substituting eqn (4) into eqn (5) gets eqn (6). Eqn (7) is the relational expression between resistivity and conductivity, and they are reciprocal to each other,  $\sigma$  is the conductivity. Substituting eqn (7) into eqn (6) gets eqn (8). Eqn (8) is the relation expression between the local conductivity and local electric field strength.