Supporting Information

Phenylboronic acid-modified polyethyleneimine assisted neutral polysaccharide of weight-resolution analysis with a nanopipette

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Content

1. Synthesis of PEI_{1800}-oBA

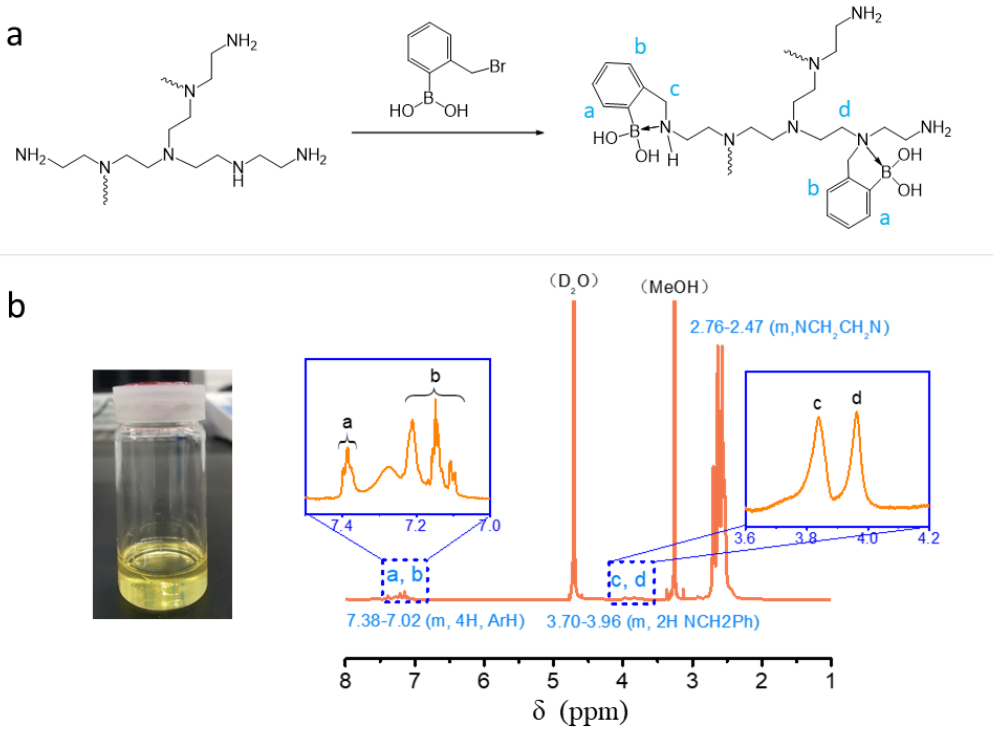


Figure S1. (a) Synthesis of PEI_{1800}-oBA, (b) the photo of the synthesis compound (left) and the 1H NMR spectra at 300 MHz in D_2O (right): δ 7.39 (br s, 2H), 7.11 (br s, 2H), 3.65 and 3.45 (br s, 2H), and 2.66-2.47 (m, 88H).

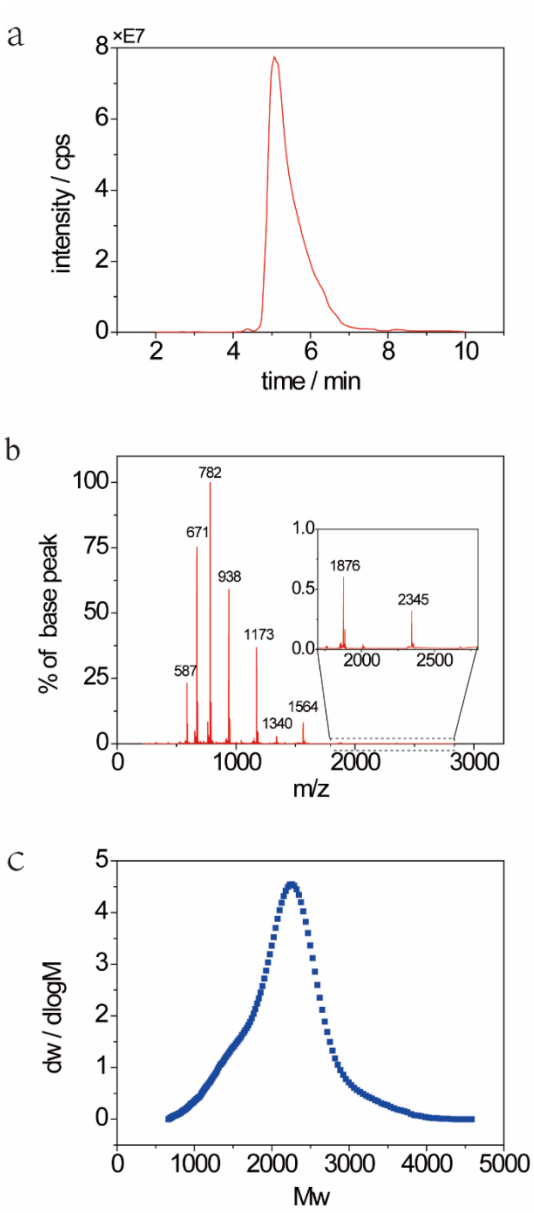


Figure S2. Liquid chromatography tandem mass spectrometry LC-MS and Gel Permeation Chromatography (GPC) analysis of the prepared PEI_{1800}-oBA molecular. Chromatograms (a), mass spectra (b), GPC spectral (c) of PEI_{1800}-oBA.

Table S1. The Mw averages for the prepared PEI_{1800}-oBA

Mp	Mn	Mw	Mz	Mz+1	Mv	PD
2265	1910	2067	2205	2328	2045	1.0822

2. Nanopore fabrication and the diameter estimate

Nanopipette fabrication: Quartz capillaries (QF100-50-10), with an outer diameter of 1.0 mm and inner diameter of 0.5 mm were obtained from Sutter Instrument. All glass capillaries were thoroughly treated by immersion in freshly prepared piranha solution (98% H_2SO_4 : 30% H_2O_2 = 3:1 v/v) for approximately 2 h. The capillaries were then thoroughly rinsed with deionized water and ethanol several times and dried under N_2 gas. Before use, the cleared capillaries were dried at 80 °C for 1 h. A CO_2-laser P-2000 puller system (Sutter Instruments Co. Ltd) was used to fabricate the nanopipette with the following settings: Heat = 760, Fil = 4, Vel = 29, Del = 140, and Pull = 168. The tip diameters of the nanopipettes were approximately 20 nm and were characterized by SEM and ionic conductance.

The diameter estimate: The electrochemical measurement to estimate the nanopore diameter according to the classical equation (S1). [S1]

$$a = \frac{1}{\pi k R tan\theta/2} \qquad \text{(Equation S1)}$$

R is the measured nanopipette resistance, k is the specific resistance of the electrolyte used (k= 7.6 S/m in 1M LiCl), θ is the cone angle (θ=18° in Figure S1b), a is the diameter of the nanopore at the tip of the nanopipette.

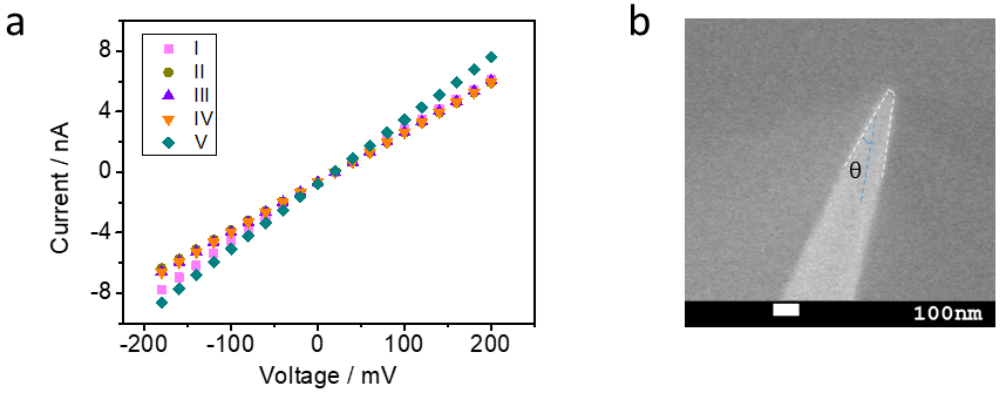


Figure S3. (a) The I-V response of the 5 different nanopipettes (1 M LiCl, 10 mM Tirs-HCl, pH 7.4); (b) Scanning electron microscope image of the presented nanopipette.

3. Calculation of the baseline fluctuation

The baseline fluctuation is quantitatively expressed as I_{RMS}, which is directly calculated as the following:

$$I_{RMS} = \sqrt{\Delta I^2(t)} \qquad \text{(Equation S2)}$$

The root-mean-square (RMS) noise I_{RMS} is the electrical current through the pore, and $\Delta I(t)$ represents the fluctuation of the current I(t) deviating from its mean value. [S2]

4. High PEI_{1800}-oBA concentration

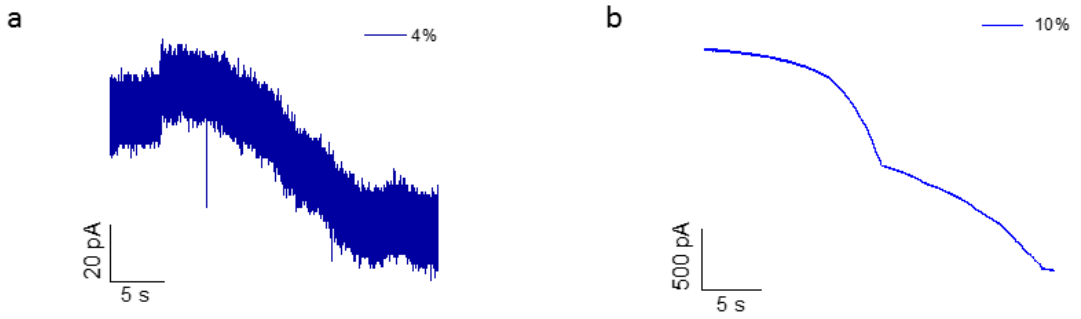


Figure S4. The current trace for 7 µg/ml dextran 70 mixed with (a) 4% and (b) 10% PEI_{1800}-oBA in 1M LiCl (10 mM Tris-HCl, pH 7.4) at 300 mV, respectively.

5. Scatter plot of ΔI *vs* Δt for dextran 70 with different concentration of PEI_{1800}-oBA

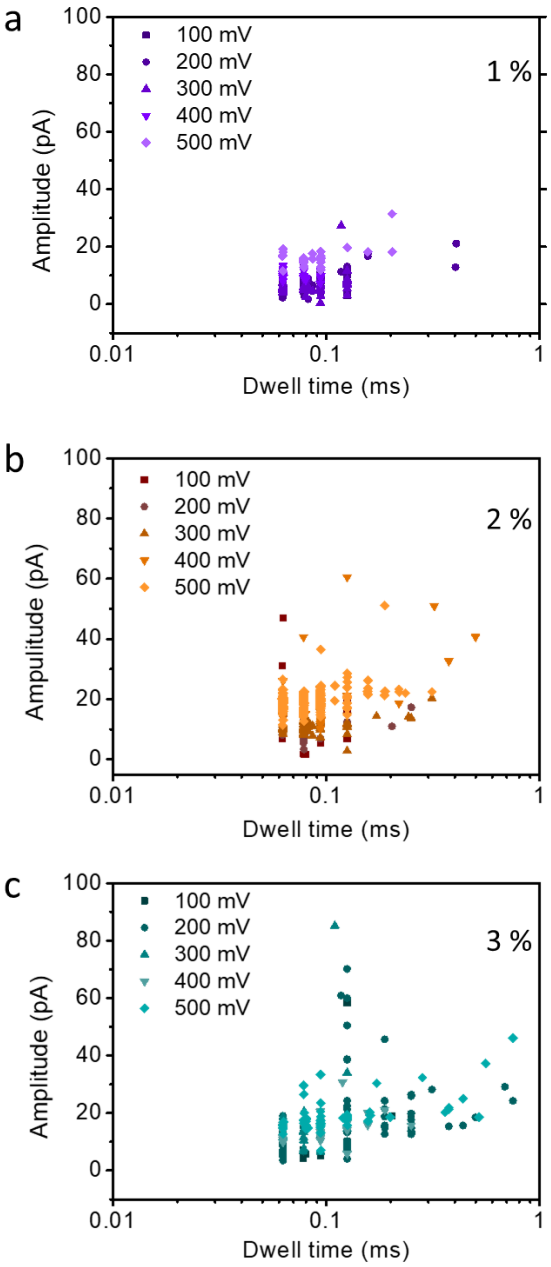


Figure S5. Scatter plot of current blockades *vs.* dwell times of 7 μg/ml dextran 70 (1M LiCl, 10 mM Tris-HCl, pH 7.4) mixed with different concentration of PEI_{1800}-oBA (a. 1%, b. 2%, c. 3%) at different applied voltage (100-500mV).

6. The TEM image and Zeta potential of PEI_{1800}-oBA mixing with dextran 70

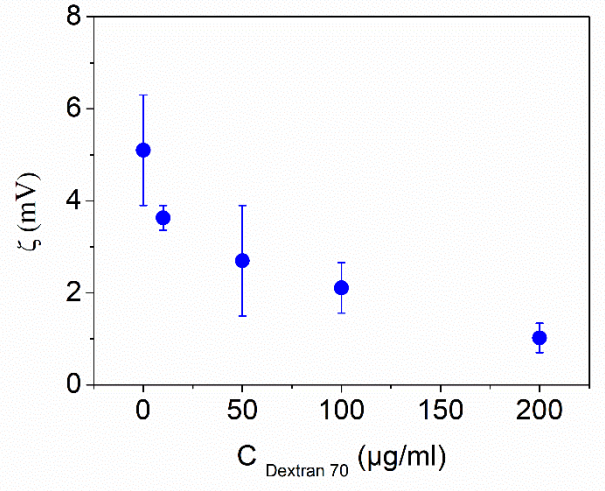


Figure S6. Zeta potential for PEI_{1800}-oBA mixing with different concentration of dextran 70 (0, 10, 50, 100, 200 μg/ml).

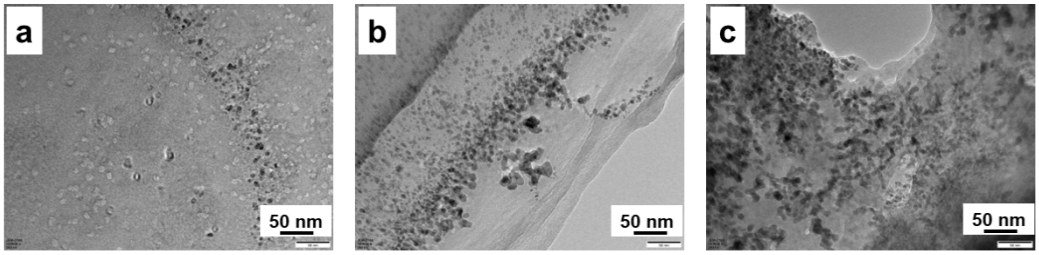


Figure S7. TEM image of PEI_{1800}-oBA mixed with different concentration of dextran 70. (a) is without dextran 70; (b) is mixed with 100 μg/ml; (b) is mixed with 200 μg/ml.

7. The signal-to-noise ratio with PEI_{1800}-oBA concentration

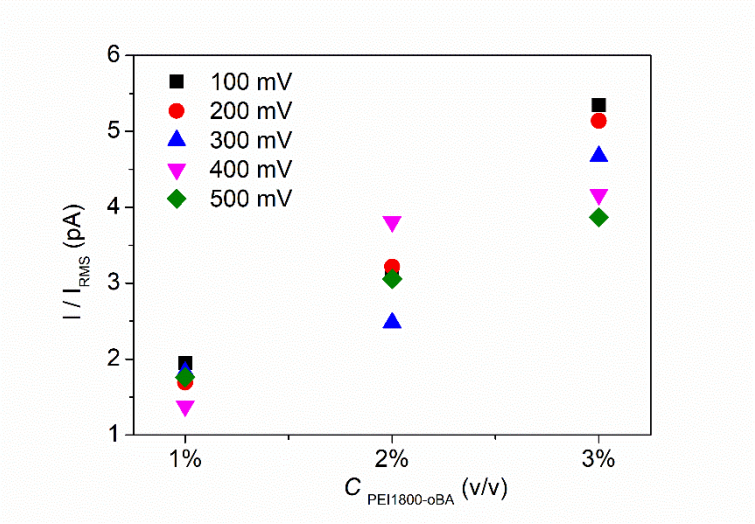


Figure S8. Signal-to-noise ratio of 7 μg/ml dextran 70 mixed with different concentrations (1%, 2%, and 3%) of PEI_{1800}-oBA at different voltages (100, 200, 300, 400, and 500 mV) in a 1 M LiCl (10 mM Tris-HCl, pH 7.4) buffer solution.

8. Current trace for different concentration of dextran 70

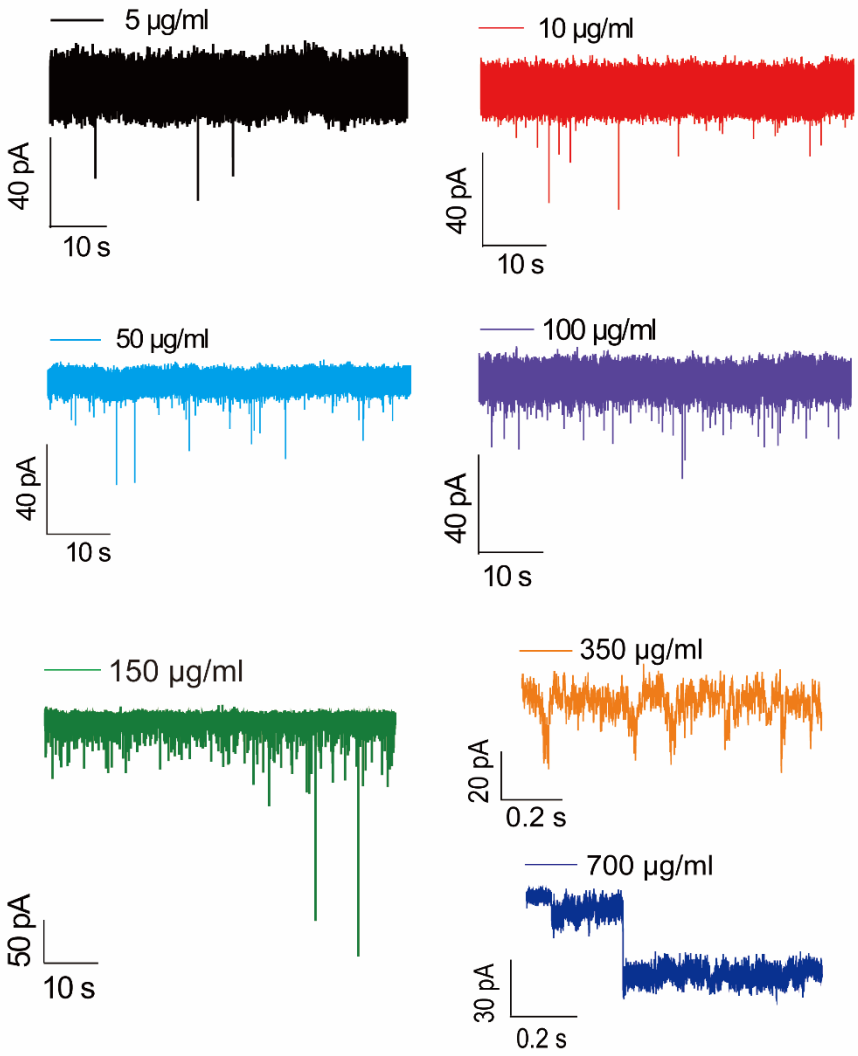


Figure S9. The current trace for different concentration of dextran 70 (5, 10, 50, 100, 150, 350 and 700 μg/ml) mixed with 3% PEI_{1800}-oBA at 300 mV voltage (1 M LiCl, 10 mM Tris-HCl, pH 7.4), respectively.

9. Compared with other dextran detection methods

Table S2. The proposed method compared with other dextran detection methods.

Detection method	Molecular weight	Detection range	Detection time	Ref.
GPC[a]	70 000	0.025-1 mg/ml	20 min	S3
HPLC[b]	70 000	0.1-5.0 mg/ml	30 min	S4
ELISA[c]	4 000	26.3-1174.9 ng/ml	60 min	S5
Ic-ELSA[d]	2 000	4.39-544.43 ng/ml	60 min	S6
Nanopipette	70 000	1-100 μg/mL	10 min	This method

[a] Gel Permeation Chromatography

[b] High Performance Liquid Chromatography

[c] Enzyme Linked Immunosorbent Assay

[d] Indirect Competitive Enzyme-Linked Immunosorbent Assay

10. Scatter plot of scatterplots of ΔI vs Δt for dextrans (20, 40, 70)

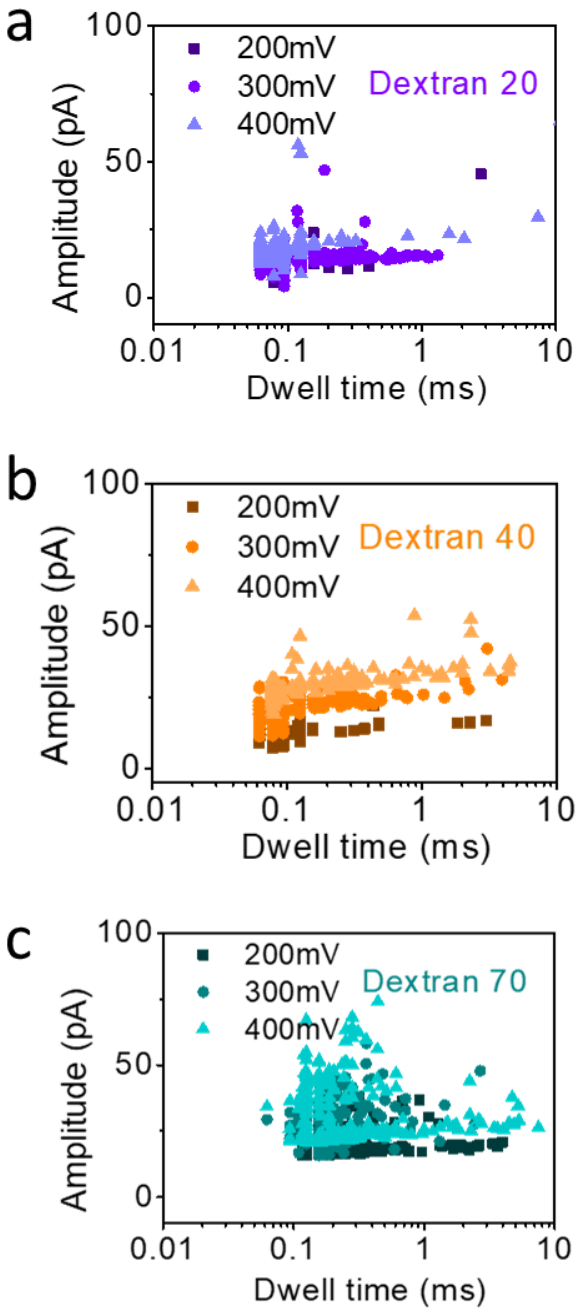


Figure S10. Scatter plot of scatterplots of current blockades *vs.* dwell times for three different molecular weights dextran (1 μM; dextran 20, dextran 40, and dextran 70) mixed with 3% PEI1800-oBA at 300 mV voltage (1 M LiCl, 10 mM Tris-HCl, pH 7.4).

11. EDC analysis for dextran 20 and dextran 40 detection at 200 mV

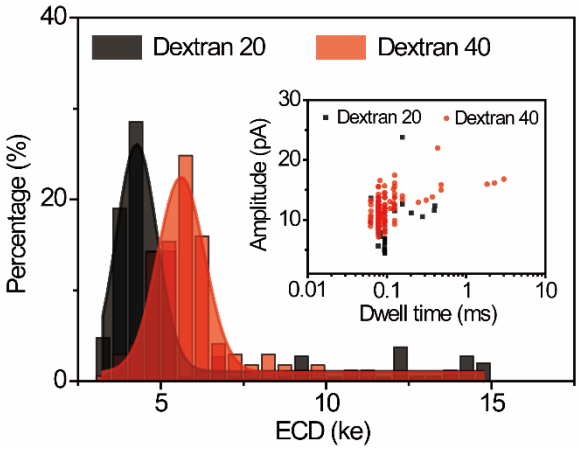
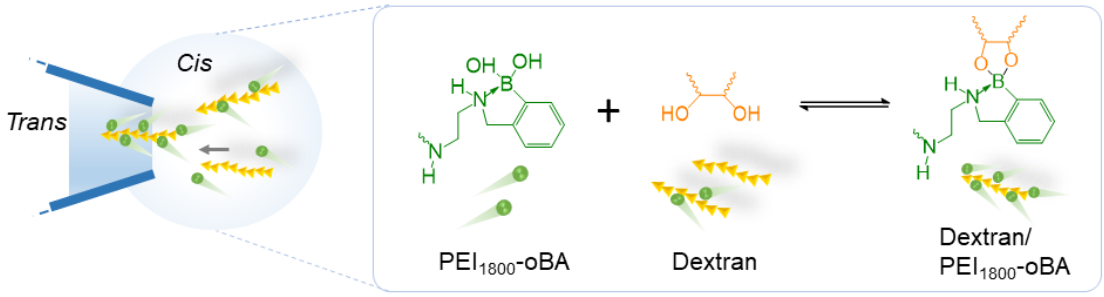


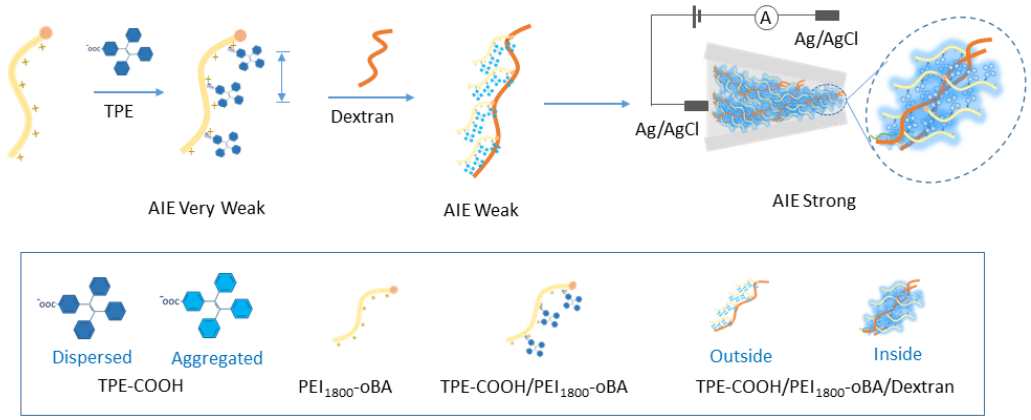
Figure S11. ECD histogram for dextran 20 and dextran 40 detection at 200 mV (1 M LiCl, 10 mM Tris-HCl, pH 7.4).

12. Schematic of the reversible reaction process of PEI_{1800}-oBA with dextran



Scheme S1. Schematic of PEI_{1800}-oBA and dextran mixture based on nanopipette and the reversible reaction process of PEI_{1800}-oBA with dextran.

13. Schematic of the AIE fluorescence detection



Scheme S2. Schematic of the AIE fluorescence detection for PEI_{1800}-oBA/Dextran complex translocation based on nanopipette detection under the negative voltage applied.

14. TPE-COOH concentration optimization

Stock solutions of dextran 20 (1 μM), dextran 40 (1 μM), dextran 70 (1 μM), and PEI_{1800}-oBA (10 mg/mL) were prepared using 1 M LiCl (10 mM Tris-HCl, pH 7.4). A stock solution of TPE-COOH (1 mM) was prepared with DMF. All of the solutions were diluted with 1M LiCl (10 mM Tris-HCl, pH 7.4) before use. For TPE-COOH/PEI_{1800}-oBA mixtures fluorescence detection, different concentrations of TPE-COOH were mixed with 3% PEI_{1800}-oBA in 96-well plates.

Figure S8 shows the fluorescence imaging and intensity of different concentrations of TPE (10-50 μM) in 1 M LiCl mixed with PEI_{1800}-oBA (3%). With increasing TPE concentration, the fluorescence intensity increased. Thus, the TPE-COOH /PEI_{1800}-oBA complex formed by TPE-COOH adsorbs on the surface of PEI_{1800}-oBA. The fluorescence intensity curve suggested that 3% PEI_{1800}-oBA needed 40 μM TPE-COOH to reach saturation.

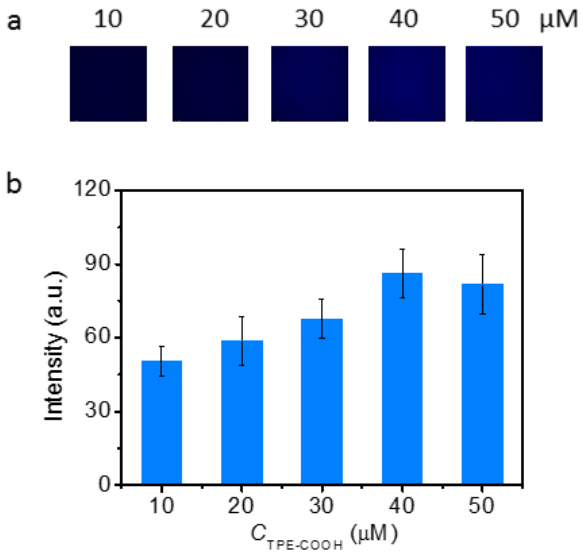


Figure S12. (a) The fluorescence image and (b) the corresponding fluorescence intensity of 1 μM dextran 70 and 3 % PEI_{1800}-oBA complex with different concentration of TPE-COOH (10, 20, 30, 40, and 50 μM).

15. Fluorescence intensity sum in Figure 4b

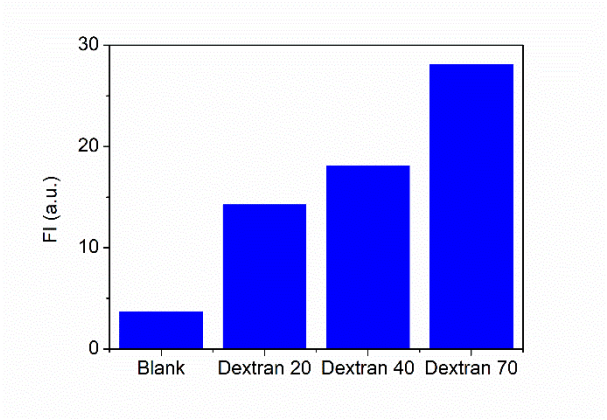


Figure S13. Fluorescence intensity sum in Figure 4b of different molecular weight dextrans (1 μM, without dextran, dextran 20, 40, and 70) mixed with 3% PEI_{1800}-oBA and 30 μM TPE-COOH under 300 mV voltage for 30 min (1 M LiCl, 10 mM Tris-HCl, pH 7.4).

16. ECD histogram for dextran mixtures

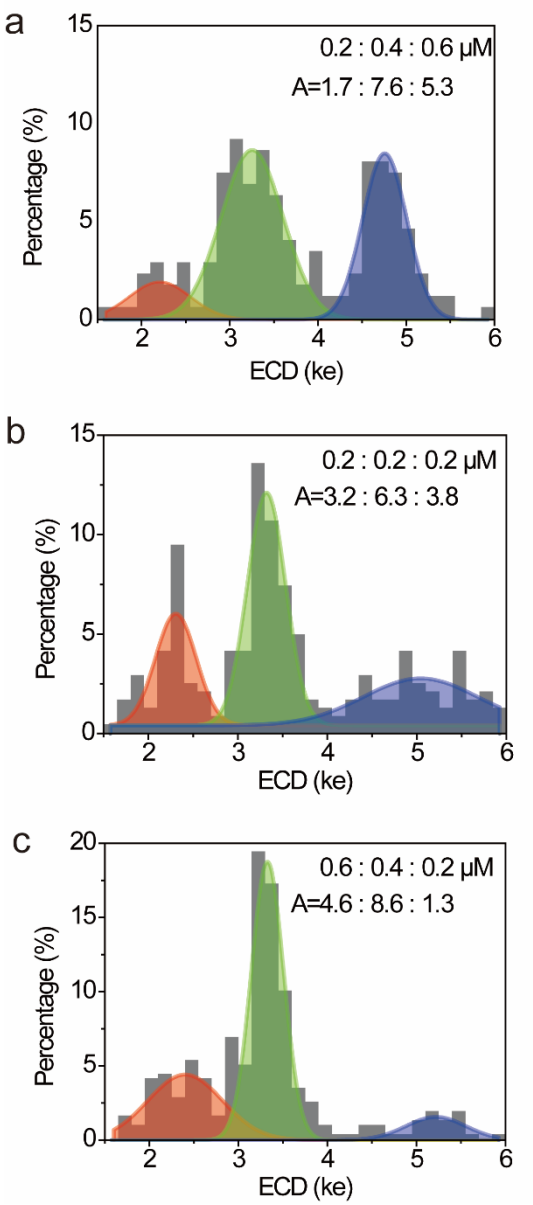


Figure S14. ECD histogram for dextran mixtures (dextran 20: dextran 40: dextran 70) with respective concentration ratios of (a) 0.2: 0.4: 0.6 μM, (b) 0.2: 0.2: 0.2 μM, and (c) 0.6: 0.4: 0.2 μM mixed with 3% PEI_{1800}-oBA at 300 mV voltage (1 M LiCl, 10 mM Tris-HCl, pH 7.4).

17. The recovery for SZK and MK eye drops samples

Table S3. The recovery for SZK and MK eye drops samples.

Sample	Added (μg/ml)	Detected (μg/ml)	Recovery (%)
ZSK	0	48	96.0
ZSK	20	68	97.1
MK	0	55	110
MK	20	72	103

Reference

[S1] Z. Zhu, X. Duan, Q. Li, R. Wu, Y. Wang, B. Li, *J. Am. Chem. Soc.* 2020, **142**, 4481−4492.

[S2] S. Liang, F. Xiang, Z. Tang, R. Nouri, X. He, M. Dong, W. Guan, *Nanotechnology and Precision Engineering*, **2020**, *3*, 9-17.

[S3] R. A. Burke, T. R. Shockley, *Journal of Chromatography B*, **1998**, 718, 115-120.

[S4] L. Wang, R. Gong, Z. Liu, *China Pharmacy*, **2015**, 26, 5126-5128.

[S5] S. Wang, Z. Li, X. Wang, E. Ni, L. Zeng, F. Luo, J. Yan, D. Liang, *Journal of Food Quality*, **2016**, 39, 408-414.

[S6] C. Li, W. Sun, L. Huang, N. Sun, X. Hua, M. Wang, F. Liu, *RSC Advances*, **2021**, 11, 517-524.