Electronic Supplementary Information

Cell-Penetrating Peptide Modified AIE Polymeric Nanoparticles by Miniemulsion Polymerization and Application for Cell Fluorescence Imaging

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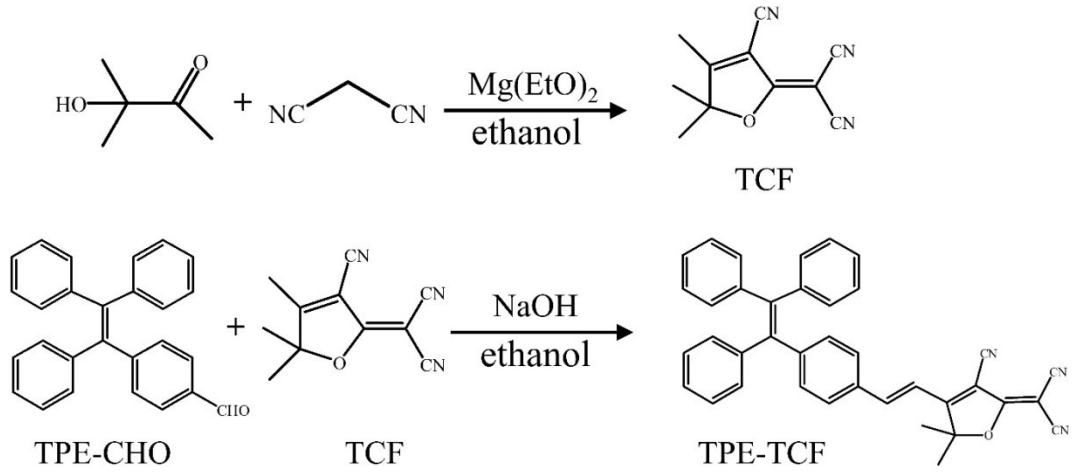
Synthesis of 2-(3-cyano-4,5,5-trimethylfuran-2(5H)ylidene)-malononitrile (TCF)

TCF was synthesized through the reaction of malononitrile with 3-hydroxy-3-methylbutan-2-one according to the method reported in previous publication (Scheme S1).[1] Typically, malononitrile (7.0 g, 106 mmol) and magnesium ethoxide ($Mg(EtO)_2$, 3.9 g, 34 mmol) were added into a 100 mL two-necked round-bottomed flask under a nitrogen atmosphere. 3-Hydroxy-3-methylbutan-2-one (4 mL, 38 mmol) and 50 mL of ethanol were injected into the flask in sequence. The reaction was carried out at 60 °C for 10 h. After cooled down to room temperature, the reaction mixture was dissolved with 100 mL of dichloromethane (DCM, AR, Shanghai Guoyao Chemistry Co., Ltd.) and filtered by sand core funnel. The filtrate was steamed to a solid state at 35 °C by rotary evaporation. The sample was further purified by silica

gel column chromatography by using DCM as the eluent, and was recrystallized from ethanol.

Synthesis of 2-(3-cyano-5,5-dimethyl-4-(4-(1,2,2-triphenylvinyl)styryl)furan -2(5H) ylidene)malononitrile (TPE-TCF)

4-(1,2,2-Triphenylvinyl)benzaldehyde (TPE-CHO, 0.36 g, 1.0 mmol), TCF (0.24 g, 1.2 mmol), and NaOH (2.1 mg) were added to a 100 mL two-necked round-bottomed flask. The flask was evacuated under vacuum and purged with nitrogen for four times. Ethanol (40 mL) was added into the flask, and the reaction was carried out at 90 °C under reflux condition for 10 h. The reaction product was extracted with DCM. The filtrate was dried at 35 °C by rotary evaporation, and the dried sample was purified by silica gel column chromatography by using petroleum ether (AR, Hangzhou Gaojing Chemistry Co., Ltd.)/DCM/ethyl acetate (AR, Wuxi Zhangwang Chemistry Co., Ltd.) (200:100:10 by volume) as the eluent.



Scheme S1. Synthetic route of TCF and TPE-TCF.

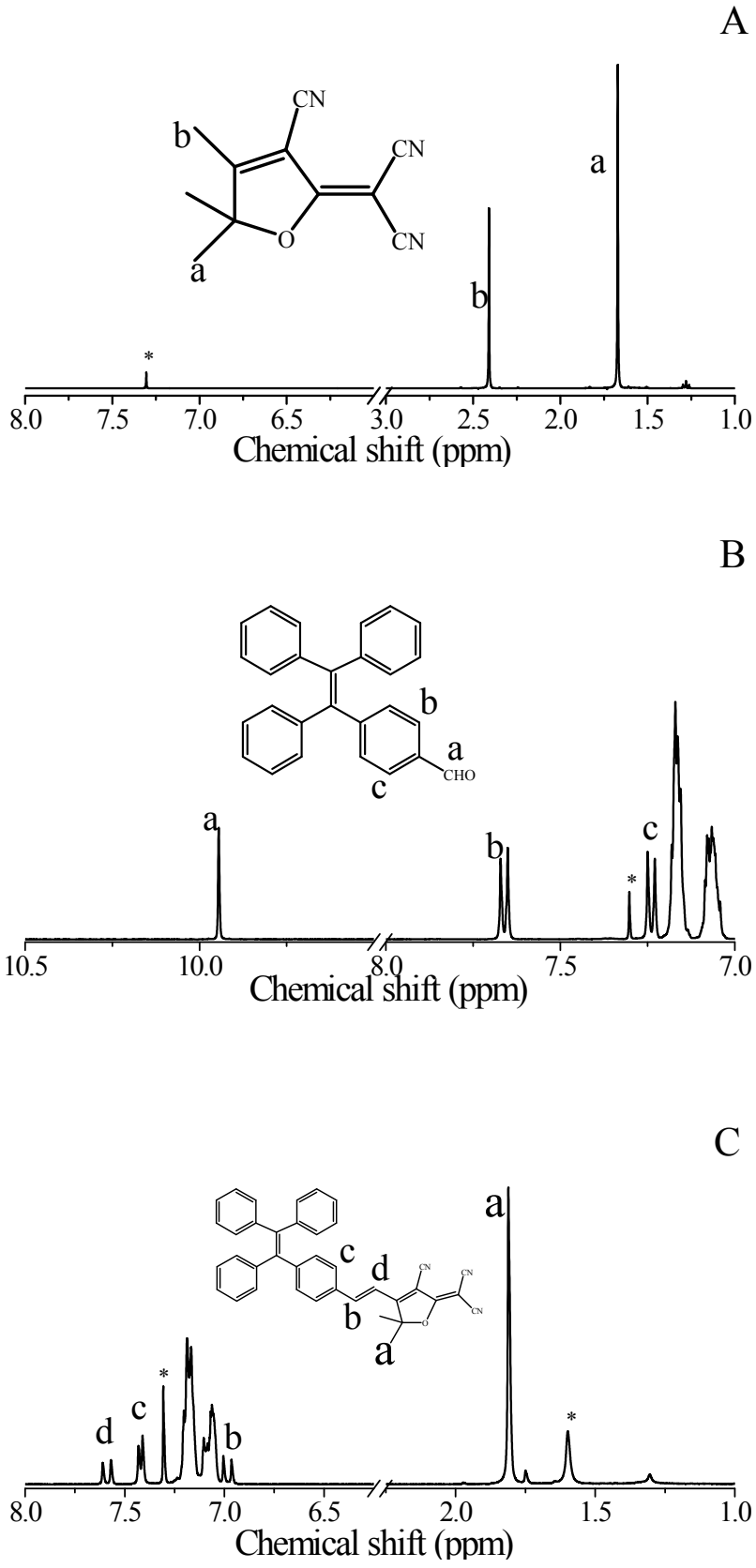


Fig. S1. ^{1}H NMR of TCF (A), TPE-CHO (B), and TPE-TCF (C) in chloroform-d (99.9%, 0.03%V/V tetramethyl silane, Cambridge Isotope Laboratories, Inc.)

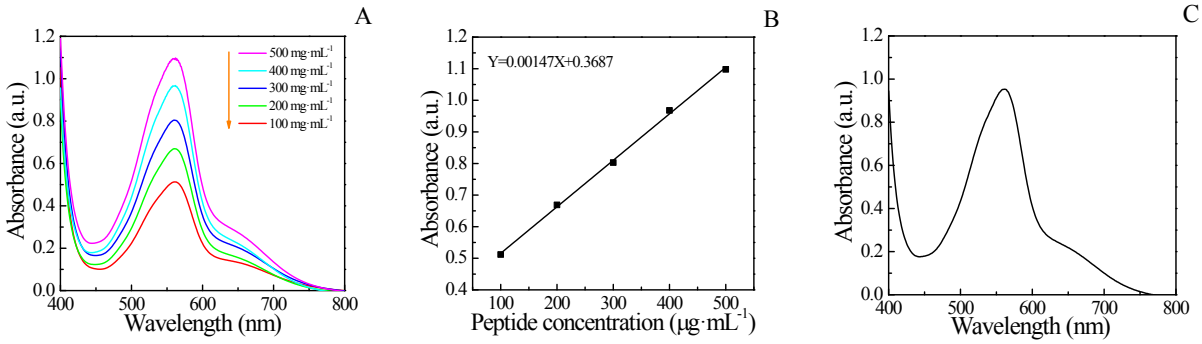


Fig. S2. UV–vis spectra of aqueous solutions with various HIV-1 Tat peptide concentrations (A). Standard curve of the absorbance at 562 nm versus the concentration of HIV-1 Tat peptide (B). UV–vis spectrum of aqueous solution containing unreacted HIV-1 Tat peptides (C).

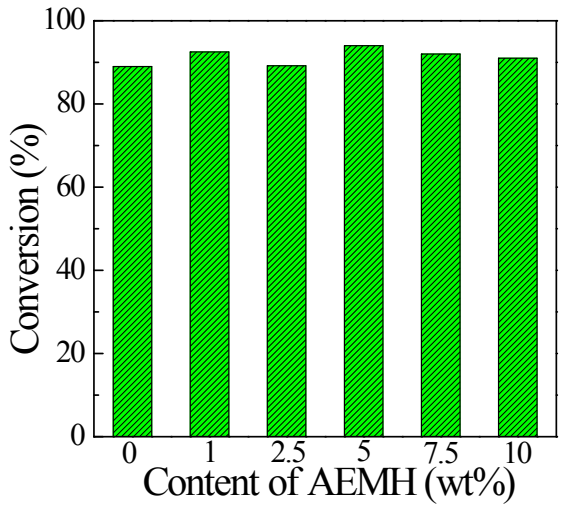


Fig. S3. Final conversions of miniemulsion polymerization systems with various AEMH contents.

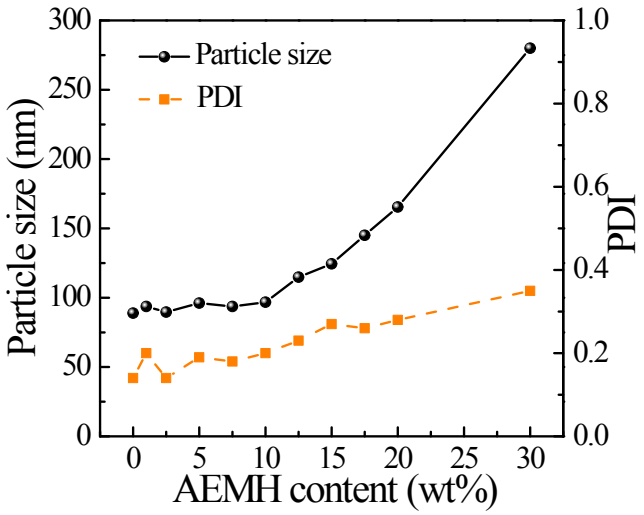


Fig. S4. Particle sizes and PDI values of AIE-NH_2-PMANPs with various AEMH contents.

Table S1. Variables for calculating the surface amino density of NH_2-AIE-PMANPs various AEMH contents and AIE PMMA NPs.

Sample name	D_n[a] (nm)	Solid content[b] (%)	V_b[c] (uL)
AIE PMMA NPs	89	7.08	41
AIE-1.0%NH_2-PMANPs	91	7.80	52
AIE-2.5%NH_2-PMANPs	95	6.40	66
AIE-5.0%NH_2-PMANPs	96	5.39	77
AIE-7.5%NH_2-PMANPs	96	3.95	128
AIE-10.0%NH_2-PMANPs	97	2.66	117

[a] Determined by DLS; [b] the solid content of emulsions after dialysis; [c] calculated through V_2-V_1 in Fig. 2A and S1.

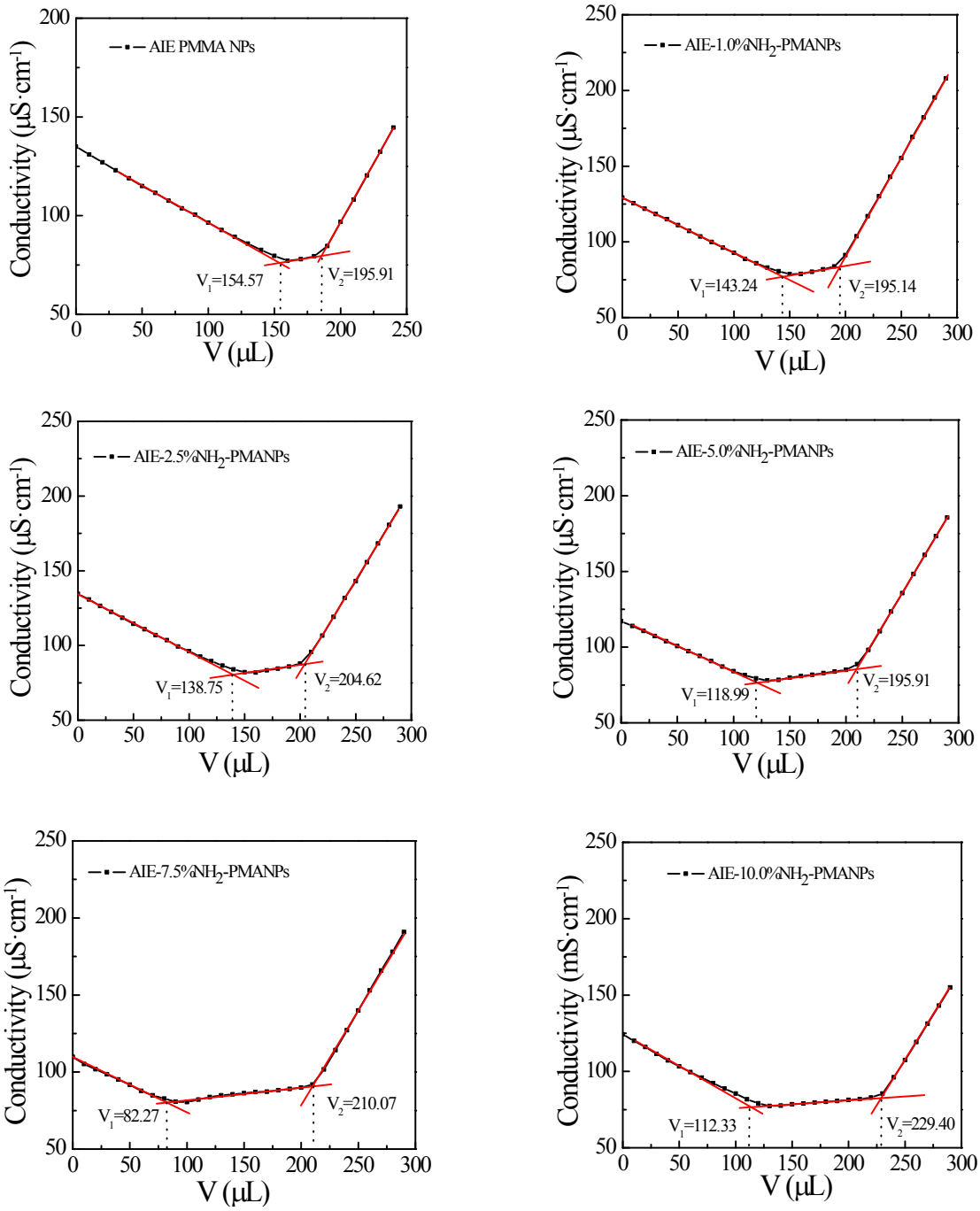


Fig. S5. Conductometric titration curves of the AIE-NH_2-PMANPs with various AEMH contents.

Reference.

1. Y. J. Wang, Y. Shi, Z. Y. Wang, Z. F. Zhu, X. Y. Zhao, H. Nie, J. Qian, A. J. Qin, J. Z. Sun and B. Z. Tang, *Chem. Eur. J.*, 2016, **22**, 9784-9791.