Supplementary Information

pH-responsive Magnetic Metal–Organic Framework Nanocomposites for Selective Capture and Release of Glycoproteins

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1. Materials

Iron(III) chloride hexahydrate ($FeCl_3 \cdot 6H_2O$), Iron(II) chloride tetrahydrate ($FeCl_2 \cdot 4H_2O$), iron nitrate nonahydrate ($Fe(NO_3)_3 \cdot 9H_2O$), bovine serum albumin (BSA), and transferrin (TRF) were purchased from Sigma Aldrich (St. Louis, MO, USA). 1, 4-phenylenebisboronic acid (PBA), terephthalic acid (PTA), polyvinyl pyrrolidone (PVP, Mw 50,000) and polyetherimide (PEI, Mw 25,000) were purchased from Aladdin (Shanghai, China). Trisodium citrate dehydrate (Na_3CT), ethylene glycol (EG), anhydrous ethanol, acetonitrile (ACN) and dimethyl formamide (DMF) were purchased from Forest Science and Technology Development Co. Ltd. (Chengdu, China). The commercial magnetic beads (Carboxylic functionalized MasterBeads with 500 nm in size) were purchased from SuperMed Trading Co. Ltd. (Shanghai, China). Other chemicals were analytical pure reagents. Deionized water was used in this work.

2. Synthesis of the SPIOs@PVP-PEI@MOF-PBA IMNCs

2.1 Synthesis of the Fe_3O_4 nanoparticles (SPIOs)

The SPIOs were synthesized by a typical hydrothermal reaction. Briefly, $FeCl_3 \cdot 6H_2O$ (1.157 g), Na_3CT (0.4 g) an NH_4Ac (3.303 g) were dissolved in EG (60 mL) in the Teflon-line stainless-steel autoclave under magnetic stirring. After stirring for one hour, the magnetic stirring bar was removed. The autoclave was heated to 200 °C and maintained for 16 h. When cooling to room temperature, the sediment were collected by an external magnetic field, and then washed with ethyl alcohol and water for several times. Finally, the SPIOs were re-dispersed in deionized water (15 mL) for subsequent use.

2.2 Synthesis of the SPIO@PVP-PEI nanospheres

The above SPIOs suspension (5 mL) was added to the mixture aqueous solution (20 mL) containing PVP (1 g) and PEI (0.4 g) under magnetic stirring for 12 h. The black products were collected using a magnet, then washed with water and DMF alternately for several times. Finally, the SPIO@PVP-PEI nanospheres were re-dispersed in DMF (2 mL) for subsequent use.

2.3 Synthesis of the SPIOs@PVP-PEI@MOF-PBA IMNCs (SPMB IMNCs)

The above SPIO@PVP-PEI nanospheres (1 mL) was dispersed in the mixture solution containing ACN (8 mL) and DMF (8 mL), $Fe(NO_3)_3 \cdot 9H_2O$ (320 mg), and PBA (160 mg). Then, the mixture was heated at 120°C for 2 h under vigorous stirring. After that, the brown products were collected by magnetic separation and washed with

DMF, ethanol, and water for five times, respectively. Finally, the obtained SPMB IMNCs were dispersed in deionized water (1 mL) for further use.

2.4 Synthesis of the SPIOs-free MOFs

The $Fe(NO_3)_3 \cdot 9H_2O$ (320 mg), and PBA (160 mg) were dispersed in the mixture solution containing ACN (8 mL) and DMF (8 mL) in the Teflon-line stainless-steel autoclave. Then, the mixture was heated at 120°C for 3 days. After that, the brown red products were collected by centrifugation at 10000 rpm for 15 min and washed with DMF and ethanol for several times, respectively. Finally, the obtained pure SPIOs-free MOFs were dried under vacuum condition.

2.5 Synthesis of SPIOs@PVP-PEI@MOF-PTA nanocomposites

The traditional SPIOs@PVP-PEI@MOF-PTA nanocomposites were prepared as above mentioned method, except that PTA was used instead of PBA as organic ligands of MOF.

2.6 Synthesis of SPIOs@PBA nanospheres

The SPIOs@PBA nanospheres were prepared by coating PBA layer on the surface of SPIOs. The above SPIOs suspension (2.5 mL) was added to the mixture solution containing DMF (6 mL), H_2O (6 mL) and PBA (200 mg) under magnetic stirring for 12 h. The black products were collected using a magnet, then washed with DMF and water alternately for several times. Finally, the SPIOs@PBA nanospheres were re-dispersed in water (1 mL) for use.

3. Protein experiments

BSA and TRF were dissolved in Britton-Robinson (BR) buffer (pH 7) to obtain 1 mg mL^{-1} of protein solution with 20% ACN, respectively. The protein mixture (TRF + BSA) were prepared with equal volume of BSA, TRF and ACN. The SPIOs@PVP-PEI@MOF-PBA IMNCs (30 μL, 900 μg) were added into the protein mixture (30 μL, 20 μg) under shaking at 35 °C for 1 h. The IMNCs were removed from the protein solution using a magnet within 10 s. The supernatant was collected for further sodium dodecyl sulfatepolyacrylamide gel electrophoresis analysis (SDS-PAGE). The protein bonded IMNCs were then washed trice with BR buffer (pH 7, 100 μL) and resuspended in protein-free BR buffer (pH 7, 20 μL). Then, 12 μL of the first supernatant, the protein-material suspension or the standard protein mixture solution was preprocessed via incubation with 5 μL of loading buffer at 99 °C for 5 min, respectively. Finally, 10 μL of the samples was characterized by SDS-PAGE analysis.

For protein release from the IMNCs, the protein-material composites were shaken in 40μL of BR buffer at pH 7 or pH 9 for another one hour after washing with BR buffer (pH 7, 100 μL). The eluent was collected for further SDS-PAGE analysis. The IMNCs after elution treatment were then washed thrice with BR buffer (pH 7 or pH 9, 100 μL) and resuspended in protein-free BR buffer (pH 7 or pH 9, 20 μL) for SDS-PAGE analysis.

The protein capture and release in the protein mixture with increased amount of BSA also was carried out. As above mentioned method, except that 2 mg mL^{-1} of BSA was used instead of 1 mg mL^{-1} of BSA.

The protein capture and release of other materials were carried out as above mentioned method, except that the commercial magnetic beads, the SPIOs@PVP-PEI@MOF-PTA nanocomposites or SPIOs@PBA nanospheres were used instead of SPIOs@PVP-PEI@MOF-PBA IMNCs.

4. Characterization

The morphologies of the samples were observed by scanning electron microscopy (SEM, Hitachi S-4800, Japan) and transmission electron microscopy (TEM, JEM-2010, Japan electronic). The zeta potential and size distribution

of the samples were calculated via dynamic light scattering (DLS, Zetasizer Nano ZS90, Malvern Company). Fourier transform infrared spectra (FTIR, PE spectrometer) were recorded with wave number range 500-4000 cm^{-1}. Powder X-ray diffraction (XRD, X' Pert Pro MPD, Philips, Netherlands) was employed to study the crystal structure of samples with angles ranging from 10° to 80°. Thermogravimetric analysis (TGA) measurements were performed with simultaneous thermal analysis (STA 449 C Jupiter, NETZSCH). The mass loss of the dried sample was monitored under N_2 at temperatures from 35 to 800 °C with a heating rate of 10 K min^{-1}. The magnetization of the dried sample was measured by a vibrating sample magnetometer (VSM, Model BHV-525, Riken Japanese Electronics Company) with field from 0 to 18,000 Oe at 300 K. Nitrogen adsorption and BJH pore size distribution were obtained at 77 K on a surface area and pore size analyzer (QuadraSorb SI, America).

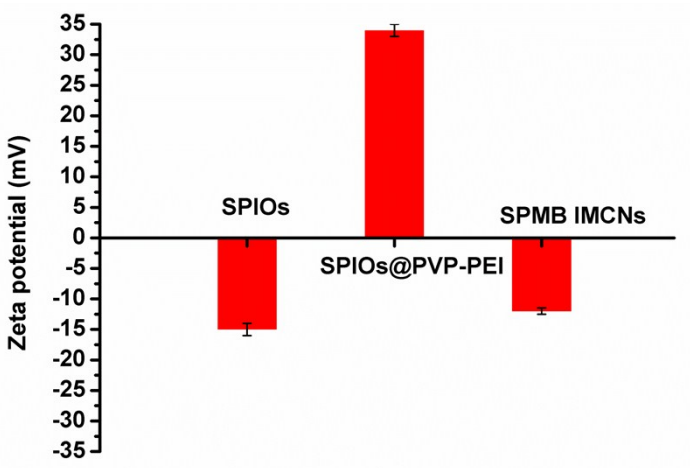
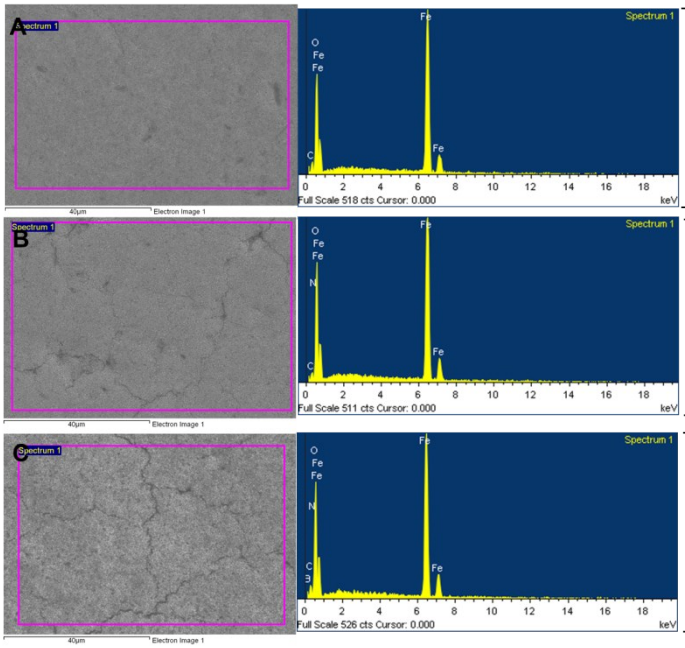


Fig. S1 Zeta poteintial of the SPIOs, SPIOs@PVP-PEI nanospheres, and SPMB IMNCs.



Element	Weight%	Atomic%
C	10.04	21.69
O	31.59	51.21
Fe	58.36	27.10
Totals	100.00	

Element	Weight%	Atomic%
C	8.20	14.92
N	12.04	18.80
O	35.94	49.12
Fe	43.82	17.16
Totals	100.00	

Element	Weight%	Atomic%
B	9.55	16.02
C	12.20	18.42
N	13.98	18.10
O	32.88	37.27
Fe	31.39	10.19
Totals	100.00	

Fig. S2 The energy dispersive X-ray (EDX) spectrum of the SPIOs (A), SPIOs@PVP-PEI nanospheres (B) and SPMB IMNCs (C).

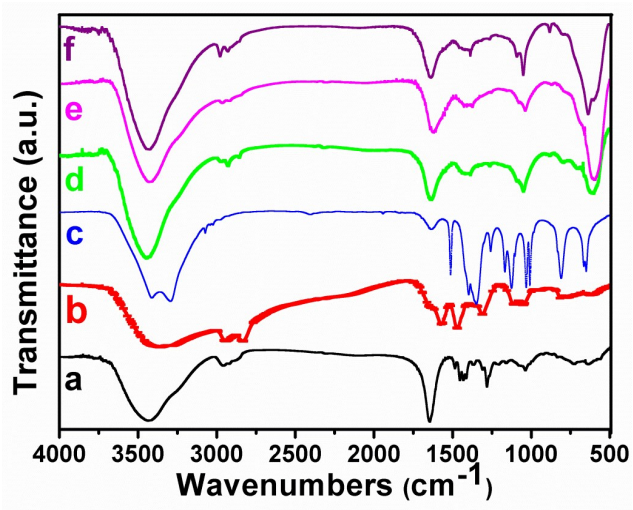


Fig. S3 FTIR spectra of (a) PVP, (b) PEI, (c) PBA, (d) SPIOs, (e) SPIOs@PVP-PEI nanospheres, and (f) SPMB IMNCs.

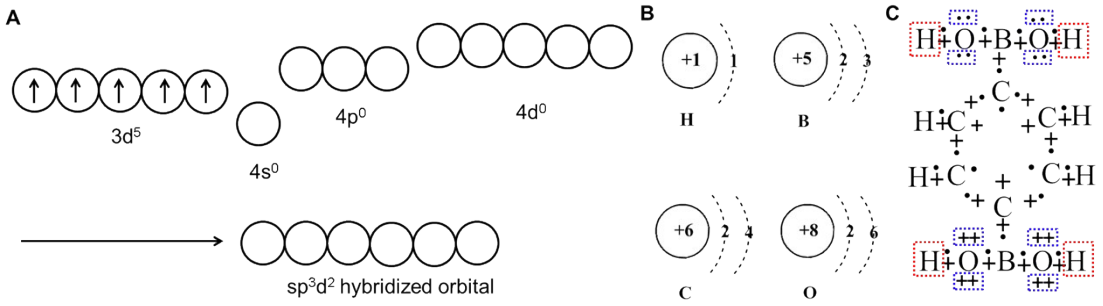


Fig. S4 (A) The electron structure of Fe^{3+}. Fe ions would offer six free orbitals via sp^3d^2 hybridization. (B) The electronic configuration of H, B, C, and O atoms. The numbers of the out most shell electron were 1 for H, 3 for B, 4 for C, 6 for O, respectively. (C) The structure of PBA. The blue frame is the lone paired electron which could possibly coordinate with Fe ions. The red frame is the hydrogen atom which could possibly be lost to form ester with *cis*-glycans of glycoproteins. The coordination with Fe ions and the recognizing glycoproteins were independent and without interference.

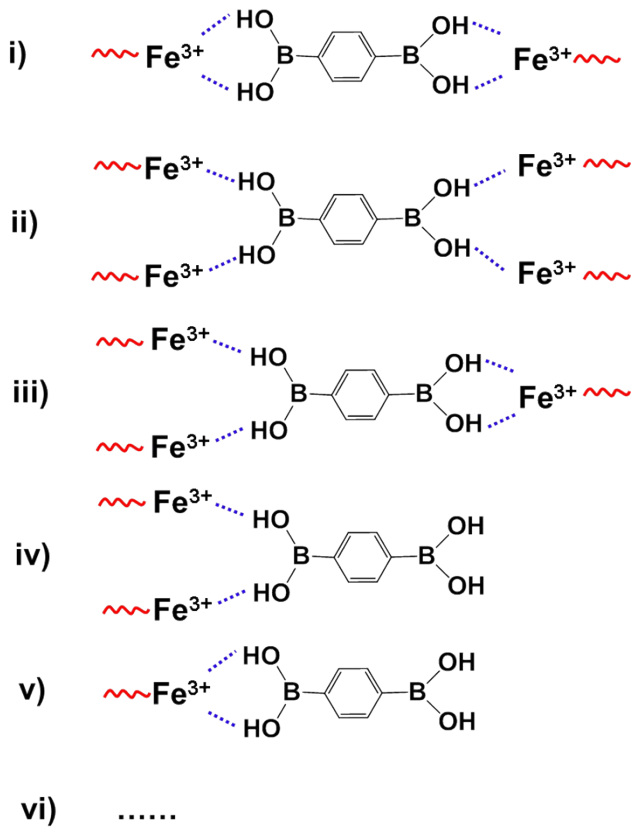


Fig. S5 The possible bonding ways of Fe^{3+} and PBA.

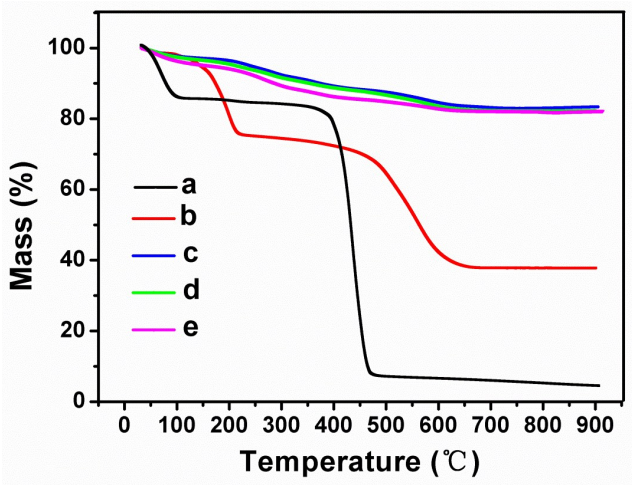


Fig. S6 TGA curves of (a) PVP, (b) PBA, (c) SPIOs, (d) SPIOs@PVP-PEI nanospheres, and (e) SPMB IMNCs.

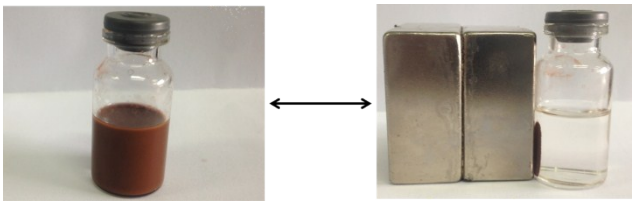


Fig. S7 The photos of the aqueous dispersion of SPMB IMNCs before and after separation with a magnet for 10 seconds.

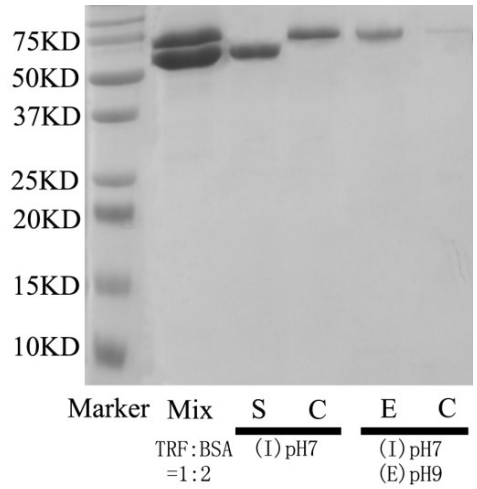


Fig. S8 SDS-PAGE analysis of the the supernatant (S), protein-material composites (C), eluate (E) of protein mixture (TRF + BSA) after treatment with the SPMB IMNCs. Lane 1, marker; Lane 2, protein mixture (TRF + BSA) before treatment; Lanes 3 and 4, supernatant and protein-material composites after treatment at pH 7; Lanes 5 and 6, eluate and protein-material composites with incubation at pH 7 and elution at pH 9. (Incabution condition: SPMB IMNCs, 900 μg; TRF, 10 μg; BSA, 20 μg)

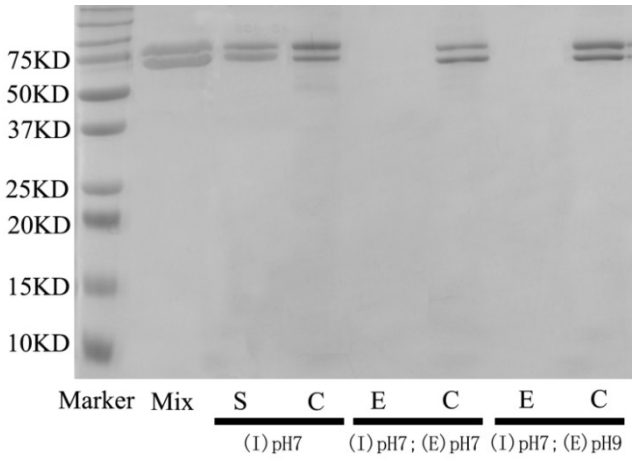


Fig. S9 SDS-PAGE analysis of the the supernatant (S), protein-material composites (C), eluate (E) of protein mixture (TRF + BSA) after treatment with the carboxylated commercial magnetic beads. Lane 1, marker; Lane 2, protein mixture (TRF + BSA) before treatment; Lanes 3 and 4, supernatant and protein-material composites after treatment at pH 7; Lanes 5 and 6, eluate and protein-material composites with incubation at pH 7 and elution at pH 7; Lanes 7 and 8, eluate and protein-material composites with incubation at pH 7 and elution at pH 9. (Incabution condition: carboxylated commercial magnetic beads, 900 μg; TRF, 10 μg; BSA, 10 μg)

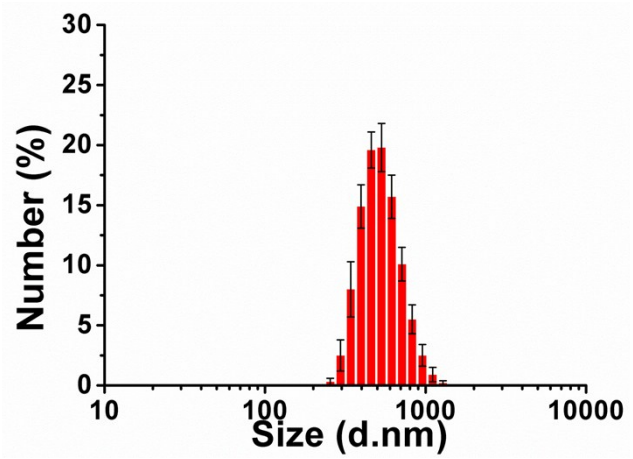


Fig. S10 Size and size distribution of the traditional SPIOs@PVP-PEI@MOF-PTA nanocomposites.

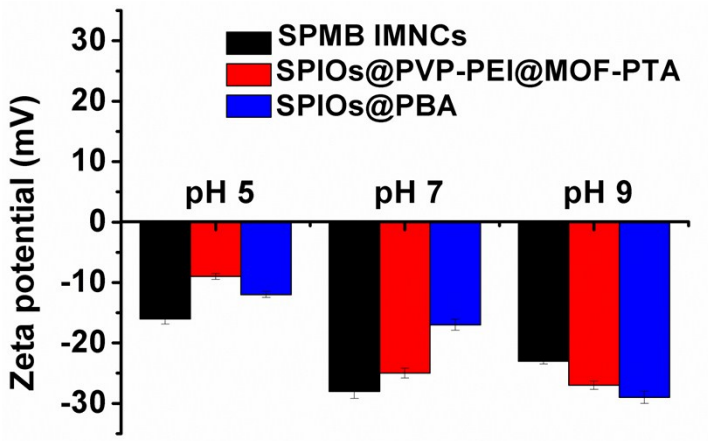


Fig. S11 Zeta potentials of the SPMB IMNCs, SPIOs@PVP-PEI@MOF-PTA nanocomposites, and SPIOs@PBA nanospheres at different pH values.

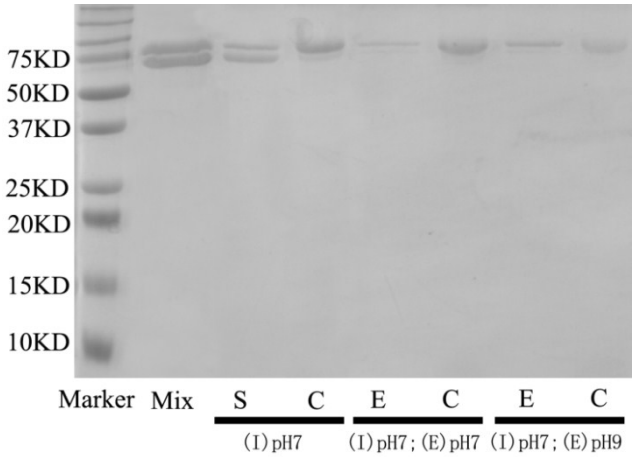


Fig. S12 SDS-PAGE analysis of the the supernatant (S), protein-material composites (C), eluate (E) of protein mixture (TRF + BSA) after treatment with SPIOs@PVP-PEI@MOF-PTA nanocomposites. Lane 1, marker; Lane 2, protein mixture (TRF + BSA) before treatment; Lanes 3 and 4, supernatant and protein-material composites after treatment at pH 7; Lanes 5 and 6, eluate and protein-material composites with incubation at pH 7 and elution at pH 7; Lanes 7 and 8, eluate and protein-material composites with incubation at pH 7 and elution at pH 9. (Incabution condition: SPIOs@PVP-PEI@MOF-PTA nanocomposites, 900 μg; TRF, 10 μg; BSA, 10 μg)

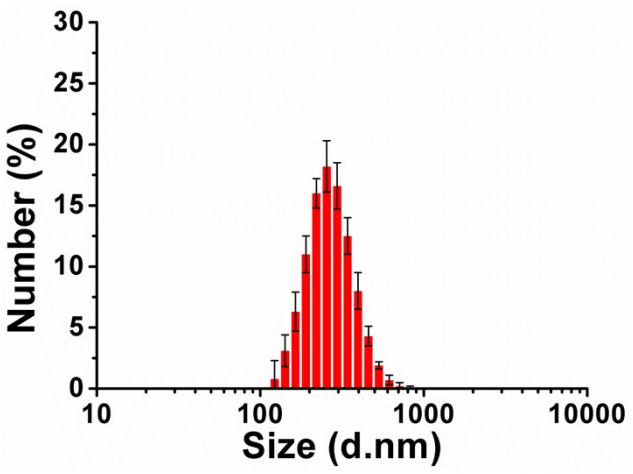


Fig. S13 Size and size distribution of the traditional SPIOs@PBA nanospheres.

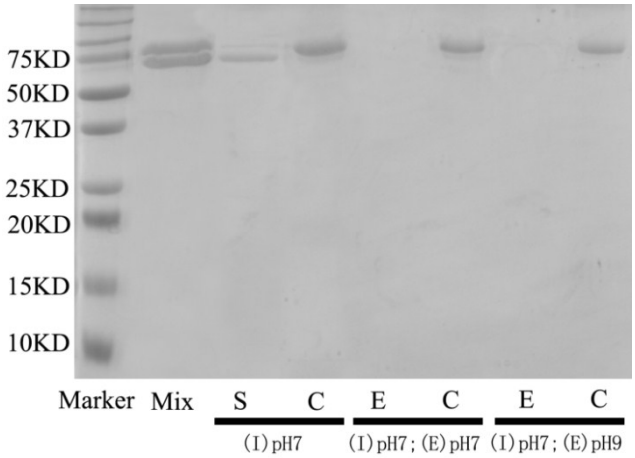


Fig. S14 SDS-PAGE analysis of the the supernatant (S), protein-material composites (C), eluate (E) of protein mixture (TRF + BSA) after treatment with SPIOs@PBA nanospheres. Lane 1, marker; Lane 2, protein mixture (TRF + BSA) before treatment; Lanes 3 and 4, supernatant and protein-material composites after treatment at pH 7; Lanes 5 and 6, eluate and protein-material composites with incubation at pH 7 and elution at pH 7; Lanes 7 and 8, eluate and protein-material composites with incubation at pH 7 and elution at pH 9. (Incabution condition: SPIOs@PBA nanospheres, 900 μg; TRF, 10 μg; BSA, 10 μg)