Supporting Information

Ensuring high selectivity for preconcentration & detection of ultra-trace cadmium by phage-functionalized metal-organic framework

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1. Experimental details

1.1. Materials and reagents

All the reagents used in the present study were analytical reagent grade. Deionized (DI) water (18 MΩ cm^{-1}) was used throughout.

A phage display heptapeptide library together with its host cell *E. coli* ER2738 (Ph.D., Phage Display Peptide Library Kit) was purchased from NEB (New England Bio-Laboratories, USA). Affimag™ γ-Fe_2O_3 (4-5 μm) was purchased from Baseline Chromtech Research Centre (Tianjin, China). Ni-NTA-Sefinose resin used for metal-loaded resin preparation was obtained from Bio Basic Inc. (Canada). The centrifugal filter device (100 kDa, 4 mL) for phage isolation was obtained from Millipore (MA, United States). LB medium (10 g of tryptone, 5 g of yeast extract, 5 g of NaCl in 1 L of deionized (DI) water, pH 7.0-7.2) with 20 mg L^{-1} tetracycline was used for host cell culture and phage amplification. All solutions were sterilized by autoclave procedure (120°C, 20 min) and the pipette tips equipped with filter cartridge were used throughout.

Zirconium chloride ($ZrCl_4$) was purchased from the Shanghai Aladdin Bio-Chem Technology Co., Ltd. (Shanghai, China). 2-aminoterephthalic acid (BDC-NH_2) was purchased from Sigma-Aldrich (St. Louis, USA). N, N-dimethylformamide (DMF) and chloroauric acid ($HAuCl_4$) were the products of the Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China).

Stock solution of cadmium (1000 mg L^{-1}) was prepared by dissolving appropriate amount of cadmium pellets in 1.0 mol L^{-1} of HNO_3. Working standards of various concentrations were obtained by diluting the stock solution. pH value of the sample solution was adjusted by using HNO_3 (1.0 mol L^{-1}) or NaOH (2.0 mol L^{-1}). Nitrate solution of metals including K^+, Na^+, Ca^{2+}, Mg^{2+}, Zn^{2+}, Fe^{3+}, Cu^{2+}, Hg^{2+}, Ni^{2+} and Pb^{2+} were prepared for the interference studies.

1.2. The preparation of cadmium loaded magnetic microbeads and metal-chelating resin

2 mL of Affimag^{TM} γ-Fe_2O_3 (1 % (w/v)) was mixed with 4 mL of iminodiacetic acid (IDA, 5 % (w/w), pH 9.16) and incubated in 4 mL of $NaHCO_3$-Na_2CO_3 (0.1 mol L^{-1}, pH 9.5) buffer solution for 24 h (37℃, 250 rpm). Then the IDA functionalized magnetic microbeads were rinsed for 3 times with DI water and stored at 4℃ in PBS buffer solution (pH 7.4). For the preparation of Cd^{2+} immobilized magnetic microbeads, 500 μL Affimag^{TM} γ-Fe_2O_3-IDA microbeads (1 % (w/v)) was incubated in 5 mL of Cd^{2+} solution (0.1 mol L^{-1}) overnight (37℃, 250 rpm). It is then rinsed for 3 times by PBS buffer solution (pH 5.8) and stored in PBS (pH 5.8) at 4℃ for further use.

M^+-loaded resin was prepared by replacing the preloaded Ni^{2+} by metal ions on the Ni-NTA-Sefinose resin. The resin beads were first washed with EDTA solution (0.5 mol L^{-1}, pH 8.0) to strip off Ni^{2+}. The metal-free resin M^- was obtained until the resin turned from blue to colorless. The M^- resin were rinsed for 3 times with DI water and stored at 4℃ for further use. Resin loading other metals was prepared in similar procedure. 1 mL of M^- resin suspension was incubated in 5 mL of metal ion solution (0.1 mol L^{-1}, M^+ represents Zn^{2+}, Pb^{2+}, Hg^{2+}, Fe^{3+} and Cu^{2+}) overnight with gentle shaking. It is then rinsed with buffer solution and stored at 4°C.

1.3. Screening for Cd(Ⅱ) binding phage

60 μL of the phage library (2×10^{12} virions, library complexity (unique clones) = ~2.8×10^9 as claimed by NEB) was dissolved in 940 μL of sterilized DI water. Then it was mixed with 500 μL of Cd^{2+} loaded magnetic microbeads and 200 μL of M^+-loaded resin beads together. The mixture was added into 1500 μL sterilized DI water and incubated for 30 min (150 rpm, 25°C). The resin beads and supernatant were discarded and the magnetic microbeads were collected with magnet and washed three times with sterilized DI water to remove the loosely bonded phages. 2 mL of EDTA solution (0.5 M, pH 8.0) was added to microbeads and shook for 40 min (250 rpm,

25℃). The eluate was further centrifuged for 15 min (8000 rpm) with a centrifugal filter device with MWCO of 100 kDa (5000 rpm, 4℃). Phages retained on the filter membrane were carefully collected, amplified, and tittered for the next round of screening. The amplified phages were purified by precipitation with PEG/NaCl (20 % (w/v) poly (ethylene glycol) (PEG)-8000, 2.5 M NaCl). 10 μL of the phage was used for phage tittering using X-gal/IPTG agar plates.

2. Supplemental tables and figures

Table S1. Temperature program of the GFAAS instrument for cadmium detection.

Step	Temperature/^{o}C	Ramp/s	Hold/s
Drying	100	15	10
Pyrolysis	350	15	10
Atomization	1900	1	3
Cleaning	2100	1	3

Table S2. Elemental percentage in UiO66-NH_2 and UiO66-NH_2@phage.

Element	Peak/eV	UiO66-NH_2 atomic/%	UiO66-NH_2@phage atomic/%
C (1s)	284.8	76.66	76.73
O (1s)	532.6	21.42	21.20
N (1s)	399.1	1.14	1.26
Zr (3d5)	182.3	0.77	0.70
Au (4f7)	83.8	0	0.11

Table S3. The kinetic parameters of Cd^{2+} adsorption by UiO66-NH_2@phage.

Adsorbent	pseudo-first-order kinetic			pseudo-second-order kinetic		
	K_f/min^{-1}	q_e/μg g^{-1}	R^2	K_s/g μg^{-1}min^{-1}	q_e/μg g^{-1}	R^2
UiO66-NH_2@phage	1.76	31.2	0.8881	0.182	13.6	0.9994

Table S4. *Langmuir* and *Freundlich* model parameters of Cd^{2+} adsorption by UiO66-NH_2@phage.

Adsorbent	*Langmuir* model			*Freundlich* model		
	K_L/L μg^{-1}	q_m/μg g^{-1}	R^2	K_F/μg g^{-1}	N	R^2
UiO66-NH_2@phage	1.01	621	0.9999	368	8.45	0.9112

Table S5. The comparison of the proposed approach in terms of analytical performances with other methods reported in the literatures for the preconcentration & detection of Cd^{2+} by SPE-AAS.

Adsorbent	Detection	LOD /ng L^{-1}	EF	Sample volume/mL	Ref.
Fe_3O_4@$FePO_4$	ETAAS	21	10.3	2	[38]
BSA-NFs	FAAS	370	40	20	[39]
L-cystine modified zeolite	FAAS	40	400	100	[40]
Amine functionalized resin	FAAS	1500	——	50	[41]
Ionic liquid modified Fe_3O_4	FAAS	500	50	50	[42]
Ionic liquid-modified TiO_2	FAAS	100	——	100	[43]
Modified nanoporous carbon	FAAS	30	165	75	[44]
Ionic imprinted polymers	ETAAS	160	12.5	25	[45]
Modified Fe_3O_4	FAAS	3710	100	100	[46]
Graphene oxide	ETAAS	5	15	1.5	[47]
Ion imprinted mesoporous silica	GFAAS	6.1	50	25	[48]
Modified chitosan	FAAS	21	90	900	[49]
UiO66-NH_2@phage	GFAAS	3.9	17.4	1	This work

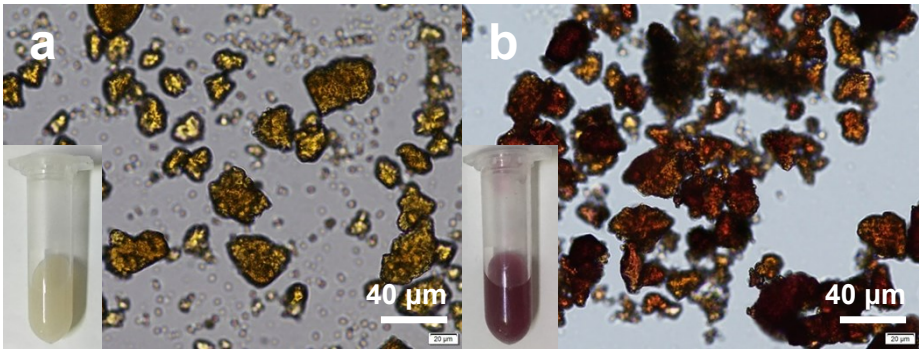

Fig. S1. Optical microscopic images of (a) UiO66-NH_2 and (b) UiO66-NH_2@phage. Inset: photograph of (a) and (b) under daylight.

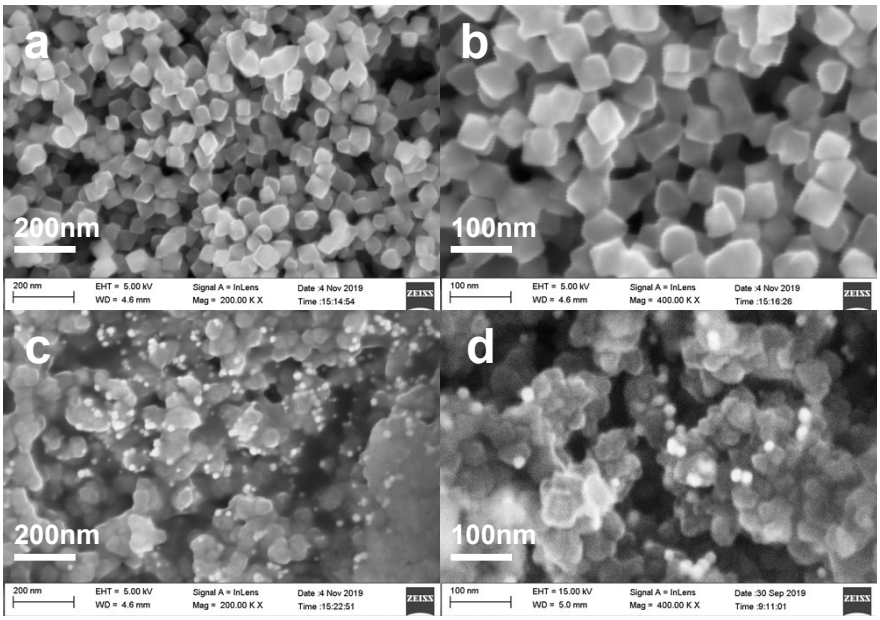

Fig. S2. SEM images of UiO66-NH_2 (a-b) and UiO66-NH_2@phage (c-d).
(a) UiO66-NH_2 (200000×), (b) UiO66-NH_2 (400000×), (c) UiO66-NH_2@phage (200000×), (d) UiO66-NH_2@phage (400000×).

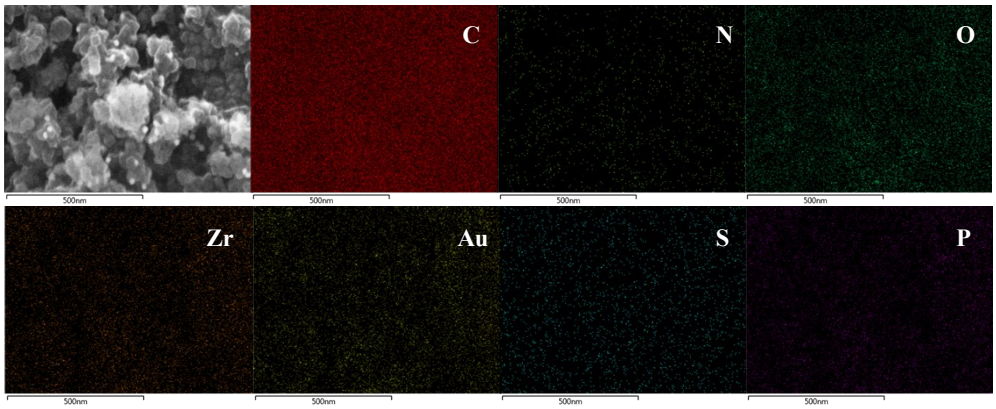

Fig. S3. EDS map of element distribution.

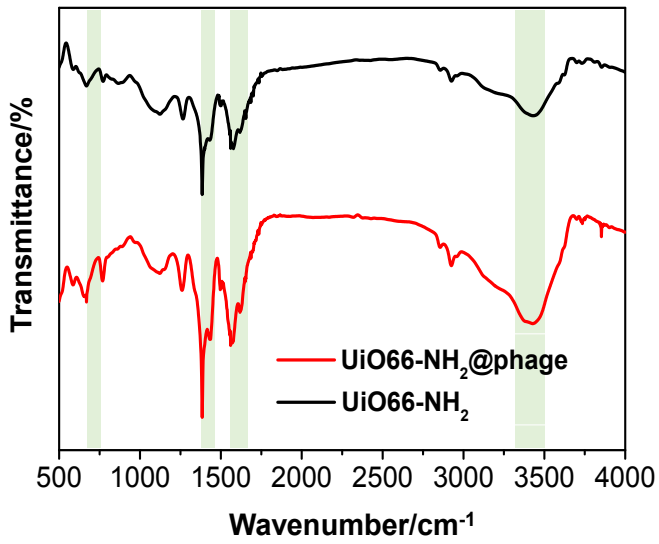

Fig. S4. FT-IR spectra of UiO66-NH_2 and UiO66-NH_2@phage.

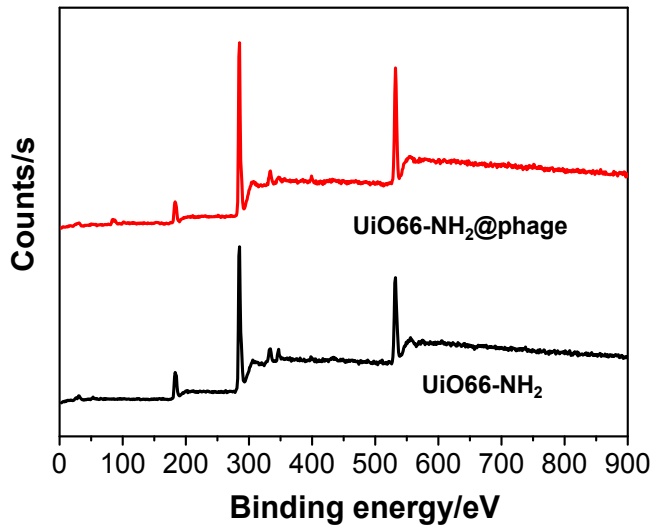

Fig. S5. XPS spectra of UiO66-NH_2 (bottom) and UiO66-NH_2@phage (top).

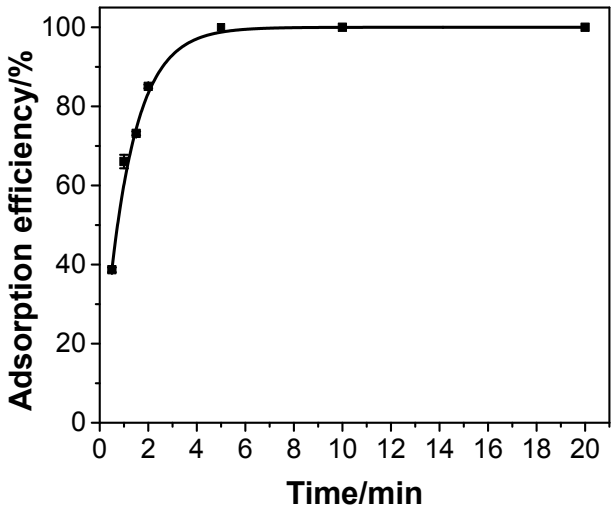

Fig. S6. The effect of reaction time on the adsorption efficiency of Cd^{2+} by the UiO66-NH_2@phage adsorbent. Sample solution: 1.0 mL (2.0 μg L^{-1} Cd^{2+}); pH: 6.0; The mass of adsorbent: 0.15 mg.

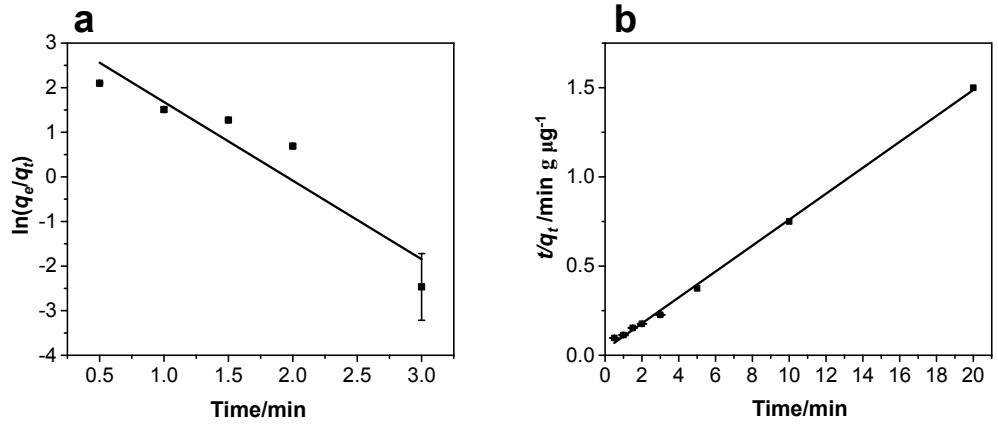

Fig. S7. (a) Pseudo-first-order kinetic and (b) pseudo-second-order kinetic plots of Cd^{2+} adsorption by UiO66-NH_2@phage. Sample solution: 1.0 mL (2.0 μg L^{-1} Cd^{2+}); pH: 6.0; The mass of adsorbent: 0.15 mg.

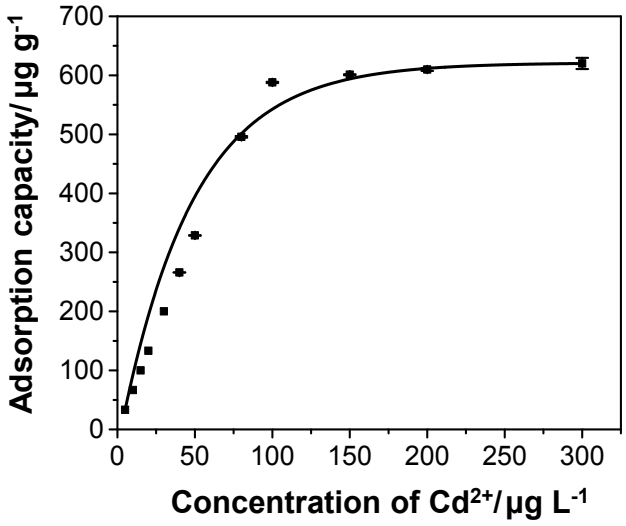

Fig. S8. The adsorption isotherm of Cd^{2+} by UiO66-NH_2@phage. Sample solution: 1.0 mL, 5-300 μg L^{-1} Cd^{2+}; pH: 6.0; The mass of adsorbent: 0.15 mg; Adsorption time: 20 min.

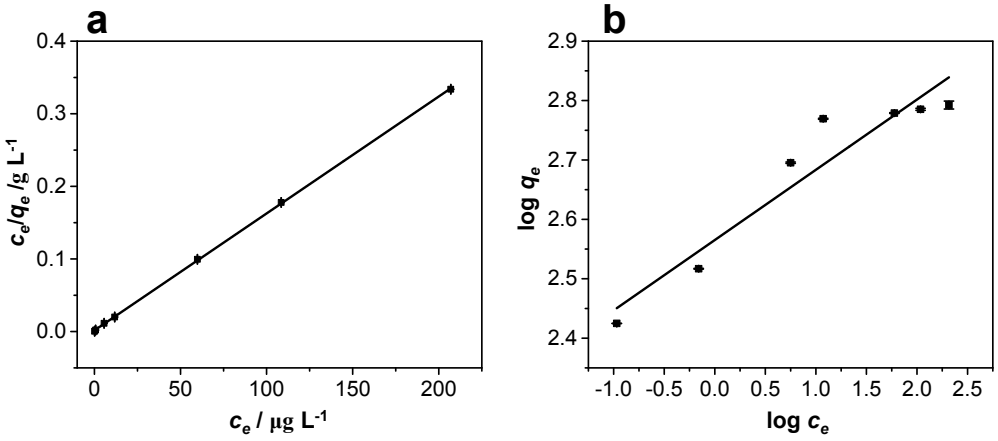

Fig. S9. Linear regression of the (a) *Langmuir* and (b) *Freundlich* adsorption isotherm. Sample solution: 1.0 mL, 5-300 μg L^{-1} Cd^{2+}; pH: 6.0; The mass of adsorbent: 0.15 mg; Adsorption time: 20 min.