Electronic supplementary information

Strategic design and fabrication of lightweight sesame ball-like hollow double-layer hybrid magnetic molecularly imprinted nanomaterials for the highly specific separation and recovery of tetracycline from milk

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Materials and regents

Dopamine (DA) was obtained from Alfa Aesar Chemical Company. Ferrous chloride ($FeCl_2 \cdot 4H_2O$), ferric chloride hexahydrate ($FeCl_3 \cdot 6H_2O$), ethanol, methanol, trihydroxymethyl aminomethane (Tris), sodium carbonate (Na_2CO_3), ammonia (NH_3), hydrochloric acid (HCl), and acetic acid (HAc) were acquired from Xi'an Chemicals Ltd. Tetracycline (TC), chloramphenicol (CAP), sulfadiazine (SD), doxycycline (DC), oxytetracycline (OTC), tetraethyl orthosilicate (TEOS), chlortetracycline (CTC), and estradiol (E2), were provided from Aladdin Ltd. (Shanghai, China). A WaterPro water system (Axlwater Corporation, TY10AXLC1805-2, China) was utilized to obtain highly filtered water. All of the reagents were of analytical grade and were utilized without further processing. The milk sample was bought at a local supermarket.

Instrumentation and conditions

The morphologies of the obtained nanomaterials were portrayed with field transmission electron microscope (TEM, jem-2100, Japan). The magnetic properties of the resultant materials were measured at room temperature using a vibrating sample magnetometer (VSM, MPMS-squid VSM-094). The Fourier transform infrared (FTIR) spectra of prepared nanomaterials in the region of 500-4000 cm^{-1} were analyzed by Nicolet AVATAR 330 FTIR spectrophotometer. The true densities of prepared nanomaterials were tested by true density analyzer (AccuPyc II 1345, micromeritics, America). The crystal forms of magnetic samples were obtained by Rigaku D/max/2500v/pcXRD diffractometer (Rigaku Co., Japan) with Cu K_α radiation. The HPLC analyses were performed on a Hitachi L-2130 HPLC system including L-2130

pump, L-2400 detector, L-2300 column oven, and WondaSil C18 column (5 μm particle size, 150 mm×4.6 mm). The column temperature was 40 °C. The mobile phase was methanol-0.05% acetic acid solution (25/75, v/v) transmitted for 15 min at a flow rate of 1.0 mL min^{-1}. The injection volume was kept as 10 μL and the column effluent was monitored at 350 nm.

The ratio of molecularly imprinted sites binding to TC

The total number of imprinted sites in HD-MMIPs is calculated according to the total amount of TC in the eluents after synthesis. As shown in Table S7, the total amount of TC eluted from HD-MMIPs is about 11.8 mg, which is equivalent to 2.7×10^{-5} mol of TC. The final mass of obtained nanomaterials is 223 mg, so the total number of imprinted sites per unit mass (N_T) of HD-MMIPs is 1.2×10^{-7} mol mg^{-1} calculated according to Eq. (S1).

Generally, specific imprinted sites and non-specific imprinted sites can contribute to sorption. The adsorption capacity (Q_{NIP} = 23.48 mg g^{-1}) of non-molecularly imprinted polymers (HD-MNIPs) is generally attributed to non-specific imprinted sites. And the adsorption capacity (Q_{MIP} = 70.23 mg g^{-1}) of molecularly imprinted polymers (HD-MMIPs) is attributed to the sum of specific imprinted sites and non-specific imprinted sites. Thus, according to Eq. (S2), the adsorption amount of TC adsorbed through the imprinted sites (Q_I) is 46.75 mg g^{-1}, which is equivalent to the number of imprinted sites binding to TC per unit mass (N_I) of HD-MMIPs is 1.1×10^{-7} mol mg^{-1}.

The formula of the ratio is defined as Eq. (6). According to the calculation, the ratio

of molecularly imprinted sites binding to TC is 91.7%.

$$N_{\mathrm{T\,or\,I}} = \frac{n_{\mathrm{T\,or\,I}}}{m} \quad \text{(S1)}$$

$$Q_{\mathrm{I}} = Q_{\mathrm{MIP}} - Q_{\mathrm{NIP}} \quad \text{(S2)}$$

Where n_T is the amount of substance eluted, n_I is the amount of substance absorbed through the imprinted sites, m refers the mass of final obtained nanomaterials, N_T represents the total number of imprinted sites per unit mass. N_I represents the number of imprinted sites binding to TC per unit mass. Q_{MIP} and Q_{NIP} (mg g^{-1}) are respectively the adsorption capacity of imprinted nanomaterials and non-imprinted nanomaterials. Q_I is the adsorption amount of TC adsorbed through the imprinted sites. $Ratio_{I/T}$ is the ration of the number of imprinted sites binding to TC in HD-MMIPs to the total number of imprinted sites in the HD-MMIPs.

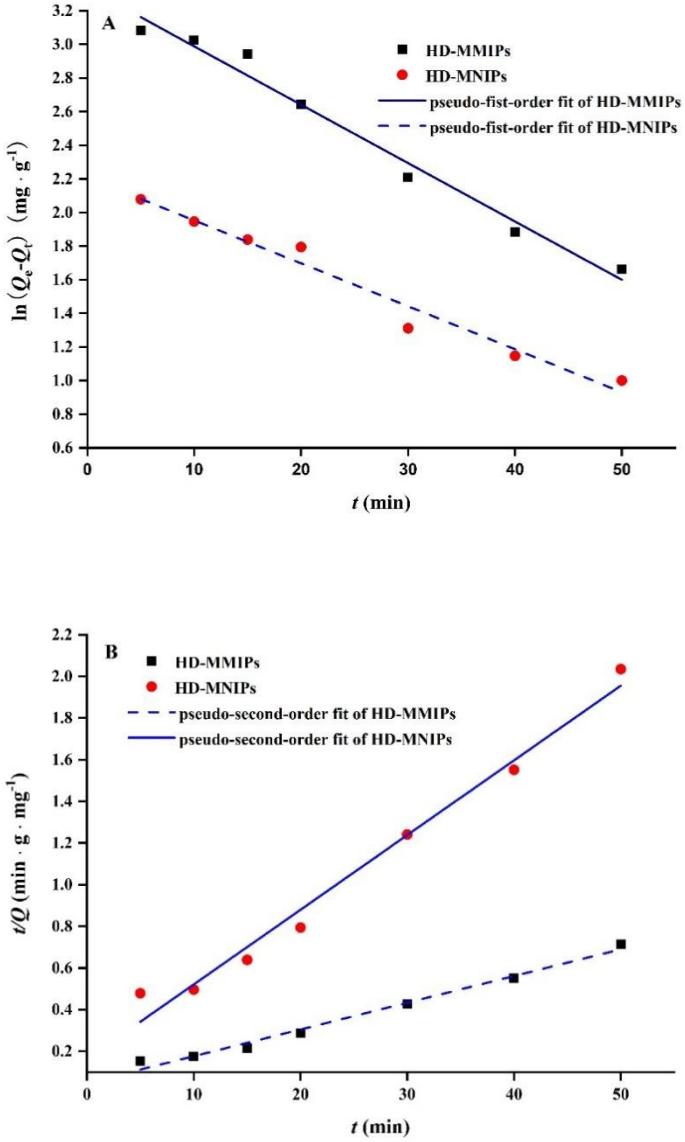

Fig. S1 pseudo-fist-order model and pseudo-second-order model to evaluate the kinetic mechanism of HD-MMIPs and HD-MNIPs.

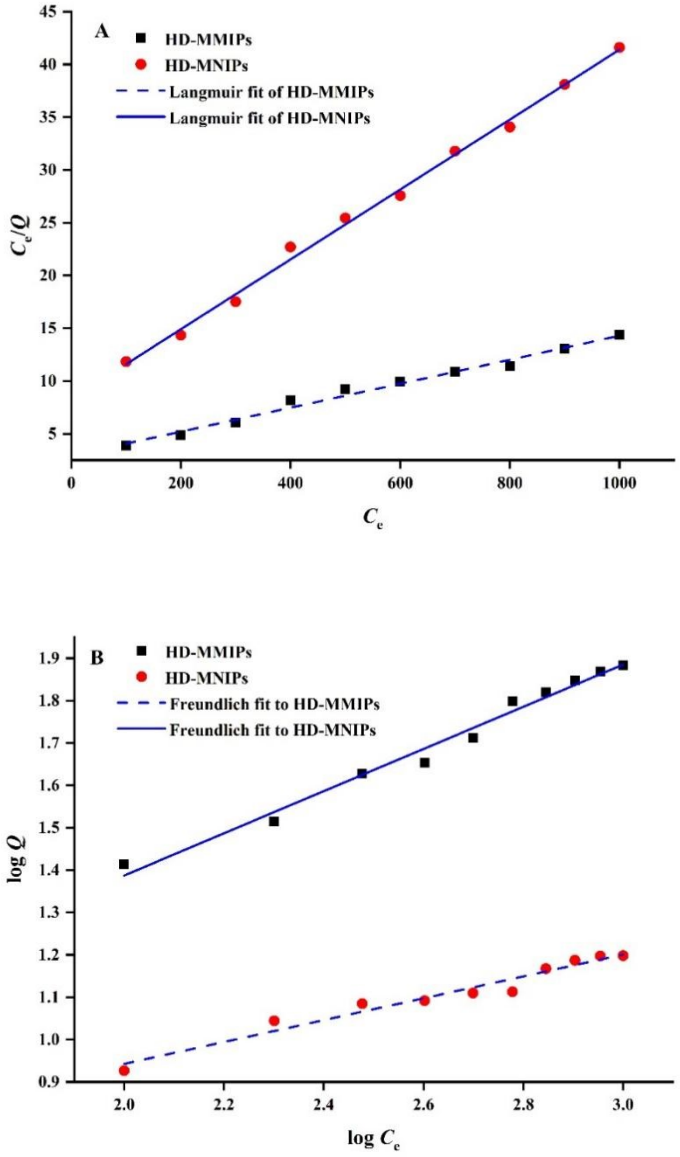

Fig. S2 *Langmuir* thermodynamic model (A) and *Freundlich* thermodynamic model (B) to evaluate the isothermal mechanism of HD-MMIPs and HD-MNIPs.

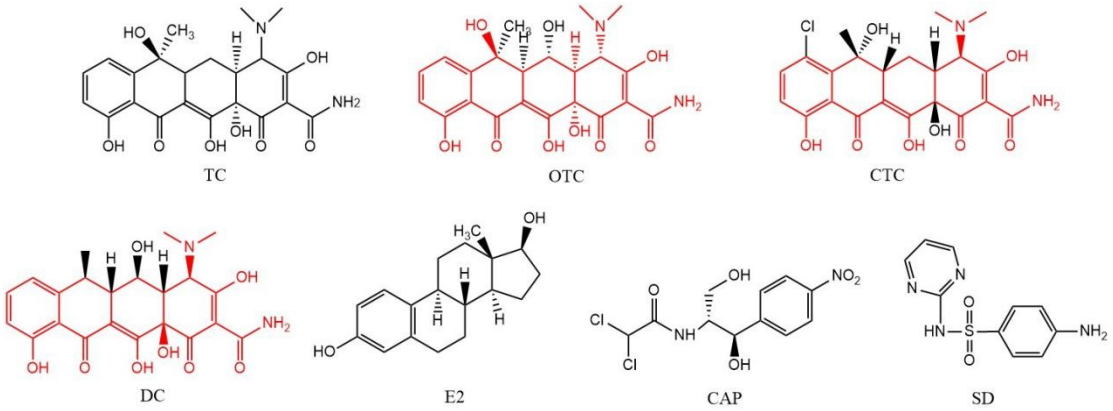

Fig. S3 Chemical structures of TC and six competitors.

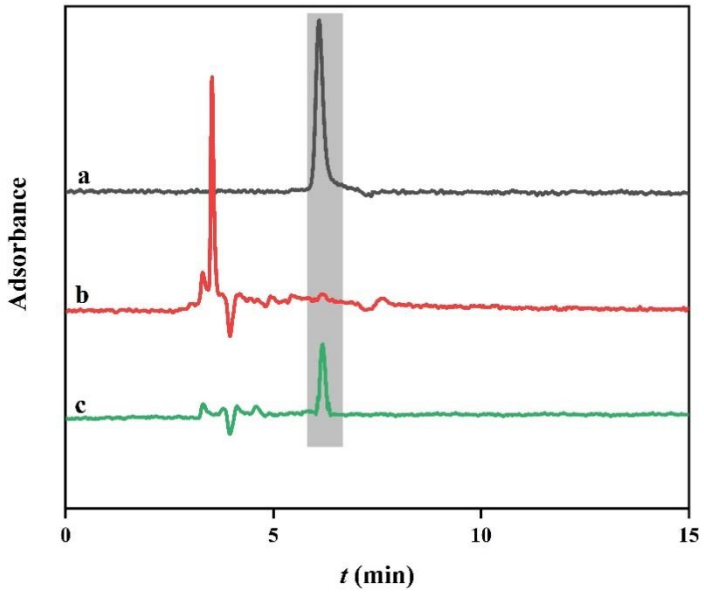

Fig. S4 Chromatograms of TC standard solution (a), treated milk sample spiked with TC at the concentration of 10 ng mL^{-1} (b), and elution of adsorbed HD-MMIPs (c).

Table S1 Equations and parameters of adsorption kinetics of HD-MMIPs and HD-MNIPs.

Model	Equations and parameters	HD-MMIPs	HD-MNIPs
pseudo-first-order	Equation	$\ln (Q_e-Q_t) =0.0347t+3.335$	$\ln (Q_e-Q_t) = 0.0205t+2.209$
	Q_e (mg g^{-1})	28.08	9.107
	K_1 (g mg^{-1} min^{-1})	0.0347	0.0205
	R^2	0.9665	0.9634
pseudo-second-order	Equation	$t / Q_t = 0.0128t + 0.0482$	$t / Q_t = 0.0359t + 0.1625$
	Q_e (mg g^{-1})	78.13	27.86
	K_2 (g mg^{-1} min^{-1})	0.0034	0.0079
	V_0 (mg g^{-1} min^{-1})	20.75	6.154
	R^2	0.9952	0.9931

Table S2 Equations and parameters of adsorption thermodynamic of HD-MMIPs and HD-MNIPs.

Model	Equations and parameters	HD-MMIPs	HD-MNIPs
Langmuir thermodynamic model	Equation	$C_e / Q = 0.0114C_e + 2.935$	$C_e / Q = 0.0311C_e + 8.286$
	Q_{max} (mg g^{-1})	87.72	30.20
	K_L (mg g^{-1})	0.0038	0.0039
	R^2	0.9988	0.9954
Freundlich thermodynamic model	Equation	$\log Q = 0.4977 \log C_e + 0.3921$	$\log Q = 0.2581 \log C_e + 0.4266$
	K_F (mg g^{-1})	2.466	2.671
	m	0.4977	0.2581
	R^2	0.9715	0.9534

Table S3 Selectivity adsorption parameters of HD-MMIPs and HD-MNIPs.

Adsorbates	Q_{MIPs}(mg g^{-1})	Q_{NIPs}(mg g^{-1})	*IF*	*SC*
TC	70.23	23.48	2.99	—
CTC	49.34	23.53	2.10	1.42
DC	48.67	23.23	2.10	1.42
OTC	42.35	24.50	1.73	1.73
E2	13.07	9.98	1.31	2.28
CAP	18.14	13.10	1.38	2.16
SD	19.12	14.26	1.34	2.23

Table S4 Reproducibility of HD-MMIPs.

Polymer	Batch	1	2	3	4	5	6	Average
HD-MMIPs	Q (mg g^{-1})	73.36	70.25	68.18	69.09	69.75	69.86	70.08
	RSD	3.9	2.6	6.8	4.6	2.2	4.3	4.06

Table S5 Density of S-MMIPs, HS-MMIPs, D-MMIPs, and HD-MMIPs.

Polymers	Density (g cm^{-3})
S-MMIPs	2.2115
HS-MMIPs	2.0773
D-MMIPs	2.3723
HD-MMIPs	2.2269

Table S6 Validation parameters of the proposed method (n = 5).

Analyte	Linearity range (ng mL^{-1})	R^2	RSDs (%)		LOD (ng mL^{-1})	LOQ (ng mL^{-1})	Recovery (%)		
			Intra-day	Inter-day			1.0 (ng mL^{-1})	5.0 (ng mL^{-1})	10 (ng mL^{-1})
TC	1.0-200	0.9991	1.8-3.6	2.5-4.5	0.83	1.25	94.8	96.8	98.5

Table S7. The concentration and amount of TC in the eluent [a].

Eluent	Concentration (mg mL^{-1})	Amount (mg)
1st	0.62	6.2
2st	0.19	1.9
3st	0.12	1.2
4st	0.08	0.80
5st	0.076	0.76
6st	0.042	0.42
7st	0.028	0.28
8st	0.013	0.13
9st	0.010	0.10
10st	0.001	0.01
Sum	/	11.8

[a] The nanomaterials are dispersed in 10 mL of ethanol-HAc (99:1, v/v) mixture each time followed by shaking for 2 h to elute the TC.