Electronic Supplementary Information (ESI)

Online highly-selective recognition of domoic acid by aptamer@MOFs affinity monolithic column with HPLC for shellfish safety monitoring

1. Experimental

1.1 Characterization of affinity monoliths

Structural morphology of the monolith was measured by Scanning electron microscope (Verios G4). FTIR spectra were measured by Fourier infrared spectrometer (Nicolet is50). Energy-dispersive spectrum (EDS) was got by the electron probe microanalyzer (Helios G4 CX). The contact angle (θ) of water on the affinity monolithic surface was measured by optical contact angle measurement (KRUSS DSA25S).

1.2 Permeability

Permeability was measured by using a LC-20AD pump (Kyoto, Japan) delivering methanol (η = 0.544 mPa·s) with different flow rates and the resulting backpressure was recorded. The permeability was calculated as below:

$$K = \frac{\eta L^2}{\Delta P t_2}$$

K is the permeability (m^2), η is the viscosity of the solvent (Pa·s), L is the length of the capillary (m), ΔP is the measured backpressure (Pa), and finally t_2 is the breakthrough time of mobile phase (min).

1.3 *SPE-LC-MS for DA analysis*

1.3.1 Sample pretreatment

The HLB solid-phase extraction column (500 mg, 6 mL, Waters Sep-pak) was treated with methanol (5 mL) and then equilibrated with Milli-Q water (5 mL). A rice sample of 40.0 g was weighted and crushed into the powder. The 4 g NaCl and 100 mL of the solution

(acetonitrile:water = 9:1, v:v) were added and vibrated by vortex for 2 min. By means of a 0.22 μm membrane, the mixture was filtered. Then a 15 mL of filtrate was adopted and diluted with 45 mL of water. A series of fortified rice samples spiked with ZEN were prepared at some concentrations ranging from 0.00 μg/kg to 10.00 μg/kg. Then, an aliquot of fortified rice sample (1 mL) was loaded onto the HLB cartridge and washed sequentially with Milli-Q water (1 mL), then the wash was discarded. After air-drying the column, the elution of ZEN was carried out with methanol (1 mL), and then the eluate was filtered through 0.22 μm filter membrane prior to the injection into HPLC-MS.

13.2 ***LC-MS***

The fortified rice samples were analyzed by a LCMS-8040 system (Shimadzu，Japan). ZEN was analyzed on a reversed-phase HPLC system (LC-20A, Shimadzu, Japan) using a Shim-pack GIST C-18 column (150 length × 2.1 mm i.d. and 5 μm particle size) and a Mass spectrometry (LCMS-8040, Shimadzu). With a gradient program at a flow rate of 0.3 mL/min, the samples were separated using a mobile phase composed of aqueous phase containing water (A) and acetonitril (B). Initial elution was performed with the mobile phase A:B = 75:25 (v/v). After 5 min, the linear gradient reached 70% of mobile phase B. After 9 min, the linear gradient reached 75% of mobile phase A, and the gradient composition returned to the initial elution and was maintained for 1 min.

The injection volume was 10 μL. Heat block temperature and DL temperature were 400 C and 250 C, respectively. Nebulizing gas flow and driving gas flow were 3 L/min and 15 L/min, respectively. Mass spectrometry was operated in electrospray ionizationin negative mode (ESI-) with multiple reaction monitoring (MRM) and the optimized mass spectrometry parameters were shown as below. The precursor-to-product ion transition m/z 317.2→175.0 was selected for the quantitative calculation of ZEN and the m/z 317.2→273.2 was used as confirmation transition.

2. Supplementary data

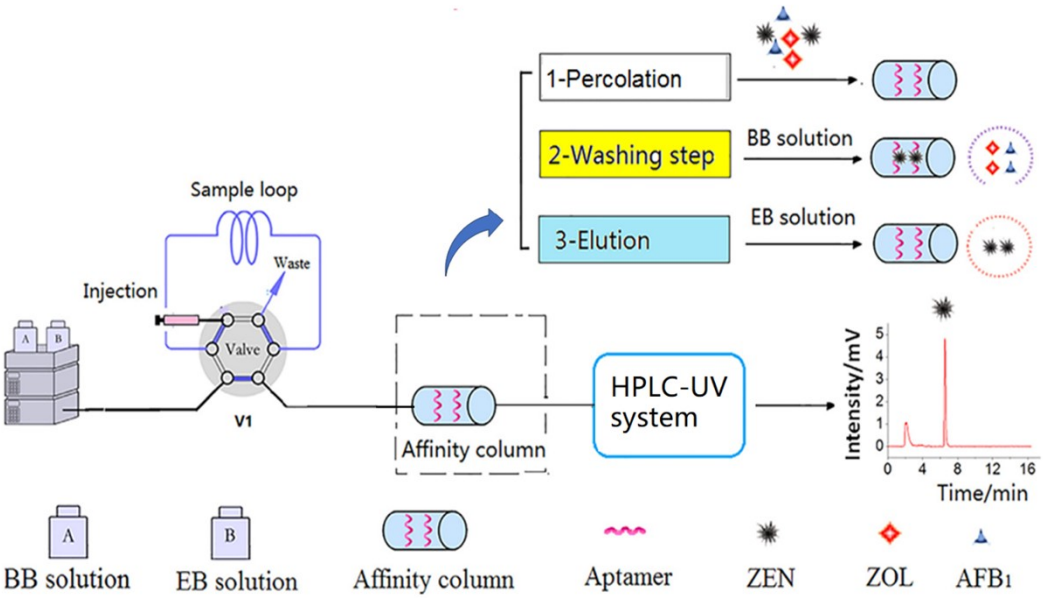

Fig. S1 Process of online recognition of DA using affinity monolith coupled with HPLC-UV

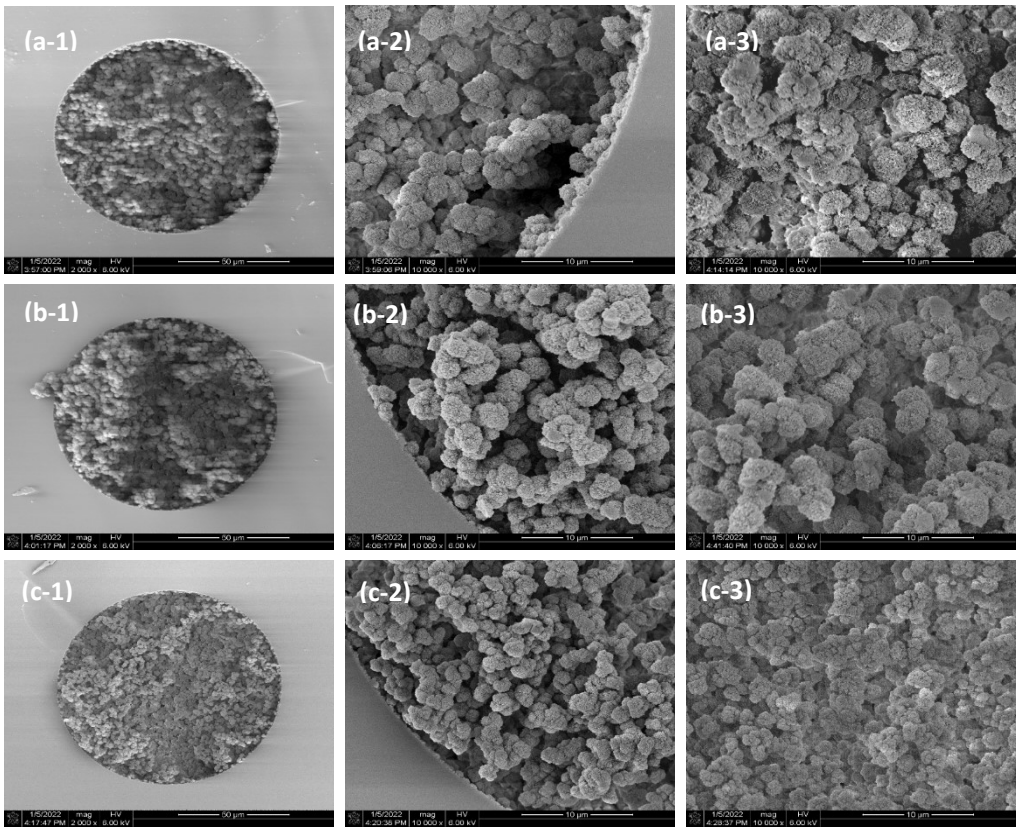

Fig. S2 Morphologies of various monoliths prepared with the recipes a-c
Numbers 1,2 and 3 represent the morphology of the polymer at different locations, 1 is the cross section, 2 is the tube wall, and 3 is the tube center.

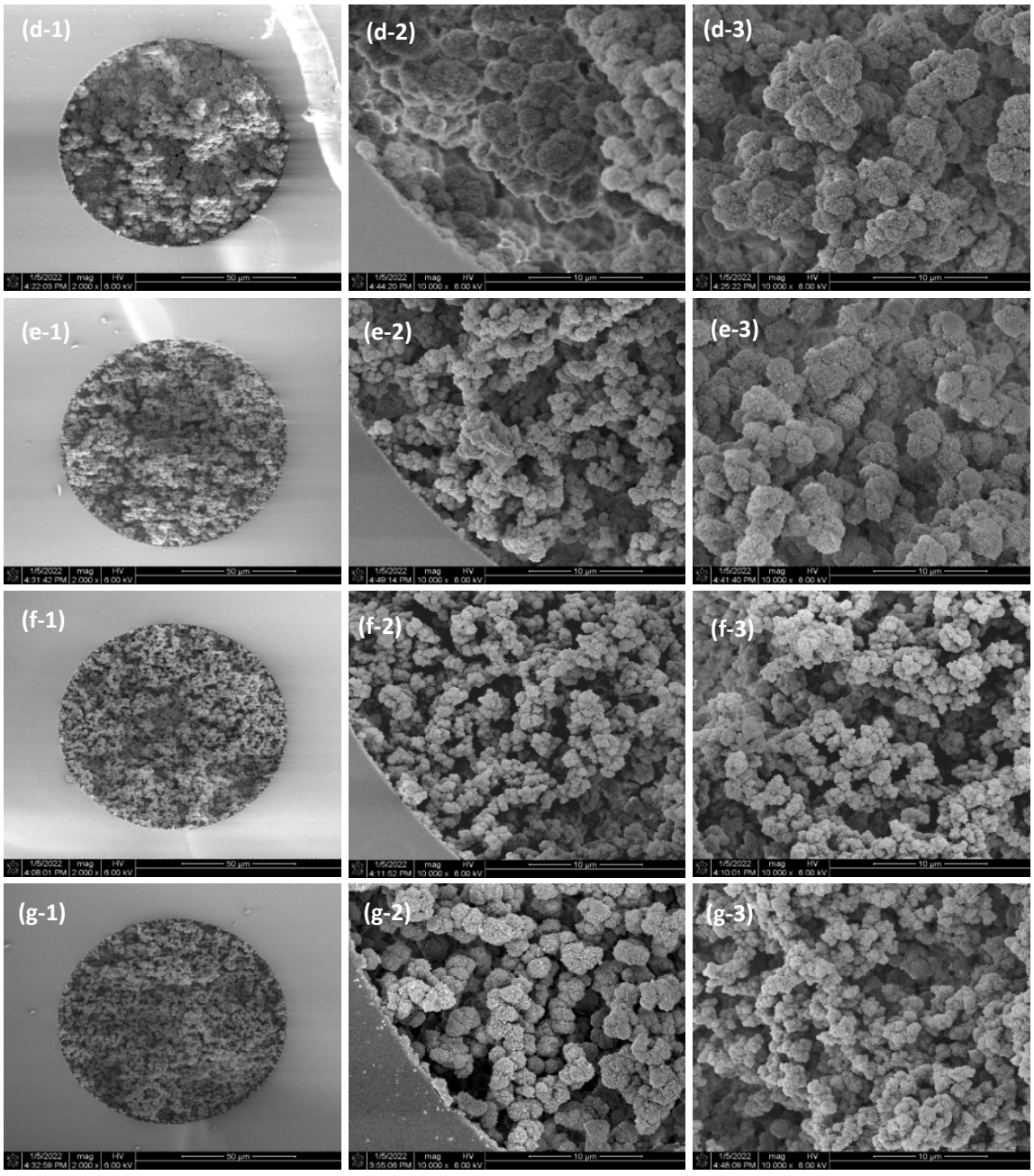

Fig. S3 Morphologies of various monoliths prepared with the recipes d-g. Numbers 1, 2 and 3 represent the morphology of monolith observed at different locations, 1 refers to the cross section, 2 refers to the tube wall, and 3 refers to the center of capillary monolithic column.

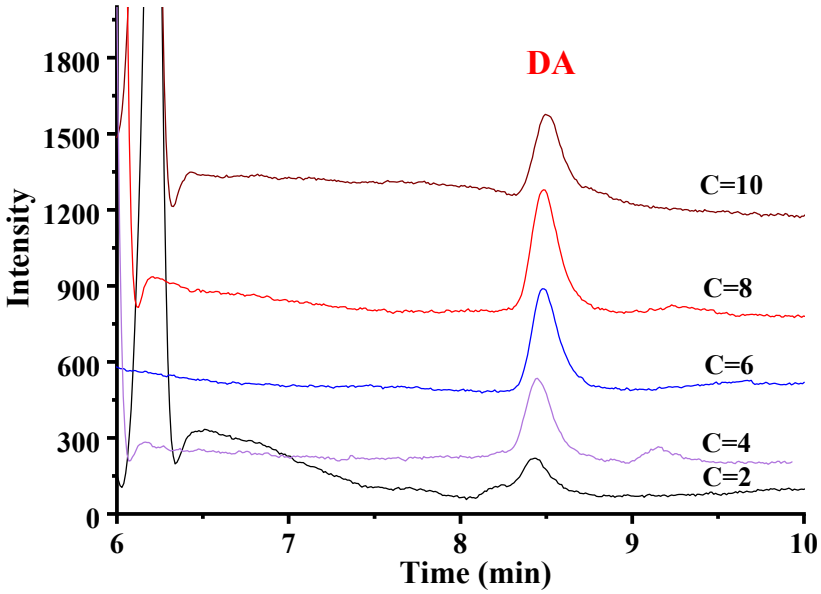

Fig. S4 Influence of cyclic assembly of ZIF-8 in monolithic column on affinity recognition. DA: 50 ng/mL; Effective length of monolith: 5cm.

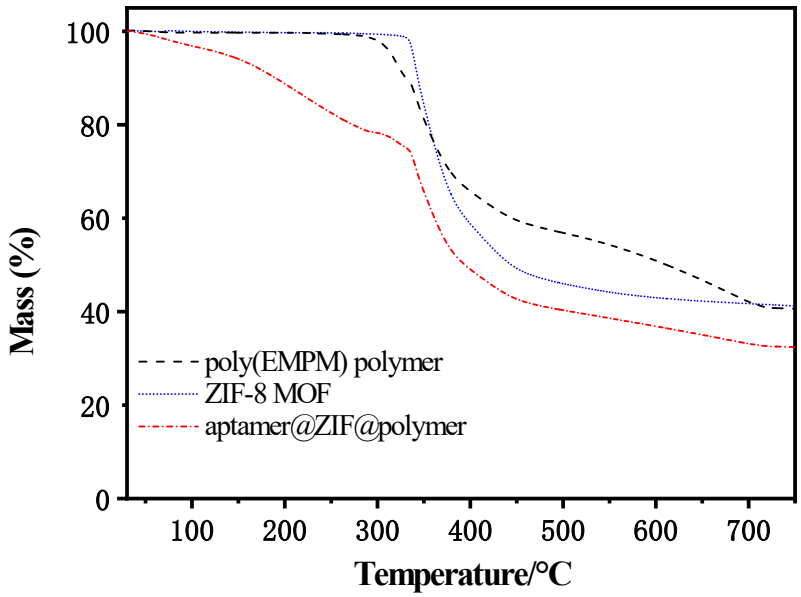

Fig. S5 TGA of ZIF-8, poly（EMPM）polymer and polyEMPM@ZIF-8@aptamer monolith

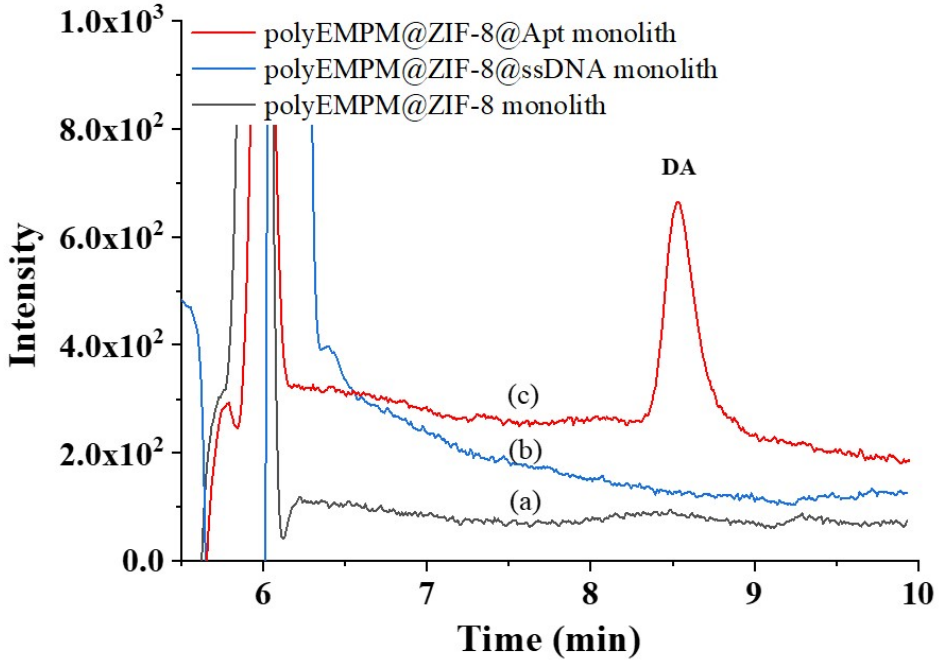

Fig.S6 Specificity performance of various monolithic columns for DA

(a) ZIF-8@poly(EMPM) monolith; (b) ssDNA@ ZIF-8@poly(EMPM) monolith; (c) aptamer@ ZIF-8@poly(EMPM) monolith

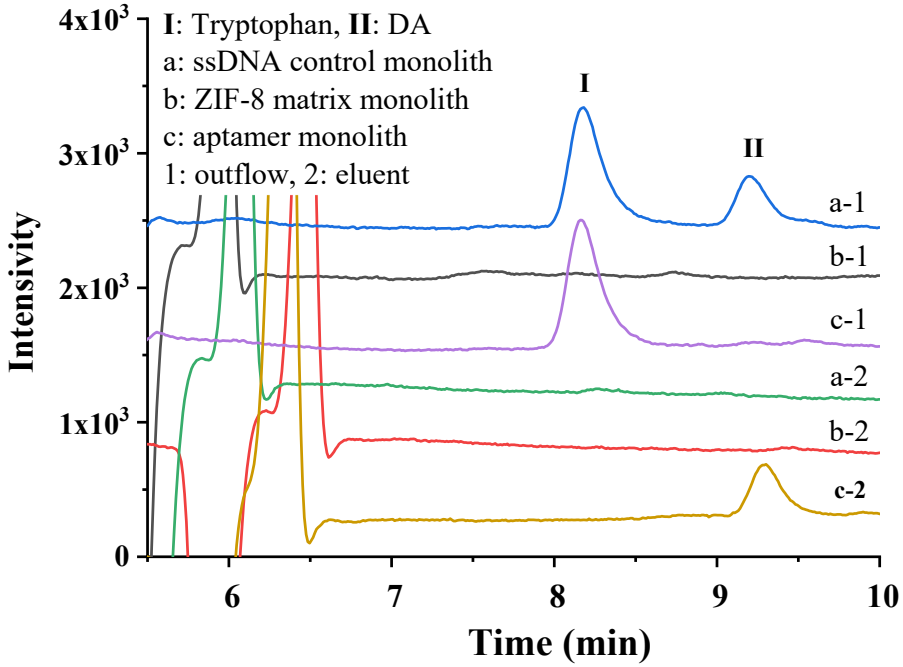

Fig. S7 Cross reactivity of different monolithic columns for DA under the interference of tryptophan

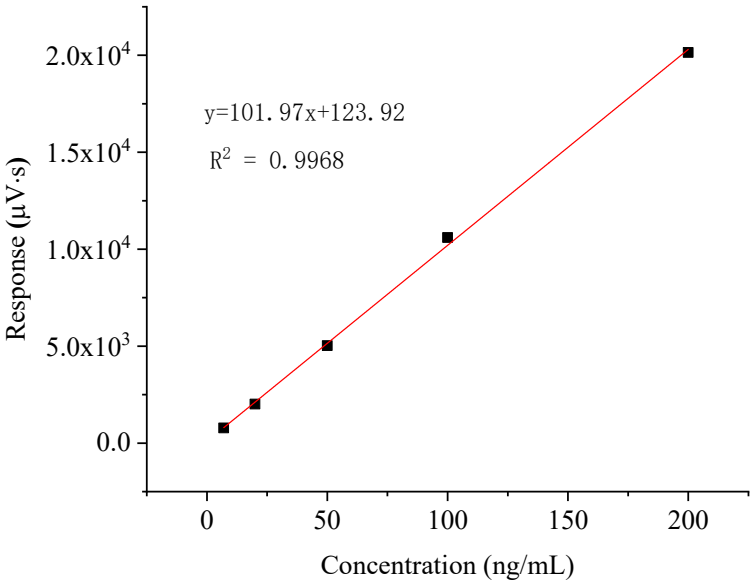

Fig. S8 Calibration curve for the detection of DA

Table S1 Element analysis of different monolithic column stationary phases.

Element	Poly(Epoxy-MA-co-POSS-MA) (%)	ZIF8@poly(Epoxy-MA-co-POSS-MA) (%)	ZIF8@Apt modified poly(Epoxy-MA-co -POSS-MA) (%)
C	56.42	51.26	51.29
O	43.58	39.47	40.25
N	0.00	8.32	6.46
Zn	0.00	0.95	0.75
P	0.00	0.00	1.25

Table S2 Coverage density of aptamer on affinity monolithic columns.

Column	n_1(nmol)	n_2(nmol)	ρ (pmol/μL)	ρ_{mean} (pmol/μL)
1	1.50	0.28	3106.7	
2	1.50	0.25	3183.1	3140.7
3	1.50	0.27	3132.2	

Table S3 Equation of calibration curve, linear concentration range and limit of detection of this method

Linear equation	R^2	Linear concentration range (ng/mL)	LOD (ng/mL)
y=101.97x+123.92	0.9968	20.0~200.0	7.0

Table S4 Reproducibility of affinity monolithic column

Colum	Concentration of DA (ng/mL)	Recovery yields of DA (%)	RSD (%)
Run-to-Run(n=3)	50.0	95.6±1.4	1.5
Day-to-Day(n=3)	50.0	96.7±1.5	1.6
Batch-to-Batch(n=3)	50.0	97.3±1.6	3.3