Electronic Supporting Information

A triazine-based conjugated microporous polymer composite for magnetic solid phase extraction of 5-nitroimidazoles coupled with UPLC-MS/MS for quantitation

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1. Chemical structures of the five 5-NDZs



Fig. S1 Chemical structures of the five 5-NDZs. a. MNZ; b. RNZ; c. SNZ;

d. TNZ; e. ONZ.



2. Choice of sorbent and investigation of the amount of TCMP in MTCMP

Fig. S2 Effects of influencing factors on recovery: (A) the amount of TCMP in MTCMP (B) Elution solvent volume for MTCMP containing 1.25 g of TCMP (C)

The type of sorbent (D) Elution solvent volume for TCMP

3. Optimization of extraction process

The enrichment efficiency of materials to the targets is affected by many factors. In order to achieve higher extraction efficiency, a series of enrichment conditions were systematically optimized including the amount of adsorbent, the concentration of the salt (NaCl), the pH of the sample solution, extraction time, the type of elute solvent, the volume of elute solvent, and the desorption time. In the optimized experiments, 10 mL of ultrapure water spiked with five 5-NDZs at the concentration of 5 ng·mL⁻¹ were applied as the simulated water samples. All experiments were conducted in triplicate.

The amount of adsorbent is an important factor affecting the recovery, in the experiment 1-11 mg of MTCMP was tested, as we can see from Fig. S3 (A), with the increasing of the amount of the material from 1 to 7 mg, the recoveries of the analytes increased gradually, and then leveled off with the further increasing from 7 to 11 mg. While the recovery of TNZ decreased all the time, the reason is the same as "3.1. Choice of materials". Take all into consideration, the amount of the adsorbent was fixed at 7 mg in the following tests.

Ionic strength can influence the extraction efficiency by affecting the solubility of the 5-NDZs and changing the speed of mass transfer. In this work, the ionic strength was studied by varying the concentration of NaCl from 0 to 0.10 mol·L⁻¹, the recoveries increased slightly with the increasing of salt concentration which could attribute to the reduce of the solubility of the 5-NDZs, and then decreased slowly due to the increase of the viscosity of the solution, but the change was not significant (Fig. S3 (B)), thus no NaCl was added in subsequent experiments.

The pH value of the sample solution can affect the extraction efficiency by changing the existing forms of the drug and adsorbent. The pKa of the five 5-NDZs are 2.58, 2.60, 2.30, 2.72, 2.62 for MNZ, RNZ, TNZ, ONZ and SNZ, respectively. When the pH is below 3, the 5-NDZs are protonated and positively charged, while when the pH is above 3, the 5-NDZs are deprotonated and negatively charged. As shown in Fig. S3 (C), the best recovery was obtained when the pH was 7, it can be inferred that the point of zero charge of MTCMP may be around 7^{S1}, when the pH was below 7, the triazine components of MTCMP was protonated and deprotonated when above 7. With the pH increasing from 2 to 7, the interaction between 5-NDZs and MTCMP varied from electrostatic repulsion to electrostatic attraction, and the intermolecular hydrogen bonding enhanced; while with the pH increasing continuously the interaction changed into electrostatic repulsion. Therefore, the adsorption of 5-NDZs on the MTCMP is mainly due to the π -interaction, electrostatic interaction and intermolecular hydrogen bond. Consequently, a pH value of 7 was selected for the extraction process.

In this work, vortex was applied in the extraction proess to facilitate the dispersion of the sorbent into the water sample. To achieve complete absorption, the vortex extraction time was investigated from 2 to 30 min, the Fig. S3 (D) shows that the adsorption equilibrium was reached at 15 min, hence 15 min was selected for the pretreatment procedure.

Elution solvent is another critical factor that can affect the extraction efficiency.

In consideration of the polarity of 5-NDZs, methanol, acetonitrile, acetone and acetic ether were studied. As illustrates in Fig. S3 (E), the satisfactory result was achieved with acetonitrile, so acetonitrile was selected as the optimal elution solvent.

In order to achieve complete elution, the effect of the volume of elution solvent was investigated in the range of 0.4-1.4 mL. As can be seen from Fig. S3 (F), the recoveries of all analytes reached the highest when the volume of acetonitrile was 1.2 mL, hence 1.2 mL was chosen for the elution of 5-NDZs.

Vortex elution time was tested from 2 to 25 min as presentes in Fig. S3 (G), the completely desorption of all the analytes was achieved at 10 min of elution time, and without obvious changing after 10 min, so the elution time was set at 10 min.



Fig. S3 Effects of influencing factors on recovery: (A) the amount of adsorbent (B) ionic strength (C) pH (D) extraction time (E) type of elute solvent (F) the volume of elution solvent (G) elution time. Reuseablity of the adsorbent: (H) Extraction cycles

4. Characterization of TCMP and MTCMP

The disappearance of C-Cl stretching vibration near 850 cm⁻¹ observed in the spectrum of TCMP and MTCMP (Fig. S4) indicated the complete substitution of the chloride atom of CC.^{38, S2, S3} The absorption band appearing at 1506-1480 and 1384 cm⁻¹ in the spectrum of MCTP and MMCTP were assigned to the stretching modes of CN in triazine ring.^{38, S2-S4} While the peak around 572 cm⁻¹ appeared in the spectrum of Fe₃O₄ and MMCTP originated from the Fe-O vibrations.^{18,24} All the above illustrated the successful synthesis of the materials.

The SEM pattern of TCMP (Fig. S5) revealed morphologies with smooth spherical shape. The appearance of irregular spheres was caused by the aggregation of particles.

As shown in Fig. S6 there are two broad diffraction peaks around 21° and 43° 2 θ , indicated the amorphous nature of the material. The broad diffraction peak (2 θ) at 21° could be assigned to the stacking of triazine linkage in the (001) plane^{S5, S6}.

The N₂ adsorption-desorption isotherm and density functional theory (DFT) pore size distribution of TCMP are presented in Fig. S7. The multi-point BET specific surface area was $660.4 \text{ m}^2 \cdot \text{g}^{-1}$, the dominant pores width centered at 0.60 and 1.23 nm, and the micropore volume (0.329 cm³·g⁻¹) was 76.69% of the total pore volume (0.429 cm³·g⁻¹). The prepared TCMP was of high specific surface area and porosity.

The elemental analysis result of TCMP is illustrated in Table S1. There existed slight deviation between the measured values and the theoretical calculated values. This could be attributed to the incomplete combustion and trapped adsorbates

including gases and water vapour, which were also discussed in other literatures^{S2, S7,}

S8.



Fig. S4 FTIR profiles of Fe_3O_4 , TCMP, and MTCMP



Fig. S5 SEM image of TCMP



Fig. S7 N₂ adsorption-desorption isotherms and pore size distribution of TCMP (inset)

Polymer	Calculated (%)					Found	l (%)	
ТСМР	С	Н	Ν	C/N	С	Н	Ν	C/N
	85.04	3.94	11.02	7.72	74.69	5.12	7.84	9.53

5. Gradient elution conditions and optimal mass spectrum parameters

Time(min)	A-0.1% formic acid water (%)	B-acetonitrile (%)	
0	92	8	
2	20	80	
2.5	92	8	
4	92	8	
2.5 4	92 92	8 8	

Table S2 The mobile phase composition and gradient elution conditions

Table S3 Mass spectrometry parameters for MS/MS analysis of the five 5-NDZs

Analyte	t_R (min)	Precursor ion (m/z)	Daughter ion (m/z)	Cone voltage (V)	Collision energy (eV)
MNIZ	1 2 2	$[M+H]^{+}$	127.94ª	38	12
MINZ	1.55	172.09	82.38	38	26
DNZ	1.57	$[M+H]^{+}$	140.08 ^a	5	12
KNZ	1.57	201.09	55.15	5	20
a) 17		$[M+H]^{+}$	127.94ª	5	12
SNZ	1.62	186.05	82.05	5	26
		$[M+H]^+$	120.93ª	15	14
TNZ	1.82	248.00	127.85	15	20
		[M+H]+	127 90ª	7	14
ONZ	1.96	220.12	82.18	7	28

^a Ion for quantification

6. Method validation

Water	Spiked (ng·mL ⁻¹)	Recovery (RSD) (%)						
sample		MNZ	RNZ	SNZ	TNZ	ONZ		
Tap water	Low ^a	99.53(5.66)	101.6(1.82)	102.3(7.97)	98.98(4.09)	103.5(1.07)		
	Middle ^b	95.37(2.44)	96.38(6.48)	98.98(3.79)	102.0(3.47)	91.12(8.56)		
	High ^c	93.40(1.73)	95.45(0.96)	95.06(4.34)	101.0(5.31)	102.2(4.43)		
	Low ^a	95.30(7.73)	84.54(6.96)	99.38(4.11)	100.7(6.05)	104.1(8.87)		
Drinking water	Middle ^b	89.66(3.54)	94.53(7.34)	86.31(8.21)	94.57(4.22)	95.69(6.22)		
Willor	High ^c	90.96(2.95)	92.57(0.23)	94.18(0.98)	96.16(4.47)	95.11(6.03)		
	Low ^a	101.3(5.16)	103.5(5.21)	104.1(8.28)	93.61(5.18)	108.6(3.83)		
River water ^d	Middle ^b	93.03(1.79)	89.91(4.94)	98.03(4.51)	83.52(9.58)	102.0(3.23)		
	High ^c	90.26(3.90)	94.25(1.28)	96.73(5.85)	98.82(4.18)	98.39(1.05)		
	Low ^a	97.20(7.81)	102.6(6.26)	100.3(2.09)	105.4(4.61)	95.05(0.36)		
aquaculture water	Middle ^b	94.69(6.24)	99.85(5.37)	90.87(8.42)	92.39(6.72)	99.09(2.05)		
water	High ^c	90.69(4.46)	110.9(4.43)	97.36(2.20)	98.02(5.41)	110.7(4.52)		
	Low ^a	97.69(5.21)	92.71(3.34)	104.1(4.98)	104.8(5.94)	87.96(8.47)		
Lake water	Middle ^b	96.24(6.87)	96.11(5.45)	93.24(5.62)	89.43(1.78)	99.26(3.29)		
	High ^c	92.55(6.35)	100.4(3.15)	97.38(1.00)	96.73(7.00)	93.85(4.17)		
	Low ^a	98.97(4.57)	99.36(4.91)	98.32(5.42)	88.17(5.54)	85.69(6.16)		
River water ^e	Middle ^b	91.25(1.85)	86.54(5.63)	91.18(3.55)	82.62(7.00)	105.0(4.44)		
	High ^c	88.85(2.33)	101.7(2.87)	95.07(3.50)	98.19(5.45)	97.08(7.67)		
	Low ^a	95.26(4.84)	98.50(4.74)	105.7(2.57)	101.6(3.35)	104.8(8.16)		
River water ^f	Middle ^b	93.14(4.12)	102.8(2.77)	98.00(5.07)	88.49(1.76)	95.15(5.67)		
water	High ^c	91.17(2.52)	95.35(3.81)	93.57(3.89)	97.15(6.80)	93.65(3.15)		

Table S4 Spiked recoveries of the five 5-NDZs in different water samples at three

different levels (n = 3)

 a 0.2 ng·mL $^{-1}$ for MNZ, 0.4 ng·mL $^{-1}$ for RNZ, 0.1 ng·mL $^{-1}$ for SNZ, TNZ and ONZ.

^{d, e, f} Different sampling point of the river water.

 $^{^{\}rm b}$ 1 ng·mL $^{\rm -1}$ for MNZ, 2 ng·mL $^{\rm -1}$ for RNZ, 0.5 ng·mL $^{\rm -1}$ for SNZ, TNZ and ONZ.

 $^{^{\}rm c}$ 15 ng·mL $^{\rm -1}$ for MNZ, 30 ng·mL $^{\rm -1}$ for RNZ, 7.5 ng·mL $^{\rm -1}$ for SNZ, TNZ and ONZ.

Table S5 Matrix effects (ME) and their precision of the proposed method in different

Analyte -	River water ^d (RSD) (%)			Aquaculture water (RSD) (%)			
	Low ^a	Middle ^b	High ^c	Low ^a	Middle ^b	High ^c	
MNZ	99.35(4.29)	97.72(2.82)	96.92(4.06)	98.39(6.62)	99.04(2.93)	98.26(2.94)	
RNZ	97.04(7.90)	99.65(2.28)	96.05(4.69)	91.11(3.98)	99.56(1.06)	98.49(5.12)	
SNZ	99.17(4.18)	95.70(5.35)	97.89(2.04)	94.72(4.00)	96.74(3.30)	98.24(6.02)	
TNZ	95.53(4.35)	91.52(5.73)	97.93(4.12)	94.04(4.73)	99.75(5.50)	97.09(4.27)	
ONZ	92.28(3.98)	88.05(5.71)	93.46(5.77)	98.15(4.79)	92.52(1.01)	93.84(5.34)	

water sample matrices (n = 3)

 a° 0.2 ng·mL⁻¹ for MNZ, 0.4 ng·mL⁻¹ for RNZ, 0.1 ng·mL⁻¹ for SNZ, TNZ and ONZ.

^b 1 ng·mL⁻¹ for MNZ, 2 ng·mL⁻¹ for RNZ, 0.5 ng·mL⁻¹ for SNZ, TNZ and ONZ.

^c 15 ng·mL⁻¹ for MNZ, 30 ng·mL⁻¹ for RNZ, 7.5 ng·mL⁻¹ for SNZ, TNZ and ONZ.

^d A sampling point of the river water.

Table S6 Determination of five 5-NDZs (ng·mL⁻¹) in different water samples (n = 3)

	Content of the analyte (ng·mL ⁻¹)							
Water sample	MNZ	RNZ	SNZ	TNZ (RSD) (%)	ONZ			
Tap water	_	_	_	-	-			
Drinking water	_	-	-	-	-			
River water ^d	_	_	_	0.083(5.40)	_			
aquaculture water	_	-	_	-	-			
Lake water	_	_	-	-	_			
River water ^e	_	-	_	0.078(5.23)	-			
River water ^f	_	_	_	_	_			

^{d, e, f} Different sampling point of the river water.

- Lower than LOQ or not detected

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