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PAPER

Preparation of hollow porous molecularly imprinted polymers and their applications to solid-phase extraction of triazines in soil samples

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Porous polymers have aroused increasing interest due to their controllable hole(s) structure in favor of mass transfer. In this work, three types of porous molecularly imprinted polymers (MIPs) using atrazine as template, namely single-hole hollow molecularly imprinted polymers (s-HMIPs), multihole hollow molecularly imprinted polymers (m-HMIPs) and porous solid molecularly imprinted polymers (ps-MIPs), were prepared, and applied as solid-phase extraction (SPE) sorbent for selective preconcentration and specific recognition of triazines in soil samples. For porous MIPs, most of the binding cavities were located in the proximity of the surface, which remarkably facilitated template removal and mass transfer. The resultant atrazine imprinted porous polymers of s-HMIPs and m-HMIPs exhibited more than triple imprinting capacity for triazines compared to the ps-MIPs, while they were all greatly superior to that of the non-imprinted polymers (NIPs). Accordingly, the s-HMIPs employed as SPE sorbent presented much higher extraction efficiency for several triazines based on the large specific surface area and high adsorption capacity in comparison with the commercial C_{18} column. The validated method was also successfully applied to soil sample analysis, and satisfactory recoveries were attained, such as 73.5–102.0% with the precision of 1.17–4.24% for five triazines at 10 μg L^{-1}. The s-HMIPs-SPE proves to be a highly-effective cleanup and enrichment method for simultaneous separation and sensitive determination of triazines in complicated samples.

1 Introduction

Molecular imprinting is known as a technique for creation of tailor-made binding sites with memory of the shape, size and functional groups for template molecule. Recently, molecularly imprinted polymers (MIPs) have aroused extensive attention and been widely applied in many fields, such as solid phase extraction (SPE)[1–3] and chemical sensors,[4,5] owing to their desired selectivity, physical robustness, thermal stability, as well as low cost and easy preparation. Although MIPs enjoy significant benefits and diverse applications, it's also involved in many problems such as incomplete template removal, low binding capacity and slow mass transfer because of highly cross-linked nature.[5] To solve those problems, more work still needs to be done, although many strategies such as surface molecular imprinting strategy,[3] molecular imprinting nanotechnologies[6] and monomolecular dendritic imprinting strategy[7] have been proposed.

In the meanwhile, the past ten years have witnessed great advances in the development of porous polymers. The main advantages recognized up to now in the use of porous polymers arise from their controllable hole structure which favors mass transfer. Up to date, various polymerization methods including suspension polymerization,[8] seed swelling polymerization,[9] W/O/W emulsion polymerization[10] and stepwise acid/alkali method[11] have been developed to prepare a variety of porous polymeric structures.

By combining molecular imprinting technology (MIT) with porous polymers preparation methods mentioned above, porous MIPs with higher binding capacity and faster mass transfer would be prepared. Single-hole hollow imprinted polymers have been tried toward specific uptake of 2,4,6,-trinitrotoluene (TNT).[12] To the best of our knowledge, there are no reported examples of porous imprinted polymers used as SPE sorbent for selective preconcentration and specific recognition of analytes of template in complicated matrices.

In this work, three types of porous MIPs using atrazine as template, namely single-hole hollow molecularly imprinted polymers (s-HMIPs), multihole hollow molecularly imprinted polymers (m-HMIPs) and porous solid molecularly imprinted polymers (ps-MIPs) were prepared. Their molecular

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recognition specificity and sensitivity were investigated in detail. The performances of the s-HMIPs used as SPE sorbents were investigated for selective preconcentration and specific recognition of triazines in soil samples, which were widely employed herbicides. Under optimal conditions, the s-HMIPs as sorbent can be successfully applied to preconcentration and separation of triazines simultaneously.

2 Experimental

2.1 Reagents

Styrene, divinylbenzene (DVB), and methacrylic acid (MAA) were purchased from Sigma-Aldrich (Shanghai, China) and distilled in vacuum prior to use in order to remove stabilizers. Benzoyl peroxide (BPO) and 2,2′-azo-bis-isobutyronitrile (AIBN) were purchased from Shanghai Chemical Reagents Company (Shanghai, China) and recrystallized in methanol prior to use. Polyvinylpyrrolidone (PVP) and poly(vinylalcohol) (PVA) as stabilizer in dispersion polymerization, dibutyl phthalate (DBP) as swelling reagent and sodium dodecyl sulfate (SDS) were supplied by Tianjin Reagent Plant (Tianjin, China). Atrazine, simetryn, propazine, simazine and ametryn were kindly provided by Binzhou Agricultural Technology Co., Ltd. (Shandong, China). Furazolidone and carbendazim were purchased from J&K Technology Limited (Beijing, China). All other reagents were used as supplied without a further purification step.

2.2 Preparation of polystyrene seed particles

Micrometre-sized, monodispersed polystyrene seed particles were prepared by dispersion polymerization.[13] Briefly, ethanol (50 mL), water (10 mL) and PVP (0.7 g) were added to a 250 mL three-necked, round-bottom flask. Under vigorous stirring, AIBN (40 mg) in styrene (5 mL) were added to the above solution. After pouring with nitrogen for 30 min at room temperature, the mixture was heated to 70 °C and polymerized for 24 h under nitrogen atmosphere. The obtained polystyrene particles were washed by ethanol three times and dried to constant weight under vacuum at 40 °C.

2.3 Synthesis of porous solid polymers

Porous solid polymers were prepared by two-step swelling polymerization.[14] Typically, 0.125 g SDS and 50 mL water were charged into a three-necked, round-bottom flask. Next, 0.2 g polystyrene seed particles were dispersed in the above solutions to form homogenous emulsion. The first step swelling was carried out by adding 0.7 mL DBP to the emulsion for 24 h, followed by the second step swelling by 5–15 mL toluene, 0.344 mL MAA, 4.0 mL DVB and 60 mg BPO. After swelling for another 24 h, the mixture was heated to 80 °C to polymerize under a nitrogen atmosphere for 24 h. Finally, the porous solid polymers were obtained by centrifuging and washing with dichloromethane and methanol. By adjusting the amount of toluene, porous solid polymers and single-hole hollow polymers could be prepared.

2.4 Preparation of hollow porous polymers

Hollow polymer particles with hole(s) in the shell were prepared by self-assembling of phase separated polymer method (SaPSeP method) involving dynamic swelling method (DSM) as reported.[15] Briefly, ethanol (14 g), water (6 g), PVA (10 mg), polystyrene (70 mg), DVB (600 μL), *p*-xylene (600 μL), and BPO (12 mg) were charged into a glass cylindrical beaker. Next, water (60 mL) was added to the mixture slowly under magnetic stirring at room temperature. Then ethanol/water medium of the (DVB/*p*-xylene)-swollen polystyrene particles was centrifuged and then dispersed in ethanol. 40–60 mM SDS was added to the final aqueous medium. Seed polymerization was carried out in a sealed glass flask under a nitrogen atmosphere at 70 °C for 24 h. By adjusting the amount of SDS, hollow polymers with single hole or multiple holes in the shell were prepared.

2.5 Preparation of atrazine imprinted porous MIPs

Typically, the atrazine porous MIPs were prepared by simply adding atrazine into the swelling solutions. During the MIPs preparation process, the molar ratio of template to functional monomer is 1 to 4 constantly. Porous atrazine MIPs were fabricated through removal of atrazine templates with solvent mixtures of methanol : acetic acid (9 : 1, v/v).

2.6 Characterization of the polymers

The prepared polymers were characterized by scanning electron microscopy (SEM), nitrogen adsorption experiments, and high performance liquid chromatography (HPLC). SEM images were recorded using a scanning electron microscope (SEM, JSM 5600 LV, operating at 20 kV) for morphological evaluation. All samples were sputter-coated with gold before SEM analysis. Nitrogen adsorption–desorption results were recorded using AUTOSORB 1 (Quantachrome Instruments, Germany). The Brunauer, Emmett and Teller (BET) method was used to determine the specific surface area. The samples were degassed in a vacuum at 300 °C prior to adsorption measurements.

Estimation of molecular recognition properties was made. The rebinding capacity and kinetics of porous MIPs for atrazine were investigated as follows: 20 mg polymer particles were dispersed in 5 mL flask containing 2.0 mL atrazine ACN solutions of various concentrations. After shaking for 24 h at room temperature, the samples were centrifuged and the supernatant solutions were collected, concentrations of which were determined using HPLC-UV (Skyray Instrument Inc., China). The binding amount of atrazine was determined by subtracting the residual amount of atrazine in solution from the total atrazine amount. Meanwhile, the binding kinetics were tested by monitoring the temporal amount of atrazine in the solutions. Selectivity experiments were carried out by using simetryn, propazine, simazine, ametryn, furazolidone and carbendazim as structural analogs. A C_{18} column with 150 mm × 4.6 mm i.d. (Arcus EP-C_{18}, 5 μm, 4.6 × 150 mm Column, Waters) was used as the analytical column. HPLC conditions employed for the triazines separation were as follows: mobile phase, acetonitrile : water (70 : 30, v/v), and specially, acetonitrile : water (35 : 65, v/v) for the simultaneous separation of the five triazines; flow rate, 1.0 mL min^{-1}; room temperature; UV detection, at 220 nm; injection volume, 20 μL.

HPLC conditions employed for furazolidone were as follows: mobile phase, acetonitrile : water (55 : 45, v/v); flow rate, 1.0 mL min^{-1}; room temperature; UV detection, at 365 nm; injection volume, 20 μL. HPLC conditions employed for carbendazim were as follows: mobile phase, methanol : water (65 : 35, v/v); flow rate, 1.0 mL min^{-1}; room temperature; UV detection, at 282 nm; injection volume, 20 μL.

2.7 MIPs-SPE procedures

A PTFE column was packed with 400 mg of s-HMIPs in 5 mL methanol solution using a wet packing method. Then the cartridge was conditioned with 5 mL of methanol. Atrazine or its structural analogs, dissolved in acetonitrile, was loaded onto the SPE cartridge at a certain flow rate. Subsequently, the cartridge was washed and eluted. The collected column-solution, washing solution and eluate were evaporated to dryness under nitrogen and the residues were redissolved in 0.6 mL acetonitrile for HPLC analysis. In order to get the highest SPE recoveries, the variables, including the flow rate of loading, loading volume and the elute solvent were studied and optimized.

2.8 Analysis of soil samples

Once the optimized MIPs-SPE experimental conditions were established, soil samples were used to demonstrate the applicability of the porous MIPs to preconcentration of triazine herbicides from complicated matrices. The soil sample was prepared as reported.[16] Briefly, dried soil samples were ground and sieved with a mesh, then 50 mL of ACN was used as extraction solvent to extract triazines from 5.0 g dried soil sample for 1 h. Then the extraction solution was concentrated with the mixture standards of atrazine, simazine, ametryn, simetryn, and propazine individual at 10, 60 and 100 μg L^{-1}, respectively. The SPE cartridge was conditioned with 5 mL methanol, and then 10 mL spiked sample was passed through. Subsequently, the column was washed with 3 mL methanol, eluted with 5 mL methanol/acetic acid (9 : 1, v/v), and then the eluted fraction was analysed by HPLC-UV.

3 Results and discussion

3.1 Preparation and characterization of porous MIPs

It is well known that traditional MIPs are confronted with some problems, such as small binding capacity, slow binding kinetics and incomplete removal of templates, because of the highly cross-linked nature of MIPs. Most of the binding cavities are not at the surface of the MIPs, and then target species are resisted from accessing the deep imprinted cavities, thus slowing the kinetics of target analyte binding, as seen in Fig. 1a.[6] Surface imprinting technology was proposed to solve these problems. However, the solid core in surface imprinted polymers was unavailable for template recognition, which lowers the binding capacity per unit mass of MIPs. In order to utilize the recognition sites utmost and improve the binding capacity per unit mass of MIPs, in the present work, hollow MIPs with hole(s) in the shell were proposed. Meanwhile, porous solid MIPs and traditional solid MIPs by precipitation polymerization (TR-MIPs) were prepared for comparing purposes. Fig. 1b describes the

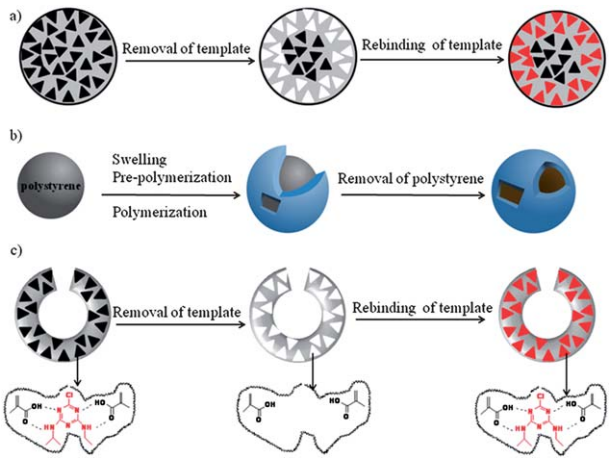

Fig. 1 Schematic illustrations for: a) process of template removal and rebinding in traditional MIPs, b) preparation process of hollow MIPs by swelling polymerization, followed by dissolution of polystyrene cores, and c) recognition process of atrazine in s-HMIPs. (▲ templates in MIPs; △ recognition sites shaped in MIPs after removal of templates; ▲ rebounded templates in recognition sites).

preparation process for hollow porous MIPs by swelling polymerization, followed by dissolving polystyrene seed particles. Fig. 1c is the schematic diagram for the distribution of recognition sites in hollow MIPs with hole(s) in the shell. For those hollow porous MIPs, most of the recognition sites are located in the surface of MIPs, including the interior and exterior surface. So the complete removal of templates can be achieved by the open interior surface. Furthermore, target species can diffuse easily into the recognition sites simultaneously from the interior and exterior of surface. As a result, higher binding capacity and faster mass transfer can be achieved thanks to the open architectures of hollow MIPs.

Initially, porous solid polymers were expected to be prepared by two-step swelling polymerization method and single-hole hollow polymers were expected to be prepared by DSM method. It is so exciting that single-hole hollow polymers could be prepared by both methods and hollow polymers with multiholes in the shell also could be synthesized *via* DSM. So, three kinds of porous polymers, namely porous solid polymers (Fig. 2B), single-hole hollow polymers (Fig. 2C) and multihole hollow polymers (Fig. 2D) were prepared in this work.

The approaches discussed above allowed imprinting molecules toward the specific molecular recognition and high-capacity uptake of target species. In the present work, atrazine, probably one of the most widely used herbicides of triazines, was used as a template, and MAA, which displayed the best effectiveness as functional monomer when used for imprinting atrazine because they can form double hydrogen bonds with atrazine,[17] was used as a functional monomer, to prepare porous MIPs *via* non-covalent interactions. Corresponding to the three kinds of porous polymers, three types of porous MIPs, namely single-hole hollow molecularly imprinted polymers (s-HMIPs), multihole hollow molecularly imprinted polymers (m-HMIPs) and porous solid molecularly imprinted polymers (ps-MIPs) were prepared. The imprinting process, as shown in Fig. 1b and Fig. 1c, was performed by simply adding atrazine molecules into the swelling

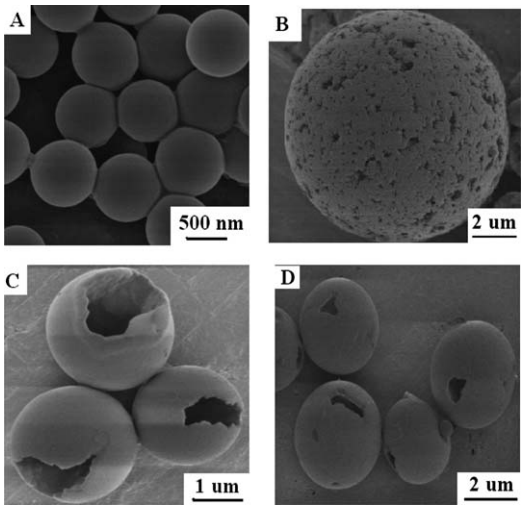

Fig. 2 SEM images of seed polystyrene particles and porous polymers. (A) Seed polystyrene particles prepared by dispersing polymerization; (B) porous solid polymers prepared by multi-step swelling polymerization; (C) single-hole hollow polymers prepared by multi-step swelling polymerization; (D) multi-hole hollow polymers prepared by DSM.

solutions. After the original atrazine templates were removed by solvent extraction, atrazine porous MIPs were obtained. The morphological evaluation for the three kinds of porous MIPs were similar to those porous polymers displayed in Fig. 2, so the SEM photographs for porous MIPs were not given repeatedly.

For the porous MIPs, the pore structure and surface area were studied using the BET adsorption method and the corresponding results are listed in Table 1. As can be seen, MIPs present a much higher specific surface area than those of the corresponding control NIPs; hollow porous MIPs exhibit much larger ones than those of the solid MIPs. The results indicate larger specific surface areas result in higher binding capacity, as can be seen from Fig. 3.

3.2 Estimation of molecular recognition properties of the porous MIPs

Small binding capacity and slow mass transfer were the main problems restricting MIPs for many years, due to the highly cross-linked nature of MIPs.[6] Higher binding capacity and faster binding kinetics were expected thanks to the open architecture of HMIPs in the present work. Static binding experiments and dynamic binding experiments were carried out to estimate the molecular recognition properties of hollow porous MIPs. As shown in Fig. 4, for the three kinds of porous MIPs, binding increased quickly and continuously along with the increase of

Table 1 Specific surface area ($m^2\ g^{-1}$) of the MIPs and corresponding NIPs

Polymers	MIPs	NIPs
Multi-hole hollow porous	390.8	298.3
Single-hole hollow porous	339.8	260.5
Solid porous	180.8	128.9
Solid	130.6	73.6

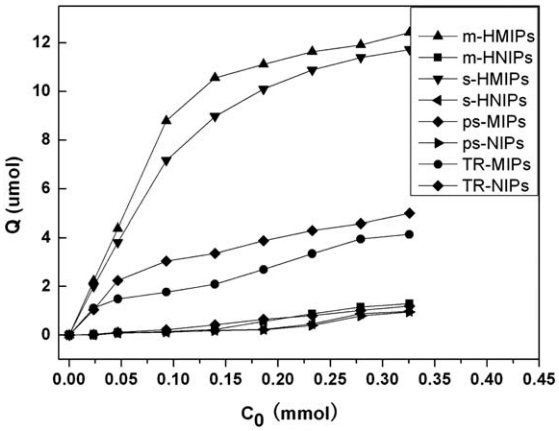

Fig. 3 Binding isotherms of porous MIPs and NIPs for atrazine in acetonitrile. Experimental conditions: $V = 2.0$ mL; mass of polymer, 20 mg; adsorption time, 24 h.

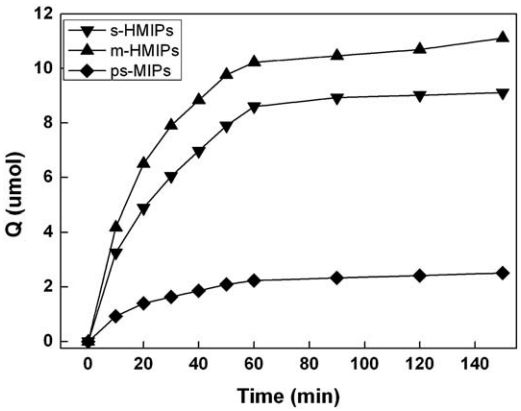

Fig. 4 Kinetic uptake of atrazine molecules onto m-HMIPs, s-HMIPs, and ps-MIPs. Experimental conditions: $V = 2.0$ mL; $C_0 = 40$ mg L^{-1}; mass of polymer, 20 mg.

initial concentration. At the same time, the binding capacity of porous polymers was higher than that of TR-MIPs. Meanwhile, m-HMIPs presented the highest binding capacity while ps-MIPs had the lowest one. The binding capacity of s-HMIPs was appreciably lower than m-HMIPs. This phenomenon could find answers from SEM images and nitrogen adsorption experiments. For the two kinds of HMIPs with the diameter of 3 μm, templates could be removed completely because most of binding sites were located in the proximity of the interior surface. Target species could easily diffuse into the recognition sites of HMIPs through two opposite directions, leading to higher binding capacity and faster mass transfer, as can be seen from Fig. 5. For ps-MIPs with diameter of 10 μm, though containing many nanopores, template removal was still difficult attributing to the big volume. So the binding capacity was much lower than HMIPs. When comparing m-HMIPs with s-HMIPs, those nanopores in m-HMIPs were in favor of template removal and mass transfer. As a result, m-HMIPs had higher binding capacity and faster mass transfer than those of s-HMIPs. The maximum binding capacity was 14.7, 13.9 and 4.6 μmol for 1.0 g of m-HMIPs, s-HMIPs and ps-MIPs, respectively. A time of 60 min was needed to reach adsorption equilibrium for m-HMIPs and

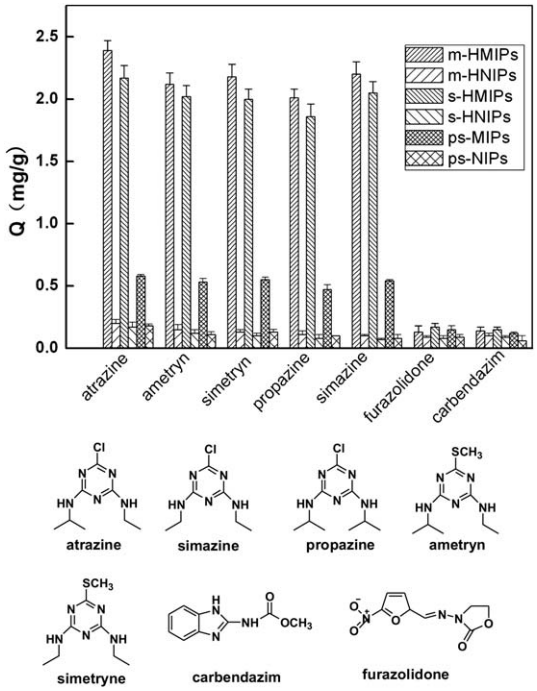

Fig. 5 Binding capacities of porous MIPs and NIPs for five triazines, furazolidone and carbendazim, and their chemical structures. Measurement conditions: polymer, 20 mg; C_0 = 40 mg L^{-1}; V = 2.0 mL; adsorption time, 24 h; room temperature.

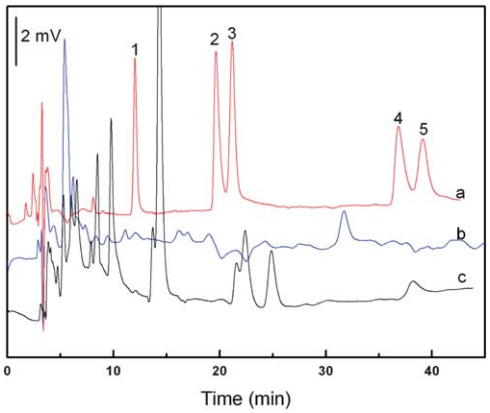

Fig. 6 HPLC-UV chromatograms of 60 μg L^{-1} triazines mixture standard spiked soil samples: (a) extraction with s-HMIPs, (b) extraction with NIPs, and (c) without extraction. (1) Simazine (2) simetryn (3) atrazine (4) ametryn (5) propazine. Experimental conditions: 10 mL spiked solutions; 400 mg s-HMIPs; wash solution, 3 mL methanol; elution solution, 5 mL methanol : acetic acid, 9 : 1, v/v; re-dissolution solution, 600 μL acetonitrile. HPLC condition: mobile phase, acetonitrile : water (35 : 65, v/v); flow rate, 1.0 mL min^{-1}; room temperature; UV detection, at 220 nm; injection volume, 20 μL.

s-HMIPs (Fig. 5) ascribing to the hollow structure. The time is much shorter than that of TR-MIPs used to get the maximum binding capacity. The binding capacity of HMIPs was 4–5 folds higher than the traditional MIPs in the same adsorption time (60 min).

In order to investigate the competitive recognition ability of all the obtained three kinds of MIPs, four triazines (simazine, simetryn, propazine and ametryn) as structural analogues and two references (furazolidone and carbendazim) as other herbicides were used. As seen from Fig. 6, for the three kinds of porous MIPs, the binding capacities for the five triazines were close while they were much higher than two references. Porous MIPs have the similar selectivity when comparing to MIPs prepared in the traditional way because the same functional monomers and same template to functional monomer ratio was used during preparation of those two kinds of MIPs. So, the binding cavity is still the same, which resulted in the same selectivity but only improving the mass transfer and binding capacity. So the obtained atrazine imprinted HMIPs could selectively bind triazines from other herbicides. The obtained HMIPs open a promising way as SPE sorbent to selective preconcentration of triazines.

3.3 Analytical performance for the triazines determination based on s-HMIPs-SPE

According to the above results, the s-HMIPs were employed as SPE sorbent for preconcentration of triazines. In order to get the highest extraction recoveries, various influence factors of SPE were studied and optimized, such as the flow rate of loading atrazine, loading volume and the elution solvent. In the column experiment, 400 mg of s-HMIPs were mixed with methanol and then packed in SPE column. After conditioning with 5 mL methanol, 5 mL of 0.1 mg L^{-1} atrazine solution in acetonitrile was passed through the column. The collected column-solution was dried under nitrogen and the residues were redissolved in 0.6 mL acetonitrile for HPLC analysis. The flow rate of atrazine solution through the packed column was a very important parameter affecting the binding of atrazine. So the flow rate in the range of 0.2–1.5 mL min^{-1} was studied. 100% of atrazine was bounded to s-HMIPs when the flow rate was less than 0.8 mL min^{-1}. Increasing the loading volume from 5 to 10 mL at the flow rate of 0.5 mL min^{-1}, 100% of atrazine still could be bound to s-HMIPs. So 0.5 mL min^{-1} and 10 mL were chosen as the optimal flow rate and loading volume, respectively, in the following work.

It is well known that the elution solvent has great influence on the recovery of atrazine. In order to get the highest recovery, different ratios of the mixed methanol : acetic acid solvents were tested. Finally, 5 mL of a mixture of methanol : acetic acid (90 : 10, v/v) was enough to achieve the complete elution of atrazine and therefore was selected.

Under the optimum condition, s-HMIPs were used for extraction of the five triazines while NIPs and traditional C_{18} cartridge purchased from Shanghai ANPEL Scientific Instrument Co., Ltd. (Shanghai, China) were used for comparison. The extraction recoveries and precisions are shown in Table 2. It was seen that triazines could not be detected at very low concentrations when using NIPs or C_{18} as sorbent. By using the s-HMIPs, the recoveries of atrazine, simazine, ametryn and simetryn ranged from 95.3–109% and 74.9–92.7%, with the relative standard deviation (RSD) of 1.3–4.4% and 1.8–3.5% at 10 and 100 μg L^{-1}, respectively. So, s-HMIPs were selected as SPE sorbent for selective extraction of triazines from real soil samples in the following work.

Table 2 SPE recovery comparisons using different sorbents for triazines acetonitrile solutions under optimal conditions[a]

	s-HMIPs		NIPs		C_{18}	
Triazines	10 μg L^{-1}	100 μg L^{-1}	10 μg L^{-1}	100 μg L^{-1}	100 μg L^{-1}	1 mg L^{-1}
Atrazine	109.3 ± 1.3	92.7 ± 1.9	ND[b]	ND	ND	64.1 ± 9.2
Ametryn	106.1 ± 1.8	80.6 ± 1.8	ND	ND	ND	34.6 ± 22.8
Simetryn	95.3 ± 4.4	74.9 ± 3.5	ND	ND	ND	40.8 ± 19.9
Propazine	45.2 ± 12.5	58.5 ± 2.4	ND	ND	ND	13.5 ± 15.6
Simazine	97.6 ± 3.4	85.6 ± 2.6	ND	ND	ND	35.8 ± 15.8

[a] SPE conditions: condition, 5 mL methanol; loading, 10 mL triazine solution in acetonitrile; washing, 3 mL methanol; eluting, 5 mL methanol/acetic acid (90 : 10, v/v) for s-HMIPs and NIPs, and 5 mL dichloromethane for C_{18} column. [b] Not detected.

Using the s-HMIPs as SPE sorbent, the analytical performances were further investigated, as shown in Table 3. The calibration curves for the detection of those five triazines were obtained by performing a linear regression analysis ranging from 0.05 to 50 mg L^{-1}. Good linearity was obtained with correlation coefficients of $R > 0.99$. The limits of detection (LOD) were 2.8–9.6 μg L^{-1} based on a signal-to-noise ratio of 3. Precision was calculated in terms of intraday repeatability ($n = 6$) and interday reproducibility (6 different days) on 0.1 mg L^{-1}. The intraday repeatability evaluated as RSD was 2.4–5.4%, and the interday reproducibility was 3.5–6.3% (Table 3).

3.4 Applications of s-HMIPs -SPE to soil sample analysis

In order to evaluate the potential applications of s-HMIPs for selective preconcentration of triazines in real samples, 10 mL soil sample solution spiked the mixture standard of atrazine, simazine, ametryn, simetryn and propazine individual at 10, 60, and 100 μg L^{-1}, respectively, were passed through the s-HMIPs filled column. Fig. 6 presents the chromatograms of triazines from soil samples. Five compounds were remarkably concentrated by the s-HMIPs-SPE (Fig. 6a), which was attributed to the fact that s-HMIPs have much higher imprinting efficiency, so the matrix effects could be drastically reduced and the triazines could be preconcentrated. While, for the NIPs-SPE, no obvious peaks of triazines were observed at the corresponding retention time (Fig. 6b), indicating the triazines were specifically adsorbed on the MIPs but negligible interaction on the NIPs. Besides, it was also extremely difficult to detect the triazines without performing the extraction preparation process because of the low concentration level and severe interferences from complicated matrices (Fig. 6c).

The validation of the s-HMIPs-SPE method was performed by examining the recoveries of spiked soil samples. The precision of the method was evaluated by calculating the RSD of the extraction at different concentration levels using the optimized procedures. The results are listed in Table 4(I). Satisfactory recoveries were obtained, such as 73.5–102.0% with precision of 1.17–4.24% at 10 μg L^{-1} except that propazine showed the lowest recovery. This demonstrated the potential applicability of the atrazine s-HMIPs-SPE for simultaneous preconcentration, separation, sensitive determination and accurate quantification of triazines in real samples. Here, the mobile phase of acetonitrile : water (70 : 30, v/v) was not used, due to an inseparable peak of ametryn and propazine and thereby inaccurate determination. Interestingly, the three triazines (simazine, simetryn and atrazine) were completely separated with less than 3.5 min, and satisfactory recoveries were obtained, as shown in Table 4(II).

Much excellent work about preparation of triazines MIPs and applications as SPE sorbent for separation and preconcentration of triazines has been reported, so some, for example,

Table 4 S-HMIPs-SPE recoveries (Rec.) and relative standard deviations (RSD, %) obtained from analysis of soil samples spiked with five kinds of triazines

		10 μg L^{-1}		60 μg L^{-1}		100 μg L^{-1}	
	Triazines	Rec. (%)	RSD	Rec. (%)	RSD	Rec. (%)	RSD
I[a]	Atrazine	102.0	4.24	99.8	1.91	93.5	1.82
	Simazine	94.5	1.17	97.6	3.52	91.8	0.91
	Ametryn	73.5	2.56	75.3	1.79	80.5	3.95
	Simetryn	89.6	1.73	93.4	2.68	93.6	2.54
	Propazine	46.5	5.12	51.2	3.54	54.5	2.15
II[b]	Atrazine	106.0	4.24	99.8	1.91	103	1.82
	Simazine	94.5	1.17	87.6	3.04	94.8	0.91
	Ametryn	98.1	1.62	87.3	1.45	117	3.95

[a] HPLC mobile phase: acetonitrile : water (35 : 65, v/v). [b] HPLC mobile phase: acetonitrile : water (70 : 30, v/v); other conditions are the same as those in Fig. 6.

Table 3 Regression equations, detection limits and relative standard deviations (RSD, %, $n = 6$) for the determination of the five triazines

Sample	Regression equation	R	LOD (μg L^{-1})	RSD (%) Interday	RSD (%) Intraday
Atrazine	$y = 1548 + 11869x$	0.9999	2.8	3.5	2.7
Simazine	$y = 350 + 17001x$	0.9999	3.2	4.2	2.4
Ametryn	$y = -180 + 12697x$	0.9988	6.2	4.9	3.2
Simetryn	$y = 720 + 13072x$	0.9992	4.3	5.7	2.9
Propazine	$y = 227 + 26315x$	0.9897	9.6	6.3	5.4

Table 5 Comparison of recoveries and LODs for triazines determination using MIPs

Template molecule	Polymerization method	Real sample	Rec. (%)	LOD	Ref.
Propazine methacrylate	Precipitation polymerization	Soil, vegetable	89–95	0.4–2.4 ng g^{-1}	18
Propazine	Bulk polymerization	Soil, corn, water	39–103	—	19
Tertbutylazine	Bulk polymerization	Nature water, sediment	—	0.05–0.2 μg L^{-1}	20
Prometryn	Bulk polymerization	Soybean, corn, lettuce, soil	81–106.1	0.012–0.09 μg L^{-1}	21
Atrazine	Precipitation polymerization	Lettuce, apple	79–98	—	22
Atrazine	Multi-step swelling polymerization	River water	95.1–101	25 pg mL^{-1}	23
Simetryn	Precipitation polymerization	Corn, soil	94.6–101	0.14 pg g^{-1}	24
Atrazine	Precipitation polymerization	Soil, millet, soybean, lettuce	71.6–126.7	2.1–8.17 μg L^{-1}	16
Atrazine	Controlled/living precipitation polymerization	Corn, lettuce	81.5–100.9	2.8 μg L^{-1}	1
Atrazine	Multi-step swelling polymerization	Soil	73.5–102	2.8–9.6 μg L^{-1}	This work

semi-covalent imprinting method,[18] surface molecular imprinting method,[16] controlled/living free radical precipitation polymerzaiton,[1] was summarized and compared with the present work, and the results are listed in Table 5. As can be seen from the table, as a new material, the porous MIPs are ideal candidates for SPE sorbent.

4 Conclusions

Three types of porous structured atrazine MIPs, including ps-MIPs, m-HMIPs and s-HMIPs, were successfully prepared by polystyrene seed polymerization in the presence of SDS, and the SPE based on s-HMIPs was greatly developed for simultaneous preconcentration, separation and determination of several triazine compounds in soil samples. The obtained atrazine HMIPs displayed excellent characteristics, such as higher binding capacity and faster mass transfer for triazines owing to the hollow core with hole(s) in the shell, compared with those of ps-MIPs. The remarkable features of HMIPs were demonstrated, namely its high preconcentration and simultaneous separation efforts for the triazines in the complicated samples. Given the high specificity and high enrichment ability of these porous MIPs, we expect that they can be applied to environmental analysis for trace pollutes in complicated matrices, and extended for slow controlled release and rapid medical diagnosis in the future. Extension of this method to preparation of cage-like MIPs with hollow core/porous shell structure and analysis of environmental estrogens are currently in progress.

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Notes and references

1 S. F. Xu, J. H. Li and L. X. Chen, *Talanta*, 2011, **85**, 282–289.
2 C. Long, Z. Mai, Y. Yang, B. Zhu, X. Xu, L. Lu and X. Zou, *J. Chromatogr., A*, 2009, **1216**, 2275–2281.
3 S. F. Xu, J. H. Li and L. X. Chen, *J. Mater. Chem.*, 2011, **21**, 4346–4351.
4 D. Lakshmi, A. Bossi, M. J. Whitcombe, I. Chianella, S. A. Fowler, S. Subrahmanyam, E. V. Piletska and S. A. Piletsky, *Anal. Chem.*, 2009, **81**, 3576–3584.
5 L. X. Chen, S. F. Xu and J. H. Li, *Chem. Soc. Rev.*, 2011, **40**, 2922–2942.
6 G. Guan, B. Liu, Z. Wang and Z. Zhang, *Sensors*, 2008, **8**, 8291–8320.
7 S. C. Zimmerman, M. S. Wendland, N. A. Rakow, I. Zharov and K. S. Suslick, *Nature*, 2002, **418**, 399–403.
8 F. M. B. Coutinho, V. G. Teixeira and C. C. R. Barbosa, *J. Appl. Polym. Sci.*, 1998, **67**, 781–787.
9 J. Ugelstad and I. Nordhuus, *J. Polym. Sci. Part A Polym. Chem.*, 1985, **72**, 225–236.
10 C. J. Mcdonald, K. J. Bouck, A. B. Chapu and C. J. Stevens, *Macromolecules*, 2000, **33**, 1593–1605.
11 M. Okubo and K. Ich, *Colloid Polym. Sci.*, 1994, **272**, 933–940.
12 G. Guan, Z. Zhang, Z. Wang, B. Liu, D. Gao and C. Xie, *Adv. Mater.*, 2007, **19**, 2370–2374.
13 A. J. Paine, W. Luymes and J. McNulty, *Macromolecules*, 1990, **23**, 3104–3109.
14 C. M. Cheng, F. J. Micale, J. W. Vanderhoff and M. S. Ei-Aasser, *J. Polym. Sci., Part A: Polym. Chem.*, 1992, **30**, 235–244.
15 H. Minami, H. Kobayashi and M. Okuto, *Langmuir*, 2005, **21**, 5655–5658.
16 Y. Zhang, R. Liu, Y. Hu and G. Li, *Anal. Chem.*, 2009, **81**, 967–976.
17 J. Matsui, H. Kubo and T. Takeuchi, *Anal. Sci.*, 1998, **14**, 699–702.
18 C. Cacho, E. Turiel, A. Martín-Esteban, D. Ayala and C. Pérez-Conde, *J. Chromatogr., A*, 2006, **1114**, 255–262.
19 E. Turiel, A. Martín-Esteban, P. Fernández, C. Pérez-Conde and C. Cámara, *Anal. Chem.*, 2001, **73**, 5133–5141.
20 I. Ferrer, F. Lanza, A. Tolokan, V. Horvath, B. Sellergren, G. Horvai and D. Barceló, *Anal. Chem.*, 2000, **72**, 3934–3941.
21 X. Hu, Y. Hu and G. Li, *J. Chromatogr., A*, 2007, **1147**, 1–9.
22 B. Mhaka, E. Cukrowska, B. T. S. Bui, O. Ramström, K. Haupt, H. Tutu and L. Chimuka, *J. Chromatogr., A*, 2009, **1216**, 6796–6801.
23 H. Sambe, K. Hoshina and J. Haginaka, *J. Chromatogr., A*, 2007, **1152**, 130–137.
24 S. Wu, Z. Xu, Q. Yuan, Y. Tang, X. Zuo and J. Lai, *J. Chromatogr., A*, 2011, **1218**, 1340–1346.