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Magnetic molecularly imprinted polymers for the fluorescent detection of trace 17β-estradiol in environmental water

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ABSTRACT

A novel core-shell structured composite of Fe_3O_4 nanoparticles and molecularly imprinted polymers (MIPs), namely, magnetic MIPs (M-MIPs), was synthesized by surface imprinting technique combined with precipitation polymerization for the selective and sensitive fluorescent detection of 17β-estradiol (17β-E_2) based on the competitive desorption of fluorescein as a fluorescent indicator from the M-MIPs. The resulting M-MIPs were well characterized, and they showed ideal spherical morphology and magnetic property. Competitive rebinding of 17β-E_2 to the corresponding recognition sites regulated the release of fluorescein and resulted in an enhanced fluorescence signal. The fluorescence increase linearly coincided with the concentration of 17β-E_2 within the range of 0.10–70 μM with a detection limit of 0.03 μM. The recovery of 17β-E_2 from spiked lake and river water samples ranged from 98.2 to 103.8% with relative standard deviations between 1.1 and 3.8%. This study successfully integrated surface imprinting, competitive adsorption, magnetic separation and fluorescent detection, and also demonstrated fast binding kinetics, easy separation and reusability, and sensitive determination of template molecules. Thus, the M-MIP-based method may provide a simple, rapid, convenient, cost-effective and environmentally friendly way for the highly selective and sensitive recognition and detection of non-fluorescent molecules at trace levels.

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1. Introduction

Endocrine disrupting compounds (EDCs) are environmental contaminants with the potential to elicit negative effects in the endocrine systems of humans and wildlife [1]. These chemicals include natural compounds, synthetic estrogens and a wide variety of organic pollutants [2]. Due to their negative effects, EDCs have become the new focus of environmental science fields. For example, 17β-estradiol (17β-E_2), one of the most active phenolic environmental estrogens (PEEs), enters the environment along with the industrial waste from female hormone drug production or through the metabolism of organisms [3]. It can affect the reproductive system or cause immune deficiency even at trace levels [4,5]. Thus, it is important to identify, monitor and remove 17β-E_2 from complicated environmental matrices.

Molecularly imprinted polymers (MIPs) have attracted widespread attention due to their unique identification and specific adsorption [6]. As a special type of adsorptive material, MIPs can effectively recognize and concentrate target molecules as well as reduce matrix interferences. They have been widely applied in purification and separation, chemical/biological sensors, drug delivery and other fields [7–11]. However, in conventional molecular imprinting, there are still many problems such as incomplete template removal, low binding capacity, poor site accessibility and slow mass transfer [12]. The use of surface imprinting technique has been proposed as a way to overcome these limitations by localizing the cavities at the surface or in close proximity to the polymer's surface [13]. Generally, polymerization occurs at the surface of solid substrates such as Fe_3O_4 nanoparticles [14–16], silica microspheres [17,18], polystyrene [19], quantum dots [20,21], TiO_2 particles [22], and Au nanoparticles [23,24].

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These nanomaterials have exhibited a high surface reactivity, large specific surface area and controllable morphology.

Recently, magnetic nanoparticles (mainly Fe_3O_4) have attracted considerable attention due to their biocompatibility and significant magnetic properties. As a novel functional material, magnetic core-shell MIPs possess both adsorptive selectivity and magnetic sensitivity, which allows for rapid separation with an external magnetic field [12]. Combining the surface imprinting technique with magnetic particles can localize the binding sites at the magnetic materials' surface. Gao et al. prepared magnetic MIPs using the surface imprinting technique for the selective separation and determination of 17β-E_2 from milk using high-performance liquid chromatography (HPLC) [25]. Our group prepared magnetic molecularly imprinted microspheres via reversible addition-fragmentation chain transfer precipitation polymerization for the selective recognition and effective removal of 17β-E_2 from water, soil and food samples coupled with HPLC determination [4]. Unfortunately, the analytical procedures are complicated and time-consuming. In view of these studies, we aspired to prepare magnetic core-shell MIPs via simple surface imprinting method and then identify and detect 17β-E_2 by virtue of an extra fluorescent substance, simplifying the analysis process and improving the efficiency.

Therefore, we developed a fluorescent detection method for 17β-E_2 utilizing its competitive adsorption with fluorescein, which acted as the fluorescent tag, onto a new magnetic core-shell MIP material that combines surface imprinting technology and the magnetic separation ability of Fe_3O_4 nanoparticles. Acrylic acid (AA) was introduced to the surface of Fe_3O_4, followed by 17β-E_2 as a template for a two-step temperature-rising precipitation polymerization. The obtained magnetic MIPs (M-MIPs) first adsorbed fluorescein to saturated equilibrium, and then 17β-E_2 was added to displace the fluorescein owing to its competitive template complementary characteristics. Subsequently, the fluorescence intensity of fluorescein indicated the concentration of 17β-E_2. The magnetic separation and binding properties of the M-MIPs for 17β-E_2 were systematically investigated. Furthermore, the M-MIPs were successfully applied to the fluorescent detection of 17β-E_2 in complicated lake and river water samples.

2. Experimental

2.1. Materials and instruments

Ferric chloride hexahydrate ($FeCl_3 \bullet 6H_2O$), acrylic acid (AA), methacrylic acid (MAA), bisphenol A (BPA), and ethylene glycol dimethacrylate (EGDMA) were purchased from Aladdin (Shanghai, China) and used as received. 2,2′-Azobisisobutyronitrile (AIBN) was obtained from Aladdin (Shanghai, China) and recrystallized in methanol prior to use. Estriol and estrone were attained from Dr. Ehrenstorfer (Germany). Hexestrol and 17β-estradiol (17β-E_2) were obtained from J&K Chemical (Shanghai, China). Fluorescein was supplied by Acros Organics USA. Ethylene glycol, anhydrous sodium acetate, polyethylene glycol (PEG) and all other affiliated reagents such as acetonitrile and sodium acetate (NaAc) were supplied by Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China) and used without further purification. The deionized water (18.2 MΩ/cm) that was used throughout the work was obtained from a Milli-Q Ultrapure water system (Millipore, Bedford, MA, USA).

The morphology evaluation was performed by transmission electron microscopy (TEM, JEM-2100F, Japan). Fourier transform infrared (FT-IR) spectra were obtained via a FT-IR spectrometer (FT/IR-4100, Japan). The specific surface areas and pore sizes of the polymers were measured by nitrogen adsorption experiments via a specific surface and pore size analysis instrument (3H-2000PS4, Beijing). A Thermo Scientific spectrophotometer (NanoDrop 2000/2000c, USA) was employed to provide UV–vis spectra. A vibrating sample magnetometer (VSM, LakeShore 7410, USA) was used to measure the magnetic properties of materials. The fluorescence measurements were carried out on a spectrofluorometer (Fluoromax-4, HORIBA)

2.2. Preparation and surface modification of Fe_3O_4 nanoparticles

Magnetic Fe_3O_4 nanoparticles were synthesized through a solvothermal reaction according to the previously reported method [26]. A solution of 2.70 g $FeCl_3 \bullet 6H_2O$, 4.30 g NaAc and 4.00 g PEG was prepared in 80 mL of ethylene glycol. The solution was transferred into a 100 mL Teflon lined stainless steel autoclave, sealed and heated at 200 °C for 10 h. After cooling to room temperature, the obtained Fe_3O_4 nanoparticles were magnetically collected and washed several times with water and ethanol. The products were dispersed in 105 mL of ethanol using ultrasonication, followed by the addition of 420 μL of acrylic acid. The reaction was carried out at room temperature for 3 h with continuous stirring. The resulting Fe_3O_4-AA nanoparticles were magnetically separated and collected, and rinsed three times with both purified water and ethanol. The final products were dispersed in 35 mL of acetonitrile for further use.

2.3. Molecular imprinting on the surface of Fe_3O_4-AA

The MAA was used as the functional monomer and the EGDMA acted as the cross-linking agent for the surface imprinting polymerization. Initially, 5 mL of the Fe_3O_4-AA nanoparticles suspension was dispersed in 80 mL of acetonitrile. Then, 17β-E_2 (0.5 mmol), MAA (2 mmol), EGDMA (10 mmol) and AIBN (20 mg) were added. The solution was degassed in an ultrasonic bath for 5 min and purged with nitrogen. A two-step temperature-rising polymerization was carried out under a nitrogen atmosphere. The polymerization was first performed in a water bath at 50 °C for 6 h, followed by 60 °C for 20 h. The resulting polymers were magnetically separated and sequentially washed with methanol/acetic acid solution (9:1, v/v) and methanol to remove both the template molecules and residual monomers. Finally, the polymers were dried under a vacuum at 30 °C for 24 h until a constant weight was achieved, and marked as M-MIPs. For comparison, magnetic non-imprinted polymers (M-NIPs) were synthesized by the identical process in the absence of the template molecules.

2.4. Fluorescence measurements determining the concentration of 17β-E_2

The fluorescence spectra were measured with an excitation wavelength of 495 nm and slit widths of 4 and 5 nm for excitation and emission, respectively. The experiment was performed in the dark. Initially, 10 mg M-MIPs were dispersed into 1 mL of 25 μM fluorescein in ethanol with ultrasonic assistance. The mixture was shaken for approximately 20 h to ensure that the M-MIPs reached saturated adsorption of the fluorescein. After that, various amounts of standard 17β-E_2 solutions were injected into the above mixture and then shaken for approximately 12 h. Subsequently, the M-MIPs were magnetically separated and the supernatant was analyzed by fluorescence spectrometry. For reference, the same procedure was followed for the measurements of M-NIPs.

2.5. Binding performances of the M-MIPs

The selective binding performance of the M-MIPs was examined using BPA, estriol, estrone and hexestrol as analogs of 17β-E_2.

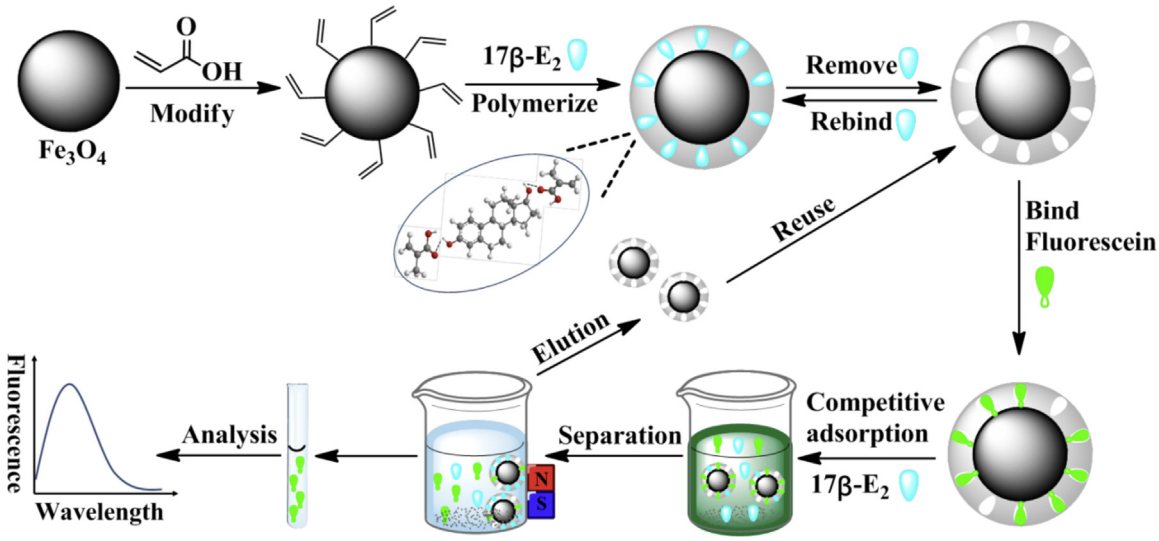

Fig. 1. Schematic illustration of the preparation process of the M-MIPs, the competitive adsorption based fluorescent detection, and the magnetic separation procedure.

The fluorescence measurements were carried out in the dark with 10 mg of the polymers, and the concentration of each of the analogs was fixed at 40 μM. Subsequent fluorescence measurements were conducted according to the method described in section 2.4.

The dynamic binding performance of the M-MIPs was tested using a solution of 10 mg of the polymer microspheres dispersed into 1 mL of 25 μM fluorescein in ethanol that was shaken for approximately 20 h. Then, specific amounts of 17β-E_2 were injected into the solution to ensure it contained 40 μM 17β-E_2. After being shaken for 5, 10, 15, 30, 45, 60, 90, 120, 150, 180 and 210 min, the M-MIPs were separated from the supernatant by an external magnetic field. The fluorescence of the supernatant was measured by fluorescence spectrometry.

2.6. Preparation and analysis of environmental water samples

Environmental water samples were used to demonstrate the practical applicability of the produced M-MIPs for the detection of 17β-E_2 by fluorescence spectrometry. Lake water samples were collected from Binzhou Medical University and river water samples were gathered from the Guangdang River located in the Laishan District of Yantai City. All the samples were filtered with a 0.45 μm filter membrane prior to use to remove suspended particles.

The analytical process was as follows. Initially, 10 mg of the M-MIPs were dispersed into 1 mL of 25 μM fluorescein in ethanol. After the M-MIPs attained saturation of the fluorescein, 20 μL of the water sample was added. The mixture was shaken for 12 h before analysis in the dark. Finally, the polymers were magnetically separated from the collected supernatant that was analyzed by fluorescence spectrometry. To verify the accuracy and practicality of the method, standard addition experiments were completed. The spiking amounts of 17β-E_2 were 2, 10 and 50 μM, respectively.

3. Results and discussion

3.1. Preparation of M-MIPs for the detection of 17β-E_2 by fluorescence

The preparation process of the M-MIPs via a two-step temperature-rising precipitation polymerization is schematically illustrated in Fig. 1. First, the Fe_3O_4 nanoparticles were prepared by a solvothermal reaction followed by the addition of AA to introduce polymerizable functional groups. Then, the imprinting process was conducted at the surface of the Fe_3O_4-AA in the presence of the template (17β-E_2) molecules, functional monomers and cross-linking agents to form uniform polymer shells. Finally, the template molecules were removed by methanol/acetic acid solution to leave the cavities that are complementary to the template 17β-E_2 in shape, size and functional groups.

The obtained M-MIPs were used to recognize and detect 17β-E_2 by means of a fluorescence competitive binding assay. In this process, it was necessary to find a fluorescent indicator/label with similar structure to 17β-E_2. A certain structural similarity between the fluorescent label and template molecule is a prerequisite for this fluorescence competitive binding assay [27,28]. Because there is a non-covalent interaction between the template (17β-E_2) and monomer (MAA), the fluorescent label could be imbedded into the binding sites by hydrogen bonding interactions. Accordingly, fluorescein was used as the fluorescent label in this work since it was similar in structure to 17β-E_2 (Fig. S1). The fluorescein was fully adsorbed and saturated the recognition sites on the M-MIPs so that the addition of 17β-E_2 resulted in the release of fluorescein from the M-MIPs into solution. Because 17β-E_2 is the original template molecule, it entered the specific recognition sites more easily and with a remarkable competitive advantage. Due to the presence and increase of fluorescein released into the sample solutions, the fluorescence intensity gradually increased, which allowed the quantity of adsorbed 17β-E_2 to be determined. Thus, successful fluorescence detection of 17β-E_2 occurred based on the competitive adsorption and selective recognition properties of M-MIPs. In addition, the M-MIPs could be rapidly and easily separated by the external magnetic field for recycled use (Fig. 1).

3.2. Characterization of the M-MIPs

The successful preparation of the M-MIPs was confirmed by FT-IR, as shown in Fig. S2. The Fe-O stretching peak at ~585 cm^{-1} was observed for bare Fe_3O_4 (a), Fe_3O_4-AA (b) and M-MIPs (c), indicating that the composition of Fe_3O_4 was not changed after modification and polymerization. The peaks at ~1745 cm^{-1} in the spectra of Fe_3O_4-AA (b) and M-MIPs (c) are ascribed to the stretching vibrations of C=O, and their presence indicates the success of the functionalization of the Fe_3O_4 particles. As shown in Fig. S2(c), the intensities of the peaks at ~1260 cm^{-1} and ~1425 cm^{-1} were enhanced due to the introduce of the functional monomer (MAA).

The TEM images of Fe_3O_4, M-MIPs and M-NIPs are shown in Fig. S3. As illustrated, the particles exhibited a spherical shape with a mean diameter of approximately 250 nm Fig. S3A shows that the uncoated Fe_3O_4 nanoparticles were of uniform size with rough surfaces. After imprinting and grafting, the nanoparticles maintained their individual spherical character with only slight agglomeration, as seen from Fig. S3B and C. Compared with bare Fe_3O_4, the surface of the M-MIPs became slightly smoother (Fig. S3B) confirming that the imprinted layer was successfully grafted onto the surface of Fe_3O_4.

The magnetic properties of the Fe_3O_4 nanoparticles and the M-MIPs were measured by VSM. As shown in Fig. S4, the shapes of the magnetization curves are similar, and the saturation magnetization of the Fe_3O_4 nanoparticles and the M-MIPs were 76.36 and 66.68 emu/g, respectively. The decrease of the magnetization value for the M-MIPs was attributed to the formation of the MIP layer. Although the existence of the MIP layer reduced the saturation magnetization value, the M-MIPs maintained the ability to rapidly respond to an external magnetic field as shown in the inset of Fig. S4. These results indicate the magnetic characteristic of the synthesized materials permitting separation by magnetic field. Separation in this manner improves the separation efficiency and replaces the centrifugation and filtration steps of previous methods, simplifying the experimental process.

The N_2 adsorption-desorption isotherms and pore size distribution of the M-MIPs and M-NIPs are shown in Fig. S5. As seen in Fig. S5A, the adsorption curves belong to a type of IV isotherm, according to the IUPAC [29,30], confirming the mesoporous structure of these particles. The Brunauer-Emmett-Teller (BET) surface area of M-MIPs under a relative pressure of ($P/P_0 = 0.19737$) was 48.04 m^2/g, higher than the compared M-NIPs (37.46 m^2/g). The pore size distribution obtained by Barrett-Joyner-Halenda (BJH) analysis can be seen in Fig. S5B. It shows prominent peaks at about 2.58 nm, indicating the narrow pore size distribution of the materials. The average pore diameter calculated from the adsorption branch of the isotherms of the M-MIPs was 14.07 nm, slightly smaller than that of the M-NIPs (14.29 nm).

3.3. Analytical sensitivity of the fluorescence detection of 17β-E_2

In this work, we established a new method for rapid identification and detection of trace estradiol based on the competitive adsorption principle of the M-MIPs. Fluorescein, which is structurally similar to 17β-E_2 with a phenolic hydroxyl group, was used as the fluorescent tag. The 17β-E_2, as the template molecule, can replace the fluorescein bound to the M-MIPs by occupying the same specific binding cavities. Subsequently, the fluorescence intensity of the suspension solution increases. According to the experimental results, there was a linear relationship between the increase of the fluorescence intensity and the concentration of 17β-E_2 added to the solution.

The equation could be expressed as

$$\Delta I/I_0 = KC_{17\beta\text{-E2}} \quad (1)$$

where $\Delta I = I - I_0$ is defined as an enhanced amount, I and I_0 are the fluorescence intensity of suspension in the presence and absence of 17β-E_2, $C_{17\beta\text{-E2}}$ is the concentration of 17β-E_2, and K represents the fluorescence enhancement constant.

The concentration of fluorescein was initially varied to determine the ideal concentration for subsequent measurements. The spectra were collected with slit widths of 3 nm for excitation and emission. Too low of a concentration would lead to a decrease in the fluorescence intensity and accuracy. On the contrary, too high of a concentration would result in low measurement sensitivity as well as large reagent consumption. The fluorescence intensity changes with the addition of 17β-E_2 were measured, as shown in Fig. 2, and

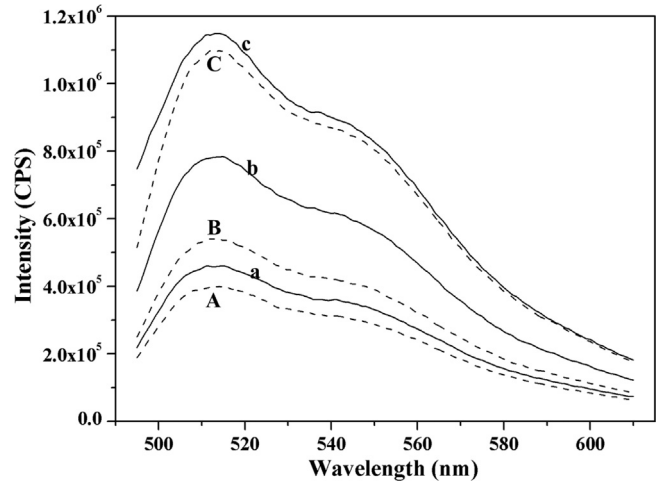

Fig. 2. Effect of fluorescein concentration on competitive efficiency of (A, a) 10 μM, (B, b) 25 μM and (C, c) 50 μM. (A, B, C) indicating fluorescence spectra without 17β-E_2, and (a, b, c) indicating fluorescence spectra in the presence of 17β-E_2. Herein, the concentration of 17β-E_2 was fixed at 100 μM and the mass of M-MIPs was 10 mg; the excitation source was 495 nm and the silt widths were 3 nm for excitation and emission.

as a result the concentration of fluorescein was chosen to be 25 μM where the greatest change in the intensity occurred leading to the largest competitive efficiency.

Under optimal conditions, the imprinting effects of polymers were investigated. As shown in Fig. 3, the fluorescence intensities were enhanced gradually with the addition of 17β-E_2, since the added 17β-E_2 could easily displace the adsorbed fluorescein from the M-MIPs and thereby increase the amount of fluorescein in the solution. By comparison, the enhancement of the fluorescence intensity of the M-MIPs (Fig. 3A) was larger than that of the M-NIPs (Fig. 3B), which indicated that the M-MIPs possessed superior adsorptive affinity for 17β-E_2. An excellent linearity was found in a wide concentration range from 0.10–70 μM with reasonable relative standard deviations (RSDs) between 0.29 and 3.6% (inset of Fig. 3A). Based on $3\sigma/s$, in which σ means the standard deviation of the blank measurements, and s means the sensitivity corresponding to the slope of the calibration graph, the limit of detection (LOD) was determined to be as low as 0.03 μM (8.17 μg/L). This value meets the requirements for trace analysis, so this method can be applied to analyze wastewater samples especially from the feed/pharmaceutical industry, where estrogens are possibly present at μg/L concentrations or higher. Therefore, the M-MIPs based fluorescence detection method can sensitively and accurately quantify the concentration of 17β-E_2, and satisfy the requirements of environmental water investigations.

3.4. Adsorption selectivity and kinetics of the M-MIPs

The recognition selectivity of the M-MIPs was investigated by using four PEEs including BPA, estriol, estrone and hexestrol as analogs of 17β-E_2. It can be seen in Fig. 4 that the fluorescence enhancement amount was the highest for template molecule 17β-E_2, followed by estriol due to the higher structural similarity than that of other three analogs. On the other hand, the fluorescence enhancement amount for 17β-E_2 using the M-NIPs was much lower than that of M-MIPs. These results can be reasonably explained as follows: a large number of tailor-made recognition sites were formed during the synthesis process of M-MIPs and led to specific adsorption towards template 17β-E_2, and there were no specific recognition sites generated on M-NIPs and showed non-specific adsorption. Consequently, the competitive adsorption ability of

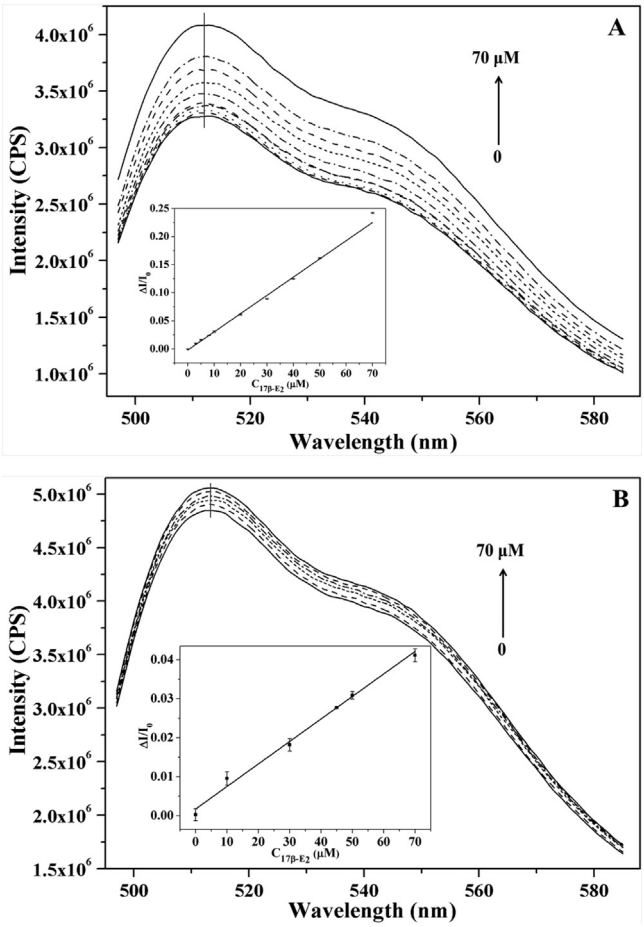

Fig. 3. The fluorescence spectra of fluorescein from competitive desorption upon exposure to different concentrations of 17β-E_2 for M-MIPs (A) and M-NIPs (B). The inset is the linear relationship between the fluorescence enhancement efficiency and the 17β-E_2 concentration. Experimental conditions: fluorescein concentration, 25 μM; mass of polymers, 10 mg; excitation wavelength, 495 nm; silt widths of excitation and emission, 4 and 5 nm, respectively.

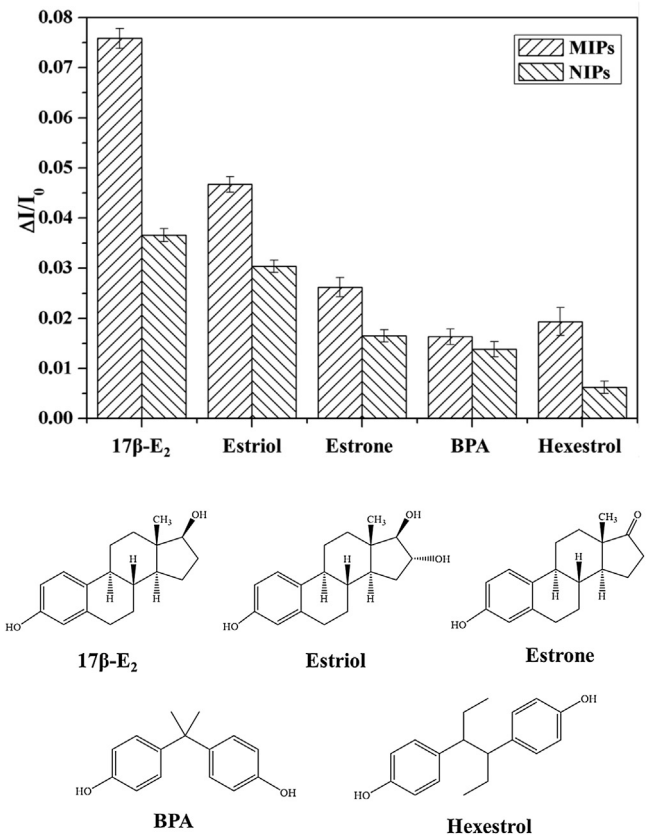

Fig. 4. The adsorption selectivity of M-MIPs and M-NIPs for 17β-E_2 and its structural analogs. Experimental conditions: fluorescein concentration, 25 μM; mass of polymers, 10 mg; concentration of all the targets, 40 μM; excitation wavelength, 495 nm; silt widths of excitation and emission, 4 and 5 nm, respectively.

17β-E_2 was significantly larger than its analogs and resulted in a considerably higher fluorescence increase due to a greater amount fluorescein being released into solution. Thus, the obtained M-MIPs are able to selectively bind 17β-E_2 compared to other PEEs, resulting in a larger quantity of released fluorescein. The obtained M-MIPs open a promising way as a competitive adsorption media for selective fluorescence detection of 17β-E_2.

The kinetic binding of the M-MIPs was also examined. As seen from Fig. S6, the relative fluorescence enhancement amount for the M-MIPs increased sharply just before the dynamic adsorption equilibrium was reached. The M-MIPs obtained about three quarters of the equilibrium adsorption capacity during 60 min and nearly reached saturation within 150 min. The M-NIPs needed a longer time to reach equilibrium. Hence, the M-MIPs had a faster binding rate with 17β-E_2 than the M-NIPs resulting in a faster displacement of fluorescein from the M-MIPs into solution, and thereby confirming the rapid pace of the fluorescence detection of 17β-E_2.

3.5. Reusability of the M-MIPs

As it is well known, reusability is an important characteristic of materials to improve economic efficiency and extend applications. To investigate the reusability, the prepared M-MIPs (10 mg) were first dispersed into fluorescein solutions (25 μM, ethanol solvent) for approximately 20 h to make sure the M-MIPs adsorbed the fluorescein to the point of saturation. Afterwards, 17β-E_2 was added into the above solution to a concentration of 50 μM. The solution was shaken for approximately 90 min and, due to the competitive adsorption of 17β-E_2, the fluorescein imbedded onto the M-MIPs was released, and then the fluorescence intensity of solution was measured. The M-MIPs were rapidly separated by an external magnetic field and after eluting with methanol/acetic acid solution (9:1, v/v) where ready for reuse. The adsorption-desorption procedure was repeated using the same sample of M-MIPs for six cycles. The measurements resulting from the six cycles had RSDs of less than 5.8%, indicating that the M-MIPs are excellently reusable.

Table 1
Spiked recoveries and relative standard deviations (RSD, %, $n = 3$) for the detection of 17β-E_2 in water samples.

Sample	Spiked (μM)	Detected[a] (μM)	Recovery[a] (%)	RSD (%)
Lake water	2	2.076	103.8	3.4
	10	10.179	101.8	2.8
	50	49.750	99.5	1.1
River water	2	1.963	98.2	3.2
	10	10.233	102.3	2.1
	50	49.268	98.5	3.8

[a] The average value of three measurements is reported.

3.6. Practical application and performance comparison

To evaluate the feasibility and practicality of the M-MIPs, they were applied to competitive adsorption of 17β-E_2 from lake water and river water samples. As listed in Table 1, high recoveries of

Table 2
Comparison of this method's analytical performance with other reported MIPs based methods for the determination of 17β-E_2.

MIPs	Detection technology	Linear range	LOD	Sample	Ref.
Fe_3O_4@MIPs	Fluorometry	0.10–70 μM	0.03 μM[a]	Lake water, river water	This work
Fe_3O_4@SiO_2-Dye-MIPs	Fluorometry	0–20 μM	0.19 μM	–	[3]
Fe_3O_4@MIPs	HPLC	0.1–50 ng/mL	0.01 ng/mL	Milk	[25]
H-MIPs[b]	HPLC	–	4.6 μg/L	Milk	[31]
Fe_3O_4@MIPs	HPLC	1.0–100 ng/mL	0.04 ng/mL	Lake water, river water, effluent	[32]

[a] 8.17 μg/L.
[b] Hollow molecularly imprinted polymers.

98.2–103.8% with RSDs between 1.1 and 3.8% were measured for the two environmental water samples, spiked with 17β-E_2 at three different concentrations. Therefore, the M-MIPs coupled to fluorometry method has been validated and is an ideal alternative to previous methods for the simultaneous separation and determination of 17β-E_2 in real water samples for pollution monitoring and abatement.

In addition, the analytical performance of our developed M-MIPs coupled to fluorometry method was compared with other reported MIPs based methods for the determination of 17β-E_2. As listed in Table 2, the LOD of our method is much lower than that of the fluorescein-coated magnetic nanoparticles combined with fluorometry method [3], as well as the reported method requiring a fluorescent label and several modification steps [3]. Meanwhile, our method presented a comparable LOD to that of the hollow MIPs coupled with HPLC method [31], or a slightly higher LOD than that of the M-MIPs combined with HPLC analysis [25,32]. While it has a comparable LOD, the HPLC determination is time consuming, high cost and solvent consuming. Overall, our developed method is simple, convenient, time and cost saving, reliable, reusable, and eco-friendly. In addition, it was successfully tested for application to the selective and sensitive determination of trace 17β-E_2 in environmental water samples.

4. Conclusions

In conclusion, a new method coupling M-MIPs competitive adsorption with fluorescent quantitation was developed by reasonably combining surface imprinting, magnetic separation and fluorescent detection, for the rapid, selective and sensitive detection of 17β-E_2 in environmental water samples. Based on the strong competitive adsorption ability of 17β-E_2 over fluorescein onto the M-MIPs, fluorescein was utilized as a fluorescent indicator for the fluorescent detection with rapid identification and accurate quantification of 17β-E_2. Owing to the simplicity, rapidity, reliability and recycled use, the presented method provides a promising route for the removal and analysis of PEEs from complicated samples. Furthermore, the M-MIPs based fluorescent detection strategy can be extended to analyze more non-fluorescent targets without the use of derivatization or inducers.

Acknowledgments

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.snb.2016.09.111.

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