Nanotechnology **20** (2009) 165601 (7pp) doi:10.1088/0957-4484/20/16/165601

# Magnetic molecularly imprinted polymer for aspirin recognition and controlled release

**Xianwen Kan, Zhirong Geng, Yao Zhao, Zhilin Wang[1] and Jun-Jie Zhu[1]**

State Key Laboratory of Coordination Chemistry, MOE Key Lab of Analytical Chemistry for Life Science, School of Chemistry and Chemical Engineering, Nanjing University, 22 Hankou Road, Nanjing 210093, People's Republic of China

E-mail: wangzl@nju.edu.cn and jjzhu@nju.edu.cn

Received 18 October 2008, in final form 13 February 2009
Published 1 April 2009
Online at stacks.iop.org/Nano/20/165601

**Abstract**

Core–shell structural magnetic molecularly imprinted polymers (magnetic MIPs) with combined properties of molecular recognition and controlled release were prepared and characterized. Magnetic MIPs were synthesized by the co-polymerization of methacrylic acid (MAA) and trimethylolpropane trimethacrylate (TRIM) around aspirin (ASP) at the surface of double-bond-functionalized $Fe_3O_4$ nanoparticles in chloroform. The obtained spherical magnetic MIPs with diameters of about 500 nm had obvious superparamagnetism and could be separated quickly by an external magnetic field. Binding experiments were carried out to evaluate the properties of magnetic MIPs and magnetic non-molecularly imprinted polymers (magnetic NIPs). The results demonstrated that the magnetic MIPs had high adsorption capacity and selectivity to ASP. Moreover, release profiles and release rate of ASP from the ASP-loaded magnetic MIPs indicated that the magnetic MIPs also had potential applications in drug controlled release.

## 1. Introduction

Molecular imprinting is now a well-established method as one of the most promising technologies for the preparation of intelligent polymer materials with specific recognition capacities for template molecules [1–6]. In the polymerization process, the specific recognition capacity of the polymer was achieved by forming a complex between the template molecule and functional monomers in the molecularly imprinted polymers (MIPs). Removal of the template molecule from the obtained polymer by simple solvent extraction reveals binding sites that are complementary in size, shape and functional group to the template molecule. Therefore, the obtained MIPs have special binding capacity, preconceived selectivity to template molecules, and also have other advantages, for instance, easy preparation, chemical stability and low cost. Owing to these features, MIPs could be used not only partially as a substitute for natural biomolecules but also as substrate-selective or separation materials under harsh conditions, which enable them to be applied in wider fields, such as biosensors, antibody and enzyme mimics, chiral separation and solid phase extraction [7–11]. In recent years, a new potential application of MIPs in drug controlled release has attracted considerable attention [12, 13]. Several works have been reported to prepare MIPs with different morphologies as delivery carriers to control release of drugs, such as monoliths, granules, membranes and microspheres [14–17]. For most of the applications, especially controlled release of drugs, the best shape of MIPs may be the spherical shape due to their excellent isotropic release capacity [18]. Among the different morphology MIPs used in the release processes, nanospherical MIPs possess better performance because the small size and high specific surface area would improve the adsorption capacity and rate [19, 20]. However, the nanosized MIPs could not be separated easily by normal separation methods like filtration, except for high-speed centrifugation. If the nanospherical MIPs were endowed with magnetic properties, they could be separated by an external magnetic field with relative rapidity and easy, cost effectively and highly efficiently

[1] Authors to whom any correspondence should be addressed.

manipulated. Furthermore, it is more important that magnetic materials could be localized to the pre-delivery sites and release of the drug to particular sites by the external magnetic field [21]. Hence, nanospherical magnetic MIPs should be suitable as an ideal polymer material for wider applications. Nonetheless, few studies have been reported on the preparation of nanospherical magnetic MIPs [22] and no study has been reported about the controlled release of drugs by nanospherical magnetic MIPs.

Aspirin (ASP), as an analgesic and antipyretic agent, is widely employed in pharmaceutical formulations for the relief of headaches, fever, muscular pains and inflammation due to arthritis or injury. However, widespread use of ASP had resulted in the problem of therapeutic intoxication from overdose, which may be found in persons with chronic inflammatory diseases taking ASP habitually [23]. This problem could be overcome by controlled release of the drug because efficiently controlled release should provide a desired rate of delivery with the therapeutic dose, at the most appropriate place in the body, to prolong the duration of pharmacological action and reduce the adverse effects, minimize the dosing frequency and enhancing patient compliance [24].

The present work proposes a simple and efficient method for the preparation of magnetic MIPs to imprint ASP. ASP, MAA and TRIM as a template molecule, a functional monomer and a cross-linker, respectively, were copolymerized in the presence of the double-bond-functionalized $Fe_3O_4$ nanoparticles to form nanospherical magnetic MIPs. The special adsorption and selective recognition capacity of magnetic MIPs were evaluated. *In vitro* controlled release of ASP from magnetic MIPs was also examined in aqueous solution.

## 2. Experimental details

### *2.1. Materials*

Trimethylolpropane trimethacrylate (TRIM) was purchased from Sigma. $\gamma$-methacryloxypropyltrimethoxysilane ($\gamma$-MPS) was purchased from Jintan Eastchina Coupling Agent Factory, China. Methacrylic acid (MAA) was distilled under reduced pressure to remove the inhibitor. 2′-azobisisobutyronitrile (AIBN) was recrystallized by ethanol. Acetonitrile used was HPLC grade. Water was purified with a Milli-Q system, whose conductivity was at a constant 18.2 MΩ. All other reagents used were of AR grade and without further purification.

### *2.2. Preparation of double-bond-functionalized $Fe_3O_4$*

$Fe_3O_4$ magnetic nanoparticles were prepared by the co-precipitation method according to the following procedure [25]. 180 ml of an aqueous solution containing 11.2 mmol $Fe^{3+}$ and 5.6 mmol $Fe^{2+}$ was heated to 50 °C. Then 12.5 ml of ammonia was added under vigorous stirring. After 30 min, the reaction was heated and kept at 90 °C for 30 min again. $N_2$ was used as the protective gas in the whole experiment. After completion of the reaction, the black precipitate was collected by an external magnetic field, washed with water and ethanol, and dried in vacuum.

To modify the magnetic nanoparticles with a double bond, 4 ml $\gamma$-MPS was dropwise added into the mixture solvents of ethanol and water (1:1, v/v) containing dispersed $Fe_3O_4$ nanoparticles and the reaction was kept for 12 h at 40 °C under $N_2$ gas. Then the product was separated and washed by ethanol for several times, and dried in vacuum.

### *2.3. Preparation of magnetic MIPs*

The magnetic MIPs were synthesized in a 100 ml flask equipped with a mechanical stirrer and a $N_2$ inlet cooling with cycled water. The mixture of 1 mmol ASP and 4 mmol MAA in chloroform which have been shaken for 10 h to form a template–monomer complex was added to 40 ml chloroform with 0.2 g dispersed double-bond-functionalized $Fe_3O_4$ magnetic nanoparticles, followed by adding 2 mmol TRIM and 50 mg AIBN. The temperature was increased to 70 °C and the reaction was allowed to proceed for 6 h. The resulting product was collected by the external magnetic field and eluted by a mixture solvent of methanol and acetic acid (9:1, v/v) for several times to extract the template molecules until the eluent was free from ASP as detected by UV–vis spectrometry (at 277 nm). The obtained polymers were finally rinsed with ethanol to remove the remaining acetic acid and dried in the vacuum desiccator for 24 h before use. For a comparison, magnetic non-molecularly imprinted polymers (magnetic NIPs) were prepared in the absence of ASP during the polymerization process and treated in the identical manner.

### *2.4. Adsorption and selectivity experiments*

20 mg of magnetic MIPs or magnetic NIPs was incubated with ASP, salicylic acid (SA) or o-amino benzoic acid (ABA) in chloroform. After incubation on a rocking table for 10 h, the polymers were separated from the suspension by the external magnetic field. The concentration of free substrate (ASP or SA) in the supernatant was measured by HPLC and the adsorption quantity was then calculated. The concentration of ABA in the supernatant was measured by UV–vis spectrometry (at 249 nm).

### *2.5. In vitro controlled release of ASP*

100 mg of magnetic MIPs or NIPs were shaken in 10 ml of 5 mM ASP in chloroform for 10 h at room temperature, and then were separated by an external magnetic field and dried under vacuum. Magnetic MIPs or magnetic NIPs loading with ASP were suspended in 40 ml of pH 6.8, 0.1 mol $l^{-1}$ phosphate buffer solution. The entire system was kept at 37 °C with gently shaking. Periodically, 0.4 ml of the release medium was withdrawn and the supernatant after magnetic separation was stored at 4 °C for HPLC analysis. The total mass of released ASP at each moment of the experiment was calculated, taking into account the aliquots taken.

### *2.6. Characterization*

*Transmission electron microscopy (TEM).* Sample size was characterized by a JEM-200CX instrument (JEOL, Japan) operating at an acceleration voltage of 100 kV. The samples for TEM measurements were prepared by placing one drop of the sample on copper grids coated with carbon.

*X-ray powder diffraction (XRD).* The structure of the powder samples was characterized by XRD (Philips, Holland). The x-ray diffraction patterns were taken from 10° to 80° ($2\theta$ value) using Cu K$\alpha$ radiation with an intensity ratio ($\alpha_1/\alpha_2$) = 0.5 and wavelengths of 1.544 39 and 1.540 56 Å, respectively.

*Fourier transform infrared spectroscopy (FTIR).* All FTIR spectroscopic measurements were performed on a Bruker IFS 66/S (Germany). The sample powders were dried and mixed with KBr and pressed into a plate for measurement.

*Vibrating sample magnetometer (VSM).* Magnetic measurements of $Fe_3O_4$ nanoparticles, double-bond-functionalized $Fe_3O_4$ nanoparticles and magnetic MIPs were carried out using a vibrating sample magnetometer (VSM, 7300, Lakeshore, USA) at room temperature under a magnetic field up to 10 kOe.

*Thermal gravimetric analysis (TGA).* TGA was conducted on a Shimadzu TGA-50 instrument (Japan) from room temperature to 600 °C with a heating rate of 10 °C min$^{-1}$ in a nitrogen flow (100 ml min$^{-1}$).

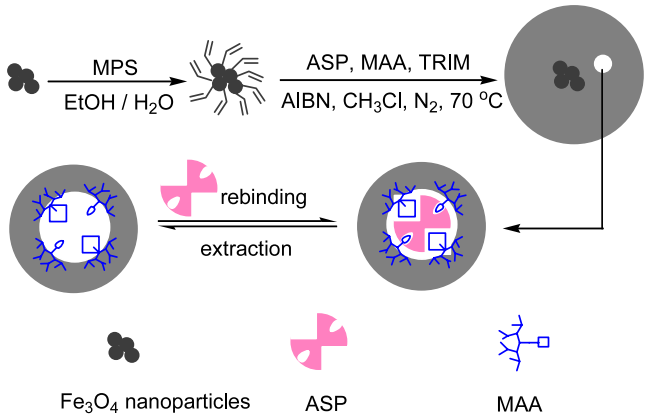
*High performance liquid chromatography (HPLC).* The concentration of ASP or salicylic acid (SA) was assayed on a Shimadzu LC-10AD (Shimadzu, Japan) HPLC system equipped with a Shimadzu RF-530 UV–vis detector. The stationary phase consisted of a column (4.6 mm × 250 mm, Agilent, USA) packed with EclipsePlus $C_{18}$, 5 $\mu$m particle size. Wavelength was set at 234 nm. The mobile phase consisted of water–phosphate buffer (pH 2.5)–acetonitrile (35:40:25, v/v). The column was eluted at a flow rate of 1.0 ml min$^{-1}$ at 37 °C.

*Ultraviolet–visible spectrum (UV–vis).* The concentrations of ASP and ABA in the supernatant were detected on a UV-2401PC spectrometer (lambda 35, Perkin Elmer Instrument, USA).

## 3. Results and discussion

### *3.1. Characterization of magnetic MIPs*

The double bond could guide the formation of MIPs at the solid support surface by radical polymerization [26]. Ding *et al* [27] reported the modification of the double bond at the $Fe_3O_4$ nanoparticle surface by replacing oleic acid with $\gamma$-MPS. Microemulsion and suspension polymerization, and nanoporous alumina template methodology have been reported to be used in the preparation of magnetic MIPs [22, 27–29]. Compared with these reported methods, the modification procedure in the present study was simplified by the directed reaction between $\gamma$-MPS and hydroxyl groups at the $Fe_3O_4$ nanoparticle surface. And, as an ideal method for preparing the spherical MIPs, precipitation polymerization was employed to synthesize the spherical magnetic MIPs without any emulsifier, suspending reagent and template being used, which would simplify the subsequent disposal process. The synthetic route




**Scheme 1.** Schematic illustration of the synthesis of magnetic MIPs. (This figure is in colour only in the electronic version)

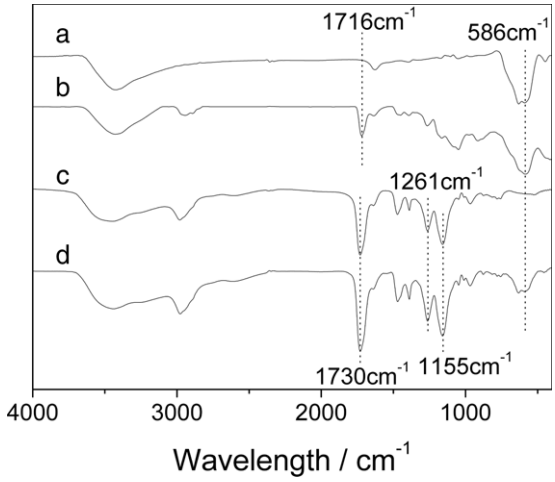



**Figure 1.** FTIR spectra of the $Fe_3O_4$ (a), double-bond-functionalized $Fe_3O_4$ (b), pure MIPs (c) and magnetic MIPs (d).

of introducing a vinyl group and further grafting MIPs onto the $Fe_3O_4$ nanoparticle surface was illustrated in scheme 1.

Figure 1 gives the FTIR spectra of $Fe_3O_4$ nanoparticles (a), the double-bond-functionalized $Fe_3O_4$ nanoparticles (b), the pure MIPs (c) and the magnetic MIPs (d). Besides the obvious Fe–O bond situated at 586 cm$^{-1}$ in both curves of a and b, a new considerably stronger band of the carbonylic group of the $\gamma$-MPS at 1716 cm$^{-1}$ in curve b indicated that the terminated double bond had been grafted onto the $Fe_3O_4$ surface. The existence of the MIPs coating to the $Fe_3O_4$ nanoparticles (curve d) was confirmed by the appearance of characteristic bands of pure MIPs (curve c). The main absorption bands of the pure MIPs situated around 1730, 1261 and 1155 cm$^{-1}$ are assigned to the following vibrations: C=O stretching vibration of carboxylic, C–O stretching vibration of symmetric and asymmetric ester, respectively [30].

The x-ray powder diffraction spectra of the bare, double-bond-functionalized and polymer-coated $Fe_3O_4$ nanoparticles are shown in figure 2. Five characteristic peaks for $Fe_3O_4$ marked by their indices (220), (311), (400), (422), (511) and (440) were observed for all the samples to reveal pure

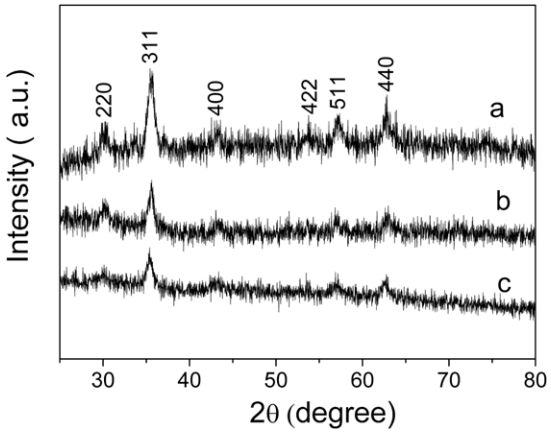



**Figure 2.** X-ray diffraction patterns of $Fe_3O_4$ (a), double-bond-functionalized $Fe_3O_4$ (b) and magnetic MIPs (c).

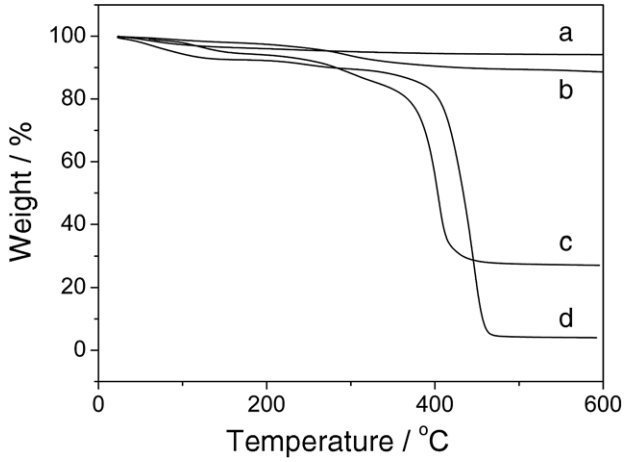



**Figure 4.** TGA curves of $Fe_3O_4$ (a), double-bond-functionalized $Fe_3O_4$ (b), magnetic MIPs (c) and pure MIPs (d).

$Fe_3O_4$ in the resultant particles [31]. The peak positions of the $Fe_3O_4$ nanoparticles are all consistent with the lower diffraction intensity before and after the polymerization, and it is found that the peaks of magnetic MIPs are obviously broadened due to lower $Fe_3O_4$ content in magnetic MIPs. This result suggested that the $Fe_3O_4$ nanoparticles were indeed incorporated into MIPs and the structure of $Fe_3O_4$ nanoparticles was not changed during the polymerization process [32].

The morphological structure of $Fe_3O_4$ nanoparticles and magnetic MIPs were detected by TEM as shown in figure 3. The effective average diameter of the $Fe_3O_4$ nanoparticles is about 12 nm, which is consistent with the XRD result calculated by Debye–Scherrer's formula. The magnetic MIPs are obvious a regular spherical shape with the black $Fe_3O_4$ nanoparticles in the outer gray MIP shell. The average diameter of the magnetic MIPs is about 500 nm. The amount of MIP coating to $Fe_3O_4$ nanoparticles can be calculated from corresponding TGA curves, as shown in figure 4. The double-bond-functionalized $Fe_3O_4$ nanoparticles displayed weight loss below 600 °C, which resulted from the loss of silica coupling agent grafted onto the $Fe_3O_4$ nanoparticles. Compared with the weight loss between pure MIPs and magnetic MIPs, the MIP grafted amount was calculated to be about 72.5% of the total weight of the synthesized magnetic MIPs.

The magnetic properties of the $Fe_3O_4$ nanoparticles, double-bond-functionalized $Fe_3O_4$ nanoparticles and magnetic MIPs were characterized by VSM at room temperature, and the results are shown in figure 5. The saturation magnetization value of the magnetic MIPs was about 15.8 emu $g^{-1}$, which is smaller than that of double-bond-functionalized $Fe_3O_4$ nanoparticles (61.3 emu $g^{-1}$). The decrease of the saturation value is most likely attributed to the existence of MIPs on the surface of $Fe_3O_4$ nanoparticles. The typical characteristics of superparamagnetic behavior were observed showing immeasurable remanence and little coercivity [33]. This superparamagnetism enabled magnetic MIPs to be separated from the suspension easily under the external magnetic field and redispersed rapidly without the external magnetic field, which should facilitate the subsequent adsorption and release experiments.

### *3.2. Adsorption properties of magnetic MIPs*

The adsorption capacity is an important factor to evaluate the special binding and selective recognition of MIPs. The adsorption experiments were conducted by varying the initial concentration of ASP from 0 to 5 mM in 2 ml chloroform in the presence of 20 mg magnetic MIPs or magnetic NIPs. The equilibrium adsorption capacity of magnetic MIPs and magnetic NIPs to ASP were determined and defined as $Q$

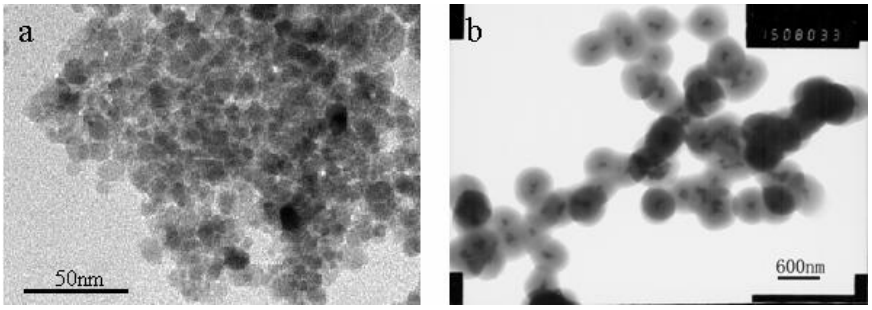



**Figure 3.** TEM images of $Fe_3O_4$ nanoparticles (a) and magnetic MIPs (b).

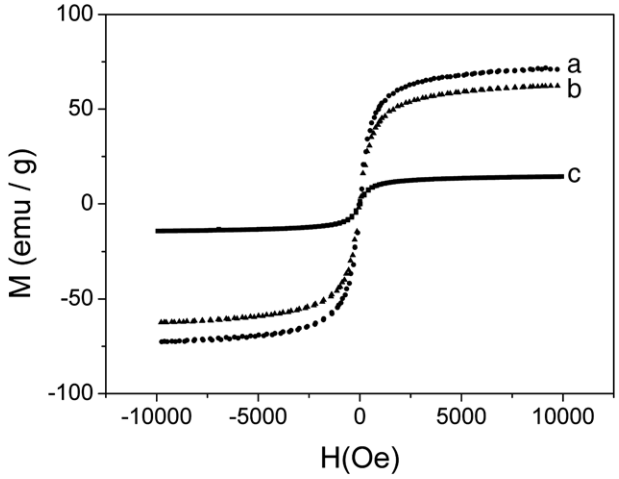



**Figure 5.** Magnetization curves obtained by VSM at room temperature of $Fe_3O_4$ (a), double-bond-functionalized $Fe_3O_4$ (b) and magnetic MIPs (c).

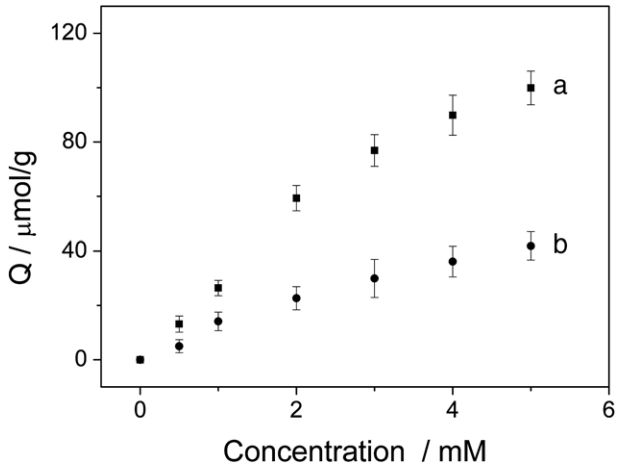



**Figure 6.** Adsorption isotherms of ASP on magnetic MIPs (a) and on magnetic NIPs (b). Amount of polymers: 20 mg, volume: 2.0 ml.

($\mu$mol $g^{-1}$), which was calculated as follows:

$$Q = (C_0 - C_s) \times V/m$$

where $C_0$ (mmol $l^{-1}$) is the initial concentration of substrate solution; $C_s$ (mmol $l^{-1}$) is the substrate concentration of the supernatant; $V$ (ml) is the volume of the initial solution; $m$ (g) is the mass of the polymers.

As shown in figure 6, the amounts of ASP absorbed on polymers increased with the increase of the initial concentration of ASP. The adsorption isotherm showed a good fit to a Langmuir model. It is obvious that the magnetic MIPs have a higher adsorption capacity than magnetic NIPs due to the specific binding cavities in the magnetic MIPs for ASP, although these two types of polymers are composed of exactly the same monomer composition.

The selectivity test of magnetic MIPs was carried out under equilibrium binding conditions using ASP, SA and ABA as substrates. The ratio of adsorption amounts obtained between the magnetic MIPs and magnetic NIPs was also compared. Here, the value of the ratio $\alpha_{(\text{imprint factor})}$ for each substrate is defined in the following equation [34]:

$$\alpha_{(\text{imprint factor})} = Q_{(\text{imprint})}/Q_{(\text{unimprint})}.$$

**Table 1.** Selectivity of magnetic MIPs and magnetic NIPs.

| Substrate | $Q$ for magnetic MIPs ($\mu$mol $g^{-1}$) | $Q$ for magnetic NIPs ($\mu$mol $g^{-1}$) | $\alpha_{(\text{imprint factor})}$ |
|---|---|---|---|
| ASP | 99.9 | 41.8 | 2.39 |
| SA | 42.7 | 28.8 | 1.48 |
| ABA | 38.9 | 22.4 | 1.74 |

Table 1 showed the adsorption amounts of three substrates on the magnetic MIPs and NIPs. The imprint factor of ASP, SA and ABA was 2.39, 1.48 and 1.74, respectively. The result indicated that the magnetic MIPs exhibited higher selectivity for ASP. This is because the imprinting process created binding sites with shape and functional group complementary to the template molecule, resulting in the magnetic MIPs having high selectivity for the template molecule, while both SA and ABA had no complementary shape and functional group matched with the imprinting cavities, resulting in the low binding capacity.

### *3.3. In vitro controlled release of ASP*

In the general preparation process of MIPs, the functional monomers initially form a complex with the template molecule, and then are polymerized in the presence of excessive cross-linkers, so that their functional groups are held in position by the highly cross-linked polymeric structure to enable the rebinding of the template molecule with a very high specificity. In the present study, the properties of magnetic MIPs were focused not only on the adsorption and selectivity capacity, but also on the controlled release capacity of the template molecule. A too highly cross-linked polymeric structure results in too strong a binding capacity to the template molecule, which leads to difficulty in controlling the release of the template molecule. Therefore, using a cross-linker at a low molar ratio with respect to the functional monomer, the obtained polymer possessed sufficient rigidity to keep the specific sites and excellent recognition properties, and at the same time, endowed the MIPs with an obvious release capacity [35]. In the present study, the molar ratio of TRIM and MAA was chosen to be 1:2.

The release kinetics of ASP from the magnetic MIPs and magnetic NIPs was subsequently conducted. 100 mg polymers were incubated in 10 ml of 5 mM of ASP in chloroform. The amounts of ASP adsorbed onto magnetic MIPs and magnetic NIPs were about 10 $\mu$mol and 4.2 $\mu$mol, respectively. After separation by external magnetic field and drying under vacuum, the ASP-loaded polymers were added to pH 6.8 PBS and the release kinetics of ASP from the two polymers was subsequently examined by HPLC. Since ASP may slowly hydrolyze in aqueous solution, the concentration of released ASP was calculated from the remaining ASP and

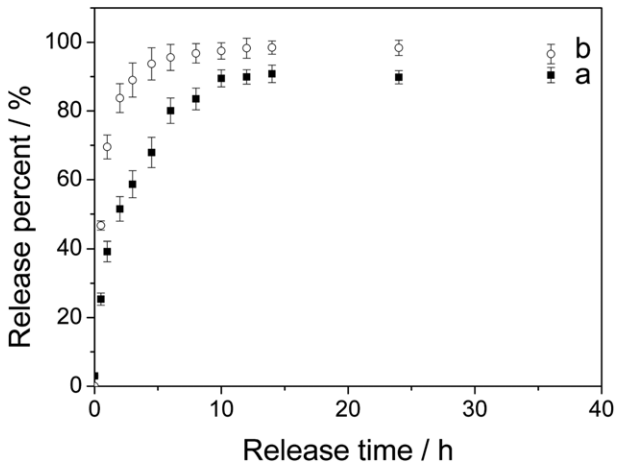



**Figure 7.** Release profiles of ASP from ASP-loaded magnetic MIPs (a) and magnetic NIPs (b) in pH 6.8 PBS solution.

produced SA, respectively. Figure 7 showed the *in vitro* release profiles of ASP from these two polymers at 37 °C. About 50% of the total ASP loaded was released in the first two hours from magnetic MIPs, while magnetic NIPs released about 85% of adsorbed ASP within 2 h. The loaded ASP onto magnetic NIPs mainly is of physical adsorption onto the polymer's surface by a weak interaction between template molecule and polymers, which have been released in the shorter time period. As for magnetic MIPs, quick release of ASP was mainly the weak adsorbed template molecule at the surface of the magnetic MIPs, which could get access to the aqueous medium without the need of long time diffusion. Then in the following period, the release of ASP slowed down and became steady finally in a controlled release manner. This steadier and slower release may be attributed to the fact that ASP was bound in the deeper binding sites. In this case the template molecules that were bound to the magnetic MIPs matrix more tightly were slowly released in the following time period.

## 4. Conclusion

The present study describes the development of magnetic molecularly imprinted polymers for special recognition and controlled release of ASP. MAA and TRIM were used to prepare MIPs at the double-bond-functionalized $Fe_3O_4$ nanoparticle surface. The as-synthesized MIPs have high magnetic responding capacity, which enable themselves to be separated from suspension by an external magnetic field. The magnetic MIPs exhibited good special binding and selectivity capacities to the template molecule. The ASP-loaded magnetic MIPs or NIPs dispersed in pH 6.8 PBS showed a controlled release ASP property. This study indicated that the prepared magnetic MIPs possess the combined properties of selective recognition and controlled release. Further objectives of this work were to synthesize small magnetic MIPs, which can pass through capillary vessels to reach the targeted region by the external magnetic field, and then release the drugs with optimized dosage.

## Acknowledgments

We greatly appreciate the support of the National Natural Science Foundation of China for Key Program (20635020 and 20535020), the Creative Research Group (20721002 and 20521503) and the General Program (20475026 and 20671051). We also thank the support of the National Basic Research Program of China (2007CB925102). Professor Huang-Hao Yang (Qingdao Key Laboratory of Analytical Technology Development) is thanked for many helpful discussions.

## References

[1] Whitcombe M J, Rodriguez M E, Villar P and Vulfson E N 1995 *J. Am. Chem. Soc.* **117** 7105
[2] Wulff G 1995 *Angew. Chem. Int. Edn Engl.* **34** 1812
[3] Matsui J, Higashi M and Takeuchi T 2000 *J. Am. Chem. Soc.* **122** 5218
[4] Yang H-H, Zhang S-Q, Tan F, Zhuang Z-X and Wang X-R 2005 *J. Am. Chem. Soc.* **127** 1378
[5] Yilmaz E, Haupt K and Mosbach K 2000 *Angew. Chem. Int. Edn* **39** 2115
[6] Xie C, Zhang Z, Wang D, Guan G, Gao D and Liu J 2006 *Anal. Chem.* **78** 8339
[7] Southard G E, van Houten K A and Murray G M 2007 *Macromolecules* **40** 1395
[8] Wang H-F, Zhu Y-Z, Yan X-P, Gao R-Y and Zheng J-Y 2006 *Adv. Mater.* **18** 3266
[9] Bolisay L D, Culver J N and Kofinas P 2007 *Biomacromolecules* **8** 3893
[10] Sambe H, Hoshina K, Moaddel R, Wainer I W and Haginaka J 2006 *J. Chromatogr.* A **1134** 88
[11] Liu X, Chen Z, Zhao R, Shangguan D, Liu G and Chen Y 2007 *Talanta* **71** 1205
[12] Sreenivasan K 1999 *J. Appl. Polym. Sci.* **71** 1819
[13] Norell M C, Andersson H S and Nicholls I A 1998 *J. Mol. Recognit.* **11** 98
[14] Piletska E V, Turner N W, Turner A P F and Piletsky S A 2005 *J. Control. Release* **108** 132
[15] Suedee R, Srichana T and Martin G P 2000 *J. Control. Release* **66** 135
[16] Bodhibukkana C, Srichana T, Kaewnopparat S, Tangthong N, Bouking P, Martin G P and Suedee R 2006 *J. Control. Release* **113** 43
[17] Duarte A R C, Casimiro T, Aguiar-Ricardo A, Simplício A L and Duarte C M M 2006 *J. Supercrit. Fluids* **39** 102
[18] Puoci F, Iemma F, Muzzalupo R, Spizzirri U G, Trombino S, Cassano R and Picci N 2004 *Macromol. Biosci.* **4** 22
[19] Perez-Moral N and Mayes A G 2004 *Langmuir* **20** 3775
[20] Li P, Rong F and Yuan C 2003 *Polym. Int.* **52** 1799
[21] Giri S, Trewyn B G, Stellmaker M P and Lin V S-Y 2005 *Angew. Chem. Int. Edn* **44** 5038
[22] Tan C J and Tong Y W 2007 *Anal. Chem.* **79** 299
[23] Thea J M, Melissa J J, Barry W A and Louis A C J 1995 *Anal. Chem.* **67** 1896
[24] Carmen A and Angel C 2004 *J. Chromatogr.* B **804** 231
[25] Yang C, Wang G, Lu Z, Sun J, Zhuang J and Yang W 2005 *J. Mater. Chem.* **15** 4252
[26] Gao D, Zhang Z, Wu M, Xie C, Guan G and Wang D 2007 *J. Am. Chem. Soc.* **129** 7859
[27] Wang X, Ding X, Zheng Z, Hu X, Cheng X and Peng Y 2006 *Macromol. Rapid Commun.* **27** 1180
[28] Ansell R J and Mosbach K 1998 *Analyst* **123** 1611
[29] Li Y, Yin X-F, Chen F-R, Yang H-H, Zhuang Z-X and Wang X-R 2006 *Macromolecules* **39** 4497

[30] Yoshimatsu K, Reimhult K, Krozer A, Mosbach K, Sode K and Ye L 2007 *Anal. Chim. Acta* **584** 112
[31] Sun H, Hong J, Meng F, Gong P, Yu J, Xue Y, Zhao S, Xu D, Dong L and Yao S 2006 *Surf. Coat. Technol.* **201** 250
[32] Ding Y, Hu Y, Zhang L, Chen Y and Jiang X 2006 *Biomacromolecules* **7** 1766
[33] Liu Z L, Ding Z H, Yao K L, Tao J, Du G H, Lu Q H, Wang X, Gong F L and Chen X 2003 *J. Magn. Magn. Mater.* **265** 98
[34] Kobayashi T, Reddy P S, Ohta M, Abe M and Fujii N 2002 *Chem. Mater.* **14** 2499
[35] Ciardelli G, Cioni B, Cristallini C, Barbani N, Silvestri D and Giusti P 2004 *Biosens. Bioelectron.* **20** 1083