Effect of Porogenic Solvent on the Morphology, Recognition and Release Properties of Carbamazepine-Molecularly Imprinted Polymer Nanospheres

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ABSTRACT: This study focus on the effect of the porogenic solvent on the morphology, recognition, and drug release of carbamazepine-molecularly imprinted polymer nanospheres prepared by precipitation polymerization. The scanning electron microscopy (SEM) images and Brunauer-Emmett-Teller (BET) analysis showed that molecularly imprinted polymer (MIP) prepared by acetonitrile exhibited a regular spherical shape at the nanoscale with a high degree of monodispersity, specific surface area of 242 m^2 g^{-1}, and pore volume of 1 mL g^{-1}, while those using chloroform and toluene produced irregular polymer particles with low specific surface area and pore volume. MIP prepared by acetonitrile/chloroform (1 : 1, v/v) showed mediator texture properties compared to MIPs obtained by acetonitrile or chloroform. Results from saturation and displacement assays indicated that the imprinted nanospheres with binding capacity of 2.85 (mg CBZ/g polymer) had high specific affinity to CBZ in contrast to nonimprinted nanospheres (1.63 mg CBZ/g polymer). The imprinted nanospheres with 2.4 selectivity factor had good recognition to CBZ than analog template of oxcarbazepine. Moreover, release studies showed that 20% of loaded CBZ was released from the imprinted nanospheres within the initial 6 h, while another 80% of CBZ was released in the following 9 days. © 2011 Wiley Periodicals, Inc. J Appl Polym Sci 121: 1118–1126, 2011

Key words: molecular imprinting; nanospheres; porogen effects; morphology; drug release

INTRODUCTION

Molecular imprinting technology provides a way to preparation of new polymer materials containing specific recognition sites for target molecules (template) that can be employed in a variety of applications, such as separation, sensors, catalysis, and drug delivery systems.[1–3] The imprinting process is performed by copolymerization of functional monomers and crosslinker in the presence of a template molecule. The functional groups on the monomers are then fixed in polymer network via chemical crosslinking of these monomers.[4] The shape, size, and chemical functionality of the recognition sites are complementary to those of the template molecule. Thus molecularly imprinted polymers (MIPs) can be a highly specific rebind of template molecule.[5] Depending on the nature of interaction between functional monomer and template involved in the imprinting and binding processes, molecular imprinting has three different approaches: covalent, noncovalent, and semicovalent.[6]

MIPs have been prepared in various configurations, including polymer monoliths, membranes, microspheres, and nanospheres for different applications. Typically, MIPs were prepared using bulk polymerization, where the resultant brittle monoliths had to be ground to create a large surface area which is molecularly imprinted, and sieved to the desired particle size. It is time consuming and yields only moderate amounts of useful MIPs, and also the irregular shape of such MIP particles is not ideal for their applications.[7] Recently, novel methods to synthesize MIPs have been reported, including precipitation polymerization,[8] suspension polymerization using fluorocarbon liquid[9] or mineral oil[10] as a

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continuous phase, swelling polymerization,[11] and grafting procedures.[12] By precipitation polymerization can provide uniform polymer particles of one or a few hundred nanometers in diameter. This method is an attractive stabilizer or surfactant-free method, and based on precipitation polymerization of functional monomer/crosslinker in the presence of a template in a high dilution solution (<5% w/v).[13] It is generally applicable to a large variety of template molecules.

In copolymerization of a mono-functional monomer and a crosslinker, two of the most important experiment parameters that determine the physical nature of the polymer product are percentage of the crosslinker and also vary based on the amount and nature of the porogen.[14] Wang et al. reported that matching the solubility parameter of the developing polymer matrix to that of the porogen was particularly important in the control of polymer morphology.[8] At either relatively low crosslinker percentages (e.g., <5%), or at higher crosslinker percentages combined with low amount of porogens (which are thermodynamically compatible with the polymer matrix), the phase separation of the polymer matrix does not occur during polymerization. The product is a gel-type polymer, which has very low specific surface area in the dry state, and poor mechanical properties. At relatively higher crosslinker percentage, and/or in higher amounts of porogen, the polymer matrix is able to phase separate from the porogen giving rise to macroporous polymer. Macroporous polymers are mechanically more robust than gel-type polymers on account of the higher percentage of crosslinker. Under high dilute conditions the primary polymer particles, which fuse to form gel-type polymers or macroporous polymers under low dilute conditions, remain in a non-aggregated state and the yield is microgel powder.[14] The dilution of the reaction system influences the final size of the polymer particles; increasing the amount of porogen leads to a decrease in the size of the polymer particles and polymerization under high dilution conditions allows obtaining the micro- and nanoparticles. In general, polymer particles in the form of microspheres and nanospheres are particularly suitable for the selective binding properties required to MIPs. The accessibility of the binding sites would be facilitated by submicron MIPs.[15]

Another important role of porogen is its effect on the complexing of functional monomers with the template before polymerization (in noncovalent imprinting).[16] The extent of the noncovalent complex is affected by the polarity of the porogen. Low polar porogens will increase complex formation by Le Chatelier's principle. On the other hand, high polar porogens tend to dissociate the noncovalent complexes, particularly protic porogens that afford a high degree of disruption to hydrogen interactions. In covalent imprinting, many kinds of porogens are employable as long as they satisfactorily dissolve all the reagents.[17]

Recently, we successfully applied MIPs as new sensor material in potentiometric detection of hydroxyzine[18] and cetirizine,[19] as new carriers for sustained release of dipyridamole,[20] and as new absorbents for solid-phase extraction of bromhexine,[21] metoclopramide,[22] verapamil,[23] and tramadol[24] from human serum and urine. The variety of applications utilizing MIPs require different MIP formats including films, irregular or spherical particles, along with precise knowledge of MIP characteristics, such as binding efficiency, binding capacity, and template release of these materials.[25] In response to this demand, MIPs were prepared in different formats by using different porogens and the influence of porogen on morphology, binding and release of template was investigated. It is commonly agreed that microspheres and nanospheres are the best MIP formats, so trying to obtain these materials by novel synthetic conditions of precipitation polymerization method. Imprinted polymers have been prepared using methacrylic acid (MAA) as a functional monomer, trimethylolpropane trimethacrylate (TRIM) as a crosslinker, azobisisobutyronitrile (AIBN) as an initiator, and acetonitrile, chloroform, mixture of acetonitrile/chloroform, and toluene as a porogen. A template chosen was carbamazepine (CBZ), an anticonvulsant drug used widely in epilepsy treatment, and MIPs were synthesized via noncovalent imprinting. The carboxylic acid groups of MAA were used to create hydrogen interactions between the polymer matrix and the functional groups of CBZ (such as $CONH_2$) (Fig. 1). The morphologies and porosities of the resulting imprinted materials were characterized by scanning electron microscopy (SEM), photon correlation spectroscopy (PCS), and Brunauer-Emmett-Teller (BET) gas adsorption measurements. Thermal characteristics of polymers were analyzed by differential scanning calorimetry (DSC). The efficiency of the noncovalent imprinting and binding kinetic were examined by measurement of binding capacity in different times by UV absorption. The binding behavior of the CBZ to imprinted polymers was compared to polymers prepared in the absence of the CBZ. The molecular recognition ability of the CBZ-imprinted polymers was studied by comparing CBZ and oxcarbazepine (template analog) binding. To study the nature of imprinted polymer particles on release profiles, CBZ release was carried out in sodium dodecyl sulfate (SDS) solution (1 wt %).

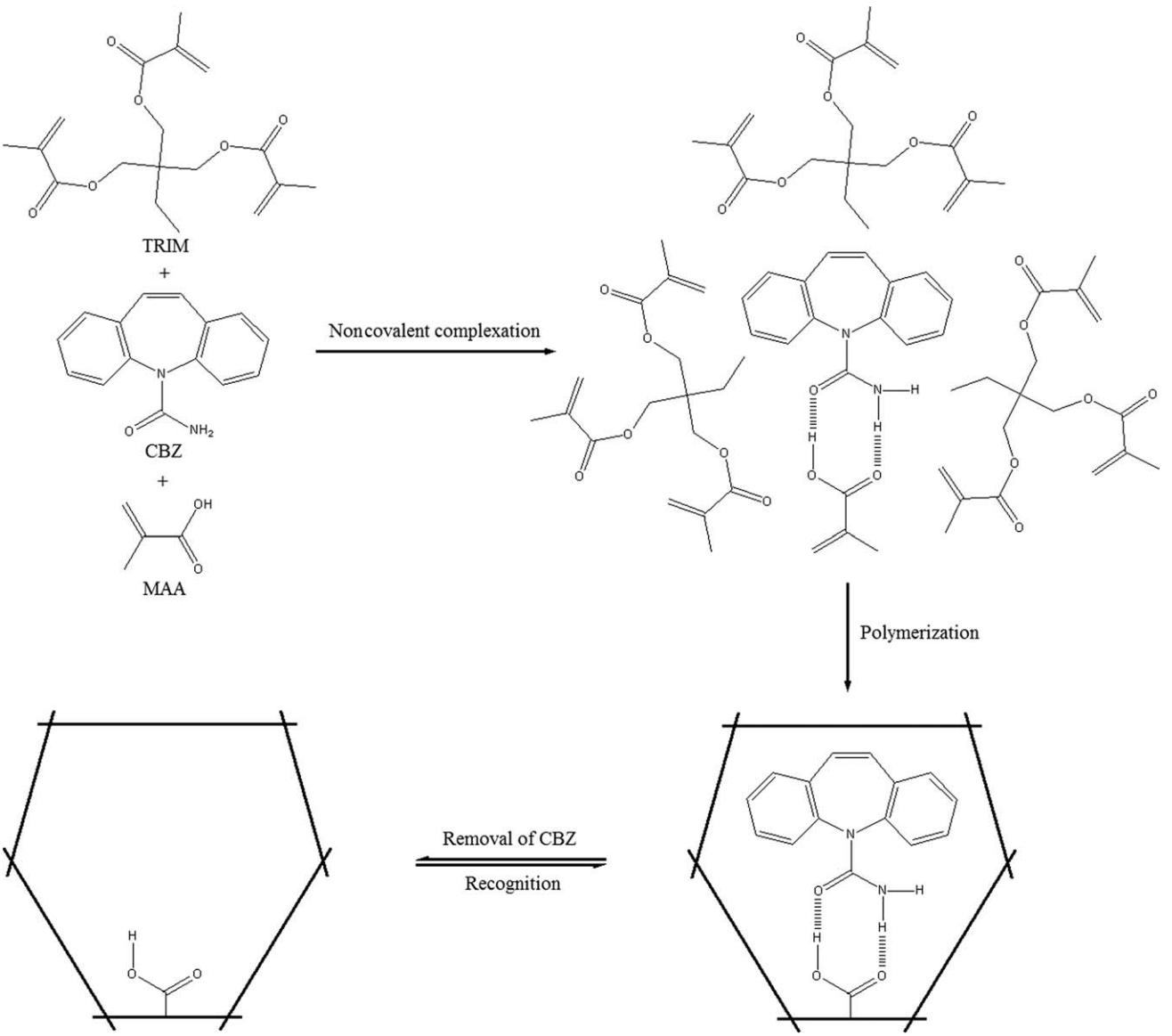

Figure 1 Schematic representation of the carbamazepine-imprinted polymer synthesis.

EXPERIMENTAL

Materials

Trimethylolpropane trimethacrylate (TRIM) and azobisisobutyronitrile (AIBN) were obtained from Σ-Aldrich (Steinheim, Germany). AIBN was re-crystallized from methanol before use. Methacrylic acid (MAA, Darmstadt, Germany) were distilled in vacuum prior to use to remove the polymerization inhibitor. Carbamazepine (CBZ) and oxcarbazepine (OCBZ) were obtained from the Ministry of Health and Medical Education (Tehran, Iran). Sodium dodecyl sulfate (SDS), acetonitrile, chloroform and toluene were purchased from Merck (Darmstadt, Germany). Dialysis tubes (Sigma dialyzes tubes M_w cutoff 12 kDa) were heated in 2 wt % solution of sodium bicarbonate and 0.05 wt % solution of ethylene diamine tetra acetic acid (EDTA) and then kept under refrigeration in 0.05 wt % solution of sodium azide until use. Other reagents and solvents of an analytical-reagent grade were used without further purification.

Synthesis of molecularly imprinted polymers

The schematic representation of the imprinting and the removal of CBZ from the imprinted polymer are shown in Figure 1. The polymers were prepared using precipitation polymerization from solutions of the composition shown in Table I at 60°C. Briefly the functional monomer, MAA, was added to a 50-mL thick walled glass tube containing CBZ dissolved in 40 mL of porogen. In this amount of porogen was

TABLE I
Preparation of Carbamazepine-Imprinted Polymers

Polymer	Template (mmol)	MAA (mmol)	TRIM (mmol)	Porogen
MIP1	0.84	1.68	1.68	Acetonitrile
NIP1	–	1.68	1.68	Acetonitrile
MIP2	0.84	1.68	1.68	Chloroform
NIP2	–	1.68	1.68	Chloroform
MIP3	0.84	1.68	1.68	Acetonitrile/Chloroform (1 : 1, v/v)
NIP3	–	1.68	1.68	Acetonitrile/Chloroform (1 : 1, v/v)
MIP4	0.84	1.68	1.68	Toluene
NIP4	–	1.68	1.68	Toluene

dissolved all reaction component. Then the mixture was sonicated in an ultrasonic bath (Tecno-Gaz, Tecna 6, Italy) for 6 min until clear solution was obtained. The solution was gently mixed for 90 min. The crosslinker, TRIM, and 0.1 mmol of the initiator, AIBN, were added and the solution was sonicated for 6 min to remove oxygen. The solution was saturated with nitrogen for 5 min and the tube sealed under nitrogen gas. The reaction vessel was inserted in a waterbath (Memmert WB14, Germany) at 50 cycles min^{-1}. Polymerization was carried out by increasing temperature from 25 to 60°C within 40 min, and thereafter kept for 20 h. The polymer particles obtained were collected by centrifugation (Sigma 3K30, Germany) at 17,000 rpm for 30 min. The template of unleached imprinted polymers were removed by batch-mode solvent extraction with 40 mL of methanol containing 10% acetic acid (v/v) five times for 1 h, until no template could be detected from the washing solvent by spectrometric measurement (at $\lambda = 285$ nm) (Scinco *S*-3100, Korea). The polymer particles were rinsed in the same volume of deionized water and acetone, and finally the resulting leached imprinted polymers were dried in oven (Memmert INB 400, Germany) at 50°C overnight. Nonimprinted polymers (NIPs) were synthesized under identical conditions without the template.

Differential scanning calorimetry (DSC)

DSC scans of polymer particles were performed on a Mettler DSC 823 (Mettler Toledo, GmbH, Switzerland) equipped with a Julabo Thermocryostate Model FT100Y (Julabo labortechnik GmbH, Germany). A Mettler Star Software System, version 9.x was used for the data acquisition. Indium was used to calibrate the instrument. The samples were scanned at a heating rate of 10°C min^{-1} in 20–470°C temperature range.

Morphologic analysis

Scanning electron microscopy (SEM, PhilipsXL30 Scanning Microscope, Philips, Netherlands) was employed to determine the shape and surface morphology of the produced polymer particles. The polymer particles were sputter coated with gold prior to the SEM measurement. The particle size and size distribution of polymer particles were measured by photon correlation spectroscopy (Malvern Zetasizer ZS, Malvern, UK). The polymer particles were suspensed in acetonitrile and measured at a fixed scattering angle of 90°.

Pore size distribution and specific surface area analysis of polymer particles were measured by nitrogen adsorption-desorption using a BELSORP-Mini II (Japan) at −196°C. Prior to measurement, 150 mg of the polymer particles were heated at 100°C for 2 h under vacuum. The specific surface area, pore volume and average pore diameter of polymer particles were obtained by Brunauer-Emmett-Teller (BET) method using BELSORP analysis software.

Saturation experiment

The binding capacity of the polymer particles was determined through saturation studies at different times. About 40 mg of polymer particles were suspended in 6 mL of 85 μ*M* of CBZ in water/acetonitrile (4 : 1, v/v) solution and mixed for 30 min. The polymer particles were separated by centrifugation at 20,000 rpm for 30 min and the supernatant was analyzed by UV spectrophotometer. The separated polymer particles were again suspended in 6 mL of initial solution (85 μ*M* of CBZ in water/acetonitrile (4 : 1, v/v)) according to the previous stage and the binding capacity obtained in the next 30 min. This process was repeated six times to determine the binding capacity per 30 min during 3 h (binding test in dynamic conditions).

For competitive analysis, selectivity of the imprinted polymers was determined by incubating fixed amounts of polymer particles with 85 μ*M* of template analog (oxcarbazepine) in 6 mL of water/acetonitrile (4 : 1, v/v) and then proceeding to binding experiments.

Accuracy (ratio between the measured concentration and the spiked concentration) was determined by analyzing on six different times (30–180 min) spiked drug samples with concentrations ranging

from 1 to 85 μ*M*. The measured concentrations were between 99.4 and 107.2% of the spiked concentrations, indicating good accuracy of the assay.

In vitro drug release

Drug release from polymer particles was evaluated using the dialysis technique in sodium dodecyl sulfate (SDS) aqueous solution (1 wt %). The good solubility of CBZ in SDS solution (1 wt %) was used to maintain sink conditions.[26] About 50 mg of polymer particles were incubated in solution of CBZ (0.75 m*M* of CBZ in water/acetonitrile (1 : 4 v/v) solution) for 1 h. The polymer particles were separated by centrifugation and unbound CBZ in supernatant was analyzed by UV spectrophotometer. The separated polymer particles were resuspended in 2 mL solution of sodium dodecyl sulfate (1 wt %) and placed in a dialysis tube which is sealed at both ends with medicell clips (Spectrum, USA). The tube soaked in 50 mL of SDS solution (1 wt %). The medium was stirred at 100 rpm. Aliquots of 2 mL were withdrawn from the medium at designated intervals and replaced with 2 mL of fresh medium. The amount of CBZ released from the polymer particles analyzed by UV spectrophotometer.

RESULTS AND DISCUSSION

Thermal analysis results

Figure 2 shows DSC thermograms of NIP1, leached and unleached MIP1, and other MIPs. All polymer particles were thermally stable up to ~ 200°C and the exothermic transitions over 200°C were related to the complex processes of thermal decomposition. NIP1 and leached MIP1 had similar thermograms. An endothermic transition was observed at 190°C of unleached MIP1, which can be explained as melting of CBZ load in polymer. The CBZ has a melting point at 190°C [Fig. 2(g)]. Comparison of peaks area at 190°C shows the drug loading of unleached MIP1 is less than the initial amount used for synthesis.

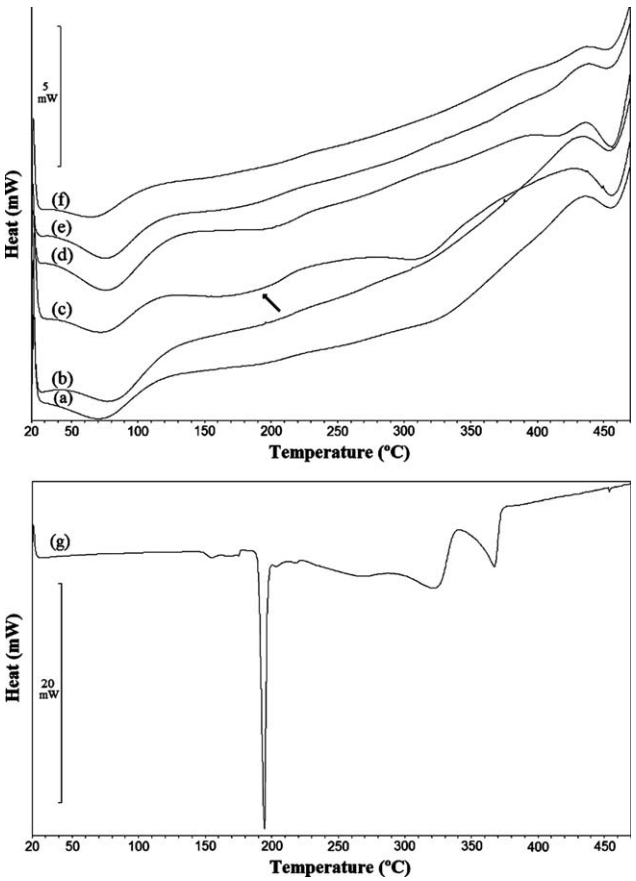

Figure 2 DSC thermograms of NIP1 (a), leached MIP1 (b), unleached MIP1 (c), MIP2 (d), MIP3 (e), NIP4 (f), and carbamazepine (g).

Morphologic analysis

Particle size and shape study of carbamazepine-imprinted polymers

The SEM micrographs (Fig. 3) for polymer particles obtained in different porogens showed appreciable differences. Figure 3(a–d) show SEM micrographs of imprinted and nonimprinted polymers prepared by acetonitrile, respectively. By using high dilution conditions of monomer in precipitation polymerization a 1 : 2 : 3 molar ratio of CBZ : MAA : TRIM were obtained of submicron polymer particles. These polymer particles were spherical and high uniformly sized in nanoscale.

It order to study the size and size distribution of the prepared poly(MAA-*co*-TRIM) nanospheres, MIP1 and NIP1 were re-suspended in acetonitrile, and characterized with photon correlation spectroscopy (PCS). The average diameter of MIP1 and NIP1 was 150 and 134 nm, respectively. Compared to the SEM micrographs in Figure 3(a–d) shows the size of nanospheres was increased about 50% after suspended in acetonitrile. Figure 4 shows a very narrow particle size distribution for nanospheres prepared by acetonitrile. The polydispersity index was 0.063 for MIP1 and 0.083 for NIP1. The monodispersed size for nanospheres is a desirable characteristic in many applications, such as competitive testing of ligand-bound, solid-phase micro extraction (SPME), and development of sensors using microsphere deposition, capillary electrochromatography, and drug delivery systems.[6] The PCS results for other polymer particles (prepared by chloroform, acetonitrile/chloroform (1 : 1, v/v), and toluene) were not acceptable, because they had high polydispersity index and therefore the PCS results had significant error.

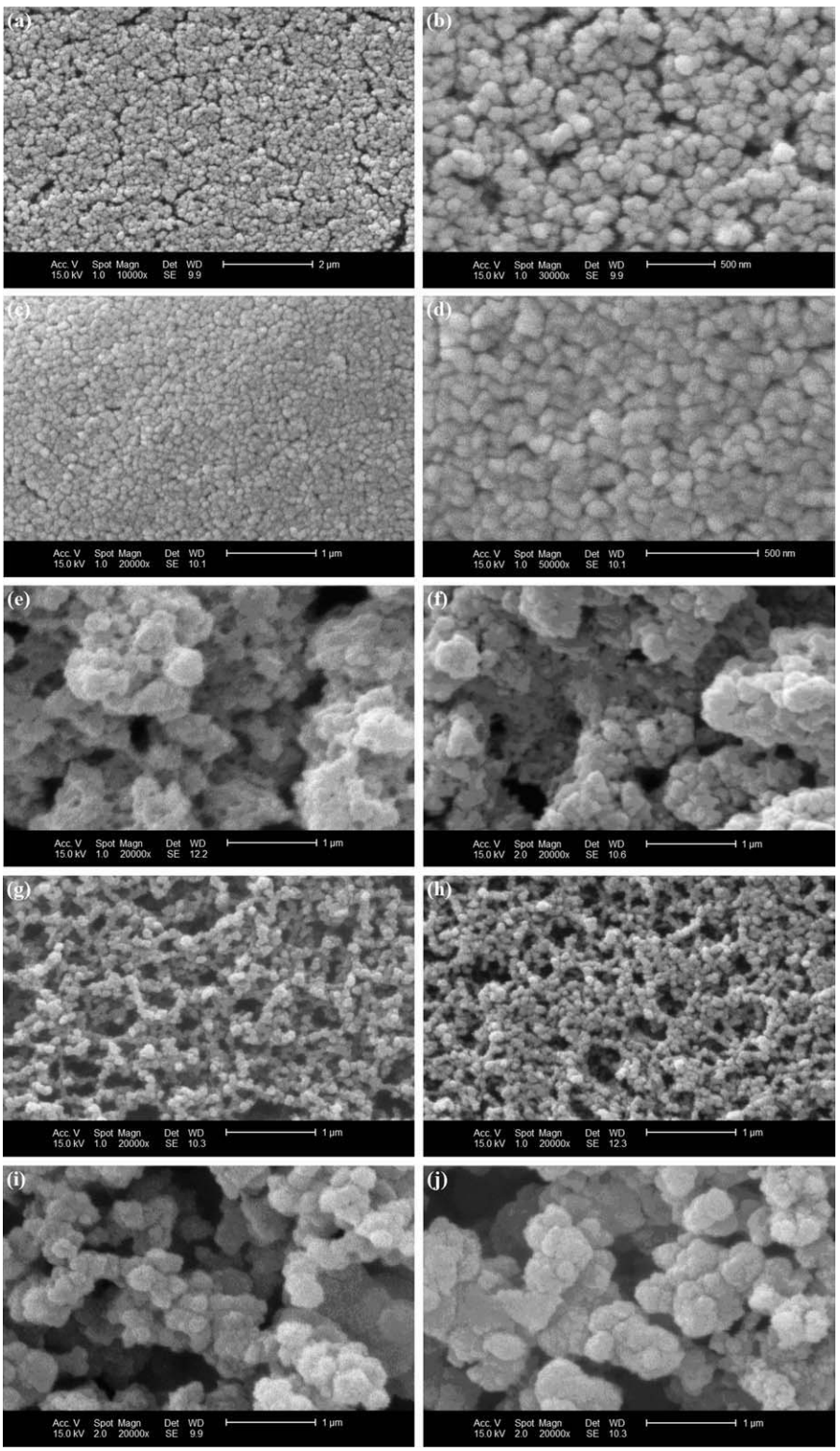

Figure 3 SEM of crosslinked poly(MAA-*co*-TRIM) particles; MIP1 (a and b), NIP1 (c and d), MIP2 (e), NIP2 (f), MIP3 (g), NIP3 (h), MIP4 (i), and NIP4 (j).

Figure 3(e,f) show SEM micrographs of imprinted and nonimprinted polymers prepared by chloroform, respectively. It is obvious that the use of chloroform leads to the formation of nanosponge aggregates which probably can be ascribed to the fact that the chlorinated porogens cause severe swelling of the polymer product.[27] The resulting polymer particles had irregular and rough morphology.

Figure 3(g,h) show SEM micrographs of imprinted and nonimprinted polymers prepared by acetonitrile/

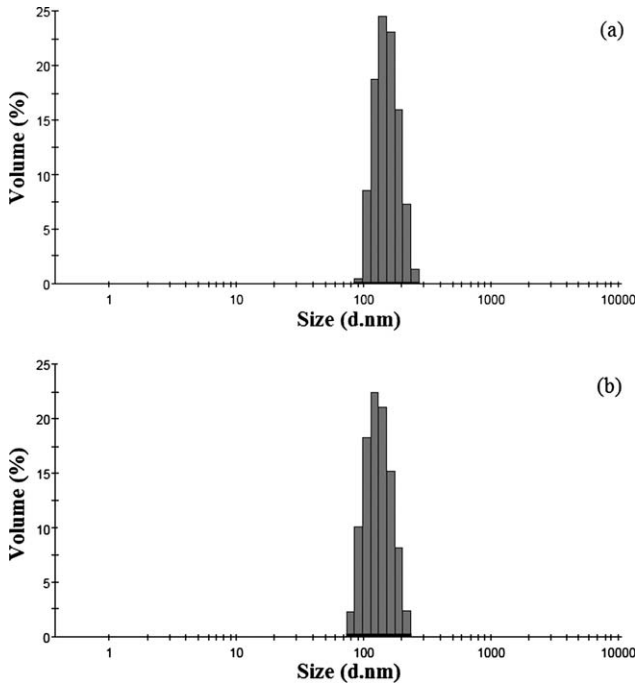

Figure 4 Particle size distribution of MIP1 (a) and NIP1 (b) measured by photon correlation spectroscopy.

chloroform (1 : 1, v/v), respectively. Micrographs show polymer particles with different size and shape (more spherical) which can be considered collections of spherical polymer particles resulting from acetonitrile and irregular polymer particles resulting from chloroform.

Figure 3(i,j) show SEM micrographs of imprinted and nonimprinted polymers prepared by toluene, respectively. The resulted polymer particles had irregular and slightly smooth morphology.

Furthermore, in all cases both MIPs and NIPs prepared with the same porogen had similar morphology, and the presence of CBZ in prepolymerization mixture had no effect on morphology of obtained imprinted particles.

Porosity measurement

Table II shows the texture properties (specific surface area, pore volume, and average pore diameter) of the MIPs and NIP1, measured by nitrogen adsorption porosimetry. These data indicate that different porogens had significant effects on the porosity of polymer particles. Nanospheres prepared by acetonitrile (MIP1 and NIP1) had high specific surface area and were mesoporous polymer particles. The polymer particles prepared by chloroform (MIP2) and toluene (MIP4) had mesoporous structure with low specific surface area and pore volume. MIP3 with the specific surface area of 121 ($m^2\ g^{-1}$) showed mediator porosity properties compared to polymer particles prepared by acetonitrile (MIP1) or chloroform (MIP2).

The porous structure of polymer results from a phase separation between the polymer and the porogen during the polymerization. When the nuclei of the growing polymer chains are fused, the cross-linked polymeric matrix is formed and a series of various pores containing the porogen are created. The porous structure makes the extraction, binding and release of the template easier. Polymers prepared in the absence of porogens are consistently too firm and dense, and have low mass transfer in binding sites. The polymers with a wide range of porosities can be produced by controlling the point of phase separation.[4]

The texture properties of MIP1 nanospheres are similar to its corresponding nonimprinted polymer (NIP1) prepared with the same porogen, therefore the addition of template to the prepolymerization mixture does not significantly affect the polymer porosity.

Kinetics of molecular binding by imprinted and nonimprinted polymers

The binding capacities of imprinted and nonimprinted polymers were determined at different times by taking samples every 30 min for 3 h and replacing the supernatant with fresh solution [Fig. 5(a)]. The initial, quick binding of CBZ was shown, such that MIPs bound 50% of total binding observed in 30 min, and in this time NIPs bound 65%, and also earlier than their MIPs reached to saturation capacity. After 90 min, the amount of bound CBZ was almost constant.

All imprinted polymers had more binding capacity than nonimprinted polymers, indicating that there were specific binding sites for CBZ. The template binding by the nonimprinted polymer can be explained with the presence of nonspecific binding due to physical adsorption, and to random interactions of the template molecules with functional groups in the polymer matrix. Regarding the binding capacity for CBZ in MIP1 (2.85 mg CBZ/g polymer) which is about 1.75 times than that of this value in NIP1 (1.63 mg CBZ/g polymer), MIP1 showed most efficient imprinted polymer with respect to the nonimprinted polymer. MIP1

TABLE II
Results of BET Analysis for Polymers Prepared by Different Porogens

Polymer	Specific surface area ($m^2\ g^{-1}$)	Pore volume (mL g^{-1})	Average pore diameter (nm)
MIP1	242	1.00	16
NIP1	268	0.95	14
MIP2	18	0.10	22
MIP3	121	1.05	35
MIP4	6	0.03	20

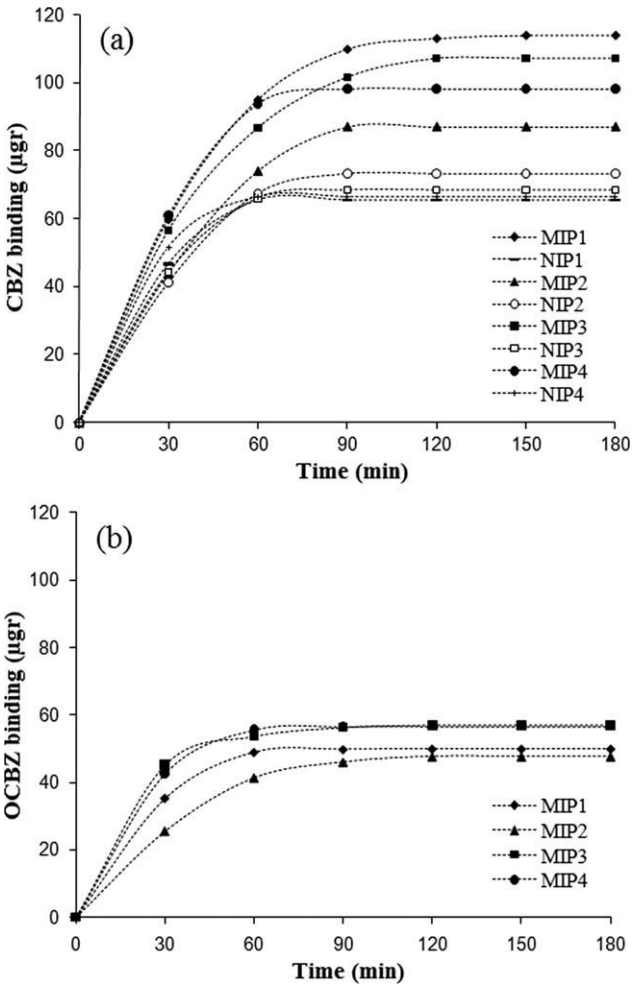

Figure 5 Template and analog template binding by 40 mg of polymers with time (continuous assay): (a) CBZ binding, (b) OCBZ binding.

nanoparticles are spherically shape with high specific surface area, which can be based on improved efficiency of solid-liquid contact and increased available binding sites for CBZ. The binding capacity in continuous assay was 2.18, 2.68, and 2.46 mg CBZ bound by 1 g of MIP2, MIP3, and MIP4, respectively, which is about 1.19, 1.57, and 1.48 times of CBZ bound by their nonimprinted polymers, respectively. As can be observed, when chloroform was used as porogen, MIP2 showed low binding capacity and imprinting efficiency. This can probably be ascribed to the fact that chlorinated porogens cause severe swelling of the polymer particles, as mentioned previously, and hence the change of swelling degree in other solutions leads to changes in the three-dimensional configuration of the functional groups taking part in binding sites, resulting in poor binding efficiency.[27]

MIP3 with better morphology (small size polymer particles with high specific surface area) than MIP2 and with undesirable characteristic of the change in the three-dimensional configuration of the functional groups taking part in binding sites (due to the presence of chloroform in polymerization mixture) toward MIP1, showed a mediator binding efficiency compared to imprinted polymers prepared by acetonitrile (MIP1) or chloroform (MIP2).

The molecular recognition ability of the imprinted polymers was studied by comparing CBZ and oxcarbazepine (OCBZ) binding by CBZ-imprinted polymers. OCBZ has a similar structure than CBZ (Fig. 6). All imprinted polymers showed better binding to CBZ than OCBZ [Fig. 5(b)]. OCBZ binding to CBZ-imprinted polymer is the result of the nonselective and nonspecific bound. The amount of OCBZ bound was almost low and equal for all imprinted polymers. After 90 min, the nonselective binding of OCBZ was stopped. After 3 h, MIP1 nanospheres bound almost 95% of the CBZ in the solution (114 μg) and only 39% of OCBZ (50 μg), confirming the presence of highly specific binding sites on the imprinted nanospheres.

By comparing CBZ and OCBZ binding the selectivity factor α, defined as the capacity for the template molecule to the capacity for the template analog molecule, was calculated. In particular, we calculated for MIP1 $\alpha_{CBZ/OCBZ} = 2.4$, MIP2 $\alpha_{CBZ/OCBZ} = 1.9$, MIP3 $\alpha_{CBZ/OCBZ} = 2$, and for MIP4 $\alpha_{CBZ/OCBZ} = 1.9$. Regarding the selectivity factor for CBZ in MIP1 imprinted nanospheres, which is about 2.4 times than OCBZ template analog as expected, MIP1 showed most efficient CBZ-imprinted polymer with respect to other imprinted polymers.

In vitro drug release studies

The pattern of drug release from MIPs and NIP1 in sodium dodecyl sulfate (SDS) solution (1% wt) is shown in Figure 7. MIP1 showed most slow release and as mentioned previously, was most efficient imprinted polymer. MIP1 released a considerable amount of CBZ during the first 6 h (about 20% of the loaded CBZ at the beginning). Subsequently another 80% of CBZ was released in 220 h. NIP1, MIP2, MIP3, and MIP4 released about 34, 32, 28, and 26% of CBZ in 6 h, respectively. Initially, the quick release of CBZ is to be ascribed to surface adsorbance and available

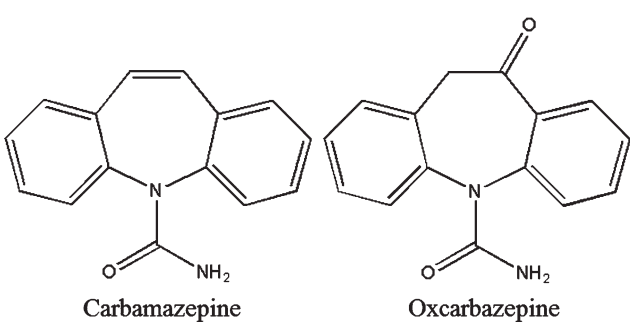

Figure 6 Chemical structure of template and template analog molecules.

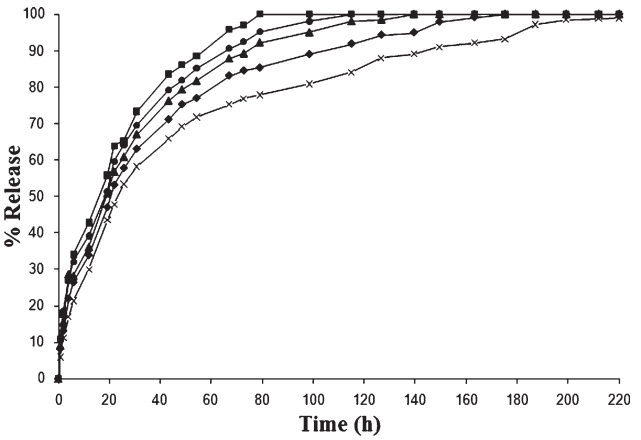

Figure 7 Pattern of drug release from MIP1 (×), NIP1 (■), MIP2 (●), MIP3 (▲), and MIP4 (◆) SDS (1 wt %).

template molecules, while those tightly bound are slowly released in the following time periods.

There was an evident difference in release kinetics between MIP1 and NIP1. NIP1 showed faster release than MIP1 and other imprinted polymers, and released a total of loaded CBZ in 80 h. The fast release of NIP1 was due to absence specific binding sites for CBZ. The total loaded CBZ of MIP2 and MIP3 released in 115 and 140 h, respectively. The fast release of MIP3 and specially MIP2 toward other MIPs were probably related to damage of the three-dimensional configuration of the binding sites by chloroform as porogen. Other reason for fast release of MIP3 toward MIP4 was most accessibility of binding sites in MIP3 (small polymer particles with high specific surface area). The total loaded CBZ of MIP4 is released in 175 h.

Using TRIM as crosslinker at a molar ratio of 3 : 2 with respect to the functional monomers, the resulted CBZ-imprinted polymer was sufficiently rigid to keep the binding sites and give to the material good recognition properties, showing at the same time appreciable release capacity.[6,28]

CONCLUSION

Imprinted polymers composed of poly(MAA-*co*-TRIM), were synthesized by precipitation polymerization allowing direct noncovalent imprinting of carbamazepine. The nature of porogen had an important role in the morphological properties of MIPs. For a given functional monomer, crosslinker and template molecule, choice of the composition of the porogen is the most important factor to control the phase separation to yield uniform MIP nanospheres. The relationships between the morphology and porosity of particles, binding and release properties were detailed, providing useful guidelines for controlling the MIP particle properties for the desired application including analytical fields and drug delivery systems. In general, the specific binding and release properties were the influence of morphology of polymer particles and the nature of porogen in polymerization reaction. Molecular imprinting was most effective when acetonitrile was used as porogen for the preparation of imprinted polymers. The MIP nanospheres (average diameter of 150 nm) revealed excellent binding properties at equilibrium binding conditions and can be used for specific capturing of template from dilute solutions. It was also confirmed that the porosity of MIPs plays an important role in their binding properties.

References

1. Wuff, G. Angew Chem Int Ed Engl 1995, 34, 1812.
2. Kriz, D.; Ramstrom, O.; Mosbach, K. Anal Chem 1997, 69, 345.
3. Perez-Moral, N.; Mayes, A. G. Anal Chim Acta 2004, 504, 15.
4. Ye, L.; Mosbach, K. React Funct Polym 2001, 48, 149.
5. Gonzalez, G. P.; Hernando, P. F.; Alegria, J. S. D. Anal Chim Acta 2006, 557, 179.
6. Ciardelli, G.; Cioni, B.; Cristallini, C.; Barbani, N.; Silvestri, D.; Giusti, P. Biosens Bioelectron 2004, 20, 1083.
7. Wulff, G.; Sarhan, A.; Zabrocki, K. Tetrahedron Lett 1973, 14, 4329.
8. Wang, J.; Cormack, P. A. G.; Sherrington, D. C.; Khoshdel, E. Angew Chem Int Ed 2003, 42, 5336.
9. Mayes, A. G.; Mosbach, K. Anal Chem 1996, 68, 3769.
10. Kempe, H.; Kempe, M. Macromol Rapid Commun 2004, 25, 315.
11. Hosoya, K.; Yoshizako, K.; Tanaka, N.; Kimata, K.; Araki, T.; Haginaka, J. Chem Lett 1994, 8, 1437.
12. Yilmaz, E.; Haupt, K.; Mosbach, K. Angew Chem Int Ed 2000, 39, 2115.
13. Ye, L.; Cormack, P. A. G.; Mosbach, K. Anal Commun 1999, 36, 35.
14. Cormack, P. A. G.; Elorza, A. Z. J Chromatogr B 2004, 804, 173.
15. Vaihinger, D.; Landfester, K.; Krauter, I.; Brunner, H.; Tovar, G. E. M. Macromol Chem Phys 2002, 203, 1965.
16. Spivak, D. A. Adv Drug Deliv Rev 2005, 57, 1779.
17. Komiyama, M.; Takeuchi, T.; Mukawa, T.; Asanuma, H. Molecular Imprinting: From Fundamentals to Applications; Wiley-VCH: Weinheim, 2003.
18. Javanbakht, M.; Fard, S. E.; Mohammadi, A.; Abdouss, M.; Ganjali, M. R.; Norouzi, P.; Safaraliee, L. Anal Chim Acta 2008, 612, 65.
19. Javanbakht, M.; Fard, S. E.; Abdouss, M.; Mohammadi, A.; Ganjali, M. R.; Norouzi, P.; Safaraliee, L. Electroanalysis 2008, 20, 2023.
20. Javanbakht, M.; Mohammadi, S.; Esfandyari-Manesh, M.; Abdouss, M. J Appl Polym Sci 2011, 119, 1586.
21. Javanbakht, M.; Namjumanesh, M. H.; Akbari-Adergani, B. Talanta 2009, 80, 133.
22. Javanbakht, M.; Shaabani, N.; Akbari-Adergani, B. J Chromatogr B 2009, 877, 2537.
23. Javanbakht, M.; Shaabani, N.; Mohammadi, A.; Abdouss, M.; Ganjali, M. R.; Norouzi, P. Curr Pharm Anal 2009, 5, 269.
24. Javanbakht, M.; Attaran, A. M.; Namjumanesh, M. H.; Esfandyari-Manesh, M.; Akbari-Adergani, B. J Chromatogr B 2010, 878, 1700.
25. Wei, S.; Mizaikoff, B. Biosens Bioelectron 2007, 23, 201.
26. Katzhendler, I.; Azoury, R.; Friedman, M. J Control Release 2000, 65, 331.
27. Chaitidou, S.; Kotrotsiou, O.; Kotti, K.; Kammona, O.; Bukhari, M.; Kiparissides, C. Mater Sci Eng B 2008, 152, 55.
28. Silvestri, D.; Borrelli, C.; Giusti, P.; Cristallini, C.; Ciardelli, G. Anal Chim Acta 2005, 542, 3.