Journal of Chromatography B, 908 (2012) 137–142

Contents lists available at SciVerse ScienceDirect

Journal of Chromatography B

journal homepage: www.elsevier.com/locate/chromb

Rapid and selective screening of melamine in bovine milk using molecularly imprinted matrix solid-phase dispersion coupled with liquid chromatography-ultraviolet detection

Hongyuan Yan*, Xiaoling Cheng, Ning Sun, Tianyu Cai, Ruijun Wu, Kun Han

Key Laboratory of Pharmaceutical Quality Control of Hebei Province & College of Pharmacy, Hebei University, Baoding 071002, China

ARTICLE INFO

Article history:
Received 13 August 2012
Accepted 13 September 2012
Available online 19 September 2012

Keywords:
Matrix solid-phase dispersion
Dummy imprinted polymers
Melamine
High performance liquid chromatography
Milk samples

ABSTRACT

A simple, convenient and high selective molecularly imprinted matrix solid-phase dispersion (MI-MSPD) using water-compatible cyromazine-imprinted polymer as adsorbent was proposed for the rapid screening of melamine from bovine milk coupled with liquid chromatography-ultraviolet detection. The molecularly imprinted polymers (MIPs) synthesized by cyromazine as dummy template and reformative methanol–water system as reaction medium showed higher affinity and selectivity to melamine, and so they were applied as the specific dispersant of MSPD to extraction of melamine and simultaneously eliminate the effect of template leakage on quantitative analysis. Under the optimized conditions, good linearity was obtained in a range of 0.24–60.0 $\mu g\,g^{-1}$ with the correlation coefficient of 0.9994. The recoveries of melamine at three spiked levels were ranged from 86.0 to 96.2% with the relative standard deviation (RSD) $\leq$ 4.0%. This proposed MI-MSPD method combined the advantages of MSPD and MIPs, and could be used as an alternative tool for analyzing the residues of melamine in complex milk samples.

1. Introduction

Melamine (1,3,5-triazine-2,4,6-triamine, MEL) is a organic compound that is often used with formaldehyde to produce MEL resin, a synthetic polymer as fire resistant and heat tolerant [1]. Owing to its high nitrogen content and low price, MEL was intentionally added to raw milk to increase its total nitrogen concentration, which will possibly result in higher measurement of protein [2]. Though, low level MEL does not pose danger to humans, high and prolonged dietary exposure to MEL can increase the incidence of urinary bladder tumors and result in the formation of lethal kidney stones [3,4], especially when combined with cyanuric acid [5]. In 2008, MEL caused the death of certain infants in China, who had drunk milk containing this compound. However, milk being a very complex substrate, the determination of a very low MEL from raw milk is difficult. Therefore, a reliable method is needed to determine MEL residues in food and particularly in dairy products for children, which is of biological, clinical, and food industry importance.

High performance liquid chromatography (HPLC) [6–10], gas chromatography [11,12], capillary electrophoresis [13,14] and spectroscopy [15,16] had been developed for the analysis of MEL in different matrices. In view of the complexity of the bio-matrix, repeated centrifugation, protein precipitation and re-extractions were often required to concentrate and purify MEL for further liquid–liquid extraction and solid-phase extraction (SPE) [17,18]. These processes were complicated, time consuming, low selectivity, and use large amounts of organic solvents. Therefore, there is considerable tendency for further improving sample pretreatment techniques, leading to simplify the procedure and enhance its selectivity [19].

Matrix solid-phase dispersion (MSPD) is one of the most promising techniques to combination the procedures of homogenization, disruption, and extraction [20–23]. It permits complete dispersion of sample with the aid of shearing forces generated by blending process and offers exhaustive extraction, clean-up and concentration of analytes in a single step, which eliminates the most of the complications of performing classical SPE process and making sample preparation easier and faster. It drastically shortens the analysis time and reduces the consumption of toxic and expensive solvents, especially for solid, semisolid and highly viscous biological samples [22,24,25]. However, the routine dispersants for extracting MEL, such as mixed-mode cation exchange (MCX), polymer cation exchange (PCX), and strong cation exchange (SCX), are lack selectivity for analytes, which would lead to the coextraction of interferents and affect the quantification of analytes. Therefore, further improving the selectivity of MSPD is still a meaningful work [26].

Molecular imprinting is a rapidly developing technique for the preparation of polymers having specific molecular recognition

* Corresponding author. Tel.: +86 312 5971107; fax: +86 312 5971107.
E-mail addresses: yanhongyuan@126.com, yanhy@hbu.edu.cn (H. Yan).

1570-0232/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.jchromb.2012.09.022

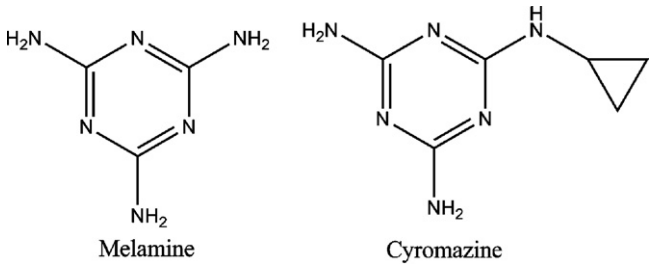

Fig. 1. Chemical structures of MEL and CYR.

properties for a given compound, its analogs or for a single enantiomer [27–29]. As the artificial polymers, MIPs are easy and rapid to prepare, very stable in harsh conditions and allow the usage of a great variety of binding/eluting conditions without the risk of losing binding activity. Due to the high selectivity and stability of MIPs, it has been widely used as a new selective adsorbent in SPE for extracting organic compounds from complex materials [30,31]. Until now, few MIPs synthesized using cyromazine as dummy template had been reported to recognize MEL by SPE method with complicated pretreatment [9,17], however, using cyromazine-MIPs as special dispersant of MSPD without redundant extraction procedure was still not available.

This work represents the first attempt of using MIPs as special dispersant of MSPD to develop a new MI-MSPD method for selective extraction MEL in milk. The water-compatible MIPs synthesized using cyromazine as a dummy template showed high affinity and selectivity to MEL, and the extracts after MI-MSPD were clean enough to inject into HPLC for further chromatographic analysis. This method not only shortened the overall procedure with the improved selectivity but also eliminated the effect of template leakage on quantitative analysis.

2. Experimental

2.1. Chemicals and reagents

MEL and cyromazine (CYR) were obtained from Fuchen Chemical Co. Ltd. (Tianjin, China) and their molecular structures were shown in Fig. 1. Ammonia and chloroform were obtained from Yongfei Chemical Reagent Co. (Tianjin, China). Methanol and acetic acid were purchased from Huadong Chemical Reagent Co. Ltd. (Tianjin, China). 2,2-Azobisisobutyronitrile (AIBN), acetonitrile, and methacrylic acid (MAA) were obtained from Kermel Chemical Reagents Development Center (Tianjin, China), and ethylene glycoldimethacrylate (EGDMA) was purchased from Sigma–Aldrich (St. Louis, MO, USA). Double-deionized water was filtered with 0.45 μm filter membrane before using.

2.2. Instrumentation and conditions

LC analysis was performed using a LC-20A system equipped with two LC-20AT Solvent Delivery Units, a SUS-20A gradient controller and a SPD-20A UV-Vis Detector (Shimadzu, Kyoto, Japan). An N-2000 data workstation (Zheda Zhineng Co. Ltd., Hangzhou, China) was used as a data acquisition system, and an ultrasonic cleaner (KQ3200E, Kunshan Instrument Co., Jiangsu, China) was set at 40 kHz. The LC condition was as follows: the ultimate AQ-C_{18} column (250 mm × 4.6 mm I.D., 5 μm) was obtained from Welch Materials, Inc. (Maryland, USA). The mobile phase was methanol–water (2:98, v/v) with a flow rate of 1.0 mL min^{-1}. The detection wavelength of the detector was set at 210 nm. All of the glassware for preparation of the samples and standard solutions were washed with deionized water and acetone and then dried at room temperature.

2.3. Preparation of the MIPs

The schematic illustration of MIPs formation was shown in Fig. 2 (each kind of polymers was synthesized in triple). CYR (2.0 mmol) and MAA (16.0 mmol) were dissolved in 10.0 mL of porogenic solvents (methanol–water, 10:1, v/v) in a 20.0 mL glass with a lid. The bottle was sonicated for 5 min and then thermostated at 4 °C for 30 min to facilitate template–monomer complex formation. Thereafter, EGDMA (50.0 mmol) and AIBN (0.36 mmol) were added, and the solution was sonicated for 5 min to dissolve it fully. The polymerization was performed at 60 °C for 24 h in a water bath. After polymerization, the monolith was crushed, ground, and sieved through a 300 mesh steel sieve. Following soxhlet extraction with 300 mL methanol–acetic acid (9:1, v/v) for 24 h to be free of the residual reagent and template molecules, the polymer was washed using 300 mL of methanol for 10 h, and then dried in vacuum oven at 60 °C for 12 h to obtain the MIP material. The non-imprinted polymers (NIPs, in the absence of template) were prepared and treated in an identical manner.

2.4. Procedure of MI-MSPD

0.1 g aliquot of milk sample was placed in a 5.0 mL glass beaker containing 0.1 g of MIPs particles, and then the mixture was blended together using a glass nail to obtain a complete disruption and dispersion of sample on solid support. Then the homogeneous mixture was loaded into an empty cartridge (60 mm × 8.0 mm, I.D.), which was pre-packed with 20 mg of MIPs particles, and then compacted by another frit on the top. The cartridge was rinsed with 3.0 mL acetonitrile and eluted with 3.0 mL methanol–ammonia (95:5, v/v). The eluent was evaporated at 50 °C to dryness under vacuum oven and the residues were re-dissolved in 0.05 mL of mobile phase for further HPLC analysis.

2.5. Absorption experiments

To investigate the binding capacity of the synthesized MIPs, static absorption and dynamic absorption test were carried out in aqueous solution. An aliquot of 20.0 mg of MIP particles was added in 10.0 mL flask containing 2.00 mL MEL solutions with various concentrations (1.00–100.0 μg mL^{-1}). After shaking in table concentrator at room temperature, the samples were centrifuged, and then the free concentration of MEL was determined by HPLC. Dynamics method was the same as static method except different times (5, 10, 20, 30, 60, and 90 min) at the constant concentration (20.0 μg mL^{-1}). The absorption quantity (Q) was calculated by subtracting the free concentration (C_{free}) from initial concentration. The same experiment was performed using the non-imprinted particles. Meanwhile, the maximum binding quantity (Q_{max}) and dissociation constant (K_D) were estimated by processing with Scatchard equation:

$$\frac{Q}{C_{free}} = \frac{Q_{max} - Q}{K_D}$$

3. Results and discussion

3.1. Synthesis and molecular recognition of the MIPs

The design criteria governing the production of the MIPs dictate that it is necessary to obtain MIPs materials capable of binding MEL selectively from aqueous samples, and the materials are also able to circumvent the problem of template bleeding [17]. MEL is polar compound, and only in polar solvents, it can be completely dissolved for bulk polymerization. In order to obtain MIPs which demonstrate specific recognition ability to MEL in aqueous media,

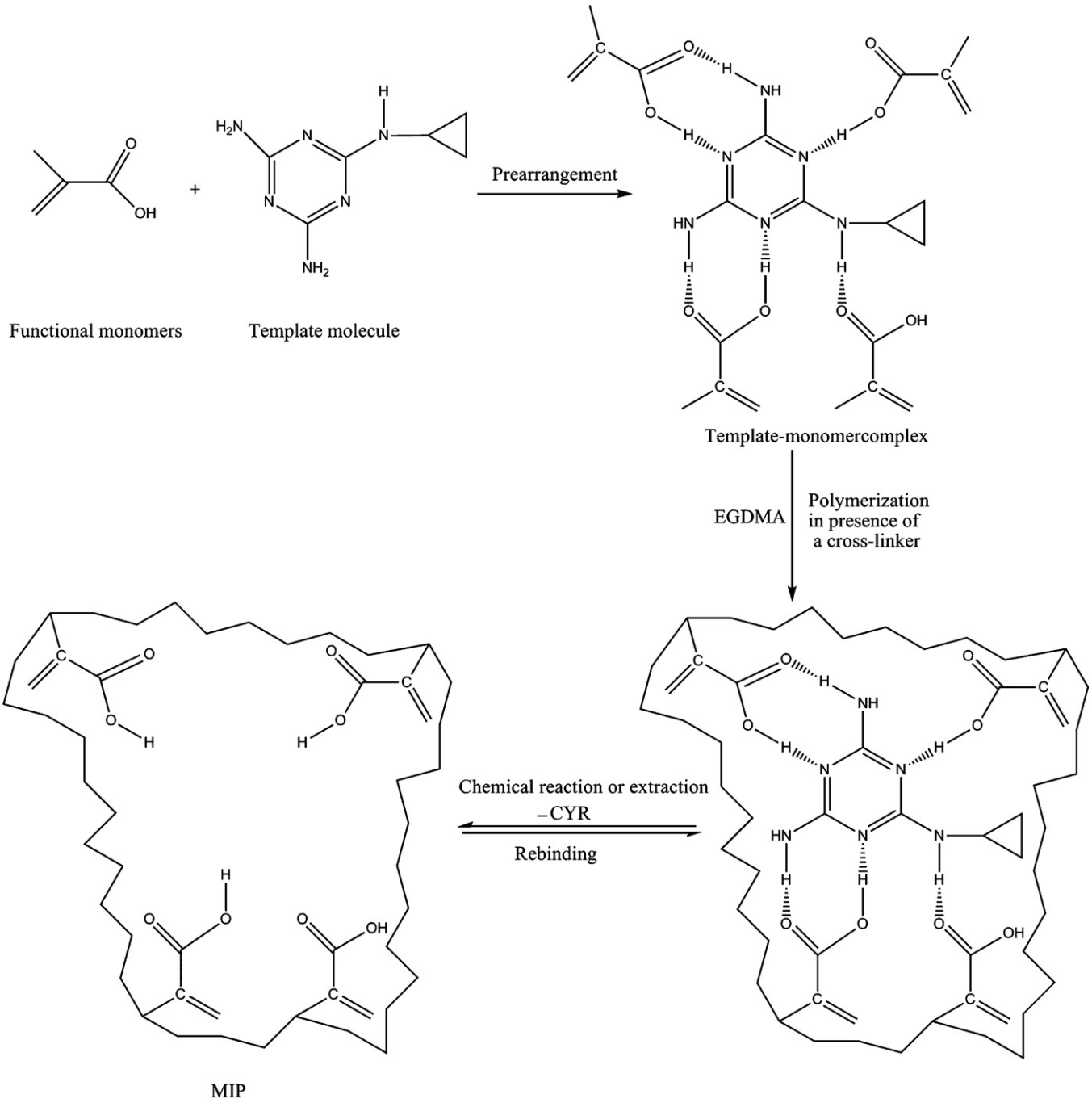

Fig. 2. Schematic illustration of the MIPs formation.

MIPs using MEL or CYR (having the common structures of MEL) as template, MAA as monomer and EGDMA as cross-linker were synthesized in polar porogenic solvents such as water containing methanol or ethanol system. The results revealed that the MIPs prepared using CYR as dummy template and MAA as monomer in methanol/water system showed good molecular recognition to MEL in aqueous media. Moreover, the proportion of water in the prepolymerization mixtures had a critical effect on the pore properties and surface area of the resulting polymers, which was due to water as the porogenic solvent not only brought all the components into one phase but also created macropore structures in the imprinted polymers [27]. Further increase of the water ratio in methanol resulted in a flexible polymer with a small surface area. Conversely, the best selective MIPs for MEL were obtained when using methanol–water (10:1, v/v) system as a porogenic solvent. Under the optimal composition, the MIP using CYR as dummy template had almost same recognition to MEL in aqueous media with MEL imprinted polymer. Besides, the peaks of MEL and CYR could separate, respectively, under the followed HPLC analysis, thus eliminated the effect of template leakage on quantitative analysis of MEL in milk samples.

3.2. Binding assays of the MIPs

The data of static absorption in Fig. 3 showed that the amounts of bound MEL on the MIPs were increased with the increase of the initial concentration of MEL, and the MIPs displayed a higher affinity than NIPs [9]. The high affinity of MIPs was also

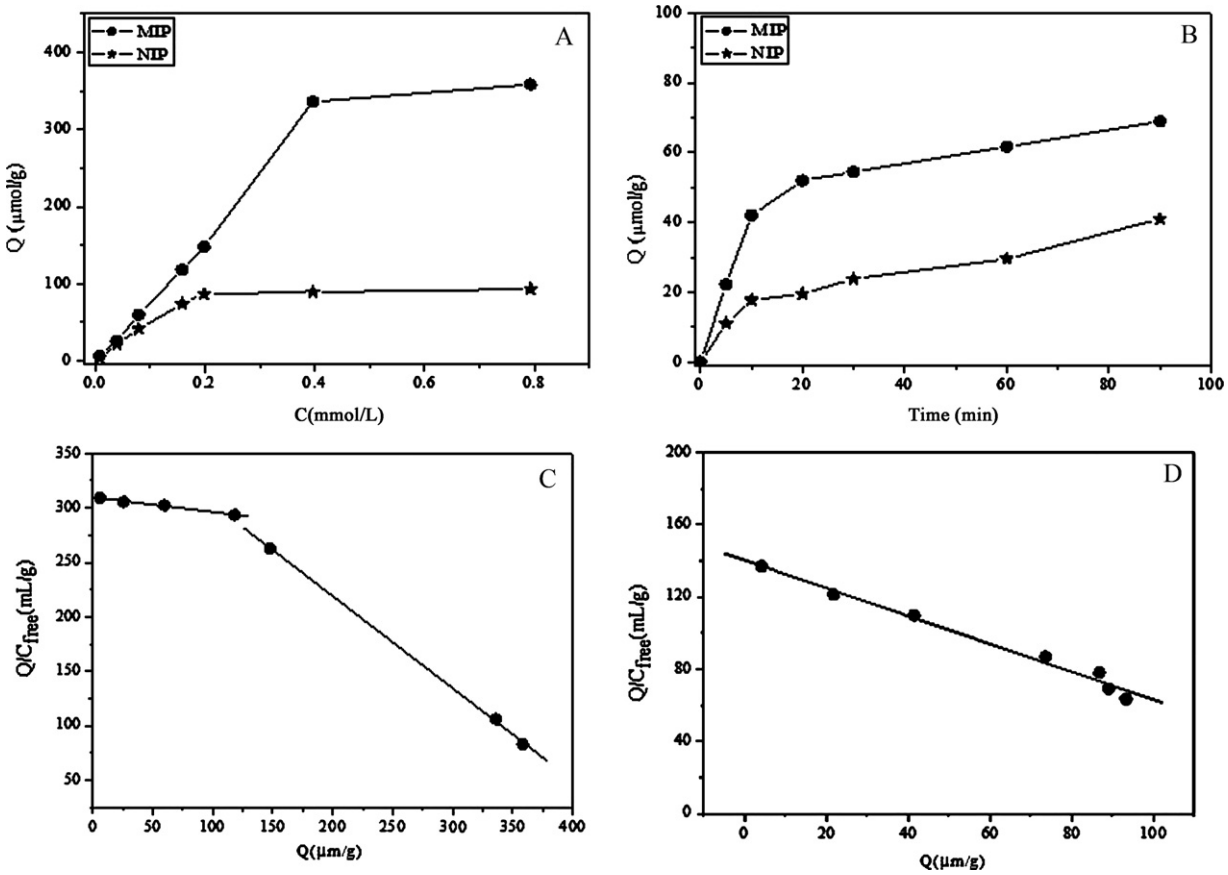

Fig. 3. Binding isotherm and Scatchard analysis of MIPs and NIPs (A: binding isotherm of MIPs and NIPs; B: dynamic adsorption of MIPs and NIPs; C: Scatchard plot of MIPs; D: Scatchard plot of NIPs).

demonstrated by dynamic adsorption, and the absorption equilibriums of MIPs and NIPs were all reached below 10 min. The Scatchard plot suggested that there were two distinct sections within the plot which can be regarded as straight lines, so it would be reasonable to assume that the binding sites can be classified into two distinct groups with specific binding properties in MIPs. For higher affinity site, the values of K_D and Q_{max} calculated from the slope and intercept of the linear portion of Scatchard analysis were 0.13 μmol L^{-1} and 0.16 mmol g^{-1}, and for lower affinity binding site were 0.84 μmol L^{-1} and 0.43 mmol g^{-1}, respectively. At the same time, only one type of binding site in NIP for MEL was observed and its K_D and Q_{max} were calculated to be 0.77 μmol L^{-1} and 0.15 mmol g^{-1}, respectively.

3.3. Optimization of MI-MSPD procedure

To achieve satisfactory recoveries of MEL in milk samples, several parameters (the ratio of sample to MIP adsorbent (S/MIP), washing and elution solvent, etc.) that affect the extraction efficiency of MI-MSPD were studied. The S/MIP ranged from 1:1 to 1:4 was evaluated and the results revealed that the ratio of 1:1 provided the best recoveries of MEL. Further increasing the proportion of adsorbent from 1:2.5 to 1:4, the recoveries reduced sharply from 91.8% to 17.0%, which possibly due to the strong absorbability of MIPs. Thereby, 1:1 was applied as the optimized S/MIP ratio in the subsequent work. Moreover, the MIPs pre-packed in the bottom of the cartridge acted as SPE adsorbent to further remove interfering matrix components and isolate analytes to perform high recoveries.

One of the outstanding advantages of MSPD is that extraction and clean-up are carried out just in a single step. Therefore, the types and volume of washing solvent are the key factors, which should be carefully selected to achieve the highest recovery for analytes while eliminating the most of interferences originated from biological matrix [19]. Different washing solvents such as methanol, acetonitrile, chloroform, water, water–methanol (1:1, v/v), water–acetonitrile (1:1, v/v), water–methanol (9:1, v/v), water–acetonitrile (9:1, v/v), and water–methanol (1:4, v/v) were investigated and the results in Fig. 4 showed that the best recovery was obtained using acetonitrile as washing solvent. For the

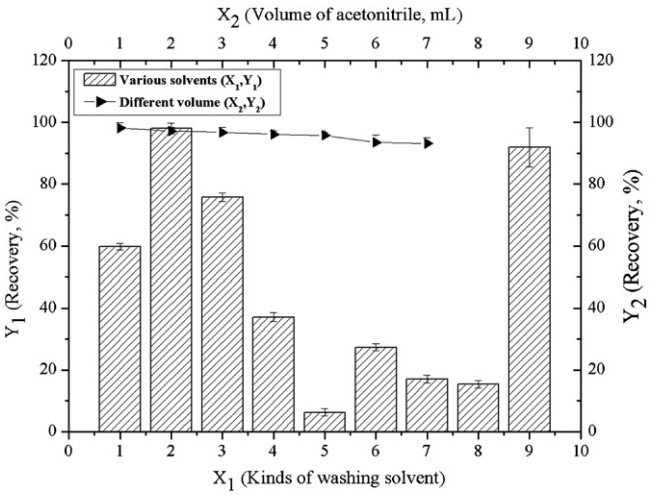

Fig. 4. Effect of washing solvents on extraction efficiency of MEL.

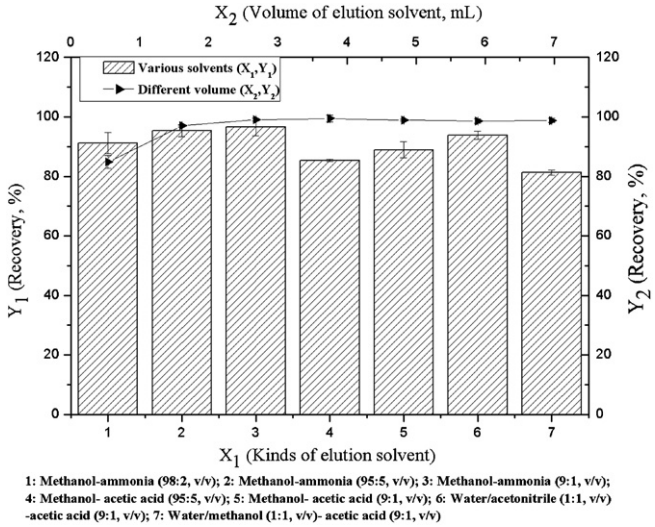

Fig. 5. Effect of elution solvents on the recovery of MEL.

purpose of the minimum volume of washing solution able to efficiently rinse the interferences, the volume of acetonitrile was ranged from 1.0 to 7.0 mL and the results showed that the recovery of MEL reduced with the increasing of acetonitrile. Considering that less solvent could not remove the interferences sufficiently, and as well as the purification efficiency and economic factors, 3.0 mL of acetonitrile was chosen as the washing solution.

On the basis of the results of washing step and the relevant references reported [17], the elution step was performed using methanol containing acetic acid and ammonia as the main eluent. The results (Fig. 5) showed that the recoveries were more than 80% for all eluents, and the highest recovery (96.6%) was obtained by methanol–ammonia (95:5, v/v) solution as eluting solvent, which due to it broken the ionic interaction between MEL with binding sites of MIPs. Different volumes (1.0, 2.0, 3.0, 4.0 5.0, 6.0, and 7.0 mL) of methanol–ammonia (95:5, v/v) were further evaluated and the results showed that the recovery of analyte increased rapidly with the increase of the elution solvent volume from 1.0 to 3.0 mL and then almost constant even further increasing the volume from 3.0 to 7.0 mL. Thereby, 3.0 mL of methanol–ammonia (95:5, v/v) was selected as elution solvent for MI-MSPD.

3.4. Comparison of MI-MSPD with conventional adsorbents-MSPD method

Different MSPD procedures using MIPs, NIPs and conventional adsorbents (PCX, MCX and SCX) as adsorbent were investigated (Fig. 6) and compared according to the previous reports [6,14]. The extracts obtained from PCX and SCX showed lower recoveries for MEL in milk (28.6% for PCX and 64.5% for SCX), which were due to their lower affinities and non-special adsorption to MEL. Although using MCX as dispersant can get higher recovery for MEL than PCX and SCX, the purification effect of washing step was unapparent. Comparatively, the highest recoveries (98.0%) with more clean extracts were obtained by using MIPs as a dispersant. The recoveries of MEL (66.0%) in NI-MSPD procedure were lower than the results of MI-MSPD. The chromatograms of MI-MSPD indicated that no interferences originated from milk matrix were observed, which demonstrated the high selectivity of the MIPs in aqueous environment.

3.5. Validation of the MI-MSPD-HPLC method

To evaluate the proposed MI-MSPD-HPLC method, the linearity, precision, accuracy, repeatability, and detection limits were investigated under the optimum condition. Calibration curves for MEL were constructed using the areas of the chromatographic peaks measured at nine increasing concentrations, in a range of 0.24–60.0 $\mu g\,g^{-1}$. Good linearity was observed throughout the concentration range, and the regression equation was $y = 4.65 \times 10^4 x + 1.30 \times 10^4$ (y: peak area of MEL; x: concentration of MEL, $\mu g\,g^{-1}$) with the correlation coefficient (r^2) = 0.9994. The precision and accuracy were determined by analyzing five replicates of the spiked samples at three concentration levels on the same day and three different days (n = 3). Intra-assay and inter-assay precision expressed as RSD were 3.8% and 4.2%, respectively. Based on the signal-to-noise ratio of 3 and 10, the LOD and LOQ for MEL was 0.05 $\mu g\,g^{-1}$ and 0.16 $\mu g\,g^{-1}$, respectively. Comparison with previously reported methods, the MI-MSPD-HPLC method significantly reduced the complicated pretreatment procedure while shortens the analysis time and reduces the consumption of toxic organic solvents [8,9,13,32,33].

3.6. Analysis of bovine milk products

The performance of MI-MSPD-HPLC method in the extraction of MEL from milk samples was investigated. Five brands of bovine milk products (fat content: 2–6% (m/v); protein content: 2–5% (m/v); carbohydrate: 2–4% (m/v); minerals: 0–13% (m/v); density: 1.023–1.030 $g\,mL^{-1}$) collected from the local markets of Baoding were pretreated under the optimized condition. The MEL contents in different milk products were in a range of 0.17–0.41 $\mu g\,g^{-1}$, which was accorded with the quality ($\leq$2.5 $mg\,kg^{-1}$) requested by China government. To investigate the effect of sample matrix on the accuracy of the MI-MSPD-HPLC method for real samples analy-

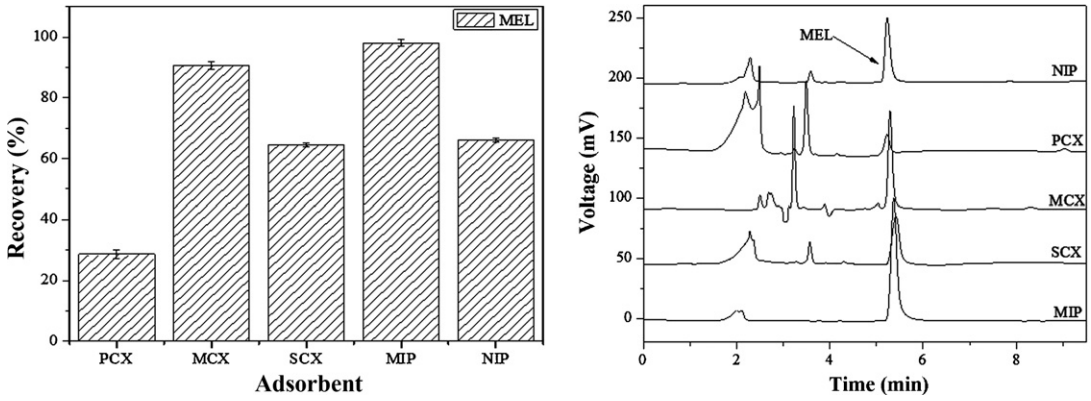

Fig. 6. Comparison of MIPs with other adsorbents.

Table 1
Recoveries of the MI-MISPD-HPLC method for spiked MEL milk samples ($n = 3$).

Spiked levels	1.5 μg g^{-1}		27.0 μg g^{-1}		60.0 μg g^{-1}	
	Recovery (%)	RSD%	Recovery (%)	RSD%	Recovery (%)	RSD%
MEL	86.0	2.8	96.2	3.2	93.7	4.0
	87.3	3.1	95.4	3.8	92.5	3.6
	90.2	3.6	92.5	2.9	90.8	2.5

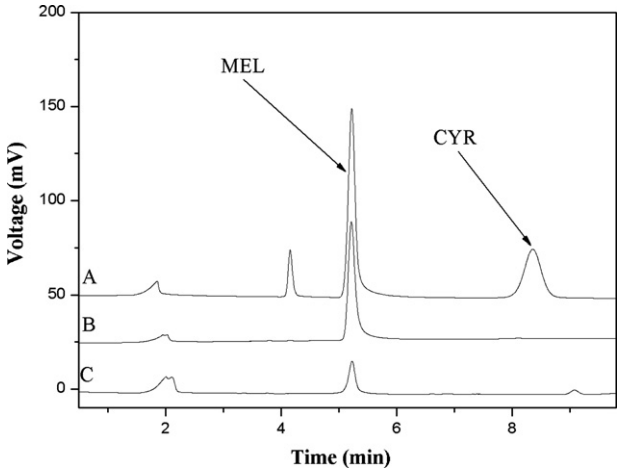

Fig. 7. Chromatograms of the milk samples (A: MEL and CYR; B: spiked sample: 2 μg g^{-1}; C: milk sample).

sis, recovery experiment was carried out by spiking three different levels of MEL into the milk samples. The results demonstrated that the MI-MSPD recovered a high recovery of MEL (86.0–96.2%) in milk with RSD ≤ 4.0% (Table 1). As seen in Fig. 7, the chromatograms of MEL in milk and spiked milk extract revealed that there was no endogenous interferences from the milk matrix, and the sample was significantly cleaner after being treated with MI-MSPD protocol, which meant the synthesized MIPs had high selectivity for analyte in a complex sample environment. Compared with the previous papers regarding the use of MIPs for the analysis of MEL, the presented method provided super purification ability and high affinity and selectivity to MEL, and it could be potentially applied for the determination of trace MEL in complicated bio-matrix samples.

4. Conclusion

A simple, convenient and high selective method (MI-MSPD-HPLC) was developed for selective purification and screening of trace level MEL in milk sample. The non-covalent MIPs synthesized by CYR as a dummy template in methanol–water systems showed high affinity, adsorption capacity, as well as remarkable impurity toward MEL in actual sample during MI-MSPD procedure. The performance characteristics, in terms of recovery, selectivity, and cleanliness of the extracts, as well as economy, indicate that the MI-MSPD-HPLC method is sufficiently accurate and precise to be used for MEL analysis in complicated bio-matrix samples.

Acknowledgements

The project was sponsored by the National Natural Science Foundation of China (20905019 and 21011140338), Natural Science Foundation of Hebei (B2010000209), and the Science Foundation of Education Department of Hebei Province (CPRC003).

References

[1] B. Wang, Y. Wang, H. Yang, J. Wang, A. Deng, Mikrochim. Acta 174 (2011) 191.
[2] Z. Zhang, M. Zhang, L. Luo, X. Yang, Y. Hu, H. Zhang, S. Yao, J. Sep. Sci. 33 (2010) 2854.
[3] B. Pushner, R.H. Poppenga, L.J. Lowenstine, M.S. Filigenzi, P.A. Pesavento, J. Vet. Diagn. Invest. 19 (2007) 616.
[4] C.G. Skinner, J.D. Thom, J.D. Osterloh, J. Med. Toxicol. 6 (2010) 50.
[5] United Nations Environment Program, OECD Screening Information Data Set (SIDS) Analysis: Melamine, 2002, available at http://www.inchem.org/pages/sids.html
[6] H. Sun, L. Wang, L. Ai, S. Liang, H. Wu, Food Control 21 (2010) 686.
[7] M. Pan, G. Fang, W. Tang, S.J. Wang, J. Chromatogr. B 878 (2010) 1531.
[8] M.S. Filigenzi, E.R. Tor, R.H. Poppenga, L.A. Aston, B. Puschner, Rapid Commun. Mass Spectrom. 21 (2007) 4027.
[9] L. He, Y. Su, Y. Zheng, X. Huang, L. Wu, Y. Liu, Z. Zeng, Z. Chen, J. Chromatogr. A 1216 (2009) 6196.
[10] H.H. Yang, W.H. Zhou, X.C. Guo, F.R. Chen, H.Q. Zhao, L.M. Lin, X.R. Wang, Talanta 80 (2009) 821.
[11] R.H. Wei, R. Wang, Q.F. Zeng, M. Chen, T.Z. Liu, J. Chromatogr. Sci. 47 (2009) 581.
[12] R.A. Yokley, L.C. Mayer, R. Rezaaiyan, M.E. Manuli, M.W. Cheung, J. Agric. Food Chem. 48 (2000) 3352.
[13] X. Li, J. Hu, H. Han, J. Sep. Sci. 34 (2011) 323.
[14] L. Meng, G. Shen, X. Hou, L. Wang, Chromatographia 70 (2009) 991.
[15] A. Philbrook, C.J. Blake, N. Dunlop, C.J. Easton, M.A. Keniry, J.S. Simpson, J. Polym. 46 (2005) 2153.
[16] D.W. Lachenmeier, E. Humpfer, F. Fang, B. Schutz, P. Dvortsak, C. Sproll, M. Spraul, J. Agric. Food Chem. 57 (2009) 7194.
[17] L. He, Y. Su, X. Shen, Y. Zheng, H. Guo, Z. Zeng, J. Sep. Sci. 32 (2009) 3310.
[18] H. Sun, L. Wang, N. Liu, F. Qiao, S. Liang, Chromatographia 70 (2009) 1685.
[19] H. Sun, F. Qiao, G. Liu, S. Liang, Anal. Chim. Acta 625 (2008) 154.
[20] J. Rubert, C. Soler, J. Mañes, Talanta 85 (2011) 206.
[21] A.L. Dawidowicz, E. Rado, J. Pharm. Biomed. Anal. 52 (2010) 79.
[22] B. Shao, H. Han, X. Tu, L. Huang, J. Chromatogr. B 850 (2007) 412.
[23] E.M. Kristenson, U.A.T. Brinkman, Trends Anal. Chem. 25 (2006) 96.
[24] D. Li, Q. Yang, Z. Wang, R. Su, X. Xu, H. Zhang, J. Sep. Sci. 34 (2011) 822.
[25] F. Qiao, H. Sun, J. Pharm. Biomed. Anal. 53 (2010) 795.
[26] S.A. Barker, J. Biochem. Biophys. Methods 70 (2007) 151.
[27] H. Yan, K.H. Row, G. Yang, Talanta 75 (2008) 227.
[28] J. Qiao, H. Yan, H. Wang, Y. Wu, P. Pan, G. Yang, Chromatographia 73 (2011) 227.
[29] H. Wang, H. Yan, M. Qiu, J. Qiao, G. Yang, Talanta 85 (2011) 2100.
[30] H. Yan, K.H. Row, Int. J. Mol. Sci. 7 (2006) 155.
[31] H. Sun, F. Qiao, G. Liu, J. Chromatogr. A 1134 (2006) 194.
[32] X. Wang, Q. Fang, S. Liu, L. Chen, J. Sep. Sci. 35 (2012) 1432.
[33] H. Miao, S. Fan, Y.N. Wu, L. Zhang, P.P. Zhou, J.G. Li, H.J. Chen, Y.F. Zhao, Biomed. Environ. Sci. 22 (2009) 87.