

Received:
2 December 2018
Revised:
14 March 2019
Accepted:
15 April 2019

Cite as: Aliya Nur Hasanah,
Driyanti Rahayu,
Rimadani Pratiwi, Tina
Rostinawati,
Sandra Megantara,
Febrina Amelia Saputri,
Khanifa Hidayati
Puspanegara. Extraction of
atenolol from spiked blood
serum using a molecularly
imprinted polymer sorbent
obtained by precipitation
polymerization.
Heliyon 5 (2019) e01533.
doi: [10.1016/j.heliyon.2019.e01533](https://doi.org/10.1016/j.heliyon.2019.e01533)



Extraction of atenolol from spiked blood serum using a molecularly imprinted polymer sorbent obtained by precipitation polymerization

Aliya Nur Hasanah*, Driyanti Rahayu, Rimadani Pratiwi, Tina Rostinawati,
Sandra Megantara, Febrina Amelia Saputri, Khanifa Hidayati Puspanegara

*Pharmaceutical Analysis and Medicinal Chemistry Department, Faculty of Pharmacy, Universitas Padjadjaran,
Jalan Raya Bandung Sumedang KM 21,5, Jatinangor, Indonesia*

* Corresponding author.

E-mail address: aliya.n.hasanah@unpad.ac.id (A.N. Hasanah).

Abstract

Atenolol (ATE) is a cardio-selective β -blocker that is used in the treatment of hypertension over extended periods. However, ATE, like propranolol, has major potential for misuse as a performance-enhancing drug in several sports. Therefore, an efficient and selective separation method is required to detect and monitor the level of ATE in the body. This paper presents a molecularly imprinted polymer with specific and selective binding to ATE using precipitation polymerization. We show that when employed in an optimized molecularly imprinted solid phase extraction (MI-SPE) protocol, recoveries of $93.65 \pm 1.29\%$ from spiked blood serum with excellent discrimination from other β -blocker drugs is possible. The methodology used in this study includes molecular modeling interaction between ATE and itaconic acid (ITA) as functional monomer, followed by determination of binding constants with spectrophotometry, synthesis of the polymer using precipitation polymerization and ending with characterization and application of polymers to extract ATE in serum. Docking analysis revealed a binding affinity between ATE and ITA of

–2.0 kcal/mol with the formation of hydrogen bonding. The association constant between ATE and ITA was studied by UV titration in two different solvents, with evidence of an association constant $6.277 \times 10^2 \text{ M}^{-1}$ measured in acetonitrile: methanol (1:1). An optimized MI-SPE protocol was developed for the extraction of ATE from spiked blood serum, obtaining recoveries of 93.65% with excellent selectivity toward other β -blocker drugs.

Keywords: Analytical chemistry, Materials chemistry, Pharmaceutical chemistry

1. Introduction

β -Blockers are an essential class of cardiovascular medications for reducing morbidity and mortality in patients with heart failure [1]. Atenolol (ATE) is part of a class of β -adrenoceptor antagonists often called β -blockers. β -blockers are competitive inhibitors and interfere with the action of hormone stimulation in β -adrenergic receptors in the nervous system [2]. ATE is reported to reduce intrinsic sympathomimetic activity. It is used for the treatment of hypertension, angina pectoris, cardiac arrhythmias, and myocardial infarction [3]. These drugs are often abused by athletes to lower the heart rate and reduce hand tremor and anxiety [4]. The World Anti-Doping Agency (WADA) prohibits the use of ATEs especially for use by athletes in some sports [5]. Therefore, monitoring of ATE levels is required to detect drug abuse, particularly in athletes, as well as for proper dose regulation for patients. Several methods have been developed to analyze ATE in plasma and urine using molecular imprinted solid phase extraction (MI-SPE) as sample preparation before analysis [6, 7, 8]. Molecular imprinting polymer (MIP) is a technique that was developed to make a polymer with specific molecular recognition to the specified compound [9, 10, 11, 12, 13]. In the synthesis of MIP, the selection of functional monomers is an important step that can affect the selectivity and performance of sorbent. Itaconic acid (ITA) is one of the polymer monomers that is widely used in this process [14, 15, 16, 17, 18, 19, 20]. According to the US Department of Energy (DOE), ITA is one of the 12 important chemical building blocks of sugar, has a group $\text{C}=\text{C}$ and two carboxyl groups, which can be precursors to various chemical transformations and can be polymerized through the formation of ester bonds and free radical polymerization [21]. MIP sorbent synthesis in this study was carried out using the precipitation polymerization method; this method is based on mixing polymer solutions consisting of functional monomers, templates, and cross-linkers with porogens. The resulting sorbent has a more homogeneous physical size, so no grinding is required and the resulting polymer generally has a better adsorption profile than the bulk polymerization method [22]. All development of MI-SPE ATE that other researcher already done [6, 7, 8], none of them using precipitation polymerization and the result of recoveries are below 80%.

According to other researcher results, our laboratory has previously made MI-SPE for ATE [23] using methacrylic acid as a functional monomer with recoveries above 90%, but there is a lack of imprinting factor compared to the non-molecular imprinted polymer (NIP). Therefore, in this study, we wish to report on the development of MI-SPE materials for the rapid pre-concentration and clean-up of ATE in biological fluid samples before HPLC analysis using ITA as the other functional monomer. So far, ITA has never been used as a monomer for ATE-MIP. The synthesis of MI-SPE ATE begins with molecular modeling interaction between ATE and ITA, continuing with the determination of binding constants using spectrophotometry and ending with the characterization and application of polymers to extract ATE in blood serum samples.

2. Materials & methods

2.1. Materials

(R)-(+)-atenolol (ATE), metoprolol tartrate (MET) and propranolol hydrochloride (PRO) were purchased from Tokyo Chemical Industry. ITA, ethylene glycol dimethacrylate (EGDMA), benzoyl peroxide (BPO), triethylamine (TEA) and trifluoroacetic acid (TFA) were purchased from Sigma Aldrich. HPLC grade acetonitrile and methanol were purchased from Fischer Scientific. Acetone, potassium bromide and acetic acid were purchased from Merck. Blood samples were provided by the Indonesian Red Cross. If not otherwise specified, all chemicals are analytical grade. The morphological evaluation analysis was carried out by JSM-6610LV JEOL Ltd. A UV-visible spectrophotometer (Analytic Jena, Specord 200) was used to detect the UV absorbance for constant association determination. Analyses of blood after extraction with the MI-SPE were performed using HPLC (Dionex Ultimate 3000 with UV detector) by isocratic elution, using a mixture of methanol/water containing 0.01% TEA (15:85) as the mobile phase and a column Merck C18G 125A (250 × 4.6 mm i.d.). The flow rate was 1 mL min⁻¹, and the detection wavelength was set at 229 nm. The column was passed through with mobile phase until the baseline was obtained before detecting the samples.

2.2. Methods

2.2.1. Analysis of the interactions in the pre-polymerization complex

In order to describe the interaction and hydrogen bonding between the template and monomers (ITA), computational predictions were carried out. Two-dimensional structures of ATE and ITA were drawn and then converted into three dimensions using the ChemBio3D Ultra 12.0 program. Geometry optimization was then carried out with the *ab initio* method (Hartree Fock, base set 3-21G) using the Games

Interface on the ChemBio3D Ultra 12.0. The docking process between ATE and ITA was undertaken using the PyRx software – Virtual Screening Tools with AutoDock Vina. The docking results were analyzed by comparing the position and type of bond formed and the value of the binding affinity of the template and each monomer using the AutoDockTools 1.5 software.

2.2.2. Determination of the association constant for the template–functional monomer with UV-Visible spectrophotometry

Monomer-template interaction was studied before polymer synthesis using UV titration. To a solution of ATE 0.001 mol/L in methanol or methanol: acetonitrile 1:1, an increasing amount of ITA was added until 10-fold excess was reached. Subsequently, absorbance was measured. Finally, a curve of the delta absorbance against the monomer concentration was constructed to determine the value of the association constant [24].

2.2.3. Preparation of the MIP and NIP

MIP and NIP were synthesized using the precipitation polymerization method, as follows. MIP was obtained by dissolving ATE (1 mmol) as a template and ITA as functional monomer (4 mmol) in 350 mL solvent in a closed vial and then sonicated for 5 min. Subsequently, EGDMA (20 mmol) was added to the solution as cross-linker then sonicated for 40 min. Then benzoyl peroxide (0.206 mmol) was added to the vial as initiator, and finally, the vial was placed in a water bath shaker at 70 °C for 24 h. The polymer thus formed was washed using methanol and water. After washing, the polymer was dried in an oven at 60 °C for 18 h. NIP was synthesized in the same way, but without the addition of templates to verify MIP results [24]. The composition of synthesized MIP and NIP ATE is presented in Table 1. The Soxhlet apparatus was used for template removal from the synthesized MIP using methanol

Table 1. The composition of MIP and NIP Atenolol.

Polymer	Functional monomer (1 mmol)	Porogen (350 mL)	Crosslinker (20 mmol)	Template (1 mmol)	Method
MIP 1	Itaconic acid	Methanol: Acetonitrile	EGDMA	Atenolol	Precipitation polymerization
NIP 1	Itaconic acid	Methanol: Acetonitrile	EGDMA	-	Precipitation polymerization
MIP 2	Itaconic acid	Methanol	EGDMA	Atenolol	Precipitation polymerization
NIP 2	Itaconic acid	Methanol	EGDMA	-	Precipitation polymerization

and acetic acid (9:1) for 24 hours. The extraction process is complete when no atenolol peaks appeared while monitored using HPLC.

2.2.4. Adsorption capacity evaluation

To evaluate the adsorption capacity, we varied the concentration of the ATE solution, i.e., 1, 2.5, 5, 7.5 and 10 mg L⁻¹. A 5 mL ATE solution was introduced into a vial containing 20 mg of MIP sorbent, then shaken using a shaker at 120 rpm for 3 h at room temperature. Next, the mixture was filtered, and the absorbance of the filtrate was measured using a UV spectrophotometer. NIP sorbents were treated in the same way as MIP. The evaluation results of MIP-SPE adsorption capacity was plotted on the Freundlich isotherm adsorption curve [24, 25].

2.2.5. Solid phase extraction

200 mg of imprinted or non-imprinted polymer particles were dry packed in 3 mL SPE cartridges using 20 µm porous polyethylene frits. Blood serum samples were prepared by centrifugation of the collected blood (from the Indonesian Red Cross) at 8,000 rpm for 5 min at 14 °C and careful collection of the transparent top layer. This was spiked with 2 mg L⁻¹ of ATE in water. Drawing on Hasanah *et al.*'s (2017) optimization results, we followed all the SPE conditions except for eluting the solvent. The final extraction protocol consisted of an initial conditioning step with 1 mL of methanol, loading 1 mL of the spiked blood sample, followed by a 1 mL acetonitrile wash and a final elution with 1 mL of 0.01% TFA: methanol (1:99). Full vacuum was applied to the cartridges between each step. To test the specificity of the polymers, an equimolar mixture of ATE, MET and PRO (1 mg L⁻¹ each) in water was spiked into the blood serum samples and applied to the SPE cartridges. The collected fractions were analyzed by HPLC using the method described above. The HPLC method for ATE analysis has previously been validated according to the ICH guideline for bioanalysis. Using these conditions, the limit of detection (LOD) and limit of quantitation (LOQ) for ATE were 0.1 and 0.4 mg mL⁻¹ respectively. Calibration curve equation of analyte were $y = 0,0063x + 0,0083$, $R^2 = 0,9995$.

2.2.6. Physical characterization of molecularly imprinted polymer

We used Fourier Transform Infra-Red (FTIR) and a scanning electron microscope (SEM) to characterize the physical properties of MIP. A total of 2 mg of MIP sorbent was crushed together with 200 mg of potassium bromide (KBr), then molded into pellets. The infrared spectrum of MIP sorbents was observed using FTIR instruments. The transmission was measured at wave numbers 4000–400 cm⁻¹. MIP sorbent functional groups were determined after extraction [25]. The surface

morphologies of the polymers were observed using SEM by placing MIP and NIP on silicon and then putting them in the SEM instrument. NIP sorbents were characterized in the same way using FTIR and SEM.

3. Results and discussion

3.1. Analysis of the interactions in the pre-polymerization complex

Based on interaction analysis in the prepolymerization complex (Fig. 1a), the binding affinity between ATE and ITA is -2.0 kcal/mol. Also, there is a hydrogen bond formed between the hydroxyl of ITA and the amine of ATE without $\pi-\pi$ interaction. The theoretical computations showed that ITA forms stable complexes with negative binding affinity. The imprinting effect of the MIP greatly influenced the interaction between the template and functional monomer that we synthesized [26]. The hydrogen bonding formed between ITA and ATE is a signal that their imprinting properties will be excellent.

We also performed interaction analysis in the pre-polymerization complex between methacrylic acid (MAA) and ITA (Fig. 1b) and found out that the binding affinity between MAA-ATE is -1.5 kcal/mol. This affinity is lower than ATE-ITA and has a good correlation with our previous result [23] that having lower recovery percentage than ATE-ITA.

3.2. Association constant determination for the functional monomer–template using UV-Visible spectrophotometry

The template molecule used was ATE. The monomer selected to form a non-covalent complex with ATE was ITA. In general, the better and stronger the interaction, the better the imprinting effect and the more stable the complex during polymerization [27]. Therefore, the selected monomers should be tested for their interaction with template molecules by non-covalent impedimetric stoichiometric study methods [28], such that interactions are determined based on the value of the association constants (K_a) obtained. For this, UV titration was performed in

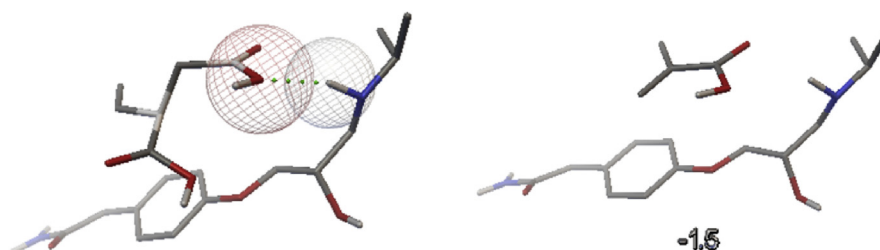


Fig. 1. Result of analysis of the interactions in the prepolymerization complex (a) ITA-ATE (b) MAA-ATE.

ATE and ITA solution. Increasing amounts of ITA were added to a solution of ATE (0.001 mol L^{-1}) in methanol and solvent mixture (methanol: acetonitrile 1:1) until at least a 10-fold excess was reached. At this point, delta absorbance was recorded, and the results plotted on the curve. The association constant was calculated based on the slope value and the graph intercept using the Benassi–Hildebrand equation [29].

Based on the association constant determined (Table 2), the ITA monomer in the methanol: acetonitrile (1:1) solvent yields the most significant association constant value with the ATE template, with a value of 6.277×10^2 . The K_a value determines the selectivity and specificity of the polymers produced [30].

The interaction between ATE and ITA is predicted to be a hydrogen interaction between the carboxylic group of ITA and the amine group of ATE as, previously predicted by analysis of the interactions in the pre-polymerization complex (Fig. 1a). Tadi confirmed the estimated interaction and Motghare (2013) [31], whose study showed the carbonyl group on the carboxylate acting as the hydrogen bond acceptor binding to the amine (-NH) as a hydrogen bond donor in ATE.

3.3. Molecular imprinted polymer synthesis

The comparison used for this synthesis was the template: monomer: crosslinker (1:4:20). The monomer must have a good affinity for the template. Sadaghi and Mo-frad (2007) and Sadeghi *et al.* (2012) [32, 33] demonstrated that the use of template monomers (1:4) provides an excellent specific affinity and high recovery of template compounds compared with NIP sorbents. The use of functional monomers with excess concentrations may lead to increased non-specific affinity in the sorbent polymer produced [34]. Based on constant association value, the monomer with the best interaction was ITA in a solvent mixture (acetonitrile: methanol 1:1). The possible interactions with the synthesized imprinting polymers of ITA monomers are presented in Fig. 2.

3.4. Adsorption capacity

The adsorption capacity was determined by an adsorption isotherm model that provides an overview of the mechanism of the interaction of ATE adsorption on the sorbent surface and sorbent characteristics by calculating the relationship of bonding parameters and the affinity distribution of the bond. In this study, the analysis used the Freundlich isotherm model. This model assumes that the tested sorbents

Table 2. Association constant results of ITA — atenolol on different solvent.

Monomer	Solvent	Template	$K_a \text{ (M}^{-1}\text{)}$
Itaconic acid	Methanol: acetonitrile	Atenolol	6.277×10^2
Itaconic acid	Methanol	Atenolol	5.43×10^2

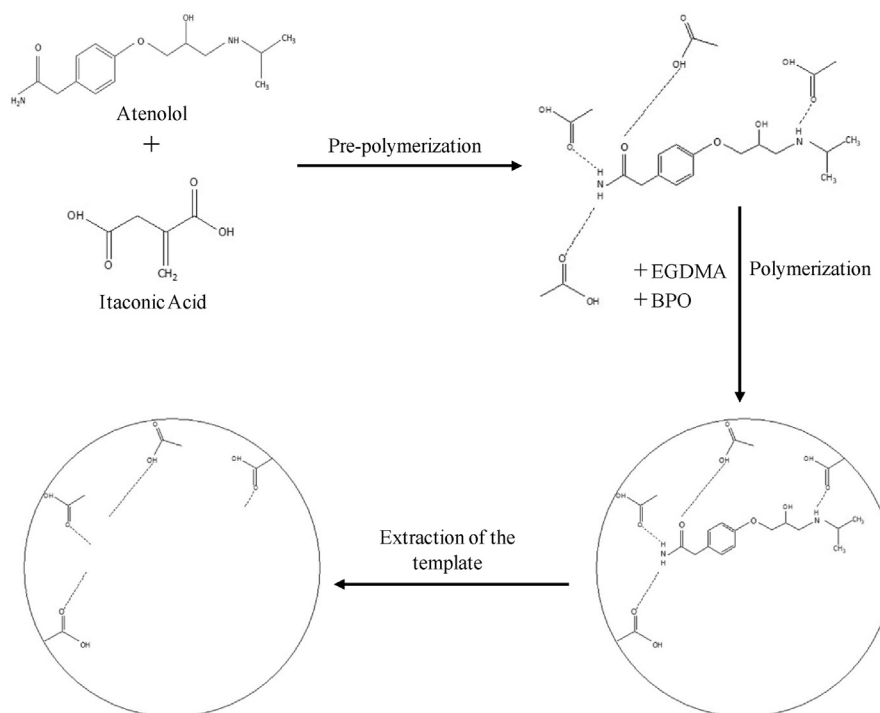


Fig. 2. Schematic diagram of MI-SPE ATE synthesis.

have various surfaces so that the bond affinity is also different in terms of the activated carbon, silica, metal and polymers, especially in imprinted polymers with non-covalent interaction approaches [35]. In this isotherm model, the heterogeneity of the sorbent is calculated exponentially as a log function, which facilitates the calculation by converting it into a linear function [36]. The Freundlich isotherm function uses two test parameters, a and m . These parameters sequentially indicate the affinity of the polymer binding and the degree of binding heterogeneity, having a value between 0 and 1. An m value equal to one indicates the sorbent bonding system is homogeneous, i.e. the surface of the polymer particles has the same binding ability, whereas a near-zero m value indicates a heterogeneous binding system [37].

Based on the significant degree of polymer affinity binding (a) shown in Table 3, MIP 1 produces a significant difference between MIP and NIP compared to MIP 2, so this polymer was then selected to be packed into an SPE cartridge and optimized for use as an alternative to the separation of ATE from biological samples.

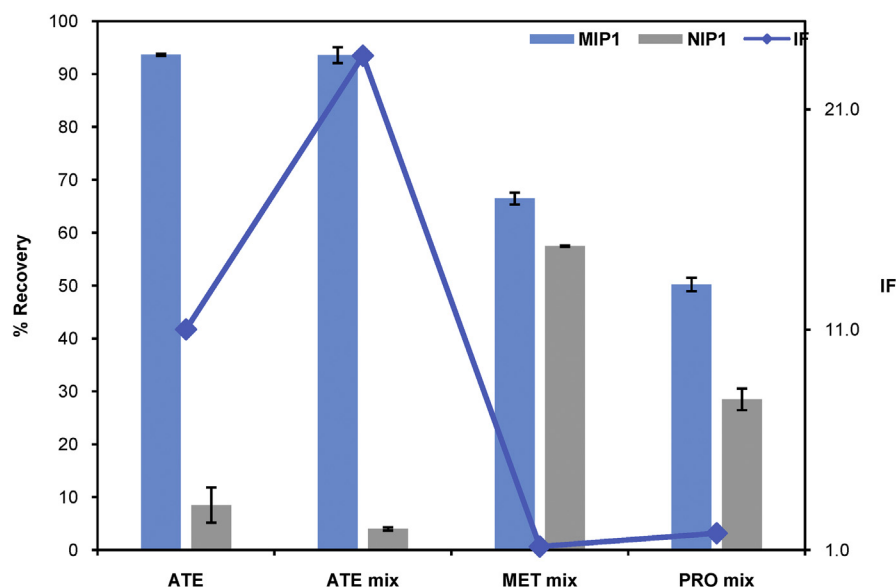
3.5. Solid phase extraction

Strong binding of ITA and ATE on pre-polymerization solution was demonstrated based on the K_a value after the polymer was synthesized. We used solid phase extraction as the tool for drug extractions. Loading of blood serum spiked with 2 mg L⁻¹ of ATE and then washed with 1 mL acetonitrile showed excellent recovery

Table 3. Adsorption isotherm linear regression Freundlich model result.

Polymer	m	a (mg/g)
MIP 1	3.039	4.250
NIP 1	0.209	0.164
MIP 2	0.603	0.269
NIP 2	0.881	0.062

and promoted specific interaction. The selectivity for ATE of MIP was investigated by determining its binding ability compared with other β -blocker drugs such as MET and PRO. The recovery of ATE up to $93.65 \pm 1.29\%$ was achieved using MIP 1, while recoveries for the competing analytes MET and PRO were $66.49\% \pm 0.88$ and $50.23\% \pm 20.62$ respectively. As seen in Fig. 3, the difference in the recoveries between MIP 1 and NIP 1 is very large for ATE. The imprinting factor (IF) of this polymer was 11.02 and increased when applied to the sample spiked with mixed β -blockers (23.43). Our previous result from MAA monomer has recoveries $92.20 \pm 1.36\%$ with IF value 1.86, showed lower imprinting efficiency of this MIP compared to our new MIP from ITA monomer. The results show that the polymer can differentiate a molecule in the same class and remember ATE molecularly. The chromatogram of the blank serum and spiked serum (with and without pre-treatment with MIP 1 ATE) is shown in Fig. 4. Previous development of ATE MI-SPE [8] only have recoveries 74.5–75.2% with IF value 4.18 this result is below our result and proved that our MI-SPE potential to be used as extraction tools of ATE from serum.

**Fig. 3.** Recoveries and imprinting factor value of blood serum spiked with ATE alone and mix of ATE with other β blockers.

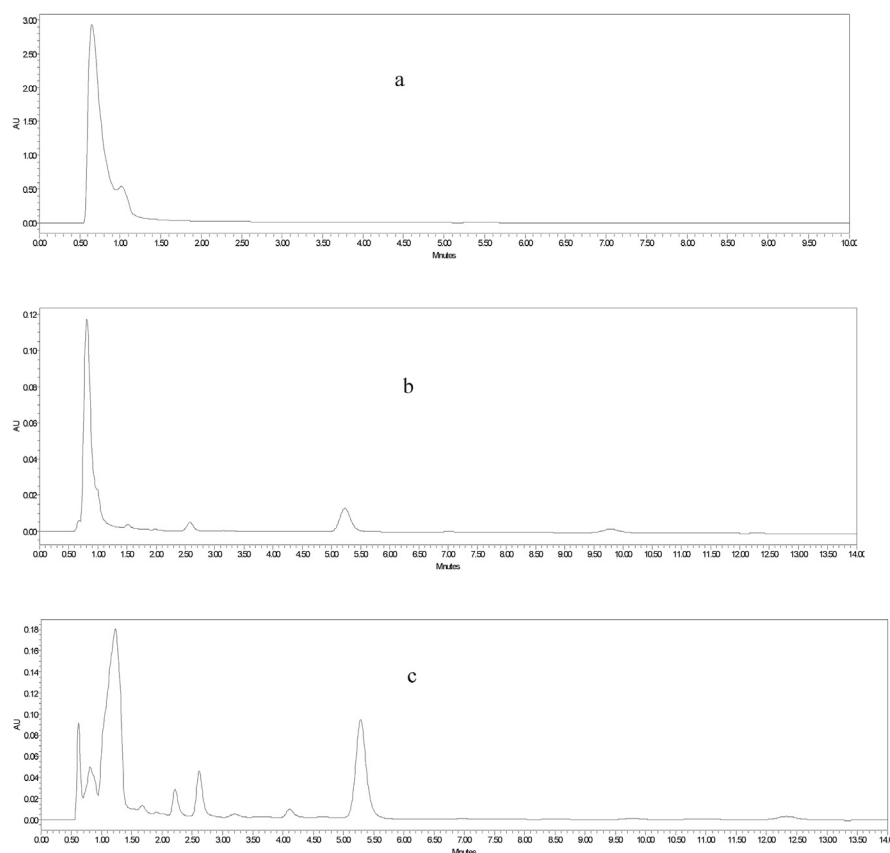


Fig. 4. Chromatogram of (a) blank sample serum, (b) spiked sample serum without MIP 1 pre-treatment, (c) spiked sample serum with MIP 1 pre-treatment.

3.6. Physical characterization of molecularly imprinted polymer

The FTIR analysis on sorbents depicted in Fig. 5 shows that MIP 1 has a spectrum that is almost identical to the NIP 1 spectrum, indicating that ATE has been extracted from the MIP matrix. Sequential C-O-C asymmetric stretching, C-H asymmetric deformation and C = O asymmetric stretching in the wave number range 1155 cm^{-1} , 1466 cm^{-1} and 1733 cm^{-1} indicating the existence of the EGDMA structure shows successful synthesis [14].

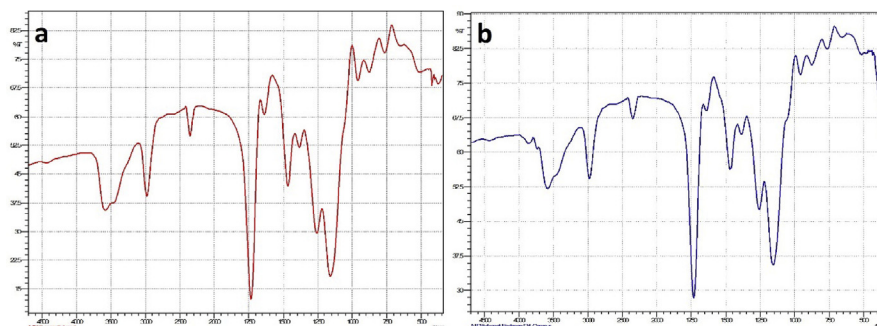


Fig. 5. FTIR spectra of MIP 1 (a) and NIP 1 (b).

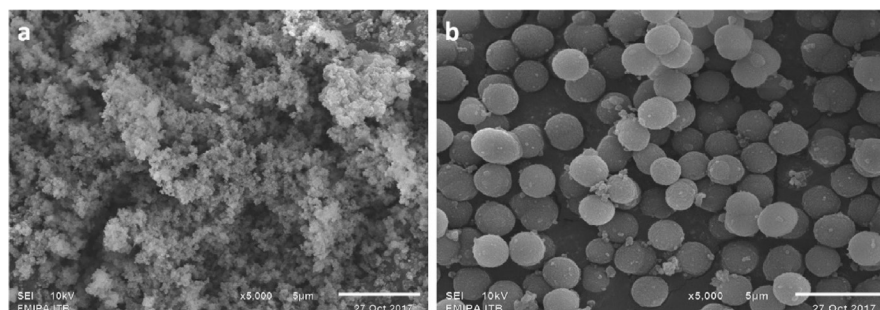


Fig. 6. SEM of MIP 1 (a) and NIP 1 (b).

After determining the analytical performance properties of MIP 1, we conducted a physical characterization of this polymer. The results of the physical characterization using SEM (Fig. 6) show that the sorbents synthesized using the precipitation polymerization method have a homogeneous particle size. It is also apparent that MIP 1 sorbents have smaller particle sizes with higher porosity compared to sorbent NIP 1, which tends to form large aggregates, influenced by the presence of a template on MIP that inhibits agglomeration in sorbent particle formation [38]. Based on BET analysis, surface area of MIP 1 and NIP 1 is 80,804 m²/g and 58,561 m²/g respectively. The high porosity level in MIP 1 shows that this polymer forms a cavity or recognition side of the target molecule, with a high porosity profile, allowing a greater adsorption area to provide better adsorption capability for ATE [14].

4. Conclusion

Our study shows that using ITA as a functional monomer in mixed porogen (methanol: acetonitrile) employing precipitation polymerization has an excellent selectivity toward ATE compared to other β -blockers when used as a sorbent in the extraction of ATE from spiked blood serum. We achieved exceptional sample clean-up and ATE recoveries of up to 93.65%. The recoveries for the competing analytes MET and PRO were 66.49% and 50.23%, respectively.

Declarations

Author contribution statement

Aliya Nur Hasanah: Analyzed and interpreted the data; Wrote the paper.

Driyanti Rahayu, Rimadani Pratiwi, Tina Rostinawati, Sandra Megantara, Febrina Amelia Saputri, Khanifa Hidayati Puspanegara: Conceived and designed the experiments; Performed the experiments.

Funding statement

This work was supported by the Ministry of Research and Higher National Education of Indonesia through the Penelitian Terapan Unggulan Perguruan Tinggi (PTUPT) research scheme.

Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

References

- [1] J.J. DiNicolantonio, H. Fares, A.K. Niazi, S. Chatterjee, F. D'Ascenzo, E. Cerrato, G. Biondi-Zoccai, C.J. Lavie, D.S. Bell, J.H. O'Keefe, β -Blockers in hypertension, diabetes, heart failure and acute myocardial infarction: a review of the literature, *Open Heart* 2 (2015), e000230.
- [2] N.R. Pai, S.S. Patil, Synthesis of atenolol impurities, *J. Chem. Pharm. Res.* 4 (2012) 375–382.
- [3] M.H. Iha, A.S. Martinez, P.S. Bonato, Enantioselective analysis of atenolol in biologic fluids: comparison of liquid–liquid and solid-phase extraction methods, *J. Chromatogr. B Anal. Technol. Biomed. Life Sci.* 767 (2002) 1–9.
- [4] C. Reardon, S. Creado, Drug abuse in athletes, *Subst. Abuse Rehabil.* 95 (2014).
- [5] J. Yu, X. Hu, R. Song, S. Xi, Molecularly imprinted polymer microspheres prepared by precipitation polymerization for atenolol recognition, 2010 Int. Conf. Adv. Mater. Manuf. Process 148–149 (2011) 1192–1198. ICAMMP 2010.
- [6] B.-C. Iacob, E. Bodoki, N. Gariano, L. Gliga, A. Bodoki, D. Spivak, S. Oprean, Improved enantioselectivity for atenolol employing pivot based molecular imprinting, *Molecules* 23 (2018) 1875.
- [7] T. Alizadeh, Synthesis of a nano-sized chiral imprinted polymer and its use as an (S)-atenolol carrier in the bulk liquid membrane, *J. Sep. Sci.* 37 (2014) 1887–1895.

- [8] Y. Gorbani, H. Yılmaz, H. Basan, Spectrofluorimetric determination of atenolol from human urine using high-affinity molecularly imprinted solid-phase extraction sorbent, *Luminescence* 32 (2017) 1391–1397.
- [9] H. Yan, H.R. Kyung, Characteristic and synthetic approach of molecularly imprinted polymer, *Int. J. Mol. Sci.* 7 (2006) 155–178.
- [10] W. Ji, R. Sun, W. Duan, X. Wang, T. Wang, Y. Mu, L. Guo, Selective solid phase extraction of chloroacetamide herbicides from environmental water samples by amphiphilic magnetic molecularly imprinted polymers, *Talanta* 170 (2017) 111–118.
- [11] M. Demirkurt, Y.A. Olcer, M.M. Demir, A.E. Eroglu, Electrospun polystyrene fibers knitted around imprinted acrylate microspheres as sorbent for paraben derivatives, *Anal. Chim. Acta* 1014 (2018) 1–9.
- [12] M. Arabi, M. Ghaedi, A. Ostovan, Development of a lower toxic approach based on green synthesis of water-compatible molecularly imprinted nanoparticles for the extraction of hydrochlorothiazide from human urine, *ACS Sustain. Chem. Eng.* 5 (2017) 3775–3785.
- [13] A. Ostovan, M. Ghaedi, M. Arabi, Q. Yang, J. Li, L. Chen, Hydrophilic multi-template molecularly imprinted biopolymers based on a green synthesis strategy for determination of B-family vitamins, *ACS Appl. Mater. Interfaces* 10 (2018) 4140–4150.
- [14] W. Zhang, X. She, L. Wang, H. Fan, Q. Zhou, X. Huang, J.Z. Tang, Preparation, characterization and application of a molecularly imprinted polymer for selective recognition of sulpiride, *Materials (Basel)* 10 (2017) 475.
- [15] G. Marcelo, I.C. Ferreira, R. Viveiros, T. Casimiro, Development of itaconic acid-based molecular imprinted polymers using supercritical fluid technology for pH-triggered drug delivery, *Int. J. Pharm.* 542 (2018) 125–131.
- [16] F. Bates, M. Busato, E. Piletska, M.J. Whitcombe, K. Karim, A. Guerreiro, M. del Valle, A. Giorgetti, S. Piletsky, Computational design of molecularly imprinted polymer for direct detection of melamine in milk, *Sep. Sci. Technol.* 52 (2017) 1441–1453.
- [17] A.R. Bagheri, M. Arabi, M. Ghaedi, A. Ostovan, X. Wang, J. Li, L. Chen, Dummy molecularly imprinted polymers based on a green synthesis strategy for magnetic solid-phase extraction of acrylamide in food samples, *Talanta* 195 (2019) 390–400.
- [18] A. Ostovan, M. Ghaedi, M. Arabi, Fabrication of water-compatible superparamagnetic molecularly imprinted biopolymer for clean separation of baclofen

- from bio-fluid samples: a mild and green approach, *Talanta* 179 (2018) 760–768.
- [19] A. Ostovan, M. Ghaedi, M. Arabi, A. Asfaram, Hollow porous molecularly imprinted polymer for highly selective clean-up followed by influential pre-concentration of ultra-trace glibenclamide from bio-fluid, *J. Chromatogr. A* 1520 (2017) 65–74.
- [20] M. Arabi, A. Ostovan, M. Ghaedi, M.K. Purkait, Novel strategy for synthesis of magnetic dummy molecularly imprinted nanoparticles based on functionalized silica as an efficient sorbent for the determination of acrylamide in potato chips: optimization by experimental design methodology, *Talanta* 154 (2016) 526–532.
- [21] S. Bednarz, K. Półciwarteck, J. Wityk, B. Strachota, J. Kredatusová, H. Beneš, A. Wesołowska-Piętak, G. Kowalski, Polymerization-crosslinking of renewable itaconic acid in water and in deep eutectic solvents, *Eur. Polym. J.* 95 (2017) 241–254.
- [22] C. Cacho, E. Turiel, A. Martin-Esteban, C. Pérez-Conde, C. Cámara, Characterisation and quality assessment of binding sites on a propazine-imprinted polymer prepared by precipitation polymerisation, *J. Chromatogr. B Anal. Technol. Biomed. Life Sci.* 802 (2004) 347–353.
- [23] A.N. Hasanah, S. Suryana, Mutakin, D. Rahayu, Evaluation performance of molecularly imprinted polymer prepared by two different polymerization method for atenolol recognition in human plasma, *Asian J. Chem.* 29 (2017).
- [24] A.N. Hasanah, R.E. Kartasasmita, S. Ibrahim, Synthesis and application of glibenclamide imprinted polymer for solid phase extraction in serum samples using itaconic acid as functional monomer, *J. Appl. Sci.* 15 (2015) 1288–1296.
- [25] A.N. Hasanah, T.N. Sari, N. Wijaya, R.E. Kartasasmita, S. Ibrahim, Study of the binding ability of molecular imprinted solid phase extraction for glibenclamide by optimizing template: monomer: crosslinker ratio, *Int. J. Chem. Sci.* 12 (2014) 863–870.
- [26] X. Song, J. Li, S. Xu, R. Ying, J. Ma, C. Liao, D. Liu, J. Yu, L. Chen, Determination of 16 polycyclic aromatic hydrocarbons in seawater using molecularly imprinted solid-phase extraction coupled with gas chromatography-mass spectrometry, *Talanta* 99 (2012) 75–82.
- [27] S.N.N.S. Hashim, R.I. Boysen, L.J. Schwarz, B. Danylec, M.T.W. Hearn, A comparison of covalent and non-covalent imprinting strategies for the synthesis of stigmasterol imprinted polymers, *J. Chromatogr. A* 1359 (2014) 35–43.

- [28] G. Wulff, K. Knorr, Stoichiometric noncovalent interaction in molecular imprinting, *Bioseparation* 10 (2001) 257–276.
- [29] P. Thordarson, Determining association constants from titration experiments in supramolecular chemistry, *Chem. Soc. Rev.* 40 (2011) 1305–1323.
- [30] L. Chen, S. Xu, J. Li, Recent advances in molecular imprinting technology: current status, challenges and highlighted applications, *Chem. Soc. Rev.* 40 (2011) 2922–2942.
- [31] K.K. Tadi, R.V. Motghare, Rational synthesis of pindolol imprinted polymer by non-covalent protocol based on computational approach, *J. Mol. Model.* 19 (2013) 3385–3396.
- [32] S. Sadeghi, A.A. Mofrad, Synthesis of a new ion imprinted polymer material for separation and preconcentration of traces of uranyl ions, *React. Funct. Polym.* 67 (2007) 966–976.
- [33] S. Sadeghi, A. Motaharian, A.Z. Moghaddam, Electroanalytical determination of sulfasalazine in pharmaceutical and biological samples using molecularly imprinted polymer modified carbon paste electrode, *Sensors Actuators B Chem.* 168 (2012) 336–344.
- [34] S. Sadeghi, M. Jahani, Selective solid-phase extraction using molecular imprinted polymer sorbent for the analysis of Florfenicol in food samples, *Food Chem.* 141 (2013) 1242–1251.
- [35] R.J. Umpleby, S.C. Baxter, M. Bode, J.K. Berch, R.N. Shah, K.D. Shimizu, Application of the Freundlich adsorption isotherm in the characterization of molecularly imprinted polymers, *Anal. Chim. Acta* 435 (2001) 35–42.
- [36] A. Lagha, N. Adhoum, L. Monser, A molecularly imprinted polymer for the selective solid-phase extraction of ibuprofen from urine samples, *Open Chem. Biomed. Methods J.* 4 (2011) 7–13.
- [37] R. Lahsini, M.R. Louhaichi, N. Adhoum, L. Monser, Preparation and application of a molecularly imprinted polymer for determination of glibenclamide residues, *Acta Pharm.* 63 (2013) 265–275.
- [38] T. Renkecz, K. László, V. Horváth, Molecularly imprinted microspheres prepared by precipitation polymerization at high monomer concentrations, *Mol. Imprinting* 2 (2014) 1–17.