ARTICLE IN PRESS

Talanta ■ (■■■■) ■■■–■■■

Contents lists available at ScienceDirect

Talanta

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

Separation of octopamine racemate on (*R*,*S*)-2-amino-1-phenylethanol imprinted polymer – Experimental and computational studies

Monika Sobiech, Teresa Żołek, Piotr Luliński, Dorota Maciejewska*

Department of Organic Chemistry, Faculty of Pharmacy, Medical University of Warsaw, Banacha 1, 02-097 Warsaw, Poland

ARTICLE INFO

Article history:
Received 5 February 2015
Received in revised form
22 May 2015
Accepted 26 May 2015

Keywords:
2-Amino-1-phenylethanol
Molecularly imprinted polymers
Molecularly imprinted solid phase extraction
Molecular modeling
Octopamine

ABSTRACT

Ten molecularly imprinted polymers coded as MIP**1**–MIP**10** were prepared by the radical bulk polymerization using (*R*,*S*)-(±)-2-amino-1-phenylethanol as the structural analog of the target analyte (*R*,*S*)-octopamine. The functional monomers, 4-vinylbenzoic acid (**1**), methacrylic acid (**2**), acrylic acid (**3**), trifluoromethacrylic acid (**4**), itaconic acid (**5**), acrylamide (**6**), isopropenylbenzene (**7**), 2-hydroxyethyl methacrylate (**8**), 2-(diethylamino)ethyl methacrylate (**9**), allylamine (**10**) were polymerized consecutively with the ethylene glycol dimethacrylate cross-linker in methanol as the porogen. On the basis of the binding capacity of (*R*,*S*)-octopamine MIP**1** with affinity factor equal to 6.37 was selected for further analysis. The affinity of polymer matrix MIP**1** was tested by the non-competitive binding experiments of eight structurally related analytes. Finally, molecularly imprinted solid phase extraction (MISPE) of (*R*,*S*)-octopamine from spiked human serum albumin was carried out in order to verify the applicability of novel sorbent. The molecular modeling was employed to rationalize the stereodifferentiation of the analytes by the stereospecific sites formed in the polymer matrix.

© 2015 Elsevier B.V. All rights reserved.

1. Introduction

4-(2-Amino-1-hydroxyethyl)phenol (octopamine) is a biogenic amine and a metabolite of tyramine, which is well established endogeneous invertebrates neuromodulator [1]. Since the discovery of various subclasses of trace amine associated receptors (TAARs), the studies of octopamine were extended on human physiology and pathophysiology [2]. The octopamine concentration in human plasma is 4.28 ± 0.28 ng mL^{-1} [2,3]. The compound occurs also in the trace level in mammalian brain. It has been hypothesized that the physiological role of octopamine might be complementary to physiological actions of neurotransmitters, and octopamine might share the same transfer systems as those of norepinephrine or dopamine. Whereas norepinephrine induces specific response by direct stimulation of postsynaptic receptors, octopamine may act to tune and refine such actions [4,5]. The turnover rate of octopamine is considerable faster than that reported for norepinephrine. It was noted, that the relative ratio of octopamine to norepinephrine in fetal life significantly varied and the concentration of octopamine is high until the third week of embriogenesis. Nevertheless, it is still little known about the role of octopamine in mammalian ontogenesis [4]. Octopamine plays also a certain role in various human disorders such as hepatic encephalopathy, schizophrenia, and renal diseases [6]. D'Andrea and co-workers investigated the role of octopamine in Parkinson's disease. Authors concluded that circulating octopamine level may hold promise as a biomarker of early stage of Parkinson's disease [2]. The effect of octopamine on α- and β-adrenoreceptors concerning lipolysis and glucose transport in mammalian fat cell have been investigated due to its possible application in obesity treatment [7]. For example *Citrus aurantium* extracts in which octopamine is present as component are already used in the dietary supplements. Recently, the stimulating properties of octopamine were identified by the World Anti-Doping Agency [8]. The compound has been prohibited in elite sport since several adverse findings concerning octopamine have been reported during doping control analyses. Moreover, the *N*-methylated analog of octopamine–synephrine, which is not banned by anti-doping authorities, might be converted *in vivo* into octopamine [9].

The accurate determination of octopamine is very difficult because of its presence in very low concentrations in complex samples (plasma or urine). A few analytical methods have been developed for its quantitative analysis including radioenzymatic assay [10], liquid chromatography [11] or capillary electrophoresis [12]. In the majority of cases the derivatization step is required to improve the sensitivity of methods. Yu and co-workers described the determination of octopamine in human plasma by capillary electrophoresis with optical fiber light-emitting diode-induced fluorescence detection [13]. Here, the main problem was the coexistence of other biogenic amines in the samples which simultaneously reacted with

* Corresponding author. Fax: +48 22 5720643.
E-mail address: dorota.maciejewska@wum.edu.pl (D. Maciejewska).

http://dx.doi.org/10.1016/j.talanta.2015.05.074
0039-9140/© 2015 Elsevier B.V. All rights reserved.

ARTICLE IN PRESS

derivatization agent decreasing the selectivity of method.

Next problem in the analytical procedures is existence a few isomeric forms of octopamine. Due to the position of hydroxy group in benzene ring, octopamine can occur in one of three isomers *ortho*, *meta*, *para* (2-, 3-, 4-). These isomers are the chiral molecules, and all can exist in the enantiomeric forms *R* and *S*. The naturally occurring form in human intracorporeal fluids is *R*-(−)-4-octopamine [14]. In the fruits extracts, in dietary supplements under certain conditions (such as high temperature or in the presence of modifier compounds), the racemization of octopamine could occurred, and both enantiomers *R*-(−) and *S*-(+) can be present [15].

The implementation of solid phase extraction (SPE) could be an advantageous step in octopamine determination for two reasons. Firstly, the preconcentration and enrichment procedure allows to isolate the analyte from complex samples. Secondly, new adsorbents dedicated to SPE give a possibility to use a dual system of enantiomeric sites to separate *R*-(−) and *S*-(+) stereoisomers simultaneously. Molecular imprinting of polymer matrices provides a straightforward procedure for obtaining the adsorbents with a desired complementarity to the chiral molecules [16,17]. In production of molecularly imprinted polymers (MIPs) the strategy that used the structural analog of analyte as the template while imprinting, provides advantage because it enables to avoid the bleeding of target analyte from the polymer matrix during the analysis [18]. Therefore, in the present studies (*R*,*S*)-2-amino-1-phenylethanol was selected as the structural analog of the target analyte (octopamine) in the imprinting process.

In this paper, the synthesis of (*R*,*S*)-2-amino-1-phenylethanol imprinted polymers as the separation materials for MISPE of octopamine racemate was presented. The affinity of the most appropriate polymer matrix was investigated by the non-competitive binding experiment of eight structurally related analytes which could exist in real samples. MISPE of (*R*,*S*)-octopamine from spiked human serum albumin was carried out in order to verify the applicability of novel sorbent.

The molecular modeling was employed to rationalize the stereodifferentiation of the analytes by the stereospecific sites formed in the polymer matix. This could be done based on the differences in the complexation energies between the chiral analyte and the cavities formed by the chiral templates [19].

2. Experimental

2.1. Materials and methods

(*R*,*S*)-(±)-2-Amino-1-phenylethanol (the template molecule, $\mathbf{T_{R/S}}$), (*R*,*S*)-(±)-1-(4-hydroxyphenyl)-2-aminoethanol hydrochloride (the target analyte, octopamine, $\mathbf{A1_{R/S}}$), 4-(2-aminoethyl) phenol hydrochloride (tyramine, **A2**), 4-(2-dimethylaminoethyl) phenol (hordenine, **A4**), (*R*,*S*)-(±)-1-(4-hydroxyphenyl)-2-methylaminoethanol (synephrine, $\mathbf{A5_{R/S}}$), 2-(3-indolyl)ethylamine (tryptamine, **A6**), 3-(2-aminoethyl)-5-hydroxyindole hydrochloride (serotonin, **A7**), and *S*-2-amino-3-(4-hydroxyphenyl)propionic acid (L-tyrosine, $\mathbf{A8_S}$) were from Sigma-Aldrich (Steinheim, Germany), and 4-(2-methylaminoethyl)phenol hydrochloride (*N*-methyltyramine, **A3**) was from Carbosynth Ltd. (Compton, United Kingdom). The functional monomers: methacrylic acid (**2**), trifluoromethacrylic acid (**4**), itaconic acid (**5**), isopropenylbenzene (**7**), 2-hydroxyethyl methacrylate (**8**), and 2-(diethylamino)ethyl methacrylate (**9**) were from Sigma-Aldrich (Steinheim, Germany), acrylamide (**6**) and allylamine (**10**) were from Fluka (Steinheim, Germany), 4-vinylbenzoic acid (**1**), acrylic acid (**3**) were from Alfa Aesar (Karlsruhe, Germany). The cross-linker, ethylene glycol dimethacrylate was purchased from Sigma-Aldrich (Steinheim, Germany). The solvents: methanol (pure as well as HPLC grade), acetone as well as hydrochloric acid (36%) were from POCh (Gliwice, Poland). The polymerization reaction initiator, 2,2′-azobisisobutyronitrile, AIBN, was from Merck (Darmstadt, Germany) and the salts, ammonium formate and ammonium acetate were from POCh (Gliwice, Poland). The monomers were purified prior to use by standard procedures (vacuum distilled or recrystallized from the appropriate solvents). All other reagents were used without purification. Ultra-pure water delivered from a Milli-Q purification system (Millipore, France) was used to prepare the water solutions. Human serum albumin was delivered from Sigma-Aldrich (Steinheim, Germany).

The stock solutions of analyzed compounds (**A1**–**A8**) were prepared by an accurate weighing of appropriate amount of each compound and dissolving in methanol (**A1**–**A7**) or in water adjusted to pH 3 with 0.1 M aq. hydrochloric acid **A8** to obtain concentration of 10 mmol L^{-1}. The standard solutions were prepared prior to use by dilution of the appropriate stock solutions with methanol–water (85:15 v/v) to obtain the required concentrations. All stock solutions were stored in dark at +8 °C.

The UV measurements were performed with a UV-1605PC spectrophotometer (Shimadzu, Germany). The calibration lines were constructed as a function of peak area under absorbance curve at λ_{max} of each compound (*y*) *vs*. concentration (*x*). Each point was measured in triplicate. The linearity of calibration lines was good with correlation coefficients $r^2 > 0.997$. The wavelength, λ_{max}, the limits of quantification, LOQ (in μmol L^{-1}) and the limits of detection, LOD (in μmol L^{-1}) were as follows: (**A1**), 275, 12.78, 4.22; (**A2**), 277, 16.26, 5.36; (**A3**) 277, 15.47, 5.10; (**A4**) 278, 16.58, 5.47; (**A5**), 276, 21.31, 7.03; (**A6**), 280, 4.43, 1.46; (**A7**), 276, 19.40, 6.40; (**A8**), 276, 21.53, 7.11. RP-HPLC was used for the determination of **A1** in human serum albumin. The HPLC system consisted of a LC 10AT pump, an CTO 10A oven, a RF 551 spectrofluorimetric detector operated at λ_{ex}=280 nm and λ_{em}=315 nm, and an SIL 20A HT autosampler (Shimadzu, Germany). Chromatographic separation was performed using a Discovery HS F5 stainless steel column (150 mm × 4.6 mm ID, 5 μm, Supelco, Bellefonte, PA, USA), preceded by a 20 mm × 4.6 mm ID, Discovery HS F5 guard column. The mobile phase consisted of 0.05 M aq. pH 3 ammonium formate–methanol 5:95 v/v delivered at a flow rate of 1 mL min^{-1}. The five-point calibration line was constructed for **A1** as a function of peak area (*x*) *vs*. concentration (*y*) in the range of 5–50 μmol L^{-1}. The linearity of calibration line, $y = 104847x + 83042$, was good with the correlation coefficients $r^2 > 0.989$, LOQ and LOD values (in μmol L^{-1}) were 7.59 and 2.50, respectively.

2.2. Synthesis of polymers

The experimental amounts of the reagents (moles, masses, and volumes) used for the preparation of the different types of polymers are listed in Table 1. The molecularly imprinted polymers (MIPs) coded as MIP**1**–MIP**10** were prepared by the radical bulk polymerization. Briefly, (*R*,*S*)-(±)-2-amino-1-phenylethanol (the template), the appropriate functional monomer, and ethylene glycol dimethacrylate, EGDMA (the cross-linker) were dissolved in methanol (the porogen) in a thick-walled glass tubes. A molar ratio of the template to the functional monomer and the cross-linker was equal to 1:4:20. At the end, the initiator of polymerization, 2,2′-azobisisobutyronitrile (AIBN), was added. The homogeneous solutions were purged with nitrogen for *ca.* 5 min and then the glass tubes were sealed. Subsequently, the polymerization was carried out under a nitrogen atmosphere for 24 h at 64 °C. The bulk rigid polymers were ground in a mortar with a pestle and wet-sieved into the particles below 45 μm diameter. Fine particles were separated by repeated decantation in acetone. 2-Amino-1-phenylethanol was removed from the polymer with continuous

Please cite this article as: M. Sobiech, et al., Talanta (2015), http://dx.doi.org/10.1016/j.talanta.2015.05.074

Download English Version:

https://daneshyari.com/en/article/7678645

Download Persian Version:

https://daneshyari.com/article/7678645

Daneshyari.com