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Environmental chiral analysis of β-blockers: Evaluation of different n-alkyl-modified SBA-15 mesoporous silicas as sorbents in solid phase extraction

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Environmental context. β-blockers are important chiral pharmaceuticals that can be found as micropollutants in environmental waters, due to an incomplete removal during wastewater treatment. They are responsible of enantioselective toxicity, so it is necessary to include enantioselectivity in environmental risk assessment. We have developed n-alkyl-modified SBA-15 mesoporous silicas that allow the extraction and preconcentration of β-blockers in water samples prior to chiral analysis.

Abstract. The extraction and preconcentration of chiral β-blockers in environmental water was evaluated by solid-phase extraction (SPE) employing a SBA-15 ordered mesoporous silica, funtionalized with alkyl chains of different length. The materials were characterized by X-ray diffraction (XRD), transmission electron microscopy (TEM), scanning electron microscopy (SEM), nitrogen adsorption-desorption isotherm measurements and elemental analysis. Important parameters on extraction efficiency, including the type and amount of sorbent and the breakthrough volume were optimized. The results obtained showed that the organic chain length played an important role in the behavior of these sorbents. Under optimized conditions, using 200 mg SBA-15-C8 as sorbent, a simple analytical method based on off-line SPE coupled to chiral capillary electrophoresis with diode array detection (SPE-chiral CE-DAD) was developed. Method detection and quantification limits were lower than 0.6 and 1.9 μg L^{-1}, respectively, for all enantiomers, with a preconcentration factor of 500-fold. The method was successfully employed to chiral analysis of atenolol, metoprolol, pindolol and propranolol in river and sewage water samples. Satisfactory recoveries (between 86 ± 2% and 98 ± 1%) and repeatability (RSD < 9 %, n = 3) were obtained.

Metoprolol was detect in sewage water with a concentration of 10.7 and 9.9 μg L^{-1} and an enantiomeric fraction of 0.52 and 0.48 for first and second enantiomer, respectively. These results emphasize the importance of enantioselective analysis for environmental risk assessment.

Additional keywords: chiral pharmaceutical compounds, environmental water, functionalized ordered mesoporous silica, solid-phase extraction

Introduction

Nowadays, pharmaceuticals have become important emerging contaminants. Thousands of different active compounds have been found over the last 20 years in different ecosystems (Grenni et al. 2018). Their presence in waters is due to its poor degradability and incomplete elimination in wastewater treatment plants. Many groups of these pharmaceuticals are chiral compounds, existing in the environment as a single enantiomer or as mixture of both enantiomers (Ribero et al. 2012; Richardson and Ternes 2014; Serrano et al. 2016; Casella et al. 2016). Therefore, when discharged into the environment, enantiomers can allow different degradation and conduct to a different variety of compounds (Ribeiro et al. 2017). B-blockers are one of the most important chiral pharmaceuticals, used in the treatment of cardiovascular disorders. As a result of the incomplete removal of these pharmaceuticals during conventional wastewater treatment, β-blockers have been found in surface waters, and are commonly detected in samples of river and drinking waters (Jelic et al. 2012; WHO 2012; León Gonzalez and Rosales-Corrado 2016). In general, each enantiomer possesses a different pharmacological activity, potency and mode of action. For example, *S*-(-)-enantiomers are usually more active than their respective *R*-(+)-isomers. In addition, some of the biotransformation pathways for β-blockers in humans are stereoselective (Aturki et al. 2011). For these reasons, chiral analysis must be taken into consideration for a better understanding of the environmental and human health impact of this type of emerging contaminants in waters. In order to characterize the enantiomeric distribution of these compounds in waters, capillary electrophoresis (CE) has become widely popular, due to its high efficiency and selectivity, simplicity,

74 versatility, low sample and chiral selector consumption (Chankvetadze 2001). Chiral
75 selectors can be used in CE as buffer additives for chiral separations, and this allows
76 saving in expensive chromatographic columns. Cyclodextrins (CDs) are by far the most
77 popular chiral selectors, due to their low toxicity, high solubility in mainly aqueous
78 background electrolytes, UV transparency and high commercial availability. β-blocker
79 enantiomers have been separated using native and derivatised CDs, achieving good
80 enantioresolution (Rs) and separation efficiency, under different optimized conditions
81 (Aturki et al. 2011). Among the wide range of CDs evaluated as chiral selectors in CE,
82 carboxymethylated-β-CD (CM-β-CD) was often used for this task, achieving good
83 resolution values (Silva et al. 2017). For example, Huang et al. (2008) reported the
84 simultaneous separation of atenolol (Rs: 1.2), metoprolol (Rs: 1.6) and propranolol (Rs:
85 2.2) using CM-β-CD with a total analysis time of 35 min. However, the main
86 disadvantage of this negatively chargeable CD is its high price and in this sense chiral
87 analytical methods for the simultaneous determination of β-blockers using cheaper
88 CDs are needed.

89 Pharmaceuticals can appear in waters in the ng L^{-1} to μg L^{-1} range (Scheurer et al.
90 2010; Maszkowska et al. 2014; Casella et al. 2016) so it is necessary to carry out an
91 extraction and preconcentration step prior to its chiral analysis. Over the last twenty
92 years, solid-phase extraction (SPE) has become the most powerful sorbent technique
93 available for rapid sample preparation, mainly due to its simplicity and limited usage of
94 organic solvents. In general, this technique involves the use of disposable cartridges
95 (filled with the sorbent) to trap analytes and separate them from the bulk of the matrix
96 of liquid samples or extracts. During SPE procedure, sample matrix can affect the

97 ability of the sorbent to extract the analytes due to competition for retention. Thus, it
98 is important to select a suitable sorbent, in order to control parameters such as
99 selectivity, affinity and capacity. This choice highly depends on the target analytes and
100 the interactions between the chosen sorbent and the functional groups of the
101 analytes. Different materials have been evaluated as SPE sorbents for the
102 determination of β-blockers (Caban et al. 2015), such us chemically modified
103 amorphous silica-based sorbents, chemically modified polymeric sorbents, hydrophilic-
104 lipophilic balanced (HLB) copolymeric sorbents, mixed-mode polymeric sorbents and
105 molecularly imprinted polymeric (MIPs) sorbents. However, despite that another
106 advantage of SPE is its versatility, as a result of the different types of sorbents
107 commercially available, some of them suffer limitations such as low capacity, long
108 equilibrium times, low selectivity and mechanical/or thermal instability. For this
109 reason, one of the main tendencies on the research related to SPE is to develop novel
110 sorbents, in order to improve the characteristics of previous ones and thereby the SPE
111 results (Augusto el al. 2013; Plotka-Wasylka et al. 2016). In this context, synthesis and
112 application of new materials, such as, ordered mesoporous silicas (OMSs), as SPE
113 sorbents has become a very interesting research area. Recently, OMSs are gaining
114 increasing application for sample preparation because of their desirable characteristics
115 (Zhao et al. 2012; Gañan et al. 2016; Sreenu et al. 2016; Wu et al. 2016; Casado et al.
116 2017a; Kejik et al. 2017). These sorbents show unique advantages for this task, as they
117 have: (1) highly ordered and size-controlled mesoporous structure, (2) extremely high
118 surface area and large pore volume, (3) thermal and chemical stability and (4)
119 flexibility for functionalization. The synthesis of OMSs functionalized with different

kinds of ligands is a good alternative to the classical amorphous silica, so efficient extraction and preconcentration of the analytes of interest can be achieved (Pérez-Fernández et al. 2014; Dahane et al. 2016). Casado et al. (2017b) have recently published a review with the most relevant achievements in the preparation and application of OMSs for xenobiotics extraction in different samples, including waters.

In this study, OMS (SBA-15 type) was prepared and functionalized by the post-synthesis method with chloro(dimethyl)silane derivatives ($(CH_3)_2$Cl-Si-R), with alkyl chains of different length, R = C3 (n-propyl), R = C8 (n-octyl) and R = C18 (n-octadecyl), to obtain SBA-15-C3, SBA-15-C8 and SBA-15-C18 materials, respectively. To evaluate the effect of the alkyl chains length on the extraction and preconcentration capacity, the resulting functionalized mesoporous silicas were characterized and evaluated as SPE sorbents to extract four enantiomeric pairs of β-blockers from water samples. Using SBA-15-C8 material as sorbent and methylated-β-CD as chiral selector, a SPE-chiral CE-DAD method was developed for the determination of atenolol (Ate), metoprolol (Met), pindolol (Pin) and propranolol (Prop) enantiomers in environmental waters, showing good precision, linearity, accuracy, method detection and quantification limits. The SPE-chiral CE-DAD method was validated and its application for the simultaneous analysis of these compounds in river and sewage waters was demonstrated. To our knowledge, this is the first work, where different n-alkyl-modified OMSs have been evaluated as SPE sorbents for the analysis of chiral emerging contaminants in water. Results demonstrate that the developed SPE-chiral CE-DAD method could have good application prospects.

Experimental

Chemicals and reagents

Poly(ethylene glycol)-block-poly(propylene glycol)-blockpolyethylene glycol, Pluronic 123 (M_{av} = 5800 g mol^{-1}), chloro(dimethyl)propylsilane ≥ 97.0% (M = 157.11 g mol^{-1}), chloro(dimethyl)octylsilane 97% (M = 206.83 g mol^{-1}), chloro(dimethyl)octadecylsilane 95% (M = 347.11 g mol^{-1}), tetraethylorthosilicate 98% (M = 208.33 g mol^{-1}), were purchased from Sigma-Aldrich (St. Louis, MO, USA). Methylated-β-CD (M-β-CD) was purchased from Fluka (Buchs, Switzerland). Hydrochloric acid 37%, toluene, diethyl ether, methanol and ethanol were purchased from Scharlau (Barcelona, Spain). All pharmaceutical standards used were of high purity grade ≥ 98 %. *S*-(−)-propranolol hydrochloride (*S*-Prop), *S*-(−)-atenolol (*S*-Ate), (±)-propranolol hydrochloride, (±)-atenolol, (±)-metoprolol, and (±)-pindolol were purchased from Sigma–Aldrich (Madrid, Spain). Water (resistivity 18 MΩ cm) used in the preparation of standard solutions was obtained from a Millipore Milli-Q System (Waters, USA).

The appropriate amount of (±)-Prop, (±)-Met, (±)-Ate and (±)-Pin was dissolved in methanol to give stock solutions with a final concentration of 1000 mg L^{-1}. For method optimization, a standard solution was prepared daily by diluting the stock solutions with running background electrolyte (BGE) to achieve a final concentration of 50 mg L^{-1} of each enantiomer. All standard solutions were filtered through a 0.45 μm pore size nylon filter membrane and stored each day at 4 ºC.

Environmental water samples

River water (pH 7.2) was collected in the Manzanares River (Madrid). Sewage water (pH 6.9) was collected in the wastewater treatment plant (effluent water) of the Rey Juan Carlos University (Mostoles, Madrid), located near to a hospital. Both waters were filtered through a 0.45 μm membrane filter (Millipore Membrane filters, 0.45 μm HA) to remove suspended particles and stored, in polyethylene bottles, at 4 ºC until analyses were done (maximum 12h after the collect).

Preparation of OMSs

SBA-15 was prepared according to the method described in our previous work (Gañan et al. 2014). Briefly, 48.4 g of Pluronic 123 were dissolved in 360 mL of water and 1440 g of 2.0 M HCl solution with stirring at room temperature. Then 102 g of tetraethylorthosilicate were added to the solution, and the mixture was stirred for 20 h at room temperature. The solid product was recovered by filtration and washed with water. A post-synthesis method was used to functionalize the material, in order to obtain SBA-15-C3, SBA-15-C8 and SBA15-C18. Surface modification of SBA-15 was carried out as follows: 5 g of SBA-15 were suspended in 50 mL of anhydrous toluene and mixed with 15% in weight with respect to the mass of SBA-15 of the chloro(dimethyl)silane derivatives (0.83 g of C3, 0.75 g C8 or 1.50 g of C18). The mixture was heated at 80 ºC for 24 h at 500 rpm. Finally, the solid was washed with two fractions of 50 mL of toluene, ethanol, and diethyl ether.

189 *Characterization of OMS*

190 Powder X-ray diffraction (XRD) pattern of the materials were recorded using a Phillips
191 Diffractometer model PW3040/00 X´Pert MPD/MRD at 45 KV and 40 mA, using a
192 wavelength Cu Kα (k = 1.5418 Å) over a range 0.4º < 2θ < 5º at room temperature.
193 Scanning electron micrographs (SEM) and morphological analysis were carried out on a
194 XL30 ESEM Philips with an energy-dispersive spectrometry system. The materials were
195 treated with a sputtering method with these parameters: sputter time 100 s, sputter
196 current 30 mA, and film thickness 20 nm using sputter coater BAL-TEC SCD 005. N_2 gas
197 adsorption–desorption isotherms were recorded using a Micromeritics ASAP 2020
198 analyzer. Conventional transmission electron microscopy (TEM) was carried out on a
199 TECNAI 20 Philips microscope operating at 200 kV, with a resolution of 0.27 nm and ±
200 70º of sample inclination, using a BeO sample holder. N_2 gas adsorption–desorption
201 isotherms were measured at the temperature of liquid nitrogen (-196 ºC) over the
202 interval of relative pressures (P/P_0) from 10^{-4} to 0.993. Before the measure of the N_2
203 adsorption–desorption isotherms, in order to remove possible volatile adsorbed
204 species, like rest of solvents used during the synthesis of the mesoporous material and
205 to eliminate physical adsorbed water, that could affect the analysis, the samples were
206 heated at 90 ºC in vacuum during 10 h in the port of degasification of the instrument.
207 Such temperature was chosen to avoid any degradation of the alkylsilane chains
208 anchored in the silica surface. The specific surface areas were calculated using the
209 Brunauer, Emmett and Teller (S_{BET}) model. The pore size distributions were calculated
210 using the Barrett–Joyner–Halenda (BJH) model on the desorption branch. Elemental

211 analysis (% C) was performed with a LECO CHNS-932 analyzer (Universidad Rey Juan
212 Carlos, Spain).
213
214 *SPE procedure*
215 β-blockers were extracted from water samples using an off-line SPE procedure. To
216 prepare the SPE cartridges, 100 or 200 mg of sorbent were packed into a 6 mL syringe
217 type cartridge (65 mm length, 11 mm diameter) plugged with porous
218 polytetrafluoroethylene disks at both ends. To prevent the material lost during sample
219 loading, a 0.45 µm pore size nylon filter membrane was also inserted at the bottom of
220 the OMSs bed. Extraction was performed using a SPE vacuum manifold 12 port model
221 connected to a vacuum pump at 7.6 psi (flow rate 1.0 mL min^{-1} aprox.). The cartridges
222 were conditioned, with 3 mL of methanol and 3 mL of deionized water. The water
223 samples were loaded to the cartridge under vacuum (flow rate 1.0 mL min^{-1} aprox).
224 Prior to the elution of the analytes with 5 mL methanol, the cartridges were flushed
225 with 3 mL deionized water and then dried for 20 min. Finally, the corresponding
226 extracts were evaporated under vacuum and reconstituted with 500 µL of BGE for
227 subsequent analysis by chiral CE.
228 In order to evaluate the SPE procedure, recoveries (R) were calculated using the
229 following equation [1]:
230 $$R\ (\%) = [Ac_{pre\text{-}SPE} / Ac_{post\text{-}SPE}] \times 100 \qquad [1]$$
231 where $Ac_{pre\text{-}SPE}$ is the corrected peak area for the analyte recorded for the water
232 sample spiked with the target β-blockers prior the SPE process and $Ac_{post\text{-}SPE}$ is the
233 corrected peak area for the analyte recorded for the water sample spiked with the

target β-blockers after the SPE process. In each case, three sample cartridges were prepared and analyzed in triplicate.

Electrophoretic separations

Electrophoretic experiments were carried out in an HP ^{3D}CE system from Agilent Technologies (Palo Alto, CA, USA) with a diode array detector (DAD). The electrophoretic system was controlled with the HP ^{3D}CE ChemStation software that included the data collection and analysis. Electrophoretic separations were carried out with uncoated fused-silica capillaries (50 µm ID, 362.1 µm OD) having 50.2 cm total length (41 cm to the detector) purchased from Polymicro Technologies (Phoenix, AZ, USA), in similar conditions in our previous work with some modifications (Silva et al. 2017). The applied voltage was 20 kV, the capillary was thermostatized at 20 ºC and the background electrolyte (BGE) was 50 mM phosphate buffer at pH 2.5 and 1.25% M-β-CD (w/v). Samples were injected by applying a pressure of 50 mbar for 5 s. The detection was performed at 220 nm for Pin and Prop enantiomers, and at 200 nm for Ate and Met enantiomers to achieve the maximum sensibility. The migration order of the enantiomers of Prop and Ate was determined with standard working solutions injected at double concentration of *S*-(−)- than *R*-(+)-enantiomer. Migration time (t_m) of the analytes were as follows: 21.43 and 22.01 min for 1-Pin and 2-Pin, 22.54 and 22.83 min for *S*-Ate and *R*-Ate, 37.32 and 38.73 min for *S*-Prop and *R*-Prop, 40.69 and 41.33 min for 1- Met and 2-Met.

257 *Analytical procedure validation*

258 The SPE-chiral CE-DAD method for Prop, Pin, Ate and Met determination in river and
259 sewage waters was validated in terms of linearity, precision, accuracy and limits of
260 detection and quantification. Linearity was estimated by a matrix matched calibration
261 curve, loading 250 mL of river or sewage water into the SPE cartridge, then the eluate
262 was spiked with the appropriate amount of a standard mixture of the four β-blockers.
263 Six calibration points, between 4 - 200 μg L^{-1} (2 - 100 μg L^{-1} for each enantiomer), were
264 prepared. Repeatability, expressed in terms of % RSD of corrected peak area, Ac (Ac =
265 peak area/t_m), was calculated from six consecutive analysis of the water sample in one
266 day at two concentration levels: 2 and 100 μg L^{-1} each enantiomer (at low and high
267 concentration levels). Within-laboratory reproducibility was calculated as % RSD of Ac
268 for all enantiomers from three consecutive days and 3 replicates for day at low
269 concentration level (2 μg L^{-1} for each enantiomer). The method detection and
270 quantification limit (MDL and MQL) were estimated by application of the
271 preconcentration factor of 500 to the concentration level corresponding to a signal-to-
272 noise ratio of 3 or 10, respectively, from the noise measured in the injection of a river
273 or sewage water sample spiked at low concentration level (2 μg L^{-1} for each
274 enantiomer).

275 To assess the accuracy of the method, β-blockers extraction recoveries were
276 determined at two spiking levels: 2 μg L^{-1} and 100 μg L^{-1} each enantiomer. R (%) were
277 calculated, according to equation [1], by comparison of the analyte electrophoretic
278 peak area measured when a spiked river or sewage water sample was extracted by SPE
279 ($Ac_{pre\text{-}SPE}$) and the analyte electrophoretic peak area measured when the same

280 unspiked river or sewage water sample was extracted by SPE and then its eluate was
281 spiked ($Ac_{post\text{-}SPE}$). All assays were performed with river or sewage water samples
282 previously analyzed, to verify the absence or presence of the target analytes.
283

284 ## Results and discussion

285 *OMSs characterization*

286 Powder XRD patterns of the SBA-15 showed three Bragg peaks at low 2ϑ angles, a
287 strong (100) diffraction peak, around 0.92 Å, as well as the (110) and (200) reflections
288 of lower intensity, at 1.55 Å and 1.81 Å, respectively, which characterize the highly
289 ordered hexagonal pore structure in this silica. The n-alkyl-modified materials exhibit
290 almost identical XRD patterns that clearly indicate that the basic SBA-15 pore structure
291 remained unchanged after surface modification. The TEM images of SBA-15 showed
292 the hexagonal array of uniform channels running parallel, with the typical honeycomb
293 appearance of SBA-15 silica (Fig. 1A and B). The SEM images revealed a uniform
294 particle size for this material, with an average size around 400 nm in one axis and
295 around 800 nm in the other axis (Fig. 1C). The particles, with the typical rope-like
296 morphology of the SBA-15, aggregated into wheat-like macrostructures, which is a
297 common feature of this type of OMSs. After functionalization, all the prepared n-alkyl-
298 modified SBA-15 silicas kept the same morphology, particle size and structure that
299 those of the as-synthesized SBA-15.

300 N_2 adsorption-desorption isotherms for SBA-15-C3, SBA-15-C8 and SBA-15-C18
301 are shown in Fig. 2A. The isotherms are type IV, according to the I.U.P.A.C.
302 classification (Sing et al. 1985), with an H1 hysteresis loop that is representative of

materials with pores of constant cross-section and cylindrical shape. In SBA-15, the S_{BET}, pore volume and BJH pore diameter were 764 $m^2\ g^{-1}$, 0.80 $cm^3\ g^{-1}$ and 55.5 Å, respectively (Table 1). After functionalization, all synthesized materials possessed lower S_{BET}, pore volume and BJH pore diameter, as consequence of the alkyl ligands attached in their surface or inside the mesopores. In these materials, surface modification takes place by forming Si–O–Si bonds due to the reaction between silanol groups (Si–OH from SBA-15) and Si–Cl groups (from chloro(dimethyl)silanes). Thus, reproducible surface coverages (without vertical polymerization) could be achieved by using monofunctional silanes in the post-synthesis modification. The textural properties of the modified silicas were maintained in the range of the mesoporous materials (Table 1), showing isotherms type IV and H1 hysteresis loop (Figure 2A), which is of extreme importance for further application of them as sorbents. As it can be seen, these materials exhibited a reduction in surface area, pore diameter and total pore volume after the functionalization. Figure 2B shows the pore size distribution of SBA-15-C3, SBA-15-C8 and SBA-15-C18.

Finally, with the percentage of C in the materials, calculated by elemental analysis, the amount of C3, C8 or C18 groups attached to the n-alkyl-modified materials (functionalization degree, *Lo*) was estimated (Table 1). Data obtained demonstrated a higher *Lo* for the material modified with shorter alkyl chains (C3) that can be explained by the possibility for the shorter chains to bind at the interior of the mesoporous channels. The longer chains (C8 and C18) are assumed to bind primarily on the surface of the SBA-15 silica, and – due to steric hindrance – have less chance to advance to the interior part of the channels. This fact explained the higher reduction in

326 the pore diameter observed in the SBA-15-C3 material (41 Å) as compared with SBA-
327 15-C8 and SBA-15-C18 materials (around 50 Å in both cases).
328
329 *Optimization of the SPE procedure with the OMSs*
330 For the SPE procedure optimization, tap water collected in the laboratory (spiked
331 at 25 μg L^{-1} each enantiomer) was used. This sample was used to check the ability of
332 the prepared sorbents (SBA-15-C3, SBA-15-C8 and SBA-15-C18) for the extraction of
333 the target β-blockers during the loading step and their potential desorption during
334 elution process, without taking into account the matrix complexity of the sample. It is
335 well known that recoveries of the extracted analytes can decrease with the increase in
336 the matrix complexity. For this reason, once the usefulness of the prepared sorbents
337 was demonstrated with this water (low complex matrix), the validation of the method
338 was carried out for environmental waters (river and sewage waters) that have more
339 complex matrix.
340 For method development, sorbents (100 or 200 mg) were packed into cartridges
341 that were previously conditioned as indicated in the Experimental section. The water
342 samples were loaded to the cartridge at a flow rate of 1.0 mL min^{-1}. Prior to the elution
343 of the analytes, the cartridges were flushed with 3 mL of deionized water and then
344 dried for 20 min. Elution was carried out with 5 mL of metanol, taking into account
345 that this solvent was selected as the more suitable for this task in previous works (Silva
346 et al. 2017). The pH of water samples was not adjusted, in order to avoid changes in
347 the composition of the samples analysed, and to reduce the consumption of reagents.
348 Thus for optimization of the SPE process, the effect of the type of sorbent (effect of

349 the n-alkyl chain length), the effect of the sample loading volume and the effect of the
350 sorbent amount was evaluated.
351
352 *Effect of the type of sorbent*
353 Firstly, a study of the effect of the n-alkyl chain length in the functionalized
354 mesoporous silicas was carried out. For this study, 100 mg of each material were used
355 as sorbent in the SPE process. As it can be seen in Table 2, recoveries obtained with
356 SBA-15-C3 were not satisfactory (between 43 ± 1% and 75 ± 7%). This fact can be
357 attributed to the low hydrophobicity of the C3 alkyl chain that was not enough to
358 quantitatively retain the target analytes inside the mesopores. On the other hand,
359 SBA-15-C8 and SBA-15-C18 showed a better retention capacity, with recovery values
360 between 79 ± 4% and 107 ± 5% for all analytes. Considering that *Lo* of SBA-15-C3 (0.45
361 mmol g^{-1}) and SBA-15-C8 (0.42 mmol g^{-1}) was quite similar, it also can be concluded
362 that the retention capacity of the sorbent increased significantly with the hydrophobic
363 properties of the material, from the increasing length of the alkyl chain (from 3 to 8
364 carbons). As shown in Table 2, recoveries obtained using SBA-15-C8 and SBA-15-C18
365 sorbents were greater than the ones obtained with bare SBA-15 (49 ± 3% and 70 ± 2%),
366 under the same conditions. Therefore, these functionalized materials were clearly
367 more effective in the extraction of the four enantiomeric pairs of β-blockers in tap
368 water than the non-modified mesoporous silica. This behavior can be explained taking
369 into account both the functional groups included in the target analytes and the
370 characteristics of the silica surface. Thus, in SBA-15-C8 and SBA-15-C18 materials the
371 target compounds can experience not only a reversed-phase sorption with the C18

groups (by hydrophobic interactions), but also polar secondary interactions with surface silanol groups (by hydrogen bonding interactions) that improves the retention of analytes. With both materials, there was not found significant difference between recoveries achieved for both enantiomers, whereby, these materials were good for the extraction of these chiral compounds.

Effect of sample loading volume (breakthrough) and sorbent amount

Since the breakthrough volume is a very important parameter in a SPE procedure (maximum volume of water sample that can be preconcentrated on the sorbent without loss of the target analyte), this parameter was evaluated using 100 mg of both SBA-15-C8 and SBA-15-C18 sorbents. As shown in Fig. 3 A and B, recoveries were satisfactory with sample volumes up to 150 mL (without adjusting the pH). From these results, it can be concluded that both materials behave in a similar way, with a slightly higher average recovery with SBA-15-C18 (around 100% for all enantiomers) in comparison to SBA-15-C8. A theoretical 300-fold preconcentration factor was achieved under these experimental conditions (preconcentration of a 150 mL sample volume).

To complete the study of the behaviour of both materials, the breakthrough volume was studied using 200 mg of sorbent packed on the cartridge. As it can be seen in Fig. 3 C and D, when the extraction process was carried out with 250 mL of water sample, recovery values were, in general, significantly increased in comparison to those obtained with 100 mg of both materials (Fig. 3A and B). However, in this case, results were different as the SBA-15-C8 enables a greater breakthrough volume, up to 250 mL, with recoveries around 80% for all enantiomers. In similar conditions, with

395 SBA-15-C18 as sorbent very low recoveries were observed for Ate enantiomers. This
396 fact can be attributed to the low hydrophobicity of Ate (Log K_{ow} = 0.16), that makes it
397 difficult the retention on the very hydrophobic C18-modified SBA-15. Therefore, a
398 volume of 250 mL was chosen to be the maximum volume of water sample for
399 extraction, and the use of 200 mg of SBA-15-C8 provided recoveries around 80 % for
400 the four enantiomeric pairs of β-blocker, allowing a theoretical 500-fold
401 preconcentration factor. The high extraction efficiency of SBA-15-C8 sorbent can be
402 attributed to the high density of n-alkyl chain in this material (*Lo* = 0.42 mmol g^{-1})
403 compared with the SBA-15-C18 sorbent (*Lo* = 0.21 mmol g^{-1}).

404 Table 5 shows some reported methods for the SPE extraction of β-blockers
405 from waters. As it can be seen, preconcentration of β-blockers in waters has been
406 carried out with different commercially available sorbents: mixed-mode polymers (e.g.
407 Strata-X-C® and Oasis MCX® cartridges), chemically modified polymers (e.g. Strata-X®
408 cartridges), hydrophilic-lipophilic balanced copolymers (e.g. Oasis HLB® cartridges),
409 molecularly imprinted polymers (e.g. SupelMIP® and MIP4SPE™ cartridges) and
410 chemically modified amorphous silicas (e.g. Strata C18-E® and Bakerbond® C18
411 cartridges). On the other hand, as it can be seen in Table 5, to the best of our
412 knowledge, there are only two previous works in the literature were “in-house”
413 synthesized OMSs have been evaluated as novel sorbents for this task. In order to
414 evaluate the effectiveness of β-blockers extraction using different commercially
415 available and “in-house” prepared SPE sorbents, recovery values and preconcentration
416 factors (PF) were compared in Table 5 (besides other parameters such as sorbent
417 amount, sample pH and type of water).

Mixed-mode strong cation-exchange polymeric sorbents have been very used, mainly Oasis MCX® cartridges (Table 5). Depending on the type of water sample, up to 2000-fold PF has been achieved in some works (Kasprzyk-Hordern et al. 2007 and 2008; Scheurer et al. 2010). However, extraction properties of this sorbent were different with respect to the target β-blocker, with very low recoveries in some studies such as 25% for Pin in effluent wastewaters (Salen et al. 2012), 10% for Pin in influent wastewaters (Piram et al. 2018) or 40% for Pro in surface waters (Kasprzyk-Hordern et al. 2008). In addition, the main disadvantage of this kind of sorbent is the need to acidify the water sample (to pH 2 - 3). Among the commercially available sorbents, Oasis HLB® (polystyrene-*N*-vinylpyrrolidone-divinylbenzene) cartridges have been widely used to extract different drugs mixtures by SPE, with wide-ranging physicochemical properties, in a variety of water samples. In general, recovery values of the target β-blockers using these cartridges were better, compared to those obtained with Oasis MCX® cartridges, so this sorbent had better potential than mixed-mode cation-exchange sorbents for the extraction of basic drugs from environmental water samples (Table 5). However, with these hydrophilic-lipophilic copolymeric sorbents (Oasis HLB®) and with other chemically modified polymers (Strata-X®) lower PF (in general ≤ 200) were achieved with some exceptions (eg. Miegé et al. 2006 and Vieno et al. 2006). On the other hand, highly selective sorbents, based on molecularly imprinted polymers (e.g SupelMIP® and MIP4SPE™ cartridges) have also been tested for selective β-blocker extraction (Table 5). These synthetic polymers have specific cavities matched to a template molecule and a retention mechanism based on molecular recognition. Good extraction properties with respect to the target analytes

441 (recoveries near 100%) but with low PF (100-fold) have been found in some works
442 (Morante-Zarcero and Sierra, 2012a; Morante-Zarcero and Sierra, 2012b). In addition
443 to the analytical performance, taking into account the cost of these sorbents, the high
444 cost of HLB and MIPs cartridges precludes their use to develop inexpensive SPE
445 procedures for routine analysis.

446 Amorphous silica-based materials, functionalized with C18 groups, have also
447 been evaluated for this task (Table 5). For example, Scheurer et al. (2010) used
448 Bakerbond C18 cartridges to extract 13 β-blockers in waters but low recoveries (16%
449 for Met in influent water, 40% for Ate in effluent water and 69% for Pro in river water)
450 were achieved. In this sense, due to the disadvantages of these kind of materials (low
451 recovery in the extraction of some compounds, instability at extreme pH, irregular
452 pore channels that making them more susceptible to blockage etc.), nowadays, they
453 have been replaced by commercially available polymeric sorbents. However, with the
454 aim to develop new silica-based sorbents that improve the characteristics of previous
455 ones, two recent works have evaluated the use of “in-house” synthesized OMS. Thus,
456 Dahane et al. (2016) used 100 mg of non-modified OMS (MCM-41 type) to extract Ate
457 and Pin, among other pharmaceuticals, in river water (pH adjusted to 2). These authors
458 achieved a PF of 100 and recoveries between 67 - 88%. More recently, Silva et al.
459 (2017) developed a SPE methodology by using octadecyl-modified SBA-15, achieving
460 good recoveries (between 96 and 105% for Pin, Ate, Prop, Met in tap, river and ground
461 waters) with a PF of 300. According to the results of this study, SBA-15-C8 has showed
462 a big extraction potential compared with other silica-based commercial sorbents (eg.
463 Strata C18-E® and Bakerbond C18®cartridges) and OMSs, since offers advantages, not

464 only for the good recovery values obtained, but also for the good PF achieved (500-
465 fold) working with environmental waters. These good results can be attributed to the
466 highly ordered and size-controlled mesoporous structure of the SBA-15-C8, besides its
467 extremely high surface area, large pore volume and high funtionalization degree.
468 Another advantage of the SPE at neutral pH, is the separation of many polar organic
469 impurities in the matrix, which were not retained due to their positive or negative
470 excess charges.

471 To summarize, based on recovery values and PFs (Table 5) among the tested
472 SPE sorbents (commercially available and "in-house" synthesized OMSs), the SBA-15-
473 C8 could be a good SPE sorbent to enrich trace of β-blockers in environmental water
474 samples and a good alternative to the most frequently used commercially sorbents
475 (Oasis MCX® and Oasis HLB®).

476

477 *Analytical performance of the SBA-15-C8 based SPE-chiral CE-DAD method*

478 First, the instrumental validation of chiral CE separation was studied in terms of
479 linearity, precision, instrumental detection limit (IDL) and instrumental quantification
480 limit (IQL). Calibration curves obtained by preparing six standard solutions (covering a
481 range of 1 to 100 mg L^{-1} for each enantiomer) were shown to be linear with
482 determination coefficients (R^2) > 0.99. To evaluate instrumental precision, a standard
483 solution with a concentration of 25 μg L^{-1} (for each enantiomer) was used.
484 Instrumental repeatability, expressed in terms of % RSD of Ac calculated from six
485 consecutive injections in one day, was found to be below 11%. Within-laboratory
486 reproducibility was calculated as % RSD of Ac calculated for three consecutive days (3

injections per day) obtaining values below 12%. The IDL and IQL, concentration level corresponding to a signal-to-noise of 3 and 10, respectively from injection of a standard solution successively diluted, were below 0.6 and 2.6 mg L^{-1} for all enantiomers, respectively.

On the other hand, analytical parameters of the SPE-chiral CE-DAD method were also evaluated and Tables 3 and 4 show the results obtained for river and sewage waters, respectively. Matrix matched calibration curves were shown to be linear, with $R^2 > 0.99$ for all enantiomers in both water samples. The existence of matrix effects was determined by comparing the slopes of the matrix-matched and the solvent-based standard calibration curves of each analyte. When comparing the slopes, it was observed that the slope values of the matrix-matched calibration curves were lower than the slopes of the solvent-based standard calibration curves, what indicates adverse influence of the matrix in the detector response. Therefore, matrix-matched calibration curves were used to quantify the target analytes in the samples, in order to compensate the errors associated with matrix effects. Precision was evaluated for river and seawage waters spiked at 2 and 100 μg L^{-1} each enantiomer and expressed in terms of RSD (%) of the Ac. In water samples, the repeatability and within-laboratory reproducibility of the procedures was good with RSD in the range of 1 - 9% and 6 - 11%, respectively, for all enantiomers. The MDL and MQL were between 0.4 – 0.6 μg L^{-1} and 1.3 -1.9 μg L^{-1}, respectively, for river and sewage waters.

510 *Real sample analysis*

511 In order to explore the analytical performance of the method in practical
512 applications, the extraction of Ate, Met, Pin and Prop was performed in river and
513 sewage waters. Table 3 shows recoveries obtained for all enantiomers in river water,
514 calculated according to equation [1] (see Experimental section). As it can be seen,
515 satisfactory recoveries in the range of 91 ± 1% – 98 ± 1% were achieved (n = 3). For
516 sewage water (Table 4), also good recoveries in the range of 86 ± 2% - 98 ± 1% were
517 achieved (n = 3). Results obtained indicate that the SPE-chiral CE-DAD method
518 developed has practical applications. In this sense, despite river and sewage waters
519 have high amount of organic matter, this fact has not produced interferences that
520 negatively affect the analytes recoveries.

521 Figure 4 shows the electropherograms obtained for tap water (spiked at 25 μg L^{-1}
522 each enantiomer), river water (unspiked) and sewage water (unspiked) registered at
523 200 nm. As it can be seen in the electropherogram of spiked tap water, the Rs
524 achieved were 1.45, 1.56 and 1.3 for Pin, Prop and Met enantiomers, respectively. For
525 Ate enantiomers, Rs was lower (in the range of 0.9-1) that is the minimum value
526 accepted for quantification, so it could be considered enough for quantitative
527 determination of these compounds in environmental samples. In the
528 electropherogram of river water peaks of β-blockers were not found, so the river
529 water analyzed was not contaminated with the target β-blockers at a concentration
530 level higher of the MDL (0.4 – 0.6 μg L^{-1}). In the electropherogram of sewage water
531 (WWTP effluent), peaks of Met enantiomers were found. The peaks identification was
532 based on the comparison of the migration times and DAD spectra of their peaks in

sewage water with those previously obtained in spiked water. For quantification purposes, the Ac values were subjected to correction with the recovery values established for them and then interpolated into their corresponding matrix matched calibration curve. The peaks found for Met correspond to a concentration of 10.7 and 9.9 μg L^{-1} for first and second enantiomer, respectively. In environmental analysis, two main descriptors are used to describe chiral signatures, the enantiomeric ratio (ER) and the enantiomeric fraction (EF) (Schuring 2013). ER described the ratio between the one enantiomer over the other, in these case the ER was 1.085. EF is the mole fraction of one enantiomer in a mixture, so for Met enantiomers EF1 was 0.52 and EF2 was 0.48 (values near to a racemic mixture). Results obtained in the sewage water analyzed confirmed the incomplete removal of Met during conventional wastewater treatment. These results are in agreement with those published by Souchier et al. (2016), that indicated the frequent presence of Met in influents and effluents of some WWTPs, with a *S*-Met enrichment in most cases (EF from 0.50 to 0.70) which extent was dependent on the WWTP.

Conclusions

As a summary, this work demonstrated that the n-alkyl-organic chain length played an important role in the behavior of functionalized SBA-15 as SPE sorbent. SBA-15-C8 was the most effective material for the extraction and preconcentration of chiral β-blockers in environmental water by SPE. The SBA-15-C8 based SPE-chiral CE-DAD method developed was successfully applied to chiral analysis of atenolol, metoprolol, pindolol

555 and propranolol in river and sewage water samples with satisfactory recoveries and a
556 preconcentration factor of 500 was achieved. The detection of metoprolol in sewage
557 water (WWTP effluent) with a concentration of 10.7 and 9.9 μg L^{-1} for first and second
558 enantiomer, respectively, emphasize the importance of enantioselectivity analyses for
559 environmental risk assessment and demonstrate that the developed method could
560 have good application prospects.
561

562 ## Conflicts of interest

563 There are no conflicts to declare.
564

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759

760 **Figure captions**

761 **Figure 1.**T EM images of SBA-15 (a) view through [100] axis and (b) through [001] axis.
762 (c) SEM image of SBA-15.

763 **Figure 2.** (a) N_2 adsorption-desorption isotherms and (b) pore size distribution of n-
764 alkyl-modified SBA-15.

765 **Figure 3.** Study of the breakthrough volume using SBA-15-C8 or SBA-15-C18 as SPE
766 sorbents. (a) 100 mg of SBA-15-C8, (b) 100 mg of SBA-15-C18, (c) 200 mg of SBA-15-C8
767 and (d) 200 mg of SBA-15-C18. Tap water spiked at 25 μg L^{-1} of each enantiomer.

Figure 4. Chiral separation obtained for pindolol (Pin), atenolol (Ate), propranolol (Pro) and metoprolol (Met) enantiomers, under optimized conditions, in river and sewage waters. BGE: phosphate buffer 50 mM at pH 2.5 and 1.25% (w/v) M-β-CD. Voltage: 20 kV, temperature: 20° C, sample injection: 50 mbar x 5.00 s.

Table 1. Textural properties and functionalization degree of SBA-15 silicas. S_{BET}: Brunauer, Emmett and Teller surface, Total pore volume were measure at relative P/P_0 =0.97, Pore diameter estimated by using the BJH (Barrett, Joyner and Halenda) model applied on the desorption branch of the isotherm, Functionalization degree = mmol of ligand per g of material obtained through the % C of the elemental analysis.

Table 2. Recoveries obtained for the target β-blockers with different sorbents. SPE conditions: 100 mg of sorbent and 100 mL of tap water spiked at 25 µg L^{-1} of each enantiomer. CE conditions: background electrolyte composed by phosphate buffer 50 mM at pH 2.5 and 1.25% (w/v) methyl-β-CD. Voltage: 20 kV, temperature: 30 °C, sample injection: 10 kV x 6 sec, Pin: Pindolol, Ate: Atenolol, Prop: Propranolol, Met: Metoprolol.

Table 3. Validation of SPE-chiral CE-DAD method for β-blockers in river water. Pin: Pindolol, Ate: Atenolol, Prop: Propranolol, Met: Metoprolol e.e.: each enantiomer Ac: Corrected peak area, MDL: Method detection limit, MQL: Method quantification limit.

Table 4. Validation of SPE-chiral CE-DAD method for β-blockers in sewage water. Pin: Pindolol, Ate: Atenolol, Prop: Propranolol, Met: Metoprolol e.e.: each enantiomer Ac: Corrected peak area, MDL: Method detection limit, MQL: Method quantification limit.

789 **Table 5. Comparative study of reported methods for the SPE extraction of β-blockers**
790 **from waters**. Ace: Acebutolol, Al: Alprenolol, Ate: Atenolol, Bet: Betaxolol, Bis:
791 Bisoprolol, Car: Carazolol, Cel: Celiprolol, Lab: Labetalol, Met: Metoprolol, Nad:
792 Nadolol, Neb: Nebivolol, Ox: Oxprenolol, Pin: Pindolol, Prop: Propanolol, Sot: Sotalol,
793 PF: preconcentration factor.

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807 **Table 1.** Textural properties and functionalization degree of SBA-15 silicas

Material	S_{BET} ($m^2\ g^{-1}$)	Pore volume ($cm^3\ g^{-1}$)	Pore diameter (Å)	L_0 (mmol g^{-1})
SBA-15	764	0.80	55.5	-
SBA-15-C3	653	0.67	41.0	0.45
SBA-15-C8	613	0.68	50.1	0.42
SBA-15-C18	650	0.70	49.2	0.24

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809 S_{BET}: Brunauer, Emmett and Teller surface, Total pore volume were measure at relative P/P_0 =0.97,

810 Pore diameter estimated by using the BJH (Barrett, Joyner and Halenda) model applied on the
811 desorption branch of the isotherm, Functionalization degree = mmol of ligand per g of material obtained
812 through the % C of the elemental analysis

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819 **Table 2.** Recoveries obtained for the target β-blockers with different sorbents

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	Analytes , Recovery ± SD (%), n = 3							
Material	1-Pin	2-Pin	*S*-Ate	*R*-Ate	*S* -Prop	*R* -Prop	1-Met	2-Met
SBA-15	68 ± 5	61 ± 4	56 ± 5	49 ± 3	66 ± 4	55 ± 2	70 ± 2	55 ± 2
SBA-15-C3	51 ± 2	43 ± 1	51 ± 5	57 ± 4	65 ± 4	50 ± 1	75 ± 7	67 ± 3
SBA-15-C8	91 ± 7	87 ± 2	97 ± 2	89 ± 2	79 ± 4	79 ± 2	93 ± 7	86 ± 5
SBA-15-C18	101 ± 6	101 ± 6	104 ± 8	104 ± 8	100 ± 6	100 ± 6	107 ± 5	96 ± 5

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822 SPE conditions: 100 mg of sorbent and 100 mL of tap water spiked at 25 µg L^{-1} of each enantiomer. CE
823 conditions: background electrolyte composed by phosphate buffer 50 mM at pH 2.5 and 1.25% (w/v)
824 methyl-β-CD. Voltage: 20 kV, temperature: 30 °C, sample injection: 10 kV x 6 sec, Pin: Pindolol, Ate:
825 Atenolol, Prop: Propranolol, Met: Metoprolol.

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Table 3. Validation of SPE-chiral CE-DAD method for β-blockers in river water

Analyte	Matrix matched calibration; R^2	Recovery ± SD (%), n = 3		Repeatability RSD Ac (%)		Within-laboratory reproducibility RSD Ac (%)	MDL (µg L^{-1})	MQL (µg L^{-1})
		2 µg L^{-1} e.e.	100 µg L^{-1} e.e.	2 µg L^{-1}e.e.	100 µg L^{-1} e.e.			
1-Pin	y = 0.317 x + 0.8; 0.998	98 ± 1	91 ± 1	4	5	7	0.5	1.7
2-Pin	y = 0.350 x + 0.6; 0.995	98 ± 1	92 ± 1	2	2	8	0.5	1.7
S-Ate	y = 0.139 x + 0.8; 0.997	94 ± 2	91 ± 1	4	8	7	0.5	1.8
R-Ate	y = 0.392 x + 0.6; 0.997	94 ± 2	95 ± 2	4	9	9	0.5	1.8
S-Prop	y = 0.262 x + 0.4; 0.992	91 ± 1	95 ± 1	6	3	9	0.4	1.3
R-Prop	y = 0.275 x + 0.2; 0.995	97 ± 2	97 ± 1	4	1	10	0.4	1.3
1-Met	y = 0.095 x + 1.0; 0.998	93 ± 2	91 ± 1	4	6	10	0.6	1.9
2-Met	y = 0.283 x + 0.2; 0.999	97 ± 1	94 ± 2	2	3	11	0.6	1.9

Pin: Pindolol, Ate: Atenolol, Prop: Propranolol, Met: Metoprolol e.e.: each enantiomer Ac: Corrected peak area, MDL: Method detection limit, MQL: Method quantification limit

Table 4. Validation of SPE-chiral CE-DAD method for β-blockers in sewage water

Analyte	Matrix matched calibration; R^2	Recovery ± SD (%), n = 3		Repeatability RSD Ac (%)		Within-laboratory reproducibility RSD Ac (%)	MDL (µg L^{-1})	MQL (µg L^{-1})
		2 µg L^{-1} e.e.	100 µg L^{-1} e.e.	2 µg L^{-1}e.e.	100 µg L^{-1} e.e.			
1-Pin	y = 0.270 x + 0.8; 0.994	94 ± 2	89 ± 2	2	3	6	0.4	1.3
2-Pin	y = 0.276 x + 0.9; 0.993	93 ± 3	92 ± 2	7	4	7	0.4	1.3
S-Ate	y = 0.149 x + 0.8; 0.995	91 ± 4	87 ± 5	5	6	7	0.6	1.9
R-Ate	y = 0.413 x - 0.2; 0.998	89 ± 2	90 ± 4	8	4	8	0.6	1.9
S-Prop	y = 0.274 x + 0.2; 0.999	91 ± 4	92 ± 3	3	4	8	0.4	1.4
R-Prop	y = 0.287 x + 0.2; 0.999	86 ± 2	98 ± 1	6	1	7	0.4	1.4
1-Met	y = 0.127 x + 0.5; 0.998	89 ± 1	91 ± 6	7	6	10	0.5	1.8
2-Met	y = 0.316 x - 0.5; 0.999	93 ± 2	92 ± 7	5	7	10	0.5	1.8

Pin: Pindolol, Ate: Atenolol, Prop: Propranolol, Met: Metoprolol e.e.: each enantiomer Ac: Corrected peak area, MDL: Method detection limit, MQL: Method quantification limit

843 **Table 5.** Comparative study of reported methods for the SPE extraction of β-blockers from waters

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β-blockers analysed	SPE sorbent	Sample	Recovery (%)	PF	Method	Comments	Ref.
Ace, Ate, Nad, Met, Prop	Strata X-C® (200 mg)	Tap water (250 mL)	67-125	83	CG-MS	Water sample adjusted at pH: 3. Not chiral analysis.	Caban et al. (2015)
Ate, Prop, Met	Oasis MCX® (60 mg)	River water (1000 mL)	60-110	2000	UPLC–MS/MS	Water sample adjusted at pH: 2.5. Not chiral analysis. Internal standard calibration.	Kasprzyk-Hordern et al. (2007)
Ace, Al, Ate, Bis, Labe, Met, Nad, Pin, Prop, Sot, Tim	Oasis MCX® (150 mg)	Effluent wastewater (250 mL)	91-108	250	LC–MS/MS	Water sample adjusted at pH: 3. Not chiral analysis. Matrix matched calibration	Lee et al. (2007)
Ace, Al, Ate, Bis, Met, Nad, Pin, Prop, Sot, Tim	Oasis MCX® (60 mg)	Influent wastewater (400 mL)	10-68	1000	HPLC-MS	Water sample acidified. Not chiral analysis. Matrix matched calibration	Piram et al. (2008)
Ate, Prop, Met	Oasis MCX® (60 mg)	Effluent wastewater (250 mL) Influent wastewater (250 mL) Surface water (1000 mL)	17-84 14-76 40-90	500 500 2000	UPLC–ESI-MS/MS	Water sample adjusted at pH: 2. Not chiral analysis. Internal standard calibration.	Kasprzyk-Hordern et al. (2008)

Ate, Met	Oasis MCX® (60 mg)	River water (150 mL) Effluent wastewater (100 mL)	71-74 82-87	750 500	LC–ESI-MS/MS	Water sample adjusted at pH: 2. Not chiral analysis. Internal standard calibration.	Al-Odaini et al. (2010)
Ate, Nad, Met, Bis, Bet	Oasis MCX® (150 mg)	Ground water (100 mL)	79-114	200	LC-TOF-MS	Water sample adjusted at pH: 3. Not chiral analysis.	Galera et al. (2011)
Ace, Ate, Met, Prop, Tim, Nad, Ox, Pin, Al	Oasis MCX® (60 mg)	Effluent wastewater (200 mL)	25-97	200	LC–ESI-MS/MS	Water sample pH not specified. Not chiral analysis. Internal standard calibration.	Salem et al. (2012)
Ace, Ate, Met, Nad, Pin, Prop	Strata X® (200 mg)	Effluent wastewater (250 mL)	62-96 28-92	50	GC-FID or GC-MS	Water sample pH not adjusted (pH: 8). Not chiral analysis. Matrix matched calibration	Caban et al. (2012)
Ace, Ate, Nad, Met, Prop	Strata X® (200 mg)	Tap water (250 mL)	63-113	50	CG-MS	Water sample pH not specified. Not chiral analysis.	Caban et al. (2015)
Ate, Prop	Oasis HLB® (200 mg)	Effluent wastewater (100 mL)	87-97	100	LC–MS/MS	Water sample adjusted at pH: 7. Not chiral analysis. Matrix matched calibration	Gómez et al. (2006)

Ox, Met, Prop, Bis, Bet	Oasis HLB® (60 mg)	River water (500 mL)	94-103	500	CG-MS	Water sample adjusted at pH: 7.5. Not chiral analysis.	Miège et al. (2006)
Ace, Ate, Met, Sot	Oasis HLB® (60 mg)	Effluent wastewater (250 mL) Influent wastewater (100 mL) Surface water (500 mL) Ground water (1000 mL)	78-101 64-108 62-105 76-93	500 200 1000 2000	LC-MS/MS	Water sample pH not specified. Not chiral analysis. Internal standard calibration.	Vieno et al. (2006)
Ate, Met, Nad, Pin, Prop, Sot	Oasis HLB® (60 mg)	Effluent wastewater (500 mL) Influent wastewater (100 mL)	50-115 56-110	100 20	HPLC-MS/MS	Water sample pH not adjusted (pH: 7). Chiral separation. Matrix matched calibration	MacLeod et al. (2007)
Neb, Met, Ate, Bis	Oasis HLB® (200 mg)	Surface water (50 mL) Influent wastewater (50 mL)	73-101 65-104	100	HILIC-MS/MS	Water sample adjusted at pH: 7. Not chiral analysis. Internal standard calibration.	van Nuijs et al. (2010)
Met, Prop, Ate	Oasis HLB® (500 mg)	Effluent wastewater (200 mL) Influent wastewater (200 mL)	69-102 69-86	200	UPLC–MS/MS	Water sample adjusted at pH: 2.5. Not chiral analysis	Yuan et al. (2014)

Met, Prop, Sot	Oasis HLB® (not specified)	Wastewater (50 mL)	21-150	100	LC–MS/MS	Water sample pH not specified. Chiral analysis. Internal standard calibration	Evans et al. (2015)
Ate, Sot, Pin, Tim, Met, Car, Prop, Bet	MIP4SPE™ (not specified)	Effluent wastewater (25 mL) Influent wastewater (25 mL)	50-110 40-112	25	LC–QqLIT-MS	Water sample neutral pH (not adjusted). Not chiral analysis. Internal standard calibration.	Gros et al. (2008)
Prop	SupelMIP™ (not specified)	River water (100 mL)	97	100	HPLC-DAD	Water sample neutral pH (not adjusted). Chiral separation. Matrix matched calibration	Morante-Zarcero and Sierra (2012a)
Met, Pin, Prop, Ate	SupelMIP™ (not specified)	River water (100 mL)	97	100	HPLC-DAD	Water sample neutral pH (not specified). Simultaneous chiral separation. Matrix matched calibration	Morante-Zarcero and Sierra (2012b)
Met, Prop, Bis, Bet, Nad, Car, Tim	C18-end capped (500 mg)	Ground water (1000 mL)	26-125	250	CG-MS	Water sample adjusted at pH: 7.5. Not chiral analysis.	Ternes et al. (1998)
Ate, Met, Nad, Bet, Bis, Car, Cel, Prop, Sot	Bakerbond C18® (not specified)	Effluent wastewater (100 mL) Influent wastewater (200 mL) River water (1000 mL)	31-84 15-49 36-92	100 200 1000	LC-MS/MS	Water sample pH not adjusted. Not chiral analysis. Matrix matched calibration	Scheurer et al. (2010)

Ace, Ate, Nad, Met, Prop	Strata C18-EC® (200 mg)	Tap water (250 mL)	20-81	50	CG-MS	Water sample pH not adjusted. Not chiral analysis.	Caban et al. (2015)
Ate, Nad, Pin, Tim, Bis, Bet	MCM-41	River water (100 mL)	67-98	100	Micro-LC-MS/MS	Water sample adjusted at pH: 2. Not chiral analysis. Standard addition calibration.	Dahane et al (2016)
Pin, Ate, Prop, Met	SBA-15-C18 (100 mg)	Tap water (150 mL) River water (150 mL) Ground water (150 mL)	72 - 118 66-106 62-105	300	CE-DAD	Water sample pH not adjusted. Simultaneous chiral separation. Matrix matched calibration	Silva et al. (2017)
Pin, Ate, Prop, Met	SBA-15-C8 (200 mg)	River water (250 mL) Effluent wastewater (250 mL)	91– 98 86 – 98	500	CE-DAD	Water sample pH not adjusted. Simultaneous chiral separation. Matrix matched calibration	This work

Ace: Acebutolol, Al: Alprenolol, Ate: Atenolol, Bet: Betaxolol, Bis: Bisoprolol, Car: Carazolol, Cel: Celiprolol, Lab: Labetalol, Met: Metoprolol, Nad: Nadolol, Neb: Nebivolol, Ox: Oxprenolol, Pin: Pindolol, Prop: Propanolol, Sot: Sotalol, PF: preconcentration factor

Figure 1

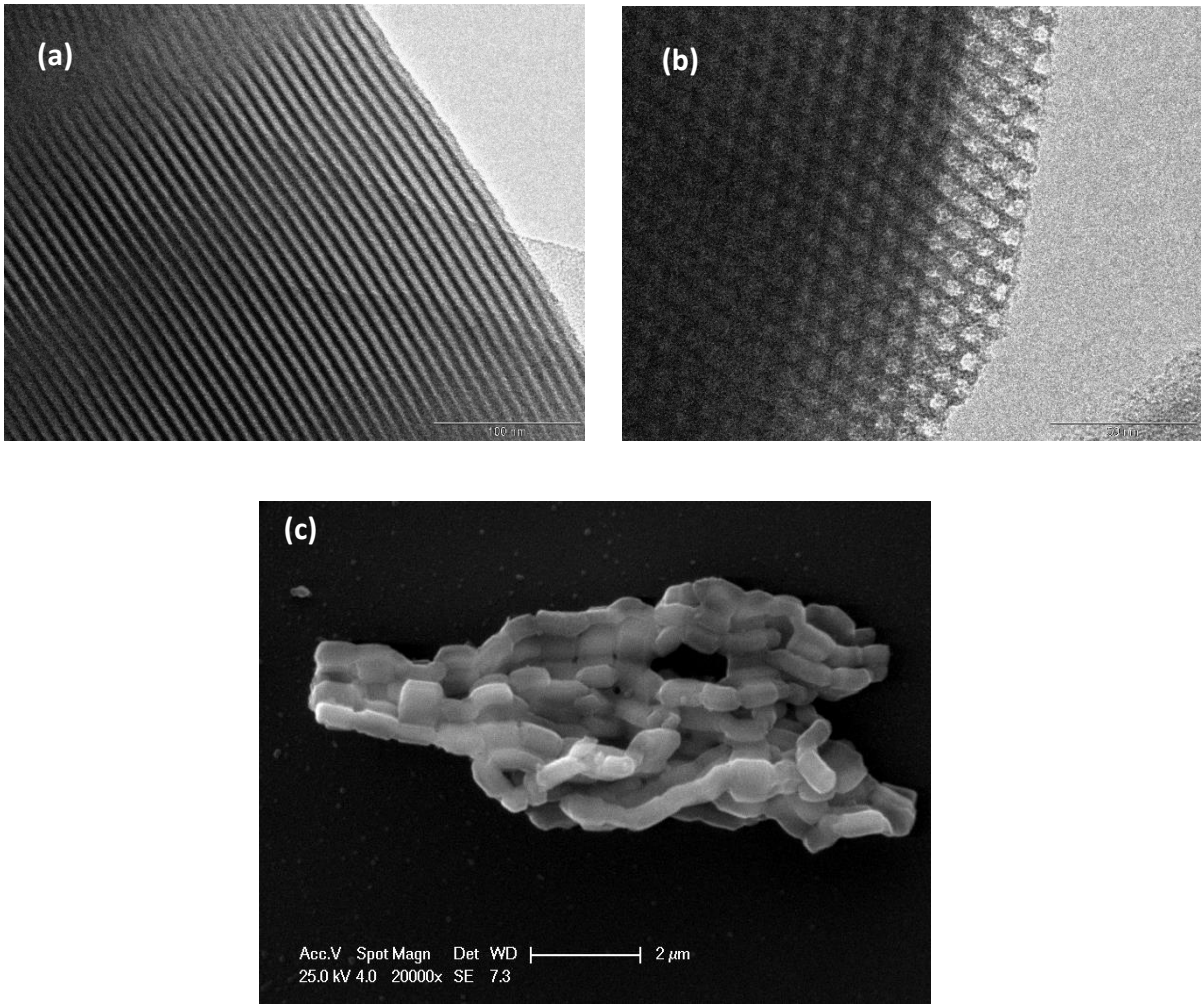
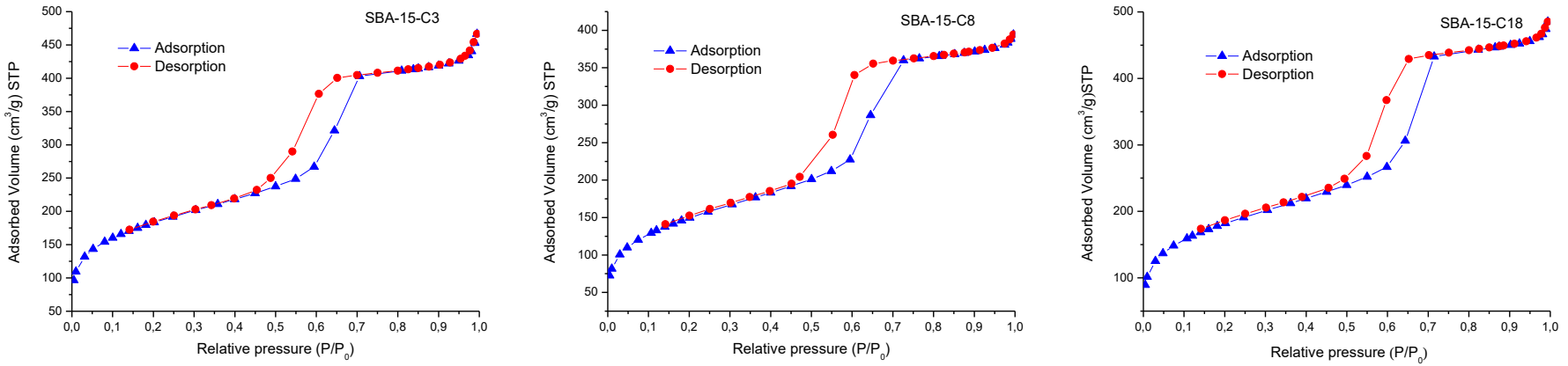


Figure 2

(a)


SBA-15-C3
Adsorption
Desorption
Adsorbed Volume (cm³/g) STP
Relative pressure (P/P0)
SBA-15-C8
Adsorption
Desorption
Adsorbed Volume (cm³/g) STP
Relative pressure (P/P0)
SBA-15-C18
Adsorption
Desorption
Adsorbed Volume (cm³/g)STP
Relative pressure (P/P0)

(b)

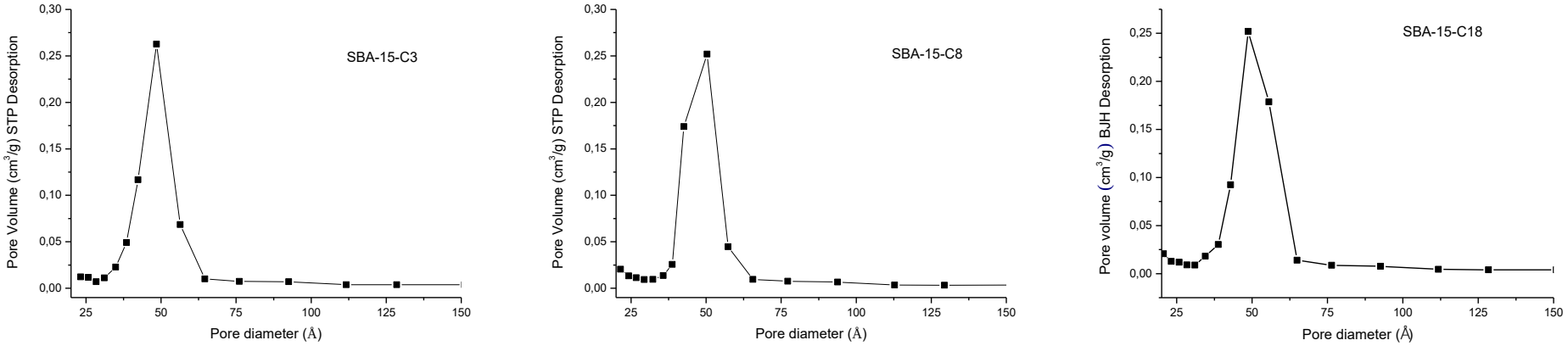

SBA-15-C3
Pore Volume (cm³/g) STP Desorption
Pore diameter (Å)
SBA-15-C8
Pore Volume (cm³/g) STP Desorption
Pore diameter (Å)
SBA-15-C18
Pore volume (cm³/g) BJH Desorption
Pore diameter (Å)

Figure 3

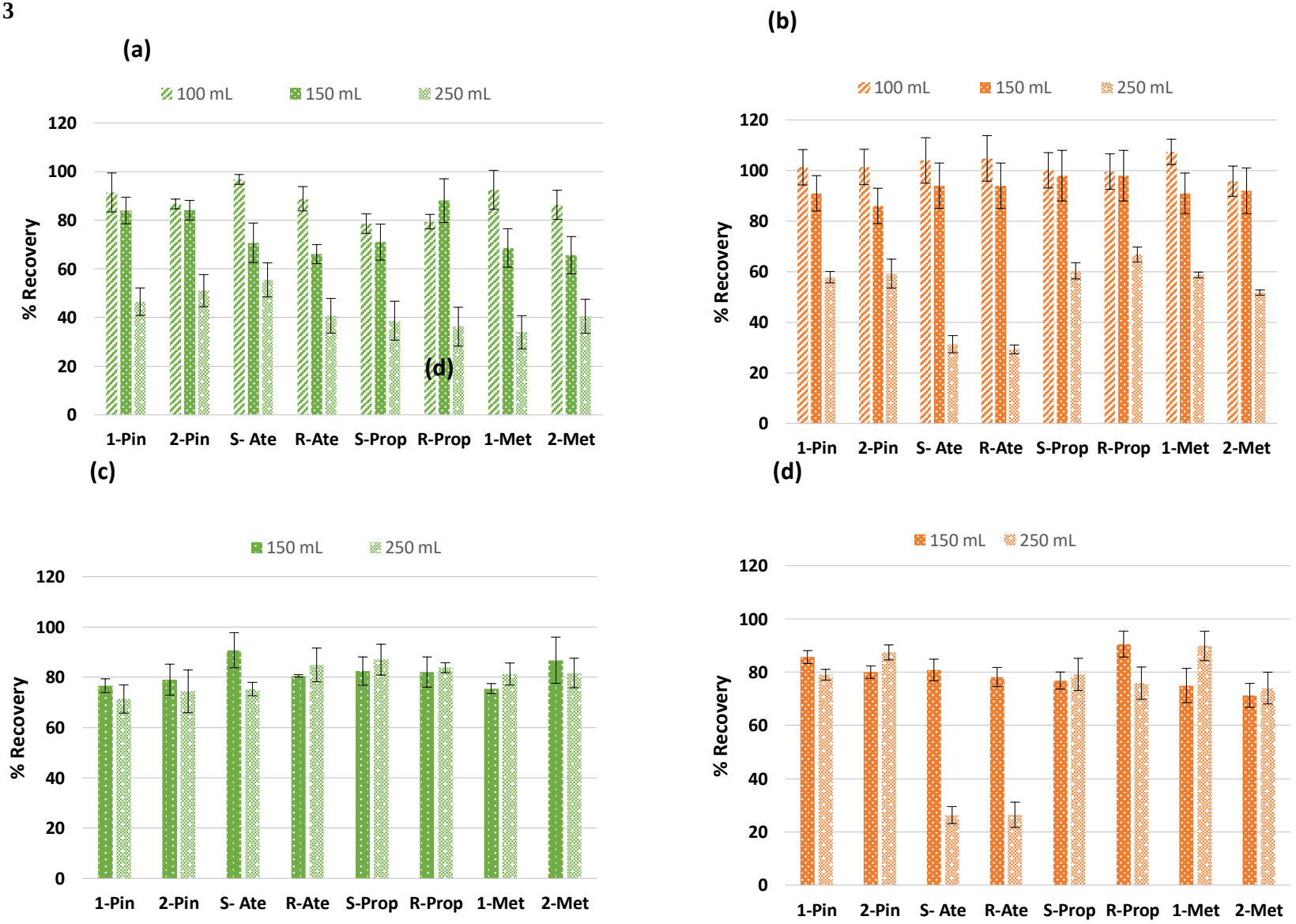
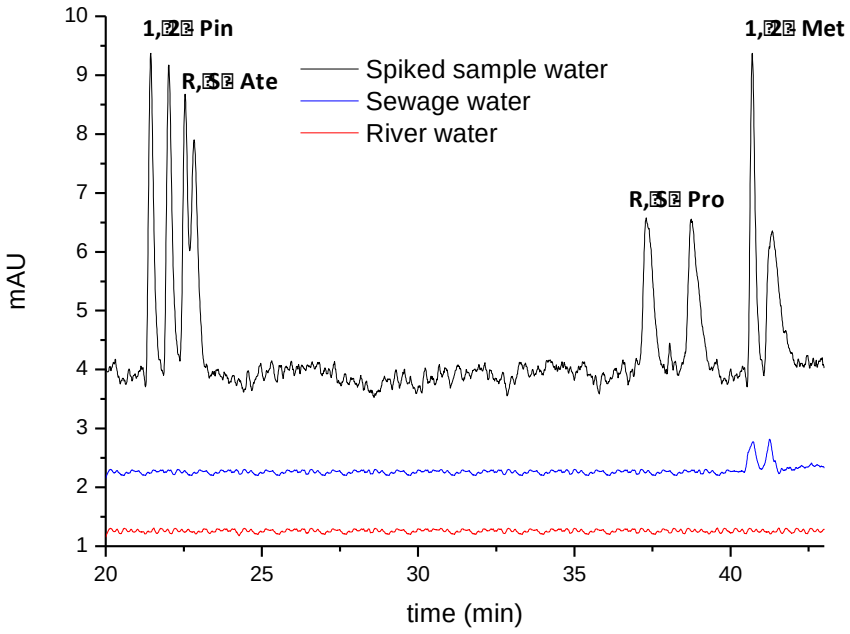

(a)
100 mL
150 mL
250 mL
% Recovery
120
100
80
60
40
20
0
1-Pin
2-Pin
S- Ate
R-Ate
S-Prop
R-Prop
1-Met
2-Met
(d)
(b)
100 mL
150 mL
250 mL
% Recovery
120
100
80
60
40
20
0
1-Pin
2-Pin
S- Ate
R-Ate
S-Prop
R-Prop
1-Met
2-Met
(c)
150 mL
250 mL
% Recovery
120
100
80
60
40
20
0
1-Pin
2-Pin
S- Ate
R-Ate
S-Prop
R-Prop
1-Met
2-Met
(d)
150 mL
250 mL
% Recovery
120
100
80
60
40
20
0
1-Pin
2-Pin
S- Ate
R-Ate
S-Prop
R-Prop
1-Met
2-Met

Figure 4


1,2-Pin
R,S-Ate
1,2-Met
R,S-Pro
Spiked sample water
Sewage water
River water
mAU
time (min)
10
9
8
7
6
5
4
3
2
1
20
25
30
35
40