Science of the Total Environment 494–495 (2014) 49–57

Contents lists available at ScienceDirect

Science of the Total Environment

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

Stereoisomeric profiling of drugs of abuse and pharmaceuticals in wastewaters of Valencia (Spain)

Pablo Vazquez-Roig [a], Barbara Kasprzyk-Hordern [b,*], Cristina Blasco [a], Yolanda Picó [a]

[a] Laboratory of Nutrition and Bromatology, Faculty of Pharmacy, University of Valencia, Av. Vicent Andrés s/n, 46100 Burjassot, Valencia, Spain
[b] University of Bath, Department of Chemistry, Faculty of Science, Bath BA2 7AY, UK

HIGHLIGHTS

- Enantiomeric profiling of chiral drugs was undertaken at three WWTPs.
- Degradation efficiency of WWTPs was compound and enantiomer dependent.
- Atenolol was enriched with either *S*(−)- or *R*(+)-enantiomer in different WWTPs.
- Amphetamine and MDMA were enriched with *R*(−)-enantiomers
- 1*S*,2*S*(+)-pseudoephedrine was more readily degradable than its diastereomer

ARTICLE INFO

Article history:
Received 31 May 2014
Received in revised form 21 June 2014
Accepted 21 June 2014
Available online xxxx

Editor: D. Barcelo

Keywords:
Pharmaceuticals
Drugs of abuse
Stereoisomers
Enantiomers
Wastewater
Chiral drugs

ABSTRACT

The enantiomeric and diastereomeric profiling of chiral pharmaceuticals (ephedrine, norephedrine, atenolol and venlafaxine) and illicit drugs (amphetamine, methamphetamine, 3,4-methylenedioxyamphetamine (MDA), 3,4-methylenedioxy-*N*-methylamphetamine (MDMA) and 3,4-methylenedioxy-*N*-ethylamphetamine (MDEA)) was undertaken over a period of fourteen consecutive days in three wastewater treatment plants (WWTPs) in the city of Valencia, Spain. Degradation efficiency of WWTPs was found to be compound and enantiomer dependent. Selective enantiomer enrichment was observed for several target analytes. Amphetamine and MDMA were enriched with *R*(−)-enantiomers. 1*S*,2*S*(+)-pseudoephedrine was found to be more readily degradable during activated sludge treatment than its diastereomer 1*R*,2*S*(−)-ephedrine. Atenolol underwent enrichment with either *S*(−)- or *R*(+)-enantiomer in different WWTPs. This unexpected enantiomeric variation in the stereoselective degradation of atenolol could be attributed to different processes utilized during activated sludge treatment. The application of (enantiomeric) profiling of wastewater revealed usage patterns of chiral drugs in the Valencia region.

1. Introduction

More than 50% of pharmaceuticals and illicit drugs currently in use are chiral (Lien et al., 2006). Although they are usually manufactured as racemic compounds, they can be stereoselectively degraded in humans (for example as a result of stereoselective metabolism) and/or during wastewater treatment as a result of microbial processes. Since activity and toxicity of these compounds are often isomer-dependent (Kasprzyk-Hordern, 2010), it is of importance to understand the influence of wastewater treatment processes in the selective degradation of chiral drugs in order to improve the performance of wastewater treatment plants (WWTPs) and to protect the receiving aquatic environment. Fluoxetine, for example, is one of the most toxic human pharmaceuticals reported so far. Its ecotoxicity is currently assessed for the racemate (Kasprzyk-Hordern, 2010). However, recent research indicates that toxic effects of fluoxetine are enantiomer dependent: *S*(+)-fluoxetine is 9.4 times more toxic to *Pimephales promelas* than *R*(−)-fluoxetine (Stanley et al., 2007). Specific inter-species toxic effects also exist among enantiomers; for example, *S*(−)-propranolol has a higher chronic toxicity to *Fathead Minnows* than its enantiomer, but the opposite is true in *Daphnia magna* (Nikolai et al., 2006).

There is a lack of information on the stereoselective and/or stereospecific fate and effects of chiral pharmaceuticals and illicit drugs in the environment. Although chiral HPLC methods have been extensively used for stereoisomer separation of drugs in pharmaceutical preparations they are usually not directly amenable to the analysis of chiral drugs in complex environmental matrices and at trace concentrations (Evans and Kasprzyk-Hordern, 2014). Recently, new HPLC methods using chiral columns packed with antibiotics or proteins coupled with

* Corresponding author. Tel.: +44 1225 385013; fax: +44 1225 386231.
E-mail address: B.Kasprzyk-Hordern@bath.ac.uk (B. Kasprzyk-Hordern).

http://dx.doi.org/10.1016/j.scitotenv.2014.06.098
0048-9697/

tandem mass spectrometry have been successfully applied in the analysis of several chiral drugs in WWTPs and/or surface waters (Bagnall et al., 2012; Kasprzyk-Hordern and Baker, 2012b; MacLeod and Wong, 2010; Nikolai et al., 2006). Among these chiral drugs are: atenolol, metoprolol, fluoxetine, venlafaxine, ibuprofen, ketoprofen, naproxen, amphetamine, methamphetamine and ephedrine (MacLeod and Wong, 2010; Nikolai et al., 2006; Barclay et al., 2012; Hashim and Khan, 2011; Fono and Sedlak, 2005; Buser et al., 1999; López-Serna et al., 2013; Kasprzyk-Hordern and Baker., 2012b).

This paper presents for the first time the results of a two week study of three WWTPs in Valencia City and surroundings aiming at estimating the occurrence and stereoselective fate of five chiral pharmaceuticals: (ephedrine, pseudoephedrine, norephedrine, atenolol and venlafaxine) and five illicit drugs (amphetamine, methamphetamine, 3,4-methylenedioxyamphetamine (MDA), 3,4-methylenedioxymethamphetamine (MDMA) and 3,4-methylenedioxyethamphetamine (MDEA)).

Table 1
Operational parameters of studied WWTPs.

Characteristics	Pinedo-I	Pinedo-II	Quart-Benager
Coordinates UTM (ETRS 89 zone 30N)	X:728552 Y: 4368031 Z: 5	X:728371 Y: 4368153 Z: 5	X:722456 Y: 4370419 Z: 22
Population served (thousands)	351,198	942,774	166,942
Flows (m^3/day)	100,602	242,580	35,903
Wastewater (% industrial/% domestic)	0/100	0/100	60/40
Treatment	AS	AS/N removal	AS/P removal
Average daily sewage flow (m^3/d)	106,537	236,396	37,998
Designed treatment capacity (m^3/d)	124,800	200,000	60,000
Influent BOD (mg/L)[a]	248.2	263.7	318.1

AS: Activated Sludge; N: Nitrogen; P: phosphorus; BOD: Biological Oxygen Demand.
[a] Annual average.

2. Materials and methods

2.1. Sampling

In a sampling campaign of two weeks (April 17–May 1 of 2012), influent and effluent 24 h-composite samples (time-proportional mode) were collected in three WWTPs serving the city of Valencia (Spain) and the surrounding towns: Pinedo-I, Pinedo-II and Quart-Benager (Fig. 1). These WWTPs have tertiary treatments but they differ in the type of technology utilized for secondary treatment: Pinedo-I utilizes only activated sludge, Pinedo-II utilizes activated sludge and phosphorus removal, and Quart-Benager utilizes activated sludge and nitrogen removal (see operational details of studied WWTPs in Table 1). Pinedo-I treats only wastewaters of the city of Valencia. Pinedo-II is the biggest and serves Valencia and surrounding towns. It has a very complete primary treatment and a secondary treatment, digestion and dehydration of sludge (200,000 m^3/day). This installation is complemented by a treatment with Densadeg, filters, sand and UV disinfection with a capacity of 350,000 m^3/day (100,000 m^3/day for regeneration of the ecological flow of Natural Park) and 250,000 m^3/day for irrigation. Quart-Benager serves the towns of the industrial belt of Valencia.

24 h-composite samples were taken in 2 L plastic bottles (polyethylene) with Teflon protected caps and were transported to the laboratory for their immediate analysis. If this was not possible, the samples were frozen at −20 °C until analysis to prevent degradation of the target residues.

2.2. Chemicals

All reference standards (±)-amphetamine (AMP), (±)-methamphetamine (MAMP), (±)-MDA, (±)-MDMA, (±)-MDEA, (−)-ephedrine (EPH), (+)-pseudoephedrine (PEPH), (±)-norephedrine (NOR), (±)-atenolol (ATE), and (±)-venlafaxine (VEN) were purchased from LGC Standards (Teddington, UK) and Sigma-Aldrich (Gillingham, UK). The surrogate standard (SS) (±)-MDA-d5 was added to the samples before solid-phase extraction (SPE). Internal standards (IS): (±)-methamphetamine-d5, (±)-MDMA-d5, (±)-MDEA-d5 and (±)-atenolol-d7

Fig. 1. Sampling locations in Valencia.