Analysis of cocaine and nicotine metabolites in wastewater by liquid chromatography–tandem mass spectrometry. Cross abuse index patterns on a major community

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HIGHLIGHTS

- Cross abuse index patterns on licit and illicit drugs by means of sewage assessment
- Data about cocaine and nicotine consumption from a Portuguese population
- Co-evaluation of cocaine abuse estimates based on local drug purity levels

ARTICLE INFO

ABSTRACT

A method based on sample preparation by solid phase extraction and analysis by liquid chromatography and mass spectrometry was validated and used for simultaneous analysis of cocaine, benzoylecgonine and cotinine in samples collected at the major wastewater treatment plant in the city of Lisbon. The aim was to estimate the consumption of both cocaine and nicotine in this community and establish an index involving both drugs supported by the relevance of nicotine as a significant anthropogenic marker. The study was made on two different weekdays during a month in order to evaluate patterns of consumption outside weekends. Cocaine and nicotine ingestion levels were back-calculated and expressed as mass of pure drugs consumed per day and per 1000 inhabitants (mean: 0.604 g and 5.860 g respectively). Cocaine was also expressed on the basis of local drug purity levels (33.7%) with a corresponding increase on dose assessments, and community drug abuse profiles. The authors sustain that this approach should always be included in drug studies of this kind allowing a better drug abuse assessment. No significant different patterns of consumption were obtained during the working days studied with the exception of one case coincident with a national holiday that showed an increased typical profile found on other non-working day studies, namely weekends. A fairly significant relationship was found between nicotine and cocaine consumption that should be further evaluated in future studies. Pharmacokinetic considerations were made and proposed for cocaine assessment based on the impact on back calculations after common simultaneous consumption of cocaine and ethanol.

1. Introduction

The analysis of sewage drug biomarkers (SDBs) has recently been established as a complementary approach for monitoring patterns and trends of illicit drug use in communities (Banta-Green et al., 2009; Berset et al., 2010; Castiglioni and Zuccato, 2011; Irvine et al., 2011; Karolak et al., 2010; Metcalfe et al., 2010; Reid et al., 2011; Terzic et al., 2010; Van Nuijs et al., 2009a; Zuccato et al., 2008). In 2012 the

first harmonized study involving drug wastewater analysis in 19 European cities has been published (Thomas et al., 2012). Moreover, in 2013, using data from this study, a critical review was made to integrally address the uncertainties associated with the estimations of community drug use through sewage analysis, proposing a set of best practice requirements (Castiglioni et al., 2013).

Cocaine (COC) remains the second most commonly used illicit drug in Europe, although prevalence levels and trends differ considerably between countries (EMCDDA, 2012). In Portugal results from the II National Population Survey on Psychoactive Substances in the Portuguese Population (15–64 years of age), the latest national survey available dated from 2007, indicate that alcohol and tobacco are the licit

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substances preferably used by the Portuguese population, and cannabis, cocaine and ecstasy, the illicit substances with lifetime prevalence respectively of 11.7%, 1.9% and 1.3% (IDT, 2011). The prevalence of cocaine use in the last 30 days/12 months has increased from 0.1/0.3 (year 2001) to 0.3/0.6 (year 2007), which was a strong incentive for selecting cocaine as the drug of choice for this sewage study in the capital of Portugal, analyzing the influents of the main wastewater treatment plant (WWTP) in the city of Lisbon, Alcântara (Fig. 1).

The aims of this work were (1) to assess the consumption of both cocaine and nicotine through the quantification of cocaine (COC), benzoylecgonine (BE) and cotinine (CT) in the influents of a major urban wastewater treatment plant, by the means of an implemented and validated liquid chromatography–tandem mass spectrometry (LC–MS/MS) analytical method; (2) to establish a relationship index between tobacco and cocaine consumption and (3) to propose a complementary data evaluation for better qualification of epidemiological studies by converting the number of cocaine doses adjusted to the average local purity levels of cocaine. This approach will produce a more realistic view on the extent of drug consumption and trafficking and is proposed as another index to be included on every sewage drug studies.

2. Material and methods

2.1. Reagents and materials

LC–MS grade acetonitrile (ACN) and methanol (MeOH) were obtained from Scharlau® (Scharlau S.L., Sentmenat, Spain). Analytical grade 37% hydrochloric acid (HCl), ammonium hydroxide (NH_4OH) and ammonium acetate (CH_3COONH_4) were purchased from Merck (Darmstadt, Germany) and VWR (Lisbon, Portugal). Ultra-pure water (18.2 MΩ·cm) was obtained from a Millipore-Direct Q3 UV system (Millipore®, Bedford, MA, USA). Individual methanolic or acetonitrile solutions (1 mg/mL) of cocaine (COC), benzoylecgonine (BE), cotinine (CT) and their deuterated analogs ($COC-d_3$, $BE-d_3$, $CT-d_3$), were of analytical grade (purity > 98%) and were purchased from Cerilliant (Round Rock, TX, USA).

Oasis HLB (500 mg, 6 mL) SPE cartridges were acquired from Waters (Barcelona, Spain) and a Waters Sep-Pak solid-phase extraction (SPE) Vacuum Manifold with 12 ports and dry vacuum system was also used in the SPE procedure.

2.2. Sample site and sample collection

Influent samples were collected from the main wastewater treatment plant (Alcântara-ETAR) in the region of Lisbon, Portugal. Samples were collected twice a week (Tuesday and Thursday) during four weeks, between the months of October and November 2011. Wastewater treatment plant (WWTP) samples were 24-h composite samples starting at midnight, collected on a time proportional sampling mode (120 min) and kept refrigerated during that process. The Alcântara WWTP serves the west part (A) and the center (A1) of the city of Lisbon (Fig. 1) and two major suburban areas — Amadora and Oeiras. The WWTP is dimensioned to an estimate design capacity of 756,000 inhabitants with a flow processing capability of 6.6 m^3/s. This system has started functioning in 1989 with a primary treatment and chlorine disinfection. Since October 2009 several major benefits have been introduced including biological treatment and ultraviolet irradiation allowing possible reuse of outflows both internal and external. During the study the total volume treated by the WWTP reached on average 123,000 m^3/day.

2.2.1. Sample preparation

All 24 h WWTP samples were transferred into 1 L polyethylene bottles, adjusted to pH = 2 with 37% HCl and stored at −20 °C until analysis. These conditions have been described as granting negligible degradation of the cocaine and metabolites (Gheorghe et al., 2008)
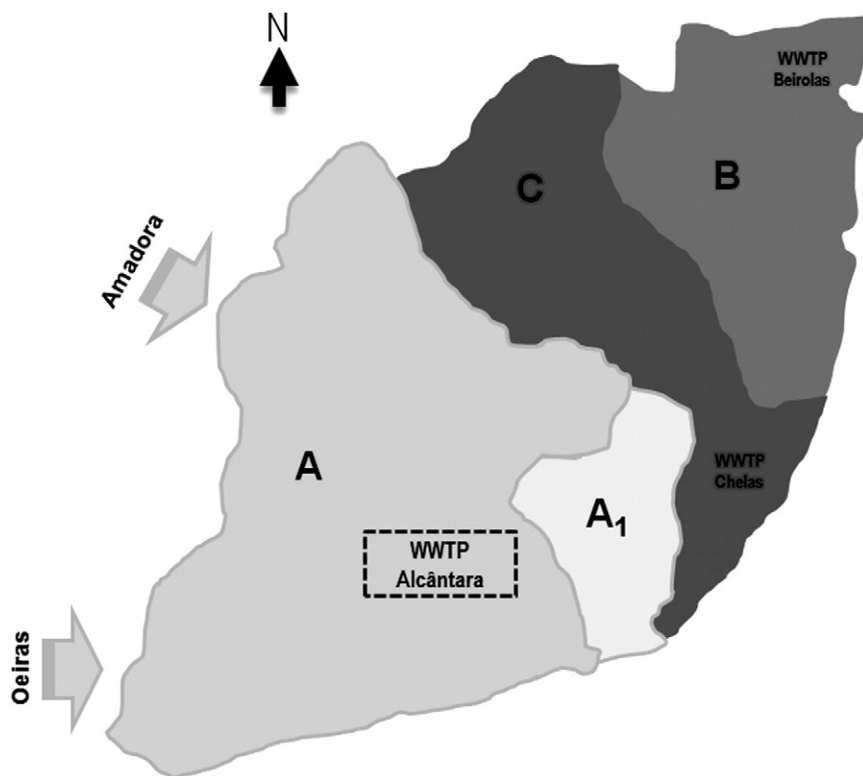

Fig. 1. Lisbon WWTP's covering areas: A (west), A1 (center), Amadora and Oeiras (major suburban areas with partial WW contributions) — Alcântara WWTP (study); B — Beirolas WWTP; C — Chelas WWTP.