Drugs of abuse and their metabolites in the urban rivers of Beijing, China:

Occurrence, distribution, and potential environmental risk

Yan Zhang ^a, Tingting Zhang ^b, Changsheng Guo ^a, Jiapei Lv ^a, Zhendong Hua ^{b,*},
Song Hou ^a, Yuan Zhang ^a, Wei Meng ^a, Jian Xu ^{a,*}

^a State Key Laboratory of Environmental Criteria and Risk Assessment, Chinese
Research Academy of Environmental Sciences, Beijing 100012, China

^b Drug Intelligence and Forensic Center of the Ministry of Public Security, Beijing
100193, China

*: To whom all correspondence should be addressed. E-mail: zdhuapku@gmail.com;
xujian@craes.org.cn

Abstract

The occurrence and distribution of five drugs of abuse and their metabolites, namely, methamphetamine (METH), amphetamine (AMP), ketamine (KET), ephedrine (EPH), and hydroxylamine (HY), were investigated in the surface water in urban rivers in Beijing, China. A total of 117 surface water samples were collected from the seven rivers in the Beijing urban areas during four different seasons. Laboratory analyses revealed that EPH and METH were the most predominant drugs, with detection frequency ranging from 94% to 100% and from 65% to 100%, respectively. High levels of METH and KET were observed in the center part of the urban areas, which was likely associated with the drug abuse and the entertainment activities. Seasonal variation of the drug occurrence showed that the highest concentration of drugs of abuse was found during winter, which was likely due to the high consumption, low temperature, and low water flows during the cold weather. By contrast, the concentration and detection frequency of AMP and HY were relatively low. Risk assessment by the risk quotient method indicated that the five drugs of abuse and their metabolites were not likely to exert biological effects on the aquatic ecosystems at current levels in the surface water. However, the potential adverse effect of drugs of abuse and their metabolites on ecosystem functioning and human health should not be neglected.

Keywords: drugs of abuse, surface water, occurrence, environmental risk, Beijing

1. Introduction

The consumption of drugs of abuse has been increasing worldwide and caused not only serious social problems but also concerns on emerging environmental contaminants. According to the “World Drug Report 2016,” a quarter of a billion people between the ages of 15 and 64 consumed illicit drugs at least once in 2014 (UNODC, 2016). The occurrence of drugs of abuse and their metabolites in rivers, lakes, coastal waters, and other water bodies has been widely reported across the world, with concentrations ranging from pg L^{-1} to ng L^{-1} . For instance, Bartelt-Hunt et al. (2009) reported the occurrence of methamphetamine (METH) in the Missouri River, influenced by sewage treatment plants (STPs) in Nebraska, USA, with concentrations ranging from 2 ng L^{-1} to 350 ng L^{-1} . Cocaine was found in 32% of the 50 randomly selected lakes across Minnesota, USA (Ferrey et al., 2015). In Taipei, ketamine (KET) has high concentrations in the in the surface waters of the Kaohsiung–Pingtung area, with concentrations up to 420 ng L^{-1} (Lin et al., 2014). The levels of drugs of abuse detected in surface water were also related to major sports activities, music festivals, celebrations, or holidays. Jiang et al. (2014) observed that the KET concentration in the surface waters in Taiwan could reach 9533 ng L^{-1} during the Youth Festival. Drugs of abuse have been detected not only in surface water but also in drinking water and groundwater (Boleda et al., 2011; Jurado et al., 2012). Mendoza et al. (2014) showed the presence of METH and EPH in tap water, with median concentrations of 3.13 ng L^{-1} and 0.61 ng L^{-1} , respectively.

Drugs of abuse may find their way into the water environment via direct dumping by clandestine manufacturers, drug addicts, and/or drug dealers (Li et al.,

2016). However, the majority of these drugs in the water was sewage-derived because the complete removal of illicit drugs could rarely be achieved by wastewater treatment procedures (Evgenidou et al., 2015; Pal et al., 2013; Postigo et al., 2008, 2010). Mackuľak et al. (2016) reported that the concentration levels of METH and AMP in the effluent of the Petržalka STPs in Slovakia were 220 ng L⁻¹ and 14 ng L⁻¹, respectively. Baker and Kasprzyk-Hordern (2011) found that the mean concentrations of METH, AMP, and KET in the effluent from seven STPs in the United Kingdom (UK) were 1, 8, and 130 ng L⁻¹, respectively. In some cases, the levels of illicit drugs in the effluent were higher than in the influent. Negative removal rates were observed for illicit drugs by Baker et al. (2012). KET was present with a large increase in concentration (range of 96–133%) in the effluent compared with the influent after a trickling filter treatment. In the natural surface water bodies, the effluent discharges from STPs have become the dominant source of these drugs.

Owing to the shortage of water resources, treated wastewater has been extensively used as an important supplementary water resource in many regions of the world for various purposes, such as agricultural and landscape irrigation, groundwater recharge, and supply for urban rivers (Xu et al., 2009). The effluent-associated contaminants were ultimately released into the surrounding environment, causing potential risks to the ecosystem. Drugs of abuse at trace levels from pg L⁻¹ to ng L⁻¹ would have adverse effects on the aquatic ecosystem and human health (Parolini et al., 2016; Zuccato et al., 2008). For instance, Binelli et al. (2012) reported that cocaine would cause significant primary DNA damage on *Dreissena polymorpha* in a short-

term experiment, even at an environmental concentration of 40 ng L⁻¹ that is typically found in Italian freshwater. At a high concentration of 5000 ng L⁻¹, AMP caused an overproduction of the reactive oxygen species to the freshwater bivalve *Dreissena polymorpha*, leading to oxidative damage (Parolini et al., 2016).

China is facing a severe water shortage problem, especially in megacities such as Beijing. The urban rivers in Beijing were mainly supplemented with treated effluents because of the lack of a natural runoff. Each day, approximately 3.3 million tons of domestic sewage were produced from residential areas, and more than 80% of wastewater was treated and reused in the city (Dai et al., 2015). Drugs of abuse, a kind of effluent-associated contaminants, have been detected in many water bodies. Limited reports on the occurrence of illicit drugs in the influents and effluents in STPs in China showed that METH and KET were frequently detected with high concentrations (Du et al., 2015; Gao et al., 2016; Khan et al., 2014; Lai et al., 2013; Li et al., 2014). In Huangpu River in Shanghai, caffeine, cotinine, and paraxanthine were detected with high detection frequency and concentrations, whereas the levels of other illicit drugs, such as pseudoephedrine, benzoylecgonine, and methadone, ranged from below the limit of detection (LOD) to several tens of ng L⁻¹ (Yao et al., 2016). Wang et al. (2016) found that METH and KET were the most abundant compounds in 36 rivers that are drained into the Bohai Sea and the north Yellow Sea. In 49 lakes and four major rivers (i.e., Yellow River, Songhua River, Pearl River, and Yangtze River) in China, the concentrations of METH, AMP, KET, and some other drugs were measured (Li et al., 2016).

To the best of our knowledge, this study is the first detailed investigation on the occurrence and spatial and temporal variations of five drugs of abuse and their metabolites, namely, METH, AMP, KET, EPH, and HY in the surface waters of the Beijing urban rivers. China National Narcotics Control Commission (2016) reported that METH and KET were the two most used synthetic drugs of abuse in the Chinese market. The drug EPH is the precursor of METH and AMP (Chiaia et al., 2008). The drug HA is the precursor of KET, and AMP is the major active metabolite of METH (Li et al., 2014; Postigo et al., 2008). The levels at sampling campaigns, which were the final environmental concentrations despite the degradation and input, were measured. Potential risks of these compounds to the aquatic ecosystem were also evaluated by a risk quotient method. Results from this study would help to understand the distribution and environmental risks of drugs of abuse associated with treated wastewater reuse in the surface water in Beijing urban rivers.

2. Materials and method

2.1 Reagents and materials

The drugs KET, AMP, METH, EPH, HY, and the internal standards (KET-HCl-*d*4, AMP-*d*8, METH-*d*8, and EPH-HCl-*d*8) were obtained from Cerilliant Corporation (Round Rock, TX). Details of the properties of these drugs are given in supplementary materials (Table S1). Methanol (MeOH) was purchased from Fisher (Poole, UK). Milli-Q water was prepared with a Milli-Q system (Millipore, MA, USA). Other reagents were purchased from the Sinopharm Chemical Reagent Co., Ltd (Beijing, China) and used without further purification.

The glass microfiber filters (GF/CTM filters), with a diameter of 47 mm and pore size of 1.2 μm , were purchased from Whatman Corporation (Meitesei, UK). Oasis MCX SPE cartridges (60 mg, 3 mL) were purchased from Waters Corporation (Milford, MA, USA). A Supelco VisiprepTM SPE Vacuum Manifold with 12 ports and a Welch 2023TM self-cleaning dry vacuum system were used for the loading of wastewater samples and for the drying of the cartridges.

2.2 Sample collection and pretreatment

Surface water samples were collected from 34 sites in Beijing urban rivers, as shown in Figure 1. Wenyu River with several tributaries is the main urban river in Beijing, which flows from northwest to southeast. The sampling sites were divided into seven parts: two sites (S1–S2) in Sha River, eight sites (W3–W10) in Wenyu River, eight sites (Q11–Q18) in Qing River, three sites (BX19–BX21) in Beixiao River, five sites (B22–B26) in Ba River, seven sites (T27–T33) in Tonghui River, and one site (X34) in Xiaozhong River. Four sampling campaigns were carried out in autumn (from the 9th to 11th of September 2015), winter (from the 19th to 28th of January 2016), spring (from 25th to 27th of April 2016), and summer (from 6th to 8th of July 2016). In each sampling campaign, a total of 34 surface water samples were collected, except during winter where only 15 samples were collected because of the availability at low temperature (approximately -5 °C). The water samples were collected in 1 L polypropylene bottles, stored in an ice box, and transported to the laboratory for analysis. All samples were extracted within 12 hours.

Prior to extraction, 500 mL of water samples were filtered through glass

microfiber GF/CTM filters to remove solid particles. After the addition of internal standards, water samples were passed through the Oasis MCX cartridges that were preconditioned with consecutive doses of 5 mL MeOH and 5 mL Milli-Q water. After being subjected to the water loading, the cartridges were washed with 10 mL of Milli-Q water and then dried under a vacuum for 30 minutes to remove excess water. Analytes were eluted from the MCX cartridges with 4 mL of 5% NH₃ in MeOH. The eluent solution with target compounds (in 10 mL glass centrifuge tubes marked with graduations) was nearly dried under a gentle nitrogen stream and then reconstituted to 1 mL with MeOH. After filtration through a 0.22 µm nylon film, the final extract was quantified using liquid chromatography with tandem mass spectrometry (LC–MS/MS).

METH, AMP, KET, and EPH were reported to be stable at 4 °C for a seven-day experiment (Castiglioni et al., 2006; Postigo et al., 2008; Senta et al., 2015). A preliminary experiment showed that HA was also stable in methanol at 4 °C, with a decrease in concentration below 7% after a seven-day storage. In this study, the target compounds were stored in sealed amber glass bottles at 4 °C and analyzed within one week after sample extraction.

2.3 Instrumental analysis

The target drugs were quantified by LC–MS/MS. The LC separation was performed using an Acquity UPLC separation module from Waters, equipped with an Acquity UPLC BEH HILIC column (2.1 mm × 100 mm, 1.7 µm). The column was maintained at 30 °C during the sample analysis. The ultrapure water with 0.2% formic

acid and 10 mM ammonium formate were used as mobile phase A, and the acetonitrile–water (9:1, v/v) with 0.2% formic acid, and 10 mM ammonium formate were used as mobile phase B. The gradient started with 100% of B for 0.1 minute, then increasing A to 30% in 4.8 minutes, followed by increasing A to 50% in 0.1 minute. The process was followed by maintaining A at 50% for 1.0 minute, then increasing B to 100% in 0.1 minute, and maintained for 2.9 minutes. The total run time was 9 minutes. The flow rate was 0.4 mL/min, and the injection volume was 1 μ L.

The MS analyses were performed on a triple quadrupole mass spectrometer 6500 (AB SCIEX, USA) equipped with an electrospray ionization (ESI) source that is operated in the positive ionization mode. The ion spray voltage was set to 5.5 kV. The collision gas and curtain gas pressures were 9.0 and 35.0 Psi, respectively. The pressures of the ion source gases 1 and 2 were all 50 Psi. The source temperature was 550 °C. Multiple reaction monitoring (MRM) analysis was used to identify analytes. The optimal quantification and confirmation transitions and their respective collision voltage and declustering potential are listed in Table S2.

As chiral pharmaceuticals, pseudoephedrine and ephedrine presented equal ionization and fragmentation patterns (Evans et al., 2015; Postigo et al., 2008, 2010; Reid et al., 2014; Senta et al., 2015). In this study, the two kinds of drugs were quantified together as the total ephedrine (EPH).

2.4 Quantification and quality control

The concentrations of target analytes in water samples were quantified using the

internal standard method based on an eight-point (0.5, 1, 2, 5, 10, 20, 50, and 100 $\mu\text{g L}^{-1}$) calibration curve, with a strong correlation coefficient ($R^2 > 0.99$). Four types of water (tap water, Milli-Q water, wastewater, and surface water) were spiked with five target drugs at 20 ng L^{-1} to evaluate the recoveries from different matrices. Each sample was analyzed in triplicate to calculate the relative standard deviation (RSD), and a non-spiked sample was analyzed to measure the background concentrations simultaneously. The relative recoveries ranged from 82% to 91% in tap water, 76% to 83% in Milli-Q water, 70% to 91% in wastewater, and 72% to 76% in surface water (Table S3). For spiking at different concentrations (5, 20, and 100 ng L^{-1}) in surface water, the recoveries ranged from 76% to 95%. Low RSDs ($< 10\%$) were obtained for each compound (Table S4). Procedural blanks consisting of ultrapure water were analyzed as the control of procedural contamination.

The LOD and limit of quantification (LOQ) were determined as the minimum detectable amount of an analyte from the eight environmental matrix spiked extract in MRM mode with a signal-to-noise ratio (S/N) of three and ten, respectively (Jelić et al., 2009). The S/N ratios were obtained by using the software MassLynx V4.1 (Waters, USA) from the data of the recovery test with the lowest spiked concentration for each compound. The LOD and LOQ of the drugs initially identified were estimated by determining the S/N of the minimum measured concentrations and extrapolating to the S/N values of ten and three (Shao et al., 2009). The LODs for each compound ranged from 0.3 ng L^{-1} to 0.7 ng L^{-1} , and the LOQs were between 0.8 ng L^{-1} and 2.0 ng L^{-1} . The details above are featured in Table 1, Table S3, and Table S4.

2.5 Risk characterization

The risk quotient (RQ), defined by the US EPA as the ratio of the potential exposure to the substances and the level at which no adverse effects are expected, was used to evaluate the environmental risk level (US EPA, 1997a, 1997b). For each individual illicit drug, the RQ was calculated through its measured environmental concentration (MEC) at each sampling site divided by the predicted no-effect concentration (PNEC). The PNEC was calculated from the available aquatic toxicity data of illicit drugs and the correlated assessment factors (AFs) (Liu et al., 2015; Mendoza et al., 2014).

$$RQ = \frac{MEC}{PNEC} \quad (1)$$

$$PNEC = \frac{Toxicity\ benchmark}{AF} \quad (2)$$

Generally, a RQ above 1 indicates a high ecological risk to aquatic organisms, $0.1 < RQ < 1$ reveals a medium risk, and a $RQ < 0.1$ represents a low risk. An exhaustive literature had reported toxicity data for algae, cladocerans, and fishes for the studied drugs. Thus, when no experimental toxicity data were available, predicted toxicity values from the US EPA Ecological Structure Activity Relationships (ECOSAR) Class Program were used. The lowest median lethal or effective concentration (LC50 or EC50) value among the three groups (algae, cladocerans, and fish) of illicit drugs was selected for the next calculations. An AF of 1000 was applied to all available data as they were all based on ECOSAR models or a limited number of short-term LC50 or EC50 values.

3. Results and discussion

3.1 Occurrence of drugs of abuse in the surface water of Beijing urban rivers

The concentrations and detection frequency of drugs of abuse in the urban surface water in Beijing are shown in Table 2. Generally, concentrations of all target compounds were in the ng L^{-1} level. EPH was detected with high levels among the five drugs in the surface water, with the highest concentration up to 185.7 ng L^{-1} . EPH was also detected with high detection frequencies. Except for two sites in summer where EPH was not detected, EPH was found in all other samples during four sampling campaigns (Table 2). The detection frequency of METH was also high ($>65\%$), indicating the ubiquity of this drug in the Beijing surface waters. The highest METH concentration could reach up to 99.5 ng L^{-1} . The concentrations of AMP and KET were relatively low; however, they were present in the surface water in autumn, with a high detection frequency of 97% and 74%, respectively. During the four sampling periods, HY was not detected during spring and summer. In autumn and winter, the detection frequency of HY was 82% and 27%, respectively, but its levels were low, with median concentrations ranging from below LOD to 1.7 ng L^{-1} .

Monitoring results of high concentrations and detection frequency of METH in this study were consistent with the seizure data from the Chinese authorities, which indicated that METH and KET were the two major drugs with increasing abuse dosage in China (China National Narcotics Control Commission, 2016). However, in the Beijing surface water, KET was not at high concentrations throughout the one-year survey, which was consistent with the effluent data from Beijing sewage

treatment plants (Du et al., 2015). The concentration levels of the target drugs in the Beijing surface water were compared with other reported results in other regions in the world, as summarized in Table S5. The METH levels (2.8–12.0 ng L⁻¹) were higher than those rivers in the US (Bartelt-Hunt et al., 2009; Loganathan et al., 2009), the UK (Baker and Kasprzyk-Hordern, 2011, 2013; Kasprzyk-Hordern et al., 2008; Zuccato et al., 2008), Italy (Zuccato et al., 2008), Ireland (Bones et al., 2007), Switzerland (Berset et al., 2010), and most areas in Spain (Bueno et al., 2011; Kasprzyk-Hordern et al., 2008; Postigo et al., 2010). Moreover, the METH levels were lower than in Taiwan (nd-917 ng L⁻¹) (Jiang et al., 2014; Lin et al., 2010) and the Pearl River (31.1 ng L⁻¹) (Li et al., 2016), but comparable to rivers in Madrid, Spain (3.1-5.0 ng L⁻¹) (Mendoza et al., 2014), Yellow River (1.3 ng L⁻¹), Songhua River (2.2 ng L⁻¹), and Yangtze River (1.9 ng L⁻¹) in China (Li et al., 2016). The AMP concentrations in this study (with median concentrations of nd-2.8 ng L⁻¹) were slightly lower than in Southern Taiwan (nd-90.3 ng L⁻¹) (Jiang et al., 2014) and Henares River in Spain (309 ng L⁻¹) (Bueno et al., 2011). KET levels (nd-1.6 ng L⁻¹) were lower than in Taiwan (nd-9533 ng L⁻¹) (Jiang et al., 2014), the UK (21.3 ng L⁻¹) (Baker and Kasprzyk-Hordern, 2011), and Pearl River in China (15.6 ng L⁻¹) (Li et al., 2016). The occurrence of EPH was reported mainly in Spain and the UK. The levels of EPH in the urban surface water in Beijing, with median concentrations of 11.5–119.0 ng L⁻¹, were lower than those in Spain (5.4 to 206.0 ng L⁻¹) (Bueno et al., 2011; Mendoza et al., 2014; Postigo et al., 2010), but higher than in the surface water in the UK (nd-14.5 ng L⁻¹) (Baker and Kasprzyk-Hordern, 2011, 2013). To the best of our

knowledge, no literature has reported the occurrence of HY in the water bodies. In this study, HY was initially reported as ranging from less than LOD to 15.9 ng L⁻¹ in the surface water of Beijing urban rivers.

3.2 Spatial distribution of drugs of abuse in the surface water of Beijing urban rivers

The spatial distribution of the target drugs in the urban river water is illustrated in Figure 2. Except in the three sites (Q12–Q14) in the Qing River, METH was detected in all river water samples, with concentrations ranging from 2.0 ng L⁻¹ to 99.5 ng L⁻¹, suggesting the popular use of METH in Beijing. Spatially, METH was present at relatively high levels in both tributaries and Wenyu River, likely owing to the low dilution in the main stem river (the average dilution factor of 2.3 was assessed according to Pernet-Coudrier et al. (2012)). At the confluence of tributaries in Wenyu River, concentrations of METH in the downstream were slightly higher than in the upstream. For instance, METH concentration in site W4 was higher than W3, and W10 was higher than W9, as shown in Figure 2. High levels at W4 and W10 were attributed to the high tributary concentrations from Beixiao River and Ba River, respectively. Sampling site B23 in Ba River had the highest METH concentration of up to 99.5 ng L⁻¹. This site was located at the downstream of Sanlitun, which was known as a recreation center with considerable night clubs. METH was expected to reach the river from these clubs by the direct and/or indirect dumping events and via wastewater effluents, contributing to the high aqueous levels in the Ba River. Relatively high levels of METH in Beixiao River and Tonghui River were associated with the surrounding urban areas, where there were many entertainment centers with

high population density. Wastewater from these entertainment centers reached adjacent STPs, where the insufficient removal of METH resulted in the high concentrations of drug residues in the natural surface water. By contrast, levels of METH in the Qing River were low. Qing River flows through the less densely populated area and receives large quantities of effluent from the Qinghe sewage treatment plant. A previous study reported that the removal rates of METH in the majority of STPs in Beijing were greater than 80% (Du et al., 2015). It confirmed that entertainment venues were important sources of METH and partly explained the high METH concentrations in the three rivers. A similar geographical pattern of METH distribution was also observed by Irvine et al. (2011), reporting that METH concentrations were much higher in the metropolitan area than in regional areas in southern Australia.

AMP was detected in all sampling sites except in T28, with concentrations ranging from below LOQ to several ng L^{-1} . Although AMP accounted for 9.20–17.47% of the total target drugs in the upstream of Qing River (sites Q11–Q14), the concentrations were relatively low (Figure 2). Selegiline, which could metabolize directly into AMP, was a legal drug used for the treatment of Parkinson's disease in China. It meant that there existed licit pathway to release AMP to sewage in Beijing. AMP was also the major metabolite of METH (Postigo, et al., 2008). The concentration ratio of AMP/METH in wastewater was less than 10%, indicating that AMP was mainly derived from METH metabolism (Du et al., 2015). In all four sampling campaigns, the AMP/METH ratios were found to be less than 10% at the

W4 site, which was far from the center of Beijing and had a low population density. The AMP was likely predominantly sourced from METH metabolism rather than from the licit pathway from this site.

EPH was the dominant drug in the surface water, which accounted for 48.99% to 92.45% of the total target drugs in all 34 sampling sites. Although EPH is the precursor drug of AMP and METH, it also has legal uses, such as in bronchodilators and nasal decongestants (Baker and Kasprzyk-Hordern, 2011). The high levels of EPH, evidenced by its high detection frequency, reflected the widespread application of EPH as a prescription drug in Beijing.

KET was present in all samples in the Sha River, Beixiao River, and Ba River during autumn, with total concentrations of four seasons ranging from 1.5–1.7 ng L⁻¹, 10.1–16.3 ng L⁻¹, and 1.5–11.8 ng L⁻¹, respectively. The detection frequencies were 75% in Wenyun River, 50% in Qing River, 86% in Tonghui River, and 0% in Xiaozhong River. In Beixiao River, KET had relatively higher concentrations and detection frequency than other rivers. This distribution pattern was mainly because the Beixiao River STP treated the wastewater from the surrounding Olympic Village, where a large number of sports competitions, commercial performances, and recreational activities are often held. The low removal efficiency of KET by STPs caused a considerable amount of KET to be released into the surface water (Andrés-Costa et al., 2014; Du et al., 2015; Yargeau et al., 2014). Similar to METH, the KET levels in the Ba River were also high. HY was used as the pharmaceutical intermediate of narcotic drugs to synthesize KET. The proportion of HY in the total drugs was low, ranging

from 0% to 8.28%. No significant differences were observed for this compound among 34 sampling sites in different urban rivers in Beijing.

3.3 Temporal variation of drugs of abuse in the surface water of Beijing urban rivers

3.3.1 Seasonal variations

Figure 3 illustrates the seasonal variations of drugs of abuse in the surface water samples in Beijing. It shows that the total concentrations of target drugs were high in winter (32.0–245.1 ng L⁻¹) and low in summer (0–140.2 ng L⁻¹) in most sampling sites. This result is consistent with the previous study that reported that target drugs, as one of the micropollutants, presented higher concentrations in the dry season (winter and spring) than in the wet season (summer and fall) in surface water (Mendoza et al., 2014). During the sampling held in January 2016, the average water temperature was 5 °C, compared to the 27 °C during July 2016. The warm temperature favored microbial activity, resulting in the enhanced biodegradation of drugs in rivers. In summer, the intense ultraviolet radiation may also trigger a higher photodegradation of drugs than in winter, contributing to the low concentrations in summer in natural waters (Bagnall et al., 2013; Lin et al., 2014). Additionally, the average annual rainfall in Beijing was 640 mm, with approximately 72.5% of the total precipitation occurring from June to August (Li et al., 2015). The dilution by heavy rainfall and surface runoff significantly affected the level of drugs of abuse in the surface water, leading to the low concentrations in the wet season.

As shown in Figure 2, EPH was the predominant compound among the five target drugs, which greatly influenced the changes in the total amounts. The

concentrations of EPH were up to 26.7–185.7 ng L⁻¹ in winter and from 3.0–106.1 ng L⁻¹ in spring. The ubiquity of EPH in the surface water in winter and spring was likely related to its therapeutic use (besides its illicit abuse) for the treatment of influenza, asthma, or hypotension (Mendoza et al., 2014). A study performed in Barcelona showed that the EPH concentrations in September and December were higher than in July because of its use against influenza (Postigo et al., 2011). The high EPH levels could be also because of its high human metabolite excretion rate, which was between 22% and 99%, but without complete elimination in STPs (Postigo et al., 2008, 2010).

By contrast, the METH concentrations in winter and spring were lower than in summer and autumn (Table 2). This finding was consistent with the situation in the Pacific Northwest in the US where the METH abuse was slightly higher in summer than in spring (Chiaia-Hernandez et al., 2011). Postigo et al. (2010) also reported that the drug abuse was more intense in July than in October. More entertainment and sports activities during summer expectedly led to the relatively high illicit drug use and resulted in high drug levels in the surface water. Factors such as the consumption dosage of drugs (Li et al., 2016), the water temperature (Postigo et al., 2008), the ultraviolet exposure (Bagnall et al., 2013; Lin et al., 2014), and river flows (Li et al., 2016; Zuccato et al., 2008) were hypothesized to affect the persistence and distribution patterns of drugs of abuse in the surface water in a one-year survey.

3.3.2 Weekly variations

A sampling campaign was performed between January 20 and 28 in 2016 at Site B23, owing to its highest total concentrations, to investigate the weekly variations of

these drugs in the river water. During this sampling week, KET and HY were not detected. The weekly variations of the other three target drugs at B23 are shown in Figure 4. Results showed that AMP concentrations were roughly constant throughout the entire week. Although the concentrations of METH and EPH fluctuated during the week, no statistically significant difference in the METH and EPH concentrations between weekdays and weekend at B23 ($p>0.5$) was observed. In several other studies, a similar trend was also observed, such as in 19 European cities (Thomas et al., 2012), Pacific Northwest (Chiaia-Hernandez et al., 2011), Brussels, Belgium (van Nuijs et al., 2011), and Hong Kong (Lai et al., 2013). However, in some other areas, an increased use of illicit drugs during weekends was reported (Huerta-Fontela et al., 2008; Lai et al., 2011; Metcalfe et al., 2010; Reid et al., 2011). Furthermore, no universal pattern in the daily variation of illicit drug use across the globe was observed. The absence or presence of differences in illicit drug use between weekdays and the weekend largely depended on a particular city or even on a particular season. Additionally, the surface water is an open system, and the pattern of weekly variations in the surface water at B23 might be influenced by the surface runoff dilution.

3.4 Environmental risk assessment

No chronic data was available for the studied drugs. The risk of HY was not evaluated in this study, given that neither chronic nor acute toxicity data were found for HY. The acute toxicity values for EPH and AMP were available from existing studies, and the toxicological data of METH and KET were calculated via ECOSAR, as shown in Table 3. The RQ values for all compounds were between 0.00 and 0.05,

suggesting that adverse effects were improbable. The environmental risk of drugs of abuse in the rivers of Rhine and Meuse in the Netherlands was evaluated by this method, with RQ values ranging from 0.0002 to 0.38 (Van der Aa et al., 2013). In a Mediterranean river (Llobregat, NE Spain), the RQ values for the illicit drugs were below 1 (López-Serna et al., 2012). This result, however, should be treated with caution. The potential exposure data were based on a punctual sampling event; though the data represented the reality of the sampling moment, they did not give a real picture of the long-term exposure of substances in the ecosystem (Mendoza et al., 2014). The limited experimental toxicological data and the use of high AFs may have an influence on the RQ values. Therefore, the potential adverse effects of illicit drugs on the ecosystem functioning and human health should not be neglected.

4. Conclusions

In this study, the occurrence and distribution of five drugs of abuse and their metabolites were investigated in the urban surface water samples in Beijing. Results showed that the levels of these drugs were site-specific and season-influenced. METH and KET were observed with high concentrations in the center part of the urban areas in Beijing, which, in turn, was related to the drug uses, the population density, and entertainment activities. Seasonal variations reflected that factors, including the water temperature, river flow, ultraviolet exposure, and rainfall, may affect the concentrations of drugs significantly. Risk assessment showed that these target drugs at their current concentration levels were not likely to exert biological effects on the aquatic ecosystem in the Beijing surface water. However, the potential adverse effects

of these drugs should not be neglected.

Acknowledgments

This work was funded by the National Science Foundation of China (No. 41673120).

References

- Andrés-Costa, M.J., Rubio-López, N., Suárez-Varela, M.M., Pico, Y., 2014. Occurrence and removal of drugs of abuse in Wastewater Treatment Plants of Valencia (Spain). *Environ. Pollut.* 194, 152-162.
- Bagnall, J., Malia, L., Lubben, A., Kasprzyk-Hordern, B., 2013. Stereoselective biodegradation of amphetamine and methamphetamine in river microcosms. *Water Res.* 47, 5708-5718.
- Baker, D.R., Kasprzyk-Hordern, B., 2011. Multi-residue analysis of drugs of abuse in wastewater and surface water by solid-phase extraction and liquid chromatography–positive electrospray ionisation tandem mass spectrometry. *J. Chromatogr. A* 1218, 1620-1631.
- Baker, D.R., Kasprzyk-Hordern, B., 2013. Spatial and temporal occurrence of pharmaceuticals and illicit drugs in the aqueous environment and during wastewater treatment: new developments. *Sci. Total Environ.* 454, 442-456.
- Baker, D.R., Očenášková, V., Kvicalova, M., Kasprzyk-Hordern, B., 2012. Drugs of abuse in wastewater and suspended particulate matter—further developments in sewage epidemiology. *Environ. Int.* 48, 28-38.
- Bartelt-Hunt, S.L., Snow, D.D., Damon, T., Shockley, J., Hoagland, K., 2009. The occurrence of illicit and therapeutic pharmaceuticals in wastewater effluent and

- surface waters in Nebraska. *Environ. Pollut.* 157, 786-791.
- Berset, J.D., Brenneisen, R., Mathieu, C., 2010. Analysis of illicit and illicit drugs in waste, surface and lake water samples using large volume direct injection high performance liquid chromatography–electrospray tandem mass spectrometry (HPLC–MS/MS). *Chemosphere* 81, 859-866.
- Binelli, A., Pedriali, A., Riva, C., Parolini, M., 2012. Illicit drugs as new environmental pollutants: Cyto-genotoxic effects of cocaine on the biological model *Dreissena polymorpha*. *Chemosphere* 86, 906-911.
- Boleda, M.R., Huerta-Fontela, M., Ventura, F., Galceran, M.T., 2011. Evaluation of the presence of drugs of abuse in tap waters. *Chemosphere* 84, 1601-1607.
- Bones, J., Thomas, K.V., Paull, B., 2007. Using environmental analytical data to estimate levels of community consumption of illicit drugs and abused pharmaceuticals. *J. Environ. Monit.* 9, 701-707.
- Bueno, M.J.M., Uclés, S., Hernando, M.D., Fernández-Alba, A.R., 2011. Development of a solvent-free method for the simultaneous identification/quantification of drugs of abuse and their metabolites in environmental water by LC–MS/MS. *Talanta* 85, 157-166.
- Castiglioni, S., Zuccato, E., Crisci, E., Chiabrando, C., Fanelli, R., Bagnati, R., 2006. Identification and measurement of illicit drugs and their metabolites in urban wastewater by liquid chromatography-tandem mass spectrometry. *Anal. Chem.* 78, 8421-8429.
- Chiaia-Hernandez, A.C., Banta-Green, C.J., Field, J.A., 2011. Interpreting

- methamphetamine levels in a high-use community. *Environ Sci. Pollut. Res.* 18, 1471-1477.
- Chiaia, A.C., Banta-Green, C., Field, J., 2008. Eliminating solid phase extraction with large-volume injection LC/MS/MS: analysis of illicit and legal drugs and human urine indicators in US wastewaters. *Environ. Sci. Technol.* 42, 8841-8848.
- China National Narcotics Control Commission, 2016. Annual Report on Drug Control in China, Beijing, China.
- Dai, G., Wang, B., Huang, J., Dong, R., Deng, S., Yu, G., 2015. Occurrence and source apportionment of pharmaceuticals and personal care products in the Beiyun River of Beijing, China. *Chemosphere* 119, 1033-1039.
- Du, P., Li, K., Li, J., Xu, Z., Fu, X., Yang, J., Zhang, H., Li, X., 2015. Methamphetamine and ketamine use in major Chinese cities, a nationwide reconnaissance through sewage-based epidemiology. *Water Res.* 84, 76-84.
- Evans, S.E., Davies, P., Lubben, A., Kasprzyk-Hordern, B., 2015. Determination of chiral pharmaceuticals and illicit drugs in wastewater and sludge using microwave assisted extraction, solid-phase extraction and chiral liquid chromatography coupled with tandem mass spectrometry. *Anal. Chim. Acta* 882, 112-126.
- Evgenidou, E.N., Konstantinou, I.K., Lambropoulou, D.A., 2015. Occurrence and removal of transformation products of PPCPs and illicit drugs in wastewaters: a review. *Sci. Total Environ.* 505, 905-926.
- Ferrey, M.L., Heiskary, S., Grace, R., Hamilton, M.C., Lueck, A., 2015.

- Pharmaceuticals and other anthropogenic tracers in surface water: A randomized survey of 50 Minnesota lakes. *Environ. Toxicol. Chem.* 34, 2475-2488.
- Gao, T., Du, P., Xu, Z., Li X., 2016. Occurrence of new psychoactive substances in wastewater of major Chinese cities. *Sci. Total Environ.* <http://dx.doi.org/10.1016/j.scitotenv.2016.09.152>
- Huerta-Fontela, M., Galceran, M.T., Martin-Alonso, J., Ventura, F., 2008. Occurrence of psychoactive stimulatory drugs in wastewaters in north-eastern Spain. *Sci. Total Environ.* 397, 31-40.
- Irvine, R.J., Kostakis, C., Felgate, P.D., Jaehne, E.J., Chen, C., White, J.M., 2011. Population drug use in Australia: a wastewater analysis. *Forensic Sci. Int.* 210, 69-73.
- Jelić, A., Petrović, M., Barceló, D., 2009. Multi-residue method for trace level determination of pharmaceuticals in solid samples using pressurized liquid extraction followed by liquid chromatography/quadrupole-linear ion trap mass spectrometry. *Talanta* 80, 363-371.
- Jiang, J.J., Lee, C.L., Fang, M.D., Tu, B.W., Liang, Y.J., 2014. Impacts of emerging contaminants on surrounding aquatic environment from a youth festival. *Environ. Sci. Technol.* 49, 792-799.
- Jurado, A., Mastroianni, N., Vázquez-Suñé, E., Carrera, J., Tubau, I., Pujades, E., Postigo, C., Alda M.L.D., Barceló, D., 2012. Drugs of abuse in urban groundwater. A case study: Barcelona. *Sci. Total Environ.* 424, 280-288.
- Kasprzyk-Hordern, B., Dinsdale, R.M., Guwy, A.J., 2008. Multiresidue methods for

the analysis of pharmaceuticals, personal care products and illicit drugs in surface water and wastewater by solid-phase extraction and ultra performance liquid chromatography-electrospray tandem mass spectrometry. *Anal. Bioanal. Chem.* 391, 1293-1308.

Khan, U., van Nuijs, A.L.N., Li, J., Maho, W., Du, P., Li, K., Hou, L., Zhang, J., Meng, X., Li, X., Covaci, A., 2014. Application of a sewage-based approach to assess the use of ten illicit drugs in four Chinese megacities. *Sci. Total Environ.* 487, 710-721.

Lai, F.Y., Bruno, R., Leung, H.W., Thai, P.K., Ort, C., Carter, S., Thompson, K., Lam, P.K.S., Mueller, J.F., 2013. Estimating daily and diurnal variations of illicit drug use in Hong Kong: a pilot study of using wastewater analysis in an Asian metropolitan city. *Forensic Sci. Int.* 233, 126-132.

Lai, F.Y., Ort, C., Gartner, C., Carter, S., Prichard, J., Kirkbride, P., Bruno, R., Hall, W., Eaglesham, G., Mueller, J.F., 2011. Refining the estimation of illicit drug consumptions from wastewater analysis: co-analysis of prescription pharmaceuticals and uncertainty assessment. *Water Res.* 45, 4437-4448.

Li, J., Hou, L., Du, P., Yang, J., Li, K., Xu, Z., Wang, C., Zhang, H., Li, X., 2014. Estimation of amphetamine and methamphetamine uses in Beijing through sewage-based analysis. *Sci. Total Environ.* 490, 724-732.

Li, K., Du, P., Xu, Z., Gao, T., Li, X., 2016. Occurrence of illicit drugs in surface waters in China. *Environ. Pollut.* 213, 395-402.

Li, W., Gao, L., Shi, Y., Liu, J., Cai, Y., 2015. Occurrence, distribution and risks of

- antibiotics in urban surface water in Beijing, China. *Environ Sci. Proc. Impac.* 17, 1611-1619.
- Lilius, H., Isomaa, B., Holmström, T., 1994. A comparison of the toxicity of 50 reference chemicals to freshly isolated rainbow trout hepatocytes and *Daphnia magna*. *Aquat. Toxicol.* 30, 47-60.
- Lin, A.Y.C., Lin, Y.C., Lee, W.N., 2014. Prevalence and sunlight photolysis of controlled and chemotherapeutic drugs in aqueous environments. *Environ. Pollut.* 187, 170-181.
- Lin, A.Y.C., Wang, X.H., Lin, C.F., 2010. Impact of wastewaters and hospital effluents on the occurrence of controlled substances in surface waters. *Chemosphere* 81, 562-570.
- Liu, J., Lu, G., Xie, Z., Zhang, Z., Li, S., Yan, Z., 2015. Occurrence, bioaccumulation and risk assessment of lipophilic pharmaceutically active compounds in the downstream rivers of sewage treatment plants. *Sci. Total Environ.* 511, 54-62.
- Loganathan, B., Phillips, M., Mowery, H., Jones-Lepp, T.L., 2009. Contamination profiles and mass loadings of macrolide antibiotics and illicit drugs from a small urban wastewater treatment plant. *Chemosphere* 75, 70-77.
- López-Serna, R., Postigo, C., Blanco, J., Pérez, S., Ginebreda, A., de Alda, M.L., Barceló, D., 2012. Assessing the effects of tertiary treated wastewater reuse on the presence emerging contaminants in a Mediterranean river (Llobregat, NE Spain). *Environ. Sci. Pollut. Res.* 19, 1000-1012.
- Mackuľák, T., Birošová, L., Bodík, I., Grabic, R., Takáčová, A., Smolinská, M.,

- Hanusová, A., Híveš, J., Gál, M., 2016. Zerovalent iron and iron (VI): Effective means for the removal of psychoactive pharmaceuticals and illicit drugs from wastewaters. *Sci. Total Environ.* 539, 420-426.
- Mendoza, A., Rodríguez-Gil, J.L., González-Alonso, S., Mastroianni, N., de Alda, M.L., Barceló, D., Valcárcel, Y., 2014. Drugs of abuse and benzodiazepines in the Madrid Region (Central Spain): Seasonal variation in river waters, occurrence in tap water and potential environmental and human risk. *Environ. Int.* 70, 76-87.
- Metcalf, C., Tindale, K., Li, H., Rodayan, A., Yargeau, V., 2010. Illicit drugs in Canadian municipal wastewater and estimates of community drug use. *Environ. Pollut.* 158, 3179-3185.
- Pal, R., Megharaj, M., Kirkbride, K.P., Naidu, R., 2013. Illicit drugs and the environment—a review. *Sci. Total Environ.* 463, 1079-1092.
- Parolini, M., Magni, S., Castiglioni, S., Binelli, A., 2016. Amphetamine exposure imbalanced antioxidant activity in the bivalve *Dreissena polymorpha* causing oxidative and genetic damage. *Chemosphere* 144, 207-213.
- Pernet-Coudrier, B., Qi, W., Liu, H., Müller, B., Berg, M., 2012. Sources and pathways of nutrients in the semi-arid region of Beijing–Tianjin, China. *Environ. Sci. Technol.* 46, 5294-5301.
- Postigo, C., de Alda, M.J.L., Barceló, D., 2010. Drugs of abuse and their metabolites in the Ebro River basin: occurrence in sewage and surface water, sewage treatment plants removal efficiency, and collective drug usage estimation.

- Environ. Int. 36, 75-84.
- Postigo, C., de Alda, M.J.L., Barceló, D., 2008. Analysis of drugs of abuse and their human metabolites in water by LC-MS²: A non-intrusive tool for drug abuse estimation at the community level. *TrAC Trends Anal. Chem.* 27, 1053-1069.
- Postigo, C., de Alda, M.L., Barceló, D., 2011. Evaluation of drugs of abuse use and trends in a prison through wastewater analysis. *Environ. Int.* 37, 49-55.
- Reid, M.J., Derry, L., Thomas, K.V., 2014. Analysis of new classes of recreational drugs in sewage: Synthetic cannabinoids and amphetamine - like substances. *Drug Test Anal.* 6, 72-79.
- Reid, M.J., Langford, K.H., Mørland, J., Thomas, K.V., 2011. Quantitative assessment of time dependent drug-use trends by the analysis of drugs and related metabolites in raw sewage. *Drug Alcohol. Depen.* 119, 179-186.
- Sanderson, H., Johnson, D.J., Reitsma, T., Brain, R.A., Wilson, C.J., Solomon, K.R., 2004. Ranking and prioritization of environmental risks of pharmaceuticals in surface waters. *Regul. Toxicol. Pharm.* 39, 158-183.
- Senta, I., Krizman, I., Ahel, M., Terzic, S., 2015. Multiresidual analysis of emerging amphetamine-like psychoactive substances in wastewater and river water. *J. Chromatogr. A* 1425, 204-212.
- Shao, B., Chen, D., Zhang, J., Wu, Y., Sun, C., 2009. Determination of 76 pharmaceutical drugs by liquid chromatography–tandem mass spectrometry in slaughterhouse wastewater. *J. Chromatogr. A* 1216, 8312-8318.
- Thomas, K.V., Bijlsma, L., Castiglioni, S., Covaci, A., Emke, E., Grabic, R.,

- Hernández F., Karolak, S., Kasprzyk-Hordern, B., Lindberg, R.H., de Alda, M.L., Meierjohann, A., Ort, C., Pico, Y., Quintana, J.B., Reid, M., Rieckermann, J., Terzic, S., van Nuijs, A.L.N., Voogt P.D., 2012. Comparing illicit drug use in 19 European cities through sewage analysis. *Sci. Total Environ.* 432, 432-439.
- UNODC. United Nations Office of Drugs and Crime, 2016. *World Drug Report*, United Nations, New York.
- US EPA. Ecological risk assessment guidance for superfund: process for designing and conducting ecological risk assessments — interim final (EPA publication no. 540/R-97/006). Washington D.C.: U.S. Environmental Protection Agency; 1997b.
- US EPA. Terms of environment: glossary, abbreviations and acronyms (EPA publication no. 175-B-97-001). Washington D.C.: U.S. Environmental Protection Agency; 1997a.
- Van der Aa, M., Bijlsma, L., Emke, E., Dijkman, E., van Nuijs, A.L.N., Van de Ven, B., Hernández, F., Versteegh, A., Voogt, P.D., 2013. Risk assessment for drugs of abuse in the Dutch watercycle. *Water Res.* 47, 1848-1857.
- van Nuijs, A.L.N., Mougel, J.F., Tarcomnicu, I., Bervoets, L., Blust, R., Jorens, P.G., Neels, H., Covaci, A., 2011. A one year investigation of the occurrence of illicit drugs in wastewater from Brussels, Belgium. *J. Environ. Monit.* 13, 1008-1016.
- Wang, D.G., Zheng, Q.D., Wang, X.P., Du, J., Tian, C.G., Wang, Z., Ge, L.K., 2016. Illicit drugs and their metabolites in 36 rivers that drain into the Bohai Sea and north Yellow Sea, north China. *Environ. Sci. Pollut. Res.* 23:16495–16503.

- Xu, J., Wu, L., Chang, A.C., 2009. Degradation and adsorption of selected pharmaceuticals and personal care products (PPCPs) in agricultural soils. *Chemosphere* 77, 1299-1305.
- Yao, B., Lian, L., Pang, W., Yin, D., Chan, S.A., Song, W., 2016. Determination of illicit drugs in aqueous environmental samples by online solid-phase extraction coupled to liquid chromatography–tandem mass spectrometry. *Chemosphere* 160, 208-215.
- Yargeau, V., Taylor, B., Li, H., Rodayan, A., Metcalfe, C.D., 2014. Analysis of drugs of abuse in wastewater from two Canadian cities. *Sci. Total Environ.* 487, 722-730
- Zuccato, E., Castiglioni, S., Bagnati, R., Chiabrando, C., Grassi, P., Fanelli, R., 2008. Illicit drugs, a novel group of environmental contaminants. *Water Res.* 42, 961-968.

Figure captions

Figure 1 Location of the sampling sites

Figure 2 Composition distribution of drugs of abuse and their metabolites in 34 sampling sites in Beijing

Figure 3 Distribution of drugs of abuse and their metabolites in urban surface water in Beijing

Figure 4 Weekly variation of drugs of abuse and their metabolites in surface water at B23

Figure 1 Location of the sampling sites

Figure 2 Composition distribution of drugs of abuse and their metabolites in 34

sampling sites in Beijing

Figure 3 Distribution of drugs of abuse and their metabolites in urban surface water in Beijing

Figure 4 Weekly variation of drugs of abuse and their metabolites in surface water at

B23

Table 1 Method validation parameters: Linear range, correlation coefficients (r^2), precision, limits of detection (LODs, S/N=3) and limits of quantification (LOQs) of five target drugs

Analytes	Linear range (ng L ⁻¹)	r ²	Precision(% RSDs)				LOD (ng L ⁻¹)	LOQ (ng L ⁻¹)
			Repeatability		Intermediate precision			
			at 5 ng L ⁻¹	at 20 ng L ⁻¹	at 5 ng L ⁻¹	at 100 ng L ⁻¹		
			(n=5)	(n=5)	(n=5)	(n=5)		
METH	1-500	0.9986	3.28	5.33	5	3	0.6	2.0
AMP	0.3-750	0.9985	8.20	4.38	2	1	0.2	0.8
KET	0.3-600	0.9990	0.68	3.98	3	3	0.3	1.0
EPH	1.0-1000	0.9987	2.46	3.08	3	1	0.3	1.0
HA	0.8-500	0.9990	7.24	4.53	4	2	0.2	0.8

Table 2 Frequency of detection, median, mean and concentration range of the five target drugs in surface water

Analytes	Autumn				Winter				Spring				Summer			
	Freq(%)	Median	Mean	Range	Freq(%)	Median	Mean	Range	Freq(%)	Median	Mean	Range	Freq(%)	Median	Mean	Range
	n=34	(ng L ⁻¹)	(ng L ⁻¹)	(ng L ⁻¹)	n=15	(ng L ⁻¹)	(ng L ⁻¹)	(ng L ⁻¹)	n=34	(ng L ⁻¹)	(ng L ⁻¹)	(ng L ⁻¹)	n=34	(ng L ⁻¹)	(ng L ⁻¹)	(ng L ⁻¹)
METH	85	6.9	14.6	nd ^a -99.5	100	12.0	15.3	3.3-57.4	79	5.2	7.1	nd-51.9	65	2.8	6.9	nd-80.6
AMP	97	2.8	3.1	nd-11.2	40	nd	1.1	nd-5.0	-	nd	nd	nd	3	nd	<LOQ	nd-8.8
KET	74	1.6	2.9	nd-16.3	27	nd	<LOQ	nd-3.9	24	nd	1.2	nd-17.1	-	nd	nd	nd
EPH	100	20.1	22.8	1.2-75.1	100	119.0	115.0	26.7-185.7	100	32.9	39.6	3.0-106.1	94	11.5	15.7	nd-50.8
HY	82	1.7	3.0	nd-15.9	27	nd	0.8	nd-4.0	-	nd	nd	nd	-	nd	nd	nd

^a Not detected.

Table 3 Predicted no effect concentrations (PNECs) (mg L⁻¹) for fish, cladocerans and green algae

Analytes	EC ₅₀ /(mg L ⁻¹)			Selected	AF	PNEC/(mg L ⁻¹)	RQ	Reference
	Fish	Cladocerans	Algae					
METH	20.51	2.51	1.97	1.97	1000	1.97×10 ⁻³	0.00-0.05	ECOSAR
AMP	28.80	2.22	3.80	2.22	1000	2.22×10 ⁻³	0.00-0.01	Lilius et al. (1994)
KET	8.34	1.13	0.72	0.72	1000	7.20×10 ⁻⁴	0.00-0.02	ECOSAR
EPH	56.00	3.62	3.91	3.62	1000	3.62×10 ⁻³	0.00-0.05	Sanderson et al. (2004)