Using environmental analytical data to estimate levels of community consumption of illicit drugs and abused pharmaceuticals†

Jonathan Bones,[a] Kevin V. Thomas[b] and Brett Paull*[a]

Received 23rd February 2007, Accepted 1st May 2007
First published as an Advance Article on the web 17th May 2007
DOI: 10.1039/b702799k

A solid phase extraction (SPE) method has been developed and applied in conjunction with a previously reported liquid chromatography tandem mass spectrometry (LC–MS–MS) procedure for the determination of illicit drugs and abused pharmaceuticals in treated wastewater and surface water samples at the ng L^{-1} level. A full method validation was also performed and determined levels of analytical sensitivity were found to lie in the 1–10 ng L^{-1} range using river water as a test sample matrix and a sample size of 500 mL. The developed procedure was successfully applied for the determination of the chosen analytes in wastewater treatment plants in Dublin, Ireland and rapidly expanding commuter towns in the surrounding counties. Cocaine was detected in 70% of the collected samples in the range of 25–489 ng L^{-1}, its primary metabolite, benzoylecognine (BZE) was also detected in the range of 22–290 ng L^{-1}. Other substances detected included morphine, Tempazepam and the primary metabolite of methadone.

Introduction

In Ireland, there has of late been increased media attention concerning the escalating use of illicit drugs, in particular cocaine, within society.[1] Such media reports appear to be corroborated by official seizure data from An Garda Síochána, wherein a search of press releases reveal no less than eighteen major hauls of cannabis, cocaine, opiates and amphetamines in the period of January to November 2006, including the largest ever seizure of heroin in this country.[2] However, accurate estimations of community drug abuse are difficult to produce and current methods for the estimation of illicit drug use within Irish society often suffer from significant shortcomings. For example, performing surveys that focus upon specific subgroups known to abuse drugs, or attempting to gauge drug abuse by measuring the numbers presenting themselves for treatment. Of course, any information regarding the extent of the 'drug problem' in Ireland is inherently valuable, especially to those governing the allocation of resources to ensure adequate funding to the health services and law enforcement in order to aid in combating a growing problem.

Several analytical procedures exist in the peer-reviewed literature for the determination of illicit drugs. However, the majority of these methods are focused on personal testing as part of usage or abstinence monitoring and toxicological studies rather than community estimations of current usage. Due to the scope of such studies the matrix investigated has always been biological in nature, spanning across plasma and serum,[3–8] to urine[9–11] and other bodily fluids such as saliva and sweat.[12–16] Hair analysis has also become a recent focus in abstinence monitoring programs due to the ability to place a time frame to usage periods in accordance with standard hair growth.[17–21] To a lesser extent, reports for the determination of mother to child substance transfer have also been published, wherein meconium was the biological matrix under investigation.[22–23] A common feature of the published methods is the widespread use of liquid chromatography (LC) in conjunction with either electrospray ionisation (ESI) or atmospheric pressure chemical ionisation (APCI) tandem mass spectrometry (MS–MS) detection, allowing for separation followed by unequivocal detection and structural information. However, the methods mentioned above rely heavily upon invasive sampling procedures.

In the past decade, a number of reports concerning the presence of pharmaceuticals and other medicines in aquatic environments have entered the public domain, wherein classes of pharmaceuticals have been detected in both fresh and marine aquatic systems at trace and ultra-trace levels.[24–29] In some instances the measured environmental concentrations (MECs) demonstrated a statistical relationship concerning the levels of the detected pharmaceutical in the environment and that consumed by the population. For example, Ashton *et al.* determined the levels of common pharmaceuticals, *e.g.* diclofenac, ibuprofen, propranolol *etc.* in treated wastewater and also in the effluent receiving waters and found that levels detected downstream of the wastewater treatment plant statistically correlated to levels detected in the plant's discharged effluent and a further statistical correlation may exist to actual levels of said drug usage within the community.[30] Daughton hypothesised that in a similar manner to medicinal compounds, residues of illicit drugs may also be detected in the aquatic environment as a result of human usage and hence

[a] *National Centre for Sensor Research, School of Chemical Sciences, Dublin City University, Glasnevin, Dublin 9, Ireland. E-mail: brett.paull@dcu.ie; Fax: +353 1 700 5503; Tel: +353 1 700 5060*
[b] *NIVA–The Norwegian Institute for Water Research, Gaustadaleen 21, NO-0189 Oslo, Norway.*

† Electronic supplementary information (ESI) available: A detailed description of the development of the SPE procedure. See DOI: 10.1039/b702799k

discharge of both parent compounds and metabolites into aquatic systems is likely to occur *via* the municipal wastewater treatment system.[31]

The first report concerning the presence of illicit materials in treated wastewater appeared in 2004, whereby low ng L^{-1} of methamphetamine and 3,4-methylenedioxy methamphetamine (MDMA or 'ecstasy') were individually detected in the effluent of two monitored wastewater treatment plants in Nevada and South Carolina in the United States, respectively. Although no further research was conducted in this instance, it was acknowledged that the detection of these two illicit drugs corroborated evidence from the United States Drug Enforcement Agency (USDEA) that both chemicals were becoming increasingly problematic in the metropolitan areas surrounding the sampled plants.[32] More recently, amphetamine was also detected in sewage sludge by Austrian researchers in the low μg kg^{-1} range. However, caution must be used when interpreting such results, as amphetamine and similar compounds may also be presented in wastewater treatment plants as metabolites of prescription drugs used in the treatment of Parkinson's disease.[33]

Clearly the determination of illicit drug residues in wastewater could provide a possible opportunity to non-invasively estimate community wide consumption of such substances. Pioneering research on this front was published by Italian researchers in 2005 where levels of cocaine and its primary metabolite, benzoylecognine (BZE), detected in surface waters and treated wastewater was used to estimate consumption of the parent narcotic within the community.[34] The approach used was to convert MECs to daily doses per thousand population equivalents (PEq) using available information regarding the volume of wastewater through the treatment plant per day and a correction factor of 2.33 to adjust benzoylecgonine MECs in favour of the parent compound, cocaine.[34] Based upon their environmental monitoring data, Zucatto *et al.* estimated that on average within the general population approximately 7 doses per 1000 PEq were consumed daily, which was further refined to 27 doses per 1000 PEq aged in the range of 15–34 years.[34] A similar study was carried out along the River Thames in London, although the results of this study have not yet been published fully.[35,36] More recently, a study has emerged where the levels of a broad suite of illicit drugs were determined using isotope dilution LC–ESI–MS–MS in the treated effluents of wastewater treatment plants in Milan, Italy and Lugano, Switzerland.[37] In both plants the majority of the target analytes were detected in the influent samples, while reduced quantities were detected in the treated effluent, suggesting the existence of removal mechanisms within the treatment plant. Interestingly, the Milan wastewater treatment plant appeared to be extremely efficient at removing illicit drug residues, with most compounds, including cocaine, being removed to levels below the limit of quantitation (LOQ) of the developed analytical method.[37]

The aims of this research were therefore, to develop a suitably sensitive analytical method for the determination of a full suite of illicit drug residues, including cocaine, in treated wastewater and receiving waters in the Dublin and Greater Dublin area of Ireland, using solid phase extraction (SPE) with LC–MS–MS. Where residues of illicit drugs are detected, it was hoped to estimate community consumption data for the said narcotic using the approach of Zucatto *et al.*[34] Treatment plants within the City of Dublin, along with smaller plants serving areas in the north and south of the capital city were selected for study.

Experimental

Chemicals and reagents

Reagent water used throughout, unless otherwise stated, was obtained from a Millipore MilliQ water purification system (Millipore, Bedford, MA, USA) and was 18.2 MΩ or greater. Methanol and dichloromethane were purchased from Labscan (Dublin, Ireland), acetone and ethyl acetate were obtained from Aldrich (Gillingham, UK). All solvents used were HPLC grade. Dichlorodimethylsilane, ammonium formate, morpholinoethane sulfonate (MES), tris(hydroxymethyl)aminomethane (TRIS), ammonium acetate, acetic acid and formic acid were also purchased from Aldrich, (all ACS reagent grade). BDH Analar grade hydrochloric acid was used for sample pH adjustment (Poole, UK). Ammonium hydroxide solution, (LC–MS additive grade) was obtained from Fluka (Steinheim, Germany). Cocaine hydrochloride, morphine sulfate salt pentahydrate, methadone hydrochloride, ketamine hydrochloride, heroin and Δ^9-tetrahydrocannabinol methanolic solution (Δ^9-THC) were purchased under license from Sigma–Aldrich (St. Louis, MO, USA). Benzoylecognine hydrate, cocaethylene, D-amphetamine sulfate salt, Tempazepam, diazepam, fluoxetine hydrochloride, lysergic acid diethylamide (LSD), papaverine hydrochloride, 3,4-methylenedioxymethamphetamine hydrochloride (MDMA) and 2-ethylidine-1,5-dimethyl-3,3-diphenylpyrrolidine perchlorate (EDDP) were purchased under license from Sigma–Aldrich (Poole, UK). Individual 100 mg L^{-1} stock solutions of each chemical were prepared in methanol and were stored at 4 °C in the dark. Working solutions were prepared from the individual stock standards using water as a diluent.

Glassware preparation

Glassware was silanised in order to minimise non specific adsorption by rinsing with a solution of 10% v/v dichlorodimethylsilane in dichloromethane, followed by rinsing twice with dichloromethane and twice with methanol, respectively.

Sample collection

24 h composite samples of treated effluent were obtained from the wastewater treatment plants listed in Table 1, (with the exception of the Shanganagh treatment works where only a grab sample could be obtained). Fig. 1 shows the sampling sites chosen and their position in relation to Dublin. Corresponding influent samples were also collected from the Ringsend wastewater treatment plant in Dublin. Grab samples of receiving water were obtained immediately downstream of the treatment plants in an attempt to investigate the effect of dilution within the aquatic environment.

Samples from the plants were collected during the week beginning November 20th 2006. Samples were extracted

Table 1 Wastewater treatment plants sampled for the presence of illicit drugs

Treatment plant	Type of treatment[a]	Population served	Through flow/ L day^{-1}
Ringsend	1^y, 2^y	1 700 000	5.00×10^8
Swords	1^y, 2^y	48 000	1.00×10^7
Shanganagh	1^y only	65 000	2.20×10^7
Leixlip	1^y, 2^y	80 000	3.00×10^7
Navan	1^y, 2^y	40 000	1.05×10^7

[a] 1^y; primary treatment, 2^y; secondary treatment using activated sludge.

within 24 h of collection and the dried SPE cartridges were immediately frozen pending instrumental analysis.

Solid phase extraction

Sorbents investigated for sample extraction included Phenomenex Strata-X™, Strata-XC™ and Strata-XCW™, all 200 mg sorbent mass pre-packed into 6 mL cartridges (Phenomenex, Macclesfield, Cheshire, UK). From initial investigations the Strata-XC™ sorbent provided the highest degree of analyte recovery and hence, was considered for further study. Prior to extraction 500 mL aqueous samples were filtered through Whatman GF–C glass micro-fibre filters to remove particulate matter (Whatman, Maidstone, UK). The filtrate was then adjusted to pH 6.0 using HCl. Prior to use, the SPE cartridge was conditioned with 2 × 6 mL of methanol and 2 × 6 mL of water, respectively. Samples were introduced by vacuum through Teflon tubing and extracted under an operating pressure of 20″ Hg on a vacuum manifold. After complete sample introduction, but without letting the cartridge run dry, the sorbent was washed with 50 mL of 10% v/v methanol in 100 mM formic acid. Finally, 500 μL of glacial acetic acid was then added to each cartridge, which was allowed to slowly percolate through the sorbent bed in order to aid with dehydration. The sorbents were then dried by vacuum aspiration for a minimum of 30 min. Elution was performed using 10 mL of 5% v/v ammonium hydroxide in 1 : 1 acetone : ethyl acetate, the elution solvent was allowed to slowly percolate through the sorbent bed and was collected in a 12 mL glass vial. The collected eluate was then reduced in volume to near dryness under a gentle stream of N_2 with heating if necessary. The resulting residue was then reconstituted in 200 μL of 30% v/v methanol in 5 mM ammonium acetate pH 4.5 fortified with 0.1 mg L^{-1} papaverine that served as an internal standard for quantitation and was transferred to an autosampler vial containing a low volume polypropylene insert for LC–MS–MS analysis.

Fig. 1 Sampling sites (☆) at wastewater treatment plants within the Greater Dublin area.

Liquid chromatography tandem mass spectrometry

LC–MS–MS analyses were performed as previously reported.[38] Product ion MS–MS transitions were used for qualitative confirmation. Quantitation was also performed using the area from the resulting product ion peak. Analytical separations were performed on a 200.0 × 3.0 mm i.d. Phenomenex Onyx monolithic C_{18} column, (Phenomenex, Macclesfield, Cheshire, UK) using the multi-step linear gradient of methanol and 5 mM ammonium acetate pH 4.5.[38]

Results and discussion

Analytical method performance

Prior to application, the performance characteristics of the developed SPE LC–MS–MS method were determined using Boyne river water as a sample matrix. Linearity was assessed using seven point curves prepared by extracting mixed analyte spike solutions in the region of 0.01 to 1 μg L^{-1} (concentrations quoted are those prior to extraction). Repeatability was determined by performing six replicate injections of a 0.20 μg L^{-1} mixed extract while reproducibility was examined using six individually extracted 1 μg L^{-1} mixed analyte solutions. The limits of detection (LOD) and LOQ were defined as signals corresponding to 3σ and 10σ, respectively, of the baseline noise in each of the extracted MS–MS production ion transition traces. The determined performance characteristics are listed in Table 2.

Linearity data were excellent within the range of concentrations extracted, calculated regression coefficients were greater than 0.99 in all cases with the exception of cocaethylene, methadone and fluoxetine, although even in these instances regression coefficients greater than 0.95 were achieved. The upper limit of linearity was set at 1 μg L^{-1}, as it was previously reported by Castiglioni *et al.* that deviations from linearity were observed when similar levels were exceeded when using MS–MS detection.[37] Extraction repeatability was determined

Table 2 Method performance data for the developed SPE LC–MS–MS procedure

Analyte	Linearity (R^2)	Precision		Sensitivity		Recovery (% ± SD)
		Intra assay (%RSD)	Inter assay (%RSD)	LOD/ng L^{-1}	LOQ/ng L^{-1}	
Morphine	0.9951	7.10	4.13	257	856	4 ± 0
MDMA	0.9967	4.92	5.22	7	22	52 ± 1
Benzoylecognine	0.9940	4.53	5.70	1	2	53 ± 3
Ketamine	0.9975	6.44	1.73	1	4	51 ± 3
Cocaine	0.9980	6.64	2.15	1	2	56 ± 2
Cocaethylene	0.9535	6.93	5.79	1	5	65 ± 3
LSD	0.9967	5.29	5.96	3	10	51 ± 3
EDDP	0.9929	6.07	3.69	2	7	59 ± 2
Methadone	0.9771	8.08	2.34	4	14	55 ± 0
Fluoxetine	0.9621	6.19	6.74	93	312	33 ± 2
Temazepam	0.9978	7.79	7.21	7	23	59 ± 3
Diazepam	0.9923	7.56	6.95	38	127	55 ± 3

using a low level spiked standard, 200 ng L^{-1}, and was on average in the range of 5–8% RSD for the six replicate extract injections. Reproducibility was determined using a higher spiking level and was in the region of 2–6% RSD for the six replicate extractions, both determined levels of precision compare well with those of Castiglioni *et al.* who achieved levels of precision $<10\%$ for repeatability and $<5\%$ for reproducibility using low mg L^{-1} level mixed analyte solutions prepared in wastewater, however, without any preconcentration.[37]

Analyte recovery was determined to be in the region of 50–65% for the most analytes. Extraction repeatability denoted by the quoted standard deviation values in Table 2 was acceptable.

Detection limits presented in Table 2, calculated as 3σ of the peak-to-peak baseline noise in each of the extracted ion chromatograms for each analyte MS–MS transition using river water as a sample matrix were all found to lie in the low ng L^{-1} region. LODs calculated compare favourably with those achieved by both Zucatto *et al.* and Castiglioni *et al.*[34,37] The calculated LOD for diazepam was higher than expected due to the low intensity of the m/z 257 MS–MS product ion. However, similar sensitivity data was obtained when calculated using the extracted ion chromatogram for the pseudo-molecular ion; m/z 285, (LOD: 36 ng L^{-1}, LOQ: 120 ng L^{-1}), and therefore due to the higher level of specificity imparted, it is recommended that the MS–MS product ion would still be used for all quantitative purposes.

Determination of illicit drug residues in wastewater and surface waters

The developed analytical method was applied for the determination of illicit drug residues in wastewater treatment plants and their receiving waters in Dublin and also in the surrounding counties of Meath and Kildare, both of which have experienced large increases in population in the last decade. In Dublin, wastewater treatment plants investigated included the Ringsend Wastewater Treatment Works, which is located at the mouth of the River Liffey on the south side of Dublin Bay and caters for all of the Greater Dublin metropolitan area (1.7 million PEq), the Swords Wastewater Treatment Works in North County Dublin, which discharges into the River Broadmeadow prior to its entry into the Malahide Estuary, and the Shanganagh Wastewater Treatment Works which is located in South County Dublin, which discharges 1.6 km offshore from Killiney Beach into the Irish Sea. Samples of treated effluent were also collected from Leixlip Wastewater Treatment Works in County Kildare which serves the towns of Leixlip, Maynooth, Celbridge and Kilcock, from which treated effluent is discharged directly into the River Liffey, and the Navan Wastewater Treatment Centre in County Meath which serves the town of Navan and discharges treated effluent into the River Boyne. All of the plants sampled employ both primary and secondary treatment using activated sludge, with the exception of the Shanganagh Wastewater Treatment Works which offers primary screening only. The Ringsend Wastewater Treatment Works also offers tertiary treatment using ultraviolet irradiation, however, tertiary disinfection is only employed during the bathing season (May to September), and therefore samples collected were not exposed to UV light.[39] Official weather data for the weekend of the 18–19th November was obtained from the Irish Meteorological Service wherein total precipitation at the Dublin Airport monitoring station was 5.3 mm.[40] Collected samples were analysed as previously described and levels of analytes detected are presented in Table 3.

From the list of target analytes only morphine, BZE, cocaine, EDDP and Tempazepam were detected in the collected wastewater and surface water samples. Morphine was detected in relatively high concentrations in the treated effluents of the Swords and Navan wastewater treatment plants. However, its presence could be attributable to medicinal use of morphine and related opiates rather than consumption of illicit heroin. Morphine was not detected in the influent of the Ringsend Wastewater Treatment Works but was present at a detectable level below the LOQ in the corresponding effluent sample. Such an observation may occur due to cleavage of glucuronide metabolites of morphine during the treatment process.[26,37]

Cocaine and its primary urinary metabolite, BZE were detected in both the influent and effluent samples of the Ringsend Wastewater Treatment Works and also in the grab sample collected from the Shanganagh Wastewater Treatment Works. Cocaine was also detected at low ng L^{-1} levels in the treated effluent of the Navan and Leixlip treatment plants. The combined presence of cocaine and BZE provides a reliable

Table 3 Concentrations (ng L^{-1}) of illicit drugs detected in collected wastewater and surface water samples

Sample	Morphine	BZE	Cocaine	EDDP	Temazepam
Ringsend influent	—	290 ± 11	489 ± 117	—	320 ± 56
Ringsend effluent	<LOQ	22 ± 4	138 ± 20	48 ± 1	126 ± 14
Swords effluent	874 ± 86	—	—	206 ± 10	—
River broadmeadow	—	—	25 ± 7	—	—
Shanganagh effluent	—	31 ± 18	77 ± 25	—	—
Killiney Beach	—	—	—	—	—
Leixlip effluent	<LOQ	—	47 ± 10	9 ± 1	106 ± 3
River Liffey	—	—	33 ± 11	—	—
Navan effluent	452 ± 86	—	111 ± 15	67 ± 10	—
River Boyne	—	—	—	—	—

indication of human consumption of cocaine. However, unlike previous reports from Zucatto *et al.* and Castiglioni *et al.* the levels of cocaine detected in the present study were higher than those of BZE.[34,37] Previous pharmacokinetic studies reveal that cocaine is readily metabolised into BZE, and therefore it was expected that higher quantities of BZE should be present compared to those of cocaine.[41] Reasons for the observed lower BZE concentrations are speculative, although it may be related to loss during the sample filtering pretreatment step. As BZE exists in the wastewater solution (pH 6.2–6.4) as a neutral zwitterion, whereas cocaine is present in cationic form, there may be a bias resulting from sorption onto particulates.

To test for cocaine stability within the wastewater samples a simple experiment was performed wherein a 10 μg L^{-1} solution of cocaine was prepared in a collected influent sample (pH 6.23) and stored in the dark at 4 °C. This solution was sampled after a period of 0, 24 and 48 h respectively, and analysed using direct injection LC–MS–MS. There was no significant negative change in the peak area of cocaine measured over the course of the 48 h period, and consequently cocaine appears to undergo only limited hydrolysis under such conditions.

Based upon the levels of both BZE and cocaine detected in the influent and effluent of the Ringsend Wastewater Treatment Works, it is estimated that the removal rates within the plant on the day of sampling were 93% and 72% for BZE and cocaine, respectively. Levels of cocaine and BZE detected in the wastewater samples were of a similar magnitude to those reported by Castiglioni *et al.* when analysing the effluents of wastewater treatment plants in Italy and Switzerland.[37]

Interestingly, cocaine was also detected in the samples (collected near the point of discharge) of the River Broadmeadow and the River Liffey. However, the levels detected were lower than those detected in the treated effluent samples, therefore suggesting dilution within the receiving water. It would be expected that levels detected would decrease significantly with increasing distance from the discharge point. Such an effect can be seen in the case of the River Boyne wherein the sample, in which no illicit drug residues were detected, was collected approximately 4 km downstream from the Navan Wastewater Treatment Centre.

EDDP, the primary metabolite of methadone, was detected in a number of effluent samples in low ng L^{-1} concentrations, although methadone itself was not detected in any of the collected samples. The presence of EDDP in the absence of methadone is somewhat surprising, although Castiglioni *et al.* detected EDDP in excess of methadone in a ratio of ~2 : 1 in wastewater treatment plants studied.[37] EDDP presence in effluent samples may relate to its existence as a charged species in solution, and would therefore be expected to persist in the aqueous phase during wastewater treatment.

Temazepam was detected in both the influent and the effluent samples of the Ringsend Wastewater Treatment Works and also in the effluent of Leixlip Wastewater Treatment Works. Temazepam is a prescribed medication and was included in the list of target analytes along with diazepam as both sedatives have the potential for abuse. However, its presence can also be a result of medicinal and therapeutic use rather than illicit or abused consumption (no information on prescription rates for Temazepam were freely available to correlate data). As was noted for both BZE and cocaine, the level of Tempazepam detected in the Ringsend treatment plant decreased from influent to effluent suggesting a removal of approximately 60% *via* either sorption or degradation during the passage of Tempazepam through the treatment works.

For the purposes of illustration, Fig. 2 depicts MS–MS product ion chromatograms showing (A) the presence of BZE and cocaine in the influent and effluent of the Ringsend Wastewater Treatment. The excellent retention time reproducibility should also be noted from Fig. 2(A) along with the clear reduction of the levels of both substances detected in the effluent sample compared to the influent sample. Fig. 2(B) depicts the suspected presence of EDDP in the effluent sample collected from the Navan Wastewater Treatment Centre (the term 'suspected' is used only due to the absence of methadone in the sample). However, Fig. 2(B) unequivocally shows the presence of trace levels of EDDP in the collected sample.

Estimation of community consumption of cocaine from environmental data

Having detected levels of cocaine in the influents and effluents of several of the examined wastewater treatment plants, an attempt was made to estimate consumption of cocaine within the community served by those treatment works.

The approach used in this instance was based upon that of Zucatto *et al.*, however, as previously mentioned, cocaine was found in greater quantities in this study than its primary urinary metabolite BZE, and therefore cocaine was used for the community consumption calculations rather than BZE.[34] Such an approach includes two assumptions: firstly, that the relative stability of the cocaine molecule in the wastewater solution (tested above); and secondly, that the source of cocaine within the wastewater comes from human excretion rather than 'dumping' of large quantities of the drug itself into the wastewater system. The latter, although an event unlikely

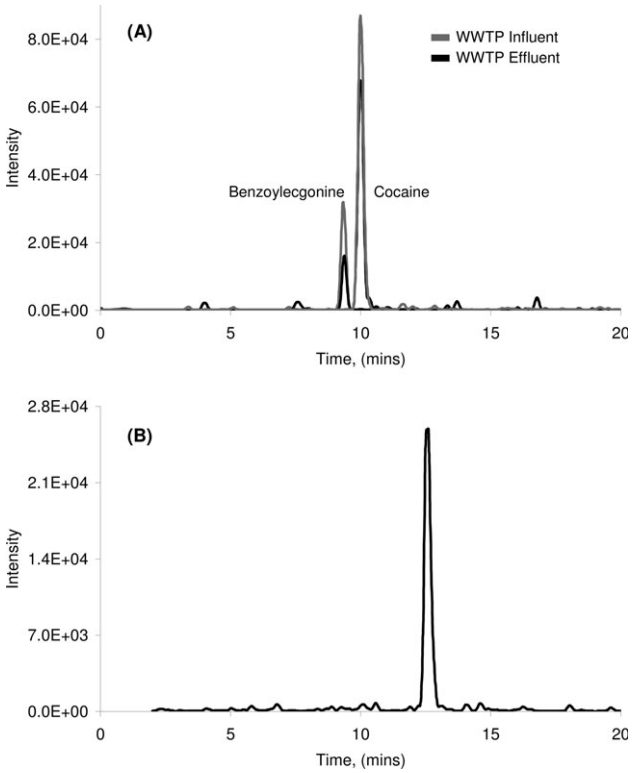

Fig. 2 (A) The presence of ng L^{-1} quantities of benzoylecognine and cocaine in the influent and effluent of the Ringsend Wastewater Treatment Works, and (B) the presence of ng L^{-1} quantities of EDDP in the treated effluent of the Navan Wastewater Treatment Centre.

to coincide with the sampling period, should be noted as a possibility.

From the levels of cocaine detected, it was possible to calculate the load of cocaine within the treatment plant on the day in which the sample was collected simply by multiplying the analytical data by the flow through the plant. This data is presented in Table 4, column 2. However, a further refinement of the data can be performed when considering the pharmacokinetic behaviour of cocaine, wherein approximately 10% of the parent dose ingested is excreted as cocaine within urine.[42] Therefore, assuming cocaine observed comes from human waste only, these data can be multiplied further to give the equivalent consumed cocaine per day.

It should be noted that the calculated results shown in Table 4 are expected to be conservative when considering the high probability that removal within the wastewater transport system or treatment plant may have occurred prior to collection of the sample. The cocaine load value quoted for the Ringsend Wastewater Treatment Works refers to that determined in the influent sample in order to minimise extra variability, such as analyte removal during treatment *etc*. The plant caters for the entire Dublin metropolitan area, (1.7 million PEq) and the sample collection period corresponds to the weekend, a time in which cocaine consumption would be thought to be at a maximum.

Table 4 Cocaine loads within the sampled wastewater treatment plants

Wastewater treatment plant	Cocaine load/ g day^{-1}	Equivalent consumed cocaine/g day^{-1}
Ringsend	224.50	2245.00
Shanganagh	1.69	16.90
Leixlip	1.41	14.10
Navan	1.17	11.70

Table 5 Estimated community consumption of cocaine within the catchments served by the sampled wastewater treatment plants

	Estimated cocaine consumption	
Catchment	Per 1000 PEq/ g day^{-1}	Per 1000 PEq/ doses day^{-1}
Ringsend	1.44	14.38
Shanganagh	0.26	2.61
Leixlip	0.18	1.76
Navan	0.29	2.91

From the cocaine load within the plant it is possible to estimate the use of cocaine by the population catchments served by the wastewater treatment plants. Combining the data from Tables 1 and 4, the estimated use of cocaine within the community is presented in Table 5.

The results presented in Table 5 compare favourably with those previously published by Zucatto *et al.*[34] The dose per day data presented in Table 5 were calculated based upon information presented by Drummer[42] which states that one average dose equals approximately 100 mg cocaine. Zucatto *et al*. further refined their data to include a positive bias against young adults in the age bracket of 15 to 34 years of age due to a reported higher consumption of cocaine by persons within this age bracket when compared to the general population. It was hoped to undertake a similar transformation in this study. However, official statistics available from the Central Statistics Office concerning the Irish population census 2006 are only preliminary in nature, and record increases in the population of Ireland in the catchment areas studied, since the last complete census, would make any such calculations unreliable.[43]

The levels of cocaine consumption estimated in this approach suggest that significant quantities of the drug are being consumed within Dublin but it can also be said that the problem of cocaine consumption is not wholly confined to the capital city but present in smaller towns outside Dublin. The detection of cocaine and its urinary metabolite in the wastewater treatment plants adds weight to the assumption that the presence of cocaine arises due to human consumption and not improper disposal of the narcotic. The problem exists in Ireland that there are currently no reliable statistics regarding cocaine consumption to compare the generated data against. The developed analytical procedure could be applied for the non-invasive routine monitoring of cocaine consumption within the community in order to assess whether usage trends are generally stable or sporadic, particularly for example during weekend and public holiday periods.

Conclusions

A suitably sensitive and validated analytical method using SPE LC–MS–MS for the determination of residues of illicit

drugs in wastewater and surface water has been described. Cocaine and corresponding metabolite residues were detected in samples of wastewater and subsequent receiving waters collected from treatment plants in Dublin city and surrounding counties. From the environmental analytical data it was possible to estimate community consumption of cocaine within the catchment areas of the sampled wastewater treatment plants. Estimations of cocaine consumption as doses per 1000 PEq per day compare well with previously published data from Italian researchers.

References

1 M. O'Halloran, *Cocaine seizures rise eight fold*, The Irish Times, Saturday October 1st 2005.
2 The Press Releases Section of the website of An Garda Síochána, viewed 5th December 2006, http://www.garda.ie/angarda/press.html.
3 G. Singh, V. Arora, P. T. Fenn, B. Mets and I. A. Blair, *Anal. Chem.*, 1999, **71**, 2021.
4 R. J. Lewis, R. D. Johnson, K. M. Angier and R. M. Ritter, *J. Chromatogr., B: Anal. Technol. Biomed. Life Sci.*, 2004, **806**, 141.
5 F. Sadeghipour and J.-L. Veuthey, *J. Chromatogr., A*, 1997, **787**, 137.
6 M. Concheiro, A. de Castro, O. Quintela, M. Lopez-Rivadulla and A. Cruz, *J. Chromatogr., B: Anal. Technol. Biomed. Life Sci.*, 2006, **832**, 81.
7 M. R. Moeller, S. Steinmeyer and T. Kraemer, *J. Chromatogr., B: Biomed. Appl.*, 1998, **713**, 91.
8 H. Z. Rofael and M. S. Abdel-Rahman, *J. Appl. Toxicol.*, 2002, **22**, 123.
9 K.-C. Wang, T.-S. Shih and S.-G. Cheng, *Forensic Sci. Int.*, 2005, **147**, 81.
10 B. Maralikova and W. Weinmann, *J. Chromatogr., B: Anal. Technol. Biomed. Life Sci.*, 2004, **811**, 21.
11 V. Bianchi and G. Donzelli, *J. Chromatogr., B: Biomed. Appl.*, 1996, **675**, 162.
12 R. Dams, C. M. Murphy, R. E. Choo, W. E. Lambert, A. P. de Leenheer and M. A. Huestis, *Anal. Chem.*, 2003, **75**, 798.
13 P. Kintz, M. Villain, M. Concheiro and V. Cirimele, *Forensic Sci. Int.*, 2005, **150**, 213.
14 M. Wood, M. Laloup, M. del Mar Remirez Fernandez, K. M. Jenkins, M. S. Young, J. G. Ramaekers, G. de Boeck and N. Samyn, *Forensic Sci. Int.*, 2005, **150**, 227.
15 K. A. Mortier, K. E. Maudens, W. E. Lambert, K. M. Clauwaert, J. F. van Bocxlaer, D. L. Deforce, C. H. van Peteghem and A. P. de Leenheer, *J. Chromatogr., B: Anal. Technol. Biomed. Life Sci.*, 2002, **779**, 321.
16 D. A. Kidwell, J. C. Holland and S. Athanaselis, *J. Chromatogr., B: Biomed. Appl.*, 1998, **713**, 111.
17 K. M. Clauwaert, J. F. van Bocxlaer, W. E. Lambert, E. G. van den Ecekhout, F. Lemierem, E. L. Esmans and A. P. de Leenheer, *Anal. Chem.*, 1998, **70**, 2336.
18 K. B. Scheidweiler and M. A. Huestis, *Anal. Chem.*, 2004, **76**, 4358.
19 F. Musshoff, F. Driever, K. Lachenmeier, D. W. Lachenmeier, M. Banger and B. Madea, *Forensic Sci. Int.*, 2006, **156**, 118.
20 M. Cheze, G. Duffort, M. Deveaux and G. Pepin, *Forensic Sci. Int.*, 2005, **153**, 3.
21 M. Villain, M. Concheiro, V. Cirimele and P. Kintz, *J. Chromatogr., B: Anal. Technol. Biomed. Life Sci.*, 2005, **825**, 72.
22 J. Gareri, J. Klein and G. Koren, *Clin. Chim. Acta*, 2006, **366**, 101.
23 S. Pichini, R. Pacifici, M. Pellegrini, J. Lozano, J. Murillo, O. Vall and O. Garcia-Algar, *Anal. Chem.*, 2004, **76**, 2124.
24 C. G. Daughton and T. A. Ternes, *Environ. Health Perspect.*, 1999, **107**, 907.
25 T. A. Ternes, *Water Res.*, 1998, **322**, 3254.
26 M. J. Hilton and K. V. Thomas, *J. Chromatogr., A*, 2003, **1015**, 129.
27 S. Zuehlke, U. Duennbier and T. Heberer, *Anal. Chem.*, 2004, **76**, 6548.
28 P. H. Roberts and K. V. Thomas, *Sci. Total Environ.*, 2006, **356**, 143.
29 P. H. Roberts and P. Bersuder, *J. Chromatogr., A*, 2006, **1134**, 143.
30 D. Ashton, M. Hilton and K. V. Thomas, *Sci. Total Environ.*, 2004, **333**, 167.
31 C. G. Daughton, Illicit Drugs in Municipal Sewage, in *Pharmaceuticals and Personal Care Products in the Environment: Scientific and Regulatory Issues*, ACS Symposium Series, The American Chemical Society, 2001, p. 348.
32 T. L. Jones-Lepp, D. A. Alvarez, J. D. Petty and J. N. Huckins, *Arch. Environ. Contam. Toxicol.*, 2004, **47**, 427.
33 A. Kaleta, M. Ferdig and W. Buchberger, *J. Sep. Sci.*, 2006, **29**, 1662.
34 E. Zucatto, C. Chiabrando, S. Castiglioni, D. Calamari, R. Bagnati, S. Schiarea and R. Fanelli, *Environ. Health*, 2005, **4**, 14.
35 J. Orr and N. Goswami, *River of cocaine*, The Telegraph, London, November 6th 2005.
36 K. Ross, *Anal. Chem.*, 2006, **78**, 13.
37 S. Castiglioni, E. Zucatto, E. Crisci, C. Chiabrando, R. Fanelli and R. Bagnati, *Anal. Chem.*, 2006, **78**, 8421.
38 J. Bones, M. Macka and B. Paull, *Analyst*, 2007, **132**, 208.
39 Dublin Drainage Consultancy, *Greater Dublin Strategic Drainage Study—Assessment of Existing Wastewater Treatment Works*, July 2002, available online at http://www.fingalcoco.ie/YourLocalCouncil/Services/Waterservices/GreaterDublinStrategicDrainageStudy, appendix E, last viewed December 15th 2006.
40 Dublin Airport weather station data for the 18/19/20th November 2006 available through http://www.meteireann.ie/climate/dailydata.asp, viewed 18th December 2006.
41 J. Ambre, *J. Anal. Toxicol.*, 1985, **9**, 241.
42 O. H. Drummer, *Forensic Sci. Int.*, 2004, **142**, 101.
43 The Central Statistics Office, *Census 2006–Preliminary Report*, available through http://www.cso.ie, viewed December 21st 2006.