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Active Pharmaceutical Ingredients Entering the Aquatic Environment From Wastewater Treatment Works: A Cause for Concern?

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Abstract

This work reports on the variation in wastewater treatment works (WwTW) influent concentrations of a wide variety of active pharmaceutical ingredients (APIs), their removal efficiency, effluent concentrations and potential risks to the aquatic environment. The research is based on data generated from two large UK-wide WwTW monitoring programmes. Taking account of removal of parent compound from the aqueous phase during treatment in combination with estimates of dilution available it is possible to prioritise the APIs of greatest risk of exceeding estimates of predicted no effect concentrations (PNEC) in receiving waters for all WwTW in the UK. The majority of substances studied were removed to a high degree, although with significant variation, both within and between WwTW. Poorer removal (between influent and effluent) was observed for ethinyloestradiol, diclofenac, propranolol, the macrolide antibiotics, fluoxetine, tamoxifen and carbamazepine. All except the last two of these substances were present in effluents at concentrations higher than their respective estimated PNEC (based on measurement of effluents from 45 WwTW on 20 occasions). Based on available dilution data as many as 890 WwTW in the UK (approximately 13% of all WwTW) may cause exceedances of estimated riverine PNECs after mixing of their effluents with receiving waters. The overall degree of risk is driven by the toxicity value selected, which in itself is controlled by the availability of reliable and relevant ecotoxicological data and

consequently the safety factors applied. The dataset and discussion, provides information to assist in the future management of these types of chemicals.

Key words: pharmaceuticals, API, wastewater, effluent, fate, risk assessment

1. INTRODUCTION

The use and environmental prevalence of pharmaceuticals increases on an annual basis due to a variety of reasons including the widening array of medical treatments available, greater availability of medicines across the world, affordability, population growth, population ageing (in some countries) and changing perspectives towards, for example, pain (Jelic et al., 2011). Active Pharmaceutical Ingredients (API) are detected throughout the environment in water, soil, sediment, sludge as well as in drinking waters in some countries (Kasprzyk-Hordern et al., 2008; Zorita et al., 2009; Wahlberg et al., 2011; Jones et al., 2014; Lees et al., 2016). Although the mere presence of pharmaceutical is not always associated with harm to the environment or human health, concerns are rising associated with antimicrobial resistance and chronic impacts on biodiversity including endocrine disrupting effects on fish (Levado et al, 2004; Jobling et al., 2005; Tyler et al., 2008). The main source of occurrence of APIs in the river environment is from human use of pharmaceuticals, via the continuous discharge of effluent from the Wastewater Treatment Works (WwTW) (Gardner et al., 2012; Melvin et al., 2016). Hence, investigating the occurrence, fate and risk of APIs is currently of great interest to regulators and the water industry alike, with a focus to better understand the loadings entering WwTW and the observed within and between works variation in removal efficiencies and concentrations often observed for APIs (Gardner, 2013).

The range of concentrations found for pharmaceuticals studied in the UK is similar to that observed in continental Europe as well as in the USA (Kolpin et al., 2002; Ashton et al., 2004; Hope et al., 2012; Bradley et al., 2016; Burns et al., 2017). Table 1 provides examples of other reported data for APIs determined as part of this research, rather than a complete list of all APIs detected in effluent and receiving waters. Other studies have also shown that there is a clear association between the number of pharmaceuticals used in a society and the levels of API found in receiving water bodies ranging from API concentration of typically less than 100 ng/l in the surface and groundwater and below 50 ng/l in treated drinking water (WHO, 2011; Furlong et al., 2017) to higher levels reported adjacent to production facilities (Phillips et al., 2010). Predicted no effect concentrations (PNECs) have been reported for some APIs below 1 ng/l and APIs such as diclofenac (CAS 15307-79-6), 17-beta-

74 estradiol (E2) (CAS 50-28-2) and 17-alpha-ethinylestradiol (EE2) (CAS 57-63-6) are on the European
75 Water Framework Directive (WFD) 'watch list' (EU, 2013). This requires member states to gather
76 monitoring data in order to assess risk to the environment, leading to significant sources of APIs
77 needing to be quantified and factors controlling the discharge of APIs carefully considered along with
78 impacts on receiving water ecology, including effects of mixtures (Bound and Voulvoulis, 2006).
79

Table 1. Average aquatic concentrations for APIs of interest to this research found in river water, as well as usage, excretion and removal in WwTW.

API	Therapeutic Class	Upstream (µg/l)	Influent (µg/l)	Effluent (µg/l)	WwTW removal (%)	Down stream (µg/l)	UK consumption (ton/year), 2009 and 2011	Excreted unchanged compound (%)
Aspirin (acetylsalicylic acid)	Anti-inflammatory/analgesics	NA	NA	NA	NA	<0.0005[b]	130[d]	<1[b]
Atenolol	Beta blocker	NA	NA	NA	NA	0-0.56[b]	28[e]	90[f]
Azithromycin	Antibiotic	NA	0.163[l]	0.030[l]	90[l]	NA	NA	NA
Carbamazepine	Antiepileptic	NA	2.593[b]	3.117[b]	ND[b]	0.0005-0.356[b]	48[e]	3[b]
Ciprofloxacin	Antibiotic	NA	1.090[l]	0.052[l]	97[l]	NA	NA	NA
Clarithromycin	Antibiotic	NA	0.524[l]	0.092[l]	91[l]	NA	NA	NA
Diclofenac	Anti-inflammatory	<0.020[a]	0.107-0.981[c]	0.599[a]	70-92[c]	0.154[a]	28[e]	15[f]
Erythromycin	Antibiotic	<0.010[a]	2.0[k]	0.109[a]	25-91[i]	0.159[a]	3[d]	25[f]
Oestrogen (E1)	Natural hormone	NA	0.042[g]	0.011-0.025[g]	58-96[g]	NA	NA	NA
Oestradiol (E2)	Contraceptive	NA	0.016[g]	0.0013-0.0039[g]	89-96[g]	NA	NA	NA
Ethinylestradiol (EE2)	Contraceptive	NA	0.0017[g]	0.00033-0.00078[g]	53-71[g]	NA	NA	NA
Fluoxetine	Psychiatric drugs	NA	0.070[k]	0.023[j]	33-100[h]	NA	6.4[m]	NA
Ibuprofen	Analgesic	0.432[a]	14.0[k]	4.201[a]	90-100[i]	1.105[a]	258[e]	10[f]
Oxytetracycline	Antibiotic	NA	1.09[l]	0.029[l]	99[l]	NA	NA	NA
Ofloxacin	Antibiotic	NA	0.081[l]	0.023[l]	89[l]	NA	NA	NA
Propranolol	Antihypertensive	0.010[a]	0.542[b]	0.093[a] 0.388[b]	28[b]	0.041[a]	15[e]	<0.5[b]
Tamoxifen	Anti-cancer	<0.010[a]	0.0002-0.015[c]	<0.010[a]	32-45[c]	<0.010[a]	NA	NA

ND = not detected; NA = not available. [a]Ashton et al., 2006; [b]Kasprzyk-Hordern et al., 2008; [c]Zhou et al., 2009; [d]2006 sales data for Wales; Kasprzyk-Hordern et al., 2008; [e]IMS figure on active ingredient sales; [f]WHO, 2011; [g]Heffley et al., 2014; [h]Clara et al., 2005; [i]Li et al., 2014; [j]Gardner et al., 2012; [k]Gardner et al., 2013;[l] Singer et al., 2014; [m] Boxall et al., 2014

Many countries have therefore started monitoring programs to investigate the exposure of APIs in order to gain a better understanding of their sources, fate and risk (Falås et al., 2012). The Chemical Investigation Program (CIP) in the UK is a large ongoing investment being undertaken by the water industry to assist the UK in meeting its obligations under the WFD to monitor concentrations of priority chemicals including APIs in WwTW influent, intermediate processes and effluent as well as assessing their risk to receiving waters (Gardner et al., 2013). The first phase of the CIP (named CIP1 here) was a project that ran from 2012-2015 with one of its aims to investigate the fate of trace substances (including 11 APIs) in influent, effluent and intermediate WwTW processes of 25 WwTW. Some of results from this program have been reported previously (Gardner et al., 2012, Gardner et al.; 2013, Jones et al.; 2013 and Comber et al., 2014). The £140 million investment in the second phase of the CIP (labelled CIP2 in this work) program builds on the outputs from CIP1 but extends the range of WwTW monitored and the number of determinands in order to in some cases measure (for WFD priority substances and priority hazardous substances) and in some cases predict (for emerging

chemicals such as APIs) the impact on receiving waters. The CIP2 determinands include 19 APIs and 4 metabolites at currently 45 WwTW on 20 occasions. In total, over 60 000 samples are to be taken, with over 2 million determinations. This study reports on the findings for APIs from the CIP1 and CIP2 programmes.

WwTWs are primarily designed to serve the purpose of removing pathogens, suspended solids and gross organic and inorganic matter, rather than the removal of the increasing numbers of modern chemicals generally present in the µg/l range or less (Melvin, 2016). It has also been observed that there is a wide variation in removal rates for different substances, both within and between WwTWs. This difference in removal rate creates large uncertainty factors for the prediction and modeling of effluent concentrations and therefore creates a challenge in conducting meaningful risk assessments. There are currently no statutory consents applied to APIs in WwTW effluent, however, there is an urgent need to better understand the risk posed by APIs in effluents to receiving waters in order to inform future investment and to design and implement better risk assessment (Gardner, 2013). The presence of APIs is not measured on a routine basis for most WwTWs owing to cost and lack of legislative drivers. Consequently, there are a number of previous studies modelling the impact of APIs based on consumption, WwTW removal and dilution but the cost of analysis generally prevents the actual measurement of APIs in effluent (Johnson et al., 2013a,b; 2015).

This study utilizes CIP 1 (11 APIs, from 25 WwTW sampled on up to 15 occasions) and the more recent CIP2 program (19 parent APIs and 4 metabolites, from WwTW sampled on 20 occasions). Although the APIs studied represent only a fraction of the total APIs in use, financial and practical constraints associated with sampling, preservation, analysis and replication meant the number of determinands needed to be controlled. However, APIs were prioritised on potential risk to the aquatic environment and all of the main classes of API have been represented (Table A1). Concentrations in the WwTW effluent have been compared with derived PNECs in receiving waters in order to generate a priority list of APIs of potential concern.

2. MATERIALS AND METHODS

2.1 Selection of Pharmaceuticals

The selection of chemicals for CIP1 is discussed elsewhere (Gardner et al., 2012). The list of candidate APIs for inclusion in CIP2 was based primarily on a prioritization study undertaken by UKWIR in 2014 (UKWIR, 2014). Unlike many previous prioritisations, which focused on usage and concentrations detected in surface waters/effluents, problem sites or substances, this study adopted a risk assessment approach by comparing the estimated environmental concentrations of nearly 150 pharmaceuticals (screened on usage and perceived hazard from a list of thousands of candidate

substances) with data for their respective effect concentrations on a variety of receptor organisms in the aquatic environment.

For the purposes of CIP2, this list was further refined by selection of substances that were considered to have the greatest potential as candidate WFD priority substances. The criteria for this selection were a) that the risk characterisation ratio (predicted concentration divided by the highest probable no effect concentration (PEC/PNEC) ranked higher than 1 in the overall 2014 UKWIR prioritisation and b) that the data supporting the derivation of a PNEC were relatively reliable and complied with the WFD approach to PNEC derivation (EU 2011). In effect, this meant that PNECs were derived using experimental rather than modelled effects, long-term effects in organisms from different trophic levels were available (though short term exposure was also considered) and assessment factors were applied according to WFD guidance (EU 2011).

The APIs prioritised were then further reviewed for their relevance to wastewater treatment, and the likelihood that the substance might be present in sewage effluents and hence discharged to surface waters (rather than being partitioned to sewage sludge). This resulted in the list of substances tabulated in Table A1 of the Electronic Supplementary Information (ESI). For the purposes of estimating risks, the PNEC values derived in the UKWIR prioritization (UKWIR 2014) were then re-examined and (where available) they were substituted with the latest estimates derived by the EU Joint Research Centre (JRC, 2015), by the pharmaceutical industry (Astra Zeneca, 2016; NSF, 2016) or published in the open literature (Murray-Smith et al., 2012). Where no PNEC was available from these sources, the ecotoxicology data applied in deriving the PNECs reported by UKWIR (UKWIR 2014) were used to deterministically estimate PNECs, according to WFD guidance (EU 2011) (Table 2 and ESI Table A1). It is recognized that as new ecotoxicity data becomes available, substance PNECs are subject to update, and the estimates of PNECs applied in the present study may not, in every case, reflect the most up to date applied or proposed PNEC for regulatory purposes (e.g. under the WFD or European Medicines Agency (EMA) Environmental Risk Assessments. However, the estimated PNECs reported here were applied in the CIP for the purposes of selection for monitoring, preliminary risk assessment and prioritization, and so remain relevant in this context, and it is beyond the objectives of the present study to derive new PNECs for each of the APIs monitored.

2.2 Sampling programme

WwTWs were selected for the CIP program on the basis of broadly representing the distribution of UK WwTWs (A1, ESI), predominantly activated sludge plants (ASP) and trickling biofilters (TF) but also Membrane bioreactors (MBR) and oxidation ditches (OD) (Table A2 of ESI).

Data used for this research were (Table A2 of ESI):

- **CIP1 program**: 25 WwTW data for primary, secondary and tertiary process for 11 APIs. Sampling for this element of the programme was conducted over a two-year period between 2011 and 2013. In this part of the programme two samples (spaced more than 4h apart to provide a degree of replication) were taken on between 10 and 15 occasions.
- **CIP2 program**: 19 APIs and 4 metabolites were sampled on 20 occasions at 45 WwTWs in the influent and effluent (not intermediate process stages, unlike CIP1) over a two-year period between 2015 and 2017.

Samples were collected on a stratified/random spot sampling basis (i.e. grab samples taken at discrete times rather than multiple integrated sampling), with sampling occasion spaced at approximately monthly intervals. A minimum of 15% of samples was taken at non-working hours (evenings and weekends) to ensure a wide a range possible of sampling intervals.

2.3 Sampling and analysis

The samples were collected in stainless steel samplers, stored in glass container and transported at 4° C to the analysis laboratories. The samples were stored a maximum of 5 days prior to analysis. This period was shown to be appropriate as not leading to more than a 20% change in determinand concentration; as confirmed before the start of the CIP sampling programme by undertaking tests of sample stability. Samples for the determination of steroid oestrogens were preserved by adding 30% hydrochloric acid and copper nitrate (Gardner, 2012). All analysis was by laboratories with ISO17025 accreditation. Prior to the programme candidate laboratories were required to undertake tests of analytical performance to demonstrate that they met the stated programme requirements for limit of detection, precision and recovery in relevant sample matrices at relevant concentrations – that is, proof of performance was required, rather that methods being stipulated. Methods used for the determination of pharmaceuticals were all based on variants of High Performance Liquid Chromatograph–Mass Spectrometry (HPLC-MS) or Gas Chromatography-Mass Spectrometry (GC-MS). Quality assurance/quality control (QA/QC) procedures, including the use of field blanks, were observed and reported for sample collection. Within laboratory QC sample pre-treatment and analysis for both laboratory tests and field sampling. Laboratories also took part in a bespoke proficiency testing scheme for pharmaceuticals. Details of the proficiency testing scheme used to ensure quality assurance is provided in A2 of the ESI. Where reported concentrations were below LOD (for the majority of substances apart from ibuprofen and tamoxifen this applied to fewer than 10% of the approximately 1000 results reported), the result was substituted at half face value - as stipulated in the

relevant daughter Directive (EC, 2009) of the WFD. There were significant instances of inter-laboratory bias or inter-regional variation, which would otherwise indicate if there was a bias in the procedure of sample handling and analysis methodology.

Table 2. Determinand abbreviations and required limits of detection and total error

		Concentration (µg/l)			
Code	**Determinand**	**PNEC[1]**	**Required LOD effluent**	**Required LOD river**	**P%[2]**
ATNL	Atenolol	148	0.01	0.01	50
ATOV	Atorvastatin	1.7	0.01	0.01	50
ATOVo	Ortho-hydroxy-atorvastatin	1.7	0.01	0.01	50
ATOVp	Para-hydroxy-atorvastatin	1.7	0.01	0.01	50
AZMY	Azithromycin	0.09	0.005	0.005	50
CBAZ	carbamazepine	2.5	0.1	0.1	50
CBAZe	10,11- epoxy-carbamazepine	2.5	0.1	0.1	50
CIPR	Ciprofloxacin	0.089	0.01	0.01	50
CLMY	Clarithromycin	0.13	0.01	0.01	50
DCF	Diclofenac	0.05	0.01	0.01	50
ERMY	Erythromycin	0.2	0.1	0.1	50
ERMYn	Norerythromycin	0.2	0.1	0.1	50
E1	Oestrone	0.003	0.001	0.001	50
E2	17β oestradiol	0.001	0.0003	0.0003	50
EE2	17α ethinyloestradiol	0.0001	0.00003	0.00003	50
FLXT	Fluoxetine	0.047	0.01	0.01	50
IBPF	Ibuprofen	0.01	0.01	0.01	50
METF	Metformin	13.45	0.1	0.1	50
PRPL	Propranolol	0.1	0.01	0.01	50
RNTD	Ranitidine	0.31	0.1	0.1	50
SERT	Sertraline	0.121	0.01	0.01	50
SERTn	Norsertraline	0.121	0.01	0.01	50
TMXF	Tamoxifen	0.49	0.005	0.005	50

[1] Estimated PNEC (ESI Table 1). [2]The target maximum tolerable error is equal to:

$$\left[(targetLOD)^2 + \left(\frac{A \times P\%}{100}\right)^2\right]^{\frac{1}{2}}$$

Where the target maximum LOD and P% are given in the table and A is the determinand concentration in the sample. Performance testing should seek to demonstrate that the tolerable total error limit is achieved by showing that precision (2 x standard deviation) and bias are respectively no larger than half the target maximum total error. Thus, for example, for a total error limit of 100 units, standard deviation should be shown not to be larger than 25 and bias should not exceed 50. LOD was defined as 3.3x the standard deviation of blank-corrected results of determinations made on a sample containing essentially no determinand (where possible in a relevant sample matrix) (Thompson and Ellison, 2013) In many cases, it was not possible to find effluent samples free from determinands in which case a synthetic sample was used.)

2.4 Data handling and analysis

The data handling and the statistical analysis were conducted with either Microsoft Excel (2016) or IBM SPSS Statistics software (version 20).

In the data handling, the replicates were averaged and this value was then used for further statistical calculations. Mean, median, maximum, minimum and percentiles were calculated from the daily average. Fraction remain was calculated from the influent concentration as a fraction of the various stages of the process. The removal was calculated as percentage from the concentration (C):

Removal (%) = $(C_{influent}-C_{effluent})/ C_{influent}$

For the purpose of this research the term 'removal' relates to the loss of specified compounds from the aqueous phase between influent and effluent (and intervening process steps where quoted). It should be noted that the term removal does not necessarily mean degradation of the API; the loss of the parent compound may be a result of a combination of partitioning to particulates and/or degradation to metabolites.

2.5 Risk assessment approach

2.5.1 Face value risk ranking

A "face value" exceedance is one in which the mean effluent concentration is greater than the relevant estimated PNEC; a "high confidence" exceedance is one for which the lower part of the 90% confidence interval about the mean effluent concentration is greater than the estimated PNEC i.e. there is 95% confidence that the mean is larger than the estimated PNEC.

2.5.2 Refined risk assessment based on estimated available dilution

Previous research has used a combination of modelled average river flows (Comber et al., 2013) and average WwTW discharge volumes to estimate dilution of effluent with receiving water. Effluent flow data was derived from measured values for larger WwTW, but estimated for works serving less than 2000 population equivalent based on water company estimates of connected population and per capita wastewater discharge to sewer (200 l/head/day- including an allowance for runoff) (Comber et al., 2007). A matrix (Table 3) of available dilution was then generated.

Table 3. Estimated dilutions available for UK WwTW

	Dilution ratio band									Total no. works
	0-1	**1-2**	**2-5**	**5-10**	**10-15**	**15-20**	**20-50**	**50-100**	**100+**	
Midpoint dilution	1	1.5	3.5	7.5	12.5	17.5	35	75	100	
Combined dist' dil'[1]	0[2]	4.4	10.6	23.6	36.5	35	70	150	200	
Population served										
<250	86	75	54	54	11	21	86	86	2656	3127
251-500	0	6	0	6	25	0	50	81	605	774
501-2000	0	5	20	20	51	46	351	285	544	1322
2001-10000	17	25	151	160	130	84	202	93	130	993
10001-50000	82	67	160	103	24	30	48	15	18	548
50001-200000	54	29	27	12	5	12	20	5	0	164
200001-1m	74	0	11	0	0	0	0	0	0	85
>1m	4	4	0	0	0	0	0	0	0	7
Total	**316**	**211**	**423**	**355**	**246**	**194**	**756**	**564**	**3954**	**7020**
%	**5**	**3**	**6**	**5**	**4**	**3**	**11**	**8**	**56**	

[1] Values used to calculate PECs in river using a Combined Distribution simulation (see A3 and Table A3). [2] A worst case scenario of zero dilution.

The next step was to generate a cumulative percentile distribution of effluent concentration data (in 10%ile intervals between 10 and 100%). This was achieved by averaging the effluent concentrations for each of the 45 WwTW sampled as part of the CIP2 survey. Step three was to divide each percentile concentration by the dilution available (using the value from the combined distribution estimate – See A3 of the ESI) to generate a PEC. The PEC can then be compared with estimated API PNECs to determine the number of WwTW at risk of exceeding the PNEC for any given each dilution band and percentile effluent concentration. An example of the risk assessment is provided in Table A4 of the ESI.

3. RESULTS and DISCUSSION

3.1 Removal efficiency for APIs

The CIP1 study generated removal data for APIs across all stages of treatment, influent, after primary settlement, secondary biological treatment and where applied, post tertiary treatment. To gain a better understand of the fate of the 11 pharmaceuticals through the treatment train, the fraction of API remaining in the effluent after treatment was calculated across all 25 WwTW in the CIP1 program (Table 4).

Each cycle of sampling was treated as an isolated entity (averaging the samples within the same day), thus simplify the ability to compare APIs removal across the diverse range of works. As seen from the

data in Table 4, most APIs are removed in the secondary biological treatment process and very little through further tertiary treatment. This corresponds well with previously published data (Stockholm Vatten, 2010). The absolute effluent concentrations (Table 4) also correspond well with those reported elsewhere for predominantly UK effluents (Table 1).

Table 4. CIP1 data for API fraction remaining throughout the process stages in the WwTW, as well as the absolute effluent concentration

Fraction of API remaining in effluent after treatment												
API	**Primary Process**			**Secondary Process**			**Tertiary Process**			**Effluent Concentration (µg/l)**		
	Median	**5-%ile**	**95-%ile**	**Median**	**5-%ile**	**95-%ile**	**Median**	**5-%ile**	**95-%ile**	**Median**	**5-%ile**	**95-%ile**
Diclofenac (DCF)	0.76	0.40	1.6	0.52	0.18	1.2	0.44	0.16	1.0	0.20	0.084	0.51
Erythromycin (ERMY)	0.79	0.26	1.7	0.52	0.11	1.2	0.44	0.08	1.1	0.43	0.052	2.0
Ethinylestradiol (EE2)	0.96	0.36	2.4	0.54	0.13	1.9	0.49	0.10	3.5	0.0003	0.0001	0.0020
Oestrone (E1)	1.0	0.59	2.1	0.28	0.02	2.4	0.10	0.01	1.2	0.0048	0.0007	0.058
Oestradiol (E2)	0.97	0.44	1.6	0.11	0.01	0.8	0.05	0.01	0.80	0.0009	0.0001	0.012
Fluoxetine (FLXT)	0.79	0.38	1.5	0.48	0.08	1.2	0.46	0.09	1.1	0.032	0.0050	0.066
Ibuprofen (IBPF)	0.83	0.39	1.3	0.04	0.00	0.2	0.01	0.00	0.21	0.19	0.0050	2.9
Ofloxacin (OFLX)	0.88	0.12	2.2	0.45	0.08	1.4	0.34	0.05	1.0	0.016	0.0050	0.14
Oxytetracycline (OXTCY)	0.66	0.13	1.6	0.16	0.00	0.6	0.13	0.01	0.54	0.21	0.019	1.1
Propranolol (PRPL)	0.91	0.52	1.4	0.68	0.14	1.2	0.65	0.16	1.2	0.14	0.042	0.32
Salicylic acid (SLCYA)	0.85	0.28	1.6	0.01	0.00	1.1	0.01	0.00	0.33	0.18	0.017	3.8

ERMY, DCF, FLXT and OXTCY were all shown to have similar removal efficiencies throughout the primary and secondary treatment processes based on the CIP1 dataset. The primary process relies mostly on removal of APIs through adsorption onto sludge (Stockholm Vatten, 2010) as retention times are relatively low and so this fits well to the data found for OXTCY, as it is previously known to adsorb strongly onto solids (Verlicchi, 2012) and found at higher concentration (4 mg/kg) in sludge compared with other APIs such as DCF, ERMY and FLXT (0.07, 0.05 and 0.12 mg/kg, respectively) (Jones et al., 2014). PRPL had overall poor removal of 35% and 26% (0.65 and 0.74 fraction remaining) between influent and effluent for CIP1 and CIP2 respectively (Table 4 and 5), which also corresponded well with previously published data of 28% removal efficacy (0.72 fraction remaining) in WwTW (Kasprzyk-Hordern et al., 2008).

In the CIP2 data set (Table 5) there was high total removal (based on comparison of influent and effluent API concentrations) of IBPF, METF, E2, ATNL, ATOVp, E1, ATOV, CIPR, ATOVo, which all had fraction remaining ratios of 0.2 or lower (i.e. better than 80% removal efficiency) (Figure 1,

and Table A5). This suggested either rapid biodegradation of the parent compound and/or adsorption to sludge. None of the substances are considered sufficiently volatile to suggest any significant loss to the atmosphere. The intermediate set of APIs consisting of SERTn, RNTD, CLMY, EE2, FLXT, DCF and ERMY, which all had fraction remaining below 0.6 (i.e. greater than 40% removal efficiency). PRPL, CBAZe, ERMYn, AZMY and CBAZ all showed poor removal through the WwTW process (Figure 1 and Table A5).

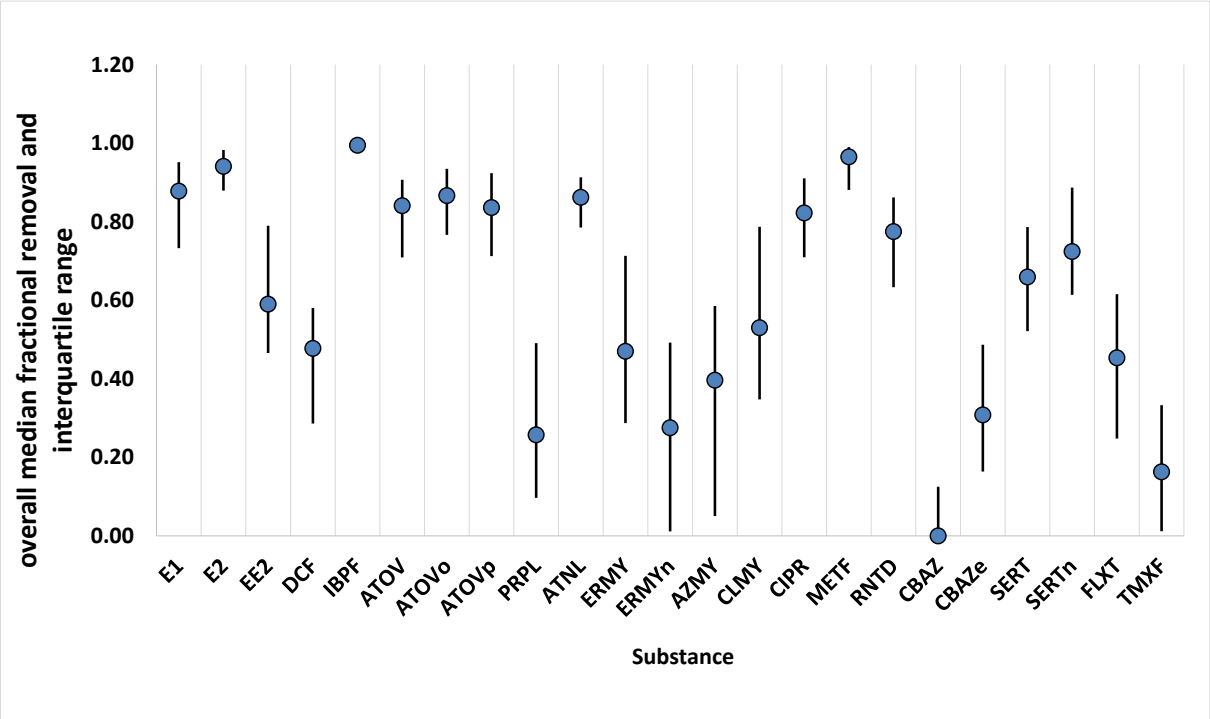

Figure 1. Fractional removal for APIs in CIP2

Table 5. Summary concentration values for CIP2 APIs (45 WwTW sampled on 20 occasions)

	Influents (µg/l)				Effluents (µg/l)		
Substance	Median of WwTW average values	25%ile	75%ile		Median of WwTW average values	25%ile	75%ile
E1	0.038	0.030	0.049		0.004	0.002	0.014
E2	0.014	0.011	0.019		0.001	0.0003	0.002
EE2	0.00051	0.00041	0.00097		0.00020	0.00014	0.00040
DCF	0.54	0.40	0.76		0.29	0.20	0.41
IBPF	18.13	12.08	21.32		0.11	0.02	0.56
ATOV	0.61	0.41	1.01		0.10	0.06	0.16
ATOVo	1.33	0.81	1.76		0.17	0.08	0.29
ATOVp	1.33	0.82	2.03		0.21	0.12	0.35
PRPL	0.260	0.171	0.354		0.174	0.119	0.245
ATNL	2.600	1.872	3.297		0.323	0.210	0.463
ERMY	0.733	0.551	1.161		0.350	0.190	0.558
ERMYn	0.060	0.050	0.091		0.050	0.027	0.050
AZMY	0.351	0.171	0.748		0.202	0.095	0.425
CLMY	0.953	0.684	1.564		0.400	0.265	0.711
CIPR	0.861	0.385	1.510		0.147	0.067	0.276
METF	129	104	208		4.8	1.7	15
RNTD	2.35	1.68	3.06		0.529	0.286	0.730
CBAZ	0.60	0.43	0.84		0.641	0.477	0.756
CBAZe	0.18	0.11	0.42		0.117	0.072	0.292
SERT	0.18	0.12	0.27		0.063	0.037	0.081
SERTn	0.12	0.10	0.21		0.033	0.016	0.045
FLXT	0.10	0.07	0.15		0.051	0.036	0.079
TMXF	0.0034	0.0026	0.0047		0.0025	0.0025	0.0028
TXP	0.0050	0.0028	0.0052		0.0050	0.0026	0.0050
BZT	2.16	1.60	3.97		1.38	1.08	2.62
TZT	1.59	1.19	2.60		1.27	0.88	1.96

Figure 2 below represents mean concentrations in the influent and effluent for selected APIs with the others shown in Figure A2 and demonstrates the degree of variability for APIs between WwTW.

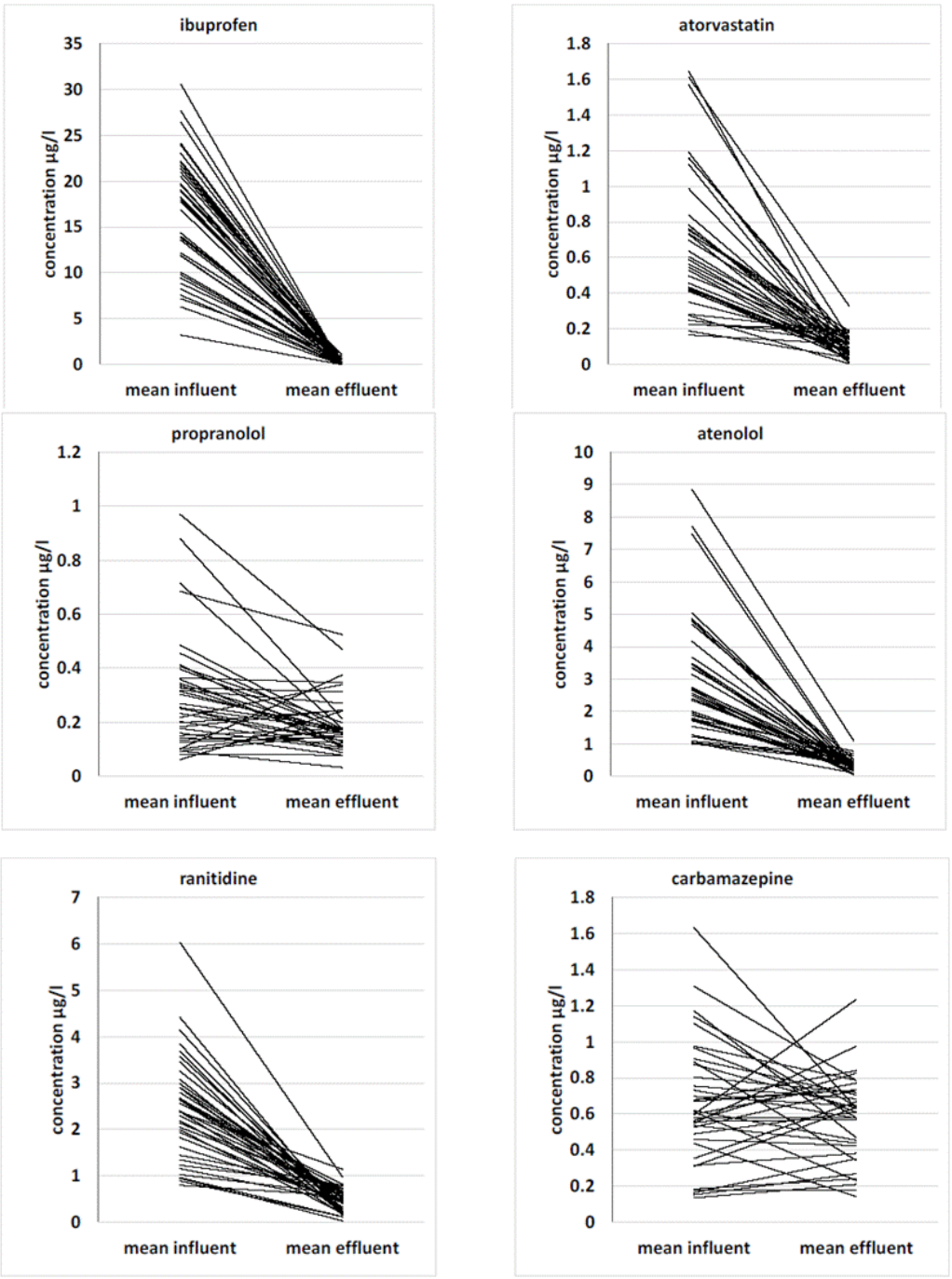

Figure 2. Graphic representation of mean concentrations of APIs in influent and effluent of individual CIP2 WwTWs

In some cases there appears to be an increase in API concentrations in the effluent compared with the influent (Figure 2 and A2 of ESI). There are three main reasons for this:

1) The hydraulic retention time (HRT) within a WwTW means that samples of influent and effluent collected at the same time (a practical requirement of the work) may not reflect actual removal efficiency owing to within works management practices, e.g. batch flow, sludge

return pumping, taking place at the time of sampling. Given HRTs vary vastly between works and types of works it was not practical to calculate nor practically sample WwTW based on their HRTs.

2) The APIs were detected at ng/l levels in a highly complex matrix (particularly the influent) therefore analytical errors may lead to apparent increase in concentrations during treatment (Jelic et al., 2011).

3) In some cases this is a real effect, for example E1 is a degradation product of E2 (Heffley et al., 2014) and so if the rate of loss of E1 during treatment is less than that of E2, then an apparent increase in E1 will occur.

3.2 What is the environmental risk of the APIs being discharged in WwTW effluent?

The median and interquartile concentration values of pharmaceuticals in influents and effluents are summarised in Table 5. Figure 3 shows a summary risk ranking of the CIP pharmaceutical group of substances in relation to the estimated predicted no-effect concentrations applied in CIP (CIP PNECs). A "face value" exceedance is one in which the mean effluent concentration is greater than the relevant estimated PNEC; a "high confidence" exceedance is one for which the lower part of the 90% confidence interval about the mean effluent concentration is greater than the estimated PNEC i.e. there is 95% confidence that the mean is larger than the estimated PNEC. Substances not shown do not figure as noteworthy exceedances.

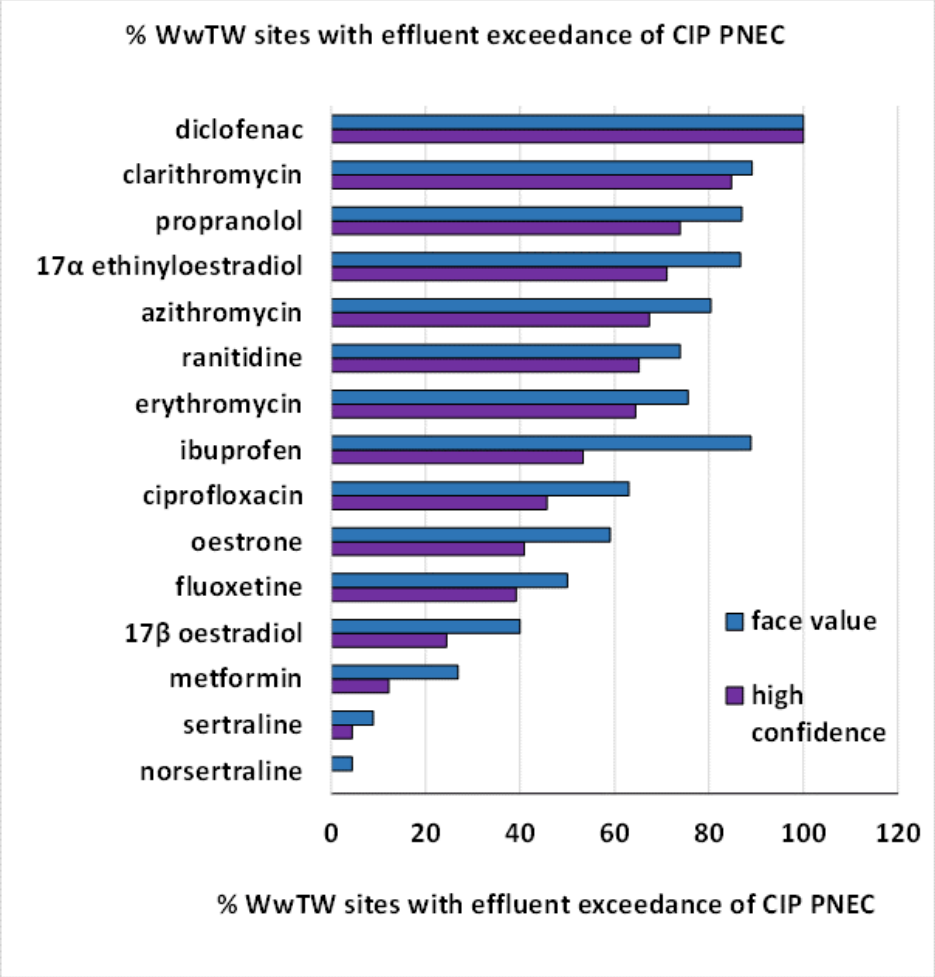

Figure 3. Risk ranking of CIP2 APIs

Figure 3 above illustrates the severity of potential non-compliances for pharmaceuticals as the ratio of the observed concentrations in effluents to the relevant estimated PNEC. This ratio represents the dilution that would be required to achieve compliance, assuming zero upstream concentrations. An important proportion of UK wastewater treatment discharges are not subject to very much greater than a twofold dilution so the potential for downstream non-compliance with PNEC values does exist on the basis of a single effluent discharge alone. Table 3 shows that over 500 WwTW has estimated dilutions of less than 2, 8% of all the WwTW in the UK. Added to this concern must be a consideration of the pharmaceutical concentrations already present in a receiving watercourse upstream of the discharge. Whilst the CIP2 programme did not include the determination of pharmaceuticals in upstream river samples such analysis was undertaken for a range of Priority Substances, including trace organic compounds that like pharmaceuticals, are primarily discharged as a result of domestic inputs to wastewater. The evidence obtained from these investigations is that the burden of upstream contamination is far from irrelevant and that discharges in the higher parts of a river catchment, for example from septic tanks and small WwTW, can raise concentrations to values

that subsequent discharges lower in the catchment only serve to maintain (Phillips et al., 2015). This is an aspect that deserves careful future examination in the context of pharmaceuticals.

Figure 4 shows that several pharmaceuticals have been shown to be present in effluents at concentrations close to, or in many cases in excess of, values that might form the basis of future regulatory limit values.

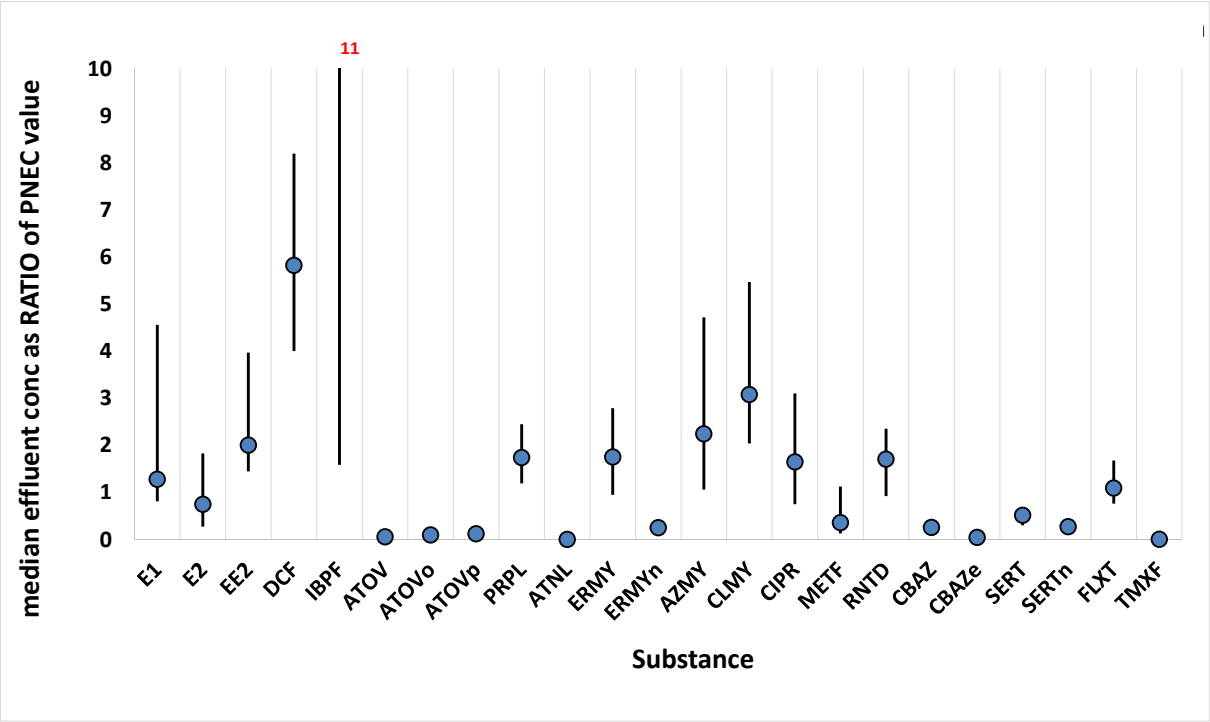

Figure 4. Required within river dilution of WwTW effluent for API concentrations to be less than their estimated PNEC.
Note median effluent concentration for ibruprofen (IBPF) as a ratio of estimated PNEC is 11.

Applying a more realistic risk assessment using estimates of available dilution for UK WwTW effluents discharged to receiving waters, combined with the measured API concentrations from the CIP2 dataset generates a similar priority ranking list in terms of the number of WwTW potentially exceeding downstream estimated PNECs after the effluent has mixed with receiving water (Figure 5). For IBPF this equates to 890 WwTW or 13% of all WwTW in the UK. This estimate is also based on the assumption that there are no significant inputs of API upstream of the WwTW in question.

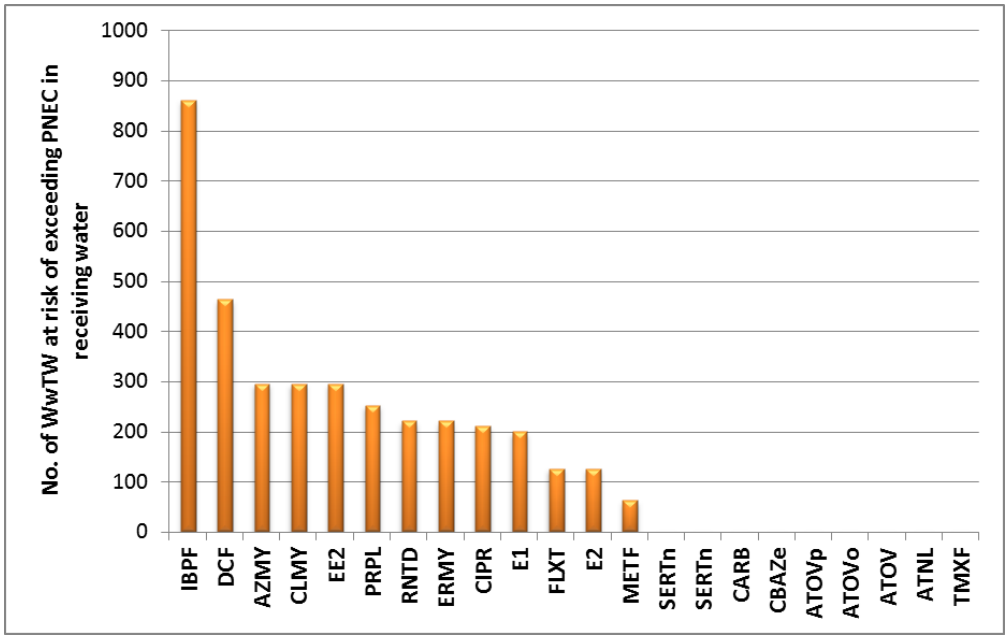

Figure 5. Number of WwTW at risk of exceeding estimated PNEC downstream of receiving water

DCF, AZMY, CLMY, EE2, PRPL, CIPR, RNTD and E1 are all predicted to exceed downstream PNECs in over 200 WwTW. Required mean removal efficiency for any tertiary treatment would range from 35% to 61% depending on the API (Table A6 of ESI). Whether the same tertiary treatment technology could be applied to all of the APIs largely depends on their physico-chemical characteristics. The use of granulated activated carbon (GAC) would require an API to have a reasonable affinity for carbon (i.e. a relatively high octanol:water coefficient - logKow) which may not always be the case for APIs with a high degree of polarity, particularly those that are charged at typical effluent pH (pH 7.5) which would include DCF, IBPF, ATOV and to a degree CIPR (pKa = 6.09). Furthermore, as can be seen from Figures 1 and 2, there are considerable variations in the removal rates between WwTW and so it may be expected to observe a similarly wide variation in removal rates and/or final effluent API concentration, if additional tertiary treatment were to be applied. This would obviously lead to a degree of uncertainty regarding possible compliance with any given in river PNEC or water quality standard.

Figures 1 and 2 show that WwTW in general, have a high (but variable) removal rate for most substances with only E2, EE2 propranolol, the macrolide antibiotics, carbamazepine fluoxetine and tamoxifen exhibiting poor removal. It is clear (and unsurprising) that more complex factors, such as the contaminant load on the WwTW, residence time in the works, overall strength of the influent, questions of operation and maintenance as well as the presence of absence of tertiary treatment "add-ons", combine at each location to result in the observed treatment performance (Zorita et al., 2009; Le-Minh et al., 2010; Deegan et al., 2011). In the wider context, the persistence of pharmaceuticals in surface waters will be determined by the degree of upstream contamination from other (in this case,

presumably WwTW) inputs higher in the river catchment. As has been seen in elements of the CIP2 programme dealing with Priority Substances, upstream contamination and lack of headroom for downstream discharges can often be more important than the local impact of a given WwTW. The likely importance of upstream inputs for pharmaceuticals is unclear. Whilst upstream inputs are inevitable in all sites except those at the top of catchments (where there may still be influences from septic tanks) the effect of such inputs is not known, but if smaller WwTW are less efficient than the predominantly larger works selected for the CIP programmes, then the risk to surface waters of exceeding estimated PNECs for APIs may be significant. Persistence and the degree and rate of breakdown in the environment are critical in this context. To fulfil their purpose pharmaceuticals need to be absorbed by the patient, to remain for sufficient time to have the desired effect and then be excreted. This means that in terms of their structure and hence fate and behaviour, pharmaceuticals tend to occupy a middle ground between substances on the one hand that are non-polar, hydrophobic, insoluble, and persistent and those that are highly polar, soluble, mobile and relatively readily biodegradable. This suggests that some degree of degradation in-river might mean that input of pharmaceuticals upstream may not be as great a risk as it is for other persistent, highly mobile priority substances such as some metals, persistent pesticides and industrial compounds.

It should, however, be noted that this assessment is based on the mixing of single APIs in effluent and receiving water under average flow conditions for a fraction of the APIs currently available and used. During summer months river flows are significantly lower than average values, yet effluent flows will remain relatively stable (accepting rain events contributing to flow in combined sewerage systems) leading to generally lower dilution available and therefore higher concentrations of effluent derived contaminants in receiving waters. Seasonal pattern of use for some APIs, antihistamines in summer, flu vaccines in winter etc, would also lead to a variable distribution of APIs in WwTW effluent and hence variable risk to receiving waters. The potential risk of mixtures is complex and requires detailed knowledge of ecotoxicology for the APIs of interest. Such assessments along with determining temporal variations in risk, require more a significantly detailed dataset (not necessarily currently available) and as such is beyond the scope of this broader risk assessment.

Whilst the objective of this research has not been to estimate costs for compliance, drawing on previous estimates of costs for API treatment based on fitting sand filters and granulated carbon sorption technology, the whole life cost (based on 2007 data) for achieving downstream compliance with the estimated IBPF PNEC would approximately £9bn (Comber et al., 2007). So for illustrative purposes it is evident that achieving compliance for all API estimated PNECs would be a substantial investment by the water industry. These estimates are only based on mixing downstream of receiving water and effluent and do not take account of any biodegradation or sorption to particulates leading to

reduced exposure which would need to be considered as part of a more detailed risk assessment prior to considering any remedial action regarding removal of APIs from WwTW effluent.

Much in relation to future compliance (and therefore cost to the water companies) will depend on the derivation method and data used to set water quality standards. The outputs of the CIPs in this case constitute a valuable risk assessment of the likely impact of whatever regulations might be introduced in the future. Of the pharmaceuticals / likely future Priority Substances, the so-called WFD watch list substance diclofenac, the steroids as well as possibly ibuprofen appear to be at risk of causing widespread exceedances of estimated PNECs in UK rivers. With respect to these substances, options of regulated use and control of patient behaviour relating to disposal of unused medicines might be enough to make a substantial difference. However, wastewater treatment solutions might turn out to be essential for the steroids, at least in the case of EE2.

4. CONCLUSIONS

As has been observed for the CIP1 program there are a high variability in the removal of APIs observed between and within the individual plants. This variation may be due to many factors such as process technology as well as regional variation. Rates of removal in wastewater treatment have also been determined. The majority of substances studied are removed to a high degree, but with a wide variation in performance. Those that are less substantially reduced in concentration are ethinyloestradiol, diclofenac, propranolol, the macrolide antibiotics, fluoxetine, tamoxifen and carbamazepine. All except the last two of these substances are present in effluents at concentration higher than their estimated respective PNECs.

If the PNECs applied in the present study were all implemented as regulatory quality standards under the WFD, the risk assessment undertaken suggests that over a 10 times dilution would be required, to ensure that some APIs (ibuprofen in this case) meet their downstream quality standards, assuming no upstream contribution to background concentrations. This could entail treatment at up to 890 WwTW to meet current PNECs.

Much in relation to the need for future action by dischargers depends on whether or not these substances are regulated and the water quality standard chosen, but if the CIP estimated PNECs are a guide to regulatory limits, then there is potential for localised non-compliance in surface waters; at least in the case of ethinyloestradiol, diclofenac, ibuprofen, propranolol and the macrolide antibiotics. Further monitoring of pharmaceuticals in surface waters to determine the temporal variations in river concentrations associated with changing river flows (and hence dilution), the persistence, and the bioavailability of APIs needs to be considered.

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REFERENCES

Ashton, D., Hilton M. And Thomas K. V. Investigating the environmental transport of human pharmaceuticals to streams in the United Kingdom. Sci Total Environ. 2004;333:167-184.

Astra Zeneca (2016) Environmental Risk Data Relating to our medicine, accessed, 21/3/17. https://www.astrazeneca.com/content/dam/az/our-company/Sustainability/Environmental_risk_data_relating_to_our_medicines.pdf

Bound, J.P., and Voulvoulis, N. Predicted and measured concentrations for selected pharmaceuticals in UK rivers: implications for risk assessment. Water Res 2006; 40: 2885-2892.

Boxall, A. B. A., Keller V.D.J. Straub J.O., Monteiro S.C., Fussell R. and Williams R.J. Exploiting monitoring data in environmental exposure modelling and risk assessment of pharmaceuticals. Environ. Internat. 2014;73:176-185.

Bradley, P.M., Journey, C.A., Button, D.T., Carlisle, D.M., Clark, J.M., Mahler, B.J., Nakagaki, N., Qi, S.L., Waite, I.R., VanMetre, P.C. Metformin and other pharmaceuticals widespread in wadeable streams of the southeastern United States. Environ. Sci. Technol. Lett. 2016;3:243-249.

Burns, E.E., Thomas-Oates, J., Kolpin, D.W., Furlong, E.T., Boxall, A.B.A. Are exposure predictions, used for prioritization of pharmaceuticals in the environment, fit for purpose? Environ. Toxicol. Chem. 2017;in press.

Clara, M., Kreuzinger N., Strenn B., Gans O. And Kroiss H. The solids retention time—a suitable design parameter to evaluate the capacity of wastewater treatment plants to remove micropollutants. Water Res. 2005;39:97-106.

Comber, S., Gardner, M., Georges, K., Thornton A. (2007) Dangerous Substances and Priority Hazardous Substances/Priority Substances under the Water Framework Directive (07/WW/17/7). UK Water Industry Research (UKWIR), 1 Queen Anne's Gate, London, UK. ISBN: 1 84057 464 X.

Comber S., Smith R., Daldorph P., Gardner M., Constantino C. and Ellor B. Development of a chemical source apportionment decision support framework for catchment management. Environ Sci Technol. 2013;47:9824-9832.

Comber S., Gardner M., Jones, V. and Ellor B. Source Apportionment of Trace Contaminants in Urban Sewer Catchments. Environ Technol. 2014;36(5):573-587.

Deegan, A.M., Shaik B., Nolan K., Urell K., Oelgemöller M., Tobin J., Morrissey A. Treatment options for wastewater effluents from pharmaceutical companies. Int J Environ Sci Technol. 2011; 8:649–666.

EC, 2009: Technical Specifications for Chemical Analysis and Monitoring of Water Status. Directive 2009/90/EC.

EU (2011) European Union Technical Report - 2011 – 055. Common Implementation Strategy for the Water Framework Directive (2000/60/EC). Guidance Document No. 27 Technical Guidance For Deriving Environmental Quality Standards. ISBN : 978-92-79-16228-2. DOI : 10.2779/43816.

EU (2013) European Union Directive 2013/39/EU of the European Parliament and of the Council of 12 August 2013 amending Directives 2000/60/EC and 2008/105/EC as regards priority substances in the field of water policy.

557 Falås, P., Andersen H.R., Ledin A., and La Cour Jansen J. Occurrence and reduction of
558 pharmaceuticals in the water phase at Swedish wastewater treatment plants. Water Sci Technol.
559 2012;66(4):783-791.
560
561 Furlong, E.T., Batt, A.L., Glassmeyer, S.T., Noriega, M.C., Kolpin, D.W., Mash, H., Schenck, K.M.
562 Nationwide reconnaissance of contaminants of emerging concern in source and treated drinking
563 waters of the Unites States: Pharmaceuticals. Sci. Total Environ. 2017;579:1629-1642.
564
565 Gardner M., Comber S., Scrimshaw M., Cartmell E., Lester J. and Ellor B. The significance of
566 hazardous chemicals in wastewater treatment works effluents. Sci Total Environ. 2012;437:363-372.
567
568 Gardner M., Jones, V., Comber S., Scrimshaw M., Coello-Garcia, T., Cartmell E., Lester J. and Ellor
569 B. Performance of UK wastewater treatment works with respect to trace contaminants. Sci Total
570 Environ. 2013;456-457:359-369.
571
572 Heffley J., Comber S., Wheeler B. and Redshaw C. Developing a modelling approach to predict
573 pharmaceutical discharges from UK sewage treatment works using steroid estrogens as a case study.
574 Environ Sci: Processes and Impacts 2014;16:2571-2580.
575
576 Henze, M., *Biological wastewater treatment: principles, modelling and design*. IWA publishing,
577 2008.
578
579 Hope, B.K., Pillsbury L., and Boling B A state-wide survey in Oregon (USA) of trace metals and
580 organic chemicals in municipal effluent. Sci Total Environ 2012;417:263-272.
581
582 IMS (2016) Quantiles IMS (Intercontinental Marketing Services) Data for active ingredient sales.
583 http://www.imshealth.com/; accessed 24th May 2016.
584
585 Jelic, A, Gros M., Ginebreda A., Cespedes-Sanchez R., Ventura F., Petrovic M. and Barcelo D.
586 Occurrence, partition and removal of pharmaceuticals in sewage water and sludge during wastewater
587 treatment. Water Res. 2011;45(3):1165-1176.
588
589 Johnson A., Dumont E., Williams R., Oldenkamp R., Cisowska I. and Sumpter J. Do concentrations
590 of ethinylestradiol, estradiol, and diclofenac in European rivers exceed proposed EU Environmental
591 Quality Standards? Environ. Sci. Tech. 2013a;47(21):12297-12304.
592
593 Johnson A., Oldenkamp R., Dumont E. and Sumpter J. Predicting concentrations of the cytostatic
594 drugs cyclophosphamide, carboplatin, 5-fluorouracil, and capecitabine throughout the sewage
595 effluents and surface waters of Europe. Environ. Tox. And Chem. 2013b;32(9):1954-1961.
596
597 Johnson A., Keller V., Dumont E. and Sumpter J. Assessing the concentrations and risks of toxicity
598 from the antibiotics ciprofloxacin, sulfamethoxazole, trimethoprim and erythromycin in European
599 rivers. Sci of the Tot. Environ., 2015, 747-755.
600
601 Jones, V., Gardner M. and Ellor B. Concentrations of trace substances in sewage sludge from 28
602 wastewater treatment works in the UK. Chemosphere 2014;111:478-484.
603
604 Jobling, S., Williams R., Johnson A., Taylor A., Gross-Sorokin M., Nolan M., Tyler C., van Aerle R.,
605 Santos E. and Brighty G. Predicted exposures to steroid estrogens in UK rivers correlate with
606 widespread sexual disruption in wild fish populations. Environ Health Persp. 2005;114:32-39.
607
608 JRC (2015) Ibuprofen Dossier, Draft Version 3.
609
610 JRC (2015) Summary Dossier Review, Draft.
611

JRC (2016) Development of the first Watch List under the Environmental Quality Standards Directive. Joint Research Centre, Report EUR 27142 EN, by Raquel N. Carvalho, Lidia Ceriani, Alessio Ippolito and Teresa Lettieri, 2015, Accessed 21/3/17

http://publications.jrc.ec.europa.eu/repository/bitstream/JRC95018/lbna27142enn.pdf

Kasprzyk-Hordern, B., Dinsdale R.M. and Guwy A.J. The occurrence of pharmaceuticals, personal care products, endocrine disruptors and illicit drugs in surface water in South Wales, UK. Water Res 2008;42(13): 3498-3518.

Kolpin, D.W., Furlong, E.T., Meyer, M.T., Thurman, E.M., Zaugg, S.D., Barber, L.B., and Buxton, H.T. Pharmaceuticals, hormones, and other organic wastewater contaminants in U.S. streams, 1999-2000: A national reconnaissance. Environ. Sci. Technol. 2002;36:1202-1211.

Le-Minh N., Khan S.J., Drewes J.E., Stuetz R.M. Fate of antibiotics during municipal water recycling treatment processes. Water Res. 2010; 44(15):4295–4323.

Lees K., Fitzsimons M., Tappin A., Snape J. and Comber S. Pharmaceuticals in soils of lower income countries: physico-chemical fate and risks from wastewater irrigation. Environ. Int. 2016; .94:712-723.

Levado R., Thibaut R., Raldua D, Martin R., and Porte C. First evidence of endocrine disruption in feral carp from the Ebro River. Tox. And Appl. Pharma. 2004;196(2):247-257.

Li, C., Cabassud C. and Guigui C. Evaluation of membrane bioreactor on removal of pharmaceutical micropollutants: a review. Desal and Water Treat 2015; 55(4):845-858.

Melvin, S.D. and Leusch F.D.L. Removal of trace organic contaminants from domestic wastewater: A meta-analysis comparison of sewage treatment technologies. Environ International 2016; 92: 183-188.

Murray-Smith, R.J. Coombe, V.T., Grönlund, M.H., Waern F., Baird J.A. Managing emissions of active pharmaceutical ingredients from manufacturing facilities: An environmental quality standard approach. Int. Env. Ass. and Man. 2012;8(2):320–330.

NSF (2016) National Science Foundation, Water and Environmental Technology Centre, Pharmaceutical PNEC list. Accessed 21/3/17. http://www.nsfwetcenter.org/wp-content/uploads/2016/10/WET-Center-Phamaceutical-PNEC-list-3.pdf

Phillips, P.J., Smith, S.G., Kolpin, D.W., Zaugg, S.D., Buxton, H.T., Furlong, E.T., Esposito, K., Stinson, B. Pharmaceutical formulation facilities as sources of opioids and other pharmaceuticals to wastewater-treatment-plant effluents. Environ. Sci. Technol. 2010;44:4910-4916.

Phillips, P.J., Schubert, C., Argue, D., Fisher, E., Furlong, E.T., Foreman, W., Gray, J., Chalmers, A. Concentrations of hormones, pharmaceuticals and other micropollutants in groundwater affected by septic systems in New England and New York. Sci. Total Environ. 2015;512-513:43-54.

Rowett C., Comber S. and Hutchinson T. The impact of natural and anthropogenic Dissolved Organic Carbon (DOC), and pH on the toxicity of triclosan to the crustacean Gammarus pulex (L.). Sci Total Environ. 2016;565:222-231.

Singer, Andrew C., Järhult J.D., Grabic R., Khan G.A., Lindberg R.H., Fedorova G., Fick J., Bowes, M.J., Olsen B. and Söderström H. Intra-and inter-pandemic variations of antiviral, antibiotics and decongestants in wastewater treatment plants and receiving rivers. *PLoS One* 9.9 2014;9(9):108621.

Stockholm Vatten, 2010. Läkemedelsrester i Stockholms vattenmiljö Förekomst, förebyggande åtgärder och rening av avloppsvatten www.stockholmvatten.se/globalassets/pdf1/rapporter/avlopp/avloppsrening/lakemedelsrapport_slutrapport.pdf (accessed sept, 2016)

Thompson, M. and Ellison, S.L.R. Towards an uncertainty paradigm of detection capability, Analytical Methods 2013;5:5857-61.

Tyler C., Jobling S. and Sumpter J. Endocrine disruption in wildlife: a critical review of the evidence. Crit. Rev. in Toxicol. 2008;28:319-361.

UKWIR (2014) Risk based Prioritisation of Pharmaceuticals UKWIR Report 14/WW/17/16 (August 2014) ISBN: 1840577355.

Verlicchi P, Al Aukidy M, Zambello E. Occurrence of pharmaceutical compounds in urban wastewater: removal, mass load and environmental risk after a secondary treatment-a review. Sci Total Environ. 2012;429:123-55.

Wahlberg, C., Björlenius B. and Paxéus N. Fluxes of 13 selected pharmaceuticals in the water cycle of Stockholm, Sweden. Water Sci and Technol. 2011;63(8):1772-1780.

WHO, 2011: Pharmaceuticals in Drinking-water, Public Health and Environment Water, Sanitation, Hygiene and Health WHO/HSE/WSH/11.05 www.who.int/water_sanitation_health/publications/2011/pharmaceuticals_20110601.pdf (accessed Sept, 2016)

Zhou, J. L., Zhang Z.L., Banks E., Grover D. and Jiang J.Q. Pharmaceutical residues in wastewater treatment works effluents and their impact on receiving river water." J Hazard Mat. 2009;166(2):655-661.

Zorita S., Mårtensson L. and Mathiasson L., Occurrence and removal of pharmaceuticals in a municipal sewage treatment system in the south of Sweden. Sci Total Environ. 2009;407(8): 2760–2770.