Article (refereed) - postprint

Johnson, Andrew C.; Oldenkamp, Rik; **Dumont, Egon**; Sumpter, John P..
2013. Predicting concentrations of the cytostatic drugs cyclophosphamide, carboplatin, 5-fluorouracil, and capecitabine throughout the sewage effluents and surface waters of Europe. *Environmental Toxicology and Chemistry*, 32 (9). 1954-1961. 10.1002/etc.2311

Running head:

Cytostatic drug concentrations in European rivers

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Words in text 3814

Words in references 1943

Words in tables 821

Predicting concentrations of the cytostatic drugs cyclophosphamide, carboplatin, 5-fluorouracil and capecitabine throughout the sewage effluents and surface waters of Europe

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Abstract - This study evaluated the potential environmental concentrations of four cytostatic (also known as cytotoxic) drugs in rivers. The antimetabolite 5-fluorouracil (5FU) and its pro-drug capecitabine were examined based on their very high use rates, cyclophosphamide (CP) for its persistence, and carboplatin for its association with the metal element platinum. The study combined drug consumption information across European countries, excretion, national water use, and sewage removal rates, to derive sewage effluent values across the continent. There was found to be considerable variation in the popularity of individual cytostatic drugs across Europe, including a 28-fold difference in 5FU use and 15-fold difference in CP use . Such variation could have a major effect on the detection of these compounds in effluent, or river water. Overall, capecitabine and CP had higher predicted levels in effluent than 5FU, or carboplatin. Predicted effluent values were compared with measurements in the literature and many non-detects could be explained by insufficient limits of detection. Linking the geographic based water resources model GWAVA with this information allowed water concentrations throughout 1.2 million km of European rivers to be predicted. The 90%ile (worst case) prediction indicated that, with the exception of capecitabine, >99% by length of Europe's rivers would have concentrations below 1 ng/L for these cytostatic drugs. For capecitabine 2.2% of river length could exceed 1 ng/L.

Key words: CYTOSTATIC, CYTOTOXIC drugs, SEWAGE effluent, RIVER, PREDICTION

INTRODUCTION

The discharge of pharmaceuticals in wastewater into the aquatic environment has been a source of discussion and concern in scientific and regulatory circles now for well over a decade [1, 2]. Chemotherapy drugs in the group known as cytostatic, cytotoxic or antineoplastic (hereafter described as cytostatic) often feature on lists of pharmaceuticals of concern which we discharge into our river systems [3, 4]. These compounds are now an increasingly popular subject of environmental research and discussion [5-7]. There is relatively little information available on the toxicity of cytostatic drugs to aquatic organisms. The information that is available suggests that cytostatic drugs are not very toxic to a variety of aquatic organisms [8-10]. However, long term exposure studies particularly with fish and including more than one generation have not yet been conducted. Therefore, it is premature to conclude that environmental concentrations of cytostatic drugs pose no risk to aquatic organisms

A greater focus has been on their potential to harm humans through water recycling. This scenario is where rivers are used both to receive sewage effluent and as a source of drinking water. A significant difference between humans and aquatic wildlife is that the former are protected by a range of water purification technologies [11, 12]. Whilst this is reassuring, there are some grounds for suggesting some of the water soluble cytostatic drugs like cyclophosphamide may still survive ozonation and pass through into drinking water [13]. Many would still consider that this is not a concern, as even the most pessimistic predictions indicate exposure to these drugs would still be considerably below levels of concern [14, 15]. Another angle is that the unborn child in the womb would be particularly vulnerable to inadvertent exposure to cytostatic drugs, due to their teratogenic potential [16]. However, a comprehensive study indicated that exposure during the foetal development phase of the second trimester onwards carried little risk of harmful effects to the unborn child [17]. Nevertheless, exposure in the embryogenesis period, which occurs in the first trimester (approximately the first 12 weeks following conception) could lead to a range of malformations [18].

Key to any assessment of the risks involved are the concentrations likely to be found in surface, or drinking water. A number of studies have been carried out to detect and analyse cytostatic drugs in sewage effluent and surface waters in recent years, with mixed levels of success. This is not an unreasonable exercise given the common administration of chemotherapy in the outpatient departments of most city hospitals [7]. However, a high proportion of these studies failed to find the selected cytostatic drugs [19-25] leaving the reader not knowing whether the drug was actually not used in that city/country, was successfully eliminated in sewage treatment, or present but at a concentration below the limit of detection (LOD)?

A number of authors have reviewed the considerable number of cytostatic drugs used in chemotherapy whose discharge might have harmful consequences for the environment [5-7]. Some are used more frequently than others as they are associated with treating the most common cancers, or used for a diverse range of cancers. This study has selected four cytostatic drugs for further study, based on either their high consumption, their persistence in the environment, or novelty. The cytostatic drug 2-[bis92-chloroethyl)amino]tetrahydro-2H-1,3,2-oxazaphosphorine 2-oxide better known as cyclophosphamide (CP), is a commonly used alkylating agent dating back to the late 1950s. It is typically given intravenously (IV) although low doses may be given orally. It needs to be metabolised by cytochrome P450 enzymes in the liver to liberate alkylating metabolites such as nor-nitrogen mustard, which can cross-link DNA [26-28]. It has been used to treat a range of cancers including brain, bone and leukemia as well as some autoimmune diseases [26]. This drug has received considerable interest from environmental scientists over the years due to its apparent persistence [29-31].

The cytostatic drug cis-diammine-1,1-cyclobutane dicarboxylate platinum (II) also known as carboplatin, is a relatively recent cytostatic drug with clinical trials carried out successfully in the mid 1980s [32]. Carboplatin is always administered via IV. The major cytostatic activity of carboplatin is binding with DNA to form intra-strand crosslinks and adducts that cause changes in the conformation of the DNA and affect DNA replication. Its use has been promoted since it has much less side effects on patients than other

75 Pt containing drugs such as cisplatin [32]. As a representative of the Pt containing group of cytostatic drugs,
76 it may be a source of Pt contamination of the aquatic environment [33].

77 The antimetabolite 5-fluorouracil (5FU) and its pro-drug capecitabine are often found near the top of the
78 consumption list for many countries. Following the observation that cancer tissues incorporated uracil much
79 more than non-malignant tissues, 5-fluorouracil (5FU) was synthesised as an antimetabolite chemotherapy
80 drug back in 1957 [34]. It is typically given to the patient via intravenous delivery. Partly due to the need to
81 give the drug via the intravenous route, it appears to be less popular than its pro-drug capecitabine, which
82 can be given orally. Indeed 5FU does not now appear to be used at all in Finland. The 5FU pro-drug pentyl
83 [1-(3,4-dihydroxy-5-methyltetrahydrofuran-2-yl)-5-fluoro-2-oxo-1*H*-pyrimidin-4-yl]carbamate, commonly
84 known as capecitabine is a drug popularly used for treating colorectal and breast cancers [35]. Unlike 5FU, it
85 is well absorbed when taken orally and in the liver is metabolised through a series of intermediates to form
86 5'-deoxy-5-flourouridine, which is then converted to 5FU by the thymidine phosphorylase enzyme. This
87 enzyme tends to be over expressed in tumour tissue [36], thus giving capecitabine greater selectivity than
88 5FU.

89 This study attempted to predict the range of possible cytostatic drug concentrations in sewage effluent
90 and surface waters for the different countries in the European Union. The method used publically available
91 consumption data, literature data on human excretion values and sewage removal rates. The major
92 refinement on previous studies was the use of varying national consumption values and wastewater
93 discharge values. The following objectives were addressed:

94 • How different are national per capita cytostatic drug consumption values?
95 • What range of concentrations in effluents might be expected given different per capita wastewater
96 discharge rates?
97 • How well do national predicted values correspond to published measured concentrations?
98 • To predict surface water concentrations throughout Europe using a geographic based water quality
99 model.

MATERIALS AND METHODS

Assessing per capita consumption rates

The approach to estimating effluent concentrations takes the drug consumption per capita for a specific nation less that prevented from being excreted as the free parent compound less that removed in sewage treatment. The effluent concentration is then calculated by dividing this figure by the per capita wastewater discharge for that nation:

$$W = \frac{(C - E - S)}{D}$$

Where C is consumption of the drug as ng/cap/d; E is the amount of the drug that is not excreted (ng/cap/d); S is the amount of the drug that is prevented from escaping into sewage effluent (ng/cap/d); D is the diluting volume of wastewater as L/cap/d; and W is the effluent concentration as ng/L.

The most critical part of any model to assess concentrations of human derived chemicals in water is obtaining accurate information on usage. Fortunately, there are some national annual consumption data on cytostatic drugs which are publically available. These can be interrogated to assess a per capita consumption value, given the population of the country at that time (Table 1). The data available ranged from as recent as one year ago to fifteen years old. Data on drug consumption for England was a summary of the responses from the major 34 health trusts, with the data originally cited as mg/1000 people over a 6 month period in 2005 [37].

Assessing per capita excretion rates and sewage removal rates

The next stage in a modelling environmental concentrations of a pharmaceutical is to ascertain how much of the parent compound is excreted unchanged by the patient. Unsurprisingly, not all humans are the same in their excretion behaviour, with such factors as age, health and co-medication all influencing the

123 percentage excreted. It is therefore advisable to survey as wide a range of literature as possible on excretion
124 rates before arriving at a mean value. Similarly, natural variations in sewage performance can influence
125 pharmaceutical removal rates in treatment. Unfortunately, the literature on removal in sewage treatment
126 for many of the cytostatic drugs is still meagre and this must be considered a weak point in modelling these
127 compounds.

128 *CYCLOPHOSPHAMIDE* In the seven references covering 74 patients receiving CP there was a
129 relatively small range in % CP excreted reported over a 24 h period (Table 2). The amount of original CP
130 excreted unchanged ranged from 11 to 20% with a weighted mean of 15.9%. Thus, these values can be used
131 as upper, lower and mean excretion rate values. There is considerable agreement that CP is a persistent
132 compound in both sewage and river water [29-31]. Thus, a simple prediction of CP concentration in sewage
133 effluent can be made, assuming the per capita wastewater discharge volume is known, and no significant
134 attenuation occurs after excretion.

135 *CARBOPLATIN* A proportion of the carboplatin taken by the patient (normally intravenously) is
136 excreted unchanged and several studies have attempted to measure the % of the parent molecule in the
137 urine over a 24 h period. Some of the reports can at first be misleading as they only measure platinum, but
138 it is possible to identify several references where the parent compound itself was measured in the urine. In
139 the five references covering 50 patients, the amount of original carboplatin excreted unchanged ranged
140 from 14 to 69% with a weighted mean of 31.7% (Table 2).

141 There are few studies on the fate of carboplatin in the environment but a number of medical
142 researchers have noted that it is not persistent in urine. The only reference which provides data that can be
143 used on this topic was one where 5 activated sludge plants were studied and an average elimination from
144 the effluent of 72% recorded from a range of 59-85% [38].

145 *5-FLUOROURACIL* As might be expected with a polar drug, the major route of excretion is in the
146 urine, with fecal excretion believed to be unimportant [39]. A potential complication with choosing an

147 appropriate excretion rate is that some treatments, such as those combined with eniluracil, give markedly
148 higher urinary excretion of unchanged 5FU [40, 41]. However, the most used combination therapies appear
149 to be with methotrexate, or leucoverin [34], which don't seem to have a dramatic effect on the 5FU amount
150 excreted in the urine. Also excretion of unchanged 5FU is much higher when given orally, which probably
151 explains the preference for IV use [42]. It was possible to identify four references where the parent
152 compound was measured in the urine in treatments without eniluracil. In the four references covering 32
153 patients examined, there was a fairly wide range in % excreted reported (Table 2). The amount of original
154 5FU excreted unchanged ranged from 2 to 39%, with a weighted mean of 4.6%. Thus, these values could be
155 used as a upper, lower and mean excretion rate values. However, before moving on to national predictions
156 for 5FU in sewage effluent, the contribution from patients taking capecitabine also needs to be taken into
157 account. The amount of 5FU excreted from patients receiving capecitabine ranged from 0.5 to 0.8%, with a
158 weighted mean of 0.7% (Table 2).

159 There have been a number of studies on the biodegradability of 5FU which give the general
160 impression that significant removal in sewage treatment is probable (Table 3). Removal would be strongly
161 dependent on biodegradation, as sorption to sewage particles appears to be low [43]. However, the studies
162 were typically carried out at high concentrations with adapted microbial populations over long time periods
163 and probably at room temperature, circumstances that may well not be typical of operational STPs in
164 Europe. Of these studies, those reported by Kiffmeyer et al. (1998) [29] and Mahnik et al. (2007) [43]
165 probably come closest to replicating the activated sludge environment most typical of European wastewater
166 treatment, albeit at μg/L rather than the ng/L concentrations which might have been more realistic. These
167 studies were most probably carried out at room temperature, which would tend to overestimate removal in
168 winter. Given a probable range of 90-99% removal from these studies, a removal rate of 95% was selected.
169 To estimate 5FU concentration in sewage effluent, the predicted value deriving from both 5FU consumption
170 (following excretion and removal in sewage treatment) and from capecitabine consumption (with its unique
171 excretion of 5FU and sewage removal) must also be included.

172 *CAPECITABINE* Fortunately, a number or references exist where unchanged capecitabine was
173 measured in the urine following administration to patients (Table 2). The amount of capecitabine excreted
174 unchanged from patients receiving capecitabine ranged from 2.6 to 3.4%, with a weighted mean of 3.1%.
175 However, it is very difficult to predict the fate of capecitabine in sewage, as no references can be found on
176 the topic other than some high concentration OECD laboratory studies carried out by Roche [15], where it
177 was described as slowly degradable. For this modelling exercise it was assumed 50% would be removed in
178 sewage treatment.

179 The summary of deduced excretion and removal rates for all the drugs is shown in Table 4. Given
180 consumption, excretion and sewage removal information, all that remains is a national per capita
181 wastewater discharge value in order to predict an effluent concentration. A per capita wastewater discharge
182 value was assumed to be the same as the national per capita water use value. Per capita water use values
183 are provided in a number of reports, including Eurostat from the EU, Environment Agency of England and
184 Wales, UN Development Programme and Environment Canada although they don't always agree with one
185 another! Where available, the Eurostat values have been used in this study. Such predicted values are likely
186 to be conservative since many STP also receive industrial waste which would further dilute the wastewater
187 and hence lower the concentrations.

188 *European river water modelling*

189 To examine potential concentrations of these cytostatic drugs throughout European surface waters,
190 the geographic-based water resources model GWAVA was used [44]. The geographic database of this model
191 includes the location and size of the human European population and their association with sewage
192 treatment plants (STPs). The flows through these STPs are incorporated with other flows and abstractions
193 into the hydrological model. The hydrology is driven by monthly climate over the period 1970-2000. The
194 chemical inputs of per capita drug consumption and removal in sewage were provided by this study. The
195 model calculates the water concentrations through a series of 177,470 grid squares of approximately 6 x 9
196 km (5 by 5 Arc minutes) dimensions. GWAVA summates all the inputs and dilutions within a cell to give a

197 value at the downstream 'outflow' location of that cell. This does not necessarily represent the highest
198 concentration that could occur at some point within that cell. However, at the scale of the European
199 continent portrayed by the model, this scale is considered to be the best compromise and will reveal the
200 exposures most likely to be faced by the majority of aquatic wildlife.

201 The main variables in modelling these cytostatics drugs were, therefore, consumption, excretion,
202 sewage removal and dilution (Table 4). GWAVA can also modify the concentrations by including a water
203 column attenuation rate, however, in the absence of such information for these compounds they were
204 assumed to be conservative once in the rivers.

205 ## RESULTS AND DISCUSSION

206 *Predicted European sewage effluent concentrations*

207 Although the incidence of different cancers are unlikely to be very different across European
208 countries, the selection of drugs to treat these diseases does vary in popularity. Indeed the popularity of
209 certain cytostatics can vary across different regions in the same country [37]. For example,
210 cyclophosphamide (CP) is more than twice as popular in Sweden than any other European country for which
211 data were obtained, with a fifteen-fold difference in possible use across Europe (Table 1, Figure 1). There is
212 an eight-fold difference in carboplatin consumption between the European countries examined, whilst for
213 capacitabine this is a four-fold difference (Table 1, Figure 1). The greatest variation is with 5FU with a
214 twentyeight-fold difference in use (Figure 1). The predicted mean effluent concentrations ranged from 2 to
215 40 ng/L for CP, 0.8 to 2.5 ng/L for carboplatin, 0.3-2.5 ng/L for 5FU, and 8.5-87 ng/L for capecitabine, which
216 reflect the original consumption preferences and differences in national per capita wastewater discharge
217 (88-230 L/cap/d) (Figure 2).

218 These model predictions suggest chemists wishing to monitor CP in sewage effluent must achieve
219 LODs below 1.7 ng/L, expect a European mean concentration of 11 ng/L and travel to Sweden to find the
220 highest concentrations in effluent. Those wishing to monitor carboplatin in sewage effluent must achieve

221 LODs below 0.8 ng/L and expect a European mean concentration of 2.4 ng/L. Carboplatin is not recorded on
222 the medical databases for Netherlands, Norway and Finland and so may not be officially prescribed in those
223 countries. Those wishing to monitor 5FU in sewage effluent must achieve LODs below 0.3 ng/L expect a
224 European mean concentration of 1.0 ng/L, with the highest predicted value of 2.5 ng/L being found in the
225 Czech Republic. Those wishing to monitor capecitabine in sewage effluent must achieve LODs below 8.5
226 ng/L expect a European mean concentration of 29.3 ng/L, with the highest predicted value of 87 ng/L being
227 found in the Czech Republic.

228 *Comparing predicted and measured effluent concentrations*

229 A major objective of this study was to compare and corroborate measured cytostatic drug
230 concentrations with those predicted here. Thirteen studies on CP in sewage effluent were examined. The
231 per capita wastewater discharge values used to predict the results were either calculated from the authors
232 own data, Eurostat 2012, UN 2006, or Environment Canada. Where it was not known, the mean European
233 per capita consumption for CP was used. Of the six references which report non detects, three can be
234 explained simply by having LODs above the predictable levels (Table 5). Where the LOD was below the
235 predicted concentration and yet still no detection made [19, 22, 23], the possibility still exists that
236 wastewater flow was greater than expected on that sampling day. Of the 8 detections, five of the
237 predictions using the method described in this paper were within an order of magnitude of those reported.

238 Three studies were found were chemists tried to measure 5FU in sewage effluents in Spain,
239 Switzerland and the USA, but these resulted in no detections [25, 45, 46]. The reported LODs were 15-21
240 ng/L, but using the prediction method described above, the mean effluent values would be expected to be
241 0.6-1.0 ng/L which would appear to explain their failures. No studies on carboplatin, or capecitabine,
242 concentrations in domestic sewage effluent could be found.

243 *Predicting European river concentrations*

The GWAVA model is providing predictions for 1.2 million km of European rivers receiving the waste from 602.8 million people so that a single run of the model with its 177,000 grid squares and 31 years of climate data generates 66 million results per chemical. All the variables discussed will play a role, but it is clear that the most important factor in predicting correctly river concentrations apart from consumption is dilution (Table 4). Different interpretations on human excretion, or sewage removal rates could change the values by up to 20-fold but dilution could change the values by up to 1000-fold! The results from model runs can be displayed in a number of different ways, such as a map showing the 50%ile concentrations across Europe for CP based on a mean excretion rate and mean sewage treatment removal (Figure 3). This is broadly equivalent to the concentration that would be recorded at a median flow for that part of a river and so might represent the typical exposure for surface waters. In this case the CP hot spots reflect not just the geography and hydrology of Europe, but also the popularity of the drug. This helps to explain the relatively low predicted concentrations in Italy compared with southern Sweden (Figure 3). The results can also be displayed as cumulative frequency curves, such as all the 90%ile concentration values for all the cells based on the highest possible human excretion rates and lowest sewage treatment removal (Figure 4). These predictions could be considered as potential worst case river concentrations such as might be associated with low summer flows. In this river scenario, the simulations indicate that 99% of European river locations would be below 0.2 ng/L for carboplatin and below 0.6 ng/L for 5FU. With CP only 0.1% of locations could exceed 1 ng/L, whilst for capecitabine 2.2% could exceed 1 ng/L in rivers (with 0.2% in the 3-41 ng/L bracket). Of course it should be remembered the highest possible concentrations would be that of undiluted sewage effluent (Figure 2).

CONCLUSIONS

Overall, the availability of fairly recent drug consumption data from reliable sources and excretion data from the medical literature has put this model in a strong starting position. The potential impact on the modelling of differences in consumption, human excretion, sewage removal and dilution range from only 1.3-fold to 1000-fold (Table 4). Getting good information on all of these factors are of course important for

precision but here differences in consumption and hydrology are the most critical to get right. The good correlation between values predicted in this study and those observed in sewage effluent for CP give grounds for encouragement. We anticipate further refinement of the model will be possible in the future as more information on the fate and behaviour of these compounds becomes available.

Returning to the original objectives of the study we found there was a surprising difference in popularity of these cytostatics drugs across European nations, which can be up to 28-fold. The predicted mean effluent concentrations ranged from 2 to 40 ng/L for CP, 0.8 to 2.5 ng/L for carboplatin, 0.3-2.5 ng/L for 5FU, and 8.5-87 ng/L for capecitabine. In the majority of cases, where data is available, it is possible to predict CP concentrations in sewage effluent to within an order of magnitude of that observed. By linking with the geographic based water quality model it is expected that the majority of European rivers would have concentrations below 1 ng/L for these cytostatics drugs

The predicted river concentrations are considerably below concentrations so far reported to have effects on aquatic wildlife [8, 9]. As even in the 90%ile prediction around 80% of European surface waters for these drugs were largely below 0.1 ng/L, there does not appear to be any widespread threat to European aquatic wildlife based on our current knowledge. The issue of water abstraction for drinking water and foetal health may still require further research. Given its potentially high effluent concentrations and good oral absorption by humans, capecitabine certainly seems worthy of further environmental research.

Acknowledgement- The authors are grateful for support from the European Union (European Commission, FP7 project PHARMAS, contract no. 265346) and acknowledge the considerable support of CEH library services at Wallingford led by A. Smith and D. Galliford.

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Table 1. National cytostatic drug consumption information normalised from annual use to per capita using the national population at the time of the survey cyclophosphamide (CP), carboplatin (Carb), 5-fluorouracil (5FU) and capecitabine (Cap)

Country	Source	Year	CP use (mg/cap/d)	Carb use (mg/cap/d)	5FU use (mg/cap/d)	Cap use (mg/cap/d)
Germany	http://www.umweltbundesamt.de/uba-info-medien/dateien/3744.htm	2002	0.0084	0.0034	0.0519	0.1838
France	AFSSAPS quoted by [5]	2008	0.0129	0.0035	0.0730	0.2164
England	UK Dept of Health quoted by [37]	2005	NA*	0.0021	NA	0.1335
Italy	OsMed 2011	2011	0.0034	NA	0.0032	0.1476
Netherlands	http://www.gipdatabank.nl/	2009	0.0027	NA	0.0185	0.3938
Austria	Drug data from [47]	1997	0.0134	0.0010	0.0410	Not used
Denmark	http://www.medstat.dk/statistics/#tabs-2	2009	0.0057	0.0010	0.0026	0.3882
Switzerland	IHA-IMS quoted by [25]	2002	0.0127	NA	0.0303	0.1701
Sweden	Apotekensservice at http://www.apotekensservice.se and Tandvårds- och Läkemedelsförmånsverket TLV; at http://www.tlv.se/beslut/sok/lakemedel/	2010	0.0357	0.0015	0.0302	0.2267
Norway	Norwegian Prescription Database at http://www.norpd.no/ or http://www.legemiddelforbruk.no	2010	0.0092	NA	0.0270	0.1167
Finland	Data from Kela (at http://www.kela.fi and http://asiointi.kela.fi/laakekys_app)	2010	0.0023	NA	5-FU not used	0.3728
Czech Republic	Data from SUKL at http://www.sukl.cz/modules/medication/search.php	2011	0.0102	0.0084	0.0198	0.4937
European mean value			0.0104	0.0030	0.0297	0.2585

*NA information not available or not found

Table 2. Proportion of cytostatic drug excreted unchanged by patients

Drug excreted	Reference	No patients	mean age	Cause	Dose (g/d)	Excretion %*
Cyclophosphamide	[28]	2	NG**	NG	1-2.6	14.5
	[48]	12	43	autoimmune	0.8-1.7	19
	[49]	16	26	bone marrow	3.5-4.2	14
	[50]	4	48	breast cancer	4.2	16.5
	[51]	19		NG	4.2	14
	[27]	6	NG	autoimmune	1.7	11
	[52]	15	44	NG	6.8-10	20
Carboplatin	[32]	14	NG*	healthy volunteers	0.03-0.9	32
	[53]	3	59	multiple cancers	0.27-0.68	69
	[51]	19	30	bone marrow	3.9	14
	[54]	7	55	ovarian cancer	0.49-0.63	41
	[55]	7	64	lung cancer	0.17	54
5FU	[56]	1	NG	Colon	1.0	39
	[57]	1	NG	Colorectal cancer	0.75	11
	[39]	8	NG	NG*	0.85	6.5
	[42]	22	61	Largely colorectal cancer	3.9	2.0
5FU from capecitabine	[36]	7	59	Range of cancers	2.0	0.5
	[58]	13	56	Range of cancers	2.1	0.7
	[59]	23	63	Range of cancers	2.0	0.7
	[60]	60	60	Colorectal cancer	4.25	0.8
Capecitabine	[36]	7	59	Range of cancers	2.0	2.9
	[58]	13	56	Range of cancers	2.1	2.6
	[59]	23	63	Range of cancers	2.0	2.7
	[60]	60	60	Colorectal cancer	4.25	3.4

*Where two excretion values were given for separate days of treatment, the average value is reported
**NG Information not provided

Table 3. Review of fate studies with 5FU in sewage environments

Reference	Environment	Concentration (μg/L)	Removal (%)	Time period	Comment
[61]	Sewage inoculums in OECD tests	9000-854,000	No removal		High concentrations were inhibitory?
[29]	Lab act. sludge pilot plant	3,000	96-100% from 2 d onwards	14 d	Population became adapted?
[43]	Act. Sludge microcosm	5	92-98% (90% at 15 h)	24 h	Replication not reported
[62]	Hospital biologically live storage tank	15-98	100%	Approx. 24 h	Biological treatment conditions unclear
[63]	Sewage inoculated biofilm column	10	97	200 d (hydraulic residence time 8 h)	Attempting to replicate soil aquifer treatment
[64]	Growth medium inoculated with act. sludge	1-50	50	50 d	A weak inoculum possibly changed from usual sludge community structure

Table 4. Summary of variables and their potential effects on the predictive modelling of cyclophosphamide (CP), carboplatin (Carb), 5-fluorouracil (5FU) and capecitabine (Cap)

Drug	Range in consumption across EU	Weighted mean and range in patient excretion values (%)	Effect on sewage influent conc.	Mean and range in sewage treatment removal (%)	Effect on sewage effluent conc.	Range in European dilution potential (m^3/cap/d) **	Effect on river conc.
CP	15-fold	15.9 (11-20)	1.8-fold	0	No difference		
Carb	8-fold	31.7 (14-69)	5-fold	72 (59-85)	2.7-fold	$2.8 - 2.7 \cdot 10^3$	1000-fold
5FU	28-fold	4.6 (2-39)*	19-fold	95 (92-99)	8-fold		
Cap	4-fold	3.1 (2.6-3.4)	1.3-fold	50 (25-75)	3-fold		

*5FU has a weighted mean of 0.7 and a range of 0.5-0.8% when excreted by patients taking capecitabine

**10%ile to the 90%ile of dilution values calculated on a cell-by-cell basis using 1970-2000 average river discharge

Table 5. Comparing literature measured cyclophosphamide with predicted values for sewage effluents

Reference	Location	Calculated water use L/cap/d	LOD (ng/L)	Reported value (ng/L)	Predicted value (ng/L)	Comments on simulation
Thomas et al 2007	VEAS STP Oslo	722	2	<2	2.0	Acceptable
Kanda et al 2003	6 x STP UK	160	23	<23	10.4	Agree
Yin et al 2010	7 x STP beijing, China	160	0.8	8.5-14.5	10.4	Agree
Buerge et al 2006	Zurich STP	630	0.3	2.1-4	3.3	Agree
Metcalfe et al 2003	18 x STP Canada	527	100	<100	3.2	Agree
Garcia-Ac et al 2011	Montreal STP Canada	329	0.5	12	5.1	Agree
Hua et al 2006	Windsor STP Canada	329	1	2.5-4	5.1	Agree
Llewellyn et al 2011	ASP H UK	200	0.04	0.2	8.3	Fail
	Biol filt L UK	300	0.12	3.6	5.5	Agree
Calamari et al 2003	Po & Lambro R, Italy	203	0.01	<0.01	1.4	Fail
Zuccato et al 2005	9 x STP Italy	203	NA*	0.6	2.7	Acceptable
Martin et al 2011	1 x STP Spain	153	1.7-2.3	0.6	10.9	Fail
Gomez-Canela et al 2012	3 x STP Spain	153	3.1	<3.1	10.9	Acceptable
Busetti et al 2009	1 x STP Australia	480	125	<125	3.5	Agree

*NA Information not provided

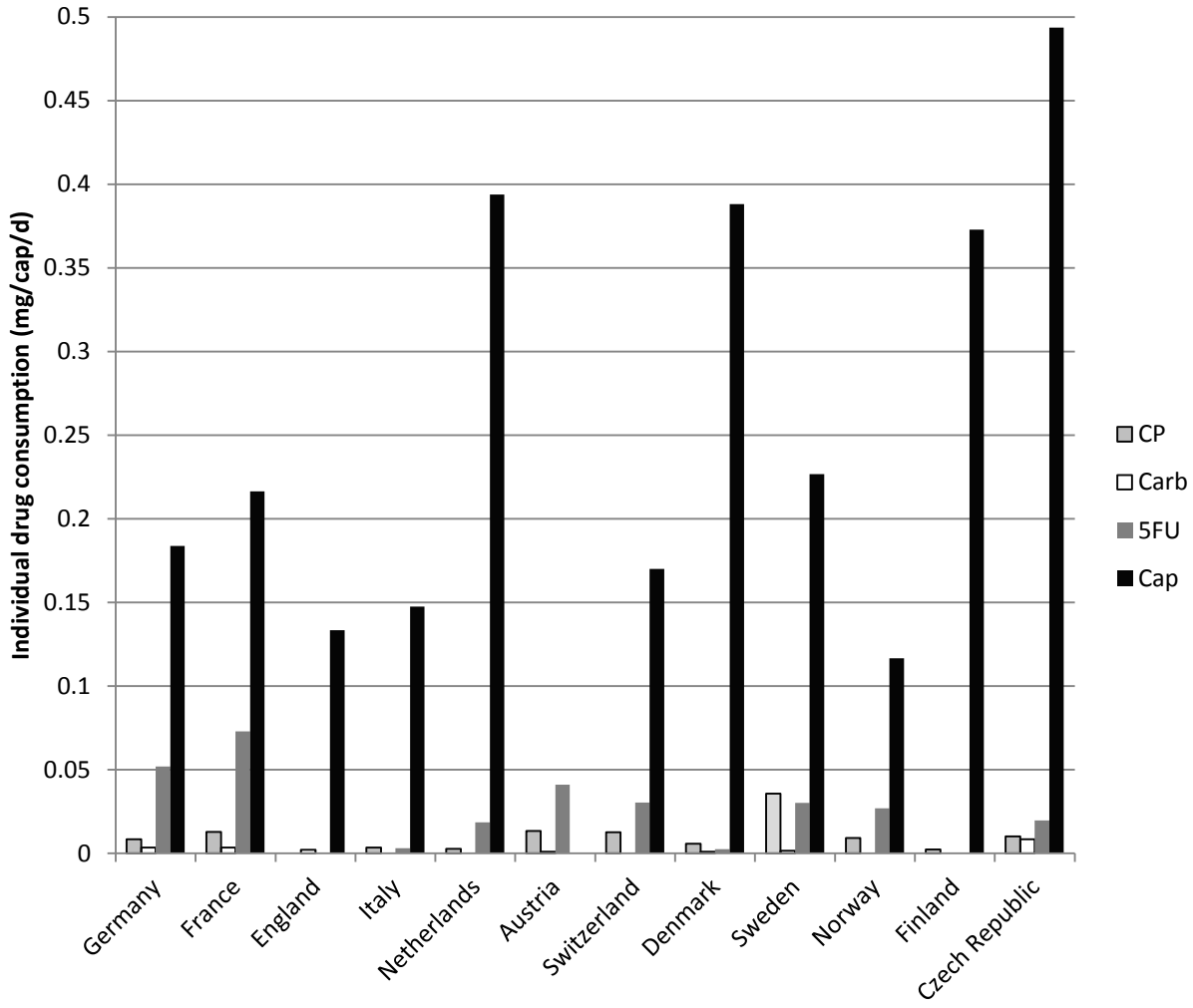

Figure 1. Variations in per capita cyclophosphamide (CP), carboplatin (Carb), 5-fluorouracil (5FU) and capecitabine (Cap) consumption between different European nations. Note these values do not all come from the same year.

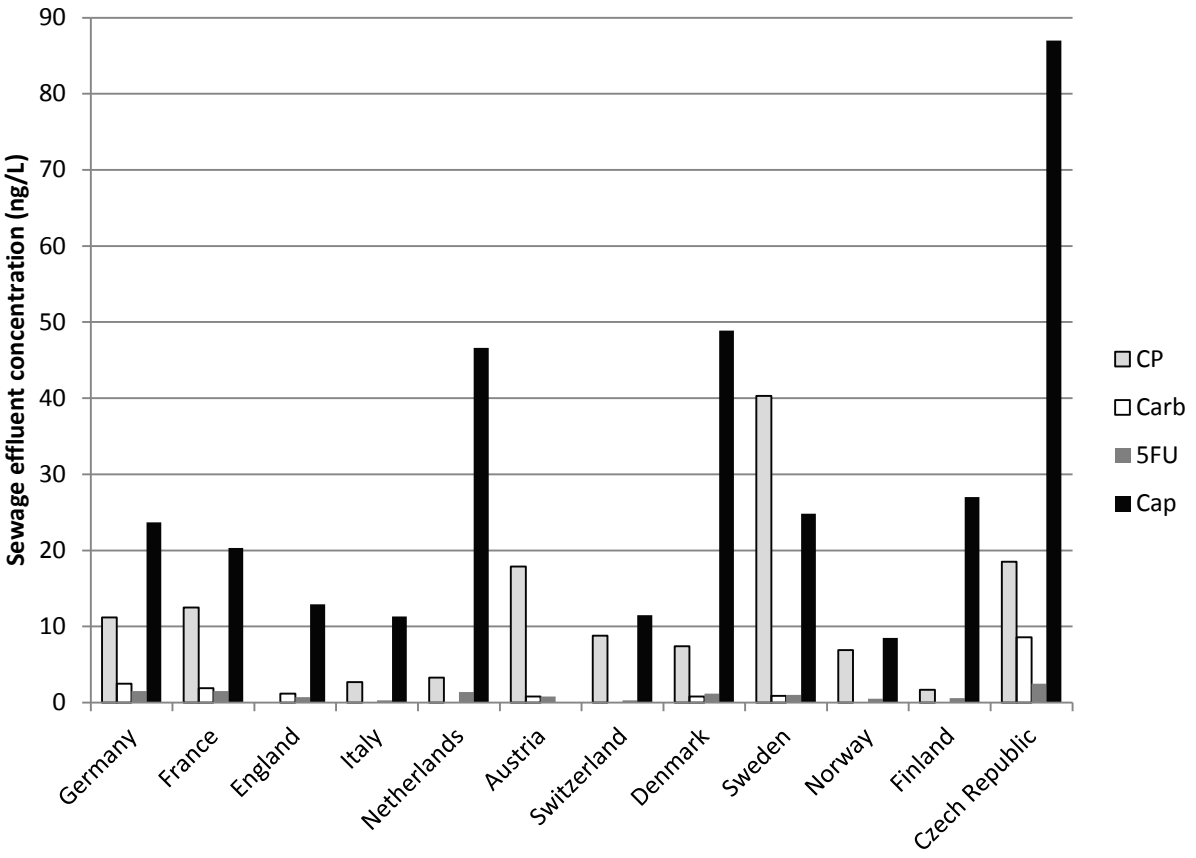

Figure 2. Predicted mean cyclophosphamide (CP), carboplatin (Carb), 5-fluorouracil (5FU) and capecitabine (Cap) concentrations in sewage effluent for European nations taking into account differing national per capita wastewater discharge values.

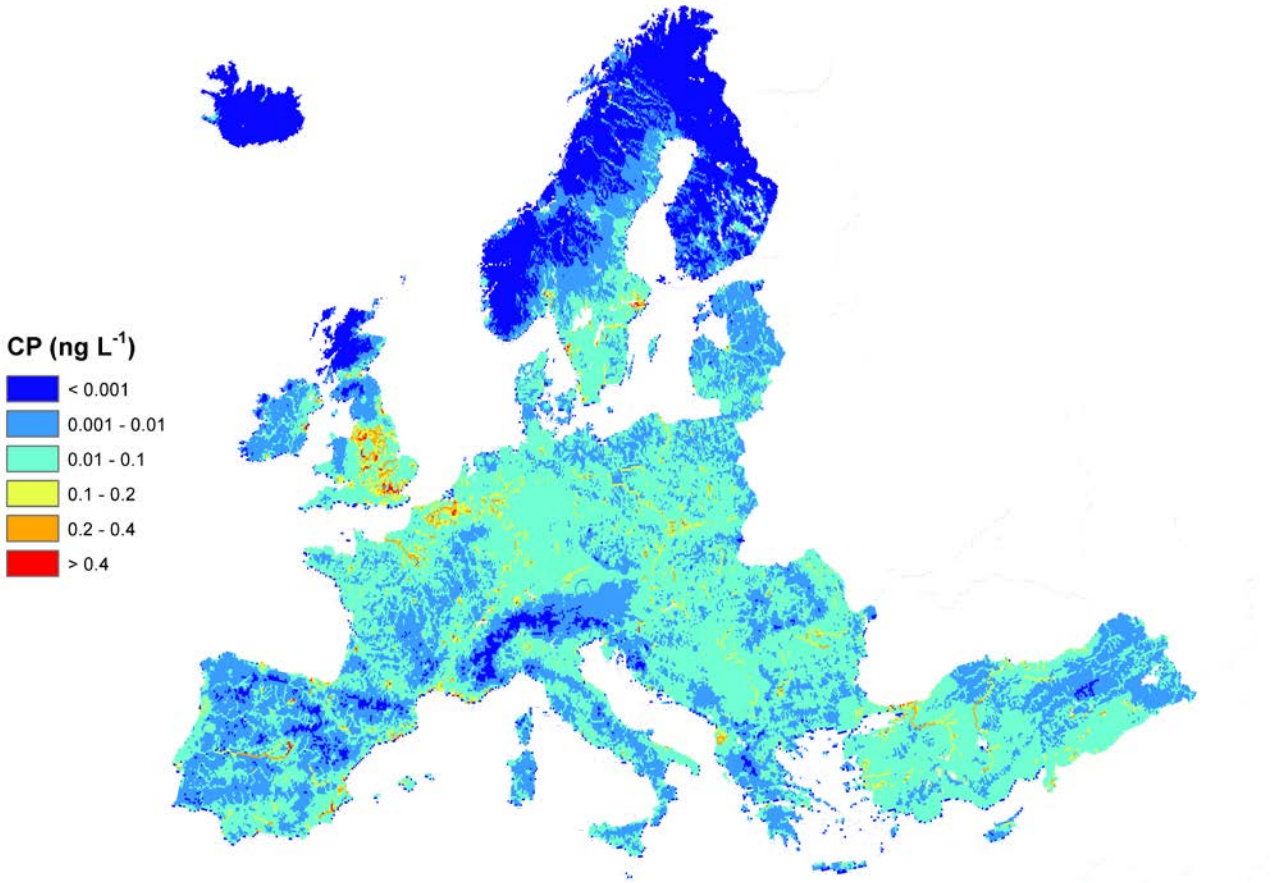

Figure 3. Predicted cyclophosphamide (CP) concentrations in surface water based on mean excretion rate, mean sewage treatment removal, and 50%ile flow across the European Continent taking into account differing national per capita consumption and wastewater discharge values from the GWAVA model.

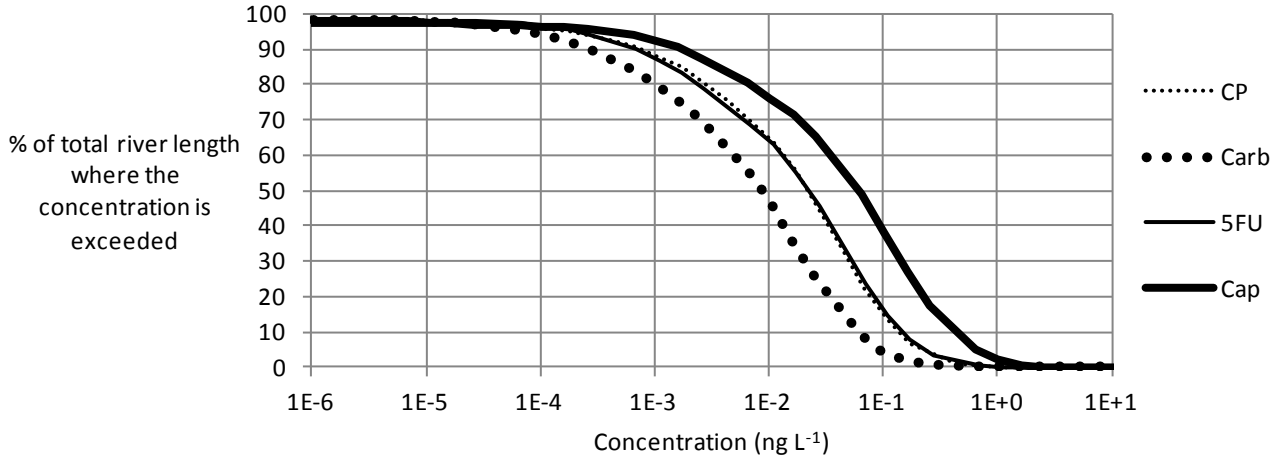

Figure 4. Predicted 90%ile concentrations for cyclophosphamide (CP), carboplatin (Carb), 5-flourouracil (5FU) and capecitabine (Cap) in surface water assuming high excretion rates and low sewage treatment removal across the whole European Continent from the GWAVA model plotted as cumulative frequency curves.