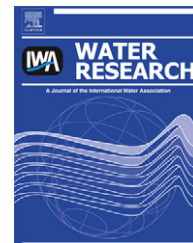


Available at www.sciencedirect.comjournal homepage: www.elsevier.com/locate/watres

Occurrence and suitability of sucralose as an indicator compound of wastewater loading to surface waters in urbanized regions

Joan Oppenheimer^{a,*}, Andrew Eaton^b, Mohammad Badruzzaman^a, Ali W. Haghani^b, Joseph G. Jacangelo^{a,c}

^a MWH Americas Inc., 618 Michillinda Avenue, Arcadia, CA 91007, USA

^b MWH Laboratories, 750 Royal Oaks Avenue, Monrovia, CA 91016, USA

^c The Johns Hopkins University, Baltimore, MD 21205, USA

ARTICLE INFO

Article history:

Received 23 February 2011

Received in revised form

11 May 2011

Accepted 12 May 2011

Available online 25 May 2011

Keywords:

Sucralose

Wastewater indicator

Pharmaceuticals and personal care products

Urban watershed

ABSTRACT

Urban watersheds are susceptible to numerous pollutant sources and the identification of source-specific indicators can provide a beneficial tool in the identification and control of input loads, often times needed for a water body to achieve designated beneficial uses. Differentiation of wastewater flows from other urban wet weather flows is needed in order to more adequately address such environmental concerns as water body nutrient impairment and potable source water contamination. Anthropogenic compounds previously suggested as potential wastewater indicators include caffeine, carbamazepine, N,N-diethyl-meta-toluamide (DEET), gemfibrozil, primidone, sulfamethoxazole, and TCEP. This paper compares the suitability of a variety of anthropogenic compounds to sucralose, an artificial sweetener, as wastewater indicators by examining occurrence data for 85 trace organic compounds in samples of wastewater effluents, source waters with known wastewater point source inputs, and sources without known wastewater point source inputs. The findings statistically demonstrate the superior performance of sucralose as a potential indicator of domestic wastewater input in the U.S. While several compounds were detected in all of the wastewater effluent samples, only sucralose was consistently detected in the source waters with known wastewater discharges, absent in the sources without wastewater influence, and consistently present in septic samples. All of the other compounds were prone to either false negatives or false positives in the environment.

© 2011 Elsevier Ltd. All rights reserved.

1. Introduction

Surface waters and shallow unconfined aquifers in urbanized regions are vulnerable to pollutants that cause impairment from previously established beneficial uses. Pathogens have been identified as the top impairment of assessed U.S. rivers and streams with major sources attributed to municipal

discharges/sewage, unspecified non-point source discharges, and urban runoff/stormwater (USEPA, 2009). Chemical indicators have been proposed as alternatives to microbial indicators as a more definitive means of identifying fecal contamination from human sources (Glassmeyer et al., 2005). Nitrogen input levels and oxic conditions are the major variables correlated with higher observed nitrate concentrations in groundwaters

* Corresponding author. Tel.: +1 626 568 6006; fax: +1 626 568 6015.

E-mail address: Joan.Oppenheimer@us.mwhglobal.com (J. Oppenheimer).

0043-1354/\$ – see front matter © 2011 Elsevier Ltd. All rights reserved.

doi:10.1016/j.watres.2011.05.014

throughout the United States (Burow et al., 2010). Quantifying water body pollutant mass loads back to contributing sources within an urbanized watershed is complex and frequently depends upon modeling strategies that incorporate estimates and uncertainties about each source's flow and pollutant concentration levels (Nix, 1994). Mass loadings are often times further attenuated by location specific fate and transport processes that are not adequately characterized. In order to effectively mitigate impaired water bodies, it is important to identify the major contributing sources, so effective best management practices can be established.

Urban water pollution originates from wastewater point source discharge of treated effluents, wastewater non-point source intrusions from septic tanks, misapplication of recycled wastewater used for irrigation, or urban runoff from mobilization of pollutants deposited to impervious surfaces through atmospheric deposition and human activities during small rain events, erosion from pervious surfaces during large storm events, and dry weather flows (Paul and Meyer, 2001). Developing an indicator specific to wastewater effluents would aid in determining the contribution of these sources toward water quality impairment in recreational waters or potable supply sources (Glassmeyer et al., 2005; Stackelberg et al., 2004).

The characteristics of an ideal wastewater indicator include: (i) source specificity, (ii) sustained effluent release because the indicator is not rapidly degraded by biological treatment processes, (iii) demonstrated analytical methodology, (iv) no attenuation during transport, and (v) virtually zero background with a sufficiently large discharge to detection level ratio able to exceed receiving water dilution factors. Traditional markers such as stable isotope ratios (e.g. $\delta^{15}\text{N}$) and inorganic ions (e.g. chloride) often lack source specificity (Gasser et al., 2010; Katz et al., 2004). Data interpretation of stable isotope ratios is also difficult due to fractionation which obscures mixing during environmental transport (Gasser et al., 2010; Fry, 2006).

Reconnaissance surveys of surface water bodies in the United States (Kumar and Xagoraki, 2010) have suggested several anthropogenic organic compounds used as pharmaceuticals, personal care products, food products, pesticides, and hospital wastes as potential chemical markers of pollutant loading due to their behavior as persistent aqueous organic pollutants (Benotti et al., 2009; Bester, 2007; Buerge et al., 2009; Focazio et al., 2008; Glassmeyer et al., 2005; Guo and Krasner, 2009; Jjemba, 2008; Standley et al., 2008; Yamamota et al., 2009). Table 1 summarizes these candidate anthropogenic organic compounds and that which is known about their characteristics to serve as a wastewater marker due to occurrence in treated wastewater effluents, resistance to secondary wastewater treatment process operations, and persistence in U.S. waters due to a sustained rate of discharge which appears to exceed environmental attenuation processes from partitioning (i.e. volatilization, sorption, biotic uptake) or transformation (i.e. biodegradation, sunlight photolysis, and abiotic hydrolysis or redox reactions). The literature presents a potential short-list of compounds that still warrant a more methodical evaluation of their performance resistance to false positives and false negatives in identifying wastewater loading to surface water bodies. Notably absent from the short-list for most studies performed in the United States are artificial sugar

substitutes which have been included in several reconnaissance surveys in Europe (Scheurer et al., 2011, 2010, 2009; Buerge et al., 2009; Brorström-Lundén et al., 2008; Loos et al., 2009). Acesulfame, cyclamate, saccharin, and sucralose are commonly consumed in Europe with per capita wastewater loads (mg/Cap d) in Switzerland estimated at 10 ± 3.4 for acesulfame, 11 ± 6.7 for cyclamate, 3.9 ± 1.7 for saccharin, and 1.5 ± 0.6 for sucralose (Buerge et al., 2009) and values in Germany observed at 4.6 and 6.9 for acesulfame, 0.07 and 0.5 for cyclamate, 0.5 and 0.6 for saccharin, and 0.11 and 0.18 for sucralose (Scheurer et al., 2009). Sucralose per capita wastewater loads have been estimated at only 0.14–0.23 mg/(Cap d) in Germany and at 1.7–2.1 mg/(Cap d) in Sweden (Neset et al., 2010). Cyclamate and saccharin are unsuitable as markers because high levels of reduction (>90%) through biological treatment result in low effluent concentrations and cyclamate is additionally unsuitable in the United States since it has been banned from distribution since 1970. Acesulfame has been suggested as the most suitable chemical marker of domestic wastewater in Europe because of its approximate ten-fold higher concentration than sucralose in European wastewater effluents (approximately 10–50 $\mu\text{g/L}$ in influents/effluents for acesulfame and <1–10 $\mu\text{g/L}$ for sucralose). No information is available on per capita wastewater loads of sucralose in the United States, but it is anticipated to be higher than Europe due to its much longer period of availability (U.S. introduction in 1988 versus 2004 in Europe) and different regional dietary habits. Sucralose degradation through wastewater treatment facilities has also been demonstrated to be minimal for measurements through full-scale facilities and laboratory-scale aerobic biodegradation reactors (Torres et al., 2011; Buerge et al., 2009; Scheurer et al., 2009, 2010; and Neset et al., 2010).

Pharmaceutical usage patterns were shown to differ between Europe and the U.S. (Sedlak and Pinkston, 2001) and differences in artificial sweetener usage and consumption is also probable. Other than one publication tracing sucralose along the salinity transect of a North Carolina river estuary impacted by municipal wastewater treatment plant effluent discharged upstream, this study is the first to evaluate the potential use of the artificial sweetener sucralose as a chemical marker for domestic wastewater input to surface waters within the United States. The evaluation considers sucralose presence in municipal wastewater effluents relative to other water sources and its susceptibility to false positives and false negatives through analysis of source waters with and without municipal wastewater discharges.

2. Materials and methods

2.1. Study sites

Samples collected for analysis were obtained from municipal wastewater treatment facility effluents, from drinking water intake sources with and without known wastewater point source discharges upstream of the intakes, and from active septic systems. The wastewater effluent locations were obtained from facilities located in Florida, Texas, northern and southern California, Illinois, and Michigan. The source waters with known upstream municipal wastewater discharges were

Table 1 – Reported detection frequencies (DF) of organic chemicals in wastewater treatment plant (WWTP) effluents, percent removal through activated sludge processes, and persistence in the environment as occurrence rank for U.S. streams and source waters.

Compound (usage)	Average DF (%) in WWTP effluent ^a	Activated sludge treatment average percent removal ^b	Occurrence rank of compounds in U.S. stream/source water ^c
acetaminophene (anti-inflammatory)	low concentration ^d	97 (n = 4)	40
atenolol (beta blocker)	low concentration ^d	61 (n = 4)	25
atrazine (pesticide)	low concentration ^d	no data	1
benzophenone (fragrance)	100	84 (n = 6)	30
bisphenol-A (plasticizer)	low concentration ^d	78 (n = 41)	3
caffeine (stimulant)	81	94 (n = 7)	not listed
1,4-dichlorobenzene (household cleaning product)	not on target list	not listed	not listed
1,7-dimethylxanthine (caffeine metabolite)	not on target list	77 (n = 1)	48
carbamazepine (mood stabilizer)	88	22 (n = 5)	4
codeine (cough suppressant)	84	29 (n = 1)	78
coprostanol (sterol)	low concentration ^d	97 (n = 1)	not listed
cholesterol (sterol)	low concentration ^d	85 (n = 1)	not listed
cotinine (nicotine metabolite)	low concentration ^d	not listed	5
N,N-diethyl-meta-toluamide (DEET) (insect repellent)	89	54 (n = 7)	22
diazinon (pesticide)	not on target list	not listed	not listed
diclofenac (anti-inflammatory)	75	44 (n = 23)	70
dehydronifedipine	not on target list	not listed	9
diltiazem (channel blocker)	not on target list	not listed	75
diphenylhydramine (antihistamine)	91	not listed	35
diuron (pesticide)	not on target list	not listed	not listed
estrone (hormone)	low concentration ^d	77 (n = 46)	73
ethyl citrate (food additive)	not on target list	not listed	28
galaxolide (HHCB) (fragrance)	100	56 (n = 25)	29
gemfibrozil (anti-cholesterol)	92	77 (n = 13)	79
ibuprofen (anti-inflammatory)	78	90 (n = 32)	93
meprobamate (muscle relaxant)	83	not listed	21
metolachlor (herbicide)	not on target list	not listed	42
naproxen (anti-inflammatory)	92	85 (n = 18)	62
4-nonylphenol (detergent)	100	78 (n = 10)	67
phenytoin (dilantin) (seizure control)	100	44 (n = 1)	23
primidone (mood stabilizer)	100	not listed	not listed
β-sitosterol (plant steroid)	not on target list	not listed	not listed
sulfamethoxazole (antibiotic)	94	58 (n = 15)	24
tonalide (AHTN) (fragrance)	100	67 (n = 20)	6
tributylphosphate (flame retardant)	not on target list	not listed	not listed
triclosan (antimicrobial)	98	60 (n = 10)	54
trimethoprim (anti-bacterial)	86	not listed	39
tris(2-butoxyethyl)phosphate (flame retardant)	not on target list	not listed	2
tris(2-chloroethyl)phosphate (flame retardant)	low concentration ^d	27 (n = 2)	20
tris(dichlorisopropyl)phosphate (flame retardant)	100	not listed	7

a Dickenson et al., 2010.

b USEPA, 2010.

c Kumar and Xagorarakis, 2010.

d <5 × limit of quantitation (LOQ).

located in northern and southern California, Colorado, Ohio, and New York. These samples were taken in the vicinity of the drinking water intake and while the percentage of wastewater present during sample collection is not known, the California location has been shown to contain sewage treatment plant effluent in the range of 20–70 percent of the flow during a severe drought year (Loraine and Pettigrove, 2006). The source waters classified without municipal wastewater discharges were located in northern and southern California, New York, Michigan, and Illinois and consisted either of rivers without upstream discharges or extremely large lakes with intakes removed from

known discharges. The septic samples were collected from systems located in Leon County and Palm Beach County, Florida.

2.2. Sample collection and preservation

Samples were collected in 40 mL amber glass vials and field preserved with either a combination of 5 mg ascorbic acid and 50 mg sodium azide (Benotti et al., 2009; Vanderford et al., 2010) or 3 mg sodium omadine and 5 mg ascorbic acid (USEPA, 2010). Samples were transported on frozen blue ice and received in the laboratory either the same day or the next

day after collection and then stored refrigerated at $< 6^{\circ}\text{C}$. Both preservatives have been shown to be effective for stabilizing many pharmaceuticals and personal care products (PPCPs) for 28 days or more under these conditions. All sample containers were provided by a single laboratory and sampling was performed with chain-of-custody documentation.

2.3. Analysis

A fully automated on-line solid phase extraction, high performance liquid chromatography, mass spectrometry-mass spectrometry (SPE-HPLC-MS/MS) system was developed for rapid analysis of various groups of compounds of emerging concern (CECs) in water matrices. A column switching technique similar to the method of Buerge et al. (2008) was used for analysis of sucralose and other PPCP's. The mass spectrometer used was an API 5000 triple quadrupole (AB/SCIEX) equipped with electrospray ionization (ESI) (TurboIonSpray®). The interface setting and collision gas pressure were manually optimized. Parameter tuning for maximum sensitivity of multi reaction mode (MRM) detection in positive and negative ion mode was carried out by means of the optimization algorithm supported by the Analyst 1.5 software using the TurboIonSpray® interface and a syringe diffusion pump

continuously supplying a standard (10 $\mu\text{L}/\text{min}$, $C = 10 \mu\text{g}/\text{L}$ for each compound). The resulting instrument values were then cross-checked for their validity under ESI conditions by flow injection MS/MS analysis (FIA-MS/MS) of the standard mixture and manual variation of the settings. Multiple mass transitions were used for each analyte (wherever there was adequate sensitivity for both transitions) to ensure unequivocal compound identification. For sucralose, both transitions were detectable at the MRL of 100 ng/L . The second transition was approximately 1/3 of the primary transition, but both transitions were more than ten times above the signal to noise ratio.

Sample extraction was carried out using an integrated Dionex UltiMate 3000 system comprised of two HPG-3200 SD binary pumps, a WPS3000 SL semi-preparative auto sampler with 2.5 mL sample loop, and a TCC-3000SD column heater equipped with a ten port switching valve (Dionex, Sunnyvale, CA). An XBridge-C18 ($2.1 \times 150 \text{ mm}$ $3.5 \mu\text{m}$ particle size) column (Waters, Milliford, MA) was used for ESI negative mode and a Luna C18(2)-HST ($3 \times 100 \text{ mm}$, $2.5 \mu\text{m}$ particle size) column (Phenomenex, Torrance, CA) was used for ESI positive mode. An Oasis HLB ($2.1 \times 10 \text{ mm}$ $25 \mu\text{m}$) cartridge (Waters, Milliford, MA) was selected as the on-line SPE sorbent which contains both hydrophilic and hydrophobic materials suitable for multiresidue analysis in environment. One of the HPG-

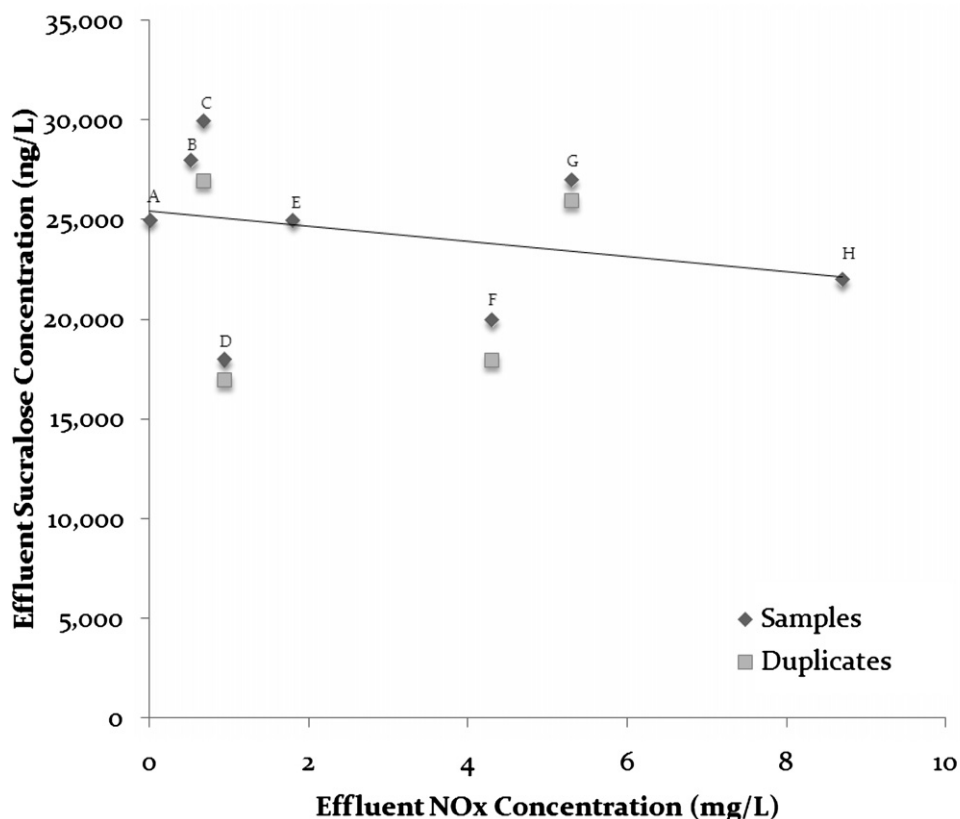


Fig. 1 – Sucralose concentration in wastewater effluents collected from facilities with varying levels of nitrification and denitrification (A = oxidization ditch with methanol feed to denitrification filter, B = bardenpho with dual media deep bed filters, C = conventional activated sludge with carousel aeration and denitrification basin; D = complete mix activated sludge with biological nutrient removal, E = activated sludge with fine bubble diffused air and filtration, F = modified Ludzak-Ettinger activated sludge, G = complete mix activated sludge with anoxic basin, H = oxidation ditch). Duplicates are a second sample bottle collected from the same site.

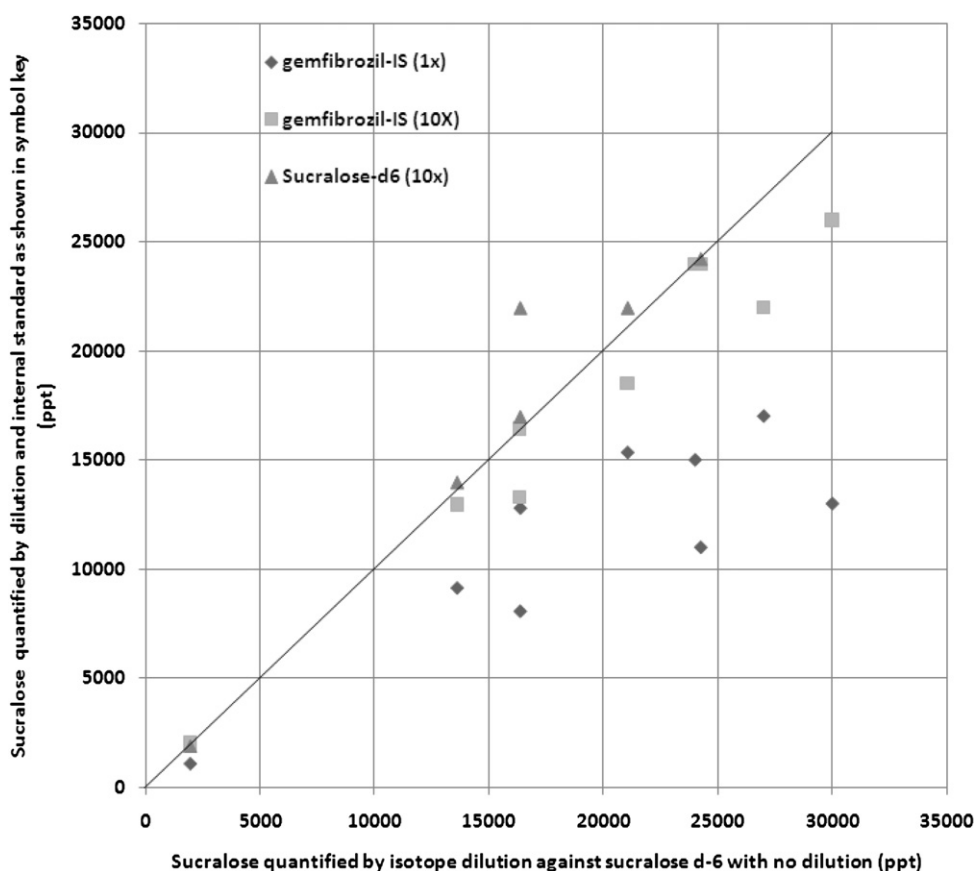


Fig. 2 – Effect of dilution factor and internal standard on sucralose quantification.

3200 SD binary pumps was used for sample delivery to the on-line SPE at a flow rate of 1.2 mL/min. Prior to extraction the short Oasis HLB cartridge was conditioned with 5 mL acetonitrile and 5 mL HPLC grade water acidified to pH 3. The delivery eluent pH was adjusted to approximately pH 3 by adding 250 µL of formic acid per 1 L of HPLC grade water. These

conditions represent a best compromise for the target analyte list and the HPLC eluant of 0.2% ammonium hydroxide and acetonitrile and the acidic pH 3 condition used to load the on-line SPE were both observed to improve the peak shape for sucralose. For both ESI modes the organic eluent B was acetonitrile and Eluent A for ESI positive mode was HPLC

Table 2 – Batch QC per Analytical Run.

QC type	Frequency	Spiking level	Acceptance criterion
calibration curve	each analytical batch ^a	minimum 5 point curve plus blank	quadratic fit with 1/× weighting and correlation coefficient of 0.99; back-calculated to +/- 30% of true value (+/- 50% at MRL level)
method blank	each analytical batch	0	< 1/3 MRL
MRL check	each analytical batch	low cal level	50–150%
continuing calibration	every 10 samples	5 × MRL level	70–130%
sample matrix spikes	every 20 samples	5 × MRL level	60–140%
closing blank	end of run	0	< 1/3 MRL
closing standard	end of run	10 × MRL	70–130%
identification criteria	each detect	retention time	+/- 1 s
identification criteria	each detect	mass transitions	2 MRM if available, ion ratio based on EU criteria ^b

a An analytical batch consists of no more than 20 samples analyzed in a single 24-h run.

b Draft SANCO 1805/2000 Rev. 1. [Revised Commission Decision 93/256 of 14 april 1993] laying down performance criteria for the analytical methods to be used for certain substances and residues thereof in live animals and animal products according to Council Directive 96/23/EC.

grade water adjusted to pH 3. For the ESI negative mode, due to the list of hormones and alkylphenols, a 0.2% solution of ammonium hydroxide was used. 2.5 mL of unfiltered sample was loaded to the 2.5 mL auto sampler loop and delivered to the on-line SPE and washed with 5% acetonitrile for 2 min, and the matrix diverted to waste. The valve position was then changed and the target analytes were refocused on the analytical column and then separated and eluted into the mass spectrometer using a gradient of 10% eluent B for 4 min, 100% eluent B at 20 min, then 5 min equilibration time at 10% eluent B again, and this conditions was kept for another 5 min during the next sample upload into the sample loop.

Sucralose and other PPCPs (85 total analytes) were analyzed using the above method, with the exception of the sucralose data provided in Fig. 1 which was performed by the Florida Department of Environmental Protection using the EPA Method 3535 offline pre concentration technique and EPA Method 8321B. For the on-line concentration method, all standards and QC samples were processed the same way. Results for 44 analytes were determined via isotope dilution and when isotopes were unavailable, a close eluting isotope was utilized. Sucralose data was quantified using both gemfibrozil and sucralose-d6 as the internal standard. Fig. 2 provides a comparison of sucralose results for a suite of samples that were evaluated using multiple quantitation techniques, including dilution and quantification against gemfibrozil as an internal standard at both 1× and 10× and also quantification against sucralose-d6 at both 1× and 10× dilutions. At low concentrations (which were less “dirty” matrices) there is minimal difference between quantitation

techniques. The sucralose signal suppression from background matrix could be largely corrected by dilution, although quantitation was typically 15% lower using 10× dilution and gemfibrozil as the internal standard as compared with sucralose d6 isotope dilution. The recovery for sucralose-d6 was greater than 50%. Since all samples quantified with gemfibrozil were diluted ten-fold or more before analysis to minimize matrix effects, data reported by either internal standard method are comparable.

2.4. Quality assurance

Minimum reporting levels (MRLs) for analytes ranged from 1 to 100 ng/L. MRLs were determined as the lowest calibration point used which also corresponds to a S/N ratio of at least 10, assuring accurate and precise quantitation (91% average recovery and relative standard deviation of 39% for sucralose and 106% average recovery and relative standard deviation of 15% for carbamazepine over a 15 month period) at the MRL levels of 100 and 5 ng/L respectively. Each analytical batch included numerous quality control (QC) samples as summarized in Table 2.

3. Results and discussion

3.1. Database evaluation of conservative indicators

A summary of the compounds detected in the wastewater effluents and two categories of source waters are presented in Table 3. Only compounds detected in at least 35% of the

Table 3 – Compounds detected in wastewater effluents and source waters with and without municipal wastewater discharges.

Compound (MRL, ng/L)	Wastewater (ww) effluents				Sources with ww discharges			Sources without ww discharges		
	mean (ng/L)	rsd (%)	detects (%)	n	detects (%)	n	detect range (ng/L)	non detects (%)	n	detect range (ng/L)
sucralose (100)	27,000	30	100	16	100	11	120–10,000	100	15	–
diuron (5)	99	78	100	12	82	11	7.5–940	80	15	5.3–6.7
simazine (5)	21	100	100	12	73	11	24–160	20	15	7.1–61
DEET (5)	269	135	100	12	73	11	2.5–67	13	15	2.2–7.1
meprobamate (5)	323	197	100	12	70	10	5.5–160	100	15	–
caffeine (10)	127	159	75	12	64	11	13–300	100	15	–
diaminochlorotriazine (5)	36	209	67	12	64	11	13–300	40	15	10–100
TCEP (5)	547	66	92	12	60	10	7.9–66	47	15	13–64
bromacil (5)	95	100	50	12	55	11	6–270	93	15	290
sulfamethoxazole (10)	907	116	80	10	55	11	17–990	100	15	–
primidone (5)	159	49	100	12	50	8	20–54	100	15	–
2,4-D (5)	248	262	83	12	44	9	11–23	60	15	7.4–21
amoxicillin (20)	1230	92	71	7	45	11	25–2200	100	14	–
iohexal (10)	4780	120	100	16	45	11	73–960	87	15	16–39
atenolol (5)	1310	1070	100	16	45	7	6.1–200	92	13	19
carisoprodol (5)	119	156	92	12	40	10	5.4–43	100	15	–
gemfibrozil (5)	360	131	83	12	40	10	13–130	100	15	–
carbamazepine (5)	416	21	100	16	36	11	31–190	100	15	–
1,7-dimethylxanthine (5)	98	160	75	12	36	11	8.9–23	100	13	–
cotinine (10)	29	86	100	8	36	11	13–27	100	15	–
dehydronifedipine (5)	119	92	92	12	36	11	12–120	87	15	7.7–70
lopressor (20)	3900	149	67	12	36	11	22–270	100	13	–
theobromine (5)	151	158	42	12	36	11	6.4–41	67	15	7.8–25

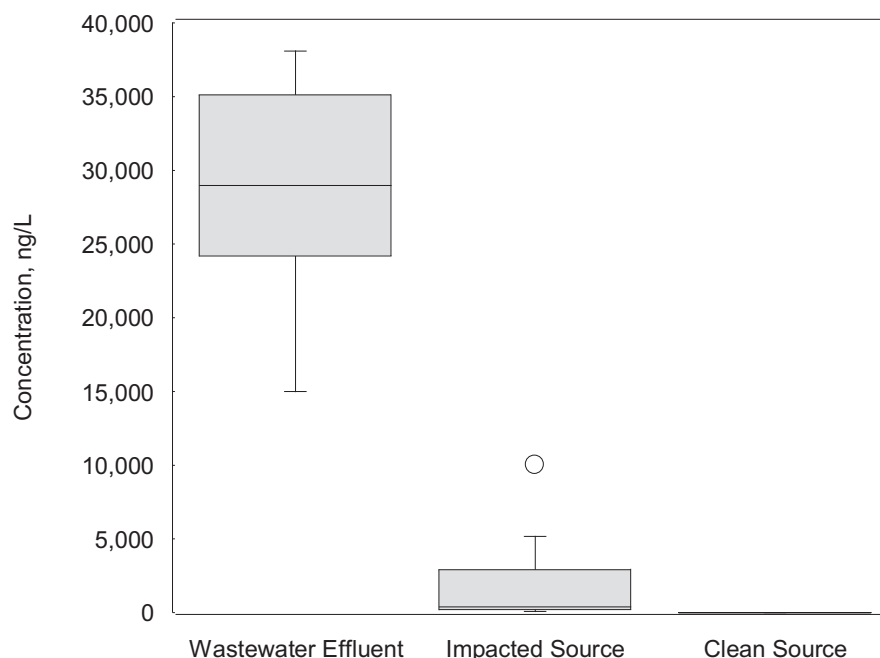


Fig. 3 – Nonparametric probability distribution of sucralose concentrations in different matrices depicted as box and whisker plots. Top and bottom of each box = 75th and 25th percentiles, respectively; top and bottom of each whisker = 90th and 10th percentile respectively; line across inside of each box = median (50th percentile); and points beyond whiskers = outliers. Clean source samples were all below the method reporting limit (MRL) and represented as a line at the 100 ng/L MRL.

sources with known wastewater discharges were captured in the table. Good markers of wastewater source loading should exhibit a high ratio of mean concentration to method reporting limit with 100 percent detection in the wastewater effluents. Low variability, denoted as % rsd in Table 3, also demonstrates a compound's potential to serve as an indicator of the extent of wastewater impact (i.e., a correlation between concentration in the receiving stream and the fraction of stream flow due to upstream wastewater discharges) provided that the compound is stable in the environment during transport. The table shows that of the compounds exhibiting 100 percent detection in wastewater effluents, the best performing compounds in terms of ratio of mean concentration to MRL and lack of false positives were sucralose, meprobamate, and carbamazepine. The low relative standard deviation of sucralose and carbamazepine also indicates their potential to serve as quantitative markers of wastewater input, provided their environmental stability is adequately demonstrated. To function as an indicator of wastewater input in the environment, a compound should also demonstrate a high detection frequency in sources with known wastewater discharges (no false negatives) as well as absence from sources without known wastewater discharge influence (no false positives) (Gasser et al., 2010). For the samples and analytical methodology utilized to generate the dataset presented in Table 3, only sucralose demonstrated no false positives or false negatives. False positives demonstrate lack of source specificity and false negatives demonstrate lack of adequate sensitivity. Amoxicillin, carbamazepine, caffeine,

cotinine, gemfibrozil, meprobamate, primidone, sulfamethoxazole exhibited false negatives, stressing the need for detectability below a 5–10 ng/L MRL. Diuron, simazine, DEET, iohexal, and atenolol, exhibited false positives as well as false negatives. The efficacy of sucralose as a marker of conventional biologically treated wastewater is further demonstrated by Fig. 1 which shows the stable sustained presence of sucralose in wastewater effluents for a suite of samples collected throughout the state of Florida, regardless of the facility's nutrient removal capabilities. Data in Fig. 1 are independent of the data presented in Table 3. Although this Fig. 1 does not include membrane bioreactor (MBR) facilities, a pilot study reported to the New Mexico Environment Department showed higher average sucralose levels of 42,400 ng/L in MBR effluent (Lee et al., 2010). This report also showed sucralose removal of approximately 40% for ozone and a biologically active filter and approximately 99% for reverse osmosis with trace levels detectable in the RO effluent indicating that sucralose is probably not appropriate as an indicator of wastewater that has gone through advanced treatment processes.

3.2. Comparison of sucralose with carbamazepine

Sucralose and carbamazepine appear to be two of the most efficacious indicators of municipal wastewater input to surface water supplies, but adequate analytical sensitivity is an important issue. Comparison of Figs. 3 and 4 demonstrates the greater spread in the sucralose data between the wastewater effluents

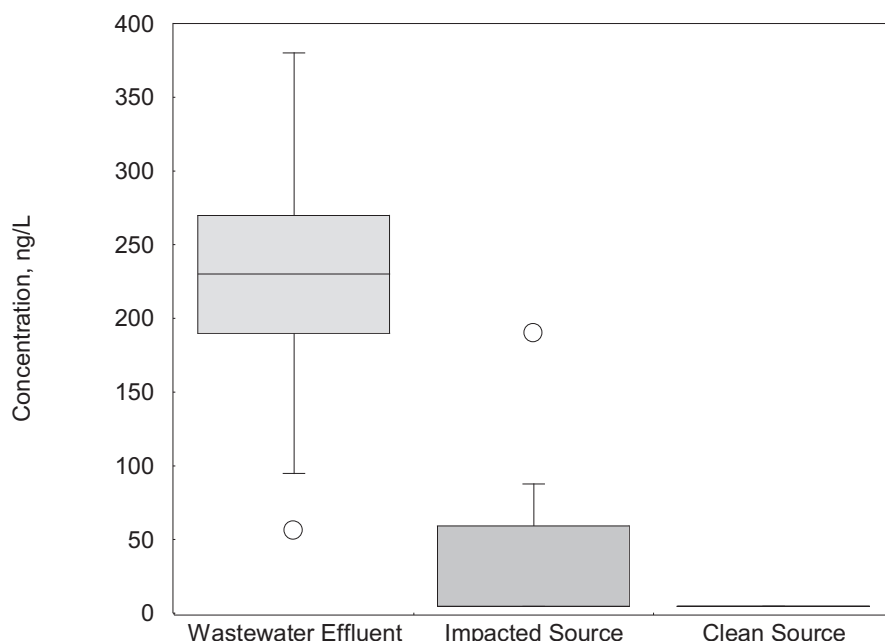


Fig. 4 – Nonparametric probability distribution of carbamazepine concentrations in different matrices depicted as box and whisker plots. Top and bottom of each box = 75th and 25th percentiles, respectively; top and bottom of each whisker = 90th and 10th percentile respectively; line across inside of box = median (50th percentile); and points beyond whiskers = outliers. Clean source samples were all below the method reporting limit (MRL) and represented as a line at the 5 ng/L MRL. For the impacted sources, 64% of the samples were left censored below the MRL and therefore only the 75th and 90th percentile and maximum values appear on the plot.

Table 4 – Comparison of sucralose and carbamazepine concentrations in single grab samples collected from eight septic systems located in two separate counties in Florida.

	Sucralose (ng/L)	Carbamazepine (ng/L)
septic 1	69,000	<5
septic 2	40,000	40
septic 3	80,000	<5
septic 4	42,000	<5
septic 5	24,000	55
septic 6	40,000	<5
septic 7	12,000	<5
septic 8	12,000	<5

and the impacted sources compared with the carbamazepine data. Sucralose therefore has greater ability to assess levels of dilution in an impacted source for sucralose and carbamazepine reporting limits of 100 ng/L and 5 ng/L respectively. Due to the differences in wastewater effluent concentrations, the performance of carbamazepine as a unique marker of municipal wastewater should be equivalent to sucralose only when the MRL of carbamazepine is less than 1 ng/L. Many laboratories, however, are not capable of achieving an MRL in the vicinity of 1 ng/L or lower for carbamazepine (Vanderford et al., 2010).

Sucralose is also a superior indicator to carbamazepine for wastewater input originating from septic system intrusion as demonstrated by the data presented in Table 4 for eight septic samples collected in Florida.

4. Conclusions

These findings suggest the superior performance of the artificial sweetener sucralose to serve as an indicator for the presence of conventional biologically treated municipal and domestic wastewater and septic system sources to water bodies in the United States. The consistently high concentrations present in municipal wastewater effluents and septic systems along with a demonstrated absence of false negatives in impaired water sources and absence of false positives in clean sources supports its usefulness as an appropriate indicator compound for contaminant inputs of human origin that have not undergone advanced treatment through processes such as reverse osmosis. Additional studies on the fate of sucralose during environmental transport need to be conducted in order to assess whether the concentration of sucralose in the receiving water can also be used to quantitatively assess the fractional contribution of wastewater input.

Acknowledgments

The authors gratefully acknowledge the WaterReuse Research Foundation's financial, technical, and administrative assistance in funding and managing the project through which a portion of this information was discovered. We are particularly indebted to Caroline Sherony in her role as the Foundation Project Officer. The comments and views detailed herein may not necessarily reflect the views of the WaterReuse Research Foundation, its officers, directors, employees,

affiliates or agents. Additional support was also received from the Florida Department of Environmental Protection, the Southwest Florida Water Management District, the South Florida Water Management District, the St Johns River Water Management District, the Orange County Utilities Department, the City of Orlando, Hillsborough County, the City of North Port and in-kind support from Miami-Dade Water and Sewer Department, JEA, and the City of Pompano Beach.

REFERENCES

- Benotti, M.J., Trenholm, R.A., Vanderford, B.J., Holady, J.C., Stanford, B.D., Snyder, S.A., 2009. Pharmaceuticals and endocrine disrupting compounds in U.S. drinking water. *Environ. Sci. Technol.* 43 (3), 597–603.
- Bester, K., 2007. *Personal Care Compounds in the Environment. Pathways, Fate, and Methods for Determination*. Wiley-VCH Verlag GmbH & Co., Weinheim, Federal Republic of Germany.
- Brorström-Lundén, E., Svenson, A., Viktor, T., Woldegiorgis, A., Remberger, M., Kaj, L., Dyem, C., Bjerke, A., Schlabach, M., 2008. Measurements of Sucralose in the Swedish Screening Program. Swedish Environmental Institute (IVL). Report B1769, January.
- Buerge, I.J., Kahle, M., Buser, H.-R., Müller, M.K., Piger, T., 2008. Nicotine derivatives in wastewater and surface waters: application as chemical markers for domestic wastewater. *Environ. Sci. Technol.* 42, 6354–6360.
- Buerge, I.J., Buser, H.-R., Kahle, M., Müller, M.D., Poiger, T., 2009. Ubiquitous occurrence of the artificial sweetener acesulfame in the aquatic environment: an ideal chemical marker of domestic wastewater in groundwater. *Environ. Sci. Technol.* 43 (12), 4381–4385.
- Burow, K.R., Nolan, B.T., Rupert, M.G., Dubrovsky, N.M., 2010. Nitrate in groundwater of the United States, 1991–2003. *Environ. Sci. Technol.* 44 (13), 4988–4997.
- Dickenson, E.R.V., Snyder, S.A., Sedlak, D.L., Drewes, J.E., 2010. Indicator compounds for assessment of wastewater effluent contributions to flow and water quality. *Wat. Res.* 45, 1199–1212.
- Focazio, M.J., Kolpin, D.W., Barnes, K.K., Furlong, E.T., Meyer, M.T., Zaugg, S.D., Barber, L.B., Thurman, M.E., 2008. A national reconnaissance for pharmaceuticals and other organic wastewater contaminants in the United States – (II) Untreated drinking water sources. *Sci. Total Environ.* 402, 201–216.
- Fry, B., 2006. *Stable Isotope Ecology*. Springer Science+Business Media, LLC, New York, N.Y.
- Gasser, G., Rona, M., Voloshenko, A., Shelkov, R., Tal, N., Pakrativ, I., Elhanany, S., Lev, O., 2010. Quantitative evaluation of tracers for quantification of wastewater contamination of potable water sources. *Environ. Sci. Technol.* 44, 3919–3925.
- Glassmeyer, S.T., Furlong, E.T., Kolpin, D.W., Cahill, J.D., Zaugg, S.D., Werner, S.L., Meyer, M.T., Kryak, D.D., 2005. Transport of chemical and microbial compounds from known wastewater discharges: potential for use as indicators of human fecal contamination. *Environ. Sci. Technol.* 39, 5157–5169.
- Guo, Y.C., Krasner, S.W., 2009. Occurrence of primidone, carbamazepine, caffeine, and precursors for n-nitrosodimethylamine in drinking water sources impacted by wastewater. *JAWRA* 45 (1), 58–67.
- Jjemba, P.K., 2008. *Pharma-ecology, the Occurrence and Fate of Pharmaceuticals and Personal Care Products in the Environment*. John Wiley & Sons, Inc., Hoboken, N.J.
- Katz, B.G., Chelette, A.R., Pratt, T.R., 2004. Use of chemical and isotopic tracers to assess nitrate contamination and ground-water age, Woodville Karst Plain, USA. *Journal of Hydrol.* 289, 36–61.
- Kumar, A., Xagoraki, I., 2010. Pharmaceuticals, personal care products and endocrine-disrupting chemicals in U.S. surface and finished waters: a proposed ranking system. *Sci. Total Environ.* 408, 5972–5989.
- Lee, C.O., Howe, K.J., Thomson, B.M., 2010. Ozone and Biofiltration as an Alternative to Reverse Osmosis for Removing PPCPs and EDCs from Wastewater. Report to New Mexico Environment Department. <http://www.unm.edu/~howe/UNM%20Howe%20Final%20PPCP%20Ozone-Biofiltration%20Report.pdf>.
- Loos, R., Gawlik, B.M., Boettcher, K., Locoro, G., Contini, S., Bidoglio, G., 2009. Sucralose screening in European surface waters using a solid-phase extraction-liquid chromatography-triple quadrupole mass spectrometry method. *J. Chromatogr.* 1216, 1126–1131.
- Loraine, G.A., Pettigrove, M.E., 2006. Seasonal variations in concentrations of pharmaceuticals and personal care products in drinking water and reclaimed wastewater in southern California. *Environ. Sci. Technol.* 40, 687–695.
- Neset, T.-S.S., Singer, H., Longree, P., Bader, H.-P., Scheidegger, R., Wittmer, A., Andersson, J.C.M., 2010. Understanding consumption-related sucralose emissions – A conceptual approach combining substance-flow analysis with sampling analysis. *Sci. Total Environ.* 408, 3261–3269.
- Nix, Stephen J., 1994. *Urban Stormwater Modeling and Simulation*. CRC Press, Inc., Boca Raton, FL.
- Paul, M.J., Meyer, J.L., 2001. Streams in the urban landscape. *Annu. Rev. Ecol. Syst.* 32, 333–365.
- Scheurer, M., Brauch, H.-J., Lange, F.T., 2009. Analysis and occurrence of seven artificial sweeteners in German waste water and surface water and in soil aquifer treatment (SAT). *Anal. Bioanal. Chem.* 394, 1585–1594.
- Scheurer, M., Storck, F.R., Brauch, J.-J., Lange, F.T., 2010. Performance of conventional multi-barrier drinking water treatment plants for the removal of four artificial sweeteners. *Wat. Res.* 44, 3573–3584.
- Scheurer, M., Rüdiger Storck, F., Graf, C., Brauch, J.-J., Ruck, W., Lev, O., Lange, F.T., 2011. Correlation of six anthropogenic markers in wastewater, surface water, bank filtrate, and soil aquifer treatment. *J. Environ. Monit.* 13, 966–973.
- Sedlak, D.L., Pinkston, K.E., 2001. Factors affecting the concentrations of pharmaceuticals released to the aquatic environment. *Wat. Res. Update* 120, 56–64.
- Stackelberg, P.E., Furlong, E.T., Meyer, M.T., Zaugg, S.D., Henderson, A.K., Reissman, D.B., 2004. Persistence of pharmaceutical compounds and other organic wastewater contaminants in a conventional drinking-water-treatment plant. *Sci. Total Environ.* 32, 99–113.
- Standley, L.J., Rudel, R.A., Swartz, C.H., Attfield, K.R., Christian, J., Erickson, M., Brody, J.G., 2008. Wastewater-contaminated groundwater as a source of endogenous hormones and pharmaceuticals to surface water ecosystems. *Environ. Toxicol. Chem.* 27 (12), 2457–2468.
- Torres, C.I., Ramakrishna, S., Chiu, C.-A., Nelson, K.G., Westerhoff, P., Krajmalnik-Brown, R., 2011. Fate of sucralose during wastewater treatment. *Environ. Eng. Sci.* 28 (5), 325–331.
- USEPA, January 2009. National Water Quality Inventory: Report to Congress, 2004 Reporting Cycle: Findings. www.epa.gov/owow/305b/2004report/factsheet2004305b.pdf.
- USEPA, August 2010. Treating Contaminants of Emerging Concern. In: A Literature Rev. Database. www.epa.gov/waterscience/ppcp/studies/results.html.
- Vanderford, B., Snyder, S., Eaton, A., Guo, C., Ternes, T., Drewes, J., Wood, C., 2010. Fourth Periodic report for evaluation of analytical methods for EDCs and PPCPs via Interlaboratory comparison. WaterRF Project 4167 April 16, 2010.
- Yamamoto, H., Nakamura, Y., Moriguchi, S., Nakamura, Y., Honda, Y., Tamura, I., Hirata, Y., Hayashi, A., Sekizawa, J., 2009. Persistence and partitioning of eight selected pharmaceuticals in the aquatic environment. Lab. photolysis, biodegradation, Sorption Experiments *Wat. Res.* 43, 351–362.