



Differential presence of anthropogenic compounds dissolved in the marine waters of Puget Sound, WA and Barkley Sound, BC

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ABSTRACT

Organic compounds were evaluated in March 2010 at 22 stations in Barkley Sound, Vancouver Island Canada and at 66 locations in Puget Sound. Of 37 compounds, 15 were xenobiotics, 8 were determined to have an anthropogenic imprint over natural sources, and 13 were presumed to be of natural or mixed origin. The three most frequently *detected* compounds were salicylic acid, vanillin and thymol. The three most *abundant* compounds were diethylhexyl phthalate (DEHP), ethyl vanillin and benzaldehyde (~600 ng L⁻¹ on average). Concentrations of xenobiotics were 10–100 times higher in Puget Sound relative to Barkley Sound. Three compound couplets are used to illustrate the influence of human activity on marine waters; vanillin and ethyl vanillin, salicylic acid and acetylsalicylic acid, and cinnamaldehyde and cinnamic acid. Ratios indicate that anthropogenic activities are the predominant source of these chemicals in Puget Sound.

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1. Introduction

Fjords on the west coast of North America are home to diverse marine ecological communities, but also are proximal to large and growing urban populations (Emmett et al., 2000; Fraser et al., 2006). Protecting the ecosystems of Puget Sound and the fjords of Vancouver Island requires understanding the myriad of natural and anthropogenic forces acting upon these waterways. Efforts to reverse declining populations of fauna and flora, mitigate threats and stressors to marine resources, and improve ecosystem health and management for long term sustainability are complex, but all require knowledge of the sources and fates of environmental pollutants entering marine systems due to the actions of man. Rapid population growth coupled with urban sprawl can have negative effects on the health of the region's environment (Fraser et al., 2006).

Marine organisms in the Pacific Northwest, particularly those in Puget Sound, already show signs of environmental stress. A large body of evidence links organic chemical exposure to a range of deleterious biological effects in marine mammals (e.g. endocrine system disruption). Salmon in Puget Sound's urban waterways have high biomarker indicators of stress, most likely due to urbanized pollution (Browne et al., 2010). Organic pollutants such as PCBs and DDTs are commonly among the stressors cited as potential causes for the recent Southern Resident Killer Whale population decline (Krahn et al., 2007). Andrews et al. (2007) hypothesized

that the six gill shark population of Puget Sound is susceptible to pollution effects because of their small migration patterns and propensity to live in waters near highly urbanized areas. Evidence such as this led Emmett et al. (2000) to suggest that the greatest threat to the coastal ocean's resources is the urbanization of the watershed (Brandenberger et al., 2008; Kuo et al., 2011b).

Increases in human populations within a watershed lead to increased release of wastewaters from industry, agriculture and households. Among the released compounds are natural and xenobiotic contaminants (chemicals which did not exist in nature before their introduction by humans) such as cooking spices, pharmaceuticals, industrial solvents, and pesticides. Many chemicals have never been evaluated in the marine environment, preventing discourse on trends and potential preventative or restorative actions. This is especially true for chemicals found in common household products (e.g. solvents, perfuming agents), which are known to have aggregated effects on humans when present as mixtures, even in exceedingly low concentrations (Caress and Steinemann, 2009; Steinemann, 2004, 2009). Their presence in aquatic environments might thus signal potential issues.

Puget Sound, Washington is a highly urbanized watershed with more than three million persons in residence. Conversely, Barkley Sound, British Columbia, has less human influence in the watershed and a lower population density. The goal of this study was to determine abundances in marine waters of a suite of chemicals commonly found in homes. These chemicals range from xenobiotics through natural compounds also synthesized for industrial purposes to natural compounds used in cooking and food preparation. Samples were collected in March 2010 and comparisons were

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made between a relatively unaltered fjord, Barkley Sound, and a highly urbanized waterway, Puget Sound.

2. Experimental

2.1. Sampling

In Barkley Sound, water samples were collected using a Niskin bottle at 11 stations aboard the *R/V Barkley Star* between March 23–27, 2010, and brought to the Bamfield Marine Sciences Center for initial workup. The cruise track included stations in Effingham Inlet, Trevor Channel, Alberni Inlet, Imperial Eagle Inlet, San Mateo

Bay, and Sarita Bay (Fig. 1). Seawater was drawn into acid-leached 4 L LDPE cubitainers and stored aboard ship until filtration commenced that evening.

In Puget Sound, samples were collected into acid-leached 1 L LDPE cubitainers by volunteers in the SoundCitizen program (<http://soundcitizen.org>). Most samples reported here were collected in the intertidal zone at beaches. Sample meta data was logged by volunteers into an on-line database and then physical samples were mailed to the University of Washington Aquatic Organic Geochemistry laboratory where they were immediately processed. An average of four days transpired between volunteer sampling and sample workup.

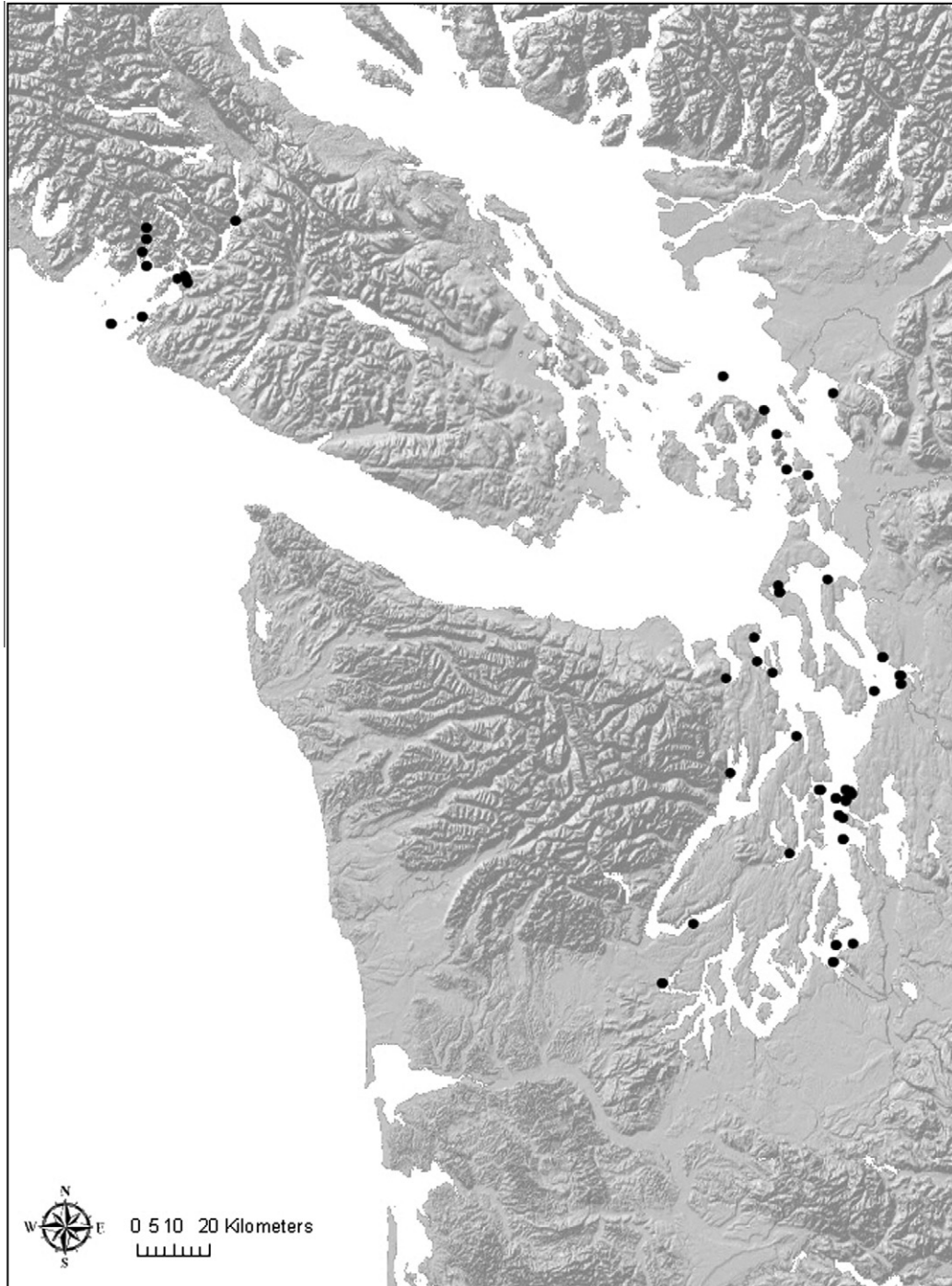


Fig. 1. Sample locations in Barkley Sound (British Columbia, Canada) and Puget Sound (Washington State, USA).

Table 1
Chemicals evaluated in this study. Column D ("designation") marks whether the chemical is a xenobiotic (1), can be of either natural or anthropogenic origin but is suspected to be of anthropogenic source (2; see Section 4) or is of unknown source (3; either natural or anthropogenic).

	CAS ID	MW (g mol ⁻¹)	D	Target ion	Qualifier ions	
					Q1	Q2
Acetylsalicylic acid	50-78-2	180.16	1	195	210	120
Benzaldehyde ^b (almond)	100-52-7	106.12	2	105	106	77
Benzyl acetate ^b (jasmine)	140-11-4	150.18	3	150	108	91
Benzyl propionate ^{b,d} (banana)	122-63-4	164.20	3	164	108	91
Carvacrol (oregano)	499-75-2	150.22	3	222	207	
β-Estradiol	50-28-2	272.38	3	416	285	232
Bisphenol A	80-05-7	228.29	1	357	372	
S(+)-Carvone ^b (caraway)	99-49-0	150.22	3	82	150	108
Caffeine	58-08-2	194.19	3	194	109	67
Chlordane	57-74-9	409.78	1	372	236	271
Cinnamaldehyde ^{b,d} (cinnamon)	104-55-2	132.16	3	132	131	103
trans-Cinnamic acid (cinnamon)	140-10-3	148.17	2	205	220	161
Coprostanol	360-68-9	388.68	3	370	355	215
Diazinon	333-41-5	304.35	1	137	179	304
Dibutyl phthalate (DBP)	84-74-2	278.34	1	149	205	223
Dicyclohexyl phthalate (DCHP)	84-61-7	330.46	1	149	167	249
Diethylhexyl phthalate (DEHP)	117-81-7	390.56	1	149	167	279
Diethylstilbestrol	56-53-1	268.35	1	412	397	383
3,4-Dihydroxybenzoic acid ^d	99-50-3	154.12	-	193	370	355
Ethyl vanillin (artificial vanilla)	121-32-4	166.17	1	167	195	238
17α-Ethynylestradiol	57-63-6	296.40	1	342	257	218
Eugenol (clove/nutmeg)	97-53-01	164.20	3	206	221	236
Ibuprofen	15687-27-1	206.28	1	160	117	263
Lilial	80-54-6	204.31	1	189	204	147
Limonene ^b (oil of lemon)	5898-27-5	136.24	2	68	93	136
Limonene oxide cis ^b (citrus)	470-82-6	152.23	2	137	109	
Limonene oxide trans ^b (citrus)	470-82-6	152.23	2	108	137	
Linalool ^c (coriander, lavender)	78-70-6	154.25	3	93	71	121
Mestranol	72-33-3	310.43	1	227	310	242
Musk ketone	81-41-1	294.31	1	279	294	
Musk Moskene	116-66-5	280.32	1	263	278	
Musk xylene	81-15-2	297.30	1	282	297	
Progesterone	57-83-0	314.46	3	314	272	
Thymol (thyme)	89-83-8	150.22	2	222	207	
Vanillin (vanilla)	121-33-5	152.15	2	194	209	224
Veratraldehyde ^b (caramelized vanilla)	120-14-9	166.17	3	166	95	
Zingerone	122-48-5	194.22	3	137	179	304

^a Internal standard.

^b Does not derivatize with BSTFA.

^c Derivatizes in pyridine but not in ethyl acetate.

^d Co-elute on DB-1 column but not on DB-5.

2.2. Solid Phase Extraction

Water samples were filtered using 47 mm glass-fiber filter (Whatman GF/F) into an acid-washed glass container, returned to their original cubitainer which was first rinsed with filtered sample water, and then acidified to a pH < 4 using HPLC-grade HCl. Blanks (di water) were processed simultaneously, including sitting in the LDPE bottles for appropriate periods of time prior to chemical extraction. A recovery standard, 3,4-dihydroxybenzoic acid was added at this point. A known volume (0.6–2.4 L) was then extracted onto a Waters Oasis HLB column as described by Keil and Neibauer (2009). Puget Sound volumes averaged 800 ml while volumes of Barkley Sound water extracted averaged 2000 ml. Extracted samples were eluted from the HLB column using ethyl acetate, brought to near-dryness, resuspended to a known volume (typically 0.5 ml) and stored at –20 °C until analysis by gas chromatography–mass spectrometer (GC–MS). Samples were prepared for gas chromatographic analysis by combining 50 µL ethyl acetate extract with 50 µL BSTFA:TMCS (99:1) in autosampler GC vials sealed with Teflon-lined caps. Samples were heated on a dry-block hot plate at 60 °C for 10 min to silylate any exchangeable hydrogens present in the extract. Some compounds do not derivatize with BSTFA in ethyl acetate and were analyzed underivatized (Table 1). GC–MS analysis was performed on an Agilent 6890N GC coupled to 5975 MSD fitted with a J&W DB-5MS column. Sam-

ples were injected in splitless mode and analyzed in both full-scan (60–380 amu) and SIM mode (ions for each compound as in Table 1). Peaks were identified by a combination of selected ions and quantified from ion-specific response factors determined from 3-point standard curves of the stock standard. All quantities were then corrected for recoveries relative to the recovery standard 3,4-dihydroxybenzoic acid. Analysis blanks were subtracted from all data reported here.

The Shapiro–Wilk's Test for Normality was used to determine whether the measured concentration ranges originated from a normally distributed population. Because the data failed ($p < 0.01$), we log-transformed the concentrations prior to additional statistical tests. A two-tailed Student's *t*-test was used to determine whether concentrations detected in Puget Sound were different than those detected in Barkley Sound. Ratios were similarly transformed and evaluated. Nondetects were not included in these analyses.

3. Results

3.1. Frequency of detection

Each of the 37 compounds was detected at least once in Puget Sound, but only 28 were detected in Barkley Sound (Fig. 2). On average, chemicals in the suite were identified ~25% of the time

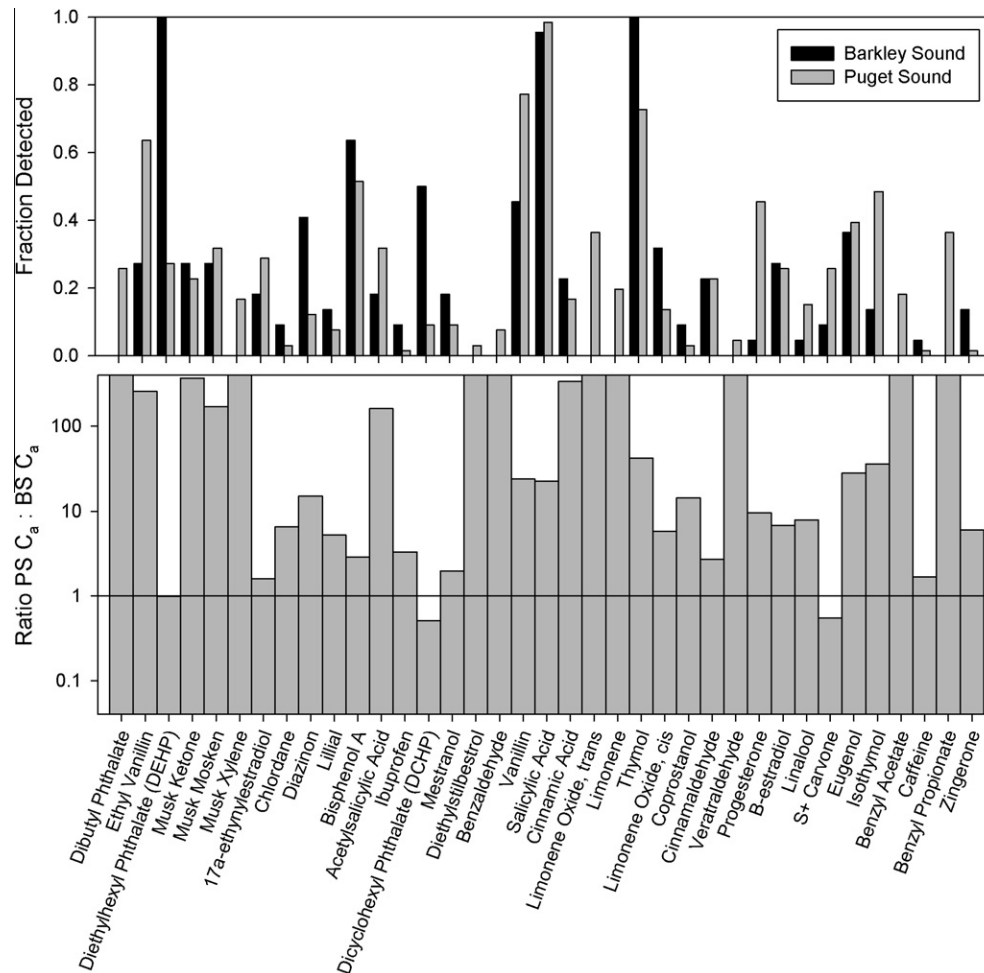


Fig. 2. Top panel: the fraction of times each chemical was detected in for Barkley Sound (dark bar) and Puget Sound (light bar). Lower panel: ratio of average concentrations for detected samples in Puget Sound relative to Barkley Sound. Samples with a full bar indicate that the chemical was not detected in Barkley Sound.

in either location and there was no statistically significant trend between detection and abundance. Because roughly 2.5 times more water was extracted in Barkley Sound, detection limits were somewhat lower in that system. This may account for the similarity in detection of xenobiotics between the urbanized system (Puget Sound) and the more pristine system (Barkley Sound). Given this, the ratio of detection frequency between fjords provides a general tool for determining in which environment certain chemicals are more prevalent. Only two compounds were observed in Barkley Sound more frequently than in Puget Sound: S(+)-Carvone ('caraway smell', a common essential oil in many plants) and dicyclohexyl phthalate (DHP; a common plasticizer in marine glues and soft plastics). All other chemicals were more frequently detected in Puget Sound than in Barkley Sound (Fig. 2).

3.2. Average concentrations

Average concentrations varied widely between compounds and between Barkley and Puget Sound (Fig. 3). Two compounds were detected at high concentrations in Puget Sound but not at all in Barkley Sound; dibutyl phthalate (a plasticizer) and benzaldehyde (a common industrial solvent). Overall, the most abundant compounds in Puget Sound were dibutyl phthalate and benzaldehyde with average concentrations of $\sim 700 \text{ ng L}^{-1}$, and ethyl vanillin with average concentrations of $\sim 1000 \text{ ng L}^{-1}$. Other abundant compounds in both fjords were salicylic acid, vanillin, and the plasticizer DEHP (Fig. 3). In Puget Sound their concentrations averaged

$\sim 300 \text{ ng L}^{-1}$ while in Barkley Sound they were observed at $\sim 15 \text{ ng L}^{-1}$ on average (Fig. 3). The observation that both locations had similar concentrations ($\sim 250 \text{ ng L}^{-1}$) of the plasticizer DEHP leads us to suspect that the data reflect contamination in our sampling procedure, but the values are reported as they are significantly above our procedural blank (which includes field blanks for the Puget Sound samples only).

4. Discussion

The human population density in the Puget Sound region is approximately $6\times$ higher than that of Barkley Sound (121 and 18 humans km^{-2} , respectively). Thus, it is perhaps expected that anthropogenic compounds are detected at higher average concentrations in the Puget Sound region. None-the-less, there are subtle signs illustrating a human impact in the relatively pristine Barkley Sound. The plasticizers diethylhexyl phthalate (DEHP) and dicyclohexyl phthalate (DCHP) were routinely ($>90\%$ of the time) detected in Barkley Sound. 17α -Ethinylestradiol, a common pharmaceutical in birth control pills, and bisphenol-A, a common plasticizer, were observed in Barkley Sound in multiple samples. This adds to the growing data indicating pollutant loads even in the most pristine marine settings.

While the observance of a xenobiotic compound is a clear indication of anthropogenic influence, it is often hard to discriminate between human and natural sources for chemicals that are derived from both. One compound like this is salicylic acid, which is

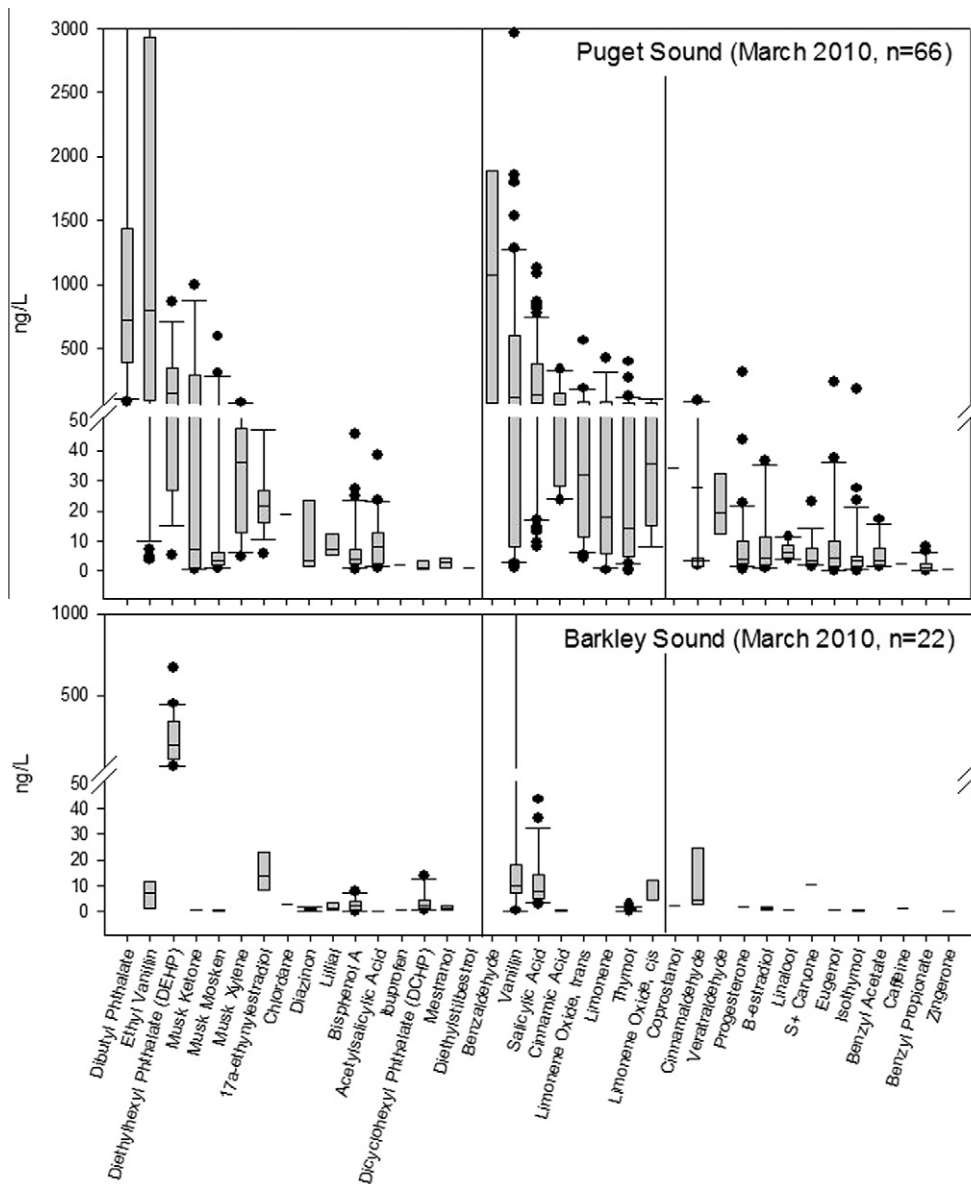


Fig. 3. Box and whisker plot of concentrations for all chemicals analyzed in Puget Sound (upper panel) and Barkley Sound (lower panel) in March 2010. Black vertical lines between diethylstilbestrol and benzaldehyde separates xenobiotics on the left from compounds with mixed sources. Black vertical line between limonene oxide (cis) and coprostanol separates compounds where anthropogenic sources are suspected to imprint over natural sources (on the left) from chemicals where there is no statistical difference between locations (Barkley and Puget Sounds) and thus the compounds are presumed to be from natural sources.

produced naturally by vascular plants (Sarkanen and Ludwig, 1971) and macroalgae (Novoa et al., 2011) is also produced industrially for various uses (acne creams), and is the first breakdown product of acetylsalicylic acid (aspirin) in the human blood stream (Bae et al., 2008). Salicylic acid is removed from sewage at high (99%) efficiency yet is often one of the most abundant non-steroidal anti-inflammatory drugs found in treated wastewater (Huang et al., 2011). It is also commonly found with other pain relievers in treated sewage effluent entering marine waters (Comeau et al., 2008). Considering that aspirin is the single most abundantly consumed pharmaceutical on earth (Jeffreys, 2004), it is reasonable to conclude that the salicylic acid commonly observed in the marine environment has a human source (Comeau et al., 2008; Huang et al., 2011; Wille et al., 2010). However, this has not been thoroughly explored and indeed may not necessarily be true.

Without resorting to additional analyses (isotopic analyses, additional compound or breakdown products), one simple way to obtain a relative measure of the influence of human activities on

the environmental composition of these two chemicals (salicylic and acetylsalicylic acids) is to calculate their ratio. In the case of salicylic and acetylsalicylic acid, this SA:ASA ratio is ~30 for human urine. Bae et al. (2008) showed that within half an hour of consumption of aspirin, the human body converts most acetylsalicylic acid to salicylic acid. SA:ASA values around 30 therefore potentially denote a system dominated by anthropogenic sources of the chemicals. Conversely, since only salicylic acid is found naturally, the ratio in a pristine system ought to be infinite. This simple ratio has no scaling function and ignores several potentially important factors such as additional breakdown of salicylic acid in the human body, differential degradation of the chemicals in the environment, etc., but it may serve as a gross first indication of a system's anthropogenic state.

Ratios can only be calculated for samples where both compounds are detected. In the case where only salicylic acid is detected, the 'infinite' ratio strongly suggests a natural source, but cannot provide proof thereof. In Barkley Sound, salicylic acid

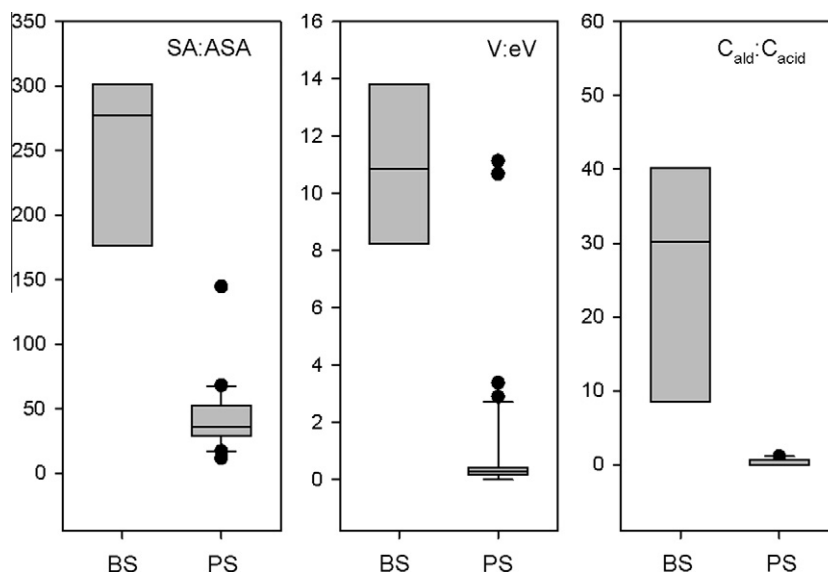


Fig. 4. Ratios of natural to anthropogenic indicators for three chemical pairs. SA:ASA is the ratio of salicylic acid to acetylsalicylic acid. V:eV is the ratio of vanillin to ethyl vanillin and $C_{ald}:C_{acid}$ is the ratio of trans-cinnamaldehyde to cinnamic acid. In each case, lower ratios indicate a more perturbed and higher ratios indicate a more natural system.

was detected in 21 of 22 samples, yet acetylsalicylic acid was co-detected only four times (Fig. 2). The SA:ASA ratio in these four cases ranged between 143 and 310. Thus, Barkley Sound seems to have a predominately natural signature for salicylic acid. In Puget Sound, salicylic acid was detected in 65 of 66 samples and acetylsalicylic acid was detected in 21 samples. The average SA:ASA ratio in Puget Sound was 42 ± 28 , much lower than observed in Barkley Sound (Fig. 4) and suggestive of a human source for these chemicals. This is further suggested by the large and statistically different (Student's *t*-test) difference in average concentration of salicylic acid in Puget Sound ($260 \pm 79 \text{ ng L}^{-1}$) versus Barkley Sound ($12 \pm 10 \text{ ng L}^{-1}$).

If these two regions reflect natural (Barkley Sound) and altered (Puget Sound) environments, respectively, they provide context to the recent findings of Comeau et al. (2008) who measured low ng L^{-1} concentrations of salicylic acid in two marine bays in NE Canada, and for Huang et al. (2011) who measured much higher values in the highly urbanized Pearl River Delta in South China. The Canadian values fall at the low end of our Puget Sound estimates, and are slightly higher than observed in Barkley Sound. Taken at face value, this suggests that their values contain a substantial component of natural salicylic acid. Alternatively, the values reported in the Pearl River delta are 2–200 \times higher than observed in Puget Sound, suggesting a greater degree of anthropogenic impact in that system. Neither of these other studies included measurement of acetylsalicylic acid, so ratios cannot be calculated for these studies.

Another example of a compound with natural and anthropogenic sources is vanillin (V). Vanillin is produced by all vascular plants and is a part of the macromolecular lignin structure (Hedges and Mann, 1979). It can be released to water via microbial degradation of lignin and also by biomass burning, which produces free vanillin as a byproduct (Kuo et al., 2011a). It is also produced by many plants as a free monomer (e.g. vanilla beans and other orchid pods contain high levels of free vanillin), and is the primary flavoring agent in vanilla. Demand for vanilla as a flavoring agent exceeds natural production (Dignum et al., 2001), so copious amounts of vanillin are produced industrially, usually from either oil or wood pulp starting materials. Along with vanillin, the xenobiotic compound ethyl vanillin (eV) is also produced synthetically,

largely because it has a 'stronger' flavor than vanilla. Typical artificial vanilla has a ratio of $\sim 1\text{--}10:1$ vanillin:ethyl vanillin (Ali et al., 2008). Unlike the salicylic–acetylsalicylic acid couple, where salicylic acid is produced both naturally and via breakdown of the xenobiotic, ethyl vanillin is not known to breakdown to vanillin during metabolism.

Similarly to the SA:ASA ratio, V:eV ratios are different between Barkley and Puget Sound (Fig. 4), with Barkley Sound having a more 'natural' composition. Ethyl vanillin was detected in Barkley Sound 27% of the samples, but was observed in 64% of the Puget Sound samples. Concentrations of eV in Puget Sound were up to three orders of magnitude higher than that observed in Barkley Sound (average concentrations of $1835 \pm 718 \text{ ng L}^{-1}$ and 7 ± 6 , respectively; Fig. 3), suggesting a strong anthropogenic signal in Puget Sound. In fact, ethyl vanillin is often one of the most abundant chemicals measured in our suite (Keil and Neibauer, 2009). Despite natural sources of vanillin, ethyl vanillin was more abundant than vanillin in Puget Sound (ratio < 1) in 32 of the 42 times they were co-detected. This intriguing observation does not make "simple" sense in light of the fact that even artificial vanilla usually contains more vanillin than ethyl vanillin (Avila et al., 2009; de Jager et al., 2007), meaning that the V:eV ratio ought always be greater than 1. The two simplest ways to explain low ratios is that eV is degraded less efficiently during sewage treatment and is thus relatively enriched upon delivery to marine waters, or it is degraded within marine waters slower than vanillin and thus its concentration tends to stabilize higher than that of vanillin. Neither case has been explored.

Finally, cinnamic acid and cinnamaldehyde can be coupled in a similar manner. In this case, the coupling is more tenuous because both are natural chemicals with multiple sources, and both are used extensively in foods (cinnamaldehyde = cinnamon) or perfumes (cinnamic acid is a precursor to many other odorants). Demand for these compounds is high enough that they are produced synthetically, but there have been no previous efforts to determine provenances for these chemicals in the environment. In nature, cinnamaldehyde is produced in far excess over cinnamic acid in most vascular plants (Sarkanen and Ludwig, 1971) but bacteria can synthesize cinnamic acid (Moore et al., 2002). Humans consume 50 million pounds of cinnamon per year in the United

States (USDA), and cinnamon is the third most valued (by economic dollars) spice imported to the US each year (after vanilla and pepper) (Buzzanell et al., 1995). After consumption by mammals, a large proportion of the cinnamaldehyde consumed is converted to cinnamic acid (Michiels et al., 2008). Thus if human consumption is the primary driver controlling the concentrations of these chemicals in the environment, cinnamic acid ought to dominate. Conversely, if natural production dominates, then cinnamaldehyde ought to be relatively more abundant.

Applying the ratio approach to cinnamaldehyde and cinnamic acid again provides evidence for a predominantly human fingerprint in Puget Sound relative to Barkley Sound. $C_{ald}:C_{acid}$ ratios in Barkley Sound are between 10 and 40, whereas in Puget Sound they are all equal to or below 1 (Fig. 4). Thus, we conclude that the majority of cinnamic acid found in Puget Sound is of anthropogenic origin.

Fifteen of the chemicals monitored are xenobiotics and thus of clear anthropogenic origin. Of the remaining 22, all of which have natural sources, we identify several as likely having a human-influenced distribution in Puget Sound. To make this inference, it is assumed that Barkley Sound represents a pristine system (even though this assumption is flawed). Average concentrations for each chemical in each sound were statistically compared (nondetects were treated as no data). Compounds with statistically higher concentrations in Puget Sound and higher frequencies of detection include vanillin, salicylic acid, cinnamic acid, thymol, limonene, limonene oxide (cis), and limonene oxide (trans). Benzaldehyde, because it was not detected in Barkley Sound, was not subjected to a statistical test but appears to be of anthropogenic source. It is found naturally (almond odor), but is an extremely common industrial solvent used as a precursor for many perfumes and cleaning agents. Thymol is a natural antioxidant found in the essential oil of many plants. It is used industrially as a safe pesticide and is found in many mouthwashes (Hu and Coats, 2008). Limonene and its oxides are common in many household products for their use as odorants (lemon, orange, pine) and for their utility as cleaning agents. These compounds are identified in Figs. 2 and 3 as being of mixed sources but having a high likelihood of being enriched in Puget Sound due to human activities.

The source (natural or anthropogenic) of the remaining compounds is not discernable with the data at hand. This list includes caffeine and veratraldehyde, compounds for which an anthropogenic source is often assumed. This is because they are used extensively in foods or drinks. Caffeine is produced by many vascular plants (Peeler et al., 2006; Power and Chesnut, 1919) and is a commonly ingested stimulant in tea and coffee (Heckman et al., 2010). Despite natural sources, it has previously been used as an indicator of human influence on marine waters (Peeler et al., 2006). We do not yet have enough data to determine the source of veratraldehyde, a compound produced by some plants (Choi et al., 2009; Cuong et al., 2001) but used extensively in such foods as waffles and kettle corn. We detect this chemical infrequently, but always at higher concentrations in Puget Sound relative to Barkley Sound.

The sterols β -estradiol and progesterone and the cholesterol hydrogenation product coprostanol are often thought to be of human origin. However, they are also produced by marine mammals, complicating source assignment. Carvone (caraway smell), eugenol (clove smell) and isothymol (also called carvacrol; smells like oregano) are all present in native plants of the Pacific Northwest (SoundCitizen – data not shown) but are also used extensively in foods. Similarly, benzyl acetate (jasmine-like smell) and benzyl propionate (pear or apple odor) are also used by the perfume industry, but are largely thought to be derived from natural products. Zingerone (gingerol breakdown product when cooking foods), is a potential marker for ginger consumption, but was rarely measured in these samples.

Whether or not these chemicals have a significant anthropogenic source in Puget Sound will require additional data. The measurement of stable carbon isotopes (^{13}C and ^{14}C) has great potential in this regard. Typical C3 plants in the Pacific Northwest have a ^{13}C composition of $\sim 27\text{‰}$ while many industrial chemicals created from petroleum starting materials have isotopic compositions that are enriched. Similarly, the ^{14}C composition of plant material is modern ($\sim 0\text{‰}$) while petroleum-based compounds do not contain ^{14}C (-1000‰). These tools give great resolving power to discriminate natural from anthropogenic sources.

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