Results of a Screening Analysis for Pharmaceuticals in Wastewater Treatment Plant Effluents, Wells, and Creeks in the Sequim-Dungeness Area

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Results of a Screening Analysis for Pharmaceuticals in Wastewater Treatment Plant Effluents, Wells, and Creeks in the Sequim-Dungeness Area

by
Art Johnson, Barbara Carey, and Steve Golding

Environmental Assessment Program
Olympia, Washington 98504-7710

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Table of Contents

List of Figures and Tables

Abstract

A screening analysis was conducted for 24 pharmaceuticals and personal care products (PPCPs) in tertiary wastewater treatment plant effluents and nearby wells and creeks in the Sequim-Dungeness area of northwest Washington. The objective was to investigate the potential for and status of PPCP contamination of area waters from application of treated wastewater via re-use programs and conventional land application. A well located in an area served by on-site septic systems also was sampled.

The following 16 compounds were detected in one or both effluents: Acetaminophen, Caffeine, Carbamazepine, Cimetidine, Codeine, Cotinine, Diltiazem, Hydrocodone, Ketoprofen, Metformin, Nicotine, Paraxanthine, Salbutamol, Sulfamethoxazole, Trimethoprim, and Estrone. Concentrations ranged from 0.26 ng/L (Estrone) to 200 ug/L (Paraxanthine). Only Caffeine, Nicotine, and the diabetes drug Metformin (tentatively identified) were consistently detected in the well and creek samples; concentrations were 25 ug/L or less. In at least some samples, Nicotine appeared to be an artifact of sampling handling procedures.

These limited results give no indication that PPCPs represent a significant concern in the wells or creeks sampled. Additional monitoring for PPCPs appears to be a low priority in connection with the Sequim and Sunland wastewater treatment plants.

Acknowledgements

The authors would like to thank the home owners and wastewater treatment plant operators in the Sequim-Dungeness area who gave permission to collect samples for this project.

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- Ann Soule, Clallam County, advised on the field work and reviewed the QA project plan and draft project report.
- Cynthia Nelson, Ecology Southwest Regional Office, provided helpful information for study planning and reviewed the draft project report.
- Dave Dougherty, Ecology Southwest Regional Office, provided helpful information on the Sequim and Sunland wastewater treatment plants.
- Karin Feddersen, Ecology Manchester Laboratory, arranged for contract laboratory services and performed the QA review of the PPCP data.
- Dale Norton, Ecology Environmental Assessment Program, supervised the work and reviewed the QA project plan and draft project report.
- Erika Wittmann, Ecology Environmental Assessment Program, did a literature search on PPCPs.
- Joan LeTourneau, Ecology Environmental Assessment Program, formatted the project report.

Introduction

Identifying emerging risks posed by previously unrecognized pollutants is one of the top five goals of the U.S. Environmental Protection Agency (EPA) Strategic Plan. Pharmaceuticals and certain personal care products (PPCPs) are a large and growing class of bioactive chemicals that, until recently, has received little attention.

Although some PPCPs are resistant to degradation, most have shorter environmental half-lives than conventional pollutants. PPCPs generally occur in surface and groundwater at ug/L to ng/L concentrations[1], far below therapeutic thresholds. There is growing concern about these compounds because many have been detected and little is known of their potential for adverse human or ecological effects. EPA has a website devoted to this issue (http://www.epa.gov/nerlesd1/chemistry/pharma).

In response to a request from Clallam County, the Washington State Department of Ecology (Ecology) conducted a screening analysis for PPCPs in the Sequim-Dungeness area of western Washington during November 2003. Sequim is a popular retirement center. Almost half the residents are over the age of 59, and more than 20 percent are 65 or older (2003). Pharmaceutical use is therefore likely higher than average for Washington communities. These facts, coupled with discharge of land applied reclaimed water and wastewater into or adjacent to surface waters, and a vulnerable surficial aquifer, made the Sequim-Dungeness a good candidate for assessing the potential for pharmaceutical contamination of state waters.

Samples for the screening study were collected of wastewater treatment plant (WWTP) effluents from the City of Sequim and the Sunland development in the Dungeness River valley, as well as from wells and creeks that could be impacted by these discharges. The objective was to investigate the potential for and status of pharmaceutical contamination of area waters from application of treated wastewater via re-use programs (Sequim) and conventional land application (Sunland). A well located in an area served by on-site septic systems also was sampled.

[1] parts-per-billion to parts-per-trillion

Study Area

The Sequim WWTP is a tertiary, high performing, reclaimed water plant that treats 0.5 - 0.6 million gallons per day (mgd). The influent is oxidized, coagulated, filtered, and disinfected. Final effluent meets Washington State Class A standards.

The treatment plant produces about 0.6 mgd of reclaimed water which goes to the city's Reuse Demonstration Site, constructed in 1999-2000 immediately north of Carrie-Blake Park (Figure 1). It is one of the first facilities of its kind in the Pacific Northwest. The reclaimed water is used for garden irrigation and wetland creation, and cooling, aeration, and flow augmentation of Bell Creek. For the past year, the effluent has been used to increase creek flow by 0.1 cfs (0.07 mgd). Unused water is currently discharged to a marine outfall outside of Sequim Bay. (David Dougherty, Ecology, personal communication, 2003; Pacific Groundwater Group, 2000).

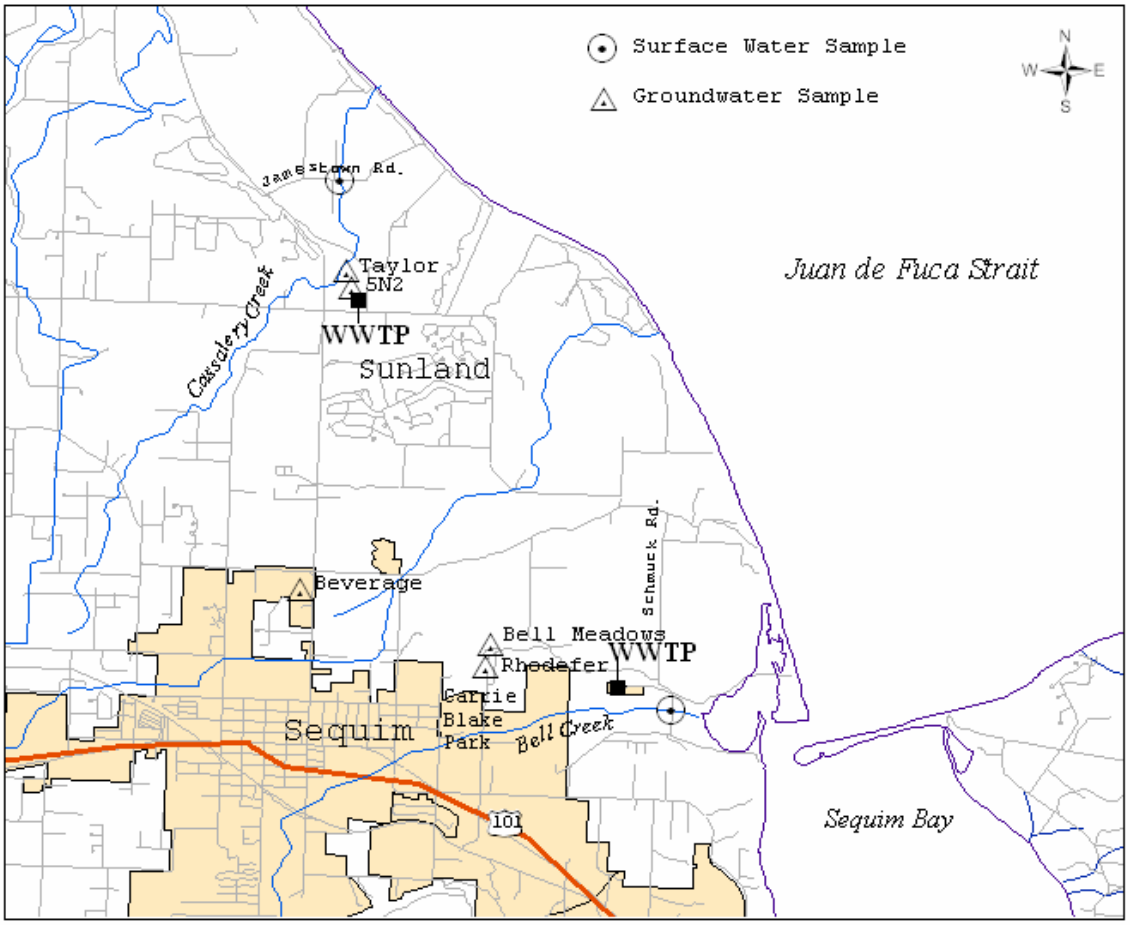

Figure 1. Study Area

Sunland has a quasi-tertiary plant that has not been approved for reclaimed water. It is a land treatment facility that applies at or above agronomic rates. The sprayfield is adjacent to the plant and has been in use since 1979. Cassalery Creek flows along the north boundary of the sprayfield. There is normally no overland discharge to the creek, but effluent could enter the creek during runoff events or via groundwater. Sunland plans an upgrade to achieve Class A reclaimed water status and intends to use treated effluent on their golf course. This upgrade may be years away. (Dougherty, 2003).

Bell and Cassalery creeks are typically fed by groundwater discharge and irrigation tailwater. Irrigation diversions are from the Dungeness River and occur year-round, but are greatest during the growing season, mid-April to mid-September. The lowest flows are in September and October. Highest flows occur during winter rains and in the spring. (Pacific Groundwater Group, 2002).

The number and density of on-site sewage systems have increased in non-sewered portions of the Sequim-Dungeness area corresponding with the population increase in recent years. Thomas et al. (1999) estimated that 7,000 on-site systems existed here in 1996. The relatively shallow depth to groundwater and lack of a low-permeability layer in some areas makes the surficial aquifer vulnerable to contamination from above.

Nitrate is an indicator of groundwater contamination from various sources, including on-site sewage systems. A statistically significant increase in nitrate since 1980, though slight, was reported in this area by Thomas et al. (1999). The largest area of high nitrate concentrations is east of the Dungeness River and north of Bell Creek, where values up to 4.3 mg/L are found. The median nitrate concentration in residential areas (1.3 mg/L) is also higher than in agricultural (0.55 mg/L) or natural grassland or forest areas (0.12 mg/L). Nitrate values are highest in residential areas that have a high density of on-site systems compared to medium density systems, and lowest in low density areas.

The Sequim-Dungeness area is a rural region with a mild and relatively dry climate (15-20" or less annual rainfall). Approximately 4,440 of Clallam County’s 65,900 residents live in the city (2003). 17,400 live in the unincorporated area around Sequim.

Chemicals Analyzed

Table 1 lists the PPCPs that were analyzed for the Sequim-Dungeness project and shows the basis for their selection. An initial target list was developed from recommendations in Daughton and Ternes (1999), a synthesis of the literature on the environmental occurrence, distribution, and effects of PPCPs. Other chemicals were then added that had been detected in a nationwide reconnaissance on the occurrence of PPCPs in surface waters, conducted by the U.S. Geological Survey (USGS) (Kolpin et al., 2002) or were under consideration for groundwater monitoring in the state of California (CDHS, 2003). The final list included several additional analytes that are routine target compounds for EPA-sponsored PPCP research being conducted by the State University of New York at Stony Brook (SUNYSB), the contractor selected for the present study.

Two steroid estrogens, Estrone and beta-Estradiol, were analyzed for this project. These are components or transformation products of drugs used in hormone replacement therapy. The birth control additive, Ethynyl Estradiol, was not analyzed as it is typically lost in the clean-up step for the method employed. It occurs in sewage effluents in lower concentrations than Estrone or beta-Estradiol, and their levels provide a relative indication of how much Ethynyl Estradiol could be present.

Table 2 lists other compounds that were considered for analysis in this project, but ultimately dropped due to lack of an adequate method, high cost, instrument problems at SUNYSB, or their being phased out of use (e.g., clofibrate).

Table 1. PPCPs Analyzed

Chemical	Use/Origin	Basis for Selection	Rank Among Top 200 U.S. Prescriptions 2002*	Rank Among Top 25 Washington State Prescriptions 2003**
Fenofibrate	Lipid regulator	1,4	129	
Carbamazepine	Antiepileptic	1,3,4		
Fluoxetine	Antidepressant	1,2,4	31	5
Codeine	Analgesic	2,4	1	3
Hydrocodone	Analgesic	2,4	1	3
Antipyrine	Analgesic	4		
Caffeine	Stimulant	2,3,4		
Paraxanthine	Caffeine metabolite	4		
Nicotine	Stimulant	4		
Cotinine	Nicotine metabolite	2,4		
Cimetidine	Ulcer drug	2,4		
Ranitidine	Ulcer drug	2,4	39	11
Diltiazem	Antianginal	2,4	92	
Nifedipine	Antianginal	4	194	
Salbutamol	Bronchial dilator	2,4	12	9
Sulfamethoxazole	Antibacterial	2,4		
Trimethoprim	Antibacterial	2,4		
Warfarin	Anticoagulant	2,4	57	23
Estrone	Hormone component	2,3,4		
beta-Estradiol	Hormone component	1,2,3,4		
Erythromycin	Antibiotic	2,4		
Acetaminophen	Anti-inflammatory	2,3,4	32	14
Ketoprofen	Anti-inflammatory	4		
Metformin	Antihyperglycemic	2,4	38	15

1 = Recommended for monitoring (Daughton and Ternes, 1999)
2 = Detected in USGS national study (Koplin et al., 2002)
3 = Under consideration for groundwater monitoring in California (CDHS 2003-draft)
4 = Routine target compound for Stony Brook University, NY
*http://www.rxlist.com/top200a.htm
**Group Health Cooperative (R. Johnson, May 29, 2003 email)

Table 2. PPCPs of Potential Interest but Not Analyzed

Chemical	Use/Origin
Gemfibrozil	Lipid regulator
Phenytoin	Antiepileptic
Primidone	Antiepileptic
17a-Ethynyl Estradiol	Oral contraceptive
Fluoroquinolone carboxylic acids	Antibiotics
Ibuprofen	Anti-inflammatory
Triclosan	Antiseptic
Clofibric acid	Lipid regulator (metabolite)
Fluvoxamine	Antidepressant
Paroxetine	Antidepressant
Ifosfamide	Antineoplastic
Cyclophosphamide	Antineoplastic
Diatrizoate (Na)	X-ray media
Iopamidol	X-ray media
Iopromide	X-ray media
Nitromusks	Fragrances
Amino musks	Fragrances
Sulfonamides	Antibiotics
Acetylsalicylic acid	Anti-inflammatory
Sildenafil citrate	Impotence drug
Methylbenzylidene camphor	Sunscreen agent
Diphenhydramine	Antihistamine

Sampling Design

Samples for PPCP screening were collected on November 17-18, 2003. A timeframe of mid-October had been initially selected for the field work as being worst-case for surface water, with the irrigation season ended and the creeks at low flow. However, the services of the contract laboratory could not be secured in time. The November data are more representative of general water quality conditions in these creeks, as opposed to extreme low flow.

The samples were analyzed for the chemicals listed in Table 1. Ancillary parameters included temperature, pH, conductivity, nitrate+nitrite-N, total suspended solids (TSS), and dissolved oxygen (groundwater only). Effluent flow rates were recorded from WWTP records and streamflow was gauged when the surface water samples were taken.

One sample each was collected of final effluents from the Sequim and Sunland WWTPs. The samples were composites of a morning and early afternoon grab. Effluent flow in WWTPs is highest in the morning, but can be more concentrated in the afternoon. A replicate effluent sample was prepared at Sunland. Sunland was selected for the replicate because of the lower level of treatment it provides.

A total of five wells were sampled: two near the Sunland sprayfield, two near the Sequim Reuse Demonstration Site, and one in an area of high nitrate concentrations (Figure 1, Table 3). The wells were selected in consultation with Ann Soule, hydrogeologist with Clallam County. The criteria for selecting wells included the following:

- The well is down-gradient of the site of interest.
- A driller's report (well log) is available for the well (if possible).
- The well is screened in as shallow an aquifer as possible – above any clay layer (if possible).
- The well is capable of producing samples representative of the groundwater.
- The well does not have a water treatment device (such as a water softener or iron treatment system) or a large storage tank that cannot be bypassed during well purging and sampling.
- The current well owner must grant access to the well.

One water sample each was collected from Bell Creek and Cassalery Creek. Bell was sampled at Schmuck Road, just before it flows into Sequim Bay (Figure 1). This site is approximately one mile below Carrie Blake Park. Cassalery was sampled at Jamestown Road, approximately ½ mile below the Sunland WWTP. A replicate sample was prepared for Bell Creek. Bell Creek was selected for the replicate because it receives effluent directly.

Table 4 shows the numbers and types of samples analyzed. This project was conducted following a Quality Assurance Project Plan prepared by Johnson and Carey (2004).

Table 3. Wells Sampled

Well ID	Location	Type of Well	Depth (ft)	Well Log?
Sunland WWTP Area				
5N2	Sunland land application site	monitoring	44	yes
Taylor Ranch Road	444 Taylor Ranch Rd.	private	49	yes
Sequim WWTP Area				
Bell Meadows Lane	154 Bell Meadows Lane	private	60?	no
Rhodefer Road	N. end N. Rhodefer Rd.	community	?	?
High Nitrate Area				
Beverage Street	885 N. Beverage St.	private	67	yes

Table 4. Number and Type of Samples Analyzed

Analysis	WWTP Effluent	Ground-water	Surface Water	Bottle Blank	Total Samples
Pharmaceuticals	3	5	3	1	12
Steroid Estrogens	3	3*	2*	0*	8
Nitrate+Nitrite-N	0	5	2	0	7
TSS	2	0	2	0	4

*Samples were lost in transport or not analyzed due to laboratory oversight

Methods

Field Procedures

Sample containers, preservation, and holding times for this project are shown in Table 5. The containers were obtained from the Ecology Manchester Environmental Laboratory, pre-cleaned to EPA QA/QC specifications (EPA, 1990).

Table 5. Field Procedures

Parameter	Minimum Sample Size	Container	Preservation	Holding Time
Pharmaceuticals	1 L	1L amber glass, teflon lid	Cool to 4°C	filter within 2 days
Steroid Estrogens	1 L	1L amber glass, teflon lid	10mL formalin, 4°C	*
Nitrate+Nitrite-N	125 mL	125 mL poly bottle	H_2SO_4 to pH<2, 4°C	28 days
TSS	1 L	1 L poly bottle	Cool to 4°C	7 days

*holding time not established

The PPCP samples were shipped by FedEx and arrived at SUNYSB the morning after collection. Formalin preservative for the PPCP analysis was added within 24 hours of sample arrival. Samples for conventional parameters were returned to the Ecology Headquarters and picked up by the Manchester Laboratory courier the next day. All samples were accompanied by a chain-of-custody record and the coolers sealed with chain-of-custody tags or tape.

Effluents

Effluent composites from the Sequim and Sunland WWTPs were collected by filling the sample containers with equal amounts of water from morning and afternoon grabs. The grabs were taken with the same glass jars used for PPCPs. The samples were kept on ice and in the dark during the compositing period. Field personnel wore nitrile gloves while doing this work.

pH and conductivity were measured when the grabs are taken. pH was determined with an Orion Model 25A meter and conductivity with a Beckman Model RB-5 conductivity bridge. A Magellan 320 GPS was used to obtain the latitude and longitude of the effluent sampling sites (Appendix A).

Groundwater

Wells selected for sampling were field located on USGS 1:24,000 quad maps. Groundwater levels were measured at each of the study wells prior to sampling, where possible. Water level measurements were made using a calibrated electric well probe or steel tape in accordance with standard USGS methods (Stallman, 1983).

Wells were purged prior to sampling. A peristaltic pump was used for the Sunland monitoring well, which does not have a pump. Domestic wells were purged using the existing pump in the well. Roughly three well volumes were purged before samples are collected. Purge water was discharged to an enclosed flow cell where temperature, pH, conductivity, and dissolved oxygen were monitored and recorded every three minutes using a WTW P4 Multi-parameter field meter. Samples were collected after flow cell measurements stabilized.

Surface Water

Water samples from Bell and Cassalery creeks were collected from the center channel directly into the appropriate sample containers. The samples were put on ice immediately on collection. pH and conductivity were measured as described above for WWTP effluents. Temperature was recorded from a meter. Streamflow was gauged with a Swoffer Model 2100 meter and top-setting rod. A Magellan 320 GPS was used to determine the latitude and longitude of the sampling sites.

Laboratory Procedures

Table 6 shows the laboratory procedures used by SUNYSB in the project.

Table 6. Laboratory Procedures

Parameter	Sample Prep Method	Extraction Method	Analytical Method
Pharmaceuticals	Filter	SPE	HPLC-MS
Steroid Estrogens	Filter	SPE	HPLC-Electrospray-MS
Nitrate+Nitrite-N	na	na	EPA 353.2
TSS	na	an	EPA 2540D

na = not applicable

The pharmaceuticals method is outlined in Kolpin et al. (2002). The compounds are extracted from filtered, one-liter water samples using solid-phase extraction cartridges. The adsorbed compounds are then eluted with methanol. The extract is reduced to near dryness under nitrogen gas and brought to a final volume of 1 mL in acetonitrile. Compounds are separated and measured by High Pressure Liquid Chromatography-Mass Spectrometry (HPLC-MS) in positive ion mode.

The estrogen method is described in Ferguson et al. (2001). Filtered, one-liter water samples are extracted by solid-phase extraction and the resulting extract purified by passing over a selective immuno-affinity extraction column. The only significant change from Ferguson et al. is that time-of-flight MS was used instead of single quadrupole MS, providing additional sensitivity and confirmation based on accurate mass.

Data Quality

The PPCP data reported here are from a wide sweep analysis to get multi-class analyte detection. Unlike the estrogen analysis, where there is an isotopically labeled internal standard for each analyte, the PPCP data use only a single isotopically labeled internal standard; more isotopically labeled standards have become available recently.

SUNYSB has a high level of confidence in the estrogen data. They consider the PPCP data to be equal or better in quality to what USGS has reported in their national studies (e.g., Kolpin et al., 2002), the most important PPCP studies to date. (Brownawell, 2004).

Metformin appeared to be present in most of the samples for the present study. This compound has poor recovery and does not separate well from the solvent peak on the HPLC. Because of uncertainties associated with its detection, Metformin is reported here as being tentatively identified.

An appreciation for the total variability associated with the data generated for this project can be gained from Table 7 which has results on the replicate effluent and surface water samples. For most of the PPCPs analyzed, the results were similar. In 11 of the 17 instances where a compound was quantified in the replicates, the results agreed within a factor of 2 or better. Five compounds were either detected in one replicate only and/or were below the limit of quantification. For unknown reasons, there was a high degree of variability associated with the Estrone analysis.

A bottle blank, prepared at Manchester Laboratory, was analyzed to detect contamination arising from sample handling procedures. This sample was carried through the field work and treated the same as the other samples. Only Nicotine was detected in the blank; the concentration was 17 ug/L. There were no known sources of Nicotine during the field work. Due to an oversight by the laboratory, the field blank was not analyzed for estrogen compounds.

Table 7. Comparison of Results on Replicate Samples
(PPCPs reported in ug/L, except Estrone and beta-Estradiol in ng/L)

Compound	Sunland WWTP Effluent	Sunland WWTP Effluent – Replicate	RPD*	Bell Creek	Bell Creek - Replicate	RPD*
Acetaminophen	47	nd	- -	nd	nd	- -
Antipyrine	nd	nd	- -	nd	nd	- -
Caffeine	30	25	17	nd	nd	- -
Carbamazepine	0.8	1.7	74	nd	nd	- -
Cimetidine	nd	nd	- -	nd	nd	- -
Codeine	2.8	2.5	12	nd	nd	- -
Cotinine	11	19	55	nd	nd	- -
Diltiazem	<LOQ	<LOQ	- -	nd	nd	- -
Erythromycin	nd	nd	- -	nd	nd	- -
Fenofibrate	nd	nd	- -	nd	nd	- -
Fluoxetine	nd	nd	- -	nd	nd	- -
Hydrocodone	nd	nd	- -	nd	nd	- -
Ketoprofen	45	42	6	nd	nd	- -
Metformin**	150	115	26	11	12	11
Nicotine	25	27	8	25	16	45
Nifedipine	nd	nd	- -	nd	nd	- -
Paraxanthine	218	194	11	nd	nd	- -
Ranitidine	4.3	3.3	29	nd	nd	- -
Salbutamol	nd	13	- -	nd	nd	- -
Sulfamethoxazole	<LOQ	10	- -	nd	nd	- -
Trimethoprim	14	19	32	nd	nd	- -
Warfarin	nd	nd	- -	nd	nd	- -
Estrone	3.5	0.29	169	0.26	nd	- -
beta-Estradiol	nd	nd	- -	nd	nd	- -

*Relative Percent Difference: range of values as percent of mean value
**tentatively identified
nd = not detected
<LOQ = below the limit of quantification

Results

Effluent and Creek Flows

Table 8 has flow data that pertain to the PPCP samples collected for the Sequim-Dungeness project.

Table 8. Effluent and Creek Flow Rates During PPCP Sample Collection (mgd)

Date (2003)	Sunland Effluent		Sequim Effluent		Cassalery Creek	Bell Creek
	Average	Irrigation Flow	Average	During Sampling		
17-Nov	0.132	0.152	0.588	0.75	1.4	2.9
18-Nov	0.117	0.181	1.06	1.5	- -	- -

Sunland effluent was collected on November 18, at which time the plant was experiencing a normal flow rate of 0.117 mgd. Effluent is stored in a pond and applied to the sprayfield only during weekdays. Irrigation rates during sampling for the PPCP study were 0.152 and 0.180 mgd.

A rainstorm occurred on November 18, becoming heavy between the morning and afternoon samples at the WWTPs. Although Sunland's flow did not appear to be affected, there was a large increase in the daily average flow for Sequim. Sequim typically has an effluent flow rate of 0.45 – 0.60 mgd compared to an average of 1.06 mgd on the 18th, one of the highest recorded for that facility. The first PPCP sample collected on the 18th was during a flow of approximately 0.75 mgd. The second sample was during a very high flow of approximately 1.5 mgd.

The creeks were sampled the day before the WWTPs. Cassalery Creek was flowing at 1.4 mgd. No other flow data were available for comparison.

Bell Creek was at 2.9 mgd during sample collection, which is close to the historical average of 3.0 mgd. Flow in Bell Creek ranges from 2.1 – 12 mgd (NPDES pre-permit application).

Sunland WWTP and Related Samples

The results for the Sunland WWTP effluent, a Sunland monitoring well, an adjacent private well, and nearby Cassalery Creek are summarized in Table 9. No data was obtained on estrogen compounds in the wells or creek, because the well 5N2 sample was broken in transport. The other two samples were not analyzed due to an oversight by the laboratory.

Two replicate samples of Sunland effluent were collected. Fourteen of the 24 compounds analyzed were detected, with 11 of these being identified in both samples. Concentrations were similar between the replicates, except for Acetaminophen, Sulfamethoxazole, and Estrone.

Almost half of PPCPs detected in Sunland effluent are common, non-prescription substances. Two compounds, Paraxanthine, a caffeine metabolite, and Metformin, used in the treatment of diabetes, were detected at concentrations ranging from approximately 100 – 200 ug/L. As noted previously, Metformin was tentatively identified. Seven compounds were detected at approximately 10 – 100 ug/L. In addition to Caffeine, Nicotine, and Cotinine (a nicotine metabolite), these included the anti-inflammatory Ketoprofen; Trimethoprim, used to treat a variety of bacterial infections; and the pain reliever Acetaminophen.

Five additional compounds - Codeine, Carbamazepine, Ranitidine, Sulfamethoxazole, and Estrone - were present at low concentrations of 10 ug/L or less. Carbamazepine is an antiepileptic, Ranitidine an ulcer drug, Sulfamethoxazole an antibacterial agent, and Estrone is a steroid hormone.

Only Nicotine, Caffeine, and Metformin were detected in Sunland monitoring well 5N2. Concentrations were 14 ug/L, 3.0 ug/L, and 1.0 ug/L, respectively. These and related compounds were among the most abundant in the effluent samples. Effluent concentrations were 2-to-20 times higher than the groundwater.

Except for Nicotine, no PPCPs were detected in the Taylor Ranch Road well. This well lies along the north edge of the WWTP sprayfield, approximately 400 yards from well 5N2.

As noted earlier, Nicotine was detected in the field blank for this project at 17 ug/L. The results reported here (Tables 8-10) have been corrected for the blank. For the Taylor Ranch Road well, the Nicotine concentration was 22 ug/L before blank correction, only slightly higher than the blank. Therefore the detection of Nicotine in this well can largely be attributed to contamination of the sample.

The same compounds identified in the Sunland monitoring well were also detected in Cassalery Creek, i.e., Nicotine, Caffeine, and Metformin. Concentrations were slightly higher than in the monitoring well and ranged from 1.9 – 25 ug/L. In this case Nicotine was substantially higher than the blank, so its presence may not be solely due to sample contamination.

There was some evidence of extremely low concentrations of Codeine, Cotinine, and Sulfamethoxazole in the wells and/or Cassalery Creek, but the concentrations were lower than the limit of quantification. Diltiazem, used in the treatment of angina, may have been present in Sunland effluent, but was also below the quantification limit.

Table 9. Results for Sunland WWTP, Adjacent Wells, and Nearby Cassalery Creek
[PPCPs reported in ug/L, except Estrone and beta-Estradiol in ng/L)]

Sample Location	Sunland WWTP	Sunland WWTP-Rep.	Sunland Monit'g Well 5N2	Taylor Ranch Rd. Residence	Cassalery Creek
Sample Type	Effluent	Effluent	Groundwater	Groundwater	Surface Water
Sample Number	474136	474137	474132	474133	474140
Collection Date	18-Nov-03	18-Nov-03	18-Nov-03	18-Nov-03	17-Nov-03
Collection Time	1050/1400	1050/1400	0930	1030	1200
Temperature (oC)	na	na	10.0	10.2	8.5
pH (S.U.)	na	na	6.9	7.1	7.8
Conductivity (umhos/cm)	465	650	386	358	230
TSS (mg/L)	2	<1	na	na	1
Nitrate-Nitrite (mg/L)	na	na	1.0	1.0	0.49
Dissolved Oxygen (mg/L)	na	na	2.7	5.5	na
Acetaminophen	47	nd	nd	nd	nd
Antipyrine	nd	nd	nd	nd	nd
Caffeine	30	25	1.0	<LOQ	1.9
Carbamazepine	0.8	1.7	nd	nd	<LOQ
Cimetidine	nd	nd	nd	nd	nd
Codeine	2.8	2.5	<LOQ	<LOQ	<LOQ
Cotinine	11	19	<LOQ	<LOQ	<LOQ
Diltiazem	<LOQ	<LOQ	nd	nd	nd
Erythromycin	nd	nd	nd	nd	nd
Fenofibrate	nd	nd	nd	nd	nd
Fluoxetine	nd	nd	nd	nd	nd
Hydrocodone	nd	nd	nd	nd	nd
Ketoprofen	45	42	nd	nd	nd
Metformin*	150	115	3.0	<LOQ	11
Nicotine	25	27	14	5.0	25
Nifedipine	nd	nd	nd	nd	nd
Paraxanthine	218	194	nd	nd	nd
Ranitidine	4.3	3.3	nd	nd	nd
Salbutamol	nd	13	nd	nd	nd
Sulfamethoxazole	<LOQ	10	nd	<LOQ	nd
Trimethoprim	14	19	nd	nd	nd
Warfarin	nd	nd	nd	nd	nd
Estrone	3.5	0.29	na	na	na
beta-Estradiol	nd	nd	na	na	na

na = not analyzed
nd = not detected
<LOQ = below the limit of quantification
*tentatively identified

Sequim WWTP and Related Samples

Sixteen compounds were detected in the Sequim WWTP effluent (Table 10). Thirteen of the 16 were also detected in Sunland effluent. The three additional compounds identified in Sequim effluent were Cimetidine, an ulcer medication; Diltiazem, for treatment of angina; and Hydrocodone, a pain reliever.

PPCP concentrations in Sequim effluent ranged from 4.2 – 200 ug/L, similar to Sunland effluent. Prescription drugs were generally present in higher concentrations than at Sunland, while concentrations of non-prescription substances were comparable.

Caffeine, Nicotine, and Metformin were detected in private wells in the vicinity of the Sequim Reuse Demonstration Site. The same compounds were detected in the Sunland well and at similar concentrations (1.0 – 7.5 ug/L vs. 1.0 – 14 ug/L). Sample contamination is the likely source of the Nicotine, as described above.

Metformin, Nicotine, and Estrone were also detected in Bell Creek. The Metformin and Nicotine concentrations compared closely to what was found in Cassalery Creek, ranging from 11 – 25 ug/L for Bell Creek. Estrone was only detected in one of the two Bell Creek replicates and at a trace concentration of 0.26 ng/L. Unlike Cassalery Creek, Caffeine was not detected here.

Table 10. Results for Sequim WWTP, Wells Adjacent to Water-Reuse Project, and Lower Bell Creek (PPCPs reported in ug/L, except Estrone and beta-Estradiol in ng/L)

Sample Location	Sequim WWTP	Bell Meadows Ln. Residence	Rhodefer Rd. Community	Bell Creek	Bell Creek – Replicate
Sample Type	Effluent	Groundwater	Groundwater	Surface Water	Surface Water
Sample Number	474135	474131	474130	474138	474139
Collection Date	18-Nov-03	17-Nov-03	17-Nov-03	17-Nov-03	17-Nov-03
Collection Time	0745 / 1335	1340	1220	1100	1105
Temperature (oC)	na	10.9	10.5	7.7	na
pH (S.U.)	na	7.2	8.0	7.2	na
Conductivity (umhos/cm)	510	422	328	347	na
TSS (mg/L)	<1	na	na	5	4
Nitrate-Nitrite (mg/L)	na	<0.10	0.17	1.7	2.1
Dissolved Oxygen (mg/L)	na	0.9	0.0	na	na
Acetaminophen	nd	nd	nd	nd	nd
Antipyrine	nd	nd	nd	nd	nd
Caffeine	21	1.0	3.8	nd	nd
Carbamazepine	43	nd	nd	nd	nd
Cimetidine	127	nd	nd	nd	nd
Codeine	12	<LOQ	<LOQ	nd	nd
Cotinine	21	<LOQ	<LOQ	nd	nd
Diltiazem	10	nd	nd	nd	nd
Erythromycin	nd	nd	nd	nd	nd
Fenofibrate	nd	nd	nd	nd	nd
Fluoxetine	nd	nd	nd	nd	nd
Hydrocodone	2.9	nd	nd	nd	nd
Ketoprofen	52	nd	nd	nd	nd
Metformin	97	7.5	3.4	11	12
Nicotine	54	6.3	1.9	25	16
Nifedipine	nd	nd	nd	nd	nd
Paraxanthine	200	nd	nd	nd	nd
Ranitidine	5.1	nd	nd	nd	nd
Salbutamol	60	<LOQ	nd	nd	nd
Sulfamethoxazole	4.2	nd	nd	nd	nd
Trimethoprim	13	nd	nd	nd	nd
Warfarin	nd	nd	nd	nd	nd
Estrone	2.6	nd	nd	0.26	nd
beta-Estradiol	nd	nd	nd	nd	nd

na = not analyzed
nd = not detected
<LOQ = below the limit of quantification
*tentatively identified

Residential Area Well

The results for the Beverage Street well are in Table 11. Although Nicotine was present in this sample, the concentration was lower than the field blank (13 ug/L vs. 17 ug/L). No other PPCPs were unambiguously detected. There was some evidence of the presence of Caffeine, Carbamazepine, Codeine, Cotinine, and Metformin.

The nitrate-nitrite concentration in this well was 3.3 mg/L, which is above background concentrations but below the Washington State Department of Health drinking water standard of 10 mg/L nitrogen for Class A public water systems. Possible sources of nitrate in the area include not only on-site sewage systems, but also domestic/commercial landscaping, historical agricultural practices, or a combination of sources. Other wells in the study had nitrate-nitrite concentrations in the range of <0.1 – 1.0 mg/L.

Table 11. Results for Private Well Affected by Septic Systems
(PPCPs reported in ug/L, except Estrone and beta-Estradiol in ng/L)

Sample Location	Beverage St. Residence
Sample Type	Groundwater
Sample Number	474134
Collection Date	18-Nov-03
Collection Time	1155
Temperature (oC)	10.5
pH (S.U.)	8
Conductivity (umhos/cm)	310
Total Suspended Solids (mg/L)	na
Nitrate-Nitrite (mg/L)	3.3
Dissolved Oxygen (mg/L)	9.7
Acetaminophen	nd
Antipyrine	nd
Caffeine	<LOQ
Carbamazepine	<LOQ
Cimetidine	nd
Codeine	<LOQ
Cotinine	<LOQ
Diltiazem	nd
Erythromycin	nd
Fenofibrate	nd
Fluoxetine	nd
Hydrocodone	nd
Ketoprofen	nd
Metformin*	<LOQ
Nicotine	nd
Nifedipine	nd
Paraxanthine	nd
Ranitidine	nd
Salbutamol	nd
Sulfamethoxazole	nd
Trimethoprim	nd
Warfarin	nd
Estrone	nd
beta-Estradiol	nd

na = not analyzed
nd = not detected
<LOQ = below the limit of quantification
*tentatively identified

Summary of Detection Frequencies

Table 12 summarizes the frequency with which the PPCPs analyzed in the Sequim-Dungeness study were detected. The results for replicate samples were pooled to calculate this statistic.

In terms of simple occurrence, the prescription drug of primary interest appears to be the antihyperglycemic Metformin. Metformin, a highly water soluble compound, ranks 15th among the top 25 most prescribed drugs in Washington (Table 1). Metformin was detected in all samples analyzed for the present study. Nicotine and Caffeine were also detected in most samples. These were the only three compounds clearly detected in groundwater or surface water, although in several cases Nicotine appeared to be an artifact of sample handling procedures.

The other detections were restricted to WWTP effluent. PPCPs common to both WWTPs included Carbamazepine, Codeine, Ketoprofen, Ranitidine, Salbutamol, Sulfamethoxazole, and Estrone. Codeine, Ranitidine, Salbutamol are among the top 25 prescriptions in Washington. The Nicotine and Caffeine metabolites Cotinine and Paraxanthine were also detected in both effluents.

Acetaminophen, Cimetidine, Diltiazem, and Hydrocodone were only found in the Sequim effluent. With the higher level of treatment achieved by the Sequim WWTP and the high effluent flow during sample collection, the detection of more compounds – some at higher concentrations – than in the Sunland effluent runs counter to expectations. However, with only one sample being analyzed, these results may not be representative of typical effluent quality.

Antipyrine, Erythromycin, Fenofibrate, Nifedipine, and Warfarin were analyzed but not detected in this study.

Table 12. Detection Frequency for PPCPs in Sequim-Dungeness Effluent, Well, and Creek Samples*

Compound	Detection Frequency (%)
Nicotine**	100
Metformin†	78
Caffeine	67
Estrone	33
Carbamazepine	22
Codeine	22
Cotinine	22
Ketoprofen	22
Paraxanthine	22
Ranitidine	22
Salbutamol	22
Sulfamethoxazole	22
Trimethoprim	22
Acetaminophen	11
Cimetidine	11
Diltiazem	11
Hydrocodone	11
Antipyrine	0
Erythromycin	0
Fenofibrate	0
Fluoxetine	0
Nifedipine	0
Warfarin	0
beta-Estradiol	0

*Replicate results were pooled.

**Some Nicotine detections appeared due to blank contamination.

†tentatively identified

Significance of Findings

The Sequim-Dungeness PPCP project was intended as a screening study to determine the presence or absence of selected PPCPs in a one-time collection of a limited number of samples. More sampling would be required to understand how frequently and at what concentrations these compounds occur in effluents, groundwater, and surface water in the Sequim-Dungeness area. The limited data obtained suggest the Sequim and Sunland effluents are not significant sources of PPCP contamination to local wells or creeks. With only one sample being analyzed, no inferences can be made as to whether on-site sewage systems are significant PPCP sources to groundwater.

Nicotine, Caffeine, and Metformin were detected in the well and creek samples. Caffeine and Nicotine are ubiquitous contaminants. USGS detected Caffeine and Nicotine in approximately 65% and 35%, respectively, of samples from 139 streams nationwide (Koplin et al., 2002). Metformin was detected in approximately 5% of the samples. The levels of these compounds found in the Sequim-Dungeness well and creek samples are far below known toxicity or therapeutic thresholds. It should be noted that WWTP effluents are not the only potential sources of these compounds in the study area.

Comparable data could not be located for similar situations involving discharge of tertiary effluents. Dr. Bruce Brownawell, PPCP researcher at Stony Brook University and director of the laboratory that did the analyses for this project, characterized the Sequim-Dungeness results as follows: “One can say that the levels are low/very low compared to most comparable wastewaters and receiving waters” (Brownawell, 2004). Screening study results suggest that the treatment and waste disposal practices of the Sequim and Sunland WWTPs are effective in dealing with PPCP concentrations in wastewater from a community with a preponderance of older citizens. The wells sampled in this survey did not have PPCPs at levels that constitute a human health hazard.

Recommendations

1. Results of this study suggest additional monitoring for personal care products (PPCPs) is a low priority in connection with the Sequim and Sunland wastewater treatment plants.

2. A more balanced perspective on the occurrence of PPCPs in Washington State surface waters would be obtained by doing a similar study for secondary effluents where there is direct discharge to receiving waters, a more typical scenario for wastewater disposal.

3. If further PPCP analyses are done, the chemicals detected in the present study should be included and additional PPCPs (e.g., Table 2) considered for analysis as current research may indicate.

References

Brownawell, B. 2004. June 23 e-mail to A. Johnson, Washington State Department of Ecology, Olympia, WA. SUNYSB, New York.

CDHS. 2003. Discussion document for draft recharge regulations, April 21, 2003. California Department of Health Services.

Daughton, C.G. and T.A. Ternes. 1999. Pharmaceuticals and Personal Care Products: Agents for Subtle Change? Environ. Health Persp. 107(6):907-942.

Dougherty, D. 2003. Personal communication. Washington State Department of Ecology, Olympia. WA.

EPA. 1990. Specifications and Guidance for Obtaining Contaminant-Free Sample Containers. OSWER Directive #93240.0-05.

Ferguson P.L., C.R. Iden, A.E. McElroy, and B.J. Brownawell. 2001. Determination of Steroid Estrogens in Wastewater by Immunoaffinity Extraction Coupled with HPLC-electrospray-MS. Anal. Chem. 73: 3890-3895.

Johnson, A. and B. Carey. 2004. Quality Assurance Project Plan: Screening for Pharmaceuticals in Wastewater Treatment Plant Effluents, Groundwater, and Surface Water in the Sequim Dungeness Area. Washington State Department of Ecology, Olympia, WA. Pub. No. 04-03-104. http://www.ecy.wa.gov/biblio/0403104.html

Kolpin, D.W., E.T. Furlong, M.T. Meyer, E.M. Thurman, S.D. Zaugg, L.B. Barber, and H.T. Buxton. 2002. Pharmaceuticals, Hormones, and Other Organic Wastewater Contaminants in U.S. Streams, 1999 - 2000: A National Reconnaissance. Environ. Sci. Technol. 36:1202-1211.

Pacific Groundwater Group. 2002. City of Sequim 2001 Hydrologic Monitoring Report. Pacific Groundwater Group, Seattle, WA.

Stallman, R.W. 1983. Aquifer-test Design, Observation, and Data Analysis: Techniques of Water Resources Investigations of the U.S. Geological Survey, Book 3, Chapter B1, 26 p.

Thomas, T., E.T. Blakemore, L.A. Goodman, and T.D. Olsen. 1999. Hydrogeologic Assessment of the Sequim-Dungeness Area, Clallam County, Washington. U.S. Geological Survey, Water-Resources Investigations Report 99-4048, 165 p.

Appendix A. Latitude and Longitude of PPCP Sampling Sites

Sampling Site	Latitude (N)	Longitude (W)
Sequim WWTP Final Effluent	48° 5.093'	123° 3.683'
Sunland WWTP Final Effluent	48° 7.004'	123° 5.773'
Sunland Monitoring Well 5N2	48° 7.148'	123° 5.746'
Taylor Ranch Road Well	48° 7.187'	123° 5.786'
Bell Meadows Lane Well	48° 5.248'	123° 4.628'
Rhodefer Road Well	48° 5.198'	123° 4.733'
Beverage Street Well	48° 5.414'	123° 6.397'
Bell Creek @ Schmuck Road	48° 5.025'	123° 3.340'
Cassalery Creek @ Jamestown Road	48° 7.620'	123° 5.969'

Appendix B. Summary of PPCP Results for the Sequim-Dungeness Project (ug/L, except Estrone and beta-Estradiol in ng/L)

Sample Location	Sunland WWTP	Sunland WWTP-Rep.	Sunland 5N2	Taylor Ranch Road	Cassalery Creek	Sequim WWTP	Bell Meadows Lane	Rhodefer Road	Bell Creek	Bell Creek - Rep.	Beverage Street	Bottle Blank
Sample Type	Effluent	Effluent	Well	Well	Water	Effluent	Well	Well	Water	Water	Well	- -
Sample Number	474136	474137	474132	474133	474140	474135	474131	474130	474138	474139	474134	474141
Collection Date	18-Nov-03	18-Nov-03	18-Nov-04	18-Nov-04	17-Nov-03	18-Nov-03	17-Nov-04	17-Nov-04	17-Nov-03	17-Nov-03	18-Nov-04	- -
Collection Time	1050/1400	1050/1400	0930	1030	1200	0745 / 1335	1340	1220	1100	1105	1155	- -
Acetaminophen	47.0	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Antipyrine	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Caffeine	29.6	25.1	1.0	<LOQ	1.9	21.3	1.0	3.8	nd	nd	<LOQ	nd
Carbamazepine	0.8	1.7	nd	nd	<LOQ	42.8	nd	nd	nd	nd	<LOQ	nd
Cimetidine	nd	nd	nd	nd	nd	127.4	nd	nd	nd	nd	nd	nd
Codeine	2.8	2.5	<LOQ	<LOQ	<LOQ	12.1	<LOQ	<LOQ	nd	nd	<LOQ	nd
Cotinine	10.8	18.9	<LOQ	<LOQ	<LOQ	21.0	<LOQ	<LOQ	nd	nd	<LOQ	nd
Diltiazem	<LOQ	<LOQ	nd	nd	nd	10.2	nd	nd	nd	nd	nd	nd
Erythromycin	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Fenofibrate	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Fluoxetine	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Hydrocodone	nd	nd	nd	nd	nd	2.9	nd	nd	nd	nd	nd	nd
Ketoprofen	45.1	42.4	nd	nd	nd	52.2	nd	nd	nd	nd	nd	nd
Metformin*	150.2	115.3	3.0	<LOQ	10.6	97.2	7.5	3.4	10.8	12.0	<LOQ	nd
Nicotine	24.7	26.8	31.5	22.3	42.7	53.7	23.6	19.2	42.1	33.0	13.2	17.3
Nifedipine	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Paraxanthine	217.8	194.2	nd	nd	nd	199.8	nd	nd	nd	nd	nd	nd
Ranitidine	4.3	3.3	nd	nd	nd	5.1	nd	nd	nd	nd	nd	nd
Salbutamol	nd	13.3	nd	nd	nd	60.2	<LOQ	nd	nd	nd	nd	nd
Sulfamethoxazole	<LOQ	10.1	nd	<LOQ	nd	4.2	nd	nd	nd	nd	nd	nd
Trimethoprim	14.0	19.5	nd	nd	nd	13.4	nd	nd	nd	nd	nd	nd
Warfarin	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Estrone	3.5	0.29	na	na	na	2.6	nd	nd	0.26	nd	nd	na
beta-Estradiol	nd	nd	na	na	na	nd	nd	nd	nd	nd	nd	na

na = not analyzed
nd = not detected
<LOQ = below the limit of quantification
*tentatively identified