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State of Washington

Quality Assurance Project Plan

Phase 2: High Summer Bacteria Concentrations in Streams

September 2010

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This plan is available on the Department of Ecology's website at www.ecy.wa.gov/biblio/1003110.html.

Data for this project will be available on Ecology's Environmental Information Management (EIM) website at www.ecy.wa.gov/eim/index.htm. Search User Study ID, NMat0003.

Ecology's Activity Tracker Code for this study is 10-153.

Waterbody Numbers:

WA-13-1100	McLane Creek
WA-14-1300	Kennedy Creek
WA-14-1750	Deer Creek
WA-15-1400	Burley Creek

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Quality Assurance Project Plan

Phase 2: High Summer Bacteria Concentrations in Streams

September 2010

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Signatures are not available on the Internet version.
SWRO – Southwest Regional Office
EAP - Environmental Assessment Program
EIM - Environmental Information Management system

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Abstract

The Washington State Department of Ecology (Ecology) conducted a study in 2008 to identify and analyze environmental conditions in streams with high bacteria levels during the summer months. The study was entitled High Summer Bacteria Concentrations in Streams. The study recommended a second monitoring phase (Phase 2) be conducted to provide additional needed information.

Ecology is conducting this study to complete the Phase 2 recommendations and research the role that streambed sediments play in contributing to high summer bacteria concentrations in South Puget Sound streams.

High bacteria concentrations in rivers and streams indicate the potential presence of harmful pathogens that pose a public health risk to the people that recreate in rivers and streams. In addition, these high bacteria streams often drain to marine waterbodies with public swimming beaches or shellfish harvesting areas. Elevated pathogen levels in the water can accumulate in shellfish tissue, making them unsafe to eat.

Each study conducted by Ecology must have an approved Quality Assurance Project Plan. This project plan describes the objectives of the study and the procedures to be followed to achieve those objectives. After completion of the study, a final report describing the study results will be posted to the Internet.

Background

Water quality specialists in Southwest Washington have recently identified a number of South Puget Sound streams which, in the summer months have high bacteria concentrations that exceed Washington State Water Quality Standards (www.ecy.wa.gov/programs/wq/swqs/criteria-freshwater/wac173201a_200-bacteria.html).

Local governments have requested more information about the cause of high summer bacteria levels to address the source of the bacteria problems. In 2008, the Washington State Department of Ecology's (Ecology) Environmental Assessment Program (EAP) undertook a project to study a population of stream bacteria data. Thurston County Environmental Health, the Squaxin Island Tribe, and Ecology's Water Quality Program, Southwest Regional Office requested the project. The project goal was to identify and analyze streams with high bacteria levels during the summer (Bell-McKinnon, 2008).

The report for this project, *High Summer Bacteria Concentrations in Streams*, compiled bacteria data and produced maps of locations with high bacteria concentrations in the summer. The report recommended that:

- A Phase 2 portion of this project should be proposed and implemented. Before analyzing any of the bacteria datasets compiled for this study, additional stream environmental parameters, including streamflow, total suspended solids (TSS), and substrate type need to be measured.
- As part of the Phase 2 project, the annotated bibliography should be reviewed and results from bacterial studies conducted in the Pacific Northwest and other regions of the U.S. compared and analyzed.

The goal of this project is to complete the Phase 2 recommendations outlined in the report.

Project Description

Why Is This Study Being Done?

Ecology is conducting this study to research the role that streambed sediments and other factors play in contributing to high summer bacteria concentrations in South Puget Sound streams.

High bacteria concentrations in rivers and streams indicate the potential presence of harmful pathogens that pose a public health risk to the people that recreate in rivers and streams. In addition, these high bacteria streams often drain to marine waterbodies with public swimming beaches or shellfish harvesting areas. Elevated pathogen levels in the water can accumulate in shellfish tissue, making them unsafe to eat.

Under certain conditions, bacteria that have been deposited in stream sediments can:

- Survive longer than those suspended in the water column.
- Re-suspend in the water column when disturbed.
- In some cases even multiply in the sediment.

Currently, little information is available as to how sediment bacteria affect bacteria levels in Washington streams. This study aims to better characterize that relationship.

The study will also serve as a framework for future Ecology sampling of sediment bacteria. Part of the project will be the development of an EAP standard operating procedure (SOP) for collecting sediment bacteria samples.

Additionally, most of the study locations are waterbodies in a Water Quality Assessment (WQA) Category (see next section) that classifies them as having violated Washington State surface water quality standards for fecal coliform (Categories 4a, 4b, and 5). The study data will provide useful information about the current status of these waterbodies and the likelihood of sediment bacteria as a potential source.

The Water Quality Assessment (WQA) and the 303(d) List

Every two years, states are required to prepare a list of waterbodies that do not meet water quality standards. This list is called the Clean Water Act Section 303(d) list. In Washington State, this list is part of the WQA process.

To develop the WQA, Ecology compiles its own water quality data along with data from local, state, and federal governments, tribes, industries, and citizen monitoring groups. All data in this WQA are reviewed to ensure that they were collected using appropriate scientific methods before they are used to develop the assessment. The list of waters that do not meet standards [the 303(d) list] is the Category 5 part of the larger assessment.

The Five Categories designate water quality as follows:

Category 1 – Meets standards for parameter(s) for which it has been tested.

Category 2 – Waters of concern.

Category 3 – Waters with no data or insufficient data available.

Category 4 – Polluted waters that do not require a Total Maximum Daily Load (TMDL) because:

4a. – Have an approved TMDL and it is being implemented.
4b. – Have a pollution control program in place that should solve the problem.
4c. – Are impaired by a non-pollutant such as low water flow, dams, culverts.

Category 5 – Polluted waters that require a TMDL – the 303(d) list.

Further information is available at Ecology's Water Quality Assessment website: www.ecy.wa.gov/programs/wq/303d.

Project Overview

EAP project staff will collect sediment and water quality data from four streams in Thurston, Mason, and Kitsap counties. EAP will conduct ten sampling events from June to September 2010. Field data collection parameters will include:

- In the water column
  - *In situ* streamflow, temperature, and conductivity measurements.
  - Continuous temperature measurements.
  - Fecal coliform bacteria (FC), turbidity, TSS, and dissolved oxygen samples.
  - Samples to determine the percentage of FC bound to suspended solids.

- In the streambed sediments
  - FC samples.
  - Total organic carbon (TOC) samples.
  - Continuous temperature measurements.

The project manager will compile data, analyze for relationships between variables (parameters), and compare to results from the Phase 1 literature review and other sediment bacteria studies. The project manager will then prepare a short technical report, with study results and discussion, which will be posted on Ecology's website.

Organization and Schedule

The following people are involved in this project. All are employees of the Washington State Department of Ecology (Table 1). Table 2 shows the schedule for completing the project work activities.

Table 1. Organization of project staff and responsibilities.

Staff (all are EAP except client)	Title	Responsibilities
Lydia Wagner Water Quality Program Southwest Regional Office Phone: (360) 407-6329	EAP Client	Clarifies scopes of the project. Provides internal review of the QAPP and approves the final QAPP.
Nuri Mathieu Direct Studies Unit Western Operations Section Phone: (360) 407-7359	Project Manager And Principal Investigator	Writes the QAPP. Oversees field sampling and transportation of samples to the laboratory. Conducts QA review of data, analyzes and interprets data, and enters data into EIM. Writes the draft report and final report.
Markus Von Prause Direct Studies Unit Western Operations Section Phone: (360) 407-7406	Field Assistant	Helps collect samples and records field information.
George Onwumere Direct Studies Unit Western Operations Section Phone: (360) 407-6730	Unit Supervisor for the Project Manager	Provides internal review of the draft QAPP and report. Reviews the project scope, approves the budget, and tracks progress. Approves the final QAPP and report.
Robert F. Cusimano Western Operations Section Phone: (360) 407-6596	Section Manager for the Project Manager	Reviews the draft QAPP, and approves the final QAPP.
Stuart Magoon Manchester Environmental Laboratory Phone: (360) 871-8801	Director	Approves the final QAPP.
William R. Kammin Phone: (360) 407-6964	Ecology Quality Assurance Officer	Reviews the draft QAPP and approves the final QAPP.

EAP – Environmental Assessment Program.
EIM – Environmental Information Management system.
QAPP – Quality Assurance Project Plan.

Table 2. Proposed schedule for completing field and laboratory work, data entry into EIM, and reports.

Field and laboratory work	Due date	Lead staff
Field work completed	September 2010	Nuri Mathieu
Laboratory analyses completed	September 2010	
Environmental Information System (EIM) database		
EIM user study ID	NMat0003	
Product	Due date	Lead staff
EIM data loaded	November 2010	Nuri Mathieu
EIM quality assurance	December 2010	George Onwumere
EIM complete	January 2011	Nuri Mathieu
Final report		
Author lead/Support staff	Nuri Mathieu	
Schedule		
Draft due to supervisor	January 2011	
Draft due to client/peer reviewer	January 2011	
Draft due to external reviewer(s)	February 2011	
Final (all reviews done) due to publications coordinator (Joan)	March 2011	
Final report due on web	April 2011	

Laboratory Budget

Table 3 summarizes the laboratory costs for the study. Table 4 shows the laboratory costs by month. Ecology's Manchester Environmental Laboratory (MEL) will perform all analyses, with the exception of grain size which will be subcontracted out.

Table 3. Laboratory costs.

Parameter	Samples	Repli-cates	Field blanks	Total samples	Cost per sample	Subtotal	Additional labor	Total
FC - MF - water	4	1	2	7	$24	$167		$167
FC - MF - water centrifuged	4	2	0	6	$24	$143	≈$50	$193
FC - MPN - sediment	4	1	n/a[1]	5	$81	$405		$405
FC - MPN - water compare	1	0	0	1	$45	$45		$45
TOC - sediment	4	1	n/a[1]	5	$44	$218		$218
TOC - water	0	0	1	1	$34	$34		$34
TSS - water	4	1	1	7	$11	$69		$69
TSS - water centrifuged	4	2	0	6	$11	$80	≈$120	$189
Turbidity - water	4	1	1	6	$11	$69		$69
							Total cost per survey =	$1,388
							Number of surveys =	10
Grain size - sediment	4	0	0	4	$197	$788[2]		$788
							Total lab costs for study =	$14,666

[1] Field blanks submitted as a water sample.
[2] Sediments analyzed for grain size during initial June survey only, not during all ten surveys.
MF – membrane filter method.
MPN – most probable number method.

Table 4. Laboratory costs by month and fiscal year.

	FY 2010	FY 2011		
Month	Jun-2010	Jul-2010	Aug-2010	Sep-2010
Number of surveys	2	2	4	2
Lab cost	$ 3,136	$ 2,776	$ 5,551	$ 2,776

FY – fiscal year.
Project laboratory costs include a 50% discount for using MEL.

Quality Objectives

Field sampling procedures and laboratory analysis inherently have associated error. Measurement quality objectives (MQOs) state the allowable error for a project. Precision and bias provide measures of data quality and are used to assess agreement with MQOs.

Table 5 outlines analytical methods, expected precision of sample replicates, and method reporting limits and/or resolution. The targets for analytical precision of laboratory analyses are based on historical performance by MEL for environmental samples taken around the state by EAP (Mathieu, 2006). The reporting limits of the methods listed in the table are appropriate for the expected range of results, and the required level of sensitivity to meet project objectives. The laboratory's quality control procedures are documented in the MEL *Lab Users Manual* (MEL, 2008) and *Quality Assurance Manual* (MEL, 2010).

Table 5. Measurement quality objectives for precision in field measurements and laboratory analysis.

Analysis	Method/ equipment	Field replicate MQO (median)	Lab duplicate MQO	Reporting limits and resolution
Field Measurements				
Discharge volume	Marsh McBirney Flow-Mate Flowmeter	10% RSD	n/a	0.01 ft/s
Water temperature[1]	YSI®	+/- 0.2° C	n/a	0.01° C
Specific conductivity	YSI®	5% RSD	n/a	0.1 umhos/cm
Dissolved oxygen[1]	SM 4500OC	+/- 0.2 mg/L	n/a	0.1 mg/L
Continuous temperature	Hobo Water Temp Pro	±0.2°C at 0 to 50°C	n/a	0.02° C
Laboratory Analyses				
FC - MPN	MPN 9221 E2	50% of replicate pairs < 50% RSD 90% of replicate pairs <100% RSD[2]	40% RPD	1.8 MPN/100 mL
FC - MF	SM 9222D	50% of replicate pairs < 20% RSD 90% of replicate pairs <50% RSD[2]	40% RPD	1 cfu/100 mL
FC - MF - centrifuged	SM 9222D	50% of replicate pairs < 50% RSD 90% of replicate pairs < 90% RSD[2]	40% RPD	1 cfu/100 mL
Turbidity	SM 2130	15% RSD[3]	20% RPD	0.5 NTU
TSS	SM 2540D	15% RSD[3]	20% RPD	1 mg/L

[1] As units of measurement, not percentages.
[2] Replicate results with a mean of less than or equal to 20 cfu/100 mL will be evaluated separately.
[3] Replicate results with a mean of less than or equal to 5X the reporting limit will be evaluated separately.
MPN – most probable number method
MF – membrane filter method
RSD – relative standard deviation.
RPD – relative percent difference.

Precision

Precision is defined as the measure of variability in the results of replicate measurements due to random error. Random error is imparted by the variation in concentrations of samples from the environment as well as other introduced sources of variation (e.g., field and laboratory procedures). Precision for replicates will be expressed as percent relative standard deviation (%RSD) and assessed following the MQOs outlined in Table 5.

Bias

Bias is defined as the difference between the population mean and true value of the parameter being measured. Field and laboratory quality control procedures, such as blanks, check standards, and spiked samples, provide a measure of any bias affecting measurement procedures. Field staff will minimize bias in field measurements and samples by strictly following measurement, sampling, and handling protocols

EAP staff will assess bias in field samples by submitting field blanks. Field staff will prepare blanks in the field by:

- For FC and TOC sediment samples, rinsing a sterile set of sampling equipment with sterile deionized water and collecting the rinse water in either an autoclaved 250 mL sampling bottle or 60 mL TOC bottle.
- For TSS, turbidity, and FC water samples, filling the bottles directly with sterile deionized water.
- Handling and transporting the samples to MEL in the same manner that the rest of the samples are processed.

For field measurements, EAP staff will:

- Minimize bias in the YSI® probe field measurements by pre-calibrating before each run.
- Assess any potential bias in probe measurements by post-checking the instrument.
- Calibrate the probe for conductivity before each run and post-check the probe afterwards using National Institute of Standards and Technology (NIST) certified conductivity standards.
- Check the probe's temperature readings before and after each run using an NIST-certified thermometer.

Table 6 contains the data quality bias objectives for post-check values.

Table 6. Measurement quality objectives for YSI® post checks.

Parameter	Units	Accept	Qualify	Reject
Conductivity*	µS/cm	≤ ± 5%	> ± 5% and ≤ ± 15%	> ± 15%
Temperature	Degrees Celsius	≤ ± 0.2	> ± 0.2 and < or = ± 0.5	> ± 0.5

* Criteria expressed as a percentage of readings; for example, buffer = 100.2 µS/cm and Hydrolab = 98.7 µS/cm; (100.2-98.7)/100.2 = 1.49% variation, which would fall into the acceptable data criteria of less than 5%.

Comparability

The membrane filter (MF) method is EAP's preferred method for FC water samples. The MF method has a faster results turnaround time, allows MEL to analyze a greater number of samples per day, and has shown to be a more precise method in past TMDL studies (Mathieu, 2006c). FC sediment samples must be analyzed using the most probable number (MPN) method, so Ecology will collect water column MPN samples for 20% of the samples to assess comparability of the MF and MPN methods at each site. This will also provide for comparison to the Washington State Department of Health shellfish growing area sampling data, where FC water samples are analyzed using an MPN method.

Comparability to previously collected data will be established by strictly following EAP protocols and adhering to data quality criteria.

Representativeness

FC bacteria values are known to be highly variable over time and space. Sampling variability can be somewhat controlled by strictly following standard procedures and collecting quality control samples, but natural spatial and temporal variability can contribute greatly to the overall variability in the parameter value. Resources limit the number of samples that can be taken at one site spatially or over various intervals of time. Laboratory and field errors are further expanded by estimate errors in seasonal loading calculations and modeling estimates.

Ecology designed the sampling regime to provide a high frequency of sampling events over a short time period to adequately characterize FC temporal patterns at each site. Additionally, the three sediment sub-samples used for the composite sample will be spread out across the sampling transect to increase the representativeness of the sample.

A lower limit of five samples per season per site is required for comparison to state criteria, which will easily be met with the current sampling design (10 samples per site).

WAC 173-201A states:
When averaging bacteria sample data for comparison to the geometric mean criteria, it is preferable to average by season and include five or more data collection events within each period…and [the period of averaging] should have sample collection dates well distributed throughout the reporting period.

Completeness

EPA has defined completeness as a measure of the amount of valid data needed to be obtained from a measurement system (Lombard and Kirchmer, 2004). The goal for this study is to correctly collect and analyze a minimum of 95% of the samples for all sites. Problems occasionally arise during sample collection that cannot be controlled, including flooding, stagnant or no flow during dry periods, or samples damaged in transit.

Sampling (Experimental) Design

Sampling Locations

Ecology staff narrowed the potential sampling locations down to 15 sites based on the bacteria maps generated during Phase 1, input from project stakeholders, and additional data analysis for FC loading (Table 7).

Table 7. Seasonal patterns in fecal coliform concentrations and loads in 15 South Puget Sound streams.

Creek Name	n =	Data Collection Period	FC Dry Season Geomean (cfu/100 mL)	Average Dry Season FC Load (billion cfu/day)	Average Wet Season FC Load (billion cfu/day)	Ratio of Dry to Wet Season FC Load
Henderson Inlet						
Woodland Creek	12	2002-2004	175	68.51	144.20	0.5
Jorgenson Creek	12	2002-2004	412	8.15	9.76	0.8
Eagle Creek	12	2002-2004	204	8.44	5.05	**1.7**
Oakland Bay						
Deer Creek[1]	14	2004-2005	44	24.68	11.98	**2.1**
Cranberry Creek[1]	15	2004-2005	41	no flow data	no flow data	no flow data
Uncle John Creek[1]	15	2004-2005	274	6.15	6.36	1.0
Little Skookum, Totten, and Eld Inlets						
Kennedy Creek[1]	56	2005-2009	45	14.44	16.85	0.9
Schneider Creek[1]	56	2005-2009	105	7.13	13.85	0.5
McLane Creek	123	1999-2002	290	147.13	129.48	**1.1**
Deschutes Watershed						
Chambers Creek	56	2005-2009	71	6.50	7.06	0.9
Nisqually Watershed						
McAllister Creek	56	2005-2009	125	no flow data	no flow data	no flow data
Kitsap County						
Burley Creek[1]	47	2002-2006	258	180.76	92.36	**2.0**
Lower Hood Canal Streams						
Big Bend Creek	14	2004-2005	136	7.48	0.89	**8.4**
Happy Hollow Creek	14	2004-2005	73	2.05	0.47	**4.4**
Twanoh Creek	14	2004-2005	92	8.30	0.18	**45.2**

Shaded bold cells indicate sites where the FC load is larger in the dry season than during the wet season.
Dry season= June to September.
Wet season= October to May.
[1] Excludes large wet season storm events.

Table 8 lists the four sampling locations chosen for the Phase 2 study. Figure 1 depicts the sampling locations and associated drainage basins.

Table 8. Sampling locations.

Creek Name	EIM User Location ID	Site Description	Latitude °N	Longitude °W
Deer Creek	OAK DEE 0	Near mouth off E Gosser Rd.	47.26076	123.00902
Kennedy Creek	SPS KENN CK	At Old Olympic Highway near mouth	47.09507	123.09127
McLane Creek	14MCLANEMC1.5	At Delphi Rd.; just upstream of Swift Creek	47.03122	122.99111
Burley Creek	KCHD-BL01	Burley Creek at Spruce Rd. bridge	47.41445	122.63132

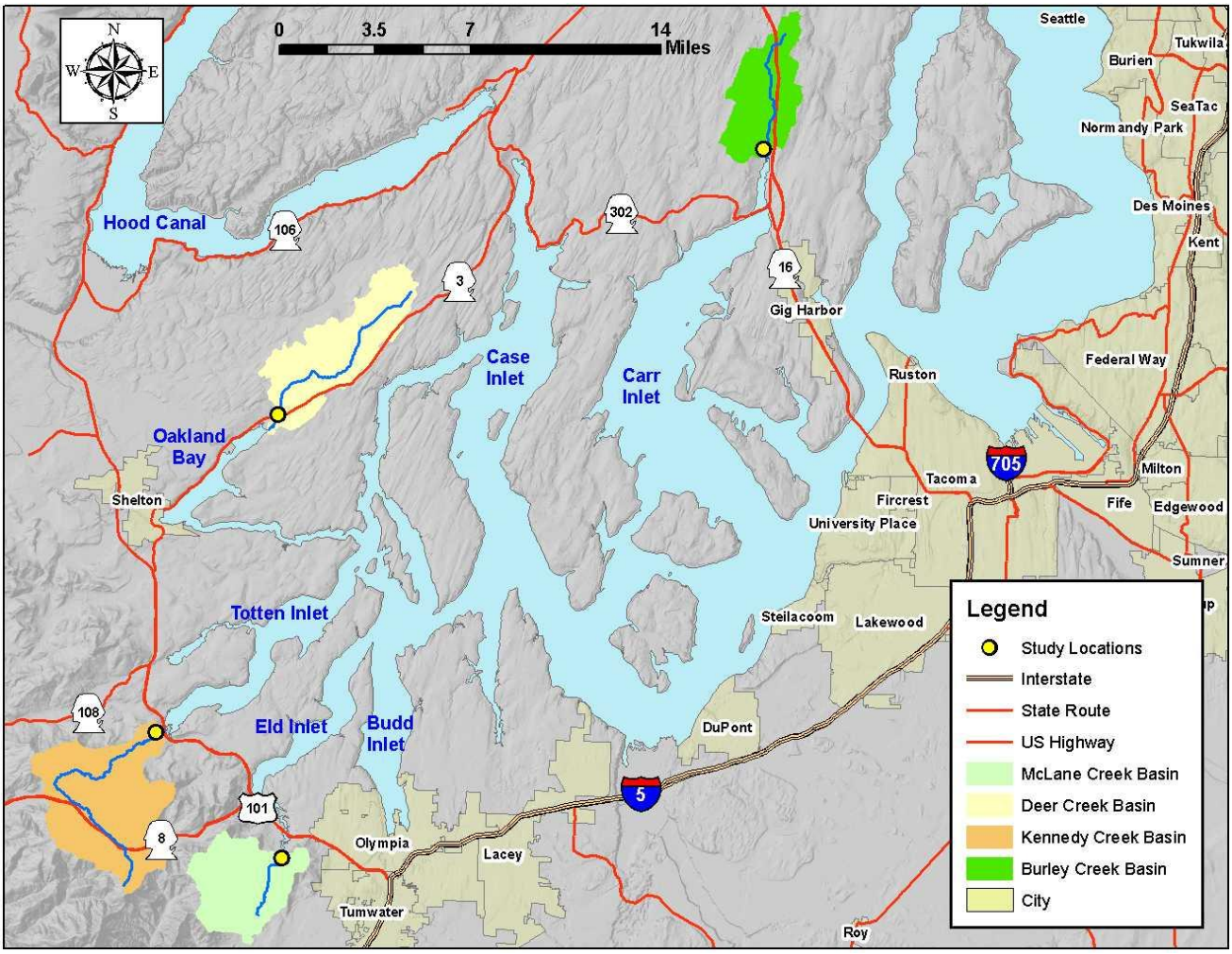

Figure 1. Study locations and associated drainage basins for the *Phase 2: High Summer Bacteria Concentrations in Streams* study.

Ecology staff made final selection of sites based on:

- Deer Creek
  - Clear pattern of higher FC concentrations and loads during summer months.
  - While the geometric mean (44 cfu/100 mL) is not very high compared to other sites, the creek mouth is located very close to a sensitive shellfish harvesting area in Oakland Bay.
  - The current WQA status is Category 1; however, the study data will be useful to:
    - TMDL implementation efforts for Oakland Bay.
    - Squaxin Island Tribe's sediment bacteria studies being conducted in Oakland Bay at the Washington State Department of Health Station 614.
- Kennedy Creek
  - Clear pattern of higher concentrations in summer months.
  - While the geometric mean (45 cfu/100 mL) is not very high compared to other sites, there are no obvious sources of FC in the watershed that might contribute to high summer bacteria concentrations.
  - The current WQA status is Category 4a and the study data will be useful to TMDL implementation efforts.
- McLane Creek
  - Clear pattern of higher FC concentrations and loads during summer months.
  - Magnitude of summer FC loading is greater than other sites investigated.
  - The current WQA status is Category 4a and the study data will be useful to TMDL implementation efforts.
- Burley Creek
  - Clear pattern of higher FC concentrations and loads during summer months.
  - Magnitude of summer FC loading is greater than other sites investigated.
  - Drains to a restricted shellfish harvesting area in Burley Lagoon.
  - The current WQA status is Category 4b and the study data will be useful to Kitsap County's pollution control program for Burley Creek.

Both Twanoh and Happy Hollow Creeks exhibited abnormally high FC loading in the summer (compared to the wet season); however, in each case a large failing septic system was discovered adjacent to the creek and has since been repaired (Mason, 2008). Both sites are located in popular summer recreation areas where increased use of the facilities during the summer likely resulted in larger bacterial loading from the failing systems during summer months.

At Big Bend Creek, several on-site septic systems are located adjacent to the sampling location near the mouth. In addition, the stream is only a little over a mile in length and had a smaller average dry season FC load than the selected sampling locations.

Eagle Creek had a large dry to wet season loading ratio; however, this was due to a single sample collected in June 2003 that resulted in a very large FC load and skewed the average dry season load.

Field Sampling Dates

Ecology field staff will conduct 10 sampling events from June to September of 2010 (Table 9).

Table 9. Sampling dates.

Day of Week	Date (in 2010)
Monday	June 14
	June 28
Tuesday	July 6
Monday	July 19
	August 2
	August 16
	August 23
	August 30
	September 20
	September 27

Study Design

Before sampling begins, field staff will:

- Visit all sampling locations to determine appropriate sampling reaches.
- Set up a permanent flow measurement transect downstream from the stream reach where water and sediment sampling occurs.
- Install continuous temperature monitoring instruments in the sediment and water column at each site.
- Take pictures of sampling reach and record observations about site characteristics.

For each site during a given sampling event field staff will:

1. Place a temperature/conductivity probe in the water just upstream of the flow transect.
2. Measure streamflow at the flow transect.
3. Record temperature and conductivity measurements.
4. Collect water column samples for dissolved oxygen, FC, turbidity, TSS, and additional FC and TSS centrifuge samples.
5. Collect sediment samples for FC and TOC.
6. Record observations of land use, recreational use, weather, etc.

During the first sampling event, field staff will collect water column and sediment samples immediately upstream of the flow transect. Field staff will flag the sampling transect and then, during the next sampling event, collect water and sediment samples immediately upstream of the flagged sampling location from the previous event. During each subsequent sampling event, the water and sediment sampling transect will move progressively upstream to avoid sampling sediments that were disturbed during a previous field survey.

During the initial sampling event, field staff will also split the composite sediment sample and have one half of the split analyzed for grain size and composition.

For sediment samples field staff will:

1. Collect three sediment samples from the sampling transect.
2. Immediately composite the samples.
3. Split the composite sample into two separate containers, one for FC analysis and one for TOC analysis.

Field staff will deploy two continuous temperature data loggers (thermistors) at each site: one to measure water temperature and another to measure sediment temperature. The thermistors will measure temperature at 30-minute intervals throughout the course of the project. Instream thermistors are deployed in the thalweg of a stream such that they are suspended off the stream bottom and in a well-mixed portion of the stream, typically in riffles or swift glides. Sediment thermistors will be buried just beneath the sediment surface.

Sampling and Measurement Procedures

Field Procedures

Field sampling and measurement protocols will follow SOPs developed by EAP for TMDL development (Table 10). Field measurements for conductivity and temperature will be collected using a calibrated YSI® probe. A dissolved oxygen sample will be collected by hand using a displacement sampler and analyzed using the Winkler titration method (APHA, 1998; Ward, 2007). Field staff will measure instantaneous flows with a Marsh McBirney Flow-mate meter. Field staff will use Hobo® Water Temp Pro V2 (Version 2) thermistors to record continuous temperature measurements.

Table 10. Sampling and measurement methods and protocols.

Parameter	Measurement/ Sample Type	Laboratory Method	Field Protocol Number
FC - MF	Grab sample	SM 9222 D	EAP012 (Mathieu, 2006a); EAP015 (Joy, 2006)
FC - MPN	Grab sample	SM 9221 E2	EAP012 (Mathieu, 2006a); EAP015 (Joy, 2006)
FC - MF - centrifuge	Grab sample	Characklis et al., 2005	EAP012 (Mathieu, 2006a); EAP015 (Joy, 2006)
FC - MPN - sediment	Composite sample	SM 9221 E	EAP069 (Mathieu, 2010 – draft)
TOC: Sediment	Composite sample	(Puget, 1986) (Puget, 1997)	EAP069 (Mathieu, 2010 – draft)
TSS	Grab sample	SM 2540 D	EAP015 (Joy, 2006)
Turbidity	Grab sample	SM 2130	EAP015 (Joy, 2006)
TSS - centrifuge	Grab sample	Characklis et al., 2005	EAP015 (Joy, 2006)
Dissolved oxygen	Displacement sample	SM 4500 OC	EAP035 (Mathieu, 2006b)
Continuous temperature	Hobo® Water Temp Pro V2	n/a	EAP044 (Bilhimer and Stohr, 2009)
Temperature and conductivity	YSI® probe	n/a	EAP010 (Ahmed, 2006)
Flow	Instantaneous	n/a	EAP024 (Sullivan, 2007)

Field staff will collect grab samples directly into pre-cleaned/sterilized containers supplied by MEL and described in the MEL *Lab Users Manual* (2008). Table 11 lists the sample parameters, containers, volumes, preservation requirements, and holding times. Field staff will store samples for laboratory analysis on ice and deliver to MEL within 24 hours of collection via either the Ecology courier or direct drop-off after sampling.

Table 11. Containers, preservation requirements, and holding times for samples collected (MEL, 2008).

Parameter	Sample Matrix	Container	Preservative	Holding Time
FC[1]	Surface water	250 or 500 mL glass/poly autoclaved	Cool to ≤10°C	24 hours
FC	Sediment	Sterile specimen cup or Whirlpak bags	Cool to ≤10°C	24 hours
TSS	Surface water	1000 mL w/m poly bottle	Cool to ≤6°C	7 days
TSS - centrifuge		4 x 250 mL w/m poly bottle	Cool to ≤6°C	7 days
TOC	Sediment	2 oz glass jar[2]	Cool to ≤6°C	14 days
Turbidity	Surface water	500 mL w/m poly bottle	Cool to ≤6°C	48 hours
Grain size	Sediment	8 oz plastic jar	Cool to ≤6°C	6 months

[1] Same for both centrifuged and un-centrifuged samples.
[2] Organic- free with Teflon- lined lids and Certificate of Analysis.

Ecology will collect replicate field samples, in a side-by-side manner, for 20% of FC samples and 10% of TSS and TOC samples to assess field and lab variability.

Field staff will check temperature monitoring stations monthly to make field measurements and to clear accumulated debris away from the instruments. Documentation of the temperature monitoring stations will include:

- GPS coordinates and a sketch of the site (during installation only).
- Depth of the instream temperature instrument (TI) under the water surface and height off the stream bottom.
- Stream temperature.
- Serial number of each instrument and the action taken with the instrument (e.g., downloaded data, replaced TI, or noted any movement of the TI location to keep it submerged in the stream).
- The date and time before the dataloggers are installed or downloaded, and the date and time after they have been returned to their location. All timepieces and PC clocks should be synchronized to the atomic clock using Pacific Daylight Savings Time. Pacific Standard Time will be reported if instruments are still in place during the time change.

Laboratory Procedures

MEL will follow their standard analytical methods following the MEL *Lab Users Manual* (MEL, 2008).

MEL will perform centrifuge analysis for FC bacteria and TSS following a method adapted from Characklis et al., 2005. A summary of the procedure is provided in Appendix A.

Quality Control Procedures

Total variation from field sampling and analytical processes will be assessed by collecting and analyzing replicate samples. Sample precision will be assessed by collecting replicates for approximately 10-20% of samples in each survey. MEL routinely duplicates sample analyses in the laboratory to determine the presence of bias in analytical methods. The difference between field variability and laboratory variability is an estimate of the sample field variability.

Field

Field sampling and measurements will follow quality control protocols described in Ecology's field sampling protocols (Table 8). If any of these quality control procedures are not met, the associated results will be qualified and used with caution or not used at all.

Prior to each sampling event, MEL will sterilize (via autoclave at 120°C):

- All sample containers used for water and sediment samples.
- The tools used to collect and composite the samples (one set for each site, plus one backup set).

Using a separate set of sterile, autoclaved equipment at each site will avoid complications associated with field sterilization of sampling equipment. Field sterilization requires the equipment to be immersed in a hypochlorite (bleach) solution for an extended period of time and then a subsequent immersion in a sodium thiosulfate solution to neutralize the bleach. Any residual bleach that was not properly neutralized could inadvertently kill off bacteria in the field sample.

The Hobo Water Temp Pro V2 instruments will have a calibration check both pre- and post-study in accordance with Ecology Temperature Monitoring Protocols (Bilhimer and Stohr, 2009). This check will document instrument bias or performance at representative temperatures. A NIST certified reference thermometer will be used for the calibration check.

A datalogger that fails pre-study calibration check will not be used. If the temperature datalogger fails the post-study calibration check, then the actual measured value will be reported along with its degree of accuracy based on the calibration check results. As a result, these data may be qualified or rejected.

Variation for field sampling of instream temperatures and potential thermal stratification will be addressed with a field check of stream temperature at all monitoring sites upon deployment, during regular site visits and during instrument retrieval at the end of the study period. Air temperature data (obtained from local weather stations) and instream temperature data for each site will be compared to determine if the instream thermistor was exposed to the air due to stream stage falling below the installed depth of the stream thermistor.

Laboratory

All samples will be analyzed at MEL. The laboratory's quality control procedures are documented in the MEL *Lab User's Manual* (MEL, 2008) and *Quality Assurance Manual* (MEL, 2010). MEL will follow standard quality control procedures (MEL, 2010).

Data Management Procedures

Field measurement data will be entered into a field book with waterproof paper in the field and then entered into Excel® spreadsheets (Microsoft, 2007) as soon as practical after returning from the field. This database will be used for preliminary analysis and to create a table to upload data into Ecology's Environmental Information Management (EIM) System.

Sample result data received from MEL by Ecology's Laboratory Information Management System (LIMS) will be exported prior to entry into EIM and added to a cumulative spreadsheet for laboratory results. This spreadsheet will be used to informally review and analyze data during the course of the project.

An EIM user study (NMat0003) has been created for this study and all monitoring data will be available via the internet once the project data has been validated. The URL address for this geospatial database is: www.ecy.wa.gov/eim/. All data will be uploaded to EIM by the EIM data engineer once it has been reviewed for quality assurance and finalized.

All spreadsheet files, paper field notes, and GIS products created as part of the data analysis will be kept with the project data files.

Audits and Reports

The project manager will be responsible for submitting a short technical report to the client for this project according to the project schedule.

Data Verification and Validation

Data Verification

MEL will provide verification for laboratory generated data. Data reduction, review, and reporting will follow the procedures outlined in the MEL *Quality Assurance Manual* (MEL, 2006). Lab results will be checked for missing or improbable data. Variability in lab duplicates will be quantified using the procedures outlined in the MEL *Quality Assurance Manual* (MEL, 2006). Any estimated results will be qualified and their use restricted as appropriate. A standard case narrative of laboratory quality assurance/quality control results will be sent to the project manager for each set of samples.

Field notebooks will be checked for missing or improbable measurements before leaving each site. The Excel® Workbook file containing field data will be labeled "DRAFT" until data verification and validity are completed. Data entry will be checked against the field notebook data for errors and omissions.

Field replicate sample results will be compared to quality objectives in Table 3. Data requiring additional qualifiers will be reviewed and verified by the project manager.

Data Validation

The project manager will validate data received from LIMS by:

- Checking for omissions against the "Request for Analysis" forms.
- Checking result values against expected range of results and data from previous surveys.

After data verification is complete, all field, laboratory, and flow data will be entered into Ecology's EIM system. An independent data reviewer will validate the EIM data by checking for errors following standard EAP protocols.

Once the EIM data has been validated, the project manager will compile all project data in a data summary report. Internal (within Ecology) and external (project stakeholders) reviewers will provide validation of the report.

Data Quality (Usability) Assessment

The project manager will verify that all measurement and data quality objectives have been met for each monitoring station. If the objectives have not been met (such as percent RSD for sample replicates exceeds the MQO), then the project manager will decide how to qualify the data and how it should be used in the analysis or whether it should be rejected.

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Appendices

Appendix A. Summary of Centrifuged Analyses

Summary of Centrifuged Fecal Coliform (FC) Procedure

1. For each sampling event, MEL will receive five to six 250 mL FC samples with the parameter labeled as "FCMF-centrifuge"

   Note: actual sample volume will be close to 200 mL.

2. Each 250 mL sample bottle will be placed in a centrifuge and spun at 2000 rpm for 10 minutes.

   Note: Each corresponding non-centrifuged FC sample will be left out (not refrigerated) during the centrifuge process to address comparability issues between the handling of centrifuged and non-centrifuged samples.

3. Following centrifugation, approximately 100-150 mL of the supernatant will be removed from the top of the sample using a vacuum flask and hose assembly connected to a pipette.

   Note: the aperture of the pipette must be at least 0.1 mm in diameter (100 µm).

4. The 100-150 mL aliquot of supernatant will then immediately be processed as a normal FC membrane filter (FCMF) sample following Standard Methods (SM) 9222D (APHA, 1998).

Outline of Centrifuged TSS Procedure (no microbiology analysis involved)

1. Field staff will collect one 1000 mL sample and four 250 mL samples from each site.
2. MEL will analyze the 1000 mL sample as a normal TSS sample following SM 2540D.
3. MEL will receive the four 250 mL samples (per site) with the parameter labeled as "TSS-centrifuge."
4. MEL will centrifuge the four 250 mL sample together at 2000 rpm for 10 minutes.
5. Following centrifugation, approximately 175-200 mL of the supernatant will be removed from the top of each 250 mL sample (in the same manner described above) and all four will be composited into a new 1000 mL container (approximately 700-800 mL of sample).
6. The composited sample will then be analyzed as a normal TSS sample following SM 2540D.

Appendix B. Glossary, Acronyms, and Abbreviations

Glossary

Clean Water Act: A federal act passed in 1972 that contains provisions to restore and maintain the quality of the nation's waters. Section 303(d) of the Clean Water Act establishes the TMDL program.

Conductivity: A measure of water's ability to conduct an electrical current. Conductivity is related to the concentration and charge of dissolved ions in water.

Dissolved oxygen: A measure of the amount of oxygen dissolved in water.

Fecal coliform (FC): That portion of the coliform group of bacteria which is present in intestinal tracts and feces of warm-blooded animals as detected by the product of acid or gas from lactose in a suitable culture medium within 24 hours at 44.5 plus or minus 0.2 degrees Celsius. FC are "indicator" organisms that suggest the possible presence of disease-causing organisms. Concentrations are measured in colony forming units per 100 milliliters of water (cfu/100 mL).

Geometric mean: A mathematical expression of the central tendency (an average) of multiple sample values. A geometric mean, unlike an arithmetic mean, tends to dampen the effect of very high or low values, which might bias the mean if a straight average (arithmetic mean) were calculated. This is helpful when analyzing bacteria concentrations, because levels may vary anywhere from 10 to 10,000 fold over a given period. The calculation is performed by either: (1) taking the nth root of a product of n factors, or (2) taking the antilogarithm of the arithmetic mean of the logarithms of the individual values.

Parameter: A physical chemical or biological property whose values determine environmental characteristics or behavior.

Pathogen: Disease-causing microorganisms such as bacteria, protozoa, viruses.

Pollution: Such contamination, or other alteration of the physical, chemical, or biological properties, of any waters of the state. This includes change in temperature, taste, color, turbidity, or odor of the waters. It also includes discharge of any liquid, gaseous, solid, radioactive, or other substance into any waters of the state. This definition assumes that these changes will, or are likely to, create a nuisance or render such waters harmful, detrimental, or injurious to (1) public health, safety, or welfare, or (2) domestic, commercial, industrial, agricultural, recreational, or other legitimate beneficial uses, or (3) livestock, wild animals, birds, fish, or other aquatic life.

Reach: A specific portion or segment of a stream.

Relative percent difference (RPD): The absolute value of the difference between duplicates expressed as a percent of the duplicate mean.

Streamflow: Discharge of water in a surface stream (river or creek).

Total Maximum Daily Load (TMDL): A distribution of a substance in a waterbody designed to protect it from exceeding water quality standards. A TMDL is equal to the sum of all of the following: (1) individual wasteload allocations for point sources, (2) the load allocations for nonpoint sources, (3) the contribution of natural sources, and (4) a margin of safety to allow for uncertainty in the wasteload determination. A reserve for future growth is also generally provided.

Total suspended solids (TSS): Portion of solids retained by a filter.

Turbidity: A measure of water clarity. High levels of turbidity can have a negative impact on aquatic life.

Watershed: A drainage area or basin in which all land and water areas drain or flow toward a central collector such as a stream, river, or lake at a lower elevation.

303(d) list: Section 303(d) of the federal Clean Water Act requires Washington State to periodically prepare a list of all surface waters in the state for which beneficial uses of the water – such as for drinking, recreation, aquatic habitat, and industrial use – are impaired by pollutants. These are water quality limited estuaries, lakes, and streams that fall short of state surface water quality standards and are not expected to improve within the next two years.

Acronyms and Abbreviations

Following are acronyms and abbreviations used frequently in this report.

EAP	Environmental Assessment Program (Ecology)
FC	(See Glossary above)
e.g.	For example
Ecology	Washington State Department of Ecology
EIM	Environmental Information Management database
EPA	U.S. Environmental Protection Agency
et al.	And others
GIS	Geographic Information System software
MEL	Manchester Environmental Laboratory
MF	Membrane filter
MPN	Most probable number
MQO	Measurement quality objective
RPD	Relative percent difference
RSD	Relative standard deviation
SOP	Standard operating procedure
TMDL	(See Glossary above)
TOC	Total organic carbon
TSS	(See Glossary above)
WAC	Washington Administrative Code
WQA	Water Quality Assessment

WRIA	Water Resources Inventory Area
WWTP	Wastewater treatment plant

Units of Measurement

°C	degrees centigrade
cfs	cubic feet per second
dw	dry weight
ft	feet
g	gram, a unit of mass
kg	kilograms, a unit of mass equal to 1,000 grams.
m	meter
mg	milligram
mgd	million gallons per day
mg/Kg	milligrams per kilogram (parts per million)
mg/L	milligrams per liter (parts per million)
mL	milliliters
mm	millimeter
NTU	nephelometric turbidity units
s.u.	standard units
µg/L	micrograms per liter (parts per billion)
µS/cm	microsiemens per centimeter, a unit of conductivity