

Washington Department of FISH and WILDLIFE



# Quality Assurance Project Plan for Status and Trends Monitoring of Marine Nearshore Mussels

# for the Regional Stormwater Monitoring Program and Pierce County

June 2015 WDFW Publication no. FPT 15-04

### **Publication information**

This Quality Assurance Project Plan is available on the Department of Ecology's Regional Stormwater Monitoring Program (RSMP) website for National Pollutant Discharge Elimination System (NPDES) municipal stormwater permittees at <a href="http://www.ecy.wa.gov/programs/wg/stormwater/municipal/status.html">http://www.ecy.wa.gov/programs/wg/stormwater/municipal/status.html</a>.

Data for the RSMP will be available on Ecology's Environmental Information Management (EIM) website at <u>www.ecy.wa.gov/eim/index.htm</u>. Search Study ID, RSMP\_PMNM2015. Data from Pierce County will be under Study ID RSMP\_PC\_PMNM2015.

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# Introduction

# Development of a Stormwater Monitoring Strategy for the Puget Sound Region

The Stormwater Work Group (SWG) is a coalition of federal, tribal, state, and local governments; business, environmental, and agricultural entities; and academic researchers. All SWG members have interests and a stake in the Puget Sound watershed. The SWG was convened by the Puget Sound Partnership (PSP) and the Washington State Department of Ecology (Ecology) in October 2008 to develop a regional stormwater monitoring strategy and to recommend monitoring requirements in National Pollutant Discharge Elimination System (NPDES) stormwater permits issued by Ecology. In 2012, the SWG became the first "topical workgroup" included in the Puget Sound Ecosystem Monitoring Program (PSEMP), an organization designed to coordinate regional monitoring efforts to assist in providing information to support Puget Sound recovery efforts.

An overall strategy for stormwater monitoring and assessment for the Puget Sound region was developed by the SWG in 2010 (SWG, 2010a). This strategy, summarized in Appendix A, included recommendations for status and trends monitoring in small streams and in the Puget Sound nearshore, with a focus on an integrated approach to quantify stormwater pollutant impacts in Puget Sound, and providing information to efficiently, effectively, and adaptively manage stormwater to reduce harm to the ecosystem.

The SWG also recommended a specific NPDES municipal permittee-funded plan for monitoring the effects of stormwater under the permits in the Puget Sound region (SWG, 2010b). The resulting program, a subset of the overall strategy, is called the Regional Stormwater Monitoring Program (RSMP). Specifically, the RSMP includes status and trends monitoring of water quality and "watershed health" (physical habitat, sediment chemistry, and biological communities) in small streams in the Puget Sound lowlands; and of sediment quality, bacteria, and contaminants in mussels in the marine nearshore of Puget Sound. The RSMP status and trends monitoring follows a probabilistic sample design (SWG, 2010a) such that data gathered can be summarized across the Puget ecoregion. Additional information about the experimental design, the goals, and the objectives for status and trends and other monitoring in the RSMP can be found in Appendix A of this report, in SWG (2010a and 2010b), and at the RSMP website http://www.ecy.wa.gov/programs/wq/stormwater/municipal/rsmp/rsmp.html).

## Scope of this Quality Assurance Project Plan

This Quality Assurance Project Plan (QAPP) defines the status and trends mussel monitoring in the Puget Sound nearshore that will be conducted by the Washington Department of Fish and Wildlife (WDFW) staff and volunteers for the RSMP. In addition, this QAPP defines the mussel monitoring that will be conducted by Pierce County as part of their NPDES permit Special Condition S8.B obligation. Pierce County selected permit option 2 of S8.B for status and trends monitoring and therefore will conduct mussel monitoring at a jurisdictionally intensified scale. This QAPP defines the site confirmation and sampling protocols that WDFW and Pierce County will follow while conducting mussel monitoring, as well as the data and reports that will be produced to document monitoring results. This QAPP was developed in accordance with Ecology's QAPP guidelines (Lombard and Kirchmer, 2004).

# WDFW, Pierce County and Ecology Roles

WDFW staff and volunteers and Pierce County will conduct monitoring at assigned sites in Puget Sound nearshore areas along their jurisdictions within the period from October 2015 to February 2016. The key completion dates for the required monitoring activities, including site confirmation, field work, and delivery of mussels to the WDFW's Olympia laboratory are summarized in Table 1. Table 2 lists key WDFW and Ecology staff responsible for monitoring activities detailed in this QAPP. Appendix C lists the key Pierce County staff, monitoring responsibilities, and mussel sites.

Due	Item	Description
April 30, 2015	Draft QAPP submitted	WDFW submits draft QAPP to Ecology for review.
May 15, 2015	Revised QAPP	Pierce County submits revised draft QAPP to Ecology.
June 30, 2015	Final QAPP approved	Final QAPP completed and accepted by Ecology.
August 31, 2015	Site selection and verification	WDFW and Pierce County have confirmed all sites to be monitored, including sufficient additional sites to sample if sampling attempted at any of the original sites is unsuccessful. Send site list to RSMP Coordinator.
October 2015	Mussel cages deployed	WDFW and Pierce County deploy mussel cages at the required number of nearshore sites.
February 2016	Mussel cages retrieved and mussels delivered to WDFW	WDFW and Pierce County retrieve mussel cages from the required number of nearshore sites and deliver the mussels, alive on ice, to the WDFW Marine Resources Laboratory in Olympia on the morning following retrieval.
February - March, 2016	Send samples to laboratories.	WDFW submits frozen mussel tissue samples to the RSMP contracted laboratories for chemical analysis.
October - February, 2017 and 2018	Round 2: deploy and retrieve mussels, deliver mussels to WDFW	Pierce County will conduct a second round of mussel monitoring at the same sites sampled in 2016-2017 and delivers mussels to WDFW. WDFW submits frozen mussel tissue samples to the contracted laboratories for chemical analysis.

 Table 1. Key completion dates for QAPP, monitoring activities, and reports for status and trends

 monitoring in the Puget Sound nearshore.

Ecology Staff	Administration of Stormwater Permits and RSMP			
Name, Program, Location	Role	Responsibility		
Brandi Lubliner - WQP Lacey, WA	RSMP Coordinator	Ongoing implementation and administration of RSMP. Reviews and approves completed project deliverables from WDFW's and permittees' monitoring efforts. Coordinate for data QA.		
Chris Montague-Breakwell WQP-SWRO: Lacey, WA	Permit Manager Ecology's contact for stormwater permittees including Pierce County. Reviews monitoring reports for permit compliance.			
Randall Marshall –WQP Lacey, WA	WQP Quality Assurance OfficerDraft template QAPP review and approval.			
WDFW Staff	Administration of Mussel Monitoring			
Name, Program, Location	n Role Responsibility			
Jennifer Lanksbury - WDFW Olympia, WA	Mussel Monitoring Coordinator	RSMP contractor to provide ongoing implementation and administration of mussel monitoring, including laboratory processing of mussels, data review, analysis and final report on the mussel monitoring efforts.		

Table 2. Ecology and WDFW project staff and responsibilities

NWRO - Northwest Regional Office; SWRO - Southwest Regional Office; EIM - Environmental Information Management database; WQP - Water Quality Program; WDFW - Washington Department of Fish and Wildlife

WDFW will coordinate with an aquaculture facility to provide mussels for all the RSMP and Pierce County nearshore monitoring sites. WDFW will contract with analytical laboratories for all mussel tissue chemistry analyses. Pierce County will coordinate their mussel purchase and analysis through WDFW.

WDFW will obtain a <u>Hydraulic Project Approval (HPA)</u>, a <u>Shellfish Transfer Permit</u>, and a Memorandum of Understanding (MOU) with the Washington Department of Natural Resources (DNR) to access <u>State-Owned Aquatic Lands (SOAL)</u> for all RSMP and Pierce County mussel monitoring activities. WDFW staff and volunteers and Pierce County will perform reconnaissance and verification of the RSMP and Pierce County monitoring sites, respectively, and acquire any *other* permits or permissions (outside those listed above) necessary to access their approved sites, including but not limited to permission to access privately-owned, city, county, port, or tribal property, or state or federal park lands.

WDFW will process all RSMP and Pierce County mussels for biological and chemical analysis, compile the results, conduct a quality assurance (QA) and quality control (QC) review of the data, and submit the data to EIM. Ecology staff will review the biological and chemistry data, notify WDFW of any problems regarding data quality, and will coordinate the final upload to EIM. The RSMP Coordinator will review all monitoring reports and Ecology permit managers will review Pierce County annual reports for compliance purposes.

### **Coordination and Training**

Pierce County will contribute data collection information and results to their permit manager and the RSMP Coordinator. During the summer of 2015 WDFW will provide training for WDFW staff and volunteers and Pierce County staff regarding mussel cage deployment and retrieval. This training will take the form of a webinar or document (i.e. self-train) to ensure comparability of results for both programs. Pierce County is required to use mussels prepared by WDFW on the day(s) of mussel cage deployment. On the morning(s) following mussel cage retrieval WDFW staff and/or volunteers and Pierce County are required to transport their mussels to the WDFW Marine Resources Laboratory in Olympia for processing. These requirements ensure comparability of results for the RSMP nearshore mussel study.

### **Timeline for Mussel Monitoring Field Work:**

- 1) Determine candidate monitoring sites
- 2) Reconnaissance and verification of suitability for monitoring at candidate sites:
  - a) Obtain permission to access site and place monitoring cage there
    - b) Visit site during daylight low tide:
      - i) Assess accessibility and safety
      - ii) Determine type(s) of anchor(s) needed to secure mussel cage to substrate
- 3) Determine which permits/permissions (in addition to those mentioned below) are necessary for monitoring and obtain them prior to monitoring:
  - a) The following blanket permits and permissions will be provided by WDFW 1. HPA, 2. Shellfish Transfer Permit, and 3. MOU with DNR to access SOAL.
- 4) Purchase and assemble equipment and supplies (i.e. cages/anchors, GPS devices, etc.)
- 5) Obtain bagged mussels from WDFW at aquaculture facility:
  - a) Date/time set by WDFW
- 6) Deploy cages with mussels to designated monitoring sites during evening October low tides:
  - a) Record field measurements and site data
- 7) Retrieve cages during evening February low tides:
  - a) Record field measurements and site data
  - b) Place mussels on bags on ice in cooler, hold overnight
- 8) Mussels delivered in coolers to WDFW Marine Resources Laboratory in Olympia, WA the morning after collection.

#### ----- End of Pierce County responsibility ------

- 9) WDFW post-sample processing:
  - a) Determine percent mortality of mussels in cages
  - b) Measure, shuck and dry subset of mussels for determination of condition index
  - c) Measure, shuck and homogenize subset of mussels into wet tissue composites

- d) Freeze wet tissue composites and remaining mussels
- 10) WDFW will transport wet tissue composites to contract analytical laboratories for chemical analysis

### Laboratory Selection

Mussel tissue composites will be analyzed for contaminants at two local laboratories recommended and contracted by WDFW (Table 3). Pierce County mussel tissue composites will be transported to these same laboratories by WDFW, along with all the other RSMP samples. To maintain quality assurance of the analytical data, analysis of mussel samples from Pierce County will occur at these same laboratories over both sampling seasons. WDFW will contract with these laboratories for the RSMP, and Pierce County can enter into this contract on a per site cost sharing basis.

Laboratory Name	Analytical Purpose	Address	Phone
Northwest Fisheries Science Center Laboratory (NWFSC)	Mussel tissue conventional and organic contaminants (persistent organic pollutants – POPS), and replicate samples	2725 Montlake Blvd East Seattle, WA 98112-2097	(206) 860-3325
King County Environmental Lab (KCEL)	Mussel tissue metals and replicate samples.	322 West Ewing Street Seattle WA 98119-1507	(206) 477-7200

Table 3. Laboratories selected for sample processing and analysis.

# Site Selection and Evaluation

The sampling site selection and evaluation process is described here for suitable mussel monitoring sites for the RSMP and Pierce County. Suitability is based largely on a field visit to candidate sites in the spring months of the sampling year. WDFW and Pierce County will provide a table listing the decisions and reasons for site selection or disqualification resulting from site evaluations to the RSMP Coordinator by August 31, 2015. Additional site suitability details to be considered on the day of sampling are described in the sections of this QAPP detailing the sampling methods.

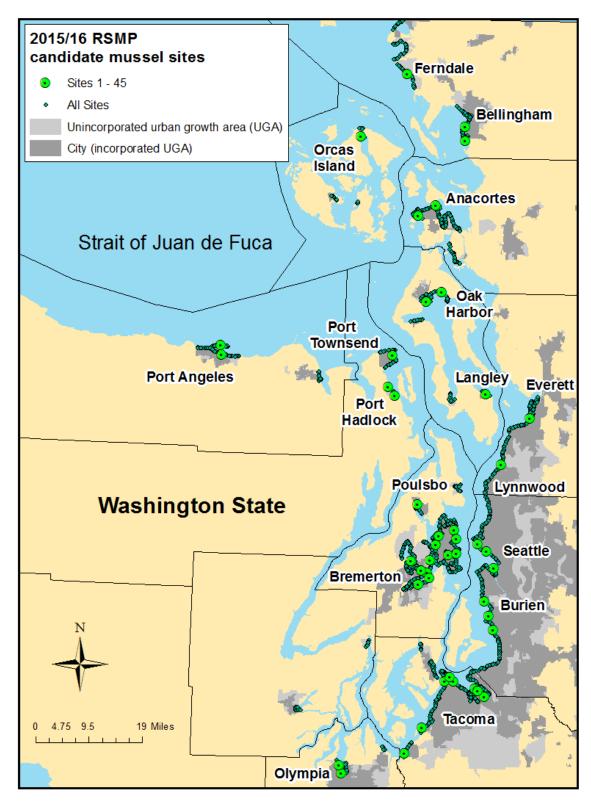
## Site Lists

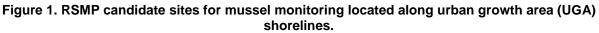
The 2015/16 RSMP and Pierce County mussel sampling site locations come from the RSMP's <u>Puget Sound Mussel Monitoring</u> sample design. The intent of the study design was to create a random list of sites, using a Generalized Random Tesselation Stratified (GRTS) model for drawing spatial samples, from a population of sites along urban growth areas (UGAs) of the Puget Sound. Each site represents an average shoreline length of 800 meters (m); a GRTS-computed weight for each site of 799.8942 m. WDFW advised the RSMP to use an 800 m length of shoreline to represent a mussel site based on criteria used by the National Centers for Coastal Ocean Science's COAST National Status & Trends <u>Mussel Watch Contaminant</u> <u>Monitoring</u> program. This shoreline length was also supported by results from a mussel contaminant study conducted in 2012/13 by the Tacoma Pierce County Health Department in collaboration with WDFW. Results of that study are available in the document titled "Mussel Watch Gradient Report - Hylebos Waterway and Ruston Way" (Callahan, Hanowell, Jensen, 2014).

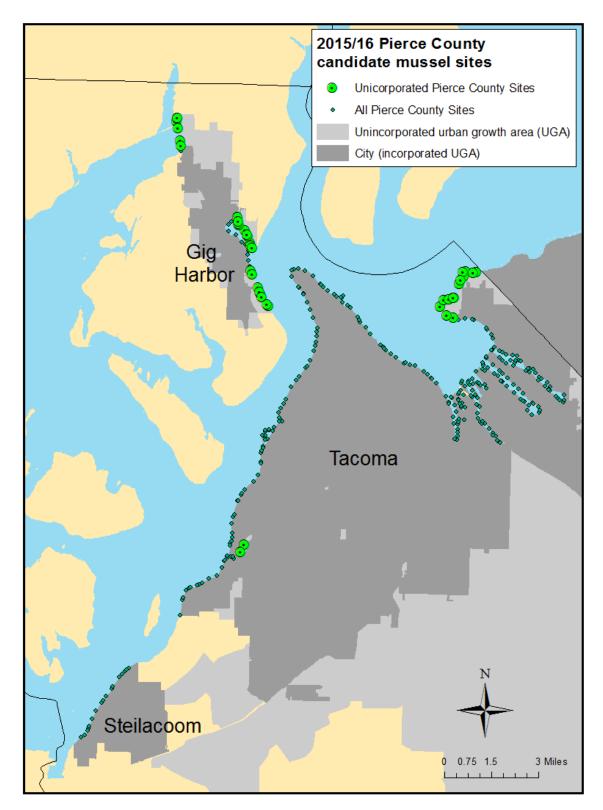
The GRTS algorithm that created the Puget Sound Mussel Monitoring sample draw resulted in a total of 2,048 sites in Puget Sound's UGAs, of which 40 locations are required for RSMP monitoring in 2015/16. WDFW staff and volunteers will evaluate candidate sites from this list (with the exception of sites within Pierce County) in numerical order from lowest to highest until 45 sites have been confirmed. The five extra confirmed sites will provide a number of reserve (i.e. contingency) sites, in case one of the original 40 sites is rejected on the date of deployment.

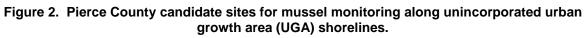
The number of sites required for monitoring by Pierce County is stated in permit condition(s) S8.B.1.b.ii for that county. Pierce County must sample the first eight (8) qualifying shoreline sites in their unincorporated UGAs from the Puget Sound Mussel Monitoring sample draw. It is recommended that Pierce County also have two extra sites in reserve on the date of deployment.

Figure 1 shows the initial 45 RSMP candidate sites (large circles) and the remaining sites (smaller circles); the first 100 RSMP sites are also listed in Appendix B. Figure 2 shows the 41 candidate Pierce County sites in unincorporated UGAs (large circles), as well as and the remaining sites in incorporated areas of Pierce County (smaller circles); the 41 unincorporated Pierce County sites are listed in Appendix C and are also available on Ecology's RSMP website.









### **Site Evaluation**

The initial list of required candidate sites for the RSMP and Pierce County must be verified by a field crew to determine suitability for sampling. Visiting a candidate site in the daylight during low tide, well in advance of monitoring, is important for evaluating accessibility, safety, and suitability of the site, which will include an evaluation of the intertidal substrate at 0 to -1.5 feet mean lower low water (MLLW).

### **Overview of Site Layout**

Each candidate site's coordinates mark a location in the center of an 800 meter (m) long shoreline segment within the Puget Sound (hereafter called the candidate "site center"). The site center is located in the high intertidal zone. Figure 3 illustrates the layout of the sampling locations at each candidate marine site. Extending from the candidate site center (shown with a star in Figure 3) in a straight line perpendicular to the shoreline and into the subtidal zone are three distinct marine sampling locations.

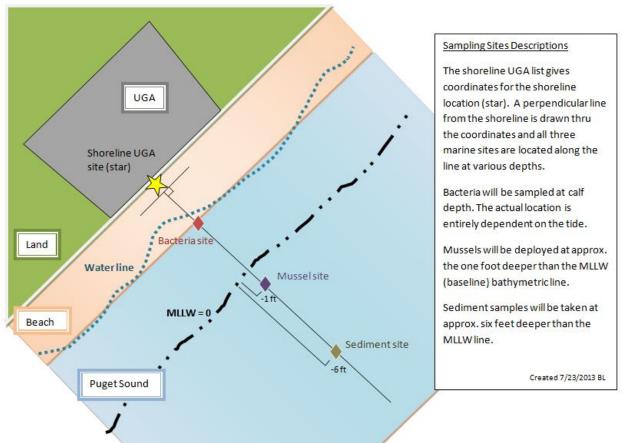


Figure 3. RSMP marine nearshore site layout along shoreline. Each nearshore site is located in the center of an 800 meter shoreline segment.

The first of the three locations (at the waterline), is intended for sampling of bacteria, the second location (in the intertidal zone) is designated for mussel cage deployment, and the third location (in the subtidal zone) is intended for sediment sampling. Bacteria and sediment sampling for the

RSMP are described in separate QAPPs. The intertidal mussel monitoring site will be placed perpendicular to the target coordinates of the candidate site, at a depth of between 0 to -1.5 feet MLLW.

## Criteria for Selecting a Suitable Sampling Site

The suitability of a mussel sample site will be determined using the criteria outlined below. Field crews must evaluate the suitability criteria outlined below at the site center. If the site center is not suitable, then the field crew will evaluate conditions up to 400 meters (1312 feet or 0.25 mile) in either direction along the shoreline until the *closest suitable location* relative to the site center is found.

Suitability of a candidate site is determined by the following criteria:

- Condition 1 the site is NOT within a marina or port (i.e. where multiple motorized vessels are kept in the water), and
- Condition 2 the site can be safely accessed and worked on in the winter, during nighttime low tides, and
- Condition 3 permission of property owners and/or tenants is granted prior to sampling, and
- Condition 4 there is suitable substrate or a location for anchoring/securing a mussel cage at the site.

If a location other than the site center is chosen, then the reason for disqualification of the site center must be documented and the alternate site coordinates must be recorded. If all 800 m of a candidate site are not suitable, then the reason for disqualification must be documented, including photos, and alternate candidate sites must then be visited, in numerical order from the site list, and verified for replacement.

### Accessibility Criteria

These criteria concern whether access to a candidate site is permitted by the land owners, and if the site can be safely accessed and sampled throughout the year. A site may also be deemed unsuitable or impracticable for sampling if more than one hour is required to access the site from the nearest parking location.

### Permission

If the mussel cage is to be placed on private or commercially-owned tidelands, or private property must be traversed to gain access to public tidelands, permission must be granted from the land owner(s) prior to monitoring. Useful shoreline information can be gained from a desktop evaluation of candidate sites (i.e. search Google maps, public records, etc.) and a good faith effort to contact owners or tenants. In some cases it might be necessary to obtain a special license, easement, or other legal document from a commercial or government property (i.e. Port Authority, City/County park, Tribe, etc.) to access and place a mussel cage on their property.

Property owners will be contacted well in advance of (i.e. several months before) cage deployment. This will ensure adequate time to explain the needs and timing of the study and to obtain permission to access the property during night-time low tides. In some cases keys or gate codes may be necessary to allow field crew access after business hours. <u>Property owners should</u>

be reminded the day before mussel cage deployment and removal that workers will be on their property soon.

### Permits

WDFW will obtain a blanket <u>HPA</u>, <u>Shellfish Transfer Permit</u>, and MOU with the DNR to access <u>SOAL</u> for all RSMP mussel monitoring activities. These permits and permissions will also cover sites monitored by Pierce County, as long as guidelines for mussel monitoring laid out in this QAPP are followed.

WDFW is responsible for obtaining any *other* permits or permissions (outside those listed above) necessary to conduct mussel monitoring work at the RSMP approved sites, including but not limited to site access permits for privately-owned, city, county, port authority, or tribal properties, or state or federal lands. Similarly, Pierce County is responsible for any other permits and permission at their sites. For instance, A <u>Scientific Research Permit</u> is required when conducting research (including mussel monitoring) within the boundaries of a Washington State Park. Application for this permit must be sent to Washington State Parks (<u>http://www.parks.wa.gov/stewardship/</u>) at least two weeks prior to mussel monitoring.

### Safety

Field work, particularly in coastal environments, has an inherent risk of danger and environmental conditions can often be unpredictable. Mussel site reconnaissance, deployment, and retrieval pose a number of potential safety hazards including: unstable terrain (i.e. deep mud or cobbles/boulders), incoming tides, breaking waves, exposure to extreme temperatures, and sudden changes in weather. Field crews will evaluate each candidate site for safety. Appropriate reasons for disqualifying a candidate site for monitoring may include:

- route of entry or intertidal area is unstable or unsafe (e.g. sucking mud, quicksand),
- hostile people or animals are present.

### **Intertidal Physical Criteria**

These criteria concern the conditions of the intertidal substrate at a candidate site for mussel monitoring. To be considered suitable for mussel cage placement, the intertidal area at the candidate site's center (or within 400 meters of the site center) must:

- have a substrate (i.e. mud, sand, cobble) into which a helical/screw anchor or rebar stakes can be driven, to secure the mussel cage, OR
- have some kind of structure to which the mussel cage can be tied or secured (e.g. steel or concrete pilings or other fixed points on-site) this is especially important in high energy environments. *However, no cages will be affixed to or placed next to creosote-treated material.*

## **Documentation of Site Evaluations**

Site evaluators must verify all sites given the suitability criteria above. Documentation of observations from both the desktop and field visits will be recorded in a Field Log. Site evaluators will provide a table listing the decisions and reasons for site selection or disqualification resulting from the site evaluations to the RSMP Coordinator by August 31, 2015.

### Site ID and Site Name

### Site ID

Once appropriate sample sites are identified, site evaluators will use the unique, pre-assigned "Site ID", which can be found in Appendices B and C under the "SITE\_ID" column, to identify each individual site. Each "Site ID" will be entered into the 2015/16 RSMP Mussel Monitoring Datasheet (Appendix D) during sampling. The Site ID will eventually become the "Location ID" in Ecology's EIM database and serves as the unique site identifier that relates the sampled sites to the GRTS study design, and is denoted as PSS13175-XXXXX where the "X" number changes for each site.

### Location Name

Site evaluators will assign a unique and appropriate "Location Name" to each of their sampling sites. The Location Name should be succinct, and is limited to 40 characters by the EIM database. The name may be general or describe the location (e.g. Tacoma, or Commencement Bay, or Ruston Waterfront, or Steilacoom) or be more specific descriptor like a nearby stream/river, neighborhood/street, marine location, or other identifying landmark (e.g. Thea Foss, or Hylebos Waterway, or Point Defiance, or Days Island, or Ferry Terminal).

Some examples of appropriate Site Names:

- Tacoma Titlow Park
- Commencement Bay Blair Waterway
- Point Defiance Ferry Terminal
- Ruston Way Dickman Mill Park
- Thea Foss Waterway 11<sup>th</sup> St Bridge

### Order #

There is another field in EIM called the "Study\_Specific\_Location\_ID" that is unique to the study. This field will be populated by a concatenation of the ORDER # (Column A) and the acronym "SUGA" which stands for Shoreline along Urban Growth Area; for example "044-SUGA".

# **Quality Objectives**

The quality objectives for nearshore mussel monitoring described here are to obtain and analyze sufficient numbers of high quality mussel tissue samples to meet the goals and objectives of the RSMP program (Table 4).

Table 4. Summary of mussel tissue composites to be collected and analyzed for chemical
contaminants during this study.

Purpose	Location	Timing	Composites	Replicates
Baseline samples	Aquaculture source	October	6	6
RSMP mussel sites	Various	January/February	40	1 per site
Pierce County sites	Various	January/February	8	1 per site
Lab QA samples	Various	Aliquots taken during chemical analysis	5	5 <sup>a</sup>
Total			59	

<sup>a</sup> two QA samples per batch of 12

# **Field Measurements**

WDFW staff and volunteers and Pierce County will record the GPS coordinates of the mussel cage at each deployment site with individual GPS units. Each field team will record the make and model of their GPS unit and the accuracy of the GPS reading when taken. In addition, all GPS devices used in this study will be set to North American Datum 83 (NAD83) for comparability and coordinates will be recorded in decimal degree format. The specifications for many GPS receivers indicate accuracy within 3 to 15 meters (10 to 50 feet) 95% of the time (http://www.gps-basics.com). Since mussel sites will be placed at least a half mile apart, this level of accuracy is acceptable for the RSMP's study purposes.

Measurements of tidal stage, substrate type, and upland and shoreline characteristics are taken by field staff during a sample collection event. WDFW staff and volunteers and Pierce County must meet measurement quality objectives (MQOs) listed in Table 5. Collection methods, reporting requirements, and quality control (QC) procedures summarized in the *Measurement Procedures* and *Quality Control Procedures* sections of this QAPP are intended to provide field measurement data that meet MQOs and RSMP objectives.

procedures. See 2015/16 RSMP Mussel Monitoring Datasheet (Appendix D).					
Parameter	Expected Range Of Results	Technique/ Instrument	Measurement Method	QA/QC	
Time of cage deployment and retrieval	12:00 - 24:00	Clock	Read from clock and reported in military time	Careful observation	
GPS coordinates	N/A	GPS device or mobile device with GPS application	Set GPS device to NAD83, record in decimal degrees (e.g. 47.5893, - 122.3953)	Record accuracy of coordinates at reading (e.g. ±15ft)	
Wave energy	Flat, calm, wind chop, swells, breaking waves	Visual examination	Visual examination of sea near cage	Careful observation	
Beach exposure level	Exposed, moderately exposed, sheltered	Visual examination	Visual examination of beach within <sup>1</sup> / <sub>2</sub> mile in either direction of cage	Careful observation	
Time of most recent low tide (MLLW)	12:00 - 24:00	NOAA tides and currents website <u>http://www.protid</u> <u>es.com/washingto</u> <u>n/</u>	Read from harmonic or subordinate tidal gauge station nearest to monitoring site	Accurate reading of information from website	
Height of most recent low tide (MLLW)	0 to -4 ft.	NOAA tides and currents website	Read from harmonic or subordinate tidal gauge station nearest to monitoring site	Accurate reading of information from website	
Precipitation	None, steady rain, showers, snow, hail	Visual examination	Visual examination of atmosphere	Careful observation	
Majority (>50%) Substrate Type	Bedrock- hardpan, cobble-gravel mix, sand- gravel mix, sand, sand-mud mix, mud-silt	Visual examination	Visual examination within 200 foot radius of cage	Careful observation	
Aquatic vegetation coverage	None (<1%), 1- 20%, 20-40%, 40-60%, 60- 80%, 80-100%	Visual examination	Visual examination within 200 foot radius of cage	Careful observation	
Aquatic vegetation type	None, eelgrass, kelps, fucus, ulva, other	Visual examination	Visual examination within 200 foot radius of cage	Careful observation, may include mix of types	
Freshwater inputs	Natural streams, rivers, outfalls	Visual examination	Visual examination within 200 foot radius of cage	Careful observation, may include mix of types	
Adjacent upland land-	Wide range of choices (see	Visual examination	Visual examination of beach within <sup>1</sup> / <sub>2</sub> mile in	Careful observation, may include mix of	

# Table 5. Mussel monitoring field parameters: field methods, reporting limits, and QA/QC procedures. See 2015/16 RSMP Mussel Monitoring Datasheet (Appendix D).

Parameter	Expected Range Of Results	Technique/ Instrument	Measurement Method	QA/QC
use type	Appendix C)		either direction of cage	types
Erosion control structures	None, hard, soft. Includes materials used	Visual examination	Visual examination of beach within ½ mile in either direction of cage	Careful observation and documentation
Abandoned or derelict structures	No/Yes, type	Visual examination	Visual examination of beach within ½ mile in either direction of cage	Careful observation and documentation
Man-made structures on beach	N/A	Visual examination	Visual examination of beach within ½ mile in either direction of cage.	Careful observation, may include mix of types
Current shoreline use	Wide range of choices (see Appendix C)	Visual examination	Visual examination of beach within ½ mile in either direction of cage.	Careful observation, may include mix of types
Construction of structures on beach touching water	Treated wood, concrete, steel, other	Visual examination	Visual examination of beach within <sup>1</sup> / <sub>2</sub> mile in either direction of cage.	Careful observation, may include mix of types
Outfalls	N/A	Visual examination	Visual examination of beach within <sup>1</sup> / <sub>2</sub> mile in either direction of cage.	Careful observation, may include mix of types
Potential sources of pollutants	N/A	Visual examination	Visual examination of beach within <sup>1</sup> / <sub>2</sub> mile in either direction of cage.	Careful observation, may include mix of types

\*Field-measured parameters follow manufacturer's website guidelines for calibrations.

# Laboratory Measurements

The objective for laboratory processing is to evaluate the biological metrics (mortality and condition index) of the transplanted mussels, while the objective for analytical chemistry is to evaluate the target analytes, with limits of detection sufficient to identify and measure the analytes. The RSMP will use mussels from a single source for the cages to minimize variability. Baseline samples of the mussel stock sufficient for both RSMP and Pierce County uses will be conducted by WDFW.

Mussel tissue chemical analyses will be conducted at the Northwest Fisheries Science Center Laboratory to ensure comparability of results. All work is expected to follow the laboratory methods and meet laboratory QC requirements of the analytical methods outlined in this QAPP. These requirements can be found in detail in the Puget Sound Estuary Program protocols (PSEP, 1986, 1997a, b, c) and in the peer-reviewed standard operating procedures (SOPs) for each test. Following are three tables listing the minimum QA criteria for organic chemicals and metals analyzed in mussels for this study (Tables 6, 7 and 8).

## Precision

Precision is monitored and controlled within batches using laboratory replicates of field samples and across batches by analyzing Standard Reference Materials (SRM) of applicable matrix i.e., tissue. Cross-batch precision is expressed as the relative standard deviation (RSD) for repeated measurements. The RSD of analyte responses relative to the internal standard must be  $\leq 15\%$  for the repetitions.

## Bias

Bias or accuracy of samples is evaluated by comparing measured SRM values with National Institute of Standards and Technology (NIST) certified values. In addition for persistent organic pollutants (POPs), concentrations of  $\geq$ 70% of individual analytes are to be within 30% of either end of the 95% confidence interval of the reference values.

## Comparability

The SOPs described in this document (Sloan et al. 2014; Sloan, Brown et al. 2004; Sloan, Brown et al. 2006) are consistent with other concurrent and future sampling efforts that could be used as comparison for mussels. In addition, methods detailed here are consistent with ongoing WDFW monitoring of contaminants in other Puget Sound species.

Although not necessary for the current project, comparability with historical NOAA Mussel Watch or other data will require some targeted evaluation. The performance-based nature of current analytical procedures is designed to allow the broadest comparability with other similar programs, however some discrepancies will exist with new vs. older mussel monitoring programs. For example, PCB Aroclors vs. PCB congeners that will be used in this study. This issue will be addressed in future efforts to fully expand and establish a mussel-monitoring program in Puget Sound.

# Table 6. Quality assurance criteria for PCBs, PBDEs, PAHs, and OCPs. Reproduced from Table 8in Sloan et al. (2006).

Quality assurance element	Minimum frequency	Acceptance criteria
Instrument calibration	Once every batch of samples	Analyte concentrations are to
	or once every two batches in	be calculated using point-to-
	one continuous analytical	point calibration with at least
	sequence	four concentration levels of
		calibration standards.
Continuing calibration	At start and end of every	The RSD of the analyte
	analytical sequence and every	responses relative to the
	10 or fewer field samples	internal standard is to be
		≤15% for the repetitions.
Reference materials:	One with every batch of 20 or	Concentrations of ≥70% of
Sediment: NIST SRM 1944,	fewer field samples	individual analytes are to be
NIST SRM 1941b		within 30% of either end of
Mussel tissue NIST SRM		the 95% confidence interval of
1974b		the reference values. These
Blubber: NIST SRM 1945		criteria do not apply to
Fish tissue: NIST SRM 1946,		analytes with concentrations
NIST SRM 1947		below their LOQ with the
		lower LOQ is within or greater
		than the 95% confidence
		interval, nor to those analytes
		known to have coeluting
		compounds.
Method blank	One with every batch of 20 or	No more than 5 analytes in a
	fewer field samples	method blank are to exceed
		2x lower LOQ. Samples are
		not corrected for analytes
		found in the blank.
Sample replicates (i.e.	One with every 20 or fewer	RSDs are to be ≤15%
duplicates or triplicates)	field samples.	(equivalent to relative percent
		difference ≤30% for
		duplicates) for ≥90% of the
		analytes that have
		concentrations ≥1 ng/g.
Internal standards/surrogates	At least one internal	The recoveries are to be 60-
	standard/surrogate is added	130%.
	to every sample	
Interlaboratory comparisons	At least one per year	In conjunction with the NIST
		or the IAEA.

Quality Control Element	The Description of Element		Control Limit
Element		Implementation	Tissue
Method Blank (MB)	Interference-free matrix to assess overall method contamination	1 per sample batch	± MDL
Spike Blank (SB)	Interference-free matrix containing all target analytes	1 per sample batch	85 - 115%
Standard Reference Material (SRM)	Certified reference material from NIST or NRCC that is digested with samples.	1 per solid or tissue sample batch, if applicable	80-120% <sup>c</sup>
Laboratory Control Sample (LCS)	Certified reference material from a source other than NIST or NRCC	1 per solid or tissue sample batch, if applicable	80-120% <sup>c</sup>
Matrix Spike (MS)	Sample matrix spiked with all/subset of target analytes prior to digestion	1 per sample batch	75-125%
Lab Duplicate (LD) <sup>a</sup> ,	Self-explanatory	1 per sample batch	RPD ≤ 20%

Table 7. Required batch quality control measures and quality assurance criteria for mercury viaCVAA. Reproduced from KCEL SOP 604v6.

<sup>a</sup> No calculation performed when both sample and duplicate values < RDL

<sup>c</sup> Or varies due to control charting

Table 8. Required batch quality control measures and quality assurance criteria for the ICP-MS
metals As, Cd, Cu, Pb and Zn. Reproduced from KCEL SOP 624v2.

Quality		- Frequency of	Frequency of	Control Limit
Control Element	Description of Element	Implementation	Tissue	
Method Blank (MB)	Interference-free matrix to assess overall method contamination	1 per QC batch	± MDL	
Spike Blank (SB)	Interference-free matrix containing all target analytes	1 per QC batch	85% - 115%	
Standard Reference Material (SRM)	Certified reference material from NIST or NRCC that is digested with samples.	1 per solid or tissue sample batch, if applicable	80-120% <sup>b</sup>	
Laboratory Control Sample (LCS)	Certified reference material from a source other than NIST or NRCC	1 per solid or tissue sample batch, if applicable	80-120% <sup>b</sup>	
Matrix Spike (MS)	Sample matrix spiked with all/subset of target analytes prior to digestion	1 per QC batch	75% -125%	
Lab Duplicate (LD) <sup>a</sup>	Self-explanatory	1 per QC batch or MSD.	$\leq 20\%$ RPD, when at least one value is > RDL	

<sup>a</sup> No calculation performed when both sample and duplicate values < RDL

<sup>b</sup>Or varies due to control charting

### Representativeness

Mussels used for this study will be of the species *Mytilus trossulus* (bay or foolish mussel), which is indigenous to intertidal habitats in the Puget Sound. As recommended in the *Standard Guide for Conducting In-situ Field Bioassays with Caged Bivalves* (ASTM E2122-02, 2007), mussels for this study will come from an aquaculture facility. The source will be Penn Cove Shellfish, Inc. in Penn Cove, Whidbey Island, Washington. The advantage of using mussels from this facility is that all individuals will be of similar ages from the same population, will have a similar genetic and environmental history and are expected to be relatively uncontaminated. In addition, Penn Cove Shellfish, Inc. is the only local aquaculture farm that raises *M. trossulus*.

The target size of mussels selected for transplantation will be based on the median size ( $\pm$  5 mm) of 100 randomly selected adult (approximately 11 months old and larger than 45 mm) mussels available when bagging begins. Based on previous measurements taken at Penn Cove Shellfish on August, 2012, mussels selected for transplantation will likely measure between 50 – 60 mm in shell length.

Since the Puget Sound on average receives its highest amount of rainfall in the winter months, the sampling period chosen for this study (October – January/February) represents a period when input of contaminants from stormwater runoff is at its potential highest. Mussel cages will be placed on the intertidal substrate between 0 to -1.5 feet mean lower low water (MLLW), with mussels suspended approximately 40 cm above the substrate. The placement of cages is meant to simulate contaminant conditions experienced by most nearshore biota in the intertidal zone during the winter in Puget Sound.

## Completeness

The goal of this study is to collect and analyze mussel tissue from 40 randomly selected sites from the Puget Sound shoreline UGAs, however, some cages may be lost due winter storms, vandalism or theft.

Based on the number of individuals used to determine the condition of mussels from National Mussel Watch Program sites (Kim et al. 2006), a sample size of 12 mussels from each site will be selected for determination of condition index (CI). For tissue chemistry analysis a composite size of about 32 individuals (200g of soft tissue) per site (cage) was selected to optimize the amount of tissue available for analysis at the two chemistry laboratories. This mass is based on previous experience with the same laboratories, and allows enough tissue for reanalysis (if needed) and archiving small (20 g) subsamples. The number of mussels per composite was selected to balance representativeness of the population with the labor and time constraints related to processing samples. Our goal will be achieved if we are able to create a tissue composite from every site.

# **Sampling Procedures**

This section describes field and lab sampling procedures. The following sampling procedures are outlined in time-sensitive order. Field activities should be conducted by at least two people. Activities can be parsed into tasks to be accomplished by one or more persons at a given time. Mussel monitoring methods will, in general, follow those described below. A complete list of field materials required for mussel cage deployment and retrieval can be found in Appendix H.

# **Preparation for Field Work**

## Safety

Mussel site reconnaissance, deployment, and retrieval pose a number of potential safety hazards to field crew, including unstable terrain (i.e. deep mud or cobbles/boulders), incoming tides, breaking waves, exposure to extreme temperatures, and sudden changes in weather. A contact person will be designated at the office to which field personnel report at pre-designated times.

WDFW staff/volunteers and Pierce County staff will develop a site-specific safety plan including at a minimum the following elements. To ensure their safety, all field crew members are required to follow these safety guidelines:

- Do not go to the monitoring site alone; use a minimum of two people.
- Wear appropriate clothing for thermal and water protection.
- Be alert to breaking waves wear a life jacket if appropriate.
- Avoid falls wet rocks and logs are slippery.
- Avoid getting stuck in deep (i.e. sucking) mud.
- Wear gloves: protect hands from cuts and samples from contamination.
- Bring a cell phone or other means of two-way communication to call for emergency response in the field if needed.

It is possible that during deployment or retrieval, invasive species (e.g. benthic invertebrates or marine plants) could collected on equipment or clothing (e.g. boot treads). All material not retained for analyses or archiving will be rinsed near the sampling location with water.

## Field Log

The lead scientist at WDFW and Pierce County will maintain a water-resistant field logs with detailed notes for each major monitoring-related activity detailed below. Information recorded will include:

- Name and location of project
- Field personnel
- Sequence of events and/or changes in plans or procedures
- Unusual circumstances that may affect interpretation of results

If a candidate mussel monitoring site is found to be unsuitable, the reasons for rejecting the site must be recorded in the Field Log. Alternate candidate sites must be visited and verified.

### Field Datasheets

WDFW and Pierce County will print a 2015/16 RSMP Mussel Monitoring Datasheet (Appendix D) on water-resistant paper for each verified and usable site. These datasheets will be filled out with data from each mussel monitoring site at the time of deployment and saved to complete at the time of cage retrieval.

## Chain-of-Custody

A *Mussel Chain-of-Custody form* (Appendix E) will be used to track mussel possession during the field and laboratory portion of the study. The chain-of-custody (COC) will be initiated by WDFW for each monitoring site to track possession of mussel bags (i.e. start of monitoring) and will be maintained by each party responsible for the mussels until all samples are relinquished to the WDFW Marine Resources Laboratory in Olympia.

## **Equipment Preparation**

### Decontamination, Prevention of Spread of Invasive Species

RSMP and Pierce County will conduct field work and clean equipment to prevent the spread of invasive species. Staff and equipment that contact multiple surface waters will, at a minimum, be cleaned according to Ecology's SOP EAP070, *Minimizing the Spread of Aquatic Invasive Species* (Ecology, 2012). These procedures will be followed at the end of each work day or upon leaving a water body before entering another. Some areas are designated to be of "Extreme Concern"; these areas are shown in several maps at the following link: www.ecy.wa.gov/programs/eap/InvasiveSpecies/AIS-PublicVersion.html

### Cages

WDFW and Pierce County will obtain plastic-coated, wire mesh cages (anti-predator cages, Figure 4) with the following attributes:

- Size =  $16 \times 16 \times 16$  inches (length x width x height)
- Mesh opening =  $1.25 \times 2.5$ cm
- Removable lids.

Acceptable cages are sold at McKay Crab and Shrimp Gear, in Brinnon, Washington.



Figure 4. Anti-predator mussel monitoring cage (lid shown inside cage) with 30-inch screw anchor and bent-tip rebar stake.

To dissipate any potential surface contaminants, cage owners will either 1) soak cages and anchoring materials to be used for monitoring in water for 24 hours prior to use, or 2) wash the cages and anchoring materials with a high pressure hose using fresh water.

#### Anchors

WDFW and Pierce County will obtain anchoring devises suitable for anchoring their cages into the substrate at their individual monitoring sites. WDFW recommends using a screw anchor (30-inch shaft recommended) and four bent-tip rebar stakes to anchor cages in mud, sand or sand/cobble beaches. Large cable ties (3 to 5 foot long) may be used as alternate anchoring devices to secure cages to fixed objects like non-creosote pilings or boulders. In addition, cinder blocks may be purchased and used in combination with cable ties and/or rebar stakes as anchoring devices.

### **Mussel Preparation**

WDFW will coordinate with Pierce County to arrange mussel pick-up from the aquaculture facility and delivery of mussels post-deployment to the WDFW Marine Resources Lab in Olympia. The exact location and day of pick up will be announced with at least a month notice, but is currently planned for October 2015 and January 2016.

### **Preparation of Study Population**

The following sections describe the procedure WDFW will follow for harvesting, measuring, and bagging mussels at Penn Cove Shellfish, Inc. a commercial aquaculture facility, in preparation for subsequent deployment in anti-predator mesh cages at sites around the greater Puget Sound.

The protocols described below are based on procedures outline in the *Standard Guide for Conducting In-situ Field Bioassays with Caged Bivalves* (ASTM E2122-02, 2007). Although the

*Standard Guide* initially mentions several possible cage types for in-situ field tests with caged bivalves, the majority of their subsequent field measurement and sampling methods are based on the assumption that the researcher is using individually compartmentalized mussels in cages suspended in the water column. In this study mussels will not be individually compartmentalized; they will be grouped together within their cages. In addition, cages will be deployed in the intertidal zone on the substrate, not suspended in the water column. Thus although the methods outline here are based on guidance from the *Standard Guide for Conducting In-situ Field Bioassays with Caged Bivalves* modifications have been made where necessary to accommodate the specifics needs of the RSMP.

### **Determination of Mussel Size Range**

The target size of mussels selected for bagging and subsequent transplantation will be based on the median size ( $\pm$  5 mm) of 100 randomly selected adult (approximately 11 months old and larger than 45 mm) mussels available the day before bagging begins. Based on previous measurements taken at Penn Cove Shellfish, Whidbey Island on August, 2012, mussels selected for transplantation will likely measure between 50 - 60 mm in shell length.

### **Mussel Presort**

The presorting, measuring, and bagging described below will take place during the September prior to deployment, allowing time for inclement weather.

WDFW staff and volunteers will obtain live mussels for cage deployment during normal, periodic harvest operations conducted by Penn Cove Shellfish, Inc. aquaculture staff. Penn Cove Shellfish, Inc. grows mussels attached to 20 foot sections of rope hanging under floating docks. Penn Cove staff harvest mussels by removing them from the ropes and cleaning them with specially designed brushes aboard a harvesting vessel. WDFW staff and volunteers will divert live, cleaned mussels from this operation to a nearby beach, where sorting, measuring and bagging will occur.

During the beach sorting, measuring and bagging mussels will be kept in the shade, so as not exposed them to direct sunlight for long periods of time. Mussels will be held in ambient seawater in coolers while they wait processing. Using a knife or scissors we will select mussels that fall within the desired size range and, if necessary, separate them from one another by cutting their byssal threads. Care will be taken not to pull or tear the byssal threads, so as not to damage the byssal glands. The cleaned and separated mussels will then be replaced into a cooler filled with ambient Penn Cove seawater.

WDFW will monitor the water temperature inside this seawater holding cooler with a thermometer, to ensure it stays within  $\pm 5^{\circ}$  C of current Penn Cove surface temperature, and change water as needed to maintain suitable water quality.

### Measuring and Bagging

WDFW staff and volunteers will take presorted mussels from the holding cooler and measure their shell length. Only intact mussels with no cracks in their shells and that respond to physical stimulation by tightly closing their shells will be selected for measuring and bagging. Mussels that do not meet these requirements will be discarded.

### Measuring

Mussels will be randomly selected from the holding cooler. WDFW staff and volunteers will measure shell length (umbo to farthest posterior margin) using a digital caliper with measurement accuracy of 0.1 mm. Length measurements will be manually recorded onto a waterproof paper data sheet.

### Bagging

Sixteen (16) measured mussels will be placed into a heavy duty mesh bag measuring 20 inches in length. WDFW staff and volunteers using a cable tie will divide the bag into two sections with eight mussels in each section. The finished mussel bags will have two separate sections providing ample space for the mussels to feed and grow.

WDFW staff and volunteers will affix a plastic identification tag with a unique number to each finished bag. This number will be noted alongside the measurements of the mussels for that specific bag. Once the identification tag is affixed to the filled mussel bag the bag will be placed into another holding cooler filled with ambient Penn Cove seawater. The seawater in these coolers will be maintained in the same fashion as described above.

### Presoak period

Once a sufficient number of mussel bags have been processed, WDFW staff and volunteers will affix them to a 20-foot weighted line, spaced approximately six inches from each other. Approximately 40 bags will be placed along each line. When a line is filled with bags, Penn Cove Shellfish staff will hang the line under one of their aquaculture platforms. Each line of bagged mussels will be marked with an identification flag indicating the range of bag ID numbers hanging on that line. The location of the line will be noted in the Field Notebook. The finished mussel bags will be left to soak at Penn Cove Shellfish for at least 10 days before they are removed from the water for deployment in mesh cages. The 10+ day period following mussel bagging is intended to allow the mussels a resting period after they are separated, sorted, cleaned and bagged. This allows them time to re-cluster prior to deployment (Andral et al, 2011; Benedicto et al, 2011; Galgani et al, 2011).

### **Mussel Cage Deployment and Retrieval**

WDFW staff and volunteers and Pierce County will place their pre-bagged mussels in wire mesh cages that will be anchored to the substrate with a combination of screw anchors, rebar stakes, and/or concrete blocks as described below. If necessary and possible, some cages may be tied (using large nylon cable ties) to steel or concrete pilings or other fixed points on-site. <u>No cages will be affixed to creosote-treated material</u>.

### **Deployment/Retrieval Dates**

WDFW staff and volunteers and Pierce County will deploy and retrieve their caged mussels during low tide times in the late fall (October 2015) and late winter (January – February 2016), respectively. Deployment and retrieval will occur during one of the preferred dates listed in Table 9 below, with alternate dates to be used only when necessary, such as in the event of a storm or other hazardous condition that precludes field work on the preferred date.

# Table 9. Potential deployment and retrieval dates for RSMP mussel monitoring in 2015/16. Dates are based on predicted low tides at Seattle, Elliott Bay harmonic station (NOAA).

Low Tide Event	Deployment Dates	Retrieval Dates
Preferred	September 30-October 3, 2015	January 20-24, 2016
Alternate	October 27-31, 2015	February 6-10, 2016

#### Baseline Tissue Sampling

At the time of deployment WDFW will sub-sample the bagged mussels from the aquaculture facility to assess the baseline biological and chemical conditions of the starting population. Pierce County has no responsibilities for baseline sampling.

#### Deployment

WDFW staff and volunteers and Pierce County field crews deploying mussel cages (hereafter referred to collectively as "deployers") <u>must be on site</u> to deploy the mussel cage at the time of the zero MLLW on the night of deployment. Proper timing ensures that the field crew can place the mussel cage at 0 to -1.5 feet MLLW (i.e. at the water line at the moment of, or just after, the daily lowest low tide) with plenty of time to work before the incoming tide.

#### Pick Up and Transport RSMP-approved Mussels to the Monitoring Site

Deployers will go to Penn Cove Shellfish, Inc. on Whidbey Island on the afternoon of the low tide on which they will deploy the cage. Deployers will provide a cooler(s) of sufficient size, half filled with ice, to transport the mussels on the date of pick-up. Each deployer will get four bags of mussels (16 mussels per bag) per mussel cage to be deployed. The four mussel bags will be placed into a large plastic Ziploc bag(s) marked with the name of the site(s) where the cage(s) will be deployed. The bagged mussels will be placed in the cooler on bagged ice. <u>Mussels must not come into contact with ice melt water during transportation.</u>

At this time WDFW will initiate a COC form (Appendix E) unique to each monitoring site for which mussels are being transferred. The deployers <u>must keep</u> these forms for later use upon retrieval and delivery of mussels to the WDFW processing laboratory.

Deployers will transport the bagged mussels on ice directly to the deployment site(s) and deployed on the same night they were received from the aquaculture facility, to minimize time out of the water.

#### Secure the Mussels into the Cage

Deployers must wear powder-free nitrile laboratory gloves when handling the mussel bags.

At the mussel site deployers will affix the four mussel bags to the top quarter (¼) of the antipredator cage, so that they span the width of the cage and are spaced evenly apart (Figure 5). Once installed the mussel bags should hang well above the bottom of the cage. Use 8-inch cable ties to secure the end of each bag to the sides of cage, so that the bags are stretched across the middle of the cage and all mussels are an equal height above the bottom (Figure 5). After the mussel bags are fastened inside the cage, record the four mussel bag ID numbers on the 2015/16 *RSMP Mussel Monitoring Data Sheet* (Appendix D), then secure the cage's lid in place with at least eight 8-inch cable ties (two per edge, Figure 6). Sea stars can get through relatively small (0.5 x 1 inch) openings, so it is important not to leave any gaps. If desired, cable ties can be trimmed to about one inch length after they have been fastened.



Figure 5. Mussel bags affixed to the top quarter (1/4) of an anti-predator cage, lid not shown.



Figure 6. Anti-predator cage lid secured in place with at least two 8-inch cable ties per edge (red circles).

### Secure the Cage to the Substrate

Once the mussels are attached inside the cage and the lid is secured, deployers will anchor the cage to the substrate in the intertidal zone between 0 to -1.5 feet MLLW. <u>Timing is critical to</u>

ensure proper placement relative to tidal height; the cage must be installed at or just below the water line when the lowest low tide of the day reaches zero feet.

Whenever possible cages should be anchored to the substrate using a screw anchor (30-inch shaft recommended) and four rebar stakes. The helical anchor must be screwed as deeply into the substrate as possible, leaving only a few inches of the shaft and the top eye hole visible. Screwing in the anchor will require a lever (to turn the anchor) and substantial downward pressure. Figure 7 illustrates use of the lever. Heavy-duty gloves are recommended for installing the screw anchor and the rebar stakes.



Figure 7. Helical, earth or screw anchors and lever used to screw anchor into the substrate. The red arrow indicates the 30-inch long anchor shaft that is recommended.

Once the anchor is installed, the cage will be placed next to the helical anchor and secured to the anchor using two 8-inch cable ties. In addition, rebar stakes should be pounded through the top and/or sides of the cage, taking care to avoid driving the stakes through the mussel bags. Deployers may also cable tie the stakes to the cage (Figure 8).



Figure 8. Mussel monitoring cage driven through with bent-tip rebar stakes (on the far end) and secured to a helical anchor with cable ties. For better cage anchoring, 3-4 rebar stakes are recommended.

If a screw anchor and rebar stakes are not adequate and more or different anchoring is needed, the cage may be secured with large (3 to 5 foot long) cable ties to a <u>non-creosote</u>, fixed object (i.e. piling or pole) or secured to a cement block(s) that will act as a weighted anchor (Figure 9). <u>No cages should be affixed to creosote-treated material</u>.



Figure 9. Examples of additional cage anchoring methods.

#### **Field Measurement Procedures**

This section describes field measurement processes to be conducted by WDFW staff and/or volunteers and Pierce County (hereafter collectively called "field personnel"). Data generated as described in this section will be entered into Excel spreadsheets and verified for accuracy. The original datasheets and the Excel spreadsheets with entered and quality checked data will be delivered to WDFW within one month of creation. Results will be entered into Ecology's EIM database along with the rest of the RSMP data by WDFW staff.

Once the mussel cage has been deployed and anchored to the site, deployers will record field measurements and observations on the 2015/16 RSMP Mussel Monitoring Site Datasheet

(Appendix D) and SAVE the datasheet to be finished during retrieval. Table 10 lists field measurements and observations deployers must make at the time of mussel cage deployment and retrieval. Deployers will also take digital photos confirming proper deployment of the mussel cage.

Field Measurements
Time of cage deployment/retrieval
GPS coordinates and accuracy
Field Observations/Estimates
Wave energy
Precipitation
Beach exposure
Substrate Type
Aquatic vegetation cover and types
Freshwater inputs
Adjacent upland land use
Erosion control structures
Shoreline use
Anthropogenic structures on beach
Outfalls present
Potential sources of pollutants

#### Table 10. Field measurement and observation parameters.

Global Positioning System (GPS) coordinates of the mussel cage will be recorded at each mussel monitoring site. All coordinates will be recorded in decimal degree format (e.g. 47.5893 latitude, -122.3953 longitude). Deployers will ensure that their GPS device or app has been set to use the North American Datum 83 (NAD83) geodetic reference system. The specifications for many GPS receivers indicate accuracy within 3 to 15 meters (10 to 50 feet) for 95% of measurements (http://www.gps-basics.com). Deployers will also document the make/model of the GPS unit used to obtain GPS coordinates. If a downloadable navigation application (app) on a smart phone is used to obtain GPS coordinates, the name and manufacturer of the app must be noted.

### Retrieval

Mussel retrieval will take place during MLLW periods within a specific range of dates to be announced by WDFW (see Table 9). WDFW staff and volunteers and Pierce County (hereafter collectively called the "retrievers") must remove their monitoring cages during the WDFW designated low tide period. Arriving on site at the time of MLLW ensures that retrievers can find and remove the mussel cage when it is totally exposed, with plenty of time to work before the incoming tide.

Upon arrival at the caged mussel site, the retrievers will take a digital photo of the cage, to document its condition, including structural integrity and degree of biofouling. Afterwards the retrievers will fill out the small retrieval section of the 2015/16 RSMP Mussel Monitoring Site Datasheet (Appendix D).

After field measurements, while wearing nitrile laboratory gloves, the retrievers will remove the four bags of mussels from the cage, keeping the mussels in the bags and the mesh intact, and place the bagged mussels immediately into a large, pre-labeled Ziploc bag(s). The Ziploc bag(s) will be placed into a cooler with bagged ice. This double barrier bagging method will ensure that mussels do not come into contact with any ice melt water during holding.

The cages and ALL anchoring devices and other paraphernalia will be removed from the beach; nothing from the monitoring project should be left behind. Upon finishing the removal the retrievers will fill out the bottom half of the matching Chain of Custody (COC) form (Appendix E), which will be kept with the cooler until it is delivered to the WDFW Marine Resources Laboratory in Olympia the following morning (see address below).

### Mussel Transport

Retrievers will hold the mussels overnight on ice in a cooler. Care will be taken to avoid freezing the mussels during holding (i.e. do not leave the cooler outside if the temperature drops below freezing). The retrievers will deliver the live mussels and matching COC form to WDFW for processing the morning following retrieval. Mussels should be delivered as early as possible to the WDFW Marine Resources Laboratory in Olympia (see address below), to ensure adequate time to process the mussels in the laboratory, especially if multiple cages are to be processed in one day.

Deliver mussels to: WDFW - Marine Resources Laboratory 1111 Washington St SE, 6th Floor Olympia, WA 98504-3150

# Laboratory Processing of Mussels

This section describes the laboratory measurement processes to be conducted by WDFW staff and volunteers. Data generated as described in this section will be entered into Excel spreadsheets and verified for accuracy. Results will be entered into WDFW's PSEMP database by WDFW staff.

## Lab Forms

Two forms will be used to track mussel samples as they are processed in the lab: *the Specimen Form* (Appendix F) records information and biological metrics for each mussel that is processed for a composite sample, while the *Tissue Resection Logs* (Appendix G) is used to document which individual mussels are included in each composite sample. These forms will be printed on waterproof paper to facilitate use in the lab environment. In addition a daily log (lab notebook) of operations will be maintained to record each day's activity, including the number of samples processed, observations, problems, resolutions, etc.

## **Equipment Cleaning Procedure**

Anything that may contact portions of a mussel subject to contaminant analysis will be cleaned before use. A "clean" work surface (lab counter, cutting board, sorting tray, instruments, etc.) will be covered by at least one layer of new aluminum foil, which will be changed between composites. "Clean" stainless steel dissection tools and grinding apparatus (hand grinder and cutting blades) will be 1) washed in warm soapy water (Terg-A-Zyme®), 2) thoroughly rinsed three times under warm running tap water, 3) rinsed with deionized water (held in Teflon squeeze bottle), 4) rinsed with isopropyl alcohol (held in a Teflon squeeze bottle), and then 5) placed on aluminum foil for air drying.

The same clean instruments/surfaces will be used repeatedly, without re-cleaning, on mussels contributing to the same composite. Afterwards, these instruments/surfaces will be subjected to the complete cleaning procedure prior to the processing of a new composite. Lab personnel will change nitrile gloves between composites.

## Mussels for Mortality, Condition Index, and Chemistry

Each mussel site will be represented by a cage that contains four individually numbered bags of mussels (64 individuals). WDFW lab staff will receive cages and bags of mussels the day after retrieval and complete the field portion of the COC form. WDFW lab staff will then determine the mortality in each mussel bag and select a random set of 12 mussels from the four bags to measure condition index. The remaining live mussels will be stored in a labeled plastic Ziploc type bag at -20°C until tissue resectioning for chemical analysis can take place. The length of mussel storage between retrieval and chemical analysis will not exceed three months.

### Mortality

WDFW lab staff will assess individual mussel bags for dead or moribund mussels within 36 hours of receiving the mussels. Dead or moribund mussels will be counted, recorded and removed. Mussels will be considered moribund if the animal is unable to tightly close its valves when stimulated. Mussels will be considered dead if there is no soft tissue inside the valves, or if the mussel soft tissue inside is putrefied.

### **Condition Index**

After dead mussels have been removed, condition index will be determined on 12 randomly selected mussels, according to the method reported by Kagley (2003) as follows:

Condition index (CI) = dry weight (g) of soft tissue/shell length (mm) X 100.

If needed, byssal threads and barnacles will be removed from the shell of the mussels prior to measuring, to prevent exterior debris from interfering with measurements. Shell length will be measured from the umbo to the farthest posterior margin (Figure 10) to the nearest tenth of a millimeter (0.1 mm) using a digital caliper. Total Shell Length (TSL) will be recorded on Specimen Forms.

Mussels will be opened by inserting a scalpel blade between the bivalve shells and severing the posterior and anterior adductor muscles (Figure 11). The shells will be spread apart at the hinge

to reveal the soft tissue. At this point, the remaining byssal fibers will be cut from the byssal gland using scissors. Then, if necessary, the tissue will be gently rinsed of sediment and foreign material with care not to lose pieces of tissue, using a Teflon squeeze bottle filled with DI water. After draining excess water, a scalpel will be used to scrape all the mussel soft tissue (including the adductor muscle) from the shell onto a pre-weighed drying pan. The wet weight of the soft tissue will be measured to the nearest tenth of a gram (0.1g) using a bench scale and recorded on the Specimen Form. Pans of mussel tissue will then be placed in a drying oven set at 120°C until the weight is constant (approximately 18 hours). After cooling to room temperature the resulting dry weight will then be recorded to the nearest tenth of a gram (0.1g) on the Specimen Form.

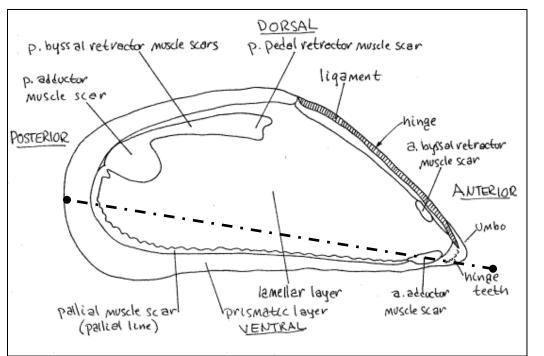


Figure 10. External anatomy of *Mytilus edulis* (Ruppert, Fox, and Barnes 2004).

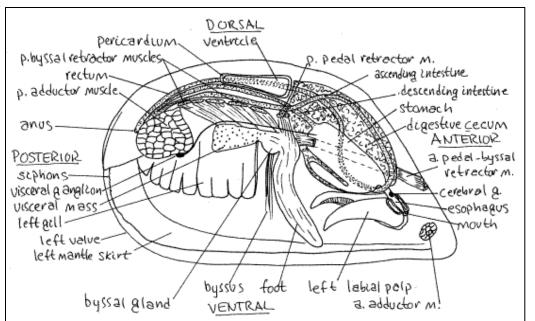


Figure 11. Internal anatomy of *Mytilus edulis* (Ruppert, Fox, and Barnes 2004).

### Preparing Composite Samples for Chemical Analysis

Previously frozen mussels will be thawed and prepared for tissue resectioning using the following procedure, which is a modification of Field Procedure 11.7 from the *Standard Guide for Conducting In-situ Field Bioassays with Caged Bivalves* (ASTM E2122-02, 2007). WDFW lab staff will wear clean nitrile gloves and change gloves between each sample. Lab staff will also maintain two sets of instruments per site; one set of tools to open the mussel, and one set of tools to remove tissue from the shell into the jar.

Prior to shucking the mussels for the soft tissue, byssal threads, sediment, biofouling, and barnacles will be removed from the shell of the mussels using scissors and gloved hands. Mussels will be rinsed several times with DI water to further remove external debris to reduce the risk of cross contamination after the mussels are opened.

Once cleaned and thawed sufficiently, lab staff will open each mussel by inserting a clean scalpel blade between the bivalve shells, severing the posterior and anterior adductor muscles (Figure 22). The shells will be spread apart at the hinge to reveal the soft tissue. The remaining byssal fibers will then be trimmed from the byssal gland using scissors. If necessary, the tissue will be gently rinsed of sediment and foreign material with care not to lose pieces of soft mussel tissue using a Teflon squeeze bottle filled with DI water. Excess water will be allowed to drain from the specimen. Using a scalpel, all soft tissue (including the adductor muscle) will be scraped into a clean I-CHEM (Class 200) glass sample jar.

Tissue from approximately 32 individual mussels from each sample site will be combined into a single pre-labeled composite sample jar, with the goal of collecting approximately 200 grams of tissue for each composite sample. Each mussel's tissue weight will be recorded on the Tissue Resection Log as it is added to the jar. After 32 mussels are added to the jar the total tissue weight will also be recorded. Each composite sample will then be frozen for later

homogenization. Unused whole mussels and cleaned (empty) mussel shells will be placed into a labeled Ziploc bag and re-frozen until the conclusion of the study.

After creation of composite samples, tissues will be ground in their original jars until a homogenous mixture is achieved. Partially thawed samples will be ground using a Bamix hand mixer to a consistency resembling pudding. Homogeneity will be determined by visual inspection. Once homogenized, subsamples will be placed in smaller I-Chem jars to allow for distribution of samples between several labs and for sample archiving.

### Sample Storage

All mussel composite samples and subsamples will be labeled and frozen to -20°C and held in a WDFW Marine Laboratory freezer until transfer to the analytical labs or their final archival destination. The location and conditions of all mussel composite samples will be recorded in a standard laboratory notebook used to track tissue samples for the WDFW-PSEMP program. The temperature of the WDFW-PSEMP program freezer is set at -20° C and is continuously monitored through data loggers tracked by Washington State Enterprise Services. Any temperature anomalies will trigger an alarm, triggering on-site maintenance staff to contact a laboratory supervisor from a priority list of supervisors, for immediate attention. In addition, this freezer is backed up by emergency generators in case of power outage.

# **Chemical Analyses**

## **Number of Samples**

The maximum number of samples to be submitted for chemical analysis in this study is expected to be 54; 40 RSMP samples, eight Pierce County samples, and six baseline samples. It is expected that the POPs analysis will also generate five laboratory quality control samples.

## Sample Preparation Method(s)

Homogenized composite mussel tissue samples will be shipped to the analytical labs frozen. The analytical labs will thaw and thoroughly mix the tissue samples with clean utensils to ensure adequate homogeneity prior to sample preparation for chemical analysis.

# Analytes

The POPs, metals, and conventional analytes to be measured are listed in Tables 11, 12 and 13. Composited somatic mussel tissue will be the only matrix analyzed for chemical contaminants.

## Persistent Organic Pollutants (POPs)

All POPs in this study will be analyzed according to Sloan et al. (2014). This analytical method is consistent with previous WDFW studies. In brief, this method comprises three steps: (a) extraction, (b), cleanup by silica/aluminum columns and size-exclusion high-performance liquid chromatography (SEC HPLC), and (c) quantitation of chlorinated hydrocarbons (CHs) and aromatic hydrocarbons (AHs) using gas chromatography /mass spectrometry (GC/MS) with selected-ion monitoring (SIM). Samples are extracted using accelerated solvent extraction (ASE with methylene chloride), which provides an extract that can be used for AH, CH recovery and gravimetric lipid evaluation. This method also includes alterations to typical GC/MS methods to stabilize the instrument and improve accuracy such as chemical ionization filaments (to increase source temperature), employing a cool on-column injection system in the GC, a guard column before the analytical column, and point-to-point calibration to improve data fit over the full range of GC/MS calibration standards (Sloan et al. 2014).

### Sensitivity: Limit of Quantitation (LOQ) and Method Detection Limit (MDL)

For all POPs in this study the lower Limit of Quantitation (LOQ) "for a given analyte in a given sample is the concentration that would be calculated if the analyte had a GC/MS response area equivalent to that analyte's area in the lowest level CS used in the calibration for that analyte (not all levels are used for some analytes). When an analyte is not detected in a sample or has an area that is smaller than its area in the lowest level CS used, the concentration of the analyte in that sample is reported to be less than the value of its LOQ." (Sloan et al. 2014). Typically LOQ values for POPs that have been reported to WDFW by this method are in the range of 0.2 to 0.8 ng/g wet weight (Table 11).

EPA defines Method Detection Limit (MDL) in Appendix A to 40 CFR Part 136 as the "minimum concentration of a substance that can be measured and reported with 99 percent confidence that the analyte concentration is greater than zero and is determined from analysis of

a sample in a given matrix containing the element". In this study, the metal's MDLs are concentrations that cannot be detected or detected at a concentration less than the associated method detection limit considering tissue sample detection limits are affected by the sample mass used, matrix and polyatomic/isobaric interferences. The MDL is the lowest concentration at which a sample result will be reported. Table 12 lists the respective method detection limits for the metals of concern in this study (Hg, As, Cu, Zn, Cd, and Pb). They range from 0.10 to  $0.00038 \mu g/g$  wet weight.

Persistent organic pollutants (POPs):			Limit of Quantitation - LOQ (wet weight)	Expected Range (wet weight)
Polychlorinated biphenyl (PCB) congeners	40	Sloan et al. 2004	0.2-0.8 ng/g	LOQ to 20 ng/g
Polybrominated diphenylethers (PBDEs) congeners	11	Sloan et al. 2004	0.2-0.8 ng/g	LOQ to 20 ng/g
Organochlorine pesticides (OCPs)	25	Sloan et al. 2004	0.2-0.8 ng/g	LOQ to 20 ng/g
Polycyclic Aromatic Hydrocarbons (PAHs)	45	Sloan et al. 2004	0.2-0.8 ng/g	LOQ to 20 ng/g

Table 11. Persistent organic pollutants (POPs) to be measured in this study.

### Expected range of results

The range of concentrations for POPs in this study is from the LOQ (typically between 0.2 and 0.8 ng/g wet weight) to 20 ng/g wet weight for individual PCB or PBDE congeners, OCP isomers, or PAH analytes. The range of concentration of metals should be from the limit of detection (approximately 0.005  $\mu$ g/g) to 5  $\mu$ g/g wet weight.

## Metals

All metals analyses will be performed by the King County Environmental Laboratory (KCEL). The metals mercury, arsenic, cadmium, copper, zinc, and lead will be analyzed by two methods. Mercury will be analyzed via automated cold vapor atomic absorption spectrometry following King County Environmental Laboratory Standard Operating Procedure (KCEL SOP) 604. This SOP incorporates elements of EPA 245.1 revision 3, SW-846 7470, 7471B and PSEP 1997. Arsenic, cadmium, copper, zinc, and lead will be analyzed via Thermo Elemental X Series II CCT (Collision Cell Technology) Inductively Coupled Plasma Mass Spectrometer (ICP-MS) following KCEL SOP 624. This SOP incorporates elements of EPA 200.8 revision 5.4, SW-846 6020A February 2007, ILM05.3 Exhibit D part B, and PSEP 1997. Total solids will be analyzed via KCEL SOP 307v3 to facilitate reporting metals data in both dry and wet weight concentrations.

	No.		Method Detection	Expected Range
Metals	Analytes	Method	Limit (wet weight)	(wet weight)
Total mercury (Hg)	1	KCEL SOP 604 <sup>b</sup>	0.00038 µg/g	MDL to $5 \mu g/g$
Lead (Pb)	1	KCEL SOP 624 <sup>c</sup>	0.004 µg/g	MDL to 5 µg/g
Arsenic (As)	1	KCEL SOP 624	0.004 µg/g	MDL to $5 \mu g/g$
Zinc (Zn)	1	KCEL SOP 624	0.10 µg/g	MDL to $5 \mu g/g$
Copper (Cu)	1	KCEL SOP 624	0.008 µg/g	MDL to $5 \mu g/g$
Cadmium (Cd)	1	KCEL SOP 624	0.002 µg/g	MDL to $5 \mu g/g$

Table 12. Metals to be measured in this study.

<sup>b</sup> KCEL SOP 604; <sup>c</sup> KCEL SOP 624

## Conventionals

Lipid content will be performed by NOAA. Samples will be extracted using accelerated solvent extraction (ASE with methylene chloride), which provides an extract that can be used for gravimetric lipid evaluation (Sloan et al. 2014). Percent solids (total solids) analyses will be performed by the KCEL. Total solids will be analyzed gravimetrically using Standard Methods 2540-G as described below.

Conventional parameters	No. Analytes	Method	Method Detection Limit (wet weight)	Expected Range (wet weight)
Lipid content (% total extractables)	1	gravimetric	0.1%	0.5 to 3%
Dry Weight (%)	1	gravimetric	0.005%	10-20%

Table 13. Conventionals to be measured in this study.

## Stable Isotopes

Stable isotopes of carbon (<sup>13</sup>C) and nitrogen (<sup>15</sup>N) will be measured by Mass Spectrometry (following Herman et al. 2005) after preparation as follows:

- 1. Homogenized tissue samples freeze-dried overnight
- 2. Freeze-dried tissue pulverized in a micro-ball mill
- 3. 0.4 to 0.6 mg powder of each sample placed into separate tin cups, in triplicate
- 4. Combusting samples in a Costech elemental analyzer attached to a Thermo-Finnegan Delta Plus Isotope Ratio Mass Spectrometer

Values are calibrated with internal standards every ten samples. Unenriched histidine is used as a control material to evaluate set-to-set reproducibility, analyzed after every 25 samples. Stable isotope results are expressed in "delta" ( $\delta$ ) notation in ‰:

$$\delta Z = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1000 \text{ (1)},$$

where Z is  ${}^{15}$ N or  ${}^{13}$ C,

 $R_{\text{sample}}$  is the ratio  ${}^{15}\text{N}/{}^{14}\text{N}$  or  ${}^{13}\text{C}/{}^{12}\text{C}$  for the tissue sample, and

 $R_{standard}$  is the ratio of  ${}^{15}N/{}^{14}N$  or  ${}^{13}C/{}^{12}C$  of standards (atmospheric air for nitrogen and Pee Dee Belemite limestone for carbon.

## **Percent Lipids**

Percent lipids in each sample are represented by total extractables, according to Sloan et al. 20042014. Briefly samples from the extraction step of the POP analyses will be evaporated and compared to the mass of the original, unextracted sample (paraphrasing from Sloan et al. 2014):

- The pan containing the sample for total extractables from Section 3 is placed on a covered rack in the hood and the solvent is allowed to completely evaporate (approximately 1–2 hours).
- The pan is dried in a 50°C oven for 2 hours, then cooled in a desiccator overnight.
- The pan is weighed to the nearest 0.0001g and the weight is recorded as the "Pan w/TE" weight.
- The percent total extractables (% TE) content of the sample is calculated as follows:

% TE = [(Pan w/TE – Pan) x (ASE Vial w/Extract – ASE Vial) x 100%]/[(ASE Vial w/Extract – ASE Vial w/o TE Extract) x Sample Weight].

## Percent Solids (Dry Weight) Determination

The percent of the sample as dry weight is determined by simple drying of tissues according to Standard Methods 2540-G (paraphrasing):

- Pre-homogenized tissue (1 + 0.5 g) is placed into the pan, and the pan is weighed to the nearest 0.0001 g. The weight is recorded as the "Pan w/Wet Sample" weight.
- The pan is placed in a drying oven at 105°C for 4 hours to overnight, then cooled in a desiccator for at least an hour. The pan is weighed to the nearest 0.0001 g, and the weight is recorded as the "Pan w/Dry Sample" weight.
- The percent dry weight of the sample is determined as follows:

% Dry Weight = [(Pan w/Dry Sample – Pan) x 100%]/(Pan w/Wet Sample – Pan).

# **Quality Control Procedures**

All mussels used for the RSMP study and those used by Pierce County, will come from a single Puget Sound aquaculture facility (Penn Cove Shellfish, Inc., Whidbey Island). Thus all the mussels deployed to RSMP and Pierce County study sites will originate from the same population, be of a similar age, have a similar genetic and environmental history and are expected to be relatively uncontaminated. In addition, all composite samples of mussels produced from RSMP and Pierce County sites will be analyzed by the same two laboratories (see Table 3).

Once the mussels have been collected and delivered to the WDFW Marine Resources Lab they will no longer be under Pierce County control. At that point WDFW and the RSMP contracted labs will have control of the samples and responsibility for laboratory quality control (QC) procedures. Laboratory processing and analysis of both the RSMP and Pierce County mussel samples will be performed by WDFW.

# Field QC

Field personnel will follow measurement and QC methods specified in Table 5, to obtain consistent field measurements specified in this QAPP. Training on mussel deployment, retrieval, and how to take field measurements will be provided by WDFW staff by the summer of 2015. This training will take the form of a webinar or document (i.e. self-train), to ensure comparability of results between the Pierce County, WDFW staff and volunteers.

Field personnel will ensure photos are taken of the fully installed mussel cage, for verification of proper technique. In addition, field personnel are expected to fill in ALL sections of the 2015/16 RSMP Mussel Monitoring Datasheet (Appendix D), as well as in the Chain of Custody Form (Appendix E) provided in this QAPP. Field personnel will perform in-field reviews of their datasheets before leaving the study site, to ensure all data is recorded correctly.

## **Instrument Check**

A GPS accuracy of 5-10 meters (15-30 feet) will provide adequate representation of the physical location of collected mussels. Field personnel will ensure that backup GPS units are available in the field should the unit currently in use fail.

## WDFW Processing Laboratory QC

All laboratory data generated by WDFW during mussel processing will be examined visually using Excel filters and sorting procedures to identify gross formatting or transcription errors. Data values will be compared with expected ranges to identify potential outliers. In addition preliminary tables of summary statistics and scatter plots will be created to examine the data.

# Analytical Laboratory QC

Quality control procedures, quality assurance criteria and corrective actions for POPs data are detailed in Sloan et al. (2014). Briefly, precision is monitored and controlled within batches using laboratory replicates of field samples (2 replicates run for every batch of 12 samples) and across batches by analyzing Standard Reference Materials (SRMs –one per batch). Cross-batch precision is expressed as the relative standard deviation (RSD) for repeated measurements. The RSD of analyte responses relative to the internal standard must be  $\leq 15\%$  for the repetitions. For POPs analysis, accuracy of samples is evaluated by comparing measured SRM values with National Institute of Standards and Technology (NIST) certified values. A SRM of applicable matrix will be selected to be analyzed i.e., tissue. Concentrations of  $\geq 70\%$  of individual analytes are to be within 30 % of either end of the 95% confidence interval of the reference values. One method blank is run for every 20 or fewer field samples. No more than 5 analytes in a method blank are to exceed 2x the lower LOQ before corrective action is taken. The corrective action will be to re-extract and re-analyze the affected samples and if necessary, qualify the sample data. At least one internal standard (surrogate) is added to each sample, with acceptable recoveries ranging from 60 to 130%.

Quality control measure and quality assurance criteria for metals data are detailed in Table 7 and Table 8. Briefly, precision is monitored and controlled within batches using laboratory replicates of field samples (one per batch). Accuracy of analysis is evaluated by comparing measured standard reference material (SRM) values and a laboratory control sample (LCS) with the respective certified values. A SRM of applicable matrix will be selected to be analyzed i.e., tissue. Method blanks and spikes are evaluated for overall run and process contamination. These are run every batch as is applicable.

All analytical laboratory data will be examined visually using Excel filters and sorting procedures to identify gross formatting or transcription errors. Raw analyte concentrations will be compared with expected ranges to identify potential outliers. In addition preliminary tables of summary statistics, scatter plots, and time trend plots will be created to examine the new data.

# Data Management

WDFW will format all digitized field and laboratory data into a structure compatible with the PSEMP-Toxics in Biota (TIB) database. The TIB database is a relational database created in Access, with separate tables for (1) field effort data, (2) biological characteristics of individuals used to create samples, (3) many-to-many cross reference for individuals-to-composites, (4) sample tracking, condition and summary statistics, and (5) chemical analyses. The TIB database is stored on a WDFW server, which is backed up nightly as part of an automated network backup service provided by WDFW Information Technology (IT) Services.

# Field Data

WDFW staff and volunteers and Pierce County field personnel will be collecting and managing data from field work during deployment and retrieval of mussel samples. All data will be managed and stored by the field personnel responsible for each site. Field measurements and observations will be recorded on the 2015/16 RSMP Mussel Monitoring Site Datasheet (Appendix D) printed on waterproof paper. A new field datasheet will be completed at every mussel monitoring site, and data on sites rejected during reconnaissance will be recorded in a separate Field Log.

Field data will be digitized (placed into Excel spreadsheets) and all entries will be independently verified for accuracy by another individual on the project team. This data will be incorporated into annual reports and electronic reports by WDFW and Pierce County (see Monitoring Reports section below). Reports and data will be submitted to Ecology in the format required.

## Audits

The WDFW mussel monitoring lead will routinely coordinate all activities with staff and volunteers to ensure the field sampling locations are suitable, deployment and retrieval of mussels and the COC form is properly filled out. Laboratories will alter the WDFW lead if timeframes are not met, or samples are lost. The WDFW will take corrective actions where necessary to ensure adequate timeframes and safe sample delivery.

## Laboratory Data

WDFW staff will digitize (place into Excel spreadsheets) laboratory measurements and observations recorded on Specimen Forms (Appendix F) and Tissue Resection Logs (Appendix G). All entries will be independently verified for accuracy by another individual on the project team.

Data received from the analytical laboratories will be in Excel spreadsheets in various formats. WDFW staff will format these data into a structure compatible with the TIB database and incorporate the data accordingly.

## **Data Storage**

All datasheets, photographs, and printed or electronic data generated for this project will be stored by WDFW and Pierce County in organized filing systems for paper and electronic files. These files may be sought by Ecology for permit compliance review and audit purposes and must be maintained according to the records retention requirements for all documents related to the permits. Location and measurement data will be evaluated through the data verification process outlined in this QAPP. Acceptable results will be used by scientists to prepare a summary report and entered into Ecology's EIM database.

# **Data Verification and Quality Assessment**

WDFW and Pierce County project leads will examine and verify all field-generated data to ensure:

- Specified methods and protocols were followed.
- Data are consistent, correct, and complete, with no errors or omissions.
- Data specified in the Sampling Process Design section were obtained.
- Results for QC samples as specified in the *Measurement Quality Objectives* and *Quality Control* sections accompany the sample results.
- Established criteria for QC results were met.
- Data qualifiers are properly assigned where necessary.

# Field Data

Throughout the duration of field sampling, the field personnel leads and crew members are responsible for implementation of sample-collection procedures. The field lead is also responsible for a systematic review of all field documentation generated (e.g., datasheets, field logs, chain-of-custody sheets, sample labels) to ensure data entries and labels are consistent, correct, and complete, with no errors or omissions. This review should be completed prior to leaving the site where the measurements were made.

Data usability assessment follows verification. This involves a detailed examination of the data package using professional judgment to determine whether the quality objectives have been met. WDFW and Pierce County project managers will examine the complete field data packages (i.e. hard copy datasheets and Excel spreadsheets) to determine compliance with procedures outlined in this QAPP and referenced SOPs. WDFW and Pierce County project managers will also ensure that the MQOs have been met and determine if the quality of the field data is usable for the RSMP objectives.

## Laboratory Data

Data generated by the analytical labs will be reviewed by analytical lab staff for out-of-bounds values, transcription errors and other problems by at least two chemists. Final review is conducted by a lab manager who approves data before they are released to the client. Prior to database entry WDFW will review the data by comparing results with similar species or matrices in the PSEMP-TIB database. Individual data, means, and standard deviations will be plotted and putative outliers evaluated for validity. Evaluation of the validity of putative outliers will include reviewing all collection, biological, and analytical data for potential transcription errors, communication with analytical labs to verify reported values are correct, and evaluation of biological covariates that might explain otherwise unanticipated values.

The success of meeting data quality objectives is evaluated based on the outcome of quality control procedures during analytical procedures. Typically if QC criteria are not met the

problem is identified, corrected, and sample (or extract) re-run. In cases where QC criteria have not been met and there is not enough tissue to be reanalyzed, the data are to be censored with appropriate qualifiers to allow an objective evaluation of the usability of the final record. Rejected data are censored with an "R" or equivalent qualifier. Based on (1) a long history of employing these methods to measure target analytes in a wide range of Puget Sound biota matrices, (2) the range of data values we expect in this study, and (3) appropriate (tenth-of-ppb) limits of quantitation, we expect rejected data to be rare, with the singular possible exception of potential blank contamination for naphthalene-compounds.

Non-detected analytes will be censored with a "<LOQ" or "U" qualifier. The value reported for non-detected analytes will be the LOQ or Method Detection Limit, depending on analytical procedure. It is the responsibility of data users to decide how to use data censored as not-detected. Previous experience with data from similar studies for the target analytes in this study suggest that summed totals will be dominated by substantial concentrations of a number of individual analytes.

# **Monitoring Reports**

## 2015/16 RSMP Mussel Monitoring Progress Report

WDFW staff will provide a progress report of the 2015/16 RSMP mussel monitoring effort, in the form of an oral presentation, to the Stormwater Work Group (SWG) in the summer of 2016. This progress report will include an update of work-to-date on the RSMP mussel monitoring project and recommendations for future changes to the program.

## 2015/16 RSMP Mussel Monitoring Summary Report

WDFW will produce a summary report on the biological, chemical, and geographic data from the 2015/16 RSMP mussel monitoring survey and Pierce County's 2015/16 mussel monitoring survey, due to Ecology on June 30, 2017. This report will include an assessment of the extent and magnitude of chemical contamination of mussels in UGAs of the Puget Sound, tables and graphs with summary statistics, maps of contaminant distributions, and recommendations for refining future rounds of RSMP monitoring. In addition, RSMP mussel monitoring results will be compared with results from WDFWs Toxic Contaminants in Puget Sound's Nearshore Biota: A Large-Scale Synoptic Survey Using Transplanted Mussels (Mytilus trossulus) (Lanksbury et al. 2014) report, where appropriate. The format will be a WDFW agency report.

## **Pierce County Mussel Monitoring Reports**

Pierce County must provide a detailed summary of the previous calendar year's mussel monitoring activities. This detailed monitoring report is due to Ecology as an attachment to the permittee's annual stormwater monitoring report, due on March 31 of 2016, 2017, and if needed, 2018. The report must include all information listed below. All associated data will also be uploaded to Ecology's EIM database and made available to the public via Ecology's web site (www.ecy.wa.gov/eim/myEIM.htm). The information contained in Pierce County's 2015/16

mussel monitoring summary report will be incorporated into the 2015/16 mussel monitoring summary report produced by WDFW.

Pierce County's project lead is responsible for describing their mussel monitoring efforts. The monitoring report will include a complete discussion of the mussel monitoring effort and must include the items detailed below in Table 14.

Table 14. Reporting requirements.						
Category	Reporting Requirement					
<u>0''</u>	Documentation of the site confirmation process, including desktop evaluation and field visits for each of the required number of assigned sites.					
Site Confirmation	List of sites disqualified and specific reasons for disqualification.					
Commination	List of final sites. In a table, provide final GPS coordinates for each site and the distances from the initial GPS locations provided in the Master Sample					
	Description of upland land use adjacent to the site sampled.					
	Description of intertidal habitat, substrate, and vegetation at the site sampled.					
Site Information	Description of man-made structures on the beach or in the water at the site sampled.					
	Field measurements and observations at each site.					
Site	Deployment and retrieval information (date, time, weather, mussel bag numbers, anchors used etc.).					
Activities	Field measurements (water temperature, salinity).					
	Photo documentation.					
Concerns	Narrative description of any deviations from this QAPP, including any delays, problems, and resolutions in conducting required monitoring activities.					
Costs	Estimated monitoring costs for each required monitoring program component.					
Signature	Designated official (General Condition G19) signature					

#### Table 14. Reporting requirements.

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# **Glossary, Abbreviations and Acronyms**

### Glossary

**National Pollutant Discharge Elimination System (NPDES):** National program for issuing, modifying, revoking and reissuing, terminating, monitoring, and enforcing permits, and imposing and enforcing pretreatment requirements under the Clean Water Act. The NPDES program regulates discharges from wastewater treatment plants, large factories, and other facilities that use, process, and discharge water back into lakes, streams, rivers, bays, and oceans.

**Pollution:** 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.

**Stormwater:** The portion of precipitation that does not naturally percolate into the ground or evaporate but instead runs off roads, pavement, and roofs during rainfall or snow melt. Stormwater can also come from hard or saturated grass surfaces such as lawns, pastures, playfields, and from gravel roads and parking lots.

### Acronyms and Abbreviations

app	application (downloadable onto cellular phones)
As	Arsenic
CI	Condition index
COC	Chain of custody
Cd	Cadmium
Cu	Copper
DI	Deionized (water)
DNR	Washington Department of Natural Resources
Ecology	Washington State Department of Ecology
EAP	Environmental Assessment Program
EIM	Environmental Information Management system
EPA	U.S. Environmental Protection Agency
GIS	Geographic information system software
GPS	Global positioning system
GRTS	Generalized random tessellation stratified
Hg	Mercury
HPA	Hydraulic Project Approval
KCEL	King County Environmental Lab
MLLW	Mean lower low water
MOU	Memorandum of understanding
MQO	Measurement quality objective
MQO	0

NAD83 NIST NOAA NPDES NWFSC OCP PAH Pb PBDE PCB POPs PSAMP PSEMP PSEMP PSP QA QAPP QC RSD RSMP SOAL SOP SRM SUBgroup SWAMPPS SWG	North American Datum 83, geodetic reference system National Institute of Standards and Technology National Oceanic and Atmospheric Administration National Pollutant Discharge Elimination System Northwest Fisheries Science Center Organochlorine pesticide Polycyclic Aromatic Hydrocarbon Lead Polybrominated diphenylethers Polychlorinated biphenyl Persistent organic pollutants Puget Sound Assessment and Monitoring Program (now PSEMP) Puget Sound Ecosystem Monitoring Program (formerly PSAMP) Puget Sound Partnership Quality assurance Quality Assurance Project Plan Quality control Relative standard deviation Regional Stormwater Monitoring Program State-Owned Aquatic Land Standard operating procedure Standard reference material Marine Nearshore Status and Trends Subgroup Stormwater Assessment and Monitoring Program for Puget Sound Stormwater Work Group
Subgroup	
SWAMPPS	Stormwater Assessment and Monitoring Program for Puget Sound
SWG	Stormwater Work Group
TIB	Toxics in Biota group, part of PSEMP
UGA	Urban growth area
WDFW	Washington Department of Fish and Wildlife
Zn	Zinc

## Units of Measurement

°C	degrees centigrade
cm	centimeter
ft	feet
g	gram
km	kilometer, a unit of length equal to 1,000 meters
m	meter

# Appendices

## Appendix A. Regional Stormwater Monitoring Strategy

## Background

The Puget Sound Stormwater Work Group (SWG) was assembled in 2008 at the request of the Washington State Department of Ecology (Ecology) and the Puget Sound Partnership (PSP) to develop recommendations for a monitoring and assessment strategy to improve our understanding of the effects of stormwater in the Puget Sound region. In 2010, the SWG finalized the overall strategy for monitoring in the document *2010 Stormwater Monitoring and Assessment Strategy for the Puget Sound Region (SWAMPPS)* (SWG, 2010a). These recommendations (SWG, 2010b) were submitted to Ecology and the PSP for consideration in the development of an integrated stormwater monitoring program focused on the Puget Sound region. The 2010 Strategy included "55 Key Recommendations" for a new stormwater assessment and monitoring program.

The 2010 Strategy describes four components of a robust program: status and trends monitoring of receiving waters impacted by stormwater runoff; effectiveness studies to evaluate best management practices and programmatic approaches to manage stormwater; source identification and diagnostic monitoring to improve pollution reduction efforts; and research to increase knowledge of stormwater effects on biota and treatment approaches to reduce effects.

The SWG followed the 2010 Strategy with 33 recommendations for municipal permit monitoring (http://www.ecy.wa.gov/programs/wq/psmonitoring/ps\_monitoring\_docs/SWworkgroupDOCS/S WGfinalreportoct292010.pdf). These recommendations outlined a plan for implementing a core subset of the 2010 Strategy through municipal stormwater permits issued to local governments in Puget Sound.

### Status and trends- marine mussel monitoring design

### Goals

One of the goals of the RSMP nearshore status and trends monitoring program is to use marine mussels (*Mytilus* sp.) as an indicator species to evaluate contaminant conditions in Puget Sound's nearshore biota. The study design involves distributing cage-protected mussels from a common source along Puget Sound's shoreline to synoptically evaluate the geographic extent and magnitude of nearshore contamination. The goals include:

- 1. Assess the tissue contaminant concentrations of Puget Sound biota in the nearshore urban areas, defined as being inside established Urban Growth Area (UGA) boundaries.
- 2. Document geographic patterns.
- 3. Document natural and human-caused changes over time in Puget Sound nearshore biota.

- 4. Identify existing challenges to the health of nearshore biota and, where possible, provide data to help target sources.
- 5. Support nearshore research activities by making available uniformly collected, high quality data.
- 6. Provide nearshore data to assist the SWG, the PSP, and others in measuring the success of stormwater and other environmental management programs.

### Objectives

Specific objectives of nearshore mussel monitoring include:

- 1. Characterize the spatial extent of tissue contamination in nearshore biota residing inside the UGA sampling frame using mussels (*Mytilus* sp.) as the primary indicator organism.
- 2. Track changes in tissue contamination over time inside the UGA sampling frame to answer the question; is biota health improving, deteriorating, or remaining the same?

### Scale of Monitoring

Status and trends is intended to report results at a high level of statistical confidence; as such, a probabilistic random stratified sampling design was selected for the nearshore urban and non-UGAs. This approach was developed by EPA as a spatially-balanced, generalized random tessellation stratified (GRTS) multi-density survey design

(<u>http://epa.gov/nheerl/arm/designing/design\_intro.htm</u>) and is described by Stevens (1997, 2003, 2004), and Stevens and Olsen (1999). A Puget Sound shoreline sampling frame (which is linear) was generated by Sitka Technology Group, LLC using the stratified design and populated with sites for the stormwater permittees.

Monitoring for this QAPP is focused on a single landscape scale, the shoreline parallel to cities and UGAs. A shoreline sampling frame for Puget Sound was defined to include the basins, channels, and embayments of Puget Sound from the US/Canada border to the southern-most bays and inlets near Olympia and Shelton, to Hood Canal, and to portions of Admiralty Inlet, the San Juan Islands, and the eastern portion of the Strait of Juan de Fuca. The shoreline master sample sampling frame was targeted to the land-based UGA boundaries within the Puget Sound basin.

Sampling points were generated to populate the shoreline sampling and sub-sampling (linear) frames using the GRTS design, providing a random and spatially balanced site selection process. From this design the <u>Puget Sound Mussel Monitoring</u> sample draw was generated, resulted in a total of 2,048 sites in Puget Sound's UGAs. The first 100 sites in the Puget Sound Mussel Monitoring sample draw are shown in Figure 1; the first 50 sites in unincorporated Pierce Co UGAs are listed in Appendix B.

## Assumptions underlying the design

This monitoring program design is based on several assumptions; #1) for the purposes of assessing stormwater impacts, the study design characteristics take into account the desire for Puget Sound-scale estimates at a high confidence level (80-90%) and potential for stratification of samples into other categories (e.g., land uses). The confidence level (i.e. the reliability of the result) is determined by the variance of the indicator variable and the sample size within populations (www.epa.gov/nheerl/arm/surdesignfaqs.htm).

The SWG also assumes #2) that two assessment regions Urban Growth Area (UGA) and non-UGA are different. This assumption is based on the differences in stormwater management efforts required by permits inside UGA boundaries, and the differences in overall land use. Shorelines and nearshore areas in Puget Sound in urban and urbanizing areas are assumed to be more (or differently) influenced than shorelines and nearshore areas outside urban and urbanizing areas. The RSMP will monitor the shoreline and nearshore within the UGA assessment area. Data from prior WDFW mussel monitoring in areas considered non-UGA will be used for comparison, where available and appropriate.

This monitoring design also assumes #3) that the sites will be useable over the long term. The site layout is designed for a long-term monitoring program rather than for a targeted study. This study design assumes that general trends in nearshore ecosystem health can be described with the parameters outlined in this QAPP.

### Regional stormwater monitoring objectives

This monitoring framework is designed to answer the following core broad-scale monitoring questions:

- What are the status and trends of water quality and biota (i.e. mussel) tissue quality in Puget Sound Nearshore areas?
- What are the status and trends of the water quality and biota (i.e. mussel) tissue quality in Puget Sound nearshore areas adjacent to Urban Growth Areas (UGAs)?

In addition, site-specific evaluations of data can be useful for answering questions at local scales and will improve stormwater managers' understanding of nearshore condition and biota stressors.

### Coordination

A programmatic objective of the 2010 Strategy is to efficiently allocate limited resources for monitoring activities. Toward this objective the RSMP marine mussel monitoring is being conducted by WDFW who has conducted the vast majority of other marine mussel monitoring in Washington State. As such results will be comparable to prior monitoring results.

#### Scale of regional monitoring

Monitoring for this QAPP is focused on monitoring marine intertidal quality, using biotic endpoints, at two landscape scales:

- Puget Sound-wide
- Adjacent to Phase I and Phase II UGAs within the Puget Sound.

These areas are the focus of important stormwater management, resource conservation and protection efforts. Information generated for each of these regions can be useful to Ecology, local governments, and agencies managing aquatic resources that are impacted by stormwater. Since management for improvements usually occurs at a local scale, the RSMP monitoring design aims at providing information on the health of nearshore biota (i.e. mussels) and sediment quality at UGA or sub-basin scales. The focus on small watersheds in the nearshore environment is understood and readily used by local governments, who are likely to participate in data collection efforts and become users of data generated by the monitoring program.

#### Indicators

The SWG (SWG, 2010) recommended monitoring specific biota (i.e. mussels), habitat, and chemical indicators related to stormwater runoff and stormwater impacts. The basic list of parameters comes from existing state status and trends study designs. For this QAPP the mussel monitoring is most heavily based on findings from the recent WDFW Mussel Watch Pilot Expansion project (Lanksbury et al., 2014), which demonstrated that transplanted mussels can be used successfully on a large scale to characterize patterns of nearshore contamination in the greater Puget Sound. In that study transplanted mussels provided data on the extent and magnitude of contamination in Puget Sound nearshore environments and offered insight into how contamination in nearshore biota is related to upland land-use patterns.

## Appendix B. 2015/16 RSMP Puget Sound Mussel Monitoring Pilot Site List

This table shows the first 100 candidate sites on the mussel monitoring list. A complete RSMP site list is available on Ecology's RSMP website at <u>http://www.ecy.wa.gov/programs/wq/stormwater/municipal/rsmp/rsmpdocs/RSMP2015-Musselsites.xlsx</u>, and contains more accompanying information.

USE_ORDER	SITE_ID	REGION	COUNTY_NM	CITY_NM	UGA_NM2	LAT_DD	LON_DD
1	PSS13175-000001	South Sound	Thurston	Olympia	Olympia - Incorporated UGA	47.04765	-122.91126
2	PSS13175-000002	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.50204	-122.38594
3	PSS13175-000003	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.68262	-122.50706
4	PSS13175-000004	Strait of Georgia	Whatcom		Cherry Point - Unincorporated UGA	48.85755	-122.7363
5	PSS13175-000005	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.29181	-122.52806
6	PSS13175-000006	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.61871	-122.52759
7	PSS13175-000007	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.64877	-122.4175
8	PSS13175-000008	Admiralty Inlet	Jefferson		Jefferson Co Unincorporated UGA	48.04868	-122.77652
9	PSS13175-000009	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.25521	-122.37604
10	PSS13175-000010	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.64458	-122.57753
11	PSS13175-000011	Strait of Georgia	Whatcom	Bellingham	Bellingham - Incorporated UGA	48.72568	-122.50606
12	PSS13175-000012	Whidbey Basin	Island	Oak Harbor	Oak Harbor - Incorporated UGA	48.2969	-122.57945
13	PSS13175-000013	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.29253	-122.4951
14	PSS13175-000014	Central Sound	Kitsap	Bremerton	Bremerton - Incorporated UGA	47.57101	-122.60648
15	PSS13175-000015	Strait of Georgia	Skagit	Anacortes	Anacortes - Incorporated UGA	48.4923	-122.67746
16	PSS13175-000016	Central Sound	Snohomish	Edmonds	Edmonds - Incorporated UGA	47.85424	-122.33472
17	PSS13175-000017	South Sound	Thurston	Olympia	Olympia - Incorporated UGA	47.06878	-122.91975
18	PSS13175-000018	Central Sound	King	Burien	Burien - Incorporated UGA	47.46333	-122.36868
19	PSS13175-000019	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.66154	-122.49952
20	PSS13175-000020	Eastern Strait of Juan de Fuca	Clallam	Port Angeles	Port Angeles - Incorporated UGA	48.1178	-123.42336
21	PSS13175-000021	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.30376	-122.51146
22	PSS13175-000022	Central Sound	Kitsap		Port Orchard - Unincorporated UGA	47.55888	-122.59715
23	PSS13175-000023	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.62206	-122.49572
24	PSS13175-000024	Admiralty Inlet	Jefferson		Jefferson Co Unincorporated UGA	48.0268	-122.74896
25	PSS13175-000025	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.27454	-122.41519
26	PSS13175-000026	Central Sound	Kitsap		Central Kitsap - Unincorporated UGA	47.60311	-122.59829
27	PSS13175-000027	Strait of Georgia	Whatcom	Bellingham	Bellingham - Incorporated UGA	48.68975	-122.50434
28	PSS13175-000028	Whidbey Basin	Island	Oak Harbor	Oak Harbor - Incorporated UGA	48.27141	-122.63749

USE_ORDER	SITE_ID	REGION	COUNTY_NM	CITY_NM	UGA_NM2	LAT_DD	LON_DD
29	PSS13175-000029	Central Sound	Kitsap	Poulsbo	Poulsbo - Incorporated UGA	47.74626	-122.65216
30	PSS13175-000030	Central Sound	Kitsap	Port Orchard	Port Orchard - Incorporated UGA	47.54111	-122.64058
31	PSS13175-000031	San Juan Archipelago	San Juan		Eastsound - Unincorporated UGA	48.69258	-122.91127
32	PSS13175-000032	Whidbey Basin	Snohomish	Everett	Everett - Incorporated UGA	47.97529	-122.22664
33	PSS13175-000033	South Sound	Pierce	DuPont	DuPont - Incorporated UGA	47.10396	-122.67593
34	PSS13175-000034	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.5871	-122.35304
35	PSS13175-000035	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.66726	-122.56549
36	PSS13175-000036	Eastern Strait of Juan de Fuca	Clallam	Port Angeles	Port Angeles - Incorporated UGA	48.14204	-123.42576
37	PSS13175-000037	South Sound	Pierce	Steilacoom	Steilacoom - Incorporated UGA	47.16998	-122.61066
38	PSS13175-000038	Central Sound	Kitsap		Bremerton - Unincorporated UGA	47.60149	-122.66985
39	PSS13175-000039	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.63128	-122.38082
40	PSS13175-000040	Admiralty Inlet	Jefferson	Port Townsend	Port Townsend - Incorporated UGA	48.13084	-122.76251
41	PSS13175-000041	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.26899	-122.40166
42	PSS13175-000042	Central Sound	Kitsap	Bremerton	Bremerton - Incorporated UGA	47.57617	-122.62899
43	PSS13175-000043	Strait of Georgia	Skagit	Anacortes	Anacortes - Incorporated UGA	48.52109	-122.61104
44	PSS13175-000044	Whidbey Basin	Island	Langley	Langley - Incorporated UGA	48.03641	-122.39957
45	PSS13175-000045	Central Sound	King	Normandy Park	Normandy Park - Incorporated UGA	47.42844	-122.3508
46	PSS13175-000046	Central Sound	Kitsap		Kingston - Unincorporated UGA	47.78584	-122.49468
47	PSS13175-000047	Strait of Georgia	Whatcom		Birch Bay - Unincorporated UGA	48.89548	-122.78201
48	PSS13175-000048	Central Sound	Snohomish	Mukilteo	Mukilteo - Incorporated UGA	47.92779	-122.30929
49	PSS13175-000049	Central Sound	Pierce	Gig Harbor	Gig Harbor - Incorporated UGA	47.33837	-122.59049
50	PSS13175-000050	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.58137	-122.52673
51	PSS13175-000051	Central Sound	King	Shoreline	Shoreline - Incorporated UGA	47.73996	-122.37688
52	PSS13175-000052	Eastern Strait of Juan de Fuca	Clallam	Port Angeles	Port Angeles - Incorporated UGA	48.12584	-123.45576
53	PSS13175-000053	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.27687	-122.40846
54	PSS13175-000054	Central Sound	Kitsap		Silverdale - Unincorporated UGA	47.60765	-122.70792
55	PSS13175-000055	Strait of Georgia	Whatcom	Bellingham	Bellingham - Incorporated UGA	48.71193	-122.51908
56	PSS13175-000056	Whidbey Basin	Skagit		Swinomish - Unincorporated UGA	48.39735	-122.53921
57	PSS13175-000057	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.24649	-122.4313
58	PSS13175-000058	Central Sound	Kitsap	Bremerton	Bremerton - Incorporated UGA	47.58171	-122.63607
59	PSS13175-000059	Strait of Georgia	Skagit		Anacortes - Unincorporated UGA	48.49191	-122.5752

USE_ORDER	SITE_ID	REGION	COUNTY_NM	CITY_NM	UGA_NM2	LAT_DD	LON_DD
60	PSS13175-000060	Strait of Georgia	Skagit	Anacortes	Anacortes - Incorporated UGA	48.46759	-122.58601
61	PSS13175-000061	Central Sound	Pierce		Tacoma - Unincorporated UGA	47.31948	-122.42765
62	PSS13175-000062	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.70579	-122.51677
63	PSS13175-000063	Strait of Georgia	Whatcom	Blaine	Blaine - Incorporated UGA	48.99194	-122.76634
64	PSS13175-000064	Whidbey Basin	Snohomish	Everett	Everett - Incorporated UGA	48.00545	-122.23047
65	PSS13175-000065	South Sound	Thurston	Olympia	Olympia - Incorporated UGA	47.04624	-122.91204
66	PSS13175-000066	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.52018	-122.3952
67	PSS13175-000067	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.72014	-122.5472
68	PSS13175-000068	Eastern Strait of Juan de Fuca	Clallam		Clallam Bay - Unincorporated UGA	48.2537	-124.26875
69	PSS13175-000069	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.29627	-122.53121
70	PSS13175-000070	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.60279	-122.54731
71	PSS13175-000071	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.69051	-122.40332
72	PSS13175-000072	Admiralty Inlet	Jefferson	Port Townsend	Port Townsend - Incorporated UGA	48.10934	-122.76823
73	PSS13175-000073	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.24386	-122.40519
74	PSS13175-000074	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.63316	-122.5775
75	PSS13175-000075	Strait of Georgia	Whatcom	Bellingham	Bellingham - Incorporated UGA	48.74124	-122.49268
76	PSS13175-000076	Whidbey Basin	Island	Oak Harbor	Oak Harbor - Incorporated UGA	48.29186	-122.62405
77	PSS13175-000077	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.28453	-122.48499
78	PSS13175-000078	Central Sound	Kitsap	Bremerton	Bremerton - Incorporated UGA	47.5722	-122.6812
79	PSS13175-000079	Strait of Georgia	Skagit	Anacortes	Anacortes - Incorporated UGA	48.50897	-122.68452
80	PSS13175-000080	Central Sound	Snohomish	Edmonds	Edmonds - Incorporated UGA	47.84736	-122.33911
81	PSS13175-000081	South Sound	Thurston	Olympia	Olympia - Incorporated UGA	47.05885	-122.90304
82	PSS13175-000082	Central Sound	King	Burien	Burien - Incorporated UGA	47.48479	-122.36117
83	PSS13175-000083	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.6499	-122.51928
84	PSS13175-000084	Eastern Strait of Juan de Fuca	Clallam	Port Angeles	Port Angeles - Incorporated UGA	48.14039	-123.41527
85	PSS13175-000085	South Sound	Pierce	University Place	University Place - Incorporated UGA	47.21093	-122.57912
86	PSS13175-000086	Central Sound	Kitsap		Central Kitsap - Unincorporated UGA	47.64996	-122.62115
87	PSS13175-000087	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.63493	-122.49368
88	PSS13175-000088	Admiralty Inlet	Jefferson		Jefferson Co Unincorporated UGA	48.0262	-122.7495
89	PSS13175-000089	Central Sound	Pierce	Tacoma	Tacoma - Incorporated UGA	47.26864	-122.41851
90	PSS13175-000090	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.60749	-122.5754

USE_ORDER	SITE_ID	REGION	COUNTY_NM	CITY_NM	UGA_NM2	LAT_DD	LON_DD
91	PSS13175-000091	Strait of Georgia	Whatcom	Bellingham	Bellingham - Incorporated UGA	48.70809	-122.51623
92	PSS13175-000092	Whidbey Basin	Island	Oak Harbor	Oak Harbor - Incorporated UGA	48.26813	-122.62896
93	PSS13175-000093	Central Sound	Kitsap	Poulsbo	Poulsbo - Incorporated UGA	47.74002	-122.65058
94	PSS13175-000094	Central Sound	Kitsap		Gorst - Unincorporated UGA	47.53297	-122.68628
95	PSS13175-000095	San Juan Archipelago	San Juan		Eastsound - Unincorporated UGA	48.71375	-122.91795
96	PSS13175-000096	Whidbey Basin	Snohomish	Everett	Everett - Incorporated UGA	47.9872	-122.21815
97	PSS13175-000097	South Sound	Mason		Allyn - Unincorporated UGA	47.39273	-122.82395
98	PSS13175-000098	Central Sound	King	Seattle	Seattle - Incorporated UGA	47.58327	-122.37223
99	PSS13175-000099	Central Sound	Kitsap	Bainbridge Island	Bainbridge Island - Incorporated UGA	47.67379	-122.56095
100	PSS13175-000100	Eastern Strait of Juan de Fuca	Clallam	Port Angeles	Port Angeles - Incorporated UGA	48.14008	-123.44849

## Appendix C. Peirce County's Puget Sound Mussel Monitoring Program Details

### Background on Permit Defined Monitoring

Ecology issued NPDES municipal stormwater permits for Phase I and Phase II communities (Ecology, 2012a,b) effective August 2013 through July 2018. All permittees located in Puget Sound were given two options to comply with the permits' Special Condition S8.B for status and trends monitoring requirements.

Option 1: Pay a prescribed amount into a pooled fund to support RSMP Status and Trends monitoring. These permittees' role is limited to providing permit-defined amounts of funding for coordinated implementation of monitoring at sites throughout the Puget Sound region.

#### Or

Option 2: Conduct their own status and trends monitoring at specific, assigned sites inside their jurisdictional boundaries, following the same protocols as those used for the RSMP.

In fall 2013, Pierce County, and the City of Redmond officially selected the second option. This Quality Assurance Project Plan (QAPP) defines the permit-required small streams status and trends monitoring that will be conducted by Pierce County. The City of Redmond does not have marine shoreline and therefore does not have a nearshore monitoring requirement. This QAPP serves as the Ecology-approved "RSMP QAPP" referenced in the permits. This appendix defines Peirce County's unique monitoring program information such as staff, roles and responsibilities, and mussel monitoring sites. All other procedures and sampling protocols are defined in the QAPP will be followed by WDFW and Pierce County

### Pierce County Project Staff and Responsibilities

Pierce County will conduct mussel monitoring at eight suitable nearshore sites in their jurisdictions from October 2015 through February 2016; exact timeline may vary slightly as determined by WDFW mussel monitoring lead. Pierce County must submit this completed appendix to their Ecology permit manager by May 15, 2015 (Table 1) for approval prior to sampling. Pierce County's responsibilities for mussel monitoring are defined throughout this QAPP, and are briefly summarized below.

- Conduct site suitability, secure permissions and report to the RSMP Coordinator and permit manager on the sites to be monitored.
- Collect sorted bagged mussels from WDFW when notified by WDFW mussel monitoring lead or RSMP coordinator that they are ready
- Install mussel monitoring cages and deploy mussels when notified WDFW mussel monitoring lead or RSMP coordinator to begin monitoring
- Conduct field site measurements at the time of deployment
- Retrieve mussels and remove all monitoring equipment from field when notified WDFW mussel monitoring lead or RSMP coordinator to end monitoring

- Conduct field site measurements at the time of retrieval
- Send all field data to WDFW mussel monitoring lead according to the timeline described in this QAPP.
- Enter field data into Ecology's Environmental Information Management (EIM) database
- Submit a mussel monitoring summary as part of the permit required annual report.

Permittee project staff and Phase I Permittees	· · · ·	tation of Stormwater Permit Monitoring
Name/Contact	Role	Responsibility
Carla Vincent <u>cvince2@co.piercel.wa.us</u> (253)798-2467	NPDES Stormwater Monitoring Project Manager	Manage overall compliance activities; verify whether QAPP is followed and monitoring data are of known and acceptable quality; ensure adequate training of staff, complies with corrective action requirements; oversees data QA/QC and submission to EIM; oversees annual report preparation
Scott Groce, Water Quality Specialist 3 (253)798-2477	Field Lead	Manage and oversee monitoring activities and sampling decisions; coordinate with WDFW mussel monitoring lead for mussel collection and delivery, manage equipment maintenance; manage internal and external field teams, prepare reports, performs data QA/QC and submission to EIM
Corrie Lee, Water Quality Specialist 2 <u>clee@co.pierce.wa.us</u> (253)798-6822	Field Assistant	Assist in site selection and confirmation, collecting and processing field samples; deliver samples, perform equipment maintenance, assist with report preparation and data entry into EIM.
Berl Eldridge, Water Quality Specialist 2 <u>beldrid@co.pierce.wa.us</u> (253)798-2248	Field Assistant	Assist in site selection and confirmation, collecting and processing field samples; deliver samples, perform equipment maintenance, assist with report preparation and data entry into EIM.
Jeff Barney, Water Quality Specialist 2 barney@co.pierce.wa.us (253)798-3073	Field Assistant	Assist in site selection and confirmation, collecting and processing field samples; deliver samples, perform equipment maintenance, assist with report preparation and data entry into EIM.

#### Permittee project staff and responsibilities.

### Ecology Project Staff and Responsibilities

Ecology's RSMP Coordinator will either approve or comment on the permittees' completed QAPPs and transmit approval or comments to the permittee via the permittee's Ecology Regional Permit Manager by June 15, 2015. After the sampling is completed and the permittee has completed quality assurance (QA) and quality control (QC) review of the data and submitted it to EIM, Ecology staff will review and notify the permittees with data quality corrections and when the data is ready for final upload to EIM. The RSMP Coordinator will review monitoring reports. Ecology permit managers will review all submittals for compliance purposes. Ecology staff and their responsibilities are listed in table below.

Ecology Staff	Administration of	Administration of Stormwater Permits								
Name, Program, Location	Role	Responsibility								
Brandi Lubliner - WQP Lacey, WA	RSMP Coordinator	Ongoing implementation and administration of RSMP. Reviews and approves completed QAPPs and project deliverables from permittees' monitoring efforts.								
Chris Montague-Breakwell WQP-SWRO: Lacey, WA	Permit Manager	Ecology's contact for stormwater permittees including Pierce County. Reviews QAPP and monitoring reports for permit compliance.								
WQP staff, Lacey, WA	EIM Coordinator	Reviews and QAs data submitted by permittees and RSMP contractors.								

### Ecology project staff and responsibilities

SWRO: Southwest Regional Office EIM: Environmental Information Management database WQP: Water Quality Program

## Pierce County's List of Sites

This table shows 41 unincorporated Pierce County sites sorted from the complete RSMP nearshore site list available on Ecology's RSMP website. The complete list also contains more accompanying information; <u>http://www.ecy.wa.gov/programs/wq/stormwater/municipal/rsmp/status.html</u>.

USE_ORDER	SITE_ID	REGION	COUNTY_NM	UGA_NM2	LAT_DD	LON_DD
61	PSS13175-000061	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31948	-122.42765
113	PSS13175-000113	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.33586	-122.5785
161	PSS13175-000161	South Sound	Pierce	Gig Harbor - Unincorporated UGA	47.38545	-122.62723
177	PSS13175-000177	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.30708	-122.56682
185	PSS13175-000185	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.30314	-122.44444
249	PSS13175-000249	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31732	-122.42943
353	PSS13175-000353	South Sound	Pierce	Gig Harbor - Unincorporated UGA	47.37649	-122.62436
433	PSS13175-000433	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.30957	-122.56843
441	PSS13175-000441	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.30621	-122.44251
481	PSS13175-000481	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.33832	-122.58275
505	PSS13175-000505	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31375	-122.43195
625	PSS13175-000625	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.33027	-122.57511
689	PSS13175-000689	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.31741	-122.57391
697	PSS13175-000697	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.29803	-122.43569
737	PSS13175-000737	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.34197	-122.58376
817	PSS13175-000817	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.33341	-122.57648
865	PSS13175-000865	South Sound	Pierce	Gig Harbor - Unincorporated UGA	47.3866	-122.62639
881	PSS13175-000881	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.30118	-122.56156
953	PSS13175-000953	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.30738	-122.43515
1005	PSS13175-001005	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31935	-122.42073
1085	PSS13175-001085	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31927	-122.4296
1121	PSS13175-001121	South Sound	Pierce	Gig Harbor - Unincorporated UGA	47.37414	-122.62384
1137	PSS13175-001137	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.32903	-122.57445
1201	PSS13175-001201	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.30783	-122.56761
1209	PSS13175-001209	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.30644	-122.43886
1273	PSS13175-001273	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31553	-122.4311
1377	PSS13175-001377	South Sound	Pierce	Gig Harbor - Unincorporated UGA	47.38242	-122.62599
1393	PSS13175-001393	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.32796	-122.5735
1445	PSS13175-001445	South Sound	Pierce	Pierce Co Unincorporated UGA	47.19088	-122.57378

USE_ORDER	SITE_ID	REGION	COUNTY_NM	UGA_NM2	LAT_DD	LON_DD
1457	PSS13175-001457	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.31578	-122.57286
1465	PSS13175-001465	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.2991	-122.44004
1505	PSS13175-001505	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.33907	-122.58334
1649	PSS13175-001649	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.30544	-122.56587
1841	PSS13175-001841	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.33383	-122.57684
1889	PSS13175-001889	South Sound	Pierce	Gig Harbor - Unincorporated UGA	47.38711	-122.62666
1893	PSS13175-001893	South Sound	Pierce	Pierce Co Unincorporated UGA	47.18725	-122.57572
1905	PSS13175-001905	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.30208	-122.56241
1977	PSS13175-001977	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.30709	-122.43604
2017	PSS13175-002017	Central Sound	Pierce	Gig Harbor - Unincorporated UGA	47.34014	-122.58342
2029	PSS13175-002029	Central Sound	Pierce	Tacoma - Unincorporated UGA	47.31903	-122.42258

## Appendix D. 2015/16 RSMP Mussel Monitoring Datasheet

#### Washington State 2015/16 RSMP Mussel Monitoring Site Datasheet

#### Washington State 2015/16 RSMP Mussel Monitoring Site Datasheet

	DEPLOYMENT			Current shoreline use	e (check all that apply): Deat ra	mp/launch:  Boathouse/st	ed: Bridge: Breakwater:					
Site ID:	Site Name:			Dock/pier/wharf;	Floating home; Marina;	Nooring buoy; Outfall;						
Bag numbers:				Road; Shipyard	l or terminal; Utilities; Other	:						
Deployer name(s):				Dock/pier/wharf mate	rial: Creosote Other treate	d wood; 🗌 Concrete; 🗌 Ste	el; 🗌 Other:					
Recorder name:				Piling/dolphin materia	al: Creosote Other treated	wood; Concrete; Steel;	Other:					
Deployment date:		Time cage anchored:			p; Yes - Estimated Number:	Used for:						
Cage GPS location (decimal degrees)	Latitude:	Longitude:	Accuracy (± XX feet):	Туре:	oint of flow onto beach): Size: Condition:	(i.e. mouth dian						
GPS make/model or a (must be set to datum N/				Outfall (pipe, culvert, po Type:	bint of flow onto beach): Size: Condition:	(i.e. mouth dian	<u>neter);</u>					
Anchors - type/number	r used:				bint of flow onto beach): Size:	(i.e. mouth dian	neter);					
	WATER & WEATHER	CONDITIONS (at cage)		Туре:	Condition:							
	ave energy: 🗌 Flat; 📄 Calm; 📄 el: 📄 Exposed; 📄 Moderately exp	Wind chop; Swells;	Breaking waves	Other obvious sources	s of pollution (oil slicks, seeps, etc.)							
Time of most recent L	LOW tide:	Height of most recent LO	W tide (feet):									
Precipitation: 🗌 Non	e; 🗌 Steady rain; 🗌 Showers;	Snow; 🗌 Hail		Additional comments/	observations (it's a good idea to r	ote landmarks that will help v	ou find the cage later!):					
	HABITAT (within 20	0 foot radius of cage)										
Substrate type – selec	ct ONE category that describes the	majority (at least 50%) of su	bstrate around cage:									
Bedrock, hardpan	Cobble-gravel mix Sand-grav	el mix 🛛 Sand	ud mix 🗌 🛛 Mud, silt 🗌									
Aquatic vegetation co	overage - percent substrate around	cage covered by seagrasse	s and/or algae:	TAKE PHOTOS of the deployed cage and surrounding substrate, including any interesting observations!								
None (<1%)	1 - 20% 🗌 20 - 40% 🗌	40 - 60% 🗌 60	0 - 80% 🗌 80 - 100% 🗌	RETRIEVAL INFORMATION								
Type of aquatic veget	tation present: 🗌 None; 🗌 Eelgr	ass; 🗌 Kelps; 🔲 Fucus;	Ulva; Other	TAKE PHOTO of the mussel cage BEFORE removal, to document condition of cage.)								
Natural streams/rivers	present: No; Yes; Na	atural spring/freshwater seep	): 🗌 No; 🗌 Yes	Site ID:	Site Name							
Other habitat comment	s/observations:			Bag numbers:								
				Retriever name(s):								
				Recorder name:								
				Retrieval date:		Time cage removed	t:					
visi	ANTHROPOGENIC STRUC ible from cage up to 400 m (1300			Cage GPS location (decimal degrees)	Latitude:	Longitude:	Accuracy (± XX feet):					
Adjacent upland use (	check all that apply):			GPS make/model or a								
	Commercial; Industrial; Ma		Public access;	(must be set to datum N	VAD83) Durces of pollution (oil slicks, seep	e etc.)2						
Rural residentia	al; 🗌 Undeveloped; 🔲 Urban resi	dential; 🗌 Other		ANT NEW ODVIOUS SC	burces of poliution (oil slicks, see	s, etc.)?						
	Ires (i.e. armoring of shoreline):	-		Additional commenter	observations (including condition	of CAGE on retrieval major	changes in babitat or structures					
	d, riprap, etc.)% shorelin			around cage):			ananyos an navitat or surukulics					
	, large woody debris, etc.) -		(↑ creosote in armoring?)									
	structures (includes old pilings, docks	, half-sunken boats, metal piec	es, etc.)									
🗌 No; 🗌 Yes, ty	ype/makeup:											

# Appendix E. Chain of Custody Form

### Mussel Chain-of-Custody Form - Washington State RSMP Mussel Monitoring

Start of monitori	ng : receive muss	els from	WDFW and	deploy at I	nussel	monito	ring site							
Release of bagged mussels from WDFW	Mussel bag ID numbers: WDFW staff name (print and	sign):					DATE	TIME						
at aquaculture facility	Receiver name and effiliation	(print):												
Exchange of bagged	Mussel bag ID numbers:						DATE	TIME						
mussels - secondary mussel possession, if	Relinguisher name and affilia	tion (print):												
needed	Receiver name and effiliation	(print):												
	Mussel bag ID numbers:		1		1		1							
Deployment of prepared mussels at	Site name and ID:													
monitoring site	Deployer name and affiliation	(print):					DATE	TIME						
	SAVE this form	and finish fill	ing it out at the	end of the mo	nitoring pe	eriod_(see	below)	-						
End of monitoring					-									
Retrieval Instructions: Wee seal, and put on ice in cool Resources Building, 1111 V	er overnight. DO NOT FRE	EZE. On the m												
	Mussel bag ID numbers:		1		1		I							
Retrieval of mussels from monitoring site	Site name and ID:	Site name and ID:												
	Retriever name and affiliation	n (print):					DATE	TIME						

Exchange of mussels -	Mussel bag ID numbers:			DATE	TIME
for overnight possession or transportation, if	Relinquisher name and affi	lation (print):			
different from retriever	Receiver name and effiliation	n (print):			

Release of bagged mussels to WDFW processing lab	Mussel bag ID numbers:				DATE	TIME
	Releaser name and affiliation	n (print):				
processing into	WDFW staff (print and sign)	•				

## **Appendix F. Specimen Form**

SPECIMEN FORM

su	RVEY I	D:			STATI	ON ID:								COL	LEC	TIO	N DA	TE:										
Γ								9		2		H	sto	Ā				_	Ct	nemis					Fin	Clip		
				Sex		Length <sup>3</sup>	Weight	Age Structure <sup>4</sup>	Origin <sup>5</sup>	Blood Plasma	Bile (FACs)	Liver / Hepato	Gonad	Liver – CYP1A	Liver – DNA	Liver Individual	Liver Comp	Muscle Individual	Muscle Comp	Whole Body	Whole Body No Liver	Whole Body No Entrails	Ovary/Egg	Spawned Eggs	Adipose	Genetics		
	Effort ID	Species <sup>1</sup>	FishID	0-7 1-M 2-F	Maturity <sup>2</sup>	FT \$C mm	gm	N() O() I() S()	н/w	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N	Y/N				Y/N	Y/N	Y/N		Observations
1																												
2																		<u> </u>										
3																				_								
3 4 5																		-		_								
6																		-										
6 7																												
8																												
9																												
10																											de ID	
8 9 10 11																											Sample ID	
12																												
13 14 15 16																												
14																												
15																												
16																												
17 18																		<u> </u>										
18 19																	_	-										
19 20																	-	-		-								
20																												PSAMP 4/07

<sup>1</sup>E8 - English sole, 8F - starry flounder, 88 - staghorn soulpin, CR - sopper rookfish, QB - quiliback rookfish, BR - brown rookfish, PH - herring, LC - lingood X - solo salmon, T - shinosk salmon, DC - Dungeness srab, GC - graceful orab, MT-Mytilus trossulus

<sup>2</sup>R - ripe NR - not ripe

<sup>2</sup>Circle: F - fork length T - total length S - standard length C - crab carapace width

<sup>4</sup>Check: N - none taken O - otolith I - Interopercule S - scale

<sup>6</sup> For salmon origin: H - hatchery W - wild

RECORDER:\_\_\_\_\_

Page

of

## Appendix G. Tissue Resection Log

TISSUE RESECTION LOG

SU	RVEY ID		ST/	ATIONID:		SAM	PLE TYPE:	nole Fish Musc () ()		Gonad	Blle Hepato () ()
				Wet Weight							
	Species <sup>1</sup>	FishID	Tissue <sup>3</sup> (g)	Empty Jar (g)	Sample (g)	SampleID	Date Collected	Date Composited	Days to Resection	Obs	servations
1											
2											
3											
4											
5											
6											
7											
8											
9											
10											
11											
12											
13											
14											
15											
16											
17											
18											
19											
20											

<sup>1</sup>E8 - English sole, 8F - starry flounder, 88 - staghorn soulpin, CR - oopper rookfish, QB - quillback rookfish, BR - brown rookfish, PH - herring, LC – lingood X - ooho salmon, T - ohinook salmon, DC - Dungeness orab, GC - graceful orab, MT-Mytilus trossulus

POWP STI

<sup>2</sup>Tissue Wt: grams of tissue taken from an individual fish.

<sup>9</sup>Sample Wt: total grams of tissue in a sample.

Page of \_\_\_\_\_

## **Appendix H. Field Equipment List**

Field materials required for DEPLOYMENT for each mussel monitoring site.

- 1. Anti-predator cage:
  - a. 16x16x16 inch wire mesh cube with removable lid (Figures 5-7, 9 and 10)
    - i. Cage manufactured by McKay Crab and Shrimp Gear in Brinnon, WA
  - b. Anchoring devices:
    - i. 4 four-foot long, bent-tip rebar stakes (Figures 5 and 10)
    - ii. 1 thirty-inch long helical/ screw anchor + a short pipe (i.e. lever) to screw it in/out (Figures 5, 8 and 9)
    - iii. Cinder block(s) optional, may be necessary for high exposure sites (Figure 10)
    - iv. 2 five-foot and 2 three-foot long ties optional, to secure cage to cinder block(s) or fixed items on site (i.e. non-creosote pilings, metal pipes, etc.) (Figure 10)
  - c. Cable ties (i.e. Zip ties) includes double the amount needed, in case some ties break (Figures 7 and 9):
    - i. 8 four-inch long ties to secure RSMP-study plaque to cage (do in advance)
    - ii. 52 eight-inch long ties, ~75 pound tensile strength, UVB-resistant 8 to secure the ends of 4 mussel bags to the sides of the cage, 8 to hold lid closed, 8 to secure cage to rebar stakes, 2 to secure cage to helical anchor
  - d. Reflector band and/or reflector/colored flagging helps field crew to re-locate cage during retrieval (Figure 7, yellow band on right side of cage)
  - e. Wire cutters to remove/reposition cable ties
  - f. Mallet to pound in rebar stakes
- 2. Mussel Installation and Removal:
  - a. Four bags of 16 live, Ecology-approved\* mussels with bag ID numbers (Figure 6)
  - b. Cooler with ice in sealed plastic bags
  - c. Nitrile, powder-free laboratory gloves 2 pairs for each field crew member
- 3. On-site Measurement:
  - a. GPS device set to <u>North American Datum 83 (NAD83)</u>
- 4. Data Recording:
  - a. Deployment/Retrieval Datasheet (Appendix C) printed on water-proof paper
  - b. Clipboard with pencils
- 5. Other:
  - a. Flashlights and/or lanterns, with extra batteries and/or a lighter
  - b. Cellular telephone, fully charged
  - c. Appropriate attire for the weather
  - d. Heavy-duty or leather gloves to protect hands during anchor installation
  - e. Printed permission and/or permits required (if any)

- f. Key(s) or pass code to access site (if needed)
- g. Garbage bag for broken zip ties

\* Ecology approved mussels (*Mytilus sp.*) to be deployed at all of the RSMP study sites will come from a single aquaculture source, to ensure a uniform starting condition in the sample population.

Field materials required for RETRIEVAL at each mussel monitoring site:

- 1. Mussel Removal:
  - a. Lever to unscrew helical anchor (Figure 8)
  - b. Wire cutters to remove cable ties
  - c. 4 gallon-sized Ziploc bags, labeled with site name/date
  - d. Cooler with ice in sealed plastic bags
  - e. Nitrile, powder-free laboratory gloves 2 pairs for each field crew member
- 2. On-site Measurements:
  - a. GPS device set to North American Datum 83 (NAD83)
  - b. Data Recording:
  - c. Retrieval form (Appendix D) printed on water-proof paper
  - d. Chain-of-custody form (Appendix E) printed on water-proof paper
  - e. Clipboard with pencils
- 3. Other:
  - a. Flashlights and/or lanterns, with extra batteries and/or a lighter
  - b. Cellular telephone, fully charged
  - c. Appropriate attire for the weather
  - d. Heavy-duty or leather gloves to protect hands during anchor removal
  - e. Printed permission and/or permits required (if any)
  - f. Key(s) or pass code to access site (if needed)
  - g. Garbage bag for broken zip ties