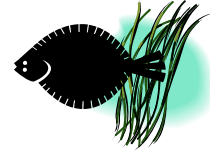


SHREFFLER ENVIRONMENTAL

*Research and Consulting in Fisheries Biology,
Nearshore Ecology, & Ecological Restoration*



December 18, 2008

Mr. David Freed
Project Coordinator
Clallam County Marine Resources Committee
223 E. 4th Street, Suite 15
Port Angeles, WA 98362

Re: 2008 MRC-MESA Pilot Field Project, Contract #332.08.034

Dear Mr. Freed:

This letter serves as the final report for the 2008 MRC-MESA Pilot Field Project, deliverable #5 under Contract #332.08.034.

Background

Because of the threat of oil pollution from large scale oil shipment through the Strait of Juan de Fuca, the U.S. EPA sponsored the Marine Ecosystems Analysis (MESA) Program in the 1970s. Under MESA, studies involving biological characterizations, physical oceanography, trajectory modeling, pollutant monitoring, and fate and effects of oil were implemented. These studies represent one of the most comprehensive baseline data sets ever gathered along the Strait.

Based on the outcomes from a 2007 MRC workshop with some of the original MESA researchers, Shreffler Environmental was contracted to develop a pilot field project to re-visit MESA sites sampled for intertidal and subtidal benthos in 1976 and 1977 (Nyblade 1978, 1979).

Study Purpose

Develop and initiate a pilot field project to: 1) collect benthic invertebrate samples with trained citizen volunteers using the same sites and same sampling protocols as Nyblade (1978); and 2) analyze the data to assess changes or long-term trends in the intertidal benthos community over the last 30 years.

Study Goals

1. Develop field sampling procedures, QA/QC procedures, and safety procedures for field data collection.
2. Purchase and/or borrow all necessary equipment for the field sampling.
3. Recruit and train a group of citizen volunteers that will do the field sampling.
4. Photograph the field sampling and laboratory sorting.
5. Assess changes or trends that have occurred between 1977 and 2008 in the abundance and biomass of the three dominant phyla of the intertidal benthos community (i.e., Annelida, Mollusca, Arthropoda)
6. Submit a pilot study progress report to MRC.

Field Methods

The intent of this pilot field project was to replicate, to the extent feasible, the same field sampling procedures that were used by Nyblade 1978 (*The Intertidal and Shallow Subtidal Benthos of the Strait of Juan De Fuca, Spring 1976 – Winter 1977*. NOAA Technical Memorandum ERL MESA 26).

The field methods used in this pilot project have previously been outlined in detail in the Final Field Sampling Plan (8-12-08), and are briefly summarized below.

Study Sites

The MRC selected two of Nyblade's 10 original sites from the 1970s to re-sample in August 2008:

- Dungeness Spit (exposed gravel)
- Jamestown (sandy mud).

Quadrat Locations

Samples were taken along a 100-m transect line stretched parallel to the water at +0 ft Mean Lower Low Water (MLLW) within a portion of the study site that was representative of the substrate type being sampled (i.e. gravel at Dungeness Spit, sandy mud at Jamestown Beach). The +0 ft MLLW elevation was determined using NOAA tidal predictions and groundtruthed by a surveyor. The MRC chose to sample at the +0 ft elevation, because this is the intertidal elevation that showed the greatest invertebrate abundance and biomass in the 1978-1979 sampling.

The sampling team recorded the GPS coordinates for temporary wooden stakes placed at the start and end of each transect line. The team also recorded compass coordinates and distances to fixed landmarks above the Ordinary High Water Mark (OHWM).

Quadrat locations were randomly determined and chosen without bias. Quadrats that were obviously not representative of the substrate type being sampled were rejected and another quadrat randomly chosen at the same location (e.g., gravel was not sampled at the sandy mud site or vice versa).

Methods of Sampling Quadrats



0.05m² quadrat at Dungeness

A 0.05 m² (22.5 cm x 22.5 cm) stainless steel quadrat frame was driven into the sediment 15 cm deep at each sampling location. The sediment from within the frame was removed using hand trowels and hands, then placed in a 3.5-gallon bucket and preserved in 10% buffered formalin. In the laboratory, the sediment samples from the 0.05 m² quadrat were dead-sieved through a 1.0 mm screen to retain small invertebrates (primarily Annelida and Arthropoda).



0.25m² quadrat at Jamestown

A 0.25 m² (50 cm x 50 cm) stainless steel quadrat frame was driven into the sediment 30 cm deep at each sampling location. The sediment from within the frame was removed using hand trowels and shovels and live-sieved in the field through a 12.5 mm screen to retain large invertebrates. All organisms retained on the screen were preserved in 500 ml jars filled with 10% buffered formalin. All samples were containerized and labeled separately for laboratory sorting and analysis.

The sample locations for both quadrat sizes were randomly chosen (using a random number table) and did not overlap. Samples from the two quadrat sizes were containerized separately and not combined.

Sample Number (replicates)

Just as in 1977-1978, five replicates for both quadrat sizes were collected at Dungeness Spit and three replicates for both quadrat sizes were collected at Jamestown Beach.

Preservation

Each sample jar received a unique field sample ID recorded on both the field data sheet and chain-of-custody form. All samples were fixed in the field in 10% buffered formalin (37% formaldehyde buffered with borax). After two weeks in buffered formalin, the samples were transferred to 70% ethanol for long-term preservation.

Supporting Measurements

The following supporting measurements were taken in addition to the field samples:

1. Temperature and salinity were measured approximately 0.25 m below the water surface. Temperature was recorded to the nearest 0.1 °C with a digital thermometer. Salinities were measured to the nearest 0.1 ‰ with a refractometer.
2. Prevailing weather conditions were recorded.
3. Each quadrat was photographed before sediment samples were taken.

Laboratory Methods



In laboratory M115 at Peninsula College, trained MRC volunteers and Peninsula College students under the supervision of Shreffler Environmental sorted each field sample into 3 jars—one jar for each major phylum (Annelida, Mollusca, and Arthropoda). The number of individuals (total abundance) and the wet weight (biomass) of each phylum was recorded. Total abundances and biomass of annelids, mollusks, and arthropods were compared between 1977 and 2008 for each of the two sites (i.e., Dungeness Spit, Jamestown Beach). All samples were archived at Peninsula College for potential further taxonomy work by a recognized expert to identify specimens to the species level.

Quality Assurance (QA) and Quality Control (QC) Procedures

To ensure that data was accurately and consistently collected and recorded, all volunteers followed standard procedures to minimize variation of methods among sites and among field samplers and lab sorters:

- Before leaving a field site, Shreffler Environmental confirmed that chain of custody forms were correctly completed for all preserved samples.
- Shreffler Environmental checked 100% of all entered field data for completeness and accuracy.
- Shreffler Environmental trained all volunteers how to identify benthic invertebrates to phylum (i.e., Annelida, Mollusca, and Arthropoda) in the laboratory.
- Shreffler Environmental ensured that sample sorting was conducted in accordance with required laboratory procedures.
- Shreffler Environmental checked a minimum of 10% of the sorted samples (and in many instances 100%) to verify the accuracy of each volunteer's identification of invertebrates to the correct phylum.

Results

Six samples (3 of each quadrat size) were successfully collected at Jamestown Beach on August 14, 2008. The following day, ten samples (5 of each quadrat size) were collected at Dungeness Spit. However, there were no invertebrates found in the Dungeness Spit sediment samples from the 0.025m² quadrat size that were sieved through a 12.5 mm screen in the field.

We compared benthic invertebrate wet weights (biomass) and numbers of individuals (total abundance) between Nyblade's data from summer 1977 and our data from summer 2008 for Jamestown Beach and Dungeness Spit (Table 1). Table 1 combines all samples from the 0.05 m² and 0.25 m² quadrats. Hence, data values are normalized to grams/m² and number of individuals/m².

No mollusks were found at either Jamestown Beach or Dungeness Spit in our 2008 samples. In 1997, Nyblade found 572 mollusks/m² at Jamestown Beach and none at Dungeness Spit.

The biomass of annelids at Jamestown Beach was higher in 2008 (190 g/m²) than 1977 (<36 g/m²), but the number of individuals was lower in 2008 (15,416/m²) than 1977 (18,148/m²). In contrast, the biomass of arthropods at Jamestown Beach was lower in 2008 (20/m²) than 1977 (<68/m²), but the number of individuals was higher in 2008 (9336/m²) than 1977 (2628/m²).

At Dungeness Spit, no annelids were found in 1977, but we found 740 annelids/m² in 2008 with a total wet weight of <1 gram. We also found a higher number of arthropods in 2008 (640/m²) than 1977 (140/m²), but the total wet weight was slightly lower in 2008 (<1 g/m²) than 1977 (<2 g/m²).

The field sampling at Jamestown Beach took eight people four hours to complete. The field sampling at Dungeness Spit took eight people five hours to complete. The lab training and sorting took a total of 184 person hours to complete, over three different days (18 people spent 6 hours on 11-12-08, 5 people spent 4 hours on 11-14-08, and 8 people spent 7 hours on 12-1-08).

Discussion

The six study goals were met with significant contributions by David Freed, 12 MRC volunteers, Dr. Jack Ganzhorn (Peninsula College (PC) Fisheries and Aquaculture Program Coordinator), and 14 PC students. From this perspective, the pilot study was a success.

Logistics & Level of Effort

As a pilot project, one of the important goals of this study was to figure out the logistics of the field sampling and laboratory sorting, and to answer the question of what could be accomplished using citizen volunteers rather than multiple researchers (as in the 1970s MESA studies).

What is clear from this pilot study is that both the lab and field efforts required significantly more volunteer hours and consultant hours than anticipated. In particular, the lab sorting was a time consuming effort, given that most of the volunteers had never sorted or identified marine invertebrates previously. In addition to the MRC volunteers and PC students that helped with the lab sorting, Jack Ganzhorn, David Freed, and Dave Shreffler each donated 15-20 hours assisting with the sorting.

One of the lessons of the field sampling was that it is physically demanding, because of the need to carry multiple heavy buckets loaded with sediment, as well as the two stainless steel sampling quadrats and miscellaneous other field gear. The level of effort required for both the field and lab has implications for future studies, as outlined in the recommendations section below.

Findings

One of the most interesting findings of this pilot project was the complete absence of mollusks at Jamestown Beach in comparison to the relatively high number of mollusks ($572/m^2$) found by Nyblade in 1977. It is unclear whether this may be due to: a) a change in substrate that resulted in a corresponding change of clam distribution; we noted significantly more gravel at this site than the sandy mud Nyblade reported in 1977; b) not enough samples; three samples of both quadrat sizes is probably the minimum necessary to characterize this habitat (note: we collected three, because this was the same number of samples collected by Nyblade in 1977); c) over-harvesting; while it is possible that all mollusks have been harvested from this site between 1977 and 2008, this seems unlikely; or d) a population collapse (mollusks completely died off) at this site between 1977 and 2008.

Without having the benefit of invertebrate identification to the species level for our 2008 samples, it is difficult to ascertain why the biomass of annelids at Jamestown Beach was higher in 2008 than 1977, but the number of individuals was lower in 2008 than 1977. One possible explanation is that there were fewer individuals in 2008, but the few individuals represented were larger and thus weighed more (e.g., a few large polychaete worms in 2008, as opposed to many small oligochaete worms in 1977). The explanation for differences between 1977 and 2008 in the biomass and abundances of arthropods at Jamestown Beach may be the exact opposite. In other words, the biomass of arthropods at Jamestown Beach could have been lower in 2008 than 1977 and the number of individuals higher in 2008 than 1977, because in 2008 there were many small individuals (e.g., gammarid amphipods) that did not weigh as much as larger individuals (e.g., mud shrimp) found in 1977.

It is unclear why we found no invertebrates in the Dungeness Spit sediment samples from the $0.025 m^2$ quadrat size, whereas Nyblade did find arthropods in samples from this quadrat size in 1977. For the $0.05 m^2$ quadrat samples, it is also confounding why we found 740 annelids/ m^2 in 2008 and Nyblade found none, and why we found higher numbers of arthropods in 2008 than Nyblade found in 1977. These differences could be, in part, because we sampled at a slightly higher elevation than Nyblade. Due to the high degree of swash (the rush of water up the beach slope following each break of the waves) on August 15, 2008, we were unable to sample at exactly +0.0 ft MLLW. The closest we could get without our quadrats getting inundated was approximately +0.5 ft MLLW.

In comparison to Dungeness Spit, Jamestown Beach showed far higher biomass and abundance of benthic invertebrates in both 1977 and 2008. The Dungeness Spit site is on a much more exposed, high energy beach, where many benthic invertebrate species have a tougher time surviving than in more protected sites like Jamestown Beach.

It is important to remember that our findings represent a “snapshot” of the benthic invertebrate fauna at Jamestown Beach and Dungeness Spit. We sampled on one day during one season (Summer) at one elevation (+0 ft MLLW) at two sites. In comparison, Nyblade sampled quarterly (Spring, Summer, Fall, Winter) at three elevations (+0 ft MLLW, +3 ft MLLW, and +6 ft MLLW) at 10 sites. Thus, we need to be careful about drawing “conclusions” from the 2008 pilot study.

Recommendations

While the pilot study was an overall success, we certainly learned some important lessons along the way that may benefit future studies.

- *The MRC should consider involving PC students in any future field sampling and lab sorting.* Dr. Jack Ganzhorn has expressed interest in collaborating if the MRC decides to expand the benthic invertebrate sampling along the Strait. Based on the total person hours to collect samples at two sites (72 hrs), and sort the samples in the lab to the phyla level (184 hrs), future sampling at +0 ft MLLW at all 10 of Nyblade’s original sites would take an estimated 360 person hours in the field and 920 person hours in the lab. Sampling at all 3 intertidal elevations sampled by Nyblade (+0, +3, and +6 ft MLLW) would require at least three times the level of effort as calculated above (1080 person hours in the field and 2760 person hours in the lab).
- *The MRC should consult with a statistician to determine if the numbers of replicates chosen by Nyblade in the original MESA studies was enough.* Three samples for each quadrat size at Jamestown Beach seemed inadequate. Would we have found mollusks at Jamestown Beach, if we had taken more samples?
- *The MRC could consider abandoning sampling with the 0.25 m² quadrats.* A lot of effort was expended in the field on sampling invertebrates within the larger 0.25m² quadrats, but no organisms were found in the five samples at Dungeness Spit and very few organisms were found in the 3 samples at Jamestown Beach (only a few larger polychaete worms). The 0.25 m² quadrat and live-sieving in the field through the 12.5 mm screen is best suited to capture mollusks, but we did not find any mollusks at either site. In the future, the MRC may want to consider sampling with the smaller 0.05 m² quadrat at all of the 0.25 m² quadrat sites. In other words, there would be twice as many 0.05 m² quadrat samples and no 0.25 m² quadrat samples. The tradeoff is this would decrease the field time, but increase the lab sorting time.
- *If the MRC were to pursue the above recommendation, they would likely want to have 1 or 2 additional 0.05 m² quadrats made.*
- *The MRC should consider recording wet weights to the nearest 0.01 gram rather than 0.1 gram, as specified by Nyblade (1978).*

- *The MRC needs to assess the tradeoffs of only identifying benthic invertebrates to the phylum level.* While sorting to the phylum level rather than the species level significantly reduces the amount of time involved in lab sorting, it correspondingly reduces the MRC's ability to draw inferences from the results. Many questions about changes in biomass and abundances of benthic invertebrates between 1977 and 2008 could be better answered with data at the species level than data at the phylum level.
- *To better understand the absence of mollusks at both Jamestown Beach and Dungeness Spit, the MRC might want to devise a study specifically designed to answer these questions:*
 - “Why were no mollusks found at these sites in 2008?”
 - Are mollusks missing from other sites where they were abundant in 1977?
 - Are there natural events (e.g., changes in substrate, increased water temperature, etc.) or human-caused events (e.g., pollution; oil spills, overharvesting, etc.) that might explain the apparent mollusk declines?

It is my understanding that the MRC would like me to make a presentation that summarizes the 2008 Pilot Field Project. I am happy to do so. Please call me or send an email to schedule the presentation.

Sincerely,

Dave Shreffler

Table 1. Benthic Invertebrate Comparison between Nyblade (1979) and MRC (2008) Samples

(all samples from 0.05m² and 0.25m² quadrats combined)

Jamestown Beach Study Area				
Data Source	TOTAL WET WEIGHT (Grams/Meter²)			
	Annelida	Mollusca	Arthropoda	All Phyla combined
Nyblade - Summer 1977	<36	<28	<68	<132
MRC - Summer 2008	190	0	20	210
Data Source	TOTAL NUMBER OF INDIVIDUALS/Meter²			
	Annelida	Mollusca	Arthropoda	All Phyla combined
Nyblade - Summer 1977	18148	572	2628	21348
MRC - Summer 2008	15416	0	9336	24752
Dungeness Spit Study Area				
Data Source	TOTAL WET WEIGHT (Grams/Meter²)			
	Annelida	Mollusca	Arthropoda	All Phyla combined
Nyblade - Summer 1977	0	0	<2	<2
MRC - Summer 2008	<1	0	<1	<2
Data Source	TOTAL NUMBER OF INDIVIDUALS/Meter²			
	Annelida	Mollusca	Arthropoda	All Phyla combined
Nyblade - Summer 1977	0	0	140	140
MRC - Summer 2008	740	0	640	1380