

Memorandum

Date: April 12, 2011

To: Pacific Shellfish Institute

From: Puget Sound Restoration Fund

Re: NMAI Economic Study- Liberty Bay Case Study, Subtask 8a

Introduction

On the US West coast intertidal molluscan restoration and enhancement efforts have focused on the native west coast (Olympia) oyster, *Ostrea lurida*. Once abundant in suitable habitat from Baja, Mexico to Sitka Alaska (Dall, 1914), native oysters with few exceptions were commercially extinct on the west coast by the 1930s. Though still found in portions of their historic range, extant populations are mostly limited to remnant aggregations where habitat characteristics remain favorable. Today, many locations in Puget Sound (Washington State) are habitat limited (otherwise suitable sites lacking emergent substrate or subject to excessive sediment loading) and generally restricted to places where existing populations can support regular recruitment. Successful oyster enhancement efforts are generally characterized by a dynamic equilibrium between the availability of hard substrate supporting regular recruitment events and predation pressure, competition for space, disease and sediment loading among other physical factors; all are factors that limit population expansion. Appropriate habitat for long-term population stability must also have the appropriate physical attributes with respect to intertidal exposure, temperature and salinity conditions to maintain the quality of oyster habitat.

A major impetus for rebuilding Olympia oyster populations derives from the potential ecosystem services associated with building larger and more robust aggregations of oysters. According to The Nature Conservancy, “restoring shellfish ecosystems is a critical step in saving our coastal bays and estuaries and the many forms of life that depend on them.” (TNC, *Shellfish Reefs At Risk: A Global Analysis of Problems and Solutions*). According to NOAA (*Habitat Connections*, 2008) “Olympia oyster beds have high biodiversity because they provide a physical habitat structure ideal for juvenile fish and crustaceans, worms, and foraging nekton and birds... Nutrient cycling is also an important role of native oysters. Feces and pseudo-feces deposited by oysters enter the benthic nutrient cycle, and are converted to various forms of nitrogen and carbon...These nutrient pathways represent a crucial linkage between benthic and pelagic realms within the estuarine ecosystem.” Further, the ecosystem benefit of

boosting populations locally goes well beyond the enhancement footprint itself. Having a pelagic larval form, Olympia oysters can contribute larvae on an inlet-scale and increase the population base overall.

Olympia oyster reintroduction efforts in Puget Sound have been underway since 1999. Recent strategies have targeted areas with limited settlement structure where there otherwise remains larval production emanating from nearby populations. Distributing a base layer of shell in these areas allows native oysters to re-occupy historic habitat while also preserving the genetic integrity of local populations. This has been the strategy of the Puget Sound Restoration Fund (PSRF) since 2005 and has resulted in a variety of restoration projects throughout Puget Sound.

Shellfish restoration and enhancement efforts are a relatively costly activity involving the large-scale addition of oyster shell to serve as a basement layer for pelagic larvae of Olympia oysters to colonize. By necessity, these activities often co-opt existing habitat, whether degraded or not, in order to alter habitat to make more productive for native oyster settlement and growth. As a consequence, the costs and benefits of enhancement and restoration efforts are especially important to document as they relate to potential short and long-term ecosystem benefits for both oysters and other organisms in terms of biodiversity, water quality and nutrient cycling. Especially important to document of course are specific population trends for the target species relative to the amount of effort expended in habitat creation. In addition, the placement of emergent substrate enables the colonization of and/or utilization of the created habitat by a great number of sessile and mobile invertebrates, fishes, birds, mammals, marine plants and other organisms. The potential for increased biodiversity including ecological services associated with native oyster enhancement and restoration activities, though quite evident have not been very well documented.

The Liberty Bay economic NMAI case study focuses on a discussion of the restoration process itself, the costs of enhancement using Pacific oyster shell as a basement layer, results of several years of enhancement activity in terms of native oyster abundance and associated diversity of other invertebrates and the anticipated ecosystem benefits associated with increased oyster abundance and biodiversity within native oyster beds.

A native oyster stock-rebuilding effort was initiated in 2005 on a 10-acre intertidal site in Liberty Bay (Scandia) near Poulsbo, WA. The project was conducted in accordance with a PSRF Memorandum of Agreement (MOA) with the Washington Department of Fish and Wildlife (WDFW) to “enhance native oyster habitat through December 31, 2012”. On this and other select beaches in Puget Sound, PSRF has experimented with habitat enhancement as a native oyster stock-rebuilding technique. The general approach involves placement of seasoned Pacific oyster shell (cultch), onto intertidal mudflats to provide a basement layer of material to help firm the bottom and provide emergent settlement substrate to oysters. Habitat enhancement at the Scandia location has allowed native oysters to re-occupy historic habitats while also minimizing impact to the

heterogeneity of local populations. The overall objective with these enhancements has been to catalyze repeated juvenile recruitment and to enhance benthic conditions to facilitate adult survival. The long-term goal is a functional, self-sustaining native oyster bed in the lower intertidal zone of this beach. The expectation is that along with the expansion of the native oyster bed there would be the potential for an increase in overall ecosystem services provided to Liberty Bay through increased biodiversity and provision of habitat for mobile invertebrates and fishes including salmonids.

Case Study Area - Habitat Enhancement in Scandia Using Pacific Oyster Shell

In 2004, Washington Department of Fish and Wildlife (WDFW), Puget Sound Restoration Fund (PSRF) and Baywater, Inc. personnel conducted a survey of intertidal sites on the Scandia side of Liberty Bay, near the village of Poulsbo, Washington that contained beds of remnant Pacific oyster shell that had persisted following the cessation of commercial oyster culture activities in 1992. Because of a legal settlement between the State of Washington and Coast Seafood, tidelands were forfeited to the State of Washington and placed under the management of the WDFW in 1990-91 (Jim Donaldson, personal communication). No activity associated with authorized shellfish culture or harvest has been associated with this site since that time. Of interest, however, were observations by McMillan (1931) who noted that native oysters thrived in Liberty Bay during periods in the early 20th century that included substantial degraded water quality. Scattered piles of Pacific oyster shell were observed in 2004 on the tidelands, which were occupied by a healthy population of native oysters that included multiple age classes (Figure 1). This discovery stimulated a series of substrate enhancement projects conducted by the PSRF over the ensuing four years (2005-2008). Substrate enhancements were based on the understanding that intertidal oyster populations in Liberty Bay were limited by the availability of hard substrates for settlement and subsequent recruitment of oysters.



Figure 1. Remnant Pacific oyster shell, native clamshell at Liberty Bay Scandia with native oysters of several size classes indicated by arrows.

The Liberty Bay (Scandia) site selected for shell enhancements is dominated by mud substrates intermixed with remnant piles of Pacific oyster shell cultch. While a few live Pacific oysters are associated with the remnant cultch material, both native oysters and a rich diversity of native and non-native invertebrates have colonized the Pacific oyster shell that remained. Molluscs associated with the remnant oyster shell include Olympia oysters, the (introduced) Japanese oyster drill (*Ocenebrellus inornatus*), the moon snail (*Polinices lewisii*) and several (introduced) suspension feeding gastropods (*Crepidula spp.*). Bivalves in addition to oysters include native horse clams (*Tresus nuttallii*) and native littleneck clams (*Protothaca staminea*) among others. The intertidal substrates dominated by Pacific oyster shell are composed largely of old oyster shell material. Significant numbers of shore crabs, red rock crabs (*Cancer productus*) and graceful crabs (*C. gracilis*) are found on or adjacent to shell substrates. Most importantly, however, is the presence of a significant population of Olympia oysters (including multiple size classes) residing on the remnant cultch material, indicating that this site in northeast Liberty Bay had been colonized by native oysters on more than one occasion.

In the years since commercial oyster culture ceased Liberty Bay shorelines have been significantly altered through extensive urbanization on the east and north sides of the bay. On the Scandia side of the bay, less intrusive land use changes have occurred though the shoreline adjacent to enhancement sites are extensively built out with homes, docks and bulkheads. In addition, Liberty Bay supports extensive plankton blooms, generally observed from late spring through late September, significantly affecting visibility and other water quality parameters (J. Davis, personal observations 2005-2010).

Of primary interest prior to making initial shell enhancements was the question of how best to utilize cultch materials for optimal recruitment of native oysters. In Puget Sound bays native oysters predominantly occupy low intertidal tide flats that are characterized by shallow pools of seawater, flowing seeps and shallow intertidal channels. Extremes of heat and cold air temperature tends to limit distribution of native oysters to settlement on hard substrate in relatively damp or permanently wet low intertidal or shallow subtidal areas (Hopkins 1935, 1937). Exceptions exist, however, as populations of native oysters also may occupy high intertidal lagoons. Indeed, native oysters also occupy shaded undersides of bulkhead rocks at considerably higher intertidal heights (up to +2'). Baker (1995) reports native oysters occupying considerably higher intertidal habitat (2m above MLLW), as well.

A pilot shell enhancement project was initiated in 2005 on 0.15 acre at an approximate tidal elevation of -0.5' to -1.5' below MLLW. This resulted in subsequent juvenile recruitment that year with oyster density initially estimated in excess of 90 oysters m². This initial success resulted in WDFW biologists recommending expansion of native oyster enhancement efforts in Liberty Bay. The specific location of the 2005-2008



Figure 2. View of 2007 two-acre enhancement plot shortly after shell was spread on intertidal tideflats.

project is situated in the tidal waters of Liberty Bay, Washington, northwest of Pearson Point in Kitsap County: Section 22, Township 26 North, Range 01 East WM (Figure 3). Of the 46 acres owned by Washington State, 10 acres were considered ideally suited for native oyster enhancement by virtue of tidal elevation and evidence of oyster recruitment. The site is composed

primarily of lower intertidal mudflats located between approximately -1.0 ft. and -3.5 ft. elevation (MLLW) and

consists almost entirely of soft substrates interspersed with piles of remnant shell consisting mostly of Pacific oyster and native littleneck clam shell. Three additional enhancement projects were completed between 2006 and 2008. Including the 2005 pilot project (0.15-acre area of mudflats was enhanced with 100 cubic yards of clean Pacific oyster shell) an additional 0.5-acre of intertidal mudflats was enhanced with the placement of 180 cubic yards of clean Pacific oyster shell in 2006. Additional enhancements in 2007 and 2008 resulted in 2 and 5-acre sites enhanced with 900 and 1,000 cubic yards of shell deposited, respectively (Figure 2). In each case clean oyster shell was barged on site at high tide and the shell sprayed into the water utilizing a fire hose over the designated tidelands during the late spring or summer months and generally prior to the predicted appearance of native oyster larvae in the water column. While shell plots varied in size by year and in the volume of shell deposited all of the plots are generally located on contiguous tidelands at the same approximate tidal elevation (Figure 3).

Following the placement of cultch material onto the tidelands monitoring efforts ensued over the last five years that varied in scope and intensity. These have included ongoing monitoring for intertidal seawater and intertidal bed temperature at various tidal elevations, annual oyster recruitment surveys and a variety of other studies that have attempted to address utilization of the Scandia tidelands by native fish including salmonids and other mobile invertebrates, monitoring for the presence of epibenthic organisms and monitoring for the presence of native oyster larvae in the water column.

NMAI funding for the Liberty Bay Case Study in 2009-10 enabled a comprehensive mapping and biological monitoring effort on selected enhancements in Scandia. Our intent was to investigate the costs associated with modifying primarily muddy substrates to increase the availability of emergent hard structure (through the addition of clean Pacific oyster shell) for native oyster settlement and subsequent recruitment. In addition to potentially boosting native oyster populations, we were interested in evaluating other ecosystem benefits (primarily species richness and biodiversity) that may be associated with the addition of cultch materials. The availability of enhanced

sites that differed in age afforded an additional assessment of whether oyster density, species richness and species diversity demonstrated trends based on time since deposition of emergent substrate materials. In this case we focused on plots that had been created in 2005 (4 years since deposition of shell), 2007 (2 years since deposition of shell) and 2008 (1 year since deposition of shell) and compared biological parameters to remnant plots that had been in place since at least 1993 (>15 years).

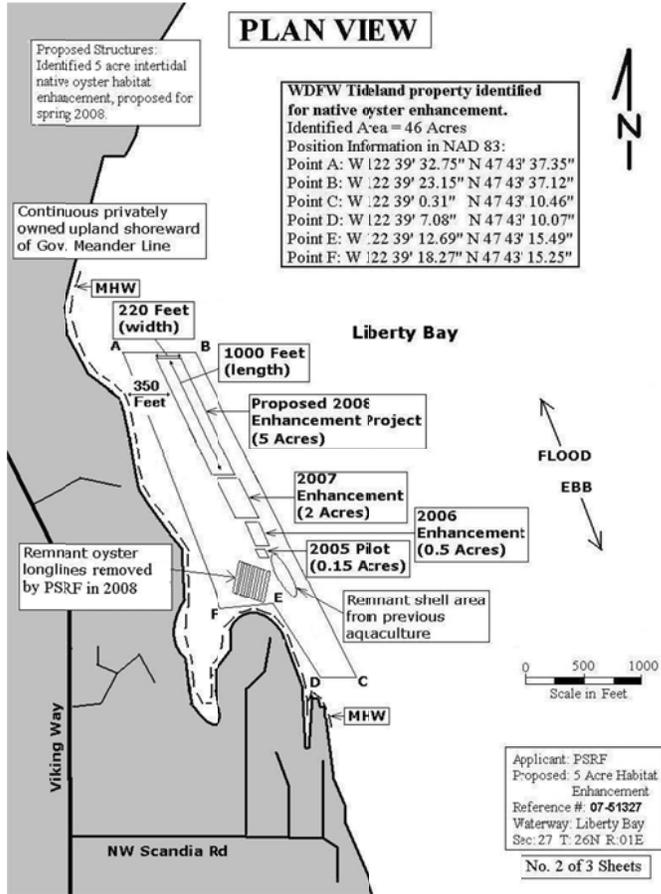


Figure 3. Location of enhanced shell plots in Liberty Bay near Poulsbo, WA.

Materials & Methods

Sampling for the presence and abundance of sessile and mobile invertebrates associated with intertidal habitats supplied with Pacific oyster shell focused on the 2005, 2007 and 2008 experimental shell plots and adjacent habitat associated with remnant Pacific oyster shell (the 2006 0.5 acre site was not selected for sampling due to the presence of massive barnacle sets that had occurred early in 2007). A primary goal of the work was to assess whether differences in abundance of native oysters and other sessile and mobile invertebrates in Liberty Bay was correlated with plots that differed in both overall area and age. The existence of plots established in 2005, 2007 and 2008

afforded this opportunity to assess abundance and diversity along a recent temporal gradient and compare biological parameters there with oyster abundance and size structure and biodiversity associated with older remnant oyster beds.

Sampling was conducted utilizing multiple ($1/16 \text{ M}^2$) quadrats placed both haphazardly and randomly on shell plots of different age during low tide events in summer 2009 (June -August). Prior to sampling, the perimeter of each shell plot was delineated using a hand-held GPS (Trimble Nomad 900GL Ultra-rugged Handheld) and mapping software (ArcPad). The perimeter of the plot was initially mapped by walking along the plot margin while inputting GPS position information. Following the establishment of the area to be surveyed random positions for quadrat placement were generated. Quadrats were placed on the substrate and sampled by removing all emergent substrate and any associated mobile organisms. Materials collected were placed into plastic bagging and transported by boat to the sorting area. Shell cultch and associated materials were initially rinsed through a 2mm sieve using a portable 12V. pump. All biogenic material was removed from the sampled cultch and identified to species. Native oysters and other bivalves were measured to the nearest mm using a digital caliper. In order to estimate the volume of the cultch material sampled for each quadrat location cultch materials (with all epibiont material removed) were placed into a mesh plastic bag and the bag placed into a 10L liter beaker of seawater previously positioned on an electronic scale tarred to zero. Taking care to avoid touching the bottom or sides of the beaker, the bagging holding cleaned and rinsed cultch was suspended into the beaker displacing seawater equal to the volume of the added cultch material. The apparent increase in recorded mass (subtracting the volume of the bagging) corresponded to the volume of shell materials contained in the $1/16 \text{ M}^2$ quadrat. Data was recorded by hand and later entered into a spreadsheet for subsequent statistical analysis.

The abundance of small crustaceans including harpacticoid copepods and including species known to be potential prey items for out-migrating juvenile salmonids was assessed in Liberty Bay on and adjacent to the 2007 2-acre shell plot in late April 2010. Sampling was accomplished using a specially modified device designed to suction biological materials off of the surface of bottom substrates. The epibenthic sampler consists of a perforated plastic PVC collar (20cm diameter) containing a 12V. bilge pump (Rule 1200). Perforations in the collar are covered with 100-micron screen. The sampler was placed onto the substrate surface and water forced through the perforations on the side of the collar when the pump is activated, effectively suctioning biological material off the surface of bottom substrates. Sample water exiting the sampler through plastic tubing is passed through a 100 micron screen and all materials retained fixed in 10% formalin in seawater and later transferred to 70% ETOH. Seven samples were taken for the presence of epibenthic organisms on and off emergent shell material, respectively on April 30, 2010. Samples were processed on site and later assessed for species present and numerical abundance including species commonly observed to constitute juvenile salmonid prey items using standard taxonomic methods for the PSRF in January 2011 by the Jeffrey Cordell Laboratory (University of Washington).

Results and Discussion

The GIS product of our survey, shown in Figure 4, illustrates the distribution of sampling locations across the range of habitat on the Scandia property; the majority of which are the result of enhancements 2005-2008. A total of 142 quadrat samples were taken on 2005, 2007 and 2008 enhanced plots plus another 68 samples taken on the remnant oyster shell piles. Our estimates from 2009 report the following for native oyster abundance in Table 1.

Table 1. Results of native oyster population assessments at Scandia near Poulsbo, WA

	Area (m ²)	Sample Number	Density Estimate(m ⁻²)	Coefficient of Variation %	Estimate Precision %	Population Estimate
Remnant Shell	13,741	68	50.35	21 %	42 %	691,884
2005 pilot	2,441	48	88.00	15 %	29 %	214,821
2006 Enhancement	1,450	19*	4.60*	27 %	54 %	6,716
2007 Enhancement	5,824	41	17.95	32 %	62 %	104,549
2008 Enhancement	14,108	53	1.21	60 %	118 %	17,036

*2006 Enhancement was surveyed in May 2008; sample unit was ¼ m².

One result of shell enhancement activity at Scandia was a 173% increase in space occupied by emergent settlement habitat. Natural recruitment of oyster juveniles has occurred in all enhancement plots; however, the magnitude of recruitment was highly variable from year to year. There is a positive correlation (R=0.66) between the residence time of the habitat and oyster abundance. Older habitat, in this case, has simply been exposed to greater numbers of potential recruitment events. As shown in Table 1, oyster abundance has increased about 50%, from 691,884 animals (estimated population abundance within the remnant shell) to a total of 1.03 million across all emergent shell habitat. It should be noted that no oysters were found outside of the oyster shell habitat, whether enhanced in 2005-2008 or residing on remnant shell piles. Reconnaissance efforts throughout greater Liberty Bay have found other oyster aggregations; these are mostly associated with remnant Pacific oyster shell habitat left by previous commercial oyster operations.

Table 2. Species data from 2009 emergent shell habitat samples.

	<u>Species</u>	<u>Remnant</u>	<u>2005</u>	<u>2007</u>	<u>2008</u>
1	<i>Alia gausapata</i>	178	264	150	53
2	<i>Amphiodia occidentalis</i>				3
3	<i>Crepidula dorsata</i>	192	823	751	201
4	<i>Crassostreagigas</i>		1	1	
5	<i>Cancer gracilis</i>				
6	<i>Crepidula nummaria</i>	158	665	387	654
7	<i>Clinocardium nuttallii</i>	1	1	8	15
8	<i>Euspira lewisii</i>		1		
9	<i>Hemigrapsusoregonensis</i>	7			1
10	<i>Haminaea sp.</i>				26
11	<i>Lophopanopeusbellus</i>	8	26	25	13
12	<i>Lottiaochracea</i>	4	1	1	
13	<i>Lirulariasuccincta</i>		1		
14	<i>Mopialialignosa</i>	9	5	1	2
15	<i>Metacarcinus magister</i>			1	
16	<i>Metridium senile</i>	1			
17	<i>Modiolusmodiolus</i>				1
18	<i>Macomanasuta</i>	1	4	31	51
19	<i>Mytilustrossulus</i>	50	5	10	66
20	<i>Nucellalamellosa</i>				1
21	<i>Nassariusmendicus</i>	48	233	125	59
22	<i>Oedignathusinermis</i>	3			
23	<i>Ocinebrellus inornatus</i>	10	1	1	7
24	<i>Ostrea lurida</i>	214	267	46	4
25	<i>Oenopota sp.</i>	1		1	1
26	<i>Pododesmusceprio</i>	4	28	23	
27	<i>Protothaca staminea</i>	3		2	19
28	<i>Pinnixatubicola</i>				6
29	<i>Pagurus sp.</i>		1	3	
30	<i>Searlesiadiria</i>				1
31	<i>Saxidomusgiganteus</i>			1	
32	<i>Tresus capax</i>			1	

Emergent habitat samples using quadrat-sampling methods were used to collect data on oyster density, associated macrofauna and shell volume within shell enhancements differing in area and age. Data on the range of species observed in both enhanced and remnant shell plots are summarized in Table 2.

A subsequent analysis focused on explaining whether shell volume (a measure of available substrate) and barnacle cover, two factors that are variable throughout the study area, predicts the abundance of native oyster and species abundance for a variety of other species associated with emergent habitat.

Sample data from the 2005 pilot enhancement indicate a potential interaction between shell volume and oyster abundance in samples. Older shell plots including both the



Figure 4. Scandia shoreline aerial photograph with discrete plot polygons and sampling locations from summer 2009 survey.

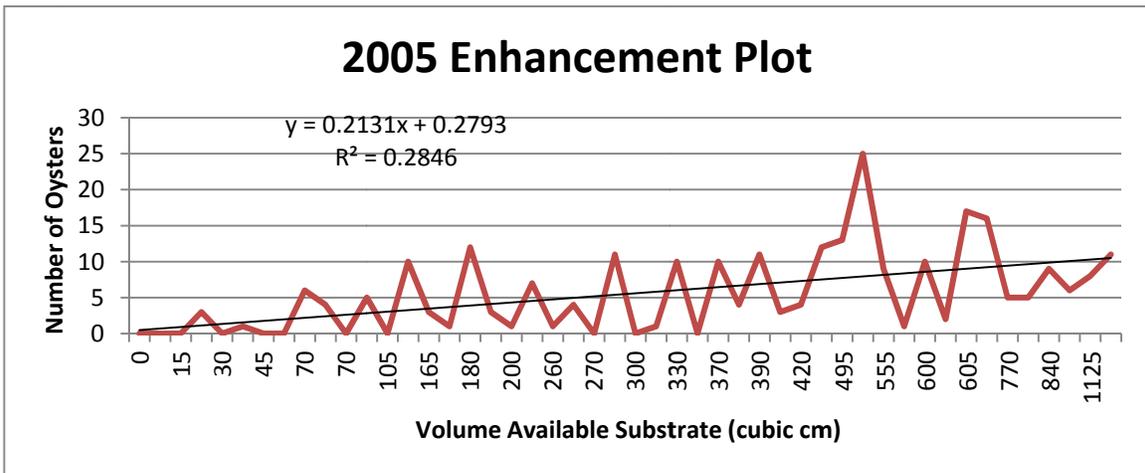


Figure 5. Oyster abundance vs. emergent shell volume from the 2005 pilot enhancement.

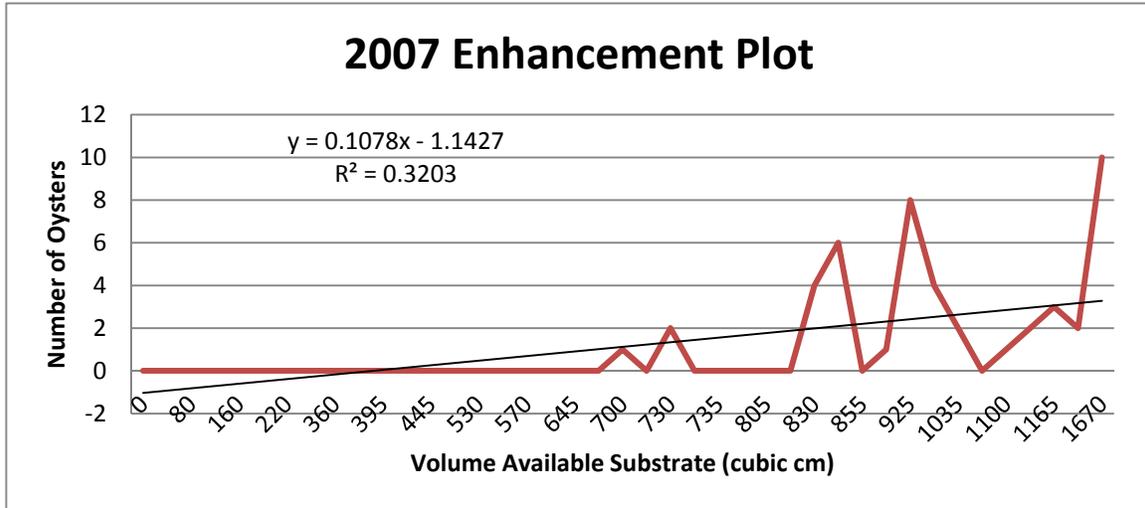


Figure 6. Oyster abundance vs. emergent shell volume from the 2007 enhancement.

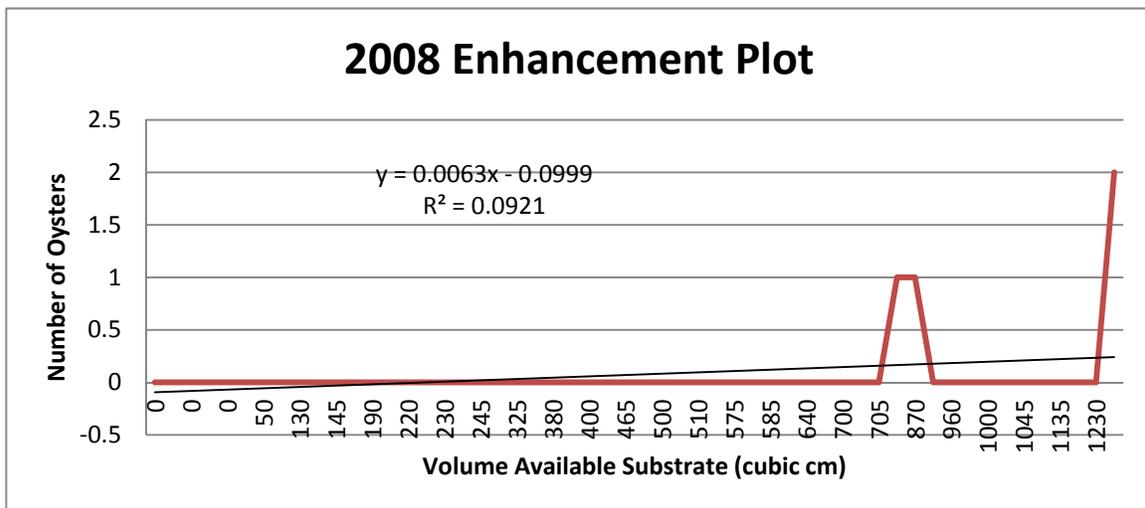


Figure 7. Oyster abundance vs. emergent shell volume from the 2008 enhancement.

remnant shell and 2005 enhanced plot demonstrate a weak statistical relationship between total available shell (estimated as cultch volume) and oyster abundance (Figure 5). For plots established in 2007 and 2008 no significant relationship between shell volume and oyster abundance was observed (Figures 6 and 7). From this data it appears that a weak but statistically significant association exists for increased oyster abundance to be associated with increased volume of available substrate for both the remnant oyster bed and adjacent 2005 plot, suggesting for this Liberty Bay site that plots exposed to a minimum of three (potential) recruitment events may be necessary before substrate availability becomes a potential factor limiting oyster recruitment.

Data on macro-fauna (other than native oysters) sampled from emergent habitat on the different enhancement sites show species richness (S') ranging from 17 to 20 species across all plots (Figure 9). Observed variability in species richness (S') was also relatively similar across all plots. Biodiversity values, calculated using the Shannon-Weiner biodiversity index (H'), estimated the “evenness” of numbers in the diversity of species present (Figure 9). In this case the oldest habitat (remnant shell) showed increased relative biodiversity compared to the enhancement plots created in 2005, 2007 and 2008, suggesting that age of plot may have an important effect on increasing greater “evenness” in numerical abundance among the variety of species present.

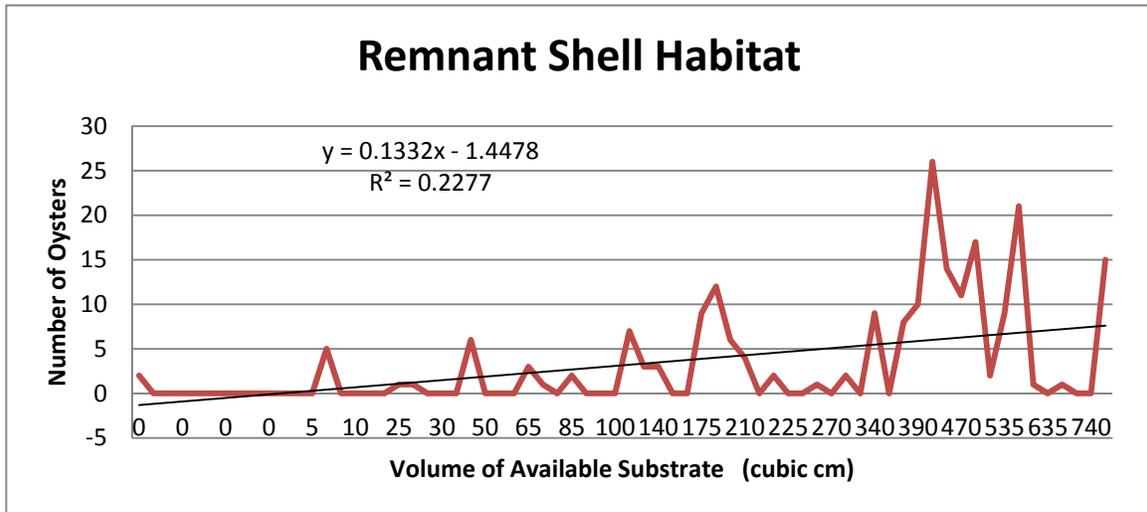


Figure 8. Oyster abundance vs. emergent shell volume from the 2007 enhancement.

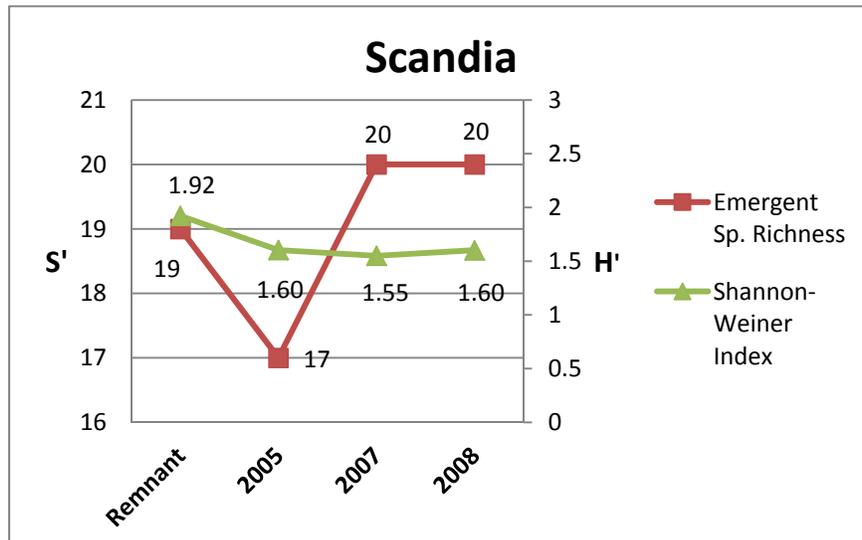


Figure 9. Emergent macrofauna species richness (S') and biodiversity (H') estimates for Scandia habitat plots.

For the specific case of barnacle cover sampled within the various plots Generalized Linear Models (GLMs) were employed to analyze the relationship between community species richness (S') and hypothesized effect factors of 1) Emergent shell volume, and 2) relative barnacle cover. GLMs were used to correlate shell volume to species using 3 models (Table 3). Data were first normalized for low sample size and both Gaussian and Poisson distributions utilized to fit the models. Examining the results for plot location (spacial/temporal gradient) across the models, AIC values were compared which use a “maximum likelihood” statistical approach. Model 1 (Shell Volume vs. S') demonstrated the best fit for the data with shell volume explaining the behavior of the S' data better than relative barnacle cover, or both (Models 2 & 3). Barnacle cover was consistently a poor covariate. The R^2 values per plot location shows that we were able to explain only 32% of the behavior of the data with our best fit in Model 1. While there was a trend for shell volume to correlate with S' , it was not an accurate predictor of species richness within the study area.

Analyses for the same parameters (shell volume and relative barnacle cover) relative to oyster abundance were also made. All possible GLM models for parameters and interaction combinations related to oyster abundance in samples were assessed. Models looking at all parameters (Plot * Volume * Barnacle + interactions) fit data for oyster abundance the best. Results suggest that models incorporating more parameters were better correlated with oyster abundance. None of the models tested reported any significant effect on oyster abundance. This was likely due to overall low observed density of oysters for all plots sampled. This was especially the case for 2007 and 2008 plots that had experienced only limited oyster recruitment (2 years and 1 year, respectively).

Results of epibenthic sampling indicated a highly significant effect (t-test; p value <.05) between the presence of epibenthic organisms (including species known to be salmonid prey) and emergent shell materials (Figure 10A and B). Salmonid prey species observed were mainly harpacticoid copepods within the genus *Tisbe* spp. At both locations, taxa richness was higher at the shell site as compared to the mud site (Figure 10A). Seven juvenile salmon prey taxa were found at Liberty Bay (Tables 4 and 5). At Liberty Bay, densities of total invertebrates, total harpacticoid copepods, and salmon prey taxa were significantly higher at the shell site as compared to the mud site (t-test, alpha = 0.05—Table 4). Table 5 lists all species observed in epibenthic samples from Liberty Bay. The presence of habitat structure in the form of shell appears to have resulted in increased abundance of total epibenthic organisms including many harpacticoid copepods (including salmon prey organisms).

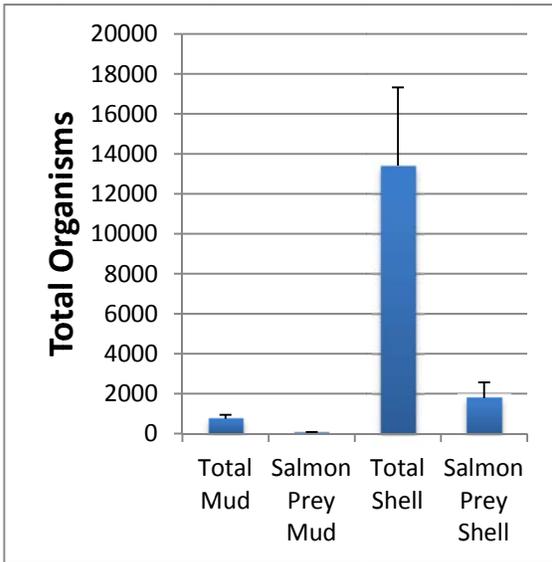


Figure 10A

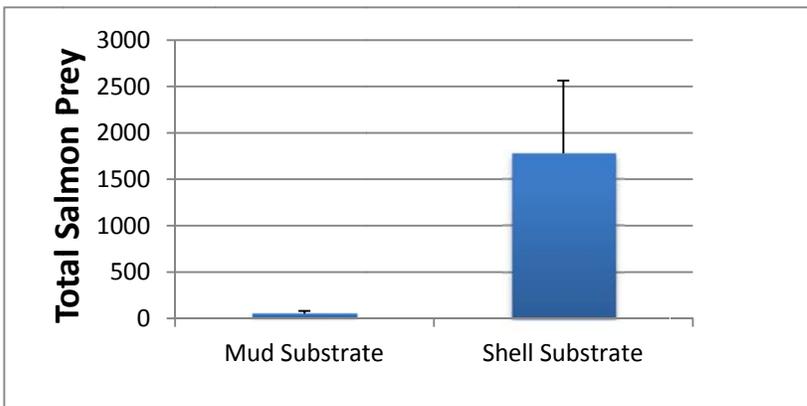


Figure 10B

Figure 10A. Epibenthic organism abundance on and off emergent Pacific oyster shell material on the Liberty Bay 2007 shell enhancement sampled in late April, 2010. Figure 10B. Total abundance of salmonid prey (mainly *Tisbe* spp.) sampled on and off emergent shell material, respectively.

Plot	<u>Emergent Volume vs. Sample Richness (model 1)</u>				<u>Sample Richness vs. Emergent Volume AND Barnacle Cover (model 2)</u>				<u>Sample Richness vs. Barnacle (model 3)</u>			
	R ² (adjusted)	Degrees of Freedom	AIC*	ΔAIC	R ² (adjusted)	Degrees of Freedom	AIC*	ΔAIC	R ² (adjusted)	Degrees of Freedom	AIC*	ΔAIC
All	0.33	209.00	872.72	0.00	0.12	207.00	874.93	2.21	0.12	208.00	930.69	57.97
Remnant	0.17	66.00	280.81	0.00	0.15	64.00	283.88	3.07	0.03	65.00	291.88	11.07
2005	0.12	46.00	205.32	0.00	0.10	44.00	207.85	2.52	0.10	45.00	207.04	1.72
2007	0.34	39.00	166.46	0.00	0.31	37.00	170.15	3.68	0.01	38.00	184.26	17.80
2008	0.34	52.00	221.71	0.00	0.34	50.00	223.95	2.24	0.04	51.00	243.00	21.29

* Note - AIC values should be compared based on the same color. You cannot compare the AIC values across plots because they use different data. The lowest AIC value represents the best model fit. R² values were adjusted for low sample size.

Table 3. GLM regression model results for shell volume (Model 1), relative barnacle cover (Model 3), and combined (Model 2) as correlated with species richness (S').

	Taxa Richness	Total Density	Harpacticoid Density	Salmon Prey Density	T-test Alpha Levels
Liberty Bay Shell	32	13407.1	6785.7	1807.1	Total: 0.018 Harpacticoids: 0.013
Liberty Bay Mud	23	742.89	310.1	57.1	Salmon Prey: 0.052

Table 4. Taxness richness, densities (numbers m-2), and t-test results from Liberty Bay epibenthic sampling.

Table 4. Epibenthic invertebrate taxa identified from Liberty Bay, 27 May 2010.

Phylum	Subphylum	Class	Subclass	Order	Taxon	Salmonid prey/ non prey
Arthropoda	Crustacea	Malacostraca	Peracarida	Amphipoda	<i>Monocorophium</i> sp.	prey
Arthropoda	Crustacea	Malacostraca	Peracarida	Isopoda	Epicaridea	
Arthropoda	Crustacea	Malacostraca	Peracarida	Cumacea	<i>Cumellavulgaris</i>	prey
Arthropoda	Crustacea	Malacostraca	Peracarida	Cumacea	<i>Nippoleuconhinumensis</i>	non prey
Arthropoda	Crustacea	Malacostraca	Peracarida	Tanaidacea	<i>Leptocheliasavignyi</i>	prey
Arthropoda	Crustacea	Malacostraca	Eucarida	Decapoda	Brachyura	non prey
Arthropoda	Crustacea	Malacostraca	Eucarida	Decapoda	<i>Neotrypaea</i> larvae	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Calanoida	Calanoida	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Cyclopoida	Cyclopinidae	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Cyclopoida	Cyclopoida	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Ameira</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Amonardianormani</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Amphiascoides</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Amphiascoides</i> sp. A	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Amphiascopsiscinctus</i>	non prey

Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Amphiascusundosus</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Amphiascus</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Cletodes</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Dactylopusiacrassipes</i>	prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Dactylopusiavulgaris</i>	prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Diarthrodes</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	Ectinosomatidae	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Enhydrosoma</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	Harpacticoidacopepodids	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Harpacticus-obscurus</i> group	prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	Laophontidacopepodids	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Longipedia</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Mesochra</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Normanella</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Orthopsylluslinearis</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Paradactylopodia</i> sp.	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Paralaophontecongenera</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Paralaophonteperplexa</i> group	
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Parastenheliahornelli</i>	non prey

Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Robertsonia cf. knoxi</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Stenheliapeniculata</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Tachidiustriangularis</i>	non prey
Arthropoda	Crustacea	Maxillopoda	Copepoda	Harpacticoida	<i>Tisbe</i> spp.	prey
Arthropoda	Crustacea	Maxillopoda	Ostracoda	Podocopida		
Annelida		Polychaeta			Polychaeta larvae/juveniles	non prey

Conclusions:

Increasing the quantity of emergent shell material in Liberty Bay has had the overall effect of increasing native oyster density. The effects of adding shell appears related to the length of time shell material has been deposited though no strong statistical relationships were established for this trend. Weak, statistically significant relationships were seen for both the remnant shell and 2005 plots demonstrating a relationship between mean oyster abundance and the volume of substrate material sampled. Both the 2007 (2 acre) and 2008 (5 acre) did not demonstrate any relationship between shell volume and oyster density; in the case of the 2008 plot there was little evidence of oyster settlement overall over the intervening 12 months. Previous research has demonstrated that annual recruitment of native oysters on this site is variable and that the utilization of emergent habitat at this location likely depends primarily upon both the supply of native oyster larvae and subsequent survival of new recruits. Previous work has also demonstrated that oyster larvae are not particularly abundant in the water column in this part of Liberty bay; the last major oyster settlement event occurring in 2007 at this site. Importantly, however, it also appears that the presence of emergent substrate remains effective as oyster habitat for many years; remnant Pacific oyster shell has been onsite here for at least 17 years following the cessation of commercial oyster culture activities there in 1992. This observation is in stark contrast to oyster restoration activities in Chesapeake Bay where shell loss is extensive and continuous without the presence of live oysters contributing to reef development (Mann, 2011).

The cost of enhancing mudflats with hard substrate is considerable. For the years 2005-2008 four enhancement plots were established in Liberty Bay Scandia totaling approximately 23,823 square meters or 5.88 acres. PSRF estimates the cost of establishing enhancement plots at \$50,000 per acre when Pacific oyster shell is deposited on mudflats or other substrates without significant amounts of emergent substrate. This represents an investment of \$294,000 in oyster restoration for the WDFW site in Scandia (PSRF 2010). These estimates assume that shell is applied at a similar density over the full acreage "enhanced." In fact, this was not the case for this project as earlier enhancements received larger volumes of shell material (the major cost associated with the enhancement) relative to the area treated. Important to consider, however, is the question of what volume of shell (cubic meters per acre) is actually necessary to provide adequate habitat for oyster recruitment and the establishment of viable oyster populations. For sites with high potential native oyster recruitment greater amounts of shell material may be warranted.

These monetary investments helped restore ecosystem services in the form of native oyster abundance and filtration, increased biodiversity and species richness and increased salmonid prey species. The 2,180 cubic yards of shell distributed on tideflats over the course of the project enhanced 5.88 acres of native oyster habitat, increased the native oyster population by 50% (from 691,884 oysters to 1,030,000 oysters) and provided a basement layer of shell that will be available for recruitment in future years. Results of epibenthic sampling indicated a highly significant effect (t-test; p value <.05) between the presence of epibenthic organisms (including seven species known to be salmonid prey) and emergent shell materials (Figure 10A and B). According to Jeff Cordell at University of Washington's Fisheries Research Institute, the

availability of food resources is the most significant factor affecting salmon survival. “It has been demonstrated that faster growth as juveniles = better survival.” Enhancing native oyster habitat in the lower intertidal zone with Pacific oyster shell resulted in a 173% increase in space occupied by emergent settlement habitat and appears to have resulted in increased abundance of total epibenthic organisms, including many harpacticoid copepods known to be salmon prey organisms.

A factor not specifically addressed in this study may also bear on the observed results. Japanese oyster drills (*Ocenebrellus inornatus*) are known to be relatively abundant at this site and are effective predators on a variety of sessile invertebrates, including native oysters. Drill density was observed to be highest on the remnant oyster plot compared to the enhancement plots though snails have been observed on other plots as well in high abundance in other years (Table 2). Predator density may be a significant factor contributing to the control of overall abundance of oysters and other invertebrates, including barnacles regardless of the overall availability of emergent substrate. The relationships are highly variable from site to site however; in some cases increasing density of native oysters reduced the per capita impact of drills, a classical type II functional response (Buhle and Ruesink, 2009). The effects of predator density on prey (native oysters) need to be better assessed in future studies as drill abundance is a potentially important determinant of native oyster abundance overall.

Emergent shell material offered a site for settlement for a large variety of marine invertebrates requiring hard substrate for at least a portion of their life cycle. Within shell plots, overall species diversity and richness trended higher in plots containing increasing volumes of emergent substrate but the overall relationships were weak. Overall, shell volume was not determined to be an accurate predictor of species richness within the study area. The possibility that the emergent substrate had not been on site for a long enough period of time cannot be discounted and it is likely that the effects of adding emergent substrate take a number of years for organisms to saturate the available habitat. It may be the case that species diversity is not particularly influenced by the presence of open substrate. As was the case for a plot established in Liberty Bay in 2006 adjacent to the three study plots described here, a massive barnacle (*Balanus glandula*) population was established on this plot, utilizing virtually 100% of the available shell cover and appearing to limit the settlement of other species (including native oysters) to a significant degree.

Abundance and diversity of epibenthic organisms in Liberty Bay assessed on the 2007 shell plot and compared to adjacent mud substrates showed a strong and significant relationship with overall abundance and species diversity (including species known to be juvenile salmonid prey (e.g. *Tisbe* spp.)) significantly greater on habitat characterized by emergent substrate. The relationships between fish utilization and native oyster enhancements using Pacific oyster shell were not specifically investigated, however the presence of juvenile salmonid prey on native oyster shell plots point to the potential utilization of these habitats by salmonids - a putatively important ecosystem service afforded by restoration projects of this kind.

The valuation of ecosystem services are relatively simple to evaluate when food, fuels and fiber are considered. These are services that have a long history of valuations gained through

traditional markets. Non-market benefits abound however and until recently there was little effort to value these within traditional market driven economies. This is due mainly to the difficulty in directly deriving appropriate market valuations. With respect to near-shore marine systems, this has especially been the case as benefits associated with increased numbers of an historically important native oyster, increased overall diversity, provision of habitat and other attributes having perceived social benefits remain difficult to define in economic terms.

Nonetheless, the importance and potential benefit derived from non-traditional ecosystem services make conceptualization of these services critical to develop as pressures on near-shore environments increase. The ability of marine bivalves to both clarify the water column through filtration activities and sequester nitrogen and phosphorus are important to document on a watershed basis for populations of marine bivalves. Unlike shellfish aquaculture where shellfish are removed from the watershed at harvest the removal of nutrients at harvest is not a prime consideration for restoration activities. As native oyster beds increase in size and ecological complexity, however, a suite of other benefits will likely emerge. Foremost is the potential through benthic pelagic coupling for native oysters to help facilitate nitrification and denitrification processes. Recent work suggests that complex habitats associated with oyster beds may significantly enhance ecosystem services related to nitrogen sequestration in estuaries subject to high nutrient loading (Cornwall et al., 2011). These processes have not been demonstrated in native oyster beds and remain a prime focus with renewed interest to better define and characterize.

Complex habitat associated with the creation of emergent substrate for settlement of native oysters has demonstrated that oyster abundance can be increased along with significant increases in abundance and species diversity of associated invertebrates. Particularly noteworthy was the finding that epibenthic organisms (mainly harpacticoids) increased in both abundance and species diversity on emergent shell material. This factor alone has the potential to justify greater attention to rebuilding near shore habitats where hard substrates have either been removed or altered.

Tradeoffs between short-term economic gain and conservation of ecosystems for the services they provide are critical to define using risk assessment techniques (Abson and Termansen 2011). An emphasis on better evaluating risk associated with loss of critical ecosystem services is needed though it may be useful to define loss of ecosystem function in terms of readily recognized economic and cultural values. There are many examples from the literature describing the effects of environmental degradation on the public's perception and use of the natural environment. Using an example from marine systems, we can relatively easily define economic losses to individuals and communities due to degradation of surface waters associated with excess nutrient loading of surface waters, subsequent eutrophication and build-up of excess and noxious aquatic vegetation. Direct, economic losses include lower property valuations resulting in decreased property tax income to municipalities, reduced income to parks and public beaches and decreased income from tourism generally as the public curtails use of the near-shore environment. These social and culturally important losses are relatively easy to quantify compared to placing a defined economic value on the public benefit of growing shellfish for the myriad of ecosystem services they provide. Placing a value on more easily

defined economic loss (decreased utilization of the beach and water for recreation) may have the effect of assisting the public to recognize and therefore value the growing of shellfish simply for the social benefit associated with nutrient removal, clarifying water, etc.

For the case of oyster restoration efforts in Liberty Bay, better defining potential salmonid use for out-migrating smolts from Dogfish Creek may be critically important. Higher survivorship of salmonids or other fishes due to greater abundance of prey items associated with restored native oyster beds may translate into enhanced sport-fishing opportunities for fishers - a clear economic benefit with wide public appeal. Framing the debate in economic terms that focus on traditionally valued ecosystem services (more oysters for the public to harvest and more fish to catch) may help to better define ecosystem valuations for less traditional ecosystem services that are nonetheless fundamentally important to the structure and ecological function of near-shore marine habitats.

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