INDIAN ISLAND FORAGE FISH MONITORING PROJECT
CLOSE-OUT REPORT FOR FISCAL YEAR 2011
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Submitted by
Randy Hatch and Shannon Miller
Point No Point Treaty Council, Fisheries Services Program
360-297-3422
7999 N.E. Salish Lane, Kingston, WA 98346

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ABSTRACT

This report summarizes sampling conducted at Naval Magazine Indian Island from October 2011 to April 2014 aimed at developing a standardized sampling protocol to establish annual indices of surf smelt (*Hypomesus pretiosus*) and Pacific sand lance (*Ammodytes hexapterus*) spawn densities. A substrate core sampler was used to retrieve random samples from the upper intertidal zone of known spawning sites during semi-lunar tides. Incubating embryos (EMB) were then separated from substrates using an elutriation process. Catch rates for smelt EMB during Study Year 3 (mean CPUE, 723 ± 187 EMB/ft²) were similar to those in Study Year 2 but were approximately 10% higher than those of Study Year 1. Sand lance catch rates during Study Year 3 (mean CPUE, 735 ± 196 EMB/ft²) were approximately 40% lower compared to Study Year 2 but approximately 75% higher than in Study Year 1. Relative precision (RP) of the 95% confidence interval for total annual density averages of both species improved during Study Year 3 (smelt RP 25.9%, sand lance RP 26.7%) compared to Study Year 2 (smelt RP 26.5%, sand lance RP 34.7%) and Study Year 1 (smelt RP 30.5%, sand lance RP 35.5%). A longer time series of data will help clarify whether or not changes in annual egg deposition of the two species are a reflection of overall population abundance or if other factors are involved.
INTRODUCTION

One issue of great concern to Tribes related to Puget Sound ecosystem restoration is the development of appropriate indicators and benchmarks of ecosystem health and tying them to recovery goals established by resource co-managers. The Puget Sound Partnership (PSP) Action Agenda has identified a robust food web as an integral component in sustaining healthy populations of native species, and has identified forage fish as an essential component of that robust food web. While the PSP has focused on Pacific herring (Clupea pallasii pallasii) as a key sentinel for forage fish species and has established a herring recovery goal as one of sixteen Ecosystem Recovery Targets for Puget Sound, surf smelt (Hypomesus pretiosus) and Pacific sand lance (Ammodytes hexapterus) also provide significant contributions to the forage fish populations in Puget Sound.

The Point No Point Treaty Council and its member Tribes are particularly concerned about the role of forage fish in the food web because of the importance of these fish as prey to salmonid species that are a key part of Northwest tribal cultural heritage and are integral to tribal treaty fishing rights. The Action Agenda has identified the protection and recovery of salmon as a critical strategy in the restoration of upland and terrestrial ecosystems (PSP 2012)

Additionally, the Action Agenda has focused on near shore marine habitats and estuaries as critical indicators of the health of Puget Sound, and has prioritized the use of complete, accurate and recent information with regard to these habitats in shoreline planning and decision making. This project contributes to that strategy by providing annual quantitative, site-specific estimates of forage fish spawn which can be used by local planning agencies to direct future development in the shoreline planning process.
Little research has been applied to providing quantitative estimates of surf smelt spawn deposition. Most previous investigations of surf smelt spawning in Puget Sound have been qualitative, presence/absence studies (Penttila 2000, Long et al 2005). The Washington Department of Fish and Wildlife (WDFW) did attempt to provide quantitative estimates of the total annual surf smelt spawn deposition on one central Puget Sound beach by collecting core samples of spawning substrates (Wildermuth 1993), but the results were not used to establish long-term indices of spawning success. Concerted forage fish survey efforts by WDFW were discontinued in 1997 due to lost funding; current efforts by the state to assess forage fish stock status, outside of Pacific herring, is limited to analysis of historical forage fish data and inclusion of these data in existing habitat models (Pierce et al. 2009).

The method of separating forage fish eggs from substrate fines used in most presence/absence studies incorporated the mechanical separation of eggs and fines by applying agitation techniques to the egg/substrate mixture in a water solution (i.e. winnowing; Moulton and Penttila 2001). While this method is successful at separating most of the eggs from substrate fines, it is time consuming, and the level of effort applied to the winnowing process is difficult to standardize. A standardized technique that produces accurate egg counts during this stage could speed up the separation process and allow quantitative estimates of the total recovery of eggs.

Penttila (2007), summarized gaps in the knowledge of surf smelt geographical distribution and life history within Puget Sound. These shortcomings include incomplete identification of existing surf smelt spawning sites, as well as the lack of a cost effective method of stock assessment. This project will attempt to at least partially address these shortcomings through the development of an efficient, quantitative survey procedure for estimating surf smelt reproductive success. Puget Sound Treaty Tribes should find the procedures useful if they wish
to confirm the existence and location of forage fish stocks within their usual and accustomed fishery area, or if they intend to initiate intertidal aquaculture projects in Puget Sound, and wish to determine the potential impact to forage fish stocks within their project sites. Additionally, new information collected on surf smelt spawning activity will add to the current forage fish spawning database maintained by WDFW.

The third year of this project focused on the continued development of quantitative sample collection and sample analysis protocols for surf smelt and Pacific sand lance spawn deposition. Specific objectives addressed in this report were to 1) sample specific index sites within Kilisut Harbor to establish an annual index of surf smelt and Pacific sand lance spawning success within the study area, 2) collect data on surf smelt and Pacific sand lance larval incubation and emergence, 3) assess the precision of our sampling protocols, and 4) investigate potential spawning cues in relation to observed spawning densities.

STUDY AREA

Indian Island (Figure 1), located in Jefferson County, Washington, southeast of Port Townsend, was chosen as the initial study site for this project due to the high level of security and the tribal access provided by the Navy to relatively undisturbed intertidal areas. The Island is 5 miles long by 1.5 miles wide and totals 2,716 acres. Indian Island is bounded by Kilisut Harbor to the east, Port Townsend Bay to the west and north, and Oak Bay and Portage Canal to the south. The Island uplands are owned entirely by the U.S. Navy and are used primarily for the handling and storage of naval ordnance. Tidelands surrounding the Island exhibit a mixture of ownership between the U.S. Federal Government and the State of Washington.
Figure 1. Map of the study area including surf smelt spawning locations documented by past studies (yellow regions) and putative spawning locations documented by PNPTC (blue regions).
METHODS

Index Site Selection

Putative surf smelt spawning areas around Indian Island were identified by the presence of specific substrate types and through prior documentation of surf smelt spawn by WDFW and the North Olympic Salmon Coalition (NOSC). We established putative spawning areas as those beach segments in the +5 ft and +10 ft (Seattle District) tidal elevation range having substrate characterized by a sand/small gravel mixture, usually with fine shell fragments incorporated. Substrate composition in these areas was approximately 80% by weight of materials in the size range of 1-7 mm (Pentilla 1978). The start and stop points of each beach segment exhibiting these characteristics were marked by their Latitude/Longitude coordinates using a hand-help GPS unit. The continuous length of each segment was then estimated using a surveyor’s tape. All marked beach segments were then divided into 100 ft transects and each transect was assigned a unique identification number. Three beach segments were then selected to serve as index sampling sites. These segments were not selected randomly but were chosen based on their proximity to vehicle access points and the presence of previously documented surf smelt spawning activity (Long et al 2005, Pierce et. al 2009). One pair of 100 ft transects were then selected randomly from the pool of available transects within each of the three index sites.

Field Procedures

Each index site was sampled twice a month on semi-lunar tides from September-April using a randomized survey design. This design was characterized by randomly selecting a start point within the first 10 feet of each transect. A collection point was then randomly chosen within four linear feet below to one linear foot above the transect midline (+7.5 foot tidal elevation) at 10 foot intervals for a total of 10 subsamples per transect. This five linear foot
boundary for subsampling was chosen because data from Study Year 1 indicated that most eggs of both species were deposited in the +5 to +8 foot tidal elevation range.

Each subsample point was marked with a survey flag and a quadrat of spawning substrate was removed with a custom-built, rectangular sampling frame (Figure 2). The sampling frame consisted of three 12 in x 14 in stainless steel panels riveted together with a supporting crossbar attached across the top of the open front. Two angled stainless steel flanges were fixed to the exterior sides of the frame and two flat metal depth guides were fixed to the interior sides to control the depth of sample retrieval. The sampling frame was pushed into the substrate to a depth of one inch until the outer flanges made solid contact with the beach surface. A boundary across the exact midline of the sampler were marked and materials in front this mark were moved aside. A stainless steel capture tray was then forced into the interior of the sampling frame using the interior depth guides to isolate a consistent 12 in wide x 6 inch long x 1 inch deep slice of spawning substrate.

The frame and capture tray were lifted upward and the contents transferred to heavy gauge plastic bags. Individual tags were then placed in each subsample bag indicating the date, beach segment, transect and subsample location. Each subsample was then rinsed with seawater through a series of graded sieves to condense the bulk samples down to particles in the 0.5 – 1 mm grain size. Each condensed sample was then transferred to 1 L Whirl-Pak bags, preserved with a 5% formalin solution and stored in the lab for subsequent egg separation and identification.

During Study Year 1, it was assumed that any proto-larval from surf smelt and sand lance collected in intertidal substrates were not truly larvae but rather egg-stage embryos that had ruptured the egg case during formalin preservation. This assumption was based on discussions
with Dan Penttila who noted that it was unlikely that larvae would remain in the beach substrates after hatching and past investigations had not enumerated larval smelt and sand lance in beach substrates. Based on this evidence, we categorized all surf smelt and sand lance not contained within an egg case as late-eyed stage eggs. However, early egg analyses during Study Year 2 showed large numbers of yolk-sac stage larvae in the beach substrates. Subsamples of these larvae were sent to Dan Penttila and confirmed as larval stage fishes, not egg stage individuals. Thus during Study Years 1 and 2, surf smelt and sand lance larvae were enumerated as such.

**Figure 2.** Photo of the quadrat sampling frame used to retrieve quantitative subsamples of beach substrates bearing eggs of intertidal spawning forage fishes.
Lab Procedures

Separation of forage fish eggs and larvae (hereafter referred to as embryos; EMB) from the substrate samples was aided by the use of an elutriation device modified from the design described in Whitman et al. (1983). The elutriator consisted of an inverted 0.3 gal plastic cone with a section of 35 mesh bolting cloth and clear plastic tubing clamped to the narrow end of the cone (Figure 3). The other end of the plastic tubing was clamped to a faucet and the cone was held over the sink at a slight angle with a ring stand and adjustable clamp. A five gal plastic bucket with a hole cut in the bottom was positioned under and around the cone to prevent the loss of outflowing materials. A 35 mesh sieve was fitted into the opening in the bottom of the bucket to retain the egg-bearing materials removed by the elutriator. Each condensed subsample was placed into the wide end of the cone and water was pumped up through the narrow end of the cone for five minutes at a rate of two to three gallons per minute. The resulting upward force of the rinse water caused heavier materials in the subsample (e.g. sand grains and small pebbles) to circulate within the cone without flowing out the top. Lighter materials (e.g. forage fish embryos, shell fragments and silt) were forced out of the funnel and into the retention sieve at the bottom of the bucket. Materials in the retention sieve were then rinsed into a glass beaker for egg identification and enumeration. Larval fish were differentiated from ruptured egg stage embryos by the following characteristics: 1) the absence of any egg-case material adhered to the embryo, 2) an elongated, not coiled, body form, and 3) a diminished yolk-sac.

For low density samples (samples containing < 200 EMB), every EMB present was enumerated, identified to species and assigned a developmental stage. For high density samples (samples containing > 200 EMB), a volumetric subsampling method commonly used in zooplankton count applications was used to enumerate EMB (Frolander 1968). This
subsampling involved diluting egg bearing materials with water in a graduated beaker. The
diluted sample was then mixed thoroughly by hand until the materials were evenly distributed.
A Hensen-Stempel pipette was then used to draw three separate two ml aliquots that were placed
in a Bogoroff counting chamber and examined under a dissecting microscope at a 4x
magnification. A total count for each species and developmental stage was then extrapolated by
multiplying the number of EMB counted per ml by the total diluted volume of egg materials
present. Counts were averaged and expressed in a catch-per-unit-effort (CPUE) term of
EMB/ft\(^2\) of beach material. The result was an EMB CPUE data set characterized by species,
developmental stage, sample date and transect location.

Figure 3. Photo of the elutriation device used to separate forage fish eggs from beach substrates.
**Data Analyses**

There is no hypothesis testing presented in this progress report as its goal is descriptive in nature. Descriptive statistics (means, standard deviations, coefficients of variation, and confidence intervals) are the main tools of analysis. However, the efficacy of the sampling approach was evaluated by calculating the relative precision of the 95% confidence interval among years. The target precision for egg abundance estimates was for confidence intervals that did not vary more than ±30% from sample means. The efficacy of the elutriation process for removing EMB from beach substrates was tested using seeded samples. The results of the seeded trials are discussed in the summary report for Study Year 1 (Hatch and Miller 2012). Inter-annual comparisons of EMB catches are also provided using descriptive statistics.

**RESULTS**

**Catch Summary for Study Year 3**

During Study Year 3 (September 2013-April 2014), we collected 476 samples that yielded an estimated 172,127 smelt (mean CPUE, 95% Confidence Interval of 723 ± 187 EMB/ft²) and 174,832 sand lance (735 ± 196 EMB/ft²; Figure 4). Smelt CPUE peaked in October (1,485 ± 481 EMB/ft²; Figure 5) and sand lance CPUE peaked in November (2,743 ± 1,394 EMB/ft²; Figure 5). Beach 4 yielded average smelt densities of 110 ± 41 EMB/ft² and negligible sand lance densities (Figures 6-8). Beach 6 yielded average smelt densities of 1,328 ± 385 EMB/ft² and average sand lance densities of 766 ± 217 EMB/ft². Beach 9 yielded average smelt densities of 345 ± 186 EMB/ft² and average sand lance densities of 1,299 ± 547 sand EMB/ft² (Table 1).
Table 1. Summary catch statistics by index site and year for surf smelt and Pacific sand lance.

<table>
<thead>
<tr>
<th>Index Site</th>
<th>Species</th>
<th>Statistic</th>
<th>Study Year 1</th>
<th>Study Year 2</th>
<th>Study Year 3</th>
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<tr>
<td>4</td>
<td>Smelt</td>
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<td>762</td>
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<td></td>
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<td></td>
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<td></td>
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<td>205</td>
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<tr>
<td>4</td>
<td>Sand Lance</td>
<td>Negligible Catch</td>
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<td>*</td>
<td>*</td>
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<td></td>
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<td>256</td>
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<td>29</td>
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<td>Coefficient of Variation (%)</td>
<td>272</td>
<td>224</td>
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<td>Sand Lance</td>
<td>Mean CPUE</td>
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<td>632</td>
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<td>549 - 982</td>
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<td>530</td>
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<td></td>
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<td>42</td>
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<tr>
<td></td>
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<td>Coefficient of Variation (%)</td>
<td>328</td>
<td>296</td>
<td>255</td>
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We collected an estimated 3,698 surf smelt larvae (15.5 ± 5.3 larvae/ft²) and 20,210 Pacific sand lance larvae (112.6 ± 36.5 larvae/ft²) during Study Year 3. Larval abundance of smelt peaked in December (Mean CPUE of 40 ± 31.5 smelt larvae/ft²). Larval abundance of Pacific sand lance peaked in January (409 ± 215 sand lance larvae/ft²). Quantitative sampling indicated that larval emergence diminished by April when mean larvae CPUE approached 0 larvae/ft² for both species.

Figure 4. Surf smelt (*Hypomesus pretiosus*) and Pacific sand lance (*Ammodytes hexapterus*) mean egg catch-per-unit-efforts by study year. Error bars represent the 95% confidence interval.
Figure 5. Surf smelt (*Hypomesus pretiosus*) and Pacific sand lance (*Ammodytes hexapterus*) mean egg catch-per-unit-efforts by month. Error bars represent the 95% confidence interval.

Figure 6. Surf smelt (*Hypomesus pretiosus*) and Pacific sand lance (*Ammodytes hexapterus*) mean egg catch-per-unit-efforts at index site 4 by study year. Error bars represent the 95% confidence interval.
Figure 7. Surf smelt (*Hypomesus pretiosus*) and Pacific sand lance (*Ammodytes hexapterus*) mean egg catch-per-unit-efforts at index site 6 by study year. Error bars represent the 95% confidence interval.

Figure 8. Surf smelt (*Hypomesus pretiosus*) and Pacific sand lance (*Ammodytes hexapterus*) mean egg catch-per-unit-efforts at index site 9 by study year. Error bars represent the 95% confidence interval.
Inter-Annual Comparisons

Catch rates for smelt embryos during Study Year 3 (723 ± 187 EMB/ft²) were very similar to those observed in Study Year 2 (727 ± 193 EMB/ft²) but were approximately 10% higher than catch rates from Study Year 1 (648 ± 198 EMB/ft²). Pacific sand lance catch rates during Study Year 3 (735 ± 196 EMB/ft²) were approximately 40% lower compared to Study Year 2 (1220 ± 425 EMB/ft²) and approximately 75% higher than in Study Year 1 (189 ± 67 EMB/ft²). During Study Year 3, smelt catches peaked in October (1,485 ± 481 EMB/ft²) and sand lance peaked in November (2,743 ± 1,394 EMB/ft²). Smelt catches in Study Year 2 peaked in September (3,675 ± 1350 EMB/ft²) and sand lance peaked in November (4,560 ± 2028 EMB/ft²). Smelt catches in Study Year 1 peaked in November (3,731 ± 1,221 EMB/ft²) and sand lance also peaked in November (1,822 ± 635 EMB/ft²). Natural mortality of smelt EMB (expressed as the percentage of dead eggs per total catch) was 27% in Study Year 3 compared to 33% in Study Year 2 and 16% in Study Year 1.

Relative precision (RP) of the 95% confidence interval for total annual density averages of both species improved during Study Year 3 (smelt RP 25.9%, sand lance RP 26.7%) compared to Study Year 2 (smelt RP 26.5%, sand lance RP 34.7%) and Study Year 1 (smelt RP 30.5%, sand lance RP 35.5%).

Assessment of Potential Spawning Cues

Results from Year 1 indicated moon phase as a potential spawning cue as most young eggs (1/2 coil stage or younger) of both species were captured when the moon was 30% full or less. Similar results were observed in Study Year 2 with most young eggs of both species occurring when the moon was 40% full or less. Analysis of moon phase in relation to catch rates
during Study Year 3 is pending. Average water temperatures during Study Year 3 (8.8 °C) were similar to those observed during Study Year 2 (8.9 °C) and slightly higher than Study Year 1 (8.6 °C; Figure 9). However, any association between changes in water temperature and inter-annual changes in catch rates has yet to be clarified.

Figure 9. Mean daily water temperatures recorded near the study area from September 1 through April 30 of study years 1, 2 and 3.

**DISCUSSION**

The patchiness of egg counts seen in this study may be a result of spawning behavior and microhabitat conditions rather than of the sample design. Pentilla (1978) reported surf smelt egg densities that varied by as much as an order of magnitude among samples within homogenous stretches of spawning substrate. Both surf smelt and sand lance exhibit similar spawning behavior characterized by large numbers of fish loosely grouped near the water’s edge followed by a sudden coalescing of numerous smaller groups which release eggs and milt in a frenetic manner. Visual observations of sand lance spawning made during our field sampling suggested
that eggs are released in distinct patches ranging from 1-3 feet in diameter, often distributed in
no discernible pattern. Therefore, random sampling shortly after a spawning event may result in
some quadrats falling directly on spawning points with others falling on areas where no eggs
were deposited. Wave action would presumably disperse eggs both medially and laterally, and
thus decrease density patchiness as eggs incubate on the beach. However, our study found
relatively high variance among subsamples even for eggs at later stages of development. One
explanation for this result may be differential mortality among groups of incubating eggs at the
microhabitat scale. The risks of mortality induced by desiccation, predation or other causes
could vary greatly within relatively short stretches of beach (Lee and Levings 2007).

Catch rates for smelt showed much less inter-annual variation than did those for sand
lance. One explanation for this difference may be that surf smelt show stronger site fidelity to
spawning areas than sand lance and therefore their annual egg deposition may be more
predictable. Martin and Swiderski (2001) conducted phylogenetic analysis of beach spawning
fishes and determined that Osmerids (including surf smelt) belong to a clade characterized by
ancestral anadromy. Their work supports the hypothesis that beach spawning in Osmerids is a
modification of anadromy and is characterized by synchronized seasonal spawning in habitats
separate from those typically used by adults for non-reproductive purposes. The relatively
narrow range of substrate types preferred by surf smelt spawning would suggest at least some
degree of homing. However, preliminary genetic stock analysis in Puget Sound has not
supported this hypothesis (Penttila 2007). A more direct explanation for the differences in inter-
annual catch rates is that our indices are reflecting differences in overall abundance between the	
two species among years. A longer time series of data will help clarify whether or not changes
in annual egg deposition of the two species moves in tandem or if other interspecific factors are involved.

Our results indicate a greater spatial overlap of egg deposition between surf smelt and Pacific sand lance than suggested by previous research. Although past studies have verified that eggs of both species can be found co-incubating in the same substrates, the consensus has been that sand lance typically prefer substrates with finer grain sizes found at lower tidal elevations (lower limit +5 feet) than surf smelt (lower limit +7 feet; Penttila 2007). We were concerned that these presumed differences in substrate preferences would necessitate separate sampling protocols for the two species. In our study however, high egg densities of both species were observed at tidal elevations between the +6 and +7.5 elevation marks. Furthermore, visual observation of sand lance spawning on an incoming tide on several occasions during November of all three Study Years indicated that fish did not initiate egg deposition until water levels had reached a tidal height of approximately +6.5 feet. Based on these observations, we consider a single sampling protocol sufficient for simultaneously sampling egg deposition of both species.

Although spawn deposition of the two species does not appear to be segregated by tidal elevation, other spatial or temporal preferences may exist. In our study, the index site nearest the mouth of the harbor (site 4) yielded negligible sand lance eggs whereas the site nearest the head of the harbor (site 9) yielded the highest sand lance catches in two of three study years. Beach segments near the head of the harbor are in a depositional zone and tend to have finer substrates than segments near the mouth of the harbor. Sand lance may be preferentially selecting for these substrate types or may be influenced by other factors associated with beach segments near the head of the harbor. In all three study years, sand lance catch rates peaked during November whereas smelt catches peaked in November during Study Year 1, September during Year 2 and
October during Year 3. We have yet to fully investigate explanations for these differences. However, it is possible that sand lance rely on some suite of predictable spawning cues (e.g. photoperiod, tidal cycles) more so than do smelt.

The elutriation method developed in our study appears to be an effective, quick and easily standardized method for separating surf smelt eggs from substrate particles. The method was less effective in recovering sand lance eggs however (97% recovery rate for smelt, 74% recovery rate for sand lance; Hatch and Miller 2012). One explanation for this dissimilarity may be that differences in the physical properties between the two egg types affect their buoyancy. Surf smelt eggs are typically larger and adhere to one or two substrate particles at a single attachment site. Sand lance eggs are smaller and are fully encapsulated in sand grains after fertilization. Therefore, sand lance eggs are less buoyant in relation to the surrounding substrates than surf smelt eggs and less likely to be rinsed upwards during elutriation. However, the recovery rate of sand lance eggs appears to be fairly consistent and thus a correction factor may be appropriate to account for unrecovered eggs.

Modifications and expansions are planned for the fourth year of this study. A major goal of the second phase of the study is to expand our sampling to areas outside of Kilisut Harbor. We have established index sites at Port Gamble Bay and at Fort Townsend State Park. These new sites will not only provide data on spawning trends for a larger geographic area, but also provide an opportunity to assess the relationship between habitat rehabilitation projects and spawning success. Port Gamble Bay is currently undergoing a debris removal project on tribal tidelands. The sampling area is near one of these clean-up sites on Point Julia. The sampling area at Fort Townsend State Park is adjacent to a rock-armored bulkhead that is scheduled for partial removal in the summer of 2015. We also plan on evaluating alternative methods of separating eggs from
substrate particles. We are currently collaborating with WDFW on comparing the elutriation method to alternative methods (Appendix 1).
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More eggs in less time: New methods for improved forage fish egg detection

Phillip Dionne (WDFW); Shannon Miller (PNRTC); Ned Pittman (WDFW); Betty Bookheim (WDNR)

Introduction
To help protect Surf Smelt and Sand Lance spawning habitat from anthropogenic impacts such as shoreline development, considerable effort has been dedicated to mapping the locations of spawning beaches. Much of the description of the geographic distribution and timing of their spawning has been acquired through the collection, processing, and analyses of sediment samples to detect the presence of eggs. The eggs are small and translucent, so they are difficult to observe on the beach unless they occur in high densities. To expedite the egg discovery process the methods published in the Field Manual for Sampling Forage Fish Spawn in Intertidal Shore Regions by Moulton and Penttila (2006) were developed, and are generally accepted as the standard methods throughout Puget Sound. In an effort to improve the efficiency and consistency of sample processing, we’ve built upon these standard methods to develop and begin testing two alternative methods. The preliminary results of this testing are described below.

Methods tested

1. Dish pan method: Winnowing separation
2. Funnel method: Elutriation separation
3. Blue bowl method: Cyclonic separation

All three methods rely on the fact that Surf Smelt eggs are less dense than the sand and gravel they were laid in, but they each use a different means to separate and collect the eggs from the sediment.

Field trials

Samples were collected following the procedures described in the Field Manual for Sampling Forage Fish Spawn in Intertidal Shore Regions by Moulton and Penttila (2006), with the exception that roughly 3 to 4 times the normal volume of sediment was collected from each site. The sediment was then sieved, homogenized, and split into roughly equal quarters. Each quarter was then processed using one of the 3 methods being tested. Each quarter of a given sample was processed by the same person.

Lab processing

Once processed, each sample was preserved and analyzed in the lab using a microscope. Lab analyses were timed and followed the procedures described by Moulton and Penttila (2006). However, for some samples egg counting was terminated after reaching a specified number of eggs, or after a specified period of time. Also, for some samples several steps described in Moulton and Penttila (2006) were skipped because the volume of sediment retained was so small, that no additional reduction was warranted. Each quarter of a given sample was analyzed by the same person.

Preliminary results

The following 3 graphs are a comparison of method 2 and method 3 as a proportion of the results of method 1 for a given trial. In these figures, if the proportion is greater than 1.0, then the method performed better than method 1; if the proportion is less than 1.0, the method performed worse than method 1.

Users’ impressions

After being trained and given the opportunity to use all three methods, each user was given a simple survey to help us understand how they viewed the different methods. Generally, users ranked method 3 most favorably, followed by method 2, and method 1.

Discussion

Our preliminary results indicate that the new methods perform well and are generally preferred compared to the standard winnow method and when utilized by relatively novice users. However, each method has its strengths and weaknesses that should be considered when deciding which method may be best suited for a given study. For example, we found that the funnel method had a lower sediment capacity which resulted in longer processing times, so the funnel method may perform better than our reported results for studies that use smaller volumes of sediment. Also, if equipment cost is the deciding factor, then the standard winnow method may be the best choice.