# EXHIBIT K-2

## EVALUATION REPORT

## SMELT

## (REVISED)

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# Evaluation Report
# Migration Timing and Distribution of Columbia River Smelt In The Lower Columbia River (Revised)

The attached report provides a final summary of studies undertaken to characterize the nature and extent of eulachon (smelt) *Thaleichthys pacificus* spawning and larval migration in the lower Columbia River. The overall goal of the study was to use the information colleted to assess the potential effects on eulachon, if any, of the proposed project to deepen the Columbia River shipping channel.

The main objectives of the 2 year study were to (1) determine the presence or absence of egg deposition and larval migrants within and adjacent to specific reaches of the Lower Columbia River navigation channel; (2) use the information acquired to asses the potential effects of dredging; and (3) depending upon the outcome of objective number three, determine if any measures are necessary to minimize the potential effects of dredging to the overall eulachon population.

The findings and recommendations in general were that dredging activities associated with channel deepening are not expected to have a significant impact on migrating eulachon larvae (through entrainment), on eulachon spawning areas, or on eulachon eggs incubating in nearshore areas in the proximity of dredging activities. Impacts to smelt spawning areas from disposal are generally not a concern because most in-water disposal sites are downstream of the lowest major smelt spawning areas, which are at CRM 56-61 and 67-69. While the current construction plan has some limited inwater (flowlane) disposal in CRM 59-62, this disposal is unlikely to directly impact eulachon spawning areas because the dynamic nature of substrates within the flowlane disposal sites (which are in or adjacent to the main channel) do not provide stable surfaces that would allow an adhesive egg to incubate for 30 days. Impacts to migrating larval smelt from disposal are a concern to the agencies and though they are unsure of the level of impact, they have indicated in the attached letter that disposal not occur during the peak of the larval movement downstream. The peak out migration in 2001 was from the 2nd to the 18th of April but can vary. The period of peak larval out-migration will be determined by the agencies prior to construction, but will likely fall within, or near this period. The Corps has agreed to schedule construction dredging and disposal to avoid this period. No additional specific actions (e.g., timing restrictions) are recommended because it is unlikely that dredging associated with channel deepening would have a significant impact on eulachon.

January 9, 2003

Mr. Kim Larson
U. S. Army Corps of Engineers
Programs and Project Management Division
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Re: Recommendations for potential in-water disposal of dredged materials associated with the Columbia River Channel Improvement Project.

Dear Kim,

In November, 2002, we provided an assessment of the potential impacts of channel deepening activities on eulachon based on study findings in our final report, *Eulachon studies related to lower Columbia River channel deepening operations*, edited by David L. Ward. In our report we stated, "Disposal is generally not a concern because in-water disposal sites are downstream of the lowest major eulachon spawning area." In a recent conversation (January 3, 2002), you explained to me that hopper disposal of dredged materials associated with the Columbia River Channel Improvement Project might occur in the channel within river mile reaches 51-56 and 59-61. You asked that we clarify our recommendations in light of this information.

I have conferred with some of the members of the smelt mitigation workgroup (Dave Ward and Patty Snow with Oregon Department of Fish and Wildlife, and Brad James and Steve Manlow of Washington Department of Fish and Wildlife). What follows is our consensus opinion and recommendations for potential in-water disposal of dredged materials associated with the Columbia River Channel Improvement Project, as it relates to eulachon.

Generally, we do not expect the described disposal will affect eulachon spawning habitat. Further our 2001 study showed that eulachon larvae disperse widely and that the shipping channel was not the primary outmigration corridor. However, larval densities were greater at mid water column and near the river bottom (where dredged materials will be released), and these areas are adjacent to and immediately downstream of one reach identified as an important main stem spawning area (river mile 56 to 61) and a major spawning tributary -- the Cowlitz River. We are concerned that larval eulachon survival may be reduced by an increase in suspended particles, but we do not know a mortality rate or the magnitude of potential losses.

Our recommendations for in-water disposal are intended to protect eulachon larvae during the period of peak outmigration and in areas where they are most abundant. We recognize some losses may occur if disposal happens at anytime during the period of eulachon outmigration

(January through June). However, the eulachon migration is variable, protracted, and sporadic, and larvae disperse widely in the river. Further, we are uncertain of the mechanism or potential magnitude of losses. As such we feel a period of restriction that protects outmigrating larvae during their period of greatest abundance is appropriate.

The following recommendations for in-water disposal are based on findings from our 2001 eulachon study.

1. No in-water disposal should occur in areas shallower than 43 feet along the Washington shore between river mile 35 and 75. Eulachon were found to spawn throughout this area and this restriction will protect spawning habitat.
2. No in-water disposal should occur during the period of peak eulachon outmigration downstream from identified spawning areas (river miles 35-75). Peak eulachon outmigration in 2001 was April 2-18, but this varies in magnitude and duration among years. Since 1988, peak landings of adult eulachon have ranged from the 4th to the 16th week of the year, with most peaks falling between weeks 5 and 11. We would expect peak outmigration to fall about four weeks after peak landings. Further analysis of historic data may better define the peak outmigration period.
3. If in-water disposals are essential during the period of peak outmigration further study is needed to estimate potential eulachon losses.

Thank you for bringing plans for in-water disposal to our attention. I hope that these additional recommendations will be useful in completing an Environmental Impact Statement that will minimize fishery losses that may result from the Columbia River Channel Improvement Project.

Sincerely,

//ss//
Tom Rien
Research Project Leader
Eulachon and Sturgeon Studies

cc: Ward, Snow, Nigro (ODFW)
James, Manlow (WDFW)

# EULACHON STUDIES RELATED TO LOWER COLUMBIA RIVER CHANNEL DEEPENING OPERATIONS

Edited by:

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Final Report to U. S. Army Corps of Engineers
Contract Number W66QKZ13237198

November 2002

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# REPORT A

# PROJECT SUMMARY AND FINAL RECOMMENDATIONS

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## PROJECT SUMMARY

This report provides a final summary of studies undertaken to characterize the nature and extent of eulachon (smelt) *Thaleichthys pacificus* spawning and larval migration in the lower Columbia River. The overall goal of the studies is to use the information collected to assess the potential effects on eulachon, if any, of the proposed project to deepen the Columbia River shipping channel. The study is a cooperative effort by the Oregon Department of Fish and Wildlife, the Washington Department of Fish and Wildlife, and the U. S. Army Corps of Engineers. Field studies were conducted in fiscal years (FY) 2000 and 2001, with FY 2002 dedicated to completing data analyses and preparing reports. Previous progress was summarized in our reports of results for FY 2000 and FY 2001.

The main objectives of the studies were to (1) determine the presence or absence of egg deposition and larval migrants within and adjacent to specific reaches of the Lower Columbia River navigation channel; (2) use the information acquired to assess the potential effects of dredging; and (3) depending upon the outcome of work to address Objective (2), determine if any measures are necessary to minimize the potential effects of dredging to the overall eulachon population. Field activities in FY 2001 were geared toward meeting Objective 1. Information from both years of field work were used to meet Objective 2. This report addresses Objective 3.

Most sampling occurred between Columbia River Mile 30 and 85 (approximately Three Tree Point to the Cowlitz River mouth). In 2000 we focused all sampling within or proximate to areas that have been proposed for channel deepening. In 2001 the study area was expanded somewhat. To evaluate gears we sampled in the Cowlitz River, and we sampled for eulachon larvae upstream of the Cowlitz River mouth when it became apparent that adults had moved upstream. In 2001 sampling was conducted over a broad cross-section of the Columbia River channel to characterize the density of larval migrants relative to that in proposed channel-deepening areas. Also in 2001, sampling for eggs was conducted in relatively nearshore areas to characterize the distribution of spawning in the Columbia River.

Final project findings are detailed three subsequent reports. These reports are:

| | |
|---|---|
| Report B | Migration Timing and Distribution of Larval Eulachon *Thaleichthys pacificus* in the Lower Columbia River, Spring 2001; |
| Report C | Use of an Artificial Substrate to Capture Eulachon *Thaleichthys pacificus* Eggs in the Lower Columbia River; and |
| Report D | Characterization of Development in Columbia River Prolarval Eulachon *Thaleichthys pacificus* Using Selected Morphometric Characters. |

Highlights from these reports include:

1. Closeable plankton nets were used to collect larval eulachon at seven transects in the Lower Columbia River from river mile 34 to river mile 100 (Report B).
2. Larval abundance was highly variable through time, by cross-channel location, and with depth. Larvae were present in the Columbia River from January through May 2001. Peak abundance occurred in early/mid-April compared with mid-March in 2000 (Report B).
3. The shipping channel was not observed to be the primary migration corridor. Highest catches of larvae were observed at stations located nearer to the Washington shoreline downstream from major spawning areas (Report B).
4. Although sampling with artificial substrates demonstrated that mainstem spawning occurred throughout the study area, it appears that this input to the larval population is less significant than that from the Cowlitz River. An exception is the Barlow Point locale where larval abundance was observed to be very high (Report B).
5. Larvae were distributed throughout the water column at all sampling locations. At sampling locations situated within the shipping channel larvae were generally more abundant at the bottom and middle of the water column than at the surface (Report B).
6. Artificial substrates were used to collect eulachon eggs in the lower Columbia River. We sampled from river mile 30 to 85, with at least two artificial substrates placed in all but one mile. We did not attempt to standardize or stratify substrate placement among depths or habitat types. Depths of sampling ranged from 3 to 42 feet and distance from the riverbank ranged from 15 ft to over 300 ft. Among 147 sets, eggs were collected in 23, all between river miles 35 and 73. The greatest number of eggs were captured in river miles 56 to 61 and 67 to 69 (Report C).
7. Egg catch per unit effort varied with sampling time and location. In areas that eggs were collected the bottom composition varied, yet was dominated by medium to fine sand. The sample rate was low given the size of the study area, therefore caution was used in applying this finding (Report C).
8. Ripe eulachon were collected from the Cowlitz and Sandy Rivers and artificially spawned in the laboratory at ODFW Clackamas. Eggs were successfully incubated in water filled petrie dishes and hatched 47 days after fertilization, accumulating 752 thermal units – approximately twice that documented by previous workers (Report D).
9. Larvae were allowed to develop in petrie dishes and survived 21 days before total yolk-sac absorption. Larvae were preserved in formalin at various post-hatch ages for subsequent morphometric evaluation (Report D).
10. Trends in larval growth and yolk-sac absorption were observable over time, however, individuals of each age class showed high variability in the chosen morphometric criteria such that development was not statistically identifiable on a time scale of days (Report D).

11. Static environmental conditions in our experiment appear to have retarded larval development. Morphometric analyses of larvae collected in the field from a known spawning area (the Cowlitz River; river mile 68) and the Columbia River mainstem at river mile 34 showed that identifiable development does occur as larvae out-migrate to the estuary and ocean (Report D).

## FINAL RECOMMENDATIONS

The following assessments of the potential impacts of channel deepening activities on eulachon are based on report findings. In general, dredging activities associated with channel deepening are not expected to have a significant impact on migrating eulachon larvae (through entrainment), on eulachon spawning areas, or on eulachon eggs incubating in nearshore areas in the proximity of dredging activities. Disposal is generally not a concern because in-water disposal sites are downstream of the lowest major smelt spawning area.

1. Given the large numbers of eulachon larvae and their distribution across the river channel and throughout the water column it is unlikely that dredging associated with channel deepening would have a significant impact (through entrainment) on the migrating larval population.
2. Dredging associated with channel deepening is unlikely to directly impact eulachon spawning areas. Given the dynamic nature of substrates within the reaches proposed for channel deepening, these reaches do not provide stable surfaces that would allow an adhesive egg to incubate for 30 d.
3. Eulachon eggs incubating in near-shore areas in the proximity of dredging activities might be affected if these activities alter flow patterns or increase sedimentation. However, hydraulic models indicate dredging will not significantly alter the river's flow patterns. The average annual bed-load transport in the main river channel to expected to remain the same within the existing range.
4. Artificial spawning substrates may be a useful tool to better characterize the timing and location of eulachon spawning. Although more intensive sampling using egg substrates over multiple years would allow better identification and characterization of long-term spawning sites and relative levels of use among areas, this information is not deemed necessary at this time to assess the potential effects of channel-deepening.
5. As a precautionary measure to minimize any dredging effects on eulachon eggs, channel-deepening operations could be scheduled to avoid certain reaches at times in which the greatest number of eulachon eggs were collected during the peak spawning period. Two reaches identified by this study are river mile 56 to 61 and 67 to 69.

The following recommendations are based on the above assessments:

1. No specific actions are recommended because it is unlikely that dredging activities associated with channel deepening would have a significant impact on eulachon.
2. Dredging activities associated with channel deepening are not scheduled to occur in known areas of high spawning concentration. The most realistic and reliable strategy for reducing impacts from other dredging operations would be to avoid areas of high spawning concentration.

# REPORT B

# MIGRATION TIMING AND DISTRIBUTION OF LARVAL EULACHON IN THE LOWER COLUMBIA RIVER, SPRING 2001.

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## ABSTRACT

We sampled from 28 January through 1 June 2001 to (1) quantify timing of larval eulachon Thaleichthys pacificus emigration in the Columbia River, (2) determine cross-channel larval distribution during emigration, and (3) determine depth distribution of larvae within the shipping channel during emigration. This study was initiated to evaluate potential effects of proposed channel-deepening operations in the Columbia River on eulachon spawning and emigration. Closeable plankton nets were used to collect larval eulachon at seven transects in the Lower Columbia River from river mile 34 to river mile 100. Larval abundance was highly variable through time, by cross-channel location, and with depth. The shipping channel was not observed to be the primary emigration corridor. Larval densities outside the shipping channel were significantly greater than those inside the channel. Highest catches of larvae were observed at stations located nearer to the Washington shoreline downstream from major spawning areas. We collected larvae throughout the sampling period, but catches peaked between 2 and 18 April. Larvae occurred at all depths sampled, but densities were generally greater near the bottom and in mid-water than near the surface. Although sampling with artificial substrates demonstrated that mainstem spawning occurred throughout the study area, it appears that this input to the larval population is less significant than that from the Cowlitz River. An exception is the Barlow point locale where larval abundance was observed to be very high. Given the variability in distribution and timing of eulachon larval emigration, scheduling of dredging to reduce impacts to emigrating larvae would be confined to the short term. The most realistic and reliable strategy for reducing dredging-related impacts to eulachon would be to avoid dredging in areas of high spawning concentration.

## INTRODUCTION

**The eulachon *Thaleichthys pacificus*, an anadromous member of the smelt family (Osmeridae), spawns along the Pacific coast of North America, from the Pribilof Islands (Bering Sea) to the Klamath River in California (Wydoski and Whitney 1979). The lower Columbia River Basin supports one of the largest spawning runs of eulachon. In most years many eulachon spawn in the Cowlitz River, with somewhat fewer spawning in the mainstem Columbia River. Smaller, periodic runs occur in other tributaries including the Grays, Skamokawa, Elochoman, Kalama, Lewis, and Sandy rivers. Adult migration in the Columbia River system usually begins in December, peaks in February and continues through May (WDFW 2001).**

**Spawning eulachon females generally deposit eggs in areas where the substrate consists of coarse sand/fine gravel, and where water flows are "moderate" in velocity (Hart and McHugh 1944; Smith and Saalfeld 1955). Eggs adhere to the surface of the substrate and incubate over a period of about 30-40 days, depending on temperature. Upon hatching the larvae become part of the drift as (presumably) passive plankters and are rapidly transported out to sea (Hart and McHugh 1944; Hart 1973) where they rear in near-shore marine areas at moderate to shallow depths (Barraclough 1964).**

**Historically, the commercial catch of eulachon in the Columbia River system has generally been strong, yet variable. Recent annual returns, based on commercial landings, were relatively stable until 1994 when a sharp decline occurred. This trend of lower annual returns of spawning adults continued through 1999. Although the 2000 and 2001 spawning runs in the lower Columbia River appear stronger (according to catch data), the relative magnitude is difficult to quantify as restrictive fishery management strategies imposed in response to the recent decline in returns severely reduced commercial effort.**

**Mechanisms controlling eulachon recruitment and survival are poorly understood. Conditions in the freshwater environment during eulachon spawning may influence productivity. This study was initiated to evaluate the potential effects of proposed channel-deepening operations in the Columbia River (USACE 1999) on eulachon spawning and migration.. Dredging activity has the potential to impact eulachon through entrainment of spawning adults (Larson and Moehl 1990; McGraw and Armstrong 1990) and possible smothering of developing eggs by increased turbidity and suspended sediment in the vicinity of operations (Morton 1977; Prussian et al. 1999). Entrainment of developing eggs and migrating larvae has not been documented but remains a concern. In response to these concerns the U.S. Army Corps of Engineers (USACE) contracted the Oregon Department of Fish and Wildlife (ODFW) and the Washington Department of Fish and Wildlife (WDFW) to identify eulachon spawning sites within proposed channel deepening areas and to characterize the spatial and temporal distribution of eulachon larvae in the mainstem Columbia River during the migration period.**

**Preliminary results from the first year of the study (Howell and Uusitalo 2001) showed that eulachon larvae were widely distributed throughout the river during migration. Sampling limitations precluded determining the relative importance of the shipping channel as a migration corridor relative to the rest of the river. The objectives for this study in 2001 were to (1) quantify the timing of larval eulachon migration in the Columbia River, (2) determine cross-channel larval distribution during migration, and (3) identify the depth distribution of larvae within the shipping channel during migration.**

## METHODS

### Study Area

Previous studies have documented large spawning concentrations of eulachon in the Cowlitz River, Washington. During field sampling in the spring of 2000 (Howell and Uusitalo 2001) we found the highest densities of migrating larvae in the Columbia River downstream of the confluence with the Cowlitz River at Columbia River kilometer (RK) 110 (Figure 1; all river distances are reported using National Oceanographic and Atmospheric Administration river distances). During 2001 the majority of our effort was therefore concentrated downstream of the Cowlitz River to maximize larval catch rates.

Only one sampling transect was located up river from the confluence of the Cowlitz and Columbia rivers.

### Sampling Gear and Methods

We used a plankton net deployed from an anchored vessel to capture eulachon larvae. The net was a typical ring net design comprising a tapered nylon sock (3.35 m length, 300 µm mesh) lashed to a stainless steel circular frame (0.61 m inside diameter).

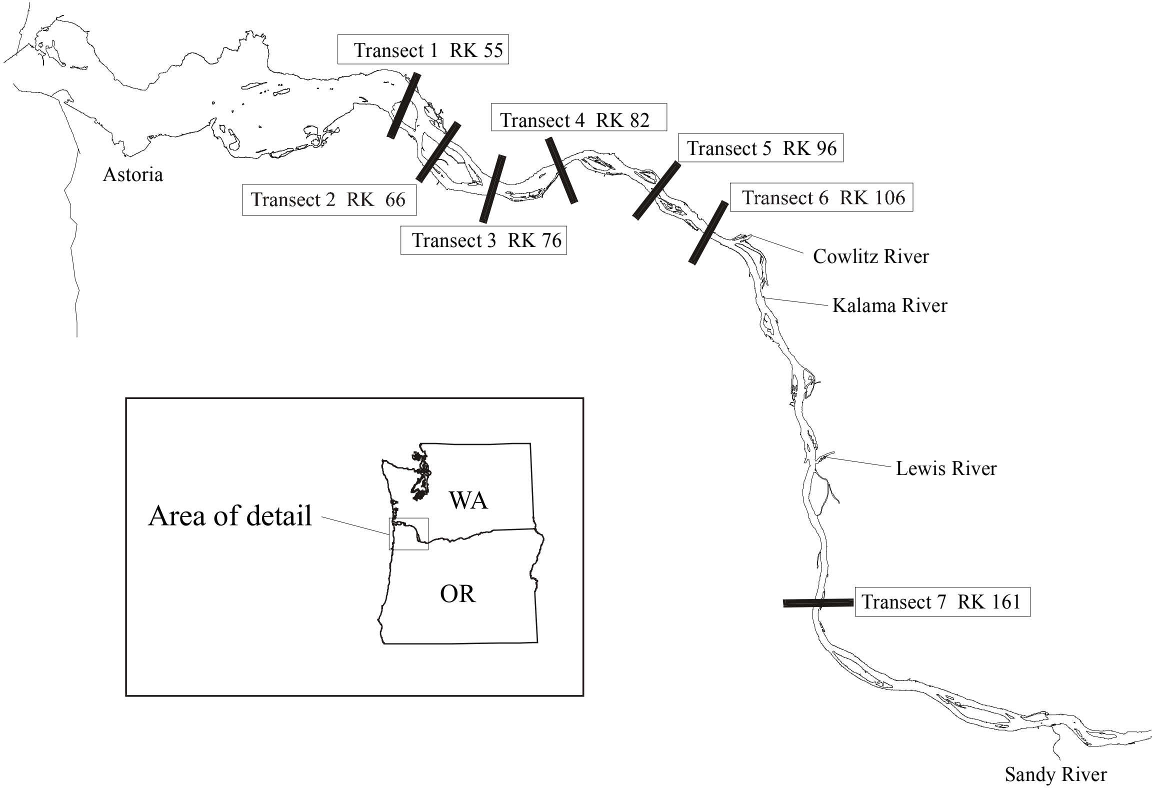

**Figure 1. 2001 lower Columbia River, larval eulachon migration study site, showing location of larval eulachon sampling transects, listed by transect name and Columbia River kilometer (RK).**

**Samples were collected in an 8.9-cm, two-piece polyvinyl chloride (PVC) collection bucket attached to the end of the sock. Spherical lead weights (2.54 kg, 9.07 kg or both) were attached to the frame base. The net was closeable via a row of choke rings placed around the sock approximately 1.3 m behind the mouth. Water flow was measured with a digital flowmeter consisting of a propeller/sensor mounted in the mouth of the net and connected to an onboard digital counter via a cable (Illustrations of net configuration are given in Appendix A).**

**We sampled during daylight hours on ebb tides. Vessels were anchored and we recorded Differential Global Positioning System (DGPS) co-ordinates, and water**

**temperature, depth, and turbidity readings. Plankton nets were lowered to the desired depth, allowed to fish for approximately 60 s, closed, and immediately retrieved. The flowmeter was activated when the net reached the desired sampling depth and stopped upon net closure.**

**Contents of the collection bucket were rinsed into storage jars and fixed with dilute (approximately 70%) ethyl alcohol. We added Rose Bengal stain to aid in laboratory examination.**

### Sampling Design

We sampled at seven transects in the lower Columbia River to characterize the cross-river distribution of eulachon larvae (Figure 1). Because of a variety of factors transects were not chosen randomly. Transect 1 (RK 55) is an index site for larval eulachon sampling (Price Island Index) that has been monitored by WDFW since 1994 (WDFW 2001). Transect 6 (RK 106) was chosen specifically to characterize the cross-river distribution of larvae in close proximity to a known spawning area (the Cowlitz River). We chose transects 2, 3, 4 and 5 (RK's 64, 75, 82 and 97 respectively) to reflect the heterogeneity in river morphology and relative position of the shipping channel within the study area. Some transects included side channels of the river, some were deeper closer to the Oregon shore, and others were deeper closer to the Washington shore. We sampled transects 1-6 eight times between 28 January and 01 June 2001 (Table 1).

During the 2001 eulachon spawning run a substantial number of adults migrated upriver of the confluence of the Cowlitz and Columbia rivers, some going as far as Bonneville Dam (RK 234). In response to expanded spawning by the strong eulachon run, we added Transect 7 in mid-season at RK 160, upstream of the confluence of Lewis and Columbia rivers (RK 139) but downstream of the confluence of the Sandy and Columbia rivers (RK 194). We sampled Transect 7 on 24 April, 27 April, 07 May, and 10 May 2001.

Each transect line was drawn roughly perpendicular to the river flow, from riverbank to riverbank. Along each transect line we established five sampling stations positioned at intervals across the river (Appendix B). At least one station at each transect was located within the shipping channel. Stations were numbered 1 through 5 across transects from the Washington shore to the Oregon shore. The number of samples collected at a station varied depending on depth. At shallow stations (< 3 m) we took samples from the bottom of the water column only. At stations of intermediate depth (≥ 3 m and ≤ 8 m) we took samples from the bottom and surface of the water column. At the deepest stations (> 8 m) we took samples from the bottom, middle and surface of the water column. For each of these depth strata we took three replicate samples in succession to account for short-term variability in larval density.

### Laboratory Methods

**Over 1,800 samples were collected and brought to the lab to be analyzed. Many of these samples contained more than 5,000 eulachon larvae and some contained as many as 30,000 larvae. Given the large number of samples taken and limited time**

**available, we used a representative subsampling method to estimate total larval counts for each sample. Each sample was emptied into an Erlenmeyer flask and total sample volume (wet) was recorded. The flask was swirled to ensure random mixing and approximately 20% of the total sample volume was poured into a graduated cylinder. The subsample was then poured into a Petrie dish and we used a dissecting microscope to count all larvae. Total sample counts were estimated by extrapolation based on subsample volumes.**

Table 1. Summary of sampling periods during 2001 larval migration study.

| Sampling Period | Dates | Comments |
|---|---|---|
| 1 | 30 January – 14 February | Gear testing |
| 2 | 28 February – 2 March | |
| 3 | 14 March – 16 March | |
| 4 | 2 April – 5 April | |
| 5 | 11 April – 13 April | |
| 6 | 16 April – 18 April | |
| 7 | 01 May – 03 May | Transect 1 not sampled |
| 8 | 11 May – 16 May | Transects 2-4 not sampled |

Catch rate for larvae was estimated as catch per cubic meter in each sample.

### Data Analysis

We estimated larval eulachon density for each sample based on laboratory count and the estimated volume of water filtered through the plankton net tow using the following digital flowmeter formula:

$$V = R\left(\frac{1}{61} m / revolution\right)A$$

Where

V = volume sampled ($m^3$),
R = revolution count from flow meter, and
A = area of net opening ($m^2$).

We examined the catch frequencies of larvae to describe the form of the catch distribution and found that the data possessed a strong negative binomial distribution with several outliers (Figure 2). Attempts to normalize the error terms by transforming data as log (catch rate + 1) failed. Consequently we elected to use non-parametric methods to test for significant differences in larval catch rate among sampling strata. We used the Mann-Whitney rank sum test for paired comparisons and Kruskal-Wallis Analysis of Variance (K-W ANOVA) for multiple comparisons. Following a significant result from the K-W ANOVA a further test was performed to isolate differences among groups (Dunn's Test when group sample size was unequal; Student-Newman-Keuls test when group sample size was equal). All tests were performed at the $\alpha = 0.05$ level of significance.

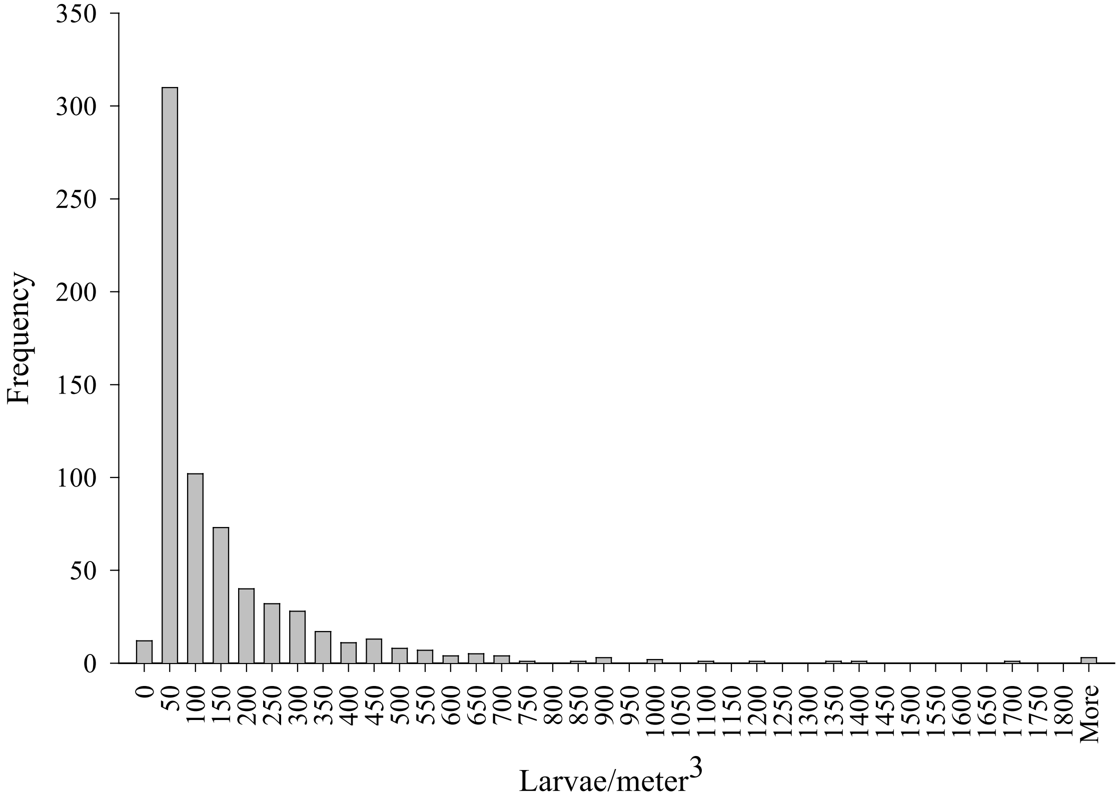

Figure 2. Density frequencies of larval eulachon collected in the Lower Columbia River during the peak of migration, spring 2001. Note negative binomial distribution.

### Migration Timing

To quantify timing of larval eulachon migration we pooled data from transects 1-6 and compared larval densities among the eight sampling periods. Contracted sampling precluded using information from Transect 7. Because most larvae were collected in periods 4-6, subsequent analyses for transects 1-6 were limited to data collected during hese peak migration periods. We also compared larval densities among transects to evaluate distribution during peak migration.

### Cross-Channel Larval Distribution

**We pooled data from transects 1-6 to compare larval eulachon densities within and outside the shipping channel during peak migration. We also compared larval densities within and outside the shipping channel for each transect. To evaluate distribution of larval eulachon across the Columbia River we pooled data from transects 1-6 for each of the five sampling stations and compared larval densities among the pooled stations. We also compared larval densities among stations for each transect.**

**Similar but separate analyses were conducted for Transect 7. Timing of sampling was different at Transect 7, and Transect 7 was relatively distant from the other transects.**

### Vertical Distribution Within the Shipping Channel

**To evaluate vertical distribution of larval eulachon within the shipping channel during peak migration we pooled data from all stations (transects 1-6) within the channel and compared larval densities among the three depth strata (bottom, mid-water, and surface). We also compared larval densities among depths at shipping channel stations for each transect. Results from Transect 7 were again interpreted separately.**

## RESULTS

### Migration Timing

Larval eulachon density varied throughout the season (Figure 3), but catches peaked between 02 April and 18 April (corresponding to sampling periods 4, 5 and 6). We found a significant difference ($P < 0.001$) in larval density among the eight sampling periods. Larval densities during periods 4, 5, and 6 were significantly greater than in all other periods but did not differ significantly from each other ($P > 0.05$). We found a significant difference ($P < 0.001$) in larval density among individual transects during peak migration, although only Transect 6 differed significantly (lower larval density) from the others ($P < 0.05$; Figure 4).

## Cross-Channel Larval Distribution

> Larval densities outside the shipping channel were significantly greater ($P < 0.001$) than densities inside the channel when data from all transects 1-6 were combined. We found no significant differences between larval densities within and outside the shipping channel at transects 1, 2, and 4 ($P = 0.524$, 0.961, 0.969 respectively) when these transects were analyzed individually (Figure 5). Larval densities were significantly greater outside the shipping channel for transects 3, 5 and 6 ($P = 0.031$, $P < 0.001$, $P < 0.001$ respectively) when these transect were analyzed individually (Figure 5). Furthermore, when limiting analyses to stations ≥ 12.2 m in depth (the minimum depth of the shipping channel), we found that larval densities were significantly greater outside the shipping channel than within the shipping channel ($P < 0.001$).

With data from transects 1-6 combined, larval densities decreased across the river from the Washington shore to the Oregon shore (Figure 6). Station 1 larval densities were significantly greater than those at all other stations, station 2 densities were significantly greater than those at stations 3, 4, and 5, and station 3 densities were significantly greater than those at station 4 ($P < 0.05$ in all cases). This trend is most apparent in transects 5-7 (Figure 7).

Larval densities were also significantly greater ($P = 0.033$) at stations located outside the shipping channel than stations located within the channel at Transect 7 (Figure 7).

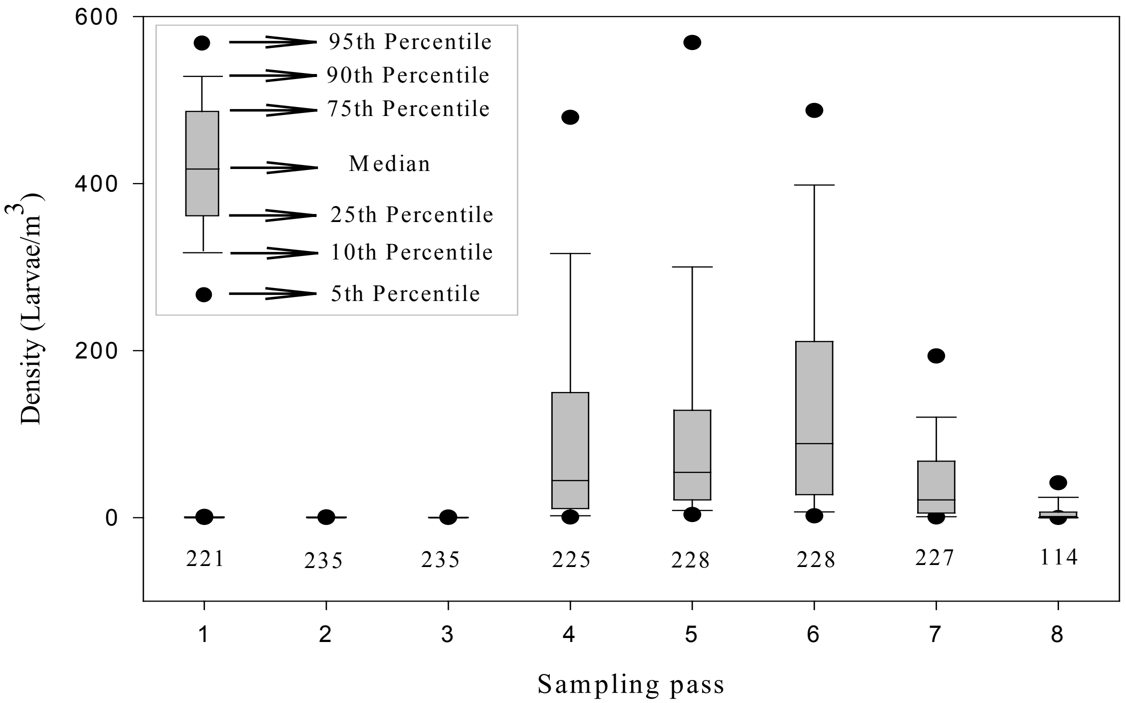

Figure 3. Larval eulachon densities in the Lower Columbia River during spring, 2000. Numbers below plots indicate sample size.

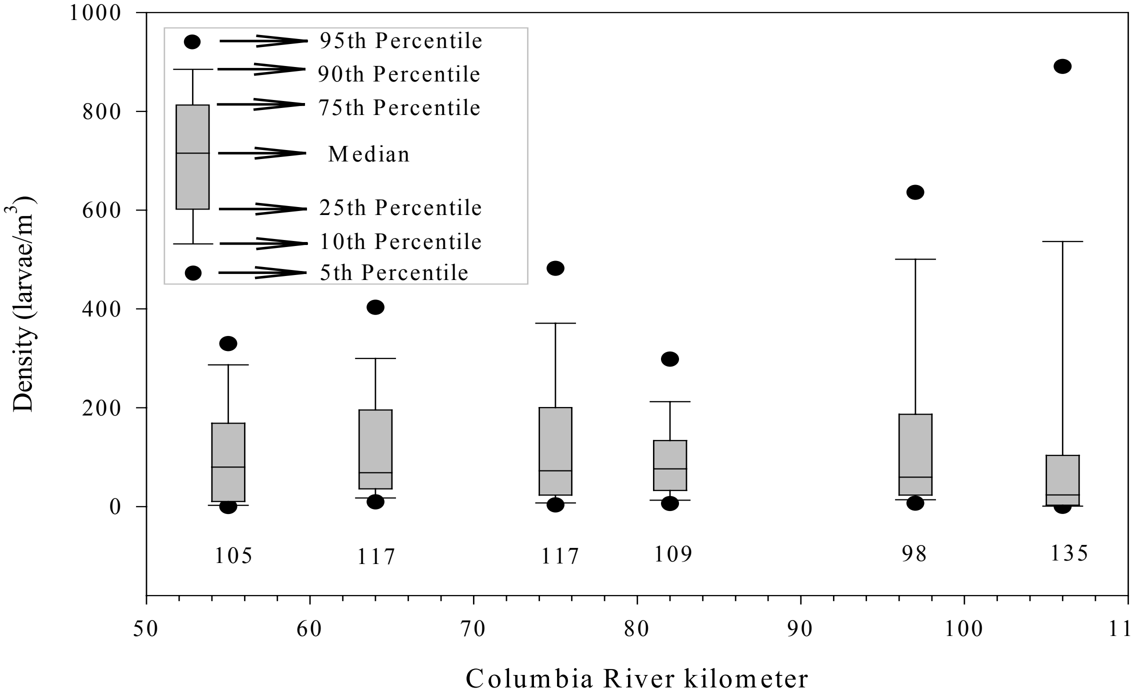

Figure 4. Larval eulachon densities at various sites in the Lower Columbia River during peak migration, spring 2001. Numbers below plots indicate sample size.

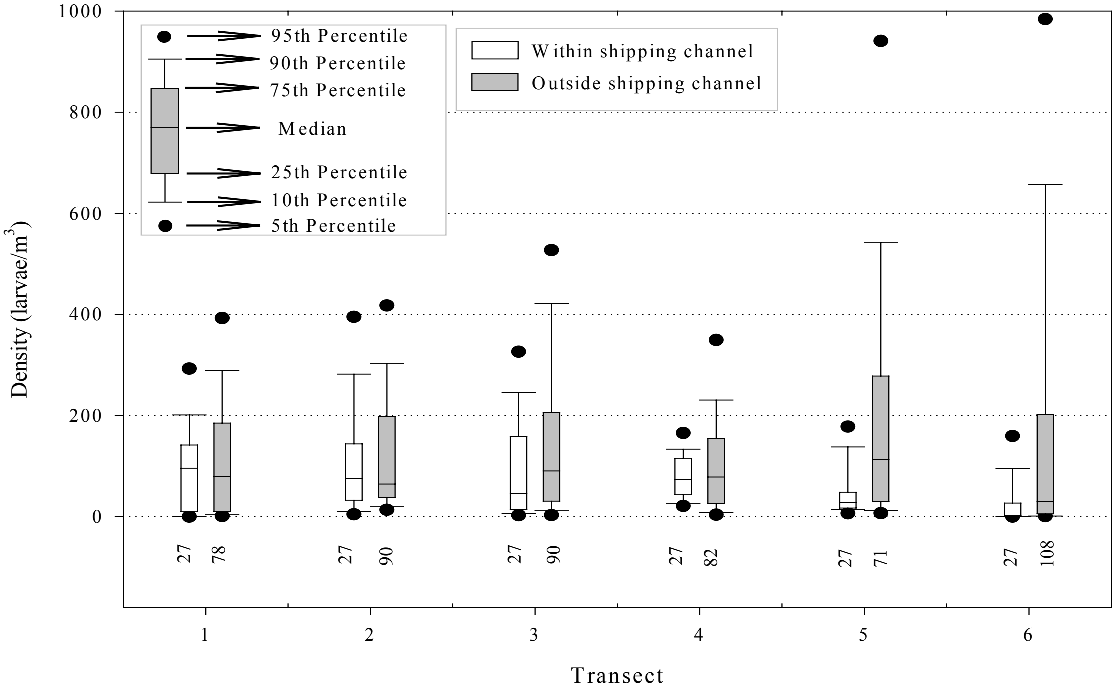

Figure 5. Larval eulachon densities inside and outside the Lower Columbia River shipping channel during the peak of migration, spring 2001. Numbers below plots indicate sample size.

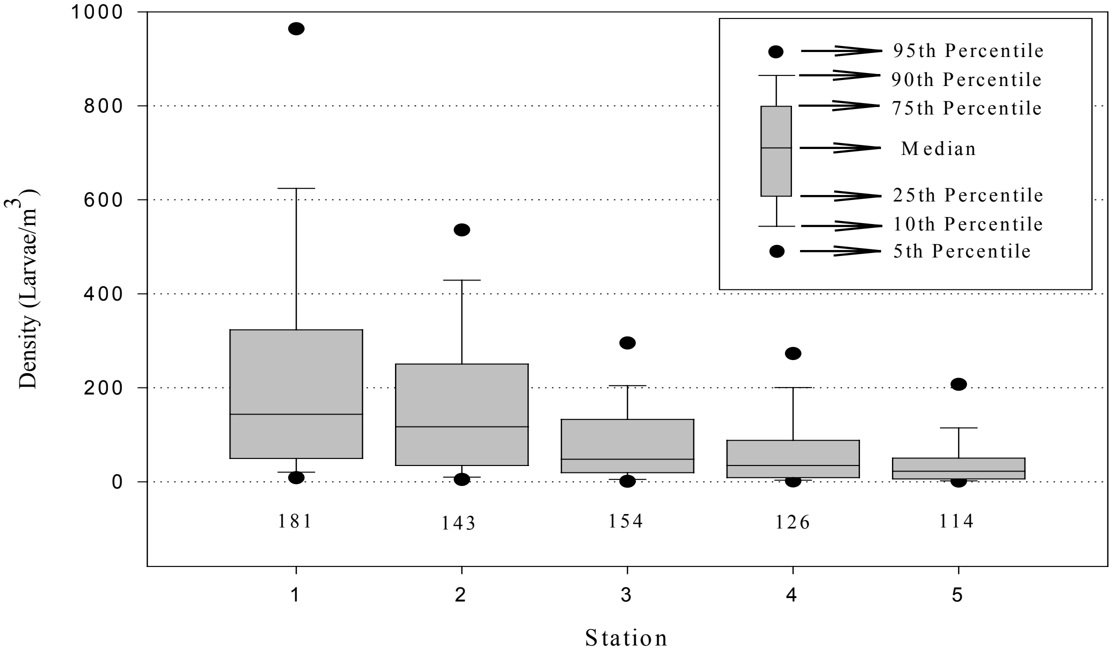

Figure 6. Cross channel distribution of larval eulachon in the Lower Columbia River during the peak of migration, spring 2001. Numbers below plots indicate sample size.

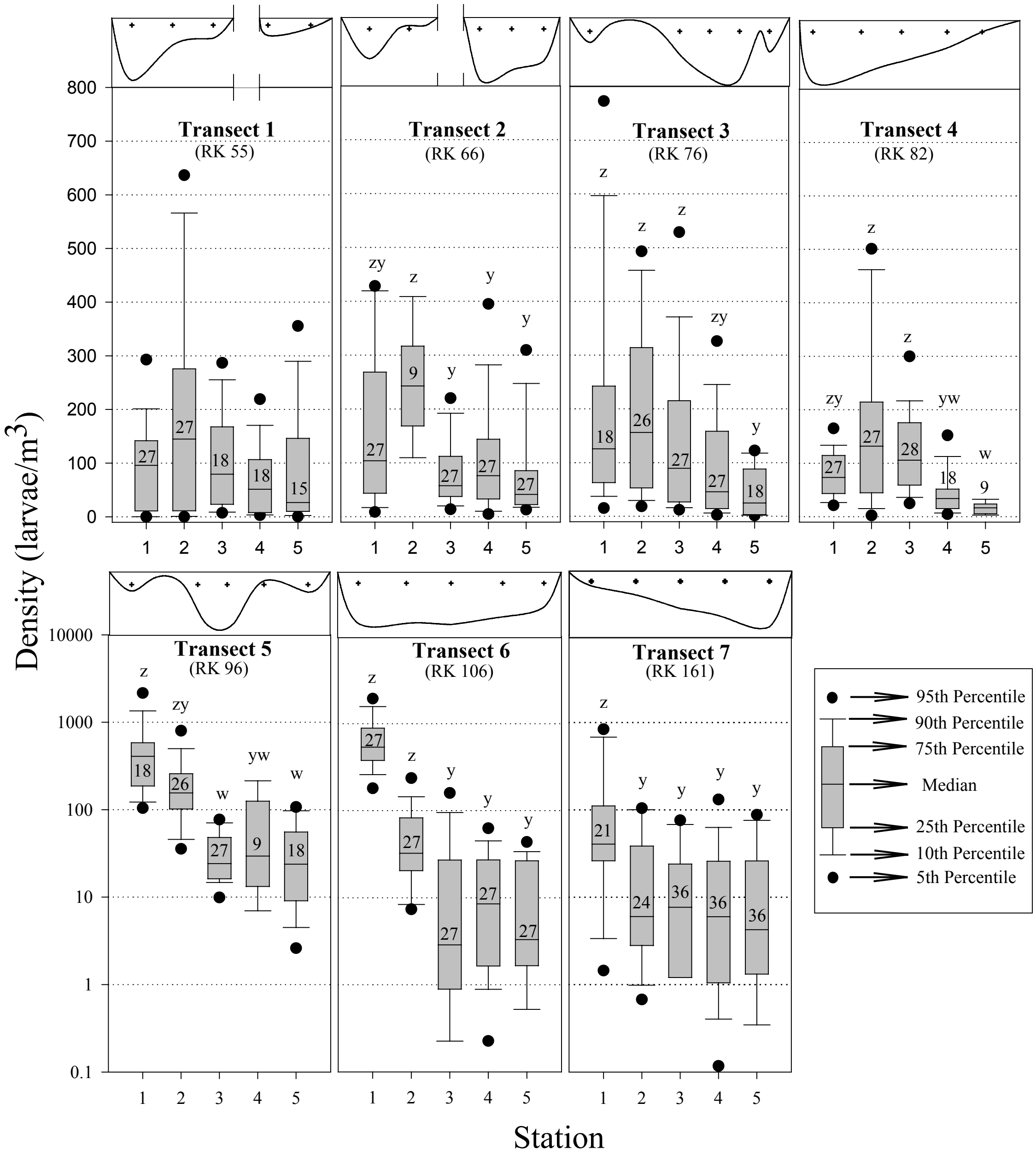

Figure 7. Cross river eulachon larval distibution at seven transects in the Lower Columbia River during the peak of migration, spring 2001. Data groups with letters contain significant differences in larval density among stations (Kruskall-Wallace ANOVA; P<0.05); within these groups, catches without a letter in common differ (P<0.05). Note logarithmic scale for transects 5 -7. Schematics of channel morphology at each transect are shown above each data group. Scaling is not consistent among plots.

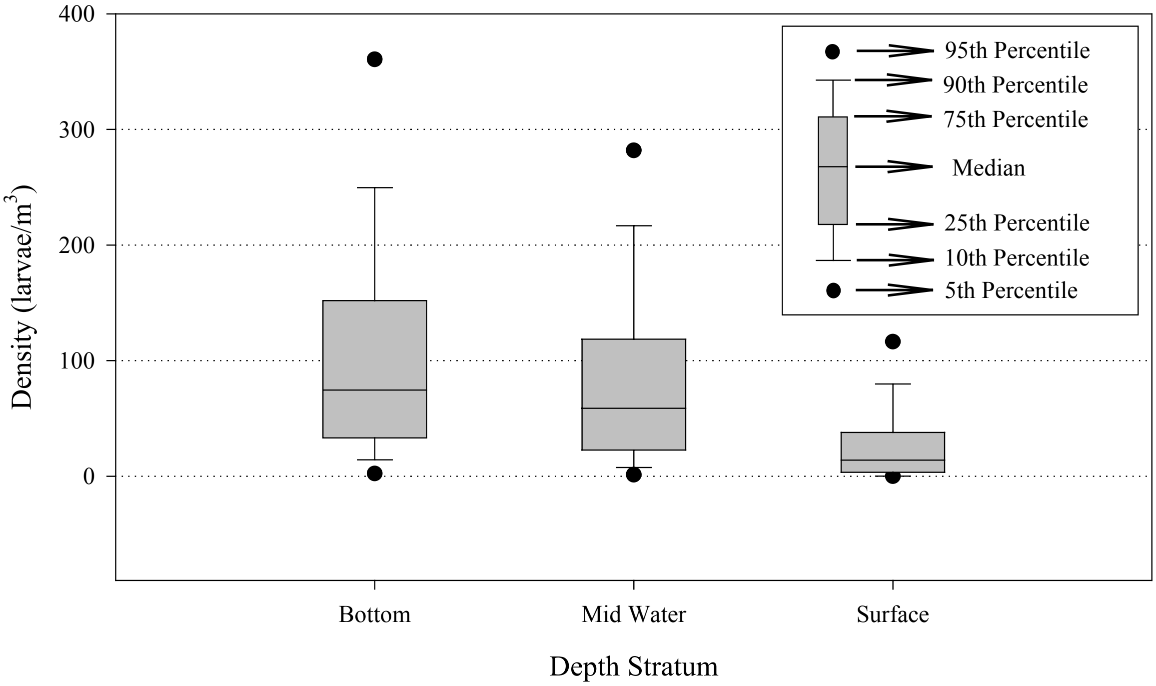

Figure 8. Vertical distribution of larval eulachon in the shipping channel of the Lower Columbia River during peak migration, spring 2001. Each plot represents 54 samples.

Catches at Station 1 were significantly greater than catches at all other stations. We found no significant differences in larval densities among stations 2, 3, 4 and 5.

## Vertical Distribution Within the Shipping Channel

We found a significant difference ($P < 0.001$) in larval densities among depth strata of sampling stations within the shipping channel (Figure 8). Larval densities did not differ significantly ($P > 0.05$) between bottom and mid-water strata; however, both bottom and mid-water larval densities were significantly greater ($P < 0.05$ in both cases) than surface larval density.

We found no significant differences in larval density among depth strata for shipping channel stations of transects 1, 4 and 5 when these were analyzed separately ($P = 0.076$, 0.067, 0.093 respectively). We did find significant differences in larval densities among depth strata for shipping channel stations of transects 2, 3 and 6 ($P = 0.006$, $< 0.001$, 0.018 respectively). Larval densities in bottom samples were significantly greater than in surface samples ($P < 0.05$ in all cases), but were not significantly different than mid-water samples ($P > 0.05$ in all cases). Mid-water larval densities were greater than surface densities for transects 2 and 3 ($P < 0.05$ for both) but not for transect 6 ($P > 0.05$).

Larval densities differed significantly among depth strata at Transect 7 sampling stations within the shipping channel ($P < 0.001$). Larval densities in bottom samples were significantly greater than densities in mid-water and surface samples ($P < 0.05$ for both).

## DISCUSSION

**The eulachon spawning migration of 2001 was one of the largest in recent years (WDFW 2001). Substantial numbers of adults migrated as far upriver as Bonneville Dam (RK 234) and into all the major lower Columbia River tributaries (Grays, Elochoman, Kalama, Lewis, and Sandy rivers). Commercial landings from the Columbia River were slightly higher than recent years but were still low compared to historical catches (WDFW 2001). This was possibly a result of restrictions on total effort during the season, because catch per unit effort (pounds per delivery) was the highest recorded since 1993.**

The timing of adult and larval migrations was later in 2001 than in 2000 (Howell and Uusitalo 2001). Peak larval densities for 2001 were recorded in early/mid April as compared to mid March in 2000. The late arrival of adults in 2001 may have been influenced by water temperature of the river. Generally, temperatures below 4 $^{o}$C inhibit entry of spawning eulachon adults into the Columbia River (Smith and Saalfeld 1955). During 2001, the bulk of adult spawners entered the river after mid February, when temperature of the Columbia River exceeded 4 $^{o}$C.

**Within the study area, migrating larval eulachon were more abundant outside the shipping channel than within the channel, with densities highest along the Washington shore. This was especially evident at transects 5 and 6. Transect 6 is located approximately 3 km downstream from the mouth of the Cowlitz River, which is a well documented spawning area for eulachon (Smith and Saalfeld 1955; Hymer 1994; WDFW 2001). Transect 5 is located < 2 km downstream from Barlow Point (Washington shore), which was identified as a likely location of eulachon spawning (Romano et al.2002).**

**Cross-channel distribution of larvae at Transect 3 does not reflect the trend seen at transects 5 and 6 despite its location downstream of, and in close proximity to, Eagle Cliff (RK 82), which is also a documented eulachon spawning site (Loeffel 1954; Smith and Saalfeld 1955). Although eulachon eggs were collected on artificial substrates in the vicinity of Eagle Cliff (Romano et al. 2002) the number of eggs caught at this location was low despite substantial sampling effort. These observations suggest that the majority of spawning in the study area could have occurred in the Cowlitz River and the Columbia River in the vicinity of Transect 5. The trends seen in successive downstream transects would then be the result of gradual cross-channel dispersion of larvae.**

Cross-channel distribution of larvae at Transect 7 (RK 161) suggests that some mainstem spawning may have occurred on the Washington shore. This is important because spawning in the Columbia River has never been recorded upstream of Martin's Bluff (RK 128; Loeffel 1954) and we collected no eggs above the mouth of the Kalama River (RK 117) during our study of eulachon spawning distribution (Romano et al. 2002). Transect

7 is also located upstream of all Columbia River tributaries in which eulachon are known to spawn, with the exception of the Sandy River (RK 194; Figure 1). Large numbers of adult eulachon were observed in the Sandy River in 2001 (personal observation, lead author) where they presumably spawned. . The even distribution of larvae across the river through stations 2 – 5 at Transect 7 suggests cross-river dispersal from a major upstream source such as the Sandy River.

> Our finding that larval eulachon density in the shipping channel was greatest in the lower portion of the water column is consistent with observations made by Loeffel (1954) and Smith and Saalfeld (1955). It is unclear what mechanisms might affect the distribution of larvae in the water column. Anecdotal laboratory observations suggest larval eulachon exhibit pelagic swim up behavior (Wendler 1937; Howell 2002) and positive phototropism (Howell 2002). This is an adaptive behavior documented in other ichthyoplankton species to facilitate feeding, lateral transport, and predator avoidance (Fortier and Leggett 1983; Manuel and O'Dor 1997). Eulachon larvae subsist on yolk sac contents on their migration to rearing areas in the Columbia River estuary and Pacific Ocean, where exogenous feeding is assumed to begin (Smith and Saalfeld 1955). Given the limits of yolk sac storage, rapid flushing to the ocean may be crucial for survival. We speculated that vertical migration of larvae into the top of the water column on ebb tides (where velocities generally are greater), might expedite the journey. Our results however do not support, and in fact somewhat contradict this theory. The lower Columbia River is subject to strong tidal influences that produce complex, turbulent flow conditions and because larval eulachon are relatively weak swimmers, depth distributions are most likely dictated by local hydraulic conditions.

Sampling for this study was conducted during daylight hours and on ebb tides only. This study design does not allow analysis of any effects of diel and tidal cycles on larval distribution. In addition, inter-annual variation in spawning site locations and run size may also influence distribution. In a year of high spawner abundance such as 2001, larval abundance was not significantly greater in proposed dredging areas than in other areas of the river. In years of high abundance, dredging-related mortality (through entrainment) may not be significant relative to the population as a whole. Without data from multiple seasons it is not possible to know how larval distribution during migration differs (if at all) in years of low abundance. In addition, mechanisms controlling eulachon recruitment and survival are poorly understood, and little is understood on how variability in habitat conditions in the freshwater environment affects larvae survival.

> Given the variability in distribution and timing of eulachon larval migration, scheduling of dredging to reduce impacts to migrating larvae would be confined to the short term. Unlike spawning runs of most anadromous salmonid species, where estimates of stock size provide the basis for development of reliable forecasts, no developed forecasting or assessment model exists for eulachon. Currently only in-season commercial monitoring exists to evaluate run size and timing. Perhaps the most realistic and reliable strategy for reducing dredging related impacts to eulachon

would be to avoid dredging in areas of high spawning concentration. This would require more research on the annual variation in use of specific spawning areas.

## ACKNOWLEDGMENTS

We thank Peter Barber, Brad Cady, Jody Gabriel, Michele Hughes, Robin Mills, Francesca Saenz, Steve West, and Nicole Wolters for assistance in field collection of the data and laboratory analyses. John DeVore (WDFW) and David Ward (ODFW) helped with initial study conception, design, and analysis of data. David Ward (ODFW) and Brad James (WDFW) reviewed a draft of this manuscript. Kim Larson (USACE) provided assistance as project manager. The U. S. Army Corps of Engineers funded this work (contracts W66QKZ10745277 and W66QKZ13237198).

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Romano, M. D., M. D. Howell, and T. A. Rien. 2002. Use of an artificial substrate to capture eulachon eggs in the lower Columbia River. Report C i*n* D. L. Ward editor. Eulachon studies related to Lower Columbia River channel deepening operations. Final report by the Oregon Department of Fish and Wildlife and Washington Department of Fish and Wildlife to the U. S. Army Corps of Engineers. Contract Number W66QKZ13237198.

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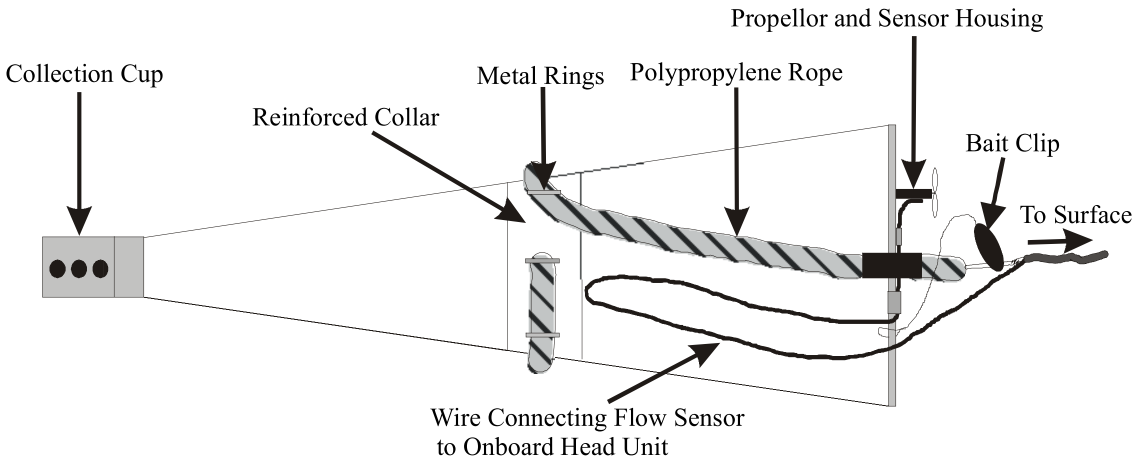
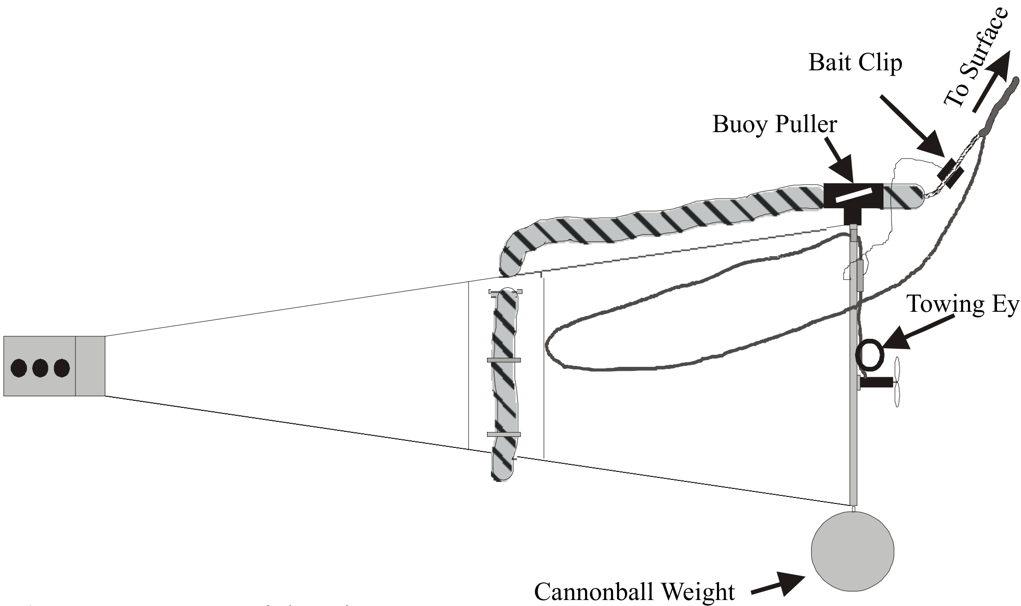
# APPENDIX A

Schematic diagrams of modified plankton net gear used in 2001 larval eulachon sampling

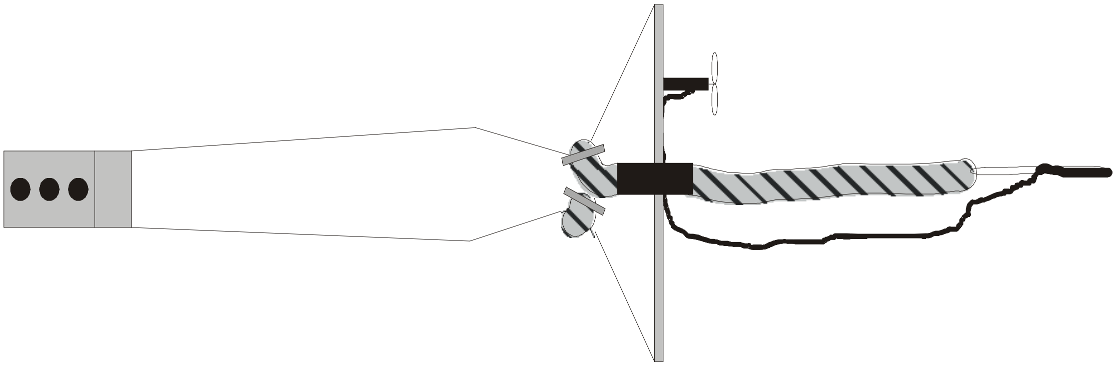


I) Net Open - Plan View

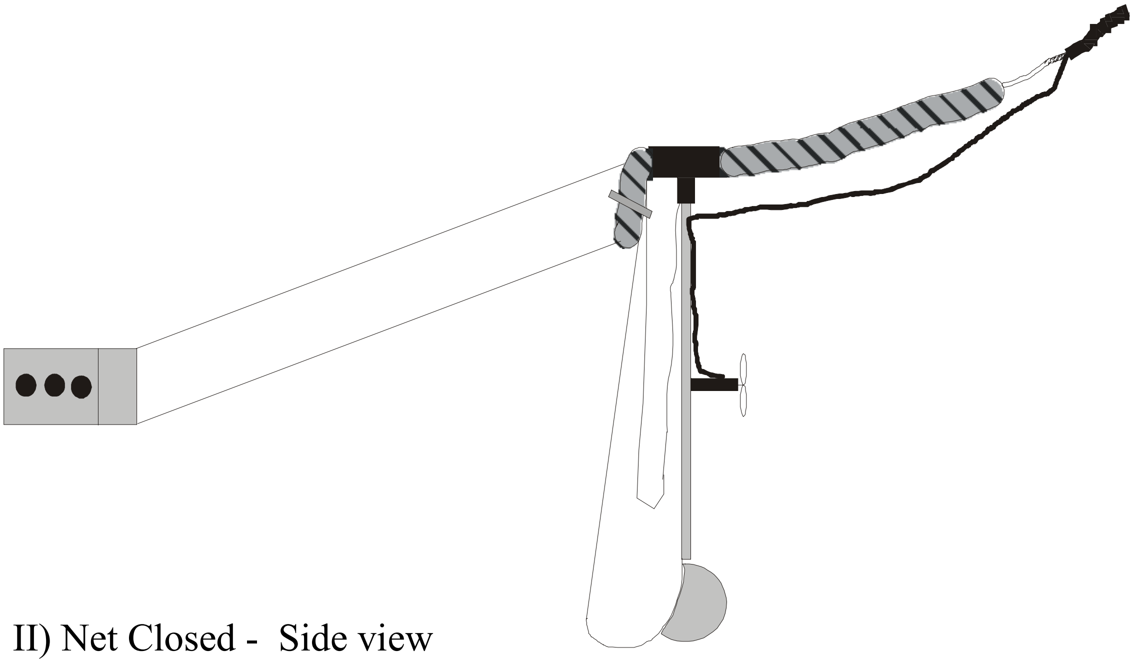


II) Net Open - Side view

**Appendix Figure A-1. Schematic diagrams of modified plankton net used in 2001 USACE larval smelt sampling.**

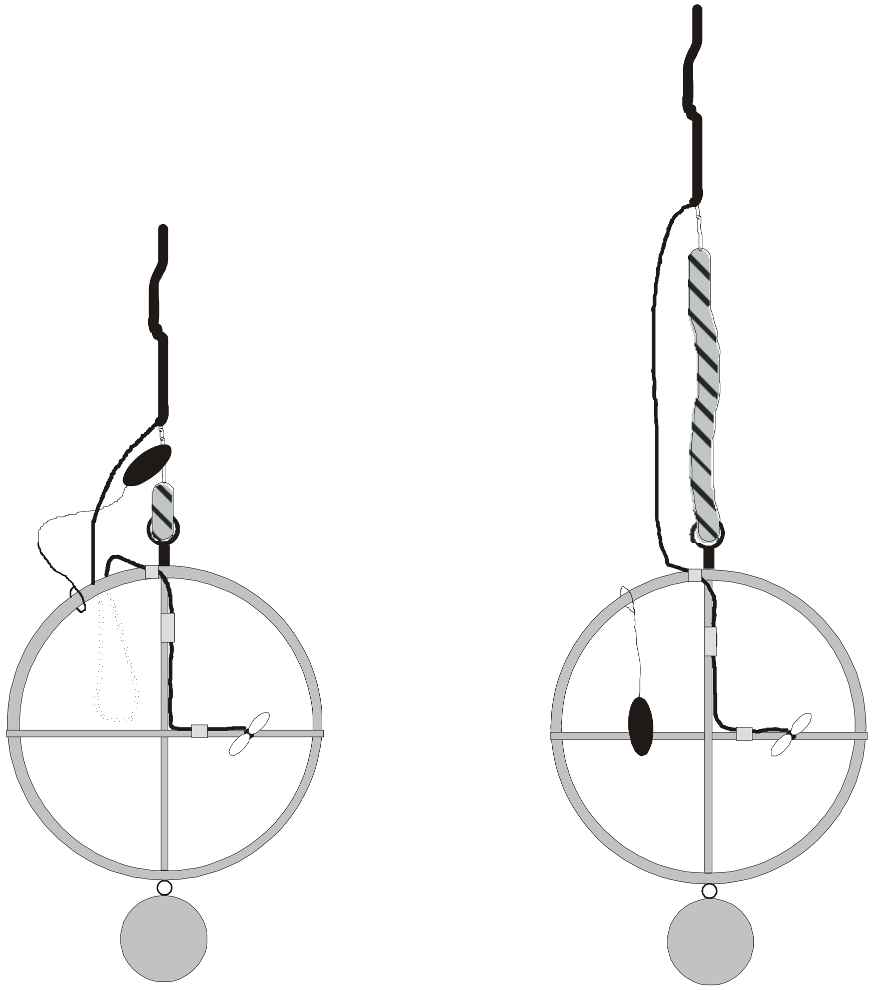


I) Net Closed - Plan View



II) Net Closed - Side view

Appendix Figure A-2. Schematic diagrams of modified plankton net used in 2001 USACE larval smelt sampling.

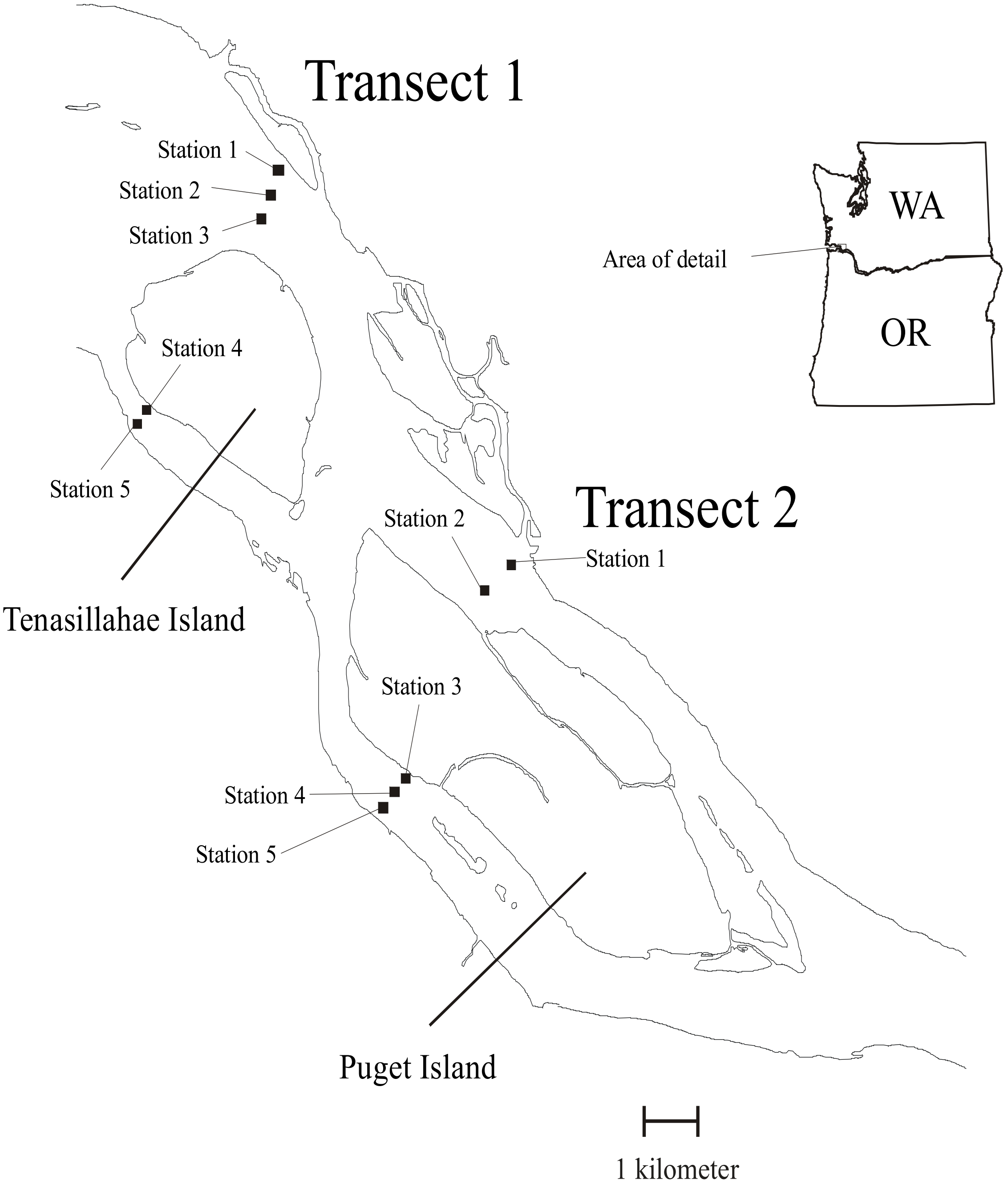


I) Net Open - Forward View II) Net Closed - Forward View

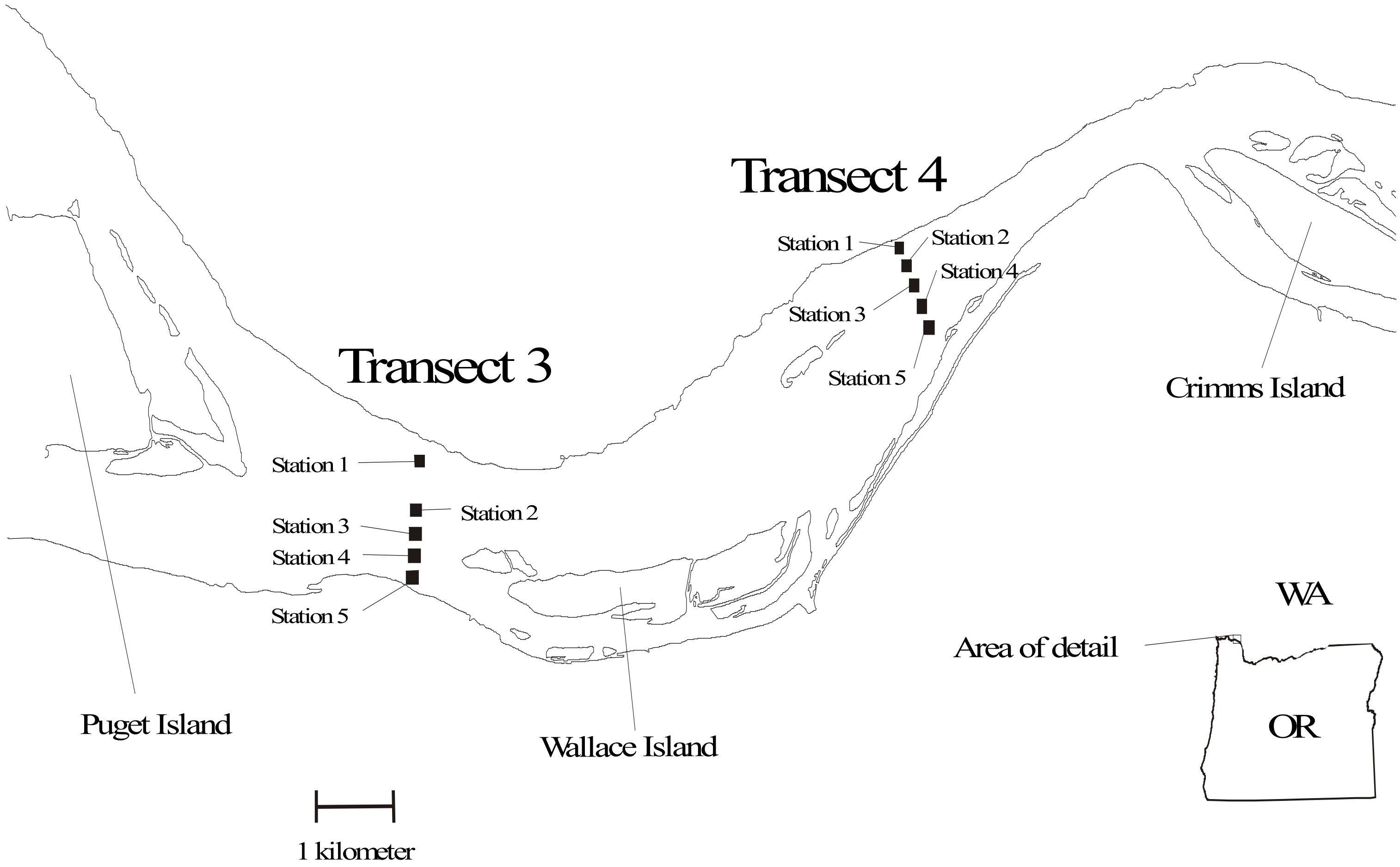
Appendix Figure A-3. Schematic diagrams of modified plankton net used in 2001 USACE larval smelt sampling.

# APPENDIX B

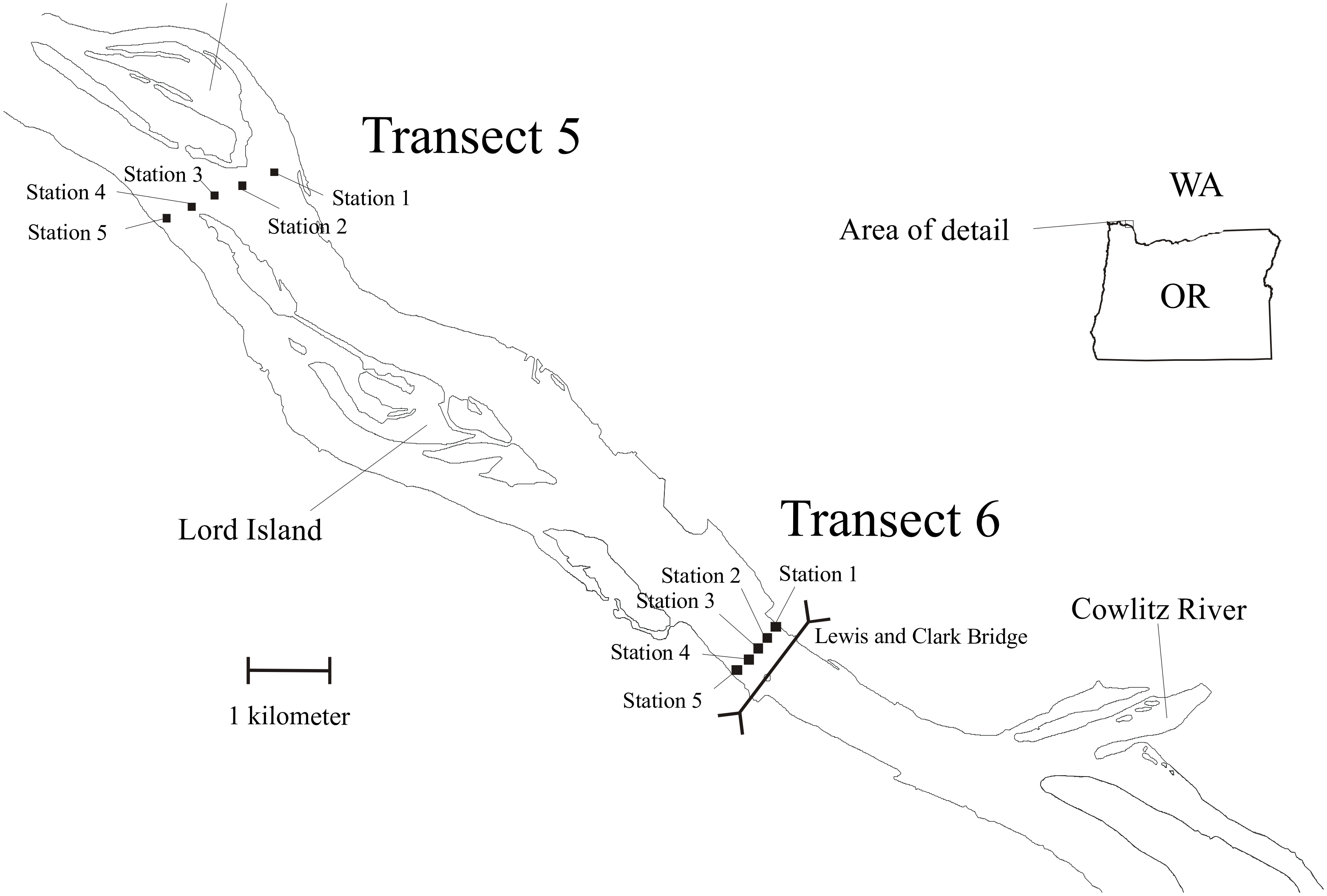
Larval eulachon sampling sites, 2001



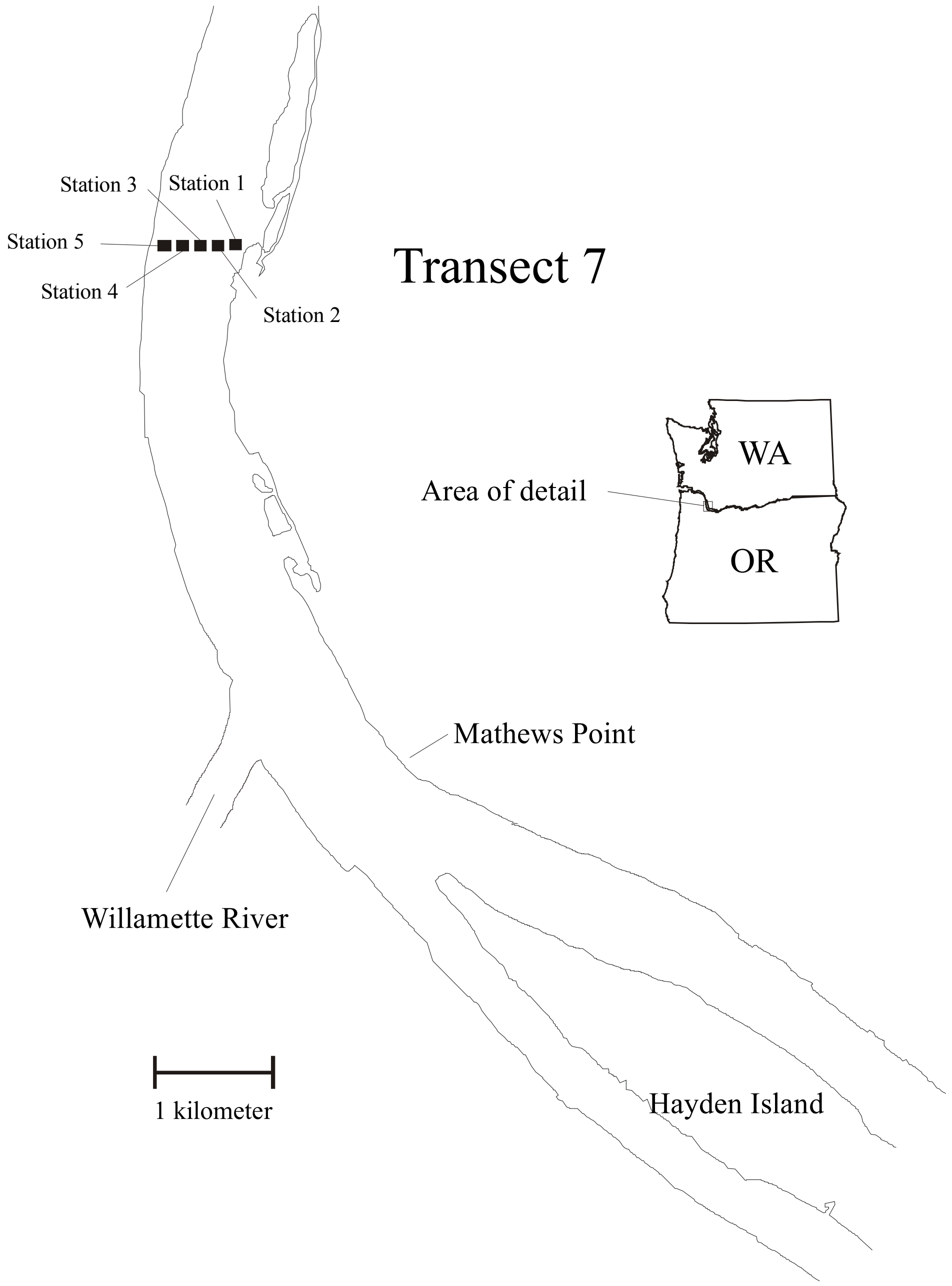
**Appendix Figure B-1. 2001 USACE larval smelt sampling sites, Transect 1 (RK 55) and Transect 2 (RK 66), lower Columbia River.**



**Appendix Figure B-2. 2001 USACE larval smelt sampling sites, Transect 3 (RK 76) and Transect 4 (RK 82), lower Columbia River.**



**Appendix Figure B-3. 2001 USACE larval smelt sampling sites, Transect 5 (RK 96) and Transect 6 (RK 106), lower Columbia River.**



Appendix Figure B-4. 2001 USACE larval smelt sampling sites, Transect 7 (RK 161), lower Columbia River.

## REPORT C

# USE OF AN ARTIFICIAL SUBSTRATE TO CAPTURE EULACHON EGGS IN THE LOWER COLUMBIA RIVER

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Final Report to U. S. Army Corps of Engineers
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## ABSTRACT

We used artificial substrates to collect eulachon (Thaleichthys pacificus) eggs in the lower Columbia River from river kilometer (RK) 48 to 137 over the period 26 February – 29 March 2001. This method has been used to capture eggs of other species, but this is the documented use of substrate frames to catch eulachon eggs. We did not attempt to standardize, stratify or randomize substrate placement among depths or habitat types, but typically two substrates were fished every 1.6 km. Depths of sampling ranged from 1 to 13 m, and distance from the riverbank ranged from 5 to >90 m. Eggs were collected in 23 of 147 sets, all between RK 55 and 120. We captured eggs throughout the sampling period and peak catch rates occurred on 9 and 13 March 2001. The greatest numbers of eggs were captured in RK 90 to 98 and RK 107 to 111. The bottom composition of areas in which eggs were collected varied, but was dominated by medium to fine sand. We conclude that in 2001 eulachon spawned over a wide range of the mainstem lower Columbia River. Further, we believe that artificial substrates can be a useful tool to assist in identifying the timing and location of eulachon spawning.

## INTRODUCTION

The eulachon *Thaleichthys pacificus* is an anadromous fish that spawns in the lower reaches of 30 to 40 coastal rivers and streams, from the Klamath River drainage in California, to the Bering Sea, Alaska. The Columbia River supports one of the world's largest spawning populations of the species (DFO 1999), and is the site of an important commercial and recreational eulachon fishery. Historically, commercial landings of eulachon in the Columbia River have been highly variable. In 1948 and again in 1951 the commercial catch of eulachon in the Columbia River exceeded 450,000 kg, yet in 1992 and again in 1994 the commercial catch fell below 450 kg. A sharp decline in commercial landings occurred in 1990 and continued through 1999. It is unclear how much of the decline is caused by a decrease in the number of spawning eulachon or by in-season adjustments to harvest regulations. The 2001 commercial eulachon fishery exceeded 79,400 kg and was considered to be quite strong (WDFW and ODFW 2001).

Eulachon spawning in the Columbia River generally begins in January or February and is completed by late April. Spawning adults have been observed in the river as early as December. Active spawning has been observed in tributaries of the Lower Columbia River including the Cowlitz, Kalama, Lewis, and Sandy rivers (Smith and Saalfeld 1955). Little is known about the spawning distribution of eulachon in the mainstem lower Columbia River. Eulachon eggs have been caught in plankton nets in the lower Columbia River but exact spawning locations have proved difficult to locate. Smith and Saalfeld (1955) identified two locations in the mainstem Columbia River as eulachon spawning locations (one upriver of the mouth of the Kalama River at river kilometer (RK) 117 and the other near RK 82). These findings were based on the presence of spent and partially spent fish in commercial catches from these areas.

Depending on her size, a female eulachon can produce from 17,000 to 60,000 eggs (Hart and McHugh 1944; Smith and Saalfeld 1955). Eulachon eggs are small, with an average diameter between 0.8 and 1.0 mm. Eulachon eggs contain a double membrane, the outer of which ruptures shortly after fertilization and remains attached to the egg by a single point, forming a short stalk or peduncle. The free edges of the outer membrane are highly adhesive and capable of sticking to substrate material (Hart and McHugh 1944). Smith and Saalfeld (1955) reported that eulachon spawn primarily over substrates of fine pea-sized gravel.

The lower Columbia River is routinely dredged to maintain a shipping channel with a minimum depth of 12.2 m and minimum width of 182 m. The U. S. Army Corps of Engineers (USACE) has proposed an increase in dredging operations to deepen the existing channel, which would allow larger vessels access to the ports of Longview, WA and Portland, OR (USACE 1999). To assess potential impacts of channel deepening operations on eulachon, the USACE contracted with the Oregon Department of Fish and Wildlife (ODFW) and the Washington Department of Fish and Wildlife (WDFW) to characterize the eulachon spawning run and larval migration from the lower Columbia River. The objective of this study was to locate and characterize eulachon spawning sites within the lower Columbia River, including the proposed channel-deepening area.

This study is based on the assumption that if eggs are captured on an artificial substrate then adult fish have spawned in the immediate vicinity. Prior to this study, artificial substrates had not been used to catch eulachon eggs. Artificial substrates have been used to collect white sturgeon *Acipenser transmontanus* eggs in the Columbia River (McCabe and Beckman 1990; Parsley et al. 1993; McCabe and Tracy 1994) and rainbow (American) smelt *Osmerus mordax* eggs in Maine (Rothschild 1961). This method has proven useful in identifying spawning locations of both species. The artificial substrates used in the present study were based on the design of McCabe and Beckman (1990). Similar to white sturgeon eggs, eulachon eggs are demersal and highly adhesive. Eulachon eggs however are much smaller than white sturgeon eggs and prior to this study it was not clear if this would affect our results.

## METHODS

### *Study Area*

This study was conducted in the lower Columbia River, from RK 48 to RK 137 (near the mouth of the Lewis River; Figure 1). Several points throughout this area are being considered as potential channel deepening, or in-river dredge spoil disposal sites for the proposed USACE channel-deepening project (USACE 1999). A shipping channel is currently maintained throughout the length of the study area.

### *Artificial Substrate Construction*

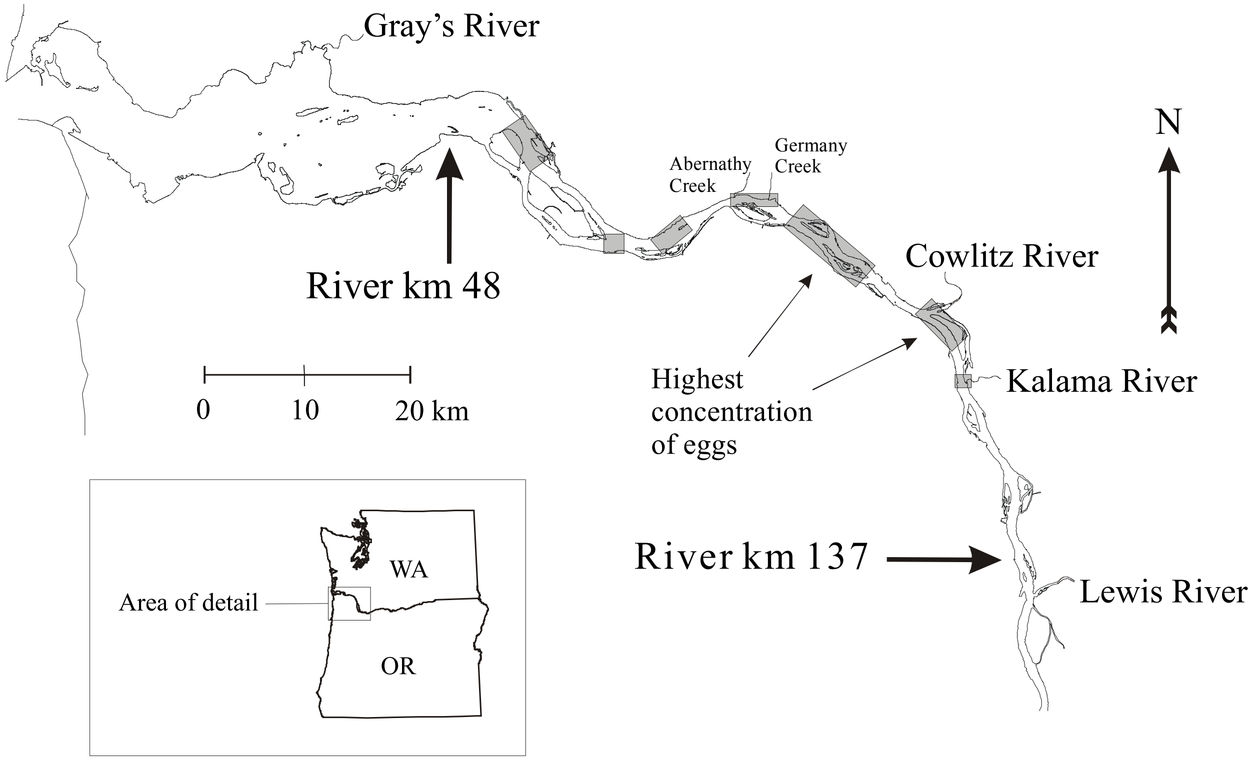


**Figure 1. Artificial-substrate sampling was conducted from RK 48 – 137 in the Columbia River. Shaded areas indicate sampling sites where eulachon eggs were successfully captured 26 February – 29 March 2001.**

The artificial substrates used in this study were constructed following the methods outlined by McCabe and Beckman (1990), with the exception of the substrate material available for eggs to adhere. The frames consisted of a 76-cm x 91-cm angle-iron outer-frame with strips of flat iron bar to provide support. Three strips of iron bar were welded into place on one side of the frame and three more were secured with nuts and bolts on the other side. Two 76-cm x 91-cm pieces of commercially available, low nap indoor-outdoor carpet material, placed back to back, were used as an egg adhesion surface. The carpet was secured in the frame by the two sets of flat iron bar. Securing the iron bar on one side of the frame with nuts and bolts facilitated easy removal of the carpet pieces. Although McCabe and Beckman (1990) utilized latex-coated animal hair material as an egg adhesion surface, we chose carpet material with a 2 to 4 mm nap depth because of concerns that the depth of animal hair would make it difficult to find the smaller eulachon eggs.

We used two types of anchors to secure substrates in place on the river bottom. A three fluke anchor constructed of steel bars (13 mm diameter), PVC pipe (8 cm diameter) and concrete, similar to those employed by McCabe and Beckman (1990) was used on approximately 25 % of the substrates. The remaining 75 % were secured with a 9-kg pyramid shaped lead weight. Buoy lines were connected to all substrates to mark the location. Length of line used depended on water depth and velocity. We generally used line equivalent to approximately 1.5 times the depth to account for tidal variation. The location of each substrate was recorded from a Global Positioning System (GPS) unit on board the deploy vessel. Upon retrieval, the location of the substrate was taken from the onboard GPS unit to determine if the substrate had moved from its original location.

### *Sampling Methods*

Sampling in the lower Columbia River with artificial substrates commenced on 26 February and lasted until 29 March 2001. We sampled from RK 48.3 to RK 136.8, with at least two artificial substrates placed every 1.6 RK (except between RK 59.6 and RK 61.2 within which only one substrate was placed). We made no attempt to standardize or stratify substrate placement among depths or habitat types. Depths of sampling ranged from 0.9 to 12.8 m and distance from the riverbank ranged from 5 m to greater than 90 m. Sampling at greater depths was problematic because substrates tended to get covered in silt when placed in deeper water. Generally, substrates placed deeper and farther from the riverbank tended to silt in the most, whereas substrates placed close to the riverbank in shallow water tended to silt in the least.

We partitioned sampling into three rounds. The first sampling round was conducted from 26 February to 28 February 2001. Sampling in this round was limited to river kilometers 80.5 – 83.7, and 7 substrates were set. Substrates were initially placed in locations believed to be eulachon spawning locations (Smith and Saalfeld 1955) to test artificial substrates as a viable means of catching eulachon eggs. The second sampling round was conducted from 8 March to 14 March 2001, and a total of 17 substrates were deployed. These were placed over a wider range of the river, from RK 86.9 – RK 109.4. During the final sampling round, which lasted from 19 March to 29 March 2001, artificial substrates were set from RK 48.3 – RK 136.8, to characterize spawning distribution over a larger area. At least two substrates were set every 1.6 km (except that only one substrate was placed between RK 59.6 and RK 61.2).

All substrates were left in the water for a minimum of 18 hours to ensure that sampling occurred throughout an entire tide cycle and during both day and night. Most substrates were allowed to fish for <24 hours; however one substrate was not retrieved on the first attempt and had to be retrieved at a lower tide. The substrate fished for 40 hours and was not included in our analysis. Two other substrates were lost through unknown causes.

We used a Ponar grab sampler to determine composition of bottom material at sites where eggs were caught. Bottom samples collected in the field were brought back to the lab for analysis. In the lab, samples were dried and then passed through a series of sieves to determine particle size. We used a modified Wentworth classification (Orth 1983) to classify particle size of bottom material. Some locations consisted of bottom material with a particle size that was too large to sample with the Ponar device. These locations were directly adjacent to rip rap riverbanks. Additionally, we were unable to sample the bottom material at three sites. River velocity was too strong at one site (mouth of the

Kalama River) to sample with the Ponar device. Two other sites (mouth of Abernathy Creek) were dry because of a decrease in river level when we returned to sample the bottom material.

## RESULTS

Only one of the seven substrates we deployed during the first sampling round caught eggs, for a success rate of 14.3%. The substrates fished for a total of 71.2 h (effort), and three eggs were caught, resulting in a catch per unit effort (CPUE) of 0.04 eggs/h. The average set depth was 7.0 m, and the average catch was 0.43 eggs/set.

Of the 17 substrates set during the second round of sampling, nine caught eggs, yielding a success rate of 52.9%. Effort totaled 449.3 h during this period for a CPUE of 0.17 eggs/h. The average set depth was 4.7 m, and the average catch was 1.88 eggs/set.

Thirteen of 123 substrates set during the final sampling round caught eggs for a success rate of 10.6%. Effort totaled 2,691 h during this period for a CPUE of 0.02 eggs/h. The average set depth was 7.4 m, and the average catch was 0.36 eggs/set.

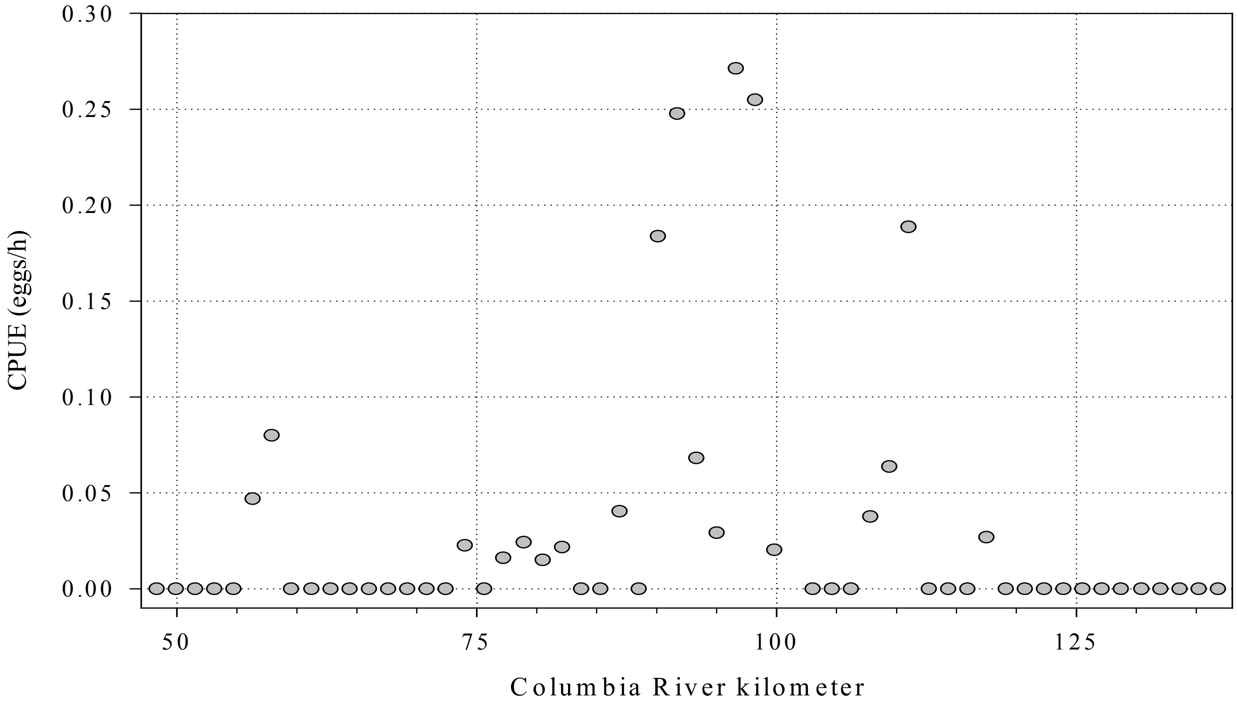
We set 147 substrates over the study, 23 of which successfully caught eulachon eggs for an overall success rate of 15.6%. A total of 122 eggs were caught for an average of 0.84 eggs/substrate. Egg catches in a single set ranged from zero to 27. Catch rates were highest between RK 90 and RK 98 and between RK 107 and RK 111 (Figure 2). Daily CPUE ranged from 0 eggs/h to 0.20 eggs/h (Figure 3). The greatest daily CPUE occurred on 9 March (0.20 eggs/h of effort) and 13 March (0.16 eggs/h of effort). Sampling days that occurred before 9 March and after 13 March averaged less than 0.08 eggs/h of effort. River bottom composition at sites where eggs were caught varied, yet was dominated by medium to fine sand (Table 1).

## DISCUSSION

Although we believe this is the first time artificial substrates have been used to collect eulachon eggs, artificial substrates have been used to collect eggs of the rainbow (American) smelt (Rothschild 1961). Similar to eulachon, rainbow smelt are broadcast spawners, laying highly adhesive eggs that readily become attached to stream substrates. Rothschild (1961) used 3.1-cm x 12.7-cm strips of heavy canvas as an egg-depositional surface, attached to a glazed black ceramic tile (11.4 cm x 11.4 cm). Since 1988, artificial substrates have been used in the Columbia River to collect white sturgeon eggs (McCabe and Beckman 1990; Parsley et al. 1993; McCabe and Tracy 1994). Although the design employed by Rothschild (1961) was effective at catching smelt eggs, we chose to follow the design of McCabe and Beckman (1990) because it had been successfully used in the high flows of the Columbia River.

We caught eulachon eggs over a larger area of the river (RK 56.3 to RK 117.5) than previously described as spawning habitat (Smith and Saalfield 1955). The area in which we caught the highest concentration of eggs (between RK 90.1 and RK 98.2) has not previously been documented as a spawning location.

**Figure 2. Egg catch/h for each river 1.6 kilometers sampled. Sampling was conducted between river kilometers 48 and 137.**



The strength of the eulachon spawning run in the lower Columbia River varies throughout the course of a single season; therefore, CPUE may vary spatially and temporally with the peak of spawning. This may explain why CPUE was highest on 9 and 13 March; however, this result must be interpreted cautiously because sampling on any one day was limited to a small area of the river (most days approximately 9.7 river kilometers were covered). Observed temporal differences in CPUE may be influenced by daily spatial differences in sampling.

Although catch varied among sample times and locations, we have shown that eulachon eggs can be caught with artificial substrates. This is important because the presence of viable eggs indicates that spawning is occurring in the vicinity. This is the first known study to show that eulachon spawning occurs at many points throughout the lower Columbia River, from RK 56.3 (Price Island) to RK 117.5 (mouth of the Kalama River). We were unable to use substrates within areas designated for channel deepening because of the dynamic nature of the bottom in these areas. In 2000 we attempted to fish several artificial substrates in areas proposed for deepening and the frames were quickly buried

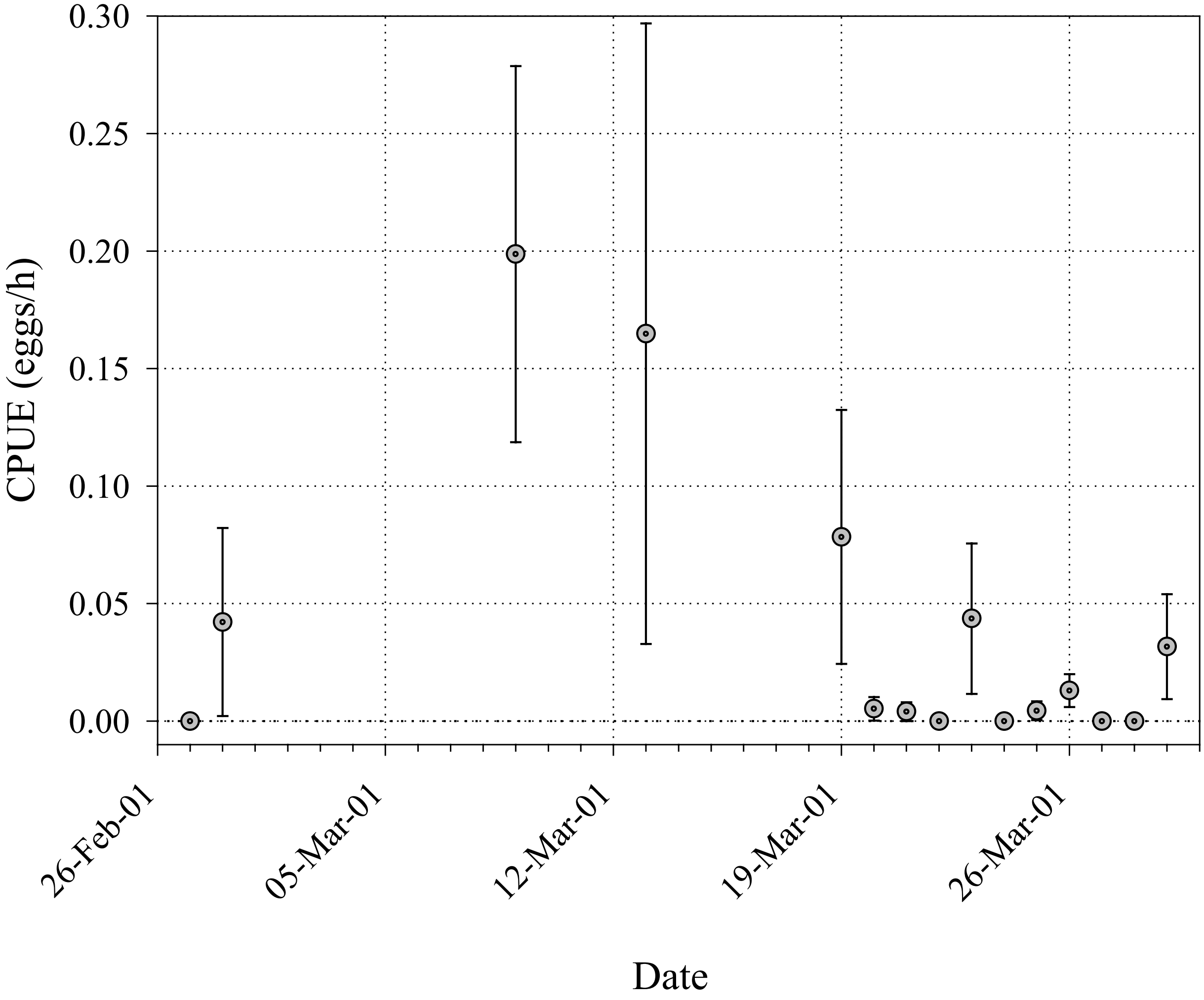


**Figure 3. Egg catch/h listed by date each artificial substrate was retrieved. No substrate frames were examined 1-8, 10-12, or 14-18 March 2001. Bbars represent one standard error.**

in sand, which made them ineffective and difficult to retrieve. Substrate movement is typical in the sand wave environment found on the bottom of the riverine areas proposed for channel deepening. Sand waves in this reach are generally large, with heights of 1.8-3.7 m and lengths up to 150-m (personal communication with Karl Eriksen, Portland District, USACE). Given the dynamic nature of channel substrates, we believe these areas do not provide stable surfaces that would allow an adhesive egg to incubate for 30 d. We considered that eggs incubating in near-shore areas in the proximity of dredging activities might be affected if these activities alter flow patterns or increase sedimentation; however, hydraulic models completed by the USACE indicate dredging will not alter the river's flow patterns. The USACE also expects the average annual bedload transport in the main river channel to remain within the existing range of 75,000-300,000 $m^3$/yr (USACE 1999). To reduce unforeseen impacts on eulachon eggs, channel-

**Table 1. Summary of artificial substrate data for sites where eggs were caught. RK = river kilometer. CPUE = eggs/h. Bottom composition based on a modified Wentworth classification.**

| RK | Date | Depth (m) | Eggs | CPUE | Bottom composition |
|---|---|---|---|---|---|
| 56.3 | 03/29/01 | 11.6 | 3 | 0.14 | Large particles associated with rip rap |
| 57.9 | 03/29/01 | 9.4 | 5 | 0.24 | Medium to fine sand |
| 74.0 | 03/26/01 | 8.5 | 1 | 0.05 | Coarse to medium sand with pebbles |
| 77.2 | 03/26/01 | 9.4 | 1 | 0.05 | Mixed cobble and pebbles |
| 78.9 | 03/26/01 | 9.1 | 1 | 0.05 | Medium to fine sand |
| 80.5 | 03/25/01 | 4.6 | 1 | 0.05 | Coarse to medium sand |
| 82.1 | 02/27/01 | 12.8 | 3 | 0.13 | Pebble |
| 86.9 | 03/08/01 | 1.8 | 1 | 0.03 | No sample taken |
| 86.9 | 03/08/01 | 1.8 | 3 | 0.10 | No sample taken |
| 90.1 | 03/08/01 | 3.0 | 18 | 0.62 | Cobble and pebble mix |
| 90.1 | 03/08/01 | 7.6 | 2 | 0.07 | Medium to fine sand |
| 91.7 | 03/19/01 | 9.8 | 11 | 0.49 | Medium to fine sand |
| 93.3 | 03/19/01 | 7.3 | 3 | 0.13 | Medium to fine sand |
| 93.3 | 03/08/01 | 4.6 | 2 | 0.07 | Large particles associated with rip rap |
| 95.0 | 03/19/01 | 8.5 | 2 | 0.09 | Medium to fine sand |
| 96.6 | 03/08/01 | 4.0 | 14 | 0.51 | Medium to fine sand |
| 98.2 | 03/12/01 | 0.9 | 3 | 0.13 | Medium to fine sand |
| 98.2 | 03/12/01 | 12.8 | 27 | 1.20 | Medium to fine sand |
| 99.8 | 03/21/01 | 4.0 | 1 | 0.04 | Medium to fine sand |
| 107.8 | 03/23/01 | 5.5 | 4 | 0.15 | Medium to fine sand |
| 109.4 | 03/08/01 | 2.7 | 5 | 0.19 | Medium to fine sand |
| 111.0 | 03/23/01 | 8.2 | 10 | 0.37 | Medium to fine sand |
| 117.5 | 03/20/01 | 9.8 | 1 | 0.05 | No sample taken |

deepening operations could be scheduled to avoid areas in which we caught the greatest number of eulachon eggs during the typical peak spawning period.

## ACKNOWLEDGMENTS

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# REPORT D

# CHARACTERIZATION OF DEVELOPMENT IN COLUMBIA RIVER PROLARVAL EULACHON *Thaleichthys pacificus* USING SELECTED MORPHOMETRIC CHARACTERS.

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Final Report to U. S. Army Corps of Engineers
Contract Number W66QKZ13237198

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ABSTRACT

**Objectives of this study were to artificially propagate eulachon larvae, establish a time/temperature dependent developmental baseline, and subsequently evaluate the efficacy of the methodology used. Adult brood fish were collected from the Cowlitz and Sandy rivers, transported to propagation facilities, held until ripe, and artificially spawned in the laboratory. Eggs were successfully incubated in water filled petrie dishes and hatched 47 days after fertilization, accumulating 752 thermal units – approximately twice that documented by previous workers. At the peak of hatching, larvae were transferred to a chiller and allowed to develop at 53 oF. Larvae were allowed to develop in petrie dishes and survived 21 days before total yolk-sac absorption. Larvae were preserved in 10% buffered formalin at post-hatch ages of 0, 6, 12, 24, 36 and 48 hours, and then subsequently at intervals of 24 hours until total yolk sac absorption. Total length, snout to vent length, and yolk sac length of preserved fish were measured to characterize larval development. Newly hatched larvae showed a marked curvature in the anterior portion of their bodies and gradually assumed a straighter form after approximately 72 hours. Coiling proved problematic for obtaining true measurements of total and snout to vent lengths. Yolk sac measurements were unaffected by coiling and showed a more linear trend over time. Results indicated that changes in larval morphology were not identifiable over a time scale of even a few days. Static environmental conditions in our experiment appear to have retarded larval development. Morphometric analyses of larvae collected in the field from known spawning areas showed that identifiable development does occur as larvae migrate to the estuary and ocean. Repetition of this experiment with propagation conducted under more natural conditions might lead to increased developmental rates allowing changes to be identified over short time periods; however, this may be precluded by the high morphological variability in individuals observed in each age class.**

## Introduction

Active spawning of eulachon *Thaleichthys pacificus* has been observed in various tributaries of the lower Columbia River, including the Cowlitz, Kalama, Lewis, and Sandy rivers (Smith and Saalfeld 1955). However, little is known about the spawning distribution of eulachon in the mainstem Columbia River.

Catches of very recently hatched eulachon larvae in plankton net hauls might indicate proximal spawning grounds and hence provide a useful tool for mapping and defining spawning habitat distribution in the mainstem Columbia River. To achieve this, developmental observations from larvae of known ages are required to provide a baseline against which larval assemblages collected in the field can be compared.

Upon hatching, eulachon larvae are incorporated into the drift and, depending on local current velocities, are presumably transported substantial distances from their spawning grounds in relatively short periods of time. For an aging methodology to be an effective tool in pinpointing spawning areas, short-term developmental changes measured on an hourly time scale must be identified.

Eulachon larvae have been successfully propagated by several workers for various studies that include spawning substrate preference (Wendler 1937), assessment of possible population heterogeneity in the lower Columbia River and its tributaries (Delacy and Batts 1963), and effects of temperature on incubation periods (Smith and Saalfeld 1955). Although Parente and Snyder (1970) provide a pictorial record and discussion of egg and (to a limited extent) larval development, a systematic, quantitative assessment of eulachon larval development has not been previously described as it has for related species such as the rainbow smelt *Osmerus mordax* (Cooper 1978). Given the lack of information regarding eulachon larval development, the objectives of this study were to artificially propagate larvae, establish a time/temperature dependent developmental baseline and subsequently evaluate the efficacy of the methodology used.

## Methods

### Artificial Propagation

**Adult brood fish were collected with dip-nets from the Cowlitz River (6 March 2001) and Sandy River (14 March 2001). Broodstock collected from the Cowlitz River (39 males and 42 females) were transported to facilities of the Oregon Department of Fish and Wildlife (ODFW) in Clackamas and held in large circulating tanks (males and females separated) until ripe. The fish matured rapidly in the holding tanks - probably a result of high water temperatures observed in the Clackamas spring water (57 $^o$F) relative to the Cowlitz River (43 $^o$F). Despite the separation of the sexes, all of the females extruded their eggs and subsequently died during the night of 11 March 2001. However, one female from this batch had been sacrificed for initial fertilization experiments on 8 March. Broodstock collected from the Sandy River (15 males and 12 females) were ripe upon collection and were artificially spawned on 14 and 15 March 2001.**

Eggs were manually stripped from females into glass Petrie dishes and covered with milt from ripe males. Water was added to activate the spermatozoa and thus initiate fertilization. The eggs/milt were gently stirred with the caudal fin of a eulachon (Wendler 1937) to ensure adequate mixing of sex products.

After a short period (minutes) the eggs were gently washed with fresh water and then transferred to two incubation environments consisting of 1) McDonalds jars supplied with water from a closed system cooled by a portable chiller unit to 48$^o$F, and 2) water-filled petrie dishes placed into a walk-in chiller to incubate at a constant temperature of 48 $^o$F. During the incubation period, water in the petrie dishes was changed daily and dead/fungused eggs were removed. All water used in the propagation experiments came from the local spring at the ODFW Clackamas complex.

**At the peak of hatching, larvae were transferred to water-filled Petrie dishes so that each dish contained larvae emergent within a 30-minute time period. Larvae were then transferred to the chiller and allowed to develop. Chiller temperature was adjusted to 53 $^{o}$F to reflect temperature of the Columbia River.**

**Larvae were preserved in 10% buffered formalin at post-hatch ages of 0, 6, 12, 24, 36 and 48 hours, then subsequently at intervals of 24 hours until total yolk sac absorption. Preservation of the 24-hour age class failed so no results from this time period are included.**

### Morphometric Characters

**Basic morphometric observations were chosen to characterize larval development. Morphometric measurements included total length, snout to vent length, and yolk sac length (Figure 1). Measurements were made with an ocular micrometer read to the nearest 0.1mm. Ten larvae per age class were examined.**

Measurements were also obtained from larvae taken in plankton net tows from the Cowlitz River and the lower Columbia River shipping channel in the vicinity of Price Island (river kilometer 55). Fifty individuals from each sample were randomly selected for characterization. These measurements were taken to compare development in larvae taken from a known spawning area (Cowlitz River) against those from a location in the study area assumed to be substantially downstream from major spawning areas.

## DATA ANALYSIS

Analysis of Variance procedures was used to test for significant differences in yolk sac length between age classes of propagated larvae. Tukey's multiple comparison procedure was used to isolate differences among groups. Linear regression analyses were conducted to define at-hatching values for each of the morphometric characters. T-tests were used to test for significant differences in morphometric characters between larvae taken from the Cowlitz River and the Columbia River. Tests were conducted at $\alpha = 0.05$ level of significance.

## RESULTS

**The majority of the fertilized eggs (approximately 60,000 eggs) were placed in the McDonalds jar system to incubate. No eggs survived after a day in this environment because of the failure of the chiller unit and stresses induced by turbulence on the eggs. Eggs incubated in the petrie dishes were initially subject to high mortality and fungusing as a result of overcrowding. We reduced egg densities and mortality was reduced.**

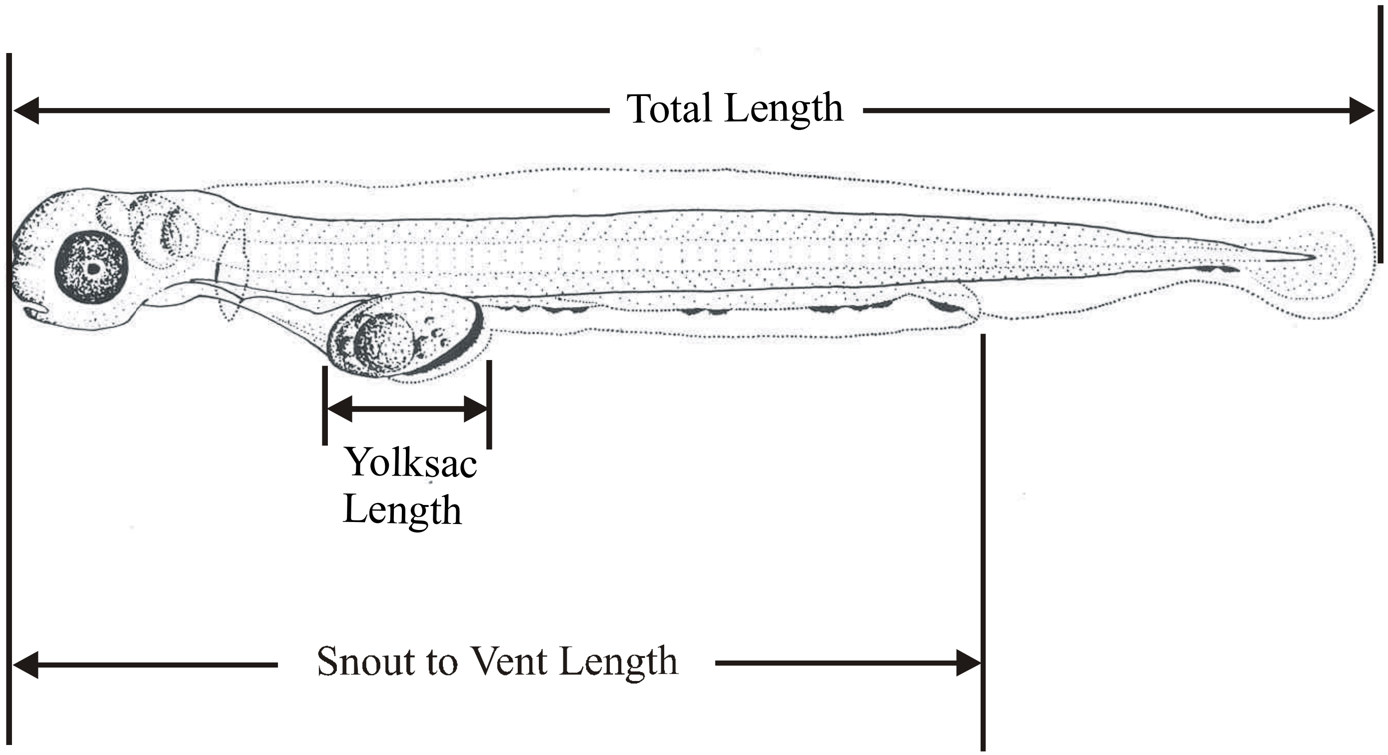


Figure 1. Morphometric characters used in eulachon larval development study.

Eggs fertilized on 8 March began hatching on 24 April and continued through 29 April. Eggs fertilized on 14 March began hatching 01 May and continued through 10 May. For each batch, first hatch occurred approximately 47 days after fertilization. Water
temperature throughout this period was a constant 48 °F. Using 32 °F as the assumed biological zero for eulachon (Delacy and Batts, 1963) the eggs accumulated 752 Thermal Units (TU's) before hatching.

Most larvae were observed to emerge tail first from their egg casing. Initial observations showed that the time between rupture of the egg membrane and full emergence of the larvae varied widely from a few minutes to several hours (see Parente and Snyder, 1970 for a pictorial record of a eulachon larva hatch sequence). Many individuals at this stage showed a marked curvature in the anterior portion of their bodies (probably a remnant of coiling in the egg) and gradually assumed a straighter form after approximately 72 hours (Figure 2). Coiling proved problematic for obtaining true measurements of total and snout to vent lengths. Data for these characters appears to show a relatively rapid rate of growth from hatching to approximately 72 hours, after which length increases were less pronounced (Figure 3). This is attributable to coiled larvae straightening out over time (Figure 2). Yolk sac measurements were unaffected by coiling and showed a more linear trend over time (Figure 3).

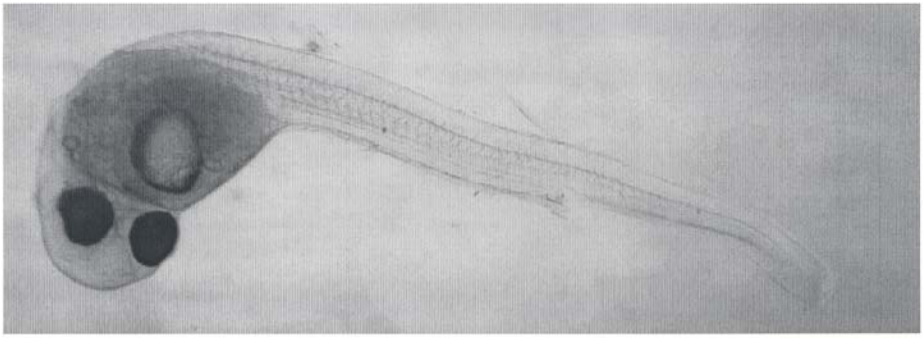
Larvae were strongly attracted to and stimulated by light sources. When placed in water-filled beakers most individuals exhibited pelagic swim-up behavior, remaining at the water surface for extended periods. Yolk absorption was complete 21 days after hatching and few individuals survived beyond this age.

Mean total length of larvae at hatching (0 hours) was 4.3 mm (+/- 0.51 SD). Mean snout-to-vent length at hatching was 3.0 mm (+/- 0.82 SD). Mean yolk sac length at hatching was 0.86 mm (+/- 0.14 SD). However, at hatching total length and snout-to-vent length means were assumed invalid because of imprecise measurements taken from coiled individuals. Linear regression analysis of total length and snout-to-vent length with 0, 6, 12 and 36-hour age class data removed gave a total length at-hatching value of 5.7mm ($R^2$=0.372, SE = 0.39) and snout to vent length at-hatching value of 4.1 mm ($R^2$=0.333, SE=0.38). Linear regression analysis of all yolk sac data gave an at-hatching length of 0.8 mm ($R^2$ = 0.746, SE = 0.118). Analysis of variance showed no statistically significant differences in mean yolk sac length between age classes 0, 6, 12, 36, 72 and 96 hours post-hatch (Tukey multiple comparison, $P > 0.05$).

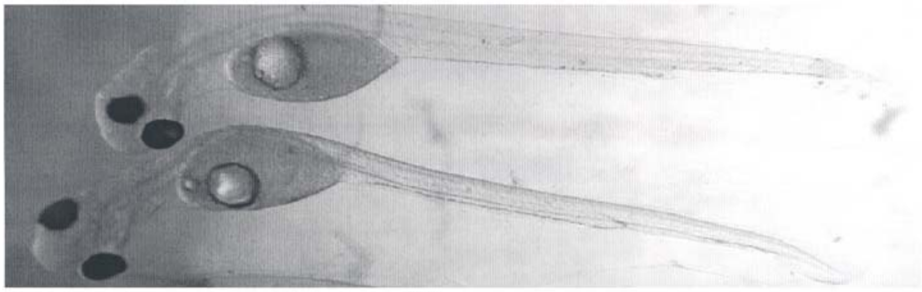
Significant differences between the mean values of each morphometric character were observed between larval assemblages collected in the Cowlitz River and in the Columbia River in the vicinity of Price Island (t-tests, $P < 0.001$ in all cases; Table 1 and Figure 4).

## DISCUSSION

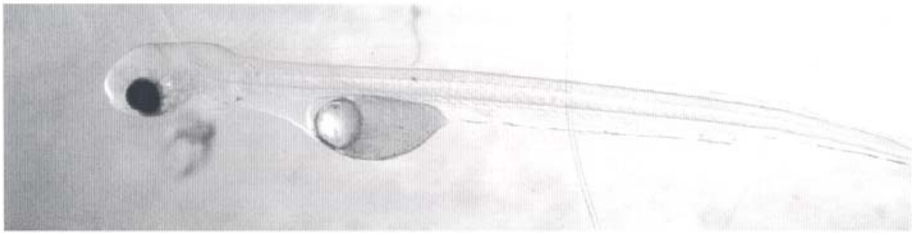
Our results show that changes in eulachon larval morphology are not identifiable over a time scale of even a few days. Since it is likely that currents in the Columbia River would carry larvae substantial distances (kilometers) in the matter of only a few hours it appears unlikely that spawning areas could be located using our proposed methodology. However, morphometric data from larvae collected from the Columbia and Cowlitz rivers



0 Hours post hatch.



36 hours post hatch.



72 hours post hatch.

Figure 2. Diminishment of remnant egg coiling over time in artificially propagated larval eulachon.

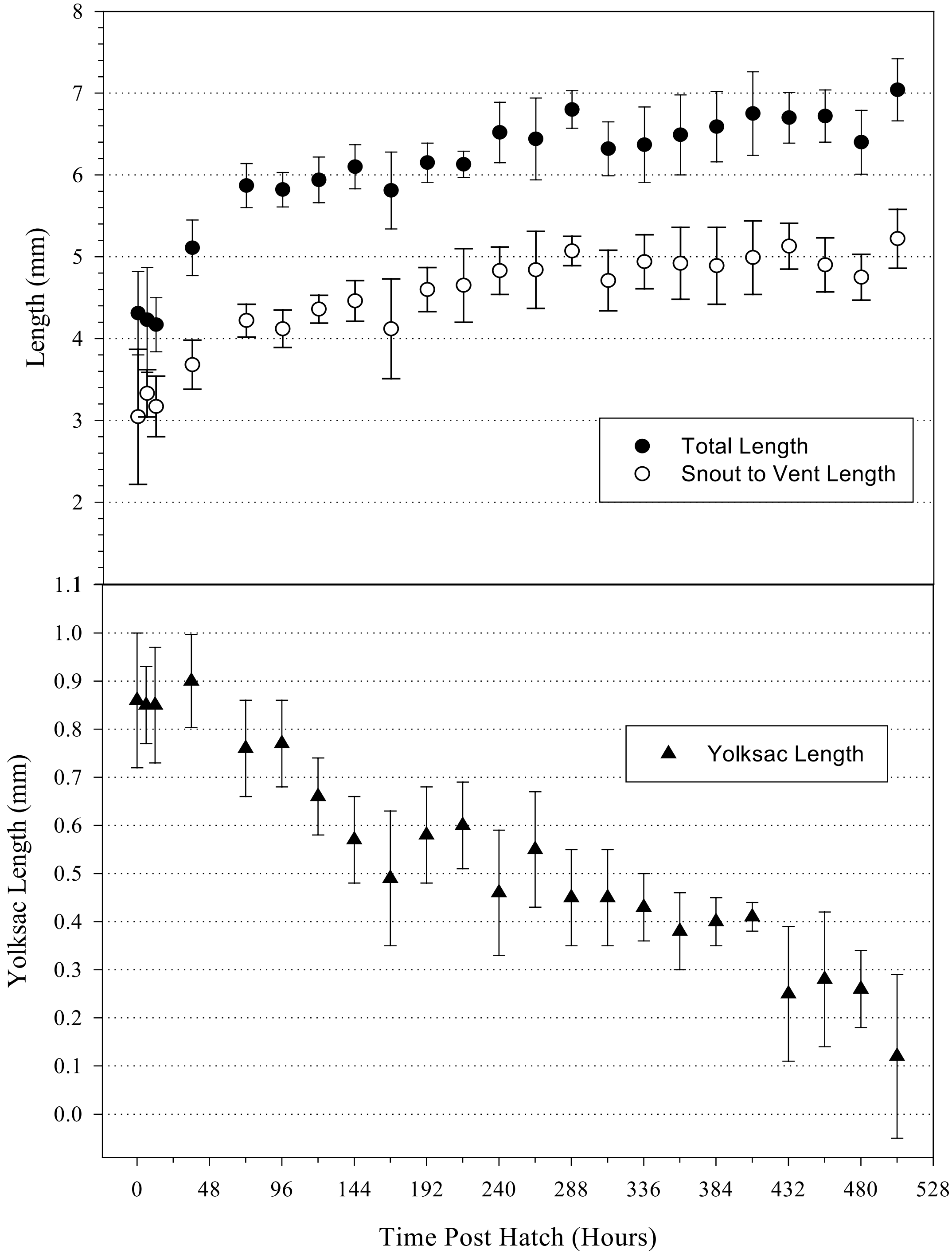


Figure 3. Morphological changes in artificially propagated eulachon larvae over time. Individual plots are means with error bars representing 1 standard deviation.

Table 1. Mean (standard deviation) morphometric measurements (mm) of

larval eulachon taken from the Cowlitz and Columbia rivers in April 2001.

| Origin | Number examined | Total length | Snout to vent length | Yolk sac length |
|---|---|---|---|---|
| Cowlitz River | 50 | 5.5 (0.5) | 4.0 (0.4) | 0.9 (0.1) |
| Columbia River | 50 | 6.1 (0.4) | 4.5 (0.3) | 0.5 (0.1) |

suggest that identifiable development does occur as larvae migrate to the estuary and ocean (Figure 4, Table 1).

In this study eggs accumulated 752 TU's before hatching – a figure markedly greater than that observed in previous investigations. Smith and Saalfeld (1955) reported TU's of 378 & 369.6 from hatchery experiments in 1946 (Kalama River Hatchery) and 1949 (University of Washington School of Fisheries), whereas Delacy and Batts (1963) found a range of 349.7 to 387.9 TU's in their investigations. Wendler (1937) reported larvae hatched 24 days after fertilization at a mean water temperature of 40.7 $^{o}$F during incubation - translating to approximately 209 TU's. Adult eulachon first entered the Cowlitz River in the first week of March 2001 (personal observation) and plankton net sampling by staff of Washington Department of Fish and Wildlife showed larvae were present in substantial numbers during the last week of March. Water temperature in the Cowlitz during this time was around 48$^{o}$F. This leads to a very rough estimate of 336 TU's for egg incubation in the Cowlitz this year (21 days at 48$^{o}$F), a figure close to those reported in previous experiments.

**It is unclear why the incubation period was so protracted in our study although eggs were incubated in the absence of light, continuous water exchange, and temperature fluctuations. This provided relatively static environmental conditions compared to those in all previous studies and under natural conditions. Environmental conditions in our experiment may also have protracted the development of larvae. In Parente and Snyder (1970) a photograph of a six-day-old eulachon larva shows almost complete utilization of yolk sac contents – a stage reached after almost 21 days for larvae in our experiment.**

**The remnant coiling observed in many hatchlings in our experiment could be considered a useful qualitative descriptor of larval age. However it was not seen in any larvae collected from the Cowlitz River despite the close proximity of the sampling location to spawning areas. This suggests the characteristic was also an artifact of our experimental conditions.**

**Repetition of this experiment with propagation conducted under more natural conditions might lead to increased developmental rates allowing changes to be identified over short time periods. However, the high morphological variability in individuals observed in each age class in our study (Figure 3) as well as those larvae taken from the Cowlitz River (a site where all individuals are presumably of very similar ages; Figure 4) might still preclude this.**

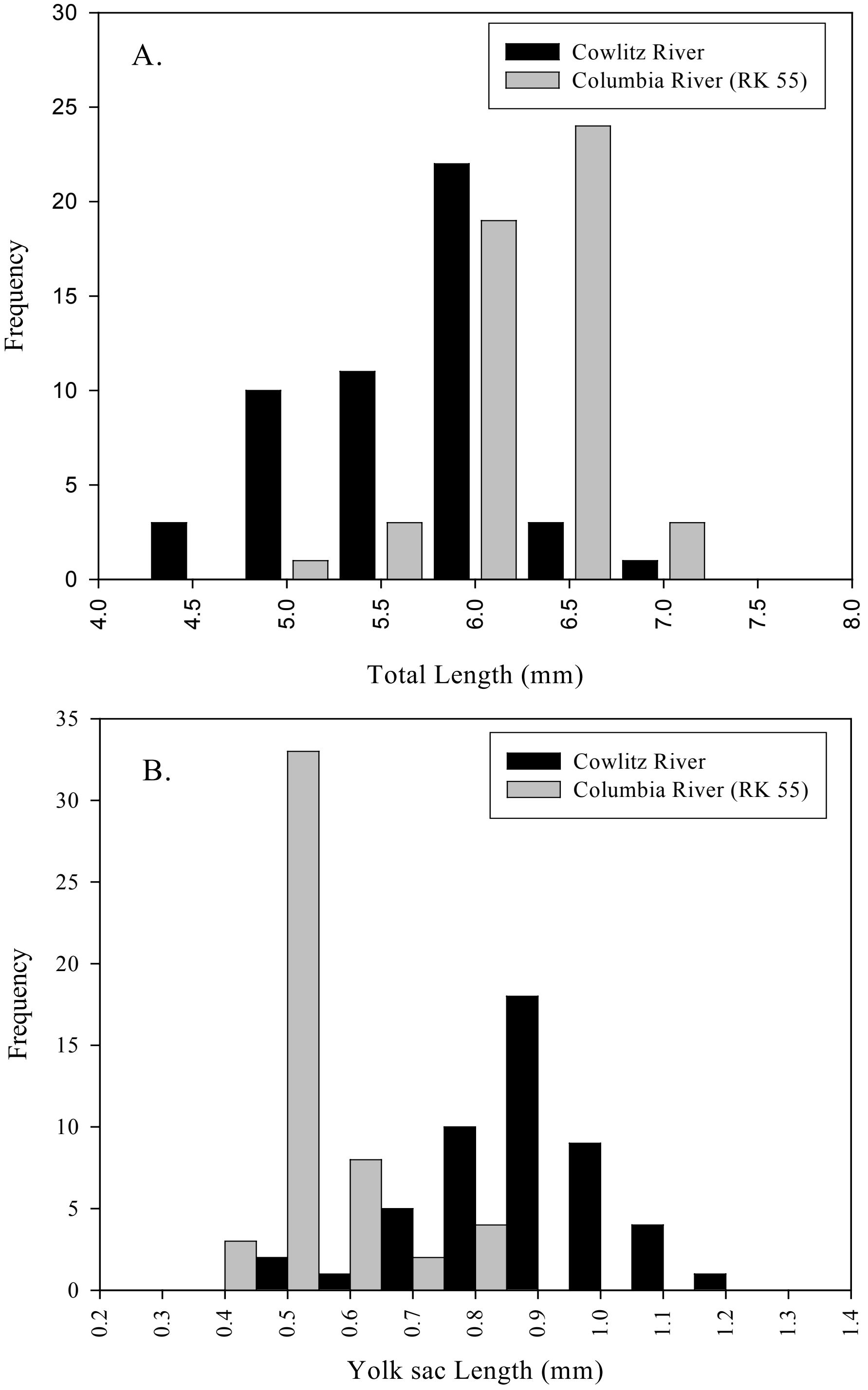


Figure 4. Length frequency distributions for A) total length and B) yolksac length in prolarval eulachon taken in plankton net tows in the Cowlitz and Columbia Rivers during April, 2001. N = 50 larvae for each location.

## ACKNOWLEDGMENTS

We thank Peter Barber, Jody Gabriel, Robin Mills, and Steve West for assistance in laboratory analysis. John DeVore (WDFW) helped with initial study conception. Brad James (WDFW), Dave Ward (ODFW) and Tom Rien (ODFW) reviewed a draft of this manuscript. Kim Larson (USACE) provided assistance as project manager. This work was funded by the U. S. Army Corps of Engineers (contracts W66QKZ 10745277 and W66QKZ13237198).

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