Chapter

Aquatic Ecology of the Elwha River Estuary Prior to Dam Removal

By Jeffrey J. Duda, Matthew M. Beirne, Kimberly Larsen, Dwight Barry, Karl Stenberg, and Michael L. McHenry

Abstract

The removal of two long-standing dams on the Elwha River in Washington State will initiate a suite of biological and physical changes to the estuary at the river mouth. Estuaries represent a transition between freshwater and saltwater, have unique assemblages of plants and animals, and are a critical habitat for some salmon species as they migrate to the ocean. This chapter summarizes a number of studies in the Elwha River estuary, and focuses on physical and biological aspects of the ecosystem that are expected to change following dam removal. Included are data sets that summarize (1) water chemistry samples collected over a 16 month period; (2) beach seining activities targeted toward describing the fish assemblage of the estuary and migratory patterns of juvenile salmon; (3) descriptions of the aquatic and terrestrial invertebrate communities in the estuary, which represent an important food source for juvenile fish and are important water quality indicators; and (4) the diet and growth patterns of juvenile Chinook salmon in the lower Elwha River and estuary. These data represent baseline conditions of the ecosystem after nearly a century of changes due to the dams and will be useful in monitoring the changes to the river and estuary following dam removal.

Introduction

As juvenile salmon migrate from freshwater rearing areas, they undergo the physiological, behavioral, and life history changes necessary for transition to life in the ocean. Estuaries are recognized as an important part of this migration (Simenstad and others, 1982; Bottom and others, 2005b). Some salmon species, such as Chinook (Oncorhynchus tshawytscha) and chum (O. keta) can spend extended periods (weeks to months) in the estuarine environment, whereas others such as pink salmon (O. gorbuscha) largely forego extended use of estuarine areas in favor of ocean rearing (Groot and Margolis, 1991). It is hypothesized that salmon use estuarine habitats because they provide a higher growth potential, facilitate the physiological transition to saltwater conditions, and reduce the risk of predation (Quinn, 2005). All of these factors may play a role in higher growth and survival during the subsequent marine life history phase (Ruggerone and others, 2009). Estuarine fish assemblages fluctuate seasonally, based on migration timing, habitat structure and connectivity, and physical factors such as salinity and temperature. Estuaries vary in size (see Duda and others, 2011, figs. 1.8-1.10, chapter 1, this report) and in the relative influence

of freshwater and marine drivers important for ecological processes. This affects the amount, complexity, biological productivity, and physical conditions of estuarine habitats, which varies among river systems. Although the role of estuaries has been deemed important for some Puget Sound Chinook populations (Simenstad and others, 1982; Beamer and Larsen, 2004), the significance of the Elwha River estuary and nearshore (coastal marine water zone extending from high tide elevation to the limit of the photic zone) to Chinook salmon populations is relatively unknown. We set out to describe the existing conditions of some key ecological components of the lower Elwha River and its estuary prior to dam removal.

Research efforts focused on collecting physical (water chemistry; see also Magirl and others, 2011, chapter 3, this report, for salinity and temperature data) and biological data from throughout the estuary and adjacent riverine and nearshore areas in the Elwha River study area (fig. 7.1) prior to dam removal. Water samples were collected monthly to measure nutrient concentrations from stations dispersed in the lower river (2 sites), in the estuary (4 sites), and in the nearshore (1 site). Seasonal use of the estuary and nearshore by salmon during their migration to the marine environment also was estimated. We studied fish distribution, abundance, and migration timing for all salmonids using the Elwha River estuary and examined the diet and growth rates of juvenile Chinook salmon. The aquatic and terrestrial macroinvertebrate communities of the estuary also were surveyed. These animals play a central role as prey for salmon and are indicators of water quality that should be responsive to physical changes associated with dam removal. Other studies are being conducted on fish ecology by colleagues elsewhere in the watershed (for examples, see Burke and others, 2008; Connolly and Brenkman, 2008; McHenry and Pess, 2008; Pess and others, 2008, Brenkman and others, 2008a, b; Duda and others, in press), in the nearshore proximal to the Elwha River, and in the Strait of Juan de Fuca (for example, Shaffer and others, 2009). Juvenile Chinook salmon samples from these studies were obtained to assist in estimating several life history traits, including age and growth (otolith microstructure), feeding (stomach contents), and genetics (fin clips). This information should help provide a better understanding of Chinook salmon life histories and help inform adaptive management of the population following dam removal and recolonization of the Elwha River watershed.



Figure 7.1. Sampling locations where water nutrients, benthic and terrestrial insects, and fish samples were collected in the Elwha River, Washington, study area.

Water Nutrients

Inorganic nutrients are important constituents for cell growth and often limit population growth and drive competitive interactions; therefore, they play a central role in determining presence and abundance patterns of biota. The availability of essential nutrients in the water column can limit the amount of biological productivity and thus the character of the entire ecosystem. Measurement of nutrients through time also can reflect patterns in seasonal biological productivity, as nutrients are diminished during periods of high growth (for example of algae and other primary producers during the summer growing season) and are higher during periods of biological dormancy. The availability of nutrients in any watershed depends on both biological and physical properties. Natural forces such as rainfall, geology, atmospheric deposition, and sedimentation drive levels of available nutrients, which are then consumed by autotrophic organisms and further modified by allochthonous energy inputs from the surrounding landscape. Other factors largely caused by human activity, such as resource extraction, fertilization, and industrial pollution also can affect the nutrient levels in a watershed.

Previous water nutrient sampling of main stem, side channel, and tributary habitats throughout the Elwha River watershed has indicated that, for its size and condition, the Elwha River is oligotrophic, or low in nutrients (Munn and others, 1999; Duda and others, in press). Although these earlier studies had broad spatial coverage of the watershed, they were temporally restricted to base (summer) flows and did not include water samples from the estuary or nearshore. This chapter provides comparable nutrient samples from the estuary and nearshore and a time series over a 16-month period. This baseline assessment of spatial and temporal trends in nutrient levels of the water column will be useful for comparisons during and following dam removal.

Water Sample Collection and Laboratory Analysis of Nutrient Concentrations

Water samples were collected monthly from seven locations in the lower Elwha River and estuary (fig. 7.1). Sites included one main stem (Lower Elwha River – main stem) and one side channel (Lower Elwha River – side channel) location in the lower Elwha River; the groundwater fed outflow of the tribal hatchery (Bosco Creek) that serves as a primary source of surface water to the east estuary; the tidally influenced lentic sloughs east of the river mouth in the east estuary (ES1 and ES2); the disconnected lake to the west of the river mouth (Dudley Pond); and the surf zone (Nearshore - Strait of Juan de Fuca) 25 m east of the river mouth in the Strait of Juan de Fuca. Samples were collected across a range of tidal conditions at approximately 30-day intervals, with interruptions in sample collection at all sites in November 2006 (inclement weather) and July 2007 (processing error).

Water samples were collected for analysis of total nitrogen (TN), total phosphorous (TP), dissolved inorganic nitrogen (nitrate [NO₃⁻], ammonia $[NH_4^+]$) and phosphate (PO_4^{-3}) . Water collection (500 mL) and sample bottles (60 mL) were acid washed (10 percent HCl) prior to sample collection. At each site, all sampling equipment and bottles were rinsed three times with ambient water. Samples were collected at the same depth for each site, generally 60 percent of the water column depth at least 1 m from shore. An inverted water collection bottle was submerged, filled, and capped at depth, to ensure that no water was collected from the surface. Samples for dissolved nutrients were filtered in the field using a 0.45-µm pore size syringe filter. Once collected, samples were kept on ice in the field and frozen in the laboratory prior to analysis by the University of Washington Marine

Chemistry Laboratory, Seattle, Wash. This procedure and laboratory were the same that were used by Morley and others (2008) and Duda and others (in press) for samples collected throughout the Elwha River watershed.

Total nitrogen and total phosphorous were analyzed using the persulfate digestion method of Valderrama (1981). The procedure of Armstrong and others (1967) was modified for the analysis of NO₃ and NO2. Water samples were passed through a cadmium (Cd) column where the nitrate (NO_2) was reduced to nitrite (NO₂). This NO₂ was then diazotized with sulfanilamide and coupled with N-(1-naphthyl)-ethylenediamine to form an azo dye. The sample was then passed through a 15 mm flowcell and absorbance measured at 540 nm. The procedure is the same for the NO_2 analysis less the Cd column. Nitrate concentration equals the $(NO_3 +$ NO_2) concentration minus the NO_2 concentration. A modification of the Slawyk and MacIsaac (1972) procedure was used for the analysis of ammonium (NH_{4}) . Water samples were treated with phenol and alkaline hypochlorite in the presence of ammonia (NH₂) to form idophenol blue (Berthelot reaction). Sodium nitroferricyanide was used as a catalyst in the reaction. Precipitation of calcium (Ca) and magnesium (Mg) hydroxides was eliminated by the addition of sodium citrate complexing reagent. The sample stream then was passed through a 55 °C heating bath, then through a 50 mm flowcell and absorbance was measured at 640 nm. Phosphate was analyzed using a modification of the Bernhardt and Wilhelms (1967) method. Ammonium molybdate was added to water samples to produce phosphomolybdic acid, which then was reduced to phosphomolybdous acid following the addition of dihydrazine (or hydrazine) sulfate. The sample was passed through a 50 mm flowcell and absorbance was measured at 820 nm.

Spatial Patterns of Water Nutrients

Fourteen water samples were collected from June 2006 until September 2007 at most locations (table 7.1). The concentration of each nutrient by month was graphed and smoothed curves were created using a cubic spline interpolation in SPSS Inc. (2010). The data are presented in the form of sparklines, a method of data visualization that uses small, highresolution graphics coupled with words and numbers (Tufte, 2006). By creating small, time series graphics for each location and water chemistry constituent (with maximum, average, and minimum values provided), spatial and temporal trends in water chemistry can be compared in a single figure.

The time series showed seasonal variations in nutrient levels and differences among sites (figs. 7.2A and 7.2B). Concentrations of nutrients (TN, TP, PO_4 and NO_3), at the nearshore zone of the Strait of Juan de Fuca generally were higher than sites in the Elwha

River and its estuary. An exception was Bosco Creek with the highest NH₄ concentration, possibly due to the hatchery operations at the source of the surface water supply, which concentrates the animal waste byproducts associated with large-scale fish production. Higher concentrations of TP, TN, NO₃, and PO₄ were measured in Bosco Creek than in the other five sites of the lower Elwha River and estuary. Following Bosco Creek and the nearshore, the two locations in the east estuary (ES1 and ES2) had the next highest levels of nutrient concentrations. Samples from the Elwha River (main stem and side channel) were consistently the lowest of all sites for most nutrients. This result is consistent with previous studies documenting the oligotrophic nutrient status of the river (Munn and others, 1999; Duda and others, in press). Levels of nitrogen and phosphorous generally increased along the gradient from freshwater-estuarine-marine waters, which is a result consistent with expectations.

Temporal Patterns of Water Nutrients

The highest temporal variability in water nutrient patterns was in the Strait of Juan de Fuca and Bosco Creek, which was indicated by the widest confidence intervals around their mean values across all samples (table 7.1). Oscillations at both these sites were during months that did not follow a consistent seasonal pattern (figs. 7.2A and 7.2B). The estuarine and lower Elwha River samples, however, displayed a more typical seasonal pattern. During spring and summer, when primary production in the river and estuary increases with increasing day length, levels of most nutrients were lower than levels during winter, when primary productivity wanes. Interestingly, Dudley Pond in the western part of the estuary, which is disconnected from the Elwha River by a dike, was out of phase with the east estuary during the autumn and winter periods, especially for TP, PO₄, and NH₄.

Table 7.1. Average concentration of total and dissolved nutrients at seven sites in the lower Elwha River, its estuary, and the nearshore surf zone of the Strait of Juan de Fuca, Washington, 2006 and 2007.

[All values are in micrograms per liter (standard deviation). Locations are shown in figure 7.1. Samples were collected at approximately 30 day intervals from June 2006 to September 2007, with interruptions in sample collection at all sites in November 2006 and July 2007. **Abbreviations:** MS, main stem; SC, side channel; ES1, east estuary 1; ES2, east estuary 2]

Location	Total phosphorus	Total nitrogen	Dissolved phosphate	Dissolved nitrate	Dissolved nitrite	Dissolved ammonia
Lower Elwha – MS	13 (4)	83 (48)	2 (2)	16 (17)	0.2 (0.1)	2 (3)
Lower Elwha – SC	12 (4)	105 (60)	2 (2)	24 (17)	0.2 (0.2)	4 (5)
Bosco Creek	32 (12)	275 (81)	14 (8)	76 (54)	3 (2)	79 (51)
ES1	16 (6)	139 (73)	5 (5)	28 (35)	0.2 (0.2)	13 (12)
ES2	18 (3)	178 (87)	6 (3)	43 (55)	0.6 (0.7)	18 (18)
Dudley Pond	16 (4)	163 (91)	3 (2)	7 (7)	0.3 (0.2)	10 (11)
Nearshore	49 (18)	284 (123)	30 (16)	132 (82)	2 (2)	12 (12)



Figure 7.2. Sparkline graphs showing time series of water chemistry species (*A*) total phosphorous and total nitrogen and (*B*) phosphate, nitrate, and ammonia, from the lower Elwha River, its estuary complex, a spring fed tributary draining into the east estuary, and the nearshore surf zone of the Strait of Juan de Fuca near the mouth of the Elwha River, Washington, 2006–07.



Figure 7.2—Continued

Nutrients and Dam Removal

The Elwha river is oligotrophic (Munn and others, 1999; Duda and others, in press), as measured values of nitrogen and phosphorous throughout the watershed are at low levels that are likely contributing to relatively low levels of primary productivity (Morley and others, 2008). This assessment, based on synoptic measurements taken during summer low flows, probably is affecting the nutrient status of the estuary. Results from the 2 freshwater Elwha River sites were similar to those reported by Duda and others (in press) for 12 other lower Elwha River sites during summer low flows (from 2005 and 2006). In that study, sites in the regulated sections of the Elwha River (downstream of the Glines Canyon and Elwha Dams) were significantly different from the upper unregulated section for phosphate, but not dissolved inorganic nitrogen. Duda and others (in press, fig. 5) reported that phosphate and dissolved inorganic nitrogen values in regulated sections of the Elwha River were significantly lower than in other unregulated rivers of the Pacific Northwest with salmon runs. However, they could not explain the significant differences in phosphate between the regulated and unregulated sections of the Elwha River.

How the levels of these nutrients in the lower Elwha River and its estuary will respond after dam removal is not known. Significant changes are expected in sediment supply, bed particle size, and nutrients supplied by salmon, all of which could affect the nutrient status of the sites. The supply of sediment is expected to increase dramatically in some places during and following dam removal (Czuba and others, 2011, chapter 2, this report; Konrad, 2009). Immediately following dam removal, fine-grained sediments stored in the reservoir bottoms and the deltas will be released and transported downstream.

Previous research has indicated that sediments released from reservoirs can change the nutrient status downstream (Stanley and Doyle, 2002). For example, phosphorous often is retained in reservoir sediments and the release of these sediments during dam removal could increase nutrient levels downstream during the short term. In the longer term, the reservoirs will not trap phosphorous, which will be transported to downstream reaches. The net effect of this material on nutrient levels after dam removal when sediment is transported to the lower Elwha River, estuary, and nearshore, is not clear. Based on an analysis of Lake Mills reservoir and delta sediments, Cavaliere (2010, p. 54) reported that, "The Elwha River sediments from Lake Mills have limited P (phosphorous) available for algal and plant growth. The water quality after the dams are removed is unlikely to be harmed by the release of excess P (phosphorous)..." from reservoir sediments. The low levels of nutrients held in reservoir sediments coupled with high rates of suspended sediment transport could limit any release of stored nutrients to the lower Elwha River and estuary.

If following dam removal and recolonization of upper parts of the watershed causes salmon to return to the river in larger numbers than the current populations, as projections suggest, then at least during some times of the year the nutrient status of the river may change. Based on estimates of future salmon returns following full recovery, 1,275–10,900 kg of nitrogen and 210–1,350 kg of phosphorous derived from salmon could be input annually into the Elwha River (estimates based on assumptions of Munn and others [1999] and projected spawners presented by Ward and others [2008]). If phosphorous and nitrogen are limiting primary productivity in the Elwha River, then increases in these nutrients may have important implications for the Elwha River ecosystem. It is not clear that an increase in salmon numbers will result in clear increases in water column nutrients, but salmon-derived nutrients may provide significant temporal increases in the biomass and growth rates of resident biota (for example, Bilby and others, 1996; Schuldt and Hershy, 1995; Wipfli and others, 1998; Chaloner and Wipfli, 2002; Duda and others, in press).

Aquatic and Terrestrial Macroinvertebrate Assemblages of the Elwha River Estuary

The current configuration of the Elwha River estuary has been relatively static for much of the period since dam construction, especially in recent decades (see Warrick and others, 2011, chapter 3, this report). The sediment deficit and related effects of the dams that led to the simplified, incised channel of the lower river (Pohl, 2004; Draut and others, 2008, 2011; Kloehn and others, 2008) also has contributed to the stability of the estuarine complex. This is most notable in the age and size classes of woody vegetation on the east estuary, particularly red alder and red elderberry (see Shafroth and others, 2011, chapter 8, this report). Effects to the estuary following dam removal will include sediment deposition, a likely associated increase in channel formation, and perhaps over the long term the development of multiple river mouths. This could result in dramatic changes to plant community associations and age class distributions, which can affect macroinvertebrate community assemblages.

The estuary is an important habitat for some juvenile salmon during their migration to the ocean in large part because of the food supply available, especially aquatic and terrestrial invertebrates (Simenstad and others, 1982). The Lower Elwha Klallam Tribe initiated a baseline assessment of macroinvertebrate assemblages in the estuary, including benthic macroinvertebrates and terrestrial insects in the emergent and shrub transitionhabitat zones. Describing the current conditions of the macroinvertebrate communities is an important monitoring goal, for their sensitivity to environmental changes and importance as a prey source for juvenile salmon.

Benthic Macroinvertebrates of the Elwha River Estuary

Sampling and Analysis Methods for Benthic Macroinvertebrates

Samples of benthic macroinvertebrates residing in the sediments throughout the Elwha River estuary were collected to establish baseline data on species diversity, relative abundance, and availability to juvenile salmonids throughout their migration season (spring and summer). Sampling occurred on three occasions (May, July, and September) in 2007. During each occasion, 22 samples were collected from sites in the east and west estuary. This sampling was near our juvenile salmon beach seining sites in the estuary. The 22 samples were distributed among the east estuary (ES1 [3 locations], ES2 [8 locations], the channel connecting ES1 and ES2 [2 locations; hereafter referred to as the intraestuarine channel [IEC]), and the west estuary (Dudley Pond [4 locations] and WESC [5 locations]) (fig. 7.1).

A spring-loaded 6-in petite Ponar grab sampler was attached to a nylon rope and deployed while wading (where depth allowed) or from a boat (fig. 7.3).



Figure 7.3. Lower Elwha Klallam Tribe fisheries technicians sampling sediments in the Elwha River, Washington, estuary using a petite Ponar grab sampler. (Photograph taken by Matt Beirne, Lower Elwha Klallam Tribe, April 17, 2006.)

The grab sampler surface area and volume were 0.023 m^2 and 2.4 L, respectively. Where the substrates were not conducive to acquiring an adequate sample volume, the grab sampler had to be manually forced into the substrate. Sediment grabs were weighed, photographed, and characterized by color and texture.

Sediment grab samples were sorted using a series of four metal sieves with successive openings of 3.55, 2.0, 1, and 0.5 mm (500 μ m). A garden hose with an adjustable nozzle was used to separate the inorganic sediment fraction gently from the organic material, detritus, and macroinvertebrates. The material remaining in the bucket was then sieved through a custom 0.5 mm stainless steel sieve fashioned from a plastic tray with the bottom replaced by a welded sheet of 500 μ m mesh. The primarily organic material that was retained in the sieves was gently separated using water sprayed into clear plastic tubs 32.5 cm \times 75 cm for subsequent sorting.

Immediately after sieving, the remaining material was sorted using forceps, typically within 24 hours of sample collection. A squirt bottle with 70 percent ethanol was used to agitate any remaining organisms from the organic material after all macroinvertebrates were removed. Sample processing time typically averaged 2 hours per sample, including sieving and sorting. Sorted samples were preserved in 10 percent formalin, which served as a fixative for soft-bodied organisms. After 1 week, specimens were filtered, rinsed, and transferred to 70 percent ethanol. Most insects from the orders Ephemeroptera, Plecoptera, and Trichoptera were identified to genus. All other taxa were identified to family level where possible, or the next identifiable taxonomic level (for example, class, order). Arthur Frost, a local expert in macroinvertebrate identification, completed all taxonomic identification.

Sediment grab samples also were used to provide general characterizations of epibenthic/ benthic habitat types in the estuary. Eight classes of sediment descriptions were used to characterize sediment (table 7.2). The greatest homogeneity was in WESC and was exclusively comprised of silty material typically overlayed with a detritus layer of leaf litter and some marine derived algae. The isolated estuary remnant of the west estuary (Dudley Pond) showed the greatest variability ranging from fine silt to gravel and was typically associated with a green, gelatinous organic fraction, likely derived from the decomposition of the heavy green algal mats that form on the surface of this water body (Duda and others, 2011, chapter 1, fig. 1.12, this report). The east estuary tended to lack the detrital/algal epibenthic component detected in the west estuary and was variable in texture from fine silt to sandy gravel. It was predominantly (66 percent) comprised of silt, with the remainder ranging from silty sand to siltysandy gravel. The west estuary, which is surrounded by dense, early successional willow/alder vegetation, seems to receive considerably greater detrital inputs and may not receive the

Table 7.2.Classification of sediment type from benthicmacroinvertebrate samples collected in the Elwha River estuary,Washington, May–September 2007.

[All values are number of grab samples. Abbreviation: WESC, west estuary channel]

Sediment category	East estuary	WESC	Dudley Pond
Silt	26	12	1
Silty-sand	2	0	0
Sandy-silt	6	0	1
Sandy	2	0	0
Silty sand-gravel	1	0	0
Sandy gravel	1	0	3
Silty organic	0	3	5
Organic gelatinous	0	0	2

same intensity or frequency of tidal flushing as in the east estuary habitats. The east estuary is significantly larger than WESC and appears to have a broader zone of emergent vegetation and shrub transitional habitat, coupled with greater tidal surges from the west and flushing from the east (Bosco Creek). This likely contributes to the small amount of detritus in the epibenthic environment.

Benthic Macroinvertebrate Assemblages of the Elwha River Estuary

During all months, we collected 40 macroinvertebrate taxa (5,943 individuals) in the east estuary, 29 taxa (3,343 individuals) in WESC and 25 taxa (1,840 individuals) in Dudley Pond in the benthic grab samples collected in 2007. These taxa belonged to 29 major taxonomic groups (Order or greater) in three phyla (Arthropoda, Nematoda, Mollusca). Of the 53 unique taxa (including a single "unknown"), 16 were detected in all three locations, 16 were detected in only the east estuary, 6 were only in Dudley pond, and 6 were only in WESC. Most taxa were identified to family (22) or genus (17), with the remainder identified to order or suborder (6), class (6), or phyla (1; nematodes). A rarefaction species accumulation curve for all sites, using the Chao1 estimator (Chao, 1984) in the program Primer (version 6; Clarke and Gorley, 2006), indicated that we were still detecting new taxa as we added more samples, a result typical of abundant and diverse groups of organisms (fig. 7.4). However, the species accumulations were different among the three locations, as the curves appeared to level off in the east estuary, but were still climbing in Dudley Pond and WESC. The taxa diversity



Figure 7.4. Species accumulation curves for benthic invertebrate samples , showing the observed and estimated taxa richness in the Elwha River estuary, Washington in 2007. (Estimated values from rarefaction using the Chao1 estimator.)

that was estimated with the Shannon diversity index (H') indicated that diversity was low and did not vary among sites or months (fig. 7.5). When averaging across sampling sites and locations, amphipods and dipterans were the dominant taxa representing 50 and 21 percent of identified specimens, respectively (table 7.3). The proportion of dipterans and amphipods was similar in May and September; however, in July the proportion of dipterans in samples increased, whereas amphipods decreased. Spatially, samples collected from WESC seemed depauperate in amphipods when compared to Dudley Pond and the east estuary (table 7.3). This may be due to the paucity of sandy sediments in WESC relative to Dudley Pond and the east estuary. Grab samples indicated that

amphipods and isopods seemed to prefer sandy substrates to silty or muddy substrates. In September, the total abundance of macroinvertebrates of WESC was only 8.1 percent and 12.9 percent of that detected in Dudley Pond and the east estuary sites, respectively. One possible explanation for this was the hydrologic isolation of WESC and lower flows of the Elwha River may have resulted in higher water temperatures and lower dissolved oxygen levels than conditions at other sites. Water quality data (from a Conductivity, Temperature, Depth (CTD) logger; details in Magirl and others, 2011, chapter 4, this report) collected at the time of sampling, seem to corroborate this, although some differences in sampling dates in September limits our ability to infer this from the data.



Figure 7.5. Shannon diversity index scores of the macroinvertebrate community from sediments in the East Estuary, WESC, and Dudley Pond sampling locations of the Elwha River estuary, Washington, in May, July, and September 2007.

Table 7.3. Average number per sample (standard deviation) of amphipods and dipterans, two major components of the macroinvertebrate community inhabiting sediments in the Elwha River estuary, Washington, May, July, and September 2007.

[Values in parenthesis are standard deviation. Abbreviations: n, number; WESC, west estuary channel]

Location	Taxon	Мау	July	September						
East estuary (n = 13)	Amphipods	45 (72)	2 (5)	220 (440)						
	Dipterans	25 (22)	52 (26)	34 (38)						
WESC $(n = 5)$	Amphipods	0	2 (4)	3 (4)						
	Dipterans	89 (65)	107 (69)	28 (27)						
Dudley Pond $(n = 4)$	Amphipods	14 (14)	187 (317)	180 (135)						
	Dipterans	20 (11)	45 (18)	11 (8)						
	Average percentage of total assemblage									
All sites	Amphipods	26 (31)	11 (24)	28 (37)						
combined	Dipterans	46 (32)	64 (31)	41 (35)						

The density of dipterans was greater in WESC than other areas of the estuary in May and July, but was not different from the east estuary in September (table 7.4). The greater densities of dipterans in WESC may be attributed partly to the significant epibenthic detrital layer in contrast to Dudley Pond and east estuary, which may provide greater cover or habitat heterogeneity for macroinvertebrate fauna. More amphipods were detected in Dudley Pond than at the other sites and Dudley Pond was the only site with significant densities of isopods.

A suite of complementary nonparametric multivariate statistics was used to analyze the assemblage structure of benthic macroinvertebrates across months and sites using Primer software. The data were placed into a rectangular data matrix of taxa (rows) by samples (columns). These data were square root transformed to reduce the effects of numerically dominant taxa and matrix wide non-detections (Clarke and Warrick, 2001; McCune and Grace, 2002). Treating each sampling month separately for sediment samples, a triangular resemblance matrix was created, with each cell of the matrix giving the pair-wise similarity between sites based upon the Bray-Curtis distance. Next, non-metric multidimensional scaling (nMDS) was used to graphically analyze whether the assemblage structure of individual sites were grouped by east estuary, west estuary, and Dudley Pond locations. Pairs of sites with similar macroinvertebrate assemblages (in terms of taxa present and their abundances, exclusive of joint-absences) occur closer together in nMDS plots than dissimilar pairs of sites, which occur farther apart. These analyses were done using the full data set (a mixture of taxonomic classifications) and taxa aggregated by orders. The results were similar between these two data sets and results are shown for the full data set unless otherwise indicated.

Based on the graphical analysis of nMDS plots, benthic invertebrates in estuarine sediments differed among the three estuary locations in May and July, but not in September (fig. 7.6). We followed up the graphical analysis with an Analysis of Similarities (ANOSIM), a non-parametric analog to Analysis of Variance (ANVOA), which calculates a ratio of rank similarities (R) that takes a value between 1 and 0. At R = 1, within-group sites are more similar to each other than any sites from other groups, whereas when R approaches 0 similarities among sites do not differ among groups. We used a permutation test with 999 iterations to develop a null distribution to test whether the observed R value was statistically significant.
 Table 7.4.
 Average density of common macroinvertebrate taxa from benthic samples collected in the Elwha River estuary, Washington,

 May, July, and September 2007.

[Average density is number per square meter. Values in parenthesis are standard deviation. Samples were collected using a petite Ponar grab sampler. Abbreviations: WESC, west estuary channel; n, number]

Tayon	W	WESC (n = 5 sites)			ley Pond (n = 4 s	ites)	East estuary (n = 13 sites)			
Taxon	Мау	July	September	Мау	July	September	Мау	July	September	
Acarina	26 (58)	43 (75)	9 (19)	43 (61)	11 (22)	11 (21)	20 (42)	76 (102)	80 (116)	
Amphipoda	0	104 (161)	147 (164)	606 (588)	8,083 (13,711)	7,802 (5,840)	1,951 (3,106)	93 (215)	9,506 (19,040)	
Diptera	3,860 (2,816)	4,622 (2,970)	1,229 (1,170)	887 (488)	1,948 (777)	487 (368)	1,102 (948)	2,261 (1,117)	1,495 (1,665)	
Isopoda	9 (19)	9 (19)	0	1,840 (2,141)	1,309 (1,491)	5,702 (7,298)	0	0	3 (12)	
Odonata	86 (118)	43 (97)	95 (166)	0	0	32 (65)	3 (12)	3 (12)	23 (52)	
Oligochaeta	1,930 (3,084)	1,350 (2,082)	95 (213)	357 (450)	86 (146)	2,943 (5,316)	113 (99)	1,129 (1,900)	632 (1,154)	
Ostracoda	9 (19)	182 (224)	35 (56)	1,742 (2,393)	1,407 (2,813)	3,105 (5,923)	173 (395)	113 (197)	782 (1,887)	
Trichoptera	17 (39)	9 (19)	0	11 (22)	0	0	13 (27)	86 (126)	0	



B. July nMDS (stress = 0.13)







Figure 7.6. Nonmetric multidimensional scaling plots (nMDS) showing assemblage structure of sediment dwelling macroinvertebrates of the Elwha River estuary, Washington, during (*A*) May, (*B*) July, and (*C*) September 2007. (Unitless nMDS plots based on Bray-Curtis similarity of square root transformed data.)

The ANOSIM analysis confirmed the patterns from the nMDS plots. Differences among estuary locations were significantly different in May (R = 0.50, P = 0.001) and July (R = 0.27, P = 0.03) but were not significantly different in September (R = 0.07, P = 0.26). Although statistically significant, the moderate (May) to low (July) R values indicate that the assemblages in the different locations of the estuary showed a fair amount of overlap.

To determine which specific taxa were driving similarities within and differences among estuary locations, we used the SIMPER analysis in Primer. This test decomposes the Bray-Curtis dissimilarity between each pair of sites in the triangular resemblance matrix by estimating the per-taxon contribution to the dissimilarity. The average contribution then is estimated across all comparisons within and among groups; taxa with a large average contribution and a small average variance generally are good discriminating taxa driving differences in assemblage structure between pairs of locations.

In May, the differences in assemblage structure between the east estuary and WESC sites were caused by differences in abundance patterns of Dipterans (27.2 percent of the average dissimilarity), Amphipoda (25.6 percent), and Oligochaeta (23.2 percent). There were more Corophium sp. amphipods in the east estuary and high abundances of Oligochaetes, Tanypodinae, Tanytarsini, and Chironomini diperans in WESC. Differences in assemblage structure between the east estuary and Dudley Pond in May were caused by Ostracoda (26.6 percent of the average dissimilarity), Isopoda (23.8 percent), and Amphipoda (20.0 percent), with higher abundances of Ostracoda and

Sphaeromatidae (Isopoda) in Dudley Pond and higher *Corophium* sp. abundances in the east estuary. Finally, abundances of Ostracoda (20.1 percent of the average dissimilarity), Diptera (18.4 percent), Isopoda (17.2 percent), and Oligochaeta (16.8 percent) were most responsible for the dissimilarity between WESC and Dudley pond. Again, the higher abundances of Ostracoda and Sphaeromatidae in Dudley Pond coupled with the higher abundances of Oligochaeta, Tanytarsini, Chironomini, and Tanypodinae in WESC were responsible for the differences between the locations.

In July, differences between the east estuary and WESC were again caused by different abundance patterns of Diptera (27.8 percent of the average dissimilarity), Oligochaeta (25.6 percent), and Amphipoda (8.6 percent) and Ostracoda (9.4 percent). Top taxa contributing from these groups were Tanytarsini, Tanypodinae, Chironomini, *Corophium* sp., and Chironomidae pupa.

Terrestrial Macroinvertebrates of the Elwha River Estuary

Sampling and Analysis Methods for Terrestrial Macroinvertebrates

Flight-intercept traps (hereafter fallout traps) were used to survey terrestrial insects, in the east estuary. Terrestrial insect sampling was not conducted in WESC or Dudley Pond. Traps were set during May, June, July, and September 2007. These periods generally span the season of highest invertebrate density and the time of maximum juvenile salmon use of the estuary. Five traps were placed in each of three main habitat types: littoral, shrub, and forest canopy (fig. 7.1). Littoral habitats occurred in shoreline areas with an open canopy and emergent marsh vegetation (for example, Juncus sp.; Carex sp.). Shrub habitats were areas dominated by woody shrubs such as black hawthorn (*Crataegus* douglasii), Nootka rose (Rosa nutkana), and twinberry (Lonicera involucrata). Forest habitats were dominated by red alder (Alnus rubra), big leaf maple (Acer macrophyllum), salmonberry (Rubus spectabilis), and red elderberry (Sambucus racemosa). Traps were set, on average, within 1, 6, and 50 m of the estuarine shoreline for the littoral, shrub, and forest canopy zones, respectively. Because we were interested in associating terrestrial macroinvertebrate diversity and abundance patterns with food availability for juvenile salmonids, our analysis was limited to samples associated with the littoral and shrub zones.

Each trap was constructed from a shallow 32×75 cm plastic tub filled with about 5 cm of water and several drops of dish soap to break up the surface tension and facilitate retention of fallen insects. Because the traps were placed on the ground, we attempted to minimize the potential for inundation from tidal fluctuations raising the water level above the lip of the tub, topping the traps, and swamping any captured insects. Around the perimeter of the trap, PVC pipes were driven into the ground; PVC eyelets were attached to the tub and secured through the PVC pipes. This helped allow the tubs to rise and fall with the tide, while they remained in the sampling location (fig. 7.7). After 3 days, the contents of each trap were passed through a 500 micron sieve and preserved in 70 percent ethanol.



Figure 7.7. Two flight-intercept (fallout) traps placed in littoral habitat of the Elwha River estuary, Washington. Each trap is attached with rings to PVC stakes, allowing the trap to rise and fall during tidal fluctuations. (Photograph taken by Matthew M. Beirne, Lower Elwha Klallam Tribe, April 26, 2006.)

Terrestrial Macroinvertebrate Assemblages of the Elwha River Estuary

Across all months, 106 (17 orders) and 73 (14 orders) taxa in the shrub (n = 2,960 individuals) and littoral (2,333) fallout traps, respectively, were collected. Forty-eight taxa were detected in both habitats, 25 taxa were detected only in littoral habitat, and 58 taxa were detected only in shrub habitats. Most taxa were identified to family (88) and genus (25), and the rest were identified to Order (13), superfamily (2), class (2), or phyla (1; nematodes). As with the sampling of benthic invertebrates in the sediment, the rarefaction species accumulation curve lacked a clear asymptote, indicating that this sampling effort probably underrepresented the diversity of the insect communities in these habitats (fig. 7.8). The rarefaction curves were of a similar shape between littoral and shrub habitats. Shannon diversity was higher in the shrub samples than in the littoral samples across all months (fig. 7.9). A large amount of literature exists on patterns of invertebrate diversity and many factors could be responsible for the greater insect diversity, including a greater vegetative diversity (Siemann and others, 1998), biomass (Haddad and others, 2001), and structural complexity (Lawton, 1983) in the shrub transitional zone.

The same suite of nonparametric statistics described previously for sediment samples was used to determine



Figure 7.8. Species accumulation curves for adult invertebrate samples in littoral and shrub habitats and both habitats combined, showing observed and estimated taxa richness in the east estuary of the Elwha River, Washington, in 2007. (Estimated values are from rarefaction using the Chao1 estimator.)



Figure 7.9. Shannon diversity index scores of the macroinvertebrate community from flight-intercept (fallout) traps deployed in shrub and littoral habitat sampling locations of the east estuary of the Elwha River, Washington, in May, June, July, and September 2007.

the assemblage structure of the terrestrial insect community. As with the benthic macroinvertebrate samples, these tests were performed on the full data set and a data set aggregated to order. Results were similar for nMDS and nearly identical for ANOSIM results, so only the results from the full data set are presented here unless otherwise noted. After square root transforming the taxa-by-site matrix for all months combined, an nMDS was run using the Bray-Curtis similarity measure. Unlike the multivariate results for sediment samples (which did not show clear separation among habitats between months), the terrestrial

macroinvertebrates were different across space and time. The nMDS plot showed clear separation between the shrub and littoral insect communities. as well as differences among months. In the shrub and littoral communities, samples from September grouped separately from the May, June, and July samples, which were largely grouped together (fig. 7.10). A two-way ANOSIM analysis confirmed the clear separation shown in the nMDS plots for both location (R = 0.93, P = 0.001) and month (R = 0.63, P = 0.001). A pairwise test showed that June and July were most similar (R = 0.34, P = 0.001) and May and September (R = 0.93, P = 0.001) were most different in their terrestrial macroinvertebrate assemblage structure (across all habitat groups).

A SIMPER analysis of the data was completed to find which groups of invertebrates were responsible for differentiating littoral and shrub habitat. There was a higher abundance of Collembola (21.9 percent of the average dissimilarity), Hymenoptera (11.3 percent), Acarina (10.9 percent), and Araneae (9.6 percent) in the shrub samples and a higher abundance of Dipterans (12.7 percent) and Odonata (7.4 percent) in the littoral samples.



Figure 7.10. Nonmetric multidimensional scaling plots (nMDS) showing assemblage structure of adult invertebrates captured with flight-intercept (fallout) traps placed in (*A*) shrub and (*B*) littoral habitats of the east estuary of the Elwha River, Washington, in 2007. (Unitless nMDS plots based on Bray-Curtis similarity of square root transformed data.)

Juvenile Salmon in the Elwha River Estuary

The restoration of prime spawning and rearing habitat in the Elwha River upstream of two dams that have been in place for nearly a century is expected to benefit salmon as populations recolonize a watershed that once produced large runs (Wunderlich and others, 1994; Pess and others, 2008; Ward and others, 2008; Winter and Crain, 2008). A critical component of salmon population recovery in the Elwha River following dam removal will be from the maintenance or increase in life history and genetic diversity, which will in turn depend in part upon habitat use and performance within the Elwha River estuary (Bottom and others, 2005a). The capacity of the Elwha River estuary to support increased population sizes and life-history diversity of salmon populations will be determined by physical and biological factors, such as food sources, sediment deposition, temperature, salinity, and other habitat features.

The emigration of juvenile Chinook salmon from natal spawning grounds is a critical phase of their life history. Date of spawning, competition, food availability, local genetic adaptation, and variable life history strategies in populations determines the timing and duration of the emigration period, which also varies among species in the same river (Groot and Margolis, 1991). As described by Myers and others (1998), the high potential for interaction among these factors, coupled with spatial and temporal variability driven by natural and anthropogenic factors, creates complexity in determining factors responsible for trends in salmon population size, or phenotypically driven shifts in life history expression.

In addition to these basic ecological questions of interest to fisheries biologists, the timing and abundances of emigrating juvenile salmon, along with estimates of run size, serves fisheries managers tasked with conservation of salmon populations. Finally, estimating vital rates associated with juvenile emigration has been used to track the effects of restoration actions geared toward salmon conservation (Bottom and others, 2005a).

Estimates of the species-specific timing and relative abundance patterns of anadromous juvenile Pacific salmonids during their migration from the Elwha River were made using various techniques throughout the lower river, the estuary, and adjacent nearshore areas (fig. 7.1).

Migration of Juvenile Salmon from the Lower Elwha River

Estimating In-River Migrants Using a Rotary Screw Trap

The Lower Elwha Klallam Tribe has been operating a 2.4 m rotary screw trap at river kilometer 0.5 since 2005 to characterize the emigration of juvenile salmonids from the Elwha River (fig. 7.11). The trap is manufactured from a perforated metal cone mounted between two pontoons. Two tapered flights are wrapped around the inside of the cone, causing the trap to rotate with the flow of the river. The trap is suspended between two pontoons and this barge-like structure is anchored to shore using steel cables. As downstream flow progresses through the trap, smolts traveling downstream are swept into the wide end of the cone and are forced into a trap box at the narrow end of the cone, where they can be identified, measured, and returned to the river

(Volkhardt and others, 2007). The trap is monitored daily to process captured smolts. Occasionally, wood debris or other factors will cause the trap to stop working and no data are collected. For these instances, the daily catch total is estimated based on the total hours of operation during the day in question. If entire days are missed (a rare occurrence), then catch is estimated for missed days using a non-linear (LOESS) regression (G.R. Pess and others, National Oceanic and Atmospheric Administration, written commun., 2010).

Operation of the screw trap in the Elwha River typically is from February or March until mid-June when the Washington Department of Fish and Wildlife (WDFW) releases juvenile Chinook salmon from the Elwha River hatchery. Because of the size of the release (about 3 million fingerlings per year) and the proximity of the release point to the trap, operation of the rotary screw trap ceases upon hatchery release. The data collected from the rotary screw trap provide emigration timing, species composition, length, and genetic samples for part of the emigration period, mostly the wild-origin component of the population. Population estimates are determined by calculating the efficiency of the trap, which is estimated by releasing multiple groups of 1,000 dye-marked (Bismark brown) juvenile chum salmon above the trap and then counting the number of the marked fish caught in the trap downstream. This total then can be used to convert the catch into an estimate of total population size (Volkhardt and others, 2007; G.R. Pess and others, National Oceanic and Atmospheric Administration, written commun., 2010). The estimated trap efficiency was 3.5 percent in 2006 and 4.7 percent in 2007.



Figure 7.11. A 2.5 meter rotary screw trap operated at river kilometer 0.5 by the Lower Elwha Klallam Tribe fisheries office for counting juvenile salmon emigrating from the Elwha River, Washington. (Photograph taken by Michael L. McHenry, Lower Elwha Klallam Tribe, May 26, 2010.)

Results from the Elwha River Screw Trap, 2006–07

An estimated 119,357 (\pm 3,443, 95 percent confidence interval) and 14,309 (\pm 3,440, 95 percent confidence interval) age-0 Chinook salmon migrated past the Elwha River screw trap in 2006 and 2007, respectively (fig. 7.12). This estimate is based on 3,376 (2006) and 266 (2007) captures at the screw trap and does not include hatchery fish or "stream type" age-1 Chinook salmon smolts. However, these numbers should be used with caution. The large difference in smolt numbers between 2006 and 2007 may be explained, in part, by the large flood that occurred on November 6, 2006 (peak flow of 346 m³/s [12,219 ft³/s] reported by Curran and others, [2009]), which may have scoured redds and affected recruitment of the 2007 brood year. In 2006, the trapping did not begin until mid-March, but trapping in previous years indicated that migration timing typically starts earlier than this date in the Elwha River. Additionally, trapping efficiency is variable from year-to-year. Finally, a road closure in spring 2007 precluded deployment of the large screw trap. A smaller 1.5 m trap was used for the 2007 outmigration estimate. Rather than provide estimates of the emigrating population size, the data are provided to show general trends in run timing and juvenile abundance in 2006 and 2007, when estuary use and diet analyses were conducted.



Figure 7.12. Run timing and estimated daily emigration numbers of juvenile Chinook salmon (age-0) based on rotary screw trap data collected by the Lower Elwha Klallam Tribe in 2006 and 2007, Elwha River, Washington. Data from G.R. Pess and others, National Oceanic and Atmospheric Administration, written commun., 2010.

Migration of Juvenile Salmon to the Elwha River Estuary

Detection of Estuary Migrants from Beach Seining

Beach seining techniques were used to sample fish populations in the Elwha River estuary on the east side of the river (fig. 7.13). Due to the shallow nature of much of the estuary and the presence of woody debris obstructing proper deployment of the net, access to suitable seining sites was limited to three primary sites in ES1 (1 site) and ES2 (2 sites) and one secondary site (IEC) (fig. 7.1). Seining started in mid-March and ended in December (2006) or September (2007). Generally, samples were collected every 2 weeks during daylight hours. Samples were collected on 13 occasions each year during 2006 and 2007 (table 7.5). The dimensions of the beach seine were 38 m long \times 2 m deep, with a 2 m \times 2 m bag in the center of the net. Mesh size was 3.18, 6.35, and 31.75 mm, for the bag, center panel, and wings, respectively.



Figure 7.13. Lower Elwha Klallam Tribe fisheries technicians and students from Peninsula College deploying a beach seine used to sample fish assemblages in the Elwha River estuary, Washington. (Photograph taken by Patrick Shafroth, U.S. Geological Survey, July 20, 2006.)

Table 7.5.Monthly sampling effort (number of occasions and seine hauls) used to estimate the fish assemblage structure of the eastElwha River estuary, Washington, 2006 and 2007.

		20	06		2007					
Month	Sampling occasions	ES1 seine hauls	ES2 seine hauls	IEC seine hauls	Sampling occasions	ES1 seine hauls	ES2 seine hauls	IEC seine hauls		
March	1	2	1	0	2	4	4	2		
April	2	3	3	0	2	4	4	4		
May	2	4	3	0	2	4	7	3		
June	1	2	2	0	2	3	4	1		
July	1	2	2	0	2	2	6	2		
August	2	3	4	0	0	0	0	0		
September	2	4	4	0	2	2	2	1		
October	1	2	2	0	0	0	0	0		
November	0	0	0	0	0	0	0	0		
December	1	0	2	0	0	0	0	0		

[Locations are shown in figure 7.1. Abbreviations: ES1, east estuary 1; ES2, east estuary 2; IEC, intraestuarine channel]

Results from Beach Seining in Elwha River Estuary, 2006–07

Sixteen species of fish were identified in the east estuary during 2006–2008 (table 7.6), including five species of salmonids (Chinook, coho, chum, steelhead, cutthroat trout) and one species of charr (bull trout, *Salvenlinus confluentus* captured in 2008). The catch per unit effort (CPUE) was calculated by dividing the number of fish caught by the number of seine hauls. Data from 2006 and 2007, but not 2008, are presented.

The most abundant fish taxa detected, adjusted for CPUE and summed across all sampling occasions, was threespined stickleback (Gasterosteus aculeatus; CPUE, 306 in 2006 and 1,823 in 2007), Pacific staghorn sculpin (Leptocottus armatus; CPUE, 180 and 134), unidentified sculpin spp. (Cottidae sp.; CPUE, 150 and 136), and starry flounder (Platichthys stellatus; CPUE, 99 and 93). The most abundant salmonid species detected were Chinook (CPUE, 136 in 2006 and 73 in 2007), coho (CPUE, 70 and 32), and chum (CPUE, 24 and 20) (table 7.6).

The patterns of seasonal estuary use were similar between 2006 and 2007 for all three salmonid species (fig. 7.14). Juvenile Chinook were caught in the estuary from March through October, with peak abundances in June. Coho showed a similar pattern, with a peak in June, but numbers dropped off more steeply during summer compared with Chinook, to a level where they were rarely detected. Chum were present in relatively lower numbers across all months, with an earlier peak in March or April and were not detected after June. The number of Chinook likely was influenced by hatchery releases from the State of Washington Elwha River Fish Hatchery about 3.2 km upstream. These fish were not externally marked prior to release (however, they do receive thermal marks on their otoliths); therefore, estimates of the proportion of hatchery- to wild-origin could not be made. A sub sample (n=115) of fish collected over the entire season in 2006, however, showed a ratio of 75:40 for wild- versus hatchery-origin juvenile Chinook salmon (see Juvenile Growth by Habitat, 2006 Brood Year).

Coastal storms that occurred in winter of 2006 resulted in flooding that significantly changed the morphology of the Elwha River mouth and reduced access to the east estuary in spring and summer of 2007. A large flood on November 6, 2006, coupled with high tidal conditions (+2.81 m, MLLW) and significant storm surges, resulted in a sediment bar forming, which appeared to preclude fish passage to the east estuary (ES1 and ES2) during 2007, except during high tides and high river flow events. As noted by Shaffer and others (2009), this sediment bar likely disrupted fish movements into the east estuary. Their results showed significant differences in abundances of juvenile salmon between sites in this study in the east estuary and their significantly smaller site in WESC. Results also indicate that the entire fish assemblage of the east estuary was different between 2006 and 2007, probably due to the formation of the sediment bar. The change in CPUE values between 2006 and 2007 were negative for Chinook (-46.3 percent), coho (-54.6 percent), and chum (-14.9 percent salmon (table 7.6).

Table 7.6. Total counts and catch per unit effort of the fish assemblage during beach seining in the eastern Elwha River estuary, Washington, 2006 and 2007.

[Abbreviations: CPUE, catch per unit effort; –, not applicable]

	20	006	2	.007	CPUE
	Total catch	CPUE	Total catch	CPUE	change (percent)
		Salmon	ids		
Chinook	266	136	129	73	-46.3
Coho ¹	127	70	37	32	-54.6
Chum	34	24	38	20	-14.9
Steelhead ²	26	24	44	30	27.1
Cutthroat Trout	1	0.5	2	0.8	60.0
		Sculpi	ns		
Pacific Staghorn Sculpin	350	180	282	134	-25.7
Prickly Sculpin	0	0	22	12	-
Cottid spp.	274	150	324	136	-9.5
		Flatfis	sh		
English Sole	1	0.5	0	0	-100
Starry Flounder	191	99	190	93	-6.0
Flatfish spp.	37	18	0	0	-100
		Forage l	Fish		
Surf Smelt	3	1.5	0	0	-100
Osmerid spp.	15	12	0	0	-100
		Othe	r		
Shiner Perch	2	1	0	0	-100
Threespine Stickleback	591	306	2,675	1,823	496.7

¹Total does not include two coho "jacks."

²Most were hatchery fish from the Lower Elwha Klallam Tribe hatchery that were released into the estuary.

Other pelagic fish, such as English sole (*Parophrys vetulus*), surf smelt (*Hypomesus pretiosus*), other osmerids, and shiner perch (*Cymatogaster aggregata*) were detected in 2006 but not in 2007.

Mean lengths of juvenile salmon in the Elwha River estuary generally increased through the sampling season (fig. 7.15). An outlier was a steep increase in average length of coho salmon in April of 2006 attributed to two coho "jacks" (adult fish who stay in the ocean for less than 1 year and return to the river earlier than their cohort) caught in the east estuary.



Figure 7.14. Catch per unit effort (number of fish captured on occasion *i*/ number of seine hauls on occasion *i*) of juvenile (*A*) Chinook, (*B*) coho, and (*C*) chum salmon detected in the east estuary of the Elwha River estuary, Washington, in 2006 and 2007.



Figure 7.15. Average total length of juvenile (*A*) Chinook and (*B*) coho salmon captured during beach seining in the east estuary of the Elwha River, Washington, during 2006 and 2007. Coho results for April 2006 include two "jacks."

Estuary and Nearshore Habitat Use by Juvenile Chinook Salmon Inferred from Otolith Analysis

Salmonid otolith microstructure can identify different juvenile life history strategies and events (for example see, Neilson and others, 1985; Volk and others, 1995). Previous work on more extensive and complex estuarine habitats in Puget Sound has shown that juvenile Chinook display distinct developmental/ life history "checks" and patterns of growth on the otolith microstructure (Beamer and Larsen, 2004: Lind-Null and others, 2008; Lind-Null and Larsen, 2009, 2010). These distinct checks (a disruption in the microstructure growth pattern) usually corresponded with a transition between habitat types followed by distinct growth patterns, allowing us to quantify differential habitat use. Checks on juvenile salmonid otoliths can be induced (1) naturally, when fish migrate from one habitat zone to another (for example, freshwater to saltwater); (2) naturally, during other life history events (for example, hatching or emergence); and (3) artificially, by thermal marking or other manipulation (such as ponding checks when hatchery fish are moved between rearing vessels).

Otolith Microstructure Methods

Processing and Analysis of Juveniles

In the laboratory, fish fork length (mm) and weight (g) were measured, the sagittal otoliths were extracted from the left and right side of each fish, otoliths were measured by length and weight, and the left otolith was embedded in resin, which was then baked to harden. Each resin block was mounted onto a slide and then ground and polished on both sides to a thin cross-section, which revealed the otolith microstructure for analysis. If the left otolith was composed of vaterite, a crystalline transparent morph of calcium carbonate, which lacks clearly defined growth increments, the right sagittal otolith was substituted for analysis (Neilson, 1992).

Next, images of the ground otoliths were imported into Image Pro Plus image analysis software (ver. 5.1.2) using a CCD color video camera and a Zeiss compound microscope. Two main analyses were of interest: (1) the identification of hatchery fish through thermal marks on the otolith, and (2) the interpretation of otolith microstructure for estimates of residence and growth. On each saved image, a consistent radial axis was drawn and then increments were marked along the axis using standard techniques (Stevenson and Campana, 1992). The distance between marks provides increment width, an indirect measurement of daily growth. Checks indicative of habitat transition also were identified. The number of marks and their average width provided estimates of residence time and growth in designated habitat zones.

River, Estuary, and Nearshore Otolith Microstructural Patterns of Juveniles

Samples were collected from the lower Elwha River, the tidally influenced region of the estuary near the mouth of the river (ES1, ES2, IEC, WESC; locations shown in fig. 7.1), and a few other sites in the river just upstream of the river mouth. Samples also were collected from nearshore areas accessible by beach seine in the Strait of Juan de Fuca east and west of the Elwha River mouth (provided by colleagues; Kurt Fresh, National Oceanic and Atmospheric Administration Fisheries and Shaffer and others, 2009). Based on work in other Puget Sound river systems (Beamer and Larsen, 2004; Lind-Null and others, 2008; Lind-Null and Larsen, 2009, 2010) we divided our sampling a priori into freshwater (FW), forested riverine tidal (FRT). emergent forested transition (EFT), estuarine emergent marsh (EEM), and nearshore (NS) habitat areas. These areas are defined as FW, main stem and side channels, above tidal influence, in the lower Elwha River; FRT, main stem and side channels/sloughs within tidal influence, generally occurring in the uppermost estuary; EFT, a transition between riparian forest and salt marsh, characterized by cattails and scrub/ shrub habitat (ES2, IEC); EEM, true salt marsh (lowermost estuary), cyclically inundated by the tide and characterized by salt tolerant plants (ES1, WESC); and NS, saltwater, shallow sub-tidal and intertidal areas.

We processed 266 juvenile Chinook salmon, collected between March and October 2006 and between May and September 2007. We attempted to collect equivalent numbers of fish in each habitat zone, equally divided between two periods (before and after the release of hatchery fish on June 15th). About one-half of the samples were collected prior to the June release of hatchery fish. Approximately 3 million juvenile Chinook are released from the Elwha River Hatchery operated by the WDFW after they are thermally marked and raised to a certain size. Thermal marking occurs at one of two rearing facilities (Sol Duc and Hurd Creek). Each facility produces a unique mark by exposing the fish to short-term temperature manipulations during early embryonic development, creating easily recognized patterns of dark, distinct increments on their otoliths. These patterns stand in stark contrast to those produced in otoliths from wild fish (compare fig. 7.16 with fig. 7.17; see also Volk and others, 1999). We familiarized ourselves with the unique thermal marking patterns (fig. 7.16) on the otolith microstructure of Elwha River Hatchery fish. This was done for the 2006 and 2007 outmigrant years by processing 25 reference otoliths collected from the two facilities in 2006 and from images available at the North Pacific Anadromous Fish Commission website (for 2006 and 2007). Eighteen sub-yearlings collected in 2006 from



Figure 7.16. Thermally marked juvenile Chinook salmon otolith (ear stone) collected prior to release from the Washington Department of Fish and Wildlife Elwha River hatchery, Washington. Brackets highlight the clear and unique marking patterns present due to a thermal marking process whereby water temperatures are fluctuated during embryonic development to slow and speed growth. This results in a specific pattern that is representative of a particular hatchery, or even brood year, and can be used for positive stock identification. (Photograph taken by Karl Stenberg, U.S. Geological Survey, January 24, 2008.)

the Dungeness River Hatchery also were processed to determine if any mixing occurred between the non-thermally marked Dungeness River Hatchery juveniles and wild-origin Elwha River juveniles in the Elwha River nearshore habitat. Differences in microstructural patterns are more subtle when comparing unmarked hatchery otoliths to wild fish otoliths. Daily growth increments formed in otoliths from hatchery Chinook salmon, immediately after the onset of exogenous feeding, are usually wider and more uniform in width than increments of wild-origin fish. Additionally, hatchery Chinook salmon frequently produce a release/ponding check when they are released from the hatchery or transported to holding ponds (Zhang and others, 1995). Later in the season, Chinook salmon from other river systems could be captured near the Elwha; however, the goals of this study are met simply by the ability to separate Elwha and Dungeness individuals from other stocks.



Figure 7.17. Otolith (ear stone) displaying characteristic growth patterns of wild-origin Chinook salmon captured in freshwater or upper estuary habitats of the Elwha River, Washington. The otolith microstructure depicts points of development (hatch, emergence from the gravel, first feeding) and a freshwater microstructural pattern used as a reference when analyzing otoliths from fish caught in other habitats downstream. (Photograph taken by Karl Stenberg, U.S. Geological Survey, January 24, 2008.)

Processing and Analysis of Adults

The overall objective was to collect otoliths from wild Elwha adult returns to analyze the otolith microstructure from the juvenile portion of the adult otoliths for life history, growth, and habitat utilization. The 2008 and 2009 adult returns were sub-sampled to correspond to juvenile Chinook outmigrants previously collected from the 2005 and 2006 brood years (2006 and 2007 collections). Forty-four and 53 otolith samples, respectively, were collected from September to mid-October in 2008 and 2009, from spawned-out adult Chinook salmon in the lower Elwha River (mouth to river kilometer 4.8). Elwha Klallam Tribal biologists collected samples during the height of the fall Chinook spawning season in the Elwha River.

All 97 samples from the 2008 and 2009 collections were initially aged (table 7.7). The otoliths were placed in deionized water and viewed with a dissection microscope. With the aid of transmitted and reflected light, the number of annuli present were visually

determined. Both sagittal otoliths were used to determine the age of each fish. The left sagittal otolith was then processed (embedded and ground, sulcus-side down) and visually analyzed to determine hatchery or wild origin and on occasion to obtain a more accurate age determination. The continued analysis of the successful wild-origin spawners for life-history information and habitat utilization from the juvenile portion of their otolith microstructure was not possible due to small sample size (see results, below).

Table 7.7. Origin, age, gender, and size (fork length) of adult Chinook salmon collected by Lower

 Elwha Klallam Tribal biologists in the lower Elwha River, Washington, 2008 and 2009.

[Origin was determined by otolith analysis, which reveals whether a fish was born in a hatchery or the wild. **Marked:** Thermal otolith mark. **Origin:** Elwha River juvelile Chinook salmon were thermally marked and reared at two different hatcheries, the Sol Duc and the Hurd Creek Hatcheries. **Abbreviations:** mm, millimeter; Y, yes; N, no; N/A, not applicable; cwt, coded-wire tag]

Date sampled	Fish identifier	River kilometer	Age (years)	Sex	Fork length (mm)	Marked	Origin
			2	2008			
September 16	16	0.0-1.0	3	М	670	Y	Hurd Creek (cwt)
September 17	17	1.6-4.8	4	F	850	Y	Sol Duc
	18	1.6-4.8	4	F	940	Ν	Wild
	19	1.6-4.8	4	М	1,040	Ν	Wild
	20	1.6-4.8	4	F	920	Ν	Wild
	21	1.6-4.8	4	F	760	Y	Hurd Creek
	22	1.6-4.8	4	F	920	Y	Sol Duc
	23	1.6-4.8	4	М	900	Ν	Wild
	24	1.6-4.8	3	Μ	790	Y	Sol Duc
	25	1.6-4.8	4	F	930	Y	Hurd Creek
	26	1.6-4.8	3	Μ	850	Y	Hurd Creek
September 18	27	0.0-3.2	4	М	1,020	Ν	Wild
	28	0.0-3.2	4	Μ	1,030	Ν	Wild
	29	0.0-3.2	4	F	890	Ν	Wild
	30	0.0-3.2	3	М	800	Y	Sol Duc
	31	0.0-3.2	3	F	750	Y	Sol Duc
	32	0.0-3.2	3	М	750	Y	Sol Duc
	33	0.0-3.2	3	М	790	Y	Sol Duc
	34	0.0-3.2	5	М	1,070	Ν	Unknown
	35	0.0-3.2	3	М	780	Y	Sol Duc
	36	0.0-3.2	3	F	700	Y	Sol Duc
September 22	37	0.0 - 1.0	3	М	860	Y	Sol Duc
	38	0.0 - 1.0	4	F	780	Y	Sol Duc

 Table 7.7.
 Origin, age, gender, and size (fork length) of adult Chinook salmon collected by Lower

 Elwha Klallam Tribal biologists in the lower Elwha River, Washington, 2008 and 2009.—Continued

[Origin was determined by otolith analysis, which reveals whether a fish was born in a hatchery or the wild. **Marked:** Thermal otolith mark. **Origin:** Elwha River juvelile Chinook salmon were thermally marked and reared at two different hatcheries, the Sol Duc and the Hurd Creek Hatcheries. **Abbreviations:** mm, millimeter; Y, yes; N, no; N/A, not applicable; cwt, coded-wire tag]

Date sampled	Fish identifier	River kilometer	Age (years)	Sex	Fork length (mm)	Marked	Origin
			2008—	Continued			
September 29	39	0.3-3.2	4	М	940	Y	Sol Duc
	40	0.3-3.2	3	F	640	Y	Sol Duc
	41	0.3-3.2	4	F	790	Y	Sol Duc
	42	0.3-3.2	3	М	790	Y	Sol Duc
	44	0.3-3.2	4	Μ	810	Y	Sol Duc
	45	0.3-3.2	3	F	690	Y	Sol Duc
	46	0.3-3.2	3	Μ	840	Y	Sol Duc
September 30	47	3.2-3.7	4	Μ	960	Ν	Wild
	48	3.2-3.7	4	F	750	Y	Hurd Creek
	49	3.2-3.7	3	F	650	Y	Sol Duc
	50	3.2-3.7	3	М	700	Y	Sol Duc
	51	3.2-3.7	3	F	700	Y	Sol Duc
	52	3.2-3.7	3	Μ	670	Y	Sol Duc
	53	3.2-3.7	3	Μ	640	Y	Sol Duc
	54	3.2-3.7	3	F	650	Y	Sol Duc
	55	3.2-3.7	4	Μ	840	Ν	Wild
October 1	56	2.9-3.2	3	Μ	690	Y	Sol Duc
	57	2.9-3.2	4	F	910	Ν	Wild
	58	2.9-3.2	4	F	840	Y	Sol Duc
	59	2.9-3.2	5	М	1,050	Ν	Unknown
October 6	60	0.0-4.8	3	F	690	Y	Sol Duc
			2	009			
September 22	61	2.9-3.2	4	N/A	N/A	Y	Sol Duc
	62	2.9-3.2	4	N/A	N/A	Y	Sol Duc
	63	2.9-3.2	4	N/A	N/A	Y	Sol Duc
	64	2.9-3.2	5	N/A	N/A	Ν	Wild
	65	2.9-3.2	4	N/A	N/A	Y	Sol Duc
	66	2.9-3.2	4	N/A	N/A	Y	Sol Duc
	67	2.9-3.2	4	N/A	N/A	Y	Sol Duc
	68	2.9-3.2	4	F	760	Y	Sol Duc
	69	2.9-3.2	4	F	750	Y	Sol Duc
September 23	70	2.4-2.7	4	F	750	Y	Sol Duc
	71	2.4-2.7	4	F	790	Y	Sol Duc
	72	2.4-2.7	4	F	710	Y	Sol Duc
	73	2.4-2.7	4	F	680	Y	Sol Duc
	74	2.4-2.7	4	М	780	Y	Sol Duc
	75	2.4-2.7	4	М	770	Y	Sol Duc
	76	2.4-2.7	4	F	730	Y	Sol Duc
	77	2.4-2.7	4	F	660	Y	Sol Duc

Table 7.7. Origin, age, gender, and size (fork length) of adult Chinook salmon collected by Lower

 Elwha Klallam Tribal biologists in the lower Elwha River, Washington, 2008 and 2009.—Continued

[Origin was determined by otolith analysis, which reveals whether a fish was born in a hatchery or the wild. **Marked:** Thermal otolith mark. **Origin:** Elwha River juvelile Chinook salmon were thermally marked and reared at two different hatcheries, the Sol Duc and the Hurd Creek Hatcheries. **Abbreviations:** mm, millimeter; Y, yes; N, no; N/A, not applicable; cwt, coded-wire tag]

Date sampled	Fish identifier	River kilometer	Age (years)	Sex	Fork length (mm)	Marked	Origin
			2008—	-Continued			
September 24	78	0.5-1.0	4	М	870	Y	Sol Duc
_	79	0.5 - 1.0	4	М	800	Y	Sol Duc
	80	0.5 - 1.0	4	М	820	Y	Sol Duc
	81	0.5 - 1.0	4	М	870	Y	Sol Duc
September 30	82	2.2-2.4	4	М	830	Y	Sol Duc
	83	2.2-2.4	4	М	740	Y	Sol Duc
October 1	84	2.2-2.4	4	F	860	Y	Sol Duc
	85	2.2-2.4	4	F	790	Y	Sol Duc
	86	2.2-2.4	4	F	860	Y	Sol Duc
	87	2.2-2.4	4	М	890	Y	Sol Duc
October 5	88	2.4-2.7	4	М	840	Y	Sol Duc
	89	2.4-2.7	4	М	680	Y	Sol Duc
	90	2.4-2.7	4	М	780	Y	Sol Duc
	91	2.4-2.7	4	F	840	Y	Sol Duc
	92	2.4-2.7	4	F	780	Y	Sol Duc
	93	2.4-2.7	4	М	730	Y	Sol Duc
	94	2.4-2.7	4	F	810	Y	Sol Duc
	95	2.4-2.7	4	М	860	Y	Sol Duc
	96	2.4-2.7	4	М	810	Ν	Wild
	97	2.4-2.7	4	F	810	Y	Sol Duc
October 1	98	2.4-2.7	4	F	850	Y	Sol Duc
October 13	99	0.0-0.8	4	F	850	Y	Sol Duc
	100	0.0 - 0.8	4	М	870	Y	Sol Duc
	631	0.0 - 0.8	4	F	870	Y	Sol Duc
	632	0.0 - 0.8	4	F	870	Y	Sol Duc
	633	0.0 - 0.8	4	F	830	Y	Sol Duc
	634	0.0 - 0.8	4	F	880	Y	Sol Duc
	635	0.0 - 0.8	4	М	950	Y	Sol Duc
	636	0.0 - 0.8	4	М	N/A	Y	Sol Duc
	637	0.0 - 0.8	4	F	830	Y	Sol Duc
	638	0.0 - 0.8	4	F	900	Y	Sol Duc
October 15	654	0.0 - 0.8	4	F	950	Y	Sol Duc
	655	0.0 - 0.8	4	F	830	Y	Sol Duc
	656	0.0-0.8	4	F	N/A	Y	Sol Duc
October 13	657	0.0-0.8	4	F	880	Y	Sol Duc
October 13	658	0.0-0.8	4	М	1,020	Y	Sol Duc

Otolith Microstructure Results

Juvenile Growth by Habitat, 2006 and 2007 Brood Years

We separated 115 (2006) and 142 (2007) emigrating Chinook juveniles into marked hatchery origin (fig. 7.16) and unmarked wild or natural origin (figs. 7.17 and 7.18) based on otolith microstructure patterns. Of the 115 Chinook from 2006, 41 were of hatchery origin (40 Elwha River Hatchery and 1 Dungeness River Hatchery), 3 were stream type (age-1) outmigrants, and 71 were wild Elwha

River (age-0). Of the 142 Chinook from 2007, 98 were of hatchery origin (90 from the Elwha River and 8 from the Dungeness River) and 44 were wild Elwha River (age-0). Of the 97 fish collected in the nearshore, 65 were from the Elwha River Hatchery and 8 were from the Dungeness River Hatchery. We analyzed 107 wild-origin juvenile Chinook otoliths (8 samples were unavailable due to poor otolith quality or processing damage). Three life history checks (hatch, emergence, and first feed) were visually and quantitatively located on these samples (figs. 7.17 and 7.18). The increments

became more consistent and identifiable across the radial axis beyond the checks associated with emergence and first feed. Reference samples of juveniles collected from freshwater sites were used to characterize a microstructural reference pattern beyond emergence and first feed unique to the FW habitat zone. Samples collected in the estuary (FRT, EFT, and EEM) and NS were analyzed visually and quantitatively for checks and increased growth (inferred from larger increment widths) beyond the microstructural reference pattern observed on the samples caught in FW habitat.



Figure 7.18. Otolith (ear stone) displaying characteristic microstructure growth patterns of wild-origin Chinook salmon captured in the lower estuary (2006 and 2007) or nearshore (2006) habitats of the Elwha River, Washington. The otolith microstructure depicts points of development (hatch, emergence from the gravel, first feeding), a freshwater reference pattern, an estuarine check where increments are transitioning between freshwater and estuary residence, and increased increment widths beyond the check that are indicative of higher growth potential in some estuarine habitats. (Photograph taken by Karl Stenberg, U.S. Geological Survey, January 24, 2008.)

The results for wild juvenile Chinook salmon revealed two distinct otolith microstructural patterns in 2006 and three distinct patterns in 2007 based on the criteria mentioned previously. The first pattern represented fish resident or captured in FW and FRT (uppermost estuary) habitats for both sampling years (fig. 7.17), because no distinct check or increased growth were apparent on the otoliths of fish caught in FW and FRT habitats. The second pattern (fig. 7.18), characterized by the presence of an estuarine check followed by increased daily growth, represented fish captured in EFT, EEM (lowermost estuary), and NS habitat types in 2006 and in EFT and EEM habitat types only in 2007. The 2007 collection further revealed a third check associated with entrance into the NS habitat type. The estuarine check was comprised of a series of increments with spacing representative of FW and estuarine growth, indicative of a transition period for the fish. The increments beyond the estuarine check were consistently wider and had higher contrast than increments associated with residence in the FW or FRT habitats prior to the check. An estuarine check alone or the check followed by a number of increments representing increased growth was not displayed on the otolith microstructure of fish collected in EFT, EEM, and NS habitats in 2006 until mid May (5 of 10), even though samples were collected in March and April (5 and 3 samples, respectively). The other five samples in May were split between no representation of estuarine residence and presence of an estuarine check at the edge of the otolith. Most of the remaining samples collected from June through September 2006 (20 of 23) displayed an estuarine check followed by an increase in growth. Two of the three samples not displaying increased growth, one in July and one in August, displayed no sign of estuary residence, and the third sample, from August, displayed the estuarine check at the otolith edge.

No samples were collected in the estuary before May 2007, so it is not known what the otolith pattern of early migrants was in 2007. However, 42 of 44 samples from 2007 displayed the estuarine check, with an increase in growth following the check. The absence of increased growth associated with fish captured in the lower estuary (EFT and EEM) before mid-May and the four caught thereafter could be due to lower food availability, which is consistent with stomach content results or lower temperatures affecting growth rates of these early emigrating fish. A more likely scenario emerges, however, given the placement of the estuarine check near the edge of the otolith. Fish collected in March and April may have been caught immediately upon their entrance into the lower estuary habitat and therefore these fish did not have a chance to experience the higher growth potential of the estuary. Some of the fish collected in the middle of the season also were fresh arrivals, whereas those collected later in the season were more likely to have had enough residence time to display increased growth.

A similar scenario of collecting fish shortly after their arrival may also explain the few individuals displaying a check indicative of entrance into NS habitat. A NS check was identified in a single individual in 2006. Although otoliths were collected from EFT and EEM habitats, no unique patterns of growth or checks associated with transition and growth were determined for these two habitat types.

It is not known whether a lack of unique patterns among the five habitat zones is due to small sample size, limitations of the technique, or minimal differentiation among habitat zones. This differentiation, seen in other river systems of Puget Sound (for example, Skagit River), often is caused by habitat specific differences in food availability, water temperature, water chemistry, salinity, spatial complexity, or a combination of these variables. The Elwha River does not have a large and heterogeneous estuary, as opposed to other Puget Sound systems (see Duda and others, 2011, chapter 1, this report), which could have contributed to the minimal differentiation observed on the otolith microstructure.

Once the different patterns of growth associated with transition from freshwater (FW and FRT capture sites) to lower estuary (EFT and EEM sites) and nearshore habitats were identified, the mean increment width (MIW) was used as an indirect measure of growth within these broad habitat categories. The number of daily increments assigned in habitats also was used to determine habitat-specific residence time.

The region of the otolith corresponding to freshwater growth was similar in MIW regardless of the habitat where individual fish were caught, with an average of 3.20 microns (fig. 7.19). The region of the otoliths assigned as lower estuary and nearshore growth showed an increase in MIW by 27 percent (4.06 microns). The average size of juvenile Chinook salmon increased as they migrated successively among freshwater, estuary, and nearshore environments (fig. 7.19B). The average estuarine residence time determined from Chinook captured in the estuary (a minimum estimate, as the fish were sacrificed) was 16 days (range = 4 to 41 days) (fig. 7.19B).Not surprisingly, the average estuarine residence time determined from Chinook salmon captured in the NS habitat showed a longer average residence of 25 days (range = 5 to 50 days), as this number is a more complete estimate because the individuals were captured after they left the estuary.

Based on a preliminary analyses, mostly wild-origin fish were using the lower estuary (EFT and EEM), whereas mostly hatchery fish were using the NS. Therefore, competition between hatchery and wild fish may be minimized in the estuary because most hatchery juveniles seem to bypass this habitat in favor of the Strait of Juan de Fuca. However, additional study would be required to confirm this hypothesis.





An interesting observation was made with three FW and four FRT Chinook salmon collected on July 12, 2006. These Chinook salmon displayed fork lengths and otolith lengths/radial distances that were much smaller than all other fish collected around this time, looking more similar to samples caught in April than to other samples caught in July. Upon examination of "developmental checks" (hatch and emergence relative to capture), we surmised that these fish were progeny of late-spawning parents. Additional sampling would be required to determine whether these results suggest a unique life-history strategy.

Juvenile Dispersal Patterns in Elwha Estuary and Strait of Juan de Fuca, 2007 Brood Year

Because the aforementioned sediment bar formation in the east estuary reduced the catch of wild-origin fish in 2007, an otolith analysis was used to determine origin of 97 juveniles sampled from June 12 to September 11, 2007, in littoral drift cells (drift cells are discreet nearshore areas that receive local sources of sediment) adjacent to the Elwha in the Strait of Juan de Fuca (n = 98) by WDFW (Shaffer and others, 2009) and NOAA (K. Fresh, unpublished data, 2007) as well as from May 17 to August 28, 2007, in the Elwha River estuary (n = 45). The goal was to assess the dispersal of wild- and hatchery-origin (Elwha River hatchery or Dungeness River hatchery) juvenile Chinook salmon in nearshore waters to the east and west of the Elwha River. Samples were collected by beach seine from 16 locations (ordered from west to east): Pysht River, Crescent Bay, Freshwater Bay, east of the Elwha River mouth, Port Angeles Harbor, and Dungeness River. Elwha River estuary samples were collected from the east (ES1, ES2, IEC) and WESC. A random sample of individuals from each sampling occasion was collected, when available, for otolith extraction. Samples were processed as described previously and were compared to reference collections to determine hatchery or wild origin.

Most individuals collected from the Strait of Juan de Fuca were to the east of the Elwha River, although effort was concentrated in these areas. Across all sampling locations in the Strait of Juan de Fuca, most samples were Elwha River hatchery-origin (65) and wildorigin (44) fish. Only eight individuals of Dungeness River hatchery origin were detected (fig. 7.20). Of the Elwha River estuary fish, the largest numbers were collected in WESC (Shaffer and others, 2009), mostly because the sediment bar that was deposited following winter flooding restricted access to the east estuary. Throughout the estuary, 20 wild-origin and 25 hatchery-origin fish were identified.

For management and recovery purposes, Chinook salmon populations are apportioned into evolutionary significant units (Waples, 1995). The Puget Sound Chinook evolutionarily significant unit (ESU) ranges from the Puget Lowlands west to and including the Elwha River. The Strait of Juan de Fuca west of the Elwha River and the coastal rivers comprise the Washington Coast ESU. Populations in these two geographically adjacent ESUs have distinct genetic and life history characteristics (Myers and others, 1998), although Elwha River Chinook salmon appear to be a transition between the two units. Based on coded-wire tag (CWT) recoveries from commercial harvest, Washington Coast ESU Chinook tend to favor a more northern ocean migration, to southeast Alaskan waters, compared to Puget Sound ESU populations. Ocean migration of Elwha River Chinook is more typical of other Puget Sound ESU populations, with most CWT tags occurring in British Columbia and the rest occurring in southeast

Alaska, coastal Washington, and Puget Sound (Puget Sound Indian Tribes and Washington Department of Fish and Wildlife, 2010). It is not known what proportion of the eastward migrating Elwha River Chinook juveniles become resident in Puget Sound, migrate north to the Pacific Ocean through the Strait of Georgia, or return westward to the Strait of Juan de Fuca towards the Pacific Ocean.

Analysis of Adult Chinook Salmon Otoliths

Forty-four Chinook adult returns collected in 2008 were segregated into wild and hatchery origin based on their otolith microstructure (table 7.7). Of these fish, 32 were of hatchery origin, 10 were of wild origin, and 2 were of unknown origin. Therefore, 76 percent of the known-origin Chinook analyzed were hatchery-reared based on the presence of a thermal mark. Twentytwo Chinook were aged age-3, 20 were age-4, and 2 were age-5. Two of the age-3 fish were Hurd Creek marked, 20 were Sol Duc marked, and no fish were wild-origin Elwha age-3 fish. Of the age-4 fish, 3 were marked at Hurd Creek, 7 were marked at Sol Duc, and 10 were wild-orgin Elwha fish. The two age-5 individuals did not have thermal marks, however, it is not known whether they were of hatchery origin because the 2003 brood year pre-dates the Elwha River thermal marking program.

In 2009, all age-3, age-4, and age-5 adult hatchery Chinook would be thermally marked. Of the 53 Chinook adult returns collected in 2009, 51 (96 percent) were of hatchery origin, and two adults were wild-orgin Elwha fish. Fifty-two Chinook were age-4 and 1 was age-5. No age-3 fish were present in the sub-sample. Fifty-one of the age-4 fish were marked at Sol Duc, and one was wild-orgin Elwha fish. The one age-5 fish was of wild origin.



EXPANATION



Figure 7.20. Aerial photographs and pie charts showing results of otolith analysis identifying origin of juvenile Chinook salmon captured during beach seining by Washington Department of Fish and Wildlife (Shaffer and others, 2009) and (K. Fresh, National Oceanic and Atmospheric Administration, unpub. data, 2007). (Origins were from Washington Department of Fish and Wildlife, Elwha, or Dungeness hatcheries and Elwha River wild-origin.) Pie charts depict the proportion of individuals from each location.

Combined returns from both collection years, showed only one wild adult (2009, age-4) corresponding to our 2 years of juvenile out-migrant sampling (2006–07), obviously too few samples to proceed with any life history or estuarine utilization analyses. Our preliminary work from 2008 and 2009 revealed that a significant part of the adult Chinook salmon naturally spawning in the Hunt Road Complex (outside of the index area that WDFW uses for spawner surveys in the lower Elwha River) were of hatchery origin. Combining both collection years, 87 percent of the adult Chinook otoliths examined were of hatchery origin (excluding two unknown origin age-5 fish from the 2008 collection).

Diet and Feeding Strategies of Elwha River Juvenile Salmon

Collection of Stomach Contents for Diet Analysis

Few studies of juvenile salmonid diet in estuarine environments of the central Strait of Juan de Fuca have been completed and no known studies

are published for the Elwha River. The relatively static nature of structural features of the Elwha River estuary over recent decades (Warrick and others, 2009; Shafroth and others, 2011, chapter 8, this report) provides an ideal opportunity to determine the nature of juvenile salmon diets prior to the anticipated changes to these conditions following dam removal. Estuarine seining activities in 2006 and 2007 provided an opportunity to characterize the diet of juvenile salmonids in the estuary. A subsample of the juvenile salmonids detected with beach seining throughout the migration period were selected at random and then sampled for diet. The stomachs of fish sacrificed for otolith analysis were dissected. Some of these fish were collected in the Elwha River above tidal influence. For the purpose of this report, most diet analyses were conducted with juvenile Chinook salmon due to their higher relative abundance (hatchery and wild) and their value to other ongoing scientific studies in the Elwha River basin. Stomach content samples from 149 individuals were collected (99 in 2006 and 50 in 2007) for juvenile Chinook salmon, with

44 from the Elwha River and 105 from the estuary. Stomach content samples also were collected from juvenile coho, steelhead, and cutthroat trout from the Elwha River estuary, but samples were not in high enough numbers during the sampling period for comparative diet analysis.

Stomachs were flushed with gastric lavage (fig. 7.21), following standard techniques (Meehan and Miller, 1978; Cordell and others, 1999, 2001) for collection of stomach contents of juvenile salmonids. Across all sampling occasions, mean length of Chinook was 80 mm (standard deviation [SD] 21) and 88 mm (SD 16) in 2006 and 2007, respectively. Juvenile salmonids were anesthetized with tricaine methanesulfonate (MS-222) until lightly sedated. The stomach contents of the fish were then gently flushed using a 10 or 20 cc syringe filled with freshwater. Stomach contents were either strained through a 106 micron sieve or through a section of nylon attached to a PVC pipe. The stomach contents were preserved in 70 percent ethanol.



Figure 7.21. Gastric lavage of a juvenile Chinook salmon. Flushed contents of its stomach consist of various invertebrate taxa. (Photograph taken by Matt Beirne, Lower Elwha Klallam Tribe, July 13, 2006.)

Results of Diet Analysis for Elwha River Juvenile Chinook Salmon

The diets of juvenile salmonids (all species) in the Elwha River estuary were comprised of a diverse macroinvertebrate assemblage. In 2006, more than 1,100 macroinvertebrates, representing 23 higher level taxonomic groups (order or greater) were collected from stomach lavage and dissections of fish caught in the estuary and lower Elwha River. In 2007, 1,563 macroinvertebrates, representing 16 higher level taxonomic groups were collected in diet samples. When looking at Chinook salmon diets, most of higher taxonomic groups were detected in diets during both years. However, 12 macroinvertebrate orders were detected only in 2006 samples and 2 orders were unique to 2007 samples (table 7.8). When both years were combined, the average number of different taxa per stomach was similar between juvenile Chinook salmon captured in the river (mean = 4.48, SD = 2.5) and in the estuary (mean = 4.72, SD = 2.9) and the overall frequency of taxa richness per stomach was similar (fig. 7.22). **Table 7.8.**Major macroinvertebrate types in stomach contentsof juvenile Chinook salmon samples collected in the lower ElwhaRiver, Washington, and its estuary complex, 2006 and 2007.

[Macroinvertebrate types are by order, unless indicated otherwise. Samples were collected in 2006 (99 individuals) and 2007 (50 individuals). Fewer unique taxa are represented in 2007 likely because samples were collected only in the estuary, whereas in 2006 samples collected in the lower Elwha River also were included]

	Perce	ntage
wacroinvertebrate taxa	2006	2007
Acarina (mites and ticks)	0.2	0
Amphipoda (amphipod crustaceans)	8.9	3.6
Aranaceae (spiders)	0.7	0
Chilopoda (Class; centipedes)	0.1	0
Coleoptera (beetles)	1.4	0.9
Collembola (springtails)	0.3	0.2
Diptera (true flies)	78.5	90.1
Ephemeroptera (mayflies)	4.6	0.2
Hemiptera (true bugs)	1.2	0.8
Hymenoptera (ants, wasps)	0.4	0.6
Lepidoptera (moths and butterflies)	0.2	0
Megaloptera (snakefly)	0.1	0
Malacostraca (Class; mysid shrimps)	0	0.9
Nematoda (Phylum; roundworms)	0.5	0.2
Nueroptera (brown lacewing)	0.1	0
Odonata (dragonfly and damselfly)	0.1	0.4
Oligochaeta (Class; aquatic worms)	0.1	0
Osteichthyes (Class; fishes)	0.1	0
Ostracoda (Class; seed shrimp)	0	0.3
Plecoptera (stoneflies)	0.9	0.3
Psocoptera (barkflies)	0.3	0.1
Thysanoptera (thrips)	0.5	0
Trichoptera (caddisfly)	1.0	0
Turbellaria (Class; flatworms)	0.1	0

Of the 16 higher taxonomic groups of macroinvertebrates present in the diet, 10 were different orders of insects. Dipterans (2-winged flies), representing 11 families were the most abundant taxa, representing 80 and 88 percent of the total number of macroinvertebrates identified from Chinook salmon diet samples in 2006 and 2007, respectively. Amphipods, representing 14 and 5 percent, were the second most abundant taxonomic group in the Chinook diet in 2006 and 2007, respectively. Chironomids were the predominant family of dipterans represented in the diet of all salmonids, with Ceratopogonids, Empidids, and Sphaerocerids represented



Figure 7.22. Frequency of taxa richness (number of unique taxa) in stomach contents collected from juvenile Chinook salmon captured in (*A*) the lower Elwha River above tidal influence and (*B*) the Elwha River estuary, Washington.

in smaller numbers (fig. 7.23). The life stage of consumed dipterans shifted from predominantly pupa in early summer to primarily adult forms in late summer, a pattern that was consistent between years (fig. 7.24). Chironomid larva were consumed at relatively low levels throughout spring and summer. Chironomid larvae provide a valuable food source for fish and other aquatic organisms due to the relatively high levels of protein and high digestibility (Armitage and others, 1995). Although caloric value varies with species, season, and life history, it usually ranges between 4.6 and 6.1 kcal g⁻¹ (Armitage and others, 1995).



Figure 7.23. Average number of individuals per stomach of four main dipteran families in samples from juvenile Chinook salmon Elwha River estuary, Washington, during May–August 2006 and 2007.



Figure 7.24. Average number of chironomids per stomach by life forms (adult, larva, pupa) for all sites in the lower Elwha River and its estuary during May–August 2006 and 2007.

Comparison of Diet Overlap Among Habitat Locations

Habitat-specific diet of juvenile Chinook salmon were examined by comparing the dietary overlap between samples collected from freshwater compared with estuary (2006) and the east and west parts of the estuary (2007). Schoener's index of diet overlap (Schoener 1970) is calculated as:

$$\alpha = 1 - 0.5 \left(\sum_{i=1}^{n} \left| P_{xi} - P_{yi} \right| \right), \tag{1}$$

where

 P_{xi} is proportion of food category *i* in diet of juvenile Chinook salmon in habitat *x*, P_{yi} is proportion of food category *i* in diet of juvenile Chinook salmon in habitat *y*,

n is number of food categories.

Values of α greater than or equal to 0.6 indicate significant dietary overlap. Standard diversity metrics, based on the orders of invertertebrates, also were calculated for comparisons. Dietary overlap between freshwater and estuary habitats was significant for March–May ($\alpha = 0.61$) and June– September ($\alpha = 0.68$) (table 7.9). In March–May, a higher number of orders was measured for juvenile Chinook salmon prey in freshwater samples with less diversity and evenness compared with the estuary samples. A higher proportion of dipterans was selected in freshwater (85 percent of total dietary items) than in estuary habitat (56 percent diptera and 38 percent amphipods). Interestingly, this pattern was inverse during June–September, as more dipterans were present in estuary diets (91 percent) than in freshwater diets (63 percent diptera and 23 percent mayflies, mostly of the genus *Baetis*). In 2007, a comparison of dietary overlap between the east and west estuary detected a high degree of overlap in March–May ($\alpha = 0.91$) and June–September ($\alpha = 0.95$).

Comparison of Prey Use versus Prey Availability in the Elwha River Estuary

As a vital requirement for animal growth, foraging is one of the primary factors in determining the fitness of an organism. Variables such as prey caloric value, handling time, and encounter rate all interact to affect food preference, with some theories suggesting that optimal foraging strategies are evolutionarily driven (Emlen, 1966). Most descriptive studies for juvenile Chinook salmon are limited to comparing the relative frequencies of prey taxa in the diet and in the environment. We examined patterns in prey use of juvenile Chinook salmon in using two approaches. The first was with modified Costello Diagrams (Costello, 1990; Amundsen and others, 1996), which graphically examines the proportion and frequency of occurrence of prey in the diet independent of prey availability. The second, Standard Forage Ratio (SFR; Manley 1974), constructs ratios of prey specific use and patterns of prey abundance in the environment.

 Table 7.9.
 Summary statistics of diet composition (insect orders) and dietary overlap of Chinook salmon collected during two seasons (March–May and June–September) in the lower Elwha River (2006), Washington, and its estuary (2006 and 2007).

[Diet items for fish in each habitat type were tallied by order and the total number of orders present (S), the Shannon diversity (H' Prey), evenness (E Prey), and the sum of all prey individuals (N Prey) for all sampled stomachs (N) were calculated. Diet composition was compared between the lower Elwha River (multiple locations between RKms 0.5 and 0.0) and the East estuary in 2006 and between the East estuary and WESC in 2007 using Schoener's (1970) index of proportional similarity (α). **Dipt_Freq.:** frequency of dipteran occurrence. **Abbreviation:** FW, freshwater; WESC, west estuary channel]

Year	Months	α	Location	N	S Prey	H' Prey	E Prey	N Prey	Dipt_Freq. (percent)
2006	March–May	0.61	FW	17	13	0.76	0.30	306	79
			East estuary	19	9	0.95	0.43	322	94
	June-September	0.68	FW estuary	27	13	1.21	0.47	819	85
			East estuary	36	15	0.54	0.20	270	100
2007	March-May	0.91	WESC	16	4	0.30	0.22	342	75
			East estuary	12	10	0.58	0.25	151	83
	June-September	0.95	WESC	11	7	0.22	0.11	227	100
			East estuary	12	9	0.41	0.19	211	92

Modified Costello diagrams were used to interpret diet information of juvenile Chinook salmon collected in the freshwater and estuarine habitats of the Elwha River (fig. 7.25). This graphical method constructs a diagram of percentage of occurrence of each food item (unique taxon) on the ordinate and the prey-specific abundance on the abscissa. Prey-specific abundance is the proportional abundance of a particular taxon in only those fish stomachs in which it was present. Amundsen and others (1996) provided guidance for interpreting the distribution of prey items in the modified Costello diagram in terms of prey importance, feeding strategy, and niche contribution. The distribution of points along the prey-specific abundance axis indicates whether fish exhibit specialized (51-100 percent) or generalized (1–50 percent) feeding. A diagonal line from the origin to the upper right corner of the diagram indicates prey importance, as rare taxa are found in the lower left quadrant of the diagram and abundant taxa are found in the upper right quadrant of the diagram. The other diagonal, from the upper left to lower

right, indicates the contribution of a prey taxon to the niche breadth of the predator. Points in the upper left quadrant indicate high selectivity among individual variability on the prey items consumed, which determines niche breadth, whereas points occurring in the lower right quadrant indicate generalized feeding on the same prey items across individuals (Amundsen and others, 1996; Vile and others, 2005).

Feeding strategies inferred from modified Costello diagrams (fig. 7.25) showed that juvenile Chinook salmon specialized on chironomids in the lower Elwha River and the eastern and western parts of the estuary. Chironomid pupa were particularly dominant in the estuary and to a lesser extent in the lower Elwha River where larval orthocladiinae and mayflies of the genus *Baetis* were also important. Most other prey species were rare and fed on in a generalized fashion by Chinook. The only other non-dipteran prey of any importance was the amphipod genus *Corophium* in the eastern Elwha estuary.



Figure 7.25. Modified Costello diagram showing feeding strategy of juvenile Chinook salmon in three habitats of the Elwha River, Washington. Diagrams modified from Costello, 1990, and Amundsen and others, 1996. Upper left panel shows three interpretive axes related to niche contribution prey importance (rare to dominant), and feeding strategy (generalization to specialization).

Prey use and prey availability patterns were estimated using SFR in the estuary where macroinvertebrate abundance in the diet as well as in the environment was sampled (fig. 7.26). Diet data from 2006 and 2007 was pooled; benthic and terrestrial (from shrub and littoral habitat) macroinvertebrate data were collected from the east and west estuary in 2007. Data were split into three seasons: early (March through May), mid (June through July) and late (August through September). Terrestrial and benthic prey availability data for the early and late seasons were collected in May and September 2007, respectively. For the mid season, benthic invertebrate samples collected in July and the average of terrestrial samples collected in June and July were used. The SFR for prey orders was calculated using the formula provided by Manley (1974):

$$P_i = \frac{\beta_i x_i}{\sum_{i=1}^k \beta_i x_i},\tag{2}$$

where

 β_i is proportion of prey in the diet,

 x_i is proportion of prey in the environment,

i equals 1, 2...*k*.

Values of P_i greater than 0.5 indicate specialization, whereas values less than 0.5 indicate that use is in proportion to availability.

Taxon	Early Season		Mid Season		Late Season					
Acarnia	0	0		•	0	٥	0	\bigcirc		
Amphipoda		\bigcirc			\bigcirc			\bigcirc		
Araneae	\bigcirc	٥		•		٥	\bigcirc		۰	
Coleoptera	٥		•	•	\bigcirc		0	۰		EXPANATIO
Collembola	\bigcirc		•	\bigcirc		۰	\bigcirc			75 percent
Diptera	\bigcirc						\bigcirc	\bigcirc		50 percent
Hemiptera	\bigcirc			\bigcirc		٠	\bigcirc			25 percent
Hymenoptera	\bigcirc		۰	\bigcirc		•	\bigcirc		•	20 percent
Odonata	0	0		•	0		0	0	•	
Ostracoda		\bigcirc			\bigcirc	٥		\bigcirc	۰	
Plecoptera					٥	ø			۰	
Thysanoptera	•			0		0	0			
Trichoptera	•	0		0	0	•	0		•	
L	estilal h	Aquatic	Dilet	estial b	auatic	Dilet	estilal .	Aquatic	Dilet	1



The SFR ratios indicate that the two main prey taxa, Amphipods and Dipterans, generally were not positively selected for, but rather were selected because they were the most abundant prey available (table 7.10). An exception was selectivity for Amphipods in the mid season (SFR = 0.5), although the calculated value was marginally positive. These results are in contrast to the interpretation of feeding strategy based on Costello diagrams, although the importance of Dipterans and Amphipods was apparent using both methods. Those cases that did show highly positive selectivity generally were those represented by few fish and therefore probably are not reliable representations of juvenile Chinook

salmon preference. Coleopterans were positively selected for in all seasons and Trichoptera were positively selected for in the early and late seasons.

Other studies in estuarine environments in the Pacific Northwest also have determined that Dipterans, particularly chironomids, and Amphipods are important prey sources. Shreffler and others (1992) diet study of the Puyallup River estuary in south Puget Sound also determined that chironomids were the most abundant prey consumed by fall Chinook salmon, with Amphipods (particularly of the genus Corophium), Plecopterans, and Cladocerans as secondary prey. Grey and others (2002) also determined that Dipterans, Amphipods, and Trichopterans were of importance in control and restored sites of the Salmon River estuary in Oregon. Roegner and others (2004), working in various habitats of the Columbia River estuary, also found that chironomids and Amphipods were the dominant prey source for juvenile Chinook salmon.

A more comprehensive analysis, for example bioenergetic modeling (see Beauchamp, 2009; Cordell and others, 2011), would give a better estimate of how food availability, consumption, and environmental factors are playing a role in the growth efficiencies of juvenile Chinook salmon in the Elwha River and how those factors relate to the ongoing restoration efforts.

Table 7.10.Average Standard Forage Ratio values for juvenile Chinook salmon diet forindividuals captured in the Elwha River estuary, Washington, in three seasons during 2006and 2007.

[Standard Forage Ratio from Manley (1974). Values in **bold** indicate prey that were positively selected for by juvenile Chinook salmon in each season. Environmental values of prey availability based on data collected in 2007. Values could not be computed during a given season when a prey taxon was not present in the diet (a) or in the environment (b). **Abbreviations:** N, number of individuals captured; n, (standard deviation) number of fish with taxon present; na, not applicable]

Macoinvertebrate taxon	Early season (March–May) N = 20	Mid season (June–July) N = 58	Late season (August–September) N = 20
Acarina	a	0.24 (0.15, n = 2)	а
Amphipoda	0.39 (0.40, n = 13)	0.50 (0.33, n = 20)	0.004 (0.001, n = 6)
Araneae	0.25 (0.12, n = 2)	0.66 (0.08, n = 1)	0.10 (na, n = 1)
Coleoptera	0.97 (0.01, n = 2)	0.54 (0.24, n = 12)	0.83 (0.11, n = 5)
Collembola	0.38 (0.35, n = 3)	0.32 (0.40, n = 2)	а
Diptera	0.35 (0.38, n = 18)	0.42 (0.42, n = 57)	0.42 (0.45, n = 20)
Hemiptera	0.42 (0.35, n = 5)	0.49 (0.31, n = 7)	0.22 (0.17, n = 5)
Hymenoptera	0.44 (na, n = 1)	0.40 (0.30, n = 6)	0.19 (0.18, n = 3)
Odonata	а	а	0.77 (na, n = 1)
Ostracoda	а	0.28 (na, n = 1)	0.01 (na, n = 1)
Plecoptera	a, b	1.0 (na, n = 1)	b
Thysanoptera	а	0.97 (na, n = 1)	а
Trichoptera	0.82 (0.08, n = 3)	0.30 (0.33, n = 6)	0.95 (0.07, n = 3)

Summary

The capacity of the Elwha River estuary to support increased population sizes and life-history diversity of salmon populations will be determined by physical and biological factors, such as food sources, sediment deposition, temperature, salinity, and other habitat features. Our results showed that juvenile salmon use the Elwha River estuary for extended periods and show increased growth when they feed in the estuary. Chinook, coho, and chum salmon were the most common salmonids detected during 2 years of beach seining in the estuary. Fish assemblages in the Elwha River estuary varied between 2 years of sampling (2006 and 2007) likely due to stormdriven physical changes that precluded access to outmigrating juvenile salmon as well as marine fish movements in the estuary.

The differences in fish assemblage structure in the Elwha River estuary suggest that even modest volumes of sediment may create barriers that preclude fish access to critical estuarine habitat. This has management implications in the context of Elwha River restoration that may warrant a more active approach to maintaining fish access during periods of heavy sediment deposition following dam removal. Adaptive management planning, already underway for the lower main stem, would be well informed by taking into consideration the potential sediment impacts on fish access to estuarine habitat. Although significantly elevated turbidity levels are anticipated in the months to years following dam removal, the magnitude of these levels is difficult to anticipate, particularly in the Elwha River estuary. The extent to which elevated post-dam removal turbidity levels in the estuary may adversely affect juvenile salmonid life history and feeding efficiency also is uncertain. Simenstad and others (1982) postulated that naturally elevated turbidity in estuaries during periods of typical juvenile salmonid residence might reduce predator efficiency and benefit their survival. The concentration threshold at which the benefits of elevated turbidity are replaced by adverse life history effects is uncertain and will be a potential subject of study during the dam removal and restoration process.

The effects of dam removal in the Elwha River estuary, like that of the entire river, is likely to be complex, with ecological processes and functions changing across multiple spatial and temporal scales. In the near term, high sediment loads due to erosion of delta deposits in the two reservoirs could have a negative impact on many habitats of the river and estuary, which could change the carrying capacity for species across trophic levels. Conversely, in the long term, as salmon recolonize the watershed and populations rebuild, greater life-history diversity of salmonids will be a key component of successful recolonization and the maintenance of self-sustaining populations (Schindler and others, 2010).

Future studies of the Elwha River estuary, during and following dam removal, should examine the effects of physical changes due to sediment. Macroinvertebrate and fish communities are likely to respond if sediment deposition has large effects on physical factors such as substrate composition and turbidity. Therefore, collection of complementary data to that presented herein would allow examination of potential affects to these communities. Additional data collection, such as sediment traps and turbidity sensors, would help explain the level of changes experienced by biological communities in the estuary. Repeating many of the data collection efforts described in this chapter will be essential in documenting the effects of dam removal and salmon recolonization on the Elwha River estuary.

Acknowledgments

This project involved the coordination with and assistance from researchers from several institutions. including state and Federal agencies. We acknowledge and are grateful for the assistance of Anne Shaffer (Washington Department of Fish and Wildlife [WDFW]) who assisted with seining activities and collected valuable complementary data, and provided specimens for otolith analysis. Kurt Fresh, Anna Kagley, and Joshua Chamberlin (National Oceanic and Atmospheric Administration) also assisted by contributing samples for otolith analysis. Jeff Grimm (WDFW) provided reference otolith samples. Scott Williams and staff of the Dungeness hatchery provided assistance with obtaining hatchery samples. We would like to thank the following individuals for assisting with field work: Ernest "Sonny" Sampson, Larry Ward, Mel Elofson, Mitch Boyd, Raymond Moses (Lower Elwha Klallam Tribe); Rebecca Paradis, Bryan Hara, David Kirner, Jimmy McDougall, Brian Matne, and Michelle Surrette (Peninsula College); Dorothy Chase, Christopher Konrad, Cecile Mathieu, Steve Rubin, Guy Gelfenbaum, and Jon Warrick (U.S. Geological Survey). Database management and mapping were provided by Randall McCoy (Lower Elwha Klallam Tribe) and Theresa Olson (USGS). Special thanks are due to Arthur Frost for completing all of the macroinvertebrate taxonomic identification. This chapter benefitted from discussions with Rich Sheibley (USGS), Jeff Cordell (University of Washington), and Tom Friesen (Oregon Department of Fish and Wildlife). Funding was provided by the USGS Multi-disciplinary Coastal Habitats in Puget Sound (MD-CHIPS) program and from a grant to the Lower Elwha Klallam Tribe from the Environmental Protection Agency. Use of trade names is for convenience of the reader and does not constitute endorsement by the U.S. government.

References Cited

Amundsen, P.-A., Gabler, H.-M., and Staldvik, F.J., 1996, A new approach to graphical analysis of feeding strategy from stomach content data— Modification of the Costello (1990) method: Journal of Fish Biology, v. 48, p. 607-614.

Armitage, P.D., Cranston, P.S., and Pinder, L.C.V., eds., 1995, The Chironomidae—Biology and ecology of nonbiting midges: London, Chapman and Hall, 572 p.

Armstrong, F.A.J., Stearns, C.R., and Strickland, J.D.H., 1967, The measurement of upwelling and subsequent biological processes by means of the Technicon autoanalyser and associated equipment: Deep Sea Research, v. 14, p. 381-389.

Beamer, E.M., and Larsen, K.A., 2004, The importance of Skagit delta habitat on the growth of the wild ocean-type Chinook in Skagit Bay—Implications for delta restoration: Skagit River System Cooperative, 6 p., accessed June 20, 2011, at <u>http://www.</u> <u>skagitcoop.org/index.php/documents/</u>.

Beauchamp, D.A., 2009, Bioenergetic ontogeny—Linking climate and mass-specific feeding to life-cycle growth and survival of salmon, *in* Zimmerman, C., and Krueger, C.C. eds., Pacific Salmon—Ecology and management of western Alaska's populations: Bethesda, Md., American Fisheries Society Symposium 70, p. 53-72.

Bernhardt, H., and Wilhelms, A., 1967, The continuous determination of low level iron, soluble phosphate and total phosphate with the autoanalyzer: Technicon Symposia, v. 1, p. 385-389.

- Bilby, R.E., Fransen, B.R., and Bisson, P.A., 1996, Incorporation of nitrogen and carbon from spawning coho salmon into the trophic system of small streams—Evidence from stable isotopes: Canadian Journal of Fisheries and Aquatic Sciences, v. 53, p. 164-173.
- Bottom, D.L., Jones, K.K., Cornwell, T.J., Gray, A., and Simenstad, C.A., 2005a, Patterns of Chinook salmon migration and residency in the Salmon River estuary (Oregon): Estuarine, Coastal, and Shelf Science, v. 64, p. 79-93.
- Bottom, D.L., Simenstad, C.A., Burke, J., Baptista, A.M., Jay, D.A., Jones, K.K., Casillas, E., and Schiewe, M.H., 2005b, Salmon at river's end—The role of the estuary in the decline and recovery of Columbia River salmon: U.S. Department of Commerce, National Oceanic and Atmospheric Administration Technical Memo NMFS-NWFSC-68, 246 p.
- Brenkman, S.J., Mumford, S.L., House, M., and Patterson, C., 2008a, Establishing a baseline for detection of fish pathogens in Pacific salmonids prior to dam removal and subsequent recolonization by anadromous fish in the Elwha River, Washington: Northwest Science, v. 82 (special issue), p. 142-152.
- Brenkman, S.J., Pess, G.R., Torgersen, C., Kloehn, K.K., Duda, J.J., and Corbett, S.C., 2008b, Predicting recolonization patterns and interactions between potamodromous and anadromous salmonids in response to dam removal in the Elwha River, Washington State: Northwest Science, v. 82 (special issue), p. 91-106.

- Burke, B.J., Frick, K.E., Moses, R.E., and McHenry, M.L., 2008, Movements by adult Coho salmon in the Lower Elwha River, Washington: Northwest Science, v. 82 (special issue), p. 119-127.
- Cavaliere, E., 2010, Elwha River sediments—Phosphorous dynamics under diverse environmental conditions: Bellingham, Wash., Western Washington University, M.S. Thesis, 73 p.
- Chaloner, D.T., and Wipfli, M.S., 2002, Influence of decomposing Pacific salmon carcasses on macroinvertebrate growth and standing stock in southeastern Alaska streams: Journal of the North American Benthological Society, v. 21, p. 430-422.
- Chao, A., 1984, Non-parametric estimation of the number of classes in a population: Scandanavian Journal of Statistics, v. 11, p. 265-270
- Clarke, K.R., and Gorley, R.N., 2006, Primer v6—User manual/ tutorial: Plymouth, United Kingdom, PRIMER-E, 190 p.
- Clarke, K.R., and Warrick, R.M., 2001, Change in marine communities—An approach to statistical analysis and interpretation, 2nd ed.: Plymouth, United Kingdom, PRIMER-E, 174 p.
- Connolly, P.J., and Brenkman, S.J., 2008, Fish assemblage, density, and growth in lateral habitats within natural and regulated sections of Washington's Elwha River prior to dam removal: Northwest Science, v. 82 (special issue), p. 107-118.

- Cordell, J.R., Stamey, M., Tanner, C., and Aitken, J.K., 2001, Fish assemblages and juvenile salmon diets at a breached-dike wetland site, Spencer Island, Washington 1999: Seattle, Wash., School of Aquatic and Fishery Sciences, University of Washington, SAFS-UW-0104, 11 p.
- Cordell, J.R., Tanner, C., and Aitken, J.K., 1999, Fish assemblages and juvenile salmon diets at a breached-dike wetland site, Spencer Island, Washington 1997–98: Seattle, Wash., Fisheries Research Institute, University of Washington, FRI-UW-9905, 15 p.
- Cordell, J.R., Toft, J.R., Gray, A., Ruggerone, G.T., and Cooksey, M., 2011, Functions of restored wetlands for juvenile salmon in an industrialized estuary: Ecological Engineering, v. 37, p. 343-353.
- Costello, M.J., 1990, Predator feeding strategy and prey importance—A new graphical analysis: Journal of Fish Biology, v. 36, p. 261-263.
- Curran, C.A., Konrad, C.P., Dinehart, R., and Moran, E., 2009, Estimates of sediment load prior to dam removal in the Elwha River, Clallam County, Washington: U.S. Geological Survey Scientific Investigations Report 2009-5221, 28 p.
- Czuba, C.R., Randle, T.J., Bountry, J.A., Magirl, C.S., Czuba, J.A., Curran, C.A., and Konrad, C.P., 2011, Anticipated sediment delivery to the lower Elwha River during and following dam removal, chap. 2 *of* Duda, J.J., Warrick, J.A., and Magirl, C.S., eds., Coastal habitats of the Elwha River, Washington—Biological and physical patterns and processes prior to dam removal: U.S. Geological Survey Scientific Investigations Report 2011-5120, p. 27-46.

- Draut, A.E., Logan, J.B., and Mastin, M.C., 2011, Channel evolution on the dammed Elwha River, Washington: Geomorphology, v. 127, p. 71-87.
- Draut, A.E., Logan, J.B., McCoy, R.E., McHenry, M., and Warrick, J.A., 2008, Channel evolution on the Lower Elwha River, Washington, 1939–2006: U.S. Geological Survey Scientific Investigations Report 2008-5127, 26 p.
- Duda, J.J., Coe, H.J., Morley, S.A., and Kloehn, K.K., in press, Establishing spatial trends in water chemistry and stable isotopes (δ^{15} N and δ^{13} C) in the Elwha River prior to dam removal and salmon recolonization: River Research and Applications, DOI:10.1002/rra.1413.
- Duda, J.J., Warrick, J.A., and Magirl, C.S., 2011, Coastal and lower Elwha River prior to dam removal—History, status, and defining characteristics, chap. 1 *of* Duda, J.J., Warrick, J.A., and Magirl, C.S., eds., Coastal habitats of the Elwha River, Washington—Biological and physical patterns and processes prior to dam removal: U.S. Geological Survey Scientific Investigations Report 2011-5120, p. 1-26.
- Emlen, J.M., 1966, The role of time and energy in food preference: The American Naturalist, v. 916, p. 611-617.
- Gray, A., Simenstad, C.A., Bottom, D.L., and Cornwell, T.J., 2002, Contrasting functional performance of juvenile salmon habitat in recovering wetlands of the Salmon River estuary, Oregon, U.S.A: Restoration Ecology, v. 10, p. 514-526.
- Groot, C., and Margolis, L., 1991, Pacific salmon life histories: Vancouver, British Columbia, University of British Columbia Press, 384 p.

- Haddad, N.M., Tilman, D., Haarstad, J., Ritchie, M., and Knops, J.M.H., 2001, Contrasting effects of plant richness and composition on insect communities: a field experiment: The American Naturalist, v. 158, p. 17-35.
- Kloehn, K.K., Beechie, T.J., Morley, S.A., Coe, H.J., and Duda, J.J., 2008, Influence of dams on river-floodplain dynamics in the Elwha River, Washington: Northwest Science, v. 82 (special issue), p. 224-235.
- Konrad, C.P., 2009, Simulating the recovery of suspended sediment transport and river-bed stability in response to dam removal on the Elwha River, Washington: Ecological Engineering, v. 35, p. 1104-1115.
- Lawton, J.H., 1983, Plant architecture and the diversity of phytophagous insects: Annual Review of Entomology, v. 28, p. 23-39.
- Lind-Null, A., and Larsen, K., 2009, Pre-restoration habitat use by Chinook salmon in the Nisqually estuary using otolith analysis—An additional year: U.S. Geological Survey Open-File Report 2009-1106, 18 p.
- Lind-Null, A., and Larsen, K., 2010, Otolith analysis of pre-restoration habitat use by Chinook salmon in the Delta-Flats and nearshore regions of the Nisqually River estuary: U.S. Geological Survey Open-File Report 2010-1238, 28 p.
- Lind-Null, A., Larsen, K., and Reisenbichler, R., 2008, Prerestoration habitat use by Chinook salmon in the Nisqually estuary using otolith analysis: U.S. Geological Survey Open-File Report 2008-1021, 16 p.

- Magirl, C.S., Curran, C.A., Sheibley, R.W., Warrick, J.A., Czuba, J.A., Czuba, C.R., Gendaszek, A.S., Shafroth, P.B., Duda, J.J., and Foreman, J.R., 2011, Baseline hydrologic studies in the lower Elwha River prior to dam removal, chap. 4 *of* Duda, J.J., Warrick, J.A., and Magirl, C.S., eds., Coastal habitats of the Elwha River, Washington—Biological and physical patterns and processes prior to dam removal: U.S. Geological Survey Scientific Investigations Report 2011-5120, p. 75-110.
- Manly, B.F.J., 1974, A model for certain types of selection experiments: Biometrics, v. 30, p. 281-294.
- McCune, B., and Grace, J.B., 2002, Analysis of ecological communities: Gleneden Beach, Oreg., MJM Software Design, 300 p.
- McHenry, M.L., and Pess, G.R., 2008, An overview of monitoring options for assessing the response of salmonids and their aquatic ecosystems in the Elwha River following dam removal: Northwest Science, v. 82 (special issue), p. 29-47.
- Meehan, W.R., and Miller, R.A., 1978, Stomach flushing—Effectiveness and influence on survival and condition of juvenile salmonids: Journal of the Fisheries Research Board of Canada, v. 35, p. 1359-1363.
- Myers, J.M., Kope, R.G., Bryant, G.J., Teel, D., Lierheirmer, L.J., Wainwright, T.C., Grant, W.S., Waknitz, F.W., Neely, K., Lindley, S.T., and Waples, R.S., 1998, Status review of Chinook salmon from Washington, Idaho, Oregon, and California: National Oceanic and Atmospheric Administration Technical Memorandum NMFS-NWFSC-36, 443 p.

- Morley, S.A., Duda, J.J., Coe, H.J., Kloehn, K.K., and McHenry, M.L., 2008, Benthic invertebrates and periphyton in the Elwha River basin: current conditions and predicted response to dam removal: Northwest Science, v. 82 (special issue), p. 179-196.
- Munn, M.D., Black, R.W., Haggland, A.L., Hummling, M.A., and Huffman, R.L., 1999, An assessment of stream habitat and nutrients in the Elwha River basin—Implications for restoration: U.S. Geological Survey Water-Resources Investigations Report 98-4223, 53 p.
- Neilson, J.D., 1992, Sources of error in otolith microstructure, *in* Stevenson, D.K., ed., Otolith microstructure examination and analysis: Canadian Special Publications in Fisheries and Aquatic Sciences, v. 117. p. 115-125.
- Neilson, J.D., Green, G.H., and Bottom, D., 1985, Estuarine growth of juvenile Chinook salmon (*Oncorhynchus tshawytscha*) as inferred from otolith microstructure: Canadian Journal of Fisheries and Aquatic Sciences, v. 42, p. 899-908.
- Pess, G.R., McHenry, M.L., Beechie, T.J., and Davies, J., 2008, Biological impacts of the Elwha River dams and potential salmonid responses to dam removal: Northwest Science, v. 82 (special issue), p. 72-90.
- Pohl, M., 2004, Channel bed mobility downstream from the Elwha dams, Washington: The Professional Geographer, v. 56, p. 422-431.
- Puget Sound Indian Tribes and Washington Department of Fish and Wildlife, 2010, Comprehensive management plan for Puget Sound Chinook: Olympia, Wash., Harvest Management Component: Washington Department of Fish and Wildlife, 230 p., accessed January 20, 2010, at http://wdfw.wa.gov/ publications/00854/wdfw00854.pdf.

- Quinn, T.P., 2005, The behavior and ecology of Pacific salmon and trout: Seattle, Wash., University of Washington Press, 378 p.
- Roegner, G.C., Bottom, D.L., Baptista, A., Burke, J., Hinton, S., Jay, D.A., Simenstad, C.A., Casillas, E., and Jones, L., 2004, Estuarine habitat and juvenile salmon—Current and historical linkages in the lower Columbia River and estuary, 2002: Report of the National Marine Fisheries Service to the U.S. Army Corps of Engineers, Portland District, Seattle, Wash., accessed June 20, 2011, at <u>http://www.nwfsc.noaa.gov/</u> assets/26/7300_07212010_141544_ Roegner.et.al.2004-rev.pdf.
- Ruggerone, G.T., Nielsen, J.L., and Agler, B.A., 2009, Linking marine and freshwater growth in western Alaska Chinook salmon Oncorhynchus tshawytscha: Journal of Fish Biology, v. 75, p. 1287-1301.
- Shafroth, P.B., Fuentes, T.L., Pritekel, C., Beirne, M.M., and Beauchamp, V.B., 2011, Vegetation of the Elwha River estuary, chap. 8 of Duda, J.J., Warrick, J.A., and Magirl, C.S., eds., Coastal habitats of the Elwha River, Washington—Biological and physical patterns and processes prior to dam removal: U.S. Geological Survey Scientific Investigations Report 2011-5120, p. 225-248.
- Schindler, D.E., Hilborn, R., Chasco, B., Boatright, C.P., Quinn, T.P., Rogers, L.A., and Webster, M.S., 2010, Population diversity and the portfolio effect in an exploited species: Nature, v. 465, p. 609-612.
- Schoener, T.W., 1970, Nonsynchronous spatial overlap of lizards in a patchy environment: Ecology, v. 51, p. 408-418.

Schuldt, J.A., and Hershey, A.E., 1995, Effect of salmon carcass decomposition on Lake Superior tributary streams: Journal of the North American Benthological Society, v. 14, p. 259-268.

Shaffer, J.A., Beirne, M., Ritchie, T., Paradis, R., Barry, D., and Crain, P., 2009, Fish habitat use response to anthropogenic induced changes of physical processes in the Elwha estuary, Washington, USA: Hydrobiologia, v. 636, p. 179-190.

Shreffler, D.K., Simenstad, C.A., and Thom, R.M., 1992, Foraging by juvenile salmon in a restored estuarine wetland: Estuaries, v. 15, p. 204-213.

Siemann, E., Tilman, D., Haarstad, J., and Ritchie, M., 1998, Experimental tests of the dependence of arthropod diversity on plant diversity: The American Naturalist, v. 152, p. 738-750.

Simenstad, C.A., Fresh, K.L., and Salo, E.O., 1982, The role of Puget Sound and Washington coastal estuaries in the life history of Pacific salmon—An unappreciated function, *in* Kennedy, V.S., ed., Estuarine comparisons: New York, Academic Press, p. 343-364

Slawyk, G., and MacIssac, J.J., 1972, Comparison of two automated ammonium methods in a region of coastal upwelling: Deep Sea Research, v. 19, p. 521-524.

SPSS Inc., 2010, PASW statistics 18 Core System User's Guide: Chicago, Ill., SPSS Inc., 406 p. Stanley, E.H., and Doyle, M.W., 2002, Trading off—The ecological effects of dam removal: Frontiers in Ecology and the Environment, v. 1, p. 15-22.

Stevenson, D.K., and Campana, S.E., eds., 1992, Otolith microstructure examination and analysis: Oceans, Ottawa, Canada, Department of Fisheries, Canadian Special Publication of Fisheries and Aquatic Sciences No. 117, 132 p.

Tufte, E.R., 2006, Beautiful Evidence: Cheshire, Conn., Graphics Press, 213 p.

Valderrama, J.C., 1981, The simultaneous analysis of total nitrogen and total phosphorus on natural waters: Marine Chemistry, v.10, p. 109-122.

Vile, J.S., Friesen, T.A., and Reesman, M.J., 2005, Diets of juvenile salmonids and introduced fishes of the lower Willamette River, *in* Friesen, T.A., ed., Biology, behavior, and resources of resident and anadromous fish in the Lower Willamette River—Final report of Research, 2000-2004: Clackamas, Oreg., Oregon Department of Fish and Wildlife, 246 p.

Volk, E., Mortensen, D.G., and Wertheimer, A.C., 1995, Nondaily otolith increments and seasonal changes in growth of a pink salmon (*Oncorhynchus gorbuscha*) population in Auke Bay, Alaska, *in* Secor, D.H., Dean, J.M., and Campana, S.E., eds., Recent developments in fish otolith research: Columbia, S.C., University of South Carolina Press, p. 211-225. Volk, E., Schroder, S.L., and Grimm, J.J., 1999, Otolith thermal marking: Fisheries Research, v. 43, p. 205-219.

Volkhardt, G.C., Johnson. S.L., Miller, B.A., Nickelson, T.E., and Seiler, D.E., 2007, Rotary screw traps and inclined plane screen traps, *in* Johnson, D.H., Shrier, B.M., O'Neal, J.S., Knutzen, J.A., Augerot, X., O'Neil, T.A., and Parsons T.N., eds., Salmonid field protocols handbook—Techniques for Assessing Status and Trends in Salmon and Trout Populations: Portland, Oreg., American Fisheries Society and State of the Salmon, 478 p.

Waples, R.S., 1995, Evolutionary significant units and the conservation of biological diversity under the Endangered Species Act, *in* Nielsen, J.L., and Powers, G.A., eds., Evolution and the aquatic ecosystem—Defining unique units in population conservation: Bethesda, Md., American Fisheries Society Symposium 17, Monterey California, May 1994, p. 8-27.

Ward, L., Crain, P., Freymond, B., McHenry, M., Morrill, D., Pess,
G., Peters, R., Shaffer, J.A., Winter,
B., and Wunderlich, B., 2008,
Elwha River fish restoration plan—
Developed pursuant to the Elwha
River Ecosystem and Fisheries
Restoration Act, Public Law 102495: Seattle, Wash., U.S. Department
of Commerce, National Oceanic
and Atmospheric Administration
Technical Memorandum NMFSNWFSC-90, 169 p.

- Warrick, J.A., Draut, A.E., McHenry, M.L., Miller, I.M., Magirl, C.S., Beirne, M.M., Stevens, A.W., and Logan, J.B., 2011, Geomorphology of the Elwha River and its delta, chap. 3 of Duda, J.J., Warrick, J.A., and Magirl, C.S., eds., Coastal habitats of the Elwha River, Washington—Biological and physical patterns and processes prior to dam removal: U.S. Geological Survey Scientific Investigations Report 2011-5120, p. 47-74.
- Warrick, J.A., George, D.A.,
 Gelfenbaum, G., Ruggiero, P.,
 Kaminsky, G.M., and Beirne, M.,
 2009, Beach morphology and change along the mixed grain-size delta of the dammed Elwha River, Washington:
 Geomorphology, v. 111 p. 136-148.

Washington Department of Fish and Wildlife, 2010, Comprehensive management plan for Puget Sound Chinook—Harvest Management Component: Puget Sound Indian Tribes and the Washington Department of Fish and Wildlife, 237 p.

- Winter, B.D., and Crain, P., 2008, Making the case for ecosystem restoration by dam removal in the Elwha River, Washington: Northwest Science, v. 82 (special issue), p. 13-28.
- Wipfli, M.S., Hudson, J.P., and Caouette, J.P., 1998, Influence of salmon carcasses on stream productivity— Response of biofilm and benthic macroinvertebrates in southeastern Alaska, USA: Canadian Journal of Fisheries and Aquatic Sciences v. 55, p. 1503-1511.
- Wunderlich, R.C., Winter, B.D., and Meyer, J.H., 1994, Restoration of the Elwha River ecosystem: Fisheries, v. 19, p. 11-19.
- Zhang, Z., Beamish, R.J., and Riddell, B.E., 1995, Differences in otolith microstructure between hatcheryreared and wild Chinook salmon (*Oncorhynchus tshawytscha*): Canadian Journal of Fisheries and Aquatic Sciences, v. 52, p. 344-352.

This page intentionally left blank