Winter Feeding Ecology of Coho Salmon (*Oncorhynchus kisutch*), Steelhead (*O. mykiss*), and Cutthroat Trout (*O. clarkii*) in the Skokomish River, Washington

by

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ABSTRACT

Winter Feeding Ecology of Coho Salmon (*Oncorhynchus kisutch*), Steelhead (*O. mykiss*), and Cutthroat Trout (*O. clarkii*) in the Skokomish River, Washington

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The Skokomish River, Washington state, has frequent flood events which combined with other factors have caused severe ecological disruption to juvenile salmonid (*Oncorhynchus* spp.) habitat. Although most coho salmon populations in this area are depressed, the habitat is still utilized. The Army Corps of Engineers is performing a General Investigation to plan ecosystem restoration and flood risk management; this diet study characterizes the winter diets of juvenile salmonids in this system in order to inform these investigations and their consequential decisions.

To accomplish this, the diets of juvenile salmonids from four habitat types were assessed; mainstem, tributaries, backwaters, and off-channel ponds. The diets of 223 coho salmon (*O. kisutch*), 31 rainbow trout (*O. mykiss*), and 9 cutthroat trout (*O. clarkii*) were assessed to characterize winter feeding habits. The diets of juvenile salmonids in these different habitat types were compared for prey abundance, relative importance of prey items, stomach fullness, diet breadth, and diet overlap. In addition, the condition factor, fish weights, and fork lengths of fish from the different habitats were also compared.

Prey weights of the salmonids diets indicated that the majority was comprised of benthic macroinvertebrates; Chironomidae, Ephemeroptera, Plecoptera, Trichoptera, other Diptera, Megaloptera, and Oligochaetes. Ephemeroptera and Chironomidae had highest index of relative importance and proportions by weight for coho salmon. Mean stomach fullness for coho salmon was highest in backwaters and tributaries, and did not vary between habitat types. Mean stomach fullness for coho salmon did vary significantly among sites within habitat types: tributaries Swift and Vance Creek were significantly higher than Hunter Creek (P = 0.004 and P = 0.0001, respectively); backwater site South Fork downstream of Vance Creek confluence was significantly higher than North Fork site 2-26 (upstream of X) (P = 0.002). Mean diet breadth was highest in the mainstem and lowest in backwaters and there were no significant differences habitat types. Coho salmon diets in the mainstem overlapped significantly with tributaries (0.62) and ponds (0.78) (Horn's Index).

Mean condition factor values for coho salmon were not significantly different between habitat types. However, there were significant differences in mean condition factor for coho salmon within mainstem sites, tributary sites, and pond sites: the North Fork mainstem site 2-26 values were significantly higher than South Fork mainstem (at the North Fork confluence) ($P = 0.005$); Hunter Creek and Vance Creek values were significantly higher than Swift Creek ($P = 0.002$ and $P = 0.012$, respectively); Skokomish pond 6-22 was mean values were significantly higher than Skokomish pond 6-14 ($P = 0.021$).

Mean weights and FL for coho salmon were not significantly different between habitat types. However, there were significant differences for coho salmon mean weights and mean FL within mainstem sites, backwater sites, and pond sites: mainstem site 2-31 values were significantly higher than the mainstem site at the South Fork and North Fork confluence ($P = 0.002$ weight, $P = 0.008$ FL); the values at the South Fork backwater site downstream of the Vance Creek confluence were significantly higher than the North Fork backwater site 2-26 ($P = 0.013$ weight, $P = 0.041$ FL); Skokomish pond site 6-22 values were significantly higher than Skokomish pond 14 ($P < 0.0001$ weight, $P = 0.0001$ FL); Skokomish pond site 6-21 values were significantly higher than Skokomish pond 14 ($P = 0.005$ weight, $P = 0.024$ FL) .

The overall mean weights, fork lengths, and stomach fullness of coho salmon in this system were lower than typical means for these species in Washington State during the same time of year, suggesting that Skokomish River fish are relatively small and their growth may be food limited. Although diets in the different habitats varied, the lack of differences in fish size suggests that the overall response is similar. The habitats examined in this diet study served different functions for the fish and each is essential for their over-wintering diets. All the habitats assessed in this system are supporting a necessary food base for juvenile salmonids and need to be preserved and further restored. Before any flood remediation work commences in this area, special emphasis needs to be placed on protecting current backwater areas, maintaining access into tributaries, and establishing a system of beaded channels which have demonstrated they are excellent sources of forage food and refuge.

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(d) Introduction

Flooding in the Skokomish River Valley is considered chronic and extensive, with generally 10 to 15 events that are classified as floods annually (classified flood stage is approximately 241 m^3s^{-1}/8,500 cfs) (USACE 2000). The highest peak discharge recorded on the lower Skokomish River of 1,036 m^3s^{-1}/36,600 cfs occurred in 1990 (USACE and USFWS 2008). Base flows in the mainstem are approximately 6 m^3s^{-1}/205 cfs (Peters et al. in prep.). Recorded studies of Skokomish River flooding date back to 1941 by the United States Army Corps of Engineers (Corps), and several have been performed since. Most recently, the Corps was charged with addressing this flooding after the completion of a project management plan for the feasibility study of the Skokomish River basin in 2006 (USACE 2006). The Corps are considering levee and dike removals and/or setbacks, sediment control structures, re-opening of side channels or oxbows, riparian planting, and dredging of certain sections of the mainstem which experience sub-surface flow (USACE and USFWS 2008). Goals of the Corps are ecosystem restoration and flood risk management; by nature the designs and implementation of these projects may temporarily reduce the availability of juvenile salmonid overwintering habitat and prey food base.

Winter is a critical time for these fish because they are shifting their diets to foraging from mainly drift feeding, and their ability to digest food reduces significantly when the temperature declines, often making the cost of acquiring food to exceed the benefit of doing so. Emaciation is common for fish over-wintering in temperate environments, such as the Skokomish River basin. Fish in these areas experience shorter growing seasons which precede periods of scarce winter resources; this places time constraints on acquiring sufficient energy reserves to survive the winter (Schultz and Conover 1999; Post and Parkinson 2001). Over-wintering juvenile salmonids require access into areas of refuge which can protect them from high flows, predators, and can offer sources of forage food. The Skokomish River habitat conditions are poor, reflecting a disturbed system, and any actions taken towards habitat-altering improvements will likely influence the over-wintering salmonid populations.

The Skokomish River has several factors contributing to its current condition; the river currently has low-levels of channel connectivity, partially due to the intermittent levels of remediation. The placement of dikes and levees have narrowed water flow areas, concentrated and accelerated flows, enhanced flooding, and cut off access for fish into side-channels. Other contributors to the river's condition are heavy logging in the lower watershed combined with reduced flows from the installation of two dams on the North Fork of the river. The diminished flow has caused reduced sediment transport and increased deposition; the aggradation has caused the channel bed to rise approximately 4 meters (Jay and Simenstad 1996).

The Skokomish Valley floodplain is the primary point of access for coho salmon (*Oncorhynchus kisutch*) into most tributaries in the lower watershed. The nature of the floodplain environment causes fish to seek refuge during high-flow events; high flows in winter have caused reductions in juvenile salmonid abundance (Cederholm et al. 1997), and reduced macroinvertebrate densities proportional to the flood magnitude (Robinson et al. 2004). With increases in water flows, the fish caught in the system will seek refuge by moving up to 30 km downstream (Peterson 1982a), into ponds which provide a thermal refuge (Swales and Levins 1989), or from the mainstem to off-channel ponds, sloughs, and wetlands that may not have been accessible during the summer (Quinn 2005). Access into these areas is critical during high flows for avoiding predators, preventing displacement downstream, providing refuge, and to find alternative sources of food through the winter season.

Aquatic food sources appear to be more available and important than terrestrial food sources for juvenile salmonids in winter (Nakano and Murakami 2001). Juvenile coho salmon (*Oncorhynchus kisutch*) diets during the winter in Oregon tributaries showed that aquatic invertebrates accounted for 75% of the total mass ingested, and were primarily comprised of aquatic chironomid larvae (Diptera), baetid mayfly nymphs (Ephemeroptera), limnephilid caddisfly larvae (Trichoptera), and winter stonefly nymphs (primarily Capniid) (Olegario 2006). Recently disturbed habitat is usually in poor condition and contains low complexity, making them less capable of sustaining adequate levels of aquatic invertebrates. Skokomish River fish are likely to more susceptible to emaciation because of the combined effects of the river's flood state, the effects of winter, and the continually disturbed, and poor habitat conditions.

Sections containing adequate winter habitat lose fewer fish during freshets and maintain higher numbers of coho salmon in winter than sections without these characteristics (Robinson et al. 2004). Acceptable winter habitat for juvenile salmonids often includes access into stream sections containing deep pools, log jams, and undercut banks with tree roots and debris (Tschaplinski and Hartman 1983), or areas which allow access into intermittent headwater streams (Olegario 2006), tributaries, backwaters, and ponds. During fall and early winter, migrations into riverine ponds and runoff streams appear to coincide with freshets (Cederholm and Scarlett 1982). Channels that are associated with ponds or swamps form highly productive habitat for overwintering fish (Peterson and Reid 1984). A fish's ability to actively access alternative habitats increases their likelihood of survival.

Habitat containing healthy benthic macroinvertebrate populations like ponds, backwaters, and small tributaries, tend to offer substantial alternative food sources. These types of alternative habitats are especially critical for juvenile salmonids that are transitioning into winter feeding habits. Connected refuge habitats such as tributaries, side-channels (oxbows), and ponds offer an alternative source of feeding for the fish. The degree to which an aquatic environment is able to support fish populations is, in part, directly related to the relative abundance of certain aquatic insects (McCafferty 1981).

The abundance of flying and terrestrial insect's declines with temperature, causing the fish to shift their patterns of acquiring food from drift feeding to foraging for benthic macroinvertebrates available during winter. Although it is known that juvenile salmonids primarily consume food organisms that are drifting aquatic insects and the larval stages of terrestrial insects (Quinn 2005), the specifics of their winter diet is not well understood. Knowledge of which types of winter refuge habitat may be sustaining populations of Skokomish River juvenile salmonids would be valuable information to the Corp's remediation actions.

This research evaluates the diets of juvenile coho salmon, steelhead (*O. mykiss*), and cutthroat trout (*O. clarkii*) in four habitat types for foraging habits during the winter months. Although more data is available on coho salmon than other salmonid species, site specific diet information on over-wintering populations is lacking. Diet analysis of juvenile salmonids in this area may provide benefits such as (1) baseline diet characterization, (2) abundance and importance of prey types, (3) quantifying diet overlap for fish among habitat types, (4) quantifying diet breadth and stomach fullness associated with habitat types, and (5) evaluation of which types of habitat may provide best foraging opportunities.

(e) Background

The first European and American settlers began arriving in the Skokomish River valley around 1850, and the first logging camps were established in 1887 (Amato 1995). By the early 1900’s most of the Skokomish floodplain had been converted into pasture (Canning et al. 1988), and by the 1920’s heavy logging of the area had commenced. In 1930 the Cushman Hydroelectric project was completed and consisted of two dams and two powerhouses, which diverted approximately 40% of the Skokomish River delta’s annual mean runoff from the North Fork for power production (Jay and Simenstad 1996). This also eliminated approximately 19.3 km (12 miles) of prime Chinook salmon (*O. tshawytscha*) spawning grounds (Skokomish Tribe and WDFW 2007). The diverted water flows down a pipe from Lake Kokanee, the reservoir below Lake Cushman, and out into Hood Canal about 5 km north of the mouth of the Skokomish River, near Potlatch, Washington.

Figure 1.-Water is diverted from the North Fork Skokomish River, Washington by two Cushman Dams and is piped and released near Potlatch, Washington (arrow points to location where the water is discharged).

Diversion of the water flow from the North Fork and several other factors combine and contribute to the current condition of the river basin habitat. The loss of gravel recruitment into the estuary has diminished eelgrass bed production, and has caused a reduction in the estuary biotic zone (USACE 2000). Aggradation has caused heightened water levels resulting in a loss of deltaic surface area, decreased mesohaline mixing zones, loss of low intertidal habitat and eelgrass beds (Jay and Simenstad 1996), and has caused frequent and substantial flooding. Eelgrass is a critical nursery environment for salmonids, and the loss of intertidal marsh combined with the loss of subtidal estuary area reduces available rearing habitat and refuge areas for juvenile salmonids (USACE 2006); approximately one third of the original marsh areas have been lost to agricultural activities (Bortelson et al. 1980).

In addition to the physical burden, the flooding incurs a heavy financial burden on the Skokomish Indian Tribe, Skokomish Valley residents, other local residents, and taxpayers. The Skokomish Indian Reservation residents are more frequently and severely affected by the flooding than those in the Skokomish Valley above the U.S. 101 Bridge (Canning et al. 1988). A feasibility study, the General Investigation (GI), was initiated using the Corps Puget Sound and Adjacent Water study authority, and funds were provided by the House of Representatives to study the flooding problems in the Skokomish River Basin (USACE 2006).

The Corps is responsible for taking actions to alleviate the flooding, and they intend on restoring proper natural function to the Skokomish River basin while reducing flood damages to valley residents including the Skokomish Indian Tribe (USACE and USFWS 2008). The Corps is currently in the feasibility phase of the GI to address ecosystem restoration and flood risk management. The U.S. Fish and Wildlife Service, in Lacey, Washington, are currently performing a survey of the distribution, abundance, and out-migration of juvenile salmonids and resident fish in the Skokomish River Basin. This diet research has been performed in conjunction with fish sampling and assessments for the GI. One goal of performing this diet study as a part of the GI was to provide the Corps of Engineers with more information on habitat types that are providing food sources to over-wintering populations of juvenile salmonids.

(f) Study area

The Skokomish River basin is situated at the southeast corner of Hood Canal, a fjord tributary to Admiralty Inlet and the Strait of Juan de Fuca (Figure 1) (Peters et al. in prep.). It drains approximately 622 km^2 of the northern part of Mason County (SRBLIT 1994). The basin has the largest estuary and intertidal delta in the Hood Canal basin (HCCC 2005); tidal influence in the mainstem of the Skokomish extends to near the confluence of the South Fork and North Fork (Canning et al. 1988). The mean and diurnal tidal ranges at Union, Washington (on the outer edge of the delta) are 2.4 m (7.8 ft) and 3.6 m (11.81 ft), respectively.

The climate in the Skokomish River basin is generally a temperate, marine climate with wet winters and dry summers (Peters et al. in prep.). The upper portions of the watershed receive approximately 304 cm (120 inches) of rain annually, while the lower portions, near Hood Canal receive approximately 152 cm (60 inches) of rain annually (Peters et al. in prep.). Almost 90% of this annual rain falls from September through April (Canning et al. 1988), and stream flows are fed the rest of the year by snow melt (USDA 1995). Historical daily peaks in precipitation of 15 - 17 cm (6-7 inches) occur between November and February (Phillips 1968).

The Skokomish River originates in a steep 640 km^2 drainage on the southeast side of the Olympic Mountains (Jay and Simenstad 1996), and enters the southern-most point of Hood Canal. The Skokomish River consists of three distinct sections, the North Fork, South Fork, and the mainstem where the North and South Fork converge. The entire river is approximately 128.7 km (HCCC 2005) comprised of 14.5 km of converged mainstem, 53.6 km of North Fork and 44.3 km of South Fork (WDOE 1985). There are three main tributary subbasins, the South Fork (269 km^2), the North Fork (305 km^2), and Vance Creek (64 km^2) (Jay and Simenstad 1996), with tributary streams totaling approximately 434.5 km, with Vance Creek (17.8 km) being the largest (WDOE 1985).

Both the North and South Forks originate in the mountainous areas of Olympic National Park and Olympic National Forest. The uppermost watershed is primarily heavily timbered, while the downstream areas have been extensively logged and suffer from heavy siltation (WDOE 1985). Approximately 80% of the South Fork subbasin has been clear-cut since 1947 (Canning et al. 1988), and an mean of 2.8 km km^{-2} of logging roads have been constructed in the areas of the South Fork subject to timber cutting (Jay and Simenstad 1996).

The South Fork of the Skokomish River originates in the Capitol Peak region of the southern Olympic Range and generally drains southeast for greater than 32 km, with the upper 7.3 km cutting through very narrow, steep-sloped valleys (SRBLIT 1994). The river bottom is primarily rubble and boulders, with some bedrock, low occurrence of gravel riffles (WDF 1975) and is predominately poorly sorted gravels and cobbles (Jay and Simenstad 1996). Most South Fork tributaries exhibit steep mountain stream characteristics; narrow and confined channels, cascades and rapids, rubble and boulder bottoms (SRBLIT 1994). Near river mile 7.0 the South Fork flows through a narrow, deep, steep-walled canyon and abruptly opens into the broad lower river valley (SRBLIT 1994). The gradient in the valley is moderate and contains excellent gravel substrate, however, it is unstable and influenced by erosion, channel changes and shifting gravel bars (SRBLIT 1994).

The North Fork originates in the Mount Skokomish-Mount Stone region and at river mile 28.0 meets and then flows through Lake Cushman and Lake Kokanee. The diverted water is piped through a spillway to the City of Tacoma Power Generating Facility on Hood Canal about 5 km north of the mouth of the river. The remaining mean annual flow in the North Fork is approximately 3 m^3s^{-1}, which prior to diversion was near 27 m^3s^{-1} (Canning et al. 1988). This 96% (USACE 2006) reduction of flow causes an estimated 70% loss of sediment transport (Jay and Simenstad 1996). The upper drainage (upstream of the dams) has precipitous gradients, numerous cascades and falls, and contains predominantly rubble and boulder stream bottom (SRBLIT 1994). Downstream of the dams, the North Fork is characterized by low gradients, eroded banks and heavy siltation at the confluence, and is predominately poorly sorted gravels and cobbles (Jay and Simenstad 1996).

The mainstem Skokomish River is approximately 16 km long and extends to a relatively large estuary at Hood Canal (Peters et al. in prep.). The tidal influence extends approximately 14.5 km (9 miles) upstream to the confluence of the North and South forks (Jay and Simenstad 1996). This section of the river is in a floodplain, is relatively low gradient, and contains sediments that are largely sand and gravel (Jay and Simenstad 1996). Overall, the Skokomish River currently contains approximately 111 river km (69 miles) of anadromous fish habitat (SRBLIT 1994).

Several anadromous and resident salmonid and other non-game fish can be found in the Skokomish River watershed system. Twenty-three species of fish have been found in the mainstem and South Fork of the river (Watershed Management Team 1995). Most of these are salmonids, including Chinook salmon, coho salmon, chum salmon (*O. keta*), steelhead, cutthroat trout, bull trout (*Salvelinus confluentus*), and mountain whitefish (*Prosopium williamsoni*), which are common in the Skokomish River (Peters et al. in prep). Sockeye salmon (*O. nerka*) and pink salmon (*O. gorbuscha*) were historically found in the Skokomish River (Peters et al. in prep). Five species of sculpin (*Cottus* sp.) are found in the Skokomish River, including prickly sculpin (*C. asper*), coast range sculpin (*C. alecticus*), riffle sculpin (*C. gulosus*), Reticulate sculpin (*C. perplexus*), and shorthead sculpin (*C. confusus*) (Mongillo and Hallock 1997). River lamprey (*Lampetra ayersii*), western brook lamprey (*L. richardsoni*) and Pacific lamprey (*L. tridentata*) have also been observed in the basin (Peters et al. in prep).

Four fish found in the Skokomish River are listed as threatened under the Endangered Species Act of 1973 or have Evolutionary Significant Units (ESU) there; Chinook salmon (a Puget Sound ESU), chum salmon (Hood Canal Summer-run), steelhead (Hood Canal Winter Steelhead), and bull trout (NMFS 1999; USFWS 1988; USFWS 1999). Salmonid populations within the North Fork watershed above the dams consist of landlocked steelhead, cutthroat trout, brook trout (*Salvelinus fontinalis*), sockeye salmon, mountain whitefish, and a lucastrine stock of Chinook salmon and bull trout (Brenkman 2001). Pink salmon, spring Chinook, and early chum have been extirpated from the South Fork of the river (WDNR 1997).

(g) Methods

Fish collections

Coho salmon, steelhead, and cutthroat trout were collected from tributaries, off-channel ponds, lateral backwaters, and the mainstem from January 2009 through March 2009 using seining and electrofishing techniques (Table 1 and Figure 2). These sample sites were randomly selected from the GI study sites used in its overall biological sampling plan (Peters et al. in prep.); they were selected using Generalized Random Tessellation Stratified (GRTS) Spatially-Balanced Survey Designs for Aquatic Resources (Stevens and Olsen 1999). GRTS was used to designate sites for the biological sampling completed for the Skokomish River GI (Peters et al. in prep.). Because these selected sites have the same spatial distribution as the stream network from which they were drawn, measurements made at them can be used to infer conditions within the entire network for the purposes of the GI (USACE and USFWS 2008). For a complete list of the coordinates where fish for the diet analysis were obtained please see Table 2.

Stomach samples were collected from habitats within these stream/river reaches or pond transects (Figure 2). River/stream reaches were centered on the points developed from the GRTS selection process. The length of the reach where sampling was completed was equal to ten times the bank-full width or 150 m, whichever was greatest (Table 2). Pond samples were collected along two 150 m by 3 m transects; one near-shore and one off-shore. These two transects were centered on the GRTS selected point. For this diet study, if a sufficient sample size was not obtained along these transects the sample area was increased until an adequate sample size was obtained.

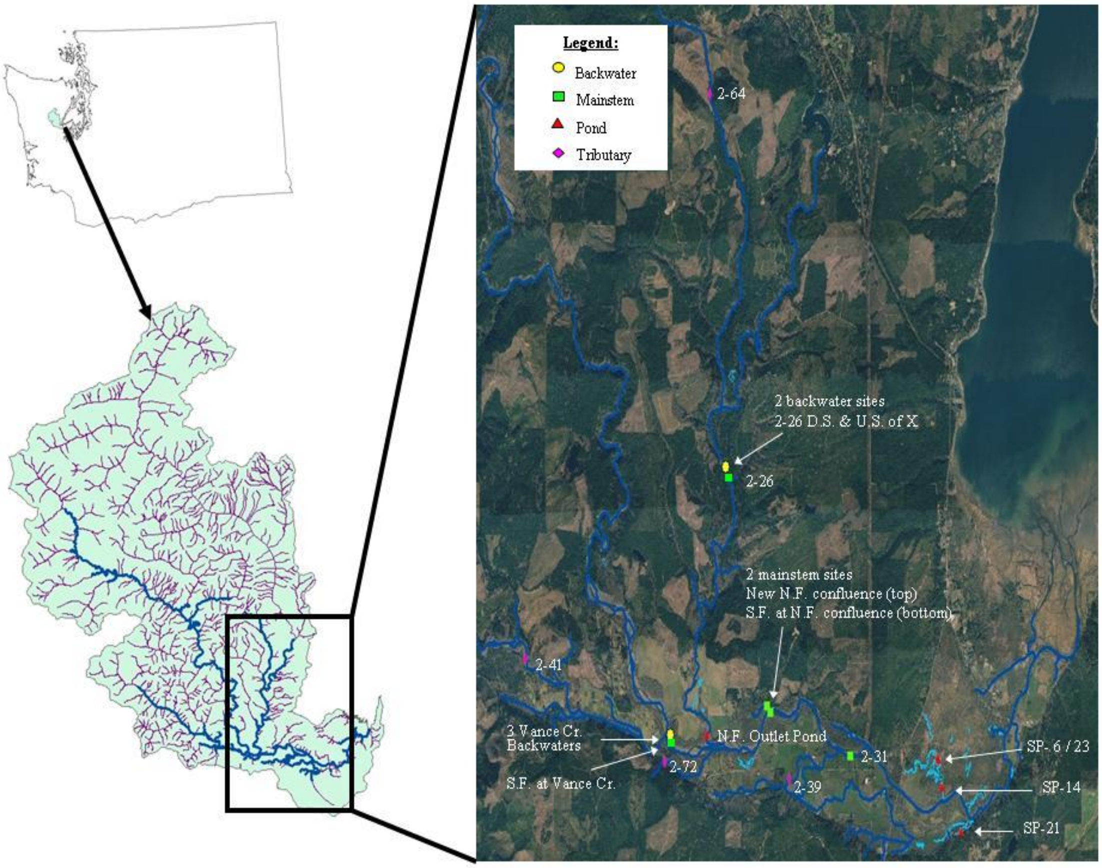


Figure 2.-Map of the study area and all sample site locations for diet study, lower Skokomish River, January through March 2009. Each marker indicates one site except for the backwater sites. Site identification numbers are used to identify site locations, see Table 2 for a detailed list of site location characteristics.

Water quality measurements were collected at most sites. Sites in which measurements were unobtainable due to equipment failure are notated by dashes in Table 2. Water quality values collected include turbidity (ntu), dissolved oxygen (DO), temperature (°C), pH (Table 2). GPS coordinates (UTM NAD 83) were collected with a Garmin (model GPS map76S), and bank full widths (BFW) were also collected (Table 2). Temperature and pH measurements were collected using a Yellow Springs Instrument (YSI) (model 60), and Dissolved Oxygen (DO) measurements were collected using a YSI (model 85). All instruments were calibrated prior to use.

There are several techniques of classifying habitat. For this study the mainstem is defined as the section of the river in which flow doesn't branch off of the main flow of the river, including braided channels. Braided channels are overlapping islands which form more than two flow channels, and are generally unstable (Arend 1999a). Tributaries are considered streams and creeks that actively transported water from the surrounding drainage basin and converged with the mainstem of the Skokomish River. Backwater habitat was defined as water collections that occur laterally with the mainstem of the river, and for this study include locations in tributaries that are near its convergence with the mainstem. Backwaters generally consist of standing or very slowly moving water that connected to the mainstem in some manner, and is fed by underground seepage or river flow. Ponds are considered off-channel habitat as they usually form with higher flows and remain after flow conditions have stabilized. Ponds are generally connected to the mainstem by relatively small egress channels that may become intermittent during low-flow conditions (i.e. summer).

Fish were caught at night, between one to three hours after dusk because research has shown that nighttime fish counts far exceeded daytime counts in the winter; fish generally hide and are inactive during the daytime (Roni and Fayram 2000). In less structurally restrictive areas a 5-mm square size mesh pole seine, with 1.8 m depth, 1.8 m width, and 9.1 m length was used to collect fish from banks, ponds, and other shallow and slower moving water. A Smith Root, Inc. (model LR-24) backpack electro-fisher was used to stun fish found in various log jams and around other in-water structures where use of the pole seine was ineffective. A Smith Root, Inc. raft Electrofisher (model

GPP) on a modified NRS Otter Raft was used in deeper areas (ponds) where foot access was prohibitive. Fish were removed from the nets and placed in buckets for processing.

Fish processing and stomach content sampling

After fish collections were completed, fish were anaesthetized with MS-222, and identified. They were measured for fork length (FL, nearest mm) and weight (nearest 0.01 g). The scale (OHaus Scout Pro, model SP402) was calibrated each sampling day using a standard 200-g weight. A modified pesticide applicator was used to lavage fish for stomach contents (Foster 1977). The sprayer was fitted with an adapted copper nozzle of appropriate length and diameter; selected for fish length and girth. Stomach contents were flushed into a 425 µm mesh sieve, and then rinsed into a whirl-pack bag. Water-proof data labels containing site number, sample number, date, and fish information (species, length, and weight) were also placed inside sample bags. Stomach samples were then placed on dry ice and kept in a cooler. Fish were allowed full recovery and returned to the location of capture. Stomach samples were transported back to the laboratory, and placed in a freezer until later analysis.

Only fish greater than 54 mm fork length (FL) and weighing more than 1.67 grams were sampled for stomach contents because fish were anesthetized and the lavage process needed to be expedited in order to minimize impacts to fish. More time would have been required to change lavage nozzle heads to a size necessary to perform gastric lavage on smaller fish. Additionally, due the small size of fish under 54 mm FL and 1.67 g, it was unclear if the lavage process would have harmed them. The nozzle selected was the appropriate size for the majority of the fish that could be collected and lavaged with the least amount of time and potential harm.

Laboratory analysis

The samples were thawed by placing the frozen whirl-pack sample bags into a shallow tub of cold water until thawed. Stomach contents were rinsed into a Petri dish, and then sorted with the aid of a dissecting scope (American Optical Stereo Star Model A0569 with zoom of 0.7 x 3.0 x). Aquatic insects were identified to family, genus, and species if possible and only to order if more specific identification was impossible. Some items were categorized differently than the other prey items because they offered little to no caloric value to the diet. These items included rocks, plant material, unidentifiable inorganic matter, and Trichoptera cases, which were categorized as “other”, and were not included in data analysis. Coleoptera presented a special issue and were classified as terrestrial insects here, however, they are known to frequently move between aquatic and terrestrial environments and could have been characterized as aquatic (McCafferty 1981).

Miscellaneous insect parts that were unidentifiable were distributed amongst the other insect contents in the stomach sample according to their percent proportion of mass. Stomach contents were assigned prey codes (Appendix A), cataloged, and preserved in 95% ethanol. McCafferty (1981) and Merritt and Cummins (1996) were referenced for aquatic entomology identification. Ingested fish were classed to genus, and species if possible. Fish were identified by external characteristics if they are not too degraded, or by diagnostic bones (i.e., cleithrum and dentaries) if the fish tissue was well-digested. Sorted contents were dabbed on absorbent towels for at least three seconds before weighing to the nearest 0.0001 g on a balance (Denver Instrument M-220).

Data Analysis

Pielou’s method considers the cumulative diversity and quantity of prey items to determine if adequate sample sizes have been collected; the purpose of using Pielou’s method is to perform unbiased data analysis (Hoffman 1978). In Pielou’s method, H_k is plotted versus k, the number of pooled stomachs, and as the stomachs are pooled, H_k initially tends to increase, and if the number of pooled stomachs (k) is large enough, H_k should level off. The following formula was applied to calculate Pielou’s method;

$$H_k = (1/N_k) \log (N_k !/\pi N_{ki})$$

where: H_k = the diversity in k pooled stomachs (k = 1 to n),

N_k = the number or individuals in these stomachs, and

N_{ki} = the number of individuals in the i^{th} species in k pooled stomachs.

Pielou’s method was first calculated on each of the individual sample sets from each habitat, and then all stomach samples were combined to represent each of the four habitat types as a whole (separately for each species of predator fish). See Appendix C and D for results of the determination of adequate sample size for individual sample sets for coho salmon and trout, respectively.

To quantify prey abundance, values of percent proportion by weight (%W_i), percent proportion by number (%N_i), and percent frequency of occurrence (%O_i) (Liao et al. 2001; Chipps and Garvey 2007) were calculated;

$$\%W_i = \frac{100W_i}{\sum_{i=1}^{n} W_i},$$

$$\%O_i = \frac{100O_i}{\sum_{i=1}^{n} O_i},$$

$$\%N_i = \frac{100N_i}{\sum_{i=1}^{n} N_i},$$

where: n = total number of prey categories found in a given sample,

%W_i = percent proportion by weight of prey type,

%N_i = percent proportion by number of prey type,

%O_i = frequency of occurrence*.

* the count of stomachs containing a specific prey item divided by the total number of stomachs with food in them

Proportion by number (%N_i) can allow small prey items to represent a dominant component of the diet, while proportion by weight (%W_i) emphasizes the relative contribution of larger prey, and frequency of occurrence (%O_i) can describe how often a particular prey item was eaten; however, none of these alone can provide an indication of the relative importance of prey to the overall diet (Chipps and Garvey 2007).

The four tributaries from which stomach samples were collected varied in their physical characteristics and were separated and grouped to compare the diets between streams with similar characteristics. For coho salmon, Vance Creek diet information was compared against Hunter Creek and Swift Creek combined. Vance Creek is a valley tributary, has its own subwatershed, and has a steep gradient. Hunter Creek and Swift Creek were grouped because they were different from Vance Creek and were very similar to each other; lower velocity, and lack a watershed to feed their flow. Vance Creek would have been grouped with McTaggart Creek and compared to Swift and Hunter Creeks if coho salmon had been captured there.

It is believed that compound indices like percent index of relative importance (%IRI) can represent all the unique properties affecting individual measures, and capture more information than do single, component measures; %IRI can therefore provide a more balanced view of fish's diets (Chipps and Garvey 2007). The %IRI values were calculated according to Bowen (1996);

$$\%IRI = \frac{100 IRI_i}{\sum_{i=1}^{n} IRI_i}, \qquad \text{where:} \quad IRI = \%O_i\,(\%N_i + \%W_i).$$

Comparisons here will utilize both the percent proportion by weight and the index of relative importance; this study is primarily concerned with the overall importance of prey items in the diet, and both characteristics are valuable. Only prey items which constituted a significant proportion of the diet according to prey weights (%W_i) and importance (%IRI) will be represented in the graphs, the remainders are grouped as other. Additionally, prey items which constituted a significant proportion of the diet according to the %IRI will be represented in tables; the remainders are grouped as other.

Stomach fullness values can be used to describe the quantity of prey items being obtained if collected during the appropriate times within a 24 h period, depending on seasonal temperature (Beauchamp et al. 2007). Stomach fullness (K_f) values for each fish was calculated as;

$$K_f = [\text{wet weight of stomach contents (g)} \,/\, \text{wet weight of stomach contents (g)} - \text{wet weight of fish (g)}] * 100.$$

Diet breadth is commonly used to describe the degree of species present (Levins 1968), and was calculated to compare diet diversity. Diet breadth was calculated for each species in each habitat type;

$$B = \frac{1}{\sum p_i^2},$$

where p_i = the proportion of the diet that is comprised of food type i.

Index values range between 1 (meaning there is only one prey type present, i.e. there is no diet breadth) and infinity. Values less than two indicate little diet breadth (Tabor et al. 2001).

Niche overlap indices, or diet overlap indices, are often used to measure the extent of resource overlap among different species, or to infer competition (Chipps and Garvey 2007). Diet overlap was determined with Horn’s index of overlap because it is the method recommended when data are expressed as biomass rather individual prey counts (Krebs 1989; Chipps and Garvey 2007). The use of prey counts can be misleading because prey items can be small, have low caloric content, and may require more effort to obtain than larger prey items of equal mass. High prey counts don’t necessarily indicate they are acquiring enough caloric sustenance. Horn’s index uses proportions by weight and has been shown to be the least biased when the following are present: changing quantities of resources, resource distribution, and uneven sample sizes (Horn 1966; Krebs 1989; Chipps and Garvey 2007).

Horn's index values were tabulated and the most specific information possible for prey items was used to maximize the robustness of the index values. For several prey items identification was only possible to order or class due to their small size. Most prey items were classable to order and family and several were identifiable to species (Appendix E). Horn's index values were calculated from the relative proportions by weight;

$$C\lambda = \frac{2\sum_{i=1}^{s} X_i * Y_i}{\sum_{i=1}^{s} X_i^2 + \sum_{i=1}^{s} Y_i^2}$$

where:

$C\lambda$ = Horn's index of diet overlap,

X_i = proportion of total diet of species/habitat type X contributed by food category *i*,

Y_i = proportion of total diet of species/habitat type Y contributed by food category *i*, and

S = food categories.

Overlap index values can range from 0 (no overlap) to 1 (complete overlap). Diet overlap is commonly considered biologically significant when values exceed 0.60 (Zaret and Rand 1971). Coho salmon diet overlap was compared between all four habitat types; backwaters, mainstem, ponds, and tributaries. Steelhead diet overlap was compared between backwaters and mainstem, and then compared between coho salmon diet in backwaters and mainstem. Cutthroat trout diet overlap was compared between coho salmon diet in tributaries.

Mean weights and FL's were used to determine Fulton's condition factor;

$$K = (W/L^3) * 100{,}000$$

where: K = condition factor (metric),
W = weight of fish (g),
L = length of fish (mm).

Mean weights, FL, condition factor, and stomach fullness were then compared using nested analysis of variance ($P < 0.05$) to evaluate whether the effects of habitat on fish weight, fork length, and condition factor were significant for coho salmon between habitats types; calculated with statistical software program SAS (SAS Institute Inc.). One sample site was excluded from mainstem, backwaters, and ponds to produce a balanced design with equal sample size to allow a nested ANOVA to be completed; tributaries caused this exclusion because only three tributary sites produced enough coho salmon to compare. Multiple comparison Tukey's Honestly-Significant-Difference Test calculated pair-wise comparisons to determine between which population means significant differences exist ($P < 0.05$) (Zar 1984, 1999).

Condition factor values are meant to represent a fish's overall robustness (Anderson and Neuman 1996; Cunjak and Power 1987) and to indicate the level of tissue energy reserves a fish has; based on the assumption that a fish in good condition would demonstrate faster growth rates, greater reproductive potential, and demonstrate higher survival than those with lower condition factor levels, given comparable environmental conditions (Pope and Kruse 2007). There are commonly associated factors with condition like the season, environment, and spatial variations that can influence fish condition; however, if interpreted correctly, condition factor can be used to characterize environmental components of fish habitat (Pope and Kruse 2007). The formula applied to calculate condition factor was not species specific; a more species specific formula may have produced more accurate condition values. Therefore, coho salmon and trout condition factors were not compared due to the allopatric growth differences between species.

(h) Results

Stomach collections and site characteristics

A total of 309 salmonid stomach samples were collected from January to March 2009, which included 226 coho salmon, 64 steelhead, and 18 cutthroat trout samples (Table 1). The early February attempts to capture coho salmon in the mainstem and backwaters, 2/3/09 and 2/5/09 respectively, yielded few coho salmon (Table 1); steelhead were the abundant species captured, and were alternately sampled. It was notable that cutthroat trout were the only species of salmonid captured at McTaggart Creek. Coho salmon were consistently captured at all pond sites; only one pond site yielded any rainbow or cutthroat trout (Table 1).

Table 1.-Habitat types and sample locations of each sampling effort. The numbers in parentheses are site identification numbers. The centers of the site transect reaches are labeled "X"; reaches were ten times the bank full width, or 150 m, whichever was greater. Skokomish pond site SP-6/22 was one very large pond, but contained two designated sites, SP-6 and SP-22.

Habitat type and sample site	Sampling date	coho salmon	steelhead	cutthroat trout
MAINSTEM				
Mainstem (2-31)	1/29/09	10	11	0
South Fork at Vance confluence	2/03/09	1	14	0
North Fork at confluence	2/12/09	15	0	0
South Fork above and below North Fork confluence	2/17/09	11	0	0
North Fork mainstem at log jam in braided channel (2-26)	3/25/09	15	1	0
	# Total	**52**	**26**	**0**
TRIBUTARIES				
Hunter Creek (2-39)	1/28/09	20	3	0
McTaggart Creek (2-64)	2/26/09	0	0	15
Swift Creek - upper (2-72)	3/03/09	15	0	0
Vance Creek - upper (2-41)	3/04/09	14	6	0
	# Total	**49**	**9**	**15**
BACKWATERS				
South Fork at Vance Creek confluence	2/02/09	5	15	0
South Fork - downstream of Vance Creek confluence	3/05/09	13	2	0
South Fork - downstream of Vance Creek confluence	3/10/09	13	4	0
North Fork - downstream of X (2-26)	3/25/09	15	0	0
North Fork - upstream of X (2-26)	3/25/09	15	0	0
	# Total	**61**	**21**	**0**
PONDS				
Outlet from mainstem to North Fork	2/05/09	16	0	0
Skokomish pond (SP-14)	2/19/09	18	0	0
Skokomish pond (SP-21)	2/23/09	15	8	3
Skokomish pond (SP-6/22)	2/24/09	15	0	0
	# Total	**64**	**8**	**3**
	Total Fish Collected	**226**	**64**	**18**
	% Total	**73**	**21**	**6**

Water quality characteristics were similar for all habitats (Table 2). The site temperatures in the mainstem, tributary, and backwater habitats ranged between 4.0 – 7.0°C and were colder than ponds, which ranged from 5.8 – 9.2 °C. Dissolved oxygen (DO) levels at the mainstem, tributary, and backwater sites ranged from 10.3 – 15.6 mg/L and were higher than ponds, which ranged from 7.23 – 9.0 mg/L. The pond site called “outlet from mainstem to the North Fork” was physically different than the other three pond sites, because it was smaller, and was closely connected to the mainstem; the other three pond sites were relatively larger bodies of water, with lower flow velocities and were more appropriately categorized as off-channel habitat. The pond outlet from mainstem to the North Fork had D.O. of 11.03, which was very similar to the other habitat types with faster moving water.

Table 2.-Water quality and habitat data from the sites where fish were collected for diet analysis. The numbers in parentheses after sample locations are site identification numbers. The centers of the site transects are labeled "X"; transects were ten times the bank full width, or 150 m, whichever was greater. Values collected were turbidity (Turb., ntu), dissolved oxygen (D.O., mg/L), water temperature (°C), pH, GPS coordinates (NAD 83) (X LAT, X LONG), GPS accuracy (Acc., +/- m), bank full widths (BFW, meters), and reach lengths (meters). Dashes indicate values which were not collected.

Habitat type and sample site	**Turb. ntu**	**D.O. mg/L**	**°C**	**pH**	**X LAT**	**X LONG**	**Acc. +/-m**	**BFW**	**Reach Length**
MAINSTEM									
Mainstem (2-31)	-	-	7.0	-	485432	5241226	-	-	-
South Fork at Vance Creek confluence	1.23	15.40	4.0	7.44	-	-	-	-	-
North Fork at mainstem confluence	3.50	10.66	5.0	6.78	483355	5240897	6.0	17.1	150
South Fork above and below North Fork confluence	1.40	11.70	4.0	7.50	483442	5240795	6.0	53.1	531
North Fork mainstem at log jam in braided channel (2-26)	3.40	15.58	5.9	7.60	482343	5244545	9.7	15.0	150
TRIBUTARIES									
Hunter Creek (2-39)	-	-	7.0	-	483916	5239798	11.2	12.3	150
McTaggart Creek (2-64)	0.56	10.30	6.3	6.36	481959	5250255	13.5	3.0	150
Swift Creek - upper (2-72)	-	11.47	7.1	7.24	482812	5248130	22.0	8.4	150
Vance Creek - upper (2-41)	0.91	11.36	6.0	-	477388	5241632	21.0	30.0	300
BACKWATERS									
South Fork at Vance Creek confluence	1.23	15.40	4.5	7.44	-	-	-	-	-
South Fork - downstream of Vance Creek confluence	0.91	11.36	6.9	-	480833	5240387	21.0	30.0	300
South Fork - downstream of Vance Creek confluence	1.23	15.40	6.4	7.44	-	-	-	-	-
North Fork - downstream of X (2-26)	3.40	15.58	5.9	7.60	482343	5244545	9.7	15.0	150
North Fork - upstream of X (2-26)	3.40	15.58	5.9	7.60	482343	5244545	9.7	15.0	150
PONDS									
Outlet from mainstem to the North Fork	0.93	11.03	5.8	7.44	481832	5240230	5.5	-	700
Skokomish pond (SP-14)	-	-	-	-	487715	5239661	-	-	150
Skokomish pond (SP-21)	1.95	7.23	9.2	6.90	488188	5238991	-	-	150
Skokomish pond (SP-6)	-	9.00	8.5	6.25	487627	5240087	-	-	150
Skokomish pond (SP-22)	-	9.00	8.5	6.25	487613	5240130	-	-	150

Sample size

According to Pielou's method only coho salmon were collected in sufficient quantity to claim an adequate sample size was collected across all four of the habitat types (Figure 3). See Appendices C and D for the results of Pielou's methods on the original sample sets of all fish collected, including fish greater than 100 mm FL. After stomach samples collected from fish greater than 100 mm FL were excluded from the analysis, Pielou's method was re-calculated on the remaining samples (Figures 3 and 4).

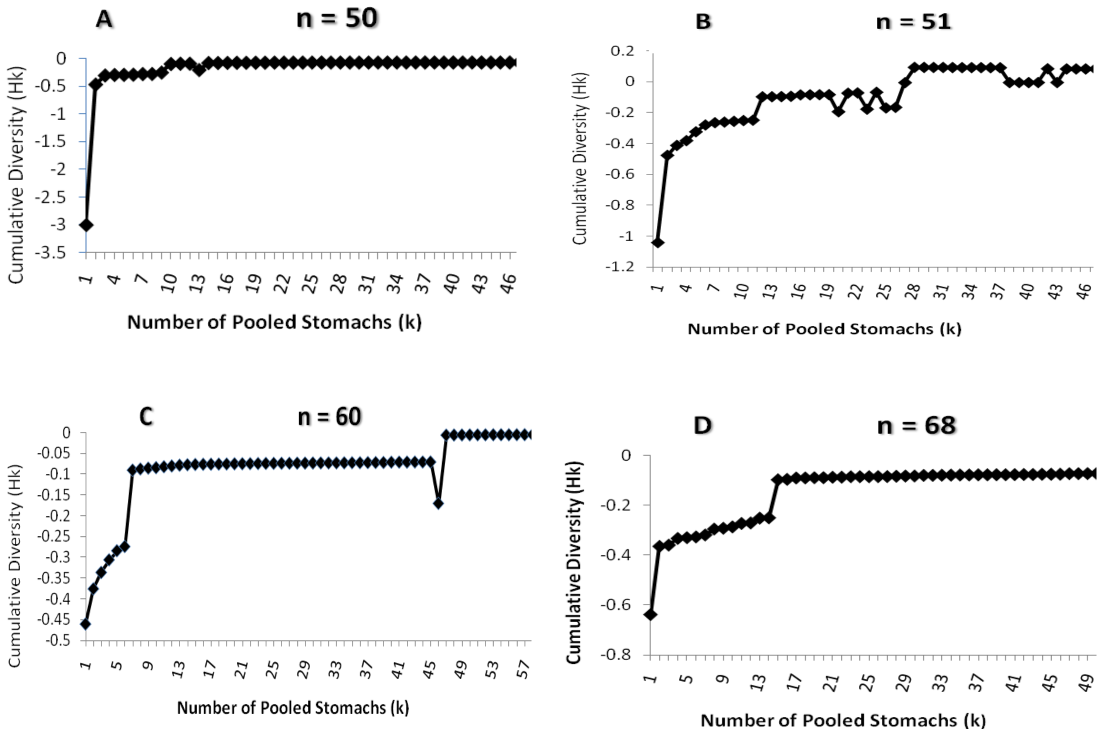


Figure 3.-Results of Pielou's method to determine adequate sample size within each of the habitat types for coho salmon; (A) mainstem, (B) tributaries, (C) backwaters, and (D) ponds. Sample sizes are indicated at the top of each graph (n). Values for cumulative diversity (Hk) initially start out low, then increase and level off when adequate sample size has been reached (Hoffman 1978).

Steelhead and cutthroat trout were also collected, but sample sizes were too small from each habitat (Figure 4 B) to perform the same analysis performed for coho salmon. Because the trout sample sizes were too small for each of habitat types in which they were collected, it was not possible to estimate the diet characteristics for these species in all habitat types. Adequate sample sizes for steelhead were only collected in mainstem and backwaters based on Pielou's method (Figure 4 A and C). An adequate sample size was collected for cutthroat trout in tributaries (Figure 4 D).

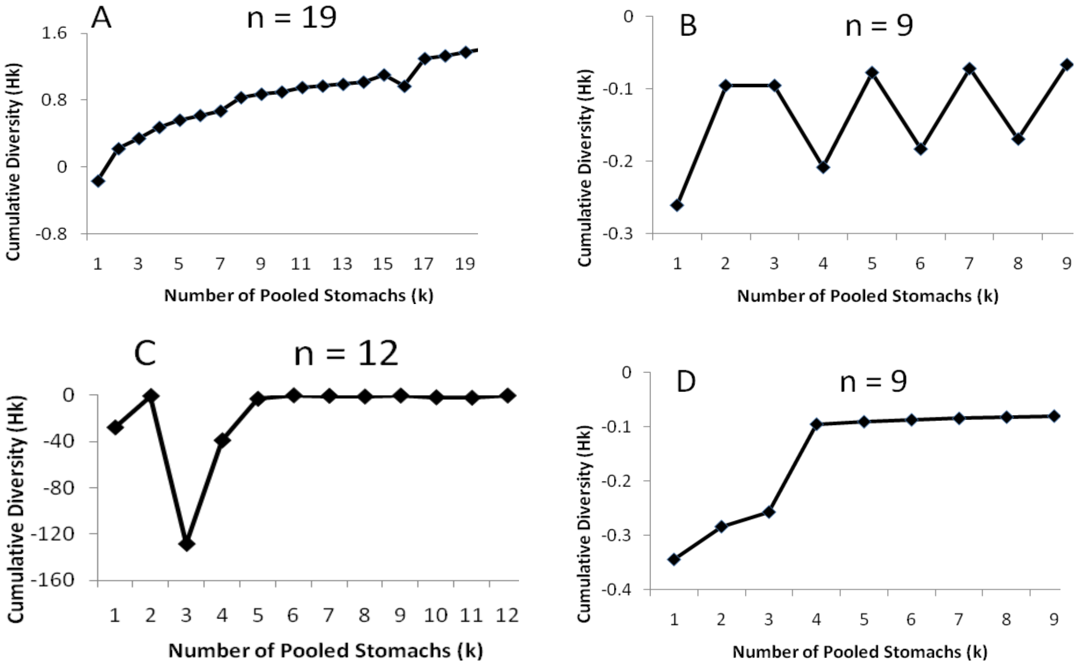


Figure 4.-Results of Pielou's method to determine adequate sample size within each of the habitat types. Steelhead in panel (A) mainstem, (B) tributaries, and (C) backwaters, and cutthroat trout in (D) tributaries. Sample sizes are indicated at the top of each graph (n). Values for cumulative diversity (Hk) initially start out low, then increase and level off when adequate sample size has been reached (Hoffman 1978). Steelhead in tributaries (B) was not used in analyses because these results indicate the sample size was too small.

Diet composition - Prey Abundance and Importance

Coho salmon fed primarily on benthic macroinvertebrates; aquatic insect nymphs or larvae which comprised a mean 48% by weight and 94% by number for all habitat types combined (Table 3). Coho salmon diets contained comparatively low proportions of exuvia and prey items categorized as “Other”. The “Other” category is comprised of prey items that had relatively low frequencies of occurrence. Steelhead and cutthroat trout also fed primarily on aquatic insect nymph and larvae and consumed much higher proportions (by weight) of prey items categorized as “Other”.

Table 3.-Percent proportion by weight (%Wi), and by number (%Ni) of aquatic and terrestrial insects. The “Other” categories are comprised of varying combinations of Arachnida, Amphipoda, Collembola, Copepoda, fish eggs, Hemiptera, Homoptera, Hydracarina, Hymenoptera, Lepidoptera, Mollusca, plant material, and unidentified organic matter.

	Coho salmon				**Steelhead**		**Cutthroat trout**
% Proportion by weight	**main-stem**	**tributaries**	**backwaters**	**tributaries**	**main-stem**	**backwaters**	**tributaries**
Aquatic insects	58.88	68.56	75.99	73.08	70.22	62.73	73.08
Terrestrial insects	19.33	26.89	15.19	4.44	0.00	0.10	4.44
Exuvia	9.04	0.04	3.27	0.04	2.24	1.45	0.04
Other	12.75	4.51	5.55	22.44	27.54	35.72	22.44
% Proportion by number							
Aquatic insects	97.11	96.68	97.98	98.31	94.59	99.69	98.31
Terrestrial insects	2.74	3.09	1.91	1.69	0.00	0.31	1.69
Other	5.63	6.41	3.93	3.38	5.41	0.00	3.38

Ephemeroptera comprised over 23% (by weight) of the overall salmonid diet, with almost 18% of that being the family Baetidae (Appendix E). While Ephemeroptera, Trichoptera, and Plecoptera were present in comparatively lower proportions (by weight), the frequency of occurrence for these prey items was consistently high for coho salmon in all habitats. Oligochaeta comprised nearly 15% (by weight) of the overall fish diets, while Plecoptera was notable at 9% and Trichoptera was nearly 6%.

Prey categories important to coho salmon (by weight) were Ephemeroptera, Chironomidae, Plecoptera, Trichoptera, Megaloptera, and Oligochaeta (Figure 5 and Appendix F). Ephemeroptera had the highest proportions (by weight) of all other prey items, which occurred in backwaters (44%). Ephemeroptera was also the prey item that was the highest proportions (by weight) in the mainstem (17%). Chironomidae was the most important prey item in tributaries (39%). For ponds, the prey item in highest proportions (by weight) was Oligochaeta (32%) and Megaloptera (17%). Megaloptera and Oligochaeta are comparably large prey items, which contributes to their high proportions by weight; they have low frequencies of occurrence, but when found Megaloptera appear to be of great importance (by weight) in ponds, and Oligochaeta (by weight) in all habitats.

Several prey items had relatively high frequency of occurrence values and for coho salmon, those prey items were Chironomidae, Diptera larvae, Ephemeroptera, Plecoptera, Trichoptera, Megaloptera, Coleoptera, and Crustacea (Appendix F). Chironomidae were a very frequently occurring prey item, with a frequency of occurrence of 84% for coho salmon in tributaries and backwaters, and over 48% in mainstem and ponds. Diptera larvae were also highly frequent, with highest frequency of occurrence values in the mainstem (42%) and lowest in ponds (15%). Ephemeroptera followed a similar trend being most frequently occurring in the mainstem (76%) and the least frequent in ponds (47%). Plecoptera and Trichoptera were occurred less frequently and values were highest in backwaters (51% and 43%) and lowest in ponds (25% and

13%), respectively. Coleoptera frequency of occurrence was highest in tributaries (35%) and lowest in ponds (6%). Crustacea and Mollusca were less frequently occurring, but most frequently occurred in ponds.

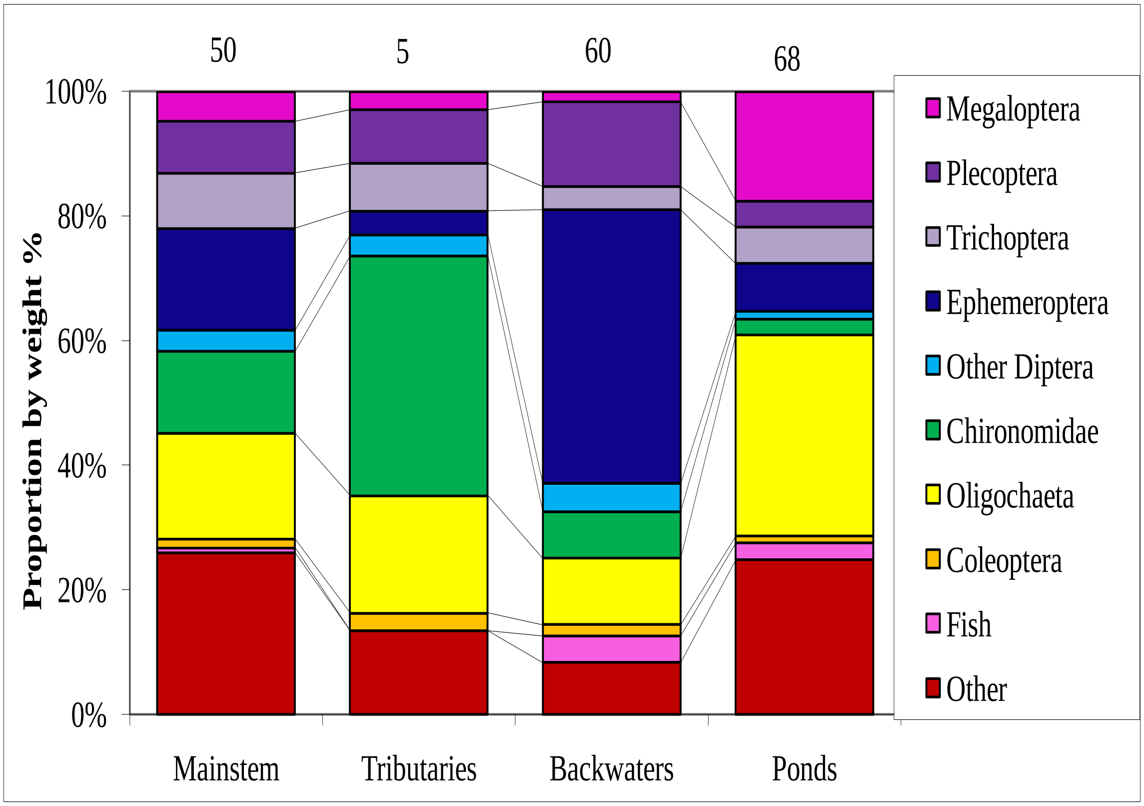


Figure 5.-Percent proportion by weight (%Wi) of major prey categories present in coho salmon diets, lower Skokomish River, January through March 2009. Samples sizes are listed at the top of the graph. The "Other" category includes a mixture of Arachnida, Amphipoda, Collembola, Copepoda, fish eggs, Hemiptera, Hydracarina, Hymenoptera, Lepidoptera, Mollusca, plant material, and unidentified organic matter.

Megaloptera had a mean frequency of occurrence value of 9.4% and it comprised almost 5% of the coho salmon diet (by weight). Frequency of Megaloptera was highest in ponds at 17.5 (%Oi) and lowest in the mainstem at 4.8 (%Oi) (Appendix F). Diptera occurred in every habitat type in this diet study and most frequently occurred in the mainstem (42%), and least frequently occurred in ponds (14%). Diptera larvae had consistently higher frequency of occurrence values for coho in all habitats, and for cutthroat trout in tributaries. The Diptera families present in the diets of these fish were Dixidae, Ceratopogonidae, Simuliidae, and Chironomidae; together they comprise 3.3% of the overall diets (by weight), of which the family Simuliidae comprised the majority at 2.3% (by weight).

For coho salmon in tributaries, Chironomidae comprised a substantial proportion (by weight) in Vance Creek (47%). Trichoptera proportions (by weight) were only slightly higher in Hunter Creek and Swift Creek combined (15%) than Vance Creek (9%). Oligochaeta comprised substantial proportions (by weight) in Swift Creek (34%). All other prey categories were fairly similar between creeks.

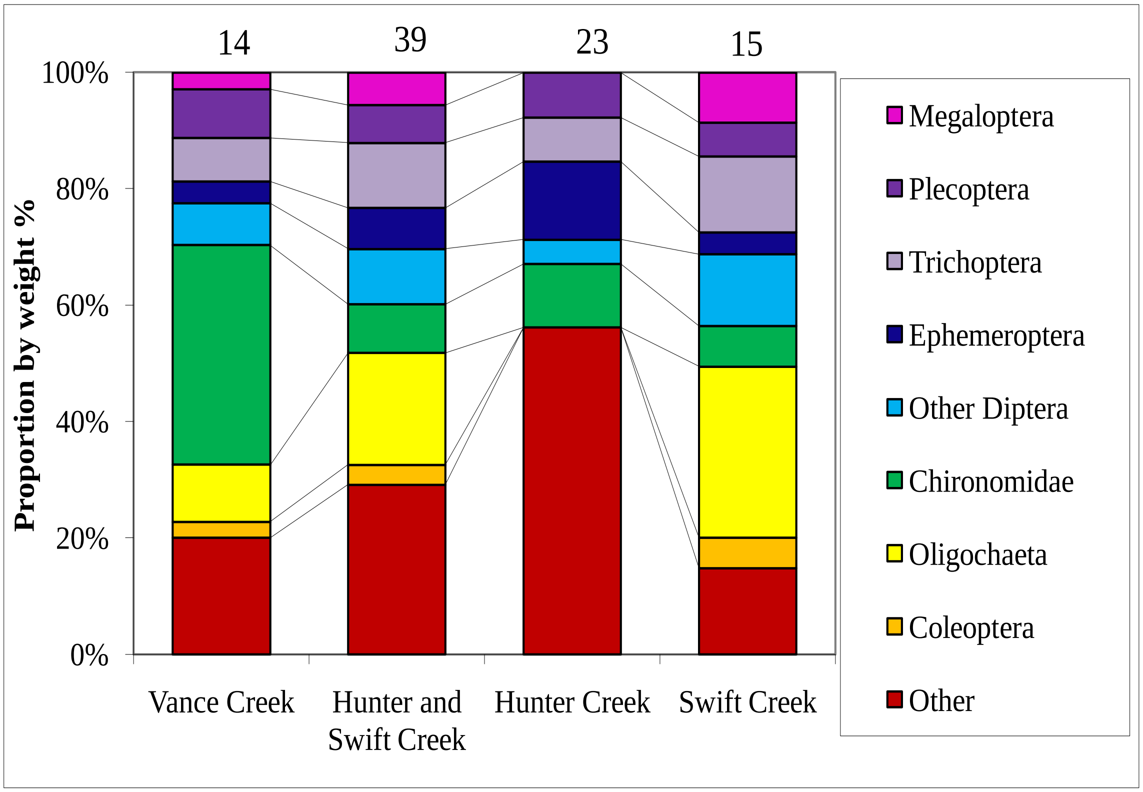


Figure 6.-Percent proportion by weight (%Wi) of major prey categories present in coho stomachs in tributaries of the lower Skokomish River, January through March 2009. Samples sizes are listed at top of graph. The “Other” category includes a mixture of Arachnida, Amphipoda, Collembola, Copepoda, fish eggs, Hemiptera, Hydracarina, Hymenoptera, Lepidoptera, Mollusca, plant material, and unidentified organic matter.

For steelhead and cutthroat trout items which were of obvious importance (by weight) to trout were Ephemeroptera in the backwaters (34%) and Plecoptera (28%) and Trichoptera (30%) in tributaries (Appendix G). Plecoptera and Trichoptera were observed in the greatest proportions (by weight) in trout from the mainstem (27% and 30% respectively).

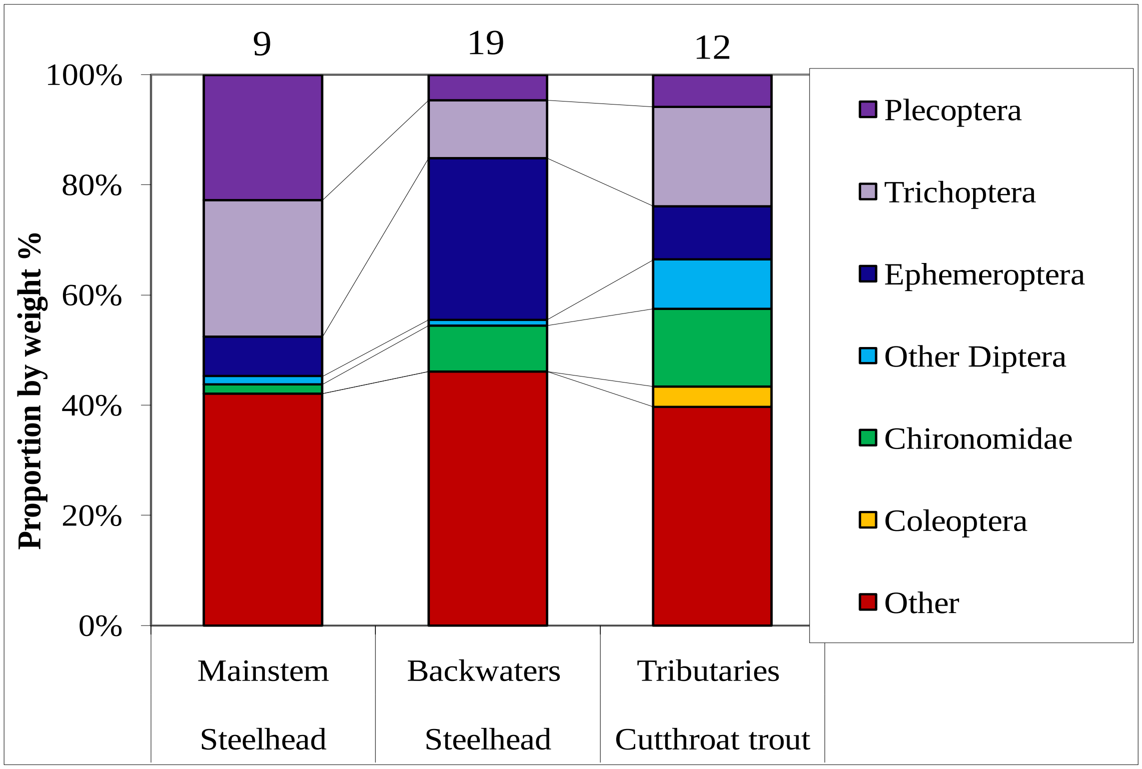


Figure 7.-Percent proportion by weight (%Wi) of the major prey categories present in cutthroat trout and steelhead, lower Skokomish River, January through March 2009. Samples sizes are listed at the top of the graph. The "Other" category includes a mixture of Arachnida, Collembola, Copepoda, Homoptera, Hydracarina, Mollusca, Lepidoptera, plant material, and unidentified organic matter.

For steelhead and cutthroat trout the prey categories Chironomidae, Diptera larvae, Ephemeroptera, Plecoptera, Trichoptera, and Coleoptera had the highest frequency of occurrence values between all habitats (Appendix G). Chironomidae most frequently occurred in backwaters (75% and 89%) and the least frequently occurred in the mainstem (20%) (Appendix G). For trout, the frequencies of occurrence values (of prey items present in appreciable amounts) were highest in tributaries and backwaters, and lowest in the mainstem (Appendix G). Large amounts of exuvia indicates they are drift feeding (Tippets and Moyle 1978), and for these trout the proportions of exuvia were less than 10 percent of any of their diets across all habitats, indicating that they were mainly foraging in benthic areas (Table 3). For trout in this diet study, the prey items of most importance (by weight) were Trichoptera, Plecoptera, Ephemeroptera, Chironomidae, and Diptera larvae (Appendix G and Figure 7).

The prey items with the highest importance (%IRI) values for coho salmon were Chironomidae and Ephemeroptera (Figure 8 and Appendix H). Chironomidae was the most important prey item (%IRI ranged 32% – 49%) for all fish in all habitats, except the mainstem. Ephemeroptera was the prey item of highest importance (%IRI) for coho salmon in the mainstem (23%). Trichoptera was the prey item of highest importance (%IRI) in the mainstem for steelhead (29%). Chironomidae %IRI for coho salmon was highest in tributaries at 49%, and lowest in the mainstem at 18%. The importance of Chironomidae jumps to near 50% while Ephemeroptera importance drops to 3% for coho salmon in tributaries (Figure 8).

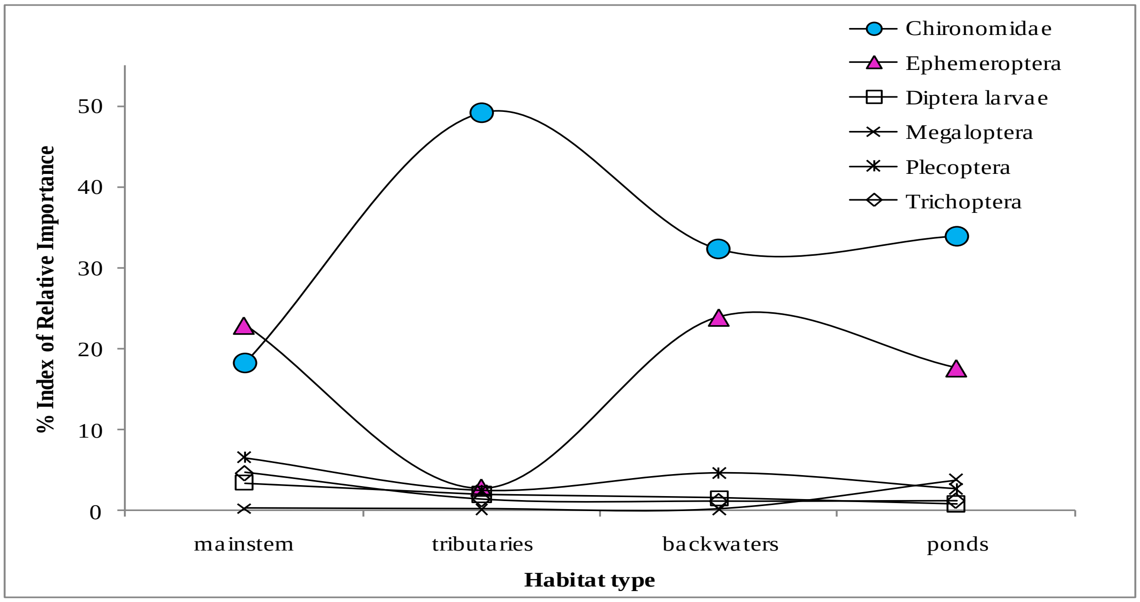
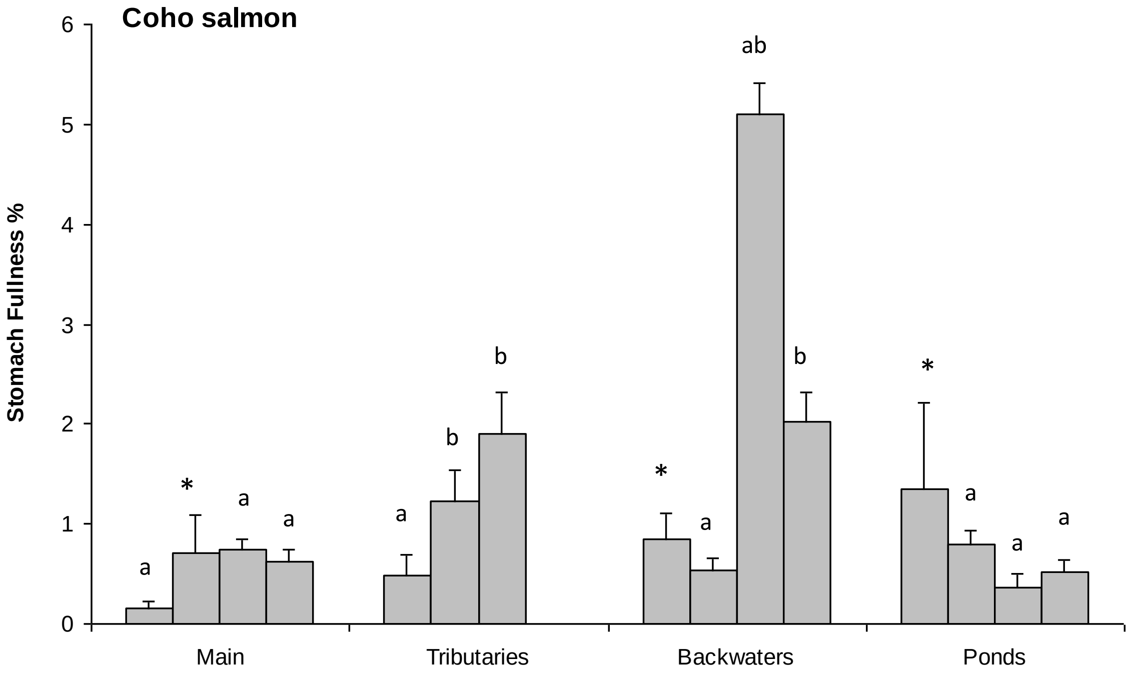


Figure 8.-Percent index of relative importance of major prey items for coho salmon, in the Skokomish River, January through March 2009.

Stomach Fullness - Diet Breadth – Diet Overlap

Mean stomach fullness values for coho salmon were not significantly different between habitat types. However, there were significant differences for coho salmon stomach fullness within tributary and backwater sites (Figure 9). Coho salmon mean stomach fullness at the tributary site Swift Creek were significantly higher than Hunter Creek ($P = 0.004$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$). Coho salmon mean stomach fullness at the tributary site Vance Creek were also significantly higher than Hunter Creek ($P = 0.0001$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$). Coho salmon mean stomach fullness at the South Fork backwater site downstream of the Vance Creek confluence were significantly higher than the North Fork backwater site 2-26, upstream of X (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$). Stomach fullness values were highest for coho salmon in backwaters at 5%, and were the lowest in the mainstem sites (Figure 9).



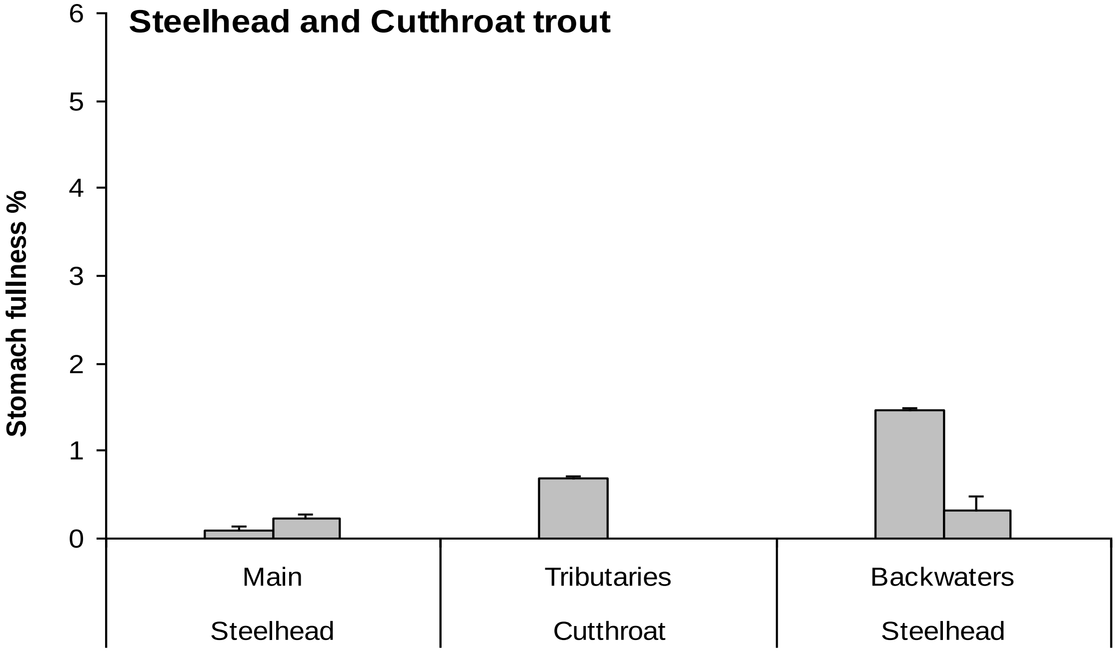


Figure 9.-Mean percent stomach fullness for coho salmon, steelhead, and cutthroat trout (+ standard error), in the Skokomish River, January through March 2009. Each bar represents a different sample site. Within each habitat type, bars with different letters are significantly different (nested ANOVA and Tukey's HSD; $P < 0.05$). *Bars with no letters were sites not statistically compared.

Diet breadth were not significantly different between habitat types (ANOVA; *P* = 0.05). Coho salmon had highest diet breadth in the mainstem and ponds (Figure 10). Tributaries provided the second highest diet breadth values for coho salmon and cutthroat trout diets. Diet breadth levels ranged overall from approximately 2 to 10. Backwaters provided the lowest diet breadth values for coho salmon and trout. The mainstem appears to be providing the highest overall opportunities for diet diversity.

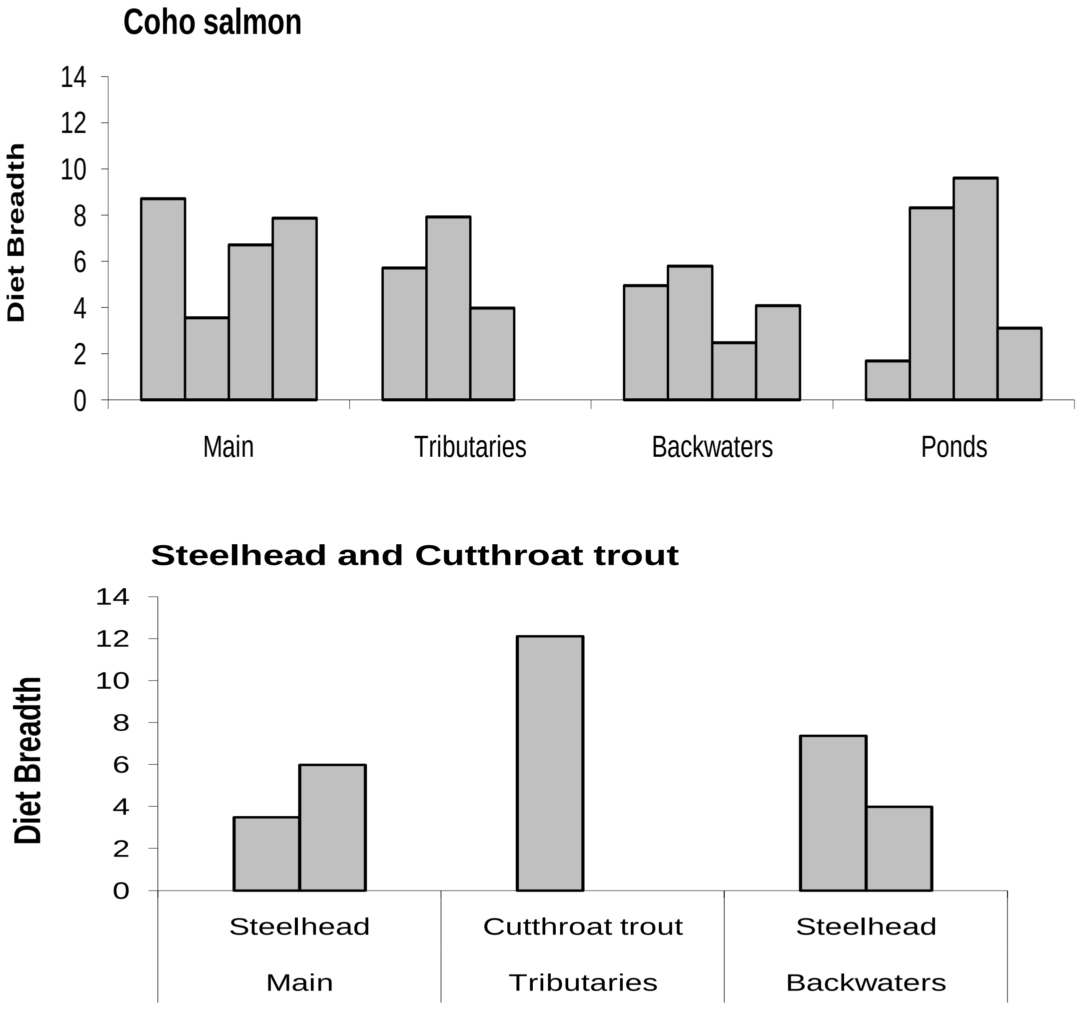


Figure 10.-Results of diet breadth determinations for coho salmon, steelhead, and cutthroat trout, in the Skokomish River, January through March 2009. Index values range between 1 (one prey type present, i.e. there is no diet breadth) and infinity. Values less than two indicate little diet breadth (Tabor et al. 2001). Each bar represents a different sample site.

Approximately 44% of the salmonid diets exhibited overlap (4 out of 9) according to Horn's diet overlap index (Table 4). Coho salmon exhibited some diet overlap in mainstem and ponds and to a lesser degree in mainstem and tributaries. The highest diet overlap occurred between coho salmon and steelhead in backwaters and they also overlapped to a lesser extent in the mainstem. No diet overlaps occurred between coho salmon and cutthroat trout (Table 4). The remaining species and habitat types showed no presence of diet overlap.

Table 4.-Results of Horn's index of overlap for coho salmon, steelhead, and cutthroat trout, in the Skokomish River, January through March 2009. Values greater than 0.60 are considered to have diet overlap, and are totally overlapped with values equal to 1. Each habitat type represents all the sample sites combined.

Species	Habitat	Overlap?	Species	Habitat	Horn's Index
Coho	**backwaters**	**Yes**	**Steelhead**	**backwaters**	**0.786**
Coho	**mainstem**	**Yes**	**Coho**	**ponds**	**0.775**
Coho	**mainstem**	**Yes**	**Coho**	**tributaries**	**0.618**
Coho	**mainstem**	**Yes**	**Steelhead**	**mainstem**	**0.608**
Coho	tributaries	No	Cutthroat	tributaries	0.581
Coho	backwaters	No	Coho	mainstem	0.560
Coho	tributaries	No	Coho	ponds	0.448
Coho	backwaters	No	Coho	tributaries	0.335
Coho	backwaters	No	Coho	ponds	0.258

Fish size and condition

Mean weights for coho salmon were not significantly different between habitat types. However, there were significant differences for coho salmon weights within mainstem sites, backwater sites, and pond sites. Coho salmon weights at mainstem site 2-31 were significantly higher than the South Fork (at the North Fork confluence) (P = 0.002) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 11). Coho salmon mean weights at the backwater site on the South Fork downstream of the

Vance Creek confluence were significantly higher than in the North Fork backwater site 2-26 (*P* = 0.013) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 11). Coho salmon mean weights at Skokomish pond site 6-22 were significantly higher than those in Skokomish Pond 14 ($P < 0.0001$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 11). Coho salmon mean weights at Skokomish pond site 6-22 were significantly higher than those in Skokomish Pond 21 (*P* = 0.005) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 11).

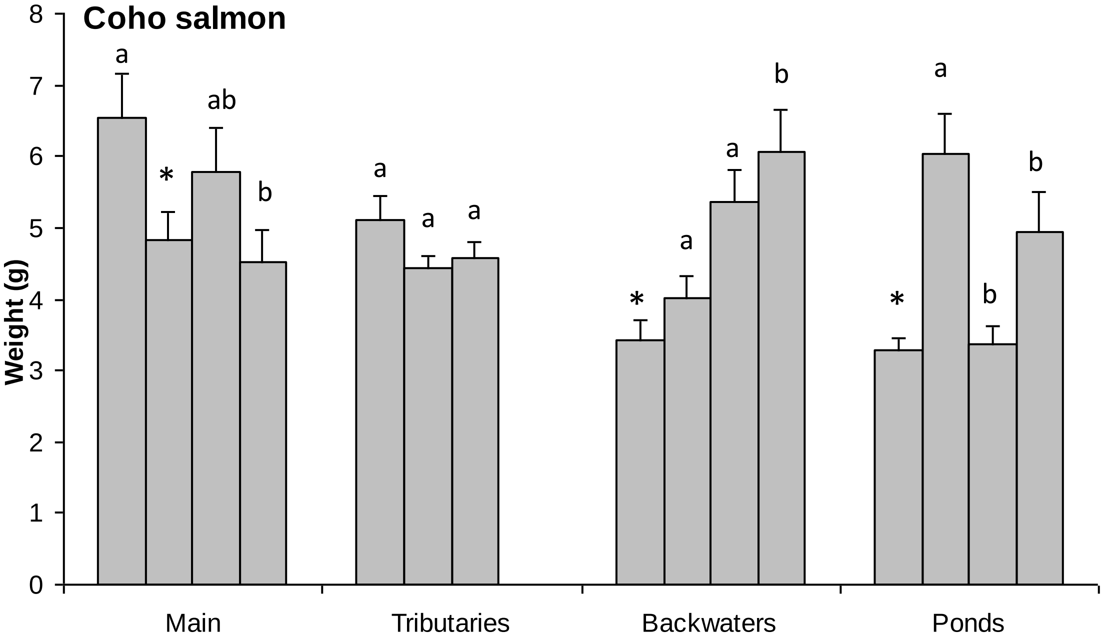


Figure 11.-Mean weights for coho salmon (+ standard error) in four habitat types analyzed in the diet analysis, lower Skokomish River, Washington. Each bar represents a different sample site. There were no significant differences in mean weights between habitat types (nested ANOVA).   Within each habitat type, bars with different letters are significantly different (nested ANOVA and Tukey's HSD; $P < 0.05$). *Bars with no letters were sites not statistically compared.

Coho salmon mean FL's were not significantly different between habitat types. However, there were significant differences for coho salmon FL within mainstem sites, backwater sites, and pond sites. Coho salmon mean FL's at the mainstem site 2-31 were significantly higher than the South Fork (at the North Fork confluence) ($P = 0.008$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 12). Coho salmon mean FL's at the backwater site on the South Fork, downstream of the Vance Creek confluence, were significantly higher than in the North Fork backwater site 2-26 ($P = 0.041$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 12). Coho salmon mean FL's on Skokomish pond site 6-22 were significantly higher than those in Skokomish Pond 14 ($P = 0.0001$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 12). Coho salmon mean FL's on Skokomish pond site 6-22 mean weights for coho salmon were significantly higher than those in Skokomish Pond 21 ($P = 0.024$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 12).

The sites which contained the coho salmon with the greatest mean FL were main channel #2-31, the South Fork backwater downstream of Vance Creek confluence, and Skokomish Pond 6-22. Fish greater than 100 - mm FL were eliminated from any analyses because the frequency distributions indicated there were two size classes present (Appendix B). There were too few greater than 100 mm FL to analyze a second size class.

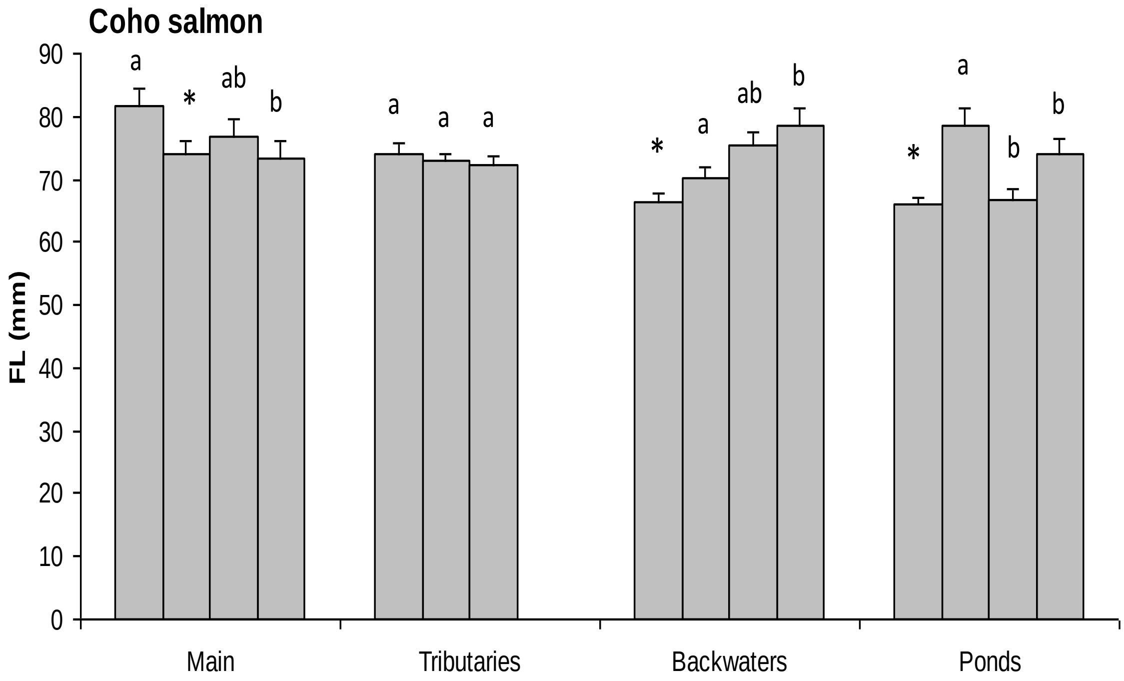


Figure 12.-Mean FL for coho salmon (+ standard error) in the four habitat types analyzed in the diet analysis, lower Skokomish River, Washington. Each bar represents a different sample site. There were no significant differences in mean FL between habitat types (nested ANOVA). Within each habitat type, bars with different letters are significantly different (nested ANOVA and Tukey's HSD; $P < 0.05$). *Bars with no letters were sites not statistically compared.

Mean condition factor values for coho salmon were not significantly different between habitat types. However, there were significant differences in the mean condition factor values for coho salmon within mainstem sites, tributary sites, and pond sites. Main channel site 2-31 mean condition factor values for coho salmon were significantly higher than values for the South Fork (at the North Fork confluence) ($P = 0.005$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 13). Tributary sites, Hunter Creek and Vance Creek, mean condition factor values for coho salmon were significantly higher than Swift Creek ($P = 0.002$ and $P = 0.012$, respectively) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$) (Figure 13). Skokomish pond site 6-22 mean condition factor values for coho salmon were significantly higher than those in Skokomish Pond 14 ($P = 0.021$) (nested ANOVA and multiple comparison

Tukey's HSD; $P < 0.05$) (Figure 13). The sites which contained coho salmon with the highest condition factor values were: the North Fork mainstem site #2-26, which was mainly comprised of a very large log jam; tributary sites Hunter Creek and Vance Creek which were very similar in that they did not have their own watersheds and were in the lower flood plain portion of the watershed; the two backwater sites downstream of Vance confluence; Skokomish pond sites 6-22 and 21, which were very similar because they were quite large, relatively shallow ponds, with some presence of deeper pool areas.

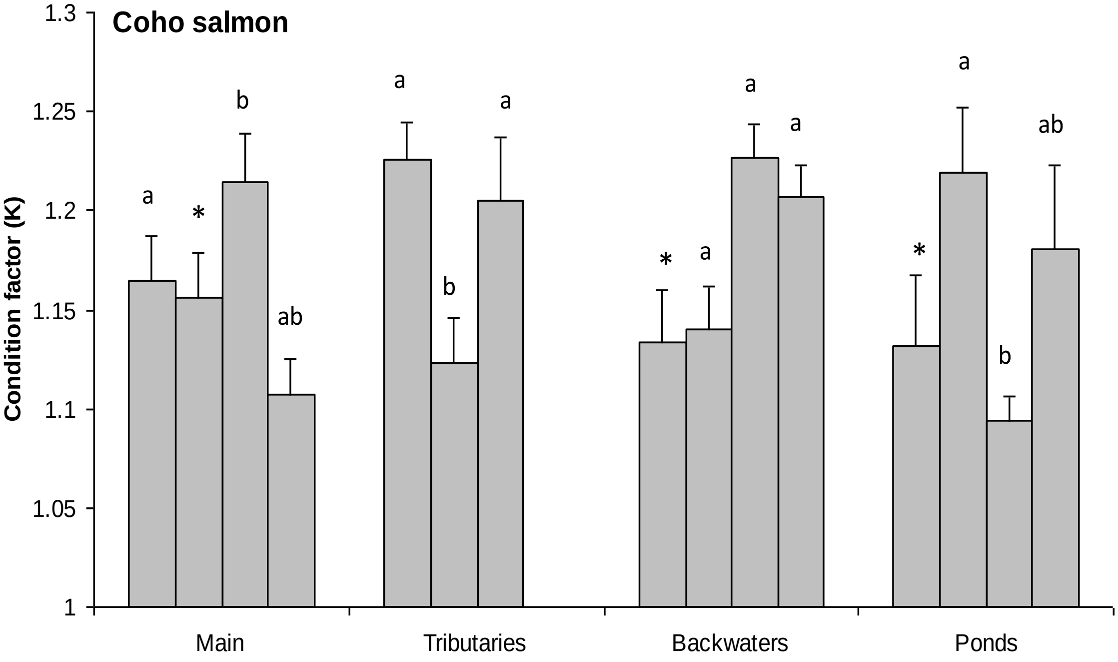


Figure 13.-Mean condition factor for coho salmon, cutthroat trout, and steelhead (+ standard error), lower Skokomish River, January through March 2009. Each bar represents a different sample site. There were no significant differences in mean condition factor between habitat types (nested ANOVA). Within each habitat type, bars with different letters are significantly different (nested ANOVA and Tukey's HSD; $P < 0.05$). *Bars with no letters were sites not statistically compared.

Steelhead mean weights were slightly higher in the mainstem compared to backwaters and cutthroat trout in tributaries (Figure 14). The mean FL and condition factors did not vary much between cutthroat trout and steelhead. Sample sizes for these collections were too small to perform equal sample size nested ANOVA.

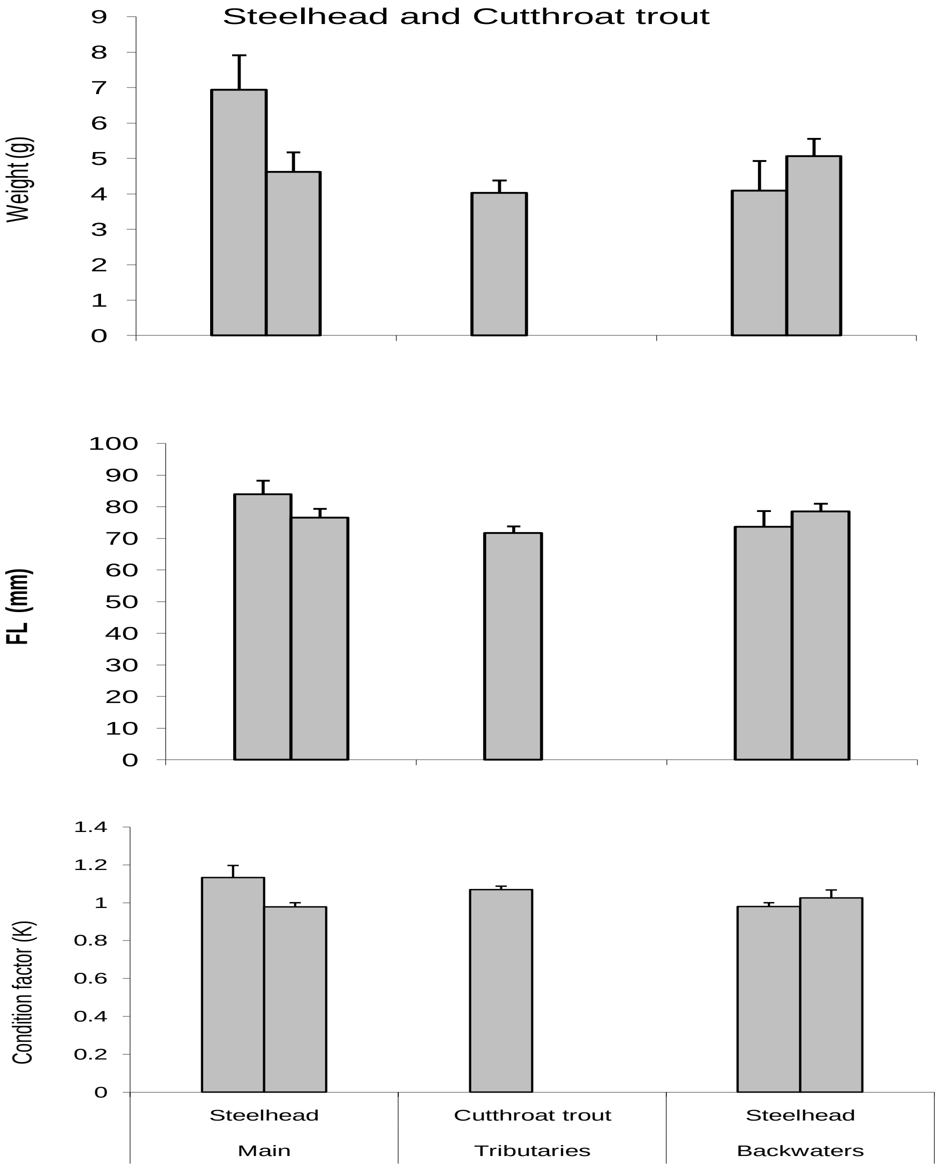


Figure 14-Mean weight, FL, and condition factor for cutthroat trout and steelhead (+ standard error) lower Skokomish River, January through March 2009.

(i) **Discussion and Conclusions**

The winter diets of coho salmon, steelhead, and cutthroat trout consisted primarily of benthic macroinvertebrates (aquatic insect nymphs or larvae) and contained very few terrestrial insects or fish. Aquatic insects in Skokomish River fish were a mean of 65% by weight. The availability of terrestrial insect's in forest streams declines to nearly zero during winter, while aquatic insect biomass generally peaks from December to July (Nakano and Murakami 2001). This may explain why aquatic invertebrates are more important than terrestrial invertebrates in the diets of Skokomish River fish, and also for brook trout (*Salvelinus fontinalis*) and brown trout (*Salmo trutta*) in Canadian streams during the winter (Cunjak and Power 1987). The quantities of aquatic insects ingested by Skokomish River juvenile fish were low in comparison to other systems. For example, aquatic insects comprised approximately 75% of juvenile coho salmon diets during early March in three northern California streams (Gonzales 2006). The diets of Skokomish River fish are consistent with expected winter food availability; the result of this diet analysis demonstrates that these fish are eating the most abundant food source, benthic macroinvertebrates.

Aquatic insects in the Skokomish River fish were 97% (by count). Proportions by number values are not used to discuss prey importance in this diet study because they are not reliable indicators of diet characteristics and do not accurately represent the diet. Fish could consume large quantities of prey items which could comprise relatively low caloric content (i.e. Chironomidae larvae). If prey importance were based on proportions by numbers of these types of prey items it would appear the fish's diets were acquiring copious amounts of food, which is not the case based on the results of this diet study.

The composition of a fish's diet tends to represent prey availability (Cada et al. 1986). Invertebrate prey has been shown to dominate steelhead and cutthroat trout diets because it is usually abundant (Milick 1977; Casne 1975) and takes the least amount of energy to obtain. Because the diets of the fish in this diet study primarily consist of benthic macroinvertebrates, it is likely that they are the most abundant prey items available during the winter months in these areas of the Skokomish River. During the winter many terrestrial (i.e., Trichoptera, Ephemeroptera, Diptera, Plecoptera, Coleoptera and Megaloptera) insects are in larval or transitional stages and generally occupy a niche in littoral zones or benthic areas (McCafferty 1981). Ephemeroptera and Chironomidae are the prey items of most importance to the diets of these juvenile salmonids, with Plecoptera and Trichoptera following closely behind (Appendix H).

Compound indices like percent index of relative importance (%IRI) provide a more balanced view of a fish's diet (Chipps and Garvey 2007). For coho salmon, Chironomidae had highest importance values in tributaries (49%), and lowest values in the mainstem (18%), while the reverse was true for Ephemeroptera (Appendix H). The importance of all other major prey types remained relatively similar across habitat types. Ephemeroptera and Chironomidae are very similar in the important roles they play in the diets of many fish species; all stages are important to fish diets (McCafferty 1981). In tributaries, there was a notable decline in the importance of Ephemeroptera with a simultaneous increase of Chironomidae importance (Figure 8). This could indicate that either the environment was more suitable for Chironomidae, or that the fish were selecting for Chironomidae and not Ephemeroptera.

Chironomidae larvae are the most widely adapted family of Diptera and are likely to be found in almost all inland waters (McCafferty 1981). Chironomidae and Simuliidae were present in virtually all habitats and fish sampled in this diet analysis. Chironomidae had the highest importance values in tributaries, backwaters, and ponds. Chironomidae were more abundant in tributaries for coho salmon (~39 %) and cutthroat trout (~7 %) in terms of proportions by weight (Figure 5 and 7). Chironomidae may have high importance in these fish's diets because they were more available, exist in relatively high densities, and may be easier to capture compared to other more mobile prey items.

Diet composition and stomach fullness can change dramatically over a 24-h period, and similar research shows that dusk samples capture peak stomach fullness values and provide the most representative diet samples (Beauchamp et al. 2007). The mean stomach fullness of Skokomish River juvenile salmonids was less than 20% for all fish, and these fish were collected between dusk and midnight. These stomach fullness values are lower than similar winter studies in which the mean stomach fullness ranged between 50% - 67% during winter (Cunjak and Power 1987). Winter temperatures slow evacuation rates and have been shown to limit brook trout to single daily stomach filling and emptying in southern Ontario, Canada (Cunjak et al. 1987).

Coho salmon in backwaters of the Skokomish River had the highest levels of stomach fullness at near 5% (Figure 9). In backwaters, high stomach fullness values could indicate that less effort can be extended while acquiring greater amounts of food there, however, that is assuming the fish captured were acquiring this food in habitats in which they were captured. Alternately, the high stomach fullness values in backwaters could suggest that the fish are utilizing these areas after feeding to rest and maintain low metabolic rates while they digest their food. Coho salmon mean stomach fullness in the backwater site on the South Fork, downstream of Vance Creek confluence, were significantly higher than the North Fork backwater site 2-26, upstream of X ($P < 0.05$) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$).

Coho salmon in the tributaries Vance Creek and Swift Creek had significantly higher stomach fullness values than Hunter Creek ($P = 0.0001$, and $P = 0.004$, respectively) (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$). Hunter Creek and Swift Creek occupy the lowland flood plain area, and do not have their own steep drainage area and watershed like Vance Creek does. However, Hunter Creek likely provides lesser opportunity to forage feed due to the invasive species which occupy the majority of its edge areas. While tributaries, backwaters, and ponds provided the highest values for stomach fullness they offered the lowest values of diet breadth (Figure 10).

Low diet breadth values imply that these habitat types offer limited levels of prey diversity or that the fish have selective diets during the winter months, January through March. Diet breadth values were highest for coho salmon in the mainstem, followed by tributaries and ponds; backwaters offered the lowest values (Figure 10); suggesting that mainstem and tributaries may offer more diet diversity. Sample sizes were relatively similar for coho across all four habitat types; therefore, it is unlikely that diet breadth is influenced by sample size. However, prey items were in larval/transitional stages, and were quite small; classification to more specific levels was not always possible and this may have caused diet breadth to be underestimated. Although backwaters and ponds lacked diversity compared to the mainstem and tributaries, they may compensate for it by providing higher levels of prey biomass. Higher abundance of prey biomass may be more valuable because fish growth rate is limited by food supply and consumption rate (Martin 1983).

Coho salmon diets overlapped between the mainstem and ponds and between the mainstem and tributaries (Table 4). This overlap suggests that the mainstem and tributaries, and the mainstem and ponds are providing similar levels of diet diversity. Significant levels of diet overlap are often attributed to higher abundance levels of invertebrate prey availability, especially when seasonal peaks in prey availability are occurring. The fact that little diet overlap occurred between other habitat types may be indicative of low prey abundance levels in this system, however, many prey items were too small to identify, and diet overlap values could have been underestimated. The low occurrence of diet overlap could also be attributed to the combination of winter conditions and overall poor habitat health because they can reduce the ability of these habitats to sustain stable forage food.

Overall, the health and well-being of coho salmon in this system appear to be relatively similar and typical for winter conditions. There were no significant differences in condition factor between habitat types (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$). The condition factor for coho salmon was similar between all habitat types, with a mean condition of 1.17 (Figure 13). Condition factor values for cutthroat trout and steelhead were also very similar, and didn't differ much between habitat types; however, both trout species had lower condition values than coho salmon. Similar research found that the condition of brook trout in winter was typically below 1 and that low condition factors in winter were not a function of reduced food availability or quality, but was instead due to the fish's inability to digest and assimilate more food

(Cunjak et al. 1987). A species specific formula that considers their differing growth patterns may have been appropriate to compare between coho salmon and trout condition. Condition factor is a measure of a fish's health, which is related to a fish's amount of energy stores, and small fish with lower lipid stores can exhaust their energy reserves earlier, experiencing high mortality rates sooner than larger fish with greater lipid content (Biro et al. 2004).

Although there were no significant differences in coho salmon mean weight and FL's between habitat types (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$), the mean weights and FL's in the mainstem were larger (Figures 11 and 12, respectively). Larger coho salmon mean weights and FL's in mainstem may suggest that that only the healthier, more fit, fish in the Skokomish River system can sustain activity in the mainstem, and are more capable of foraging in these areas of greater forage food diversity, although similar research suggests otherwise. Smolts originating from ponds have been shown to be generally larger than those from tributaries (Peterson and Reid 1984; Cederholm and Scarlett 1991). Peterson and Reid (1984) also found that small size may reflect the conditions of their past spring-summer rearing environment, and they could grow more rapidly when given a suitable environment (Chapman 1962; Mason 1969)

Overall, the mean FL of coho salmon in this diet study was slightly lower than typical means. Coho smolts in Washington State are usually a mean FL of 89 to 129 mm; smolting generally occurs in late spring (Wydowski and Whitney 2003). Coho salmon in winter California streams are generally a mean FL 73 mm in early March (mean 4.5 S.E.) (Gonzales 2006). The coho salmon in the Skokomish River were pre-smolts and averaged less than 76 mm FL across all habitat types (minimum 54 mm, maximum 100 mm, and median of 72 mm), and these values may have been biased by the fact that fish smaller than 54 mm were not sampled. These fish may reach mean average smolt sizes in this area by late spring, but further studies would be necessary to determine if this were occurring. The relatively small sizes of these juvenile salmonids could indicate poor habitat health year-round, an insufficient prey food base during winter, or that their growth may soon be enhanced when spring insects emerge. There were no significant differences in mean coho salmon FL between habitat types (nested ANOVA and multiple comparison Tukey's HSD; $P < 0.05$).

This study of the winter feeding ecology of juvenile salmonids in the lower Skokomish River provides small insight into a much larger ecological system. Substantial habitat degradation has occurred and is exhibited by the small size of the juvenile salmonids inhabiting it during the winter months. Because freshwater fish diets tend to shift primarily to benthic foraging in the colder weather, adequate supplies of aquatic insects and access to areas of refuge will be necessary to sustain them. Exuvia, discarded exoskeleton, is usually acquired through drift feeding, and relatively large amounts of it in the diet can indicate that drift feeding is the primary means of acquiring prey (Tippets and Moyle 1978), especially in the mainstem. These diets exhibited low amounts of exuvia, indicating that these fish are not primarily drift feeding and are relying on benthic foraging opportunities in the substrate. Therefore, maintaining healthy undisturbed habitat which can sustain juvenile salmonids and their prey base is critical to their survival in winter.

More accurate results could be produced if more sites were sampled in future diet studies. There was a lot of variability within the habitat types sampled in this diet study; as demonstrated by the figures for coho salmon weight, length, condition factor, and stomach fullness. Future research of this system should perform prey availability assessments in addition to diet sampling. Collecting multiple years of diet comparisons during the winter could also provide better descriptions of how sustainably these habitat types can provide food for juvenile salmonids over time. Adhering to a consistent collection time, between dusk and dawn, of stomach samples would prevent the possibility of obtaining stomach samples contain partially digested contents. And finally, larger sample sizes would provide better interpretive and comparative ability, ensuring the analyses were adequately representing the populations' characteristics.

(j) Management Implications and Habitat Recommendations

The conditions on the Skokomish River have been gradually accumulating since the 1800's and are not likely to be reversed in any short length of time. Conditions are continually accentuated by a combination of factors; extensive logging in the lower watershed; heavily introduced sediment from logging; reduction in sediment transport and consequential river bed aggradation; transformation of lowlands to agricultural areas; regularly frequent and increasing intensity of flooding; and the present installation of levees, dikes, and setbacks that intensify the effects of the flooding. Much more restoration is needed to provide better habitat for these fish and their food sources in these systems.

It is well established that healthier habitat is better equipped in sustaining over-wintering populations of juvenile salmonids. Pre-settlement characteristics of the Skokomish Valley describe it as densely covered by shrubs and trees associated with wet or periodically flooded soils and surveys performed in the 1860's describe few portions of the valley as inundated or swampy (Canning et al. 1988). There is evidence that the lowland reaches of the Skokomish Valley was characterized by multiple channels, numerous sloughs and old side channels, and an abundance of snags and log jams, with sluggish stream flow through multiple channels (Canning et al. 1988). This may explain why it was once prime spawning habitat for salmon. Access to healthy habitat, including side-channels and multiple area of refuge are critical to their survival.

Although Quinn (2005) and others state that several factors support smolt survival and growth, the major contributors are water flow, lengthier streams, lower gradients causing more pools in the streams, and most importantly healthy habitat and food availability. Scarcity of suitable over-wintering habitat can cause fish to migrate long distances (Peterson 1982a). Ocean survival of salmon has been shown to be closely linked with the early–life stages of growth and size during freshwater rearing (Bilton et al. 1982; Holtby and Healey 1990; Nislow et al. 1999), suggesting that a larger body size is advantageous (Olegario 2006). Access into areas of refuge appears to determine the long-term survival of these fish.

Healthy habitat needs are demonstrated by the fact that juvenile salmonids prefer habitat with certain combinations of depth, velocity, and other physical characteristics (Quinn 2005). Constructed complex habitat can be created by adding large woody debris to newly created alcoves and dammed pools; these additions showed a significant increase in over-winter survival of juvenile coho salmon in treatment streams, and increases in downstream migrant numbers (Solazzi et al. 2000). Coho salmon have been shown to selectively use deeper, slower water characteristic of pools rather than shallower, faster moving water (Healy and Lonzarich 2000). Additionally, juvenile salmonids shift from using both pool and riffle habitat to predominantly deeper water depths, as found in pool habitat, during winter conditions in small streams (Hartman 1965; Bustard and Narver 1975; Bisson and Nielsen 1983; Murphy et al. 1984).

Survey of Washington state creeks showed that coho salmon were most likely to be found in pools, while other salmonid species preferred shallower, faster water, or intermediate conditions (Bisson et al. 1988). Ponds can provide over-wintering fish benign thermal refuge during the harshest time of year, when their lipid reserves are most constrained. However due to intense predation by avian and mammalian predators (Peterson 1982b), installation of ponds will need to be specifically created to prevent predation, provide ample cover, refuge and depth. Fish will need to be able to safely forage in the benthos areas while taking refuge in the deeper portions; deeper parts of the ponds tend to have lower benthos densities (Peterson 1982b). Ponds are capable of providing ample high-quality detrital base for insect production and rich invertebrate fauna is directly associated with aquatic macrophytes (Hodkinson 1975). It is possible to maximize both survival and growth of overwintering fish by combining the productivity of shallow ponds with the cover of a riverine environment (Peterson 1982b)

Stream-restoration projects that increase the quantity of large woody debris (LWD) and pools tend to increase coho density but decrease that of other species (Roni and Quinn 2001). Productive sites for fish tend to possess hard waters with relatively high inorganic nutrient concentrations; moderate temperatures, especially in spring-fed streams where temperatures are buffered by groundwater inputs year-round; relatively low vegetative canopy coverage allowing ample sunlight to reach the streams and abundant macrophytes and mosses, or dense growths of filamentous algae (Bisson and Bilby 1998). Well developed overhanging vegetation in a riparian area can enhance the input of terrestrial invertebrates, and the presence of meandering and/or braided stream channels can be expected to increase the supply of emerging aquatic insects per unit area of forest (Nakano and Murakami 2001). Additionally, flood plains have been shown as important in the rearing of juvenile salmonids; salmon increased in size substantially faster in seasonally inundated agricultural floodplain than in the river, suggesting better growth rates (Sommer et al. 2001), and larger size is advantageous to ocean survival (Bilton et al. 1982; Holtby and Healey 1990; Nislow et al. 1999; Olegario 2006). The Skokomish River has all these characteristics or can possess them to some degree.

The Corps has proposed several options of remediation for the Skokomish River, including levee and dike removals, setbacks, sediment control structures, re-opening of side channels which experience sub-surface flow, riparian planting, and dredging portions of the mainstem (USACE and USFWS 2008). All of these actions could be beneficial if applied appropriately. However, any actions that disturb salmonid habitat, at any time of the year will likely have a substantial effect on their prey food base, especially during the winter. Disturbing the lotic and lentic areas could potentially eliminate the prey food base during their over-wintering period.

Dredging could have a substantial impact on overwintering juvenile salmonids. Dredging may isolate critical off-channel habitat or these habitats could potentially be eliminated. If dredging is performed, it must be done such that juvenile salmonids can still access tributaries and off-channel ponds (Peters pers. comm. 2010). In addition, dredging depth will influence groundwater levels, which may cause off-channel ponds to become shallower (and potentially more productive) or completely dewater (Peters pers. comm. 2010). Backwater areas will likely be completely eliminated if dredging is completed and will likely require re-creation as part of the dredging process (Peters pers. comm. 2010).

Flooding is now occurring more frequently on the Skokomish River and access into areas of refuge is essential during times of high flows. The river already has low-levels of channel connectivity caused in part by the intermittent level of remediation and, any additional channel elimination could be devastating to the fish populations. A food web analysis between birds and freshwater fishes shows that the loss or degradation of one habitat has a more detrimental effect on neighboring communities that previously recognized (Nakano and Murakami 2001).

My recommendation is that remediation activities consider the food and habitat needs of juvenile salmonids during all seasons, but particularly during winter. Peterson and Reid (1984) suggest that providing additional overwintering habitat within the lower reaches of stream systems may provide refuge to large numbers of resident and immigrant coho salmon. Considering that all the habitat types assessed in this diet study are sustaining these fish to some degree during winter will ensure that remediation efforts are successful at creating more suitable habitat for them.

Several options could alleviate pressure to these populations, including performing relatively small projects over time which would allow the disturbed habitats to achieve some form of stability before other areas of refuge are disturbed. Also, if dredging and other major alterations must occur which further reduce channel connectivity, prior installation of new areas of refuge should be performed and can be very beneficial if created properly. Installing beaded channels, as described by Cederholm and Scarlett (1991), before remediation projects commence is a low-cost technique which has been shown to successfully enhance winter habitat for coho salmon. These beaded channels contained a system of ponds which considerably increased the overwinter survival of juvenile coho salmon. These areas must be allowed enough time to build some level of ecosystem stability, measured by the presence of indicator species, before other large remediation efforts commence.

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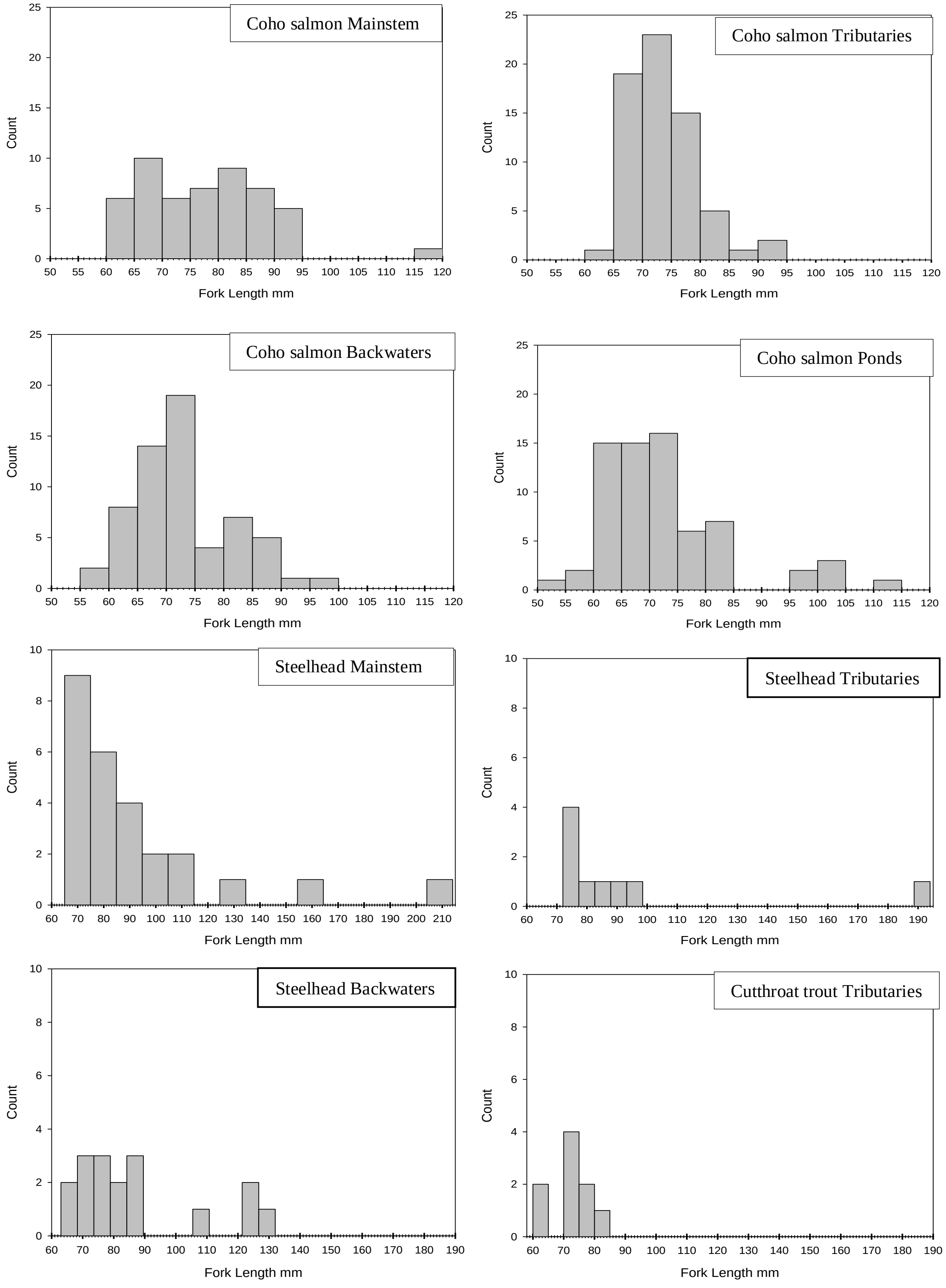
(l) Appendix A. Prey codes/categories

Code	Prey Item
10	Unidentified_Salmon_or_Trout
50	CHIRONOMIDS_MIDGES
501	Chironomid_Larvae
502	Chironomid_Pupae_and_emergent
503	Chironomid_Adults
51	OTHER_AQUATIC_DIPTERA_LARVAE
510	Unknown_Aquatic_Diptera_Larvae
511	Diptera_Larvae_Simuliidae
512	Diptera_Larvae_Ceratopogonidae
513	Diptera_Larvae_Dixidae
514	Diptera_Larvae_Chaoboridae
515	Diptera_Larvae_Tipulidae
516	Diptera_Larvae_Empididae
517	Diptera_Larvae_Athericidae
520	COLLEMBOLA_Isotomis_Springtails
53	TRICHOPTERA_Caddisflies
530	Unknown_Trichoptera
531	Trichoptera_Rhyacophilidae
532	Trichoptera_Leptoceridae
533	Trichoptera_Hydroptilidae
534	Trichoptera_Brachycentridae
535	Trichoptera_Limnephilidae
536	Trichoptera_Psychomyiidae
537	Trichoptera_Hydropsychidae
538	Trichoptera_Wood_Leaf_Cases
539	Trichoptera_Sand_Gravel_Cases
593	Trichoptera_Glossomatidae
594	Trichoptera_Polycentropodidae
54	PLECOPTERA_Stoneflies
540	Unknown_Plecoptera
541	Plecoptera_Perlidae
542	Plecoptera_Perlodidae
543	Plecoptera_Nemouridae
544	Plecoptera_Leuctridae
545	Plecoptera_Capniidae
546	Plecoptera_Chloroperlidae
595	Plecoptera_Sericostomidae
55	EPHEMEROPTERA_Mayflies
550	Unknown_Ephemeroptera
551	Ephemeroptera_Baetidae
552	Ephemeroptera_Heptageniidae
553	Ephemeroptera_Caenidae
554	Ephemeroptera_Siphlonuridae
555	Ephemeroptera_Leptophlebiidae
556	Ephemeroptera_Ephemerellidae
56	ODONATA_Dragonflies
560	Unknown Odonata
561	Odonata_Coenagrionidae
57	COLEOPTERA_Aquatic Beetles
570	Unknown_Coleoptera
571	Coleoptera_Elmidae
572	Coleoptera_Hydrophilidae
573	Coleoptera_Dystisidae
574	Coleoptera_Staphylinidae
575	Coleoptera_Noteridae
576	Coleoptera_Hydrochidae
577	Coleoptera_Circulionidae
58	OTHER_AQUATIC_INSECTS
581	Megaloptera_Sialis_Alderfly
582	Lepidoptera
583	Chrysomelidae_Aquatic_Leaf_Beetle
59	OTHER
590	Unknown
591	Exuvia_Aquatic_Insect_Exoskeleton
592	Misc_Insect_Parts
60	NEOMYSIDS
600	Neomysid_Neomysis
61	AMPHIPODS
610	Unknown_Amphipods
611	Amphipod_Gammaridae_Gammarus spp
612	Amphipod_Talitridae_Hyalella_azteca
62	Crayfish_Astacidae
63	COPEPODS
630	Unknown_Copepods
631	Copepod_Cyclopoid
632	Copepod_Calanoid
633	Copepod_Harpactacoid

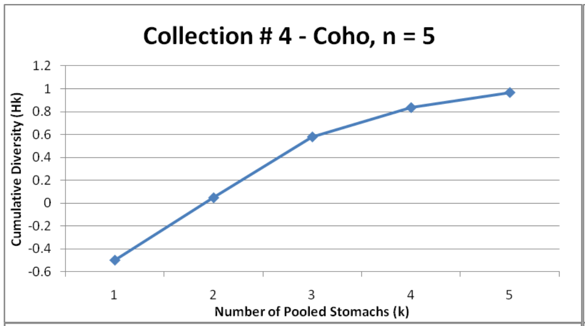
64	CLADOCERANS_Water_Fleas
65	ISOPODS_Asellidae
70	GASTROPODA_Snails_Limpets
701	Gastropod_Gyraulus
702	Gastropod_Campeloma
703	Gastropod_Goniobasis
71	PELECYPODA_Clams_Mussels
72	HIRUDINEA_Leeches
73	AQUATIC_OLIGOCHAETES
741	Aquatic_ Horsehair_Worm
75	HYDRACARINA_Water_Mites
76	OTHER_AQUATIC_INVERTEBRATES
80	Diptera_Adult_Flies
81	Hymenoptera_Ants_Bees
82	Coleoptera
83	OTHER_TERRESTRIAL_ARTHROPODS
831	Arachnids_Spiders_
832	Homoptera_Aphids_
833	Hemiptera_Water_Bugs_Arthropod
834	Thrips_Terrestrial_Arthropod
85	Terrestrial_Oligochaetes_Earthworms
86	Terrestrial_Mollusks_Slugs
87	Terrestrial_Cicadellidae_Leafhopper
90	Detritus
91	Plant_Material
92	Rocks
93	Fish_Eggs
94	Unidentified_Organic_Matter
95	Unidentified_Inorganic_Matter
96	Other
835	Centipedes_Terrestrial_Arthropod
84	Terrestrial_Isopods_Sow_Bugs

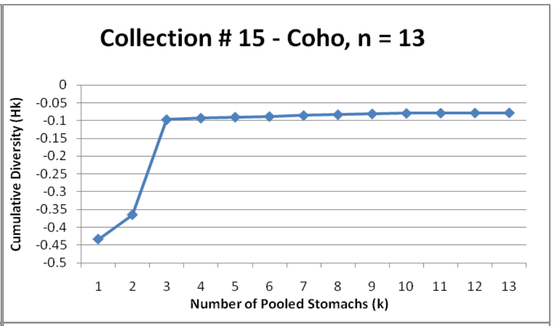
(m) **Appendix B.** Length frequency (5-mm FL increments) distributions for coho salmon in all habitat types, steelhead (10-mm FL increments) in backwaters, mainstem, and tributaries, and cutthroat trout (10-mm FL increments) in tributaries. Graphs include all fish collected, including those greater than 100-mm FL, which were excluded from all analyses.

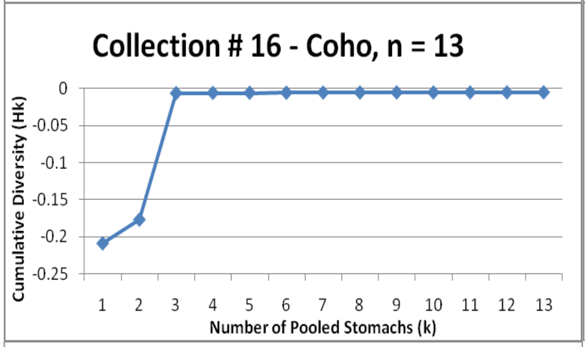


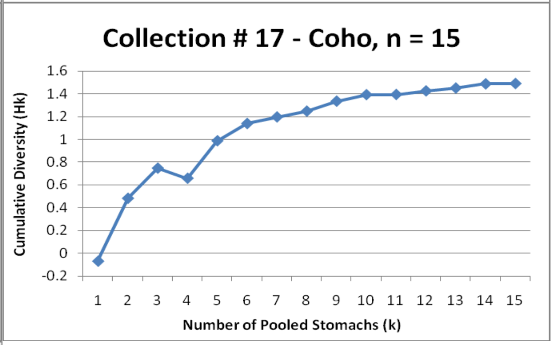
(n) **Appendix C.** Results of Pielou's method on individual sample sets for the determination of adequate sample size for coho salmon.

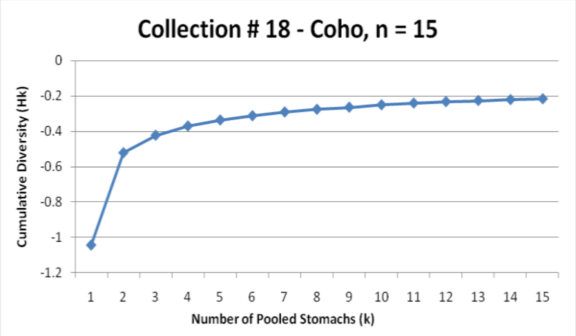
Backwaters



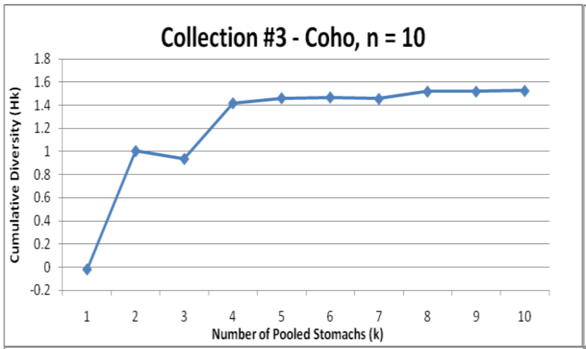


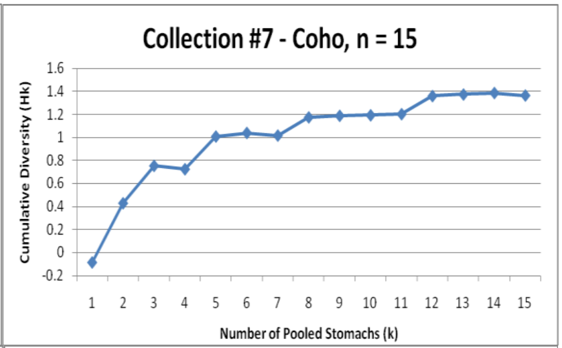


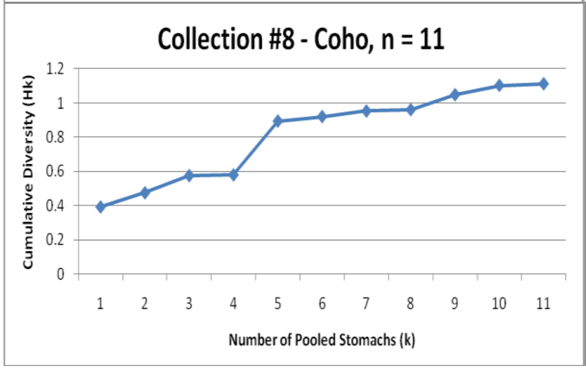


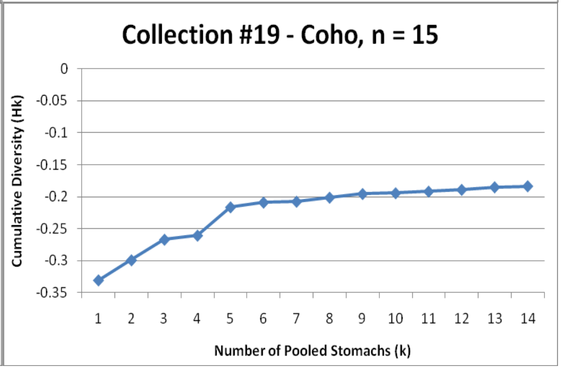


Mainstem

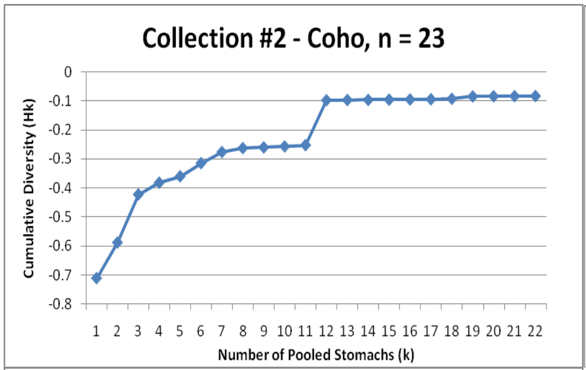


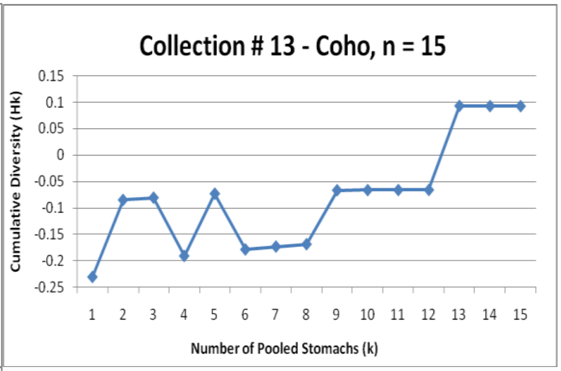


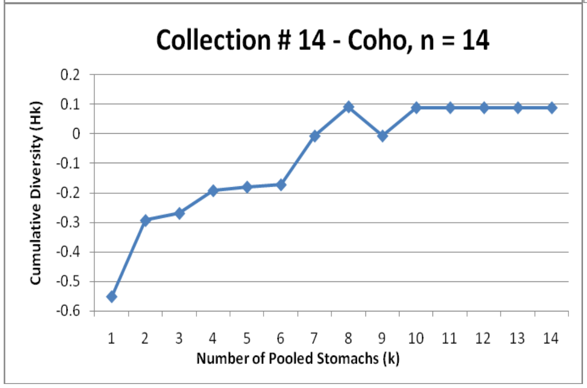




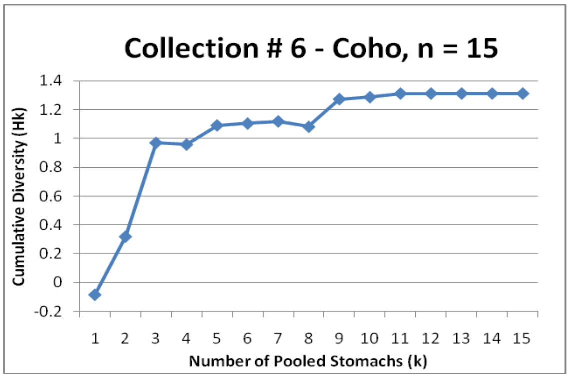
Ponds

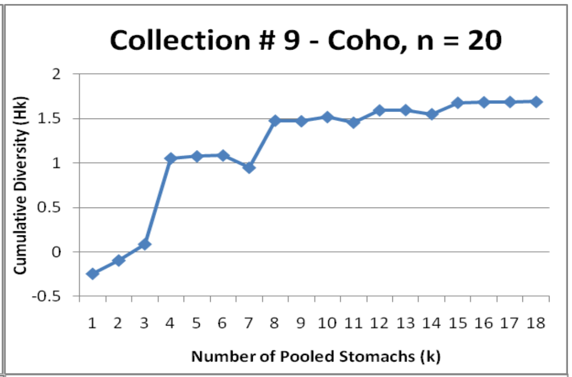


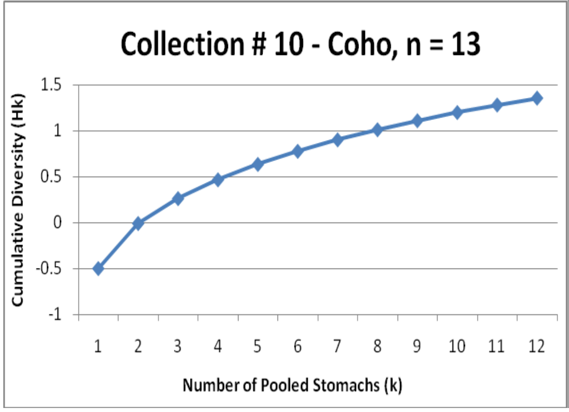




Tributaries


Collection # 6 - Coho, n = 15


Collection # 9 - Coho, n = 20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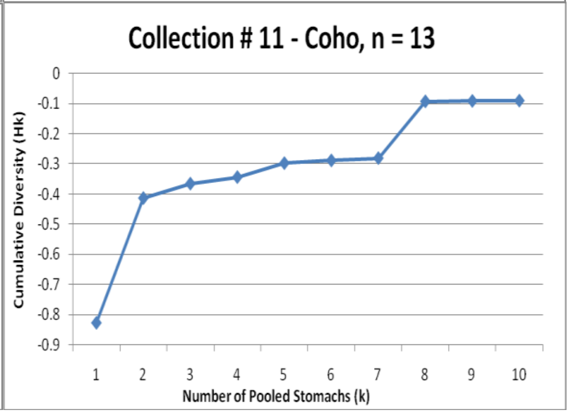
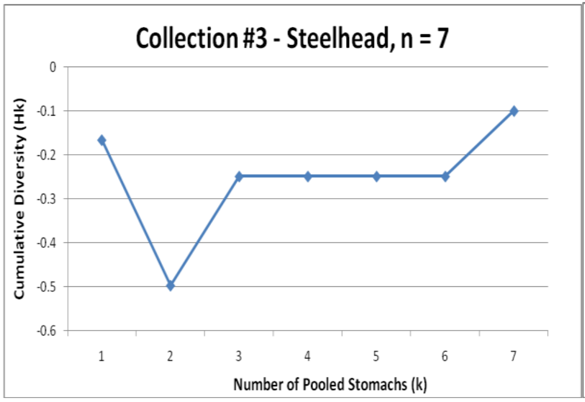
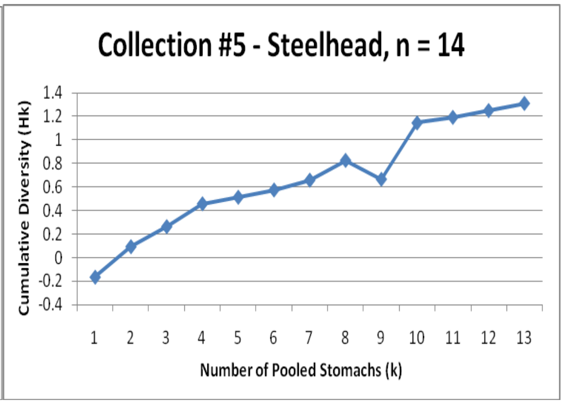

Collection # 11 - Coho, n = 13

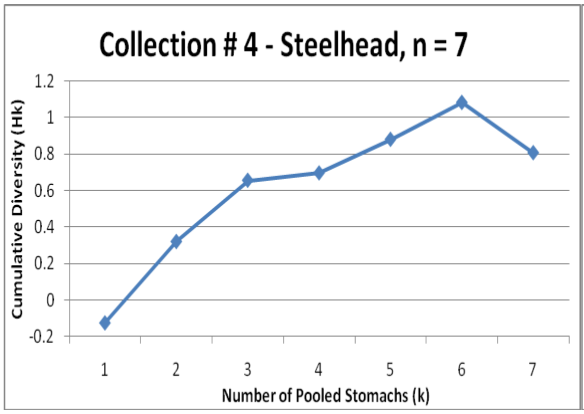
(o) **Appendix D.** Results of Pielou's method on individual sample sets for the determination of adequate sample size for steelhead and cutthroat trout.

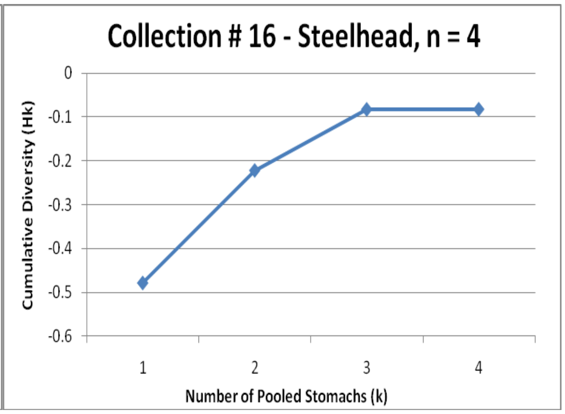
Mainstem



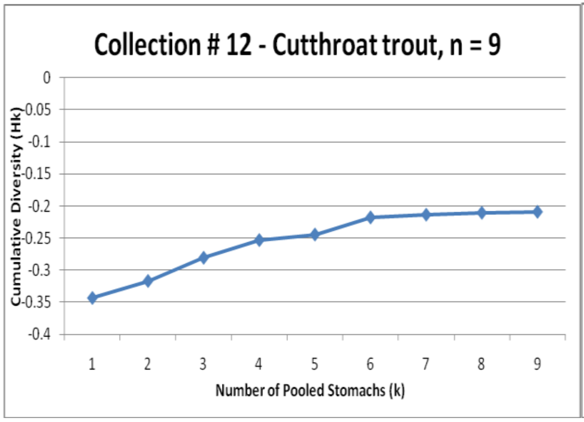


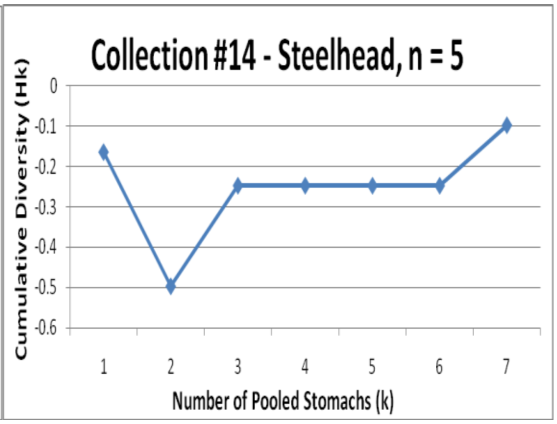
Backwaters





Tributaries





(p) **Appendix E.** Weights of all prey items for coho salmon, steelhead and cutthroat trout in all habitat types.

	Mainstem		Tributaries		Backwaters		Ponds
	coho salmon	steelhead	coho salmon	cutthroat trout	coho salmon	steelhead	coho salmon
Mollusca	0.0442		0.0232	0.0008	0.0454		0.0689
Bivalvia	0.0442		0.0232				0.0211
Gastropoda				0.0008	0.0454		0.0478
Annelida							
Oligochaeta	0.2559		0.4595		0.6171		0.6822
Nematomorpha	0.0023		0.0012	0.0078	0.0002		0.0000
Gordiacea	0.0023		0.0012	0.0078	0.0002		0.0000

Arthropoda

Insects

Aquatic

Collembola							
Isotoma	0.0024		0.0145	0.0033	0.0075		0.0074
Ephemeroptera	**0.2462**	**0.0194**	**0.0930**	**0.0288**	**2.5456**	**0.1388**	**0.1617**
Baetidae	0.1389	0.0093	0.0248	0.0040	2.1566	0.1339	0.0073
Ephemerellidae	0.0262	0.0079	0.0362	0.0127	0.0956	0.0006	0.1261
Heptageniidae	0.0671	0.0006	0.0026	0.0057	0.2835	0.0040	0.0271
Leptophlebiidae	0.0090		0.0258		0.0020		
Unknown sp.	0.0050	0.0016	0.0035	0.0064	0.0080	0.0003	0.0012
Plecoptera	**0.1257**	**0.0615**	**0.2093**	**0.0173**	**0.7907**	**0.0217**	**0.0882**
Capniidae	0.0013		0.1079		0.0079	0.0020	
Leuctridae	0.0264	0.0003	0.0092		0.6296	0.0176	0.0131
Nemouridae	0.0279		0.0698		0.0174		
Perlidae	0.0093	0.0051		0.0025			0.0303
Perlodidae	0.0541	0.0463	0.0192		0.0949		0.0216
Sericostomidae							0.0010
Unknown sp.	0.0067	0.0098	0.0031	0.0148	0.0408	0.0021	0.0221
Megaloptera							
Sialidae	0.0720		0.0718		0.0931		0.3713
Trichoptera	**0.1339**	**0.0672**	**0.1862**	**0.0537**	**0.2158**	**0.0497**	**0.1238**
Brachycentridae				0.0003			
Glossomatidae	0.0555		0.0258		0.0014		
Hydropsychidae	0.0008				0.0436	0.0011	
Hydroptilidae	0.0005						
Limnephilidae	0.0687	0.0672	0.1603	0.0525	0.1451	0.0463	0.0987
Polycentropodidae	0.0043				0.0048		
Psychomyiidae				0.0009			
Rhyacophilidae	0.0040				0.0078		
Unknown sp.	0.0001		0.0001	0.0001	0.0131	0.0023	0.0252
Chironomidae	**0.1993**	**0.0046**	**0.9388**	**0.0421**	**0.4340**	**0.0394**	**0.0526**
Adults	0.0018		0.0165		0.0200	0.0001	0.0035
Larvae	0.0512	0.0010	0.5858	0.0348	0.3005	0.0391	0.0405
Pupae and emergent	0.1463	0.0036	0.3365	0.0073	0.1135	0.0002	0.0086
Other Diptera larvae	**0.0502**	**0.0040**	**0.0819**	**0.0266**	**0.2613**	**0.0050**	**0.0273**
Ceratopogonidae	0.0001		0.0034	0.0129			0.0026
Chaoboridae							0.0032
Dixidae			0.0521				

Empididae	0.0057		0.0070		0.0139		0.0005
Simuliidae	0.0420		0.0170	0.0135	0.2336	0.0050	0.0068
Tipulidae	0.0024		0.0025		0.0138		
Unknown sp.		0.0040		0.0002			0.0141
Odonata							
Coenagrionidae							0.0111
Lepidoptera	0.0240		0.0036		0.0089		0.0076

Arthropoda

Insects

Terrestrial

Coleoptera	**0.0210**		**0.0681**	**0.0110**	**0.1038**		**0.0226**
Chrysomelidae			0.0013				
Circulionidae					0.0065		
Elmidae	0.0026		0.0262	0.0110	0.0537		
Hydrochidae	0.0146		0.0225		0.0223		0.0057
Noteridae			0.0065				
Staphylinidae	0.0002		0.0116		0.0126		0.0009
Unknown sp.	0.0035				0.0087		0.0160
Other Diptera adult							
Unknown sp.	0.0108		0.0965		0.1024	0.0004	0.0611
Homoptera							
Cicadellidae					0.0048		
Hymenoptera							
Formicidae			0.0275				

Arthropoda

Insects-Other

Other - Insect - Unknown sp.	0.1347	0.0050	0.0400	0.0021	0.1953	0.0059	0.1211
Exuvia	0.1345	0.0050	0.0010	0.0021	0.1893	0.0059	0.1066
Insect eggs					0.0060		0.0145
Trichoptera Cases	**0.0188**	**0.0474**	**0.0128**	**0.0429**	**0.0042**	**0.0669**	**0.0031**
Sand/Gravel Cases	0.0123	0.0402	0.0123	0.0323	0.0017	0.0631	0.0031
Wood/Leaf Cases	0.0065	0.0072	0.0005	0.0106	0.0025	0.0038	
Crustacea			0.0217		0.0455		0.0905
Copepoda			0.0004				0.0009
Amphipoda			0.0213		0.0455		0.0896
Arachnida							
Unknown sp.	0.0014		0.0620	0.0004	0.0518		0.0000
Hydracarina		0.0001	0.0006		0.0001		0.0081
Fish	0.0116				0.2492		0.0573
Fish Eggs	0.0116				0.0022		0.0573
Unidentified salmonid fry					0.2470		

Other

Other	0.1595	0.0141	0.0965	0.0105	0.0737	0.0781	0.1498
Plant Material	0.0524	0.0082	0.0403	0.0080	0.0733	0.0080	0.0760
Unidentified Organic Matter	0.1071	0.0059	0.0562	0.0025	0.0004	0.0701	0.0738

(q) **Appendix F.** Summary of the major prey categories; percent proportion by weight (%Wi), percent proportion by number (%Ni), and frequency of occurrence (%Oi) by habitat type for coho salmon.

	Habitat Types											
	Mainstem			Tributaries			Backwaters			Ponds		
Prey Category	%Wi	%Ni	%Oi	%Wi	%Ni	%Oi	%Wi	%Ni	%Oi	%Wi	%Ni	%Oi
Mollusca	2.97	0.43	4.00	0.96	0.07	1.96	0.00	0.00	0.00	3.27	2.96	8.82
Annelida												
Oligochaeta	17.19	0.29	3.92	18.96	0.15	8.16	10.66	0.13	8.20	32.36	0.56	4.76
Arthropods												
Insects												
Aquatic Insects												
Collembola												
Isotoma	0.16	1.59	15.69	0.60	1.53	30.61	0.13	0.24	18.03	0.35	4.26	12.70
Ephemeroptera	16.54	18.18	76.00	3.84	6.15	60.78	43.97	45.89	60.00	7.67	28.15	47.06
Plecoptera	8.45	6.49	50.00	8.64	2.76	47.06	13.66	6.55	51.67	4.18	5.93	25.00
Megaloptera												
Sialidae	4.84	0.29	3.92	2.96	0.11	6.12	1.61	0.15	9.84	17.61	2.96	17.46
Trichoptera	9.00	5.63	36.00	7.68	1.30	33.33	3.73	2.10	43.33	5.87	2.59	13.24
Chironomidae	13.39	58.01	48.00	38.74	76.96	84.31	7.50	38.54	88.33	2.49	35.71	48.53
Other Diptera larvae	3.37	5.92	42.00	3.38	7.23	39.22	4.51	4.38	36.67	1.29	3.33	14.71
Terrestrial Insects												
Coleoptera	1.41	1.59	20.00	2.81	1.19	35.29	1.79	0.56	30.00	1.07	0.74	5.88
Diptera												
Adult	0.72	0.87	7.84	3.98	1.72	32.65	1.77	1.14	47.54	2.90	3.70	12.70
Crustacea	0.00	0.00	0.00	0.90	0.37	11.76	0.79	0.09	5.00	4.29	8.33	13.24
Amphipoda	0.00	0.00	-	0.00	0.00	-	0.59	0.06	-	0.00		
Gammaridae	0.00	0.00	0.00	0.88	0.34	10.20	0.19	0.02	1.64	4.25	5.19	14.29
Unknown sp.	0.00	0.00	0.00	0.00	0.00	0.00	0.59	0.06	3.28	0.00	0.00	0.00
Copepoda												
Cyclopoida	0.00	0.00	0.00	0.02	0.04	2.04	0.00	0.00	0.00	0.04	3.15	3.17
Fish	0.78	0.00	8.00	0.00	0.00	0.00	4.30	0.06	3.33	2.72	0.00	4.41

(r) **Appendix G.** Summary of the major prey categories; percent proportions by weight (%Wi), percent proportions by number (%Ni), and percent frequency of occurrence (%Oi) by habitat type for steelhead and cutthroat trout.

	cutthroat trout			steelhead					
	Tributaries			Mainstem			Backwaters		
Prey Category	%Wi	%Ni	%Oi	%Wi	%Ni	%Oi	%Wi	%Ni	%Oi
Mollusca	0.32	0.42	11.11	0.00	0.00	0.00	0.00	0.00	0.00
Nematomorpha									
Paragordius tricuspidatus	3.17	2.54	33.33	0.00	0.00	0.00	0.00	0.00	0.00
Arthropoda									
Insects									
Aquatic Insects									
Collembola									
Isotoma	1.34	1.27	22.22	0.00	0.00	0.00	0.00	0.00	0.00
Ephemeroptera	**11.66**	**9.75**	**77.78**	**8.69**	**22.97**	**60.00**	**34.19**	**28.93**	**66.67**
Plecoptera	**7.00**	**4.24**	**66.67**	**27.54**	**6.76**	**20.00**	**5.35**	**1.89**	**33.33**
Trichoptera	**21.76**	**17.80**	**100**	**30.10**	**55.41**	**50.00**	**12.25**	**18.24**	**66.67**
Chironomidae	**17.05**	**55.08**	**88.89**	**2.05**	**8.11**	**20.00**	**9.71**	**48.11**	**75.00**
Diptera larvae	**10.77**	**7.20**	**66.67**	**1.79**	**1.35**	**5.00**	**1.23**	**2.52**	**16.67**
Terrestrial Insects									
Coleoptera	**4.44**	**1.69**	**44.44**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**	**0.00**
Diptera									
Adult	0.00	0.00	0.00	0.00	0.00	0.00	0.10	0.31	8.33

(s) **Appendix H.** Percent index of relative importance (%IRI) for major prey categories for coho salmon, steelhead, and cutthroat trout

	coho salmon				cutthroat trout	Steelhead	
Prey Category	**Mainstem**	**Tributary**	**Backwater**	**Pond**	**Tributary**	**Backwater**	**Mainstem**
Mollusca	**0.11**	**0.00**	**0.00**	**0.57**	**0.02**	**0.00**	**0.00**
Arthropoda							
Insects							
Aquatic							
Collembola	**0.23**	**0.29**	**0.02**	**0.60**	**0.17**	**0.00**	**0.00**
Ephemeroptera	**22.84**	**2.74**	**23.84**	**17.55**	**5.13**	**19.27**	**13.32**
Plecoptera	**6.46**	**2.42**	**4.61**	**2.63**	**2.30**	**1.10**	**4.81**
Megaloptera larvae	**0.17**	**0.08**	**0.07**	**3.74**	**0.00**	**0.00**	**0.00**
Trichoptera	**4.55**	**1.35**	**1.11**	**1.16**	**12.19**	**9.30**	**29.91**
Chironomidae	**18.19**	**49.23**	**32.27**	**33.9**	**34.43**	**32.97**	**1.63**
Diptera Larvae	**3.37**	**1.88**	**1.44**	**0.70**	**3.69**	**0.28**	**0.11**
Terrestrial							
Coleoptera	**0.51**	**0.63**	**0.31**	**0.11**	**0.84**	**0.00**	**0.00**
Crustacea	**0.00**	**0.06**	**0.01**	**1.74**	**0.00**	**0.00**	**0.00**
Fish	**0.05**	**0.00**	**0.06**	**0.12**	**0.00**	**0.00**	**0.00**