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# Executive Summary

This study aims to examine what marine resources are found in the mixing zones of the estuary. In the Skeena estuary there is an abundance of forage fish such as eulachon, Longfin smelt, capelin, and others. These species spend all or part of their lives in the estuary where they play a role in ecosystem function of the entire Skeena watershed. Many key ecological questions exist about these species and how they support species important to humans such as salmon. At this early stage of knowledge basic questions like presence and distribution are important. This study generated knowledge of some species such as Longfin smelt and other forage fish.



# Table of Contents

<b>Executive Summary</b> .....	2
<b>1.0 Introduction</b> .....	4
<b>2.0 Methods</b> .....	4
2.1 <i>Salmonid Diet Composition with a focus on Forage Fish in the Estuary</i>	
2.2 <i>Characterizing Sampling Locations - Location and Environmental Parameters</i>	
2.3 <i>Estuary Species Composition</i>	
2.3.1. <i>Species Identification through Genetic Analysis</i>	
2.4 <i>Estuary Delineation</i>	
<b>3.0 Results</b> .....	7
3.1 <i>Salmonid Diet Composition</i>	
3.2 <i>Salinity Profiles</i>	
3.3 <i>Genetic differentiation – Longfin and Eulachon species ID</i>	
3.3.1 <i>Genetic differentiation - Longfin and Eulachon species ID</i>	
3.4 <i>Estuary Delineation</i>	
<b>4.0 Discussion</b> .....	11
4.1 <i>Salmon Diet Composition</i>	
4.2 <i>Estuary Physical and Ecological Description</i>	
4.2.1 <i>Material Transport</i>	
4.3 <i>Estuary Species Composition</i>	
4.3.1 <i>Baseline limitations of Genetic Identification</i>	
4.3.2 <i>Species Use of the Skeena Estuary</i>	
4.3.3 <i>Valued Ecosystem Components</i>	
<b>5.0 Acknowledgements</b> .....	16
<b>6.0 References</b> .....	17
<b>7.0 Appendices</b> .....	18
7.1 <i>Appendix A. Original data attached. Fish Sample Data and Fish Analyzed Genetically</i>	
7.2 <i>Appendix B. Photos of Selected Sorted Samples</i>	
7.3 <i>Appendix C. Estuary Mapping</i>	
Map 1. <i>Skeena Estuary Salinity Profiles</i>	
Map 2. <i>Estuary Delineation</i>	

# 1.0 Introduction

This study's main objectives were to;

- A) Investigate Salmonid diet composition with a focus on forage fish
- B) Map and characterize the estuary and the sites sampled
- C) Investigate the distribution, density and habitat use of forage fish and other potential salmonid prey species in the Skeena estuary and establish whether or not the estuary contains Valued Ecosystem Components (VEC)

Determining the diet composition of salmon, especially smelts that we suspect are important prey for young salmon in the estuary, first requires the ability to locate and identify the forage fish and other potential prey species present. Since young salmon likely consume small fish this will be mostly larval and juvenile prey. For many species identification is not a problem since they can be identified using keys such as Fishbase or Ichthyoplankton Information System. However, confusion about larval species ID is a common problem, especially among closely related species like the smelts. Therefore larval fish require careful examination, and in some cases, use of genetics to separate species. The timing and location of sampling to catch young salmon was also a question. The approach we followed was to sample areas known to contain forage fish from past work, and to attempt to capture young salmon feeding on them at those locations.

## 2.0 Methods

### *2.1 Salmonid Diet Composition with a focus on Forage Fish in the Estuary:*

Fish were collected under DFO Licence Number XR 289 2011. Salmon fry and smolts vary in size and likely in size of prey preference. Therefore we sampled for very small potential prey with a plankton net and larger prey with a larger net. Both types of nets were fished at depths and locations where large numbers of larval and juvenile smelt were caught in 2010. Next, a diversity of sites were sampled to collect additional forage fish samples to address questions about the distribution and abundance of potential prey species, especially eulachon (*Thaleichthys pacificus*), Longfin smelt (*Spirinchus thaleichthys*) and capelin (*Mallotus villosus*).

A live-well was constructed to hold live salmon using a cooler and electric live-well motor similar to a bilge pump. Digital food scale and measuring trough were used to measure fish caught. Clove oil for anaesthetizing fish and trout stomach pumps were used to pump stomachs of salmon caught so they could be released alive.

Tows were oblique to the shoreline to cross eddy lines, and sample a diagonal of depths from close to the bottom at 25m, to the surface. All samples were located with a GPS position, and temperatures and salinities recorded at 1m below the surface, then at 2m intervals for depths sampled. Samples were preserved in 95% ethanol to permit genetic analysis and retained in 500ml sample jars for later sorting to suspected smelt species, other fish, shrimp and other taxa. These samples, collected in a wet environment, were completely submerged in 95% alcohol. Wet tissue samples were covered completely at all times in solution and not exposed to air until extraction of DNA occurred. A >10:1 ratio of buffer to tissue was used. Exposure of alcohol-stored tissue to air can cause cell wall fracturing and loss of DNA into the liquid buffer. Net samples were taken on August 3, 6, 14, 18, 30, 31, and September 1, 2011.

Nets used were a 2m diameter larval net, a 3m diameter larval net each with 15mm mesh, and a 0.5m diameter bongo net with 350micron mesh. Sample locations repeated those from 2010, from Kwinitza, well within upstream extent of tidal influence but above salinity, down to Site 22, approximately half way down Kennedy Island (see Map 1). Site 22 is within the Skeena estuary but close to full marine salinity (21.5ppt at 1m depth and 26.5ppt at 13m). Samples selected for genetic analysis came from a diversity of sites; sites 8, 9, 10, 64, 65, and 70. This variety of sample sites was selected to ensure as large an area as possible was checked, and in case any single site only contained one or the other species of smelt.

## *2.2 Characterizing Sampling Locations - Location and Environmental Parameters*

A GPS location was taken of each sample site with a Garmin rhino 530HCx. Temperature and salinity were measured at each location where sampling was carried out with a YSI Model 85 digital handheld SCT meter with a 50' probe at 2m intervals down to 13m. Depth at each site was recorded from a Humminbird 325 fish finder/depth sounder.

Each fish sampling site was recorded as a point. In order to characterize the estuary, transects composed of a series of point samples at 500m intervals were conducted across channel out to areas without freshwater dilution.

## *2.3 Estuary Species Composition*

### *2.3.1 Species Identification through Genetic Analysis*

All samples were preserved immediately in the field in 95% ethanol in 500ml sample jars. After samples had been preserved for 2 months a subset was selected for genetic analysis. After rinsing in fresh water under a fume hood, samples of larval fish and other taxa were picked out of the sample with eyedroppers and tweezers as suitable from pyrex baking dishes on a light table. After samples had been preserved for 2 months samples were again sorted and suspected smelts, either eulachon, capelin and/or Longfin smelt larvae were placed in small paper envelopes. Date of capture, sample number,

and length of each larvae were recorded. Samples were sent to NOAA lab in Seattle for genetic analysis.

### 2.3.2 Genetic differentiation - Longfin and Eulachon species ID Anna Elz, NOAA

DNA was extracted from caudal fin tissue using a Qiagen 96-well kit . We performed polymerase chain reaction (PCR) of the cytochrome oxidase (COI) region of the mitochondrial DNA on genomic DNA using primers from Barcode of Life database (NWFSC Marine Fish Voucher Collection project code FMV

<http://www.boldsystems.org/views/projectlist.php?searchBy=Project+Code&filter=FMV>).

The species identification assay consists of restriction fragment length polymorphisms (RFLP) of the COI product with two enzymes (ApaI and KpnI) in the same reaction at 25 degrees.

Neither enzyme cuts eulachon (in the baseline). ApaI cuts capelin and Longfin, and KpnI cuts only capelin producing three distinct banding patterns to identify each species. \*\*

Samples identified as eulachon were then sequenced to confirm species identification.

Positive control vouchers for eulachon and Longfin were also sequenced, as well as a Longfin smelt specimen for confirmation. The DNA sequences were aligned to known positive controls and compared to sequences in Genbank using the BLAST tool.

### 2.4 Estuary Delineation

Estuaries are unique ecosystems in that they have constantly fluctuating salinity due to the mixing of fresh and salt water. This poses metabolic challenges for organisms that live in estuaries as they must be capable of adapting to constantly varying salinities. At the same time, species also have a range of suitable salinity or productive zone for that species. The result is an ecosystem with a distinct makeup of species and processes that are adapted to the estuarine environment. In temperate estuaries, like the Skeena, the estuary is a critical environmental link between the freshwater and saltwater environments and the species that live there. It is particularly important to anadromous salmon species since it provides the opportunity for juveniles to gradually adjust to full strength sea water and provides nutrient rich brackish environments that allow juveniles to begin rapid growth which is critical for reducing predation. Given the importance of the estuarine environment to salmonids and the unique processes and ecosystems that occur there it is important from a management perspective to be able to delineate the physical extent of the estuarine zone. The first step in the delineation process is to classify the estuary.

In general estuaries are classified based on the character of their salinity structure. Although many estuary definitions exist one of the most accepted definitions is the one developed by Cameron and Pritchard (1963), as follows;

An estuary is a semi-enclosed and coastal body of water, with free communication to the ocean, and within which ocean water is diluted by freshwater derived from land.

Delineation of the Skeena estuary for this study involved the field measurement of salinity structure through vertical profiling and a review of previously published salinity structure studies. Classification is also based on sonar surveys, bathymetric mapping interpretation and interpretation of the estuary geomorphic context.

## 3.0 Results

### 3.1 Salmonid Diet Composition

Small numbers of salmon fry were captured in sampling; four pink salmon (*Onchorhynchus gorbuscha*), one chum (*Oncorhynchus keta*) and one chinook (*Oncorhynchus tshawytscha*). All of these had their stomachs pumped without identifiable contents. Two were retained in samples and sent along with smelts for genetic analysis in case stomach contents could be determined genetically.

### 3.2 Salinity Profiles

The estuary is a complex series of habitats between the freshwater of the river and saltwater of the ocean. Mixing of fresh and salt water occurs when the fresh water rides down over the salinity that intrudes upstream with the tide. Further mixing occurs as the tides rise and fall causing estuarine circulation. This mixing creates pockets of higher salinity even upstream of fresher water. Generally for each location salinity increases as depth increases at each site. For simplicity in trying to understand use of these mixing zones and the habitat preferences of the species found, salinities were averaged for each site where fish samples were collected. In order to better characterize the estuary salinity profiles were determined for the transects made.

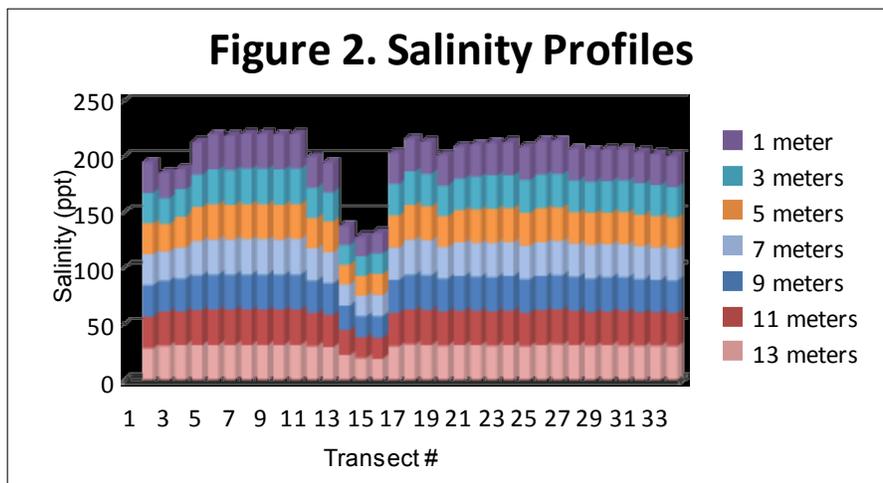


Figure 2: Salinity profiles for the Skeena Estuary study. Map 1 (Appendix B) shows the location of the profiles.



As expected, we found juvenile (averaged 40mm approximately) smelts that could not be differentiated between eulachon and Longfin smelt. These species cannot be sorted by eye until they are at least 40-50mm when gill rakers can be counted. In order to differentiate between these and other smelt species in samples it was necessary to use genetic markers, markers that first had to be developed. We collected samples for genetic analysis, separated these samples to suspected species and sent them to a NOAA lab for genetic confirmation.

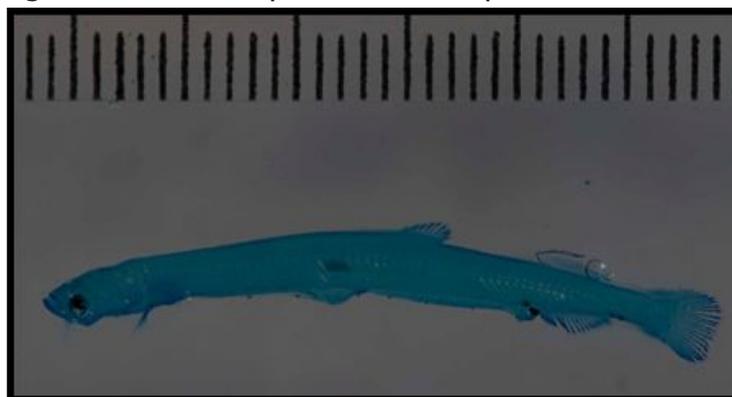
### *3.3.1 Genetic differentiation - Longfin and Eulachon species ID Anna Elz, NOAA*

Fifty unknown larval smelt were processed as well as two salmon smolts. In initial analysis, five samples were identified as eulachon and the remainder Longfin smelt. A Chinook salmon produced a banding pattern matching Longfin smelt and a Pink salmon fry produced a product matching eulachon. The DNA sequence data for the five specimens identified as eulachon by RFLP did not confirm those results. Those five specimens were actually Longfin smelt. The sequence data for the Longfin smelt specimen did confirm that ID and matched the vouchered samples.

Once Longfin smelt identification was possible we found Longfins and Panadaliidae (pink) shrimp were the most abundant species collected. Sites 064 and 9 possess by far the greatest density of biomass found in this study; astounding densities of 292/m<sup>3</sup> and 155/m<sup>3</sup> for Longfin and 487/m<sup>3</sup> for shrimp. The highest biomass of LF and shrimp was found in the lowest freshwater areas, just upstream of salinity. Longfin smelt were found in all salinities (0 – 27ppt) at low densities but very concentrated at upstream end of salinity in 0ppt. Together with Longfin smelt, shrimp were also found concentrated at these same sites, at low densities basically at all sites, and in high densities just above salinity.

A single recently hatched 6mm eulachon larvae was captured with the plankton net in 0ppt at Kwinitza several kms upstream of uppermost salinity detected. Other species were found throughout the mixing zones, at lower densities. Of note, other smelt species caught in high densities in previous work that sampled earlier in the year, such as capelin, were not detected in this sampling (Fig. 1).

Figure 1. 30mm Capelin Larvae Captured in 2010



Pacific sandfish (*Trichodon trichodon*) were found here within a range of salinity from 15 – 21ppt and not elsewhere. Other studies have found them commonly in nearshore waters in the Gulf of Alaska but in more marine environments with higher salinities of 30-33ppt (Thedinga et al. 2006). Walleye pollock (*Theragra chalcogramma*) were caught at one location from 75-105mm. Walleye pollock are known to use large estuaries and these fish were likely about 6 months old.

(<http://www.nwfsc.noaa.gov/publications/techmemos/tm44/walleyepollock.htm>) Pacific lamprey (*Entosphenus tridentatus*) were caught as both juveniles (microptamia) of 190mm and as ammocoetes or larvae that filter feed in sand for 3 to 7 years (Oregon Fish and Wildlife Service Factsheet). Starry flounder (*Platichthys stellatus*) were caught in low (0.73ppt) and zero salinity.

**Table 2. Forage Fish Species in the Skeena Estuary**

Common Name	Latin	Abundance Estimate	Source
Capelin	<i>Mallotus villosus</i>	Abundant	Pers. Obs.
Eulachon	<i>Thaleithys pacificus</i>	500tonnes in 2001 run	Lewis et al. 2001
Pacific herring	<i>Clupea pallasii</i>	Abundant	Pers. Obs.
Longfin smelt	<i>Spirinchus thaleichthys</i>	Abundant	This study
Pacific lamprey	<i>Entosphenus tridentatus</i>	Abundant	This study
Pacific sandfish	<i>Trichodon trichodon</i>	Apparently abundant	This study
Sandlance	<i>Ammodytes hexapterus</i>	Abundant	Pers. Obs.
Staghorn sculpin	<i>Leptocottus armatus</i>	Unknown	This study
Starry flounder	<i>Platichthys stellatus</i>	Apparently abundant	This study
Surf smelt	<i>Hypomesus pretiosus</i>	Unknown	Pers. Obs.
Walleye pollock	<i>Theragra chalcogramma</i>	Unknown	This study

### 3.4 Estuary Delineation

Map 1 (Appendix B) shows the location of the salinity profiles across the estuary. Map 2 (also in Appendix B) shows estuary delineation for this project (the red lines on map 2 show the approximate dimensions of the three estuary subdivisions). The delineation is strongly reflective of the observed salinities from the profiling conducted for this study and the sampling conducted by Environment Canada in the 1970s (Hoos, 1975). The estuary is broken into three major subdivisions because of the complexity of habitats and geomorphic environments found there. The subdivisions include, 1) the Upper estuary, a largely riverine, freshwater form delineated up to the maximum extent of tidal influence at the Kasiks confluence, 2) Mid Estuary, a fjord estuary form characterized by soft sediments, boulder glacial lag deposits and mixed salinity gradients, and 3) the Outer Estuary with a salinity structure of freshwater over saltwater. Yellow lines at the extreme north and south of the estuary indicate where the delineation is indeterminate. In the extreme north the boundary between the Skeena and Nass estuaries is indeterminate because there is simply no clear

demarcation between the brackish water contributions from the two watersheds. A meaningful demarcation between the two estuaries may not be possible. In the extreme south the yellow line indicates the limit of sampling.

## 4.0 Discussion

### 4.1 *Salmon Diet Composition*

Salmon fry are known to make differing use of the Skeena estuary. Pink salmon migrate to the estuary immediately upon hatching in May where they move out into shallow estuary channels, along the beaches and sandbanks of Flora and DeHorsey banks (Hoos, 1975). Sockeye seaward migration takes place from April to June, peaking in Mid-May. Upon reaching the estuary the majority stay right in the river mouth, or on Flora Bank near Kitson Island where they remain for a few weeks to a month (Hoos, 1975). Coho have a longer estuary residence time, with juveniles remaining in the shallow waters of the sand banks, particularly of Inverness Passage, for several weeks to two months before moving out to sea. Chum fry emerge in the spring and move directly to the estuary where they remain from May until August or September, mainly in Inverness Passage (Hoos, 1975). Chinook fry migrate downstream in the spring after spending from a few days to a year in their natal stream (Quinn, 2006). Upon reaching the estuary they take up residence in the estuary shallows of Inverness Passage for the summer, leaving in September (Hoos, 1975). Life-history type 3 chinooks spend from early summer to fall in estuaries and it was these we were most likely to catch, and possibly the lone chinook smolt we did catch. Nothing is known of the Skeena estuary residence of Steelhead prior to movement to the open sea.

In spite of a reasonable knowledge of juvenile salmon presence in the Skeena estuary, little is known about their diet composition while there. This is a very important knowledge gap. In this study, the sampling methods used, and/or timing of study failed to capture enough salmon fry and smolts to investigate diet composition. This, in spite of the capture of 1144 Longfin smelt averaged 54mm in length, about the same length as the pink salmon fry captured. As well, many Longfins larger than the salmon fry were caught, probably with a higher burst speed and greater ability to avoid the net. Therefore the timing of sampling was most likely a reason we missed the salmon.

In this study sampling was done mid-channel and not along stream sides or in intertidal marshes. Salmon are well-known to use intertidal habitats along the stream side but we attempted to catch salmon where greatest densities of forage fish were known to be. Therefore both the timing and sampling locations contributed to the low numbers of salmon captured and therefore lack of information obtained on salmon diet composition.

## *4.2 Estuary Physical and Ecological Description*

The Skeena estuary is one of the more physically complex estuaries on the west coast of North America due to its variation in salinity structure, the effect of multiple channels, distributaries and islands that dissect the delta and the outer Islands that enclose both the Skeena and Nass Estuaries. The Mid Estuary is partially confined by Kennedy and Smith islands and the numerous adjoining sand bars and banks along the delta front. The outer estuary is partially confined by Stevens, Porcher and the Melville / Dundas group and the Tree Knob group. The result is a wide range of salinity structures across the three estuary subdivisions. Salinity transects in the northern portion of the outer estuary (the yellow line at the north end of the outer estuary on Map 2 (Appendix C)) indicate a strong freshwater influence that suggests that there is no clear demarcation between the Skeena and Nass estuaries and the unit may be most accurately described as a "Mega Estuary". Therefore, ecosystem function is linked and fisheries values are shared to some extent between the Skeena and Nass estuaries.

The Upper Estuary (the upstream extent of tidal influence) is predominantly fresh water with a largely riverine geomorphic character. Fish habitats are characterized by braided sands and gravels at the western extreme grading to mudflats and lateral sedge flats downstream. Glacial boulder lag deposits in the western portion of the upper estuary likely provide some cover for fishes but for the most part good quality cover is limited to the braided sections and tributary junctions.

The Mid Estuary is a very dynamic environment due to the interaction of tides, Skeena River outflow and wind driven circulation. For the most part the mid estuary has a well mixed salinity structure but that does not preclude the presence of a stable salt wedge at some times of the year in certain locations. Bottom substrates are predominantly sand in the exposed areas of the main channel and silt and clay in the more protected embayments such as beside Robertson Bank. High current velocities in the western channel portion of the mid estuary favour the formation of mega-ripples and dunes contributing to the complex geomorphology and fish habitats of that zone. Map 1 (Appendix B) shows the location of the mega-ripples and dunes as the area marked by "A". Between "A" and "C" there is a deep scour hole likely formed by turbulent scour at the highest tides and currents. The scour hole was sampled with the larval net and was found to have an abundance of shrimp, Pacific Sandfish and other estuarine species as well as an abundance of fine particulate organic material and some salmon fry (although it was late in the season). This area is thought to have a unique and stable estuarine ecology and may have a stable salt wedge due to its depth and enclosed morphology. The fine particulate matter sequestered in the scour hole most likely contributes physical cover to this productive habitat unit as well as a source of nutrients.

The delta front of the estuary lies roughly on a line between Ridley Island in the north and Gibson Island at the entrance to Grenville channel in the south. The Skeena delta front is formed by sandy sediments out of Inverness, Marcus and Telegraph Passages that settles out

once they reach the relatively quiescent waters of the outer estuary. Shallow sandy banks form at the outer edge of the delta (at "C" and "B" on Map 1) and are some of the most productive rearing habitats in the estuary. Flora Bank ("B" on Map1), in particular, was noted by Hoos (1976) as one of the most productive habitats of the Skeena Estuary.

The Outer Estuary (see Appendix C. Map 2) is an extensive area of partially enclosed estuarine character with a distinct salinity structure and strong density gradient evidenced by the pronounced surface layering of diluted seawater over top of full strength sea water. The extent of the fresh water lens likely varies seasonally and yearly based on differences in climate and river runoff from both the Skeena and Nass Rivers. Sediments in the outer estuary are fine grained marine type sediments reflecting the more quiescent settling environment of the outer estuary and the fact that coarser sediments are selectively settled out in the mid and upper estuary (Hoos, 1976). The salinity profiling undertaken for this study indicates a plume of diluted sea water that projects out from the mid estuary in a somewhat laminar lens out beyond Melville Island. The profiling here also indicated some laminar instabilities in the plume at point "D" on map 1 that indicate there is a secondary mixing process causing delamination of the plume at that point. It is not clear whether that secondary mixing is the result of mixing between Skeena and Nass plumes or is the Mega-estuary plume mixing turbulently with incoming full strength seawater from the west.

#### *4.2.1 Material Transport*

Each year the Skeena River transports millions of tonnes of dissolved, suspended and bedload material to the estuary in a process that is collectively referred to as "Material Transport". This includes natural sources of sediments and organic materials and anthropogenic sources such as sewage treatment and industrial effluents and accidental inputs (oil spills, train derailments etc). The estuary is the final repository for most of this material and therefore the transport and more importantly the fate of material transported to the estuary is a primary management concern. Material fate and transport of anthropogenic inputs is the single most important management issue for the estuary.

Predicting environmental effects and designing mitigation / cleanup strategies for anthropogenic inputs to the estuary is very difficult given the poor state of knowledge around estuary material transport and ecology and the highly complex nature of the area. Each of the estuary subdivisions will have its own mechanisms for sorting and storing anthropogenic inputs making prediction of environmental effects very difficult.

Fishes, like eulachon, that rely on small scale channel hydraulics to deposit and incubate their eggs in backwater hydraulic settings could be seriously affected by solid phase contaminants that would tend to accumulate in those same backwater areas. The scour hole off Parry Point sampled during this study is a similar hydraulic setting where it is likely that anthropogenic inputs will be forced to accumulate in an area of very high biological activity.

### 4.3 Estuary Species Composition

All of the fish species, crustaceans (mainly Panadaliidae pink shrimp) and invertebrates observed in this study, together with species known to exist here (see Hoos, 1975), are potential prey for salmonids. Each of these species has a niche in the system and is therefore important to the whole ecosystem and in some way supports abundance of salmonids. Significantly, this study found the first actual record of Longfin smelt spawning in B.C. (Ramona de Graff, pers. comm). Larval Pacific lamprey were captured in abundance, and are a known food source for salmon.

Notable ecological details about some of these forage fish are as follows;

1. It now appears that eulachon larvae do not remain in the estuary for any extended period of time. Previous assumptions that eulachon larvae drift out to sea shortly after hatching are probably true. The capture of a single eulachon larvae in August is consistent with the occasional capture of mature adult eulachon in gillnets set for sockeye salmon in July (pers. obs). Whereas the majority of eulachon spawning occurs in the spring in March, this larvae was produced by a later spawning event. Eulachon eggs incubate in reduced flow areas between the upstream extent of tidal influence at Kasiks and the upstream extent of salinity near Kwinitza (Lewis et al 2001, Kelson 2010).
2. High densities of larval and spawning mature Longfin smelt are found in the fresh water, just above uppermost salinity in the estuary. These areas have tidally affected flows that are slowed and reversed with each tidal cycle. This productive zone for Longfins is also where some forms of anthropogenic inputs being transported downstream by the river would be slowed and accumulate. Although Longfins are non-commercial and unharvested they are an abundant forage fish that must be trophically significant, both as predator and prey.
3. There are now genetic markers allowing separation of larval Longfin smelt from eulachon.

#### 4.3.1 Baseline limitations of Genetic Identification Anna Elz, NOAA

A diagnostic assay depends on limited intra-species variability to differentiate species by assessing variation within and amongst species throughout their geographic range. This assay has been developed with a very limited baseline for all species and therefore is considered preliminary in its diagnostic capabilities (See Table 3 for baseline details). The ability to distinguish eulachon from Longfin smelt using this assay was dependent on variation at a particular base that the ApaI enzyme would recognize and cut. The samples that were initially identified as eulachon by RFLP in this study had variation at this cut site that matched eulachon sequence and was not represented in our

baseline. Our baseline had only two Longfin smelt specimens from British Columbia. It is not known if the variation exists in eulachon that would make them appear as Longfin smelt. This is the first test of the assay and more work will be required to expand the baseline and test unknowns. For now all we can say is 5 out of 50 were misidentified suggesting the assay is 90% effective, but that could change if the samples come from another area.

\*\*Separate enzymes cut white and night smelt but were not used for this study.

The assay is only as good as the baseline. Prior to this current work, our baseline was very deficient geographically for Longfin smelt in particular since there were only two from BC. The reason those samples "looked" like eulachon initially is because those five samples had a variable site in the cut region, that matched eulachon. The assay was developed based on other samples that did not exhibit that variation. So the bad news is I need to do more work to develop a better assay, and the good news is that now we have more samples in the baseline with the addition of your samples to help us do that.

#### *4.3.2 Species Use of the Skeena Estuary*

We now have a better idea of the distribution and abundance of some species in the estuary, especially in the upper areas of salinity and around the uppermost edge of salinity. Eulachon, for example, spawn in the uppermost part of the Skeena estuary at the furthest upstream extent of tidal influence down to the upstream extent of salinity. Spawning occurs mainly in March but in lesser, on-going spawning events into the summer. Eggs incubate in lower-flow areas attached to fine substrate (Lewis et al, 2009). Larvae drift out to sea immediately upon hatching and spend very little time in estuarine circulation. This level of knowledge is a minimum which should be developed for many other species. For example, little is known about halibut or Steelhead residence time and ecological relationships in the estuary, both very important species. River-specific knowledge of the life histories of forage fish life, suspected foundations of estuary ecology, is limited. We can say that there is a diversity and great abundance of forage fish species present in the Skeena estuary that undoubtedly helps to support predators.

#### *4.3.3 Valued Ecosystem Components*

The Canadian Environmental Assessment Agency defines Valued Ecosystem Components (VEC) as:

The environmental element of an ecosystem that is identified as having scientific, social, cultural, economic, historical, archaeological or aesthetic importance. The value of an ecosystem component may be determined on the basis of cultural ideals or scientific concern. Valued ecosystem components that have the potential to interact with project components should be included in the assessment of environmental effects.

Clearly the diversity and abundance of forage fish and other biota found in this study are ecologically important and potentially at-risk from anthropogenic inputs transported downstream or otherwise introduced in the Skeena estuary, and must therefore be considered as VECs.

## **5.0 Acknowledgements**

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## 6.0 References

Canadian Environmental Assessment Agency, Policy and Guidance Glossary of Terms, [www.ceaa-acee.gc.ca](http://www.ceaa-acee.gc.ca)

DeGraff, R. B.C. Shorespawners Alliance, pers. Comm.

Fishbase. <http://www.fishbase.org/search.php>

Gustafson, R. G, W.H. Lenarz, B.B. McCain, C.C. Schmitt, W.S. Grant, T. L. Builder, and R.D. Method. Status Review of Pacific Hake, Pacific Cod and Walleye Pollock from Puget Sound, Washington. NOAA Technical Memorandum NMFS-NWFSC-44 National Marine Fisheries Service. Seattle, WA. November, 2000

Hoos, L. M., 1976. The Skeena River Estuary: Status of Environmental Knowledge to 1975. Environment Canada Special Estuary Series No. 3.

Ichthyoplankton Information System. <http://access.afsc.noaa.gov/ichthyo/index.cfm>

Kelson J. Skeena 2010 Eulachon Habitat Use Study, March 2010. AFS Report to Department of Fisheries and Oceans, Prince Rupert, B.C. .

Lewis, A, J. Kelson, S. Faulkner, and I. Murphy. 2009. Skeena River Eulachon 2001. Consultant's report prepared for BC Hydro, Burnaby.

Thedinga, J.F., S.W. Johnson, and D.G. Mortensen. 2006. Habitat, age, and diet of a forage fish in southeastern Alaska: Pacific sandfish (*Trichodon trichodon*). Fishery Bulletin 104:631-637.

<http://www.fws.gov/oregonfwo/Species/Data/PacificLamprey/Documents/012808PL-FactSheet.pdf>

Quinn, T. P. The Behavior and Ecology of Pacific Salmon and Trout. American Fisheries Society, 2005.

## **7.0 Appendices**

*7.1 Appendix A. Spreadsheet of Original Fish Sample Data ...attached.  
Table of Fish Analyzed Genetically.*

7.2 *Appendix B. Photos of Selected Sorted Samples*











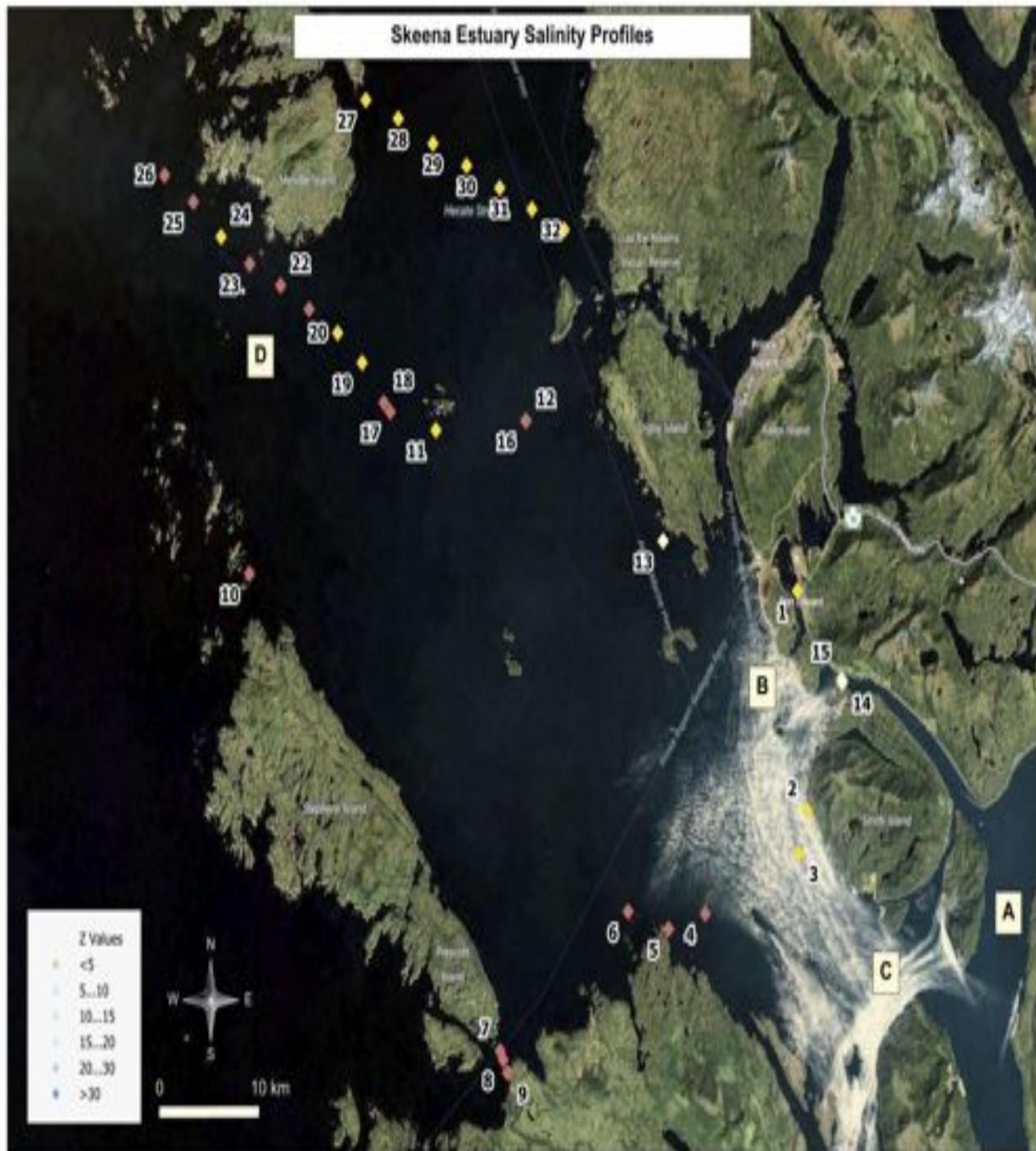








7.3 Appendix C Estuary Mapping



Map 1: Salinity profile location map.



Map 2. Estuary Delineation