

Early Marine Ecology of Pacific Salmon: Interactions with Sea Lice

by

Michael Harold Howard Price  
B.Sc., University of Victoria, 2003

A Thesis Submitted in Partial Fulfillment  
of the Requirements for the Degree of

MASTER OF SCIENCE

in the Department of Biology

© Michael Harold Howard Price  
University of Victoria

All rights reserved. This thesis may not be reproduced in whole or in part, by photocopy or other means, without the permission of the author.

## **Supervisory Committee**

Early Marine Ecology of Pacific Salmon: Interactions with Sea Lice

by

Michael Harold Howard Price  
B.Sc., University of Victoria, 2003

### **Supervisory Committee**

Dr. John D. Reynolds, Department of Biology; Department of Biological Sciences, Simon Fraser University

**Co-Supervisor**

Dr. Barry W. Glickman, Department of Biology

**Co-Supervisor**

Dr. Steve J. Perlman, Department of Biology

**Departmental Member**

Dr. John P. Volpe, School of Environmental Studies

**Outside Member**

## Abstract

### Supervisory Committee

Dr. John D. Reynolds, Department of Biology; Department of Biological Sciences, Simon Fraser University

**Co-Supervisor**

Dr. Barry W. Glickman, Department of Biology

**Co-Supervisor**

Dr. Steve J. Perlman, Department of Biology

**Departmental Member**

Dr. John P. Volpe, School of Environmental Studies

**Outside Member**

Pacific salmon (*Oncorhynchus* spp.) are key elements of ecological systems, and play an important role in the cultural foundation of human societies. All species of wild salmon face multiple, simultaneous threats, with habitat degradation likely playing a key role in survival. Open net-pen salmon farms can degrade important nursery marine habitat for wild juvenile salmon by disrupting natural salmonid host-parasite dynamics. The first two chapters in this thesis examine louse parasitism of wild juvenile chum (*Oncorhynchus keta*), pink (*O. gorbuscha*), and sockeye salmon (*O. nerka*) in relation to their marine migration past salmon farms. I compare sites of low and high exposure to salmon farms, and include two areas without farms on British Columbia's central and north coasts to assess baseline infection levels. Louse prevalence and abundance were lowest and most similar to natural baseline levels at low exposure sites, and highest at high exposure sites in all farm regions. A significantly greater proportion of the lice infecting juvenile chum and pink salmon were *Lepeophtheirus salmonis* at high exposure sites. *Caligus clemensi* was the principal louse species infecting all juveniles in

areas without salmon farms, and at low exposure sites within salmon farm regions; *C. clemensi* was also the dominant louse to infect juvenile sockeye that migrated past farms. Mixed-effects modelling results showed that exposure to salmon farms was the most consistent factor to explain the variation in louse infection levels, and support my hypothesis that salmon farms are a major source of sea lice on juvenile wild salmon in regions with salmon farms.

I discovered that juvenile sockeye at one particular location within the Georgia Strait hosted unusually high lice levels; this location was situated at a distance from salmon farms, but near a farm salmon processing facility. Upon further investigation, I found live sea lice, *Lepeophtheirus salmonis*, mucus, and fish tissue in effluent discharged from the processing facility. Sea lice transmitted from this source may pose a threat to wild salmon populations, and the release of potentially untreated offal, including blood water, is of considerable concern. These results form the third chapter in my thesis.

Given the challenges facing juvenile salmon in general, and sockeye from the Fraser River in particular (i.e., 2009 was the lowest return on record), and because poor habitat conditions within Georgia Strait are considered the major cause of the recent decline in Fraser River sockeye, this raises the question as to whether food limitations are a factor. The final chapter in my thesis examines the prey assemblage, diet composition, and foraging selectivity of juvenile sockeye, and investigates whether food limitations can be detected during early migration through Georgia Strait. Juvenile sockeye demonstrated high prey diversity, with preference for particular prey. Prey were more concentrated in the north, which may help explain migratory behavior of

juveniles through the study region, and temporal similarities in sockeye foraging success may reflect short-term food resource stability. Moreover, I could not find evidence of food limitations that might suggest juvenile sockeye were strongly food deprived during the years of this study.

Finally, my thesis explores how best to conserve salmon populations given the multitude of stressors. Because stressors often interact to produce compound effects and unpredictable results, ranking the overall threats in order of severity may not be useful. Instead, the most successful ranking system may be in terms of reducing harm where possible. For juvenile salmon during their early marine migration, risks posed by salmon farms can be more easily mitigated than the far-reaching effects on ocean productivity of climate change and ocean acidification, or predator removal. I recommend we begin here.

## Table of Contents

Supervisory Committee.....	ii
Abstract.....	iii
Table of Contents.....	vi
List of Tables.....	vii
List of Figures.....	ix
Acknowledgements.....	xi
Dedication.....	xii
General Introduction.....	1
Chapter 1.....	11
Evidence of farm-induced parasite infestations on wild juvenile salmon in multiple regions of coastal British Columbia, Canada.	
Chapter 2.....	31
Sea louse infection of juvenile Pacific sockeye salmon ( <i>Oncorhynchus nerka</i> ) in relation to marine salmon farms on Canada’s west coast.	
Chapter 3.....	58
Fish processing facilities: new challenge to marine biosecurity in Canada.	
Chapter 4.....	68
Zooplankton dynamics and prey selectivity of Fraser River sockeye salmon( <i>Oncorhynchus nerka</i> ) during their early marine migration in British Columbia, Canada.	
General Discussion.....	88
Bibliography.....	96

## List of Tables

### Chapter 1

Table 1.1. Summary of mean capture-site and biological data values for juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined at Bella Bella (BB), Finlayson (Fin), Broughton Archipelago (BA), and Georgia Strait (GS) during 2007-2008.....23

Table 1.2. Mean prevalence (P), abundance (A), and intensity (I) of sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined during 2007-2008 at Bella Bella (BB), Finlayson (Fin), Broughton Archipelago (BA), and Georgia Strait (GS) and their exposure to salmon farms (low or high)...24

Table 1.3. Percentages of sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) at different life stages: Copepodid (Cop), Chalimus A (Ch A), Chalimus B (Ch B), Pre-adult (Pre-A), adult (A), infecting juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon at Bella Bella (BB), Finlayson (Fin), and Georgia Strait (GS), and their exposure to salmon farms (low or high) .....25

Table 1.4. List of candidate models and resulting Akaike's Information Criteria (AIC) scores used to determine which factors most influence sea louse prevalence on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon.....26

Table 1.5. Summed Akaike's Information Criteria (AIC) weights ( $\sum\omega_i$ ) across the top model set to rank parameters by relative importance in predicting sea louse prevalence levels on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon.....27

### Chapter 2

Table 2.1. Stock proportion estimates and standard deviations for genetically identified juvenile sockeye salmon (*Oncorhynchus nerka*) caught on British Columbia's north coast (NC) and Discovery Islands (DI).....49

Table 2.2. Summary statistics and sea louse infection rates on juvenile sockeye salmon (*Oncorhynchus nerka*) caught either upstream (Up) or downstream (Down) of salmon farms at north coast (NC) or Discovery Islands (DI) region. All morphometric and abiotic values represent the mean, except sea lice infection rates.....50

Table 2.3. List of candidate models and resulting Akaike's Information Criteria (AIC) scores used to determine which factors most influence *Caligus clemensi* abundance on juvenile sockeye salmon (*Oncorhynchus nerka*).....51

Table 2.4. Summed Akaike's Information Criteria (AIC) weights ( $\sum \omega_i$ ) across the top model set to rank parameters by relative importance in predicting *Caligus clemensi* abundance on juvenile sockeye salmon (*Oncorhynchus nerka*).....52

Table 2.5. List of candidate models and resulting Akaike's Information Criteria (AIC) scores used to determine which factors most influence *Lepeophtheirus salmonis* abundance on juvenile sockeye salmon (*Oncorhynchus nerka*).....53

### Chapter 3

Table 3.1. Contents from effluent samples retrieved during five-minute collections from the outflow pipe of a farm salmon processing facility on Canada's west coast.....65

### Chapter 4

Table 4.1. Summary statistics (sample mean and standard error) for juvenile sockeye salmon examined during 2009 and 2010. The sample size for fish fork-length and mass was the same as the number of stomachs in 2009, and  $n = 93$  in 2010; the number of collection sites for sea surface temperature and salinity readings were 12 (2009) and 18 (2010).....81

Table 4.2. Zooplankton taxa identified during 2009 and 2010 plankton surveys; percent abundance (A) is the number of individuals of a given taxon divided by the total number of individuals of all taxa, and frequency of occurrence (F) is the frequency of a given taxon in the plankton.....82

Table 4.3. Prey taxa identified in juvenile sockeye stomachs during 2009 and 2010; relative abundance (A) is the number of individuals of a given taxon divided by the total number of individuals of all taxa, and frequency of occurrence (F) is the frequency of a given taxon in all sockeye stomachs.....83

## List of Figures

### Chapter 1

Figure 1.1. Study area, including 3 salmon farm regions (A-C), the non-salmon farm area of Bella Bella, and all associated sampling sites for juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined for sea lice in British Columbia during 2007-2008.....28

Figure 1.2. 95% Confidence Intervals +/- 2 SE of combined mean sea louse (*Lepeophtheirus salmonis* and *Caligus clemensi*) abundance on juvenile chum (*Oncorhynchus keta*) and pink salmon (*O. gorbuscha*) examined at sites of low (open circle) or high (solid circle) exposure to salmon farms for all years combined: B.B. is Bella Bella, Fin is Finlayson, B.A. is Broughton Archipelago, and G.S. is Georgia Strait.....29

Figure 1.3. 95% Confidence Intervals +/- 2 SE of combined mean sea louse (*Lepeophtheirus salmonis* and *Caligus clemensi*) abundance on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined among 4 areas of coastal British Columbia, and their associated farm salmon production in metric tonnes (MT); B.B. is Bella Bella, Fin is Finlayson, B.A. is Broughton Archipelago, and G.S. is Georgia Strait.....30

### Chapter 2

Figure 2.1. Sockeye collection sites relative to salmon farms. Downstream boundary encircles all sites situated downstream of at least one salmon farm given the direction of prevailing oceanic flow and fish movement; all other sites are considered upstream.....54

Figure 2.2. 95% Confidence Intervals +/- 2 SE of the means for *Caligus clemensi* abundance on sockeye salmon in regions with varying exposure to salmon farms. N.C. is north coast where there are no farms, D.I. upstream is Discovery Islands locations upstream of all salmon farms (open circle), and D.I. downstream is Discovery Islands locations downstream of all salmon farms (closed circle) on British Columbia's coast.....55

Figure 2.3. 95% Confidence Intervals +/- 2 SE of the means for *Lepeophtheirus salmonis* abundance on sockeye salmon in regions with varying exposure to salmon farms. N.C. is north coast where there are no farms, D.I. upstream is Discovery Islands locations upstream of all salmon farms (open circle), and D.I. downstream is Discovery Islands locations downstream of all salmon farms (closed circle) on British Columbia's coast.....56

Figure 2.4. Sea louse abundance (*Caligus clemensi* at top, and *Lepeophtheirus salmonis* at bottom) over time on Atlantic salmon at named salmon farms among the Discovery Islands during concurrent sockeye collection in 2007 and 2008 (shaded grey).....57

### Chapter 3

Figure 3.1. Collection site of effluent samples retrieved from a farm salmon processing facility on Canada's Pacific coast.....66

Figure 3.2. Effluent discharged from the outflow pipe of a farm salmon processing facility on Canada's Pacific coast.....67

#### **Chapter 4**

Figure 4.1. Study area of Georgia Strait and Johnstone Strait and all associated sampling sites for zooplankton and juvenile sockeye salmon (*Oncorhynchus nerka*) examined during 2009-2010.....84

Figure 4.2. Distribution of zooplankton density (m<sup>3</sup>) in relation to distance '0', the southern-most collection site in 2010.....85

Figure 4.3. Electivity scores (a species-specific measure of prey selection) for primary prey species identified in juvenile sockeye stomachs during 2009-2010. A value of -1.0 indicates lowest selectivity (i.e., never in the diet, but present in zooplankton samples), and 1.0 indicates the highest selectivity (i.e., present in the diet, but never in zooplankton samples).....86

Figure 4.4. Feeding index, expressed as percent body weight of stomach contents, in relation to the migration of juvenile sockeye through the study region during 2009-2010. Migration day is the number of days since the start of sampling in a given year.....87

## Acknowledgments

I wish to thank the Heiltsuk, Homalco, Kitasoo, and MTTC Nations in whose traditional territories the research occurred. I am grateful to C. Aries, S. Bergh, D. Brown, F. Campbell, T. Campbell, J. Eriksson, H. Humchitt, J. Lawson, K. Poppe, I. Reid, R. West, and A. Woods for assistance in the field; C. Carr-Harris, D. Kwaii, S. Latham, A. Morton, S. Proboszcz, T. Roscovich, and A. Rosenberger for technical assistance; D. Braun, B. Connors, C. Darimont, A. Gottesfeld, M. Hocking, M. Krkosek, C. Orr, W. Palen, and R. Routledge for analytical advice; and the biologists at the Fisheries and Oceans molecular genetics laboratory for genetic identifications of sockeye salmon.

I am indebted to the Raincoast Conservation Foundation for financial and project support, and the Natural Sciences and Engineering Research Council for an Industrial Post-graduate Scholarship. I also acknowledge project support from the British Columbia Pacific Salmon Forum, Coastal Alliance for Aquaculture Reform, David and Lucille Packard Foundation, Gordon and Betty Moore Foundation, Mountain Equipment Co-op Environment Fund, Pacific Salmon Commission, Redden Net, Ritchie Foundation, Sandler Foundation, SOS Marine Conservation Foundation, Vancouver Foundation, and Watershed Watch Salmon Society.

Finally, I wish to thank Dr. John Reynolds and Dr. Barry Glickman for their role as my academic supervisors, and Dr. John Volpe and Dr. Steve Perlman for serving on my committee. I am particularly grateful to Dr. Reynolds for his timely encouragement.

## Dedication

I dedicate this thesis in three parts:

The first part I dedicate to my life partner, *Clare*, who has remained a pillar of strength for me even during the most tumultuous of times; your support during this project will never be forgotten.

The second part I dedicate to my son, *Anian Lloyd William*; you inspire me to understand and protect our planet. The future of wild salmon undoubtedly rests in your hands.

The final part I dedicate to my parents, *Wally* and *Linda Price*, who have supported me throughout life.

## General Introduction

Pacific salmon (*Oncorhynchus* spp.) are key elements of ecological systems (Naiman *et al.* 2002; Gende *et al.* 2002; Hocking and Reynolds 2011), and play an important role in the cultural foundation of human societies (Campbell and Butler 2010). All species of wild salmon face multiple, simultaneous threats, which can include: hatcheries, pollution, harvest, introduced species, contaminants, habitat loss, salmon farms, and the overarching effects of climate change (BCPSF 2009; Healey 2011). Salmon in British Columbia (BC), Canada are thought to be at 50% of their historic abundance (Northcote and Atagi 1997), with at least 142 populations extinct, and 624 considered at high extinction risk as of 1996 (Slaney *et al.* 1996).

A wealth of knowledge regarding Pacific salmon has been amassed (e.g., Groot and Margolis 1991), yet we fail to understand many important details of their life history. For example, the early marine period remains poorly understood relative to the late marine and freshwater stages (Welch *et al.* 2009). Specifically, mortality agents responsible for survival during this period remain in question (Welch *et al.* 2011). It has long been thought that mortality incurred during the early marine migration of salmon is an important factor limiting overall abundance (Ricker 1976; Peterman 1982; Beamish *et al.* 2004). Marine survival is believed to be a function of early marine growth (Farley *et al.* 2007; Duffy and Beauchamp 2011), with mortality primarily occurring during two phases: soon after juveniles enter the ocean environment (predation-based), and after the first summer season when slower growing individuals have a higher probability of mortality (growth-based; Beamish and Mahnken 2001; Beamish *et al.* 2004). Both phases are

undoubtedly coupled with ocean conditions in general, and food resource availability in particular.

The quality of nursery habitat for juvenile salmon is deteriorating. Salmon migrate along the near-shore habitat of bays and inlets of the continental shelf during their first summer at sea (Quinn 2005). These nursery grounds are among the most highly productive ecosystems on earth, yet are also largely threatened by human activities (Halpern *et al.* 2009). In BC, the single largest nursery ground for juvenile salmon is the Strait of Georgia: a coastal sea that is also at the receiving end of waste discharges and contaminants from the activities of several million human inhabitants (Ross 2006). Juveniles can be exposed to the cumulative effects of pollution, fishing, introduction of invasive species, and climate change, all of which reduce habitat quality. Food-web dynamics appear to be deteriorating due to possible trophic mismatches (Johannessen and Macdonald 2009), and poor habitat conditions in Georgia Strait may have played a role in the recent decline of at least one species of salmon (i.e., sockeye; Peterman *et al.* 2010).

Juvenile salmon may be experiencing food limitations. Juvenile pink (*Oncorhynchus gorbuscha*), chum (*O. keta*), and sockeye salmon (*O. nerka*) primarily consume zooplankton during their first summer at sea (Healey 1980; Landingham *et al.* 1996). Zooplankton production in the Strait of Georgia has been declining since 2001, and a change in their community assemblage may also be underway (Johannessen and Macdonald 2009). Warming ocean temperatures as predicted by climate change may replace superior dietary sources of prey with less nutritious food for juvenile salmon, and promote a mismatch between the timing of the zooplankton biomass peak and the

presence or abundance of juveniles in this region (Mackas *et al.* 2007; Johannessen and Macdonald 2009; Healey 2011). Knowledge of the feeding habits and prey abundance for juvenile salmon during their migration through Georgia Strait is necessary to determine whether food limitations occur (Landingham *et al.* 1996; Schabetsberger *et al.* 2003), yet this information is incomplete and antiquated.

Open net-pen salmon farms are exacerbating habitat degradation for juvenile salmon. Salmon farming is the fastest growing agriculture sector globally (FAO 2008). In BC, there are more than 130 salmon farm tenures, and nearly all are situated in near-shore marine areas along juvenile salmon migration routes (BCPSF 2009). The open net-pens that are used to grow salmon on farms allow for the direct exchange of water and effluent with the surrounding habitat. As such, waste from farms in the form of untreated nutrients and potentially harmful chemicals is discharged into the surrounding marine environment. This can have considerable local effects on marine life (Goldburg and Naylor 2005), and produce broad impacts beyond salmon at both the low and high ends of the food web (Naylor *et al.* 2003). Salmon farm waste increases sediment organic content, and can elevate levels of heavy metals in fish prey and predators through contaminant cycling (Debruyne *et al.* 2006). Importantly, the direct flow of water through net-pens enables the transmission of fish pathogens between farm and wild salmon.

The transmission of pathogens to wildlife frequently occurs where host populations are concentrated into dense aggregations (Daszak *et al.* 2000; McCallum and Dobson 1995). Salmon farms are reservoirs of host populations, and the intensive growing conditions facilitate the

amplification of pathogens (Murray and Peeler 2005; Murray 2008). For example, salmon farms hold domestic fish, mainly Atlantic salmon (*Salmo salar*), in high densities for months in the same location (i.e., 15-30 kg/m<sup>3</sup> for up to 24 months; Marine Harvest Corporate 2008). These crowded conditions promote pathogen transmission within the farm, and open net-pens enable the release of pathogens to the surrounding environment. Farm fish hosting even small numbers of lice can collectively produce large numbers of louse eggs and infectious larvae (Heuch and Mo 2001; Heuch *et al.* 2005; Orr 2007). Although the number of fish diseases that infect salmon farms is extensive, evidence of transmission to wild populations is limited; this may be because diseased organisms are difficult to detect if not tracked (Gozlan *et al.* 2006). One easily detectable pathogen is the sea louse, an ectoparasite commonly associated with farm-origin epizootics (Marty *et al.* 2010), and depressed adjacent wild salmon populations in regions with salmon farms (Krkosek *et al.* 2007a, 2011a; Connors *et al.* 2010; Krkosek and Hilborn 2011).

Sea lice are important pathogens of salmon. Caligid sea lice (mainly *Lepeophtheirus salmonis* and *Caligus* spp.) are the most widespread marine parasites affecting domestic and wild fish, and have now emerged as important pathogens in many coastal marine areas (Costello 2006, 2009; Krkosek 2010). *Lepeophtheirus salmonis* and *Caligus clemensi* are the two most common species found on wild salmon in BC. The impact of sea lice is host size dependent, with fewer lice required to induce negative effects on smaller fish (Bjorn and Finstad 1997). Sea lice feed on surface tissues of their hosts, which can lead to many problems especially for small juvenile fish (Costello 2006; Pike and Wadsworth 2000). Sea lice can compromise osmoregulation (Bjorn and Finstad 1997), induce behavioral changes that increase predation risk (Krkosek *et al.* 2011b),

reduce growth rates and, in sufficient numbers, result in host death (Costello 2009; Morton and Routledge 2005; Krkosek *et al.* 2006). Sea lice also have been shown to serve as vectors for the spread of fish diseases (Nese and Enger 1993). For example, Atlantic salmon that were exposed to salmon lice from infectious salmon anaemia-infected fish, suffered high mortalities (Nylund *et al.* 1994).

Salmon farms transmit sea lice to wild juvenile salmon. Acute sea lice infestations are common on farmed salmon in Europe and North America (Penston *et al.* 2008; Marty *et al.* 2010). It is generally accepted that sea lice on farm salmon were transferred from wild fish, because farm salmon enter the marine environment without lice (Marine Harvest Corporate 2008). However, debate continues over whether the stationary, high host-density populations on farms in nearshore areas reverse the transmission to out-migrating juvenile wild salmon. Until recently, sea lice epizootics have been rare in wild fish populations (Costello 2009), though brief localised outbreaks have occurred (Parker and Margolis 1964; Beamish *et al.* 2009). Wild juvenile salmon in areas not exposed to salmon farms routinely host low levels of sea lice (i.e., < 5% of juveniles infected; Morton *et al.* 2004; Krkosek *et al.* 2007b; Gottesfeld *et al.* 2009), although Beamish *et al.* (2009) describe a sea lice outbreak on juvenile salmon in an area far from salmon farms. Concomitantly, juvenile wild salmon swimming past farms are frequently infected with sea lice (Tully *et al.* 1999; Heuch *et al.* 2005; Krkosek *et al.* 2005a, 2006). Despite the strong correlation between sea lice on wild juvenile salmon and the presence of salmon farms, there remains a lack of information on lice levels from farmed salmon, which could be compared with concurrent data on wild juvenile salmon.

Alternative explanations beyond farm-origin lice have been suggested for the repeated occurrence of sea lice on wild juvenile salmon in fish farming regions. Factors such as temperature and salinity are often cited because sea louse growth in lab-based trials depends strongly on temperature and salinity (Pike and Wadsworth 2000; Costello 2006). The presence and abundance of wild fish hosts have also been cited (e.g., Beamish *et al.* 2007; Beamish *et al.* 2009), and combined with the above potential factors, adds to the uncertainty of sea louse origin on wild juvenile salmon.

Juvenile pink and chum salmon are the most frequently reported species infected with lice. Most investigations on sea lice infections of wild juvenile salmon have focused on pink and chum in the Broughton Archipelago, where the first epizootic in BC was observed (Morton and Williams 2003). Pink salmon populations have declined in this region, and there is evidence that farm-origin lice may be partly responsible (PFRCC 2002; Krkosek *et al.* 2007a, 2011a; Ford and Myers 2008; Krkosek and Hilborn 2011). Pink and chum salmon are of particular concern because of their small size and undeveloped immune system at the time of sea entry, which may increase their sensitivity to sea lice infection (Morton *et al.* 2004; Bjorn and Finstad 1997).

Recent research has raised concern that sea lice from salmon farms may infect juvenile sockeye salmon in northern Georgia Strait (Morton *et al.* 2008). This region is home to the northeast Pacific's largest salmon farm industry, and hosts one of the largest migrations of salmon in the world (primarily to and from the Fraser River; Hartt and Dell 1986). Sockeye is the Pacific

Ocean's most economically and culturally important salmon species, and several populations from the Fraser River are endangered (IUCN 2008). Productivity of Fraser River sockeye has been declining since the early 1990s, with 2009 being the lowest on record, prompting the Canadian government to launch a Judicial Inquiry to investigate the cause of the decline and identify imminent threats to their survival (Cohen Commission 2010). Determining whether sockeye are at risk to sea lice transmission from salmon farms during their early marine migration is highly relevant to conservation and management efforts.

In addition to questions about the role of salmon farms in pathogen transmission, there are also questions about potential impacts of processing facilities. Effluent released from facilities processing farm salmon in Europe are considered a risk factor in the spread of fish diseases (Vagsholm *et al.* 1994; Jarp and Karlsen 1997). Untreated blood, tissue, and mucus from infected fish pose a serious risk of disease transmission to wild fish (Totland *et al.* 1996), and the disinfection of farm salmon waste from processing has been effective at diminishing disease transmission in some salmon farm regions of Europe (Murray *et al.* 2010). Because processors and salmon farms are often separated by large distance, three under-appreciated disease processes might occur: i) pathogens may be transferred to new regions, ii) extant but discrete pathogen populations may experience genetic recombination with new strains, leading to resistance of commonly used chemical treatments, and iii) non-target populations, such as wild salmon, will likely face unpredictable interaction effects with the introduced pathogens. While farm-origin sea lice populations rise and fall as salmon farms are stocked, harvested, and fallowed, processing plants have the capability of continuous pathogen release. Examining

whether farm salmon processing facilities pose a pathogen risk to wild fish is another important step towards understanding current threats to the early marine survival of salmon.

My thesis investigates some potential stressors that may be influencing the early marine survival of salmon in BC. Chapter 1 examines the multiple potential causes and correlates of sea lice infections on wild juvenile pink and chum salmon from four regions in British Columbia. Because there is a paucity of information on lice levels in salmon farm regions beyond the Broughton Archipelago, I extend this comparative investigation to include the salmon farm regions of Finlayson to the north and Georgia Strait to the south. I also compare lice levels on juveniles in each region to an area without farms (Bella Bella). Factors such as temperature, salinity, and exposure to salmon farms are tested for their influence on sea lice infection levels using mixed-effects modelling. All samples were collected and examined for sea lice during 2007 and 2008, and the initial manuscript was submitted to the Canadian Journal of Fisheries and Aquatic Sciences prior to my enrollment at the University of Victoria (January 2010). As a registered graduate student, I resubmitted the manuscript with major revisions to the Methods, Results, and Discussion sections, stemming primarily from a complete reanalysis of the data using mixed effects modelling.

Juvenile sockeye were incidentally caught in the Georgia Strait region during the investigation of Chapter 1; these fish hosted the highest sea louse infection levels. As such, Chapter 2 examines parasite infection of wild juvenile sockeye. I compare infection rates on fish from locations that vary in their exposure to farms within the Georgia Strait, and I compare infection levels to a

region without salmon farms (north coast). I use molecular genetics techniques to determine the origins of the juvenile sockeye, and employ mixed-effects modelling to examine factors that best explain sea lice abundance (i.e., temperature, salinity, exposure to salmon farms). All samples were collected and examined for sea lice during 2007 and 2008, and genetic analyses were performed by biologists at the Fisheries and Oceans molecular genetics laboratory, prior to my enrollment at the University of Victoria. As a registered graduate student, I assembled and analyzed the data, and wrote the entire paper that led to the submission and successful publication of the manuscript in Public Library of Science ONE.

I discovered that juvenile sockeye at one particular location within the Georgia Strait hosted unusually high lice levels. This location was situated at a distance from salmon farms, but near a farm salmon processing facility. Chapter 3 examines whether sea lice could survive travel from salmon farms to a processing facility, mechanical disturbance during processing, and final treatment of effluent before release into the marine environment. All work for this chapter was performed while I was a registered student.

Given the challenges facing juvenile salmon in general, and sockeye from the Fraser River in particular (i.e., 2009 was the lowest return on record), and because poor conditions within Georgia Strait are considered the major cause of their recent decline (Peterson *et al.* 2010), this begs the question of whether food limitations are a factor. Chapter 4 examines the prey assemblage, diet composition, and foraging selectivity of juvenile sockeye, and investigates whether food limitations can be detected during their early migration through Georgia Strait.

Except for the collection and analyses of samples during 2009, all work for this chapter was performed while I was a registered student.

## Chapter 1

Evidence of farm-induced parasite infestations on wild juvenile salmon in multiple regions of coastal British Columbia, Canada.

Published in slightly modified form in Price, M.H.H., Morton, A., and Reynolds, J.D. 2010. Evidence of farm-induced parasite infestations on wild juvenile salmon in multiple regions of coastal British Columbia, Canada. *Canadian Journal of Fisheries and Aquatic Science* 67: 1925-1932.

## Abstract

Salmon farms are spatially concentrated reservoirs of fish host populations that can disrupt natural salmonid host-parasite dynamics. Sea lice frequently infect farm salmon and parasitize sympatric wild juvenile salmonids, with negative impacts on survival in Europe and Pacific Canada. I examined louse parasitism of wild juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon from three salmon farming regions in British Columbia (Finlayson, Broughton Archipelago, and Georgia Strait). I compared sites of low and high exposure to farms, and included an area without farms (Bella Bella) to assess baseline infection levels. Louse prevalence and abundance were lowest and most similar to natural baseline levels at low exposure sites, and highest at high exposure sites in all farm regions. A significantly greater proportion of the lice were *Lepeophtheirus salmonis* at high exposure sites. Exposure to salmon farms was the only consistently significant factor to explain the variation in prevalence data, with a secondary role played by salinity. My results support the hypothesis that salmon farms are a major source of sea lice on juvenile wild salmon in salmon farming regions, and underscore the importance of using management techniques that mitigate threats to wild stocks.

## Introduction

Disease outbreaks are an increasing threat to wildlife, exacerbated by increases in the human population and domesticated animals (Macdonald and Laurenson 2006; Thirgood 2009). The most common route of transmission to wildlife is from artificial reservoirs of host populations (McCallum and Dobson 1995; Daszak *et al.* 2000). Marine salmon farms located along near-shore wild salmon migration routes provide spatially concentrated host populations that can

serve as reservoirs and perturb the dynamics of natural salmonid host-parasite systems (Krkosek *et al.* 2006, 2009; Costello 2009). Sea lice (*Lepeophtheirus salmonis* and *Caligus* spp.) frequently infect farm salmon, and many studies in Europe have identified farm-origin lice as those that parasitize sympatric wild salmonids (MacKenzie *et al.* 1998; Tully *et al.* 1999; Bjorn and Finstad 2002). Moreover, parasite outbreaks from salmon farms have been implicated in the collapse of wild sea trout (*Salmo trutta*) and Atlantic salmon (*Salmo salar*) populations in Norway, Scotland, and Ireland (McVicar 1997, 2004).

In Pacific Canada, recurrent parasite infestations from farms to wild juvenile pink (*Oncorhynchus gorbuscha*) and chum (*O. keta*) salmon have been well documented in the Broughton Archipelago (Krkosek *et al.* 2005a, 2006; see my Figure 1.1), where the first epizootic in British Columbia (BC) was observed (Morton and Williams 2003). Pink salmon populations have shown a general decline in this region, and there is evidence that farm-origin lice may be partly responsible (PFRCC 2002; Krkosek *et al.* 2007a, 2011a; Ford and Myers 2008; Krkosek and Hilborn 2011). Additionally, a recent investigation has shown parasite outbreaks on wild salmon in a salmon farm region south of the Broughton - in nearby Georgia Strait (Morton *et al.* 2008). Given the current intensity of farmed salmon produced in BC, and the proposed expansion of the industry, there is concern that lice outbreaks and negative impacts on wild salmon populations could occur elsewhere.

Alternative explanations have been suggested for the origins of sea lice on wild juvenile salmon in fish-farming regions (i.e., Brooks 2005; Beamish *et al.* 2007; Jones and Hargreaves 2007).

Factors such as temperature, salinity, and presence and abundance of wild fish hosts have been cited. Adding to this uncertainty is a paucity of information on lice levels in farm regions beyond the Broughton Archipelago. Additionally, there is a lack of information on lice levels in regions without salmon farms, which could be compared with concurrent data gathered in active farm regions. Moreover, no investigation has examined the relationship between lice levels on wild juveniles and the total amount of salmon produced on farms in a region.

In this study I examine multiple potential causes and correlates of lice infections on juvenile chum and pink salmon from four regions in BC. I extend my comparative investigation beyond the Broughton Archipelago to include Bella Bella (an area without salmon farms), and the salmon farming regions of Finlayson to the north, and Georgia Strait to the south.

## **Materials and methods**

I collected early marine phase juvenile chum and pink salmon from four regions along the BC coast during March to June 2007 and 2008 (Figure 1). Capture locations were selected based on the probability of exposure of juvenile salmon to active salmon farms, categorized as: high (< 1 km from active farms), or low (4 to 40 km upstream from farms; Figure 1). Some low exposure sites were relatively near an active salmon farm (4 km). However, these sites were situated along migration corridors upstream of the predominant flow from farms or across large channels that juveniles at the time of capture are known not to cross. Thus, I considered the exposure probability at those sites to be low. This way of categorizing exposure matches similar studies within the Broughton Archipelago and Georgia Strait (Morton *et al.* 2004, 2005, 2008). It would

not have been appropriate to treat probability of exposure as a continuous variable based on distance from the nearest farm, because that would have ignored the movements of fish from upstream (pre-exposure) to downstream (post-exposure) of farms. I determined the activity status of farms (i.e., active or fallow) and annual production harvest during the years of study from the Oceans and Marine Fisheries Branch of the British Columbia Ministry of Environment (BCMOE 2009; Figure 1.1). Sites near farms that were fallow in a given year were considered low exposure. Dates and frequency of surveys varied slightly between regions: Finlayson, 23 April - 22 June (bi-weekly), 2008; Bella Bella, 17 April - 15 June (weekly), 2007-2008; Broughton Archipelago, 20 March - 15 May (bi-weekly), 2007; Georgia Strait, 22 April - 14 June (bi-weekly), 2007-2008. At each site, juveniles were corralled by beach seine (50 m long, 6 mm mesh) from a boat, and in all regions except Georgia Strait, subsets of 30-100 juveniles per set were haphazardly selected and live-sampled for sea lice using methodology described by Krkosek *et al.* (2005b). Because this technique broadly identifies chalimus-stages of lice into only two stages and not by species, I modified this approach in Finlayson and Bella Bella by euthanising and collecting only those juvenile salmon that hosted a louse. All infected juveniles were frozen and sent to a lab for louse and host species identification, and fork length and weight measurements as described by Morton *et al.* (2008). To assess observer accuracy during non-lethal sampling, I also euthanised 3 juveniles per sampling day in Finlayson and Bella Bella (n = 100) that were judged to be louse-free, and later assessed them for louse presence using a dissecting microscope; no fish had lice. Only adult and copepodid stages of sea lice were identified in the Broughton Archipelago (for reasons explained above; but all lice were counted), and fish species and fork length were recorded without weights according to non-lethal field

assessment methods (Krkosek *et al.* 2005b). In Georgia Strait, entire subsets of juveniles (30-50 per site/week) were euthanized and lethally assayed for sea lice as described above. I recorded sea surface salinity and temperature at each collection site per sampling event among all regions using a calibrated YSI 85 multi-function meter. Measures of lice infection rates are as follows: Prevalence is the number of hosts infected with lice (expressed as a percentage), Abundance is the total number of lice divided by the total number of hosts (infected and uninfected), and Intensity is the mean number of lice per infected host (Margolis *et al.* 1982).

I was interested in which factors most influence whether a sea louse infects a juvenile salmon in BC. Accordingly, based on the literature cited in the Introduction, I formulated a priori hypotheses relating fish capture sites to the prevalence of sea lice on juveniles captured at those sites. Specifically, I hypothesized that fish from locations that were more exposed to farms would have higher louse prevalence, and that high temperature and salinity would also be correlated with high lice loads (because sea louse growth in lab-based trials depends strongly on temperature and salinity; Pike and Wadsworth 2000; Costello 2006).

I used generalized linear mixed effects modelling to account for the hierarchical nature of the sampling, where multiple sampling events at a given location were treated as random factors nested within location, which itself was a random factor nested within a region. I included exposure category (low or high) nested within region, temperature, salinity, and fork length as fixed factors to examine their influence on sea louse infection levels on juvenile salmon. My initial model selection included analyses for each louse and host species, respectively; however,

because results were broadly similar for each analysis, I combined louse prevalence and host species for simplification of presentation (minor differences are discussed in the Results). Thus, for the model, I averaged prevalence for combined sea louse species (*L. salmonis* and *C. clemensi*) for all host individuals (pink and chum) within a sampling event (i.e., replicate; n = 296), and transformed prevalence data using an arcsine square root function to correct for unequal variances and non-normality. I tested a set of candidate models using Akaike's Information Criterion (AIC), and then evaluated  $\Delta AIC$  to select the best approximating model(s). I made appropriate inference using  $\Delta AIC < 4$  to describe the top model set. Finally, I summed Akaike weights ( $\omega_i$ ) across the top model set for each variable to rank them by importance (Burnham and Anderson 1998; Anderson *et al.* 2001).

I performed a Chi-square test to examine whether the ratio of louse species abundances change in accordance to a given salmon farm region and associated differences in farm salmon production. I generated all analyses using SPSS 16.0 for Mac (SPSS 2007).

## Results

I assessed a total of 13 426 juvenile chum and pink salmon over 296 sampling episodes for sea louse parasitism across the 4 regions during 2007-2008. Juvenile salmon were largest in Georgia Strait and smallest in the Broughton Archipelago, and both pink and chum salmon were larger on average at high exposure sites in all farm regions except pinks in Georgia Strait, where they were larger at low exposure sites (Table 1.1). Sea surface salinity and temperature were higher on average at sites of high exposure than at sites of low exposure.

Louse prevalence and abundance were lowest for both chum and pink salmon in all farm regions at sites of low exposure, and most similar to Bella Bella where there are no farms (Table 1.2; Figure 1.2). In a comparison among the four regions, combined louse abundance was highest in Georgia Strait where salmon production is greatest (Figure 1.3). Increases in combined louse abundance between sites of low and high exposure ranged from a 2.4-fold increase at Georgia Strait to 7.1-fold increase at Finlayson to 30.5-fold increase in the Broughton Archipelago. A greater proportion of the lice were *L. salmonis* at sites of high exposure ( $\chi^2 = 3.814$ ,  $df = 1$ ,  $p = 0.000$ ), and lice at all locations were dominated by larval stages (copepodid and chalimus; Table 1.3).

Model selection and multi-model inference suggested that exposure + salinity was the best predictor of louse prevalence on juvenile salmon given the set of candidate models (Table 1.4). Specifically, louse prevalence increased at sites of high exposure to salmon farms, and this was most prominent in the regions with the highest salmon production (Figure 1.3); 3 of 3 models in the top model set (0-4  $\Delta$ AIC) contained exposure.

Summing the Akaike weights across top models ranked the variable exposure ( $\sum \omega_i = 0.974$ ) higher than salinity and temperature by factors of 2.5 and 3.8, respectively (Table 1.5). Although mixed-effects modelling results were broadly similar for each louse and host species, an exception was an increase in the positive effect of chum salmon host-length on the prevalence of *C. clemensi*.

## Discussion

My study shows associations between salmon farms and infestations of sea lice on wild juvenile salmon across a large area of coastal BC. Specifically, I show regional differences in parasitism of juvenile salmon between areas with and without salmon farms, as well as within-regional differences between sites of differing exposure levels. Within salmon farmed regions, juveniles at low exposure sites hosted fewer sea lice and were most similar in infection levels to regions without salmon farms. Overall, exposure to farms was the most important factor to explain louse prevalence. Finally, the proportion of *L. salmonis* infection increases in concert with farm salmon production.

Because louse parasitism of juvenile salmon at low exposure sites in active farm regions is most similar to levels in a region that lacks salmon farms, this suggests a ‘baseline’ designation that can enable regional comparisons. Juvenile chum and pink salmon examined at sites of low exposure in Finlayson and the Broughton Archipelago hosted spatially uniform louse prevalence averaging less than 5%. These rates are most similar to Bella Bella where farms are absent (3.5%), and correspond with those reported elsewhere in coastal BC without farms (Morton *et al.* 2004; Krkosek *et al.* 2007b; Gottesfeld *et al.* 2009). However, juveniles at low exposure sites in Georgia Strait hosted higher louse levels than all other peripheral areas; though levels were significantly lower than in high exposure locations within the region. The large number of farms in this area, the high complexity of waterways, and evidence of long-distance transmission capability of farm-origin lice (> 30 km; Krkosek *et al.* 2006; Costello 2009), suggest that louse transmission in this region confounds point sources as previously described (Morton *et al.* 2008).

The consistent relationship between elevated louse levels near salmon farms over all regions examined strongly suggests farm-induced parasite transmission to wild fish. Farm fish hosting even small numbers of lice can collectively produce large numbers of louse eggs and infectious larvae (Heuch and Mo 2001; Heuch *et al.* 2005; Orr 2007). Both juvenile chum and pink salmon hosted elevated lice levels in all regions and years at sites of high exposure compared with sites of low exposure. These results are consistent with previous research in farm areas of Europe (Tully *et al.* 1999; Bjorn and Finstad 2002) and locally in the Broughton Archipelago and Georgia Strait (Krkosek *et al.* 2005a, 2006; Morton *et al.* 2008). I add to this evidence, a 7.1-fold increase in louse abundance near farms in the northern region of Finlayson compared to sites of low exposure. Although this is the first demonstration of elevated lice levels on BC's north-central coast, the lower parasitism compared to other farm areas is most likely due to low salmon production in the region.

Although environmental parameters have been considered contributors to elevated louse parasitism of juvenile salmon, the data I present here suggest they are not the primary factors predicting prevalence levels in areas that are exposed to open net-pen salmon farms. Other work has shown that sea louse growth is strongly dependent on, and positively correlated with, salinity and temperature (Pike and Wadsworth 2000; Costello 2006). However, I found only moderate positive associations between salinity and louse prevalence, and only at sites of high exposure to salmon farms. Size (length) of juveniles also did little to predict louse prevalence. Instead, my analyses show that exposure to farms was the most important factor to explain louse prevalence.

Moreover, louse abundance is coupled with the amount of salmon produced in a given farm region. For example, regional louse abundance (combined low and high exposure sites) increased from 0.13 at Finlayson, to 0.24 in the Broughton Archipelago, to 0.65 in Georgia Strait, with associated farmed salmon production of 1 911 MT, 16 174 MT, and 17 005 MT, respectively.

A comparison of infections by the two louse species provides further insights into the potential for salmon farms to alter natural parasite dynamics. Juvenile salmon at sites of low exposure in all regions were most infected by *C. clemensi*, which is consistent with other BC areas where farms are absent (Morton *et al.* 2004; Krkosek *et al.* 2007b; Gottesfeld *et al.* 2009). This species is not salmon-specific, unlike *L. salmonis*. However, parasitism by *L. salmonis* increased in all regions at sites of high farm exposure, and became the dominant louse infecting juveniles near Broughton and Georgia Strait farms where farmed salmon production is highest. This proportional shift may contribute to the relationship between increased fish aquaculture intensity and decreasing wild salmon populations observed in Europe and eastern and western Canada by Ford and Myers (2008). *L. salmonis* are locally associated with juvenile chum and pink mortality (Morton and Routledge 2005; Krkosek *et al.* 2006), and have been implicated in contributing to the population collapse of pinks in the Broughton Archipelago (PFRCC 2002; Krkosek *et al.* 2007a, 2011a; Krkosek and Hilborn 2011). Accordingly, these data should alert managers to the potential for high juvenile salmonid mortality and associated population level impacts on numerous wild salmon stocks migrating through the high-intensity farm salmon production region of Georgia Strait.

### *Conservation implications*

Sea lice from salmon farms threaten vulnerable wild salmon populations in BC (Krkosek 2010), heightening the urgency required for Canada to develop an effective conservation-based salmon aquaculture policy. Infection levels are correlated with the amount of salmon produced in a given farm region; the alternative explanations beyond farm-origin lice that I tested here have less support. These findings should concern resource managers, as current wild salmon populations on the BC coast are under multiple human stressors, and many populations are at low levels (English *et al.* 2006; Price *et al.* 2008). Moreover, salmon farms have been specifically implicated in the decline or collapse of several local wild salmon populations (Krkosek *et al.* 2007a, 2011a; Ford and Myers 2008; Connors *et al.* 2010; Krkosek and Hilborn 2011). Given the increased production and site expansion proposed by the salmon farm industry, associated effects from farms may intensify and ultimately challenge the sustainability of ecosystems and economies along BC's entire coast (Krkosek 2010). Threats from salmon farms to wild salmon can be mitigated by reducing the number of fish per farm, limiting the number of farms in a region, and moving farms from migration routes and juvenile salmon habitats as has been implemented in some wild salmon sensitive areas of Norway (Heuch *et al.* 2005; Krkosek 2010). Ultimately, a switch to closed-containment aquaculture offers the best solution to the problem of transmission of diseases to wild fish.

**Table 1.1.** Summary of mean capture-site and biological data values for juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined at Bella Bella (BB), Finlayson (Fin), Broughton Archipelago (BA), and Georgia Strait (GS) during 2007-2008.

Region	Exposure	Salinity	Temperature	Species	Fork length	Mass
BB	low	20.1 ‰	10.6 °C	Chum	4.79 cm (0.02)	1.28 g (0.02)
				Pink	4.68 cm (0.03)	1.21 g (0.04)
Fin	low	25.2 ‰	9.4 °C	Chum	4.75 cm (0.05)	0.70 g (0.00)
				Pink	4.34 cm (0.03)	1.20 g (0.16)
	high	26.3 ‰	9.5 °C	Chum	5.23 cm (0.07)	2.27 g (0.22)
				Pink	4.80 cm (0.05)	1.18 g (0.10)
BA	low	27.6 ‰	10.4 °C	Chum	4.19 cm (0.17)	–
				Pink	3.76 cm (0.24)	–
	high	21.5 ‰	9.5 °C	Chum	4.48 cm (0.31)	–
				Pink	3.92 cm (0.26)	–
GS	low	24.9 ‰	10.4 °C	Chum	5.41 cm (0.45)	2.19 g (0.08)
				Pink	5.87 cm (0.76)	2.54 g (0.17)
	high	27.6 ‰	12.1 °C	Chum	5.93 cm (0.39)	2.71 g (0.08)
				Pink	5.61 cm (0.67)	2.19 g (0.07)

Note: Measurement units include: salinity (‰), temperature (°C), fork length (cm), and mass (g); Standard error for mean fork length and mass are in parentheses.

**Table 1.2.** Mean prevalence (P), abundance (A), and intensity (I) of sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined during 2007-2008 at Bella Bella (BB), Finlayson (Fin), Broughton Archipelago (BA), and Georgia Strait (GS) and their exposure to salmon farms (low or high).

Chum											
Region	Year	Exposure	# Fish	<i>L. salmonis</i>			<i>C. clemensi</i>			Combined prevalence*	Combined abundance
				P	A	I	P	A	I		
BB	2007	low	1 504	2.1	0.0	1.0	2.2	0.0	1.1	4.2	0.0
	2008	low	1 916	0.5	0.0	1.0	2.5	0.0	1.0	2.9	0.0
Fin	2008	low	317	0.0	0.0	0.0	1.3	0.0	1.0	1.3	0.0
		high	372	3.2	0.1	2.0	16.1	0.2	1.0	19.1	0.3
BA	2007	low	910	0.5	0.0	1.0	1.3	0.0	1.2	1.8	0.0
		high	717	13.5	0.2	1.2	15.1	0.2	1.4	25.9	0.4
GS	2007	low	674	15.6	0.2	1.4	19.9	0.3	1.4	29.8	0.5
	2008	low	169	7.1	0.1	1.3	10.1	0.1	1.4	15.4	0.2
	2007	high	884	18.4	0.3	1.6	24.8	0.3	1.4	37.3	0.6
	2008	high	635	23.6	0.6	2.4	17.2	0.4	1.9	37.2	1.0

Pink											
Region	Year	Exposure	# Fish	<i>L. salmonis</i>			<i>C. clemensi</i>			Combined prevalence*	Combined abundance
				P	A	I	P	A	I		
BB	2007	low	479	1.3	0.0	1.0	2.3	0.0	1.1	3.5	0.0
	2008	low	955	0.4	0.0	1.0	2.7	0.0	1.0	3.1	0.0
Fin	2008	low	774	0.3	0.0	1.0	4.3	0.0	1.0	4.5	0.0
		high	741	3.5	0.1	4.6	14.7	0.2	1.0	18.5	0.2
BA	2007	low	473	0.8	0.0	1.0	0.2	0.0	1.0	0.8	0.0
		high	697	20.4	0.2	1.2	24.2	0.4	1.5	37.7	0.6
GS	2007	low	82	20.7	0.3	1.3	15.9	0.2	1.5	32.9	0.5
	2008	low	79	8.8	0.1	1.1	11.4	0.1	1.1	19.0	0.2
	2007	high	538	37.4	0.7	1.8	20.3	0.3	1.4	48.9	1.0
	2008	high	510	12.7	0.2	1.4	18.0	0.3	1.7	27.3	0.5

Note: Combined Prevalence\* includes *L. salmonis*, *C. clemensi*, and unidentified chalimus A and B stages.

**Table 1.3.** Percentages of sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) at different life stages: Copepodid (Cop), Chalimus A (Ch A), Chalimus B (Ch B), Pre-adult (Pre-A), adult (A), infecting juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon at Bella Bella (BB), Finlayson (Fin), and Georgia Strait (GS), and their exposure to salmon farms (low or high).

Region	Exposure	<i>Lepeophtheirus salmonis</i>						<i>Caligus clemensi</i>				
		# lice	Cop	Ch A	Ch B	Pre-A	A	# lice	Cop	Ch A	Ch B	A
BB	low	68	8.3	33.3	16.7	8.3	33.3	135	12.8	53.2	17.0	17.0
Fin	low	2	0.0	0.0	33.3	33.3	33.3	38	17.4	65.2	8.7	8.7
	high	61	23.5	48.2	14.1	7.1	7.1	171	11.6	44.2	11.6	32.6
GS	low	189	10.2	37.1	18.4	14.7	19.6	236	10.2	47.6	28.2	14.0
	high	1046	8.0	38.7	20.0	17.0	16.2	840	8.0	38.9	30.8	22.3

Note: Broughton Archipelago is not included due to unidentified species of chalimus stages as a result of live-sampling.

**Table 1.4.** List of candidate models and resulting Akaike's Information Criteria (AIC) scores used to determine which factors most influence sea louse prevalence on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon.

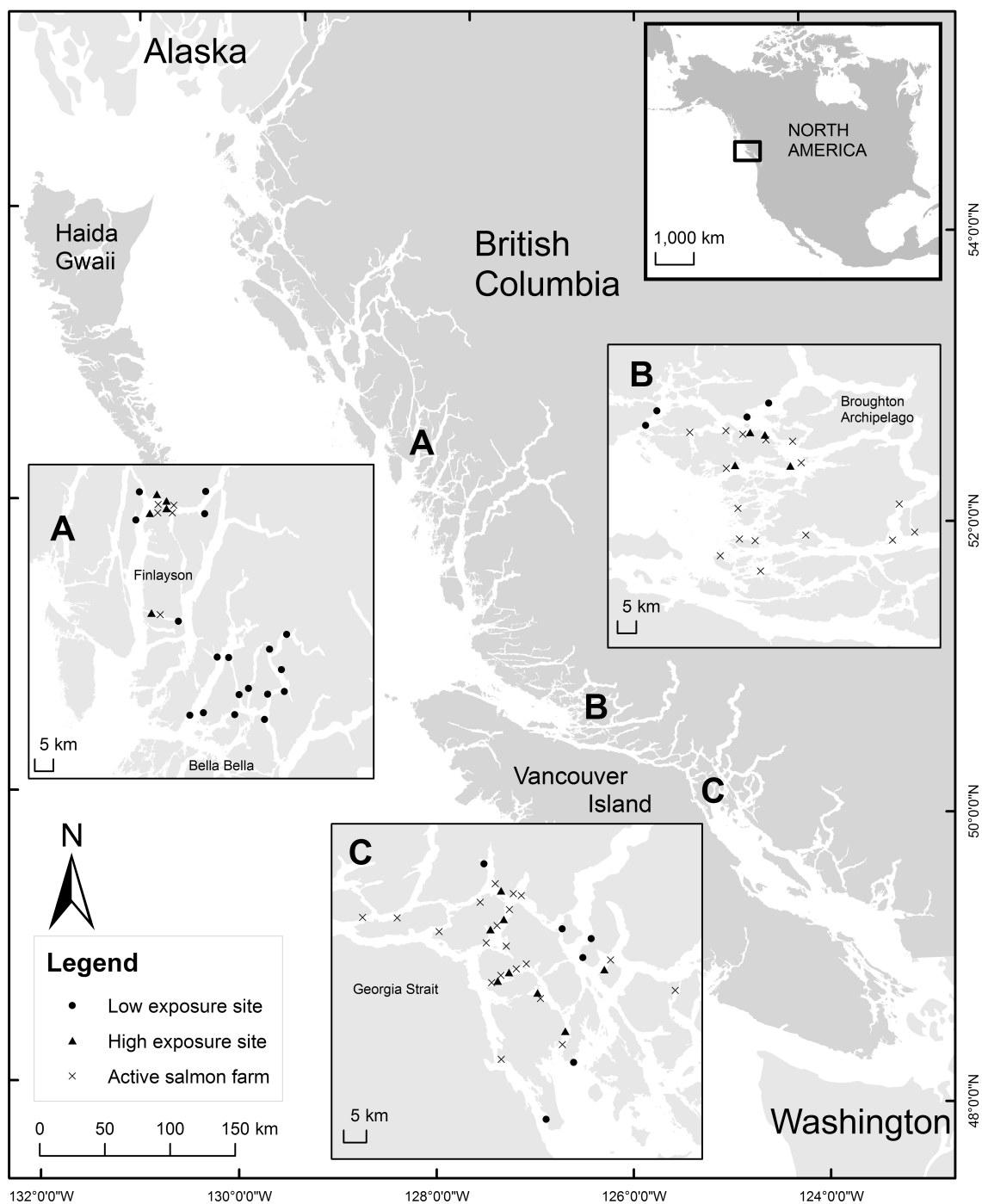
Model	K	AIC	$\Delta$ AIC	$\omega_i$
Exposure + salinity *	4	-255.069	0.000	0.394
Exposure *	3	-254.660	0.409	0.321
Exposure + temperature *	4	-254.231	0.838	0.259
Exposure + length	4	-248.094	6.975	0.012
Length	3	-221.395	33.674	0.000
Salinity	3	-213.518	41.551	0.000
Temperature	3	-212.535	42.535	0.000
Salinity + temperature	4	-205.107	49.962	0.000
Salinity + temperature + (salinity x temperature)	5	-194.389	60.680	0.000

Note: Model structure, number of parameters + intercept + covariance structure (K), AIC,  $\Delta$ AIC, and Akaike weight ( $\omega_i$ ) are included; Exposure is fish exposure to farms (low or high), Salinity is sea surface salinity, Temperature is sea surface temperature, and Length is host fork-length. The symbol '\*' denotes models of the top model set.

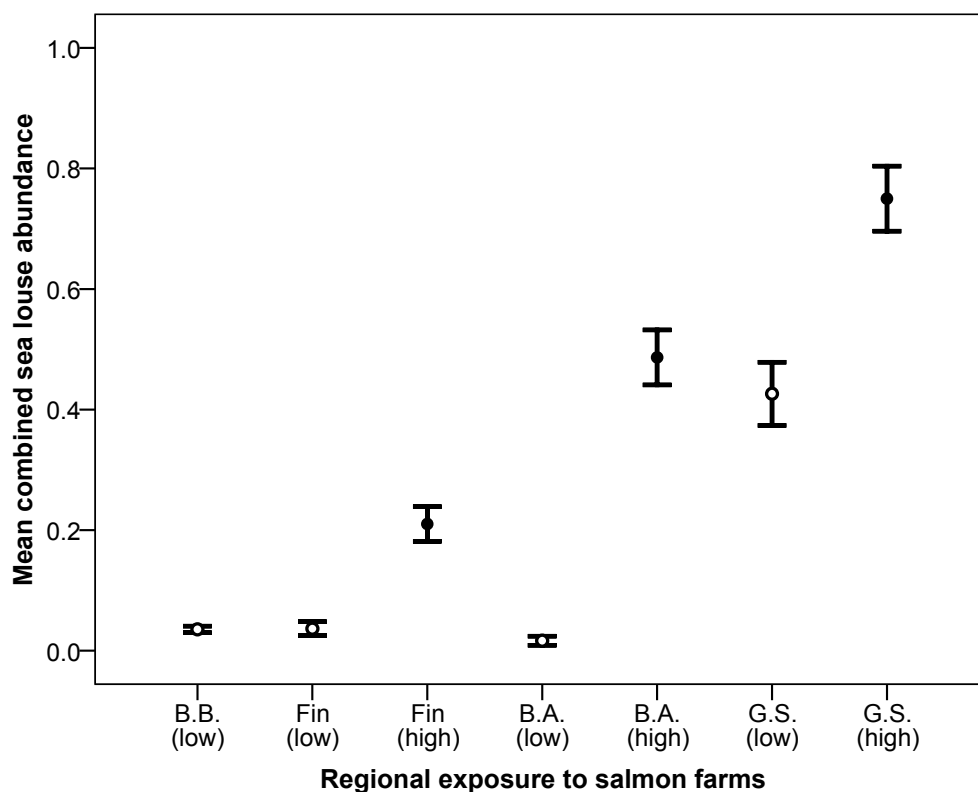
**Table 1.5.** Summed Akaike’s Information Criteria (AIC) weights ( $\sum\omega_i$ ) across the top model set to rank parameters by relative importance in predicting sea louse prevalence levels on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon.

Parameter	$\sum\omega_i$	Direction of highest louse prevalence
Exposure	0.974	High exposure to salmon farms
Salinity	0.394	Higher salinity
Temperature	0.259	Higher temperature

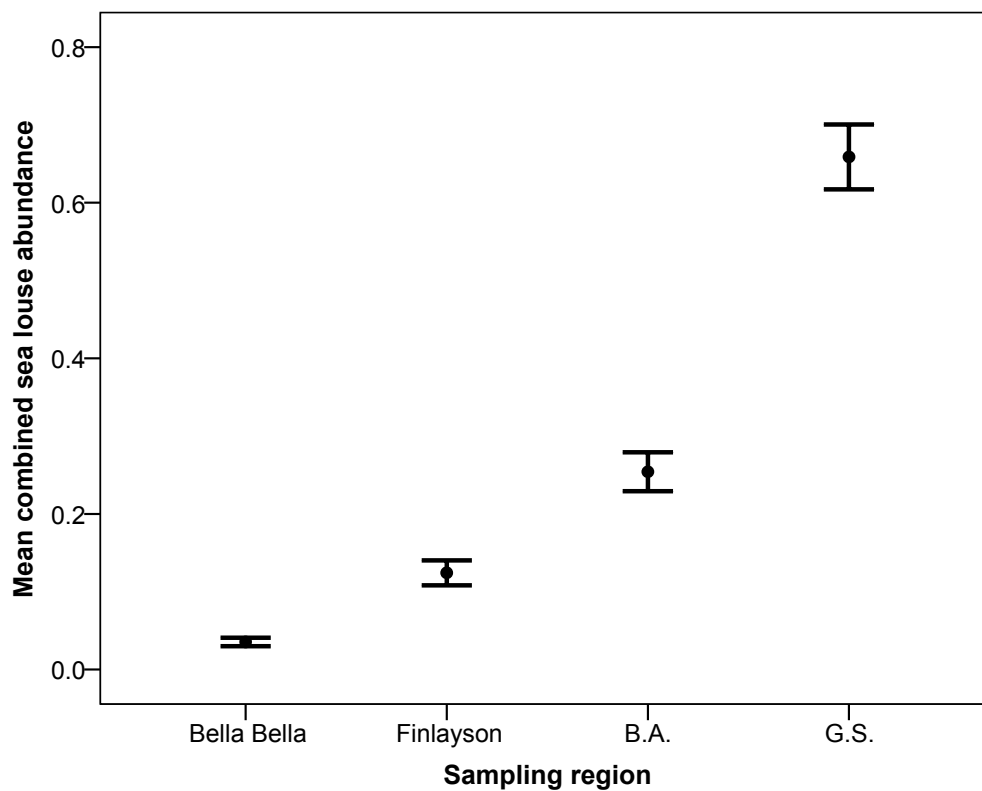
Note: Exposure is fish exposure to farm influence (low or high), Salinity is sea surface salinity, and Temperature is sea surface temperature.



**Figure 1.1.** Study area, including 3 salmon farm regions (A-C), the non-salmon farm area of Bella Bella (lower inset map A), and all associated sampling sites for juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined for sea lice in British Columbia during 2007-2008.



**Figure 1.2.** 95% Confidence Intervals  $\pm$  2 SE of combined mean sea louse (*Lepeophtheirus salmonis* and *Caligus clemensi*) abundance on juvenile chum (*Oncorhynchus keta*) and pink salmon (*O. gorbuscha*) examined at sites of low (open circle) or high (solid circle) exposure to salmon farms for all years combined: B.B. is Bella Bella, Fin is Finlayson, B.A. is Broughton Archipelago, and G.S. is Georgia Strait.



**Figure 1.3.** 95% Confidence Intervals  $\pm$  2 SE of combined mean sea louse (*Lepeophtheirus salmonis* and *Caligus clemensi*) abundance on juvenile chum (*Oncorhynchus keta*) and pink (*O. gorbuscha*) salmon examined among 4 areas of coastal British Columbia, and their associated farm salmon production in metric tonnes (MT): Bella Bella (0), Finlayson (1 911), Broughton Archipelago (B.A., 16 174), and Georgia Strait (G.S., 17 005).

## Chapter 2

Sea louse infection of juvenile Pacific sockeye salmon (*Oncorhynchus nerka*) in relation to marine salmon farms on Canada's west coast.

Published in slightly modified form in Price, M.H.H., Proboszcz, S.L., Routledge, R.D., Gottesfeld, A.S., Orr, C., and Reynolds, J.D. 2011. Sea louse infection of juvenile Pacific sockeye salmon (*Oncorhynchus nerka*) in relation to marine salmon farms on Canada's west coast. Public Library of Science ONE 6: e16851.

## Abstract

Pathogens are growing threats to wildlife. The rapid growth of marine salmon farms over the past two decades has increased host abundance for pathogenic sea lice in coastal waters, and wild juvenile salmon swimming past farms are frequently infected with lice. I used genetic analyses to determine the origin of sockeye from Canada's two most important salmon rivers, the Fraser and Skeena; Fraser sockeye migrate through a region with salmon farms, and Skeena sockeye do not. I compared lice levels between Fraser and Skeena juvenile sockeye, and within the salmon farm region I compared lice levels on wild fish either before or after migration past farms. I matched the latter data on wild juveniles with sea lice data concurrently gathered on farms. Fraser River sockeye migrating through a region with salmon farms hosted an order of magnitude more sea lice than Skeena River populations, where there are no farms. Lice abundance on juvenile sockeye in the salmon farm region were substantially higher downstream than upstream of farms for the two common species of lice: *Caligus clemensi* and *Lepeophtheirus salmonis*, and changes in their proportions between two years matched changes on the fish farms. Mixed-effects models show that position relative to salmon farms + migration year best explained both *C. clemensi* and *L. salmonis* abundance on sockeye. This is the first study to demonstrate a potential role of salmon farms in sea lice transmission to juvenile sockeye salmon during their critical early marine migration. Moreover, it demonstrates a major migration corridor past farms for sockeye that originated in the Fraser River, which contains a complex of populations that are the subject of conservation concern.

## Introduction

Pathogens are growing threats to wildlife (Macdonald and Laurenson 2006; Thirgood 2009). The spread of infectious pathogens commonly occurs when humans bring wildlife into increased contact with infected domestic animals (Dobson and Foufopoulos 2001; Otterstatter and Thomson 2008). Ensuing epizootics have devastated wild populations, as illustrated by the transmission of rabies from domestic dogs to wild carnivores (Daszak *et al.* 2000; Power and Mitchell 2004), *Pasteurella* from domestic to wild sheep (Jessup *et al.* 1991), and *Crithidia bombi* from commercial to wild bumble bees (Otterstatter and Thomson 2008).

Caligid sea lice (mainly *Lepeophtheirus salmonis* and *Caligus* spp.) are the most widespread marine parasites affecting domestic and wild fish, and have now emerged as important pathogens in many coastal marine areas (Costello 2006, 2009; Krkosek 2010). Sea lice feed on surface tissues of their hosts, which can lead to many problems especially for small juvenile fish (Costello 2006; Pike and Wadsworth 2000). Sea lice can compromise osmoregulation (Bjorn and Finstad 1997), induce behavioral changes that increase predation risk (Krkosek *et al.* 2011b), reduce growth rates and, in sufficient numbers, result in host death (Costello 2009; Morton and Routledge 2005; Krkosek *et al.* 2006). Sea lice also have been shown to serve as vectors for the spread of fish diseases (Nese and Enger 1993; Nylund *et al.* 1994).

The transmission of pathogens to wildlife frequently occurs where host populations are concentrated into dense aggregations (Daszak *et al.* 2000; McCallum and Dobson 1995). The recent global expansion of marine salmon farming is one such situation in which concentrated

reservoir populations may dramatically alter the natural transmission dynamics of salmonid host-parasite systems (Orr 2007; Costello 2009; Fraser 2009; Marty *et al.* 2010). In natural systems, migratory allopatry (the spatial separation of age classes) of wild salmon creates a barrier to parasite transmission (Krkosek *et al.* 2007b). Conversely, salmon farms hold domestic fish, mainly Atlantic salmon (*Salmo salar*), in high densities for months in the same location (i.e., 15-30 kg/m<sup>3</sup> for up to 24 months; Marine Harvest Corporate 2008). These crowded conditions facilitate parasite and disease transmission within the farm, and enable exponential population growth of pathogens and release to the surrounding environment (Murray and Peeler 2005; Murray 2008). Juvenile wild salmon swimming past salmon farms are frequently infected with sea lice (Price *et al.* 2010; Marty *et al.* 2010), and studies have implicated sea lice from farms in the decline of some wild salmonid populations in Europe and North America (Costello 2009; Heuch *et al.* 2005; Krkosek *et al.* 2007a, 2011a; Krkosek and Hilborn 2011).

Recent research has raised concern that sea lice from salmon farms may infect juvenile sockeye salmon (*Oncorhynchus nerka*) in an area of Canada's west coast between Vancouver Island and the mainland known as the Discovery Islands (Morton *et al.* 2008). This region is home to the northeast Pacific's largest salmon farm industry and hosts one of the largest migrations of salmon in the world (primarily to and from the Fraser River; Hartt and Dell 1986). Sockeye is the Pacific Ocean's most economically and culturally important salmon species, and several populations from the Fraser River are endangered (IUCN 2008). Productivity of Fraser River sockeye has been declining since the early 1990s, with 2009 being the lowest on record, prompting the Canadian government to launch a Judicial Inquiry to investigate the cause of the decline and

identify imminent threats to their survival (Cohen Commission 2010). The early marine phase of sockeye remains one of the least understood (Welch *et al.* 2009), yet has received the most attention in the search for answers to declining sockeye productivity (Peterman *et al.* 2010). Thus, determining whether sockeye are at risk from sea lice transmission from salmon farms during their early marine migration is highly relevant to conservation and management efforts.

In this study I examined parasite infection of wild juvenile sockeye from two geographically separated regions of Pacific Canada: one with salmon farms, and one without. Within the farm region, I compared infection rates on fish from locations that vary in their exposure to farms. I used molecular genetic techniques to determine the origins of the fish, and I employed mixed-effects modelling to examine factors that best explain sea lice abundance.

## **Materials and methods**

I collected juvenile sockeye from marine waters surrounding the Discovery Islands, an area containing 18 active salmon farms, from April 22 to June 15, 2007 ( $n = 381$ ) and May 31 to July 3, 2008 ( $n = 510$ ), and acquired samples retained from the north coast of British Columbia, an area without salmon farms, from May 26 to July 5, 2007 ( $n = 369$ ; Figure 2.1). Up to five replicate sets of samples were obtained from each site, each year, in the Discovery Islands (1-50 juvenile sockeye salmon per sample), and during 2007 on the north coast (1-129 juvenile sockeye salmon per sample). I used a beach seine (50 m long, 1.5 m deep, 6 mm mesh) among the Discovery Islands to capture sockeye, and a surface trawl-net (18 m long, 5 m opening, 4.6 m deep) was used on the north coast. The trawl-net was fitted with a rigid holding box at the far end

designed for live capture and to minimize the loss of scales and ectoparasites (Holst and McDonald 2000). Sea surface salinity and temperature were recorded during each sampling event in both regions using a YSI-30 SCT meter. Fish were immediately frozen and labeled for subsequent laboratory analyses in which individual fish were thawed and assayed for sea lice using a dissecting microscope. Species of motile (i.e., sub-adult and adult) stages of sea lice were directly identified by morphology (Kabata 1972; Johnson and Albright 1991); younger copepodid and chalimus stage lice were removed from the fish, mounted on permanent slides and examined under a compound microscope for determination based on detailed morphology (Kabata 1972; Johnson and Albright 1991).

I proportionately sampled previously frozen tissues for genetic determination in the Discovery Islands from juveniles retained at each capture location, per sampling event, each year (i.e., 1/3 from 2007,  $n = 92$ ; 1/5 from 2008,  $n = 114$ ), and placed them individually in vials of 95% ethanol. Fresh tissue from all sockeye ( $n = 478$ ) on the north coast were placed individually in vials of 99% ethanol. Tissue samples from both regions were analyzed at the Fisheries and Oceans Canada (DFO) molecular genetics laboratory in British Columbia. DNA was extracted from tissue (Withler *et al.* 2000), and samples were analyzed for polymerase chain reaction products at 14 microsatellite loci (Beacham *et al.* 2004). I considered amplification at a minimum of 7 loci as adequate for estimating stock origin as previous surveys of the microsatellite variation in Fraser River sockeye at 6 loci indicated differentiation among populations (Withler *et al.* 2000). Individuals were assigned to source populations using mixed stock analysis techniques employing Bayesian mixture modeling (Pella and Masuda 2001) using

the software program cBayes. Stock proportions were determined by comparing one mixture (north coast 2007) to a baseline comprising 227 sockeye populations, and two mixtures (Discovery Islands 2007 and 2008) to a baseline comprising 85 sockeye populations (Beacham *et al.* 2004, 2005). The reported stock composition estimates with corresponding standard deviations were derived from combined posterior distributions using the last 1 000 iterations from 10 Monte Carlo Markov runs of 20 000 iterations.

To test for spatial patterns in sea lice on sockeye, I organized capture locations within the Discovery Islands based on whether each site was: upstream (a position on the juvenile sockeye migration route where fish likely had not passed a salmon farm), or downstream (a position where fish must have passed at least one salmon farm), given the net movement of juvenile sockeye through the region (Groot and Cooke 1987); downstream collection sites are encircled within Figure 2.1. The ocean environment surrounding the Discovery Islands is estuarine, with a net-northward flow predominating during the months of my study (Thomson 1981). Fish captured downstream of a salmon farm could only have arrived at that location by swimming past a salmon farm, and my results on genetic origins of the fish substantiated this. However, sockeye caught at two sites considered upstream of a salmon farm may have swum past a farm before capture because of fish movements or strong tidal currents, and the close proximity to a farm. Although I consider these occurrences infrequent, they may have contributed to the variability in louse infection levels observed at these sites. One additional site upstream of farms and near a farm salmon processing facility emerged as a clear outlier. Because such outliers can exercise undue influence on inferences based on regression-style statistical models (Kleinbaum

*et al.* 2008), yet can also provide important insight, I singled out this site for special consideration. Finally, I placed collection sites from the north coast in a third category: no farms.

Marine Harvest Canada (MHC) is the only salmon farm company to report sea louse average abundance; raw sea louse data were not reported publicly at the time of my study. I used average *Caligus clemensi* abundance and *L. salmonis* motile abundance provided online to estimate sea louse trends on six MHC farms in the Discovery Islands during 2007-2008; sea louse data were not provided for the other 12 farms operating in the region. For periods without reported information, I calculated average abundance using the previous and subsequent values.

I was interested in which factors most influence sea louse infection levels on juvenile sockeye in BC. Accordingly, based on the literature cited in the Introduction, I formulated a priori hypotheses relating fish capture sites to the abundance of sea lice on juveniles captured at those sites. Specifically, I hypothesized that fish from locations that were downstream to salmon farms would have higher louse abundance than those upstream to farms and where there are no farms, and that high temperature and salinity would also be correlated with high lice loads (because sea louse growth in lab-based trials depends strongly on temperature and salinity; Pike and Wadsworth 2000; Costello 2006).

I used generalized linear mixed effects modelling to account for the hierarchical nature of the sampling, where multiple sampling events at a given location were treated as random factors nested within location. I included position relative to salmon farms (downstream, upstream, or no

farms), temperature, salinity, and year as fixed factors to examine their influence on sea louse infection levels on juvenile sockeye. I ran the complete suite of models ( $n = 15$ ) of all subsets of the four factors on total abundance of each louse species (*C. clemensi* and *L. salmonis*), and I ran analyses with and without the outlier site excluded. Because results were broadly similar, and due to the statistical problems of including the outlier site (mentioned above and in the Discussion section), I report findings with the outlier excluded. I averaged louse abundance within a sampling event (i.e., replicate;  $n = 96$ ) to avoid pseudo-replication, and transformed abundance data ( $y$ ) to  $\log_e(y + 0.5)$  to correct for unequal variances and non-normality. I tested a set of candidate models using Akaike's Information Criterion ( $AIC_c$ ) for small sample sizes, and then evaluated  $\Delta AIC_c$  to select the best approximating model. I made appropriate inference using  $\Delta AIC_c < 4$  to describe the top model set. Finally, I summed Akaike weights ( $\omega_i$ ) across the top model set for each variable to rank them by importance (Burnham and Anderson 1998; Anderson *et al.* 2001).

Because the spatial distribution of upstream/downstream collection sites assumes a northbound migration, juveniles caught downstream of farms may have been influenced by extended residency time (i.e., increased exposure to sea lice, which may lead to epizootics; Krkosek *et al.* 2009). I tested for this potential effect independently of the effect of position relative to farms by using linear regression analysis: latitude coordinates were assigned for each collection site, and average lice levels on fish at a given site were compared with other sites within upstream and downstream categories separately. I generated all analyses using SPSS 16.0 for Mac (SPSS 2007).

## Results

Genetic analyses confirmed that the majority of juvenile sockeye on the north coast were from the Skeena, Nass, and adjacent watersheds (98.3% combined), and thus they were unlikely to have been influenced by salmon farms further south before capture (Table 2.1; Figure 2.1).

Conversely, all sockeye migrating through the Discovery Islands region were either from the Fraser River (85%) or nearby Johnstone and Queen Charlotte Strait rearing lakes (15%), and may have been influenced by salmon farms depending on their location.

Sea louse abundances on the north coast for *C. clemensi* and *L. salmonis* combined were an order of magnitude lower than in the Discovery Islands (Table 2.2). Within the Discovery Islands, *C. clemensi* was the principal louse species infecting sockeye in both years, and most abundant on fish downstream of salmon farms (Figure 2.2). The maximum infection intensity of *C. clemensi* was highest downstream of farms in 2007 (28 lice per fish) compared to upstream sites (16 lice per fish), and equal throughout the region in 2008 (9 lice per fish).

Sockeye at downstream locations were larger on average than upstream locations, particularly in 2008 (Table 2.2). The difference in body size before and after farms in 2008 was driven largely by fish collected at two locations on two separate days; by excluding these fish, the difference in weight between downstream and upstream fish is reduced from 3.89 g to 1.73 g, and yet lice levels on fish downstream of farms remain significantly higher than fish upstream of farms ( $t = 8.349$ ,  $df = 745$ ,  $p = 0.000$ ). The relationship between migration distance and louse infection

levels on juvenile sockeye was not statistically significant for either upstream ( $r^2 = 0.191$ ,  $df = 9$ ,  $p = 0.207$ ) or downstream ( $r^2 = 0.083$ ,  $df = 30$ ,  $p = 0.115$ ) categories.

Excluding sockeye caught at the outlier site among the Discovery Islands in 2008, which hosted the highest levels of either louse species during that year, *L. salmonis* was most abundant on juveniles downstream of salmon farms, and more abundant in 2008 compared to 2007 (Figure 2.3). In correspondence with the hypothesized contributions of salmon farms to these wild fish, MHC farms hosted more *C. clemensi* during the out-migration period in 2007 than 2008, and more *L. salmonis* in 2008 than 2007 (Figure 2.4).

Mixed-effects modelling showed some variation in results depending on louse species. Model selection suggested that position to salmon farms + migration year was the best predictor of *C. clemensi* abundance on juvenile sockeye given the set of candidate models (Table 2.3).

Specifically, *C. clemensi* abundance increased on sockeye downstream of salmon farms, and was most prominent in the migration year of 2007; 5 of 5 models in the top model set (0-4  $\Delta AIC_c$ ) contained position to salmon farms. Summing the Akaike weights across top models for *C. clemensi* ranked the variable position ( $\sum \omega_i = 0.927$ ) higher than year, temperature, and salinity by factors of 1.2, 3.2, and 11.2, respectively (Table 2.4). Position to farms + migration year was the only model in the top model set to explain *L. salmonis* abundance (Table 2.5).

## Discussion

I have demonstrated a potential role of open net-pen salmon farms in transmission of sea lice to wild juvenile sockeye salmon. Most juvenile sockeye assessed for sea lice originated either in the Fraser or Skeena watershed, thus providing a novel comparison of sea louse infection between Canada's largest sockeye rivers. Moreover, genetics results demonstrate a major migration corridor past farms for fish that originated in the Fraser River, a complex of populations that have been the subject of concern due to declining productivity since the early 1990s, and a collapse in 2009 followed by a substantial rebound in 2010.

Juvenile sockeye salmon in both regions were primarily infected by *C. clemensi*, which is consistent with juvenile pink and chum salmon in areas without salmon farms in the north Pacific (Morton *et al.* 2004; Krkosek *et al.* 2007b). The predominance of *C. clemensi* routinely shifts to *L. salmonis* for pink and chum in regions with intensive salmon farming (Krkosek *et al.* 2005a; Morton *et al.* 2008; Marty *et al.* 2010), and this was shown for those species in the Discovery Islands during the years of my study (Price *et al.* 2010). Most of the sockeye I examined among the Discovery Islands were caught in mixed schools with *L. salmonis*-infected juvenile pink and chum. Thus, the predominance of *C. clemensi* on sockeye suggests that sockeye either show higher resistance to *L. salmonis*, or heightened susceptibility to *C. clemensi*; alternatively, perhaps *C. clemensi* has a preference for sockeye, or *L. salmonis* prefers juvenile pink and chum salmon. This warrants future experimental work.

Juvenile sockeye migrating along the north coast hosted an order of magnitude fewer sea lice

than those migrating through the Discovery Islands. Wild juvenile salmon in Europe and North America consistently host low levels of sea lice during their early marine migration in areas without salmon farms (Morton *et al.* 2004; Krkosek *et al.* 2007b; Gottesfeld *et al.* 2009), though brief localized outbreaks have occurred (Parker and Margolis 1964; Beamish *et al.* 2009). Louse parasitism of juveniles is frequently higher for sustained periods in regions with salmon farming (Tully *et al.* 1999; Heuch *et al.* 2005; Krkosek *et al.* 2005a). Factors beyond the absence of farm salmon on the north coast may have contributed to the significantly lower lice levels on sockeye compared to the Discovery Islands. In particular, differences in lice levels may be due to the use of different sampling gear or different environmental conditions, though I did incorporate the two key conditions known to affect sea louse infection levels into my analyses: salinity and temperature. My analyses show that the lower infection rates for *C. clemensi* on the north coast cannot be explained by salinity and temperature alone. The primary strength of my study was the comparison of infection levels before and after fish had been exposed to salmon farms within the Discovery Islands.

Parasitism of sockeye by *C. clemensi* in the Discovery Islands was higher on juveniles downstream of salmon farms than on those upstream of farms. These findings are consistent with previous research on juvenile pink and chum salmon in this region, and elsewhere in the north Pacific (Morton *et al.* 2008; Price *et al.* 2010). Farm data provide further evidence that *C. clemensi* was abundant on farm salmon while juvenile sockeye migrated through the region, particularly during the higher infection year of 2007 (BCMAL 2007, 2008; see my Figure 2.4). Although the position of sockeye relative to salmon farms was the primary factor to explain my

data, I need to consider alternative explanations. First, the spatial distribution of upstream/downstream collection sites assumes a northbound migration. Juveniles caught downstream of farms were consistently larger than upstream sockeye, which may be evidence for extended residency time (i.e., increased exposure to sea lice, which may lead to epizootics; Krkosek *et al.* 2009). However, the relationship between migration distance and louse infection levels on juveniles was not statistically significant within either upstream or downstream categories. Importantly, excluding fish caught at two locations downstream of farms reduced the difference in weights, yet lice levels remained significantly higher downstream of farms. Moreover, juveniles from different populations within the Fraser River are not of equal size, and they vary in their migration timing through the study region (M. Price unpublished data); thus, size may not be a simple metric for residency time and deserves further examination. Second, because *C. clemensi* is a generalist parasite, non-salmonids such as Pacific herring (*Clupea pallasii*) may have been a local source for lice (as has been hypothesized elsewhere; Beamish *et al.* 2009). I also consider this unlikely to account for *C. clemensi* increases on sockeye downstream of farms, as pelagic fishes would need to assume a similar spatial distribution (i.e., more fishes downstream of farms) over consecutive years, and there is no evidence for this.

Similar to *C. clemensi*, parasitism of sockeye by *L. salmonis* was higher in the Discovery Islands than the north coast, and lice levels further increased for juveniles downstream of salmon farms. Notably, the year of highest infection among the Discovery Islands was the opposite for each louse species infecting sockeye: *L. salmonis* was most abundant in 2008, *C. clemensi* was most abundant in 2007, and farm salmon in this region showed similar inter-annual trends for each

species. Mixed-effects modelling further showed that migration year + position to farms best explained *L. salmonis* total abundance, indicating significant inter-annual variation in *L. salmonis* abundance on sockeye that is consistent with farm salmon. Farm salmon hosted lice well before sockeye began migrating through the region, and are the most likely source of infection.

Sockeye among the Discovery Islands were most infected with *L. salmonis* at the outlier site compared to all other sites. This site was approximately 8 km upstream from a farm salmon processing facility where large numbers of live sea lice, primarily nauplii, were recently recorded from the effluent (see Chapter 3). Tidal currents here (i.e., Discovery Passage) can transport particles this distance in a single tide-cycle (Thomson 1981), which suggests that the processing facility may have been a source for lice on sockeye. This also suggests that other ‘upstream’ locations may have been exposed to farm-origin lice (and may explain the significantly higher lice levels on sockeye at all upstream sites compared to the north coast), but to a lesser degree than downstream locations. Alternatively, this single location may have been home to a large congregation of resident fishes that were heavily infected with sea lice. Although I caught only sockeye during this single capture event, I have caught juvenile pink and chum salmon with relatively low lice levels at that location previously. Note that while I cannot justify including this outlier site in my formal statistical tests because it is inconsistent with the model assumptions, when I included the outlier in the analysis (the invalidity of the inferences notwithstanding), the primary conclusions remained essentially the same. Hence, this unique observation, though it does not critically impinge on the results of the study, is important in that

it suggests the need for heightened attention towards the potential role of processing plants in sea lice dynamics.

Does *C. clemensi* pose a threat to sockeye salmon? Research to date has not examined the effects of this sea louse on wild juvenile Pacific salmonids, though significant fin damage by larval stage lice has been documented (Parker and Margolis 1964). *Caligus clemensi* is smaller than *L. salmonis*, and is thought to cause less mechanical damage to juvenile pink and chum salmon (Morton and Routledge 2005; Krkosek *et al.* 2007b; Costello 2009). Moreover, juvenile sockeye are larger and have developed scales at the time of ocean entry compared to juvenile pink and chum; thus, it is unlikely that the average number of *C. clemensi* observed on sockeye (2-3 lice/fish) would cause direct mortality for healthy fish. However, evidence is mounting that marine parasites, such as sea lice, can induce behavioral changes that may result in higher mortality rates for hosts (Webster *et al.* 2007; Krkosek *et al.* 2011b). The transition from freshwater to marine environments is one of the most physiologically demanding phases for salmon (Quinn 2005), and overall marine survival appears to depend on rapid early marine growth (Beamish *et al.* 2004). Even low levels of parasitic infection may be harmful during this critical period. Moreover, the presence and abundance of sea lice on juvenile sockeye may be a proxy for other farm-origin pathogens. Given the high intensities of *C. clemensi* observed on some juveniles in this study (i.e., up to 28 lice/fish), concern is justified, and research should be undertaken to understand the extent of threat posed.

There is considerable interest in understanding the factors that affect survival of juvenile sockeye in the marine environment, and specifically whether salmon farms are contributing to declines. Sockeye productivity in many Canadian river systems has declined over the last decade, including the Skeena River; thus multiple contributing factors other than farm-origin parasites are likely responsible for reduced sockeye productivity. However, unlike most other systems, Fraser River sockeye experienced a record-low return in 2009, triggering a federal Judicial Inquiry (Cohen Commission 2010). Although the effect of sea louse parasitism on juvenile sockeye acting in isolation may arguably be small, it could be important when combined with multiple stressors (Finstad *et al.* 2007). Negative impacts of salmon farms on wild populations have been indicated in other parts of the world (Ford and Myers 2008; Costello 2009), and in juvenile pink, and coho salmon populations on the west coast of Canada (Krkosek *et al.* 2007a, 2011a; Connors *et al.* 2010). A recent study found no correlation between numbers of lice on farms and adult pink salmon returns in the Broughton Archipelago, which is located between my southern and northern sites (Marty *et al.* 2010). This study, based on a nine-year time series, lacked full statistical comparisons of productivity in regions without salmon farms. Other more recent studies that included such comparisons reported significant declines in productivity of pink salmon in relation to salmon farms (Krkosek and Hilborn 2011; Krkosek *et al.* 2011a).

My evidence suggests that salmon farms are elevating parasite levels on Fraser River sockeye during their critical early marine migration; to establish the link more definitively between farms and wild fish would require collaborative work with the salmon farm industry as has begun in Europe and the Broughton Archipelago (Penston *et al.* 2008; Marty *et al.* 2010). Ultimately, risks

to wild salmon posed by salmon farms can be more easily mitigated than the far-reaching effects on ocean productivity of climate change and ocean acidification. Options already recommended include removal of farm salmon from migration routes of juvenile sockeye from the Fraser (Statement from Think Tank of Scientists 2009), and transitioning of salmon farms to closed-containment facilities (LABC 2007). At minimum, the Discovery Islands' migration corridor requires a co-ordinated aquaculture management plan to minimize the exposure of wild juvenile sockeye to sea lice.

**Table 2.1.** Stock proportion estimates and standard deviations for genetically identified juvenile sockeye salmon (*Oncorhynchus nerka*) caught on British Columbia's north coast (NC) and Discovery Islands (DI).

Stock Origin	NC 2007	DI 2007	DI 2008
	Estimate (SD)	Estimate (SD)	Estimate (SD)
Chilko Lake (Fraser River)	0.0 (0.0)	22.8 (4.7)	26.9 (3.9)
Quesnel Lake (Fraser River)	0.0 (0.1)	33.4 (5.2)	3.1 (1.9)
Shuswap Lake (Fraser River)	0.0 (0.1)	0.0 (0.2)	57.9 (4.1)
Other Fraser River	0.0 (0.2)	5.4 (2.8)	11.0 (2.7)
Washington and Oregon	0.0 (0.0)	0.0 (0.2)	0.0 (0.1)
West Coast Vancouver Island	0.0 (0.1)	0.0 (0.2)	0.1 (0.4)
Johnstone and Queen Charlotte Straits	0.0 (0.1)	37.8 (4.9)	0.6 (0.6)
Queen Charlotte Strait to Skeena estuary	2.2 (0.9)	0.0 (0.5)	0.0 (0.4)
Skeena River estuary	3.1 (0.9)	0.0 (0.2)	0.0 (0.2)
Babine Lake (Skeena River)	85.0 (1.9)	0.0 (0.2)	0.0 (0.1)
Other Skeena River	7.7 (1.4)	0.0 (0.2)	0.0 (0.1)
Nass River	0.9 (1.2)	0.0 (0.2)	0.0 (0.2)
Queen Charlotte Islands	0.2 (0.4)	0.0 (0.5)	0.0 (0.3)
Southeast Alaska	0.7 (0.6)	0.6 (0.9)	0.3 (0.6)

**Table 2.2.** Summary statistics and sea louse infection rates on juvenile sockeye salmon (*Oncorhynchus nerka*) caught either upstream (Up) or downstream (Down) of salmon farms at north coast (NC) or Discovery Islands (DI) region. All morphometric and abiotic values represent the mean, except sea lice infection rates.

Region	Position to farms	Year	Total fish	Fork length	Mass	Salinity	Temperature	<i>C. clemensi</i>				<i>L. salmonis</i>			
								P	A	I	Nm	P	A	I	Nm
NC	Up	2007	369	8.17 cm	5.21 g	16.97 ‰	9.80 °C	0.09	0.17	1.97	0.97	0.01	0.01	1.00	1.00
DI	Up	2007	163	7.26 cm	3.91 g	25.42 ‰	10.79 °C	0.29	1.10	3.83	0.92	0.05	0.05	1.00	0.78
	Down	2007	218	7.76 cm	5.08 g	27.38 ‰	10.94 °C	0.84	4.83	5.72	0.95	0.09	0.09	1.05	1.00
	Up	2008	60	8.98 cm	8.15 g	25.98 ‰	14.72 °C	0.40	0.95	2.31	0.72	0.05	0.05	1.00	0.33
	Down	2008	400	10.30 cm	12.04 g	28.47 ‰	9.64 °C	0.62	1.61	2.60	0.55	0.21	0.30	1.42	0.31
	Outlier	2008	50	9.22 cm	8.50 g	30.00 ‰	9.00 °C	0.92	3.60	4.42	0.70	0.42	0.64	1.52	0.94

Note: P is louse prevalence, A is louse abundance, I is louse intensity, and Nm is the proportion of combined non-motile life stages (copepodid and chalimus I to IV).

**Table 2.3.** List of candidate models and resulting Akaike's Information Criteria ( $AIC_c$ ) scores used to determine which factors most influence *Caligus clemensi* abundance on juvenile sockeye salmon (*Oncorhynchus nerka*).

Model	K	$AIC_c$	$\Delta AIC_c$	$\omega_i$
Position + Year *	2	434.844	0.000	0.444
Position + Year + Temperature *	3	436.230	1.386	0.222
Position *	1	437.627	2.783	0.111
Position + Year + Salinity *	3	438.195	3.351	0.083
Position + Temperature *	2	438.629	3.785	0.067
Position + Year + Temperature + Salinity	4	439.642	4.798	0.040
Position + Salinity	2	440.973	6.129	0.021
Position + Temperature + Salinity	3	442.065	7.221	0.012
Year + Salinity	2	451.602	16.758	0.000
Year + Temperature + Salinity	3	452.187	17.343	0.000
Salinity	1	452.717	17.873	0.000
Temperature + Salinity	2	453.326	18.482	0.000
Year + Temperature	2	458.641	23.797	0.000
Year	1	460.199	25.355	0.000
Temperature	1	462.927	28.083	0.000

Note: Model structure, number of parameters + intercept + covariance structure (K),  $AIC_c$ ,  $\Delta AIC_c$ , and Akaike weight ( $\omega_i$ ) are included; Position is position of juvenile sockeye relative to salmon farms (no farms, upstream, or downstream), Year is migration year, Salinity is sea surface salinity, and Temperature is sea surface temperature. The symbol '\*' denotes models of the top model set.

**Table 2.4.** Summed Akaike's Information Criteria ( $AIC_c$ ) weights ( $\sum\omega_i$ ) across the top model set to rank parameters by relative importance in predicting *Caligus clemensi* abundance on juvenile sockeye salmon (*Oncorhynchus nerka*).

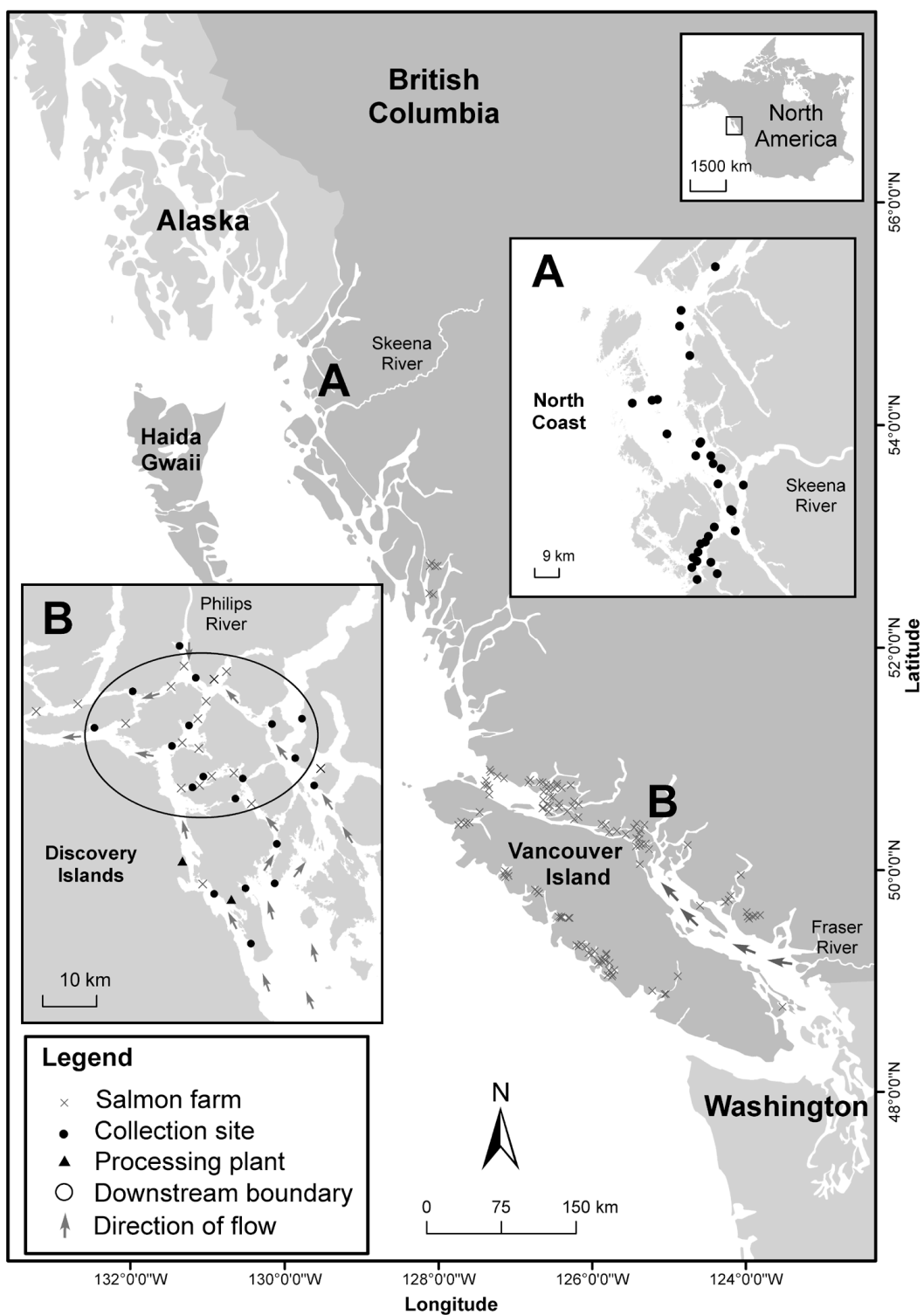
Parameter	$\sum\omega_i$	Direction of highest louse abundance
Position to farms	0.927	Downstream of salmon farms
Year	0.749	Migration year of 2007
Temperature	0.289	Higher temperature
Salinity	0.083	Higher salinity

Note: Position to farms is position of juvenile sockeye relative to salmon farms (no farms, upstream, or downstream), Year is migration year, Salinity is sea surface salinity, and Temperature is sea surface temperature.

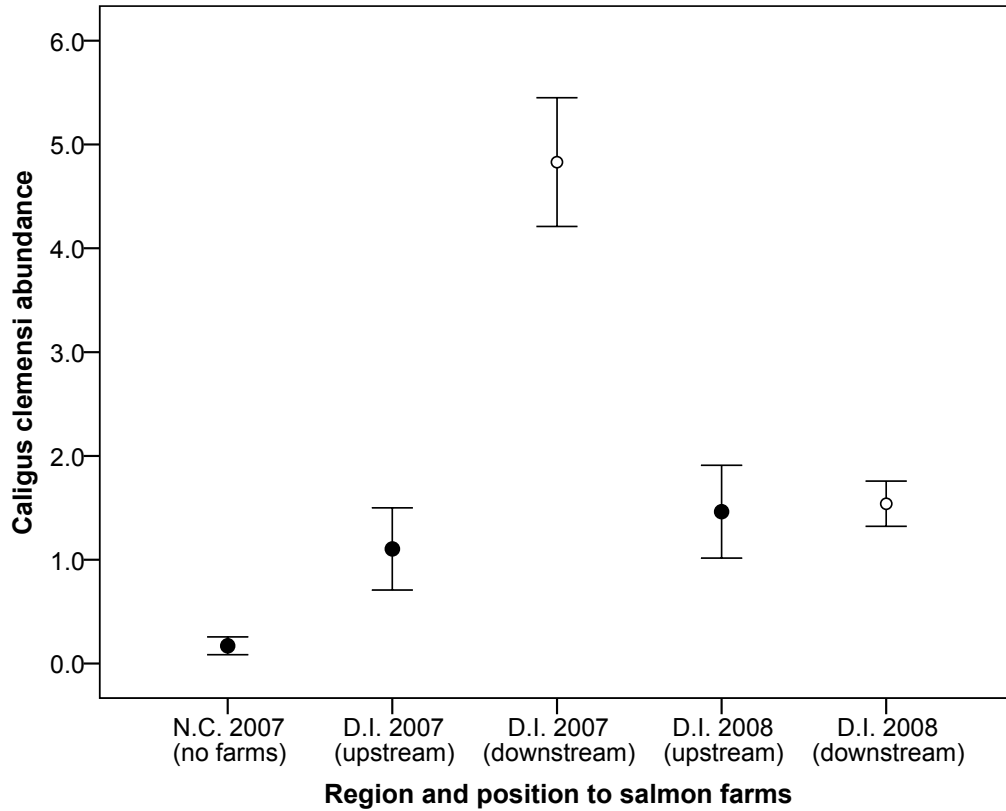
**Table 2.5.** List of candidate models and resulting Akaike's Information Criteria ( $AIC_c$ ) scores used to determine which factors most influence *Lepeophtheirus salmonis* abundance on juvenile sockeye salmon (*Oncorhynchus nerka*).

Model	K	$AIC_c$	$\Delta AIC_c$	$\omega_i$
Position + Year *	2	-93.924	0.000	0.834
Year	1	-89.045	4.879	0.073
Position + Year + Temperature	3	-88.335	5.589	0.051
Year + Temperature	2	-85.558	8.366	0.013
Position + Year + Salinity	3	-85.015	8.909	0.010
Position	1	-84.512	9.412	0.008
Year + Salinity	2	-83.957	9.967	0.006
Year + Temperature + Salinity	3	-83.521	10.403	0.005
Position + Temperature	2	-80.952	12.972	0.002
Position + Year + Temperature + Salinity	4	-79.777	14.147	0.000
Position + Salinity	2	-75.681	18.243	0.000
Position + Salinity + Temperature	3	-72.573	21.351	0.000
Temperature + Salinity	2	-71.749	22.175	0.000
Salinity	1	-71.668	22.256	0.000
Temperature	1	-59.525	34.399	0.000

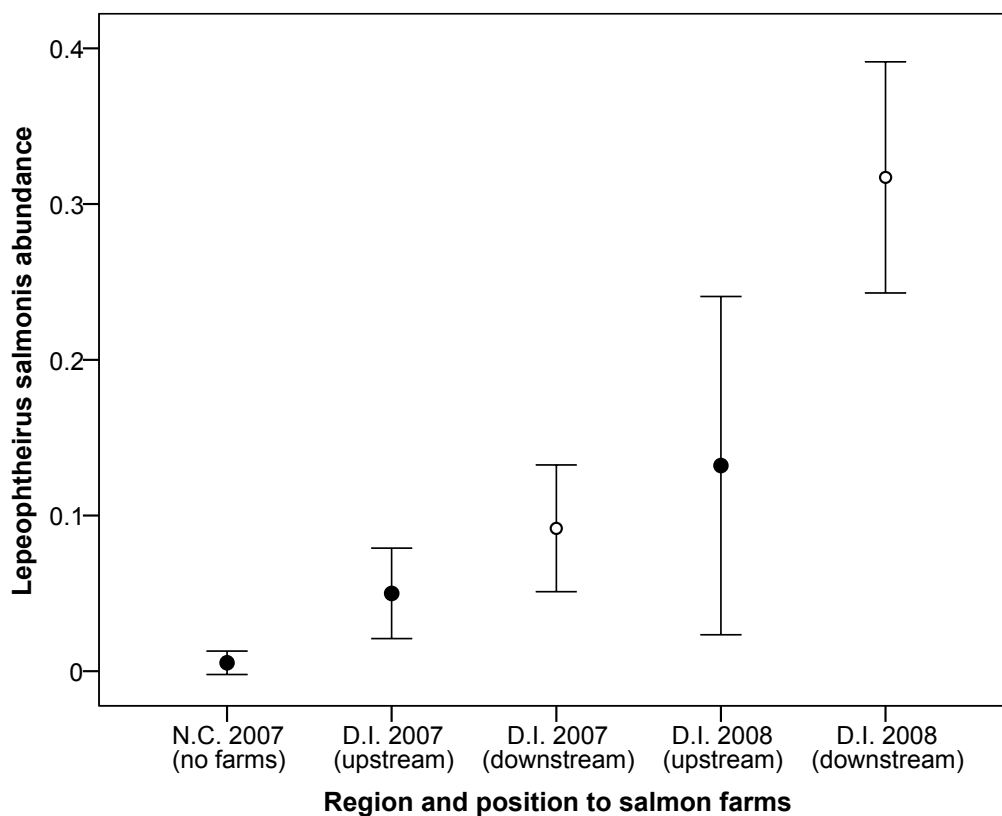
Note: Model structure, number of parameters + intercept + covariance structure (K),  $AIC_c$ ,  $\Delta AIC_c$ , and Akaike weight ( $\omega_i$ ) are included; Position is position of juvenile sockeye relative to salmon farms (no farms, upstream, or downstream), Year is migration year, Salinity is sea surface salinity, and Temperature is sea surface temperature. The symbol '\*' denotes models of the top model set.



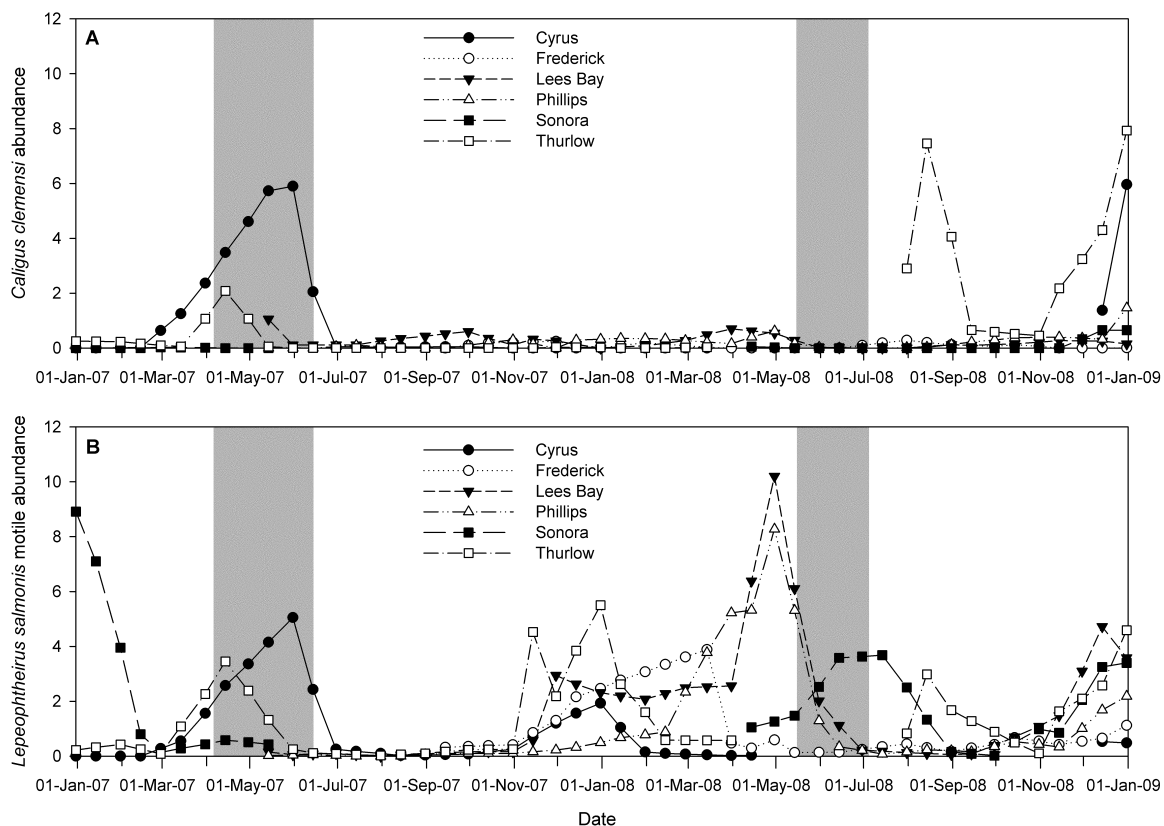
**Figure 2.1.** Sockeye salmon collection sites relative to salmon farms. Downstream boundary encircles all sockeye collection sites situated downstream of at least one salmon farm given the direction of prevailing oceanic flow and fish movement; all other collection sites are considered upstream.



**Figure 2.2.** 95% Confidence Intervals  $\pm$  2 SE of the means for *Caligus clemensi* abundance on sockeye salmon in regions with varying exposure to salmon farms. N.C. is north coast where there are no farms, D.I. upstream is Discovery Islands locations upstream of all salmon farms (open circle), and D.I. downstream is Discovery Islands locations downstream of all salmon farms (closed circle) on British Columbia's coast.



**Figure 2.3.** 95% Confidence Intervals  $\pm$  2 SE of the means for *Lepeophtheirus salmonis* abundance on sockeye salmon in regions with varying exposure to salmon farms. N.C. is north coast where there are no farms, D.I. upstream is Discovery Islands locations upstream of all salmon farms (open circle), and D.I. downstream is Discovery Islands locations downstream of all salmon farms (closed circle) on British Columbia's coast.



**Figure 2.4.** Sea louse abundance (*Caligus clemensi* at top, and *Lepeophtheirus salmonis* at bottom) over time on Atlantic salmon at named salmon farms among the Discovery Islands during concurrent sockeye collection in 2007 and 2008 (shaded grey).

## **Chapter 3**

Fish processing facilities: new challenge to marine biosecurity in Canada

## Abstract

The transmission of pathogens is a common consequence of animal food production. Marine salmon farms and their processing facilities can serve as sources for virulent fish pathogens; my study is the first to confirm the broadcast of a live fish pathogen from a farm salmon processing facility into the marine waters of Canada's Pacific coast. I found live sea lice, *Lepeophtheirus salmonis*, mucus, and fish tissue in effluent from the processing facility. Sea lice transmitted from this source may pose a threat to wild salmon populations, and the release of potentially untreated offal, including blood water, is of considerable concern. Further research is needed to quantify the extent to which processing facilities collect and subsequently release sea lice, and whether more virulent fish pathogens are present in effluent. I recommend salmon farming nations, including Canada, develop mandatory bio-security programs to ensure farm salmon processing facilities prevent broadcast of live infectious fish pathogens into wild fish habitat.

## Introduction

Humans often play the role of catalyst in the spread of pathogens in the environment, such as when infected domestic animals are brought into contact with wildlife (Daszak *et al.* 2000). The transmission of *Pasteurella* from domestic to wild sheep (Jessup *et al.* 1991), and *Crithidia bombi* from commercial to wild bumblebees (Otterstatter and Thompson 2008), are two such examples. Consequently, disease management has become an intrinsic part of animal food production.

Fish farming is the fastest growing agriculture sector globally (FAO 2008). Pathogen-mediated

effects on marine life as a result of mariculture's global proliferation is not well understood, though increased rates of transmission and emergence of pathogens are documented (Nowak 2007). Intensive growing conditions in open net-pens facilitate the introduction, amplification, and subsequent broadcast of pathogens (Murray and Peeler 2005); thus, marine salmon farms serve as point sources for exceptional pathogen contributions. Although the number of fish diseases that plague salmon farms is extensive, evidence of transmission to wild populations is limited. This may be because diseased organisms are difficult to detect if not tracked (Gozlan *et al.* 2006).

Disease transmission between fish occurs through contact with tissue, blood, or mucus from infected fish (Totland *et al.* 1996), and pathogens can disseminate over long distances via farm-to-farm infections (Saksida 2006) and infected parasites (Nyland *et al.* 1994). Farm-origin fish diseases may be further broadcast through the release of untreated effluent from slaughterhouses processing infected farm salmon (Vagsholm *et al.* 1994; Jarp and Karlsen 1997), which may be far removed from source farms. I was interested in examining the potential for a farm salmon processing facility on Canada's west coast to act in an analogous fashion, and transmit infectious pathogens to wild salmon. One easily detectable pathogen is the salmon louse, *Lepeophtheirus salmonis*, an ectoparasite commonly associated with farm-origin epizootics (Marty *et al.* 2010), and depressed adjacent wild salmon populations in regions with salmon farms (Krkosek *et al.* 2007a, 2011a; Connors *et al.* 2010; Krkosek and Hilborn 2011). I therefore asked: "Can sea lice survive travel from salmon farms to processing facilities, mechanical disturbance during processing, and final treatment of effluent before release?"

## Material and methods

Effluent samples were collected from the Walcan fish processing facility situated off the east coast of Vancouver Island, BC (Figure 1). During sampling, the facility was processing Atlantic salmon reared in net-pens on the west coast of Vancouver Island (~450 km via the most direct ocean route). Effluent was collected from an outflow pipe (20 cm diameter; Figure 2) located 27 m below the surface using a plankton-net (50 cm diameter, 125  $\mu\text{m}$  mesh) during two daylight-hour dives at slack tide on February 3 and 8, 2010. The plankton-net was folded over the opening to prevent water from entering during the descent to the pipe. Once at the pipe, the plankton-net was held over the opening for five minutes. However, the net was full within seconds, and more effluent billowed out rather than filtered through the net during our collection period due to the fluid's high viscosity and discharge volume. After five minutes, the opening of the plankton-net was refolded and carried to the surface. The effluent samples were transferred to one-litre glass bottles, packed in ice, and transported to the laboratory for processing. In the lab, samples were filtered using a 200  $\mu\text{m}$  mesh, and residue was sorted and identified using a dissecting microscope (10-30X) within 36 hours of collection. A sub-sample of three fish scales recovered from the effluent were selected at random and sent to Canada's Fisheries and Oceans sclerochronology laboratory for species identification.

## Results

Volumetrically, mucus dominated both samples. Fish scales, tissue fragments, and blood flakes were the most commonly identified discrete items (Table 1); fish scales ranged from 5-8 mm in diameter, and were positively identified as adult Atlantic salmon, *Salmo salar*. More than 100

*Lepeophtheirus salmonis* eggs and living nauplii were also retained from each five-minute sample, as well as several live male and gravid *L. salmonis* females with attached egg-strings.

## **Discussion**

Processors of farm salmon are an unacknowledged point source of a non-trivial pathogen on Canada's Pacific coast. Although salmon farms routinely experience sea lice epizootics (Marty *et al.* 2010; Krkosek 2010), processing facilities of farm salmon have not previously been identified as a source for lice. Hundreds of live sea lice (eggs, larvae, and adults) were recovered directly from the effluent of a facility processing fish transported across numerous natural boundaries, resulting in a net-addition of lice to the region. Accurately quantifying the magnitude of pathogen release from the processing facility is difficult given the inherent variation in numbers of fish processed at any given time. However, considering that processing effluent from diseased farm salmon has been identified as a source for more virulent fish pathogens than sea lice (Vagsholm *et al.* 1994; Jarp and Karlsen 1997), future research aimed at identifying potential pathogens, and quantifying associated magnitude of release, is warranted.

Because processors and farms are often separated by large distance, three under-appreciated disease processes might occur. First, pathogens may be transferred to new regions. Fish scales retrieved from effluent were positively identified as Atlantic salmon, and the sea lice recovered in this study likely originated on infected Atlantic salmon farmed in a distant region (see Figure 3.1). Second, extant but discrete pathogen populations may experience genetic recombination favoring resistant strains. Loss of sensitivity to chemotherapeutants by sea lice is a consequence

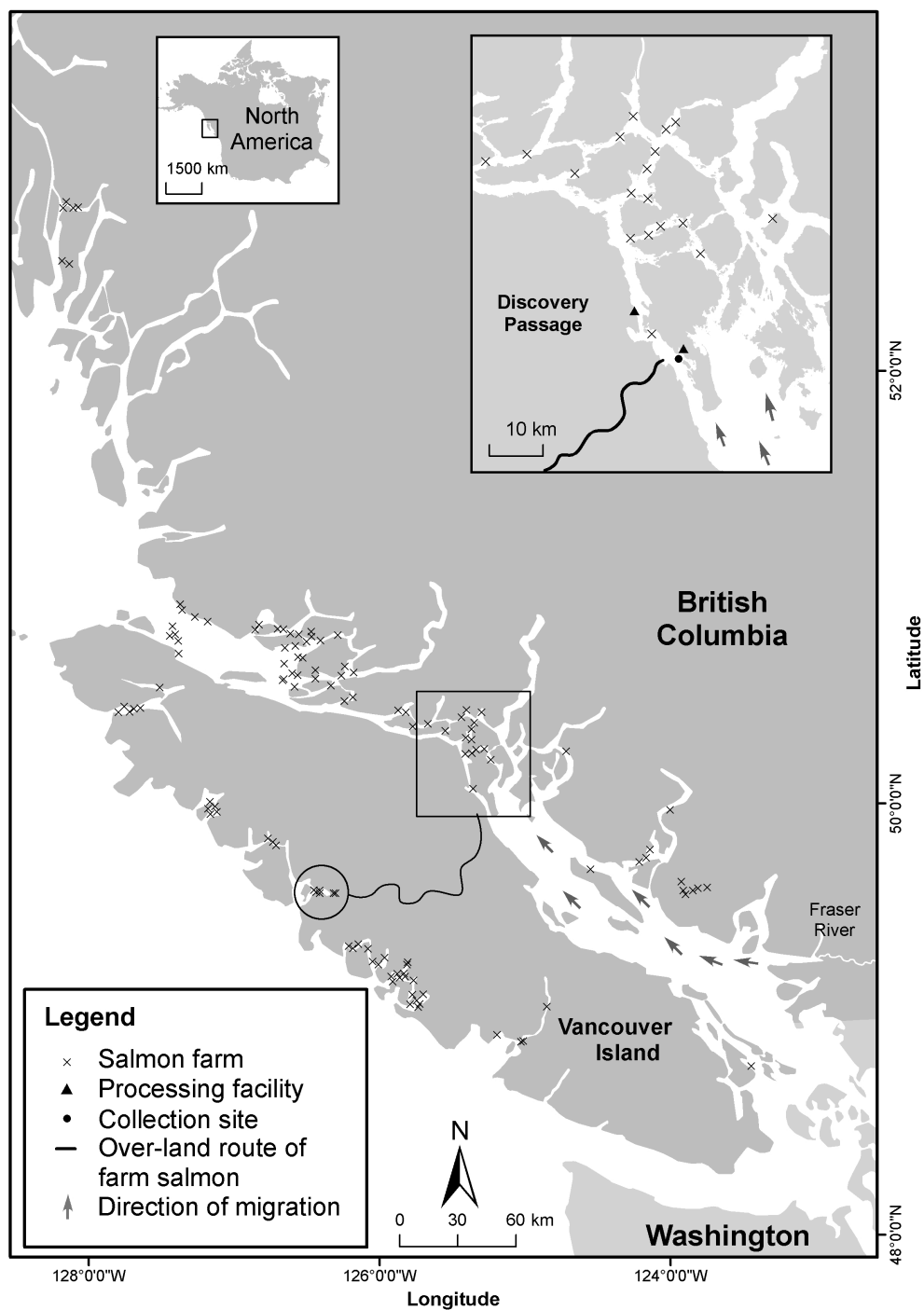
of salmon farming (BurrIDGE *et al.* 2010). Resistance dynamics that are typically constrained within a population may spill over to new populations when pathogens and parasites are transferred between geographical regions. My work suggests that lice had survived overland transport on refrigerated farm fish to a processor across natural and management boundaries, which could promote accelerated resistance of sea lice to commonly used therapeutants. Finally, non-target populations will likely face unpredictable interaction effects with introduced pathogens. The processing facility I investigated is situated along a primary corridor utilised by Canada's largest annual migration of wild juvenile salmon (predominantly from the Fraser River). Wild Fraser River sockeye caught near the facility hosted the highest numbers of *Lepeophtheirus salmonis* of any other site near and far from salmon farms in the region (Price *et al.* 2011). Sea lice from salmon farms are a known threat to already vulnerable wild salmon populations in British Columbia (Krkosek *et al.* 2007a, 2011a; Connors *et al.* 2010; Krkosek and Hilborn 2011), but see Marty *et al.* (2010), and processing facilities may intensify infection pressure to vulnerable fish. Importantly, while farm-origin sea lice populations will rise and fall as salmon farms are stocked, harvested, and fallowed, processing plants have the capability of continuous pathogen release.

Current bio-security regulations in Canada appear inadequate to mitigate the threat of pathogen release. Canada's guidelines for fish processors require fine screening of effluent before release, using a minimum mesh size of 0.71 mm (Environment Canada 1975). The recovery of adult sea lice, fish tissue, and fish scales measuring up to 8 mm reveals a deficiency in mandatory physical treatment of effluent. The viability of sea lice and eggs from effluent shows a similar deficiency

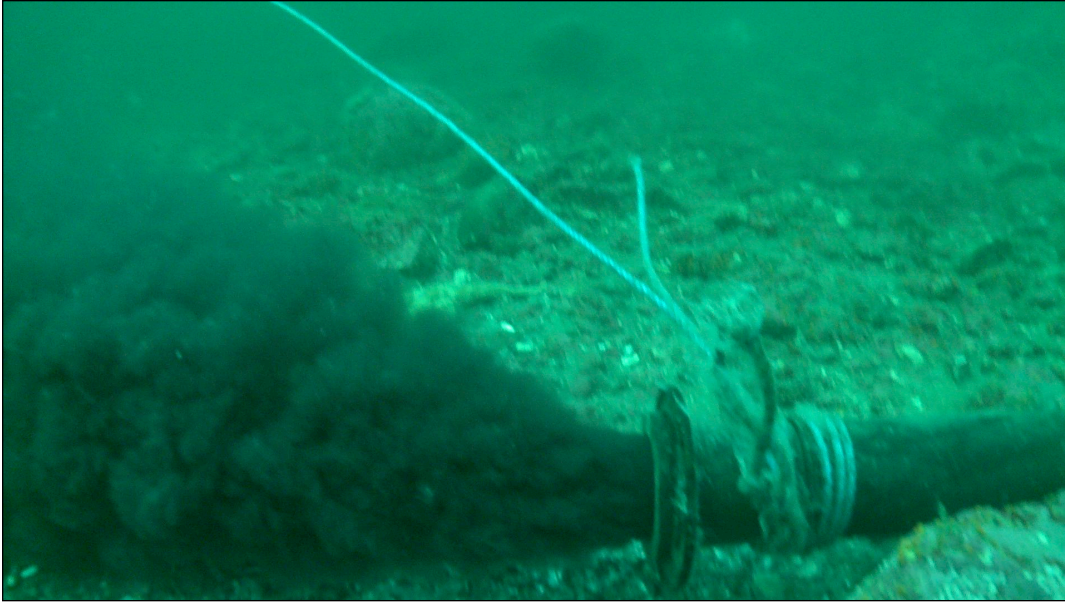
in the use of disinfection or chemical treatments to reduce pathogen transmission. Untreated blood, tissue, and mucus from infected fish pose a serious risk of disease transmission to wild fish (Totland *et al.* 1996) and adjacent farm fish (Vagsholm *et al.* 1994; Jarp and Karlsen 1997), and the disinfection of farm salmon waste from processing has been effective at diminishing disease transmission in some regions of Europe (Murray *et al.* 2010). I recommend development of a mandatory bio-security program in Canada to ensure that facilities processing farm salmon minimize transmission of infectious fish pathogens to wild populations. To be effective, such a policy would require, at a minimum, the disinfection and mechanical treatment of effluent before release, and a robust monitoring effort to ensure compliance as is implemented in Europe (Murray *et al.* 2010).

**Table 3.1.** Contents from effluent samples retrieved during five-minute collections from the outflow pipe of a farm salmon processing facility on Canada's west coast.

Contents	Collection 1	Collection 2
Mucus/blood flakes/tissue shards	Present	Present
Fish scales	436	401
<i>L. salmonis</i> eggs/nauplii	129	102
<i>L. salmonis</i> adult males	4	2
<i>L. salmonis</i> gravid females	7	-
Egg-strings with eggs	3	4
Empty egg-strings	2	3



**Figure 3.1.** Collection site of effluent samples retrieved from a farm salmon processing facility on Canada's Pacific coast.



**Figure 3.2.** Effluent discharged from the outflow pipe of a farm salmon processing facility on Canada's Pacific coast.

## Chapter 4

Zooplankton dynamics and prey selectivity of Fraser River sockeye salmon (*Oncorhynchus nerka*) during their early marine migration in British Columbia, Canada.

## **Abstract**

Sockeye salmon (*Oncorhynchus nerka*) from the Fraser River have experienced some serious challenges recently, and poor habitat conditions in the Georgia Strait have been suggested as a possible cause of their long-term decline. My study examines the prey assemblage, diet composition, and foraging selectivity of juvenile Fraser sockeye during 2009 and 2010, and investigates whether food resources might be a limiting factor in their early marine survival. Sockeye showed high prey diversity, with preference for euphasiid, amphipod, decapod, terrestrial insect, fish, egg, and cumacean prey. Calanoid copepods, the most abundant available prey, were not strongly selected in either year. Zooplankton densities increased with increasing distance north, and approached statistical significance. Sockeye appeared to have an abundant prey resource pool while migrating through south-coast British Columbia, and there was consistency in foraging success between years and throughout the migration period. Moreover, I could not find evidence of resource limitations that might suggest juvenile sockeye were strongly food deprived during the years of this study. However, more work is needed to test whether juveniles are obtaining a sufficient ration to grow and survive their first winter at sea, and to determine whether the timing of their migration coincides with peak prey abundance under the influence of warming ocean conditions.

## **Introduction**

Sockeye salmon (*Oncorhynchus nerka*) is the northeast Pacific's most economically and culturally important salmon species. Populations of sockeye inhabiting southern portions of their range are in decline (IUCN 2008), and those returning to the Fraser River in particular have

experienced some serious challenges recently. The 2009 return of sockeye to the Fraser was the lowest in 50 years, prompting the Canadian government to launch a Judicial Inquiry to investigate the cause of the decline and identify imminent threats to the survival of sockeye (Cohen Commission 2010). Despite a substantial rebound in 2010, there has been a consistent decrease in productivity of most Fraser River sockeye populations that has occurred since the early 1990s (Peterman and Dorner 2011).

Mortality of salmon during their early marine life stage has long been thought to be important in limiting overall abundance (Ricker 1976; Peterman 1982; Beamish *et al.* 2004). Marine survival of juveniles is believed to be a function of their early marine growth (Farley *et al.* 2007; Duffy and Beauchamp 2011), with mortality occurring during two distinct phases (Beamish and Mahnken 2001). The first phase is assumed to be predation-based and occurs early after juveniles enter the marine environment, and the second phase occurs after the first summer when slower growing individuals have a higher probability of mortality (Beamish *et al.* 2004). Both phases are likely linked to ocean conditions in general, and available food resources in particular. Poor habitat conditions for sockeye during their early marine migration from the Fraser River are thought to be the major cause of their recent decline (Peterman *et al.* 2010). However, a substantial rebound in 2010 may indicate a return to favourable ocean conditions.

Georgia Strait is a primary migration corridor for sockeye leaving the Fraser River (Groot and Cooke 1987; Price *et al.* 2011). This coastal sea also receives emissions and waste discharges from the activities of several million humans (Ross 2006). Resident biota can be exposed to the

cumulative effects of pollution, fishing, habitat destruction, introduction of invasive species, and climate change (Johannessen and Macdonald 2009). Secondary production in Georgia Strait has been declining since at least 2001 (Johannessen and Macdonald 2009), and a change in the zooplankton assemblage may also be underway. Knowledge of the feeding habits and prey abundance for Fraser River sockeye during their migration through Georgia Strait is necessary to determine whether food limitations occur (Landingham *et al.* 1996; Schabetsberger *et al.* 2003), yet this information is incomplete and antiquated.

An expert panel of scientists recently recommended the undertaking of an ecological investigation of Georgia Strait to advance our knowledge concerning the state of Fraser sockeye productivity (Peterman *et al.* 2010). My study is a first step towards this goal, with an aim to understand factors influencing the early marine survival of Fraser sockeye as they migrate through Georgia Strait. I examine the prey assemblage, diet composition, and foraging selectivity of juvenile sockeye, and specifically investigate whether the timing of migration through Georgia Strait is a factor in their feeding success, and whether food limitations have occurred.

## **Materials and methods**

I collected juvenile sockeye from inland marine waters of northern Georgia Strait and southern Johnstone Strait from May 24 to July 20, 2009 ( $n = 162$ ) and May 14 to June 21, 2010 ( $n = 186$ ; Figure 4.1), as part of a broader ecological investigation. Capture locations were selected based on their proximity to salmon farms, where each location was sampled weekly; up to 10 juveniles per location were retained for diet analysis when the number of fish caught at a site exceeded 50.

The northern Georgia Strait and southern Johnstone Strait region hosts the largest juvenile sockeye salmon migrations on Canada's west coast (Groot and Cooke 1987), and the time period matches the migration of juvenile Fraser River sockeye through the study region (Price *et al.* 2011). Given the northbound migration of sockeye through the study region, the most northern sites were not sampled during the initial two weeks, and the most southern sites were not sampled during the final two weeks; the southern-most site was only sampled once during mid-May in 2010. All sockeye collections occurred during daylight hours, which is the primary feeding time of juvenile salmonids for this latitude and time of the year (Perry *et al.* 1996). I used a modified purse seine (70 m long, 10 m deep, 6 mm mesh) to capture sockeye; after pursing the net, the catch was concentrated in the bunt of the seine, and fish were dip-netted out from the seine, humanely euthanised, and immediately placed in 1 L bottles of 7% formalin-seawater solution. In the laboratory, individual fish were weighed and measured (fork length) prior to removing stomachs. Sea surface temperature and salinity were recorded during each sampling event using a YSI-30 SCT meter (Table 4.1).

Stomach contents were examined using a Wild M-7 dissecting microscope, weighed to the nearest 0.01 g (wet weight), and counted and divided into taxonomic categories. The percentages per taxonomic category for major components of each stomach were estimated visually. I used a feeding index ( $I_F$ ) as a measure of foraging success, expressed as the percentage of body weight consisting of food items, which standardized for differences in body size using the formula:

$$I_F = M_{sc} (M)^{-1} 100$$

where  $M_{sc}$  is the mass of the stomach contents in grams, and  $M$  is fish mass in grams (Farley *et al.* 2007). I used relative abundance ( $A$ ) as the primary metric to rank prey taxa, expressed as percent contents and calculated as:

$$\%A_{i,f} = A_{i,f} (\sum A_{i,f})^{-1} 100$$

where  $A_{i,f}$  is the total of prey category  $i$  in the  $f^{\text{th}}$  sockeye salmon stomach sampled for diet analysis. I also calculated the frequency-of-occurrence of each major prey group ( $F$ ), expressed as a percent, and calculated as:

$$\%F = O(S)^{-1} 100$$

where  $O$  is the occurrence of a prey item, and  $S$  is the number of samples examined (sockeye stomachs or zooplankton). Finally, I examined prey selectivity by juvenile sockeye salmon using the electivity index ( $E_i$ ):

$$E_i = (r_i - p_i) / (r_i + p_i)$$

where  $r_i$  is the numerical proportion of the  $i^{\text{th}}$  taxon in the stomachs; and  $p_i$  is the proportion of the same taxon in the environment (Ivlev 1961). The electivity values provide a species-specific measure of prey selection by allowing a comparison of stomach contents to available prey.

Values for  $E_i$  range from  $-1.00$  to  $1.00$ , where  $1.00$  indicates the highest selectivity (i.e., present in the diet, but never in the zooplankton samples), and  $-1.00$  indicates lowest selectivity (i.e., never in the diet, but present in the zooplankton samples).

I also retained zooplankton samples at each sockeye location in 2009 ( $n = 16$ ) and 2010 ( $n = 20$ ) using a plankton net (1 m diameter, 125  $\mu\text{m}$  mesh) towed vertically from a depth of 20 m. Juvenile sockeye more often select prey from surface waters than at a depth of 50 m

(Landingham *et al.* 1996). Zooplankton samples from each location were placed in 1 L bottles of 10% formalin-seawater solution. In the laboratory, plankton samples were decanted into a 63  $\mu\text{m}$  sieve, and rinsed under a stream of water to remove formalin. The sample was returned to a 1 L bottle, topped-up with water, and run through a Folsom splitter to generate sub-samples.

Subsamples were placed on a counting dish and the relative abundance of zooplankton taxa were determined using a dissecting microscope. Zooplankton density ( $ZD$ ) was calculated for 2010 samples only (no flow meter was used in 2009), using the formula:

$$ZD = A/V$$

where  $A$  is the total abundance of zooplankton per sample, and  $V$  is the corresponding volume to pass through the plankton-net ( $V = \pi r^2 * d$ ;  $r$  is the radius of the plankton-net,  $d$  is the distance of the tow measured with a RIGO flow meter).

I tested whether zooplankton density depended on latitudinal distance using linear regression analysis. Distance was set to zero for the southern-most collection site, and all other sites were measured in kilometers from this reference location using the shortest linear distance by sea. I also tested whether foraging success depended on migration timing using linear regression analysis. Migration day was set to zero for the first collection of stomachs in a given sampling year, and all other subsequent days were sequentially counted. I transformed feeding index data ( $y$ ) to  $\log_e(y + 0.5)$  to correct for non-normality and unequal variances, and generated all analyses using SPSS 18.0 for Mac (SPSS 2010).

## Results

Calanoid copepods were the most abundant group in plankton samples of 2009 and 2010, followed by barnacles; both groups were ubiquitous throughout the study area (Table 4.2). Remaining zooplankton were much less abundant in each year. The combined group 'other' in 2009 included several fish larvae and the marine ectoparasite, *Caligus clemensi*. Total zooplankton densities increased with increasing distance north, which approached statistical significance ( $r^2 = 0.171$ ,  $df = 17$ ,  $p = 0.088$ ; Figure 4.2), and showed the largest aggregations where northern Georgia Strait and southern Johnstone Strait converge.

Juvenile sockeye showed high prey diversity across the study area (Table 4.3). Of 67 246 food items identified, calanoid copepods were the most common item in both abundance and frequency of occurrence for 2009 and 2010; euphasiids were the second most numerically consumed prey in both years. An adult female *C. clemensi* was found in a sockeye stomach in 2010. Despite the large prey diversity, juvenile sockeye in both years consistently and strongly selected for euphasiid, amphipod, decapod, terrestrial insect, fish, egg, and cumacean prey ( $E_i > 0.75$ ), and routinely selected against brachyuran and cladoceran prey (Figure 4.3); calanoid copepods, the most abundant available prey, were not strongly selected for in either year. Although foraging success did not differ between migration years ( $t = 0.947$ ,  $df = 240$ ,  $p = 0.345$ ), the percentage of empty stomachs was higher in 2009 (7.0%) compared to 2010 (0.0%); foraging success remained relatively constant over the migration period ( $r^2 = 0.002$ ,  $df = 29$ ,  $p = 0.806$ ; Figure 4.4).

## Discussion

My study describes the available zooplankton community as prey for Fraser River sockeye salmon in coastal British Columbia. Prey were more concentrated in the north, which may help explain migratory behaviour of juveniles through the study region. Juvenile sockeye demonstrated high prey diversity, with preference for particular prey, and temporal similarities in foraging success that may reflect short-term food resource stability. Furthermore, results suggest that sockeye may currently be experiencing favorable ocean conditions.

Zooplankton are dominated by copepods, with the largest aggregations at the Johnstone Strait front. Copepods were the most abundant prey group in the epipelagic zooplankton community, which is consistent for this region (Legare 1957; Harrison *et al.* 1983), and the marine waters of northern British Columbia and southeast Alaska (Landingham *et al.* 1996). Zooplankton abundance was not spatially uniform, however. For example, zooplankton densities increased with increasing distance north, with the highest densities recorded at the approach of Johnstone Strait. Although there remains a paucity of zooplankton data for my study area (i.e., northern Georgia Strait and southern Johnstone Strait), phytoplankton densities during spring in Georgia Strait are highest in the northern and southern regions where the most complete mixing of fresh water and sea water occur (Hutchinson and Lucas 1931; Stockner *et al.* 1979; Parsons *et al.* 1981; Harrison *et al.* 1983). Because zooplankton aggregate where food is abundant, such as at biological fronts (Johannessen and Macdonald 2009), my data likely reflect actual differences in zooplankton densities, and may partly explain the rapid migration of juvenile sockeye through Georgia Strait (i.e., 26 km per day; Welch *et al.* 2009).

Juvenile sockeye that migrated through Georgia and Johnstone Straits primarily consumed zooplankton. Prey diversity was high, consisting of more than 12 taxa each year, which matches previous studies in Georgia Strait and other regions of the northeast Pacific (Healey 1980; Brodeur and Pearcy 1990; Healey 1991; Landingham *et al.* 1996; Haegele 1997). Despite the large numbers of species consumed, my results show evidence of selectivity for particular prey. Sockeye routinely selected for euphasiid, amphipod, decapod, terrestrial insect, fish, egg, and cumacean prey, and avoided abundant brachyuran and cladoceran prey; calanoid copepods, the most abundant and consumed prey, were not strongly selected in either year. Calanoid copepods have been described as a major diet item for juvenile salmon generally, but are not considered preferred prey (Pearcy 1992), and sockeye have been shown to avoid numerically dominant copepods elsewhere (Landingham *et al.* 1996). However, the ability to switch to alternative prey (such as copepods) when preferred prey are limiting, may be an important factor in the early marine survival of sockeye. Previous research has shown larval fish as principal prey for sockeye during some years and in certain habitats (Hartt and Dell 1986; Landingham *et al.* 1996), and the low consumption of fish observed in my study may reflect limited availability of this potentially important food resource.

This is the first report of adult *Caligus clemensi* in zooplankton and stomach samples of sockeye salmon, and may provide insight into a poorly understood ecology. *Caligus clemensi* is a common external parasite of Atlantic salmon (*Salmo salar*) on salmon farms in the study region, and juvenile sockeye (Korman 2011; Price *et al.* 2011). Little is known as to how *C. clemensi*

maximize their chance of finding a suitable host, but in addition to the broadcast of eggs in marine waters, it may involve adults dispersing in the plankton. In Europe, adult *C. elongatus* are commonly found in the plankton (Neilson *et al.* 1987), and are known to transfer between fish (Bruno and Stone 1990). However, it remains unknown as to whether *C. clemensi* in the North Pacific behave similarly. The capture of adult *C. clemensi* in marine waters and within a sockeye stomach suggests a possible dispersal mechanism beyond the broadcast of eggs that may involve adult transfer; such a mechanism would enable the redistribution of lice among hosts, and prevent host mortality and loss of lice habitat (Costello 2009). Additionally, their mechanism for host attachment may involve attracting hosts by appearing as a crustacean prey, and upon approach, may evade predation and attach to the host as has been observed for *Lepeophtheirus salmonis* (Connors *et al.* 2008); the adult louse in a sockeye stomach may represent a failed attempt of parasitism.

Temporal similarities of sockeye foraging success may reflect short-term food resource stability. Predictions of climate warming effects on Fraser River sockeye include changes in the abundance and distribution of prey in Georgia Strait, which are thought to result in a migration-timing mismatch with juveniles and nearshore productivity (Healey 2011). Peak abundance of copepods in Georgia Strait is believed to have advanced by two months since the 1960s, and the recent decline in total zooplankton biomass is thought to be due to a mismatch with phytoplankton availability (Johannessen and Macdonald 2009). Juvenile sockeye that migrated through Georgia and Johnstone Straits during 2009 showed comparable foraging success with juveniles that migrated through the region in 2010. Foraging success also remained relatively

unchanged for juveniles throughout the migration period (late May to early July). Coupled with the consistent prey assemblage consumed by sockeye between years, these results suggest that the prey resource pool for juvenile sockeye has been stable for the two years of this study. These results further suggest that prey are abundant even during their peak migration period, which occurred from the second week of June to the end of June. Importantly, the question remains as to whether this apparent resource stability for juveniles during the first few months at sea relate to marine survival and consistent adult returns in subsequent years.

It is unlikely that juvenile sockeye experienced food limitations during the years of this study.

The percentage of body weight consisting of food for sockeye averaged 1.9% during 2009 and 2010. This is the highest recorded for sockeye during their first summer at sea, and comparable with other juvenile salmon for this region and elsewhere. For example, juvenile sockeye examined along British Columbia's north coast showed an average feeding success between 0.4% and 0.9%; levels considered at the time to indicate food limitations (Healey 1991).

Concomitantly, averages for chum, Chinook, and coho in the Strait of Georgia (0.73% - 1.15%; Healey 1980), and coho in Oregon and Washington (1.2% - 2.7%; Fisher and Pearcy 1988) were reported as evidence for an adequate food supply (Healey 1991).

The percentage of juvenile sockeye with empty stomachs (7.0% in 2009; 4.3% overall) further suggests that food limitation (if present during the years of this study) was not widespread.

Previous studies of juvenile sockeye have shown a range in the percentage of empty stomachs from 0% to 75% (Healey 1991; Landingham *et al.* 1996; Haegele 1997; Farley *et al.* 2007), and

coho and Chinook from 2.9% to 12.8% (Brodeur 1992). Although Healey (1991) was the only study to suggest food deprivation with 9% empty stomachs, juvenile sockeye that out-migrated from the Fraser during 1992 and 1993 hosted 47% and 75% empty stomachs, respectively (Haegele 1997), and returned in reduced numbers as adults. Thus, my results suggest that juvenile sockeye were not food deprived during their migration through Georgia and Johnstone Straits during 2009-2010. What my data cannot show, however, is whether juveniles are obtaining a sufficient ration to grow and survive their first winter at sea; an important question that would benefit from future work.

In conclusion, during the years of this study juvenile Fraser River sockeye appeared to have an abundant prey resource pool while migrating through south-coast British Columbia. Consistency in foraging success between years and throughout the migration period reinforces my assertion. The comparison of my results to the previous ecological investigation by Haegele (1997) suggests that ocean conditions for juvenile sockeye have likely returned to a favorable regime, and may result in improved early marine survival. Furthermore, these data should help fisheries managers understand future abundance scenarios in the context of food resources.

**Table 4.1.** Summary statistics (sample mean and SE) for juvenile sockeye salmon examined during 2009 and 2010. The sample size for fish fork-length and mass was the same as the number of stomachs in 2009, and  $n = 93$  in 2010; the number of collection sites for sea surface temperature and salinity readings were 12 (2009) and 18 (2010).

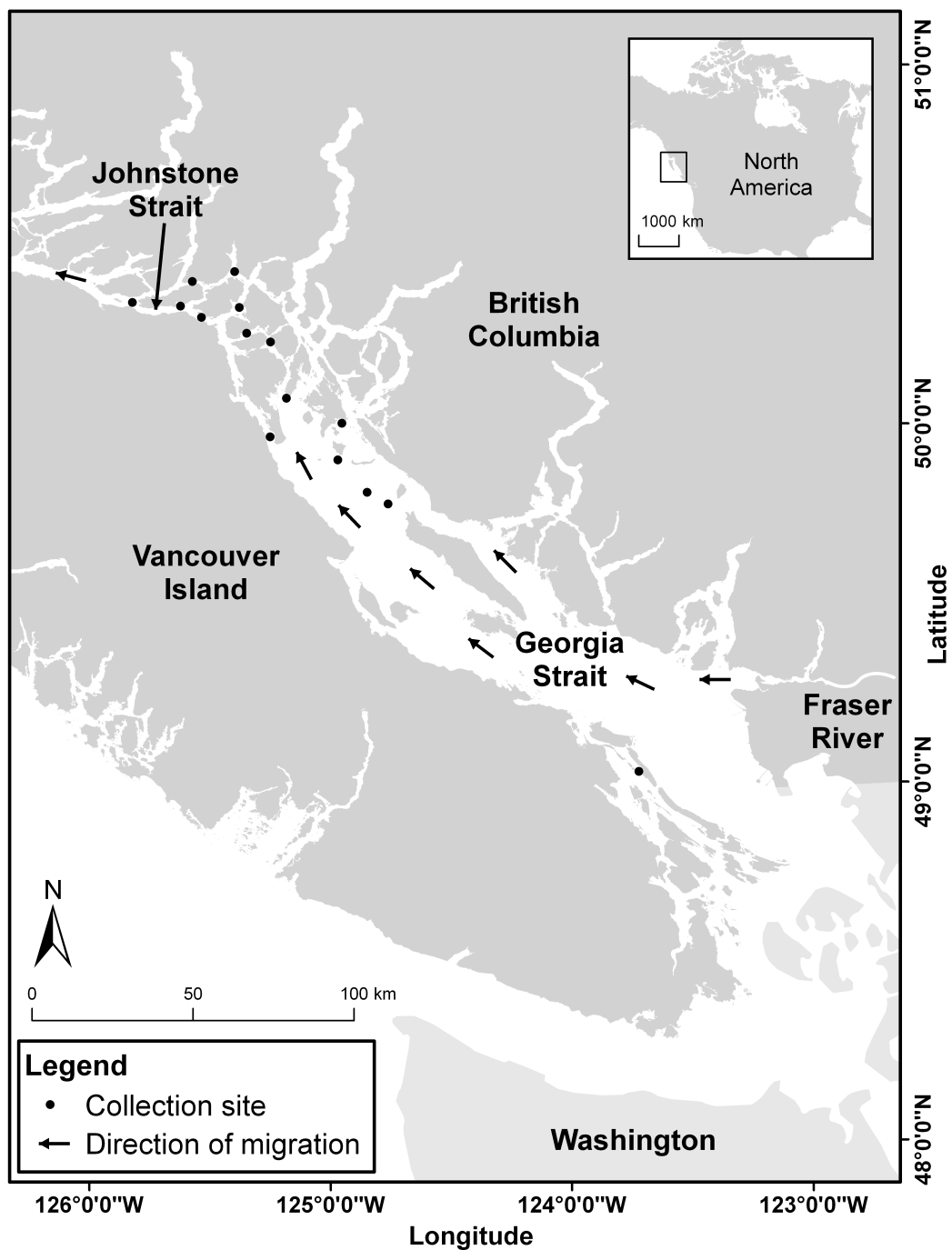
<b>Variable</b>	<b>2009</b>	<b>2010</b>
Number of stomachs	162	186
Fork length (cm)	10.49 (1.02)	16.89 (1.35)
Mass (g)	12.41 (0.39)	12.22 (4.08)
Temperature (°C)	11.57 (0.76)	11.41 (0.37)
Salinity (‰)	29.05 (0.32)	28.65 (0.40)

**Table 4.2.** Zooplankton taxa identified during 2009 and 2010 plankton surveys; percent abundance (A) is the number of individuals of a given taxon divided by the total number of individuals of all taxa, and frequency of occurrence (F) is the frequency of a given taxon in the plankton.

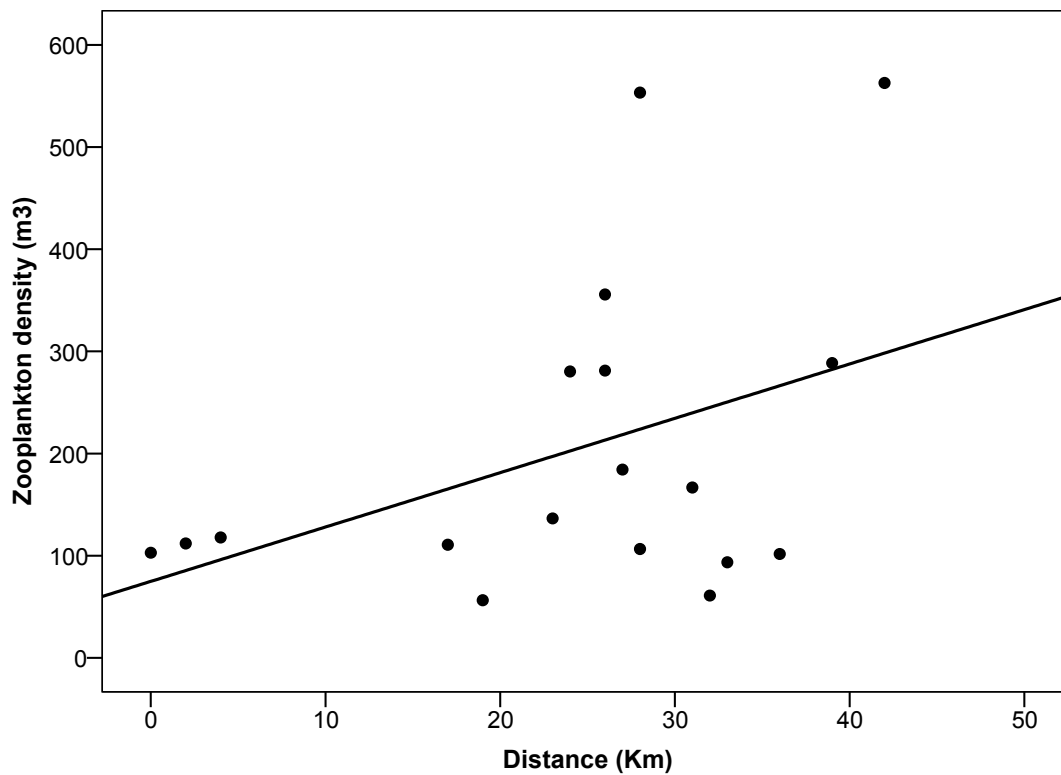
Taxon	2009		2010	
	A (%)	F (%)	A (%)	F (%)
Copepoda	68.7	100.0	74.9	100.0
Brachyura	12.3	100.0	17.8	100.0
Euphasacea	2.3	100.0	1.8	83.3
Cyclopoida	0.5	50.0	1.7	88.9
Polychaeta	0.6	100.0	1.4	100.0
Cnidaria	2.7	88.9	1.3	100.0
Oikopleura	9.1	100.0	0.6	100.0
Pteropoda	0.2	18.8	0.3	77.8
Harpacticoida	1.8	56.3	0.2	16.7
Decapoda	0.1	6.4	0.2	66.7
Cladocera	3.2	87.5	0.2	94.4
Mollusca	0.4	12.5	0.2	38.9
Echinoderm	0.7	81.3	0.1	77.8
Other	0.3	50.0	0.2	77.8

**Table 4.3.** Prey taxa identified in juvenile sockeye stomachs during 2009 and 2010; relative abundance (A) is the number of individuals of a given taxon divided by the total number of individuals of all taxa, and frequency of occurrence (F) is the frequency of a given taxon in all sockeye stomachs.

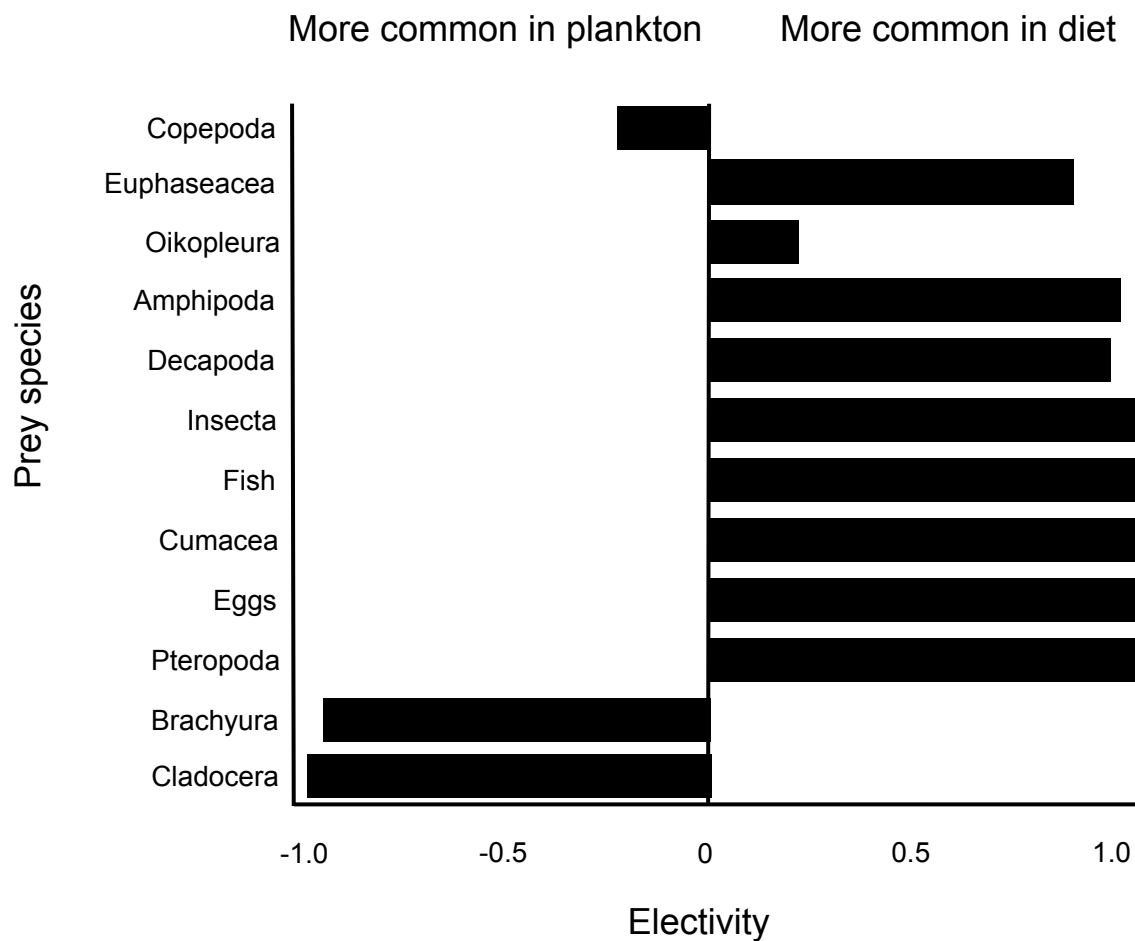
Taxon	2009		2010	
	A (%)	F (%)	A (%)	F (%)
Copepoda	49.9	76.7	32.1	83.9
Euphasacea	16.6	66.3	21.2	62.9
Oikopleura	10.2	39.9	4.4	22.0
Amphipoda	7.5	50.3	15.5	67.7
Decapoda	5.7	33.7	13.1	45.7
Insecta	2.9	15.3	0.4	5.9
Fish	2.9	12.9	11.0	15.1
Cumacea	2.1	9.8	0.1	3.8
Eggs	2.1	3.7	1.1	3.8
Pteropoda	0.1	9.8	0.7	25.3
Brachyura	0.1	32.5	0.4	24.2
Cladocera	0.1	18.0	0.1	16.7



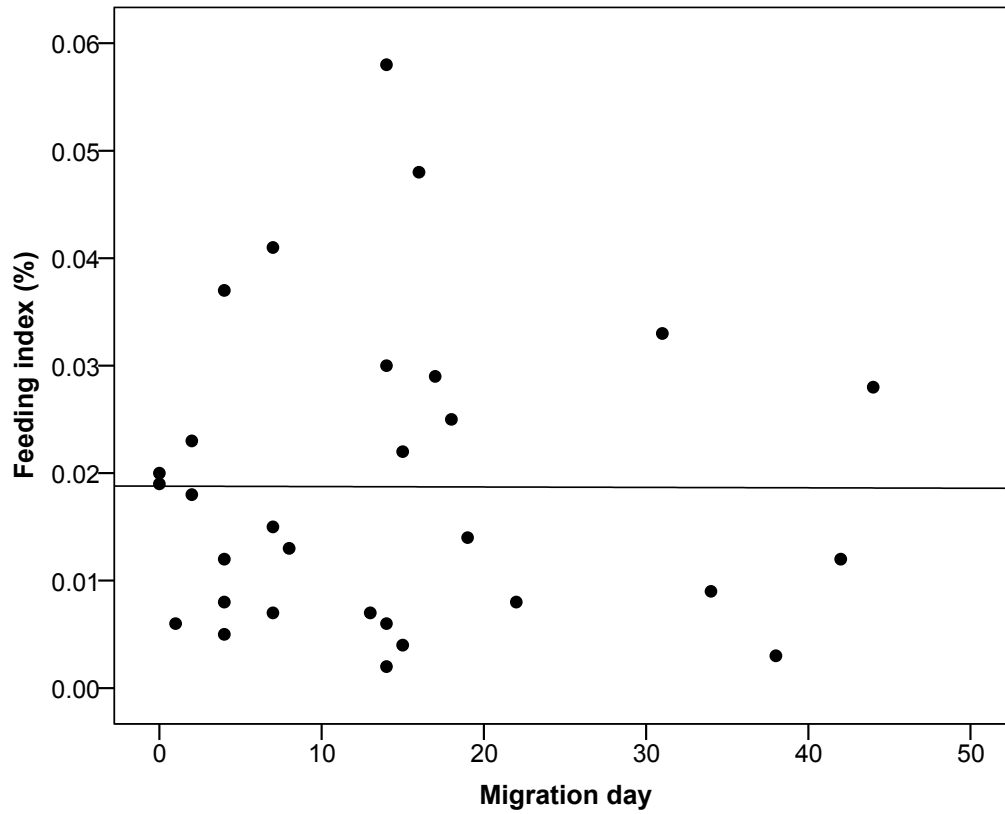
**Figure 4.1.** Study area of Georgia Strait and Johnstone Strait and all associated sampling sites for zooplankton and juvenile sockeye salmon (*Oncorhynchus nerka*) examined during 2009-2010.



**Figure 4.2.** Distribution of zooplankton density (m<sup>3</sup>) in relation to distance '0', the southern-most collection site in 2010.



**Figure 4.3.** Electivity scores (a species-specific measure of prey selection) for primary prey species identified in juvenile sockeye stomachs during 2009-2010. A value of  $-1.0$  indicates lowest selectivity (i.e., never in the diet, but present in zooplankton samples), and  $1.0$  indicates the highest selectivity (i.e., present in the diet, but never in zooplankton samples).



**Figure 4.4.** Feeding index, expressed as percent body weight of stomach contents, in relation to the migration of juvenile sockeye through the study region during 2009-2010. Migration day is the number of days since the start of sampling in a given year.

## General Discussion

A wealth of knowledge regarding Pacific salmon has been amassed (e.g., Groot and Margolis 1991), yet we fail to understand many important details of their life history (Welch *et al.* 2009, 2011). The transition from freshwater to the marine environment is one of the most challenging phases for salmon, as they encounter a suite of new predators and prey (Quinn 2005). Juveniles must grow rapidly to avoid the initial risk of predation, and reach a size and condition that will increase their chances of surviving the first marine winter when food resources are considerably reduced (Beamish and Mahnken 2001; Beamish *et al.* 2004; Duffy and Beauchamp 2011). Factors that slow growth may include: increased predation, competition, reduced food quantity or quality, and pathogens (such as parasites). Reduced habitat quality can enhance the negative effects these factors have on juvenile salmon, and each of these factors, in turn, reduce the quality of habitat. My thesis has explored some of these factors, and here I consider my major findings in light of concerns about sustainability of salmon.

Salmon farms reduce habitat quality by enhancing pathogen infection. My studies show strong associations between open net-pen salmon farms and infections of sea lice on wild juvenile salmon across a large area of coastal BC. Specifically, I show regional differences in parasitism of juvenile salmon between areas with and without salmon farms, as well as within-regional differences between sites of differing exposure levels. I consider the latter to be the stronger test of the hypothesis that salmon farms are significant contributors of sea lice to wild juvenile salmon, because it controls for larger variation among regions in potentially confounding factors such as salinity and temperature. Within the salmon farmed regions of Finlayson, Broughton

Archipelago, and Georgia Strait, juvenile pink, chum, and sockeye salmon hosted significantly more sea lice when exposed to salmon farms. Although mixed-effects modelling showed some variation in results depending on louse and host species, exposure to salmon farms was the most consistent predictor of both *C. clemensi* and *L. salmonis* abundance on juvenile salmon.

A dynamic equilibrium in nature exists between host and parasite, such that parasites are limited by host defenses and an ability to evade the host's responses (Wikel *et al.* 1994). Parasites are typically confined to a few hosts within the population, resulting in a low probability of an extinction event occurring due to high infestation levels (Esch and Fernandez 1993).

Approximately 5% of wild juvenile pink and chum salmon have been shown to host a single sea louse during their early marine migration in areas without salmon farms (Morton *et al.* 2004; Krkosek *et al.* 2007b), and my data from Bella Bella show similar results. Moreover, lice infection of juveniles at locations far from farms within the salmon farmed regions of Finlayson and Broughton Archipelago were equally low, which suggests that juveniles at these locations had not been exposed to salmon farms, and it is likely that a dynamic equilibrium between host and parasite had been maintained.

Epizootics are the result of an imbalance in the host-parasite equilibrium, and salmon farms can perturb the balance of natural salmonid host-parasite systems (Krkosek *et al.* 2006; Costello 2009; Krkosek 2010). Wild juvenile pink and chum salmon hosted consistently elevated levels of sea lice near salmon farms in all salmon farmed regions, which at times approached 50% of fish examined. *Lepeophtheirus salmonis* was the dominant louse infecting juvenile pink and chum

near salmon farms, and this particular louse has been shown to compromise osmoregulation (Bjorn and Finstad 1997), induce behavioral changes that increase predation risk (Krkosek *et al.* 2011b), and result in host death (Costello 2009; Morton and Routledge 2005; Krkosek *et al.* 2006). Moreover, *L. salmonis* is locally associated with juvenile pink and chum mortality (Morton and Routledge 2005; Krkosek *et al.* 2006), and has been implicated in contributing to the population collapse of pinks in the Broughton Archipelago (PFRCC 2002; Krkosek *et al.* 2007a, 2011a; Krkosek and Hilborn 2011; but see contrary results below). It is likely that the infection levels observed on pink and chum near salmon farms in my study will compromise the health of these fish, and may contribute to population-level impacts as has been documented in the Broughton Archipelago (Krkosek *et al.* 2007a, 2011a; Krkosek and Hilborn 2011), and suggested for Georgia Strait (Ford and Myers 2008).

A laboratory study has inferred that louse-induced mortality of wild pink salmon is negligible for juveniles  $> 0.7\text{g}$  in size (Jones and Hargreaves 2009). Given that many wild juveniles I sampled were  $> 0.7\text{g}$ , it may be argued that negative impacts are unlikely to be significant. However, there is a three orders of magnitude difference in infection period between the above laboratory study (i.e., hours and days) and the field conditions experienced by juveniles in my study (i.e., 2 months). Moreover, laboratory studies on lice-induced mortality of wild juvenile pink salmon ignore the indirect effects of infection mentioned previously. For example, louse-infected juveniles show behavioral changes that increase their risk to predation (Krkosek *et al.* 2011b). An investigation that includes both a laboratory and field-based component is needed to understand the potential impacts of sea lice on wild juvenile chum salmon, and further research

aimed at both direct and indirect effects on juvenile pink salmon is required to quantitatively estimate survival.

Do sea lice pose a threat to juvenile sockeye salmon? Juvenile sockeye are primarily infected by the generalist sea louse *C. clemensi*, and research to date has not examined the effects of this louse on sockeye or any other wild juvenile salmonid, though significant fin damage by larval stage lice has been documented on juvenile pinks (Parker and Margolis 1964). *Caligus clemensi* is smaller than *L. salmonis*, and is thought to cause less mechanical damage to juvenile pink and chum salmon (Morton and Routledge 2005; Krkosek *et al.* 2007b; Costello 2009). Moreover, juvenile sockeye are much larger and have developed scales at the time of ocean entry compared to juvenile pink and chum; thus, it is unlikely that the average number of *C. clemensi* observed on sockeye (2-3 lice/fish) exposed to salmon farms would cause direct mortality for healthy fish. However, the effect of sea louse parasitism on juvenile sockeye could be important when combined with multiple stressors (Finstad *et al.* 2007). Sublethal effects, such as reduced body growth due to parasitism, may explain elevated mortality of *L. salmonis*-infected juvenile coho in the Broughton Archipelago, which are larger and presumably more robust to infection than pink salmon (Connors *et al.* 2010; Krkosek *et al.* 2011a). Moreover, the presence and abundance of sea lice on juvenile sockeye may be a proxy for other farm-origin pathogens that go undetected. Given the high intensities of *C. clemensi* observed on some juveniles in my study (i.e., up to 28 lice/fish), concern is justified, and research should be undertaken to understand the extent of threat posed to individual fish and populations.

Processors of farm salmon may further threaten juvenile salmon by pathogen transmission. My data show that a farm salmon processing facility located along a major migratory corridor for juvenile salmon from the Fraser River is a point source of a live fish pathogen. Live sea lice recovered at the outflow pipe of this facility suggest that the effluent had not been treated prior to release. Of considerable concern is the additional recovery of blood, tissue, and mucus; factors shown to pose a significant pathogen risk to fish (Totland *et al.* 1996). Pathogens can disseminate over long distances via farm-to-farm infections (Saksida 2006) and infected parasites (Nyland *et al.* 1994), and farm-origin fish diseases may be further broadcast through the release of untreated effluent from slaughterhouses processing infected farm salmon (Vagsholm *et al.* 1994; Jarp and Karlsen 1997), which may be far removed from source farms. Farmed salmon commonly host fish pathogens (BCMAL 2007, 2008; Korman 2011), and while farm-origin pathogens will rise and fall as salmon farms are stocked, harvested, and fallowed, processing facilities have the capability of continuous pathogen release. At minimum, this facility is undoubtedly polluting sensitive early marine habitat for juvenile salmon with a parasite known to inflict population-level impacts. It is imperative that future research investigates the potential pathogens discharged in processor effluent and, if present, quantifies the associated magnitude of pathogen release, and examines whether salmon migrating through effluent-discharged waters are hosts of associated fish diseases.

Fish that are sub-lethally stressed may be more sensitive to pathogen attacks, and can have reduced tolerance to additional stressors (Iversen *et al.* 2005). Juvenile Atlantic salmon that swam through polluted waters and were subsequently infected by sea lice from salmon farms

showed reduced survival compared to equally infected juveniles that did not (Finstad *et al.* 2007). It is reasonable to believe that juvenile salmon that migrate from the Fraser River and through nursery waters of Georgia Strait are exposed to poor, stressful conditions. Indeed, this coastal sea is at the receiving end of waste discharges and contaminants from the activities of several million human inhabitants (Ross 2006). Importantly, zooplankton production (primary prey for juvenile sockeye, pink, and chum) in Georgia Strait has been declining since 2001 (Johannessen and Macdonald 2009), and the question remains as to whether juveniles are obtaining a sufficient ration to grow and survive their first winter at sea. If the fitness of juvenile salmon is reduced before they are exposed to salmon farms, subsequent pathogen attacks may exacerbate lowered fitness, and lead to higher rates of mortality than otherwise might occur.

Marine habitats of Pacific salmon are predicted to warm by 2-5 °C or more over the next century (Mackas *et al.* 2007; Healey 2011), and this degree of warming will have uncertain but potentially devastating effects on salmon and their ecosystems (Beaugrand and Reid 2003; Beamish 2008). Wild juvenile salmon may be particularly vulnerable to temperature increases during their early marine migration in BC. For example, zooplankton abundance is expected to shift northward and peak earlier under warmer conditions (Mackas *et al.* 2007), which would lead to poorer feeding conditions for juveniles, and this pattern may be underway currently in Georgia Strait (Johannessen and Macdonald 2009). The abundance and distribution of predators may also change with warmer seas, resulting in increased levels of predation on juvenile salmon. Warmer coastal sea temperatures experienced by juvenile Fraser River sockeye have been

correlated with reduced survival, and increased sea temperatures may be a proxy for reduced food or increased predation levels (Hinch *et al.* 1995).

All species of wild salmon face multiple, simultaneous challenges, and wild juvenile salmon migrating through coastal seas may be specifically threatened by contaminants, and pathogen transmission from salmon farms and processing facilities of farmed salmon; food limitations may compound these threats. One of the gravest concerns is that these threats may act together, and that the overarching effect of climate warming may elevate even a minor threat into a significant one (Healey 2011). Pathogens, such as sea lice, are typically of minor consequence to juvenile salmon and salmon populations in areas without salmon farms. However, it has been shown within areas of the Atlantic and Pacific Oceans that there are negative correlations between the number of farmed salmon and decreases in exposed salmon populations, relative to nearby farm-free areas (Ford and Myers 2008). Because sea louse growth in lab-based trials depends strongly, in part, on temperature (Pike and Wadsworth 1999; Costello 2006), warmer coastal seas may increase the prevalence and virulence of these pathogens on wild juvenile salmon.

How can we best conserve salmon populations given the multitude of stressors? It may be easiest to simply prioritize the known threats in order of their severity, as a way towards lessening the overall burden these stressors currently have on salmon in BC. For example, predation by marine mammals or other fishes in nursery habitats might arguably be primarily responsible for juvenile salmon mortality during the first few months at sea (Christensen and Trites 2011); perhaps we should focus our attention on the removal of predators. Pre-spawning mortality due to increasing

water temperatures in the Fraser River may be considered one of the largest overall threats to salmon returning to the Fraser River (Hinch and Gardner 2009), and it may theoretically seem appropriate to focus effort on reducing these threats as a way to increase salmon abundance. However, mitigation measures to reduce these stressors are unlikely to be practical or achievable. Moreover, given the way stressors often interact to produce compound effects and unpredictable results, ranking the overall threats in order of severity may not be useful. Instead, the most successful ranking system may be in terms of ‘do-ability’ (i.e., reducing harm where possible; Harvey 2008). For juvenile salmon during their early marine migration, risks posed by salmon farms can be more easily mitigated than the far-reaching effects on ocean productivity of climate change and ocean acidification, or predator removal. I recommend we begin here.

Options already recommended for minimizing pathogen risks to wild salmon include removal of farm salmon from migration routes of juvenile salmon (Statement from Think Tank of Scientists 2009), and transitioning of salmon farms to closed-containment facilities (LABC 2007). I further recommend the development of a mandatory bio-security program in Canada to ensure that facilities processing farm salmon minimize transmission of infectious fish pathogens to wild populations. To be effective, such a policy would require, at a minimum, the disinfection and mechanical treatment of effluent before release, and a robust monitoring effort to ensure compliance as is implemented in Europe (Murray *et al.* 2010). These are all easily achievable mitigation measures that, if implemented, will undoubtedly help wild salmon populations.

## Bibliography

- Anderson, D.R., Link, W.A., Johnson, D.H., and Burnham, K.P. 2001. Suggestions for presenting the results of data analyses. *Journal of Wildlife Management* 65: 373-378.
- Beacham, T.E., Lapointe, M., Candy, J.R., McIntosh, B., MacConnachie, C., Tabata, A., Kaukinen, K., Deng, L., Miller, K.M., and Withler, R.E. 2004. Stock identification of Fraser River sockeye salmon using microsatellites and major histocompatibility complex variation. *Transactions of the American Fisheries Society* 133: 1117-1137.
- Beacham, T.E., Candy, J.R., McIntosh, B., MacConnachie, C., Tabata, A., Kaukinen, K., Deng, L., Miller, K.M., and Withler, R.E. 2005. DNA-level variation of sockeye salmon in Southeast Alaska and the Nass and Skeena Rivers, British Columbia, with applications to stock identification. *North American Journal of Fisheries Management* 25: 763-776.
- Beaugrand, G., and Reid, P.C. 2003. Long-term changes in phytoplankton, zooplankton and salmon related to climate. *Global Change Biology* 9: 801-817.
- Beamish, R.J. 2008. Impacts of climate and climate change on key species in the fisheries of the North Pacific. North Pacific Marine Science Organization (PICES), Sidney, B.C. PICES Sci. Rep. No. 35.
- Beamish, R.J., and Mahnken, C. 2001. A critical size and period hypothesis to explain natural regulation of salmon abundance and the linkage to climate and climate change. *Progress in Oceanography* 49: 423-437.
- Beamish, R.J., Mahnken, C., and Neville, C.M. 2004. Evidence that reduced early marine growth is associated with lower marine survival of coho salmon. *Transactions of the American Fisheries Society* 133: 26-33.
- Beamish, R.J., Neville, C.M., Sweeting, R.M., Jones, S.R.M., Ambers, N., Gordon, E.K., Hunter, K.L., and McDonald, T.E. 2007. A proposed life history strategy for the salmon louse, *Lepeophtheirus salmonis*, in the subarctic Pacific. *Aquaculture* 264: 428-440.
- Beamish, R.J., Wade, J., Pennell, W., Gordon, E., Jones, S., Neville, C., Lange, K., and Sweeting, R. 2009. A large, natural infection of sea lice on juvenile Pacific salmon in the Gulf Islands area of British Columbia, Canada. *Aquaculture* 297: 31-37.
- Bjorn, P.A., and Finstad, B. 1997. The physiological effects of salmon lice infection on sea trout post smolts. *Norwegian Journal of Freshwater Research* 73: 60-72.
- Bjorn, P.A., and Finstad, B. 2002. Salmon lice, *Lepeophtheirus salmonis* (Krøyer), infestation in sympatric populations of Arctic char, *Salvelinus alpinus* (L.), and sea trout, *Salmo trutta*

(L.), in areas near and distant from salmon farms. ICES Journal of Marine Science 59: 131-139.

British Columbia Ministry of Agriculture and Lands (BCMAL). 2007. Fish health program annual report. Available: [http://www.agf.gov.bc.ca/ahc/fish\\_health2007.pdf](http://www.agf.gov.bc.ca/ahc/fish_health2007.pdf) via the internet. Accessed 28 January 2011.

British Columbia Ministry of Agriculture and Lands (BCMAL). 2008. Fish health program annual report. Available: [http://www.agf.gov.bc.ca/ahc/fish\\_health/Fish\\_Health\\_Report\\_\\_\\_\\_2008.pdf](http://www.agf.gov.bc.ca/ahc/fish_health/Fish_Health_Report____2008.pdf) via the internet. Accessed 28 January 2011.

British Columbia Ministry of the Environment (BCMOE). 2009. Aquaculture production statistics for British Columbia 2007-2008, areas 3.5, 3.3, and 3.2. Oceans and Marine Fisheries Branch. Victoria, BC.

British Columbia Pacific Salmon Forum (BCPSF). 2009. Final report and recommendations to the Government of British Columbia.

Brodeur, R.D. 1992. Factors related to variability in feeding intensity of juvenile coho salmon and Chinook salmon. Transactions of the American Fisheries Society 121: 104-114.

Brodeur, R.D., and Percy, W.G. 1990. Trophic relations of juveniles Pacific salmon off the Oregon and Washington coast. Fishery Bulletin 88: 617-636.

Brooks, K.M. 2005. The effects of water temperature, salinity, and currents on the survival and distribution of the infective copepodid stage of sea lice (*Lepeophtheirus salmonis*) originating on Atlantic salmon farms in the Broughton Archipelago of British Columbia, Canada. Reviews in Fisheries Science 13: 177-204.

Bruno, D.W., and Stone, J. 1990. The role of saithe, *Pollachius virens* L., as a host for the sea lice, *Lepeophtheirus salmonis* Kroyer and *Caligus elngatus* Nordmann. Aquaculture 89: 201-207.

Burnham, K.P., and Anderson, D.R. 1998. Model selection and inference: a practical information-theoretic approach. Springer-Verlag, New York.

Burridge, L., Weis, J.S., Cabello, F., Pizarro, J., and Bostick, K. 2010. Chemical use in salmon aquaculture: A review of current practices and possible environmental effects. Aquaculture 306: 7-23.

Campbell, S.K., and Butler, V.L. 2010. Archaeological Evidence for Resilience of Pacific Northwest Salmon Populations and the Socioecological System over the last ~7,500 years. Ecology and Society 15: 17. Available: <http://www.ecologyandsociety.org/vol15/>

- [iss1/art17/](#) via the internet. Accessed 15 August 2011.
- Christensen, V., and Trites, A.W. 2011. Predation on Fraser River sockeye salmon. Cohen Commission Technical Report 8. Vancouver, BC. 129 p.
- Cohen Commission. 2010. Commission of inquiry into the decline of Sockeye salmon in the Fraser River. Available: <http://www.cohencommission.ca/en/TermsOfReference.php> via the internet. Accessed 28 January 2011.
- Connors, B.M., Krkosek, M., and Dill, L.M. 2008. Sea lice escape predation on their host. *Biology Letters* 4: 455–457.
- Connors, B.M., Krkosek, M., Ford, J., and Dill, L.M. 2010. Coho salmon productivity in relation to salmon lice from infected prey and salmon farms. *Journal of Applied Ecology* 47: 1372-1377.
- Costello, M.J. 2006. Ecology of sea lice parasitic on farmed and wild fish. *Trends in Parasitology* 22: 475-483.
- Costello, M.J. 2009. How sea lice from salmon farms may cause wild salmonid declines in Europe and North America and be a threat to fishes elsewhere. *Proceedings of the Royal Society of London B* 276: 3385-3394.
- Daszak, P., Cunningham, A., and Hyatt, A. 2000. Emerging infectious diseases of wildlife: threats to biodiversity and human health. *Science* 287: 443-449.
- Debruyn, A.M.H., Trudel, M., Eyding, N., Harding, J., McNally, H., Mountain, R., Orr, C., Urban, D., Verenitch, S., and Muzumder, A. 2006. Ecosystem effects of salmon farming increase mercury contamination in wild fish. *Environmental Science and Technology* 40: 3489-3493.
- Dobson, A., and Foufopoulos, J. 2001. Emerging infectious pathogens of wildlife. *Philosophical Transactions of the Royal Society of London B* 356: 1001–1012.
- Duffy, E.J., and Beauchamp, D.A. 2011. Rapid growth in the early marine period improves the marine survival of Chinook salmon (*Oncorhynchus tshawytscha*) in Puget Sound, Washington. *Canadian Journal of Fisheries and Aquatic Sciences* 68: 232-240.
- English, K.K., Oeacock, D., and Spilsted, B. 2006. North and central coast core stock assessment program for salmon. Prepared for Pacific Salmon Foundation and Fisheries and Oceans Canada by LGL Limited Environmental Research Associates and Fisheries and Oceans Canada, Sidney, BC.

- Environment Canada. 1975. Fish processing operations liquid effluent guidelines. Regulations, codes and protocols report EPS 1-WP-75-1.
- Esch, G.W., and Fernandez, J.C. 1993. A functional biology of parasitism. Chapman and Hall.
- Farley, E.V., Murphy, J.M., Adkison, M., and Eisner, L. 2007. Juvenile sockeye salmon distribution, size, condition and diet during years with warm and cool spring sea temperatures along the eastern Bering Sea shelf. *Journal of Fish Biology* 71: 1145-1158.
- Finstad, B., Kroglund, F., Strand, R., Stefansson, S.O., Bjorn, P.A., Rosseland, B.O., Nilsen, T.O., and Salbu, B. 2007. Salmon lice or suboptimal water quality - reasons for reduced postsmolt survival? *Aquaculture* 273: 374-383.
- Fisher, J.P., and Percy, W.G. 1988. Growth of juvenile coho salmon (*Oncorhynchus kisutch*) off Oregon and Washington, USA, in years of differing coastal upwelling. *Canadian Journal of Fisheries and Aquatic Sciences* 45: 1036-1044.
- Food and Agriculture Organization of the United Nations (FAO). 2008. The state of world fisheries and aquaculture 2008. Available: <ftp://ftp.fao.org/docrep/fao/011/i0250e/i0250e.pdf>. via the internet. Accessed 14 March 2011.
- Ford, J.S., and Myers, R.A. 2008. A global assessment of salmon aquaculture impacts on wild salmonids. *Public Library of Science Biology* 6: 411-417.
- Fraser, N.L. 2009. Sea-cage aquaculture, sea lice, and declines of wild fish. *Conservation Biology* 23: 599-607.
- Gende, S.M., Edwards, R.T., Willson, M.F., and Wipfli, M.S. 2002. Pacific salmon in aquatic and terrestrial ecosystems. *Bioscience* 52: 917-928.
- Goldburg, R.J., and Naylor, R.L. 2005. Future seascapes, fishing, and fish farming. *Frontiers in Ecology and the Environment* 3: 21-28.
- Gottesfeld, A.S., Proctor, B., Rolston, L.D., and Carr-Harris, C. 2009. Sea lice, *Lepeophtheirus salmonis*, transfer between wild sympatric adult and juvenile salmon on the north coast of British Columbia, Canada. *Journal of Fish Diseases* 32: 45-57.
- Gozlan, R.E., Peeler, E.J., Longshaw, M., St-Hilaire, S., and Feist, S.W. 2006. Effect of microbial pathogens on the diversity of aquatic populations, notably in Europe. *Microbes and Infection* 8: 1358-1364.
- Groot, C., and Cooke, K. 1987. Are the migrations of juvenile and adult Fraser River sockeye

- salmon (*Oncorhynchus nerka*) in near-shore waters related? In: Smith, H.D., Margolis, L., Wood, C.C., editors. Sockeye salmon (*Oncorhynchus nerka*) population biology and future management. Canadian Special Publication of the Fisheries and Aquatic Sciences 96. pp. 53-60.
- Groot, C., and Margolis, L. 1991. Pacific salmon life histories. UBC Press, Vancouver, BC. 564 p.
- Haegle, C.W. 1997. The occurrence, abundance, and food of juvenile herring and salmon in the Strait of Georgia, British Columbia in 1990 to 1994. Canadian Manuscript Report of Fisheries and Aquatic Sciences 2390.
- Halpern, B.S., Ebert, C.M., Kappel, C.V., Madin, E.M.P., Micheli, F., Perry, M., Selkoe, K.A., and Walbridge, S. 2009. Global priority areas for incorporating land–sea connections in marine conservation. Conservation Letters 2: 189-196.
- Harrison, P.J., Fulton, J.D., Taylor, F.J.R., and Parsons, T.R. 1983. Review of the biological oceanography of the Strait of Georgia: pelagic environment. Canadian Journal of Fisheries and Aquatic Sciences 40: 1064-1094.
- Hartt, A.C., and Dell, M.B. 1986. Early oceanic migrations and growth of juvenile Pacific salmon and steelhead trout. International North Pacific Fisheries Commission Bulletin 46: 1–105.
- Harvey, B. 2008. Nowhere to hide: salmon versus people in the 21st century. A report to the B.C. Pacific Salmon Forum. Victoria, B.C.
- Healey, M.C. 1980. The ecology of juvenile salmon in Georgia Strait, British Columbia. In: McNeil, W.J., and Himsworth, D.C., editors. Salmonid ecosystems of the North Pacific. pp. 203-229.
- Healey, M.C. 1991. Diets and feeding rates of juvenile pink, chum, and sockeye salmon in Hecate Strait, British Columbia. Transactions of the American Fisheries Society 120: 303-318.
- Healey, M.C. 2011. The cumulative impacts of climate change on Fraser River sockeye salmon (*Oncorhynchus nerka*) and implications for management. Canadian Journal of Fisheries and Aquatic Sciences 68: 718-737.
- Heuch, P.A., and Mo, T.A. 2001. A model of salmon louse production in Norway; effects of increasing salmon production and public management measures. Diseases of Aquatic Organisms 45: 145-152.

- Heuch, P.A., Bjorn, P.A., Finstad, B., Holst, J.C., Asplin, L., and Nilsen, F. 2005. A review of the Norwegian Action Plan against salmon lice on salmonids: the effects on wild salmonids. *Aquaculture* 246: 79–92.
- Hinch, S.G., Healey, M.C., Diewart, R.E., Thomson, K.A., Hourston, R., Henderson, M.A., and Juanes, F. 1995. Potential effects of climate change on marine growth and survival of Fraser River sockeye salmon. *Canadian Journal of Fisheries and Aquatic Sciences* 52: 2651-2659
- Hinch, S.G., and Gardner, J. 2009. Proceedings of the conference on early migration and premature mortality in Fraser River late-run sockeye salmon. Forest Sciences Centre, the University of British Columbia, 16–18 June 2008. Pacific Fisheries Resource Conservation Council, Vancouver, B.C. Available: [http://www.psc.org/pubs/LateRun/R-69\\_LateRun-SockeyeConf\\_2009\\_final.pdf](http://www.psc.org/pubs/LateRun/R-69_LateRun-SockeyeConf_2009_final.pdf) via the internet. Accessed 26 August 2011.
- Hocking, M.D., and Reynolds, J.D. 2011. Impacts of salmon on riparian plant diversity. *Science* 331: 1609-1612.
- Holst, J.C., and McDonald, A. 2000. FISH\_LIFT: a device for sampling live fish with trawls. *Fisheries Research* 48: 87-91.
- Hutchinson, A.H., and Lucas, C.C. 1931. The epithalassa of the Strait of Georgia: salinity, temperature, pH and phytoplankton. *Canadian Journal of Research* 5: 231-284.
- International Union for the Conservation of Nature. 2008. Pacific sockeye salmon (*Oncorhynchus nerka*) added to IUCN red-list. Available: <http://www.iucnredlist.org/apps/redlist/details/135301/0> via the internet. Accessed 30 December 2010.
- Iversen, M., Finstad, B., McKinley, R.S., Eliassen, R.A., Carlsen, K.T., Evjen, T., 2005. Stress responses in Atlantic salmon (*Salmo salar* L.) smolts during commercial well boat transports, and effects on survival after transfer to sea. *Aquaculture* 243: 373–382.
- Ivlev, V.S. 1961. *Experimental Ecology of the Feeding of Fishes*. Yale University Press, New Haven, Connecticut. 302 p.
- Jarp, J., and Karlsen, E. 1997. Infectious salmon anaemia (ISA) risk factors in sea-cultured Atlantic salmon *Salmo salar* (L.). *Diseases of Aquatic Organisms* 28: 79-86.
- Jessup, D.A., Boyce, W.M., and Clarke, R.K. 1991. Diseases shared by wild, exotic and domestic sheep. In: Renecker LA, Hudson RJ, editors. *Wildlife production: conservation and sustainable development*. University of Alaska. pp. 438–445.
- Johannessen, S.C., and Macdonald, R.W. 2009. Effects of local and global change on an inland

- sea: the Strait of Georgia, British Columbia, Canada. *Climate Research* 40: 1-21.
- Johnson, S.C., and Albright, L.J. 1991. The developmental stages of *Lepeophtheirus salmonis* (Kroyer, 1837) (Copepoda: Caligidae). *Canadian Journal of Zoology* 69: 929-950.
- Jones, S.R.M., and Hargreaves, N.B. 2007. The abundance and distribution of *Lepeophtheirus salmonis* (Copepoda: Caligidae) on pink (*Oncorhynchus gorbuscha*) and chum (*O. keta*) salmon in coastal British Columbia. *Journal of Parasitology* 93: 1324-1331.
- Jones, S.R.M., and Hargreaves, N.B. 2009. Infection threshold to estimate *Lepeophtheirus salmonis*-associated mortality among juvenile pink salmon. *Diseases of Aquatic Organisms* 84: 131-137.
- Kabata, Z. 1972. Development stages of *Caligus clemensi* (Copepoda: Caligidae). *Journal of the Research Board of Canada* 29: 1571-1593.
- Kleinbaum, D.G., Kupper, L.L., Nizam, A., and Muller, K.E. 2008. *Applied regression analysis and other multivariable methods*, fourth edition. Belmont, CA. pp. 287.
- Korman, J. 2011. Summary of information for evaluating impacts of salmon farms on survival of Fraser River sockeye salmon. Cohen Commission Technical Report 5A. Vancouver, BC. 65 p.
- Krkosek, M. 2010. Sea lice and salmon in Pacific Canada: ecology and policy. *Frontiers in Ecology and the Environment* 8: 201-209.
- Krkosek, M., Lewis, M.A., and Volpe, J.P. 2005a. Transmission dynamics of parasitic sea lice from farm to wild salmon. *Proceedings of the Royal Society of London B* 272: 689-696.
- Krkosek, M., Morton, A., and Volpe, J.P. 2005b. Nonlethal assessment of juvenile pink and chum salmon for parasitic sea lice infections and fish health. *Transactions of the American Fisheries Society* 134: 711-716.
- Krkosek, M., Lewis, M.A., Morton, A., Frazer, L.N., and Volpe, J.P. 2006. Epizootics of wild fish induced by fish farm. *Proceedings of the National Academy of Science in the U.S.A.* 103: 15506-15510.
- Krkosek, M., Ford, J.S., Morton, A., Lele, S., Myers, R.A., and Lewis, M.A. 2007a. Declining wild salmon populations in relation to parasites from farm salmon. *Science* 318: 1772-1775.
- Krkosek, M., Gottesfeld, A., Proctor, B., Rolston, D., Carr-Harris, C., and Lewis, M. 2007b. Effects of host migration, diversity and aquaculture on sea lice threats to Pacific salmon

- populations. Proceedings of the Royal Society of London B 274: 3141–3149.
- Krkosek, M., Morton, A., Volpe, J.P., and Lewis, M.A. 2009. Sea lice and salmon population dynamics: effects of exposure time for migratory fish. Proceedings of the Royal Society of London B 276: 2819-2828.
- Krkosek, M., and Hilborn, R. 2011. Sea lice (*Lepeophtheirus salmonis*) infestations and the productivity of pink salmon (*Oncorhynchus gorbuscha*) in the Broughton Archipelago, British Columbia, Canada. Canadian Journal of Fisheries and Aquatic Sciences 68: 17-29.
- Krkosek, M., Connors, B.M., Morton, A., Lewis, M.A., Dill, L.M., and Hilborn, R. 2011a. Effects of parasites from salmon farms on productivity of wild salmon. Proceedings of the National Academy of Science in the U.S.A. *In press*.
- Krkosek, M., Connors, B.M., Ford, H., Peacock, S., Mages, P., Ford, J.S., Morton, A., Volpe, J.P., Hilborn, R., Dill, L.M., and Lewis, M.A. 2011b. Fish farms, parasites, and predators: implications for salmon population dynamics. Ecological Applications 21: 897-914.
- Landingham, J.H., Sturdevant, M.V., and Brodeur, R.D. 1996. Feeding habits of juvenile Pacific salmon in marine waters of southeastern Alaska and northern British Columbia. Fishery Bulletin 96: 285-302.
- Legare, J.E.H. 1957. The Qualitative and quantitative distribution of plankton in the Strait of Georgia in relation to certain oceanographic factors. Journal of the Fisheries Research Board of Canada 14: 521-552.
- Legislative Assembly of British Columbia (LABC). 2007. Special committee on sustainable aquaculture: final report volume one. Available: [http://www.leg.bc.ca/CMT/38thparl/\\_\\_\\_\\_session-3/aquaculture/reports/Rpt-AQUACULTURE-38-3-Volume1-2007-MAY-16.pdf](http://www.leg.bc.ca/CMT/38thparl/____session-3/aquaculture/reports/Rpt-AQUACULTURE-38-3-Volume1-2007-MAY-16.pdf) via the internet. Accessed 28 January 2011.
- Macdonald, D.W., and Laurenson, M.K. 2006. Infectious disease: inextricable linkages between human and ecosystem health. Biological Conservation 131: 143–150.
- Mackas, D.L., Batten, S., and Trudel, M. 2007. Effects on zooplankton of a warmer ocean: recent evidence from the Northeast Pacific. Progress in Oceanography 75: 223–252.
- MacKenzie, K., Longshaw, M., Begg, G.S., and McVicar, A.H. 1998. Sea lice (Copepoda: *Caligidae*) on wild sea trout (*Salmo trutta L.*) in Scotland. ICES Journal of Marine Sciences 55: 151-162.
- Margolis, L., Esch, G.W., Holmes, J.C., Kuris, A.M., and Schad, G.A. 1982. The use of ecological terms in parasitology (report of an ad hoc committee of the American Society of

- parasitologists). *Journal of Parasitology* 68: 131–133.
- Marine Harvest Corporate. 2008. Sustainability report. Oslo, Norway.
- Marty, G.D., Saksida, S.M., and Quinn, T.J. 2010. Relationship of farm salmon, sea lice, and wild salmon populations. *Proceedings of the National Academy of Science of the U.S.A.* doi: 10.1073/pnas.1009573108.
- McCallum, H., and Dobson, A.P. 1995. Detecting disease and parasite threats to endangered species and ecosystems. *Trends in Ecology and Evolution* 10: 190-194.
- McVicar, A.H. 1997. Disease and parasite implications of the coexistence of wild and cultured Atlantic salmon populations. *ICES Journal of Marine Sciences* 54: 1093-1103.
- McVicar, A.H. 2004. Management actions in relation to the controversy about salmon lice infections in fish farms as a hazard to wild salmonid populations. *Aquaculture Research* 35: 751-758.
- Morton, A., and Williams, R. 2003. First report of a sea louse, *Lepeophtheirus salmonis*, infestation on juvenile pink salmon *Oncorhynchus gorbuscha* in nearshore habitat. *Canadian Field-Naturalist* 117: 634-641.
- Morton, A., Routledge, R., Peet, C., and Ladwig, A. 2004. Sea lice (*Lepeophtheirus salmonis*) infection rates on juvenile pink (*Oncorhynchus gorbuscha*) and chum salmon (*Oncorhynchus keta*) in the nearshore marine environment of British Columbia, Canada. *Canadian Journal of Fisheries and Aquatic Sciences* 61: 147–157.
- Morton, A., and Routledge, R. 2005. Mortality rates for juvenile pink *Oncorhynchus gorbuscha* and chum *O. keta* salmon infested with sea lice *Lepeophtheirus salmonis* in the Broughton Archipelago. *Alaska Fisheries Research Bulletin* 11: 146-152.
- Morton, A., Routledge, R.D., and Williams, R. 2005. Temporal patterns of sea louse infestation on wild Pacific salmon in relation to the fallowing of Atlantic salmon farms. *North American Journal of Fisheries Management* 25: 811-821.
- Morton, A., Routledge, R., and Krkosek, M. 2008. Sea lice infestation of wild juvenile salmon and herring associated with fish farms off the east-central coast of Vancouver Island, British Columbia. *North American Journal of Fisheries Management* 28: 523-532.
- Murray, A.G. 2008. Using simple models to review the application and implications of different approaches used to simulate transmission of pathogens among aquatic animals. *Preventive Veterinary Medicine* 88: 167–177.

- Murray, A.G., and Peeler, E.J. 2005. A framework for understanding the potential for emerging diseases in aquaculture. *Preventive Veterinary Medicine* 67: 223–235.
- Murray, A.G., Munro, L.A., Wallace, I.S., Berx, B., Pendrey, D., Fraser, D., and Raynard, R.S. 2010. Epidemiological investigation into the re-emergence and control of an outbreak of infectious salmon anaemia in the Shetland Islands, Scotland. *Diseases of Aquatic Organisms* 91: 189-200.
- Naiman, R.J., Bilby, R.E., Schindler, D.E., and Helfield, J.M. 2002. Pacific salmon, nutrients, and the dynamics of freshwater and riparian ecosystems. *Ecosystems* 5: 399–417.
- Naylor, R.L., Eagle, J., and Smith, W.L. 2003. Salmon aquaculture in the Pacific Northwest: a global industry. *Environment* 45: 19-39.
- Neilson, J.D., Perry, R.I., Scott, J.S., Valerio, P. 1987. Interactions of caligid ectoparasites and juvenile gadids on Georges Bank. *Marine Ecology Progress Series* 39: 221-232.
- Nese, L., and Enger, R. 1993. Isolation of *Aeromonas salmonicida* from salmon lice, *Lepeophtheirus salmonis* and marine plankton. *Diseases of Aquatic Organisms* 16: 79–81.
- Northcote, T.G., and Atagi, D.Y. 1997. Pacific salmon abundance trends in the Fraser River watershed compared with other British Columbia systems. In Stouder, D.J., Bisson, P.A., and Naiman, R.J., editors. *Pacific salmon and their ecosystems: status and future options*. Chapman and Hall, New York, New York, USA. pp. 199-219.
- Nowak, B.F. 2007. Parasitic diseases in marine cage culture – An example of experimental evolution of parasites? *International Journal of Parasitology* 37: 581-588.
- Nylund, A., Hovland, T., Hodneland, K., Nilsen, F., and Løvik, P. 1994. Mechanisms for transmission of infectious salmon anemia (ISA). *Diseases of Aquatic Organisms* 19: 95–100.
- Orr, C. 2007. Estimated sea louse egg production from marine Harvest Canada farmed Atlantic salmon in the Broughton Archipelago, British Columbia, 2003-2004. *North American Journal of Fisheries Management* 27: 187-197.
- Otterstatter, M.C., and Thomson, J.D. 2008. Does pathogen spillover from commercially reared bumble bees threaten wild pollinators? *Public Library of Science ONE* 3: e2771.
- Parker, R.R., and Margolis, L. 1964. A new species of parasitic copepod, *Caligus clemensi* sp. Nov. (Caligoida: Caligidae), from pelagic fishes in the coastal waters of British Columbia. *Journal of the Fisheries Research Board of Canada* 21: 873-889.

- Parsons, T.R., Stronach, J., Borstad, G.A., Louttit, G., and Perry, R.I. 1981. Biological fronts in the Strait of Georgia, British Columbia, and Their Relation to Recent Measurements of Primary Productivity. *Marine Ecology Progress Series* 6: 237-242.
- Pearcy, W.G. 1992. *Ocean ecology of North Pacific salmonids*. Washington Sea Grant, Seattle, Washington. 170 p.
- Pella, J., and Masuda, M. 2001. Bayesian methods for analysis of stock mixtures from genetic characters. *Fishery Bulletin* 99: 151-167.
- Penston, M.J., Millar, C.P., and Davies, I.M. 2008. Reduced *Lepeophtheirus salmonis* larval abundance in a sea loch on the west coast of Scotland between 2002 and 2006. *Disease of Aquatic Organisms* 81: 109-117.
- Perry, R.I., Hargreaves, B., Waddell, B.J., and Mackas, D.L. 1996. Spatial variations in feeding and condition of juvenile pink and chum salmon off Vancouver Island, British Columbia. *Fisheries Oceanography* 5: 73-88.
- Peterman, R.M. 1982. Nonlinear relation between smolts and adults in Babine Lake sockeye salmon (*Oncorhynchus nerka*) and implications for other salmon populations. *Canadian Journal of Fisheries and Aquatic Sciences* 39: 904-913.
- Peterman, R.M., Marmorek, D., Beckman, B., Bradford, M., Mantua, N., Riddell, B.E., Scheuerell, M., Staley, M., Wieckowski, K., Winton, J.R., and Wood, C.C. 2010. Synthesis of evidence from a workshop on the decline of Fraser River sockeye. Available: <http://www.psc.org/pubs/FraserSockeyeDeclineWorkshopIntro.pdf> via the internet. Accessed 30 December 2010.
- Peterman R.M., and Dorner, B. 2011. Fraser River sockeye production dynamics. Cohen Commission Technical Report 10. Vancouver, BC. 134 p.
- PFRCC (Pacific Fisheries Resource Conservation Council). 2002. Advisory: the protection of Broughton Archipelago pink salmon stocks. Report to the Minister of Fisheries and Oceans and B.C. Minister of Agriculture, Food, and Fisheries. Vancouver, BC.
- Pike, A.W., and Wadsworth, S.L. 2000. Sea lice on salmonids: their biology and control. *Advances in Parasitology* 44: 233-337.
- Power, A.G., and Mitchell, C.E. 2004. Pathogen spillover in disease epidemics. *American Naturalist* 164: S79-S89.
- Price, M.H.H., Darimont, C.T., Temple, N.F., and MacDuffee, S.M. 2008. Ghost runs: management and status assessment for Pacific salmon (*Oncorhynchus* spp.) returning to

- British Columbia's central and north coasts. *Canadian Journal of Fisheries and Aquatic Sciences* 65: 2712-2718.
- Price, M.H.H., Morton, A., and Reynolds, J.D. 2010. Evidence of farm-induced parasite infestations on wild juvenile salmon in multiple regions of coastal British Columbia, Canada. *Canadian Journal of Fisheries and Aquatic Sciences* 67: 1925-1932.
- Price, M.H.H., Proboszcz, S.L., Routledge, R.D., Gottesfeld, A.S., Orr, C., and Reynolds, J.D. 2011. Sea louse infection of juvenile sockeye salmon in relation to marine salmon farms on Canada's west coast. *Public Library of Science ONE* 6: e16851.
- Quinn, T.P. 2005. *The behavior and ecology of Pacific salmon and trout*. Vancouver: UBC Press.
- Ricker, W.E. 1976. Review of the rate of growth and mortality of Pacific salmon in salt water, and non-catch mortality caused by fishing. *Journal of the Fisheries Research Board of Canada* 33: 1483-1524.
- Ross, P.S. 2006. Fireproof killer whales (*Orcinus orca*): flame-retardant chemicals and the conservation imperative in the charismatic icon of British Columbia, Canada. *Canadian Journal of Fisheries and Aquatic Sciences* 63: 224-234.
- Saksida, S.M. 2006. Infectious haematopoietic necrosis epidemic (2001 to 2003) in farmed Atlantic salmon *Salmo salar* (L.) in British Columbia. *Diseases of Aquatic Organisms* 72: 213-223.
- Schabetsberger, R., Morgan, C.A., Brodeur, R.D., Potts, C.L., Peterson, W.T., and Emmett, R.L. 2003. Prey selectivity and diel feeding chronology of juvenile Chinook (*Oncorhynchus tshawytscha*) and coho (*O. kisutch*) salmon in the Columbia River plume. *Fisheries Oceanography* 12: 523-540.
- Slaney, T.L., Hyatt, K.D., Northcote, T.G., and Fielden, R.J. 1996. Status of anadromous salmon and trout in British Columbia and Yukon. *Fisheries* 21: 20-38.
- SPSS. 2007. SPSS 16.0 for Mac. SPSS Inc., Chicago, Ill.
- SPSS. 2010. SPSS 18.0 for Mac. SPSS Inc., Chicago, Ill.
- Statement from Think Tank of Scientists. 2009. Adapting to change: managing Fraser sockeye in the face of declining productivity and increasing uncertainty. Available: <http://www.fish.bc.ca/files/FraserSockeyeThinkTankStatement.pdf> via the internet. Accessed 28 January 2011.

- Stockner, J.G., Cliff, D.D., and Shortreed, K.R.S. 1979. Phytoplankton ecology of the Strait of Georgia, British Columbia. *Journal of the Fisheries Research Board of Canada* 36: 657-666.
- Thirgood, S. 2009. New perspectives on managing wildlife diseases. *Journal of Applied Ecology* 46: 454-456.
- Thomson, R.E. 1981. Oceanography of the British Columbia coast. *Canadian Special Publication of Fisheries and Aquatic Sciences* 56.
- Totland, G.K., Hjeltnes, B.K., and Flood, P.R. 1996. Transmission of infectious salmon anaemia (ISA) through natural secretions and excretions from infected smolts of Atlantic salmon *Salmo salar* during their presymptomatic phase. *Diseases of Aquatic Organisms* 26: 25-31.
- Tully, O., Gargan, P. Poole, W.R., and Whelan, K.F. 1999. Spatial and temporal variation in the infestation of sea trout *Salmo trutta L.* by the caligid copepod *Lepeophtheirus salmonis* (Kroyer) in relation to sources of infection in Ireland. *Parasitology* 119: 41-51.
- Vagsholm, I., Djupvik, H.O., Willumsen, F.V., Tveit, A.N., and Tangen, K. 1994. Infectious salmon anaemia (ISA) epidemiology in Norway. *Preventive Veterinary Medicine* 19: 277-290.
- Webster, S.J., Dill, L.M., and Butterworth, K. 2007. The effect of sea lice infestation on the salinity preference and energetic expenditure of juvenile pink salmon (*Oncorhynchus gorbuscha*). *Canadian Journal of Fisheries and Aquatic Sciences* 64: 672-680.
- Welch, D.W., Melnychuk, M.C., Rechisky, E.R., Porter, A.D., Jacobs, M.C. Ladouceur, A., McKinley, R.S., and Jackson, G.D. 2009. Freshwater and marine migration and survival of endangered Cultus Lake sockeye salmon (*Oncorhynchus nerka*) smolts using POST, a large-scale acoustic telemetry array. *Canadian Journal of Fisheries and Aquatic Sciences* 66: 736-750.
- Welch, D.W., Melnychuk, M.C., Payne, J.C., Rechisky, E.R., Porter, A.D., Jackson, G.D., Ward, B.R., Vincent, S.P., Wood, C.C., and Semmens, J. 2011. In situ measurement of coastal ocean movements and survival of juvenile Pacific salmon. *Proceedings of the National Academy of Science of the U.S.A.* 108: 8708-8713.
- Wikel, S.K., Ramachandra, R.N., and Bergman, D.K. 1994. Tick-induced modulation of the host immune response. *International Journal for Parasitology* 24: 59-66.
- Withler, R.E., Le, K.D., Nelson, R.J., Miller, K.M., and Beacham, T.D. 2000. Intact genetic structure and high levels of genetic diversity in bottlenecked sockeye salmon, *Oncorhynchus nerka*, populations of the Fraser River, British Columbia, Canada. *Canadian*

Journal of Fisheries and Aquatic Sciences 57: 1985–1998.