Estimating the dispersal of *Lepeophtheirus salmonis* using routine counts among aquaculture sites in southwest New Brunswick

A Thesis
Submitted to the Graduate Faculty
in Partial Fulfillment of the Requirements
for the Degree of

Doctor of Philosophy

Department of Health Management
Faculty of Veterinary Medicine
University of Prince Edward Island

Marianne Parent
Charlottetown, PEI
January 17, 2024

© 2024, M.I. Parent

THESIS/DISSERTATION NON-EXCLUSIVE LICENSE

Family Name: Parent	Given Name, Middle Name (if applicable): Marianne I.
Full Name of University: University of Prince Edward Island	
Faculty, Department, School: Faculty of Veterinary Medicine, Department of Health Management, Atlantic Veterinary College	
Degree for which thesis/dissertation was presented: Doctor of Philosophy	Date Degree Awarded: February 6, 2024
Thesis/dissertation Title: Estimating the dispersal of *Lepeophtheirus salmonis* using routine counts among aquaculture sites in southwest New Brunswick	
Date of Birth. It is **optional** to supply your date of birth. If you choose to do so please note that the information will be included in the bibliographic record for your thesis/dissertation.	

In consideration of my University making my thesis/dissertation available to interested persons, I,
Marianne Parent

hereby grant a non-exclusive, for the full term of copyright protection, license to my University,
The University of Prince Edward Island:

(a) to archive, preserve, produce, reproduce, publish, communicate, convert into any format, and to make available in print or online by telecommunication to the public for non-commercial purposes;
(b) to sub-license to Library and Archives Canada any of the acts mentioned in paragraph (a).

I undertake to submit my thesis/dissertation, through my University, to Library and Archives Canada. Any abstract submitted with the thesis/dissertation will be considered to form part of the thesis/dissertation.

I represent that my thesis/dissertation is my original work, does not infringe any rights of others, including privacy rights, and that I have the right to make the grant conferred by this non-exclusive license.

If third party copyrighted material was included in my thesis/dissertation for which, under the terms of the *Copyright Act*, written permission from the copyright owners is required I have obtained such permission from the copyright owners to do the acts mentioned in paragraph (a) above for the full term of copyright protection

I retain copyright ownership and moral rights in my thesis/dissertation, and may deal with the copyright in my thesis/dissertation, in any way consistent with rights granted by me to my University in this non-exclusive license.

I further promise to inform any person to whom I may hereafter assign or license my copyright in my thesis/dissertation of the rights granted by me to my University in this non-exclusive license.

Signature	**Date** February 7, 2024

University of Prince Edward Island
Faculty of Veterinary Medicine
Charlottetown

Certification of Thesis Work

We, the undersigned, certify that **Marianne Parent**, candidate for the degree of Doctor of Philosophy, has presented her thesis with the following title:

Estimating the dispersal of *Lepeophtheirus salmonis* using routine counts among aquaculture sites in southwest New Brunswick

that the thesis is acceptable in form and content, and that a satisfactory knowledge of the field covered by the thesis was demonstrated by the candidate through an oral examination held on January 17, 2024.

Approval verified by the Examination Committee Chair, Dr. Sonja Saksida, for Examination Committee members:

Dr. Alexander (Sandy) Murray (External), Dr. Krishna Thakur, Dr. Mark Fast, Dr. K. Larry Hammell

[redacted]

Dr. Sonja Saksida

Feb 6, 2024

Date

Abstract

Sea lice *Lepeophtheirus salmonis* requires intense management practices for the Atlantic salmon aquaculture industry in the Bay of Fundy, New Brunswick (NB). This dissertation aims to improve the knowledge of the dispersal of *L. salmonis* (commonly referred to as salmon lice) in the study area, which may aid in planning decisions for sea lice mitigation strategies using monitoring counts from the Fish-iTrends© database. The biology, history, and trends of sea lice counts and treatments were described to identify possible confounding factors that may be important during the model-building process. The impact of the internal and external infestation pressures on the abundance of adult female (AF) sea lice for the Bay of Fundy, NB, for 2009-2018 was estimated using a multivariable linear mixed model. This model was used to compare different connectivity measures of dispersal distance and assess their impact when calculating external infestation pressure on modelling the AF sea lice abundance. A multivariable autoregressive model for the abundance of AF sea lice was built for 2016-2021 and employed for short-term predictions at the end of the summer of 2020. The multivariable linear mixed model and the multivariable autoregressive model were compared using an overlapping subset of the datasets (2016-2018). The estimations of infestation pressures, dispersal distance and other predictors, and results from the generation of novel connectivity measures may have implications for the management of the NB aquaculture industry. The results suggest that the current area-wide management structure may need to be revised, and

synchronizing mitigation strategies within and among sites could reduce infestation pressure.

Acknowledgments

I sincerely thank Henrik Stryhn and Raphaël Vanderstichel, my co-supervisors, for their guidance in this project. They have provided support during each aspect of the project, from data management to manuscript writing. Henrik and Raph have supported me during personal events during the period of my thesis. Their dedication to my success is a large part of how I am completing this dissertation. Thank you to Larry Hammell, Mark Fast and Jon Grant for their role on the supervisory committee and for the time and effort they applied to edit my manuscripts. Thank you, Larry, for being the voice of industry members in my work. Larry has been a crucial to the development and maintenance of the Fish-iTrends© database, and without his work, mine would not have been possible. I want to acknowledge the financial support of the Ocean Frontiers Institute (OFI), Long Island University and Atlantic Fisheries Fund for this project. I want to thank the New Brunswick Atlantic salmon producers for reporting data to the Fish-iTrends© database. Thank you to the Atlantic Veterinary College Centre for Aquatic Health Sciences and M. Sanford for the management of the Fish-iTrends© database. The Atlantic Canada Fish Farmers Association, Newfoundland Aquaculture Industry Association, and the Nova Scotia Aquaculture Association fund the Fish-iTrends© database.

Dedication

I want to dedicate this thesis to my family. Patrick has supported me during all parts of this project. He did not hesitate to support me during the challenges of sickness, having children, a pandemic, and more. This would have taken much longer without his help on the sidelines. Thank you. To my mother, you supported my educational goals the whole way, even when I strongly doubted myself, thank you.

Table of Contents

Estimating the dispersal of *Lepeophtheirus salmonis* using routine counts among aquaculture sites in southwest New Brunswick 1
Abstract iv
Acknowledgments vi
Dedication vii
Table of Contents viii
List of Tables xiii
List of Figures xvi
List of Abbreviations xxi
Glossary xxii
1 Introduction 1
1.1 Salmonids 1
1.2 Salmon production 2
1.2.1 General 2
1.2.2 World 4
1.2.3 Canada 5
1.3 Sea lice 11
1.3.1 Species 11
1.3.2 Biology of sea lice 13
1.3.3 Pathology secondary to sea lice infestation 19
1.4 Other diseases of farmed salmon 22
1.5 Mitigation of sea lice 24
1.5.1 Management strategies 24
1.5.2 Estimated costs of sea lice management 38
1.5.3 Global and local trends 39
1.6 Monitoring and surveillance of sea lice 41
1.7 Connectivity and infestation pressure 43
1.7.1 General concepts of dispersal and connectivity 43
1.7.2 Dispersal distance of sea lice 48
1.7.3 Connectivity measures 58
1.7.4 Internal and external infestation pressure 60
1.8 Sea lice models of dispersal and infestation 67
1.8.1 Statistical vs. simulation models 67
1.8.2 Empirical vs. theoretical models 69

1.8.3 Models for prediction vs. risk factors 70
1.8.4 Model predictors: highlight on temperature 72
1.9 Thesis objectives 76
1.10 References 79
2 Atlantic salmon aquaculture industry in the Bay of Fundy, New Brunswick 108
2.1 Introduction 108
2.1.1 Bay of Fundy, New Brunswick coast 108
2.1.2 Aquaculture and sea lice in the Bay of Fundy, NB 116
2.1.3 Objective 123
2.2 Materials and methods 123
2.3 Results 126
2.3.1 Observations 126
2.3.2 Sea lice abundances 128
2.3.3 Sea lice treatments 131
2.3.4 Water temperature 133
2.4 Discussion 134
2.4.1 Sea lice counts and abundances 134
2.4.2 Sea lice abundances vary by location 138
2.4.3 Sea lice abundances vary seasonally 140
2.4.4 Treatments 142
2.5 Conclusions 144
2.6 References 145
3 Estimating the dispersal of *Lepeophtheirus salmonis* sea lice within and among Atlantic salmon sites of the Bay of Fundy, New Brunswick 154
3.1 Abstract 154
3.2 Introduction 155
3.3 Materials and methods 160
3.3.1 Study area and population 160
3.3.2 Selection and preparation of the outcome and abundances 161
3.3.3 Data structure 162
3.3.4 Defining internal and external infestation pressure 163
3.3.5 Baseline abundance of adult female sea lice 164
3.3.6 Topical and in-feed treatments 164

3.3.7 Sea surface temperature 165
3.3.8 Modelling 166
3.4 Results 168
3.4.1 Descriptive statistics of the observations modelled 168
3.4.2 Final model 176
3.4.3 Fixed effects 180
3.4.5 Random effects 184
3.5 Discussion 184
3.5.1 Contributions of internal and external infestation pressure 184
3.5.2 Treatments and temperature 188
3.5.3 Hierarchical levels 191
3.5.4 Study design and limitations 191
3.6 Conclusions 195
3.7 References 196
3.8 Supplemental information 204
3.8.1 Data 204
3.8.2 Generation of weights 205
3.8.3 Final model 209
3.8.4 Sensitivity analyses 210
3.8.5 References 221
4 Estimation and comparison of connectivity measures for the dispersal of *Lepeophtheirus salmonis* sea lice among Atlantic salmon farms in New Brunswick, Canada 222
4.1 Abstract 222
4.2 Introduction 223
4.3 Materials and methods 227
4.3.1 Study area and population 227
4.3.2 Connectivity measures 228
4.3.3 Model 233
4.4 Results 239
4.4.1 Comparison of connectivity measures 239
4.4.2 Model 241
4.4.3 Depth 244
4.5 Discussion 245

4.5.1 New connectivity measures 245
4.5.2 Connectivity measures 245
4.5.3 Impact on the model 246
4.5.4 Study design and limitations 254
4.6 Conclusions 257
4.7 References 258
4.8 Supplemental information 264
4.8.1 External infestation pressure 264
4.8.2 Euclidean distance for the comparison of various connectivity measures 274
4.8.3 References 276
5 Predicting the dispersal of *Lepeophtheirus salmonis* in the Bay of Fundy, New Brunswick 277
5.1 Abstract 277
5.2 Introduction 277
5.3 Materials and methods 281
5.3.1 Study area and data structure 281
5.3.2 Preparation of the outcome 282
5.3.3 Calculation of internal and external infestation pressures 283
5.3.4 Topical and in-feed treatments 284
5.3.5 Time at sea 286
5.3.6 Modelling 292
5.4 Results 295
5.4.1 Descriptive statistics of the observations modelled 295
5.4.2 Final model 297
5.5 Discussion 310
5.6 Conclusions 322
5.7 References 324
5.8 Supplemental information 329
5.8.1 Temperature 329
5.8.1 Treatments 329
5.8.2 Internal and external infestation pressure 330
5.8.3 Final model 333
5.8.4 In-sample short term prediction 334

5.8.5 Sensitivity analyses 335
5.8.6 References 345
6 Discussion 346
6.1 Summary 346
6.2 Comparison of models 348
6.2.1 Objective 348
6.2.2 Materials and methods 349
6.2.3 Results 350
6.3 Internal and external infestation pressures 357
6.4 Connectivity and dispersal distance 364
6.4.1 Connectivity measures 364
6.4.2 Dispersal distance 367
6.5 Treatments 369
6.6 Temperature and salinity 373
6.7 Time at sea 374
6.8 Prediction 375
6.9 Data and future studies using the Fish-iTrends© 376
6.10 Recommendations to the New Brunswick Atlantic salmon aquaculture stakeholders 379
6.11 Conclusions 382
6.12 References 384

List of Tables

Table 1.1. Summary of treatments for *Lepeophtheirus salmonis* as available in New Brunswick, Canada. Lufenuron is applied at the hatchery, while all others are applied at the cage-level.

Table 2.1. Published (Chang et al., 2007; NB Department of Energy and Resource Development/Agriculture, 2017) stocking years for the culture of Atlantic salmon within Bay Management Areas (BMA). BMA 4 2006-2010 (Jones et al., 2012); same location, BMA 3C 2017-present.

Table 2.2. Summary of counts of sea lice between July 2009 and November 2018 for the Bay of Fundy, NB. PAAM: pre-adult and adult male, AF: adult female, 25 and 75%; 25 and 75th percentiles. n = 63,432.

Table 3.1. Number of observations and segments per hierarchical level. n = 11,658, n/a = not applicable, † = nested groups of the final multivariable linear mixed model for the abundance of adult female *Lepeophtheirus salmonis* with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km.

Table 3.2. Number of observations (number of segments) categorized by the Bay Management Areas (BMA) and three temperature profiles (temp. cat.).

Table 3.3. Descriptive statistics for the variables potentially predicting the outcome variable of the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice. Results of the univariable linear mixed model are summarized by the Akaike's information criterion (AIC; null model AIC = 32,486). SD: standard deviation; IIP: internal infestation pressure; PAAM: pre-adult and adult male sea lice; EIP: external infestation pressure; n = 11,658.

Table 3.4. Multivariable linear mixed model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice; n = 11,658.

Table 3.5. Estimated coefficient change (difference/average against the final model (n = 11,658) with a cutoff of |25 %|): - no change, ↑ increase, and ↓ decrease for the multivariable linear mixed model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. n_{obs}: number of observations, n_{seg}: number of segments, Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice.

Table 3.6. Estimated coefficients (coef.) and standard errors (SE) for the final multivariable linear mixed model for the abundance of adult female *Lepeophtheirus salmonis* and adjusted models with the omitted predictor $baseline_{AF}$: model 2 with final model sample and model 3 without constraint of final model sample. Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice. Final and model 2: n_{obs} = 11,658, n_{seg} = 2,398. Model 3: n_{obs} =17,762, n_{seg} = 3,723.

Table 4.1. Quantified distances among scaled connectivity measures using Euclidean distance. Connectivity measures were seaway distance at 25 m (SWD_{25}) and 100 m resolutions (SWD_{100}), percent surface area of water along seaway path with a square window area of 250 m (SA_{250}), 500 m (SA_{500}), and 1,000 m (SA_{1000}), topographical distance (TD) with adjustment of the cost of movement along an incline by factors of 10 (TD_{10}), 100 (TD_{100}), 500 (TD_{500}), and 1,000 (TD_{1000}), and volume of water along seaway path (VW) with adjustment of the number of cells included in the estimation to 250 m (VW_{250}), 500 m (VW500), and 1,000 m (VW_{1000}).

Table 5.1. Descriptive statistics for the variables potentially predicting the outcome variable of the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice. Results of the univariable mixed autoregressive model are summarized by the Akaike's information criterion (AIC; null model AIC = 6445). SD: standard deviation; IIP: internal infestation pressure; PAAM: pre-adult and adult male sea lice; EIP: external infestation pressure; n = 3,137 site-weeks.

Table 5.2. Multivariable mixed autoregressive model (AR[2]) for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice with an external infestation pressure weighted with a kernel density estimate of seaway distance. Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice; n = 3,137 weeks.

Table 5.3. Temperature profiles for the AR(2) correlation structure to the errors of the multivariable autoregressive linear mixed effects model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice; n = 3,137 site-weeks.

Table 5.4. Mean (standard deviation) of the observed and predicted adult female (AF) *Lepeophtheirus salmonis* sea lice and the normalized prediction error over 8 weeks in the late summer of 2020 for 14 site-cycles using two methods of prediction: 1) predictions calculated using residuals that were the difference between the observed and predicted, and 2) predictions calculated using residuals set to 0.

Table 5.5. Estimated coefficient change (difference/average against the final model (n = 3,137 site-weeks, AIC = 6080) with a cutoff of |25 %|): - no change, ↑ increase, and ↓ decrease for the multivariable mixed autoregressive model for

the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice. Temperature: sea surface temperature; IIPAF: internal infestation pressure of AF; IIPPAAM: internal infestation pressure of pre-adult and adult male; EIPAF: external infestation pressure of AF; TAS: time at sea in weeks.

Table 6.1. Number of observations (counts and weeks) per hierarchical level for datasets from Chapter 2-4 (1) and Chapter 5 (2a and b) for June 2016-November 2018. Dataset 2b was restricted to observations (weeks) that occur after the first topical treatments were applied during the production cycle. n/a not applicable, † = nested groups in multilevel mixed effects linear regression for the abundance of adult female (AF) *Lepeophtheirus salmonis*; ‡ = nested groups in the multivariable autoregressive linear mixed effects model for the abundance of AF *L. salmonis*.

Table 6.2. Estimate coefficients (coef.) and standard errors (SE) for the final multivariable linear mixed effects regression model (dataset 1; model 1) and the final multivariable autoregressive linear mixed effects model (dataset 2a and b; models 2 and 3, respectively). The linear mixed effects model and autoregressive linear mixed effects model were applied to a limited list of predictors (datasets 1 and 2a; models 4 and 5, respectively): internal infestation pressure (IIP) of adult female (AF) *Lepeophtheirus salmonis* (IIP_{AF}), IIP of pre-adult and adult male (IIP_{PAAM}), external infestation pressure of AF (EIP_{AF}), and sea surface temperature. The Bay of Fundy, New Brunswick, Canada, study area (A) and Bay Management Areas (BMA) in the southwest coast and Passamaquoddy Bay (B) and Grand Manan Island (C). The locations of the BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (NB Department of Energy and Resource Development/Agriculture, and Fisheries, 2017). base = baseline.

List of Figures

Figure 1.1. Reproduced figure from Chapter 2. The Bay of Fundy, New Brunswick, Canada, study area (A) and Bay Management Areas (BMA) in the southwest coast and Passamaquoddy Bay (B) and Grand Manan Island (C). The locations of the BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (NB Department of Energy and Resource Development/Agriculture, and Fisheries, 2017).

Figure 1.2. Life cycle stages of development of *Lepeophtheirus salmonis* with the location of this ectoparasite in the water column.

Figure 1.3. Diagram of the concepts of metapopulation and subpopulations of Atlantic salmon farm sites in the Bay of Fundy, New Brunswick, Canada, that aid in the understanding of the concept of connectivity among farms by the parasite *Lepeophtheirus salmonis*.

Figure 1.4. Illustration of a timeline for the development of *Lepeophtheirus salmonis*. The treatment reset for lice reflects the assumed decrease in sea lice abundance secondary to the application of a treatment.

Figure 1.5. Direct comparison of Fisheries and Oceans Canada and UKMO remote-sensing sea surface temperatures for 29 matched sites (Atlantic salmon aquaculture site and DFO Station) in the Passamaquoddy Bay, New Brunswick, Canada for the period July 2009 to November 2018.

Figure 2.1. The Bay of Fundy, New Brunswick, Canada, study area (A) and Bay Management Areas (BMA) in the southwest coast and Passamaquoddy Bay (B) and Grand Manan Island (C). The locations of the BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (NB Department of Energy and Resource Development/Agriculture, and Fisheries, 2017).

Figure 2.2. Boxplot of sea lice abundances ([lice+1]/fish) in the Bay of Fundy, New Brunswick, for July 2009 to November 2018 per season (A) and Bay Management Areas (B). Seasons were defined using the equinox and solstices. n = 63,214.

Figure 2.3. Mean sea lice abundances ([lice+1]/fish) per month +/- standard error for *Caligus elongatus* and *Lepeophtheirus salmonis* for the Bay of Fundy, New Brunswick, for July 2009 to November 2018 (A). *L. salmonis* life stages were counted as chalimus, pre-adult and adult male (PAAM), and adult female (AF). Mean number of topical and oral treatments per cage for the same time period (B). Oral treatments are reported in cage-days of treatment.

Figure 2.4. Total number of topical treatments by method of application applied annually in the Bay of Fundy, New Brunswick. DS: deltamethrin skirt, AT:

azamethiphos tarpaulin, AS: azamethiphos skirt, AW: azamethiphos wellboat, A.: azamethiphos missing method of application, WW: thermal, H: Hydrolicer®, HT: hydrogen peroxide tarpaulin, HW: hydrogen peroxide wellboat, and H.: hydrogen peroxide missing method of application. n_{total} = 6,296

Figure 2.5. Average daily sea surface temperature (extracted from the UKMO OSTIA SST remote sensing product [UK Met Office, 2005]) per aquaculture site for the duration of the study (A) and 2017 excerpt (B). Horizontal line represents the mean temperature for the plot. Vertical lines (B) are the seasonal equinox and solstices. n = 11,157, n_{2017} = 1,332.

Figure 3.1. The Bay of Fundy, New Brunswick, Canada, study area demonstrating Bay Management Areas (BMA) and Atlantic salmon aquaculture sites. Sites included (black circle) and excluded (grey circle) from the final multivariable linear mixed model for the abundance of adult female *Lepeophtheirus salmonis*. Abundances of sea lice were not available from sites from Maine, USA (white circle). The locations of sites and BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (Department of Energy and Resource Development/Agriculture, and Fisheries, 2017).

Figure 3.2. Boxplots of the mean normalized count of adult female (AF) *Lepeophtheirus salmonis* sea lice after treatment (AF count divided by the number of fish counted and offset by 1) included in the final multivariable linear mixed model with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. Counts were categorized by treatment (D: deltamethrin, AT: azamethiphos tarpaulin, AS: azamethiphos skirt, AW: azamethiphos wellboat, WW: warm water, Hy: Hydrolicer®, and HP: hydrogen peroxide) and temperature profiles (L: 3-7 °C, M: 8-11 °C, and H: 12-16 °C). n = 11,658.

Figure 3.3. Estimated exponentiated coefficient and 95 % confidence interval (CI) for the external infestation pressure (EIP) predictor weighted with a Gaussian kernel density estimate (KDE) and the Akaike's information criterion (AIC) reported from the multivariable linear mixed model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice where the bandwidth of the KDE was increased from 5 to 40 km. Scale is the mean normalized count of AF sea lice after treatment (AF count divided by the number of fish counted and offset by 1).

Figure 3.4. Estimated mean normalized count of adult female (AF) *Lepeophtheirus salmonis* sea lice (AF count divided by the number of fish counted and offset by 1) after treatments applied in the Bay Management Area (BMA) 1 against sea surface temperature at median values of the continuous predictors of the final multivariable linear mixed model with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. D: deltamethrin, AT: azamethiphos tarpaulin, AS: azamethiphos skirt, AW:

azamethiphos wellboat, WW: warm water, and HP: hydrogen peroxide. The Hydrolicer® was not applied in BMA 1 between 2009 and 2018.

Figure 3.5. Organization of ‘segments’ of sea lice counts with time and topical treatments.

Figure 4.1. Depictions of the methods for the generation of connectivity matrices used in the production of Gaussian kernel density estimate weights for the calculation of external infestation pressure. A: seaway distance; B: percent surface area of water along seaway path with a square window area; C: topographical distance; D: volume of water along seaway path. Additional matrices were produced by adjusting the resolution (A), increasing the covered area to 250 m, 500m, and 1,000 m (B and D), and increasing the cost of movement along an incline by multiplying by a factor of 10, 100, 500, and 1,000 (C).

Figure 4.2. Scatter plot matrix of selected connectivity measures directly (above diagonal) and external infestation pressure (EIP_{AF}) predictors as calculated from the connectivity matrices (below diagonal). The values in the top left corner of the plots are the Pearson’s correlation (ρ) for pairs of connectivity measures and concordance correlation coefficients (ρ_c) for pairs of EIP_{AF}. SWD_{100}: seaway distance with 100 m resolution; SA_{250}: surface area with a square window area of 250 m; TD_{100}: topographical distance with 100x cost of movement along an incline; TD_{500}: topographical distance with 500x cost of movement along an incline; TD_{1000}: topographical distance with 1,000x cost of movement along an incline; VW: volume of water along seaway path.

Figure 4.3. Multiplicative regression coefficient for the external infestation pressure (EIP) on abundance of adult female (AF) *Lepeophtheirus salmonis* and 95 % confidence interval (CI) as varied by the Gaussian kernel density estimate (KDE) bandwidth (5-15 km) which was the weight for the calculated EIP. The Akaike’s information criterion (AIC) is also reported for the multivariable linear mixed model for the abundance of AF sea lice as varying according to the KDE bandwidth of the EIP. EIP and AIC were calculated using various connectivity measures: A: surface area with a square window area of 1,000 m; B: topographical distance with 100x cost of movement along an incline; C: topographical distance with 500x cost of movement along an incline; and D: volume of water along seaway path. Red markers represent the estimated exponentiated coefficient and 95 % CI for the EIP and the AIC for the KDE bandwidth 10 km reported in Chapter 3 model where weights were seaway distances at 100 m resolution. Scale is the mean normalized count of AF sea lice after treatment (AF count divided by the number of fish counted and offset by 1).

Figure 4.4. Example of a grid overlayed over a map of the islands east of Grand Manan Island, New Brunswick, where cells have an approximate width and height of 25 m and are assigned values of 1 for coastal waters and ‘No Data’ (nd) for land. The resulting raster image of the waterway map for the same area (B).

Figure 5.1. In-sample short term predictions (8 weeks; red points) of the abundance of AF *Lepeophtheirus salmonis* for single selected sites from each of the Bay Management Areas (BMA) in the Bay of Fundy, New Brunswick, Canada, using a multivariable mixed autoregressive model with 2nd order function. The vertical grey line marks the last date prior to predictions (week 36 of year 2020). Predictions were calculated using the observed values (black points) to generate residuals.

Figure 5.2. The square root of the prediction error for 14 sites (n = 112 observations) across the prediction horizon of 8 weeks. The prediction errors were calculated for in-sample short term predictions of the abundance of adult female *Lepeophtheirus salmonis* using a multivariable mixed autoregressive model.

Figure 5.3. Site contributions (arrows) from neighbouring sites (A-F) to the external infestation pressure of a selected site (black circle) in the Bay Management Area 1 of the Bay of Fundy, New Brunswick, Canada. Colour of the arrows are on the scale of white to orange linearly proportional to the contributions. Site contributions of F are twice the contributions of other sites (A-E). Dashed arrows (A, B and D) point to sites beyond map bounds at seaway distances of 6.5 km, 13.8 km, and 10.0 km, respectively.

Figure 5.4. Ratio of the predicted abundance of the week before to the week after mechanical treatment (thermal and Hydrolicer®) of adult female (AF) *Lepeophtheirus salmonis* sea lice against sea surface temperature in the Bay Management Area (BMA) 1 for mean values of the other continuous predictors. Estimates were produced using the final multivariable autoregressive linear mixed effects model (AR[2]).

Figure 5.5. Estimated mean normalized count of adult female (AF) *Lepeophtheirus salmonis* sea lice (AF count divided by the number of fish counted and offset by 1) against time at sea (TAS) in weeks in the Bay Management Area (BMA) 1 for mean values of the continuous predictors of the final multivariable mixed autoregressive model (AR[2]).

Figure 5.6. Total number of topical treatments applied annually in the Bay of Fundy, New Brunswick, for 2018 to early June 2021. n = 247

Figure 5.7. The standard deviation of the prediction error for 14 sites (n = 112 observations) across the prediction horizon of 8 weeks. The prediction errors were calculated for in-sample short term predictions of the abundance of adult female *Lepeophtheirus salmonis* using a multivariable mixed autoregressive model and as calculated without the influence of the correlation parameters (residual set to zero).

Figure 6.1. The external infestation pressure of *Lepeophtheirus salmonis* sea lice (non-log scale) against the time at sea in weeks for Atlantic salmon at

aquaculture sites in the Bay of Fundy, New Brunswick. The observations were modelled in the multivariable mixed effects time series model in Chapter 5. Locally weighted scatter plot smoothing curve overlay the observations (red dash). Aggregate plot (A) and divided per the Bay Management Areas (BMA) (B).

Figure 6.2. Direct comparison of Fisheries and Oceans Canada and UKMO remote-sensing sea surface temperatures for 29 matched sites (Atlantic salmon aquaculture site and DFO Station) in the Passamaquoddy Bay, New Brunswick, Canada for the period July 2009 to November 2018.

Figure 6.3. Total number of topical treatments applied annually in the Bay of Fundy, New Brunswick, for 2018 to early June 2021. n = 247.

List of Abbreviations

AF	adult female
AGD	amoebic gill disease
AIC	Akaike's Information Criterion
AVC	Atlantic Veterinary College
BC	British Columbia
BKD	bacterial kidney disease
BMA	Bay Management Area
CAF	external contributing adult females
CAHS	Centre for Aquatic Health Sciences
DFO	Department of Fisheries and Oceans, Canada
EIP	external infestation pressure
EMB	emamectin benzoate
FAO	Food and Agriculture Organization of the United Nations
GIS	geographic information system
HSMI	heart and skeletal muscle inflammation
IHNV	infectious hematopoietic necrosis virus
IIP	internal infestation pressure
ISAV	infectious salmon anemia virus
NB	New Brunswick
NL	Newfoundland
NS	Nova Scotia
PAAM	pre-adult male and female, and adult male
PMRA	Pest Management Regulatory Agency
PRV	piscine reovirus
RSC	revised statutes of Canada
SIS	susceptible-infectious-susceptible epidemic model
SRS	salmonid rickettsial syndrome/septicaemia
USA	United States of America

Glossary

Agent: parasite, bacteria or virus that may cause disease.

Anadromous: fish that spawn and develop in freshwater then migrate to a marine environment for their adult life.

Bathymetry: different landforms of the seafloor, also referred to as underwater topography.

Co-infection: a host fish infected by more than one pathogen such as *L. salmonis* and ISAV.

Connectivity: the exchange of an agent among sites resulting from dispersal and is dependent on multiple factors in a spatiotemporal context.

Connectivity measure: measure that estimates the dispersal distance among sites for an area.

Depth: average distance from the seafloor to the water surface.

Dispersal: movement of sea lice outward from one site.

Dispersal distance: measure of distance (Euclidean, seaway or other) for the dispersal of sea lice.

Effective dispersal: the event occurs when there are sufficiently large number of sea lice to allow detection to be labelled as a successful parasitic infestation.

Euclidean distance: 1) shortest distance between two sites irrespective of the landscape, also described as ‘as the crow flies.’ 2) method for the calculation of the distance between two points.

External infestation pressure: infestation pressure applied by *L. salmonis* as a sum of the contribution from neighbouring farm sites, also known as regional sources of sea lice infestation.

Fallowing: a management practice where sites are free of farmed salmon hosts for a period prior to re-stocking with a new batch of smolt, for the purpose of reducing exposure of new fish to pathogens.

Fish-iTrends©: data management system operated by the Centre for Aquatic Health Sciences (CAHS) at the Atlantic Veterinary College (AVC) of the University of Prince Edward Island.

Habitat: location in the environment that has biological significance.

Habitat patch: location that is preferred by the organism for biological or behavioural reasons.

Harvest: salmon are removed from the cages at a site for the purposes of sale (commercial processing) or culling.

Hydrodynamic connectivity: the connection of farm sites through the movement of water.

Infestation pressure: measure that represents the dose of exposure of parasitic stages of sea lice to potential salmon hosts, also known as infection pressure for non-parasitic agents (i.e., bacterial, and viral).

Internal infestation pressure: infestation pressure applied by *L. salmonis* from within the farm site, also known as farm sources of sea lice infestation.

Lecithotropic: life stages that depend on lipid and protein resources from the yolk sac and do not otherwise feed.

Metapopulation: defined group of agents where there is limited dispersal among metapopulations.

Mobile lice: pre-adult and adult male and female *L. salmonis* life stages that are capable of active movement, mostly on the same host; adult male may be capable of swimming to new hosts in very close proximity.

Percent surface area of water: a cumulative average of water percentage within a defined spatial window that follows along the seaway path between two sites, abbreviated to SA, with a subscript value describing the area (m) included in the spatial window.

Persistence: chemotherapeutant that has a prolonged lifespan in the environment, either in the water column or bound to sediment

Planktonic lice: nauplii and copepodid *L. salmonis* life stages that are passively transported by water currents and demonstrate mild swimming capabilities; also known as pelagic life stages.

Raster image: two-dimensional graphical representations of spatial information along a grid of square cells with a defined constant dimension.

Reservoir host: fish species that is infested by sea lice but does not suffer disease from it; sea lice cannot complete its life cycle on it.

Seaway distance: shortest distance through water between two sites, abbreviated to SWD, where the subscript describes the dimension of the raster image.

Seaway path: path between two sites that has the shortest seaway distance.

Sentinel cage: cages stocked with hatchery fish, placed off site but within the coastal marine environment, to monitor regional levels of sea lice infestation or other infectious diseases.

Site: Atlantic salmon farm site.

Source: site from which agent originates, also known as outflux.

Sink: destination site, also known as influx.

Spatial window: square with its centroid located along the seaway path.

Stocking: smolt from regional hatcheries are added or relocated to cages at a site.

Topographic distance: a measure of the distance between two locations while accounting for topographic relief (i.e., elevation or bathymetry), abbreviated to TD, where the subscript describes the cost of movement along an incline. This cost was applied as a depth weighting factor where acute changes in elevation were assigned a larger factor to provide for a greater resistance to vertical movement (i.e., depth).

Volume of water: a volume of water within a defined spatial window that follows along the seaway path between two sites, while accounting for topographic relief, abbreviated to VW, with a subscript value describing the area (m) included in the spatial window.

Zooplankton: animal component of plankton which is a diverse group of organisms that drift in the water, includes *L. salmonis* in its free-living larval stages.

1 Introduction

1.1 Salmonids

Salmonidae (Jarocki or Schinz, 1822) are a family classification of marine, brackish, and fresh teleost fish (Froese and Pauly, 2023). The species included in this family are the Atlantic salmon (*Salmo salar* [Linnaeus, 1758]), Pacific salmon (e.g., Chinook salmon, *Oncorhynchus tshawytscha* [Walbaum, 1792] and Coho salmon, *O. kisutch* [Walbaum, 1792]), trout (e.g., Rainbow trout, *O. mykiss* [Walbaum, 1792] and Brown or Sea trout, *S. trutta* [Linnaeus, 1758]), and char (e.g., Arctic char, *Salvelinus alpinus* [Linnaeus, 1758]), among other species (Froese and Pauly, 2023).

In general, the salmonids are anadromous, meaning they spawn and develop in freshwater and migrate to a marine environment for their adult life. Occasional groups such as *S. salar* remain in freshwater and are non-anadromous or “landlocked” (for review, see Hutchings et al., 2019). The salmon freshwater habitat (defined as the patch of environment that has biological significance [Girvetz and Greco, 2007]) are streams and rivers. At sea, Atlantic salmon were demonstrated to select environments with water temperature range of 14 to 20 °C and mortalities were noted at temperatures above 25 °C (for review, see Breau, 2012). Atlantic salmon smolts transferred to sea cages are exposed to salinities of 30 to 32 ppt. The Canadian 2021 code of practice for the care and handling of farmed salmonids recommends water temperatures between 4 and

18 °C, without providing advice for salinity ranges (National Farm Animal Care Council, 2021).

Salmonids are environmentally, culturally, and economically important. Salmon are considered keystone species on which many other animals and environments rely (Hyatt and Godbout, 2000). Salmon have been managed by many Indigenous groups for thousands of years (Atlas et al., 2021). Wild salmon are harvested in salt and in freshwater for commercial and sporting purposes. Multiple salmonid species are farmed and the most prominent is the Atlantic salmon (Naylor et al., 2021).

1.2 Salmon production

1.2.1 General

The farming of Atlantic salmon is divided into two periods as it is an anadromous species. The first period includes spawning and development of the eggs to the early smolt stage in freshwater at hatchery sites. The eggs develop into alevins, fry and parr for approximately 8 to 16 months when they reach 40 to 120 g (Jones, 2004). The smolt are transported to seawater sites using a combination of specialized trucks, and boats (Erikson et al., 2022). There are certain production systems where the smolt are not moved into sea sites but instead are moved to fresh, brackish, or saltwater tanks during land-based farming (Davidson et al., 2016). At sea, the cages may be square or circular and have many designs. The most common type of cage is the floating high-density polyethylene (HDPE) cage (for a review, see Cardia and Lovatelli, 2015). The sea cages are

typically grouped and may be connected by floating walkways. The transfer of smolt typically occurs in the spring (April to June in the Northern Hemisphere) and in some cases in the fall (November to December). In saltwater, Atlantic salmon develop for approximately 12 to 24 months until they are 4-5 kg or greater (L. Hammell, pers. comm.). The market-sized fish are harvested (removed) from the cages, transported via specialized boat and trucks, and processed for market.

There are multiple strains of Atlantic salmon, wild and domesticated populations (Skaala et al., 2005; Withler et al., 2005; Roberge et al., 2006; FAO, 2020). The wild Atlantic salmon have two distinct lineages: European and North American populations (Withler et al., 2005). Domesticated salmon have been developed multiple times from wild stock through the selection of traits such as improved growth rates and resistance to bacterial diseases (Roberge et al., 2005). Most of the NB production utilizes Canadian strains (L. Hammell, pers. comm.).

Atlantic salmon are maintained in cages at densities of 30 kg/m^3 on average and is regulated by government (L. Hammell, pers. comm.). Infectious diseases can be transmitted among individuals in a cage, among cages in a site (Jaworski and Holm, 1992) and among sites (e.g., McClure et al., 2005; Gustafson et al., 2007; Amundrud and Murray, 2009; Viljugrein et al., 2009). Transmission of infectious and parasitic agents among sites can occur through the water, sharing of improperly sterilized equipment and individuals visiting multiple sites. Additional

details on infectious diseases of concern can be found in Sections 1.3.3 and 1.4 while transmission/dispersal is discussed in detail in Section 1.7.

1.2.2 World

Pisciculture supplies fish globally to support the traditional fishery practices of commercial sea and freshwater fisheries. Worldwide pisciculture has contributed 46.0 % of the total global fish production in 2016-2018 (FAO, 2020). In 2017, 59.6 % of stocks were maximally sustainably fished (i.e., the fishing practices are considered biologically sustainable) and 34.2 % were overfished (FAO, 2020). The salmon market was relatively resilient to the uncertainties and challenges in global fish production of the 2019-2021 COVID-19 pandemic (Murray et al., 2021; FAO, 2022). Despite these challenges, global salmon production increased to produce 2.8 million tonnes in 2021 (FAO, 2022).

Atlantic salmon production dominated the farmed salmon market and provided greater than half the total global salmon market in 2018 (FAO, 2020). Salmonid production globally is dominated by the Norwegian and Chilean industries (FAO, 2020). Japan, Canada, Scotland, the Faroe Islands, Northeastern USA, Tasmania, and a few other countries also farm Atlantic salmon (FAO, 2020).

Salmon fisheries and aquaculture efforts are performed along the entire coast of Norway in fjords and coastal areas. The Norwegian aquaculture industry is characterized by large production units that overlap with the migration pathways of wild salmonids: Atlantic salmon, Arctic char, and sea trout (Myksvoll et al.,

2018). In 2021, Norway produced 387.6 million Atlantic salmon and production increased by an estimated 10 % from 2020 (FAO, 2022). Norway also farms Rainbow trout (Myksvoll et al., 2018).

The Chilean aquaculture industry occurs at sites among the islands and fjords of the Patagonian Fjords System that is at the southwestern coast abutting the Pacific Ocean (Niklitschek et al., 2013). There are feral populations of Chinook salmon in the Patagonia area because of governmental and private efforts to introduce salmon for recreational fishing (for review, see Niklitschek et al., 2013). In 2021, Chile harvested a total biomass of 723,650 tonnes of Atlantic salmon (FAO, 2022). Growth rate of salmon production has declined for the period 2005-2017 and production level reached a biological limit for sustained production (Marine Harvest, 2018). Chilean salmon production includes the Rainbow trout, Coho salmon and Atlantic salmon (Price et al., 2017).

Additional international review of fisheries and aquaculture production can be found in the regularly published GLOBEFISH Highlights (e.g., FAO, 2022).

1.2.3 Canada

1.2.3.1 Atlantic Canada

Salmon production on the East Coast of Canada is generally limited to the aquaculture industry, however there is recreational fishing, with extremely limited wild stocks (Government of New Brunswick, 2023). Marine sites produce Atlantic salmon primarily and to a smaller degree trout of multiple species. Sites are in

the provinces of New Brunswick (NB), Nova Scotia (NS), and Newfoundland and Labrador (NL). New Brunswick is the largest producer of Atlantic salmon in Atlantic Canada when in 2021 it produced 27,423 metric tonnes (20.2 % of total 136,090[1] metric tonnes Canadian production value) compared to the 8,592 metric tonnes (6.3 %) for NS (Statistics Canada, 2022) and an estimated 15,904 metric tonnes (11.7 %) in NL (Newfoundland and Labrador Department of Fisheries, Forestry and Agriculture, 2022).

The following is a brief overview of the NB Atlantic salmon aquaculture industry as the thesis research focuses on this study area. Chapter 2 will further describe the NB Atlantic salmon aquaculture industry including the environment, a history of the monitoring practices, and a descriptive analysis of the sea lice counts in 2009-2018. Additional details on the NB Atlantic salmon industry can be found in manuscripts and reports by Chang and Page (Chang et al., 2011; Chang and Page, 2014; Chang et al., 2014; Chang and Coombs, 2016).

The NB aquaculture industry is most heavily present in the Bay of Fundy in Charlotte County (Figure 1.1A). Aquaculture in the area first began in 1978 at Deer Island in Passamaquoddy Bay as a research program (Bailey et al., 1983) and has since expanded to cover the southwestern coast of the province. There

[1] As calculated for the four salmon producing Canadian provinces utilizing the total production value submitted to Statistics Canada (NB, NS and BC) and the Newfoundland and Labrador Department of Fisheries, Forestry and Agriculture.

are more than 90 designated lease areas with approximately 40 active sites (Flaherty et al., 2019), and these are allocated into Bay Management Areas (BMAs [Figure 1.1B and C]). The sites are allocated by the provincial government under the Bay of Fundy Marine Aquaculture Site Allocation Policy (New Brunswick Department of Agriculture Fisheries and Oceans, 2013). The BMAs were developed for the purpose of managing Infectious Salmon Anaemia Virus (ISAV [New Brunswick Department of Agriculture Fisheries and Oceans Canada, 2013]; more in Section 1.5.1.1), an internationally notifiable infectious disease that caused severe mortality events starting in 1997 and continues to cause sporadic outbreaks (L. Hammell, pers. comm.).

The Bay of Fundy, NB, is characterized by water temperatures that vary seasonally from 1 °C to 15 °C (Peterson et al., 2001; Jones et al., 2012; Gautam et al., 2016). The salinity is approximately 30 ppt or higher (Greenberg et al., 1984; Drinkwater, 1987) with substantial vertical mixing due to high tides. Temporary variations in salinity may occur during freshet events, when fresh water from surrounding rivers and estuaries discharge into the ocean (e.g., St. Croix River estuary [Fife et al., 2015]) following heavy rains or melting snow.

As of 2014, the salmon farms harvested their fish at an average of 4.7 kg (range of 3.6-6.4 kg) after an average growing period in sea cages of 25 months with a range of 20-30 months (Chang et al., 2014). The Atlantic salmon total biomass for production in the area is not published, in part because the maximum

potential number fish permitted at a site and allowable production levels were discontinued for preference toward a performance-based management approach (Chang and Coombs, 2016). From 2006-2012, the area stocked approximately 6-8 million smolts per year and these smolt originated from 10-14 active salmon hatcheries in the province (Chang et al., 2014). The production of finfish, which is mostly Atlantic salmon, peaked in 2002 at approximately 38,900 metric tonnes (Chang et al., 2014; New Brunswick Department of Agriculture, Aquaculture and Fisheries, 2022). The Government of New Brunswick stated that there may be opportunities for the growth of the industry through improvement of productivity (e.g., increased research on sea lice reduction), the application of new technologies (e.g., land-based post-smolt production facilities) and aquaculture of different species of finfish (e.g., Arctic char), to name a few (New Brunswick Department of Agriculture, Aquaculture and Fisheries, 2022).

The Bay of Fundy is also the site for Atlantic salmon aquaculture in the state of Maine, USA (Figure 1.1). The sites from BMA 1 (Passamaquoddy Bay) neighbour Cobscook Bay which has approximately 9 active leases to produce Atlantic salmon, Rainbow trout and other species around the Moose and Cooper islands (Maine Department of Marine Resources, nd). The production output for the Cobscook Bay is unknown. Therefore, the thesis focused solely on the Canadian aquaculture sites in the Bay of Fundy, NB.

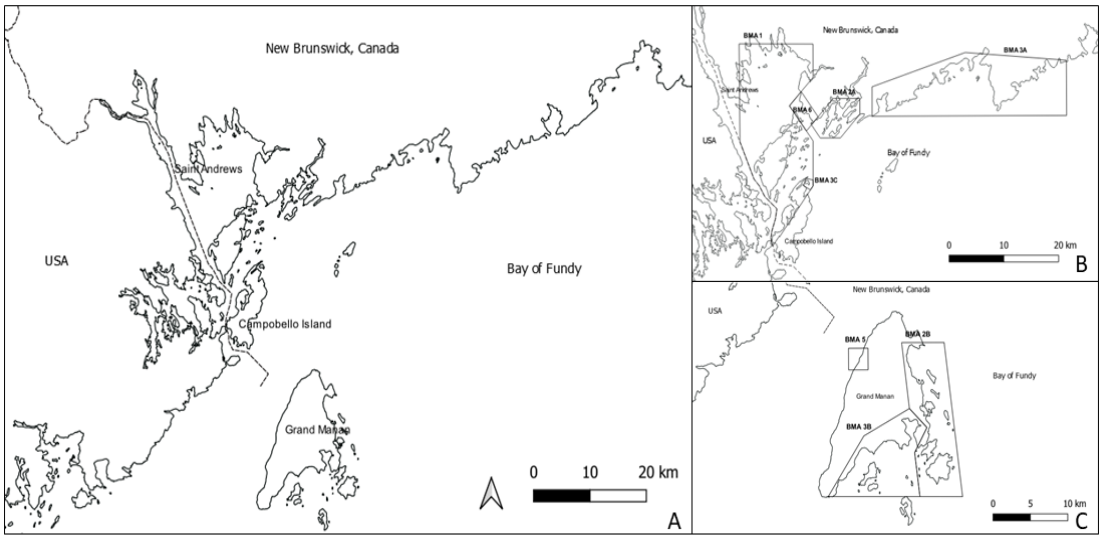

Figure 1.1. Reproduced figure from Chapter 2. The Bay of Fundy, New Brunswick, Canada, study area (A) and Bay Management Areas (BMA) in the southwest coast and Passamaquoddy Bay (B) and Grand Manan Island (C). The locations of the BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (NB Department of Energy and Resource Development/Agriculture, and Fisheries, 2017).

The East Coast has small populations of wild salmonids (Department of Fisheries and Oceans, 2014; Government of Canada, 2019). In the Bay of Fundy, there are the inner (Government of Canada, 2019) and outer (Department of Fisheries and Oceans, 2014) Bay of Fundy Atlantic salmon populations. The habitat for these populations may overlap with aquaculture sites (i.e., farms; Chang et al., 2011), but the migration patterns for these small populations are mostly unknown. Their disease status and levels of potential parasitic infestations are unknown, and for these studies, interactions with wild populations are assumed to be non-existent for the aquaculture sites in the Bay of Fundy.

1.2.3.2 West Coast

British Columbia (BC) is Canada's main province to produce Atlantic salmon. The main production area is the Broughton Archipelago and sites are otherwise located among the islands on the East and West Coast of Vancouver Island (Department of Fisheries and Oceans, 2021). Atlantic salmon is the main farmed species, but there is also Chinook salmon, Coho salmon, and Rainbow trout production to a small degree. BC contributed 84,171 metric tonnes of salmon in 2021 (Statistics Canada, 2022), which accounted for 61.8 % of the total Canadian production.

The West Coast production area has a greater variation in salinity and reduced variation in temperature, compared to the East Coast. In the Broughton Archipelago, Byrne et al. (2018) measured monthly average water temperatures and observed ranges between 6.9 and 16.7 °C at the surface (≤ 1 m depth) and 6.9 and 10.5 °C at 6 m below the surface. The measured monthly average salinity for 2012-2014 was 19-35 ppt at the surface and 24-34 at 5 m (Byrne et al., 2018). The salinity patterns reflect seasonal trends where it decreased from May to August, increased from September to November, and remained stable for the remainder of the year at approximately 35 ppt (Byrne et al., 2018).

The Broughton Archipelago is an area for spawning and development of wild salmonids. There is research into these wild populations to address general concerns about potential transmission of disease agents and pests among

farmed and wild salmon in the area (e.g., Saksida et al., 2007). Overall, the public perception of aquaculture is more negative on the West Coast compared to the East Coast, which may be due to an increased number of environmental non-governmental organizations in BC with stronger public campaigns against marine aquaculture (Coletto et al., 2011; Flaherty et al., 2019). Of the Atlantic salmon producing provinces, NB has the more positive perception of aquaculture (Kraly et al., 2022), which may be in part due to local ownership and job creation (Chang et al., 2014). The social, political, and environmental perceptions of the public in relation to salmon aquaculture serve as an important driver for industry and governmental policies and regional management practices and are further discussed by Flaherty et al. (2019) and Kraly et al. (2022).

1.3 Sea lice

1.3.1 Species

Sea lice are marine copepods of the family Caligidae that are ectoparasites of fish. There are multiple species of sea lice that are of concern because they have considerable impact on farmed and wild salmonids. The following is a summary of the species of interest, their biological characteristics, and diseases they cause. Additional review information can be found in Fast and Dalvin (2020).

Lepeophtheirus salmonis (Krøyer, 1837) is a marine ectoparasite of fish and is commonly known as the “salmon louse” as it primarily infests salmonids with a preference for Atlantic salmon (Pike and Wadsworth, 1999). There are two subspecies, *salmonis* (Krøyer, 1837) and *oncorhynchi* (Skern-Mauritzen et al.,

2014) located in the Atlantic and Pacific oceans, respectively (Skern-Mauritzen et al., 2014; Walter and Boxshall, 2023). Yazawa et al. (2008) proposed that *L. salmonis oncorhynchi* diverged from *L. salmonis salmonis* when the species migrated across the Bering strait approximately 2.5-11 million years ago. *L. salmonis salmonis* has a range that extends across the Atlantic Ocean affecting salmon in Eastern Canada and Northern Europe (Todd et al., 2004; Boxaspen, 2006; Glover et al., 2011; Besnier et al., 2014). *L. salmonis oncorhynchi* are limited to southwestern coast of Canada. There are multiple differences among these subspecies (for review, see Brooker et al., 2018), and a few examples include different protease secretions (Fast et al., 2002), reduced disease on farmed salmon in BC (Saksida et al., 2007), and increased tolerance to low salinity in the Pacific subspecies compared to the Atlantic one (Johnson and Albright, 1991a; Bricknell et al., 2006). Within the families of *L. salmonis salmonis* there was noted differences in tolerance to water temperature and salinity variation; however, the results for the salinity tests could not be “disentangled” from background mortality (Ljungfeldt et al., 2017).

Another important species of sea lice is *Caligus rogercresseyi* (Boxshall and Bravo, 2000 [Walter and Boxshall, 2023]) in Chile (Bravo, 2003) that mainly infests salmonids (Fast and Dalvin, 2020). Additional *Caligus* species that are occasionally noted are *Caligus clemensi* (Parker and Margolis, 1964), *Caligus elongatus* (von Nordmann, 1832), and *Caligus curtus* (Müller O.F., 1785 [Walter and Boxshall, 2023]).

The different species of sea lice infest farmed Atlantic salmon to different degrees. In NB, *L. salmonis salmonis* abundance is greater than *C. elongatus* and *C. curtus* since the mid-1990s (Westcott et al., 2004; Chang et al., 2011). In BC, *L. salmonis oncorhynchi* infestations are greater than *C. clemensi* and infestations were more sporadic (Saksida et al. 2010).

1.3.2 Biology of sea lice

Since *L. salmonis salmonis* is the major pest for the NB aquaculture industry, the summary review of the biological aspects of sea lice will focus on this species. The morphology of the louse will not be described, but those descriptions can be found in Johnson and Albright (1991b) and Schram (1993).

L. salmonis undergo eight life stages in their development (Hamre et al., 2013). Figure 1.2 presents a schematic of the life cycle. In *L. salmonis*, eggs are released by the adult female in a structure called an egg string, which remains attached for several days until the eggs hatch as nauplii (plural form of nauplius; Johnson and Albright, 1991a). A female produces two egg strings, and interestingly, if an egg string gets detached from the progenitor, the eggs can develop successfully, as demonstrated under laboratory conditions (Eisenhauer et al., 2020). After hatching, the nauplius 1 molts into nauplius 2 followed by the copepodid stage. The nauplii and early copepodid are pelagic (free-living in the water column; Schram, 2004) with some vertical swimming capabilities (Johnsen et al, 2016); therefore, they can be considered part of the zooplankton (defined

as the animal component of plankton which is a diverse group of organisms that drift in the water). These stages generally remain in the top 5 m of the water column (Costelloe et al., 1996; Heuch et al., 1995, 2005; Johnsen et al., 2014, 2016). The nauplii and early copepodid are lecithotrophic as they depend on lipid and protein resources from the yolk sac and do not otherwise feed (Johnson and Albright, 1991b). These life stages are difficult to study under field conditions as they are difficult to capture using tools and techniques commonly used for the study of pelagic zooplankton (Penston et al., 2004, 2011; Asplin et al., 2011; Skarðhamar et al., 2019). Sea lice are found lower concentrations of 0 to 24 individuals per m^3 (Fernandez-Gonzalez et al., 2022) compared to other plankton (approximately 100 individuals per m^3) and have high spatiotemporal variations and patchy distributions (Skarðhamar et al., 2019).

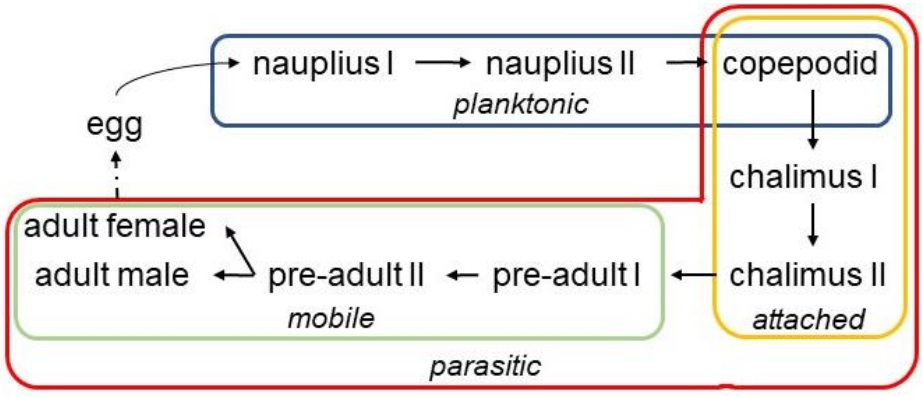

Figure 1.2. Life cycle stages of development of *Lepeophtheirus salmonis* with the location of this ectoparasite in the water column.

The copepodid is parasitic and will attach to a host if available or will succumb to starvation (Brooker et al., 2018). The ability of the copepodid to infest a host

varies from the time since the molt as it is dependent on the yolk reserves; the mid-life copepodid with the decreasing lipid resources may be the most active and thus capable to infest a host (Cook et al., 2010; Brooker et al., 2018). On the host, the copepodid will attach reversibly or commit to settling (Brooker et al., 2018). Once attached, the copepodid will molt into the chalimus 1 stage that is securely attached to the host using a frontal filament (for review, see Pike et al., 1993). A note here, there is the recent publication that argues that the terms “chalimus” and “pre-adult” should be dropped and instead be part of the copepodid stage I-V (Piasecki et al., 2023). For this thesis and the pending publications related to this work, the terms chalimus and pre-adult are maintained. The chalimus 2 molts into the pre-adult 1 that is motile on the host. The pre-adult stage is characterized by its motility where the frontal filament is detached from the host (*Caligus* sp.: Kim, 1993; Lin and Ho, 1993; Piasecki, 1996; and *Lepeophtheirus* sp. Astensrud, 1990). The pre-adult 1 molts into pre-adult 2 followed by the adult stage. In *Caligus* sp., there are 4 chalimus stages followed by a molt into the adult stage (Piasecki, 1996; Hamre et al., 2013). The attached and mobile life stages feed on the host (additional details on the feeding activities and consequent pathology can be found in Section 1.3.3). The mobile life stages may move among salmon hosts (Bruno and Stone, 1990; Jaworski and Holms, 1992).

Sexual dimorphism begins at the chalimus 1 stage (Hamre et al., 2013) but is difficult to observe under field conditions. In the field, the adult female louse (AF)

is easily differentiated from other stages by the presence of the paired egg strings and larger size. The male louse is less easily differentiated from the pre-adult stages and is therefore commonly grouped into the pre-adult and adult male category (PAAM [Elmoslemany et al., 2013]).

Adult female sea lice produce eggs over multiple generations (Ritchie et al., 1993; Mustafa et al., 2000a). Under laboratory conditions, adult female lice were observed to produce up to 10-11 pairs of egg strings during infestation on Atlantic salmon (Heuch et al., 2000; Mustafa et al., 2000a). The egg strings produced earlier had greater viability compared to later in the life span of the adult female (Mustafa et al., 2000a), and Heuch et al. (2000) noted that the first pair of egg strings were significantly shorter compared to subsequent production. Some of these adult female lice survived up to 7 months (Mustafa et al., 2000a) and 191 days at 7.2 °C (Heuch et al., 2000). The production of new eggs strings occurred within approximately 10 days of removal of the extruded eggs during laboratory tests (Heuch et al., 2000). There is also seasonal variability in the production of the size and number of the eggs in the egg strings (Tully, 1989; Ritchie et al., 1993; Tully and Whelan, 1993). At the lower temperature evaluated (7.1 °C), Heuch et al. (2000) observed that the egg strings were longer and had a greater number of eggs compared to the higher temperature of 12.2 °C. However, the greater number of eggs did not translate to greater viability, where at 7 °C there was a larger proportion of non-viable eggs (Heuch et al., 2000). There have been multiple attempts to estimate the average number of eggs in

pairs of egg strings produced by adult female *L. salmonis*, and Costello (1993) listed values ranging from 107 to 1,220. The host species was demonstrated to affect the number of eggs produced (Johnson, 1993) and other factors such as the health and diet of the host, and genetic differences in the lice may affect variation in the fecundity, too (Johnson and Albright, 1992; Brooker et al., 2018). The high fecundity of the sea lice is part of the overall success of this parasite (Brooker et al., 2018).

The development of each stage of *L. salmonis* is dependent on environmental conditions (for review, see Fast and Dalvin, 2020). The main factors responsible for the rate of development are water salinity (Bricknell et al., 2006; Wright et al., 2016) and temperature (Tucker et al., 2000; Stien et al., 2005; Samsing et al., 2016; Hamre et al., 2019). In optimal environments, such as temperature between 15-20 °C there was noted 100 % hatching and development success (Samsing et al., 2016). Planktonic stages have greater susceptibility to low salinity compared to the parasitic stages (Wright et al., 2016) but still there is decreased survival with decreasing salinity (Groner et al., 2016a). Brooker et al. (2018) proposed that attached stages obtain protection from close contact to the host and the ingested host tissue. The copepodid required a salinity of 30 ppt or higher for successful host infestation (Bricknell et al., 2016). The variation of salinity in BC was proposed as one of the factors controlling the abundance of *L. salmonis* (Brooks, 2005; Elmoslemany et al., 2015; Arriagada et al., 2016). Stien et al. (2005) demonstrated that each life stage growth rate depends on the water

temperature and there is a positive relationship despite a lack of agreement on specific times per stage at specific temperatures (Gravil, 1996; Boxaspen and Naess, 2000; Samsing et al. 2016). At temperatures below 6 °C and above 21 °C, the development, egg production and capability to infest a host are reduced (Samsing et al., 2016; Brooker et al., 2018; Hamre et al., 2019).

Generally, male *L. salmonis* infesting Atlantic salmon develop at a rate of approximately 40 days at 10 °C, and females take approximately 50 days at the same temperature (Johnson and Albright, 1991a; Stien et al., 2005). The growth rates of other species of sea lice vary compared to *L. salmonis*; for example, the copepodid of *C. elongatus* takes a larger proportion of the time to total development compared to *L. salmonis* (Hamre et al., 2019).

Sea lice development is a major field of research as it is considered vital to understanding the biology of this pest, the transmission among host fish and identifying methods for their mitigation. Hamre et al. (2019) and Stien et al. (2005) are some of the foremost references of *L. salmonis* development under laboratory conditions. Utilizing the information from the laboratory, Revie et al. (2005) and others (Amundrud and Murray, 2009) have modelled the development of *L. salmonis* in the field to improve the understanding of sea lice infestation observed at aquaculture sites. Multiple other models have been developed since (e.g., Murray and Salama, 2016). Additional discussions of sea lice models can be found under Section 1.8.

1.3.3 Pathology secondary to sea lice infestation

Sea lice infestation results in direct and indirect pathology of the host fish. Tully and Nolan (2002) reviewed the host-parasite interactions. The feeding activities of the pre-adult 1 and 2 and adult life stages cause damage to the mucus layer and dermis of the fish (Kabata, 1974), on which they feed as well as any fresh blood that may result from the tissue damage (Brandal et al., 1976). Severe ulceration and infestation may result in anemia (González et al., 2016).

The mucus layer and dermis are the primary barriers to the infestation by sea lice and are the site for non-specific (i.e., innate) immune responses to infestation (Brooker et al., 2018). The sea louse can excrete factors and enzymes to suppress or redirect host immune responses during infestation (Fast, 2014). Non-specific immune responses to the attachment of the sea louse included transient skin melanisation (Fast and Dalvin, 2020), increased protease activity in the mucous layer of the skin (Ross et al., 2000), and decreased phagocytic activities in the blood (Mustafa et al., 2000b). The skin of infested fish had differential gene expression per individual (Robledo et al, 2018a). The innate responses, such as melanisation, may play a role in reducing AF *L. salmonis* infestation (Fast and Dalvin, 2020). The innate immune response to sea lice infestation is dependent on host variables such as the species, age, health, and others (Brooker et al., 2018).

The disruption of the skin barrier results in disorder of the host osmoregulatory mechanisms necessary for optimal health and can produce physiological changes consistent with a stress response (Grimnes and Jakobsen, 1996). Fjelldal et al. (2020) and others (Grimnes and Jakobsen, 1996) found that Atlantic salmon had a poorer ability to osmoregulate during infestations with *L. salmonis*. Juvenile fish may succumb to osmoregulatory failure secondary to infestation by sea lice (Brauner et al. 2012; Patanasatienkul et al., 2013); the size and age at which the juveniles may die depends on the species (< 1 g in Pink [Jones and Hargreaves, 2009], and approximately < 60 g in Atlantic salmon [M. Fast, pers. comm.]). Ives et al. (2023) estimated *L. salmonis* sub-lethal physiological effects at 0.08 lice per g of the host and 50 % probability of mortality onset at 0.24 lice per g of host (95 % confidence interval 0.1 to 0.67 lice per g). These estimates were derived from a literature review of studies assessing the physiological impacts of lice on salmon as the sub-lethal and lethal effects vary according to the species of salmon and lice, farmed or wild stock salmon, life stage of the lice, laboratory conditions, and other factors. The life stage of the sea lice impacted the lethality of the infestation (Ives et al., 2023); pre-adult and adult sea lice resulted in lower survival compared to chalimus life stages (Grimnes and Jakobsen, 1996; Hvas and Bui, 2022). Another important factor is the potential for the host to contract a sufficient dose of viable pathogen, in this case the sea lice (Schmid-Hempel and Frank, 2007).

The adaptive immune response is activated after attachment and feeding of the sea lice. An inflammatory response results in a greater humoral than cellular immune response (Th_2-type regulatory pathway [Braden et al., 2015]). Atlantic salmon, sockeye salmon (*Oncorhynchus nerka* [Walbaum, 1792]) and chum salmon (*Oncorhynchus keta* [Walbaum, 1792 {Froese and Pauly, 2023}]) have genotype and phenotype characteristics that increase their susceptibility to infestation by *L. salmonis* (Johnson and Albright, 1992; Fast and Dalvin, 2020). Pink salmon (*Oncorhynchus gorbuscha* [Walbaum, 1792 {Froese and Pauly, 2023}]) and Coho salmon infestation by *L. salmonis oncorhynchi* resulted in a strong immune response and rejection of the lice (Fast et al., 2002; Johnson and Albright, 1992). Infestation by *C. rogercresseyi* resulted in up-regulation of genes for the innate immune response including interleukins, interferons, and complement components (Robledo et al., 2018a). Interleukins 8 and 1β and tumour necrosis factor α-1 have been identified and proposed as important components for the rejection of *L. salmonis* infestation in pink salmon (Fast, 2014). Valenzuela-Muñoz et al. (2023) found Atlantic salmon resistant phenotypes to *C. rogercresseyi* had an upregulation of genes associated with tissue repair compared to salmon families labelled as susceptible.

The tissue damage and incited stress response from sea lice infestation may affect the growth, fecundity, and survival of fish (Pike and Wadsworth, 1999; Tully and Nolan, 2002; Heuch et al., 2005). Under laboratory conditions, specific growth rates were lower in infested compared to control Atlantic salmon (Fjelldal

et al., 2020). All these effects could have a significant impact on the production of Atlantic salmon. Another challenge to the salmon is the possible secondary infections after and during infestation by sea lice (Tully and Nolan, 2002; Pike and Wadsworth, 1999; Heuch et al., 2005). The state of co-infection may exacerbate disease and is discussed with additional details in Section 1.4.

1.4 Other diseases of farmed salmon

There are multiple pathogens of concern that affect the production of Atlantic salmon. Some of these are the infectious salmon anemia virus (ISAV), furunculosis, bacterial kidney disease (BKD), salmonid rickettsial syndrome/septicaemia (SRS), amoebic gill disease (AGD), and piscine reovirus (PRV)/heart and skeletal muscle inflammation (HSMI); not all of these pathogens of concern are present in NB. Farmed salmon can be infected with these pathogens while being infested by sea lice, which is termed a co-infection, and may be the result of synergistic effects between the pathogens (Long et al., 2019). Another concern is the potential for adverse events secondary to sea lice treatments for fish suffering from gill damage secondary to AGD or other pathogens.

Co-infections between these pathogens and sea lice may develop from an infestation of sea lice that results in immunosuppression and secondary pathogen invasion (Fast, 2014; Braden et al., 2015) or from the disease of these

other pathogens of concern and secondary infestation by sea lice (e.g., Lhorente et al., 2014).

ISAV is particularly relevant to the NB aquaculture industry because of outbreaks in 1996-1997 (Hammell and Dohoo, 2005) and 2002-2004 that occurred in the Bay of Fundy, NB, and the Quoddy region of Maine (Gustafson et al., 2007). Sea lice infestation had been suspected to increase risk of infection by ISAV (Hammell and Dohoo, 2005) as sea lice were demonstrated to transmit viruses including ISAV (Nylund et al., 1993, 1994). An unconditional association was observed between greater sea lice bath treatment frequency and a decreased risk of ISAV (Hammell and Dohoo, 2005). In Chile, an increased abundance of *C. rogercresseyi* was associated with increased prevalence of ISAV (Valdes-Donoso et al., 2013). Infestation by *L. salmonis* followed by ISAV infection was associated with increased mortality rates of Atlantic salmon compared to uninfected controls and ISAV infected groups only (Barker et al., 2019). Barker et al. (2019) proposed the mechanism behind this increased susceptibility to ISAV following infestation was the manipulation of genes that increased successful settlement such as the decrease of MCH1-β in coinfected fish.

It is theorized that *L. salmonis* may function as a reservoir for *Aeromonas* sp., the genus that includes multiple species of interest as the causative agents of furunculosis (Nese and Enger, 1993), and other ectoparasites may as well. *Aeromonas* sp., BKD and AGD have not been specifically evaluated as co-

infectious pathogens with sea lice as far as this author is aware. However, infection by one of these pathogens may increase risk of infestation by sea lice. For example, BKD can cause decreased appetite (Pirhonen et al., 2000) and decreased growth (Sandell et al., 2015) such that the salmon may be less able to perform behaviours to avoid sea lice and to produce sufficient immune response to avoid high infestation rates. Infestation by sea lice may predispose the host fish to these pathogens and the fish may have worse disease outcomes. Co-infection with sea lice and, separately, infectious hematopoietic necrosis virus (IHNV; which is not endemic to Atlantic Canada [Long et al., 2019]) and *Piscirickettsia salmonis*, which often co-occurs with *Renibacterium salmoninarum*, the causative agent of SRS (Lhorente et al., 2014) have been evaluated. These co-infections resulted increased mortality (Lhorente et al., 2014; Long et al., 2019).

1.5 Mitigation of sea lice

1.5.1 Management strategies

There are multiple management strategies for sea lice. Management strategies of prevention and control can be differentiated as preventative, i.e., to reduce the rate of new infestations, and treatments that aim to control the current infestation (Barrett et al., 2020). Prevention strategies include area-wide management with coordinated fallowing efforts. Treatments can be immediate, such as the application of hydrogen peroxide, or continuous, such as with the use of cleaner fish that remove lice on an ongoing basis (Barrett et al., 2020). A final effort for

the minimization of sea lice infestation is the harvesting of salmon (i.e., removing susceptible hosts). Guragain et al. (2021) reviewed sea lice infestation management practices.

Monitoring practices can also be considered part of a management strategy. Sea lice are regularly counted by staff or members of a governing body. These counts aid veterinarians and farmers to make decisions about when to schedule treatments (additional discussion on treatments in Section 1.5.1.2). These counts can further be amalgamated into a database to aid in area-wide management decisions (additional discussion on monitoring in Section 1.6).

1.5.1.1 Zonal management

Area-wide management efforts occur at multiple levels. At the lowest level, there is the cage where biosecurity measures are employed, such as chemical bath treatments, to decrease the abundance and spread of pathogens from one cage to another. Then, there is the site, which is an amalgamation of multiple cages, where biosecurity measures are also employed. Biosecurity measures can include strategies such as cleaning equipment and personnel clothing, in the case of bacterial or viral pathogens, or site-level in-feed chemotherapeutants.

Zones also exist by grouping together sets of neighbouring sites for the coordination of the production cycles and management practices for salmon farming. Typically, zones are determined by the governing body that assigns the aquaculture site leases, and each lease is assigned to a specific zone. The

zones are developed for the purpose of prevention a specific pathogen, such as the Bay Management Areas (BMAs) in NB which were revised in 2006 for improved ISAV risk management (Chang et al., 2007). In Norway, the prescribed areas are production zones that were developed from clusters of sites identified using connectivity results from a hydrodynamic model combined with a sea lice simulation model (Myksvoll et al., 2018), and in Chile ‘barrios’ were designed with the intension to manage ISAV after the 2007-2009 outbreak (Iizuka and Zanlungo, 2016). The management zones allow for coordinated efforts within an area during scenarios of pathogen outbreak or for other purposes.

Commonly, fallowing is applied within management zones. Fallowing is defined as a time where sites are free of salmon prior to stocking a new batch of smolt, for the purpose of reducing the presence of infectious/parasitic agents (i.e., reduce exposure pathogens). Fallowing has been demonstrated to be effective to reduce sea lice infestation (Bron et al., 1993; Toorians and Adams, 2020), and the incidence of ISAV, salmon pancreas disease virus, infectious pancreatic necrosis virus (Werkman et al., 2011) and AGD (Douglas-Helders et al., 2004). Fallowing can be synchronized within a prescribed area (all sites within a zone fallow within a close time range and occurs when sites stock and harvest synchronously, too), partial (mixture of synchronized and unsynchronized fallowing practices within a zone) and unsynchronized (fallowing is applied to a site irrespective of the timing of the fallowing period at neighbouring sites within a zone [Werkman et al., 2011]). Murray and Salama (2016) applied the Werkman

et al. (2011) susceptible-infectious-susceptible (SIS) epidemiological model to assess co-ordinated fallowing and treatment of sea lice; synchronization of fallowing within a management area results in a slow increase of sea lice numbers in newly stocked sites.

The concept of synchrony in management strategies can be applied to either zones or sites. Single-year class zone and/or site management is achieved by the stocking of salmon within a short period and the coordination of treatments among cages at a site. It is common in the Atlantic salmon industry to have synchronized stocking of cages at a site (usually within a month or so), as it was showed that single generation sites reduced the incidence of sea lice and other disease (Jackson et al., 1997), and management is easier with regards to feeding and stocking/harvesting activities. With synchronized stocking and harvesting of all fish within a site, producers refer to these as production cycles; one production cycle includes a group of salmon on one site from the moment they are stocked with smolts until the last fish is harvested.

1.5.1.2 Treatments

There are multiple active mitigation strategies, which are generally referred to as treatments, for sea lice (Table 1.1; Guragain et al., 2021). Overton et al. (2019) and Guragain et al. (2021) review the treatments available. These treatments may be time-consuming, costly, resource-intensive (with the use of equipment and personnel), and potentially associated with detrimental effects on the

environment. Chemotherapeutants are applied topically in a water bath or administered orally via the feed. Here, chemotherapeutants include drugs and pesticides, which are different in their regulations. There are also physical/mechanical removal techniques that are more recent treatments for the management of sea lice. A selection of treatments is reviewed below as they are relevant to the study area; there are currently only four therapeutic treatments available/commonly used in the Bay of Fundy, NB.

Table 1.1. Summary of treatments for *Lepeophtheirus salmonis* as available in New Brunswick, Canada. Note that lufenuron is exclusively given to fish in hatcheries, while all others are given to fish or applied in cages at sea. The duration of effect refers to an observed decrease in sea lice during future counting events rather than residual effects secondary to absorption (duration of action).

Common name	**Main producer**	**Active ingredient**	**Drug class**	**Method of action**	**Method of application**	**Duration of effect**	***L. salmonis* life stages affected**
SLICE	Merck Animal Health, USA	emamectin benzoate	avermectin, macrocyclic lactone	disruption of neurotransmission (chloride-gated channels)	in-feed	6-10 weeks	chalimus, pre-adult, adult (Davies and Rodger, 2000)
Ivomec®	Boehringer Ingelheim, Germany	ivermectin	avermectin, macrocyclic lactone	disruption of neurotransmission (chloride-gated channels)	in-feed	1 month	chalimus, pre-adult and adult (Johnson and Margolis, 1993; Davies and Rodger, 2000)
Noromectin®	Norbrook Laboratories Ltd., United Kingdom	ivermectin	avermectin, macrocyclic lactone	disruption of neurotransmission (chloride-gated channels)	in-feed	1 month	chalimus, pre-adult, adult (Davies and Rodger, 2000)
Imvixa™	Elanco Animal Health, USA	lufenuron	benzoyl phenyl urea	inhibition of chitin synthesis, formation, and deposition	in-feed	6.6-9 months (Junquera et al., 2019)	chalimus and pre-adult (Junquera et al., 2019)

Table 1.1 continued next page.

Table 1.1 continued.

Common name	Main producer	Active ingredient	Drug class	Method of action	Method of application	Duration of effect	*L. salmonis* life stages affected
Lepsidon™	Ewos® Cargill, Inc., Canada	diflubenzuron	benzoyl phenyl urea	inhibition of chitin synthesis, formation, and deposition	in-feed	3 weeks (like teflubenzuron)	chalimus and pre-adult at molt (PMRA, 2003)
Calicide®	Nutreco ARC Ltd., Netherlands	teflubenzuron	benzoyl phenyl urea	inhibition of chitin synthesis, formation, and deposition	in-feed	3 weeks (Campbell et al., 2006)	chalimus and pre-adult at molt (PMRA, 2003)
Alpha Max®	Zoetis Inc., USA	deltamethrin	pyrethroid	disruption of neurotransmission (sodium-gated channels)	bath: tarp or wellboat	2 weeks (Whyte et al., 2014)	chalimus, pre-adult and adult (Overton et al., 2019)
Excis®	Novartis Animal Health Limited, Switzerland	cypermethrin	pyrethroid	disruption of neurotransmission (sodium-gated channels)	bath: tarp or wellboat	2 weeks (Jimenez et al., 2013)	chalimus, pre-adult and adult (Davies and Rodger, 2000; Jimenez et al., 2013)
Salmosan® Vet	Benchmark Animal Health, UK	azamethiphos	organophosphate	disruption of neurotransmission (acetylcholinesterase)	bath: skirt, tarp or wellboat	2 weeks (Barisic et al., 2019)	pre-adult and adult (Overton et al., 2019)

Table 1.1 continued next page.

Table 1.1 continued.

Common name	Main producer	Active ingredient	Drug class	Method of action	Method of application	Duration of effect	*L. salmonis* life stages affected
Nuvan®, formely Aquaguard	Ciba-Geigy Agrochemicals Ltd., UK (Jones et al., 1992)	dichlorvos	organophosphate	disruption of neurotransmission (acetylcholinesterase)	bath	Approx. 2 weeks (Bron et al., 1993)	post-chalimus (Wootten et al., 1982; Rae, 1999)
Interox® Paramove 50™	Solvay Chemicals Inc., Belgium	hydrogen peroxide	inorganic compound	gas bubbles in haemolymph	bath: tarp or wellboat	immediate (Overton et al., 2019	adults and pre-adults (Davies and Rodger, 2000)
Aquaparox 50™	Alpha Chemical Limited, Canada	hydrogen peroxide	inorganic compound	gas bubbles in haemolymph	bath: tarp or wellboat	immediate (Overton et al., 2019)	adults and pre-adults (Davies and Rodger, 2000)
Salartect® 350 and 500	Brenntag, UK	hydrogen peroxide	inorganic compound	gas bubbles in haemolymph	bath	immediate (Overton et al., 2019)	chalimus, pre-adult and adult (Davies and Rodger, 2000)
Hydrolicer®	Hydrolicer Production AS, Norway	n/a	n/a	high water pressure physically detaches lice	treatment vessel	immediate	mobile and adults (Overton et al., 2019)

Table 1.1 continued next page.

Table 1.1 continued.

Common name	Main producer	Active ingredient	Drug class	Method of action	Method of application	Duration of effect	*L. salmonis* life stages affected
Thermal		n/a	n/a	detach after exposure to warm water (Overton et al., 2019)	treatment vessel	immediate	mobile (Overton et al., 2019; Nilsson et al., 2023)
cleaner fish: lumpptckers: *Cyclopterus lumpus*	n/a	n/a	n/a	predation	co-habit with salmon	variable (Imsland and Reynolds, 2022)	gravid female and some chalimus stages (PMRA, 2003)

The topical administration of treatments to farmed fish is achieved with the skirt, tarp or wellboat. A skirting method involving a temporary skirt around the circumference of a cage was employed but discontinued. Tarping is when the circumference and the bottom of a cage is temporarily enclosed with a tarpaulin. After administering the treatment, the tarp is removed and any unabsorbed chemotherapeutant is released into the surrounding water body (Burridge and Van Geest, 2014). The wellboat method of application involves the transfer of the fish from a cage into a large well (i.e., a container; the number of fish held in the well from the sea cage depends on the size of the fish and the volume of the well) in a boat where a treatment is applied and the fish are held for the duration of the treatment and then are transferred back to their original or different cage at the site. The in-feed method of application means that the chemotherapeutant is mixed in or applied to the surface of the pelleted feed and when the fish are fed, they receive the medication during feeding.

The chemotherapeutants released into the water column after removal of the tarp, discharged from a wellboat, and the uneaten and digested remnants of medicated feed may have unintended effects on the environment and animals in the surrounding area (Burridge et al., 2010; Burridge and Van Geest, 2014). A couple of examples follow. Hydrogen peroxide, after release from bath treatments, is considered to have low environmental risk and may affect crustaceans in the immediate vicinity resulting in mortality (Burridge and Van Geest, 2014). Azamethiphos has been associated with mortality of the European lobster (*Hommarus gamarus* [Linnaeus, 1758 {DecaNet, 2023}]; Parsons et al.,

2020) and sublethal chronic effects in adult American lobster (Burridge and Van Geest, 2014). The is also potential for persistence and bioaccumulation; azamethiphos may persist in the water column and bioaccumulate (Burridge and Van Geest, 2014). Regulating bodies (e.g., NB province [Burridge and Van Geest, 2014]), the Provincial Environmental Monitoring Program (New Brunswick Department of Agriculture Fisheries and Oceans, 2013), and the Pest Management Regulatory Agency (PMRA) of Health Canada (PMRA, 2003) evaluate chemotherapeutants and potential environmental adverse effects for the registration and use of chemotherapeutants for the management of sea lice infestations. Farmers, managers, and veterinarians use chemotherapeutants under strict government regulation and approval.

The alternative to chemotherapeutants is the use of non-medicinal, i.e., mechanical, methods to remove sea lice. The Hydrolicer® is a technology where the salmon are pumped through a pressure washer system that removes mobile lice (Overton et al., 2019). The thermal treatment method involves the transfer of the salmon from the cage to a wellboat treatment chamber with a set temperature of up to 34°C. At this higher temperature, the mobile lice are removed as they are inactivated, and the salmon are transferred back to a cage (Overton et al., 2019). The mechanical treatments may affect the welfare of the salmon as wounds to the dermis and gills, and mortality were observed (Overton et al., 2019).

Treatments vary in their methods of action, duration, and effectiveness on various stages of sea lice (Table 1.1). The method of action and effectiveness of

a treatment affects the duration and activity on a subset of the life stages of the sea lice. Other variables such as environmental conditions will also impact these treatment factors; for example, hydrogen peroxide bath duration is decreased at higher water temperatures, otherwise the hydrogen peroxide may kill the salmon (Overton et al., 2019). The toxicity of hydrogen peroxide to salmon at high water temperature highlights a concern for the welfare of the salmon being treated and consideration toward welfare and to reduce mortality was examined by Overton et al. (2019) for multiple treatments.

The effectiveness of a treatment has varied per the location and period (Torrissen et al., 2013) as a result of overuse or over-reliance (SEARCH, 2006; Aaen et al., 2015). Sea lice that survive and demonstrate reduced sensitivity to treatments transmit genes to their offspring resulting in ineffective treatments at mitigating sea lice infestations. Sea lice have developed resistance to chemotherapeutic (e.g., dichlorvos [Torrissen et al., 2013]) and thermal treatments (Ljungfeldt et al., 2017). Resistance can exist to multiple drugs simultaneously such as the *L. salmonis salmonis* multiresistant to organophosphates and pyrethroids (Fjørtoft et al., 2021). The reduced sensitivity to treatments were observed in the field, and monitoring this reduced sensitivity has associated challenges, such as the variability of sensitivity according to the life stage and sex of the sea louse, daily and seasonal variability, and many other factors as described by Westcott et al. (2018) for the optimization of a bioassay for emamectin benzoate efficacy.

The decision made by the farmer, manager, and veterinarian to apply treatments for the mitigation of sea lice infestation are complex as there are many factors involved as described above. With regards to the timing of the application of treatments, the decision can be directed by the sea lice counts. In certain areas, there are regulations of threshold level of sea lice per fish at which active mitigation strategies must be employed. In BC, a count of 3.0 mobile lice (equivalent to 0.9 AF per fish) is used as the threshold to treat during March 1 to June 30, which corresponds with the out-migration of wild salmonids in the area (Vormedal and Larsen, 2021). In Norway, there is a traffic light system which has two threshold values (Sandvik et al., 2021). There are typically regulations surrounding the actions to take if the threshold was exceeded; in BC, the farmer must apply treatments that reduce the infestation to levels under 3.0 mobile lice within 42 days (Vormedal and Larsen, 2021). In the Faroe Islands, the maximum level of sea lice is an absolute requirement rather than a threshold, which means that an exceedance of the threshold results in a penalty point, and these penalty points have an effect on the number of salmon the site can stock during the next production cycle (Vormedal and Larsen, 2021). The application of thresholds and absolute requirements may result in the biases in the monitoring data (Jeong et al., 2023).

1.5.1.3 Prevention strategies

Barrett et al. (2020) performed a meta-analysis of additional prevention strategies for sea lice management. A few interesting prevention strategies were the plankton mesh “skirt,” repellent and host cue masking, breed-selection practices

for louse resistance, and vaccination. There are different barrier technologies, such as the plankton mesh “skirt” that reduce the encounters between the parasitic copepodid and salmon host. Compounds (e.g., evaluation of garlic oil in Cobscook Bay, Maine, USA [R. Morefield, pers. comm.]) are released into the water column or added into feed (included in the term of functional feed) that repels or masks the hosts cues reducing the post-encounter infestation success (Barrett et al., 2020).

Selective breeding programs are another avenue for the management of sea lice infestation in Atlantic salmon (for a review of selective breeding in aquaculture species, see Houston, 2017). There is phenotypic and genetic variation in susceptibility to *L. salmonis* among strains of Atlantic salmon that may provide sources for selective breeding programs for improved resistance to infestation and disease (e.g., Gharbi et al., 2015; Robledo et al., 2018a; Valenzuela-Muñoz et al., 2023). The use of genomic selection has been applied and demonstrated to provide an advantage of host resistance to sea lice compared to pedigree selection programs (Tsai et al., 2016; Correa et al., 2017). One challenge of breed-selection for resistance to sea lice is that it may increase susceptibility to other diseases of concern. There is concurrent breed-selection for other diseases, such as AGD (Robledo et al., 2018b).

There is great interest in the development and use of a vaccine against sea lice because it would reduce the need for topical and oral treatments, effectively reducing a substantial portion of the costs and potential detrimental effects on the environment that is related to the mitigation of this pest. Vaccines for *S. salar* are

at the prototype stage to reduce the burden of *C. rogercresseyi* (Casuso et al., 2022) and *L. salmonis* (Swain et al., 2020). The prototype vaccines against *C. rogrecresseyi* were developed from the proteins cathepsin and peritrophin that are necessary to the louse for many functions including hemoglobin digestion and intestinal protection against host-pathogen interactions (Casuso et al., 2022). The prototype vaccines for *L. salmonis* were based on the housekeeping protein P0 (highly conserved among species [Swain et al., 2020]), and midgut proteins P30 and P33 (Contreras et al., 2020). The laboratory evaluation of these prototype vaccines provided modest reductions in sea lice abundances (e.g., reduction 42.5 % AF with egg strings by the peptide P0 [Swain et al., 2020]). Further development and research of the vaccines is considered necessary (Swain et al., 2020).

1.5.2 Estimated costs of sea lice management

In addition to the welfare and health issues directly affecting the fish, lice are also responsible for economic losses to the producers in the form of the salmon's reduced growth rate (Grimnes and Jakobsen, 1996) and downgrading of the harvested fish product (Mustafa et al., 2001). In Eastern Canada in 2001, farms infested with sea lice that did not apply sea lice treatments had an estimated economical loss of 0.45 US dollars per kg (Mustafa et al., 2001). The costs to the farmers for the management of the lice infestation includes the cost of the therapeutant (e.g., chemotherapeutant applied in feed), the use and maintenance of wellboats and other equipment, the time and resources from staff for the application, regular lice counts associated with monitoring activities (pre and post

treatment as required by the General Regulations), and others. The approximate yearly cost of sea lice to the aquaculture industry was 305 million Euros in 2008 (Costello, 2009b), which is equivalent to approximately 610 million US dollars in 2022[2]. More recently, Abolofia et al. (2017) estimated that infestation cost approximately 0.46 US dollars per kg of harvested biomass (equivalent to 0.55 US dollars per kg in 2022[3]).

Other costs associated to sea lice infestation are difficult to estimate. Environmental toxicity may occur secondary to the application of pesticides to fish in coastal waters that may impact non-intended inhabitants of the surrounding area. There is potential for transmission of the pest from farmed salmon to their wild counterparts (Murray and Gillibrand, 2006; Middlemas et al., 2013). The sea louse, its mitigation treatments and related environmental concerns all impact the social perception and acceptance of the aquaculture industry (Torrissen et al., 2013; Flaherty et al., 2019; Kraly et al., 2022). Sea lice and the environmental concerns are part of the reason for the development of land-based production sites (Song et al., 2019).

1.5.3 Global and local trends

The current trends in the mitigation of sea lice includes synchronous and single generation zonal management that is informed by regular monitoring of lice

[2] Calculated using the 2008 average yearly conversion rate for Euros to USD (1.47) and the USA average inflation rate from 2008 to 2022 (1.36; U.S. Bureau of Labor Statistics, 2023).
[3] Calculated using the USA average inflation rate from 2017 to 2022 (1.19; U.S. Bureau of Labor Statistics, 2023).

counts and hydrodynamic and particle-tracking models (e.g., Sandvik et al., 2016b). There is also a movement away from chemotherapeutants for the preferential development and use of mechanical and non-chemical treatments.

One challenge that applies specifically to BMA 1 is the neighbouring area with active sites in the state of Maine, which belong to another jurisdiction. No data of sea lice counts are available from the salmon producers in that area (R. Morefield, pers. comm.). However, from the analysis of drug and pesticide utilization in USA aquaculture performed by Love et al. (2020), we can deduce that this area also has sea lice; for example, 28 % of the production cycles from 2014-2017 applied emamectin benzoate.

The treatments listed in Table 1.1 included those no longer in use, such as cypermethrin, dichlorvos and diflubenzuron which have not been used since 2009. A breakdown of the use of specific treatments for the area is described in Chapter 2 between 2009 and 2018, and Chapter 6 between 2018 and 2021. In general, NB has followed the trend toward non-chemical treatments, favouring Hydrolicer® and thermal treatments. The area has not benefitted yet from bio-physical modelling input to aid in the management of sea lice, but the veterinarians, farmers and researchers have worked together to develop and maintain a monitoring system for *L. salmonis,* and database of sea lice counts was instituted in 2009.

1.6 Monitoring and surveillance of sea lice

Since the 1990s, managers, farmers, and veterinarians began counting sea lice on a small sample of fish from farms semi-regularly in order to help with decisions for the application of management strategies. Monitoring and surveillance can help identify rises in sea lice that need treatment and anticipate trends of sea lice based on environmental predictors (e.g., Revie et al., 2002, 2003). Norway and the Faroe Islands have active surveillance systems for sea lice infestations as mitigation treatments must be applied if a threshold is exceeded. In NB, there is no threshold, but there is a monitoring system of the sea lice with documentation of counts into a data management system. These monitoring systems produce data that can be visualized for trends in time and can be used in the construction of models as was achieved in Chapters 3 to 5. The quality of the data output from the monitoring and surveillance systems are dependent on the entire system from the person counting, to the farmer/manager, veterinarian, and data-manager. The data-analyst/researcher are dependent on the previous persons and the data produced to generate accurate results.

The monitoring practices engender a lot of data and to manage these, counts of sea lice and records of treatments are often aggregated into databases. In Chile, there is Sernapesca's Sistema de Fiscalización de la Acuicultura (SIFA) and Informe Ambiental para la Acuicultura (INFA) databases for weekly counts of *C. rogercresseyi*. Scotland has a new web-based database (“Sea Lice Data” under http://aquaculture.scotland.gov.uk/) which contains mean weekly counts of AF *L.*

salmonis. Norway has the web based BarentsWatch database (https://www.barentswatch.no/en/) which contains aggregated weekly counts of AF *L. salmonis* as well as mobile and stationary lice. In Canada, there is the user-protected Fish-iTrends© database (https://www.fishitrends.com) for the East Coast and on the west coast, the Department of Fisheries and Oceans Canada audits the reported values directly.

The frequency of monitoring counts varies as per the regulation for the area and country. In Chile, the regulation stated biweekly counts were to be submitted for the period 2008-2012 (Bravo, 2003; Hamilton-West et al., 2012) and since then it has been increased to a weekly submission (Kristoffersen et al., 2013). In Eastern Canada, it is the purview of the provinces to regulate commercial fisheries and aquaculture. The Atlantic salmon aquaculture in the Bay of Fundy, NB, is under the authority of the Province of New Brunswick and sea lice monitoring occurs under the General Regulations 91-158 (Government of New Brunswick, 2020).

In NB, the submissions of sea lice counts must be made weekly and outlining treatments as part of an integrated sea lice management program starting after 2008 (Chang et al., 2011). The program outlines standardized sampling protocols and reporting of counts (Elmoslemany et al., 2013). The regulation states that a minimum of 5 salmon must be counted for sea lice in 6 or more cages per site weekly (based on work from Treasurer and Pope, 2000), and before and after a treatment, if the temperature is above 5 °C (Government of New Brunswick, 2020). At temperatures below 5°C, counting events are mandated monthly

(Government of New Brunswick, 2020). These data are collected into the Fish-iTrends© web-based data management system that was implemented in 2009 and is operated by the Centre for Aquatic Health Sciences (CAHS) at the Atlantic Veterinary College (AVC) of the University of Prince Edward Island, Prince Edward Island, Canada (for additional information, see Gautam et al., 2016, 2017). The Fish-iTrends© is supported by the industry associations in the provinces of NB, NS and NL. Additional information on the history of sea lice counting in NB is reviewed in Chapter 2.

1.7 Connectivity and infestation pressure

1.7.1 General concepts of dispersal and connectivity

The concept of connectivity was first developed to study the movements of animals and plants (organisms) in terrestrial environments (i.e., 'landscape connectivity' [Tischendorf and Fahrig, 2000]) but has since been employed in marine ecology, particularly for the study of benthic marine species that have a pelagic larval stage (Siegel et al., 2003; Cowen and Sponaugle, 2009). The concept of connectivity is equally applicable to viruses (e.g., ISAV [Gautam et al., 2018]) and bacteria (e.g., SRS transmission in Chile [Rees et al., 2014]) as it is to parasites (e.g., *L. salmonis* [Adams et al., 2012]).

Dispersal is defined as the movement of an agent away from the source (outflux), its progenitor, to a destination (sink or influx) for settlement (e.g., benthic species such as oysters [Cowen and Sponaugle, 2009]) or transmission to a new host (e.g., sea lice and salmon). The transmission of infestation of sea lice occurs through the horizontal transmission of the pelagic lice through the water and it

develops into the parasitic copepodid. Alternative modes of transmission as with vectors (e.g., sea lice carrying ISAV) can also be applied within the concept of connectivity. The exchange of an agent among sites resulting from dispersal is dependent on multiple factors in a spatiotemporal context; this is the concept of connectivity (Cowen and Sponaugle, 2009). The agent moves within the environment with respect to its behavioural and physical needs and this is dependent on the characteristics of the environment that contains elements that are accessible or not (ecological resource: Le Corre et al., 2012), and where it is accessible, may contain its preferred habitat (i.e., habitat patch: location that is preferred by the organism for biological or behavioural reasons [Girvetz and Greco, 2007]). The agent's movement may be impeded or facilitated by its environment as it moves between habitat patches (Tischendorf and Fahrig, 2000; Cowen and Sponaugle, 2009; Le Corre et al., 2012). The marine environment is complex; the water currents, presence and distances of neighbouring salmon farms, salinity, temperature, and many other factors may affect the level of *L. salmonis* infestation on an individual salmon. One factor absent in NB but present in other areas that can affect infestation is the presence of a potential reservoir wild population (e.g., sea trout in northeastern Atlantic countries [Middlemas et al., 2013]), or even specific reservoir host (host for *L. salmonis* that does not suffer from the disease), such as the three-spined stickleback (*Gasterosteus aculeatus* Linnaeus, 1758 [Froese and Pauly, 2023]) in BC for *L. salmonis oncorhynchi* (Fast and Dalvin, 2020).

The habitat patches of the sea lice are different for the life stages. The pelagic lice are present in the water column and have a continuous habitat patch (i.e., the top layer of coastal waters). The parasitic stages are limited to the farm sites, its habitat patch, although the mobile life stages may transfer among salmon in a cage and among cages at a site (Jaworski and Holms, 1992; Costello, 2009a). Considering the parasitic stages, a collection of farm sites in close proximity, or as defined within a BMA, are a subpopulation, and the collection of the BMAs in the Bay of Fundy, NB, is a metapopulation (Figure 1.3). The definition of metapopulation is strictly defined in marine ecology such that there is limited dispersal among metapopulations (Sale et al., 2006; Van Nouhuys, 2016); in other words, there is limited dispersal of sea lice between NB and another metapopulation of sea lice (e.g., Norway). It should be noted that the *L. salmonis* population shares genetic markers throughout the whole of the Northern Atlantic Ocean (Todd et al., 2004; Boxaspen, 2006; Glover et al., 2011; Besnier et al., 2014), but the concept of metapopulation does not mean that there is zero dispersal among the metapopulations, but that the process of mixing/transmission is rare (Van Nouhuys, 2016). In NB, and other areas, the definitions of the habitat patch, subpopulation and metapopulation (Figure 1.3) were not determined through ecological or genetic analysis but serve to aid in understanding the connection of farms in the greater environment. In truth, there may be a different pattern to the populations of sea lice in the Bay of Fundy, NB, for example the Passamaquoddy Bay and Grand Manan Island may be metapopulations with rare sharing of sea lice individuals. The definition of these

divisions of population would have to be evaluated, likely through genetic or epigenetic analysis, and for this to be achieved, a unique marker would have to be identified. A successful demonstration of subpopulations in the area may present the opportunity to apply better coordinate mitigation strategies (Tully and Nolan, 2002; Aldrin et al., 2013; Adams et al., 2015; Cantrell et al., 2019) and apply some control to sea lice movements, such that is described as a “firebreak” (Green, 2010; Samsing et al., 2017, 2019).

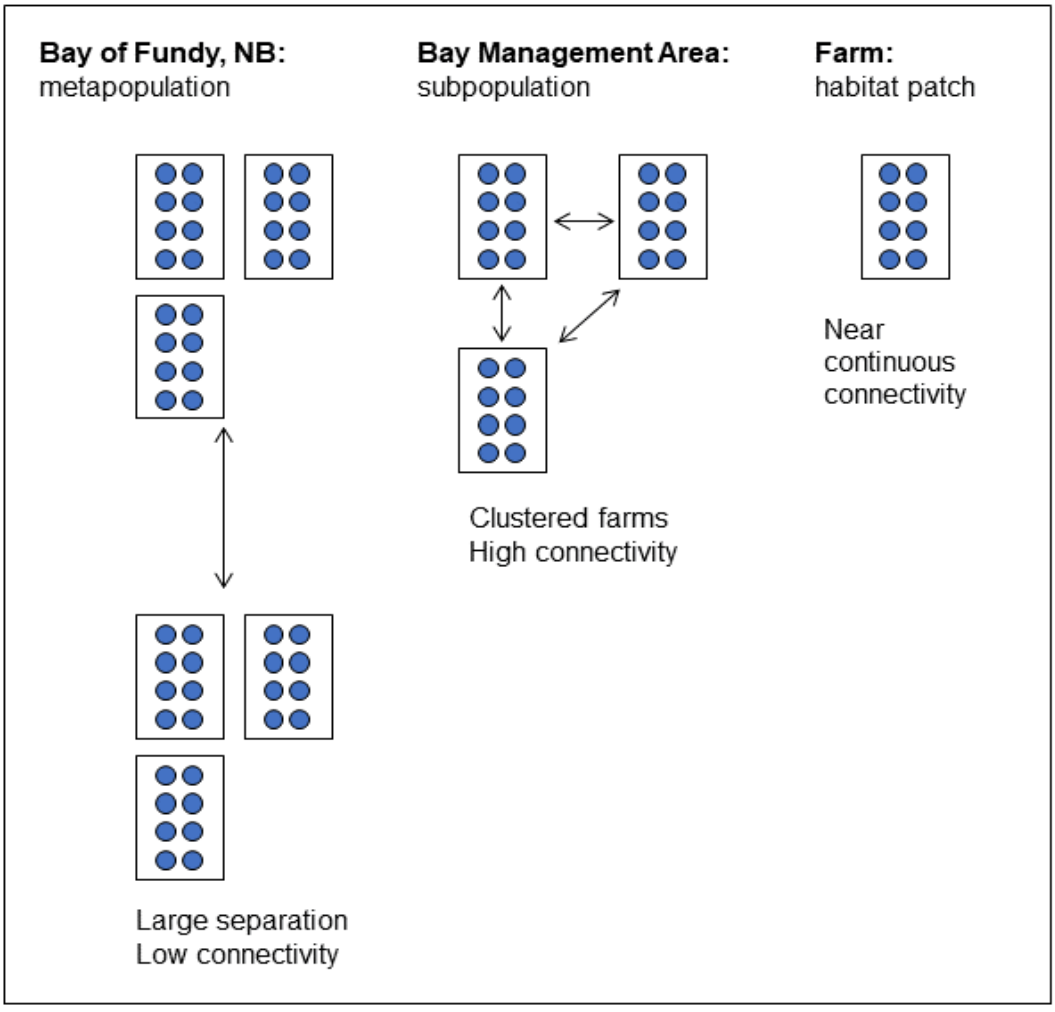

Figure 1.3. Schematic of the concepts of metapopulation and subpopulations of Atlantic salmon farm sites in the Bay of Fundy, New Brunswick, Canada, that aid

in the understanding of the concept of connectivity among farms by the parasite *Lepeophtheirus salmonis*.

It should be noted that the term connectivity can be used in other ways, too. It can be understood as the process that "drives the exchange of individuals and alleles (i.e., connectivity) among populations with many marine metapopulations" (Sale et al., 2006; D'Aloia et al. 2015). The term 'hydrodynamic connectivity' is used to describe the connection of farm sites through the movement of water (e.g., Gautam et al., 2018; Bravo et al., 2020). In the study of the concept of connectivity, there is also the refinement of the concept toward structural and functional connectivity as described in Tischendorf and Fahrig (2000).

The resulting dispersal data for the agent among habitat patches can be displayed as a square matrix (e.g., Gautam et al., 2018; Bravo et al., 2020; Aleynik et al., 2022) showing measures of connectivity from every site to every site, where the rows and columns are the site identifiers, and the values represent the movement of the organism or agent between each pair of sites. Generally, rows and columns represent the source/origin and sink/destination/receiver, respectively. The values for origin to destination and destination to origin may be the same with resulting symmetrical values across the diagonal, or the values for origin to destination are different than for destination to origin with asymmetrical values across the diagonal of the matrix. A connectivity matrix can be a matrix of distances among sites, or it can be the probability of the exchange of the agents between pairs of habitat patches (Cowen and Sponaugle, 2009). Tully and Nolan (2002) defined connectivity among farms as a value between 0 (no connectivity) and 1 (continuous

connectivity). In studies that utilized bio-physical models, the matrix entries may consist of a probability that a particle emitted from a source site reaches a sink site (Cantrell et al., 2020) or it may represent a transfer rate among nodes (Bacher et al., 2016). Harrington et al. (2022) proposed that next-generation matrices be used for the study of marine metapopulations to standardize connectivity matrices across various modelling frameworks and could give results toward the determination of the persistence of sea lice populations. The next-generation matrices are produced from the conversion of differential equation models for the development of sea lice into generation time (i.e., number of new individuals produced in each age), and the sum of the columns of the matrix can be used to measure the individual contribution of habitat patches to a metapopulation over a generation (Harrington et al., 2022).

1.7.2 Dispersal distance of sea lice

1.7.2.1 Biological variables in the dispersal of sea lice

The dispersal of sea lice is dependent on multiple factors. The sea louse must survive from hatching, reach a host in the copepodid stage despite the challenges of poor swimming capability, currents, tides, and other water movements, survive the limited duration of the yolk sac energy reserves (Brooker et al., 2018), and survive predation (Connors et al., 2008). Then the sea lice must parasitize a host, develop to sexual maturity, and achieve reproduction for there to be persistence of the infestation. An effective dispersal occurs when there are sufficiently large numbers of sea lice to allow detection to be labelled as a successful parasitic infestation.

Probabilities can be assigned for stages of development, survival, and mortality, and for an effective dispersal event (e.g., Harrington et al., 2023). Toorians and Adams (2020) estimated probabilities for successful dispersal of *L. salmonis*. There was a 1 % to 3 % probability of successful dispersal within a site (i.e., self-infestation) from a population of *L. salmonis* between 8 and 14 °C, and among sites, population density of sea lice increased over time if the site influx was above a threshold of 0.0075 (Toorians and Adams, 2020). An alternative to probabilities is rates which can be assigned to the various stages of survival and development of the sea lice. A mortality rate can be assigned to the pelagic life stages of sea lice (e.g., Revie et al., 2005) and this rate will affect the distance covered during the movement of the sea lice among habitat patches.

Environmental variables such as the water temperature and salinity will impact the rate of development and rate of mortality (Boxaspen and Naess, 2000; Revie et al., 2005; Groner et al., 2016a; Samsing et al., 2017), and will impact the probability of successful dispersal (Toorians and Adams, 2020). A sea louse that develops more rapidly due to high water temperature will expend its energy reserves more rapidly and may risk dying if it does not find a suitable host in time (Brooker et al., 2018).

1.7.2.2 Estimation of the dispersal distance of sea lice

Because dispersal is part of understanding the connectivity of sea lice in an area, there have been multiple studies with the goal to estimate the dispersal distance of sea lice (e.g.,Gillibrand and Willis, 2007; Amundrud and Murray, 2009; Stucchi

et al., 2011; Asplin et al., 2004, 2011, 2014). There are two major ways of achieving this goal. The first method is biological observation. This can be conducted through several ways. Copepodid can be sampled in the water column at a distance from sites with the use of trawls or other plankton sampling equipment and techniques (e.g., Penston et al., 2004, 2011; Skarðhamar et al., 2019). This provides very strong empirical evidence of where free-living stages of lice can be found in coastal waters; however, a major disadvantage of this method is the difficulty in capturing the pelagic life stages as they are present in very low concentrations in the water column (Penston et al. 2004, 2011; á Norði et al. 2015, 2016; Skarðhamar et al., 2019). Another way is to estimate this by counting attached sea lice on fish. This can be performed on the farm site (e.g., Gautam et al., 2016; Aldrin et al., 2017; Gislason, 2017; Kristoffersen et al., 2013, 2014, 2018; Zalcman et al., 2021), on fish in sentinel cages (e.g., Adams et al., 2012; Pert et al., 2014; Sandvik et al., 2016b, 2021) and on wild fish (e.g., Rees et al., 2015). Sampling sea lice in the water column or on fish in sentinel cages or wild fish are methods that are logistically demanding and expensive (Skarðhamar et al., 2019). Sandvik et al. (2016a, 2020) have utilized sentinel cages to capture the copepodids that move among farm sites. The salmon were removed and euthanized after 3 weeks at sea and the lice are counted for the number of fish collected and an infestation level for the cage was estimated (Sandvik et al., 2016b). This involved the use of additional equipment such as cages and boats, smolt that are not reared for harvest and sale, and scheduling a team for the setup and frequent collection of the salmon for the estimation of the

infestation level. The estimation of infestation levels using sentinel cages may be an underestimate of the true abundance of sea lice on wild fish (Ulgenes, 2018).

The use of monitoring counts on farmed salmon does not give a direct answer to the question of dispersal of sea lice. A sea lice count on a farm site does not differentiate the sea lice infestation from within the site (i.e., self-infestation) and from neighbouring sites. The dispersal distance can be estimated using statistical models. Statistical models were developed from sea lice monitoring counts and utilized to estimate the dispersal distance of sea lice (e.g., Kristoffersen et al., 2013; Elghafghuf et al., 2020). The dispersal distance was estimated from a range of kernel-weighted intensity of mean-reported abundance of AF sea lice abundance and the comparison of the model best fit (e.g., Kristoffersen et al., 2013; Elghafghuf et al., 2020).

Numerical simulations can also be used to estimate dispersal distance, and connectivity, and this is generally achieved with the use of particle-tracking hydrodynamic models. Hydrodynamic models model the physical properties of water movement in three-dimensional space in coastal waters. Simulated particles are released from sites within the hydrodynamic model and their movements and concentrations are observed. The simulated particles can represent sea lice and may be assigned biological parameters such as survival rates (e.g., Amundrud and Murray, 2009; Asplin et al., 2014; Salama et al., 2016; Kragesteen et al., 2018). The application of parameters that represent the biological properties of sea lice is an advantage (e.g., Amundrud and Murray,

2009; Salama et al., 2016) and may affect the estimations of the connectivity among sites (Cowen et al., 2006).

Relevant to the study area, were the application of zones of influence or water exchange using a 5 km radius circle centred on the farm site, and the application a three-dimensional particle-tracking model to estimate tidal excursion areas (Chang et al., 2005a,b, 2006a,b, 2007; Page et al., 2005). The interactions among farms identified through the overlaps of the buffer zones and the model-derived tidal excursions were important for the determination of the BMAs (Chang et al., 2005a,b, 2006a,b, 2007; Page et al., 2005). Dispersal of sea lice was also estimated through the measurement of mean flow rates around sites (Nelson et al., 2017). Nelson et al. (2017) estimated a dispersal distance range of 8 km in Passamaquoddy Bay, NB.

There are multiple estimates for dispersal distance. Using an empirical model, Elghafghuf et al. (2020) estimated a dispersal distance of 12.5 km for the region of Grand Manan, NB, a part of the study area. For other aquaculture areas and utilizing various modelling approaches, estimates of dispersal distances were 6 km (Amundrud and Murray, 2009; Salama et al., 2016), 7-12 km (Gillibrand and Willis, 2007) and even as far as greater than 100 km (Asplin et al., 2014). A common range for dispersal is 20-30 km (Krkošek et al., 2005; Kristoffersen et al., 2013; Asplin et al., 2011, 2014; Salama et al., 2013, 2016; Aldrin et al., 2019). These estimates likely vary due to the spatiotemporal variability, environmental characteristics such as the geometry of the coast (e.g., Adams et al., 2015) and estimation approach. This information is relevant not only because

of its importance for understanding connectivity, but because it may inform disease management practices (Groner et al., 2016b).

1.7.2.3 Dispersal kernel

One statistical method to model dispersal of sea lice is to create weights that decay over space, such as a kernel-weighing of abundance (e.g., Kristoffersen et al., 2013; Elghafghuf et al., 2020). Kernel weighting abundance of sea lice in statistical models is usually done with a Gaussian kernel density estimate on seaway distance (e.g., Aldrin et al., 2013, 2017; Cantrell et al., 2018; Elghafghuf et al., 2020). This kernel weighting of distance is the dispersal kernel, and it is defined as a probability density function that can be integrated to yield the probability of successful dispersal over a given distance (D'Aloia et al., 2015). Dispersal kernels are an important concept in landscape ecology and have been reviewed for other species (Chiswell, 2012; Nathan et al., 2012).

The Gaussian distribution was selected as the transformation for seaway distance of choice in multiple studies (e.g., Kristoffersen et al., 2013). The Gaussian distribution applies symmetrical radial weights for any given distance around a farm site (Nekouei et al., 2018). There are multiple arguments for the use of the Gaussian distribution for the dispersal kernel of *L. salmonis*. In terms of biology, the parasitic copepodid decrease exponentially away from a site as the copepodid are moved by currents and their ability to parasitize a host is limited by the energy reserves of the yolk sac (Brooker et al., 2018). Previous studies used an exponential transformation for the seaway distance, like in

studies of viruses (Aldrin et al., 2011) and this transformation was applied to sea lice thereafter (Jansen et al., 2012). Aldrin et al. (2013) evaluated several distributions for the transmission kernel of *L. salmonis* and found that the curve was steeper than the exponential curve at short seaway distances and levelled off at intermediate distances. More generally, Siegel et al. (2003) examined planktonic larval duration in a nearshore current using numerical simulations. These authors concluded that a Gaussian distribution had an excellent fit for different experiments of planktonic larvae distributions with short and long planktonic larval duration, as defined by a short or long interval to settle and short or long ability to remain in the plankton (Siegel et al., 2003). The decision to select a Gaussian kernel was also based on the link with the traditional advection-diffusion approaches, which are numerical models that represent the complex mixing of oceanic waters (Siegel et al., 2003).

For the case of the sea lice in the Bay of Fundy, NB, the sea lice dispersal kernel may be Gaussian as the site has large tides with anisotropic patterns and Chiswell (2012) estimated that as the ocean becomes more dispersive the kernel approached a Gaussian distribution. However, the author noted that this was counter-intuitive, and other research (Cowen et al., 2006; Huret et al., 2010) have hypothesized that anisotropy of ocean currents results in asymmetry in dispersal kernels. Other distributions of movement exist and are studied for other agents, such as the pollen of the Eurasian black poplar (*Populus nigra* L. [Niggemann et al., 2012]) and planktonic larvae of the benthic red snapping shrimp *Alpheus immaculatus* (Knowlton and Keller, 1883 [DecaNet eds., 2023]) (Chiswell, 2012),

but these are studies consider larger study areas than the coastal study area; we can expect for these species a distribution of movement with an isotropic decay. The estimation of dispersal kernels and connectivity depend on the biological and physical constraints of the model (Cowen et al., 2006). The hydrodynamic and particle-tracking models to simulate the dispersal of particles are likely a better approach than the simple Gaussian distribution; but where a hydrodynamic model is unavailable, a Gaussian distribution may be suitable for our modelling purposes.

The movement of sea lice away from a site may be non-Gaussian due to currents and other environmental factors. Chiswell (2012) concluded that dispersal kernels cannot be assumed as Gaussian distributions as they depend on several factors, such as the amount of time that the species has to settle, which in terms of sea lice, would mean that the period from hatching to copepodid attachment and limited by the yolk sac reserves and development rates which are dependent on environmental variables of temperature and salinity. The dispersal kernel can be estimated only with knowledge of the furthest distance of dispersal (term used is long-distance dispersal events) otherwise the tail of the distribution is unknown (D'Aloia et al., 2015). Chiswell (2012) also found that if the variability of the larval duration was large (i.e., the variation in the probability of settling/attachment varies for early vs. late arrivals to a host, and not the time from hatching to settlement) then the dispersal kernel approached a non-Gaussian distribution. Aldrin et al. (2013) modelled the abundance of sea lice and applied a Box-Cox transformation to allow for variation

in the shapes of the distance function. These authors found that the estimate had a steeper curve than the exponential distance function (Aldrin et al., 2013). This author did not find any other studies that estimated the dispersal kernel of sea lice at the time of writing. The application of the Gaussian distribution in the models produced in Chapters 3 to 5 may overestimate the dispersal distance if the true distribution is steeper as observed by Aldrin et al. (2013).

The empirical determination of the kernel dispersal for sea lice would require an improvement of the detection of copepodid in the water column and an extensive sampling regime to be able to make an estimate of the distribution. Another method for the estimation of the dispersal of sea lice is the biophysical model, i.e., where biological parameters are applied to particles during simulations within a hydrodynamic model. Where biophysical models have previously been developed, the evaluation of alternate dispersal kernels may be evaluated. In the Bay of Fundy, NB, there are no biophysical models available at this time, but the tidal excursions data provided by Chang and Page (Chang et al., 2005a,b, 2006a,b, 2007; Page et al., 2005) may provide additional information into the dispersal kernel of sea lice from sites.

1.7.2.4 Use of dispersal and connectivity knowledge to inform disease management

Disease management may be informed by knowledge of the dispersal of sea lice and the connectivity among sites. With the knowledge of the dispersal distance and which sites are connected, managers can make informed decisions to

effectively control/mitigate the transmission of that disease (Sandvik et al., 2016a). The dispersal distance information can be applied by separating sites to distances greater than the effective dispersal distance, i.e., the pelagic sea lice that exceed that distance are not detectable and do not produce a successful infestation. The epidemiologically significant distance is a distance where a higher proportion of sea lice on average are transmitted (Adams et al., 2012; Salama et al. 2016, 2018) and it is within the effective dispersal distance. A key term there is ‘on average’ because a biophysical model for Loch Linnhe, Scotland, demonstrated that some close sites may not be connected due to specific current patterns and may be connected to sites further away (Salama et al., 2018). There is also temporal variation in the dispersal of sea lice in response to environmental variables such the effect of spring freshet (i.e., large freshwater input from rivers that is secondary to the snow melting and higher precipitation volumes in the spring) which resulted in a decrease in the survival rate of the simulated particles that represented sea lice in Cantrell et al. (2019).

Information about dispersal distance is also related to the possible mitigation strategy to space out sites and remove intermediary sites (‘firebreaks’) so that the sites are at epidemiological significant distances so that transmission is impeded (Green, 2010; Samsing et al., 2017, 2019). This would organize sites into clusters, and multiple studies have identified the organization of clusters using biophysical models (Stene et al. 2014; Adams et al. 2012, 2015; Patanasatienkul et al., 2015; Murray and Gubbins 2016; Samsing et al. 2017; Cantrell et al. 2018; Salama et al., 2018).

The information that is provided by estimates of dispersal of sea lice is important when trying to better understand the potential interaction/infestation of wild fish (e.g., Krkošek et al., 2005, 2011). There are concerns regarding the potential of farms to amplify the parasite and result in a higher infestation pressure for wild fish (e.g., Kristoffersen et al. 2018; Nekouei et al. 2018) as farmed salmon account for the majority of sea lice produced (Dempster et al., 2021). Krkošek et al. (2005, 2011) applied advection-diffusion submodels for the dispersal of sea lice transported away from the farm sites as they overlay the migration routes of wild salmonids. A challenge to the incorporation of wild salmonids in models of sea lice is that farms are set in location, so they are considered a 'point source,' where wild salmonids are motile and follow a migration route from a river to the ocean, they are a 'distributed source' (Krkošek et al., 2005).

1.7.3 Connectivity measures

The term connectivity measure was employed in the field of landscape ecology (e.g., Kindlmann and Burel, 2008) to describe a matrix that represents the connectivity among sites. This matrix is an array of measures, and the measures can be true distances such as seaway distance or another variable such as a measure that incorporates bathymetry.

Seaway distance is a common measure for connectivity: Grand Manan, NB (Elghafghuf et al., 2020), Broughton Archipelago, BC (Rees et al., 2015), Norway (Aldrin et al., 2013, 2017, 2019), and Chile (Kristoffersen et al., 2013). This connectivity measure is simple, where the distance between two sites is the shortest distance while remaining within a water environment, as sea lice

movement is impossible on land (in landscape ecological terms land is a hard barrier for sea lice [Girvetz and Greco, 2007]). Euclidean distance (shortest distance between the two farms irrespective of the landscape) is not typically employed because of the hard barrier which is land.

In landscape ecology, there are other measures for the movement of agents (e.g., Wang, 2020). And some are not as computationally expensive as hydrodynamic and particle-tracking models. There is topographic distance that measures a distance among two sites while accounting for the topographic relief (elevation of land) (e.g., Bista et al., 2021). These authors defined the dispersal distance as the topographic distance in km such that the distance accounted for the changes in elevation as the Himalayan red pandas, *Ailurus fulgens* (F. Cuvier, 1825), moved within the landscape (Bista et al., 2021). While it may be unconventional to apply landscape ecology techniques to the marine environment, these techniques could be helpful in describing connectivity and dispersal of sea lice because bathymetry (depth) influence water movements. Other measures extrapolated from bathymetric data, such as the volume of water and the percentage of water adjacent to sites, may help explain connectivity among the sites in the study area. These measures of connectivity can be easily calculated from currently available maps (DFO bathymetry map) using freely available geographic information system (GIS) software and R. This is especially useful when hydrodynamic and particle-tracking models are not available or feasible. Connectivity measures derived from bathymetry data using landscape ecology techniques are described in Chapter 4.

1.7.4 Internal and external infestation pressure

1.7.4.1 General concept and terms

Infestation pressure is a measure that represents the dose of exposure of parasitic stages of sea lice to potential salmon hosts. Infestation pressure can occur within a farm (internal infestation pressure [IIP]), where the parasitic copepodid will attach to a host within the same farm site as its progenitor. If the copepodid originated from another site, this infestation is called external infestation pressure (EIP). The wording 'infestation pressure' is referred to as '*infection* pressure' by others (Aldrin et al., 2013; Kristoffersen et al., 2013, 2014), but the term 'infection pressure' should be reserved for viruses, bacteria, and endoparasites, while infestation pressure for ectoparasites. There are also other terms used to refer to the internal and external infestation pressures. Kristoffersen et al. (2013) referred to "internal (farm) and external (regional) sources of sea lice infection." Earlier still, Tully and Nolan (2002) used the term 'colonization pressure' to refer to the similar concept of EIP regarding sea lice.

There can be another source of infestation external to the site and that is the contribution of copepodids from wild fish or escaped farmed salmonids (Aldrin et al., 2013). The contribution from this external source was deemed small in that study (Aldrin et al., 2013) and is typically ignored in most studies (e.g., Johansen et al., 2011; Aldrin et al., 2017). One consideration of the EIP contribution from wild fish may be its seasonality as the wild fish migrate near salmon farms (Wallace, 1998; Revie et al., 2005).

1.7.4.2 Estimating internal and external infestation pressure

Multiple life stages of the sea louse can be considered to calculate infestation pressure. The variable of time is also part of the consideration as it is possible to use different life stages from different time periods; for example, the AF from three weeks ago will explain the observed AF observed today (Figure 1.4). Previous studies have used lags of 2 weeks (Kristoffersen et al., 2013, 2018; Aldrin et al., 2019) and 1 month (Aldrin et al., 2013). Therefore, infestation pressure was modelled as a predictor in multiple ways in a multitude of studies (e.g., Aldrin et al., 2013; Kristoffersen et al., 2013, 2014, 2018).

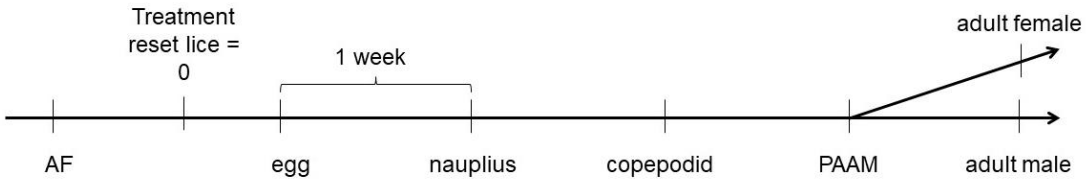

Figure 1.4. Illustration of a timeline for the development of *Lepeophtheirus salmonis*. The treatment reset for lice reflects the assumed decrease in sea lice abundance secondary to the application of a treatment.

In general, these authors estimated the IIP as the abundance of AF multiplied by the number of fish at the farm site (Aldrin et al., 2013; Kristoffersen et al., 2014, 2018). This simple calculation of IIP can be clarified by adding an estimated number of louse eggs produced adjusted with development time in degree days according to the temperature and a daily mortality rate (Kristoffersen et al., 2018). It can also be by adjusted using temperature dependent variables for fecundity and mortality (Kristoffersen et al., 2014; Aldrin et al., 2017).

The EIP can be calculated as the summation of contributions of the abundance of sea lice multiplied by the number of fish sampled adjusted by the Gaussian kernel-weighed seaway distance with a lag (Aldrin et al., 2013; Kristoffersen et al., 2013, 2018). Kristoffersen et al. (2014) calculated the EIP differently, where it was the difference of the total infestation pressure and internal infestation pressure; the total infestation pressure was the sum of the IIP for the sites within a range of 100 km and weighted with a Gaussian kernel to the seaway distance. These authors also produced a simpler estimate of EIP (termed the external contributing adult females [CAF]) as the reported abundance of AF from neighbouring sites with a kernel-weighted seaway distance (Kristoffersen et al., 2014). The simpler calculation of EIP (external CAF) produced a model with a poorer fit (Kristoffersen et al., 2014). In Aldrin et al. (2017), the authors wanted to adjust the measure of the seaway distances to simulate variation in the dispersal in a farm and time dependent manner; they achieved this by applying a modifying factor that was farm and time dependent. The EIP calculation was improved upon again in Aldrin et al. (2019) by adding parameters for non-linear dependencies from the contributions from neighbouring farms and the number of fish at previous lags. These adjustments to the EIP allowed for the assessment of the effects of adding farm sites and increasing the number of fish that are potential hosts for the sea lice (Aldrin et al., 2019). Elghafghuf et al. (2018) used state-space models to evaluate different methods for the estimation of the EIP: 1) fixed lag (e.g., Kristoffersen et al., 2013 as described above), 2) degree days, 3) quantitative life cycle, and 4) weighted average lag structure approaches were

evaluated against each other. The fixed lag approach provided a simple method for the calculation of the EIP and produced a model with low AIC compared to models with EIP and IIP calculated with the other approaches (Elghafghuf et al., 2020). Kragesteen et al. (2021) estimated EIP on a per-farm basis for the first 150 days of the production; their results had high variability of EIP among farms (0.002 to 0.1 lice per salmon per day) and they demonstrated that EIP was dependent on the mean level of gravid female lice 15 to 45 days prior. The calculations EIP per-farm utilized the number of fish at the farm and this is not possible where there is a lack of data on fish numbers (e.g., Fish-iTrends database). The lack of data on fish numbers would also affect the weighting of contributions from different farms to EIP as done in other studies (e.g., Kristoffersen et al., 2013).

The estimation methods for the EIP in frameworks that employ hydrodynamic and particle-tracking models are different to the methods described above. The EIP was calculated as the proportion of surviving copepodids to the total number of particles released from the source site (Adams et al., 2012; Salama et al., 2018). The EIP was estimated as a daily density of particles for each grid section of the grid cell for the study area (Cantrell et al., 2018). Sandvik et al. (2016) integrated the densities of particles per grid cell over relevant time periods and translated the result to a density of the parasitic copepodids (particles) per m^2.

1.7.4.3 Comparing internal and external infestation pressure

It is relevant to the management of sea lice to know if the infestation pressure is stronger within the farm site (IIP) or from neighbouring sites (EIP); thus, the pressures are often compared in order to make recommendations towards management practices to reduce the pressures and abundance of lice. Where the IIP is larger than the EIP, management practices towards the control of sea lice at the site are important and could be achieved through near synchronous treatment of all cages with treatments to reduce the abundance of lice.

Aldrin et al. (2013) concluded that the EIP had a larger effect than the IIP to the level of two thirds of the additive contribution; the third contributor to the infestation was an external unknown source, such as wild fish. The situation of greater EIP compared to IIP was hypothesized as 1) the survival of pre-adults and adults was greater for the consecutive monitoring counts, 2) the produced eggs hatched and developed into parasitic copepodids that attached to salmon within the site where they were produced, 3) there are few parasitic copepodids arriving from neighbouring sites as transported passively for the period examined (Aldrin et al., 2013). The points 1 and 2 may be the result of a positive feedback system, which means that the IIP increases continuously as the produced eggs develop into adults that produce more eggs in an exponential cycle (Adams et al., 2012).

At stocking, there is no IIP as the smolt were free of sea lice as they originated from a freshwater environment at the hatchery (Kristoffersen et al., 2014; Aldrin

et al., 2013, 2017). The EIP is particularly relevant for the salmon at stocking as it will drive the initial infestation of a freshly stocked site; this EIP is dependent on the density of sea lice present at the neighbouring sites (Aldrin et al., 2013). The situation of a greater IIP compared to EIP was observed in Chile (Kristoffersen et al., 2013) and Grand Manan, NB (Elghafghuf et al., 2020). These authors proposed that the situation of the larger IIP vs. EIP was due to 1) asynchronous treatment applications, 2) treatments applied had better effectiveness against the adult life stages compared to the juvenile life stages, 3) there was a large component of flushing mobile life stages out of the sites prior to the infestation of that site, and 4) the improved management of sea lice infestation at their site compared to their neighbours' sites.

There are likely many factors involved in the difference in the magnitude of the effects of IIP to EIP. The modelling framework, underlying assumptions, location, empirical sea lice densities, management strategies employed, environmental variables and other factors likely play a role in the effects of the IIP and EIP.

1.7.4.4 Assumptions, limitations, and validation

The IIP and EIP are calculated using varying sea lice life stages and lags. There are multiple assumptions behind the method of calculating these infestation estimates. The decisions on life stages and lags utilized in the calculation depend specifically on biological information. The development rates of the sea lice life stages, natural mortality or secondary to treatments, and reproduction rate can each be modelled extensively (e.g., Aldrin et al., 2017, 2019) from the biological

information that drives these estimates. Brooker et al. (2018) reviewed the models for these rates of the biology of sea lice and concluded that there was no consensus on the parameters or equations. The basic science of development rates (e.g., Stien et al., 2005), and egg production (Heuch et al., 2000; Mustafa et al., 2000a), among many others, are crucial to the models that then inform mitigation strategies. There are also other variables related to the infestation of sea lice that are difficult to model. For example, the contact time necessary for successful attachment and the competition for space that is dependent on the density of attached and mobile sea lice currently on the host (Frenzl, 2014). Researchers select averages of the biological rates and processes for application in models of sea lice (e.g., Aldrin et al., 2017).

Another important assumption for infestation pressures is the additive relationship between IIP and EIP, which was first stated by Aldrin et al. (2013) and other models applied this assumption (e.g., Elghafghuf et al., 2020). The relationship between IIP and EIP may be non-additive; for example, the infestation pressures may be additive until a threshold is reached where the competition of sea lice for ideal attachment sites on hosts may render the relationship non-additive. Further analysis into the relationship between IIP and EIP should be carried out but may be difficult to accomplish due to the challenges of estimating dispersal. One method may be to estimate the EIP at the time of stocking; however, this may not reflect the EIP during the entire production cycle as the stocked smolt are younger, smaller, and may have been administered chemotherapeutants at the hatchery.

The issues with the use of a Gaussian kernel dispersal apply here, too, because the infestation pressure typically includes a kernel weighting in its calculation. Cantrell et al. (2018) commented that the infestation pressure may not decrease proportionally to an increase in distance from the source for sea lice as it would do for water-borne viruses (Murray and Gubbins, 2016). Infestation pressure is kernel smoothed in space especially compared to biophysical models (Kristoffersen et al., 2018).

The validation of infestation pressure measures was demonstrated in Kristoffersen et al. (2018). For the study area, there were sentinel cage sea lice monitoring results to compare to the calculated infestation pressures. The modelled infestation compared to the observed sea lice in sentinel cages explained 39% of the variation in the observed (Kristoffersen et al., 2018). Their model using infestation pressure outperformed an alternative model using temperature (Kristoffersen et al., 2018). Sandvik et al. (2016, 2021) and others (Salama et al., 2018) utilized observed counts of sea lice in sentinel cages to calculate the relative operating characteristic (ROC) to estimate an index of accuracy.

1.8 Sea lice models of dispersal and infestation

1.8.1 Statistical vs. simulation models

The previous discussion of dispersal distance and infestation pressure has introduced many of the key studies for sea lice models of dispersal and infestation. There are many types of models and studies because the life cycle of sea lice and its environment are complex, and it is a pressing issue to increase

our knowledge base of sea lice to mitigate them appropriately. Salama and Rabe (2013) reviewed the different models published on the dispersal and infestation of sea lice.

Statistical and numerical models of sea lice were among the first models developed to evaluate the dispersal and infestation of sea lice (e.g., delay-differential equations [Revie et al., 2005]). This is because they do not require the development of a hydrodynamic model for the study area and do not require the same computing processing power and time to run the hydrodynamic model. Examples of statistical models recently published are the deterministic model with empirical estimates of relative risk of sea lice transmission among farms (Kristoffersen et al., 2014), partly stage-structured model (Aldrin et al., 2019) and matrix population model (Toorians and Adams, 2020). The matrix population model by Toorians and Adams (2020) incorporated aspects of previous statistical models (e.g., delay-differential equations [Revie et al., 2005] and matrix models [Groner et al., 2014, 2016a]) to estimate a probability of successful dispersal.

Simulation models are numerical models that generate data to examine the effects under controlled circumstances (e.g., Stucchi et al. 2011; Adams et al. 2012; Asplin et al. 2014; Samsing et al. 2017; Salama et al. 2013, 2016, 2018) and they are a more recent addition to the tools available for sea lice modelling activities because they are computationally expensive (Cantrell et al., 2021). Within the simulation models are the agent-based model and within that category are the coupled biological-physical (i.e., biophysical) models that combine hydrodynamic and particle tracking models; multiple terms for these models may

lead to confusion (Cantrell et al., 2020). Cantrell et al. (2020) summarized biophysical models for modelling marine pathogen dispersal. Examples of agent-based models where the particles respond to environmental parameters include Stucchi et al. (2011), Adams et al. (2012) and Asplin et al. (2014), where for example the survivability of free-living stages of sea lice depend on the exposure to various salinity levels along its track. The particles may also have biological properties that improve the simulation of behaviours, such as vertical swimming capacity (e.g., Stucchi et al., 2011; Adams et al. 2015; Salama et al. 2016; Cantrell et al. 2018), in search of more favourable conditions to improve survival rates. An advantage of simulations is the ability to examine multiple scenarios (e.g., Cantrell et al., 2019), and even extend the parameters beyond their observed ranges to simulate hypothetical scenarios.

1.8.2 Empirical vs. theoretical models

Empirical datasets can be used during statistical and simulation modelling (e.g., Cantrell et al., 2021). Statistical models based on empirical datasets can be useful for the identification of risk factors and associations (e.g., Kristoffersen et al., 2018; Elghafghuf et al., 2018, 2020). Empirical abundances of sea lice can be used to validate theoretical models (e.g., Sandvik et al., 2016a, 2020, 2021), or identify emergent behaviour of networks (e.g., Cantrell et al., 2021). The major limitation of empirical models is the reliance on the primary-user, which is the farmer, manager, or other staff, that inputs data into the database.

Theoretical modelling includes the biophysical models of sea lice. Biophysical models include theoretical equations, assumptions, and averages of biological

rates (e.g., Revie et al., 2005). The biophysical model may be completely theoretical if the particles are released from a source site without prior definition of the abundance of sea lice (e.g., Sandvik et al., 2016a; Cantrell et al., 2018; Aleynik et al., 2022) or may be adjusted with empirical data to better represent a true scenario of egg production density (e.g., Sandvik et al., 2020, 2021). A disadvantage of biophysical models is the impossibility of applying the model to a different area as it is completely dependent on the hydrodynamic model for a specific study area. In comparison, empirical models may be more easily adjusted to a new study area with the use of a dataset of sea lice abundances for that location. To have empirical models that represent the area and produce robust estimates, ideally a dataset with a minimum of 3 to 5 years is needed.

1.8.3 Models for prediction vs. risk factors

Risk factor studies contribute to the understanding of the biological, ecological, and epidemiological processes underlying sea lice dispersal and infestation/disease. These studies contribute to the understanding of the causal relationships among risk factors and disease/infestation. These causal relationships are often complicated and multifactorial; therefore, large-scale cohort studies help to identify and disentangle these relationships. Repeated cohort studies provide more evidence to support or modify our hypotheses of certain relationships.

In contrast, models for prediction purposes rely on an appropriate set of predictors as developed through previous modelling endeavours examining risk factors. Both model types are useful for the translation of the knowledge of

dispersal and infestation of sea lice into actionable goals for the farmer, manager, veterinarian, and other stakeholder of the salmon aquaculture industry. Elghafghuf et al. (2021) predicted abundances of sea lice using a statistical model and empirical data. Simulation studies can produce predictions such that the Institute of Marine Research publishes regular reports for the aquaculture industry in the Norwegian coast (Myksvoll et al., 2018).

A disadvantage of models for the evaluation of risk factors is the possibility of inconsistent results, such as changes in the significance of predictors (e.g., wind [Revie et al., 2003; Cantrell et al., 2018]). This may render decisions for model-building for prediction purposes more difficult (Cantrell et al., 2019), but as additional risk factor studies are carried out, the weight of the evidence shifts toward supporting true causal relationships. A disadvantage of prediction models is the condition upon which the prediction was completed. For example, the predictions in Aldrin et al. (2017) were calculated knowing the application of treatments during the forecasting window. The application of such a prediction tool would have to be adjusted for use in the field to either produce results without any treatments or by selection of available treatments by the farmer or manager that is using the prediction tool. There are additional challenges for predictive models, which include overfitting with too many predictors, and the selection of an inappropriate window for prediction, among others. A challenge specific to biophysical models used in prediction is the current inability to translate the number or density of particles to an absolute risk value (Frenzl,

2014); however, some recent publications are working toward that end (e.g., Toorians and Adams, 2020).

1.8.4 Model predictors: highlight on temperature

1.8.4.1 Remote-sensed vs. *in-situ* measurement of temperature

Sea water temperature is a crucial variable in the biology of the sea lice as it determines the rate of development of the different life stages. Therefore, it was important to include it in our analyses. Data would optimally be collected at the cage during sea lice counts at recorded constant depths across sites for the study area. However, the quality of in-situ (farm site) temperature measurements was poor, and the data were sparse. We used United Kingdom Meteorological Office (UKMO) sea surface temperatures (UK Met Office, 2005) as these were demonstrated to have a high concordance correlation coefficient and index of agreement with in-situ measurements in the Broughton Archipelago (west coast of Canada [Thakur et al., 2018]). In our study area, Elghafghuf et al. (2020) used interpolation techniques to replace the in-situ missing values with UKMO sea surface temperatures.

A limitation to the UKMO sea surface temperature data was the measurement of the temperature at the surface only. Water temperature correlates with depth and may impact the activity and population dynamics of sea lice. The Fish-iTrends data lack consistent in-situ temperature measurements taken at consistent/standard depths. The UKMO temperature had a daily temporal resolution; the temperatures are measured with satellite data and under cloud

cover apply a Numerical Weather Prediction to input missing values (UK Met Office, 2005). Future models such as FVCOM (hydrodynamic model) may help account for depth and spatiotemporal variation of temperatures within those model time periods.

A small study was conducted to compare the UKMO sea surface temperatures and measured water temperatures to determine if the use of the UKMO sea surface temperatures were acceptable. The following is a small comparison of the UKMO temperature to a dataset of *in-situ* water temperatures in the Passamaquoddy Bay, NB.

1.8.4.2 Materials and methods

The dataset of water temperatures in the Passamaquoddy Bay were collected at Fisheries and Oceans Canada stations (Robinson, 2019). The sites from our study area were selected by identifying the closest DFO stations within 3 km of the approximate centre points (centroids) of sites using R. The methods for the selection of centroids of sites was described in the methods for Chapters 3 and 5.

The Fisheries and Oceans Canada (DFO) temperatures were compared to the UKMO sea surface temperatures for the selected sites/stations and at dates when the measurements were performed over the date range 01 July 2009 to 05 November 2018. A concordance correlation coefficient was calculated for all depths and stratified by depth (< 5 m, ≥ 5 to < 10 m, and ≥ 10 m) of the DFO

data. The depth for the UKMO temperatures was 0 m as it was from a remote-sensing product.

Analyses were conducted in Stata® v16.

1.8.4.3 Results

In Passamaquoddy Bay, 29 sites were matched to their nearest DFO station. A total of 1,817 temperature measurements were available for comparison. The DFO station temperatures were highly correlated (ρ_c = 0.832, SE 0.007) with our UKMO temperatures without accounting for depth (Figure 1.5). With stratification of the temperatures per depth, the correlation between DFO and UKMO temperatures remained high. At depths less than 10 m, where farmed salmon reside, the concordance correlation coefficient was 0.917 for the depth less than 5 m (n = 231, SE 0.010) and 0.909 for the depth 5-10 m (n = 253, SE 0.011).

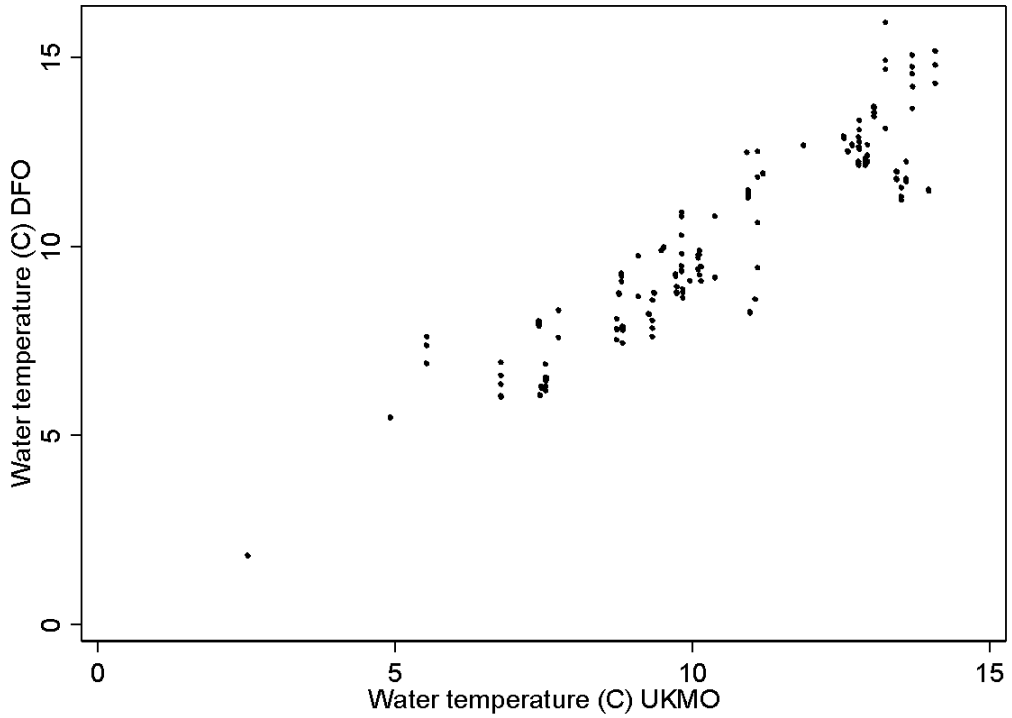

Figure 1.5. Direct comparison of Fisheries and Oceans Canada and UKMO remote-sensing sea surface temperatures for 29 matched sites (Atlantic salmon aquaculture site and DFO Station) in the Passamaquoddy Bay, New Brunswick, Canada for the period July 2009 to November 2018.

1.8.4.4 Discussion

The UKMO remote-sensing product was highly correlated with 'ground-truthed' DFO temperature measurements up to a depth of about 10 m. Therefore, it was deemed acceptable to use the UKMO remote-sensing product as a replacement for the *in-situ* temperatures for our study. The DFO temperatures measurements did not have the spatial and temporal resolution for use of these data in our models, and the UKMO temperatures had daily grid values for the study area and time period.

1.9 Thesis objectives

The sea louse *L. salmonis* infests the Atlantic salmon in the aquaculture industry in the Bay of Fundy, NB. This has been an issue for the industry since the parasite was observed at higher abundances in the mid-1990's (Hogans, 1995). At sufficient intensity, *L. salmonis* infestation causes wounds (Tully and Nolan 2002; Johnson et al., 2004), decrease the ability to osmoregulate (Grimnes and Jakobsen, 1996; Fjelldal et al., 2020) and immunocompromise its host (Fast, 2014). Severe sea lice infestation is responsible for health and welfare issues to the salmon, and economic losses to the producers (Mustafa et al., 2001). The mitigation practices for *L. salmonis* are multiple from zonal management to the application of chemotherapeutants. This dissertation's aims improve on the knowledge of the dispersal and connectivity of *L. salmonis* sea lice in the Bay of Fundy, NB, to arm the farmers, managers, veterinarians, and other stakeholders with information to improve on their mitigation practices. The models developed for this project made use of the monitoring counts from the Fish-iTrends© data management system for the Bay of Fundy, NB, for the period 2009 to 2021. This dissertation contains five additional chapters, and these are organized as follows:

The intention of Chapter 2 was to describe the trends of sea lice counts, treatments, and sea surface temperature between 2009 and 2018 specifically in the Bay of Fundy area, in southwest New Brunswick. Data were described to observe annual, seasonal, and spatial trends, and to identify possible confounding factors. These confounding factors may be used during the model building process during subsequent chapters.

The objective of Chapter 3 was to estimate the impact of the internal and external infestation pressures on the abundance of AF sea lice for the Bay of Fundy, NB, for the period 2009-2018. The model constructed was used for the empirical estimation of the average seaway dispersal distance of sea lice. The data were organized into segments as identified by counts immediately after a chemotherapeutic bath treatment, which allowed for a resetting of the count immediately after a treatment event. The results of this study had implications for the management of the NB aquaculture industry as it may suggest the need for a different BMA design for the synchronization of site production cycles (i.e., stocking and harvesting), as well as for synchronizing mitigation strategies among sites to maximize efficiency. This chapter was published in the *Journal of Fish Diseases* (https://doi.org/10.1111/jfd.13511).

The objectives of Chapter 4 were to compare different connectivity measures of dispersal distance and assess their impact when used in the calculation of external infestation pressure on the modelling of abundance of the AF sea lice in the Bay of Fundy, NB. The analysis employed the previously published model, same study area and dataset as published in Chapter 3. This chapter is under review in the *Journal of Experimental Marine Biology and Ecology*. The supplemental information for the chapter was submitted as a separate manuscript and is under review in the journal *MethodsX*.

The objectives of Chapter 5 were twofold. The first was the construction of a time series model for the abundance of AF sea lice in the Bay of Fundy, NB, for the period 2016-2021. The second objective was to illustrate the predictive

capabilities of the time series model for short-term predictions at the end of the summer in 2020. The results, and in particular the estimated infestation pressures, will be compared to the previously published estimates that were produced in Chapter 3 for the same study area. The manuscript derived from this chapter will be submitted for publication.

The last chapter of the thesis discussed the major results of the infestation pressures, temperature and treatment-temperature interactions, and dispersal distance from previous chapters (3-5). It also had the objective to compare the final models presented in Chapters 3 and 5 using an overlapping subset of the datasets that were used to produce the models (2016-2018).

1.10 References

á Norði, G., Simonsen, K., Danielsen, E., Eliasen, K., Mols-Mortensen, A., Christiansen, D. H., Steingrund, P., Galbraith, M., and Patursson. (2015). Abundance and distribution of planktonic *Lepeophtheirus salmonis* and *Caligus elongatus* in a fish farming region in the Faroe Islands. Aquaculture Environment Interactions, 7(1), 15-27. https://doi.org/10.3354/aei00134

á Norði, G., Simonsen, K., and Patursson, Ø. (2016). A method of estimating *in situ* salmon louse nauplii production at fish farms. Aquaculture Environment Interactions, 8, 397-405. https://doi.org/10.3354/aei00185

Aaen, S. M., Helgesen, K. O., Bakke, M. J., Kaur, K., and Horsberg, T. E. (2015). Drug resistance in sea lice: A threat to salmonid aquaculture. Trends in Parasitology, 31(2), 72–81. https://doi.org/10.1016/j.pt.2014.12.006

Abolofia, J., Asche, F., and Wilen, J. E. (2017). The Cost of lice: Quantifying the impacts of parasitic sea lice on farmed salmon. Marine Resource Economics, 32(3), 329-349. https://doi.org/10.1086/691981

Adams, T., Black, K., Macintyre, C., Macintyre, I., and Dean, R. (2012). Connectivity modelling and network analysis of sea lice infection in Loch Fyne, west coast of Scotland. Aquaculture Environment Interactions, 3, 51-63. https://doi.org/10.3354/aei00052

Adams, T. P., Proud, R., and Black, K. D. (2015). Connected networks of sea lice populations: Dynamics and implications for control. Aquaculture Environment Interactions, 6(3), 273-284. https://doi.org/10.3354/aei00133

Aldrin, M., Huseby, R. B., Stien, A., Grøntvedt, R. N., Viljugrein, H., and Jansen, P. A. (2017). A stage-structured Bayesian hierarchical model for salmon lice populations at individual salmon farms – Estimated from multiple farm data sets. Ecological Modelling, 359, 333-348.

Aldrin, M., Jansen, P. A., and Stryhn, H. (2019). A partly stage-structured model for the abundance of salmon lice in salmonid farms. Epidemics, 26(July 2018), 9-22. https://doi.org/10.1016/j.epidem.2018.08.001

Aldrin, M., Lyngstad, T., Kristoffersen, A., Storvik, B., Borgan, Ø., Jansen, P. A. (2011). Modelling the spread of infectious salmon anaemia among salmon farms based on seaway distances between farms and genetic relationships between infectious salmon anaemia virus isolates. Journal of the Royal Society Interface, 8, 1346–1356. http://dx.doi.org/10.1098/rsif.2010.0737

Aldrin, M., Storvik, B., Kristoffersen, A. B., and Jansen, P. A. (2013). Space-Time Modelling of the Spread of Salmon Lice between and within Norwegian Marine Salmon Farms. PLoS ONE, 8(5), 1-10. https://doi.org/10.1371/journal.pone.0064039

Aleynik, D., Adams, T., and Davidson, K. (2022). Optimizing the connectivity of salmon farms: Role of exposure to wind, tides, and isolation. In Md. N. Islam and S. M. Bartell (Eds.), Global Blue Economy: Analysis, Developments, and Challenges (1st ed., pp. 61-86). CRC Press. https://doi.org/10.1201/9781003184287-3

Amundrud, T. L., and Murray, A. G. (2009). Modelling sea lice dispersion under varying environmental forcing in a Scottish sea loch. Journal of Fish Diseases, 32(1), 27-44. https://doi.org/10.1111/j.1365-2761.2008.00980.x

Arriagada, G., Vanderstichel, R., Stryhn, H., Milligan, B., and Revie, C. W. (2016). Evaluation of water salinity effects on the sea lice *Lepeophtheirus salmonis* found on farmed Atlantic salmon in Muchalat Inlet, British Columbia, Canada. Aquaculture, 464, 554-563. https://doi.org/10.1016/j.aquaculture.2016.08.002

Asplin, L., Boxaspen, K., and Sandvik, A. D. (2004). Modelled distribution of salmon lice in a Norwegian fjord. ICES CM, 11-23. http://www.ices.dk/products/CMdocs/2004/P/P1104.pdf

Asplin, L., Boxaspen, K. K., and Sandvik, A. D. (2011). Modeling the Distribution and Abundance of Planktonic Larval Stages of *Lepeophtheirus salmonis* in Norway. In S. Jones and R. Beamish (Eds.), Salmon Lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). John Wiley and Sons, Inc.

Asplin, L., Johnsen, I. A., Sandvik, A. D., Albretsen, J., Sundfjord, V., Aure, J., and Boxaspen, K. K. (2014). Dispersion of salmon lice in the Hardangerfjord. Marine Biology Research, 10(3), 216-225. https://doi.org/10.1080/17451000.2013.810755

Astensrud, M. 1990. Molting and mating *Leopeophtheirus pectoralis* (Copepoda, Caligidae). Journal of the Marine Biology Association of the United Kingdom. 70: 269-281. https://doi.org/10.1017/S002531540003596

Atlas, W. I., Ban, N. C., Moore, J. W., Tuohy, A. M., Greening, S., Reid, A. J., Morven, N., White, E., Housty, W. G., Housty, J. A., Service, C. N., Greba, L., Harrison, S., Sharpe, C., Butts, K. I. R., Shepert, W. M., Sweeney-Bergen, E., Macintyre, D., Sloat, M. R., and Connors, K. (2021). Indigenous Systems of Management for Culturally and Ecologically Resilient Pacific Salmon (*Oncorhynchus* spp.) Fisheries. BioScience, 71(2), 186-204. https://doi.org/10.1093/biosci/biaa144

Bacher, C., Filgueira, R., and Guyondet, T. (2016). Probabilistic approach of water residence time and connectivity using Markov chains with application to tidal embayments. Journal of Marine Systems, 153, 25-41. https://doi.org/10.1016/j.jmarsys.2015.09.002

Bailey, J. K., Loudanslager, E. J., Glebe, B. D., Saunders, R. L., and Schom, C. B. (1983). Salmon Genetics Research Program, North American Salmon Research Center. http://www.dfo-mpo.gc.ca/Library/193404.pdf

Barisic, J., Cannon, S., and Quinn, B. (2019). Cumulative impact of anti-sea lice treatment (azamethiphos) on health status of Rainbow trout (Oncorhynchus mykiss, Walbaum 1792) in aquaculture. Scientific Reports, 9(1), 16217. https://doi.org/10.1038/s41598-019-52636-1

Barker, S. E., Bricknell, I. R., Covello, J., Purcell, S. L., Fast, M. D., Wolters, W., and Bouchard, D. A. (2019). Sea lice, *Lepeophtheirus salmonis* (Krøyer 1837), infected Atlantic salmon (*Salmo salar* L.) are more susceptible to infectious salmon anemia virus. PLoS ONE, 14(1), 1-28. https://doi.org/10.1371/journal.pone.0209178

Barrett, L. T., Oppedal, F., Robinson, N., and Dempster, T. (2020). Prevention not cure: A review of methods to avoid sea lice infestations in salmon aquaculture. Reviews in Aquaculture, 12(4), 2527-2543. https://doi.org/10.1111/raq.12456

Besnier, F., Kent, M., Skern-Mauritzen, R., Lien, S., Malde, K. et al. (2014). Human-induced evolution caught in action: SNP-array reveals rapid amphi-atlantic spread of pesticide resistance in the salmon ectoparasite *Lepeophtheirus salmonis*. BMC Genomics, 15, 937

Bista, D., Baxter, G. S., Hudson, N. J., Lama, S. T., Weerman, J., and Murray, P. J. (2021). Movement and dispersal of a habitat specialist in human-dominated landscapes: a case study of the red panda. Movement Ecology, 9(1), 1-15. https://doi.org/10.1186/s40462-021-00297-z

Bjørn, P. A., and Finstad, B. (1997). The physiological effects of salmon lice infection on sea trout post smolts. Nordic Journal of Freshwater Research, 73, 60-72.

Bjørn, P. A., Finstad, B., and Kristoffersen, R. (2001). Salmon lice infection of wild sea trout and Arctic char in marine and freshwaters: The effects of salmon farms. Aquaculture Research, 32(12), 947-962. https://doi.org/10.1046/j.1365-2109.2001.00627.x

Boxaspen, K. (2006). A review of the biology and genetics of sea lice. ICES Journal of Marine Science, 63(7), 1304-1316. https://doi.org/10.1016/j.icesjms.2006.04.017

Boxaspen, K., and Naess, T. (2000). Development of eggs and the planktonic stages of salmon lice (*Lepeophtheirus salmonis*) at low temperatures. Contributions to Zoology, 69(1), 51-55.

Braden, L. M., Koop, B. F., and Jones, S. R. M. (2015). Signatures of resistance to *Lepeophtheirus salmonis* include a TH_2-type response at the louse-salmon interface. Developmental and Comparative Immunology 48, 178-191.

Brandal P. O., Egidius, E., and Romslo, I. (1976). Host blood: A major food component for the parasitic copepod *Lepeophtheirus salmonis* Krøyer, 1838 (Crustacea: Caligidae). Norwegian Journal of Zoology, 24, 341-343.

Brauner, C. J., Sackville, M., Gallagher, Z., Tang, S., Nendick, L., and Farrell, A. P. (2012). Physiological consequences of the salmon louse (*Lepeophtheirus salmonis*) on juvenile pink salmon (*Oncorhynchus gorbuscha*): Implications for wild salmon ecology and management, and for salmon aquaculture. Philosophical Transactions of the Royal Society B, 367, 1770-1779. https://doi.org/10.1098/rstb.2011.0423

Bravo, S. (2003). Sea lice in Chilean salmon farms. Bulletin of the European Association of Fish Pathologists, 23(4), 197-200.

Bravo, F., Sidhu, J., Bernal, P., Bustamante, R., Condie, S., Gorton, B., Herzfeld, M., Jimenez, D., Mardones, F., Rizwi, F., and Steven, A. (2020). Hydrodynamic connectivity, water temperature, and salinity are major drivers of piscirickettsiosis prevalence and transmission among salmonid farms in Chile. Aquaculture Environment Interactions, 12, 263-279. https://doi.org/10.3354/aei00368

Breau, C. (2012). Knowledge of fish physiology used to set water temperature thresholds for in-season closures of Atlantic salmon (*Salmo salar*) recreational fisheries. DFO Canadian Science Advisory Secretariat Report 2012/163, 24p.

Bricknell, I. R., Dalesman, S. J., O'Shea, B., Pert, C. C., and Luntz, A. J. M. (2006). Effect of environmental salinity on sea lice *Lepeophtheirus salmonis* settlement success. Diseases of Aquatic Organisms, 71(3), 201-212. https://doi.org/10.3354/dao071201

Bron, J. E., Sommerville, C., Wootten, R., and Rae, G. H. (1993). Fallowing of marine Atlantic salmon, *Salmo salar* L., farms as a method for the control of sea lice, *Lepeophtheirus salmonis* (Krøyer, 1837). Journal of Fish Diseases, 16(5), 487-493. https://doi.org/10.1111/j.1365-2761.1993.tb00882.x

Brooker, A. J., Skern-Mauritzen, R., and Bron, J. E. (2018). Production, mortality, and infectivity of planktonic larval sea lice, *Lepeophtheirus salmonis* (Krøyer, 1837): Current knowledge and implications for epidemiological modelling. ICES Journal of Marine Science, 75(4), 1214-1234. https://doi.org/10.1093/icesjms/fsy015

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(3), 177-204. https://doi.org/10.1080/10641260500207109

Bruno, D. W., and Stone, J. (1990). The role of saithe, *Pollachius virens* L., as a host for the sea lice, *Leopeophtheirus salmonis* Krøyer and *Caligus elongatus* Nordmann. Aquaculture, 89(3-4), 201-207. https://doi.org/10.1016/0044-8486(90)90125-7

Burridge, L. E., and Van Geest, J. L. (2014). A review of potential environmental risks associated with the use of pesticides to treat Atlantic salmon against infestations of sea lice in Canada. DFO Canadian Science Advisory Secretariat Research Document 2014/002. 39 p.

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. https://doi.org/10.1016/j.aquaculture.2010.05.020

Byrne, A. A., Pearce, C. M., Cross, S. F., Jones, S. R. M., Robinson, S. M. C., Hutchinson, M. J., Miller, M. R., Haddad, C. A., and Johnson, D. L. (2018). Planktonic and parasitic stages of sea lice (*Lepeophtheirus salmonis* and *Caligus clemensi*) at a commercial Atlantic salmon (*Salmo salar*) farm in British Columbia, Canada. Aquaculture, 486, 130-138. https://doi.org/10.1016/j.aquaculture.2017.12.009

Campbell, P. J., Hammell, K. L., Dohoo, I. R., and Ritchie, G. (2006). Historical control clinical trial to assess the effectiveness of teflubenzuron for treating sea lice on Atlantic salmon. Diseases of Aquatic Organisms, 70, 109-114.

Cantrell, D., Filgueira, R., Revie, C. W., Rees, E., Vanderstichel, R., Foreman, M. G. G., Wan, D., and Grant, J. (2019). The relevance of larval biology on spatiotemporal patterns of pathogen connectivity among open-marine salmon farms. Canadian Journal of Fisheries and Aquatic Sciences, 77(3), 505-519. https://doi.org/10.1139/cjfas-2019-0040

Cantrell, D. L., Groner, M. L., Ben-Horin, T., Grant, J., and Revie, C. W. (2020). Modeling pathogen dispersal in marine fish and shellfish. Trends in Parasitology, 36(3), 239-249. https://doi.org/10.1016/j.pt.2019.12.013

Cantrell, D. L., Rees, E. E., Vanderstichel, R., Grant, J., Filgueira, R., and Revie, C. W. (2018). The use of kernel density estimation with a bio-physical model provides a method to quantify connectivity among salmon farms: Spatial planning and management with epidemiological relevance. Frontiers in Veterinary Science, 5, 1-14. https://doi.org/10.3389/fvets.2018.00269

Cantrell, D., Vanderstichel, R., Filgueira, R., Grant, J., and Revie, C. W. (2021). Validation of a sea lice dispersal model: Principles from ecological agent-based models applied to aquatic epidemiology. Aquaculture Environment Interactions, 13, 65-79. https://doi.org/10.3354/AEI00390

Casuso, A., Valenzuela-Muñoz, V., Benavente, B. P., Valenzuela-Miranda, D., and Gallardo-Escárate, C. (2022). Exploring sea lice vaccines against early

stages of infestation in Atlantic salmon (*Salmo salar*). Vaccines, 10(7), 1063-1085. https://doi.org/10.3390/vaccines10071063

Chang, B. D., and Coombs, K. A. (2016). A history of the Southwest New Brunswick Aquaculture Environmental Coordinating Committee: The first 25 years, 1989-2014. Canadian Technical Report of Fisheries and Aquatic Sciences 3152. Ottawa, ON: Government of Canada Fisheries and Oceans.

Chang, B. D., Coombs, K. A., and Page, F. H. (2014). The development of the salmon aquaculture industry in Southwestern New Brunswick, Bay of Fundy, including steps toward integrated coastal zone management. Aquaculture Economics and Management, 181(1), 1-27. https://doi.org/10.1080/13657305.2014.855952

Chang, B. D., and Page, F. H. (2014). Historical and spatial trends in selected aspects of the finfish aquaculture industry in southwestern New Brunswick, Bay of Fundy, 1978-2012. Canadian Technical Report of Fisheries and Aquatic Sciences 3112. Ottawa, ON: Government of Canada Fisheries and Oceans.

Chang, B. D., Page, F. H., Beattie, M. J., and Hill, B. W. H. (2011). Sea Louse Abundance on Farmed Salmon in the Southwestern New Brunswick Area of the Bay of Fundy. In S. Jones and R. Beamish (Eds.), Salmon Lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2005a). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry of Cobscook Bay, Maine and adjacent Southwestern New Brunswick. Canadian Technical Report of Fisheries and Aquatic Sciences 2598. St. Andrews, NB: Government of Canada Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2005b). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry of Letete Passage, Back Bay, Bliss Harbour, and Lime Kiln Bay in Southwestern New Brunswick. Canadian Technical Report of Fisheries and Aquatic Sciences 2599. St. Andrews, NB: Government of Canada Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2006a). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry of Eastern Grand Manan Island, Bay of Fundy. Canadian Technical Report of Fisheries and Aquatic Sciences 2621. St. Andrews, NB: Government of Canada Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2006b). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry in Passamaquoddy Bay, Southwestern New Brunswick. Canadian Technical Report of Fisheries and

Aquatic Sciences 2622. St. Andrews, NB: Government of Canada Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Lawton, P., Singh, R., and Greenberg, D. A. (2007). Evaluation of Bay Management Area Scenarios for the Southwestern New Brunswick Salmon Aquaculture Industry: Aquaculture Collaborative Research and Development Program Final Project Report. Canadian Technical Report of Fisheries and Aquatic Sciences 2722. St. Andrews, NB: Government of Canada Department of Fisheries and Oceans. https://doi.org/10.1017/CBO9781107415324.004

Chiswell, S. M. (2012). Non-Gaussian larval dispersal kernels in Gaussian ocean flows. Aquatic Biology, 16, 203-208. https://doi.org/10.3354/ab00466

Connors, B. M., Krkošek, M., and Dill, L. M. (2008). Sea lice escape predation on their host. Biology Letters, 4, 455-457. https://doi.org/10.1098/rsbl.2008.0276

Contreras, M., Karlsen, M., Villar, M., Olsen, R. H., Leknes, L. M., Furevik, A., Yttredal, K. L., Tartor, H., Grove, S., Alberdi, P., Brudeseth, B., and de la Fuente, J. (2020). Vaccination with ectoparasite proteins involved in midgut function and blood digestion reduces salmon louse infestations. Vaccines, 8(1), 32-49. https://doi.org/10.3390/vaccines8010032

Cardia, F., and Lovatelli, A. (2016). Aquaculture operations in floating HDPE cages: A field handbook. FAO Fisheries and Aquaculture Technical Paper 593. Rome: Food and Agriculture Organization of the United Nations and Ministry of Agriculture of the Kingdom of Saudi Arabia. http://www.fao.org/3/a-i4508e.pdf

Coletto, D., Di Francesco, L., and Morrison, J. (2011). Seafood survey: Public opinion on aquaculture and a National Aquaculture act: A survey of 1,200 Canadians. Ottawa, ON: Abacus Data.

Cook, P. F., McBeath, S. J., Bricknell, I. R., and Bron, J. E. (2010). Determining the age of individual *Lepeophtheirus salmonis* (Krøyer, 1837) copepodids by measuring stored lipid volume; Proof of principle. Journal of Microscopy, 240, 83-86. https://doi.org/10.1111/j.1365-2818.2010.03417.x

Correa, K., Lhorente, J. P., Bassini, L., López, M.E., Di Genova, A., Maass, A., Davidson, W. S., Yáñez, J. M. (2017). Genome wide association study for resistance to *Caligus rogercresseyi* in Atlantic salmon (*Salmo salar* L.) using a 50K SNP genotyping array. Aquaculture, 472, 61-65. https://doi.org/10.1016/j.aquaculture.2016.04.008

Costello, M.J. (1993). Review of methods to control sea lice (Caligidae: Crustacea) infestations on salmon (*Salmo salar*) farms. In G. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 219-252). West Sussex, UK: Ellis Horwood Ltd.

Costello, M. J. (2009a). 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 B: Biological Sciences, 276 (1672), 3385–3394. https://doi.org/10.1098/rspb.2009.0771

Costello, M. J. (2009b). The global economic cost of sea lice to the salmonid farming industry. Journal of Fish Diseases, 32, 115-118. https://doi.org/10.1111/j.1365-2761.2008.01011.x

Costelloe, M., Costelloe, J., and Roche, N. (1996). Planktonic dispersion of larval salmon-lice, *Lepeophtheirus salmonis*, associated with cultured salmon, *Salmo salar*, in Western Ireland. Journal of the Marine Biological Society of the United Kingdom, 76, 141-149.

Cowen, R. K., Paris, C. B., and Srinivasan, A. (2006). Scaling of connectivity in marine populations. Science, 311(5760), 522-527. https://doi.org/10.1126/science.1122039

Cowen, R. K., and Sponaugle, S. (2009). Larval dispersal and marine population connectivity. Annual Review of Marine Science, 1, 443-466. https://doi.org/10.1146/annurev.marine.010908.163757

D'Aloia, C. C., Bogdanowicz, S. M., Francis, R. K., Majoris, J. E., Harrison, R. G., and Buston, P. M. (2015). Patterns, causes, and consequences of marine larval dispersal. PNAS, 112(45), 13940-13945. https://doi.org/10.1073/pnas.1513754112

Davidson, J., May, T., Good, C., Waldrop, T., Kenney, B., Terjesen, B. F., and Summerfelt, S. (2016). Production of market-size North American strain Atlantic salmon *Salmo salar* in a land-based recirculation aquaculture system using freshwater. Aquacultural Engineering, 74, 1-16. https://doi.org/10.1016/j.aquaeng.2016.04.007

Davies, I. M., and Rodger, G. K. (2000). A review of the use of ivermectin as a treatment for sea lice [*Lepeophtheirus salmonis* (Krøyer) and *Caligus elongatus* Nordmann] infestation in farmed Atlantic salmon (*Salmo salar* L.). Aquaculture Research, 31(11), 869-883. https://doi.org/10.1046/j.1365-2109.2000.00510.x

DecaNet eds. (2023). DecaNet. *Alpheus immaculatus* Knowlton and Keller, 1983. Accessed through: World Register of Marine Species at: https://www.marinespecies.org/aphia.php?p=taxdetails&id=514897 on 2023-05-12

DecaNet eds. (2023). DecaNet. *Homarus americanus* H. Milne Edwards, 1837. Accessed through: World Register of Marine Species at: https://www.marinespecies.org/aphia.php?p=taxdetails&id=156134 on 2023-04-17

Dempster, T., Overton, K., Bui, S., Stien, L. H., Oppedal, F., Karlsen, Coates, A., Phillips, B. L., & Barrett, L. T. (2021). Farmed salmonids drive the abundance, ecology and evolution of parasitic salmon lice in Norway. Aquaculture Environment Interactions, 13, 237–248. https://doi.org/10.3354/AEI00402

Department of Fisheries and Oceans Canada. (2014). Archived – The Scotian Shelf: An Atlas of Human Activities – Atlas. Accessed at: http://www.inter.dfo-mpo.gc.ca/Maritimes/Oceans/OCMD/Atlas/Marine-Traffic on 2018-01-08

[map] DFO Aquaculture Management Division. (2021). 2020 Marine Finfish Aquaculture in British Columbia. Government of Canada Department of Fisheries and Oceans.

Douglas-Helders, G. M., Weir, I. J., O'Brien, D. P., Carson, J., and Nowak, B. F. (2004). Effects of husbandry on prevalence of amoebic gill disease and performance of reared Atlantic salmon (*Salmo salar* L.). Aquaculture, 241(1-4), 21-30. https://doi.org/10.1016/j.aquaculture.2004.07.026

Drinkwater, K. F. (1987). Mean Temperature and Salinity Conditions Near the Entrance to the Bay of Fundy, 1951-80. NAFO Science Council Studies, 71-73.

Eisenhauer, L., Solvang, T., Alver, M., Krause, D. F., and Hagemann, A. (2020). Dispersal of salmon lice (*Lepeophtheirus salmonis* Krøyer, 1837) egg strings from open-cage salmon farming: a neglected source for infestation dynamics. Aquaculture Research, 51, 4595-4601. https://doi.org/10.1111/are.14805

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2021). State-space modeling for inter-site spread of sea lice with short-term population predictions. Ecological Modelling, 452. https://doi.org/10.1016/j.ecolmodel.2021.109602

Elghafghuf, A., Vanderstichel, R., St-Hilaire, S., and Stryhn, H. (2018). Using state-space models to predict the abundance of juvenile and adult sea lice on Atlantic salmon. Epidemics, 24, 76-87. https://doi.org/10.1016/j.epidem.2018.04.002

Elmoslemany, A., Revie, C. W., Milligan, B., Stewardson, L., and Vanderstichel, R. (2015). Wild juvenile salmonids in Muchalat Inlet, British Columbia, Canada: Factors associated with sea lice prevalence. Diseases of Aquatic Organisms, 117(2), 107–120. https://doi.org/10.3354/dao02939

Elmoslemany, A., Whyte, S. K., Revie, C. W., and Hammell, K. L. (2013). Sea lice monitoring on Atlantic salmon farms in New Brunswick, Canada: Comparing

audit and farm staff counts. Journal of Fish Diseases, 36(3), 241-247. https://doi.org/10.1111/jfd.12051

Erikson, U., Rosten, C., Klebert, P., Aspaas, S., and Rosten, T. (2022). Live transport of Atlantic salmon in open and closed systems: Water quality, stress and recovery. Aquaculture Research, 53(11), 3913–3926. https://doi.org/10.1111/are.15895

FAO. (2020). The State of World Fisheries and Aquaculture: Sustainability in Action. Rome: FAO's Office for Corporate Communication Publishing group. https://doi.org/10.4060/ca9229en

FAO. (2022). GLOBEFISH Highlights – International markets for fisheries and aquaculture products, 2nd issue 2022, January-December 2021 Statistics. Globefish Highlights No. 2-2022. Rome: Food and Agriculture Organization of the United Nations. https://doi.org/10.4060/cc1350en

Fast, M. D. (2014). Fish immune responses to parasitic copepod (namely sea lice) infection. Developmental and Comparative Immunology, 43(2), 300-312. https://doi.org/10.1016/j.dci.2013.08.019

Fast, M. D., Burka, J. F., Johnson, S. C., and Ross, N. W. (2003). Enzymes released from *Leopeophtheirus salmonis* in response to mucus from different salmonids. Journal of Parasitology, 89, 7-13.

Fast, M. D., and Dalvin, S. (2020). Lepeophtheirosis (*Lepeophtheirus salmonis*). In P. T. K. Woo, J.-A. Leong, and K. Buchmann (Eds.), Climate Change and Infectious Fish Diseases (p. 529). Wallingford, UK; Boston, USA: CAB International.

Fast, M. D., Ross, N. W., Mustafa, A., Sims, D. E., Johnson, S. C., Conboy, G. A., Speare, D. J., Johnson, G., and Burka, J. F. (2002). Susceptibility of Rainbow trout *Oncorhynchus mykiss*, Atlantic salmon *Salmo salar* and Coho salmon *Oncorhynchus kisutch* to experimental infection with sea lice *Lepeophtheirus salmonis*. Diseases of Aquatic Organisms, 52, 57-68.

Fernandez-Gonzalez, V., Ulvan, E. M., Sanchez-Jerez, P., Diserud, O. H., Toledo-Guedes, K., Casado-Coy, N., Klebert, P., & Uglem, I. (2022). Abundance of sea lice larvae in plankton samples: Determination of optimal sample sizes. Aquaculture, 551. https://doi.org/10.1016/j.aquaculture.2022.737919

Fife, F. J., Goreham, R. L. M., and Page, F. H. (2015). A century of monitoring station Prince 6 in the St. Croix River estuary of Passamaquoddy Bay. Department of Fisheries and Oceans Canada; Ecosystems and Oceans Science; Environment Canada.

Fjelldal, P. G., Hansen, T. J., and Karlsen, Ø. (2020). Effects of laboratory salmon louse infection on osmoregulation, growth and survival in Atlantic salmon. Conservation Physiology, 8(1), 1-10. https://doi.org/10.1093/conphys/coaa023

Fjørtoft, H. B., Nilsen, F., Besnier, F., Stene, A., Tveten, A. K., Bjørn, P. A., Aspehaug, V. T., and Glover, K. A. (2021). Losing the “arms race”: Multiresistant salmon lice are dispersed throughout the North Atlantic Ocean. Royal Society Open Science, 8(5), 210265-210275. https://doi.org/10.1098/rsos.210265

Flaherty, M., Reid, G., Chopin, T., and Latham, E. (2019). Public attitudes towards marine aquaculture in Canada: Insights from the Pacific and Atlantic coasts. Aquaculture International, 27(1), 9-32. https://doi.org/10.1007/s10499-018-0312-9

Frenzl, B. (2014). Understanding key factors associated with the infection of farmed Atlantic salmon by the salmon louse *Lepeophtheirus salmonis*. Doctoral dissertation, University of Stirling. http://hdl.handle.net/1893/22320

Froese, R., and D. Pauly. (Eds.). (2023). FishBase. Salmonidae Jarocki or Schinz, 1822. Accessed through: World Register of Marine Species at: https://www.marinespecies.org/aphia.php?p=taxdetails&id=125587 on 2023-01-11

Gautam, R., Boerlage, A. S., Vanderstichel, R., Revie, C. W., and Hammell, K. L. (2016). Variation in pre-treatment count lead time and its effect on baseline estimates of cage level sea lice abundance. Journal of Fish Diseases, 39, 1297-1303. https://doi.org/10.1111/jfd.12460

Gautam, R., Price, D., Revie, C. W., Gardner, I. A., Vanderstichel, R., Gustafson, L., Klotins, K., and Beattie, M. (2018). Connectivity-based risk ranking of infectious salmon anaemia virus (ISAv) outbreaks for targeted surveillance planning in Canada and the USA. Preventive Veterinary Medicine, 159, 92-98. https://doi.org/10.1016/j.prevetmed.2018.09.001

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017). Effect of timing of count events on estimates of sea lice abundance and interpretation of effectiveness following bath treatments. Journal of Fish Diseases, 40(3), 367-375. https://doi.org/10.1111/jfd.12519

Gharbi, K., Matthews, L., Bron, J., Roberts, R., Tinch, A., and Stear, M. (2015). The control of sea lice in Atlantic salmon by selective breeding. Journal of the Royal Society Interface, 12, 20150574-20150582. doi: 10.1098/rsif.2015.0574

Gillibrand, P. A., and Willis, K. J. (2007). Dispersal of sea louse larvae from salmon farms: Modelling the influence of environmental conditions and larval behaviour. Aquatic Biology, 1(1), 63-75. https://doi.org/10.3354/ab00006

Girvetz, E. H., and Greco, S. E. (2007). How to define a patch: A spatial model for hierarchically delineating organism-specific habitat patches. Landscape Ecology, 22(8), 1131-1142. https://doi.org/10.1007/s10980-007-9104-8

Gislason, H. (2017). Statistical modelling of sea lice count data from salmon farms in the Faroe Islands. Journal of Fish Diseases, 41(6), 973-993. https://doi.org/10.1111/jfd.12742

Glover, K. A., Stølen, Å.B., Messmer, A., Koop, B. F., Torrissen, O. and Nilsen, F. (2011). Population genetic structure of the parasitic copepod *Lepeophtheirus salmonis* throughout the Atlantic. Marine Ecology Progress Series, 427, 161-172. https://doi.org/10.3354/meps09045

González, M. P., Muñoz, J. L. P., Valerio, V., and Vargas-Chacoff, L. (2016). Effects of the ectoparasite *Caligus rogercresseyi* on *Salmo salar* blood parameters under farm conditions. Aquaculture, 457, 29-34. https://doi.org/10.1016/j.aquaculture.2016.01.027

Government of Canada. (2019). Atlantic Salmon inner Bay of Fundy population: Action Plan, 2019. Accessed at: https://www.canada.ca/en/environment-climate-change/services/species-risk-public-registry/action-plans/Atlantic-salmon-inner-bay-fundy-2019.html on 2020-04-11.

Government of New Brunswick. (2018). General Regulation, NB Reg 91-158, 1-14. http://canlii.ca/t/544vm

Government of New Brunswick. (2023). Fish: Regulations Summary: Regular Season 2023-Winter 2024. Accessed at: https://www2.gnb.ca/content/dam/gnb/Departments/nr-rn/pdf/en/Fish/Fish.pdf on 2023-04-18.

Gravil, H. R. (1996). Studies on the biology and ecology of the free swimming larval stages of *Lepeophtheirus salmonis* (Krøyer, 1838) and *Caligus elongatus* Nordmann, 1832 (Copepoda: Caligidae). Doctoral dissertation, University of Stirling. http://hdl.handle.net/1893/2380

Green, D. M. (2010). A strategic model for epidemic control in aquaculture. Preventive Veterinary Medicine, 94(1-2), 119-127. https://doi.org/10.1016/j.prevetmed.2009.12.004

Greenberg, D. A. (1984). A Review of the Physical Oceanography of the Bay of Fundy. In D. C. Gordon, Jr., and M.J. Dadswell (Eds.), Update of the Marine Environmental Consequences of Tidal Power Development in the Upper Reaches of the Bay of Fundy. Canadian Technical Report Fisheries and Aquatic Sciences 1256. Dartmouth, Canada: Government of Canada Department of Fisheries and Oceans. publications.gc.ca/pub?id=9.577998&sl=0

Grimnes, A., and Jakobsen, P. J. (1996). The physiological effects of salmon lice infection on post-smolt of Atlantic salmon. Journal of Fish Biology, 48(6), 1179-1194. https://doi.org/10.1111/j.1095-8649.1996.tb01813.x

Groner, M.L., Gettinby, G., Stormoen, M., Revie, C.W., Cox, R. (2014). Modelling the impact of temperature-induced life history plasticity and mate limitation on the

epidemic potential of a marine ectoparasite. PLoS ONE 9 (2), e88465-e88476. https://doi. org/10.1371/journal.pone.0088465.

Groner, M. L., McEwan, G. F., Rees, E. E., Gettinby, G., and Revie, C. W. (2016a). Quantifying the influence of salinity and temperature on the population dynamics of a marine ectoparasite. Canadian Journal of Fisheries and Aquatic Sciences, 73, 1281-1291. https://doi.org/10.1139/cjfas-2015-0444

Groner, M. L., Rogers, L. A., Bateman, A. W., Connors, B. M., Frazer, L. N., Godwin, S. C., Krkošek, M., Lewis, M. A., Peacock, S. J., Rees, E. E., Revie, C. W., and Schlägel, U. E. (2016b). Lessons from sea louse and salmon epidemiology. Philosophical Transactions of the Royal Society B: Biological Sciences, 371(1689), 20150203-20150213. https://doi.org/10.1098/rstb.2015.0203

Guragain, P., Tkachov, M., Båtnes, A. S., Olsen, Y., Winge, P., and Bones, A. M. (2021). Principles and methods of counteracting harmful salmon-arthropod interactions in salmon farming: Addressing possibilities, limitations, and future options. Frontiers in Marine Science, 8, 1-19. https://doi.org/10.3389/fmars.2021.701793

Gustafson, L. L., Ellis, S. K., Beattie, M. J., Chang, B. D., Dickey, D. A., Robinson, T. L., Marenghi, F. P., Moffett, P. J., and Page, F. H. (2007). Hydrographics and the timing of infectious salmon anemia outbreaks among Atlantic salmon (*Salmo salar* L.) farms in the Quoddy region of Maine, USA and New Brunswick, Canada. Preventative Veterinary Medicine, 78, 35-56. https://doi.org/10.1016/j.prevetmed.2006.09.006

Hamilton-West, C., Arriagada, G., Yatabe, T., Valdés, P., Hervé-Claude, L. P., and Urcelay, S. (2012). Epidemiological description of the sea lice (*Caligus rogercresseyi*) situation in Southern Chile in August 2007. Preventive Veterinary Medicine, 104, 341-345. https://doi.org/10.1016/j.prevetmed.2011.12.002

Hammell, K. L., and Dohoo, I. R. (2005). Risk factors associated with mortalities attributed to infectious salmon anaemia virus in New Brunswick, Canada. Journal of Fish Diseases, 28(11), 651-661. https://doi.org/10.1111/j.1365-2761.2005.00668.x

Hamre, L. A., Eichner, C., Caipang, C. M. A., Dalvin, S. T., Bron, J. E., Nilsen, F., Boxshall, G., and Skern-Mauritzen, R. (2013). The salmon louse *Lepeophtheirus salmonis* (Copepoda: Caligidae) life cycle has only two chalimus stages. PLoS ONE, 8(9), 1-9. https://doi.org/10.1371/journal.pone.0073539

Hamre, L., Bui, S., Oppedal, F., Skern-Mauritzen, R., and Dalvin, S. (2019). Development of the salmon louse *Lepeophtheirus salmonis* parasitic stages in temperatures ranging from 3 to 24°C. Aquaculture Environment Interactions, 11, 429-443. https://doi.org/10.3354/aei00320

Harrington, P. D., Cantrell, D. L., Foreman, M. G. G., Guo, M., and Lewis, M. A. (2023). Timing and probability of arrival for sea lice dispersing between salmon farms. Royal Society Open Science, 10(2), 220853-220873. https://doi.org/10.1098/rsos.220853

Heuch, P. A., Bjørn, P. A., Finstad, B., Holst, J. C., Asplin, L., and Nilsen, F. (2005). A review of the Norwegian "National Action Plan Against Salmon Lice on Salmonids": The effect on wild salmonids. Aquaculture, 246(1-4), 79-92. https://doi.org/10.1016/j.aquaculture.2004.12.027

Heuch, P. A., Nordhagen, J. R., and Schram, T. A. (2000). Egg production in the salmon louse [*Lepeophtheirus salmonis* (Krøyer)] in relation to origin and water temperature. Aquaculture Research, 31(11), 805-814. https://doi.org/10.1046/j.1365-2109.2000.00512.x

Heuch, P. A., Parsons, A., and Boxaspen, K. (1995). Diel vertical migration – A possible host-finding mechanism in salmon louse (*Lepeophtheirus salmonis*) copepodids. Canadian Journal of Fisheries and Aquatic Sciences, 52(4), 681-689. https://doi.org/10.1139/f95-069

Houston, R. D. (2017). Future directions in breeding for disease resistance in aquaculture species. Revista Brasileira de Zootecnia, 46(6), 545-551. https://doi.org/10.1590/S1806-92902017000600010

Huret, M., Petitgas, P., and Woillez, M. (2010). Dispersal kernels and their drivers captured with a hydrodynamic model and spatial indices: A case study on anchovy (*Engraulis encrasicolus*) early life stages in the Bay of Biscay. Progress in Oceanography, 87(1-4), 6-17. https://doi.org/10.1016/j.pocean.2010.09.023

Hutchings, J. A., Ardren, W. R., Barlaup, B. T., Bergman, E., Clarke, K. D., Greenberg, L. A., Lake, C., Piironen, J., Sirois, P., Sundt-Hansen, L. E., and Fraser, D. J. (2019). Life-history variability and conservation status of landlocked Atlantic salmon: An overview. Canadian Journal of Fisheries and Aquatic Sciences, 76(10), 1697-1708. https://doi.org/10.1139/cjfas-2018-0413

Hvas, M., and Bui, S. (2022). Energetic costs of ectoparasite infection in Atlantic salmon. Journal of Experimental Biology, 255(1). https://doi.org/10.1242/JEB.243300

Hyatt, K. D., and Godbout, L. (2000). A review of salmon as keystone species and their utility as critical indicators of regional biodiversity and ecosystem integrity. In L. M. Darling (Ed.), Proceedings of a Conference on the Biology and Management of Species and Habitats at Risk (p. 520). BC Ministry of Environment, Lands and Parks and University College of the Cariboo.

Iizuka, M., and Zanlungo, J. P. (2016). Environmental collapse and institutional restructuring: The sanitary crisis in the Chilean salmon industry. In Chile's Salmon Industry: Policy Challenges in Managing Public Goods (pp. 109–135). Springer Japan. https://doi.org/10.1007/978-4-431-55766-1_4

Imsland, A. K. D., and Reynolds, P. (2022). In lumpfish we trust? The efficacy of lumpfish *Cyclopterus lumpus* to control *Lepeophtheirus salmonis* infestations on farmed Atlantic salmon: A Review. Fishes, 7(5), 220-240. https://doi.org/10.3390/fishes7050220

Jackson, D., Deady, S., Leahy, Y., and Hassett, D. (1997). Variations in parasitic caligid infestations on farmed salmonids and implications for their management. ICES Journal of Marine Science, 54, 1104-1112. https://academic.oup.com/icesjms/article/54/6/1104/673951

Jansen, P. A., Kristoffersen, A. B., Viljugrein, H., Jimenez, D., Aldrin, M., Stien, A. (2012). Sea lice as a density-dependent constraint to salmonid farming. Proceedings of the Royal Society B, 279 (1737), 2330–2338. https://doi.org/10.1098/rspb.2012.0084

Jaworski, A., and Holm, J. CHR. (1992). Distribution and structure of the population of sea lice, *Lepeophtheirus salmonis* Krøyer, on Atlantic salmon, *Salmo salar* L., under typical rearing conditions. Aquaculture Fisheries Management, 23(5), 577-589. https://doi.org/10.1111/j.1365-2109.1992.tb00802.x

Jeong, J., Arriagada, G., and Revie, C. W. (2023). Targets and measures: Challenges associated with reporting low sea lice levels on Atlantic salmon farms. Aquaculture, 563, 1-7. https://doi.org/10.1016/j.aquaculture.2022.738865

Jimenez, D. F., Revie, C. W., Hardy, S. P., Jansen, P. A., and Gettinby, G. (2013). Multivariate evaluation of the effectiveness of treatment efficacy of cypermethrin against sea lice (*Lepeophtheirus salmonis*) in Atlantic salmon (*Salmo salar*). BMC Veterinary Research, 9(1), 258-267. https://doi.org/10.1186/1746-6148-9-258

Johansen, L. H., Jensen, I., Mikkelsen, H., Bjørn, P. A., Jansen, P. A., and Bergh, O. (2011). Disease interaction and pathogens exchange between wild and farmed fish populations with special reference to Norway. Aquaculture, 315(3-4), 167-186. https://doi.org/10.1016/j.aquaculture.2011.02.014

Johnsen, I. A., Asplin, L. C., Sandvik, A. D., and Llinares, R. M. S. (2016). Salmon lice dispersion in a Northern Norwegian fjord system and the impact of vertical movements. Aquaculture Environment Interactions, 8, 99-116. https://doi.org/10.3354/aei00162

Johnsen, I., Fiksen, Ø., Sandvik, A., and Asplin, L. (2014). Vertical salmon lice behaviour as a response to environmental conditions and its influence on regional dispersion in a fjord system. Aquaculture Environment Interactions, 5(2), 127-141. https://doi.org/10.3354/aei00098

Johnson, S. C. (1993). A comparison of development and growth rates of *Lepeophtheirus salmonis* (Copepoda: Caligidae) on naive Atlantic (*Salmo salar*) and Chinook (*Oncorhynchus tshawytscha*) salmon. In G. Boxshall and D. Defaye

(Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 68-80). West Sussex, UK: Ellis Horwood Ltd.

Johnson, S. C. and Albright, L. J. (1991a). Development, growth, and survival of *Lepeophtheirus salmonis* (Copepoda: Caligidae) under laboratory conditions. Journal of the Marine Biological Association, 71(2): 425-436. https://doi.org/10.1017/S0025315400051687

Johnson, S.C. and Albright, L J. (1991b). The developmental stages of *Lepeophtheirus salmonis* (Krøyer, 1837) (Copepoda: Caligidae). Canadian Journal of Zoology, 69, 929-950. https://doi.org/10.1139/z91-138

Johnson, S. C., and Albright, L. J. (1992). Comparative susceptibility and histopathology of the response of naive Atlantic, chinook and Coho salmon to experimental infection with *Lepeophtheirus salmonis* (Copepoda: Caligidae). Diseases of Aquatic Organisms, 14, 179-193. https://doi.org/10.3354/dao014179

Johnson, S. C., and Margolis, L. (1993). Efficacy of ivermectin for control of the salmon louse *Lepeophtheirus salmonis* on Atlantic salmon. Diseases of Aquatic Organisms, 17, 101-105.

Jones, M. 2004. Salmonidae: Cultured Aquatic Species Information Programme. Fisheries and Aquaculture Division. Rome: Food and Agriculture Organization of the United Nations. Accessed at: https://www.fao.org/fishery/en/culturedspecies/salmo_salar/en on 2023-07-23.

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(2), 131-137. https://doi.org/10.3354/dao02043

Jones, P. G., Hammell, K. L., Dohoo, I. R., and Revie, C. W. (2012). Effectiveness of emamectin benzoate for treatment of *Lepeophtheirus salmonis* on farmed Atlantic salmon *Salmo salar* in the Bay of Fundy, Canada. Diseases of Aquatic Organisms, 102(1), 53-64. https://doi.org/10.3354/dao02517

Junquera, P., Hosking, B., Gameiro, M., and MacDonald, A. (2019). Benzoylphenyl ureas as veterinary antiparasitics. An overview and outlook with emphasis on efficacy, usage and resistance. Parasite, 26, 26-59. https://doi.org/10.1051/parasite/2019026

Kabata, Z. (1974). Mouth and mode of feeding of Caligidae (Copepoda), parasites of fishes, as determined by light and scanning electron microscopy. Journal of the Fisheries Research Board of Canada, 31, 1583-1588.

Kim, I.-H. (1993). Developmental stages of *Caligus punctatus* Shiino, 1955 (Copepoda: Caligidae). In G. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 16-29). West Sussex, UK: Ellis Horwood Ltd.

Kindlmann, P., and Burel, Æ. F. (2008). Connectivity measures: A review. Landscape Ecology, 23, 879-890. https://doi.org/10.1007/s10980-008-9245-4

Kragesteen, T., Simonsen, K., Visser, A., and Andersen, K. (2018). Identifying salmon lice transmission characteristics between Faroese salmon farms. Aquaculture Environment Interactions, 10, 49-60. https://doi.org/10.3354/aei00252

Kragesteen, T. J., Simonsen, K., Visser, A. W., and Andersen, K. H. (2021). Estimation of external infection pressure and salmon-louse population growth rate in Faroese salmon farms. Aquaculture Environment Interactions, 13, 21–32. https://doi.org/10.3354/aei00386

Kraly, P., Weitzman, J., and Filgueira, R. (2022). Understanding factors influencing social acceptability: Insights from the media portrayal of salmon aquaculture in Atlantic Canada. Aquaculture, 547, 737497-737506. https://doi.org/10.1016/j.aquaculture.2021.737497

Kristoffersen, A. B., Rees, E. E., Stryhn, H., Ibarra, R., Campisto, J. L., Revie, C. W., and St-Hilaire, S. (2013). Understanding sources of sea lice for salmon farms in Chile. Preventive Veterinary Medicine, 111(1-2), 165-175. https://doi.org/10.1016/j.prevetmed.2013.03.015

Kristoffersen, A. B., Jimenez, D., Viljugrein, H., Grøntvedt, R., Stien, A., and Jansen, P. A. (2014). Large scale modelling of salmon lice (*Lepeophtheirus salmonis*) infection pressure based on lice monitoring data from Norwegian salmonid farms. Epidemics, 9, 31-39. https://doi.org/10.1016/j.epidem.2014.09.007

Kristoffersen, A. B., Qviller, L., Helgesen, K. O., Vollset, K. W., Viljugrein, H., and Jansen, P. A. (2018). Quantitative risk assessment of salmon louse-induced mortality of seaward-migrating post-smolt Atlantic salmon. Epidemics, 23, 19-33. https://doi.org/10.1016/j.epidem.2017.11.001

Krkošek, M., Connors, B. M., Morton, A., Lewis, M. A., Dill, L. M., and Hilborn, R. (2011). Effects of parasites from salmon farms on productivity of wild salmon. PNAS, 108(35), 14700-14704. https://doi.org/10.1073/pnas.1101845108

Krkošek, M., Lewis, M. A., and Volpe, J. P. (2005). Transmission dynamics of parasitic sea lice from farm to wild salmon. Proceedings of the Royal Society B: Biological Sciences, 272(1564), 689-696. https://doi.org/10.1098/rspb.2004.3027

Le Corre, N., Guichard, F., and Johnson, L. E. (2012). Connectivity as a management tool for coastal ecosystems in changing oceans. In M. Marcelli (Ed.), Oceanography (p. 235-258). IntechOpen. https://doi.org/10.5772/27704

Lhorente, J. P., Gallardo, J. A., Villanueva, B., Carabaño, M. J., and Neira, R. (2014). Disease resistance in Atlantic salmon (*Salmo salar*): Coinfection of the intracellular bacterial pathogen *Piscirickettsia salmonis* and the sea louse *Caligus*

rogercresseyi. PLoS ONE, 9(4), e95397-e95404. https://doi.org/10.1371/journal.pone.0095397

Lin, C.-L., and Ho, J. (1993). Life history of *Caligus epidemicus* Hewitt parasitic on tilapia (*Oreochromis mossambicus*) cultured in brackish water. In G. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 5-15). West Sussex, UK: Ellis Horwood Ltd.

Ljungfeldt, L. E. R., Quintela, M., Besnier, F., Nilsen, F., and Glover, K. A. (2017). A pedigree-based experiment reveals variation in salinity and thermal tolerance in the salmon louse, *Lepeophtheirus salmonis*. Evolutionary Applications, 10(10), 1007-1019. https://doi.org/10.1111/eva.12505

Long, A., Garver, K. A., and Jones, S. R. M. (2019). Synergistic osmoregulatory dysfunction during salmon lice (*Lepeophtheirus salmonis*) and infectious hematopoietic necrosis virus co-infection in sockeye salmon (*Oncorhynchus nerka*) smolts. Journal of Fish Diseases, 42(6), 869-882. https://doi.org/10.1111/jfd.12989

Love, D. C., Fry, J. P., Cabello, F., Good, C. M., and Lunestad, B. T. (2020). Veterinary drug use in United States net pen salmon aquaculture: Implications for drug use policy. Aquaculture, 518, 734820-734832. https://doi.org/10.1016/j.aquaculture.2019.734820

[map] Maine Department of Marine Resources. (nd). Aquaculture Leases. Accessed at: https://www.maine.gov/dmr/aquaculture/maine-aquaculture-leases-and-lpas/aquaculture-web-map on 2023-01-17.

Marine Harvest. (2018). Salmon Farming Industry Handbook. Marine Harvest ASA (p. 112). https://mowi.com/wp-content/uploads/2019/04/2018-salmon-industry-handbook-1.pdf

McClure, C., Hammell, K., Stryhn, H., Dohoo, I., and Hawkins, L. (2005). Application of surveillance data in evaluation of diagnostic tests for infectious salmon anemia. Diseases of Aquatic Organisms, 63, 119-127. https://doi.org/10.3354/dao063119

Middlemas, S. J., Fryer, R. J., Tulett, D., and Armstrong, J. D. (2013). Relationship between sea lice levels on sea trout and fish farm activity in Western Scotland. Fisheries Management and Ecology, 20(1), 68-74. https://doi.org/10.1111/fme.12010

Murray, A. G., and Gillibrand, P. A. (2006). Modelling salmon lice dispersal in Loch Torridon, Scotland. Marine Pollution Bulletin, 53, 128-135. https://doi.org/10.1016/j.marpolbul.2005.09.013

Murray, A. G., and Gubbins, M. (2016). Spatial management measures for disease mitigation as practiced in Scottish aquaculture. Marine Policy, 70, 93-100. https://doi.org/10.1016/j.marpol.2016.04.052

Murray, A. G., Ives, S. C., Smith, R. J., and Moriarty, M. (2021). A preliminary assessment of indirect impacts on aquaculture species health and welfare in Scotland during COVID-19 lockdown. Veterinary and Animal Science, 11. https://doi.org/10.1016/j.vas.2021.100167

Murray, A. G., and Salama, N. K. G. (2016). A simple model of the role of area management in the control of sea lice. Ecological Modelling, 337, 39-47. https://doi.org/10.1016/j.ecolmodel.2016.06.007

Mustafa, A., Conboy, G. A., and Burka, J. F. (2000a). Life-span and reproductive capacity of sea lice, *Lepeophtheirus salmonis*, under laboratory conditions. Special Publication – Aquaculture Association of Canada.

Mustafa, A., MacWilliams, C., Fernandez, N., Matchett, K., Conboy, G. A., and Burka, J. F. (2000b). Effects of sea lice (*Lepeophtheirus salmonis* Krøyer, 1837) infestation on macrophage functions in Atlantic salmon (*Salmo salar* L.). Fish and Shellfish Immunology, 10(1), 47-59. https://doi.org/10.1006/fsim.1999.0229

Mustafa, A., Rankaduwa, W., and Campbell, P. (2001). Estimating the cost of sea lice to salmon aquaculture in eastern Canada. Canadian Veterinary Journal, 42(1), 54–56. https://doi.org/10.1111/j.1365-2761.2008.01011.x

Myksvoll, M. S., Sandvik, A. D., Albretsen, J., Asplin, L., Johnsen, I. A., Karlsen, Ø., Kristensen, N. M., Melsom, A., Skardhamar, J., and Ådlandsvik, B. (2018). Evaluation of a national operational salmon lice monitoring system – From physics to fish. PLoS ONE, 13(7), 1-25. https://doi.org/10.1371/journal.pone.0201338

Nathan, R., Klein, E. K., Robledo-Arnuncio, J. J., and Revilla, E. (2012). Dispersal kernels: Review. In J. Clobert, M. Baguette, T. G. Benton, and J. M. Bullock (Eds.), Dispersal Ecology and Evolution (1st issue, pp. 187-210). Oxford University Press. https://doi.org/10.1093/acprof:oso/9780199608898.001.0001

National Farm Animal Care Council. (2021). Code of Practice for the Care and Handling of Farmed Salmonids. Ottawa, Canada: Canadian Aquaculture Industry Alliance.

Naylor, R. L., Hardy, R. W., Buschmann, A. H., Bush, S. R., Cao, L., Klinger, D. H., Little, D. C., Lubchenco, J., Shumway, S. E., and Troell, M. (2021). A 20-year retrospective review of global aquaculture. Nature, 591(7851), 551-563. https://doi.org/10.1038/s41586-021-03308-6

Nekouei, O., Vanderstichel, R., Thakur, K., Arriagada, G., Patanasatienkul, T., Whittaker, P., Milligan, B., Stewardson, L., and Revie, C. W. (2018). Association between sea lice (*Lepeophtheirus salmonis*) infestation on Atlantic salmon farms and wild Pacific salmon in Muchalat Inlet, Canada. Scientific Reports, 8(1). https://doi.org/10.1038/s41598-018-22458-8

Nelson, E. J., Robinson, S. M. C., Feindel, N., Sterling, A., Byrne, A., and Pee Ang, K. (2017). Horizontal and vertical distribution of sea lice larvae (*Lepeophtheirus salmonis*) in and around salmon farms in the Bay of Fundy, Canada. Journal of Fish Diseases, 41(6), 885-899. https://doi.org/10.1111/jfd.12692

Nese, L., and Enger, O. (1993). Isolation of *Aeromonas salmonicida* from salmon lice *Lepeophtheirus salmonis* and marine plankton. Diseases of Aquatic Organisms, 16, 79-81.

New Brunswick Department of Agriculture, Aquaculture and Fisheries. (2022). New Brunswick Finfish Aquaculture Growth Strategy 2022-2030. Fredericton, Canada: New Brunswick Department of Agriculture, Aquaculture and Fisheries.

New Brunswick Department of Agriculture, Fisheries and Ocean. (2013). Bay of Fundy Marine Aquaculture Site Allocation Policy. Accessed at: http://www.gnb.ca/0177/e-fundy.asp#amend on 2018-01-11

[map] New Brunswick Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). Bay of Fundy Marine Aquaculture Sites. 1:125000. New Brunswick, Canada.

Newfoundland and Labrador Department of Fisheries, Forestry and Agriculture. (2022). Seafood Industry Year in Review 2021. St. John's, Canada: Newfoundland and Labrador Department of Fisheries, Forestry and Agriculture.

Niggemann, M., Wiegand, T., Robledo-Arnuncio, J. J., and Bialozyt, R. (2012). Marked point pattern analysis on genetic paternity data for uncertainty assessment of pollen dispersal kernels. Journal of Ecology, 100, 264-276. https://doi.org/10.1111/j.1365-2745.2011.01854.x

Niklitschek, E. J., Soto, D., Lafon, A., Molinet, C., and Toledo, P. (2013). Southward expansion of the Chilean salmon industry in the Patagonian Fjords: Main environmental challenges. Reviews in Aquaculture, 5(3), 172-195. https://doi.org/10.1111/raq.12012

Nilsson, J., Barrett, L. T., Mangor-Jensen, A., Nola, V., Harboe, T., and Folkedal, O. (2023). Effect of water temperature and exposure duration on detachment rate of salmon lice (*Lepeophtheirus salmonis*); Testing the relevant thermal spectrum used for delousing. Aquaculture, 562, 738879-738886. https://doi.org/10.1016/j.aquaculture.2022.738879

Nylund, A., Hovland, T., Hodneland, K., Nilsen, F., and Lovik, P. (1994). Mechanisms for transmission of infectious salmon anaemia (ISA). Diseases of Aquatic Organisms, 19(2), 95–100. https://doi.org/10.3354/dao019095

Nylund, A., Wallace, C., and Hovland, T. (1993). The possible role of *Lepeophtheirus salmonis* (Krøyer) in the transmission of infectious salmon

anaemia. In G. A. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (pp. 367–373). Ellis Horwood.

Overton, K., Dempster, T., Oppedal, F., Kristiansen, T. S., Gismervik, K., and Stien, L. H. (2019). Salmon lice treatments and salmon mortality in Norwegian aquaculture: A review. Reviews in Aquaculture, 11(4), 1398-1417. https://doi.org/10.1111/raq.12299

Parsons, A. E., Escobar-Lux, R. H., Sævik, P. N., Samuelsen, O. B., and Agnalt, A. L. (2020). The impact of anti-sea lice pesticides, azamethiphos and deltamethrin, on European lobster (*Homarus gammarus*) larvae in the Norwegian marine environment. Environmental Pollution, 264, 114725-114736. https://doi.org/10.1016/j.envpol.2020.114725

Patanasatienkul, T., Sanchez, J., Rees, E. E., Krkošek, M., Jones, S. R. M., and Revie, C. W. (2013). Sea lice infestations on juvenile chum and pink salmon in the Broughton Archipelago, Canada, from 2003 to 2012. Diseases of Aquatic Organisms, 105(2), 149-161. https://doi.org/10.3354/dao02616

Patanasatienkul, T., Sanchez, J., Rees, E. E., Pfeiffer, D., and Revie, C. W. (2015). Space-time cluster analysis of sea lice infestation (*Caligus clemensi* and *Lepeophtheirus salmonis*) on wild juvenile Pacific salmon in the Broughton Archipelago of Canada. Preventive Veterinary Medicine, 120(2), 219-231. https://doi.org/10.1016/j.prevetmed.2015.03.006

Penston, M. J., McKibben, M.A., Hay, D.W., Gillibrand, P.A. (2004). Observations on open-water densities of sea lice larvae in Loch Shieldaig, Western Scotland. Aquaculture Research, 35, 793−805. https://doi.org/10.1111/j.1365-2109.2004.01102.x

Penston M. J., McBeath, A. J. A., and Millar, C. P. (2011). Densities of planktonic *Lepeophtheirus salmonis* before and after an Atlantic salmon farm relocation. Aquaculture Environment Interactions, 1, 225−232. https://doi.org/10.3354/aei00022

Pert, C., Fryer, R., Cook, P., Kilburn, R., McBeath, S., McBeath, A., Matejusova, I., Urquhart, K., Weir, S., McCarthy, U., Collins, C., Amundrud, T., and Bricknell, I. (2014). Using sentinel cages to estimate infestation pressure on salmonids from sea lice in Loch Shieldaig, Scotland. Aquaculture Environment Interactions, 5(1), 49-59. https://doi.org/10.3354/aei00094

Pest Management Regulatory Agency (PMRA), Government of Canada. (2003). Integrated Pest Management. Overview: Integrated Pest Management of Sea Lice in Salmon Aquaculture: Overview. Health Canada, Ottawa, ON. Catalogue H114-9/2003E. publications.gc.ca/pub?id=9.686494&sl=0

Peterson, R. H., Page, F., Steeves, G. D., Wildish, D. J., Harmon, P., and Losier, R. (2001). A survey of 20 Atlantic salmon farms in the Bay of Fundy: Influence of environmental and husbandry variables on performance. Canadian Technical

Report of Fisheries and Aquatic Sciences 2337. Ottawa, ON: Government of Canada Fisheries and Oceans.

Piasecki, W. (1996). The developmental stages of *Caligus elongatus* von Nordmann, 1832 (Copepoda: Caligidae). Canadian Journal of Zoology, 74(8), 1459-1478. https://doi.org/10.1139/z96-161

Piasecki, W., Venmathi Maran, B. A., and Ohtsuka, S. (2023). Are we ready to get rid of the terms “chalimus” and “preadult” in the caligid (Crustacea: Copepoda: Caligidae) life cycle nomenclature? Pathogens, 12(3), 460-485. https://doi.org/10.3390/pathogens12030460

Pike, A. W., Mordue, A. J., and Ritchie, G. (1993). The development of *Caligus elongatus* Nordmann from hatching to copepodid in relation to temperature. In G. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 51-60). West Sussex, UK: Ellis Horwood Ltd.

Pike, A. W., and Wadsworth, S. L. (1999). Sea lice on salmonids: Their biology and control. Advances in parasitology, 44, 233-337. https://doi.org/10.1016/S0065-308X(08)60233-X

Pirhonen, J., Schreck, C. B., Gannam, A. (2000). Appetite of Chinook salmon (*Oncorhynchus tshawytscha*) naturally infected with bacterial kidney disease. Aquaculture, 189, 1-10.

Price, D., Ibarra, R., Sánchez, J., and St-Hilaire, S. (2017). A retrospective assessment of the effect of fallowing on piscirickettsiosis in Chile. Aquaculture, 473, 400-406. https://doi.org/10.1016/j.aquaculture.2017.02.034

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

Rae, G. (1999). Sea lice, medicines and a national strategy for control. Fish Veterinary Journal, 3, 46-51.

Raynard, R. S., Bricknell, I. R., Billingsley, P. F., Nisbet, A. J., Vigneau, A., and Sommerville, C. (2002). Development of vaccines against sea lice. Pest Management Science, 58(6), 569-575. https://doi.org/10.1002/ps.474

Rees, E. E., Ibarra, R., Medina, M., Sanchez, J., Jakob, E., Vanderstichel, R., and St-Hilaire, S. (2014). Transmission of *Piscirickettsia salmonis* among salt water salmonid farms in Chile. Aquaculture, 428-429, 189-194. https://doi.org/10.1016/j.aquaculture.2014.03.031

Rees, E. E., St-Hilaire, S., Jones, S. R. M., Krkošek, M., DeDominicis, S., Foreman, M. G. G., Patanasatienkul, T., and Revie, C. W. (2015). Spatial patterns of sea lice infection among wild and captive salmon in Western Canada. Landscape Ecology, 30(6), 989-1004. https://doi.org/10.1007/s10980-015-0188-2

Revie, C. W., Robbins, C., Gettinby, G., Kelly, L., and Treasurer, J. W. (2005). A mathematical model of the growth of sea lice, *Lepeophtheirus salmonis*, populations on farmed Atlantic salmon, *Salmo salar* L., in Scotland and its use in the assessment of treatment strategies. Journal of Fish Diseases, 28(10), 603-614. https://doi.org/10.1111/j.1365-2761.2005.00665.x

Revie, C. W., Gettinby, G., Treasurer, J. W., Rae, G. H., and Clark, N. (2002). Temporal, environmental and management factors influencing the epidemiological patterns of sea lice (*Lepeophtheirus salmonis*) infestations on farmed Atlantic salmon (*Salmo salar*) in Scotland. Pest Management Science, 58(6), 576-584. https://doi.org/10.1002/ps.476

Revie, C. W., Gettinby, G., Treasurer, J. W., and Wallace, C. (2003). Identifying epidemiological factors affecting sea lice *Lepeophtheirus salmonis* abundance on Scottish salmon farms using general linear models. Diseases of Aquatic Organisms, 57(1-2), 85-95. https://doi.org/10.3354/dao057085

Ritchie, G., Mordue, A. J., Pike, A. W., and Rae, G. H. (1993). The reproductive output of *Lepeophtheirus salmonis* adult females in relation to seasonal variability of temperature and photoperiod. In G. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 153-165). West Sussex, UK: Ellis Horwood Ltd.

Roberge, C., Einum, S., Guderley, H., and Bernatchez, L. (2006). Rapid parallel evolutionary changes of gene transcription profiles in farmed Atlantic salmon. Molecular Ecology, 15(1), 9-20. https://doi.org/10.1111/j.1365-294X.2005.02807.x

[dataset] Robinson, S. Data of: Passamaquoddy Bay monthly Conductivity Temperature and Depth (CTD) sampling (1989 - 2018). Published: October 2019. Coastal Ecosystems Science Division, Fisheries and Oceans Canada, St. Andrews, NB. https://open.canada.ca/data/en/dataset/12184962-7879-4214-aef0-b31162f04a27

Robledo, D., Gutiérrez, A. P., Barría, A., Yáñez, J. M., and Houston, R. D. (2018a). Gene expression response to sea lice in Atlantic salmon skin: RNA sequencing comparison between resistant and susceptible animals. Frontiers in Genetics, 9(AUG). https://doi.org/10.3389/fgene.2018.00287

Robledo, D., Matika, O., Hamilton, A., and Houston, R. D. (2018b). Genome-wide association and genomic selection for resistance to amoebic gill disease in Atlantic salmon. G3: Genes, Genomes, Genetics, 8(4), 1195-1203. https://doi.org/10.1534/g3.118.200075

Ross, N., Firth, K., Wang, A., Burka, J., and Johnson, S. (2000). Changes in hydrolytic enzyme activities of naïve Atlantic salmon *Salmo salar* skin mucus due to infection with the salmon louse *Lepeophtheirus salmonis* and cortisol implantation. Diseases of Aquatic Organisms, 41, 43-51. https://doi.org/10.3354/dao041043

Saksida, S., Constantine, J., Karreman, G. A., and Donald, A. (2007). Evaluation of sea lice abundance levels on farmed Atlantic salmon (*Salmo salar* L.) located in the Broughton Archipelago of British Columbia from 2003 to 2005. Aquaculture Research, 38(3), 219-231. https://doi.org/10.1111/j.1365-2109.2007.01651.x

Saksida, S. M., Morrison, D., and Revie, C. W. (2010). The efficacy of emamectin benzoate against infestations of sea lice, *Lepeophtheirus salmonis*, on farmed Atlantic salmon, *Salmo salar* L., in British Columbia. Journal of Fish Diseases, 33(11), 913-917. https://doi.org/10.1111/j.1365-2761.2010.01192.x

Salama, N. K. G., Collins, C. M., Fraser, J. G., Dunn, J., Pert, C. C., Murray, A. G., and Rabe, B. (2013). Development and assessment of a biophysical dispersal model for sea lice. 36, 323-337. https://doi.org/10.1111/jfd.12065

Salama, N. K. G., Dale, A. C., Ivanov, V. V., Cook, P. F., Pert, C. C., Collins, C. M., and Rabe, B. (2018). Using biological-physical modelling for informing sea lice dispersal in Loch Linnhe, Scotland. Journal of Fish Diseases, 41(6), 901-919. https://doi.org/10.1111/jfd.12693

Salama, N. K. G., Murray, A. G., and Rabe, B. (2016). Simulated environmental transport distances of *Lepeophtheirus salmonis* in Loch Linnhe, Scotland, for informing aquaculture area management structures. Journal of Fish Diseases, 39(4), 419-428. https://doi.org/10.1111/jfd.12375

Salama, N. K. G., and Rabe, B. (2013). Developing models for investigating the environmental transmission of disease-causing agents within open-cage salmon aquaculture. Aquaculture Environment Interactions, 4(2), 91-115. https://doi.org/10.3354/aei00077

Sale, P. F., Hanski, I., and Kritzer, J. P. (2006). The Merging of Metapopulation Theory and Marine Ecology: Establishing the Historical Context. In J. P. Kritzer and P. F. Sale (Eds.), Marine Metapopulations (pp. 3-28). Burlington, USA: Elsevier. https://doi.org/10.1016/B978-0-12-088781-1.50004-2

Samsing, F., Johnsen, I., Dempster, T., Oppedal, F., and Treml, E. A. (2017). Network analysis reveals strong seasonality in the dispersal of a marine parasite and identifies areas for coordinated management. Landscape Ecology, 32(10), 1953-1967. https://doi.org/10.1007/s10980-017-0557-0

Samsing, F., Johnsen, I., Treml, E. A., and Dempster, T. (2019). Identifying ‘firebreaks’ to fragment dispersal networks of a marine parasite. International Journal for Parasitology, 49(3-4), 277-286. https://doi.org/10.1016/j.ijpara.2018.11.005

Samsing, F., Oppedal, F., Dalvin, S., Johnsen, I., Vågseth, T., and Dempster, T. (2016). Salmon lice (*Lepeophtheirus salmonis*) development times, body size, and reproductive outputs follow universal models of temperature dependence. Canadian Journal of Fisheries and Aquatic Sciences, 73, 1841-1851. https://doi.org/10.1139/cjfas-2016-0050

Sandell, T. A., Teel, D. J., Fisher, J., Beckman, B., Jacobson, K. C. (2015). Infections by *Renibacterium salmoninarum* and *Nanophyetus salmincola* Chaplin are associated with reduced growth of juvenile Chinook salmon, *Oncorhynchus tshawytscha* (Walbaum), in the Northeast Pacific Ocean. Journal of Fish Diseases, 38, 365-378. 10.1111/jfd.12243

Sandvik, A. D., Bjørn, P. A., Ådlandsvik, B., Asplin, L., Skardhamar, J., Johnsen, I. A., Myksvoll, M., and Skogen, M. D. (2016a). Toward a model-based prediction system for salmon lice infestation pressure. Aquaculture Environment Interactions, 8, 527-542. https://doi.org/10.3354/AEI00193

Sandvik, A. D., Johnsen, I. A., Myksvoll, M. S., Sævik, P. N., and Skogen, M. D. (2020). Prediction of the salmon lice infestation pressure in a Norwegian fjord. ICES Journal of Marine Science, 77(2), 746-756. https://doi.org/10.1093/icesjms/fsz256

Sandvik, A. D., Skagseth, Ø., and Skogen, M. D. (2016b). Model validation: Issues regarding comparisons of point measurements and high-resolution modeling results. Ocean Modelling, 106, 68-73. https://doi.org/10.1016/j.ocemod.2016.09.007

Sandvik, A. D., Dalvin, S., Skern-Mauritzen, R., and Skogen, M. D. (2021). The effect of a warmer climate on the salmon lice infection pressure from Norwegian aquaculture. ICES Journal of Marine Science, 78(5), 1849-1859. https://doi.org/10.1093/icesjms/fsab069

Schmid-Hempel, P., and Frank, S. A. (2007). Pathogenesis, virulence, and infective dose. PLoS Pathogens, 3(10), 1372-1373. https://doi.org/10.1371/journal.ppat.0030147

Schram, T. A. (1993). Supplementary descriptions of the developmental stages of *Lepeophtheirus salmonis* (Krøyer, 1837) (Copepoda: Caligidae). In G. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (2nd ed., pp. 30-47). West Sussex, UK: Ellis Horwood Ltd.

Schram, T. A. (2004). Practical identification of pelagic sea lice larvae. Journal of Marine Biological Association of the United Kingdom, 84, 13-110. https://doi.org/10.1017/S0025315404008963h

SEARCH Consortium. (2006). Sea lice resistance to chemotherapeutants: A handbook in resistance management. Sacramento, USA.

Siegel, D. A., Kinlan, B. P., Gaylord, B., and Gaines, S. D. (2003). Lagrangian descriptions of marine larval dispersion. Marine Ecology Progress Series, 260, 83-96. https://doi.org/10.3354/meps260083

Skaala, Ø., Taggart, J. B., and Gunnes, K. (2005). Genetic differences between five major domesticated strains of Atlantic salmon and wild salmon. Journal of Fish Biology, 67, 118-128. https://doi.org/10.1111/j.0022-1112.2005.00843.x

Skarðhamar, J., Fagerli, M. N., Reigstad, M., Sandvik, A. D., and Bjørn, P. A. (2019). Sampling planktonic salmon lice in Norwegian fjords. Aquaculture Environment Interactions, 11, 701-715. https://doi.org/10.3354/AEI00342

Skern-Mauritzen, R., Torrissen, O., and Glover, K. A. (2014). Pacific and Atlantic *Lepeophtheirus salmonis* (Krøyer, 1838) are allopatric subspecies: *Lepeophtheirus salmonis salmonis* and *L. salmonis oncorhynchi* subspecies novo. BMC Genetics, 15, 1-9. https://doi.org/10.1186/1471-2156-15-32

Song, X., Liu, Y., Pettersen, J. B., Brandão, M., Ma, X., Røberg, S., and Frostell, B. (2019). Life cycle assessment of recirculating aquaculture systems: A case of Atlantic salmon farming in China. Journal of Industrial Ecology, 23(5), 1077–1086. https://doi.org/10.1111/jiec.12845

St-Hilaire, S., Patanasatienkul, T., Yu, J., Kristoffersen, A. B., Stryhn, H., Revie, C., Ibarra, R., Tello, A., and McEwan, G. (2018). Tool for predicting *Caligus rogercresseyi* abundance on salt water salmon farms in Chile. Preventative Veterinary Medicine, 158, 122-128. https://doi.org/https://doi.org/10.1016/j.prevetmed.2018.08.001

StataCorp (2019). *Stata: Release 16.* Statistical Software. StataCorp LLC, TX, USA.

[dataset] Statistics Canada. (2022). Aquaculture Statistics. Catalogue No. 32-10-0107-01

Stene, A., Hellevik, C. C., Fjørtoft, H. B., and Philis, G. (2022). Considering elements of natural strategies to control salmon lice infestation in marine cage culture. Aquaculture Environment Interactions, 14, 181-188. https://doi.org/10.3354/aei00436

Stien, A., Bjørn, P. A., Heuch, P. A., and Elston, D. A. (2005). Population dynamics of salmon lice *Lepeophtheirus salmonis* on Atlantic salmon and sea trout. Marine Ecology Progress Series, 290, 263-275.

Stucchi, D. J., Guo, M., Foreman, M. G. G., Czajko, P., Galbraith, M., Mackas, D. L., and Gillibrand, P. A. (2011). Modeling Sea Lice Production and Concentrations in the Broughton Archipelago, British Columbia. In S. Jones and R. Beamish (Eds.), Salmon Lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). John Wiley and Sons, Inc.

Swain, J. K., Carpio, Y., Johansen, L. H., Velazquez, J., Hernandez, L., Leal, Y., Kumar, and Estrada, M. P. (2020). Impact of a candidate vaccine on the dynamics of salmon lice (*Lepeophtheirus salmonis*) infestation and immune response in Atlantic salmon (*Salmo salar* L.). PLoS ONE, 15, e0239827-e0239838. https://doi.org/10.1371/journal.pone.0239827

Tischendorf, L., and Fahrig, L. (2000). On the usage and measurement of landscape connectivity. Oikos, 90(1), 7-19. https://www.jstor.org/stable/3547105?seq=1andcid=pdf-

Thakur, K. K., Vanderstichel, R., Barrell, J., Stryhn, H., Patanasatienkul, T., and Revie, C. W. (2018). Comparison of remotely-sensed sea surface temperature and salinity products with in situ measurements from British Columbia, Canada. Frontiers in Marine Science, 5, 1-11. https://doi.org/10.3389/fmars.2018.00121

Todd, C.D., Walker, A. M., Ritchie, M. G., Graves, J. A. and Walker, A. F. (2004) Population genetic differentiation of sea lice (*Lepeophtheirus salmonis*) parasitic on Atlantic and Pacific salmonids: Analyses of microsatellite DNA variation among wild and farmed hosts. Canadian Journal of Fisheries and Aquatic Sciences 61, 1176-1190.

Toorians, M. E. M., and Adams, T. P. (2020). Critical connectivity thresholds and the role of temperature in parasite metapopulations. Ecological Modelling, 435, 109258-109267. https://doi.org/10.1016/j.ecolmodel.2020.109258

Torrissen, O., Jones, S., Asche, F., Guttormsen, A., Skilbrei, O. T., Nilsen, F., Horsberg, T. E., and Jackson, D. (2013). Salmon lice – Impact on wild salmonids and salmon aquaculture. Journal of Fish Diseases, 36(3), 171-194. https://doi.org/10.1111/jfd.12061

Treasurer, J. W., and Pope, J. A. (2000). Selection of host sample number and design of a monitoring programme for ectoparasitic sea lice (Copepoda: Caligidae) on farmed Atlantic salmon, *Salmo salar*. Aquaculture, 187, 247–260. www.elsevier.nlrlocateraqua-online

Tsai, H.-Y., Hamilton, A., Tinch, A. E., Guy, D. R., Bron, J. E., Taggart, J. B., Gharbi, K., Stear, M., Matika, O., Pong-Wong, R. (2016). Genomic prediction of host resistance to sea lice in farmed Atlantic salmon populations. Genetic Selection Evolution. 48(1), 47-58. https://doi.org/10.1186/s12711-016-0226-9

Tucker, C. S., Sommerville, C., and Wootten, R. (2000). The effect of temperature and salinity on the settlement and survival of copepodids of *Lepeophtheirus salmonis* (Krøyer, 1837) on Atlantic salmon, *Salmo salar* L. Journal of Fish Diseases, 23(5), 309-320. https://doi.org/10.1046/j.1365-2761.2000.00219.x

Tully O. (1989). The succession of generations and growth of the caligid copepods *Caligus elongatus* and *Lepeophtheirus salmonis* parasitising farmed Atlantic salmon smolts (*Salmo salar*). Journal of Marine Biological Association of the United Kingdom, 69, 279-287.

Tully, O., and Nolan, D. T. (2002). A review of the population biology and host-parasite interactions of the sea louse *Lepeophtheirus salmonis* (Copepoda: Caligidae). Parasitology, 124, S165-S182. https://doi.org/10.1017/S0031182002001889

Tully, O., and Whelan, K. F. (1993). Production of nauplii of *Lepeophtheirus salmonis* (Krøyer) (Copepoda: Caligidae) from farmed and wild salmon and its relation to the infestation of wild sea trout (*Salmo trutta* L.) off the West Coast of Ireland in 1991. Fisheries Research, 17(1-2), 187-200. https://doi.org/10.1016/0165-7836(93)90018-3

[dataset] UK Met Office (2005). OSTIA Level 4 Sea Surface Temperature Analysis. Ver. 1.0. PO.DAAC, CA, USA. Retrieved from https://doi.org/10.5067/GHOST-4FK01.

Ulgenes, I. (2018). Possible underestimation of salmon lice (*Lepeophtheirus salmonis*) abundance using sentinel cages. Master thesis, Inland Norway University of Applied Sciences, Hamar. http://hdl.handle.net/11250/2585127

[dataset] U.S. Bureau of Labor Statistics. (2023). Consumer Price Index. Series Id CUUR0000SA0.

Valenzuela-Muñoz, V., Gallardo-Escárate, C., Valenzuela-Miranda, D., Nuñez-Acuña, G., Benavente, B. P., Alert, A., and Arevalo, M. (2023). Transcriptome signatures of Atlantic salmon – Resistant phenotypes against sea lice infestation are associated with tissue repair. Genes, 14(5), 986. https://doi.org/10.3390/genes14050986

Valdes-Donoso, P., Mardones, F. O., Jarpa, M., Ulloa, M., Carpenter, T. E., and Perez, A. M. (2013). Co-infection patterns of infectious salmon anaemia and sea lice in farmed Atlantic salmon, *Salmo salar* L., in Southern Chile (2007-2009). Journal of Fish Diseases, 36(3), 353-360. https://doi.org/10.1111/jfd.12070

Van Nouhuys, S. (2016). Metapopulation Ecology. ELS, 2009, 1-9. https://doi.org/10.1002/9780470015902.a0021905.pub2

Viljugrein, H., Staalstrøm, A., Molvær, J., Urke, H. A., and Jansen, P. A. (2009). Integration of hydrodynamics into a statistical model on the spread of pancreas disease (PD) in salmon farming. Diseases of Aquatic Organisms, 88, 35-44. https://doi.org/10.3354/dao02151

Vormedal, I., and Larsen, M. L. (2021). Regulatory Processes for Setting Sensitive-Period Sea-lice Thresholds in Major Salmon Producer Jurisdictions: An Evaluation. Aquaculture Stewardship Council.

Wallace, C. (1998). Possible causes of salmon lice *Lepeophtheirus salmonis* (Krøyer, 1837) infections on farmed Atlantic salmon, *Salmo salar* L., in a Western Norwegian fjord-situated fish farm: Implications for the design of regional management strategies. Master thesis, University of Bergen, Norway. University of Bergen Library.

Walter, T.C. and Boxshall, G. (2023). World of Copepods Database. *Caligus rogercresseyi* Boxshall and Bravo, 2000. Accessed through: World Register of Marine Species at:

https://www.marinespecies.org/aphia.php?p=taxdetails&id=135566 on 2023-08-18.

Wang, I. J. (2020). Topographic path analysis for modelling dispersal and functional connectivity: Calculating topographic distances using the topoDistance package. Methods in Ecology and Evolution, 11(2), 265-272. https://doi.org/10.1111/2041-210X.13317

Werkman, M., Green, D. M., Murray, A. G., and Turnbull, J. F. (2011). The effectiveness of fallowing strategies in disease control in salmon aquaculture assessed with an SIS model. Preventive Veterinary Medicine, 98(1), 64-73. https://doi.org/10.1016/j.prevetmed.2010.10.004

Westcott, J. D., Hammell, K. L., and Burka, J. F. (2004). Sea lice treatments, management practices and sea lice sampling methods on Atlantic salmon farms in the Bay of Fundy, New Brunswick, Canada. Aquaculture Research, 35(8), 784-792. https://doi.org/10.1111/j.1365-2109.2004.01101.x

Westcott, J. D., Stryhn, H., Burka, J. F., and Hammell, K. L. (2008). Optimization and field use of a bioassay to monitor sea lice *Lepeophtheirus salmonis* sensitivity to emamectin benzoate. Diseases of Aquatic Organisms, 79, 119-131. https://doi.org/10.3354/dao01887

Whyte, S. K., Westcott, J. D., Jimenez, D., Revie, C. W., and Hammell, K. L. (2014). Assessment of sea lice (*Lepeophtheirus salmonis*) management in New Brunswick, Canada using deltamethrin (Alphamax®) through clinical field treatment and laboratory bioassay responses. Aquaculture, 422-423, 54-62. https://doi.org/10.1016/j.aquaculture.2013.11.027

Withler, R. E., Supernault, K. J., and Miller, K. M. (2005). Genetic variation within and among domesticated Atlantic salmon broodstocks in British Columbia, Canada. Animal Genetics, 36(1), 43-50. https://doi.org/10.1111/j.1365-2052.2004.01220.x

Wootten, R., Smith, J. W., and Needham, E. A. (1982). Aspects of the biology of the parasitic copepods *Lepeophtheirus salmonis* and *Caligus elongatus* on farmed salmonids, and their treatment. Proceedings of the Royal Society of Edinburgh: Section B. Biological Sciences, 81(3), 185-197. https://doi.org/10.1017/s0269727000003389

Wright, D. W., Oppedal, F., and Dempster, T. (2016). Early-stage sea lice recruits on Atlantic salmon are freshwater sensitive. Journal of Fish Diseases, 39(10), 1179-1186. https://doi.org/10.1111/jfd.12452

Yazawa, R., Yasuike, M., Leong, J., von Schalburg, K. R., Cooper, G. A., Beetz-Sargent, M., Robb, A., Davidson, W. S., Jones, S. R. M., and Koop, B. F. (2008). EST and mitochondrial DNA sequences support a distinct Pacific form of salmon louse, *Lepeophtheirus salmonis*. Marine Biotechnology, 10(6), 741-749. https://doi.org/10.1007/s10126-008-9112-y

Zalcman, E., Burroughs, A., Meyer, A., Hillman, A., Sadler, R., Madin, B., Mackenzie, C., Ward, M. P., Stevenson, M., Happold, J., Hutchison, J., Gallardo Lagno, A. L., Cameron, A., and Cowled, B. (2021). Sea lice infestation of salmonids in Chile between 2011 and 2017: Use of regulatory data to describe characteristics and identify risk factors. Aquaculture, 530, 735752-735763. https://doi.org/10.1016/j.aquaculture.2020.735752

2 Atlantic salmon aquaculture industry in the Bay of Fundy, New Brunswick

2.1 Introduction

2.1.1 Bay of Fundy, New Brunswick coast

2.1.1.1 Location

The Bay of Fundy is a large recess of the Gulf of Maine off the North Atlantic Ocean. Its greatest depth is less than 150 m and generally less than 50 m with an irregular sea bottom (Buzeta et al., 2003). The area considered is the southwestern shore of New Brunswick (NB), Canada, located in the Bay of Fundy directly east of the Canadian/American border. It is part of the Northwest Atlantic Fisheries Organization (NAFO) area 4X with a marine surface area of 5,129 km^2 (Chang et al., 2007). Salmon aquaculture is prescribed in the Bay Management Areas (BMA; Figure 2.1 and Table 2.1). In Maine, USA, aquaculture occurs in Cobscook Bay and around Moose Island (Department of Energy and Resource Development/Agriculture, 2017) and was not included in the study. Hereafter, the study area is referred to as the Bay of Fundy, NB.

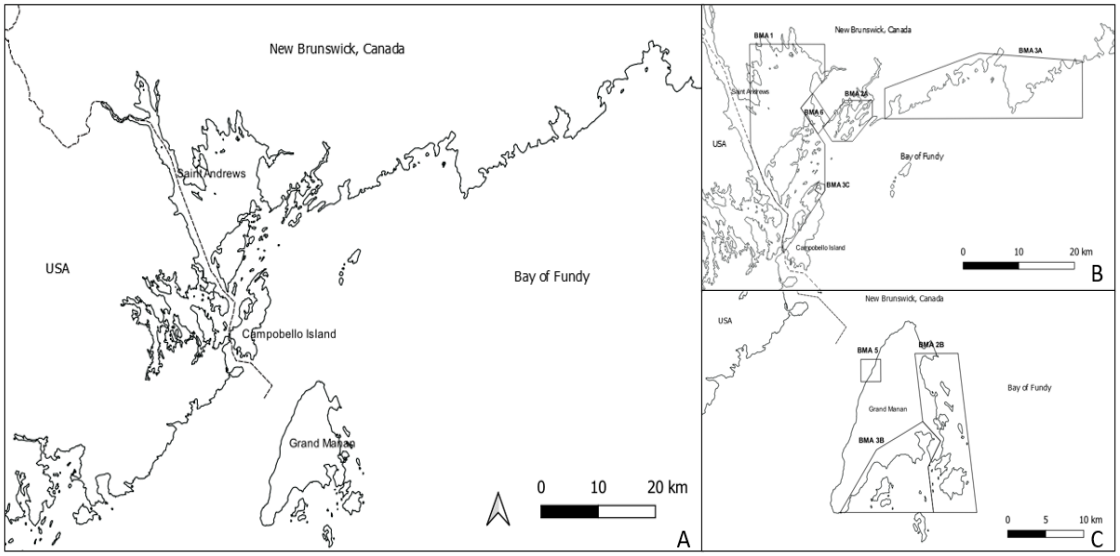

Figure 2.1. The Bay of Fundy, New Brunswick, Canada, study area (A) and Bay Management Areas (BMA) in the southwest coast and Passamaquoddy Bay (B) and Grand Manan Island (C). The locations of the BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (Department of Energy and Resource Development/Agriculture, and Fisheries, 2017).

Table 2.1. Published (Chang et al., 2007; Department of Energy and Resource Development/Agriculture, 2017) stocking years f(shaded area) or the culture of Atlantic salmon within Bay Management Areas (BMA). BMA 4 2006-2010 (Jones et al., 2012); same location, BMA 3C 2017-present.

BMA	Location	Year 20XX									
		09	10	11	12	13	14	15	16	17	18
1	Passamaquoddy Bay, Deer Island, most of Campobello Island										
2A	Back Bay, Bliss Harbour, Lime Kiln Bay										
2B	Eastern Grand Manan Island, White Head Island										
3A	Eastern mainland coast of Southwest NB from Beaver Harbour to Haley's Cove										
3B	Southern Grand Manan Island, excluding White Head Island										
4/3C	Anse Sawyers Cove										
5	Dark Harbour, Grand Manan Island										
6	Letete Passage										

2.1.1.2 Water temperature

The water temperature of the Bay of Fundy has a broad range that varies daily and according to the tide, depth, season, and distance from shore. Typically, water temperature measurements are at their lowest point in February and March at approximately 1 °C and peak at approximately 15 °C in August and September (Peterson et al., 2001; Jones et al., 2012; Gautam et al., 2016). The range of temperature is wide and may differ from other areas; Norway has a similar range of 3 to 18 °C (Aldrin et al., 2013; Asplin et al., 2014), but Chile has a narrower range of 6 to 18 °C (Zalcman et al., 2021).

The lower limit of the water temperature range of the Bay of Fundy extends beyond the range of water temperatures investigated for effects on individual sea lice under laboratory conditions, which has usually ranged from 3 to 24 °C (Stien et al., 2005; Samsing et al., 2016; Hamre et al., 2019). The effects of water temperature on sea lice are complex as reviewed by Brooker et al. (2018). Water temperature impacts the fecundity, rates of egg hatching, rates of life stage development, rates of survival, and behaviour of sea lice (Brooker et al., 2018). The overall conclusion is that, within the temperature range normally encountered in fish farming, sea lice generally develop and reproduce at a more rapid rate at higher water temperatures (Stien et al., 2005; Samsing et al., 2016; Hamre et al., 2019).

The impacts of water temperature on these biological processes thus affects the dispersal of sea lice. At warmer water temperatures, the planktonic life stages of

the sea louse develop and expend their energy reserves more rapidly therefore have a shorter time in which to disperse (Pike and Wadsworth, 1999). Conversely, at colder water temperatures, planktonic lice develop slowly which results in the opportunity to disperse greater distances but have a greater probability of mortality (Graham et al., 2008; Samsing et al., 2016).

Another consideration for water temperature is the treatment response. In the Bay of Fundy, topically applied treatments produced a more immediate change in sea lice abundance when temperatures were above 10 °C (Gautam et al., 2017b). In the autumn, as defined as after August 31 and when water temperatures were under 10 °C, there was low post-treatment abundance of sea lice (Gautam et al., 2017b). Sea lice abundance at temperatures under 10 °C and after August 31 also preserve/provide more time for the measurement of the treatment effect.

2.1.1.3 Salinity

The salinity of the Bay of Fundy is generally constant. At the mouth of the Bay of Fundy, the water salinity was measured at 33 ppt (Greenberg et al., 1984). At the mouth of the Passamaquoddy Bay (at site Prince 5), the mean minimum surface salinity was measured at 30.5 ppt (standard deviation [SD] 1.30 ppt) in May; otherwise, salinity was above 31 ppt for all measured depths and for all months of the year (Drinkwater, 1987). Previous research in sea lice in the Bay of Fundy (Jones et al., 2012; Elmoslemany et al., 2013; Gautam et al., 2016, 2017a,b; Elghafghuf et al., 2020) did not comment on the salinity of the area presumably

due to its consistency, and lack of available data. Salinity data were not captured in the Fish-iTrends database.

At 30 ppt, the salinity of the area is optimal for population growth of *L. salmonis* (Bricknell et al., 2006). Temporary reductions in salinity may occur due to freshet events, which decrease surface salinity due to the increased input of freshwater from estuaries secondary to large rainfall and snow melting events. Freshet events occur primarily at the St. Croix River estuary (Fife et al., 2015) and Magaguadavic River, but the effect on the study area is expected to be isolated spatially and temporally. The effects of reduced salinity on sea lice were reviewed by Brooker et al. (2018); salinities below 29 ppt appear to reduce survival rates, reduce fecundity, increase rates of life stage development, and change the behaviour of the sea lice. The effects of salinity lensing were observed in British Columbia, where freshet events affected an increased risk of wild juvenile salmonids having lice as salinity increased until a maximum risk at ≥ 15 ppt (Elmoslemany et al., 2015). In farmed salmonids, Arriagada et al. (2016) observed short (< 30 ppt) and long-term effects on mobile lice abundances; reduced salinities 15 to 45 days prior likely decreased copepodid and chalimus abundances thereby affecting adult lice abundances. Lower salinities measured 61 to 75 days prior may have affected a decrease in the mobile lice abundances by reducing the mating/fecundity success of the parents of that cohort (Arriagada et al., 2016).

2.1.1.4 Habitat and use

The area is host to the commercial aquaculture of Atlantic salmon primarily, and habitat to many species of wildlife (Buzeta et al., 2003). The habitat and migration paths of wild Atlantic salmon (*Salmo salar* [Linnaeus, 1758]; WoRMS, 2020) populations of the inner (Government of Canada, 2019) and outer (Department of Fisheries and Oceans, 2014) Bay of Fundy may overlap with aquaculture sites (i.e., farms; Chang et al., 2011). However, their population size is small (Department of Fisheries and Oceans, 2014; Government of Canada, 2019) and the probability of transmission from wild to cultured populations is likely very low.

The probability of cultured populations acting as the source for new infestations of wild Atlantic salmon populations is unknown. Transmission of sea lice from cultured to wild Atlantic salmon populations through seeding of the environment with more infective copepodid stages is possible, especially in situations of high sea lice abundance at aquaculture sites (Chang et al., 2011; Dempster et al., 2021). Wild postsmolt salmon captured near the east coast of Grand Manan did not have any *L. salmonis* during the sampling period of 2001 to 2003 (Lacroix and Knox, 2005). In 2002, all captured wild fish in the Magaguadavic River, which drains into the Passamaquoddy Bay, had low intensities of sea lice (defined as 20 or fewer lice per fish [Carr and Whoriskey, 2004]). No recent literature was obtained regarding sea lice infestation of wild Atlantic salmon populations in the study area.

2.1.1.5 Hydrodynamics

The Bay of Fundy has an extreme tidal range with strong tidal currents (Garrett, 1972; Greenberg 1983). The major current entering the Bay of Fundy is the Western Nova Scotian Inflow that originates from the Scotian Shelf Coastal Current in the Eastern Gulf (Aretxabaleta et al., 2009). The current exits the Bay along the southwestern coast of NB and east of Grand Manan and creates the Bay of Fundy Gyre to the northeast of Grand Manan (Aretxabaleta et al., 2009).

The dominant tidal component of the Bay of Fundy is the principal semidiurnal lunar constituent (Garrett, 1972; Greenberg 1983), which means the gravitational forces of the moon exceed those of the sun and demonstrate a M_2 frequency. The tidal current exchange for the Bay of Fundy is 12.42 hours (Garrett, 1972; Greenberg 1983). Averaged tidal current magnitudes range from 0.8-2.0 m/s (Godin, 1968; Greenberg, 1983; Brooks, 1994; Hannah et al., 2001).

Hydrodynamic modelling of the Bay of Fundy, NB, occurred for the development of the BMA (Chang et al., 2007). Circulation model-derived tidal excursion areas for aquaculture sites were estimated using a three-dimensional particle-tracking model developed by Greenberg et al. (2005) and modified for the Bay of Fundy, NB (Page et al., 2005; Chang et al., 2005a,b, 2006a,b, 2007). The models had an estimated maximum displacement of particles with a mean of 4.1 km (SD 3.4 km) for 94 sites (Chang et al., 2007). This hydrodynamic modelling data was previously used to demonstrate the spatio-temporal distribution of Infectious

Salmon Anemia virus (ISAv) in the Passamaquoddy Bay (Gustafson et al., 2007; Gautam et al., 2018).

Hydrodynamic and particle-tracking modelling for the purpose of the evaluation of sea lice dispersal has not been performed for the Bay of Fundy, NB. Amundrud and Murray (2009) and others (e.g., Adams et al., 2012) have demonstrated environmental factors such as wind and freshwater supply affect the dispersal of sea lice. Tidal mixing was a notable driver for the dispersal of particles representing sea lice during simulations of less than 14 days for Loch Torridon, Scotland (Amundrud and Murray, 2009).

2.1.2 Aquaculture and sea lice in the Bay of Fundy, NB

Production values of farmed Atlantic salmon in our study area increased by about 18 % over the past decade (Chang and Page, 2014; L. Hammell, pers. comm.). In 2009, 24,000 tons of salmon were produced for a value of 159 million CAD, which accounted for 23.9 % and 26.5 % of total Canadian salmon production volume and value, respectively (100,212 tons and 599 million CAD; Statistics Canada, 2020). In 2018, NB produced 28 metric tonnes of salmon for a value of 271 million CAD (23.0 % and 24.3 % of total Canadian salmon production volume and value [123,184 tons and 1,056 million CAD], respectively; these values were not adjusted according to inflation [Statistics Canada, 2020]).

Sea lice control is a challenge for the farmed salmon industry in the Bay of Fundy, NB. In 2010, high abundances of *Lepeophtheirus salmonis* (Krøyer, 1837; WoRMS, 2020) resulted in high mortality rates of salmon that were stocked that

year if they were untreated (Saksida et al., 2015) due to lack of available treatments and recent development of widespread emamectin benzoate (Slice®, Merck Animal Health, USA) resistance (Chang et al., 2011; Igboeli et al., 2012). The area has not had high post-smolt mortality due to uncontrolled sea lice since 2010 (L. Hammell, pers. comm.). Although physiologic or economic effects were not part of this research, studies in other areas have concluded that high numbers of sea lice can affect the salmon by grazing on mucus, skin and underlying tissue causing bleeding, increased mucus production (Tully and Nolan 2002; Johnson et al., 2004) and immunocompromise of its host (Fast, 2014). Additional information on the pathophysiological effects of sea lice on salmon can be found in reviews by Boxaspen (2006) and Torrissen et al. (2013).

The cost to farmers related to sea lice problems were estimated at 6.16-6.18 CAD per kg of Atlantic salmon produced in 2000 (Mustafa et al., 2021) and approximately 6.8 million CAD in 2006 (Costello, 2009). A more recent economic analysis was performed in Norway and estimated the cost of average infestation and scenario for no treatments was between 2.3 and 13.1 % of revenue (Abolofia et al., 2017). The primary cost to sea lice appears to be in the control methods (both medicinal and non-medicinal applications) employed to maintain sufficiently low sea lice abundance to reduce potential impacts on wild salmon and other farmed salmon populations.

2.1.2.1 History of aquaculture and organization in Bay Management Areas

The NB Atlantic salmon aquaculture industry began in 1978 as a research and

development initiative by the Salmon Genetics Research Program of the North American Salmon Research Center (Bailey et al.,1983). Chang et al. (2011; Chang and Page, 2014) provides additional details on the history of Atlantic salmon aquaculture in the area. Aquaculture sites were organized into 10 Sea Louse Management Zones in 1995 (Chang et al., 2011). The BMA framework was created in 2000 with 22 areas to manage ongoing ISAV outbreaks that began in 1996 (Chang et al., 2011, 2014), standardize industry practices, improve production efficiency, and promote environmental sustainability (New Brunswick Department of Agriculture Fisheries and Oceans Canada, 2013). There were 21 BMA (revised to 22 in 2002) and these zonal health management units utilized a 2-year rotational stocking system. The framework was revised in 2006 for improved risk management of the ISAv, implementing a 3-year rotation stocking system with a mandatory fallowing period (time during which no fish are cultured in an area in order to reduce the sea lice burden), and integrating other resource users (Chang et al., 2007). The BMA 4 was active during 2006 to 2010 at which time those sites were merged with the BMA 1 (Jones et al., 2012). The BMA 3C replaced the BMA 4 with the same boundary locations in 2017 (Department of Energy and Resource Development/Agriculture, 2017). In 2017, there were 8 BMA (Department of Energy and Resource Development/Agriculture, 2017; Figure 2.1; Table 2.1). The stocking of smolt in cages is regulated according to the 3-year rotation system (Table 2.1). The site production cycle is defined by the stocking to harvest dates at site and area levels.

2.1.2.2 History of sea lice and monitoring practices

There are records of *L. salmonis* on wild Atlantic salmon prior to the development of aquaculture in the Bay of Fundy, NB (Chang et al., 2011; Torrissen et al., 2013). The first outbreak of *L. salmonis* was confined to a single site at Dark Harbour, Grand Manan Island, in 1984 where up to 100 sea lice per fish were reported (L'Aventure, 1987; Stuart, 1990; Chang et al., 2011). The second outbreak was in 1994 and started in the Letang area; *L. salmonis* abundance increased to problematic levels from 2 to 84 lice per infested fish over the period of August-September to October (Hogans, 1995). Other species of lice were also present: *Caligus elongatus* (Nordmann, 1832) and *Caligus curtus* (Müller O.F., 1785; Hogans and Trudeau, 1989; WoRMS, 2020). *C. elongatus* abundance was higher than *L. salmonis* prior to 1990 (Hogans and Trudeau, 1989) and it was treated in the mid-1990s (L. Hammell, pers. comm.). The abundance of *C. elongatus* did not increase in New Brunswick to a point of concern (Chang et al., 2011) and the neighbouring province of NS has required occasional control of *C. elongatus* (L. Hammell, pers. comm.). *C. curtus* abundance remained low (Chang et al., 2011).

A monitoring program for the abundance of sea lice on cultured Atlantic salmon was conducted between October 1994 and August 1996 by the NB government (Chang et al., 2011). The plan was to sample a minimum of one site per Sea Louse Management Zone per month (Chang et al., 2011). Ten years later, a survey of 8 of 11 active sites in the Passamaquoddy Bay was performed from May to December (MacPhee and Moore, 2007). Sea lice counts remained in

private and company veterinary services records (Jones et al., 2012, 2013) until Fish-iTrends was initiated in 2009.

An integrated sea lice management program was initiated by the NB Department of Agriculture, Aquaculture and Fisheries in 2009 in response to anecdotal reports of resistance to the main sea lice treatment (emamectin benzoate; Chang et al., 2011). The program had standardized sampling protocols and reporting of counts with monitoring of treatment effectiveness (Elmoslemany et al., 2013). Part of the program was the Fish-iTrends web-based data management system that is operated by the Centre for Aquatic Health Sciences (CAHS) at the Atlantic Veterinary College (AVC) of the University of Prince Edward Island, Prince Edward Island, Canada. It was a system that was developed to manage sea lice data, and their treatments, in Atlantic salmon marine sites for the salmon farmers of NB initially and eventually, in 2016, included Nova Scotia and Newfoundland and Labrador, Canada. Data access is limited to producers, aquatic veterinarians and the AVC-CAHS for real-time comparisons and visualization of sea lice abundance trends and production of summaries used by producer associations (Gautam et al., 2016).

Sampling salmon for sea lice counts involves opportunistically sampling of Atlantic salmon attracted using feed, captured, and anesthetized for sea lice counting (Jones et al., 2012). The NB regulation requires counts of sea lice on samples of 5 or more salmon in 6 or more cages at each site weekly if the sea temperature is above 5 °C and prior to and after bath treatments (General Regulation 91-158 [Government of New Brunswick, 2020]). Sea lice counts when

sea temperatures are below 5°C are mandated monthly (Government of New Brunswick, 2020) because it is more difficult to access sites and fish due to weather conditions (i.e., unsafe for humans and fish), and because salmon don't readily rise to the surface for sampling.

2.1.2.3 Topical and oral treatments for sea lice management

A number of sea lice treatments have been employed since 1984 (Chang et al., 2011); treatments include topical and systemic (oral) administration of parasiticides, and other topical removal methods. Keen et al. (2020) and Burridge and Van Geest (2014) reviewed sea lice treatments in Canada.

In summary, multiple topical parasiticides, or pesticides, have been used in the Bay of Fundy, NB. Early topical treatments included dichlorvos (Nuvan® CIBA Laboratories Limited, United Kingdom [UK]; L'Aventure, 1987), azamethiphos (Salmosan® Vet, Benchmark Animal Health, UK), hydrogen peroxide (Salartect® 350 and 500, Brenntag, UK), and cypermethrin (Excis®, Novartis Animal Health Limited, Switzerland; Burridge and Van Geest, 2014). Topical treatments were halted between 2000 and 2009 for the preferential use of the in-feed parasiticide emamectin benzoate (Burridge and Van Geest, 2014). Emamectin benzoate became available to farmers under emergency registration and was preferentially used because of the poor efficacy of other treatments and its activity against all life stages of *L. salmonis* (Burridge and Van Geest, 2014). Azamethiphos, hydrogen peroxide (Interox® Paramove® 50™, Solvay Chemicals Inc., Belgium, and Aquaparox 50™, Alpha Chemical Limited, Canada), and deltamethrin (Alpha

Max®, Zoetis Inc., USA) were used starting in 2010 (Burridge and Van Geest, 2014). These topical parasiticides are applied to the salmon using bath application methods where the cage is fully enclosed (using a tarpaulin), or the salmon are temporarily transferred into a wellboat. Partial enclosures (referred to as cage skirts) were utilized briefly in 2009-2010 but were quickly abandoned due to their suboptimal delivery of treatments.

Other topical delousing methods include the Hydrolicer® (Hydrolicer Production AS, Norway; Overton et al., 2019), warm water/thermal (Grøntvedt et al., 2015; Overton et al., 2019), and cleaner fish treatments (Gonzalez and de Boer, 2017). The Hydrolicer® is a system that physically removes the sea lice by using inverse water turbulence and was efficient at removing 82-100 % of mobile lice (Overton et al., 2019). The thermal delousing method involves bathing the salmon in warm water up to 34 °C for approximately 20 to 30 seconds (Overton et al., 2019). These are relatively new topical anti-louse control methods: 2015 in Norway (Overton et al., 2019) and 2018 in Atlantic Canada.

Systemic parasiticides were employed since 1994-1995 (Chang et al., 2011) for sea lice control and were administered to the salmon through feed. Avermectins (Ivomec®, Boehringer Ingelheim, Germany, and Noromectin®, Norbrook Laboratories Ltd., United Kingdom), emamectin benzoate, and teflubenzuron (Calicide®, Nutreco ARC Ltd., Netherlands) were administered (Burridge and Van Geest, 2014). Beginning in 2017, lufenuron (Elanco Animal Health, USA) has been administered in-feed at some hatcheries prior to transferring smolt to the sea cages but has not been used since 2020 (L. Hammell, pers. comm.).

Farm companies submit sea lice relevant data to the Fish-iTrends data management system, including the method and timing of the application of the topical and systemic (oral) administration of parasiticides, and topical mechanical removal methods applied to manage the sea lice problem at the cage-level.

2.1.3 Objective

The Bay of Fundy, NB, is a large area with a complex environment. There are 36 active aquaculture sites (Department of Energy and Resource Development/Agriculture, and Fisheries, 2017) registered by the NB provincial government and all are required to participate in the industry-wide database for the counts of sea lice. The Fish-iTrends database presents a unique opportunity for the descriptive analysis of sea lice trends in the Atlantic salmon aquaculture industry for the Bay of Fundy, NB.

The intention of this chapter is to describe the trends of sea lice counts, treatments, and sea surface temperature between 2009 and 2018. Data will be described to observe annual, seasonal, and spatial trends. These descriptive statistics will serve to provide a summary of the Fish-iTrends data and identify possible confounding factors for the process of modelling the data in subsequent chapters.

2.2 Materials and methods

Sea lice count and treatment data from aquaculture sites in the Bay of Fundy, NB, between July 2009 and early November 2018, were extracted from the Fish-iTrends data management system and included in this descriptive study. Count data consisted of routine counts of *Caligus* sp., and the *L. salmonis* life stages of

chalimus, pre-adult and adult male (PAAM), and adult female (AF; with and without egg strings) on farmed Atlantic salmon from several cages at each site during their production cycle (see Gautam et al., 2016 for detailed sampling description). Sea lice counts were adjusted for the number of salmon counted (e.g., number of AF counted/number of fish counted). Abundances of sea lice were transformed using the natural logarithm of sea lice per fish with an offset of 1. BMA 5 and 6 were excluded from the descriptive analysis as they are a conservation area for wild salmon and limited to species other than Atlantic salmon, respectively.

Topical treatments were summarized as proportions of annual applications. Hydrogen peroxide treatments (Paramove® 50™ and Aquaparox 50™) were combined into a single category. In-feed treatments provided to fish within cages were summarized as the number of days of applications at a cage of each parasiticide. There were no data on lufenuron applications at the hatcheries that supplied Atlantic salmon to the sites in our study area.

The frequency of observations (i.e., counts) and abundances of sea lice were summarized according to season and production cycle. Seasons were defined per the equinox and solstices: winter December 21-March 19, spring March 20-June 20, summer June 21-September 21, and autumn September 22-December 20. Production cycles were defined for a site according to the stocking and harvesting dates which were submitted by the farmers and managers into the Fish-iTrends data management system. Cages and sites were uniquely identified

according to their specific production cycle. Summaries were not adjusted for fish group identifiers as the movement of fish among cages at a site are complex.

The United Kingdom Meteorological Office's operational sea surface temperature and sea ice analysis (UKMO OSTIA) produces daily global sea surface temperature estimates (UK Met Office, 2005) which were utilized for the study area. This remote sensing product was selected for multiple reasons. The temperature recorded at the site does not have the corresponding measurement depth and there were many missing values. The UKMO temperature had the highest concordance correlation coefficient and index of agreement with *in situ* water temperature measurements taken on aquaculture sites in the Broughton Archipelago, in British Columbia, Canada (Thakur et al., 2018). Daily temperature values were extracted for the centroid location of each aquaculture site polygon utilizing the 'raster' package, version 3.0-12 (Hijmans et al., 2020) in R. The spatial resolution was 1/20 degrees, which is approximately 6 km at the 45 degree latitude of the Bay of Fundy, NB; sites that occupied the same temperature grid shared temperature values which may result in mild spatial clustering as a few farms are within 6 km of a neighbouring farm in the study area. Water temperature trends were summarized specifically for the year 2017 because this year was proposed for the development of a hydrodynamic model of the Bay of Fundy, NB.

The trends of sea lice counts and abundances, treatments, and sea surface temperature were presented in tables and figures. All analyses, unless otherwise specified, were conducted in Stata® v16.1 (2019, StataCorp).

2.3 Results

2.3.1 Observations

There was a total of 63,214 observations (i.e., count events) between 06 July 2009 and 04 November 2018. The most frequent observations were performed in 2014; a total of 8,771 observations were recorded in 45 sites for an average of 195 observations per site. Observations were organized into 1,091 cages and these were distributed among 70 sites. There was a total of 191 site-cycles (short for site-production cycles) and 2,467 cage- cycles (short for cage-production cycles). The number of active sites varied annually; there were approximately 40 active sites per year, and a maximum of 49 sites were active in 2015.

Observations were most frequently performed for sites in the BMA 1 (n = 21,268) followed by the BMA 2B (n = 12,530) and 2A (n = 11,877). The BMA 1, 2A and 3B had active sites during the entire study period suggesting fallowing may not synchronized for the management area. BMA 2B and 3A had active sites starting in 2010. BMA 3C had active sites starting in 2017.

There was a total of 191 production cycles and 38,771 production days during the study period; on average production cycles were less than a year, with 203 production days. The BMA 1 had 69 production cycles and 11,300 production days (Figure 2.2B). BMA 1 had an additional 2,644 production days compared to the BMA 2A, which had the 2nd largest number of production days with 8,556. The BMA 2B had 34 production cycles and contributed 8,287 production days. The BMA 3A contributed 20 production cycles and 3,060 production days. The BMA 3B had 30 production cycles and 6,377 production days. The BMA 3C had

2 production cycles and 1,091 production days. There were 33,828 counts performed at cages in the first 365 days at sea since stocking of the first cage at a site, 27,665 in the 2nd year at sea (365 to 729 days at sea) and 1,721 in the 3rd year (great than 730 days at sea).

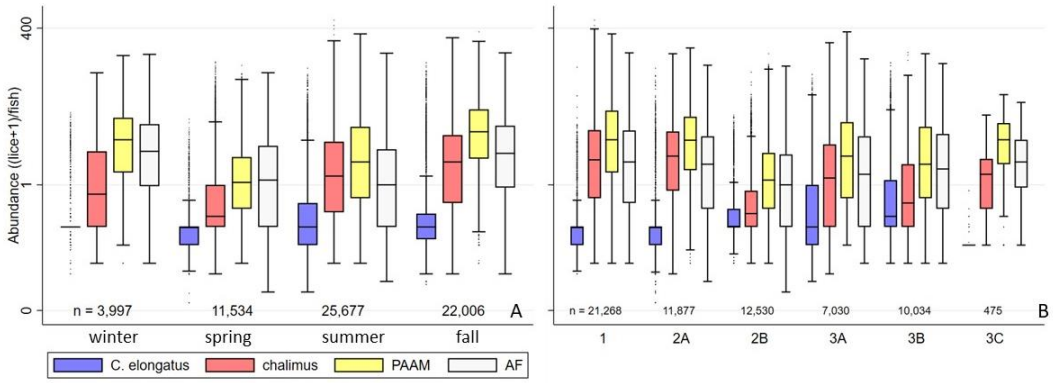

Figure 2.2. Boxplot of sea lice abundances ([lice+1]/fish) in the Bay of Fundy, New Brunswick, for July 2009 to November 2018 per season (A) and Bay Management Areas (B). Seasons were defined using the equinox and solstices. n = 63,214

A minimum of 5 fish were counted for over 96.5 % of the observations (n = 60,242). The counts are performed less frequently in winter (n = 3,997) and at water temperatures under 5 °C (n = 3,155). Sea lice counts were performed most frequently in the summer (n = 25,677) and then in autumn (n = 22,006; Figure 2.3). In 2010, 2012-2014, and 2017, the number of observations performed in the autumn were near or exceeded the number of observations performed during the summer.

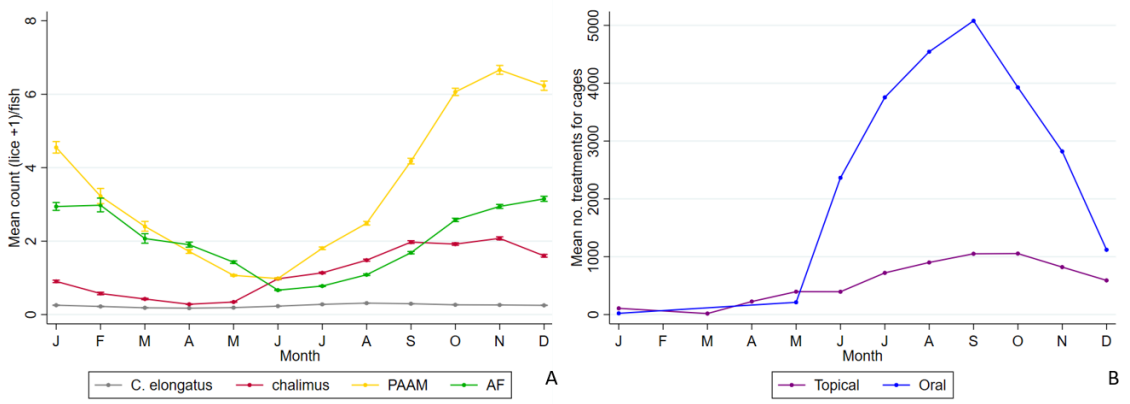

Figure 2.3. Mean sea lice abundances ([lice+1]/fish) per month +/- standard error for *Caligus elongatus* and *Lepeophtheirus salmonis* for the Bay of Fundy, New Brunswick, for July 2009 to November 2018 (A). *L. salmonis* life stages were counted as chalimus, pre-adult and adult male (PAAM), and adult female (AF). Mean number of topical and oral treatments per cage for the same time period (B). Oral treatments are reported in cage-days of treatment.

2.3.2 Sea lice abundances

All sea lice counts were strongly right skewed with a substantial percentage of zero counts (9.9 to 66.6 %; Table 2.2). There were few *Caligus* sp. per fish for the majority of the study period; the species of *Caligus* was not identified, but it was assumed that the majority were *C. elongatus* based on previous published data. The chalimus, PAAM and AF *L. salmonis* life stages had medians of more than 1 louse per fish. The maximum value for the range of the *L. salmonis* exceeded 120 lice per fish (all stages).

Table 2.2. Summary of counts of sea lice between July 2009 and November 2018 for the Bay of Fundy, NB. PAAM: pre-adult and adult male, AF: adult female, 25 and 75%; 25 and 75th percentiles. n = 63,432

Variable	Missing (n)	Zero (n)	≤ 1 louse/fish (n)	≤ 5 lice/fish (n)	Median (lice/fish)	25 % (lice/fish)	75 % (lice/fish)	Range (lice/fish)
Caligus sp.	874	42,081	56,465	60,388	0	0	0.2	0, 106.0
L. salmonis								
Chalimus	48	16,174	32,269	48,609	1.0	0	4.6	0, 540.0
PAAM	51	6,260	19,857	37,458	3.2	0.7	11.0	0, 346.1
AF	204	12,842	27,517	45,347	1.6	0.2	6.0	0, 154.8

C. elongatus abundance was highest in the summer and lowest in the winter (mean 0.29, SD 3.39 and mean 0.24, SD 2.40, respectively; Figure 2.2A). Chalimus abundance increased from winter into autumn; in autumn, its mean was 1.94 (SD 5.27). The abundance of PAAM was highest in autumn (mean 6.17, SD 4.55) followed by the winter (mean 4.36, SD 4.62). Although fewer observations occurred in winter, the abundance of AF was highest (2.85, SD 5.03) followed by the autumn (mean 2.70, SD 5.00).

The abundances of *Caligus* sp. were lower than the abundances of each life stage of *L. salmonis* (Figure 2.2). PAAM *L. salmonis* had the highest abundance compared to the chalimus and AF life stages. The mean daily abundance of *C. elongatus* increased in 2013 compared to the other years. The mean monthly abundance of AF was higher for the years 2016 to 2018 compared to the previous years. There was a cyclical trend to the mean daily counts of sea lice annually. Generally, the mean abundance of sea lice was lowest in May and was highest in October for the years 2009 to 2018 (Figure 2.3). *C. elongatus* mean monthly abundances remained near 0 during the whole year. The *L. salmonis* chalimus life stage mean monthly abundances decreased between November and April and increased between April and September; the mean monthly abundances were fairly constant between September and November. The mean monthly abundances of PAAM increased between June to November and decreased from November to June. The AF mean monthly abundances decreased from February to June, increased from June to October and were generally constant between October to February.

The mean abundance of sea lice differed by BMA and had large variability (Figure 2.2B). In Grand Manan Island, the BMA 2B had smaller mean abundances compared to the BMA 3B (AF mean 0.98, SD 4.57 vs. mean 1.62, SD 5.71).

2.3.3 Sea lice treatments

A total of 6,296 topical sea lice treatments were applied to cages during the study period. Topical treatments varied by month (Figure 2.3B) and year (Figure 2.4). Deltamethrin was the most frequent topical treatment in 2009 and was discontinued after 2010. The skirt method of application was employed for the application of deltamethrin and azamethiphos, with few uses in 2012 after which it was discontinued. Hydrogen peroxide applied using a wellboat and azamethiphos applied using a tarpaulin were by far the most frequently performed topical treatments. The Hydrolicer® was introduced in 2018. Warm water topical exposure applications began in 2015 and in 2018, it was the most frequently employed topical management tool. The proportion of hydrogen peroxide treatments applied by wellboat was greatest from 2011 to 2015. Out of 6,296 total treatments, the proportion with azamethiphos (applied using a tarpaulin) increased from 2010 to 2017.

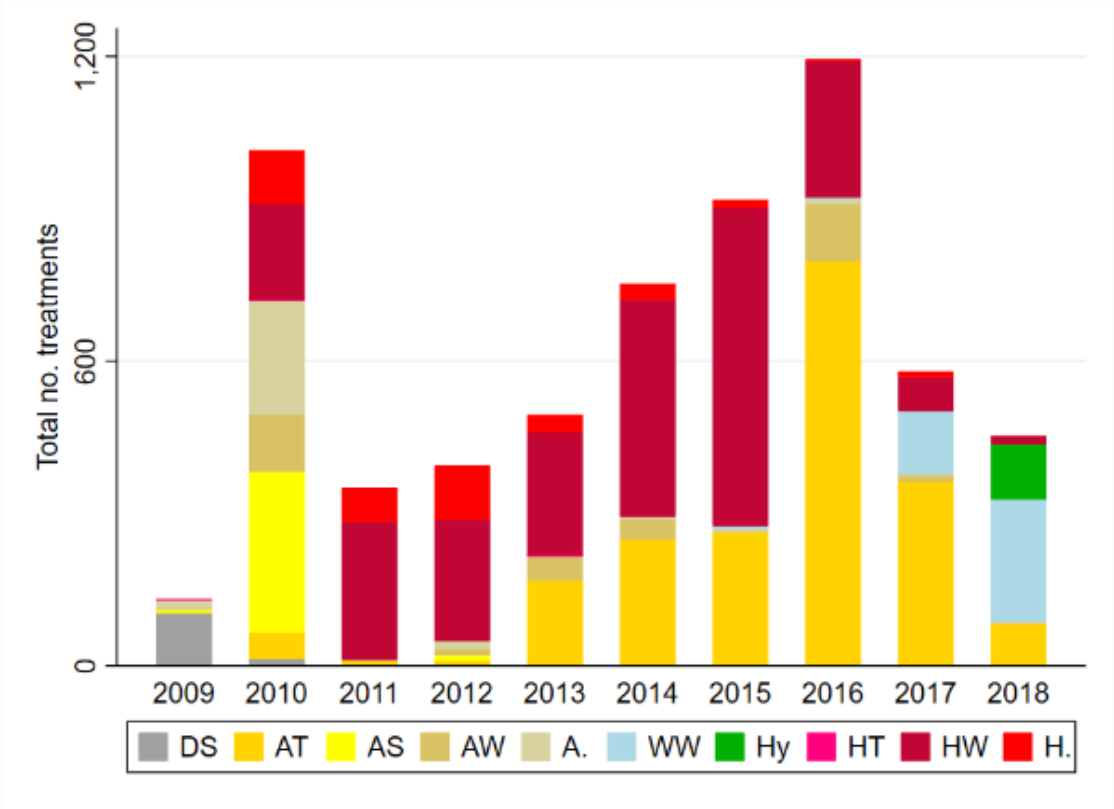

Figure 2.4. Total number of topical treatments by method of application applied annually in the Bay of Fundy, New Brunswick. DS: deltamethrin skirt, AT: azamethiphos tarpaulin, AS: azamethiphos skirt, AW: azamethiphos wellboat, A.: azamethiphos missing method of application, WW: thermal, Hy: Hydrolicer®, HT: hydrogen peroxide tarpaulin, HW: hydrogen peroxide wellboat, and H.: hydrogen peroxide missing method of application. n_{total} = 6,296

Topical treatment uses differed by location. The BMA 1 and 2A had the most frequent treatment applications over the study period. In BMA 1, a third of topical treatments were azamethiphos applied by tarpaulin and another third were hydrogen peroxide applied by wellboat. The BMA 2A had the same breakdown of topical treatments. A large number of treatments were applied in 2010 and 2016 compared to the other years; the BMA 1 had the most frequent use of topical treatments applications during those years. The BMA 2B had 348 topical treatments compared to the BMA 3B with 657 topical treatments.

There was a total of 23,978 cage-days of oral sea lice treatments applied to cages. Each treatment day for each cage is represented by one cage-day. For example, 10 cages treated with emamectin benzoate for 7 days is represented by 70 cage-days of emamectin benzoate treatment. The most frequent application of in-feed treatments was applied in 2013. Avermectins were applied starting in 2010. Emamectin benzoate was used in at least one site in the NB industry every year of the study period. Avermectins had the largest proportion of application days, followed by emamectin benzoate. Teflubenzuron was applied the least frequently and was discontinued after 2011. In-feed applications were most commonly applied during the months of May to December and the largest number of daily applications was in September (n = 5,079; Figure 2.3).

2.3.4 Water temperature

Sea surface temperature demonstrated a cyclical pattern that repeated annually (Figure 2.5A). The distribution of the water temperature measurements had an approximate Gaussian distribution. The mean water temperature across the study period was 10.9 °C (n = 11,157, SD 3.0). The mean of the annual minimum water temperature between 2009 and 2018 was 3.1 °C (n = 10, SD 1.9 °C) and the mean of the annual maximum was 15.4 °C (n = 10, SD 0.7 °C). However, during the time period, a minimum of 0.8 °C and maximum of 16.3 °C were observed. The year 2015 had the largest range of water temperature (n = 1,357, mean 10.6 °C, SD 3.1 °C, maximum 0.8°C, minimum 16.0 °C).

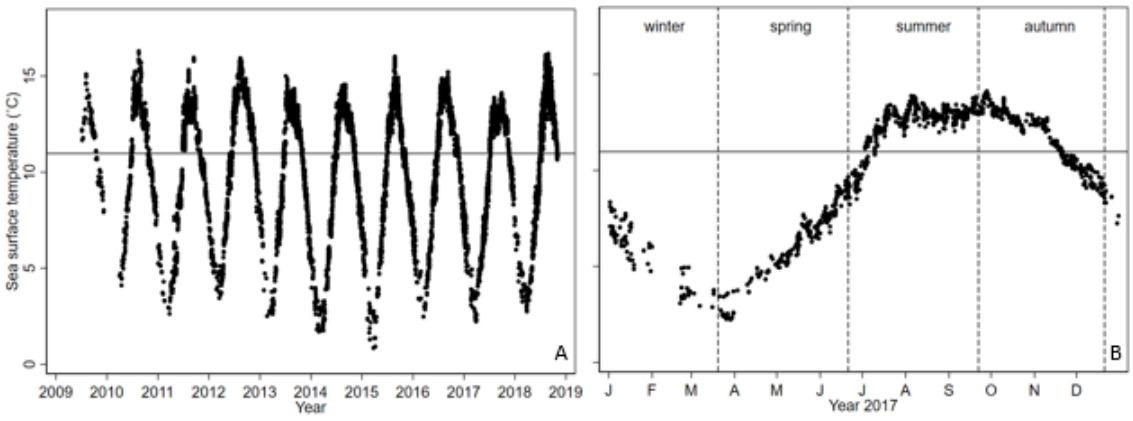

Figure 2.5. Average daily sea surface temperature (extracted from the UKMO OSTIA SST remote sensing product which has a 6 km resolution [UK Met Office, 2005]) per aquaculture site for the duration of the study (A) and 2017 excerpt (B). The horizontal line represents the mean temperature for the plot. Vertical lines (B) are the seasonal equinox and solstices. n = 11,157, n_{2017} = 1,332

In 2017, the mean water temperature during the winter was 5.9 °C (n = 103, SD 1.7 °C) and had a range of 2.9 °C to 9.2 °C (Figure 2.5B). In the spring, the temperature had a range of 2.2 °C to 9.4 °C with a mean of 6.4 °C (n = 284, SD 1.8 °C). During the summer, the mean water temperature was 12.3 °C (n = 516, SD 1.2 °C) and had a range of 8.2 °C to 13.6 °C. The autumn had a mean water temperature of 11.8 °C (n = 429, SD 1.4 °C) with a range of 8.7 °C to 14.1 °C.

2.4 Discussion

2.4.1 Sea lice counts and abundances

This study is the most extensive historical descriptive analysis of sea lice counts in the Bay of Fundy, NB, as it covers a period of 10 years. This descriptive study adds to the previous sea lice trend analyses performed in the area (Chang et al., 2011; MacPhee and Moore, 2007). Subsets of these data have been previously employed for the evaluation of treatments, timing of count events, and other studies (Jones et al., 2012, 2013; Elmoslemany et al., 2013; Gautam et al., 2016,

2017a,b; Elghafghuf et al., 2020). This research demonstrates that the Fish-iTrends data management system is a valuable tool for the evaluation of sea lice trends in the Bay of Fundy, NB, and highlights the possibility of additional historical analyses.

The NB regulations have a set of requirements for counts (General Regulation 91-158 [Government of New Brunswick, 2020]) and these were met for the majority of the observations. Previous studies that utilized the sea lice count data from the Bay of Fundy, NB, limited evaluations to observations with five or more fish; those authors did not report the proportion with fewer than 5 fish per observation (Jones et al., 2012, 2013; Gautam et al., 2016, 2017a,b). A recent study did not exclude observations with fewer than 5 fish and instead, calculated a mean abundance of sea lice at the site level (Elghafghuf et al., 2020).

The upper limit of the range of *L. salmonis* per fish (Table 2.2) exceeded 120 lice (all stages) per fish, which is an estimate for the maximum number of lice that can reasonably be counted within the limited time (L. Hammell, pers. comm.). Counts above 120 lice per fish are likely a crude estimate based on a count of sea lice in a small area of the fish and then multiplied for the entire fish. However, these large values of lice per fish accounted for a small percentage (< 1 %) of the counts.

L. salmonis remains a parasite of ongoing concern for the Atlantic salmon aquaculture industry. The estimated average sea lice per fish were lower in previous studies in the area (Chang et al., 2011). In 2006, the average number of

L. salmonis sea lice/fish was above 3 in September (MacPhee and Moore, 2007). *L. salmonis* PAAM abundances were higher than chalimus and AF in this study as observed in Grand Manan between 2010 and 2016 (Elghafghuf et al., 2020). In Passamaquoddy Bay between May and June 2007, the mean abundance of *L. salmonis* chalimus per fish was smaller than the mean abundance of the other life stages per fish (MacPhee and Moore, 2007). Chalimus are more often under-counted compared to PAAM presumably because they were smaller and more difficult to identify by inexperienced counters, as determined by Elmoslemany et al. (2013). Also, chalimus have a limited life span as they develop into PAAM and likely have the most attrition (Stien et al., 2005). The adult male portion of PAAM will be present for a longer period compared to the pre-adult portion as it develops into male and female adults. Therefore, under situations of low but constant recruitment, it is expected that there are lower counts of chalimus (L. Hammell, pers. comm.). The AF counts were smaller than PAAM because the PAAM were a combination of the pre-adult (male and female) and adult male as per standard industry practice because these life stages are difficult to differentiate (Elmoslemany et al., 2013).

C. elongatus was present during the entire study period but abundance was low as previously noted for the Bay of Fundy, NB (Peterson et al., 2001; Chang et al., 2011). Comparatively, Chile has a greater burden from *Caligus* species. The *Caligus* species and *L. salmonis* lice in the Bay of Fundy, NB, share the same habitat as proposed for *L. salmonis* and *Caligus clemensi* (Parker and Margolis, 1964; WoRMS 2021) in BC (Patanasatienkul et al., 2015).

The true abundance of *C. elongatus* may be higher than estimated in this study; all life stages are combined for reporting into the Fish-iTrends database. As a general rule, sea lice in the anesthetic tote were not counted or reported; mobile *C. elongatus* may be underrepresented if they were more likely to detach in the anesthetic bath and were not counted, as suggested by a small unpublished study with *C. clemensi* (Saksida et al., 2007). Moreover, the chalimus life stage of *L. salmonis* in this study may include chalimus of the *C. elongatus*, as it was determined it is difficult to accurately differentiate the chalimus of *L. salmonis* and *C. clemensi* in the field and would require expert analysis under laboratory conditions (Saksida et al., 2007). Therefore, *Caligus* species should continue to be recorded. The Norwegian Atlantic salmon aquaculture industry observed an increase in the abundance of presumed *C. elongatus* starting around 2017 (Imsland et al., 2019a,b cited in Hemmingsen et al., 2020). In our study area, the abundances of *C. elongatus* were highest for the southern coast of Grand Manan and the southwestern coast of NB.

The defined production cycles per site were useful for the hierarchical organization of the data and could be used during future evaluation of these data. This list of production cycles can be shared with stakeholders to guide the industry to improve their management practices. Although the Fish-iTrends database may contain some errors regarding precise stocking and harvest dates, the evaluation of the site stocking data suggested that stocking salmon was occurring outside of the published stocking years (Table 2.1). This suggests that the fallowing sites may be occurring primarily at the site-level rather than at the

BMA-level, and synchronization of fallowing should be synchronized within the zonal health management units. This may have implications for higher disease transmission rates as demonstrated by Werkman et al. (2011). Partial or unstructured fallowing of sites as modelled through network analysis had higher transmission rates of pathogens such as ISAV (Werkman et al., 2011). A more thorough evaluation of the fallowing schedule in the Bay of Fundy, NB, is outside the scope of this descriptive study.

2.4.2 Sea lice abundances vary by location

The abundances of *L. salmonis* were highest for the BMA 1 and we propose this finding was due to the relatively short distance between sites within this BMA. Generally, the BMAs 1, 2A and 3A had similar trends to each other in sea lice abundances. The BMA 3A had high abundances of sea lice despite its characteristics of a coastal area with presumably greater currents compared to the BMAs 1 and 2A that are in Passamaquoddy Bay.

BMA 3C abundances represented only a recent addition (2017) of Atlantic salmon production and therefore had limited contribution to the duration of the study period. Another consideration was the water current for the area, where the BMA 3C was near the open waters of the Bay of Fundy. It would be interesting to further evaluate the abundances of sea lice of the BMA 1 and 3C to account for differences in water currents. The BMA 1 could be subdivided into Passamaquoddy Bay north of Deer Island, between Deer Island and Campobello Island, and south of Campobello Island; we anticipate abundances of *L. salmonis* to be different in these areas.

The abundances of sea lice were smaller on the east coast of Grand Manan (BMA 2B) compared to the south coast (BMA 3B) despite the BMA 2B having a greater number of active sites and production days compared to the BMA 3B. The lower abundance of sea lice on the east coast of Grand Manan suggests that sea lice were not being transmitted from the southwestern coast of NB (BMA 3A) even though the dominant Bay of Fundy current passes along the southwest coast of NB and then east of Grand Manan (Aretxabaleta et al., 2009). It is possible that there is diffusion of sea lice and reduced transmission (Saksida et al., 2015), the lice were not surviving the distance, or they were being swept up in the gyre that occurs to the northeast of Grand Manan (Aretxabaleta et al., 2009). The higher abundance of sea lice in the BMA 3B may be explained by a lower depth-averaged speed on the south coast of Grand Manan (Aretxabaleta et al., 2008). Lower current speeds would decrease transportation of sea lice away from the area (Amundrud and Murray, 2009). Finally, differences between the BMA 2B and 3B may be due to differences in the management of sea lice within these BMA; for example, BMA 2B may have better synchronization of fallowing compared to BMA 3B.

The finding that the east coast of Grand Manan has lower abundances of sea lice compared to the south coast may suggest that the east coast of Grand Manan has a minimal risk of transmission of sea lice to wild salmon. The east coast of Grand Manan was identified in the early 2000s as an area of potential concern as sea lice were observed on captured wild salmon (Carr and Whoriskey, 2004; Lacroix and Knox, 2005). The effect of sea lice exchange between wild Atlantic

salmon and aquaculture sites in the Bay of Fundy, NB, should be further evaluated as some studies have found sea lice from aquaculture sites negatively impacted wild salmon (e.g., Torrissen et al., 2013; Rees et al., 2015; Kristoffersen et al., 2018). There may also be transfer of sea lice from wild to farmed salmon; the evaluation of this question should centre around the confirmation of wild salmon movement in the area and estimation of external infestation pressure soon after stocking if both BMAs 2B and 3B were fallowed synchronously.

2.4.3 Sea lice abundances vary seasonally

The abundances of sea lice observed during the period of 2009 to 2018 were similar to that of 1994 to 1996 when abundance was highest in the summer and autumn, and lowest in winter and spring with variation among sites (Chang et al., 2011). In the Broughton Archipelago, BC, abundances of sea lice were highest during September to December (Saksida et al., 2007). In Europe, sea lice abundances were highest during the late summer and early autumn (Revie et al., 2002; Rikardsen, 2004; Hayward et al., 2011).

The variation for each group of sea lice (*C. elongatus* and each life stage of the *L. salmonis*) were large when summarized by season, but the monthly variation was smaller. Season remains a common way to outline trends in the abundances of sea lice (e.g., Rikardsen, 2004; Whyte et al., 2013); however, season is usually further defined by the month because the definition of seasons is different between studies (e.g., Whyte et al., 2013: winter-spring from January to June and summer-fall from July to December vs. Gautam et al., 2016: spring, early

summer, late summer, and autumn). Seasonality trends of counts of *L. salmonis* seemed to be reduced after 2013 in Scotland (Hall and Murray, 2018).

Another method to evaluating trends in seasonality is with the use of environmental variables that vary naturally, such as water temperature. Rittenhouse et al. (2016) and others set biological delay differential equation models to water temperature and salinity to allow for seasonal variation. In other studies, water temperature has been used to define the seasons (e.g., Gautam et al., 2016). There may be additional factors that affect the variation of sea lice according to seasons. These may include wind speeds and direction, storm events, amount of daylight, and variation in salinity, and others (Brooker et al., 2018). Seasonal effects may be also evaluated by including a modelled sinusoidal function representing unexplained seasonal variation and comparing its effects to models with parameters of location, water temperature and salinity as performed by Kristoffersen et al. (2013). Factors such as wind, storms and salinity will be indirectly incorporated during the evaluation of dispersal of sea lice during application of hydrodynamic models in a subsequent chapter.

Our data demonstrated a lag of approximately 2 months between an increase in water temperature and an increase in the abundances of *L. salmonis* life stages. It was expected that there is a lag of one to multiple weeks for the development of a new sea lice population in response to changes in the environment (Aldrin et al., 2013; Aldrin et al., 2019; Rees et al., 2015; Elghafghuf et al., 2020). Historically, Chang et al. (2011) did not evaluate a temperature lag influencing the abundance of sea lice. Further evaluation of the relationship between water

temperature and abundance of sea lice for the Bay of Fundy, NB, will be presented in further chapters. It was anticipated that the observed lag was caused by the application of treatments. The mean number of oral and topical treatments applied were highest for the months of July to October. This increase in the frequency of treatments preceded the rise in the abundances of PAAM and AF, likely as farm management anticipated sea lice increase and addressed them early.

Finally, it must be noted that the annual water temperature range extremes measured between 2009 and 2018 were higher than that measured by Peterson et al. (2001) in 1995-1996 with a difference of 0.3 °C at the minimum and 1.1 °C at the maximum. This would impact the rates of development, reproduction, and infestation of sea lice in the area by generally producing an increase in those rates (Brooker et al., 2018). Should trends in rising sea temperatures continue in the Bay of Fundy, NB, negative impact on the sea lice situation for the NB aquaculture industry may occur (Fast and Dalvin, 2020).

2.4.4 Treatments

Over the study period, there was a trend for topical treatments to move away from topical parasiticides to other delousing methods, particularly mechanical and thermal delousing (i.e., the Hydrolicer® and thermal). These technologies were developed as alternatives that do not release parasiticides to the environment, and are not prone to parasiticide resistance (e.g., Igboeli et al., 2012) or adverse effects on non-target species (e.g., Parsons et al., 2020).

Despite the increasing use of mechanical and thermal delousing, azamethiphos

treatments were the most frequent applications historically. Azamethiphos was used between 1995 and 2002 and was given emergency registration again in 2009 (Burridge and Van Geest, 2014). Recently, a mutant allele associated with the improved survival to organophosphates (Carpio et al., 2011) was demonstrated to be present across the North Atlantic Ocean with samples including the Bay of Fundy, NB (Kaur et al., 2017). The lower numbers of wild refugia for sea lice in the Bay of Fundy may decrease their potential protective effect in the selection rate for chemical resistance (McEwan et al., 2015).

The second most applied topical parasiticide over the study period was hydrogen peroxide. According to Burridge and Van Geest (2014), the application of hydrogen peroxide was used sparingly between 2000 and 2010. Our data confirmed that statement for the years 2009 and 2010 when wellboats were not in operation, and subsequent introduction of wellboat applications increased the frequency of hydrogen peroxide treatments until the year 2016 when new tarpaulin methods facilitated targeted use of azamethiphos.

The systemic parasiticide, emamectin benzoate, has been used since 1999 (Burridge and Van Geest, 2014) despite the development of resistance in 2009 (Jones et al., 2012, 2013; Whyte et al., 2013, 2016) after which time the dose was increased (L. Hammell, pers. comm.). Igboeli et al. (2012) noted that maximal effective concentration that caused 50 % mortality was 4-26 times greater in 2011 compared to 2002-2004. The evaluation of the efficacy of emamectin benzoate and all other treatments were outside of the scope of this chapter, but their effect on sea lice will be presented in subsequent chapters.

2.5 Conclusions

The Bay of Fundy, NB, is a large area with complex environmental influences, including large tidal excursions, wide seasonal water temperature ranges, and heterogeneous current patterns. This area has a successful Atlantic salmon aquaculture industry that is challenged by sea lice, *L. salmonis*. Salmon farmers count sea lice frequently to monitor trends and manage delousing options; the Fish-iTrends database was developed to manage reported data. The Fish-iTrends database therefore presents a unique opportunity for the descriptive analysis of the sea lice related observations and practices of the Atlantic salmon aquaculture industry in the Bay of Fundy, NB. The intention of this chapter was to describe the trends of sea lice counts, treatments, and sea surface temperature between 2009 and 2018. As previously noted for the area, the abundances of *C. elongatus* and *L. salmonis* varied annually, seasonally, and spatially. The abundances of sea lice responded to water temperature and treatments applied at the sites. Trends in the abundances of sea lice and treatments will continue to be influenced by treatment response patterns, reduced chemical involvement, and regional stocking densities.

2.6 References

Abolofia, J., Asche, F., and Wilen, J. E. (2017). The Cost of lice: Quantifying the impacts of parasitic sea lice on farmed salmon. Marine Resource Economics, 32(3), 329–349. https://doi.org/10.1086/691981

Adams, T., Black, K., Macintyre, C., Macintyre, I., and Dean, R. (2012). Connectivity modelling and network analysis of sea lice infection in Loch Fyne, west coast of Scotland. Aquaculture Environment Interactions, 3, 51–63. https://doi.org/10.3354/aei00052

Aldrin, M., Jansen, P. A., and Stryhn, H. (2019). A partly stage-structured model for the abundance of salmon lice in salmonid farms. Epidemics, 26, 9–22. https://doi.org/10.1016/j.epidem.2018.08.001

Aldrin, M., Storvik, B., Kristoffersen, A. B., and Jansen, P. A. (2013). Space-time modelling of the spread of salmon lice between and within Norwegian marine salmon farms. PLoS ONE, 8(5), 1–10. https://doi.org/10.1371/journal.pone.0064039

Amundrud, T. L., and Murray, A. G. (2009). Modelling sea lice dispersion under varying environmental forcing in a Scottish sea loch. Journal of Fish Diseases, 32(1), 27–44. https://doi.org/10.1111/j.1365-2761.2008.00980.x

Aretxabaleta, A. L., Mcgillicuddy, D. J., Smith, K. W., and Lynch, D. R. (2008). Model simulations of the Bay of Fundy Gyre: 1. Climatological results. Journal of Geophysical Research, 113, 10027. https://doi.org/10.1029/2007JC004480

Aretxabaleta, A. L., Mcgillicuddy, D. J., Smith, K. W., Manning, J. P., and Lynch, D. R. (2009). Model simulations of the Bay of Fundy Gyre: 2. Hindcasts for 2005-2007 reveal interannual variability in retentiveness. Journal of Geophysical Research, 114, 9005. https://doi.org/10.1029/2008JC004948

Arriagada, G., Vanderstichel, R., Stryhn, H., Milligan, B., and Revie, C. W. (2016). Evaluation of water salinity effects on the sea lice *Lepeophtheirus salmonis* found on farmed Atlantic salmon in Muchalat Inlet, British Columbia, Canada. Aquaculture, 464, 554–563. https://doi.org/10.1016/j.aquaculture.2016.08.002

Bailey, J. K., Loudanslager, E., Glebe, B. D., Saunders, R. L., and Schom, C. B. (1983). Salmon Genetics Research Program, North American Salmon Research Center. St. Andrews, NB.

Boxaspen, K. (2006). A review of the biology and genetics of sea lice. ICES Journal of Marine Science, 63(7), 1304–1316. https://doi.org/10.1016/j.icesjms.2006.04.017

Bricknell, I. R., Dalesman, S. J., O'Shea, B., Pert, C. C., and Luntz, A. J. M. (2006). Effect of environmental salinity on sea lice *Lepeophtheirus salmonis*

settlement success. Diseases of Aquatic Organisms, 71(3), 201–212. https://doi.org/10.3354/dao071201

Brooker, A. J., Skern-Mauritzen, R., and Bron, J. E. (2018). Production, mortality, and infectivity of planktonic larval sea lice, *Lepeophtheirus salmonis* (Krøyer, 1837): Current knowledge and implications for epidemiological modelling. ICES Journal of Marine Science, 75(4), 1214–1234. https://doi.org/10.1093/icesjms/fsy015

Brooks, D.A. (1994). A model study of the buoyancy-driven circulation the Gulf of Maine. Journal of Physical Oceanography, 24:2387-2412.

Burridge, L. E., and Van Geest, J.L. (2013). A review of potential environmental risks associated with the use of pesticides to treat Atlantic salmon against infestations of sea lice in southwest New Brunswick, Canada. DFO Canadian Science Advisory Secretariat Research Document (Vol. 050).

Buzeta, M., Singh, R., Andrews, S., and Brunswick, N. (2003). Identification of significant marine and coastal areas in the Bay of Fundy Oceans and Coastal Management Division Fisheries and Oceans Canada. St. Andrews, NB.

Carpio, Y., Basabe, L., Acosta, J., Rodríguez, A., Mendoza, A., Lisperger, A., … Estrada, M. P. (2011). Novel gene isolated from *Caligus rogercresseyi*: A promising target for vaccine development against sea lice. Vaccine, 29(15), 2810–2820. https://doi.org/10.1016/j.vaccine.2011.01.109

Carr, J., and Whoriskey, F. (2004). Sea lice infestation rates on wild and escaped farmed Atlantic salmon (*Salmo salar* L.) entering the Magaguadavic River, New Brunswick, 723–729. https://doi.org/10.1111/j.1365-2109.2004.01094.x

Chang, B. D., and Page, F. H. (2014). Historical and spatial trends in selected aspects of the finfish aquaculture industry in southwestern New Brunswick, Bay of Fundy, 1978-2012. Canadian Technical Report of Fisheries and Aquatic Sciences 3112. Ottawa, ON: Government of Canada Fisheries and Oceans.

Chang, B. D., Page, F. H., Beattie, M. J., and Hill, B. W. H. (2011). Sea Louse Abundance on Farmed Salmon in the Southwestern New Brunswick Area of the Bay of Fundy. In S. Jones and R. Beamish (Eds.), Salmon Lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2005a). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry of Cobscook Bay, Maine and adjacent southwestern New Brunswick. Canadian Technical Report of Fisheries and Aquatic Sciences 2598. St. Andrews, NB: Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and

McCurdy, E. P. (2005b). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry of Letete Passage, Back Bay, Bliss Harbour, and Lime Kiln Bay in Southwestern New Brunswick. Canadian Technical Report of Fisheries and Aquatic Sciences 2063. St. Andrews, NB: Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2006a). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry of Eastern Grand Manan Island, Bay of Fundy. Canadian Technical Report of Fisheries and Aquatic Sciences 2621. St. Andrews, NB: Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2006b). Water circulation and management of Infectious Salmon Anemia in the salmon aquaculture industry in Passamaquoddy Bay, Southwestern New Brunswick. Canadian Technical Report of Fisheries and Aquatic Sciences 2622. St. Andrews, NB: Department of Fisheries and Oceans.

Chang, B. D., Page, F. H., Losier, R. J., Lawton, P., Singh, R., and Greenberg, D. A. (2007). Evaluation of Bay Management Area Scenarios for the Southwestern New Brunswick Salmon Aquaculture Industry: Aquaculture Collaborative Research and Development Program Final Project Report. Canadian Technical Report of Fisheries and Aquatic Sciences, 2722. https://doi.org/10.1017/CBO9781107415324.004

Costello, M. J. (2009). The global economic cost of sea lice to the salmonid farming industry. Journal of Fish Diseases, 32, 115–118.

Dempster, T., Overton, K., Bui, S., Stien, L. H., Oppedal, F., Karlsen, Coates, A., Phillips, B. L., & Barrett, L. T. (2021). Farmed salmonids drive the abundance, ecology and evolution of parasitic salmon lice in Norway. Aquaculture Environment Interactions, 13, 237–248. https://doi.org/10.3354/AEI00402

Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). Bay of Fundy Marine Aquaculture Sites [map]. 1:125000. New Brunswick, Canada.

Department of Fisheries and Oceans Canada. (2014). Archived - The Scotian Shelf: An Atlas of Human Activities - Atlas.

Drinkwater, K. F. (1987). Mean Temperature and Salinity Conditions Near the Entrance to the Bay of Fundy, 1951-80. NAFO Science Council Studies, 71–73.

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

Elmoslemany, A., Revie, C. W., Milligan, B., Stewardson, L., and Vanderstichel,

R. (2015). Wild juvenile salmonids in Muchalat Inlet, British Columbia, Canada: Factors associated with sea lice prevalence. Diseases of Aquatic Organisms, 117(2), 107–120. https://doi.org/10.3354/dao02939

Elmoslemany, A., Whyte, S. K., Revie, C. W., and Hammell, K. L. (2013). Sea lice monitoring on Atlantic salmon farms in New Brunswick, Canada: Comparing audit and farm staff counts. Journal of Fish Diseases, 36(3), 241–247. https://doi.org/10.1111/jfd.12051

Fast, M. D. (2014). Fish immune responses to parasitic copepod (namely sea lice) infection. Developmental and Comparative Immunology, 43(2), 300–312. https://doi.org/10.1016/j.dci.2013.08.019

Fast, M. D., and Dalvin, S. (2020). Lepeophtheirosis (*Lepeophtheirus salmonis*). In P. T. K. Woo, J.-A. Leong, and K. Buchmann (Eds.), Climate Change and Infectious Fish Diseases (p. 529). Wallingford, UK; Boston, USA: CAB International.

Fife, F. J., Goreham, R. L. M., and Page, F. H. (2015). A century of monitoring station Prince 6 in the St. Croix River estuary of Passamaquoddy Bay. Department of Fisheries and Oceans Canada; Ecosystems and Oceans Science; Environment Canada.

Garrett, C.J.R. (1972). Tidal resonance in the Bay of Fundy and Gulf of Maine. Nature, 238, 441-443.

Gautam, R., Boerlage, A. S., Vanderstichel, R., Revie, C. W., and Hammell, K. L. (2016). Variation in pre-treatment count lead time and its effect on baseline estimates of cage level sea lice abundance. Journal of Fish Diseases, 39, 1297–1303. https://doi.org/10.1111/jfd.12460

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017a). Effect of timing of count events on estimates of sea lice abundance and interpretation of effectiveness following topical treatments. Journal of Fish Diseases, 40, 367–375. https://doi.org/10.1111/jfd.12519

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017b). Evaluating topical treatment effectiveness in the control of sea lice burdens on Atlantic salmon in New Brunswick, Canada. Journal of Fish Diseases, 40, 895–905. https://doi.org/10.1111/jfd.12569

Godin, G. (1968). The 1965 current survey of the Bay of Fundy: A new analysis of the data and an interpretation of results. Manuscript Report Series 8, 97 pp. Marine Science Branch, Energy Mines and Resources, Ottawa.

Gonzalez, E.B. and de Boer, F. (2017). The development of the Norwegian wrasse fishery and the use of wrasses as cleaner fish in the salmon aquaculture industry. Fishery science 83, 661-670.

Government of Canada. (2019). Atlantic Salmon inner Bay of Fundy population: Action Plan, 2019.

Government of New Brunswick. (2018). General Regulation, NB Reg 91-158, 1–14. http://canlii.ca/t/544vm

Graham, E.M., Baird, A.H., and Connolly, S.R. (2008). Survival dynamics of scleractinian coral larvae and implications for dispersal. Coral Reefs, 27(3): 529–539. doi:10.1007/s00338-008-0361-z.

Greenberg, D.A. (1983). Modeling the mean barotropic circulation in the Bay of Fundy and Gulf of Maine. Journal of Physical Oceanography, 13: 886-904.

Greenberg, D. A. (1984). A Review of the Physical Oceanography of the Bay of Fundy. Update of the Marine Environmental Consequences of Tidal Power Development in the Upper Reaches of the Bay of Fundy, 9–30.

Greenberg, D.A., Shore, J.A., Page, F.H., and Dowd, M. (2005). A finite element circulation model for embayments with drying intertidal areas and its application to the Quoddy Region of the Bay of Fundy. Ocean Modelling 10: 211–231.

Grøntvedt, R.N., Nerbøvik, I.K.G., Viljugrein, H., Lillehaug, A., Nilsen, H., and Gjevre, A.G. (2015). Norwegian Veterinary Institute's Report Series: 13. Thermal de-licing of salmonid fish – documentation of fish welfare and effect. Norwegian Veterinary Institute.

Hall, L. M., and Murray, A. G. (2018). Describing temporal change in adult female *Lepeophtheirus salmonis* abundance on Scottish farmed Atlantic salmon at the national and regional levels. Aquaculture, 489, 148–153. https://doi.org/10.1016/j.aquaculture.2018.01.040

Hamre, L., Bui, S., Oppedal, F., Skern-Mauritzen, R., and Dalvin, S. (2019). Development of the salmon louse *Lepeophtheirus salmonis* parasitic stages in temperatures ranging from 3 to 24°C. Aquaculture Environment Interactions, 11, 429–443. https://doi.org/10.3354/aei00320

Hannah, C.G., Shore, J., Loder, J.W., and Naimie, C.E. (2001). Seasonal circulation on the western and central Scotian Shelf. Journal of Physical Oceanography 31: 591-615.

Hayward, C. J., Andrews, M., and Nowak, B. F. (2011). *Lepeophtheirus salmonis* - A Remarkable Success Story. In S. Jones and R. Beamish (Eds.), Salmon Lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley and Sons, Inc.

Hemmingsen, W., MacKenzie, K., Sagerup, K., Remen, M., Bloch-Hansen, K., and Dagbjartarson Imsland, A. K. (2020). *Caligus elongatus* and other sea lice of the genus *Caligus* as parasites of farmed salmonids: A review. Aquaculture, 522, 735160. https://doi.org/10.1016/j.aquaculture.2020.735160

Hijmans, R.J., van Etten, J., Sumner, M., Cheng, J., Baston, D., Bevan, A., … Wueest, R. (2019). raster: Geographic Data Analysis and Modeling. R package version 3.0-12. https://CRAN.R-project.org/package=raster

Hogans, W. E. (1995). Infection dynamics of sea lice, *Lepeophtheirus salmonis* (Copepoda: Caligidae) parasitic on Atlantic salmon (*Salmo salar*) cultured in marine waters of the lower Bay of Fundy. Canadian Technical Report of Fisheries and Aquatic Sciences 2067. St. Andrews, NB: Department of Fisheries and Oceans.

Hogans, W.E. and Trudeau, D.J. (1989). Preliminary studies on the biology of sea lice, *Caligus elongatus*, *Caligus curtus* and *Lepeophtheirus salmonis* (Copepoda: Caligoida) parasitic on cage-cultured salmonids in the lower Bay of Fundy. Canadian Technical Report of Fisheries and Aquatic Sciences 1715. St. Andrews, NB: Department of Fisheries and Oceans.

Igboeli, O. O., Fast, M. D., Heumann, J., and Burka, J. F. (2012). Role of P-glycoprotein in emamectin benzoate (SLICE®) resistance in sea lice, *Lepeophtheirus salmonis*. Aquaculture, 344–349, 40–47. https://doi.org/10.1016/j.aquaculture.2012.03.026

Johnson, S. C., Treasurer, J. W., Bravo, S., and Nagasawa, K. (2004). A review of the impact of parasitic copepods on marine aquaculture. Zoological Studies, 43(2), 229–243. Retrieved from http://www.sinica.edu.tw/zool/zoolstud/43.2/229.pdf

Jones, P. G., Hammell, K. L., Dohoo, I. R., and Revie, C. W. (2012). Effectiveness of emamectin benzoate for treatment of *Lepeophtheirus salmonis* on farmed Atlantic salmon *Salmo salar* in the Bay of Fundy, Canada. Diseases of Aquatic Organisms, 102(1), 53–64. https://doi.org/10.3354/dao02517

Jones, P. G., Hammell, K. L., Gettinby, G., and Revie, C. W. (2013). Detection of emamectin benzoate tolerance emergence in different life stages of sea lice, *Lepeophtheirus salmonis*, on farmed Atlantic salmon, *Salmo salar* L. Journal of Fish Diseases, 36(3), 209–220. https://doi.org/10.1111/jfd.12022

Kaur, K., Helgesen, K. O., Bakke, M. J., and Horsberg, T. E. (2015). Mechanism behind resistance against the organophosphate azamethiphos in salmon lice (*Lepeophtheirus salmonis*). PLoS ONE, 10(4), 1–20. https://doi.org/10.1371/journal.pone.0124220

Keen, J. (2020). The effects of the anti-sea lice chemotherapeutants Salmosan® and Interox® Paramove® 30 on marine zooplankton. Master of Environmental Toxicology, Simon Fraser University, Burnaby, Canada.

Kristoffersen, A. B., Qviller, L., Helgesen, K. O., Vollset, K. W., Viljugrein, H., and Jansen, P. A. (2018). Quantitative risk assessment of salmon louse-induced mortality of seaward-migrating post-smolt Atlantic salmon. Epidemics, 23, 19–33. https://doi.org/10.1016/j.epidem.2017.11.001

Kristoffersen, A. B., Rees, E. E., Stryhn, H., Ibarra, R., Campisto, J. L., Revie, C. W., and St-Hilaire, S. (2013). Understanding sources of sea lice for salmon farms in Chile. Preventive Veterinary Medicine, 111(1–2), 165–175. https://doi.org/10.1016/j.prevetmed.2013.03.015

L'Aventure, J. (1987). The case history of an Atlantic salmon farm. In: Atlantic Canada Aquaculture Workshop − Proceedings, Volume II (Salmonids). Ed., Roache, J.F. Canada Department of Fisheries and Oceans, Atlantic Fisheries Technology Program (AFTP), General Education Series, No. 5, pp. 28–34

Lacroix, G. L. and Knox, D. (2005). Distribution of Atlantic salmon (*Salmo salar*) postsmolts of different origins in the Bay of Fundy and Gulf of Maine and evaluation of factors affecting migration, growth, and survival. Canadian Journal of Fisheries and Aquatic Sciences, 62(6), 1363–1376. https://doi.org/10.1139/f05-055

McEwan, G. F., Groner, M. L., Fast, M. D., Gettinby, G., and Revie, C. W. (2015). Using agent-based modelling to predict the role of wild refugia in the evolution of resistance of sea lice to chemotherapeutants. PLoS ONE, 10(10), 1–23. https://doi.org/10.1371/journal.pone.0139128

MacPhee, D. and Moore, M. (2007). A retrospective study of sea lice dynamics and treatments on aquaculture sites in Passamaquoddy Bay, New Brunswick in 2006. Report for the New Brunswick Department of Agriculture and Aquaculture, St. George, NB, 20 p.

Mustafa, A., Rankaduwa, W., and Campbell, P. (2001). Estimating the cost of sea lice to salmon aquaculture in eastern Canada. Canadian Veterinary Journal, 42(1), 54–56. https://doi.org/10.1111/j.1365-2761.2008.01011.x

New Brunswick Department of Agriculture Fisheries and Oceans Canada. (2013). Bay of Fundy Marine Aquaculture Site Allocation Policy.

Overton, K., Dempster, T., Oppedal, F., Kristiansen, T. S., Gismervik, K., and Stien, L. H. (2019). Salmon lice treatments and salmon mortality in Norwegian aquaculture: A review. Reviews in Aquaculture, 11(4), 1398–1417. https://doi.org/10.1111/raq.12299

Page, F. H., Chang, B. D., Losier, R. J., Greenberg, D. A., Chaffey, J. D., and McCurdy, E. P. (2005). Water Circulation and Management of Infectious Salmon Anemia in the Salmon Aquaculture Industry of Southern Grand Manan Island, Bay of Fundy. Canadian Technical Report of Fisheries and Aquatic Sciences 2595. St. Andrews, NB: Department of Fisheries and Oceans.

Parsons, A. E., Escobar-Lux, R. H., Sævik, P. N., Samuelsen, O. B., and Agnalt, A. L. (2020). The impact of anti-sea lice pesticides, azamethiphos and deltamethrin, on European lobster (*Homarus gammarus*) larvae in the Norwegian marine environment. Environmental Pollution, 264, 114725. https://doi.org/10.1016/j.envpol.2020.114725

Patanasatienkul, T., Sanchez, J., Rees, E. E., Pfeiffer, D., and Revie, C. W. (2015). Space-time cluster analysis of sea lice infestation (*Caligus clemensi* and *Lepeophtheirus salmonis*) on wild juvenile Pacific salmon in the Broughton Archipelago of Canada. Preventive Veterinary Medicine, 120(2), 219–231. https://doi.org/10.1016/j.prevetmed.2015.03.006

Peterson, R.H., Page, F., Steeves, G.D., Wildish, D.J., Harmon, P., and Losier, R. (2001). A survey of 20 Atlantic salmon farms in the Bay of Fundy: influence of environmental and husbandry variables on performance. Canadian Technical Report of Fisheries and Aquatic Sciences 2337. St. Andrews, NB: Department of Fisheries and Oceans.

Pike, A W. and Wadsworth, S. L. (1999). Sea lice on salmonids: Their biology and control. Advances in Parasitology, 44. https://doi.org/10.1016/S0065-308X(08)60233-X

Rees, E. E., St-Hilaire, S., Jones, S. R. M., Krkošek, M., DeDominicis, S., Foreman, M. G. G., … Revie, C. W. (2015). Spatial patterns of sea lice infection among wild and captive salmon in western Canada. Landscape Ecology, 30(6), 989–1004. https://doi.org/10.1007/s10980-015-0188-2

Revie, C. W., Gettinby, G., Treasurer, J. W., Rae, G. H., and Clark, N. (2002). Temporal, environmental and management factors influencing the epidemiological patterns of sea lice (*Lepeophtheirus salmonis*) infestations on farmed Atlantic salmon (*Salmo salar*) in Scotland. Pest Management Science, 58(6), 576–584. https://doi.org/10.1002/ps.476

Rikardsen, A. H. (2004). Seasonal occurrence of sea lice *Lepeophtheirus salmonis* on sea trout in two north Norwegian fjords. Journal of Fish Biology, 65(3), 711–722. https://doi.org/10.1111/j.1095-8649.2004.00478.x

Rittenhouse, M. A., Revie, C. W., and Hurford, A. (2016). A model for sea lice (*Lepeophtheirus salmonis*) dynamics in a seasonally changing environment. Epidemics, 16, 8–16. https://doi.org/10.1016/j.epidem.2016.03.003

Saksida, S., Bricknell, I., Robinson, S., and Jones, S. (2015). Population ecology and epidemiology of sea lice in Canadian waters. DFO Canadian Science Advisory Secretariat Science Advisory Report, 2015/114(February), 34 p.

Saksida, S., Constantine, J., Karreman, G. A., and Donald, A. (2007). Evaluation of sea lice abundance levels on farmed Atlantic salmon (*Salmo salar* L.) located in the Broughton Archipelago of British Columbia from 2003 to 2005. Aquaculture Research, 38(3), 219–231. https://doi.org/10.1111/j.1365-2109.2007.01651.x

Samsing, F., Oppedal, F., Dalvin, S., Johnsen, I., Vågseth, T., and Dempster, T. (2016). Salmon lice (*Lepeophtheirus salmonis*) development times, body size, and reproductive outputs follow universal models of temperature dependence. Canadian Journal of Fisheries and Aquatic Sciences, 73, 1841–1851. https://doi.org/10.1139/cjfas-2016-0050

StataCorp (2019). Stata: Release 16. Statistical Software. StataCorp LLC, TX, USA.

Statistics Canada; Government of Canada. (2020). Table 32-10-0107-01 Aquaculture, production and value. https://doi.org/10.25318/3210010701-eng

Stien, A., Bjørn, P. A., Heuch, P. A., and Elston, D. A. (2005). Population dynamics of salmon lice *Lepeophtheirus salmonis* on Atlantic salmon and sea trout, 290, 263–275.

Stuart, R. 1990. Sea lice, a maritime perspective. Bulletin of the Aquaculture Association of Canada 90, 18–24.

Thakur, K. K., Vanderstichel, R., Barrell, J., Stryhn, H., Patanasatienkul, T., and Revie, C. W. (2018). Comparison of remotely-sensed sea surface temperature and salinity products with in situ measurements from British Columbia, Canada. Frontiers in Marine Science, 5, 1–11. https://doi.org/10.3389/fmars.2018.00121

Torrissen, O., Jones, S., Asche, F., Guttormsen, A., Skilbrei, O. T., Nilsen, F., … Jackson, D. (2013). Salmon lice - impact on wild salmonids and salmon aquaculture. Journal of Fish Diseases, 36(3), 171–194. https://doi.org/10.1111/jfd.12061

Tully, O. and Nolan, D. T. (2002). A review of the population biology and host-parasite interactions of the sea louse *Lepeophtheirus salmonis* (Copepoda: Caligidae). Parasitology, 124 Suppl(2002), S165–S182. https://doi.org/10.1017/S0031182002001889

UK Met Office (2005). GHRSST Level 4 OSTIA Global Foundation Sea Surface Temperature Analysis. CA, USA. Retrieved from https://doi.org/10.5067/GHOST-4FK01

Werkman, M., Green, D. M., Murray, A. G., and Turnbull, J. F. (2011). The effectiveness of fallowing strategies in disease control in salmon aquaculture assessed with an SIS model. Preventive Veterinary Medicine, 98(1), 64–73. https://doi.org/10.1016/j.prevetmed.2010.10.004

Whyte, S. K., Westcott, J. D., Elmoslemany, A., Hammell, K. L., and Revie, C. W. (2013). A fixed-dose approach to conducting emamectin benzoate tolerance assessments on field-collected sea lice, *Lepeophtheirus salmonis*. Journal of Fish Diseases, 36(3), 283–292. https://doi.org/10.1111/jfd.12055

WoRMS Editorial Board. (2021). World Register of Marine Species. https://doi.org/10.14284/170

3 Estimating the dispersal of *Lepeophtheirus salmonis* sea lice within and among Atlantic salmon sites of the Bay of Fundy, New Brunswick

3.1 Abstract

The objective of this study was to estimate the impact of infestation pressures on the abundance of the parasitic sea louse, *Lepeophtheirus salmonis*, in the Bay of Fundy, New Brunswick (NB), Canada, using the Fish-iTrends database for the years 2009-2018. Infestation pressures were calculated as time-lagged weighted averages of the abundance of adult female (AF) sea lice within a site (internal infestation pressure: IIP) and among sites (external infestation pressure: EIP). The EIP weights were calculated from seaway distances among sites and a Gaussian kernel density for bandwidths of 5 to 60 km. The EIP with a bandwidth of 10 km had the best fit, as determined with Akaike's Information Criterion, and historical AF sea lice abundance. This estimated dispersal distance of 10 km was similar to previous studies in Norway, Scotland, and in New Brunswick. The infestation pressures estimated from empirical AF sea lice abundance within and among sites significantly increased the abundance of AF sea lice ($p < 0.001$). This study concludes that sea lice burdens within Atlantic salmon farms in the Bay of Fundy, NB, are affected by within site management and could be improved by synchronizing treatments between sites.

3.2 Introduction

The Atlantic salmon, *Salmo salar* (Linnaeus: WoRMS, 2020), aquaculture industry in the Bay of Fundy, New Brunswick (NB), Canada, has been exposed to the parasitic sea louse, *Lepeophtheirus salmonis* (Krøyer: WoRMS, 2020), since its early development in the 1980's (Hogan and Trudeau, 1989) and its presence has required management since the mid-1990's (Hogans, 1995). *L. salmonis* negatively affects salmon welfare by rasping mucus, skin and underlying tissue causing the potential for increased mucus production, skin wounds (Tully and Nolan 2002; Johnson et al., 2004) and immunocompromising its host (Fast, 2014). Moreover, sea lice have been identified as a potential transmission route for the infectious salmon anemia virus (ISAv), which itself can cause high mortality rates in Atlantic salmon (Nylund et al., 1993, 1994), and can be further exacerbated by sea lice infection (Barker et al., 2019).

Atlantic salmon producers seek to reduce sea lice abundance on their fish to mitigate the negative effects caused by this parasite and to keep in check the potential for adult female sea lice to reproduce. The control of *L. salmonis* involves increased costs of production due to decreased growth rate and treatment (Costello, 2006; Abolofia et al., 2017).

Aside from industry-wide measures such as fallowing and single year class area management, control options for sea lice include the use of chemotherapeutic agents and non-medicinal operations (see Overton et al., 2019 for review). The effectiveness of these treatments varies with the drug or operation used, timing, method of application, sea lice life stage, season, pre-treatment abundance level

(Gautam et al., 2017b), frequency of application (Revie et al., 2005), and potential resistance of sea lice to the drug (e.g., emamectin benzoate [Slice®, Merck Animal Health, USA]; Westcott et al., 2008; Igboeli et al., 2012; Jones et al., 2012).

The Fish-iTrends is a web-based data management system that compiles data on sea lice and their treatments in Atlantic salmon marine sites for the province of NB and other Atlantic provinces (Gautam et al., 2016; additional details found in Supplemental Information). This database aggregates sea lice counts for certain life stages from all aquaculture sites in their respective Bay Management Areas (BMA; additional details in Chang et al., 2011).

The sea louse undergoes multiple moults in its 8 life stages (see Johnson and Albright, 1991; updated by Hamre et al., 2013). The life stages of note are the adult male and female sea lice, the planktonic stages (nauplii I and II, and copepodid), and the pre-adult stages (I and II). The planktonic stages are passively transported by water currents (Amundrud and Murray, 2009). Water salinity and temperature affects the rate of development, mortality (Stien et al., 2005; Groner et al., 2016) and attachment of each life stage (Tucker et al., 2000). Sea lice development was more rapid at higher water temperatures (Stien et al., 2005; Hamre et al., 2019), while salinities below 22 ppt reduced survival (Groner et al., 2016). Modelling of sea lice infestation therefore often accounts for maturation and mortality in relation to water salinity and temperature (e.g., Revie et al., 2005).

Modelling the effective distance that planktonic sea lice travel can be accomplished with multiple methods: physical and biophysical models, and empirical methods (e.g., plankton tows in Nelson et al., 2017). Physical models are numerical (i.e., oceanographic) models of currents that respond to temperature, depth, wind, and other physical characteristics of the water body (e.g., Murray and Gillibrand, 2006). The physical models can be adjusted with biological estimates of mortality, survival, and development rates of the sea lice (e.g., Revie et al., 2005; Asplin et al., 2011, 2014).

A different approach to the estimation of dispersal distance and the impact of sea lice among aquaculture sites is with the use of the concept of infestation pressure. Simply, infestation pressure is a measure that represents the dose of exposure of infectious stages of sea lice to potential fish hosts. The infestation pressure can originate from within a site (internal infestation pressure, or IIP) and be derived from neighbouring sites (external infestation pressure, or EIP). Both sources of infestation pressure can be calculated using several methods (e.g., Aldrin et al., 2013; Kristoffersen et al., 2013, 2014, 2018; Aldrin et al., 2019). Cantrell et al. (2018, 2019) defined infestation pressure as the density of particles representing sea lice using a biophysical model in the Broughton Archipelago, British Columbia (BC). Sandvik et al. (2016) defined infestation pressure on a scale of low (0-1 louse) to high (> 10 lice) resulting from a biophysical model informed from field data. The present study, amongst others (e.g., Elghafghuf et al., 2020), propose data-driven methods for the estimation of infestation pressure.

Elghafghuf et al. (2020) evaluated multiple estimation approaches for infestation pressure using Fish-iTrends data from the aquaculture sites in Grand Manan, NB (see Figure 3.1). Five methods were evaluated using a multivariate autoregressive state-space model. The IIP and EIP were calculated using the fixed lag approach of mean reported adult female (AF) sea lice abundance with a time lag of one to four weeks (Kristoffersen et al., 2013), and other approaches (Kristoffersen et al., 2014, 2018; Aldrin et al., 2019). The fixed lag approach had the lowest Akaike's information criterion (AIC) for the model that included both the IIP and EIP. The authors concluded that an EIP calculated with the abundance of sea lice weighted by a Gaussian KDE bandwidth of 12.5 km resulted in the lowest AIC (Elghafghuf et al., 2020).

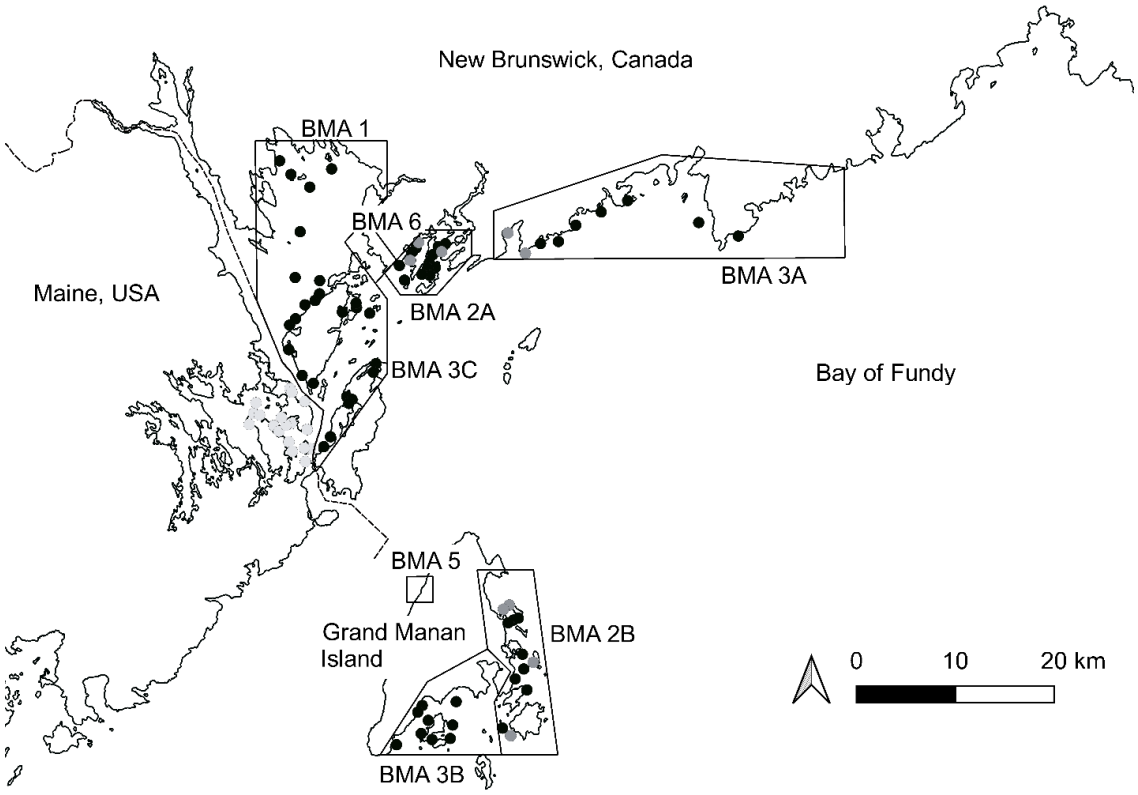

Figure 3.1. The Bay of Fundy, New Brunswick, Canada, study area demonstrating Bay Management Areas (BMA) and Atlantic salmon aquaculture sites. Sites included (black circle) and excluded (grey circle) from the final multivariable linear mixed model for the abundance of adult female *Lepeophtheirus salmonis*. Abundances of sea lice were not available from sites from Maine, USA (white circle). The locations of sites and BMA were adapted from the New Brunswick Bay of Fundy Marine Aquaculture sites map of 2017 (Department of Energy and Resource Development/Agriculture, and Fisheries, 2017).

The bandwidth of the Gaussian KDE may be taken to represent the dispersal of planktonic sea lice among neighbouring sites. Kristoffersen et al. (2013) determined the model with the lowest AIC had a kernel weighted intensity of mean reported abundance of gravid AF sea lice adjusted with a KDE of 30 km. Other reports suggest a range of dispersal distances from 7 to 200 km. The variation in dispersal distance is the result of highly variable environmental conditions such as wind (Salama et al., 2018), current speed and direction

(Asplin et al., 2014), distance from shore (Penston et al., 2004), methodology to obtain the estimate, among others.

The dispersal of sea lice, for the whole of the Bay of Fundy, NB, has not been previously estimated. Our objective was to estimate the impact of the IIP and EIP on the abundances of sea lice on farmed Atlantic salmon in the Bay of Fundy, NB. This information has implications for the management of the NB aquaculture industry as it may suggest the need for a different BMA design for the synchronization of site production cycles (i.e., stocking and harvesting), as well as for synchronizing mitigation strategies among sites to maximize efficiency. The Fish-iTrends database offers the opportunity for empirical estimation of the maximum dispersal distance of sea lice using modelling of the IIP, EIP, and treatments.

3.3 Materials and methods

3.3.1 Study area and population

All available sea lice (*L. salmonis*) information from aquaculture sites in the Bay of Fundy, New Brunswick (Figure 3.1), between July 2009 and November 2018, were retrieved from the Fish-iTrends data management system and included in this study. Data consisted of routine counts of pre-adult and adult male (PAAM) and AF (with and without egg strings) sea lice on Atlantic salmon from several cages at each site during their production cycle (additional details found in Supplemental Information; see Gautam et al., 2016 for detailed sampling description). The Bay of Fundy adjoins the state of Maine, USA (Figure 3.1), and

data on sea lice abundance and active sites from the Maine aquaculture industry were not available for this study.

3.3.2 Selection and preparation of the outcome and abundances

Adult female sea lice counts were selected as the outcome variable for modelling. AF sea lice counts had lower variance than PAAM sea lice counts during comparison of audit technician and site staff counts, suggesting higher repeatability of AF sea lice counts within cages (Elmoslemany et al., 2013). This is perhaps not surprising since PAAM sea lice counts are based purely on morphology, which include two life stages with a disproportionate sex ratio (i.e., pre-adult males and females, and adult males). In addition, AF sea lice develop more slowly than their male counterparts (Hamre et al., 2019); therefore, the rate of population growth is limited by this slowest life stage.

Sea lice abundances were calculated for each observation (i.e., count date; Eq. 3.1). The normalized counts of sea lice are the count of sea lice divided by the number of fish sampled with the offset of 1.

$$\text{abundance of } \mathrm{AF}_{ic}(t) = \ln\left({}^{(\mathrm{AF}_{ic}+1)}/_{\text{no. fish}}\right) \qquad \text{Eq. 3.1}$$

Where *AF* ~ adult female sea lice for *c* ~ cage within site i and *t* ~ time (day).

Caligus elongatus and *L. salmonis* chalimus sea lice were also recorded but were not considered suitable to be included in the modelling due to their small numbers.

3.3.3 Data structure

Sea lice abundances were measured at the cage level. The aquaculture industry is hierarchically structured with multiple cages within sites and multiple sites within BMAs. Regulations stipulate when sites can stock and harvest fish within BMAs (production cycles), therefore, cages and sites were uniquely identified according to their specific production cycle within BMAs.

Observations (counts) occurred as repeated measurements of sea lice throughout the production cycle. Observations were further grouped into segments, where a segment was defined as the first observation after a topical treatment followed by all additional observations until the following treatment within a production cycle. This organization of observations into segments also allowed us to account for the treatment effect on lice abundance.

A segment was limited to a single cohort of salmon, as defined by salmon with the same fish group identifier, cage, site, and production cycle. The fish group identifier was used in the Fish-iTrends database to follow groups of salmon that were sometimes split or moved between multiple cages during the production cycle, including at the time of a wellboat treatment.

Descriptors for the segments included their length (the number of days from the start to the end of the segment, including the treatment date), size (the number of observations that contribute to the segment), and maximum gap (the largest number of days between observations of a segment). Short segments occurred when there were multiple treatments in short succession within a cage. Evaluation of the segments using the length, size and maximum gap information

identified segments with extreme length, size and maximum gap that resulted in the creation of strict inclusion criteria for modelling.

3.3.4 Defining internal and external infestation pressure

The internal and external infestation pressures were estimated using a fixed lag approach, similar to Elghafghuf et al. (2020) and Kristoffersen et al. (2013). The fixed lag is the time for AF sea lice to produce eggs and develop into a new generation of AF and PAAM sea lice. The lag time for PAAM sea lice abundance was set at 28 days, and for AF sea lice abundance at 21 days. These lag times were chosen based upon a combination of a literature review, and the estimated most rapid development at the sea temperature annual maximum in the study area (15 °C near Saint Andrews, NB [World Sea Temperature, 2020]). The closest analog to our settings was an estimated AF sea lice that contributed to the next generation of PAAM sea lice at a neighbouring site with a lag of approximately four weeks at water temperatures of 14 °C (Aldrin et al., 2019).

The dose of exposure of infectious stages of sea lice to potential fish hosts within a site (IIP) was represented by the triangular weighted 15 day average of AF and PAAM sea lice abundances divided by the sum of the weights at a site 21 and 28 days prior to an observation, respectively. The EIP consisted of the triangular-weighted temporal and Gaussian KDE-weighted spatial average abundance of AF sea lice at other sites 21 days prior to an observation (additional details found in Supplemental Information). Gaussian KDE weights used seaway distances among study sites, which were calculated with the ‘gdistance’ package, version 1.3-1 (van Etten and de Sousa, 2019) in R (R Core Team, 2020). Averages of AF

and PAAM sea lice abundances were calculated across all observations (whether part of a segment or not) for a site, and no adjustments were made for observations that occurred prior to and after treatments.

3.3.5 Baseline abundance of adult female sea lice

The abundance of sea lice after a bath treatment was hypothesized to be associated with the abundance of AF and PAAM sea lice prior to the application of the treatment (Gautam et al., 2017a). Gautam et al. (2017a) defined a treatment as the application of azamethiphos (Salmosan® Vet, Benchmark Animal Health, United Kingdom) or hydrogen peroxide (Interox® Paramove® 50^{TM}, Solvay Chemicals Inc., Belgium) using a tarp enclosure or wellboat. The abundance of sea lice prior to a treatment was defined as the baseline abundance of AF sea lice and was identified for each segment. The baseline abundance of AF sea lice was the first observation within 7 days prior to and including the treatment date, provided it was indicated in the database as a pre-treatment count on the treatment date; observations also had to belong to the same cohort of salmon.

3.3.6 Topical and in-feed treatments

The topical treatments at the start of a segment were assigned to all the observations within that segment. Hydrogen peroxide (Interox® Paramove® 50^{TM} and Aquaparox 50^{TM}, Alpha Chemical Limited, Canada) treatments were combined into a single category. Azamethiphos treatments were categorized according to their method of application: tarpaulin, skirt, or wellboat.

A dichotomous variable was created to represent the application of an in-feed

treatment (emamectin benzoate, teflubenzuron [Calicide®, Nutreco ARC Ltd., Netherlands], and avermectin [Ivomec®, Boehringer Ingelheim, Germany, and Noromectin®, Norbrook Laboratories Ltd., United Kingdom]) for a group of fish (i.e., cage level) prior to the start of a segment: present (1) and absent (0).

3.3.7 Sea surface temperature

The UKMO OSTIA SST product (UK Met Office, 2005) was utilized for the sea surface temperature (hereafter, temperature) of the study area. This remote sensing product was selected instead of the *in-situ* temperature measurements to avoid missing values and inconsistencies in measuring technique. Thakur et al. (2018) found that the UKMO OSTIA SST product had the highest concordance correlation coefficient and index of agreement compared to measurements of temperature taken on aquaculture sites in the Broughton Archipelago between 2003 and 2016.

Daily temperature values were extracted for the duration of the study for the centroid location of each aquaculture site polygon utilizing the ‘raster’ package, version 3.0-12 (Hijmans et al., 2020) in R. The spatial resolution was 1/20 degrees, which is approximately 6 km at the 45 degree latitude of the Bay of Fundy, NB; sites that occupied the same temperature grid shared temperature values.

3.3.8 Modelling

3.3.8.1 Inclusion criteria

Segment inclusion for modeling was strict. Segment length was constrained to 5 and 300 days, inclusive. Segment size was constrained to three and 20 observations, inclusive. Segments with a large gap between observations (greater than and equal to 90 days) were excluded. The BMAs 5 and 6 were not included in the modelling of the outcome as the BMA 5 is a conservation area for wild salmon and BMA 6 is for species other than Atlantic salmon. Moreover, there were no segments in the BMA 6 and only 4 segments with 13 observations in the BMA 5. A sensitivity analysis was performed to evaluate the effect of the strict inclusion criteria on the model (additional details found in Supplemental Information).

3.3.8.2 Multivariate linear mixed model

A multilevel mixed-effects linear regression (Pinheiro and Bates, 2000) was used to model the abundance of AF sea lice. The mixed-effects model is robust, improves the estimation of standard errors where there is clustering of the outcome and allows the assignment of an error correlation structure. Random effects included the site-cycle, cage-cycle, and segments. BMA was set as a fixed effect.

Models were fit with the restricted maximum likelihood estimation unless nested models were compared. The AIC was used to compare models with different residual correlation structure, different random effects levels, and the EIP with

different bandwidths. The final model was utilized to compare the EIP calculated with bandwidths of 5 to 60 km. The model was fit with an exponential correlation structure to the errors, stratified by three temperature profiles (low = 3-7 °C, medium = 8-11 °C, and high = 12-16 °C). The exponential correlation structure was selected to account for non-uniform time gaps between observations.

The model assumptions of linearity, homogeneity of variance, and normality of residuals were assessed graphically for each level of the hierarchy (additional details found in Supplemental Information). The linearity of temperature effects was evaluated using locally weighted scatterplot smoothing (LOWESS) plots of the outcome against the temperature for each treatment. A quadratic regression and fractional polynomial regression were used to evaluate other transformations of the interaction term that could improve the model fit.

The effect of the loss of observations with the inclusion of the $baseline_{AF}$ predictor was evaluated (additional details found in Supplemental Information). The predicted temperatures for each treatment were calculated for the BMA 1 at the median values (in brackets) of IIP_{AF} (1.5), IIP_{PAAM} (2.0), EIP_{AF} at 10 km (0.3), and $baseline_{AF}$ (2.3).

All analyses, unless otherwise specified, were conducted in Stata® (2019, StataCorp), and statistical significance was set at $p < 0.050$.

3.4 Results

3.4.1 Descriptive statistics of the observations modelled

There was a total of 11,658 observations and 2,398 segments included in the final multivariable linear mixed model for the abundance of AF sea lice in Atlantic salmon sites in the Bay of Fundy between 2009 and 2018. Segments had on average 8.3 observations across an average of 70.2 days. The BMA 1, 2A, 2B, 3A, 3B, and 3C were included and had an average of 1,943.0 observations per group and 400.0 segments per group (Table 3.1). Observations were most frequent in the BMA 1 and 2A (48.1 % and 28.9 %, respectively). The most frequent observations were found in the highest temperature category (12-16 °C), followed by the middle (8-11 °C), and lowest categories (3-7 °C; Table 3.2). A total of 62 sites produced 114 site-cycles; 28 sites (45.2 %) had a single production cycle, 19 sites (30.7 %) had 2, 12 sites (19.4 %) had 3, and 3 sites (4.8 %) had 4 production cycles. There was a total of 685 cages that produced a total of 1,001 cage-cycles.

Table 3.1. Number of observations and segments per hierarchical level. n = 11,658, n/a = not applicable, † = nested groups of the final multivariable linear mixed model for the abundance of adult female *Lepeophtheirus salmonis* with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km.

		Observations per group			Segments per group		
Group var.	No. groups	Min.	Mean	Max.	Min.	Mean	Max.
BMA	6	86	1943.0	5609	18	400.0	1,173
Site	62	6	356.5	698	1	38.7	178
Site-cycle†	114	1	102.3	418	1	21.0	121
Cage	685	1	17.0	80	1	3.5	17
Cage-cycle†	1,001	1	11.6	53	1	2.4	11
Segment†	2,398	1	4.9	19	n/a	n/a	n/a

Table 3.2. Number of observations (number of segments) categorized by the Bay Management Areas (BMA) and three temperature profiles (temp. cat.).

BMA\Temp. cat.	3-7 °C	8-11 °C	12-16 °C	Total BMA
1	422 (89)	2,384 (461)	2,803 (623)	5,609 (1,173)
2A	235 (33)	1,064 (174)	2,072 (472)	3,371 (679)
2B	108 (25)	209 (40)	249 (50)	566 (115)
3A	126 (33)	258 (52)	594 (127)	978 (212)
3B	202 (39)	506 (102)	340 (60)	1,048 (201)
3C	0 (0)	0 (0)	86 (18)	86 (18)
Total temp. cat.	1,093 (219)	4,421 (829)	6,144 (1,350)	11,658 (2,398)

The normalized count of AF sea lice (AF count divided by the number of fish counted and offset by 1) was strongly right skewed. Therefore, these observations were log-transformed to obtain the abundance of AF sea lice (Eq. 3.1). The abundance of AF sea lice (i.e., outcome [Eq. 3.1]) had a mean of 1.20 (SD 1.52). The IIP_{AF} and EIP_{AF} had means of 1.15 (SD 1.34) and 0.28 (SD 0.36), respectively (Table 3.3). The $baseline_{AF}$ mean was 1.84 (SD 1.44). The IIP_{PAAM} was 1.96 (SD 1.20). The inclusion of $baseline_{AF}$ as a predictor in the model resulted in the loss of 6,104 observations and 1,325 segments. The average EIP_{AF} with a bandwidth of 10 km was 0.28 (SD 0.36). The range of seaway distances among the centroid locations of sites in our study area was 0.4 to 79.2 km. The median seaway distance for the modelled sites was 26.1 km.

Azamethiphos was applied the most frequently (n_{obs} = 7,021, n_{seg} = 1,491) of all the treatments (Figure 3.2). The tarpaulin method of application was the most common method of application for azamethiphos (n_{obs} = 5,491, n_{seg} = 1,228). The Hydrolicer® use was started in 2018 and had the fewest applications (n_{obs} = 58, n_{seg} = 12).

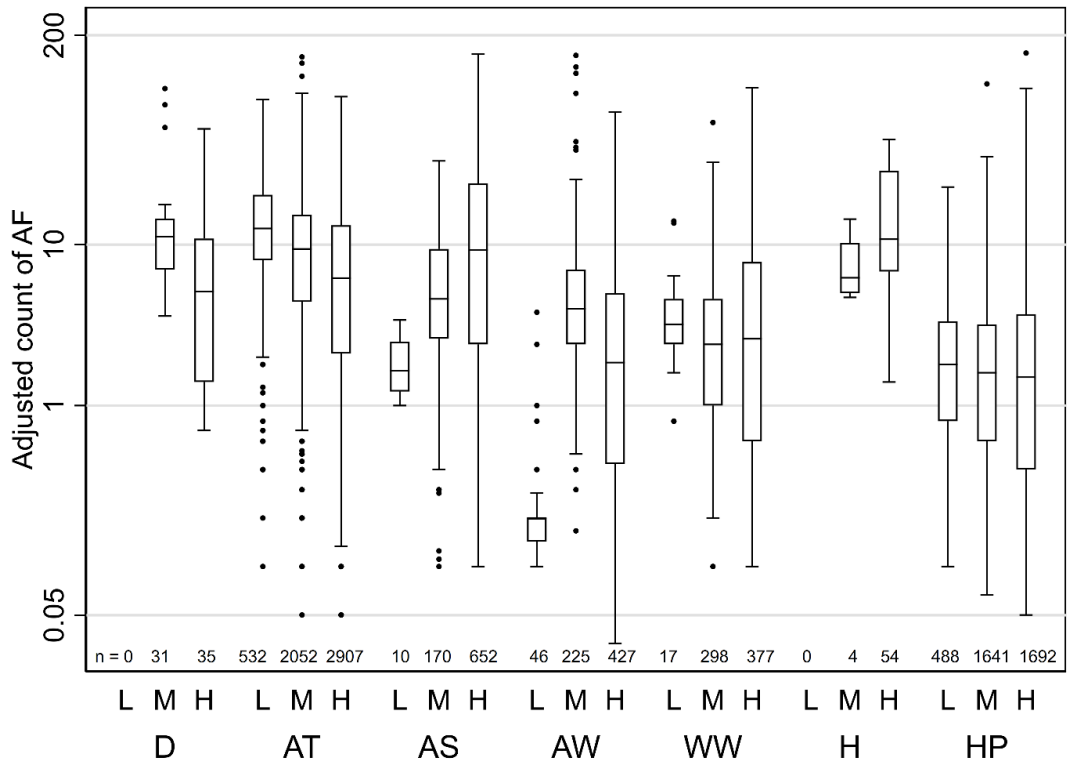

Figure 3.2. Boxplots of the mean normalized count of adult female (AF) *Lepeophtheirus salmonis* sea lice after treatment (AF count divided by the number of fish counted and offset by 1) included in the final multivariable linear mixed model with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. Counts were categorized by treatment (D: deltamethrin, AT: azamethiphos tarpaulin, AS: azamethiphos skirt, AW: azamethiphos wellboat, WW: warm water, H: Hydrolicer®, and HP: hydrogen peroxide) and temperature profiles (L: 3-7 °C, M: 8-11 °C, and H: 12-16 °C). n = 11,658.

Treatments were applied differently within the study area. Deltamethrin (Alpha Max®, Zoetis, USA) was applied in the BMA 1 and 2a and only prior to 2012 (when it was available for emergency use). The Hydrolicer® (Hydrolicer Production AS, Norway) was utilized in the BMA 3A and 3B. Azamethiphos, warm water and hydrogen peroxide treatments were applied in most of the BMAs at some point during the study period. The choice of treatments varied by temperature, where deltamethrin and the Hydrolicer® treatments were applied at

higher temperatures (range 8.8-15.1 °C and 11.2-15.6 °C, respectively) compared to other treatments (range 1.8-16.2 °C). The mean normalized count of AF sea lice after hydrogen peroxide treatments at each temperature were lower than the mean normalized count of AF sea lice for other treatments.

Table 3.3. Descriptive statistics for the variables potentially predicting the outcome variable of the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice. Results of the univariable linear mixed model are summarized by the Akaike's information criterion (AIC; null model AIC = 32,486). SD: standard deviation; IIP: internal infestation pressure; PAAM: pre-adult and adult male sea lice; EIP: external infestation pressure; n = 11,658.

Variable	Variable description	Mean (SD)	Mean (SD) abundance of AF sea lice after treatment	AIC
IIP_{AF}	Temporal weighted average abundance of AF sea lice with 21 day time lag	1.15 (1.34)		31,511
IIP_{PAAM}	Temporal weighted average abundance of PAAM with 28 day time lag	1.96 (1.20)		32,094
EIP_{AF}	Temporal and KDE spatial weighted average abundance of AF sea lice with 21 day time lag	0.28 (0.36)		31,852
$Baseline_{AF}$	Abundance of AF sea lice within and including 7 days prior to topical treatment	1.84 (1.44)		31,327
Temperature	Sea surface temperature from UKMO OSTIA product (°C)	11.34 (2.49)		32,374
Treatment	Topical treatment that was applied at the start of a segment			32,479
Azamethiphos Tarpaulin			1.90 (1.29)	

Table 3.3 continued next page.

Table 3.3 continued.

Variable	Variable description	Mean (SD)	Mean (SD) abundance of AF sea lice after treatment	AIC
Treatment	Temporal weighted average abundance of AF sea lice with 21 day time lag	1.15 (1.34)		31,511
Azamethiphos Skirt			1.51 (1.64)	
Azamethiphos Wellboat			0.84 (1.69)	
Deltamethrin			1.80 (1.07)	
Warm water			1.31 (1.27)	
Hydrolicer®			2.43 (0.94)	
Hydrogen peroxide			0.78 (1.39)	
In-feed treatment	Emamectin benzoate or avermectin was applied to a cage prior to the start of a segment			32,483
No			1.18 (1.67)	
Yes			1.20 (1.43)	

The application of an in-feed treatment prior to a segment occurred for most of the modelled segments (n_{seg} = 2,053). The mean abundance of AF sea lice after topical treatment was not significantly different following the application of in-feed treatment (t = -0.66, p = 0.509).

3.4.2 Final model

The final multilevel mixed-effects linear regression model included the variables IIP_{AF}, IIP_{PAAM}, EIP_{AF} weighted with a KDE bandwidth of 10 km, $baseline_{AF}$, BMAs, temperature and treatments, and their interaction (Table 3.4). It had site-cycle, cage-cycle, and segment random effects with an exponential correlation structure to the errors, stratified by three temperature profiles. The in-feed treatment dichotomous variable was non-significant (coefficient = -0.092, p = 0.179) and was not retained in the final model. No violations of model assumptions of linearity, homoscedasticity and normality were observed.

Table 3.4. Multivariable linear mixed model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice; n = 11,658.

Variable	Coefficient	95 % Confidence interval	p-value
IIP_{AF}	0.188	0.156, 0.219	< 0.001
IIP_{PAAM}	0.095	0.069, 0.121	< 0.001
EIP_{AF}	0.478	0.374, 0.581	< 0.001
$Baseline_{AF}$	0.287	0.254, 0.320	< 0.001
Temperature (°C)	-0.111	-0.126, -0.096	< 0.001
Treatment			< 0.001
Azamethiphos Tarpaulin	baseline		
Azamethiphos Skirt	-1.966	-2.624, -1.307	
Azamethiphos Wellboat	-1.492	-2.066, -0.919	
Deltamethrin	-0.953	-3.573, 1.668	
Warm water	-3.074	-3.630, -2.517	
Hydrolicer®	0.932	-3.564, 5.427	

Table 3.4 continued next page.

Table 3.4 continued.

Variable	Coefficient	95 % Confidence interval	p-value
Treatment			
Hydrogen peroxide	0.188	0.156, 0.219	< 0.001
Treatment-Temperature interaction	0.095	0.069, 0.121	< 0.001
Azamethiphos Tarpaulin*Temp			
Azamethiphos Skirt*Temp			
Azamethiphos Wellboat*Temp			
Deltamethrin*Temp			
Hydrogen peroxide			
Treatment-Temperature interaction			
Warm water*Temp	0.222	0.176, 0.267	
Hydrolicer®*Temp	-0.020	-0.363, 0.323	
Hydrogen peroxide*Temp	0.154	0.131, 0.176	

Table 3.4 continued next page.

Table 3.4 continued.

Variable	Coefficient	95 % Confidence interval	p-value
BMA			0.108
1	baseline		
2A	-0.015	-0.230, 0.200	
2B	-0.269	-0.571, 0.033	
3A	0.269	-0.043, 0.581	
3B	-0.005	-0.257, 0.247	
3C	-0.494	-1.196, 0.208	
Constant	1.521	1.284, 1.758	

3.4.3 Fixed effects

3.4.3.1 Internal and external infestation pressures

The IIP_{AF} and IIP_{PAAM} were significant predictors in the final model (both $p < 0.001$). An increase of the IIP_{AF} by one unit affected the abundance of AF sea lice by a factor of 1.207 (i.e., a relative increase of 20.7 %) when other predictors remained constant. Similarly, a one unit increase in the IIP_{PAAM} resulted in a relative increase of the abundance of AF sea lice by 11.0 %.

The EIP_{AF} with a KDE bandwidth of 10 km was a significant predictor for the outcome ($p < 0.001$). The predictor remained significant at $p < 0.001$ for KDE bandwidths of 5 to 60 km. The coefficient of the EIP_{AF} had a bimodal distribution with increasing KDE bandwidths; a peak occurred at the 13 km bandwidth (Figure 3.3 [range for 40-60 km not displayed in graph]). The estimated coefficient was smallest at a bandwidth of 5 km and had a second nadir at 25 km. The model fit with the EIP_{AF} weighted with a KDE with a 10 km bandwidth had the lowest AIC. The AIC decreased with KDE bandwidths of 5 to 9 km with a range of ΔAIC of 0.4 to 24.8. The ΔAIC increased to 51.0 at 40 km. The abundance of AF sea lice increased by 61.2 % for every increase of one unit of the EIP_{AF} 10 km predictor when other predictors remained constant.

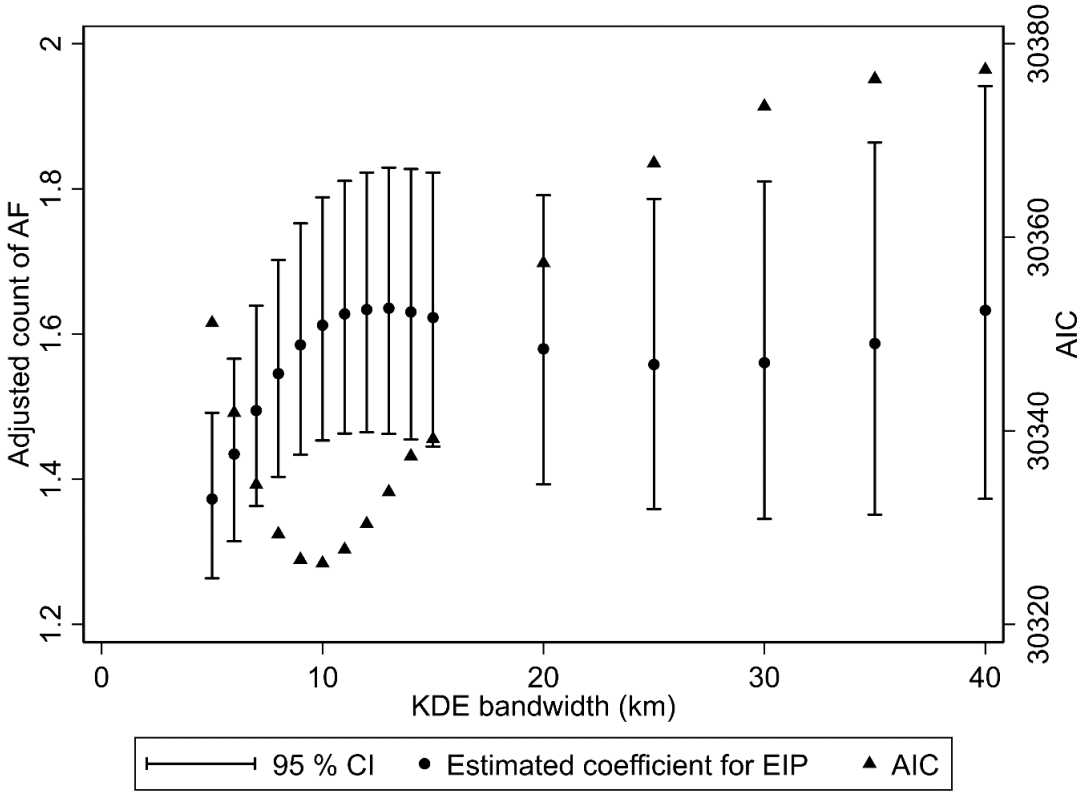

Figure 3.3. Estimated exponentiated coefficient and 95 % confidence interval (CI) for the external infestation pressure (EIP) predictor weighted with a Gaussian kernel density estimate (KDE) and the Akaike's information criterion (AIC) reported from the multivariable linear mixed model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice where the bandwidth of the KDE was increased from 5 to 40 km. Scale is the mean normalized count of AF sea lice after treatment (AF count divided by the number of fish counted and offset by 1).

3.4.3.2 Baseline abundances of AF sea lice

The $baseline_{AF}$ was a significant predictor in the final model ($p < 0.001$). A one unit increase in the $baseline_{AF}$ resulted in an increase of 33.3 % of the AF sea lice abundance when other predictors remained constant.

3.4.3.4 Temperature, treatments, and their interaction

Treatments and the treatment-temperature interaction were significant predictors in the final model (Wald Chi square tests, $p < 0.001$). A quadratic regression and polynomial regression of temperature with default powers and two dimensions did not provide an improvement to the linear treatment-temperature interaction. The association between the abundance of AF sea lice and temperature was linear for most treatments; the azamethiphos applied by skirt and warm water by wellboat treatments had a slight curve, while the azamethiphos applied by tarpaulin had a stronger curving trend line.

The abundance of AF sea lice decreased with increasing temperature for the treatments deltamethrin, and azamethiphos applied by tarpaulin and wellboat as demonstrated in Figure 3.4 using median values for the average abundances of AF sea lice in BMA 1. With the same reference values, the estimated abundance of AF sea lice increased with increasing temperature for the treatments azamethiphos applied by skirt, warm water, and hydrogen peroxide.

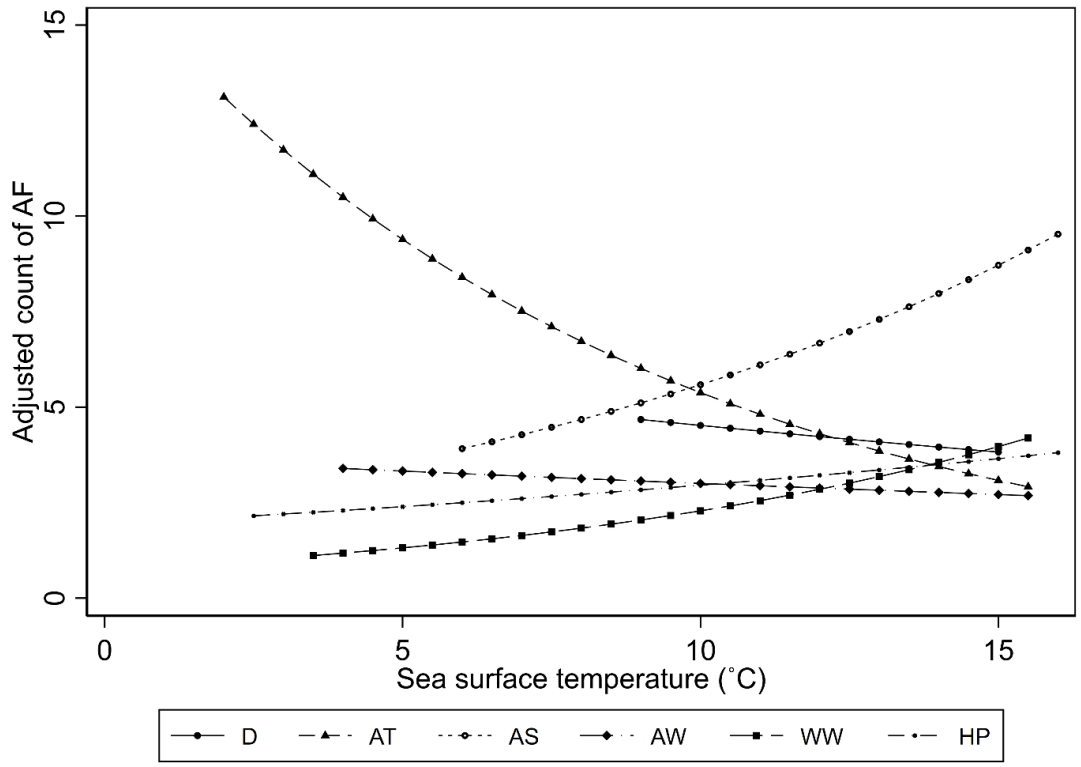

Figure 3.4. Estimated mean normalized count of adult female (AF) *Lepeophtheirus salmonis* sea lice (AF count divided by the number of fish counted and offset by 1) after treatments applied in the Bay Management Area (BMA) 1 against sea surface temperature at median values of the continuous predictors of the final multivariable linear mixed model with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. D: deltamethrin, AT: azamethiphos tarpaulin, AS: azamethiphos skirt, AW: azamethiphos wellboat, WW: warm water, and HP: hydrogen peroxide. The Hydrolicer® was not applied in BMA 1 between 2009 and 2018.

At the median temperature (11.7 °C), deltamethrin, azamethiphos by wellboat, warm water, and hydrogen peroxide treatments were estimated to decrease AF sea lice abundances by 4.0 %, 34.8 %, 38.0 %, and 28.5 %, respectively (data not shown; snapshot of data from Figure 3.4). The application of azamethiphos by skirt resulted in an increase of the abundance of AF sea lice by 46.2 % at the median temperature.

3.4.3.5 Bay Management Areas

The BMAs were non-significant in the final model (Wald Chi square test, p = 0.108). The univariable linear mixed model with the BMAs produced an AIC of 32,478, which was slightly smaller than the null model AIC of 32,486. The BMAs were retained as a fixed effect in the final model as a potential cofounder.

3.4.5 Random effects

The site-cycle random effect had a variance of 0.14 (95 % confidence interval [CI] 0.10-0.21). The variances of the cage-cycle and segment random effects were near zero. The residual variances accounted for 80.0 to 91.6 % of the total variance in the final model. The intraclass correlation coefficient (ICC) for 2 observations in the same site-cycle was between 0.09 and 0.20. The autocorrelation of the errors was between 0.92 and 0.94 per day difference.

3.5 Discussion

3.5.1 Contributions of internal and external infestation pressure

This study had the objective to estimate the impact of the infestation pressures on the abundances of sea lice for the Atlantic salmon aquaculture sites of the Bay of Fundy, NB. The estimated infestation pressures had a significant impact on the abundance of AF for the NB aquaculture sites.

The model estimated the EIP had a dispersal distance of AF abundance of 10 km for the NB aquaculture sites using empirical evidence. This displacement distance is near the extrapolated 8 km proposed by Nelson et al. (2017) based on approximate mean flow rates of 10 cm/s around sites. This result was similar

to previously reported estimates from Norway (Aldrin et al., 2013), Scotland (Murray and Gillibrand, 2006; Gillibrand and Willis, 2007; Amundrud and Murray, 2009), and Grand Manan (Elghafghuf et al., 2020). Other studies estimated larger dispersal distances between 20 and 30 km (Krkošek et al., 2005; Asplin et al., 2011, 2014; Kristoffersen et al., 2013; Salama et al., 2013, 2016, Aldrin et al., 2019). The variability in the estimate of the EIP is likely a response to differing environments and modelling approaches. Water current characteristics, for example, are drastically different in Scotland (Murray et al., 2011) and NB, and likely have a large effect on the dispersal distance estimated. Modelling approaches likely also affect the estimate of the EIP; Aldrin et al. (2013, 2019) estimated 10 km and 20-30 km for two Norwegian rivers using different models with data from the Norwegian aquaculture industry.

The infestation pressure within sites (standardized estimated coefficient IIP_{AF}: 0.165, 95 % CI 0.137-0.192) was an important predictor in the model. Our estimated IIP was larger than the estimate of zero for Grand Manan produced in the state space model by Elghafghuf et al. (2020). This difference in IIP may be due to the transition matrix of the state space model accounting for the development of PAAM to AF.

The IIP_{PAAM} standardized coefficient (0.075, 95 % CI 0.054-0.096) was smaller than the other two estimated infestation pressures. This suggested the count of PAAM sea lice is not as important in estimating the abundance of AF sea lice. One reason may be that PAAM sea lice is a combination of adult males, and pre-adult that will mature into female and male in an approximate 1:1 ratio (Hamre et

al., 2019).

The standardized estimated coefficient of the EIP_{AF} (0.114, 95 % CI 0.089-0.138) was smaller than the standardized coefficient of the IIP_{AF}, but not the IIP_{PAAM}. The larger impact of the IIP compared to the EIP in this model may be associated with the length of time necessary to complete topical treatments at a site. Nelson et al. (2017) captured planktonic lice at higher concentrations near farms and posited sites amplify sea lice densities, and planktonic lice are rapidly displaced from sites, but concentrate in proximity to the originating sites. Aldrin et al. (2013) estimated the IIP of lice abundance was larger than the EIP. Based on hydrodynamic models, Adams et al. (2012) noted a higher rate of self-infestation compared to external infestation and concluded self-infestation may result in positive feedback, meaning that the population of sea lice on the site would increase over time because of the reproductive output of AF sea lice. Conversely, EIP was greater than IIP for Grand Manan (Elghafghuf et al., 2020) and Chile (Kristoffersen et al., 2013). These authors had multiple hypotheses for the larger effect of the EIP compared to the IIP: asynchronous treatments, better treatment effectiveness against adults compared to the juvenile life stages (Elghafghuf et al., 2018, 2020), flushing out of the planktonic life stages prior to the infestation of that site, and improved management of sea lice infestation at their site compared to the infestation at their neighbours' sites. The methodology for estimation of IIP, EIP, environmental characteristics, sea lice densities and management, and aquaculture industry may all contribute to the differences in the effects of the EIP compared to the IIP.

Another source of variation in the estimates of infestation pressures are the lag times. In this study, the infestation pressures were defined by fixed lag times of 21 (AF) and 28 days (PAAM) to produce the outcome of the abundance of AF. Elghafghuf et al. (2020) found that a fixed lag of three weeks produced the best model fit for the outcome of the abundance of PAAM. In other studies (e.g., Aldrin et al., 2017, 2019), lags of approximately three to four weeks resulted in infestation at neighbouring sites. This lag suggests planktonic lice removed from a site by currents disperse at a low concentration (Nelson et al., 2017). During the approximate three to four weeks dispersal, the nauplii (I and II) lice develop into the copepodid prior to the encounter with a neighbouring site. The effect of lag time on the infestation pressures is unknown and should be evaluated in future sensitivity studies.

The standardized coefficient of the $baseline_{AF}$ (0.272, 95% CI 0.241-0.304) was larger than that of the infestation pressures. This was a surprising finding and suggests the infestation pressure within sites is more important for mitigation of sea lice compared to the infestation pressure originating from other sites. We hypothesize the effect is larger than the estimated infestation pressures because the $baseline_{AF}$ is the abundance prior to treatment and not an average of abundances over a 15 day window as for the IIP_{AF}, IIP_{PAAM}, and EIP_{AF}.

These estimates of the internal and external infestation pressures for the Bay of Fundy, NB, have implications for the management of sites. The larger IIP compared to EIP suggests that improvement of mitigation strategies for within site control of sea lice has the potential to decrease the IIP which contributes to

maintaining the infestation within a site (Aldrin et al., 2013). The EIP of 10 km suggests that neighbouring sites within 10 km should apply synchronous treatments. This implies that the majority of sites in the Bay of Fundy, NB, need to have the common goal for sea lice treatment timing because more than 75 % of sites had less than 1 km seaway distance from their nearest neighbour (Chang et al., 2011). The limited number of wellboats and tarpaulin systems presents a logistical constraint to synchronizing the treatments of entire sites in tandem; in-feed sea lice treatments are a potential method for the synchronized treatment of sea lice at multiple sites that avoids the requirement for additional equipment purchases. The EIP also has implications for the stocking of sites after fallowing and shortly after treatments because under these scenarios the sites have low to no abundances of sea lice (Aldrin et al., 2013; Kristoffersen et al., 2013, 2014).

3.5.2 Treatments and temperature

Temperature had a significant effect on sea lice abundance. Temperature is an important controlling factor for sea lice as demonstrated under laboratory (Hamre et al., 2019), simulation and modelling (Revie et al., 2005; Stien et al., 2005; Stucchi et al., 2011), and field conditions (Samsing et al., 2016). The time spent in each life stage was observed to decrease with increasing temperature (Stien et al., 2005; Samsing et al., 2016). Samsing et al. (2016) observed that the proportion of copepodids that successfully attached out of the total number of copepodids in the suspension (infestation success) was highest at 10 °C compared to 5 °C and 20 °C.

The effects of temperature on the outcome varied by treatment. There are

multiple contributing factors to this association: the treatment dosage and exposure time, method of application, and efficacy. Sea lice treatments are prescribed within an acceptable range of dosage and exposure time (Burridge et al., 2010). For example, hydrogen peroxide prescription guidelines are for application between 8 and 13 °C (Overton et al., 2017, 2019); at low temperatures there is poor treatment efficacy (Treasurer et al., 2000) and at elevated temperatures there is potential for high salmon mortality (Thomassen, 1993). Between 8 and 13 °C, the dosage of hydrogen peroxide needed to achieve similar efficacy decreases with increasing water temperature (Overton et al., 2017, 2019).

We found that the effect of hydrogen peroxide treatments was diminished with increasing temperature. Perhaps the exposure time was shortened for the application of hydrogen peroxide treatments at higher water temperatures, resulting in decreased activity time on the sea lice and thus reduced effectiveness of this treatment. An alternative, proposed by Overton et al. (2017), was to apply hydrogen peroxide treatment to salmon pumped from cages into the wellboat that contains cold seawater chilled on the boat or sourced from greater depths or a land-based facility.

The abundance of AF sea lice was decreased after warm water treatment, but this effect was diminished with increasing temperatures. This finding may be due to the acclimation of the sea louse to temperature change stress. Under laboratory settings, nauplii were demonstrated to adapt to a temperature rise of 20 °C after acclimation at 10 °C (Borchel et al., 2018). The ability to survive a

drastic rise in temperature was dependent on the acclimation temperature as demonstrated with the water flea, *Daphnia pulex* (Leydig; Williams et al., 2012).

The method of application influenced the treatment response on the abundance of AF sea lice. Azamethiphos by skirt resulted in an increase of the abundance of AF sea lice as compared to the tarpaulin and wellboat methods of application for all temperatures. We suggest that the skirt provided poor control of the treatment compared to the other 2 methods of application. The skirt application method involved enclosing only the sides of a cage while keeping the bottom open and stopped being used in the region by 2013. The tarpaulin and wellboat methods provide complete enclosure of the water used for treatment, and since 2013 have been the only available methods to administer azamethiphos.

The predictor of in-feed treatment applied prior to a segment was non-significant in the final model. We anticipate the effect was minimal because in-feed treatments were frequently applied, and there may be resistance to the in-feed treatments (e.g., Jones et al., 2013; Whyte et al., 2013) reducing the impact of the variable in the model. Additionally, this predictor was particularly prone to information bias, where treatment durations may not have overlapped very well with segment lengths. Specifically, segments may have been mislabeled if the duration of the application of in-feed treatment extended only a few days into the segment and the half-life of the chemotherapeutic agent was shorter than the length of the segment. In-feed treatments do not have an immediate sea lice response and have a much longer duration of impact making them more difficult to assess independently of the other factors. Proper evaluation of in-feed

treatments would require a different modelling approach than was used in this study.

3.5.3 Hierarchical levels

The BMA are a unique example of zonal health management and were retained in the model despite being a non-significant predictor. This finding may imply that the BMA boundaries are not optimally effective for the management of *L. salmonis*; importantly, the BMA were designed for the management of ISAv and not specifically for sea lice (Chang et al., 2011).

Most of the variance in the abundance of AF sea lice occurred at the residual level; the variance was strongly temperature dependent. The estimated ICC for cage-cycle were much lower than the ICC estimated from another study in our study area. Elmoslemany et al. (2013) reported clustering of cages within sites with ICC values of 0.60 for AF and 0.78 for PAAM sea lice counts reported in 2011. This discrepancy may be due to the exclusion of treatments and the use of segments. Elmoslemany et al. (2013) excluded treatments as a variable in the analyses; this would increase the variability explained at the cage level. Our analysis included segments as a hierarchical level within cage-cycles. The inclusion of segments would reduce cage level clustering.

3.5.4 Study design and limitations

3.5.4.1 Study design

This was a retrospective cohort study that utilized complex historical data with integral hierarchical structures. The linear mixed model was selected because it

is well adapted to these types of data. The model allowed for estimation of infestation pressures in the Bay of Fundy, NB, using the Fish-iTrends database.

The EIP were estimated from seaway distances between sites. Ideally, the dispersion of sea lice that contribute to the estimated EIP should be validated using sentinel cages (Salama et al., 2013; Sandvik et al., 2016). However, there may be discrepancies between abundances of sea lice measured at sites and from sentinel cages (Salama et al., 2018). Alternatively, investigating the genetic parentage of individual sea lice could improve the knowledge of lice dispersion locally (Cantrell et al., 2020).

The EIP can also be estimated from hydrodynamic models. Therefore, future research with these data will estimate the EIP using an updated hydrodynamic model for the Bay of Fundy, NB. Multiple estimates of the EIP will be generated for different scenarios such as storm events with high winds and annual freshwater input secondary to snow and ice melting (i.e., spring freshet) using methods similar to Filgueira et al. (2012). The EIP will be estimated from matrices of averaged volumetric water exchange for each link of the finite element grid of the hydrodynamic model (Filgueira et al., 2012).

3.5.4.2 Limitations

The final model estimated the coefficients for the IIP and EIP that are averages in time and space. The infestation pressures were estimated for many of the sites in the Bay of Fundy, NB, for the years 2009 to 2018. The application of different methods to the dataset may reveal variability in the impact of infestation

pressures estimated (e.g., Kristoffersen et al., 2014; Cantrell et al., 2019).

The estimates of infestation pressure from this study demonstrated the potential for larval infestation. The fixed lag approach to the estimates of infestation pressure means that the biological effects of temperature cannot be separated from salinity. We know from previous studies that low salinity increases sea lice mortality rates and higher temperatures increase sea lice development rates (Stien et al., 2005; Groner et al., 2016). Due to high tidal exchanges, salinity in this area of the Bay of Fundy, NB, does not vary greatly and remains near the optimal salinity (30 ppt; Chang et al., 2011; Page et al., 2015) for sea lice development and infestation (Bricknell et al., 2006; Arriagada et al., 2016). Therefore, a quantitative life cycle approach could be applied (Kristoffersen et al., 2014) to evaluate the rates of development and mortality in light of temperature variation in our study area. However, Elghafghuf et al. (2020) found that the quantitative life cycle approach did not perform better compared to the fixed lag approach.

The fixed lag approach can be utilized to evaluate the effects of temperature variation, and other environmental parameters such as weather and wind when combined with hydrodynamic modelling. Hydrodynamic models will inform the estimates of the EIP and these effects will be evaluated in future studies. The model was limited to the available data in Fish-iTrends. There were no data from the neighbouring Maine Atlantic salmon aquaculture sites in the Bay of Fundy. There were no records of the spatial and temporal distribution of cages within sites, and the number of salmon stocked in cages. A sensitivity analysis

evaluated a range of 270 to 840 thousand stocked fish and found little change in the estimated coefficients for the IIP and EIP (Elghafghuf et al., 2020). Kristoffersen et al. (2014) concluded that the EIP was proportional to the abundance of salmon at neighbouring sites. Therefore, the Fish-iTrends database would improve its usefulness for infestation pressure investigations if stocking densities and fish biomass were included.

This model cannot characterize the movement of sea lice in and out of sites (influx and outflux) as performed by other researchers (Adams et al., 2012; Cantrell et al., 2018). Data on the interactions between sites as performed with hydrodynamic connectivity studies may provide useful information in conjunction to the findings in this study.

Inherent to this retrospective cohort study is potential for misclassification and selection bias, and measurement error. Misclassification bias would occur when site staff counters recorded no sea lice (n_{obs} = 733) when the fish was infested. Measurement errors would occur due to the variability of site staff counter experience (Elmoslemany et al., 2013). Measurement error may be most likely to occur with larger numbers of sea lice on a fish (Aldrin et al., 2013). Measurement error may be present in the UKMO sea surface temperature remote sensing product as proximity to land may result in temperature contamination of the temperature at low resolutions. There is also the possibility for selection bias due to non-random sampling of salmon (Gautam et al., 2016b) and the use of the $baseline_{AF}$ predictor in the model. The $baseline_{AF}$ reduced the total number of observations and was evaluated further in a small sensitivity study (the additional

details found in Supplemental Information). We expect these biases and errors to be homogeneous across the study period and area. Furthermore, we expect the presence of any biases or errors in our model to move the effects toward the null hypothesis.

3.6 Conclusions

Our objective is to estimate the impact of the infestation pressures on the abundances of *L. salmonis* sea lice for the Bay of Fundy, NB. The abundance of AF sea lice was modelled using a multivariable linear mixed model to obtain estimates of the IIP and EIP using the Fish-iTrends database. The effect of the IIP and EIP on the abundances of AF were significant in the final model. The EIP with a Gaussian KDE bandwidth of 10 km had the lowest AIC and represents the dispersal distance for infestation of a neighbouring site, which is consistent with previous findings in regional and global areas with salmon aquaculture. Future research will focus on improving the estimate of the EIP with hydrodynamic data and evaluating the infestation pressures under different scenarios. We conclude from this study that site management of sea lice has its greatest impact on the site in question and synchronizing treatments between sites remains an important component of overall success in controlling sea lice at fish farms in Bay of Fundy.

3.7 References

Abolofia, J., Asche, F., and Wilen, J. E. (2017). The Cost of lice: Quantifying the impacts of parasitic sea lice on farmed salmon. Marine Resource Economics, 32, 329–349. https://doi.org/10.1086/691981

Adams, T., Black, K., Macintyre, C., Macintyre, I., and Dean, R. (2012). Connectivity modelling and network analysis of sea lice infection in Loch Fyne, west coast of Scotland. Aquaculture Environment Interactions, 3, 51–63. https://doi.org/10.3354/aei00052

Aldrin, M., Storvik, B., Kristoffersen, A. B., and Jansen, P. A. (2013). Space-time modelling of the spread of sea lice between and within Norwegian marine salmon farms. PLoS ONE, 8, 1–10. https://doi.org/10.1371/journal.pone.0064039

Aldrin, M., Huseby, R. B., Stien, A., Grøntvedt, R. N., Viljugrein, H., and Jansen, P. A. (2017). A stage-structured Bayesian hierarchical model for sea lice populations at individual salmon farms - Estimated from multiple farm data sets. Ecological Modelling, 359, 333–348.

Aldrin, M., Jansen, P. A., and Stryhn, H. (2019). A partly stage-structured model for the abundance of sea lice in salmonid farms. Epidemics, 26, 9–22. https://doi.org/10.1016/j.epidem.2018.08.001

Amundrud, T. L., and Murray, A. G. (2009). Modelling sea lice dispersion under varying environmental forcing in a Scottish sea loch. Journal of Fish Diseases, 32, 27–44. https://doi.org/10.1111/j.1365-2761.2008.00980.x

Arriagada, G., Vanderstichel, R., Stryhn, H., Milligan, B., and Revie, C. W. (2016). Evaluation of water salinity effects on the sea lice *Lepeophtheirus salmonis* found on farmed Atlantic salmon in Muchalat Inlet, British Columbia, Canada. Aquaculture, 464, 554–563. https://doi.org/10.1016/j.aquaculture.2016.08.002

Asplin, L., Boxaspen, K. K., and Sandvik, A. D. (2011). Modeling the Distribution and Abundance of Planktonic Larval Stages of *Lepeophtheirus salmonis* in Norway. In S. Jones and R. Beamish (Eds.), Sea lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Asplin, L., Johnsen, I. A., Sandvik, A. D., Albretsen, J., Sundfjord, V., Aure, J., and Boxaspen, K. K. (2014). Dispersion of sea lice in the Hardangerfjord. Marine Biology Research, 10, 216–225. https://doi.org/10.1080/17451000.2013.810755

Barker, S. E., Bricknell, I. R., Covello, J., Purcell, S. L., Fast, M. D., Wolters, W., and Bouchard, D. A. (2019). Sea lice, *Lepeophtheirus salmonis* (Krøyer 1837), infected Atlantic salmon (*Salmo salar* L.) are more susceptible to infectious salmon anemia virus. PLoS ONE, 14(1), 1–28. https://doi.org/10.1371/journal.pone.0209178

Borchel, A., Komisarczuk, A. Z., Rebl, A., Goldammer, T., and Nilsen, F. (2018). Systematic identification and characterization of stress-inducible heat shock proteins (HSPs) in the sea louse (*Lepeophtheirus salmonis*). Cell Stress & Chaperones, 23, 127–139. https://doi.org/10.1007/s12192-017-0830-9

Bricknell, I. R., Dalesman, S. J., O'Shea, B., Pert, C. C., and Luntz, A. J. M. (2006). Effect of environmental salinity on sea lice *Lepeophtheirus salmonis* settlement success. Diseases of Aquatic Organisms, 71, 201–212. https://doi.org/10.3354/dao071201

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. https://doi.org/10.1016/j.aquaculture.2010.05.020

Cantrell, D., Filgueira, R., Revie, C. W., Rees, E., Vanderstichel, R., Foreman, M. G. G., … Grant, J. (2019). The relevance of larval biology on spatiotemporal patterns of pathogen connectivity among open-marine salmon farms. Canadian Journal of Fisheries and Aquatic Sciences, 77, 505–519. https://doi.org/10.1139/cjfas-2019-0040

Cantrell, D. L., Groner, M. L., Ben-Horin, T., Grant, J., and Revie, C. W. (2020). Modeling pathogen dispersal in marine fish and shellfish. Trends in Parasitology, 36, 239–249. https://doi.org/10.1016/j.pt.2019.12.013

Cantrell, D. L., Rees, E. E., Vanderstichel, R., Grant, J., Filgueira, R., and Revie, C. W. (2018). The use of kernel density estimation with a biophysical model provides a method to quantify connectivity among salmon farms: Spatial planning and management with epidemiological relevance. Frontiers in Veterinary Science, 5, 1–14. https://doi.org/10.3389/fvets.2018.00269

Chang, B. D., Page, F. H., Beattie, M. J., and Hill, B. W. H. (2011). Sea Louse Abundance on Farmed Salmon in the Southwestern New Brunswick Area of the Bay of Fundy. In S. Jones and R. Beamish (Eds.), Sea lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Costello, M. J. (2006). Ecology of sea lice parasitic on farmed and wild fish. Trends in Parasitology, 22, 475–483. https://doi.org/10.1016/j.pt.2006.08.006

Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). *Bay of Fundy Marine Aquaculture Sites* [map]. 1:125000. New Brunswick, Canada.

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

Elghafghuf, A., Vanderstichel, R., St-Hilaire, S., and Stryhn, H. (2018). Using state-space models to predict the abundance of juvenile and adult sea lice on Atlantic salmon. Epidemics, 24, 76–87. https://doi.org/10.1016/j.epidem.2018.04.002

Elmoslemany, A., Whyte, S. K., Revie, C. W., and Hammell, K. L. (2013). Sea lice monitoring on Atlantic salmon farms in New Brunswick, Canada: Comparing audit and farm staff counts. Journal of Fish Diseases, 36, 241–247. https://doi.org/10.1111/jfd.12051

Fast, M. D. (2014). Fish immune responses to parasitic copepod (namely sea lice) infection. Developmental and Comparative Immunology, 43, 300–312. https://doi.org/10.1016/j.dci.2013.08.019

Filgueira, R., Grant, J., Bacher, C., and Carreau, M. (2012). A physical-biogeochemical coupling scheme for modeling marine coastal ecosystems. Ecological Informatics, 7, 71–80. https://doi.org/10.1016/j.ecoinf.2011.11.007

Gautam, R., Boerlage, A. S., Vanderstichel, R., Revie, C. W., and Hammell, K. L. (2016). Variation in pre-treatment count lead time and its effect on baseline estimates of cage level sea lice abundance. Journal of Fish Diseases, 39, 1297–1303. https://doi.org/10.1111/jfd.12460

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017a). Effect of timing of count events on estimates of sea lice abundance and interpretation of effectiveness following topical treatments. Journal of Fish Diseases, 40, 367–375. https://doi.org/10.1111/jfd.12519

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017b). Evaluating topical treatment effectiveness in the control of sea lice burdens on Atlantic salmon in New Brunswick, Canada. Journal of Fish Diseases, 40, 895–905. https://doi.org/10.1111/jfd.12569

Gillibrand, P. A., and Willis, K. J. (2007). Dispersal of sea louse larvae from salmon farms: Modelling the influence of environmental conditions and larval behaviour. Aquatic Biology, 1, 63–75. https://doi.org/10.3354/ab00006

Groner, M. L., McEwan, G. F., Rees, E. E., Gettinby, G., and Revie, C. W. (2016). Quantifying the influence of salinity and temperature on the population dynamics of a marine ectoparasite. Canadian Journal of Fisheries and Aquatic Sciences, 73, 1281–1291. https://doi.org/10.1139/cjfas-2015-0444

Hamre, L., Bui, S., Oppedal, F., Skern-Mauritzen, R., and Dalvin, S. (2019). Development of the sea louse *Lepeophtheirus salmonis* parasitic stages in temperatures ranging from 3 to 24°C. Aquaculture Environment Interactions, 11, 429–443. https://doi.org/10.3354/aei00320

Hamre, L. A., Eichner, C., Caipang, C. M. A., Dalvin, S. T., Bron, J. E., Nilsen, F., … Skern-Mauritzen, R. (2013). The sea louse *Lepeophtheirus salmonis*

(Copepoda: Caligidae) life cycle has only two chalimus stages. PLoS ONE, 8(9), 1–9. https://doi.org/10.1371/journal.pone.0073539

Hijmans, R.J., van Etten, J., Sumner, M., Cheng, J., Baston, D., Bevan, A., … Wueest, R. (2019). raster: Geographic Data Analysis and Modeling. R package version 3.0-12. https://CRAN.R-project.org/package=raster

Hogans, W. E. (1995). Infection dynamics of sea lice, *Lepeophtheirus salmonis* (Copepoda: Caligidae) parasitic on Atlantic salmon (*Salmo salar*) cultured in marine waters of the lower Bay of Fundy (Vol. 2067). Retrieved from http://search.proquest.com/docview/16997923?accountid=27308

Igboeli, O. O., Fast, M. D., Heumann, J., and Burka, J. F. (2012). Role of P-glycoprotein in emamectin benzoate (SLICE®) resistance in sea lice, *Lepeophtheirus salmonis*. Aquaculture, 344–349, 40–47. https://doi.org/10.1016/j.aquaculture.2012.03.026

Johnson, S. C., and Albright, L. J. (1991). The developmental stages of *Lepeophtheirus salmonis* (Krøyer, 1837) (Copepoda: Caligidae). Canadian Journal of Zoology, 69(4), 929–950. https://doi.org/10.1139/z91-138

Johnson, S. C., Treasurer, J. W., Bravo, S., and Nagasawa, K. (2004). A review of the impact of parasitic copepods on marine aquaculture. Zoological Studies, 43, 229–243.

Jones, P. G., Hammell, K. L., Dohoo, I. R., and Revie, C. W. (2012). Effectiveness of emamectin benzoate for treatment of *Lepeophtheirus salmonis* on farmed Atlantic salmon *Salmo salar* in the Bay of Fundy, Canada. Diseases of Aquatic Organisms, 102, 53–64. https://doi.org/10.3354/dao02517

Jones, P. G., Hammell, K. L., Gettinby, G., and Revie, C. W. (2013). Detection of emamectin benzoate tolerance emergence in different life stages of sea lice, *Lepeophtheirus salmonis*, on farmed Atlantic salmon, *Salmo salar* L. Journal of Fish Diseases, 36(3), 209–220. https://doi.org/10.1111/jfd.12022

Kristoffersen, A. B., Rees, E. E., Stryhn, H., Ibarra, R., Campisto, J. L., Revie, C. W., and St-Hilaire, S. (2013). Understanding sources of sea lice for salmon farms in Chile. Preventive Veterinary Medicine, 111, 165–175. https://doi.org/10.1016/j.prevetmed.2013.03.015

Kristoffersen, A. B., Jimenez, D., Viljugrein, H., Grøntvedt, R., Stien, A., and Jansen, P. A. (2014). Large scale modelling of sea lice (*Lepeophtheirus salmonis*) infection pressure based on lice monitoring data from Norwegian salmonid farms. Epidemics, 9, 31–39. https://doi.org/10.1016/j.epidem.2014.09.007

Kristoffersen, A. B., Qviller, L., Helgesen, K. O., Vollset, K. W., Viljugrein, H., and Jansen, P. A. (2018). Quantitative risk assessment of sea louse-induced

mortality of seaward-migrating post-smolt Atlantic salmon. Epidemics, 23, 19–33. https://doi.org/10.1016/j.epidem.2017.11.001

Krkošek, M., Lewis, M. A., and Volpe, J. P. (2005). Transmission dynamics of parasitic sea lice from farm to wild salmon. Proceedings of the Royal Society B: Biological Sciences, 272, 689–696. https://doi.org/10.1098/rspb.2004.3027

Murray, A. G., Amundrud, T. L., Penston, M. J., Pert, C. C., and Middlemas, S. J. (2011). Abundance and Distribution of Larval Sea Lice in Scottish Coastal Waters. In S. Jones and R. Beamish (Eds.), Sea lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Murray, A. G., and Gillibrand, P. A. (2006). Modelling sea lice dispersal in Loch Torridon, Scotland. Marine Pollution Bulletin, 53, 128–135. https://doi.org/10.1016/j.marpolbul.2005.09.013

Nelson, E. J., Robinson, S. M. C., Feindel, N., Sterling, A., Byrne, A., and Pee Ang, K. (2017). Horizontal and vertical distribution of sea lice larvae (*Lepeophtheirus salmonis*) in and around salmon farms in the Bay of Fundy, Canada. Journal of Fish Diseases, 41, 885–899. https://doi.org/10.1111/jfd.12692

Nylund, A., Wallace, C., and Hovland, T. (1993). The possible role of *Lepeophtheirus salmonis* (Krøyer) in the transmission of infectious salmon anaemia. In G. A. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (pp. 367–373). Chichester, West Sussex: Ellis Horwood.

Nylund, A., Hovland, T., Hodneland, K., Nilsen, F., and Lovik, P. (1994). Mechanisms for transmission of infectious salmon anaemia (ISA). Diseases of Aquatic Organisms, 19, 95–100. https://doi.org/10.3354/dao019095

Overton, K., Dempster, T., Oppedal, F., Kristiansen, T. S., Gismervik, K., and Stien, L. H. (2019). Sea lice treatments and salmon mortality in Norwegian aquaculture: a review. Reviews in Aquaculture, 11, 1398–1417. https://doi.org/10.1111/raq.12299

Overton, K., Samsing, F., Oppedal, F., Stien, L. H., and Dempster, T. (2017). Lowering treatment temperature reduces salmon mortality: a new way to treat with hydrogen peroxide in aquaculture. Pest Management Science, 74, 535–540. https://doi.org/10.1002/ps.4751

Page, F. H., Losier, R., Haigh, S., Bakker, J., Chang, B. D., McCurdy, P., and Beattie, M. (2015). Transport and dispersal of sea lice topical therapeutants from salmon farm net-pens and well-boats. St. Andrews, NB.

Penston, M. J., Mckibben, M. A., Hay, D. W., and Gillibrand, P. A. (2004). Observations on open-water densities of sea lice larvae in Loch Shieldaig, Western Scotland. Aquaculture Research, 35, 793–805.

https://doi.org/10.1111/j.1365-2109.2004.01102.x

Pinheiro, J. C., and Bates, D. M. (2000). Mixed-Effects Models in S and S-PLUS. (J. Chambers, W. Eddy, W. Hardle, S. Sheather, and L. Tierney, Eds.), Statistics and Computing (1st ed.). New Y: Springer-Verlag New York, Inc.

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

Revie, C. W., Robbins, C., Gettinby, G., Kelly, L., and Treasurer, J. W. (2005). A mathematical model of the growth of sea lice, *Lepeophtheirus salmonis*, populations on farmed Atlantic salmon, *Salmo salar* L., in Scotland and its use in the assessment of treatment strategies. Journal of Fish Diseases, 28, 603–614. https://doi.org/10.1111/j.1365-2761.2005.00665.x

Salama, N. K. G., Collins, C. M., Fraser, J. G., Dunn, J., Pert, C. C., Murray, A. G., and Rabe, B. (2013). Development and assessment of a biophysical dispersal model for sea lice, 36, 323–337. https://doi.org/10.1111/jfd.12065

Salama, N. K. G., Dale, A. C., Ivanov, V. V., Cook, P. F., Pert, C. C., Collins, C. M., and Rabe, B. (2018). Using biological–physical modelling for informing sea lice dispersal in Loch Linnhe, Scotland. Journal of Fish Diseases, 41, 901–919. https://doi.org/10.1111/jfd.12693

Salama, N. K. G., Murray, A. G., and Rabe, B. (2016). Simulated environmental transport distances of *Lepeophtheirus salmonis* in Loch Linnhe, Scotland, for informing aquaculture area management structures. Journal of Fish Diseases, 39, 419–428. https://doi.org/10.1111/jfd.12375

Samsing, F., Oppedal, F., Dalvin, S., Johnsen, I., Vågseth, T., and Dempster, T. (2016). Sea lice (*Lepeophtheirus salmonis*) development times, body size, and reproductive outputs follow universal models of temperature dependence. Canadian Journal of Fisheries and Aquatic Sciences, 73, 1841–1851. https://doi.org/10.1139/cjfas-2016-0050

Sandvik, A. D., Bjørn, P. A., Ådlandsvik, B., Asplin, L., Skardhamar, J., Johnsen, I. A., … Skogen, M. D. (2016). Toward a model-based prediction system for sea lice infestation pressure. Aquaculture Environment Interactions, 8, 527–542. https://doi.org/10.3354/AEI00193

StataCorp (2019). *Stata: Release 16.* Statistical Software. StataCorp LLC, TX, USA.

Stien, A., Bjørn, P. A., Heuch, P. A., and Elston, D. A. (2005). Population dynamics of sea lice *Lepeophtheirus salmonis* on Atlantic salmon and sea trout, 290, 263–275.

Stucchi, D. J., Guo, M., Foreman, M. G. G., Czajko, P., Galbraith, M., Mackas, D. L., and Gillibrand, P. A. (2011). Modeling Sea Lice Production and

Concentrations in the Broughton Archipelago, British Columbia. In S. Jones and R. Beamish (Eds.), Sea lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Thakur, K. K., Vanderstichel, R., Barrell, J., Stryhn, H., Patanasatienkul, T., and Revie, C. W. (2018). Comparison of remotely-sensed sea surface temperature and salinity products with in situ measurements from British Columbia, Canada. Frontiers in Marine Science, 5, 1–11. https://doi.org/10.3389/fmars.2018.00121

Thomassen, J.M. (1993) Hydrogen peroxide as a delousing agent for Atlantic salmon. In G.A. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice, (1st ed., p. 378). Ellis Horwood Series in Aquaculture and Fisheries Support, Chichester, UK: CRC Press.

Treasurer, J. W., Grant, A., and Davis, P. J. (2000). Physical constraints of bath treatments of Atlantic salmon (*Salmo salar*) with a sea lice burden (Copepoda: Caligidae). Contributions to Zoology, 69. https://doi.org/10.1163/18759866-0690102014

Tucker, C. S., Sommerville, C., and Wootten, R. (2000). The effect of temperature and salinity on the settlement and survival of copepodids of *Lepeophtheirus salmonis* (Krøyer, 1837) on Atlantic salmon, *Salmo salar* L. Journal of Fish Diseases, 23, 309–320. https://doi.org/10.1046/j.1365-2761.2000.00219.x

Tully, O., and Nolan, D. T. (2002). A review of the population biology and host-parasite interactions of the sea louse *Lepeophtheirus salmonis* (Copepoda: Caligidae). Parasitology, 124, S165–S182. https://doi.org/10.1017/S0031182002001889

UK Met Office (2005). OSTIA Level 4 Sea Surface Temperature Analysis. Ver. 1.0. PO.DAAC, CA, USA. Retrieved from https://doi.org/10.5067/GHOST-4FK01 [2020-03-21].

van Etten, J., and de Sousa, K. (2019). gdistance: Distances and Routes on Geographical Grids. R package version 1.1-4. https://CRAN.R-project.org/package=gdistance

Westcott, J. D., Stryhn, H., Burka, J. F., and Hammell, K. L. (2008). Optimization and field use of a bioassay to monitor sea lice *Lepeophtheirus salmonis* sensitivity to emamectin benzoate. Diseases of Aquatic Organisms, 79, 119–131. https://doi.org/10.3354/dao01887

Whyte, S. K., Westcott, J. D., Elmoslemany, A., Hammell, K. L., and Revie, C. W. (2013). A fixed-dose approach to conducting emamectin benzoate tolerance assessments on field-collected sea lice, *Lepeophtheirus salmonis*. Journal of Fish Diseases, 36(3), 283–292. https://doi.org/10.1111/jfd.12055

Williams, P. J., Dick, K. B., and Yampolsky, L. Y. (2012). Heat tolerance, temperature acclimation, acute oxidative damage and canalization of haemoglobin expression in Daphnia. Evolutionary Ecology, 26(3), 591–609. https://doi.org/10.1007/s10682-011-9506-6

WoRMS Editorial Board. World Register of Marine Species. VLIZ. doi:10.14284/170

World Sea Temperature (2020). Saint Andrews Sea Temperature. Retrieved from https://www.seatemperature.org/north-america/canada/saint-andrews.htm [2020-08-04].

3.8 Supplemental information

3.8.1 Data

The Fish-iTrends is a web-based data management system that is operated by the Centre for Aquatic Health Sciences (CAHS) at the Atlantic Veterinary College (AVC) of the University of Prince Edward Island, Prince Edward Island, Canada (Gautam et al., 2016). The system was developed to collect information on sea lice and their treatments in Atlantic salmon marine sites for the provinces of NB, Nova Scotia, and Newfoundland and Labrador, Canada. Data access is limited to producers, aquatic veterinarians, and the CAHS for real-time analysis of sea lice abundance and production of summaries for other stakeholders.

A MySQL query of the Fish-iTrends database was performed for all NB Atlantic salmon marine site records between 06 July 2009 and 01 November 2018. Producer, owner, manager, and company information were withheld. Data consisted of sea lice counts of *L. salmonis* chalimus, pre-adult and adult male (PAAM), and adult female (AF), and *C. elongatus* on several Atlantic salmon from several cages at each site. The NB regulation requires counts of sea lice on samples of a minimum of 5 salmon in 6 cages at a site weekly if the sea temperature is above 5 °C (Government of New Brunswick, 2020) and a minimum of 10 fish immediately prior to and after a bath treatment. Sea lice counts for sea temperatures below 5°C are mandated monthly (Government of New Brunswick, 2020) because it is more difficult to access sites due to weather conditions and because fewer salmon rise to the surface for sampling.

The AF and PAAM counts of sea lice were divided by the number of fish sampled, which was often 10 fish. These AF/fish and PAAM/fish were transformed using a natural logarithmic function as the counts were skewed. An offset of 1 was applied as there were many zero values of the counts.

Separate MySQL queries were performed to obtain topical and in-feed treatments applied at cages within sites. Topical and in-feed treatments were combined with the count data. Data were not available for the in-feed therapeutic Lufenuron treatment applied at the hatchery, which started in 2017. Lufenuron administration reporting is submitted to the regulators, but the data of administration for this treatment are not recorded in the Fish-iTrends database.

Data were assessed with Stata® (2019, StataCorp). The data were analyzed for incomplete and inaccurate entries, labelled, and sorted according to date of the count by cage at a site.

3.8.2 Generation of weights

3.8.2.1 Temporal weights

Abundances of sea lice were averaged because counts at the cage level were not performed at 7 day intervals. A small sensitivity analysis was conducted to summarize the number of observations within varying temporal windows (11 to 31 days) for the calculation of the average abundances of AF sea lice. Empirical evidence supported the use of a time window of 15 days; this time window had the fewest zero counts for the narrowest window size.

AF sea lice abundance and PAAM sea lice abundance averages were calculated

using a triangular distribution across the 15 day window (AF: [14, 28] and PAAM: [21, 35] days prior to an observation). The IIP average abundances of AF sea lice were multiplied by the temporal weight and divided by the sum of the weights to normalize these values (Eq. 3.2 and 3.3). Normalization of the abundances meant that the averages did not reflect the number of observations that contributed to the calculation. This calculation was repeated for the IIP average abundance of PAAM sea lice. In comparison, the EIP average abundances of AF sea lice were not normalized.

$$\overline{\ln \mathrm{AF}}_i(t) = \sum_{c;s\in\Theta_t} w(s,t)\ln \mathrm{AF}_{ic}(s) \Big/ \sum_{c;s\in\Theta_t} w(s,t) \qquad \text{Eq. 3.2}$$

$$\overline{\ln \mathrm{PAAM}}_i(t) = \sum_{c;s\in\Theta_t} w(s,t)\ln \mathrm{PAAM}_{ic}(s) \Big/ \sum_{c;s\in\Theta_t} w(s,t) \qquad \text{Eq. 3.3}$$

Where: Average abundances of $\ln \mathrm{AF}_{ic}$ and $\ln \mathrm{PAAM}_{ic}$ were obtained via $\mathrm{AF}_{ic}(t) = \ln\left(^{(\mathrm{AF}_{ic}+1)}\Big/_{\text{no. fish sampled}}\right)$

c ~ cage within site i

t ~ target date (day); s ~ other times

$w(s,t)$ weight for time s at time t (based on time distance between *s* and *t*); triangular distribution weights $w(s,t) = {}^{(7.5-min(7.5,|s,t|))}\Big/_{56.25}$, $w(s,t) \in [-7.5, 7.5]$,

θ_t ~ time window around time t, i.e., the interval (t - 7, t + 7)

Note: In the expressions 1 and 2, it was understood that the summations are over cages c within sites i and times s where $\ln \mathrm{AF}_{ic}(s)$ and $\ln \mathrm{PAAM}_{ic}(s)$ exist.

3.10.2.2 Spatial weights

Seaway distances among sites were calculated using the 2017 published map

(PDF format) of sites in the Bay of Fundy, NB (Department of Energy and Resource Development/Agriculture, and Fisheries, 2017). The map of sites was converted into a raster image file with a 100 x 100 unit grid in QGIS (QGIS.org, 2018). The raster image was geo-referenced using landmarks between the image and Google Maps. This geo-referenced image was formatted to the NAD83(CSRS)/New Brunswick Stereographic (EPSG:2953) projection. Approximate centre points (centroids) for each NB site were selected manually. This map of data points was imported to R (R Core Team, 2020) using the ‘rgdal’ package (version 1.4-8; Bivand et al., 2019).

A waterway map for the Bay of Fundy, NB, was created in QGIS by taking the difference between a designed large bounding box (polygon) and the land boundary (Administrative Area; GADM, 2018). The projection for the waterway map was converted to NAD83. This vector map was converted to a raster layer with water cost values as 1 and land values as 'No Data.' The waterway raster image was imported into R and converted to a 100 x 100 m grid size using the ‘raster’ package (version 3.0-12; Hijmans et al., 2020). A transition raster was created from the waterway raster using the 'gdistance' package (version 1.1-4; van Etten and de Sousa, 2012) in R utilizing a mean function to calculate the transition values from the grid values, 8 directions for the connection of grid cells and a symmetrical matrix output. An East-West direction correction was applied to the transition layer and produced a matrix with corrected values. The seaway distances (least-cost distance among sites) were calculated for the approximate centroids for all sites within the corrected transition waterway layer. Distances

among sites were converted to km.

The output matrix of distances among sites was used to calculate the weights using the Gaussian Kernel Density Estimate (KDE). KDE was estimated for the distance between each pair of sites and for kernel bandwidths of 5 to 60 km. The Gaussian distribution was set to maintain a consistent precision across the range of bandwidths. KDE weights smaller than e^{-18} were set to zero. In Stata, the weights were merged by site with the dataset containing the segments, IIP_{AF} and IIP_{PAAM}, and the average AF sea lice abundances for the summation procedure to generate the EIP_{AF}.

The summation procedure consisted of multiplying the weight (KDE) by the average abundance of AF sea lice for every pair of sites and generated a matrix of spatially and temporally weighted abundances. The sum of the values across the row represented the EIP for a site with all its neighbours (Eq. 3.3). This summation procedure was repeated for each kernel bandwidth.

$$\mathrm{EIP} = \sum_{j \in N_i} w_{ij}\, \overline{\ln \mathrm{AF}}_j(t-21) \qquad \text{Eq. 3.3}$$

Where: $\overline{\ln \mathrm{AF}}_i(t)$ is the triangular temporal average abundances of AF sea lice for site i at time t.

w_{ij} are Gaussian kernel weights (e.g., Elghafghuf et al., 2020) computed based on seaway distance between sites i and other sites j. Gaussian kernel function bandwidth was evaluated at different values (i.e., 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 20, 25, 30, 35, 40, 45, 50, 55, and 60 km)

t ~ target date (day)

j ~ other sites near site i (i.e., N_i~ neighbour sites of i)

Note: In the expression, it was understood that the summations are over j at time t where $\ln\mathrm{AF}_j(t-21)$ exists.

3.8.3 Final model

The final model described was a multilevel mixed-effects linear regression with site-cycle, cage-cycle, and segment random effects, and an exponential correlation structure to the errors, stratified by three temperature profiles (Eq. 3.4). Segments are defined by the first count after a bath treatment and end with the last count prior to the next bath treatment, and are limited to a site-cycle and cage cycle (Figure 3.5).

$$\begin{aligned}\overline{lnAF} = {} & \beta_0 + \beta_1 IIP_{AF} + \beta_2 IIP_{PAAM} + \beta_3 EIP_{AF} + \beta_4 baseline_{AF} \\ & + \beta_5 wT + \beta_6 Azamethiphos\ Tarp + \beta_7 Azamethiphos\ Skirt \\ & + \beta_8 Azamethiphos\ Wellboat + \beta_9 Deltamethrin \\ & + \beta_{10} Warm\ water + \beta_{11} Hydrolicer \\ & + \beta_{12} wT * Azamethiphos\ Tarp \\ & + \beta_{13} wT * Azamethiphos\ Skirt \\ & + \beta_{14} wT * Azamethiphos\ Wellboat + \beta_{15} wT * Deltamethrin \\ & + \beta_{16} wT * Warm\ water + \beta_{17} wT * Hydrolicer + \beta_{18} BMA\ 1 \\ & + \beta_{19} BMA\ 2A + \beta_{20} BMA\ 2B + \beta_{17} wT * Hydrolicer \\ & + \beta_{23} BMA\ 3C + \mu_{sc} + \mu_{cc} + \mu_{seg} + \varepsilon_{seg,sc,cc}\end{aligned} \qquad \text{Eq. 3.4}$$

Where: wT: water temperature in °C
sc: site-cycle
cc: cage-cycle
seg: segment
$\varepsilon_{seg,sc,cc(1)}, \ldots, \varepsilon_{seg,sc,cc(11,658)}$ are ~N(0, $\sigma^2_{seg,sc,cc}$ is dependent on $g_{(i)}$),
$\varepsilon_{seg,sc,cc}$ within a segment are dependent on time distance in days,
$\varepsilon_{seg,sc,cc}$ between segments are independent,
$\mu_{site-cycle}$'s (~$\mathrm{N}(0,\sigma^2_{sc})$) are the random site-production cycle effects,
$\mu_{cage-cycle}$'s (~$\mathrm{N}(0,\sigma^2_{cc})$) are the random cage-production cycle effects,
$\mu_{segment}$'s (~$\mathrm{N}(0,\sigma^2_{seg})$) are the random segment cycle effects, and
$g_{(i)}$ is the group of $\mathrm{wT_{seg}}$ and have $\sigma^2_{g_{(i)}}$.

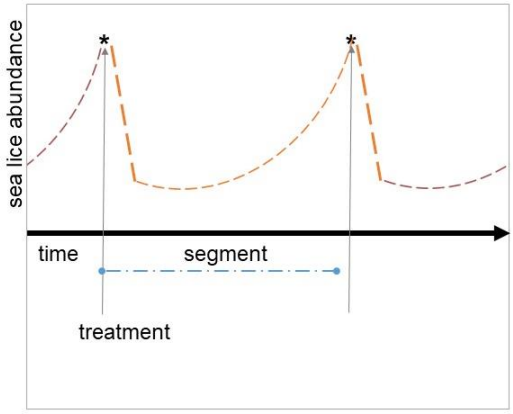

Figure 3.5. Organization of ‘segments’ of sea lice counts with time and topical treatments.

3.8.4 Sensitivity analyses

3.8.4.1 Evaluation of the effect of the inclusion criteria

We considered that these relatively strict criteria may introduce bias or misrepresentation of the population; therefore, these criteria were relaxed, and the effect was evaluated. The segment length was extended to 500 days. The upper limit of the segment size was removed. The maximum gap in a segment was extended to 120 days. Models were run for each of these changes and for all changes simultaneously. The relative difference of the estimates from each model were compared against the model with the strictest inclusion criteria (i.e., final model).

The relaxation of the upper limits of the inclusion criteria of segment maximum gap, segment length and segment size simultaneously provided an additional 3 site-cycles, and 62 cage-cycles. This resulted in changes to the estimated coefficients that were considered minor (Table 3.5). The relative difference of estimated coefficients was sensitive to minor differences due to the small size of

the estimates. The relationships and directions of the coefficients remained the same.

The profile of segments changed by the BMA. This suggested that sites within certain BMA treat less frequently (longer segments) compared to others (shorter segments). As segments increased the effect of $baseline_{AF}$ increased. This trend may be explained by the inclusion of more cages with fewer treatments; therefore, the impact of a louse prior to treatment has a greater effect.

The relaxation of the inclusion criteria lower limits was not evaluated in this sensitivity analysis. Short segments occurred when there were multiple treatments in short succession within a cage. We expect the potential selection bias related to the inclusion of short segments would reduce the effect of infestation pressures.

Table 3.5. Estimated coefficient change (difference/average against the final model (n = 11,658) with a cutoff of |25 %|): - no change, ↑ increase, and ↓ decrease for the multivariable linear mixed model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice with an external infestation pressure weighted with a kernel density estimate that had a bandwidth of 10 km. n_{obs}: number of observations, n_{seg}: number of segments, Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice.

Variable and categories	Segment length extended	Segment size extended	Maximum gap extended	All 3 extended
n_{obs}	11,706	13,009	12,180	14,276
n_{seg}	2,402	2,464	2,508	2,610
IIP_{AF}	-	-	-	-
IIP_{PAAM}	-	-	-	-
EIP_{AF}	-	-	-	-
$Baseline_{AF}$	↑	↑	↑	↑
Temperature (°C)	-	-	-	-
Treatment				
Azamethiphos Tarpaulin	baseline			
Azamethiphos Skirt	-	-	-	-
Azamethiphos Wellboat	-	-	-	-

Table 3.5 continued next page.

Table 3.5 continued.

Variable and categories	Segment length extended	Segment size extended	Maximum gap extended	All 3 extended
Treatment				
Deltamethrin	-	-	-	↓
Warm water	-	-	-	-
Hydrolicer®	-	-	-	↑
Hydrogen peroxide	-	-	-	-
Treatment-Temperature interaction				
Azamethiphos Tarp*Temp	baseline			
Azamethiphos Skirt*Temp	-	-	-	-
Azamethiphos Boat*Temp	-	↑	-	-
Deltamethrin*Temp	-	-	-	↓
Warm water*Temp	-	-	-	-
Hydrolicer®*Temp	-	-	↑	↑
Hydrogen peroxide*Temp	-	-	-	-

Table 3.5 continued next page.

Table 3.5 continued.

Variable and categories	Segment length extended	Segment size extended	Maximum gap extended	All 3 extended
BMA				
1	baseline			
2A	-	↑	↓	↑
2B	↑	-	-	↓
3A	-	↑	-	↑
3B	↑	↑	↓	↑
3C	-	↓	-	↓

3.10.4.2 Evaluation of the effect of potential outliers

Evaluation of model fit revealed a few sites and segments were potential outliers. These findings were based on the evaluation of the standardized residuals; it should be noted that Stata has poor performance regarding standardized residuals.

Many of the sites identified as potential outliers originated from within the BMA 1. This finding may be due to the higher frequency of cages in the BMA 1. We noted that observations with extreme standardized residuals occurred late in the production cycle and in segments following treatment almost exclusively with the azamethiphos tarpaulin and hydrogen peroxide treatments. There were no trends noted in the continuous variables that could explain these potential outliers.

Although the variances of the random effects of segments were minute, the plot of the BLUP demonstrated two segments were completely different from the rest. Both segments originated from the same site and the 3rd production cycle. It was noted that the segments were treated with azamethiphos applied with a tarpaulin and were the 11th segment for that cage in that production cycle. Otherwise, these 2 segments were average.

The final model without these observations from sites with high standardized residuals produced lower IIP_{PAAM} and BMA 3B estimated coefficients, and higher BMA 2A and 3C estimated coefficients. Other estimated coefficients were robust to the removal of these observations. In a separate model, the two segments identified contributed 36 observations; removal of these segments resulted in

significant changes to most of the estimates. The estimated coefficients of the IIP_{AF} and IIP_{PAAM} decreased, the EIP_{AF} increased, and each of the BMA increased. At 7 and 9 °C, the deltamethrin treatment estimated coefficient increased, but decreased at 11.5 °C. The Hydrolicer® treatment estimates decreased at 7, 9 and, 11.5 °C

3.10.4.3 Evaluation of the effect of baseline abundance of sea lice

The $baseline_{AF}$ predictor was kept in the model despite the loss of observations (34.4% observations and 35.6% segments). This decision was made for multiple reasons. The first was for biological reasons as the abundance of sea lice after treatment is thought to be associated with the abundance immediately prior to treatment (Gautam et al., 2017a). Second, the $baseline_{AF}$ loss of observations was related to cohort issues where groups of salmon were sometimes split between multiple cages during the event of a treatment. Also, this predictor had a significant effect in the final model. Finally, the sensitivity analysis did not change the estimated coefficients by a large magnitude.

The sensitivity analysis involved the comparison of the final model and two models adjusted with the omission of the $baseline_{AF}$ predictor (Table 3.6). The first adjusted model utilized the sample from the final model and the second adjusted model was run without that limitation.

The infestation pressures (IIP_{AF}, IIP_{PAAM}, and EIP_{AF}) remained significant predictors in the model. The estimated IIP_{PAAM} coefficient decreased when the

$baseline_{AF}$ was included in the model. This finding suggests that the final model may underestimate the IIP_{PAAM}.

The estimated coefficient for the Hydrolicer® treatment changed by a large magnitude, but that is due to the large standard error for this predictor (final model SE 2.29). The Hydrolicer® treatment was the least frequently applied.

From this small sensitivity analysis, we concluded that there was not a strong selection bias. If any selection bias were present due to the inclusion of the $baseline_{AF}$ then it would affect a change toward the null.

Table 3.6. Estimated coefficients (coef.) and standard errors (SE) for the final multivariable linear mixed model for the abundance of adult female *Lepeophtheirus salmonis* and adjusted models with the omitted predictor $baseline_{AF}$: model 2 with final model sample and model 3 without constraint of final model sample. Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice. Final and model 2: n_{obs} = 11,658, n_{seg} = 2,398. Model 3: n_{obs}=17,762, n_{seg} = 3,723.

Variable and categories	Final model		Model 2		Model 3	
	Coef.	SE	Coef.	SE	Coef.	SE
IIP_{AF}	0.188	0.016	0.283	0.015	0.275	0.012
IIP_{PAAM}	0.095	0.013	0.096	0.014	0.141	0.011
EIP_{AF}	0.478	0.053	0.625	0.053	0.676	0.044
$Baseline_{AF}$	0.287	0.017	omitted		omitted	
Temperature (°C)	-0.111	0.008	-0.115	0.008	-0.112	0.007
Treatment						
Azamethiphos Tarpaulin	baseline					
Azamethiphos Skirt	-1.966	0.336	-2.124	0.343	-2.621	0.257
Azamethiphos Wellboat	-1.492	0.292	-2.118	0.302	-1.726	0.240
Deltamethrin	-0.953	1.337	-0.894	1.355	-0.823	0.926
Warm water	-3.074	0.284	-3.258	0.290	-2.844	0.217

Table 3.6 continued next page.

Table 3.6 continued.

Variable and categories	Final model		Model 2		Model 3	
	Coef.	SE	Coef.	SE	Coef.	SE
Treatment						
Hydrolicer®	0.932	2.294	0.190	2.316	-1.693	0.891
Hydrogen peroxide	-2.134	0.129	-2.399	0.133	-2.303	0.109
Treatment-temperature interaction						
Azamethiphos Tarp*Temp	baseline					
Azamethiphos Skirt*Temp	0.200	0.026	0.189	0.027	0.212	0.021
Azamethiphos Boat*Temp	0.091	0.025	0.129	0.025	0.114	0.020
Deltamethrin*Temp	0.078	0.112	0.067	0.113	0.056	0.079
Warm water*Temp	0.223	0.023	0.239	0.023	0.218	0.017
Hydrolicer®*Temp	-0.020	0.175	0.038	0.177	0.171	0.066
Hydrogen peroxide*Temp	0.154	0.012	0.150	0.012	0.161	0.010

Table 3.6 continued next page.

Table 3.6 continued.

Variable and categories	Final model		Model 2		Model 3	
	Coef.	SE	Coef.	SE	Coef.	SE
BMA						
1	baseline					
2A	-0.015	0.110	-0.125	0.113	-0.124	0.105
2B	-0.269	0.154	-0.232	0.160	-0.270	0.141
3A	0.269	0.159	0.181	0.164	0.118	0.129
3B	-0.005	0.129	0.027	0.133	0.007	0.118
3C	-0.494	0.358	-1.001	0.366	-1.078	0.359
Constant	1.521	0.121	2.127	0.120	2.029	0.109

3.8.5 References

Bivand, R., Keitt, T., Rowlingson, B., Pebesma, E., Sumner, M., Hijmans, R., … Rundel, C. (2019). rgdal: Bindings for the 'Geospatial' Data Abstraction Library. R package version 1.4-8. https://CRAN.R-project.org/package=rgdal

Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). *Bay of Fundy Marine Aquaculture Sites* [map]. 1:125000. New Brunswick, Canada.

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

GADM. (2018). World/Canada/New Brunswick. Retrieved from https://gadm.org/maps/CAN/newbrunswick.html

Gautam, R., Boerlage, A. S., Vanderstichel, R., Revie, C. W., and Hammell, K. L. (2016). Variation in pre-treatment count lead time and its effect on baseline estimates of cage-level sea lice abundance. Journal of Fish Diseases, 39(11), 1297–1303. https://doi.org/10.1111/jfd.12460

Government of New Brunswick. (2020). General Regulation, NB Reg 91-158, 1–14. Retrieved from https://www.canlii.org/en/nb/laws/regu/nb-reg-91-158/latest/nb-reg-91-158.html

Hijmans, R.J., van Etten, J., Sumner, M., Cheng, J., Baston, D., Bevan, A., … Wueest, R. (2019). raster: Geographic Data Analysis and Modeling. R package version 3.0-12. https://CRAN.R-project.org/package=raster

QGIS.org (2018). QGIS Geographic Information System. Open Source Geospatial Foundation Project. http://qgis.org.

StataCorp (2019). *Stata: Release 16.* Statistical Software. StataCorp LLC, TX, USA.

UK Met Office (2005). GHRSST Level 4 OSTIA Global Foundation Sea Surface Temperature Analysis. Ver. 1.0. PO.DAAC, CA, USA. Dataset accessed [2020-03-21] at https://doi.org/10.5067/GHOST-4FK01.

van Etten, J., and de Sousa, K. (2019). gdistance: Distances and Routes on Geographical Grids. R package version 1.1-4. https://CRAN.R-project.org/package=gdistance

4 Estimation and comparison of connectivity measures for the dispersal of *Lepeophtheirus salmonis* sea lice among Atlantic salmon farms in New Brunswick, Canada

4.1 Abstract

Atlantic salmon aquaculture sites in the Bay of Fundy, New Brunswick (NB), share a common coastal body of water, which potentiates the exchange of free-swimming stages of the parasite *Lepeophtheirus salmonis* among sites, where each site is an aggregation of salmon hosts for the attached and mobile stages of the parasite. The measure of seaway distances among sites is commonly used to capture some of the effects of connectivity and transmission patterns among sites. However, we propose novel measures to quantify connectivity, some with the use of bathymetry data to improve on seaway distances, but without extensive hydrodynamic modelling. We compared seaway distance at differing grid resolutions, topographical distance, percent surface area of water along seaway path, and volume of water along seaway path directly and indirectly. These connectivity measures were indirectly compared through the external infestation pressure (EIP) of the model for the abundance of adult female *L. salmonis* from empirical data stored in the Fish-iTrends database from 2009-2018. The percent surface area of water along seaway path improved the model fit as compared to the seaway distance with 100 m grid resolution. An estimate of dispersal distance of 7-10 km was produced through the comparison of optimal model fit with EIP using the Akaike information criterion (AIC), supporting the continued use of area-wide parasite management in the Bay of Fundy, NB.

4.2 Introduction

The Atlantic salmon, *Salmo salar* (Linnaeus [WoRMS, 2022]), aquaculture industry in the Bay of Fundy, New Brunswick (NB), Canada, contributed to a production of 28 thousand tonnes earning up to 270 million $CAD in 2018 (Statistics Canada, 2020). Since the mid-1990's (Hogans, 1995), the parasitic sea louse, *Lepeophtheirus salmonis salmonis* (Krøyer [WoRMS, 2022]) has necessitated monitoring and the implementation of management practices by the Atlantic salmon aquaculture industry in the Bay of Fundy, NB. Producers and managers of aquaculture farms (hereafter, sites) mitigate the effects of this parasite using fallowing, single year class area management, and control options that include chemotherapeutic agents and non-medicinal removal (see Overton et al., 2019 for review).

The implementation of mitigation strategies against *L. salmonis* is improved by the knowledge of connectivity among sites (Adams et al., 2015; Samsing et al., 2017). Connectivity is the concept that an animal or pathogen's movement among habitat patches (which are locations with distinct characteristics responsive to the biology of the animal or pathogen [Girvetz and Greco, 2007]) is facilitated or impeded by its environment (Tischendorf and Fahrig, 2000; Cowen and Sponaugle, 2009; Le Corre et al., 2012). In the case of *L. salmonis* in the Bay of Fundy, NB, sites act as habitat patches because they are distinct aggregations of salmon hosts for the non-free-swimming stages of lice (the attached copepodid, and the mobile stages: chalimus I and II, pre-adult I and II, and adults). Although wild salmon occur in areas between sites, sites can be

considered habitat patches due to the limited number of wild salmonids that can serve as hosts for lice in comparison to the population clusters within sites that contribute meaningfully to transmission of lice among sites. Connectivity among sites occurs as they share the same ecological resource, the coastal body of water, thus allowing the exchange of drifting material (Le Corre et al., 2012) including planktonic stages of sea lice (nauplii I and II, and early copepodid [Cantrell et al., 2018]) and egg strings that will develop into those planktonic stages (Eisenhauer et al., 2020). Typically, the nauplii I are the first planktonic stage considered as they emerge directly from the egg string attached to the gravid female, however, Eisenhauer et al. (2020) recently demonstrated that egg strings detached from the female continue to develop and may be transported by currents and contribute to the dispersal of the sea lice. The knowledge of connectivity may be applied to interrupt the transmission of planktonic stages of sea lice among habitat patches (i.e., sites) by separating sites at epidemiologically meaningful distances (Samsing et al., 2017, 2019).

Epidemiologically meaningful distances can be defined as the distance greater than the effective dispersal distance for planktonic sea lice. The effective dispersal is the movement of eggs and planktonic sea lice by water currents from their point of release (Shanks et al., 2003) that results in successful parasitism of salmon and ensuing reproduction (adapted for sea lice from Siegel et al., 2003). The concept of effective dispersal has the outcome of successful parasitism which is a probability and not a binary success/failure outcome as the outcome of sea lice counted on salmon needs to be sufficiently large to allow for detection

and therefore label it successful parasitism. Effective dispersal distances of *L. salmonis* sea lice have been estimated using a data-driven approach in various regions around the world (e.g., Aldrin et al., 2013, 2017; Kristoffersen et al., 2013; Elghafghuf et al., 2020). In the Bay of Fundy, NB, the study in Chapter 3 estimated a dispersal distance of 10 km using a multilevel mixed-effects linear regression model by quantifying the external infestation pressure (EIP; infestation pressure, a measure that represents the dose of exposure of parasitic stages of sea lice to potential fish hosts) from the abundance of adult female (AF) sea lice weighted by a Gaussian kernel density estimate (KDE) of seaway distances among sites. For Grand Manan Island, NB, Elghafghuf et al. (2020) estimated a dispersal distance of 12.5 km using a state-space model of the infestation pressure supplied by sea lice abundance weighted by seaway distance. Biophysical models, which are a combination of hydrodynamic models with particle-tracking models, (e.g., Salama et al., 2013; Asplin et al., 2014; Johnsen et al., 2016; Samsing et al., 2017; Cantrell et al., 2018, 2021) can also estimate dispersal distances of planktonic lice but require intensive modelling processes.

We propose refinement of seaway distance measures for new connectivity measures that require bathymetry data, but without extensive ocean current modelling, to estimate the dispersal distance of *L. salmonis* sea lice in the Bay of Fundy, NB. These new connectivity measures may provide insight into the observed variation of *L. salmonis* sea lice abundance at salmon farms (Aldrin et al., 2017). Conceptually, topographic distance is a land-based ecological measure of the distance among habitat patches accounting for topographic relief

(elevation of land) and estimates dispersal-driven geographical connectivity (Wang, 2019). Topographic distance has been used successfully to study dispersal patterns and evaluate habitat fragmentation on land (e.g., Bista et al., 2021). However, this connectivity measure has not been employed for water environments, as far as the authors are aware, and it may prove useful to understand the dispersal of *L. salmonis* sea lice. Contiguous to topography is the measure of depth, bathymetry, which may influence *L. salmonis*, as depth affects their distributions (Frenzl, 2014). The volume of water along the seaway path among sites can be calculated using the bathymetric maps for the area and is proposed to affect the dispersal of sea lice through the assumption that larger volumes of water result in greater dilution of planktonic sea lice and may impede successful dispersal. Additionally, water volume is associated with physical properties when modelling coastal water movements and may be indirectly associated with coastal hydrodynamic patterns. In a similar fashion, the cumulative percent surface area of water along a pre-determined buffer (e.g., 500 m) surrounding the seaway path may also reflect shallower waters nearshore, thus potentially affecting the dispersal of sea lice as eggs and planktonic lice experience slower current speeds nearshore compared to open waters (Shanks, 2009).

The objectives of this study were to compare different connectivity measures of dispersal distance and assess their impact when used in the calculation of external infestation pressure on the modelling of abundance of the parasitic sea louse, *L. salmonis*, in the Bay of Fundy, NB. We utilized a previously published

model (Chapter 3) to perform these comparisons, using the same study area and dataset, as a framework for more direct comparisons of the different connectivity measures through their estimated dispersal distances and their impacts of infestation pressures on the abundance of *L. salmonis*.

4.3 Materials and methods

4.3.1 Study area and population

Data were extracted from the Fish-iTrends data management system that is operated by the Centre for Aquatic Health Sciences (CAHS) at the Atlantic Veterinary College (AVC) of the University of Prince Edward Island, Prince Edward Island, Canada (for more detailed description, see Gautam et al., 2016). The dataset was composed of counts of sea lice (*L. salmonis* and *Caligus elongatus* [von Nordmann {WoRMS, 2022}]) from regular monitoring and counts associated with pre- and post-treatments (topical and oral therapeutants) for the mitigation of these lice for all active Atlantic salmon aquaculture sites in the Bay of Fundy, NB, between July 2009 and November 2018. The monitoring and pre- and post-treatment counts consisted of pre-adult and adult male (PAAM) and AF (with and without egg strings) sea lice from several cages at each site during their production cycle (see Gautam et al., 2016 for detailed data records description). There were 62 active sites included in the study and these were used for the calculation of the connectivity measures presented henceforth. Additional details of the data and query of the Fish-iTrends database can be found in Chapter 3.

4.3.2 Connectivity measures

The connectivity measure of seaway distance with 100 m grid resolution (Chapter 3) and 25 m grid resolution, and the new connectivity measures of topographic distance, volume of water, and percent surface area of water were calculated for all pairs of sites in the study area.

Except for seaway distances, the connectivity measures were constructed from an interpolation of depth points from the regional depth map (bathymetry map) obtained from D. Greenberg and the Department of Fisheries and Oceans Canada (DFO). An elevation raster (25 m grid resolution) was produced that contained data of the average height of the water columns for cells among sites. Additional details are found in Chapter 4's supplemental information (section 4.8).

Relationships between selected connectivity measures for all pairs of sites were represented graphically by a matrix plot and quantified by the Pearson's correlation coefficient (ρ) and Euclidean distance (L_2). Prior to analysis, each connectivity measure was scaled by dividing by the standard deviation of the connectivity measure across all pairs of sites. The scaling process was performed to ensure that the spread of the points among connectivity measures was the same. Euclidean distance represents a "straight-line" distance between two points in high-dimensional abstract space and was used to quantify the deviations between the connectivity measures across all pairs of sites (additional information provided in Chapter 4's supplemental information [section 4.8]).

Additional details for the generation of all connectivity measures can be found in Chapter 4's supplemental information (section 4.8).

4.3.2.1 Seaway distance

The original model was constructed with a Gaussian KDE weighted spatial average abundance of adult female (AF) sea lice that utilized seaway distance at a resolution of 100 metres (SWD_{100}; Figure 4.1A). The seaway distances (least-cost distance between sites) among sites were calculated using the 'costDistance' function of the 'gdistance' package (version 1.1-4 van Etten, 2012) in R (R Core Team, 2020) for the approximate centroids of sites with a 100 x 100 m grid in QGIS. The procedure was repeated with a finer resolution using a 25 x 25 m grid for analysis in this study (SWD_{25}).

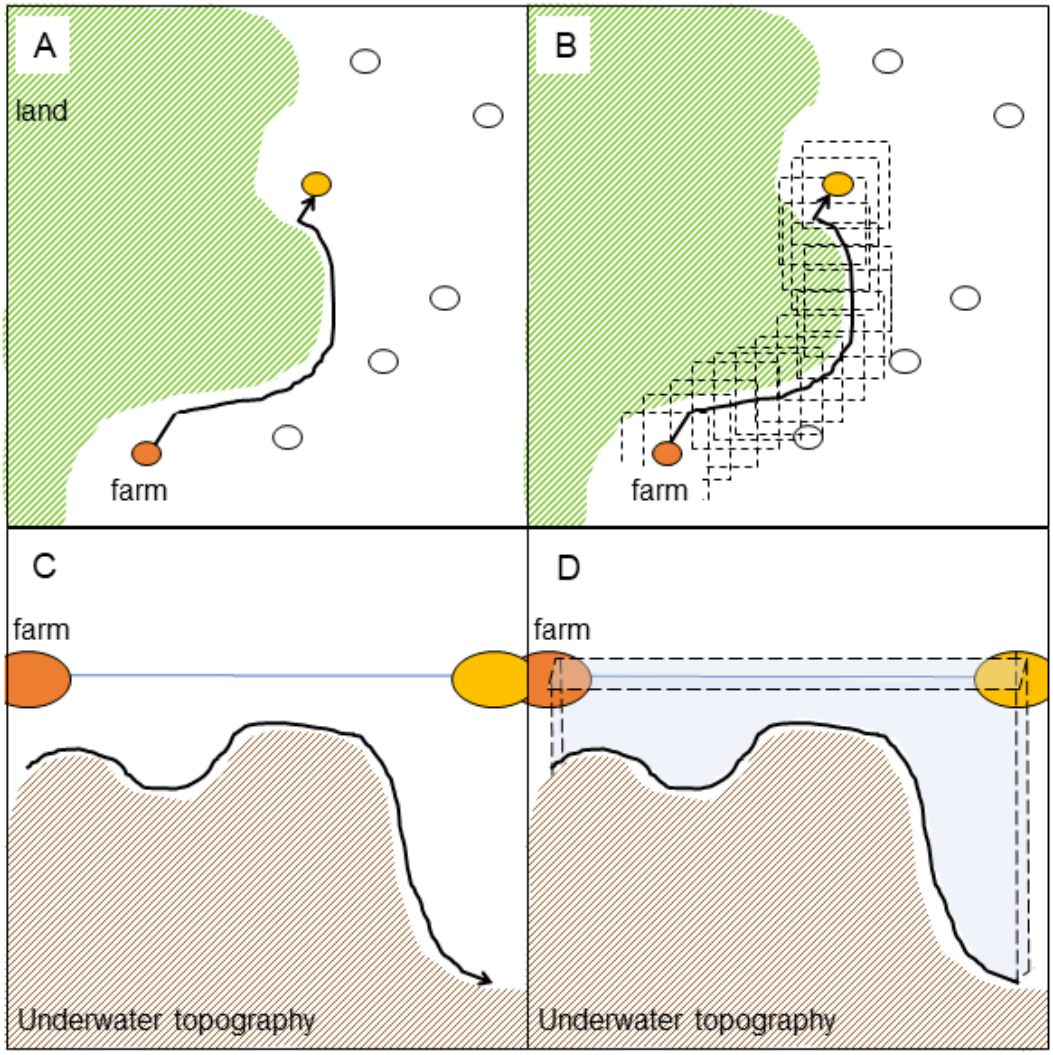

Figure 4.1. Depictions of the methods for the generation of connectivity matrices used in the production of Gaussian kernel density estimate weights for the calculation of external infestation pressure. A: seaway distance; B: percent surface area of water along seaway path with a square window area; C: topographical distance; D: volume of water along seaway path. Additional matrices were produced by adjusting the resolution (A), increasing the covered area to 250 m, 500m, and 1,000 m (B and D), and increasing the cost of movement along an incline by multiplying by a factor of 10, 100, 500, and 1,000 (C).

4.3.2.2 Topographical distance

The ‘topoDist’ function of the ‘topoDistance’ package (version 1.0.1 Wang, 2019) was applied to the elevation raster (TD; Figure 4.1B). The function calculated the distance along the shortest topographic path among sites. The distance in a path is the sum of distances across the individual cells in the raster image, where a total of 8 possible directions can be taken from one cell to the next neighbouring cell; and all eligible paths progress entirely through water. For each cell in a 25 x 25 m grid, the topographical distance was calculated as the hypotenuse of the width of the horizontal distance and the difference in depth travelled through the cell (m). Weights were applied to the raster prior to applying the ‘topoDist’ function and produced topographic paths where the cost of movement along an incline increased by factors of 10, 100, 500 and 1,000 (TD_{10}, TD_{100}, TD_{500} and TD_{1000}, respectively).

4.3.2.3 Percent surface area of water along seaway path

The topographic least cost paths among sites were calculated using the ‘topoLCP’ function of the ‘topoDistance’ package (version 1.0.1 Wang, 2019). A square window area was designated by selecting 250 m in each cardinal direction from the centroid of a site (SA_{250}; Figure 4.1C). Within the window, the average percentage of water was calculated using the function ‘focal’ from the ‘raster’ package (Hijmans et al., 2020) in R. The window was then moved one cell (25 x 25 m) along the least cost path between the pair of sites and the average percentage of water was calculated again. The least cost path is the

shortest distance between a pair of sites with adjustment by a resistance-based measure (e.g., Wang et al., 2009) set to the waterway map raster. The resulting least cost paths among sites were perfectly correlated to the seaway distance (ρ and concordance correlation coefficient [ρ_c] = 1.00). The procedure for the average percentage of water for a cell was repeated for every cell along the seaway paths among sites and resulted in a raster map of cumulative average of water percentage. The window area for the calculation of the average percentage of water was adjusted to distances of 500 m and 1,000 m (SA_{500} and SA_{1000}, respectively).

4.3.2.4 Volume of water along seaway path

The topographic least cost paths among sites were calculated using the 'topoLCP' function of the 'topoDistance' package (version 1.0.1 Wang, 2019). The depth along the topographic least cost paths among sites were extracted from the bathymetry raster layer. The volume of water along the least cost path (VW; Figure 4.1D) was obtained by multiplying the average depth per cell with the area constant of a cell (25 x 25 m cell = 625 m^2) to obtain the volume of water for that cell. The number of cells included in the estimation of volume of water was adjusted to 250 m, 500 m, and 1,000 m (VW_{250}, VW_{500} and VW_{1000}, respectively).

4.3.3 Model

4.3.3.1 Data structure

Data were hierarchically structured with multiple cages within sites and multiple sites within bay management areas (BMAs). Production cycles (stock and harvest of salmon within BMAs) further defined groups of fish. Observations (counts) occurred as repeated measurements of sea lice at a cage in a production cycle at a site. These observations were grouped into segments, where a segment was defined as the first observation after a topical treatment followed by all additional observations until the following treatment within a production cycle and limited to a single cohort of salmon (defined by salmon with the same fish group identifier, cage, site, and production cycle). Additional details on the organization of observations into segments is described in Chapter 3.

4.3.3.2 External infestation pressure

The EIP consisted of the triangular-weighted temporal and Gaussian KDE-weighted spatial average abundance of AF sea lice (variable EIP_{AF}). The temporal weight was calculated for the abundance of AF sea lice at other sites centred at a fixed lag of 21 days prior to an observation as estimated by the most rapid development of the sea lice from egg to adult female at the annual maximum water temperature of 15 °C. All observations for a site were included (whether part of a segment or not), and no adjustments were made for observations that occurred prior to and after treatments. The spatial portion of the EIP_{AF} Gaussian KDE weights was calculated at bandwidths of 5 to 15 km for

each of the linearly scaled connectivity measures among active sites ([13 connectivity measures + original SWD_{100}] x 11 KDE bandwidths = 154 EIP_{AF}; Table 4.1). The Gaussian KDE weights were calculated using the 'gdistance' package, version 1.3-1 (van Etten and de Sousa, 2019) in R for each of the connectivity matrices, which were the connectivity measure organized as a matrix of the measure among all pairs of sites. Additional details for the calculation of EIP_{AF} can be found in the supplemental information.

Table 4.1. Quantified distances among scaled connectivity measures using Euclidean distance. Connectivity measures were seaway distance at 25 m (SWD_{25}) and 100 m resolutions (SWD_{100}), percent surface area of water along seaway path with a square window area of 250 m (SA_{250}), 500 m (SA_{500}), and 1,000 m (SA_{1000}), topographical distance (TD) with adjustment of the cost of movement along an incline by factors of 10 (TD_{10}), 100 (TD_{100}), 500 (TD_{500}), and 1,000 (TD_{1000}), and volume of water along seaway path (VW) with adjustment of the number of cells included in the estimation to 250 m (VW_{250}), 500 m (VW_{500}), and 1,000 m (VW_{1000}).

Connectivity measure	SWD_{25}	SWD_{100}	SA_{250}	SA_{500}	SA_{1000}	TD	TD_{10}	TD_{100}	TD_{500}	TD_{1000}	VW	VW_{250}	VW_{500}	VW_{1000}
SWD_{25}	–													
SWD_{100}	0.34	–												
SA_{250}	5.57	5.61	–											
SA_{500}	6.01	6.00	1.28	–										
SA_{1000}	6.66	6.63	2.75	1.56	–									
TD	0.02	0.35	5.57	6.01	6.67	–								
TD_{10}	1.13	0.35	5.59	6.17	6.98	1.11	–							
TD_{100}	14.59	14.76	15.36	16.31	17.46	14.58	14.58	–						
TD_{500}	28.89	29.06	28.83	29.68	30.68	28.88	28.88	15.25	–					
TD_{1000}	31.99	32.15	31.84	32.65	33.61	31.98	31.98	18.64	3.65	–				
VW	20.64	20.55	18.84	17.96	16.78	20.64	20.64	30.64	40.95	43.25	–			
VW_{250}	21.12	21.03	19.34	18.44	17.25	21.12	21.12	31.18	41.46	43.76	0.72	–		
VW_{500}	21.24	21.15	19.55	18.64	17.43	21.25	21.25	31.38	41.70	43.99	1.23	0.66	–	
VW_{1000}	21.61	21.52	20.06	19.15	17.91	21.62	21.62	31.88	42.22	44.51	2.17	1.68	1.09	–

The EIP_{AF} was always included as a fixed effect in the models. Comparisons of models, with the different versions of EIP_{AF}, constituted an indirect (though relevant in the context) way of comparing the connectivity measures. The various EIP_{AF} were also compared using the matrix correlation plot and the concordance correlation coefficient for selected pairs.

4.3.3.3 Data manipulation for variables other than external infestation pressure

The variables of internal infestation pressure (IIP) of AF (IIP_{AF}) and PAAM (IIP_{PAAAM}), baseline abundance of AF ($baseline_{AF}$), topical treatments, and sea surface temperature (hereafter, temperature) remain unchanged from the previous publication (Chapter 3). Here follows a summary of the data manipulations performed for the formulation of these variables.

The abundances were calculated as the normalized counts of AF sea lice divided by the number of fish sampled with the offset of 1 for each observation (i.e., count date). This calculation was repeated for the PAAM. The IIP_{AF} and IIP_{PAAM} were estimated using a triangular weighted 15 day average of AF and PAAM abundances with fixed lags of 21 days for AF and 28 days for PAAM. The baseline abundances of AF were identified for each segment and were defined by the abundance of AF in the first observation within 7 days prior to and including the treatment date.

The topical treatments applied at the start of a segment were assigned to all the observations within that segment. Hydrogen peroxide (Interox® Paramove®

50™, Solvay Chemicals Inc., Belgium, and Aquaparox 50™, Alpha Chemical Limited, Canada) treatments were combined into a single category. Azamethiphos (Salmosan® Vet, Benchmark Animal Health, United Kingdom) treatments were categorized according to their method of application: tarpaulin, skirt, or wellboat. Although the skirt method of application of azamethiphos treatments were scarce and only occurred prior to 2013 these were not excluded from the previously published model (Chapter 3).

The daily estimated temperature values were extracted from the UKMO OSTIA SST product (UK Met Office, 2005) for the duration of the study for the centre points (centroids) of each aquaculture site polygon utilizing the 'raster' package, version 3.0-12 (Hijmans et al., 2020) in R. The spatial resolution was 1/20 degrees, which is approximately 6 km at the 45 degree latitude of the Bay of Fundy, NB; sites that occupied the same cell shared the same temperature values.

4.3.3.4 Depth

Where some connectivity measures rely on a bathymetry map, the depth (m) at the centroids of sites was extracted for direct evaluation, as a fixed effect, in the model. The measurement of depth is an average height from the seafloor to the water surface; the depth is an estimated average due to the variation generated by lunar and solar tides, water currents, winds, and other factors.

4.3.3.5 Modelling

A multilevel mixed-effects linear regression (Pinheiro and Bates, 2000) was used to model the abundance of AF sea lice. Random effects included the site-cycle, cage-cycle, and segments. BMAs were set as a fixed effect. The model was fit with an exponential correlation structure to the errors, stratified by three temperature profiles (low: ≤ 7.0 °C, medium: > 7.0 to ≤ 11.5 °C, and high: > 11.5 °C). The exponential correlation structure was selected to account for non-uniform time gaps between observations. The model assumptions of linearity, homogeneity of variance, and normality of residuals were assessed graphically for each level of the hierarchy. Additional detail and description were published in Chapter 3.

The model was fit with each variation of the EIP_{AF} produced from the various connectivity measures and range of KDE bandwidths (n = 154). These model versions were compared using the Akaike information criterion (AIC).

Depth was introduced as a fixed effect predictor into the original model (SWD_{100}) and a natural log transformation was applied, based on the results from fractional polynomial regression analyses. Depth was also examined as a potential confounder for EIP_{AF}, IIP_{AF} and IIP_{PAAM} in the model fit with each of the variations of the EIP_{AF} at a KDE bandwidth of 10 km (n = 14). An interaction between depth and BMAs was evaluated using the Wald Chi^2 test, as depth was noted to vary per BMAs.

All analyses, unless otherwise specified, were carried out in Stata® v16.1

(StataCorp, 2019), and statistical significance was set at $p < 0.050$.

4.4 Results

4.4.1 Comparison of connectivity measures

The Euclidean distances were calculated for 14 connectivity measures using 62 active sites. This resulted in 91 pairwise comparisons of connectivity measures for a total of 1,953 pairs of sites (Table 4.1). The Euclidean distances (L_2) had a large range from 0.02 (SWD_{25} and TD) to 44.51 (TD_{1000} and VW_{1000}). Certain connectivity measures (SWD_{100}, SA_{250}, TD_{100}, TD_{500}, TD_{1000}, and VW) were selected for their variability and were presented in Figure 4.2 with concordance correlation coefficients for their relevant EIP_{AF}. Figure 4.2 displays the scatter plots of the adjusted count of AF below the diagonal and the scatter plots of the Euclidean distances above the diagonal for the selected connectivity measures.

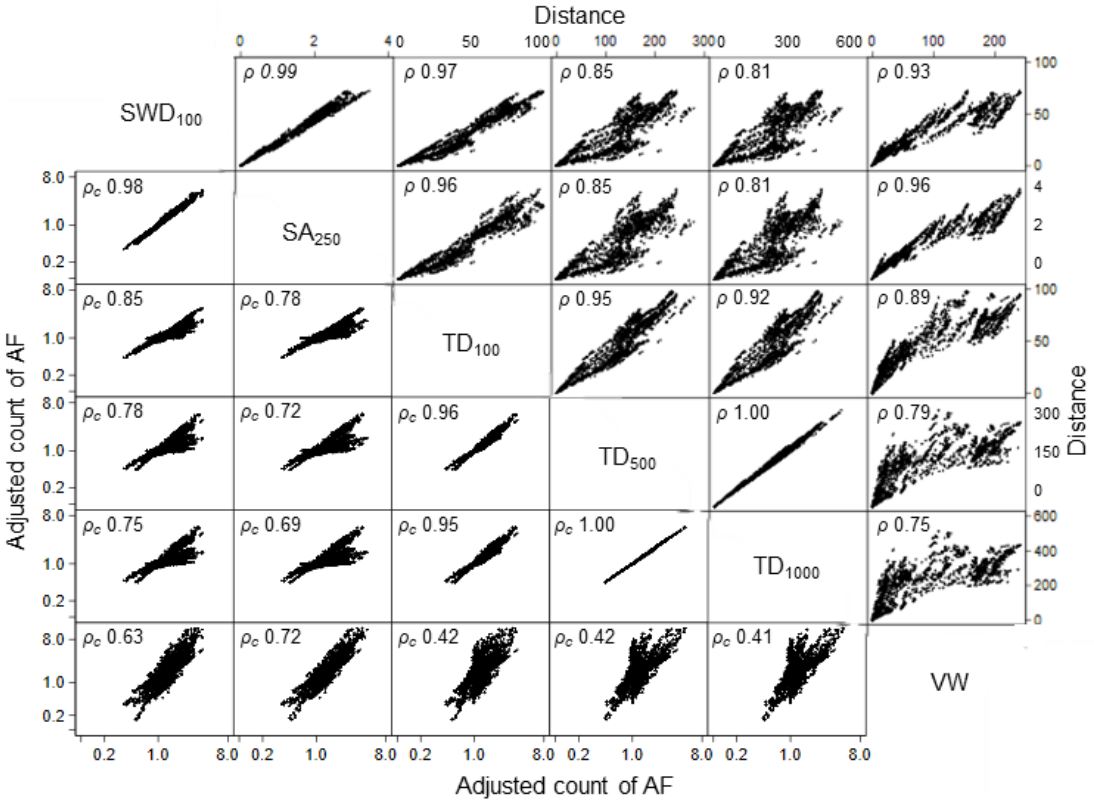

Figure 4.2. Scatter plot matrix of selected connectivity measures directly (above diagonal) and external infestation pressure (EIP_{AF}) predictors as calculated from the connectivity matrices (below diagonal). The values in the top left corner of the plots are the Pearson's correlation (ρ) for pairs of connectivity measures and concordance correlation coefficients (ρ_c) for pairs of EIP_{AF}. SWD_{100}: seaway distance with 100 m resolution; SA_{250}: surface area with a square window area of 250 m; TD_{100}: topographical distance with 100x cost of movement along an incline; TD_{500}: topographical distance with 500x cost of movement along an incline; TD_{1000}: topographical distance with 1,000x cost of movement along an incline; VW: volume of water along seaway path.

SWD_{25} was very similar to SWD_{100} as demonstrated by a small Euclidean distance of 0.34 and perfect concordance correlation of the EIP_{AF} (ρ_c = 1.00). TD and TD_{10} were the same as SWD_{100} with Euclidean distances of 0.12 (EIP_{AF} ρ_c = 1.00). All derivations of SA were similar to SWD_{100} (EIP_{AF} ρ_c SA_{250} = 0.98, SA_{500} = 0.96, and SA_{1000} = 0.93) and Euclidean distance increased slightly with increasing size of the window area (L_2 SA_{250} = 5.61, SA_{500} = 6.00, and SA_{1000} =

6.63). The derivations of VW did not vary against each other (EIP_{AF} ρ_c = 1.00) but were very different to the other connectivity measures (Figure 4.2). Among all pairwise comparisons to VW, VW and SA_{1000} had the shortest Euclidean distance with 16.78, and TD_{1000} and VW_{1000} the largest distance. The adjustment of the cost of movement along an incline for the TD connectivity measure resulted in a large increase in the Euclidean distance compared to SDW_{100} (L_2 TD_{100} = 14.76, TD_{500} = 29.06, and TD_{1000} = 32.15) and decrease in correlation (EIP_{AF} ρ_c TD_{100} = 0.85, TD_{500} = 0.78, and TD_{1000} = 0.75).

4.4.2 Model

The model had 11,658 observations organized into 114 site-production cycles, 1,001 cage-production cycles and 2,398 segments (Chapter 3). The coefficients and confidence intervals for the internal infestation pressure of AF (IIP_{AF}) and PAAM (IIP_{PAAM}), baseline abundance of AF ($baseline_{AF}$), BMAs, temperature, treatments, and temperature-treatment interaction did not vary greatly (results not presented) compared to those previously published (Chapter 3).

The dispersal distance was identified at the nadir of the AIC during comparison of versions of the model with variation of the KDE bandwidths for each of the connectivity measures. SWD_{25} affected a very slight improvement of AIC compared to the original model with SWD_{100} at a KDE bandwidth of 10 km (ΔAIC = 0.40). The nadir of the AIC for the SA derived connectivity measures occurred at a shorter KDE bandwidth compared to the original model (Chapter 3). The SA_{1000} had the shortest estimated dispersal distance at 7 km and improved the AIC of the model by 6.36 units (Figure 4.3A). The smaller square window area for

the calculation of the percent surface area of water (SA_{250} and SA_{500}) had an estimated dispersal distance of 8 km and improved the AIC by 5.27 and 6.63 units, respectively. The AIC and estimated dispersal distance from TD and VW were larger than the values obtained for the original model for the range of KDE bandwidths evaluated (e.g., Figure 4.3B-D for TD_{100}, TD_{500}, and VW, respectively).

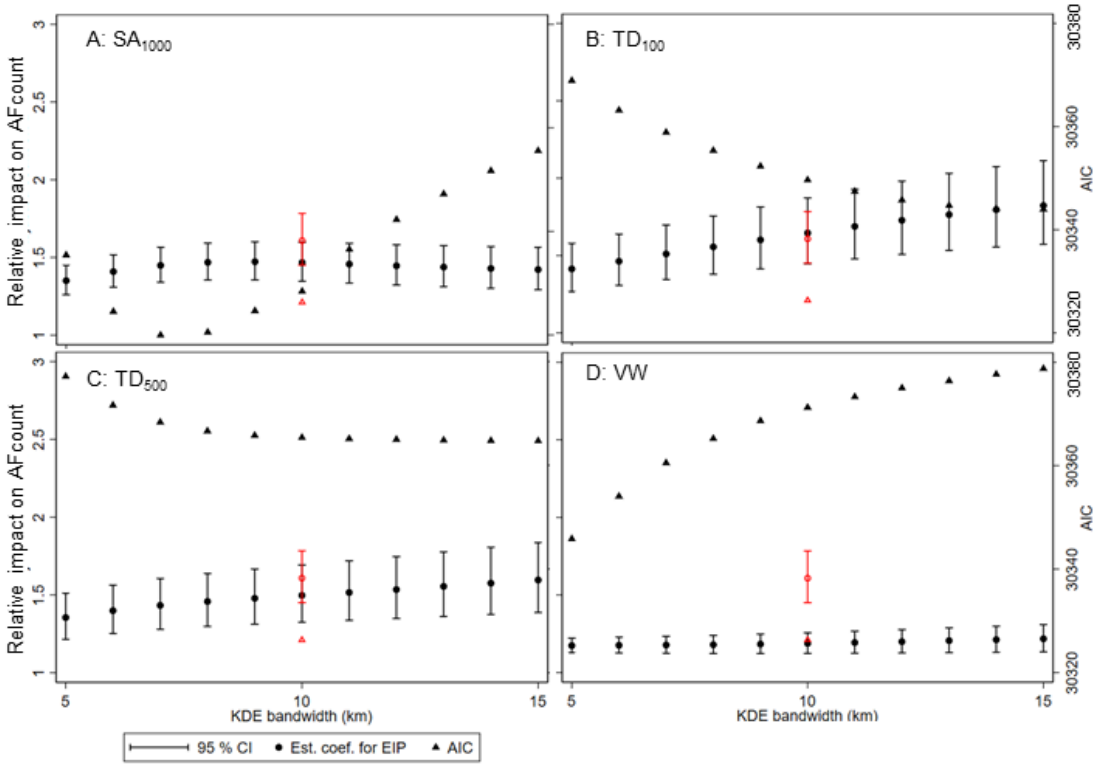

Figure 4.3. Multiplicative regression coefficient for the external infestation pressure (EIP) on abundance of adult female (AF) *Lepeophtheirus salmonis* and 95 % confidence interval (CI) as varied by the Gaussian kernel density estimate (KDE) bandwidth (5-15 km) which was the weight for the calculated EIP. The Akaike's information criterion (AIC) is also reported for the multivariable linear mixed model for the abundance of AF sea lice as varying according to the KDE bandwidth of the EIP. EIP and AIC were calculated using various connectivity measures: A: surface area with a square window area of 1,000 m; B: topographical distance with 100x cost of movement along an incline; C: topographical distance with 500x cost of movement along an incline; and D: volume of water along seaway path. Red markers represent the estimated exponentiated coefficient and 95 % CI for the EIP and the AIC for the KDE bandwidth 10 km reported in Ch. 3 model where weights were seaway distances at 100 m resolution. Scale is the mean normalized count of AF sea lice after treatment (AF count divided by the number of fish sampled counted and offset by 1).

The EIP_{AF} was not a significant predictor in each version of the model. The EIP_{AF} for all derivations of TD at a KDE bandwidth of 10 km were non-significant ($p >$ 0.050). An increase by one unit of the EIP_{AF} using the connectivity measure of SWD_{25} and a KDE bandwidth of 10 km affected the abundance of AF sea lice by

a factor of 1.61 (i.e., a relative increase of 61.3 %) when other predictors remained constant ($p < 0.001$). The EIP_{AF} weighted with a KDE bandwidth of 10 km for the connectivity measures of SA resulted in a relative increase in the abundance of AF sea lice by 56.8 %, 52.2 %, and 47.0 % for SA_{250}, SA_{500}, and SA_{1000}, respectively. At the identified nadir of the AIC for SA_{250}, a one unit increase in the EIP_{AF} resulted in a relative increase of the abundance of AF sea lice by 54.3 % ($p < 0.001$). Similarly, the SA_{500} and SA_{1000} affected a relative increase in the abundance of AF sea lice by 51.2 % and 45.1 %, respectively (both $p < 0.001$). The EIP_{AF} using the other connectivity measures were not summarized here since SWD_{25} and SA derivations were the only versions to improve the fit of the model.

4.4.3 Depth

Depth at sites had a multimodal distribution with a median of 12.7 m, standard deviation (sd) of 7.6 m and range of 1.7 to 41.4 m. The addition of depth as a fixed effect in the model with EIP_{AF} calculated from SWD_{100} resulted in a coefficient of 0.14 and it was a non-significant predictor ($p > 0.050$) and resulted in an increase in the AIC ($\Delta AIC = 91.50$). There was no confounding of depth with the EIP_{AF}, IIP_{AF} and IIP_{PAAM} as calculated with all connectivity measures in their corresponding models (percent change of the EIP_{AF} coefficient was under or equal to 6.3 %, $IIP_{AF} \leq 0.6$ %, and $IIP_{PAAM} \leq 0.4$ %). It appeared that depth varied by the BMAs (data not shown); however, the impact of depth varying by BMA was non-significant (Wald Chi^2 test $p = 0.986$).

4.5 Discussion

4.5.1 New connectivity measures

Seaway distance is a common measure of connectivity among sites (e.g., Kristoffersen et al., 2014, 2018; Elghafghuf et al., 2020). The connectivity measures examined in this study are possible alternatives to seaway distance and possible surrogates to environmental factors that may impact the dispersal of sea lice. The recent construction of a finite-volume ocean model (FVCOM) for the study area (D. Greenberg, unpublished) may be used to study sea lice in the Bay of Fundy, NB, but this was outside the scope of this study.

4.5.2 Connectivity measures

Certain connectivity measures were very similar (SWD_{25}, SA_{250}, SA_{500}, SA_{1000} TD, and TD_{10}) while others were very different (TD_{100}, TD_{500}, TD_{1000}, VW_{250}, VW_{500} and VW_{1000}) to the original connectivity measure of SWD_{100}. The variations found among certain connectivity measures were due to the different methodology and environmental characteristics they represent.

The direct comparison of the connectivity measures demonstrated a larger dispersion of points as compared to the EIP_{AF}, which were an indirect method of comparing the connectivity measures (Figure 4.2). This indicates that the calculations of the EIP_{AF} using the various connectivity measures produced a smoothing effect. The relationship between the EIP_{AF} from SWD_{100} and SA_{250} was linear, which may be in part due to the perfect correlation between seaway distance and least cost path calculations. Other connectivity measures had linear relationships and were similar to each other (TD_{100} and TD_{500}; TD_{500} and TD_{1000}).

Interestingly, the relationship between certain connectivity measures (SWD_{100} and TD_{100}; SA_{250} and TD_{100}; SA_{250} and TD_{500}) diverges into two lines above a certain adjusted count of AF and the cause is unknown. The comparison of the measures directly did not inform the reason for divergence further. Other connectivity measures have matrices that do not demonstrate any clear trends (SWD_{100} and TD_{500}; SWD_{100} and TD_{1000}; SA_{250} and TD_{1000}; and VW with all others).

4.5.3 Impact on the model

4.5.3.1 External infestation pressure

The EIP_{AF} calculated with certain connectivity measures explained some of the variation in the abundance of AF sea lice 3 weeks prior at sites in the Bay of Fundy, NB. The higher grid resolution (25 m vs. 100 m) of seaway distances among sites for the calculation of EIP_{AF} did not improve model fit. Therefore, further analyses using seaway distance can use the less computationally demanding seaway distance at the lower grid resolution of 100 m.

All derivations of the connectivity measure of percent surface area of water had a slightly larger impact on the outcome of the abundance of AF sea lice and appear to be an adequate alternative, and perhaps an improvement on the typical connectivity measure of seaway distance as the calculated AIC was lower using these connectivity measures compared to SWD_{100}. SA_{1000} with a KDE of 7 km resulted in the lowest AIC and a standardized estimated coefficient of 0.118 (95 % confidence interval [CI] 0.093-0.142). SA_{250} and SA_{500} with a KDE of 8 km had

similar standardized estimated coefficients to SA_{1000} (0.117, 95 % CI 0.092-0.141 and 0.118, 95 % CI 0.093-0.143, respectively). Additional research into the connectivity measure of the percent surface area of water and similar connectivity measures may be of interest to further examine the relationship between the percent surface area of water and the effect on sea lice among sites. The connectivity measure of percent surface area of water may be a surrogate for coastal effects on the dispersion of sea lice. Sites that are close to land and each other have lower cumulative percent surface area of water compared to pairs of sites that occur away from land and from each other. A biophysical simulation model of sea lice for Scotland predicted higher sea lice density and prevalence closer to shore compared to sites further away from the coast (Aleynik et al., 2022). There may be two processes occurring where there is lower cumulative percent surface area of water: 1) decreased water flow resulting in lower transmission (decreased EIP) and 2) decreased water flushing resulting in higher transmission of sea lice within the site (increased IIP). Where there are slower current speeds nearshore due to the drag of land as compared to open waters (Shanks, 2009), we wanted to further investigate if low percent surface area of water (i.e., coastal areas) may be associated with impeded dispersal of sea lice. The results of this model allowed for descriptive analysis of this hypothesis in comparison to seaway distance. There was no difference of the standardized residuals between SA_{1000} (mean 0.02, sd 1.08) and SWD_{100} (mean 0.02, sd 1.07) for the BMA 2A, which has many islands and thus lower percent surface area of water among its sites. In this situation, the islands may be

associated with shoals and produce constrictions that increase the flow rate as compared to locations with open water. In BMA 2A, the standardized residuals for VW (mean 0.03, sd 1.08) were slightly larger than the residuals listed above suggesting that depth may be a contributing factor to the observations that sea lice accumulate closer to shore. There may also be other mechanisms in place that affect connectivity (e.g., Siegel et al., 2008) and explain why the percent surface area of water had a slightly greater impact on sea lice than seaway distance. Further research is necessary to explain the relationship between percent surface area of water and dispersion of sea lice; simulations and modelling using biophysical models may provide this insight.

The EIP_{AF} calculated using the connectivity measures of topographic distance and volume of water among sites did not improve model fit; seaway distance explained the variation in sea lice better than these novel connectivity measures. TD_{100} produced the largest standardized estimated coefficient of EIP_{AF} of all the topographic distance connectivity measures (0.097, 95 % CI 0.072-0.122). TD_{1000} had the lowest standardized estimated coefficient of EIP_{AF} at 0.084 (95 % CI 0.058-0.110). The volume of water connectivity measures had EIP_{AF} standardized estimate coefficients between 0.070 and 0.074 for VW_{1000} and VW, respectively. This does not imply that these novel connectivity measures were not useful but may have limited applicability in the examination of the effects of dispersal of sea lice from neighbouring sites.

Topographic distance among sites may not be a useful connectivity measure in the context of this model as the distribution of sea lice in the water column may

not have a strong effect on the dispersal distance in our study area. The smaller effect of topographic distance compared to seaway distance may be due to the complex nature of water movement and effect of depth on the distribution of sea lice. *L. salmonis* remains in the top 0-4 m depth, but that may be affected by factors such as light, pressure and salinity (Frenzl, 2014). *L. salmonis* exhibit a diel cycle with positive phototaxis (Heuch et al., 1995; Penston et al., 2004). Recently, nauplii were found in deeper waters (10-17 m) during the day (Nelson et al., 2017). Aarflot et al. (2019) determined that bathymetry did not constrain the vertical distributions of small zooplankton (captured using a mesh size of 180-1,000 µm and would include the *L. salmonis* nauplii and copepodid of 0.4-0.7 mm in length, if present [Boxaspen, 2006]). Johnsen et al. (2016) found that the vertical distribution of simulated lice depended on the parameter of lice copepodid swimming velocity; slow swimmers (10^{-4} m/s) did not aggregate at the surface of the water column as effectively as rapid swimmers (10^{-3} m/s). Depth at sites represent a simplification of bathymetry, and this predictor was not significant in the model using EIP_{AF} calculated with $SWD_{100,}$ and neither was it a statistical confounder for the EIP_{AF} calculated with any of the connectivity measures. Topographic distance can be reconsidered as a connectivity measure during analyses of genetic variability to examine dispersion as suggested by Cantrell et al. (2020) as it has been demonstrated to be effective for multiple species on land (e.g., Goldberg and Waits, 2010).

The volume of water among sites did not have lower AIC, compared to seaway distance, as the volume which contains sea lice may be constant among sites. It

remains that this is an interesting, proposed connectivity measure as it can be considered as a surrogate to dilution of planktonic sea lice where larger volumes of water may impede successful dispersal from a site to another. It may be that this surrogate measure does not capture the variability of tides and ocean mixing that affect dispersal of sea lice (Harrington et al., 2022). Tides and ocean mixing affect turbulence in the water column among many other variables, and higher levels of turbulence can reduce infectivity by dispersing concentrations of sea lice, and possibly increase the probability of convergence of sea lice with the host (i.e., salmon) and possible mates (i.e., mobile lice attached to salmon [Tanaka et al., 2021]). In the biophysical simulation by Johnsen et al. (2016), sea lice particles were distributed at higher densities along land where the particles swam vertically until a maximum turbulent vertical velocity was reached (5×10^{-4} m/s) at which point the simulated lice sank. Tanaka (2019) noted turbulence avoidance behaviour in zooplankton that differed by feeding mode. In general, a higher turbulence intensity resulted in a decrease of the zooplankton observed in the top 0-10 m of the water column (Tanaka, 2019). Turbulence avoidance behaviour included instantaneous escape jumps of greater than 10 cm/s that were estimated to be sufficient to escape areas of high turbulence for calmer waters (Tanaka, 2019). The literature on the motility of sea lice in relation to turbulence is limited; the copepodid *Lepeophtheirus pectoralis* (Müller O.F. [WoRMS, 2022]) displayed increased activity when exposed to water turbulence, but the short-burst swimming was not quantified (Bron et al., 1994). In *L. salmonis,* swim bursts of greater than 15 cm were demonstrated in response to increased

pressure (Bron et al., 1994). Furthermore, the feeding modes described in the study did not include lecithotrophic zooplankton (life stage that depends solely on the yolk for sustenance). This paucity of information therefore highlights an area for basic research that may aid in further studies that employ the concept of infestation pressure or biophysical models.

The impact of the EIP_{AF} for each of these connectivity measures was smaller compared to the internal infestation pressures (standardized estimated coefficients IIP_{AF}: 0.164, 95 % CI 0.136-0.191 and IIP_{PAAM}: 0.075, 95 % CI 0.055-0.096); similar effects were observed in the original model for SWD_{100} (Chapter 3). The larger impact of the IIP compared to EIP is multifactorial (Adams et al., 2012; Aldrin et al., 2013). Harrington et al. (2022) proposed that local currents may play a role in maturation of the sea lice within and around a site. These authors also found that waterways driven by diffusion (mixing due to winds and tidal flow) rather than advection (general seaward flow due to river output), such as the study area in the Bay of Fundy, NB, have a lower probability of sea lice movement among sites (as driven by advection) and have a greater probability of infestation when arrival does occur (as driven by diffusion). The conclusion remains that management of sea lice must occur at the site first and consider sea lice transmission to the site from neighbouring sites.

The evaluation of the multiple connective measures through the estimation of EIP highlighted that the contribution of the AF abundance to the estimated EIP is small because of the weight of the connectivity measure adjusted by KDE bandwidth. The weight of the KDE connectivity measure dampens the AF counts

severely. The calculated value of the EIP_{AF} of SWD_{100} at KDE 10 km was on average 0.7 % (95 % CI 0.18-1.24 %) compared to the calculated AF abundance contributed from neighbouring sites. The adjustment of the KDE bandwidth and the connectivity measure affected a small change in the dampening effect. The EIP_{AF} therefore is strongly influenced by the weight of the connectivity measure and the bandwidth and likely affects the significance of this predictor in the model.

4.5.3.2 Dispersal distance

The dispersal distances obtained with the EIP_{AF} estimated using the percent surface area of water were shorter than previously noted at 10 km for the Bay of Fundy, NB, and 12.5 km for Grand Manan using seaway distance (Chapter 3 and Elghafghuf et al., 2020, respectively). These shorter dispersal distances were closer to an extrapolated 8 km based on approximate mean flow rates of 10 cm/s around sites (Nelson et al., 2017). This supports the previous conclusion that percent surface area of water performs adequately and maybe even better than seaway distance at explaining the variation in AF sea lice abundance. However, these versions of dispersal distance do not point to one being more important than another as it is a large area, and some connectivity measures may represent dispersal better than others in certain areas.

The results of this data-driven iterative process and multilevel mixed-effects linear regression model were that dispersal distance for *L. salmonis* in the Bay of Fundy, NB, was 7-10 km as estimated using percentage surface area (7 km) and

seaway distance (10 km). These results suggest that when sites are located at distances under 7-10 km apart, the EIP likely affect sea lice management at a site. For sites located at distances greater than 10 km, the impact of the EIP is likely very small and therefore contributing very little to the infestation of a neighbouring site. These results are based on the estimation of EIP for AF sea lice abundance weighted by a connectivity measure adjusted by a Gaussian KDE bandwidth. The KDE bandwidth implies that the ability to fit the data is best at the bandwidth in the model that resulted in the lowest AIC; for example, capturing all sites within 10 km seaway distance resulted in a model with lower AIC compared to capturing all sites at 9 km. The other aspect to consider is the Gaussian distribution which was selected for the KDE calculation. This assumption may not fit for all circumstances as some close neighbours (which using Gaussian KDE implies higher impact, larger EIP) may not affect each other due to high flushing rates.

The dispersal distance estimates of 7-10 km support the use of area-wide management to mitigate the effect of sea lice transmission from neighbouring sites. The aquaculture industry of the Bay of Fundy, NB, have practiced area-wide management using the current form of the BMAs since 2006. Within the current structure of the BMAs, there are multiple sites located within seaway distances under 10 km. While this 10 km cannot be considered as a cut-point for decision-making, it signifies that there is a larger likelihood of impact among neighbouring farms at that distance. It can be suggested that the BMA structure be revisited to create new groupings that can aid in the management of sea lice.

Clinical evidence points to the overlap of transmission of sea lice among BMAs as currently occurring (L. Hawkins, pers. comm.). A new BMA structure should take into consideration the estimated dispersal distance and apply the concept of the mitigation strategy to ‘disconnect’ sea lice populations (Samsing et al., 2017, 2019); in practice this would involve creating clusters of sites and removing intermediary sites that connect between two clusters.

4.5.4 Study design and limitations

The estimation of the EIP_{AF} and dispersal distance vary according to the study area and methods employed (e.g., Murray et al., 2011; Aldrin et al., 2013, 2017, 2019). This study used the same model and dataset to examine the effects of various connectivity measures; therefore, the effects of differing environments and modelling approaches do not apply.

The comparison of connectivity measures through the EIP_{AF} was limited; therefore, more direct comparison was achieved using Euclidean distance. The direct comparison demonstrated differences among the connectivity measures which may indicate that estimates of EIP_{AF} using the various connectivity measures have minimal nominal value. However, this limitation does not obscure the finding that the EIP_{AF} is often a significant contributor to the infestation of sites and IIP is greater than EIP on average for the study area for 2009-2018.

Another limitation to the use of multiple connectivity measures is their relevance to different spatial scales. This means that the use of connectivity measures other than seaway distance are less intuitive. Research and results from new

connectivity measures would require a calibration with a measure that is currently understood and utilized.

The connectivity measures were symmetrical for sites in the Bay of Fundy, NB; in other words, the strength of the connectivity was the same for the direction of site ‘A’ to ‘B’ and ‘B’ to ‘A.’ The assumption of symmetrical connectivity among sites is currently employed elsewhere (e.g., Aldrin et al., 2017; Elghafghuf et al., 2020). However, studies that utilized hydrodynamic models (e.g., Cantrell et al., 2018) demonstrated that the strength of the connection between sites is not necessarily symmetrical. A hydrodynamic model for the area was not available at the time of analysis and will likely support asymmetrical connectivity among sites. The result of this assumption on the effects of the external infestation pressure is unknown; further evaluation is necessary with the advent of a hydrodynamic model for the Bay of Fundy, NB.

The dispersal success of sea lice is based on the statistical approach and definitions of the study. In our study the sea lice on salmon were detected by a sea lice counter and the successful parasitism event was defined in a linear mixed-effects model as the abundance of AF and PAAM lice counted at the site (IIP_{AF} and IIP_{PAAM}) and the sum of the abundance of AF sea lice per fish from all neighbouring sites within a KDE-adjusted connectivity measure (EIP_{AF}). The probability of the dispersal success event for sea lice from each potential source (i.e., neighbouring sites or wild fish) is unknown because it is dependent on the ability of the counter to identify the sea lice and count them (e.g., Gautam et al., 2016) and the capacity for the counted sample to accurately measure the

population level of lice. Both factors contribute to the probability of detection, which is unknown in the aquaculture sites of the Bay of Fundy, NB. The probability of detection was estimated at 0.78 copepodid for an area of 5,760 km^2 around a site at Norwegian sites using a combination of sea lice counts at sentinel cages and a biophysical model (Sandvik et al., 2016). The capacity for the counted sample to accurately measure the population of sea lice is influenced by the sea lice's passive and limited active movement in the water, their ageing and mortality processes, and their proficiency at identifying and attaching to the host. Our study's data-driven approach based on the Fish-iTrends data, indirectly accounts for the unsuccessful dispersal of sea lice as these lice were not counted. Our approach has the advantage that it accounts for the variability in lice counts and certain environmental conditions that are ignored in some biophysical models (e.g., Aleynik et al., 2022) and avoids the paradox of zero connectivity in single habitat patch environments that may occur with methods that rely on particle-tracking (Tischendorf and Fahrig, 2000). The major disadvantage of this approach is that the success of dispersal cannot be estimated as can be achieved in biophysical models where a set concentration of sea lice is released as particles that are assigned parameters of ageing and mortality rates (constant rate, e.g., Sandvik et al., 2016 and variable rate, e.g., Revie et al., 2005).

The fixed effect predictor of depth at sites may not be an adequate representation of depth as an environmental variable of interest. The depth at a site was estimated at its centroid location according to a 2017 geo-referenced

published map of sites in the Bay of Fundy, NB (Department of Energy and Resource Development/Agriculture, 2017). The range of depths among sites was large and the depth at a site may vary for its entire area; however, we do not have access to the geo-referenced coordinates for all the cages that make up the sites. Another challenge was the closure of certain sites during the study period that make it more difficult to obtain coordinates for a retrospective study such as this. Other methods of estimating depth at sites may provide more insight into the relationship between depth and sea lice abundance at the site-level.

4.6 Conclusions

Different connectivity measures for the dispersal of *L. salmonis* were compared directly and indirectly as a component of the EIP during the modelling of the abundance of the AF sea lice. Seaway distance was a previously explored connectivity measure that served as a comparison for the novel connectivity measure of percent surface area of water among sites that performed slightly better as evaluated using AIC. The percent surface area of water among sites should be further explored as it may explain part of the transmission of sea lice in the Bay of Fundy, NB.

The dispersal distance of *L. salmonis* was estimated to 7-10 km using the optimal fit of the model with the EIP calculated with different connectivity measures. This range of distances may be of importance to the management of sea lice in the Bay of Fundy, NB, to decrease the transmission of sea lice from neighbouring sites. In practice, this may be achieved through the creation of clusters of sites with the removal of intermediary sites that connect two clusters of sites.

4.7 References

Aarflot, J. M., Aksnes, D. L., Opdal, A. F., Skjoldal, H. R., and Fiksen, Ø. (2019). Caught in broad daylight: Topographic constraints of zooplankton depth distributions. Limnology and Oceanography, 64(3), 849–859. https://doi.org/10.1002/lno.11079

Adams, T., Black, K., Macintyre, C., Macintyre, I., and Dean, R. (2012). Connectivity modelling and network analysis of sea lice infection in Loch Fyne, west coast of Scotland. Aquaculture Environment Interactions, 3, 51–63. https://doi.org/10.3354/aei00052

Adams, T. P., Proud, R., and Black, K. D. (2015). Connected networks of sea lice populations: Dynamics and implications for control. Aquaculture Environment Interactions, 6(3), 273–284. https://doi.org/10.3354/aei00133

Aleynik, D., Adams, T., Davidson, K. (2022). Optimizing the connectivity of salmon farms: Role of exposure to wind, tides, and isolation. In Md. N. Islam, S. M. Bartell (Eds.), Global Blue Economy: Analysis, Developments, and Challenges (1st ed., pp. 61–86). CRC Press. https://doi.org/10.1201/9781003184287-3

Aldrin, M., Huseby, R. B., Stien, A., Grøntvedt, R. N., Viljugrein, H., and Jansen, P. A. (2017). A stage-structured Bayesian hierarchical model for sea lice populations at individual salmon farms ― Estimated from multiple farm data sets. Ecological Modelling, 359, 333–348.

Aldrin, M., Storvik, B., Kristoffersen, A. B., and Jansen, P. A. (2013). Space-time modelling of the spread of sea lice between and within Norwegian marine salmon farms. PLoS ONE, 8, 1–10. https://doi.org/10.1371/journal.pone.0064039

Aldrin, M., Jansen, P. A., and Stryhn, H. (2019). A partly stage-structured model for the abundance of sea lice in salmonid farms. Epidemics, 26, 9–22. https://doi.org/10.1016/j.epidem.2018.08.001

Asplin, L., Johnsen, I. A., Sandvik, A. D., Albretsen, J., Sundfjord, V., Aure, J., and Boxaspen, K. K. (2014). Dispersion of salmon lice in the Hardangerfjord. Marine Biology Research, 10(3), 216–225. https://doi.org/10.1080/17451000.2013.810755

Bista, D., Baxter, G. S., Hudson, N. J., Lama, S. T., Weerman, J., and Murray, P. J. (2021). Movement and dispersal of a habitat specialist in human-dominated landscapes: A case study of the red panda. Movement Ecology, 9(1), 1–15. https://doi.org/10.1186/s40462-021-00297-z

Boxaspen, K. (2006). A review of the biology and genetics of sea lice. ICES Journal of Marine Science, 63(7), 1304–1316. https://doi.org/10.1016/j.icesjms.2006.04.017

Bron, J. E., Sommerville, C., and Rae, G. H. (1994). Aspects of the behaviour of copepodid larvae of the salmon louse *Lepeophtheirus salmonis* (Krøyer, 1837). In G. A. Boxshall and D. Defaye (Eds.), Pathogens of Wild and Farmed Fish: Sea Lice (pp. 125–142). Chichester, West Sussex: Ellis Horwood.

Cantrell, D. L., Groner, M. L., Ben-Horin, T., Grant, J., and Revie, C. W. (2020). Modeling pathogen dispersal in marine fish and shellfish. Trends in Parasitology, 36(3), 239–249. https://doi.org/10.1016/j.pt.2019.12.013

Cantrell, D. L., Rees, E. E., Vanderstichel, R., Grant, J., Filgueira, R., and Revie, C. W. (2018). The use of kernel density estimation with a bio-physical model provides a method to quantify connectivity among salmon farms: Spatial planning and management with epidemiological relevance. Frontiers in Veterinary Science, 5(Oct), 1–14. https://doi.org/10.3389/fvets.2018.00269

Cantrell, D., Vanderstichel, R., Filgueira, R., Grant, J., and Revie, C. W. (2021). Validation of a sea lice dispersal model: principles from ecological agent-based models applied to aquatic epidemiology. Aquaculture Environment Interactions, 13, 65–79. https://doi.org/10.3354/AEI00390

Cowen, R. K., and Sponaugle, S. (2009). Larval dispersal and marine population connectivity. Annual Review of Marine Science, 1, 443–466. https://doi.org/10.1146/annurev.marine.010908.163757

Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). Bay of Fundy Marine Aquaculture Sites [map]. 1:125000. New Brunswick, Canada.

Eisenhauer, L., Solvang, T., Alver, M., Krause, D. F., and Hagemann, A. (2020). Dispersal of salmon lice (*Lepeophtheirus salmonis* Krøyer, 1837) egg strings from open-cage salmon farming: a neglected source for infestation dynamics. Aquaculture Research, 51, 4595–4601. https://doi.org/10.1111/are.14805

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394Fast 2014

[dataset] Fish-iTrends data management system for farm sea lice counts, program owned and operated by CAHS, AVC, UPEI, Charlottetown, Canada; data submitted and owned by private fish farm companies. Queried on [2022-06-30]

Frenzl, B., Stien, L. H., Cockerill, D., Oppedal, F., Richards, R. H., Shinn, A. P., … Migaud, H. (2014). Manipulation of farmed Atlantic salmon swimming behaviour through the adjustment of lighting and feeding regimes as a tool for salmon lice control. Aquaculture. https://doi.org/10.1016/j.aquaculture.2013.12.012

Gautam, R., Boerlage, A. S., Vanderstichel, R., Revie, C. W., and Hammell, K. L. (2016). Variation in pre-treatment count lead time and its effect on baseline estimates of cage-level sea lice abundance. Journal of Fish Diseases, 39(11), 1297–1303. https://doi.org/10.1111/jfd.12460

Girvetz, E. H., and Greco, S. E. (2007). How to define a patch: A spatial model for hierarchically delineating organism-specific habitat patches. Landscape Ecology, 22(8), 1131–1142. https://doi.org/10.1007/s10980-007-9104-8

Goldberg, C. S., and Waits, L. P. (2010). Comparative landscape genetics of two pond-breeding amphibian species in a highly modified agricultural landscape. Molecular Ecology, 19(17), 3650–3663. https://doi.org/10.1111/j.1365-294X.2010.04673.x

Harrington, P. D., Cantrell, D. L., Foreman, M. G. G., Guo, M., and Lewis, M. A. (2022). Calculating the timing and probability of arrival for sea lice dispersing between salmon farms, 1–32.

Heuch, P. A., Parsons, A., and Boxaspen, K. (1995). Diel vertical migration ─ A possible host-finding mechanism in salmon louse (*Lepeophtheirus salmonis*) copepodids. Canadian Journal of Fisheries and Aquatic Sciences, 52(4), 681–689. https://doi.org/10.1139/f95-069

Hijmans, R.J., van Etten, J., Sumner, M., Cheng, J., Baston, D., Bevan, A., … Wueest, R. (2020). raster: Geographic Data Analysis and Modeling. R package version 3.0-12. https://CRAN.R-project.org/package=raster

Hogans, W. E. (1995). Infection dynamics of sea lice, *Lepeophtheirus salmonis* (Copepoda: Caligidae) parasitic on Atlantic salmon (*Salmo salar*) cultured in marine waters of the lower Bay of Fundy (Vol. 2067). St. Andrews, NB.

Johnsen, I. A., Asplin, L. C., Sandvik, A. D., and Llinares, R. M. S. (2016). Salmon lice dispersion in a northern Norwegian fjord system and the impact of vertical movements. Aquaculture Environment Interactions, 8, 99–116. https://doi.org/10.3354/aei00162

Kristoffersen, A. B., Jimenez, D., Viljugrein, H., Grøntvedt, R., Stien, A., and Jansen, P. A. (2014). Large scale modelling of salmon lice (*Lepeophtheirus salmonis*) infection pressure based on lice monitoring data from Norwegian salmonid farms. Epidemics, 9, 31–39. https://doi.org/10.1016/j.epidem.2014.09.007

Kristoffersen, A. B., Qviller, L., Helgesen, K. O., Vollset, K. W., Viljugrein, H., and Jansen, P. A. (2018). Quantitative risk assessment of salmon louse-induced mortality of seaward-migrating post-smolt Atlantic salmon. Epidemics, 23, 19–33. https://doi.org/10.1016/j.epidem.2017.11.001

Kristoffersen, A. B., Rees, E. E., Stryhn, H., Ibarra, R., Campisto, J. L., Revie, C. W., and St-Hilaire, S. (2013). Understanding sources of sea lice for salmon farms

in Chile. Preventive Veterinary Medicine, 111(1–2), 165–175. https://doi.org/10.1016/j.prevetmed.2013.03.015

Le Corre, N., Guichard, F., and E. Johnson, L. (2012). Connectivity as a Management Tool for Coastal Ecosystems in Changing Oceans. In M. Marcelli (Ed.), Oceanography (pp. 235–258). IntechOpen. https://doi.org/10.5772/27704

Manly, B. F. J. (2004). Measuring and testing multivariate distances. In Multivariate Statistical Methods: A Primer (3rd ed., pp. 59–74). Florida, USA: Chapman & Hall/CRC Press.

Murray, A. G., Amundrud, T. L., Penston, M. J., Pert, C. C., and Middlemas, S. J. (2011). Abundance and Distribution of Larval Sea Lice in Scottish Coastal Waters. In S. Jones and R. Beamish (Eds.), Sea lice: An Integrated Approach to Understanding Parasite Abundance and Distribution (1st ed., p. 726). West Sussex, UK: John Wiley & Sons, Inc.

Nelson, E. J., Robinson, S. M. C., Feindel, N., Sterling, A., Byrne, A., and Pee Ang, K. (2017). Horizontal and vertical distribution of sea lice larvae (*Lepeophtheirus salmonis*) in and around salmon farms in the Bay of Fundy, Canada. Journal of Fish Diseases, 41, 885–899. https://doi.org/10.1111/jfd.12692

Overton, K., Dempster, T., Oppedal, F., Kristiansen, T. S., Gismervik, K., and Stien, L. H. (2019). Sea lice treatments and salmon mortality in Norwegian aquaculture: a review. Reviews in Aquaculture, 11, 1398–1417. https://doi.org/10.1111/raq.12299

Penston, M. J., Mckibben, M. A., Hay, D. W., and Gillibrand, P. A. (2004). Observations on open-water densities of sea lice larvae in Loch Shieldaig, Western Scotland. Aquaculture Research, 35, 793–805. https://doi.org/10.1111/j.1365-2109.2004.01102.x

Pinheiro, J. C., and Bates, D. M. (2000). Mixed-Effects Models in S and S-PLUS. (J. Chambers, W. Eddy, W. Hardle, S. Sheather, and L. Tierney, Eds.), Statistics and Computing (1st ed.). New Y: Springer-Verlag New York, Inc.

QGIS Development Team. (2018). QGIS Geographic Information System. Open Source Geospatial Foundation Project. Retrieved from http://qgis.osgeo.org

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

Revie, C.W., Robbins, C., Gettinby, G., Kelly, L., Treasurer, J.W., 2005. A mathematical model of the growth of sea lice, *Lepeophtheirus salmonis*, populations on farmed Atlantic salmon, *Salmo salar* L., in Scotland and its use in the assessment of treatment strategies. J. Fish Dis. 28, 603–614. https://doi.org/10.1111/j.1365-2761.2005.00665.x

Salama, N. K. G., and Rabe, B. (2013). Developing models for investigating the environmental transmission of disease-causing agents within open-cage salmon aquaculture. Aquaculture Environment Interactions, 4(2), 91–115. https://doi.org/10.3354/aei00077

Samsing, F., Johnsen, I., Dempster, T., Oppedal, F., and Treml, E. A. (2017). Network analysis reveals strong seasonality in the dispersal of a marine parasite and identifies areas for coordinated management. Landscape Ecology, 32(10), 1953–1967. https://doi.org/10.1007/s10980-017-0557-0

Sandvik, A. D., Bjørn, P. A., Ådlandsvik, B., Asplin, L., Skardhamar, J., Johnsen, I. A., … Skogen, M. D. (2016). Toward a model-based prediction system for salmon lice infestation pressure. Aquaculture Environment Interactions, 8, 527–542. https://doi.org/10.3354/AEI00193

Shanks, A. L. (2009). Pelagic larval duration and dispersal distance revisited. Biological Bulletin, 216(3), 373–385. https://doi.org/10.2307/25548167

Shanks, A. L., Grantham, B. A., and Carr, M. H. (2003). Propagule dispersal distance and the size and spacing of marine reserves. Ecological Applications, 13(1), s159–s169.

Siegel, D. A., Kinlan, B. P., Gaylord, B., and Gaines, S. D. (2003). Lagrangian descriptions of marine larval dispersion. Marine Ecology Progress Series, 260, 83–96. https://doi.org/10.3354/meps260083

Siegel, D. A., Mitarai, S., Costello, C. J., Gaines, S. D., Kendall, B. E., Warner, R. R., and Winters, K. B. (2008). The stochastic nature of larval connectivity among nearshore marine populations. Proceedings of the National Academy of Sciences of the United States of America, 105(26), 8974–8979. https://doi.org/10.1073/pnas.0802544105

StataCorp (2019). Stata: Release 16.1. Statistical Software. StataCorp LLC, TX, USA

Statistics Canada; Government of Canada. (2020). Table 32-10-0107-01 Aquaculture, production and value. https://doi.org/10.25318/3210010701-eng

Tanaka, M. (2019). Changes in vertical distribution of zooplankton under wind-induced turbulence: A 36-year record. Fluids, 4(4). https://doi.org/10.3390/fluids4040195

Tanaka, M., Genin, A., Endo, Y., Ivey, G. N., and Yamazaki, H. (2021). The potential role of turbulence in modulating the migration of demersal zooplankton. Limnology and Oceanography, 66, 855–864. https://doi.org/10.1002/lno.11646

Tischendorf, L., and Fahrig, L. (2000). On the usage and measurement of landscape connectivity. Oikos, 90(1), 7–19.

[dataset] UK Met Office (2005). OSTIA Level 4 Sea Surface Temperature Analysis. Ver. 1.0. PO.DAAC, CA, USA. Retrieved from https://doi.org/10.5067/GHOST-4FK01 [2020-03-21].

van Etten, J., and de Sousa, K. (2012). gdistance: Distances and Routes on Geographical Grids. R package version 1.3-1. https://CRAN.R-project.org/package=gdistance

Wang, I. J., Savage, W. K., and Bradley Shaffer, H. (2009). Landscape genetics and least-cost path analysis reveal unexpected dispersal routes in the California tiger salamander (*Ambystoma californiense*). Molecular Ecology, 18(7), 1365–1374. https://doi.org/10.1111/j.1365-294X.2009.04122.x

Wang, I. (2019). topoDistance: Calculating Topographic Paths and Distances. R package version 1.0.1. https://cran.r-project.org/package=topoDistance

WoRMS Editorial Board. (2022). World Register of Marine Species. VLIZ. doi:10.14284/17

4.8 Supplemental information

4.8.1 External infestation pressure

Sea lice are parasitic copepods affecting wild and cultured salmon. They disperse through water transport among aquaculture sites via larval stages; the connectivity measure is an estimate of the dispersal distance of these larval stages among sites. The seaway distance between two sites is the more commonly used method to capture connectivity measures among sites, which supersedes the more primitive Euclidean distance measure. In addition, the study in Chapter 4 presented alternatives to using only seaway distances to describe connectivity measure, some of these used coastal water depth (bathymetry) data in the calculations. More specifically, three connectivity measures were proposed: 1) topographical distance, 2) percent surface area of water and 3) volume of water. The derivation of these connectivity measures used data from 63 Atlantic salmon farm site locations in the Bay of Fundy, New Brunswick (NB), Canada, and geographical characteristics of the coastal water body (and depths) in the study area. The connectivity measures were also directly compared using Euclidean distance. The Euclidean distance used for the comparison of the connectivity measures (as further described in section 2) should not be confused with the Euclidean distance among a pair of sites.

The external infestation pressure (EIP) equation was presented in Chapter 3 and used again in Chapter 4. The calculation uses a temporal weight and a spatial weight to adjust the abundance of sea lice, as recorded and made available in the Fish-iTrends database. The spatial weight adjusts for the dispersal of sea lice

from neighbouring farms (i.e., sites) to a site according to a connectivity measure, following a decaying function (Gaussian kernel). The abundance of adult female (AF) sea lice obtained from the Fish-iTrends database are multiplied by these weights and summed across all connections with neighbours for a site.

4.8.1.1 Generation of temporal weights

The 3-week lagged abundance of AF lice is an average across a 15-day window ([13.5, 28.5] days prior to an observation) using a triangular distribution. The decision for the abundance of AF lice 3 prior to an observation was described in Chapter 3. The width of the temporal window was determined during a small sensitivity analysis (additional details in Chapter 3). The date 3 weeks prior to an observation was identified and, if present, this abundance of AF lice was assigned the highest weight. Abundances 7 days prior and 7 days after this date were assigned decreasing weights toward the average abundance. The abundances of AF lice were multiplied by the weights and summed, then divided by the sum of the weights to normalize these values (Eq. 4.1)

$$\overline{lnAF}_{it} = \sum_{c;s\in\Theta_t} w(s,t)\, lnAF_{ic}(s) \Big/ \sum_{c;s\in\Theta_t} w(s,t) \qquad \text{Eq. 4.1}$$

Where: logarithmic abundances were obtained as $lnAF = ln\left({(AF+1)}\Big/{no.fish}\right)$

c ~ cage within site i

t ~ target date (day) typically 3 weeks prior to an observation

s ~ other times in time window $\theta_t = (t - 7.5, t + 7.5)$

$w(s,t)$ weight for time *s* at time *t* (based on time distance between *s* and *t*); triangular distribution weights $w(s,t) = {(7.5 - |s-t|)}/{56.25}$

Note: In the expression, it was understood that the summations are over cages c within sites i and at times s where $lnAF_{ic}(s)$ exists.

4.7.1.2 Generation of spatial weights

The EIP was calculated as the sum of the mean AF abundances as calculated previously (Eq. 4.1) and adjusted with a spatial weight. The spatial weight represents the relationship between sites and their neighbours as evaluated using the different connectivity measures. The connectivity measures were calculated according to the equations below (Eq. 4.2 to 4.7).

The generation of spatial weights utilized raster images which are two-dimensional graphical representations of spatial information along a grid of square cells with a defined constant dimension (e.g., 25 m, so each cell is 25 x 25 m). The cells themselves contain data; for example, a waterway map could have a constant value of 1 for grid cells which are mostly within coastal waters and missing values for those within land (Figure 4.4). Raster images could also store bathymetry data, assigned as average depth (m) within each cell.

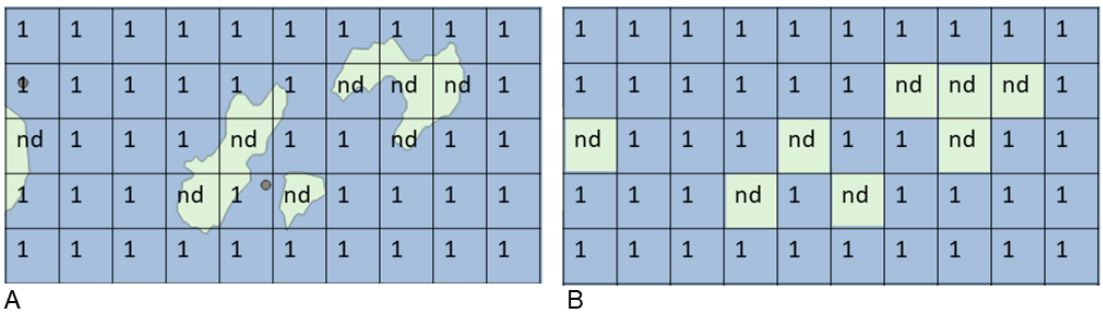

Figure 4.4. Example of a grid overlayed over a map of the islands east of Grand Manan Island, New Brunswick, where cells have an approximate width and height of 25 m and are assigned values of 1 for coastal waters and 'No Data' (nd) for land. The resulting raster image of the waterway map for the same area (B).

4.7.1.2.1 Site locations

Approximate centroids of sites were manually generated from a raster image of the 2017 published map (PDF format) of sites in the Bay of Fundy, NB (Department of Energy and Resource Development/Agriculture, 2017). This raster image was geo-referenced using landmarks between the image and Google Maps and was projected to the NAD83(CSRS)/New Brunswick Stereographic (EPSG:2953) projection in QGIS (QGIS.org, 2018).

4.7.1.2.2 Depth/elevation base map

The connectivity measures of topographical distance and volume of water along seaway path utilized bathymetry data obtained from D. Greenberg and the Department of Fisheries and Oceans Canada (DFO). The bathymetry dataset was originally generated for a finite-volume ocean model (FVCOM, D. Greenberg, unpublished), and was provided as depths for 110,099 unstructured point locations in the Bay of Fundy, NB. Depths were linearly interpolated onto a 25 x 25 m grid which overlayed the study area using the ‘interp’ function of the ‘akima’ package (version 0.6-3.4 Akima and Gebhardt, 2021) in R (R Core Team, 2020). The depth measurements represented an average distance from the seafloor to the water surface; depth is an estimated average due to the variation generated by lunar and solar tides, water currents, winds, and other factors. The projection of the bathymetry map for the study area was converted to the NAD83(CSRS)/New Brunswick Stereographic projection (EPSG:2953) in QGIS. Depths for aquaculture sites were extracted from the bathymetry raster dataset to

represent average depth of water for each site by using the ‘extent’ function and a vector of length 4 along the *x* and *y* axes.

4.7.1.2.3 Waterway base map

Using the same grid dimensions from the depth raster layer (25 m) a waterway map was created by clipping out land boundaries (Administrative area; GADM, 2018) within a large regional bounding box (polygon) and assigning a constant value of 1 for those grid cells mostly within coastal waters (land values were stored as 'No Data').

The R commands below can be implemented:

```
# Bathymetry raster (negative = deeper)
bath_25m<-raster("Bathymetry - 25m - NBProj.tif")
elev_r<-(bath_r)*-1

# Land Boundary
land_poly <-readOGR("Land Boundaries - NBProj.shp")

# Water boundary
water_poly <-readOGR("Waterboundaries - NBProj.shp")

# Crop around all NB sites (2445000, 2527000, 7280000, 7350000)
bb <- extent(2445000, 2527000, 7280000, 7353000)
bath_NB <- crop(bath_r, bb)

# Farm locations from QGIS
farms <-readOGR("NB Farms - points_joined - MAP 2013 - NBProj.shp")

# Correct inter-cell conductance values
# rp is the raster of the water boundary
tr<-transition(rp,transitionFunction=mean,directions=8,symm=TRUE)
trC<-geoCorrection(tr,type="c",multpl=FALSE)

# Create matrices of sites
sites_coords<-as.data.frame(sites)
site.mat=data.matrix(sites_coords[,c("coords.x1","coords.x2")])

# Create raster with values of 1 for water, and Land = NA
```

```
# Bathymetry raster
r_bath_NB <- mask(bath_NB, water_poly)
water_r <- r_bath_NB
water_r[water_r>=-1000]= 1

# Create matrix of logical values
water_r0[is.na(water_r0)]= 0

# Elevation raster
r_elev_NB <- mask(elev_NB, water_poly)
```

4.7.1.3 Connectivity measures

The equation for the commonly used seaway distance measure was provided below for reference, and the three newly proposed connectivity measures follow. Seaway distance was examined at two grid resolutions (100 and 25 m). The other connectivity measures adjusted for increased resistance along an incline (topographic distance) and increased area for the calculation (percent surface area of water and volume of water). These connectivity measures were used in the generation of spatial weights for the EIP to examine their potential effects on sea lice abundance in a linear mixed effects model. These connectivity measures were compared both directly using Euclidean distance and indirectly through the EIP (additional description of these can be found in Chapter 4).

4.7.1.3.1 Seaway distance

Using the waterway raster base map and site locations, the seaway distances among sites were calculated using the ‘costDistance’ function of the ‘gdistance’ package version 1.1-3 (van Etten and de Sousa, 2012) in R. The ‘costDistance’ function employs the Dijkstra algorithm to find the minimum distance $D(i,j)$ between two sites i and j (Eq. 4.2). The distance is calculated along a path u

from i and j which is selected to have the smallest distance (also known as least-cost distance, lowest waterway cost and seaway distance) among paths between i and j. The distance in a path is added up from distances across the individual cells in the raster image, where a total of 8 possible directions can be taken from one cell to the next neighbouring cell. It is understood that all eligible paths progress entirely through water cells. The calculation can be formalized as follows:

$$D(i,j) = \min_{u \in V_{ij}} \left\{ \sum_{k=1}^{n(u)} d(u_{k-1}, u_k) \right\} \quad \text{Eq. 4.2}$$

where V_{ij} is the collection of all paths $u = (u_0, u_1, \ldots, u_{n(u)})$ through connected raster cells that connect sites i and j (i.e., $u_0 = i, u_{n(u)} = j$, and every (u_{k-1}, u_k) is a pair of neighbouring cells, $k = 1, \ldots, n(u)$), and $d(u_{k-1}, u_k)$ is the Euclidean (surface) distance within the cell pair (u_{k-1}, u_k).

The R command below can be implemented:

```
# Calculate seaway distance
distsites2sites<-costDistance(trC,site.mat,site.mat)
```

4.7.1.3.2 Topographical distance

Using the waterway base map, depth (elevation) base map and site locations, the topographical distances among sites were calculated. The addition of a depth base map to the calculations results in a three-dimensional scheme. The topographical distances were calculated using the 'topoDistance' (version 1.0.1; Wang, 2019) which uses the 'costDistance' function of the 'gdistance' package (van Etten and de Sousa, 2012). The 'topoDistance' function finds the minimum topographical distance $tD(i,j)$ between two sites i and j and accounts for depth data (Eq. 4.3). For each cell, the topographical distance is the hypotenuse of the

width of the horizontal distance (within the 25 m grid cell) and the difference in depth (v) travelled through the cell (in m) (Eq. 4.4). A cost of movement was applied to the topographical distance as a depth weighting factor (i.e., f = 1, 10, 100, 500 and 1,000); acute changes in elevation were assigned a larger factor to provide for a greater resistance to vertical movement (i.e., depth). This concept of resistance to movement was implemented as it is applied in landscape ecology where the organism may be less likely to move across certain environments (Wang, 2020).

$$tD(i,j;f) = \min_{u \in V_{ij}} \left\{ \sum_{k=1}^{n(u)} d_t(u_{k-1}, u_k; f) \right\}, \quad \text{with} \qquad \text{Eq. 4.3}$$

$$d_t(u_{k-1}, u_k; f) = \sqrt{d(u_{k-1}, u_k)^2 + f \cdot \left(v(u_{k-1}) - v(u_k)\right)^2} \qquad \text{Eq. 4.4}$$

Where, as for Equation 4.2, V_{ij} is the collection of paths from i to j, u_{k-1} and u_k are the individual neighbouring cells, $d(u_{k-1}, u_k)$ is the Euclidean (surface) distance within the cell pair (u_{k-1}, u_k), and additionally v_{k-1} and v_k are the depths of cells, so that $|v_{k-1} - v_k|$ equals the change in depth within the cell pair (u_{k-1}, u_k), and f is the depth weighting factor. The path with minimal topographical distances between sites i and j obtained from Equation 4.3 we call u^{td}, where, for simplicity, the indices for sites i and j have been suppressed. The path u^{td} was calculated for depth weighting factor 1.

The R commands below can be implemented:

```
# Calculate the topographic distance
# Depth weighting factor = NONE (1)
tdist <- topoDist(r_elev_NB, coordinates(farms_NB), paths = FALSE)

# Calculate the topographic distance with depth weighting factor
# E.g., depth weighting factor = 10
r_bath_NB10 <- r_bath_NB * 10
tdist <- topoDist(r_bath_NB10, coordinates(farms_NB), paths = FALSE)
```

4.7.1.3.3 Percent surface area of water along seaway path

The percent surface area of water along a seaway path $pD(i,j)$ was calculated using the waterway raster base map and site locations. The percent of surface area of water was calculated along the path u^{td} identified as having the smallest distance $tD(i,j)$ between two sites i and j (Eq. 4.3). This path was conveniently generated with the 'topoDistance' package (version 1.0.1 Wang, 2019), but could have otherwise been generated in R, without the use of bathymetry data. Along the path u^{td}, the percent surface area of water was calculated as the average of percentage of water; the sum of waterway cells (1) with an area of 25 m^2 divided by a variable area s (Eq. 4.5). The area for the calculation of the percent surface area of water was square and had its centroid located along the path u^{td}. The distance from the centroid was varied as 10, 20, 40 and 200 cells in the horizontal directions (i.e., grid of 250 m = 441 cells, 500 m = 1,681 cells, and 1,000 m = 6,561 cells).

$$pD(i,j;s) = \sum_{k=0}^{n(u^{td})} pw(u_k;s) \qquad \text{Eq. 4.5}$$

where $u^{td} = (u_0, \ldots, u_{n(u^{td})})$ is the path of the topographical distance $tD(i,j)$ between sites i and j (from the previous section), and $pw(u_k;s)$ is the proportion of water cells in a square window size s around cell u_k.

The R command below can be implemented:

```
# Generate square window
# E.g., 250 m grid ~ (10 · 2 + 1)^2 = 21x21 = 441 cells
f_250 = matrix(c(rep(1,441)), nrow=21)

# Calculate the percent surface area of water along seaway path
water_250 <- focal(water_r0, w=f_250, fun=mean, na.rm=TRUE)
```

4.7.1.3.4 Average volume of water along seaway path

The volume of water along seaway path $vD(i,j)$ was calculated using the waterway base map, depth (elevation) base map and site locations. The volume of water was calculated along the path u_{td} (see previous sections for description of u^{td}). Along the path u^{td}, the volume of water was calculated as the sum of the volume of water for cells with width of 25 m and depth v for a variable area s (see section 2.3.3 for a description of s) (Eq. 4.6).

$$vD(i,j;s) = 25^2 \sum_{k=0}^{n(u^{td})} md(u_k;s) \qquad \text{Eq. 4.6}$$

Where $u^{td} = (u_0, \ldots, u_{n(u^{td})})$ is the path of the topographical distance $tD(i,j)$ between sites i and j (from the previous section), and $md(u_k,s)$ is the mean depth (based on the previously introduced variable v) among cells in a square window of size s around cell u_k.

The R commands below can be implemented:

```
# Calculate the volume of water along seaway path
# E.g., square window 250 m around, 441 grid cells
vlm_water_NB250 <- focal(r_bath_NB, w=f_250, fun=sum, na.rm=TRUE)
vlm_water_NB250_avg <- vlm_water_NB250/441
```

4.7.1.4 Summation procedure

The output matrices of each connectivity measure (site i x site j) were used to calculate the spatial weights by using the Gaussian kernel density estimate (KDE) in R. The value of the Gaussian density function was calculated for the value of x that was equal to the 'distance' of the connectivity measure between site i and site j. This 'distance' of the connectivity measure was in km for seaway distance; the other connectivity measures were linearly scaled to match the

range of 0 to 80 km for the seaway distances calculated in the study area (connectivity measures were multiplied as follows: SA_{250}, SA_{500} and SA_{1000} · 0.02; TD, TD_{10} · 0.001; TD_{100} · 0.0008; TD_{500} · 0.0003; TD_{1000} · 0.00015; VW, VW_{250}, VW_{500}, VW_{1000} · 0.0002). The Gaussian density function bandwidth was varied (i.e., 1-15 in single increments, and 20 to 60 in increments of 5; for seaway distance, these values are in km).

The temporally weighed mean AF abundances 3 weeks prior were multiplied by each of the weights generated using the KDE to produce temporally and spatially weighed AF abundances (Eq. 4.7). The sum of these weighed abundances represented the EIP for a site with its neighbours.

$$\overline{\ln\mathrm{AF}}_{EIP,t} = \sum_{j \in N_i} w_{ij}\, \overline{\ln\mathrm{AF}}_{j,t-21} \qquad \text{Eq. 4.7}$$

Where: $\overline{\ln\mathrm{AF}}_{jt}$ is the triangular temporal average abundances of AF sea lice for site j at time t calculated in Equation 4.1.

w_{ij} are Gaussian kernel weights (e.g., Elghafghuf et al., 2020) computed based on connectivity measure (Eq. 4.2 to 4.7) between sites i and other sites j. Gaussian kernel function bandwidth was evaluated at different values

t ~ target date (day)

j ~ other sites near site i (i.e., N_i ~ neighbour sites of i)

Note: The summation procedure over all neighbouring sites j with the weights w_{ij}(based on the connectivity measure) will be zero or very small for sites j that are far away from site i. However, it is easier to consider the external infestation pressure as a sum over all sites.

4.8.2 Euclidean distance for the comparison of various connectivity measures

Euclidean distance is a classical metric for distance between points in high-dimensional space and it was used for the comparison of the seaway distance

and proposed connectivity measures. This Euclidean distance is used in a different context to the Euclidean distance that is a straight-line distance between 2 sites. In this context, the points are pairs of values of a connectivity measure across all pairs of sites. The dimension (p) of the space equals the number of pairs between sites, i.e., p = 1,953. Distances are computed for pairs among the 14 connectivity measures, each scaled by dividing by the standard deviation of the connectivity measure (across all pairs of sites). The normalization by the respective standard deviations is needed to ensure that the distances are on comparable scales. The Euclidean distance in p-dimensional space is a generalization of the hypotenuse of a triangle formed by 2 points in 2-dimensional space, and the resulting formula is shown in Equation 4.8.

$$L_2(x,y) = \sqrt{\sum_{j=1}^{p} \left(\frac{x_j}{\sigma_x} - \frac{y_j}{\sigma_y}\right)^2} \qquad \text{Eq. 4.8}$$

Where $x = (x_1, \ldots, x_p)$ and $y\,(y_1, \ldots, y_p)$ in p-dimensional space for two connectivity measures, σ ~ standard deviation of the connectivity measure, and j ~ pairs of sites.

4.8.3 References

Akima, H., Gebhardt, A., Petzold, T., Maechler, M. (2021). akima: Interpolation of Irregularly and Regularly Spaced Data. R package version 0.6-2.3. https://cran.r-project.org/package=akima

Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). Bay of Fundy Marine Aquaculture Sites [map]. 1:125000. New Brunswick, Canada.

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

GADM. (2018). World/Canada/New Brunswick. Retrieved from https://gadm.org/maps/CAN/newbrunswick.html

QGIS Development Team. (2018). QGIS Geographic Information System. Open Source Geospatial Foundation Project. Retrieved from http://qgis.osgeo.org

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

StataCorp (2019). Stata: Release 16. Statistical Software. StataCorp LLC, TX, USA.

van Etten, J., and de Sousa, K. (2012). gdistance: Distances and Routes on Geographical Grids. R package version 1.1-3. https://CRAN.R-project.org/package=gdistance

Wang, I. (2019). topoDistance: Calculating Topographic Paths and Distances. R package version 1.0.1. https://cran.r-project.org/package=topoDistance

Wang I.J. (2020) Topographic path analysis for modeling dispersal and functional connectivity: calculating topographic distances using the topoDistance R package. Methods in Ecology and Evolution, 11:265-272.

Zhan, C., Menon, S., Gao, P. (1993). A directional path distance model for raster distance mapping. Lecture Notes in Computer Science (Including Subseries Lecture Notes in Artificial Intelligence and Lecture Notes in Bioinformatics), 716 LNCS, 434–443. https://doi.org/10.1007/3-540-57207-4_2

5 Predicting the dispersal of *Lepeophtheirus salmonis* in the Bay of Fundy, New Brunswick

5.1 Abstract

Lepeophtheirus salmonis are routinely counted for monitoring purposes by the Atlantic salmon farmers in the Bay of Fundy, New Brunswick (NB), Canada. These data are collected in the Fish-iTrends® database. A subset of 2016-2021 data was used to generate a predictive model for adult female (AF) sea lice abundance. A multivariable autoregressive linear mixed effects model (AR[2]) was fit with treatments, infestation pressures (a measure that represents the dose of exposure of parasitic stages of sea lice to potential fish hosts) within (internal) and among sites (external), and other predictors. The treatments were categorized by duration and type. The effect of the mechanical treatment decreased with increasing sea surface temperature. In-sample predictions had good accuracy. A one standard deviation increase in the external infestation pressures (EIP_{AF}) affected a significant relative increase of the abundance of AF by 5 % ($p = 0.002$) when other model predictors were kept constant. Sites separated by short seaway distances had stronger EIP_{AF} compared to sites with more considerable distances. This model may be helpful for managers and farmers regarding sea lice mitigation strategies on salmon farms in the Bay of Fundy, NB.

5.2 Introduction

The Atlantic salmon, *Salmo salar* (Linnaeus [WoRMS, 2022]), aquaculture industry in the Bay of Fundy, New Brunswick (NB), Canada, monitors the

parasitic sea louse, *Lepeophtheirus salmonis salmonis* (Krøyer [WoRMS, 2022]), and, since 2009, keeps a record of their findings in the Fish-iTrends® data management system. The database holds information regarding counting events of *L. salmonis* and delousing treatments, which is essential for management, monitoring, and regulatory purposes (for additional detail, see Gautam et al., 2017a). Counting events include multiple steps, namely the selection of cages on a farm or aquaculture site (hereafter, site), the capture and anesthesia of numerous fish, and the counting of three *L. salmonis* life stages (for additional detail, see Jones et al., 2012). The provincially regulated monitoring program requires each site to perform counts every week and before and after treatments (Jones et al., 2012; General Regulation 91-158 [Government of New Brunswick, 2020]); however, the frequency of counts is relaxed to once a month when sea temperatures fall below 5°C (Government of New Brunswick, 2020). This results in a longitudinal repeated measures dataset for sea lice abundance, with uneven temporal patterns since the counting events were performed on given days but occurred sporadically (i.e., not every day for every cage at a site). These uneven gaps in the time series may be short interruptions in sampling events (e.g., adverse weather conditions or when air temperature is so cold that it could damage the fish to remove them from the water to count lice) or lengthy interruptions during fallowing periods (ranging from 29 to 75 weeks [Elghafghuf et al., 2020]) when a site was left empty after harvest and before stocking the next group of smolts.

A mixed linear regression model of adult female (AF) sea lice abundance was published from the longitudinal repeated measures dataset of cage-level counts (Chapter 3). We expanded on the previously published work to create a relatively simple model capable of short-term predictions. A simple model that can generate accurate predictions may aid in planning decisions for sea lice mitigation strategies (Aldrin et al., 2017; Elghafghuf et al., 2018, 2021; St-Hilaire et al., 2018). The previously published model could not be employed for prediction purposes as it had a complex organizational structure of count data that were organized into segments (defined as counts after a topical treatment followed by all additional counts until the next treatment within a site and production cycle [Chapter 3]). The segment organization of the data was unsuitable for predictions due to the irregular arrangement of the segments, as they depended on the irregular temporal pattern of treatment applications.

Counts were performed daily at cages and submitted into Fish-iTrends directly producing a daily resolution to the data, with multiple irregular time gaps (Chapter 3). Short-term predictions were better suited to a weekly scale to represent management perspectives, as is more commonly applied in other predictive studies (e.g., 1 week: Elghafghuf et al., 2018, 2021; St-Hilaire et al., 2018 and 10 days: Myksvoll et al., 2018). The weekly scale of the predictions may be selected because the sea lice count data were submitted into the data management system as site-level weekly mean abundances (e.g., the Norwegian BarentsWatch) or to complement the regulations for the minimum frequency of weekly counts of sea lice (Elghafghuf et al., 2018, 2020). Additionally, few

published papers describe the methods for calculating the site-level weekly mean abundances, either because the information was not recorded in their database or simply because they were not published (e.g., the week's starting day was not defined by Elghafghuf et al. [2020]). The daily cage-level resolution of the Fish-iTrends® data offers the opportunity to describe the averaging methods to obtain site-level weekly abundances of sea lice.

A state-space model was previously published that produced accurate short-term weekly predictions for the Atlantic salmon aquaculture sites around Grand Manan Island, NB (Elghafghuf et al., 2021). State-space models are powerful but complex, and a simpler model of NB's Atlantic salmon industry would be beneficial. More specifically, a time series regression would be suitable for this task. One approach was the distributed lag model used to answer environmental epidemiological questions (Katsouyanni et al., 1996; Bhaskaran et al., 2013). This approach was deemed inadequate for the complex structure and hierarchy of sea lice data and the lack of an explicit intervention; multiple treatment applications were performed at numerous sites without spatiotemporal patterns. A mixed effects model, however, can handle the complexity of the sea lice data and be apprehensible for the industry.

Both the state-space model, constructed for Grand Manan (Elghafghuf et al., 2020, 2021) and the mixed effects model, with the segment organization for the entire Bay of Fundy (Chapter 3), employed predictive covariates of sea lice infestation pressures. The infestation pressures represent the dose of exposure of parasitic stages of sea lice to potential fish hosts (Chapter 3). The infestation

pressure is organized for the site and production cycle (internal infestation pressure: IIP) and among sites (external infestation pressure: EIP). Including infestation pressures in the prediction models resulted in slightly better prediction accuracy than models without infestation pressures (Elghafghuf et al., 2021).

The primary objective of this work was to develop a predictive model for longitudinal data of AF *L. salmonis* abundance for the production cycles in the Bay of Fundy, NB, and to demonstrate its predictive capabilities with an illustrative example. We used recent data (2016-2021) and adjusted for the effects of infestation pressures and topical and in-feed treatments. The estimation of infestation pressures was necessary for the model, and a secondary objective was to compare these infestation pressures with the previously published estimates for the same area, but that differ in time, methodology, and data than were previously published (Chapter 3).

5.3 Materials and methods

5.3.1 Study area and data structure

L. salmonis count and treatment information from aquaculture sites in the Bay of Fundy, NB, between January 2016 and July 2021, were extracted from the Fish-iTrends® data management system (n = 41,240 count events). Routine counts included the pre-adult male and female, and adult male (PAAM) and AF (with and without egg strings) *L. salmonis* sea lice on Atlantic salmon. Counts of sea lice were repeated measures within a cage where multiple cages were organized within sites and numerous sites within Bay Management Areas (BMAs). Three companies operated in the Bay of Fundy, NB, and were included in our analyses

as a categorical predictor. Counts were labelled as ‘weekly,’ ‘pre-treatment count,’ and ‘post-treatment count,’ depending on when they were applied to a cage. Production cycles further defined cohorts of fish at sites (i.e., site-[production] cycles). The production cycles are not directly recorded in the Fish-iTrends data management system but were identified using the recorded variables of stocking and harvesting dates. The production cycle began when the first fish were stocked and ended with the last count at a site prior to fallowing and around the harvest date. Fish-iTrends Groups of fish within a cage had a unique identifier per site to follow the fish movement among cages that may have occurred during treatment events.

5.3.2 Preparation of the outcome

The observational unit for analysis was the sites’ weekly mean abundances of AF sea lice (site-weeks equivalent to farm-weeks, as referred to by other authors). The weekly time scale was a logical choice for prediction since it reduced the temporal variation of counts that occurred at the cage-level and complemented the regulation requirement of weekly counts. The selection of AF as the outcome was determined by the lower variance of AF compared to PAAM (Elmoslemany et al., 2013). The abundances were calculated as the natural logarithm of the sea lice count divided by the number of fish sampled, with an offset of 1 (additional details are found in the Supplemental Information). The average abundance of AF sea lice was calculated for a week (the week was defined as Monday to Sunday) and per site with cage-weighting. The fish group identifier used in the Fish-iTrends® database to follow groups of salmon that may be split or moved

between cages during the production cycle was ignored during the averaging process as the aggregation occurred at the site-level.

5.3.3 Calculation of internal and external infestation pressures

The internal infestation pressure (IIP) and external infestation pressure (EIP) were calculated using the previously described site-week average abundance of sea lice. The IIP was calculated for AF (IIP_{AF}) and PAAM (IIP_{PAAM}) as 3- and 4-week lagged abundances, respectively. The decision for the lags was based on a review of the literature and mean sea temperature for the study area (for additional detail, see Chapter 3).

The EIP_{AF} was calculated as the sum of the Gaussian kernel density estimate (KDE)-weighted spatial average abundance of AF with a 3-week lag from other sites (i.e., neighbours). The spatial component was calculated as the seaway distance (least-cost distance) at 100 m resolution among the manually selected approximate centre points (centroids) for sites using the ‘gdistance’ package, version 1.1-3 (van Etten and de Sousa, 2012) in R (R Core Team, 2020). The weights were estimated from the seaway distances among sites using the Gaussian KDE with a kernel bandwidth of 10 km. The seaway distance is commonly used to estimate the dispersal of sea lice and was used to compare other estimates of dispersal (Chapter 4). The 10 km kernel bandwidth represented the optimal displacement distance for AF sea lice among sites and was selected from a previous sensitivity analysis comparing various kernel bandwidths (Chapter 3). The Gaussian KDE weights were then multiplied by the abundance of AF with a 3-week lag for other sites, and these values were

summed to represent the EIP of a site. The equation for calculating the EIP_{AF} using site-level weekly abundances of AF sea lice is presented in the Supplemental Information.

5.3.4 Topical and in-feed treatments

Treatments were considered as three separate predictors in the analysis; the variables were labelled as follows: $Week_{\dagger}$ where † represents the treatment type (C: short-term chemical, M: mechanical, and I: in-feed chemical). The predictors were primarily created according to the duration of effect for topical (chemotherapeutic agents and non-medicinal operations with short duration of effect) and in-feed/oral treatments (chemotherapeutic agents with prolonged duration of effect [$Week_{I*}$]). The in-feed treatments included emamectin benzoate (Slice®, Merck Animal Health, USA) and avermectins (Ivomec®, Boehringer Ingelheim, Germany, and Noromectin®, Norbrook Laboratories Ltd., United Kingdom). The short treatment category was subdivided into chemical ($Week_{C*}$) and mechanical treatments ($Week_{M*}$). The chemical treatments included hydrogen peroxide (combined Interox® Paramove® 50^{TM}, Solvay Chemicals Inc., Belgium, and Aquaparox 50^{TM}, Alpha Chemical Limited, Canada) and azamethiphos (Salmosan® Vet, Benchmark Animal Health, United Kingdom). The mechanical treatments included thermal (also warm water) treatment and Hydrolicer®.

Each treatment was constructed to have 4 categories based on the temporal relation of a given week to a treatment: B: before, Tx: treatment, A: after, and O: other. A treatment week ($Week_{\dagger Tx}$) may not have an associated $Week_{\dagger B}$ and

$Week_{\dagger A}$ if no counts were performed the week before and/or after the treatment week. A treatment week was identified as having any treatment applied between Monday to Sunday. Treatment weeks may occur as consecutive weeks, and different types of treatments may occur during the same week.

Chemical and mechanical treatment predictors were categorized as a treatment week ($Week_{CTx}$ and $Week_{MTx}$), a week before the treatment week ($Week_{CB}$ and $Week_{MB}$), a week after the treatment week ($Week_{CA}$ and $Week_{MA}$), and any other week that did not fit the previous criteria ($Week_{CO}$ and $Week_{MO}$). The $Week_{MTx}$ was further subdivided into its components of thermal and Hydrolicer® treatments to evaluate the impacts of each. A one-week duration of effect, i.e. reduction of sea lice abundance, was assumed for topical treatments as applied in other publications (e.g., Elghafghuf et al., 2018).

The in-feed treatment predictor was categorized similarly to the short-term treatment predictors but with two critical differences. The week of treatment and the week before the treatment were combined ($Week_{IB}$). The $Week_{IA}$ encompassed four weeks as the effect of in-feed treatments was believed to have residual effects (Elghafghuf et al., 2018, 2020); a laboratory trial demonstrated that the concentration of emamectin benzoate residues decreased until day 45 after oral administration, with a peak at day 5 (Whyte et al., 2011).

Table 5.1 lists these treatment variables with a brief description.

5.3.5 Time at sea

Atlantic salmon growth was controlled in the model using the number of weeks since the salmon were stocked at a site (time at sea: TAS). To note, salmon were stocked at a site over often over several weeks, and the stocking date in the database was for the date that the first cage at a site was stocked. The TAS represented salmon growth as the age, size, and weight may be associated with variation in the abundance of sea lice (for review, see Frenzl, 2014). However, information related to salmon's age, size, and weight was not recorded in Fish-iTrends®.

Table 5.1. Descriptive statistics for all variables in the study, potentially used to predict the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice. Results of the univariable mixed autoregressive model were summarized by Akaike's information criterion (AIC; null model AIC = 6,442). SD: standard deviation; IIP: internal infestation pressure; PAAM: pre-adult and adult male sea lice; EIP: external infestation pressure; n = 3,137 site-weeks.

Variable and categories	Variable and category descriptions	Mean (SD)	Mean (SD) abundance of AF sea lice for the category	AIC
IIP_{AF}	Internal infestation pressure; temporal weighted average abundance of AF sea lice with a 3-week time lag	1.38 (4.91)		6,443
IIP_{PAAM}	Internal infestation pressure; temporal weighted average abundance of PAAM with a 4-week time lag	2.29 (5.19)		6,401
EIP_{AF}	External infestation pressure; temporal and KDE spatial weighted average abundance of AF sea lice with a 3-week time lag	1.07 (1.38)		6,430
Temp	Sea surface temperature from UKMO OSTIA product (°C)	11.21 (2.31)		6,433
TAS	Time at sea: number of weeks since the stocking of a cage at a site	44.0 (28.3)		6,186

Table 5.1 continued the next page.

Table 5.1 continued.

Variable and categories	Variable and category descriptions	Mean (SD)	Mean (SD) abundance of AF sea lice for the category	AIC
Mechanical treatment	Combined thermal topical and Hydrolicer® treatments			6,314
$Week_{MB}$	A single week before treatment week if it exists; n =128 site-weeks		3.87 (4.09)	
$Week_{MTx}$	Week of mechanical treatment application; n = 382 site-weeks		2.53 (4.28)	
$Week_{MA}$	Single week after treatment week, if exists; n = 133 site-weeks		2.18 (3.20)	
$Week_{MO}$	Week that does not fit other criteria; n =2,494 site-weeks		1.57 (4.75)	
Chemical treatments	Combined hydrogen peroxide and azamethiphos applications			6,435
$Week_{CB}$	A single week before treatment week if it exists; n = 65 site-weeks		3.82 (4.79)	

Table 5.1 continued the next page.

Table 5.1 continued.

Variable and categories	Variable and category descriptions	Mean (SD)	Mean (SD) abundance of AF sea lice for the category	AIC
Chemical treatments				
$Week_{CTx}$	Week of chemical treatment application; n = 405 site-weeks		2.47 (4.46)	
$Week_{CA}$	Single week after treatment week, if exists; n = 72 site-weeks		3.66 (3.91)	
$Week_{CO}$	Week that does not fit other criteria; n = 2,595 site-weeks		1.59 (4.64)	
In-feed treatments	Combined emamectin benzoate and avermectin in-feed treatments			6,443
$Week_{IB}$	Combined 2 weeks of single week before treatment and week of in-feed treatment application; n = 764 site-weeks		0.85 (3.78)	
$Week_{IA}$	Weeks 1-4 after treatment week, if exists; n = 545 site-weeks		1.79 (3.47)	
$Week_{IO}$	Week that does not fit other criteria; n = 1,828 site-weeks		2.34 (4.99)	

Table 5.1 continued the next page.

Table 5.1 continued.

Variable and categories	Variable and category descriptions	Mean (SD)	Mean (SD) abundance of AF sea lice for the category	AIC
BMA	Bay Management Area			6,451
1	Passamaquoddy Bay, Deer Island, most of Campobello Island; 14 sites; n = 681 site-weeks		2.29 (5.33)	
2A	Back Bay, Bliss Harbour, Lime Kiln Bay; 8 sites; n = 649 site-weeks		2.84 (5.36)	
2B	Eastern Grand Manan Island, White Head Island; 11 sites; n = 907 site-weeks		1.69 (3.43)	
3A	Eastern mainland coast of Southwest NB from Beaver Harbour to Haley's Cove; 5 sites; n = 305 site-weeks		1.68 (5.27)	
3B	Southern Grand Manan Island, excluding White Head Island; 8 sites; n = 454 site-weeks		1.86 (4.92)	
3C	Anse Sawyers Cove; 2 sites; n = 141 site-weeks		1.99 (4.54)	

Table 5.1 continued next page.

Table 5.1 continued.

Variable and categories	Variable and category descriptions	Mean (SD)	Mean (SD) abundance of AF sea lice for the category	AIC
Company	Owners of the aquaculture sites in the Bay of Fundy, NB			6,446
1	35 sites; n = 2,385 site-weeks		1.98 (4.64)	
2	11 sites; n = 627 site-weeks		2.11 (4.99)	
5	2 sites; n = 125 site-weeks		2.64 (3.71)	

5.3.6 Modelling

5.3.6.1 Inclusion criteria

The dataset was restricted to the BMAs where Atlantic salmon were produced; the BMA 5 and 6 were not included (n = 349 counts, 0.8 %). Cages must have been counted at a frequency greater than 5 counting events for a site-cycle (excluded n = 369 counts, 0.9 %). The observations were limited to production cycles starting in 2016 to 2020. Observations for production cycles that began in 2021 were not included (n = 150 counts).

5.3.6.2 Multivariable autoregressive linear mixed effects model

A multivariable autoregressive linear mixed effects model (Rosner et al., 1985) was used to predict the weekly average abundance of AF sea lice. Random effects were for the site-cycles. The BMAs were applied as fixed effects (Chapter 3). The sea surface temperature (hereafter, temperature) fixed effects were extracted from the UKMO OSTIA SST product (UK Met Office, 2005) as previously described (Chapter 3) and centred at the median of 11.85 °C. The treatment predictors (chemical, mechanical and in-feed) reference values were set to the week before the treatment ($Week_{CB}$, $Week_{MB}$, and $Week_{ITx}$) to examine the effects of before and after treatment. A fractional polynomial regression with default powers and two dimensions was used to examine potential transformations for improved fit of the TAS predictor. Random slopes for time were applied as dichotomized TAS (categorized as prior and after 44 weeks at sea) to explore the possibility that the TAS was not constant between site-cycles

and this model was compared for improved fit to the model without random slopes.

The autocorrelation and partial autocorrelation functions (ACF and PACF) were utilized to evaluate autoregressive processes that may fit the residual errors. These ACF and PACF plots were assessed for each site-cycle. The model was fitted with a second-order autoregressive (AR[2]) process correlation structure to the errors, stratified by three temperature profiles for the site-cycle hierarchical level. The temperature profiles were low 6-9 °C, medium 10 °C, and high 11-12 °C, which were defined from the mean temperature for the site-cycles (SD 0.7 °C). To evaluate the potential effect of IIP_{AF}, alternate residual structures were applied; first-order autoregressive (AR[1]) and independent structures of the residual errors were used with stratification by temperature profiles.

An adjusted version of the time series ‘without winter gaps’ was created for site-cycles with intervals of 5 weeks or more during the winter (defined as December to March, inclusive). These gaps were replaced by a single week to evaluate the effect of correlation (exponential decay) in time since the model considered observations across a large interval to be independent.

Models were fit with the restricted maximum likelihood estimation unless nested models were compared. Akaike’s information criterion (AIC) was used to compare models with different residual correlation structures. The model assumptions of linearity, homogeneity of variance, and normality of residuals were assessed graphically for each level of the hierarchy.

Influential observations were described and evaluated in the Supplemental Information. Two observations were removed after evaluation as these represent possible gross errors.

Unless otherwise specified, all analyses were conducted in Stata® v17 (StataCorp, 2021), and statistical significance was set at $p < 0.05$.

5.3.6.3 Prediction

An illustration of the model's capability was produced with an in-sample prediction with a horizon of eight weeks. The prediction start date (week 36 of the year 2020, corresponding to August 31-September 6, 2020) was purposefully selected to make predictions during periods of high abundance of sea lice (late summer) and with a large training dataset (≥ 2,500 site-weeks) to which the model was fitted.

The prediction for a site-week was calculated as the sum of the linear and random effects and the predicted residuals for lag weeks 1 and 2 multiplied by the autoregressive coefficients for lag weeks 1 and 2 (Pinheiro and Bates, 2000). The equation for the prediction is found in the Supplemental Information. The highest temperature category of the random effects was selected to match the mean weekly temperature for the prediction period. After the first week of prediction, the predicted residual was replaced by a residual calculated from the difference between the observed abundance of AF and the prediction from the first week. The predictions for weeks 1 to 8 were also calculated without predicted residuals or residuals calculated from the difference between the

observed and predicted values of AF; the residuals for lag weeks 1 and 2 were set to 0. With this latter method, the predictions were entirely driven by the linear and random effects of linear prediction. Additional details are found in the Supplemental Information.

The accuracy of the predictions was assessed using the normalized prediction error. The normalized prediction error was calculated as the difference between the observed and residual AF abundance divided by the standard deviation of the random effects, squared (see the Supplemental Information for additional information).

5.4 Results

5.4.1 Descriptive statistics of the observations modelled

There were 3,137 site-weeks modelled over 261 weeks from June 6, 2016, to June 7, 2021. There were 77 production cycles for 48 sites. On average, the site-cycles lasted 40.7 weeks with a range from 6 to 74 weeks. On several occasions (8.4 %; 274/3,137 site-weeks) cages at sites were counted multiple times per week (range of 1 to 5 cage/site-cycle/week).

The abundances of sea lice (AF and PAAM) were right-skewed; therefore, these were transformed with a natural logarithmic function (additional details are found in the Supplemental Information). A descriptive summary of potential variables for the multivariable model of the abundance of AF is in Table 5.1. The AF abundances were highly correlated (0.82) with means of 2.05 (standard deviation [SD] 4.68) and 1.38 (SD 4.91) for the outcome and IIP_{AF}, respectively. The mean IIP_{PAAM} was larger than the mean $IIP_{AF,}$ which was larger than the mean EIP_{AF}.

The EIP_{AF} was calculated from weights derived from seaway distances with a mean of 29.07 km (SD 19.14 km) among sites.

The mean weekly temperature was 11.21 °C (SD 2.31 °C; Table 5.1) with a range of 3.85 °C to 16.16 °C for 2016-2021. There were a larger number of observations for the 10 °C temperature category of the random effects with 1,725 site-weeks (55.0 %). The 6-9 °C category had 415 site-weeks (13.2 %), and 11-12 °C had 997 site-weeks (31.8 %). The mean AF abundances for the temperature profiles were 1.52 (SD 4.48) for 6-9 °C, 1.93 (SD 4.66) for 10 °C and 2.41 (SD 4.62) for 11-12 °C. The predicted period temperature (weeks 37 to 40) had a mean of 13.11 °C (n = 508 site-weeks, SD 0.71 °C, range 11.32-15.42 °C).

The Atlantic salmon spent, on average, 44.0 site-weeks (SD 28.3) at sea (TAS), with a range of 4 to 123 site-weeks. The fractional polynomial regression estimated a square root function as the best transformation for the TAS predictor.

A total of 353 treatment events occurred in the sample of counts modelled. Mechanical, chemical, and in-feed treatments were applied on a site within one week (n = 173 of 1,000 site-weeks, 17.3 %), but most extended beyond one week (n = 827, 82.7 %). There were 405 site-weeks of chemical treatments ($Week_{CTx}$) and 382 site-weeks of mechanical treatments ($Week_{MTx}$; Table 5.1). The Hydrolicer® and thermal treatments were applied during 89 and 293 site-weeks, respectively. The week before chemical treatments ($Week_{CB}$) had a mean AF abundance of 3.82 (SD 4.79) and decreased to 3.66 (SD 3.91) for the week after chemical treatments ($Week_{CA}$). The week before mechanical treatment

($Week_{MB}$) had a mean AF abundance of 3.87 (SD 4.09), and the week after mechanical treatment ($Week_{MA}$) was 2.18 (SD 3.20). A total of 100 site-weeks had both chemical and mechanical treatments applied. There were 1,316 site-weeks when no treatments were applied ($Week_{MO,}$ $Week_{CO}$ and $Week_{IO}$), and the mean AF abundance was 2.36 (SD 5.21) during those weeks.

5.4.2 Final model

The final multivariable autoregressive linear mixed effects model (AR[2]) had the continuous predictors IIP_{AF}, IIP_{PAAM}, EIP_{AF}, temperature, and TAS, and the categorical predictors of mechanical treatments, mechanical treatments and temperature interaction, chemical treatments, in-feed treatments, and BMA (Table 5.2). The autoregressive 2nd order had good fit according to the ACF and PACF descriptive plots (data not shown). The random effects were the site-cycle, and the AR(2) structure of the errors was stratified by temperature. Temperature and BMA fixed effects results were similar to the previously published model (Chapter 3). The company variable was not retained in the final model as it was non-significant (Wald Chi-square test, p = 0.914).

Table 5.2. Multivariable autoregressive linear mixed effects model (AR[2]) for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice with an external infestation pressure weighted with a kernel density estimate of seaway distance. Temperature: sea surface temperature; IIP: internal infestation pressure; EIP: external infestation pressure; PAAM: pre-adult and adult male sea lice; n = 3,137 weeks.

Variable and their categories	Coefficient	95 % Confidence interval	p-value
IIP_{AF}	-0.002	-0.037, 0.034	0.926
IIP_{PAAM}	0.104	0.076, 0.133	<0.001
EIP_{AF}	0.243	0.091, 0.394	0.002
Temperature (°C), centered	-0.043	-0.066, -0.021	<0.001
$TAS^{0.5}$	0.422	0.379, 0.466	<0.001
Mechanical treatments			<0.001
$Week_{MB}$	baseline		
$Week_{MTx}$	-0.264	-0.362, -0.166	
$Week_{MA}$	-0.604	-0.718, -0.491	
$Week_{MO}$	-0.381	-0.485, -0.277	

Table 5.2 continued the next page.

Table 5.2 continued.

Variable and their categories	Coefficient	95 % Confidence interval	p-value
Mech. treatment-Temperature interaction			0.005
$Week_{MB}$*centered Temp	baseline		
$Week_{MTx}$*centered Temp	0.057	0.008, 0.107	
$Week_{MA}$*centered Temp	0.057	0.000, 0.114	
$Week_{MO}$*centered Temp	-0.006	-0.055, 0.043	
Chemical treatments			0.045
$Week_{CB}$	baseline		
$Week_{CTx}$	0.011	-0.132, 0.154	
$Week_{CA}$	-0.129	-0.296, 0.038	
$Week_{CO}$	-0.151	-0.296, -0.006	
In-feed treatments			<0.001
$Week_{ITx}$	baseline		
$Week_{IA}$	0.052	-0.024, 0.129	
$Week_{IO}$	-0.229	-0.355, -0.102	

Table 5.2 continued the next page.

Table 5.2 continued.

Variable and their categories	Coefficient	95 % Confidence interval	p-value
BMA			0.327
1	baseline		
2A	0.157	-0.195, 0.508	
2B	0.082	-0.238, 0.402	
3A	0.410	-0.017, 0.837	
3B	0.345	-0.034, 0.724	
3C	0.072	-0.506, 0.650	
Constant	-1.692	-2.090, -1.294	

There were 2 to 35 consecutive site-weeks range and non-consecutive time values (gaps) of 2 to 32 weeks. The model ran with the ‘without winter gaps’ time series and produced changes greater than 25 % for only two of the coefficients of IIP_{AF} and $Week_{MB}$*Temp (additional details are found in the Supplemental Information).

No violations of model assumptions of linearity, homoskedasticity and normality were observed.

5.4.2.1 Prediction

The training dataset had a total of 2,658 site-weeks with 76 site-production cycles. Eight weeks of prediction points were calculated from 112 site-weeks and 14 site-production cycles. The predictions were computed using the observed values to estimate the residuals and were shown in Figure 5.1. The plot illustrated a single site-cycle from each BMAs to maintain the site's anonymity. The predictions generally followed the trends in observed AF abundances.

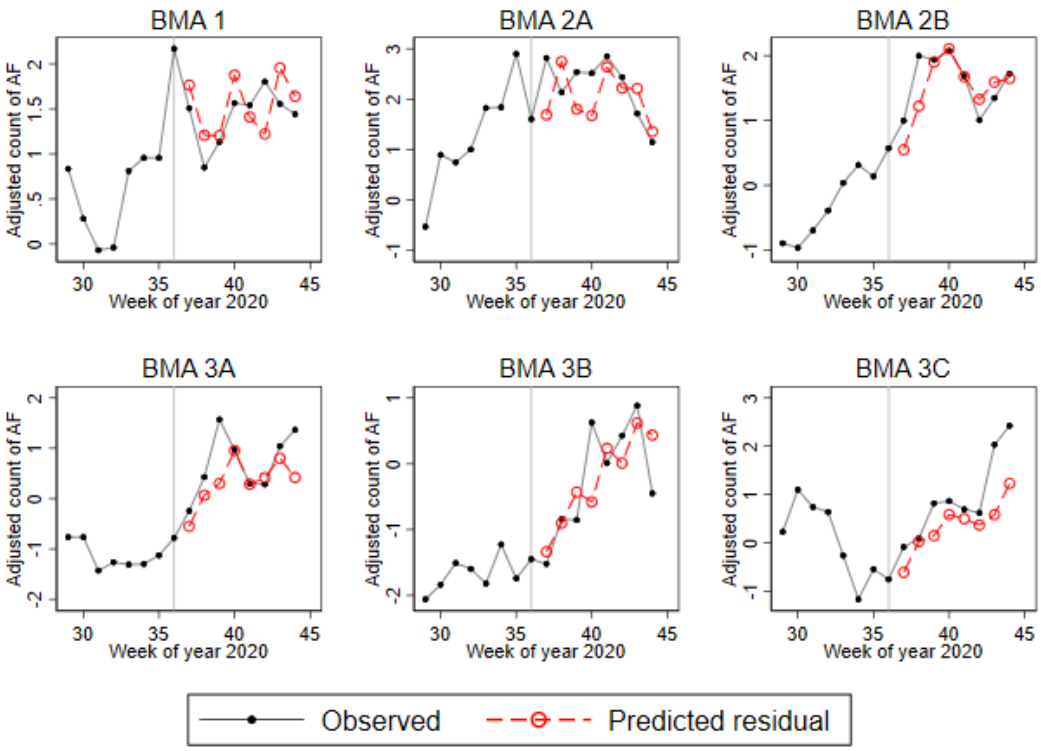

Figure 5.1. In-sample short-term predictions (8 weeks; red points) of the abundance of adult female *Lepeophtheirus salmonis* for single selected sites from each of the Bay Management Areas (BMA) in the Bay of Fundy, New Brunswick, Canada, using a multivariable autoregressive linear mixed effects model (AR[2]). The vertical grey line marks the last date before predictions (week 36 of 2020). Predictions were calculated using the observed values (black points) to generate residuals.

The median of the normalized prediction error was 0.13, with a 25th percentile of 0.04 and a 75th percentile of 0.51. Weeks 2 and 5 had single sites with greater than 2 standard deviations of the prediction error (Figure 5.2). The predictions calculated using residuals set to 0 had a slightly larger mean normalized prediction error (median 0.27, 25th percentile 0.05, 75th percentile 0.95). No trend was noted for an increase in the prediction error across the prediction horizon for either of the methods of calculating the predictions (additional information is found in the Supplemental Information).

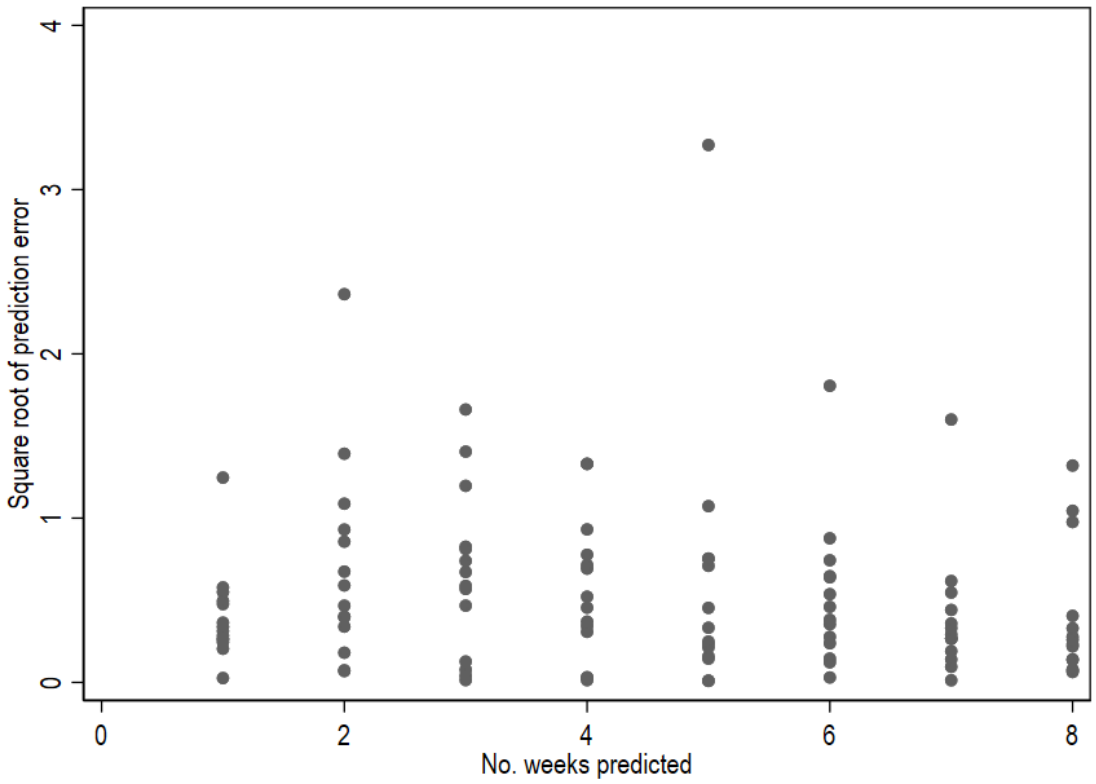

Figure 5.2. The square root of the prediction error for 14 sites (n = 112 observations) across the prediction horizon of 8 weeks. The prediction errors were calculated using a multivariable mixed autoregressive model for in-sample short-term predictions of adult female *Lepeophtheirus salmonis* abundance.

5.4.2.2 Fixed effects

5.4.2.2.1 Internal and external infestation pressures

The IIP_{AF} was not significantly different from zero in the final model (p = 0.926). The effect of the IIP_{AF} was examined under varying orders of the autoregressive function: AR(2), AR(1), and the independent residual structures were specified for the random-effects equation. The AR(1) and independent alternate residual structures did not improve the model (ΔAIC = 66 and ΔAIC = 1,170, respectively) but demonstrated that IIP_{AF} was a significant predictor. The model with the AR(1)

estimated an abundance of AF that increased by a factor of 1.08 (i.e., a relative increase of 7.7 %; 95 % confidence interval [CI] 1.04-1.12; $p < 0.001$) for an increase of one unit of the IIP_{AF} when all other predictors were kept constant. The model with the independent structure to the errors estimated a relative increase of 35.3 % of the abundance of AF sea lice (95% CI 1.30-1.40; $p < 0.001$) for one unit change in the IIP_{AF}.

The effect of the IIP_{PAAM} was a relative increase of 11.0 % of the abundance of AF ($p < 0.001$) for an increase in one unit change in the IIP_{PAAM} when other predictors were kept constant.

A one-unit increase in the EIP_{AF} resulted in a relative increase of 27.5 % of the abundance of AF ($p = 0.002$) when other predictors were kept constant in the final model. An example of the contributions of EIP_{AF} was calculated and illustrated in Figure 5.3. The sites (pseudonyms A-F) in the sample were active sites for the study period; the site abundance values were chosen for demonstration. Five areas (A-E) contributed an abundance of 1.1 AF sea lice, and a site (F) contributed twice that abundance, resulting in a total EIP_{AF} of 4.07. The site with high exposure also has a short seaway distance (2.25 km) from the evaluated site and, therefore, had the most substantial EIP_{AF} contribution (dark orange) compared to the other sites in the example. Longer seaway distances produce lower EIP_{AF} contributions. Of the sites with an abundance of 1.1 AF, site E has a more substantial contribution (pale orange) because it was the closest to the site evaluated (500 m). Sites A and C were located a seaway distance of 6.46 km and 1.77 km, respectively, away from the considered site and

contributed an intermediary value to the EIP_{AF}. Sites B and D have longer seaway distances (13.75 and 9.98 km, respectively), which were close to the KDE bandwidth of 10 km and contribute very little to the EIP_{AF} of the evaluated site.

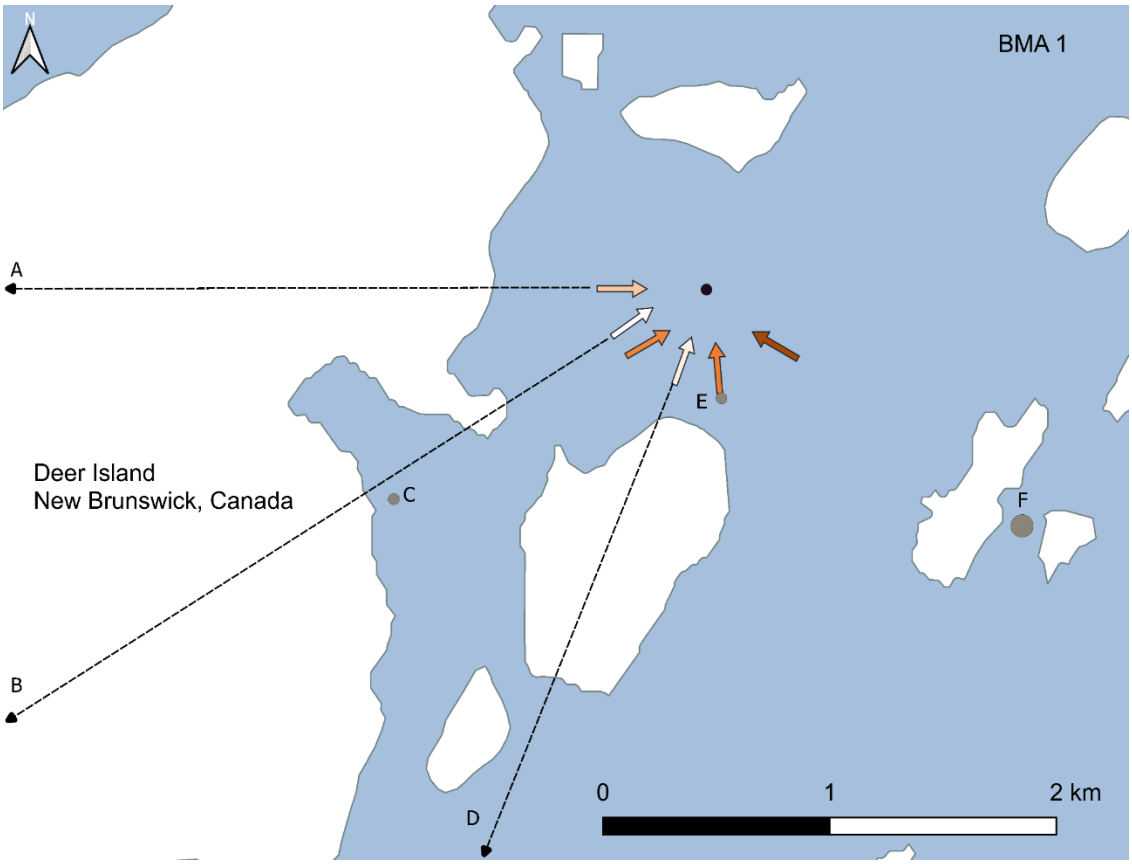

Figure 5.3. Site contributions (arrows) from neighbouring sites (A-F) to the external infestation pressure of a selected site (black circle) in the Bay Management Area 1 of the Bay of Fundy, New Brunswick, Canada. The colour of the arrows was on the scale of white to orange, linearly proportional to the contributions. Site F's contributions were twice those of other sites (A-E). Dashed arrows (A, B and D) point to sites beyond map bounds at seaway distances of 6.5 km, 13.8 km, and 10.0 km, respectively.

5.4.2.2.2 Treatments and temperature and their interaction

The effects of applying mechanical treatments varied by temperature (Figure 5.4; Wald Chi-square test, $p = 0.005$). Mechanical treatments had a reduction of the

estimated AF sea lice abundance during comparison of the $Week_{MA}$ to the $Week_{MB}$ (referent) for the range of temperatures presented (6.5-15.0 °C). However, the effectiveness was reduced at higher temperatures; $Week_{MA}$ had relative decreases of the AF abundances of 52.0 % at 9.56 °C and 42.0 % at 12.89 °C. At a low temperature (25^{th} percentile: 9.56 °C), there was a relative decrease of 32.6 % of the AF abundance for the $Week_{MTx}$ compared to the $Week_{MB}$.

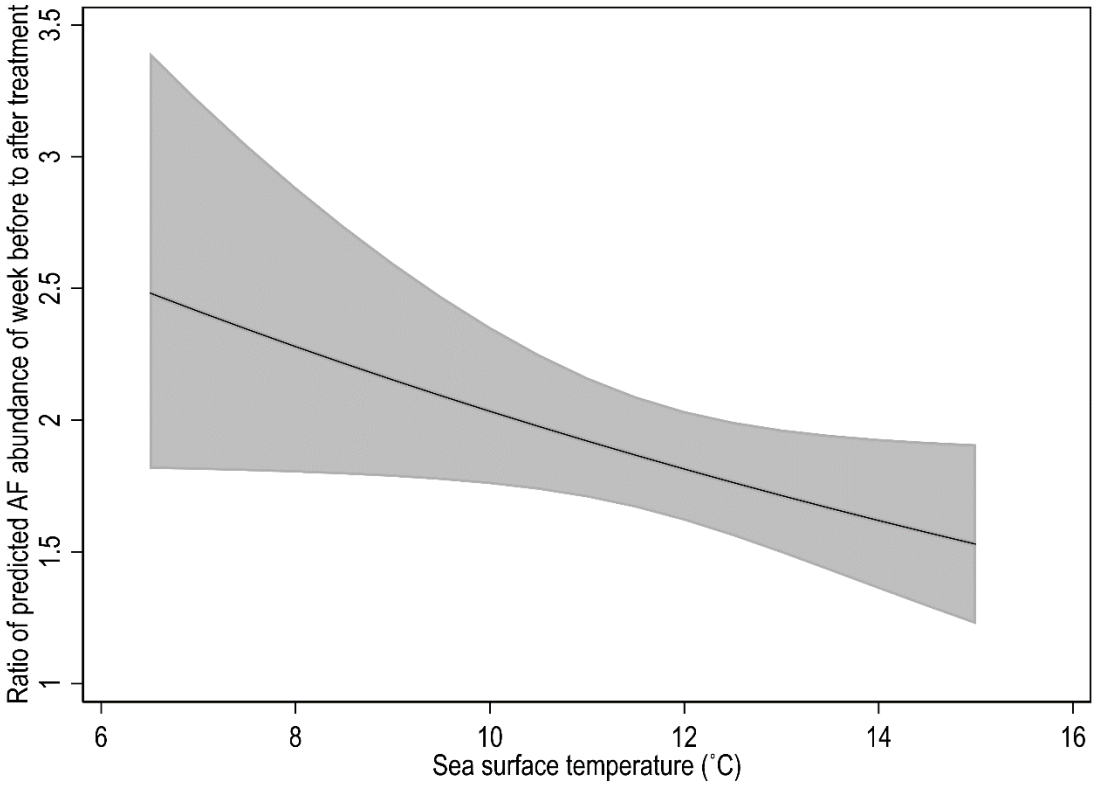

Figure 5.4. Ratio of the predicted abundance of the week before to the week after mechanical treatment (thermal and Hydrolicer®) of adult female (AF) *Lepeophtheirus salmonis* sea lice against sea surface temperature in the Bay Management Area (BMA) 1 for mean values of the other continuous predictors. Estimates were produced using the final multivariable autoregressive linear mixed effects model (AR[2]).

The $Week_{MTx}$ was divided into thermal and Hydrolicer® treatments to evaluate their respective effects further. The likelihood ratio test was non-significant (Chi-square, $p = 0.726$); therefore, there was no evidence for splitting the category $Week_{MTx}$ into its component treatments.

Chemical treatments were borderline significant in the final model (Wald Chi square test, $p = 0.042$) and did not interact with temperature (Wald Chi square test, $p = 0.567$). The $Week_{CTx}$ resulted in a slight relative increase (1.1 %) in AF abundance compared to $Week_{MB}$. The $Week_{CA}$ had a relative decrease in the abundance of AF by 12.1 % compared to the $Week_{CB}$.

In-feed treatments were significant in the model (Wald Chi square test, $p < 0.001$). There was no significant interaction between in-feed treatments and temperature (Wald Chi square test, $p = 0.480$). There was a relative increase in the abundance of AF by 5.3 % compared to the in-feed treatment week of application and the week prior to application ($Week_{ITx}$), when other predictors were kept constant.

5.4.2.2.3 Time at sea

Counts were performed at a median of 46 weeks at sea; the maximum was 123 weeks at sea. A fractional polynomial model suggested transformations with powers of -2 and 0, and 0.5. The square root transformation of TAS was selected for its parsimony, and it provided an improved fit compared to the linear scale (ΔAIC = 42). The model with the TAS random slopes did not improve the model fit (likelihood ratio test Chi-square with 2 degrees of freedom = 1.37, $p = 0.505$).

There was a relative increase in AF abundance of 52.5 % for one SD increase in $TAS^{0.5}$ when other predictors were kept constant ($p < 0.001$). The estimated abundance of AF increased over 4 to 121 weeks at sea as shown for BMA 1 during $Week_{MA}$, $Week_{CO}$, and $Week_{IO}$, and at average values of the continuous predictors (Figure 5.5).

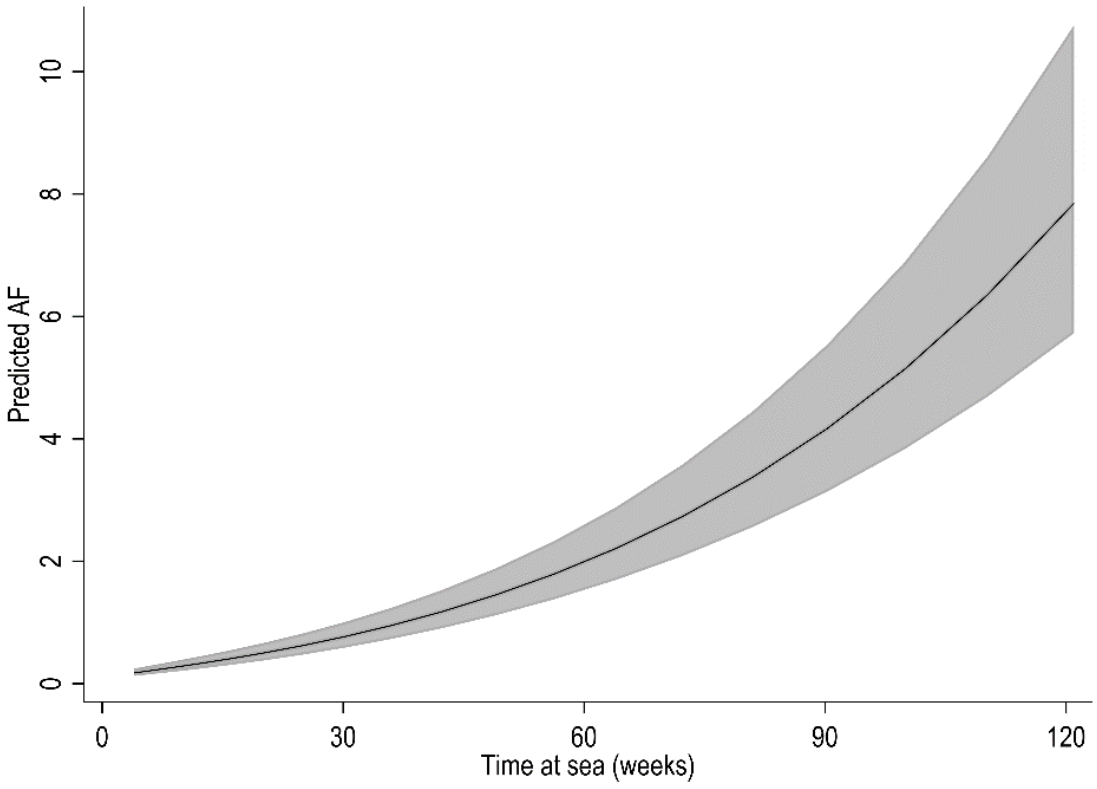

Figure 5.5. Estimated mean adult female (AF) *Lepeophtheirus salmonis* sea lice against the time at sea (TAS) in weeks in the Bay Management Area (BMA) 1 for mean values of the continuous predictors after mechanical treatment was applied. Estimates were produced using the final multivariable autoregressive linear mixed effects model (AR[2]).

5.4.2.3 Random effects

The random effects parameters included the site-cycle variance and AR(2) structure of residual errors with three temperature profiles. The site-cycle

variance of the errors was 0.08 (95% CI 0.02-0.32). The first AR coefficient (Φ_1) decreased with increasing temperature profiles (Table 5.3). The second AR coefficient (Φ_2) was highest for the 11-12 °C category, followed by the 10 °C and 6-9 °C categories. The variance was largest for the Φ_2 6-9 °C (47.96 %) and Φ_2 10 °C (34.00 %). The estimated variances of the errors were largest for the lowest temperature category, followed by the 10 °C and 11-12 °C categories.

Table 5.3. Temperature profiles for the AR(2) correlation structure to the errors of the multivariable autoregressive linear mixed effects model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice; n = 3,137 site-weeks.

Temperature category	Estimate	95 % Confidence interval
6-9 °C		
Φ_1	0.775	0.671, 0.879
Φ_2	0.085	-0.024, 0.194
Variance of the errors	1.221	0.902, 1.652
10 °C		
Φ_1	0.679	0.626, 0.733
Φ_2	0.152	0.960, 0.209
Variance of the errors	0.920	0.765, 1.106
11-12 °C		
Φ_1	0.546	0.480, 0.612
Φ_2	0.274	0.203, 0.346
Variance of the errors	0.954	0.731, 1.244

5.5 Discussion

5.5.1 Predicting the abundance of adult female *L. salmonis*

The objective was to illustrate the predictive capability of a model of the abundance of AF sea lice in the Bay of Fundy, NB. The model constructed was a multivariable autoregressive linear mixed effects model (AR[2]) of the abundance of AF sea lice. The illustration was accomplished with an in-sample and short-term weekly prediction for several sites during the summer of 2020. The predictions had good accuracy, which was expected with an in-sample approach. This model could serve as a basis for managers and farmers to make decisions about managing their salmon, such as applying a treatment considering the predicted rise in the abundance of AF sea lice.

The predictions sometimes over-estimated and sometimes under-estimated the observed AF *L. salmonis* abundance at varying weeks in the prediction horizon and varied by site-cycle. This overestimation of abundance was also observed in the model and predictions from Aldrin et al. (2017). These authors proposed that the stochastic nature of infestation and other variables, such as salinity, affected the ability of the model to predict the abundance of sea lice accurately.

The use of observed values for the calculation of residuals did not improve much the model's prediction accuracy compared to the method of calculation of the residuals without the use of observed values (set at zero). The fixed and random effects of the predictions drove the prediction to a greater degree than the effects of the correlation parameters. The prediction with the largest accuracy was in the

first week. There was no increase in the prediction error along the prediction horizon; this finding was likely due to the small dataset for prediction across 8 weeks (n = 14 sites) with a high level of variability.

The evaluation of a site in the BMA 1 demonstrated a situation with a much poorer prediction outcome (data not shown). The stocking of this site occurred close to the date selected for the start of prediction (week 36 of the year 2020), and there were already high abundances of sea lice. The model's fixed effects could not predict this high abundance of AF lice well because a more considerable value for the predictor $TAS^{0.5}$ was required for a suitably high prediction. The random effects for the site-production cycle increased the estimate of the AF abundance, but it was too slight for it to be fully compensated.

Implementing this model for prediction purposes would require reliable prediction of the temperature used in the model (Elghafghuf et al., 2021). The prediction of infestation pressures would be necessary for the use of this model for a prediction horizon greater than 4 weeks as the IIP_{AF} and EIP_{AF} were dependent on the abundance of AF sea lice with a 3-week lag, and IIP_{PAAM} was dependent on the abundance of PAAM with a 4-week lag. Predictions could be produced for scenarios without treatment applications, or various scenarios could be produced according to topical and/or in-feed treatments.

5.5.2 Contributions of internal and external infestation pressure

The infestation pressures were estimated for their use in the predictive model with the dataset 2016-2021. The infestation pressure among sites (EIP_{AF}) and

within sites (IIP_{AF} and IIP_{PAAM}) were important predictors for the abundance of AF sea lice. However, the effect of the IIP_{AF} had to be evaluated with alternate structures to the residual errors. The standardized estimated coefficient for IIP_{AF} in the final model was null (0, 95 % CI -0.04, 0.04), where the AR absorbed the effect of the IIP_{AF}. The null IIP_{AF} matches the null effect of IIP estimated in the state space model produced for Grand Manan (Elghafghuf et al., 2020). However, the effect was significant for IIP_{AF} in the model with the AR(1) and independent residual error structures, which highlights a technical aspect rather than biological aspect of the significance of the IIP_{AF} in the model. The IIP_{AF} standardized estimated coefficient for the model with the AR(1) was 0.08 (95 % CI 0.04, 0.11) and more significant with the independent structure (standardized estimated coefficient 0.32, 95 % CI 0.28, 0.35). The IIP_{AF} standardized estimated coefficient for the model with the Fish-iTrends® data for 2009-2018 was between the estimates for the AR(1) and independent structure of residual errors (Chapter 3). In the region of Grand Manan, Elghafghuf et al. (2021) found that their predictive model had negligible effects from the IIP on the outcome of the abundance of PAAM.

The standardized estimated coefficient of EIP_{AF} was 0.05 (95 % CI 0.02, 0.08) for the final model. The effect of the EIP_{AF} smaller effect than the IIP_{AF} standardized estimated coefficients with the AR(1) and independent structures of residual errors. The finding that IIP_{AF} was greater than the effect of EIP_{AF} duplicates the conclusion for the model produced with the Fish-iTrends® data for 2009-2018 (Chapter 3). The larger effect of IIP compared to EIP was observed in other

studies (Adams et al., 2012; Aldrin et al., 2013). This finding may be explained by a reproductive output of AF sea lice that increases over time at a site, resulting in self-infestation (concept of positive feedback) greater than the infestation force produced by the dispersal of planktonic lice from neighbouring farms. It is also possible that the effect of the IIP_{AF} was larger due to the duration of treatments, as it was noted that many treatment applications occur over one week at a site. The opposite relationship between EIP_{AF} and IIP_{AF} has been observed in other studies in Grand Manan (Elghafghuf et al., 2020) and Chile (Kristoffersen et al., 2013). The standardized estimated coefficient of EIP_{AF} was smaller than previously reported (Chapter 3). The difference in the strength of the effects of EIP and IIP among studies may be due to the divergence in methodology for estimating these predictors. For instance, the mean temperature increased from 2016 to 2021 compared to the mean temperature of 10.9 °C (SD 3.0) from 2009 to 2018, and this difference highlights the fact that despite no evidence of an interaction, there may be unmeasured confounders that could have changed between the two timelines and inclusion criteria.

The standardized estimated coefficient of IIP_{PAAM} (0.11, 95 % CI 0.08, 0.14) was larger than the IIP_{AF} in the final model and the model with the AR(1) structure of the residual errors but smaller than the model with the independent structure. Compared to the previously reported IIP_{PAAM} for 2009-2018, the standardized estimated coefficient was larger, but the 95 % CI overlapped from 0.08 to 0.10 (Chapter 3). The significance of this predictor in the current and previous study

(Chapter 3) suggested that the estimation of infestation pressures should consider both AF and PAAM within a site.

The contributions of IIP larger than EIP to the abundance of AF sea lice imply that the management of sea lice in the Bay of Fundy, NB, should primarily focus on within-site control of sea lice. The improvement of mitigation strategies, for example, the application of treatments to all active cages at a site over a shorter time interval, may decrease the IIP by decreasing the planktonic sea lice burden and reducing self-infestation (Aldrin et al., 2013). However, farmers should remember that their neighbours contribute to their site lice infestation. Mitigation strategies to reduce EIP may include synchronizing in-feed treatments and continued use of fallowing periods (Aldrin et al., 2013; Kristoffersen et al., 2013, 2014). The synchronization of in-feed treatments on the potential for the development of resistance is unknown (Arriagada et al., 2017). However, the risk of resistance may be decreased as the time to next treatment may be extended with synchronization (Arriagada et al., 2017) reducing the sea lice exposure to sub-lethal levels of the chemotherapeutant, which may increase the risk of resistance development (Lam et al., 2020).

5.5.3 Treatments and temperature

The treatments applied between 2016 and 2021 were included in the model to control for their potentially confounding effects, and the study was not explicitly designed to evaluate the efficacy of the treatments, per se. Additionally, the model accounted for multiple other predictor variables that make it difficult to

understand the treatment effects completely. Despite this, the mechanical, chemical, and in-feed treatments significantly affected the estimated abundance of AF in the model.

The effects of mechanical treatments varied by temperature. The ratio of predicted abundance of the mechanical treatments for the $Week_{MB}$ to $Week_{MA}$ demonstrated decreased effectiveness with increasing temperature (Figure 5.4). The relationship between thermal (i.e., warm water) application and temperature was previously noted (Chapter 3). The Hydrolicer® application and temperature association was weak in the previously published model (95 % CI crosses zero in Chapter 3). In Chapter 3, we proposed that the diminished capacity of warm water treatment to reduce the abundance of AF was due to the acclimation of sea lice. In other words, during the spring and early summer at lower temperatures, the thermal treatment seemed to cause a greater shock to the sea lice, potentially because of a larger temperature difference compared to sea lice in the late summer and early fall that were acclimated to higher temperatures. Borchel et al. (2018) demonstrated that the nauplii of *L. salmonis* have thermotolerance as they can survive rapid increases in temperature if exposed to an intermediate temperature prior to heat shock. The effect of thermal treatments for delousing was recently evaluated (Nilsson et al., 2023). The employed range was a 20-22.5 °C difference between the seawater and the treatment water temperature, per the Norwegian industry guidelines (28-34 °C; Nilsson et al., 2023). In the study area, there was a difference of 12-21 °C at low temperature

and 18-27 °C at high temperature (based on UKMO SST 6.9 to 16.0 °C during days when thermal treatments were applied).

Chemical treatments did not vary by temperature in this model, as was observed with hydrogen peroxide in Chapter 3. The combination of hydrogen peroxide with azamethiphos might have masked these temperature variable effects. Bioassays for hydrogen peroxide exposure of *L. salmonis* at temperatures of 5 and 10 °C demonstrated a slight decrease in survival from 73 % to 67 %, respectively (VanIderstine et al., 2020). In Chapter 3, the effects of azamethiphos were categorized per method of application (tarpaulin, skirt, and wellboat) and demonstrated variable effects concerning temperature. There were minimal changes to the sensitivity of *C. rogercresseyi* to azamethiphos in the temperature range 12 to 15 °C with a more effective concentration value at 9 °C (González-Gómez et al., 2023).

The temperature was a significant predictor for sea lice abundance and the development rate of the louse (Samsing et al., 2016; Hamre et al., 2019) and infestation success was dependent on temperature (Samsing et al., 2016). Additionally, the temperature may direct mitigation strategies, such as treatment dosage and exposure time (Overton et al., 2017, 2019). Increasing temperatures were associated with higher mortality after treatment (Overton et al., 2019).

This study design improved the ability to observe the effects of in-feed treatments compared to the 2009-2018 study with the segmented data organization (Chapter 3). In-feed treatments were typically applied early in the production

cycle, and this predictor may capture the first period at sea. It is possible that the association observed between sea lice counts and the in-feed treatments may be confounded by the time at sea. The in-feed treatments may also be influenced by the application of lufenuron (Imvixa™, Elanco™ Animal Health Inc., USA) to smolt at the hatcheries immediately prior to stocking, for which we have no records of such hatchery-based treatments in the database.

Considering these findings, mechanical and chemical treatments reduced the abundance of AF when the week before and after treatment were compared. The mechanical treatments' ability to reduce the abundance of AF should be further evaluated at various temperatures. The efficacy of treatments should be evaluated with other methods than presented in this study, such as Gautam et al. (2017b) for the study area. This information would guide veterinarians in prescribing treatments for those more efficacious at the measured temperature at the time of application.

5.5.4 Time at sea

The abundance of AF increased with TAS as observed by others (e.g., Revie et al., 2002). The TAS was a continuous variable and may represent the growth of the salmon as the fish increased in size (weight and surface area) and age. The TAS was equivalent to the 'count week' used by Kristoffersen et al. (2014), a significant predictor and EIP in the abundance of PAAM. Other studies have mixed findings of an association between the abundance of sea lice and the size and age of fish (for review, see Frenzl, 2014). In short, the size of Atlantic salmon

may affect the settlement of sea lice where larger salmon are easier for the louse to identify in the water column for infestation, present a greater surface area for attachment, and decrease the competition for space (Frenzl, 2014). The age of the salmon may also affect the settlement of lice. Transport and smoltification (the period of morphological and physiological changes that occur in anadromous fish allowing them to survive in saltwater) increase the susceptibility of young smolt to stress and infection, while post-smolt demonstrated decreased stress responses compared to parr (Madaro et al., 2016). The swimming depth of the fish may also predispose younger fish to infestation, as younger fish (0.5 kg) swam at shallower depths compared to older fish (4.0 kg; 2-5 m and 7-10 m, respectively) during the day (Korsøen et al., 2012). Smaller fish have higher densities of sea lice (Tucker et al., 2002), which is a function of the count of sea lice per the surface area of the fish; for a large and a small fish with the exact count of sea lice, the small fish will have a greater density of sea lice because its surface area is smaller.

Another predictor that may be used to represent the growth of salmon in the model was the individual weight of the salmon (Todd et al., 2000; Glover et al., 2004) and the average weight for the group (Kristoffersen et al., 2013). However, these data were not available in the Fish-iTrends® database. A predictor for the age/size of the salmon, using an earlier extraction of the dataset, was also not included in Chapter 3 and among other studies (e.g., Amundrud and Murray, 2009; Elghafghuf et al., 2020, 2021).

5.5.5 Winter

The ‘without winter gaps’ time series was applied to the final model to evaluate the effect of correlation in time. Where this time series did not improve the model fit and the coefficients did not change considerably the counts of sea lice before and after these large gaps in time, the observations before and after the winter period were deemed independent.

5.5.6 Study design and limitations

5.5.6.1 Study design

This study developed a predictive model using estimated infestation pressures in the Bay of Fundy, NB, using empirical AF sea lice abundances for the years 2016-2021 from the Fish-iTrends® database. The multivariable autoregressive linear mixed effects model was selected as it is well suited to the repeated measures of longitudinal data with a hierarchical structure. Moreover, the mixed effects model is relatively simple and could be employed for prediction purposes that may benefit salmon aquaculture veterinarians and managers. A single equation model was selected for simplicity; however, it could be argued that a model with equations for the first year at sea (defined as stocking to first winter at sea) and the second to third year at sea (defined as after the first year at sea until harvesting) may be better suited. This avenue would have the advantage of separating the first year at sea from the other production years as it may be different for several reasons, including higher abundance for the second year at sea populations (Revie et al., 2002; Saksida et al., 2007), more in-feed

treatments and fewer topical treatments, and the possible in-feed chemotherapeutic effects of lufenuron that may have been applied at the hatchery prior to stocking the smolt (2017-2020). Other studies have utilized multiple equation models (e.g., Kristoffersen et al., 2013; Aldrin et al., 2019). Aldrin et al. (2019) selected a multivariate zero-inflated negative binomial distribution that treated adult females and other mobile lice as separate time series because they have different parameter estimates. The disadvantage of the multiple equation models would be the added complexity during prediction and decreased apprehensibility by the industry.

The time series utilized all counts and contained many time gaps. The mixed model was appropriate for managing time gaps without imputing sea lice counts, as Elghafghuf et al. (2020) did when using state-space models in the Grand Manan region. Limiting the data to consecutive counts, as employed by Kristoffersen et al. (2014), would not be possible with our dataset, as it would severely limit the sample size and potentially introduce a selection bias into the study.

The model's residual error structure was complex; it was an autoregressive second-order with three temperature profiles. The null IIP_{AF} in the AR(2) model reflected a lack of ability to pick up on the variation in explained terms. Other residual error structures were evaluated. The study with the Fish-iTrends® data for 2009-2018 utilized an exponential structure with three temperature profiles to its residual errors (Chapter 3). The exponential structure functions similarly to the AR(1) (Simpson et al., 2010). For the region of Grand Manan, an AR(1) function

was found suitable for the development of AF (Elghafghuf et al., 2020). Elghafghuf et al. (2018) remarked that high-order autoregressive structures may be necessary to develop juvenile lice into adults and produce offspring from the adults. The temperature categories for the structure of the residual errors were different in this model compared to the previous model as the categories must be constructed from the average temperature at the lowest hierarchical level, which in this case is the site-cycle as compared to the segment in Chapter 3.

The repeated counts of sea lice were averaged weekly for all the cages at a site, irrespective of the manager-contributed label of a count occurring before and after treatment (i.e., 'pre-treatment' and 'post-treatment' counts, respectively). The labels were effectively ignored as an entire week (Monday to Sunday) was categorized as a treatment application. The consolidation of 'pre-treatment', 'post-treatment' and regular weekly counts may result in reversion to the mean and decrease the variability of the outcome and infestation pressures. Comparatively, Elghafghuf et al. (2020) were more temporally precise in identifying the treatments. They limited the mean abundance of lice to the counts labelled 'post-treatment' if they occurred the same week as a topical treatment.

5.5.6.2 Limitations

The present multivariable autoregressive linear mixed effects model was used to predict the abundance of AF *L. salmonis* with reasonable accuracy. This model did not account for multiple sources of variation (e.g., latent and observation errors) as was achieved with a state-space model in Grand Manan, NB

(Elghafghuf et al., 2021). Like that model, our model did not include parameters for the larval stage (Elghafghuf et al., 2021). Adding predictors such as seawater salinity and parameters that vary per larval stage may improve the prediction accuracy and decrease prediction variability.

The estimates of the infestation pressures from this model for the date range 2016-2021 differed from those previously published for 2009-2018 (Chapter 3). These results highlighted the variability in the estimates of infestation pressures according to the different methodologies and datasets (e.g., Kristoffersen et al., 2014; Cantrell et al., 2019). The addition of an autocorrelation structure to the residual errors may have masked the actual effect of IIP vs. EIP, making the comparison between them difficult to fully assess due to a combination of autocorrelation of counts and the 'entangling' of life stages in the autocorrelation. The ‘segment’ approach (Chapter 3) may better identify the effect of IIP vs EIP, as an estimate for the IIP_{AF} was observed from that model.

The evaluation of the time series for this study demonstrated a possible selection of cages for counts. For example, the same cage with high sea lice abundance was counted twice, resulting in a large weekly average. This may affect the averaging at the site and week level and produce selection and information biases that may dilute the observed treatment effects.

5.6 Conclusions

A mixed autoregressive model was constructed to predict the abundance of AF sea lice and estimate infestation pressures that affect the abundance of lice in

New Brunswick, Canada. The model accounted for topical and in-feed treatments, time at sea, and BMA while modelling the abundance of AF sea lice. The model had a good level of accuracy for the prediction horizon of 4 weeks and could be considered for further development and may aid in decision-making processes for the management of sea lice on salmon farms in the Bay of Fundy, NB.

The AF abundance is complex at a site and impacted by multiple factors, including the contributions of IIP and EIP. Farmers and managers of aquaculture sites can utilize these results to better plan mitigation strategies at their sites and within Bay Management Areas. Mitigation strategies for sea lice should focus on the site while monitoring for trends of sea lice from their neighbours to adjust and maximize the impact of their mitigation strategies.

5.7 References

Adams, T., Black, K., Macintyre, C., Macintyre, I., and Dean, R. (2012). Connectivity modelling and network analysis of sea lice infection in Loch Fyne, west coast of Scotland. Aquaculture Environment Interactions, 3, 51-63. https://doi.org/10.3354/aei00052

Aldrin, M., Huseby, R. B., Stien, A., Grøntvedt, R. N., Viljugrein, H., and Jansen, P. A. (2017). A stage-structured Bayesian hierarchical model for salmon lice populations at individual salmon farms - Estimated from multiple farm data sets. Ecological Modelling, 359, 333-348. http://dx.doi.org/10.1016/j.ecolmodel.2017.05.019

Aldrin, M., Jansen, P. A., and Stryhn, H. (2019). A partly stage-structured model for the abundance of salmon lice in salmonid farms. Epidemics, 26, 9-22. https://doi.org/10.1016/j.epidem.2018.08.001

Aldrin, M., Storvik, B., Kristoffersen, A. B., and Jansen, P. A. (2013). Space-time modelling of the spread of sea lice between and within Norwegian marine salmon farms. PLoS ONE, 8, 1-10. https://doi.org/10.1371/journal.pone.0064039

Amundrud, T. L., and Murray, A. G. (2009). Modelling sea lice dispersion under varying environmental forcing in a Scottish sea loch. Journal of Fish Diseases, 32, 27-44. https://doi.org/10.1111/j.1365-2761.2008.00980.x

Arriagada, G., Stryhn, H., Sanchez, J., Vanderstichel, R., Campistó, J. L., Rees, E. E., Ibarra, R., and St-Hilaire, S. (2017). Evaluating the effect of synchronized sea lice treatments in Chile. Preventive Veterinary Medicine, 136, 1–10. https://doi.org/10.1016/j.prevetmed.2016.11.011

Bhaskaran, K., Gasparrini, A., Hajat, S., Smeeth, L., and Armstrong, B. (2013). Time series regression studies in environmental epidemiology. International Journal of Epidemiology, *42*(4), 1187-1195. https://doi.org/10.1093/ije/dyt092

Borchel, A., Komisarczuk, A. Z., Rebl, A., Goldammer, T., and Nilsen, F. (2018). Systematic identification and characterization of stress-inducible heat shock proteins (HSPs) in the salmon louse (*Lepeophtheirus salmonis*). Cell Stress and Chaperones, 23(1), 127-139. https://doi.org/10.1007/s12192-017-0830-9

Bui, S. (2016). The behavioural resistance and response of Atlantic salmon to the ectoparasite *Lepeophtheirus salmonis*. Doctoral thesis, University of Melbourne, Australia. http://hdl.handle.net/11343/127923

Cantrell, D., Filgueira, R., Revie, C. W., Rees, E., Vanderstichel, R., Foreman, M. G. G., … Grant, J. (2019). The relevance of larval biology on spatiotemporal patterns of pathogen connectivity among open-marine salmon farms. Canadian Journal of Fisheries and Aquatic Sciences, 77, 505-519. https://doi.org/10.1139/cjfas-2019-0040

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2021). State-space modeling for inter-site spread of sea lice with short-term population predictions. Ecological Modelling, 452. https://doi.org/10.1016/j.ecolmodel.2021.109602

Elghafghuf, A., Vanderstichel, R., St-Hilaire, S., and Stryhn, H. (2018). Using state-space models to predict the abundance of juvenile and adult sea lice on Atlantic salmon. Epidemics, 24, 76-87. https://doi.org/10.1016/j.epidem.2018.04.002

Elmoslemany, A., Whyte, S. K., Revie, C. W., and Hammell, K. L. (2013). Sea lice monitoring on Atlantic salmon farms in New Brunswick, Canada: Comparing audit and farm staff counts. Journal of Fish Diseases, 36(3), 241-247. https://doi.org/10.1111/jfd.12051

Frenzl, B. (2014). Understanding key factors associated with the infection of farmed Atlantic salmon by the salmon louse *Lepeophtheirus salmonis*. Doctoral dissertation, University of Stirling. http://hdl.handle.net/1893/22320

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017a). Effect of timing of count events on estimates of sea lice abundance and interpretation of effectiveness following topical treatments. Journal of Fish Diseases, 40, 367-375. https://doi.org/10.1111/jfd.12519

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., & Hammell, K. L. (2017b). Evaluating bath treatment effectiveness in the control of sea lice burdens on Atlantic salmon in New Brunswick, Canada. Journal of Fish Diseases, 40(7), 895–905. https://doi.org/10.1111/jfd.12569

Glover, K. A., Nilsen, F., and Skaala. (2004). Individual variation in sea lice (*Lepeophtheirus salmonis*) infection on Atlantic salmon (*Salmo salar*). Aquaculture, 241(1-4), 701-709. https://doi.org/10.1016/j.aquaculture.2004.07.030

González-Gómez, M. P., Spinetto, C., Oyarzo, C., Álvarez, D., Oyarzún, R., & Rivas, M. (2023). Effects of temperature on azamethiphos sensitivity profile of *Caligus rogercresseyi*. Aquaculture, 575. https://doi.org/10.1016/j.aquaculture.2023.739824

Government of New Brunswick. (2020). General Regulation, NB Reg 91-158, 1-14. http://canlii.ca/t/544vm

Hamre, L., Bui, S., Oppedal, F., Skern-Mauritzen, R., and Dalvin, S. (2019). Development of the salmon louse *Lepeophtheirus salmonis* parasitic stages in

temperatures ranging from 3 to 24°C. Aquaculture Environment Interactions, 11, 429-443. https://doi.org/10.3354/aei00320

Jones, P. G., Hammell, K. L., Dohoo, I. R., and Revie, C. W. (2012). Effectiveness of emamectin benzoate for treatment of *Lepeophtheirus salmonis* on farmed Atlantic salmon *Salmo salar* in the Bay of Fundy, Canada. Diseases of Aquatic Organisms, 102(1), 53-64. https://doi.org/10.3354/dao02517

Katsouyanni, K., Schwartz, J., Spix, C., Touloumi, G., Zmirou, D., Zanobetti, A., Wojtyniak, B., Vonk, J. M., Tobias, A., Ponka, A., Medina, S., Bacharova, L., and Anderson, H. R. (1996). Short term effects of air pollution on health: a European approach using epidemiologic time series data: the APHEA protocol. Journal of Epidemiology and Community Health, 50(1): 12-18.

Korsøen, Ø. J., Dempster, T., Oppedal, F., & Kristiansen, T. S. (2012). Individual variation in swimming depth and growth in Atlantic salmon (*Salmo salar* L.) subjected to submergence in sea-cages. Aquaculture, 334–337, 142–151. https://doi.org/10.1016/j.aquaculture.2011.12.031

Kristoffersen, A. B., Rees, E. E., Stryhn, H., Ibarra, R., Campisto, J. L., Revie, C. W., and St-Hilaire, S. (2013). Understanding sources of sea lice for salmon farms in Chile. Preventive Veterinary Medicine, 111, 165-175. https://doi.org/10.1016/j.prevetmed.2013.03.015

Kristoffersen, A. B., Jimenez, D., Viljugrein, H., Grøntvedt, R., Stien, A., and Jansen, P. A. (2014). Large scale modelling of sea lice (*Lepeophtheirus salmonis*) infection pressure based on lice monitoring data from Norwegian salmonid farms. Epidemics, 9, 31-39. https://doi.org/10.1016/j.epidem.2014.09.007

Lam, C. T., Rosanowski, S. M., Walker, M., and St-Hilaire, S. (2020). Sea lice exposure to non-lethal levels of emamectin benzoate after treatments: A potential risk factor for drug resistance. Scientific Reports, 10(1). https://doi.org/10.1038/s41598-020-57594-7

Madaro, A., Olsen, R. E., Kristiansen, T. S., Ebbesson, L. O. E., Flik, G., & Gorissen, M. (2016). A comparative study of the response to repeated chasing stress in Atlantic salmon (*Salmo salar* L.) parr and post-smolts. Comparative Biochemistry and Physiology – Part A: Molecular and Integrative Physiology, 192, 7–16. https://doi.org/10.1016/j.cbpa.2015.11.005

Myksvoll, M. S., Sandvik, A. D., Albretsen, J., Asplin, L., Johnsen, I. A., Karlsen, Ø., Kristensen, N. M., Melsom, A., Skardhamar, J., and Ådlandsvik, B. (2018). Evaluation of a national operational salmon lice monitoring system – From physics to fish. PLoS ONE, 13(7), 1-25. https://doi.org/10.1371/journal.pone.0201338

Nilsson, J., Barrett, L. T., Mangor-Jensen, A., Nola, V., Harboe, T., and Folkedal, O. (2023). Effect of water temperature and exposure duration on detachment rate

of salmon lice (*Lepeophtheirus salmonis*); testing the relevant thermal spectrum used for delousing. Aquaculture, 562. https://doi.org/10.1016/j.aquaculture.2022.738879

Overton, K., Dempster, T., Oppedal, F., Kristiansen, T. S., Gismervik, K., and Stien, L. H. (2019). Sea lice treatments and salmon mortality in Norwegian aquaculture: a review. Reviews in Aquaculture, 11, 1398-1417. https://doi.org/10.1111/raq.12299

Overton, K., Samsing, F., Oppedal, F., Stien, L. H., and Dempster, T. (2017). Lowering treatment temperature reduces salmon mortality: a new way to treat with hydrogen peroxide in aquaculture. Pest Management Science, 74, 535-540. https://doi.org/10.1002/ps.4751

Pinheiro, J. C., and Bates, D. M. (2000). Mixed-Effects Models in S and S-PLUS. In J. Chambers, W. Eddy, W. Hardle, S. Sheather, L. Tierney (Eds.), Statistics and Computing (1st ed.). Springer-Verlag New York, Inc. 528p.

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

Revie, C. W., Gettinby, G., Treasurer, J. W., Rae, G. H., and Clark, N. (2002). Temporal, environmental and management factors influencing the epidemiological patterns of sea lice (*Lepeophtheirus salmonis*) infestations on farmed Atlantic salmon (*Salmo salar*) in Scotland. Pest Management Science, 58(6), 576-584. https://doi.org/10.1002/ps.476

Rosner, B., Muñoz, A., Tager, I., Speizer, F., and Weiss, S. (1985). The use of an autoregressive model for the analysis of longitudinal data in epidemiologic studies. Statistics in Medicine, 4(4), 457-467. https://doi.org/10.1002/sim.4780040407

Saksida, S., Constantine, J., Karreman, G. A., and Donald, A. (2007). Evaluation of sea lice abundance levels on farmed Atlantic salmon (*Salmo salar* L.) located in the Broughton Archipelago of British Columbia from 2003 to 2005. Aquaculture Research, 38(3), 219-231. https://doi.org/10.1111/j.1365-2109.2007.01651.x

Samsing, F., Oppedal, F., Dalvin, S., Johnsen, I., Vågseth, T., and Dempster, T. (2016). Salmon lice (*Lepeophtheirus salmonis*) development times, body size, and reproductive outputs follow universal models of temperature dependence. Canadian Journal of Fisheries and Aquatic Sciences, 73, 1841-1851. https://doi.org/10.1139/cjfas-2016-0050

Simpson, S. L., Edwards, L. J., Muller, K. E., Sen, P. K., and Styner, M. A. (2010). A linear exponent AR(1) family of correlation structures. Statistics in Medicine, 29(17), 1825-1838. https://doi.org/10.1002/sim.3928

St-Hilaire, S., Patanasatienkul, T., Yu, J., Kristoffersen, A. B., Stryhn, H., Revie, C., Ibarra, R., Tello, A., and McEwan, G. (2018). Tool for predicting *Caligus*

rogercresseyi abundance on salt water salmon farms in Chile. Preventative Veterinary Medicine, 158, 122-128. https://doi.org/10.1016/j.prevetmed.2018.08.001

StataCorp (2021). Stata: Release 17. Statistical Software. StataCorp LLC, TX, USA.

Todd, C. D., Walker, A. M., Hoyle, J. E., Northcott, S. J., Walker, A. F., and Ritchie, M. G. (2000). Infestations of wild adult Atlantic salmon (*Salmo salar* L.) by the ectoparasitic copepod sea louse *Lepeophtheirus salmonis* Krøyer: Prevalence, intensity and the spatial distribution of males and females on the host fish. Hydrobiologia, 429(1-3), 181-196. https://doi.org/10.1023/a:1004031318505

Tucker, C. S., Sommerville, C., and Wootten, R. (2002). Does size really matter? Effects of fish surface area on the settlement and initial survival of *Lepeophtheirus salmonis*, an ectoparasite of Atlantic salmon *Salmo salar*. Diseases of Aquatic Organisms, 49(2), 145-152. https://doi.org/10.3354/dao049145

UK Met Office (2005). OSTIA Level 4 Sea Surface Temperature Analysis. Ver. 1.0. PO.DAAC, CA, USA. Retrieved from https://doi.org/10.5067/GHOST-4FK01.

van Etten, J., and de Sousa, K. (2012). gdistance: Distances and Routes on Geographical Grids. R package version 1.1-3. https://CRAN.R-project.org/package=gdistance

VanIderstine, C. R., Poley, J. D., Whyte, S. K., Purcell, S. L., & Fast, M. D. (2021). Hydrogen peroxide treatment and its impacts on *Lepeophtheirus salmonis* originating from the Bay of Fundy, Canada. Journal of Fish Diseases, 44(6), 757–769. https://doi.org/10.1111/jfd.13299

Whyte, S. K., Westcott, J. D., Byrne, P., and Hammell, K. L. (2011). Comparison of the depletion of emamectin benzoate (SLICE®) residues from skeletal muscle and skin of Atlantic salmon (*Salmo salar*), for multiple dietary dose regimens at 10°C. Aquaculture, 315(3-4), 228-235. https://doi.org/10.1016/j.aquaculture.2011.02.043

WoRMS Editorial Board (2022). World Register of Marine Species. Available from https://www.marinespecies.org at VLIZ. https://doi.org/10.14284/170

5.8 Supplemental information

5.8.1 Temperature

The UKMO OSTIA SST product (UK Met Office, 2005) was utilized for the sea surface temperature (hereafter, temperature) of the study area. This remote sensing product was selected instead of the *in-situ* temperature measurements to avoid missing values and inconsistencies in measuring technique. Thakur et al. (2018) found that the UKMO OSTIA SST product had the highest concordance correlation coefficient and index of agreement compared to measurements of temperature taken on aquaculture sites in the Broughton Archipelago between 2003 and 2016.

Daily temperature values were extracted for the duration of the study for the centroid location of each aquaculture site polygon utilizing the ‘raster’ package, version 3.0-12 (Hijmans et al., 2020) in R. The spatial resolution was 1/20 degrees, which is approximately 6 km at the 45 degree latitude of the Bay of Fundy, NB; sites that occupied the same temperature grid shared temperature values.

5.8.1 Treatments

A descriptive analysis was not conducted for December 2018 to June 2021 as this was for an extraction from the Fish-iTrends performed after completing the descriptive analysis reported in Chapter 2. A summary of treatments was produced (Figure 5.6) to aid during the presentation of the studies and comparison of the methods of treatment across the time range for the entire study (2009-2021). The application of treatments varied annually as observed in

Chapter 2. The total number of treatments increased from 2018 to 2020 for a total of 208 treatment events. In-feed treatments were applied during this same period and accounted for an additional 122 treatment events from 2018-2020 (Figure 5.6).

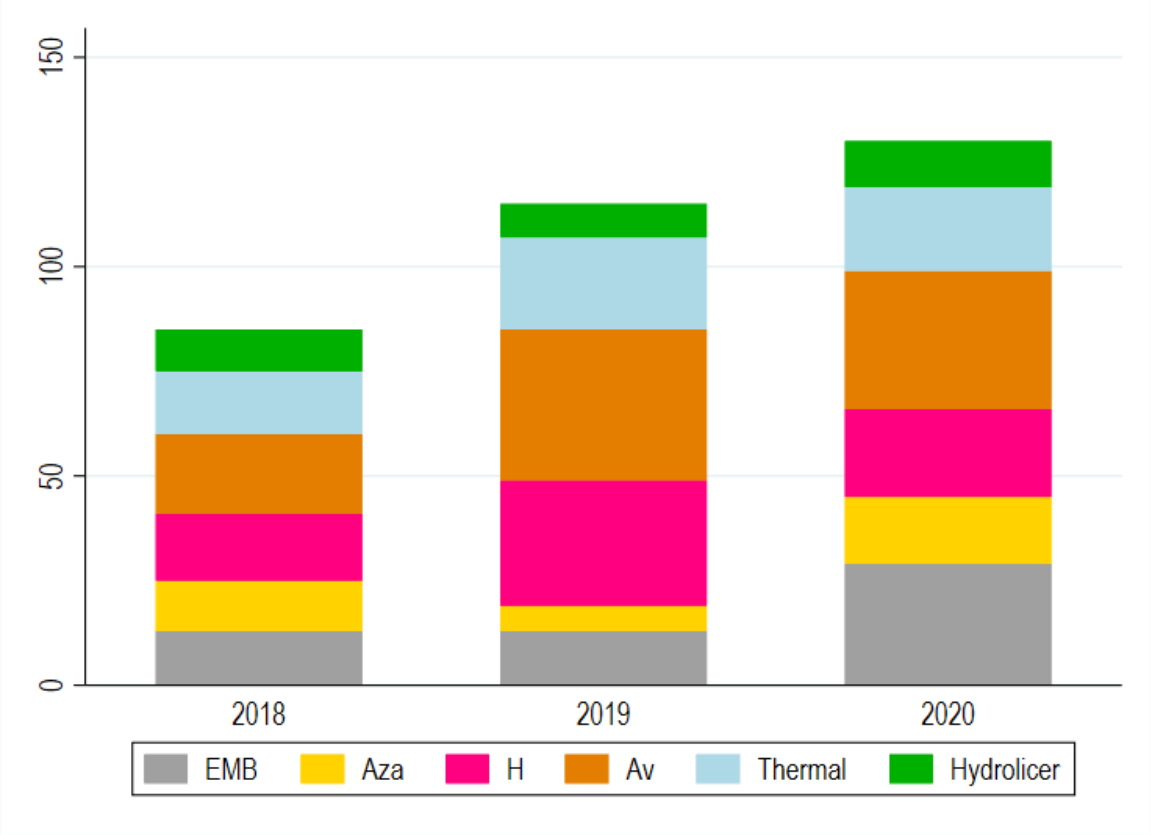

Figure 5.6. Total number of treatments applied annually in the Bay of Fundy, New Brunswick, for 2018 to 2020. EMB: emamectin benzoate, AZA: azamethiphos, H: hydrogen peroxide, Av = avermectins. n = 330

5.8.2 Internal and external infestation pressure

5.8.2.1 Equation for site-week internal infestation pressure

The abundances of *Lepeophtheirus salmonis* sea lice (adult female [AF] and pre-adult and adult male [PAAM]) were averaged for a week (Monday to Sunday, International Organization for Standardization 8601) at a site with cage weighting

(outcome and IIP_{AF}: Eq. 5.1 and IIP_{PAAM}: Eq. 5.2). There were instances where a cage was sampled multiple times during the week; therefore, cage weighting was applied. Cage-weighting did not result in a large difference to the calculated weekly average abundance of sea lice (Pearson's correlation coefficient [ρ] and concordance correlation coefficient [ρ_c] = 1.00). These site-week abundances were lagged 3 and 4 weeks to represent the IIP_{AF} and IIP_{PAAM}, respectively. There was no temporal weighting applied as was performed for the IIP in Chapter 3 because abundances were averaged weekly.

$$\overline{lnAF}_{it} = \sum_{c;s\in\Theta_t} w(c,t)\, lnAF_{ic}(s) \Big/ \sum_{c;s\in\Theta_t} w(c,t) \qquad \text{Eq. 5.1}$$

$$\overline{lnAF}_{it} = \sum_{c;s\in\Theta_t} w(c,t)\, lnPAAM_{ic}(s) \Big/ \sum_{c;s\in\Theta_t} w(c,t) \qquad \text{Eq. 5.2}$$

Where: logarithmic abundances were obtained as $lnAF = ln\left({}^{(AF+1)}\Big/_{no.fish}\right)$

c ~ cage within site i

t ~ target week, typically 3 weeks prior to an observation for AF and 4 weeks for PAAM

θ_t ~ week as defined by days s: Monday to Sunday

$w(c,t)$ ~ weight for the number of times a cage c was counted for a site i during a week t (range: 1-5 cages/site/week), $w(c,t) = {}^{1}/_{lnAF_{ct}}$

5.8.2.2 Equation for site-week external infestation pressure

The external infestation pressure for the AF was the sum of the mean abundance of sea lice from its neighbours 3 weeks prior (Eq. 5.3). A detailed description of the generation of the Gaussian kernel density estimate (KDE) spatial weights

was previously published (Chapters 3 and 4). In short, the seaway distances were calculated for the approximate centroids for all aquaculture sites extracted from the 2017 published map (Department of Energy and Resource Development/Agriculture, and Fisheries, 2017) within a corrected transition waterway layer. The Gaussian KDE function was applied to the matrix of seaway distances among sites to produce spatial weights. The weight was multiplied by the mean abundance of AF (average calculated as per Equation 5.1) for the neighbouring sites and these values were summed across the row.

Vector and raster image work was completed in QGIS (QGIS.org, 2018) with the NAD83(CSRS)/New Brunswick Stereographic (EPSG:2953) projection. Otherwise, transformations and calculations were performed in R (R Core Team, 2020) using the packages ‘rgdal’ (version 1.4-8; Bivand et al., 2019), ‘raster’ (version 3.0-12; Hijmans et al., 2020), and 'gdistance' (version 1.1-4; van Etten and de Sousa, 2012).

$$\overline{\mathrm{lnAF}_{it,N}} = \sum_{j \in N_i} w_{ij} \overline{\mathrm{lnAF}_{j,t-3}} \qquad \text{Eq. 5.3}$$

Where:

t ~ target week, 3 weeks prior to an observation for AF

i ~ site

N ~ all neighbour sites of site i, j ~ other sites

w_{ij} ~ Gaussian kernel density estimated weights (e.g., Elghafghuf et al., 2020) at bandwidth of 10 km computed based on seaway distance between site s and j

5.8.3 Final model

The final model described was a multivariable mixed effects autoregressive regression with site-cycle random effects and a second order autoregressive function structure to the errors that was stratified by three temperature profiles (Eq. 5.4). Treatments were organized by duration of effects and chemotherapeutic agents vs. mechanical (i.e., non-medicinal) operations. The equation presents the individual count for a site-cycle.

$$\begin{aligned}\overline{\ln \mathrm{AF}} = {} & \beta_0 + IIP_{AF}\beta_1 + IIP_{PAAM}\beta_2 + EIP_{AF}\beta_3 + SST\beta_4 + Week_{MO}\beta_5 \\ & + Week_{MTx}\beta_6 + Week_{MA}\beta_7 + \mathrm{SST} * Week_{MO}\beta_8 \\ & + SST * Week_{MTx}\beta_9 + SST * Week_{MA}\beta_{10} \\ & + Week_{CO}\beta_{11} + Week_{CTx}\beta_{12} + Week_{CA}\beta_{13} \\ & + Week_{PO}\beta_{14} + Week_{PA}\beta_{15} + BMA_{2A}\beta_{16} \\ & + BMA_{2B}\beta_{17} + BMA_{3A}\beta_{18} + BMA_{3B}\beta_{19} + BMA_{3C}\beta_{20} \\ & + \mathrm{TAS}^{0.5}\beta_{21} + \varepsilon + \mu\end{aligned} \qquad \text{Eq. 5.4}$$

Where:

IIP_{AF} ~ internal infestation pressure of adult female (AF) *L. salmonis* as calculated in Equation 5.1

IIP_{PAAM} ~ internal infestation pressure of pre-adult and adult male (PAAM) *L. salmonis* as calculated in Equation 5.2

EIP_{AF} ~ external infestation pressure of AF as calculated in Equation 5.3

SST ~ sea surface temperature in °C

Mechanical treatments: combined topical thermal and Hydrolicer® applications

- $Week_{MO}$: week that does not fit other criteria
- $Week_{MTx}$: week of treatment application
- $Week_{MB}$: 1 week prior to treatment week
- $Week_{MA}$: 1 week after treatment week

Chemical treatments: combined hydrogen peroxide and azamethiphos applications

- $Week_{CO}$: week that does not fit other criteria
- $Week_{CTx}$: week of treatment application
- $Week_{CB}$: 1 week prior to treatment week
- $Week_{CA}$: 1 week after treatment week

In-feed treatments: combined emamectin benzoate and avermectins oral applications

- $Week_{IO}$: week that does not fit other criteria
- $Week_{ITx}$: combined single week prior to treatment week and week of treatment application
- $Week_{IA}$: weeks 1-4 after treatment week

BMA ~ Bay Management Area

TAS ~ time at sea, in weeks

sc ~ site-cycle

β ~ coefficient parameter

ε are ~N(0, σ_{sc}^2 is dependent on g), within a site-cycle are dependent on time distance in weeks, and between site-cycle are independent

μ are the random site-cycle effects and ~N(0, σ_{sc}^2)

g ~ group of SST_{sc} and have σ_g^2

5.8.4 In-sample short term prediction

5.8.4.1 Prediction

The predictions were calculated for a total of 8 weeks after week 36 of the year 2020 using the equations presented in Pinheiro and Bates (2000 [Eq. 5.5]). The term a_t was the linear and random effects estimated abundance of AF sea lice using the multivariable mixed effects AR(2) model. The correlation parameters of the AR(2) model ($\Phi_1 \; and \; \Phi_2$) were the autoregressive coefficients for lag week 1 and lag week 2 for the high temperature category (11-12 °C). The residuals ($\hat{\epsilon}$) were calculated from the difference of the observed abundance of AF sea lice to the prediction of that week or set to zero. Setting the residuals to zero was done to examine the effects of the estimated abundance without the influence of the correlation parameters.

$$\hat{y}_{it} = \Phi_1 \hat{\epsilon}_{i,t-1} + \Phi_2 \hat{\epsilon}_{i,t-2} + a_{it} \quad \text{Eq. 5.5}$$

Where:

t ~ week

i ~ site

$\hat{\epsilon}_t$ ~ residual at time t, the lag between 2 observations was 1 unit of time (1 week)

$\Phi \geq 0$, and it was defined through a recursive difference equation (more in Pinheiro and Bates, 2000).

5.8.4.2 Prediction accuracy

The prediction error pe_{it} was calculated as the difference between the observed abundance of AF sea lice and the prediction divided by the standard deviation, squared, for the highest temperature category (Eq. 5.6).

$$pe_{it} = \left(\frac{obs.\overline{lnAF_{it}} - \hat{y}_{it}}{sd} \right)^2 \quad \text{Eq. 5.6}$$

Where: $\overline{\mathrm{lnAF}_{it}}$ was defined in Equation 5.1 as the weekly mean abundance of AF sea lice; in-sample observations (obs.) and predicted ($\hat{y}_{it}$ as calculated in Equation 5.5)

t ~ week

i ~ site

sd ~ error standard deviation for the high temperature category (11-12 °C)

5.8.5 Sensitivity analyses

5.8.5.1 Predictions and prediction errors

A summary of the predictions generated from the model is listed in Table 5.4.

The normalized prediction error for the predictions calculated using residuals set

are shown in Figure 5.7. These normalized prediction errors were not very different that the prediction errors calculated using the residuals that were the difference of the observed and predicted AF abundance.

Table 5.4. Median (standard deviation) of the observed and predicted adult female (AF) *Lepeophtheirus salmonis* sea lice and the normalized prediction error over 8 weeks in the late summer of 2020 for 14 site-cycles using two methods of prediction: 1) predictions calculated using residuals that were the difference between the observed and predicted, and 2) predictions calculated using residuals set to 0.

Predicted week	Observed AF	Method 1		Method 2	
		Predicted AF	Normalized prediction error	Predicted AF	Normalized prediction error
1	0.56 (1.36)	0.20 (1.21)	0.11 (0.39)	0.71 (0.88)	0.11 (0.79)
2	0.16 (1.18)	0.57 (1.08)	0.28 (1.47)	0.70 (0.92)	0.45 (1.43)
3	1.17 (1.03)	0.33 (0.91)	0.40 (0.83)	0.77 (0.89)	0.32 (0.82)
4	0.95 (0.91)	1.14 (1.00)	0.24 (0.60)	0.83 (0.98)	0.65 (0.54)
5	1.08 (1.41)	0.66 (0.81)	0.09 (2.81)	0.86 (0.85)	0.37 (2.15)
6	0.87 (1.09)	0.82 (1.04)	0.18 (0.84)	0.91 (0.78)	0.23 (0.61)
7	1.31 (0.77)	1.03 (0.88)	0.08 (0.66)	1.11 (0.76)	0.20 (0.86)
8	1.40 (0.88)	1.28 (0.66)	0.06 (0.54)	1.10 (0.77)	0.19 (1.33)

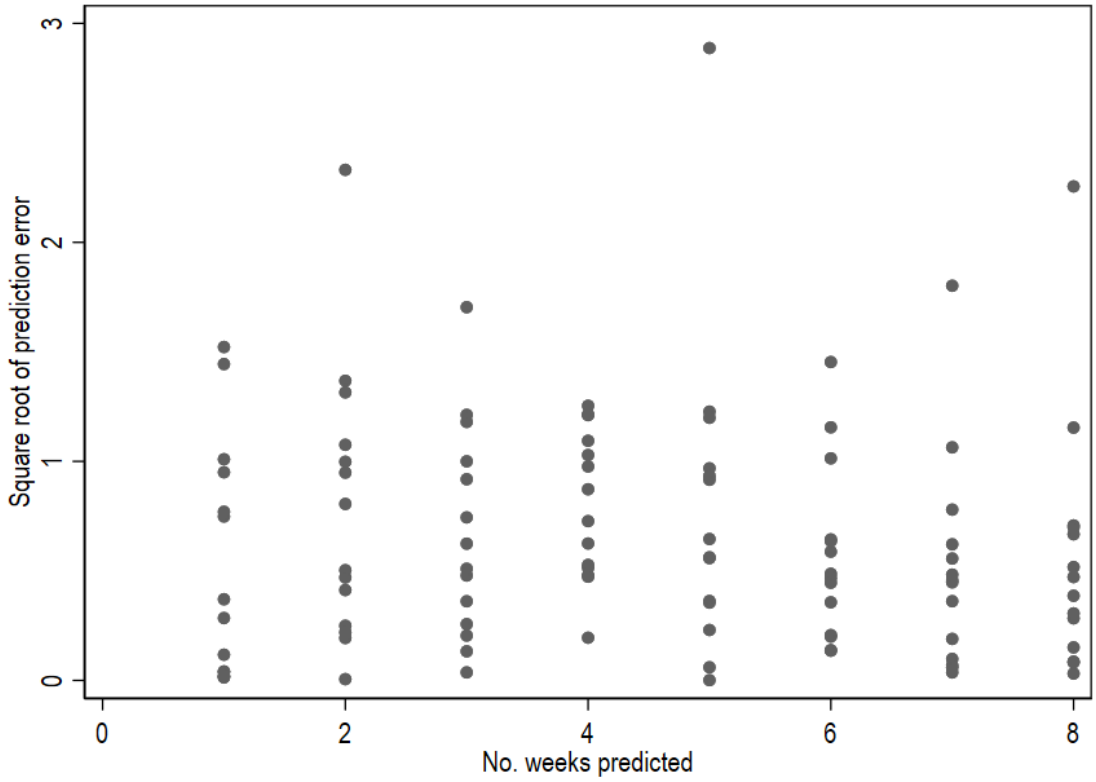

Figure 5.7. The square root of the prediction error for 14 sites (n = 112 observations) across the prediction horizon of 8 weeks. The prediction errors were calculated for in-sample short term predictions of the abundance of adult female *Lepeophtheirus salmonis* using a multivariable mixed autoregressive model and as calculated without the influence of the correlation parameters (residual set to zero).

5.8.5.2 Evaluation of the effect of potential outliers

Influential observations were identified using the standardized residuals in the evaluation of the model's fit. These observations were examined in detail at the cage and site-levels. The coefficients were evaluated for six observations (site-weeks) by looking at the percent change (Table 5.5). The percentage change was calculated as the difference of the coefficients of the final model with the model evaluated divided by the average of the two values.

Table 5.5. Estimated coefficient change (difference/average against the final model (n = 3,137 site-weeks) with a cutoff of |25 %|): - no change, ↑ increase, and ↓ decrease for the multivariable mixed autoregressive model for the abundance of adult female (AF) *Lepeophtheirus salmonis* sea lice. Temperature: sea surface temperature; IIP_{AF}: internal infestation pressure of AF; IIP_{PAAM}: internal infestation pressure of pre-adult and adult male; EIP_{AF}: external infestation pressure of AF; TAS: time at sea in weeks.

Variable and categories	Excl. obs. 1	Excl. obs. 2	Excl. obs. 3	Potential outlier 1	Potential outlier 2	Potential outlier 3
$n_{site\text{-}weeks}$	3,138	3,138	3,138	3,136	3,136	3,136
IIP_{AF}	↑	↑	↓	↑	-	↑
IIP_{PAAM}	-	-	-	-	-	-
EIP_{AF}	-	-	-	-	-	-
Temperature (°C)	-	-	-	-	-	-
$TAS^{1/2}$	-	-	-	-	-	-
Mechanical treatments						
$Week_{MO}$	Baseline					
$Week_{MB}$	-	-	-	-	-	-
$Week_{MTx}$	-	↓	-	-	-	-
$Week_{MA}$	-	-	-	-	-	-

Table 5.5 continued next page.

Table 5.5 continued.

Variable and categories	Excl. obs. 1	Excl. obs. 2	Excl. obs. 3	Potential outlier 1	Potential outlier 2	Potential outlier 3
Mechanical treatment-temperature interaction						
$Week_{MO}$*Temp	Baseline					
$Week_{MB}$*Temp	↓	↓	-	-	-	↓
$Week_{MTx}$*Temp	-	↓	-	-	-	-
$Week_{MA}$*Temp	-	-	-	-	-	-
Chemical treatments						
$Week_{CO}$	Baseline					
$Week_{CB}$	-	-	↓	-	-	-
$Week_{CTx}$	-	-	-	-	-	↑
$Week_{CA}$	↑	↑	-	-	↑	-
In-feed treatments						
$Week_{IO}$	Baseline					
$Week_{ITx}$	-	-	-	-	-	-
$Week_{IA}$	-	-	-	-	-	-

Table 5.5 continued next page.

Table 5.5 continued.

Variable and categories	Excl. obs. 1	Excl. obs. 2	Excl. obs. 3	Potential outlier 1	Potential outlier 2	Potential outlier 3
BMAs						
1	Baseline					
2A	-	-	-	-	-	-
2B	-	-	-	-	-	-
3A	-	-	-	-	-	-
3B	-	-	-	-	-	-
3C	-	-	-	-	-	-

The estimates for the predictors IIP_{PAAM}, EIP_{AF}, temperature, TAS, in-feed treatments and BMAs were robust to the reintroduction of three excluded observations and the removal of three potential outliers.

Three observations were excluded from the model as outliers after their evaluation. These observations had extremely low standardized residuals that resulted from multiple counts with 0-1 AF sea lice that occurred in the fall. Two observations were labelled as treatment weeks; excluded observation 2 had both chemical and mechanical (thermal and azamethiphos) treatments applied and excluded observation 3 had a mechanical (Hydrolicer®) treatment applied. Excluded observation 1 was labelled as a week before mechanical and chemical treatments (thermal and avermectin).

The decision to exclude these observations from the model was based on the deviation from the observed trends of the abundance of AF sea lice against the week for the duration of the production cycle for the site. The IIP_{AF} was sensitive to the reintroduction of these observations into the modelled dataset, and the magnitude of its effect increased. The reintroduction of the excluded observations reduced the effect of $Week_{MB}$*Temp. The reintroduction of the excluded observation 2 affected a decrease in $Week_{MTx}$ and $Week_{MTx}$*Temp. Although the excluded observations were not labelled as a week after chemical treatment, the effect increased for $Week_{CA}$.

One other potential outlier (no. 3) had low standardized residuals with a count of 0 AF sea lice that occurred in October. This site-week was labelled as $Week_{CB}$.

This observation was kept in the dataset as other weekly average abundances were low to 0 for the months of September and October at this site. The effect of the IIP_{AF} increased when this observation was removed from the dataset. Although labelled as $Week_{MO}$, the removal of potential outlier 3 affected a decrease in the effect of the $Week_{MB}$*Temp. The effect of the $Week_{CTx}$ increased with the removal of the potential outlier 3.

Two observations (outlier 1 and 2) had extremely high standardized residuals. One of these observations occurred when there were a large number of counts for that site-week. The other observation was labelled as a $Week_{MTx}$ (thermal) and had high abundance of AF sea lice for just two counts. The removal of the potential outlier 1, but not 2, affected an increase in the effect of the IIP_{AF}. The $Week_{CA}$ effect increased at the removal of the potential outlier 2.

In conclusion, the coefficient of the IIP_{AF} was very small and small changes to its value resulted in a large percent change. The effects noted on the mechanical and chemical treatment predictors suggest that caution should be applied during their interpretations.

5.8.5.3 Evaluation of the effect of adjusted timeseries

The effect of the adjusted timeseries ‘without winter gaps’ was evaluated using the percent change as was calculated for the evaluation of the potential outliers. The ‘without winter gaps’ time series effects were not summarized in a table as only the IIP_{AF} and $Week_{MB}$*Temp had a change greater than 25 %. Both these predictors increased with the ‘without winter gaps’ time series. The percent

change of the IIP_{AF} was large because the coefficient was small as previously discussed.

5.8.6 References

Bivand, R., Keitt, T., Rowlingson, B., Pebesma, E., Sumner, M., Hijmans, R., … Rundel, C. (2019). rgdal: Bindings for the 'Geospatial' Data Abstraction Library. R package version 1.4-8. https://CRAN.R-project.org/package=rgdal

Department of Energy and Resource Development/Agriculture, Aquaculture, and Fisheries. (2017). *Bay of Fundy Marine Aquaculture Sites* [map]. 1:125000. New Brunswick, Canada.

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31, 100394. https://doi.org/10.1016/j.epidem.2020.100394

GADM. (2018). World/Canada/New Brunswick. Retrieved from https://gadm.org/maps/CAN/newbrunswick.html

Hijmans, R.J., van Etten, J., Sumner, M., Cheng, J., Baston, D., Bevan, A., … Wueest, R. (2020). raster: Geographic Data Analysis and Modeling. R package version 3.0-12. https://CRAN.R-project.org/package=raster

Pinheiro, J. C., Bates, D. M. (2000). Mixed-Effects Models in S and S-PLUS. In J. Chambers, W. Eddy, W. Hardle, S. Sheather, L. Tierney (Eds.), Statistics and Computing (1st ed.). Springer-Verlag New York, Inc. 528p.

QGIS.org (2018). QGIS Geographic Information System. Open Source Geospatial Foundation Project. http://qgis.org.

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

Thakur, K. K., Vanderstichel, R., Barrell, J., Stryhn, H., Patanasatienkul, T., and Revie, C. W. (2018). Comparison of remotely-sensed sea surface temperature and salinity products with in situ measurements from British Columbia, Canada. Frontiers in Marine Science, 5, 1-11. https://doi.org/10.3389/fmars.2018.00121

[dataset] UK Met Office (2005). OSTIA Level 4 Sea Surface Temperature Analysis. Ver. 1.0. PO.DAAC, CA, USA. Retrieved from https://doi.org/10.5067/GHOST-4FK01.

van Etten, J., and de Sousa, K. (2019). gdistance: Distances and Routes on Geographical Grids. R package version 1.1-4. https://CRAN.R-project.org/package=gdistance

6 Discussion

6.1 Summary

This thesis evaluated the connectivity of *L. salmonis* sea lice among Atlantic salmon aquaculture sites in the Bay of Fundy, NB, by estimating infestation pressures and dispersal of farm-sourced sea lice. Information on sea lice infestation in farmed salmon is important to the producers in terms of money and time (e.g., Abolofia et al., 2017) and the planktonic life stages of *L. salmonis* (nauplii I and II, and young copepodid) exist in the water column in a manner described as a “black box” (Morefield and Hamlin, 2022). The exact movements of the planktonic life stages are unknown but crucial to the infestation of the sea lice within and among sites (internal [IIP] and external infestation pressure [EIP], respectively), which has important ties to decisions regarding mitigation strategies for the sea lice. The marine environment adds complexity to the study of sea lice infestation and dispersal as it strongly impacts the movement of the planktonic life stages (e.g., Amundrud and Murray, 2009) and rate of development of sea lice (Hamre et al., 2019).

In Chapter 2, a descriptive analysis was performed to describe the industry and sea lice data collected thus far in the monitoring program and evaluate the variables that may be confounding factors in models produced in subsequent chapters. A short historic review of the Atlantic salmon aquaculture industry in the Bay of Fundy, NB, was provided in tandem with a brief review of the critical environmental variables of water temperature and salinity.

Two multivariable models were developed to estimate the infestation pressures and dispersal of the sea lice, considering the complex life stages of the sea lice and their complex environment. The first was a multivariable linear mixed model constructed to estimate the average IIP and EIP of sea lice in the Bay of Fundy, NB (Chapter 3: using “segments”). The dispersal seaway distance of sea lice was estimated by selecting the lowest AIC while evaluating the model and adjusting the EIP with a range of KDE kernel bandwidths.

We proposed that there may be connectivity measures other than seaway distance that could inform the connectivity and dispersal of sea lice in the Bay of Fundy, NB. Four connectivity measures were evaluated using the previously published model from Chapter 3 (Chapter 4). The dispersal distance was also determined for these different connectivity measures.

A second model constructed was a multivariable autoregressive linear mixed effects model (AR[2]) to estimate the abundance of AF sea lice in the Bay of Fundy, NB, and it was used to illustrate short-term predictions (Chapter 5: using “time series”). This model’s estimates of infestation pressure were also evaluated and compared to the results of the previous model in Chapter 3.

The current chapter further compares the models from Chapters 3 and 5, as these were constructed using different methods and datasets. Following, infestation pressures, connectivity and dispersal distance, treatments and other major points are discussed with contrast across the chapters and insight from the

results of the comparison of the models. Discussion of future directions for research are embedded within each of the subsections (6.3 and later).

6.2 Comparison of models

6.2.1 Objective

The models presented in Chapters 3 and 5 produced different estimates. These different results are likely due to the different modelling approaches and datasets. These different models (Chapters 3 and 5) provided a unique opportunity to compare different methods and datasets within the same study area when they overlap. In Chapter 3, the data were organized into segments where a topical treatment occurred and followed by all the observations (counts of sea lice) until the next topical treatment for a cage at a site within a production cycle. Comparatively, Chapter 5 did not have this segment organization and the counts were averaged weekly at the site-level. These models were constructed using different datasets from the Fish-iTrends© database; Chapter 3 included 2009 to 2018, and Chapter 5 was based on 2016 to 2021. The comparison across numerous studies has demonstrated variable estimates of infestation pressures, dispersal distance and other predictors (e.g., Aldrin et al., 2019). Moreover, these studies vary by the environment, an essential factor in estimating connectivity, dispersal, and infestation.

This comparison of the models from Chapters 3 and 5 for the period 2016 to 2018 will shed light on the influence of the methods and datasets on the results of infestation pressures. The comparison will allow for the assessment of the internal validity of the methods and data. Chapters 3 and 5 models will be applied

to an overlapping subset of the datasets to achieve these objectives. Additionally, simplified Chapters 3 and 5 models will be applied to the subset of the datasets to examine the effects of temperature and infestation pressures on their own.

6.2.2 Materials and methods

The datasets for Chapters 2-4 (July 2009 to November 2018) and Chapter 5 (June 2016 to June 2021) were restricted to overlapping time. The site-production cycles included in the analysis were active sites for the overlapping time modelled in Chapters 3-5. This produced two datasets: 1 (Chapters 2-4) and 2a (Chapter 5). Dataset 2a was further restricted to average weekly abundances after the first topical treatment events (dataset 2b). Dataset 2b was created to compare more closely the observations from later in the production cycle as the inclusion of observations early in the production cycle in Chapter 5 may potentially affect the estimation of IIP and EIP compared to Chapter 3, which lost the early observations of the production cycle due to the organization into segments where topical treatments typically begin later in the production cycle.

The multilevel mixed effects linear regression with an exponential correlation structure to the errors, stratified by three temperature profiles (low = 3-7 °C, medium = 8-11 °C, and high = 12-16 °C) constructed in Chapter 3 was applied to dataset 1 (model 1). The multivariable autoregressive linear mixed effects model with (AR[2]) process correlation structure to the errors, stratified by three temperature profiles for the site-cycle hierarchical level (low = 6-9 °C, medium = 10 °C, and high = 11-12 °C) constructed in Chapter 5 was applied to datasets 2a and 2b (models 2 and 3). These models were also run with only the infestation

pressures (IIP_{AF}, IIP_{PAAM} and EIP_{AF}) and temperature to evaluate these effects with datasets 1 and 2a (models 4 and 5). All models were fit with restricted maximum likelihood estimation. Model assumptions were not re-examined.

Analyses were conducted in Stata® v17, and statistical significance was set at $p < 0.050$.

6.2.3 Results

The datasets overlapped from June 6, 2016, to November 8, 2018. The number of observations and site-weeks for each of the subsets of data were summarized in Table 6.1. The restriction of dataset 2a to observations after the early production cycle (dataset 2b) resulted in a loss of 328 site-weeks. Dataset 2b had a smaller mean number of site-weeks per site-cycle, 23.3 (range of 4 to 52 site-weeks), compared to dataset 2a, with a mean of 36.4.

Table 6.1. Number of observations (counts and weeks) per hierarchical level for datasets from Chapters 2-4 (1) and Chapter 5 (2a and b) for June 2016-November 2018. Dataset 2b was restricted to observations (weeks) after the first topical treatments were applied during the production cycle. n/a not applicable, † = nested groups in multilevel mixed effects linear regression for the abundance of adult female (AF) *Lepeophtheirus salmonis*; ‡ = nested groups in the multivariable autoregressive linear mixed effects model for the abundance of AF *L. salmonis*.

	Dataset		
	1	**2a**	**2b**
Counts †	2,068	5,370	2,651
Weeks ‡	473	911	583
Grouping variable			
BMA	6	6	6
Site	25	25	25
Site-production cycle †‡	25	25	25
Cage	34	n/a	n/a
Cage-production cycle †	220	n/a	n/a
Segment †	484	n/a	n/a

The restriction of the data to an overlapping time frame resulted in estimated coefficients for the IIP_{AF} and IIP_{PAAM} that were smaller in the time series model (2) compared to the segment model (1; Table 6.2). Including the observations early in the production cycle (model 2, dataset 2a) resulted in smaller IIP estimated coefficients compared to the estimated coefficients from model 3 without those observations (model 3, dataset 2b). The IIP_{AF} remained non-significant in model 2 ($p = 0.557$) as in the final model produced in Chapter 5. In the models restricted to the predictors of the infestation pressures and the temperature, the estimated coefficient of IIP_{AF} was much larger in model 4 and it became significant ($p < 0.001$).

The EIP_{AF} had an effect almost 2.5 times larger in the segment model (1; $p < 0.001$) compared to the time series model (2; $p > 0.083$). The restriction to observations later in the production cycle resulted in a slight increase in the

effects of the EIP_{AF}. In model 3, the EIP_{AF} increased slightly compared to model 2 but remained smaller than in model 1. The estimated coefficient of the EIP_{AF} was greater in the models restricted to the predictors of the infestation pressures and temperature (model 4: $p < 0.001$ and 5: $p = 0.005$) compared to models 1 and 2.

Table 6.2. Estimate coefficients (coef.) and standard errors (SE) for multiple models (1 to 5) that utilized datasets (ds) from 2016 to 2018. The final multivariable linear mixed effects regression model (model type “segments” [seg]) and the final multivariable autoregressive linear mixed effects model (model type “time series” [ts]) were applied (models 1 to 3) and limited to the following predictors: internal infestation pressure (IIP) of adult female (AF) *Lepeophtheirus salmonis* (IIP_{AF}), IIP of pre-adult and adult male (IIP_{PAAM}), external infestation pressure of AF (EIP_{AF}), and sea surface temperature centred at 11.85 °C (models 4 and 5). base = baseline, ─ predictor not in the model.

Variable and categories	Model 1: seg, ds 1		Model 2: ts, ds 2a		Model 3: ts, ds 2b		Model 4: seg, ds 1		Model 5: ts, ds 2a	
	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE
IIP_{AF}	0.091	0.037	0.021	0.035	0.129	0.045	0.271	0.034	0.030	0.035
IIP_{PAAM}	0.089	0.034	0.074	0.029	0.123	0.035	0.072	0.035	0.069	0.030
EIP_{AF}	0.834	0.119	0.338	0.195	0.380	0.229	1.173	0.119	0.565	0.202
$Baseline_{AF}$	0.393	0.047	─		─		─		─	
Temperature, centred	-0.075	0.021	-0.048	0.056	0.028	0.068	-0.002	0.016	-0.009	0.027
Time at $sea^{0.5}$	─		0.430	0.042	0.275	0.046	─		─	
Treatment										
Azamethiphos Tarp	base.		─		─		─		─	
Aza. Wellboat	-0.715	0.164	─		─		─		─	

Table 6.2 continued next page.

Table 6.2 continued.

Variable and categories	Model 1: seg, ds 1		Model 2: ts, ds 2a		Model 3: ts, ds 2b		Model 4: seg, ds 1		Model 5: ts, ds 2a	
	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE
Treatment										
Thermal	-0.595	0.122	–		–		–		–	
Hydrolicer®	0.428	0.343	–		–		–		–	
H_2O_2	-0.141	0.228	–		–		–		–	
Treat-temp interaction										
Aza.Tarp*Temp	base.		–		–		–		–	
Aza.Wellboat*Temp	0.078	0.057	–		–		–		–	
Thermal*Temp	0.177	0.029	–		–		–		–	
Hydrolicer®*Temp	-0.014	0.178	–		–		–		–	
H_2O_2*Temp	-0.030	0.094	–		–		–		–	
Mechanical treatment										
$Week_{MB}$	–		base.		base.					
$Week_{MTx}$	–		-0.154	0.119	-0.280	0.151	–		–	

Table 6.2 continued on the next page.

Table 6.2 continued.

Variable and categories	Model 1: seg, ds 1		Model 2: ts, ds 2a		Model 3: ts, ds 2b		Model 4: seg, ds 1		Model 5: ts, ds 2a	
	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE
Mechanical treatment										
$Week_{MA}$	—		-0.410	0.142	-0.543	0.174	—		—	
$Week_{MO}$	—		-0.328	0.126	-0.412	0.158	—		—	
Mech-temp. interaction										
$Week_{MB}$*Temp	—		base.		base.					
$Week_{MTx}$*Temp	—		0.047	0.053	0.012	0.071	—		—	
$Week_{MA}$*Temp	—		-0.006	0.067	-0.068	0.082	—		—	
$Week_{MO}$*Temp	—		0.016	0.055	-0.069	0.068	—		—	
Chemical treatment										
$Week_{CB}$	—		base.		base.					
$Week_{CTx}$	—		-0.011	0.121	-0.074	0.153	—		—	
$Week_{CA}$	—		-0.095	0.140	-0.086	0.178	—		—	
$Week_{CO}$	—		-0.101	0.119	-0.112	0.150	—		—	

Table 6.2 continued next page.

Table 6.2 continued.

Variable and categories	Model 1: seg, ds 1		Model 2: ts, ds 2a		Model 3: ts, ds 2b		Model 4: seg, ds 1		Model 5: ts, ds 2a	
	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE	Coef.	SE
In-feed tx										
$Week_{ITx}$	–		base.		base.					
$Week_{IA}$	–		-0.024	0.072	0.005	0.101	–		–	
$Week_{IO}$	–		-0.121	0.133	-0.055	0.183	–		–	
BMA										
1	base.		base.		base.					
2A	0.085	0.325	0.475	0.540	0.617	0.523	–		–	
2B	0.241	0.385	0.313	0.578	0.280	0.562	–		–	
3A	0.433	0.326	0.477	0.535	0.669	0.517	–		–	
3B	0.510	0.318	0.650	0.579	0.733	0.516	–		–	
3C	-0.303	0.447	0.209	0.580	0.430	0.558	–		–	
Constant	-0.004	0.301	-1.848	0.541	-1.015	0.523	0.414	0.110	0.633	0.154

The predictor of treatment and the interaction term of treatment with temperature in model 1 were significant (Wald test, $p < 0.001$). The topical treatments (mechanical and chemical) and the interaction term of mechanical treatment with temperature were no longer significant in model 2 as compared to the final model in Chapter 5 (Wald test, $p > 0.050$). The loss of the early production cycle in the dataset affected a change in the strength of the estimated coefficients for the treatment terms, but these were not significant. The other treatment predictors were also non-significant in models 2 and 3 (Wald test, $p > 0.050$).

Temperature had a negative estimated coefficient in models 1 and 2 and near zero in models 4 and 5. The negative and small coefficient for temperature was a surprising finding as this is an essential variable for the sea lice development rate. The temperature was positive for model 3 with the dataset that excluded the early observations (2b).

6.3 Internal and external infestation pressures

In comparing the models (section 6.2), five models were produced from two datasets for 2016-2018 to assess the internal validity of the models and datasets presented in this thesis. The segment and time series models were similar in that they both had a positive effect on the abundance of AF sea lice, but the strength of the effect varied. The comparison of the models 4 and 5, with the predictors limited to the infestation pressures and temperature, illustrated that the effects of the infestation pressures were stronger for the “segment” type models. However,

if a decision had to be made between the “segment” and “time series” model types, it should be based on the questions posed and the ease of data management. For these reasons, the time series model may be a better selection than the segment model as it can be applied with fewer data management steps and can perform predictions that may be useful for the industry.

It was determined that infestation pressures play an important role in the prediction of the abundance of AF sea lice. The coefficient of the IIP_{AF} was null in the final multivariable autoregressive linear mixed effects model from Chapter 5, but the IIP_{AF} was significant with the adjustment of that model’s residual error and the previously observed significant IIP_{AF} in Chapters 3 and 4. The IIP was statistically significant in multiple studies (e.g., Kristoffersen et al., 2013) and was not significant in other studies (e.g., Elghafghuf et al., 2020). The IIP_{PAAM} was significant in the final multivariable autoregressive linear mixed effects model from Chapter 5 and its effect was not altered with the changes applied to the model’s residual error structure. The IIP_{AF} and IIP_{PAAM} are important predictors for the abundance of AF sea lice in the Bay of Fundy, NB. Important IIP conveys that managers and farmers should mitigate sea lice at their farms to reduce the burden of sea lice in the future. Mitigation practices to reduce IIP include methods to reduce the reproductive output of AF sea lice at the farm site, such as those that reduce adult sea lice (e.g., cleaner fish that remove gravid females [PMRA, 2003]). Flushing the site, as would be expected in some geographical locations where tidal currents prevent re-infestations, would reduce IIP but may

increase the EIP to the neighbours. It also means that prediction models should consider the IIP for the site even if the predictor is non-significant.

The IIP estimates were calculated in Chapter 3 as the temporally weighted averages for 3 and 4 weeks past for AF and PAAM, respectively. In Chapter 5, the IIP was not temporally weighted; the IIP were 3 and 4-week lagged weekly averages of AF and PAAM, respectively. A benefit to the non-weekly average for the IIP was that there were fewer losses from missing observations, demonstrated in the small sensitivity analysis for Chapter 3, where a temporal triangular average reduced the zero counts. In Chapter 5, observations were lost when there were no observations at the lagged time points for IIP_{AF} and IIP_{PAAM}. The potential loss of observations where there were no available observations of AF and PAAM is a disadvantage to the methods of weekly average. However, a substantial benefit is the ease of the weekly average calculation. The effect of this loss of observations in the autoregressive model was partly evaluated using the ‘winter gaps’ time series, as during the spring, there would not be available lagged values for the AF and PAAM. The model runs with the ‘winter gap’ time series had a larger estimated coefficient of the IIP_{AF} compared to the AR(2) model, but the coefficient's size remained small. We conclude that the loss of observations from the missing values at the lagged time points in Chapter 5 had a minimal effect on the IIP estimated.

The EIP presented in Chapters 3, 4, and 5 were significant predictors in the models. The estimates of EIP informed us that there are potential movements of the sea lice among sites, and we can conclude that neighbouring sites contributed to the infestation of sites for 2009-2021. In addition to reducing IIP at the farm site, reducing the EIP should be a goal for farmers and managers. One way to achieve this is by applying a different organizational structure to the site locations within the Bay of Fundy, NB. An example of a different organization structure would be the creation of clusters of sites where synchronous treatment applications may be applied, and intermediary sites that connect two clusters are removed. This is the application of the concept of 'firebreaks' (Samsing et al., 2019) that reduces the connectivity among clusters of sites and the overall EIP for the area. This would mean changing the current organization of the BMAs and the sites within the BMAs so that groups of sites are closely connected, and small sites intermediary to the clusters are closed or removed ('firebreak') to decrease the dispersal of sea lice. A network analysis combined with a hydrodynamic and biological model was used to identify the 'firebreaks' in Norway (Samsing et al., 2019). A similar application of 'firebreaks' in NB may be more difficult in NB due to differences in the geography of the area (i.e., NB does not have fjords as in Norway).

The EIP demonstrated interesting patterns when plotted against the time at sea (TAS; Figure 6.1), a variable calculated from the stocking date of the salmon for each site and production cycle (Chapter 5). The IIP vs. TAS had similar trends to

the EIP (not shown). The stocking of salmon at sites is regulated to be coordinated according to the BMA that the site belongs to (NB Department of Energy and Resource Development/Agriculture, 2017). The locally weighted scatter plot smoothing (LOWESS) curve demonstrated a trend for an increase in EIP as the cohort of salmon developed until approximately 90 weeks at sea, when there was a decrease in the EIP (Figure 6.1A). The build-up of EIP was steepest from the stocking date to approximately 20 weeks at sea when the rate decreased slightly until about 90 weeks at sea. The steepest increase of the EIP was for the first year at sea, after which there was a decrease and then another increase in the EIP in the second year but with greater variability. The decrease in pressure after 90 weeks at sea was partially due to the decrease in the number of observations, likely due to the start of the harvest of the salmon, as salmon are grown for approximately 18 months, and 90 weeks is equivalent to 20 months. In other words, this decrease in the EIP is likely due to removing the potential hosts for the infestation by sea lice. The LOWESS curve showed a similar trend of increasing EIP from the stocking date for each of the BMA until a particular time that the pressure decreased (Figure 6.1B). One BMA had a different trend, such that the EIP curiously followed a sinusoidal-like curve. The classification of the EIP by BMA more clearly illustrated that the EIP had two major peaks during the production cycle. Most stocking occurred between April and June for 2016-2021 (74.0 % or 57/77 site-cycles), so the troughs in the EIP occur at approximately the winter, and the peaks occur at approximately the first

and second summer at sea. Further exploration of the trends of the EIP against the time at sea may provide more insight into the management of sea lice according to the number of weeks in the production cycle and can be a topic for future studies.

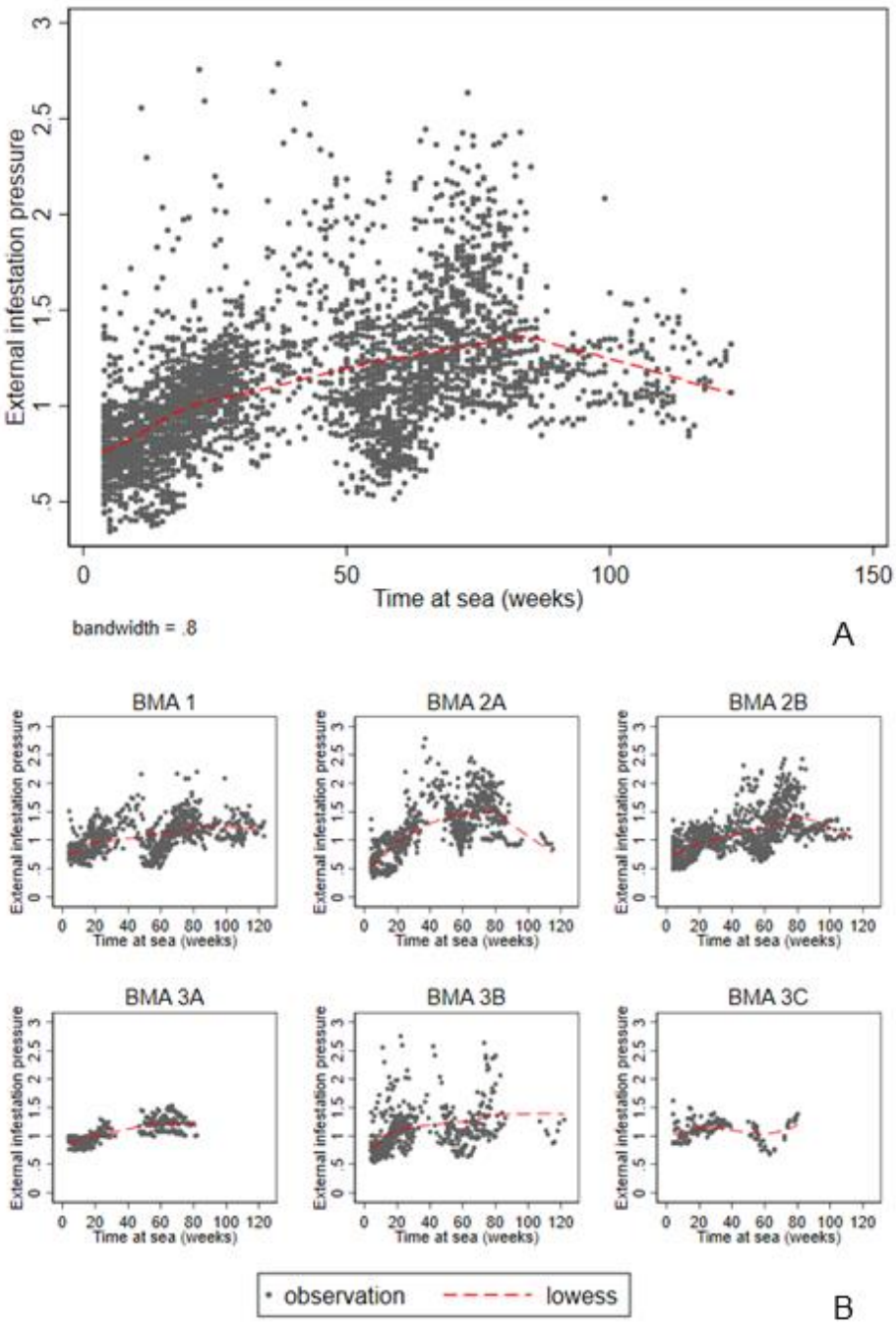

Figure 6.1. The external infestation pressure of *Lepeophtheirus salmonis* sea lice (non-log scale) against the time at sea in weeks for Atlantic salmon at aquaculture sites in the Bay of Fundy, New Brunswick. The observations were modelled in the multivariable mixed effects time series model in Chapter 5. Locally weighted scatter plot smoothing curve overlay the observations (red dash). Aggregate plot (A) and divided per the Bay Management Areas (BMA) (B).

The strength of the effect of the EIP was smaller than the IIP in the models in Chapters 3 and 5; these models utilized large ranges of time (2009-2018 and 2016-2021, respectively). The larger IIP than the EIP effects can be translated to a need to focus on management at the site to reduce sea lice burden (e.g., chemotherapeutant applications) and, as a secondary effort, apply management practices that reduce sea lice incoming from neighbouring farm sites (e.g., that is available but was not applied to the study area are sea lice netting). However, other studies of infestation pressures for other areas have demonstrated that the EIP had a larger estimated effect than the IIP (e.g., Grand Manan: Elghafghuf et al., 2020). The estimated coefficient of the EIP was greater than the IIP_{AF} and IIP_{PAAM} for the models produced to compare the methods and datasets (Section 6.2). The IIP being larger than the EIP may be dependent on the period of data modelled. Future studies should examine the relationship between IIP and EIP.

6.4 Connectivity and dispersal distance

6.4.1 Connectivity measures

The EIP was calculated using spatially and temporally weighted empirical data. The spatial weights were calculated as the KDE adjustment of the connectivity measure among pairs of sites. Recall that the connectivity measure is a measure to estimate the dispersal distance among sites, and the dispersal distance is one variable inherent to the movements among sites that are facilitated or impeded by the environment.

The most commonly used metric is seaway distance, the shortest distance between two sites by waterway (e.g., Elghafghuf et al., 2021). Seaway distance at a resolution of 100 m was used in Chapter 3, and it was determined that a finer resolution (up to 25 m) was not necessary for this model (Chapter 4). Where seaway distance is the most common and 100 m resolution sufficient, this connectivity measure was used for the model in Chapter 5, in which predictions were made.

The proposed connectivity measure of the percent surface area along a seaway path performed slightly better (i.e., slightly lower AIC) than the seaway distance (Chapter 4). This may be because water closer to land behaves differently than water further from land due to aspects such as depth and turnover due to tides. Describing the reasoning behind this connectivity measure will require additional research as it may demonstrate confounding effects. In the chapter, we proposed that the percent surface area along a seaway path may be a surrogate for coastal effects on the dispersion of sea lice in the Bay of Fundy, NB. A hydrodynamic model (e.g., FVCOM) would likely inform the influence of the coast on the dispersion of sea lice. The benefits of the FVCOM are the hourly movements of the water currents that respond to temperature, salinity, tides, and weather patterns, and it can produce bidirectional connectivity measures. A FVCOM was being developed during the thesis project but was not ready to be employed within a reasonable time frame. Future work will incorporate FVCOM

output for the year 2017 and attempt to summarize the output as a connectivity measure for comparison within the model of Chapter 4.

The other connectivity measures examined, topographical distance and volume of water along a seaway path, did not improve the model fit compared to seaway distance. Both connectivity measures included a measure of depth in their calculation. This and the non-significant predictor of depth at sites could be interpreted as bathymetry (depth) having negligible effect on the sea lice dispersal. Depth may influence sea lice since there are sea lice present throughout the water column as they react to the light (Penston et al., 2004) and other variables (Frenzl, 2014). However, where these connectivity measures did not improve the model fit (i.e., seaway distance explained the variation in the abundance of AF lice better), these connectivity measures may still impact sea lice. Further exploration of these connectivity measures may help explain some of the variation that is yet unexplained in models using seaway distance as a connectivity measure.

The heretofore description of the connectivity measures was indirect as they were part of the calculation for the derived predictor EIP. The KDE and EIP calculations had a robust smoothing effect on the variation observed among the 'distances' for the connectivity measures among sites. Despite this, we could identify differences in the effect of EIP in the model.

The four connectivity measures and their adjustments were also directly compared in Chapter 4 using Euclidean distance. The major finding was that some of these connectivity measures were very different from the seaway distance (e.g., topographical distance with an adjustment of the cost of movement along an incline by a factor of 1,000), and others were very similar (e.g., topographical distance with a factor of 1). This comparison of the connectivity measures did raise additional questions, such as the noted divergence of the observations in a selection of scatter plots (e.g., seaway distance against topographical distance with an adjustment of the cost of movement along an incline by a factor of 10). Future research into connectivity measures is necessary to explain the divergence of the observations. It is unknown at this time.

6.4.2 Dispersal distance

The dispersal distance was estimated during a sensitivity analysis with varying bandwidths of the KDE to determine the distance that best fit the observed data. The bandwidth of the KDE was used to adjust the seaway distance to produce the spatial weight necessary for calculating the EIP. This dispersal distance is, therefore, strongly dependent on the form of the function of the KDE; a Gaussian distribution was used in this model as was utilized in previous studies (e.g., Elghafghuf et al., 2020). A greater distance from the site received fewer infective copepodids that could infest a neighbouring site as demonstrated in the example in Chapter 5. The Gaussian distribution is isotropic (i.e., the spread of planktonic

sea lice life stages is even in all directions from the site; however, the EIP is anisotropic because the spread is impacted by land where the KDE with Gaussian curve is applied to seaway distances. An example of increasing distances that lead to decreased EIP was shown in Figure 5.4. The actual distribution of planktonic life stages may be different and more complex because the dispersal kernel is difficult to estimate (e.g., Aldrin et al., 2013). An entire study could be constructed around estimating the appropriate dispersal kernel (i.e., distribution) for the KDE. The similar results of the dispersal distance estimated in the models presented in this thesis and the distances published in other studies (e.g., Elghafghuf et al., 2020) support a conclusion of this dispersal distance as it was repeatable using different methods and assumptions (e.g., a distribution that decayed with temperature [Aldrin et al., 2019]).

The dispersal distance of planktonic sea lice is an estimate that can be used for the management of sea lice and during applications of the concept of ‘firebreaks’ (Samsing et al., 2019) The dispersal distance for Atlantic salmon farm sites in the Bay of Fundy, NB, was estimated at a seaway distance of 10 km (Chapter 3) and the percent surface area along a seaway path with an area of 1,000 m of 7 km (Chapter 4). Elghafghuf et al. (2020) estimated a slightly larger dispersal seaway distance of 12.5 km for Grand Manan. The dispersal distance may vary according to specific areas within the study area; the 7-10 km range represents an average for the entire area. Other areas of the world have estimated more considerable dispersal distances between 20 and 30 km (e.g., Aldrin et al.,

2019), which may be due to differences in the environment and methodologies employed.

6.5 Treatments

Treatments were included in the constructed models (Chapters 3, 4 and 5). The studies and models were not designed to evaluate the efficacy of treatments; therefore, caution should be used when interpreting the treatment estimates and trends were noted. Instead, the inclusion of treatments was used as a controlling factor when estimating infestation pressures. Despite this, the modelling results demonstrated an interaction between treatments and temperature. In Chapter 3, the effect of hydrogen peroxide and thermal treatments decreased with increasing temperature. The effect of azamethiphos increased with increasing temperature. In Chapter 5, the effect of thermal and Hydrolicer® treatments (combined into a mechanical category of treatments) decreased with increasing temperature. These temperature-treatment effects could be a leftover from seasonal trends, which should be examined further. A real interaction between temperature and treatment is an avenue for further research where treatment efficacy considers temperature. The research results may guide clinical decision-making such that a veterinarian selects the appropriate treatments according to the seasonal adjustments.

The predictor $baseline_{AF}$, which was the observation of AF abundance prior to a segment, was significant in the final multivariable linear mixed effects regression

model in Chapter 3. There was no equivalent for $baseline_{AF}$ in the final multivariable autoregressive linear mixed effects model. The post treatment weeks ($Week_{MA}$, $Week_{CA}$ and $Week_{IA}$) were compared to the week before treatment ($Week_{MB}$, $Week_{CB}$ and $Week_{ITx}$) showed that the abundance of AF was higher for the before weeks than after. These results support that treatments are reducing the abundance of AF as the abundance is higher for the week prior to treatment and lower for the week after treatment. The effectiveness of treatments was not an objective for these studies, and the combination of 'pre-treatment' and 'post-treatment' counts in Chapter 5 limit our ability to comment fully on the effectiveness of sea lice treatments in the Bay of Fundy, NB.

Although not relevant to this project, a change was made to the application method of azamethiphos for treatment as of 2021 that may affect future evaluations of treatment effects and efficacy. Bath treatments with azamethiphos were extended in time due to changes in application methods than previously applied (L. Hammell, pers. comm.).

Other treatments were applied to the sites not accounted for in the analysis because these were not included in the Fish-iTrends© database. One of the possible treatments that can affect sea lice abundance early in the production cycle is the application of lufenuron (Imvixa™, Elanco™ Animal Health Inc., USA). Mowi Canada East applied lufenuron to salmon prior to the transfer of smolt from the hatcheries to sea in 2018 and 2019 (A. Borchardt, pers. comm.).

Kelly Cove Salmon Ltd. (a division of Cooke Aquaculture) applied lufenuron at the hatcheries for pilot studies (2013-2015 and 2019) and for treatment in 2017-2019 (L. Hawkins, pers. comm.). The hatcheries that had lufenuron applied supplied smolt to complete BMAs in some instances: 2018 BMA 1, 2019 BMA 2A (A. Borchardt and L. Hawkins, pers. comm.), and 2017 ¾ of BMA 2B (L. Hawkins, pers. comm.). The Veterinary Drugs Directorate of Health Canada denied the application of lufenuron at the hatcheries for stocking fall smolt of 2019 (L. Hawkins, pers. comm.). This information affects all models as the application range was included in Chapters 3-4 (2009-2018) and Chapter 5 (2016-2021). The application of lufenuron on select cages in BMAs, such as for pilot studies, may not significantly impact the model. However, where entire BMAs were treated in 2017-2019, this would reduce sea lice abundance from stocking to 6.6-9 months into the production cycle (Junquera et al., 2019). To note, there is a paucity of published literature on the efficacy of lufenuron, and there is also uncertainty regarding resistance to the compound. The application of lufenuron was, therefore, a potential confounder. Lufenuron treatment may also influence other model predictors, such as the time at sea and seasonality effects. The implications were that the ability to model the abundance of sea lice was poorer for specific spatiotemporal locations, such as early after stocking; however, in the infestation pressures vs. TAS (e.g., EIP: Figure 6.1), there was an increasing trend soon after stocking and not the expected 6-month wait until the increase of the abundance of sea lice. The unaccounted application of

lufenuron had the potential for misclassification that would result in the infestation pressure estimates to err toward the null hypothesis. To fully understand the effects of lufenuron in the data, a few approaches can be considered: 1) adding a categorical variable for the dates and sites where lufenuron was putatively applied, and 2) obtaining the treatment data from Elanco™. The latter was investigated, and we met with Elanco™ team members to assess the feasibility of data sharing and linking to the Fish-iTrends© data. We determined that where the administration of the lufenuron occurs at the hatcheries, and the hatcheries supply multiple sites in separate locations, and this makes the data much more complex.

Similar to the application of lufenuron, our models did not assess sea lice treatment with cleaner fish. Cleaner fish were used in 2019 BMA 1, 2020-2021 BMA 3 (unknown if section A or B [A. Borchardt, pers. comm.]), and a few sites in 2020 BMA 2B and 2021 BMA 1 (L. Hawkins, pers. comm.). During pilot studies, cleaner fish were also used before 2019 in Grand Manan (L. Hawkins, pers. comm.). These treatments with cleaner fish would reduce the measured abundance of sea lice (Imsland and Reynolds, 2022). However, since the application was sparse and the effects on the models were likely minimal, the potential bias would lead the estimate towards the null hypothesis.

6.6 Temperature and salinity

Temperature is an important parameter for the development of sea lice (Hamre et al., 2019). The rate of sea lice development increases with increasing temperature (Hamre et al., 2019). In all of the models in Chapters 3, 4 and 5, this predictor was significant. The results presented were small and negative estimated coefficients, the opposite of the expectation of a positive association between temperature and sea lice abundance. This negative association between the abundance of AF *L. salmonis* and temperature may be due to the complex nature of the relationship of the sea lice with temperature in light of the treatments. Another factor driving the association between temperature and the abundance of AF sea lice may be the early production cycle, as the estimated coefficient for temperature of model 3 was positive. Where most stocking occurs in the spring in the study area, the early production cycle would include lower temperatures; therefore, excluding the early production cycle, observations would increase the mean temperature of the observations. This increased mean temperature may be associated with improved reproductive success and increased development rates, as demonstrated during matrix population modelling by Groner et al. (2014).

The effects of temperature on the development and survival of sea lice are affected by salinity (Groner et al., 2016). The constructed models did not include salinity measures because the *in-situ* site salinity records are poor. Including salinity would improve our knowledge of the effects of salinity on the abundance

of sea lice. One way to include salinity in models is using remote-sensing salinity measurements. The sea surface salinity SMOS-OI product (EU Copernicus Marine Service Information, 2021) has measurements that may be used but could not be utilized within this thesis as they were published in 2021. A challenge to using this remote-sensing product is the spatial resolution of approximately 45 km, which means the study area has poor coastal resolution. Implementing a salinity measure in the model may be helpful; however, the effects of salinity may be small. The freshet and other freshwater input into the Passamaquoddy Bay that may affect the abundance of sea lice in the area occur mainly during the spring, and there are few counts performed during the same period.

6.7 Time at sea

The TAS was a significant predictor in the model for Chapter 5. It has a linear relationship with the abundance of AF sea lice; as the TAS increased, so did the abundance of AF sea lice, as demonstrated in Figure 5.5. The ‘count week’ equivalent to TAS was used by Kristoffersen et al. (2014) and was a significant predictor in their model. The weight of the salmon had a positive association with the abundance of juvenile lice in Chile; this parameter was considered a proxy for the age of the salmon (Kristoffersen et al., 2013). Other studies had demonstrated the opposite results; smaller fish had higher densities of sea lice (number of sea lice per cm of surface area [Tucker et al., 2002]).

A similar predictor was not included in the model for Chapter 3 because, with the segment approach, the TAS could not have been calculated for segments as the lengths of these varied in length. A discrete variable for the representation of the TAS may be created for the segments, and it is likely that adding a version of the TAS in the segment model would improve the model fit as it did in Chapter 5. The reasoning behind the improved model fit is that the TAS may be a representation of the growth of the salmon, and there are otherwise no predictors that represent the growth of the salmon and fish size (weight and surface area) and age, which have associations with the abundance of sea lice. The TAS is likely a vital predictor to include in future models for the study area and prediction scenarios.

6.8 Prediction

The model's predictive capabilities in Chapter 5 were illustrated with an in-sample example and showed high prediction accuracy. This model may be adapted for prediction with a horizon of 3 weeks if predicted temperatures and decisions for treatments are provided in the model. The treatments will need to be decided for the future weeks to make the predictions, or the other option is to perform sensitivity analyses as the treatments are varied to observe the effects on the abundance of AF. The infestation pressures can be calculated from the lagged observations of the AF and PAAM. A predictive model for the aquaculture industry in the Bay of Fundy, NB, could aid in planning decisions for mitigation strategies as suggested for the state-space model for Grand Manan (Elghafghuf et al., 2021). A way to adapt such a model for ease of use by stakeholders is to

implement an app, like Shiny (Posit Software, Boston, USA), that can be implemented with R and is available online and could be password-protected to maintain confidentiality if desired.

6.9 Data and future studies using the Fish-iTrends©

The Fish-iTrends© database provides a valuable dataset that can utilized to a greater degree. The descriptive statistics as produced for Chapter 2 may be applied to produce real-time plots and reports to share with stakeholders and the public. It was understood that stakeholders of the Atlantic salmon industry in the Bay of Fundy, NB, may be apprehensive about public opinion in light of the strong opposition on the west coast in British Columbia. However, it is possible to increase the accessibility of sea lice data as demonstrated in other countries (e.g., Norway's Salmon Lice Data Sharing project from the HUB Ocean Foundation). The publication of raw data and programming code for research projects such as this one may improve the assessment of the study validity and allow for replication or expansion of the research as recommended by Sargeant and O'Connor (2014) and the "The REporting of studies Conducted using Observational Routinely-collected health Data (RECORD) Statement" for research in human populations (Benchimol et al., 2015).

Additionally, we propose that future research using the Fish-iTrends© database should include the opinions of the farmers, managers and veterinarians involved in sea lice counts. This can be achieved by applying a mixed methods approach

and could be beneficial as these stakeholders would have a more considerable influence on the development of the models. The benefits of this approach would be to better focus future research and funds towards models that produce exploitable outcomes. That is not to say that this research does not present any avenues for improving the mitigation of sea lice, but that a combination of qualitative and quantitative approaches may aid in developing tools and techniques for these stakeholders to use to mitigate sea lice.

The Fish-iTrends© database was constructed as a monitoring tool for *L. salmonis* with a prescribed sampling design and is well suited for studies with observational designs. The observational study design can be affected by selection, information, and confounding biases (Dohoo et al., 2014). For example, selection bias may arise due to the non-random sampling of salmon in the cages (Gautam et al., 2016). The aquaculture industry has a hierarchical organization. Thus, the data had this hierarchical organization and repeated measurements (counts), which implies non-independence among observations at each level (Dohoo et al., 2014). It should be noted that the hatcheries supply to more than one site, resulting in a cross-classification (Dohoo et al., 2014), but this information was not recorded in the Fish-iTrends© management system. The Fish-iTrends© data management system was designed with a specific function in mind to recognize confidentiality issues. However, it would benefit immensely from the addition of measurements of biomass and temperature and salinity measurements at set depths. The addition of salmon biomass data at sites and

cages may aid in constructing future models rather than depending on sensitivity analyses as performed by Elghafghuf et al. (2020). It was part of the General Regulation 91-158 (Government of New Brunswick, 2018) that the weight and length of salmon sampled be submitted to the registrar or a third-party database (e.g., Fish-iTrends©). However, in this case, this information was not submitted into the Fish-iTrends©. In all cases, the study design and model selection were made to minimize bias and work within the limitations of the dataset.

A note of little importance to the current project but may impact future analysis of Fish-iTrends© data were the changes in the regulations for counts submitted into the database. Chapter 2 outlined the General Regulation 91-158 (Government of New Brunswick, 2018). These regulations were repealed and not in force as of 22 November 2022. The proclamation of the new Aquaculture Act and supporting regulations were applied in April 2022 (Government of New Brunswick, nd). The new regulations (C. Leavitt, pers. comm.) were not published as of the writing of this document but had the following changes compared to General Regulation 91-158 (Government of New Brunswick, 2018):

- if a site has 6 or fewer cages, sample all cages
- if there is a high sea lice burden expected in specific cages within a site, samples must be taken from 2 of those cages, then 4 cages at random
- if treatment is scheduled, cages scheduled for treatment should be among those cages sampled

- counts must be performed within 4 days after treatment

These changes to the regulations should produce more consistent data that may aid in evaluating treatment effects and efficacy. Clarifying the timing for post-treatment counts may demonstrate a more robust response to treatment applications. Gautam et al. (2017) noted that the lag between the treatment and post-treatment count was an important predictor for the abundance of AF and PAAM.

Another change that may impact the industry and research using the sea lice monitoring data is the proposed review of the BMA structures (March 2023) and planned implementation by December 2025 (Government of New Brunswick, nd). This is because the BMAs were initially designed to manage ISAV outbreaks. Ideally, the BMAs should help reduce the connectivity of all pathogens, particularly for each type of agent (viral, bacterial, and parasitic), which share similarities in transmission patterns.

6.10 Recommendations to the New Brunswick Atlantic salmon aquaculture stakeholders

In light of the studies presented in this thesis, Atlantic salmon producers, farmers, and managers in NB should consider the following recommendations. First and foremost is to continue the data submission into the Fish-iTrends data management system. The Fish-iTrends data management system is well developed for annual reporting of sea lice and can be accessed for research studies such as this one. Further additions to the Fish-iTrends website could be

considered, such as a dashboard for the prediction of AF sea lice. This dashboard would be a refined product of the predictive model demonstrated in Chapter 5. Such a dashboard would require funding and farmer/scientist collaboration to develop a useful and user-friendly product.

NB aquaculture farming stakeholders should continue support of an integrated pest management plan and should also consider funding support toward studies to evaluate the efficacy of the treatments applied at varying water temperatures. The results of treatment efficacy trials would greatly benefit the producers and veterinarians toward selection of the most effective treatments and combination of treatments for *L. salmonis* mitigation (Jackson et al., 2016; Thompson et al., 2023). Combining treatments may already be prevalent, as is the case in Newfoundland with emamectin benzoate and azamethiphos treatment (Hamoutene et al., 2022). The results in Chapters 3 and 5 indicated that certain treatment effects varied per the water temperature at application. This is relevant to the current management practices at farms in NB, but especially since the sea water temperatures are increasing as a result of climate change (Fast and Dalvin, 2020). It is recommended that research on the apparent field treatment responses and the emerging resistance of treatments should be repeated every few years (e.g., Aaen et al., 2015) and Fish-iTrends can be an asset for this data management.

Additional recommendations for the mitigation of sea lice centre around geophysical characteristics of the Bay of Fundy, NB. The management zones should be redesigned as their original function was for the management of ISAV. Sea lice disperse across the BMAs suggesting the current structure does not contribute very much to reducing the external infestation pressure during the management of sea lice. The area could be divided into two large management zones thereby combining BMAs 1, 2A, 3A and 3C into one zone and BMAs 2B and 3B into another zone. Such a division is supported by the 10 km seaway dispersal distance identified in Chapter 3. Smaller management zones may be considered, but a hydrodynamic-particle tracking model would be necessary to identify networks and clusters. Within these suggested management zones, stocking and fallowing should be synchronized. Treatment synchronization should be achieved for the first year at sea, when there is a greater importance of the external infestation pressure to infest the newly stocked salmon hosts (Kragesteen et al., 2021). After the first year at sea, farmers and managers can apply treatments as they deem fit to decrease the internal infestation pressure, when external infestation pressure impacts are smaller than internal infestation pressure. For the process of synchronizing treatments, there should be a focus on in-feed treatments across many farms, although the in-feed treatments appeared to be less effective than other treatments in our studies. The focus on in-feed treatments is a result of the limited numbers of wellboats and other de-lousing equipment necessary for topical, thermal and Hydrolicer® delousing. A

sense of community should be promoted with strong communication across farms and companies; supporting knowledge exchange among Swiss dairy farmers was demonstrated to improve adoption of measures to decrease greenhouse gas emissions (Kreft et al., 2023). The infestation pressures should be re-evaluated regularly using the Fish-iTrends data and additional design changes to the management zones could be considered.

A final recommendation would be to develop agreements with the neighbouring Maine aquaculture industry stakeholders. With the current design of the BMAs, BMA 1 may be impacted by the sea lice from the Atlantic salmon farms in Maine. The effects of fallowing would be diminished from the external infestation pressure applied by Maine farms if they do not have similar fallowing schedules. Fostering the development of a database for sea lice counts, such as the Fish-iTrends, would also be beneficial for the Maine aquaculture stakeholders, but could also be used in future NB studies of sea lice counts.

6.11 Conclusions

The objectives were met to estimate the infestation pressures for *L. salmonis* for the Atlantic salmon aquaculture sites of the Bay of Fundy, NB. The estimation of infestation pressures and the dispersal of sea lice through the external infestation pressure was essential to inform us about the connectivity of lice among sites in the study area. New subjects for the connectivity of sea lice in the Bay of Fundy were identified. The percent surface area of water among sites may be a

surrogate for the relationship between coastal areas and sea lice abundance and should be further researched.

The infestation pressure within a site had a stronger impact on the abundance of AF sea lice compared to the effect of the infestation pressure among the sites in the models for the period 2009-2021. This information highlights the need for adequate control of sea lice at the site to reduce re-infestation of the salmon. It remains that the EIP was an important predictor in the models, and mitigation strategies should account for the output of sea lice from near neighbours. Area-wide management (currently BMA-level; e.g., fallowing) should continue, and synchronization of treatments for close neighbours should be considered. Another method to reduce the EIP for the Bay of Fundy, NB, would be to apply the concept of ‘firebreaks’ that would create clusters of sites and remove intermediary sites.

Depending on the treatment, treatments decreased the abundance of AF sea lice in the area. Interactions noted between treatments and temperature suggest an avenue for research that may provide prescribing criteria that vary per temperature measurement at the time of application.

6.12 References

Aaen, S. M., Helgesen, K. O., Bakke, M. J., Kaur, K., & Horsberg, T. E. (2015). Drug resistance in sea lice: A threat to salmonid aquaculture. Trends in Parasitology, 31(2), 72–81. https://doi.org/10.1016/j.pt.2014.12.006

Abolofia, J., Asche, F., and Wilen, J. E. (2017). The Cost of lice: Quantifying the impacts of parasitic sea lice on farmed salmon. Marine Resource Economics, 32(3), 329-349. https://doi.org/10.1086/691981

Aldrin, M., Storvik, B., Kristoffersen, A. B., and Jansen, P. A. (2013). Space-time modelling of the spread of salmon lice between and within Norwegian marine salmon farms. PLoS ONE, 8(5), 1-10. https://doi.org/10.1371/journal.pone.0064039

Aldrin, M., Jansen, P. A., and Stryhn, H. (2019). A partly stage-structured model for the abundance of salmon lice in salmonid farms. Epidemics, 26, 9-22. https://doi.org/10.1016/j.epidem.2018.08.001

Amundrud, T. L., and Murray, A. G. (2009). Modelling sea lice dispersion under varying environmental forcing in a Scottish sea loch. Journal of Fish Diseases, 32(1), 27-44. https://doi.org/10.1111/j.1365-2761.2008.00980.x

Benchimol, E. I., Smeeth, L., Guttmann, A., Harron, K., Moher, D., Petersen, I., … Langan, S. M. (2015). The REporting of studies Conducted using Observational Routinely-collected health Data (RECORD) Statement. PLoS Medicine, 12(10), 1-22. https://doi.org/10.1371/journal.pmed.1001885

Dohoo, I., Martin, W., and Stryhn, H. (2014). Veterinary Epidemiologic Research. (S. M. McPike, Ed.) (2nd ed.). Charlottetown, PEI: VER Inc.

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2020). Estimating sea lice infestation pressure on salmon farms: Comparing different methods using multivariate state-space models. Epidemics, 31(July 2019), 100394. https://doi.org/10.1016/j.epidem.2020.100394

Elghafghuf, A., Vanderstichel, R., Hammell, L., and Stryhn, H. (2021). State-space modeling for inter-site spread of sea lice with short-term population predictions. Ecological Modelling, 452. https://doi.org/10.1016/j.ecolmodel.2021.109602

EU Copernicus Marine Service Information (CMEMS). (2021). SSS SMOS L4 OI – LOPS-v2021. Marine Data Store (MDS). https://doi.org/10.1175/JTECH-D-20-0093.1

[dataset] Fish-iTrends© data management system for farm sea lice counts, program owned and operated by CAHS, AVC, UPEI, Charlottetown, Canada;

data submitted and owned by private fish farm companies. Dataset 2009-2018 queried on [2019-03-11]

[dataset] Fish-iTrends© data management system for farm sea lice counts, program owned and operated by CAHS, AVC, UPEI, Charlottetown, Canada; data submitted and owned by private fish farm companies. Dataset 2016-2021 queried on [2022-06-30]

Frenzl, B., Stien, L. H., Cockerill, D., Oppedal, F., Richards, R. H., Shinn, A. P., Bron, J. E., and Migaud, H. (2014). Manipulation of farmed Atlantic salmon swimming behaviour through the adjustment of lighting and feeding regimes as a tool for salmon lice control. Aquaculture. https://doi.org/10.1016/j.aquaculture.2013.12.012

Gautam, R., Boerlage, A. S., Vanderstichel, R., Revie, C. W., and Hammell, K. L. (2016). Variation in pre-treatment count lead time and its effect on baseline estimates of cage-level sea lice abundance. Journal of Fish Diseases, 39(11), 1297-1303. https://doi.org/10.1111/jfd.12460

Gautam, R., Vanderstichel, R., Boerlage, A. S., Revie, C. W., and Hammell, K. L. (2017). Effect of timing of count events on estimates of sea lice abundance and interpretation of effectiveness following bath treatments. Journal of Fish Diseases, 40(3), 367-375. https://doi.org/10.1111/jfd.12519

Government of New Brunswick. (2018). General Regulation, NB Reg 91-158. 1-14. https://www.canlii.org/en/nb/laws/regu/nb-reg-91-191/latest/nb-reg-91-191.html

Government of New Brunswick: Department of Agriculture, Aquaculture and Fisheries. nd. New Brunswick Finfish Aquaculture Growth Strategy 2022-2030.

Groner, M. L., Gettinby, G., Stormoen, M., Revie, C. W., and Cox, R. (2014). Modelling the impact of temperature-induced life history plasticity and mate limitation on the epidemic potential of a marine ectoparasite. PLoS ONE, 9(2), e88465. https://doi.org/10.1371/journal.pone.0088465

Groner, M. L., McEwan, G. F., Rees, E. E., Gettinby, G., and Revie, C. W. (2016). Quantifying the influence of salinity and temperature on the population dynamics of a marine ectoparasite. Canadian Journal of Fisheries and Aquatic Sciences, 73(May), 1281–1291. https://doi.org/10.1139/cjfas-2015-0444

Hamoutene, D., Oldford, V., and Donnet, S. (2022). Drug and pesticide usage for sea lice treatment in salmon aquaculture sites in a Canadian province from 2016 to 2019. Scientific Reports, 12(1). https://doi.org/10.1038/s41598-022-08538-w

Hamre, L., Bui, S., Oppedal, F., Skern-Mauritzen, R., and Dalvin, S. (2019). Development of the salmon louse *Lepeophtheirus salmonis* parasitic stages in temperatures ranging from 3 to 24°C. Aquaculture Environment Interactions, 11, 429-443. https://doi.org/10.3354/aei00320

Imsland, A. K. D., and Reynolds, P. (2022). In lumpfish we trust? The efficacy of lumpfish Cyclopterus lumpus to control *Lepeophtheirus salmonis* infestations on farmed Atlantic salmon: A Review. Fishes, 7(5). https://doi.org/10.3390/fishes7050220

Jackson, D., Moberg, O., Stenevik Djupevåg, E. M., Kane, F., & Hareide, H. (2018). The drivers of sea lice management policies and how best to integrate them into a risk management strategy: An ecosystem approach to sea lice management. Journal of Fish Diseases, 41(6), 927–933. https://doi.org/10.1111/jfd.12705

Junquera, P., Hosking, B., Gameiro, M., and MacDonald, A. (2019). Benzoylphenyl ureas as veterinary antiparasitics. An overview and outlook with emphasis on efficacy, usage and resistance. Parasite, 26(26), 33. https://doi.org/10.1051/parasite/2019026

Kragesteen, T. J., Simonsen, K., Visser, A. W., & Andersen, K. H. (2021). Estimation of external infection pressure and salmon-louse population growth rate in Faroese salmon farms. Aquaculture Environment Interactions, 13, 21–32. https://doi.org/10.3354/aei00386

Kreft, C., Huber, R., Schäfer, D., and Finger, R. (2023). Quantifying the impact of farmers' social networks on the effectiveness of climate change mitigation policies in agriculture. Journal of Agricultural Economics. https://doi.org/10.1111/1477-9552.12557

Kristoffersen, A. B., Jimenez, D., Viljugrein, H., Grøntvedt, R., Stien, A., and Jansen, P. A. (2014). Large scale modelling of salmon lice (*Lepeophtheirus salmonis*) infection pressure based on lice monitoring data from Norwegian salmonid farms. Epidemics, 9, 31–39. https://doi.org/10.1016/j.epidem.2014.09.007

Kristoffersen, A. B., Rees, E. E., Stryhn, H., Ibarra, R., Campisto, J. L., Revie, C. W., and St-Hilaire, S. (2013). Understanding sources of sea lice for salmon farms in Chile. Preventive Veterinary Medicine, 111(1–2), 165–175. https://doi.org/10.1016/j.prevetmed.2013.03.015

Morefield, R. D., and Hamlin, H. J. (2022). Larval salmon lice *Lepeophtheirus salmonis* exhibit behavioral responses to conspecific pre-adult and adult cues. Diseases of Aquatic Organisms, 149, 121-132. https://doi.org/10.3354/dao03666

Penston, M. J., Mckibben, M. A., Hay, D. W., Gillibrand, P. A. (2004). Observations on open-water densities of sea lice larvae in Loch Shieldaig, Western Scotland. Aquaculture Research, 35, 793-805. https://doi.org/10.1111/j.1365-2109.2004.01102.x

Pest Management Regulatory Agency (PMRA), Government of Canada. (2003). Integrated Pest Management. Overview: Integrated Pest Management of Sea Lice in Salmon Aquaculture: Overview. Health Canada, Ottawa, ON. Catalogue H114-9/2003E. publications.gc.ca/pub?id=9.686494&sl=0

Samsing, F., Johnsen, I., Treml, E. A., and Dempster, T. (2019). Identifying 'firebreaks' to fragment dispersal networks of a marine parasite. International Journal for Parasitology, 49(3-4), 277-286. https://doi.org/10.1016/j.ijpara.2018.11.005

Sargeant, J. M., and O'Connor, A. M. (2014). Issues of reporting in observational studies in veterinary medicine. Preventive Veterinary Medicine, 113(3), 323-330. https://doi.org/10.1016/j.prevetmed.2013.09.004

StataCorp (2021). Stata: Release 17. Statistical Software. StataCorp LLC, TX, USA

Thompson, C. R. S., Madaro, A., Nilsson, J., Stien, L. H., Oppedal, F., Øverli, Ø., Korzan, W. J., & Bui, S. (2023). Comparison of non-medicinal delousing strategies for parasite (*Lepeophtheirus salmonis*) removal efficacy and welfare impact on Atlantic salmon (*Salmo salar*) hosts. Aquaculture International. https://doi.org/10.1007/s10499-023-01167-8

Tucker, C. S., Sommerville, C., and Wootten, R. (2002). Does size really matter? Effects of fish surface area on the settlement and initial survival of *Lepeophtheirus salmonis*, an ectoparasite of Atlantic salmon *Salmo salar*. Diseases of Aquatic Organisms, 49(2), 145–152. https://doi.org/10.3354/dao049145

Fast, M. D., and Dalvin, S. (2020). Lepeophtheirosis (*Lepeophtheirus salmonis*). In P. T. K. Woo, J.-A. Leong, and K. Buchmann (Eds.), Climate Change and Infectious Fish Diseases (p. 529). Wallingford, UK; Boston, USA: CAB International.