12-2014

Phenology and Effects of Dams on the Success of Atlantic Salmon Smolt Migrations in the Penobscot River, Maine

Daniel S. Stich

Recommended Citation

Stich, Daniel S., "Phenology and Effects of Dams on the Success of Atlantic Salmon Smolt Migrations in the Penobscot River, Maine" (2014). *Electronic Theses and Dissertations*. 2244.
http://digitalcommons.library.umaine.edu/etd/2244

PHENOLOGY AND EFFECTS OF DAMS ON THE SUCCESS OF ATLANTIC SALMON SMOLT MIGRATIONS IN THE PENOBSCOT RIVER, MAINE

By

Daniel S. Stich

A.A. Schenectady County Community College 2006

B.T. State University of New York College of Agriculture and Technology at Cobleskill 2008

M.S. Virginia Polytechnic Institute and State University 2011

A DISSERTATION

Submitted in Partial Fulfillment of the

Requirements for the Degree of

Doctor of Philosophy

(in Wildlife Ecology)

The Graduate School

The University of Maine

December 2014

Advisory Committee:

Joseph Zydlewski, Associate Professor of Wildlife, Fisheries, and Conservation Biology
Michael Bailey, US Fish and Wildlife Service Fisheries Biologist
Michael Kinnison, Professor of Evolutionary Applications
John Kocik, National Marine Fisheries Service Fishery Biologist
Gayle Zydlewski, Associate Professor of Marine Sciences

DISSERTATION ACCEPTANCE STATEMENT

On behalf of the Graduate Committee for Daniel Stich I affirm that this manuscript is the final and accepted dissertation. Signatures of all committee members are on file with the Graduate School at the University of Maine, 42 Stodder Hall, Orono, Maine.

Dr. Joseph Zydlewski, Date

Associate Professor of Wildlife,

Fisheries, and Conservation Biology

LIBRARY RIGHTS STATEMENT

In presenting this dissertation in partial fulfillment of the requirements for an advanced degree at the University of Maine, I agree that the Library shall make it freely available for inspection. I further agree that permission for "fair use" copying of this dissertation for scholarly purposes may be granted by the Librarian. It is understood that any copying or publication of this dissertation for financial gain shall not be allowed without my written permission.

Signature:

Date:

PHENOLOGY AND EFFECTS OF DAMS ON THE SUCCESS OF ATLANTIC SALMON SMOLT MIGRATIONS IN THE PENOBSCOT RIVER, MAINE

By Daniel S. Stich

Dissertation Advisor: Dr. Joseph Zydlewski

An Abstract of the Thesis Presented
in Partial Fulfillment of the Requirements for the
Degree of Doctor of Philosophy
(in Wildlife Ecology)
December 2014

Atlantic salmon *Salmo salar* populations are diminished throughout their range and high marine mortality is among the drivers of the failure of many stocks to recover. A goal of salmon recovery is to maximize the number of juvenile ‘smolts’ entering the ocean to offset loss therein. Dam removals and changes to hydropower allocation in Maine’s largest river, the Penobscot River, have occurred as part of the Penobscot River Restoration Project (PRRP). These activities, in addition to stocking have the potential to influence the number of smolts reaching the ocean. Telemetry was used to investigate factors influencing initiation of migratory behavior, movement rates, migratory route, and survival through freshwater (FW) before and after changes to the system resulting from the PRRP, and behavior and survival of smolts during estuary migration.

Initiation of migration was influenced by smolt development, stocking location and environmental conditions. Smolts with the greatest gill Na^+, K^+-ATPase activity (physiological development) initiated migration 24 hours sooner than fish with the lowest gill NKA activity. Fish with the greatest cumulative temperature experience (accumulated thermal units: ATU) initiated migration 5 days earlier than those with lowest ATU. Smolts released furthest upstream initiated migration earlier than those released downstream, and movement rate increased 5-fold from upstream to the estuary. Movement rate increased from 2.8 $km\cdot h^{-1}$ to 5.4 $km\cdot h^{-1}$ in reaches where dams were removed, and decreased from 2.1 $km\cdot h^{-1}$ to 0.1 kmh^{-1} after powerhouse construction.

Proportional use of the Stillwater Branch was low (0.12, 95% CI = 0.06 – 0.25), and survival through the dams therein was relatively high (0.99) prior to installation of new powerhouses, decreasing slightly thereafter. Survival at Milford Dam, now the lowermost dam in the main-stem, was low (0.91) prior to increased power generation, whereas survival at Great Works and Veazie Dams was high (0.99 and 0.98) prior to removal. Survival was higher through free-flowing reaches (> $0.99\cdot km^{-1}$) than reaches containing dams (*c*. $0.95\cdot km^{-1}$). Survival was reduced at high (> 2000 m^3s^{-1}) or low (<300 m^3s^{-1}) flow, and was optimal between 12 °C and 17 °C. Survival increased following dam removal, but survival through those dams was high before removal. The greatest increase in survival (8%) followed turbine shutdown at Howland Dam.

Smolts experiencing greatest ATU arrived in the estuary 8 days earlier than those experiencing lowest ATU. Estuary arrival date was 10 days later for fish experiencing high flow than for fish experiencing low flow. Fish released furthest upstream arrived in the estuary 3 days later than those stocked further downstream, but moved 0.5 $km\cdot h^{-1}$ faster through the estuary. Estuary survival decreased by 40% with increasing number of dams passed (from 2 to 9). Estuary movement rate and survival both peaked in mid-May, and slowed from FW to ocean, likely resulting from tidal influences. Smolts became increasingly surface-oriented during passage from FW to ocean as salt water (SW) became more prevalent. In laboratory experiments, preference for SW by never exceeded

50% during smolt development. Thus, smolts likely select low salinity (i.e. surface) waters during migration through coastal areas.

Smolts with low gill NKA activity spent greater time in FW reaches of the estuary than those with high gill NKA activity. However, there was no difference in travel time through SW reaches of the estuary based on gill NKA activity. Fish with the highest gill NKA activity incurred 25% lower mortality through the estuary than fish with lowest gill NKA activity, and survival was lowest where SW was prevalent. These results underscore the importance of physiological preparedness on performance and the delayed effects of dams on survival of smolts during estuary migration, ultimately affecting marine survival estimates.

ACKNOWLEDGMENTS

This work is the result of a large number of people and groups who have invested time, effort, and financial support in excess of what could possibly have been accomplished by a single person, and I am indebted to each of them for their contributions. Financial support for this research was provided by the National Fish and Wildlife Foundation, the American Recovery and Reinvestment Act (ARRA) and National Oceanic and Atmospheric Administration (NOAA) Open Rivers Initiative through the Penobscot River Restoration Trust and NOAA National Marine Fisheries Service (National Marine Fisheries Service). Additional funding was provided by Brookfield Renewable Power (Great Lakes Hydro America), Maine Department of Marine Resmyces (DMR), The Nature Conservancy, The Penobscot Indian Nation, The University of Maine, U.S. Geological Survey (USGS), Maine Cooperative Fish and Wildlife Research Unit, and The West Enfield Fund. The U.S. Fish and Wildlife Service, Green Lake National Fish Hatchery provided juvenile Atlantic salmon and daily temperature data for smolt rearing pools in all years of this study, and was instrumental in facilitating the marking and stocking of hatchery-reared smolts for this work. The Maine DMR (Randy Spencer, Peter Ruksznіz, and Kevin Gallant) and Brookfield Renewable Power (Kevin Bernier and Dan Anderson) were key in trapping wild smolts for use in this research. In-river temperature data were made available by the Maine DMR and the U.S.G.S. John Beeman (U.S.G.S. Western Fisheries Research Center) loaned radio-telemetry equipment during 2012. Access to private property was provided by Brookfield Renewable Power (c/o Scott Hall), the Eddington salmon Club (c/o Buck Owen), The Medway Sno-rovers (c/o David Violette), The Penobscot Indian Nation (c/o Sparky

Clark), and Chip Reir. I acknowledge substantial contributions to this project by Michael Bailey, Cory Gardner, Christopher Holbrook, and Andrew O'Malley (University of Maine) both prior to and during my involvement. I thank Matthew Altenritter, Megan Altenritter, Phillip Dionne, Stephen Fernandes, Catherine Johnston, Kevin Lachapelle, and Matthew Wegener (School of Marine Sciences, University of Maine) as well as Graham Goulette and James Hawkes (NMFS NEFSC) for their collaborative efforts in maintaining the acoustic receiver network in the Penobscot River, the estuary, and the bay. Additionally, I would like to thank Alia Al-Humaidhi, George Aponte Clarke, Wesley Ashe, Charlie Baeder, John Banks, Meg Begley, Keith Boyer, Brandon Capron, Sparky Clark, Aubrey Curley, Matthew Dzaugis, Anitra Firmenich, Graham Goullete, Graham Griffin, Ann Grote, Ed Hughes, Chris Introne, Betsy Irish, Lisa Izzo, Mike Kinnison, Ian Kiraly, Christine Lipsky, Phillip Lung, George Maynard, Dan McCaw, Jeff Murphy, Ana Rapp, Silas Ratten, Margo Relford, Laura Rose-Day, Joshua Royte, Rory Saunders, Tim Sheehan, Doug Sigmyney, Justin Stevens, Chandler Smith, Erin Snook, Fred Trasko, Haley Vieman, Trevor Violette, Dan Weaver, Fred Yost, and Gayle Zydlewski, and Orion Zydlewski for assistance in the field, lab, and/or as reviewers for the various components of this manuscript.

I thank my advisory committee, especially my advisor, Joe Zydlewski, for patient and thoughtful mentoring on and off campus that helped to make my time here about more than just a degree. I would like to acknowledge the support of my friends and family, especially my parents (Cathy and Steve Stich) and mother in law (Trudy Hinkle) without whose multi-faceted support this experience would have been far less enjoyable, and very likely impossible. Finally, I am forever indebted to my wife, Ashley Stich, and my daughters, Adeline and Aurelia Stich, for the sacrifices they have made over the years so that I could pursue my dreams, and for their unwavering love and support that transcend state lines, international boundaries, and periods of physical and mental absence in the name of fish.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1

PHENOLOGY AND MOVEMENT RATES OF ATLANTIC SALMON SMOLTS IN FRESH WATER

INTRODUCTION

Seaward migration through freshwater and estuarine habitats to ocean feeding grounds represents a critical transition in the life-history of anadromous fishes (Zydlewski and Wilkie 2013). In Atlantic salmon *Salmo salar* smolts, migration is associated with high predation risk (Blackwell et al. 1997; Kocik et al. 2009; Hawkes et al. 2013), dynamic environments (Thorstad et al. 2012), and new food sources (Haugland et al. 2006). Seaward migration by smolts occurs in four primary phases: initiation, downstream migration (fresh water), estuary passage, and early marine migration (McCormick 2013). A building body of evidence suggests ocean temperatures play a critical role in the success of early marine migrants (Friedland 1998; Friedland et al. 2003, 2014), and sea surface temperatures are predictive of the timing of smolt arrival in estuaries throughout the world (Otero et al. 2014). Timing of estuary arrival and ocean entry are important for survival (Stich et al. in review). However, the arrival of smolts in the ocean is determined by the integration of cues experienced by smolts far from the point of ocean entry (McCormick et al. 1998). As such, these cues play a critical role in determining when smolts initiate migration and how fast these fish move.

Smolting in Atlantic salmon involves synchronous changes in the physiology, morphology, and behavior that prepare the salmon for a life at sea (McCormick et al. 1998). Smolting includes, among other changes, increased saltwater tolerance and upregulation of enzymes involved with ion transport (such as gill Na^+, K^+-ATPase [NKA] activity). Loss of territoriality, inhibition of positive rheotaxis, and adoption of schooling behavior also are characteristics of smolting (McCormick et al. 1998). Environmental cues, particularly photoperiod, control physiological transformation (McCormick et al. 1987), and onset of migratory behavior (Whalen et al. 1999; Zydlewski et al. 2005; Zydlewski et al. 2014) in hatchery- and wild-reared Atlantic salmon smolts. These processes are adapted for effectively transitioning to the marine environment. Dams can interrupt migration of Atlantic salmon smolts through injury (Stier and Kynard 1986; Mathur et al. 2000; Music et al. 2010) or migratory delay (Holbrook et al. 2011). Delays at dams result in increased risk of predation (Poe et al. 1991; Blackwell and Juanes 1998) or may result in mismatch of physiological preparedness for ocean entry and timing of ocean entry through temperature-related loss of smolt characteristics (McCormick et al. 1999; McCormick et al. 2009; Marschall et al. 2011) and high mortality during early marine migration (Petrosky and Schaller 2010). Such an effect has been implicated in significant dam-related estuary mortality (Stich et al. in review). Recent dam removals, such as those in the Kennebec and Penobscot Rivers (Day 2006) in Maine, USA allow examination of how dams, in conjunction with other influences, affect migration behavior.

Many rivers with Atlantic salmon rely heavily on stocking for population persistence (USASAC 2014). In light of low marine survival, a primary goal of management is to maximize the number of smolts leaving coastal systems (Hansen et al. 2012; Russell et al. 2012). Decisions about where and when to stock hatchery-reared smolts not only influence survival in freshwater based on the presence of dams (Holbrook et al. 2011), but also can influence timing of ocean entry (Otero et al. 2014). The complex relationships between timing, stocking location, and environmental conditions with ecology and survival of smolt migration are poorly characterized.

The goal of this study was to investigate influences of dams, stocking decisions (where and when to stock), and environmental conditions on movement rates of smolts through freshwater and determine how those same factors influence initiation of migration by hatchery-stocked smolts. I had two specific objectives to this end. First, I used acoustic telemetry data from 2005 through 2014 in the Penobscot River to model how movement rate of hatchery- and wild-reared smolts was influenced by 1) presence of dams and head ponds, 2) distance from the ocean, 3) environmental conditions (e.g., temperature and discharge), and 4) characteristics indicative of physiological development. Second, I used telemetry data to model relationships between initiation of migration behavior by hatchery-reared smolts and stocking conditions, including 1) stocking location, 2) temporal variability in environmental conditions at stocking, and 3) spatial variability in release locations used.

METHODS

Study site

The Penobscot River (Figure 1.1) is the largest river in Maine, USA, and drains an area of approximately 22 000 km^2. Limited wild spawning of Atlantic salmon occurs in the system, and this is extensively supplemented by annual stocking of eggs and fry in headwater streams (USASAC 2014). Because these life stages are not marked, naturally reared hatchery fish are morphologically indistinguishable from wild-spawned fish and all are referred to hereafter simply as ‘wild’. Peak migration of wild smolts generally occurs during late April and early May each year (USASAC 2014). While exact estimates of the proportion of the Penobscot River smolt run made up by stocked fish is not known, sampling in the bay suggests that more than 90% of the run results from smolt stocking (Sheehan et al. 2011). Consequently, most (*c*. 83%) of the spawners that return to the Penobscot River each year result from the stocking of hatchery-reared (hereafter ‘hatchery’) smolts (USASAC 2012).

Beginning in 2009, a suite of large-scale conservation activities were initiated as part of a basin-wide restoration project, the Penobscot River Restoration Project (PRRP; Day 2006). The goal of the PRRP was to balance the production of hydropower in the Penobscot River with the revitalization of 11 species of diadromous fishes in the catchment, including Atlantic salmon. The PRRP resulted in significant changes to the hydro system (i.e. location and operation of hydropower dams) in the Penobscot River. These changes include 1) the removal of two main-stem dams in the lower river (Great Works [rkm 58, June 2012] and Veazie [rkm 45, July 2013], Figure 1.1) and 2) decommissioning of a third dam (rkm 99, Howland, Figure 1.1) in the mouth of the Piscataquis River (Federal Energy Regulatory Commission [FERC] 2009).

Other changes in the Penobscot River have resulted from the conditions of The Lower Penobscot River Basin Comprehensive Settlement Accord (‘Settlement Accord’; FERC 2004) that governed the re-allocation of hydropower throughout the river. The Settlement Accord allowed for increased hydropower generation at six facilities in the river (FERC 2004). Generating capacity was increased at Milford Dam at rkm 60 in the main-stem by raising head pond elevation and increasing the number of turbines at

Milford Dam (FERC 2009). Concurrently, increased head-pond elevation at Stillwater Dam (FERC 2005), and the construction of new powerhouses at Stillwater and Orono Dams (2013) doubled hydropower generation at each dam (Figure 1.1).

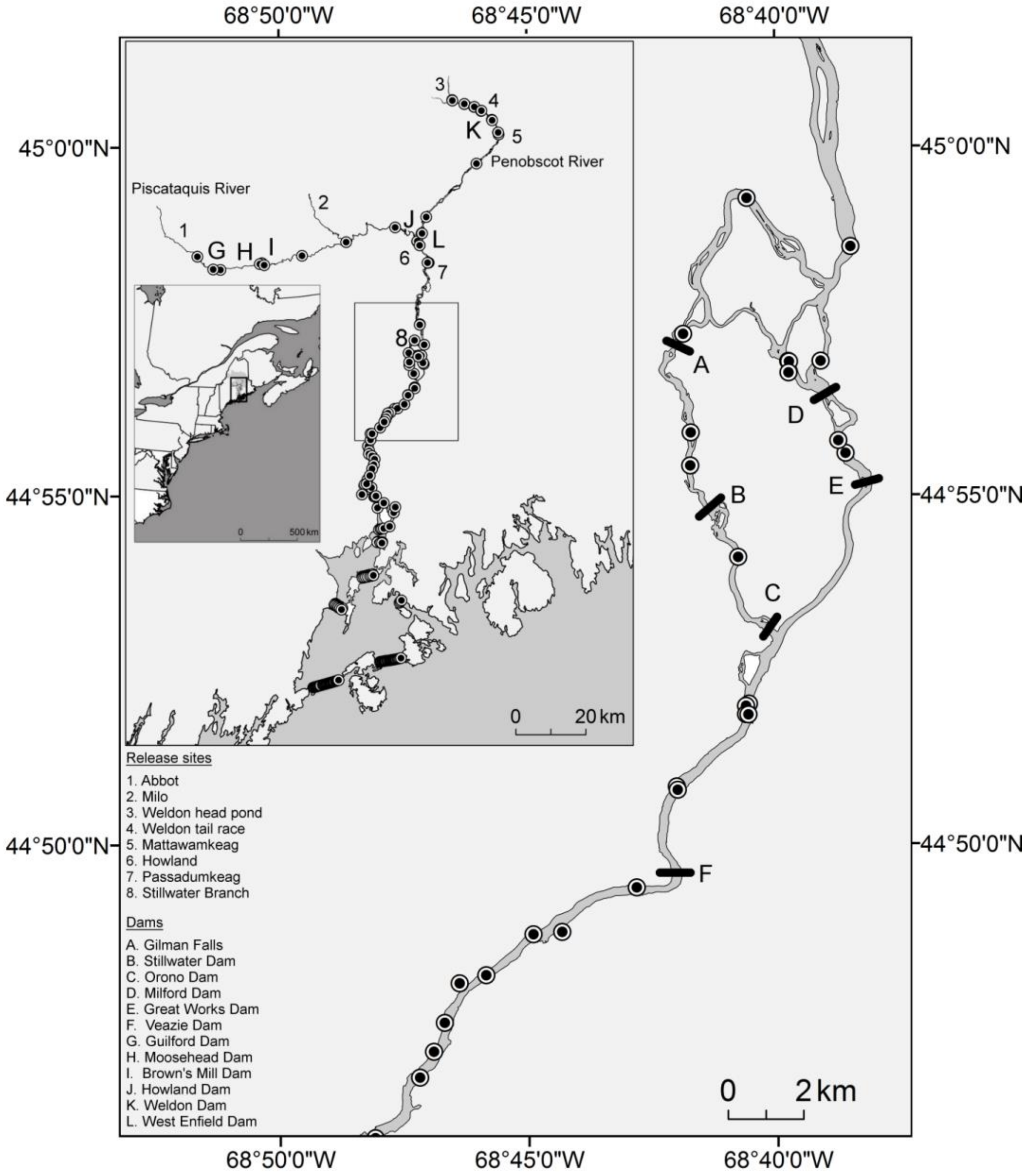


Figure 1.1. Map of the Penobscot River in Maine, USA. Shown are location in North America (small inset), locations of acoustic receivers, release sites and dams, and potential migratory routes in the lower river through the Stillwater Branch or the main-stem Penobscot River around a large island. Head of tide is located immediately downstream of Veazie Dam (F).

Acoustic receiver array

From 2005 through 2014, a network of stationary acoustic receivers was deployed from the headwaters of the Penobscot River and its primary tributary, the Piscataquis River, downstream to the Gulf of Maine (Figure 1.1). The receiver network was deployed collaboratively in all years by the United States Geological Survey (USGS) Maine Cooperative Fish and Wildlife Research Unit, the University of Maine, and the National Oceanic and Atmospheric Administration (NOAA) National Marine Fisheries Service Northeast Fisheries Science Center. Depending on year of the deployment, the network included up to 200 VR2 and/or VR2-W acoustic receivers (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) scanning continuously at 69 kHz. Receivers in the freshwater reaches and in the Penobscot Estuary were moored on bottom using reinforced cement anchors. In the bay, receivers were suspended approximately 10 m below the surface of the water in the bay. Where necessary, multiple receivers were deployed across a transect to provide adequate coverage across wider reaches, and detections of fish at these receivers were pooled as a single location. Detections of fish at receivers downstream of the head of tide at Veazie Dam (Figure 1.1, F) were pooled as a single, terminal detection for this study.

Acoustic tagging and releases

From 2005 through 2014, a total of 2,056 Atlantic salmon smolts was acoustically tagged and released in the Penobscot River (Table 1.1), of which 1,639 (80%) were 18-month, hatchery smolts from the U.S. Fish and Wildlife Service (USFWS) Green Lake National Fish Hatchery (GLNFH) and 417 (20%) were wild smolts. Fish handling procedures and acoustic tagging methods follow those of Holbrook et al. (2011) and Stich et al. (2014) and were identical in all years (2005–2014). Smolts were anaesthetized using a 100 mg·L^{-1} solution of MS-222 (buffered with 20-mmol $NaHCO_3$; pH = 7.0), fork length (L_F; mm) and mass (g) were measured. Condition factor (K) for each fish was calculated as:

$$K = \frac{\text{Mass} \times 100\,000}{L_F^3} \qquad \text{(Equation 1.1)}$$

For each fish, a small (1-cm) incision was made slightly offset from the ventral line and about 1-cm posterior to the pectoral fin girdle. An acoustic tag was inserted intraperitoneal and the incision was closed with two simple, interrupted knots using 4-0 absorbable vicryl sutures (Ethicon, Somerville, New Jersey, USA). Model V7-2L (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) tags were used in 2005 and for wild fish in 2011. In all other years, I used model V9-6L or V9-6x tags (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada). Expected battery life of the tags was 80-82 days in all years. Numbers of fish and release sites varied among years (Table 1.1). Hatchery smolts were released at up to five locations per year, and wild smolts at up to three locations per year.

Gill NKA activity

A nonlethal gill biopsy (4-6 filaments) was taken from the front, left gill arch of each fish prior to tagging. Individual biopsies were stored at -80°C in 100 µL SEI buffer (250 mM sucrose, 10 mM Na_2-EDTA, 50 mM imidazole) for later analysis of gill NKA (enzyme code 3.6.3.9; IUBM 1992) activity (expressed as µmol ADP·mg protein^{-1}·h^{-1}) using the method of McCormick (1993). Concentration of NADH at 25°C and 340 nm was used to measure kinetic rate of ouabain-inhibitable ATP hydrolysis, and protein concentration in gill samples was determined using the bicinchoninic acid (BCA) method (Smith et al. 1985). Gill samples from each fish were analyzed in triplicate for gill NKA activity and protein concentration.

Environmental data

Mean daily water temperature data for each year were collected from the USGS gage (USGS gage station 01036390) at West Enfield Dam (Figure 1.1) where available. I used mean daily water temperature data collected by Maine DMR for years (2005 and 2006) during which gage data were not available (Randy Spencer, Maine DMR, unpublished data). Discharge (Q, $m^3 \cdot s^{-1}$) data used in all analyses were mean daily values collected from the USGS West Enfield gage. Accumulated thermal units (ATU) experienced by wild fish each year were calculated from river temperature data between 1 January and capture date of individual fish. I calculated ATU over the period from 1 January to release date for hatchery smolts using mean daily temperatures (°C) from outdoor rearing pools at GLNFH for each year (A. Firmenich, US Fish and Wildlife Service, unpublished data). Photoperiod was calculated from ordinal date and latitude for 1) capture location of wild fish or 2) GLNFH for hatchery fish using the ‘geosphere’ package in R (R Development Core Team 2014).

Table 1.1. Descriptive statistics for acoustically tagged smolts from 2005 through 2014. Stocking locations with release rkm, number released (n), mean release date (Date) and mean (± SD) of fork length (L_{F}, mm), mass (g), and gill Na^{+}, K^{+}-ATPase activity (NKA, μmol ADP·mg protein^{-1}·h^{-1}) of acoustically tagged wild(W)- and hatchery (H) Atlantic salmon smolts released in Penobscot and Piscataquis Rivers 2005–2014.

Year	Release Site	Origin	n	LF	Mass)	NKA	Date
2005	Howland (99)	H	150	189(± 11)	75(± 15)	6.6(± 2.0)	23 Apr
2005	Mattawamkeag (144)	H	40	189(± 15)	77(± 21)	5.2(± 1.5)	14 Apr
2005	Milo (142)	H	85	191(± 11)	77(± 14)	8.1(± 1.9)	24 Apr
2005	Weldon (149)	W	60	180(± 18)	53(± 17)	9.1(± 1.9)	26 May
2006	Milo (142)	H	72	197(± 11)	87(± 18)	5.0(± 1.2)	24 Apr
2006	Weldon (149)	H	146	199(± 15)	87(± 19)	4.7(± 1.9)	24 Apr
2006	Weldon (149)	W	73	189(± 8)	63(± 8)	4.2(± 0.9)	8 May
2009	Milo (142)	H	100	180(± 8)	62(± 9)	3.3(± 1.4)	30 Apr
2009	Passadumkeag (92.3)	H	100	180(± 9)	63(± 10)	3.0(± 0.8)	29 Apr
2010	Abbot (187)	W	75	169(± 8)	45(± 7)	4.7(± 1.1)	2 May
2010	East Branch (162)	W	74	181(± 13)	55(± 13)	4.7(± 0.9)	7 May
2010	Milo (142)	H	100	189(± 11)	72(± 13)	4.5(± 1.0)	24 Apr
2010	Passadumkeag (92.3)	H	100	186(± 11)	69(± 14)	4.6(± 1.1)	24 Apr
2011	Abbot (187)	W	75	146(± 8)	29(± 5)	2.6(± 1.2)	11 May
2011	East Branch (162)	W	60	168(± 21)	46(± 18)	3.6(± 1.5)	24 May
2011	Milo (142)	H	100	192(± 13)	76(± 18)	5.0(± 1.5)	25 Apr
2011	Passadumkeag (92.3)	H	100	194(± 13)	77(± 17)	5.4(± 1.5)	27 Apr
2012	Abbot (187)	H	72	199(± 10)	84(± 14)	3.3(± 1.3)	20 Apr
2012	East Branch (162)	H	85	200(± 11)	85(± 14)	3.5(± 1.6)	20 Apr
2013	Abbot (187)	H	75	185(± 11)	70(± 13)	2.8(± 2.0)	16 Apr
2013	East Branch (162)	H	82	185(± 9)	71(± 11)	2.7(± 2.1)	16 Apr
2014	Abbot (187)	H	75	190(± 11)	70(± 13)	5.1(± 1.9)	2 May
2014	East Branch (162)	H	82	208(± 177)	66(± 12)	5.5(± 2.8)	2 May
2014	Stillwater (62)	H	75	190(± 9)	69(± 10)	4.9(± 2.0)	2 May

Movement rate

Detections of wild and hatchery smolts at upstream and downstream receivers that delimited reaches were used to calculate movement rates through each of the reaches. Movement rate was expressed as kilometers per hour (R_{ij}, km·h^{-1}), and was calculated as distance traveled in reach j ($D_{ij,}$ km) divided by time of first detection at each location (T_{ij}) for each fish (i) using:

$$R_{ij} = \left(\frac{D_{ij}}{T_{ij}} \right) \qquad \text{(Equation 1.2)}$$

I recognize that velocity also has been expressed as body lengths per second (bl·s^{-1}) in other systems (see Thorstad et al. 2012). However, because of the scale of movements (generally > 1 km), the duration of most movements measured (hours or days), and the fact that I actually was assessing velocity, I chose to present the results of this study in terms of km·h^{-1}; however based on potential concerns about the relation of velocity to body length, I ran models using bl·s^{-1}as the response variable and found no differences in the results. As such, I present km·h^{-1} for the reasons given above as well as for simplicity.

Models of movement rate

I used generalized linear mixed-effects models (GLMM; Zuur et al. 2009) in the ‘lme4’ package in R (R Development Core Team 2014) to estimate effects of covariates on movement rate. I included an individual-based random effect on the intercept in all models to account for repeated and unequal numbers of observations for each fish. Movement rate (km·h^{-1}), must be greater than zero and was right skewed. Therefore, I

$\log_e$-transformed movement rate prior to analysis. Variables used to represent fish characteristics included rearing history (hatchery or wild), condition factor (K), gill NKA activity, release rkm, and release date. Environmental variables included location within the catchment (rkm), photoperiod (i.e., day length) on the first date of each movement, discharge, temperature, and reach type (dam, free-flowing, or head pond). I used an information-theoretic approach to model selection based on Akaike's information criterion (Burnham and Anderson 2002) to evaluate the relative support for models containing *a priori* combinations of covariate effects on movement rates of hatchery and wild fish. I considered a covariate to have a statistically significant effect if the 95% confidence interval for the coefficient did not overlap zero. To evaluate the relative support for candidate models I used Akaike's information criterion, corrected for sample size (AICc; Burnham and Anderson 2002). I assumed models with ΔAICc < 2.0 to have similar support (Burnham and Anderson 2002). The probability that any given model was the best in the candidate set was estimated as the AICc weight (w_i). Approximation of a variance inflation factor ($\hat{c}$) for the most-parameterized model indicated reasonable model fit ($\hat{c} \leq 1.00$), so model selection was not adjusted.

Assessing effects of dams on movement rate

I sought to assess changes in movement rate through the lower Penobscot River following the removal of Great Works and Veazie Dams in 2012 and 2013 and the addition of a new powerhouse at both Stillwater and Orono Dams in the Stillwater Branch in 2013. Because Milford Dam was located about 2 km upstream of Great Works Dam, and because Veazie Dam formerly was located near the head of tide in the

Penobscot River Estuary, I examined changes in movement rates through four reaches between the tailrace of Milford Dam and the head of the estuary in the main-stem of the Penobscot River (rkm 44–59) to assess effects of dam removal on movement rate. I examined changes in movement rate throughout the Stillwater Branch (rkm 51–63). I tested for shifts in median movement rate through the main-stem Penobscot River in years before (2005–2013) and after the removal of *both* Great Works and Veazie Dams (2014) and before (2005–2013) and after (2014) completion of powerhouse construction at Stillwater and Orono Dams using Wilcoxon ranked sums tests (Zar 1999).

Initiation of migratory behavior by hatchery smolts

I assumed that a minimum downstream movement greater than 5 km by hatchery smolts was indicative of the initiation of migratory behavior (hereafter ‘initiation’) for this study. Initiation by acoustically tagged smolts was indexed as the total amount of time taken by each tagged smolt to move the first 8–25 km (depending on proximity of receiver locations to release sites) in a downstream direction. This range was used because of variability in the first downstream receiver location that could be used to define initiation for each release site. Given average movement rate, this introduced a potential error of up to 8 hours in initiation time based on variability in distance to first location.

Models of migration initiation

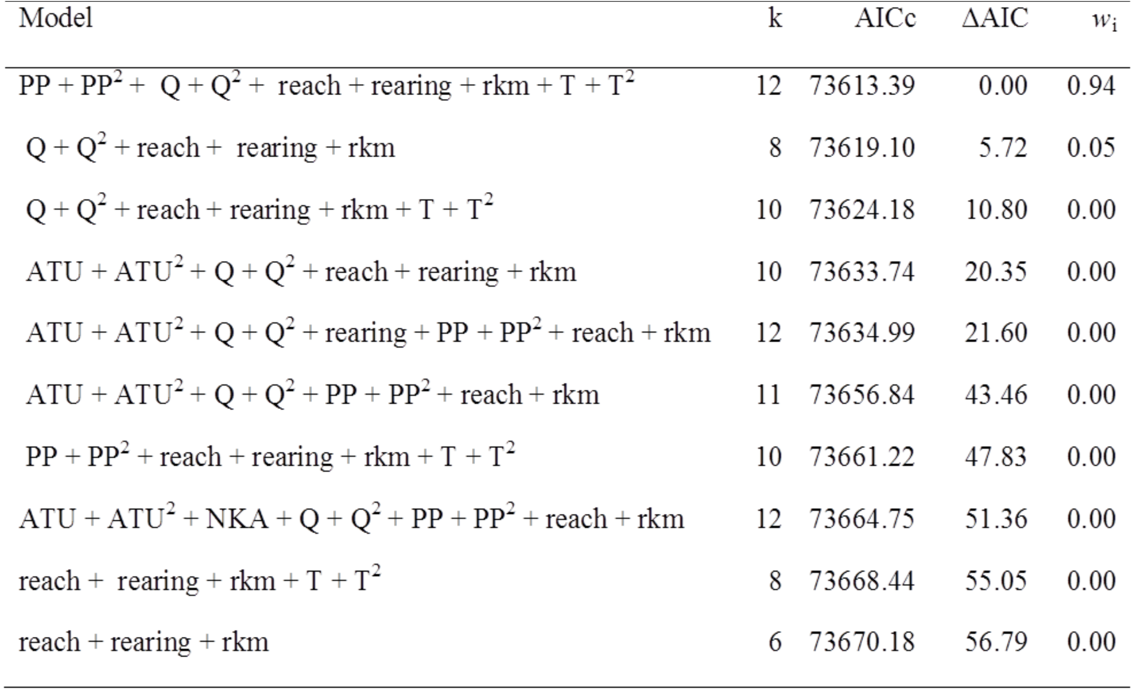
I used general linear models (Montgomery et al. 2006) to estimate effects of the smolt development (gill NKA activity and ATU), distance of release from the ocean, and environmental conditions (discharge, photoperiod, and temperature) on migration initiation. The response variable (time to initiation) was $\log_e$ transformed prior to analysis because it was right-skewed, and because negative predictions of time were not biologically realistic. All results are presented on the real scale of the variable (hours). I investigated potential support for non-linear effects of ATU, discharge, photoperiod, and temperature through the inclusion of a second-order term for these covariates in a subset of models. Because the distance used for assigning initiation varied between release sites (see above), I incorporated the distance between release and acoustic receivers as an explanatory variable in all models of initiation. Otherwise, model selection and goodness of fit were addressed in the manner used for models of movement rate (above).

RESULTS

Movement rate of wild and hatchery smolts

The best predictors of Atlantic salmon smolt movement rate were discharge, release site, photoperiod (day length), physiological development, rearing history, and temperature (Table 1.2), while distance to first location appeared to have the smallest effect on initiation. Average movement rate of Atlantic salmon smolts during freshwater migration in the Penobscot River 2014 was faster through free-flowing reaches of the system (2.1 $\text{km}\cdot\text{h}^{-1}$) than through reaches that contained dams (1.9 $\text{km}\cdot\text{h}^{-1}$) or head ponds (1.8 $\text{km}\cdot\text{h}^{-1}$, Figure 1.2 and Table 1.3). Movement rate did not differ among reaches that contained dams compared to reaches that contained head ponds (Figure 1.2). The reduction in movement rate at dams and head ponds was 6% greater in wild fish than in hatchery fish (Figure 1.2).

Table 1.2. Model-selection statistics for the ten best models used to quantify variation in movement rate ($km \cdot h^{-1}$) by Atlantic salmon smolts in the Penobscot River, Maine USA 2005–2014. Number of parameters estimated in each model is k, $AICc_i$ is the Akaike information criterion for each i^{th} model, $\Delta AICc_i$ is the difference between the AICc of each i^{th} model and the best model in the candidate set, and w_i is the relative probability that each i^{th} model is the best in the candidate set. Explanatory variables are defined as: accumulated thermal units (ATU), discharge (Q), distance between release and relocation (Distance), fork length (L_F), gill NKA activity (NKA), kilometers from ocean (rkm), photoperiod (PP), reach type (‘reach’, types= dam, free-flowing, or head pond), rearing history (‘rearing’, hatchery or wild), and river temperature (T).



Model	k	AICc	ΔAIC	w_i
PP + PP^2 + Q + Q^2 + reach + rearing + rkm + T + T^2	12	73613.39	0.00	0.94
Q + Q^2 + reach + rearing + rkm	8	73619.10	5.72	0.05
Q + Q^2 + reach + rearing + rkm + T + T^2	10	73624.18	10.80	0.00
ATU + ATU^2 + Q + Q^2 + reach + rearing + rkm	10	73633.74	20.35	0.00
ATU + ATU^2 + Q + Q^2 + rearing + PP + PP^2 + reach + rkm	12	73634.99	21.60	0.00
ATU + ATU^2 + Q + Q^2 + PP + PP^2 + reach + rkm	11	73656.84	43.46	0.00
PP + PP^2 + reach + rearing + rkm + T + T^2	10	73661.22	47.83	0.00
ATU + ATU^2 + NKA + Q + Q^2 + PP + PP^2 + reach + rkm	12	73664.75	51.36	0.00
reach + rearing + rkm + T + T^2	8	73668.44	55.05	0.00
reach + rearing + rkm	6	73670.18	56.79	0.00

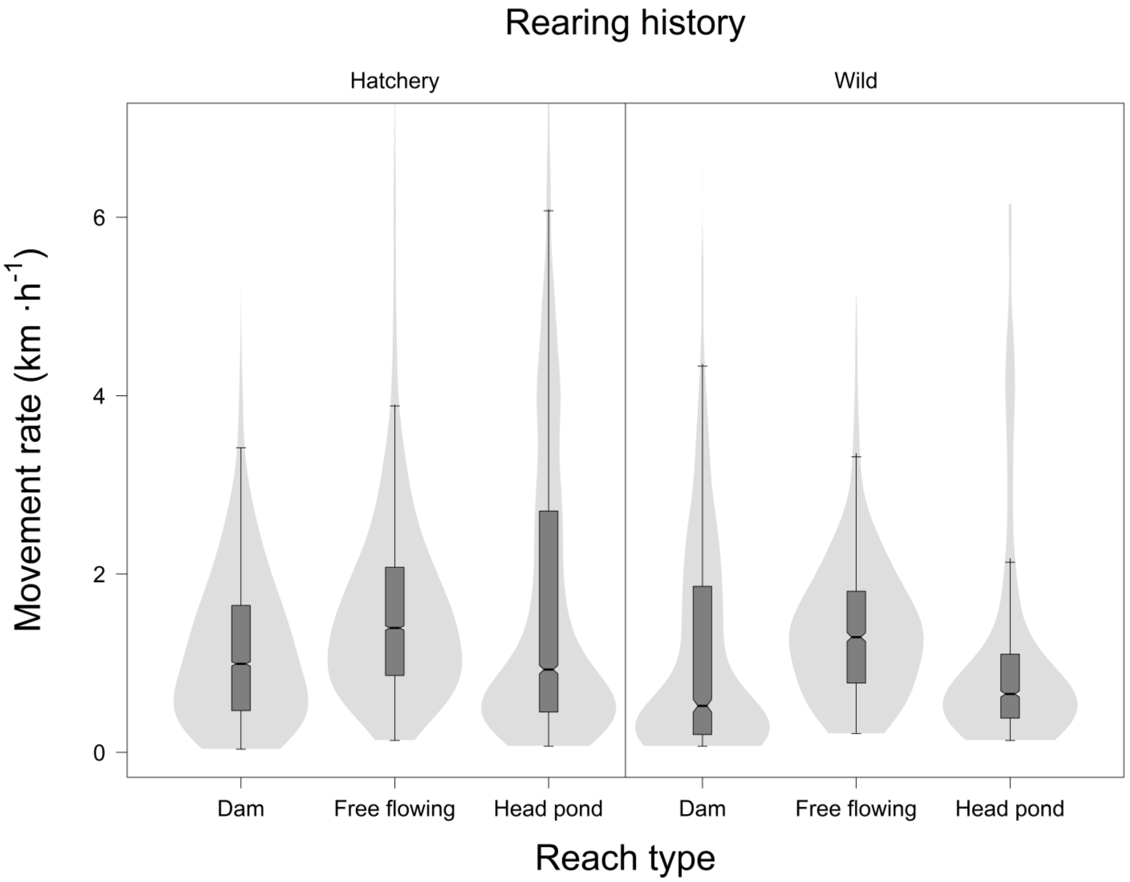
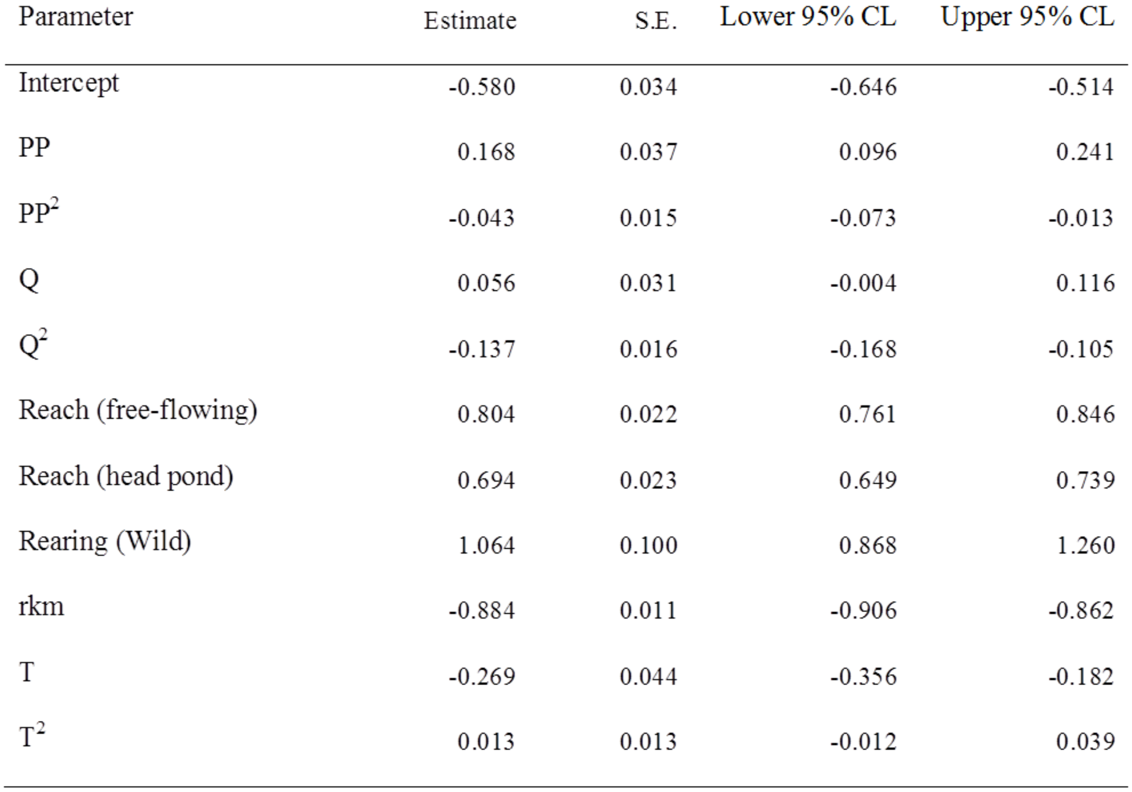


Figure 1.2. Changes in movement rate of Atlantic salmon smolts in the Penobscot River with respect to reach type and rearing history (hatchery or wild). Notches in boxes for hatchery (left panel) and wild (right panel) smolts indicate median movement rate within each reach type, box ends indicate 75% confidence intervals, and whiskers are 95% CI. The light gray polygons are violin plots showing density of predicted movement rate for each rearing history in each reach type.

Discharge (Q) affected movement rate in a complex fashion (Table 1.3). Movement rate of smolts was slowest at lowest or highest discharges observed (212–1 580 $m^3 \cdot s^{-1}$), and fastest at discharges (*c*. 750 $m^3 \cdot s^{-1}$) near the middle of this range (Figure 1.3a). Movement rate of smolts increased with photoperiod until late in the smolt run, at which point the relationship became highly variable due to low sample sizes (Figure 1.3b and Table 1.3). Temperature was inversely related to movement rate, with slower movement rates at higher temperatures. Over the range of temperatures observed (4–16 °C), movement rate decreased by about 8 km·h^{-1} (Figure 1.3d), although the relationship was highly variable at the highest temperatures due to low sample sizes.

Table 1.3. Mean and 95% confidence limits (CL) of estimated regression coefficients for the best model of the movement rate of Atlantic salmon smolts in the Penobscot River, Maine USA 2005–2014. Explanatory variables are defined as: discharge (Q), kilometers from ocean (rkm), photoperiod (PP), reach type (‘reach’, types= dam, free-flowing, or head pond), rearing history (‘rearing’, hatchery or wild), and river temperature (T).



Parameter	Estimate	S.E.	Lower 95% CL	Upper 95% CL
Intercept	-0.580	0.034	-0.646	-0.514
PP	0.168	0.037	0.096	0.241
PP^2	-0.043	0.015	-0.073	-0.013
Q	0.056	0.031	-0.004	0.116
Q^2	-0.137	0.016	-0.168	-0.105
Reach (free-flowing)	0.804	0.022	0.761	0.846
Reach (head pond)	0.694	0.023	0.649	0.739
Rearing (Wild)	1.064	0.100	0.868	1.260
rkm	-0.884	0.011	-0.906	-0.862
T	-0.269	0.044	-0.356	-0.182
T^2	0.013	0.013	-0.012	0.039

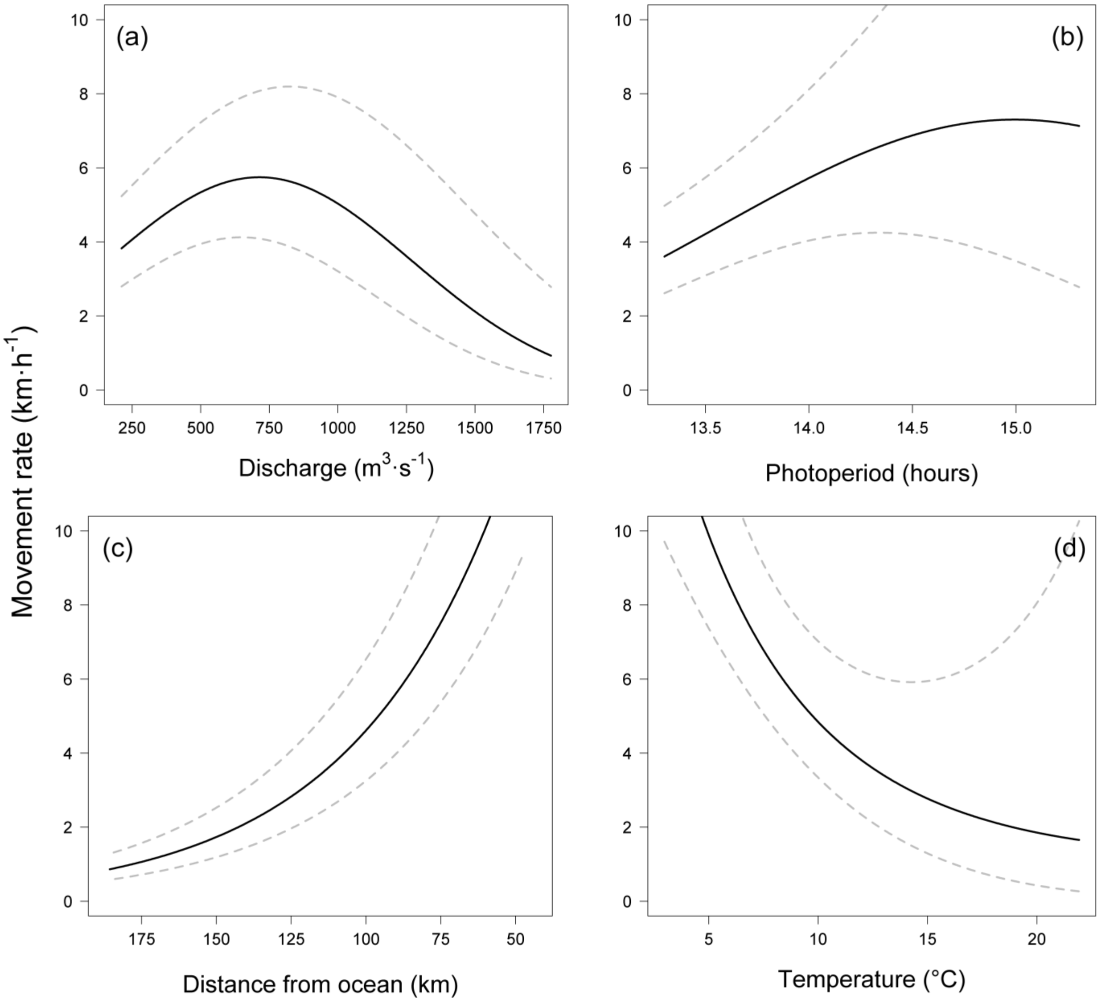


Figure 1.3. Covariate effects on freshwater movement rate of smolts 2005–2014. Shown are plots of relationships between freshwater movement rate of migrating Atlantic salmon smolts and a) discharge at West Enfield Dam ($m^3 \cdot s^{-1}$), b) photoperiod (hours), c) distance from the ocean (in km), and d) water temperature (°C) in the Penobscot River. Mean predicted movement rate ($km \cdot h^{-1}$) from each covariate is shown by the solid black line and 95% confidence interval is indicated by the dashed gray lines.

Movement rate increased substantially from the most upstream reaches of the catchment (rkm 180) to the most downstream (rkm 45) freshwater reaches (Table 1.3). This resulted in an increase in movement rate of about 500% during the course of downstream migration from headwaters to the estuary (Figure 1.3c). The increase in movement rate was more gradual in the upstream reaches of the river than in downstream reaches, and the most rapid increase in movement rate occurred downstream of rkm 100 (Figure 1.3c).

Movement rates in the main-stem of the Penobscot River increased following the removal of Great Works and Veazie Dams when compared to the previous 7 years (Wilcoxon ranked sum test, $W = 205124$, $P < 0.001$). Median movement rate was 2.8 $km \cdot h^{-1}$ through the impacted reaches between Milford Dam and the head of the Penobscot River Estuary during years 2005–2013, but doubled during 2014 (5.5 $km \cdot h^{-1}$; Figure 1.4a). Movement rate slowed through the Stillwater Branch after the installation of new powerhouses (Wilcoxon ranked sum test, $W = 14088$, $P < 0.001$). Following installation of new powerhouses on the Stillwater Branch in 2013, median movement rate through impacted reaches in 2014 was only 0.1 $km \cdot h^{-1}$, more than an order of magnitude slower than the long-term median of 2.2 $km \cdot h^{-1}$ during the previous seven years (Figure 1.4b). This result was despite reductions in smolt movement rate during 2013 due to ongoing powerhouse construction in the Stillwater Branch (Figure 1.4b).

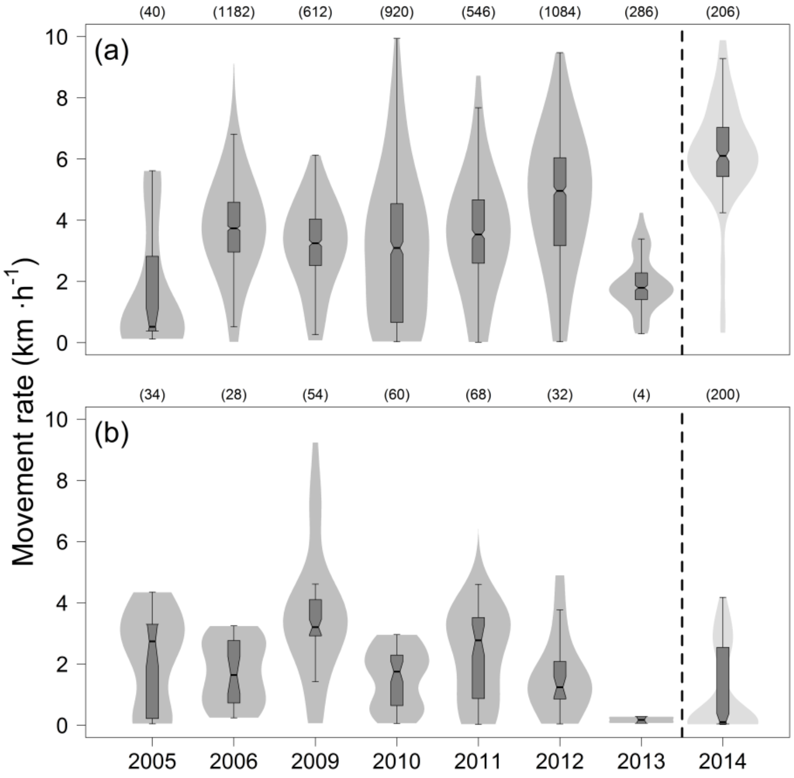
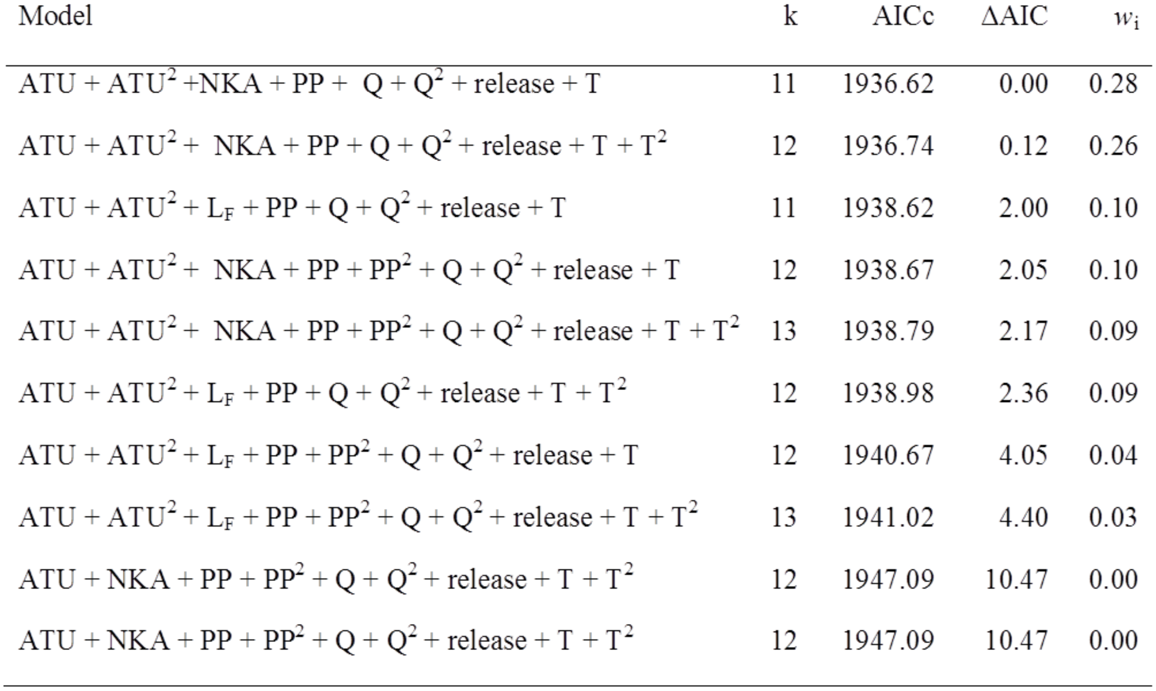


Figure 1.4. Annual changes in movement rates of smolts 2005–2014. Movement rates of Atlantic salmon smolts during each year of this study through a) the river reach between Milford Dam and Penobscot River Estuary, Maine and b) the reach from the upstream end of the Stillwater Branch and the first receiver downstream of the confluence of the Stillwater Branch with the main-stem Penobscot. The vertical line represents the time after which both Veazie and Great Works Dam were removed and new powerhouses had been installed at Stillwater and Orono Dams. Box-ends represent the inner quartile range, whiskers represent 95% CI, and the bold line in the boxes represent median movement rate. The shadows behind boxes are violin plots showing density of observations, and sample size is given above each plot. The slow movement through the Stillwater Branch in 2013 coincided with drawdown for construction, but was used in the 2005–2013 movement rates for Wilcoxon tests.

Time to initiation of migratory behavior by hatchery smolts

Initiation of migration by hatchery smolts was related to physiological development, environmental conditions at release, and the distance from the ocean at which fish were released (Table 1.4). Hatchery smolts that were better prepared for saltwater entry (measured as gill NKA activity) initiated migratory behavior faster than fish that had lower gill NKA activity (Figure 1.5a and Table 5). Over the range of gill NKA activity observed in hatchery smolts (0.9–16.3 μmol ADP·mg protein^{-1}·h^{-1}), time to initiation of migratory behavior was reduced by about 1 day on average (Figure 1.5a).

Table 1.4. Model-selection statistics for the ten best models used to quantify variation in the amount of time to initiate migration by hatchery Atlantic salmon smolts after release into the Penobscot River catchment, Maine USA 2005–2014. Number of parameters estimated in each model is k, $AICc_i$ is the Akaike information criterion for each i^{th} model, $\Delta AICc_i$ is the difference between the AICc of each i^{th} model and the best model in the candidate set, and w_i is the relative probability that each i^{th} model is the best in the candidate set. Explanatory variables are: accumulated thermal units (ATU), discharge (Q), distance between release and relocation (Distance), fork length (L_F), gill NKA activity (NKA), photoperiod (PP), release rkm from ocean (release), and temperature (T).



Model	k	AICc	ΔAIC	w_i
ATU + ATU^2 +NKA + PP + Q + Q^2 + release + T	11	1936.62	0.00	0.28
ATU + ATU^2 + NKA + PP + Q + Q^2 + release + T + T^2	12	1936.74	0.12	0.26
ATU + ATU^2 + L_F + PP + Q + Q^2 + release + T	11	1938.62	2.00	0.10
ATU + ATU^2 + NKA + PP + PP^2 + Q + Q^2 + release + T	12	1938.67	2.05	0.10
ATU + ATU^2 + NKA + PP + PP^2 + Q + Q^2 + release + T + T^2	13	1938.79	2.17	0.09
ATU + ATU^2 + L_F + PP + Q + Q^2 + release + T + T^2	12	1938.98	2.36	0.09
ATU + ATU^2 + L_F + PP + PP^2 + Q + Q^2 + release + T	12	1940.67	4.05	0.04
ATU + ATU^2 + L_F + PP + PP^2 + Q + Q^2 + release + T + T^2	13	1941.02	4.40	0.03
ATU + NKA + PP + PP^2 + Q + Q^2 + release + T + T^2	12	1947.09	10.47	0.00
ATU + NKA + PP + PP^2 + Q + Q^2 + release + T + T^2	12	1947.09	10.47	0.00

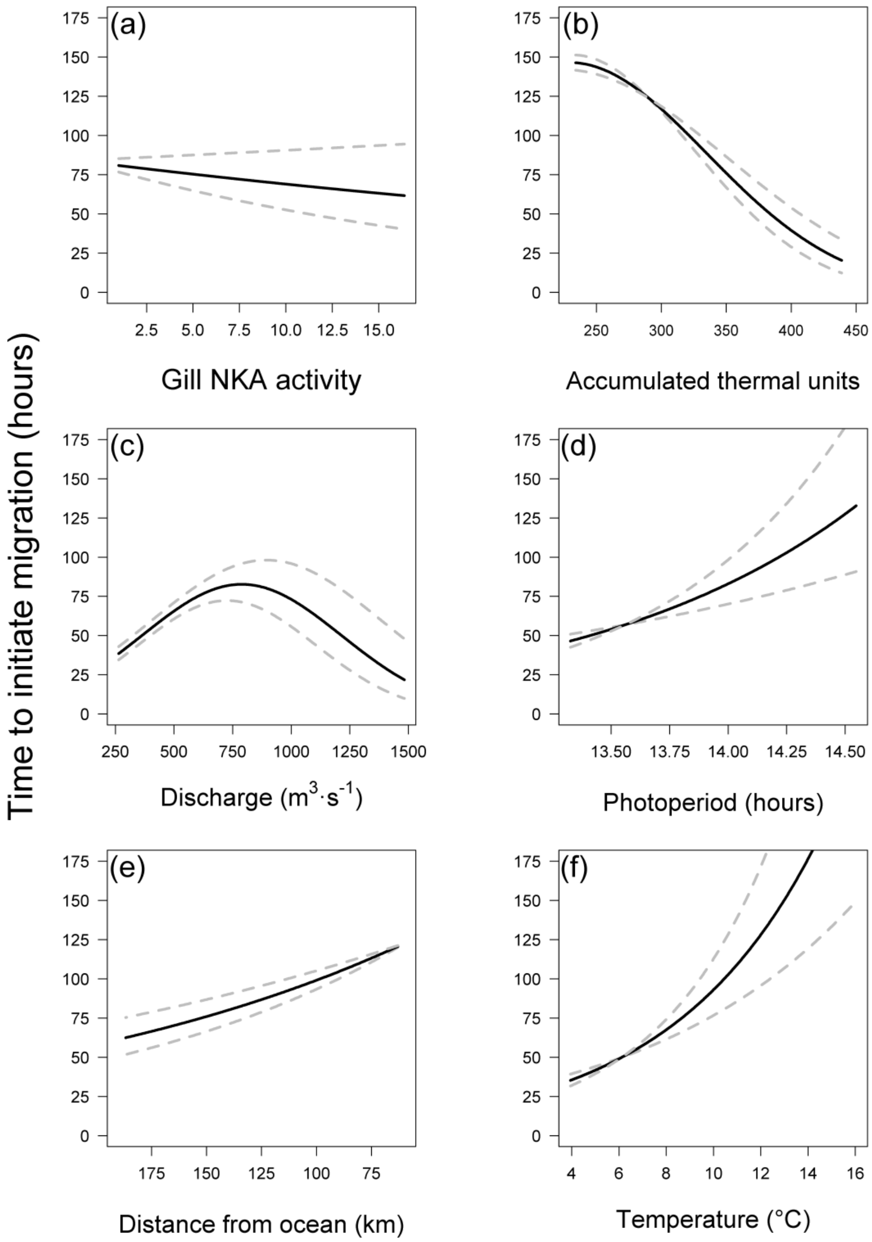
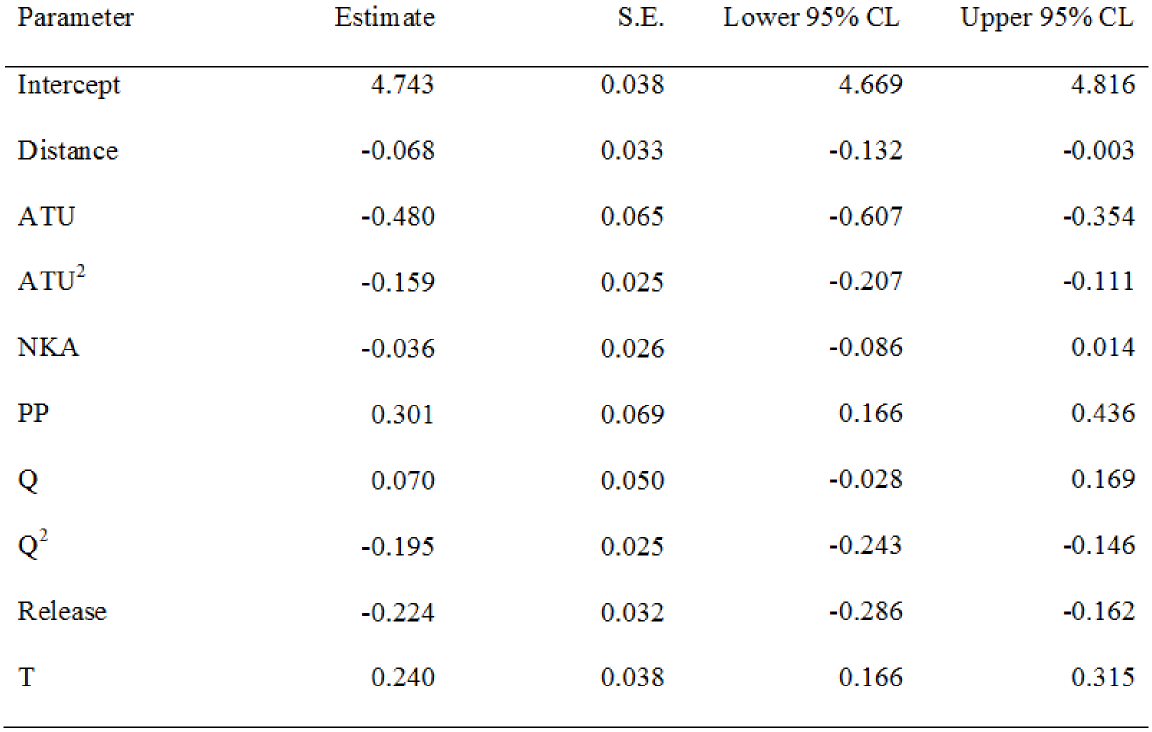


Figure 1.5. Covariate effects on initiation of migration by hatchery-reared smolts 2005–2014. Shown are effects of: a) gill NKA activity, b) accumulated thermal units (ATU), c) discharge at West Enfield Dam, d) photoperiod on date of release, e) distance of release upstream of ocean, and f) water temperature in the Penobscot River at release.

The ATU experienced by all fish in this study was below theoretical thresholds for loss of smolt characteristics (500 ATU: Handeland et al. 2004) and indicated that most smolts were released before or near the peak of smolting (300-400 ATU). Hatchery smolts with greater ATU initiated migration sooner after stocking than smolts with lower ATU (Figure 1.5b and Table 5). Fish with the greatest ATU (439) initiated migration more than 100 hours sooner after stocking than those fish with the lowest ATU (234; Figure 1.5b). This relationship also was non-linear, and the reduction in time to initiate migration was most rapid for fish that experienced 300–400 ATU (Figure 1.5b).

Table 1.5. Mean and 95% confidence limits (CL) of estimated regression coefficients from the best model of time to initiation of migration by hatchery smolts after stocking in the Penobscot River 2005–2014. Explanatory variables are defined as: accumulated thermal units (ATU), discharge (Q), distance between release and relocation (Distance), gill NKA activity (NKA), photoperiod (PP), release rkm from ocean (release), and river temperature (T).



Parameter	Estimate	S.E.	Lower 95% CL	Upper 95% CL
Intercept	4.743	0.038	4.669	4.816
Distance	-0.068	0.033	-0.132	-0.003
ATU	-0.480	0.065	-0.607	-0.354
ATU^2	-0.159	0.025	-0.207	-0.111
NKA	-0.036	0.026	-0.086	0.014
PP	0.301	0.069	0.166	0.436
Q	0.070	0.050	-0.028	0.169
Q^2	-0.195	0.025	-0.243	-0.146
Release	-0.224	0.032	-0.286	-0.162
T	0.240	0.038	0.166	0.315

Initiation of migration occurred later at intermediate discharges (*c*. 750 $m^3 \cdot s^{-1}$), with reduced time to initiation at very low or very high flows (Figure 1.5c and Table 5). Hatchery smolts stocked early in the year initiated migration sooner after release than those fish released later in the year (Figure 1.5d and Table 5), resulting in an increase of about 85 hours (3.5 days) to time of initiation over the range of release dates (12 April–8 May) used in this study (Figure 1.5d). Similarly, initiation of migration took longer with increasing temperatures (Figure 1.5f and Table 1.2), resulting in a change of about 125 hours (5.2 days) across the range of temperatures (3.9–16.1 °C) observed at release date (Figure 1.5f).

Finally, hatchery smolts released at a greater distance from the ocean initiated migration sooner after release than did fish released nearer to the ocean (Figure 1.5e and Table 5). Over the range of release locations used in this study (63–187 km to ocean), the average time for smolts to initiate migratory behavior was about 60 hours (2.5 days) longer for fish released nearest to the ocean compared to fish released furthest upstream (Figure 1.5e).

DISCUSSION

A global theme in Atlantic salmon recovery plans is to maximize the number of smolts entering the marine environment (Hansen et al. 2012). To do this requires managers to maximize survival in freshwater rivers and estuaries. Recent work has demonstrated that estuary survival in the Penobscot River is linked to physiological development of smolts, timing of estuary arrival, and delayed effects of dam passage during freshwater migration (Stich et al., in review). For the first time, I now have a comprehensive picture of smolt migration in a single river system, before and after dam removal, including information about factors affecting freshwater survival (Holbrook et al. 2011; Stich et al. 2014), initiation of freshwater migration and rates of movement (this study), and how experiences in freshwater affect smolt physiology and estuary survival (Stich et al. in review). Additionally, much of the information (e.g., ATU, discharge, photoperiod, and temperature) used to facilitate annual decision making is available in near real time on the Penobscot River. The integration of these data into decision frameworks for recovery has the potential to inform management in a way that could help maximize the number of smolts leaving the Penobscot River through strategic stocking. This will be especially important given continued reliance on hatchery supplementation and the implications of the present study regarding where and when to stock smolts based on developmental status, environmental conditions, and barriers to migration.

Rate of movement and time to initiate migration can influence when fish arrive in the estuary. Timing of arrival in the estuary has important consequences for survival based on physiological development of smolts (Stich et al. in review) and ecological conditions in estuaries and coastal waters (Thorstad et al. 2012). These conditions include ocean temperature (Otero et al. 2014) the presence of predators (Hawkes et al. 2013), or the presence of other migrants (Svenning et al. 2005). In addition to environmental influences on freshwater movement rate and initiation that dictate estuary arrival timing, I was able to uncover important influences of stocking decisions and dams on when and how fast smolts undergo the freshwater phase of migration.

Movement rate

I used two indicators of smolt development to investigate behavior of smolts during the freshwater migration in this study: ATU and gill NKA activity. Previous work has demonstrated that ATU experienced by smolts is a strong predictor of the initiation of migratory behavior in hatchery (Zydlewski et al. 2005) and wild (Sykes et al. 2009) salmon smolts. The freshwater movement rate of smolts was fastest during the seasonal peak of the run (Figure 1.3). Smolts also moved faster at cool temperatures than at warm temperatures (Figure 1.3), and this relationship bottomed out after about 12 °C, toward the end of the smolt run. These results indicate that smolts may move faster during the period at which their physiological development is optimal for ocean entry. In fact, loss of smolt characteristics was previously observed toward the end of the Penobscot River run (McCormick et al. 1999).

When variability in environmental conditions was accounted for, I found that movement rate of smolts was slower in reaches of the Penobscot River that contained head ponds or dams associated with hydropower projects than it was through free-flowing reaches of the river. Reduced movement rates previously have been observed through dams for Atlantic salmon (Holbrook et al. 2011; Norrgård et al. 2013) and for Pacific salmon (Ransom et al. 2008). In this study, the contrast was most pronounced in the movement rates of wild smolts through free-flowing river reaches and river reaches that contained head ponds (Figure 1.2). This effectively increases the ATU experienced by smolts prior to estuary arrival, which could lead to loss of physiological smolt characteristics (McCormick et al. 1999; Handeland et al. 2004). Therefore, delays at dams in the Penobscot River could result in estuary mortality (Stich et al., in review) as a result of mismatches (*sensu* Cushing 1969) between physiological development and timing of arrival in the estuary (Marschall et al. 2011). Given that the effects of dams on movement rates were more pronounced in wild smolts than in hatchery smolts, estuary mortality resultant from delays at dams could be greater in wild smolts than in hatchery smolts.

The removal of two main-stem dams in the Penobscot River during the years of this study allowed us to compare movement rates of Atlantic salmon smolts through standardized reaches of the river before and after the removal of the structures. Similarly, the construction of two powerhouses in the Stillwater Branch afforded during this study afforded us the ability to compare movement rates before and after increases in generating capacity in that part of the river. My results showed a 96% increase in movement rate after the removal of the two dams compared to median movement rates in the same reaches during seven years prior to dam removal (Figure 1.4). I also found that movement rate decreased markedly through a migration route in the lower river (the Stillwater Branch) following the addition of a second powerhouse at each of the dams located therein (Figure 1.4). Although movement rates through impacted reaches have only been collected for a single year after dam removal and hydropower re-allocation, the results are, at minimum, deserving of attention. Furthermore, the methods used in the present study provide both a framework for analysis and a baseline of information moving into the future and will have implications for similar assessments in other systems.

Previous studies of Atlantic salmon smolt migrations in the Penobscot River have demonstrated that acute mortality through reaches containing Great Works and Veazie

Dams was low relative to reaches containing other main-stem dams in the catchment prior to dam removal (Holbrook et al. 2011; Stich et al. 2014). Similarly, acute mortality incurred through passage of Stillwater and Orono Dams was low before construction of new powerhouses (Holbrook et al. 2011; Stich et al. 2014). Subsequent research found, however, that a significant component of smolt mortality in the Penobscot River Estuary was related to delayed effects of dam passage in freshwater, and that the removal of Great Works and Veazie Dams is likely to improve estuary survival of smolts (Stich et al., in review). Similar results have been observed in Pacific salmon smolts in recent years (Budy et al. 2002; Schaller et al. 2012). My results demonstrate that the removal of main-stem dams could also improve smolt survival during estuary passage by reducing delay at these facilities, thus decreasing potential for predation (Blackwell and Juanes 1998) and temperature-related loss of physiological smolt characteristics such as elevated gill NKA activity (McCormick et al. 1999). Counter to this potential positive change in the main-stem is the decrease in movement rates through the Stillwater Branch that could potentially result in increased estuary mortality following construction of new powerhouses. This may be particularly relevant given that survival of smolts in the Penobscot River Estuary also was recently related to gill NKA activity and number of dams passed in freshwater (Stich et al., in review). Additionally, previous work has shown that more fish use the main-stem as a migratory route than the Stillwater Branch (Holbrook et al. 2011; Stich et al. 2014), so a net increase in movement rate through the lower river is expected for smolts. These results highlight the importance of considering effects beyond acute mortality when assessing the effects of main-stem dam removal on anadromous fish migrations.

Initiation

Physiological development and environmental conditions were important predictors of the time to initiate migratory behavior after stocking of hatchery Atlantic salmon smolts. These findings could have important implications for strategic stocking of hatchery smolts in the Penobscot River and elsewhere. Other work has shown that smolts stocked as much as three weeks apart arrive in estuaries at similar times (McCormick et al. 2014). This suggests a tradeoff in the decision by hatchery fish to migrate based on development and/or environmental cues. It is known that physiological development in Atlantic salmon smolts is entrained by seasonal changes in photoperiod (McCormick et al. 1987) and is regulated by changes in temperature (McCormick et al. 2002). Physiological development of Atlantic salmon smolts previously has been observed to coincide with onset of migratory behavior in laboratory experiments (Zydlewski et al. 2014) and field studies (McCormick et al. 2003; McCormick et al. 2013). Influences of local environmental conditions and habitat features also might physically override or inhibit station holding behavior in smolts rather than incite an active ‘choice’ to migrate. Intensity of exposure to environmental changes (such as in photoperiod, temperature or discharge) in upper regions of a watershed might be increased relative to those in the lower river due to the local characteristics of the river, such as depth or channel width. The nature of the mechanism controlling differences among reaches of a river could provide an interesting line of inquiry for future work.

I found that the time to initiation of downstream migration decreased when smolts had greater thermal experience (ATU) during hatchery rearing. This result previously has been observed in laboratory studies of Atlantic salmon (Zydlewski et al. 2005; Zydlewski et al. 2014) and in field studies of Chinook salmon (Sykes et al. 2009). It is important to note that the range of ATU experienced by smolts in the present study was below upper thresholds that have been observed to inhibit migratory behavior in Atlantic salmon smolts (Zydlewski et al. 2005; Zydlewski et al. 2014; McCormick 2009). Loss of physiological smolt characteristics occurs at high temperatures (McCormick et al. 1999), and cessation of migratory behavior also occurs at high temperatures (Zydlewski et al. 2014). In the present study, I observed a gradual increase in the amount of time required for initiation of migratory behavior until about 10 °C, after which the amount of time for initiation increased rapidly with temperature. Interestingly, this is approximately the same temperature at which daily counts of migrating smolts also begin to decline in laboratory studies (Zydlewski et al. 2014). My results suggest that the accumulated thermal units experienced by smolts, in addition to a threshold temperature may be important for initiation of migratory behavior.

Implications for conservation hatcheries

The results of this study have important implications for two decisions that are made annually at conservation hatcheries charged with Atlantic salmon smolt stocking: 1) when to stock fish each spring, and 2) where to stock those fish. Inherent in these decisions is the need to minimize residency time in fresh water and associated risk of loss of smolt characteristics and increased predator exposure. Thus, annual decision making about when and where to stock fish can be greatly facilitated by incorporating knowledge about factors that influence how soon fish begin to migrate, how fast they move, and how well they survive.

Measures of smolt development and environmental conditions were important predictors of when hatchery-stocked smolts initiated migration (Figure 1.5). Given the strong relationships between time to initiation of migration and 1) ATU, and 2) temperature, hatcheries potentially could use both ATU and river temperature as annual indicators for when to stock smolts. My results demonstrated that hatchery stocking of smolts in the Penobscot River has occurred in advance of the number of accumulated thermal units (about 500 ATU) that can connote loss of smolt characteristics, and thus is predicted to increase the number of hatchery smolts that actually migrate to the ocean. Knowledge of smolt development could be used to inform stocking of hatchery smolts as well based on the relationship between physiological development and initiation. However, the relation of initiation to both ATU and river temperature provides a simpler, less expensive planning tool and these factors were quantitatively better predictors of initiation than gill NKA activity. Although developmental indices were not related to movement rate, temperature was (Figure 1.3). Thus, information about temperature could be used to balance time to initiate migratory behavior with movement rate based on when fish are stocked. Daily data on temperature also are already collected multiple times a day throughout the catchment and at salmon hatcheries.

Daily discharge in the Penobscot River can be used as another source of information about when to stock hatchery smolts based on its relation to initiation of migration and movement rate. It took fish longer to initiate migration under intermediate flows (Figure 1.3), but once migrating this was when fish moved most rapidly (Figure 1.5). Thus, information about discharge (like temperature) could be used to optimize when fish are stocked based on relation to initiation of migration and movement rate.

Similar to temperature data, information about discharge in the Penobscot River also is collected several times a day in various locations.

I found two important results that could be used to inform decisions about where to stock smolts: 1) stocking location, and 2) locations of dams and head ponds. Stocking location has the potential to influence movement rate during migration as well as initiation of migratory behavior. Hatchery smolts moved much faster through the lower river than through reaches further upstream (Figure 1.3). Conversely, fish released in the upper watershed initiated migration sooner after stocking than fish released nearer to the ocean (Figure 1.5). As with the influences of temperature and discharge, these results suggest a need to balance stocking location based on differential effects on initiation and movement rate.

Stocking location also is related to the number of dams fish pass as well as which dams fish pass in the Penobscot River. My results clearly demonstrated that movement rate was reduced through reaches containing dams or head ponds associated with dams (Figure 1.2), and that changes to the hydro system have the potential to influence movement rate during migration (Figure 1.4). These results underscore the importance of considering the locations of dams and head ponds with respect to stocking location. Furthermore, my results are average movement rates through a given reach. It is important to note that passage of multiple dams will compound effects on movement rate if only in an additive sense, although multiplicative effects have been observed (Norrgård et al. 2013). This should be important given that dams also are known sites of elevated mortality of Atlantic salmon smolts (Holbrook et al. 2011; Norrgård et al. 2013; Stich et al. 2014), and these structures can have spatially removed effects on migration (Marschall et al. 2011) and even result in delayed mortality during estuary passage (Stich et al. in review). Following the removal of Veazie Dam in 2013, about 20 km of lotic habitat was reclaimed in the lower Penobscot River between the now lowermost dam (Milford) and the estuary. This resulted in the stocking of all smolts in the tail race of Milford Dam in an effort to minimize in-river and estuary mortality incurred through dam passage in the Penobscot River. There are clear tradeoffs between homing/straying rates and smolt-to-spawn survival that must be considered for such a stocking strategy. Research suggests that incidence of straying increases when smolts are released nearer to the ocean, but losses to straying do not offset gains in survival until stocking occurs at the coast (Gunnerød et al. 1988). Similarly, although release within the Penobscot River effects homing to natal streams, the number of fish reaching the lower river (at which point most returning adults currently are trucked to hatcheries) was unaffected by stocking location (Gorsky et al. 2009). Thus, stocking below dams in the Penobscot River likely will increase the number of adults that return for spawning.

CHAPTER 2

SURVIVAL OF ATLANTIC SALMON *SALMO SALAR* SMOLTS THROUGH A HYDROPOWER COMPLEX IN THE LOWER PENOBSCOT RIVER, MAINE USA

INTRODUCTION

Despite extensive efforts to restore Atlantic salmon *Salmo salar* L. 1758, in the USA, total adult returns remain low (NRC, 2004; Saunders et al. 2006). Historically low numbers of *S. salar* led to the federal listing of the species in Downeast Maine, USA waters in 2000 (USFWS and NOAA, 2000), and the Penobscot and Merrymeeting Bay watersheds in 2009 (USFWS and NOAA, 2009). The total number of *S. salar* that returned to all USA waters in 2011 was 4,167 fish (USASAC, 2012). The majority of these fish (75%) returned to the Penobscot River in Maine. As the largest returning run of *S. salar* in the United States, the Penobscot River population has been one focus of a major restoration effort in recent years. The Penobscot River Restoration Project (PRRP) was initiated in 1999 by hydropower companies, conservation groups, state and federal agencies, the Penobscot Indian Nation, and the Penobscot River Restoration Trust (Day 2006). One goal of the PRRP is to balance the restoration of sea-run fisheries (11 species) with hydropower production in the River. Pursuant to this goal, the Penobscot River Restoration Trust (PRRT) purchased the two most seaward dams in the Penobscot for removal (Great Works Dam and Veazie Dam) and a third dam (Howland) for decommissioning and construction of a fish bypass (Day 2006; FERC, 2009).

Dams were cited as the primary cause for the decline of *S. salar* in the Penobscot River (NRC, 2004) and they impede both the upstream migration of adult salmon and the downstream migration of smolts (Holbrook et al. 2009; Holbrook et al. 2011). Although all dams alter the physical environment of riverine ecosystems, some have more-pronounced effects on fish migration than others (Hall et al. 2010). In general, the most seaward dams in heavily impounded systems present comparatively greater disturbances than do upstream dams in terms of system connectivity, total area affected, species richness, or relative abundance of individual species (Vannote et al. 1980; Schlosser 1982; Herbert and Gelwick 2003; Hall et al. 2010). Furthermore, dams are known to cause mortality to downstream-migrating salmonids through migratory delay and entrapment (Keefer et al. 2012) increased predation (Poe et al. 1991), and physical injury (Mathur et al. 2007).

With the removal of Great Works Dam (2012) and Veazie Dam (2013; see Figure 2.1), Milford Dam is now the lowermost barrier to anadromous fish passage in the Penobscot River (Opperman et al. 2011), and is known to be a site of relatively high *S. salar* smolt mortality (Holbrook et al. 2011). Prior studies suggest that the majority of downstream-migrating smolts use the main-stem of the Penobscot as opposed to an alternate migration route around Marsh Island, the Stillwater Branch (Shepard 1991; Holbrook et al. 2011). Therefore, most of these fish must pass Milford Dam before seawater entry, although precise estimates only exist for two years of passage data (Holbrook et al. 2011). These attributes have made Milford Dam a focus for research and assessment regarding anadromous fish passage and survival, as well as for future improvements to upstream and downstream fish passage (Opperman et al. 2011). In addition, two operational dams (Stillwater and Orono Dams) in the Stillwater Branch (Figure 2.1) currently are undergoing construction of new facilities that will increase power generation and head-pond height through that route (Day 2006; Opperman et al. 2011). The regulatory requirement for downstream passage of *S. salar* smolts is a survival of 96% at each of these dams (NMFS, 2012). A baseline of knowledge about fish passage through this complex of dams (hereafter the "Marsh Island hydropower complex") prior to the implementation of restoration efforts will be necessary for assessment of future improvements of fish passage in the lower river, and for determining the combined effects of dam removal and operational changes on the survival of federally endangered *S. salar* smolts during seaward migration in the lower river.

The goals of this study were 1) to estimate proportional use of migratory routes and the apparent survival rates for *S. salar* smolts through the Marsh Island hydropower complex using a combination of acoustic- and radio-telemetry data, and 2) to determine the effects of in-river discharge and fish characteristics (fork length [L_F], mass, and rearing origin) on path choice and survival through this section of the river. To achieve these goals, the first objective of this study was to estimate proportional use of two migratory routes (Penobscot and Stillwater) by *S. salar* smolts and to estimate path-specific survival using six years of acoustic telemetry data. The second objective of the study was to estimate path-specific survival through the powerhouse and spillway of Milford Dam using radio-telemetry data from 2010 and 2012. Finally, data from both acoustic and radio telemetry are used to characterize variability in selection of migratory route and survival in relation to river discharge. The results of this study will be useful for making decisions about management of downstream fish passage through the complex of dams in the lower Penobscot River and assessing the overall effect of the Penobscot River Restoration Project on downstream passage of *S. salar* smolts.

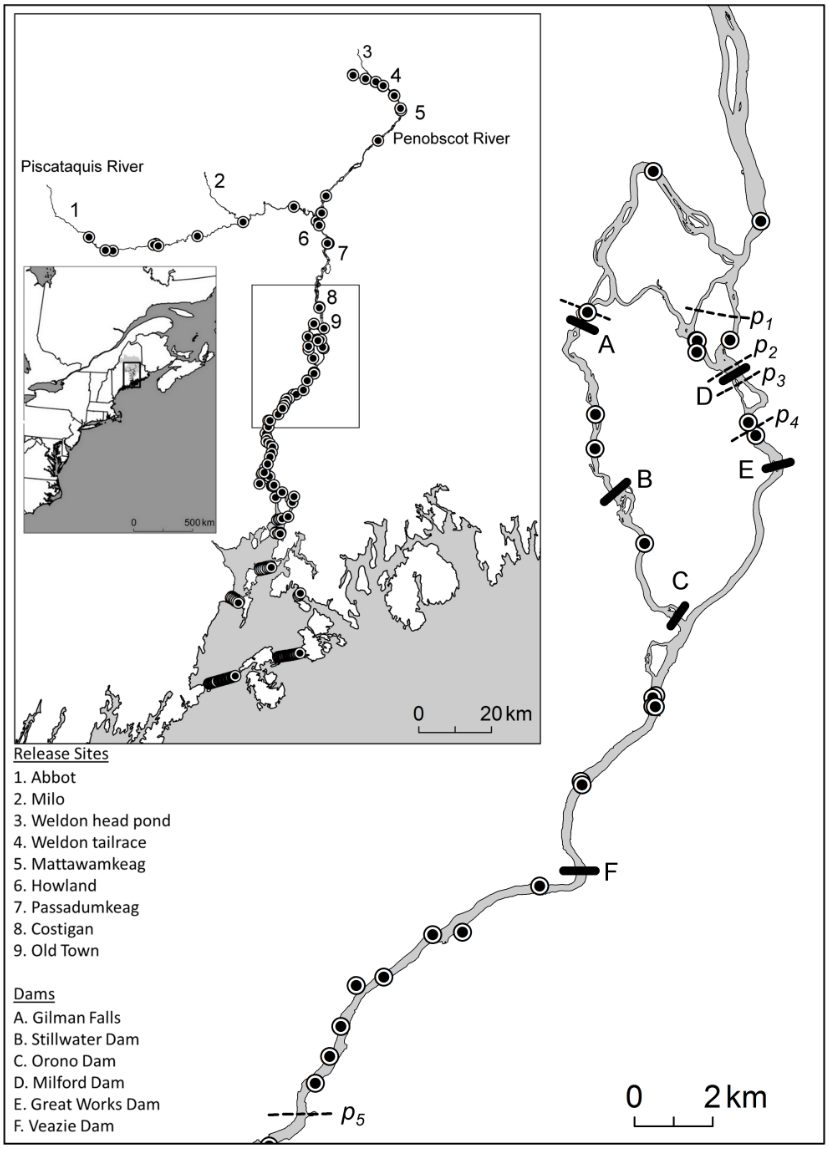


Figure 2.1. Map of Penobscot River catchment and Marsh Island. Shown are location in North America in the small inset, coverage of the acoustic telemetry network and release sites for tagged *S. salar* smolts (acoustic and radio) in the large inset, and locations of dams and coverage of radio-receiver network in the large map. Release sites for acoustic- and radio-telemetry studies are numbered, and the dams in the lower Penobscot River are represented by solid lines and are lettered. Circles represent locations of acoustic receivers. Dashed lines represent locations of one or more radio receivers used in the radio-telemetry study of passage at Milford Dam, each with multiple antennas. The detection occasions used in radio-telemetry models are indicated by the letter *p* with subscripts corresponding to detection occasions shown in the radio schematic in Figure 2.3.

METHODS

Main-stem dams

Milford Dam is located between the City of Old Town and the Town of Milford at river kilometer (RKM) 61on the main-stem of the Penobscot River in Maine, USA (Figure 2.1). The current site of the hydropower project is the natural fall line in the Penobscot River (Opperman et al. 2011). Milford Dam is approximately 6.1 m high, and spans 353 m across the river. The powerhouse at the project, located on the eastern shore of the river, contains six generating turbines, with a maximum authorized generation of about 9 megawatts (MW). Current fish passage facilities at the site include an eel ladder and a Denil fish way for upstream fish passage, as well as a log sluice between the powerhouse and the spillway for downstream fish passage (FERC, 2009). Construction of a new fish elevator for upstream passage is ongoing and is anticipated to be completed in spring 2014. Discharge into the Stillwater Branch is controlled primarily through increases and decreases in head pond level at Milford Dam up to about 430 m^3s^{-1}, at which point the facility can no longer control spill to the main-stem (FERC 2004). Currently, the dam redirects about 30% of total discharge in the lower Penobscot into the Stillwater Branch and under legal agreements involved with the Penobscot River Restoration Project this can be increased to 40% of total river discharge once new powerhouses in the Stillwater Branch are on-line (FERC, 2004).

Great Works Dam (Figure 2.1) was removed from the main-stem of the Penobscot River during summer 2012; just after the final year of this study. The former Great Works project was located at RKM 59, was 6.1 m high and 331 m across (FERC, 2009). The powerhouse had 11 horizontal turbines and generating capacity of 7.9 MW. Fish passage facilities at the former Great Works Dam included two Denil fish ways for upstream passage.

Veazie Dam (Figure 2.1) was formerly located at RKM 45 in the main-stem of the Penobscot River, and was removed in summer 2013; a year after this study. The project consisted of two powerhouses, one with 15 turbines and another with 3 turbines, with a maximum generating capacity of 8.4 MW (FERC, 2009). The dam was 10 m high and 257 m across, with a slot fish way for upstream passage. Operations at both Great Works and Veazie Dams were subject to periodic shutdowns for regulatory purposes, which had the potential to affect smolt survival during the final two years of the study. Although records for turbine shut downs were not publically available to correlate with survival estimates for any of the dams, the Penobscot River Restoration Trust was required to shut down turbines during the smolt migration period per their permitting conditions during 2011 and 2012.

Stillwater Branch dams

Two operational hydropower dams will remain in the 16.9-km Stillwater Branch after the Penobscot River Restoration Project. Stillwater Dam (Figure 2.1) is located at RKM 60 (from the mouth of the Penobscot River) on the Stillwater Branch, is 6.7 m high, and 524 m across (NMFS, 2012). The original powerhouse contains four horizontal turbines and has a generating capacity of about 2.0 MW. The additional powerhouse being constructed at the Stillwater project will have 3 vertical turbines and will add 2.2 MW to the total generating capacity of the Stillwater facility, more than doubling the capacity for power generation. Current fish passage facilities at Stillwater Dam include a downstream bypass discharging into the tail race and two eel-passage facilities. No further upstream passage will be constructed at Stillwater under the license amendment for this facility, although a new downstream bypass facility will replace the existing structure (NMFS, 2012).

Orono Dam is located in the Town of Orono at RKM 55 (from the mouth of the Penobscot River) on the Stillwater Branch, at the confluence of the Stillwater with the main-stem of the Penobscot River (Figure 2.1). The dam is 7.6 m high, and is 358 m across, with a powerhouse containing four turbines that have a total generating capacity of 2.3 MW (NMFS, 2012). The new powerhouse being constructed at Orono Dam will add three vertical turbines that have total generating capacity of 3.7 MW, more than doubling the total capacity of the Orono project. Current fish passage facilities at the Orono project include a downstream fish way and an upstream eel-passage facility. The upgrades to the project will include construction of an additional downstream bypass, as well as a fish trap used to catch upstream-migrating fishes for transport to the main-stem Penobscot (NMFS, 2012).

Acoustic receiver array

Prior to the start of the *S. salar* smolt run during each year of this study, stationary acoustic receivers (VR2 and VR2-W; Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) were deployed in the Penobscot River cooperatively by the University of Maine, in cooperation with U.S. Geological Survey (USGS) Maine Cooperative Fish and Wildlife Research Unit, and the National Oceanic and Atmospheric Administration (NOAA). All receivers contained omnidirectional hydrophones that scanned continuously at 69 kHz. The number and type of receivers deployed in the Penobscot River watershed varied slightly between years. The number of receivers deployed in the watershed increased through time as new units were purchased and as new release sites were added. The acoustic receiver array used in 2005 and 2006 was described in Holbrook et al. (2011). Up to 198 acoustic receivers were deployed in a given year, providing detection coverage from the headwaters of the East Branch Penobscot and Piscataquis Rivers through outer Penobscot Bay for years 2009 through 2012 (Figure 2.1). Despite differences in arrays between years, the configuration and proximity of acoustic receivers in and around the Marsh Island hydropower complex were virtually identical through all years of the acoustic telemetry study. Acoustic receivers deployed in the Penobscot River and in the estuary were moored to reinforced cement anchors on the river bottom. Acoustic receivers deployed in the Penobscot Bay were tethered approximately 10 m below the surface of the water. Multiple receivers were deployed where the width of the river exceeded the detection range of acoustic receivers or where obstructions (e.g. islands) prevented complete coverage with a single deployment, and detections for all receivers at such locations were pooled as single encounter events for survival analyses.

Acoustic tagging and releases

From 2005 through 2012 1,669 *S. salar* smolts, either wild-reared or from the U.S. Fish and Wildlife Service (USFWS) Green Lake National Fish Hatchery, were acoustically tagged and released by the University of Maine and USGS for studies of in-river movements and survival during downstream smolt migration through the Penobscot

River (Table 2.1). Acoustic tagging methods were described in detail by Holbrook et al. (2011) and identical procedures were used in all years from 2005 through 2012. Smolts were anesthitized using a 100 $mg \cdot L^{-1}$ solution of MS-222, L_F (mm) and mass (g) were measured before fish were placed ventral side up in a surgical saddle. A small (1-cm) incision was made slightly offset from the ventral line and about 1-cm posterior to the pelvic fin girdle. An acoustic tag was inserted intraperitoneal and the incision was closed with two simple, interrupted knots using 4-0 absorbable vicryl sutures (Ethicon, Somerville, New Jersey, USA). Model V7-2L acoustic tags (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) were used in 2005, as well as for wild-origin fish tagged in 2011. Expected battery life of V7-2L tags was 80 days for tags used during 2005, and 69 days for tags used during 2011. In all other years, acoustic transmitters used were model V9-6L (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) with expected battery life of 80–82 days. Total time for each surgery was less than two minutes. *Salmo salar* smolts of wild and hatchery origin were released at up to four different sites in a single year, although the numbers of fish and release sites varied between years (Table 2.1).

Table 2.1. Descriptive statistics for acoustically tagged smolts used to assess survival through the Marsh Island hydropower complex. Number, origin, tag type, mean fork length (L_F, mm) and release site of *S. salar* tagged and released within the Penobscot River drainage each year of study from 2005- 2012. Numbers in parentheses to the right of L_F measurements represent the standard deviation of L_F (mm) in each release group.

Year	Release site	Tag Type	Origin	Number Released	L_F(SD)
2005	Howland	Acoustic	Hatchery	150	189 (11)
	Mattawamkeag	Acoustic	Hatchery	40	185 (12)
	Milo	Acoustic	Hatchery	85	191 (11)
	Weldon tailrace	Acoustic	Wild	60	178 (18)
2006	Milo	Acoustic	Hatchery	72	196 (11)
	Weldon tailrace	Acoustic	Hatchery	146	198 (15)
	Weldon tailrace	Acoustic	Wild	73	190 (11)
2009	Milo	Acoustic	Hatchery	100	180 (8)
	Passadumkeag	Acoustic	Hatchery	100	180 (9)
2010	Abbott	Acoustic	Wild	75	169 (8)
	Milo	Acoustic	Hatchery	100	189 (11)
	Passadumkeag	Acoustic	Hatchery	100	186 (11)
	Weldon head pond	Acoustic	Wild	74	180 (13)
	Old Town	Radio	Hatchery	58	198 (12)
2011	Abbott	Acoustic	Wild	75	146 (8)
	Milo	Acoustic	Hatchery	100	191 (13)
	Passadumkeag	Acoustic	Hatchery	100	197 (32)
	Weldon head pond	Acoustic	Wild	60	162 (19)
2012	Abbott	Acoustic	Hatchery	74	199 (11)
	Weldon head pond	Acoustic	Hatchery	85	199 (11)
	Costigan	Radio	Hatchery	130	201 (15)
Grand total	All		All	1857	-

Radio receiver array

A total of 13 data-logging radio receivers (models SRX400 and SRXDL; Lotek Wireless, Newmarket, Ontario, Canada) were used to detect radio-tagged *S. salar* smolts during migration through Milford Dam in 2010 and 2012 (Figure 2.1). Individually coded radio transmitters spanning three frequencies were used in order to minimize tag collisions while allowing for an acceptable cycling time on radio receivers. At least two frequencies were used in each release group. Radio receivers were set to scan each of three frequencies for 3 seconds on each antenna. This resulted in total cycling times that ranged from 9 seconds in receivers with one antenna to 36 seconds in receivers with four antennas. The radio-receiver array differed slightly between 2010 and 2012 based on smolt release locations. In 2012, smolts were released further upstream than in 2010 to increase detection probabilities; therefore, an extra pair of radio receivers was deployed between the release location and Milford Dam in 2012. The location at which the additional pair of receivers was deployed in 2012 corresponded with the release locations that were used in 2010 (Old Town and p_1; Figure 2.1). Multiple receivers, each with multiple antennas, were deployed at each detection site above and below Milford dam (a total of 5 receivers above the dam [p_2 in Figure 2.1 and Figure 2.3] and two below[p_3 in Figure 2.1 and Figure 2.3]) to ensure that path choice could be determined. Two receivers were deployed just downstream of the Milford tailrace, each with one antenna (p_4 in Figure 2.1 and Figure 2.3). Finally, two receivers were deployed at a private residence downstream in the estuary (p_5 in Figure 2.1) to allow for estimation of survival in the tailrace of Milford Dam. Receivers were pooled as a single encounter location where multiple receivers or antennas were used to obtain adequate coverage across the width of the river. Because the release site used in 2012 resulted in the possibility of fish moving into the Stillwater Branch, and out of the main-stem of the river, a radio receiver was placed below the upper-most dam on the Stillwater Branch (Gilman Falls; Figure 2.1) so that these fish ($n = 1$) could be excluded from analysis of passage at Milford Dam.

Radio tagging and release

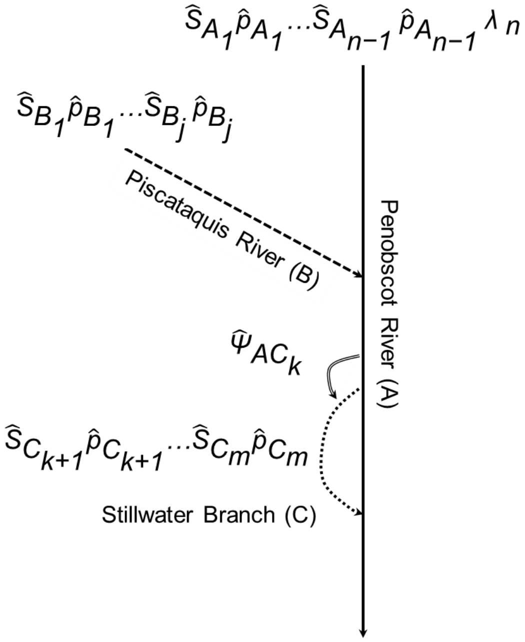
Salmo salar smolts were radio tagged with NTC-3-2 coded nano tags weighing approximately 0.5 g with 24-cm trailing-whip antenna, 2-second burst rate, and 31-d battery life (Lotek Wireless, Newmarket, Ontario, Canada) using a modification of the shielded needle method (Ross and Kleiner 1982). Fish were anesthitized using a 100 $mg{\cdot}L^{-1}$ solution of MS-222, and L_F (mm) and mass (g) were measured prior to surgery (Table 2.1). Smolts were placed ventral-side up in a v-shaped saddle, and a small (0.5-cm) incision was made offset from the ventral line and about 1-cm posterior to the pectoral fin girdle. Radio tags were tested and the antenna inserted into a 20-gage, deflected-tip septum needle. The needle was inserted through the ventral incision and passed from inside the peritoneal cavity through the body wall posterior and dorsal to the pelvic fin. The needle was removed, leaving only the antenna in the opening through the body wall. The radio tag was gently pushed into the peritoneal cavity and the ventral incision was closed with a single interrupted knot using 4-0 absorbable vicryl sutures (Ethicon, Somerville, NJ, USA). Mean time for radio surgeries was approximately 1 minute.

In 2010, 58 *S. salar* smolts from the USFWS Green Lake National Fish Hatchery were radio tagged to assess passage and survival through Milford Dam. An additional 25 fish were tagged with dummy tags of identical dimensions and held in the Green Lake National Fish Hatchery for three weeks to assess tag loss and tagging-related mortality. *Salmo salar* smolts were released on 15 May 2010 about 1 km upstream of Milford Dam (Old Town; Table 2.1, Figure 2.1). Half of the fish were stocked from the east bank of the river, and half were stocked from the west bank. In 2012, 130 hatchery-reared *S. salar* smolts were released in four groups over the course of one week in order to reduce the risk of not detecting smolts at individual receivers in the array. Smolts were released at a public boat launch on the east bank, approximately 8 km upstream of Milford Dam (Costigan; Figure 2.1) during 22 April 2012 through 28 April 2012 (Table 2.1).

Movement and survival through Marsh Island hydropower complex

Multi-state (MS) Cormack-Jolly-Seber mark-recapture survival models were developed and analyzed in program MARK (White and Burnham 1999) to estimate the proportional use of the Stillwater Branch and main-stem Penobscot River, as well as path-specific survival rates through each route using acoustic telemetry data. The logit-link function was used to model all parameters in acoustic multistate models. While I use the term survival throughout the present study for simplicity, these estimates reflect only apparent survival and not true survival of *S. salar* smolts as the only data used in these models were detections of fish at each receiver location, and information about whether fish were alive or dead was not available. Although only those estimates of survival relevant to the Marsh Island hydropower complex are reported in the present study, these estimates are based on MS models that incorporated detections at acoustic receivers through the entire acoustic array. These “whole-system” survival models were constructed separately for each year due to differences in the acoustic-receiver array between years at locations outside of the Marsh Island hydropower complex. Due to differences in migratory histories of hatchery and wild fish resultant from release locations in the Penobscot and Piscataquis Rivers, migratory route and survival also were modelled separately for hatchery and wild fish within years.

Three states were used in the development of acoustic-MS survival models in each year: State-1) the main-stem of the Penobscot River (A) from the upper-most interval to Penobscot Bay, State-2) the Piscataquis River (B), and State-3) the Stillwater Branch (C) as an alternative migratory route through the Marsh Island hydropower complex (Figure 2.2).



Year	Origin	Variable occasions			
		j	*k*	*m*	*n*
2005	All	3	7	10	16
2006	All	3	7	10	16
2009	Hatchery	4	8	10	32
2010	Hatchery	5	6	7	25
2010	Wild	9	11	14	31
2011	Hatchery	4	8	10	30
2011	Wild	10	13	17	19
2012	Hatchery	10	13	15	16

Figure 2.2. Diagram of the parameters estimated each year in acoustic multi-state models of *S. salar* smolt survival in the Penobscot River 2005—2012. The schematic (left) illustrates the three states used to model survival in each year (A = main-stem Penobscot, B = Piscataquis River, and C =Stillwater Branch), and includes variables (*j*,*k*,*m*,*n*) that indicate differences in the number of parameters estimated in each year. The Table 2.1n the diagram contains the value of each variable in each model each year, and can be used to reconstruct annual survival models. As an example, in 2005: survival ($\widehat{\boldsymbol{S}}$) and detection probability ($\hat{\boldsymbol{p}}$) were estimated for intervals 1-16 (*n*) in state A (main-stem Penobscot), survival and detection probability were estimated for intervals 1-3 (*j*) in state B (Piscataquis), the state-transition probability for movement into the Stillwater ($\widehat{\boldsymbol{\Psi}}_{AC}$) was estimated in interval 7 (*k*), and survival and detection probabilities were estimated in the Stillwater (state C) during intervals 8 (*k* + 1) through 10 (*m*).

The parameters estimated in the acoustic-MS survival models varied each year (Figure 2.2). Survival ($\widehat{S}$) and detection probability ($\widehat{p}$) were estimated in the main-stem of the Penobscot from release ($\widehat{S}_{A_1}\widehat{p}_{A_1}$) through interval *n* - 1 ($\widehat{S}_{A_{n-1}}\widehat{p}_{A_{n-1}}$) each year, and the joint probability of detection and survival (λ) was estimated during interval *n* each year. Survival and detection probability were estimated in the Piscataquis River each year from release ($\widehat{S}_{B_1}\widehat{p}_{B_1}$) to interval *j* ($\widehat{S}_{B_j}\widehat{p}_{B_j}$). The probability of moving into the main-stem of the Penobscot River from the Piscataquis River ($\widehat{\Psi}_{BA_j}$) given survival in state B during interval *j* was fixed to 1.00 during interval *j* each year. The proportion of fish that migrated through the Stillwater Branch each year was estimated as the state-transition probability for movement from the Penobscot River into the Stillwater Branch ($\widehat{\Psi}_{AC_k}$) during interval *k* and the probability of remaining in the main-stem ($\widehat{\Psi}_{AA_k}$) was $1 - \widehat{\Psi}_{AC_k}$. Survival and detection probabilities in the Stillwater Branch were estimated each year from interval *k* +1 ($\widehat{S}_{C_{k+1}}\widehat{p}_{C_{k+1}}$) through interval *m* ($\widehat{S}_{C_m}\widehat{p}_{C_m}$), and the state-transition probability for movement from the Stillwater Branch into the main-stem ($\widehat{\Psi}_{CA_m}$) given survival during interval *m* was fixed to 1.00. All parameters not shown (Figure 2.2) or described above were fixed to zero during model estimation.

Path-specific survival through Milford Dam using radio telemetry

Multistate survival models were developed and analyzed in program MARK (White and Burnham 1999) to estimate survival and determine proportional passage through two potential paths through Milford Dam by radio-tagged *S. salar* smolts (the spillway or powerhouse). Due to lack of sample sizes required for estimation of a third transition probability, fish passage through a log sluice on the face of the dam was included in the estimate of passage via the spillway. The radio-MS models used for assessment of smolt passage through Milford Dam consisted of six detection events (Figure 2.3). The logit link function was used to model all parameters in all models. In both years of the radio-telemetry study, a downstream, estuarine radio-receiver station was established such that *S. salar* smolt survival could be estimated through all intervals of interest (Figure 2.3). Detections at each receiver location were used to construct individual encounter histories from release to the Penobscot Estuary for all radio-tagged fish. Passage path through Milford Dam (spillway or powerhouse) was discriminated by fine-tuning radio receivers at various locations at the dam and the probability of using the spillway ($\hat{\Psi}_{2AA}$) or powerhouse ($\hat{\Psi}_{2AB}$) for passage through the dam was estimated (Figure 2.3). Each of the two potential passage paths was used as a state in the individual encounter histories, and state-specific survival ($\hat{S}$) and detection probability ($\hat{p}$) were estimated for passage through the spillway ($\hat{S}_{3A}, \hat{p}_{2A}$; Figure 2.3) and the powerhouse ($\hat{S}_{3B}, \hat{p}_{2B}$; Figure 2.3).

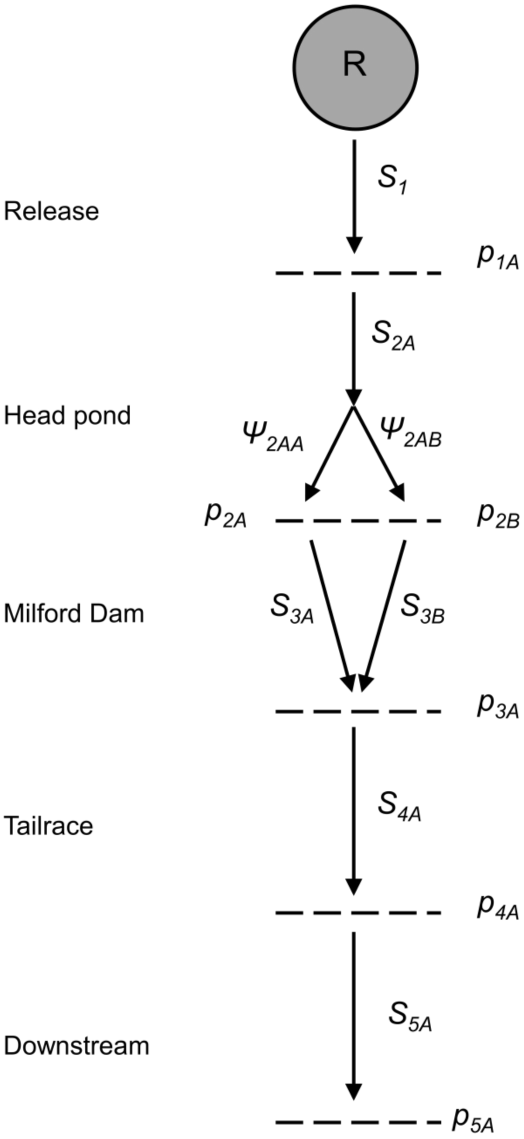


Figure 2.3. Schematic representation of multi-state survival model used to estimate path choice and path-specific survival of *S. salar* smolts through Milford Dam using radio-telemetry data. The *R* represents release, $\hat{\boldsymbol{p}}$ indicates detection probability at each occasion after release, $\hat{\boldsymbol{S}}$ is survival in each reach and in each state ($\hat{\boldsymbol{S}}_A$ = main-stem/spillway survival, and $\hat{\boldsymbol{S}}_B$ = powerhouse survival). Estimates of detection probability ($\hat{\boldsymbol{p}}_{5A}$) and survival $\hat{\boldsymbol{S}}_{5A}$) are confounded during the final interval of the radio-telemetry models, and so λ is the joint probability of survival and detection estimated in the final reach. A description of each interval used in radio-telemetry models is given to the left of the schematic.

In 2010, the release of radio-tagged smolts coincided with draw-down of the Milford head pond for installation of flashboards at downstream facilities (this was serendipitous, not a study design detail). Given the timing and degree of the drawdown, in combination with the narrow timeframe of passage by *S. salar* smolts, all of the radio-tagged fish that successfully passed Milford Dam in 2010 did so by way of the powerhouse; therefore, all state-transition probabilities were fixed to zero and are not included in the parameters reported in model results for 2010. *Salmo salar* smolt releases in 2012 occurred during variable discharge conditions; however, water was being spilled over the top of the dam during the majority of the 2012 smolt season. Because discharge conditions and intervals (i.e., distance between receivers) used in survival estimation varied between 2010 and 2012, survival was modelled separately for each year of the radio-telemetry study.

Model fit and selection

To assess fit of acoustic- and radio-MS survival models, an over-dispersion parameter, $\hat{c}$, was estimated for the saturated model(s) each year using program U-CARE (Choquet et al. 2009). In all cases, models were structured such that $\hat{c}$ was < 2 and adequate model fit was achieved prior to analysis of competing hypotheses. After assessing the fit of each of the full models, candidate models of *S. salar* smolt survival were chosen, *a priori*, to determine the (hypothesized) relative importance of variability in survival ($\widehat{S}$) and detection probabilities ($\widehat{p}$) among river reaches in models for each year. Probability of using each passage path (spillway or powerhouse) in radio-telemetry models and each migratory route (Stillwater or Penobscot) in acoustic models was estimated as an interval-specific state-transition probability ($\hat{\Psi}$) in each model.

An information-theoretic approach to model selection, based on corrected Akaike information criterion ($\mathrm{AIC_c}$; Burnham and Anderson 2002), was used to determine whether survival varied between reaches of the river by comparing models with constant survival between reaches to models with reach-specific survival rates. The relative support for candidate models was evaluated as the difference in $\mathrm{AIC_c}$ between the best model and each i^{th} model (Δ_i), and the relative probability of each model being the best was represented using $\mathrm{AIC_c}$ weights (w_i; Burnham and Anderson 2002). Models for which $\Delta_i \leq 2.0$ were considered to have similar support to the best model in each candidate model set (Burnham and Anderson 2002).

Effect of discharge on movement and survival around Marsh Island

Simple linear regression was used to obtain a characterization of the relationship between discharge and estimated mean survival through the reach of the main-stem Penobscot River containing Milford Dam across all years using survival estimates from both radio- and acoustic-telemetry studies. Due to constraints on the possible values of survival [0, 1], survival estimates were logit-transformed prior to analysis. I determined that the variances of individual survival estimates did not influence the results of the regression when the results were compared to a weighted least squares regression. Therefore, for sake of simplicity, the results of ordinary least squares regression are presented graphically on the real scale of the response variable with tick marks spaced on the logit scale. Mean daily discharge values (m^3s^{-1}) from the USGS gage upstream of

Milford at West Enfield Dam were used to characterize mean discharge during the window of time that smolts passed through Milford Dam each year.

Generalized linear mixed models (GLMM; Zuur et al. 2007) were used to assess the relationship between discharge and individual migration route (Stillwater Branch or main-stem Penobscot River), with year as a random effect on the intercept in all models using the ‘lme4’ package (Bates et al. 2013) in R (Version 3.0.1, R Development Core Team 2013). Only those fish (n=759) for which passage path was known were used for the analysis, and the results of the GLMM were compared to predictions from multistate models to assure that predictions were not biased due to the exclusion of detection probability for fish that were omitted due to unknown passage path. The model used a logit-link function and the response was binary (1 = Stillwater, 0 = main-stem Penobscot). Discharge experienced by individual fish prior to choosing a migratory route was characterized using mean of daily discharges at West Enfield Dam from the time a fish was first located 0.5 km upstream of West Enfield Dam until the time that it was first detected at Milford Dam or in the Stillwater Branch (mean travel time = 4 days for smolts). West Enfield Dam is located approximately 40 km upstream of the Marsh Island hydropower complex, on the main-stem of the Penobscot, immediately upstream from the mouth of the Piscataquis River (Figure 2.1). Although I recognize that proportional distribution of discharge between the Stillwater Branch and main-stem Penobscot around Marsh Island would have provided an ideal measurement of discharge for this analysis, these data were not available and discharge at West Enfield Dam offered the best available information about discharge carrying fish to the Marsh Island hydropower complex. The ability of the hydropower company to control the distribution of flows at

Milford Dam is lost (due to maximum pond height) at discharges of approximately 430 m^3s^{-1}. At discharges less than 430 m^3s^{-1}, operations at Milford Dam maintain proportional flow of approximately 30% of total river discharge to the Stillwater Branch (FERC, 2004). It was, therefore, hypothesized that total discharge through the lower river, as measured at West Enfield Dam would provide a biologically meaningful predictor of the probability that smolts used the Stillwater Branch that could be indirectly related to hydropower operations in the Marsh Island complex and also would provide comparisons in the future following operational changes. To test the null hypothesis that choice of migratory route was not related to discharge, models of migratory route that did or did not include discharge were compared using AIC_c (described above; Burnham and Anderson 2002). Fish characteristics (rearing history and L_F) that had the potential to influence choice of migratory route were also investigated using model selection. Approximation of the overdispersion parameter $\hat{c}$ for the most parameterized model in the candidate set indicated that the models were not overdispersed ($\hat{c} \leq 1$); therefore, model selection was not adjusted.

RESULTS

Path choice around Marsh Island

In all years and for all release groups, the fully reach-dependent parameterizations for survival and detection probability in MS models based on acoustic-telemetry data were the most parsimonious, and therefore model selection for these models are not shown. The mean (95% CI) annual probabilities of using the Stillwater Branch ($\hat{\Psi}_{AC_j}$from acoustic MS survival models) ranged from 0.04 (0.01 – 0.11) to 0.25 (0.13 – 0.45), with an overall mean of about 0.12 across years (Figure 2.4). Individual-based GLMMs of path choice indicated that of the factors hypothesized to affect proportional use of the Stillwater Branch, discharge at West Enfield Dam had the greatest influence; it was the only covariate included in all models that had a meaningful amount of support in the candidate model set, and it was the only covariate included in the best model (Table 2.2). Use of the Stillwater Branch increased with discharge within the observed range of discharges during the smolt window during 2005 through 2012 (Figure 2.5). Based on observed flows over the six years of this study, the overall mean (95% CI) probability of using the Stillwater Branch in any given year, according to the GLMM used to model individual migration route, was 0.12 (0.06 – 0.25) conditional on flow (Table 2.3). This conditional mean is identical to the mean probability of using the Stillwater estimated in multistate models.

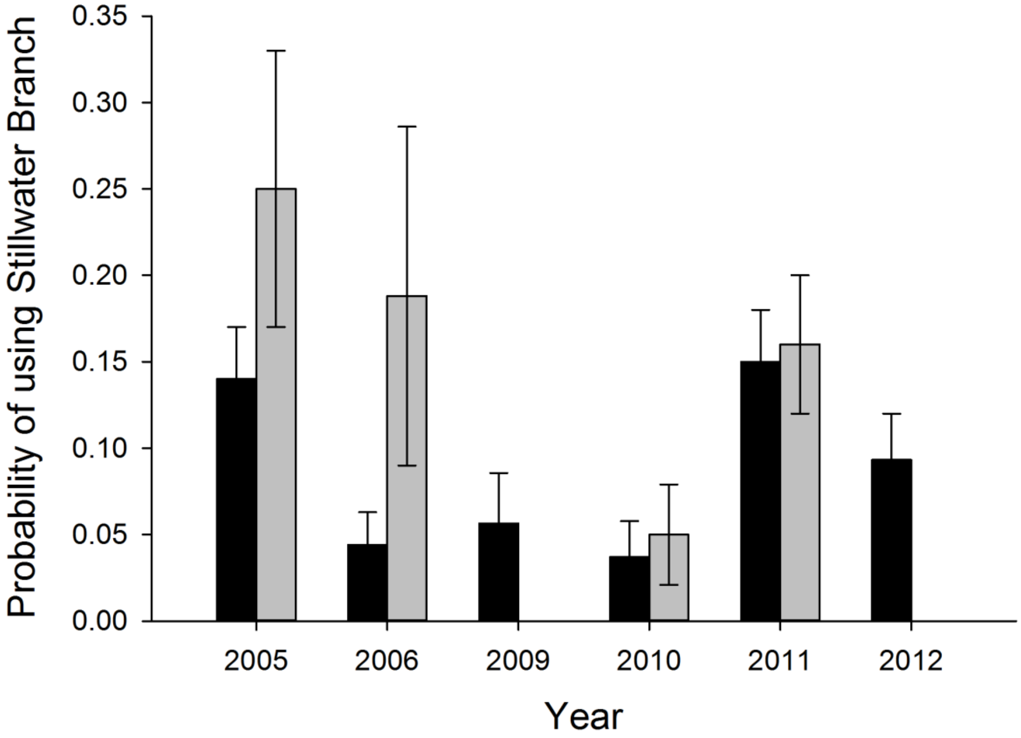


Figure 2.4. Annual probability of using Stillwater Branch. Shown are annual estimates of the mean ± S.E. probability of *S. salar* smolt migration through the Stillwater Branch estimated using acoustic multi-state models of smolt movement and survival in the Penobscot River during six years from 2005 through 2012 for wild (gray) and hatchery (black) smolts.

Table 2.2. Model selection for probability of using the Stillwater Branch. Model selection statistics for GLMMs used to characterize relationships between the probability of *S. salar* smolts using the Stillwater branch for migration and several factors of interest, including rearing history (ORIGIN: hatchery or wild), fork length (FL), and discharge measured at West Enfield Dam (DISCHARGE). All models included a random effect of year on the intercept (not shown in table), which accounts for one of the estimated parameters in each model. Symbols in table are defined as number of parameters (k), corrected Akaike-information criterion (AIC_c), the difference in AIC_c between the best model and the i^{th} model ($\mathit{\Delta}_i$), and the relative probability that the i^{th} model is the best model in the candidate set (w_i).

Model	k	AIC_c	$\mathit{\Delta}_i$	w_i
DISCHARGE	3	567.442	0.000	0.287
DISCHARGE + L_F	4	567.454	0.012	0.286
DISCHARGE + ORIGIN	4	567.507	0.065	0.278
DISCHARGE + L_F + ORIGIN	5	568.972	1.530	0.134
ORIGIN	3	574.948	7.506	0.007
L_F	3	575.585	8.143	0.005
L_F + ORIGIN	4	576.309	8.867	0.003

Table 2.3. Regression coefficients for covariate effects on probability of using the Stillwater Branch. Parameter estimates for the model of p(Stillwater Branch) that included all covariates (p(Stillwater Branch) ~ DISCHARGE), showing direction of relations between p(Stillwater Branch) and discharge. Symbols are defined as the logit-scale parameter estimates (β_j), standard error (S.E.), critical value of the test statistic (z), and the p-value for the test (P).

Parameter	β_j	*S.E.*	z	P
INTERCEPT	-3.0131295	0.3656585	-8.240	< 0.001
DISCHARGE	0.0013813	0.0003941	3.505	< 0.001

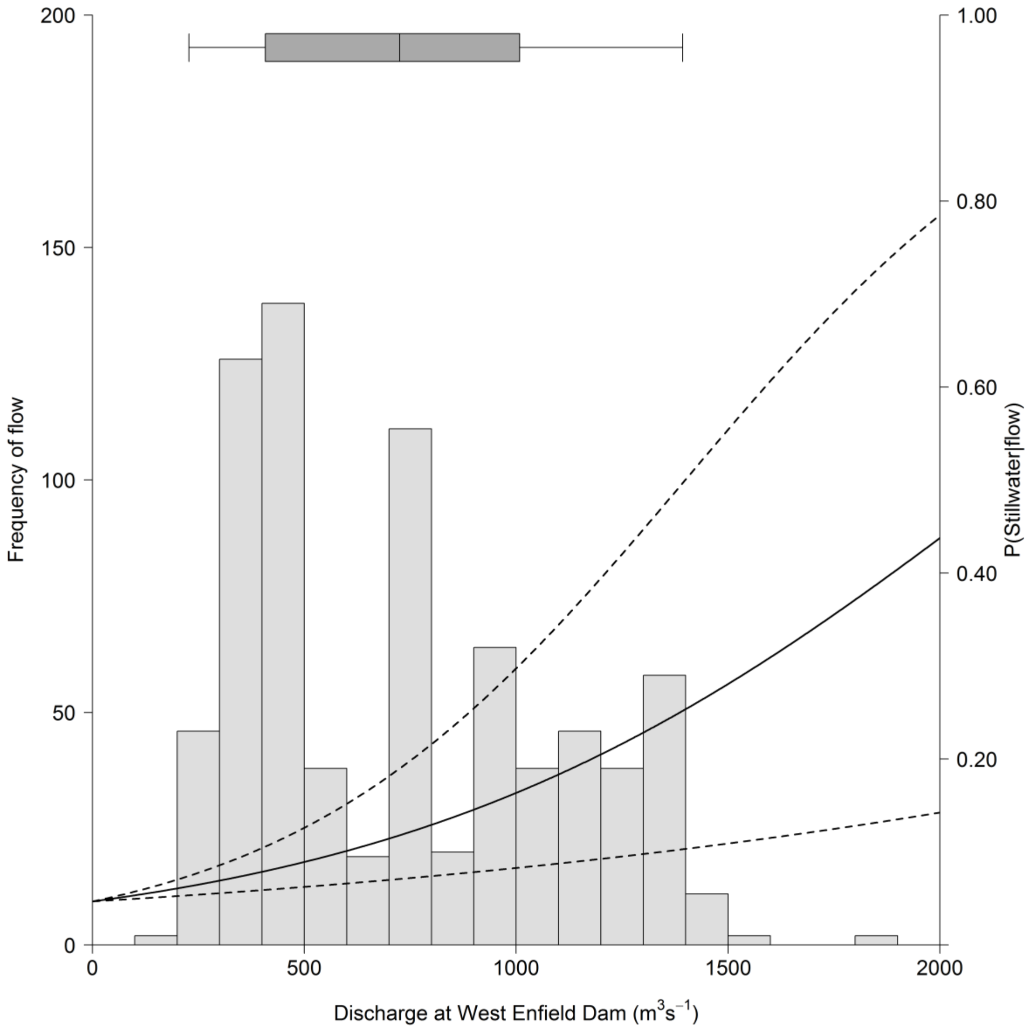


Figure 2.5. Relationship between discharge and use of Stillwater Branch. Shown is frequency of observed mean daily discharge values (histogram bars) during the 2005—2012 *S. salar* smolt runs compared to predicted proportional use of the Stillwater Branch (solid curve) and asymmetric 95% prediction intervals (dashed lines). The horizontal boxplot at the top of the plot indicates median value of observed discharge, the box ends represent the inner quartile for values of observed discharge, and the whiskers represent the 95% confidence limits of observed discharge values during smolt runs 2005-2012.

Survival around Marsh Island

Estimated survival of *S. salar* smolts (from acoustic-MS models) varied between reaches and between states during passage through the Marsh Island hydropower complex (Figure 2.6). Survival through the complex generally was higher for smolts that migrated through the Stillwater Branch than for smolts that migrated through the main-stem of the Penobscot River. Acoustic-telemetry estimates of mean (95% CI) *S. salar* smolt survival through the 1-km reach of the main-stem Penobscot containing Milford Dam ranged from 0.75 (0.51 – 0.89) to 1.00 (1.00 – 1.00) during 2005 through 2012. In contrast, per-kilometer survival through any of the free-flowing (undammed) reaches in the Penobscot River was ≥ 99% in all years (data not shown). Survival at the main-stem dams, Veazie (0.99 ± 0.00) and Great Works Dams (0.98 ± 0.02), that were removed was higher than at Milford Dam (0.91 ± 0.02) in all six years of this study. Similarly, mean survival across years at the two dams in the Stillwater Branch was high at the Stillwater (0.97 ± 0.02) and Orono Dams (1.00 ± 0.00).

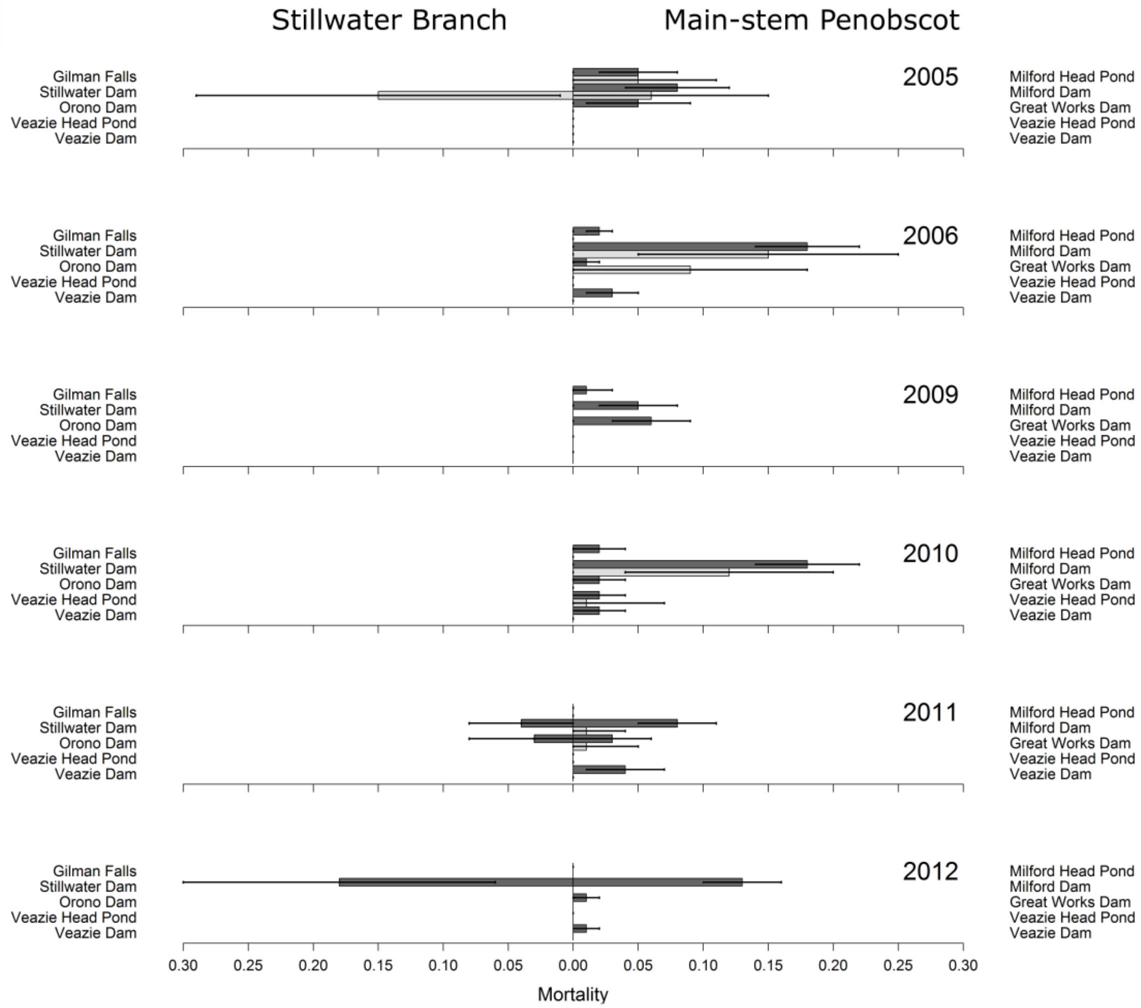


Figure 2.6. Mortality through the Marsh Island hydropower complex. Reach-specific mortality (calculated as one minus apparent survival in each reach) of acoustically tagged *S. salar* smolts of wild (light gray) and hatchery (dark gray) origin through the Stillwater Branch (left) and main-stem Penobscot River (right) during passage of the Marsh Island hydropower complex in each year of study from upstream (top of each plot) to downstream (bottom of each plot). Names of the reaches in each migration route are shown to side of plots, and correspond to intervals containing dams in the acoustic array shown in Figure 2.1. Mortality during the final two reaches (Veazie Head Pond and Veazie Dam) occurred downstream of the confluence of Stillwater Branch and Penobscot River, and therefore was experienced by all fish, regardless of migration route.

Movement and survival through Milford Dam

The most parsimonious models for the 2010 and 2012 radio-telemetry analyses differed between years and model selection for multistate radio-telemetry models are presented with the results (Table 2.4). No loss of tags or tagging-related mortality was observed in fish that were dummy tagged as part of the 2010 radio-telemetry study.

In 2010, a drawdown of the Milford head pond coincided with the radio-telemetry study such that any smolts passing through Milford Dam must have done so via the powerhouse. Therefore, estimates of path choice and of survival through the spillway were not made in 2010, although model selection suggested that survival did vary between reaches of the study area (Table 2.4). The mean (95% CI) survival of *S. salar* smolts through the Milford powerhouse was 0.90 (0.79 – 0.95) in 2010 according to models based on radio-telemetry locations. In 2012, discharges allowed for estimation of path-specific survival through Milford dam using multi-state models based on radio-telemetry locations. The 2012 radio-telemetry study indicated that estimated mean survival of *S. salar* smolts did not differ between the powerhouse (0.88, 95% CI: 0.42 – 0.99) and the spillway (0.88, 95% CI: 0.76-0.94; Figure 2.7). This finding was corroborated by the fact that the model using state-specific survival rates did not receive a meaningful amount of support in the candidate model set of 2012 radio-telemetry models of smolt survival through Milford Dam (Table 2.4). The wide confidence intervals for individual estimates of survival through the powerhouse suggest that precision of the powerhouse survival estimate may have been low owing to the small probability of smolts using that movement path (0.09, 95% CI: 0.05 – 0.16) in 2012.

Table 2. 4. Model-selection statistics for the 2010 and 2012 radio-telemetry models of *S. salar* smolt survival through Milford Dam. Parameters estimated in the multi-state (MS) mark-recapture survival models were survival ($\widehat{\boldsymbol{S}}$), detection probability ($\widehat{\boldsymbol{p}}$), and state-transition probabilities ($\widehat{\boldsymbol{\psi}}$) for transitions between river/spillway (state A) and the powerhouse (state B) at Milford Dam. Symbols in the table heading are defined as in Table 2.2. Reported number of parameters does not include parameters fixed for maximum likelihood estimation (e.g. $\Psi_{3BA} = 0.00$ for MS models used in 2012).

Year	Model	k	AIC$_c$	Δ_i	w_i
2010	$S_{(reach)}\,p_{(.)}$	6	152.187	0.000	0.854
	$S_{(reach)}\,p_{(reach)}$	10	155.913	3.725	0.133
	$S_{(.)}p_{(reach)}$	6	160.574	8.386	0.013
	$S_{(.)}p_{(.)}$	2	267.000	114.813	0.000
2012	$S_{(reach)}p_{(state*reach)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	11	615.538	0.000	0.935
	$S_{(.)}p_{(state*reach)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	10	621.094	5.557	0.058
	$S_{(reach)}p_{(reach)}\ \Psi_{AB(reach)}\Psi_{BA(reach)}$	9	626.331	10.793	0.004
	$S_{(state*reach)}p_{(state*reach)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	10	628.413	12.876	0.002
	$S_{(state*reach)}p_{(reach)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	11	630.504	14.967	0.001
	$S_{(.)}p_{(reach)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	7	630.887	15.350	0.000
	$S_{(reach)}p_{(.)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	7	682.472	66.934	0.000
	$S_{(state*reach)}p_{(.)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	10	688.694	73.156	0.000
	$S_{(.)}p_{(.)}\Psi_{AB(reach)}\Psi_{BA(reach)}$	3	737.870	122.332	0.000

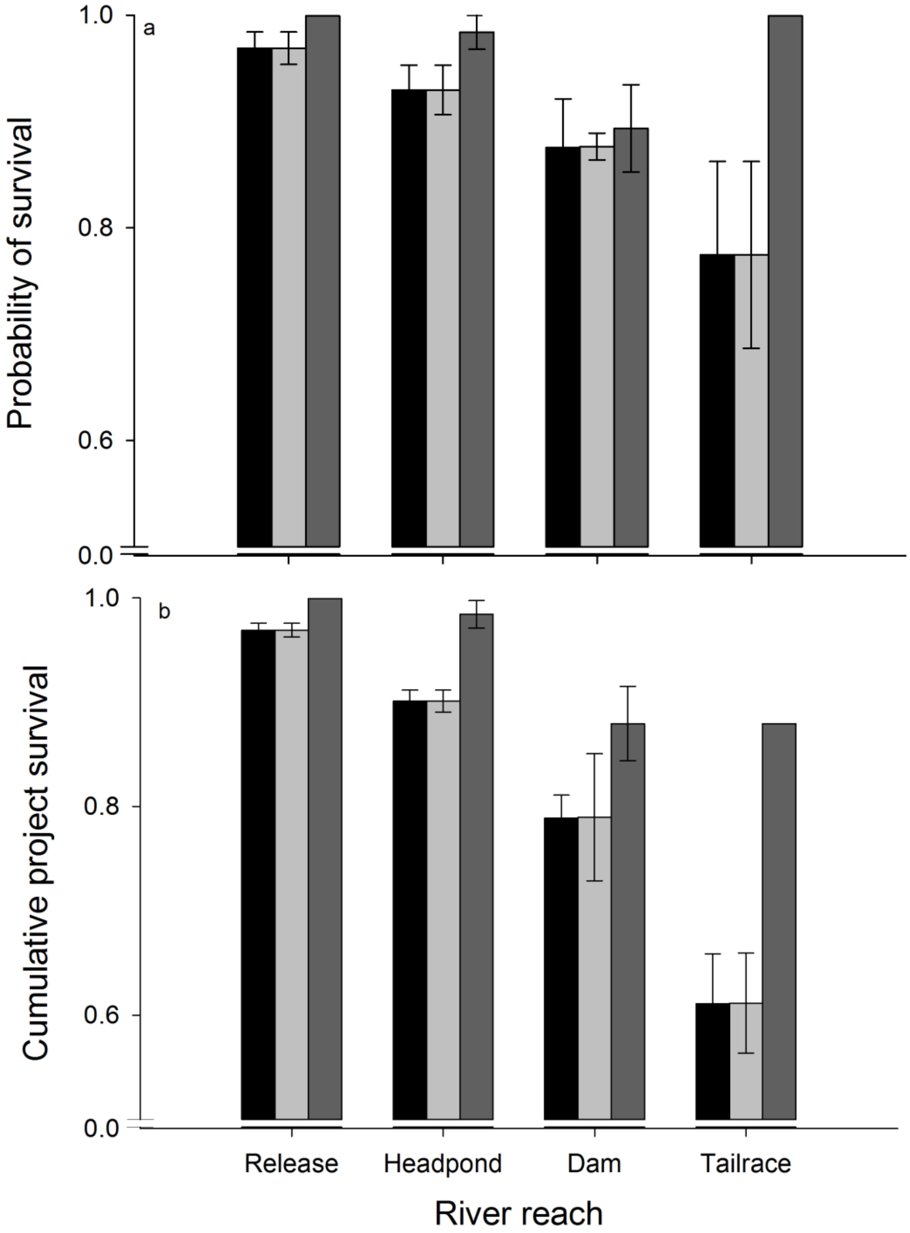


Figure 2.7. Plots of survival through Milford Dam. Shown are (a) reach-specific, and (b) cumulative survival of radio-tagged *S. salar* smolts through the Milford Dam powerhouse and spillway during 2010 and 2012 radio-telemetry studies. Model selection suggested that there was no difference between survival through the powerhouse and spillway in 2012, as is indicated by the high degree of overlap between the two estimates. Black bars represent survival through spillway in 2012, light-gray bars represent survival through the powerhouse route during 2012, and dark-gray bars represent survival through the powerhouse path in 2010.

Discharge experienced by smolts in each year was found to explain a relatively large amount of variation in estimated smolt survival through Milford Dam ($R^2 = 0.44$), and had a positive influence on smolt survival (simple linear regression, df = 15, $F = 11.89$, $P < 0.01$; Figure 2.8).

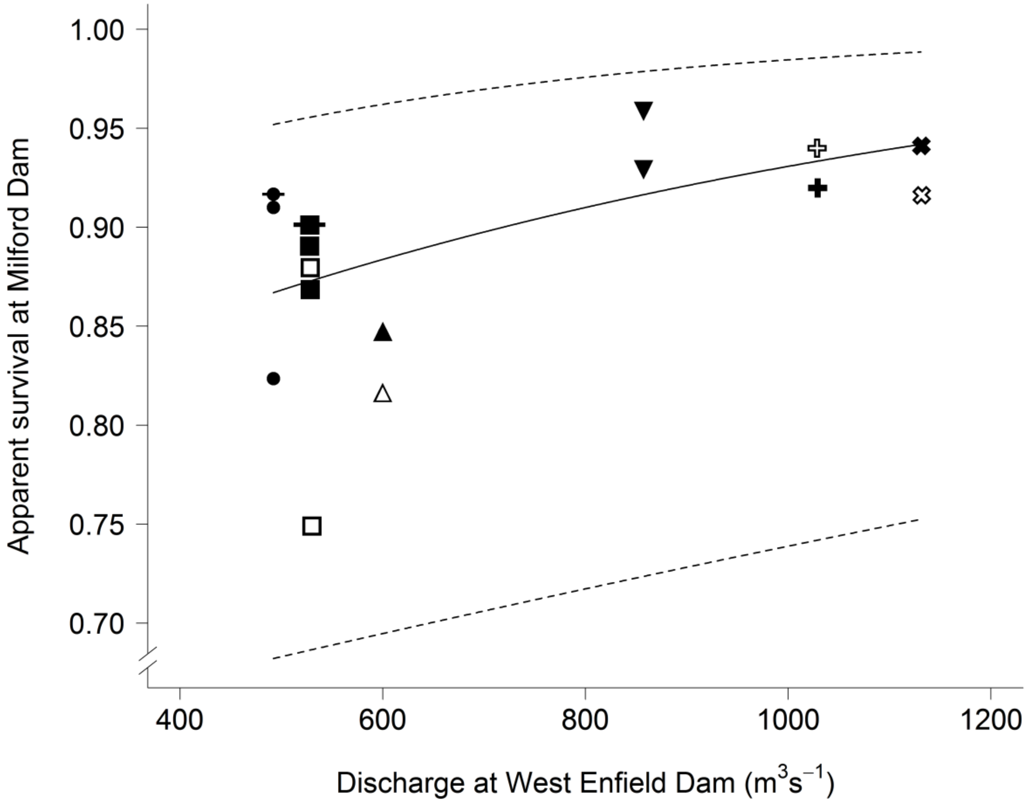


Figure 2.8. Plot of relationship between discharge and survival at Milford Dam. Simple linear regression used to characterize the relationship between mean daily discharge at West Enfield Dam during the smolt run for each year and estimated annual probabilities of *S. salar* smolt survival during passage of Milford Dam by each release cohort of acoustic- and radio-tagged fish. A logit-transformation was used on the response in the analysis and as such the y-axis is labeled with probabilities but is scaled on the logit. Symbols represent survival estimates from 2005 (upside-down triangles), 2006 (x), 2009 (triangles), 2010 (squares), 2011 (+), and 2012 (circles). Within each year, open symbols correspond to survival estimates for wild smolts and closed symbols are for hatchery-reared smolts. Radio-telemetry estimates in 2010 and 2012 are indicated by strike-through.

DISCUSSION

Passage through the Marsh Island hydropower complex represents a critical transition during downstream migration of the federally endangered *S. salar* population in the Penobscot River, Maine USA. This hydropower complex represents the final set of physical barriers to downstream migration in the Penobscot River. To reach the free-flowing portion of the river (and eventually the estuary) all of the out-migrating smolts in this system must pass either through Milford Dam on the east side of Marsh Island by using the main-stem Penobscot River or through the west side using the Stillwater Branch with its two operational dams (Stillwater Dam and Orono Dam). The present study provides a baseline of information about fish passage through the Marsh Island hydropower project before anticipated changes to discharge around the island, installation of new powerhouses at Stillwater and Orono dams, and installation of new downstream-passage facilities at each of those facilities.

Movement and survival through the main-stem Penobscot River

Milford Dam represents a potential impediment to restoring effective downstream passage of *S. salar* in the main-stem of the Penobscot River. It also offers the greatest opportunity for improvement of smolt passage in the lower river. By virtue of its location in the watershed (the lowest remaining dam in the main-stem), Milford Dam may be predicted to affect the success of diadromous fish migrations more than many of the other dams in the system. A large proportion (75% — 94%) of the total number of migrating *S. salar* smolts in the Penobscot watershed passes Milford Dam each year. Smolt survival through Milford Dam averaged 91% over the six years this study (range = 75–100 %).

Survival at Milford dam is among the lowest of dams in the system (Holbrook et al. 2011). Survival past this dam is also lower than the combined effects of dams in the alternative migration route (the Stillwater Branch in 2005-2012) through the Marsh Island hydropower complex (present study). Estimated survival through Milford Dam is also low relative to salmonid smolt survival at larger dams with greater generating capacity throughout Pacific Coast systems such as the Columbia River (e.g. Ferguson et al. 2007; Skalski et al. 2009). Taken together, these facts suggest that Milford Dam is the most-limiting impediment to *S. salar* smolt migrations in the lower 100 km of the Penobscot River. Furthermore, if passage at Milford Dam is not improved, then the regulatory requirement of 96% passage will not likely be met in the future.

Smolt survival through Milford Dam was estimated under a wide range of discharges during the six years of this study, and survival through the dam was found to be positively related to discharge experienced by fish during the smolt run each year. This observation is consistent with observations on other systems in which higher survival is observed past impoundments under higher flow conditions (e.g. Connor et al. 2003; Smith et al. 2003). These findings indicate that there may be some potential for regulation of upstream discharge at dams in the upper Penobscot River to be useful as a tool for managers to improve downstream passage success of smolts at Milford Dam (Connor et al. 2003). Indeed, by increasing discharge at regulated dams upstream of those examined in the present study during peak migration, smolt survival might be improved at Milford Dam through mechanisms related to passage and path choice.

While path choice through dams can often influence survival, it is notable that there did not appear to be any differences in path-specific estimates of survival between smolts that used the powerhouse or the spillway at Milford Dam. The probabilities of survival through the powerhouse (88%) and the spillway (88%) were strikingly similar, and they agreed well with the annual survival estimated for acoustically tagged smolts through Milford Dam (91%). This suggests that the mechanism resulting in increased survival during high discharge is not likely to be related to passage path (powerhouse or spillway) at the Milford Dam, and could potentially be a result of decreased passage time (Smith et al. 2003) and thus reduced exposure to physical injury at dam structures and from predators congregating above and below the dam (Venditti et al. 2000; Antalos et al. 2005) during high-discharge events (Raymond 1979). Similarly, mortality experienced by smolts at Great Works and Veazie Dams did not appear to be directly related to turbine passage because mortality during 2005-2010 at these facilities was similar to mortality during years in which turbines were shut down during the smolt run (2011 and 2012).

In future assessments of the results of the PRRP, it is important to understand and differentiate between the acute effects of management actions on individual species and the integrated effects of the project as a whole. The benefits of conservation efforts in the Penobscot River are likely to be species-specific and responses to restoration efforts also will be specific to life-history stages for any species. The removal of Great Works and Veazie Dams is expected to improve upstream passage of adult *S. salar* (Holbrook et al. 2009; NMFS 2012) and will increase access to nearly 100% of historical habitat for other species such as Atlantic sturgeon *Acipenser oxyrinchus* Mitchill 1815, shortnose sturgeon *A.brevirostrum* Lesueur1818, Atlantic tomcod *Microgadus tomcod* Walbaum 1972, and striped bass *Morone saxatillis* Walbaum 1792 (Trinko Lake et al. 2012). Thus, restoration efforts in the Penobscot River are expected to provide benefits to adult *S. salar* in addition to various life-history stages of several other species. However, the results of the present study suggest that the benefits afforded to *S. salar* smolts through the Penobscot River Restoration Project will be minimal in the lower main-stem Penobscot because estimated smolt survival at the two dams that were removed in the main-stem, Great Works Dam (99%) and Veazie Dam (98%), were already high prior to the removal of those dams (at least during the period studied). Rather, for smolts using the main-stem of the river, improved passage will depend largely upon anticipated improvements to downstream passage at Milford Dam or use of the alternative migratory path through the Stillwater Branch.

Movement and survival in the Stillwater Branch

Though only 6-25% of fish use the Stillwater Branch, survival through this migratory route historically has been high relative to survival through the main-stem Penobscot River. In most years survival was near 100% at Orono and Stillwater dams prior to PRRP actions. The estimated survival of 1.00 at the Orono facility in all years indicates that there may have been some difficulty in estimating survival at this dam due to the small number of fish using the Stillwater Branch; however; inspection of empirical relocation data at sites above and below the dam also suggest that survival was near 1.00 at this facility in all years. Even estimates of minimum survival based on empirical data (0.97) that ignore detection probability suggest that the per-kilometer rate of survival ($0.99 \cdot km^{-1}$) was indistinguishable from survival in free-flowing reaches of the river ($0.99 \cdot km^{-1}$; Holbrook et al. 2011). In all years of the present study but one, mean passage success at each dam in the Stillwater Branch was higher than the minimum standards for passage (96%) that will be required under the species protection plans for the two dams (NMFS, 2012). If downstream passage success through the Stillwater Branch is reduced below these historically high survival rates, by the addition of new generating capacity, the net result of the restoration project for *S. salar* smolts will be an overall reduction in survival through the Stillwater Branch, even if performance standards for downstream passage are met. This is because the criteria of 96% survival at each dam could compound into a cumulative survival of just 92% through the two dams in the Stillwater Branch. Based on historically high (and therefore difficult-to-estimate) survival in the Stillwater, combined with the small numbers of fish that use the migratory route each year, studies that stock tagged fish directly in the Stillwater may provide the most useful method for assessing possible future changes in passage success at these dams.

Proportional use of the Stillwater Branch by out-migrating smolts was variable among the six years of the present study, and as many as 25% of migrating smolts used this route each year. Operational and structural changes at Stillwater and Orono Dams in the Stillwater Branch increase total energy production from 4.3 MW to 10.2, more than doubling the capacity of these dams over the pre-restoration configuration. Legal provisions exist that will allow for modest increases to discharge in the Stillwater Branch from the current level of 30% of upstream (main-stem) discharge to 40% of upstream discharge (FERC, 2004). While the proportional use of the Stillwater Branch by smolts is clearly related to bulk flow in the lower river (Figure 2.5) the importance of the proportional distribution of flows between the main-stem and the Stillwater Branch in this relationship remains unclear. In the future, data about relative distributions of flow through each branch of the lower river could provide invaluable information about effects of management on smolt passage.

The effects of operational and structural changes in the Stillwater Branch Dams have the potential to affect smolt survival in the lower river in two ways. First, if discharge through the Stillwater Branch is increased, I hypothesize that a greater proportion of migrating smolts will use this migration route. Second, with increased generation and number of turbines, I hypothesize that Stillwater Branch smolt survival could decrease below historic rates, especially at Orono Dam where mean annual survival was near 100% during all 6 years of the present study. Thus, as in the main-stem of the Penobscot River, it seems likely that there will be no net gain in smolt survival through the Stillwater Branch through the actions of the PRRP. In the future, monitoring changes in discharge in the Stillwater Branch, concurrent with smolt survival, will be imperative for evaluating the success of the restoration project with respect to *S. salar* smolts.

Uncertainty in restoration

Predicting the influence of large-scale conservation efforts for any given species involves some understanding of the uncertainty surrounding expected results (Simenstad et al. 2006; Millar et al. 2007). Despite the utility of basin-scale restoration as a conservation tool (Opperman et al. 2011) the results of the present study indicate that the individual effects of specific dams have important, site-specific and species-specific consequences for restoration of downstream fish passage (improvements in fish survival, in this case) within large-scale conservation projects. This demonstrates the importance of monitoring individual sites for adaptive management and governance within basin-wide restoration projects (Gunderson and Light 2006; Opperman et al. 2011; Trinko Lake et al. 2012). In the Penobscot River, management agencies will have a good, working knowledge of the baseline conditions for survival of smolts by which progress can be measured. Few systems have such an unambiguous quantification of both the sites and magnitudes of loss during downstream migration.

Continued monitoring of passage through the hydropower complex in the lower river will provide the ability to assess management strategies and hydropower operations through the complex. Importantly, uncertainty in the effectiveness of downstream passage facilities and proportional discharge through the Marsh Island hydropower complex strongly suggests that monitoring will be fundamental for understanding biological changes in the river in response to ongoing changes in dam operation, and ultimately for determining the effects of the Penobscot River Restoration Project on the success of *S. salar* smolt passage in the lower Penobscot River.

Even in “natural” systems, the transition into the lower river and estuary of coastal systems is known to be a period of high mortality for *S. salar* smolts, owing to high rates of predation (Blackwell et al. 1997; Kocik et al. 2009) and increased susceptibility to both physical and physiological stressors (McCormick et al. 1998). The mortality experienced during this transition can be exacerbated due to the direct and indirect effects of dams such as disorientation, migratory delays (Mathur et al. 2000; Keefer et al. 2012), increased exposure to predators (Poe et al. 1991; Blackwell and Juanes 1998), and physical injury (Stier and Kynard 1986; Zydlewski et al. 2010) caused during dam passage.

The present study only examined acute mortality at dams in the lower Penobscot. It is possible that smolts experiencing different conditions through the two migration routes in the lower river also express different responses to the stressors encountered during later estuary migration and seawater entry. Fish passing dams that have increased rates of mortality may also experience elevated rates of delayed mortality downstream (Schreck et al. 2006). In the future, these considerations may become increasingly important in determining the overall effects of changes in the main-stem of the Penobscot River and the Stillwater Branch, and may hold previously unrecognized benefits for improvement of downstream migration of *S. salar* smolts.

CHAPTER 3

CATCHMENT-WIDE SURVIVAL OF WILD AND HATCHERY-REARED ATLANTIC SALMON SMOLTS IN A CHANGING SYSTEM

INTRODUCTION

Emigration from fresh water to estuaries, and ultimately the ocean, is a period of high mortality in the life-history of anadromous fishes. Mortality can result from acclimation to novel environments and food sources (Larsson et al. 2011), physiological challenges (Järvi 1989), and predation (Antalos et al. 2005; Blackwell et al. 1997). Anthropogenic perturbations to aquatic systems (e.g., pollution, dams, and climate change) may reduce viability of migratory fish populations. Dams reduce the structural and functional connectivity of migratory corridors (Herbert and Gelwick 2003; Hall et al. 2010; Branco et al. 2014) through physical inhibition (Keefer et al. 2012), and physiological impairment (Zydlewski et al. 2010). Mortality can occur due to dam-related injury (Mathur et al. 2000) or elevated predation risk (Poe et al. 1991; Blackwell and Juanes 1998).

Atlantic salmon *Salmo salar* L. stocks in North America diminished beginning in the 1800s, due to pollution, poor land-use practices, dams and overfishing (Haines 1992; Parrish et al. 1998). Many of the stocks in the southern range of the species are listed as critically endangered in Canada and the United States. Distribution of anadromous *S. salar* in the US is now restricted to several rivers in Maine that constitute what remains of the Gulf of Maine distinct population segment (U.S. Fish and Wildlife Service and National Marine Fisheries Service 2000). Most of the remaining runs in the US are maintained through intensive hatchery supplementation, a practice that has been in use since the 1800s (Moring 2000), although limited wild spawning does occur (U.S. Atlantic Salmon Assessment Committee 2014). More than 0.5 million smolts have been stocked each year since 1977 and these fish have constituted the majority (75%) of returns to the U.S. during that time period (U.S. Atlantic Salmon Assessment Committee 2014). Despite stocking of *S. salar* at virtually every juvenile life stage over the past several decades, adult returns to the U.S. remain at multi-decadal lows.

One major driver of reduced *S. salar* stock abundances is the effect of ocean conditions on early marine growth (Friedland 1998; Friedland et al. 2000) and survival (Salminen et al. 1995; Friedland et al. 2003a, b). Few options exist for improving marine survival (Hansen et al. 2012; Russell et al. 2012), and therefore a primary tool for recovery of imperiled stocks is to maximize the number of smolts that reach the ocean to offset high mortality there. Changes to links between marine and freshwater ecosystems (Friedland 1998), as well as impediments to passage (such as dams) in migration corridors can limit population recovery (Parrish et al. 1998; Johnsen et al. 2011).

The transition from a territorial parr to a migratory smolt is a complex suite of physiological, morphological, and behavioural transformations that prepare the fish for a life at sea (McCormick et al. 1998). The timing of smolting is controlled by photoperiod and temperature, defining a limited period of preparedness for saltwater entry, the ‘physiological smolt window’ (McCormick et al. 1998). This developmental period is coincident with favorable environmental conditions for saltwater entry, the ‘ecological smolt window’ (McCormick 2013). The overlap between the physiological and ecological smolt windows reflects the adaptive nature of smolting, resulting in optimal probability of survival during a suiTable 3.period each year (McCormick et al. 1998). Migratory delay, physiological impairment, and mortality can reduce the number of smolts reaching the ocean. Accumulating effects of passing multiple dams can further interfere with migration (Branco et al. 2014, Stich et al. in review). There is a cumulative effect of passing multiple dams, which results in elevated mortality during the early marine phase of salmonid migrations in both Pacific (Budy et al. 2002; Schreck et al. 2006) and Atlantic (Stich et al. in review) rivers. The magnitude of freshwater migratory mortality occurring from the passage of multiple dams is still not well characterized.

Among the extant stocks of *S. salar* in the U.S., the Penobscot River population is the largest, contributing to more than 75% of total U.S. adult returns each year since the 1970s (U.S. Atlantic Salmon Assessment Committee 2014). A major restoration project (the Penobscot River Restoration Project, PRRP), started in 2004 with the goal of balancing hydropower production in the river with the restoration of sea-run fishes (11 species). Major changes in hydropower generation have occurred in the Penobscot River since 2009 as part of the PRRP (Day 2006). Specific details of changes to hydropower dam operations and downstream fish passage resulting from combined effects of dam removal and hydropower reallocation in the Penobscot are described fully by the Federal Energy Regulatory Commission (2009) and National Marine Fisheries Service (2012a, b). Some changes were linked directly to restoration actions taken within the PRRP; others resulted from legal provisions of the Lower Penobscot River Basin Comprehensive Settlement Accord (Federal Energy Regulatory Commission 2004).

Starting in spring 2010, generating turbines at Howland (Figure 3.1 G), Great Works (Figure 3.1, E) and Veazie (Figure 3.1, F) Dams were shut down during the smolt migration each year until the removal of Great Works Dam in summer 2012 and the removal of Veazie Dam during summer 2013 as part of the species protection plan (National Marine Fisheries Service 2012a). Seasonal shutdowns continue at Howland Dam. Hydropower generation at Milford Dam (Figure 3.1, D) increased in spring 2012 with the addition of two turbines. Concurrent with these actions, hydropower generation at the Stillwater (Figure 3.1, B) and Orono (Figure 3.1, C) Dams was increased by raising head pond elevation (Federal Energy Regulatory Commission 2005). A secondary powerhouse was constructed at both the Stillwater and Orono Dams during 2013, which approximately doubled the generating capacity of each facility prior to the 2014 smolt migration (National Marine Fisheries Service 2012b). Finally, the owner of the dams in the lower river is licensed to increase flow diversion to the Stillwater Branch from 30% to 40% of total river discharge, after which ability to control flow based on ponding at facilities around the island is lost (Federal Energy Regulatory Commission 2004).

The goal of this study was to use a multi-annual data set to assess the effects of tributary-specific management actions on the number of smolts reaching the ocean in the largest extant population of *S. salar* in the U.S., the Penobscot River stock, and determine what contribution freshwater reaches in the Penobscot River made to the total mortality within that stock from 2005 through 2014. Information about how changes to the hydro-system have affected survival would provide a necessary tool for assessing management actions, but a unified framework for analysis was needed. Therefore, the primary objective of this study was to use acoustic telemetry data from more than 1,800 *S. salar* smolts to estimate survival throughout the Penobscot River and its tributaries using multi-state mark-recapture models from eight years of smolt migrations before and after changes to hydropower projects in the catchment. As a secondary objective, I evaluated effects of survival covariates related to management.

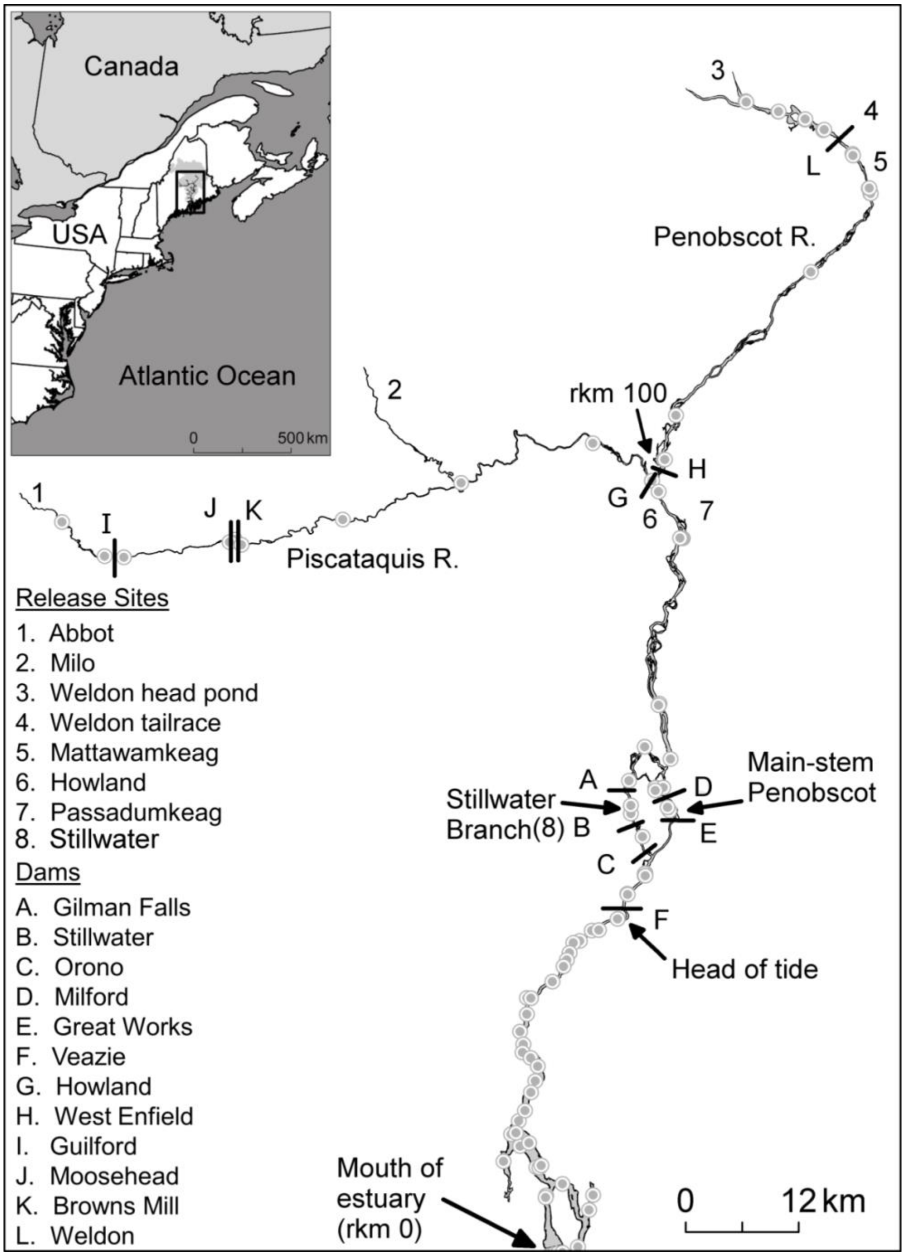


Figure 3.1. Acoustic receiver network. Shown are map of the Penobscot River catchment, showing location in North America (inset), locations of acoustic receivers (grey circles), release sites for acoustically tagged fish throughout the river, and locations of dams (bold lines). Not shown are 40^+ acoustic receivers deployed from the mouth of the estuary to the ocean.

METHODS

Study area

The Penobscot River (Figure 3.1) is the largest river in Maine, USA, comprising a drainage area of about 22,000 km^2. Despite that the Penobscot River stock of *S. salar* is the largest in the U.S., abundance has been low since the mid-to-late 20th century (Trinko Lake et al. 2011), being further diminished in recent years. As part of the species' recovery plan, *S. salar* has been stocked throughout the catchment at egg, fry, parr, and smolt life-stages (U.S. Atlantic Salmon Assessment Committee 2014). Egg and fry plantings have occured in headwater streams, and contribute proportionally few fish, in combination with wild spawning, to the smolt run each spring (U.S. Atlantic Salmon Assessment Committee 2014). Most migrating smolts are 18-month-old, hatchery-reared smolts stocked in the main-stem. As a result, the majority of the adult run (*c.* 83%) is made up of hatchery-stocked smolts with few naturally reared counterparts (U.S. Atlantic Salmon Assessment Committee 2012).

All *S. salar* (stocked or naturally reared) in the upper Penobscot River enter the main-stem at river kilometer (rkm) 100 during migration, passing the Howland Dam (Figure 3.1, G) or West Enfield Dam (Figure 3.1, H) near the confluence of the Upper Penobscot River and the Piscataquis River. Smolts approach the Marsh Island hydropower complex at rkm 60, where most (88%) remain in the main-stem to the east, and the remainder (12%) use the Stillwater Branch to the west (Figure 3.1) before entering the estuary (Stich, Bailey and Zydlewski 2014).

On the east side of Marsh Island (Penobscot River) smolts passed two dams until the removal of the Great Works Dam (Figure 3.1, E) in 2012 and Veazie Dam in 2013.

Previous estimates of annual survival through Great Works Dam (98%) and Veazie Dam (99%) were high relative to other dams in the Penobscot, and thus little improvement (*c.* 1%) in smolt survival is anticipated in that reach of the main-stem as a result of dam removal (Holbrook et al. 2011; Stich et al. 2014). Cumulative survival during migration through the Stillwater Branch (*c.* 96%), on the west side of Marsh Island, historically has been higher than cumulative survival through the main-stem (*c.* 88%), owing largely to low survival at Milford Dam (91%; Figure 3.1, D). Based on historically high passage efficiency, survival through the Stillwater and Orono Dams in the Stillwater Branch is not expected to increase with the doubling of hydropower generation at those facilities (Stich et al. 2014), although changes in survival through Milford Dam with the addition of two generating turbines and increased head pond height are somewhat less predictable. However, use of the Stillwater Branch by smolts increases with total river discharge (Stich et al. 2014), and as such the cumulative survival of smolts through the lower river has the potential to change based on flow diversion and use of the Stillwater Branch by smolts as well.

Acoustic tagging and releases

From 2005 through 2014 smolts were acoustically tagged (n = 2,056: Table 3.1) and released into the Penobscot catchment at locations ranging in distance to the mouth of the estuary from 63 to 187 rkm (Figure 3.1), of which 1,823 were subsequently relocated. Of the relocated fish 1,504 were hatchery-reared smolts from the U. S Fish and Wildlife Service (USFWS) Green Lake National Fish Hatchery (GLNFH), and the remaining 319 were wild-reared smolts captured in the Piscataquis or Penobscot River

(Table 3.1). Acoustic tagging methods were described in detail by Holbrook, Kinnison and Zydlewski (2011) and Stich, Bailey and Zydlewski (2014); identical procedures were used in all years from 2005 through 2014 of the present study. Briefly, smolts were anaesthetized using a 100 mg·L-1 solution of MS-222 (buffered with 20-mmol $NaHCO_3$; pH=7.0). A small (1-cm) incision was made slightly offset from the ventral line and 1-cm posterior to the pectoral fin girdle. An acoustic tag was inserted and the incision was closed with two simple, interrupted knots using 4-0 absorbable vicryl sutures (Ethicon, Somerville, New Jersey, USA). Model V7 acoustic tags (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) with a pinger volume of 136 dB were used in 2005 and for wild fish in 2011. Expected battery life of V7 tags was 69–80 days. In all other years, model V9 acoustic tags (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) with battery life of 80–82 days and volume of 151 dB were used.

Table 3.1. Data summary for acoustically tagged *Salmo salar* smolts released in the Penobscot River 2005–2014. Shown are number of fish relocated from release group (n), and within release groups, means of accumulated thermal units (ATU), discharge experienced during migration (Q, $m^3 \cdot s^{-1}$), photoperiod at release (PP, hours), and daily temperature experienced during migration (T, °C). The number in the parentheses under *n* is initial size of release group.

Year	Origin	Release Site	*n*	ATU	Q	PP	T
2005	Hatchery	Howland	103(150)	262	1416	13.9	7.8
2005	Hatchery	Mattawamkeag	10(40)	221	1058	13.4	5.9
2005	Hatchery	Milo	85(85)	268	1407	13.9	7.5
2005	Wild	Weldon	34(60)	417	1014	15.2	11.1
2006	Hatchery	Milo	66(72)	316	401	13.9	8.3
2006	Hatchery	Weldon	135(146)	333	489	13.9	9.2
2006	Wild	Weldon	46(73)	346	454	14.5	13.3
2009	Hatchery	Milo	96(100)	387	689	14.2	10.4
2009	Hatchery	Passadumkeag	97(100)	378	728	14.1	10.1
2010	Wild	Abbot	74(75)	376	297	14.3	14.0
2010	Wild	Weldon Head Pond	65(74)	447	283	14.5	13.5
2010	Hatchery	Milo	100(100)	364	351	13.9	10.1
2010	Hatchery	Passadumkeag	98(100)	364	359	13.9	9.7
2011	Wild	Abbot	74(75)	241	1068	14.7	10.9
2011	Wild	Weldon Head Pond	26(60)	390	743	15.2	15.0
2011	Hatchery	Milo	88(100)	228	1195	14.0	7.4
2011	Hatchery	Passadumkeag	97(100)	239	1231	14.0	8.1
2012	Hatchery	Abbot	72(72)	336	670	13.8	8.8
2012	Hatchery	Weldon Head Pond	84(85)	336	620	13.8	8.7
2013	Hatchery	Abbot	70(75)	253	715	13.5	6.7
2013	Hatchery	Weldon Head Pond	80(82)	253	665	13.5	7.4
2014	Hatchery	Abbot	68(75)	338	950	14.3	8.7
2014	Hatchery	Weldon Head Pond	80(82)	338	900	14.3	9.0
2014	Hatchery	Stillwater	75(75)	338	1005	14.3	8.3

Acoustic array

Smolts were tracked using an array of stationary VR2 and VR2-W acoustic receivers (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada). The array was deployed prior to tagging each year cooperatively between the United States Geological Survey (USGS) Maine Cooperative Fish and Wildlife Research Unit, the University of Maine, and the National Oceanic and Atmospheric Administration (NOAA). Coverage extended from rkm 187 in the Piscataquis River and rkm 165 in the East Branch of the Penobscot River to the mouth of Penobscot Bay (rkm -45; Fig.1). Numbers and locations of acoustic receivers used varied during the study years, and as such I restricted my analysis to locations that were common to most years. Acoustic receivers deployed on river bottom with 45-kg concrete anchors in the freshwater and estuary reaches, while receivers in the bay were tethered 10 m below the surface. Multiple receivers were deployed in a transect across the river to achieve cross-sectional coverage where needed; smolt detections at these locations were pooled for statistical analyses. For the purpose of this study, all acoustic receivers from the second location downstream of Veazie Dam (rkm 43.5) to the bay were pooled as a terminal detection event.

Multistate survival model

Survival was estimated in the Penobscot River 2005–2014 using multi-state (MS) mark-recapture models (Figure 3.2). Spatially explicit capture histories were created for each smolt using detections at acoustic receivers during one-way, downstream migration (Figure 3.2). To accommodate two upstream sources (Piscataquis River and East Branch), and two migration routes through the Marsh Island complex, three 'states' were used, in addition to a non-detected (absorbing) state. Detection in the Penobscot River (A), Piscataquis River (B), or Stillwater Branch (C) defined the state occupied by fish (Figure 3.2). Fish were assigned a zero (0) for locations at which they were not detected. Within each state apparent survival (S), probability of detection (p), and probability of movement into the other two states (ψ) were estimated. Survival estimated from these models is 'apparent' rather than 'true' survival (confirmation of dead fish is generally not possible), but I use the term 'survival' throughout for simplicity.

Parameters of MS models were estimated using a hierarchical (state-space) modeling framework (Calvert et al. 2009) in WinBUGS (Lunn et al. 2000) from the 'R2WinBUGS' package (Sturtz et al. 2005) in R (version 3.1.0; R Development Core Team 2014). The use of MS models allows for separate estimation of S and ψ while accounting for imperfect detection. The probability of detecting a smolt was contingent upon the state occupied by fish and probability of survival within that state. The state occupied by fish was conditional on the probability of moving between states in the previous interval, as well as on the initial state occupied, which was known (stocking location). Therefore, the likelihood for MS models incorporated components describing the state and the observation processes.

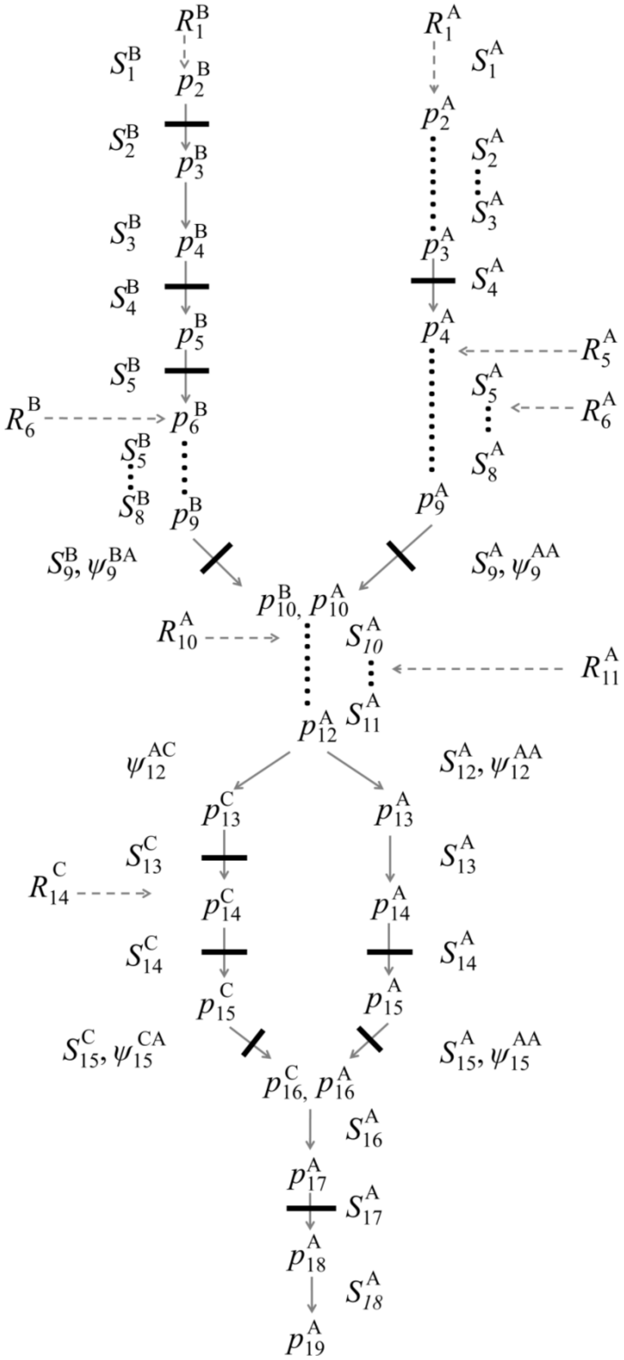


Figure 3.2. Multistate modeling framework for freshwater survival. Schematic representation of multi-state mark-recapture models used to estimate apparent survival (*S*), probability of detection (*p*), and state-transition probabilities (*ψ*) of acoustically tagged *Salmo salar* smolts within each reach of the main-stem Penobscot River (A), the Piscataquis River (B), and the Stillwater Branch (C). The symbol ‘R’ represents release events that occurred at location *t* within state *h*. Bold lines represent dams.

In the state matrix of the MS models (Table 3.2), the probability of occupying a given state (h') during a given interval (t + 1) was based on the state of an individual (i) during the previous interval (t), the probability of survival in state h during interval t (S_t^h), and the probability of moving from one state (h) to another (h') immediately prior to interval t + 1, given survival during interval t ($\psi_t^{h,h'}$). Because of this the state-process model was conditioned on the state at first observation (stocking tributary was known, see Figure 3.2). As such, the likelihood used in the state-process model was defined (see Kéry and Schaub 2012) by the following component equations:

$$z_i fs_i = fs_i \qquad \text{(Equation 3.1)}$$

and

$$z_{i,t+1} | z_{i,t} \sim \text{categorical}\left(\boldsymbol{\Omega}_{z_{i,t},\, 1\ldots S,\, i,\, t}\right) \qquad \text{(Equation 3.2)}$$

where $z_{i,t}$ was the true state of each individual i at interval t, f was the state (s) of each fish at first encounter. The probability of an individual's true state was a categorical distribution described by the 4-dimensional matrix $\boldsymbol{\Omega}$ (Table 3.2) in which the first dimension was the observed state z at interval t, the second dimension was the vector of true states (1…S) at interval t + 1, the third dimension was the individual fish (i), and the fourth dimension represented interval, t (see Kéry and Schaub 2012 for a general structure).

Table 3.2. State-process matrix for multi-state mark-recapture models. Shown is the probability that an individual occupies state *h′* at interval *t*+1 given the true state *h* of the individual at interval *t*, the probability of surviving interval *t*, and the probability of changing states immediately before *t* + 1. The matrix is reduced from its general form to reflect parameter constraints imposed by biological constraints of the system and the downstream nature of *S. salar* smolt migrations.

	True state at $t+1$			
True state at t	Penobscot (A)	Piscataquis (B)	Stillwater (C)	Absorbing (0)
Penobscot (A)	$S_t^{\mathrm{A}}\left(1-\psi_t^{\mathrm{AC}}\right)$	0	$S_{\mathrm{t}}^{\mathrm{A}}\left(1-\psi_t^{\mathrm{AA}}\right)$	$1-S_t^{\mathrm{A}}$
Piscataquis (B)	$S_t^{\mathrm{B}}\left(1-\psi_t^{\mathrm{BB}}\right)$	$S_t^{\mathrm{B}}\left(1-\psi_t^{\mathrm{BA}}\right)$	0	$1-S_t^{\mathrm{B}}$
Stillwater (C)	$S_t^{\mathrm{C}}\left(1-\psi_t^{\mathrm{CC}}\right)$	0	$S_{\mathrm{t}}^{\mathrm{C}}\left(1-\psi_t^{\mathrm{CA}}\right)$	$1-S_t^{\mathrm{C}}$
Absorbing (0)	0	0	0	1

Detection probability (p) was estimated separately for each interval in each state (p_t^h) in the MS models (Figure 3.2). The likelihood for the observation process, conditional on the state of individual, i, was defined as (see Kéry and Schaub 2012):

$$y_{i,t}|z_{i,t} \sim \text{categorical}(\mathbf{\Theta}_{Z_{i,t},\,1\ldots O,\, i\,,t}) \qquad \text{(Equation 3.3)}$$

where y was the observed state of individual i at interval t given the true state of that individual at interval t, and y was defined to have a categorical distribution described by the four-dimensional matrix, $\boldsymbol{\Theta}$ (Table 3.3). The first element of $\boldsymbol{\Theta}$ was the vector of true states, the second element was the vector of observed states (O), the third element was the individuals (i), and the fourth was interval, t (see Kéry and Schaub 2012).

Table 3.3. Observation-process matrix for multi-state mark-recapture models. Shown is the probability of being detected in a given state in interval *t* conditional on the true state of individuals at interval *t*. States of individuals were assumed to be known without error given that they were detected.

	Observed state at *t*			
True state at *t*	Penobscot (A)	Piscataquis (B)	Stillwater (C)	Absorbing (0)
Penobscot (A)	p_t^{A}	0	0	$1- p_t^{\mathrm{A}}$
Piscataquis (B)	0	p_t^{B}	0	$1- p_t^{\mathrm{B}}$
Stillwater (C)	0	0	p_t^{C}	$1- p_t^{\mathrm{C}}$
Absorbing (0)	0	0	0	1

Base model description

Multi-state models require that a parallel model structure operates across states with regard to survival and observation within states, as well as movement between them. In reality, there are biological constraints to this structure when modeling survival of smolts during downstream migration through a river. In these models, I assume that migration occurs in one direction (downstream), and thus some states can only be occupied by individuals during some intervals (see Figure 3.2). Within the state- and observation-process matrices, constraints were imposed on survival, state transition, and detection probabilities based on biological constraints of migration through the study system (Table 3.2). First, I did not allow downstream-migrating fish to transition to the Piscataquis River (B) from the Penobscot River (A) or the Stillwater Branch (C) during any interval, so ψ_t^{AB} and ψ_t^{CB} both were fixed to zero for all reaches (Figure 3.2 and Table 3.2).

We assumed smolts that failed to move downstream represented mortality even if this was not confirmed, because failed migrants typically make little to no population contribution due to high overwinter mortality (Horton et al. 2009). The only interval during which fish could move from the Piscataquis River into the Penobscot was $t = 9$ when the probability of transition was one given that fish survived (i.e. $\psi^{\mathrm{BA}}_{1\ldots8} = 0$, $\psi^{\mathrm{BA}}_{9} = 1$, and $\psi^{\mathrm{BA}}_{10\ldots19} = 0$), and no fish could move from the Piscataquis River directly into the Stillwater Branch ($\psi^{\mathrm{BC}}_{1\ldots19} = 0$) given the intervening main-stem Penobscot River. It was impossible to detect a fish in the Piscataquis River (state B) after $t = 8$, therefore $S^{\mathrm{B}}_{10\ldots19}$ and $p^{\mathrm{B}}_{9\ldots19}$ were fixed to one. The only interval during which fish could move from the Penobscot River (state A) into the Stillwater Branch was at $t = 12$, therefore $\psi^{AC}_{1\ldots11}$ and $\psi^{AC}_{13\ldots19}$ were fixed to zero. Fish could be located in the Stillwater Branch only when $t = 12\ldots14$; therefore, $p^{\mathrm{C}}_{1\ldots11}$, $p^{\mathrm{C}}_{15\ldots19}$, $S^{\mathrm{C}}_{1\ldots12}$, and $S^{\mathrm{C}}_{16\ldots19}$ all were fixed to one. Probability of fish moving from the Stillwater Branch into the main-stem Penobscot River after interval $t = 15$ was fixed to one given survival during interval $t = 15$.

Based on the constraints imposed above, the only state-transition probability estimated within MS models was the probability of moving into the Stillwater Branch from the Penobscot River during interval $t = 12$ (ψ^{AC}_{12}), and this parameter was assigned a uniform prior distribution between 0 and 1. Similarly, survival through intervals for which no constraint was applied ($S^{\mathrm{A}}_{1\ldots19}$, $S^{\mathrm{B}}_{1\ldots9}$, and $S^{\mathrm{C}}_{13\ldots15}$) and detection probabilities that were not constrained ($p^{\mathrm{A}}_{1\ldots19}$, $p^{\mathrm{B}}_{1\ldots8}$, and $p^{\mathrm{C}}_{12\ldots14}$) were assigned uniform prior distributions between zero and one.

We included the size of acoustic transmitters as an individual covariate of detection probability in the above MS survival model and all subsequent models that modified the structure of this ‘base model’ (Fig 3). Acoustic tag model (V7 = 0, V9 = 1) was included as a fixed effect. Because I previously have found that larger (i.e. louder) tags (V9) were easier to detect than smaller (V7) tags, I used a prior distribution for the effect of tag type defined as B(1, 1), producing a uniform distribution between zero and one. Similarly, I have observed an inverse relationship between freshwater discharge (Q) and detection. The prior for this covariate effect was defined as for tag type. For all intervals (t) in which detection $\left(p_t^h\right)$ was not fixed to one, the posterior probability $\left(\hat{p}_t^h\right)$ was modeled as a function of fixed effects (β_j) of tag model ('Tag_i') used and discharge (Q_i) for individual fish (i) using a logit link function as (Equation 3.4):

$$\text{logit}\left(\hat{p}_t^h\right) = \ln\left(\frac{p_t^h}{1\text{-}p_t^h}\right) + \beta_1 \times \text{Tag}_i + \beta_2 \times Q_i$$

Model estimation

I used Markov chain Monte Carlo methods to estimate survival, state-transition, and detection probabilities for the base model and all extensions thereof (see below). I ran three Markov chains for each parameter in each model, and chose random starting values for each individual chain from the prior distribution of each parameter. I used a burn-in of 3,000 samples and then sampled another 30,000 values from the posterior distribution of each parameter, keeping every third sample to reduce autocorrelation between samples and to increase the number of independent samples (effective sample size; Kruschke 2010). This resulted in a total of 1,000 burn-in samples in each chain, and 10,000 samples from the posterior distribution of each chain for each parameter estimated, yielding a total of 30,000 samples from which to construct the posterior distribution of each parameter. I assessed convergence of Markov chains using the

Gelman and Rubin convergence diagnostic ($\hat{r} \approx 1.00$ at convergence). I monitored the number of independent samples from the posterior distribution of each parameter (effective sample size) to ensure adequate sampling (Kruschke 2010). Unless otherwise specified, survival estimates are presented as mean (95% credible interval) in the results.

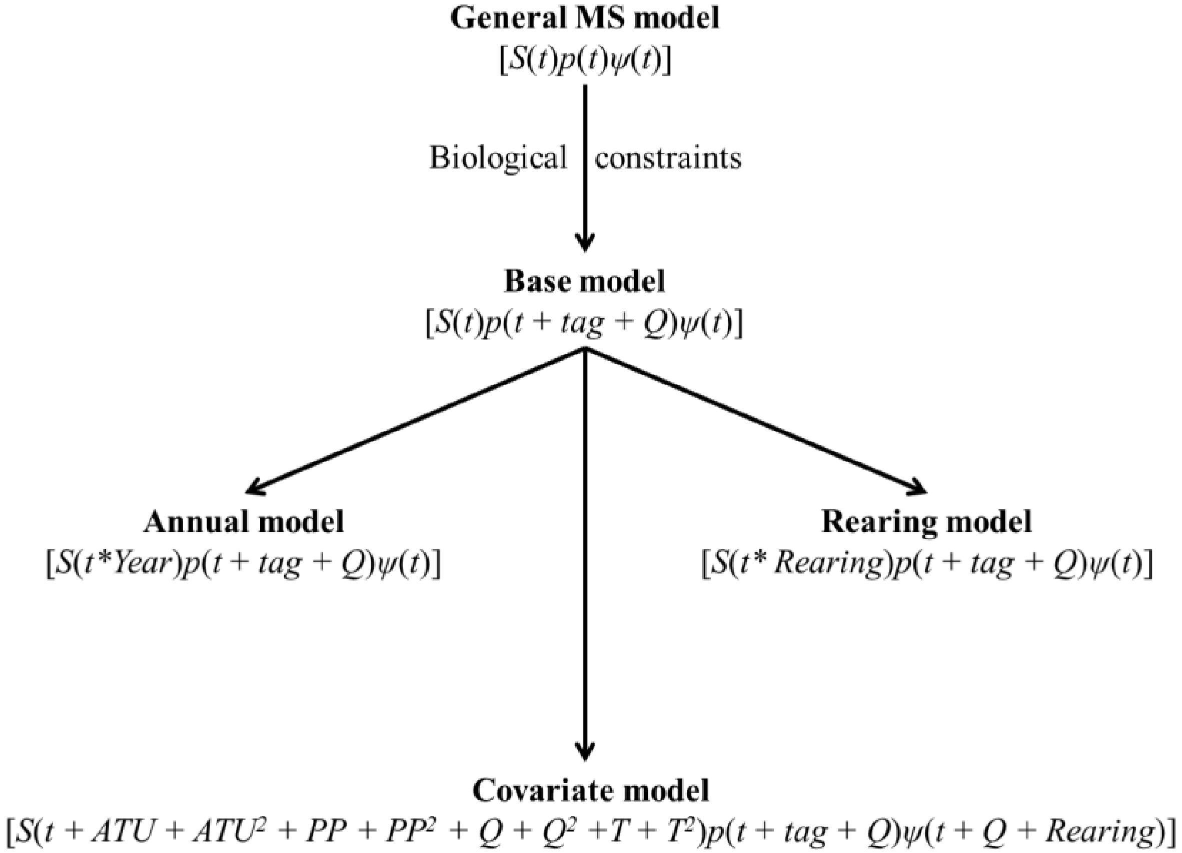


Figure 3.3. Model construction process for freshwater survival. Schematic of model development for multi-state mark-recapture models used to estimate interval-specific (indicated by '*t*') survival and detection probabilities, as well as proportional use of the Stillwater Branch by *Salmo salar* smolts in the Penobscot River catchment 2005–2014. The general MS model structure is given at the top, and is extended to include effects of tag model (*tag*) and discharge (*Q*) on detection probability (base model). The base model was further extended to estimate 1) annual survival ('Annual model') and effects of rearing history on survival ('Rearing model'), and 2) linear and quadratic fixed effects of discharge, accumulated thermal units (*ATU*), photoperiod (*PP*), and temperature (*T*) on survival, as well as effects of discharge and rearing history on use of the Stillwater Branch.

Derived quantities

To standardize survival as a per-km rate, survival ($\hat{S}_t^h$) for each interval (t) in each state (h) was raised to the power of one divided by interval length (D_t, km) to which the estimate corresponded ($\left[\hat{S}_t^h\right]^{\frac{1}{D_t}}$). This approach allowed direct comparisons of the posterior distributions of estimated survival within each interval. I calculated cumulative survival of *S. salar* smolts from the Piscataquis River to the estuary using per-kilometer survival through each interval as (Equation 3.5):

$$\hat{S}_{\text{Piscataquis}} = \prod_{t=1}^{9} \hat{S}_t^{\text{B}^{D_t}} \times \prod_{t=10}^{12} \hat{S}_t^{\text{A}^{D_t}} \times \left[\left(1-\hat{\psi}_{12}^{\text{AC}}\right) \times \prod_{t=15}^{15} \hat{S}_t^{\text{A}^{D_t}} + \hat{\psi}_{12}^{\text{AC}} \times \prod_{t=13}^{15} \hat{S}_t^{\text{C}^{D_t}} \right] \times \prod_{t=16}^{17} \hat{S}_t^{\text{A}^{D_t}}$$

Cumulative survival of smolts migrating from the upper main-stem of the Penobscot River to the estuary was calculated from the posterior distributions of per-kilometer survival in a similar fashion (Equation 3.6):

$$\hat{S}_{\text{Main-stem}} = \prod_{t=1}^{12} \hat{S}_t^{\text{A}^{D_t}} \times \left[\left(1-\hat{\psi}_{12}^{\text{AC}}\right) \times \prod_{t=15}^{15} \hat{S}_t^{\text{A}^{D_t}} + \hat{\psi}_{12}^{\text{AC}} \times \prod_{t=13}^{15} \hat{S}_t^{\text{C}^{D_t}} \right] \times \prod_{t=16}^{17} \hat{S}_t^{\text{A}^{D_t}}$$

In order to determine the cumulative effects of dam passage on the number of smolts arriving in the estuary, I compared cumulative survival derived from the above equations to the expected survival through the system in the absence of dams. To do this, I replaced survival through intervals containing dams or head ponds associated with dams with the posterior mean of estimated survival through all intervals that did not contain dams or head ponds.

Environmental covariates

I extended the base model to include date (photoperiod), discharge, and temperature experienced by smolts on survival (Figure 3.3). Photoperiod was calculated from latitudes in the watershed and day of year for detections. Temperature and discharge date were obtained from the U.S. Geological Survey river gage at West Enfield Dam.

Prior distributions for survival in the base model were modified to incorporate these factors in a ‘covariate model’ (Figure 3.3). As above, survival in each interval within each state $\left(S_t^h\right)$ was uniform between 0 and 1[U(0,1)], such that survival $\left(\widehat{S}_t^h\right)$ was modeled as a function of location (t) and individual fish (i) using a logit link function to estimate the fixed effect (β_j) of the j^{th} covariate (X_{ij}) as (Equation 3.7):

$$\operatorname{logit}\left(\widehat{S}_t^h\right) = \ln\left(\frac{S_t^h}{1\text{-}S_t^h}\right) + \sum_{j=1}^{k=8} \beta_j \times X_{ij} \ldots \beta_k \times X_{ik}$$

The prior distribution for each of the β_j, was constructed as a logit-transformation of a uniform distribution [U(0,1)] that was used to constrain the values to the probability scale. This resulted in a normal prior distribution for each coefficient on the logit scale [N(0,1.8)]. All covariates were standardized prior to analysis to speed model convergence and facilitate comparison between effects. I first ran a full additive model that included linear and quadratic terms each for 1) photoperiod, 2) discharge, 3) temperature, and 4) the accumulated thermal units experienced by smolts from 1 January to tagging (ATU, *sensu* Sykes, Johnson and Shrimpton 2009). These factors have been identified as major drivers of smolt migrations (McCormick et al. 1998). My rationale for including second-order terms for each of the environmental covariates was that there theoretically is some threshold after which effects on smolt survival might reverse or asymptote during migration. I ran the covariate model once (using settings described above), and used the posterior distributions for the coefficient estimates from the first run as priors to run the model a second time and estimate a probability that each covariate was retained (Tenan et al. 2014). To do this, I multiplied the coefficient of each covariate in the model by a random draw from a Bernoulli (i.e. 0 or 1) distribution (γ_j) with p = 0.5 for each of the environmental covariates (one each for the linear and quadratic term) as (Equation 3.8):

$$\text{logit}\left(\widehat{S}_t^{\,h}\right) = \ln\left(\frac{S_t^h}{1\text{-}S_t^h}\right) + \sum_{j=1}^{k=8} \gamma_j \times \beta_j \times X_{ij} \ldots \gamma_k \times \beta_k \times X_{ik}$$

and estimated the probability of each covariate being in the model as the mean of the Bernoulli draws for samples that were retained (see Tenan et al. 2014).

I used the same process to investigate influences of discharge and rearing history on the probability of using the Stillwater Branch (ψ_{12}^{AC}) as a migration route through the lower river, where the prior on ψ_{12}^{AC} was uniform between zero and one [U(0,1)], and the effects of discharge and rearing history were estimated as (Equation 3.9):

$$\text{logit}\left(\widehat{\psi}_{12}^{\text{AC}}\right) = \ln\left(\frac{\psi_{12}^{\text{AC}}}{1 - \psi_{12}^{\text{AC}}}\right) + \sum_{j=1}^{k=2} \gamma_j \times \beta_j \times X_{ij} \ldots \gamma_k \times \beta_k \times X_{ik}$$

Rearing history

I extended the base model to estimate mean survival for hatchery- and wild-reared fish across years to assess influence of rearing history (Figure 3.3). Because I was unsure of how survival might vary, I specified separate slopes and intercepts for survival probability in each reach. Based on consideration of model size and estimability of parameters, I did not include environmental covariates in this ‘rearing model’, with the exception of detection covariates (tag and discharge, Equation 3.4). The prior distributions for survival probabilities in the base model were therefore modified to allow independent estimation of survival for hatchery and wild fish (i.e., random group effect). This was accomplished by incorporating a random group effect for each estimated probability.

Tracking annual survival

The efficacy of management and conservation activities within the Penobscot River to increase smolt survival was assessed by extending the base model to estimate survival separately for each year (Figure 3.3). I included acoustic tag model and discharge as covariates in the detection model for this ‘annual model’, and estimated detection across years because I did not foresee sources of detection heterogeneity other than these variables. I compared survival in years preceding management actions to survival in years following those actions. I examined effects of three types of management: 1) dam removals at Great Works Dam (2012) and Veazie Dam (2013; 2) turbine shut downs during the smolt migration at Howland (2010–2014), Great Works (2010–2012), and Veazie Dams (2010–2013); and 3) increases in hydropower generation at Milford (2012–2014), Orono (spring 2013–2014), and Stillwater Dams (2013–2014).

RESULTS

Multi-annual trends in survival and use of Stillwater Branch

Detection probability was higher for smolts tagged with the larger (model V9) acoustic tags than the smaller (model V7) tags (Table 3.4). The probability of detecting the larger tag was 73% (95% CRI = 71–76%) greater than the smaller tag. Regardless of tag type, the probability of detection decreased with increasing discharge. Over the range of discharges (212– 2,164 $m^3 \cdot s^{-1}$), detection probability decreased from 0.81(0.80–0.82) to 0.028 (0.023–0.034).

Survival of smolts in the free-flowing (i.e., unimpounded) reaches of the river was high across years (> $0.99 \cdot km^{-1}$). Survival through reaches containing dams was notably lower than survival through unimpounded reaches or reaches that contained head ponds (Figure 3.4). Mortality through some reaches containing dams was as much as five times loss through unimpounded river sections (Appendix A).

Table 3.4. Covariate effects on survival, detection, and state-transition probabilities in freshwater. Mean, 95% credible intervals (CRI) and parameter inclusion probabilities (PIP) for each of the covariates used to model detection probability, apparent survival, and the probability of transitioning into the Stillwater Branch from the main-stem Penobscot River ($\boldsymbol{\psi_{12}^{AC}}$) during migration through the lower river by *Salmo salar*.

Parameter modeled	Covariate	Mean	S.D.	2.5%	97.5%	PIP
Detection (*p*)	Tag Model	1.007	0.066	0.878	1.139	1.000
	Discharge	-0.988	0.026	-1.039	-0.937	1.000
Survival (*S*)	Discharge	1.392	0.086	1.224	1.565	1.000
	Discharge^2	-0.536	0.037	-0.609	-0.463	1.000
	ATU	0.182	0.077	0.030	0.333	0.425
	ATU^2	0.064	0.036	-0.006	0.134	0.317
	Photoperiod	-0.977	0.086	-1.147	-0.808	1.000
	Photoperiod^2	-0.001	0.030	-0.059	0.058	0.129
	Temperature	1.676	0.082	1.514	1.837	1.000
	Temperature^2	-0.279	0.020	-0.319	-0.240	1.000
Transition to Stillwater (ψ_{12}^{AC})	Discharge	0.159	0.090	-0.190	0.336	0.630
	Rearing history	-0.282	0.238	-0.741	0.198	0.183

Mean probability of using the Stillwater Branch was 0.113 (0.096–0.131) among years. Use of the Stillwater Branch increased with increasing discharge. Although the 95% CRI for this effect overlapped zero, there was a relatively high probability (0.62) that the effect of discharge was included in the best model (Table4). Conversely, there was little support for differential use of the Stillwater Branch between rearing histories (inclusion probability = 0.18, Table 3.4).

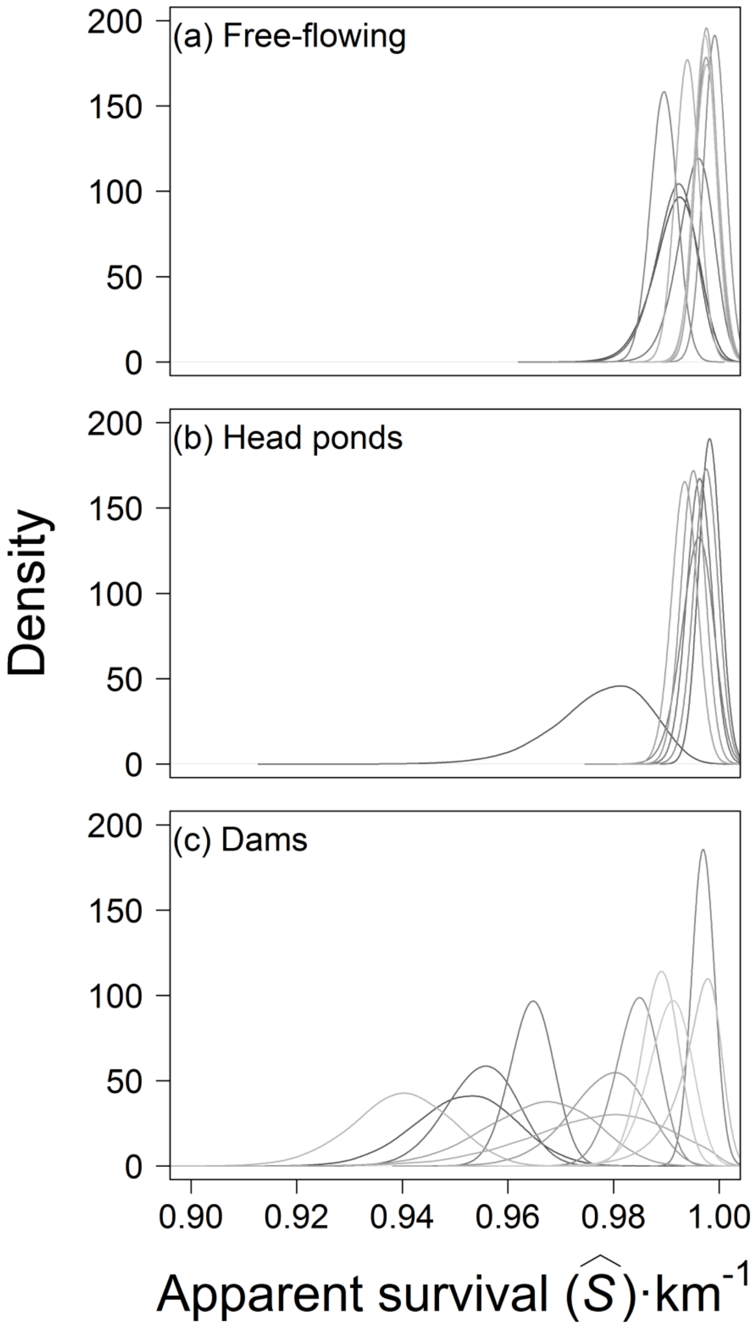


Figure 3.4. Freshwater survival by reach type. Shown are density plots for posterior distributions of estimated survival in (a) reaches that do not contain impoundments or head ponds, (b) reaches that contained head ponds, and (c) reaches of the river that contained dams.

There was high cumulative mortality during migration from the most upstream reaches of the catchment to the estuary. Cumulative survival to the mouth of the estuary was 0.453 (0.416–0.490) for fish emigrating from the upper Penobscot River (rkm 165), and was 0.480 (0.443–0.515) for fish emigrating from the upper Piscataquis River (rkm 187). Much of this loss occured through reaches of the freshwater system associated with dams that, in terms of coverage in the system, represent a minority of the study river (Figure 3.4). As a frame of reference, the reaches containing dams in the Penobscot River catchment accounted for about 32 rkm of the more than 250 rkm through which survival was estimated in the present study, or about 15% of the total study system.

When survival through intervals containing dams and head ponds was replaced with mean survival through free-flowing intervals, cumulative survival of smolts from the Piscataquis River was 0.609 (0.555–0.661), and cumulative survival of smolts from the upper main-stem of the Penobscot River was 0.600 (0.539–0.657). These results indicate a decrease in cumulative survival probability of 0.12 (20% reduction) for smolts from the Piscataquis River, and a decrease in cumulative survival probability of 0.15 (25% reduction) for smolts from the main-stem Penobscot River due to the effects of dams and head ponds associated with dams.

Based on comparisons of survival from wild- and hatchery-reared smolts, there was little difference in survival among rearing histories (Appendix B). In general, the trend in survival estimates for hatchery and wild fish was similar (Fig 5). However, in the Stillwater Branch, where all reaches contained dams, survival of hatchery-reared fish was similar to free-flowing reaches while survival of wild fish was lower at two of the dams (Figure 3.5).

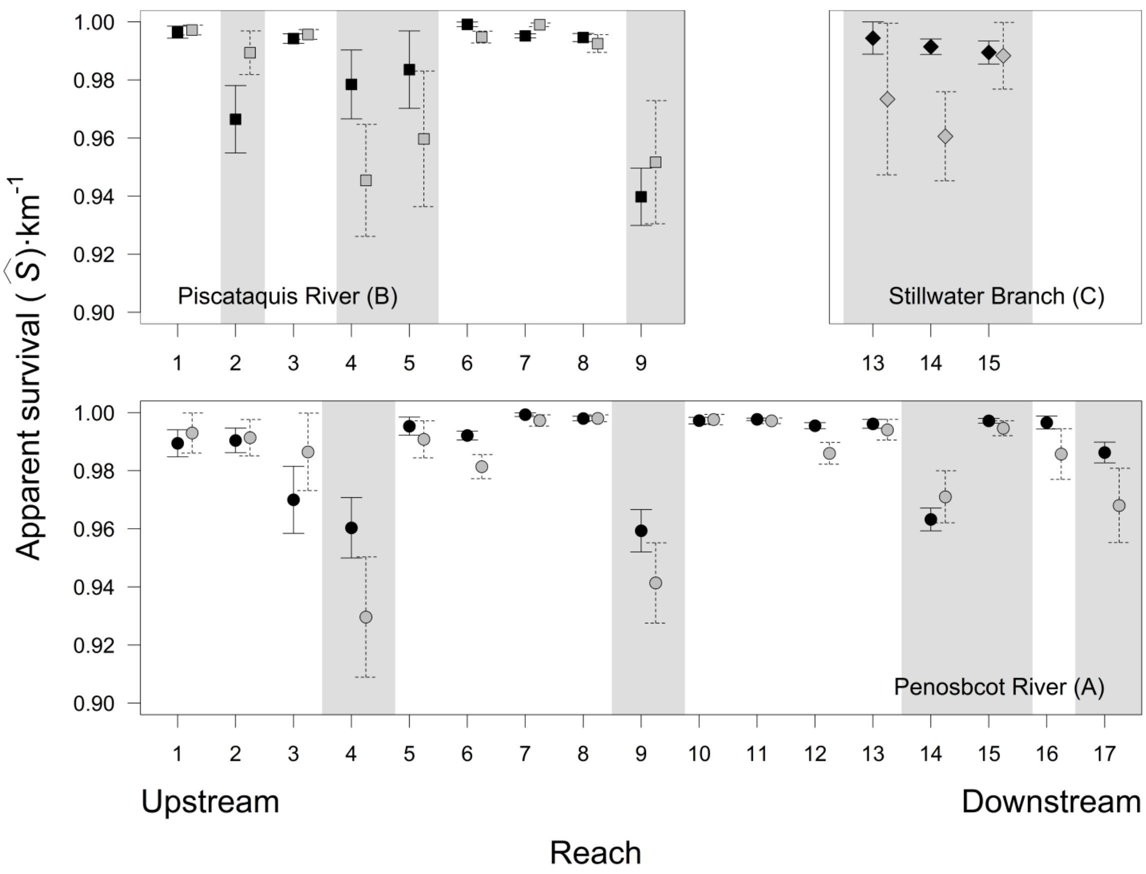


Figure 3.5. Freshwater survival of wild and hatchery-reared smolts. Mean (± S.D.) estimated apparent survival (per km) of hatchery- (black) and wild-reared (gray) *Salmo salar* smolts through discrete reaches of the Penobscot River catchment in all years 2005–2014 from release locations to the head of tide in the Penobscot River (circles, state A), the Piscataquis River (squares, state B), and the Stillwater Branch (diamonds, C). Reach numbers and states correspond to parameters in the model schematic (Figure 3.2) as well as those in Appendix B. Gray panels indicate reaches containing dams.

Environmental influences on survival

Smolt survival increased with increasing discharge until about 1,200 m^3·s^{-1}(Figure 3.6a), but decreased at discharges higher than that. Over the range of observed discharge (212– 2,164 m^3·s^{-1}) survival increased from 0.034 (0.032–0.036) at the lowest discharge to 0.712 (0.649–0.790) at intermediate discharge of about 1200 m^3·s^{-1} (Figure 3.6a). The estimated regression coefficients for the first- and second-order terms used to estimate effects of discharge on survival both excluded zero, suggesting that both were important predictors of survival (Table 3.4). Inclusion probabilities for these terms also indicated both were supported (Table 3.4).

The thermal regime (ATU) experienced by *S. salar* smolts prior to tagging was positively related to survival (Figure 3.6b). Fish that experienced the greatest ATU (warmer development period) had 47% higher survival (mean = 0.686, 95% CRI = 0.510-0.822) than fish experiencing the lowest ATU (mean = 0.467, 95% CRI = 0.449–0.485). Although the 95% CRI of the coefficient for linear effect of ATU did not overlap zero, the probability that the variable was included in the best model was less than 0.50 (Table 3.4). The 95% CRI for the quadratic term overlapped zero, and there was a low probability of inclusion for the quadratic term (0.32), suggesting that a quadratic effect of ATU on survival was not important (Table 3.4).

I found strong evidence for a relationship between smolt survival and mean water temperature experienced during migration (Figure 3.6d). Survival was lowest (approximately zero) at temperatures below 5 °C, after which survival increased rapidly until reaching 0.93 (0.86–0.97) near 12 °C. Survival remained high with increasing temperature from about 12 °C to about 19 °C, after which smolt survival began to decrease again (Figure 3.6d). Both the linear and quadratic effects of temperature were strongly supported based on lack overlap of coefficients with zero, and the high probability that both terms were included in the model (Table 3.4).

Smolts migrating later in the year (photoperiod) had lower survival than those migrating earlier (Figure 3.6c). Survival decreased from 0.865 (0.853–0.878) in the earliest part of the smolt run to about 0.044 (0.015–0.123) in the latest part of the run

(Figure 3.6c). The estimated coefficient for the quadratic effect of photoperiod on survival was essentially centered at zero, and the 95% CRI overlapped zero considerably (Table 3.4). The inclusion probability for the quadratic effect (0.129) indicated low probability that the term was included in the model (Table 3.4). Conversely, both the 95% CRI of the regression coefficient and the inclusion probability indicated strong support for inclusion of the linear term (Table 3.4).

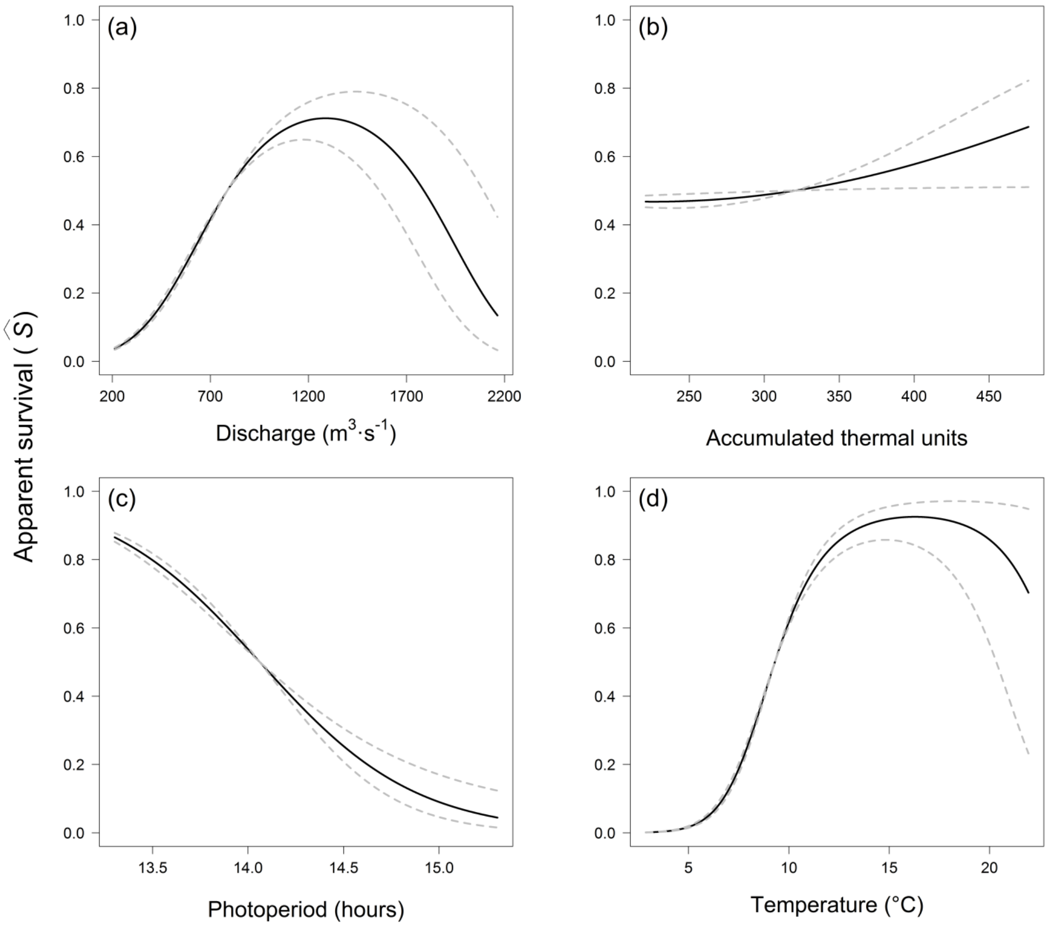


Figure 3.6. Environmental influences on survival. Shown are relationships between environmental covariates and apparent survival of *Salmo salar* smolts in the Penobscot River, Maine, USA showing effects of a) discharge, b) accumulated thermal units from 1 January to release date, c) photoperiod (day length), and d) water temperature in the river during migration.

Changes in annual survival following management actions

The removal of Great Works Dam in 2012 and Veazie Dam in 2013 increased smolt survival, but the increase was small. The result was an increase of 0.005 in smolt survival following removal of Great Works Dam, and an increase of 0.014 following removal of Veazie Dam (Figure 3.7).

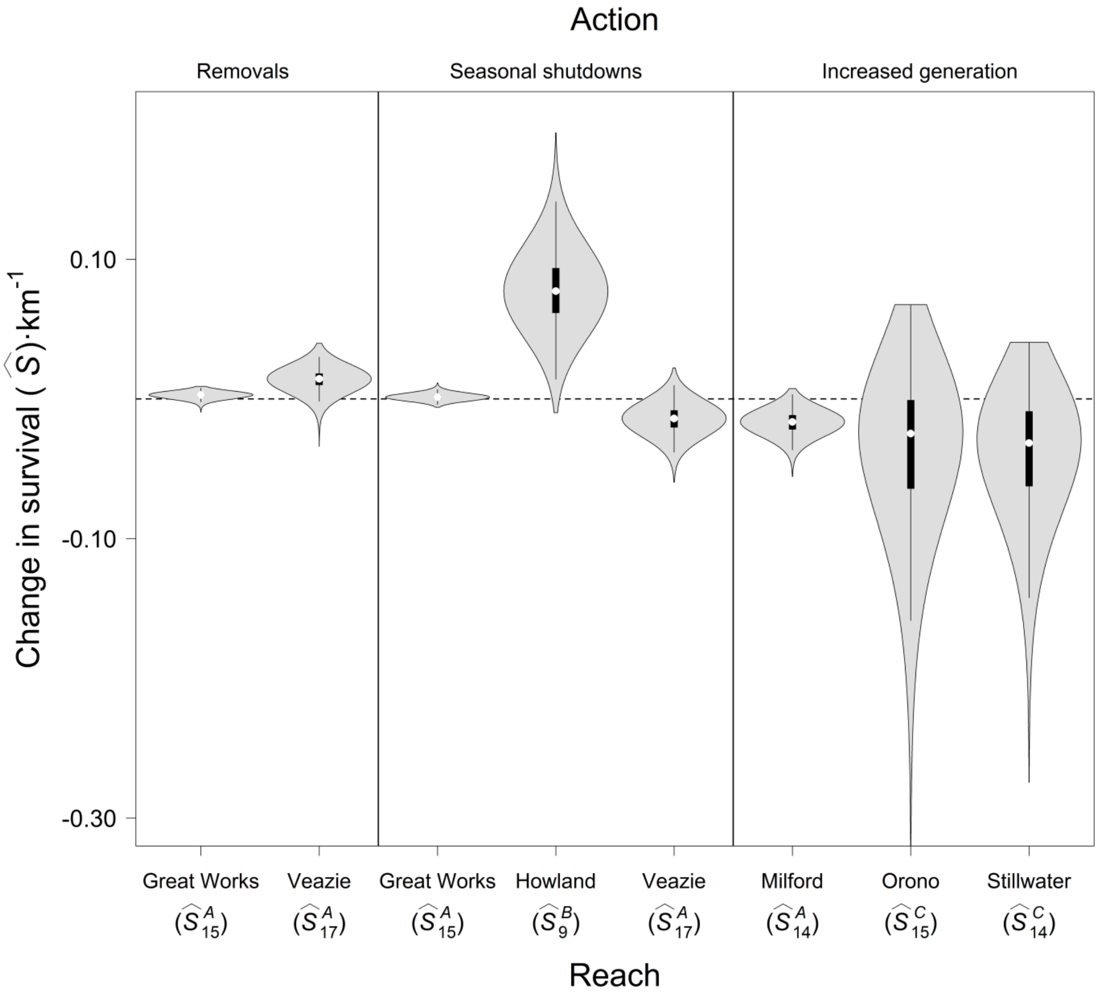


Figure 3.7. Changes in freshwater survival following changes in hydro system. Estimated mean (95% CRI) change in survival of *Salmo salar* smolts through impacted reaches following three different types of changes (dam removal, seasonal turbine shutdowns during the smolt run, or increased power generation) to dams in the Penobscot River catchment (GW = Great Works, HD = Howland, MD = Milford, SW = Stillwater, and VZ = Veazie). Survival parameters in parentheses correspond to parameters in the model schematic (Figure 3.2) as well as those in Appendix C.

Seasonal shutdowns of hydropower generation at three facilities in the Penobscot River were varied in efficacy. At Howland Dam, a marked increase in smolt survival (+ 0.078) coincided with shutdowns in 2010 (Figure 3.7). However, shutdowns at Veazie (2010–2013) and Great Works Dams (2010–2012) were more ambivalent. Minimal change in survival followed turbine shutdown at Great Works Dam (+ 0.001), but survival at Veazie Dam appeared to decrease (- 0.014) slightly following implementation (Figure 3.7).

Changes to survival also were minimal but negative (- 0.017) at Milford Dam following increases in head pond height and addition of two turbines (Figure 3.7). Survival also decreased following construction of two new powerhouses (2013) and increased generation (2014) at Stillwater (- 0.040) and Orono Dams (- 0.039) in the Stillwater Branch. However, when survival was estimated separately for each year in the Stillwater Branch, there was a high degree of uncertainty in the estimates for most years prior to 2013 based on the low probability of using that migratory route (Appendix C).

DISCUSSION

Effects of dams and changes in the hydro-system

Dams remain the single largest impediment to successful migration of *S. salar* in freshwater systems throughout the world (Parrish et al. 1998). In the Penobscot River, survival through dams was reduced relative to free-flowing reaches of the system, resulting in five times greater mortality at some facilities when compared to free-flowing reaches. Mortality at dams can occur as a result of increased exposure to predators through migratory delay (Poe et al. 1991; Keefer et al. 2012) or physical injury during passage (Stier and Kynard 1986; Mathur et al. 2011). Smolts can incur delayed mortality from dam passage due to physical injuries (Music et al. 2011) that impair osmoregulatory ability in estuaries (Zydlewski et al. 2010) where predation is high (Hawkes et al. 2013), resulting in dam-related estuary mortality (Budy et al. 2002; Schaller et al. 2012; Stich et al. in review).

The cumulative probability of survival (0.45) of *S. salar* smolts during emigration from headwaters to the estuary in the Penobscot River demonstrates that a large proportion of this population is lost during the freshwater phase of the smolt migration each year. Much of the mortality incurred by smolts in the river is incurred through those short river reaches that contain dams. The passage of dams in this system results in a cumulative decrease of 0.15 in the probability of smolt survival compared to what might be expected in a free-flowing system (0.60). Dams contribute 30% of the total mortality incurred during this freshwater migration. These results indicate that ongoing recovery and management activities (such as dam removal and improvements to fish passage) continue to have demonstrated potential to increase the number of fish entering the estuary.

A growing body of evidence demonstrates the utility of assessing proposed changes to hydro systems at both catchment and local-project scales to balance multiple uses of river systems (Ziv et al. 2012; Null et al. 2014). Ongoing management and restoration activities in the Penobscot River have the potential to increase survival of smolts during freshwater and estuary migration through dam removal and seasonal turbine shutdowns (Figure 3.7). The removal of main-stem dams in the river increased the per-kilometer rate of smolt survival through the lower river. Because survival was previously high around these facilities (Holbrook et al. 2011; Stich et al. 2014), the increases in survival at Great Works and Veazie Dams to levels seen in other free-flowing river reaches suggests that dam removal has some benefit, even if modest. As the lower-most dam, all smolts passed Veazie, so any change at this facility demonstrates the potential to enhance recovery of *S. salar* (a change of 0.01 in survival translates to a difference of several thousand fish).

Seasonal turbine shutdowns and increases in hydropower generation had dam-specific effects on smolt survival (Figure 3.7), but there are some general trends that could inform seasonal management of hydropower and site-location for changes to generation in the future. At dams where survival was already high (Great Works and Veazie), turbine shutdown had little (and mixed) influence on smolt survival. However, where survival was low without shutdowns I observed increases in survival concurrent with this action. Prior to seasonal turbine shutdowns, the probability of survival through the reach containing Howland Dam (range = 0.75–0.92) was among the lowest in the entire catchment (Appendix C). Concurrent with seasonal turbine shutdowns, survival through the reach containing Howland Dam increased such that it now surpasses survival at small dams further upstream in the Piscataquis River.

Increases in hydropower generation, indicated dam-specific responses by smolts, as was the case for the response to seasonal turbine shutdowns. At Milford Dam, survival was historically low relative to free-flowing reaches and other reaches containing dams (Holbrook et al. 2011; Stich et al. 2014). Smolt survival exhibited little change following increases in generation at this facility concurrent with head pond increase and addition of two turbines. Conversely, survival at Stillwater and Orono Dams, where survival historically was high (Holbrook et al. 2011; Stich et al. 2014) decreased by 4% at each facility following the addition of a new powerhouse (Figure 3.7). Based on the above results, it is evident that the greatest increases in survival through dam removal and seasonal turbine shutdowns can be achieved at sites where survival is reduced relative to free-flowing reaches. Conversely, the greatest reductions in survival resultant from increases in hydropower generation can be expected at facilities through which survival is high.

My results demonstrate that the number of smolts entering the marine environment could be increased through specific stocking strategies. By stocking below dams in the Penobscot River (as was done in 2014), the number of smolts entering saltwater is expected to increase by a minimum of about 55% through avoidance of mortality at dams and within free-flowing reaches of the river. This does not include reductions of up to 40% in estuarine and/or early marine mortality that otherwise result from delayed effects of dam passage (Budy et al. 2002; Schaller et al. 2012; Stich et al., in review). The number of smolts exiting the Penobscot Estuary previously has been related to the number of adult returns (Sheehan et al. 2011), so this gain is expected to translate directly to an increased number of returning adult *S. salar*. There are potential implications of these management actions on the imprinting and homing of *S. salar*, but adult salmon need only reach the main-stem of the Penobscot River (after which they are taken for captive breeding), and straying of adults is offset by increases in smolt-to-adult survival (Gunnerød et al. 1988). Such a stocking strategy may provide a means of increasing relative adult returns to the Penobscot River in the face of low marine survival until other conservation measures can be put in place in the estuary or marine environments (Hansen et al. 2012).

Environmental variability in smolt survival

I was able to identify important environmental influences on *S. salar* smolt survival in freshwater that can be directly incorporated into the decision-making framework for smolt stocking and the regulatory framework for hydropower dams with respect to smolt passage. Smolt survival was highest early in the run each year, at intermediate discharges, and at temperatures between 10 °C and 20 °C (Figure 3.6). Previous studies have shown that salmon smolts stop moving at temperatures above 20 °C in laboratory experiments (Martin et al. 2009; Zydlewski, Stich and McCormick 2014), and may be considered losses due to high overwinter mortality (Horton et al. 2009). In the future, survival could be optimized with respect to temperature by informing decisions about when to stock hatchery-reared smolts.

Low survival of Pacific salmon smolts also has been observed at high temperatures in freshwater (Newman and Rice 2002). Pacific salmon smolt survival also has previously been shown to increase with increasing discharge (Kjelson and Brandes 1989; Newman 2003), but those studies used only flows well below the 50th percentile observed in the present study, even though rivers in which this trend has been documented (e.g. Columbia and San Joaquin Rivers) are substantially larger than the Penobscot River. This difference is due, in large part, to intensive regulation of the quantity of water that is diverted for human uses in those systems (see Kjelson and Brandes 1989) compared to the Penobscot River, in which most dams are operated as ‘run-of-river’ (National Marine Fisheries Service 2012a,b). However, the data suggest an upper threshold to this relationship, after which further increases in flow reduce survival. Thus, my results have uncovered some of the complexities of environmental influences on salmonid smolt survival that may previously have gone unnoticed in other systems.

Rearing history and implications for conservation hatcheries

The similarity in survival trends among rearing histories (Figure 3.5) suggests that the actual rearing of fish in hatcheries apparently has little influence on survival of out-migrating smolts in the freshwater reaches of this system. Because the majority (*c*. 90%) of smolts leaving the Penobscot River Estuary results from hatchery stocking of smolts (Sheehan, Renkawitz and Brown 2011), trends in freshwater survival of hatchery-reared smolts likely will be representative of broader population trends until wild smolt production increases. Based on similarities, hatchery-reared smolts provide a useful surrogate for the study of smolt survival in lieu of wild-reared smolts. Wild-reared *S. salar* smolts are a precious commodity in most rivers based on multi-decadal lows in adult returns (U.S. Atlantic Salmon Assessment Committee 2014). Ultimately, increasing natural reproduction and rearing are the goal of most recovery plans for the species (*see* National Marine Fisheries Service and U.S. Fish and Wildlife Service 2005). As such, any limitation to trapping, handling, and otherwise interrupting migration of wild smolts can only benefit species recovery.

I do not suggest that hatchery and wild smolts in the Penobscot River or other systems are fully exchangeable or even equivalent in terms of survival, long-term marine performance, or other life-history aspects. In fact, a large number of other studies have documented differential performance of hatchery- and wild-reared smolts globally (e.g. Jonsson et al. 2003; Saloniemi et al. 2004; Jokikokko et al. 2006), and wild fish generally are found to out-perform hatchery fish in a variety of ways. The similarity in survival among rearing histories in the Penobscot River likely reflects the artificial nature of high mortality associated with dam passage. This result indicates that the cause of mortality from dams is not being mitigated by phenotypic responses of wild-reared smolts, and that active management of hydro-systems for maintenance of these stocks will likely be required in the future.

Resource managers may target optimal release timing of hatchery-reared smolts using knowledge about relationships between survival, environmental conditions, and hydropower operations in the system. The findings of this study could be integrated with information about onset of migratory behavior and freshwater movement rates of *S. salar* smolts in the Penobscot River (Stich et al., unpublished), and information about smolt physiology, the timing of estuary arrival, and early marine survival in this system (Stich et al., in review) to produce decision models to assist with decisions about hatchery supplementation of this smolt run in the future. Moving into the future, such a synthesis has the potential to assist in the management and recovery of critically endangered *S. salar* stocks throughout the world in the form of a support tool.

The model developed in this study provides a standardized approach for assessing changes to *S. salar* smolt survival in the Penobscot River in the future and can be modified for use with other species or in other systems. By standardizing locations used for estimating survival among years, and by standardizing survival estimates within those reaches as per-kilometer rates, the methods used in this study allowed for direct comparison of survival among reaches and years within the catchment. I was able to compare survival between rearing histories of *S. salar* smolts throughout the catchment, and identify environmental influences on smolt survival across years. I also was able to compare changes in survival from year to year associated with management and conservation activities catchment-wide and within specific tributaries of the Penobscot River. As such this framework offers a means to target potential restoration activities (shutdown periods, bypasses, and dam removals) and assess whether they meet their intended goals.

CHAPTER 4

LINKING BEHAVIOR, PHYSIOLOGY, AND SURVIVAL OF ATLANTIC SALMON SMOLTS DURING ESTUARY MIGRATION

INTRODUCTION

The links between the freshwater experience of anadromous fishes and marine survival are poorly understood in general (McCormick et al. 2009). Information about these relationships could have timely implications for the management and conservation of fisheries. Elevated marine mortality in recent years is thought to be a driver behind the failure of many endangered Atlantic salmon *Salmo salar* populations to recover (Chaput et al. 2005; Chaput 2012; Miller et al. 2012; Mills et al. 2013; Lacroix 2014), due in large part to changes in ocean climate (Friedland et al. 2003; Mills et al. 2013; Friedland et al. 2014). However, high mortality during migration through freshwater and estuarine corridors (Thorstad 2012a; Hayes and Kocik 2014) also likely contributes to reduced population sizes (Parrish et al. 1998). Estimates of marine survival also often include estuarine mortality due to difficulty in separating these processes (Friedland 1998). A better understanding of factors that influence estuary mortality could help to enhance management of Atlantic salmon stocks in the face of changing ocean climates (Mills et al. 2013). Despite the importance of estuarine habitats during migration, little is known about Atlantic salmon smolt behavior and survival in North American estuaries compared to freshwater and near-shore marine environments (Weitkamp et al. 2014).

The transition of Atlantic salmon smolts to saltwater is recognized as a period of high mortality in estuaries (Lacroix 2008; Kocik et al. 2009; Dempson et al. 2011) and fjords (Gudjonsson et al. 2005; Svenning et al. 2005; Thorstad et al. 2012a). This period is marked by high predation risk (Hvidsten and Lund 1988; Kocik et al. 2009; Hawkes et al. 2013), physiological stresses (Handeland et al. 1997), and novel environmental conditions (McCormick et al. 1998). In response to these challenges, smolting involves a synchronous suite of changes in physiology, morphology, and behavior that, in concert, enhance the probability of successful saltwater entry (McCormick et al. 1998). Therefore, seasonal timing of smolt runs is important for smolt survival during saltwater entry (McCormick et al. 1998; Thorstad et al. 2012a).

Smolt survival during estuary passage is theoretically maximized by environmental conditions, along with the presence or absence of predators (Kocik et al. 2009) and sympatric prey buffers (Svenning et al. 2005) during a brief annual period of weeks, characterizing the ‘ecological smolt window’ (McCormick et al. 1998). To match this ecological window, smolts experience a period of peak physiological preparedness for saltwater entry, the ‘physiological smolt window’ (McCormick et al. 1998). Gill Na^+, K^+-ATPase (gill NKA) activity in Atlantic salmon is one enzyme that has been found to be a useful indicator of smolt development and preparedness for saltwater entry (Zaugg 1982; McCormick et al. 1987, 1989). During smoltification, gill NKA activity peaks during spring, resulting in a period of increased saltwater tolerance (Duston and Saunders 1990; McCormick 2013). As a result, smolts transferred to salt water have increased gill NKA activity, osmoregulatory ability, and survival relative to parr in laboratory settings (McCormick et al. 2003). Evidence relating smolt survival in the wild to gill NKA activity is conspicuously absent from this body of work, and long term performance is not clearly linked to higher gill NKA activity (Zydlewski and Zydlewski 2012).

The natural timing of estuary arrival, movement rates through estuaries, and ultimately smolt survival during estuary migration are likely affected by the presence of dams. Estuarine mortality that occurs naturally from causes such as predation and physiological challenges (Handeland et al. 1997; Blackwell et al. 1997; Halfyard et al. 2013) can be exacerbated by anthropogenic influences such as passage of dams in freshwater. This might occur through migratory delay (Keefer et al. 2012), increased predation (Poe et al. 1991; Blackwell and Juanes 1998) and physical injuries (Stier and Kynard 1986; Mathur et al. 2011) that can result in physiological impairment (Zydlewski et al. 2010) and reduced survival during estuary passage. However, the presence and magnitude of dam-related estuary mortality (i.e. delayed effects of dams) during estuary passage by Atlantic salmon remains uncertain (Stich et al. 2014). Information about dam-related estuary mortality could have important implications for recovery activities involving Atlantic salmon stocks, including dam removals such as those occurring in the Kennebec and Penobscot Rivers in Maine (Day 2006).

The goal of this study was to quantify movement behavior and survival of Atlantic salmon smolts in the Penobscot River Estuary, Maine, relative to potential drivers of these population characteristics. I classified these drivers into three main categories: 1) individual fish characteristics (fork length, mass, condition factor, and rearing history), 2) migratory history (number of dams passed, release distance from ocean, migratory route), and 3) environmental variability (cumulative temperature experienced, discharge, and photoperiod). My first objective was to relate estuary arrival date and movement rate of smolts to individual fish characteristics, migratory history, and environmental variability from 2005 through 2013. The second objective of this study was to estimate survival of Atlantic salmon smolts during migration through the estuary from 2005 through 2013, using Cormack-Jolly-Seber (CJS) mark-recapture models and quantify effects of movement behavior (estuary arrival date and movement rate), individual fish characteristics, migratory history of fish, and environmental variability on survival.

METHODS

Study site

The Penobscot River (Figure 4.1) is the largest watershed within Maine, encompassing approximately 22,000 km^2. Atlantic salmon have been stocked throughout the watershed at egg, fry, parr, and smolt life-stages during the last several decades, and limited wild spawning occurs (USASAC 2014). Exact estimates of the proportion of the Penobscot River smolt run made up by stocked fish is not known, but sampling in the bay suggests that more than 90% of the run results from smolt stocking (Sheehan et al. 2011). As a result, the majority of the adult run (*c*. 83%) is made up of hatchery-stocked smolts (USASAC 2012). Hatchery stocking generally occurs less than 160 river kilometers (rkm) above the mouth of the estuary in the Penobscot River and its tributaries (see Figure 4.1). The peak of the smolt emigration from wild rearing sites occurs between late April and early May most years (USASAC 2014).

The migratory history of smolts in the Penobscot River depends on rearing history, stocking locations, migratory routing through the lower river, and inter-annual changes to hydropower operations throughout the catchment. All smolts stocked or reared in the upper reaches of the catchment enter the main-stem of the Penobscot River at river kilometer 100, either by passing Howland Dam (Figure 4.1, G) from the Piscataquis River or by passing West Enfield Dam (Figure 4.1, H) from the Upper Penobscot River. Upon reaching the lower freshwater portion of the Penobscot River (60 rkm from the mouth of the estuary), smolts can use one of two migratory paths around a large island: the main-stem Penobscot to the east (88% of smolts), or the Stillwater Branch (12 % of smolts) to the west (Figure 4.1).

On the east side of the island (main-stem Penobscot) smolts passed two dams until the removal of the Great Works Dam (Figure 4.1, E) in 2012. Concurrently, hydropower production was increased at two dams (Stillwater and Orono) in the Stillwater Branch to the west. The details of changes to operations of the Stillwater (Figure 4.1, B) and Orono (Figure 4.1, C) Dams are described in Stich et al. (2014). Briefly, hydropower generation was approximately doubled at each of these two dams following the addition of a second powerhouse at each facility in spring 2014. On the west side of the island (Stillwater Branch), smolts pass three dams through which survival was high relative to the dams in the main-stem during 2005–2013 prior to changes in hydroelectric generation. This resulted in a cumulative difference in survival of 10% between passage routes around the island (Holbrook et al. 2011; Stich et al. 2014). Finally, smolts traveling either route would then pass the Veazie Dam, at the head of tide, until 2013 when that dam was removed. Dependent upon stocking location, migratory route through the Lower Penobscot River, and year of stocking (because of Great Works Dam removal), smolts stocked in freshwater may have passed 2–9 dams before entering the estuary. This study occurred prior to the removal of Veazie Dam (Figure 4.1, F).

The Penobscot River Estuary spans 45 km from the mouth of the estuary to the head of tide, which coincides with the former location of Veazie Dam (Figure 4.1, F). The upper estuary (from p_1 to p_6 in Figure 4.1) is tidally influenced but uniformly fresh water (Imhoff and Harvery 1972), while the middle estuary (between p_6 and p_{10} in Figure 4.1) is characterized by mixing of fresh and salt water (Seiwell 1932; Stich et al., in review), and the lower estuary is physically and chemically stratified (Imhoff and Harvey 1972).

Acoustic receiver array

An array of stationary VR2 and VR2-W acoustic receivers (Amirix Vemco Ltd., Halifax, Nova Scotia) was deployed in the Penobscot River, Estuary, and Bay prior to the start of the Atlantic salmon smolt run each year of the study. Deployment was coordinated collaboratively between the U.S. Geological Survey (USGS) Maine Cooperative Fish and Wildlife Research Unit, the University of Maine, and National Oceanic and Atmospheric Administration (NOAA; Figure 4.1). All receivers monitored continuously on a frequency of 69 kHz using omnidirectional hydrophones, and were moored to the bottom of the estuary on cement anchors. Where necessary, multiple receivers were deployed in a single location to achieve coverage over the entire width of the channel. Detections at all receivers within a discrete cross section were pooled as a single site for data analysis. A total of 11 acoustic receiver locations within the estuary (and several in the bay) were common to all arrays of the present study (2005–2006, 2009–2013). During all years, fish detections at all (about 40) receivers in the bay (downstream of Fort Point: p_{10} in Figure 4.1) were pooled as a single, final detection event for all analyses.

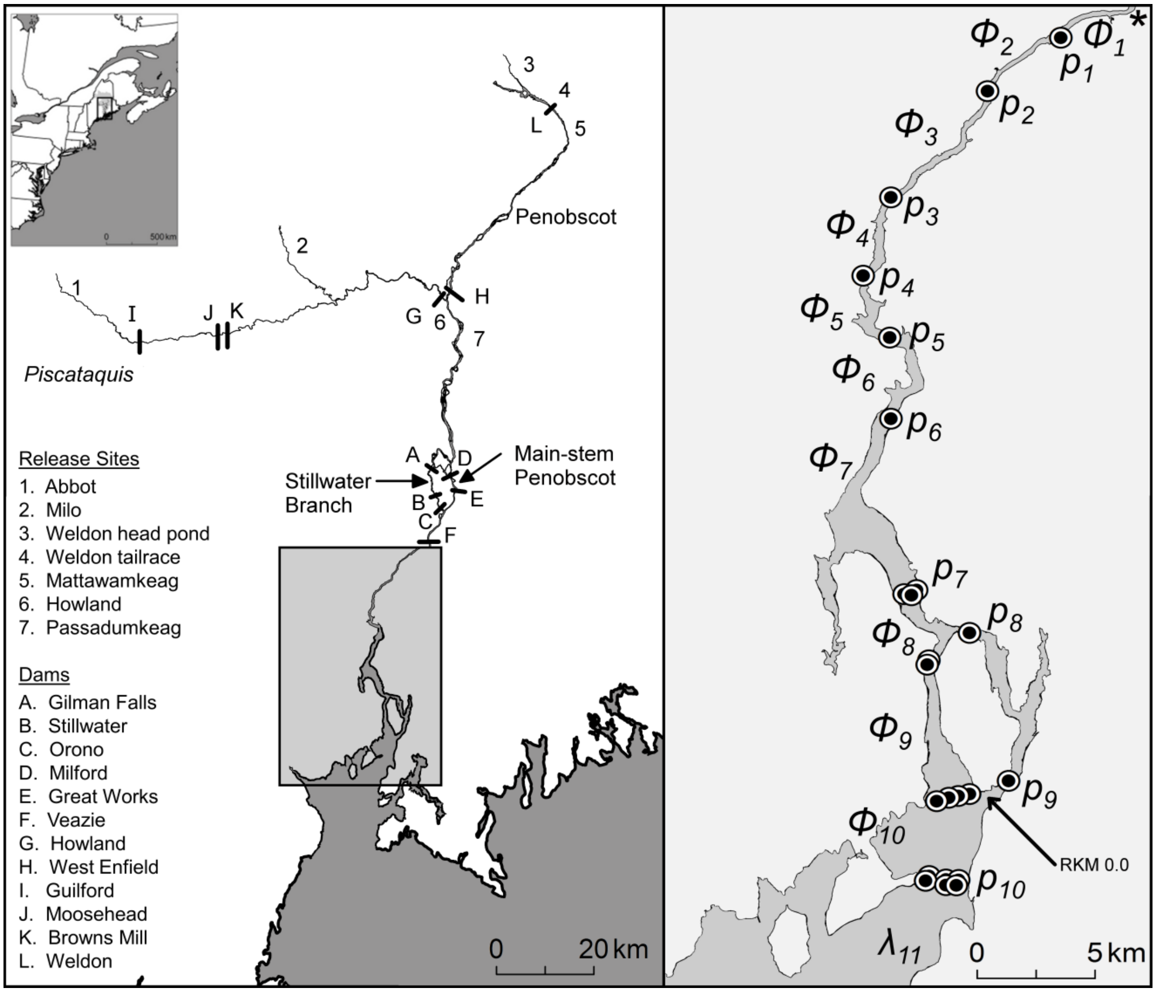


Figure 4.1. Acoustic telemetry array used for assessing estuary behavior and survival. Map of the Penobscot River Watershed in Maine, USA showing location in North America (top left inset), and locations of tributaries, dams, and release sites in the Penobscot River (left panel). The right panel shows locations of acoustic receivers used to detect Atlantic salmon smolts in the estuary. Parameters associated with Cormack-Jolly-Seber (CJS) survival models are: detection probability at each location following release (p_t), apparent survival within reaches between locations (ϕ_t), and λ_{11} (product of ϕ_{11} in final reach and p_{11} at final receiver location (40 receivers in bay, not shown). The symbol * indicates point of virtual release in the estuary for CJS models.

Fish measurements, acoustic tagging, and releases

From 2005 through 2013 a total of 1,824 Atlantic salmon smolts was acoustically tagged (Table 4.1) and released throughout the Penobscot watershed in fresh water (Figure 4.1). Of these fish, 941 fish were later relocated during estuary migration and used in this study, including 800 hatchery-reared smolts from the U.S. Fish and Wildlife Service (USFWS) Green Lake National Fish Hatchery (GLNFH), and 141 wild-reared smolts. Acoustic tagging methods were described in detail by Holbrook et al. (2011) and Stich et al. (2014). Identical procedures were used in all years of the present study (2005–2013). Briefly, individual smolts were anaesthetized using a 100 $mg \cdot L^{-1}$ solution of MS-222 buffered to pH 7.0 (using 20-mmol $NaHCO_3$), fork length ($L_{F;}$ mm) and their mass (g) was measured.

For each smolt, a 1-cm incision was made offset from the ventral line and 1-cm posterior to the pectoral fin girdle. An acoustic tag was inserted intraperitoneal and the incision was closed with two simple, interrupted knots using 4-0 absorbable vicryl sutures (Ethicon, Somerville, New Jersey, USA). Model V7-2L (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) tags were used in 2005, as well as for wild-origin fish tagged in 2011. Expected battery life of V7-2L tags was 80 days during 2005, and 69 days in 2011. In all other years, I used model V9-6L acoustic transmitters (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada) with expected battery life of 82 days (except during 2006 when battery life of V9-6L transmitters was 80 days). Model V7 tags were 7 mm in diameter, 18.5 mm long, and weighed 1.6 g in air (0.75 g in water), while model V9 tags were 9 mm in diameter, were 20 mm long , and weighed 3.3 g in air (2.0 g in water).

Table 4.1. Descriptive statistics for acoustically tagged fish used in assessment of estuary behavior and survival. Release sites, river kilometer of release sites (rkm), rearing history (Origin), number (n), as well as mean and standard deviation (SD) of fork length (L_F, mm), gill Na^+, K^+-ATPase activity (gill NKA; μmol ADP·mg protein^{-1}·h^{-1}), and mass (g) of Atlantic salmon smolts acoustically tagged and released throughout the Penobscot River and Estuary 2005–2013. Summary statistics for L_F, Mass, and gill NKA are for fish used in the current study. The number in parenthesis under 'n' indicates total number originally released in each group.

Year	Release site	rkm	Origin	L_F	Mass	gill NKA activity	n
2005	Howland	99	Hatchery	189 (11)	75 (15)	6.58 (1.95)	90 (150)
	Mattawamkeag	144	Hatchery	185 (12)	69 (15)	6.07 (1.83)	3 (40)
	Milo	142	Hatchery	191 (11)	77 (14)	8.15 (1.94)	44 (85)
	Weldon tailrace	149	Wild	178 (18)	52 (16)	9.08 (1.85)	24 (60)
2006	Milo	142	Hatchery	196 (11)	87 (18)	4.86 (1.24)	38 (72)
	Weldon	149	Hatchery	199 (15)	87 (19)	4.78 (1.85)	53 (146)
	Weldon tailrace	149	Wild	189 (9)	62 (10)	4.12 (1.10)	14 (73)
2009	Milo	142	Hatchery	180 (8)	62 (9)	3.29 (1.38)	73 (100)
	Passadumkeag	92	Hatchery	180 (9)	63 (9)	3.02 (0.82)	77 (100)
2010	Abbot	187	Wild	169 (8)	45 (7)	4.68 (1.10)	19 (75)
	Weldon head pond	162	Wild	180 (14)	55 (13)	4.81 (0.92)	15 (74)
	Milo	142	Hatchery	189 (11)	72 (13)	4.53 (1.04)	63 (100)
	Passadumkeag	92	Hatchery	186 (11)	69 (13)	4.63 (1.13)	77 (100)
2011	Abbot	187	Wild	146 (8)	29 (5)	2.61 (1.17)	55 (75)
	Weldon head pond	162	Wild	163 (19)	42 (16)	3.30 (1.41)	14 (60)
	Milo	142	Hatchery	191 (13)	75 (18)	4.94 (1.47)	55 (100)
	Passadumkeag	92	Hatchery	194 (13)	76 (17)	5.42 (1.49)	74 (100)
2012	Abbot	187	Hatchery	199 (10)	84 (14)	3.35 (1.33)	54 (72)
	Weldon head pond	162	Hatchery	200 (11)	85 (14)	3.48 (1.59)	46 (85)
2013	Abbot	187	Hatchery	185 (11)	70 (13)	2.80 (2.13)	20 (75)
	Weldon head pond	162	Hatchery	185 (9)	71 (11)	2.75 (2.04)	33 (82)
All	-	-	-	187 (16)	71 (19)	4.39 (1.92)	941 (1824)

Covariates of estuary arrival date, movement rate, and survival

I collected data about several hypothesized covariates of estuary arrival date, movement rate, survival, or detection probability of Atlantic salmon smolts during estuary migration. Broadly, I categorized these covariates as characteristics of individual fish, characteristics of the migratory history of a fish, and environmental covariates. All continuous covariates were standardized prior to each analysis (i.e., timing, movement, and survival analyses) to facilitate ease of interpretation among covariates. Covariate effects were considered statistically significant if the 95% confidence interval (CI) of the estimated coefficient for the covariate did not include zero.

Characteristics of individual fish

Five covariates were used to represent characteristics of individual fish in models: fork length (L_F, mm), Fulton condition factor (K), gill NKA activity, model of acoustic tag that was used (V7 or V9), and rearing history (wild or hatchery). To measure gill NKA activity, a nonlethal gill biopsy (4-6 filaments) was taken from the front, left gill arch of each fish prior to tagging. Individual biopsies were stored at -80°C in 100 μL SEI buffer (250 mM sucrose, 10 mM Na_2-EDTA, 50 mM imidazole) for later analysis of gill NKA (enzyme code 3.6.3.9; IUBM 1992) activity (expressed as μmol ADP$\cdot$mg protein$^{-1}\cdot h^{-1}$) using the method of McCormick (1993). Concentration of NADH at 25°C and 340 nm was used to measure kinetic rate of ouabain-inhibiTable 4.ATP hydrolysis, and protein concentration in gill samples was determined using the bicinchoninic acid (BCA) method (Smith et al. 1985). Gill samples were analyzed in triplicate for gill NKA activity and protein concentration and averaged.

Migratory history of individuals

I used five covariates to represent the migratory history of each fish: 1) release date, 2) location (in river kilometer) of release, 3) migratory routing through the lower Penobscot River), 4) number of dams passed during migration (to evaluate dam-related estuary mortality), and 5) median movement rate through the estuary (only in survival models). Based on differences in performance between freshwater migration routes in the lower river (Holbrook et al. 2011; Stich et al. 2014), I hypothesized that differential use of the Stillwater Branch or main-stem might result in differences in estuary arrival date, movement rates, and/or dam-related estuary mortality based on dams in each route. Because of imperfect detection at acoustic receivers in the lower river, use of the Stillwater Branch by smolts was included as a binary covariate in statistical analyses, and fish with unknown migratory route were assigned the mean value of the covariate (Stillwater = 1, main-stem = 0, unknown = 0.5).

Smolts released in the freshwater reaches of the Penobscot River passed 2–9 dams from stocking locations to the head of tide during years of this study (2005–2013). To test whether or not smolts experienced delayed mortality in the estuary due to the passage of dams (hereafter ‘dam-related estuary mortality’) or behavioral effects from dam passage, I examined the relationship of the number of dams a fish passed to estuary arrival date, movement rate, and survival in the estuary. The number of dams passed by each smolt was conditional on migratory route in the lower river (main-stem or Stillwater Branch) because the number of dams differed between routes. The number of dams passed also depended on year due to the removal of Great Works Dam in summer 2012.

Environmental covariates

I collected information about environmental covariates of estuary arrival date, movement rate, and survival that included temperature and discharge throughout the catchment, as well as photoperiod at the head of tide. In-river temperature data (R. Spencer, Maine Department of Marine Resources, unpublished data; and USGS gage station 01036390) were used to calculate accumulated thermal units (ATU) experienced by wild smolts in the watershed prior to tagging, and temperature data from outdoor rearing pools at GLNFH (A. Firmenich, US Fish and Wildlife Service, unpublished data) were used to calculate ATU for hatchery-reared smolts from 1 January to date of tagging. Photoperiod was calculated from the latitude at the head of tide in the estuary and ordinal dates using the package ‘geosphere’ in R (R Development Core Team 2014). Discharge data were obtained for the USGS gage at the West Enfield Dam (Figure 4.1, H) for each day during all years from 2005 through 2013 and used to characterize discharge in the main-stem of the Penobscot River during the period of smolt migration.

Models of estuary arrival date

Estuary arrival date was assigned as the first detection in the estuary for each smolt detected below Veazie Dam (rkm 45). I estimated effects of covariates on estuary arrival date using generalized linear models (GLMs) with a $\log_e$ (Poisson family) link function (Montgomery et al. 2006) in R. I used an information-theoretic approach to model selection to determine the relative influences of fish characteristics, migratory history, and environmental variables on estuary arrival date by comparing *a priori* combinations of covariates. I thought that it was important to account for release date regardless of what other covariates were included in the timing models; therefore, photoperiod was included in all models of estuary arrival date. Results were plotted with corresponding calendar dates to facilitate interpretation. I note that the entire smolt run occurred prior to the vernal equinox each year such that photoperiod only ever increased with progressively later calendar dates (i.e. no two dates had the same photoperiod).

I constructed models containing a single variable I classified as a 'fish characteristic' in any given model to reduce the potential for spurious effects, simplify the model set, and facilitate comparison between competing explanations for factors affecting estuary arrival date. I did not consider models that contained dams passed in addition to the rkm of release or use of Stillwater Branch because 1) there was a strong correlation between rkm of release and dams passed, and 2) dams passed was conditional on whether fish migrated through the Stillwater Branch or the main-stem in the lower river. I evaluated the relative support for candidate models using Akaike information criterion corrected for sample size (AIC_c; Burnham and Anderson 2002). Approximation of a variance inflation factor ($\hat{c}$) for the most parameterized model prior to analysis indicated reasonable model fit ($\hat{c} \approx 1.00$), so model selection was not adjusted.

Movement rate

Individual movement rates through the estuary were calculated from consecutive seaward detections of Atlantic salmon smolts for which detections at more than one receiver location in the estuary were recorded. Movement rate (R) of individual fish (i) through estuary reaches (j) was calculated as kilometers per hour (R_{ij}, km·h^{-1}) based on the distance between consecutive relocations D_{ij}, and amount of time elapsed between first detections at consecutive locations (T_{ij}) for each fish (L_{Fi}) using:

$$R_{\mathrm{ij}} = \left(\frac{D_{\mathrm{ij}}}{T_{\mathrm{ij}}} \right) \times \left(\frac{1}{L_{F\mathrm{i}}} \right) \qquad \text{(Equation 4.1)}$$

I used linear mixed-effects models (Zuur et al. 2009) in R (R Development Core Team 2014) to estimate relative effects of 1) fish characteristics, 2) migratory history, 3) detection location in the estuary, 4) and estuary arrival date on the rate of individual fish movements through the estuary. I included an individual-based random effect on the intercept term in all models to account for repeated, unequal numbers of measurements of movement rate for each fish. Movement rate was constrained to be greater than zero (i.e., predicted movement rate cannot be negative); therefore, I $\log_e$-transformed movement rate prior to analysis.

I included estuary location (in rkm), movement timing (photoperiod and photoperiod^2), and freshwater discharge in all models of movement rate because I had strong *a priori* expectations that movement rate changed in the estuary, during the smolt window and with discharge. A second-order term was included for photoperiod because I expected that movement rate would peak during the middle of the smolt run. Because I also hypothesized that movement rate would be faster at head of tide and at the mouth of the estuary than in the primary mixing zone in the middle estuary, I compared models with only a linear term for rkm to models containing a second-order (quadratic) term for receiver rkm. Other than these modifications, I followed the same process for *a priori* model construction and model selection as was used for analysis of estuary arrival date. Approximation of $\hat{c}$ for the most parameterized model indicated reasonable model fit ($\hat{c} \leq 1.00$), so model selection was not adjusted.

Survival Analysis

Detections at receiver locations were used to develop individual recapture histories (located = 1, not located = 0) for each fish during estuary passage (Figure 4.1). I used encounter histories to estimate apparent survival (hereafter ‘survival’; ϕ) and detection probability (p) of smolts in the estuary using a spatially explicit form of the Cormack-Jolly-Seber (CJS) mark-recapture modeling framework (see Figure 4.1). While the term survival is used throughout this study for simplicity, estimates reflect only apparent survival and not true survival of smolts as information about whether fish were alive or dead was not available. I conducted the survival analysis using maximum-likelihood estimation in MARK (version 7.1, White and Burnham 1999) via the ‘RMark’ package (Laake 2013) in program R, (version 3.1.0, R Development Core Team 2014). All parameters were estimated using the logit link function.

I used 12 encounter occasions (i.e., receiver locations) in the CJS models, which included for a ‘virtual release’, or starting location, at the head of tide (Figure 4.1). I accounted for differences in reach lengths (i.e., distances between receiver locations) by including reach lengths within the models. As a result, the estuary array provided for estimates of survival through 11 reaches of the Penobscot River Estuary, each standardized as per-kilometer rate within those reaches. However, estimates of survival and detection probability are confounded in the final interval of CJS models and so the joint probability of recapture and survival (λ) was estimated in the eleventh and final interval (Penobscot Bay), whereas survival was estimated separately in the first 10 reaches.

I investigated relationships between survival and 1) fish characteristics, 2) migratory history, and 3) environmental effects. I include five covariates to represent characteristics of individual fish in CJS models: 1) L_F, 2) K, 3) gill NKA activity, 4) rearing history, and 5) tag type. Based on prior work (Zydlewski, unpublished data) and the difference in signal power between tag models, I expected the smaller (V7, 136 dB) tags to have a lower probability of being detected than the larger (V9, 151 dB) tags. Therefore, tag type was included as a binary covariate of detection probability (V7 = 0, V9 = 1).

I included five covariates to represent migratory history: 1) photoperiod (to represent timing), 2) median movement rate for each fish, 3) migratory route (Stillwater or main-stem) in the lower Penobscot River, 4) release rkm, and 5) number of dams passed. I included year, estuary reach, photoperiod, and discharge in the main-stem of the river to explain spatial and temporal variation in survival. Year, estuary interval, and discharge were used to model heterogeneity detection probability.

Goodness of fit and model selection for survival analysis

I used an information-theoretic approach to model selection to compare competing hypotheses about factors affecting survival and detection probability of acoustically tagged Atlantic salmon smolts in the Penobscot River Estuary. I assessed goodness of fit (GOF) for my most general model using the median $\hat{c}$ approximation in MARK (White and Burnham 1999) to estimate overdispersion. The result of this test indicated that the most general model was slightly overdispersed ($\hat{c} \approx 1.265$); therefore, I adjusted model selection and variances of estimated regression coefficients for overdispersion, and I used quasi-Akaike's information criterion corrected for sample size ($QAIC_c$) for model selection (Burnham and Anderson 2002).

My model-selection approach had two hypothesis-driven phases. First, I identified the best spatial and temporal model structure for survival (year, reach, and/or photoperiod) while allowing detection probability to vary according to year, reach, acoustic tag model, and/or discharge. I compared *a priori* combinations of survival and detection parameterizations to determine my model for subsequent hypothesis testing. Based on my experience, I did not consider null (i.e., static or constant) models of survival or detection probability. For detection probability I only considered combinations of explanatory variables that included group (year) and reach effects. Due to limitations of sample sizes, I considered only additive group-by-reach effects (i.e., different intercepts for years in each reach but the same slopes) for both survival and detection probability. My justification for this decision was that if survival or detection probability varied between sites, then variation was likely similar in direction among years but may have varied in magnitude.

After I identified the best spatial and temporal model structure for survival and accounted for factors influencing detection probability, I constructed a model set, building on this base model, to test hypotheses based on fish characteristics and migratory history. I did not include more than a single covariate representing migratory history of fish in a given model because of known dependencies between some variables (migratory route and number of dams passed) and colinearity between others (e.g., release rkm and number of dams passed).

RESULTS

Estuary arrival date

The best model used to describe estuary arrival date explained 50% of the variation observed during the past decade (McFadden's pseudo R^2= 0.50; Faraway 2005). The mean (± SD) arrival date in the Penobscot River Estuary for Atlantic salmon smolts released in freshwater was May 9 (± 8 days). Smolts released earlier in the year (shown as photoperiod at release) arrived in the estuary at an earlier date than those fish released later in the season (Table 4.2). Over the range of release dates used in the past decade (range: April 12–May 29) arrival date was 24 days earlier for the earliest release dates than for the latest release dates (Table 4.3).

Smolts released further upstream of the estuary arrived in the estuary at a later date than those fish that were released further downstream. The difference in arrival dates between the furthest upstream and furthest downstream release sites, located approximately 90 rkm apart (range rkm 92–rkm 187), was three days (Figure 4.2a) and the effect was not statistically significant at $\alpha = 0.05$ (Table 4.3) unless discharge was not included in the same model. Observed discharge during the period between release dates and estuary arrival dates 2005–2013 ranged from a minimum of 175 $m^3 \cdot s^{-1}$ to 2,500 $m^3 \cdot s^{-1}$. Fish experiencing the greatest discharge between release date and estuary arrival date arrived in the estuary 10 days later than fish experiencing the least discharge (Figure 4.2b, Table 4.3).

Table 4.2. Model selection for estuary arrival date. Model-selection statistics for generalized linear models (GLMs) that were used to quantify relationships between date of arrival by acoustically tagged Atlantic salmon smolts in the Penobscot River Estuary 2005–2013 and fish characteristics (Rearing history [Rearing], gill NKA activity), migratory history (Photoperiod at release date [PP], Release rkm [Release], migratory route through the lower river [Stillwater; SW]), number of dams passed [Dams], and environmental conditions (accumulated thermal units [ATU], and discharge experienced from release to arrival date). Number of parameters estimated in each GLM is *k*, AIC_i is the Akaike information criteria for each i^{th} model and $\Delta AICc_i$ is the difference between the *AICc* of each i^{th} model and the best model in the candidate set, and w_i is the relative probability that each i^{th} model is the best in the candidate set. Top ten models are shown: models for which $\Delta AICc_i < 2.0$ were considered to have similar support as the best model.

Model	*k*	$AICc_i$	$\Delta AICc_i$	w_i
Discharge + PP + Rearing + Release	5	6556.57	0.00	0.16
Discharge + PP + Rearing + Release + SW	6	6556.98	0.42	0.13
Discharge + PP + Rearing	4	6558.03	1.47	0.08
ATU + Gill NKA activity + PP + Release + SW	7	6558.16	1.59	0.07
ATU + Discharge + PP + Rearing +Release	6	6558.59	2.02	0.06
ATU + Discharge + Gill NKA activity + PP + Release	7	6558.71	2.14	0.05
Discharge + PP + Rearing + SW	5	6558.72	2.15	0.05
ATU + Gill NKA activity + PP + Rearing	6	6558.82	2.26	0.05
ATU +Discharge + Gill NKA activity + PP + Release + SW	8	6558.91	2.34	0.05
ATU + Discharge + PP + Rearing + Release + SW	7	6558.96	2.40	0.05

Table 4.3. Coefficients for covariate effects on estuary movement behavior. Standardized regression coefficients, standard error (SE) and 95% confidence limits (CI) for generalized linear model (GLM) used to model estuary arrival date (as a function of photoperiod at release date (PP),release rkm, rearing history, and discharge, as well as the linear mixed-effects model used to model movement rates as a function of location (‘Estuary rkm’), discharge, rearing history, release rkm (Release), and photoperiod for date of movement (PP, PP^2).

Model	Parameter	Estimate	SE	Lower 95% CL	Upper 95% CL
Arrival date	Intercept	4.861	0.004	4.868	4.854
	Discharge	0.017	0.003	0.022	0.011
	PP	0.022	0.005	0.031	0.013
	Rearing (wild)	0.040	0.015	0.069	0.012
	Release	0.007	0.004	0.014	0.000
Movement rate	Intercept	0.306	0.019	0.270	0.343
	Discharge	0.034	0.018	0.000	0.069
	Estuary rkm	0.174	0.016	0.144	0.205
	PP	0.057	0.018	0.022	0.092
	PP^2	-0.012	0.011	-0.034	0.009
	Release	0.081	0.016	0.049	0.113

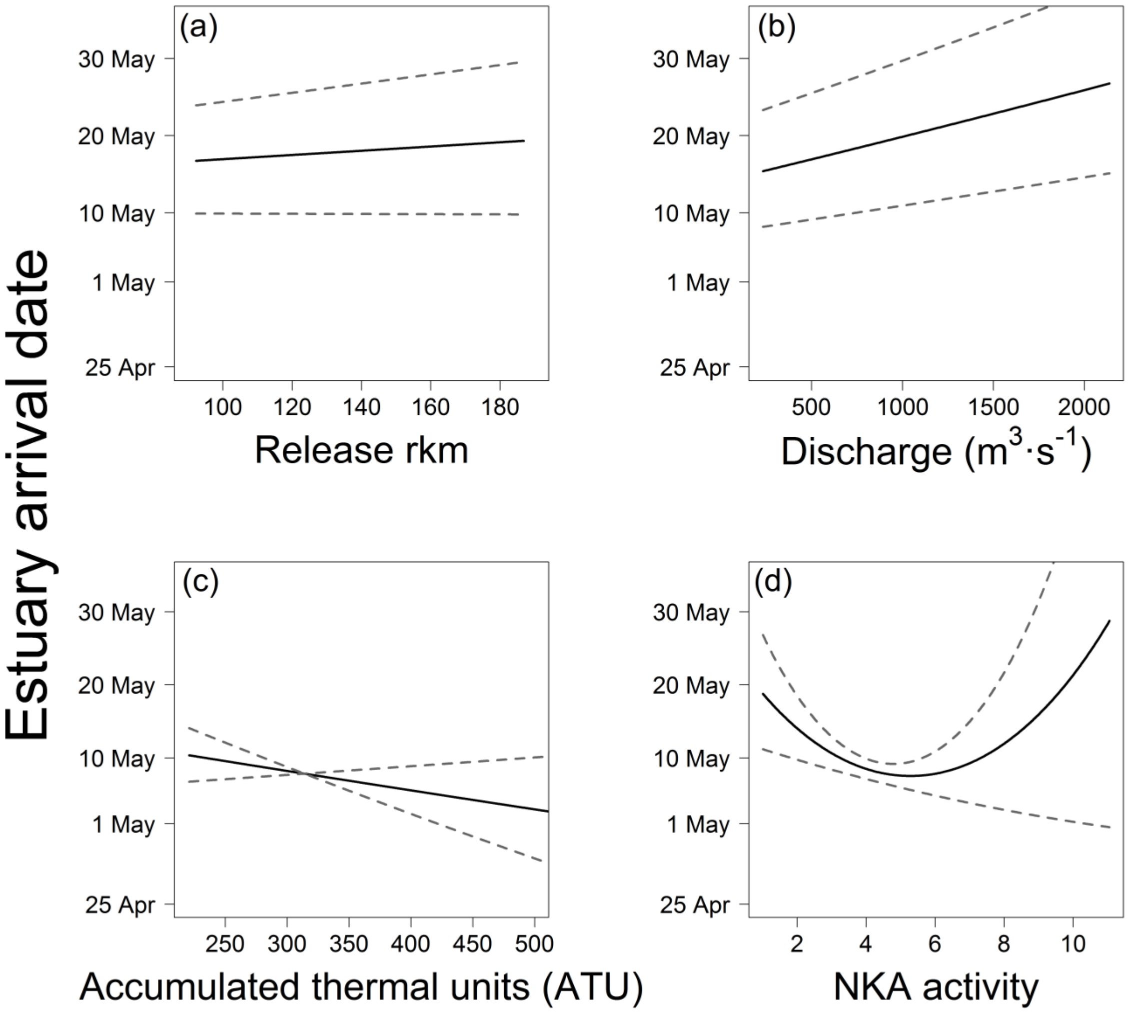


Figure 4.2. Covariate effects on estuary arrival date. Predictions from the generalized linear model relating arrival date of acoustically tagged Atlantic salmon smolts in the Penobscot River Estuary 2005–2013 to a) release rkm (upstream of estuary), b) discharge experienced from tagging to estuary arrival date, c) accumulated thermal units (ATU) experienced prior to release, and d) gill NKA activity.

Atlantic salmon smolts that were wild-reared (i.e., progeny of wild spawning, egg planting, or fry stocking) arrived in the estuary later than smolts that were reared in the hatchery (Table 4.3). Mean (± SD) estuary arrival date for wild-reared fish was May 20 (± 7 days), whereas mean estuary arrival date for hatchery-reared smolts was May 7 (± 6 days), a difference of two weeks. The majority of this differences results directly from later tagging dates of wild-reared smolts (mean = 13 May) compared to release dates of hatchery-reared smolts (mean = 24 April).

Fish experiencing a warmer thermal history (higher ATU) prior to tagging and release arrived in the estuary earlier than fish experiencing lower ATU prior to release when the effect of release date was accounted for (Table 4.2). This relationship indicated that experiencing the greatest ATU (550) arrived 8 days earlier than those fish experiencing the lowest ATU (220) prior to release (Figure 4.2c). However, ATU covaried with discharge, and discharge was a better predictor of estuary arrival date than ATU because ATU was not included in the best model and was not statistically significant at α=0.05 when discharge was included in the same model.

Estuary arrival date decreased with increasing gill NKA activity, until gill NKA activity reached 6 μmol ADP·mg protein^{-1}·h^{-1}, after which estuary arrival date began to increase with gill NKA activity (Figure 4.2d). As a result, Atlantic salmon smolts that had very low or very high gill NKA activity arrived in the estuary later than smolts near the mean gill NKA activity. Similar to ATU, gill NKA activity covaried with discharge and was neither included in the best model nor significant at α=0.05 when discharge was included in the same model.

Movement rate

Mean (± SD) movement rate of smolts through all reaches of the estuary, and among all fish was 2.27 (± 1.88) km·h^{-1}. I observed evidence of a quadratic relationship between date of movement and individual movement rate. Fish increased movement rate until the middle of the migration period (early May), after which movement rate became more variable, but appeared to asymptote or even decrease (Figure 4.3a, Table 4.3).

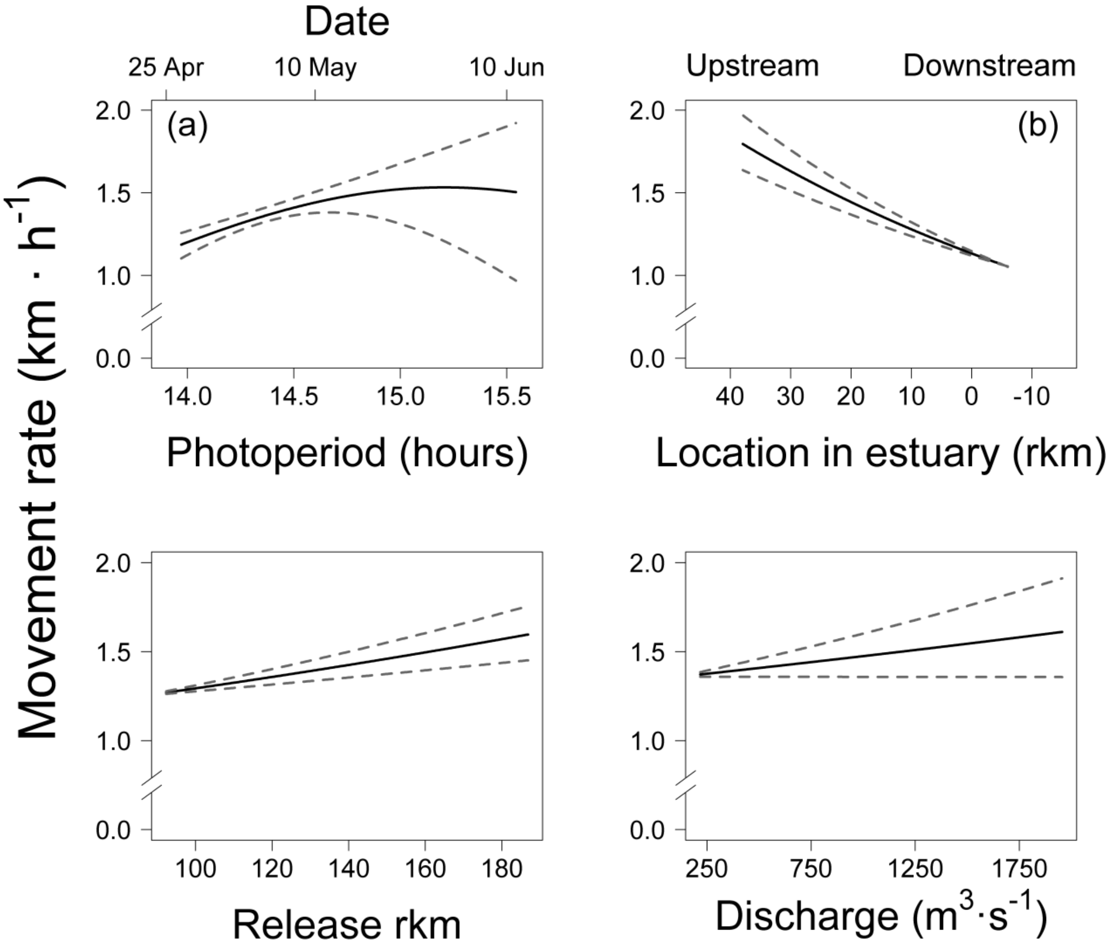


Figure 4.3. Covariate effects on estuary movement rate of smolts. Mean (solid line) and 95% CI (dashed lines) predictions from parameters of generalized linear mixed-effects model (GLMM) used to relate movement rate of acoustically tagged Atlantic salmon smolts in the Penobscot River Estuary 2005–2013 to a) photoperiod, b) location in estuary (rkm), c) release distance upstream of the estuary (rkm), and d) discharge experienced during estuary migration.

Movement rate of smolts decreased in the Penobscot River Estuary from the head of tide to the mouth of the estuary (Figure 4.3b, Table 4.3), and a second-order term for estuary rkm was not supported in the candidate model set (Table 4.4). From the head of tide to the mouth of the estuary, a distance of 50 km, movement rate of smolts decreased from a mean of 1.8 km·h^{-1} to 1.1 km·h^{-1} (Figure 4.3b).

Table 4.4. Model selection for smolt movement rate in estuary. Model-selection statistics for the ten best mixed-effects models (GLMM) that used to quantify relationships between movement rate by acoustically tagged Atlantic Salmon smolts in the Penobscot River Estuary and fish characteristics (gill NKA activity, Fulton condition factor [K], rearing history [Rearing]), migratory history (Release rkm [Release], migratory route through the lower river [SW]), and spatial/temporal factors (photoperiod of date at which movement occurred [PP, PP^2], number of dams passed [Dams], and rkm of receiver location [rkm, rkm^2]). Table headings and model selection statistics are defined as in Table 4.2.

Model	k	$AICc_i$	$\Delta AICc_i$	w_i
Discharge + PP + PP^2 + Release + rkm	7	20962.11	0.00	0.84
Discharge + PP + PP^2 + Rearing + Release + rkm	8	20967.19	5.08	0.07
Discharge + PP + PP^2 + Rearing + Release + rkm + rkm^2	8	20967.46	5.35	0.06
Discharge + PP + PP^2 + Gill NKA activity + Release + rkm	8	20969.89	7.78	0.02
Discharge + K + PP + PP^2 + Release + rkm	8	20970.48	8.37	0.01
Discharge + PP + PP^2 + Rearing + Release + rkm + rkm^2	9	20972.55	10.44	0.00
Dams + Discharge + PP + PP^2 + rkm	7	20973.60	11.49	0.00
Discharge + Gill NKA activity + PP + PP^2 + Release + rkm + rkm^2	9	20975.01	12.90	0.00
Discharge + K + PP + PP^2 + Release + rkm + rkm^2	9	20975.56	13.45	0.00
Dams + Discharge + PP + PP^2 + Rearing + rkm	8	20978.63	16.52	0.00

Smolts released further upstream in the Penobscot River moved faster than tagged smolts released at downstream sites (Figure 4.3c, Table 4.3). However, the effect of release rkm on movement rate appeared to be minor in comparison to effects of arrival date and estuary location: over the range of release rkms used in this study (92–187), maximum movement rate increased by less than 0.5 km·h^{-1}. Movement rate through the estuary increased with increasing discharge (Figure 4.3d). As with release rkm, the effect of discharge was minimal, resulting in a change of less than 0.3 km·h^{-1} over the range of discharge observed (217–1,957 m^{3}·s^{-1}).

Survival

Survival and detection probability of smolts varied spatially, temporally within years, and among years (Table 4.5). Survival was highest near the head of tide (> 0.99) and the mouth of the estuary, and was lowest in the middle estuary (0.98) where tidal influences are strongest (Figure 4.4). Smolt survival was highest during the middle of the smolt run, peaking in early May when survival was as much as 70% greater than in the early or late run. Fish arriving in the estuary very early or very late had poor survival (near zero) by comparison to those arriving during the middle of the run (Figure 4.5a).

As expected, the probability of detecting smolts was higher for fish tagged with large (model V9) acoustic transmitters than for those tagged with smaller (model V7) transmitters (Table 4.6). The mean (95% CI) probability of detecting fish increased by 0.40 (0.34–0.47) when fish were tagged using large tags instead of smaller tags. Probability of detection also was affected by discharge experienced by individual smolts during estuary migration (Table 4.5). Over the range of flows observed (217–1,957 m^{3}·s^{-}

^{1}), the mean (95% CI) probability of detection was reduced by 0.48 (0.37–0.60) during periods of greatest freshwater discharge compared to periods of lowest discharge (Table 4.6). Based on these results, year, estuary reach, and photoperiod all were included in the final parameterization for survival. The final parameterization for the detection model included year, estuary reach, tag model, and discharge experienced by individual fish during migration. I tested all other hypotheses about effects of fish characteristics and migratory history using the parameterization above as a ‘base-model’.

Table 4.5. Model selection for estuary survival. Model-selection statistics for Cormack-Jolly-Seber (CJS) mark-recapture models used to determine the appropriate spatial and temporal structure for estimating survival of acoustically tagged Atlantic salmon smolts during emigration through Penobscot River Estuary. Number of parameters estimated in each CJS model is k, $QAIC_i$ is the Akaike information criterion for each i^{th} model (corrected for overdispersion), $\Delta QAICc_i$ is the difference between the $QAICc$ of each i^{th} model and the best model in the candidate set, and w_i is the relative probability that each i^{th} model is the best in the candidate set. The top ten candidate models are shown. Covariates of apparent survival (ϕ) and detection probability included estuary reach (Reach), year, linear or quadratic effects of photoperiod (PP or PP^2), tag type, and discharge.

Model	k	$QAICc_i$	$\Delta QAICc_i$	w_i
ϕ(Reach + Year + PP + PP^2) p(Reach + Year + Tag type + Discharge)	38	7211.13	0.00	1.00
ϕ(Reach + Year + PP) p(Reach + Year + Tag type + Discharge)	37	7248.90	37.78	0.00
ϕ(Reach + Year + PP + PP^2) p(Reach + Year + Tag type)	37	7273.77	62.64	0.00
ϕ(Reach + Year + PP) p(Reach + Year + Tag type)	36	7311.32	100.19	0.00
ϕ(Reach + PP + PP^2) p(Reach + Year + Discharge)	37	7333.34	122.21	0.00
ϕ(Reach + PP+ PP^2) p(Reach + Year + Tag type + Discharge)	32	7370.72	159.59	0.00
ϕ(Reach + Year + PP) p(Reach + Year + Discharge)	36	7371.32	160.20	0.00
ϕ(Reach + Year + PP + PP^2) p(Reach + Year)	36	7390.29	179.16	0.00
ϕ(Reach + Year) p(Reach + Year + Tag type + Discharge)	36	7407.49	196.37	0.00
ϕ(Reach + Year + PP) p(Reach + Year)	35	7428.10	216.97	0.00

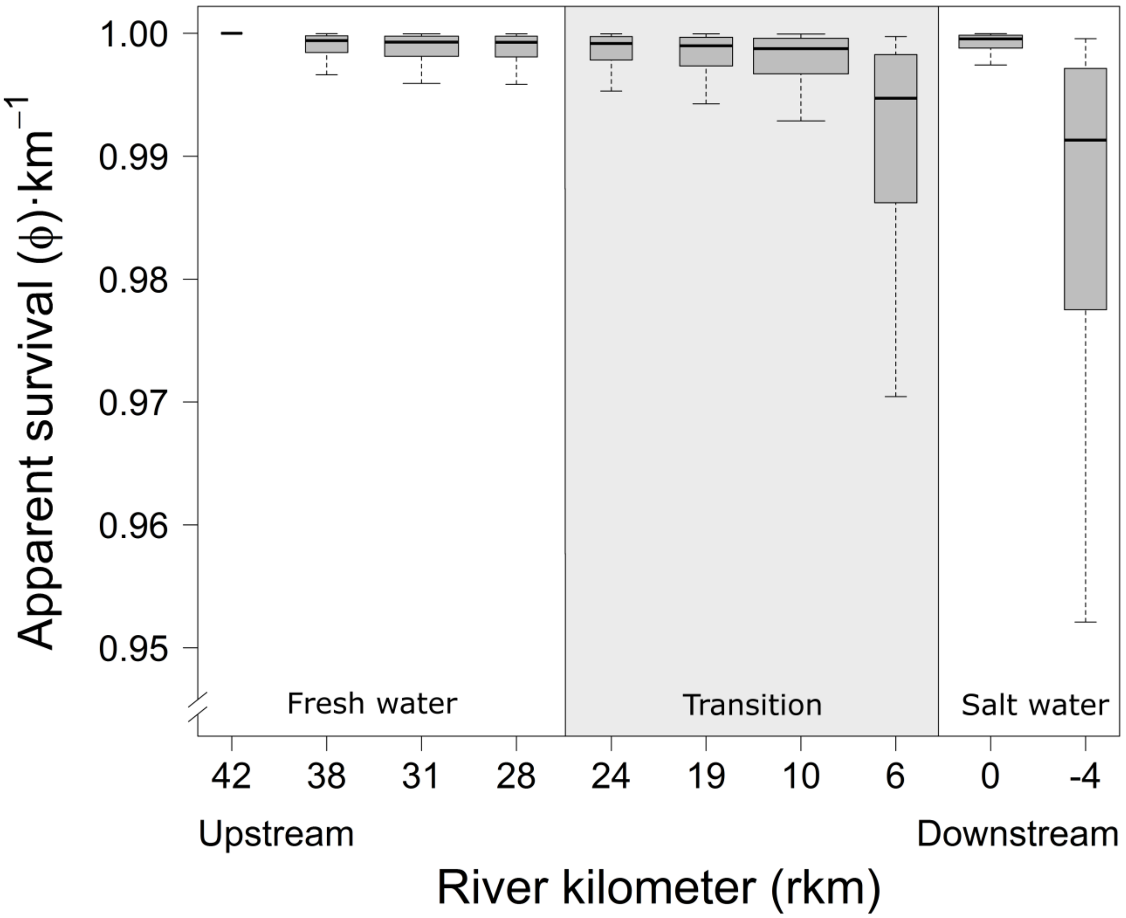


Figure 4.4. Annual smolt survival through the estuary. Estimates of mean apparent survival per kilometer of acoustically tagged Atlantic salmon smolts in the Penobscot River Estuary 2005–2013 estimated using Cormack-Jolly-Seber (CJS) models. The solid line in each box indicates median annual survival, box-ends indicate the inner quartile range and whiskers indicate 95% CI. Box widths are proportional to estuary reach lengths to which the estimates apply. From left to right, the three panels represent fresh water, the zone of transition from low salinity to high salinity, and salt water within the Penobscot River Estuary.

Table 4.6. Regression coefficients for covariate effects on estuary survival. Standardized regression coefficients (logit) and 95% confidence limits (CL) for individual covariates included in the top-ranked Cormack-Jolly-Seber (CJS) mark-recapture model used to estimate apparent survival (ϕ) and detection probability (p) of acoustically tagged Atlantic salmon smolts during emigration through Penobscot River Estuary 2005–2013. Tag type was classified as a binary covariate: smaller (V7) tags were assigned to zero, and larger (V9) acoustic tags were assigned to one.

Parameter	Covariate	Estimate	SE	Lower 95% CL	Upper 95% CL
ϕ	PP	1.631	0.110	1.414	1.847
	PP^2	-0.545	0.086	-0.713	-0.378
	Gill NKA activity	0.192	0.081	0.032	0.351
	Number of dams passed	-0.376	0.092	-0.557	-0.195
	Movement rate	-0.108	0.064	-0.233	0.018
p	Tag type	0.404	0.033	0.340	0.469
	Discharge	-0.475	0.053	-0.579	-0.371

After accounting for these effects, survival of smolts in the Penobscot River Estuary was dependent on physiological development (measured as gill NKA activity), dams passed, and the rate at which fish moved through the estuary (Table 4.7). I found strong evidence for an optimal timing of estuary passage related to survival (Figure 4.5a, Table 4.6). Survival increased until mid-May, after which survival became variable but appeared to decrease.

The number of dams passed by individual smolts had a strong, negative effect on fish survival in the estuary (Figure 4.5b, Table 4.6). Survival of smolts that passed the greatest number of dams (nine) was reduced by 40% compared to those fish that passed only 2 dams (minimum). The model including dams passed outperformed the corresponding model with release rkm by nearly 2 QAICc, suggesting that the majority of the effect of release location on survival was explained by the number of dams passed during migration, and not merely by the distance fish had migrated.

The physiological preparedness of smolts for saltwater entry (measured as gill NKA activity) at tagging was positively related to smolt survival during estuary passage (Figure 4.5c, Table 4.6). Atlantic salmon smolts with the highest gill NKA activity had 25 % greater probability of surviving the estuary migration than those fish with the lowest enzyme activity. The median movement rate of smolts was included in the best model of survival, and survival of smolts decreased slightly with increasing movement rate over the observed range of movement rates; however, the effect of this covariate was not statistically significant (Table 4.6), and the corresponding model that excluded effects of movement on survival had virtually identical support in the data.

Table 4.7. Model selection for estuary survival. Statistics for Cormack-Jolly-Seber (CJS) mark-recapture models used to estimate survival of smolts during emigration through Penobscot River Estuary 2005–2013, and to test hypothesis about fish characteristics and migratory history that influenced survival. Table headings and model-selection statistics are defined as in Table 5. The top ten candidate models are shown. Candidate models in this set included the covariates representing spatial and temporal variation ($\phi_{(\text{Reach + Year + PP + PP}^2)}$ from the best model in Table 4) in addition to the covariates shown in each model below. Covariates of apparent survival (ϕ) not described elsewhere are gill NKA activity, number of dams passed (Dams), median movement rate (R), and fork length (LF).

Model	k	$QAICc_i$	$\Delta QAICc_i$	w_i
ϕ (Dams + Gill NKA activity + R) p(time + Year + Tag type + Discharge)	41	7194.11	0.00	0.24
ϕ (Dams + Gill NKA activity) p(time + Year + Tag type + Discharge)	40	7194.28	0.17	0.22
ϕ (Gill NKA activity + Release) p(time + Year + Tag type + Discharge)	40	7195.80	1.69	0.10
ϕ (Gill NKA activity + R + Release) p(time + Year + Tag type + Discharge)	41	7195.92	1.81	0.10
ϕ (Dams + LF) p(time + Year + Tag type + Discharge)	40	7196.65	2.54	0.07
ϕ (Dams + R) p(time + Year + Tag type + Discharge)	40	7196.68	2.56	0.07
ϕ (Dams) p(time + Year + Tag type + Discharge)	39	7196.74	2.62	0.06
ϕ (Dams + LF + R) p(time + Year + Tag type + Discharge)	41	7197.03	2.92	0.05
ϕ (LF + Release) p(time + Year + Tag type + Discharge)	40	7197.51	3.40	0.04
ϕ (LF + R + Release) p(time + Year + Tag type + Discharge)	41	7198.28	4.16	0.03

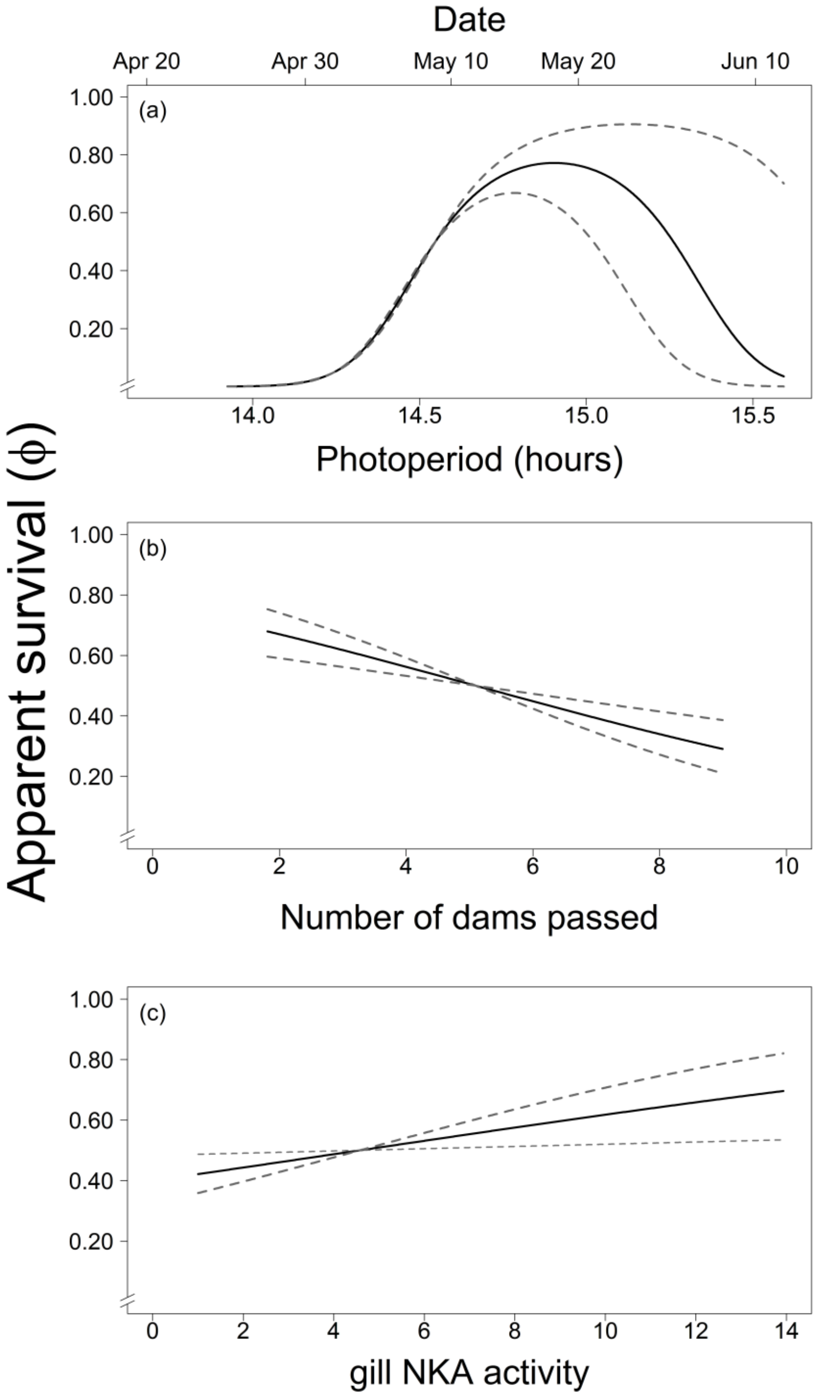


Figure 4.5. Covariate effects on estuary survival. Mean (solid line) and 95% confidence intervals (dashed lines) of predictions from parameters of the Cormack-Jolly-Seber (CJS) mark-recapture models used to estimate apparent survival (ϕ) of acoustically tagged Atlantic salmon smolt survival through the Penobscot River Estuary 2005–2013. Panels show effects of a) individual timing, b) number of dams passed by individuals, and c) gill NKA activity of individuals on estimated survival.

DISCUSSION

By linking the arrival date, movement rate, physiological preparedness, and survival of smolts over nearly a decade, I were able to improve my understanding of complex relationships and interactions between behavior and survival during estuary migration. Although a number of other studies have explored effects of factors influencing estuary arrival date, movement rate, and survival of Atlantic salmon smolts and post-smolts during early marine migration (see Thorstad et al. 2012a), few (if any) studies have had the opportunity to examine all of these processes together in a single population from distant upriver release sites all the way to the ocean. Furthermore, this study provides an unprecedented link between physiological preparedness (gill NKA activity) and performance in the wild. Similarly, this study is the first to clearly relate estuary survival of Atlantic salmon directly to delayed effects of prior dam passage.

Estuary arrival date

Smolt arrival in the Penobscot River Estuary was overwhelmingly driven by environmental conditions. I found that estuary arrival date was negatively related to both thermal history (ATU) experienced by smolts prior to tagging, as well as freshwater discharge between tagging and arrival date. Migratory behavior in Atlantic salmon smolts has been shown in multiple laboratory studies to be driven by environmental effects, namely photoperiod (Zydlewski et al. 2014) and temperature (Zydlewski et al. 2005). Those studies have shown that smolts experiencing cooler temperatures during development initiate migration at a later date. My field studies are consistent with that lab work and demonstrate that Atlantic salmon experiencing a warmer thermal history arrived in the estuary earlier than those experiencing cooler conditions. Sykes et al. (2009) reported that wild Chinook salmon *Oncorhynchus tshawytscha* smolts initiated migration later when they experienced cooler temperatures during development and during high discharge. Similarly, global trends in the timing of Atlantic salmon smolt migrations show that smolts experiencing higher discharge migrate later than those experiencing low discharge (Otero et al. 2014). Thus, my study is consistent with previous studies, and the understanding that environmental factors drive the onset of migration in smolts.

Timing of estuary arrival was related to physiological development of Atlantic salmon smolts in the Penobscot River. Fish with the lowest or greatest gill NKA activity arrived in the estuary later than fish that had intermediate gill NKA activity at release. This is consistent with behavioral adherence to the physiological smolt window for migration (McCormick et al. 1998). Arrival date for smolts with the greatest gill NKA activity was highly variable. This variability at the peak of physiological smolting may be due to heightened sensitivity to stress associated with tagging and handling (Carey and McCormick 1998).

Rates of movement

Rate of movement in the estuary by smolts was influenced by release location, date of individual movements, and location within the estuary. Smolts released further upstream in freshwater moved faster through the estuary than smolts released further downstream. These results suggest that fish released further upstream in the watershed begin to ‘catch up’ to fish released further downstream, either as a result of increased migratory speed by fish released upstream (Jokikokko and Mäntyniemi 2003) or as a result of station-holding (i.e., staging) behavior by smolts released downstream in the watershed prior to estuary arrival (e.g., Strand et al. 2010). Smolts that arrive in the estuary earliest may delay subsequent migration to synchronize ocean entry with optimal temperatures that connote improved ability to avoid predators, increased abundance of sympatric prey species, or greater food availability (Otero et al. 2014). Differences in behavioral priming (Dingle and Drake 2007) or environmental conditions experienced between different regions of the watershed (Whalen et al. 1999; Zydlewski et al. 2005) offer an alternative explanation. Positive reinforcement of downstream-movement behavior during migration might promote increased rates of migration for fish released further upstream (Zydlewski et al. 2005). Also, fish in headwater reaches of the river might experience increased intensity of exposure to environmental factors that prime migratory behavior than fish released downstream in main-stem river reaches due to smaller water volumes that respond more directly to changing conditions.

Smolts decreased migratory speed throughout the course of estuary emigration. It is likely that reductions in movement rate during estuary emigration were primarily due to changes in current velocity and tidal influences in the lower Penobscot River Estuary (Fried et al. 1978; Moore et al. 1995) compared to net discharge in freshwater reaches (net seaward movement of water). Reduction in movement rates through estuaries might also result from reversal of migratory direction during passage of tidal estuaries and bays (Kocik et al. 2009; Dempson et al. 2011; Halfyard et al. 2013). These behaviors appear to be related to tidal cycles in both estuarine (McCleave 1978; Martin et al. 2009) and coastal systems (Lacroix et al. 2005). This behavior previously has been hypothesized to relate to saltwater acclimation (Gudjonsson et al. 2005; Dempson et al. 2011; Halfyard et al. 2013). However, if related to saltwater acclimation, then the behavior may not provide an actual fitness benefit because survival is related to gill NKA activity at the time of release in freshwater, which implies that competence for saltwater entry is developed prior to estuary arrival. This finding is corroborated by results of studies in the Northeast Atlantic, where smolts acclimated in net pens within an estuary showed no improvement in survival over fish released directly into the mouth of the river (Thorstad et al. 2012b). Other laboratory experiments have found no period of acclimation upon reaching salt water (Moore et al. 1995). It is, however, possible that fish use tidal movements to minimize energetic costs through zones of net land-ward movements, especially because this is the period of migration during which smolts are thought to transition from passive to active migration (Hedger et al. 2008; Martin et al. 2009), which could be thought of alternatively as a failure in selective tidal stream transport. Further investigation of diurnal and tidal factors that influence behavior and survival on localized spatial and temporal scales could provide improved understanding of those mechanisms and might have implications for strategic stocking of hatchery-reared smolts.

Survival

Estuary arrival date, individual fish characteristics, number of dams passed, and location within the estuary all influenced Atlantic salmon smolt survival during estuary passage. I found evidence for a strong optimizing effect of estuary arrival date on the survival of Atlantic salmon smolts during estuary emigration, emphasizing the importance of timing in determining the success of migrating smolts (McCormick et al.

1998). Although this trend previously was suspected based on narrow windows of estuary passage in many Atlantic salmon smolt runs (see Thorstad et al. 2012a), this study provides strong, empirically derived evidence linking survival during estuary migration to the variability in timing of individual estuary passage. The shape and spread of the timing-survival relationship in the Penobscot River suggests that normalizing selection may occur on the timing of estuary arrival through a direct link to survival. This relationship likely occurs in response to environmental cues and releasing factors prior to onset of migration and illustrates the importance of timing for successful estuary passage (McCormick et al. 1987; Hoar 1988). This supposition is supported by the strength of release date in freshwater as a predictor of timing for both wild and hatchery fish and the effect of gill NKA activity on survival in the estuary at a later date.

Estuary arrival date was a stronger predictor of smolt survival at a gross scale than the movement rate of individual smolts. The inclusion of movement rate in the best model of survival was somewhat perplexing given the lack of significance for the covariate, and the unexpected direction of the relationship to survival. Increased movement rate through estuaries is postulated to reduce exposure to predators and environmental stressors such as pollution (McCormick et al. 1998), but my data suggest that perhaps this may not be the case. Based on the similarity between the best model for survival in the present study and the second-ranked survival model (ΔQAIC < 0.20), it seems that the inclusion of movement rate in survival models in the present study resulted in minimal improvement in model fit, and that this variable was included because it covaried with some other important variable of interest, such as number of dams passed, gill NKA activity, or location within the estuary. Based on the other results of this study, it seems likely that there could be synergistic effects of physiological preparedness and movement rate that result in changes to individual survival, but further investigation of the relationships is needed.

Atlantic salmon smolt survival in the estuary increased with increasing gill NKA activity at the time of release. This result establishes a critical link between physiological preparation of smolts in fresh water for osmoregulation in the ocean and survival, which has been long-suspected by others (Boeuf 1993; Itokazu et al. 2014). A rich literature exists describing physiological transformations of diadromous fishes (Zydlewski and Wilkie 2013), and the study of smolt physiology constitutes a large body of work within that field (McCormick et al. 1998; McCormick 2013). It is well established that gill NKA activity is a useful indicator of osmoregulatory ability (Zaugg and McLain 1972; McCormick et al. 1998; McCormick et al. 2009) and migratory readiness in salmonids (Aarestrup et al. 2000). While researchers have demonstrated performance benefits of smolts up-regulating gill NKA activity in laboratory studies relative to sublethal indicators (McCormick et al. 2009), the present study has provided a direct link between physiological development of smolts and fitness in a study of actively migrating smolts. Although gill NKA activity is not a strong predictor of long-term growth scope or ocean performance (Zydlewski and Zydlewski 2011), my results underscore the importance of physiological preparedness for successful entrance into the marine environment.

I do not suspect that reduced gill NKA activity led to direct mortality due to inability to osmoregulate in the estuary because mortality is not generally observed in laboratory studies of Atlantic salmon during the period of smolt migration. However, proximate causes of mortality such as predation (Jarvi 1990; Handeland et al. 1997;

Hawkes et al. 2013), or acidosis due to synergies between osmoregulatory stress and other forms of stress (Jarvi 1989; Price and Schreck 2003; Berli et al. 2014) have the potential to dramatically increase when osmoregulatory capacity of smolts is suboptimal (McCormick et al. 2009). Research targeting the relationships between smolt physiology and sources of direct mortality (e.g., predation) in estuaries might help to further unravel links between physiology and proximate causes of mortality in the wild.

My data strongly implicate a delayed, negative effect of dam passage on survival in the estuary, reducing estuary survival by 6–7 % per dam passed. I demonstrated that the cumulative number of dams passed (ranging from two to nine dams in this study) was an important predictor of smolt survival. This result is consistent with the work of Schaller et al. (2014), who found that number of powerhouses passed by out-migrating Chinook salmon affected marine survival. It is, however, notable that my results are the first to demonstrate this trend in Atlantic salmon migrations. Furthermore, the *delayed* dam-related mortality experienced in the 50-km Penobscot River Estuary as a result of passing nine dams was comparable in magnitude to the cumulative, *acute* mortality incurred by smolts passing those same nine dams during the 150-km freshwater migration (Holbrook et al. 2011, this study). This suggests that studies of survival at dams, which are the basis for dam permitting, may drastically underestimate the effects of those dams.

Smolts experience injuries such as descaling during passage of dams in the Penobscot River (Music et al. 2011). Such injuries can severely impair osmoregulatory ability, and impairment can persist for several days after injury (Zydlewski et al. 2010). All fish entering the Penobscot River Estuary passed at least one dam prior to estuary arrival during the critical period during which reduced osmoregulatory ability from dam-related injuries is expected to persist. Many fish passed several dams within just 24–48 hours of estuary arrival. My results suggest that this experience reduces survival of smolts during estuary passage. Reduction in osmoregulatory ability during estuary passage has previously been linked to decreased ability to avoid predators (Handeland et al. 1997; Price and Schreck 2003), which could increase mortality in estuaries. Spatial patterns in survival through Penobscot River Estuary indicate that one possible mechanism of reduced survival in the estuary is the interaction between multiple factors (such as dam-related injury, gill NKA activity, and predators), because the greatest reduction in survival occurred upon reaching saltwater reaches of the estuary where osmoregulatory perturbance would have the greatest effect.

Conservation and management implications

Mortality of smolts during the early phase of marine migration can be high (Thorstad et al. 2012a), as was the case in this study. I have synthesized the major factors associated with performance in the Penobscot River Estuary (Figure 4.6). Included are spatial and temporal components of environmental variation, timing of estuary arrival, physiological development, and dam-related estuary mortality. The smolt window in the Penobscot River is defined by factors commonly observed to control physiological (McCormick et al. 1987; Hoar et al. 1988) and behavioral smolting (Sykes et al. 2009; Zydlewski et al. 2014), such as photoperiod, temperature, and discharge (Figure 4.6). The period during which estuary survival was expected to be greater than 50% in the Penobscot River Estuary spans 2–3 weeks as a result. Environmental control of physiological and behavioral smolting suggests that this species may be particularly susceptible to chronic and acute anthropogenic stressors.

Climate change, based on my data, has the potential to squeeze Atlantic salmon against the ecological and physiological limits to adaptability. Earlier seasonal warming could result in mismatches between physiological and ecological smolt windows based on run timing (McCormick et al. 1997; Todd et al. 2012; Otero et al. 2014). Although physiological smolt development and timing of initiation of migratory behavior could change synchronously (McCormick et al. 1997) to result in shifting run times initially, these changes are also both entrained by circannual rhythms in photoperiod (Zydlewski et al. 2014) so there may be limits to how early physiological and behavioral smolting can occur (Otero et al. 2014). Resilience of many North American stocks to strong selection pressures imposed by these shifts is unknown (Hayes and Kocik 2013; Mills et al. 2013; Friedland et al. 2014).

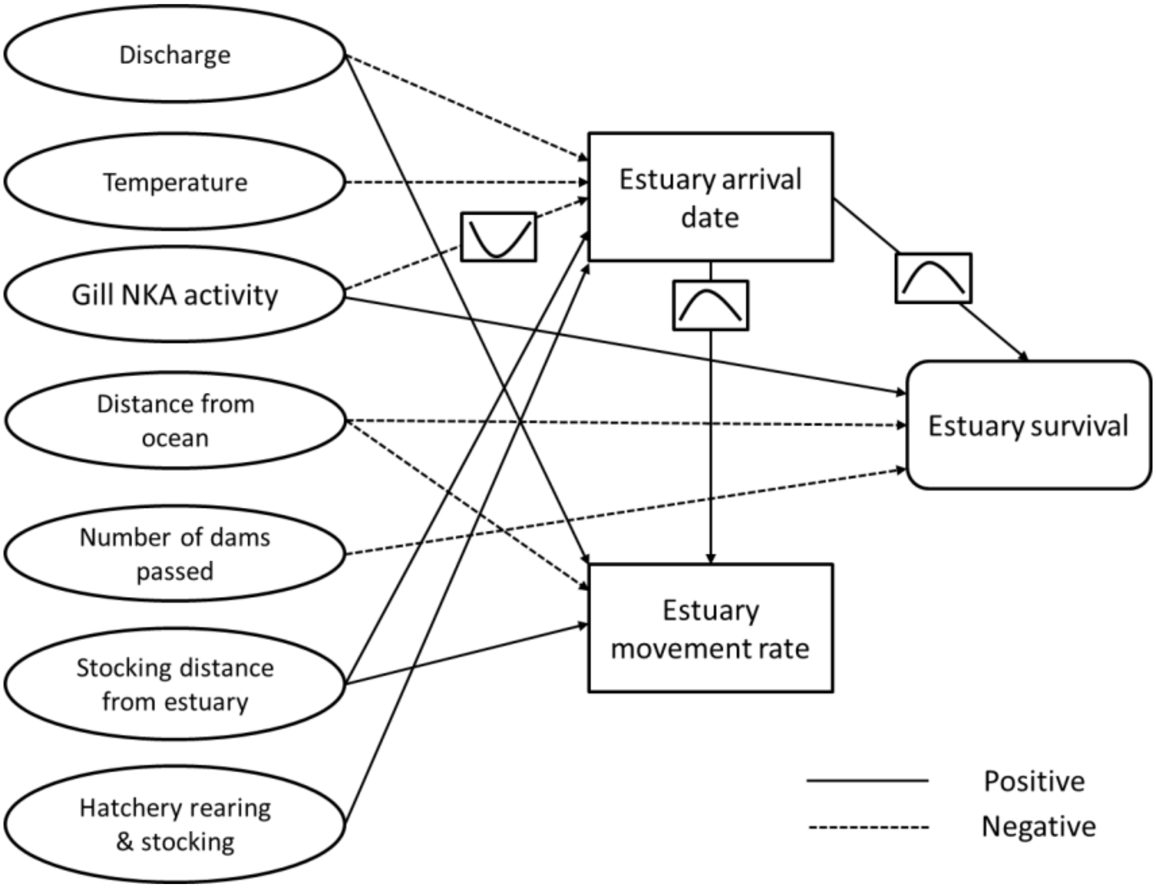


Figure 4.6. Diagram synthesizing major findings about factors affecting aspects of estuary migration of Atlantic salmon smolts during the present study. Dotted lines indicate negative relationships, whereas solid lines indicate positive influences. Assumed causality of relationships is indicated by the direction of arrows. Non-linear (quadratic) relationships are indicated by parabolas in boxes on top of arrows that describe relationships.

Successful and expedient passage of estuaries may become increasingly important based on relations between temperature, physiology, and survival (Figure 4.6). Migratory delay through dams (Keefer et al. 2012), and physical injuries incurred during dam passage (Music et al. 2011) have the potential to further promote loss of smolt characteristics (McCormick et al. 1999; Marschall et al. 2011) and impair osmoregulatory ability directly (Zydlewski et al. 2011). Recent modeling suggests the possibility that dams affect smolt migrations at spatially removed scales through migratory delay and potential mismatch in the timing of estuary arrival (McCormick et al. 2009; Marschall et al. 2011).

In the Penobscot River, dam-related estuary mortality is nearly as great as the *cumulative* mortality incurred as a result of dam passage in freshwater (Holbrook et al. 2011; Stich et al. 2014). Dam-related estuary mortality resultant from passage of a single dam during migration also has the potential to be greater than acute mortality incurred during passage of multiple hydropower projects in the Lower Penobscot River (Holbrook et al. 2011; Stich et al. 2014). This clearly highlights the importance of considering dam-related estuary mortality within regulatory frameworks used to assess effects of hydropower projects on fish passage.

The removal of Great Works and Veazie Dams will likely increase smolt survival in the Penobscot River Estuary by reducing dam-related mortality in the estuary (Figure 4.6), despite that these dams have only small effects (if any) on survival during dam passage (Holbrook et al. 2011; Stich et al. 2014). Indeed, dam-related estuary mortality resulting from each of these structures (6–7 %) was likely greater than mortality incurred during passage (0–1 %) of the facilities Holbrook et al. 2011; Stich et al. 2014). Because both of these dams were less than 12 h travel to the estuary, I hypothesize that their effects also may have been greater than dams located further upstream but this remains to be tested. Prior to removal of Veazie and Great Works Dams, 88% of Atlantic salmon smolts passed both dams (Stich et al. 2014), and 100% of smolts passed Veazie Dam during my study (2005–2013). As such, I expect that estuary survival will increase as a result of the removal of Veazie Dam alone (6%). Additional increased survival is expected for 88% of emigrating smolts due to Great Works Dam removal (6%). With the removal of these two dams, 20 km of lotic habitat was restored in the lower main-stem of the Penobscot River below the now lowermost dam (Milford Dam). This resulted in the opportunity to stock hatchery-reared smolts in the main-stem below any dams during the 2014 smolt run.

Smolts incur relatively high rates (c. 10% per km) of acute mortality at main-stem dams compared to background mortality in the Penobscot River (c. 1% per km), resulting in total loss of 40-60% of fish prior to arrival in the estuary (Holbrook et al. 2011; Stich et al. 2014). Fish passing the most dams (nine) during the present study experienced 40% increased dam-related estuary mortality compared to fish passing the fewest dams (two). Smolts stocked in 2014 passed no dams, eliminating dam-related mortality in the estuary (Figure 4.6). However, stocking nearer to the estuary may require closer attention to the physiological development of smolts (Figure 4.6) and environmental conditions (Figure 4.6). Similarly, there are potential ramifications for adult homing that result from stocking further downstream (Gorsky et al. 2009).

The number of smolts exiting the Penobscot River Estuary is expected to increase (by 12%) based on reductions in dam-related estuary mortality following the removal of the two lowermost dams. Through improvements in estuary survival by stocking below dams and monitoring smolt gill NKA activity I expect that the proportion of fish exiting the estuary each year could approximately double in a best-case scenario. Based on lack of differential survival in marine habitats after leaving Penobscot Bay (Sheehan et al. 2011), and the fact that patterns in marine mortality are similar among North American stocks (Friedland et al. 2003; Mills et al. 2013), this gain would likely translate directly to increases in returning adults.

CHAPTER 5

PHYSIOLOGICAL PREPAREDNESS AND PERFORMANCE OF *SALMO SALAR* RELATED TO BEHAVIORAL SALINITY PREFERENCES AND THRESHOLDS

INTRODUCTION

Many populations of Atlantic Salmon *Salmo salar* L. 1758 have been extirpated or are currently in decline (Parrish et al. 1998). Effects of dams in fresh water, pollution, and elevated marine mortality all have been cited as drivers of population decline and persistently low levels of abundance in recent years (NRC, 2004; Chaput 2012). These factors are related to the complex suite of physiological, morphological, and behavioral changes that *S. salar* undergo during smolting (McCormick et al. 1998). The physiological preparation of *S. salar* smolts for hypo-osmoregulation in marine environments has been widely studied from hormonal control of smolting and physiological development of salinity tolerance (Zaugg and Wagner 1973; McCormick et al. 1995; Hoar 1998) to behavioral initiation of migration (Sykes et al. 2009; Zydlewski et al. 2005, 2014) and estuary behavior and survival (Halfyard et al. 2013; Stich et al. in review). Smolting is physiologically regulated by circannual rhythms in photoperiod and temperature, which cue a suite of endocrine-driven changes to physiology, morphology and behavior (McCormick et al. 1998). The synchrony of these changes results in annual migrations from freshwater rivers to the ocean when a threshold set of conditions has been reached. The timing of these migrations is critical for successful ocean entry, and small changes in performance during this period can have major individual (e.g., death), and population consequences.

High mortality of *S. salar* smolts has been observed during passage through estuaries (Holbrook et al. 2011; Kocik et al. 2009), fjords (Dempson et al. 2011; Thorstad et al. 2012*a*), and near-coastal waters (Lacroix 2008; Thorstad et al. 2012*b*). This mortality recently has been related to physiology, experiences during freshwater migration, and behavior of individual smolts (Schreck et al. 2006; Halfyard et al. 2013; Stich et al. in review) as well as predation upon smolts (Hawkes et al. 2013) during the early marine phase of migration. The results of these studies underscore the importance of physiological, morphological, and behavioral preparations for successful estuary passage.

The timing of physiological and behavioral smolting and the timing of estuary entrance are instrumental in determining the success of smolts during estuary passage (Thorstad et al. 2012*b*). Environmental control of smolting results in a physiologically enhanced period for saltwater (SW) entry known as the physiological smolt window (McCormick et al. 1998). The timing of the physiological smolt window overlaps temporally with an ecologically opportune window for SW entry (McCormick et al. 1998). An ‘ecological smolt window’ can be defined as a period during which temperature (McCormick et al. 1999), abundance of predators (Kocik et al. 2009; Halfyard et al. 2013), and presence of sympatric migrants (Svenning et al. 2005) presumably act together to facilitate increased survival during estuary passage (McCormick et al. 1998). The overlap (match) between physiological and ecological smolt windows confers higher smolt survival during estuary passage than mismatch.

The various neuroendocrine controls of physiological smolting have been investigated in great detail during the past several decades, and it is accepted that multiple endocrine systems are involved with various aspects of smolting (McCormick et al. 1998; McCormick 2013). One reliable indicator of migratory urge and physiological development of smolts is an enzyme used in ion exchange; gill Na^+, K^+-ATPase (NKA) activity (McCormick et al. 1987; McCormick et al. 1989). The specific activity of the SW isoform of gill NKA is increased during smolting (McCormick et al. 2009), and differentiation in the cells of gills results in increased abundance of this ion-transport enzyme (McCormick 2013). Consequently, increased gill NKA activity is a useful indicator of osmoregulatory performance following exposure to SW. Recently, survival of *S. salar* smolts during estuary migration also has been related to activity of gill NKA activity immediately prior to migration (Stich et al. in review), highlighting the importance of physiological development for successful SW entry.

While *S. salar* smolts develop features for SW acclimation during migration, the rate at which smolts move through estuaries is controlled at the individual and may relate to variability in the timing of physiological development and environmental conditions experienced. These differences likely manifest during estuary migration through behavioral responses to SW upon estuary entry, and smolts may exhibit specific preferences or movement behaviors as a result of individual variability. However, individual variability in behavior observed during migration of (especially vertical distribution of fish) may be modified based on trade-offs between ion-regulation, energetic demands, and predator avoidance. The reasons for vertical movements by postsmolts during migration still are not well understood, although a number of reasons (selection of environmental gradients, predator avoidance, and feeding behaviors) have recently been suggested (Davidsen et al. 2008; Plantalech Manel-La et al. 2009; Renkawitz et al. 2012). Furthermore, it has been hypothesized that vertical movements, as well as downstream movement through estuaries, are mediated by diurnal and/or tidal rhythms (McCleave 1978; Davidsen et al. 2008) that might result in reduced energetic investment during seaward migration.

Timely information about how migratory success of fish might be affected by synergisms or mismatches between physiology and behavior during migration could have important implications for ongoing conservation and management efforts surrounding *S. salar*. This is particularly true in light of high marine mortality in recent decades (Mills et al. 2013), much of which is thought to occur during the early marine phase of migration (Friedland et al. 2003), but often is not separately accounted for because marine survival usually is estimated from smolt-to-adult return rates. The goal of this study was to explore the ontogeny of salinity preferences of *S. salar* smolts through laboratory experiments and to use the observed patterns to better understand smolt behavior during estuary migration in the Penobscot River Estuary, Maine. The specific objectives of this study were 1) to determine whether individual variability in selection of fresh or salt water by *S. salar* smolts in laboratory experiments was related to seasonal timing (ontogeny), gill NKA activity, and/or osmoregulatory performance (measured as change in NKA activity and plasma osmolality) of *S. salar*, and 2) to describe spatial and temporal patterns in depths and salinities used during estuary migration by acoustically tagged Penobscot River smolts with respect to physiological status and tidal influences in a natural system.

METHODS

Laboratory protocol

All fish used in the laboratory study were hatchery-reared, 18-month old *S. salar* smolts from the U.S. Fish and Wildlife Service (USFWS) Green Lake National Fish Hatchery (GLNFH) in Ellsworth, Maine, USA. This facility rears all of its fish in FW. Three groups of fish were transported from GLNFH to the University of Maine

Aquaculture Research Center, the first on 2 April (presmolts), the second on 6 May (smolts), and the third on 2 June (postsmolts) 2014. These groups were used to represent the variability in the seasonal timing of the Penobscot River smolt run, in which the mean annual peak of estuary arrival date is 9 May (S.D. = 8 days; Stich et al. in review). Fish were immediately transferred to a circular, 1,890-L, flow-through holding tank that contained aerated well water within 1°C of the transport tank, and were sampled for gill NKA activity (see below) about one hour after transfer. Temperature of holding tanks was gradually acclimated to reach temperatures identical to those used in salinity choice tanks described below. Throughout the course of the study, mean temperature in outdoor rearing tanks at GLNFH was 3.14 °C (S.D. = 0.08 °C) for the presmolt group, 9.57 °C (S.D. = 1.57 °C) for the smolt group, and 16.26 °C (S.D. = 0.69 °C) for the postsmolt group.

Sampled fish were anesthetized using a 100 $mg \cdot L^{-1}$ solution of tricaine methanesulfonate (MS-222) adjusted to pH 7.0 with 20-mmol $NaHCO_3$. For each fish, fork length (L_F, in mm) and mass (g) were measured. A nonlethal gill biopsy (4-6 filaments) was taken from the front, left gill arch of each fish prior to tagging. Individual biopsies were stored at -80°C in 100 μL SEI buffer (250 mM sucrose, 10 mM Na_2-EDTA, 50 mM imidazole) for later analysis of gill Na^+, K^+-ATPase (enzyme code 3.6.3.9; IUBM 1992) activity (expressed as $\mu mol\ ADP \cdot mg\ protein^{-1} \cdot h^{-1}$) using the method of McCormick (1993). Concentration of NADH at 25°C and 340 nm was used to measure kinetic rate of ouabain-inhibitable ATP hydrolysis, and protein concentration in gill samples was determined using the bicinchoninic acid (BCA) method (Smith et al.

1985). Gill samples from each individual were analyzed in triplicate for NKA activity and protein concentration.

After a biopsy was taken a small incision (*c.* 1–2 mm) was made offset from the ventral line, about 1-cm posterior to the pectoral fins and a passive integrated transponder (PIT) tag measuring 2 × 12 mm and weighing 0.1 g (Model TX1411L; Destron Fearing, St. Paul, Minnesota) was gently inserted through the opening for the purpose of individual identification. All tags and surgical equipment were disinfected in a 1 % solution of Chlorhexidine solution (Bimeda Inc., Irwindale, California; http://www.bimedaus.com/). Fish were allowed a recovery period in the holding tanks (see above) with a minimum time of 24 hours (up to 6 days) prior to any subsequent behavioral or physiological testing.

Fish were transferred individually to an automated salinity choice tank (see below) following the recovery period. Salinity choice tests were conducted in 8-hour time blocks to enable the use of a single SW-challenge tank for each time block. For testing, each fish was tested on its own in the salinity choice tank for 1 hour, after which the fish was immediately transferred to SW (salinity = 35) for 16–24 hours in an aerated 100-liter SW-challenge tank. Complete water exchange in the SW-challenge tank occurred after each 16-24 hour SW-challenge period, and the salinity choice system tank flushed after each 8-hour testing period.

After 16–24 hours of exposure to full SW, fish were anesthetized as described above, and a second gill biopsy was taken from the same gill arch of each fish (described above). A blood sample was then taken from the caudal vein of each fish using a 1-mL, 25-gage ammonium-heparinized syringe. The blood sample was transferred to a 1.8-mL centrifuge tube and centrifuged at 2,000 × gravity (*g*) for 5 minutes. Plasma was transferred to a 0.6-mL centrifuge tube, immediately frozen on dry ice, and stored at -80°C for later analysis. Plasma osmolality was measured with an Advance Instruments Model 3200 freezing-point-depression osmometer (Advanced Instruments, Inc., Norwood, Massachusetts), and was expressed in miliosmolality (mOsm). After terminal physiological samples were collected, all fish were euthanized by application of a lethal dose (250 $mg \cdot L^{-1}$) of MS-222 adjusted to pH 7.0 with 20-mmol $NaHCO_3$. The trial period for each group of 60 test smolts lasted about 7 days.

Salinity choice system

The tank system used to assess salinity choice by *S. salar* smolts, in addition to the tanks used for the SW challenge, were housed in a room that was separated from holding tanks and laboratory equipment. The choice-tank room was kept at the same temperature as holding tanks (see above), and maintained on simulated natural photoperiod. All electronic equipment in the choice-tank room was controlled externally through an automated computer system to minimize disturbance to test subjects during salinity choice experiments and SW-challenge.

The ‘shuttlebox’ system (Loligo Systems, Inc.) used to test salinity choice by *S. salar* smolts in the laboratory was previously described by Serrano et al. (2010), with minor modifications (e.g., smaller tank size and tubing) during the present study (Figure 5.1). The choice tank consisted of two circular compartments, each of 41 cm diameter and 19 cm depth. The two circular tanks were connected by a rectangular trough 10 cm long, and 7.5 cm wide to allow movement between compartments. The tank was illuminated from below with two infrared (IR) utility lamps. Movement of smolts between compartments was tracked using an IR camera (uEYE USB camera, model UI-164xSE-C, Imaging Development Systems, Obersulum, Germany), and analyzed in ShuttleSoft software (version 2.6.0 , Loligo® Systems, Tjele, Denmark) on a remote computer. The software recorded position of the fish on a Cartesian grid once per second and assigned fish position to one of the two compartments of the choice tank. To narrow the field of observation open to analysis by the IR camera and ShuttleSoft, a ‘mask’ was constructed within ShuttleSoft that restricted analysis to the dimensions of the tank. To improve resolution of IR analysis, the camera was calibrated for each fish by adjusting the location within the IR spectrum that was being analyzed, as well as the bandwidth of the spectrum being observed. Measurements taken by the camera were calibrated (in pixels) against the length of the connecting trough prior to each run. Calibration ($mm \cdot pixels^{-1}$) was conducted such that the ratio of millimeters to pixels was standardized among trials ($0.74\ mm \cdot pixels^{-1}$).

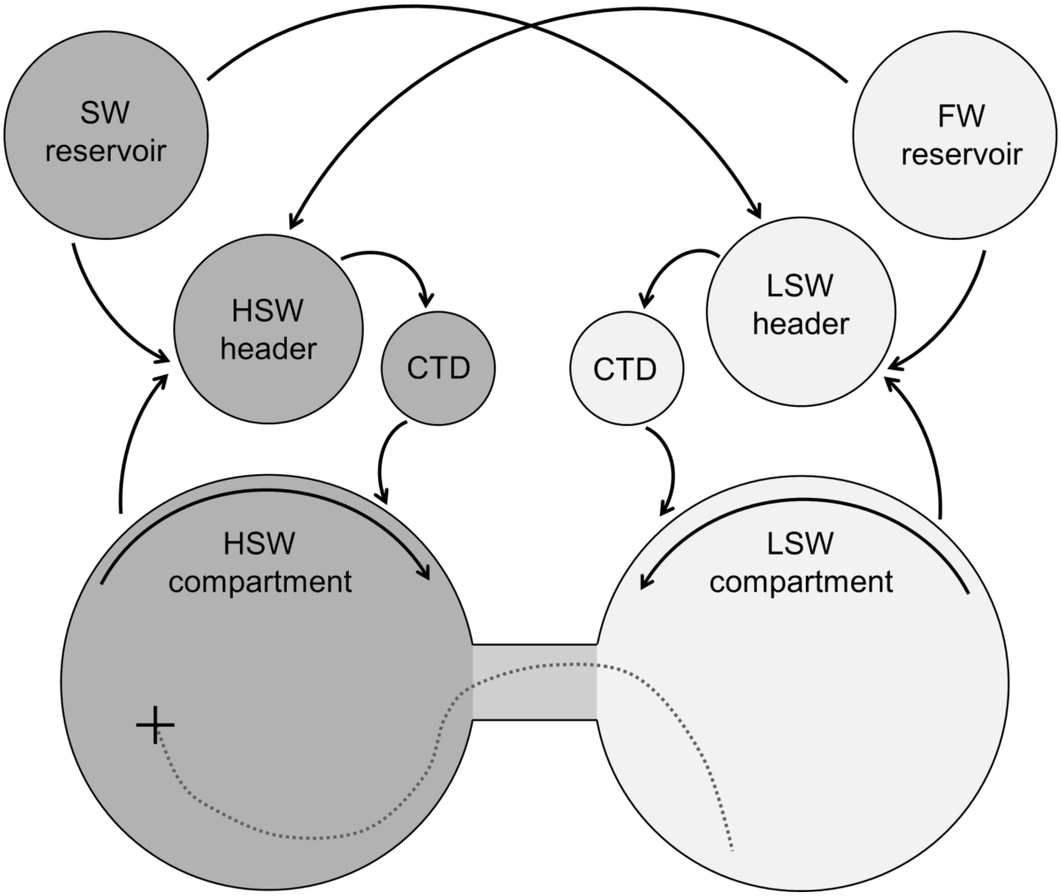


Figure 5.1. Schematic drawing of the tank system used to assess salinity choice by hatchery-reared *S. salar* smolts in the laboratory. Direction of water flow is indicated by solid arrows. The dotted line depicts an example fish track and the cross indicates the position of the fish. High-salinity water (HSW) and low-salinity water (LSW) are indicated by dark gray (HSW) and light gray (LSW) fills.

Salt concentration in the choice tank was horizontally stratified so that one compartment contained high-salinity water (HSW), and the other compartment contained low-salinity water (LSW). Inflow to each compartment was gravity-fed from cuboid header tanks, and outflow from each was controlled by constant-rate, continuous pumps (EHEIM Universal 300, EHEIM Aquatics Group, Deizisau, Germany) that returned water to the header tank for each compartment (HSW header tank, and LSW header tank). Water in the header tanks was partially re-circulated in this manner, with overflow drains located 5 cm from the top of the header tanks. Salinity in header tanks was monitored continuously using WTW-3300 conductivity and temperature meters (WTW [Wissenschaftlich-Technische-Werkstätten], Weilheim, Germany). The target salinity for each compartment (LSW and HSW) was established within ‘zones’ in the ShuttleSoft software program. Zones used for this study were circular, and excluded the connecting trough (movement through the trough was recorded, but was not assigned to a zone).

The salinity in the header tank for each zone was transmitted to Shuttlesoft software on a remote computer using a universal serial bus (USB) connection. When the salinity in the header tanks reached upper or lower threshold concentrations for their respective zones, salinity in the corresponding header tank was either increased or decreased accordingly through the activation of pumps in large fresh water (LSW) and salt water (HSW) reservoir tanks that were controlled remotely by a relay box (NI-USB-6009-DAQ-M, National Instruments, Austin, Texas) integrated into the ShuttleSoft software. Water from the reservoir tanks was pumped into the header tanks until the desired salinity was reached within a tolerance of ± 1, as measured by conductivity meters. The mean (S.D.) salinity across all trials was 31.8 (3.7) for the HSW compartment and 2.3 (3.2) for the LSW compartment.

The LSW reservoir tank contained well water that was held constant at approximately 10°C throughout the duration of this study and was operated as flow-through. Temperature was monitored continuously using the temperature meters described above. To achieve homogeneous temperatures (± 1°C) between the LSW and HSW reservoir tanks (as well as LSW and HSW header tanks and compartments of the choice tank), well water was run through a 30-m length of cross-linked polyethylene (PEX) tubing constantly as a means of heat exchange and LSW well water was periodically added to the HSW header tank as the HSW reservoir tank was filled between salinity choice trials. Both the LSW and HSW reservoirs (diameter = 0.75 m, height = 1.6 m) were polyethylene conical tanks (Chem-tainer Industries Inc., New York, New York) with 5-cm drains located approximately 15 cm from the top. Water in both tanks was aerated vigorously and continuously to remove gasses that are common to groundwater sources.

Experimental design for laboratory study

Salinity choice was observed in the three discrete smolt groups (‘presmolts’, ‘smolts’, and ‘postsmolts’) during spring 2014 using 54 to 60 individually tested fish for each group. Half of the fish in each group were tested during daylight hours and half during hours of darkness (using local sunrise/sunset times and broad spectrum lights on simulated natural photoperiod) to determine if salinity choice was directly related to daylight. I alternated the starting salinity (LSW or HSW) between compartments of the choice tank, and the diel timing (light or dark), such that half of the fish tested at night and half of the fish tested during daylight hours started in HSW. Fish always entered the choice tank on the same side to account for possible tank effects on the selection of compartments. This resulted in an initial sample size of about 15 fish in each level of a full factorial design (group × starting salinity × diel timing).

During the first SW challenge (5 presmolts), I attempted to use salinity of 40, but it was determined that this concentration was too high as it resulted in 100% mortality. All subsequent SW challenges were conducted at salinity of 35.

Laboratory data analysis

Two behavioral responses of *S. salar* were measured in the salinity choice tank as indices of SW choice: proportional use of the HSW compartment, and the maximum continuous time spent in the HSW compartment. The proportion of time spent in the HSW compartment of the salinity choice tank (‘preference’) was calculated as the amount of time that each fish occupied the HSW compartment of the choice tank divided by the total duration of the trial for that individual. Maximum continuous time spent in the HSW compartment, or ‘residence’ was calculated for each fish based on consecutive relocations (at one-second intervals) within the HSW compartment of the salinity choice tank. For all fish exposed to SW challenge, physiological performance was assessed in two ways. First, change in gill NKA activity was calculated as the difference in gill NKA activity at tagging and gill NKA activity following SW challenge. This difference is considered an indicator of upregulation of gill NKA activity. Second, plasma osmolality after exposure was measured directly (described above).

The effects of treatment group (presmolts, smolt, and postsmolt), daylight (night=0, day=1), starting compartment (LSW=0, HSW=1), time since tagging (recovery), initial gill NKA activity, change in gill NKA activity (ΔNKA), and plasma osmolality, on each of the response variables were tested using generalized linear models (GLM: Montgomery et al. 2006) in R, version 3.1.0 (R Development Core Team 2014). Fish preference is on a binary scale (0, 1) and as such a logit-transformation ($log\left[\frac{p}{(1-p)}\right]$) was used for analysis of this response. Because of the wide range of variability in residence, and because most of the values were small (i.e., variable was right-skewed), residence was analyzed assuming a negative binomial error structure and the GLMs for the residence models used a $\log_e$ link function. Approximation of a variance inflation factor($\hat{c}$) indicated that model structures were appropriate for the analyses conducted ($\hat{c} \approx 1.00$, Montgomery et al. 2006). The total, final sample sizes used for each group during analysis of behavior in the salinity choice tank and subsequent performance during SW challenge were 53 (presmolts), 60 (smolts), and 56 (postsmolts).

Competing models, for both preference and residence, were constructed to test the effects of explanatory variables on each of the behavioral responses described above. An information-theoretic approach using Akaike's information criterion corrected for sample size (AIC_c; Burnham and Anderson 2002) was used for model selection. The relative support for candidate models was evaluated as the difference in AIC_c between the best model and each i^{th} model (Δ_i), and the relative probability of each model being the best was represented using AIC_c weights (w_i; Burnham and Anderson 2002). Models for which $\Delta_i \leq 2.0$ were considered to have similar support to the best model in each candidate model set (Burnham and Anderson 2002). Covariate effects were considered to be statistically significant if 95% confidence intervals for the estimated regression coefficient did not overlap zero.

Analysis of variance (ANOVA) was used to characterize differences in gill NKA activity, change in gill NKA activity following SW challenge, and plasma osmolality between the presmolts, smolts, and postsmolts. Tukey-Kramer pairwise comparisons of means were used to determine group-level differences with a significance assumed at $p < 0.05$ (Zar 1999).

Collection of field data

The Penobscot River Estuary (see Figure 5.2) spans approximately 45 km from the mouth of the estuary (rkm 0) to the head of tide (rkm 45; Haefner 1967). The water column in the estuary is uniformly fresh at the head of tide (Imhoff and Harvery 1972), has considerable mixing of fresh and salt water in the middle of the estuary between rkm 30 and rkm 0 (Figure 5.1; Seiwell 1932), and displays stratification of fresh water and salt water occurs in the lower estuary and bay (Imhoff and Harvey 1972). Given the vertical stratification within the Penobscot River Estuary based on temperature and salinity gradients (Haefner 1967), depths at which fish migrate dictate exposure of individual smolts to elevated salinities. Due to salinity gradients in the Penobscot River Estuary, depth used by smolts likely exposes them to different salinities.

Ten hatchery-reared *S. salar* smolts from GLNFH were acoustically tagged with depth-sensing transmitters and tracked during migration from the head of tide in the Penobscot River Estuary to Penobscot Bay during spring 2013. All *S. salar* smolts were tagged using model V9P-6L tags (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada). Mean (± S.D.) L_F of acoustically tagged smolts was 192 (± 12) mm, and mean mass was 70 (± 13) g. The tags were 39 mm length, 9 mm diameter, and weighed 2.2 g in water. Estimated battery life for acoustic tags was 60 days.

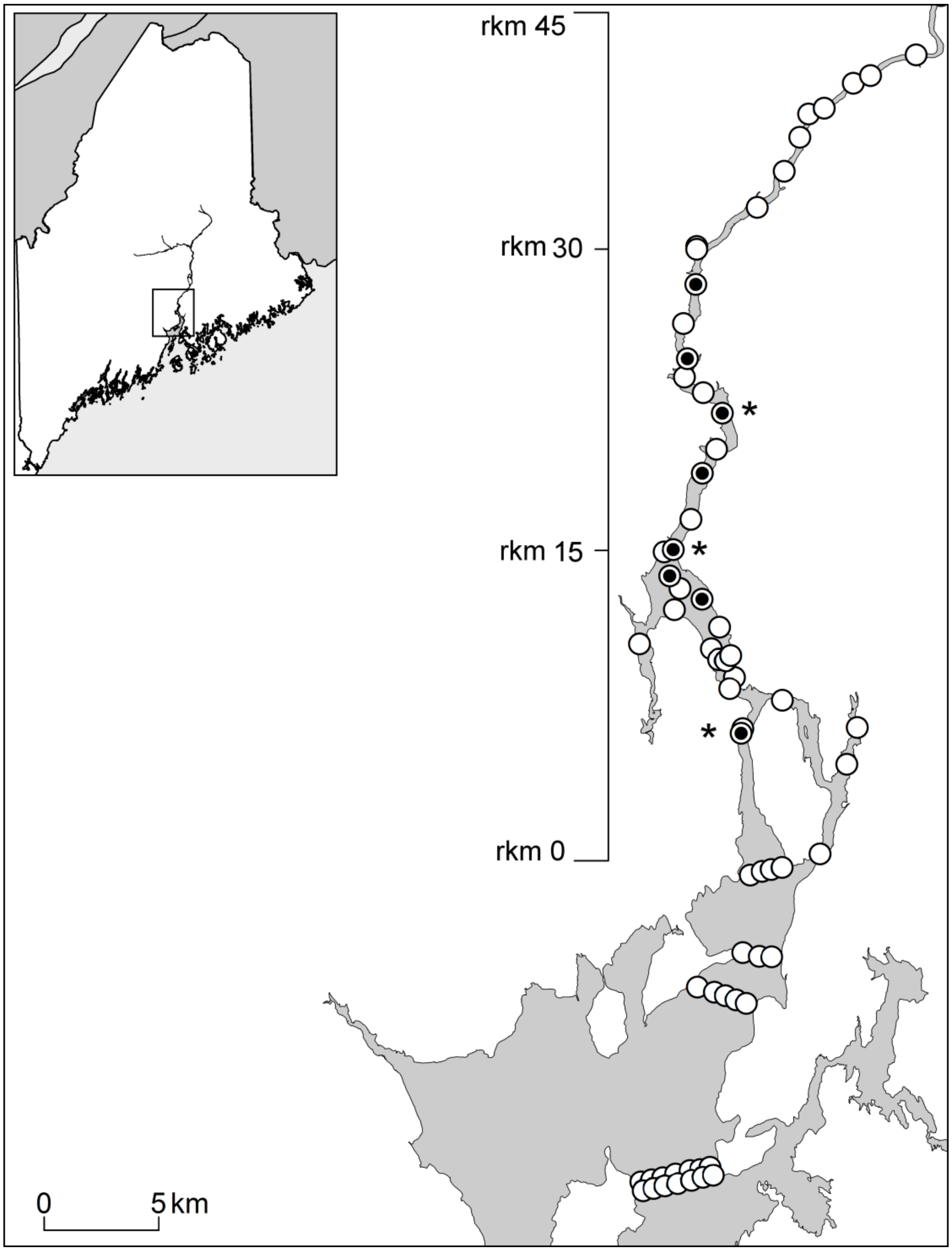


Figure 5.2. Map showing the location of VR2-W acoustic receivers and corresponding river kilometer (rkm) of deployment. Acoustic receiver locations are indicated by circles. Locations at which a conductivity, temperature, and depth sensor (CTD) was deployed on the river bottom are indicated by circles with bulls eyes. Receiver locations at which CTDs were also deployed at the top the water column are indicated by *.

Fish were tagged using previously described methods (Holbrook et al. 2011; Stich et al. 2014). Smolts were anesthetized using a 100 mg·L-1 solution of MS-222 buffered to pH 7.0 (using 20-mmol $NaHCO_3$), L_F (mm) and mass (g) were measured. A nonlethal gill biopsy (4-6 filaments) was taken from the front, left gill arch of each fish prior to tagging. Individual biopsies were stored at -80°C in 100 µL SEI buffer (250 mM sucrose, 10 mM Na_2-EDTA, 50 mM imidazole) for later analysis of NKA activity using procedures described above. A small (1-cm) incision was made offset from the ventral line and about 1-cm posterior to the pelvic fin girdle. An acoustic tag was inserted intraperitoneal and the incision was closed with two interrupted knots using 4-0 absorbable vicryl sutures (Ethicon, Somerville, New Jersey, USA).

Salmo salar smolts were tracked during migration using an array of stationary VR2-W acoustic receivers (Amirix Vemco Ltd., Halifax, Nova Scotia, Canada). The receiver array was deployed prior to tagging in a cooperative effort between the US Geological Survey (USGS) Maine Cooperative Fish and Wildlife Research Unit, the University of Maine, and the National Oceanic and Atmospheric Administration (NOAA) National Marine Fisheries Service (NMFS) Northeast Science Center. The array extended from the head of tide (river kilometer [rkm] 43.5) to Penobscot Bay (rkm -15), and included a total of 66 acoustic receivers (Figure 5.2). Acoustic receivers deployed in the Penobscot Estuary were moored to 45-kg concrete anchors deployed on the river bottom, and receivers deployed in the bay were tethered approximately 10 m below the surface. Where necessary, multiple receivers were deployed across the estuary to achieve adequate coverage in a given deployment location. All detections at receivers within these locations were pooled as a single site for analyses.

Conductivity, temperature, and depth loggers (DST-CTD, Starr-Oddi, Gardabær, Iceland) were deployed at eight receiver locations throughout the estuary in configurations of either one or two loggers (Figure 5.2). In locations at which one CTD was used, the logger was attached to an acoustic receiver mooring near the bottom of the water column. Where multiple DST-CTDs were deployed at a receiver location, one CTD was deployed in the top 2 meters of the water column and the other CTD was deployed about 0.5 m from the bottom.

Analysis of field data

Patterns in depth use during migration were investigated using data from acoustic tags. To determine if depth use was related to the presence of SW in the estuary, the relationship between depth use and distance from the mouth of the estuary was estimated using a generalized linear mixed-effects model (GLMM; Zuur et al. 2009). Individual fish identity was included as a random effect on the intercept because an unequal number of measurements were collected for each fish at each location. Because a non-linear relationship between depth use and estuary location was hypothesized, position within the estuary was represented using a linear term for estuary rkm, and a second-order function (i.e., quadratic) was included. Similarly, a GLMM with random effect of fish was used to determine whether depth used by individual fish was related to tidal cycles (incoming or outgoing) for the week during which fish moved through the estuary. The link function used in each of these models was the inverse Gaussian, because the response (depth) was continuous but constrained to be greater than zero (i.e., fish could not occupy negative depths within the water column).

The probabilities of encountering varying salinities at a given rkm in the estuary were modeled using logistic regression and salinity data for the locations with CTDs in the lower estuary (Figure 5.2). The probabilities of encountering salinities greater than concentrations from 0 to 15 (0, 5, 10, 15) were used to predict presence of SW in the Penobscot Estuary. For each SW concentration examined, water of that concentration was considered to be present (1) if a CTD recorded salinities greater than or equal to the concentration. If salinity equal to or greater than the concentration was not recorded at a site, SW greater than the corresponding concentration was considered to be absent (0). Salinity of 10 was assumed to be near isosmotic. From each salinity used, the probability that SW concentrations greater than that salinity existed at each receiver location was estimated from a logistic regression model. These probabilities were then plotted and used to assess average position of the salt wedge in the Penobscot Estuary visually and qualitatively compare salinity gradients to apparent changes in fish depth and movement rate.

Travel time through the FW reaches of the estuary was compared to gill NKA activity of individual smolts using simple linear regression to test the null hypothesis that gill NKA activity had no effect on travel time through FW (from release [rkm 43.5] to first recorded SW (rkm 30). Similarly, the relationship between gill NKA activity and SW travel time was tested using simple linear regression. A significance level of $\alpha < 0.05$ was used for both tests. Finally, the vertical and horizontal movements of fish were plotted against date and tidal cycles to assess potentially interesting patterns in behavior related to residency time in FW and tidal cycles in SW. Because tidal and diurnal cycles were confounded over the week of the field study, and because the laboratory experiment did not indicate effects of diurnal cycle on salinity preference or residence, I did not examine differences in depth use during day and night.

RESULTS

Laboratory experiment

Laboratory assays indicated significant differences between treatment groups (presmolts, smolts, postsmolts) of *S. salar* in gill NKA activity prior to tagging (ANOVA, $F = 31.21$, df = 2, 166, $P < 0.001$), change in gill NKA activity over the trial (ANOVA, $F = 4.28$, df = 2, 166, $P < 0.001$), and plasma osmolality (ANOVA, $F = 40.97$, df = 2, 166, $P < 0.001$) following SW challenge (Figure 5.3 and Table 5.1). Mean gill NKA activity (expressed as μmol ADP·mg protein^{-1}·h^{-1}) was significantly lower in presmolts (median = 3.11, S.D. = 1.35) than smolts (median = 5.78, S.D. = 1.95), and postsmolts had gill NKA activity (median = 3.66, S.D. = 1.79) intermediate to presmolts and smolts (Figure 5.3a). Similarly, presmolts increased NKA activity (median = 1.41, S.D. = 1.86) significantly more than smolts (median = 0.16, S.D. = 2.81), and postsmolts were intermediate in observed up-regulation of gill NKA activity following SW challenge (median = 0.51, S.D. = 1.60; Figure 5.3b). Congruent with changes in gill NKA activity following SW challenge, plasma osmolality (mOsm) was significantly higher in presmolts (median = 377, S.D. = 27) after SW challenge than in smolts (median = 326, S.D. = 27) or postsmolts (median = 356, S.D. = 34; Figure 5.3c). Plasma osmolality was significantly higher in postsmolts than in smolts.

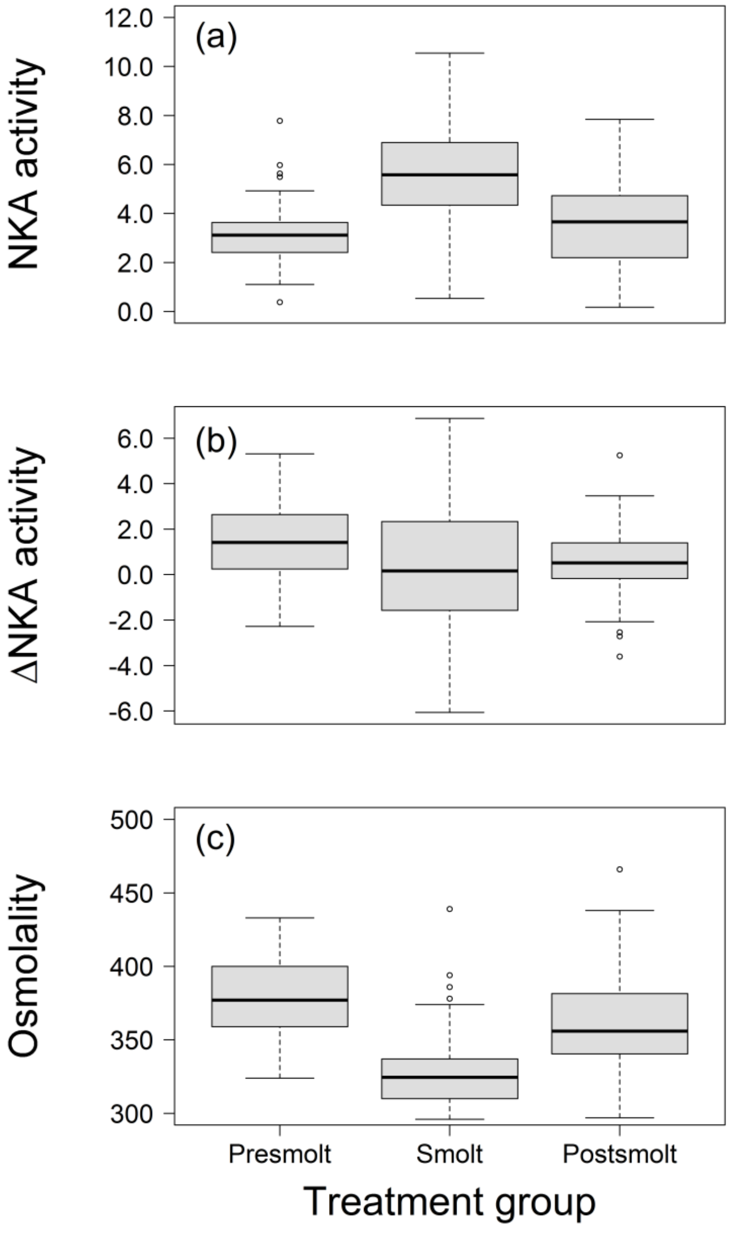


Figure 5.3. Ontogenetic changes in gill NKA activity and osmoregulatory performance. Differences in a) Gill Na^+, K^+-ATPase activity (NKA; μmol ADP·mg protein^{-1}·h^{-1}), b) change in NKA activity following SW challenge, and c) plasma osmolality for hatchery-reared *Salmo salar* smolts that were tested in salinity choice tanks early in the smolt run (Apr 3–Apr 9), in the middle of the smolt run (May 7–May 15), and late in the smolt run (Jun 4–Jun 10). Gray boxes represent 25–75% confidence intervals, and bold lines in the center of the boxes are median proportion of time spent in freshwater for each group. Whiskers indicate 95% confidence intervals.

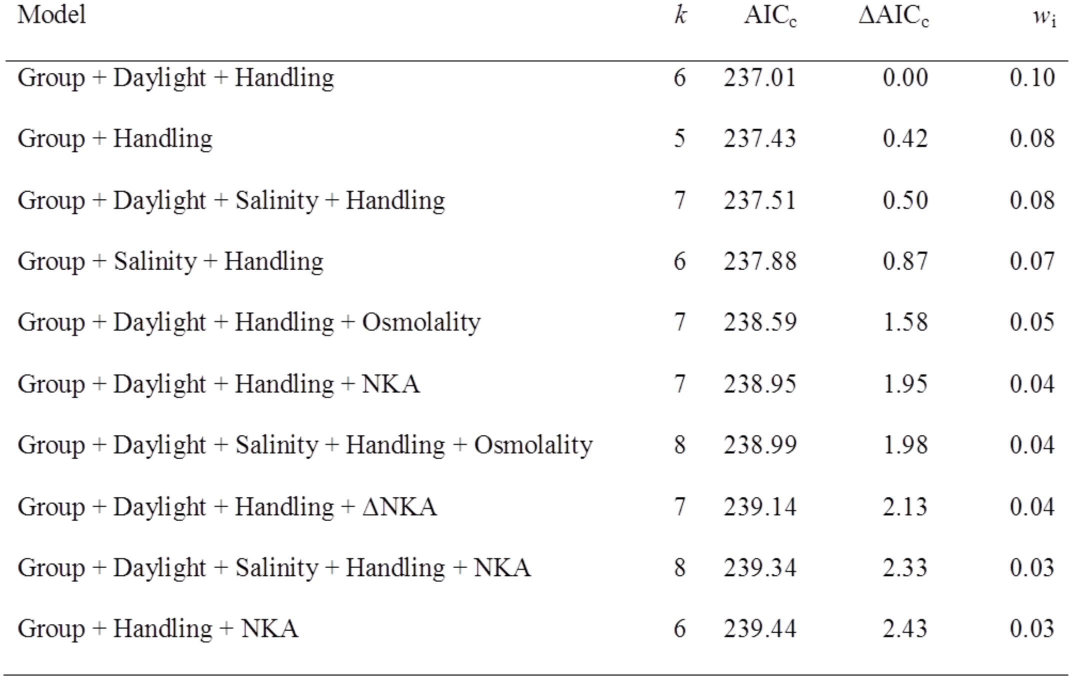
Table 5.1. Mean ± S.D. values for indicators of smolt development for fish used in the laboratory study. Symbols in table headings are defined as L_F: fork length (mm), mass (g), Fulton condition factor (K), initial and final gill NKA activity (μmol ADP·mg protein^{-1}·h^{-1}), and plasma osmolality (mOsm).

Developmental Stage	L_F	Mass	K	Initial gill NKA activity	Final gill NKA activity	Plasma osmolality
Presmolt	190 ± 12	74 ± 15	1.04 ±0 .06	3.1 ± 1.4	4.5 ± 1.5	380 ± 27
Smolt	190 ± 12	68 ± 12	0.99 ± 0.05	5.6 ± 1.9	5.8 ± 2.5	330 ± 27
Postsmolt	196 ± 10	69 ± 12	0.92 ± 0.07	3.6 ± 1.8	4.2 ± 1.4	362 ± 34

Of the fish exposed to a 24-hour, 35-ppt SW challenge, 2 mortalities were observed in the presmolt group (4%). This result was assumed to be due to reduced osmoregulatory capacity in those fish because it represented only 2 of 55 (< 4%) total fish remaining in the presmolt group and 2 of 8 (25%) of the fish in that specific SW challenge trial. In the presmolt treatment group, 13 fish (24%) exhibited moribund behavior and physical appearance following SW challenge, and 10 (18%) of the postsmolts similarly appeared to be moribund. Most fish that displayed signs of imminent mortality had extremely high (> 400 mOsm) plasma osmolality, and mean gill NKA activity of these fish (2.9 μmol ADP·mg protein^{-1}·h^{-1}) was lower than remaining fish (4.38 μmol ADP·mg protein^{-1}·h^{-1}) that did not show such signs (t-test, $t = 4.49$, df = 40, $p < 0.001$). There were no mortalities in the smolt group, and none of those fish exhibited the obvious signs of distress following SW challenge observed in fish from the presmolt and postsmolt groups.

Proportional use of the HSW compartment of the salinity choice tank (‘preference’) by fish was related to development and time after handling (Table 5.2). Presmolts used the saltwater compartment of the choice tank significantly less (mean = 0.37, S.D. =0.09) than postsmolts (mean = 0.44, S.D. = 0.09), and smolts were intermediate in their preference for SW (mean = 0.41, S.D. = 0.13; Figure 5.4a and Table 5.3). Preference for SW increased with time after initial sampling (Table 5.3). Fish tested at the end of each treatment group (*c*. 1 week) increased preference by about 10 % (95% C.I. = 3–17%) compared to fish tested 24 hours after tagging.

Table 5.2. Model selection for salinity preference in lab study. Model-selection statistics for GLMs used to test effects of treatment group (early, middle, or late), daylight (dark=0, light=1), starting salinity (Salinity: LSW=0, HSW=1), gill Na^+, K^+-ATPase activity (μmol ADP·mg protein^{-1}·h^{-1}) at tagging (NKA), change in NKA following SW challenge (ΔNKA), plasma osmolality (Osmolality), and time after tagging (Handling) on proportional use of salt water (‘preference’) by hatchery-reared *Salmo salar* smolts in salinity choice tanks. Symbols in table are defined as number of parameters (*k*), corrected Akaike-information criterion (AIC_c), the difference in AIC_c between the best model and the i^{th} model (Δ_i), and the relative probability that the i^{th} model is the best model in the candidate set (w_i).



Model	*k*	AIC_c	ΔAIC_c	w_i
Group + Daylight + Handling	6	237.01	0.00	0.10
Group + Handling	5	237.43	0.42	0.08
Group + Daylight + Salinity + Handling	7	237.51	0.50	0.08
Group + Salinity + Handling	6	237.88	0.87	0.07
Group + Daylight + Handling + Osmolality	7	238.59	1.58	0.05
Group + Daylight + Handling + NKA	7	238.95	1.95	0.04
Group + Daylight + Salinity + Handling + Osmolality	8	238.99	1.98	0.04
Group + Daylight + Handling + ΔNKA	7	239.14	2.13	0.04
Group + Daylight + Salinity + Handling + NKA	8	239.34	2.33	0.03
Group + Handling + NKA	6	239.44	2.43	0.03

Table 5.3. Covariate effects on preference and residence. Estimated regression coefficients, standard errors (S.E.), t-statistics, and p-value (*p*) for the best models of behavioral responses of hatchery-reared *Salmo salar* smolts in salinity choice tanks, including proportional use of the HSW compartment ('Preference'), and maximum continuous time spent in the HSW compartment ('Residence'). Variable names in parameter column are defined as in Table 5.2.

Response	Parameter	Estimate	S.E.	*t*	*p*
Preference	Group (presmolt)	-0.747	0.102	-7.333	< 0.001
	Group (smolt)	0.213	0.091	2.357	0.020
	Group (postsmolt)	0.342	0.092	3.720	< 0.001
	Daylight	-0.122	0.077	-1.586	0.115
	Handling	0.051	0.018	2.787	0.006
Residence	Group (presmolt)	3.683	0.218	16.894	< 0.001
	Group (smolt)	0.418	0.172	2.439	0.301
	Group (postsmolt)	0.128	0.154	0.830	0.293
	Daylight	0.355	0.128	2.757	0.006
	Handling	-0.053	0.031	-1.725	0.085
	NKA	0.254	0.044	5.811	0.010
	ΔNKA	0.131	0.035	3.716	0.114

The maximum continuous time (residence) spent in the HSW compartment of the salinity choice tank by *S. salar* was related to development, daylight, handling, gill NKA activity, change in gill NKA activity (ΔNKA) following SW challenge, and plasma osmolality (Table 5.4). Residence in the HSW compartment of the salinity choice tank was greater in smolts (mean = 286, S.D. = 482) than in presmolts (mean = 97, S.D. = 89) or postsmolts (mean = 123, S.D. = 131; Figure 5.4b). Fish occupied the HSW compartment of the salinity choice tank for longer continuous periods during darkness than during daylight hours (Table 5.3), but the mean difference was just 17 sec. (95% C.I. = 1–51 sec.). Smolts tested at the beginning of each treatment group exhibited increased residence, but the difference (mean = 13 sec., 95% C.I. = 4–14 sec.) was not significant at the test level (α = 0.05; Table 5.3). Fish with higher gill NKA activity at tagging exhibited greater residence compared to fish with lower gill NKA activity. Fish with the highest NKA activity continually resided in the HSW compartment 9 minutes longer (95% C.I. = 2–35 minutes) on average than fish with the lowest gill NKA activity (Figure 5.5a and Table 5.3). Similarly, *S. salar* with greater ΔNKA and plasma osmolality had greater residence in HSW than fish that performed more poorly during SW challenge (Figure 5.5b and Table 5.3), although ΔNKA was a better predictor of residence than was plasma osmolality. Fish that exhibited the greatest ΔNKA activity continuously occupied the SW compartment of the salinity choice tank for 80 seconds (95% C.I. = 22–223 seconds) longer than fish with lowest ΔNKA across all groups. Similarly, when ΔNKA was not included as an explanatory variable, fish that had the highest plasma osmolality following SW challenge reduced residence compared to fish with the lowest plasma osmolality, a difference of 76 seconds (95% C.I. = 2–163 seconds).

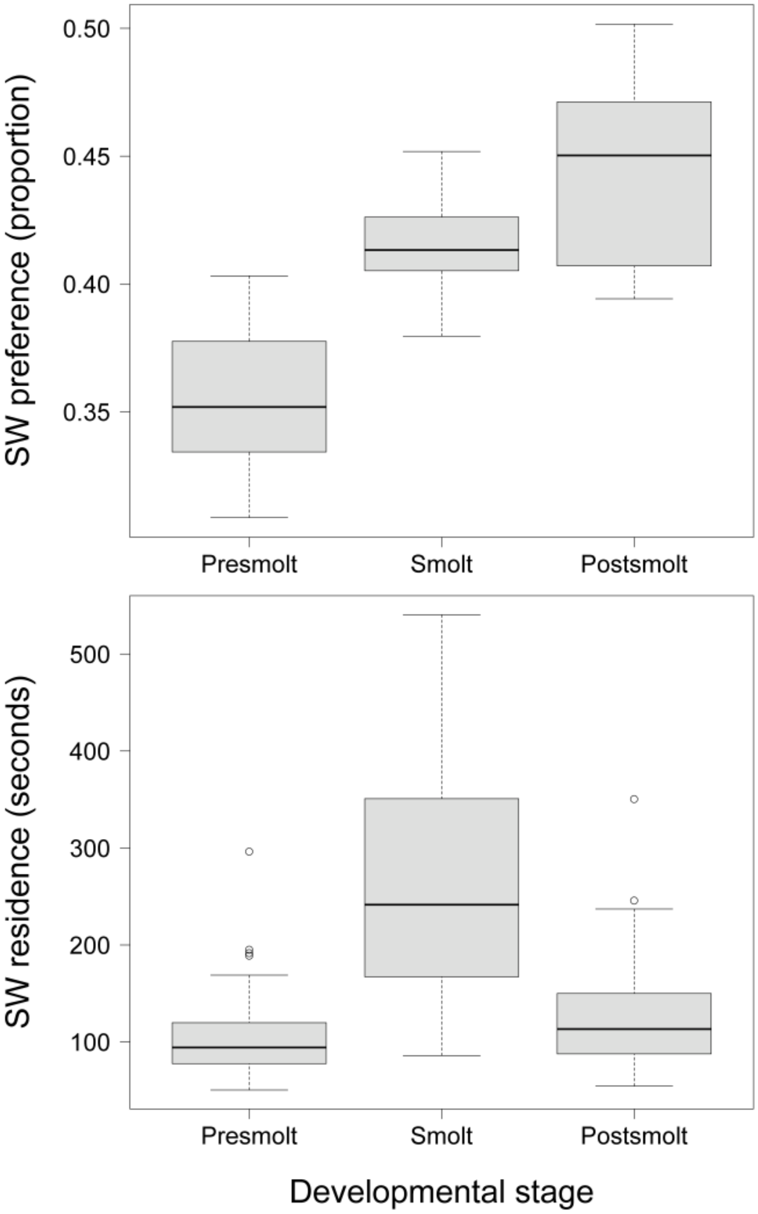


Figure 5.4. Ontogenetic shifts in salinity preference and residence time. Differences in a) proportional use of HSW in salinity choice tank (‘preference’) and b) maximum continuous time in HSW compartment of salinity choice tank (‘residence’) by hatchery-reared *Salmo salar* smolts tested early in the smolt run (Apr 3–Apr 9), in the middle of the smolt run (May 7–May 15), and late in the smolt run (Jun 4–Jun 10). Gray boxes represent 25–75% confidence intervals (C.I.), and bold lines in the center of the boxes are median proportion of time spent in freshwater for each group. Whiskers indicate 95% C.I.

Table 5.4. Model-selection statistics for GLMs used to describe maximum continuous time spent in SW compartment of salinity choice tank (‘residence’) by *Salmo salar* smolts. Model-selection statistics and symbols used for explanatory variables are defined as in TABLE 5.2.

Model	k	AIC$_c$	ΔAIC$_c$	w_i
Group + Daylight + Handling + NKA + ΔNKA	7	2008.47	0.00	0.15
Group + Daylight + Salinity + Handling + NKA + ΔNKA	8	2008.71	0.25	0.13
Group + Daylight + NKA + ΔNKA	6	2009.10	0.63	0.11
Group + Daylight + Salinity + Handling + NKA + Osmolality + ΔNKA	9	2009.23	0.77	0.10
Group + Daylight + Handling + NKA + Osmolality + ΔNKA	8	2009.33	0.87	0.10
Group + Daylight + NKA + Osmolality + ΔNKA	7	2009.45	0.99	0.09
Group + Daylight + Salinity + NKA + Osmolality + ΔNKA	8	2009.50	1.04	0.09
Group + Daylight + Salinity + NKA + ΔNKA	7	2009.53	1.07	0.09
Group + NKA + Osmolality + ΔNKA	6	2011.57	3.10	0.03
Group + Salinity + NKA + Osmolality + ΔNKA	7	2012.32	3.86	0.02

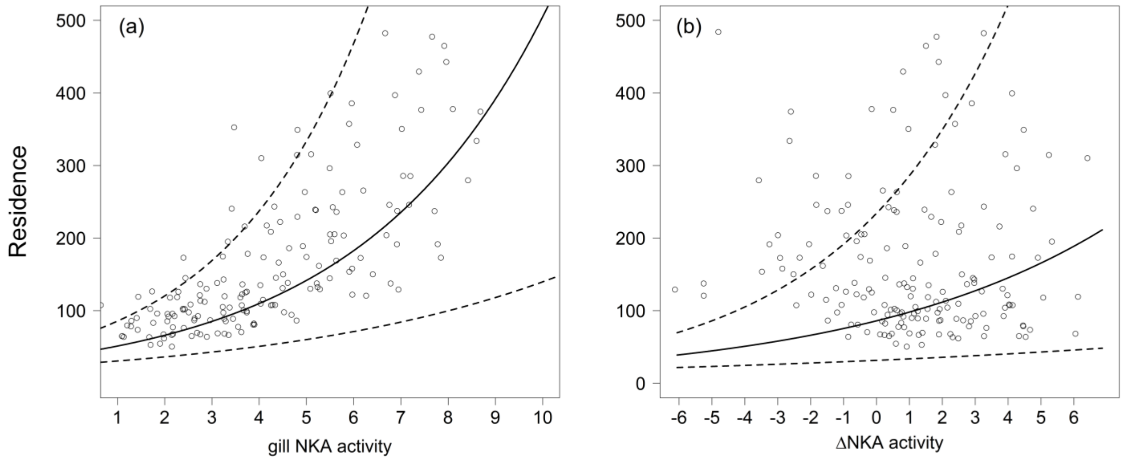


Figure 5.5. Relationship between gill NKA activity and residence time in saltwater. Shown are change in maximum continuous time spent in the HSW compartment of salinity choice tank (‘Residence’) by *Salmo salar* smolts with a) gill Na$^+$, K$^+$-ATPase activity (NKA), and b) changes in gill NKA activity following SW challenge at 35 ppt for 16–24 hours.

Field study

All (100%) of the *S. salar* smolts that were acoustically tagged survived migration from the release site to the ocean during the course of the field study, based on relocation of all individuals at the mouth of the Penobscot Bay. Minimum residency time in the estuary was 3 days (three fish), and maximum residency time in the estuary and bay for any of the fish was 7 days (two fish).

Logistic regression models used to estimate salinity throughout the estuary fit the data well ($\hat{c} \approx 1.00$), and indicated low probability of encountering SW at receiver locations until about rkm 20, at which point salinity increased gradually until full-strength SW was present in the mouth of the estuary at rkm 0 (Figure 5.6a). The slopes of the individual regression lines and the spread of these lines in the middle estuary indicated a gradual increase in mean salinity until full SW was reached at the mouth of the estuary.

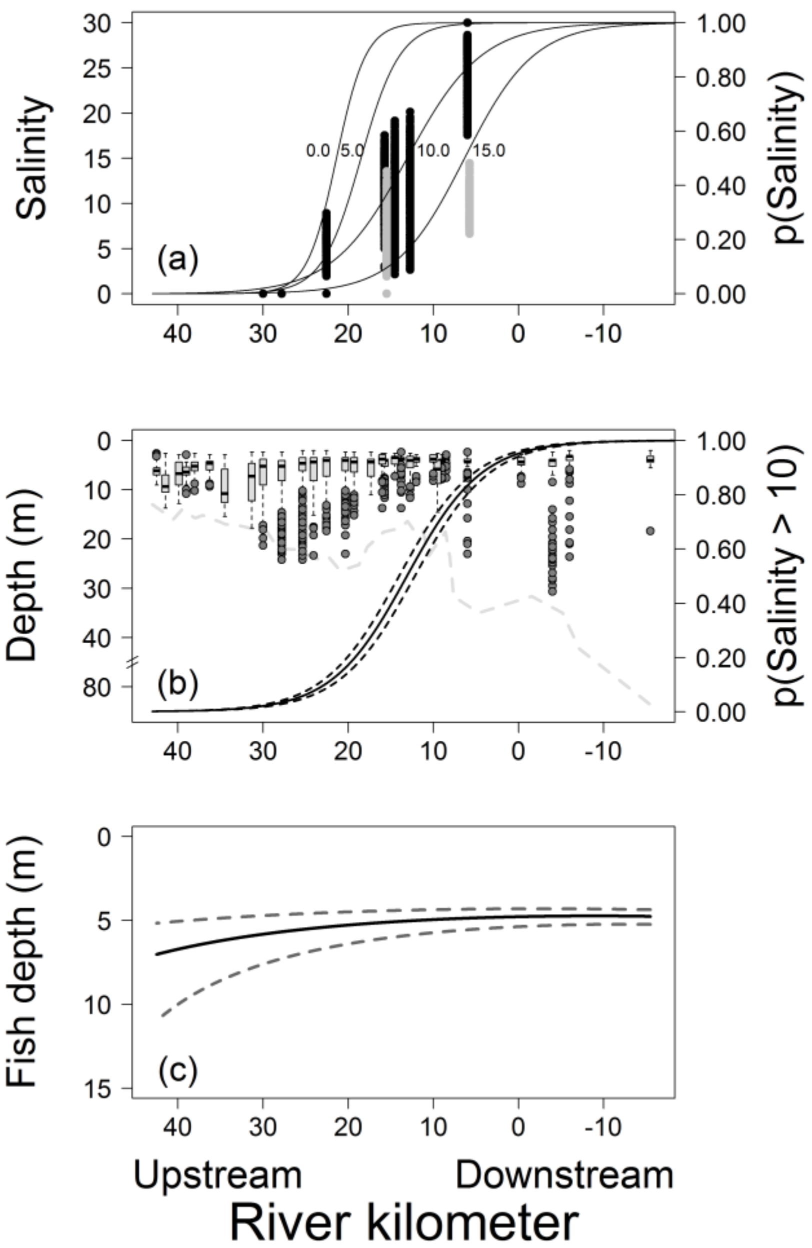


Figure 5.6. Predicted salinity (a) observed fish depth (b) and modeled fish depth (c) in the Penobscot River Estuary. The top panel (a) shows observed salinities at top (gray points) and bottom (black points) of the water column at CTD locations on the primary y-axis, and solid lines are modeled probabilities that mean salinity exceeds 0, 5, 10, or 15 at a given location in the estuary. The middle panel (b) shows box plot of depths used by *S. salar* during migration plotted against probability that mean (± 95% C.I.) salinity (black lines) in the estuary was greater than 10 at acoustic receiver locations, and mean maximum channel depth (gray, dashed line). Boxes (in b) represent 25–75% C.I., lines in the center of the boxes are median depth, and points are outliers. Whiskers indicate 95% C.I.

The overall mean (± S.D.) depth used by *S. salar* smolts during passage through the Penobscot Estuary was 5.9 (± 3.2) m. Depth use by migrating smolts was highly variable until the fish reached uniform SW conditions downstream of rkm 10 in the lower estuary (Figure 5.6b). Smolts became increasingly surface oriented during migration from the head of the estuary to the mouth of the bay. Upon reaching full salt water in the lower estuary near rkm 10, variability in the depths used by individual smolts decreased and mean depth used in SW was shallower than in FW or in the mixing zone of the Penobscot River Estuary, despite that mean maximum channel depth increased (Figure 5.6b). The quadratic relationship between estuary rkm and depth used by smolts indicated a rapid decrease in depth used, as well as in the variability of depth used, when *S. salar* smolts initially encountered the salt wedge in the middle estuary (Figure 5.6c and Table 5.5). Estimated depth use from the regression model appeared to agree well with mean observed depths. Despite that smolts became increasingly surface oriented during downstream migration, fish continued to make some use of the deepest parts of the water column for the full length of the estuary. Several smolts were observed to use water as deep as 30 m in the estuary (Figure 5.6b).

The relationship between depths used and location within the estuary was related to the presence or absence of SW (Figure 5.6a). When salt water was absent in the upper estuary, depths used by smolts was highly variable and no obvious pattern in depth used between locations was apparent. Similarly, fish were located deepest in areas of the estuary with low probability of encountering SW in the water column (Fig 6a) until they reached full SW.

Table 5.5. Estimated regression coefficients and associated standard errors for the GLMM used to estimate changes in depths used by *Salmo salar* smolts during downstream migration through Penobscot Estuary. The symbol *z* is the value of the z statistic, and *p* is the p-value for each coefficient. River kilometer (rkm) was measured from the mouth of the estuary (rkm 0). Values of rkm greater than zero were upstream of the mouth of the estuary and values of rkm less than zero were located downstream of the mouth of the estuary.

Parameter	Estimate	S.E.	*z*	*p*
Intercept	0.20901	0.01174	17.79738	< 0.001
River kilometer	-0.00046	0.00018	-2.51326	< 0.050
River kilometer^2	-0.00003	0.00000	-7.16993	< 0.001

Travel time through FW from the release site (rkm 43.5) to rkm 30 (where salinity > 0 was first detected by CTDs) was inversely related to gill NKA activity of individual smolts (simple linear regression, $R^2 = 0.53$, $F_{1,2} = 9.204$, 1, df = 8, $p < 0.05$). However, total travel time from rkm 30 to the mouth of the estuary was not related to gill NKA activity (simple linear regression, $R^2 = 0.03$, $F_{1,2} = 0.265$, 1, df = 8, $p < 0.60$). Based on examination of individual plots of horizontal and vertical movements, the differences observed in FW were a result of 'station-holding' behavior, whereby fish remained in a given location over multiple tidal cycles (e.g., Figure 5.7). When fish exhibited station-holding behavior, they generally tended to occupy deeper water than when actively migrating, although vertical movements that apparently related to tidal cycles were observed. When holding station in FW, fish tended to rise into the water column late during incoming tides or early during outgoing tides, and if they did not make seaward movement on that tidal cycle, they moved deeper later in the outgoing tide. None of the fish that exited the estuary within 3 days exhibited station-holding behavior in FW reaches of the estuary.

Upon arrival at estuary reaches in which low salinity water (0–5) was present (*c.* rkm 30), all smolts began to display directional reversals in movement (i.e., movement upstream), a behavior that ceased after entry of SW > 10 near rkm 10 in the estuary, regardless of time spent in FW reaches. Mean (± S.D.) number of reversals in these reaches was 2.7 (± 1.3), and the number of migratory reversals ranged from 1 to 5 for the fish used in this study. This behavior correlated with tidal cycle. Upstream movement generally occurred on incoming tides, and downstream movement occurred on outgoing tides in virtually all observations (Figure 5.7).

The tidal patterns in horizontal movements through the estuary were accompanied by corresponding changes in vertical movements. Fish were nearer to the surface while mobile than when holding position. Furthermore, fish became more surface oriented while embarking on movement and they increased depth as they terminated movement. Smolts were generally located deeper on incoming tides than on outgoing tides (t-test, $t = 2.8129$, df =10,783, $p < 0.05$). Once in the lower estuary (downstream or rkm 30), fish always initiated station-holding behavior at the end of an outgoing tide; continuing to hold horizontal position until the end of the subsequent incoming tide (Figure 5.7).

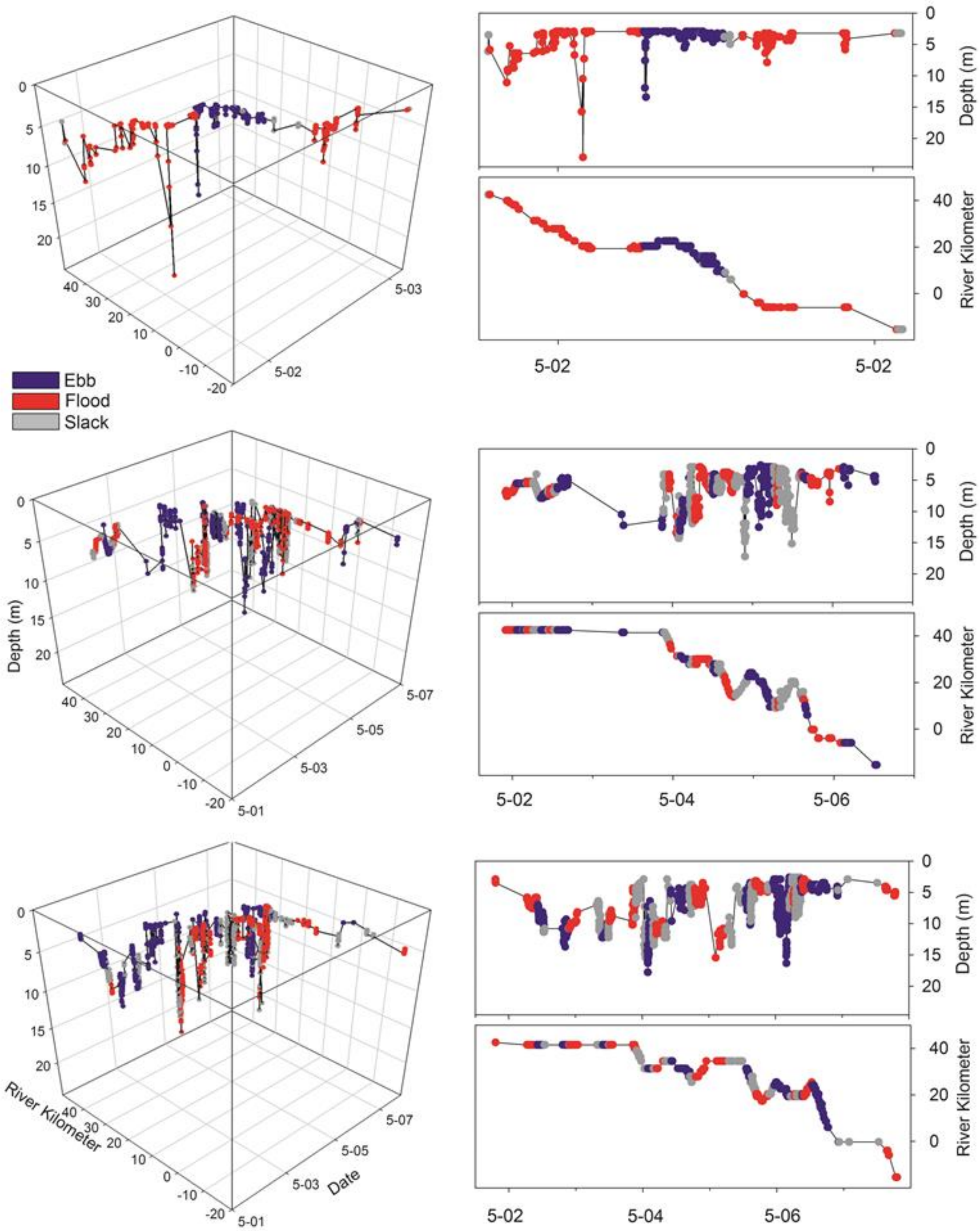


Figure 5.7. Tidal movements of smolts during estuary migration. Shown are representative plots of horizontal and vertical movements of three *S. salar* smolts during estuary migration with respect to tidal cycles for each observation (blue is ebb tide, red is flood tide, and gray is slack tide). The right panel (b) shows three-dimensional fish tracks through space and time with respect to tidal stage. The right panel (b) shows two-dimensional overlay of vertical (top panel in each) and horizontal (bottom panel in each) movements of three fish with respect to tidal stage.

DISCUSSION

Salinity preference, SW residence, and osmoregulation

The results of this study have demonstrated the utility of a novel experimental apparatus for use in monitoring salinity preference of *S. salar* smolts. Salinity choice in the laboratory study (measured as preference and residence) was successfully related to an established measure of physiological preparedness for SW entry (gill NKA activity: McCormick et al. 2012; McCormick 2013) as well as physiological performance following SW exposure (indicated by ΔNKA and demonstrated by plasma osmolality). As expected from the results of previous work (Zaugg and McClain 1970; Duston and Saunders 1995), a developmental shift in gill NKA activity was observed, and *S. salar* tested during May (i.e., smolts) had greater gill NKA activity than presmolts or postsmolts. A corresponding increase was observed in the osmoregulatory performance of *S. salar* during the course of development, as indicated by the fact that smolts had lower plasma osmolality and upregulated gill NKA activity less in response to SW exposure than either presmolts or postsmolts. These results are consistent with the results of a large body of work regarding osmoregulatory capacity during smolt development, summarized by McCormick (2013).

The behavioral responses to SW by the postsmolt group in the salinity choice tank indicate a general correspondence with some potential decoupling of the behaviors late in the ecological smolt window with respect to the timing of the physiological optimum for SW entry. An ontogenetic increase in preference was observed during the course of the laboratory experiment. Residency in the HSW compartment of the salinity choice tank also increased until the peak of the run, but was less in the postsmolt group than the smolt group. Similarly, measurements of SW tolerance increased until the peak of physiological smolting and subsequently were lower in the postsmolt group. These results could have important implications for the seasonal timing of smolt runs and effects of migration delays behind dams. Early- and late-migrating smolts tend to have reduced estuary survival relative to smolts that migrate during the peak of physiological smolt window (Stich et al. in review). It is possible that continued preference for SW late in the run, combined with reduced SW tolerance (measured as plasma osmolality or ΔNKA activity) could result in lower estuary survival of fish migrating late in the physiological smolt window. Delays such as those at dams (Keefer et al. 2012) can reduce survival of smolts in the later part of smolt runs (Marschall et al. 2011), possibly because fish might retain similar preferences for SW despite reduced osmoregulatory capacity.

It is unknown whether this behavior has direct implications for fitness in the wild or if it represents behavioral plasticity that might allow for corresponding flexibility in the period of overlap between ecological and physiological smolt windows. This result may indicate a reason for strong relationships between timing of estuary arrival and smolt survival observed in the wild (see Thorstad et al. 2012*b*), and suggests strong optimizing selection might occur on natural variability in physiological preparedness, resulting in temporally narrow, synchronous migrations that last only weeks (McCormick et al. 1998), such as in the Penobscot River (Stich et al. in review). Alternatively, behavioral preferences of smolts for salinity may be highly plastic compared to physiological underpinnings of salinity tolerance (Hutchings 2011), and as a result promote potential behavioral adaptability to changing environmental conditions driving physiological smoltification from year to year. This might have implications for the adaptability of smolts to changing climate in the future, as greater flexibility in behavioral SW preferences could promote resilience to changes in environmental releasers of migratory behavior (Zydlewski et al. 2014).

Integrating lab and field studies

It is notable that *S. salar* in the laboratory study never exhibited a demonstrable preference for SW (fish in all groups never spent more than 50% of time in HSW), even at the peak of physiological smolting. Smolts at all developmental stages avoided SW, despite the fact that gill NKA activity and SW-challenge performance measurements indicated that fish were fully competent for transition into SW based on previous research (Duston and Saunders 1990). While this has the potential to be an artifact of the tank design used in the laboratory study, it was consistent with what I observed during migration in the Penobscot River Estuary, and similar avoidance of SW previously has been observed in migrating *S. salar* smolts that use the upper, FW layers of the water column for migration (Renkawitz et al. 2012).

Depths used by smolts during estuary migration indicated a pattern consistent with salinity influence in the estuary. Depth use was highly variable in the upper estuary until fish encountered SW. Upon reaching water with elevated salinity, fish moved closer to the surface. These results indicate that migrating fish made use of the entire water column during passage of freshwater reaches, but concentrated in the top of the water column, where salinities were lower, upon reaching SW in the lower estuary. Although fish continued to make excursions into deeper (and higher salinity) water in the lower estuary as previously observed in the Penobscot River Estuary (Renkawitz et al. 2012), the frequency with which fish made vertical movements and the duration of these movements appeared to be greatly reduced in reaches of the lower estuary where mean salinity exceeded concentrations that were isosmotic (salinity ≈ 10). The mean depth during estuary migration was deeper than has previously been described for this species (Plantalech Manel-La et al. 2009; Renkawitz et al. 2012; Thorstad et al. 2012*a*). This is expected because previous studies have examined vertical movements of postsmolts in fjords and coastal bays where availability of FW is restricted to upper layers of the water column, and fish depth was observed to decrease substantially upon reaching full SW in the present study.

The use of the upper water column may occur for a number of reasons other than, or in addition to, salinity preferences, such as decreased energetic cost of migration (Moore et al. 1998), pelagic feeding (Renkawitz and Sheehan 2011), selection of warmer temperatures (Steffansson et al. 2003) or avoidance of deep-water predators (Hvidsten and Lund 1988). However, the agreement between laboratory and field studies, as well as the physical-chemical data collected, suggests that salinity preferences may also influence the depth of water occupied by smolts due to the presence of low-salinity water in the top few meters with a net seaward flow during spring. Periodic excursions to greater depths likely are not related to salinity preference, but rather feeding behavior or surface-predator avoidance, as previously has been suggested (Renkawitz et al. 2012). Based on these results, it seems that smolts in this and other systems make use of the freshwater layers of estuarine and coastal systems during migration while those layers are available, after which the fish adopt a surface-oriented disposition, likely for other reasons.

Movement rate from the release location at the head of tide to the middle estuary (rkm 30, where SW was first present) was related to gill NKA activity of individual *S. salar* smolts. Conversely, transit time from rkm 30 to the mouth of the estuary was not related to gill NKA activity. Previous research has failed to detect differences in estuarine movement rates based on gill NKA activity (Stich et al. in review) in the Penobscot Estuary, but that study did not attempt to separate movement rates based on location of SW in the estuary. It appears that *S. salar* behaviorally prepared for SW entry by prolonging their residency time in the upper estuary prior to entering SW, and not through behavioral acclimation after entry into SW, as previously has been speculated (Halfyard et al. 2012; Halfyard et al. 2013). In fact, gill NKA activity explained more than half ($R^2 = 0.53$) of the variation in travel time from release to the middle estuary (rkm 30), and virtually no support ($p > 0.60$) was found for a relationship between transit time through SW and gill NKA activity. The exact mechanisms and environmental cues controlling this behavioral preparation for SW entry currently are not well understood, but might involve both internal (e.g., circulating hormone levels) or external (e.g., temperature, current velocity, or olfaction) cues. The relationship between traverse time in the upper estuary and gill NKA activity could have important implications for optimizing stocking decisions regarding *S. salar* smolts with respect to seasonal timing, as well as timing of tidal cycles.

Tidal movements in the estuary

Salmo salar at various developmental stages of smolting generally are capable of tolerating exposure to full-strength SW for short periods (McCormick 2013). But, fish early and late in the smolt window might experience greater mortality during estuary migration due to reduced osmoregulatory performance (Duston and Saunders 1990) in conjunction with stressors (Price and Schreck 2003*b*) and other agents of osmotic perturbance during chronic exposure to SW (Handeland et al. 1996; McCormick et al. 2005; Zydlewski et al. 2010). In the lab study, this was indicated by low-level mortalities in the presmolt group, as well as observations of moribund presmolts and postsmolts following 24-hour exposure to SW and differences in plasma osmolality between these fish and those fish that showed no change in physical disposition. Based on the relationships between osmoregulatory preparedness, performance and behavioral choices in the laboratory experiment, it is expected that downstream-migrating *S. salar* might mediate behavior prior to or during estuary migrations to minimize osmoregulatory perturbance. This might occur through behavioral selection of low-salinity water (Price and Schreck 2003*b*), reduction of energetic investment in movement with respect to prevailing currents (McCleave 1978), reduced exposure to predators through selection of specific depths during day or night (Moore et al. 1995), or some combination of these behaviors (Ibbotson et al. 2006).

All migrating smolts exhibited behavioral reversals in movement direction during migration through the Penobscot River Estuary, behavior observed in other systems (Kocik et al. 2009; Dempson et al. 2011; Halfyard et al. 2013). The locations at which fish in the present study reversed movement direction always occurred within a 20-km section of the estuary, between rkm 30 and rkm 10. This section of the estuary appeared also to be the area in which SW was first encountered by smolts. While the fish observed in the present study displayed only minimal numbers of directional reversals (mean = 2.7) compared to other studies (e.g., Halfyard et al. 2012), it seems possible that this behavior might promote osmoregulatory capacity through behavioral acclimation to SW (Halfyard et al. 2012); however, this would in theory occur at the cost of increased exposure to predators and other sources of osmoregulatory perturbance (McCormick et al. 1998). As a result of the trade-off between increased osmoregulatory capacity and increased predation, the fitness benefits and associated adaptive value of this behavior seem dubious if related to SW acclimation. More likely, directional reversal of movements were related to minimizing energetic costs of movement by moving in synchrony with tides and surface currents (Gibson 2003), as suggested by plots of fish movements with respect to tide. Tidal current speeds in excess of sustained swimming capacity have been observed in the lower Penobscot Estuary (McCleave 1978), supporting the hypothesis that observed directional reversals might be related to tides rather than SW acclimation. Thus, these behaviors appear to result from contstraints to movement energetics rather than tradeoffs between osmoregulation and predator avoidance.

Selective tidal stream transport (STST) is a behavioral tactic that reduces energetic costs of migration for marine animals (Forward and Tankersley 2001), and several examples exist in a number of invertebrate (Queiroga 1998; Forward et al. 2003; Criales 2011) and fish species (Barbin 1998; McCleave and Arnold 1999; Kelly and Kimley 2012), including adult sockeye salmon (Levy and Cadenhead, 1995). Changes in depth by *S. salar* smolts in the Penobscot River Estuary suggested that fish were located significantly deeper during incoming tides than outgoing tides, a behavior that would result in increased, rather than decreased exposure to SW during tidal cycles as would be expected from the laboratory results. Although tidal movements have been observed in estuarine and coastal waters by *S. salar* smolts and postsmolts (McCleave 1978; Lacroix et al. 2005), this behavior has not previously been linked to STST due to incomplete information on individual behavior during migration. The vertical pattern observed in this study does, however, typify behaviors involved in STST (Gibson 2003) used in reducing energetic expenditures of swimming against strong currents because current velocities are reduced near boundary layers (i.e., estuary bottom) compared to the upper water column. Because all of the tagged smolts in the present study demonstrated some apparent use of STST during migration through the middle estuary, and because all fish exhibited directional reversal of movements regardless of gill NKA activity, it seems reasonable that the vertical and horizontal movements of *S. salar* smolts through the lower estuary are based on minimizing energetic cost, and not physiological acclimation for SW entry. However, there may be important interactions between osmoregulatory preparedness and osmotic perturbance incurred by increased energetic costs of migrating against currents that might manifest in terms of the duration and number of migratory reversals undertaken by individual smolts (Halfyard et al. 2012), a hypothesis that would require further investigation.

Conclusions

This study corroborates a number of existing hypotheses about *S. salar* smolt behavior. Observations of migratory behavior of smolts during estuary migration showed intriguing evidence for tide-related movement patterns and clear patterns in depth use with respect to salinity gradients through a 45-km estuary. The results of both the laboratory study and the field study suggested that, when given the choice, smolts generally use freshwater in greater relative proportion than salt water. However, the ability of smolts at various developmental stages to enter and remain in SW seemed to be related to an important enzyme used as an indicator of osmoregulatory capacity, as well as the ultimate physiological disposition of fish. The use of information from laboratory studies to guide investigation of interesting trends in estuary behavior helped uncover important spatial and temporal trends in behavior that otherwise might have gone unnoticed. Given the observation of a gradual salt-wedge in the Penobscot Estuary with respect to distance and the advanced mediation of behavior by smolts to prolong SW entrance in the wild, future experiments might investigate mechanisms by which smolts make behavioral decisions about SW entry with respect to physiology and minimal detectable salinities.

Physiological controls of salinity preferences (e.g., Iwata et al. 1990) and saltwater orientation (Otto and McInerney 1970) in salmonids may differ from physiological controls of the actual behavioral preference for SW. For example, whereas behavioral preferences for downstream movement and salinity preference are likely controlled primarily by release of thyroid hormone mediated by photoperiod (Iwata et al. 1990), physiological salinity tolerance is controlled by a suite of hormones and peptides, including thyroid hormones, cortisol, adrenocorticotropic hormone, growth hormone, insulin-like growth factor-1, and possibly others (see McCormick 2013). Furthermore, perturbations to osmotic balance might effect change in physiological processes controlling behavior (Price and Schreck 2003*a* 2003*b*) as behavior also was affected by handling in the present study. This could result in reduced behavioral preferences for SW based on physiological impairment or osmotic perturbation, as was observed in the laboratory experiment in the present study. Further investigation into the physiological underpinnings of salinity preference could have potentially important consequences for the timing and siting of smolt stocking in this system.

REFERENCES

Aarestrup, K., C. Nielsen, and A. Koed. 2002. Net ground speed of downstream migrating radio-tagged Atlantic salmon (*Salmo salar* L.) and brown trout (*Salmo trutta* L.) smolts in relation to environmental factors. Hydrobiologia 483:95–102.

Aarestrup, K., C. Nielsen, and S. S. Madsen. 2000. Relationship between Na^+, K^+-ATPase activity and downstream movement in domesticated and first-generation offspring of wild anadromous brown trout (*Salmo trutta*). Canadian Journal of Fisheries and Aquatic Sciences 57:2086–2095.

Aarestrup, K., H. Baktoft, A. Koed, D. del Villar-Guerra, and E. B. Thorstad. 2014. Comparison of the riverine and early marine migration behaviour and survival of wild and hatchery-reared sea trout *Salmo trutta* smolts. Marine Ecology Progress Series 496:197–206.

Antalos, M., D. D. Roby, D.E. Lyons., K. Collis., A. F. Evans, M. Hawbecker, and B. A. Ryan. 2005. Caspian tern predation on juvenile salmonids in the Mid-Columbia River. Transactions of the American Fisheries Society 134, 466-480.

Barbin, G. P. 1998. The role of olfaction in homing and estuarine migratory behaviour of yellow-phase American eels. Canadian Journal of Fisheries and Aquatic Sciences 55:564–575.

Bates, D., M. Maechler, M., and B. Bolker. 2013. lme4 vignette – Linear mixed-effects models using S4 classes. Available at http://cran.r-project.org/web/packages/lme4/lme4.pdf (last accessed 9 September 2013).

Berli, B. I., M. J. Gilbert, A. L. Ralph, K. B. Tierney, and P. Burkhardt-Holm. 2014. Acute exposure to a common suspended sediment affects the swimming performance and physiology of juvenile salmonids. Comparative Biochemistry and Physiology: Part A, Molecular & Integrative Physiology. doi: 10.1016/j.cbpa.2014.03.013.

Blackwell, B. F., and F. Juanes. 1998. Predation on Atlantic salmon smolts by striped bass after dam passage. North American Journal of Fisheries Management 18:936-939.

Blackwell, B. F., W. B. Krohn, N. R. Dube, and A. J. Godin. 1997. Spring prey use by double-crested cormorants on the Penobscot River, Maine, USA. Colonial Waterbirds 20:77–86.

Boeuf, G. 1993. Salmonid smolting: a pre-adaptation to the oceanic environment. Pages 105–135 in J. C. Renkin, and F. B. Jensen, editors, Fish Ecophysiology. Chapman and Hall, London, United Kingdom.

Branco, P., P. Segurado, J. M. Santos, and M. T. Ferreira. 2014. Prioritizing barrier removal to improve functional connectivity of rivers. Journal of Applied Ecology 51:1197–1206.

Budy, P., G. P. Thiede, N. Bouwes, C. E. Petrosky, and H. Schaller. 2002. Evidence linking delayed mortality of Snake River salmon to their earlier hydrosystem experience. North American Journal of Fisheries Management 22:35–51.

Budy, P., G. P.Thiede, N. Bouwes, C. E. Petrosky, and H. Schaller. 2002. Evidence linking delayed mortality of Snake River salmon to their earlier hydrosystem experience. North American Journal of Fisheries Management 221:35–51.

Burnham, K. P., and D. R. Anderson. 2002. Model selection and multimodel inference: a practical information-theoretic approach, second edition. Springer, New York, New York.

Calver, A. M., S. J., Bonner, I. D.Jonsen, J. M. Flemming, S. J. Walde, and P. D. Taylor2009. A hierarchical Bayesian approach to multi-state mark-recapture: simulations and applications. Journal of Applied Ecology 46:610–620.

Carey, J. B., and S. D. McCormick. 1998. Atlantic salmon smolts are more responsive to an acute handling and confinement stress than parr. Aquaculture 168:237–253.

Chaput, G. 2012. Overview of the status of Atlantic salmon (*Salmo salar*) in the North Atlantic and trends in marine mortality. International Counsel for the Exploration of the Sea Journal of Marine Science 69:1538–1548.

Chaput, G., C. M. Legault, D. G. Reddin, F. Caron, and P .G. Amiro. 2005. Provision of catch advice taking account of non-stationarity in productivity of Atlantic salmon (*Salmo salar* L.) in the Northwest Atlantic. International Counsel for the Exploration of the Sea Journal of Marine Science 62:131–143.

Choquet, R., J. Lebreton, O. Gimenez, A. Reboulet, and R. Pradel. 2009. U-CARE: utilities for performing goodness of fit tests and manipulating Capture-Recapture data. Ecography 32:1071-1074.

Connor, W. P., H. L. Burge , and J. R. Yearsley. 2003. Influence of flow and temperature on survival of wild subyearling fall Chinook salmon in the Snake River. North American Journal of Fisheries Management 23:362-375.

Criales, M. M., M. B. Robblee, J. A. Browder, H. Cardenas T. L. Jackson. 2011. Field observations on selective tidal-stream transport for postlarval and juvenile pink shrimp in Florida Bay. Journal of Crustacean Biology 31:26–33.

Davidsen, J. G., N. Plantalech Manel-la, O. H. Økland, E. B. Thorstad., B. Finstad, R. Sivertsgård., R. S. McKinley, and A. H. Rikardsen. 2008. Changes in swimming depths of Atlantic salmon *Salmo salar* post-smolts relative to light intensity. Journal of Fish Biology 73: 1065–1074.

Day, L. R. 2006. Restoring native fisheries to Maine's largest watershed: the Penobscot River Restoration Project. Journal of Contemporary Water Research and Education 134:29-33.

Dempson, J. B., C. J. Schwarz, D. G. Reddin, M. F. O'Connell, C. C. Mullins, and C. E. Bourgeois. 2001. Estimation of marine exploitation rates on Atlantic salmon (*Salmo salar* L.) stocks in Newfoundland, Canada. International Council for the Exploration of the Sea Journal of Marine Science 58:331–341.

Dempson, J. B., M. J. Robertson, C. J. Pennell, G. Furey, M. Bloom, M. Shears, L. M. N. Ollerhead, K. D. Clark, R. Hinks, and G. J. Robertson. 2011. Residency time, migration route, and survival of Atlantic salmon *Salmo salar* smolts in a Canadian fjord. Journal of Fish Biology 78:1976–1992.

Dingle, H., and V. A. Drake. 2007. What is migration? BioScience 57:113¬–121.

Duston, J., and R. L. Saunders. 1995. Increased winter temperature did not affect completion of smolting in Atlantic salmon. Aquaculture International 3:196–204.

Duston, J., and R. L. Saunders. 1990. The entrainment role of photoperiod on hypoosmoregulatory and growth-related aspects of smolting in Atlantic salmon (*Salmo salar*). Canadian Journal of Zoology 68:707–715.

Faraway, J. J. 2005. Extending the Linear Model with R: Generalized Linear, Mixed Effects and Nonparametric Regression Models. Chapman and Hall, London.

FERC (Federal Energy Regulatory Commission). 2004. Submittal of the Lower Penobscot River Basin comprehensive settlement accord with explanatory statement for FERC project numbers 2403, 2534, 2666, 2710, 2712, 2721, and 10981. United States Department of Energy, Washington D. C. Available from http://elibrary.ferc.gov/idmws/common/OpenNat.asp?fileID=10188480 [accessed 13 August 2014].

FERC (Federal Energy Regulatory Commission). 2005. Order modifying and approving amendment of license for FERC project number 2712-055. United States Department of Energy, Washington D. C. Available from http://elibrary.ferc.gov/idmws/common/opennat.asp?fileID=10499146 [accessed 13 August 2014].

FERC (Federal Energy Regulatory Commission). 2009. Draft environmental assessment, application for surrender of license for FERC project numbers 2403-056, 2312-019 and 2721-20. United States Department of Energy, Washington D. C. Available from http://www.penobscotriver.org/assets/FERC_Environmental_Assessment_-_Draft_8-4-09.pdf [accessed 13 August 2014].

Ferguson, J. W., B. P. Sanford, R. E. Reagan, L. G. Gilbreath, E. B. Meye, R. D. Ledgerwood, and N. S. Adams. 2007. Bypass system modification at Bonneville Dam on the Columbia River improved the survival of juvenile salmon. Transactions of the American Fisheries Society 136:1487-1510.

Forward Jr., R. B., R. A. Tankersley, and J. M. Welch. 2003. Selective tidal-stream transport of the blue crab *Callinectes sapidus,* an overview. Bulletin of Marine Science 72:347–365.

Forward Jr., R. B., and R. A. Tankersley. 2001. Selective tidal-stream transport of marine animals. Oceanography and Marine Biology 39:305–353.

Fried, S. M., J. D. McCleave, and G. W. LaBar. 1978. Seaward migration of hatchery-reared Atlantic salmon, *Salmo salar*, smolts in the Penosbcot Estuary, Maine: riverine movements. Journal of the Fisheries Research Board of Canada 35:76–87.

Friedland, K. D. 1998. Ocean climate influences on critical Atlantic salmon (*Salmo salar*) life history events. Canadian Journal of Fisheries and Aquatic Sciences 55 (Supplement 1):119–130.

Friedland, K. D., B. V. Shank, C. D. Todd, P. McGinninty, and J. A. Nye. 2014. Differential response of continental stock complexes of Atlantic salmon (*Salmo salar*) to the Atlantic Multidecadal Oscillation. Journal of Marine Systems 133:77–87.

Friedland, K. D., G. Chaput , and J. C. MacLean. 2005. The emerging role of climate in post-smolt growth of Atlantic salmon. International Council for the Exploration of the Sea Journal of Marine Science 62, 1338–1349.

Friedland, K. D., D. G. Reddin, J. R. McMenemy, and K. F. Drinkwater. 2003. Multidecadal trends in North American Atlantic salmon (*Salmo salar*) and climate trends relevant to juvenile survival. Canadian Journal of Fisheries and Aquatic Sciences 60:563–583.

Friedland, K. D., L. P. Hansen, D. A. Dunkley, and J. C. MacLean. 2000. Linkage between ocean climate, post-smolt growth, and survival of Atlantic salmon (*Salmo salar* L.) in the North Sea area. International Council for the Exploration of the Sea Journal of Marine Science 57:419–429.

Friedland, K. D., D. G. Reddin, and M. Castonguay. 2003. Ocean thermal conditions in the post-smolt nursery of North American Atlantic salmon. International Council for the Exploration of the Sea Journal of Marine Science 60:343–355.

Gibson, R. N. 2003. Go with the flow: tidal migration in marine animals. Hydrobiologia 503:153-161.

Gorsky, D., J. Trial, J. Zydlewski, and J. McCleave. 2009. The effects of smolt stocking strategies on migratory path selection of adult Atlantic salmon in the Penobscot River, Maine. North American Journal of Fisheries Mangament 29:949–957.

Gudjonsson, S., I. R. Jonsson, and T. Antonsson. 2005. Migration of Atlantic salmon, *Salmo salar*, smolt through the estuary area of River Ellidaar. Environmental Biology of Fishes 74:291–296.

Gunderson L., and S. L. Light. 2006. Adaptive management and adaptive governance in the everglades ecosystem. Policy Science 39:323-334.

Gunnerød, T., N. A. Hvidsten, and T. G. Heggberget. 1988. Open sea releases of Atlantic salmon smolts, *Salmo salar*, in Central Norway, 1971–83. Canadian Journal of Fisheries and Aquatic Sciences 45:1340–1345.

Haefner Jr., P. A. 1967. Hydrography of the Penobscot River (Maine) Estuary. Journal of the Fisheries Research Board of Canada 24:1553–1571.

Haines, T. A. 1992. New England’s rivers and Atlantic salmon. Pages 131-139 in Stroud, R. H., editor, Stemming the tide of coastal fish habitat loss. National Coalition for Marine Conservation, Savannah, Georgia.

Halfyard, E. A., A. J. F. Gibson, M. J. W. Stokesbury, D. E. Ruzzante, and F. G. Whoriskey. 2013. Correlates of estuarine survival of Atlantic salmon postsmolts from the Southern Upland, Nova Scotia, Canada. Canadian Journal of Fisheries and Aquatic Sciences 70:452–460.

Halfyard, E. A., A. J. F. Gibson, D. E. Ruzzante, M. J. W. Stokesbury, and F. G. Whoriskey. 2012. Estuarine survival and migratory behaviour of Atlantic salmon *Salmo salar* smolts. Journal of Fish Biology 81:1626–1645.

Hall, C. J., A. Jordan, and M. G. Frisk. 2010. The historic influence of dams on diadromous fish habitat with a focus on river herring and hydrologic longitudinal connectivity. Landscape Ecology 26:95-107.

Handeland, S. O., T. Järvi, A. Fernö, and S. O. Stefansson. 1997. Osmotic stress, antipredator behavior, and mortality of Atlantic salmon (*Salmo salar*) smolts. Canadian Journal of Fisheries and Aquatic Sciences 53:2673–2680.

Handeland, S. O., E. Wilinson, B. Sveinsbø, S. D. McCormick, and S. O. Stefansson. 2004. Temperature influence on the development and loss of seawater tolerance in two fast-growing strains of Atlantic salmon. Aquaculture 233:513–529.

Hansen, L. P, P. Hutchinson, D. G. Reddin, and M. L. Windsor. 2012. salmon at sea: scientific advances and their implications for management: an introduction. International Council for the Exploration of the Sea Journal of Marine Science 69:1533–1537.

Haugland, M., Holst, J. N., Holm, M., and L. P. Hansen. 2006. Feeding of Atlantic salmon (*Salmo salar* L.) post-smolts in the Northeast Atlantic. International Council for the Exploration of the Sea Journal of Marine Science 63:1488–1500.

Hawkes, J. P., R. Saunders, A. D. Vashon, and M. S. Cooperman. 2013. Assessing efficacy of non-lethal harassment of double-crested cormorants to improve Atlantic salmon smolt survival. Northeastern Naturalist 20:1–18.

Hayes, S. A., and J. F. Kocik. 2014. Comparative estuarine and marine migration ecology of Atlantic salmon and steelhead: blue highways and open plains. Reviews in Fish Biology and Fisheries 24:757–780.

Hedger, R. D., F. Martin, D. Hatin, F. Caron, F. G. Whoriskey, and J. J. Dodson. 2008. Active migration of wild Atlantic salmon *Salmo salar* smolt through a coastal embayment. Marine Ecological Press Series 355:235–246.

Herbert, M. E., and F. P. Gelwick. 2003. Spatial variation of headwater fish assemblages explained by hydrologic variability and upstream effects of impoundment. Copeia 2:273-284.

Hoar, W. S. 1988. The physiology of smolting salmonids. Pages 275-343 in W. S. Hoar and D. J. Randall editors. Fish physiology, Volume XIB. Academic Press, New York, New York.

Holbrook, C. M., M. T. Kinnison, and J. Zydlewski. 2011. Survival of migrating Atlantic salmon smolts through the Penobscot River, Maine: a prerestoration assessment. Transactions of the American Fisheries Society 140:1255–1268.

Holbrook, C. M., J. Zydlewski, D. Gorsky, S. L. Shepard, and M. T. Kinnison. 2009. Movements of prespawn adult Atlantic salmon near hydroelectric dams in the lower Penobscot River, Maine. North American Journal of Fisheries Management 29:495-505.

Horton, G. E., B. H. Letcher, M. M. Bailey, and M. T. Kinnison. 2009. Atlantic salmon (*Salmo salar*) smolt production: the relative importance of survival and body growth. Canadian Journal of Fisheries and Aquatic Sciences 66:471–483.

Hutchings, J. A. 2011. Old Wine in new bottles: reaction norms in salmonid fishes. Heredity 10:421–437.

Hvidsten, N. A., and R. A. Lund. 1988. Predation on hatchery-reared and wild smolts of Atlantic salmon, *Salmo salar* L., in the estuary of River Orkla, Norway. Journal of Fish Biology 33:121–126.

Ibbotson, A. T., W. R. C. Beaumont, A. Pinder, S. Welton,and M. Ladle. 2006. Diel migration patterns of Atlantic salmon smolts with particular reference to the absence of crepuscular migration. Ecology of Freshwater Fish 15:544-551.

Imhoff, E. A., and R. L. Harvey. 1972. Penobscot River Study. Technical Report Number 1, Environmental Studies Center, University of Maine, Orono, ME.

Itokazu, Y., R. Käkelä, J. Piironen, X. L. Guan, P. Kiiskinen, and M. Vornanen. 2014. Gill tissue lipids of salmon (*Salmo salar* L.) presmolts and smolts from anadromous and landlocked populations. Comparative Biochemistry and Physiology Part A: Molecular and Integrative Physiology 172:39–45.

IUBMB (International Union of Biochemistry and Molecular Biology). 1992. Enzyme nomenclature 1992. Academic Press, San Diego, California.

Iwata, M. 1995. Downstream migratory behaviour of salmonids and its relationship with cortisol and thyroid hormones: a review. Aquaculture 135:131–139.

Iwata, M., K. Yamauchi, R. S. Nishioka, R. Lin, and H. A. Bern. 1990. Effects of thyroxine, growth hormone and cortisol on salinity preference of juvenile coho salmon (*Oncorhynchus kisutch*). Marine and Freshwater Behaviour and Physiology 17:191–201.

Järvi, T. 1989. Synergistic effect on mortality in Atlantic salmon, *Salmo salar,* smolt caused by osmotic stress and presence of predators. Environmental Biology of Fishes 26:149–152.

Järvi, T. 1990. Cumulative acute physiological stress in Atlantic salmon smolts: the effect of osmotic imbalance and the presence of predators. Aquaculture 89:337–350.

Johnsen, B. O., J. V. Arnekleiv, L. Asplin, B. T. Barlaup, T. F. Næsje, B. O. Rosseland, S. J. Saltveit, and A. Tvede. 2011. Hydropower development- ecological effects. Pages 351–386 in Aas, Ø. , S. Einum, A. Klemetsen, and J. Skurdal, editors, Atlantic salmon ecology. Blackwell Publishing Ltd, Oxford, United Kingdom.

Jokikokko, E., I. Kallio-Nyberg, I. Saloniemi, and E. Jutila. 2006. The survival of semi-wild, wild, and hatchery-reared Atlantic salmon smolts of the Simojoki River in the Baltic Sea. Journal of Fish Biology 68:430–442.

Jokikokko, E., and S. Mäntyniemi. 2003. The survival of stocked Atlantic salmon smolts during sea run and the timing of migration in the river Simojoki, northern Finland. Aquaculture 219:431–444.

Jonsson, N., B. Jonsson, and L. P. Hansen, L. P. 2003. The marine survival and growth of wild and hatchery-reared Atlantic salmon. Journal of Applied Ecology 40:900–911.

Keefer, M. L., G. A., Taylor, D. F. Garletts, C. K. Helms, G. A. Gauthier, T. M. Pierce, and C. C. Caudill. 2012. Reservoir entrapment and dam passage mortality of juvenile Chinook salmon in the Middle Fork Willamette River. Ecology of Freshwater Fish 21:222–234.

Kelly, J. T., and A. P. Kimley. 2012. Relating the swimming movements of green sturgeon to the movement of water currents. Environmental Biology of Fishes 93:151–167.

Kjelson, M. A., and P. L. Brandes. 1989) The use of smolt survival estimates to quantify the effects of habitat changes on salmonid stocks in the Sacramento-Joaquin Rivers, California. Pages 100-115 In Levings, C. D., L. B. Holtby, and M. A. Henderson, editors, Proceedings of the national workshop on effects of habitat alteration on salmonid stocks, Canadian special publication on fisheries and aquatic sciences 105. Fisheries and Oceans Canada, St. Andrews, New Brunswick.

Kocik, J. F., J. P. Hawkes, T. F. Sheehan, P. A. Music, and K. F. Beland. 2009. Assessing estuarine and coastal migration and survival of wild Atlantic salmon smolts from the Naraguagus River, Maine using acoustic telemetry. Pages 293–310 in A. Haro, K. L. Smith, R. A. Rulifson, C. M. Moffitt, R. J. Klauda, M. J. Dadswell, R. A. Cunjak, J. E. Cooper, K. L. Beal, and T. S. Avery, editors, Challenges for diadromous fishes in a dynamic global environment. American Fisheries Society, Symposium 69, Bethesda, Maryland.

Kroglund, F., B. Finstad, S. O. Stefanson, T. O. Nilson, T. Kristensen, B. O. Rosseland, H. C. Teien, and B. Salbu. 2007. Exposure to moderate acid water and aluminum reduces Atlantic salmon post-smolt survival. Aquaculture 273:360–373.

Krucshke, J. K. 2011. Doing Bayesian data analysis: a tutorial with R and BUGS. Academic press, San Diego, California, USA.

Laake, J. L. 2013. RMark: an R interface for analysis of capture-recapture data with MARK. NOAA National Marine Fisheries Service, Alaska Fisheries Science Center Report 2013-01, Seattle, Washington.

LaBar, G. W., J. D. McCleave, and S. M. Fried. 1978. Seaward migration of hatchery-reared Atlantic salmon (*Salmo salar*) smolts in the Penobscot River estuary, Maine: open-water movements. Journal du Conseil International pour l’Exploration de la Mer 38:257–269.

Lacroix, G. L. 2008. Influence of origin on migration and survival of Atlantic salmon (*Salmo salar*) in the Bay of Fundy, Canada. Canadian Journal of Fisheries and Aquatic Sciences 65:2063–2079.

Lacroix, G. L. 2014. Large pelagic predators could jeopardize the recovery of endangered Atlantic salmon. Canadian Journal of Fisheries and Aquatic Sciences 71:343–350.

Lacroix, G. L., D. Knox, and M. J. W. Stokesbury. 2005. Survival and behaviour of post-smolt Atlantic salmon in coastal habitat with extreme tides. Journal of Fish Biology 66:485–498.

Larsson, S., T. Linnansaari, S. Vatanen, I. Serrano,and A. Haikonen. 2011. Feeding of wild and hatchery reared Atlantic salmon (*Salmo salar* L.) smolts during downstream migration. Environmental Biology of Fishes 92:631–369.

Levy, D. A., and A. D. Cadenhead. 1995. Selective tidal stream transport of adult sockeye salmon (*Onchorhynchus nerka*) in the Fraser River Estuary. Canadian Journal of Fisheries and Aquatic Sciences 52:1–12.

Lunn, D. J., A. Thomas, N. Best, and D. Spiegelhalter. 2000. WinBUGS— a Bayesian modelling framework: concepts, structure, and extensibility. Statistics and Computing 10:325–337.

Marschall, E. A., M. E. Mather, D. L. Parrish, G. W. Allison, and J. R. McMenemy. 2011. Migration delays caused by anthropogenic barriers: modeling dams, temperature, and success of migrating salmon smolts. Ecological Applications 21:3014–3031.

Martin, F. R. D. Hedger, J. J. Dodson, L. Fernandes, D. Hatin, F. Caron, and F. G. Whoriskey. 2009. Behavioural transition during the estuarine migration of wild Atlantic salmon (*Salmo salar* L.) smolt. Ecology of Freshwater Fish 18:406–417.

Martin, P., J. Rancon, G. Segura,J. Laffont,G. Boeuf, and S. Dufour. 2012. Experimental study of the influence of photoperiod and temperature on the swimming behaviour of hatchery-reared Atlantic salmon (*Salmo salar* L.) smolts. Aquaculture 362–363:200–208.

Mathur, D., P. G. Heisey, J. R. Skalski, and D. R. Kenney. 2000. Salmonid smolt survival relative to turbine efficiency and entrainment depth in hydroelectric power generation. Journal of the American Water Resources Association 36:737–747.

McCleave, J. D., and G. P. Arnold. 1999. Movements of yellow- and silver-phase European eels (*Anguilla anguilla* L.) tracked in western North Sea. International Council for the Exploration of the Sea Journal of Marine Science 56:510–536.

McCleave, J. D. 1978. Rhythmic aspects of estuarine migration of hatchery-reared Atlantic salmon (*Salmo salar*) smolts. Journal of fish Biology 12:559–570.

McCormick, S. D. 1993. Methods for non-lethal gill biopsy and measurement of Na^+,K^+-ATPase activity. Canadian Journal of Fisheries and Aquatic Sciences 50:656–658.

McCormick, S. D. 2013. Smolt physiology and endocrinology. Pages 200–237 in S. D. McCormick, A. P. Farrell, and C. J. Brauner, editors, Euryhaline Fishes. Academic Press, Oxford, United Kingdom.

McCormick, S. D., and R. L. Saunders1987. Preparatory physiological adaptations for marine life in salmonids: osmoregulation, growth and metabolism. Pages 211–229 in M. J. Dadswell, R. J. Klauda, C. M. Moffitt, and R. L. Saunders, editors, Common strategies in anadromous and catadromous fishes. American Fisheries, Society Symposium 1, Bethesda, Maryland.

McCormick, S. D., B. T. Björnsson, M. Sheridan, C. Eilertson, J. B. Carey, and M. O'Dea. 1995. Increased daylength stimulates plasma growth hormone and gill Na^+,K^+-ATPase in Atlantic salmon (*Salmo salar*). Journal of Comparative Physiology 165, 245–254.

McCormick, S. D., D. T. Lerner, M. Y. Monette, K. Nieves-Puigdoller, J. T. Kelly, and B. T. Björnsson. 2009. Taking it with you when you go: how perturbations to the freshwater environment, including temperature, dams, and contaminants, affect marine survival of salmon. Pages 195–214 in A. Haro, K. L. Smith, R. A. Rulifson, C. M. Moffitt, R. J. Klauda, M. J. Dadswell, R. A. Cunjak, J. E. Cooper, K. L. Beal, and T. S. Avery, editors, Challenges for diadromous fishes in a dynamic global environment. American Fisheries Society, Symposium 69, Bethesda, Maryland.

McCormick, S. D., J. M. Shrimpton, and J. D. Zydlewski. 1997. Temperature effects on osmoregulatory physiology of anadromous fish. Pages 279–301 in C. M. Wood and D. G. McDonald, editors. Global warming implications for freshwater and marine fish. University Press, Cambridge, United Kingdom.

McCormick, S. D., L. P. Hansen, T. P. Quinn, and R. L. Saunders. 1998. Movement, migration, and smolting of Atlantic salmon (*Salmo salar*). Canadian Journal of Fisheries and Aquatic Sciences 55(Supplement 1):77–92.

McCormick, S. D., M. F. O’Dea, A. M. Moeckel, and B. T. Bjornsson. 2003. Endocrine and physiological changes in Atlantic salmon smolts following hatchery release. Aquaculture 222:45–57.

McCormick, S. D., M. F. O’dea, A. M. Moeckel, D. T. Lerner, and B. T. Björnsson. 2005. Endocrine disruption of parr-smolt transformation and seawater tolerance of Atlantic salmon by 4-nonylphenol and 17beta-estradiol. General and Comparative Endocrinology 142:280–288.

McCormick, S. D., R. A. Cunjak, B. Dempson, M. F. O’Dea, and J. B. Carey. 1999. Temperature-related loss of smolt characteristics in Atlantic salmon (*Salmo salar*) in the wild. Canadian Journal of Fisheries and Aquatic Sciences 56:1649–1658.

McCormick, S. D., R. L. Saunders, and A. MacIntyre. 1989. Mitochondrial enzyme and Na^+,K^+-ATPase activity, and ion regulation during parr-smolt transformation of Atlantic salmon (*Salmo salar*). Fish Physiology and Biochemistry 6:231–241.

McCormick, S. D., R. L. Saunders, E. B. Henderson, and P. R. Harmon. 1987. Photoperiod control of parr-smolt transformation in Atlantic salmon (*Salmo salar*): changes in salinity tolerance, gill Na^+, K^+-ATPase activity and thyroid hormones. Canadian Journal of Fisheries and Aquatic Sciences 44:1462–1468.

McCormick, S. D., A. M. Regish, and A. K. Christensen. 2009. Distinct freshwater and seawater isoforms of Na^+/K^+-ATPase in gill chloride cells of Atlantic salmon. The Journal of Experimental Biology 212:3994–4001.

McCormick, S. D., A. M. Regish, A. K. Christensen, and B. T. Bjornsson. 2012. Differential regulation of sodium–potassium pump isoforms during smolt development and seawater exposure of Atlantic salmon. Journal of Experimental Biology 216:1142–1151.

McCormick, S. D., T. F. Sheehan, B. T. Björnsson, C. Lipsky, J. F. Kocik, A. M. Regish, and M. F. O'Dea. 2013. Physiological and endocrine changes in Atlantic salmon smolts during hatchery rearing, downstream migration, and ocean entry. Canadian Journal of Fisheries and Aquatic Sciences 70:105–118.

McCormick, S.D., R. A. Cunjak., B. Dempson, M. F. O'Dea, and J. B. Carey. 1999. Temperature-related loss of smolt characteristics in Atlantic salmon (*Salmo salar*) in the wild. Canadian Journal of Fisheries and Aquatic Sciences 56:1649–1658.

McCormick, S.D., A. Haro, D. T. Lerner, M. F. O'Dea, and A.M Regish. 2014. Migratory patterns of hatchery and stream-reared Atlantic salmon *Salmo salar* smolts in the Connecticut River, U.S.A. Journal of Fish Biology 85:1005–1022.

McCormick, S.D., J. M. Shrimpton, S. Moriyama, and B. T. Björnsson. 2002. Effects of an advanced temperature cycle on smolt development and endocrinology indicate that temperature is not a zeitgeber for smolting in Atlantic salmon. Journal of Experimental Biology 205:3533–3560.

Millar, C. I., N. L. Stephenson, S. L. Stephens. 2007. Climate change and forests of the future: managing in the face of uncertainty. Ecological Applications 17:2145-2151.

Miller, A. S., T. F. Sheehan, M. D. Renkawitz, A. L. Meister, and T. J. Miller. 2012. Revisiting the marine migration of US Atlantic salmon using historical Carlin tag data. International Counsel for Exploration of the Sea Journal of Marine Science 69:1609–1615.

Mills, K. E., A. J. Pershing, T. F. Sheehan, and D. F. Mountain. 2013. Climate and ecosystem linkages explain widespread declines in North American Atlantic salmon populations. Global Change Biology 19:3046–3061.

Monette, M. Y., and S. D. McCormick. 2008. Impacts of short-term acid and aluminum exposure on Atlantic salmon (*Salmo salar*) physiology: A direct comparison of parr and smolts. Aquatic Toxicology 86:216–226.

Montgomery, D. C., E. A. Peck, and G. G. Vining. 2006. Introduction to linear regression analysis, 4th edition. John Wiley and Sons Inc., Hoboken, New Jersey.

Moore, A., S. Ives, T. A. Mead, and L. Talks. 1998. The migratory behaviour of wild Atlantic salmon (*Salmo salar* L.) smolts in the River Test and Southhampton Water, southern England. Hydrobiologia 371/372:295–304.

Moore, A., E. C. E. Potter, N. J. Milner, and S. Bamber. 1995. The migratory behavior of wild Atlantic salmon (*Salmo salar*) smolts in the estuary of the River Conway, North Wales. Canadian Journal of Fisheries and Aquatic Sciences 52:1923–1935.

Moring, J. R. 2000. The creation of the first public salmon hatchery in the United States. Fisheries 25:6–12.

Muir, W. D., A. E. Giorgi, and T. C. Coley. 1994. Behavioural and physiological changes in yearling Chinook salmon during hatchery residence and downstream migration. Aquaculture 127:69–82.

Music, P. A., J. P. Hawkes, and M. S. Cooperman. 2011. Magnitude and causes of smolt mortality in rotary screw traps: an Atlantic salmon case study. North American Journal of Fisheries Management 30:713–722.

National Marine Fisheries Service & U.S. Fish and Wildlife Service. 2005. Recovery plan for the Gulf of Maine distinct population segment of Atlantic salmon (*Salmo salar*). National Marine Fisheries Service, Silver Spring, MD. Available at http://www.nmfs.noaa.gov/pr/pdfs/recovery/salmon_atlantic.pdf (last accessed 13 October 2014).

National Marine Fisheries Service. 2012a. Endangered species act biological opinion for Federal Energy Regulatory Commission projects 2403, and 2721. National Marine Fisheries Service, Silver Spring, MD. Available at http://www.greateratlantic.fisheries.noaa.gov/protected/section7/bo/actbiops/prrp_opinion_2012.pdf (last accessed 13 August 2014).

National Marine Fisheries Service. 2012b. Endangered species act biological opinion for Federal Energy Regulatory Commission projects 2710, 2712, 2354, 2600, and 2666. United States Department of Commerce, Washington D. C., USA.Available at http://www.nero.noaa.gov/prot_res/section7/Federal Energy Regulatory Commission-signedBOs/Black-BearHydroBO.pdf (last accessed 13 August 2014).

Newman, K. B. & J. Rice. 2002. Modeling the survival of Chinook salmon smolts outmigrating through the lower Sacramento River. Journal of the American Statistical Association 97:983–993.

Newman, K. B. 2003. Modelling paired release-recovery data in the presence of survival and capture heterogeneity with application to marked juvenile salmon. Statistical Modelling 3:157–177.

Norrgård, J. R., L. A. Greenberg, J. J. Piccolo, M. Schmitz, and E. Bergman. 2013. Multiplicative loss of landlocked Atlantic salmon *Salmo salar* L. smolts during downstream migration through multiple dams. River Research Applications 29:1306–1317.

NRC (National Research Council). 2004. Atlantic salmon in Maine. Academic Press, Washington, D.C.

Null, S. E., J. Medellín-Azuara, A. Escriva-Bou, A., M. Lent, and J. R. Lund. 2014. Optimizing the dammed: water supply losses and fish habitat gains from dam removal in California. Journal of Environmental Management 136:121–131.

Opperman, J. J., J. Royte, J. Banks, L. Rose Day, and C. Apse. 2011. The Penobscot River, Maine, USA: a basin-scale approach to balancing power generation and ecosystem restoration. Ecology and Society 16:7–25.

Otero, J., J. H. L'Abée-Lund, T. Castro-Santos, K. Leonardsson, G. O. Storvik, B. Jonsson, B. Dempson, I. C. Russell, A. J. Jensen, J.-L. Baglinière, M. Dionne, J. D. Armstrong, A. Romakkaniemi, B. H. Letcher, J. F. Kocik, J. Erkinaro, R. Poole, G. Rogan, H. Lundqvist, J. C. MacLean, E. Jokikokko, J. V. Arnekleiv, R. J. Kennedy, E. Niemelä, P. Caballero, P. A. Music, T. Antonsson, S. Gudjonsson, A. E. Veselov, A. Lamberg, S. Groom, B. H. Taylor, M. Taberner, M. Dillane, F. Arnason, G. Horton, N. A. Hvidsten, I. R. Jonsson, N. Jonsson, S. McKelvey, T. F. Naesje, Ø. Skaala, G. W. Smith, H. Saegrov, N. C. Stenseth, and L. A. Vøllestad. 2014. Basin-scale phenology and effects of climate variability on global timing of initial seaward migration of Atlantic salmon (*Salmo salar*). Global Change Biology 20:61–75.

Otto, R. G., and J. E. McInerney. 1970. Development of salinity preference in pres-smolt coho salmon, *Oncorhynchus kisutch*. Journal of the Fisheries Research Board of Canada 27:793–800.

Parrish, D. L., R. J. Behnke, S. R. Gephard, S. D. McCormick, and G. H. Reeves. 1998. Why aren’t there more Atlantic salmon (*Salmo salar*)? Canadian Journal of Fisheries and Aquatic Sciences 55(Supplement 1):281–287.

Petrosky, C. E., and H. A. Schaller. 2010. Influence of river conditions during seaward migration and ocean conditions on survival rates of Snake River Chinook salmon and steelhead. Ecology of Freshwater Fish 19:520–536.

Plantalech Manel-La, N., E. B. Thorstad, J. G. Davidsen, F. Økland, R. Sivertsgård, R. S. McKinley, and B. Finstad. 2009. Vertical movements of Atlantic salmon post-smolts relative to measures of salinity and water temperature during the first phase of the marine migration. Fisheries Management and Ecology 16:147–154.

Poe, T. P., H. C. Hansel, S. Vigg, D. E. Palmer, and L. A. Prendergast. 1991. Feeding of predaceous fishes on out-migrating juvenile salmonids in John Day reservoir, Columbia River. Transactions of the American Fisheries Society 120:405–420.

Price, C. S., and C. B. Schreck. 2003a. Effects of bacterial kidney disease on saltwater preference of juvenile spring Chinook salmon, *Oncorhynchus tshawytscha*. Aquaculture 222:331–341.

Price, C. S., and C. B. Schreck.2003b. Stress and saltwater-entry behaviour of juvenile chinook salmon (*Oncorhynchus tshawytscha*): conflicts in physiological motivation. Canadian Journal of Fisheries and Aquatic Sciences 60:910–918.

Queiroga, H. 1998. Vertical migration and selective tidal stream transport in the megalopa of the crab *Carcinus maenus*. Hydrobiologia 375–376:137–149.

R Development Core Team 2014. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Ransom, B.H., T. W. Steig, M. A. Timko, and P. A. Nealson. 2008. Basin-wide monitoring of salmon smolts at US dams. International Journal of Hydropower Dams 15:43–49.

Raymond, H. L. 1979. Effects of dams and impoundments on migrations of juvenile Chinook salmon and steelhead from the Snake River, 1966 to 1975. Transactions of the American Fisheries Society 108:505-529.

Redding, J. M., and C. B. Schreck. 1983. Influence of ambient salinity on osmoregulation and cortisol concentration in yearling Coho salmon during stress. Transactions of the American Fisheries Society 112:800– 807.

Renkawitz, M. D., and T. F. Sheehan. 2011. Feeding ecology of early marine phase Atlantic salmon *Salmo salar* post-smolts. Journal of Fish Biology 79:356–373.

Renkawitz, M. D., T. F. Sheehan, and G. S. Goulette. 2012. Swimming depth, behaviour, and survival of Atlantic salmon postsmolts in Penobscot Bay, Maine. Transactions of the American Fisheries Society 141:1219–1229.

Ross, M. J., and C. F. Kleiner. 1982. Shielded-needle technique for surgically implanting radio-frequency transmitters in fish. Progressive Fish Culturist 44:41-49.

Russell, I. C., M. W. Aprahamlam, J. Barry, I. C. Davidson,P. Fiske, A. T. Ibbotson, R. J. Kennedy, J. C. Maclean, A. Moore, J. Otero, E. C. E. Potter, and C. D. Todd. 2012. The influence of the freshwater environment and the biological characteristics of Atlantic salmon smolts on their subsequent marine survival. International Council for the Exploration of the Sea Journal of Marine Science 69:1563–1573.

Salminen, M., S. Kuikka,and E. Erkamo. 1995. Annual variability in survival of sea-ranched Baltic salmon, *Salmo salar* L: significance of smolt size and marine conditions. Fisheries Management and Ecology 2:171–184.

Salomniemi, I., E. Jokikokko, I. Kallio-Nyberg, E. Jutila, and P. Psanen. 2004. Survival of reared and wild Atlantic salmon smolts: size matters more in bad years. International Council for the Exploration of the Sea Journal of Marine Science 61:782–787.

Saunders, R., M. A. Hacheyk, and C. W. Fay. 2006. Maine’s diadromous fish community. Fisheries 31:537-547.

Schaller, H. A., C. E. Petrosky, and E. S. Tinus. 2014. Evaluating river management during seaward migration to recover Columbia River stream-type Chinook salmon considering variation in marine conditions. Canadian Journal of Fisheries and Aquatic Sciences 71:259–271.

Schlosser, I. J. 1982. Fish community structure and function along two habitat gradients in a headwater stream. Ecological Monographs 52:395–414.

Schreck, C. B. 1982. Stress and rearing of salmonids. Aquaculture 28:241–249.

Schreck, C. B., T. P. Stahl, L. E. Davis, D. D. Roby, and B. J. Clemens. 2006. Mortality estimates of juvenile spring-summer Chinook salmon in the Lower Columbia River and Estuary, 1992–1998: evidence for delayed mortality? Transactions of the American Fisheries Society 135:457–475.

Seber, G. A. F. 1982. The Estimation of animal abundance and related parameters, second edition. Macmillan, New York, NY.

Seiwell, H. R. 1932. Some physical characteristics of the water of Penobscot Bay, Maine, especially in relation to the tides and a discussion of the results obtained by duplicate measurements of specific gravity of sea water by the Knudsen and Plummet methods. Internationale Revue der Gesamten Hydrobioligie and Hydrographie 27:315–331.

Serrano, X., M. Grosell, and J. E. Serafy. 2010. Salinity selection and preference of the grey snapper *Lutjanus griseus*: field and laboratory observations. Journal of Fish Biology 76:1592–1608.

Sheehan, T. F., M. D. Renkawitz, and R. W. Brown. 2011. Surface trawl survey for US origin Atlantic salmon *Salmo salar*. Journal of Fish Biology 79:374–398.

Shepard, S. L. 1991. Report on radio telemetry investigations of Atlantic salmon smolt migration in the Penobscot River. Bangor Hydro-Electric Company, Bangor, ME.

Shrimpton, J. M., D. A. Patterson, J. G. Richards, S. J. Cooke, J. M. Schulte, S. G. Hinch, A. P. Farrell. 2005. Ionoregulatory changes in different populations of maturing sockeye salmon *Oncorhynchus nerka* during ocean and river migration. Journal of Experimental Biology 208:4069–4078.

Simenstad, C., D. Reed, and M. Ford. 2006. When is restoration not? Incorporating landscape-scale processes to restore self-sustaining ecosystems in coastal wetland restoration. Ecological Engineering 26:27-39.

Skalski, J. R., R. A. Buchanan, R. L. Townsend, T. W. Steig, and S. Hemstrom. 2009. A multiple-release model to estimate route-specific and dam passage survival at a hydroelectric project. North American Journal of Fisheries Management 29:670-679.

Smith, P. K., R. I. Krohn, G. T. Hermanson, A. K. Mallia, F. H. Gartner, M. D. Provenzano, E. K. Fujimoto, N. M. Goeke, B. J. Olson, and D. C. Klenk. 1985. Measurements of protein using bicinchoninic acid. Analytical Biochemistry 150:76–85.

Smith, S. G., W. M. Muir, E. E. Hockersmith, R. W. Zabel, R. J. Graves, C. V. Ross, W. P. Connor, and B. D. Arnsberg. 2003. Influence of river conditions on survival and travel time of Snake River subyearling fall Chinook salmon. North American Journal of Fisheries Management 23:939-961.

Solomon, D.J. 1978. Migration of smolts of Atlantic salmon (*Salmo salar* L.) and sea trout (*Salmo trutta* L.) in a chalkstream. Environmental Biology of Fishes 3:223–229.

Spencer, R. C., J. Zydlewski,and G. Zydlewski. 2010. Migratory urge and gill Na^+,K^+-ATPase activity of hatchery-reared Atlantic salmon smolts from the Dennys and Penobscot River stocks, Maine. Transactions of the American Fisheries Society 139:947–956.

Stefansson, S. O., B. T. Bjornsson, K. Sundell, G. Nyhammer, G., and S. D. McCormick. 2003. Physiological characteristics of wild Atlantic salmon post-smolts during estuarine and coastal migration. Journal of Fish Biology 63:942–955.

Stich, D. S, M. M. Bailey, J. D. Zydlewski. 2014. Survival of Atlantic salmon *Salmo salar* smolts through a hydropower complex in the lower Penobscot River, Maine USA. Journal of Fish Biology 85:1074–1096.

Stich, D. S., Kocik, J. F., Zydlewski, G. B. & Zydlewski, J. D. (In review). Linking behavior, physiology, and survival of Atlantic salmon smolts during estuary migration. Marine and Coastal Fisheries: Dynamics, Management, and Ecosystem Science.

Stier, D. J., and B. Kynard. 1986. Use of radio telemetry to determine the mortality of Atlantic salmon smolts passed through a 17-MW Kaplan turbine at a low-head hydroelectric dam. Transactions of the American Fisheries Society 115:771–775.

Strand, J. E. T., J. G. Davidsen, E. H. Jørgensen, and A. H. Rikardsen. 2010. Seaward migrating Atlantic salmon smolts with low levels of gill Na^+,K^+-ATPase activity; is sea entry delayed? Environmental Biology of Fishes 90:317–321.

Sturtz, S., U. Ligges, and A. Gelman, A. 2005. R2WinBUGS: a package for running WinBUGS from R. Journal of Statistical Software 1:1–16.

Svenning, M. A., R. Borgstrøm, T. O. Dehli, G. Moen, R. T. Barrett, T. Pedersen, and W. Vader. 2005. The impact of marine fish predation on Atlantic salmon smolts (*Salmo salar*) in the Tana estuary, North Norway, in the presence of and alternative prey, Lesser Sandeel (Ammodytes marinus). Fisheries Research 76:466–474.

Sykes, G. E., and J. M. Shrimpton. 2010. Effect of temperature and current manipulation on smolting in Chinook salmon (Oncorhynchus tshawytscha): the relationship between migratory behaviour and physiological development. Canadian Jounral of Fisheries and Aquatic Sciences 67:191–201.

Sykes, G. E., C. J. Johnson, and J. M. Shrimpton. 2009. Temperature and flow effects on migration timing of Chinook salmon smolts. Transactions of the American Fisheries Society 138:1252–1265.

Tenan, S., R. B. O'hara, I. Hendriks, and G. Tevecchia. 2014. Bayesian model selection: the steepest mountain to climb. Ecological Modelling 283:62–69.

Thorstad, E. B., F. Økland, B. Finstad, R. Sivertsgård, N. Plantalech, P. A. Bjørn, and R. S. McKinley. 2007. Fjord migration and survival of wild and hatchery-reared Atlantic salmon and wild brown trout post-smolts. Hydrobiologia 582:99–107.

Thorstad, E. B., F. Whoriskey, I. Uglem, A. Moore, A. H. Rikardsen, and B. Finstad. 2012a. A critical life stage of the Atlantic salmon *Salmo salar*: behaviour and survival during the smolt and initial post-smolt migration. Journal of Fish Biology 81:500–542.

Thorstad, E. B., I. Uglem, B. Finstad, F. Kroglund, I. E. Einarsdottir, T. Kristensen, O. Diserud, P. Arechavala-Lopez, I. Mayer, A. Moore, R. Nilsen, B. T. Björnsson, and F. Økland. 2013. Reduced marine survival of hatchery Atlantic salmon post-smolts exposed to aluminum and moderate acidification in freshwater. Estuarine, Coastal and Shelf Science 124:34–43.

Thorstad, E. B., I. Uglem, B. Finstad, smolts C. M. Chittenden, R. Nilsen, F. Økland, and P. A. Bjørn. 2012b. Stocking location and predation by marine fishes affect survival of hatchery-reared Atlantic salmon. Fisheries Management and Ecology 19:400–409.

Todd, C. D., K. D. Friedland, J. C. MacLean, B. D. Whyte, I. C. Russell, M. E. Lonergan, and M. B. Morrissey. Phenological and phenotypic changes in Atlantic salmon populations in response to a changing climate. International Council for the Exploration of the Sea Journal of Marine Science 69:1686–1698.

Trinko Lake, T. R., K. R. Ravana, and R. Saunders. 2012. Evaluating changes in diadromous species distributions and habitat accessibility following the Penobscot River Restoration Project. Marine and Coastal Fisheries: Dynamics, Management, and Ecosystem Science 4:284–293.

USASAC (United States Atlantic salmon Assessment Committee). 2012. Annual report of the U.S. Atlantic salmon Assessment Committee. USASAC Report 24, USASAC, Old Lyme, Connecticut. Available from http://www.nefsc.noaa.gov/USASAC/Reports/ [accessed 13 August 2014].

USASAC (United States Atlantic salmon Assessment Committee). 2014. Annual report of the U.S. Atlantic salmon Assessment Committee. USASAC Report 26, USASAC, Old Lyme, Connecticut. Available from http://www.nefsc.noaa.gov/USASAC/Reports/ [accessed 13 August 2014].

USFWS (United States Fish and Wildlife Service) & NOAA (National Oceanic and Atmospheric Administration). 2000. Endangered and threatened species; final endangered status for a distinct population segment of anadromous Atlantic salmon (*Salmo salar*) in the Gulf of Maine. Federal Register 65:69459-69483.

USFWS (United States fish and Wildlife Service) & NOAA (National Oceanic and Atmospheric Administration). 2009. Endangered and threatened species; determination of endangered status for the Gulf of Maine distinct population segment of Atlantic salmon. Federal Register 74:29344-29387.

Vannote, R. L., G. W. Minshall, K. W. Cummins, J. R. Sedell, and C. E. Cushing. 1980. The river continuum concept. Canadian Journal of Fisheries and Aquatic Sciences 37:130-137.

Venditti, D. A., D. W. Rondorf, and J. M. Krauft J. M. 2000. Migratory behavior and forebay delay of radio-tagged juvenile fall Chinook salmon in a Lower Snake River impoundment. North American Journal of Fisheries Management 20:41-52.

Walters, C. 1997. Challenges in adaptive management of riparian and coastal ecosystems. Conservation Ecology 1. Available at http://www.ecologyandsociety.org/vol1/iss2/art1/ (last accessed 17 September 2013).

Weitkamp, L. A., G. Goulette, J. Hawkes, M. O'Malley, and C. Lipsky. 2014. Juvenile salmon in estuaries: comparisons between North American Atlantic and Pacific salmon populations. Reviews in Fish Biology and Fisheries. doi: 10.1007/s11160-014-9345-y.

Whalen, K. G., D. L. Parrish, and S. D. McCormick. 1999. Migration timing of Atlantic salmon smolts relative to environmental and physiological factors. Transactions of the American Fisheries Society 128:289–301.

White, G. C., and K. P. Burnham. 1999. Program MARK: survival estimation from populations of marked animals. Bird Study 46:120–139.

Zar, J.H. 1999. Biostatistical Analysis. Prentice Hall, Upper Saddle River, New Jersey.

Zaugg, W. S. & L. R. McLain. 1970. Adenosinetriphosphatase activity in gills of salmonids: seasonal variations and salt water influence in coho salmon, *Oncorhynchus kisutch*. Comparative Biochemistry and Physiology 35:587–596.

Zaugg, W. S., and H. H. Wagner. 1973. Gill ATPase activity related to parr–smolt transformation and migration in steelhead trout (*Salmo gairdneri*): influence of photoperiod and temperature. Comparative Biochemistry & Physiology B: Comparative Biochemistry 45:955–965.

Zaugg, W. S., 1982. Some changes in smoltification and seawater adaptability of salmonids resulting from environmental and other factors. Aquaculture 28:143–151.

Zaugg, W. S., and L. R. McLain. 1972. Changes in gill adenosinetriphosphatase activity associated with parr-smolt transformation in steelhead trout, coho, and spring Chinook salmon. Journal of the Fisheries Research Board of Canada 29:161–171.

Ziv, G., E. Baran, S. Nam, I. Rodríguez-Iturbe, and S. A. Levin. 2012. Trading-off fish biodiversity, food security, and hydropower in the Mekong River Basin. Proceedings of the National Academy of Science of the United States of America 109:5609–5614.

Zuur, A., E. N. Ieno, N. Walker, A. A. Saveliev, and G. M. Smith. 2009. Mixed Effects Models and Extensions in Ecology with R. Springer, New York, New York.

Zydlewski, G. B, A. Haro, and S. D. McCormick. 2005. Evidence for cumulative temperature as an initiating and terminating factor in downstream migratory behavior of Atlantic salmon (*Salmo salar*) smolts. Canadian Journal of Fisheries and Aquatic Sciences 62:68–78.

Zydlewski, G. B., and J. Zydlewski. 2012. Gill Na^+,K^+-ATPase of Atlantic salmon smolts in freshwater is not a predictor of long-term growth in seawater. Aquaculture 362–363:121–126.

Zydlewski, G. B., D. S. Stich, and S. D. McCormick. 2014. Photoperiod control of the initiation of downstream migration in Atlantic salmon smolts. Journal of Fish Biology 85:1074–1096.

Zydlewski, J., and M. P. Wilkie. 2013. Freshwater to saltwater transitions in migratory fishes. Pages 254–294 in S. D. McCormick, A. P. Farrell, and C. J. Brauner, editors, Euryhaline Fishes. Academic Press, Oxford, United Kingdom.

Zydlewski, J., G. Zydlewski, and G. R. Danner. 2010. Descaling injury impairs the osmoregulatory ability of Atlantic salmon smolts entering seawater. Transactions of the American Fisheries Society 138:129–136.

APPENDIX A

MULTIANNUAL SURVIVAL ESTIMATES FOR PENOBSCOT RIVER

Table A.1. Multiannual freshwater survival estimates for acoustically tagged smolts in all years 2005–2014. Mean, S.D., and 95% credible intervals (CRI) of per-kilometer survival estimates from multi-state mark-recapture models of Atlantic salmon *Salmo salar* smolt survival in the Penobscot River, Maine, U.S.A. 2005–2014. Parameters correspond to the model schematic (Figure 2). Locations of head ponds and dams in each part of the catchment are indicated where appropriate. The variable *L* indicates the length (in kilometers) of each reach used in MS models.

Parameter	Reach	Mean	S.D.	95% CRI	L
$\hat{S}_1^A$	Weldon Head Pond 01	0.992	0.004	0.983–0.998	2.5
$\hat{S}_1^B$		0.997	0.001	0.995–0.999	7.0
$\hat{S}_2^A$	Weldon Head Pond 02	0.991	0.003	0.984–0.997	3.0
$\hat{S}_2^B$	Guilford Dam	0.978	0.007	0.962–0.991	1.5
$\hat{S}_3^A$	Weldon Head Pond 03	0.978	0.009	0.957–0.992	1.0
$\hat{S}_3^B$	Dover Head Pond	0.995	0.001	0.993–0.997	11.8
$\hat{S}_4^A$	Weldon Dam	0.952	0.009	0.933–0.970	2.4
$\hat{S}_4^B$	Dover (Moosehead) Dam	0.965	0.011	0.942–0.984	1.2
$\hat{S}_5^A$		0.995	0.003	0.989–1.000	6.6
$\hat{S}_5^B$	Brown Mills Dam	0.976	0.013	0.946–0.998	1.0
$\hat{S}_6^A$		0.989	0.002	0.986–0.992	12.4
$\hat{S}_6^B$		0.998	0.001	0.995–0.999	15.5
$\hat{S}_7^A$		0.999	0.001	0.998–1.000	13.8
$\hat{S}_7^B$		0.997	0.001	0.996–0.998	38.4

Table A.1. (Continued)

$\hat{S}_8^A$	West Enfield Head Pond	0.998	0.001	0.997–0.999	14.4
$\hat{S}_8^B$	Howland Head Pond	0.993	0.001	0.991–0.996	9.7
$\hat{S}_9^A$	West Enfield Dam	0.955	0.006	0.941–0.967	2.8
$\hat{S}_9^B$	Howland Dam	0.940	0.009	0.920–0.957	1.25
$\hat{S}_{10}^A$		0.997	0.001	0.995–0.999	6.43
$\hat{S}_{11}^A$		0.998	0.000	0.997–0.998	20.1
$\hat{S}_{12}^A$		0.994	0.001	0.992–0.996	6.0
$\hat{S}_{13}^A$	Milford Head Pond	0.996	0.001	0.993–0.998	3.9
$\hat{S}_{13}^C$	Gilman Falls	0.995	0.005	0.983–1.000	1.5
$\hat{S}_{14}^A$	Milford Dam	0.964	0.004	0.957–0.971	2.9
$\hat{S}_{14}^C$	Stillwater Dam	0.989	0.003	0.982–0.994	6.1
$\hat{S}_{15}^A$	Great Works Dam	0.997	0.001	0.995–0.998	8.0
$\hat{S}_{15}^C$	Orono Dam	0.990	0.004	0.982–0.997	4.1
$\hat{S}_{16}^A$	Veazie Head Pond	0.996	0.002	0.991–0.999	1.8
$\hat{S}_{17}^A$	Veazie Dam	0.984	0.004	0.977–0.991	1.5

APPENDIX B

MULTIANNUAL SURVIVAL OF WILD AND HATCHERY SMOLTS

Table B.1. Multiannual survival for wild and hatchery-reared smolts. Mean, S.D., and 95% credible intervals (CRI) of per-kilometer survival estimates from multi-state mark-recapture models of hatchery- and wild-reared Atlantic salmon *Salmo salar* smolt survival in the Penobscot River, Maine, U.S.A. 2005–2014. Parameters correspond to the model schematic (Figure 2). Locations of head ponds and dams in each part of the catchment are indicated where appropriate. Rearing history is indicated in the column 'Rearing'. The variable *L* indicates the length (in kilometers) of each reach used in MS models.

Parameter	Reach	Rearing	Mean	S.D.	95% CRI	L
$\hat{S}_1^A$	Weldon Head Pond 01	Hatchery	0.989	0.005	0.979–0.997	2.5
$\hat{S}_1^B$		Hatchery	0.996	0.002	0.992–0.999	7
$\hat{S}_2^A$	Weldon Head Pond 02	Hatchery	0.990	0.004	0.981–0.997	3
$\hat{S}_2^B$	Guilford Dam	Hatchery	0.966	0.012	0.941–0.986	1.5
$\hat{S}_3^A$	Weldon Head Pond 03	Hatchery	0.970	0.012	0.944–0.989	1
$\hat{S}_3^B$	Dover Head Pond	Hatchery	0.994	0.002	0.991–0.997	12
$\hat{S}_4^A$	Weldon Dam	Hatchery	0.960	0.010	0.939–0.980	2.4
$\hat{S}_4^B$	Dover (Moosehead) Dam	Hatchery	0.978	0.012	0.951–0.997	1.2
$\hat{S}_5^A$		Hatchery	0.995	0.003	0.988–1.000	6.6
$\hat{S}_5^B$	Brown Mills Dam	Hatchery	0.984	0.013	0.950–0.999	1
$\hat{S}_6^A$		Hatchery	0.992	0.002	0.989–0.995	12
$\hat{S}_6^B$		Hatchery	0.999	0.001	0.997–1.000	16
$\hat{S}_7^A$		Hatchery	0.999	0.001	0.998–1.000	14

Table B.1. (Continued)

$\hat{S}_7^B$		Hatchery	0.995	0.001	0.994–0.997	38
$\hat{S}_8^A$	West Enfield Head Pond	Hatchery	0.998	0.001	0.996–0.999	14
$\hat{S}_8^B$	Howland Head Pond	Hatchery	0.995	0.001	0.992–0.997	9.7
$\hat{S}_9^A$	West Enfield Dam	Hatchery	0.959	0.007	0.944–0.972	2.8
$\hat{S}_9^B$	Howland Dam	Hatchery	0.940	0.010	0.919–0.958	1.2
$\hat{S}_{10}^A$		Hatchery	0.997	0.001	0.995–0.999	6.4
$\hat{S}_{11}^A$		Hatchery	0.998	0.000	0.997–0.998	20
$\hat{S}_{12}^A$		Hatchery	0.995	0.001	0.993–0.997	6
$\hat{S}_{13}^A$	Milford Head Pond	Hatchery	0.996	0.002	0.993–0.999	3.9
$\hat{S}_{13}^C$	Gilman Falls	Hatchery	0.994	0.006	0.980–1.000	1.5
$\hat{S}_{14}^A$	Milford Dam	Hatchery	0.963	0.004	0.955–0.971	2.9
$\hat{S}_{14}^C$	Stillwater Dam	Hatchery	0.991	0.003	0.986–0.996	6.1
$\hat{S}_{15}^A$	Great Works Dam	Hatchery	0.997	0.001	0.995–0.999	8
$\hat{S}_{15}^C$	Orono Dam	Hatchery	0.989	0.004	0.981–0.996	4.1
$\hat{S}_{16}^A$	Veazie Head Pond	Hatchery	0.997	0.002	0.991–1.000	1.8
$\hat{S}_{17}^A$	Veazie Dam	Hatchery	0.986	0.004	0.978–0.992	1.5
$\hat{S}_1^A$	Weldon Head Pond 01	Wild	0.993	0.007	0.974–1.000	2.5

Table B.1. (Continued)

$\hat{S}_1^{B}$		Wild	0.997	0.002	0.993–1.000	7
$\hat{S}_2^{A}$	Weldon Head Pond 02	Wild	0.991	0.006	0.976–0.999	3
$\hat{S}_2^{B}$	Guilford Dam	Wild	0.989	0.008	0.971–0.999	1.5
$\hat{S}_3^{A}$	Weldon Head Pond 03	Wild	0.986	0.013	0.951–1.000	1
$\hat{S}_3^{B}$	Dover Head Pond	Wild	0.996	0.002	0.992–0.998	12
$\hat{S}_4^{A}$	Weldon Dam	Wild	0.930	0.021	0.886–0.966	2.4
$\hat{S}_4^{B}$	Dover (Moosehead) Dam	Wild	0.945	0.019	0.902–0.978	1.2
$\hat{S}_5^{A}$		Wild	0.991	0.006	0.976–0.999	6.6
$\hat{S}_5^{B}$	Brown Mills Dam	Wild	0.960	0.023	0.906–0.996	1
$\hat{S}_6^{A}$		Wild	0.981	0.004	0.973–0.989	12
$\hat{S}_6^{B}$		Wild	0.995	0.002	0.990–0.998	16
$\hat{S}_7^{A}$		Wild	0.997	0.002	0.992–1.000	14
$\hat{S}_7^{B}$		Wild	0.999	0.001	0.998–1.000	38
$\hat{S}_8^{A}$	West Enfield Head Pond	Wild	0.998	0.001	0.995–1.000	14
$\hat{S}_8^{B}$	Howland Head Pond	Wild	0.993	0.003	0.985–0.997	9.7
$\hat{S}_9^{A}$	West Enfield Dam	Wild	0.941	0.014	0.912–0.966	2.8
$\hat{S}_9^{B}$	Howland Dam	Wild	0.952	0.021	0.903–0.985	1.2
$\hat{S}_{10}^{A}$		Wild	0.998	0.002	0.993–1.000	6.4
$\hat{S}_{11}^{A}$		Wild	0.997	0.001	0.995–0.999	20

Table B.1. (Continued)

$\hat{S}^{A}_{12}$		Wild	0.986	0.004	0.978–0.992	6
$\hat{S}^{A}_{13}$	Milford Head Pond	Wild	0.994	0.004	0.985–0.999	3.9
$\hat{S}^{C}_{13}$	Gilman Falls	Wild	0.973	0.026	0.903–0.999	1.5
$\hat{S}^{A}_{14}$	Milford Dam	Wild	0.971	0.009	0.952–0.986	2.9
$\hat{S}^{C}_{14}$	Stillwater Dam	Wild	0.961	0.015	0.926–0.986	6.1
$\hat{S}^{A}_{15}$	Great Works Dam	Wild	0.995	0.003	0.989–0.999	8
$\hat{S}^{C}_{15}$	Orono Dam	Wild	0.988	0.011	0.958–1.000	4.1
$\hat{S}^{A}_{16}$	Veazie Head Pond	Wild	0.986	0.009	0.965–0.999	1.8
$\hat{S}^{A}_{17}$	Veazie Dam	Wild	0.968	0.013	0.939–0.988	1.5

APPENDIX C

ANNUAL SURVIVAL ESTIMATES FOR YEARS 2005–2014

Table C.1. Annual freshwater survival estimates for each year 2005 through 2014. Mean, S.D., and 95% credible intervals (CRI) of annual, per-kilometer survival estimates from multi-state mark-recapture models of Atlantic salmon *Salmo salar* smolt survival in the Penobscot River, Maine, U.S.A. 2005–2014. Parameters correspond to the model schematic (Figure 2). Locations of head ponds and dams in each part of the catchment are indicated where appropriate. The variable *L* indicates the length (in kilometers) of each reach used in MS models. The symbol "-" indicates years during which survival was not estimable through specific reaches based on release locations used in those years.

Parameter	Reach	Year	Mean	S.D.	95% CRI	L
$\hat{S}_1^A$	Weldon Head Pond 01	2005	-	-	-	2.5
$\hat{S}_1^B$		2005	-	-	-	7
$\hat{S}_2^A$	Weldon Head Pond 02	2005	-	-	-	3
$\hat{S}_2^B$	Guilford Dam	2005	-	-	-	1.5
$\hat{S}_3^A$	Weldon Head Pond 03	2005	-	-	-	1
$\hat{S}_3^B$	Dover Head Pond	2005	-	-	-	11.8
$\hat{S}_4^A$	Weldon Dam	2005	-	-	-	2.4
$\hat{S}_4^B$	Dover (Moosehead) Dam	2005	-	-	-	1.2
$\hat{S}_5^A$		2005	-	-	-	6.6
$\hat{S}_5^B$	Brown Mills Dam	2005	-	-	-	1
$\hat{S}_6^A$		2005	-	-	-	12.4
$\hat{S}_6^B$		2005	-	-	-	15.5
$\hat{S}_7^A$		2005	-	-	-	13.8

Table C.1. (Continued)

$\hat{S}_7^B$		2005	0.991	0.002	0.987–0.995	38.4
$\hat{S}_8^A$	West Enfield Head Pond	2005	0.995	0.004	0.986–1.000	14.4
$\hat{S}_8^B$	Howland Head Pond	2005	0.996	0.004	0.985–1.000	9.7
$\hat{S}_9^A$	West Enfield Dam	2005	0.942	0.027	0.880–0.987	2.8
$\hat{S}_9^B$	Howland Dam	2005	0.928	0.038	0.841–0.989	1.2
$\hat{S}_{10}^A$		2005	0.990	0.005	0.978–0.999	6.4
$\hat{S}_{11}^A$		2005	0.998	0.001	0.994–1.000	20.1
$\hat{S}_{12}^A$		2005	0.995	0.003	0.987–1.000	6
$\hat{S}_{13}^A$	Milford Head Pond	2005	0.994	0.005	0.982–1.000	3.9
$\hat{S}_{13}^C$	Gilman Falls	2005	0.971	0.028	0.897–0.999	1.5
$\hat{S}_{14}^A$	Milford Dam	2005	0.969	0.011	0.945–0.989	2.9
$\hat{S}_{14}^C$	Stillwater Dam	2005	0.989	0.008	0.968–1.000	6.1
$\hat{S}_{15}^A$	Great Works Dam	2005	0.996	0.003	0.989–1.000	8
$\hat{S}_{15}^C$	Orono Dam	2005	0.989	0.010	0.961–1.000	4.1
$\hat{S}_{16}^A$	Veazie Head Pond	2005	0.993	0.006	0.976–1.000	1.8
$\hat{S}_{17}^A$	Veazie Dam	2005	0.993	0.007	0.976–1.000	1.5
$\hat{S}_1^A$	Weldon Head Pond 01	2006	-	-	-	2.5
$\hat{S}_1^B$		2006	-	-	-	7
$\hat{S}_2^A$	Weldon Head Pond 02	2006	-	-	-	3

Table C.1. (Continued)

$\widehat{S}_{2}^{B}$	Guilford Dam	2006	-	-	-	1.5
$\widehat{S}_{3}^{A}$	Weldon Head Pond 03	2006	-	-	-	1
$\widehat{S}_{3}^{B}$	Dover Head Pond	2006	-	-	-	11.8
$\widehat{S}_{4}^{A}$	Weldon Dam	2006	-	-	-	2.4
$\widehat{S}_{4}^{B}$	Dover (Moosehead) Dam	2006	-	-	-	1.2
$\widehat{S}_{5}^{A}$		2006	-	-	-	6.6
$\widehat{S}_{5}^{B}$	Brown Mills Dam	2006	-	-	-	1
$\widehat{S}_{6}^{A}$		2006	0.977	0.004	0.969–0.983	12.4
$\widehat{S}_{6}^{B}$		2006	-	-	-	15.5
$\widehat{S}_{7}^{A}$		2006	0.999	0.001	0.997–1.000	13.8
$\widehat{S}_{7}^{B}$		2006	0.975	0.025	0.908–0.999	38.4
$\widehat{S}_{8}^{A}$	West Enfield Head Pond	2006	0.996	0.002	0.992–0.998	14.4
$\widehat{S}_{8}^{B}$	Howland Head Pond	2006	0.906	0.085	0.683–0.997	9.7
$\widehat{S}_{9}^{A}$	West Enfield Dam	2006	0.927	0.015	0.896–0.954	2.8
$\widehat{S}_{9}^{B}$	Howland Dam	2006	0.757	0.048	0.657–0.845	1.2
$\widehat{S}_{10}^{A}$		2006	0.994	0.003	0.986–0.999	6.4
$\widehat{S}_{11}^{A}$		2006	0.995	0.001	0.992–0.998	20.1
$\widehat{S}_{12}^{A}$		2006	0.994	0.003	0.988–0.999	6
$\widehat{S}_{13}^{A}$	Milford Head Pond	2006	0.997	0.003	0.991–1.000	3.9

Table C.1. (Continued)

$\hat{S}_{13}^{C}$	Gilman Falls	2006	0.931	0.064	0.762–0.998	1.5
$\hat{S}_{14}^{A}$	Milford Dam	2006	0.927	0.014	0.898–0.953	2.9
$\hat{S}_{14}^{C}$	Stillwater Dam	2006	0.982	0.017	0.935–1.000	6.1
$\hat{S}_{15}^{A}$	Great Works Dam	2006	0.996	0.002	0.990–1.000	8
$\hat{S}_{15}^{C}$	Orono Dam	2006	0.973	0.026	0.903–0.999	4.1
$\hat{S}_{16}^{A}$	Veazie Head Pond	2006	0.994	0.006	0.978–1.000	1.8
$\hat{S}_{17}^{A}$	Veazie Dam	2006	0.982	0.011	0.955–0.997	1.5
$\hat{S}_{1}^{A}$	Weldon Head Pond 01	2009	-	-	-	2.5
$\hat{S}_{1}^{B}$		2009	-	-	-	7
$\hat{S}_{2}^{A}$	Weldon Head Pond 02	2009	-	-	-	3
$\hat{S}_{2}^{B}$	Guilford Dam	2009	-	-	-	1.5
$\hat{S}_{3}^{A}$	Weldon Head Pond 03	2009	-	-	-	1
$\hat{S}_{3}^{B}$	Dover Head Pond	2009	-	-	-	11.8
$\hat{S}_{4}^{A}$	Weldon Dam	2009	-	-	-	2.4
$\hat{S}_{4}^{B}$	Dover (Moosehead) Dam	2009	-	-	-	1.2
$\hat{S}_{5}^{A}$		2009	-	-	-	6.6
$\hat{S}_{5}^{B}$	Brown Mills Dam	2009	-	-	-	1
$\hat{S}_{6}^{A}$		2009	-	-	-	12.4
$\hat{S}_{6}^{B}$		2009	-	-	-	15.5

Table C.1. (Continued)

$\hat{S}_{7}^{A}$		2009	-	-	-	13.8
$\hat{S}_{7}^{B}$		2009	0.975	0.025	0.909–0.999	38.4
$\hat{S}_{8}^{A}$	West Enfield Head Pond	2009	-	-	-	14.4
$\hat{S}_{8}^{B}$	Howland Head Pond	2009	0.990	0.003	0.983–0.996	9.7
$\hat{S}_{9}^{A}$	West Enfield Dam	2009	-	-	-	2.8
$\hat{S}_{9}^{B}$	Howland Dam	2009	0.967	0.019	0.922–0.995	1.2
$\hat{S}_{10}^{A}$		2009	0.998	0.002	0.992–1.000	6.4
$\hat{S}_{11}^{A}$		2009	0.997	0.001	0.995–0.999	20.1
$\hat{S}_{12}^{A}$		2009	0.996	0.002	0.991–1.000	6
$\hat{S}_{13}^{A}$	Milford Head Pond	2009	0.993	0.004	0.982–0.999	3.9
$\hat{S}_{13}^{C}$	Gilman Falls	2009	0.979	0.021	0.923–1.000	1.5
$\hat{S}_{14}^{A}$	Milford Dam	2009	0.980	0.008	0.962–0.994	2.9
$\hat{S}_{14}^{C}$	Stillwater Dam	2009	0.995	0.005	0.981–1.000	6.1
$\hat{S}_{15}^{A}$	Great Works Dam	2009	0.992	0.003	0.985–0.997	8
$\hat{S}_{15}^{C}$	Orono Dam	2009	0.959	0.018	0.918–0.987	4.1
$\hat{S}_{16}^{A}$	Veazie Head Pond	2009	0.995	0.005	0.982–1.000	1.8
$\hat{S}_{17}^{A}$	Veazie Dam	2009	0.995	0.005	0.983–1.000	1.5
$\hat{S}_{1}^{A}$	Weldon Head Pond 01	2010	0.992	0.008	0.970–1.000	2.5
$\hat{S}_{1}^{B}$		2010	0.998	0.002	0.993–1.000	7

Table C.1. (Continued)

$\hat{S}_2^A$	Weldon Head Pond 02	2010	0.993	0.007	0.975–1.000	3
$\hat{S}_2^B$	Guilford Dam	2010	0.984	0.012	0.953–0.999	1.5
$\hat{S}_3^A$	Weldon Head Pond 03	2010	0.981	0.019	0.930–1.000	1
$\hat{S}_3^B$	Dover Head Pond	2010	0.994	0.003	0.988–0.998	11.8
$\hat{S}_4^A$	Weldon Dam	2010	0.913	0.025	0.859–0.956	2.4
$\hat{S}_4^B$	Dover (Moosehead) Dam	2010	0.914	0.032	0.844–0.967	1.2
$\hat{S}_5^A$		2010	0.995	0.005	0.983–1.000	6.6
$\hat{S}_5^B$	Brown Mills Dam	2010	0.937	0.038	0.850–0.994	1
$\hat{S}_6^A$		2010	0.996	0.003	0.989–1.000	12.4
$\hat{S}_6^B$		2010	0.991	0.004	0.982–0.997	15.5
$\hat{S}_7^A$		2010	0.996	0.003	0.989–1.000	13.8
$\hat{S}_7^B$		2010	0.998	0.001	0.996–1.000	38.4
$\hat{S}_8^A$	West Enfield Head Pond	2010	0.997	0.002	0.992–1.000	14.4
$\hat{S}_8^B$	Howland Head Pond	2010	0.992	0.002	0.987–0.996	9.7
$\hat{S}_9^A$	West Enfield Dam	2010	0.905	0.026	0.847–0.950	2.8
$\hat{S}_9^B$	Howland Dam	2010	0.956	0.016	0.919–0.983	1.2
$\hat{S}_{10}^A$		2010	0.996	0.002	0.990–0.999	6.4
$\hat{S}_{11}^A$		2010	0.998	0.001	0.996–0.999	20.1

Table C.1. (Continued)

$\hat{S}_{12}^{A}$		2010	0.988	0.003	0.982–0.993	6
$\hat{S}_{13}^{A}$	Milford Head Pond	2010	0.996	0.002	0.991–1.000	3.9
$\hat{S}_{13}^{C}$	Gilman Falls	2010	0.971	0.028	0.895–0.999	1.5
$\hat{S}_{14}^{A}$	Milford Dam	2010	0.958	0.008	0.941–0.973	2.9
$\hat{S}_{14}^{C}$	Stillwater Dam	2010	0.944	0.020	0.898–0.977	6.1
$\hat{S}_{15}^{A}$	Great Works Dam	2010	0.997	0.002	0.993–1.000	8
$\hat{S}_{15}^{C}$	Orono Dam	2010	0.985	0.014	0.947–1.000	4.1
$\hat{S}_{16}^{A}$	Veazie Head Pond	2010	0.985	0.008	0.966–0.998	1.8
$\hat{S}_{17}^{A}$	Veazie Dam	2010	0.956	0.013	0.927–0.978	1.5
$\hat{S}_{1}^{A}$	Weldon Head Pond 01	2011	0.946	0.051	0.813–0.999	2.5
$\hat{S}_{1}^{B}$		2011	0.994	0.003	0.986–0.999	7
$\hat{S}_{2}^{A}$	Weldon Head Pond 02	2011	0.975	0.018	0.930–0.997	3
$\hat{S}_{2}^{B}$	Guilford Dam	2011	0.986	0.012	0.954–1.000	1.5
$\hat{S}_{3}^{A}$	Weldon Head Pond 03	2011	0.960	0.038	0.860–0.999	1
$\hat{S}_{3}^{B}$	Dover Head Pond	2011	0.996	0.002	0.991–1.000	11.8
$\hat{S}_{4}^{A}$	Weldon Dam	2011	0.959	0.033	0.877–0.999	2.4
$\hat{S}_{4}^{B}$	Dover (Moosehead) Dam	2011	0.975	0.021	0.922–0.999	1.2
$\hat{S}_{5}^{A}$		2011	0.975	0.016	0.938–0.998	6.6

Table C.1. (Continued)

$\widehat{S}_{5}^{B}$	Brown Mills Dam	2011	0.968	0.027	0.899–0.999	1
$\widehat{S}_{6}^{A}$		2011	0.995	0.005	0.981–1.000	12.4
$\widehat{S}_{6}^{B}$		2011	0.998	0.002	0.994–1.000	15.5
$\widehat{S}_{7}^{A}$		2011	0.995	0.004	0.983–1.000	13.8
$\widehat{S}_{7}^{B}$		2011	0.999	0.001	0.997–1.000	38.4
$\widehat{S}_{8}^{A}$	West Enfield Head Pond	2011	0.996	0.004	0.986–1.000	14.4
$\widehat{S}_{8}^{B}$	Howland Head Pond	2011	0.997	0.002	0.991–1.000	9.7
$\widehat{S}_{9}^{A}$	West Enfield Dam	2011	0.969	0.025	0.907–0.999	2.8
$\widehat{S}_{9}^{B}$	Howland Dam	2011	0.940	0.023	0.892–0.980	1.2
$\widehat{S}_{10}^{A}$		2011	0.994	0.004	0.985–0.999	6.4
$\widehat{S}_{11}^{A}$		2011	1.000	0.000	0.998–1.000	20.1
$\widehat{S}_{12}^{A}$		2011	0.997	0.002	0.993–1.000	6
$\widehat{S}_{13}^{A}$	Milford Head Pond	2011	0.996	0.003	0.989–1.000	3.9
$\widehat{S}_{13}^{C}$	Gilman Falls	2011	0.982	0.018	0.933–1.000	1.5
$\widehat{S}_{14}^{A}$	Milford Dam	2011	0.981	0.008	0.964–0.995	2.9
$\widehat{S}_{14}^{C}$	Stillwater Dam	2011	0.987	0.008	0.968–0.999	6.1
$\widehat{S}_{15}^{A}$	Great Works Dam	2011	0.992	0.003	0.985–0.998	8
$\widehat{S}_{15}^{C}$	Orono Dam	2011	0.985	0.011	0.956–0.999	4.1

Table C.1. (Continued)

$\hat{S}_{16}^{A}$	Veazie Head Pond	2011	0.995	0.005	0.980–1.000	1.8
$\hat{S}_{17}^{A}$	Veazie Dam	2011	0.988	0.008	0.969–0.999	1.5
$\hat{S}_{1}^{A}$	Weldon Head Pond 01	2012	0.981	0.010	0.959–0.996	2.5
$\hat{S}_{1}^{B}$		2012	0.993	0.004	0.984–0.999	7
$\hat{S}_{2}^{A}$	Weldon Head Pond 02	2012	0.991	0.006	0.975–0.999	3
$\hat{S}_{2}^{B}$	Guilford Dam	2012	0.982	0.013	0.949–0.998	1.5
$\hat{S}_{3}^{A}$	Weldon Head Pond 03	2012	0.963	0.021	0.912–0.993	1
$\hat{S}_{3}^{B}$	Dover Head Pond	2012	0.998	0.002	0.993–1.000	11.8
$\hat{S}_{4}^{A}$	Weldon Dam	2012	0.956	0.018	0.918–0.988	2.4
$\hat{S}_{4}^{B}$	Dover (Moosehead) Dam	2012	0.977	0.017	0.933–0.998	1.2
$\hat{S}_{5}^{A}$		2012	0.990	0.006	0.975–0.998	6.6
$\hat{S}_{5}^{B}$	Brown Mills Dam	2012	0.985	0.015	0.945–1.000	1
$\hat{S}_{6}^{A}$		2012	0.998	0.002	0.993–1.000	12.4
$\hat{S}_{6}^{B}$		2012	0.999	0.001	0.996–1.000	15.5
$\hat{S}_{7}^{A}$		2012	0.999	0.001	0.995–1.000	13.8
$\hat{S}_{7}^{B}$		2012	0.999	0.001	0.997–1.000	38.4
$\hat{S}_{8}^{A}$	West Enfield Head Pond	2012	0.999	0.001	0.996–1.000	14.4
$\hat{S}_{8}^{B}$	Howland Head Pond	2012	0.998	0.002	0.993–1.000	9.7
$\hat{S}_{9}^{A}$	West Enfield Dam	2012	0.980	0.011	0.954–0.996	2.8

Table C.1. (Continued)

$\widehat{S}_{9}^{B}$	Howland Dam	2012	0.979	0.016	0.938–0.999	1.2
$\widehat{S}_{10}^{A}$		2012	0.996	0.003	0.990–1.000	6.4
$\widehat{S}_{11}^{A}$		2012	0.999	0.001	0.998–1.000	20.1
$\widehat{S}_{12}^{A}$		2012	0.998	0.002	0.993–1.000	6
$\widehat{S}_{13}^{A}$	Milford Head Pond	2012	0.997	0.003	0.990–1.000	3.9
$\widehat{S}_{13}^{C}$	Gilman Falls	2012	0.947	0.050	0.812–0.999	1.5
$\widehat{S}_{14}^{A}$	Milford Dam	2012	0.961	0.011	0.937–0.980	2.9
$\widehat{S}_{14}^{C}$	Stillwater Dam	2012	0.957	0.025	0.898–0.992	6.1
$\widehat{S}_{15}^{A}$	Great Works Dam	2012	0.996	0.002	0.990–1.000	8
$\widehat{S}_{15}^{C}$	Orono Dam	2012	0.975	0.025	0.910–0.999	4.1
$\widehat{S}_{16}^{A}$	Veazie Head Pond	2012	0.994	0.006	0.977–1.000	1.8
$\widehat{S}_{17}^{A}$	Veazie Dam	2012	0.968	0.014	0.935–0.989	1.5
$\widehat{S}_{1}^{A}$	Weldon Head Pond 01	2013	0.987	0.009	0.965–0.999	2.5
$\widehat{S}_{1}^{B}$		2013	0.986	0.014	0.949–1.000	7
$\widehat{S}_{2}^{A}$	Weldon Head Pond 02	2013	0.990	0.007	0.972–0.999	3
$\widehat{S}_{2}^{B}$	Guilford Dam	2013	0.940	0.024	0.886–0.980	1.5
$\widehat{S}_{3}^{A}$	Weldon Head Pond 03	2013	0.938	0.027	0.875–0.981	1
$\widehat{S}_{3}^{B}$	Dover Head Pond	2013	0.988	0.004	0.979–0.995	11.8
$\widehat{S}_{4}^{A}$	Weldon Dam	2013	0.940	0.021	0.894–0.979	2.4

Table C.1. (Continued)

$\hat{S}_4^B$	Dover (Moosehead) Dam	2013	0.961	0.027	0.894–0.998	1.2
$\hat{S}_5^A$		2013	0.994	0.005	0.980–1.000	6.6
$\hat{S}_5^B$	Brown Mills Dam	2013	0.939	0.042	0.839–0.997	1
$\hat{S}_6^A$		2013	0.996	0.003	0.989–1.000	12.4
$\hat{S}_6^B$		2013	0.995	0.004	0.987–1.000	15.5
$\hat{S}_7^A$		2013	0.998	0.002	0.992–1.000	13.8
$\hat{S}_7^B$		2013	0.995	0.002	0.991–0.999	38.4
$\hat{S}_8^A$	West Enfield Head Pond	2013	0.998	0.002	0.993–1.000	14.4
$\hat{S}_8^B$	Howland Head Pond	2013	0.970	0.010	0.947–0.987	9.7
$\hat{S}_9^A$	West Enfield Dam	2013	0.971	0.014	0.938–0.993	2.8
$\hat{S}_9^B$	Howland Dam	2013	0.956	0.034	0.873–0.998	1.2
$\hat{S}_{10}^A$		2013	0.998	0.002	0.992–1.000	6.4
$\hat{S}_{11}^A$		2013	0.995	0.002	0.990–0.998	20.1
$\hat{S}_{12}^A$		2013	0.988	0.006	0.974–0.998	6
$\hat{S}_{13}^A$	Milford Head Pond	2013	0.967	0.012	0.940–0.987	3.9
$\hat{S}_{13}^C$	Gilman Falls	2013	0.818	0.150	0.443–0.994	1.5
$\hat{S}_{14}^A$	Milford Dam	2013	0.949	0.017	0.912–0.977	2.9
$\hat{S}_{14}^C$	Stillwater Dam	2013	0.878	0.081	0.678–0.987	6.1
$\hat{S}_{15}^A$	Great Works Dam	2013	0.997	0.003	0.990–1.000	8

Table C.1. (Continued)

$\hat{S}_{15}^{C}$	Orono Dam	2013	0.888	0.099	0.630–0.997	4.1
$\hat{S}_{16}^{A}$	Veazie Head Pond	2013	0.973	0.017	0.931–0.997	1.8
$\hat{S}_{17}^{A}$	Veazie Dam	2013	0.984	0.015	0.943–1.000	1.5
$\hat{S}_{1}^{A}$	Weldon Head Pond 01	2014	0.992	0.007	0.973–1.000	2.5
$\hat{S}_{1}^{B}$		2014	0.997	0.003	0.989–1.000	7
$\hat{S}_{2}^{A}$	Weldon Head Pond 02	2014	0.982	0.009	0.960–0.996	3
$\hat{S}_{2}^{B}$	Guilford Dam	2014	0.955	0.027	0.891–0.995	1.5
$\hat{S}_{3}^{A}$	Weldon Head Pond 03	2014	0.985	0.014	0.948–1.000	1
$\hat{S}_{3}^{B}$	Dover Head Pond	2014	0.994	0.003	0.987–0.999	11.8
$\hat{S}_{4}^{A}$	Weldon Dam	2014	0.981	0.013	0.950–0.999	2.4
$\hat{S}_{4}^{B}$	Dover (Moosehead) Dam	2014	0.962	0.026	0.898–0.998	1.2
$\hat{S}_{5}^{A}$		2014	0.992	0.005	0.979–1.000	6.6
$\hat{S}_{5}^{B}$	Brown Mills Dam	2014	0.966	0.029	0.894–0.999	1
$\hat{S}_{6}^{A}$		2014	0.998	0.002	0.993–1.000	12.4
$\hat{S}_{6}^{B}$		2014	0.998	0.002	0.994–1.000	15.5
$\hat{S}_{7}^{A}$		2014	0.997	0.002	0.992–1.000	13.8
$\hat{S}_{7}^{B}$		2014	0.999	0.001	0.998–1.000	38.4
$\hat{S}_{8}^{A}$	West Enfield Head Pond	2014	0.999	0.001	0.995–1.000	14.4
$\hat{S}_{8}^{B}$	Howland Head Pond	2014	0.998	0.002	0.992–1.000	9.7

Table C.1. (Continued)

$\hat{S}_{9}^{A}$	West Enfield Dam	2014	0.988	0.009	0.965–0.999	2.8
$\hat{S}_{9}^{B}$	Howland Dam	2014	0.979	0.018	0.932–0.999	1.2
$\hat{S}_{10}^{A}$		2014	0.997	0.002	0.991–1.000	6.4
$\hat{S}_{11}^{A}$		2014	0.996	0.001	0.993–0.999	20.1
$\hat{S}_{12}^{A}$		2014	0.991	0.005	0.980–0.998	6
$\hat{S}_{13}^{A}$	Milford Head Pond	2014	0.987	0.008	0.969–0.998	3.9
$\hat{S}_{13}^{C}$	Gilman Falls	2014	0.957	0.041	0.847–0.999	1.5
$\hat{S}_{14}^{A}$	Milford Dam	2014	0.981	0.009	0.959–0.995	2.9
$\hat{S}_{14}^{C}$	Stillwater Dam	2014	0.994	0.003	0.986–0.999	6.1
$\hat{S}_{15}^{A}$	Great Works Dam	2014	0.998	0.002	0.993–1.000	8
$\hat{S}_{15}^{C}$	Orono Dam	2014	0.990	0.006	0.977–0.999	4.1
$\hat{S}_{16}^{A}$	Veazie Head Pond	2014	0.993	0.006	0.978–1.000	1.8
$\hat{S}_{17}^{A}$	Veazie Dam	2014	0.994	0.005	0.981–1.000	1.5

BIOGRAPHY OF THE AUTHOR

Dan Stich was born in Oneida, New York, on March 17, 1983. He grew up moving around the state and fishing as he went. Dan graduated from Burnt Hills-Ballston Lake High School in 2001. He attended Schenectady County Community College, and graduated with an Associate of Arts in 2006, at which point he learned that people got paid to work with fish. Re-invigorated, Dan attended SUNY Cobleskill and graduated with a Bachelor of Technology in Fisheries and Aquaculture in 2008. He worked various jobs in the field during his time there, prior to moving to Blacksburg, VA, where he earned a Master of Science in Fisheries and Wildlife and a graduate certification in college teaching at Virginia Polytechnic Institute and State University in 2011. Dan is a candidate for the Doctor of Philosophy degree in Wildlife Ecology from the University of Maine in December 2014.