ACFFA Aquaculture Research, Science and Technology Forum

FINAL REPORT

October 26 and 27, 2016

Huntsman Fundy Discovery Centre

St. Andrews, NB

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Acknowledgements

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A special thanks to all the speakers and presenters for their participation.

Introduction

The Atlantic Canada Fish Farmers Association hosted its annual Science, Research and Technology Forum on October 26 and 27, 2016 at the Huntsman Fundy Discovery Centre in St. Andrews, New Brunswick. The annual forum is designed to support the transfer of knowledge on aquaculture related research and development projects. It creates a venue to share results, profile new technologies, and facilitate networking opportunities on variety of industry priorities. direction or knowledge gaps.

Presentations at the 2016 forum covered a variety of themes. The collaboration and communication discussions began with presentations by the Department of Fisheries and Oceans and the Canadian Aquaculture Industry Alliance. Presentations also engaged the audience in discussions regarding how to share working waterfronts, collaborative research on lobster populations, as well as wild Atlantic salmon recovery projects in the Bay of Fundy.

New technology / development issues included presentations on Arctic char aquaculture, new feed technologies, an alternative environmental monitoring tool and an innovative virtual reality system for educating youth on aquaculture operations. Genomic tools, genetic traceability and genomic selection were also topics of discussion.

Fish health issues discussed on day two focused on the infectious salmon anemia virus (ISAv) and strain types. Other presentations included discussion of the many challenges of sea lice, ulcer disease, and antimicrobial treatments.

Over 130 individuals attended the forum. They included representatives from the aquaculture industry from across Canada, local, national and international researchers, pharmaceutical and feed companies, federal and provincial regulators as well as representatives of tourism, academia, traditional fishery and conservation interests.

AGENDA

Aquaculture Research, Science and Technology Forum
HUNTSMAN FUNDY DISCOVERY CENTRE
ST. ANDREWS, NB

<u>WEDNESDAY, OCTOBER 26, 2016</u> - COLLABORATION AND INNOVATION

8:00 **Registration and Refreshments**

8:30 Welcome and Introduction – *Susan Farquharson, Executive Director, Atlantic Canada Fish Farmers Association*
8:40 Opening Remarks – *Morley Knight, Regional Director General – Maritimes, Fisheries and Oceans Canada*
8:55 Securing a Future for Responsible & Sustainable Aquaculture in Canada Together - *Ruth Salmon, Executive Director, Canadian Aquaculture Industry Alliance*
9:20 Strategic Alliances and Collaboration as Important Community Outreach Tools – *Sebastian Belle, Executive Director, Maine Aquaculture Association*
9:45 Are you an Opponent or a Collaborator – *Brad Hicks, Taplow Feeds*
10:10 Collaborative iBoF Wild Salmon Recovery Program - *Corey Clarke, Fundy National Park*

10:35 Refreshment Break

10:55 What Goes Around Comes Around, Restoring the Upper Salmon River – *Kurt Samways, Canadian Rivers Institute / University of New Brunswick*
11:20 Impact of Salmon Aquaculture on the Diversity and Health of Benthic Communities in Shallow Coastal Habitats of the Bay of Fundy – *Heather Hunt, University of New Brunswick*
11:45 Aquaculture in Virtual Reality: exploring, educating,popularizing – *Ekaterina Prasolova-Førland, University of Science and Technology Norway (Skype)*
12:10 Aquaculture Development and Profitable Commercialization of Arctic Char in Canada – *Rodrigue Yossa, Costal Zone Research Institute*

12:35 Lunch

1:35 Genomic Tools Identify the Direct Genetic Impacts of Escaped Farmed Atlantic Salmon on Wild Populations in Southern Newfoundland – *Ian Bradbury, DFO-NL*
2:00 Genetic Traceability – *Amber Garber, Huntsman Marine Science Centre*
2:25 Genomic Selection and Genome-Wide Association Studies: perspectives and possibilities - *Tiago Hori, Center for Aquaculture Technologies Canada*
2:50 Functional Transcriptomic Characterization of *Lepeophtheirus salmonis* Rejection by Coho Salmon - *Laura Braden, University of Prince Edward Island*

3:15 **Refreshment Break**

3:35 FeedKind Protein: The Future of Aquaculture Feeds – *Dennis Leong, Calysta*
4:00 Development of an Alternative Sulfide Detection Method - *David Wong, St. Andrews Biological Station*

4:30 Wrap up / Adjournment

THURSDAY, OCTOBER 27, 2016 – FISH HEALTH

8:00 **Registration and Refreshments**

8:30 Welcome and Introduction

Panel –ISAv Strains
8:40 - 10:30:

- ISAv: What Is a Strain? - *Ben Forward, RPC*
- (Significance of HPR0 in Relation to ISA Disease Caused by HPR-Deleted ISA Variants) - *Knut Falk, Norwegian Veterinary Institute*
- ISA in the Atlantic Region - *Michael Beattie (NB), Nicole O'Brien (NL), Roland Cusack* (NS)- 10:30

Refreshment Break

10:50 Atlantic Salmon Response to ISAV: An Unwelcome Guest on Extended Stay - *Nellie Gagne, Fisheries and Oceans Canada*
11:15 ISAv – When You Should Sample and Why - *Nicole O'Brien, Newfoundland Department of Fisheries and Aquaculture*
11:40 Federal Approach to Freedom Evaluation for Reportable and Emerging Diseases - *Annie Wagener, Canadian Food Inspection Agency*

12:15 **Lunch**

1:15 The Horizontal and Vertical Distribution of Sea Lice Larvae in Relation to Salmon Farms in the Bay of Fundy - *Emily Nelson, St. Andrews Biological Station*
1:40 Salmon migration: a key process for understanding lice infection in wild salmon - *Marc Trudel, St. Andrews Biological Station*
2:05 Salmosan – Committed to Sea Lice Control - *Jason Collins, Fish Vet Group*
2:30 Sea lice 2016: Trends to Inform Management Decisions in New Brunswick - *Larry Hammell, Atlantic Veterinary College*

3:00 **Refreshment Break**

3:20 Risk Factors for Treatment Failure in Antibiotic Treatments in Farmed Atlantic Salmon in Chile - *Derek Price, Atlantic Veterinary College*
3:45 An Update on the Epidemiology of Ulcer Disease - *Brett MacKinnon, Atlantic Veterinary College*

4:10 **Plenary Discussion -** What's next? What are the R&D priorities?

4:30 **Wrap up and Adjournment**

Presentation Synopses and Speaker Biographies

The speakers approved the following synopses.

Wednesday, October 26, 2016

DELIVERING HEALTHY, RESPONSIBLE, SUSTAINABLE GROWTH IN CANADA
CAIA'S NATIONAL STRATEGY: A REPORT CARD
- *presented by Ruth Salmon, Canadian Aquaculture Industry Alliance*

Demand for protein continues to rise such that by 2050, worldwide animal protein consumption will rise by nearly 73%. Aquaculture is leading the way in sustainable food production and global seafood demand is rising by 7% to 9% each year. Currently 50% of the seafood sold worldwide is farmed with the estimation that by 2030, this will increase to 62%. With the demand for seafood growing annually, Canada is well positioned to reassert its leadership.

The aquaculture industry has been ill-served by the lack of consistency in provincial and federal regulation, and the industry is not recognised in the Fisheries Act even though it is the lead regulatory legislation for the sector. CAIA's evidence and analysis has created a greater understanding of our industry, its current value and contribution, the barriers and challenges to growth, and solutions for moving forward. The new federal government's commitment to science and research is an indication of the growing support for responsible and sustainable aquaculture development in Canada.

The industry, through CAIA's approach to provide credible solutions, has also gained third party support on various fronts. The Conference Board of Canada Report "From Fin to Fork" and the Senate Committee Report on Aquaculture 2016 are examples. In the Economic Advisory Council report to Cabinet to be provided January 2017, the four main themes of Capital Infrastructure, Innovation, Labour Productivity and Trade were identified with a sector approach in the report that will highlight the aquaculture industry, among others.

On the political level, CAIA continues to work closely with the Fisheries and Ocean Canada Minister's office on all issues, presenting industry positions to all relevant Standing Committees (GM Salmon to Agriculture & AgriFood; TPP to International Trade) and continuing business risk-management discussions.

See Attached Presentation

<u>Ruth Salmon</u>
Ruth Salmon brings more than a decade of aquaculture experience to the Canadian Aquaculture Industry Alliance, having served five years as Executive Director of the BC Shellfish Growers Association and seven years as a private consultant. She has held senior positions with the Canadian agri-food industry – as General Manager of the Alberta Milk Producers Association and Advertising Manager with the Dairy Bureau of Canada. Having worked at both the provincial and national levels, Ruth takes a special interest in the promotion and expansion of the aquaculture industry across Canada.

STRATEGIC ALLIANCES AND COLLABORATION AS IMPORTANT COMMUNITY OUTREACH TOOLS
- *presented Sebastian Belle, Maine Aquaculture Association*

The presentation began with the suggestion that those of us in the aquaculture industry need to start thinking about the communities in which we work as our customers, followed by quote from Charles Darwin" In the long history of humankind those who learned to collaborate and improvise most effectively have prevailed." The quote was a reminder to make and maintain key relationships and strategic partnerships. Slides presented the results of various surveys that identified the key drivers for strategic partnerships, the types of partnership that would most benefit a company, the types of partnerships a CEO would rely on to increase market share and the priorities for most large

companies. Based on this information, the Maine Aquaculture Association (MAA) identified three key areas for strategic partnerships and collaboration – the coastal community, the tourism industry and the fishing industry.

With the relatively low cost of coastal land in Maine, many retired people and people from "away" moved into these communities increasing property values and taxes. These new community members had little connection to the commercial use of the ocean and different values and expectations with respect to how oceans and waterfronts should be used. Working waterfronts were often perceived as noisy, smelly and unsightly. As pressures on working waterfronts increased the MAA approached fisheries groups to work together and lobbied with them for tax incentives so fishing families could keep waterfront land through a working waterfront easement fund. The groups also worked together to produce information brochures for real estate agents showing these communities as working waterfronts and explaining to new residents what they could expect in their new communities.

The tourism industry in the various coastal communities in Maine were lobbying against aquaculture licence applications and requesting a moratorium as the farms were viewed as a liability not an asset to their tour operations. MAA took on this challenge with their members, offering the tourism industry tours of the various aquaculture operations and working with them to create tourism events to benefit both groups. Walking trail festivals were initiated with the farmers inviting tour groups to visit and taste their products, with materials made available by MAA for farmers to give out to visitors and other groups in the area. With events like the Permaquid Oyster Festival, Eastport Salmon and Seafood Festival and Maine Seaweed Festival now part of the Experience Maritime Maine promotion, aquaculture farms are now seen as an asset by the tourism industry and featured by the State agency.

Aquaculture is now viewed as part of the heritage of the area but this does not mean the work of collaboration stops or is easy to maintain. Another presentation of survey results identified ten key partnership / alliance challenges such as keeping the relationship active and mutually rewarding, building an ongoing win-win relationship, and allocating sufficient resources.

See Attached Presentation

Sebastian Belle

Sebastian Belle began his career as a commercial fisherman, working his way through university as a mate on offshore lobster boats. Currently Mr. Belle is the Executive Director of the Maine Aquaculture Association (MAA), a private non-profit association representing Maine shellfish and finfish growers. Prior to joining the Maine Aquaculture Association, Mr. Belle was the state aquaculture coordinator, working for the Maine Department of Marine Resources. In addition to his role as MAA Executive Director, Mr. Belle is president of Econ-Aqua, and a founding partner of TAAG. Econ-Aqua is consulting firm specializing in the farm management, financial due diligence and risk analysis and control. TAAG is an international consulting and investment firm specializing in aquaculture projects. Prior to founding TAAG, Mr. Belle was project manager of the Bluefin Tuna Project at the New England Aquarium in Boston. Before joining the aquarium, Mr. Belle was operations manager for Connors Aquaculture Inc. in Eastport, Maine, one of the largest Atlantic salmon farms in the United States. Mr. Belle holds degrees in fisheries biology and agricultural economics. Mr. Belle served as a technical consultant on over 20 major commercial aquaculture ventures for investment groups from Europe and North and South America. Before returning to North America in 1989, Mr. Belle spent four years managing a commercial scale aquaculture research and development foundation in Norway.

ARE YOU AN OPPONANT OR A COLLABORATOR?

- presented by Brad Hicks, Taplow Feeds

The development of salmonid aquaculture in North America has been significantly hampered by opposition from well-organized parties both from within government agencies and from well-funded civic society, primarily the environmental Non-Governmental Organizations (eNGO). The eNGOs working in consort with a manipulated, sympathetic press and their allies in government agencies are sculpturing public policies and regulations, which make it challenging to develop aquaculture in North America. These mislead policies and regulations and increase the cost of production in North America, making salmon farming in North America less competitive than in many other regions of the world.

These same anti-fish-farming individuals and groups are also trying to force aquaculture to develop in less desirable coastal locations. By forcing these limitations on aquaculture, the innovation and growth of the industry has been restrained. The restraining of the industry is not an unintended consequence of the desire by eNGOs to "improve" the industry but a concerned and directed effort to stifle the industry's growth.

The methods used to influence the press, the public and the government to develop public policies hostile to the development of aquaculture are primarily based on misinformation campaigns masquerading as science. These organizations publish reports in "scientific" journals which support their anti-aquaculture hypothesis. Then they use this advocacy "science" to push their agendas. They "*spew for the torrents of error"* and "*ignore abjections raised by [their] opponents"* [1] resulting in government and the public being hood-winked into thinking there is something inherently wrong with aquaculture. It then becomes easy for government to implement restrictive policy, which control the growth and development of this demonized industry.

Motivation for this behavior of the eNGOs is driven primarily by self-preservation. The eNGOs require an income. In several regions in North America the anti-fish- farming campaigns have been one of the main generators of cash for these organizations. But, their "sky is falling" rhetoric is morphing into silence as none of their cataclysmic predictions based on misinformation and deceit have come to fruition. Their influence is diminishing. Within one generation the fish farming industry will become a normal part of the economic and social fabric of coastal communities much in the way terrestrial farming is the main and most accepted activity on much of the land base.

1. The Unbearable asymmetry of bullshit, Brian Earp, Quillette, Feb. 18, 2016 http://quillette.com/2016/02/15/the-unbearable-asymmetry-of-bullshit/

See Attached Presentation

Brad Hicks

Dr. Hicks holds degrees in Fisheries and Wildlife Biology, Veterinary Medicine and Veterinary Pathology. He has been involved in the aquaculture industry in Canada and Internationally for the past 40 years. He began his professional career as a veterinary pathologist specializing in fish diseases. Later he became involved in several operating companies raising, salmon, trout, tilapia, sablefish and striped bass in both North and South America. Brad has published many articles in both peer reviewed academic journals and trade journals. Brad recently migrated into the supply side of the industry as a partner in a private feed company in BC. In addition to these revenue-generating activities, Brad has been active as an advisor and a member of several boards. He has been an advisor to the National Research Council, the Department of Fisheries and Oceans and the Ministry of Agriculture Food and Fisheries BC and Chairman of the Board of AquaNet one of the federal Networks of Centres of Excellence. He is a Director of the BC Salmon Farmers Association, a founder and Director of the Pacific Organic Seafood Association and a founder and Chairman of the Canadian Organic Aquaculture Producers. Brad chaired the Canadian "Organic Aquaculture Working Group" which drafted the "National Organic Standard for Aquaculture". Brad has also been involved many community organizations.

COLLABORATIVE IBOF WILD SALMON RECOVERY PROGRAM

- presented by Corey Clarke, Fundy National Park

Parks Canada has the responsibility to protect and represent 46 parks on behalf of Canadians. Part of the project presented focused on restoring ecosystem integrity, specifically for inner Bay of Fundy or iBoF Atlantic salmon in Fundy National Park. Without restoration efforts, it is estimated that IBoF salmon would have been extirpated from Fundy National Park by 2010. The iBoF salmon are federally listed as endangered and marine survival is thought to be currently limiting the return of spawning adults. Upon Species at Risk Act (SARA) listing in 2003, action to collect the remnant migrating smolts was undertaken. In partnership with the Department of Fisheries and Oceans' the Live Gene Bank (LGB) program Fundy National Park had been producing migrating smolts using various release strategies including releasing hatchery reared mature adults to spawn naturally and produce smolt, or releasing fry or parr from hatchery spawned adults to grow to smolt and leave the river. To improve chances of population recovery resulting from restoration efforts, managers needed to know which smolts performed best and had best potential for survival in the wild.

With the local salmon aquaculture industry, growing millions of smolts near the mouth of the Bay of Fundy a few hundred kilometers from Fundy National Park, a collaborative project started in 2009 to evaluate the smolt being produced through the LGB program. As part of a Master's project, wild captured smolt, either from a previous parr or fry release, were placed on farm sites to grow out to adult. The adults were then released into the Bay of Fundy or native river to be monitored during the spawning period. The adults released in 2011 survived to result in a 20-yr high number of adult returns in 2012. This initial research project indicated that smolts from fry releases migrated as bigger, older smolts, and made bigger adults with better surviving offspring. Seeing that less captivity resulted in more wild fitness, FNP re-focused its program on releasing mature salmon with minimal captive exposure to produce offspring with no captive exposure. Again, looking at the massive aquaculture industry capacity to produce adults in numbers not possible with a conservation hatchery plus value of marine exposure, the partnership was reinvigorated and the collaboration effort expanded to include First Nations, academic researchers, regulators and Conservation and Enforcement (C&E) officers of various stripes.

With the support of the New Brunswick Department of Agriculture, Aquaculture and Fisheries the project partners have created the world's 1st endangered salmon marine conservation farm, on a farm leased from the Grand Manan Village Council and operated by Kelly Cove Salmon / Cooke Aquaculture in Dark Harbour, Grand Manan. Named "Fundy Salmon Recovery (www.fundysalmonrecovery.com)" in 2016, the project has won multiple awards for innovative collaboration and has been the subject of a growing number of high profile media products including national TV network documentaries and a recent feature in Canadian Geographic. Pioneering wild fish culture techniques in industrial settings, innovative new partnerships and research, and unique opportunities for Canadians to connect with wild salmon conservation demonstrate that Fundy Salmon Recovery is producing more than wild salmon. It is producing new solutions, engaging community and changing discussion on collaborative conservation efforts.

See Attached Presentation

Corey Clarke

Corey leads Fundy National Park's salmon recovery program and has worked on IBoF Salmon recovery for Parks Canada in Fundy for 15 years. During his MSc program, he developed an award-winning project in collaboration with Parks Canada and the Aquaculture Industry. The project grew wild Fundy National Park smolts to maturity in marine pens, which later contributed to 20 year-high salmon observations in Fundy National Park Rivers. The findings of this project generated interest from additional groups, first nations and academics concerned with restoring wild salmon populations and a larger collaborative program is now underway. The current initiative has expended beyond Fundy National Park as partners with Fort Folly First Nation have began releasing marine-reared adult IBoF salmon into the Petitcodiac River system, the largest river of the IBoF population range.

WHAT GOES AROUND COMES AROUND, RESTORING THE UPPER SALMON RIVER

– Presented by Kurt Samways, Canadian Rivers Institute / University of New Brunswick

The science project was described as an option for researchers and students only because of the unique and special partnerships that exist as part of the Fundy Salmon Recovery program. Mature Atlantic salmon that have been raised from smolt on an aquaculture marine conservation farm are being returned to their natal rivers each Fall to spawn. The portion of the overall project overseen by the Canadian Rivers Institute is related to the monitoring of these salmon and the river systems they are being returned to ask: Are they behaving as expected and how is the river being impacted by the large numbers of salmon being released? Marine derived nutrients (MDN) such as nitrogen, phosphorous and fatty acids are deposited in the river with the return of Pacific salmon to spawn and die but little is known about the contribution of Atlantic salmon.

In the example of the Miramichi River, lost productivity is linked to the loss of the salmon population. With the historic high landings of over 3000 metric tons in 1620 it can be estimated that those salmon introduced over 100 tons of nitrogen and over nine tons phosphorous. With today's salmon population, this estimate is approximately seven tons of nitrogen and 0.6 tons of phosphorous.

The experimental design was described indicating how the question "How does the freshwater community respond to adult supplementation?" This will be answered in relation to sources of nutrients, changes in primary productivity and

behaviour of cage-reared adult salmon. Using stable isotopes of carbon and nitrogen as ecological tracers the source of nutrients was evaluated, and changes in primary production were evaluated through measurements of chlorophyll. The comparison was made between sections in the Upper Salmon River (USR) that are accessible salmon habitat or have barriers to the migration of the salmon released; as well as a comparison between similar sections of the USR and the Point Wolfe River (PWR) where there are no salmon releases. Results from both experiments indicated that the released salmon are responsible for increasing the level of MDN in the system and for overall productivity increases but with the small number of salmon released in 2015 it is too early to make any other conclusive statements.

To provide data on spawning behaviour, the adult salmon were monitored using Passive Integrated Transponder (PIT) telemetry tags, radio tags and DIDSON Sonar. Information provided from the PIT tag data to date showed there were 218 different fish detected (210 from 2016; 8 from 2015) and seven of the 2015 returning fish were still in the river. Data from the radio tagged fish showed 22 in Black Hole, a known spawning area, and one of the 39 fish detected was released in 2015.

The DIDSON (Dual frequency IDentification SONar) was deployed to identify and count returning Atlantic salmon. The DIDSON is an "Acoustic Camera" and like a medical ultrasound sonogram, it transmits sound pulses and converts the returning echoes into digital images. This allows researchers to "see" what is entering the river even in the dark and/or in zero visibility conditions. The DIDSON can distinguish between fish that are swimming side-by-side or head-to-tail and determine which direction fish are swimming. The DIDSON is also confirming that the majority of fish are retained in the system and the mature adults are migrating upriver.

See Attached Presentation

Kurt Samways

Kurt is from Saskatchewan where he received his undergraduate and master's degree at the University of Regina. His MSc focused on brook trout and habitat use throughout Quebec. After obtaining his MSc, Kurt continued east where he is completing a PhD with Rick Cunjak at UNB studying the interactions between anadromous fish and the freshwater environment. Kurt is now starting a post-doctoral project in collaboration with Fundy National Park and their adult release program.

IMPACT OF SALMON AQUACULTURE ON THE DIVERSITY AND HEALTH OF BENTHIC COMMUNITIES IN SHALLOW COASTAL HABITATS OF THE BAY OF FUNDY

– *presented by Heather Hunt, University of New Brunswick*

The coastal ecosystem in the Bay of Fundy is known for its high biodiversity, as well as its salmon aquaculture and fishing industries. Cobble habitat, which is important for many marine species including juvenile lobster, is scarce within Bay of Fundy and sometimes aquaculture sites are located near these sites. To address stakeholder concerns, the objective of this project is to quantify effects (positive and negative) of salmon aquaculture on diversity and health of benthic communities in shallow cobble habitat (5m to 10m).

The study focuses on eight site pairs with one of the pair near (~240m) and one away (~ 1200m) from a salmon aquaculture site plus reference sites (~8000m), in each of 3 aquaculture bay management areas. Trap surveys were conducted for adult lobster and bio-collectors were used to assess biodiversity of invertebrates in general and fish. Location maps, graphs showing distance of bio-collectors from aquaculture sites, and a mean daily temperature graph for all sites between July and December 2015 were discussed to provide context for the results to be presented.

Data for the 2014-2016 lobster trap surveys were presented by year separated by BMA indicating which areas had smolt, second year fish or had farms that were being fallowed. In all years, adult lobsters were caught at all study sites. Overall, the adult lobster trap data showed differences between some pairs of sites at some locations, but not all, and whether differences were significant depended on the sampling week. When a difference was found between the near and away sites of a pair, generally more lobster were found at locations away from the cage site. It was noted in discussion that there were differences in bait type used by the fishermen carrying out the sampling in different BMAs.

Bio-collectors (made of lobster trap wire mesh and filled with cobbles) are used to assess diversity and abundance, as well as exposure to chemicals and nutrients from aquaculture (metals and stable isotopes) but work on metals and

stable isotopes is just starting so initial results will not be available until 2017. The collectors are set from July to November and samples are processed in a lab to identify, count (with subsampling for small invertebrates), and measure organisms. Bio-collectors were first used at these sites in 2015.

Preliminary data for decapod crustaceans (lobster, crabs, shrimp), fish, and encrusting invertebrates in the bio-collectors were presented. In 2015, there were juvenile lobster, along with many species of crabs, shrimp and various marine fish in the bio-collectors, along with many smaller invertebrates that we are still in the process of identifying. There were no settling lobster in bio-collectors in BMA 1 but a good set in BMA 3a. It was noted that lobster settlement occurs in very specific areas and is known to be spatially patchy. There was no strong pattern with respect to the numbers of settling lobster found near or away from a farm site. When all of the decapod crustacean and fish data was analyzed together there were significant biodiversity differences between BMAs but it is recognized that there are many spatial factors that would explain these differences. At some locations, there was a significant difference in the community of organisms in bio-collectors between the near and far site pair. More analysis needs to be done to find out if these differences are consistent across site pairs or a result of spatial differences between the sites within some pairs. The samples from the bio-collectors are currently been examined to look at the smaller species of invertebrates. Research using bio-collectors in the Bay of Fundy from 2009-2015 has identified over 500 species in 14 Phyla of animals.

Current and future work in 2016 and 2017 will include additional work with the bio-collectors. A stakeholder workshop will be planned for March 2018.

See Attached Presentation

Heather Hunt

Dr. Heather Hunt joined the University of New Brunswick Saint John in 2002 and is a Professor in the Department of Biological Sciences. She earned her BSc and PhD from Dalhousie University and then completed postdoctoral fellowships at Woods Hole Oceanographic Institution and Rutgers University in the USA. Dr. Hunt is a marine ecologist with >20 years of research experience on the ecology of coastal marine invertebrates, with a current research focus on patterns of marine biodiversity and human impacts on marine ecosystems. She is the PI on a recently funded Environment Canada grant examining the effects of salmon aquaculture on the diversity and health of shallow coastal habitats. She previously led a project on infaunal invertebrates in Saint John Harbour, which developed recommendations for long-term monitoring of the Harbour. Other recent research topics include effects of sediment acidification on soft-shell clams, development of cobble-filled collectors as a tool for monitoring biodiversity, effects of scallop dredging intensity on invertebrate communities, and shifts in species' distributions in response to climate change.

AQUACULTURE IN VIRTUAL REALITY: EXPLORING, EDUCATING, POPULARIZING

– *presented by Ekaterina Prasolova-Førland, University of Science and Technology Norway*

Aquaculture has evolved to become a multi-billion-dollar industry in Norway and worldwide. The introduction of modern and advanced technological equipment and higher environmental demands motivates exploration of new training methods for fish farmers. At the same time, the knowledge of the aquaculture industry among the general public is rather limited, which might often lead to misconceptions and prejudices. Therefore, there is a need for new approaches to get people, especially youth, interested in the industry in order to secure future growth. In this talk, we presented the experiences from the development and evaluation of an educational aquaculture simulator in Virtual Reality with Oculus Rift. The simulator allows the user to visit a virtual salmon fish farm, dive into a fish cage, swim among the salmon, feed the salmon or check out the anchoring of the cage. The presentation reported evaluation results at various venues, outlining directions for future work.

http://aquaculturenorthamerica.com/profiles/virtual-reality-technology-applied-to-salmon-farming/

https://www.youtube.com/watch?v=ho6A65PuUDM

Ekaterina Prasolova-Førland

Ekaterina Prasolova-Forland is an Associate Professor at the Department of Education and Lifelong Learning, Norwegian University of Science and Technology. She received her Master's degree in Technical Cybernetics in 2000 and PhD in Computer Science in 2004 from the same university. Her research interests include innovative technologies for learning, Virtual Reality, educational and social aspects of 3D virtual worlds, educational simulations and serious games. She is author and co-author of more than 90 publications and has served on numerous program and organizing committees of scientific conferences. In 2011-2012 Dr. Prasolova-Førland participated in developing a game-based simulation for pre-deployment cultural awareness training for the Norwegian Armed Forces. She has been involved in several EU-financed projects. Dr. Prasolova-Førland is currently working on a few projects on educational applications of Virtual Reality in medicine, aquaculture, maritime industry, sports, tourism, emergency management and other areas.

AQUACULTURE DEVELOPMENT AND PROFITABLE COMMERCIALIZATION OF ARCTIC CHAR IN CANADA

– *presented by Rodrigue Yossa, Costal Zone Research Institute*

The Coastal Zones Research Institute Inc (CZRI) located in Shippagan, New Brunswick was created in 2005 and currently employs approximately 45 people. CZRI has four divisions: Laboratories and analysis services; Soil, Peatlands and Sustainable Development; Fisheries and Marine Co-products, and Aquaculture. The Aquaculture Division is under the leadership of Dr. Rodrigue Yossa.

Among the reasons for pursuing the development of an Arctic char industry are the fish's preference for cold water (6°-14°C), its toleration of high-density culture conditions, the existence of three domesticated strains already in Canada, and the fact that finished product is amenable to niche marketing. There are few complete or consistent statistics on the size of the Arctic charr industry globally, but the most recent data (2013) indicated that U.S. and Canada produced 507 MT while Iceland produced 3393 MT.

The overall objective of the Arctic char project, which began in 2014 in Canada under the leadership of CZRI, is to sustainably develop Arctic char aquaculture in Canada through collaborative efforts between government agencies, scientists and producers. To date this list of collaborators includes eight universities and research institutes, eight government agencies and seven farming industry representatives. These partners are involved in the specific objectives of the project's ten activities. These activities include broodstock pedigree development, development of fast growing, late maturing and salinity tolerant strains, feed and feeding program development, disease prevention, and efforts to enhance productivity.

Arctic char aquaculture is gaining momentum abroad and so the project partners see the potential of Arctic char as a commercial species. Going forward, in addition to completing the scientific activities, the project partners hope to develop a sustainable market for Arctic charr eggs and possibly fry, spur industry interest in Arctic charr aquaculture, and improve the productivities in current Canadian farms. To realize these goals, several challenges need to be addressed including the need for consumer awareness and education on the product, the lack of an available commercial feed dedicated exclusively to Arctic char, and need for better communication among the industry partners, scientists, and business people in Canada and abroad.

See Attached Presentation

Rodrigue Yossa

Dr. Rodrigue Yossa is the Scientific Director of aquaculture at the Coastal Zones Research Institute Inc., New-Brunswick, Canada. Rodrigue has completed a Ph.D. in animal sciences at Université Laval, Canada, a M.Sc. in aquaculture at Ghent University, Belgium, and a B.Sc. in Forestry, Wildlife and Water Engineering at the University of Dschang, Cameroon. Rodrigue is familiar with aquaculture research and development in Africa, Europe, Asia and the Americas. Rodrigue's research activities have received several awards, including the Younger Scientist Award at the 14th International Symposium on Fish Nutrition and Feeding in Qingdao (China), in 2010, and the Best Student Abstract Award at the World Aquaculture Society conference in Natal (Brazil), in 2011. After having studied, lived and worked in four continents, Rodrigue and his family now enjoy life in beautiful Acadian Peninsula, in Atlantic Canada.

GENOMIC TOOLS IDENTIFY THE DIRECT GENETIC IMPACTS OF ESCAPED FARMED ATLANTIC SALMON ON WILD POPULATIONS IN SOUTHERN NEWFOUNDLAND

- presented by Ian Bradbury, DFO-NL

Salmon aquaculture is expanding globally, and while industry Standard Operating Procedures (SOPs) for fish handling are in place and staff are trained, containment breach events can be a result of human error during operations at the farms such as harvesting. With these escapes comes the potential for genetic and ecological interactions with wild salmon populations. Interactions have been examined to some degree in Europe but this is the first research project to evaluate interactions in Atlantic Canada, using rivers in Newfoundland as the case study.

In Newfoundland (NL), there was a single large escape event that occurred in 2013, which released approximately 20 thousand farmed salmon into Bay d'Espoir and Fortune Bay. Since the entire south coast of Newfoundland is predicted to have only 20 thousand wild salmon the potential for impact from this event seemed high but the likelihood or extent of potential interactions was largely unknown. To study these interactions genetic and genomic approaches were applied to quantify the levels of interbreeding among the escapes and wild salmon. The genomic tools, or markers, had to be developed and in this case, SNPs or single nucleotide polymorphisms were used to identify escapes and hybrids. These markers first applied to wild and aquaculture salmon established baselines then used to measure levels of hybridization and introgression. The data collected can be used to model the interactions at the population level to better understand impacts to salmon populations.

The work included conducting high-resolution genome-wide genome scans using 5.6 and 220 thousand single nucleotide polymorphism arrays. From this data, SNPs were selected that allowed for accurate differentiation of farmed and wild salmon, and identification of hybrids. Once selected and verified, the panels of diagnostic SNPs were developed into genomic tools that could be rapidly and efficiently applied.

Genetic differentiation between farmed and wild salmon exists due to the combination of their different geographical origins, domestication selection and unintentional domestication effects, so the population lines differ at both neutral and adaptive regions of the salmon genome. By targeting regions of the genome that are the most highly divergent between wild and farmed fish, the test could maximize the resolution power.

Simulations showed the diagnostic panels provided accuracy in identifying both wild, farmed, and hybrids in Newfoundland. These results were validated independently using lab-made F1 hybrids and showed 100% accuracy in wild, farm and hybrid identification. Based on theses simulations and independent validation, in-field application to measure levels of hybridization was warranted.

In 2014, 2000 young of year Atlantic salmon collected at 18 river locations throughout Bay d'Espoir and Fortune Bay to screen for hybrid ancestry. The data provided evidence of extensive hybridization among wild and farmed salmon following the 2013 escape event with hybrids detected in 17 of the 18 rivers surveyed. Overall, about a third of the juveniles sampled were of hybrid ancestry. This is the first documented case of interbreeding among escapes and wild salmon in Atlantic Canada. In addition, the occurrence of farm-to-farm reproduction was demonstrated, resulting in the creation of farmed salmon offspring in the wild.

These observed hybrids can be separated into classes, first (F1) and second generation (F2) hybrids, and back crosses. There was strong evidence for later generation hybrids such as F2 suggesting that hybridization predates the 2013 escape event. The existence of later generation hybrids also indicated that a portion of the farmed-wild hybrids are viable and that long term genetic effects are possible. The proportion of wild juveniles found increases with distance from the release location and the proportion of hybrids decreases with distance. Both results are consistent with the release location as the probable source and suggested impacts are highest close to the release location.

Large scheduled rivers / populations were compared to small non-scheduled rivers / populations in terms of the proportions of groups present in the juvenile samples. It was found that large scheduled river populations had more wild juveniles and less hybrids and aquaculture salmon, while small unscheduled rivers had more hybrids and aquaculture fry compared to wild fry.

In 2015 and 2016, approximately 1500 young of year and Atlantic salmon parr were collected for screening and results are pending.

The presence of genetic impacts regionally in NL and the Maritimes was evaluated using a series of large existing SNP datasets, in conjunction with data collected. These samples were all collected prior to 2013. For both regions, the level of genetic impact declines with distance away from the center of production so significant genetic change was detected up to scales of 100's of km, and was largely absent beyond that. In the Bay of Fundy, samples showed little evidence of introgression beyond 100-500 km with the amount of introgression similar across regions (15-30% at fine scales).

Future questions to be answered include "so what?" At the individual level, how do these hybrids perform in the wild, and how do they grow, behave and survive in the freshwater and marine environments? Will there be an impact on population productivity? Estimates based on these 2013 samples from lower stretches in these specific rivers of Newfoundland may not be representative of all watersheds and importantly there is also annual variation that will need to be considered.

See Attached Presentation

Ian Bradbury

Dr. Bradbury is a research scientist with the Department of Fisheries and Oceans and is the Cox Fisheries Scientist in residence at Dalhousie University. Originally from Newfoundland, he completed his PhD in 2007 at Dalhousie University and started with DFO in 2010. His research uses genomic tools to inform the conservation and management of both marine and anadromous species from throughout Atlantic Canada. Specific work focuses on identifying the genomic basis of marine climate associated adaptation, developing genetic baselines for individual identification in multiple species, and quantifying the impacts of escaped farmed salmon on wild populations.

GENETIC TRACEABILITY

– *presented by Amber Garber, Huntsman Marine Science Centre*

Traceability is described as the ability to identify individuals over time and is often discussed in relation to a grocery store and / or consumer having the ability to view the life history of a product (e.g., a fillet when it was part of a fish, how / where it was caught). This presentation discussed genetic traceability meaning the ability to identify cultured Atlantic salmon, found in the wild, to a sea cage, hatchery, company or, most detailed, a family group.

Sampling farmed and wild salmon for later genetic evaluation is relatively easy and minimally invasive. There are various types of technologies that can be used to identify individual fish from a common group or to a family using samples collected. However, a technology using DNA from tissues such as fin clips is the easiest and least invasive. There are two types of DNA markers that are presently being used in Atlantic Canada to identify groups and families – microsatellite markers (short tandem repeats or STRs) and single nucleotide polymorphic markers (SNPs). Atlantic salmon DNA is like a human in the sense that one strand of our DNA comes from each parent. When combined each pair of alleles inherited make up each locus or marker.

There have been two methods or processes used to trace collected samples. The DNA Stand By Method identifies the origin of escaped farmed salmon when a large group of cultured individuals are identified in wild. DNA samples are collected from farms/sea cages nearby with similar sized individuals and an estimate of probability that the escaped salmon are from sampled farms (genetics + statistics) is generated.

The DNA Registry method of traceability traces cultured salmon found in the wild back to individual families from a company or may be traced back to a sea cage then hatchery. Selective breeding programs using pedigreed broodstock are critical if discussing implementation of a DNA Register or Registry program as it provides the foundation to more easily allow for traceability (at the level of genotype) from gamete contributors (parents). This method is similar to the program implemented in Maine.

Depending on the breeding program followed there are challenges to identification of individuals to specific families. Selective breeding programs using pedigreed broodstock are designed to produce fish that retain genetic variation, minimize inbreeding and are improved for all desired traits while also tracking parentage (genetic distinction). However, multiplier groups or production groups may not be tracked and maintained by family within a production

year (parentage may be unknown). Therefore, sampling and/or tracking to the level of family (individual cross) could be an additional step for a company even when a pedigreed broodstock program is in place.

See Attached Presentation

Amber Garber

Dr. Amber Garber has been involved in aquaculture and wild fisheries studies since 1998. Her primary topics of research include breeding and broodstock development, development of molecular markers for aquaculture and wild fisheries, population genetics studies, and stock enhancement. She has applied her expertise to commercial production and stock enhancement broodstock programs to varying degrees for red snapper, hybrid striped bass, Atlantic cod, rainbow trout, brook trout and Atlantic salmon. Amber has also been involved with and led experiments in fish physiology, disease challenges, sea lice management and proprietary research. Amber moved from North Carolina State University (Raleigh, North Carolina, USA) to the Huntsman Marine Science Centre (St. Andrews, New Brunswick, Canada) in 2006 where she is a Research Scientist in Aquatic Services

GENOMIC SELECTION AND GENOME-WIDE ASSOCIATION STUDIES: PERSPECTIVES AND POSSIBILITIES

- presented by Tiago Hori, Center for Aquaculture Technologies Canada

In the competitive market of aquaculture, broodstock development is becoming increasingly important and aquaculture needs to move to same direction in breeding programs as in other production systems. Phenotypic selection (selection based on physical appearance / observable traits) can have limitations and may be lengthy and costly. The genes and other genetic elements that contribute to the observed variation can be identified using genomics and therefore used for selection. An example of a genetic marker is a single nucleotide polymorphism or SNP which can be different between individuals. SNPs can be responsible for or associated with characteristics like early maturation, food conversion and resistance to infectious pancreatic necrosis (IPN) disease. Since they are heritable, the use of SNPs enables pedigree reconstruction (parent assignment) for broodstock development and the use of marker assisted selection (MAS). MAS can significantly increase genetic gains over a generation reducing the time and cost to witness results.

The genome of the Atlantic salmon took ten years to characterize as it is very complicated but now that it is publicly available, it has opened the door to better marker-based selection processes. Genome wide selection (GWS) looks for the most similar individuals or groups based on their genetic makeup. SNPs occur in intervals in the genome with each interval potentially representing a section of the genome that can potentially affect a trait of interest. If enough SNPs can be identified, the impact of each interval on a given phenotype can be estimated which allows the calculation of a genetic estimated breeding value (GEBV). GEBV are often more accurate than the traditional EBV method and can be calculated in early life, which reduces time and cost. The genome wide information also allows for estimation of relatedness and that can be used to mitigate the potential negative impacts of inbreeding depression.

There are challenges with GWS in the amount of data to be analyzed, data gaps and cost, but advantages associated with the large genetic potential in aquaculture animals and for GWS to identify multiple and / or complex traits must be considered along with the increased importance of genetic traceability to the industry.

The Center for Aquaculture Technologies is continually working to makes these approaches available and this work includes the determination of the minimum number of markers needed to get a specific result required by industry, and increasing data analysis capacity.

See Attached Presentation

Tiago Hori

Dr. Tiago Hori obtained his Ph.D. from Memorial University of Newfoundland, where he worked on developing genomic resources for the Atlantic cod. During that time, he built and characterized both normalized and subtracted cDNA libraries and helped with the construction of 20,000 features Atlantic cod cDNA microarray. These resources have been since used in several studies looking at the physiology and immune response of Atlantic cod, which have

contributed to a better understanding of the biology of this commercially relevant species. Subsequently, he worked as post-doctoral fellow at Dr. Matthew Rise's laboratory at the Ocean Sciences Centre. During that time, Dr. Hori applied diverse functional genomics techniques such as cDNA libraries, microarrays, RNA-seq and QPCR to investigate the biology of salmonids. His work with salmonids included investigating the impacts of triploidization, using functional genomics to look into environmental impacts on the brain transcriptome and applying RNA-seq to study differential growth between families of Atlantic salmon. Dr. Hori is currently the Associate Director of genomics at the Center for Aquaculture Technologies Canada (CATC), where he leads efforts into further developing the genomic tools available for commercially-important fish and their application in the Aquaculture industry. At CATC, he is working to develop cost-effective platforms for single nucleotide polymorphisms (SNPs) at low, medium and high densities, aiming to make genome wide association studies (GWAS), marker assisted selection (MAS) and genomic selection accessible to the aquaculture industry. Dr. Hori also continues his efforts in developing resources for non-model commercially relevant species, such as Atlantic sturgeon and Arctic Char, using high-throughput sequencing of SNP discovery and genotyping. Lastly, Dr. Hori in a contributing author in more than 25 peer-reviewed publications in journals such as BCM Genomics, Physiological Genomics, PLoS One and Developmental and comparative Immunology.

FUNCTIONAL TRANSCRIPTOMIC CHARACTERIZATION OF *LEPEOPHTHEIRUS SALMONIS* REJECTION BY COHO SALMON

- presented by Laura Braden, Atlantic Veterinary College

Challenges with sea lice infestation is and may continue to be an impediment to growth in the salmon aquaculture sector. The primary work focus for this research group is the host-parasite interaction between salmon and the salmon louse, specifically the mechanisms of host infection resistance. Atlantic salmon, the most susceptible host to this parasite, has a weak or absent inflammatory response and a delayed wound healing response during infection and is therefore susceptible to secondary infections and pathology associated with chronic louse infestation. Coho and pink salmon, at the other end of the spectrum, immediately respond to attachment site infection with aggressive inflammation, cellular infiltration, rapid wound repair, and rapid rejection of the parasite. The preliminary data presented described the effort to characterize the rapid rejection of lice by juvenile Coho salmon, as the mechanism involved is unknown.

The first objective was to determine if resistance is a function of life history - if resistance accompanies the switch to saltwater or if resistance increases with host size. Coho salmon smolt were exposed to saltwater over a period of 48hrs (short) or 30 days (long) then were challenged with approximately 60 L salmonis copepodids per fish over 2 hours with reduced water flow. At specific intervals starting at six hours and up to 18 days for the respective groups, fish from each study tank including controls were euthanized, lice counts completed and blood, kidney, spleen, fin, attachment sites (on infected fish) and gill samples taken. Irrespective of the time in saltwater, there was rapid rejection of lice accompanied by aggressive epithelial hyperplasia and inflammation resulting in complete parasite rejection by 10 days post-infection. This response was observed as early as 4 dpi, where attached larval L. salmonis were observed encapsulated by the hyperplastic response.

A comprehensive understanding of host-parasite interactions requires knowledge of the associated gene expression changes in both pathogen and host. Using the infected and non-infected fin samples taken from the thirty-day post smolt group, a dual RNA sequencing approach was utilized whereby the transcriptomes of both host and parasite were analyzed in the same sample from 1 to 18 days post infection. Many transcripts overexpressed by Coho were identified that had not been described before in this context, including IgE, which may indicate an allergic response within the Coho to the louse. The salmon louse transcriptome was characterized by upregulation of stress responses and chitin synthesis enzymes indicating an effect on the moulting response of the parasite.

Further analysis needs to be conducted to determine genes that are purely involved in developmental processes in the salmon louse, rather than in the stress/defense response. This dual RNA sequencing work will need to be repeated with one day post-smolt and the larger cohort (~ 60 gram) to explore expression of identified targets, and immunohistochemistry of encapsulation sites will be evaluated to identify cell populations involved this process. With immune system cells from Coho found inside the louse there is potential for a “Trojan horse” to be used by other salmon species. Identification of pathways involved in the rapid rejection by Coho salmon will provide invaluable

information that will enable development of novel control strategies against the salmon louse. For example, gene editing may be used to augment genes in Atlantic salmon so that they respond more similarly to a Coho salmon.

See Attached Presentation

Laura Braden

Laura Braden obtained her doctorate in March of this year at the University of Victoria, on Vancouver Island, BC. Her dissertation focused on determining molecular mechanisms for resistance among different species of salmon to the salmon louse. In April, she moved to PEI to take a post-doctoral fellowship with Dr. Mark Fast at the Atlantic Veterinary College. Her research at the AVC maintains a focus on host-parasite interactions. She is continuing her work from her PhD, looking at the molecular pathways and responses responsible for louse resistance in Coho salmon. Other projects include looking at the immunological responders involved during the host response to Loma salmonae (a microsporidian parasite) in rainbow trout, and to Kudoa thyrsites (a myxozoan parasite) in Atlantic salmon.

FEEDKIND PROTEIN: THE FUTURE OF AQUACULTURE FEEDS

– *presented by Dennis Leong, Calysta*

Calysta is involved in aquaculture feed production that does not involve the use of fish, animal or plant protein. This "Future Fit Feed" is produced using methane, including sustainable sources such as biogas, and is a technology that helps address climate change and food security concerns. Naturally occurring microorganisms metabolize methane as their sole source of carbon and energy, producing a nutritious, high-protein biomass. Calysta's single cell protein product is comparable to a super prime grade of fishmeal. The concept of using single cell protein in food applications is nothing novel – millions of people consume these proteins that can be produced from fungi, yeast and bacteria daily in a variety of products.

There is no agricultural land use with this technology; the process uses 77-98% less water than agricultural products and FeedKind® protein does not compete with the human food chain.

In the search for potential alternatives to fishmeal in aquaculture feeds, many are high in fiber, do not have the appropriate amino acid profile, or are difficult to scale up to produce meaningful volumes. A comparison of the nutritional profiles of various fishmeal alternatives was presented along with the amino acid and key nutrient profile of Feedkind®. Results from a study completed in cooperation with EWOS in Norway indicated the use of FeedKind Protein improves growth rates, nitrogen retention and gut health in Atlantic salmon.

Calysta's proprietary "marker" technology in the protein can be used to meet various supply chain traceability and certification program requirements.

Commercialization is underway in the UK and the groundbreaking for the first commercial plant in North America will take place later this year in collaboration with Cargill. The modular design lends itself to a phased construction process and the facility will ultimately produce 200,000 tonnes per year. Aquaculture producers will have the opportunity to test Feedkind protein in different species.

See Attached Presentation

Dennis Leong

Mr. Leong joined Calysta in 2015 and is the Vice President, Business Development. He has responsibility for all business development strategy and program execution, including broadening the Calysta portfolio of strategic partnerships. He joined Calysta from Chemtex, an international technology and engineering organization, where he was the Executive Vice President responsible for worldwide marketing and business development. At Chemtex he played an integral role in building the renewable and sustainable products business, including launching and licensing Beta Renewables' cellulosic ethanol technology. Before joining Chemtex in 2008, Mr. Leong led the marketing and business development activities for the downstream petrochemical and chemical business of Aker Solutions, a global engineering and construction contractor. Over his 20-year tenure with the group, he was based in a number of international locations including Canada, Japan, Hong Kong, Singapore and the United Kingdom. He holds a bachelor

of applied science degree in chemical engineering from the University of British Columbia and is a registered professional engineer in Canada.

DEVELOPMENT OF AN ALTERNATIVE SULFIDE DETECTION METHOD
- presented by David Wong, DFO-SABS

As organic material is consumed around aquaculture sites (uneaten feed and feces), oxygen levels decrease and sulphide levels increase such that after a certain point a reduction in the benthic community observed. Through the New Brunswick's' Environmental Monitoring Program (EMP), aquaculture sites are classified annually related to sediment sulfide concentrations found on the farm.

The traditional method for regulatory sulfide analysis has included sediment samples collected by grab or core and analysis of the sediment sample basified to pH > 12 and using a silver/sulfide ion selective electrode (ISE) to detect the sulfide ion. Various questions have been raised about this method. These include, but not limited to the instability and temperature dependence of the probe calibration, the measurement of sulfide in a sediment slurry, geochemistry differences of sediment types, the possible overestimation of sulfide due to the pH environment required for sulfide detection (Brown et al, 2011), and sediment sample storage were issues identified. Two ACRDP funded research projects were completed that evaluated and quantified these concerns, ultimately leading to work on alternative methods that would address the concerns identified with the traditional method.

A methylene blue colorimetric method was chosen as a possible alternative as it would use sediment porewater as a consistent sample matrix. The soluble sulfide present in the porewater would be "fixed" as an insoluble salt, and the ability to use 96-well microplates would allow the simultaneous analysis of many samples. To validate this method, several parameters had to be tested including its limit of quantification (LOQ), its accuracy and precision and sample storage stability. Work to compare the results of this potential testing method against the traditional ISE method also needed to be completed to ensure consistency for the regulatory program. The results of the validation work were presented along with the pros and cons of the methylene blue microplate method.

The initial cost to purchase a plate reader was the only potential con identified with this alternative method for sulphide analysis, although the time saving capacity to test a large number of samples within a short time frame (potentially up to 240 samples in three hours) would also have to be considered. In addition to eliminating many of the concerns identified with the current method, the work to date indicated that the methylene blue microplate method is comparable to the ISE method, calibration is stable and sample stability has been recorded up to 24 weeks.

See Attached Presentation

<u>David Wong</u>
David Wong is an Aquatic Science Technician for DFO in the Coastal Ecosystem Research Section at the St. Andrews Biological Station and has experience in developing and validating analytical methods in different types of matrices. He obtained his BSc in Applied Sciences and Post Graduate Diploma in Instrumental Analytical Sciences from the Robert Gordon University in Aberdeen, Scotland. He was a Study Director in the Metabolism Chemistry Department of an international contract research organisation in Scotland prior to moving to St. Andrews in 2006.

ISAv: WHAT IS A STRAIN?
- presented by Ben Forward, RPC Science and Engineering

A basic overview and anatomy of the causative agent of infectious salmon anaemia (ISA) was presented to begin the presentation. First detected in Norway in 1984, ISA outbreaks in New Brunswick started in late 90's and still occur today though the impact has been reduced due to improved management practices. The infectious salmon anaemia virus (ISAv) is an orthomyxovirus, which is an eight segment RNA virus encoding approximately ten proteins designed to aid the virus in taking over the host cell function. This process was presented pictorially.

The hemagglutinin esterase (HE) and fusion proteins on the surface of the virus communicates with the host cell, allowing the viral payload to be delivered inside the host and initiating the viral replication process. This HE protein will therefore influence ISAv virulence or the ability of the virus to invade the tissues of the host and the severity of the disease produced. Strain typing methods are employed to uniquely identify and distinguish different isolates found in each diagnostic case and to predict potential virulence. There are several strain-typing methods available though DNA sequencing of Segment 6 (HE) of the ISAv genome is currently favoured. This segment is a high polymorphic region (HPR) and deletions / deviations in this segment are used to determine strain type.

Through this strain-typing work over 40 different HPR types have been detected and sequenced since 2004. Nearly all those detected in the past five years have been new strains. Survival charts presented for three ISAv strains provided an example of virulence difference. With HPR4 infection, sick fish and mortalities will be seen within eight to ten days and by 30 days post infection, nearly 100% morality is experienced by both those salmon directly infected by injection and those infected through cohabitation. By day 40 of an experiment with HRP5 80% of the cohabitation fish will still be alive. The mortality rate seen with cohab fish in work with HRP2 between these other strains is approximately 60%.

Many factors influence the potential of ISA occurring in a group of fish at a specific location and time and there are many research questions yet to be answered about ISAv. These questions include:

- What precipitates the transition from avirulent (HRP0) to virulent (HPR deletion) – are natural reservoirs involved?
- How does the HRP0 strain of ISAv replicate in the host but not cause disease?
- What is the role of HE (Seg6) deletions in virulence regarding interaction with fusion protein (Seg5)?
- What are the relative contributions of fusion protein (Seg5) mutations to virulence?
- What is the relationship of infectivity/virulence to pathogenesis (development of disease: acute, chronic, or recurrent)?

See Attached Presentation

<u>Ben Forward</u>
Dr. Forward is Head of the Food, Fisheries, & Aquaculture department at the New Brunswick Research & Productivity Council (RPC), in Fredericton, NB, Canada. He holds a PhD in Biochemistry from the University of Victoria and a BSc with honors in Biology from the University of New Brunswick. As Department Head, he oversees three divisions providing R&D and diagnostic services in the areas of Fish Health, Microbiology, and Forensic Biology and has served as project lead on numerous applied molecular and microbiological R&D projects. He is an adjunct professor at UNBSJ, member of the Canadian Society of Forensic Science, Society for Wildlife Forensic Science, and Aquaculture Association of Canada.

SIGNIFICANCE OF HPR0 IN RELATION TO ISA DISEASE CAUSED BY HPR-DELETED ISA VARIANTS
- presented by Knut Falk, Norwegian Veterinary Institute

The World Organization for Animal Health (OIE) is an instrument for the World trade organization (WTO). The OIE maintains lists of animal, fish, and shellfish diseases that may compromise commercial activities or wild animal stocks

and it publishes a number guidelines related to disease control, including procedures for detection and diagnosis. Both the virulent HPR-deleted types, and the non-virulent HPR0 type are listed by the OIE and finding of the virus must be reported. Virulence is the capacity of a microbe to cause disease and both types of virus are found worldwide.

In Norway, there are annual small localized outbreaks of virulent HPR-deleted ISA virus with recent cases transferred by infected smolt. In eastern Canada and Chile, ISA is considered endemic; Scotland and the Faroe Islands are ISA free in principle; and western Canada, Ireland, and Tasmania have had no officially reported ISA virus detections.

Extensive PCR screening on the Faroe Islands revealed non-virulent HPR0 virus causes a transient / passing infection mostly localized to gills. Information from Norway, Scotland, Chile and eastern Canada suggest comparable prevalence in gills. HPR0 ISA virus has not been found in western Canada despite testing thousands of fish.

The hypothesis is virulent HPR-deleted ISA virus develops from non-virulent HPR0 virus. This possibility leads to various questions and challenges from a regulatory point of view:

- How often does this transition occur?
- What is the driver(s) for this transition?
- What is the risk of this transition when HPR0 ISA virus is detected?
- Are there other necessary changes needed to get a fully virulent virus?
- Is the transition a step-by-step process which include low-virulent intermediates?

The virus causing ISA has a shortened HE stalk (HPR-deleted) and a small change in the fusion protein relative to the original non-virulent HPR0 type. These changes are key factors for ISA virus virulence and disease characteristics, and together modify viral fusion activation and activity. A Faroese example representing the first field evidence of direct transition from a non-virulent HPR0 to a virulent HPR-deleted ISA virus was presented.

In this example, routine PCR screening at harvest detected a very small number of fish in one net pen with low level HRP-deleted ISA virus. There was no elevated mortality or clinical signs suggesting ISA. The next month PCR screening of 150 fish within one pen revealed 90% were ISA virus positive with higher viral levels but again there was no elevated mortality. A re-examination including sequencing of samples from the originating smolt farm revealed a very closely related HPR0 virus. The HPR0 and HPR-deleted virus could not be found in screening samples collected during seawater production in the affected farms or neighboring farms. The whole genome of both the new HPR-deleted and the related HPR0 virus were sequenced with the only difference found between them being the deletion in the HPR-region of the HE-gene and a single amino acid mutation in the F-gene.

The results provided practical support for the HPR0 hypothesis and demonstrated that deletions in the HPR-region of the HE-gene, combined with a mutation in the F-gene are the minimum requirements for a shift from a localized to a generalized infection. Transition from non-virulent HPR0 virus to a fully virulent HPR-deleted virus is suggested to be a stepwise process requiring more unknown changes to the virus involving low virulent intermediates that may be difficult to detect. Transition may have occurred late in the seawater production phase with potential stress episodes including peroxide treatment, heavy storms and ulcers. Prevention strategies suggested included all in / all out production, year class separation and good biosecurity.

See Attached Presentation

Knut Falk

Dr. Knut Falk is senior researcher at the Norwegian Veterinary Institute (NVI), which is a governmental research institute. He is also a designated World Organization for Animal Health (OIE) expert on ISA, and responsible for the OIE reference laboratory function at NVI. Knut has a DVM degree and PhD from the Norwegian School of Veterinary Science, and has worked with viral fish diseases in farmed Atlantic salmon, in particular infectious salmon anaemia (ISA), for the last 25 years. Knut's work has included first isolation of ISA virus including major contribution to characterization of the virus as well as the disease. He has been involved in both diagnostics, pathology, epidemiology and disease control related to ISA. Knut's current research is now focused on revealing pathogenetic mechanisms for ISA associated with the ISAV HPR0 type including evaluation of risk factors for this non-disease causing virus to develop into virulent disease-causing ISA.

ISA IN THE ATLANTIC REGION - NEW BRUNSWICK
- presented by Michael Beattie, NB Department of Agriculture, Aquaculture and Fisheries

New Brunswick has an active surveillance program for ISAv, which involves NB DAAF fish health personnel monitoring sites every five to six weeks in addition to the private / company veterinarians. Testing includes both IFAT and Rt-PCR with a sample held for virology if initial testing indicates the virus is present. If the HRP-deleted virus is found, the frequency of site visits increases for approximately six weeks. No cases of ISA were found in New Brunswick for 7.5 years although the HPR0 strain was reported during that time. A case is defined as two fish being found positive by two tests. However, industry has proactively harvest within four days if one fish tests positive.

Informational charts presented New Brunswick data for 2014 to 2016 that indicated the number of sites and BMAs involved in ISA cases and the strains identified. To date 36 of the 44 strains of ISAv identified have been found here with a shift to more European virulent strains being found than North American strains. From 2003 to 2015, 99% of the virulent strains were North American but in 2015/2016 this dropped to 80%.

The HRP0 strain most commonly found since 2003 is European (85%) but observations indicated that the equivalent percentage of virulent strains are not being found. In the cases from 2014-2016 where HPR0 was initially identified on 16 sites, additional sampling conducted eventually identified ISAv on only 5 of these sites. These strains were 69% North American.

Results of the screening program have shown that if HPR0 is present the IFAT will be negative and Rt-PCR will be positive 99% of the time. During a case of HPR 2a ISAv, the screening showed -IFAT and +Rt-PCR and there were no mortalities. This was found to indicate the need for the research community to work towards using the same nomenclature regarding these viral strains identified as European or North American.

Several observations and questions about ISAv were presented which will need further research to answer. Are there wild reservoir populations of ISAv? Juvenile herring and hake were seen in salmon cages prior to 2015 and 2016 ISA cases. Are HPR0 strain mutations leading to virulent forms? The New Brunswick observations of European versus North American strain types doesn't seem to support this hypothesis. Work also needs to be completed to improve ISA vaccine efficacy. Does it need to be strain type specific? Is a new adjuvant needed? Does a more stable region of segment 6 need to be chosen for DNA vaccine development and / or does segment 5 need to be added to vaccine?

See Attached Presentation

Michael Beattie
Michael Beattie is the NB DAAF Veterinarian. Michael received a BSc, (hon.) and MSc. in marine biology from the University of New Brunswick, a DVM degree from the AVC and a Marketing certification from the Norwegian School of Business. In 1997, he became a member of the Royal College of Veterinary Surgeons. Since 2003, he has served as the Chief Veterinarian for Aquaculture in the New Brunswick Department of Agriculture, Aquaculture and Fisheries. Prior to joining the Provincial government Mike was the North American Product Manager for the world's largest integrated aquaculture company, Nutreco. He was involved in uncovering new research, carrying out field trials and marketing new products.

ISA IN THE ATLANTIC REGION - NOVA SCOTIA
- presented by Roland Cusack, NS Department of Fisheries and Aquaculture

Surveillance for ISAv has been a 20 year, collaborative effort with provincial governments, industry, RPC, lab personnel and DFO/CFIA; so, it has been a team effort.

Initial detection of ISAv in Nova Scotia was not a clinical case. The virus was found in 1998 during routine broodstock screening by viral culture. It was the first finding of HPR0 in North America, the "European" strain, and no ISA was detected in progeny. The first clinical cases occurred within the same bay in 2000 and again in 2003; the viral strain was identified as HPR 3 during both occurrences. The next clinical cases did not present until 2012, after nine years of surveillance. In the spring, there was a non-clinical case in a freshwater land based facility, the first

HPR-0 NA strain ever detected. Following this event, the summer and winter of 2012 saw virulent ISAv in two locations. There were links between the sites through fishing gear and oceanography but the strains were different. The question of a reservoir for ISAv was raised. There have been no new cases of ISA since the 2012 episodes.

Nova Scotia has both an active and passive ISA surveillance programs. Marine sites are visited every four to eight weeks, and freshwater facilities are visited three times per year. Testing for ISAv is primarily done by RT-PCR, then virology and IFAT as a backup test, with histopathology completed where lesions are present. An ISA case is defined by two fish being positive by one test. Passive surveillance is through active investigation of elevated mortalities.

A new regulatory structure implemented in 2015 has increased provincial oversight for ISA monitoring and control. Farm Management Plans are required for each operational marine farm that outlines preventative measures including biosecurity program, disease surveillance plans and demonstrates emergency preparedness. A Certificate of Health for Transfer is also now required prior to any stock movement.

In terms of case management, under the Aquaculture Management Regulations the Province directs all activities including quarantine, removal and disposal of fish, fallow periods to be followed post removal, and pre-stocking testing.

Roland Cusack

Dr. Cusack graduated from St. Francis Xavier University with a degree in biology, a Masters of Science from Dalhousie in fish parasitology and a Doctor of Veterinary Medicine from the Atlantic Veterinary College. He began working on the use of thermal effluents for aquaculture in 1980 and continued working in aquaculture and fisheries topics through to 1991. At that time, he joined the Nova Scotia Department of Fisheries and Aquaculture as the clinical aquaculture veterinarian serving Nova Scotia fish farmers. He is currently the Chief Aquatic Animal Health Veterinarian for the Department

ACTIVE ISAV SURVEILLANCE

- *presented by Nicole O'Brien, NL Department of Fisheries and Land Resources*

Active ISAv surveillance data was analyzed over a 1.5 year history. The surveillance program was to conduct inspections on a minimum of 5 moribund fish on every site on a monthly basis. Stochastic model simulation was used to evaluate the surveillance program. The results of the model show that for a new site, it takes 4 months to demonstrate freedom from disease using this surveillance program. After this 4 month period, the confidence of freedom from disease will remain high unless inspections do not occur or a disease event happens. Using this information, risk-based surveillance at a regional level could be considered. If certain fish life-stages are considered higher risk, sampling frequency could be increased to gain a level of 95% confidence much quicker. Furthermore, if the consequence risk of a positive finding in a region is considered high, the area could be sampled more frequently so that the region can be declared free of the disease sooner. This regional approach may include sampling all sites within the region or rotating through the sites. In conclusion, risk-based surveillance can be utilized to demonstrate freedom from disease depending on the situation.

Nicole O'Brien

Dr. Nicole O'Brien (DVM, PhD) is a licensed aquaculture veterinarian and Veterinary epidemiologist with expertise in the areas of fish health (Atlantic salmon, cod, trout, charr, lobsters, shellfish) and evidence-based Veterinary medicine. She has worked extensively with cold water marine species from broodstock to plate during her PhD work. She has managed fish health surveillance programs for monitoring and testing cultured salmonids for reportable and economically significant viral, bacterial, fungal and parasitic pathogens in the NL aquaculture industry.

ATLANTIC SALMON RESPONSE TO ISAV: AN UNWELCOME GUEST ON EXTENDED STAY
- presented by Nellie Gagne, Fisheries and Oceans Canada

Aquaculture in marine net pens exposes fish to pathogens, which may lead to infections; therefore, there is a need to understand the pathogen / disease to mitigate the concern. Regarding ISA and ISAv there have been a number of past research project conducted including work to evaluate natural immunity vs induced by vaccination, immune response, minimal infectious dose, and effect of family, wild vs cultured fish susceptibility, epidemiology and viral strain variation. Work is continuing to evaluate the dispersal and viability of ISAv in saltwater. Studies have shown that selected populations of wild fish are more resistant to ISAv than farmed fish. Although HPR0 is non-virulent, it is infectious and can spread rapidly in a population of salmon. ISAv is thought of as a recently found virus but evidence shows that the European and North American strains came from the same ancestor virus becoming evolutionarily separate over 100 years ago. A second introduction of ISAV from Europe occurred in Atlantic Canada more recently.

Since 2012 and up to July 2016, there have been 60 ISAv notifications investigated and confirmed by the National Aquatic Animal Health Program (NAAHP). Of these ISAv-HPR deleted strains have been identified 17 times, with 12 of these cases having unique HPR variants. Only the consecutive ISA outbreaks in Newfoundland in 2012 and 2013 were found to be related strains due to horizontal transfer of the virus. Most of the virulent strains (14) were North American, with the remaining being European strains found in 2016. The newer strains of HPR deleted ISAv have lower pathogenicity. In controlled challenges in 2012, the Nova Scotia (NS) isolates and one of the Newfoundland (NL) isolate showed moderate mortalities.

The further 43 ISAv notifications resulted in HPR0 being identified, the majority (39) the European strain. The NA HPR0 strain found in NS in 2012 represent the first detection of this variant.

Prediction of outcomes from HPR-deleted strain sequencing / knowing what the strain type is, is basically not possible as the strain type does not necessarily relate to virulence. The original HPR4 ISAv strain identified in 1996 and involved in a majority of outbreaks at the time, has not been identified again, though detection of "closely" related strains with lower virulence in the field have been confirmed. It is possible that low virulent strains will continue to circulate and outbreaks of more virulent strains will appear occasionally so continued surveillance is needed. Combining increased monitoring with sequencing will also provide information on horizontal transmission.

New strains of ISAv have showed up regularly so further monitoring and sequencing may help determine if the new strains originate from HPR0, if they are mutations from circulating HPR-deleted strains, and /or if there are reservoirs in wild populations transferring ISAv on farms.

Research priorities need to focus on broodstock selection for resistance and oceanographic modeling for mitigation of horizontal transmission. This work could be combined with eDNA detection of ISAv in seawater.

See Attached Presentation

Nellie Gagné
Nellie Gagné is a scientist and head of the Molecular Biology group at DFO-Gulf region since 2001. The expertise of her group is in the development and validation of molecular diagnostics of fish diseases, the development of improved in situ assays, and research focusing on disease control in general. Atlantic salmon response to ISAV is explored through genomic. Her research on ISAV focuses on strain variability, salmon response, environmental conditions affecting the impact of ISAV, and more.

FEDERAL APPROACH TO FREEDOM EVALUATION FOR REPORTABLE AND EMERGING DISEASES
- presented by Annie Wagener, Canadian Food Inspection Agency

As part of the National Aquatic Animal Health Program (NAAHP), CFIA has been designing a population level program to declare an area free of a disease, in this case freedom from ISA. In BC, there have been no confirmed reports of ISA but some lab reports indicated ISAv detection in wild salmon in 2011. ISA could not be confirmed from the samples but investigations began.

To determine if Pacific salmon are susceptible to ISA lab challenge studies on chinook, chum, Coho, steelhead were performed. Lab challenges indicated resistance to infection. Mortality was only recorded in the first days after being injected with the virus, with no clinical signs of ISA observed and the virus only detectable at the end of the experiment in a few fish. In tests with rainbow trout and brown trout, neither had shown clinical signs of ISA, though in lab studies the virus may replicate in these hosts. During the Wild Fish Surveillance program of 2012-2013 over 8000 wild salmon were tested for ISAv using RT-PCR with no clinical occurrences or confirmed detections.

The evaluation of the existing surveillance program on salmon farms began with an introduction risk evaluation, a review of the process and diagnostic testing results from both government and industry. There are several potential pathways of ISAv introduction to fish including infected wild or farmed salmonids, other wild fish or infected equipment and there are ongoing evaluations for new risks of introduction.

A table presented the number of salmon tested during the surveillance program between 2006 and 2011 for BC farmed salmon by both industry and the provincial government. Industry alone tested over 5000 salmon, all results have been negative for both HPR0, and HRP deleted ISAv.

With the syndromic surveillance (investigation based on clinical signs) program that was already in place for farmed salmon, CFIA was very confident that the ISA seen in other parts of the world, was not present in BC farmed fish but felt that additional active surveillance was required to increase confidence that HPR0 strains were not present. During the CFIA program of 2014-2015 over 8500 salmon were tested with no suspect or confirmed positive results.

See Attached Presentation

Annie Wagener

Before moving to Prince Edward Island and joining the Canadian Food Inspection Agency (CFIA) in 2002, Dr. Annie Wagener practiced privately in the Annapolis Valley in Nova Scotia. Prior to filling her present role as a National Veterinary Science Specialist in the Aquatic Surveillance and Epidemiology Section (Ottawa), she held positions in animal health, meat hygiene, acted as District Veterinarian of PEI and Veterinarian-in-Charge of two federally inspected abattoirs in PEI. In 2013, Annie completed her Ph.D. (Epidemiology) at the Atlantic Veterinary College, and joined the Aquatics Section in July 2014. Since joining, she has been active in national surveillance in both wild and farmed fish, and as the on-site CFIA epidemiologist, collaborated with the Province of Alberta in developing and implementing surveillance for whirling disease in salmonids. As part of an inter-departmental team, Annie developed a template for assessing the surveillance systems of international trading partners. She completed evaluations of diagnostic test protocols for spring viremia of carp and salmonid alphavirus, and presently sits on an OIE working group tasked with revising chapters of the OIE Manual of Diagnostic Tests for Aquatic Animals. Annie has recently taken on the role as lead of national surveillance in farmed fish.

THE HORIZONTAL AND VERTICAL DISTRIBUTION OF SEA LICE LARVAE IN RELATION TO SALMON FARMS IN THE BAY OF FUNDY

- presented by Emily Nelson, St. Andrews Biological Station

Sea lice are a priority fish health challenge to the southwestern New Brunswick salmon farming industry requiring changes in stocking strategies and increasing costs for the industry as the sea lice develop tolerance to the few existing approved treatment products. To date, the majority of the sea lice research focuses on the attached stages, and while these larval stages are difficult to study, they are an extremely important aspect that must be explored. A better understanding of the early life history (larval) of sea lice is needed. Studies conducted to date on the larval stages in SW NB have reported very low densities; however, sea lice infections continue to occur on salmon farms in the area. The current research project builds on this research and focuses on addressing where the larvae are found on and off aquaculture sites, their vertical distribution, at what densities are the larvae leaving the farm (horizontal distribution) and whether there are areas on the farm where larvae are more prevalent.

The vertical distribution of sea lice larvae was quantified by sampling the water column using a pump system at shallow (1, 3, 6m) and deep (10, 14, 17m) depths every 90 minutes over a period of 24 hours. The majority of the larvae sampled were the nauplii stage (88%) rather than the copepodid stage (12%), however larvae in SWNB were

found throughout the water column (no significant differences between depths). When densities were pooled between shallow and deep depths, the larvae exhibited a diel cycle where they are deeper during the day and shallow at night. As sea lice larvae were found at all depths from 1-30m, modelling using only surface densities (as is the typical practise for other areas) will greatly underestimate larval densities in SW NB.

Sea lice larval densities were captured over 5 years throughout the SWNB to determine if sea lice larval densities differ between farm and reference sites. Farm densities were found to be significantly higher than reference sites, however all larval densities captured were low (<1 larvae per m3).

To determine if there are specific locations of high larval densities on a farm site testing was done over 5 years to compare inside and outside cage densities and inner versus outer areas of the cage array. The project design and results were presented indicating there was no difference in densities observed between sampling locations. To quantify the horizontal distribution of larvae leaving a farm site, 100m transects were completed with replicate sampling at various distances. Larval densities did decrease exponentially going away from cages starting at approximately 2.4 larvae/m3 at cage edge to 0.3 larvae/m3 at 100m.

Based on a given sea lice number on a site stocked with salmon, some basic calculation estimates presented indicated that the observed larval densities are drastically lower than that estimated from fish load. Further work was conducted to determine if certain site activities such as harvesting may influence the number of larvae being found and / or could additional aspects of farm structure be contributing to sea lice success. The results showed that there may be compounding factors on site which could be contributing to sea lice larval densities. Work to further evaluate the relationship between site activities and larval densities will continue along with research to capture potential seasonal variations of sea lice larvae on farms and reference sites.

See Attached Presentation

Emily Nelson

Emily Nelson, a native New Brunswicker, has an MSc from the University of New Brunswick and is an Aquatic Science Biologist for Fisheries and Oceans Canada at the St. Andrews Biological Station (New Brunswick) in Shawn Robinson's lab. Her research project focuses on sustainable aquaculture and explores the early life history stages (larval) of sea lice and their distribution in and around salmon farms in the Bay of Fundy in order to understand the ecological relationships involved with this association. The results of this research are anticipated to help provide better tools for the management and control of sea lice in order to further increase the level of sustainability of salmon farms. She has worked previously in Ottawa where she was involved with DFO's Aquaculture Collaborative Research and Development Program and is currently funded through DFO's Program for Aquaculture Regulatory Research (PARR).

SALMON MIGRATION: A KEY PROCESS FOR UNDERSTANDING LICE INFECTION IN WILD SALMON
- presented by Marc Trudel, St. Andrews Biological Station

The work presented is based on research conducted in British Columbia through the Pacific Biological Station because of declines in the Fraser River sockeye populations and that the suggestion two migration routes may explain differences in abundance. As one of the suggested migration routes passed salmon farms it was inferred by other researchers that sea lice from these farms contributed to the lower population levels on the route. Map and data were presented showing sockeye production data, migration routes with farm site locations, and distribution of Fraser River versus Harrison River stock.

A brief review of several parameters of both stocks such as size at migration and peak migration period identified differences suggesting that Harrison River sockeye were not a good control for comparison with the Fraser River sockeye. DNA analysis of another 6000 sockeye salmon showed that Harrison River sockeye salmon were using both migration corridors.

With published papers inferring that lice infection on juvenile Fraser River sockeye salmon in the Discovery Islands area was higher downstream due the salmon farms, infection levels on various salmonid and other marine species were

evaluated. Sea lice and health surveys were conducted between May and June of 2010 to 2012. Freshwater samples were obtained from Chilko Lake and lower Fraser River. Marine samples were collected from over 75 sites throughout the Strait of Georgia and Johnstone Strait. Data recorded included the type of Pacific salmon, sea lice counts on salmonid and non-salmonids captured, histology samples were taken, and sockeye salmon were screened for pathogens. The annual results were presented.

With high numbers of juvenile Pink salmon from the Fraser River in even years, juvenile salmon dominated the catches in June 2010 and 2012, while mostly non-salmonids (herring and stickleback) were found in May and June of 2011. The louse identified in 99% of cases on all salmon and non-salmonid hosts was *Caligus clemensi* with stickleback and sockeye salmon found with the highest sea lice burdens.

In the evaluation of sea lice infection with distance from the mouth of the Fraser River, data provided no evidence for dramatic increases in sea lice numbers on sockeye near the salmon farms. Evaluation of residence time of juvenile Fraser River sockeye was completed using data from rotary screw trap sampling (2012-2016) and purse seine surveys in Johnstone Strait (2014-2016). The Fraser River sockeye salmon residence time in the Strait of Georgia for 2014 was presented showing that these salmon were in the Strait of Georgia for about six to seven weeks overall, with approximately 80% of the sockeye in the Discovery Islands for about two weeks. Published data using acoustic telemetry infers that on an individual level the average residence time was estimated to be two to three days.

The changes in the sockeye population are most likely due to cumulative effects of multiple stressors such as climate changes (sea surface temperatures, ocean circulation). To further assess these interactions the life-history, migration behaviour and infection history of these sockeye prior to reaching the salmon farms must be considered.

See Attached Presentation

Marc Trudel
Dr. Trudel is a research scientist who leads multidisciplinary research program aimed at assessing the long-term effects of climate change on salmon productivity and the limits to marine ecosystems productivity for Pacific salmon. He has extensive experience in designing and managing large-scale field programs in coastal waters of British Columbia, and in studying the migration behavior of juvenile salmon. His research program has contributed to the development of leading indicators of marine survival that are used to forecast adult salmon returns in southern British Columbia and to understanding the interactions between wild and cultured salmon. He recently relocated to St. Andrews where his research will focus on aquaculture-ecosystem impacts and risk mitigation

SALMOSAN® VET – COMMITTED TO SEA LICE CONTROL
- presented by Jason Collins, Fish Vet Group

The work outlined in this presentation was undertaken by Benchmark Animal Health to not only achieve maximal sea lice clearance rates on farms, but also support the prescribing community in their decision-making.

Benchmark Animal Health is aware of the pressures facing medicine prescribers, farm health advisors and producers with respect to medicine residues. The protocols described are frequently undertaken in some regions. To support industry, this information regarding the impact on medicine residues is being provided. Along with Pharmaq, work was completed to investigate the simultaneous treatment of Salmosan Vet and Alpha Max, which is an off-label use of both products. The choice may be made by prescribers who are facing fish welfare issues due to lack of efficacy of established sea lice treatment protocols. For the trial, a commercial farm completed the treatment using standard protocols for each product. Skin and muscle samples were taken from fish before treatment, and at 24, 48, and 96 hours after treatment for residue testing. Results conducted by an outside lab as directed by regulating authorities showed no medicine residues for either product. As such, the simultaneous treatment does not affect the residues of azamethiphos or deltamethrin.

The ecotox and environmental impact of any medicine is a major factor for consideration when determining sea lice control methods and medicine prescription. Legislation surrounding treatment administration can restrict the number

of treatments resulting in limiting production and in some circumstances, hinder animal welfare. The half-life for Salmosan as calculated by historical methods is a reported 8.9 days and it is this number that all discharge regulations are based. The current understanding of the biochemistry and pharmacokinetics of Salmosan Vet is demonstrating that the product breaks down remarkably quickly in seawater so a study to calculate the half-life under farm conditions was undertaken. Based on the first study, the half-life was recalculated as 2.31 days. Work is ongoing to evaluate the impact of pH and water temperature on half-life.

Research was also undertaken to understand alternate mitigation options following Salmosan Vet treatments. In this project, the azamethiphos half-life was derived under GLP conditions per standard treatment protocol, as well as after the introduction of a treatment dose of (1500 ppm) peroxide. The resulting calculation post peroxide addition indicated a half-life of just over 45 minutes.

Working with industry to try and achieve higher lice clearance rates an "All in One" protocol has been developed to be used with low salinity treatment. This protocol consists of a 3-hour well boat treatment with low-salinity water (3-4ppt) and Salmosan Vet added for the final hour. Trials have resulted in up to 100% clearance rates for all stages of sea lice, even on units where multi-resistant populations of lice are present. This high mortality, regardless of life stage, is thought to be a result of osmoregulatory stress as the lice take in larger amounts of azamethiphos than under normal seawater conditions. The underlying mechanism is still under examination. This new application of Salmosan Vet has now been supported by tank trials aimed at establishing the efficacy of this method in populations of lice that have been previously resistant to azamethiphos. Results showed 91% reduction in all stages of sea lice after 17 hours with the remaining 9% of the lice attached but not viable.

Commercial field trials followed this lab work. The two farms involved in these trials were previously experiencing clearance rates as low as 40%. After the "All in One" treatments the two farms reported 100% lice clearance two days post treatment with very low fish mortality. The addition of Salmosan Vet for the final hour reduces the total time of a typical freshwater treatment by 5 hours but close attention should still be paid to water quality parameters. It is also recommended that only well-boat crews that are experienced in fresh water treatments be used for this new protocol.

Going forward, treatments are being undertaken on a regular basis to collect more data to inform the industry on how to achieve maximal clearance rates and maintain fish health and welfare.

Jason Collins

Jason has spent the last 16 years committed to development of sustainable aquaculture, with much of this time focused in the global salmon production regions. In his present role as Fish Vet Group's North American Sales & Technical Manager, Jason works to grow the company's diagnostic and vet service business, and in support of Benchmark Animal Health's aquaculture product portfolio. Jason's presentation will describe recent developments for Salmosan, including research undertaken, and new customer applications in Europe.

SEA LICE 2016: TRENDS TO INFORM MANAGEMENT DECISIONS IN NEW BRUNSWICK

- *presented by Larry Hammell, Atlantic Veterinary College*

The Fish-iTrends (FiT) database was designed by AVC for the New Brunswick industry in 2010 and since then it has expanded to include industry partners in Newfoundland and Nova Scotia. The presented data provided an overview of the trends from 2010-2016.

Reviewing the sea lice numbers at the industry-wide level, regardless of life stage, over this period 2016 was comparable to 2010. Sea lice annual trends by specific BMAs are influenced by the production cycle of fish stocked in the BMA since lice numbers would naturally differ depending on smolt versus harvest size fish. As graphically presented, sea lice numbers also change as the frequency of treatments is applied within an area. Automatic FiT summaries of sea lice changes from one week to the next are generated and sent to decision makers weekly.

Based on industry approved levels of access, Fish-iTrends database informs regulators regarding compliance for required reporting of sea lice counts (the table provided demonstrated site level compliance). Colour codes indicate sites that submitted counts (one colour for meeting the minimum requirement, another for submitting counts but fewer fish or cages than required). Although FiT provides a way to track compliance, it is the responsibility of provincial regulators to ensure compliance is achieved.

Fish-iTrends data can be used to evaluate time trends for treatment frequency by bath or in-feed product. Efficacy of each treatment for each life stage is compared over the years of use and based on pre-defined thresholds of lice reduction post-treatment, the proportion of successful treatments is visualized over time. Pre-treatment and post-treatment lice counts are recorded along with the timing of these counts allowing an evaluation of the best time to perform these counts to reflect the true proportion of lice removal. Based on statistical models, counts should be conducted one to three days following bath treatment (i.e. not on the same day and not after 3 days) to ensure that the estimate of effect from treatment does not reflect other changes occurring since the treatment. Similar models indicate that pre-treatment levels of sea lice can influence the estimated treatment effectiveness.

See Attached Presentation

Larry Hammell

As an aquatic veterinary epidemiologist, Dr. Larry Hammell DVM, MSc has been the lead proponent on many large, clinical research projects and partnerships with industry and government agencies. He is Co-Director of the Collaborating Centre for Epidemiology and Risk Assessment of Aquatic Animal Diseases (ERAAAD) for the World Organization for Animal Health (OIE), Professor and Associate Dean (Graduate Studies & Research) at the Atlantic Veterinary College, University of Prince Edward Island.

RISK FACTORS FOR TREATMENT FAILURE IN ANTIBIOTIC TREATMENTS IN FARMED ATLANTIC SALMON IN CHILE

- presented by Derek Price, Atlantic Veterinary College

Piscirickettsia salmonis is the bacterial pathogen responsible for the chronic disease Piscirickettsiosis or SRS. This disease is the main cause of infectious disease-related mortality in all salmonids in Chile and is spread through horizontal transmission. A surveillance and control program has been in place since 2009, but antimicrobial treatments have had variable success even though sensitivity studies report low resistance, which suggests treatment failure is multifactorial. In this study, we focused on evaluating the influence of the antibiotic product used, water temperature, average fish weight, and pre-treatment mortality level over treatment failure.

In reviewing data from the Intesal-Salmonchile database on the first antibiotic treatment for over 2000 pens in 2014, we found that approximately 45% had weekly mortality rates that were above our threshold for normal mortality (0.1%) after the treatment ended and were deemed failed treatments. We also found that the probability of failure was associated with timely treatment; the typical timing of diagnosis and start of treatment is occurring when the clinical disease is already present and the pathogen has established in the population making it harder to eradicate. The probability of failure was also higher in larger fish. As fish grow, the typical practice is to reduce the number of feedings to ensure all fish are getting the appropriate amount of feed. This practice may be impacting how the antimicrobials work because the pharmacokinetic properties of different products are not being considered when implementing certain husbandry practices such as reduction of feeding frequency.

In a separate study, we assessed over 2600 tissue samples from over 100 treatment events and we determined if the epidemiological cut-off (ECOFF) values for the antimicrobials used against *P. salmonis* in Chile were reached. The data showed a wide distribution of concentration in tissue, with a high proportion of individuals below ECOFF. Body condition appears to explain some of the variation, within a population the larger fish seem to have consumed the majority of the food presented therefore reaching higher concentrations of antimicrobial in tissue.

Future research includes an assessment of the effect of feeding frequency in antibiotic tissue concentration and treatment success, and a review of antimicrobial sensitivity surveillance programs to assess the role of resistance.

See Attached Presentation

Derek Price
Derek Price is an aquaculture veterinarian with over 10 years of field experience in the Chilean salmon farming industry completing his PhD in Veterinary epidemiology at the AVC with Dr. Sophie St-Hilaire. Dr. Price is currently working with antibiotic tissue concentration data to assess factors that influence treatment failure.

AN UPDATE ON THE EPIDEMIOLOGY OF ULCER DISEASE
- presented by Brett MacKinnon, Atlantic Veterinary College

In recent years, the Atlantic Canadian salmon farming industry has been witnessing outbreaks of ulcer disease, starting in the summer months. Lesions are typically found on the lateral side of the fish and can vary in size. The disease can cause high mortality and downgrading of the product, leading to economic losses.

It is thought that ulcer disease is caused by *Moritella viscosa*, as in the case of winter ulcer disease in Europe, but the bacterium has been difficult to isolate from field samples. There are several difficulties with managing this disease. It is believed that the causative agent of ulcer disease is endemic in the marine environment, antibiotics do not effectively control mortalities associated with this disease, and the current vaccines being used in Canada do not always prevent the disease.

The objective of the study was to investigate the temporal and spatial patterns of ulcer disease, within and between farms, to determine potential sources and transmission of infection. Pen-level mortality data, management practices, and medical records from salmon farms in Atlantic Canada between the years 2014 and 2016 were extracted and analyzed to determine the time-periods of the ulcer disease outbreaks. Results from the cage and industry level analyses of the onset, magnitude, and duration of ulcer disease outbreaks was presented.

In both 2014 and 2015, the onset of ulcer disease outbreaks occurred in cages within 3 weeks of each other on a particular farm, with the exception of one farm in 2015, suggesting a common point source exposure to the pathogen. The proportion of cages diagnosed with ulcer disease within the affected farms ranged from 8% to 100%, which suggests that cage-to-cage transmission of the pathogen is unlikely since unaffected cages were evident on most of the farms diagnosed with ulcer disease. The magnitude of cage-level total mortality associated with outbreaks of ulcer disease ranged from 0.006 to 23.3%. At the farm-level, there was high variability in the average duration of outbreaks, which ranged from 1 to 10 weeks in 2014 and 5 to 26 weeks in 2015. The variation in magnitude and duration of outbreaks may indicate that exposure to the pathogen is not uniform on a farm and/or other factors may be associated with severity of the disease including cage density, predation stress, timing of treatments, and co-infections.

Of the 29 farms analyzed during 2014 to 2016, 12 (41%) were diagnosed with ulcer disease. The pattern of disease was similar during 2014 and 2015, with the earliest outbreaks occurring in the summer and all affected farms having outbreaks by early to mid-fall. This may suggest that the pathogen is relatively widespread in the area at a specific time of the year or the pathogen is present during other times of the year but outbreaks are triggered by an environmental factor.

A laboratory study was conducted, mimicking field conditions, to determine if ulcer disease can be transmitted horizontally between tanks using an *M. viscosa* isolate from Atlantic Canada, and to describe the progression of ulcer disease. The trial lasted for 26 days and fish were sampled from control and experimental groups over this time to follow progression of disease. Typical lesions of ulcer disease occurred on fish infected via bath immersion with *M. viscosa* but there was no horizontal transmission to tanks of naïve fish that received the effluent water of exposed fish.

All fish remained on feed except those with the most severe lesions and many of the fish with mild lesions recovered by the end of the experiment. There is also evidence of this recovery occurring in the field.

The objective of the second laboratory study was to determine whether ulcer disease lesions can be induced in salmon with the extracellular products (ECP) produced by *M. viscosa*. Past histological evaluation of ulcers from field studies did not have evidence of bacteria in the lesions. One week after subcutaneous injection with *M. viscosa* ECP (in broth), several fish in each experimental tank showed signs of swelling, superficial hemorrhage and mild-moderate erosion of the epithelium in the area of the injection site. From days 12 to 17, five fish developed ulcers at the injection sites while the control fish continued to have no significant findings. It appears that the ECP of *M. viscosa* causes necrosis/swelling/hemorrhage/ulcers of Atlantic salmon tissue when injected subcutaneously under laboratory conditions.

See Attached Presentation

Brett MacKinnon

Dr. Brett MacKinnon is an MSc student at the Atlantic Veterinary College, UPEI. Her thesis is focusing on the epidemiology of ulcer disease of Atlantic Salmon in Canada. She graduated with a BSc Biology from UNB (2006) and a DVM from AVC (2012). Prior to pursuing graduate studies, she worked as an aquatic health veterinarian with the CFIA and small animal veterinarian in private practice.

PLENARY DISCUSSION - WHAT'S NEXT? WHAT ARE THE R&D PRIORITIES?

The conversation on research and development priorities focused on issues identified during presentations on ISA.

1. Wild reservoirs

There has been work completed that implicates multiple wild fish species as potential reservoirs of ISAv. It was noted by one participant that due to a poor local catch at the time, herring was brought into local fish plants from Norway, Iceland, and Greenland, which corresponds with timing and location of initial cases in 1996/97. An AVC risk factor study for ISA in 2010 included the presence of mackerel. Juvenile hake and mackerel were observed in salmon cages prior to the 2015/2016 outbreaks. Eels were also mentioned as a potential reservoir. The fact that the North American strain has been here in New Brunswick for >100yrs, pre-farming, proves the virus is maintaining itself in the marine environment long before aquaculture.

2. Migration patterns of wild fish

Once potential wild reservoirs are identified, an understanding of their migration patterns could help industry develop avoidance strategies if possible.

3. Does HPR deleted develop from HPR0?

Information from the Faroes seems to provide field evidence for this hypothesis. However, data from Atlantic Canada does not seem to support this thought. Research priorities noted were:

a. Understanding why / how HRP0 becomes virulent if this is the case
b. How HPR0 virus is replicated within host without causing disease
c. Effect of environmental conditions and stressors on mutations of ISAv. Are there environmental stressors such as temperature, pH that are potentially influencing mutations / deletions in HPR0?
d. Are there on-farm stressors?

4. Inability to culture HRP0

HPR0 cannot currently be cultured in the lab so unless a procedure can be developed to work with this virus (to grow it and infect fish in the lab) research cannot evaluate how it works or what it does within the fish, etc.

5. Nomenclature / common terminology

Part of the work that needs to be completed is to use the same nomenclature for the virus so the European, North American strains, etc., are named in the same way / based on the same set of criteria. Companies / research institutions must be identifying viral strain based on this common process.

6. Link between cases / strains ISAv – genetically.
    a. No two cases are the same in clinical presentation, mortality rates, etc between cages, sites, provinces.
    b. Sequencing may help inform horizontal transmission process

Forum Wrap-up

Research and science remains essential to ongoing development of the aquaculture industry. It continues to provide the salmon farming industry and broader stakeholders with important information on a range of topics central, while providing opportunities for collaborative projects intended to develop a sustainable industry in Canada. These include fish health, operational best practices, environmental monitoring and regulatory frameworks, as well as technological advancement.

Additional research is needed on various fronts including fish health. The pharmacokenetics and pharmacology of antimicrobials should be reviewed in the context of husbandry practices and changing environmental conditions. In addition to those items specifically identified during the plenary discussion other ISA research topics include vaccine improvements, the distribution of HRP0 via gill sampling, oceanography work to understand horizontal transmission, and eDNA testing for ISAv in the marine environment. Disease resistant broodstock development is a reoccurring priority, along with questions regarding what is the best tool(s) to use to detect the virus. Those typically used IFAT, Rt-PCR results differ and sampling site (tissue) may influence what you find. The Rt-PCR test is gold standard in some places with high through-put for tests though q-PCR is more sensitive but may be too sensitive.

Future work on ulcer disease includes diagnostic tools and treatment options but the causative agent still needs to be confirmed. Outbreaks are seen in March and during the summer months so is there a role of temperature in these outbreaks and effort is required to determine if they are caused by the same isolate. Research is needed around the carrier status of fish that survive as well as the potential role of vectors like sea lice, lumpfish.

The multi-faceted challenges of sea lice remain a research priority. Wild reservoir identification and understanding migration patterns of wild fish influenced by climate change, can add knowledge to inform management planning. Understanding the entire process of how Coho salmon respond to and eventually reject sea lice, could lead to multiple preventative and mitigation measures to be used with Atlantic salmon such as gene editing and or bolstering immune system response. Understanding larval sea lice behavior, life history and distribution may also provide direction on avoidance / preventative methods that could be employed by farmers.

Research that develops and assesses fish-health tool (ie., addressing and monitoring) need to continue. Additionally, research that provides greater understanding of wild salmon and lobster presence and abundance and interactions near aquaculture sites can contribute to productive discussions and interactions with traditional marine users.

Additional work on farm / wild salmon hybridization and potential impacts is important research that will continue with support of the industry. Research on wild salmon needs to show if there are river specific traits / local adaptation to river systems and the performance traits of hybrids also needs to be determined.

The ACFFA is committed to continuing to work on behalf of our members to identify industry research priorities and facilitate collaborative research activities.

As always, we greatly appreciate the contributions of the public and private research community in supporting our annual forum.

Participants

Last Name	First Name	Company
ABBOTT	MATTHEW	CONSERVATION COUNCIL OF NB
ARSENAULT	JANELLE	SIMCORP
BACKMAN	STEVE	SKRETTING
BACON	BEV	RDI STRATEGIES INC.
BARKER	DUANE	HUNTSMAN MARINE SCIENCE CENTER
BARRELL	JEFF	DALHOUSIE UNIVERSITY
BEATTIE	DR. MICHAEL	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
BELLE	SEBASTIAN	MAINE AQUACULTURE ASSOCIATION
BERRINGER	CHARLENE	ELANCO CANADA LTD
BLAIR	TAMMY	ST. ANDREWS BIOLOGICAL STATION
BLANCHARD	CLARENCE	FUTURE NETS AND SUPPLIES
BOSIEN	BRYAN	SIMCORP
BOURQUE	PETER	MITCHELL MCCONNELL INS.
BRACELAND	MARK	THE CENTER FOR AQUACULTURE TECHNOLOGIES
BRADBURY	IAN	DEPT. OF FISHERIES AND OCEANS
BRADEN	LAURA MARIE	ATLANTIC VETERINARY COLLEGE
BRAGDON	FIONA	DEPT. OF ENVIRONMENT & LOCAL GOV'T, NB
BREWER-DALTON	KATHY	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
BRIDGER	CHRIS	HUNTSMAN MARINE SCIENCE CENTER
BROWN	CHUCK	COOKE AQUACULTURE INC.
BRUNSDON	ERIC	ATLANTIC SALMON FEDERATION
CANAM	AMY	COOKE AQUACULTURE INC.
CHEUNG	LEO	RPC
CLARK	COREY	FUNDY NATIONAL PARK
CLEGHORN	KATHY	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
CLINE	JEFF	DEPT. OF FISHERIES AND OCEANS
COLLINS	JASON	FISHVET GROUP
COOK	SARAH	SKRETTING
COOKE	TARA	COOKE AQUACULTURE INC.
COX	KASHA	MERCK ANIMAL HEALTH

CRAIG	AARON	NORTHERN HARVEST SEAFARMS
CUSACK	ROLAND	NS FISHERS AND AQUACULTURE
DAGGETT	TARA	SIMCORP
DONKIN	ALAN	NORTHEAST NUTRITION INC.
DROST	TERRY	4 LINKS MARKETING
FALK	KNUT	NORWEGIAN VETERINARY INSTITUTE
FARQUHARSON	SUSAN	ATLANTIC CANADA FISH FARMERS ASSOCIATION
FILGUEIRA	RAMON	DALHOUSIE UNIVERSITY
FINN	JEAN	NEW BRUNSWICK DEPUTY MINISTER
FISCHER-RUSH	JONATHAN	UNIVERSITY OF NEW BRUNSWICK
FORWARD	BEN	RESEARCH PRODUCTIVITY COUNCIL
FOTI	MICHAEL	PHIBRO ANIMAL HEALTH & NUTRITION
GAGNE	NELLIE	DEPT. OF FISHERIES AND OCEANS
GAGNE	JONATHAN	ENTERPRISE SHIPPAGAN & INT'L SEAFOOD AND BAIT
GAMEIRO	MARTA	ELANCO CANADA LTD
GARBER	AMBER	HUNTSMAN MARINE SCIENCE CENTER
GRANT	JON	DALHOUSIE UNIVERSITY
GREEN	DARRELL	NEWFOUNDLAND AQUACULTURE INDUSTRY ASSOC.
GREENLAW	LEANNE	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
GRIFFIN	RANDY	COOKE AQUACULTURE INC.
GURNEY-SMITH	HELEN	DEPT. OF FISHERIES AND OCEANS
GUTHRIE	SKY	COOKE AQUACULTURE INC. -WEST COAST DIV.
HALSE	NELL	COOKE AQUACULTURE INC.
HAMMELL	LARRY	ATLANTIC VETERINARY COLLEGE
HANLEY	JAMES	ATLANTIC CANADA FISH FARMERS ASSOCIATION
HATT	BRADEN	EWOS/CARGILL
HICKS	BRAD	TAPLOW FEEDS
HODKINSON	TIM	COOKE AQUACULTURE INC.
HOGANS	BILL	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
HOLMES	JASON	NORTHEAST NUTRITION INC.
HORI	TIAGO	THE CENTER FOR AQUACULTURE TECHNOLOGIES
HOUSE	BETTY	ATLANTIC CANADA FISH FARMERS ASSOCIATION

HUNT	HEATHER	UNIVERSITY OF NEW BRUNSWICK
INGALLS	LARRY	NORTHERN HARVEST SEAFARMS
JAMES	SEAN	COOKE AQUACULTURE INC.
JONES	GINNY	ELANCO CANADA LTD
KAUFIELD	KATHY	ATLANTIC CANADA FISH FARMERS ASSOCIATION
KNIFFEN	TIM	MERCK ANIMAL HEALTH
KNIGHT	MORLEY	FISHERIES AND OCEANS
KNUDSON	TANYA	ELANCO CANADA LTD
KURKIMAKI	PETER	SKRETTING
LEADBEATER	STEVE	DEPT. OF FISHERIES AND OCEANS
LEAVITT	CORY	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
LEE	MISTY	NORTHEAST NUTRITION INC.
LEONG	DENNIS	CALYSTA INC.
LYONS	MONICA	DEPT. OF FISHERIES AND OCEANS
LYONS	TROY	GOV'T OF NEW BRUNSWICK
MACDONALD	ALICIA	ELANCO CANADA LTD
MACKINNON	ALLISON	ELANCO CANADA LTD
MACKINNON	BRETT	ATLANTIC VETERINARY COLLEGE
MCBRIARTY	GEOFFREY	KELLY COVE SALMON
MCCARTHY	JOSEPH	NORTHEAST NUTRITION INC.
MCGEACHY	SANDI	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
MCGEE	JOEL	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
MCGRATTAN	JASON	ELANCO CANADA LTD
MEDEIROS	DEAN	DEPT. OF FISHERIES AND OCEANS
MOORE	MARK	MARITIME VETERINARY SERVICES
NELSON	EMILY	DEPT. OF FISHERIES AND OCEANS
NESS	MICHAEL	PHARMAQ
NESS	MATTHEW	RESEARCH PRODUCTIVITY COUNCIL
NICHOLLS	KRIS	COOKE AQUACULTURE INC.
O'BRIEN	DR. NICOLE	GOV'T OF NL AND LABRADOR
O'HALLORAN	JOHN	AQUA VET SERVICES INT'L
PAGE	DR. FRED	DEPT. OF FISHERIES AND OCEANS

PAUL	STACEY	DEPT. OF FISHERIES AND OCEANS
PENTON	NORMAN	NL DEPT. OF FISHERIES, FORESTRY AND AGRIFOODS
PINEAU	AL	NORTHERN HARVEST SEAFARMS
PRICE	DEREK	UNIVERSITY OF PEI
QUAIATTINI	GORDON	MAPLE LEAF STRATEGIES
RAINNIE	DON	CONSULTANT
REID	GREGOR	DEPT. OF FISHERIES AND OCEANS
RITUALO	ANGELO	COOKE AQUACULTURE INC. -WEST COAST DIV.
ROBINSON	SHAWN	ST. ANDREWS BIOLOGICAL STATION
ROBINSON	TIM	FORT FOLLY HABITAT RECOVERY
RODGERS	BRAD	AMIRIX
ROSE-QUINN	TAMMY	DEPT. OF FISHERIES AND OCEANS
ROUSE	MIKE	OPPORTUNITIES NEW BRUNSWICK
SALMON	RUTH	CAIA
SAMWAYS	KURT	UNIVERSITY OF NEW BRUNSWICK
SEELEY	DAVID	SKRETTING
SLOAF	ANDY	COOKE AQUACULTURE INC. -WEST COAST DIV.
SMITH	TOM	AANS
SMITH	JAMEY	HUNTSMAN MARINE SCIENCE CENTER
SMITH	AMANDA	SIMCORP
ST. HILAIRE	SOPHIE	ATLANTIC VETERINARY COLLEGE
STEINE	NILS	PHARMAQ AS
STEWART	LEN	COOKE AQUACULTURE INC.
STONE	TIM	AMIRIX
STUART	ERICA	CANADAIAN FOOD INSPECTION AGENCY
SWEENEY	BOB	SIMCORP
SWIM	AMANDA	DEPT. OF FISHERIES AND AQUACULTURE
SYKES	PETER	AQUACULTURE ASSOCIATION OF NOVA SCOTIA
SZEMERDA	MICHAEL	COOKE AQUACULTURE INC.
TAYLOR	TOBI	ATLANTIC CANADA FISH FARMERS ASSOCIATION
TAYLOR	TOM	NORTHEAST NUTRITION INC.
TAYLOR	GARY	SKRETTING

TAYLOR	CORY	COOKE AQUACULTURE INC.
THORPE	BRUCE	DEPT. OF AGRICULTURE, AQUACULTURE & FISHERIES
TRENHOLM	DR. MICHAEL	CANADAIAN FOOD INSPECTION AGENCY
TRUDEL	MARC	DEPT. OF FISHERIES AND OCEANS
UPTON	KANA	AQUACAGE FISHERIES
WAGENER	ANNIE	CANADAIAN FOOD INSPECTION AGENCY
WIPER	JENNIFER	COOKE AQUACULTURE INC.
WONG	DAVID	DEPT. OF FISHERIES AND OCEANS
WOOD	KYLE	COOKE AQUACULTURE INC. -WEST COAST DIV.
YOSSA	RODRIGUE	COASTAL ZONES RESEARCH INSTITUTE

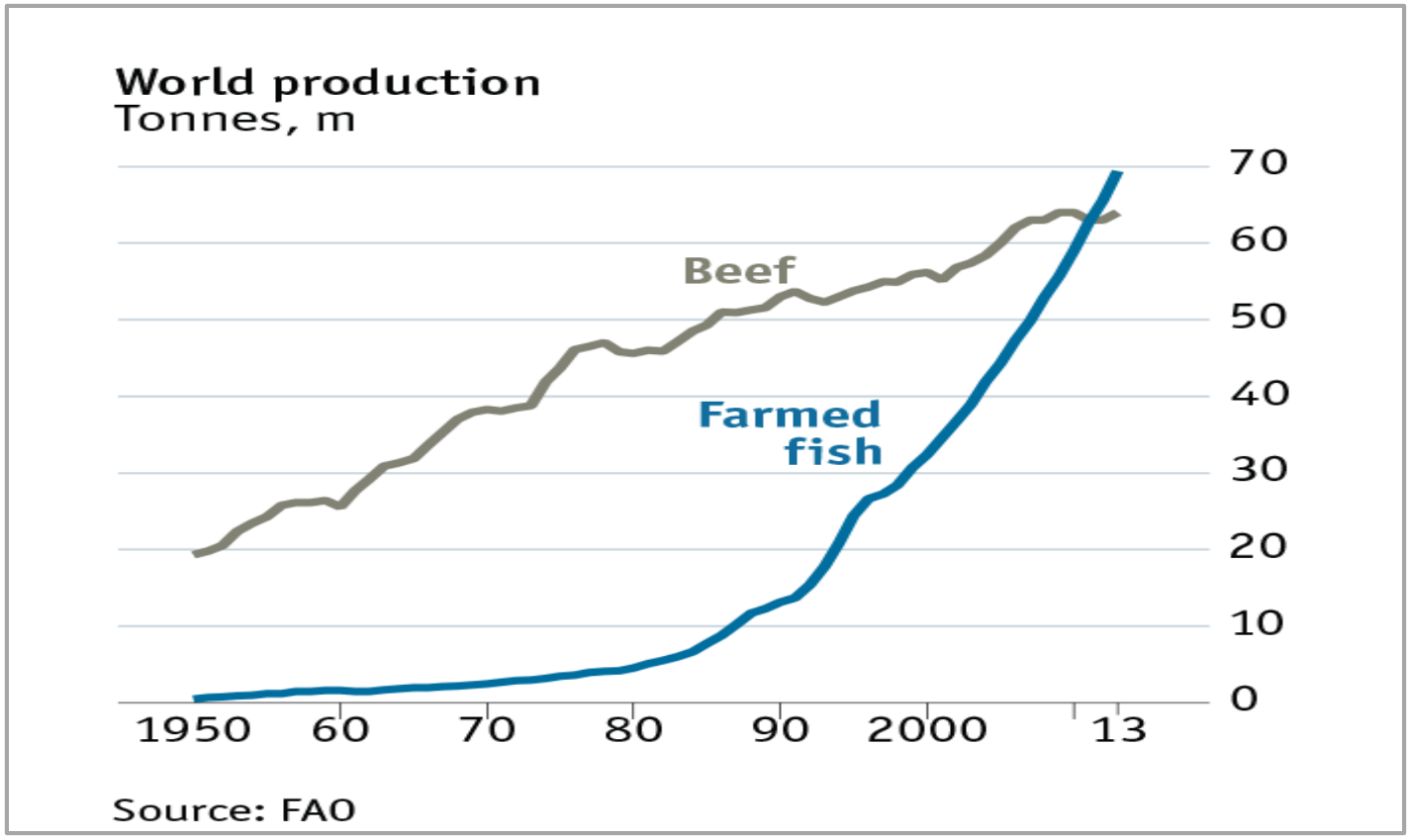
CANADIAN AQUACULTURE INDUSTRY ALLIANCE | ALLIANCE DE L'INDUSTRIE CANADIENNE DE L'AQUACULTURE

Farming Canadian Waters with Care

Delivering Healthy, Responsible, Sustainable Growth in Canada
CAIA's National Strategy: A Report Card

ACFFA Forum
October 26, 2016

Farmed Fish vs. Beef Production

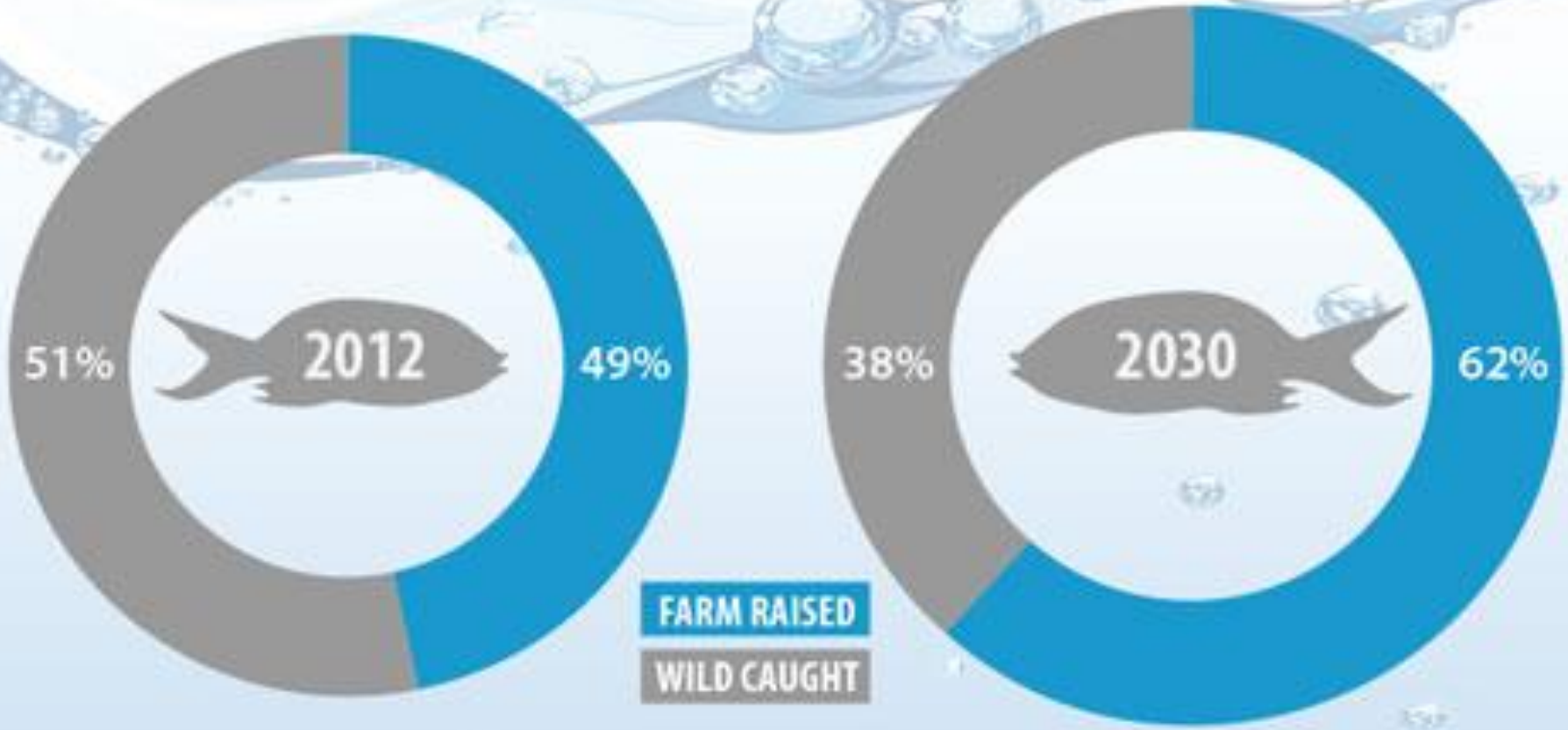


CANADIAN AQUACULTURE INDUSTRY ALLIANCE | ALLIANCE DE L'INDUSTRIE CANADIENNE DE L'AQUACULTURE

Farming Canadian Waters with Care

GLOBAL SEAFOOD CONSUMPTION

NOW vs FUTURE

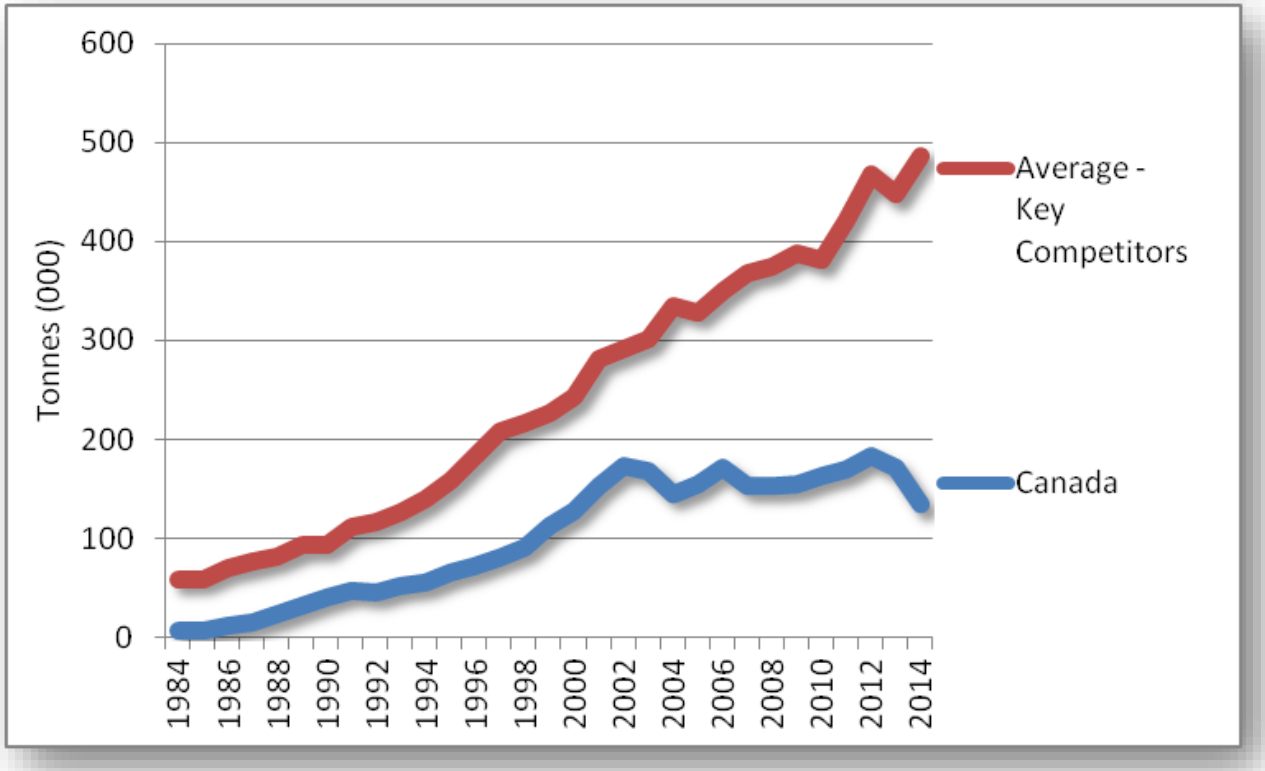


Sources: FAO FIPS (2014) // Fish to 2030 (2013)

#Fish2030

Canada falling behind key competitors

Aquaculture Production – Canada vs. Key Competitors (1984-2014)



Canada has lost **over 50%** of it's share of the world market since 2002

Source: FAO Statistics **Key competitors = Norway, Chile, U.S., Scotland, Ireland, Australia, New Zealand**

CANADIAN AQUACULTURE INDUSTRY ALLIANCE

ALLIANCE DE L'INDUSTRIE CANADIENNE DE L'AQUACULTURE

Farming Canadian Waters with Care

#1 Priority: Federal Aquaculture Act

- Science based approach to assessing risk
- Reflects unique farming characteristics
- Industry needs to be formally recognized in modern piece of legislation

Federal Gov't Commitment to Science & Research

- Science is the centre of this sector's policy and regulatory regime
- We support the government for new investments in science

Canada can learn from other jurisdictions

HAVFORSKNINGSINSTITUTTET
INSTITUTE OF MARINE RESEARCH

Institute of Marine Research - Norway

Aquaculture Science & Research Strategy - Scotland

National Strategy: Review of Key Issues

Key Issues & Priorities:

- ✓ Responsible Growth for Canada – overarching priority
- ✓ Aquaculture Act Development
- ✓ Business Risk Management tools for finfish and shellfish
- ✓ Access to Climate Change & New R&D Funding, including Aquatic MUMS program
- ✓ New species & access to broodstock policy
- ✓ Strategic Infrastructure Support
- ✓ Labour Market Development
- ✓ Access to Growing Forward III Funding

Strong Evidence-Base to Support Change

1. A Policy and Business Case for a Federal Aquaculture Act (Oct 2011)
2. Responsible Aquaculture Development In Canada - A National Strategy For New Jobs, Science-based Management, and a Healthy Food Supply (Jun 2012)
3. A Brief Primer on Government Subsidies: Overview of Methods and Sector Comparisons (Feb 2013)
4. Regulatory Cost, Economic Impacts and Overall Social Welfare Benefits of the Aquaculture Sector in Canada (May 2013)
5. Predictable Tenure/Lease/License Framework (Mar 2013)
6. Overview/Broad Elements of a new Aquaculture Act (Mar 2013)
7. Legal Elements of an Aquaculture Act (May 2013)
8. Improved Access to Feed & Fish Health Products (May 2013)
9. Farmed Seafood and Canadian Health ("Seafood Saves Lives") (Nov 2013)
10. Regulatory Reform (Nov 2013)
11. Social Licence and the Aquaculture Industry in Canada (Feb 2014)
12. Policy and Program Reforms (presentation - Apr 2014)
13. Implementing an Aquatic MUMS Program in Canada (Oct 2014)
14. Canada's Aquaculture Industry: Potential Production Growth and Footprint (Nov 2014)
15. Drafting Instructions for an Aquaculture Act (June 2015)

Responsible Growth: Key 3rd Party Support

- Conference Board of Canada Report – "From Fin to Fork"
- Senate Committee Report on Aquaculture - 2016
- Economic Advisory Council – report to Cabinet Jan 2017
  - Arrived at 4 main themes – Capital Infrastructure, Innovation, Labour Productivity and Trade.
  - Also taking a sector lens approach in the report. Using various criteria such as global growth and carbon friendly – they have agreed on **Energy, Agriculture and Aquaculture** as industries they will showcase

National Strategy: Next Steps

Political level:

- Working closely with Minister's office on all issues
- Briefing the PMO
- Continuing to present industry positions to Standing Committees (GM Salmon to Agriculture & AgriFood; TPP to International Trade)
- Outreach to all party MP's is ongoing

Gov't Officials level:

- Initiate MUMS Pilot project; advocating for full funding in 2017
- Continue negotiations and discussions re Superchill & BRM
- Initiating a revitalized Industry/Gov't Working Group

CAIA Forum November 29th

Keynote Speakers

Minister Dominic LeBlanc, Fisheries & Oceans and the Canadian Coast Guard

Opening Remarks

Andrew Pickersgill, McKinsey & Co. Economic Advisory Council

"Winning in Advantaged Sectors: Canada and Aquaculture"

MAINEAQUACULTURE

LOCAL - HEALTHY - SUSTAINABLE

GROWING MAINES FUTURE BY FEEDING AMERICA

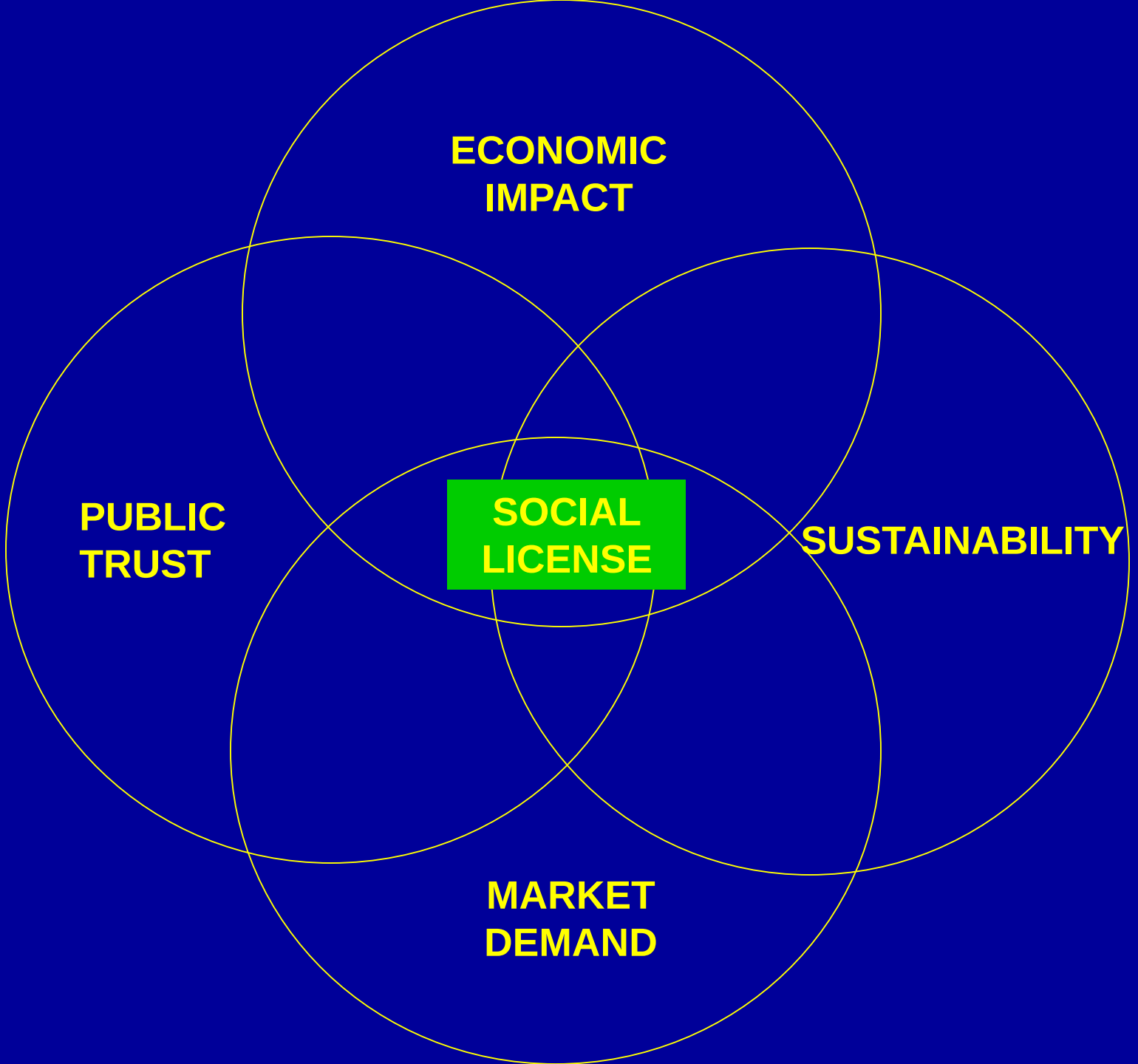
STRATEGIC ALLIANCES AND COLLABORATION AS IMPORTANT COMMUNITY OUTREACH TOOLS

“EASIER SAID THAN DONE”

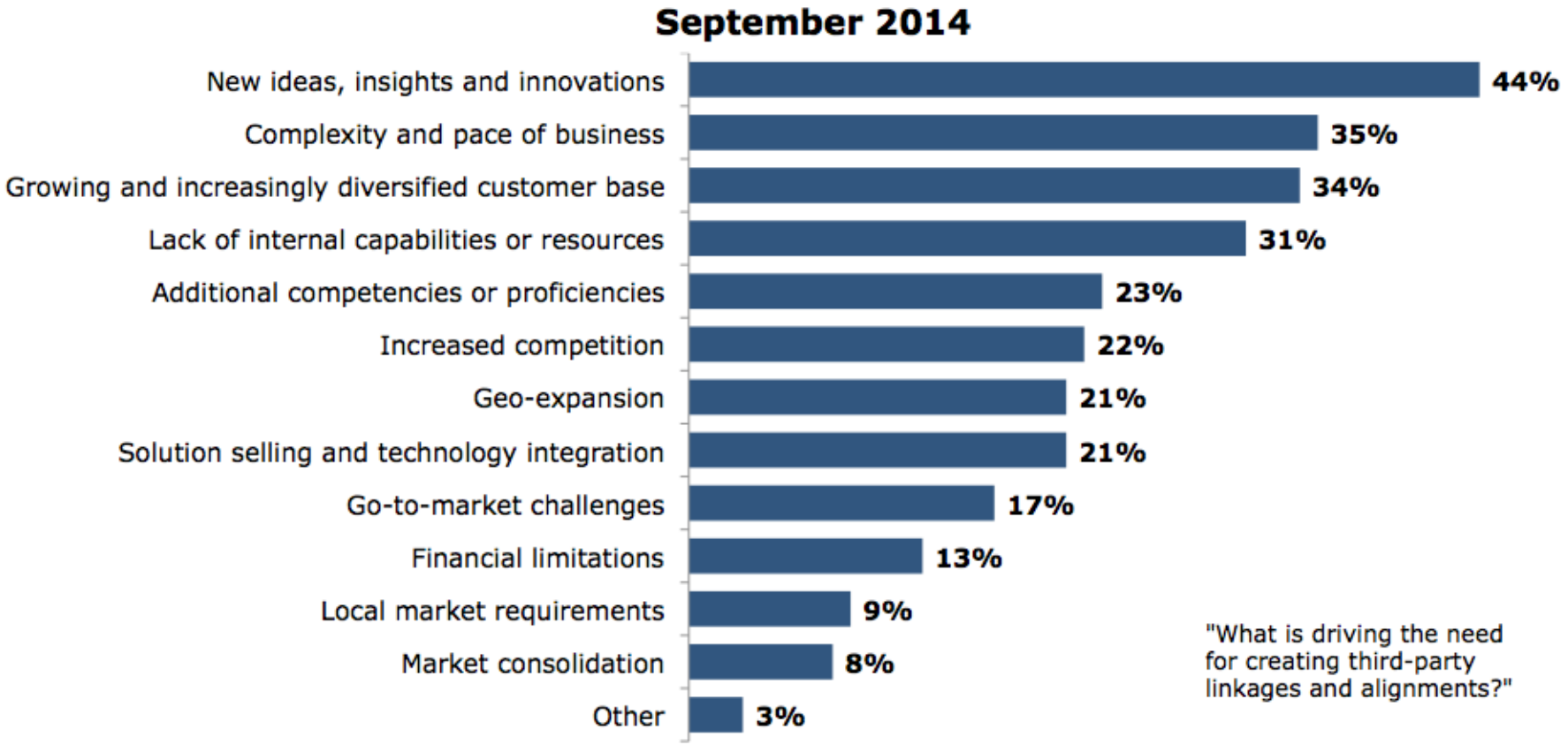
“MORE IMPORTANTLY DONE THAN SAID”

"In the long history of humankind those who learned to collaborate and improvise most effectively have prevailed."

— Charles Darwin


ECONOMIC
IMPACT
PUBLIC
TRUST
SOCIAL
LICENSE
SUSTAINABILITY
MARKET
DEMAND

STRATEGIC PARTNER KEY DRIVERS



(HSB CEO SURVEY 2014)

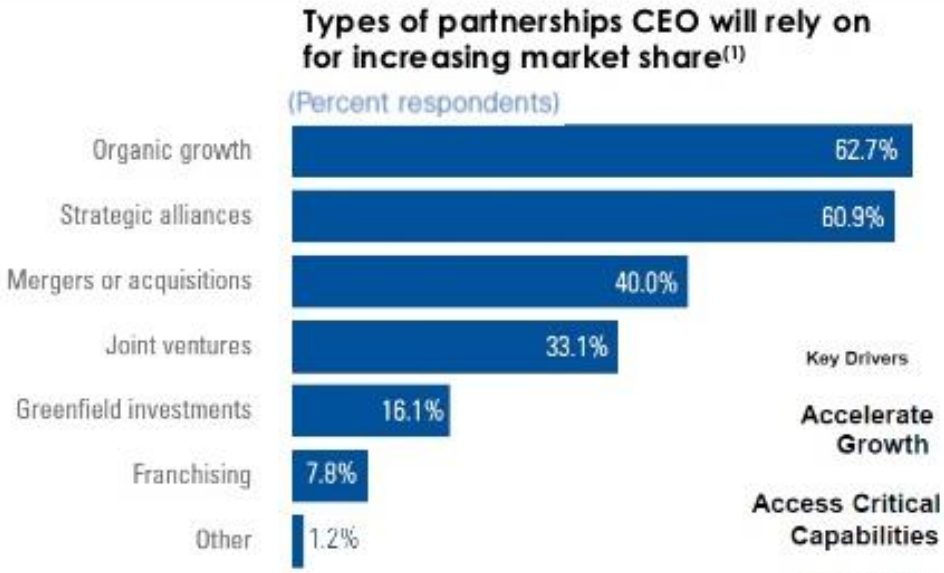
What types of partnerships, opportunities or introductions would most benefit your company?

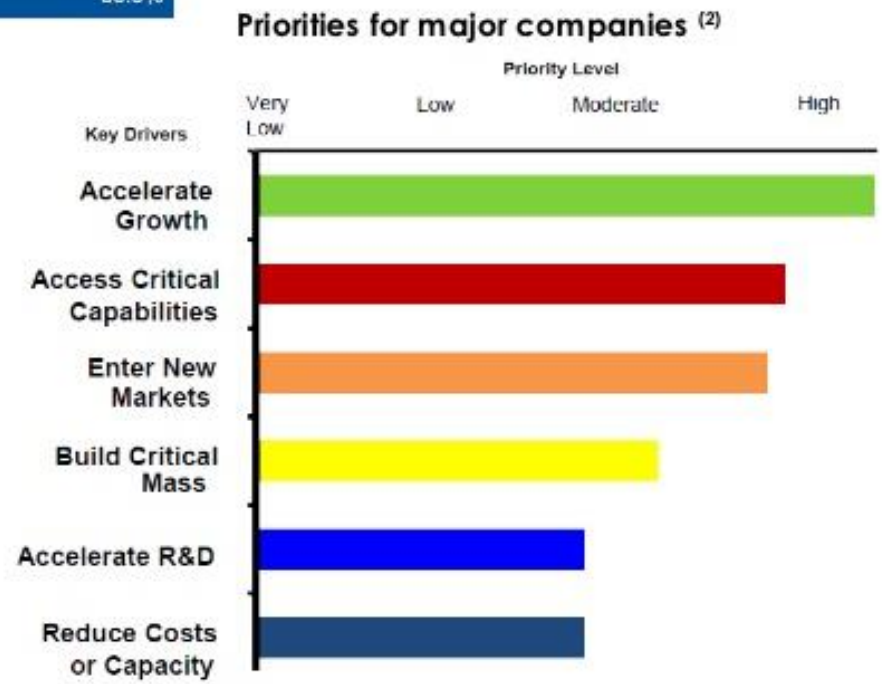
Key Findings:

Strategic partnerships that allow access to new customers and new technologies are the top concern for those surveyed. New customers result in increased market share and revenue growth, while new technologies can increase efficiency and improve offerings.

(SLOAN SCHOOL ALUMNI 2015)

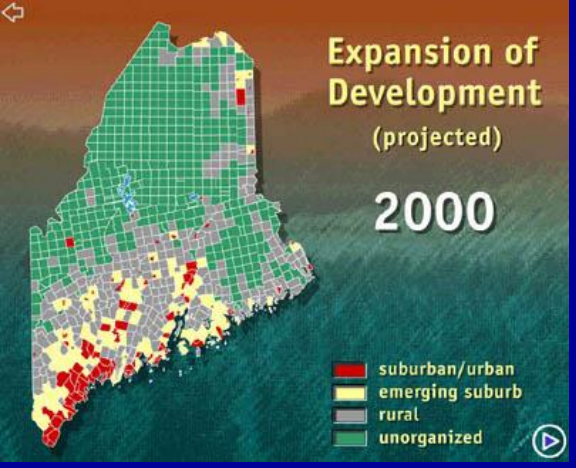
Strategic alliances

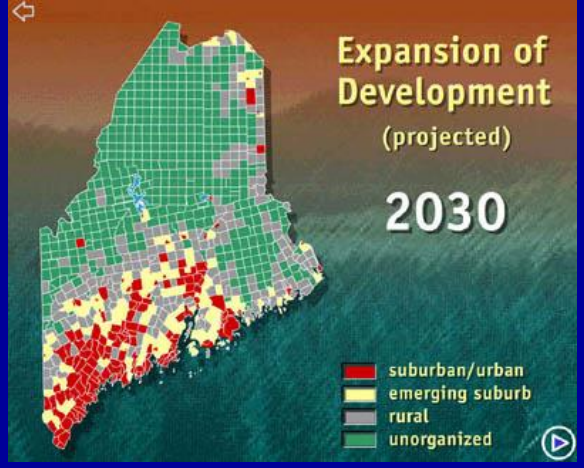


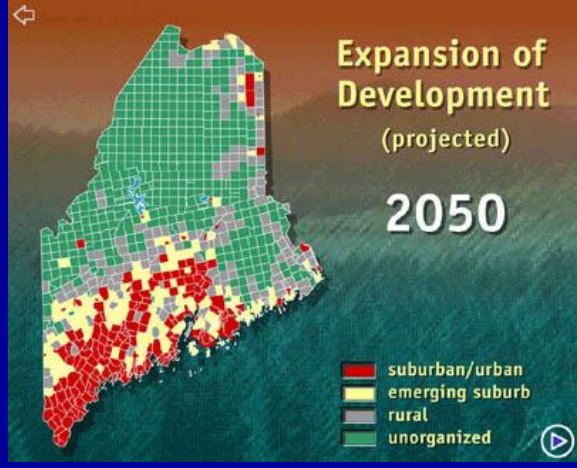


(1. KPMG 2013, 2. BOOZE ALLEN 2014)


"Like the resource it seeks to protect, wildlife conservation must be dynamic, changing as conditions change, seeking always to become more effective."
-Rachel Carson (1907-1964)







COASTAL COMMUNITY TRENDS

ME SPO

TRADITIONAL NATURAL RESOURCE BASES DEPLETED

DRAMATIC INCREASES IN PROPERTY VALUES AND TAXES

SIGNIFICANT DEMOGRAPHIC SHIFTS

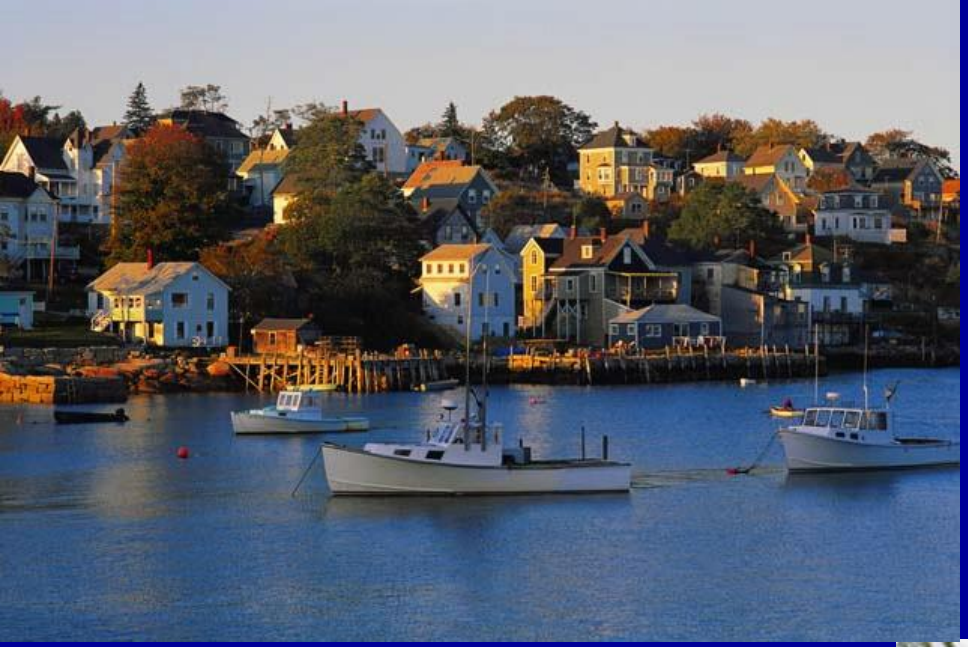
- **INCREASING % SENIORS**
- **INCREASING % "FROM AWAY"**

NON EXTRACTION RESOURCE USE BECOMES DOMINANT (LIFESTYLE/TOURISM)

NON-EXTRACTIVE RESOURCE USE SHIFTING FROM SUMMER ACTIVITY TO YEAR ROUND

REDUCTION AND DISPLACEMENT OF TRADITIONAL SOCIO-ECONOMIC GROUPS BASED ON EXTRACTIVE NATURAL RESOURCE EXPLOITATION

POLITICAL AND ECONOMIC DEVELOPMENT DRIVEN BY TOURISM, RESIDENTIAL DEVELOPMENT, AND RETIREES

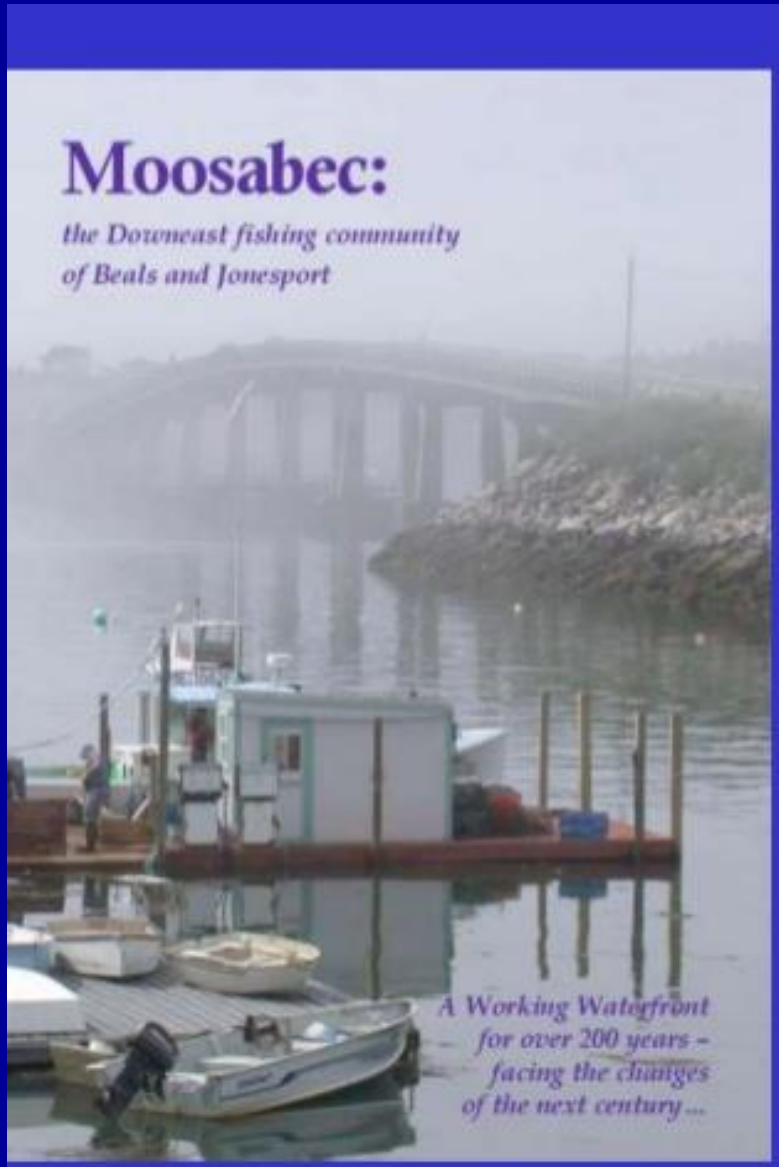


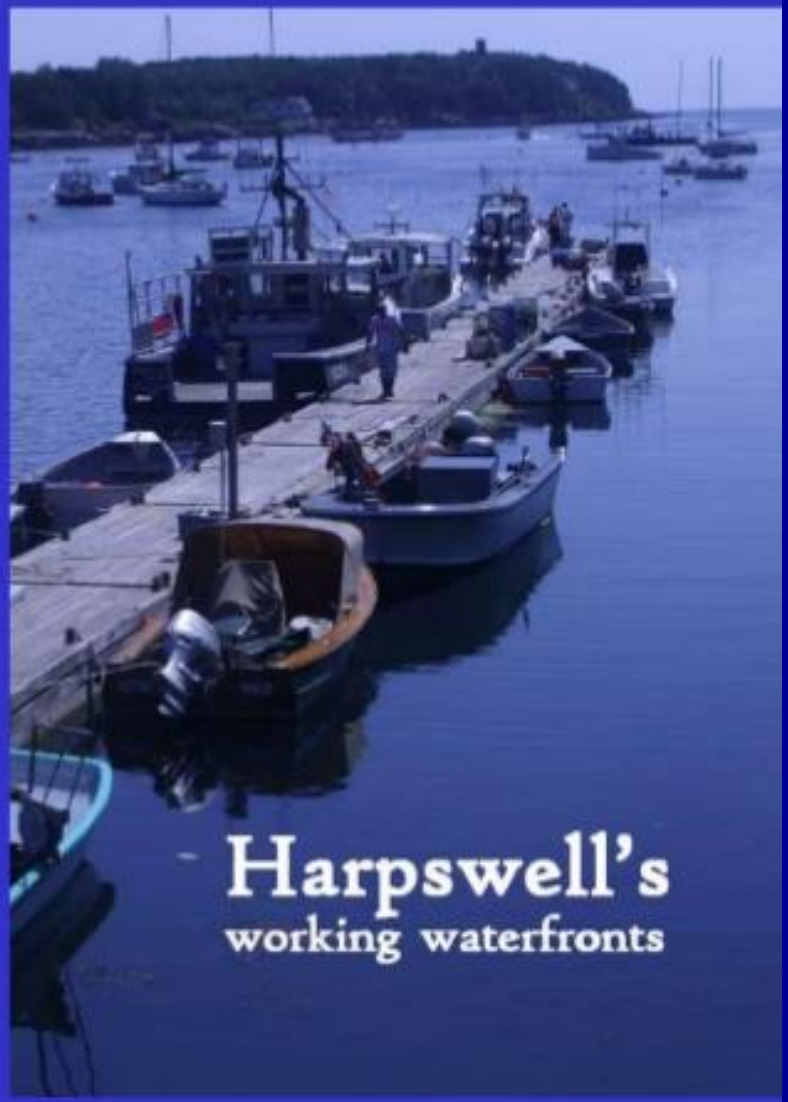
MIXED USE

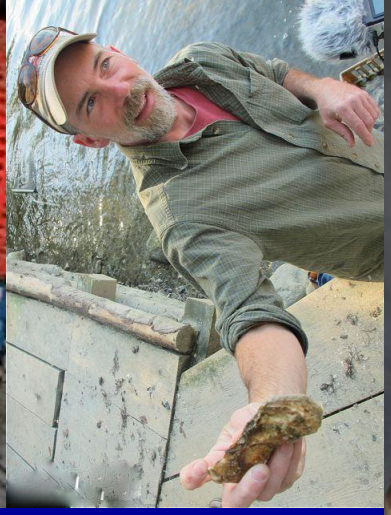
MCMANSIONS

Working Waterfront
COALITION

MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE


Moosabec:
the Downeast fishing community
of Beals and Jonesport
A Working Waterfront
for over 200 years –
facing the changes
of the next century...


Harpswell's
working waterfronts



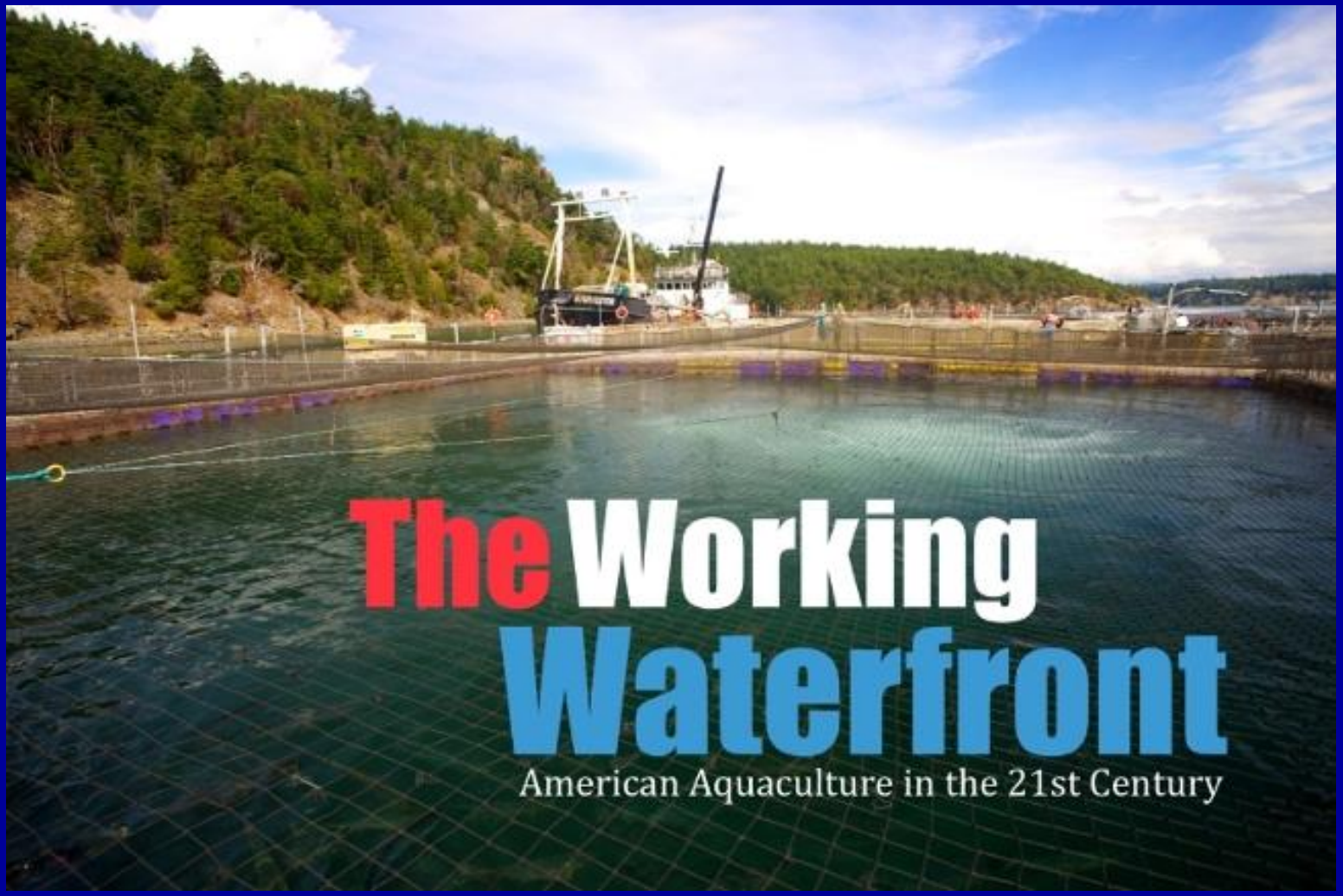
WE ARE MAINE AQUACULTURE

National Working Waterfronts & Waterways Symposium

In partnership with
Stem to Stern: Boating and Waterway Management in Florida

November 16-19, 2015 | Tampa, FL

MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE



MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE

MAINE
TOURISM
ASSOCIATION

Maine
VisitMaine.com

EXPERIENCE
MAINE
MARITIME

MAINE
Restaurant Association

MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE

DOWNEAST FISHERIES TRAIL

the Pemaquid Oyster Festival

MAINE OYSTER TRAIL

EASTPORT SALMON & SEAFOOD FESTIVAL

Eastport Salmon & Seafood Festival
Labor Day Weekend
August 30 & 31, 2014

MAINE SEAWEED FESTIVAL

MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE


860
CYR BUS


The RiverTripper
888-635-4309


The Pemaquid Oyster Festival

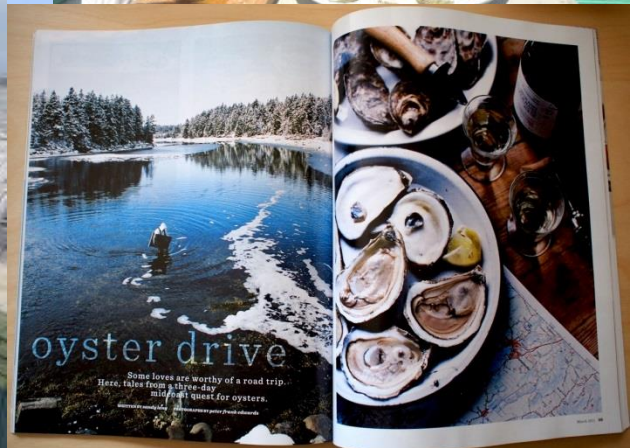

The River Tripper
Boat Rides
to the
Oyster Farms
@
12:30pm 3:30pm
1:30pm 4:30pm
2:30pm 5:30pm
$15 adults & $7 kids


OYSTER FARMS
1997
4 FARMS
500,000
OYSTERS
2013
65 FARMS
12 MILLION
OYSTERS


OYSTER
LADY


MAINE


Lincoln County
Magazine
Damariscotta
River Oysters
How they grow...
Where they go...
Miss Pig
Boothbay's calendar girl
greets motorists on Rt. 27
The Kraut's Out
September means a
new crop of sauerkraut
at Morses's in Waldoboro
Kid Stuff Crafts
Seashell candles...
A great summer project
Calendar
of Events


oyster drive
Some loves are worthy of a road trip.
Here, tales from a three-day
mid-coast quest for oysters.

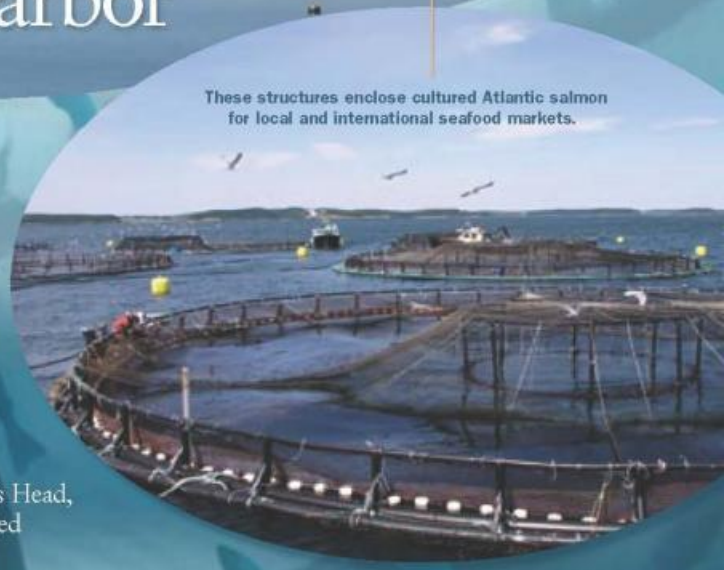



EXPERIENCE
MARITIME MAINE




MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE

Aquaculture's place in a working harbor



These structures enclose cultured Atlantic salmon for local and international seafood markets.

The changing marine industries of Broad Cove

Broad Cove has a 200-year history of marine industry that continues today in varied forms.

The circular installations floating on the water surface are enclosures for raising Atlantic salmon. Fish leap and splash while nets across the top keep out eagles, osprey, gulls, and cormorants. Young salmon are placed in the pens when they are about six inches long, and take about two years to grow to market size. Atlantic salmon is one of Maine's most economically significant seafood industries.

Beyond the salmon farm, tugboats and barges take advantage of Estes Head, the deepest natural port on the US East Coast. Shipments have included pulp and paper, cattle, and even wind turbines.

Maine
VisitMaine.com

Explore and learn more! Visit
DowneastFisheriesTrail.org

Look for fish slapping the water at feeding time to spot ACTIVE PENS. Between harvests, the pens rest—lying fallow.

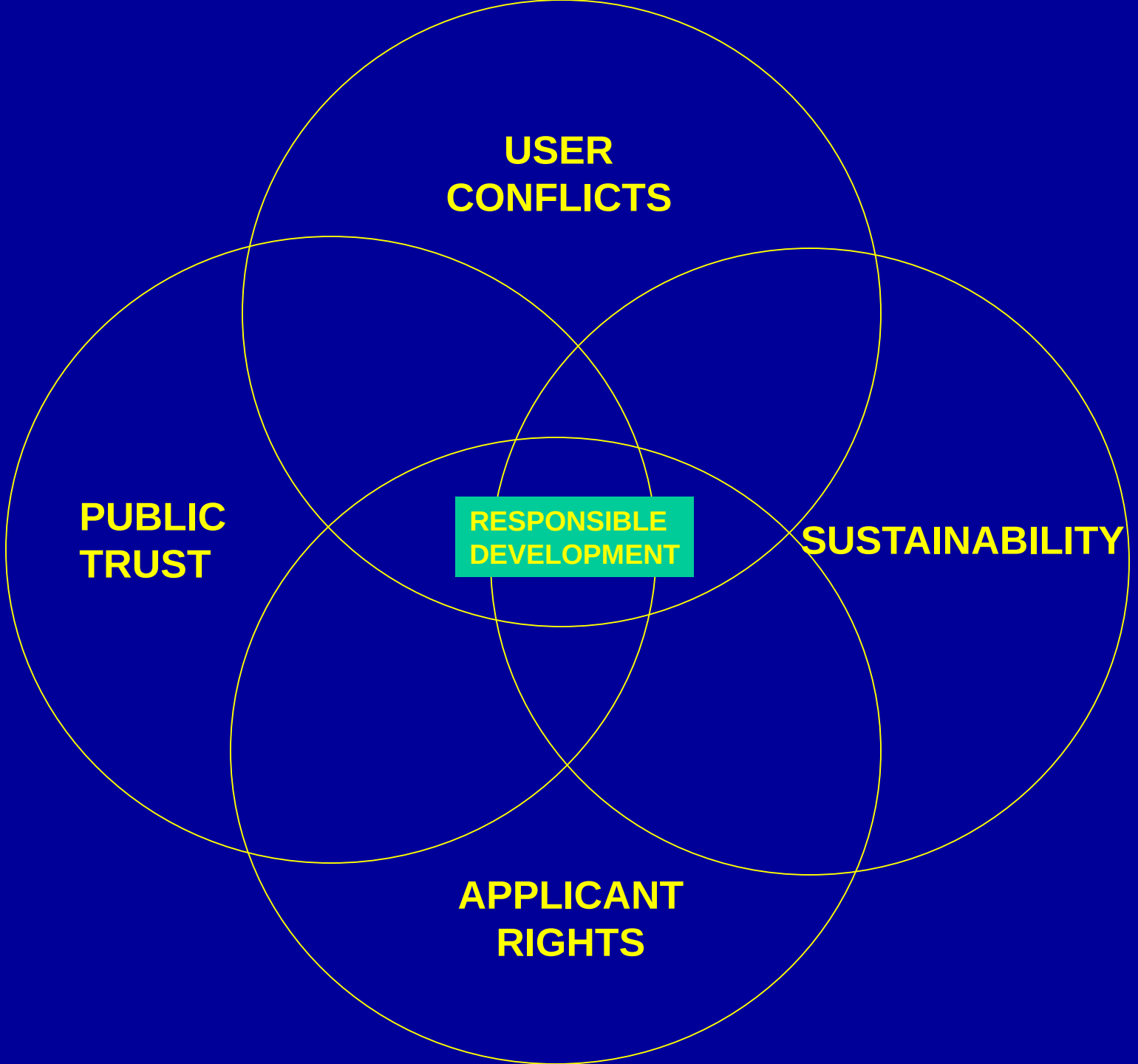
Photos: Downeast Aquaculture

Atlantic salmon take about TWO YEARS to grow to harvestable size.

Mature ATLANTIC SALMON ready for market.

KEY PARTNERSHIPS/ ALLIANCE CHALLENGES

FORTUNE 100 CEO SURVEY 2015


USER
CONFLICTS
PUBLIC
TRUST
RESPONSIBLE
DEVELOPMENT
SUSTAINABILITY
APPLICANT
RIGHTS

MAINEAQUACULTURE

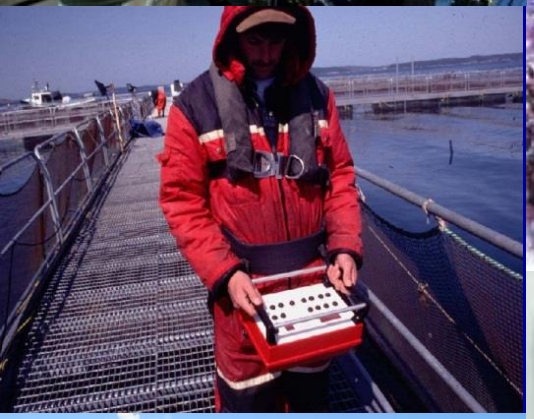
GROWING MAINES FUTURE

GOOD JOBS - RESPONSIBLE STEWARDSHIP - HEALTHY FOOD

PARTNERS (?) IN CONSERVATION

MAINE AQUACULTURE ASSOCIATION – GROWING MAINE'S FUTURE

MAINE AQUACULTURE
KEEPING WORKING WATERFRONTS WORKING



ARE YOU AN OPPONENT OF A COLLABORATOR?

Brad Hicks
Taplow Feeds
British Columbia, Canada

Global Assessment of Organic Contaminants in Farmed Salmon

Ronald A. Hites et al *Science* 09 Jan 2004:

Farmed salmon more toxic than wild salmon, study finds

Declining Wild Salmon Populations in Relation to Parasites from Farm Salmon

Martin Krkošek, et al

<u>If outbreaks continue, then local extinction is certain, and a 99% collapse in pink salmon population abundance is expected in four salmon generations [2014]</u>.
Science 318: 1772-1775 (2007)

Lethal Atlantic Virus found in Pacific Salmon

Morton 2011

The highly marine influenza virus, Infectious Salmon Anaemia (ISA) has for the first time been officially reported after being found in the Pacific on B.C.'s central coast.

NASCO

Atlantic Salmon Federation
Fédération du Saumon Atlantique

CNL(16)52

Closed containment: recent developments - costs and benefits

June 2016

BUT WHAT IF IT IS ALL BULLSHIT?

- *It can make a mess, is hard to clean up, but it does not have staying power.*
- *We hope.*

ETHICAL DELEMA for PEER REVIEWED SCIENCE

The unbearable asymmetry of bullshit

Quillette, February 18, 2016 | By Brian D Earp

Research Fellow in the Uehiro Centre for Practical Ethics at the University of Oxford

Bullshit in science

There is a veritable truckload of bullshit in science.[1] When I say bullshit, I mean arguments, data, publications, or even the official policies of scientific organizations that give every impression of being perfectly reasonable — of being well-supported by the highest quality of evidence, and so forth — but which don't hold up when you scrutinize the details. Bullshit has the veneer of truth-like plausibility. It looks good. It sounds right. But when you get right down to it, it stinks.

HOW IT WORKS

Gish Gallop

The term “Gish Gallop” was coined by the science educator Eugenie Scott in the 1990s to describe the debating strategy of one Duane Gish.

To “**spew forth torrents of error** that the evolutionist hasn’t a prayer of refuting in the format of a debate.” It also referred to Gish’s apparent tendency to simply **ignore objections raised by his opponents**.

MONEY NOT LIMITING FACTOR

Gordon and Betty Moore Foundation	$92,000,000
Packard Foundation and Hewlett Foundation	$90,000,000
Pew Charitable Trust	$82,000,000
TOTAL	$264,000,000

Vivian Krause

NOTABLE PROJECTS SUPPORTED

Atlantic Salmon Federation
David Suzuki Foundation
Tides Canada
WWF
Living Oceans
Coastal Center for Aquaculture Reform
Middle Bay Sustainable Aquaculture Institute
Aquaseed
Watershed Watch
T. Buck Suzuki Foundation
Marine protected areas CPWS
Conservation Fund
Earthlife Canada Foundation
Ecotrust Canada
Wild Salmon Center
National Environmental Trust
Pembina Foundation
Rainforest Conservation Foundation
Round River Conservation Foundation
Sierra Club of BC Foundation

Gordon and Betty Moore Foundation

$3,600,000 for “Demarketing” Farmed Salmon

SeaWeb $560,000

“to provide a high-quality tool-kit and co-ordination infrastructure for use by ENGO’s in their campaigns to shift consumer and retailer demand away from farmed salmon”

PCB - BS

Global Assessment of Organic Contaminants in Farmed Salmon

Ronald A. Hites et al

Science 09 Jan 2004:
Vol. 303, Issue 5655, pp. 226-229
DOI: 10.1126/science.1091447

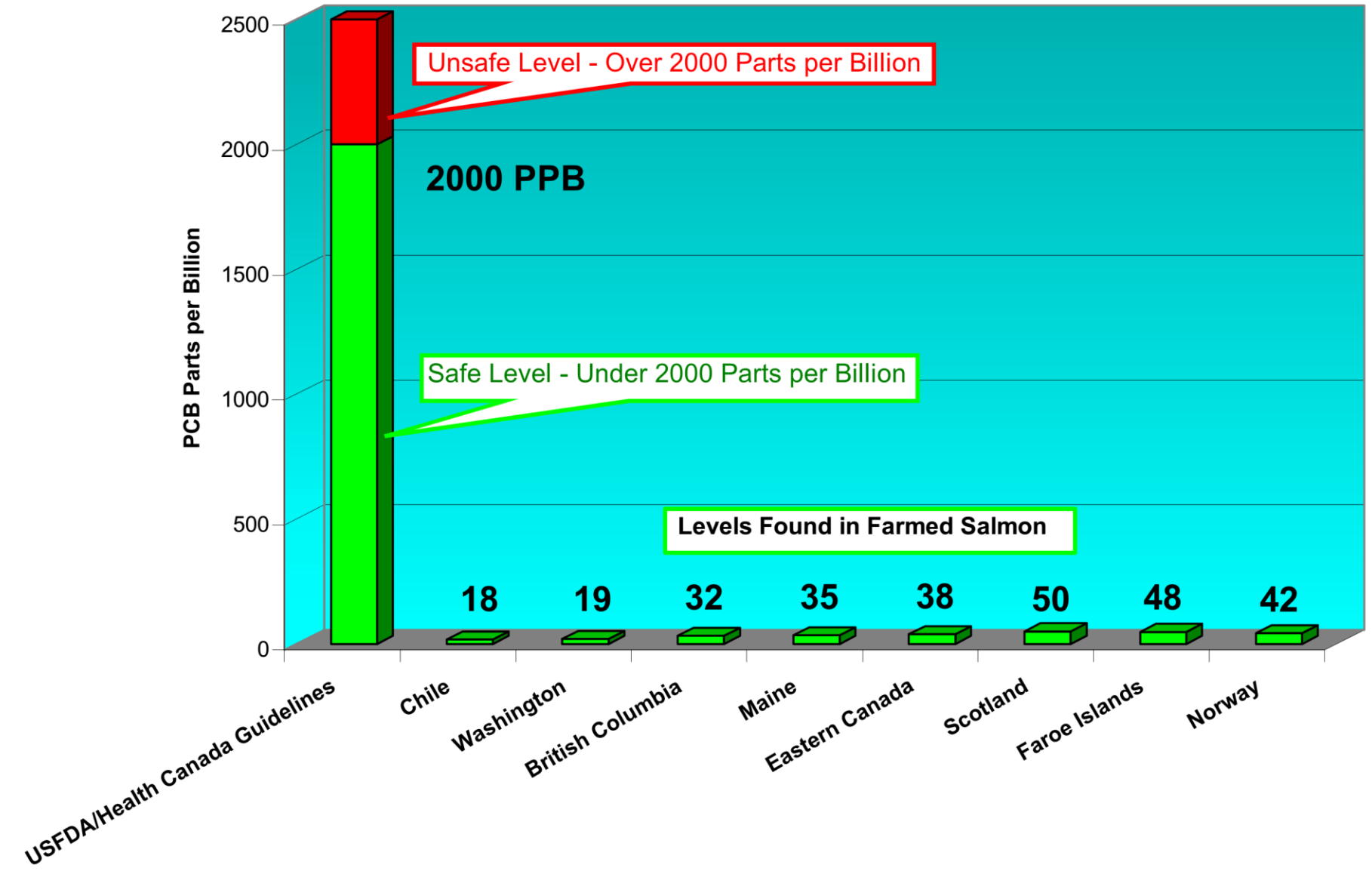
Farmed salmon more toxic than wild salmon, study finds

FOR IMMEDIATE RELEASE

Jan. 8, 2004

BLOOMINGTON, Ind. -- A study of more than two metric tons of North American, South American and European salmon has shown that PCBs and other environmental toxins are present at higher levels in farm-raised salmon than in their wild counterparts

Comparison of US Food and Drug Administration and Health Canada Guidelines for PCBs in Food Compared to Levels Found in Farmed Salmon (Science, Vol. 303, Jan. 9, 2004)



LICE - BS

Declining Wild Salmon Populations in Relation to Parasites from Farm Salmon

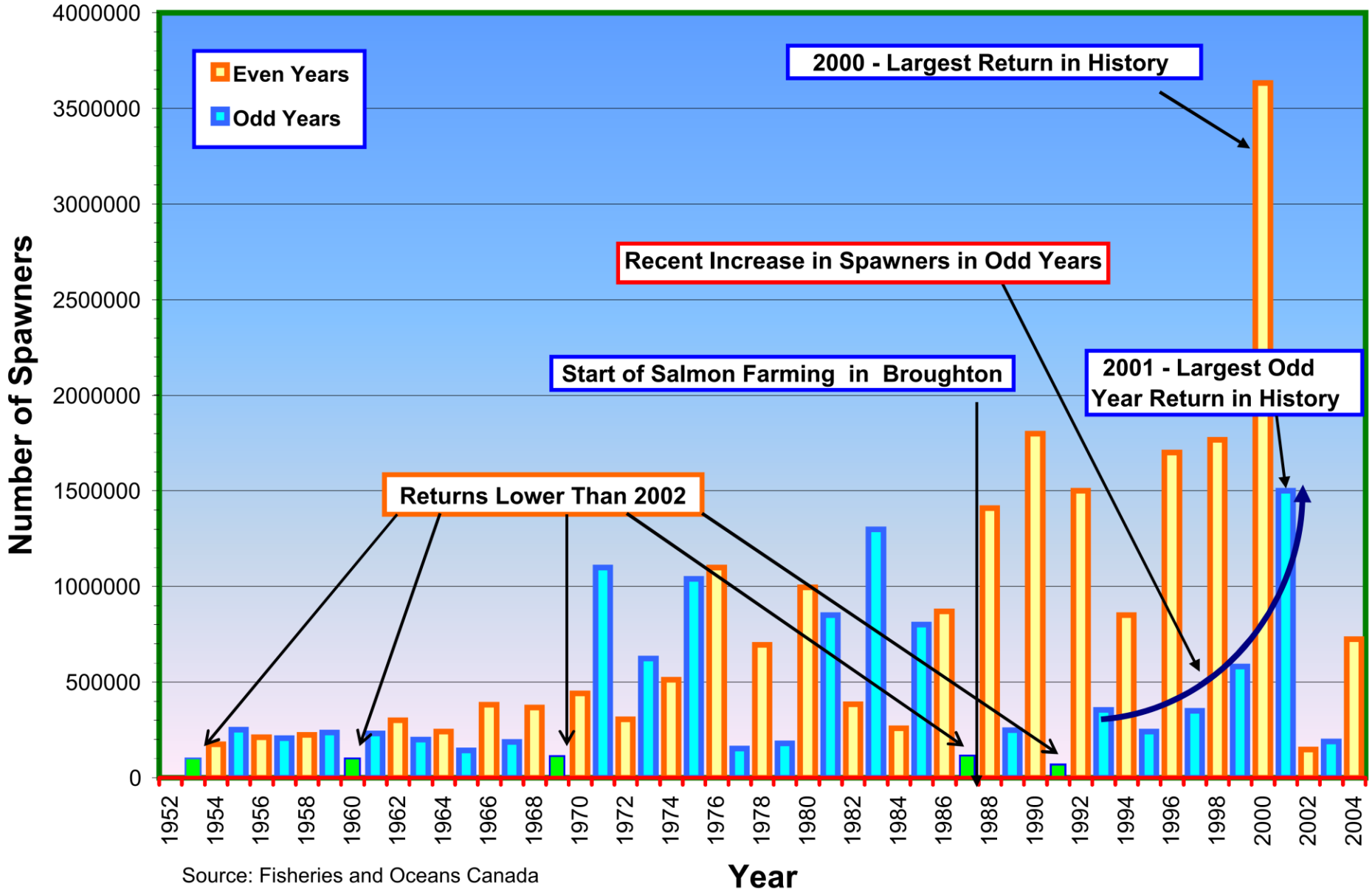
Martin Krkošek, Jennifer S. Ford, Alexandra Morton, Subhash Lele, Ransom A. Myers, Mark A. Lewis

The louse-induced mortality of pink salmon is commonly over 80% and exceeds previous fishing mortality. **If outbreaks continue, then local extinction is certain, and a 99% collapse in pink salmon population abundance is expected in four salmon generations [2014]**. These results suggest that salmon farms can cause parasite outbreaks that erode the capacity of a coastal ecosystem to support wild salmon populations.

Science 318: 1772-1775 (2007)

Escapes of major pink salmon producing rivers in the Broughton Area		
River	**2006**	**2014**
Glendale River	182,000	334,000
Kakweiken River	75,000	734,000
Ahnuhati River	10,800	23,000
Kingcome River	3,400	2,700
Wakeman River	14,700	14,000
TOTAL	285,900	1,107,700 (EXTINCT ?)

Pink Salmon Spawners in Broughton Mainland Inlets (Area 12)



Source: Fisheries and Oceans Canada

Sea lice outbreak shows no links to salmon farming in B.C.: report

Prevalence in juvenile wild salmon hit five-year high in 2015

BY RANDY SHORE, VANCOUVER SUN FEBRUARY 18, 2016

ISA - BS

Lethal Atlantic Virus found in Pacific Salmon

October 17, 2011

Contact:
Alexandra Morton, 250.974.7086 (cell)
Rick Routledge, 778.782.4478; 604.329.8712 (cell); richard_routledge@sfu.ca
Marianne Meadahl, PAMR, 778.782.3210 (o); 604.209.5770 (c); marianne_meadahl@sfu.ca

The highly contagious marine influenza virus, Infectious Salmon Anaemia (ISA) has for the first time been officially reported after being found in the Pacific on B.C.'s central coast.

Salmon-Killing Virus Seen for First Time in the Wild on the Pacific Coast

Cornelia Dean and RACHEL NUWER

Published: October 17, 2011

New York Times

COMMENTARY

By Brad Hicks

Squashing the ISA lie...

Those who have been following the prolonged saga of the possible presence of Infectious Salmon Anemia (ISA) in western waters can be comforted that the virus has not been found in the eastern north Pacific Ocean.

CREATION OF A CRISIS

This story begins in October 2011 when a researcher and associate (Dr. R. Routledge and Ms A. Morton) at Simon Fraser University (SFU) issued a press release indicating that ISAv has been found among wild pacific salmon in British Columbia. As it turns out this press release was based on preliminary results which could never be confirmed using a complete testing regime or indeed by other laboratories. Both confirmation and corroboration are required for a definitive diagnosis. It is now widely understood that this initial finding was a false result.

To add fuel to the fire, Senators Maria Cantwell, (D. Wash.), Lisa Murkowski, (R. Alaska) and Mark Begich, (D. Alaska), signed a letter calling for the US to do its own testing rather than trusting the Canadian government scientists. "We should not rely on another government – particularly one that has a motive to misrepresent its finding."

As one could imagine this inflammatory language sparked a diplomatic row and set both the Canadian and the US governments into frenetic action. The outcome was that a lot of money was going to be rapidly spent in an avalanche of activity to investigate whether or not this virus might be present in the Pacific Northwest, BC and Alaska.

BACK TO BASICS

In Canada the Canadian Food Inspection Agency

questionable lab results and then using the vast machinery of the university's public relations department to create an artificial crisis. This crisis is further exacerbated by agitating the politicians into a panicked frenzy. And the

Lab that found virus in B.C. salmon stripped of credentials after audit

ANDREA WOO VANCOUVER

The Globe and Mail
Published Wednesday, Jul. 03, 2013

NASCO

Atlantic Salmon Federation
Fédération du Saumon Atlantique

CNL(16)52

Closed containment: recent developments - costs and benefits

June 2016

Atlantic Salmon Federation
Fédération du Saumon Atlantique

The conclusions of this is that RAS technology for land based salmon grow-out is available, i.e. systems where it is technical possible to … produce a high quality salmon in RAS.

Economic profiles of commercial scale operations will soon be available via publicly supported projects such as that being built by the Namgis First Nation on Vancouver Island

REVIEW

Land-based salmon still not investor-ready

An analysis of the BC-based "Kuterra" project confirms that there is currently no sound business case to be made for rearing market-size Atlantic salmon on land.

BY BRAD HICKS

Conclusion (Earp paper)

“The amount of energy necessary to refute bullshit is an order of magnitude bigger than to produce it.”

“This is the unbearable asymmetry of bullshit I mentioned in my title, and it poses a serious problem for research integrity. Developing a strategy for overcoming it, should be a top priority for publication ethics.”

BS has been published by the anti-fish farming community in Canada as the primary driver for the development of public policy in Canada and for the regulation of fish farms and this is also unbearable.

NOT BS

THANK YOU

June 27th press release [34]

Mr. Staniford says that the press releases were published to professional journalists and that the words in the June 27th press release, namely “scam”, “liar” and “consumer fraud” are merely hooks or arresting leads. Those words are clearly capable of a defamatory meaning.

Gordon and Betty Moore Foundation

$3,600,000 for “Demarketing” Farmed Salmon

SeaWeb $560,000

“to provide a high-quality tool-kit and co-ordination infrastructure for use by ENGO’s in their campaigns to shift consumer and retailer demand away from farmed salmon”

Kibenge *et al. Virology Journal* (2016) 13:3
DOI 10.1186/s12985-015-0459-1

Virology Journal

RESEARCH Open Access

Discovery of variant infectious salmon anaemia virus (ISAV) of European genotype in British Columbia, Canada

Molly JT Kibenge[1], Tokinori Iwamoto[1,5], Yingwei Wang[2], Alexandra Morton[3], Richard Routledge[4] and Frederick SB Kibenge[1*]

Source: http://moore.org/grants-awarded.aspx

Gordon and Betty Moore Foundation

$3,600,000 for “Demarketing” Farmed Salmon

Living Oceans $1,578,000

Educate major buyers of farmed fish. ($453,000)

Agreement of at least one major salmon producer, from BC to plan for transitioning aquaculture industry to more sustainable practices, with government support [Land based, RAS, closed containment] ($1,250,000)

Gordon and Betty Moore Foundation

$3,600,000 for “Demarketing” Farmed Salmon

National Environmental Trust
Salmon Aquaculture Project $1,525,000

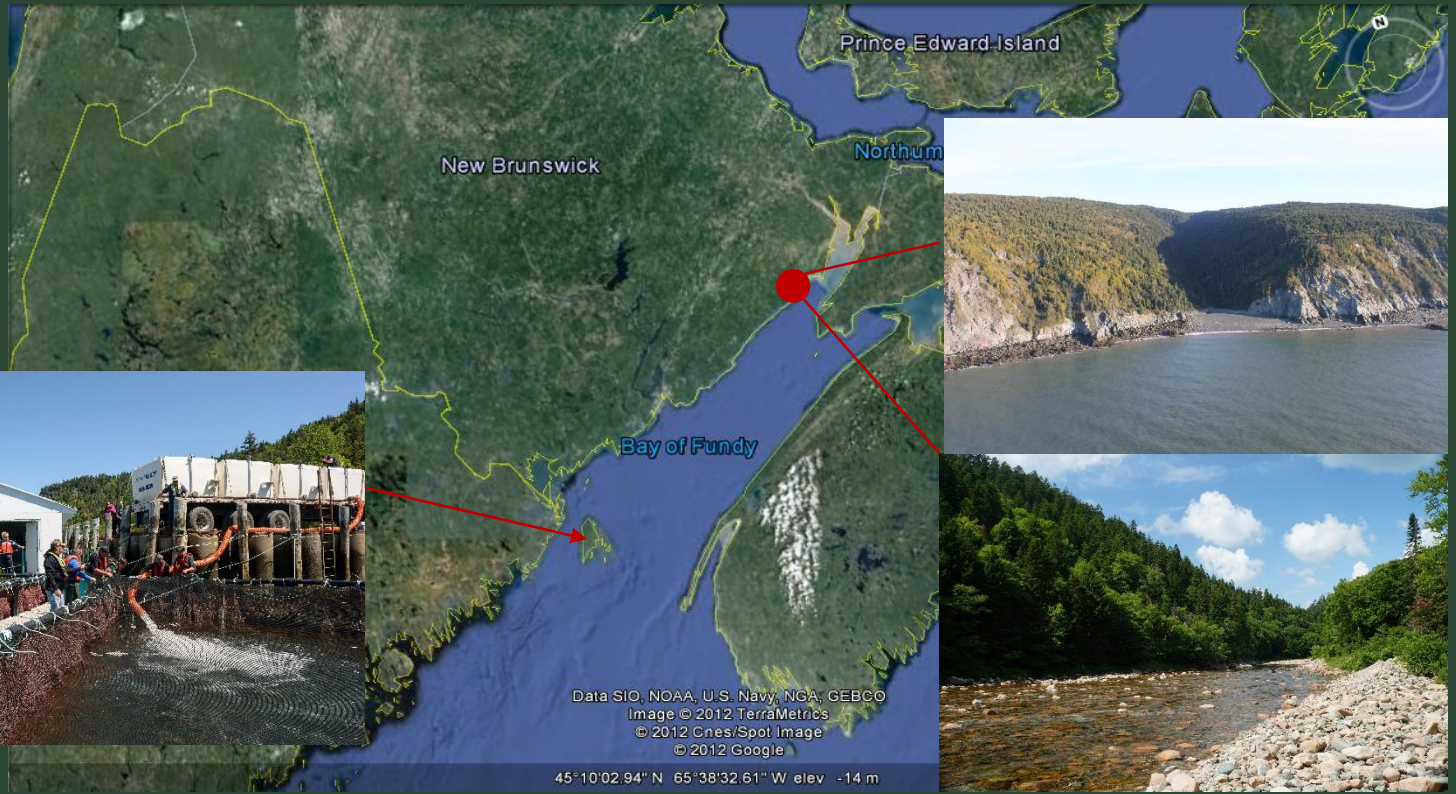
Adaptation of farmed-salmon purchasing standards which require suppliers to mitigate threats to wild fish and the environment, by major retailer and adaptation of policies consistent with codification of these market-driven reforms by regulators

Collaborative Salmon Recovery in Fundy National Park

Corey Clarke, Ecologist, Project Coordinator
Fundy National Park

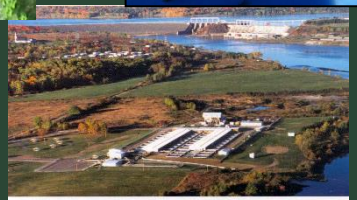
FUNDY SALMON RECOVERY

IBoF Salmon
Fundy National Park
Wild Salmon Conservation Farm



- 2003-’16 Live Gene Banking avoids extirpation
- Adults still fail to return from sea
- Smolt traits differ by release strategy

Collect remnant wild fish as Smolt



DFO LGB

Captive rear to adult

3 release strategies

Strategies produced different smolts,
Did that matter over a salmon life-time?
We partnered with industry to examine

2009-12
Reared wild smolts in
custom industrial marine pens

Animal Conservation

Animal Conservation. Print ISSN 1367

Lifelong and carry-over effects of early captive exposure in a recovery program for Atlantic salmon (*Salmo salar*)

C. N. Clarke[1]*, D. J. Fraser[2] & C. F. Purchase[3]

1 Fish Evolutionary Ecology Research Group, Environmental Science Graduate Program, Memorial University, St. John's, NL, Canada
2 Department of Biology, Concordia University, Montreal, QC, Canada
3 Department of Biology, Fish Evolutionary Ecology Research Group, Memorial University, St. John's, NL, Canada

Keywords
Atlantic salmon; captive breeding; domestication effects; early exposure; phenotypic plasticity; transgenerational.

Abstract

A full life cycle understanding of how different captive rearing strategies
wild fitness is needed for many species of conservation concern. Over th
cycle of endangered Atlantic salmon, we measured effects on wild fitness res

Less captivity = More wild fitness

FNP re-focused to Adult release = offspring with NO captive exposure
Massive industry capacity >10M Adult Salmon/yr

Results Cont…
20yr-high salmon counts in 2012
Produced 2015 smolt 5x more than expected

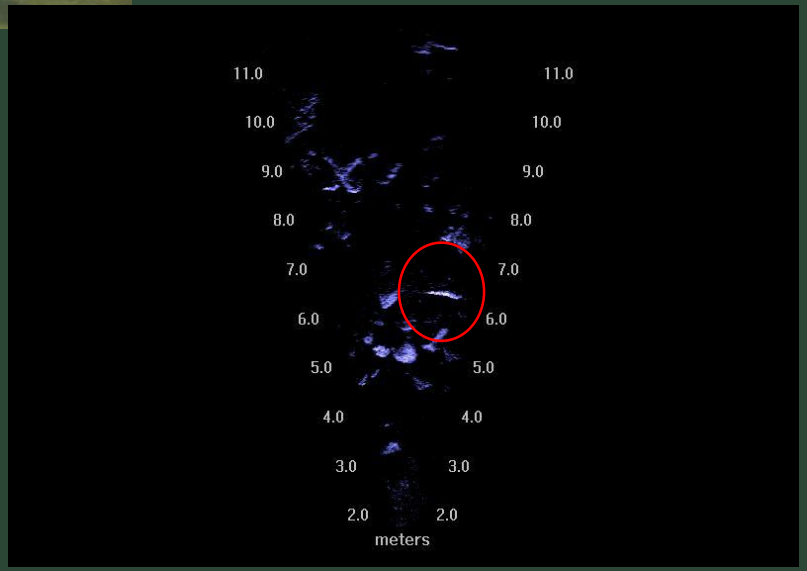
Worlds 1st endangered salmon marine farm
Operated by Cooke Aqua
Dark Harbour, Grand Manan

-Wild smolt reared in custom pens
-Released back to native rivers as adults
-Offspring free of captive exposure

Collaboration producing more than salmon:

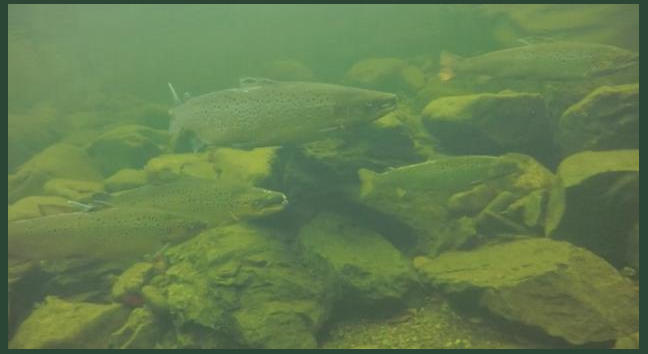
Public connections

Ecological and Conservation Knowledge (Sonar fish detection UNB)



Innovation in conservation methods

Collaboration producing more than salmon:



Multi-award winning

7 Agency Salmon Protection Coalition

Restoration of Salmon outside FNP

Public and media engagement

Diverse and practical collaborations sustainable programs

- Atlantic Canada Fish Farmers Association
- Fort Folly First Nation
- Department of Fisheries and Oceans
- Cooke Aquaculture
- New Brunswick (DAAF & DNR)
- University of New Brunswick
- Huntsman Ocean Science Centre

What Goes Around Comes Around, Restoring the Upper Salmon River

Kurt M. Samways

University of New Brunswick, Canadian Rivers Institute

Aquaculture Research, Science and Technology Forum October 26 and 27, 2016

Huntsman Fundy Discovery Centre, St. Andrews, NB

“Alone we can do so little; together we can do so much.”

- Hellen Keller

Parks Canada Parcs Canada

acffa Atlantic Canada Fish Farmers Association

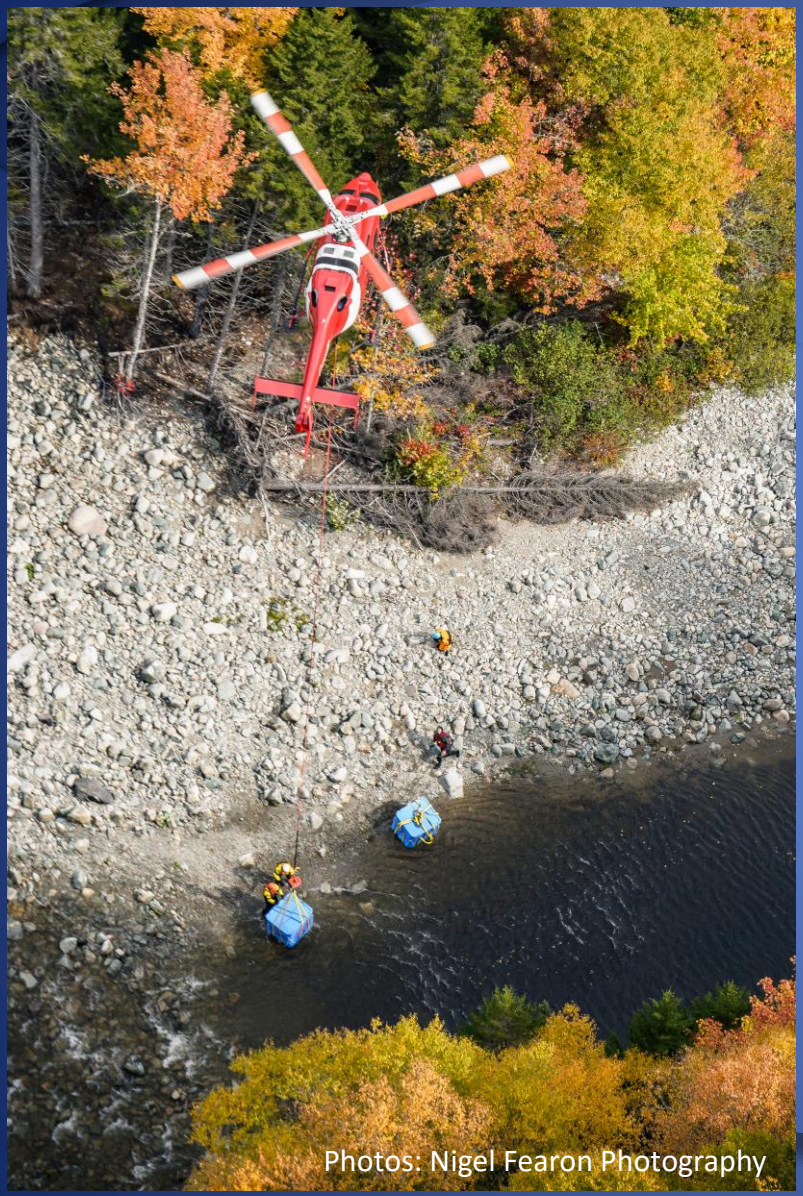
Cooke AQUACULTURE Inc.

Pêches et Océans Canada Fisheries and Oceans Canada

Inner Bay of Fundy Atlantic Salmon Fort Folly Habitat Recovery

The Atlantic Salmon CONSERVATION FOUNDATION

NB Wildlife Trust Fund Fonds de Fiducie de la Faune du N.-B.



Photos: Nigel Fearon Photography

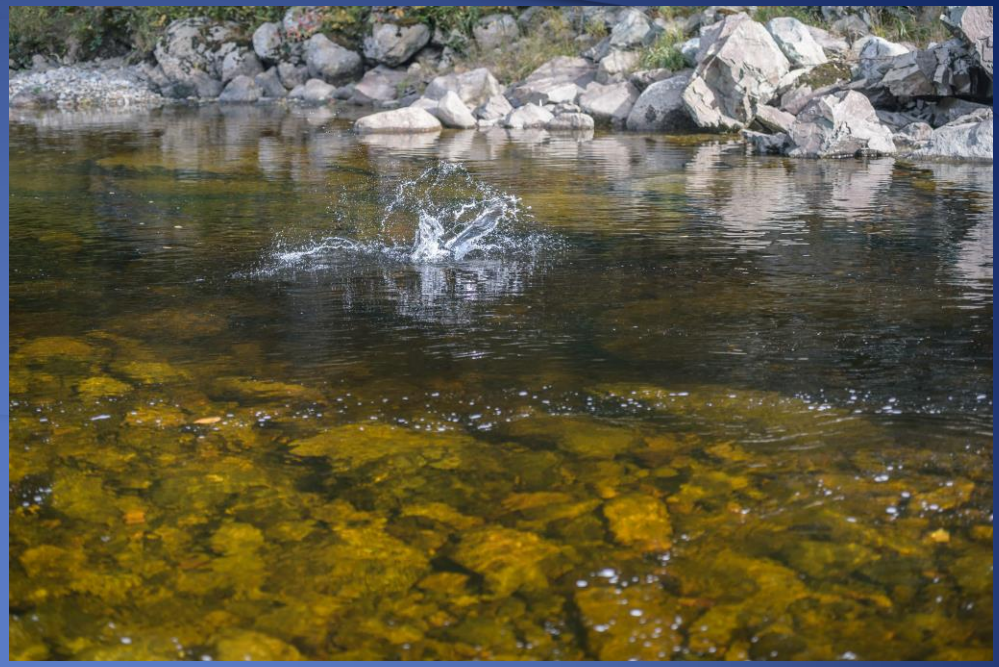
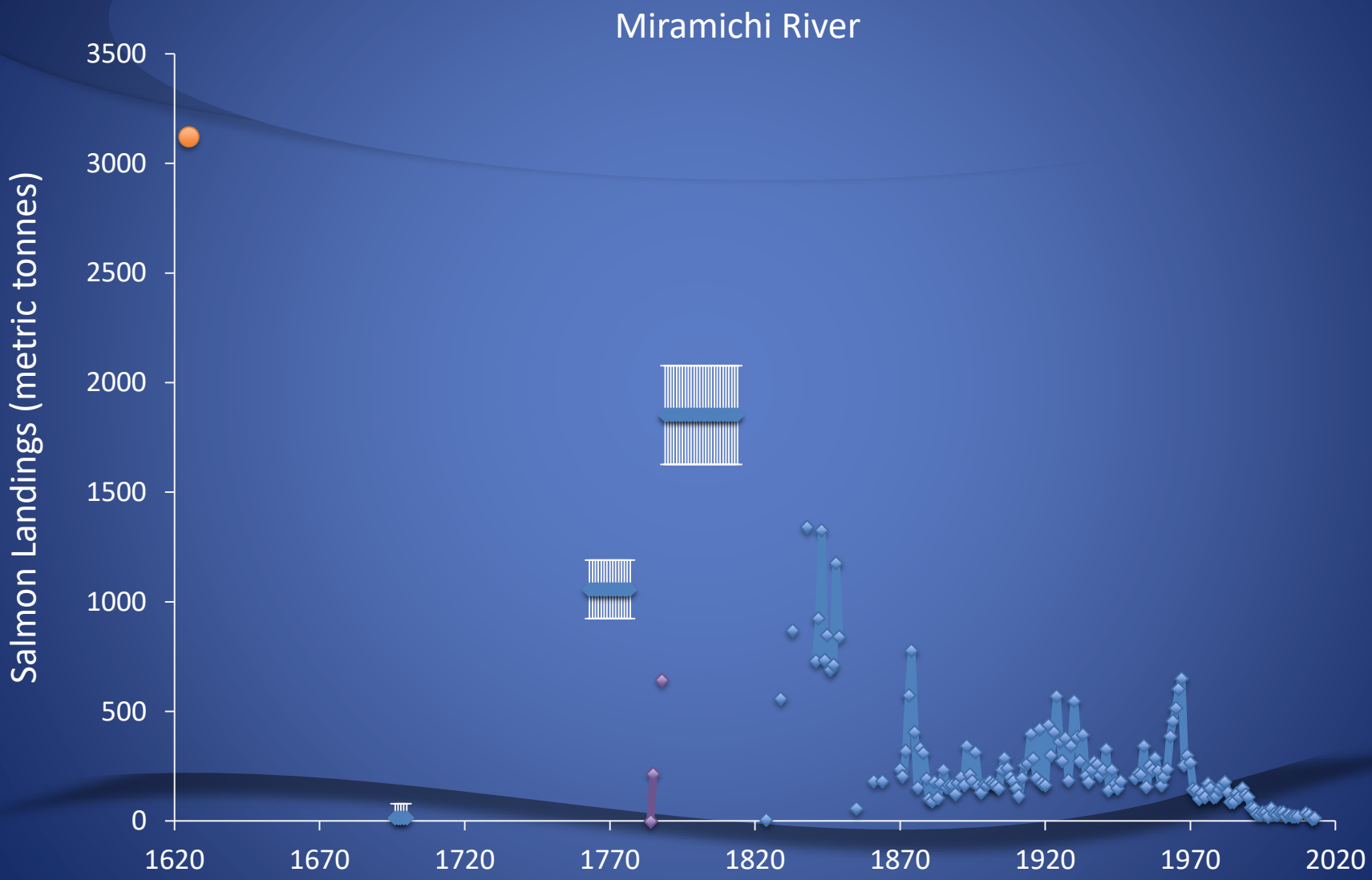


Photo: Emily Corey

Why are Atlantic salmon important?

Photo: Brittany Graham

Marine-nutrient Inputs to Atlantic Rivers



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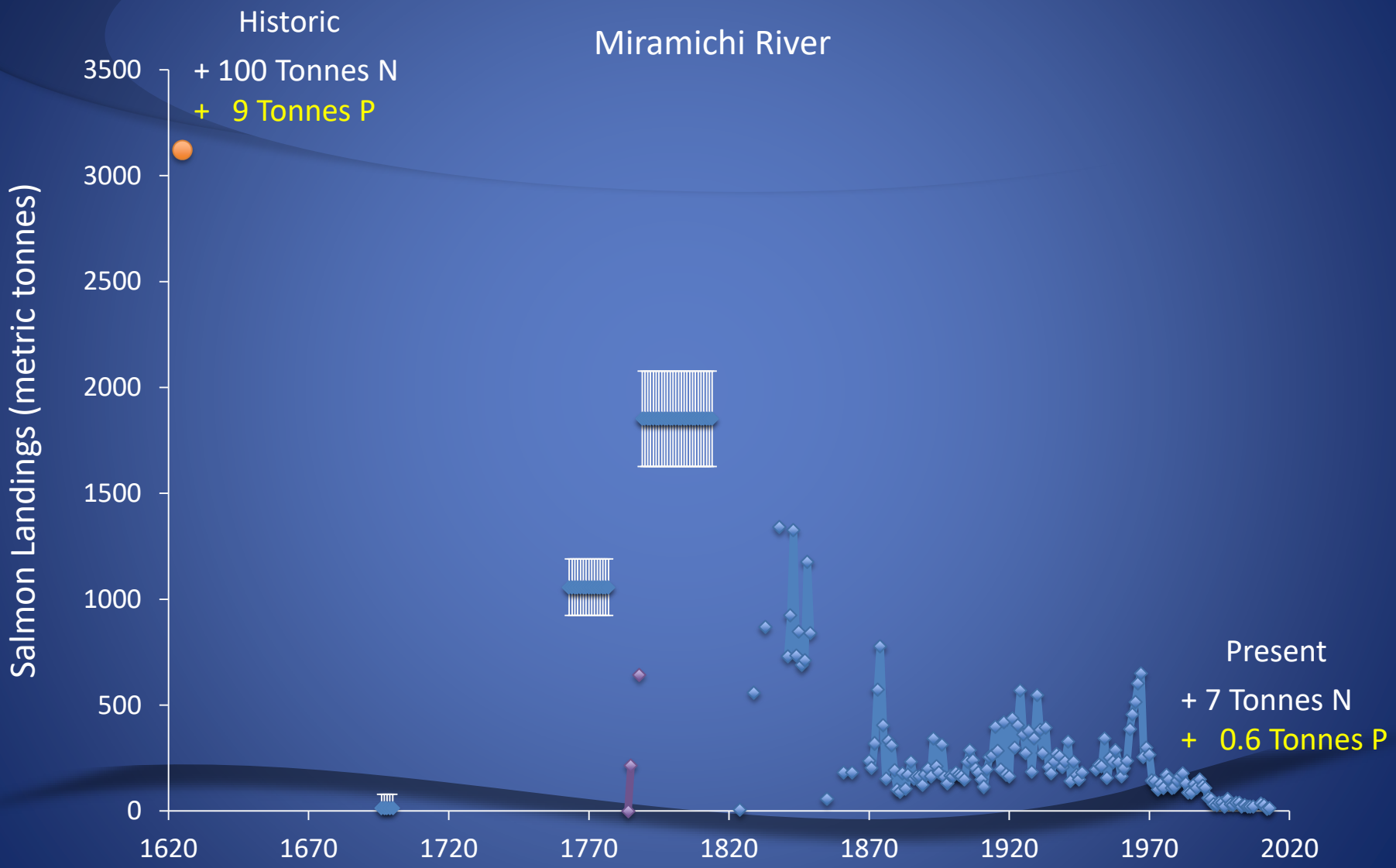
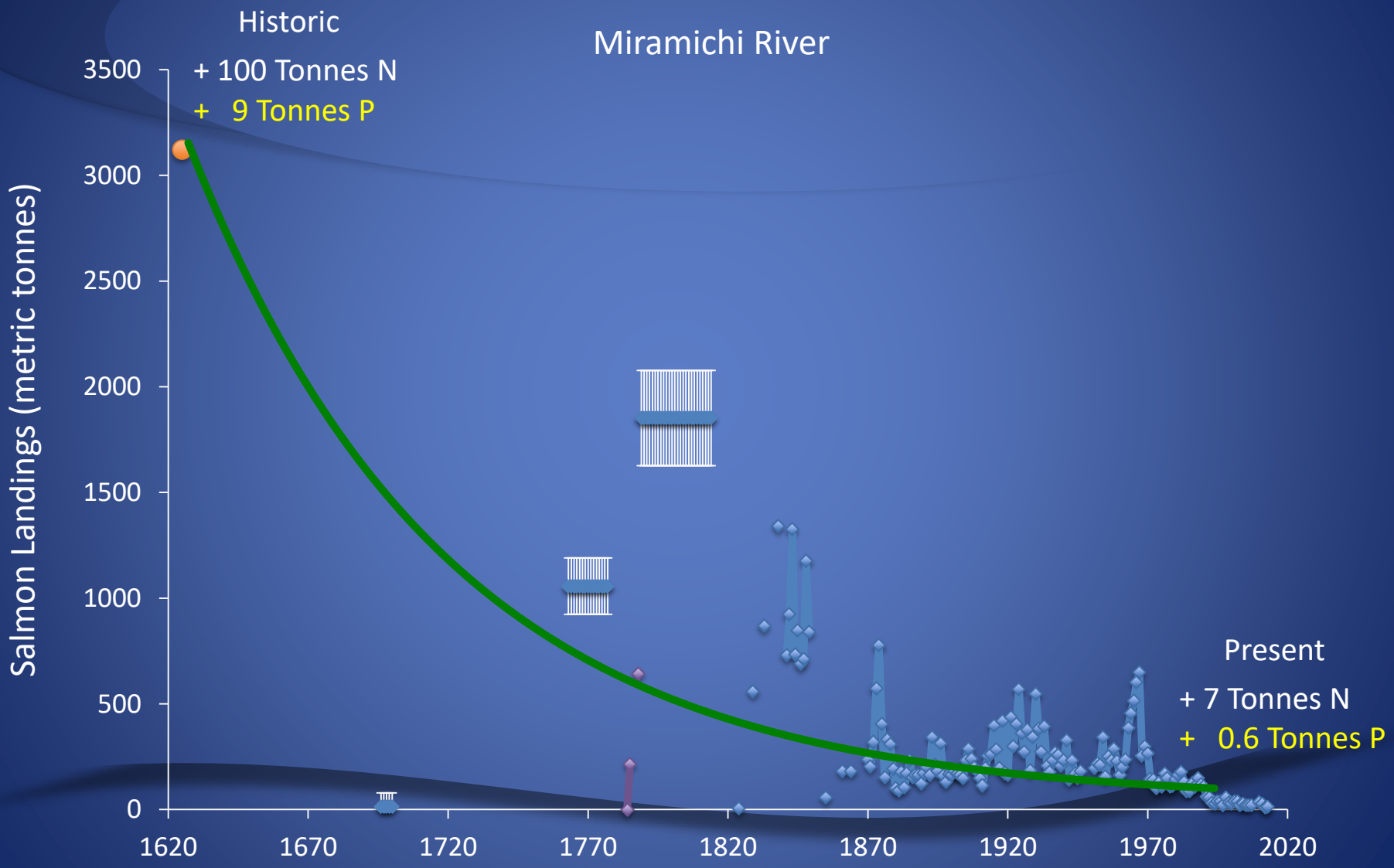


Marine-nutrient Inputs to Atlantic Rivers



Can river ecosystem health increase from adult supplementation?

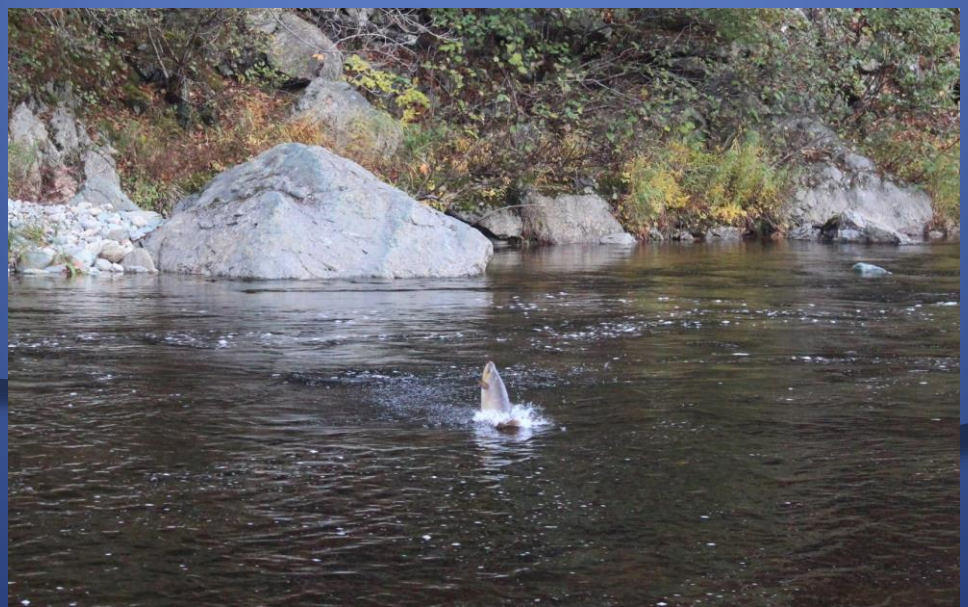
Photo: Michelle Charest

How does the Freshwater Community Respond to Adult Supplementation?

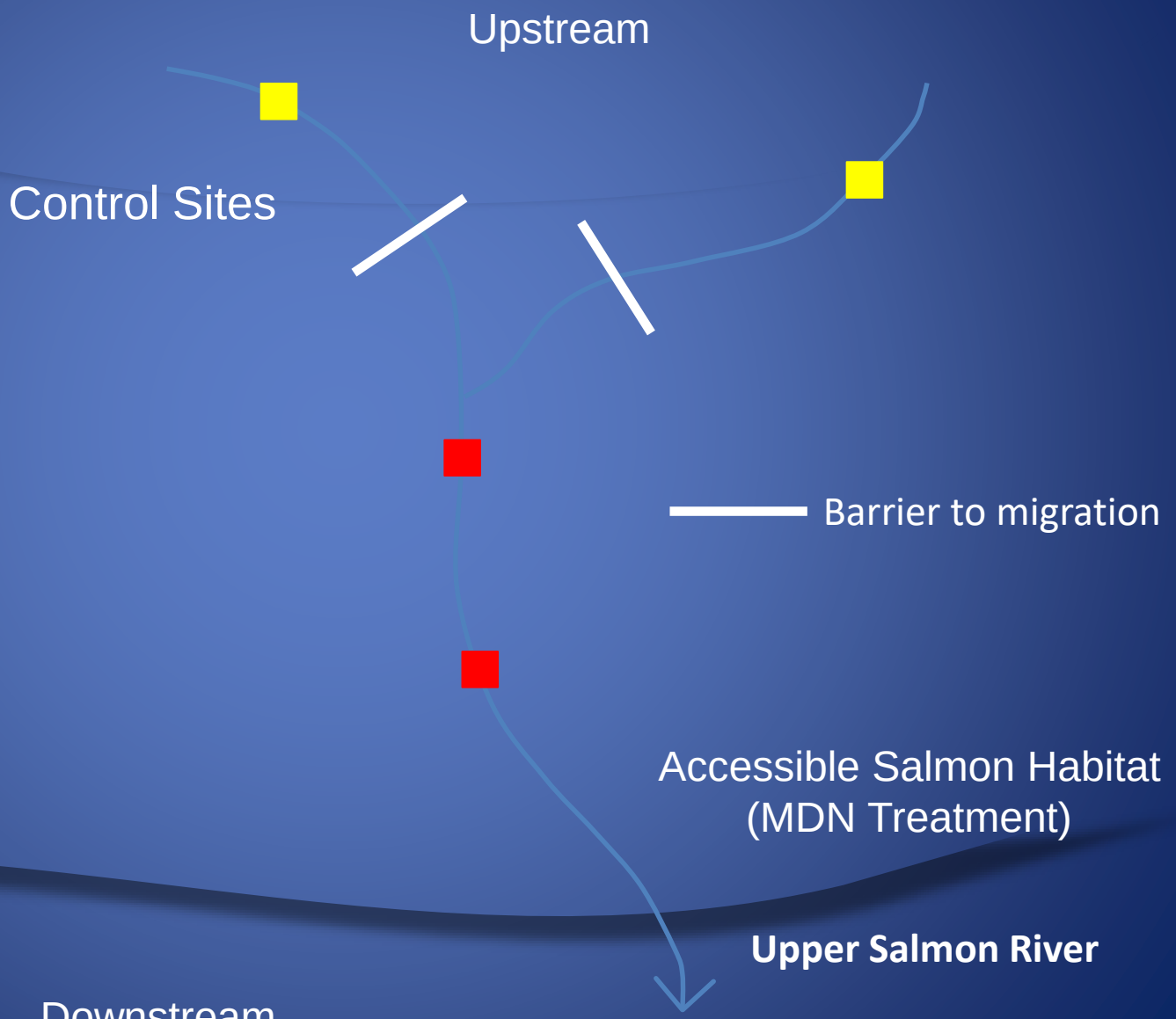
i. Sources of nutrients

i. Changes in primary productivity

i. Behaviour of cage-reared adult salmon.



Experimental Design



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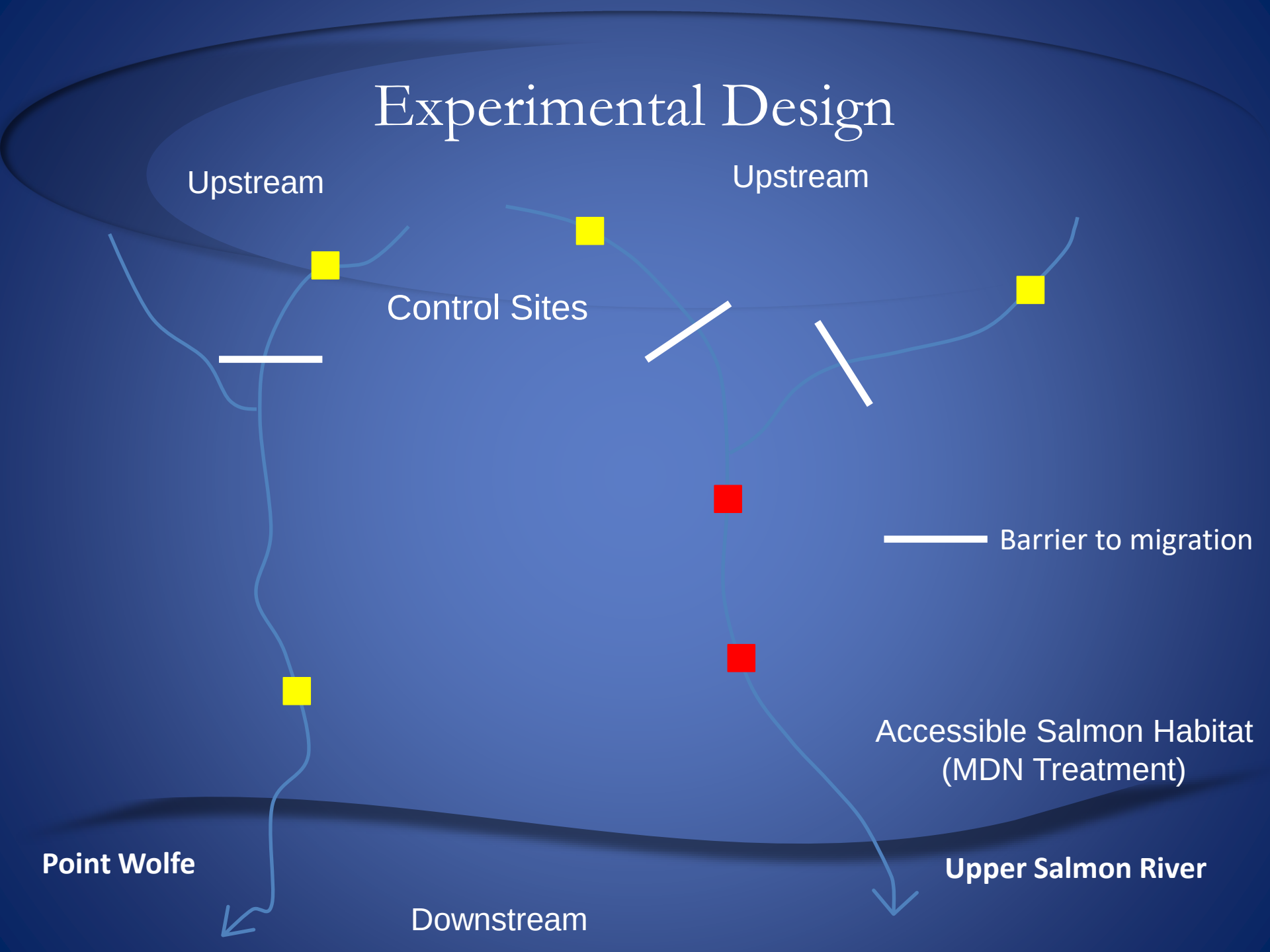

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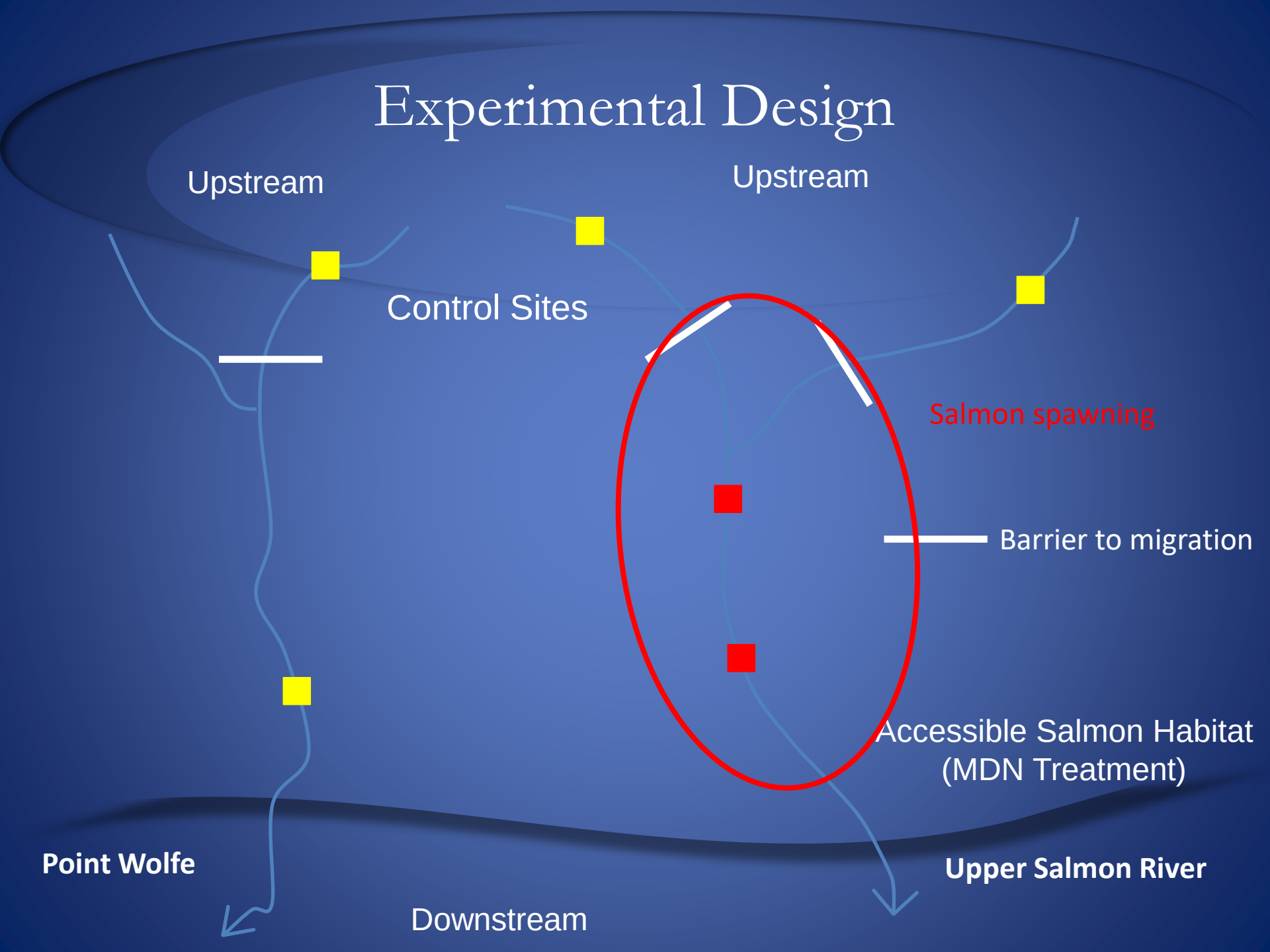


Adult Salmon in Rivers

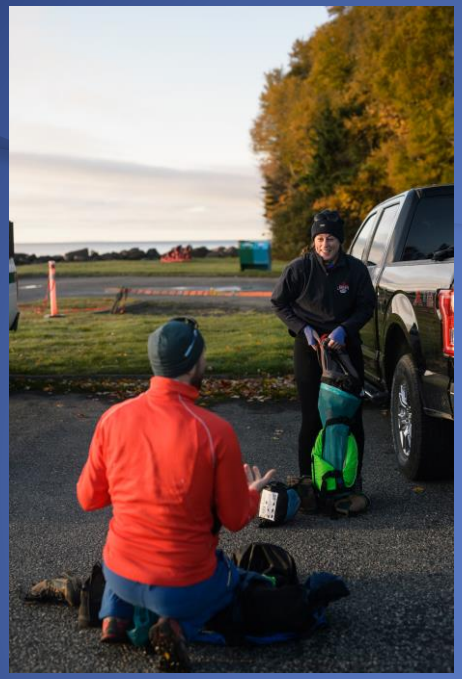
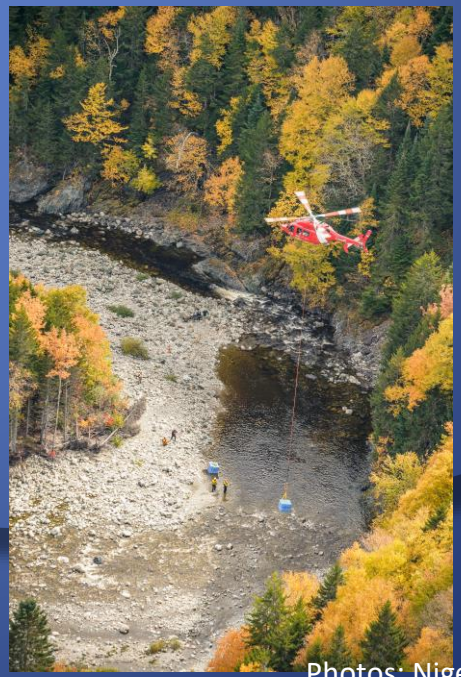
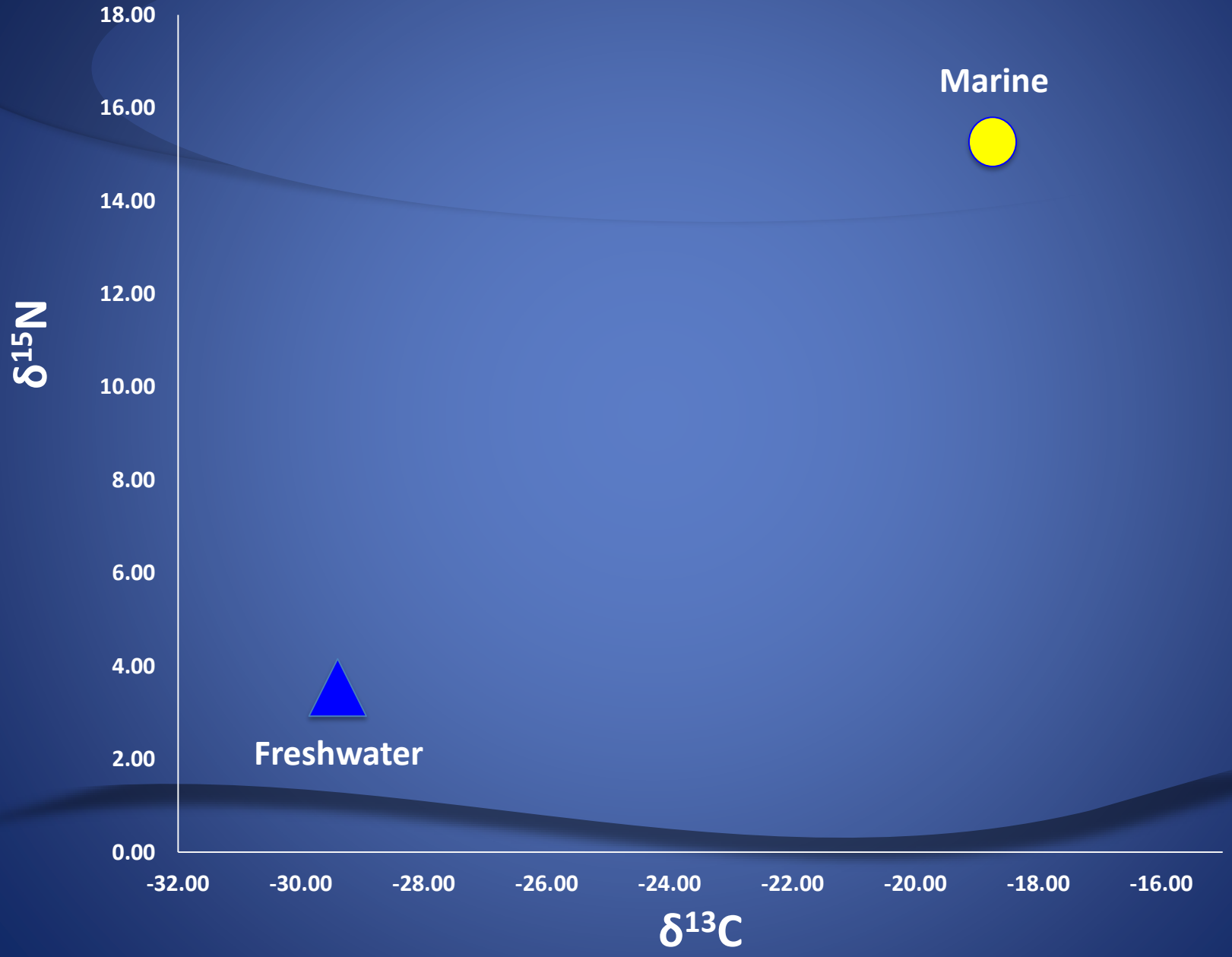


Photo: Emily Corey

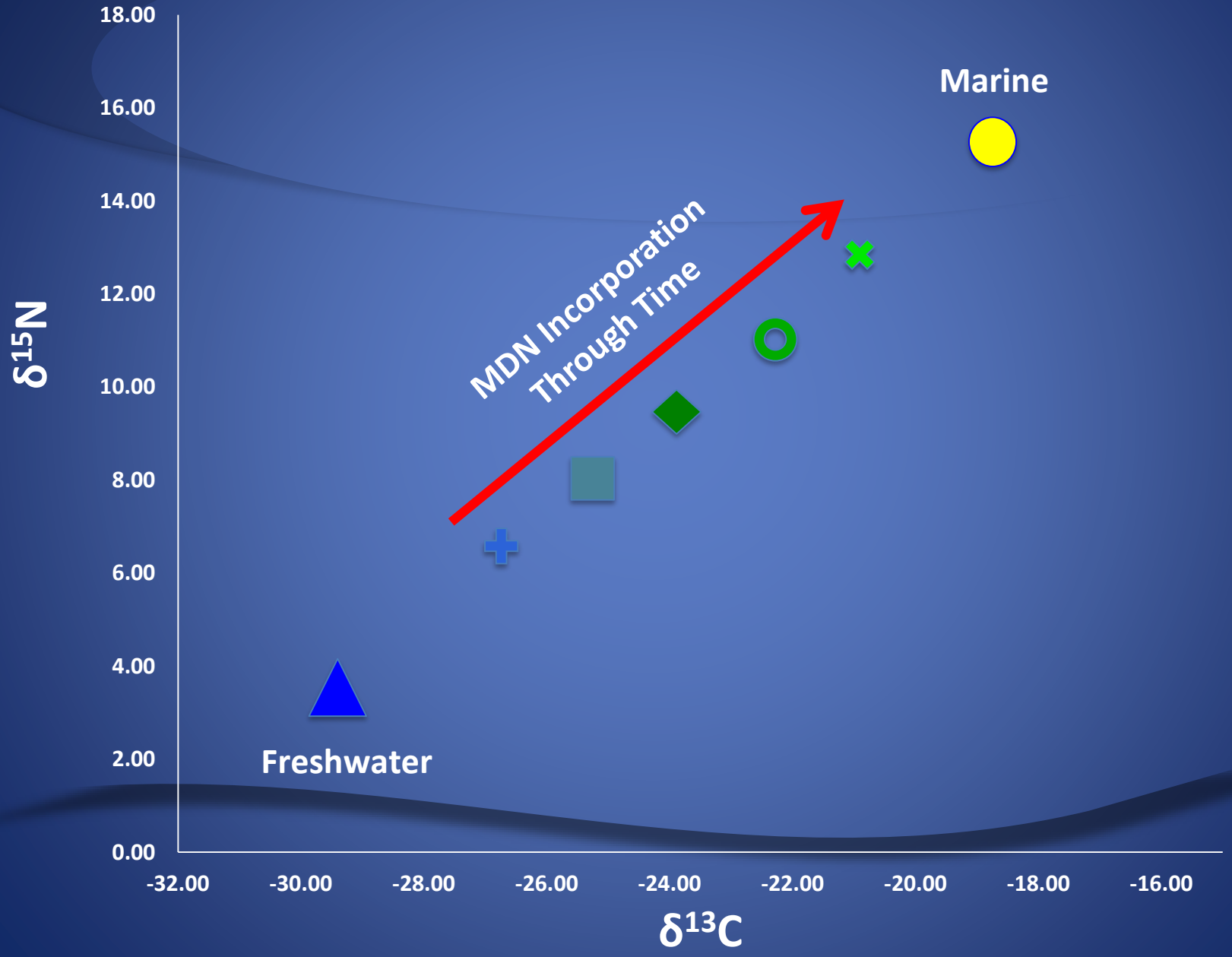


Photos: Nigel Fearon Photography

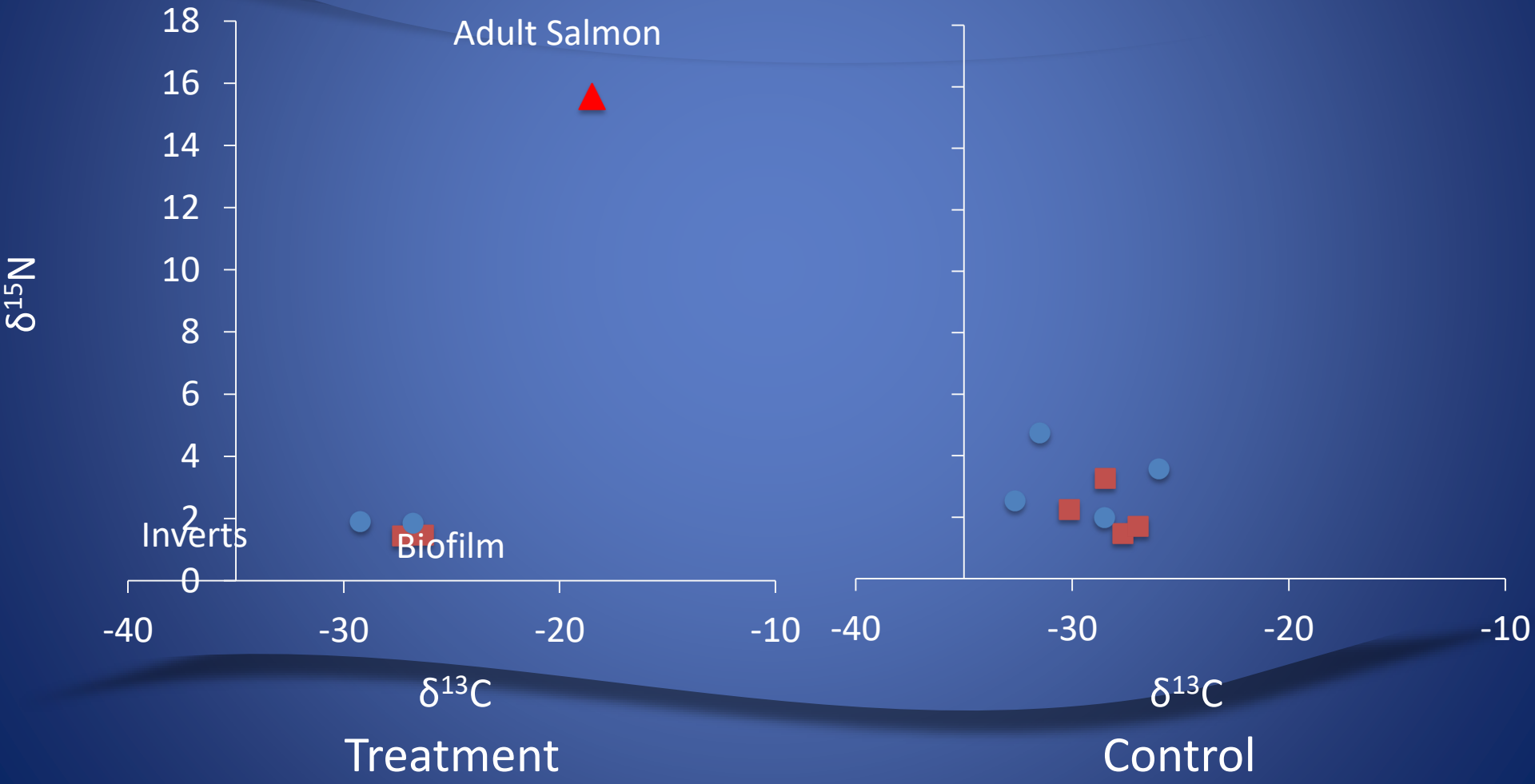
Stable Isotopes as Ecological Tracers



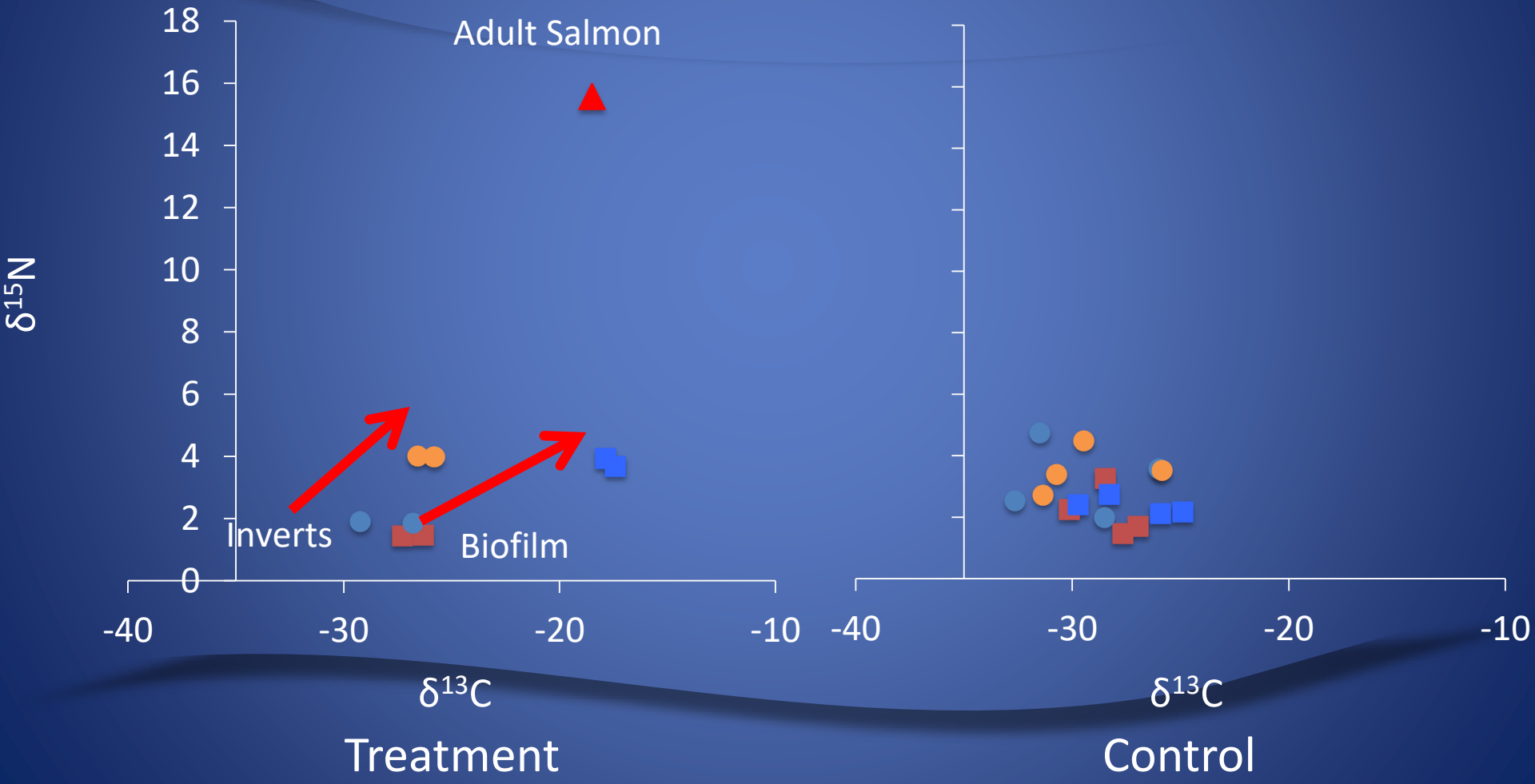
Stable Isotopes as Ecological Tracers



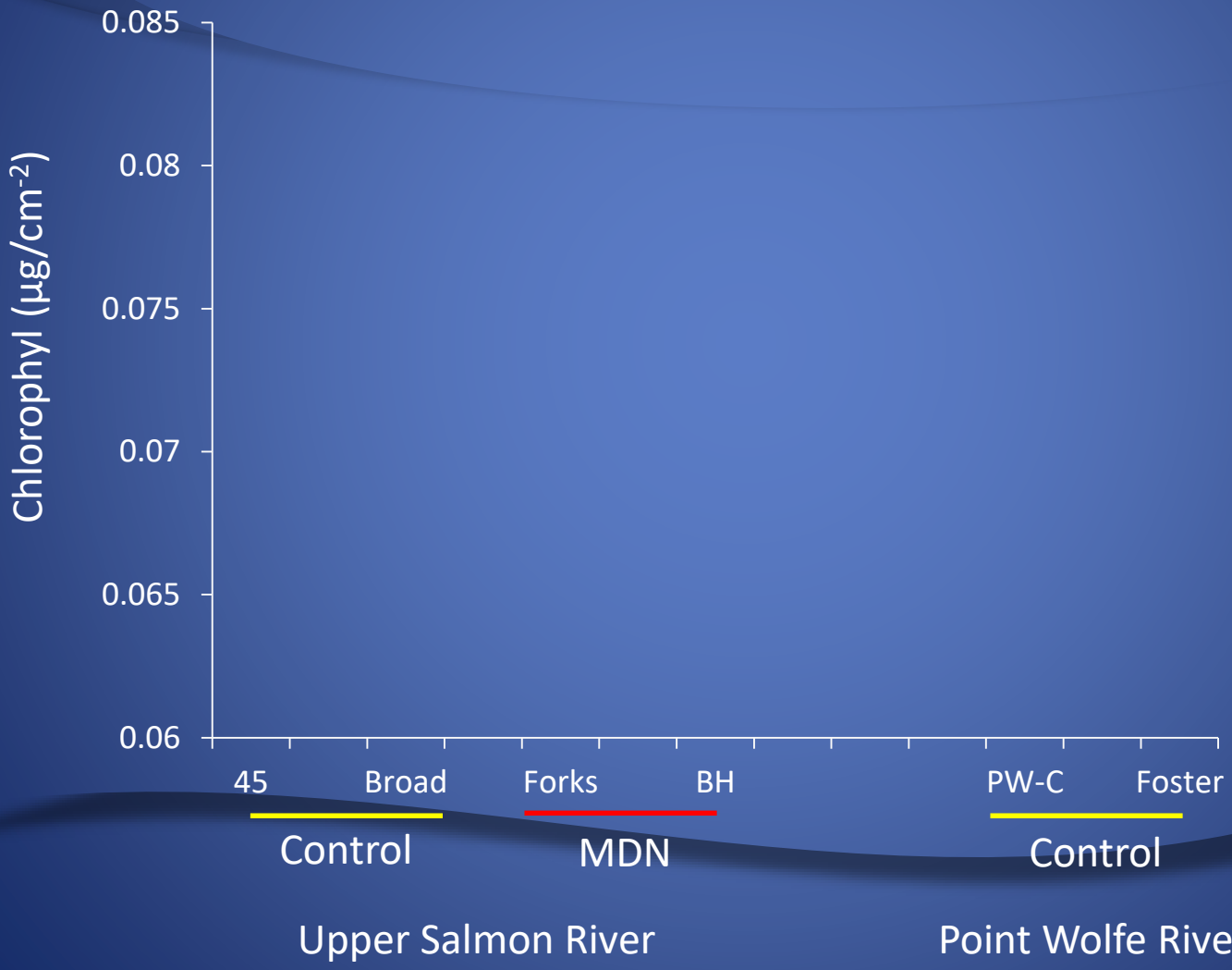
Salmon-derived Resources



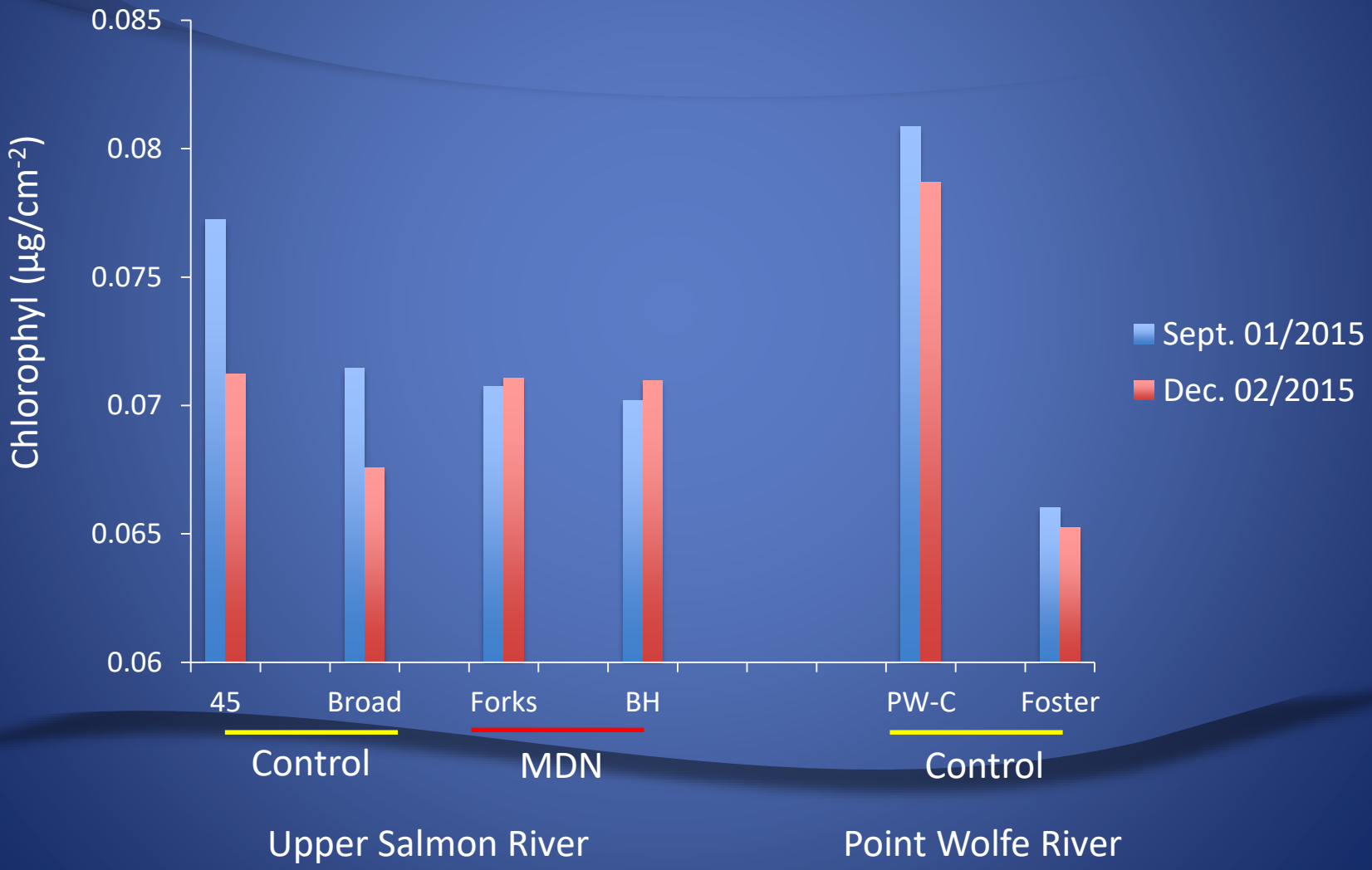
Salmon-derived Resources



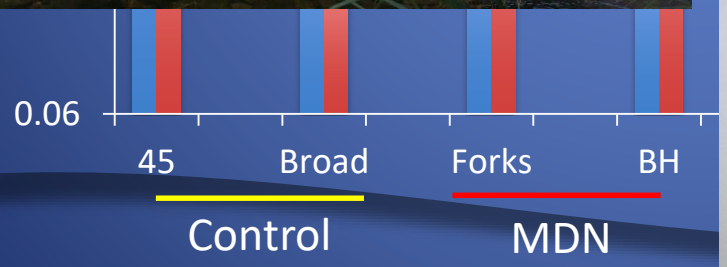
Increases in Primary Production



Increases in Primary Production



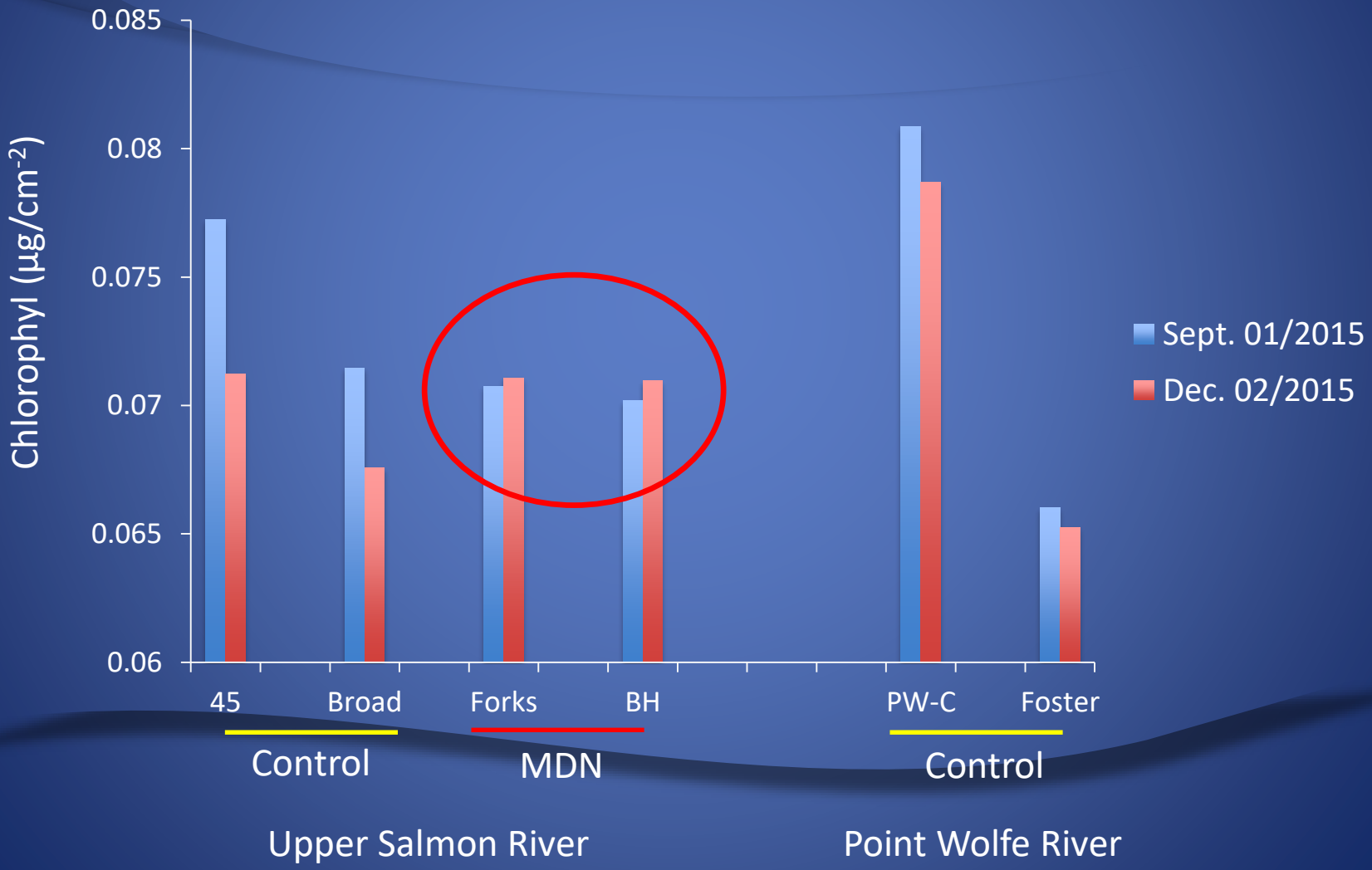
Increases in Prim



Upper Salmon River

Point Wolfe River

Increases in Primary Production



How does the Freshwater Community Respond to Adult Supplementation?

iii. Assess the efficacy of releasing cage-reared adult salmon.

A. Provide reach-scale data on spawning behaviour

a. Passive Integrated Transponder (PIT) telemetry tracking of adults

b. Radio tracking of adults

c. DIDSON Sonar

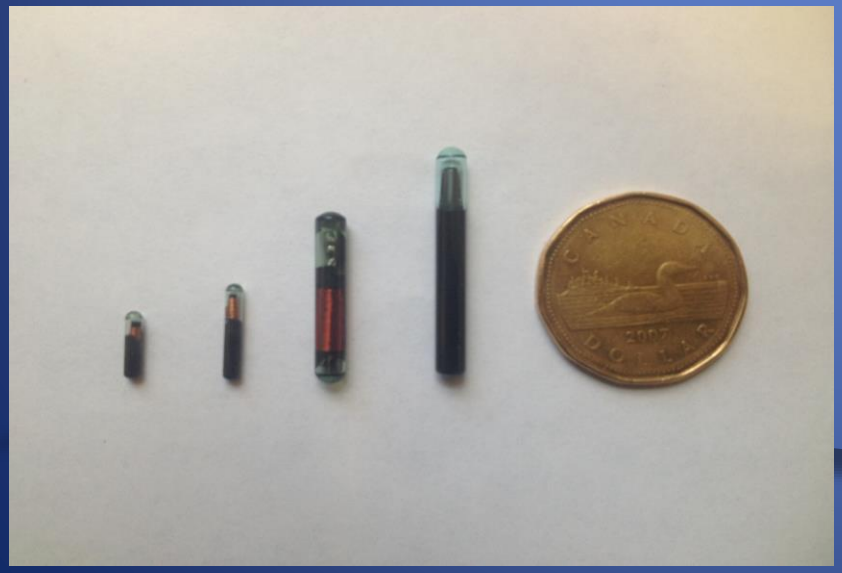
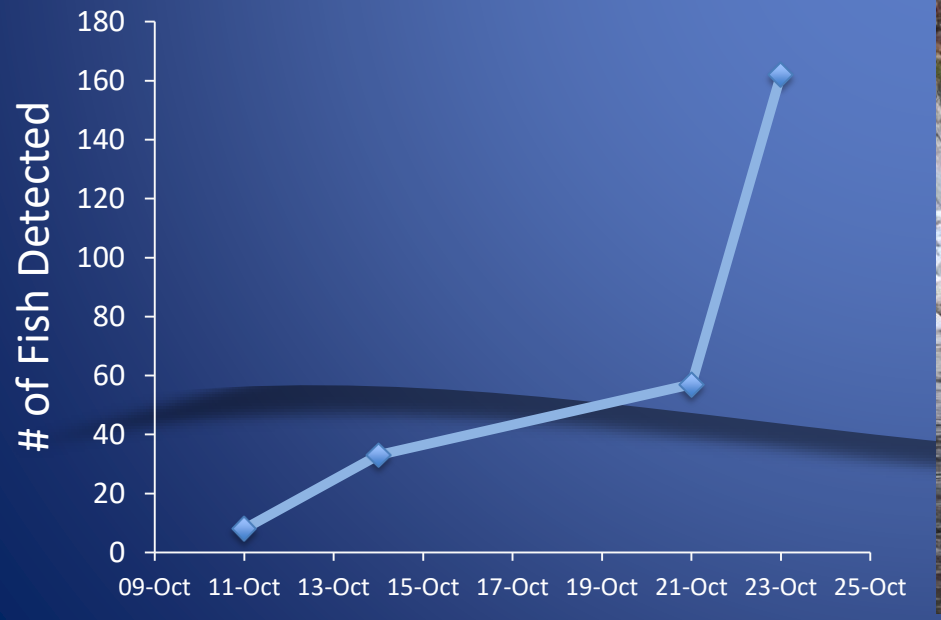


Photo: Laura Buck


Photos: Nigel Fearon Photography

Movements Based on PIT Antennas

- 218 different fish detected (210 from 2016; 8 from 2015)
- First fish returned July 28 (that was not eaten by an eagle)
- 9368 detections (7001 of those between Oct 21-23)
- 7 of the 2015 returning fish still in the river



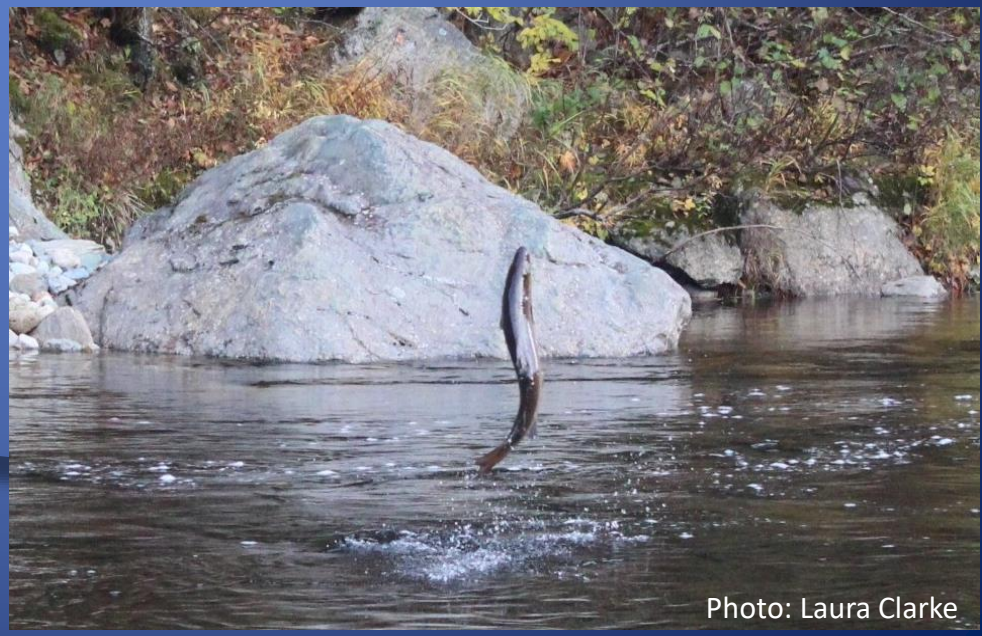


Photo: Laura Clarke

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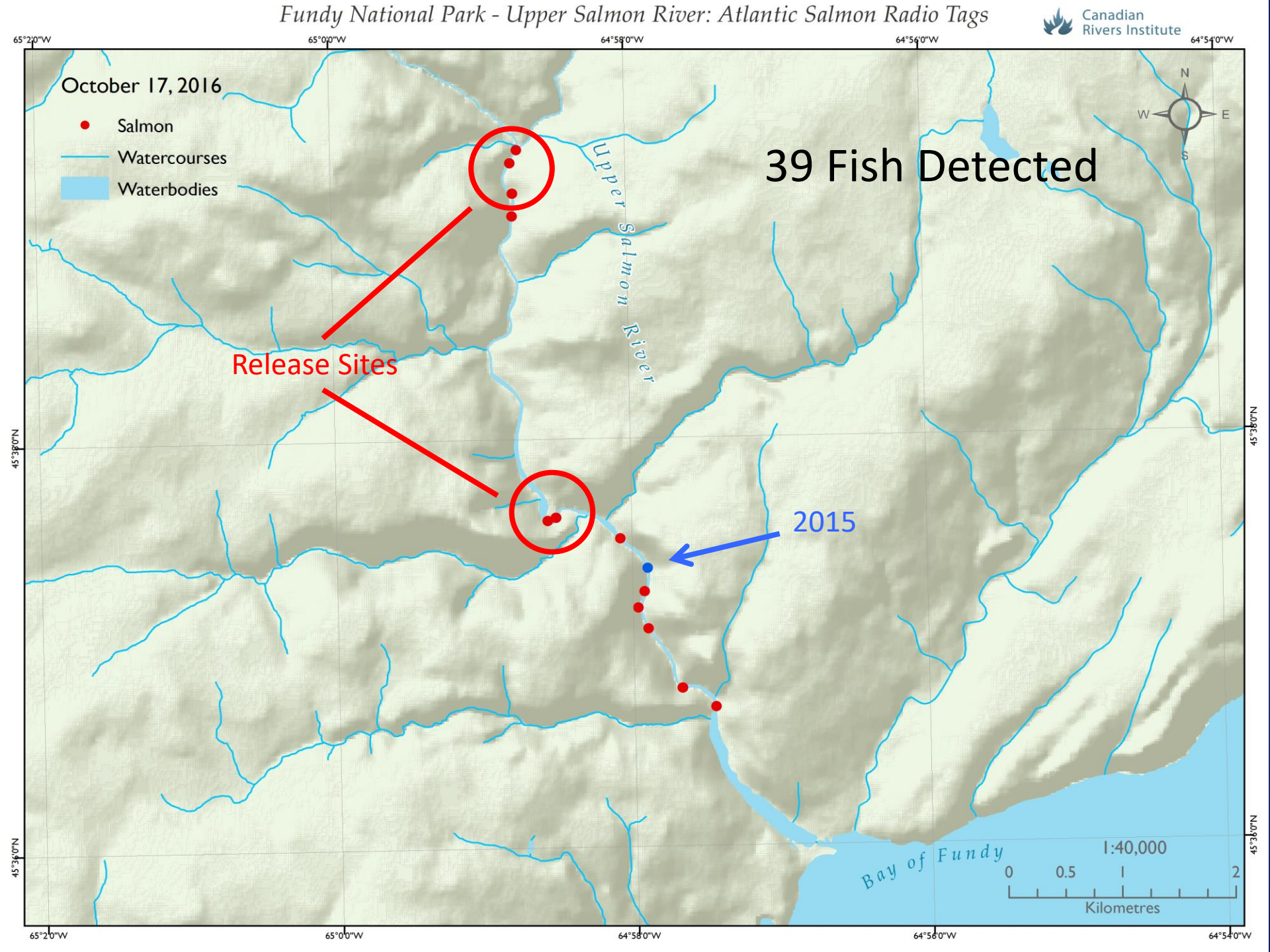
Fundy National Park - Upper Salmon River: Atlantic Salmon Radio Tags
Canadian Rivers Institute
October 17, 2016
Salmon
Watercourses
Waterbodies
39 Fish Detected
Release Sites
2015
Upper Salmon River
Bay of Fundy
1:40,000
0 0.5 1 2
Kilometres
N
W
E
S
65°2'0"W
65°0'0"W
64°58'0"W
64°56'0"W
64°54'0"W
45°38'0"N
45°36'0"N

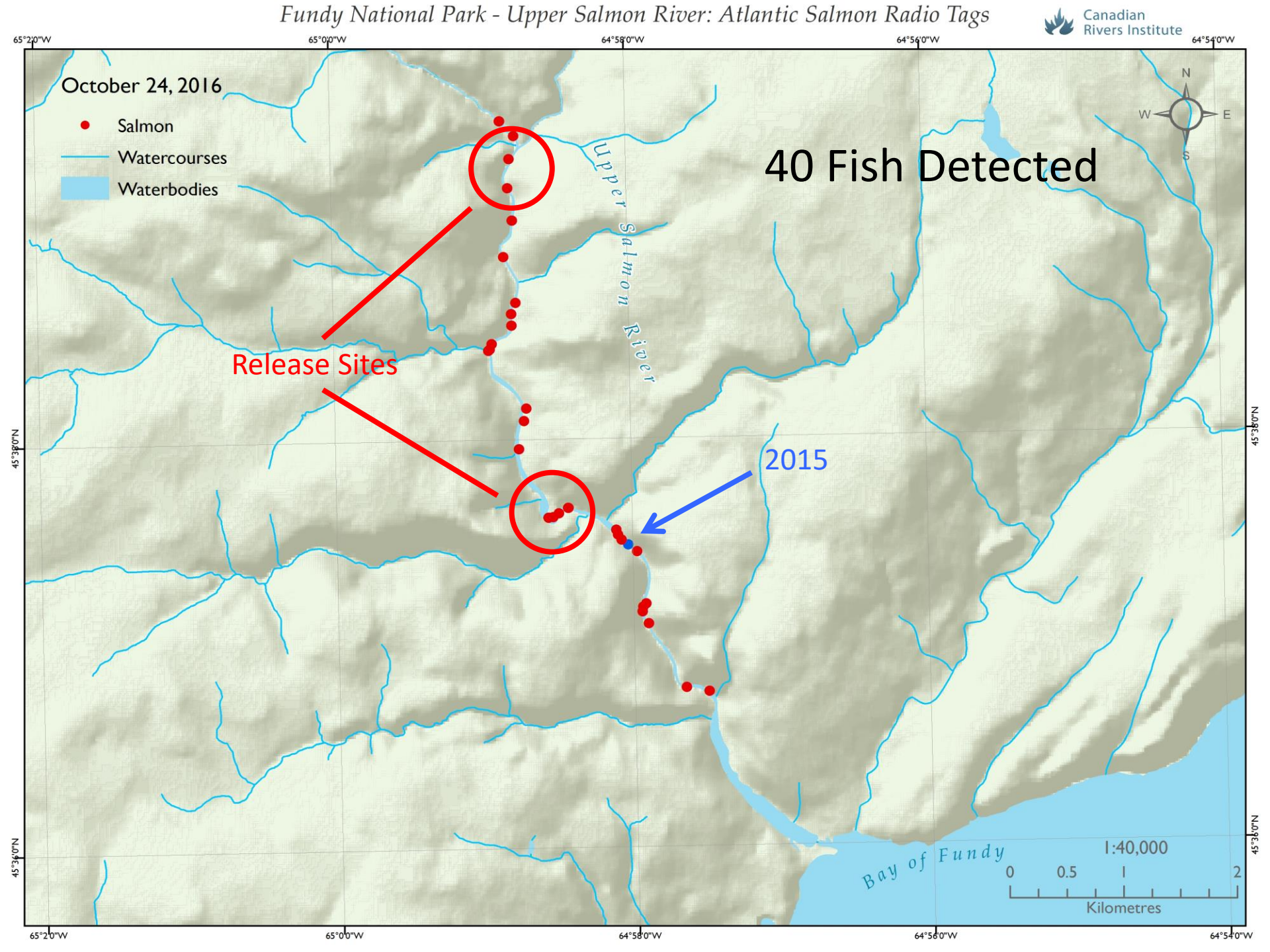
Fundy National Park - Upper Salmon River: Atlantic Salmon Radio Tags
Canadian Rivers Institute
October 24, 2016
Salmon
Watercourses
Waterbodies
40 Fish Detected
Release Sites
2015
Upper Salmon River
Bay of Fundy
1:40,000
0 0.5 1 2
Kilometres
N
W
E
S
65°2'0"W
65°0'0"W
64°58'0"W
64°56'0"W
64°54'0"W
45°38'0"N
45°36'0"N

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Photo: Lauren Fitzpatrick


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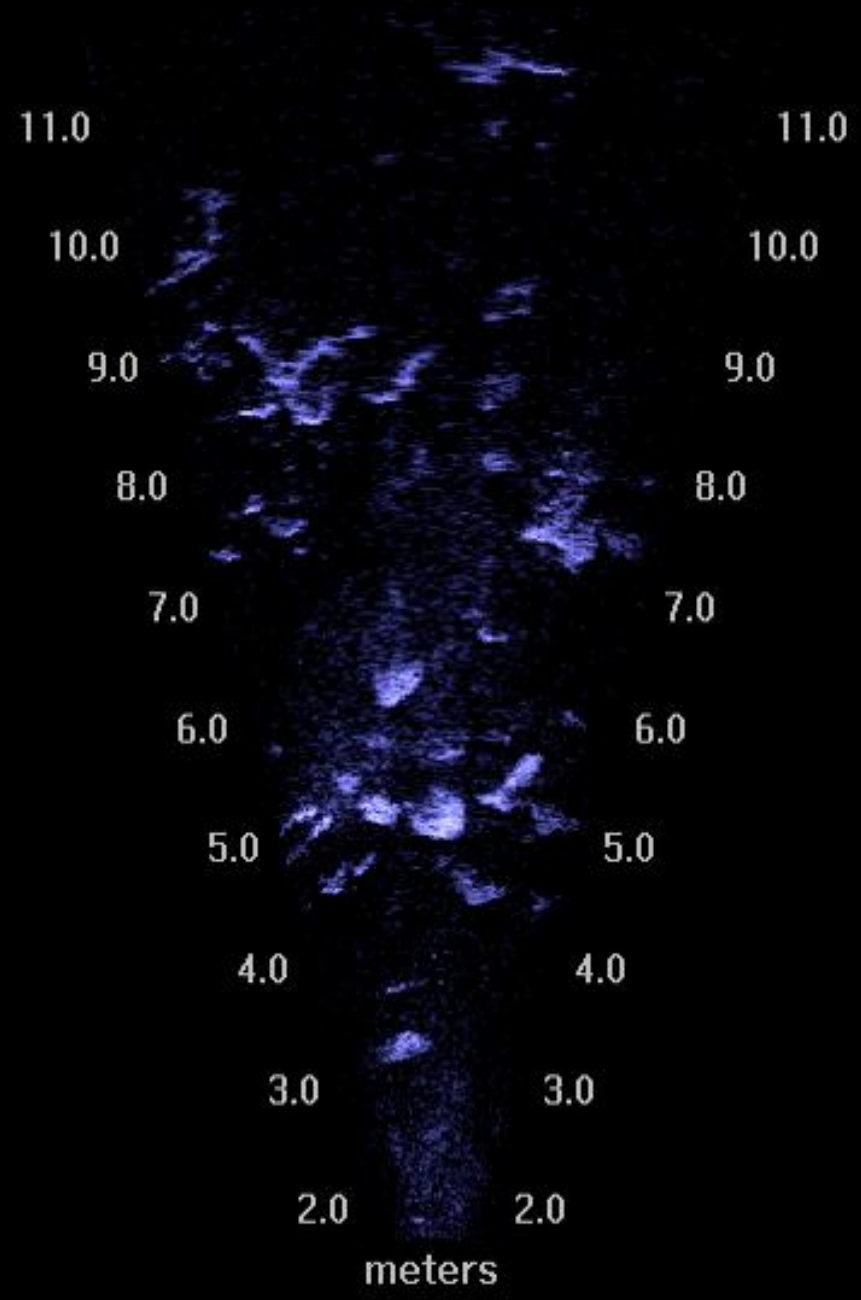
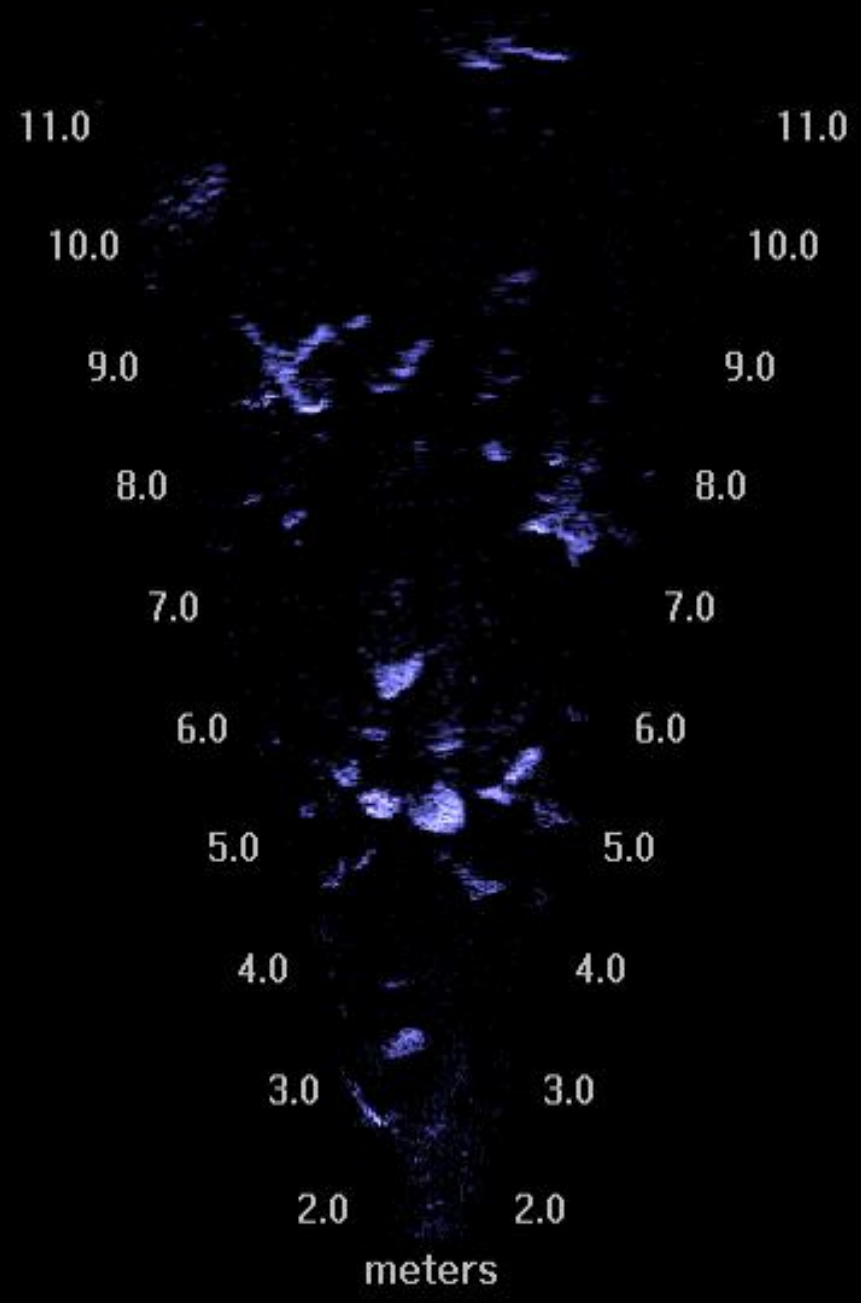

11.0
11.0
10.0
10.0
9.0
9.0
8.0
8.0
7.0
7.0
6.0
6.0
5.0
5.0
4.0
4.0
3.0
3.0
2.0
2.0
meters


11.0
11.0
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9.0
9.0
8.0
8.0
7.0
7.0
6.0
6.0
5.0
5.0
4.0
4.0
3.0
3.0
2.0
2.0
meters

Take Home Message

- Even small numbers of mature adults appear to impact food web
- Changes are likely the result of MDN and not another nutrient source
- Majority of fish retained in the system
- Mature adults migrating upriver

Photo: Nigel Fearon Photography

Thank You!

Michelle Charest, Emily Corey, Alex Parker, Samantha Petty, Colin De Coste, Lauren Fitzpatrick, Coralie Laplace, Emma Laliberte, Mark Gautreu, Tommi Linnansaari, Laura Clarke, Rick Cunjak

Questions?

Photo: Tim Robinson

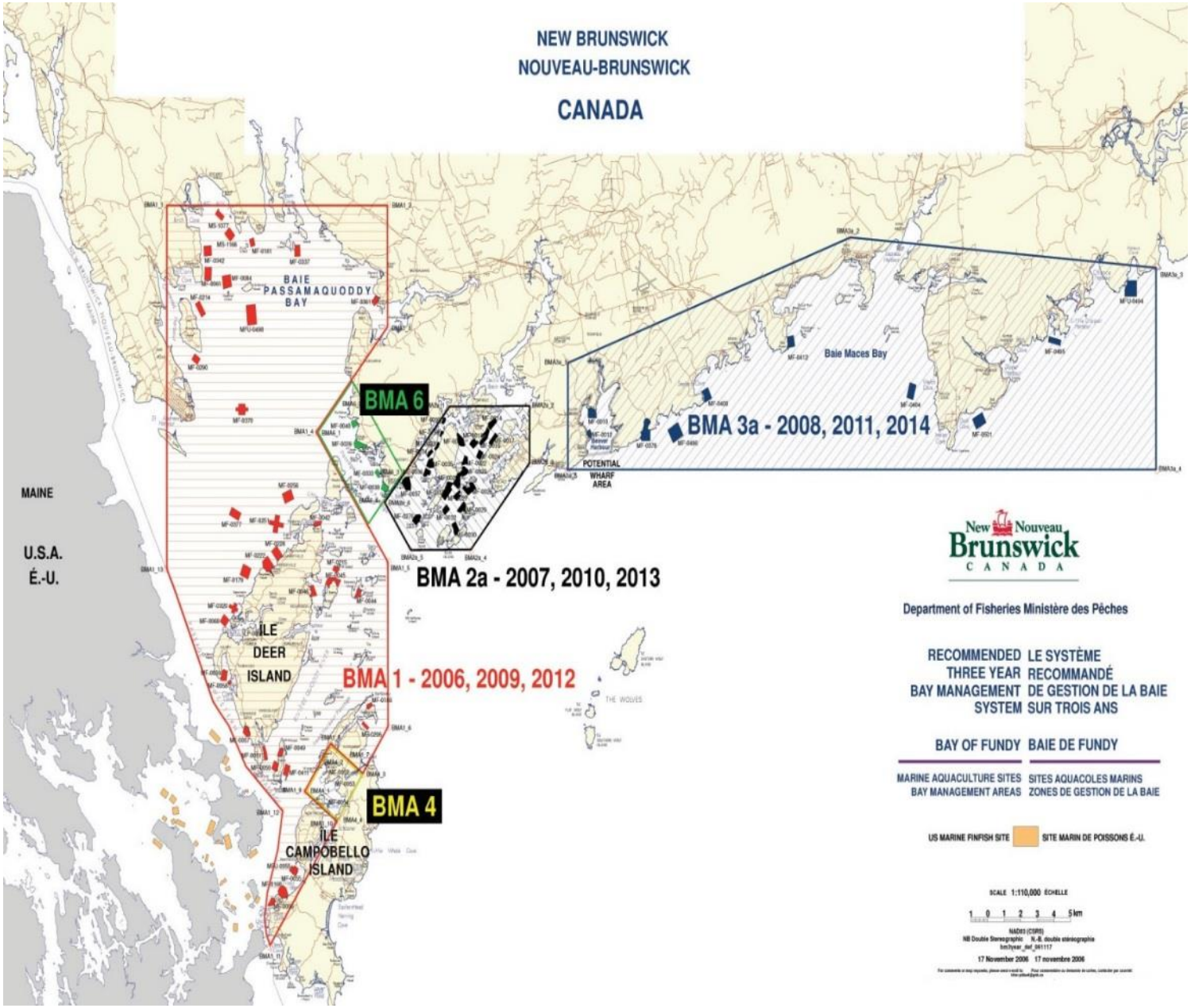
Impact of salmon aquaculture on the diversity and health of benthic communities in shallow coastal habitats of the Bay of Fundy

Heather Hunt, Rémy Rochette, Karen Kidd

EST. 1785 UNB Saint John

Coastal Ecosystems in Bay of Fundy

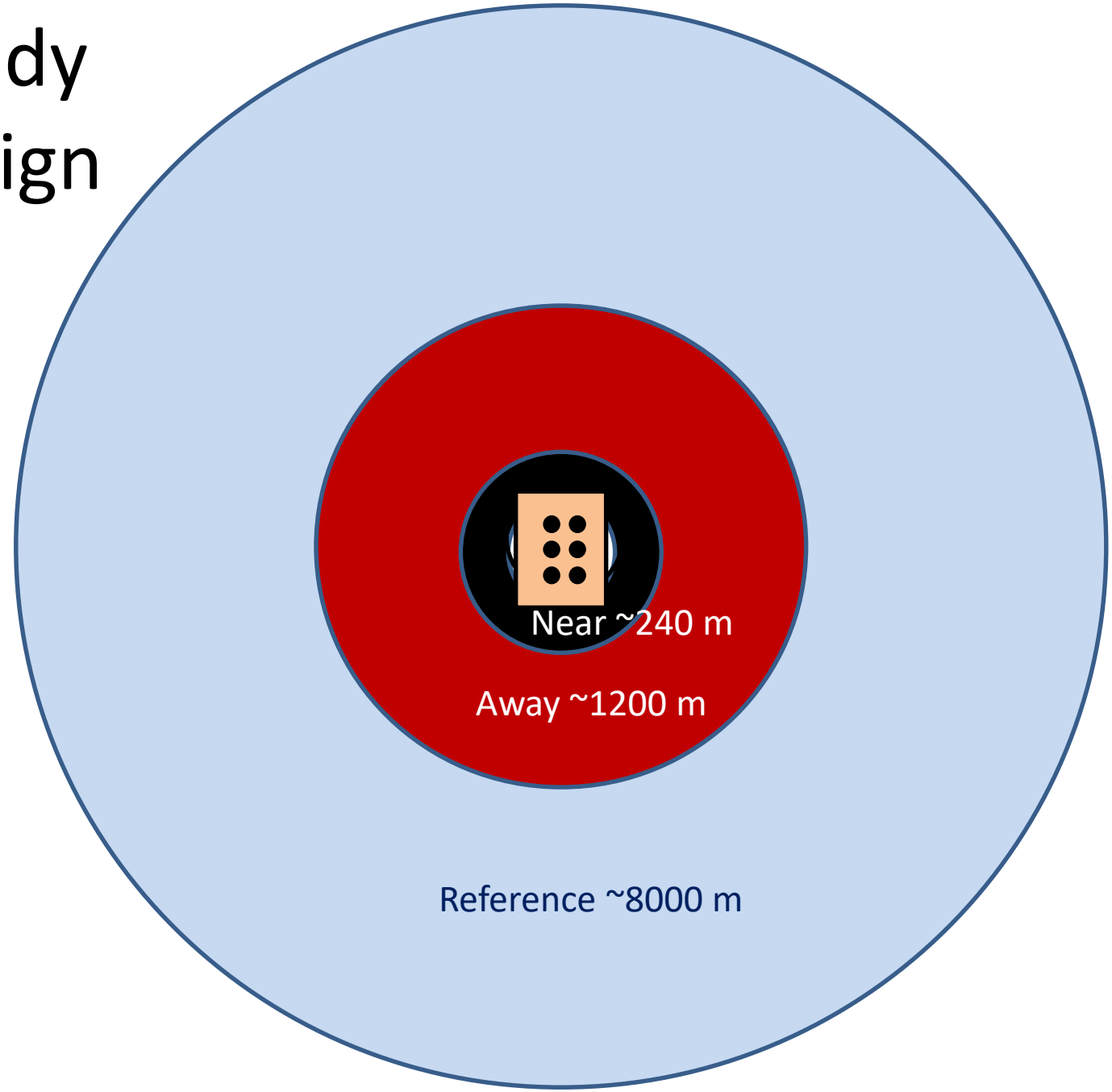
- High biodiversity
- Also salmon aquaculture and lobster fishery
- Cobble habitat
  - Scarce in Bay of Fundy
  - Important for many species e.g. juvenile lobster

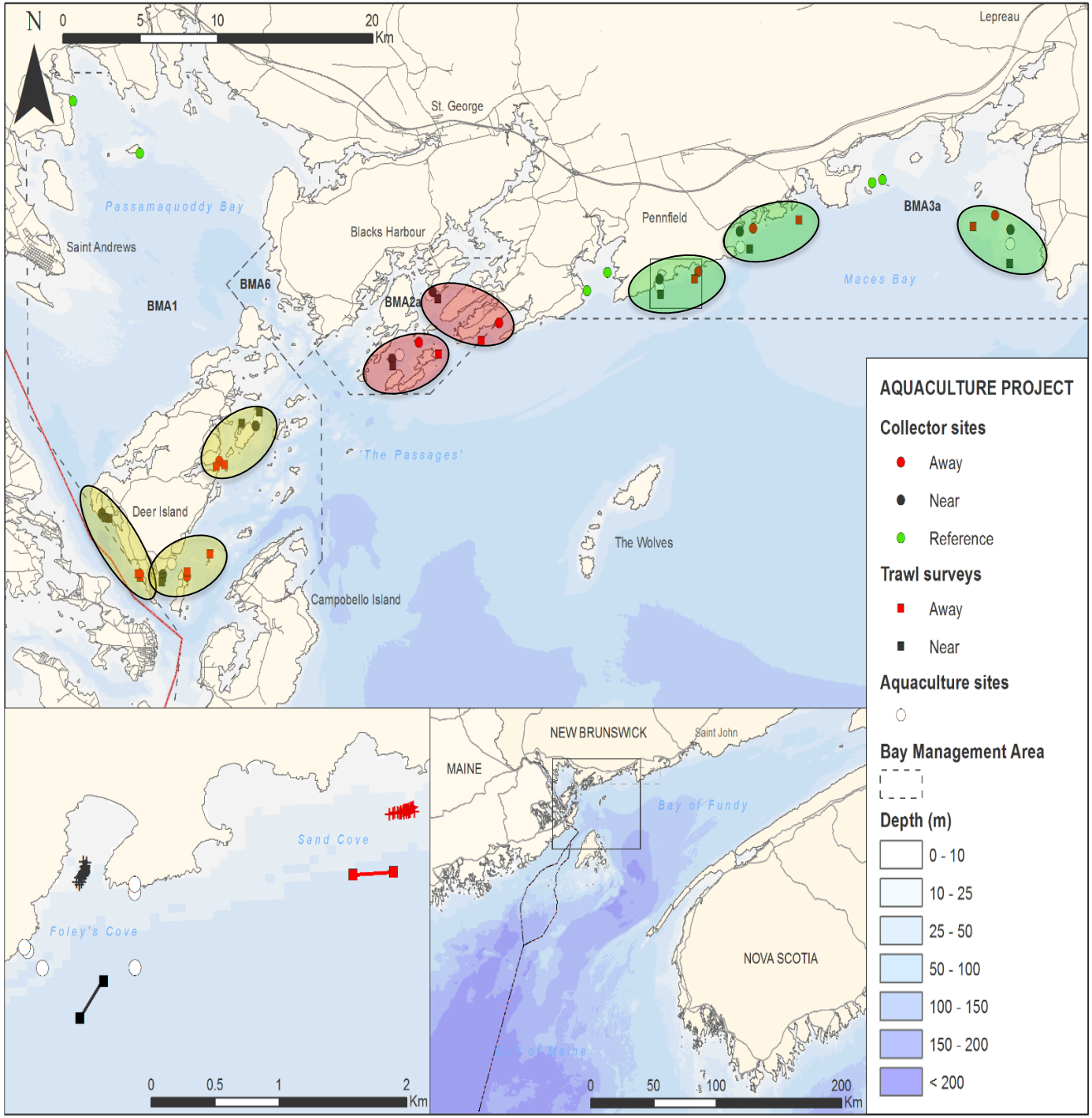

NEW BRUNSWICK
NOUVEAU-BRUNSWICK
CANADA
BAIE PASSAMAQUODDY BAY
BMA 6
Baie Maces Bay
BMA 3a - 2008, 2011, 2014
POTENTIAL WHARF AREA
MAINE
U.S.A.
É.-U.
BMA 2a - 2007, 2010, 2013
ÎLE DEER ISLAND
BMA 1 - 2006, 2009, 2012
THE WOLVES
BMA 4
ÎLE CAMPOBELLO ISLAND
New Nouveau Brunswick CANADA
Department of Fisheries Ministère des Pêches
RECOMMENDED THREE YEAR BAY MANAGEMENT SYSTEM
LE SYSTÈME RECOMMANDÉ DE GESTION DE LA BAIE SUR TROIS ANS
BAY OF FUNDY
BAIE DE FUNDY
MARINE AQUACULTURE SITES
BAY MANAGEMENT AREAS
SITES AQUACOLES MARINS
ZONES DE GESTION DE LA BAIE
US MARINE FINFISH SITE
SITE MARIN DE POISSONS É.-U.
SCALE 1:110,000 ÉCHELLE
1 0 1 2 3 4 5km
NAD83 (CSRS)
NB Double Stereographic N.-B. double stéréographie
bmthyear_def_061117
17 November 2006 17 novembre 2006

Objectives and approach

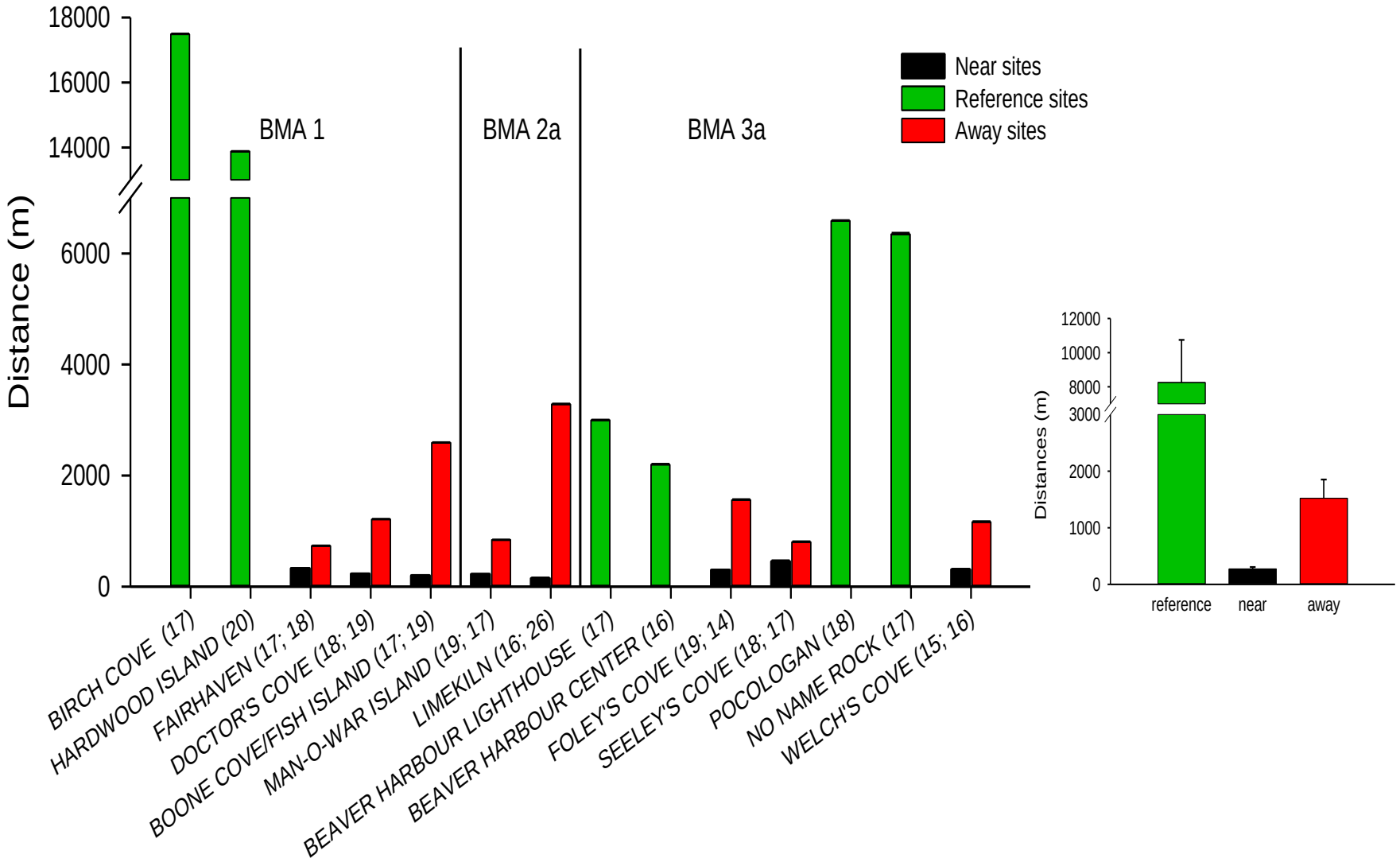
- **Objective**: quantify effects (+ and -) of salmon aquaculture on diversity and health of benthic communities in shallow cobble habitat
- **Approach**: 8 site pairs near and away from aquaculture (+ reference sites)
  - Trap surveys for adult lobster
  - Bio-collectors

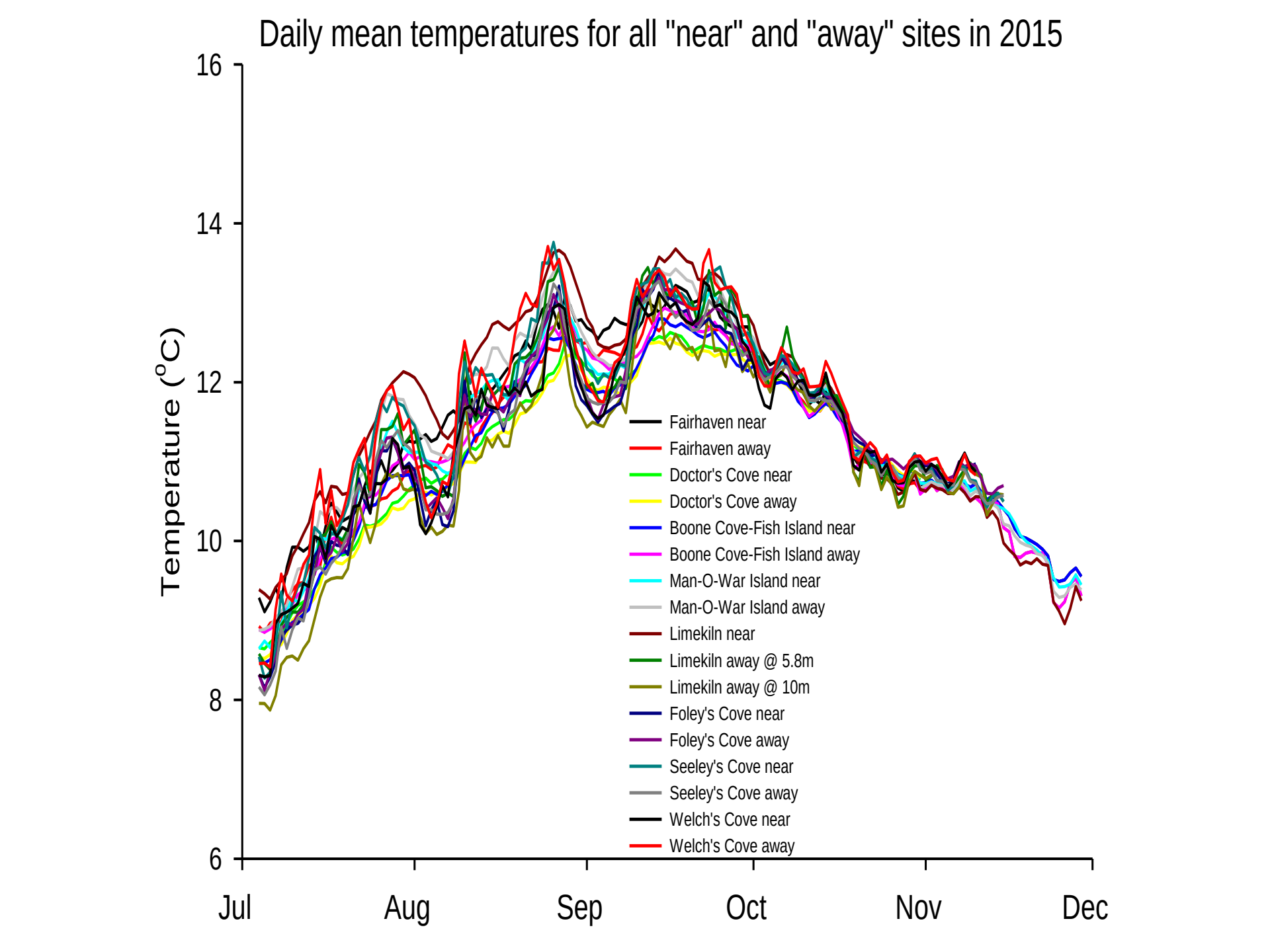
Study design




N
0
5
10
20
Km
Lepreau
St. George
Passamaquoddy Bay
Saint Andrews
Blacks Harbour
Pennfield
BMA3a
Maces Bay
BMA6
BMA1
BMA2a
'The Passages'
Deer Island
The Wolves
Campobello Island
AQUACULTURE PROJECT
Collector sites
Away
Near
Reference
Trawl surveys
Away
Near
Aquaculture sites
Bay Management Area
Depth (m)
0 - 10
10 - 25
25 - 50
50 - 100
100 - 150
150 - 200
< 200
Sand Cove
Foley's Cove
0
0.5
1
2
Km
NEW BRUNSWICK
Saint John
MAINE
Bay of Fundy
NOVA SCOTIA
Gulf of Maine
0
50
100
200
Km

Mean (+SE) distance between collectors and aquaculture pens in 2015
Number of collectors retreived are in brackets (near; away)




Daily mean temperatures for all "near" and "away" sites in 2015
Temperature (°C)
16
14
12
10
8
6
Jul
Aug
Sep
Oct
Nov
Dec
Fairhaven near
Fairhaven away
Doctor's Cove near
Doctor's Cove away
Boone Cove-Fish Island near
Boone Cove-Fish Island away
Man-O-War Island near
Man-O-War Island away
Limekiln near
Limekiln away @ 5.8m
Limekiln away @ 10m
Foley's Cove near
Foley's Cove away
Seeley's Cove near
Seeley's Cove away
Welch's Cove near
Welch's Cove away

2014-2016 Lobster trap survey

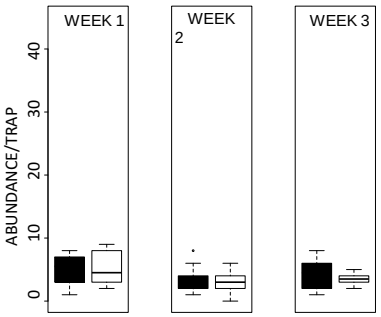
Rémy Rochette, Melanie Wiber, FNFA

2014

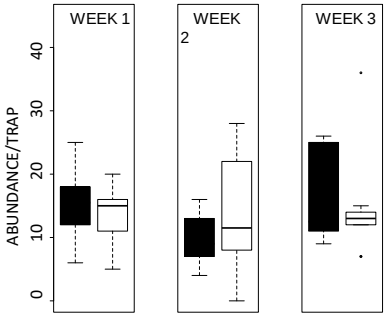
fallow BMA 1

Yr-2 fish BMA 2

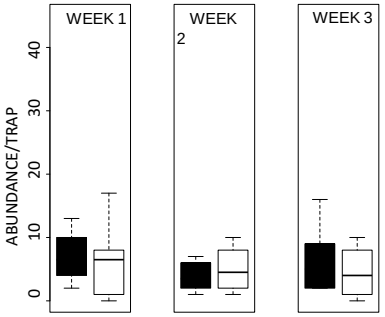
Yr-1 fish BMA 3



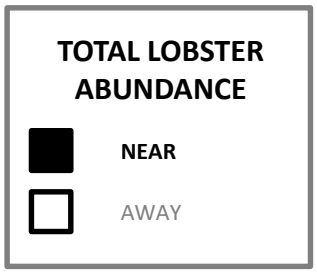
FAIRHAVEN/ROUNDMARSH

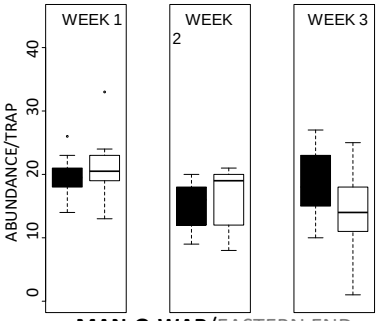


BOONE COVE/NW HARBOUR

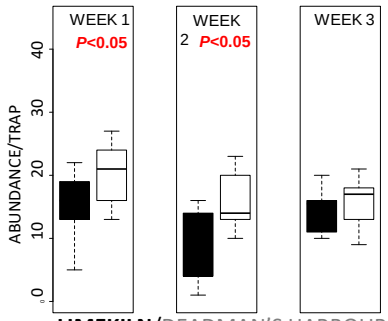


DOCTOR'S COVE/INDIAN ISLAND

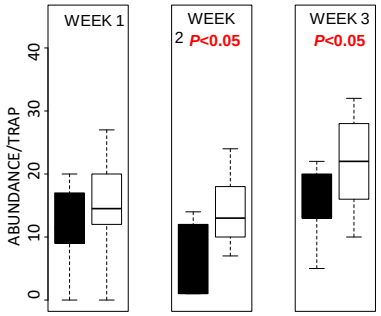




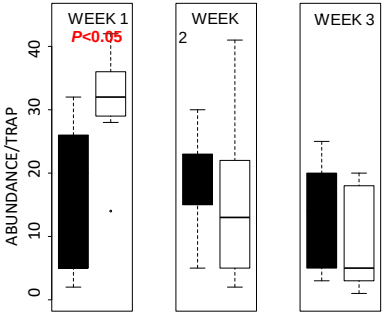
MAN-O-WAR/EASTERN END



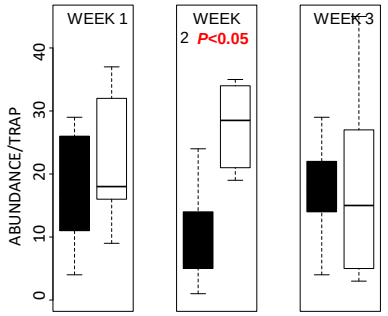
LIMEKILN/DEADMAN'S HARBOUR



FOLEY'S COVE/SAND COVE



WELCH'S COVE/N WELCH'S

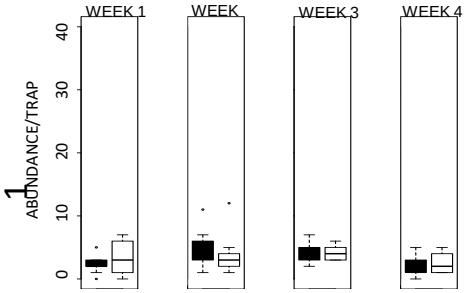


SEELEY'S COVE/SEELEY'S BASIN

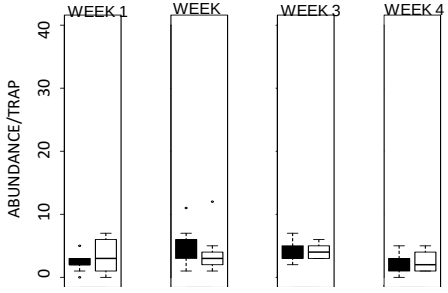
2015

Yr-1 fish

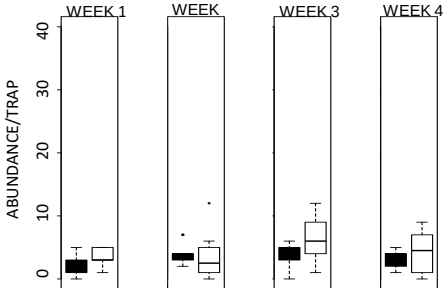
BMA 1



FAIRHAVEN/ROUNDMARSH



BOONE COVE/NW HARBOUR



DOCTOR'S COVE/INDIAN ISLAND

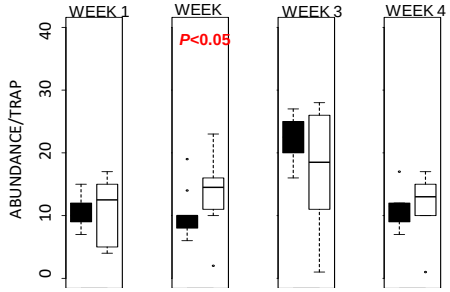
fallow

BMA 2

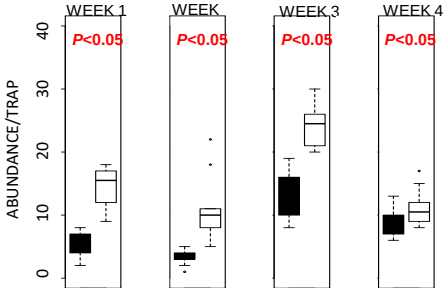
TOTAL LOBSTER ABUNDANCE

NEAR

AWAY



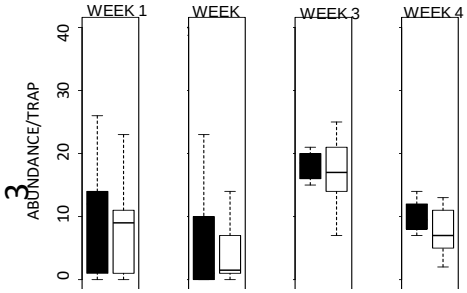
MAN-O-WAR/EASTERN END



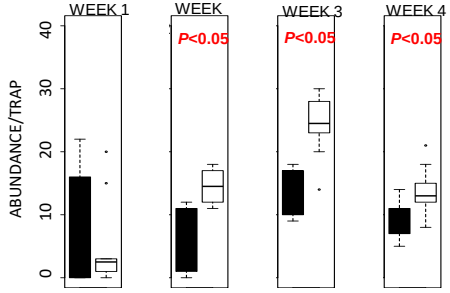
LIMEKILN/DEADMAN'S HARBOUR

Yr-2 fish

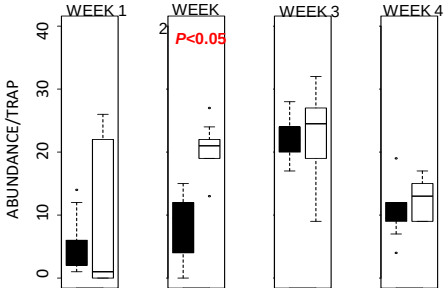
BMA 3



FOLEY'S COVE/SAND COVE



WELCH'S COVE/N WELCH'S



SEELEY'S COVE/SEELEY'S BASIN

2016

Yr-2 fish

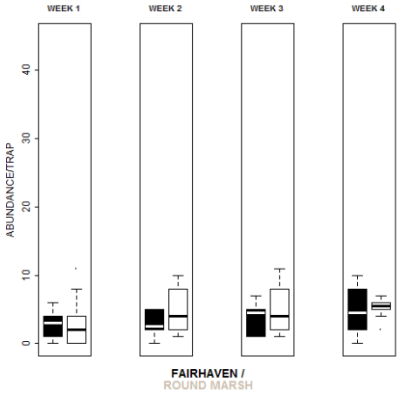
Yr-1 fish

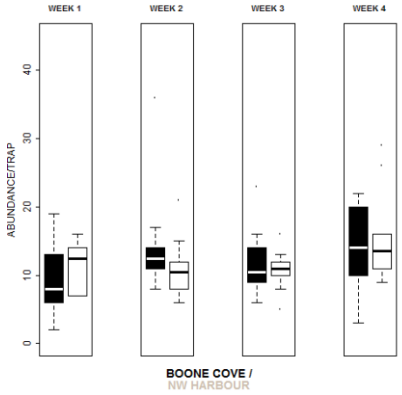
fallow

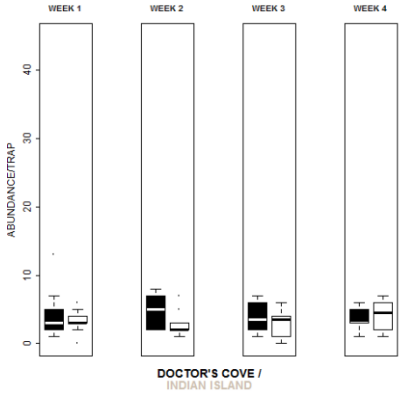
BMA 1

BMA 2

BMA 3



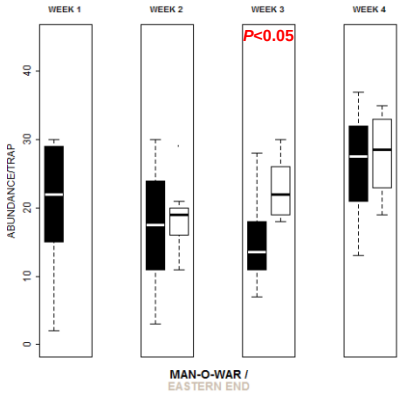


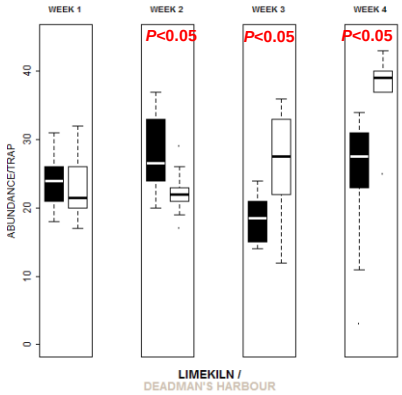


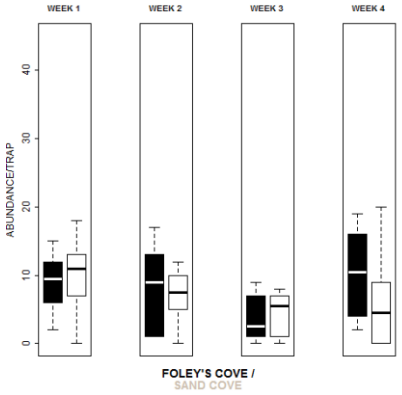
TOTAL LOBSTER ABUNDANCE

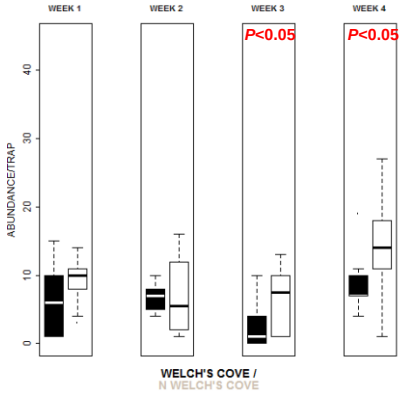
NEAR

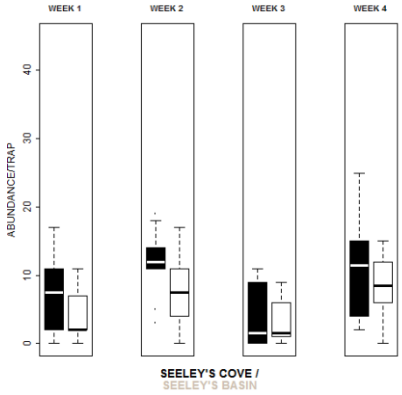
AWAY











Bio-collectors

Comparing:

(1) diversity and abundance

(2) exposure to chemicals and nutrients from aquaculture (metals and stable isotopes)

Bio-collectors

- Wire mesh (50 mm) lined with 1 mm mesh
- Filled with ~10 cm rounded rocks
- Jul-Nov
- 5-10 m depth

Bio-collector processing

- Collectors processed on land
- 2015
  - Subsampling for small organisms
  - Decapod crustaceans and fish in all
- Measurements & identification in lab

Decapod crustaceans and fishes

American lobster

Shrimps

Eualus spp. *Lebbeus* spp.

Spirontocaris spp.

Pandalus montagui

Crangon septemspinosa

Crabs

Cancer spp.
www.vitalsignsme.org

Hyas spp.

Carcinus maenas
www.pbase.com

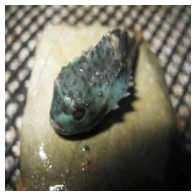
Hermit crabs

Pagurus spp.

Fishes

Rock Gunnel: *Pholis gunnellus*

Seasnail: *Liparis* sp.



Lump fish: *Cyclopterus lumpus*

Radiated shanny: *Ulvaria subbifurcata*

Cunner: *Tautogolabrus adspersus*

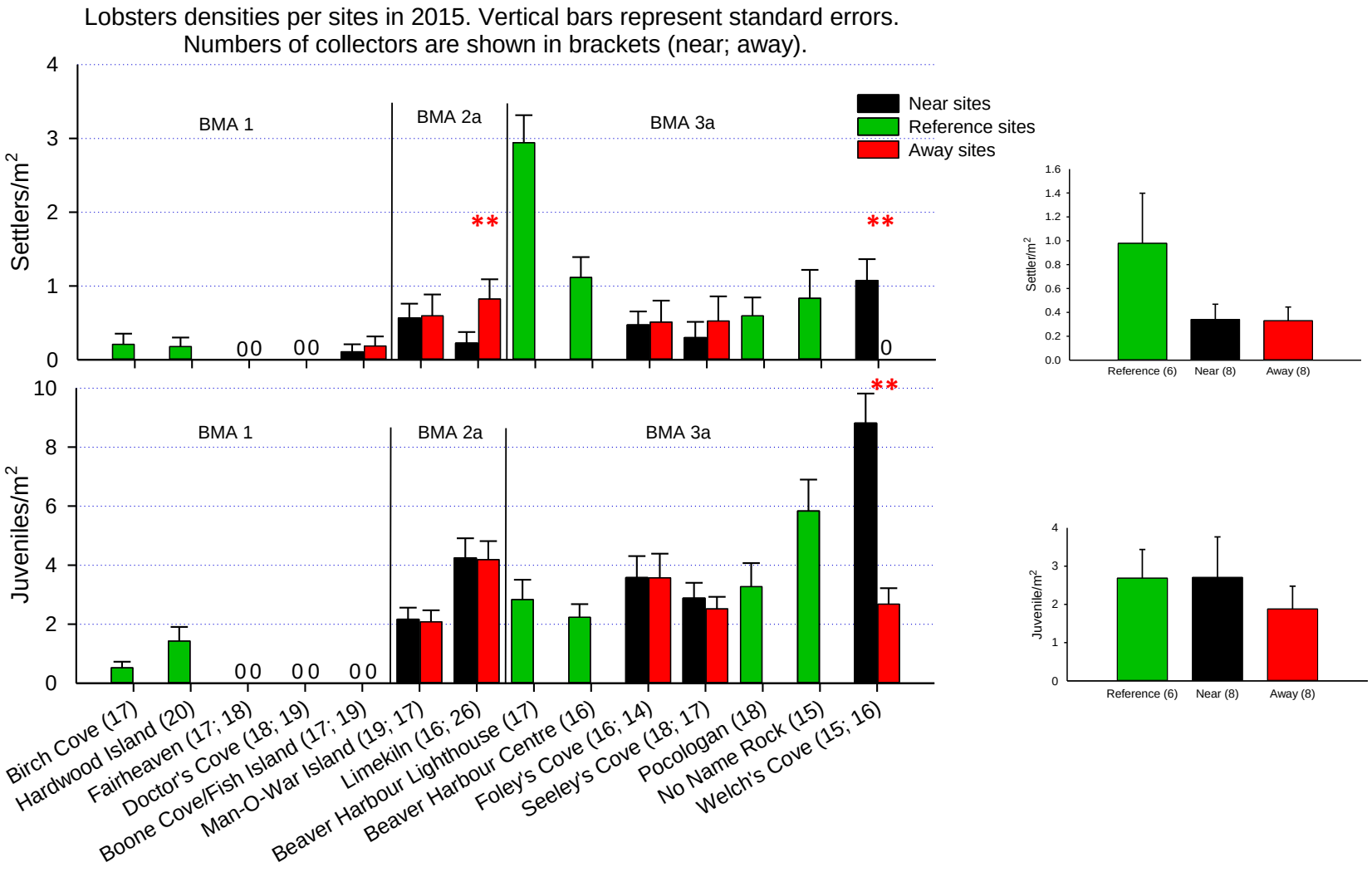
Sculpins: Myoxocephalus spp.

White hake: *Urophycis tenuis*
http://www.dfo-mpo.gc.ca/species-especes/aquatic-aquatique/white-hake-merluche-blanche-eng.htm

Eelpout: *Zoarces* sp.

Tomcod: *Microgadus tomcod*

Lobster settlers and juveniles



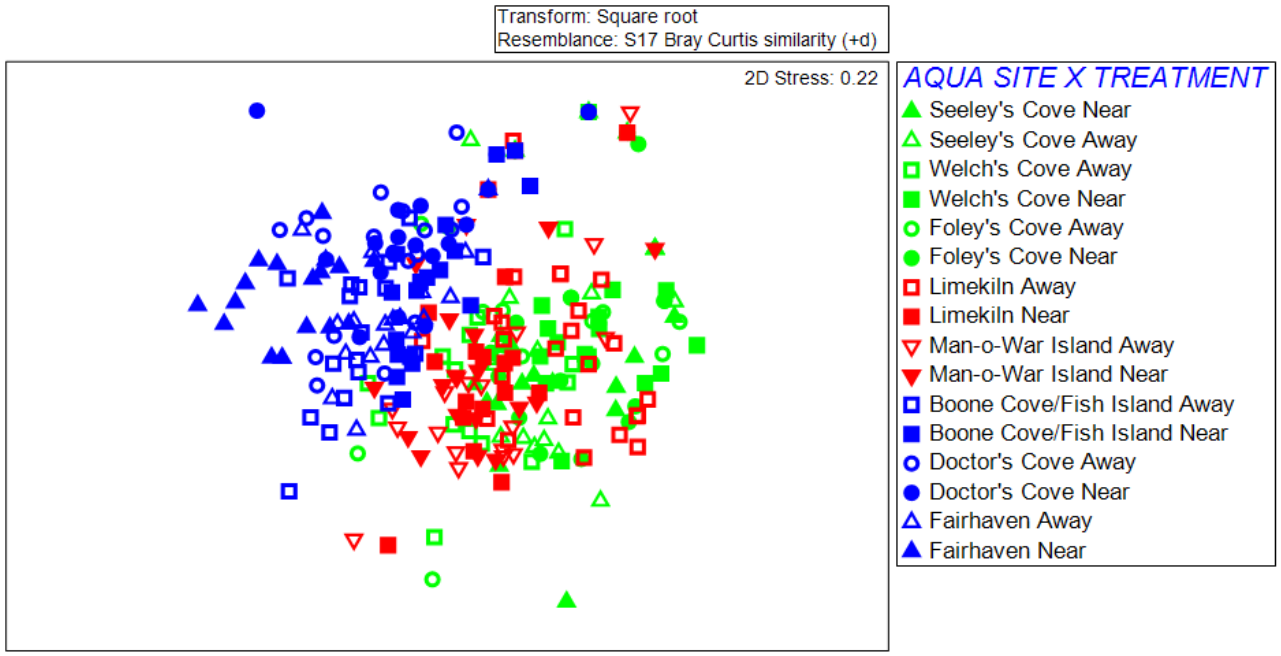
Lobster summary

	American lobster *Homarus americanus*			
	Adults 2014	**Adults 2015**	**YOY 2015**	**Adults 2016**
BMA-1	fallow	Yr-1	Yr-1	Yr-1
Fairhaven's				
Doctor's Cove				
Boone Cove				
BMA-2a	Yr-2	fallow	fallow	fallow
Man-O-War		(1/4)		(1/4)
Limekiln	(2/3)	(4/4)		1/4 2/4
BMA-3a	Yr-1	Yr-2	Yr-2	Yr-2
Foley's Cove	(2/3)			
Seeley's Cove	(1/3)	(3/4)		
Welch's Cove	(1/3)	(1/4)		(2/4)

No difference
Near < Away
Near > Away

Macro-Biodiversity

MDS of sqrt-transformed fish and decapod data



PERMANOVA table of results

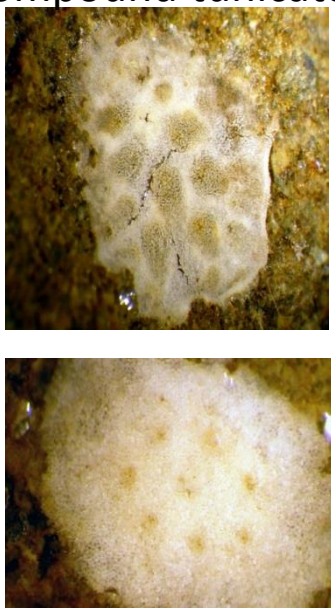
Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
BM	2	94418	47209	6.2326	0.001	973
TR	1	2028.3	2028.3	0.4099	0.779	998
AQ(BM)	5	37340	7467.9	6.569	0.001	998
BMxTR	2	12528	6264.2	1.2607	0.316	999
AQ(BM)xTR	5	24551	4910.1	4.319	0.001	999
Res	269	3.05E5	1136.9			
Total	284	4.78E5				

Encrusting species

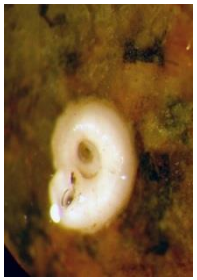
scale: 0 (none); 1 (1 to 10); 2 (11 to 100); 3 (100+)

Bryozoans

Didemnum albidum (compound tunicate)

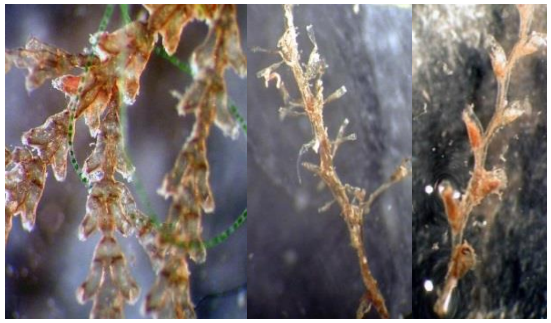


Spirorbis spp.



Anomia spp. (jingle shells)

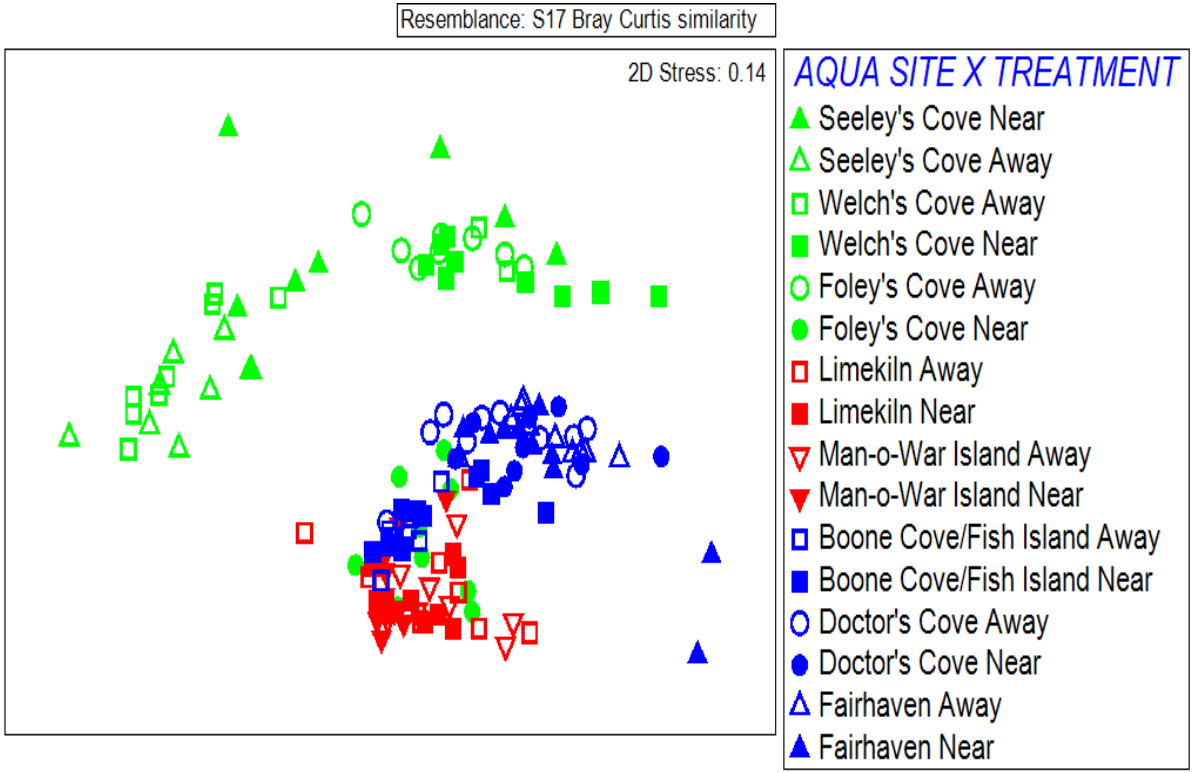
Hydroids



Encrusting species

MDS of encrusting species

log-scale abundances transformed to counts per collector

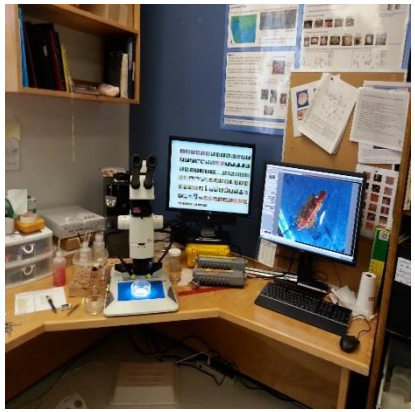


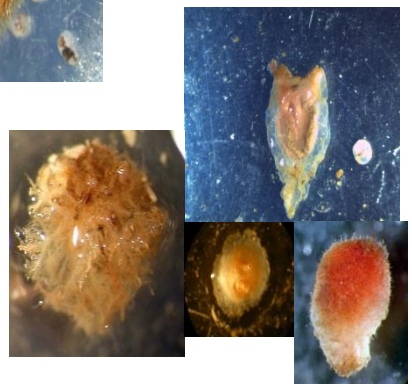
Total adjusted abundance per collector

PERMANOVA table of results

Source	df	SS	MS	Pseudo-F	P(perm)	Unique perms
BM	2	83191	41595	5.3217	0.01	951
TR	1	4626.7	4626.7	0.82575	0.473	998
AQ(BM)	5	39089	7817.7	11.728	0.001	998
BMxTR	2	11558	5779.2	1.0314	0.425	998
AQ(BM)xTR	5	28022	5604.4	8.4074	0.001	995
Res	131	87325	666.61			
Total	146	2.5593E5				

Micro-biodiversity





All animals > ½ mm in center portion of collector are being identified under magnifying glass or dissecting scope

Still in progress…

Sampling biodiversity

Bio-collectors in Bay of Fundy 2009-2015

> 500 species in 14 Phylum

- 8 sponges
- 91 molluscs
- 111 annelids
- 121 arthropods
- 11 echinoderms
- 25 bryozoans
- 24 Chordates, primarily fishes

Metal and stable isotope data

- Potential for elevated metals (copper, zinc) from antifouling compounds or micronutrients in feed
- Stable isotopes of natural elements (C, N) to assess use of aquaculture wastes
- Preliminary data on juvenile lobster in 2015
- 2016: collecting data on multiple species from bio-collectors

Current and future work

- Received funding from Environment and Climate Change Canada's Gulf of Maine Initiative
  - 2016 and 2017 work with the bio-collectors
    - Biodiversity
    - Metals
    - Stable isotopes
- Engagement of stakeholders & government agencies: Cooke Aquaculture, FNFA, DAAF, DFO
  - Working to engage other stakeholders
  - Stakeholder workshop in March 2018

Acknowledgements

- Funding: SSHERC, UNB Environmental Studies and Research Projects Fund, Environment and Climate Change Canada, NSERC, NBIF
- FNFA
- The many students who assisted with collector deployment and processing
- Huntsman Marine Science Centre staff and Fundy Spray

This project was undertaken with the financial support of:
Ce projet a été réalisé avec l'appui financier de:

Environment and Climate Change Canada | Environnement et Changement climatique Canada

Social Sciences and Humanities Research Council of Canada | Conseil de recherches en sciences humaines du Canada

Canada

Institut de recherche sur les zones côtières inc.

Coastal Zones Research Institute Inc.

Aquaculture Development and Profitable Commercialization of Arctic Charr in Canada

« Développement aquacole et commercialisation profitable de l'omble chevalier au Canada »

Rodrigue Yossa, Ph.D.

Project Leader

The Arctic Charr Project

- Atlantic Innovation Fund (AIF) project - Atlantic Canada Opportunities Agency

- Promotor: Coastal Zones Research Institute Inc., Shippagan, NB

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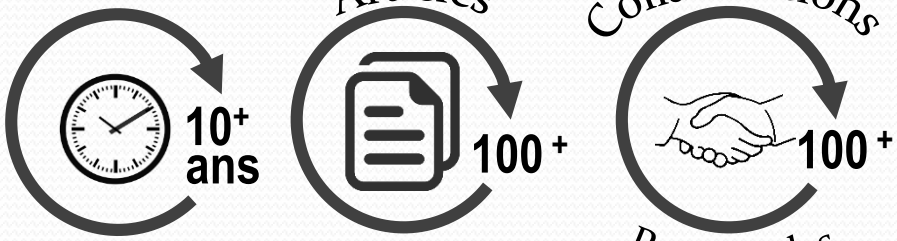
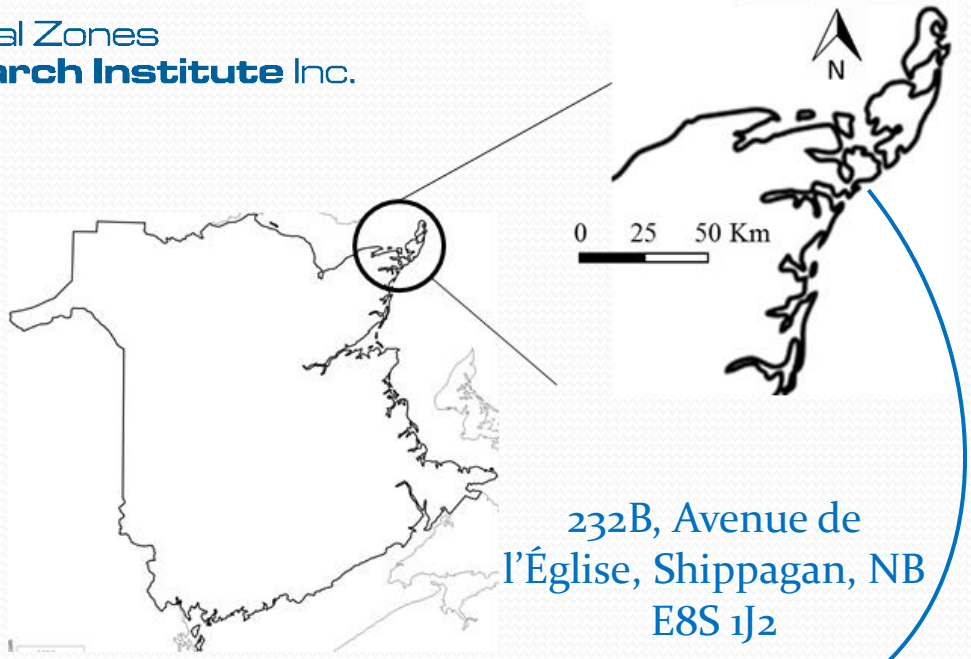
sur les zones côtières inc.

Coastal Zones
Research Institute Inc.





232B, Avenue de l'Église, Shippagan, NB
E8S 1J2

- 45 employees
- 5 PhD, 7 MSc, 12 BSc

- 3 locations

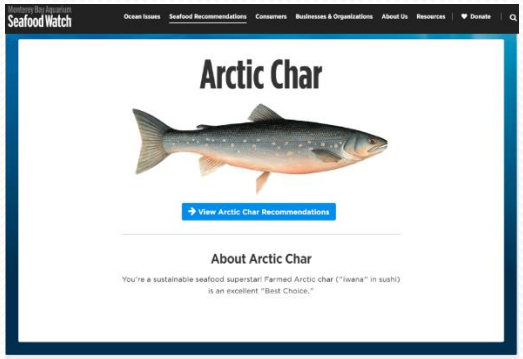
Pavillon principal,

Pavillon aquacole

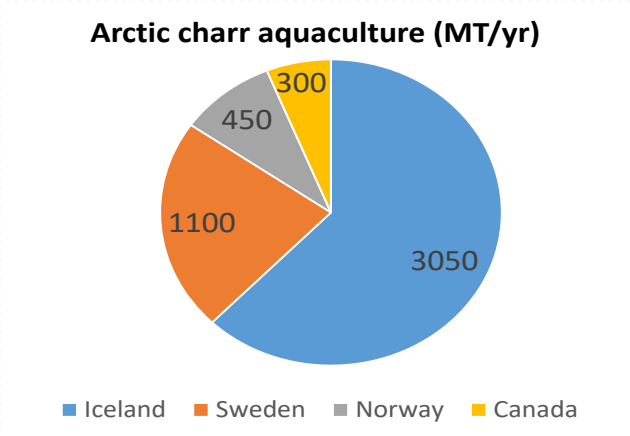
Facilities at CCNB, Caraquet

Why Arctic Charr?

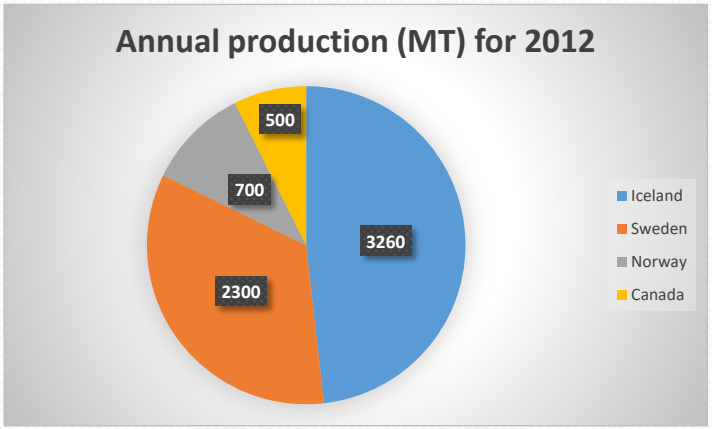
- Coldwater fish
- *Salvelinus alpinus*; 3 mains strains in Canada: Nauyuk, Tree River and Fraser
- Summerfelt et al. (2004): Arctic char
  - ✓ tolerate high-density culture conditions,
  - ✓ have an excellent fillet yield,
  - ✓ are amenable to niche marketing, and
  - ✓ are suitable for production within super-intensive recirculating systems.
- The Monterey Bay Aquarium's Seafood Watch (SFW)
  - ✓ Green "Best Choice" → well managed and farmed in environmentally friendly ways



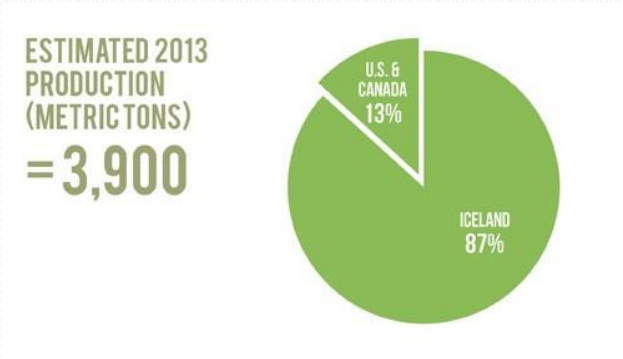
Arctic Charr Industry Update



Source: NorthCharr Project (2009-2012)



Source: Saether et al. (2013); Fisheries NO (2014)



Source: Fishchoice.com (2016)

- Incomplete/inconsistent statistics (in MT):

2011 Total = 4,900 (no US)

2012 Total = 6,760 (no US)

2013 Total = 3,900 (Iceland, Canada & US)

- 2/3 of Arctic charr is farm raised

Objective of the Arctic Charr Project

To sustainably develop Arctic charr aquaculture in Canada, through collaborative efforts between government agencies, scientists and producers.

The specific objectives of the projects represent each of the project's activities.

Relevance of the Arctic charr Project

- Canadian Arctic charr: Good reputation, Best choice, “green” farming
- Physical characteristics: Availability of land, water, technology
- Science: Best Arctic charr scientists
- Farming expertise: Highest
- Strains: Best in the World
- Public awareness: Growing
- Canada in 2015-2016:
- ✓ The Trans-Pacific Partnership (TPP) - free-trade area spanning from Chile to Japan (12 countries) +
- ✓ Canada-European Union: Comprehensive Economic and Trade Agreement (CETA)
- ✓ Canada-Korea Free Trade Agreement (CKFTA)
- Competition: None (Iceland?)

Canada · Europe
DIALOGUE TRANSATLANTIQUE
TRANSATLANTIC DIALOGUE

http://www.theglobeandmail.com/report-on-business/international-business/what-is-tpp-understanding-the-new-pacific-tradedeal/article26648948/

http://carleton.ca/ces/cu-events/the-canada-eu-comprehensive-economic-and-trade-agreement-the-never-ending-story/

Duration of the Arctic Charr Project: 2014-2019

Atelier international sur l'omble chevalier / International Workshop on Arctic Charr
Moncton, Nouveau-Brunswick - Canada - 2011

Institut de recherche sur les zones côtières inc.
Coastal Zones Research Institute Inc.

Mâle et Femelle (géniteurs) de l'IRZC - Shippagan - NB "ACADIAN CHARR"

Kickoff Meeting: *April 30th-May 2nd 2014*

Centre des congrès (rue de l'Aquarium) - Shippagan, Nouveau-Brunswick

Partners Involved in the Arctic Charr Project

- **Government and allied agencies:**

  - New Brunswick Innovation Foundation (NB)
  - Department of Agriculture, aquaculture and Fisheries (NB)
  - North Fund-Regional Development Corporation (NB)
  - BioAtlantech (now BioNB) (NB)
  - Department of Agriculture and Fisheries (NS)
  - Agri-Food Research & Development Initiative (Manitoba)
  - Ministère de l'Agriculture, Pêches et Alimentation du Québec (QC)
  - Ontario Ministry of Agriculture, Food and rural Affairs (ON)

- **Arctic charr industry:**

  - Icy Waters Ltd. (ON and Yukon)
  - Aquaculture Gaspésie Inc. (QC)
  - Delicasea (BC)
  - Ridgeland Aqua Farms Ltd. (Manitoba)
  - Pisciculture Acadienne (NB)
  - Pisciculture CJL (NB)
  - Parc Atlas (NB)

- **Universities and research institutes**

  - Dalhousie University; Truro and Halifax campuses (NS)
  - University of Guelph (ON)
  - Atlantic Veterinary College-UPEI (PEI)
  - NB Research and Productivity council/RPC (NB)
  - Université du Québec à Rimouski (UQAR)
  - Collège Communautaire du Nouveau Brunswick (NB)
  - Coastal Zones Research Institutes Inc. (NB)

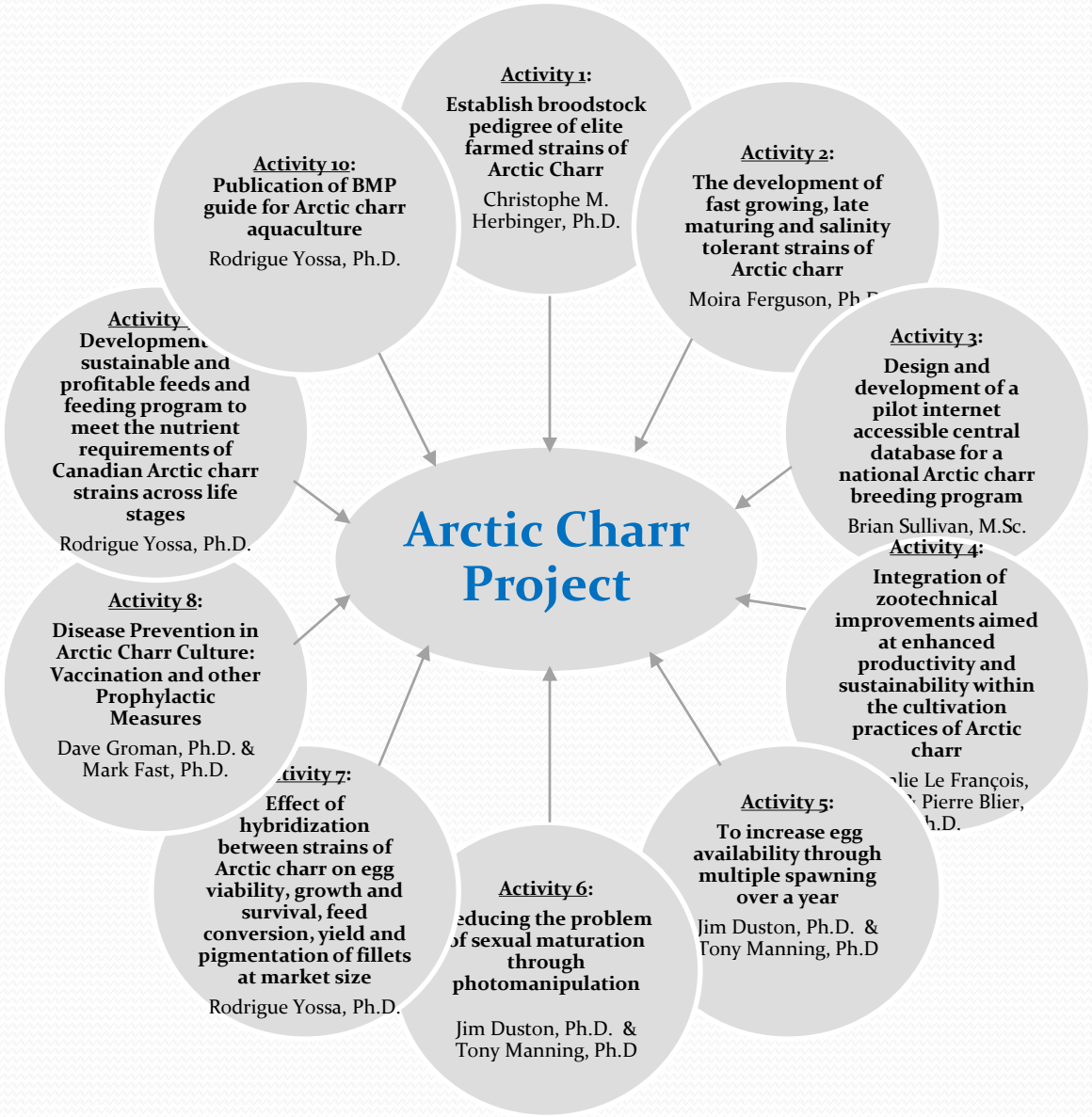
Institut de recherche sur les zones côtières inc.
Coastal Zones Research Institute Inc.

Second Meeting: *May 27-29, 2015*
Four Points by Sheraton Moncton, Moncton, New Brunswick

Institut de recherche sur les zones côtières inc.
Coastal Zones Research Institute Inc.

Last Meeting: *May 25-27, 2016*
New Brunswick Aquarium and Marine Centre, Shippagan, New Brunswick

Scientific Activities



Next steps of the Arctic Charr Project

- Complete the Scientific activities (and publish the results?)
- Develop sustainable market for Arctic charr eggs (and fry?)
- Spur industry interest in Arctic charr aquaculture
- Improve the productivities in current Canadian farms

High Ambitions

"Keeping temperature optimal year-round at an economic and sustainable rate is basically the alpha and omega of fish farming. Our new farm will do exactly that and will be producing a fish in high demand worldwide", said Árni Páll Einarsson, CEO

http://advocate.gaalliance.org/matorka-aims-to-unearth-innovation-with-arctic-char/

- Company: Matorka Holdings AG
- Country: Iceland
- Facility: world's largest land-based salmonid farm
- Water: groundwater supply, partial recirculating
- Production: Arctic charr aquaculture expected to grow from 50 metric tons (MT) to more than 3,000 MT
- Energy: "cheap" geothermal heat
- Sustainability; BAP standards & certification → carbon-neutral, chemical-free, antibiotic-free product
- First harvest: summer 2017

Thank you!

Questions, comments?

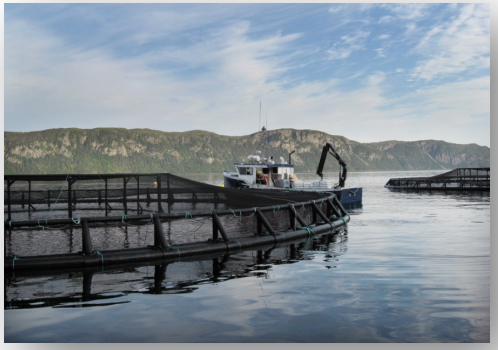
Pêches et Océans Canada | Fisheries and Oceans Canada

Genomic tools identify impacts of escaped farmed salmon

Ian Bradbury
Fisheries and Oceans Canada
Memorial University, Newfoundland
Email: ian.bradbury@dfo-mpo.gc.ca

Canada

Outline



- Background

- Approach

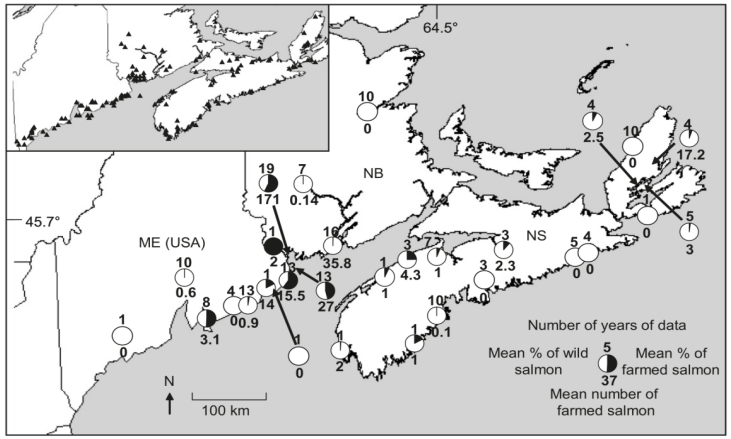
- Application

- Summary / Future steps

Salmon aquaculture expanding globally with impacts on wild salmon

- Number of salmon in culture exceeds those in the wild
- Farmed salmon escape
- Genetic and ecological interactions
- Genetic changes (50% of Norwegian rivers, Karlsson et al. 2016)

Escapes in Atlantic Canada



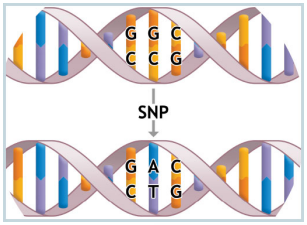
Morris et al. 2008 CJFAS

- Escapes in both Maritimes and Newfoundland
- Reports localized to region around industry
- Large escape event in Newfoundland 2013
- Interactions largely unknown

Approach

1) **Develop** genomic tools

1)

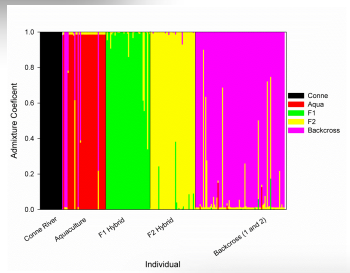


2) **Apply -** sample baselines and testing salmon throughout region

2)

3) **Measure** - levels of introgression and frequency of hybridization

3)



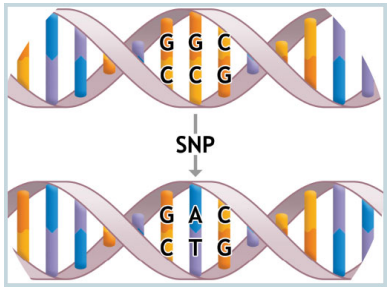
4) **Model interactions -** as functions of proximity and wild population size to **inform decision making**

4)

Genomic tool development

Goal: to identify region specific panels of collectively diagnostic markers

1) **Genome scans:** based on the 5.6K and 220K Atlantic salmon SNP arrays (CIGENE)



2) **Panel development:** markers that differentiate wild and farmed salmon as well as hybrids

3) **Rapid and efficient assays for screening:** assays developed for rapid screening, significant investments in infrastructure and training (Aquatic Biotechnology Lab)

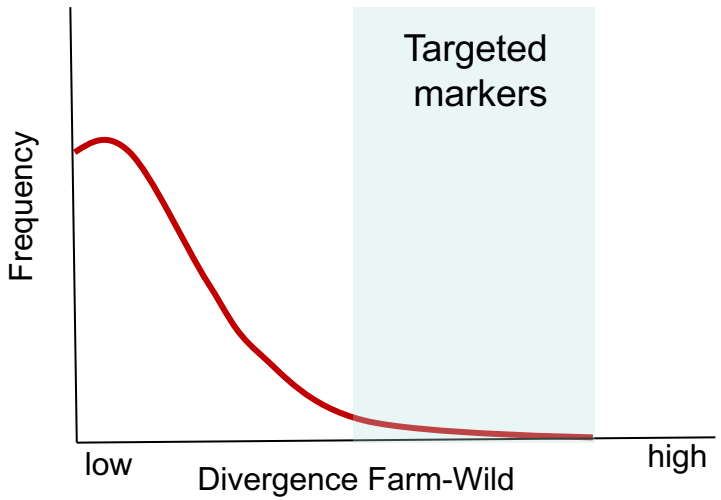
Search for “diagnostic” markers

Farmed lines differ from wild populations at both neutral and adaptive regions of the salmon genome

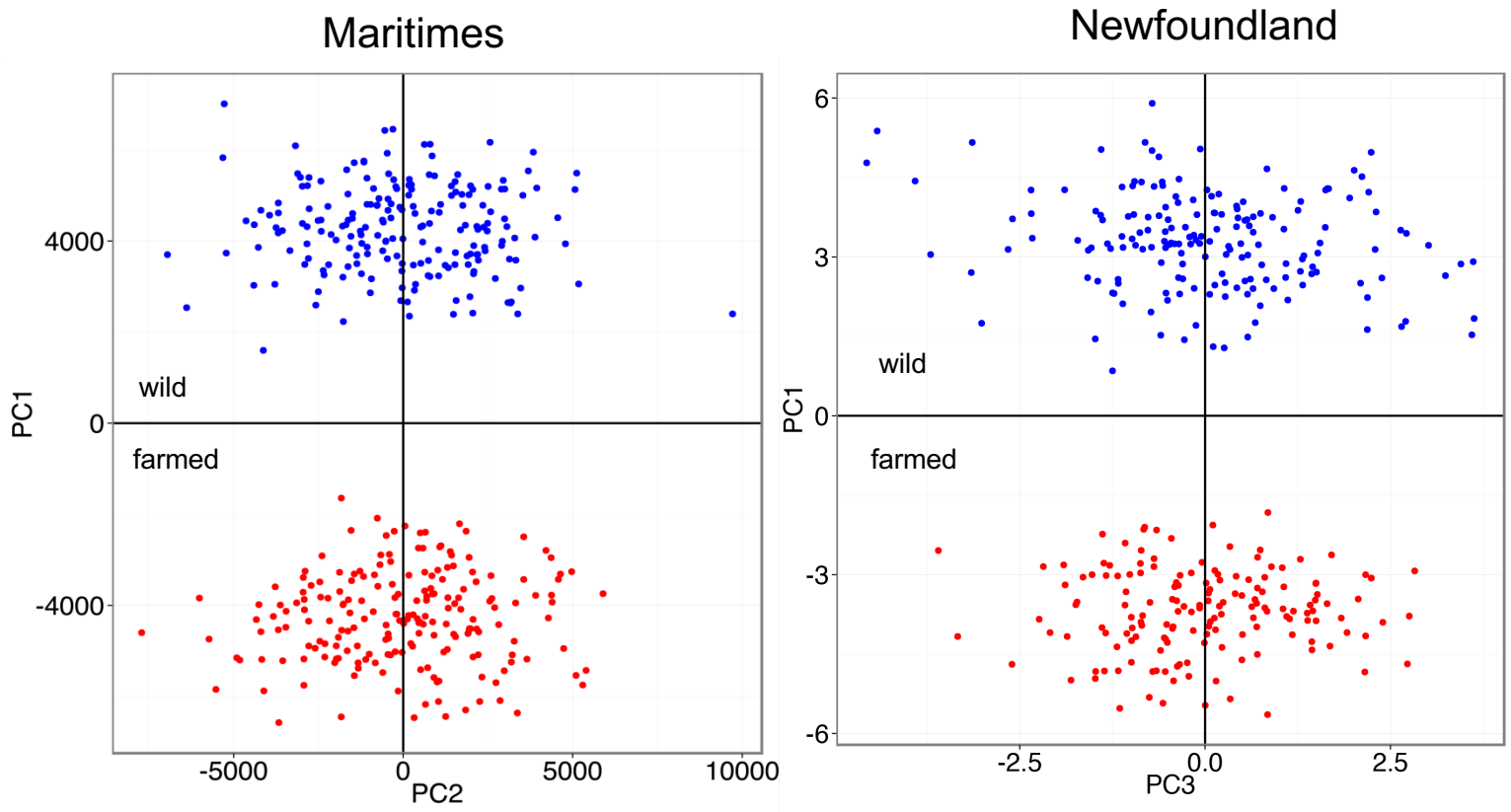
Lines (populations) chosen, domestication, and random genetic drift

Targeting regions that differ the most can maximize power for resolving potential impacts

Custom R scripts to select highly divergent independent loci (Genepopedit)



Diagnostic Panels of SNPs



Panels of diagnostic markers

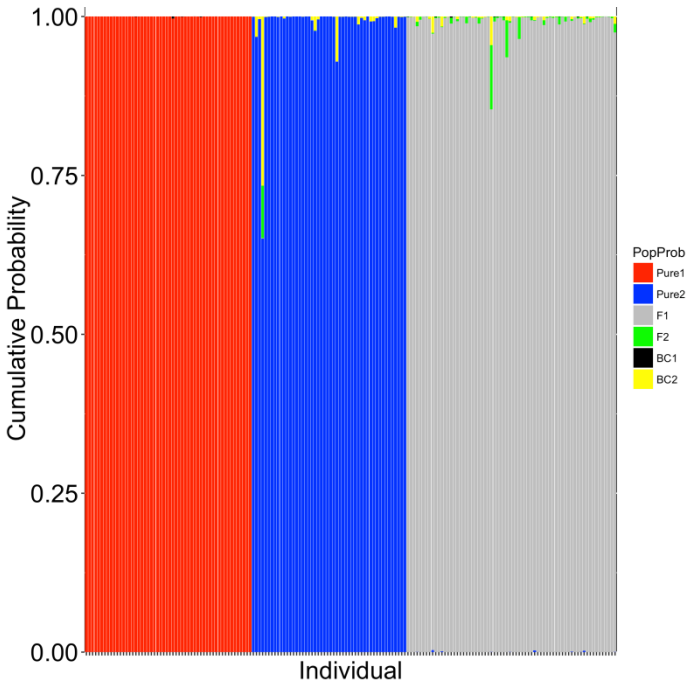
Hybrid ID - Accuracy and Power

1) Methods development

R packages for data manipulation, simulation and power analysis, and parallel processing of hybrid identification

2) Simulations and bias correction

Power evaluated using simulated hybrids and re-sampling, high grading bias addressed, highly accurate assignment



3) Validation using lab reared hybrids

Lab reared first generation (F1) hybrids used for independent power analysis, 100% accurate assignment

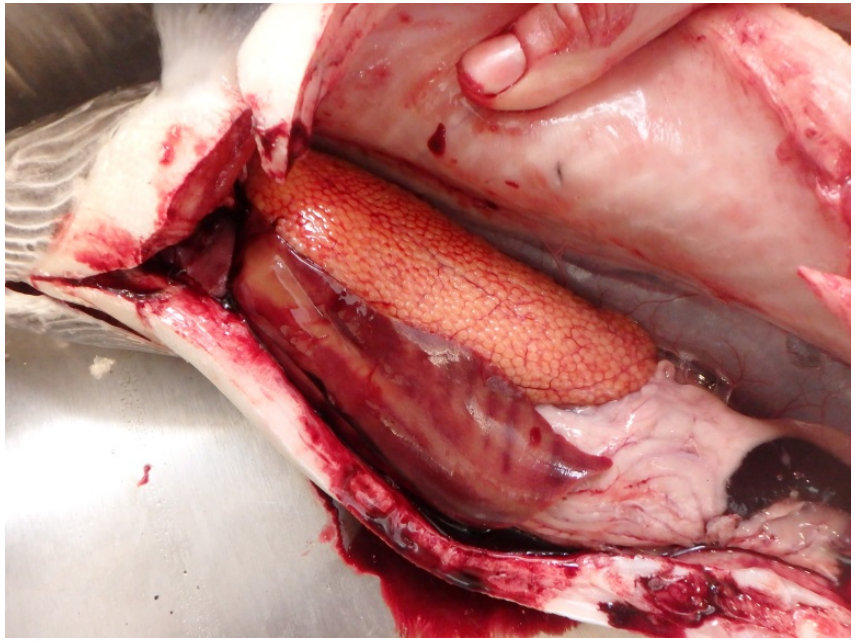
Application – Southern Newfoundland

Juvenile samples collected to screen for hybrid ancestry following 2013 escape (20K fish escaped)

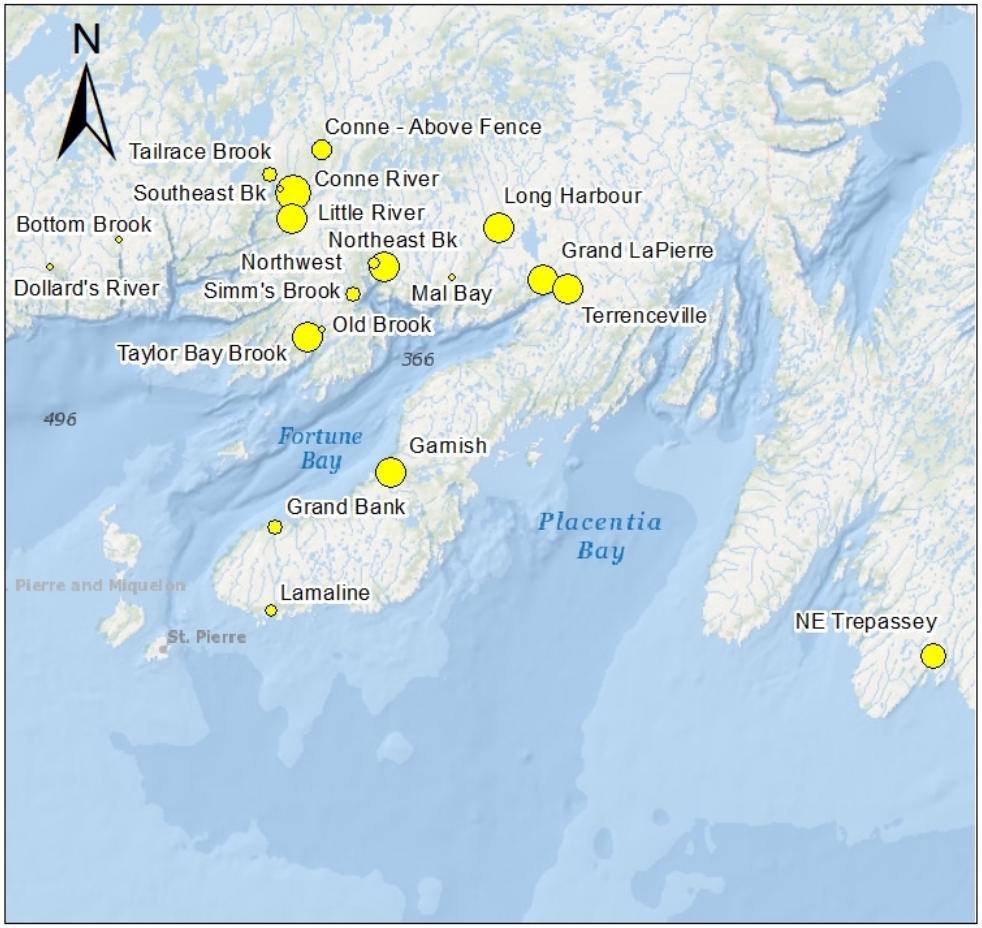
South coast population ~20K wild salmon

2014, 2000 young of year Atlantic salmon collected, 18 locations

2015 / 2016 ~1500 young of year, and Atlantic salmon parr collected for screening



Juvenile salmon collection 2014



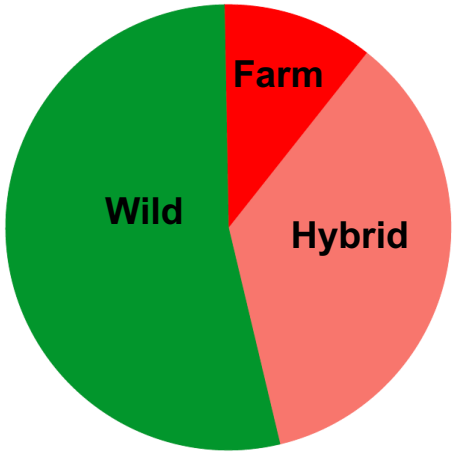
Electrofishing Totals

Summer 2015

Young of the Year

- 18 - 35
- 36 - 70
- 71 - 105
- 106 - 140
- 141 - 175
- 176 - 210
- 211 - 245

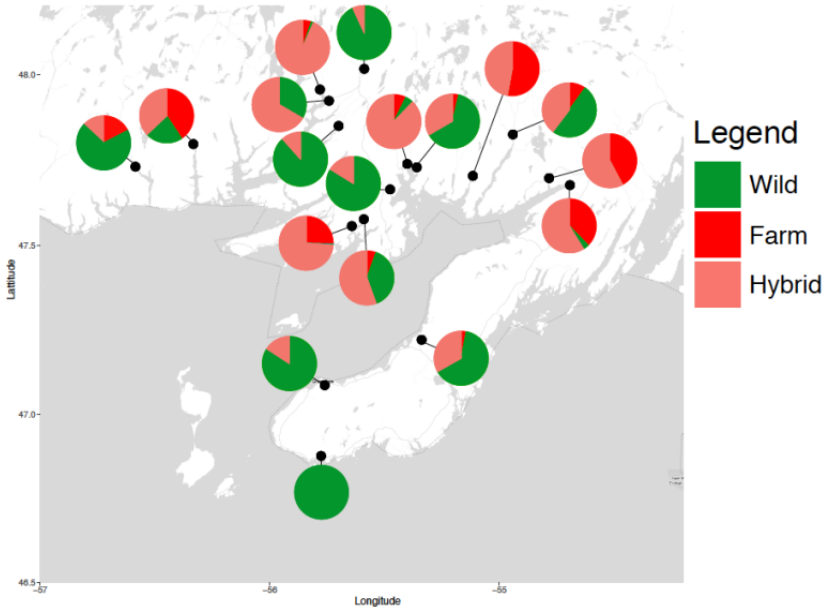
Evidence of hybridization (2014)



Data supports hypothesis of hybridization of wild and farm escaped salmon in southern Newfoundland following 2013 escape

17/ 18 rivers show evidence of hybridization (~1/3 overall individuals)

Evidence of successful pure escape reproduction at several locations



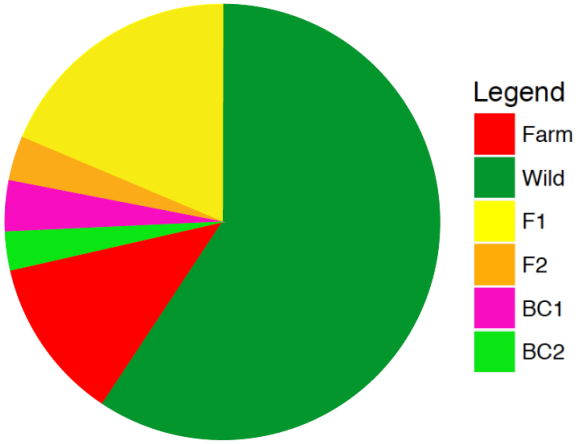
Evidence of hybridization

Breakdown of hybrids into classes

Strong support for the presence of other hybrid classes such as F2, BC1 and BC2

Hybridization prior to 2013 event and ongoing

Hybrids are viable and long term genetic impacts possible

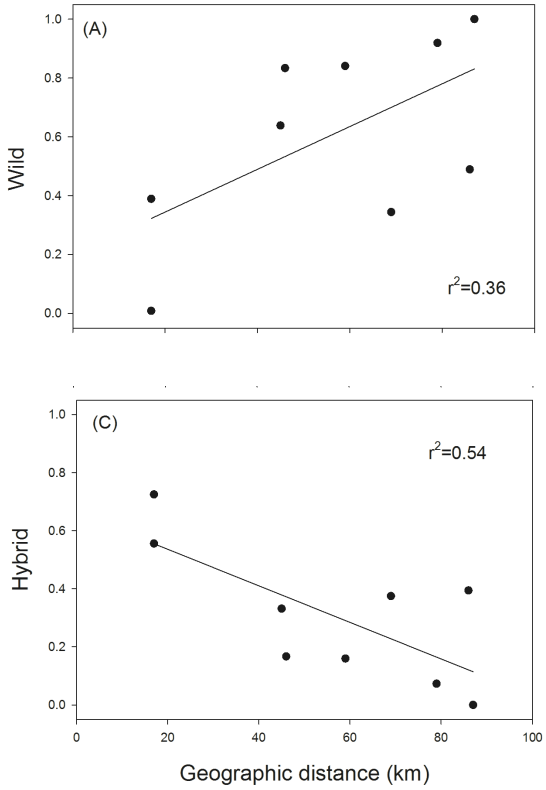


Geographic distance and impacts

Geographic distance relative to the 2013 escape location

Proportion of wild at each location increases with distance

Proportion of hybrids decreases with distance



Consistent with expected source

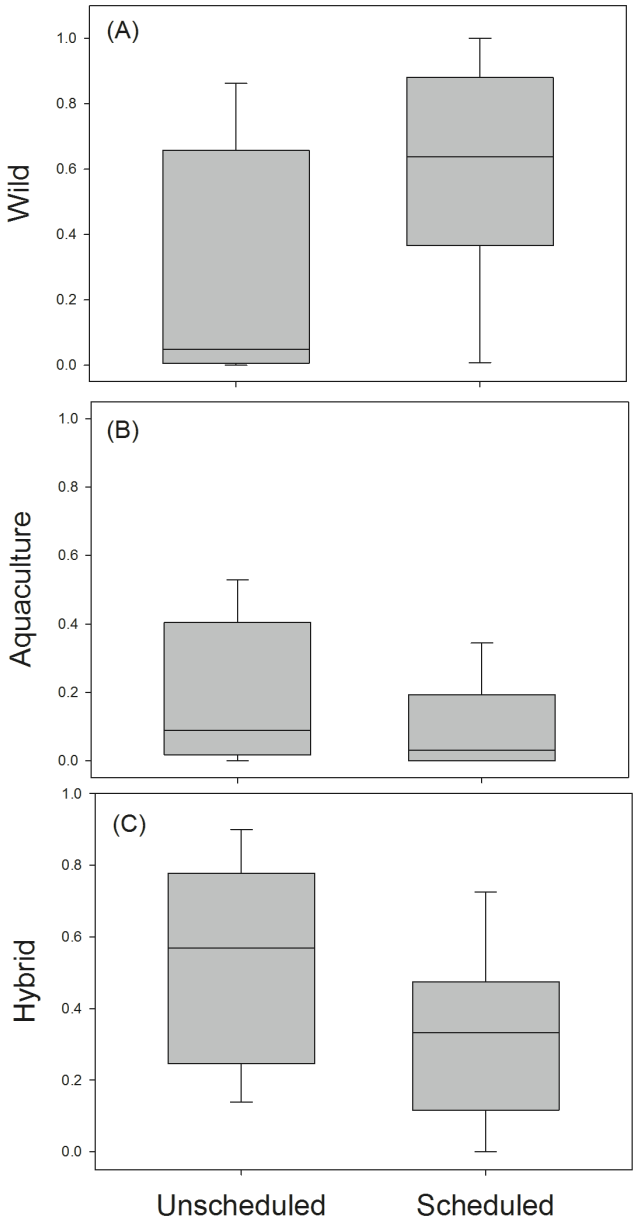
Scheduled vs non-scheduled rivers

Scheduled proxy for large and small populations

Scheduled and non-scheduled rivers differ in proportions

Scheduled rivers have more wild juveniles and less hybrids

Consistent with greater impacts on smaller populations

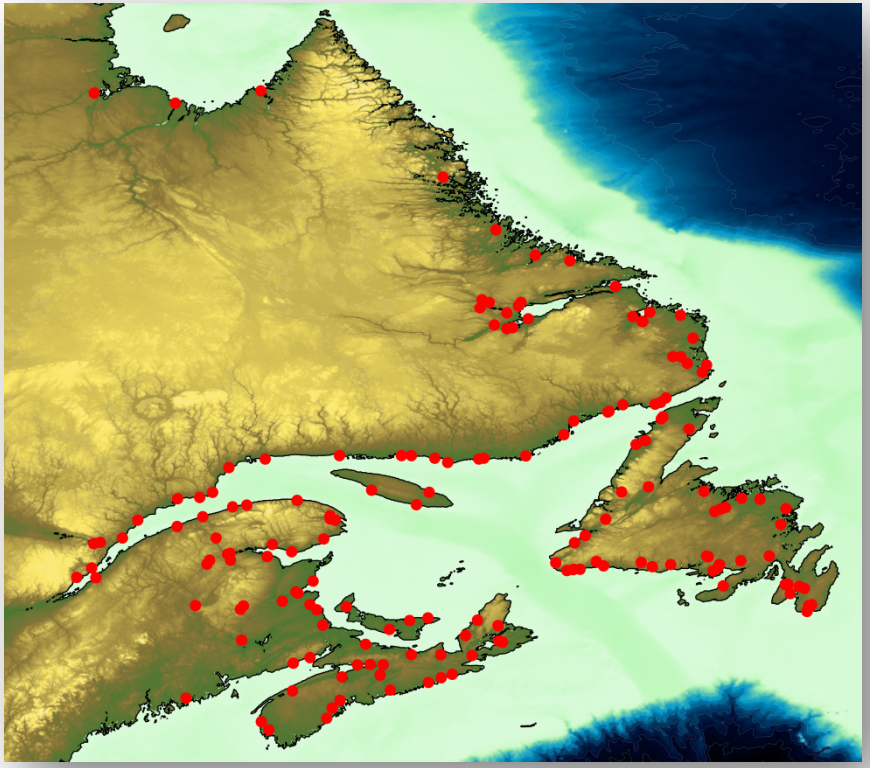


Range wide evaluation

North American range-wide examination of introgression

North American SNP baseline (n=1818 SNPs, 1710 individuals), including several aquaculture samples (Moore et al. 2014, Bradbury et al. 2015, Jeffery in prep)

All samples collected prior to 2013; Bayesian analysis of introgression following Karlsson et al. 2016



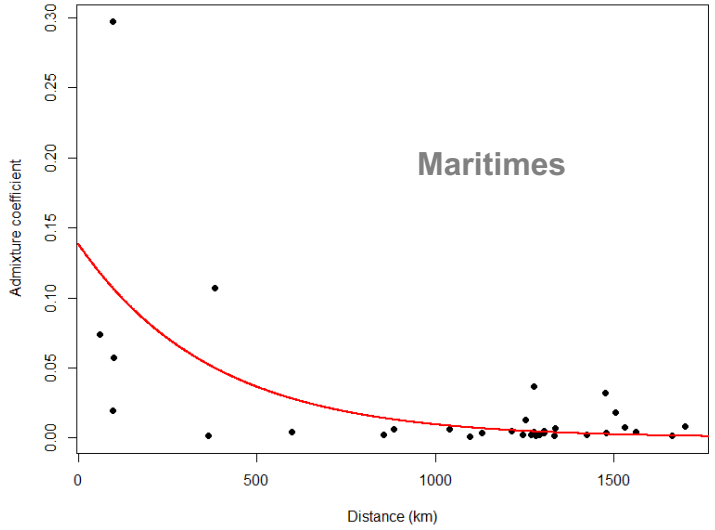
Range wide evaluation

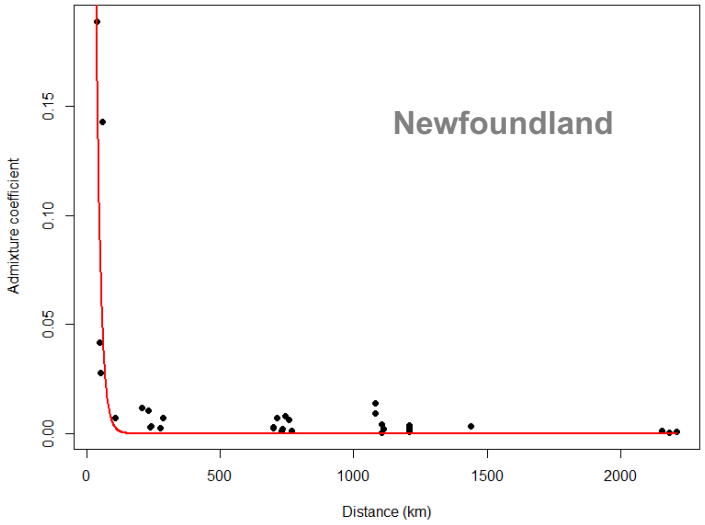
Evidence of declines in impact with distance from industry both in Newfoundland and Maritimes

Maritimes beyond 100-500 km shows little evidence of introgression – Bay of Fundy

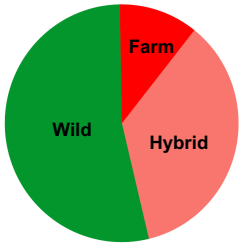
Southern Newfoundland beyond 100 km shows limited introgression – Bay d'Espoir and Fortune Bay

Amount of introgression similar across regions (15-30% at fine scales)

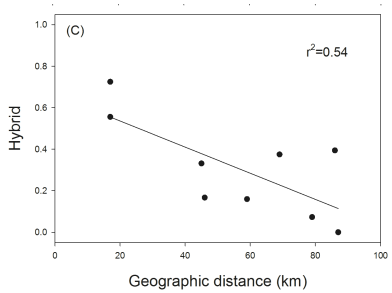




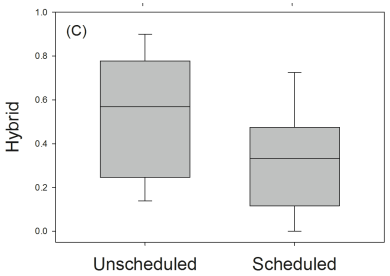
First steps towards understanding interactions



Robust and reliable tool for quantifying wild and farmed interactions in Atlantic Canada

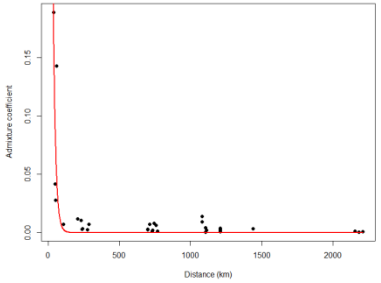


Recent hybridization and levels of successful farm-farm reproduction now measurable



Scales of impact in Newfoundland suggest <100 km (both hybridization and introgression analysis)

Impacts higher in smaller rivers consistent with observations from Norway



Beginning to spatially model the impacts (additional years, ages, and locations being analyzed)

Future questions – so what?

Impact on survival - variation in relative survival of hybrids across freshwater and marine stages

Impact on population productivity - models of potential population-level effects

Estimates may not be representative of all watersheds, most samples from lower stretches

Annual variation (2014?), samples from 2015 and 2016 for comparison, continued monitoring funded

Acknowledgements

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Industry Partners

Cooke Aquaculture
Northern Harvest Sea Farms

Fisheries and Oceans Canada

Program for Aquaculture Regulatory Research

Genomics Research and Development Initiative

NSERC CRSNG

Atlantic Salmon Federation
Fédération du Saumon Atlantique

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Huntsman Marine Science Centre, St. Andrews, NB

Genetic Traceability

Amber Garber
Huntsman Marine Science Centre
St. Andrews, New Brunswick
agarber@huntsmanmarine.ca













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What is Traceability?

- ✓ Ability to identify individuals over time
- ✓ Traceability is often discussed related to a grocery store and consumer
  - Ability of a customer to view the life history of a product (e.g., a fillet when it was part of a fish)
- ✓ In this presentation – ability to identify cultured Atlantic salmon, found in the wild, to a sea cage, hatchery, company or (most detailed) a family

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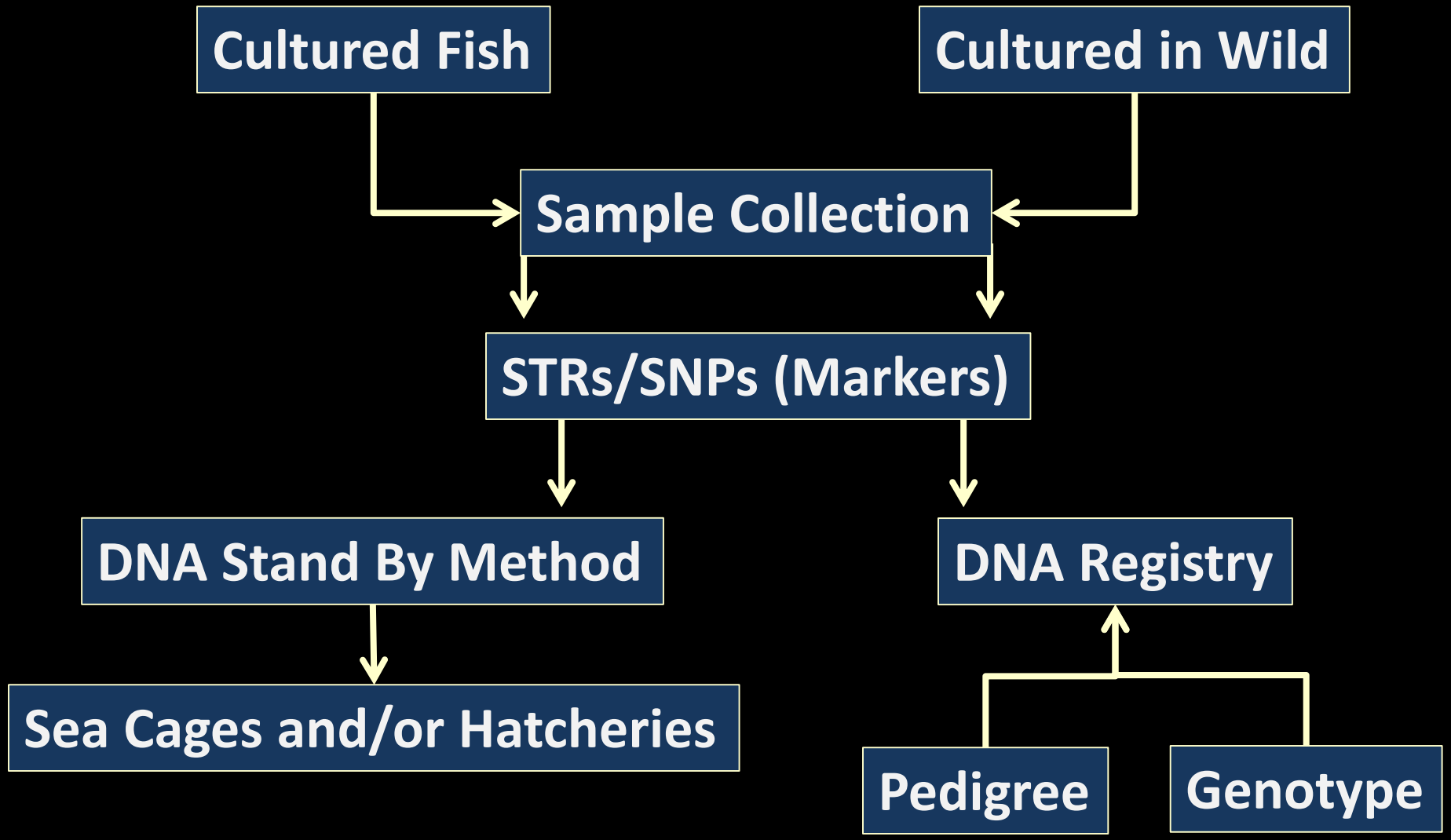
Traceability



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Collecting Samples

- ✓ Sampling easy and minimally invasive
- ✓ Likely would be collecting fin clips on live fish
  - Can be as simple as netting fish on a sea cage
  - Amount of tissue needed is minimal (e.g., piece of rice and probably gnat size actually needed/used)
- ✓ Tissue from live fish of the best quality

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Types of Genetic Technologies

- ✓ There are various types of technologies that can be used to identify individual fish from a common group or individual fish to a family
  - DNA easiest because slowest to degrade (sample doesn't have to be from a live fish)
- ✓ There are two types of markers that are presently being used in Atlantic Canada to identify groups and families – microsatellite markers (short tandem repeats, STRs) and single nucleotide polymorphic markers (SNPs)

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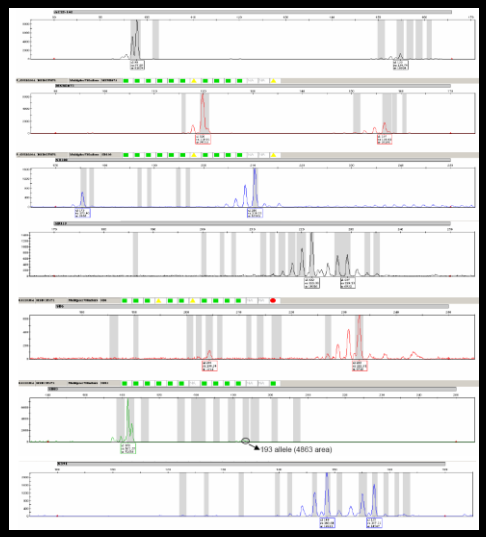
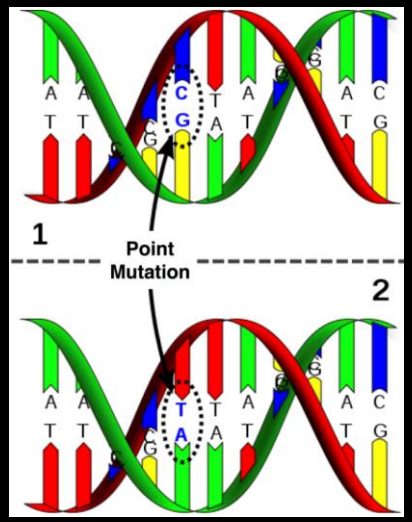
Inheritance

- ✓ An Atlantic salmon DNA is similar to a human (in a sense)…
  - One copy of our DNA comes from our mother
  - One copy of our DNA comes from our father
- ✓ A marker is typically referred to as a locus
- ✓ There are two alleles at each locus





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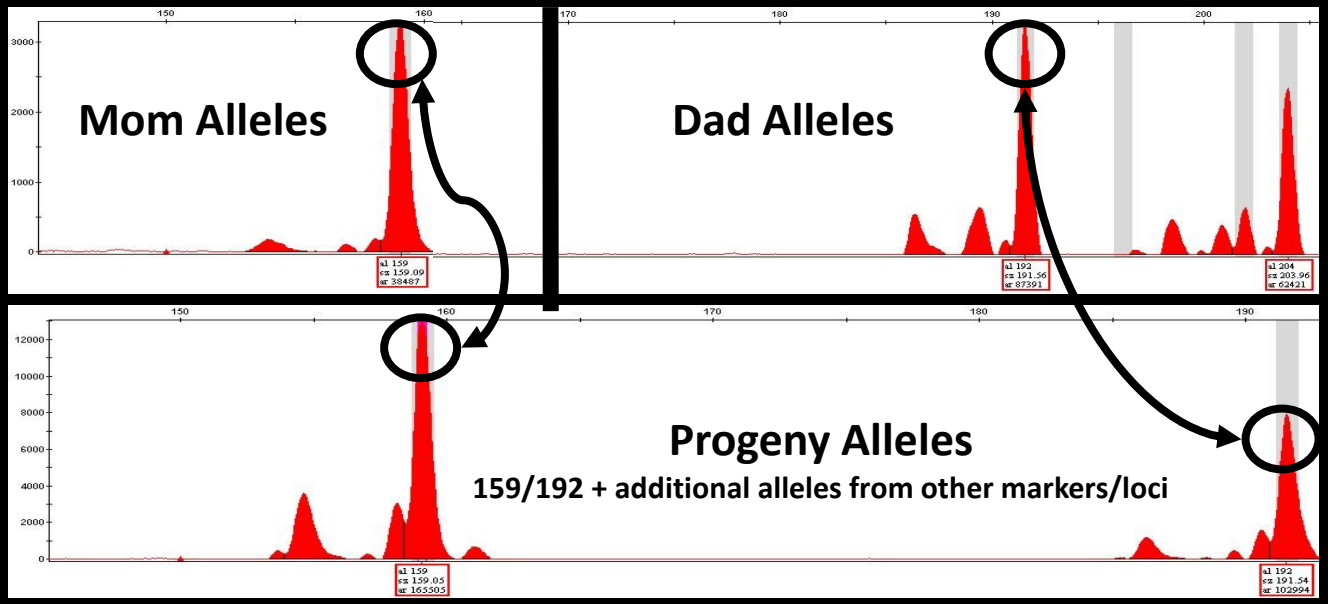
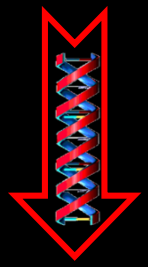
Marker Inheritance



Inheriting two alleles of up to four possibilities (1 from mom, 1 from dad)

Number of alleles at a locus and frequency dependent on population



Clustering or Identification (Statistical Programs)
Various Programs Used – Structure, Cervus, Probmax

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Let's Define Type of Traceability

- ✓ DNA Stand By Method
  - Being considered for & has been used in Norway
  - Identifies origin of escaped farmed salmon
  - Used when a large group of cultured individuals identified in wild
  - DNA samples collected from farms/sea cages nearby with similar sized individuals
  - Estimate probability escaped salmon are from sampled farms (genetics + statistics)
    - Plus may use biological data

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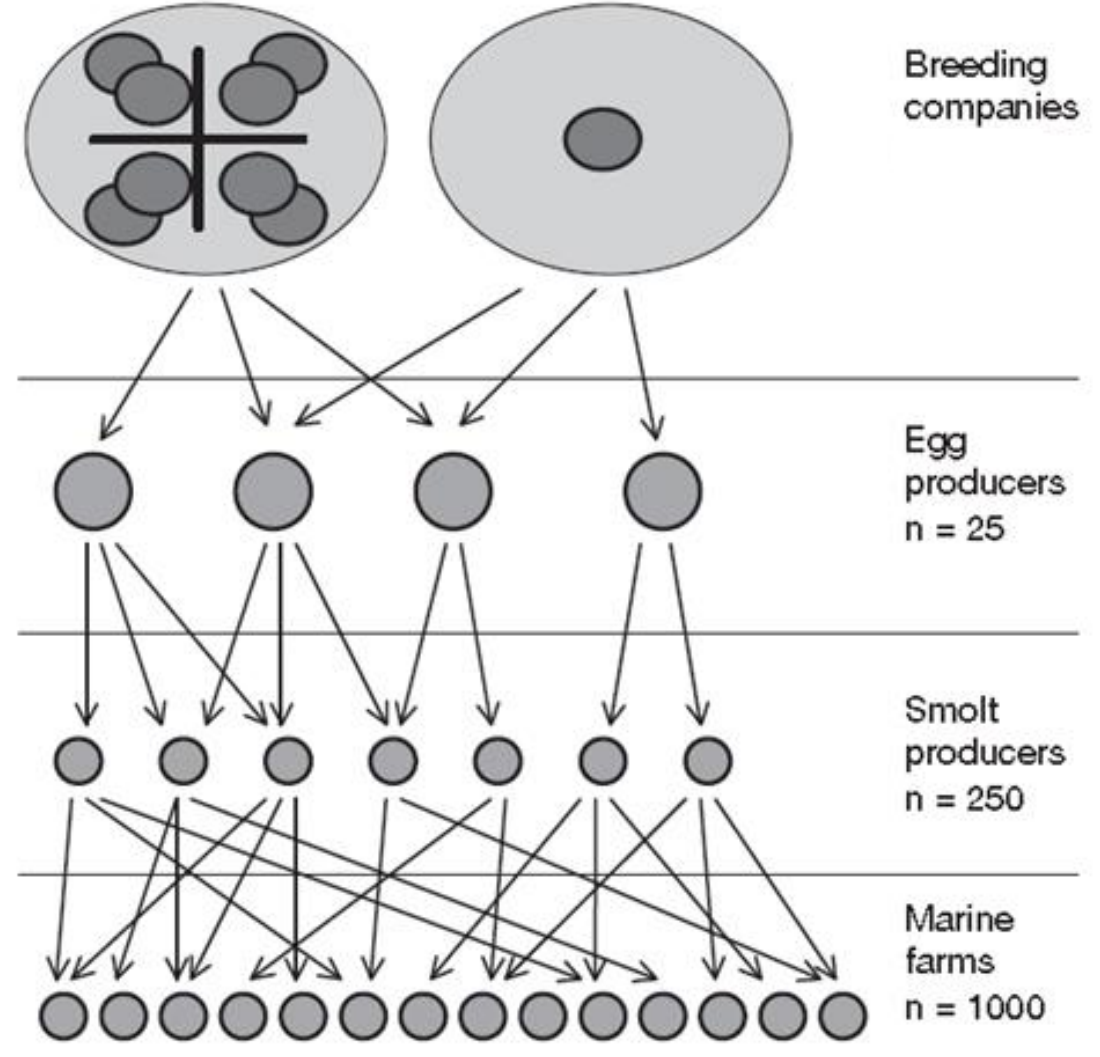
Supply Chain Example



Glover 2010 Aquacult Environ Interact

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Let's Define Type of Traceability

- ✓ DNA Registry
  - Also being considered for use in Norway
  - Similar to program implemented in Maine
  - Cultured salmon found in the wild could be traced back to individual families from a company (e.g., if genotypes and pedigrees provided) or may be traced back to a sea cage then hatchery (e.g., if DNA samples collected from each cage and hatchery)

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Broodstock/Selective Breeding Programs

- Critical if discussing implementation of a DNA Register or Registry program
- Retain genetic diversity and prevent inbreeding
- Improve traits to reduce cost of production, increase health of animal, faster time to harvest
- A broodstock program provides the foundation to more easily allow for traceability (at the level of genotype) from gamete contributors (parents) to heath tray/jar/tank → tank → sea cage → processing plant → → plate

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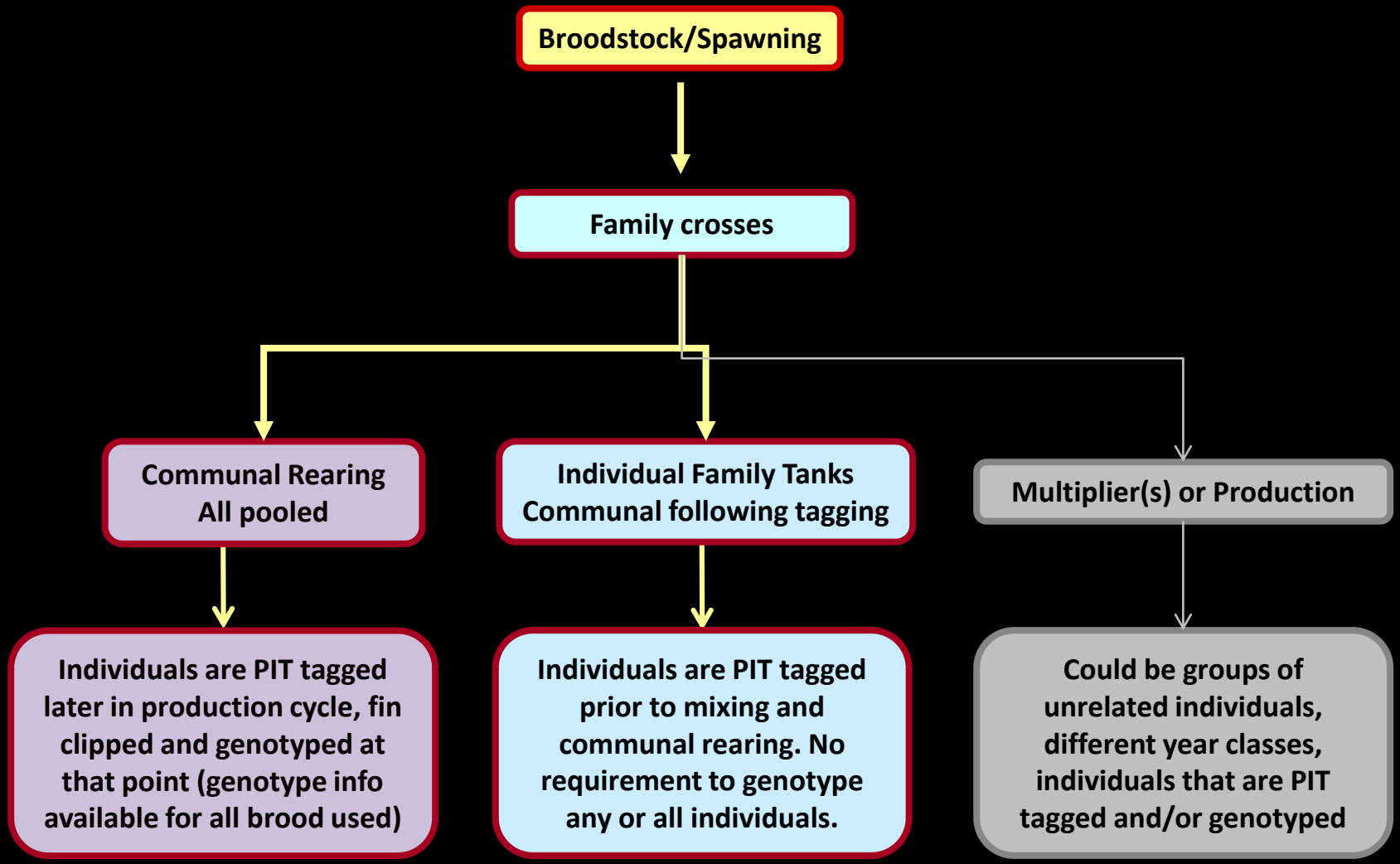
Breeding Programs (High Level Overview)



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Challenges in Identification

- Production of fish that retain genetic variation, minimize inbreeding and are improved for all desired traits ++ ***also genetically distinct***
  - ✓ A ***breeding nucleus*** retains the most genetic diversity of a broodstock program
  - ✓ ***Multiplier groups*** or ***production*** typically have reduced genetic variability overall but maximize improvement on traits (many individuals but from best of best families)
- ALSO programs combine some genetics between year classes to allow for analysis of all year classes together (e.g, use of 5 yr olds or cryo) and/or homogenize production traits

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Challenges in Identification

- ✓ Using molecular markers to identify resistance to specific genes further reduces the number of individuals available for use
  - Selecting fish based on genotype vs phenotype (genetic makeup vs physical measurements)
  - IPN resistance – one SNP – used in Norway
    - Decreases number of individuals available for spawning that are resistant
    - New technologies – e.g., Cryogenetics whole gonadal extraction – allow for the fertilization of millions of eggs with a single male
      - Physically possible to use very little genetic variation in production

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Communal, Individual, Multipliers - Registry

- ✓ **Communal**
  - Fin clipped when tagged prior to spawning – pedigree or parentage assigned with genotype
  - Cost mainly to ‘package’ data
- ✓ **Individual**
  - Tagged from individual family tank – pedigree known
  - Genotyping of individuals required at some point
- ✓ **Multiplier(s) for production**
  - Parentage may be known/unknown, tracked or not, crossing possibly between tanks, cages, sites, year classes, etc.
  - Genotyping likely an added cost

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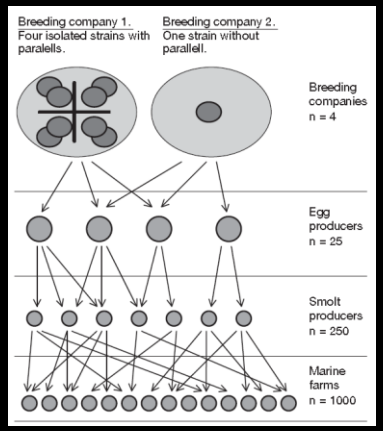
Review

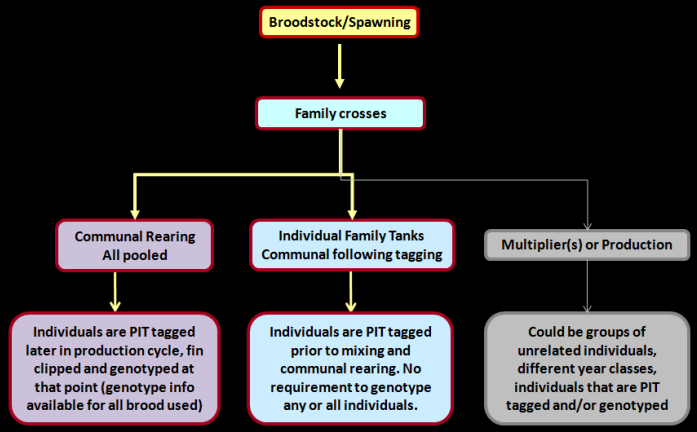
- ✓ Traceability & Genetic Traceability
- ✓ Sampling & Markers

Types of Traceability & Challenges

DNA Stand By Method



DNA Registry



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Genomic Selection and Genome-Wide Association Studies: Perspectives and Possibilities

Tiago S. Hori, Ph.D – Associate Director of Genomics

Now with AQC3 certification!

Aquaculture research facility opening in Souris

Published on August 16, 2015

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Debbie Plouffe, Center for Aquaculture Technologies Canada vice president of research, oversees the company's new Souris research facility, the only one of its kind in North America.

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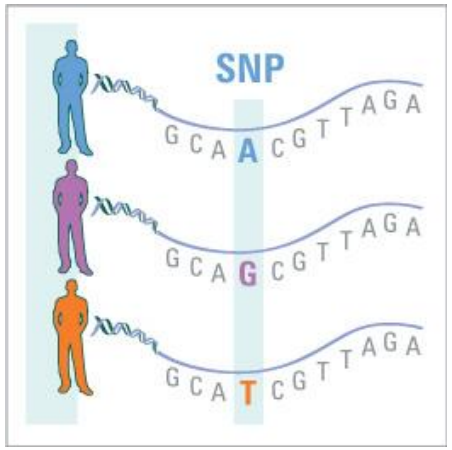
From genes to phenotypes

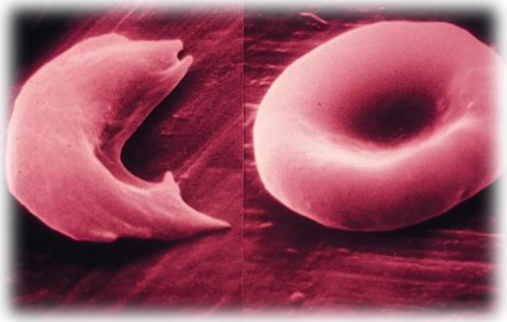
- A proportion of the variability observed in traits of animals is driven by differences in DNA sequences between individuals.
- Genes and other genetic elements that contribute to the observed variation can be identified using Genomics.

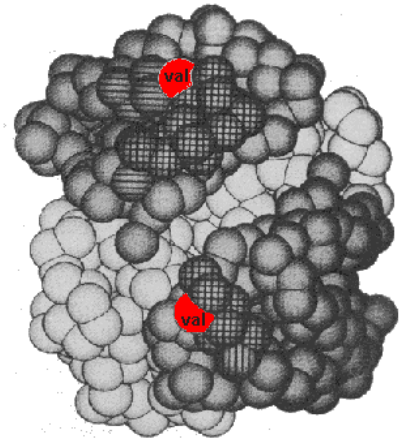
From genes to phenotypes

- Selection based on phenotypes is not a new idea.
- However, selection on phenotypes can be lengthily and costly.
- One example of genetic marker is called single nucleotide polymorphism (SNP).
- A SNP is difference in a nucleotide between individuals in a section of the genome.



- SNPs can be responsible for phenotypes, such as diseases.
- SNPs are heritable and therefore can be used for selection.





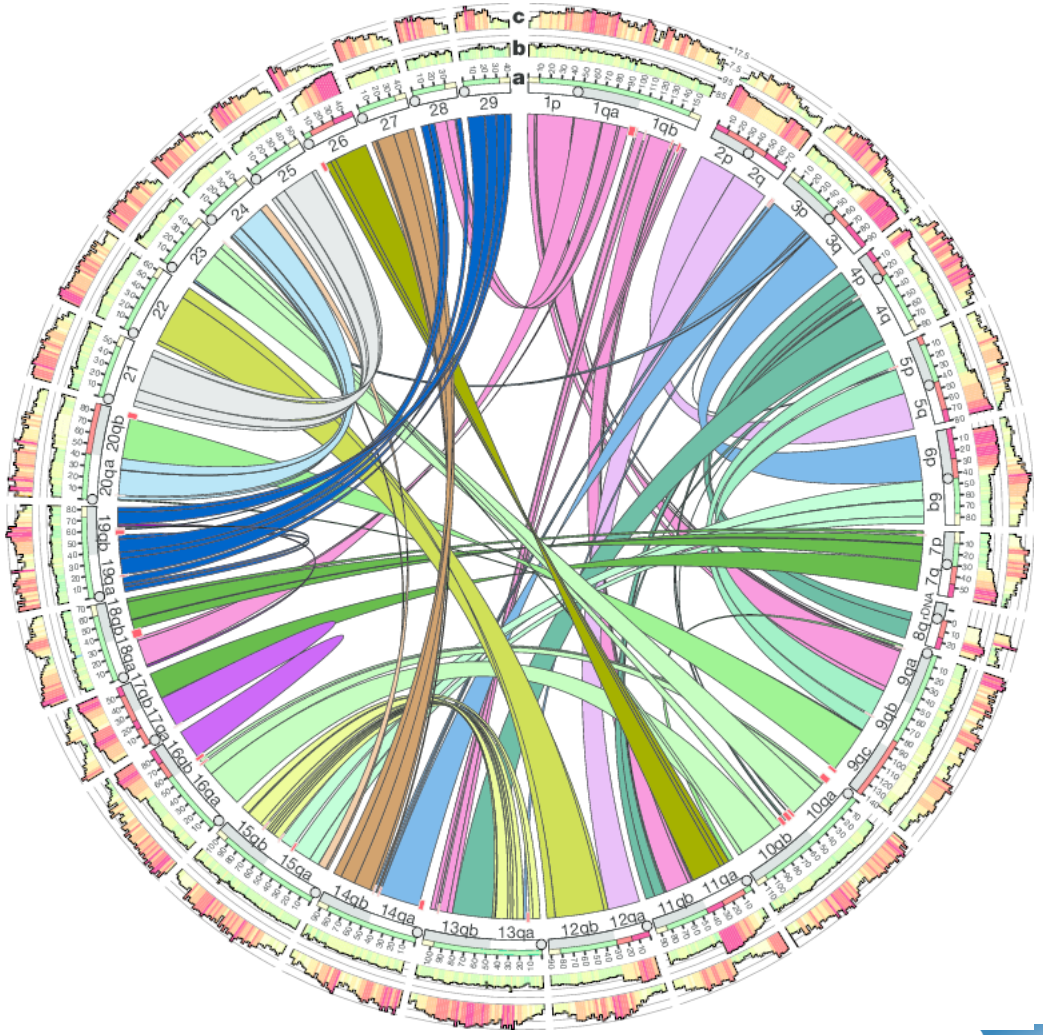
Why are SNPs relevant to Atlantic Salmon?

- In the competitive market of aquaculture, broodstock development is becoming increasingly important.
- Development of elite broodstock can reduce the use of feed and losses to disease.

- Pedigrees reconstruction is key to broodstock development.
- Marker assisted selection (MAS) can significantly increase genetic gains over a generation.
- Reduces time and cost.
- Pedigree reconstruction and MAS can be achieved using SNPs.

Genomic Resources for Atlantic salmon

- Assembled Genome
- Large number of identified SNPs
- High-density SNP arrays



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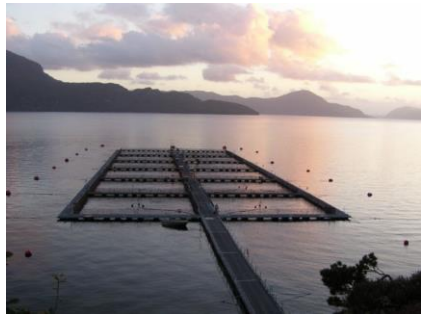
From phenotypes to genotypes

- SNPs can be linked to phenotypes and act as predictors of a phenotype.
- Predictive SNPs can be identified by correlating phenotypes with genotypes.
- This can now be done in a genome-wide fashion, leading to genome-wide association studies (GWAS).



Select many families to encompass as much variability as possible

Observe phenotypes

Genotype selected individuals (based on phenotype observations) for as many SNPs as possible. A few thousand SNPs can work, but tens of thousands are safer

Perform bioinformatics analysis of association using regression analysis to identify trait-relevant markers

Integrate markers into the broodstock development program

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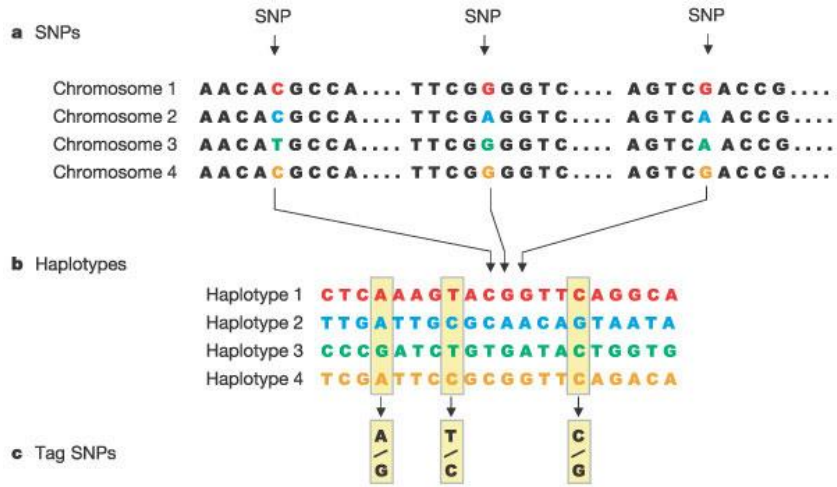
Genome wide selection (GWS)

- SNPs occur in intervals in the genome.
- Each interval represents a section of the genome that can potentially affect a trait of interest.
- If there are enough known SNPs such as these intervals are not longer than 1 cM, than the impact of each interval on a given phenotype can be estimated.
- This allows the calculation of a genetic estimated breeding value (GEBV).



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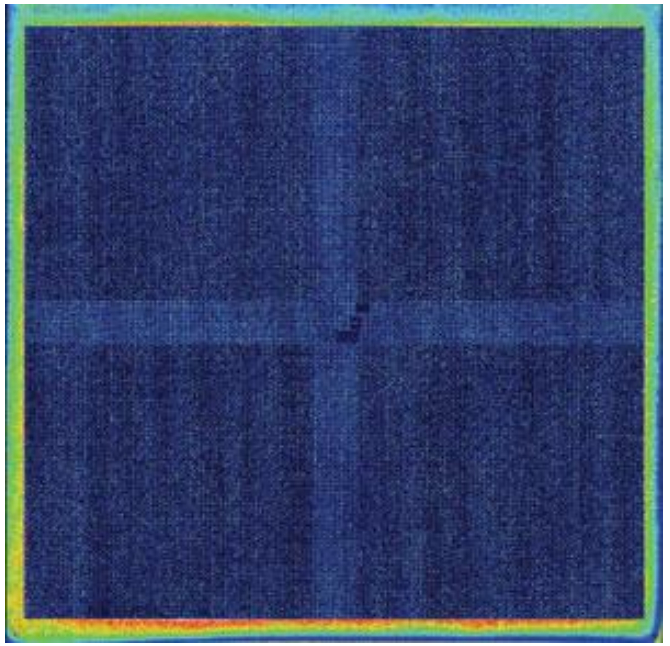
Genome wide selection (GWS)

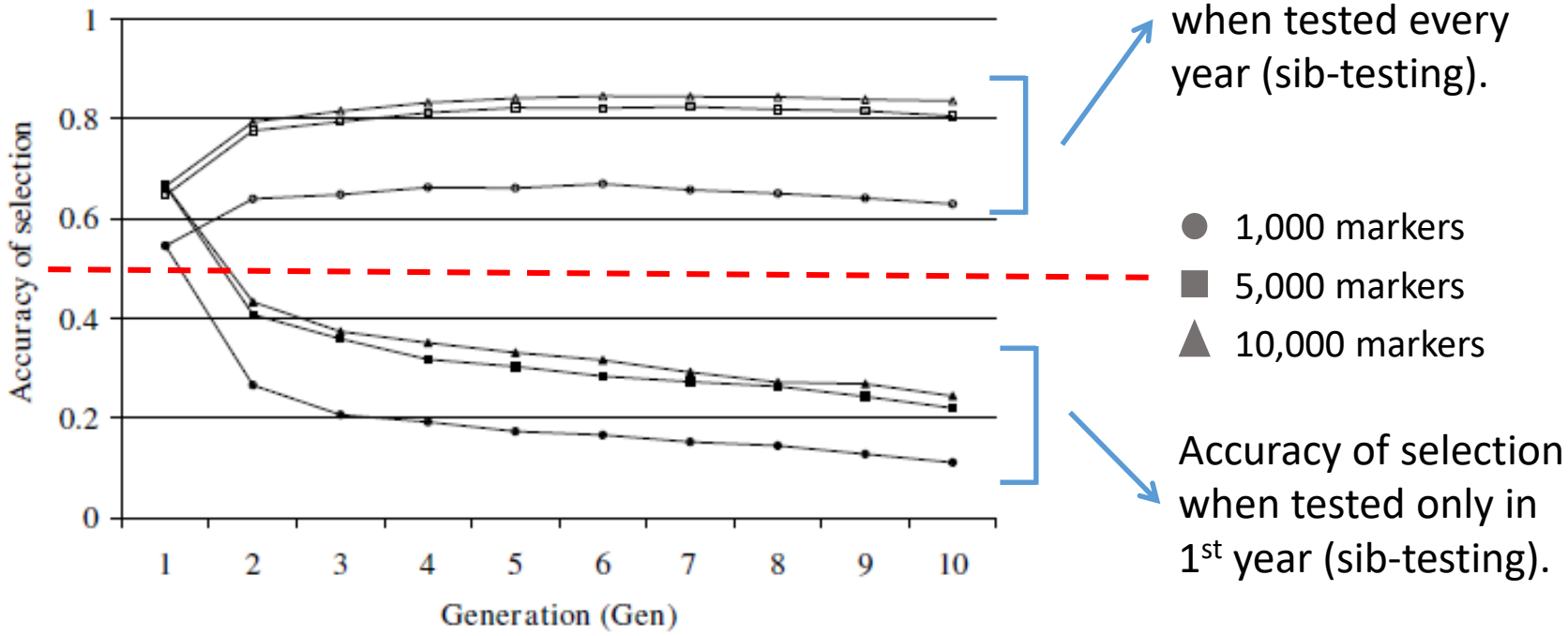
- GWS EBV accuracy is about 80% for cattle.
- GEBVs can be calculated in early life, which reduces time and cost.
- In dairy cattle genetic gains can be increased by two-fold.
- It requires a reference population, which could be a problem in livestock, but it is common practice for fish.
- The linkage map of Tilapia is about 1113 cM, so GWS at least 1200 markers.
- It has been successfully used in dairy cattle and sheep.
- One disadvantage is that one always has to genotype all the markers to calculate GEBVs.
- It requires a high-density linkage map.



Advantage of a genetic breeding program

- ***Large genetic potential in aquaculture animals***
- Evidence that growth rates in aquaculture species can ***continuously increase an average of 10% per generation*** using genetic breeding.

Testing strategies for genomic selection in aquaculture breeding programs



Sonesson and Meuwissen (2009). Genetics Selection Evolution. 41:37.

- Genome-wide selection has even greater potential for multiple &/or complex traits.

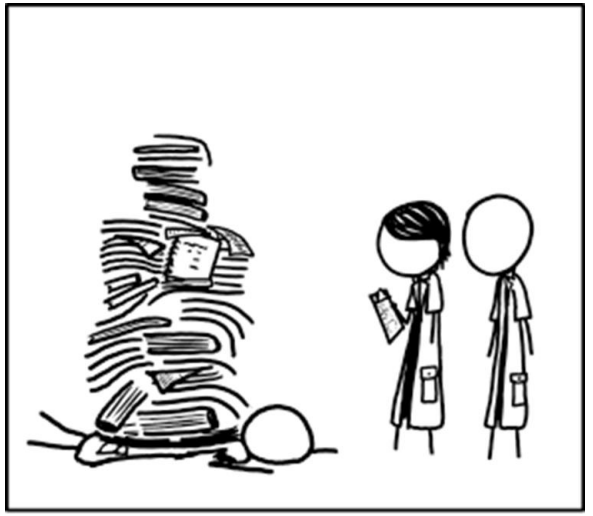
THE CENTER FOR AQUACULTURE TECHNOLOGIES CANADA

Genomic Approaches in Aquaculture

- **Challenge**:
  - Cost
  - Phenotypic gap
  - Analyzing the data can prove to be difficult
- **Benefits:**
  - Increased accuracy – leading to increased genetic gain
  - Accurate selection when traits can be measured in the selection candidates
    - e.g. Disease Resistance, Fillet Quality, Fatty Acid Deposition

Overcoming the Barriers

- **Cost**:
  - Exploring different methods of genotyping
  - Determining minimum required marker density for each application
  - Exploring different statistical approaches to genomic selection



- **Data:**
  - CATC is constantly evaluating and assembling pipelines for the analysis of big data
  - CATC has in-house capacity to analyze data for genomic selection using a variety of genotyping platforms, including sequencing, arrays and probe-based QPCR.

Pedigree reconstruction and traceability

- Raising many families in separate family tanks is very costly.
- Pit tags can be used, however, that can also be expensive and it delays family pooling.
- Parent assignment using SNPs is fast and high-throughput.
- It allows pooling at earlier life stages.
- CAT has developed parent assignment panels for several commercial species such as white-legged shrimp and Atlantic salmon.

- Traceability is becoming more important to aquaculture.
- It is relevant from environmental, regulatory and quality assurance standpoints.
- Population traceability can also be performed with SNPs and CAT has developed such technology for sturgeon and Atlantic salmon.

Characterizing the rapid rejection of salmon lice, *Lepeophtheirus salmonis*, by juvenile coho salmon, *Oncorhynchus kisutch*

Laura M. Braden[1], Tiago Hori[2], Jordan Poley[1], Phillip Byrne[3], Mark Fast[1]

[1]Department of Pathology & Microbiology, Atlantic Veterinary College, Charlottetown, PE, Canada

[2]Centre for Aquaculture Technologies Canada, Souris, PE, Canada

[3]Gulf Containment Unit, Department of Fisheries & Oceans Canada, Charlottetown, PE, Canada

UNIVERSITY of Prince Edward ISLAND

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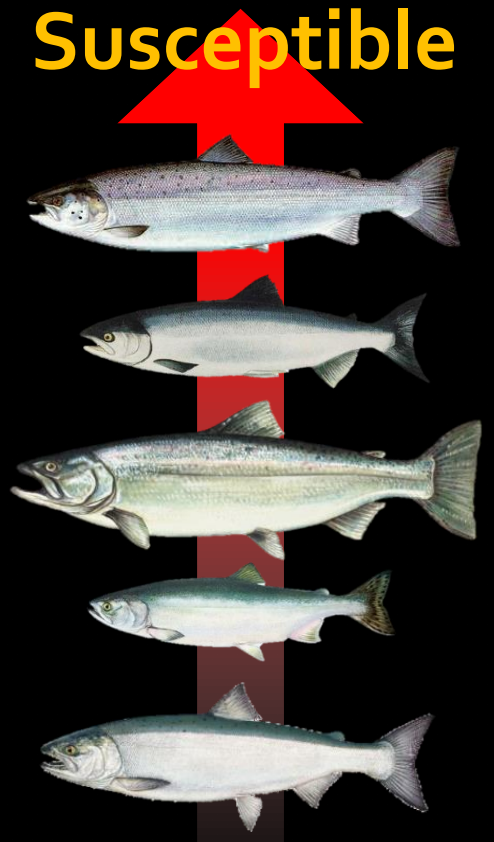
Fisheries and Oceans Canada

Variable host response to *L. salmonis*



Atlantic

Sockeye

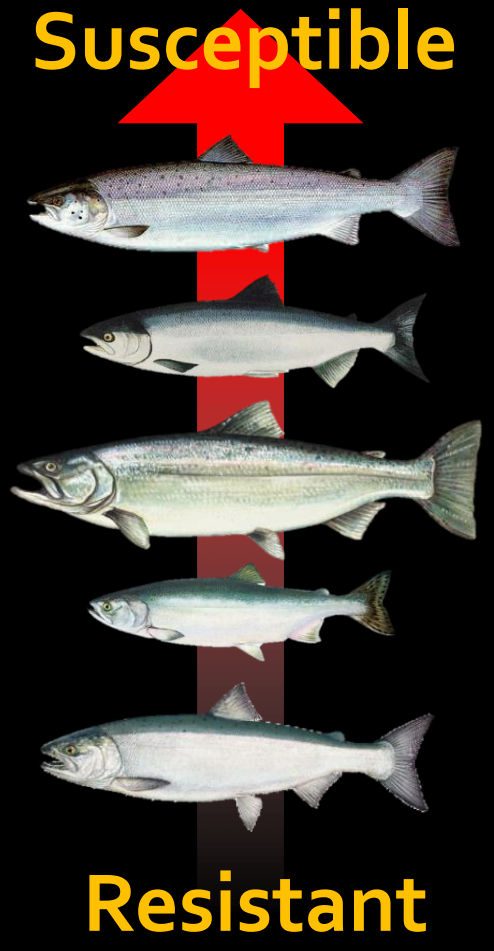
Chinook

Pink

Coho

- Delayed, weak inflammation
- Weak wound healing response
- Chronic infections

Variable host response to *L. salmonis*



Atlantic

Sockeye

Chinook

Pink

Coho

- Delayed, weak inflammation
- Weak wound healing response
- Chronic infections

- Immediate, aggressive inflammation
- Regulation of inflammation
- Rapid wound repair
- Parasite rejection

Host rejection of the salmon louse

Responses by **host** and **parasite** determine successful infection

- Species-specific variability in host responses
  - Fast et al. 2004; Jones et al. 2007; Sutherland et al. 2011, 2015; Braden et al 2012, 2015
- Parasite response determined by host species
  - Braden et al. 2016, in review *BMC Genomics*

Rapid rejection by juvenile coho salmon

- Attached parasite is engulfed by hyperplastic epithelia, aggressive cellular infiltrate
  - Johnson & Albright 1992; Fast et al 2002

Johnson & Albright, 1992

Host rejection of the salmon louse

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- Parasite response determined by host species

Mechanism/cellular effectors involved in parasite rejection are unknown

Rapid rejection by juvenile coho salmon

- Attached parasite is engulfed by hyperplastic epithelia, aggressive cellular infiltrate
  - Johnson & Albright 1992; Fast et al 2002

Johnson & Albright, 1992

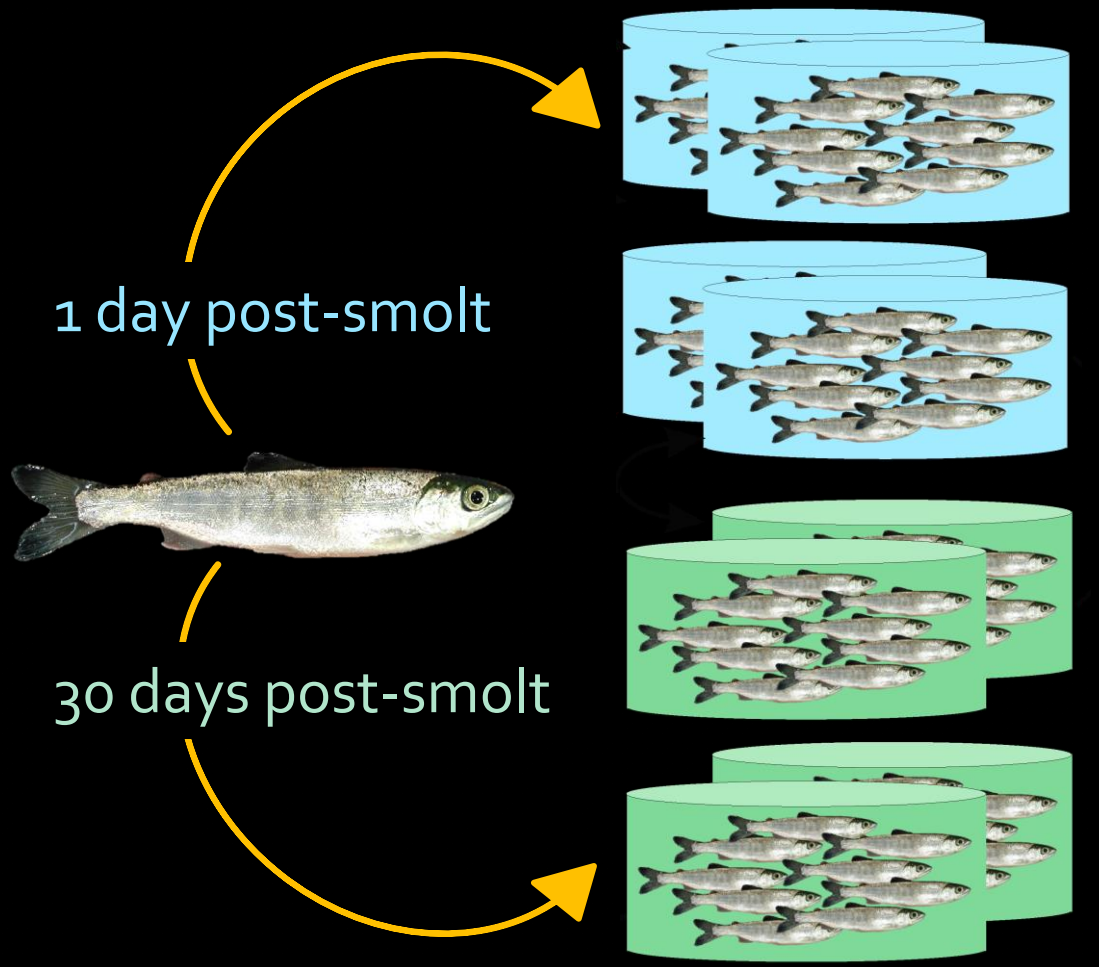
Rationale:

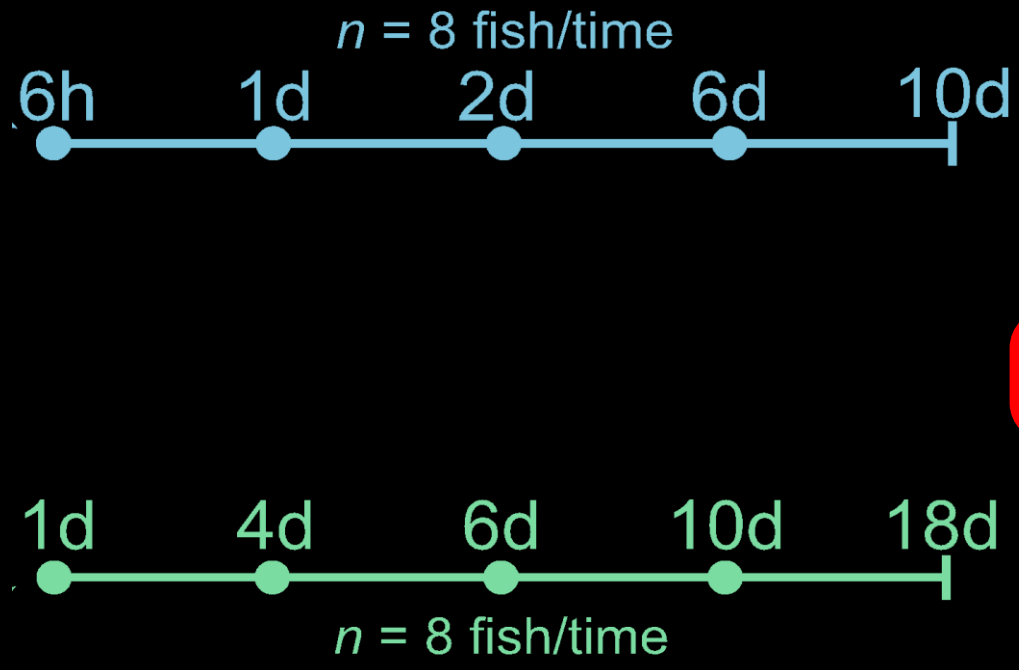
The period of rapid rejection of the salmon louse by *O. kisutch* has not been fully characterized

Objectives:

1. Use a dual RNA seq approach to assess the transcriptomic response of host and parasite during the period of rejection (~ first 2 weeks)
2. Determine if resistance is a function of life-history (e.g., if resistance accompanies switch to saltwater, if resistance increases with host size)

Experimental design



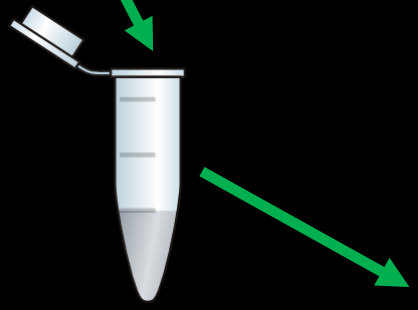


Samples:

- Gill
- Skin
- Fin
- Head kidney
- Spleen

Sequencing the host-parasite transcriptome
~ A novel dual RNAseq approach ~

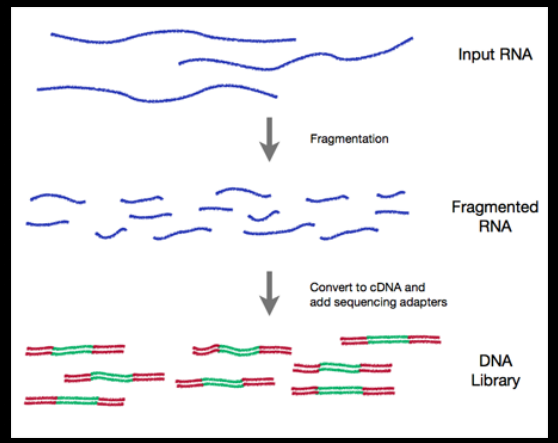
Infected fins/
Un-infected fins



Parasite & host RNA

Libraries produced from infected fins had 2-8% reads mapped to the lice genome.

RNA was used to build Illumina stranded mRNA libraries



RNA sequencing

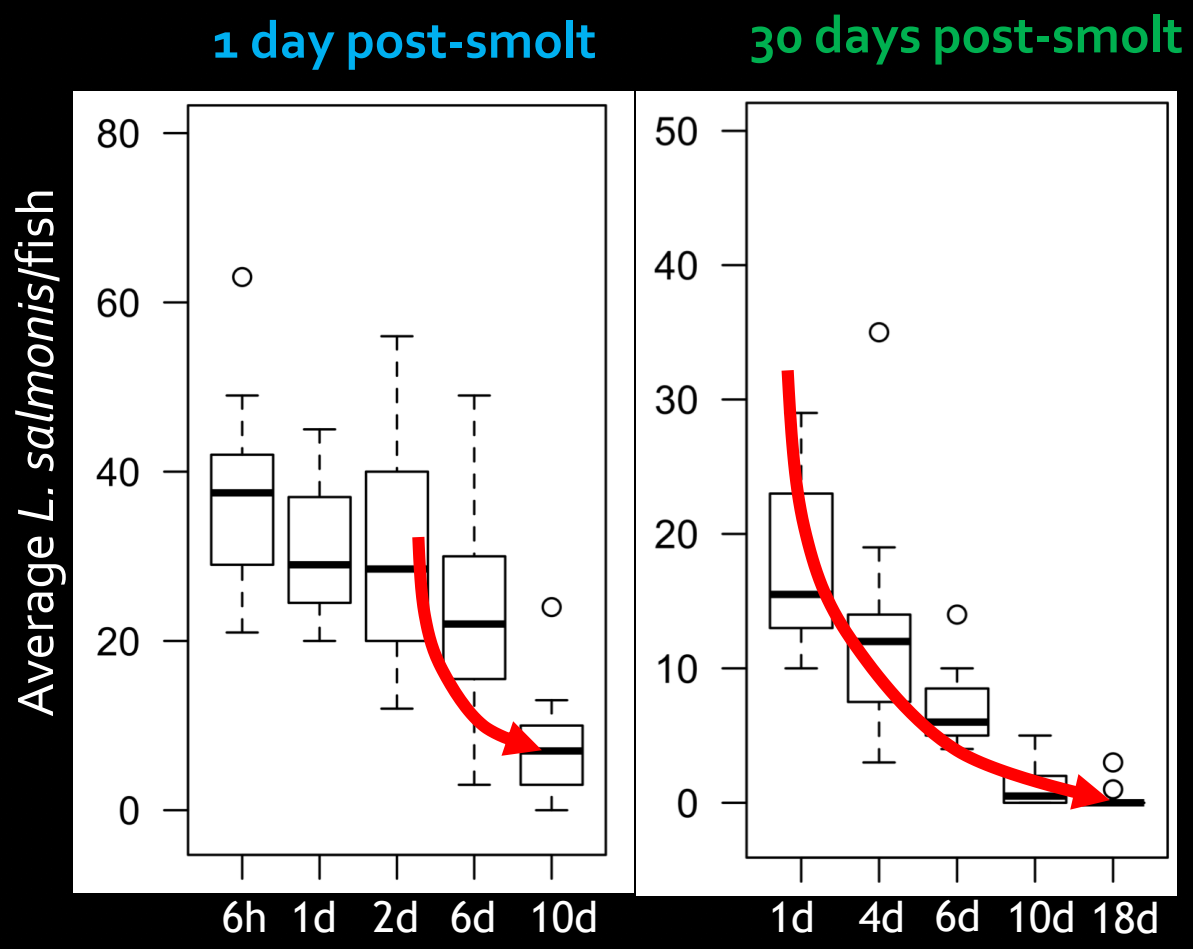
Samples were sequenced in 3 lanes of an Illumina HiSeq2500 flow-cell.

Bioinformatics

Reads were trimmed and then mapped to lice genome or Coho transcriptome

- Clustering analysis
- DEG analysis
- Functional annotation
- Pathway analysis

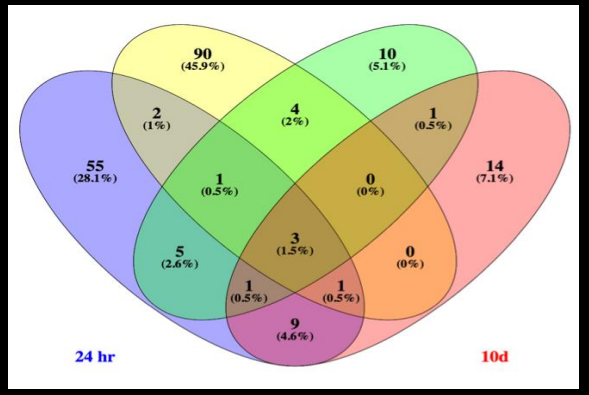
Lice are rapidly rejected by Coho

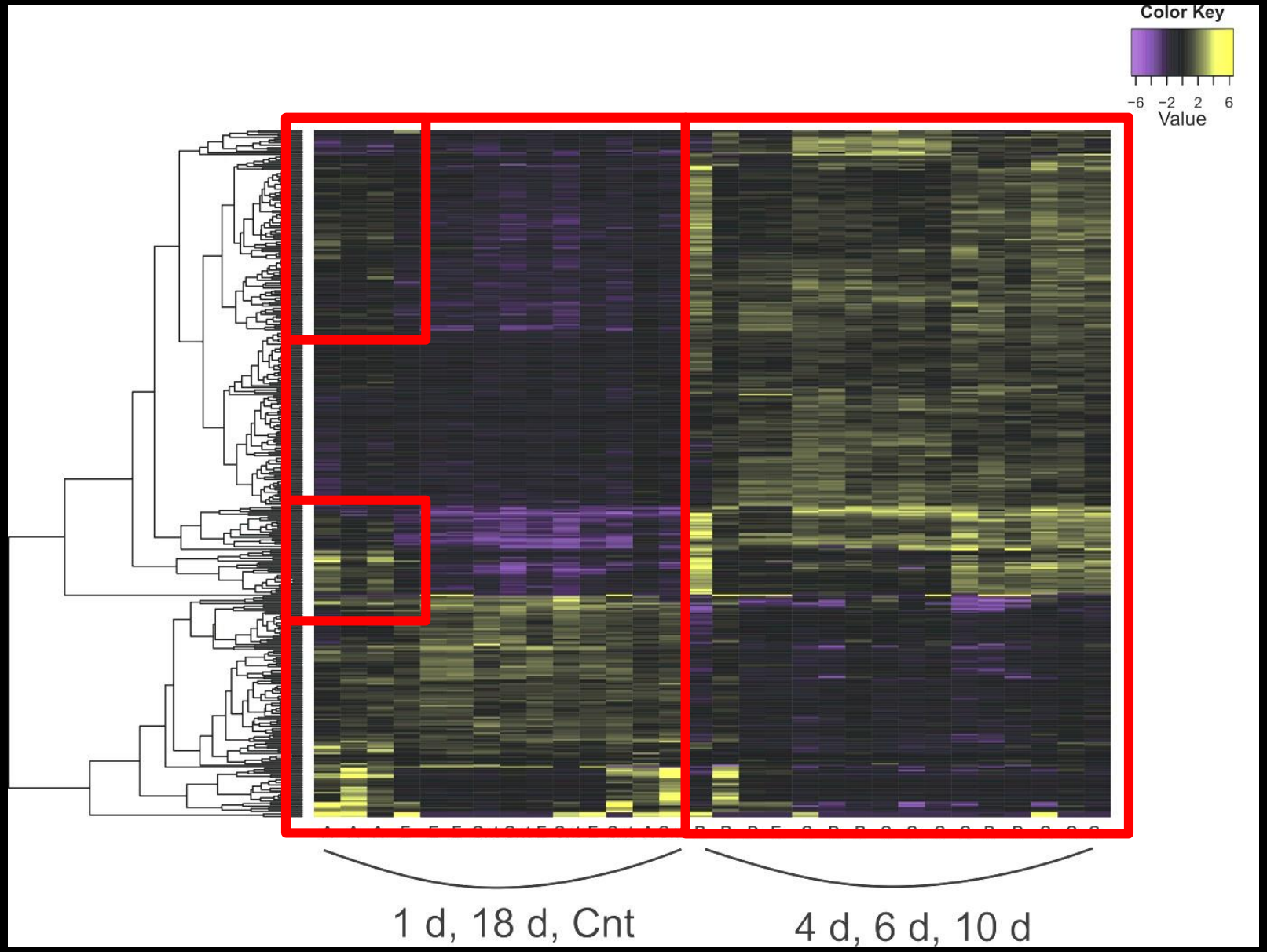


Dual RNA-seq of coho salmon fin infected with *L. salmonis* (30 day post-smolt)

Host
- Over 2000 differentially expressed & unique transcripts
- Distinct clustering between controls and infected fish

Parasite
- Over 800 differentially expressed transcripts
- No lice transcripts detected in controls or at 18 days

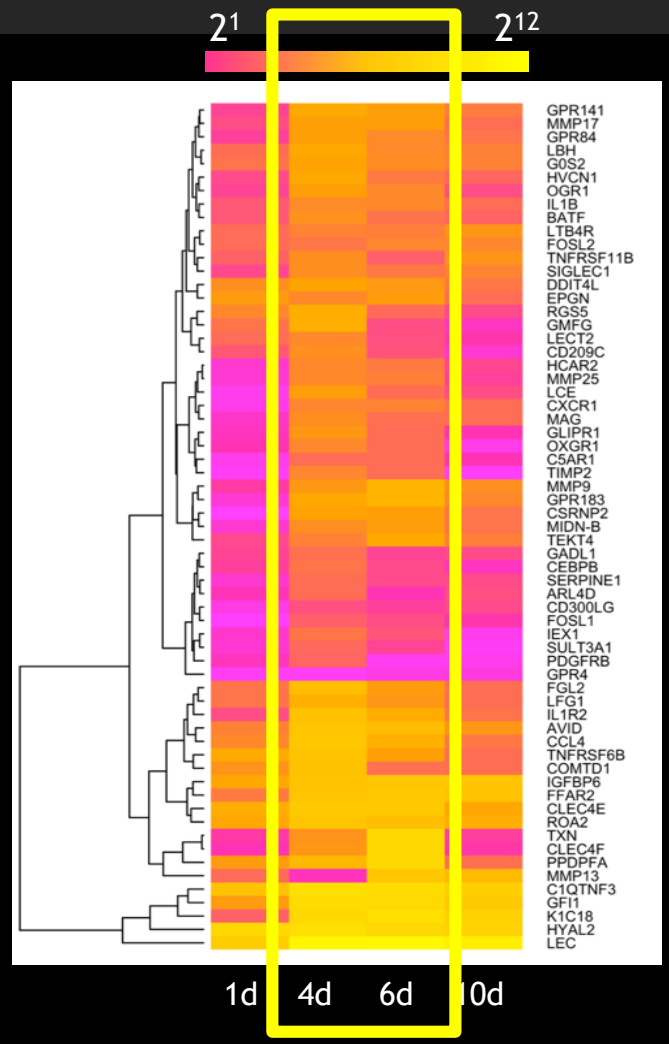




Attack response of coho salmon

Checks & balances

Zinc finger protein Gfi-1
TNF receptor superfamily member 11B
Protein lifeguard 1
Plasminogen activator inhibitor 1
Metalloproteinase inhibitor-2
Free fatty acid receptor 2
DNA damage inducible transcript 4-like protein
Basic leucine zipper transcriptional factor ATF-like



Simultaneous host-parasite responses
~ Temporal-specific response of the host ~

1 day infected
Ribosomal proteins, transcriptional activity
Energy metabolism
Inflammatory mediators

4 days infected
Cellular effectors
Growth factors
Adhesion factors

6 days infected
Acute phase response
Growth factors
Cytoskeletal components
ECM components
Effector molecules

10 days infected
Regulatory factors

18 days infected

Louse response during coho attack

1. Cuticle/chitin proteins

Cuticle protein 7
Cuticle protein 6
Cuticle protein 18.6, isoform B
Cuticle protein CP14.6 precursor
Peritrophin 1-A
Peritrophin 1
Chitin binding peritrophin-A, putative
Putative cuticle protein
Neuronal acetylcholine receptor subunit al

2. Stress proteins

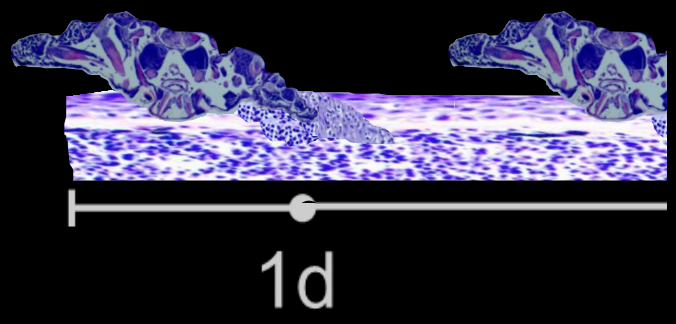
Heat shock protein beta-1
Stress protein ddr48
Nesprin-1
Catalase
15-hydroxyprostaglandin dehydrogenase
Thioredoxin domain-containing protein 17
Extracellular superoxide dismutase precursor

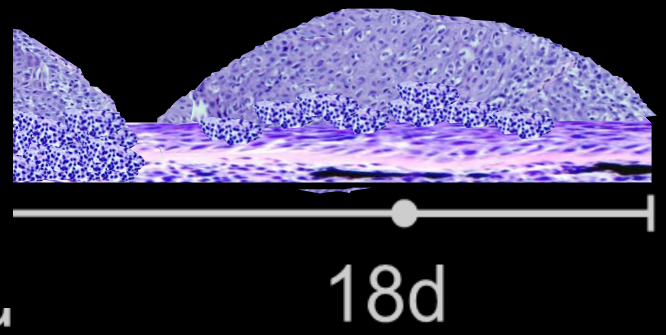
3. Virulence factors

Cathepsin L1
Matrix metalloproteinase 9
Legumain
Trypsin-1
Collagenase
Chemosensory protein 16 precursor
Chemosensory protein 3
Carboxypeptidase A2 precursor
Intestinal trypsin 2 precursor

4. Cytoskeleton proteins

Myosin heavy chain CG17927-PF isoform 7 (predicted)
Myosin-2 essential light chain
Myosin-light chain
Tropomyosin
Actin
PREDICTED: similar to Actin-5C isoform 2
Transmembrane protein, putative

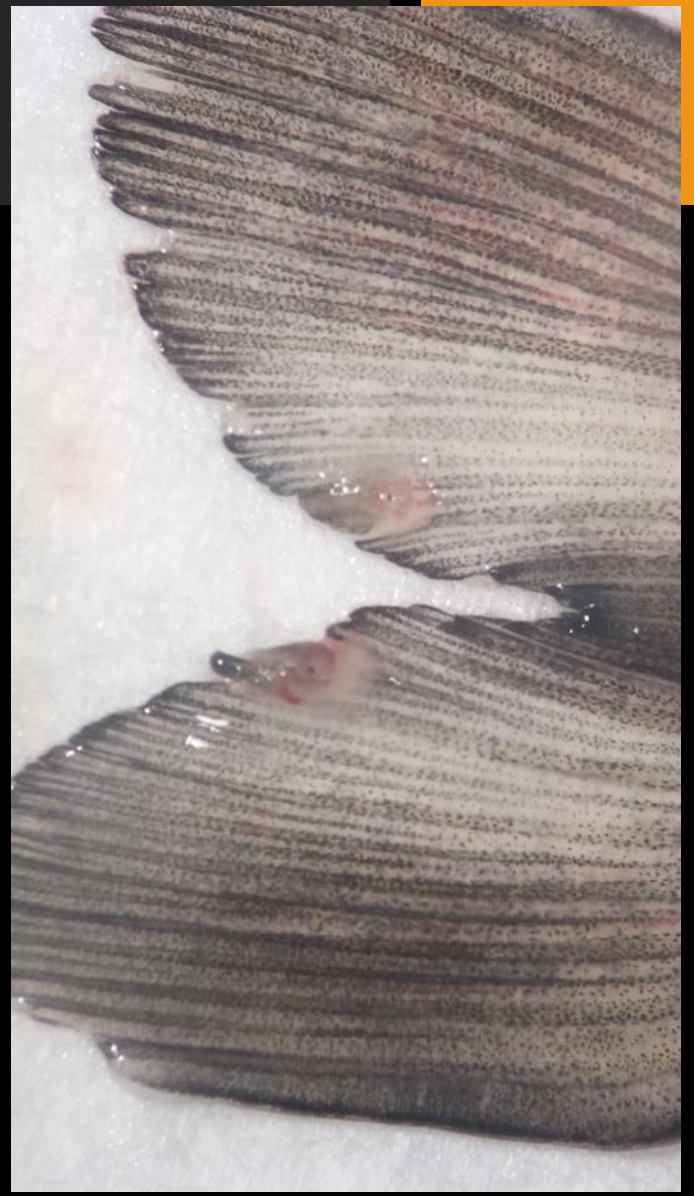




Preliminary analysis:

Dual RNA seq approach provides a 'true' snapshot of the host-parasite interaction

- Complement membrane attack involved in louse killing
  - Inflammatory cascades, tissue remodeling, cellular infiltration and growth
- Detected host novel effector molecules
  - *Fibroleukin, IgE receptor*
- Louse response to host attack indicates overall stress, oxidative-damage response
  - *Hspb1, stress protein*
  - *Catalase, glutathione peroxidase*
  - Development-associated genes
    - Cuticle proteins, chitin-binding proteins, proteinases



Summary & going forward

- The **first dual transcriptome analysis** of the salmon louse and resistant coho salmon
  - Important insights in mechanisms involved in rejection
  - Identifies regulatory responses driving **pathogenesis** & **resistance**

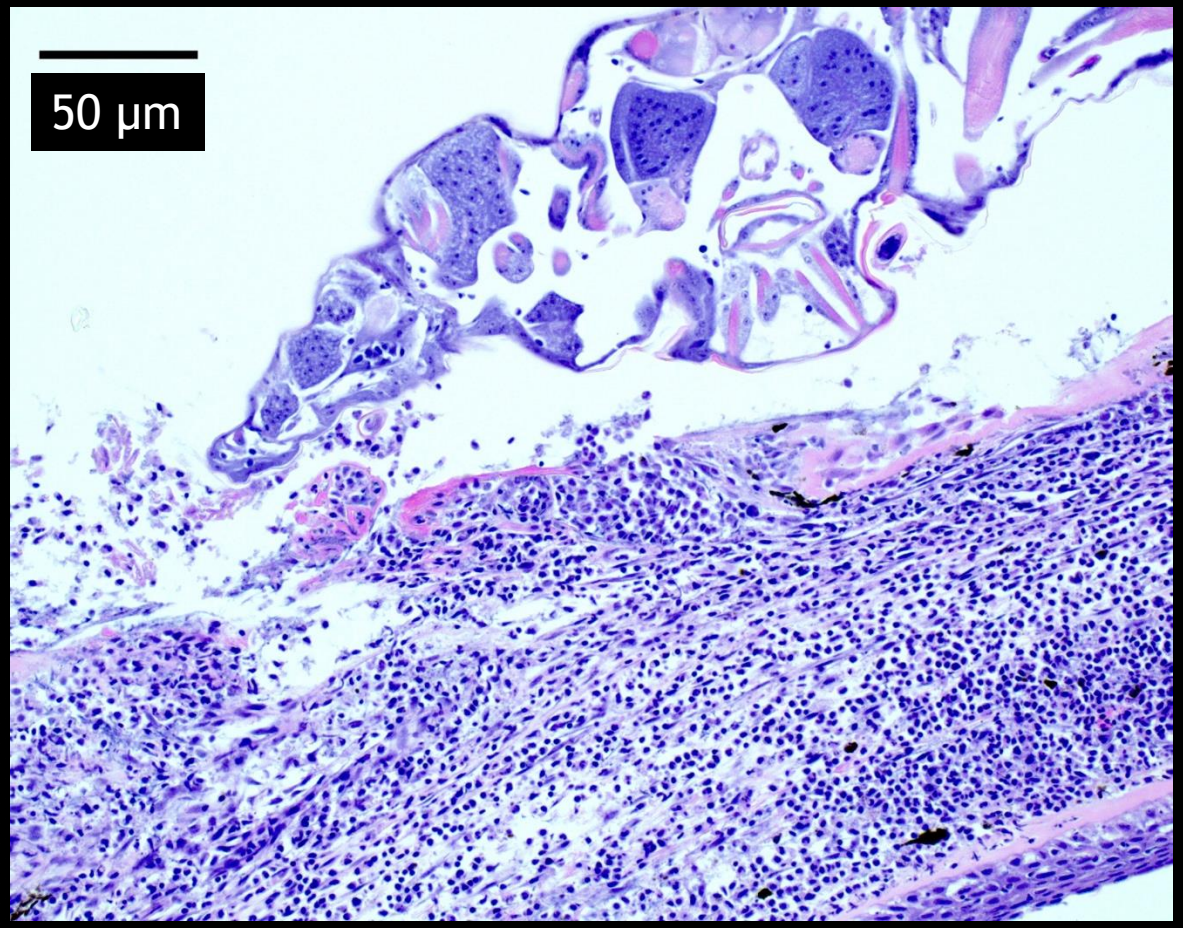
Summary & going forward

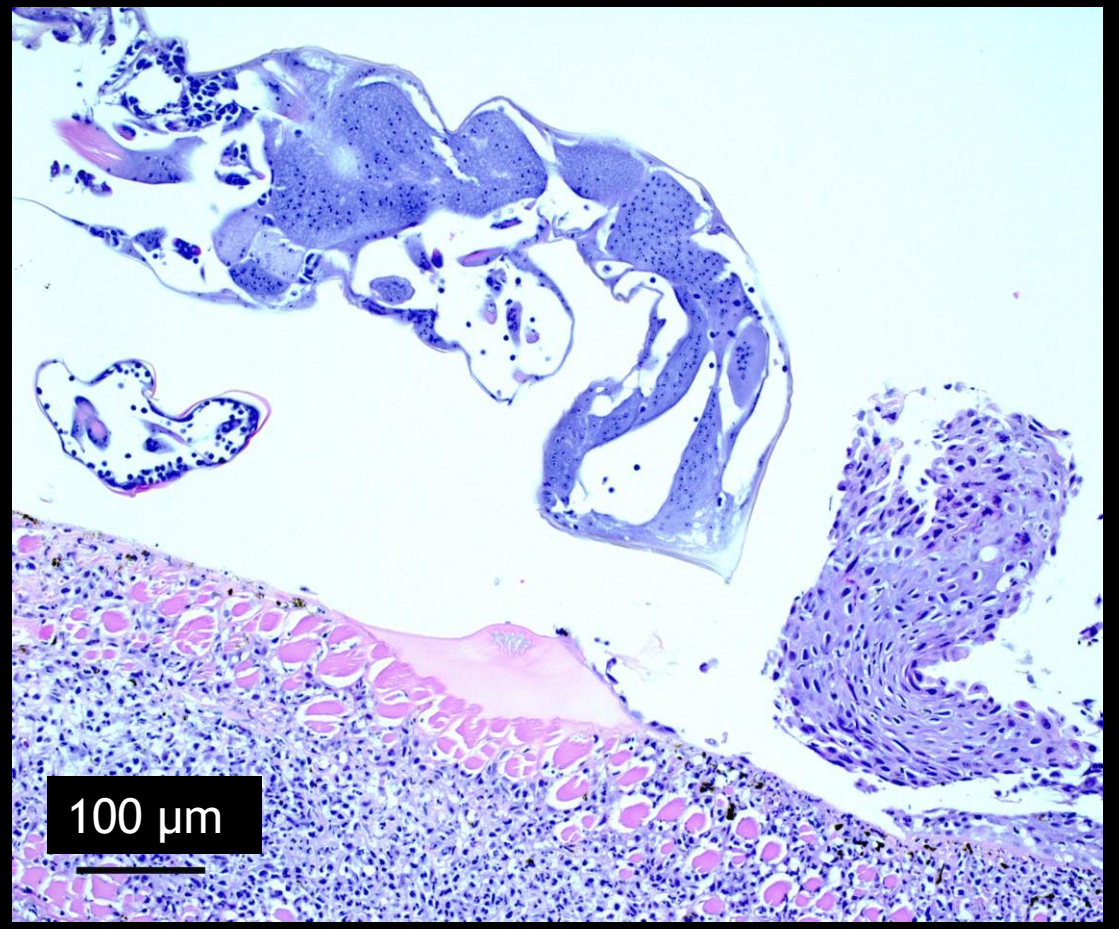
- The **first dual transcriptome** analysis of the salmon louse and resistant coho salmon
  - Important insights in mechanisms involved in rejection
  - Identifies regulatory responses

- **Next steps**
  - **Explore expression** of identified targets in 1 day post-smolt & larger cohort (~ 60 gram)
  - **Immunohistochemistry** of encapsulation sites to identify cell populations

Profound cellular infiltrate at attachment site

~ Characterization of cellular effectors ~





Acknowledgements

- Phyllis Dixon, Greg MacCallum with DFO-Gulf
- Brian Banks and staff at Nanaimo River Hatchery
- Steve Cho with DFO-Pacific
- Sampling team
  - Carter Van Iderstine, Tyson Hay, Dylan Michaud, Alyson Brown, Jessica Fry, Dr Mark Braceland
- Dr Sara Purcell

NSERC CRSNG

Elanco

Fisheries and Oceans Canada

Pêches et Océans Canada

Canada


Questions?
@Lulu_Braden
lbraden@upei.ca

FeedKind® Protein: The future of Aquaculture Feeds

Atlantic Canada Fish Farmers Association
Huntsman Fundy Discovery Centre
St. Andrews, NB
October 26, 2016

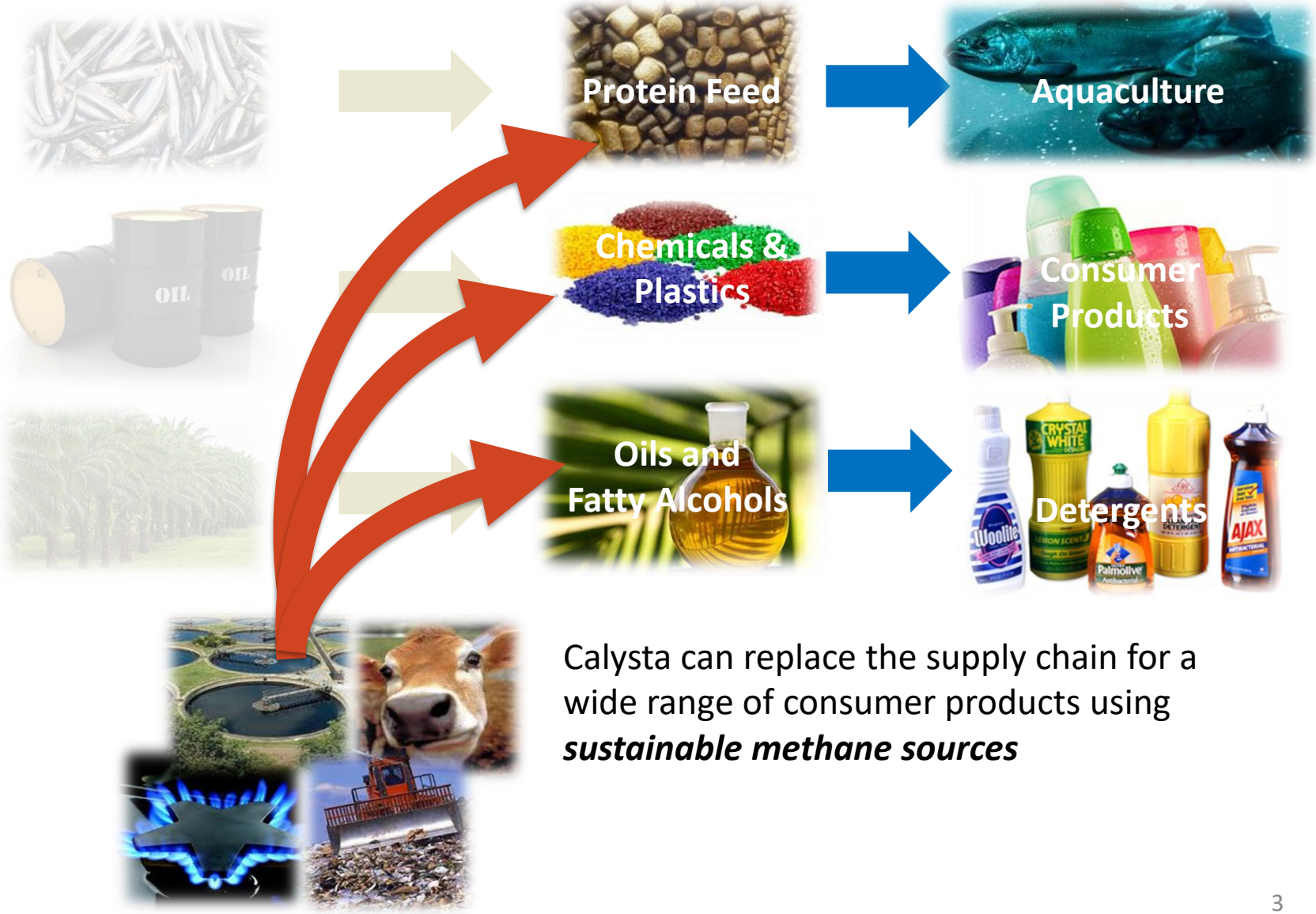
CALYSTA

CALYSTA

Calysta: What we do

Making everything from plastics to protein

CALYSTA



Calysta can replace the supply chain for a wide range of consumer products using ***sustainable methane sources***

CALYSTA

FeedKind® Protein

"Future Fit Feed"

Two Problems…One Solution

CALYSTA

Food Security

Global Warming

CALYSTA

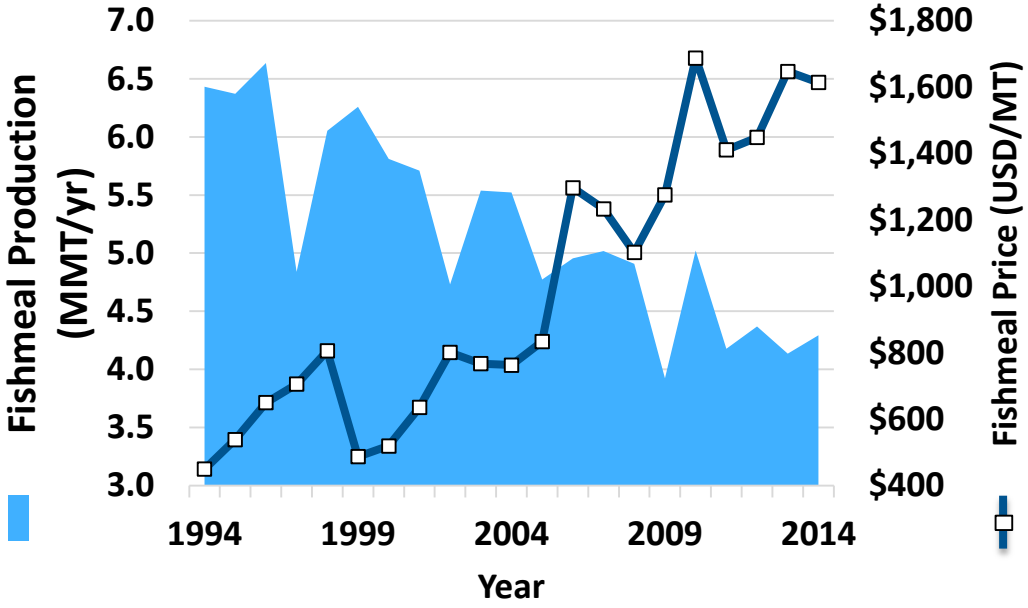
"Future Fit Feed"

- No agricultural land use
- 77-98% less water than agricultural products
- 40% improved CO_2 emissions compared to combustion of methane
- FeedKind protein does not compete with the human food chain
- No animal derived ingredients
- Helps mitigate global warming losses at 5% of global GDP (IPCC 2014)

Fishmeal is a Non-Ideal Ingredient

CALYSTA

- Demand for fishmeal continues to grow while supply is constrained by flat or shrinking wild fish populations
- Supply is highly variable and dependent on Pacific weather patterns
- Feed is the single biggest cost in the of production of aquaculture, comprising 40-50% of total production costs for salmon



"Our biggest challenge is how to meet this demand… where to find new raw materials"

Andrew Jackson, Technical Director, International Fishmeal & Fish Oil Org.

Source: World Bank Global Economic Monitor, 2014.
The 2013 Marine Harvest Salmon Industry Handbook
Undercurrent News, Oct 2013.

FeedKind® is a Natural, Non-GMO Protein Source Offering Significant Differentiating Benefits

CALYSTA

Naturally occurring microorganisms metabolize methane as their sole source of carbon and energy, producing a nutritious, high-protein biomass

Multiple Monetization Opportunities:

Supply Chain	Consumer	Sustainability	Under development
• Traceable • Consistent product • Year round production • Long shelf life • Reduced enteritis from plant proteins	• Non-GMO • Natural fermentation process • Reduced fish-in / fish-out ratio • Saturated fatty acids • No animal based ingredients	• No agricultural land use • Little water use • Additive to the human food chain	• Amino acid modifications • Omega-3 • Prebiotic and probiotic effects

EU Approval Already Received for Use in Fish and Livestock:

EU Feed Registration:

12.1.2	**Protein from *Methylococcus capsulatus (Bath)*, *Alcaligenes acidovorans*, *Bacillus brevis* and *Bacillus firmus***	Protein product of fermentation with *Methylococcus capsulatus (Bath)* (NCIMB strain 11132), *Alcaligenes acidovorans* (NCIMB strain 12387), *Bacillus brevis* (NCIMB strain 13288) and *Bacillus firmus* (NCIMB strain 13280) (1) on natural gas (approx. 91 % methane, 5 % ethane, 2 % propane, 0,5 % isobutane, 0,5 % n-butane), ammonia, and mineral salts, the crude protein is at least 65 %.	Crude protein Crude ash Crude fat

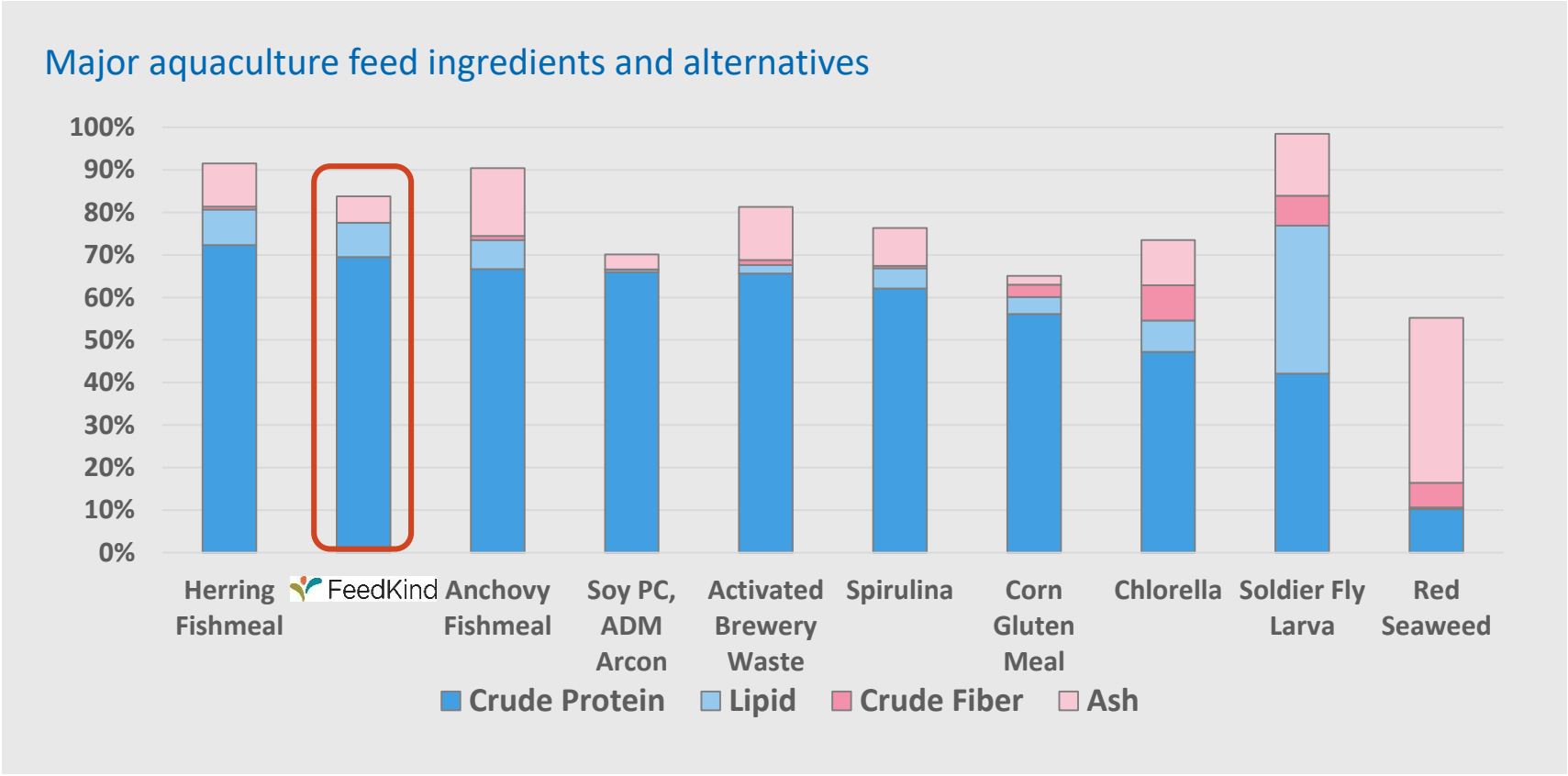
Millions of people eat single cell proteins every day

CALYSTA

Superior nutritional profile when compared to other fishmeal alternatives

CALYSTA

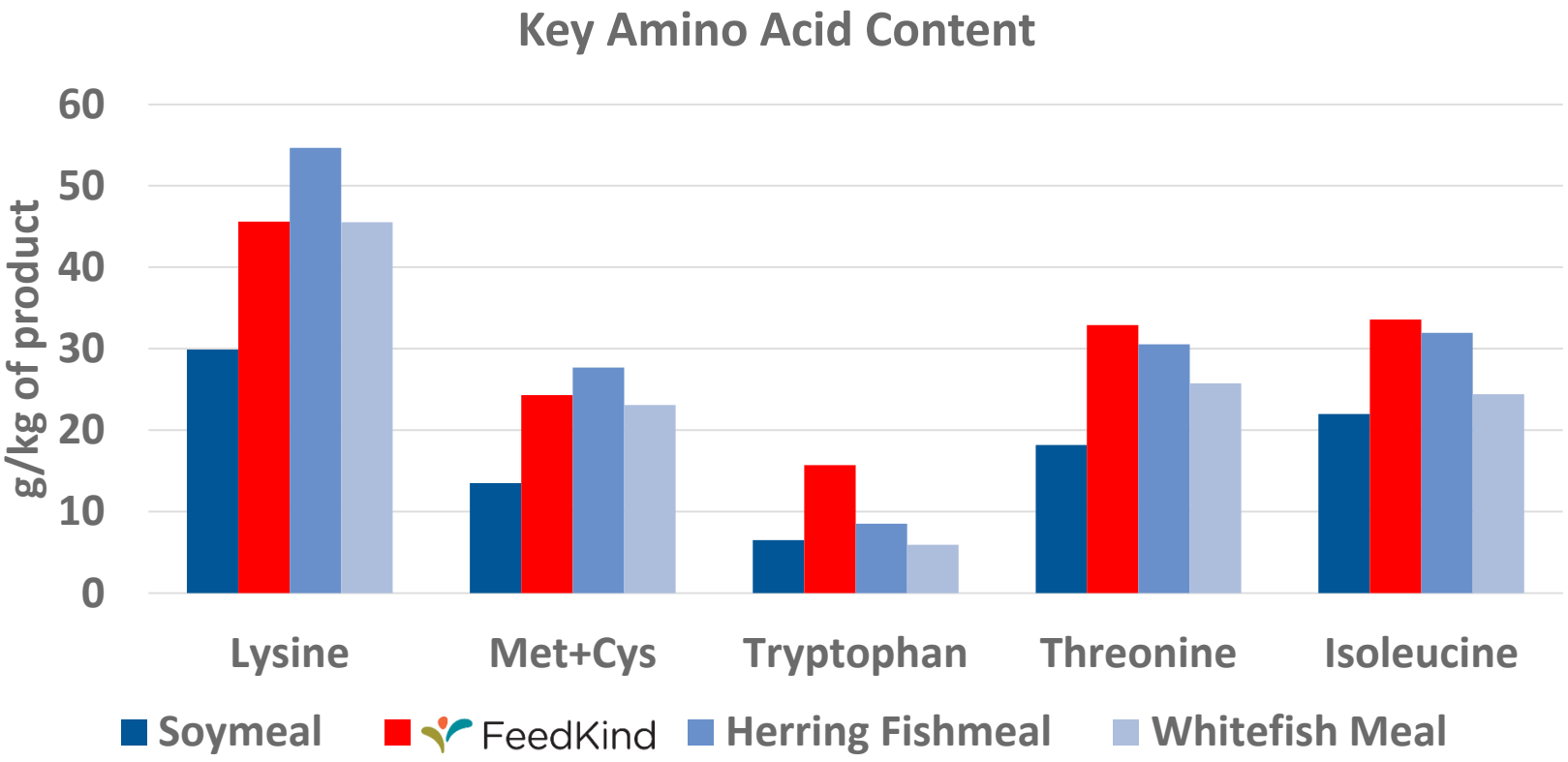
Many alternatives are plagued by high fiber or inferior amino acid profiles. Others may never scale to meaningful volumes.



Source: FAO Feed ingredients and fertilizers for farmed aquatic animals, 2009.

FeedKind® protein has a premium amino acid profile

CALYSTA



FeedKind® protein provides key nutrients

CALYSTA

High-value mineral content

High vitamin content

Nutrient	Mean	Range	Nutrient	Mean	Range
Minerals (in DM)#			Fatty acids [% of total fatty acids] ($n = 4$)*		
Phosphorus [g/kg]	14.8	13.7–15.9	C12:0	0.1	0.0–0.1
Calcium [g/kg]	2.8		C13:0	0.1	0.1–0.1
Magnesium [g/kg]	2.9		C14:0	4.2	3.9–4.4
Potassium [g/kg]	6.4		C14:1n-5	0.5	0.1–1.0
Sodium [g/kg]	2.9		C15:0	0.7	0.6–0.9
Iron [mg/kg]	317	293–341	C16:0	49.2	48.1–51.1
Manganese [mg/kg]	2.7	2.4–3.0	C16:1	36.0	32.4–39.5
Zinc [mg/kg]	22.4	22.1–22.7	C17:0	0.5	0.2–1.1
Copper [mg/kg]	83.9	79.9–87.9	C18:0	0.3	0.3–0.4
			C18:1n-9	0.2	0.1–0.5
Vitamins (in DM)*			C18:1n-7	0.2	0.2–0.3
Vitamin A [IU/g]	<1†		C18:2n-6	0.1	0.0–0.2
Vitamin E [mg/kg]	<5†		C18:3n-3	0.3	0.1–0.5
Thiamine [mg/kg]	12.1		C20:1n-11,n-9	0.1	0.0–0.2
Riboflavin [mg/kg]	73		Unidentified	7.5	4.0–12.1
Niacin [mg/kg]	130				
Inositol [mg/kg]	30				

Highly saturated, short-chain lipids

Notes: *Unpublished data; #Data from Aas et al. (2006a, 2006b); †Below detection limit.

Source: Overland et al., 2010. Review in Archives Anim Nutr.

FeedKind® protein improves growth in salmon

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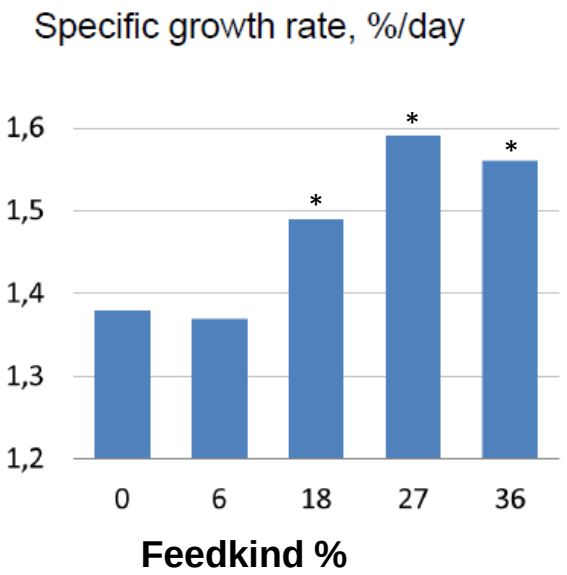
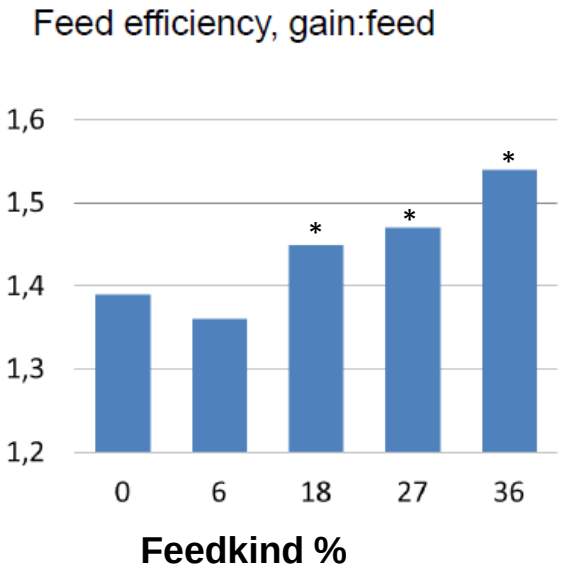
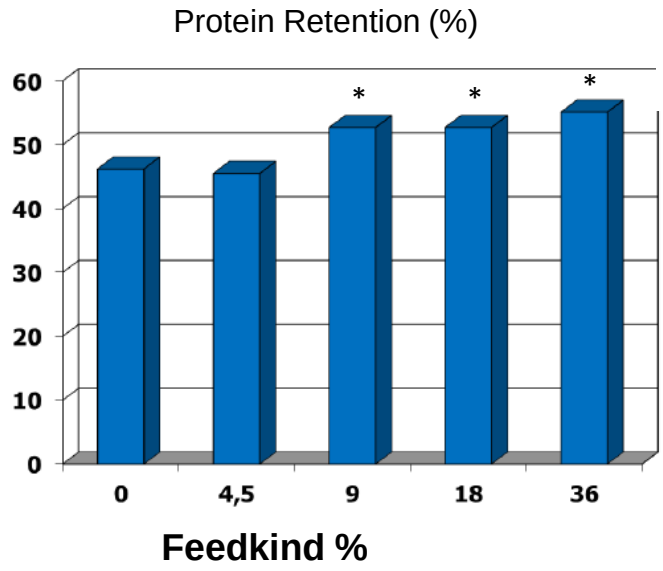






$p < 0.05$ vs 0% control

Improved growth, feed efficiency, and protein retention vs. fish meal in salmon

Source: Aas et al., 2006. Aquaculture

Consumers and Retailers Demand Traceability Throughout Their Supply Chain

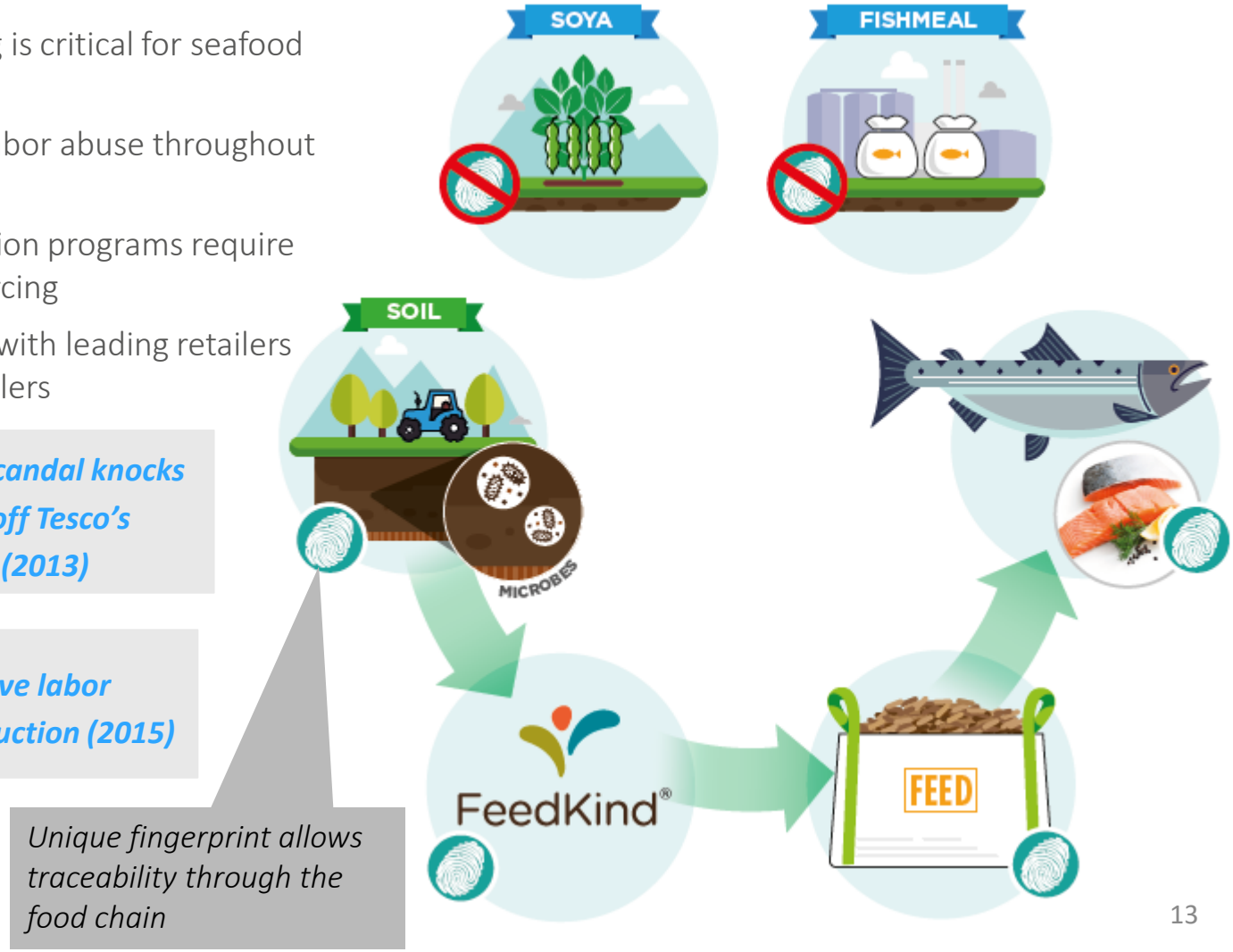
CALYSTA

Calysta's proprietary "marker" technology provides confidence on the integrity of the supply chain, a feature which legacy feed ingredients cannot offer

- Supply chain auditing is critical for seafood producers
- Must guarantee no labor abuse throughout the supply chain
- Sustainable certification programs require sustainable feed sourcing
- Ongoing discussions with leading retailers and seafood wholesalers

Horse meat scandal knocks £300 million off Tesco's market value (2013)

Costco sued over slave labor used in shrimp production (2015)



Unique fingerprint allows traceability through the food chain

Commercialization Under Way: European Fermentation Center Opens in the UK

CALYSTA

- Opening ceremony on September 19th, 2016
- Samples available for customers and regulatory agencies Q2 2017
- European location makes facility accessible to customer

European Fermentation Center in Teesside, England.

Ground-breaking on 1st Commercial Plant in North America Anticipated Q4 2016

CALYSTA

- Collaborating with Cargill on a 200,000 tonne per year production facility in North America
- Global marketing collaboration allows leveraging of Cargill global distribution channels
- Modular design lends itself to phased construction process
- 20,000 mtpa Phase I coming online Q4 2018

Cargill™

200,000

tonnes per year to be produced at commercial scale

over $300M

expected annual revenue

Opportunities

CALYSTA

- Aquaculture producers to test Feedkind protein in different species
- Offtake partners for early phases of commercial production

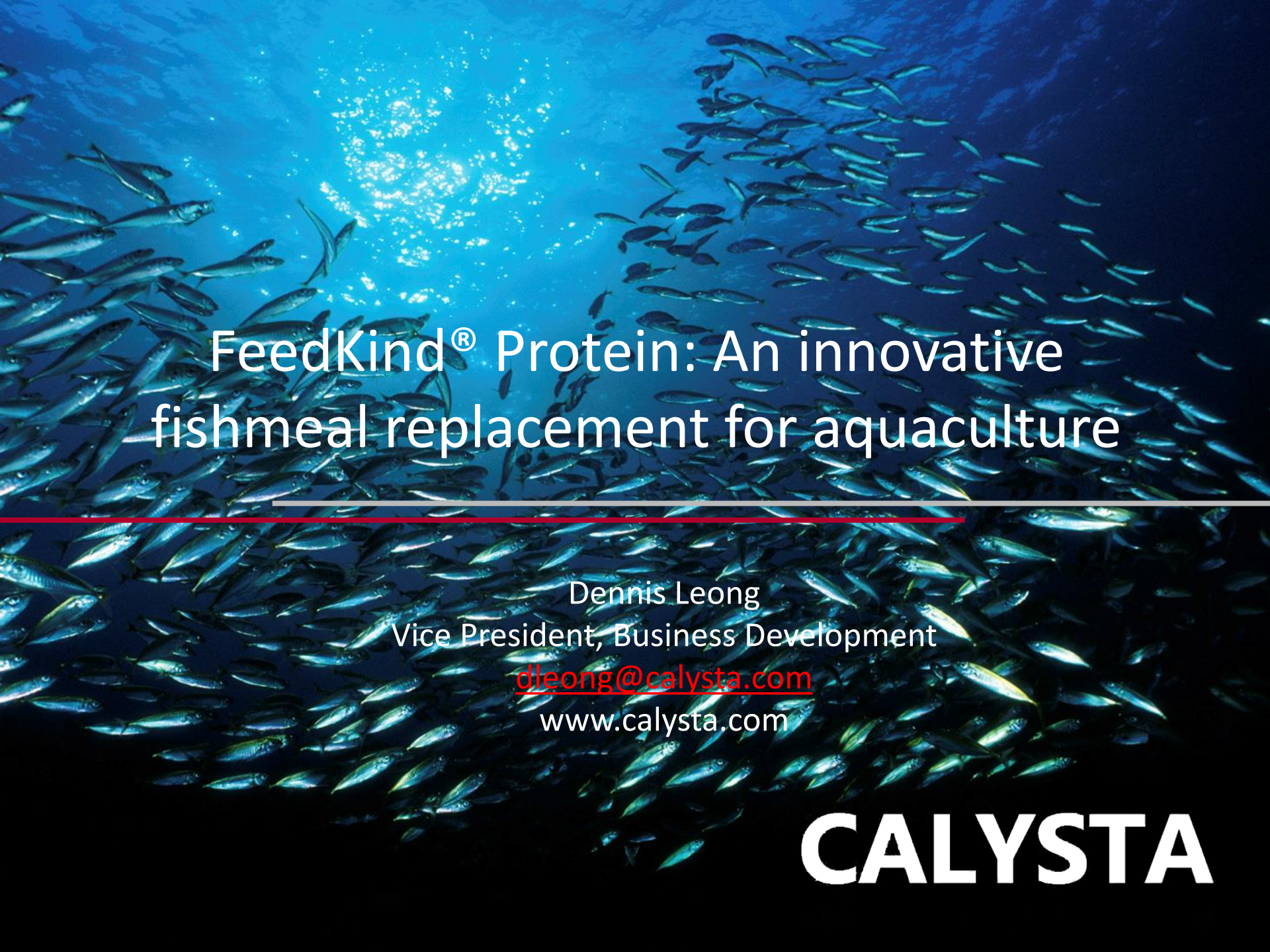
FeedKind® Protein: Future Fit Feed

CALYSTA

- Proven ✓
- Scalable ✓
- Backed by Cargill ✓



A sustainable source of protein that improves the health and quality of farmed fish and livestock



FeedKind® Protein: An innovative fishmeal replacement for aquaculture

Dennis Leong
Vice President, Business Development
dleong@calysta.com
www.calysta.com

CALYSTA

Development of an alternative sulfide detection method

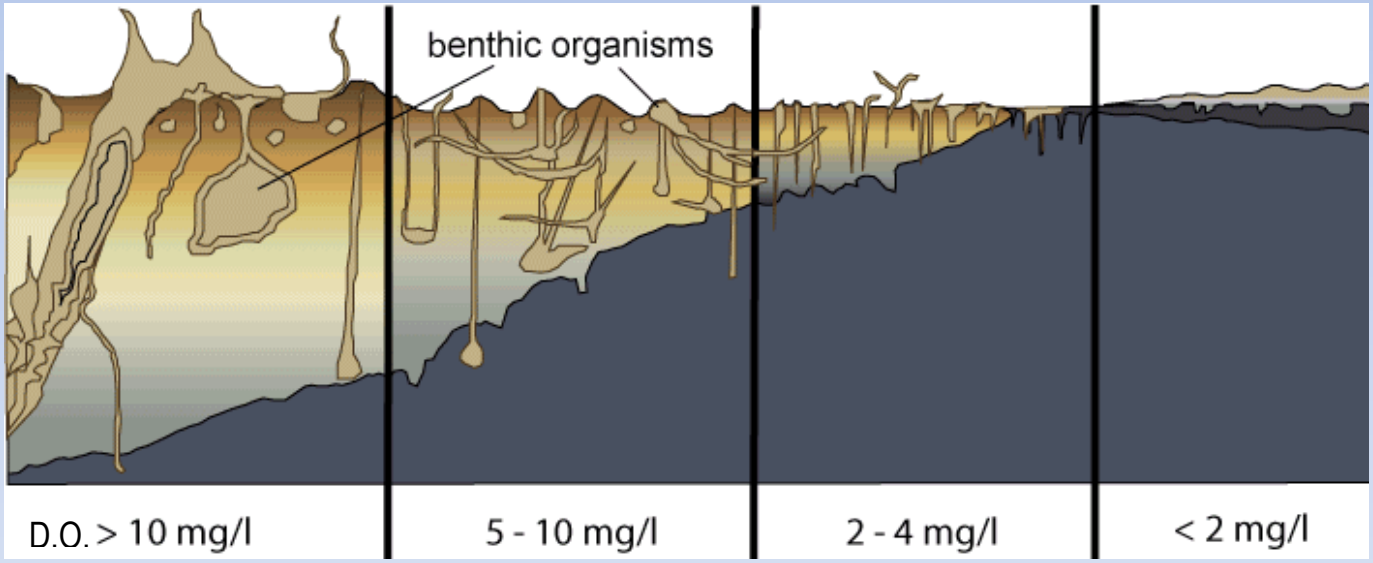
David Wong

Fisheries and Oceans Canada

St. Andrews Biological Station, NB

Primary causes of sulfide around cage sites:

- Excess uneaten feed
- Faeces

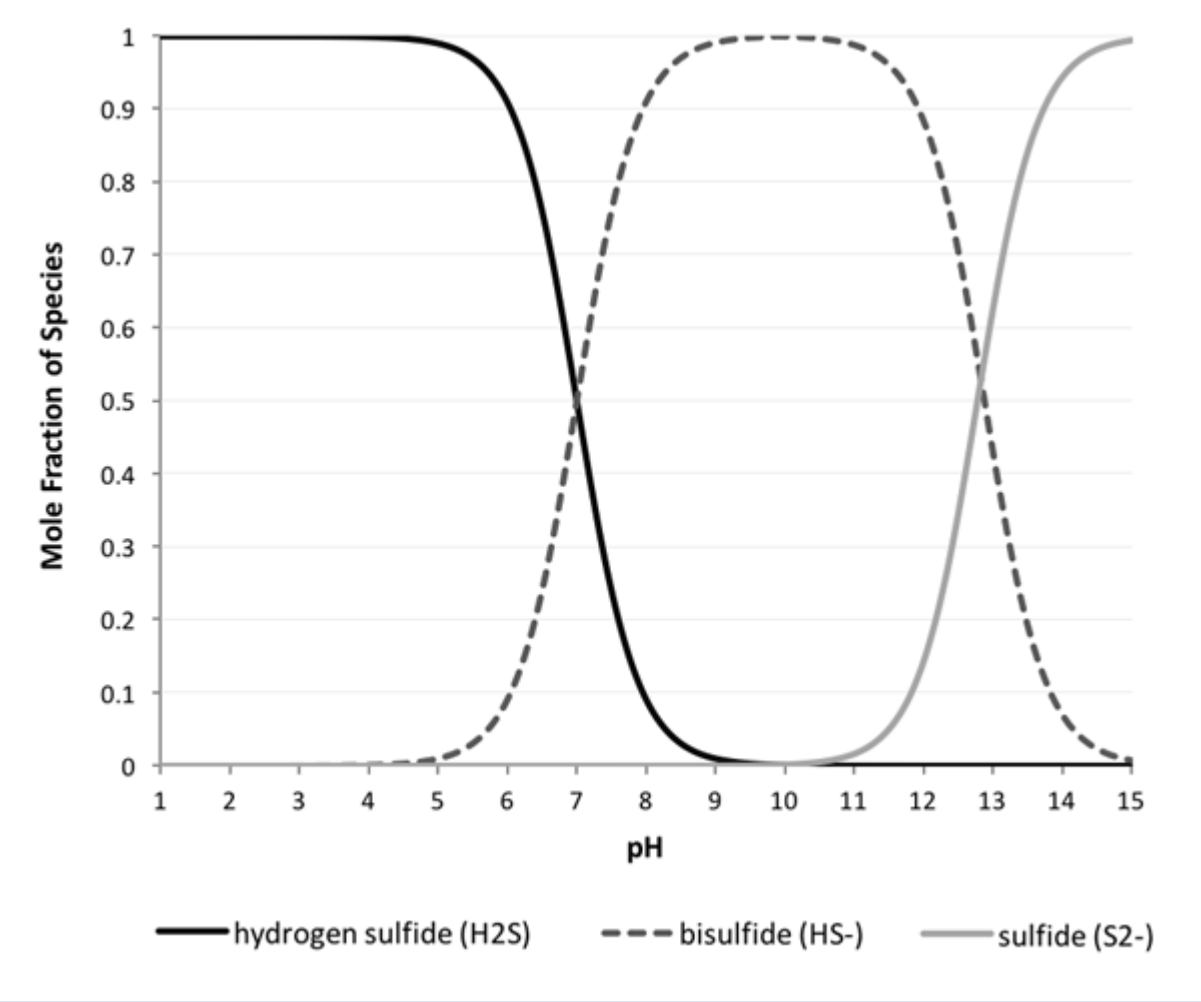


Adapted from Nilsson and Rosenberg 2000 (Marine Ecology Progress Series 197)

Site classifications related to sediment sulfide concentrations (NB DELG 2012)

Site classification		Sediment sulfide concentration (µM)
Oxic	Oxic A	≤ 750
	Oxic B	750 - 1,499
Hypoxic	Hypoxic A	1,500 - 2,999
	Hypoxic B	3,000 - 4,499
	Hypoxic C	4,500 - 5,999
Anoxic	Anoxic	≥ 6000

Sulfide species at varying pH



Traditional method for regulatory sulfide analysis:

- Sediment samples collected by grab
- Samples stored and transported (NB, NS) or analysed immediately (BC)
- Uses silver/sulfide ion selective electrode (ISE)
- Sediment sample basified to pH > 12 with SAOB
- Sulfide ion (S^{2-}) detected

ISE method - equipment

Meter

Silver/sulfide ion selective electrode

Temperature probe

ISE method – Pros and Cons

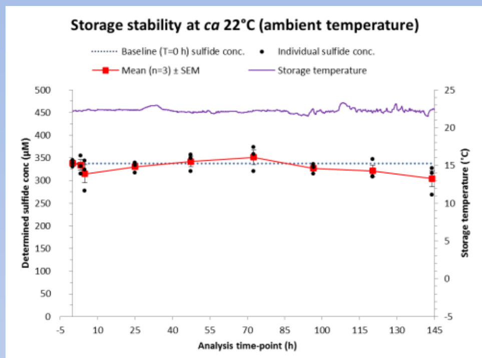
Pros

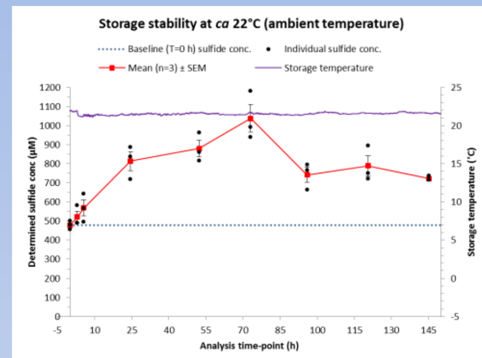
- (Relatively) inexpensive

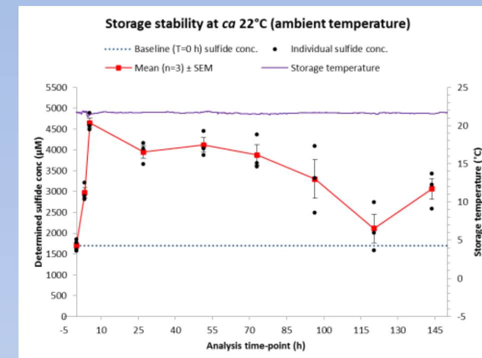
Cons

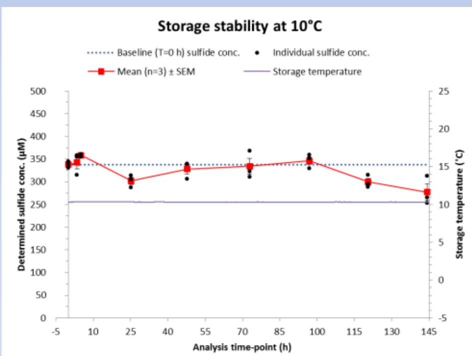
- Calibration not stable
- Temperature dependant
- Time consuming: sequential analysis
- Repeat analysis not possible – **one shot deal!!!!**
- Storage stability issues
- No extraction of porewater…measures sulfide in sediment slurry
  - Inconsistent sample matrix – **all sediments types are different!!!!!!!**
  - Possible overestimation of sulfide (Brown *et al*, 2011)

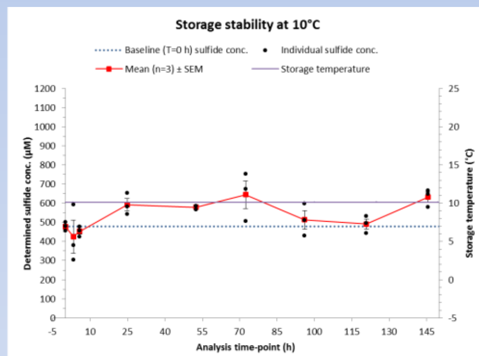
ISE method – storage stability issues

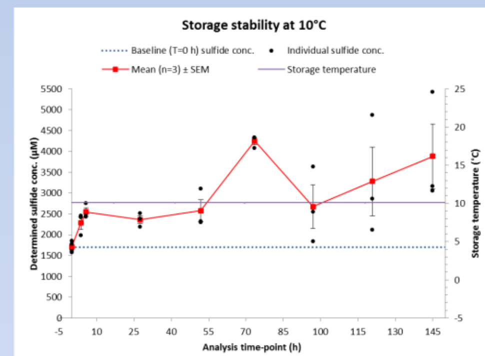


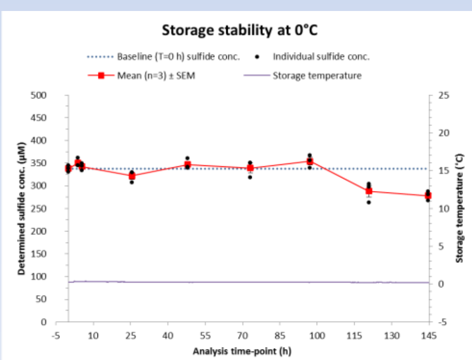


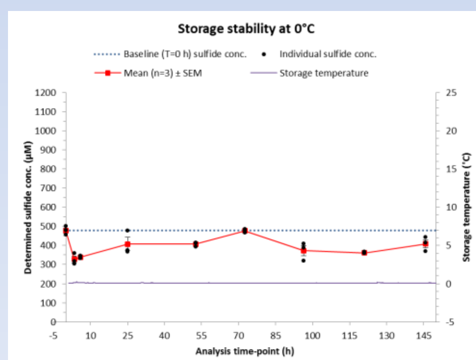


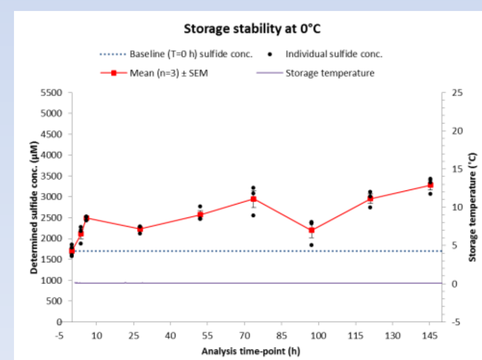










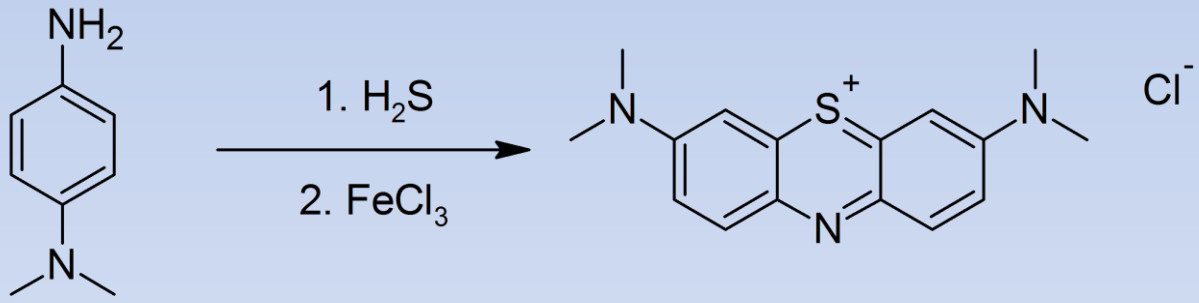


Requirements for alternative method

- Stable calibration
- Not temperature dependant
- Consistent matrix
- Storage stability of samples
- Equivalent quantification compared to ISE method
- High sample throughput

Chosen methodology:

- Sediment porewater – consistent matrix
- ‘Fix’ sulfide as an insoluble salt
- Colorimetric detection: methylene blue

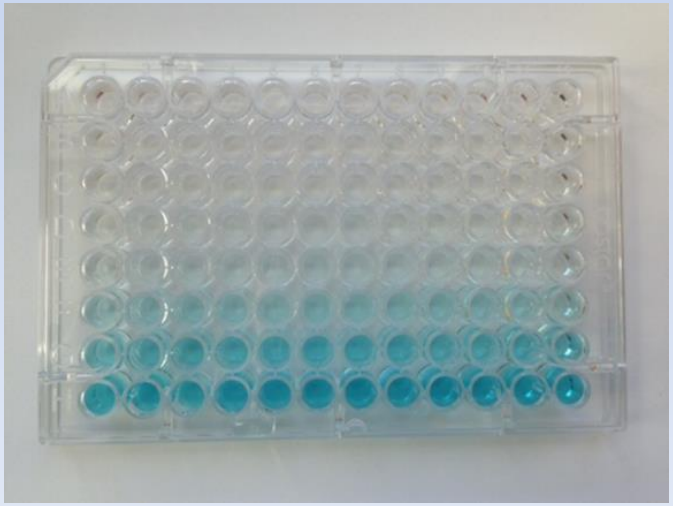


- Microplate (96 well) – simultaneous analysis of many samples

Newly developed analytical method:

- Prepare $Na_2S \cdot 9H_2O$ stock solution, titrate to obtain accurate sulfide concentration and prepare calibration standards and samples
- Transfer calibration and porewater samples (100 µL) to 1 mL of $Zn(OAc)_2$:EDTA:NaOH (1:1:0.8%, w/v) fixing solution
- Pipette calibration and fixed samples (10 µL) into 96 well plate
- Add Milli-Q water (250 µL) to each well to dilute samples
- Add DMPPDA·2HCl (20mM):$FeCl_3 \cdot 6H_2O$ (30 mM); (2:1, 40 µL)
- Incubate at ambient temperature for 5 min
- Read plate at 660 nm

Instrumental set up and example 96-well plate



Method validation parameters:

- Limit of quantification (LOQ)
- Linearity
- Accuracy and precision
- Comparison against ISE method
- Storage stability

Limit of Quantification

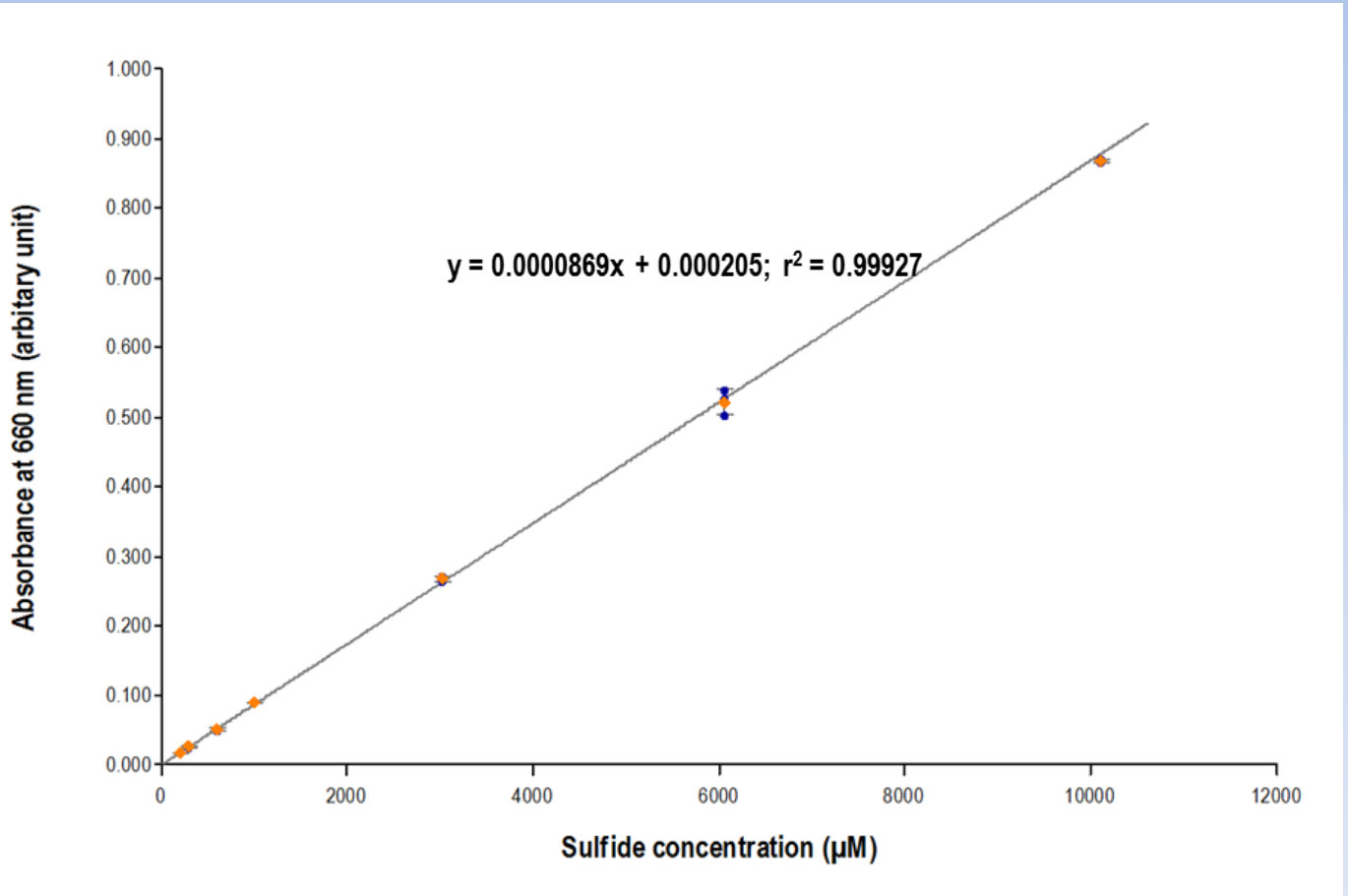
(One) definition:

”The lowest concentration at which the performance of a method or measurement system is acceptable for a specified use”

LOQ determined to be 200 µM, based on better repeatable accuracy and precision compared to 100 µM.

Linearity

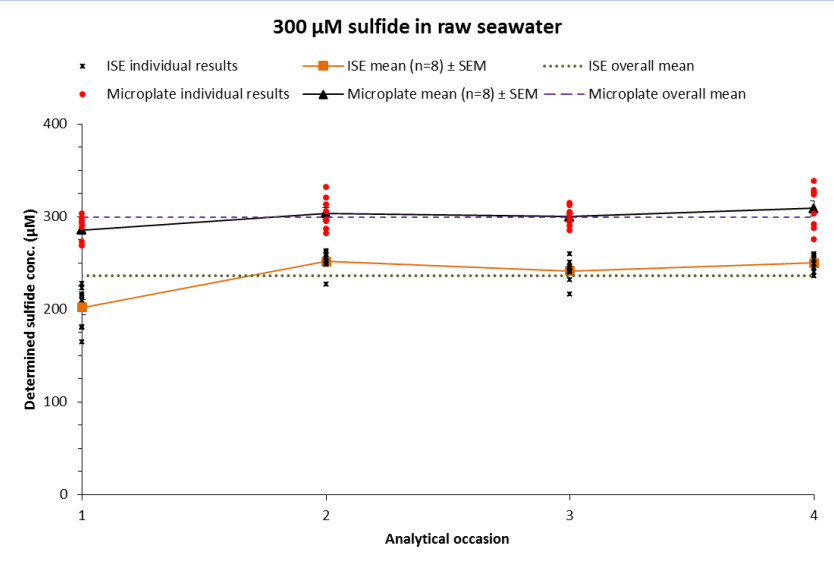
Linearity range 200 to 10,000µM

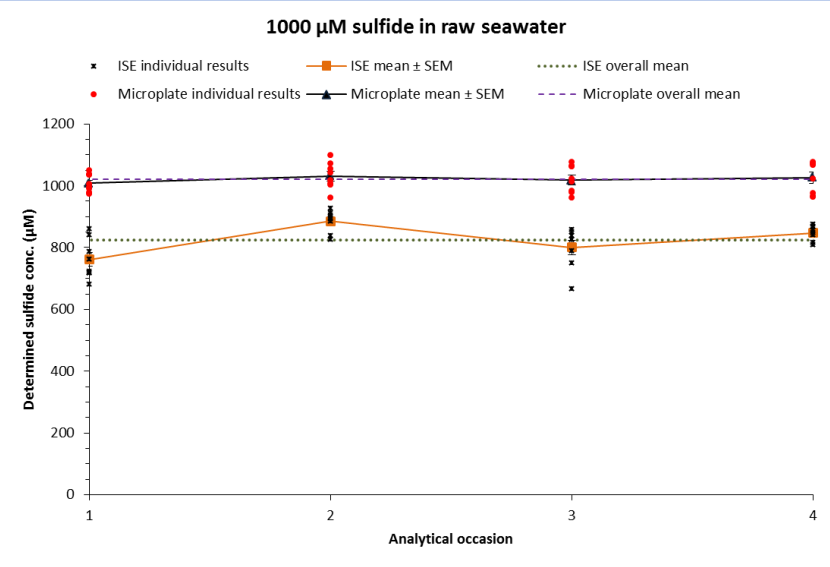


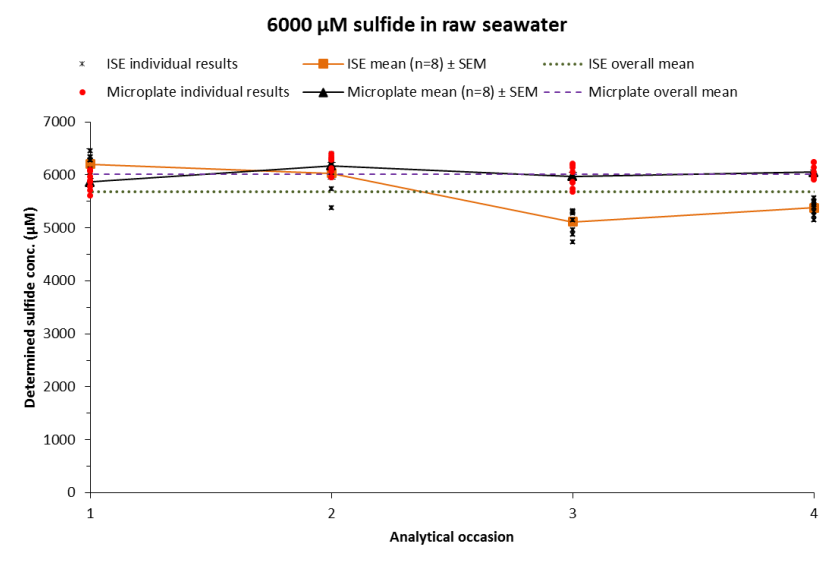
Accuracy and precision

Replicate	Nominal sulfide concentration (µM)		
	300	1000	6000
1 (n=8)	109.9%	102.6%	100.5%
2 (n=8)	97.4%	100.1%	94.3%
3 (n=8)	99.3%	100.5%	100.2%
Accuracy	102.2%	101.1%	98.3%
CV	6.6%	1.4%	3.6%

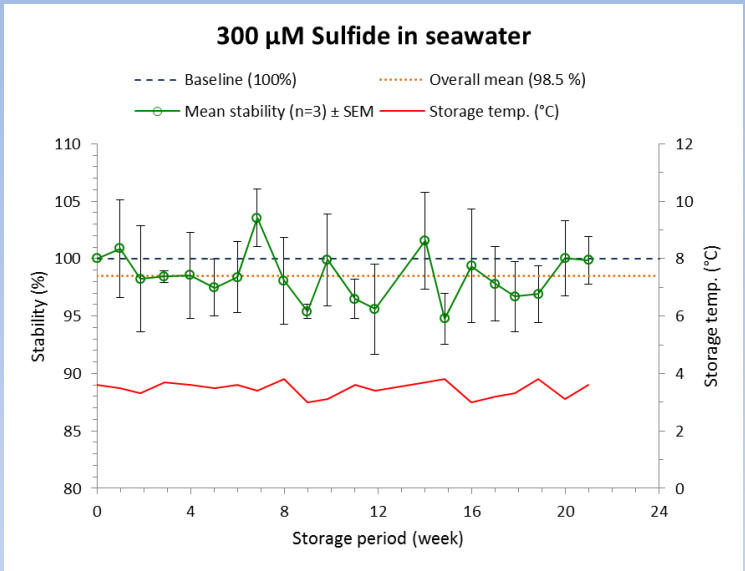
Methylene blue method *vs* ISE method

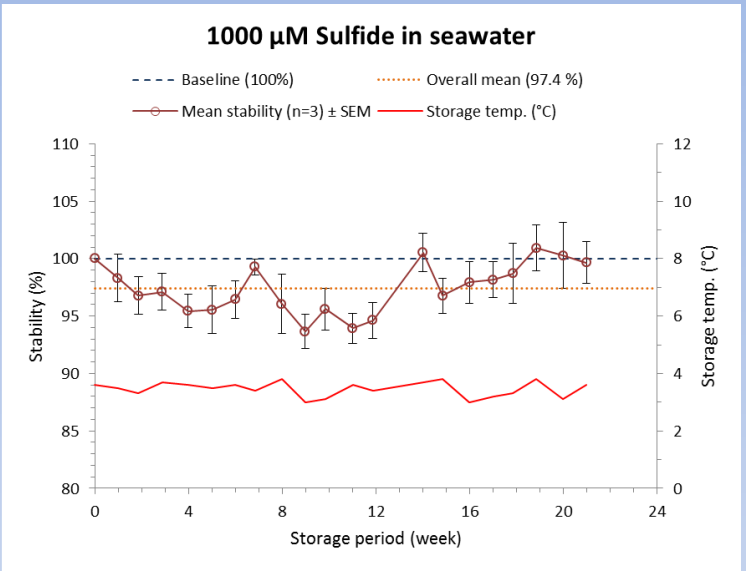


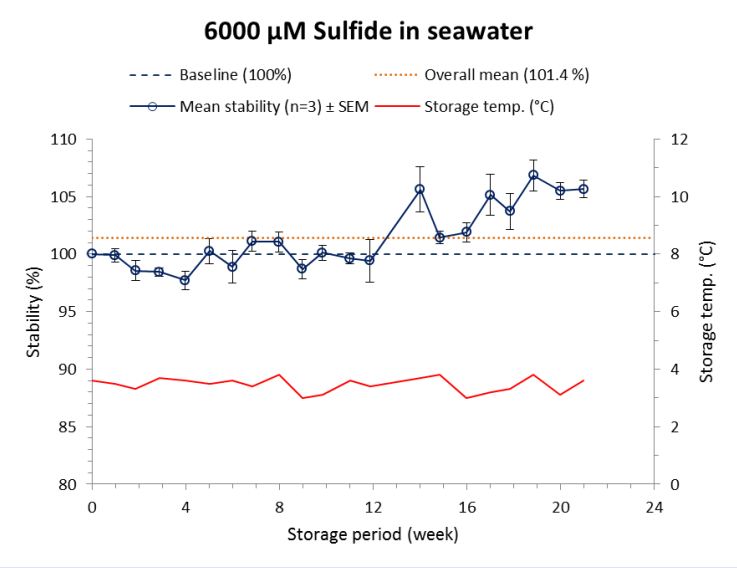


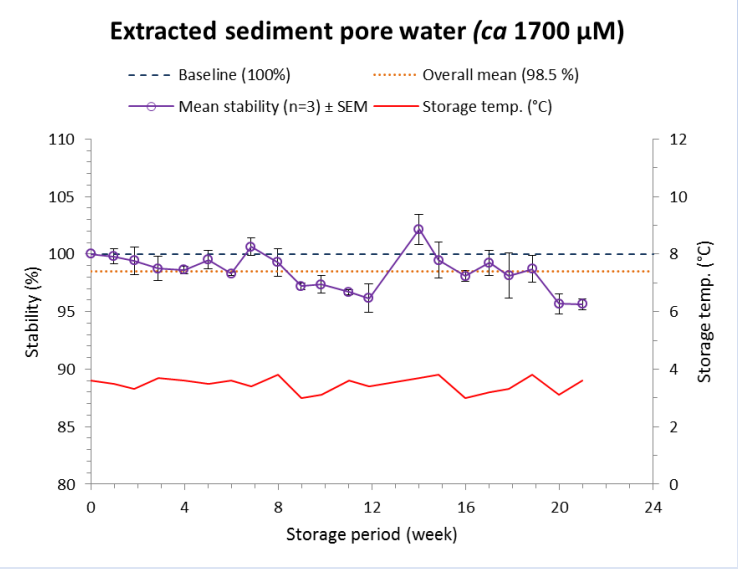


Extended storage stability at *ca* +4 °C









Methylene blue microplate method Pros and Cons

Pros

- Stable calibration
- Accurate and precise
- Low reagent volumes used
- Very high sample throughput (**up to 240 samples in 3 hours**)
- Sample stability up to 19 weeks (so far).
- Audit trail for changes to generated raw data (depending on type of plate reader)
- Comparable to ISE method
- Not temperature dependant

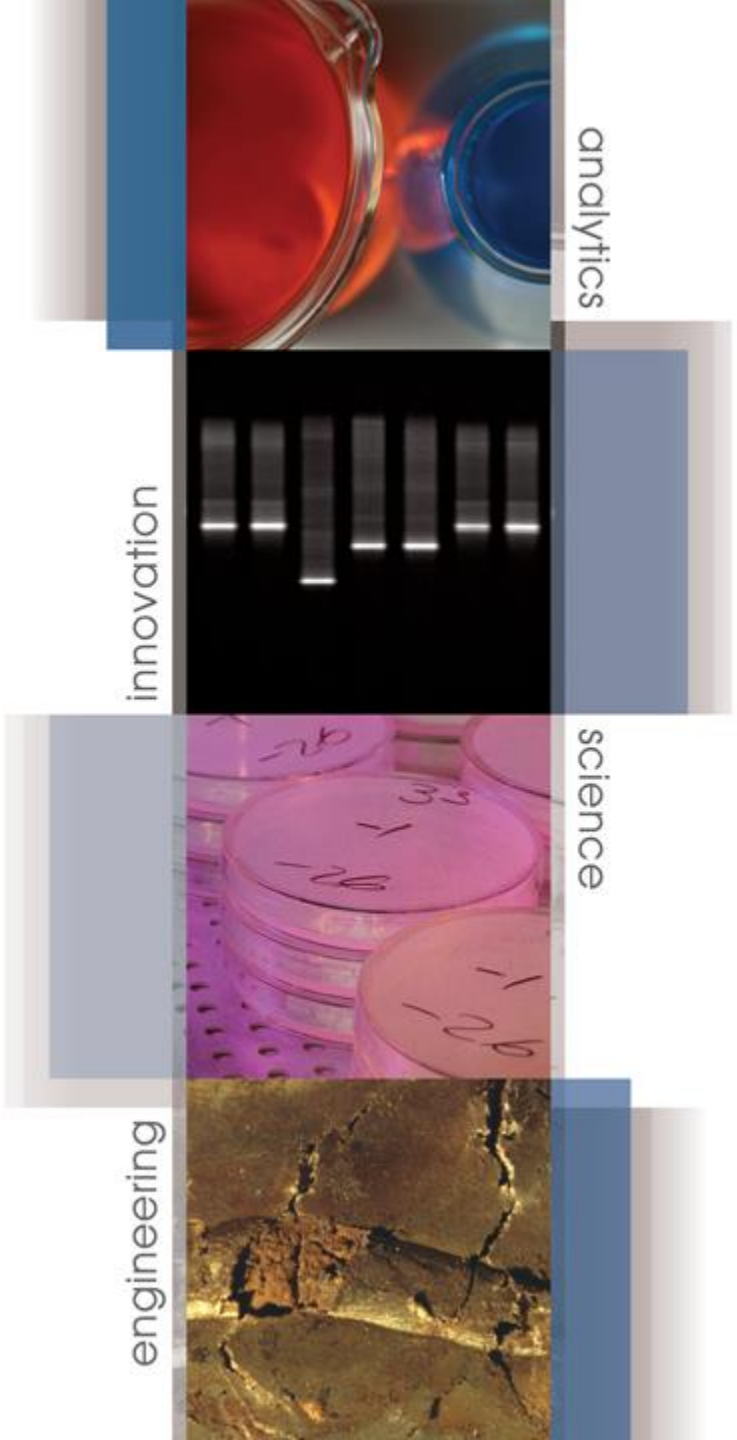
Cons

- Initial cost (depending on type of plate reader)

rpc
SCIENCE & ENGINEERING

ISAv: What is a Strain?

Benjamin S. Forward, PhD



Outline

- ISAv: basic overview and anatomy
- Strain typing tools
- Strains & virulence determinants
- Outstanding questions

ISAv

- Causative agent of infectious salmon anemia
- First detected in Norway in 1984
- Responsible for outbreaks in NB in late 90's
- Subsequently in NS and NL
- Outbreaks still occur today
- Impact reduced due to improved management practices

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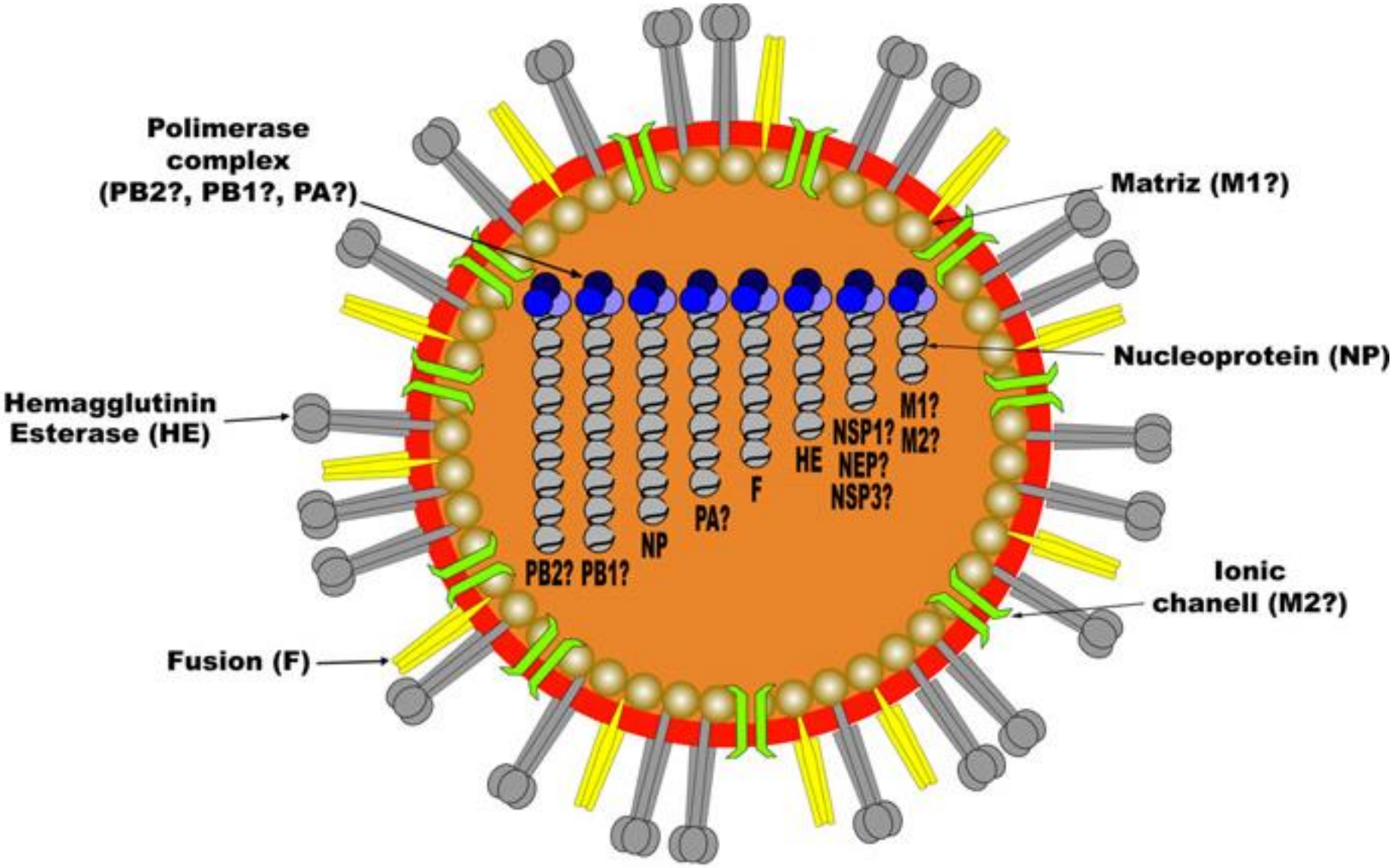
ISAv

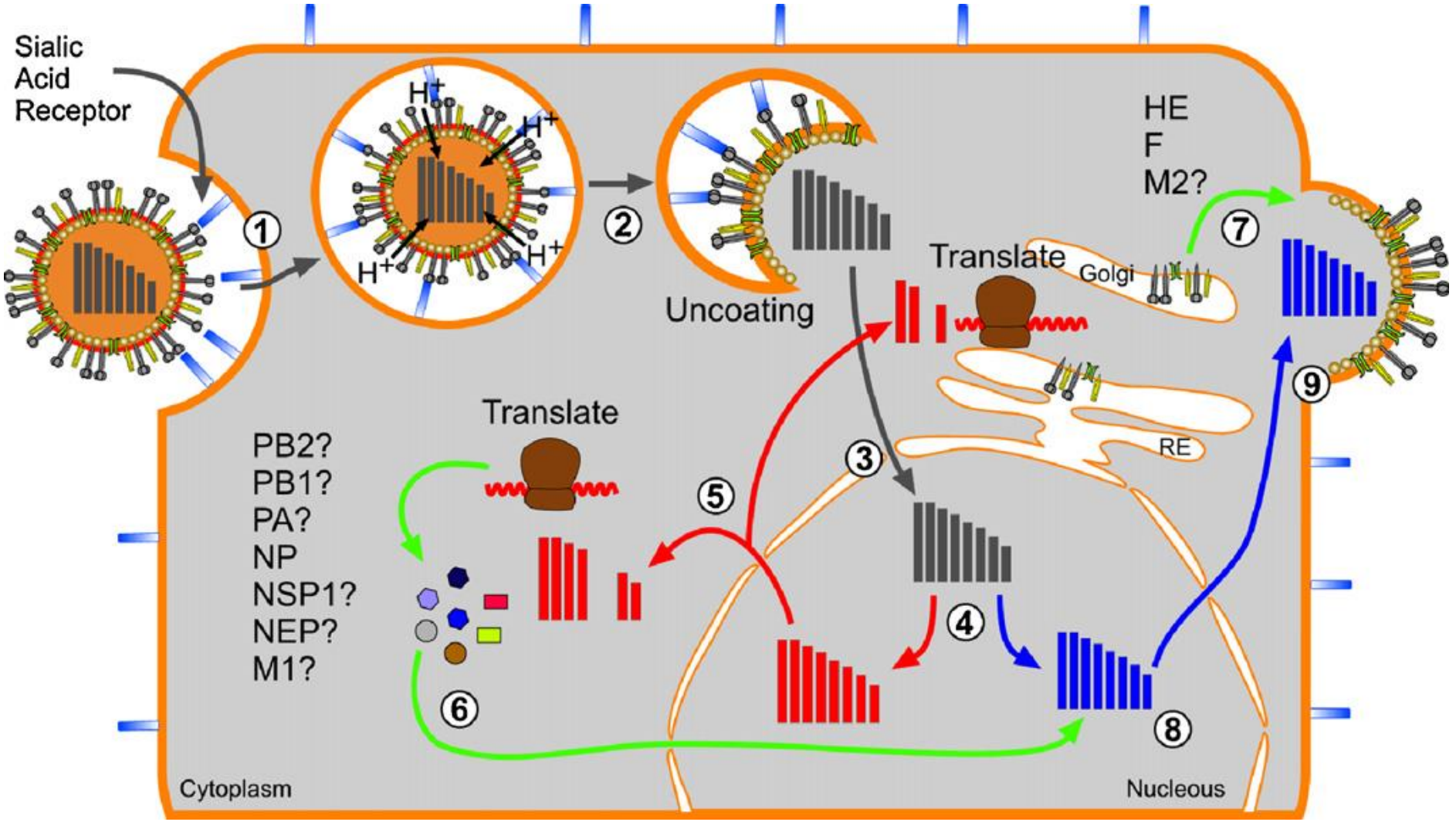
- Orthomyxovirus
- Isavirus
- 8 segment RNA virus (-ve strand)
- Encoding 10-11 different proteins

100-120 nm in diameter



Cottet et al 2010

(4-*O*-acetyl sialic acid)



Cottet et al 2010

Strain Typing Methods

Goal: to uniquely identify and distinguish different isolates and ultimately predict potential virulence

- Antibody reactivity (e.g. salmonella)
- SDS-PAGE & 2D gel electrophoresis
- DNA Typing Methods
  - RAPD, PFGE, PCR-RE
- DNA sequencing

Strain Typing Methods

Overall, there is a movement to techniques which identify and characterize the molecular determinants of virulence

- DNA sequencing and determination of inferred amino acid sequence from specific gene segments

Strain Typing ISAv

DNA sequencing of Seg 6 (HE)

- Primary indicator/marker of potential virulence
- HPR (highly polymorphic region)
- Sequence ~201 bp (67 aa HPR0)

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HPR Types

```
H0              SLGNTDTLIMREVALHKEMISKLQRNITDVKIRVDAIPPQLNQTFNTNQVEQPSTSVLSNIFISMGV
H0.a            ....................I.......................................
Hpr8(Euro)      ....................................------------------------GV
Hpr2(Euro)      ....................................--------------------MGV

HPR0.RPC#NA     G......Q....LETQR...G..S......NN............LGV...................
HPR0.RPC#Na.a   G......Q....LEAQ....G..S......NN............LGV...................
Hpr2            G......Q....LEAQ....G..G......NN............--------------------MGV
Hpr2.a          G......Q....LEAQ.G..G..G......NN............--------------------MGV
Hpr2.b          G......Q....LKAQ....G..G......NN............--------------------MGV
Hpr4            G......Q....LEAQ....G..G......NN.........----------------SNIFISMGV
Hpr4.a          G......Q....LEAQ....G..G......NN..A......----------------SNIFISMGV
Hpr4.b          G......Q....LEAQ....G..G...A..NN.........----------------SNIFISMGV
Hpr6            G......Q....LEAQ...TG..G......NN.....-------LGV..........P.........
Hpr-RPC#9       G......Q....LEAQ....G..G......NN..-----------GV...................
Hpr-RPC#10      G......Q....LEAQ....G..G......NN....--------------------.......MGV
Hpr-RPC#19      G......Q....LEAQ....G..G......NN..........---------------SNIFISMGV
```

- NA vs EU
- Virulent vs avirulent

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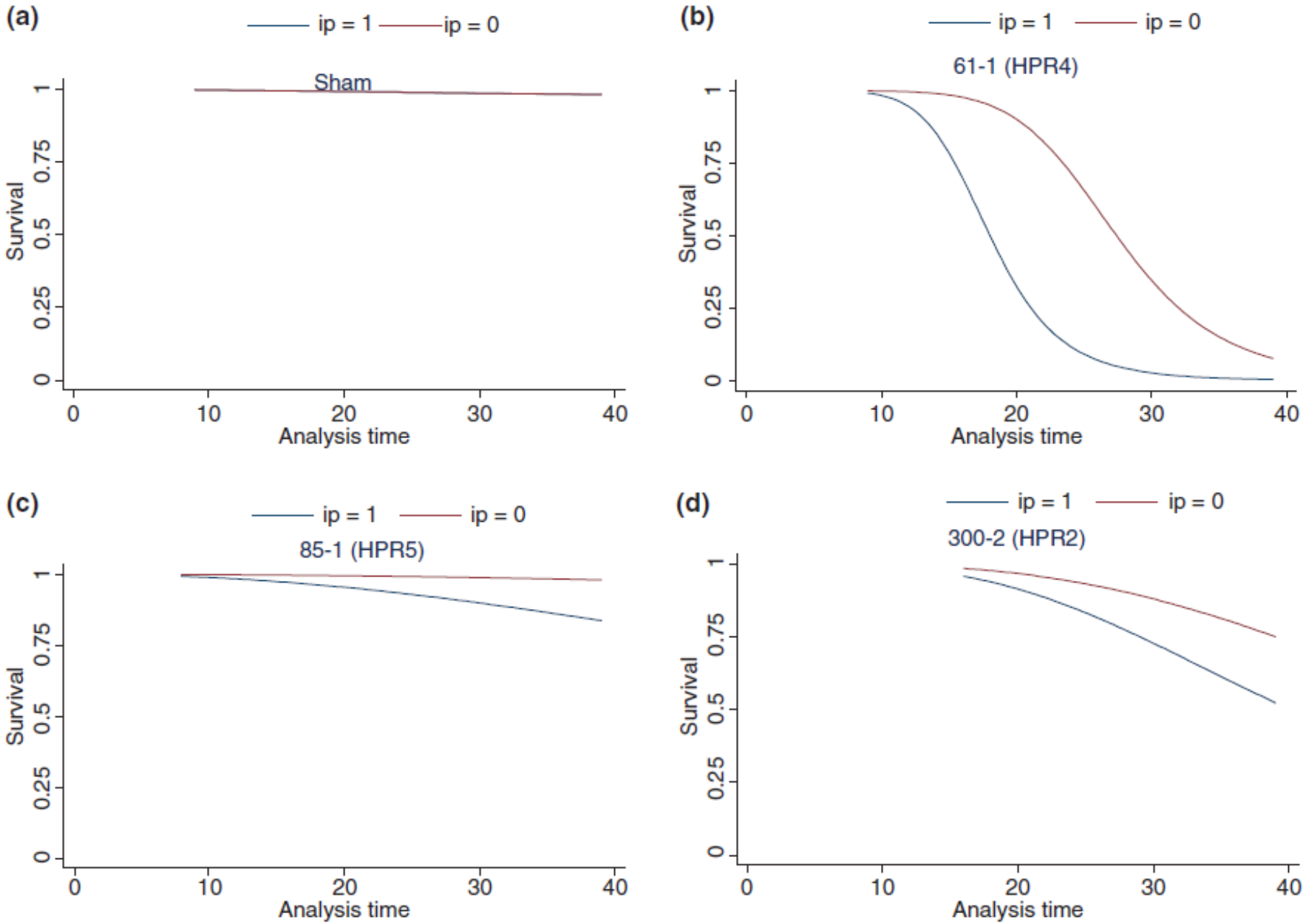


Ritchie et al 2009

Strain Stats

- Detected and sequenced over 40 different HPR types since 2004
- Nearly all in past 5 years have been new
- Still detect EU HPR0 regularly, NA HPR0 is rare

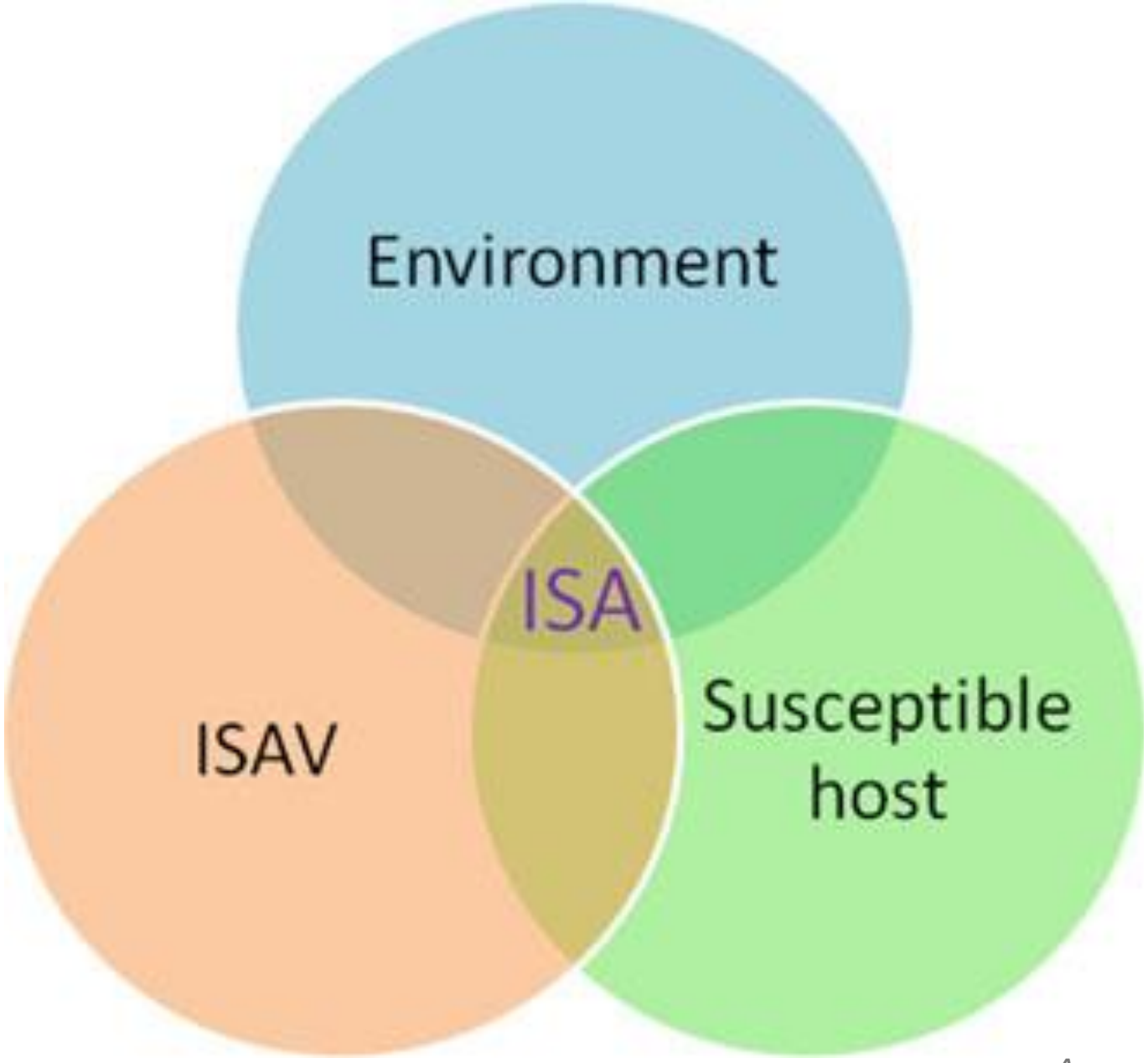
ISAv HE Deletion Role in Virulence

- Deletions do not appear to affect receptor binding (McBeath, 2011)
- Deletions do not appear to affect esterase activity (McBeath, 2011)
- Deletions do appear to influence activity of fusion protein (Fourrier 2014, 2015)

Strain Typing ISAv

Segment 5 – Fusion protein

- Also important for virulence
- Mutations affecting cleavage site likely influence fusion activity (Fourrier 2015)
- FP functionally induced by HE-Del to enhance fusion (Fourrier 2015)



Aamelfot et al 2014

Outstanding Questions

- What precipitates the transition from avirulent to virulent (HPR deletion) – natural reservoirs?
- Fully understand the role of HE (Seg6) deletions in virulence wrt interaction with fusion protein (Seg5)
- Relative contributions of fusion protein (Seg5) mutations to virulence
- Relationship of infectivity/virulence to pathogenesis

rpc

Thank You!

rpc

Significance of HPR0 in Relation to ISA Disease Caused by HPR-Deleted ISA Variants

Dr. Knut Falk, PhD, Senior scientist
Norwegian Veterinary Institute

Veterinærinstituttet
Norwegian Veterinary Institute

Outline of presentation

- Global distribution and control of ISA virus
- Infection by HPR-deleted ISAV vs. HPR0 virus
- A Faroese example representing the first field evidence of direct transition from a non-virulent HPR0 to a virulent HPR-deleted ISA virus

Virulence = The capacity of a microbe to cause disease

ISA virus international control - OIE

- Both the virulent HPR-deleted types, and the non-virulent HPR0 type are listed by the World Organization for Animal Health (OIE), i.e. findings must be reported.
- The World Organization for Animal Health (OIE)
  - Is an instrument for the World trade organization (WTO)
  - Lists animal, fish, and shellfish diseases that may compromise commercial activities or wild animal stocks. These diseases must be reported to the OIE
  - Publish a number guidelines related to disease control, including procedures for detection and diagnosis
  - Note: A country may legally stop import if any OIE-listed infection is found

Global distribution of virulent HPR-deleted ISA virus

- Norway:
  - 2-20 annual outbreaks
  - Occurs often as small localized epidemics
  - The source is often unknown, but have recently seen a few cases where virus has been transferred by infected smolt
  - Control is based on identification of diseased fish
- Eastern Canada and Chile:
  - Endemic with sporadic detections/outbreaks
- Scotland and the Faroe Islands:
  - In principle ISA free since epidemics in 1998-99, and 2000-05, respectively. However, ISAV have been detected once in each country since these epidemics.
- Western Canada, Ireland, Tasmania:
  - No officially reported ISA virus detections

Global distribution of non-virulent HPR0 ISA virus

- Extensive PCR screening on the Faroe Islands revealed (Ref. Christiansen et al. 2011, J.Gen.Virol.)
  - HPR0 virus cause a transient/passing infection, mostly localized to gills
  - All Atlantic salmon populations went through one or more short-lived infection episodes, including in smolt farms.
  - Overall prevalence of HPR0 ISA virus in random sampled gills were +/- 10%
- Published and non-published information from Norway, Scotland, Chile and Eastern Canada suggest comparable prevalence's in gills.
- HPR0 ISA virus has not been found in western Canada in spite of testing thousands of fish. There are no available information from Ireland and Tasmania.

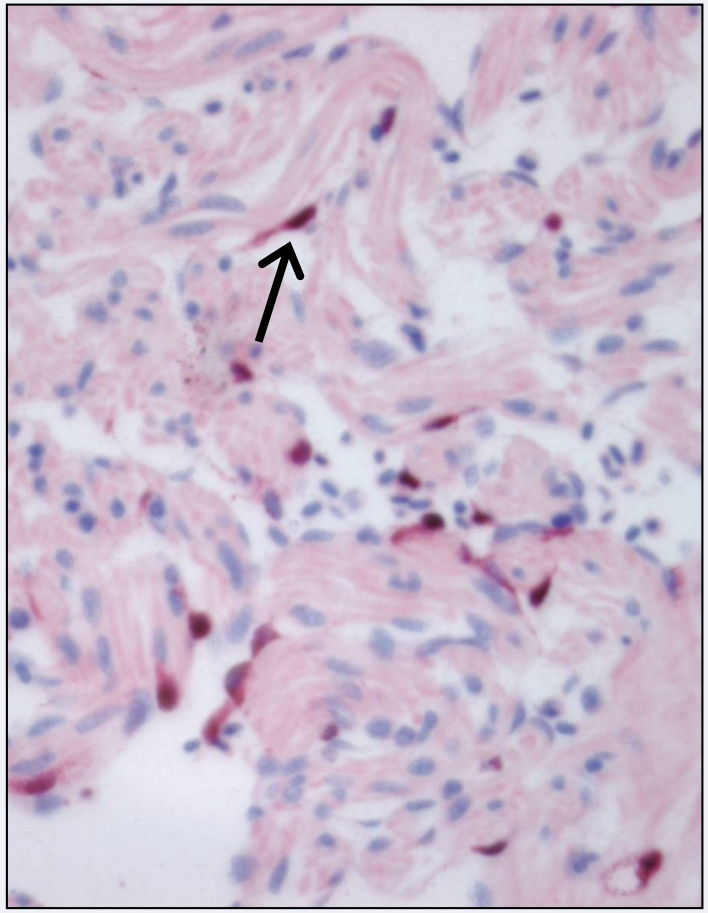
The HPR0 hypothesis

- Virulent HPR-deleted ISA virus develops from non-virulent HPR0 virus
- Questions and challenges (from a regulatory point of view):
  - How often does this transition occur?
  - What is the drivers for this transition?
  - What is the risk of this transition when HPR0 ISA virus is detected?
  - Are there other necessary changes needed to get a fully virulent virus?
  - Is the transition a step-by-step process which include low-virulent intermediates?

Differences in desease appearance between HPR0 and HPR-deleted ISAV

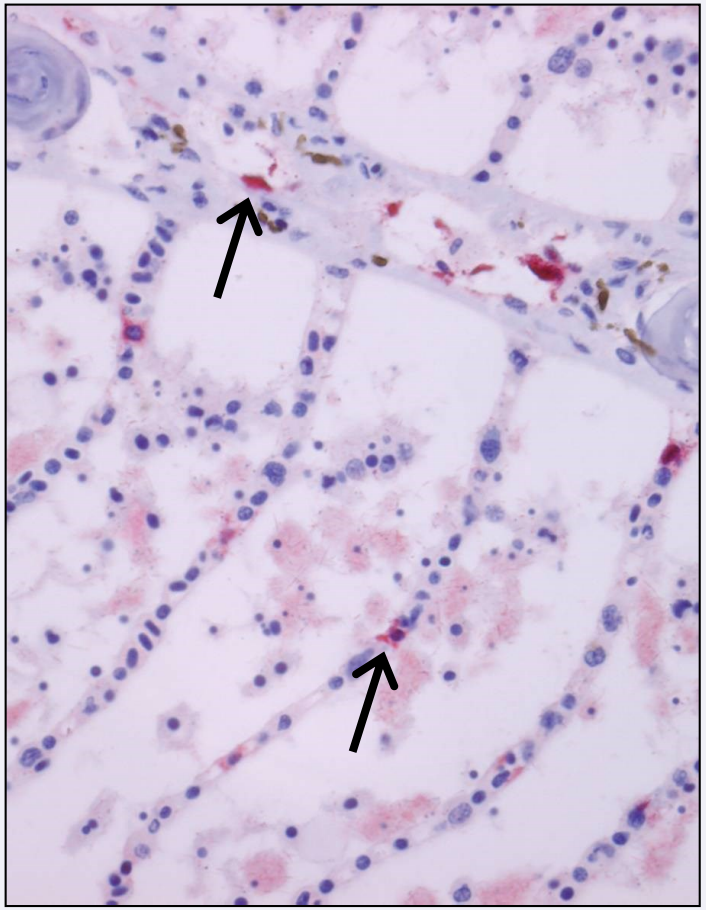
HPR-deleted ISAV	HPR0 ISAV
Virulent – cause disease	Non-virulent – no clinical signs
Generalized infection	Localized infection
Infects internal organs	Infects mucosal surface (gill, skin)
Target cells: endothelium	Target cells: epithelium
Progressing infection, often lethal	Short duration/passing (transient)

«Classical» ISA is an infection of cells lining the the blood circulatory system (i.e. endothelial cells)



Heart – infected endothelial cells are pink

(Photo: Maria Aamelfot, NVI)

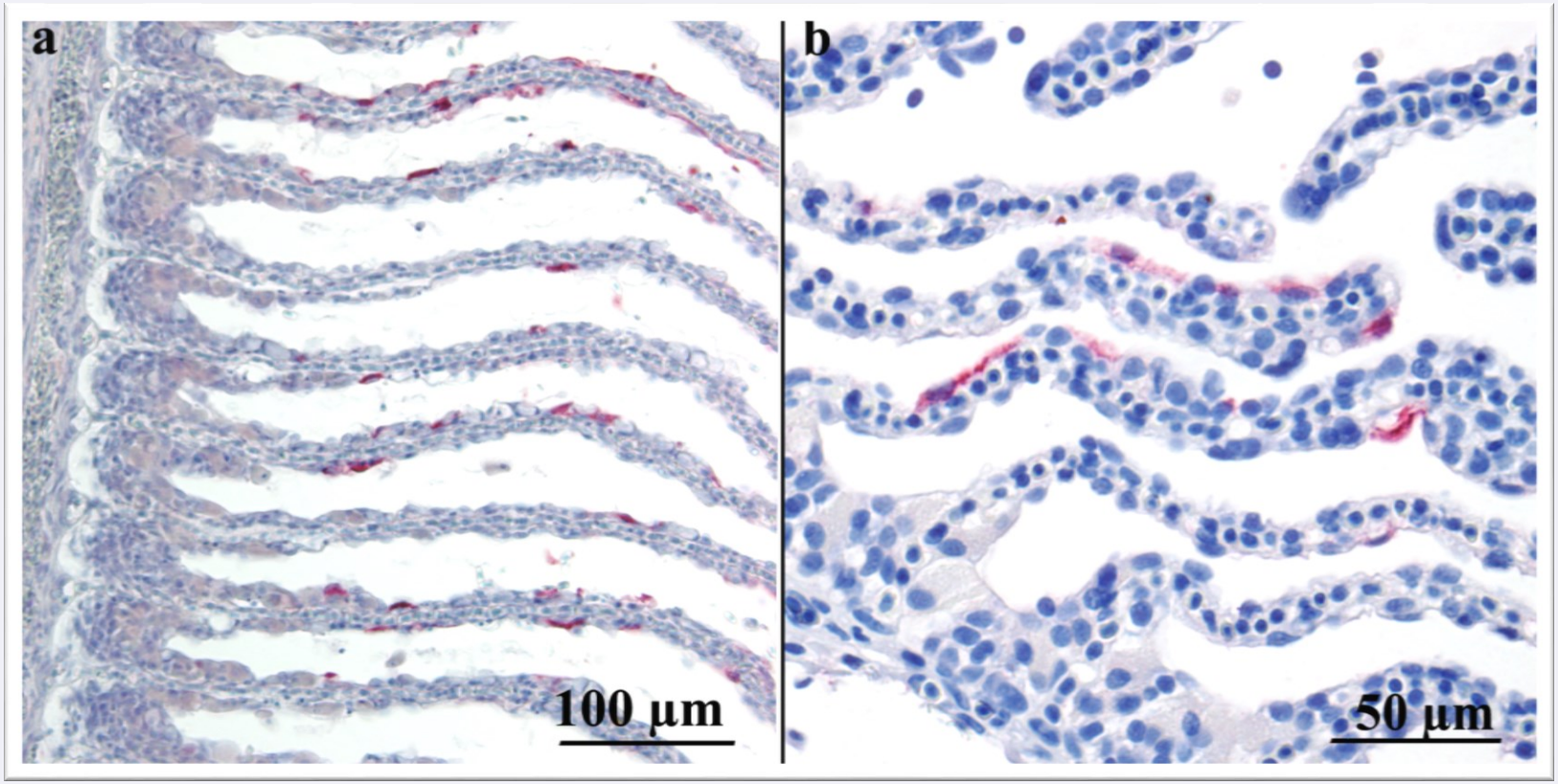


Gill – infected endothelial cells are pink

(Photo: Maria Aamelfot, NVI)

Norwegian Veterinary Institute

Infection by HPR0 ISA virus cause an epithelial infection of body surfaces (i.e. gill and skin)

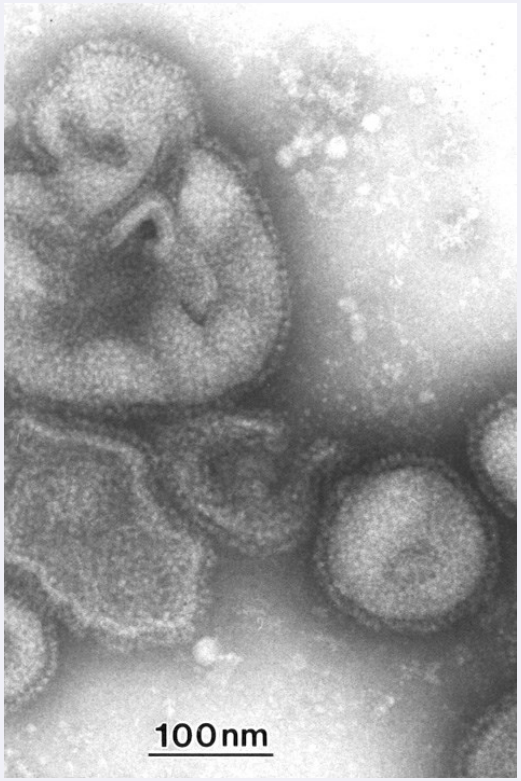


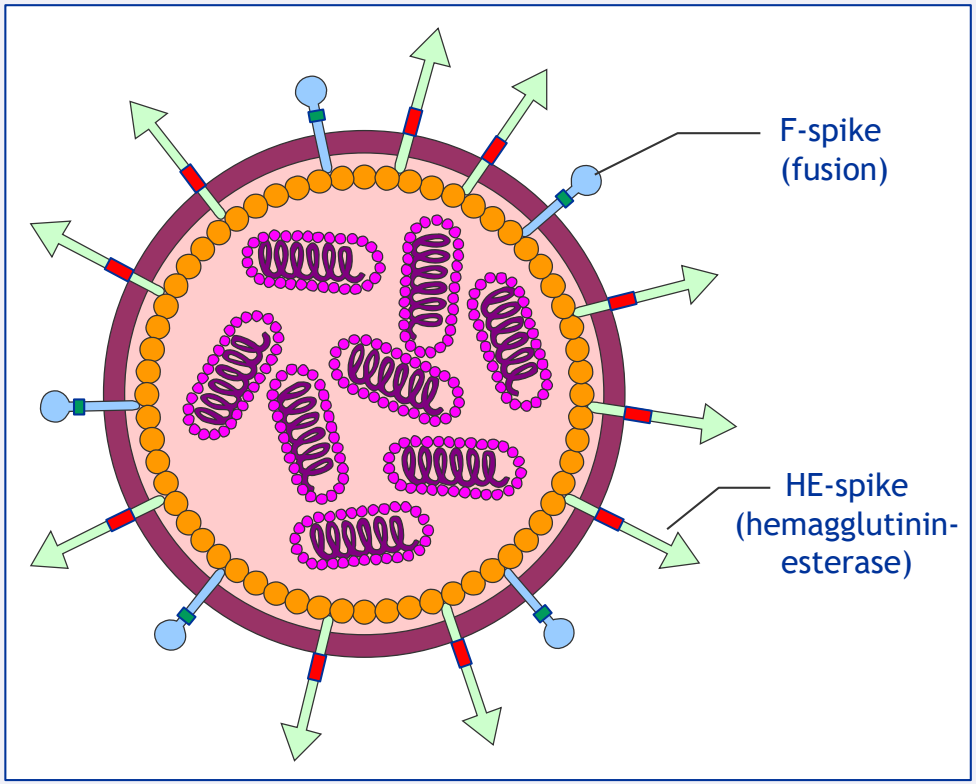
(Photo: Ole Bendik Dale, NVI)

Gill sections – infected epithelial cells are pink – no infection of circulatory system

A Faroese example representing the first field evidence of direct transition from a non-virulent HPR0 to a virulent HPR-deleted ISA virus

ISA virus structure – What are HPR types?





(Photo: Ellen Namork, Norwegian Institute of Public Health)

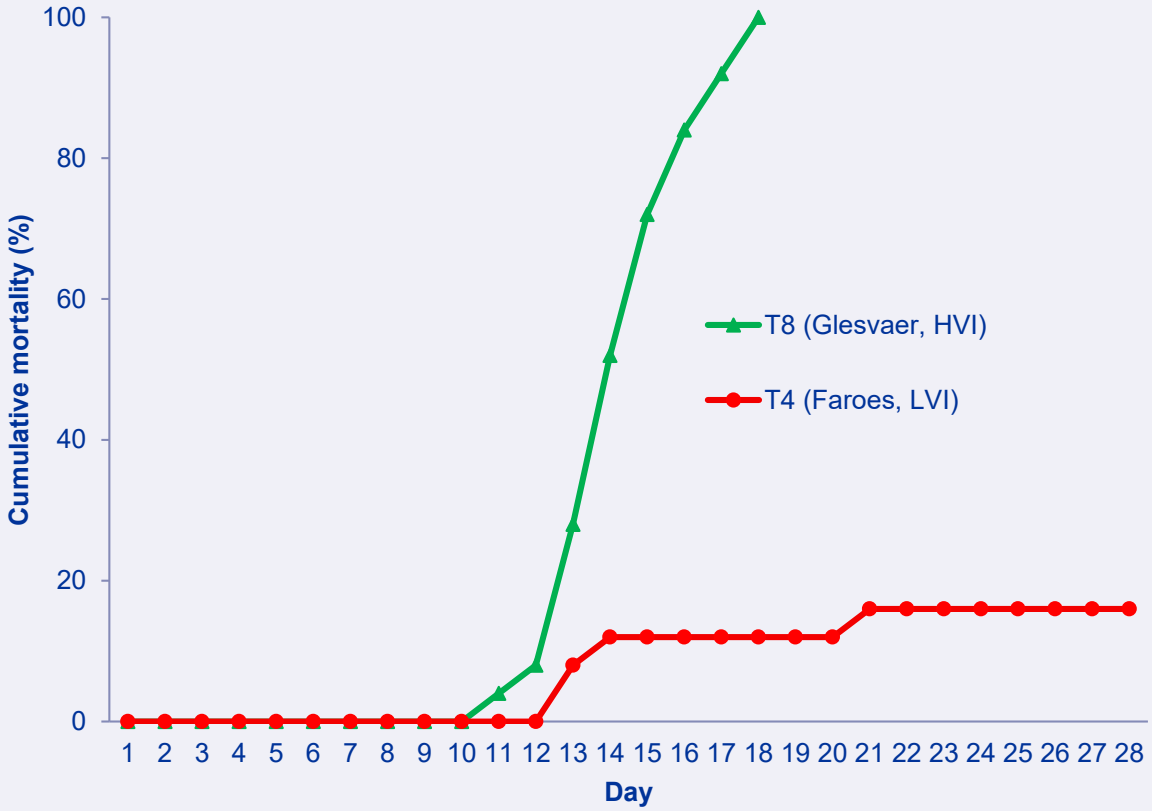
- Virus causing «classical» ISA all have a shortened HE stalk (HPR-deleted) and a small change in the F-protein relative to the original non-virulent HPR0 type

- These changes are key factors for ISA virus virulence and disease characteristics, and together modify viral fusion activation, and activity

A Faroese example representing the first field evidence of direct transition from a non-virulent HPR0 to a virulent HPR-deleted ISA virus

- January 2014:
  First detection of HPR-deleted ISA virus since 2005 during routine PCR screening at harvest. 3 out of 16 fish in one netpen tested positive with low virus levels. No elevated mortality or clinical signs suggesting ISA.
- February 2014:
  PCR screening of 150 fish revealed 90% ISA virus positives, and higher virus levels. Only one affected netpen, and no elevated mortality.
- Experimental infection confirms low virulence (disease-causing capacity), i.e low mortality (~10%).
  Classical pathological signs, but less pronounced.

Immersion (bath) challenge experiment



Norwegian Veterinary Institute

Epidemiological examinations

- The affected farm had received smolts from 4 different smolt producers.
- Examination including sequencing of previously collected samples from the smolt farm that had delivered fish to the affected net pen revealed a HPR0 virus with very closely related HE-, and F-genes.
- The HPR0 and HPR-deleted virus could not be found in 1800 screening samples collected during the seawater production phase in the affected farms and 6 neighboring farms (i.e. in the affected management area). 140 of these samples were HPR0 positive, but not related to the two related viruses.

Genome sequencing

- The whole genome of both the new HPR-deleted, and the related HPR0 virus were sequenced.
- The only difference found between the HPR0 and HPR-deleted virus was:
  - A deletion in the HPR-region of the HE-gene
  - A single amino acid mutation in the F-gene
  - Apart from these changes, they were identical

What can we learn from this example?

- We have for the first time provided practical support for the HPR0 hypothesis.
- Our results demonstrated that deletions in the HPR-region of the HE-gene, combined with a mutation in the F-gene, are the minimum requirements for a shift in infection pattern from a localized, to a generalized infection.
- However, the observed changes were not enough to obtain a fully virulent ISA virus.
- We propose that the transition from non-virulent HPR0 virus to a fully virulent HPR-deleted virus is a stepwise process requiring more unknown changes to the virus, and thus also involving low virulent intermediates that may be difficult to detect.
- The two viruses were not detected in the seawater production phase, in spite of extensive screening.
- We believe that the transition may have occurred late in the seawater production phase, and that the drivers may have been various stress episodes, including peroxide treatment, heavy storms and ulcers.

Thanks to collaborators at:

- Marine Laboratory, Aberdeen, UK:
  Alastair McBeath, Iveta Matejusova, Mickael Fourrier, and Mike Snow

- Faroese Food and Veterinary Authority:
  Debes Christiansen and Peter Østergård

- Norwegian Veterinary laboratory:
  Maria Aamelfot and Ole Bendik Dale

ISAv
Opportunities for Health Innovation

M J Beattie DVM MRCVS
Chief Veterinarian NBDAAF

ACFFA Oct 2016

New Nouveau Brunswick
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Introduction

"Frame the right question and one will move forward, fail to do so and you will take 2 steps backward"

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Historical and Present Status

- Number of cases 2015/16
- Regions of outbreaks
- Strain types encountered
- European strain types
- Virulence of strain types
- Presence/Absence of HPR0
- Observations
  - Herring and juvenile hake

ISAv Status 2014 - 2016

- 2014
  - 0 cases
- 2015
  - 16 cases
  - 3 sites
  - 2 BMA’s
- 2016
  - 10 cases
  - 7 sites
  - 3 BMA’s

- HPR 0
- HPR 2 & 2.a Euro
- HPR 14
- HPR 15 & 15a
- HPR 16
- HPR 17 Euro
- HPR 18 Euro
- HPR 19

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ISAv Status 2014-2016

- Cases by BMA and Marine Site

• BMA 1	2 cases	1 site	2016	Euro
• BMA 2a	3 cases	1 site	2015	NA
• BMA 2b	6 cases	2 sites	2015	NA
• BMA 3a	1 case	1 site	2016	Euro
• BMA 3b	14 cases	5 sites	2016	Euro 2
•				NA 3

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ISAv HPRO Implications

- BMA 1 10 cases/6 sites 1 site + Euro
- BMA 2a 3 cases/2 sites
- BMA 2b 1 case/1 site
- BMA 3a 6 cases/2 sites
- BMA 3b 12 cases/5 sites 1 site + Euro
- 1 site + Euro/NA
- 2 sites + NA
- 5/16 sites led to virulence ISAv

New Nouveau Brunswick
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ISAv HPRO vs Virulent Strains

- Virulent strains 2003-16 1% Euro/ 99% NA
- Virulent strains 2015-16 20% Euro / 80% NA

- HPRO's 2003-16 85% Euro / 15% NA

- HPRO to virulent strain at same site 2015-16
  - 31 % Euro / 69 % NA

Observations/Unanswered Q's

- Wild reservoir population
- HPR0 mutations lead to virulent forms
  - Euro vs. NA strain types
- Vaccine efficacy improvement
  - Strain type specific ?
    - Colour filter
  - New adjuvant
  - Choose more stable region of segment 6
  - Add segment 5 to vaccine

INFECTIOUS SALMON ANEMIA VIRUS: AN UNWELCOME GUEST ON EXTENDED STAY

Nellie Gagné[1]* , Francis LeBlanc[1] , Delphine Ditlecadet[1], Steve Leadbeater[2]
Fisheries and Oceans Canada, Gulf Fisheries Center, Aquatic animal Health, Moncton
Fisheries and Oceans Canada, St-Andrews Biological Station

Canada

Outline of presentation

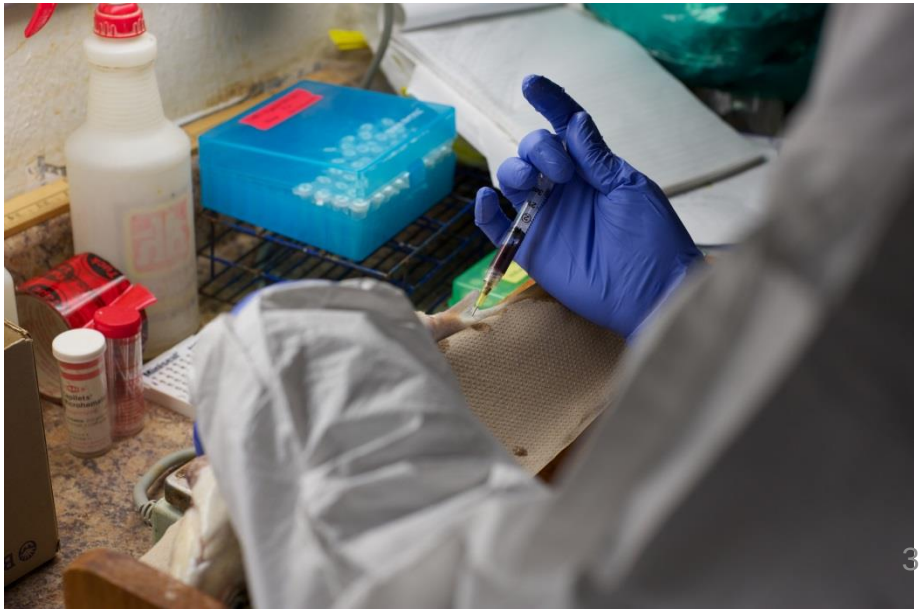
- Aquaculture and disease
- Infectious salmon anemia
- HPR0 vs HPRΔ
- Atlantic Canada, 20 years of history with ISAV
- Cases 2012 to 2016
- Where are we heading

Aquaculture and diseases associated

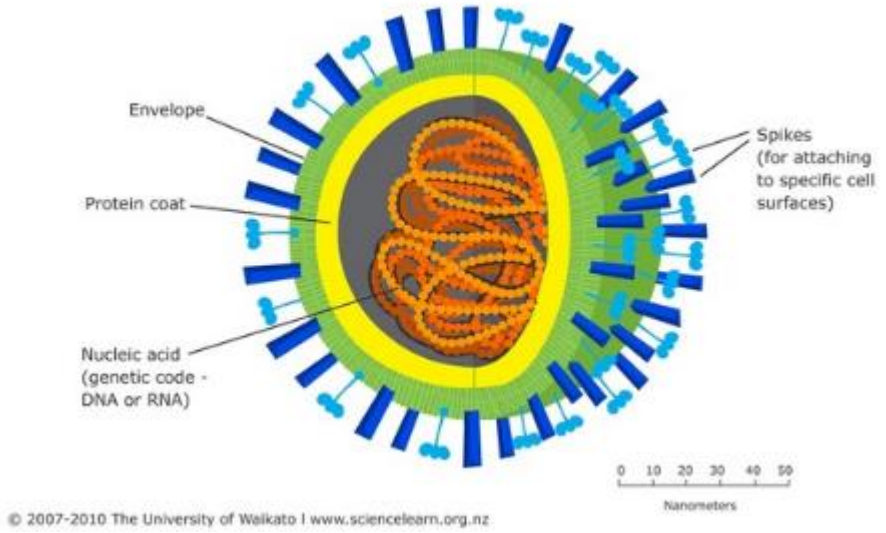
Intense rearing = stress = disease = prophylaxis, treatment, vaccines

Need to understand in order to mitigate (past research)

- Natural immunity vs induced by vaccination
- Cross-protection vs strain of virus
- Immune response (primary, secondary, by tissue and exposure)
- Dose (minimal infectious dose), shedding rate, UV and virus viability in SW
- Effect of family, wild vs cultured fish (genetic background), environment (temperature, stress) and viral strain
- etc



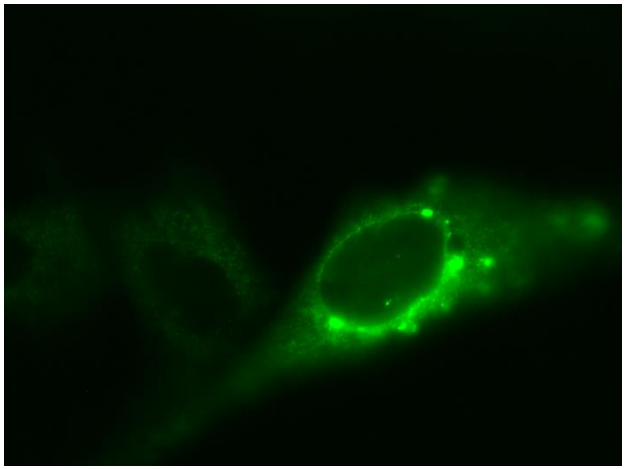
What is a virus?

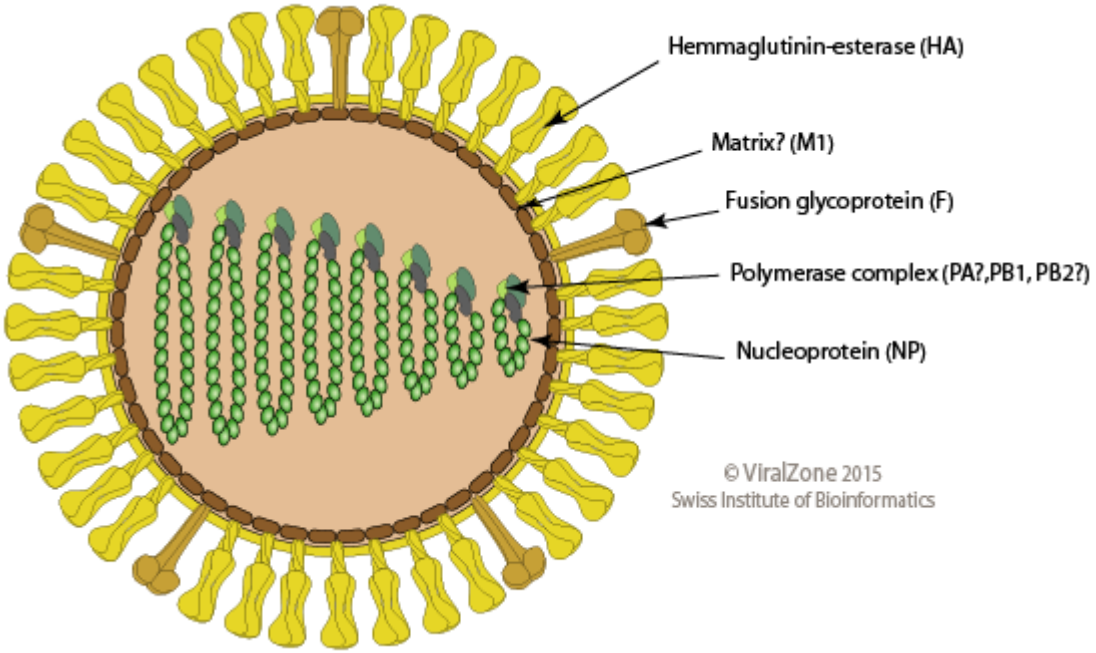


A virus is a biological agent that reproduces inside the cells of living hosts. When infected by a virus, a host cell is forced to produce many thousands of identical copies of the original virus at an extraordinary rate (Wikipedia)

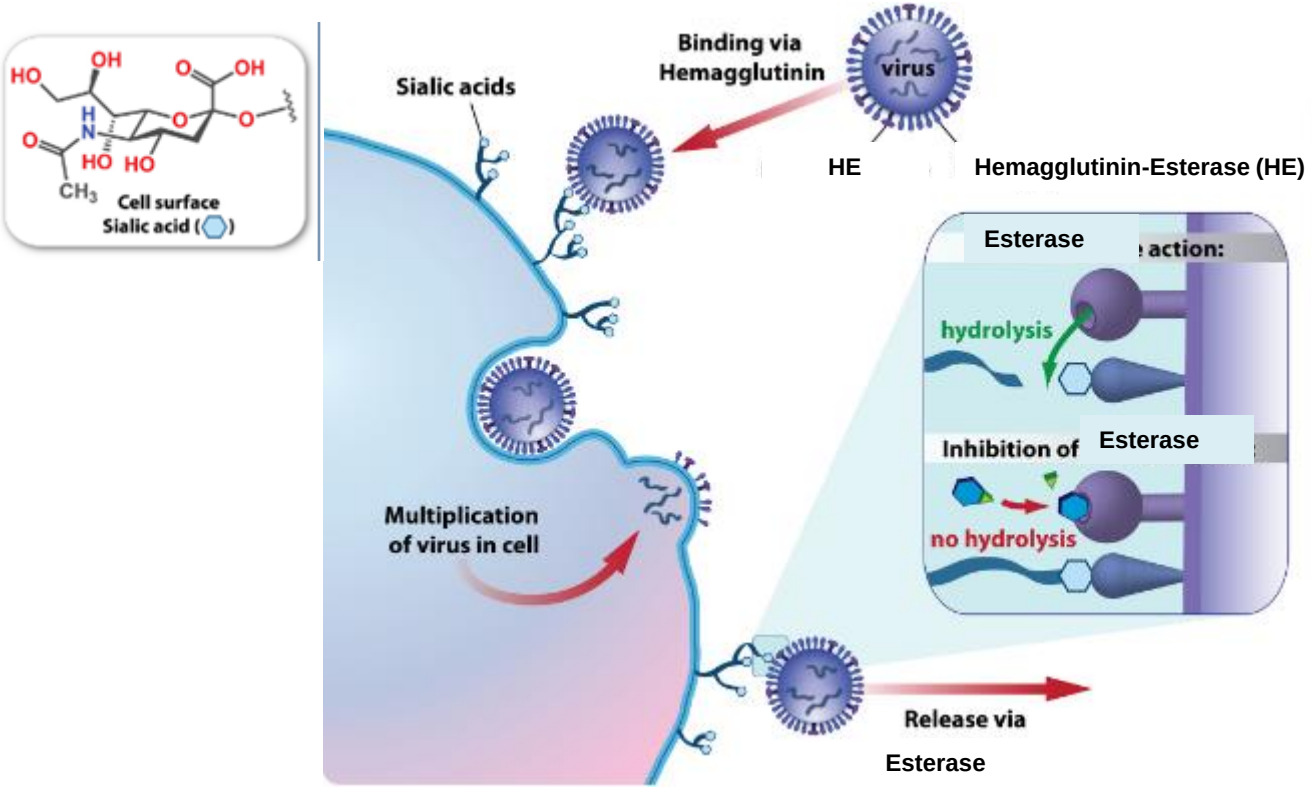
Introduction to infectious salmon anemia virus

- Family: Orthomyxoviridae, Genus: Isavirus
- Envelopped, (-)ssRNA, 8 segments (10 proteins)
- Segment 6 = hemaglutinin-esterase (HE)
- Segment 5 = fusion
- Anemia, congested blood vessels, mortality can be high



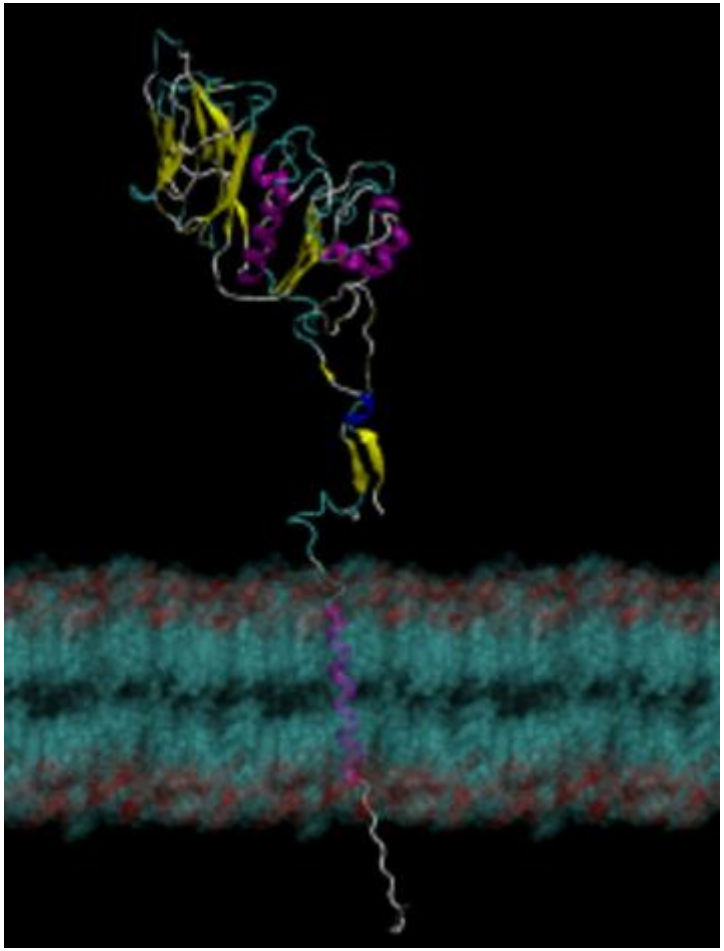


Two critical components in ISAV: HE and fusion protein

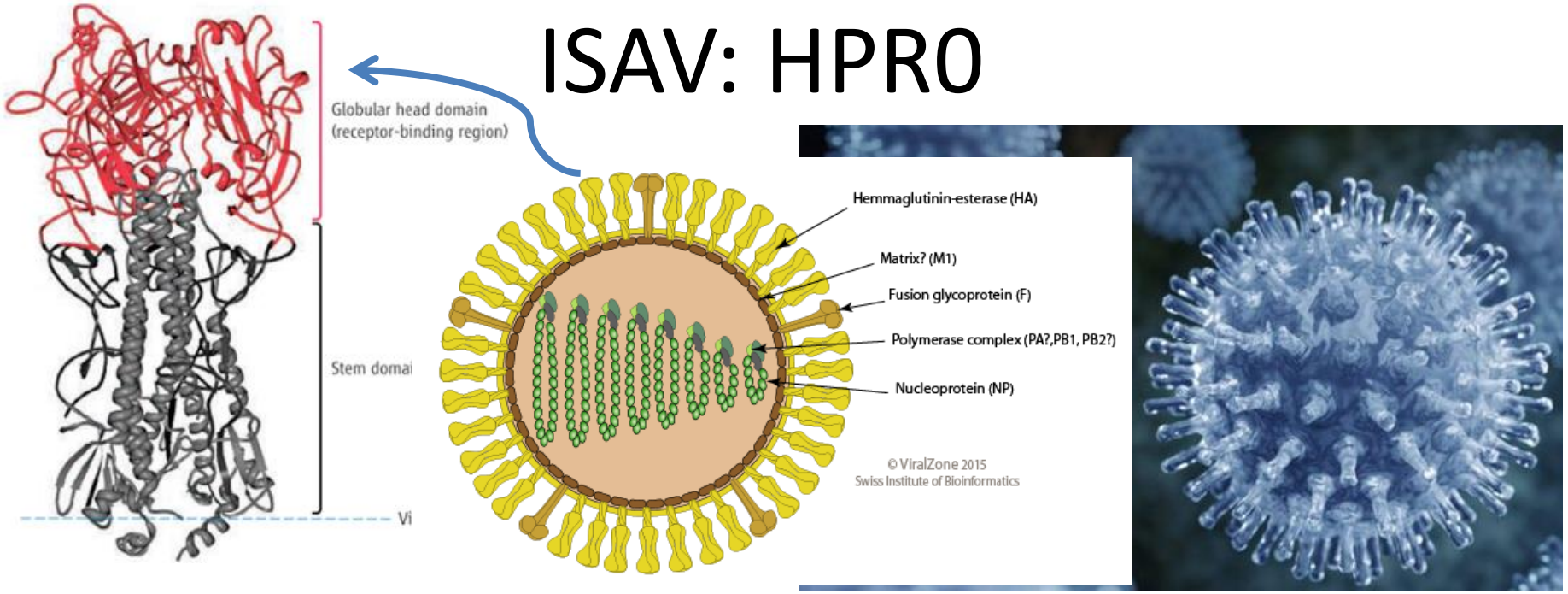


Sialic acid (a sugar) = receptor (host cell selectivity)
Hemagglutinin mediates the receptor-binding
*ISAV esterase => (allows virus escape)
Fusion protein = viral-host endosomal membrane fusion needed for entry

ISAV Hyper Variable Region (HPR)



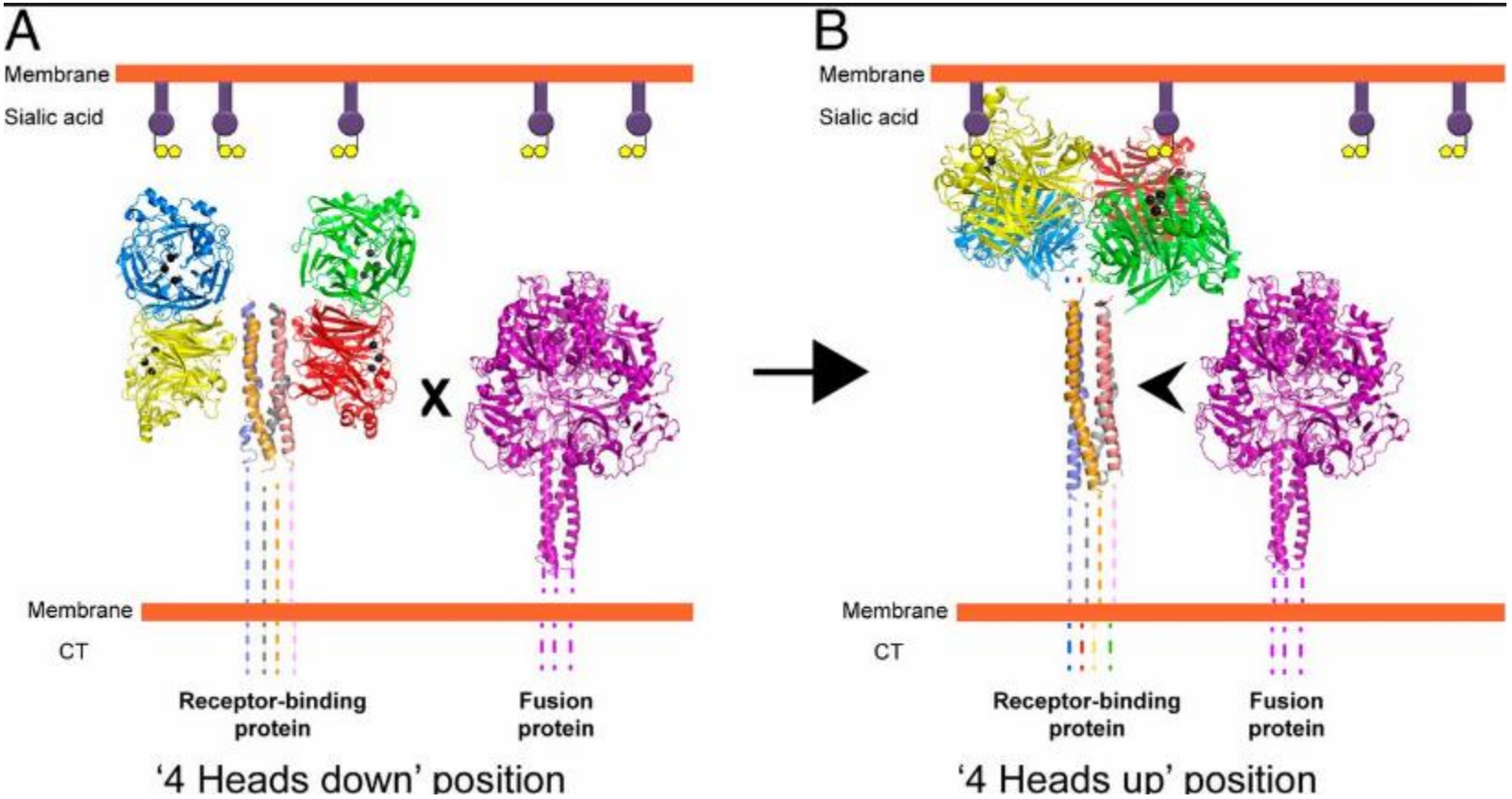
ISAV: HPR0



HPR0= non-virulent.
Infectious (can spread rapidly in a population).
Mainly detected in gills (non-systemic infections); ~10 times less detection in kidney tissues*
No symptoms
This virus binds to fish cell but can't get in (mostly).

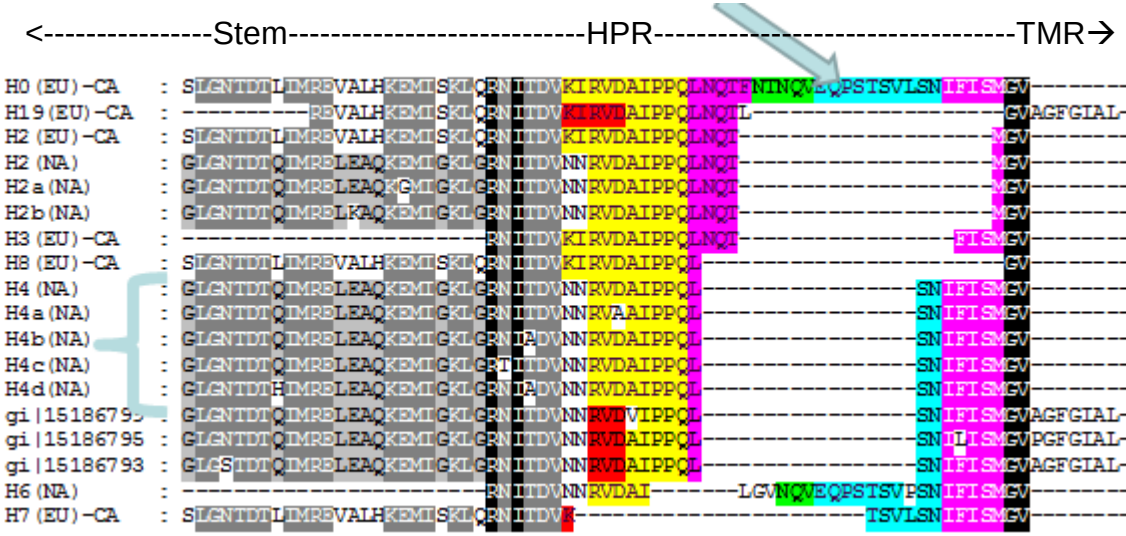
Christiansen et al 2011 "A low-pathogenic variant of infectious salmon anemia virus (ISAV-HPR0)"

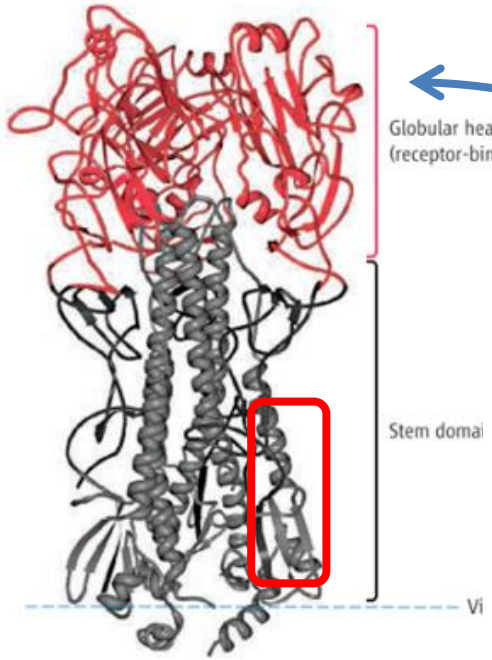
ISAV fusion with host cell

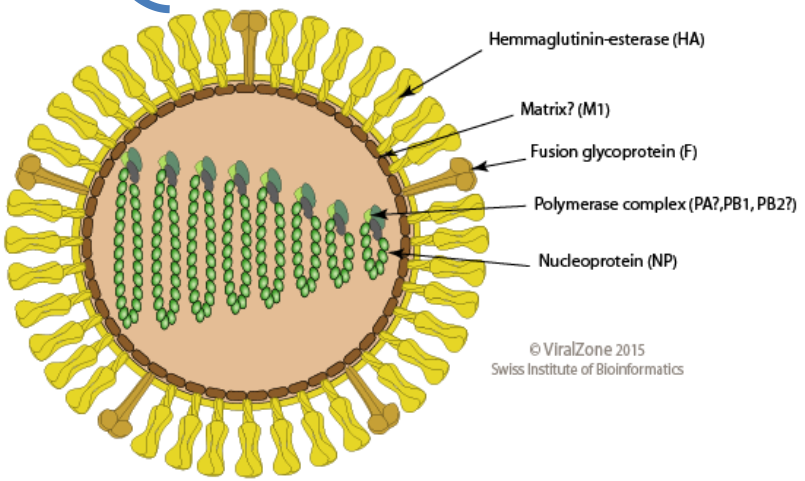


ISAV: HPRΔ

1st Condition to observe virulence:
Deletion in the hypervariable region (HPR) of the hemagglutinin= HPR Δ.
(HPR = 35 aa stretch near the membrane domain TMR)



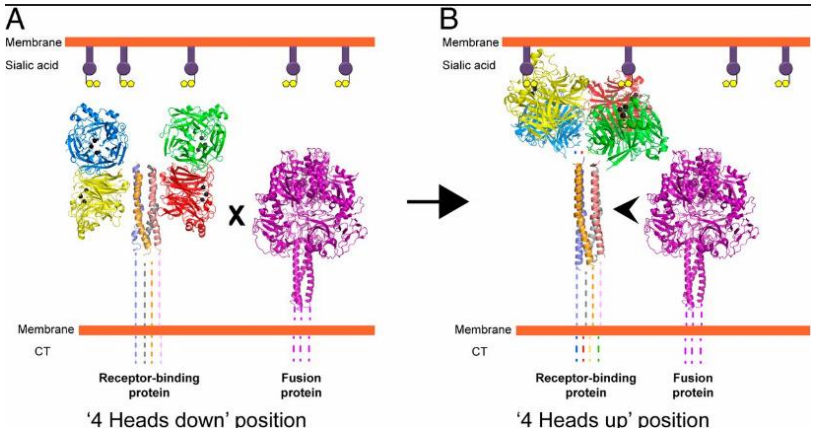


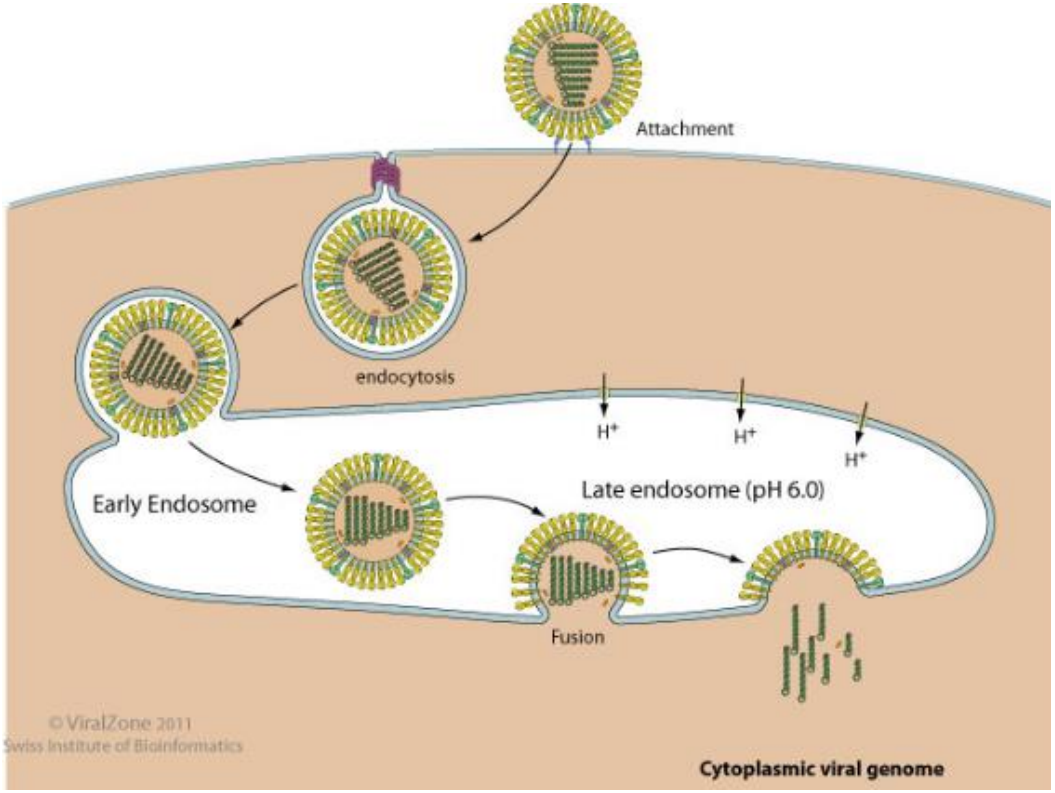


ISAV: HPRΔ

2nd Condition to observe virulence:

Fusion protein (F0) needs to be cleaved into F1/F2 by cellular protease. All virulent strain have either a Q266→L266 substitution or an insertion



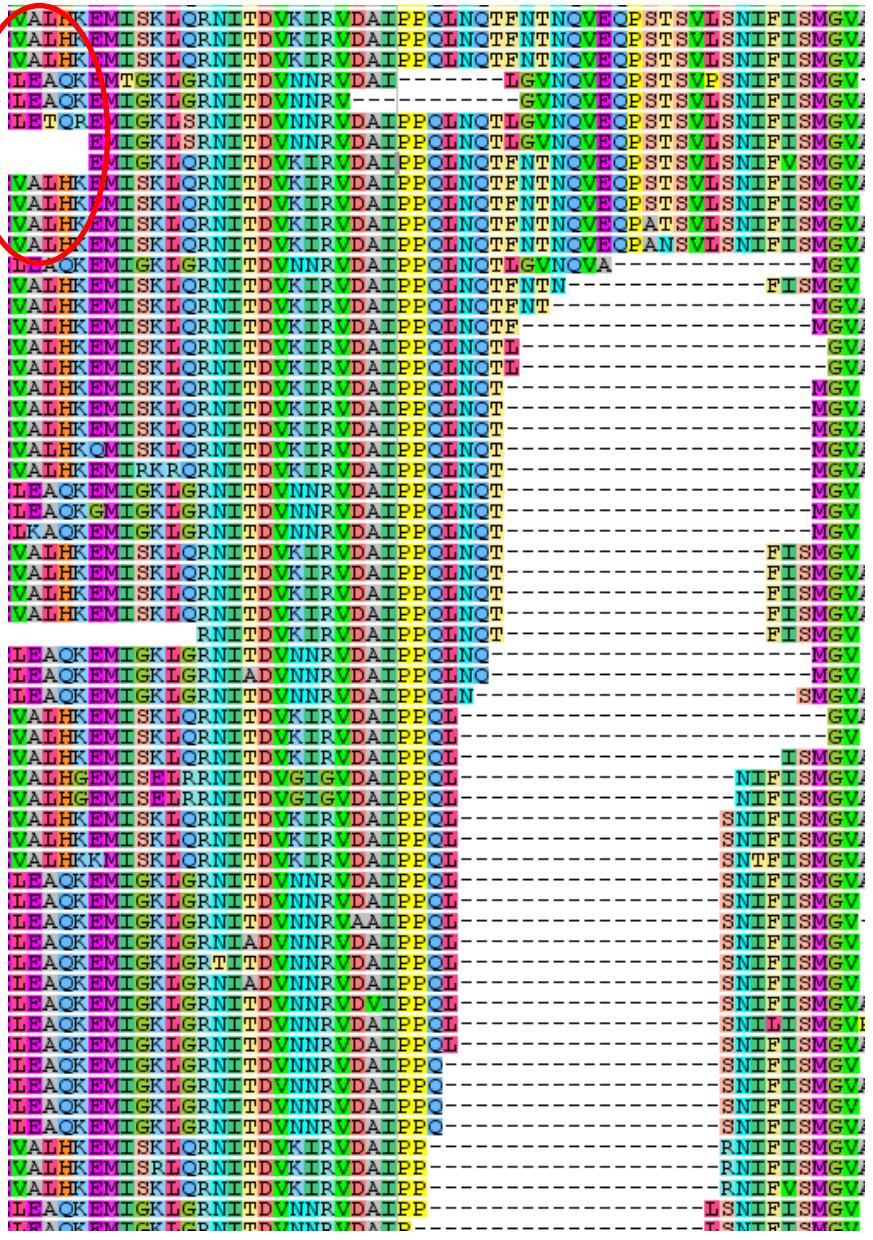


ISAV: North American vs European

Two separate introductions of ISA:

(1) Older – enough separate evolution and a clear difference between ISAV from EU and NA

(2) Recent – looks more like EU strains



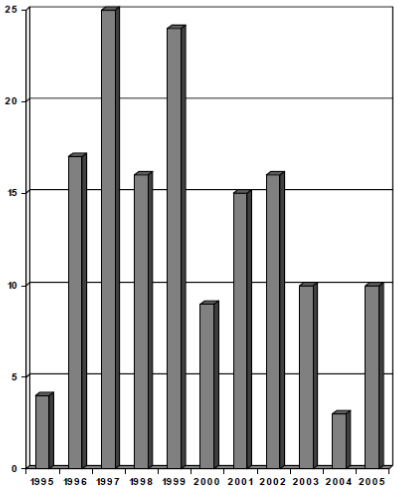
Atlantic Canada history with ISAV

ISAV affects farmed Atlantic salmon mainly in Norway (1984), Canada (1996), Scotland (1998), Chile (2007)

1996: first in NB, Canada, mostly NA HPR4
1998: first detection of EU-HPR0
Outbreaks up to 2007, then only EU-HPR0

Since 2012, recrudescence, NL, NS, NB affected, 2 -4 outbreaks per year, HPR0 continually reported
Newer HPRΔ strain are low pathogenic

Figure 1. Number of Farms Infected with ISA by Year Class in New Brunswick, Canada (updated August 2006). Data from a slide presentation by NBDAFA



Cases observed since 2012

60 ISAV notifications have been investigated and confirmed by the NAAHP (as of July 2016)

ISAV-HPRΔ strains =17 times*

- 14 ISAV NA-HPRΔ strains
- 3 ISAV EU-HPRΔ strains (in 2016).

**12 unique HPR variants*

ISAV-HPR0 strains =43 times

- 39 ISAV EU-HPR0
- 4 ISAV NA-HPR0

Cases observed since 2012

2012: 2 outbreaks in NS, 2 outbreaks in NL, all unrelated strains
- in controlled challenges, the NS isolates and one of the NL isolate showed moderate mortalities at up to 50%.
- HPR0 NA was found for the 1st time (ancestor strain of NA virulent)

2013: 4 outbreaks in NL related to previous ones (horizontal transfer), and more HPR0 EU findings. One more HPR0 NA found.
2014: only HPR0's
2015: 2 outbreaks in NB, unrelated. HPR0 EU still regularly detected.
2016: Outbreaks in NB only, some horizontal transmission of low virulent ISA HPRΔ EU and NA types.

Can we predict the outcome from sequencing?

The often asked question: what is the strain, what can you tell.
Answer: HPRΔ is not a predictor of virulence.

Hypothesis: low virulent strains will continue to circulate and outbreaks of more virulent strains will appear occasionally.

Original HPR4: we have not seen it again, we detect “closely” related ones with lower virulence.

Note: in controlled challenges, it is still affecting fish (~90% mortality)

More variability in the strains observed

Random HPR0 -> HPRΔ events leading to new strains; and/or reservoirs in wild population showing up on farms?

Conclusion

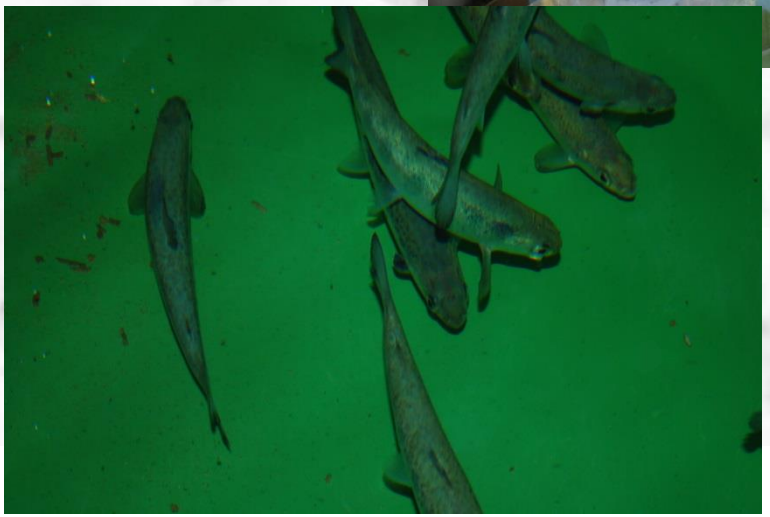
Continued surveillance is needed.

Combining increased monitoring with sequencing helps understand horizontal transmission (it does happen), but ISAV new strains show up regularly (new from HPR0 or mutant from circulating HPRΔ).

Seasonality: not a clear trend

Consider: selection for resistance; modeling for mitigation of horizontal transmission - combining with eDNA detection of ISAV in seawater...

Questions



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Canadian Food Inspection Agency




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Federal Approach to Freedom Evaluation for Reportable and Emerging Diseases

Aquatic Surveillance and Epidemiology Section

Animal Health Science Branch

Atlantic Canada Fish Farmers Association Annual Forum

St. Andrews, New Brunswick

October 27, 2016

Canada

Outline

- Brief history of ISA in BC
- Risk factors in wild fish
- Risk factors for introduction in farmed fish
- Conclusions

Brief History

- Historically, no confirmed reports of ISA in BC
- In 2011, reports of lab detections of ISA from wild BC salmon
- ISA could not be confirmed from the samples
- Wild salmon surveillance was conducted in 2012-2013
- On-farm surveillance activities were evaluated
- On farm surveillance for HPR0 carried out in 2014-2015

What are the potential pathways of ISA introduction?

- Infected wild salmonids
- Infected farmed salmonids
- Feed
- Fomites
- Vectors

Are Pacific Salmon Susceptible to ISA?

- No clinical occurrences, no confirmed detections
- Lab challenge studies on chinook, chum, coho, steelhead
  - Mortality occurs only in the first days after being injected with the virus
  - No clinical signs of ISA seen
  - Virus detectable at the end of the experiment in a few fish

Rainbow Trout and Brown Trout

- Both species present in BC
- Do not show clinical signs of ISA
- In lab studies the virus may replicate in the hosts

Wild Fish Surveillance 2012-2013

- 8006 wild salmon tested for ISAV using RT-PCR
- All species and age classes sampled
- No clinical occurrences, no confirmed detections
- Lab challenge studies on chinook, chum, coho, steelhead

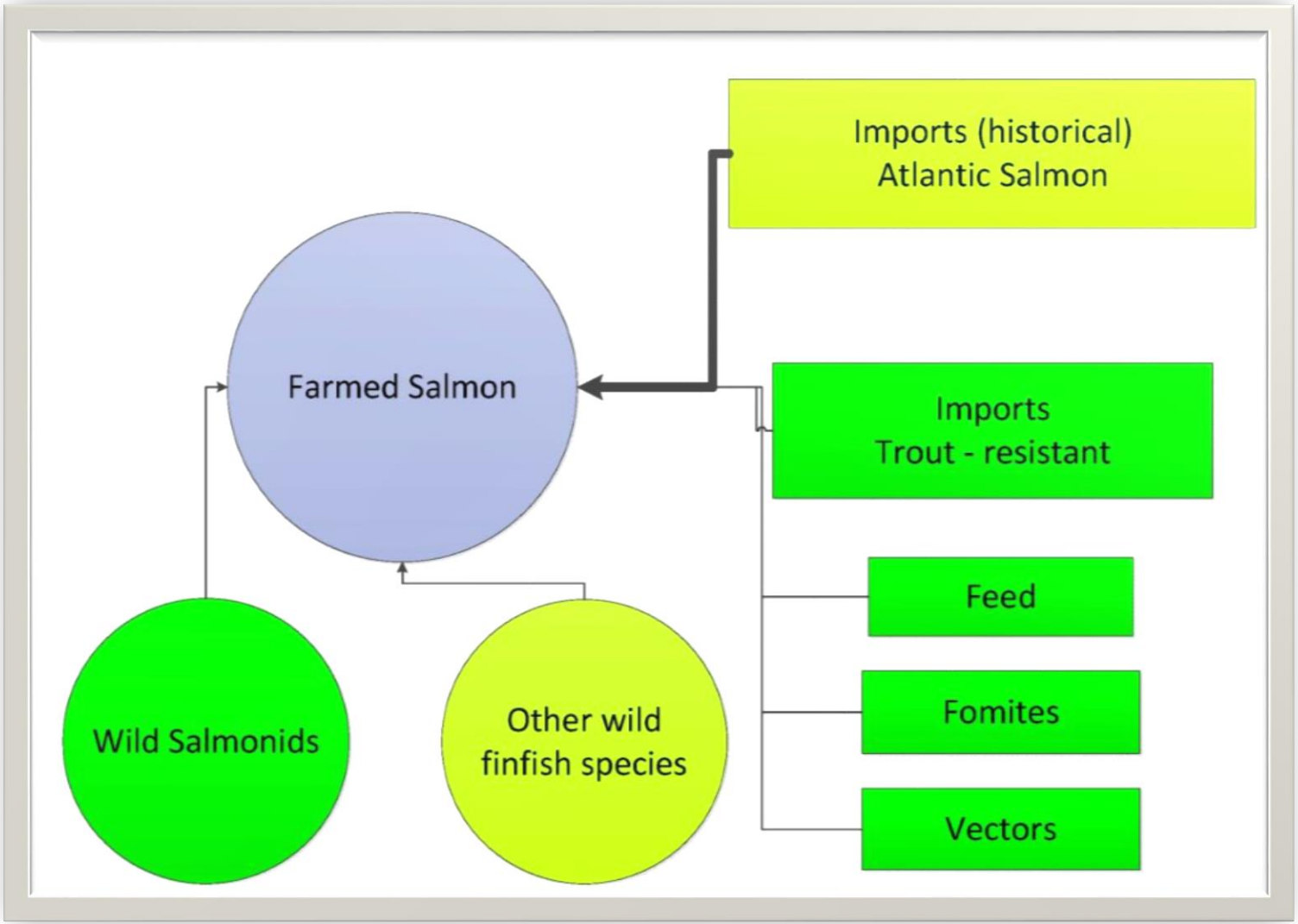
Summary – Pacific Salmon

- No confirmed historical reports
- No detections
- Lab challenges indicate resistance to infection
- No known infection in coho and rainbow trout

Evaluation of Existing Surveillance in Farmed Salmon

- Introduction Risk evaluation
- Diagnostic Testing
  - CFIA
  - Government audit
- Syndromic Surveillance

Introduction Risk to Farmed Population

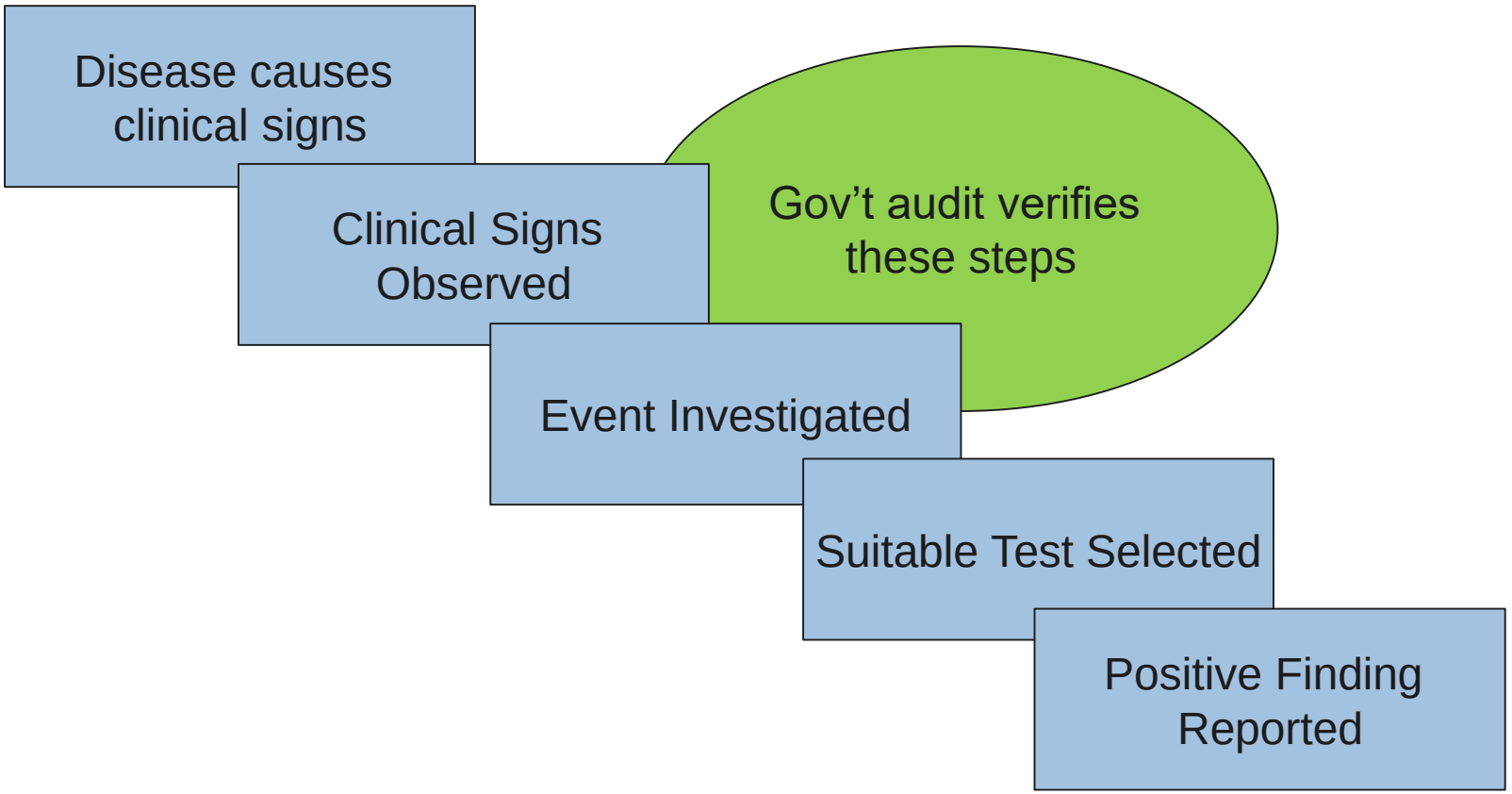


Surveillance in BC Farmed Salmon 2006-2011

Pathogen	Government Audit (2006-11)*	Industry Testing (2016-11)*	CFIA (2014-2015)
ISA HPRΔ	3183	5132	8642
ISA HPR0	3183	2789	8642

- All results negative
- *Monitoring ongoing

Syndromic Surveillance

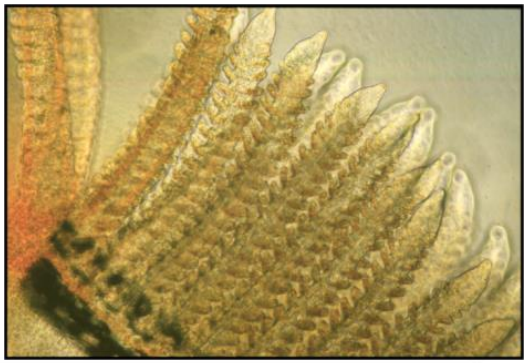


ISA HPRΔ

- No additional active surveillance required
- Syndromic surveillance increased the level of confidence
- Very confident (>99%) that the ISA seen in other parts of the world, was not present in BC farmed fish

ISAV HPR0

- Additional active surveillance required for farmed Atlantic salmon
- Confident in disease freedom (84%) but needed to achieve 95% confidence
- Syndromic surveillance would not work
- In 2014-2015, active surveillance completed
- 8462 farmed fish tested
- No suspect nor confirmed positive results



Conclusion

- The populations examined were free of ISA
- Risks of introduction of ISAV into these populations were rigorously evaluated
- If ISAV was present, it would have been detected in Atlantic salmon through the existing industry program
- Syndromic surveillance and ongoing testing maintained by existing programs was sufficient to maintain health status
- No additional active surveillance required
- Ongoing evaluations for new risks of introduction

Questions ?

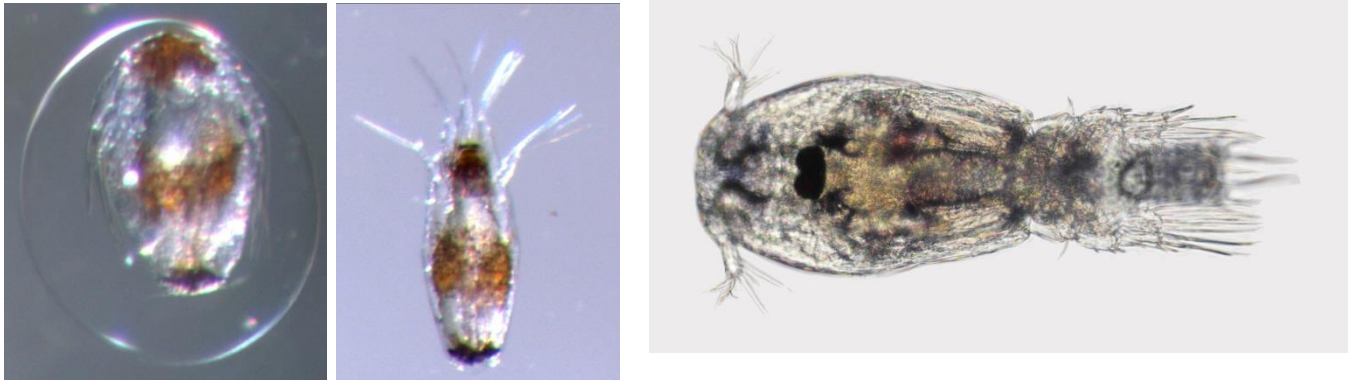
The horizontal and vertical distribution of sea lice larvae (*Lepeophtheirus salmonis*) in relation to salmon farms in the Bay of Fundy, Canada

E.J. Nelson, S.M.C Robinson, N. Feindel, A. Byrn, A. Sterling, M. Luitkus, K. Pee Ang, D. Cleaves, T.R. Lander

Canada

Context

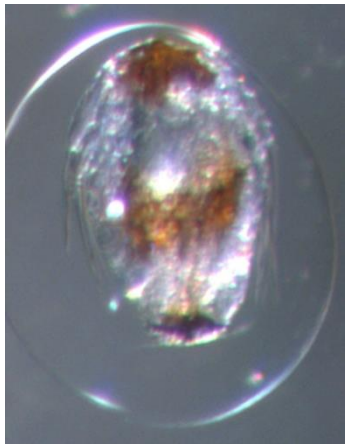
- Sea lice major problem to SW NB industry:
  - Cost approximately $75 million over last 5 years
  - Change in stocking strategies
  - Developing resistance resulting in reduced efficacy
- Early life history (larval) information needed

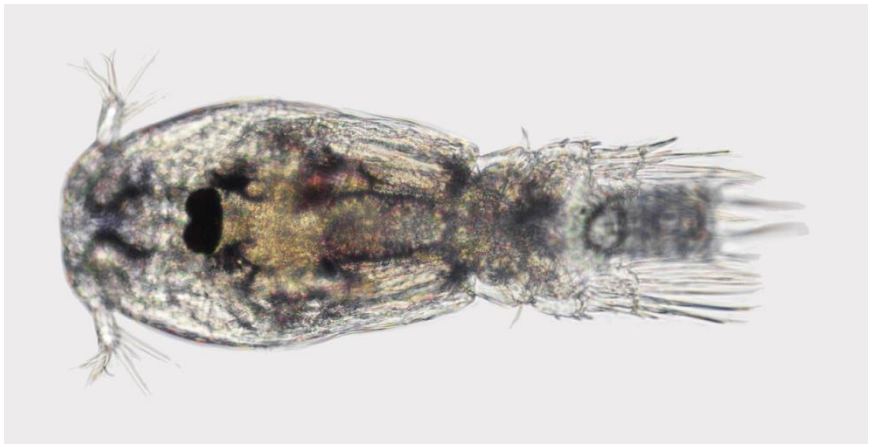


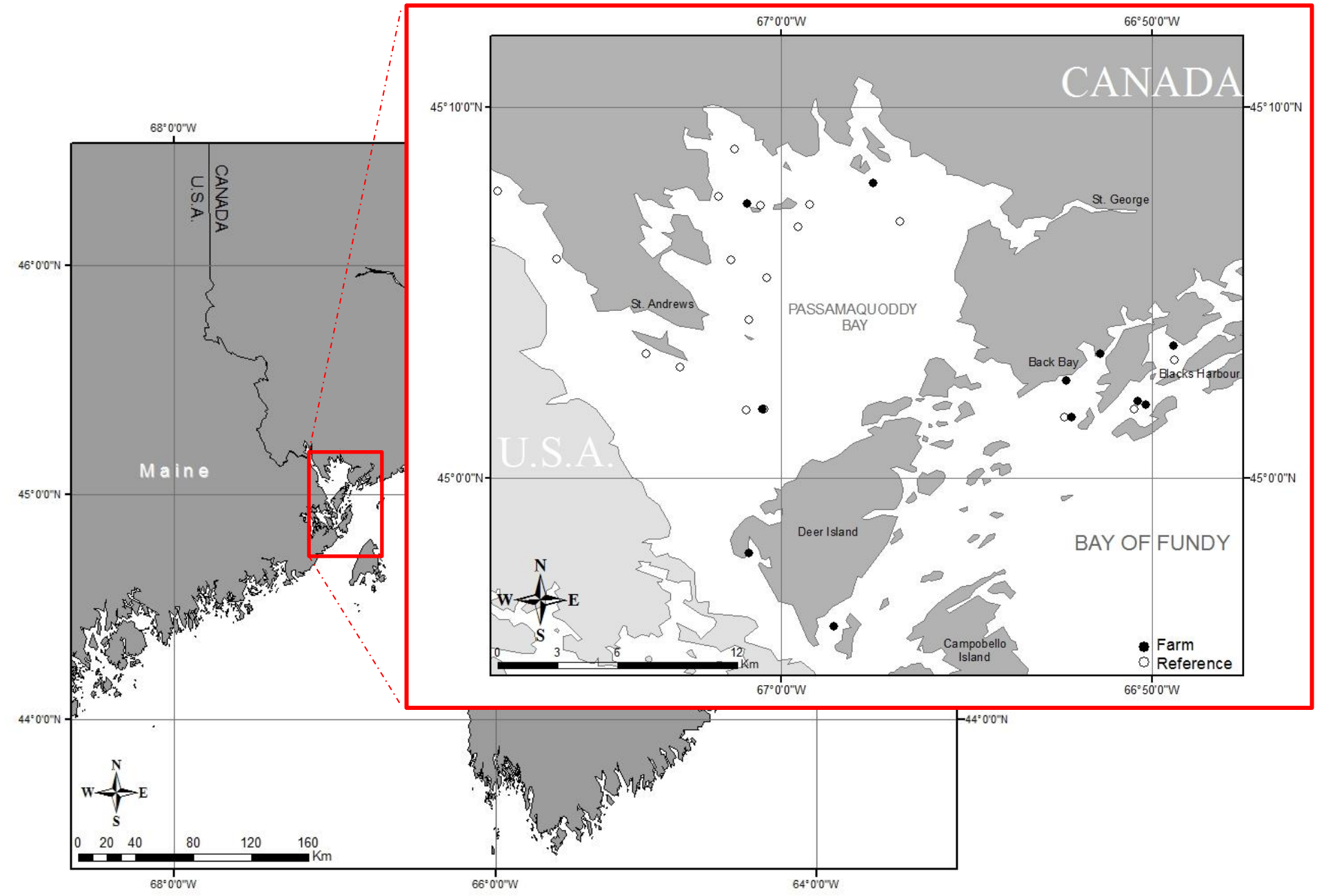
Attached chalimus stage

Context

- Our research since 2012:
  - Vertical and horizontal distribution
  - How they are staying on site / close to fish?
  - Potential to target vulnerable stages of life cycle?






68°0'0"W
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67°0'0"W
66°50'0"W
45°10'0"N
CANADA
St. George
St. Andrews
PASSAMAQUODDY
BAY
Back Bay
Blacks Harbour
U.S.A.
45°0'0"N
Deer Island
BAY OF FUNDY
N
W
E
S
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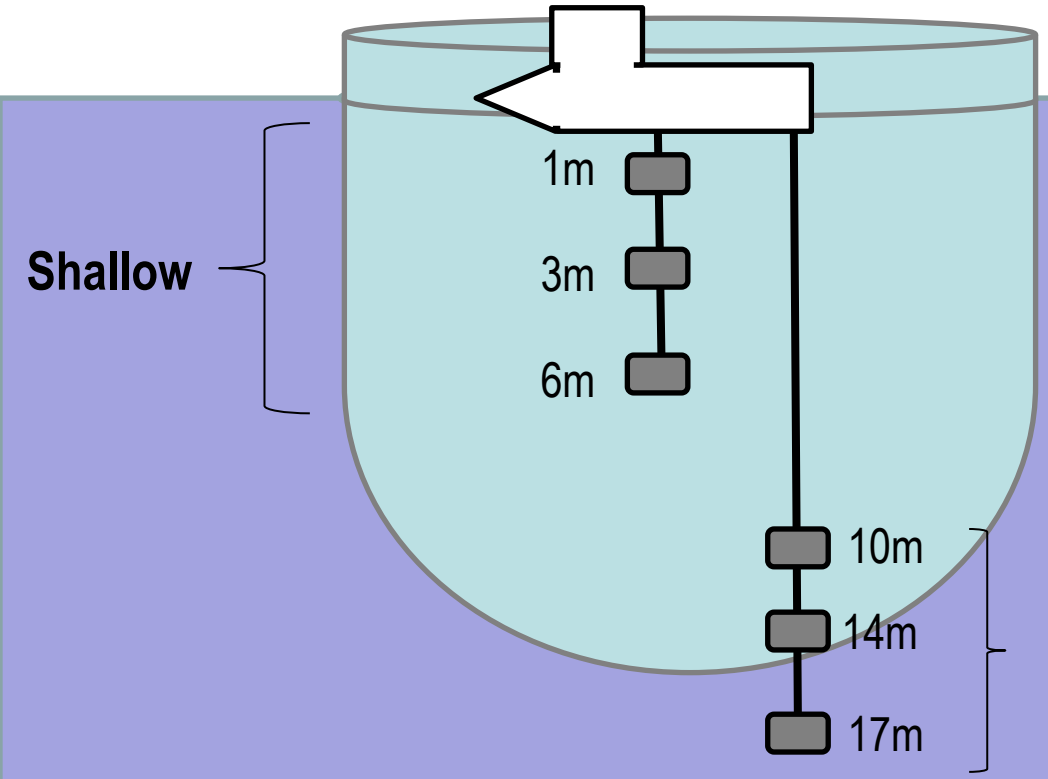
Km
Campobello
Island
Farm
Reference
67°0'0"W
66°50'0"W
45°10'0"N
45°0'0"N
44°0'0"N

Research Questions

- At what depths are larvae found (vertical distribution)?
- Do densities differ between farm and reference sites?
- Where are the larvae on site?
  - Are there areas on the farm where larvae are more prevalent?
- At what densities are the larvae leaving farm sites?
- How could the larvae stay on site and near fish?

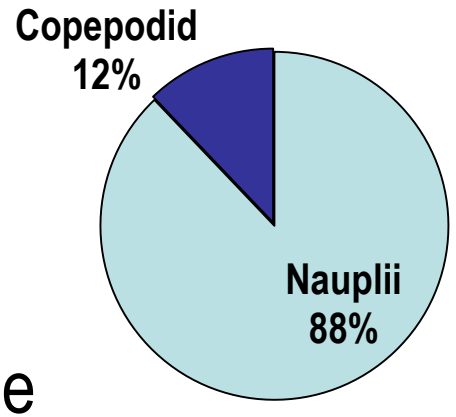
Vertical Distribution



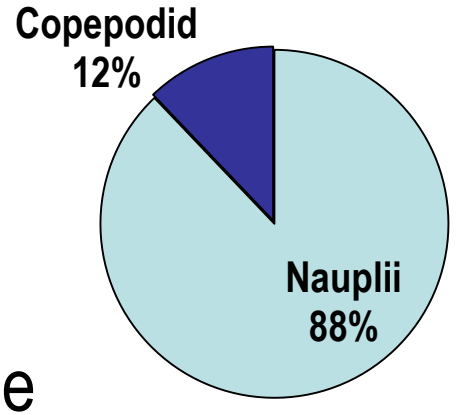
- 2 pumps
- Sampled every 90 minutes
- 2 minutes (~200L) per sample
- 3 depths (replicates of 3 each) / pump / hourly sample (N=244)

Vertical Distribution

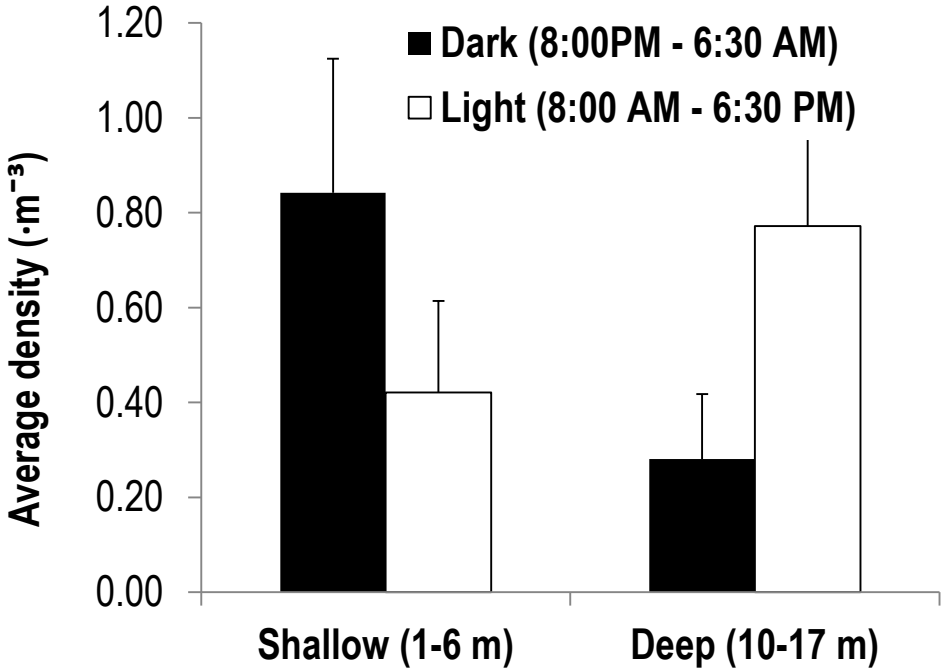
- Nauplii majority
- More or less equal densities at all depths
- When pooled shallow/deep exhibited diel cycle



Vertical Distribution



- Nauplii majority
- More or less equal densities at all depths
- When pooled shallow/deep exhibited diel cycle



- Deeper during day
- Shallow during night

Vertical Distribution

- Heuch et al. 1995
  - Copepodids in bags (shallow in day, deep in night)
  - Opposite trend, but we found nauplii majority
- In SWNB sea lice larvae found at all depths from 1-30m
- Other literature doing surface tows

- **Sea lice were found throughout water column (per m^3)**
- **If we only consider / model surface densities in SWNB we will greatly underestimate larval densities**

Farm vs. Reference Densities

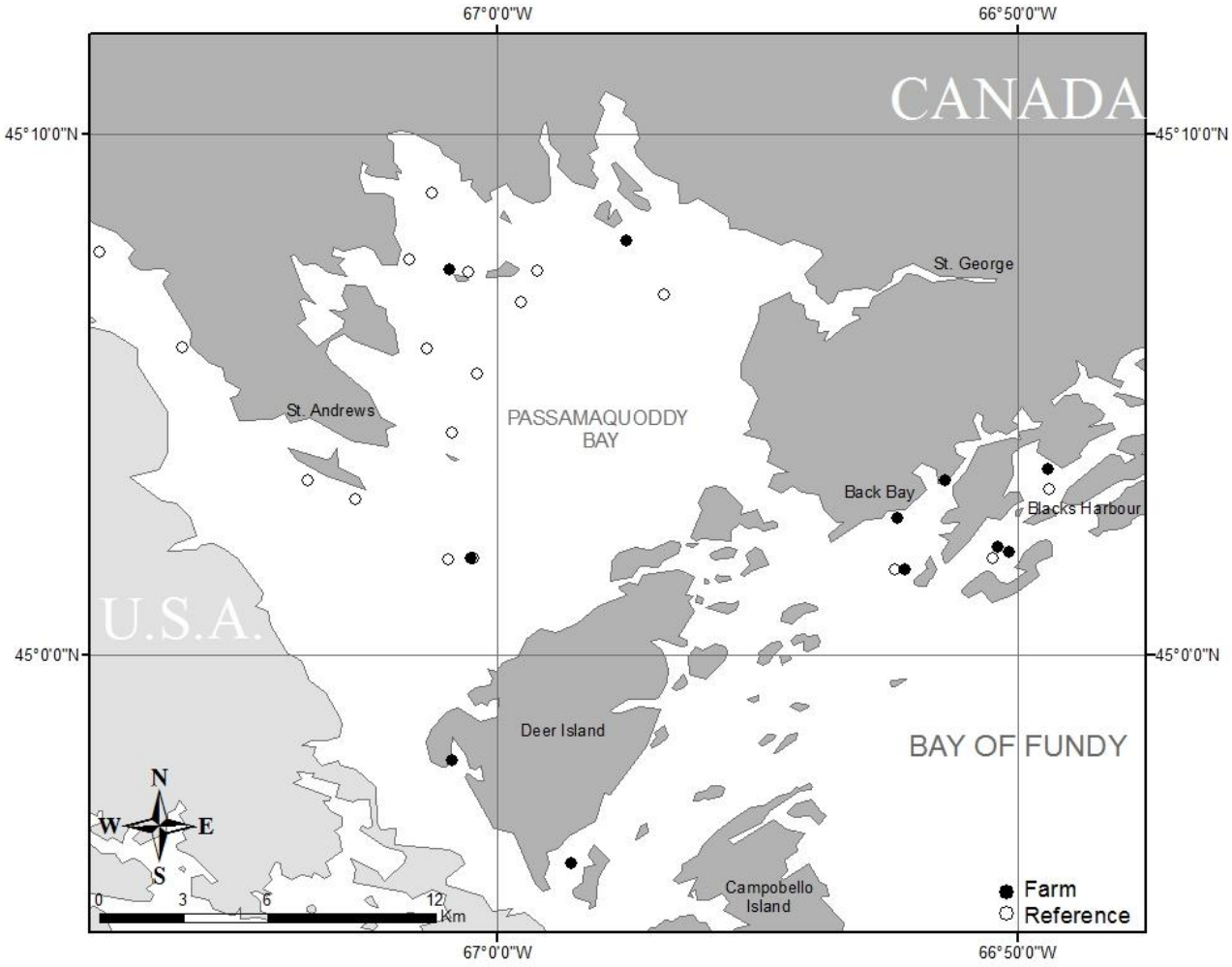
Sampling sea lice larvae in SW NB since 2012

(> 1500 samples)

11 salmon farms

17 reference sites

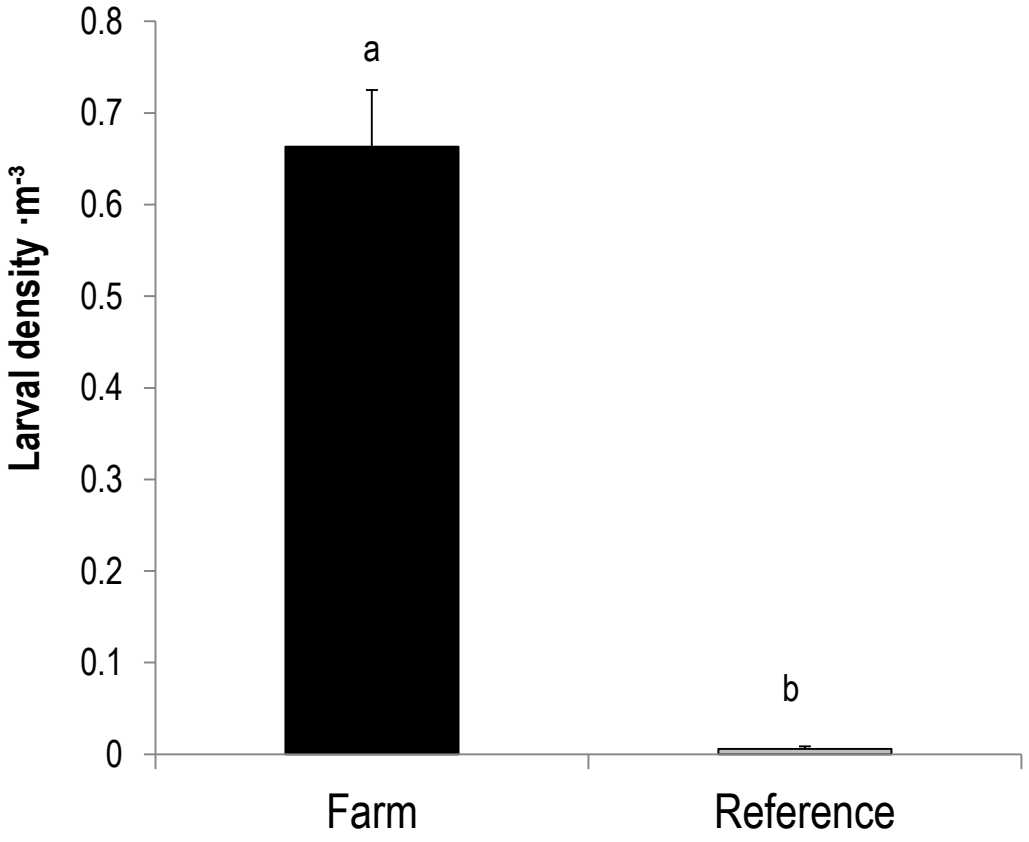
Plankton/pump samples



Farm vs. Reference Densities

Do densities differ between farm and reference sites?

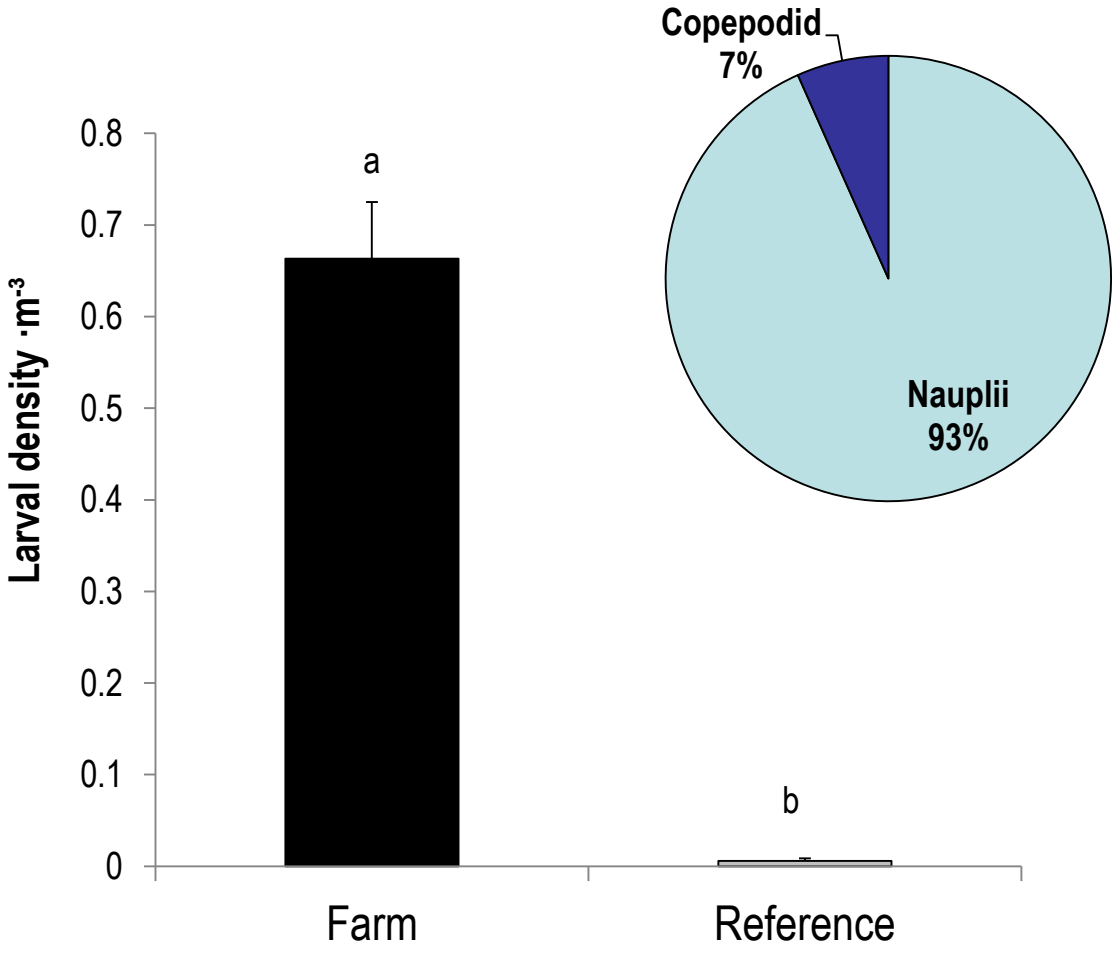
- Farm densities significantly higher
- Low densities for both (<1·m^{-3})
- Mostly nauplii



Farm vs. Reference Densities

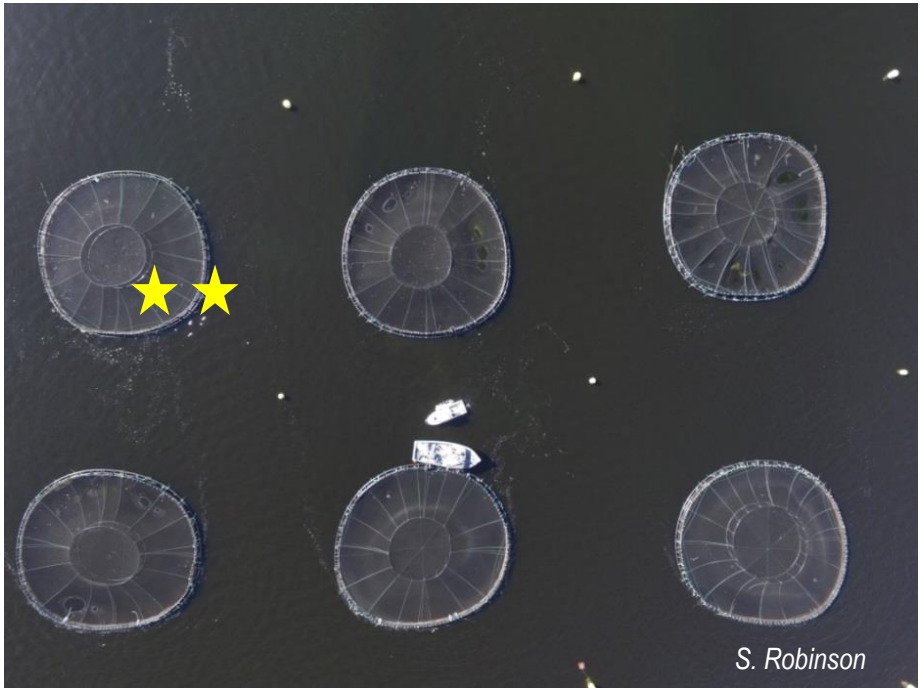
Do densities differ between farm and reference sites?

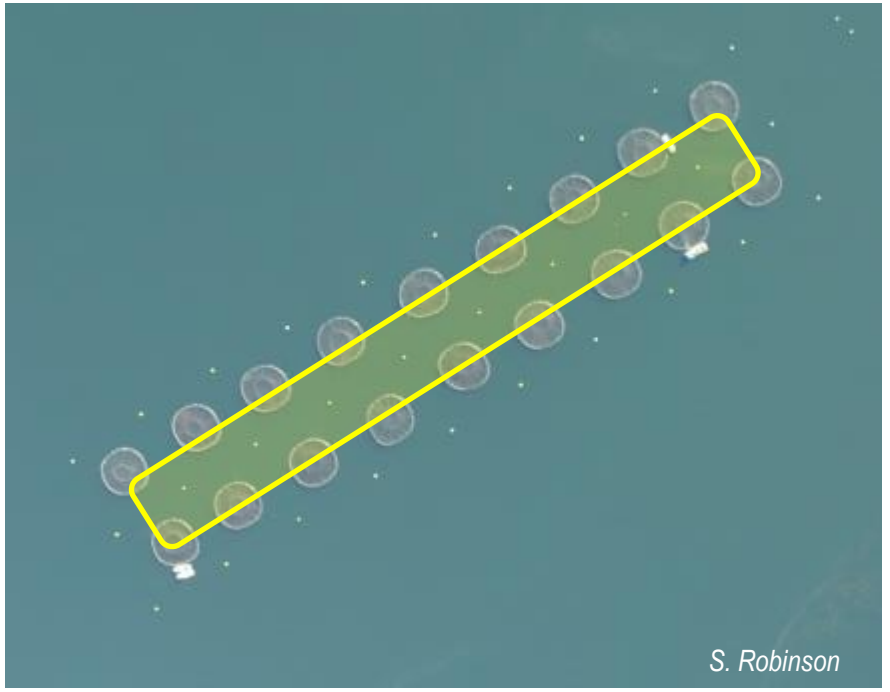
- Farm densities significantly higher ($p < 0.001$)
- Low densities for both ($<1 \cdot m^{-3}$)
- Mostly nauplii



Salmon Farm Densities (small scale)

- Where are the larvae on site? Are there high density areas?



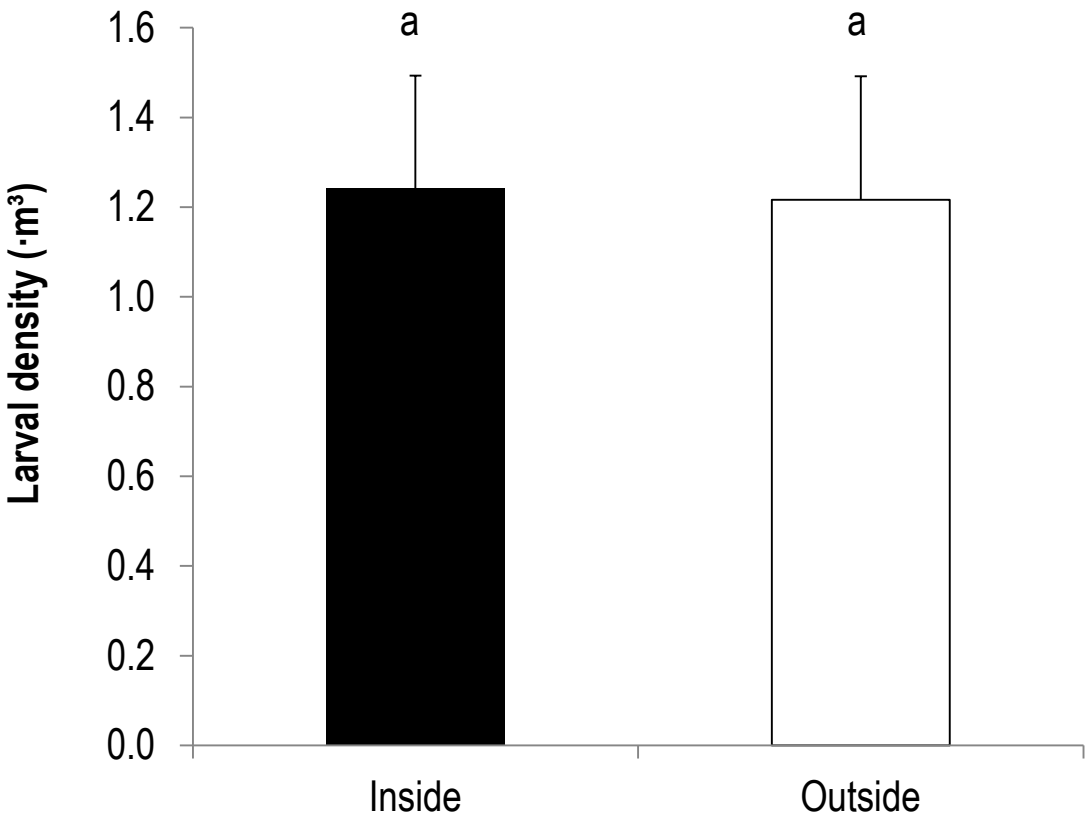


Inside cage vs. **Outside cage**
2012, 2013 and 2015
10m vertical plankton tows, 15m pump
n = 49 (paired samples)

Inner cage array vs. **Outer cage array**
2012, 2015 and 2016
15m pump samples
n = 78 each

Salmon Farm Densities (small scale)

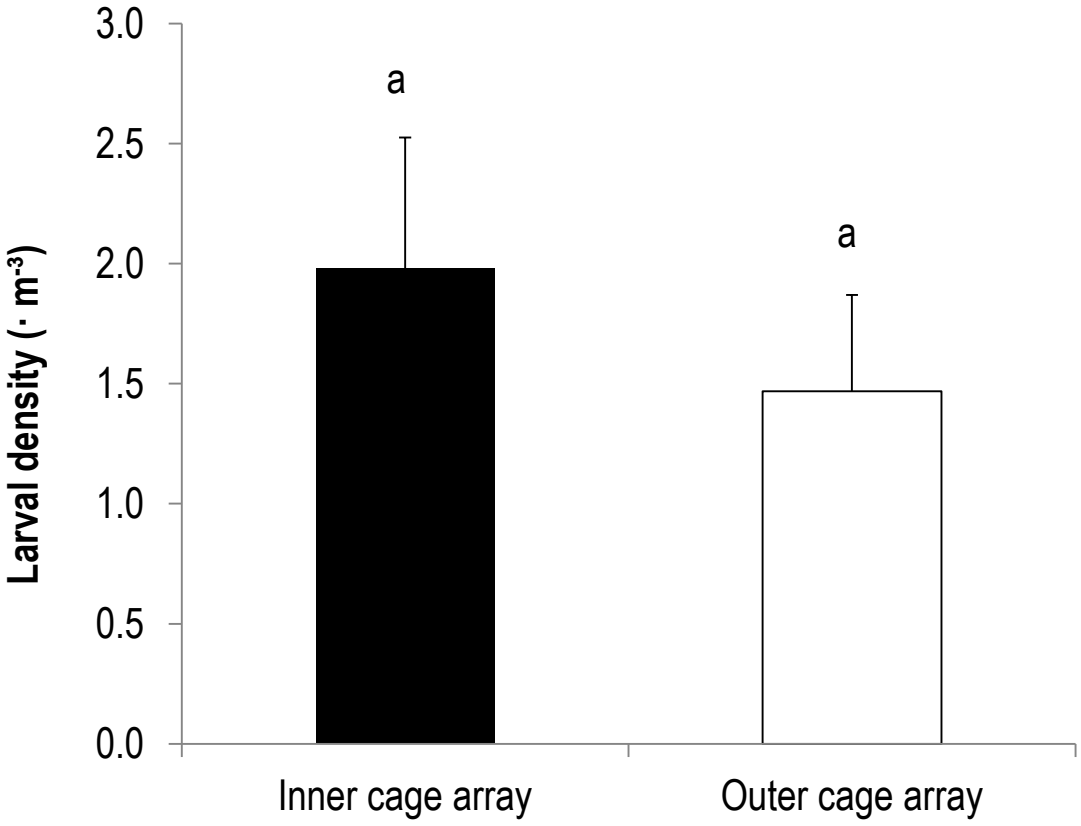
- Inside vs. Outside of cage

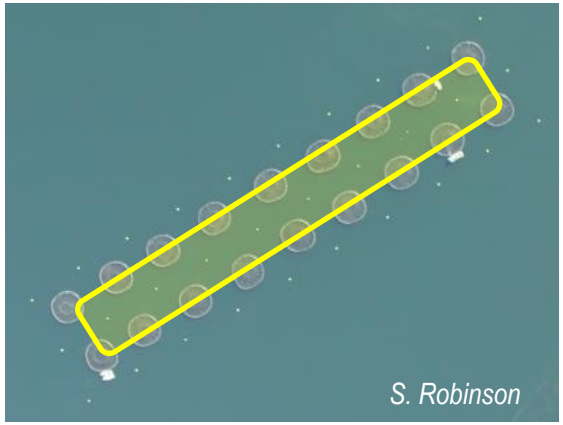


- No significant difference

Salmon Farm Densities (small scale)

- Inner vs. Outer cage array

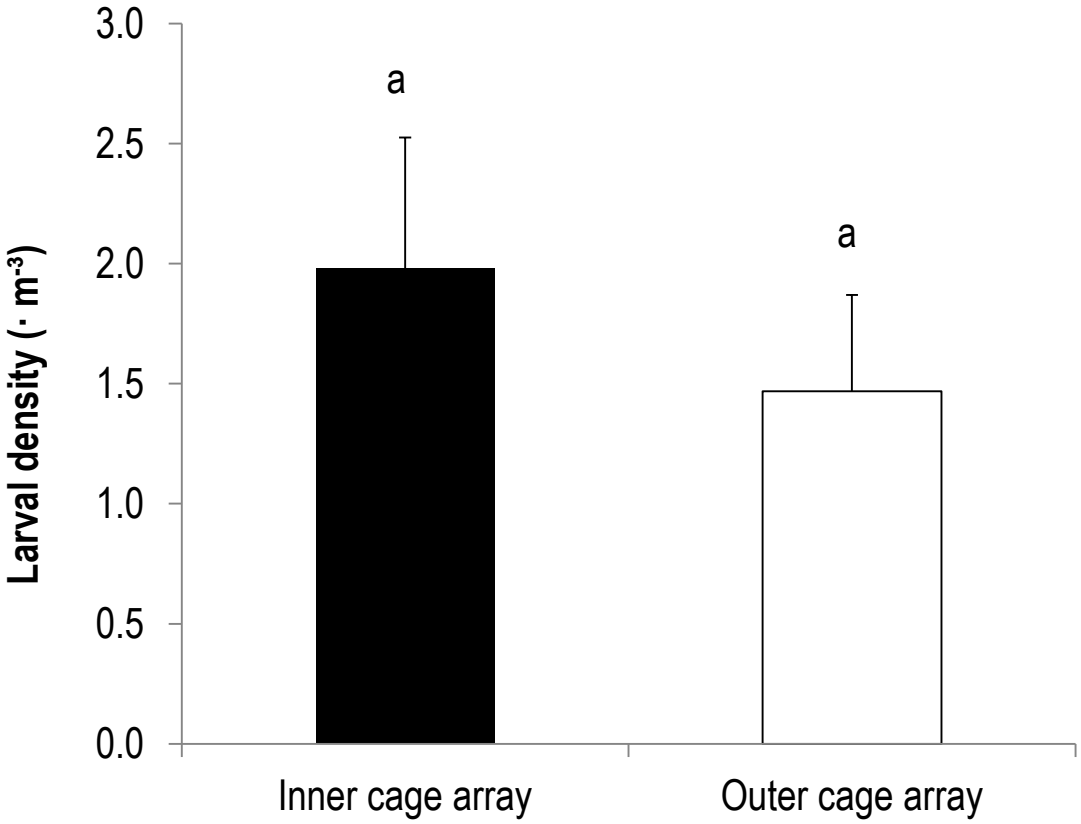


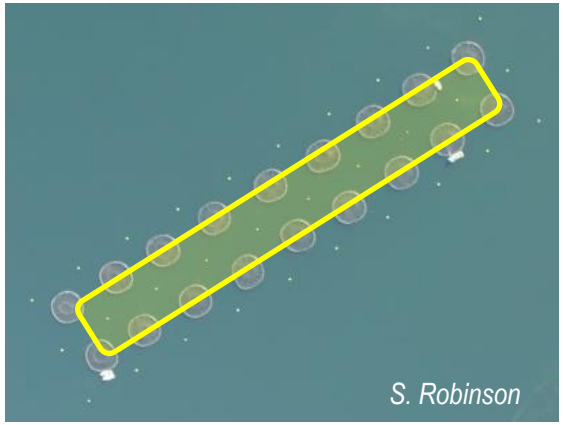


- No significant difference

Salmon Farm Densities (small scale)

- Inner vs. Outer cage array





- No significant difference

No real "hot spots" of larvae on farm site

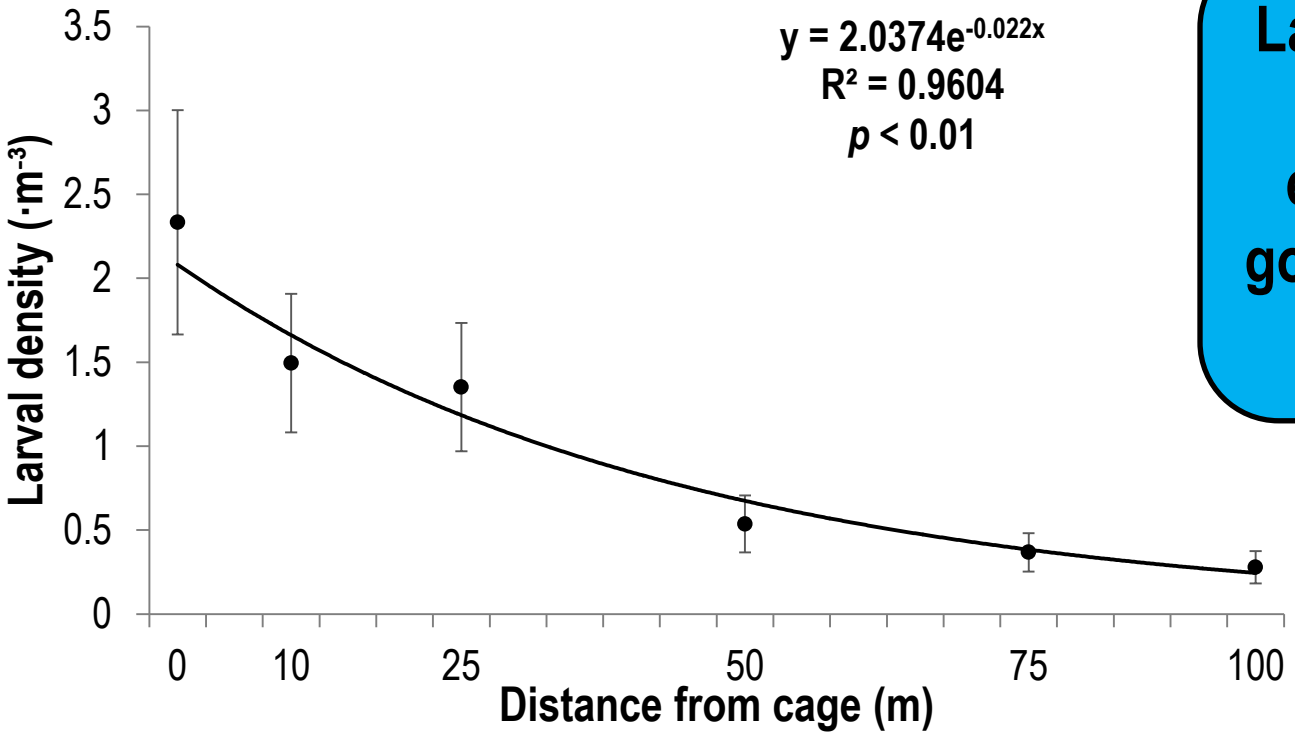
Horizontal Distribution

At what densities are the larvae leaving farm sites?

- 100 m transects in outflow direction of farm (n=3 replicates at each)

Horizontal Distribution

- Significant relationship (R=0.98) between density and distance
- Exponential decrease going away from cages



Larval densities decrease exponentially going away from cages

Main Conclusions

- Larvae found throughout water column, not just at surface in SW NB
- Mostly on farms
  - No gradients (inside/outside; inner/outer; transects)
  - No real ‘cloud’ of larvae on salmon farms
- Mostly nauplii (few copepodids)
- Compounding factors potentially on site
- Densities on nets high; close proximity to fish

Future Research / Next Steps

- Continued sampling:
  - Capture seasonal densities on farms / reference sites
  - Harvest activities
  - On-site treatments
- Paper on horizontal and vertical distribution of larvae in Bay of Fundy

Many thanks to…

Funding sources:

- DFO's Aquaculture Collaborative Research and Development Program (ACRDP)
- DFO's Program for Aquaculture Regulatory Research (PARR)

Site access:

- Cooke Aquaculture
- Grays Aquaculture

Field and Lab Assistance:

- CCGS Viola M. Davidson Captain and Crew
- Craig A. Smith, Craig G. Smith, Steve Neil, Adena Peters, Riley Walker, Karl Hanke, Paul Robertson and Rebecca Eldridge

Thank you!

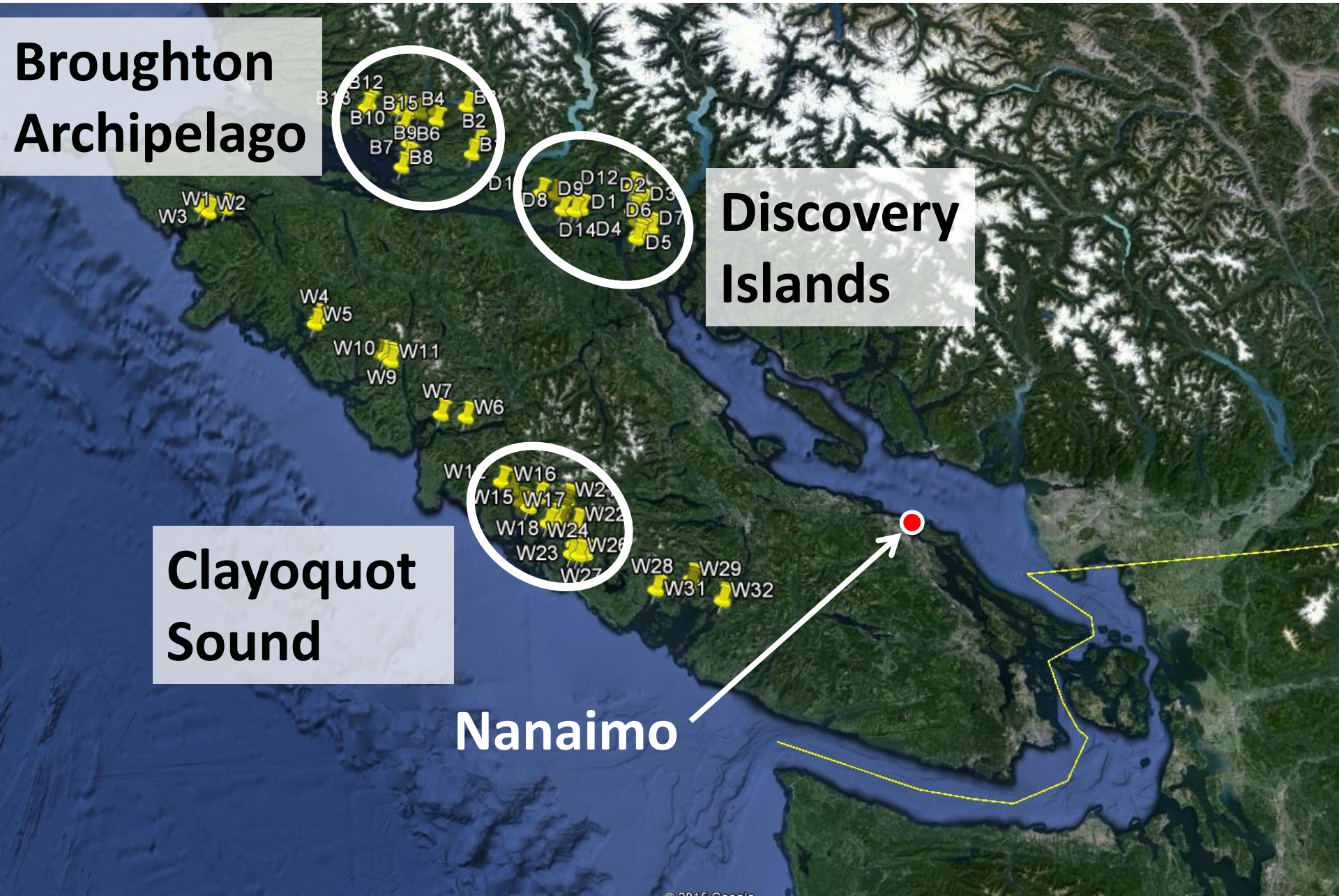
Salmon migration: a key process for understanding lice infection of wild salmon

Marc Trudel[1,*], Stewart Johnson[1], Chrys Neville[1], Simon Jones[1], and Julie Bradshaw[1]

[1]Pacific Biological Station, Fisheries and Oceans Canada, Nanaimo, BC

[*]Present address: St. Andrews Biological Station, Fisheries and Oceans Canada, St. Andrews, NB

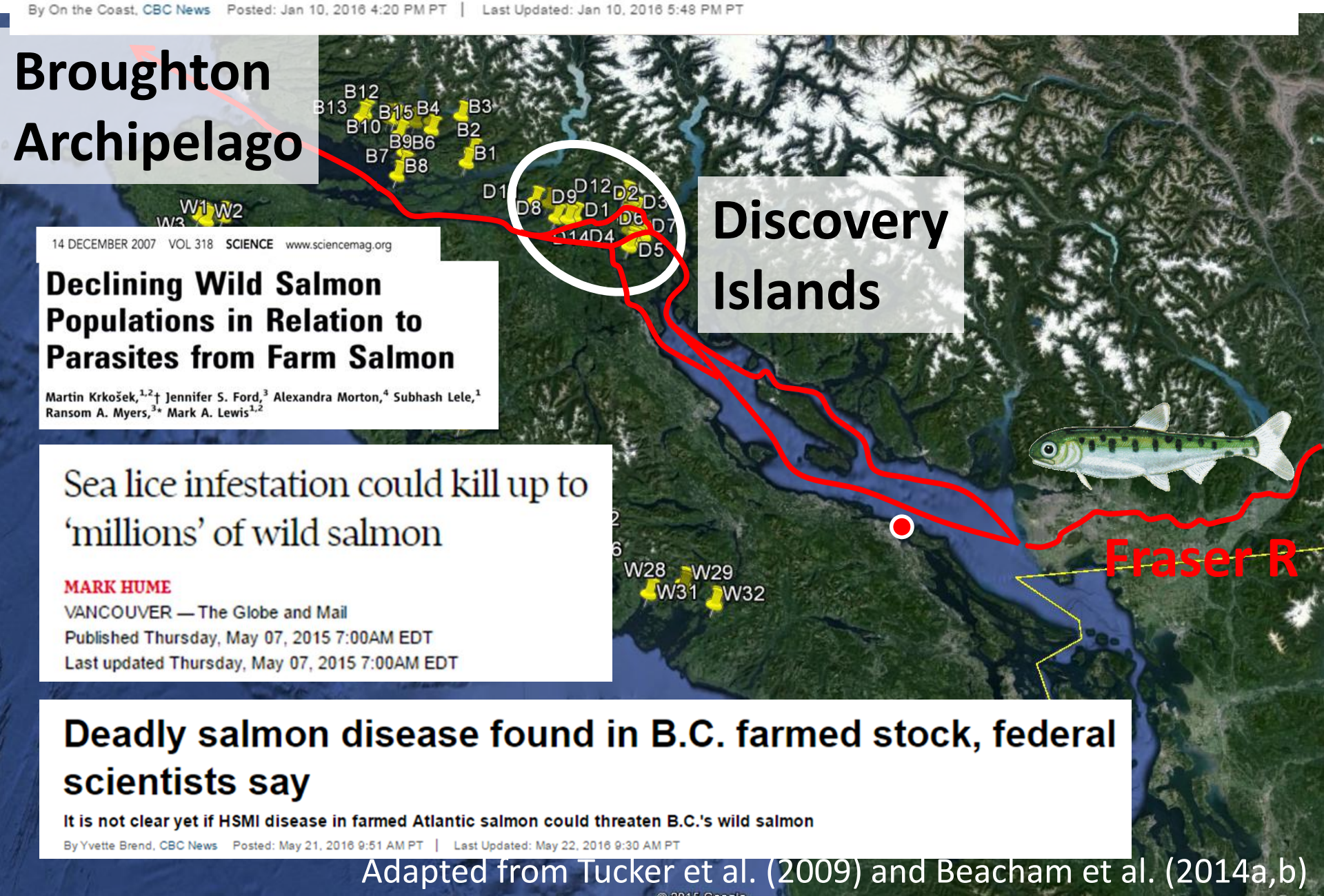
Salmon farms in Southern British Columbia



Deadly salmon virus may be in B.C. waters, study suggests

Farmed salmon industry says findings of infectious salmon anaemia are false positives

By On the Coast, CBC News Posted: Jan 10, 2016 4:20 PM PT | Last Updated: Jan 10, 2016 5:48 PM PT



14 DECEMBER 2007 VOL 318 SCIENCE www.sciencemag.org

Declining Wild Salmon Populations in Relation to Parasites from Farm Salmon

Martin Krkošek,[1,2]† Jennifer S. Ford,[3] Alexandra Morton,[4] Subhash Lele,[1] Ransom A. Myers,[3]* Mark A. Lewis[1,2]

Sea lice infestation could kill up to 'millions' of wild salmon

MARK HUME

VANCOUVER — The Globe and Mail

Published Thursday, May 07, 2015 7:00AM EDT

Last updated Thursday, May 07, 2015 7:00AM EDT

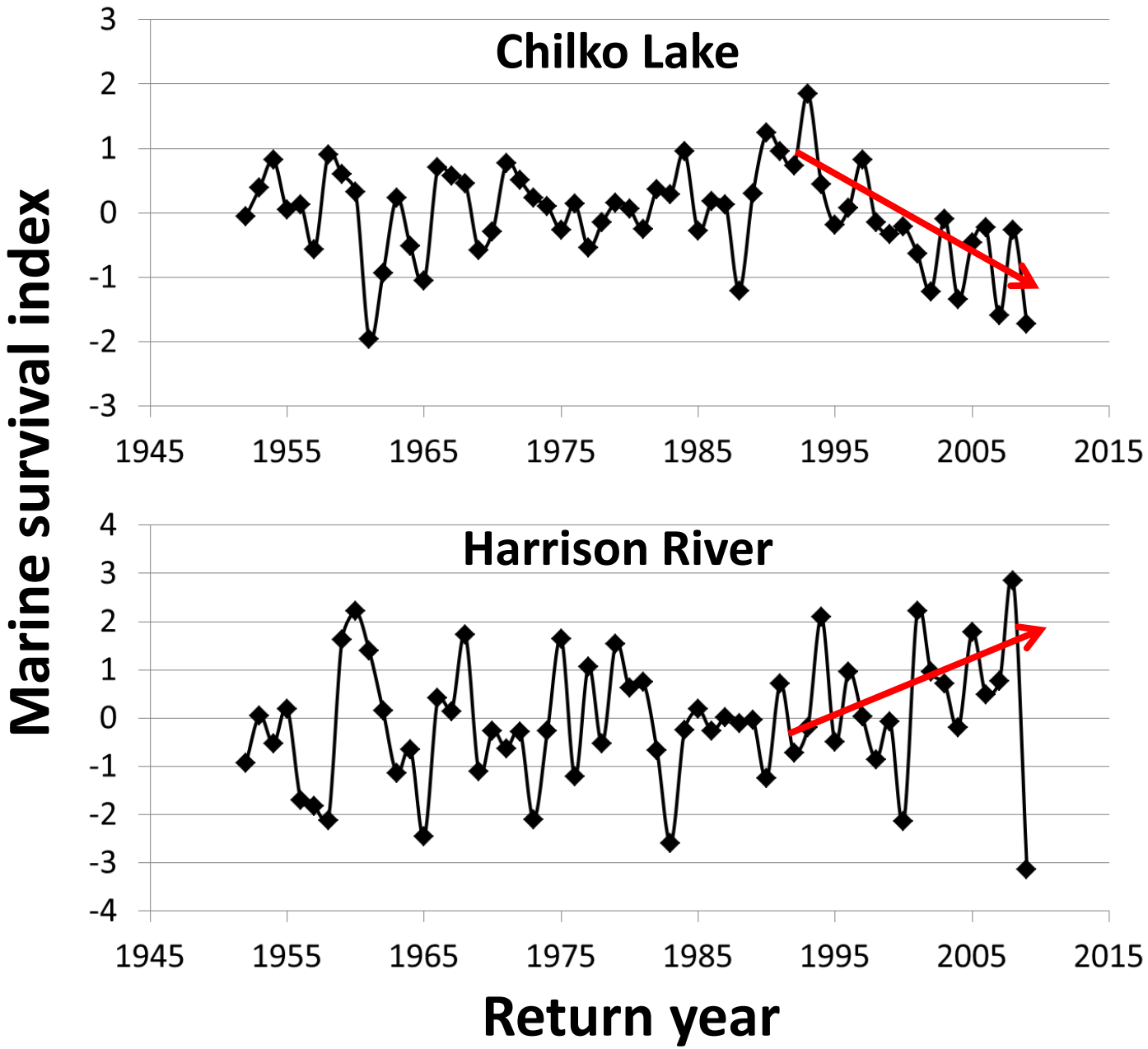
Deadly salmon disease found in B.C. farmed stock, federal scientists say

It is not clear yet if HSMI disease in farmed Atlantic salmon could threaten B.C.'s wild salmon

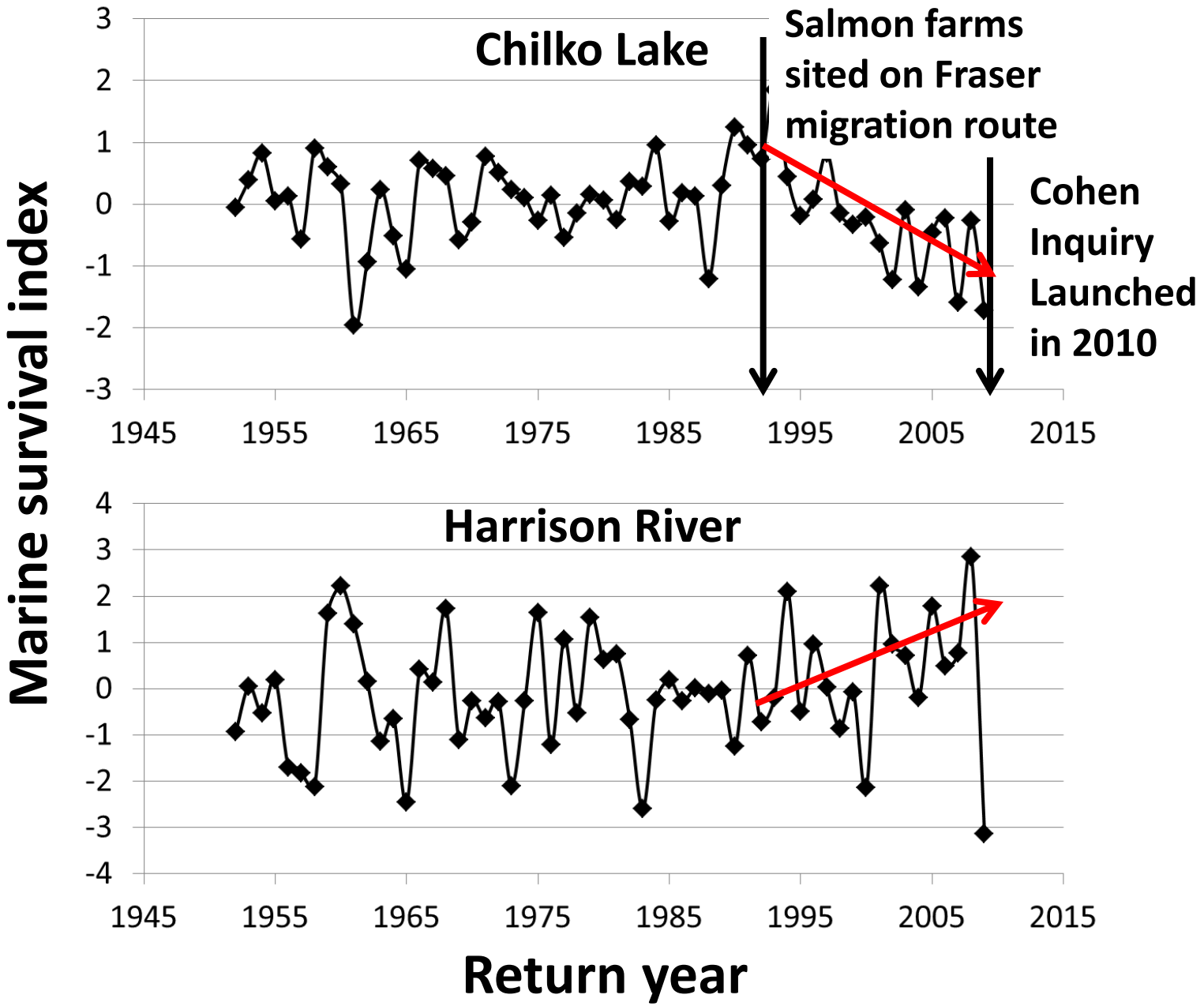
By Yvette Brend, CBC News Posted: May 21, 2016 9:51 AM PT | Last Updated: May 22, 2016 9:30 AM PT

Adapted from Tucker et al. (2009) and Beacham et al. (2014a,b)

Fraser River sockeye salmon productivity



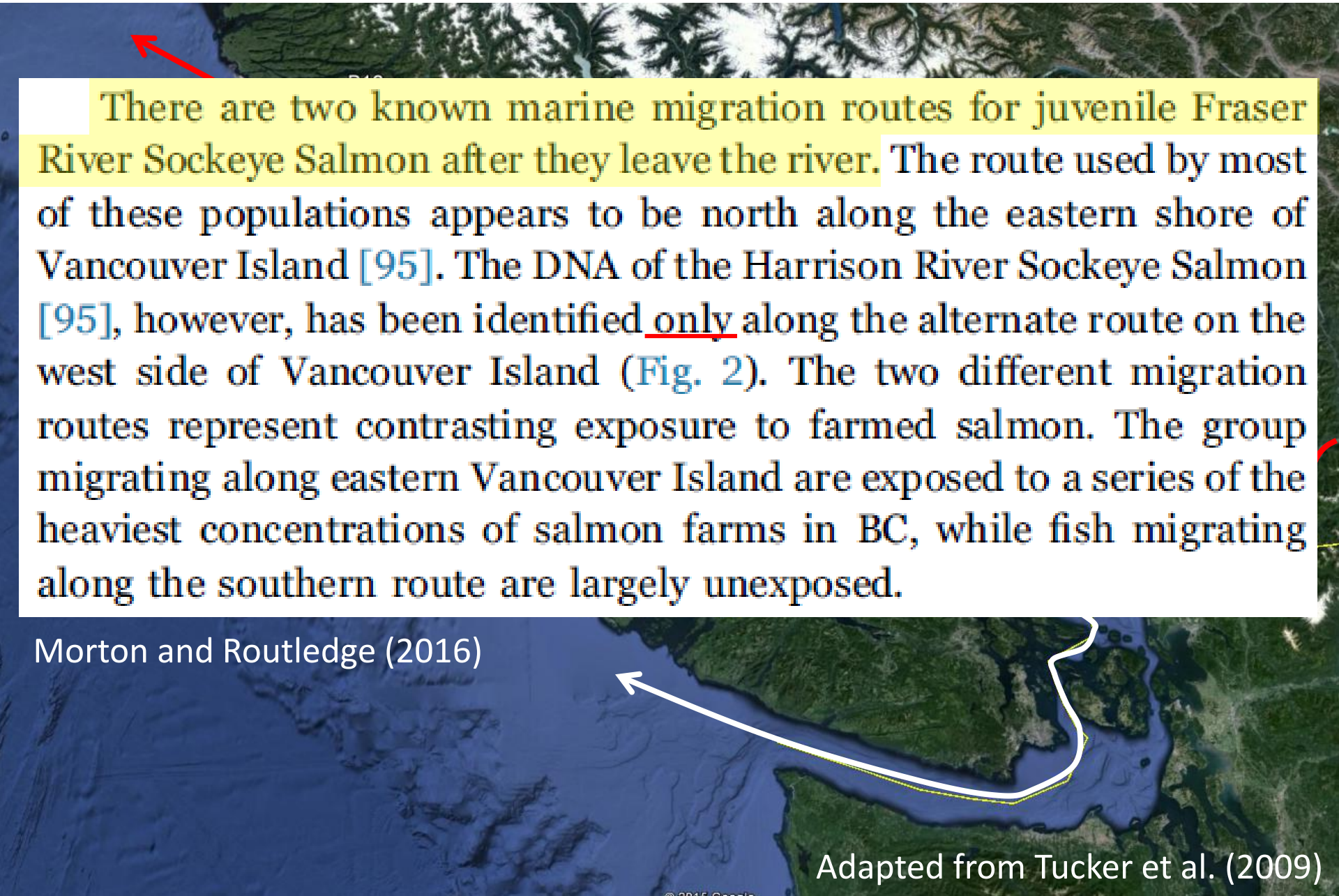
Fraser River sockeye salmon productivity



Migration route of juvenile Fraser R. sockeye

Adapted from Tucker et al. (2009)

Migration route of juvenile Fraser R. sockeye

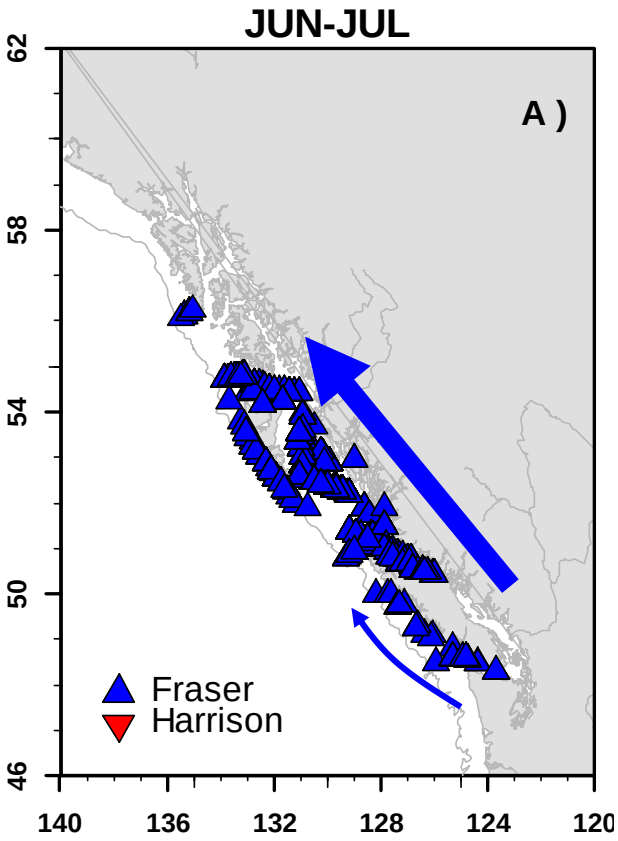


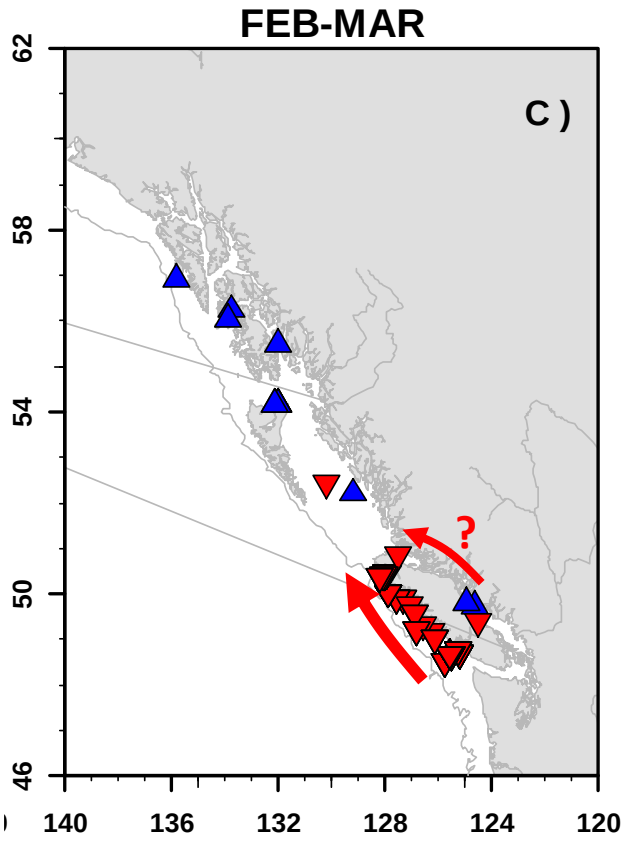
There are two known marine migration routes for juvenile Fraser River Sockeye Salmon after they leave the river. The route used by most of these populations appears to be north along the eastern shore of Vancouver Island [95]. The DNA of the Harrison River Sockeye Salmon [95], however, has been identified only along the alternate route on the west side of Vancouver Island (Fig. 2). The two different migration routes represent contrasting exposure to farmed salmon. The group migrating along eastern Vancouver Island are exposed to a series of the heaviest concentrations of salmon farms in BC, while fish migrating along the southern route are largely unexposed.

Morton and Routledge (2016)

Adapted from Tucker et al. (2009)

Juvenile Fraser River and Harrison River sockeye salmon distribution & migration





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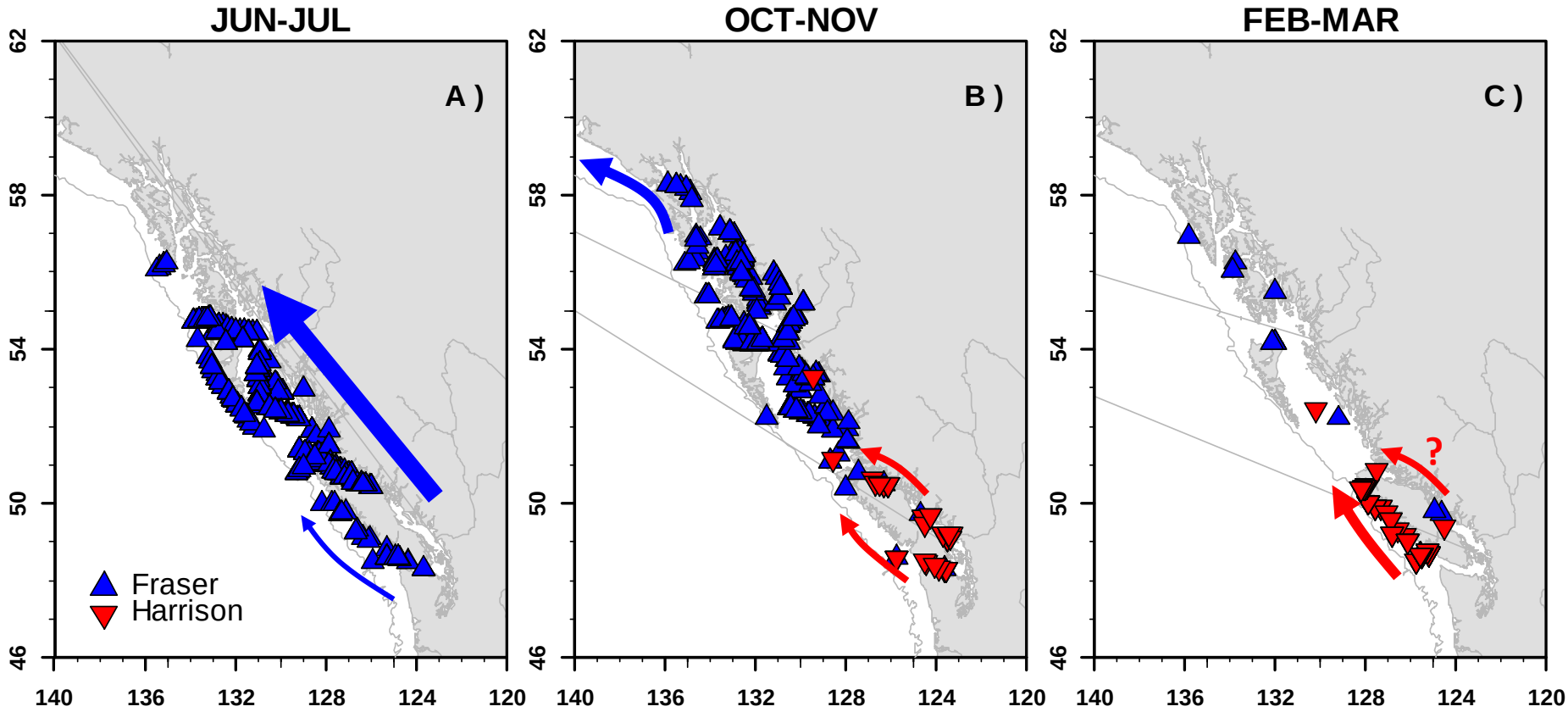
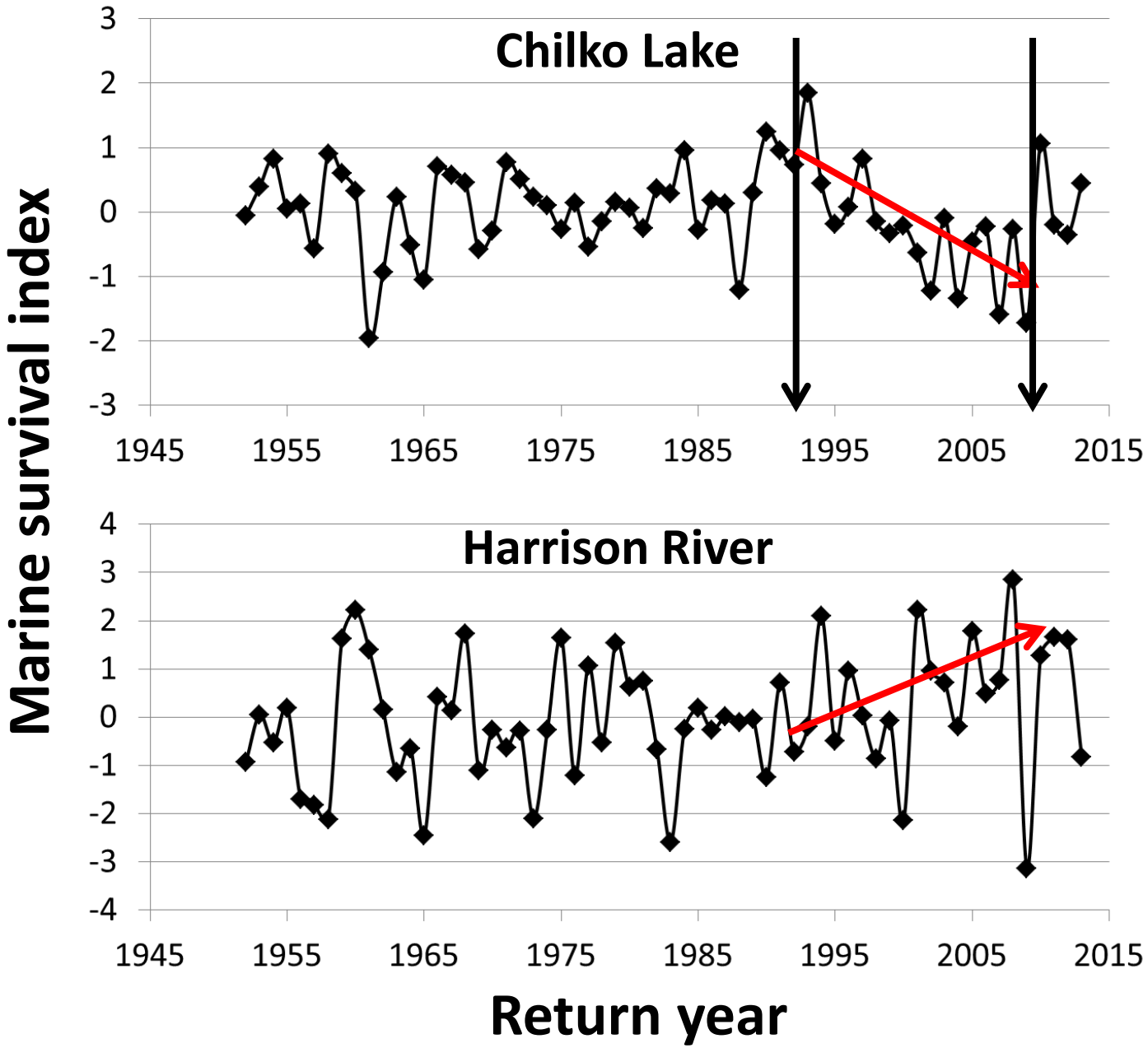


Adapted from Tucker et al. (2009), Beacham et al. (2014a, 2014b)

Are Harrison River sockeye a good control for Fraser River sockeye?

Parameter	*Fraser*	*Harrison River*
Smolt size	80-120 mm	50-60 mm
Peak migration	late April – early May	late June – early July
Residence time in the Strait of Georgia	early April – early July	mid June – February/March

Fraser River sockeye salmon productivity



North American Journal of Fisheries Management 28:523–532, 2008

DOI: 10.1577/M07-042.1

[Article]

Sea Louse Infestation in Wild Juvenile Salmon and Pacific Herring Associated with Fish Farms off the East-Central Coast of Vancouver Island, British Columbia

ALEXANDRA MORTON
Raincoast Research Society, Simoom Sound, British Columbia V0P 1S0, Canada

farms were infected with more sea lice than those in the peripheral category. Sea louse abundance on sockeye salmon and Pacific herring followed the same trends, but sample sizes were too low to support formal statistical analysis. The Pacific herring were translucent and lacked scales, and they were primarily parasitized by C. clemensi. These results suggest that the association of salmon farms with sea lice infestations of wild juvenile fish in Pacific Canada now extends beyond juvenile pink and chum salmon in the Broughton Archipelago. Canada's most abundant and economically valuable salmon populations, as well as British Columbia's most valuable Pacific herring stock, migrate through the Discovery Islands; hence, parasite transmission from farm to wild fish in this region may have important economic and ecological implications.

region of British Columbia. For pink and chum salmon we tested for the dependency of sea louse abundance on temperature, salinity, sampling period, host species, and farm exposure category. For both louse species, farm exposure was the only consistently significant predictor of sea lice abundance. Fish exposed to salmon farms were infected with more sea lice than those in the peripheral category. Sea louse abundance on sockeye salmon and Pacific herring followed the same trends, but sample sizes were too low to support formal statistical analysis. The Pacific herring were translucent and lacked scales, and they were primarily parasitized by C. clemensi. These results suggest that the association of salmon farms with sea lice infestations of wild juvenile fish in Pacific Canada now extends beyond juvenile pink and chum salmon in the Broughton Archipelago. Canada's most abundant and economically valuable salmon populations, as well as British Columbia's most valuable Pacific herring stock, migrate through the Discovery Islands; hence, parasite transmission from farm to wild fish in this region may have important economic and ecological implications.

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OPEN ACCESS Freely available online

PLoS one

Sea Louse Infection of Juvenile Sockeye Salmon in Relation to Marine Salmon Farms on Canada's West Coast

Michael H. H. Price[1,2]*, Stan L. Proboszcz[3], Rick D. Routledge[4], Allen S. Gottesfeld[5], Craig Orr[3], John D. Reynolds[6]

1 Department of Biology, University of Victoria, Victoria, Canada, **2** Raincoast Conservation Foundation, Sidney, Canada, **3** Watershed Watch Salmon Society, Coquitlam, Canada, **4** Department of Statistics and Actuarial Science, Simon Fraser University, Burnaby, Canada, **5** Skeena Fisheries Commission, Hazelton, Canada, **6** Earth to Ocean Research Group, Department of Biology, Simon Fraser University, Burnaby, Canada

Abstract

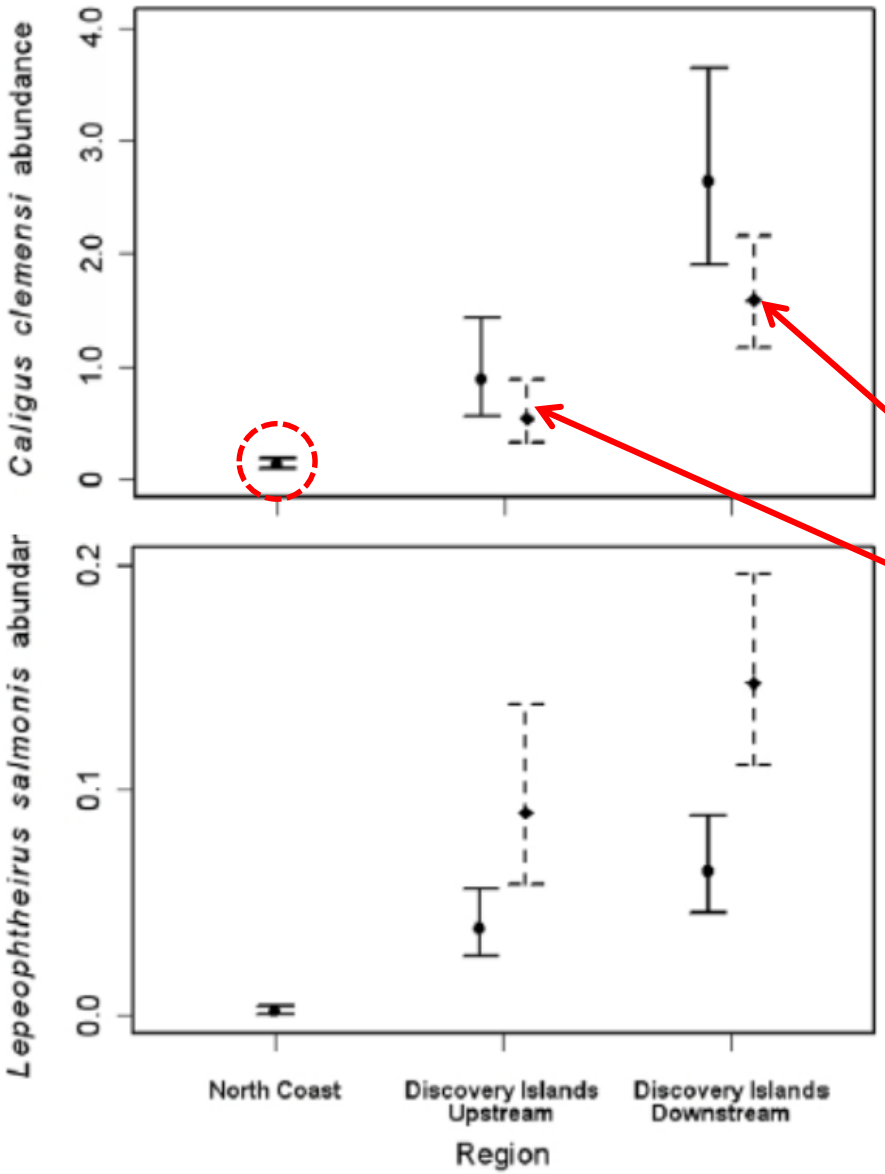
juveniles with sea lice data concurrently gathered on farms. Fraser River sockeye migrating through a region with salmon farms hosted an order of magnitude more sea lice than Skeena River populations, where there are no farms. Lice abundances on juvenile sockeye in the salmon farm region were substantially higher downstream of farms than upstream of farms for the two common species of lice: *Caligus clemensi* and *Lepeophtheirus salmonis*, and changes in their proportions between two years matched changes on the fish farms. Mixed-effects models show that position relative to salmon farms

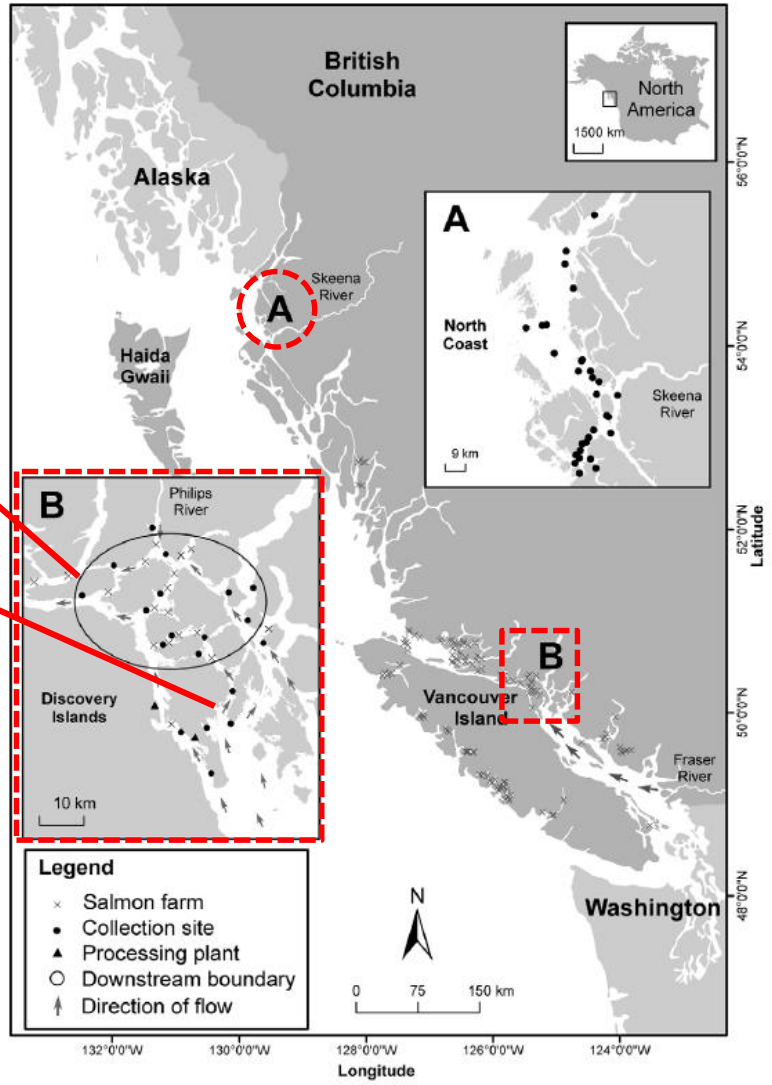
important salmon rivers, the Fraser and Skeena; Fraser sockeye migrate through a region with salmon farms, and Skeena sockeye do not. We compared lice levels between Fraser and Skeena juvenile sockeye, and within the salmon farm region we compared lice levels on wild fish either before or after migration past farms. We matched the latter data on wild juveniles with sea lice data concurrently gathered on farms. Fraser River sockeye migrating through a region with salmon farms hosted an order of magnitude more sea lice than Skeena River populations, where there are no farms. Lice abundances on juvenile sockeye in the salmon farm region were substantially higher downstream of farms than upstream of farms for the two common species of lice: *Caligus clemensi* and *Lepeophtheirus salmonis*, and changes in their proportions between two years matched changes on the fish farms. Mixed-effects models show that position relative to salmon farms best explained *C. clemensi* abundance on sockeye, while migration year combined with position relative to salmon farms and temperature was one of two top models to explain *L. salmonis* abundance.

Conclusions/Significance: This is the first study to demonstrate a potential role of salmon farms in sea lice transmission to juvenile sockeye salmon during their critical early marine migration. Moreover, it demonstrates a major migration corridor past farms for sockeye that originated in the Fraser River, a complex of populations that are the subject of conservation concern.

Source: Price et al. 2011. PLoS ONE e16851

Lice infection on juvenile Fraser River sockeye salmon in the Discovery Islands

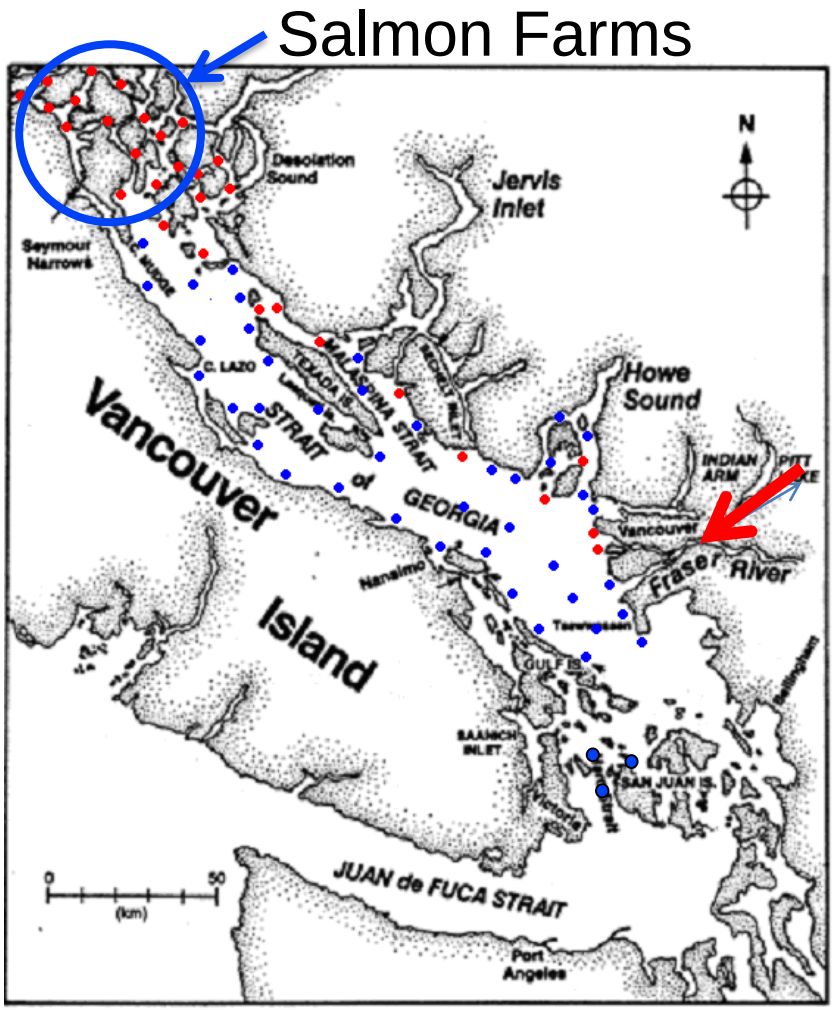




Source: Price et al. 2011. PLoS ONE e16851

Sea Lice and Health Surveys 2010-2012

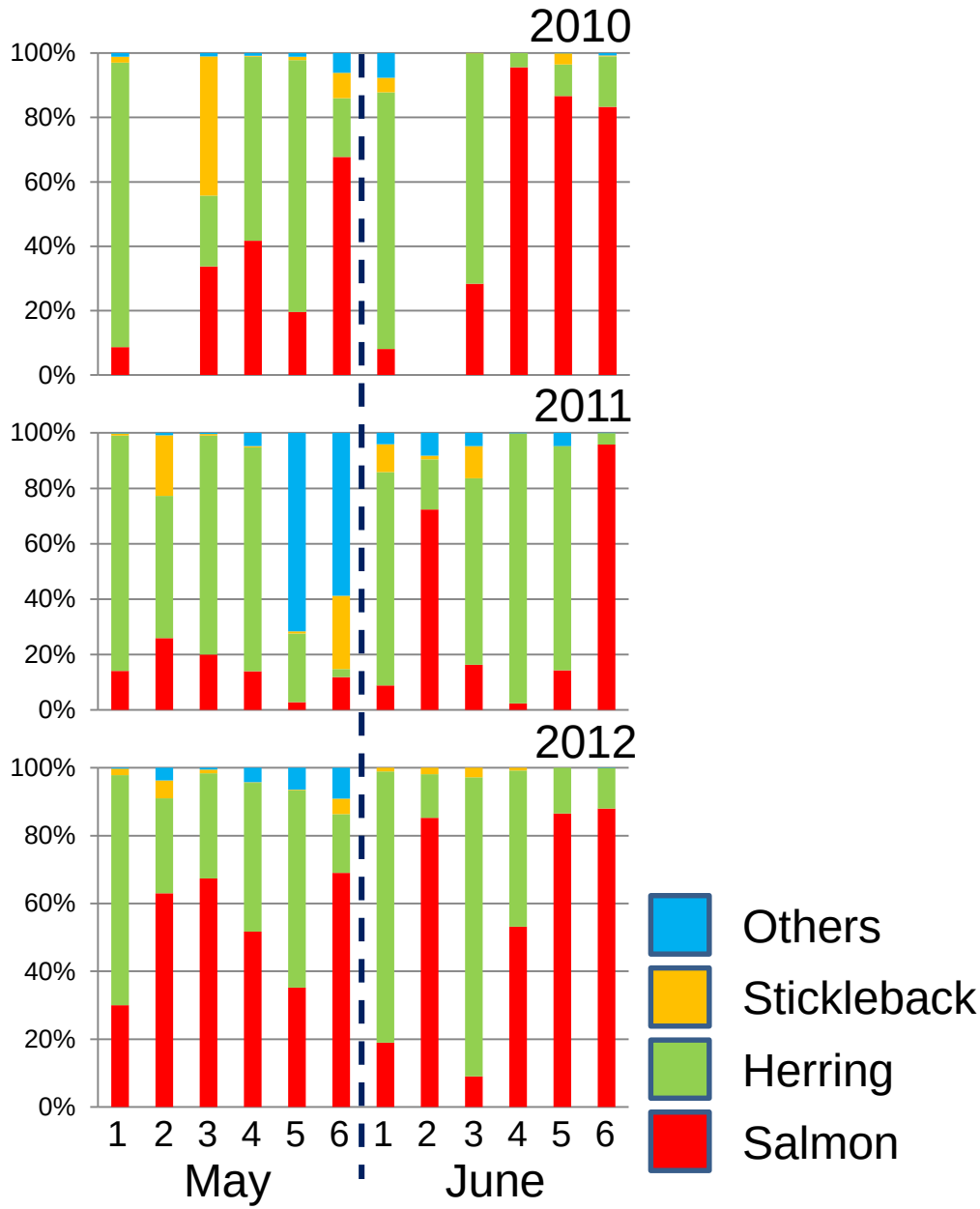
- Freshwater samples from Chilko Lake and lower Fraser River
- Marine samples collected throughout the Strait of Georgia and Johnstone Strait (75+ sites)
  - 2010 – 3 cruises
  - 2011 – 2 cruises
  - 2012 – 2 cruises.



Distribution of Sampling Sites

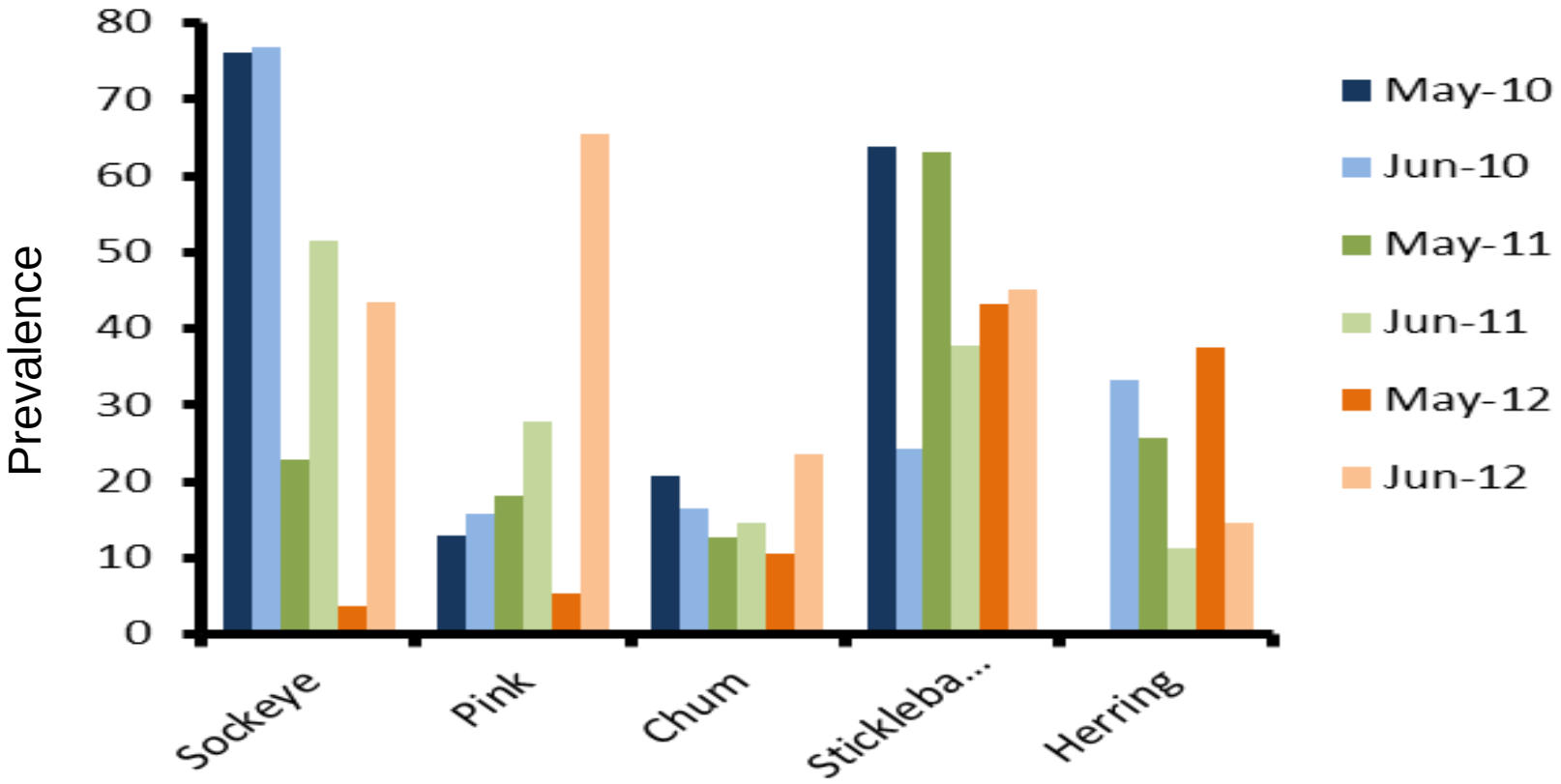
Stock ID (Chinook, Coho, Sockeye), Sea Lice (Pink, Chum, Sockeye, Non-salmonids), Histology (Chinook, Coho, Sockeye), Pathogen Screening (Sockeye), Feeding Ecology and Growth

Catch composition 2010-2012



- Few resident wild salmonids present
- Non-salmonid hosts were dominant in May and in June 2011 catches.
- Juvenile salmon were dominate in June 2010 and 2012, due in part to extremely high numbers of Pink salmon from the Fraser River in even years.
- Purse seine doesn't adequately sample some non-salmonid host species.

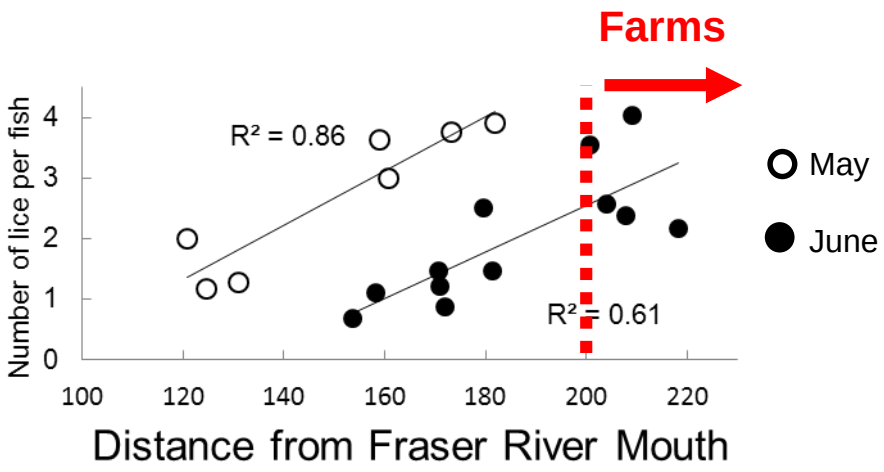
Sea lice prevalence 2010-2012



- *Caligus clemensi* was dominant on all salmon and non-salmonid hosts.
- Stickleback and Sockeye Salmon have the highest sea lice burdens.
- Stickleback are resident others migratory.

Sea Lice Infection with Distance

- Prevalence and intensity of infection are directly related to time in SW
- There is no evidence for dramatic increases in sea lice numbers on sockeye in the “impact zone” of salmon farms



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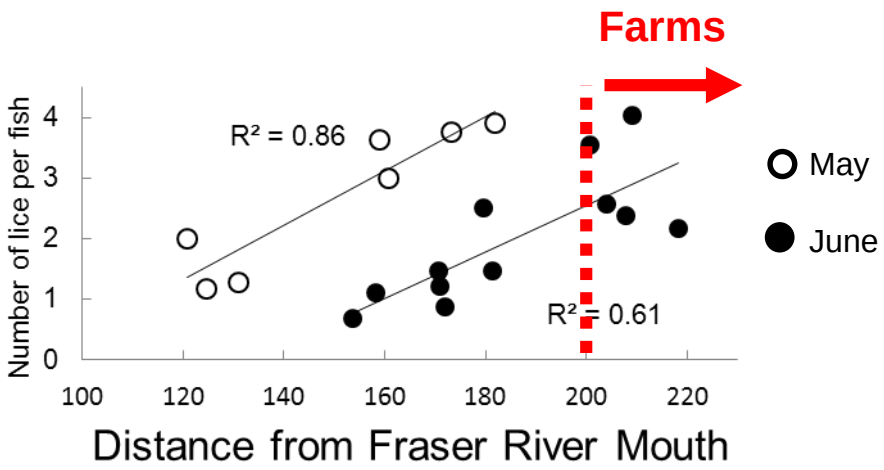
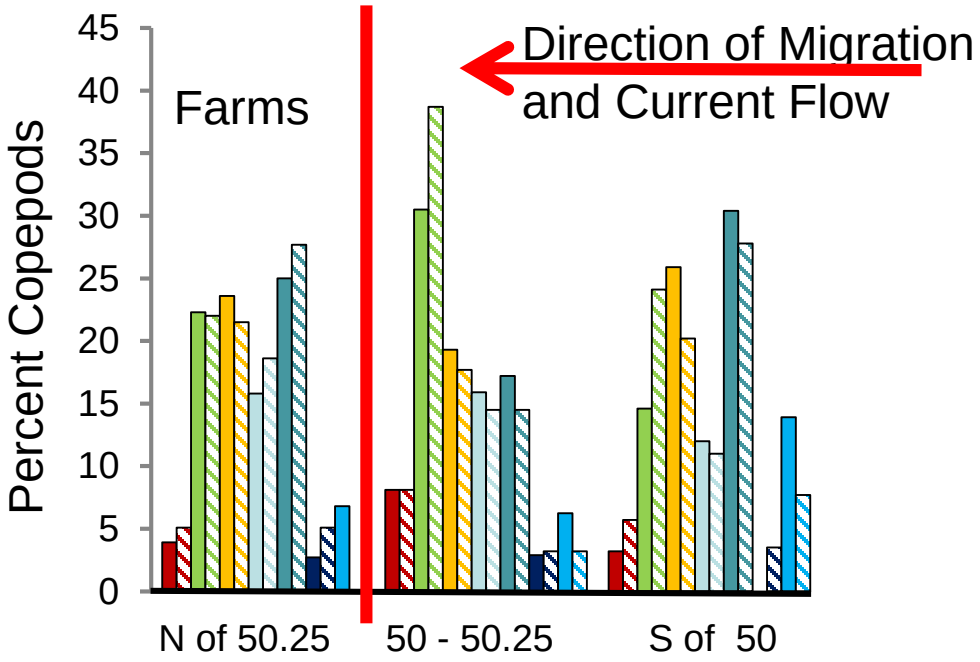
- Prevalence and intensity of infection are directly related to time in SW
- There is no evidence for dramatic increases in sea lice numbers on sockeye in the “impact zone” of salmon farms





Salmon farms in Southern British Columbia

Risk = Exposure * Consequences

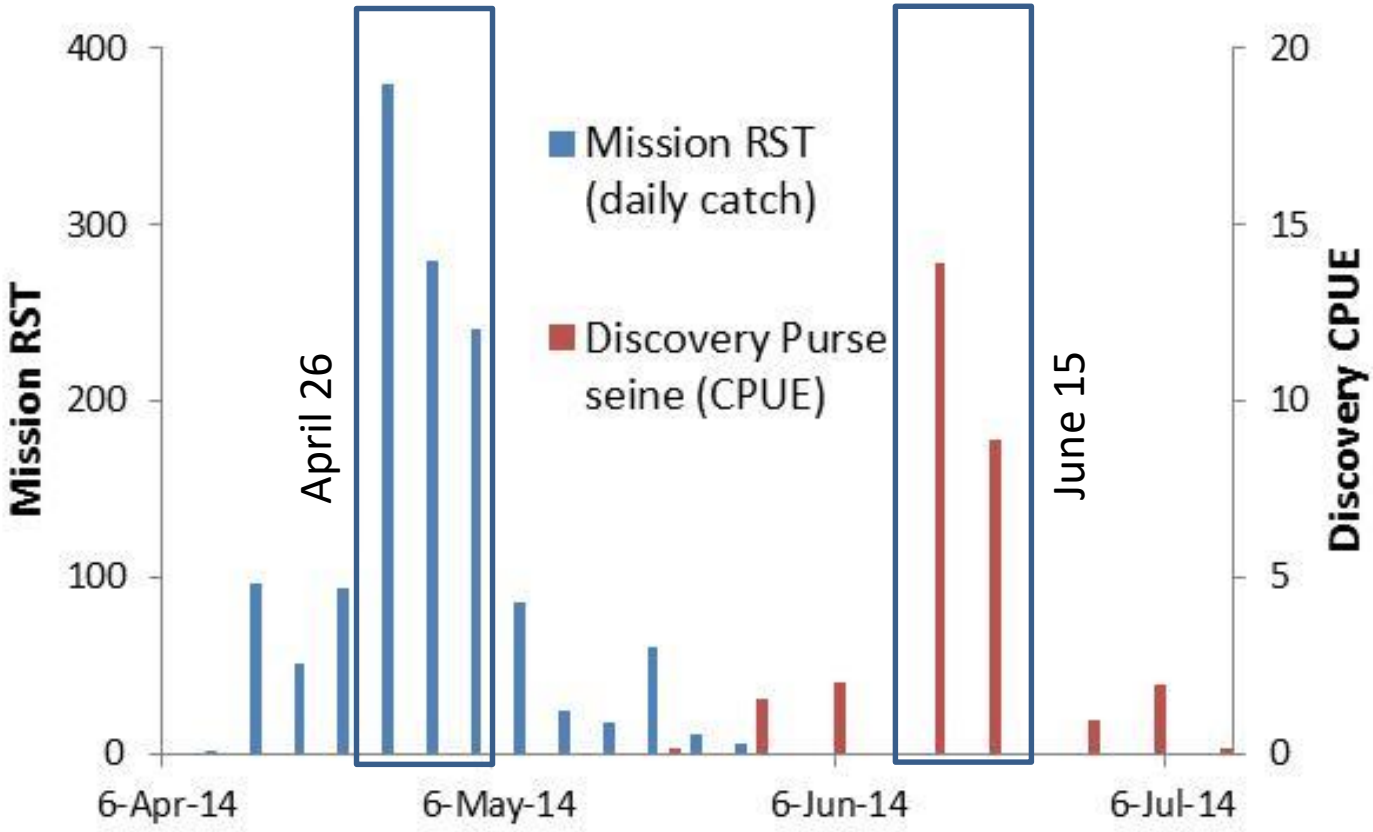
Adapted from Tucker et al. (2009) and Beacham et al. (2014a,b)

Residence time of juvenile Fraser River sockeye

Purse seine surveys in Johnstone Strait (2014-2016)

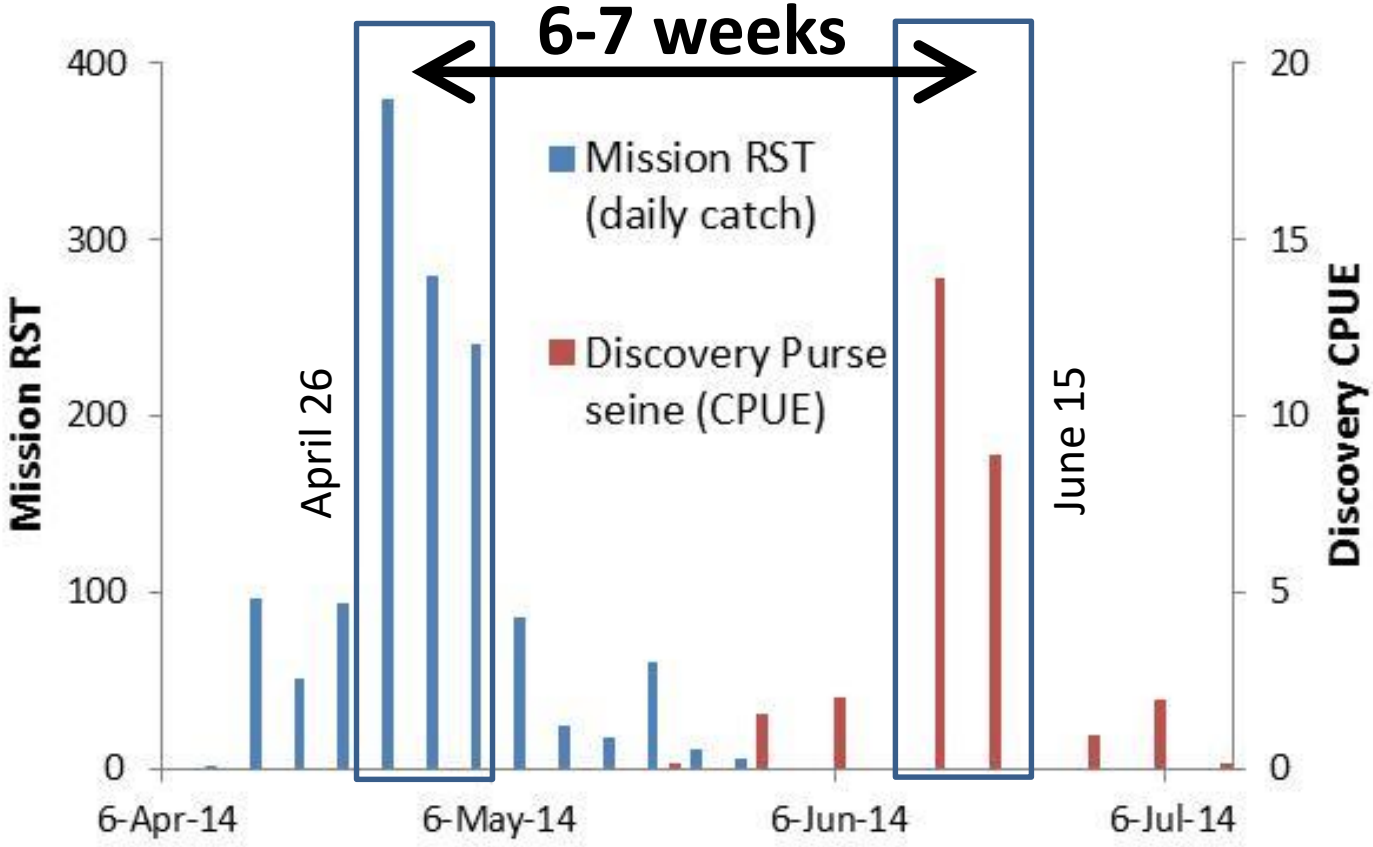
Rotary screw trap sampling at Mission (2012-2016)

Fraser River sockeye salmon residence time in the Strait of Georgia (2014)



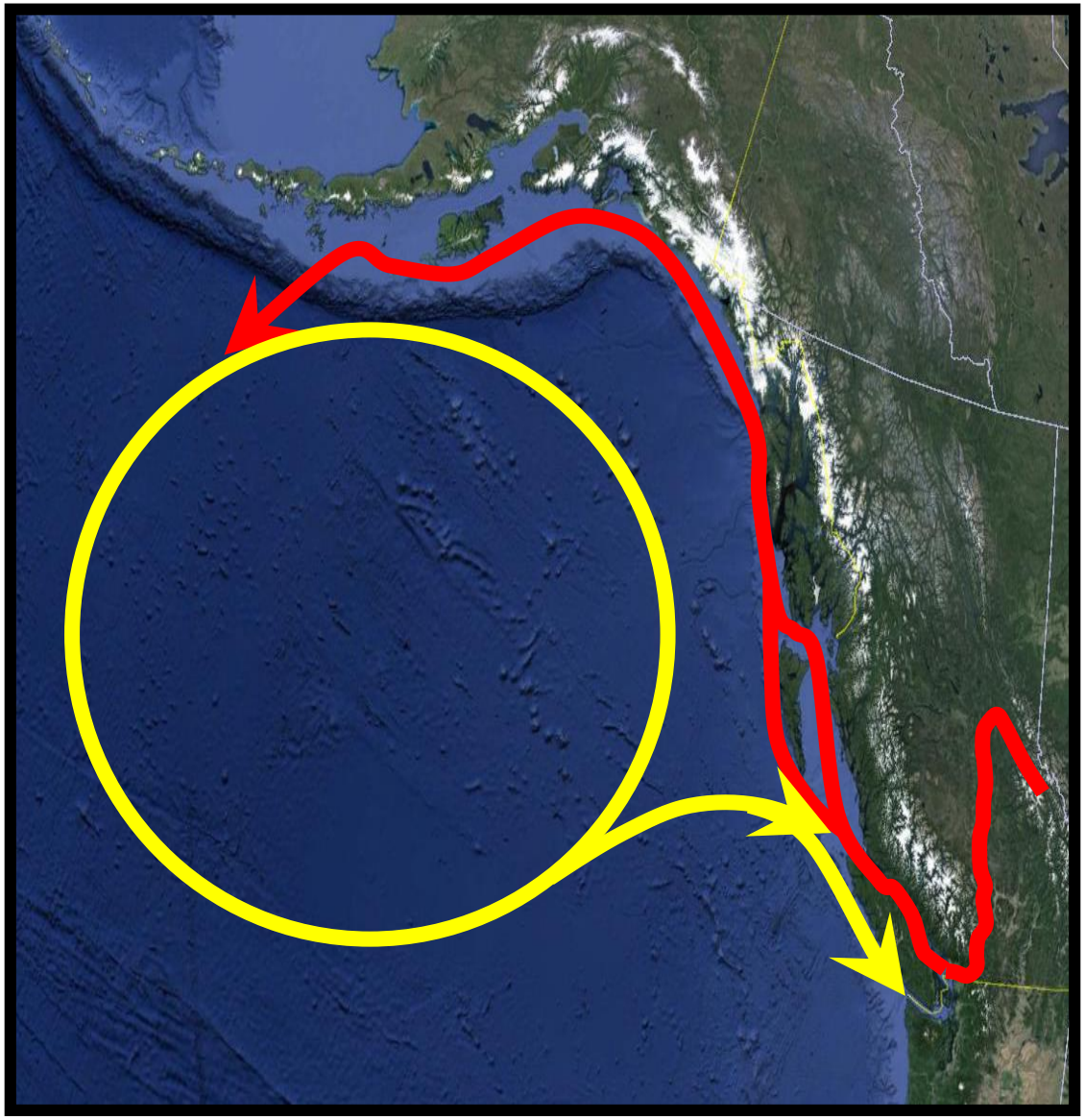
Source: Neville et al. (in press)

Fraser River sockeye salmon residence time in the Strait of Georgia (2014)

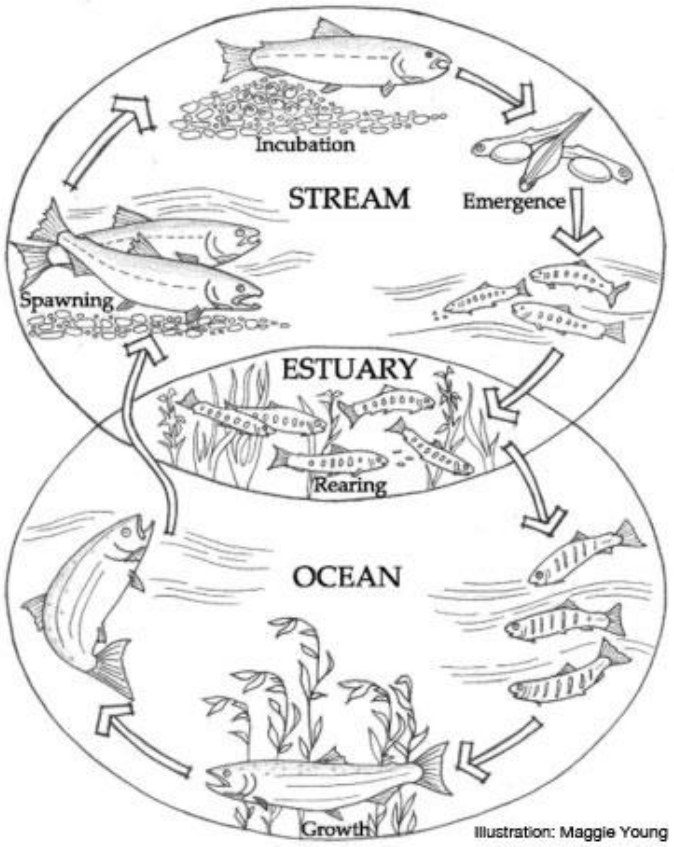


Source: Neville et al. (in press)

Cumulative impacts of multiple stressors

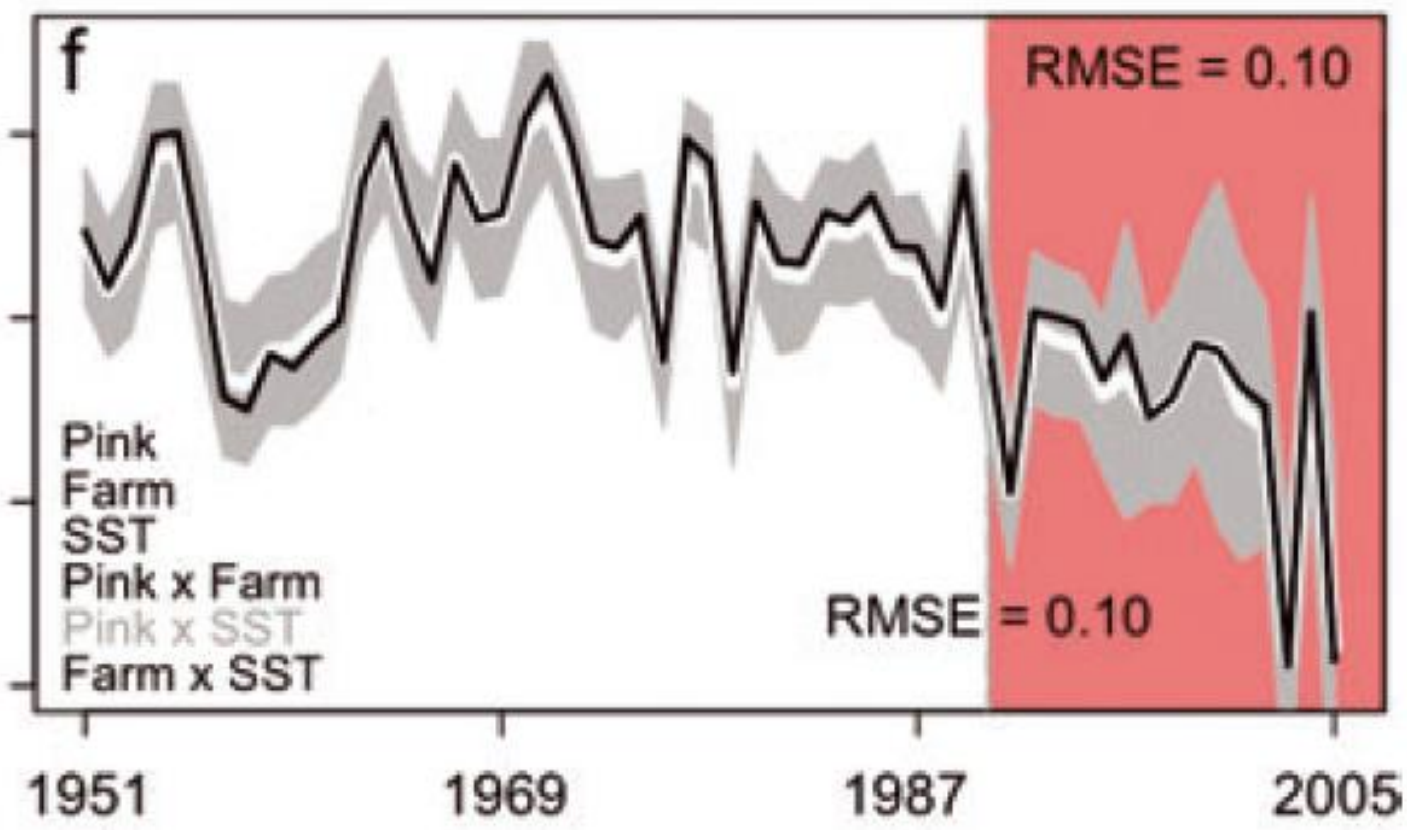


Adapted from Hartt and Dell. 1986. INPFC 46: 1-105



http://www.marinwatersheds.org/salmonids.html

Cumulative effects of climate, competition, and salmon farms on sockeye recruitment



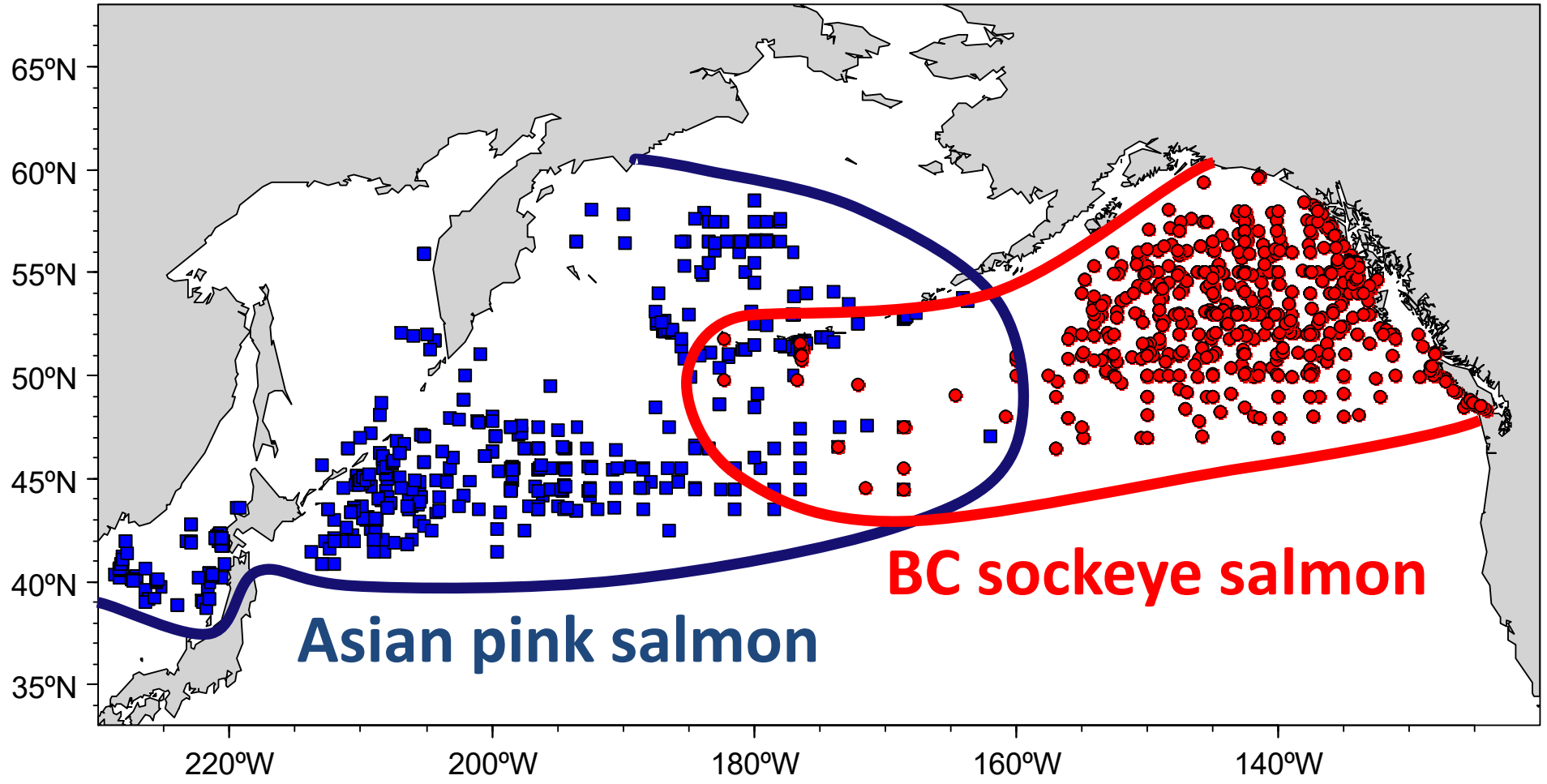
Source: Connors et al. (2012). Cons. Let.

Cumulative effects of climate, competition, and salmon farms on sockeye recruitment.

… when the contribution of Russian pink salmon to the index of pink salmon competitors was removed (Russian pink salmon are ~65% of total pink-salmon abundance from 1952 to 2010), accounting for the interaction between pink-salmon abundance and farmed-salmon production barely improved our ability to predict the decline in Fraser sockeye

Source: Connors et al. (2012). Cons. Let.

Competition between Asian pink salmon and BC sockeye salmon?



Data from www.npafc.org

Concluding remarks

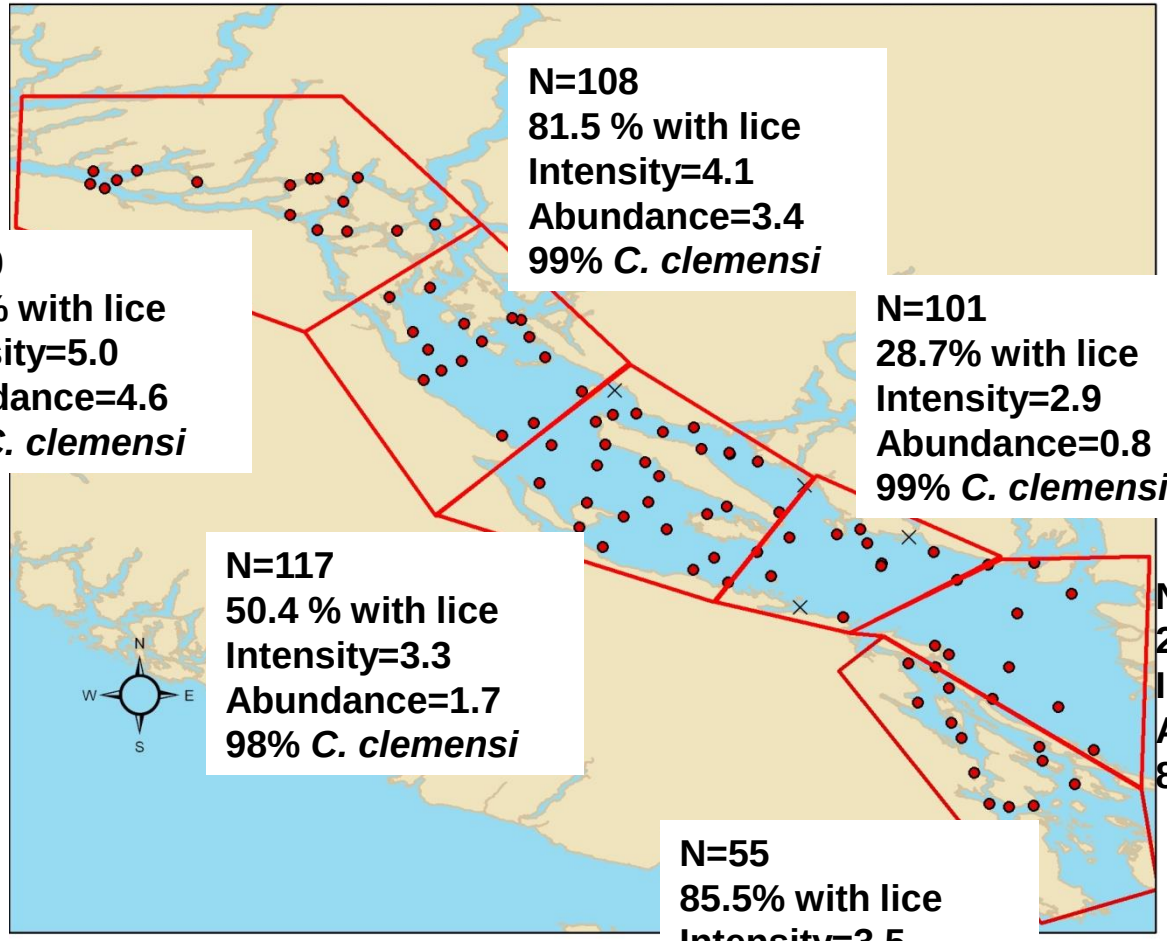
1. Life-history and migration behaviour must be considered when assessing the interactions between wild and farmed salmon

2. The infection history prior to reaching the salmon farms need to be considered to understand their dynamics.

3. Residence time of juvenile Fraser River sockeye salmon in the Discovery Islands is likely short.

Strait of Georgia Juvenile salmon survey
June 11-25, 2012

Sea lice numbers on juvenile sockeye salmon



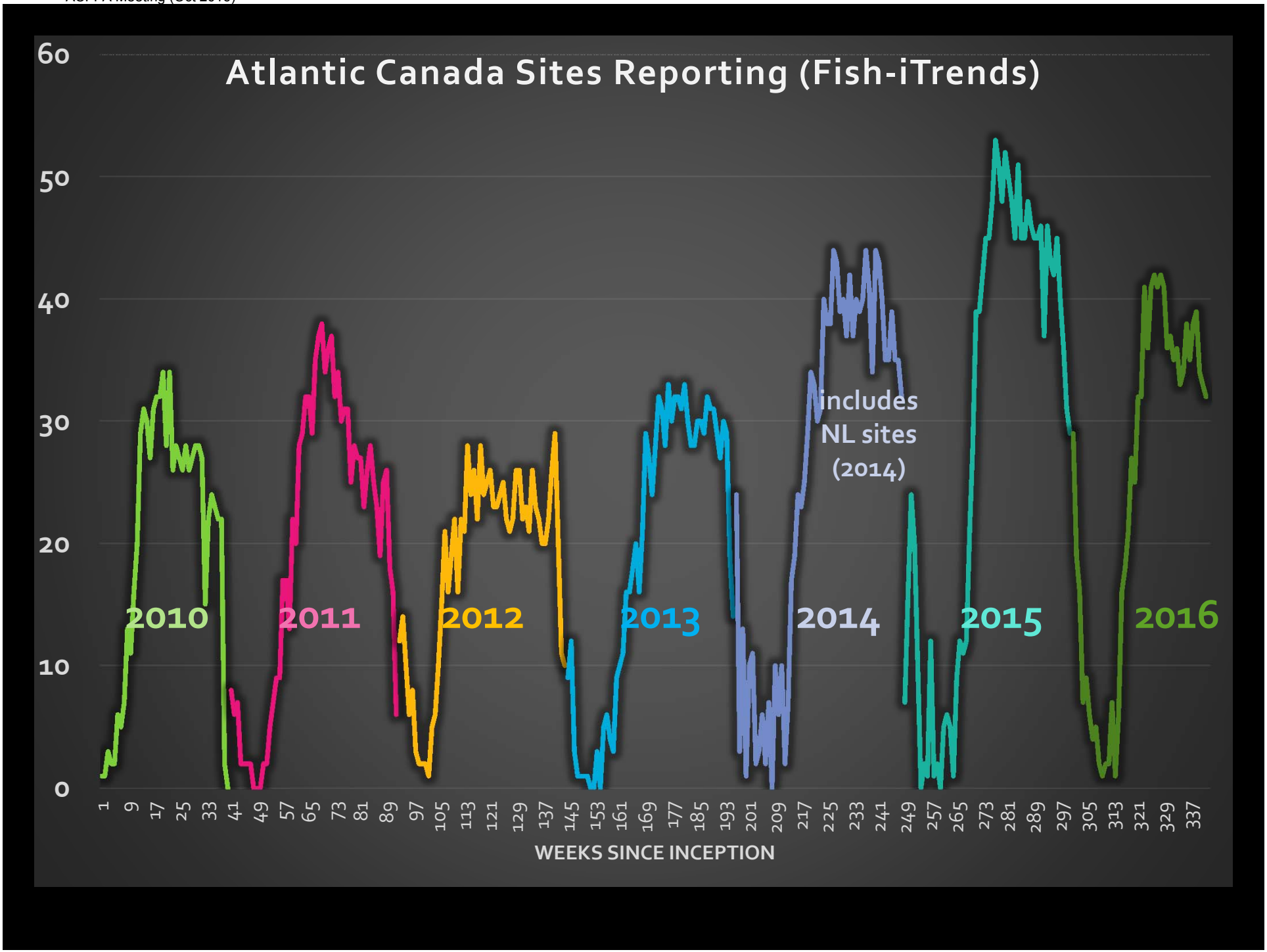
51.57%

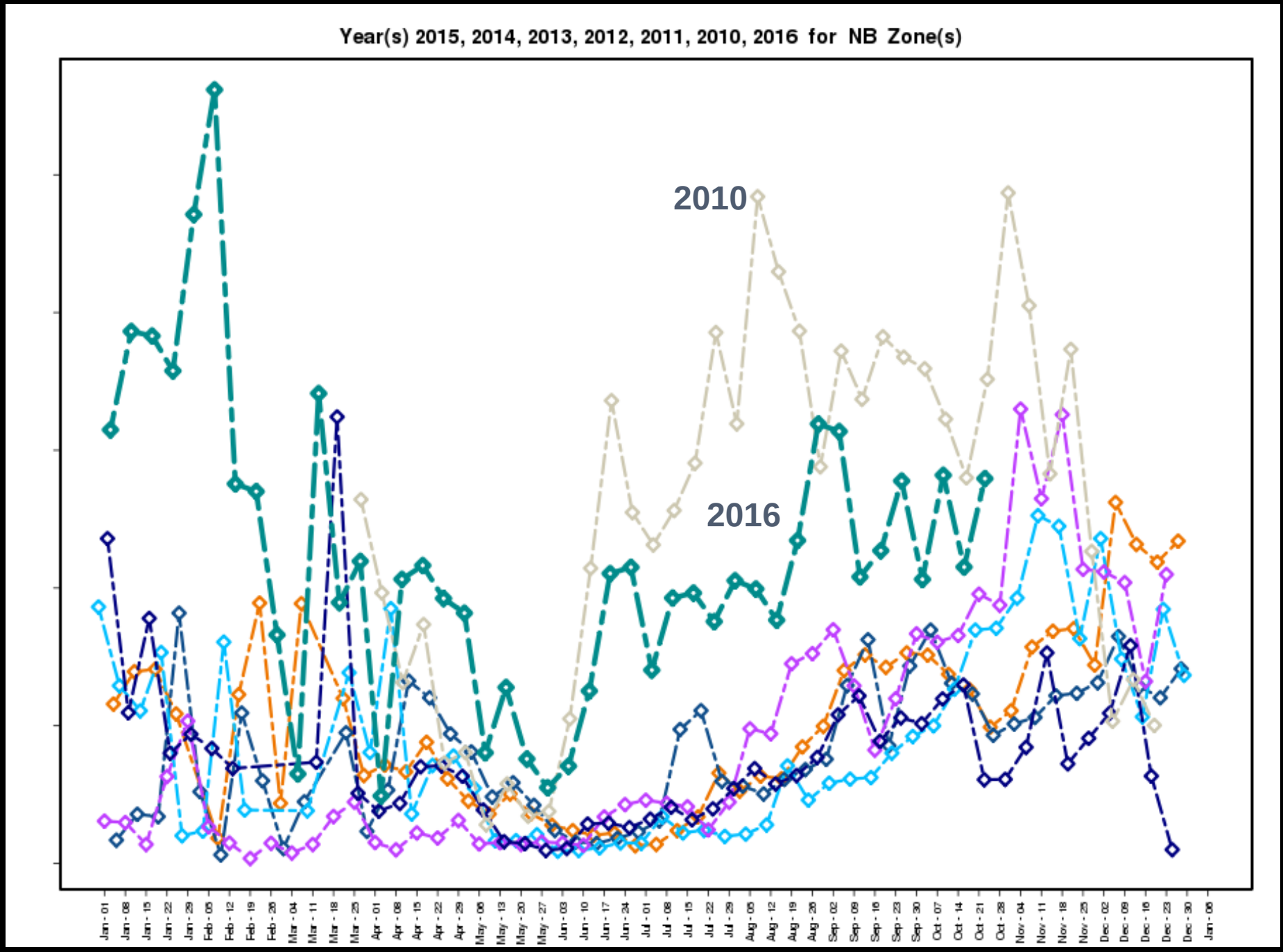
X = set with 0 sockeye catch

SEA LICE 2016

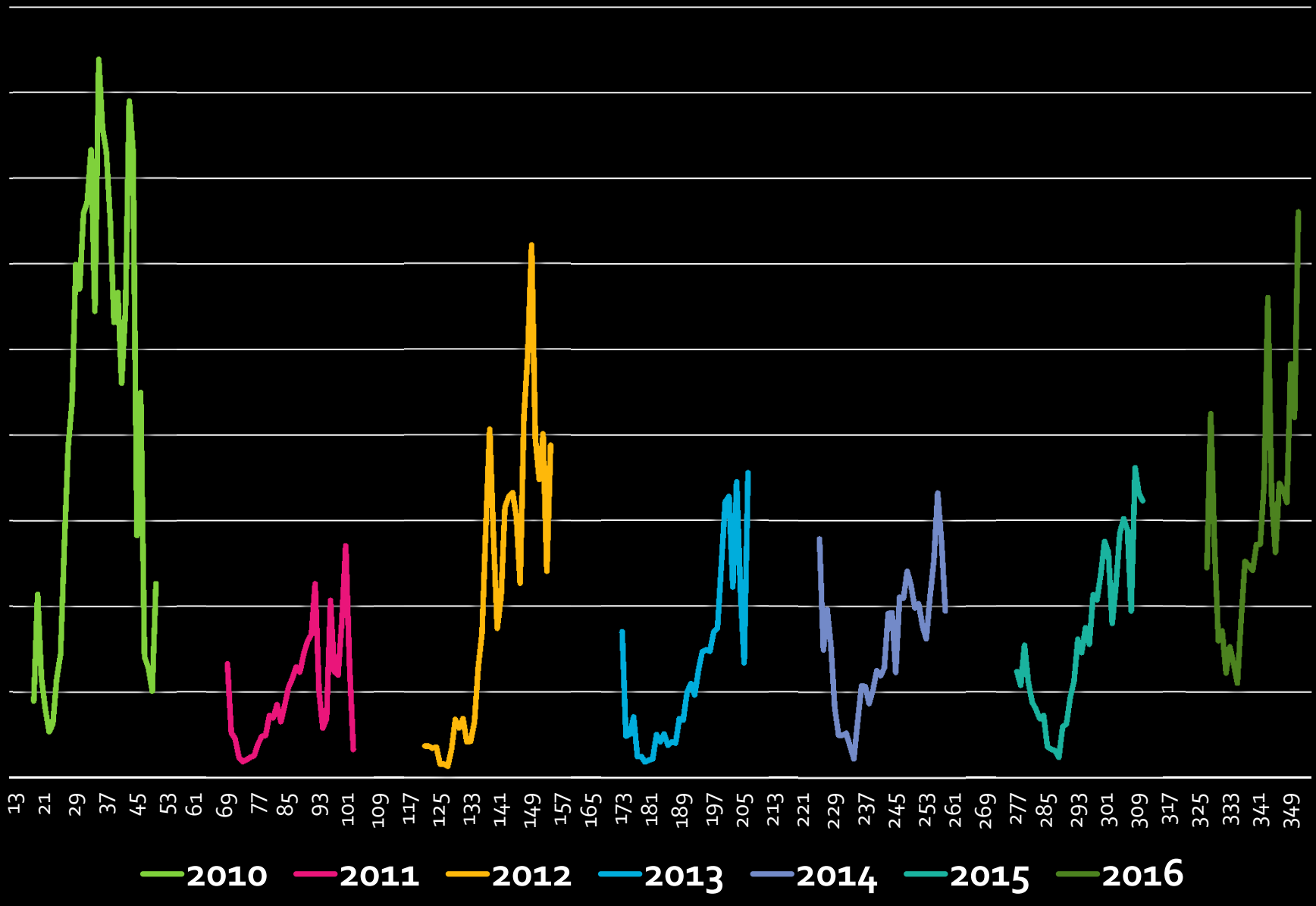
TRENDS TO INFORM MANAGEMENT DECISIONS IN NB

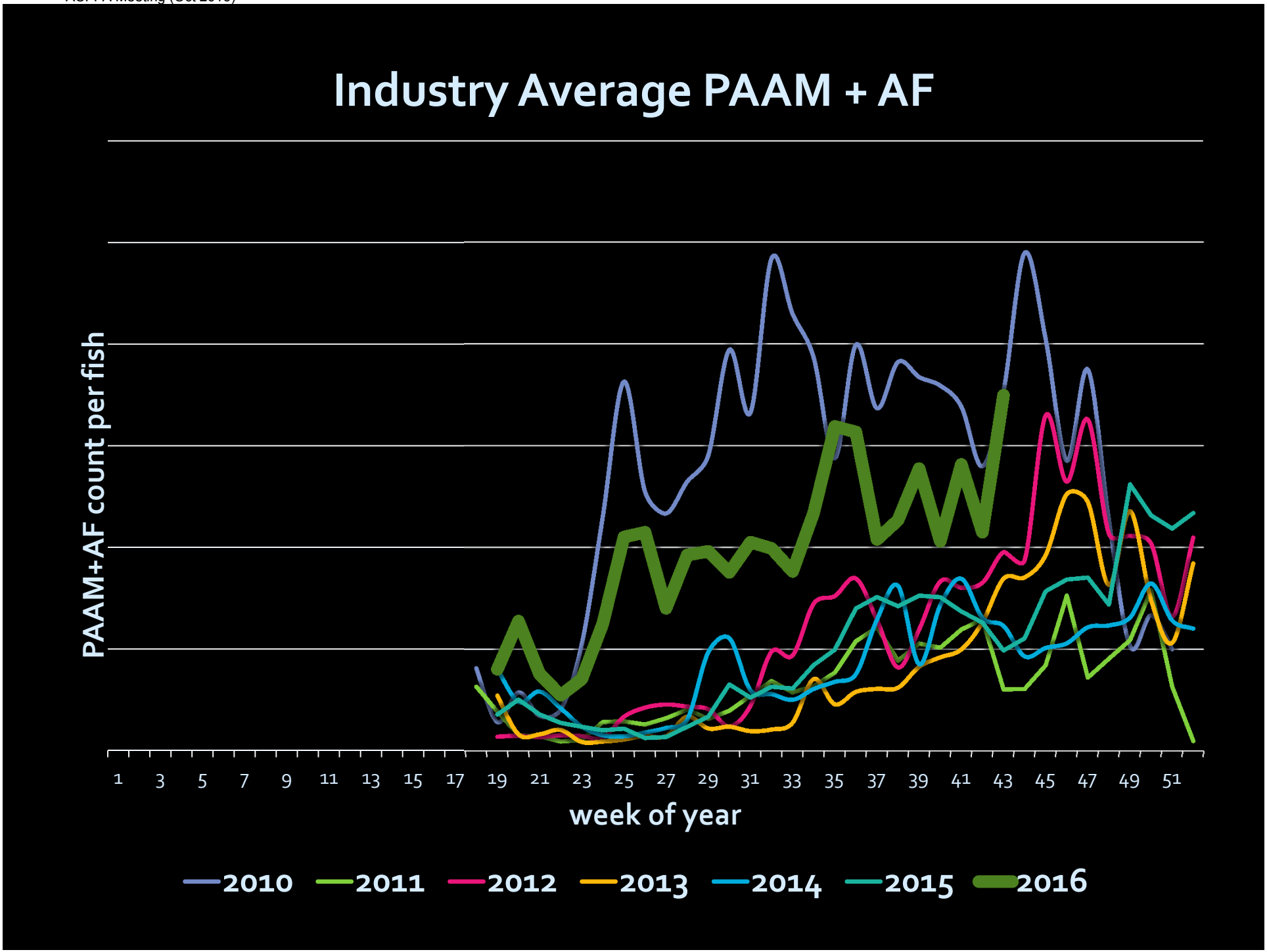
Larry Hammell
Assoc Dean, Grad Studies & Research,
Professor, Dept of Health Management,
Atlantic Veterinary College,
University of Prince Edward Island
Charlottetown, PE, Canada


Atlantic Canada Sites Reporting (Fish-iTrends)
60
50
40
30
20
10
0
2010
2011
2012
2013
2014
2015
2016
includes
NL sites
(2014)
1
9
17
25
33
41
49
57
65
73
81
89
97
105
113
121
129
137
145
153
161
169
177
185
193
201
209
217
225
233
241
249
257
265
273
281
289
297
305
313
321
329
337
WEEKS SINCE INCEPTION


Year(s) 2015, 2014, 2013, 2012, 2011, 2010, 2016 for NB Zone(s)
2010
2016
Jan - 01
Jan - 08
Jan - 15
Jan - 22
Jan - 29
Feb - 05
Feb - 12
Feb - 19
Feb - 26
Mar - 04
Mar - 11
Mar - 18
Mar - 25
Apr - 01
Apr - 08
Apr - 15
Apr - 22
Apr - 29
May - 06
May - 13
May - 20
May - 27
Jun - 03
Jun - 10
Jun - 17
Jun - 24
Jul - 01
Jul - 08
Jul - 15
Jul - 22
Jul - 29
Aug - 05
Aug - 12
Aug - 19
Aug - 26
Sep - 02
Sep - 09
Sep - 16
Sep - 23
Sep - 30
Oct - 07
Oct - 14
Oct - 21
Oct - 28
Nov - 04
Nov - 11
Nov - 18
Nov - 25
Dec - 02
Dec - 09
Dec - 16
Dec - 23
Dec - 30
Jan - 06

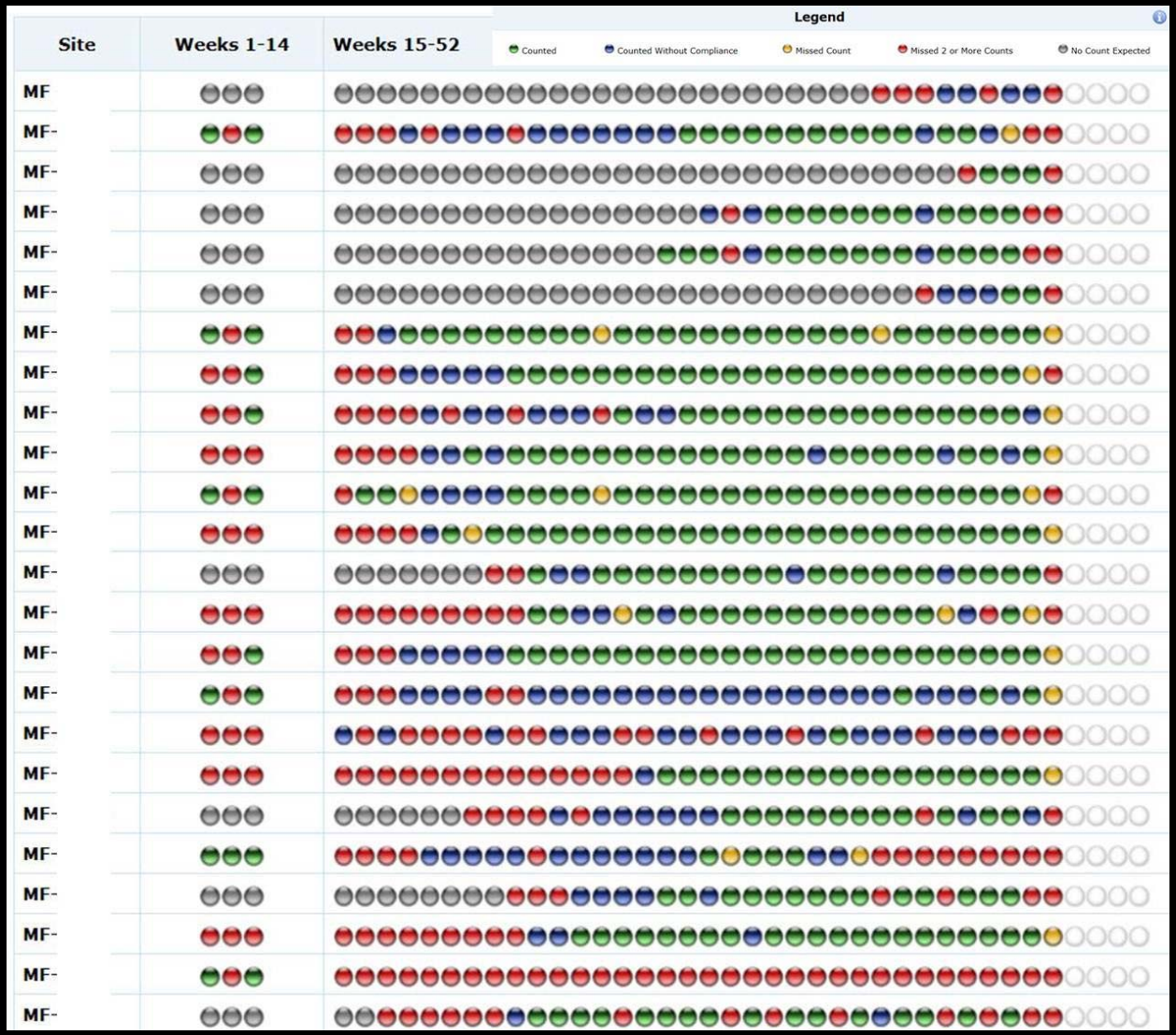
AF (industry average)

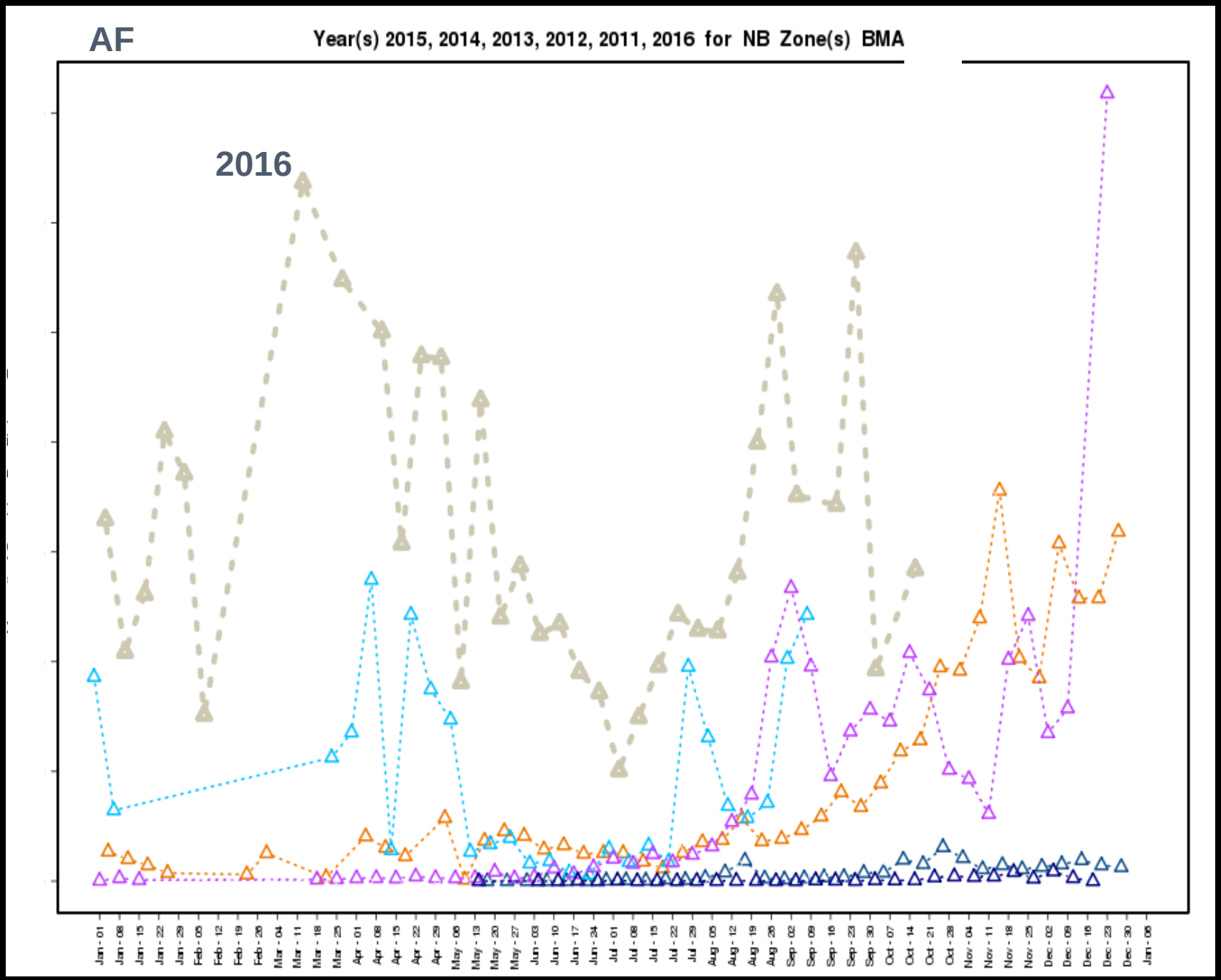


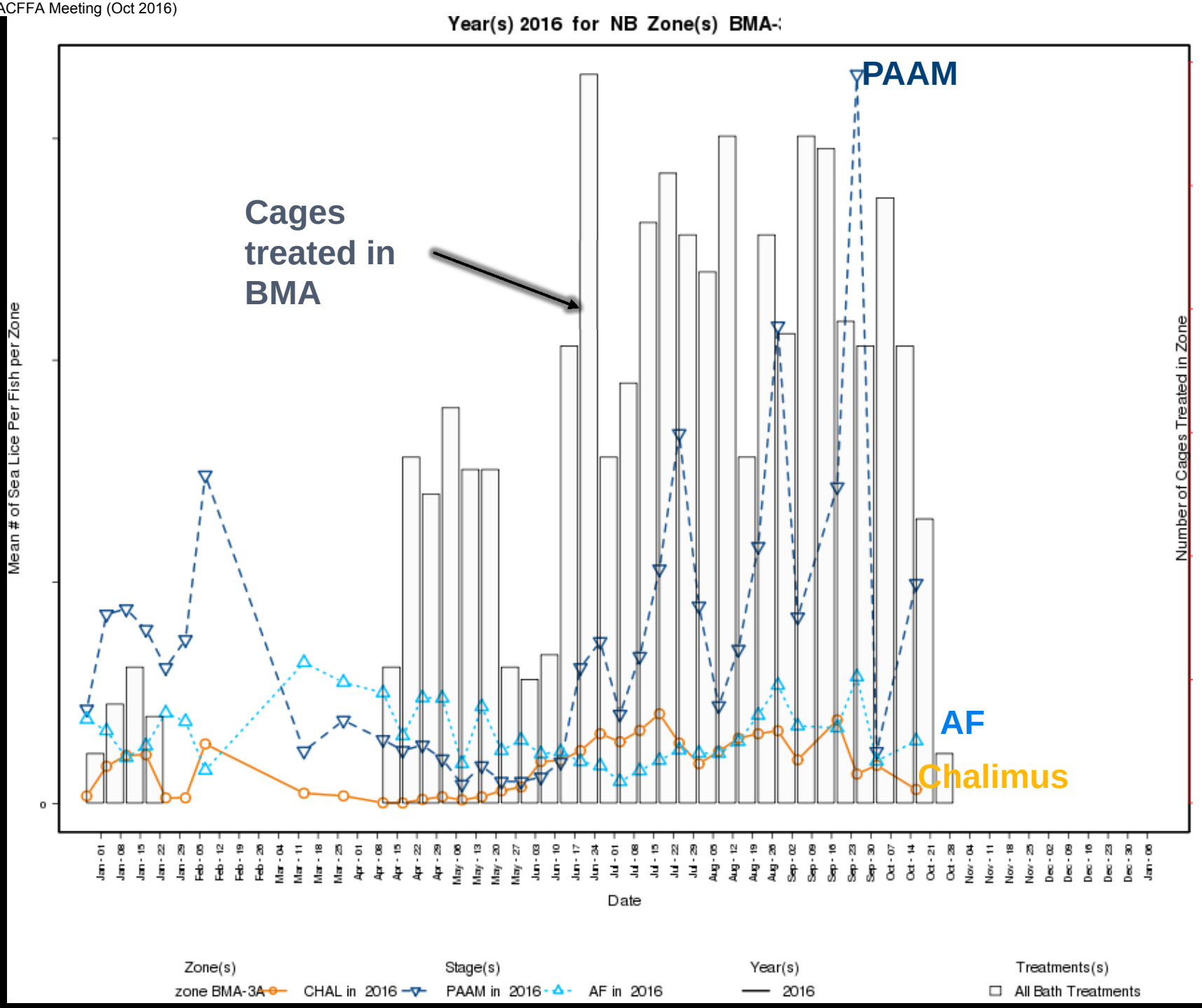

Industry Average PAAM + AF
PAAM+AF count per fish
1
3
5
7
9
11
13
15
17
19
21
23
25
27
29
31
33
35
37
39
41
43
45
47
49
51
week of year
2010
2011
2012
2013
2014
2015
2016

Count compliance

Sites reporting fish/cages to meet provincial requirement



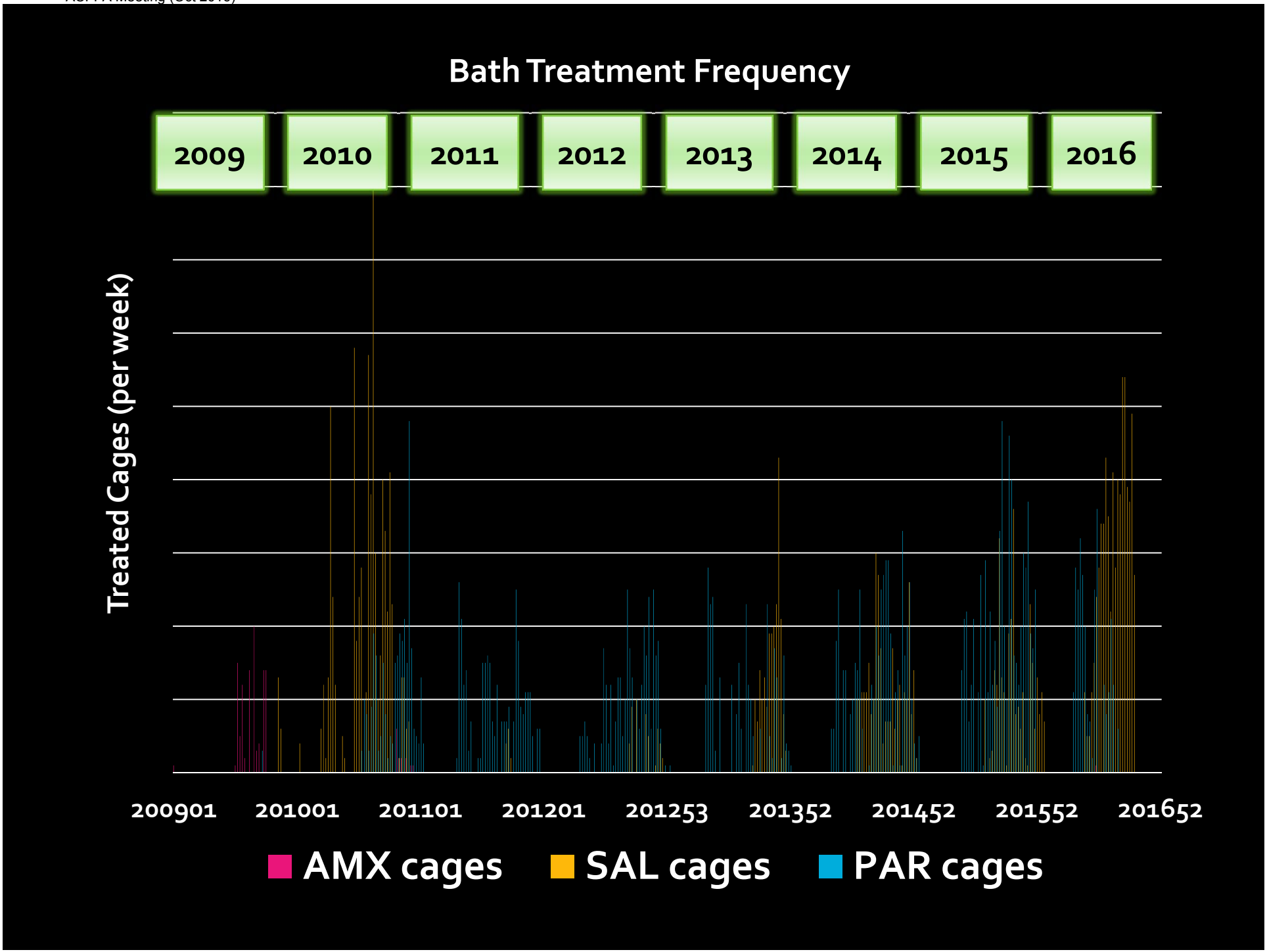

AF
Year(s) 2015, 2014, 2013, 2012, 2011, 2016 for NB Zone(s) BMA
2016
Jan - 01
Jan - 08
Jan - 15
Jan - 22
Jan - 29
Feb - 05
Feb - 12
Feb - 19
Feb - 26
Mar - 04
Mar - 11
Mar - 18
Mar - 25
Apr - 01
Apr - 08
Apr - 15
Apr - 22
Apr - 29
May - 06
May - 13
May - 20
May - 27
Jun - 03
Jun - 10
Jun - 17
Jun - 24
Jul - 01
Jul - 08
Jul - 15
Jul - 22
Jul - 29
Aug - 05
Aug - 12
Aug - 19
Aug - 26
Sep - 02
Sep - 09
Sep - 16
Sep - 23
Sep - 30
Oct - 07
Oct - 14
Oct - 21
Oct - 28
Nov - 04
Nov - 11
Nov - 18
Nov - 25
Dec - 02
Dec - 09
Dec - 16
Dec - 23
Dec - 30
Jan - 06

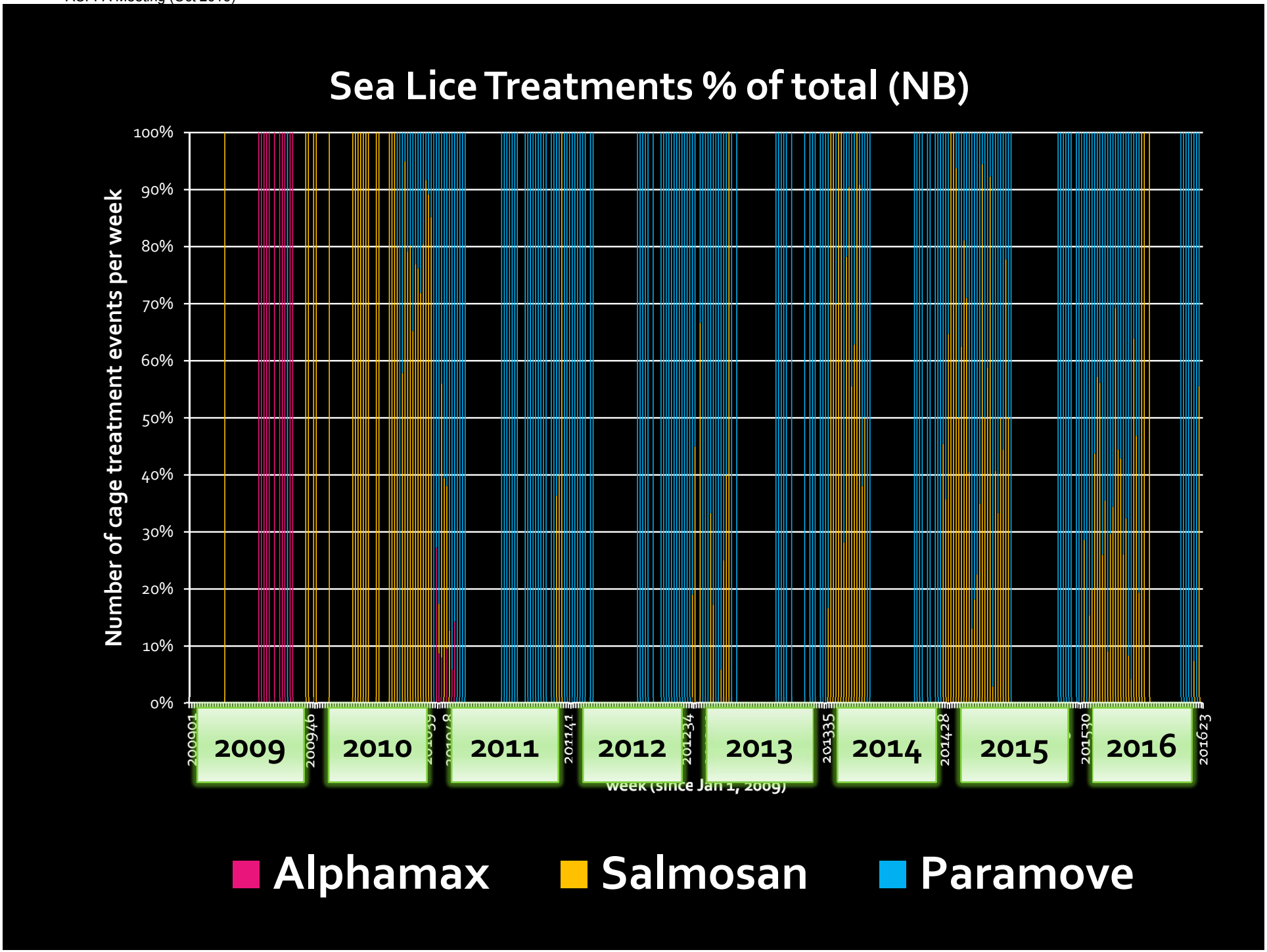

Year(s) 2016 for NB Zone(s) BMA-3
PAAM
Cages treated in BMA
AF
Chalimus
Mean # of Sea Lice Per Fish per Zone
Number of Cages Treated in Zone
0
Jan - 01
Jan - 08
Jan - 15
Jan - 22
Jan - 29
Feb - 05
Feb - 12
Feb - 19
Feb - 26
Mar - 04
Mar - 11
Mar - 18
Mar - 25
Apr - 01
Apr - 08
Apr - 15
Apr - 22
Apr - 29
May - 06
May - 13
May - 20
May - 27
Jun - 03
Jun - 10
Jun - 17
Jun - 24
Jul - 01
Jul - 08
Jul - 15
Jul - 22
Jul - 29
Aug - 05
Aug - 12
Aug - 19
Aug - 26
Sep - 02
Sep - 09
Sep - 16
Sep - 23
Sep - 30
Oct - 07
Oct - 14
Oct - 21
Oct - 28
Nov - 04
Nov - 11
Nov - 18
Nov - 25
Dec - 02
Dec - 09
Dec - 16
Dec - 23
Dec - 30
Jan - 06
Date
Zone(s)
zone BMA-3A
CHAL in 2016
Stage(s)
PAAM in 2016
AF in 2016
Year(s)
2016
Treatments(s)
All Bath Treatments

Automatic summaries emailed to industry decision makers weekly

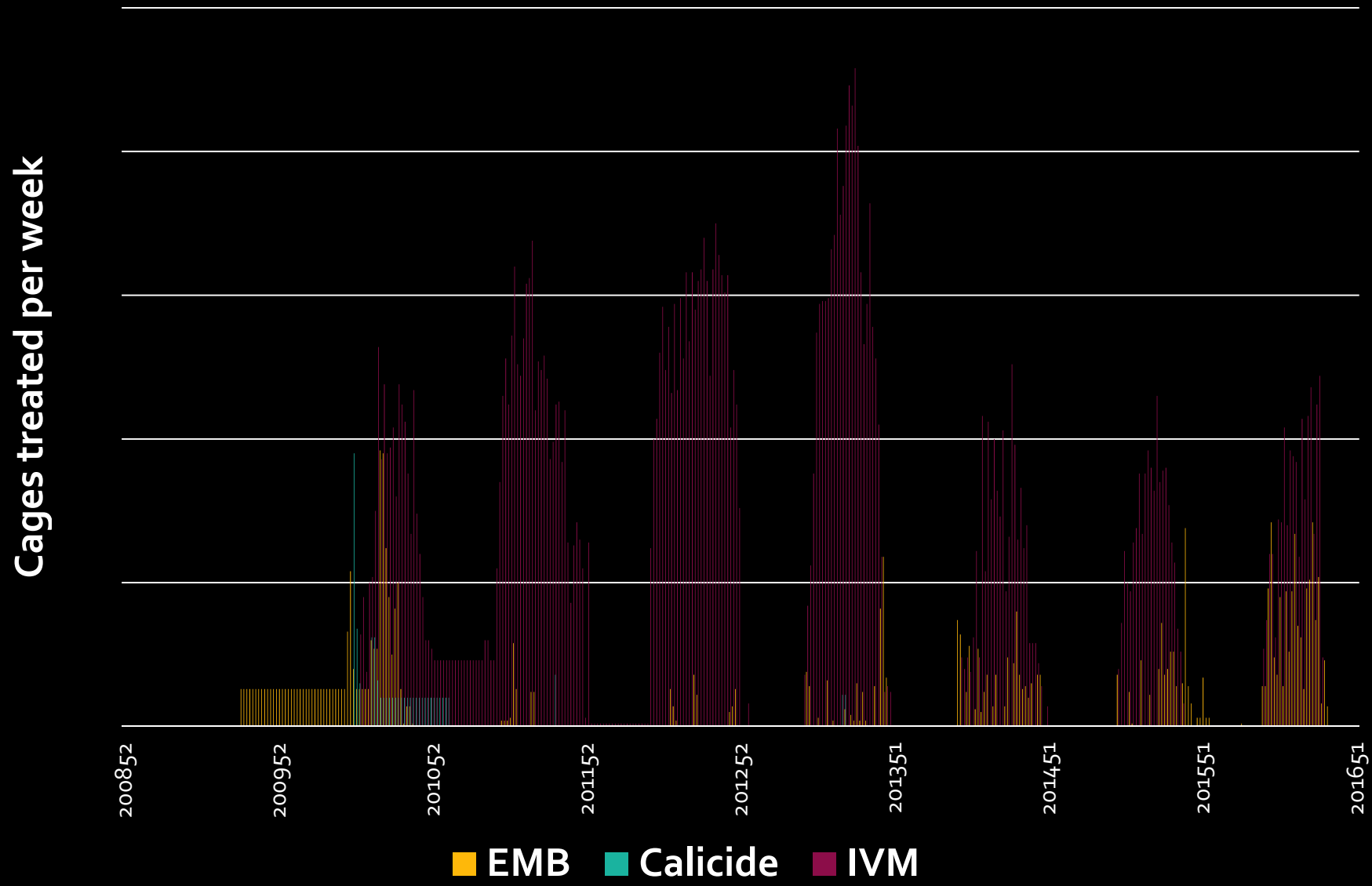
example

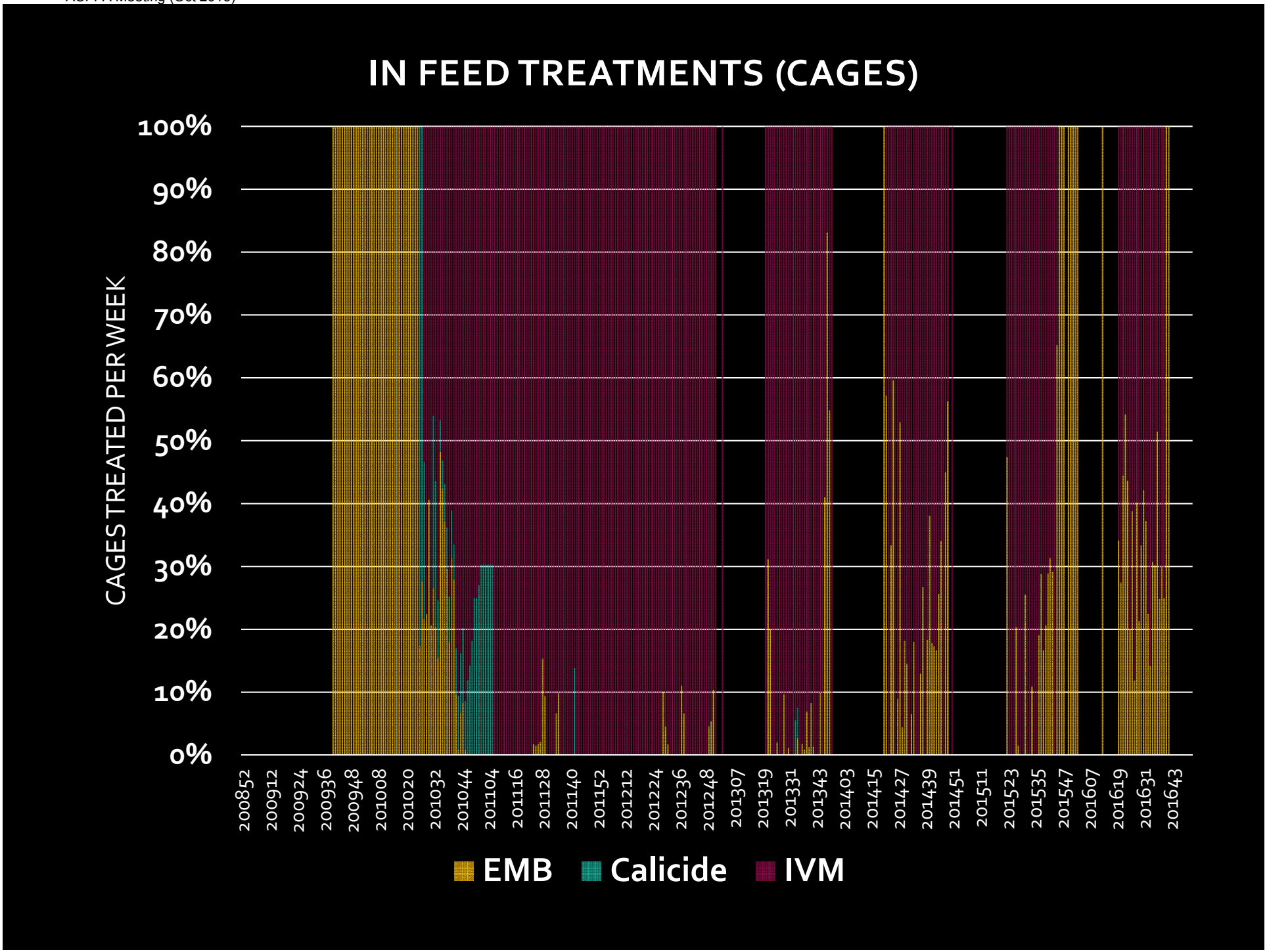
Total	Site	CHAL	PAAM	AF
+50	ID-38	-10	+53	+7
+45	ID-80	+7	+34	+4
+30	ID-18	+6	+22	+2
+8	ID-13	+3	+5	0
+8	ID-9	-1	+7	+2
+3	ID-14	0	+2	+1
+2	ID-274	0	+2	0
+2	ID-113	0	+1	+1
+2	ID-29	+1	+1	0
+2	ID-79	+2	-1	+1
+2	ID-24	0	+2	0
+2	ID-16	0	+1	+1
+1	ID-124	0	+1	0
-1	ID-10	-2	-1	+2
-1	ID-23	0	-1	0
-3	ID-8	-9	+6	0
-6	ID-17	-15	+4	+5
-17	ID-21	-14	-1	-2
-17	ID-106	-2	-11	-4
-31	ID-71	+1	-16	-16
-38	ID-12	-7	-26	-5
-46	ID-31	-6	-15	-25
-46	ID-99	-1	-35	-10
-50	ID-287	-18	-25	-7

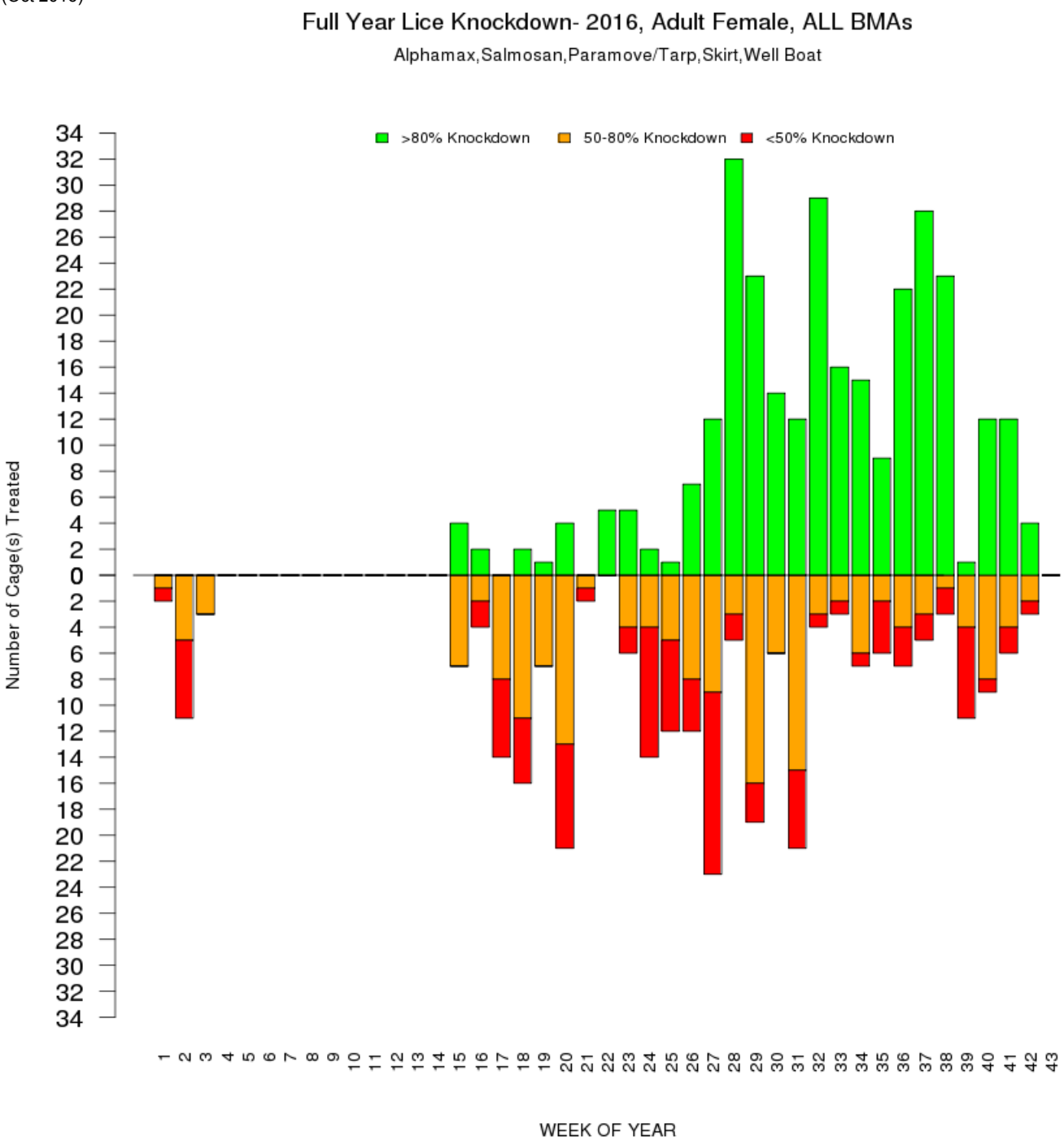

Bath Treatment Frequency
2009
2010
2011
2012
2013
2014
2015
2016
Treated Cages (per week)
200901
201001
201101
201201
201253
201352
201452
201552
201652
AMX cages
SAL cages
PAR cages

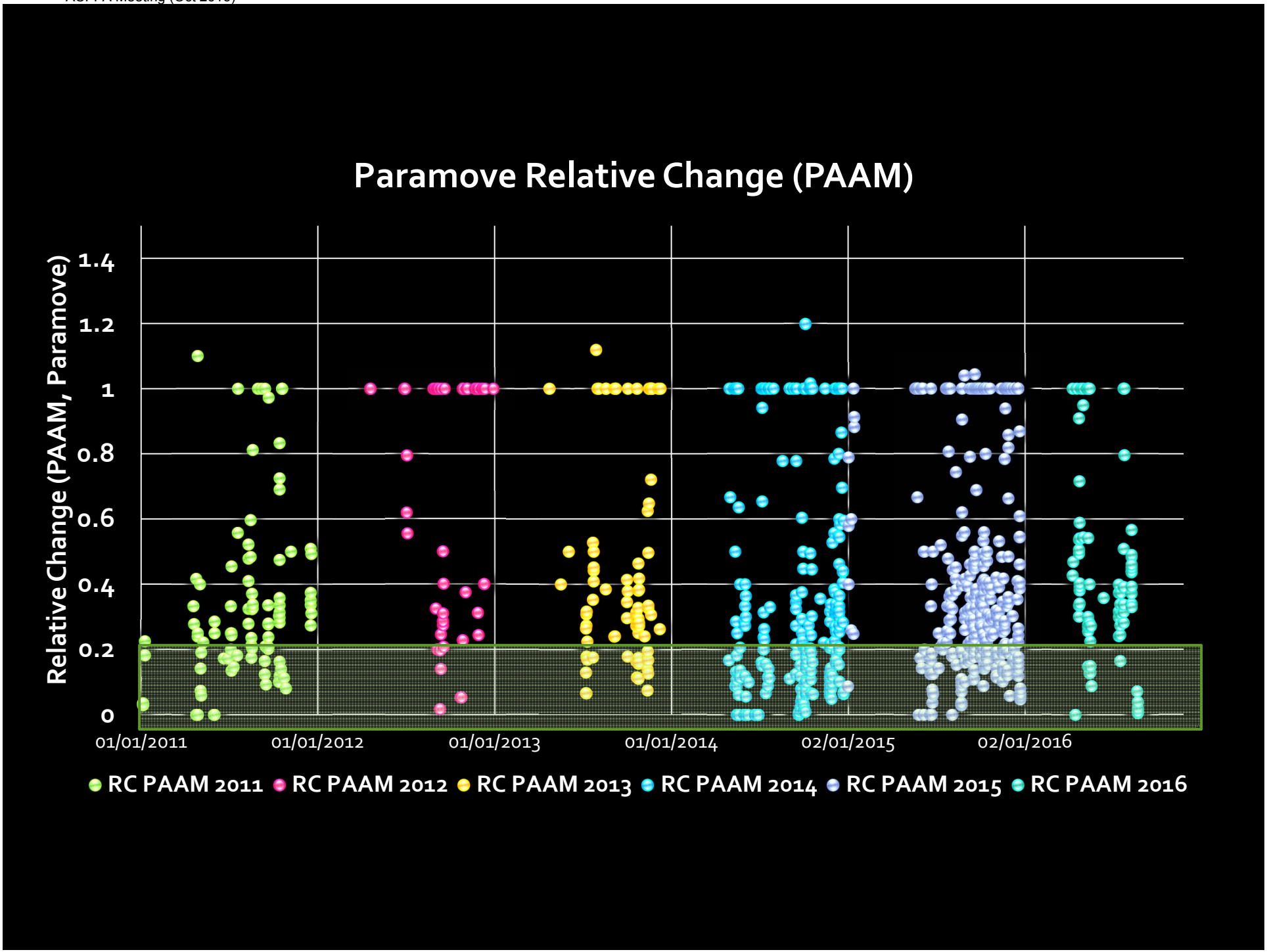

Sea Lice Treatments % of total (NB)
Number of cage treatment events per week
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
2009
2010
2011
2012
2013
2014
2015
2016
week (since Jan 1, 2009)
Alphamax
Salmosan
Paramove

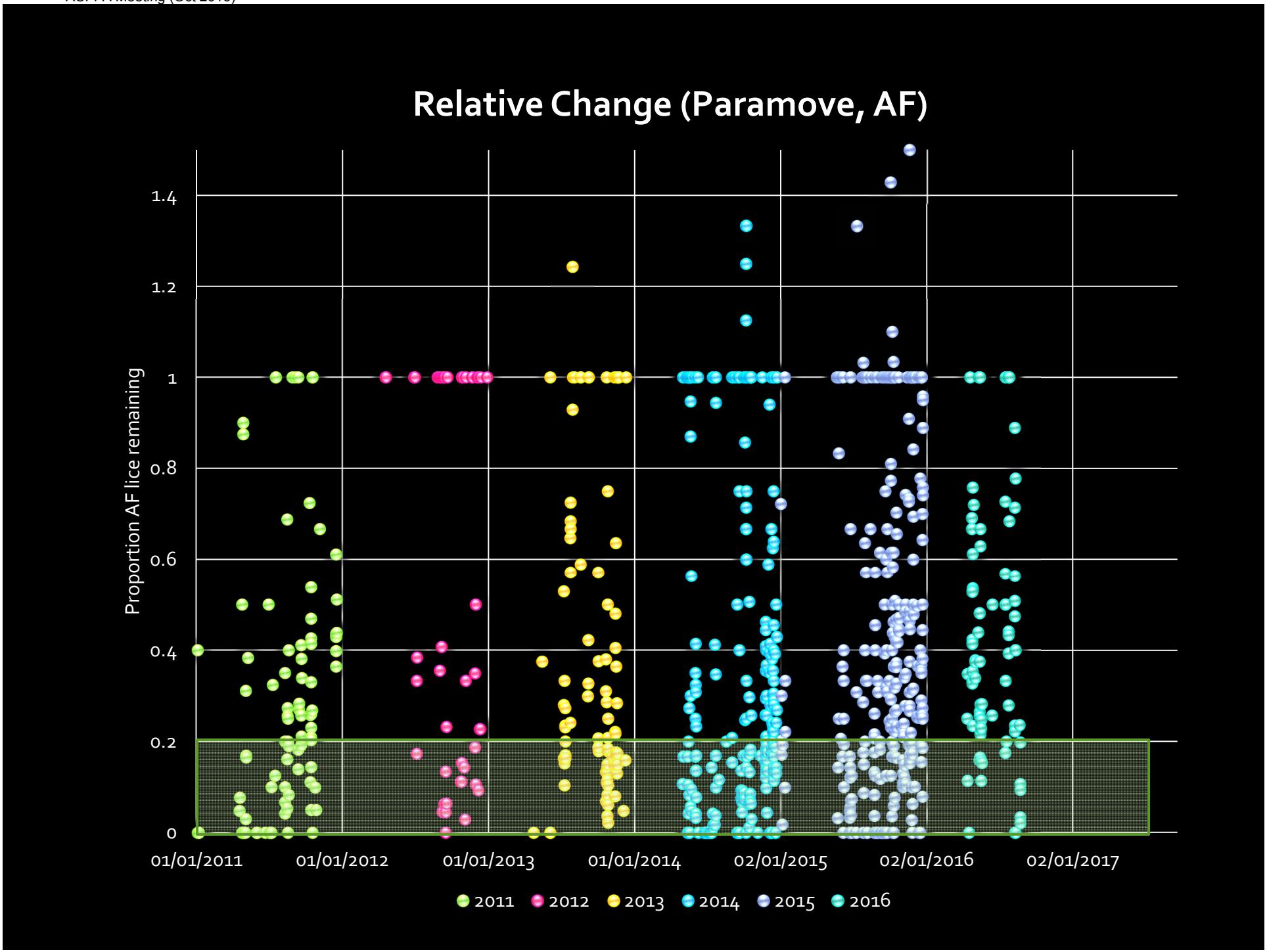
In Feed Treatments (cages)

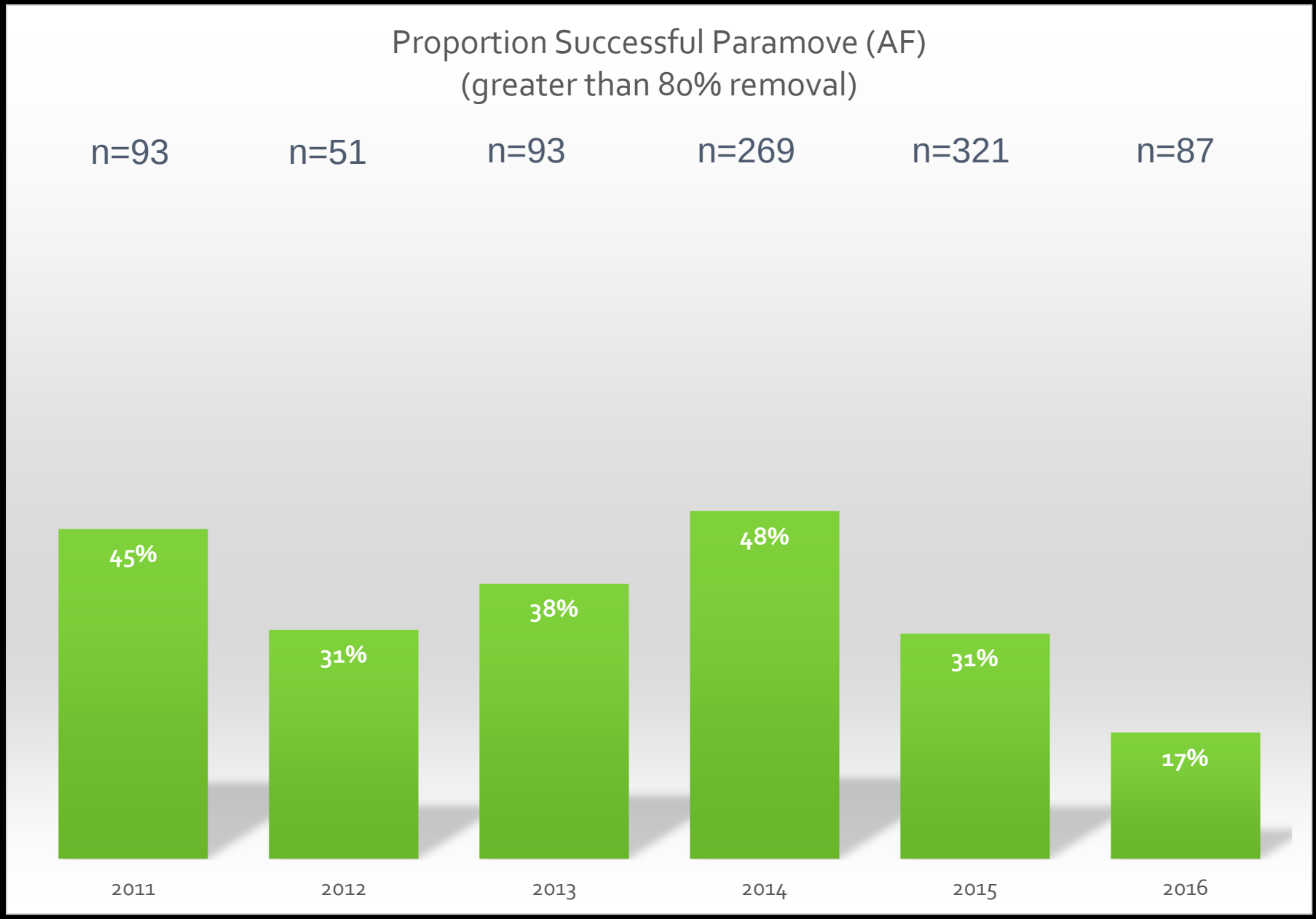



IN FEED TREATMENTS (CAGES)
CAGES TREATED PER WEEK
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
200852
200912
200924
200936
200948
201008
201020
201032
201044
201104
201116
201128
201140
201152
201212
201224
201236
201248
201307
201319
201331
201343
201403
201415
201427
201439
201451
201511
201523
201535
201547
201607
201619
201631
201643
EMB
Calicide
IVM

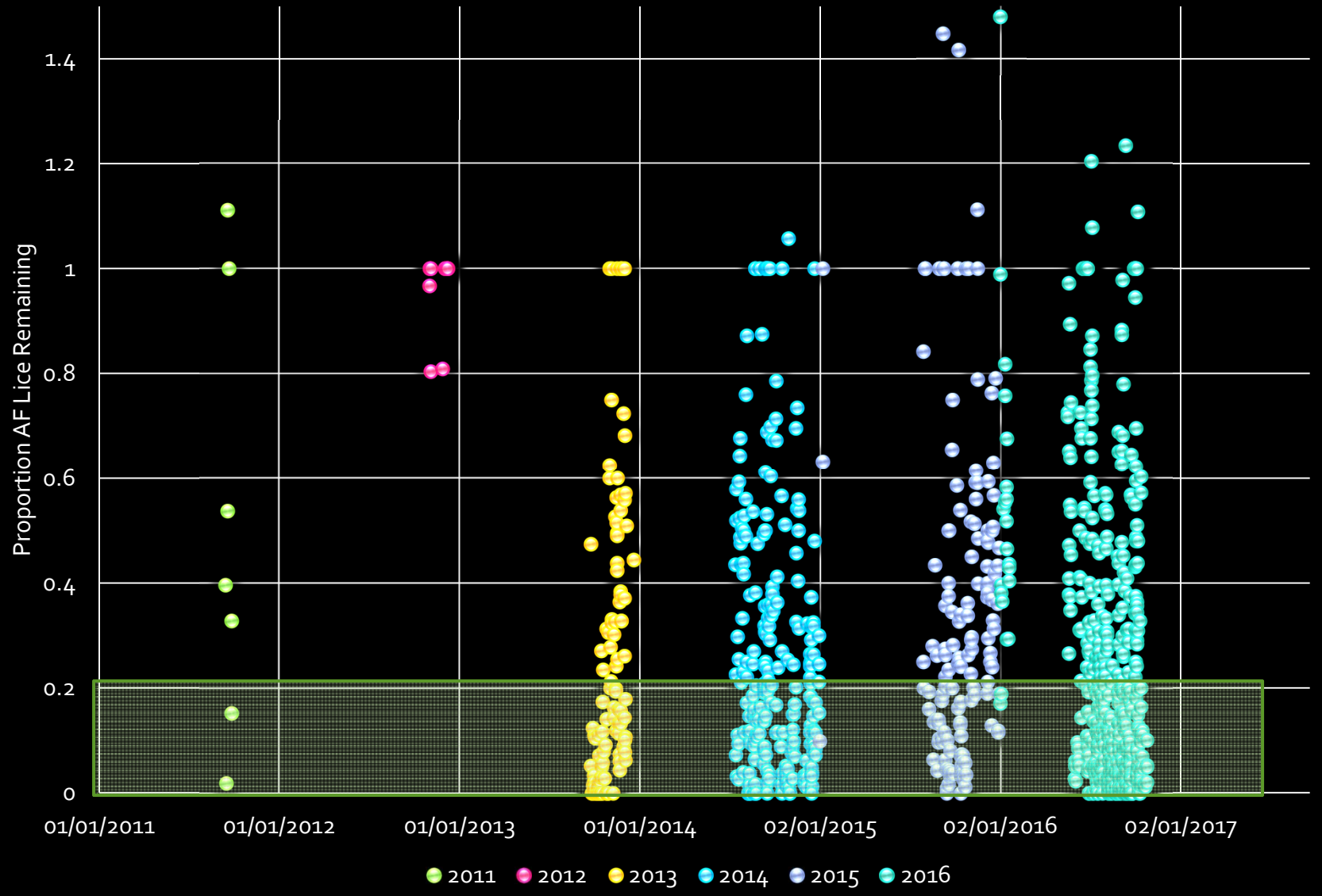

Full Year Lice Knockdown- 2016, Adult Female, ALL BMAs
Alphamax,Salmosan,Paramove/Tarp,Skirt,Well Boat
>80% Knockdown
50-80% Knockdown
<50% Knockdown
Number of Cage(s) Treated
34 32 30 28 26 24 22 20 18 16 14 12 10 8 6 4 2 0 2 4 6 8 10 12 14 16 18 20 22 24 26 28 30 32 34
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43
WEEK OF YEAR

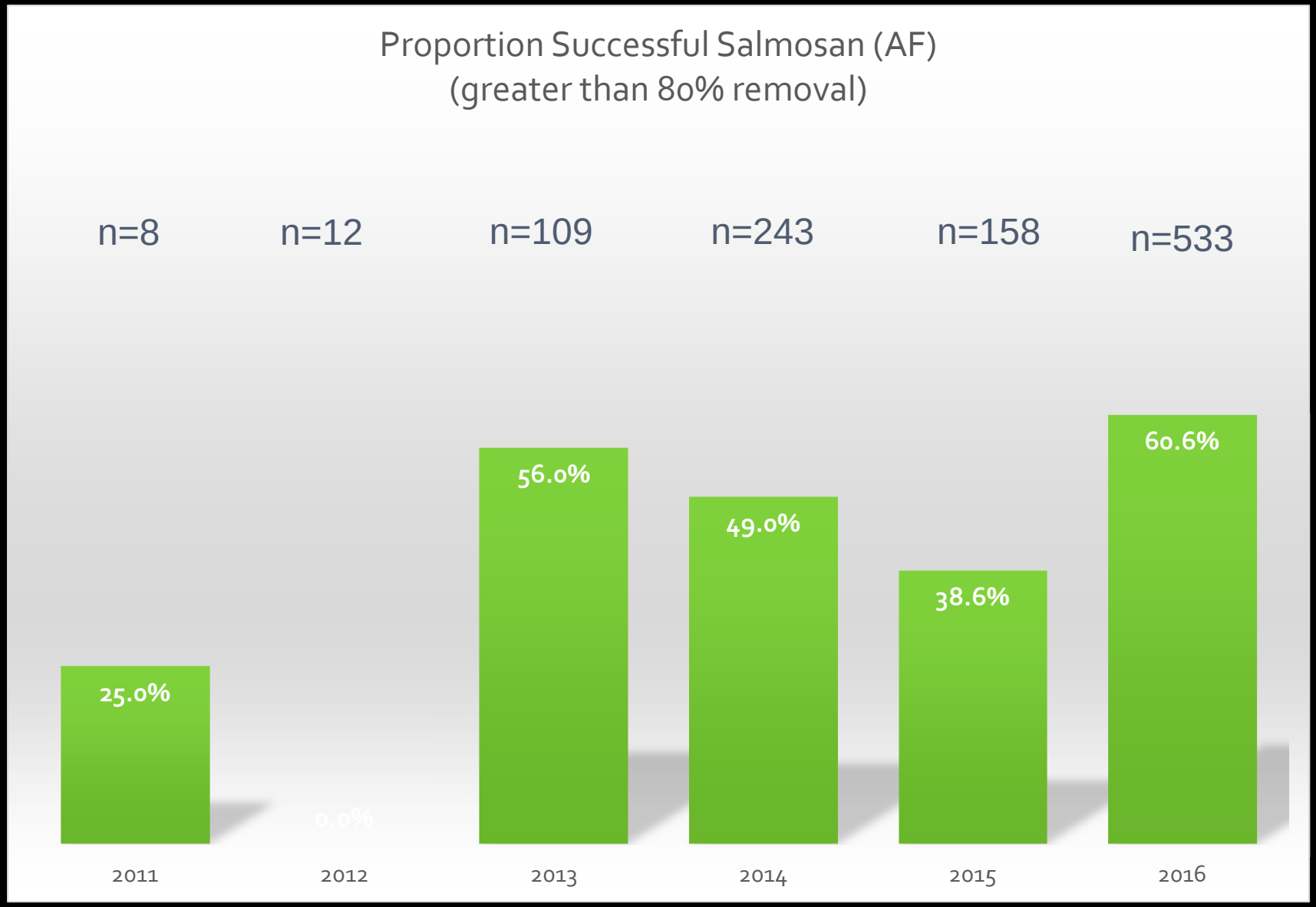

Paramove Relative Change (PAAM)
Relative Change (PAAM, Paramove)
1.4
1.2
1
0.8
0.6
0.4
0.2
0
01/01/2011
01/01/2012
01/01/2013
01/01/2014
02/01/2015
02/01/2016
RC PAAM 2011
RC PAAM 2012
RC PAAM 2013
RC PAAM 2014
RC PAAM 2015
RC PAAM 2016


Relative Change (Paramove, AF)
Proportion AF lice remaining
1.4
1.2
1
0.8
0.6
0.4
0.2
0
01/01/2011
01/01/2012
01/01/2013
01/01/2014
02/01/2015
02/01/2016
02/01/2017
2011
2012
2013
2014
2015
2016

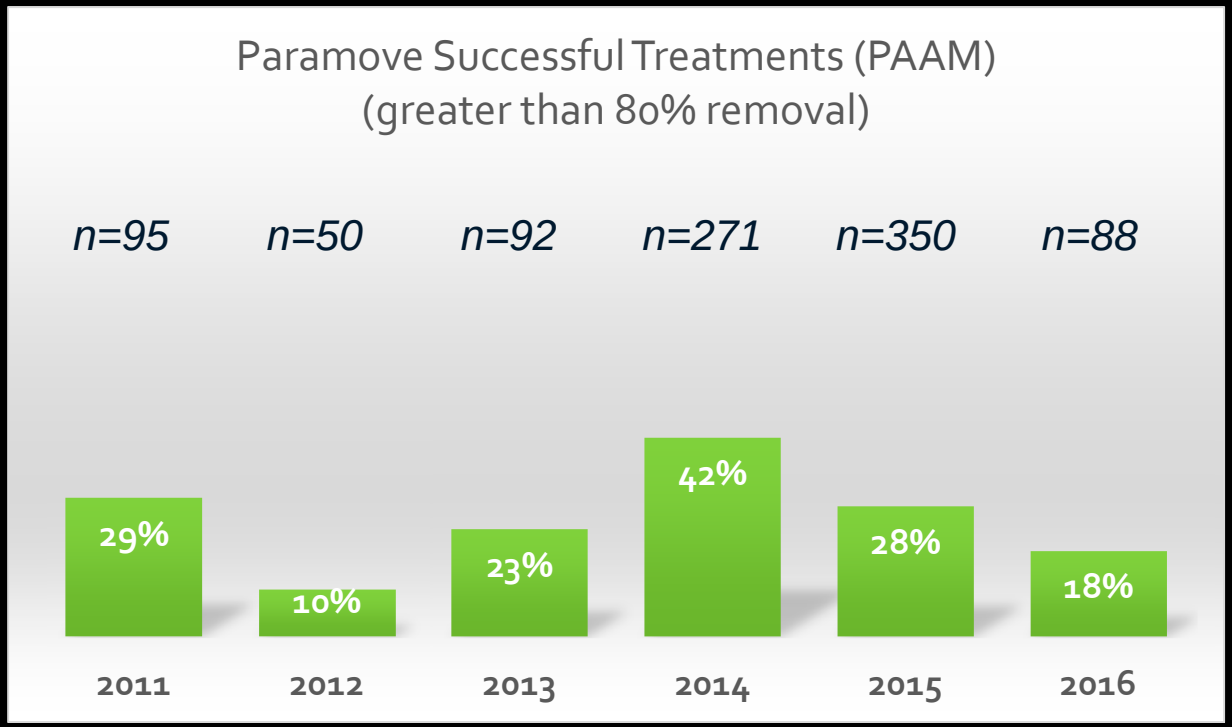

Proportion Successful Paramove (AF)
(greater than 80% removal)
n=93
n=51
n=93
n=269
n=321
n=87
45%
31%
38%
48%
31%
17%
2011
2012
2013
2014
2015
2016

Relative Change Salmosan (AF)

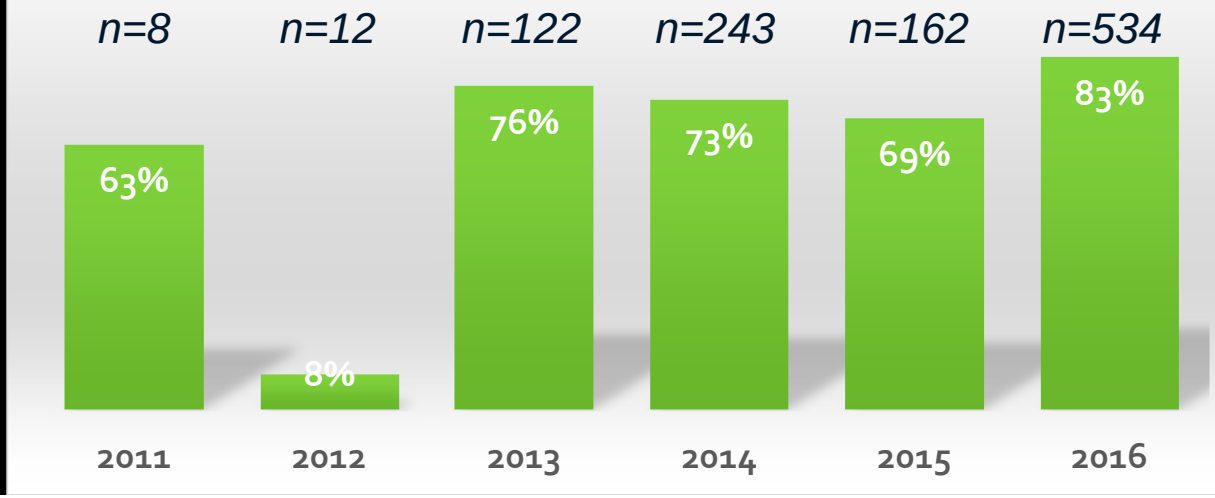


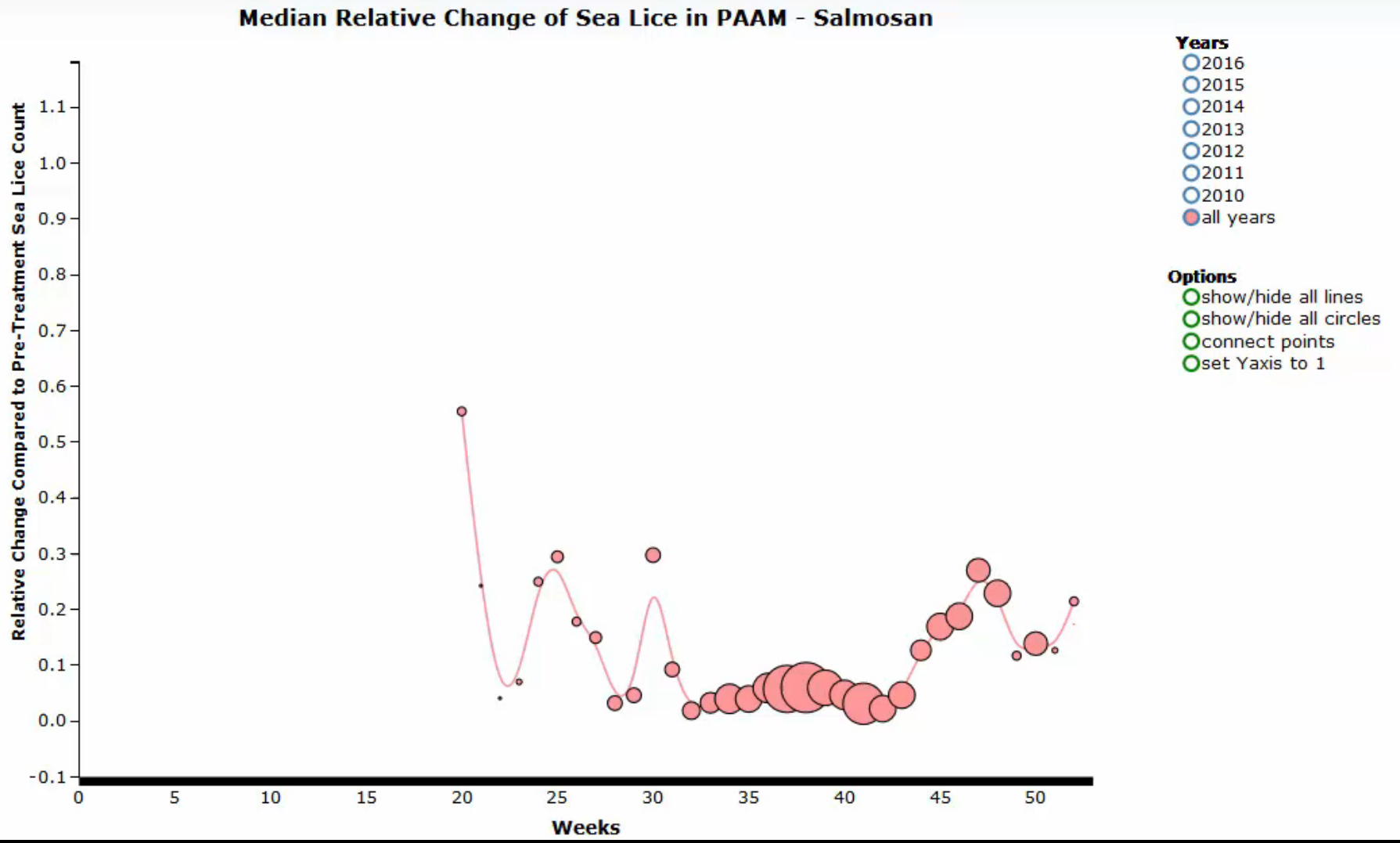

Proportion Successful Salmosan (AF)
(greater than 80% removal)
n=8
n=12
n=109
n=243
n=158
n=533
25.0%
0.0%
56.0%
49.0%
38.6%
60.6%
2011
2012
2013
2014
2015
2016

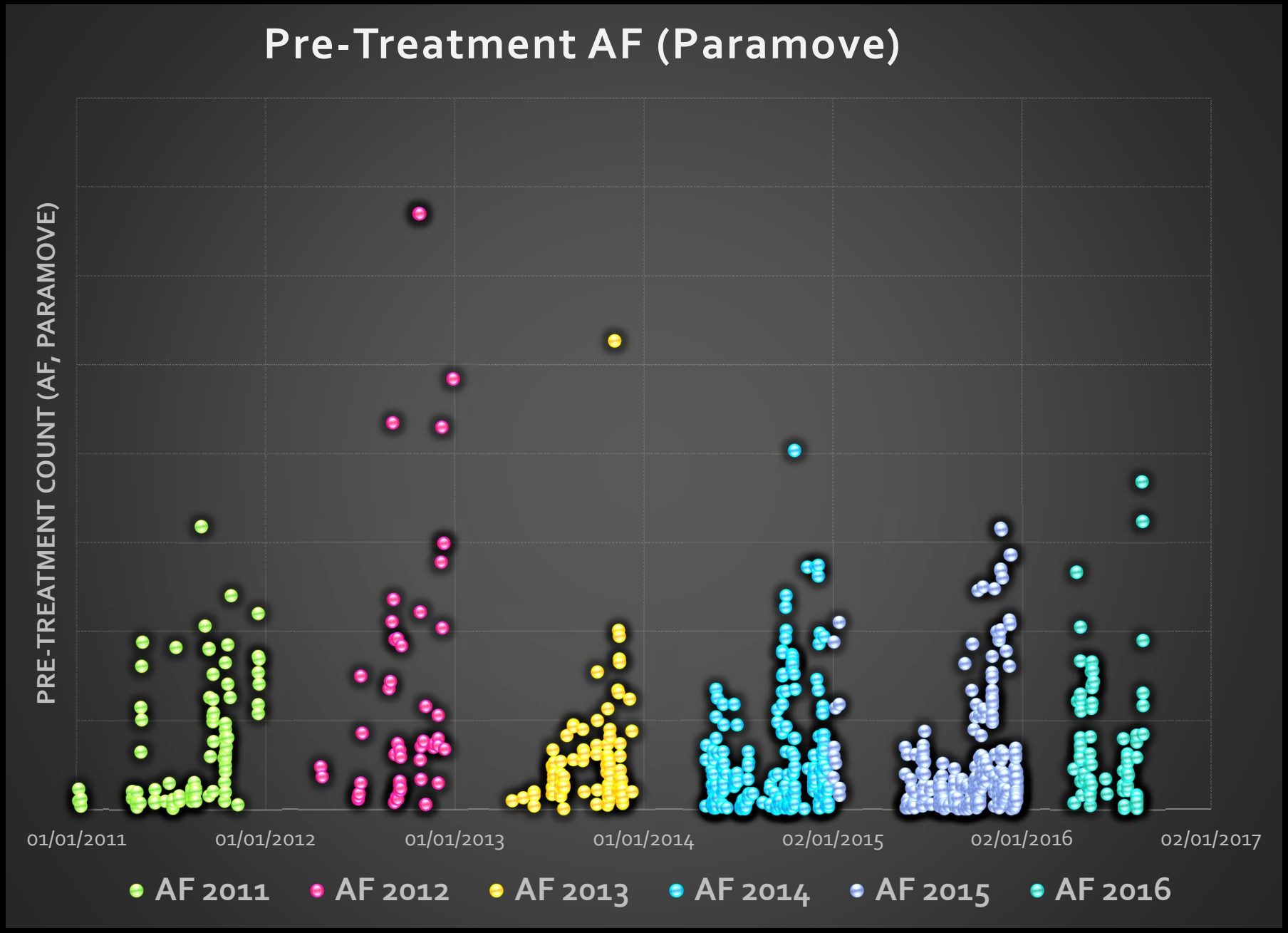
Paramove Successful Treatments (PAAM) (greater than 80% removal)

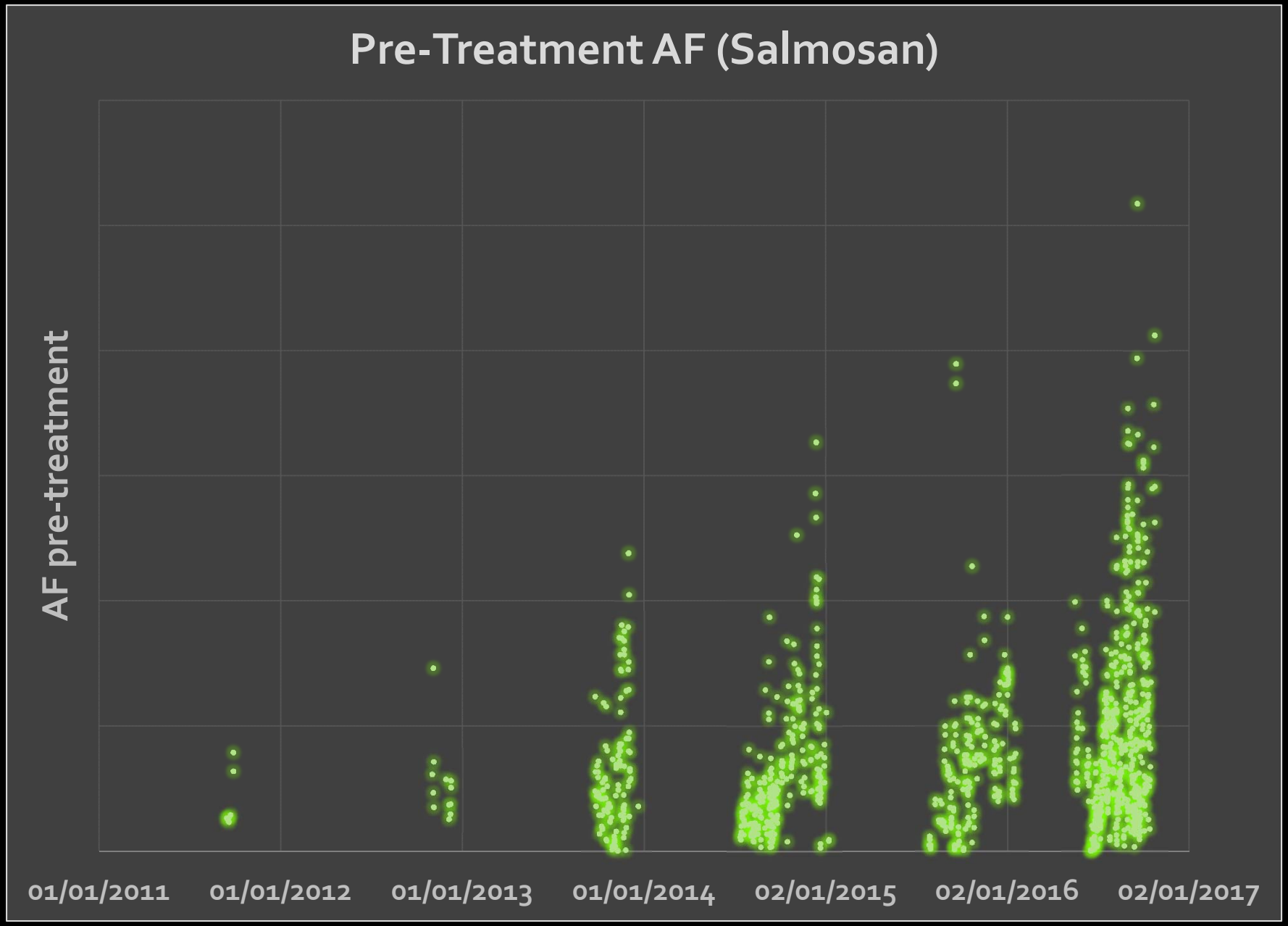


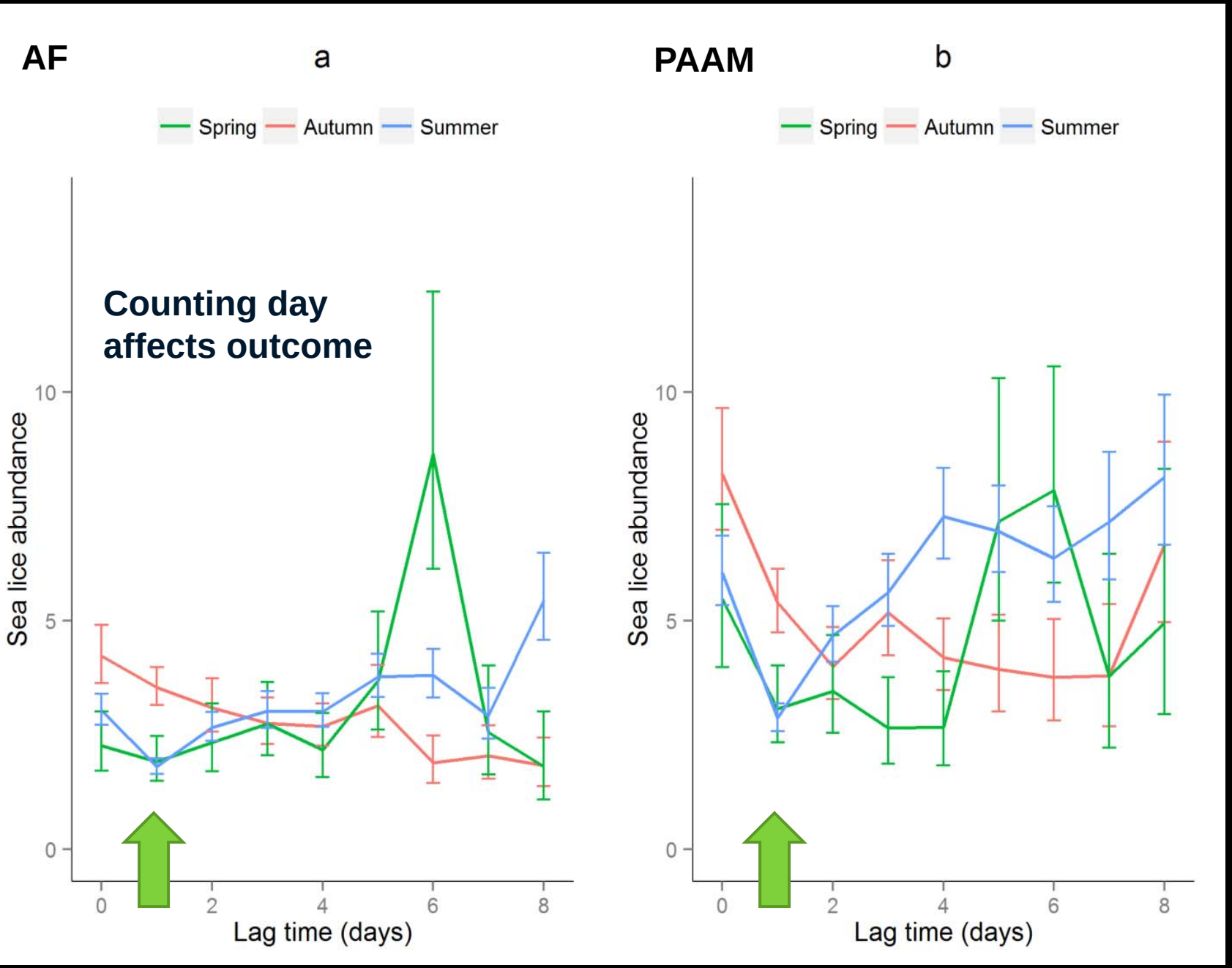
Salmosan Successful Treatments (PAAM) (greater than 80% removal)




Median Relative Change of Sea Lice in PAAM - Salmosan
Relative Change Compared to Pre-Treatment Sea Lice Count
1.1
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
-0.1
0
5
10
15
20
25
30
35
40
45
50
Weeks
Years
2016
2015
2014
2013
2012
2011
2010
all years
Options
show/hide all lines
show/hide all circles
connect points
set Yaxis to 1


Pre-Treatment AF (Paramove)
PRE-TREATMENT COUNT (AF, PARAMOVE)
01/01/2011
01/01/2012
01/01/2013
01/01/2014
02/01/2015
02/01/2016
02/01/2017
AF 2011
AF 2012
AF 2013
AF 2014
AF 2015
AF 2016


Pre-Treatment AF (Salmosan)
AF pre-treatment
01/01/2011
01/01/2012
01/01/2013
01/01/2014
02/01/2015
02/01/2016
02/01/2017



GAUTAM R, VANDERSTICHEL R, BOERLAGE A, REVIE C, **HAMMELL L**. 2016. Effect of timing of count events on estimates of sea lice abundance and interpretation of effectiveness following bath treatments. J Fish Diseases

effectiveness of treatment modality

- influenced significantly by
  - Season
  - pre-treatment level of sea lice
  - lead and lag times
- In summer, Salmosan (tarp) had greatest effectiveness for both AF and PAAM
  - when pre-treatment levels were above 10 sea lice
- in autumn, treatment performance varied significantly
  - Depended on pre-treatment levels (of two life-stages)
- Ignoring lead or lag time effect generally resulted in underestimation of treatment effectiveness

R Gautam et al (in press; J Fish Dis)

Conclusions

- Generally, higher average lice abundance compared to every other year except 2010
- Paramove treatments appear to be less effective in 2016 than previous
  - for PAAM and AF
- Salmosan treatments appear to be more effective in 2016 that previous
  - for PAAM and AF
- Pre-treatment lice count is higher than previous
  - i.e. less aggressive treatment
- Counts should be 1-3 days following bath (not day 0, and not longer than 4 days)

Risk factors for treatment failure in antibiotic treatments against Piscirickettsiosis in farmed Atlantic salmon in Chile

D. Price[1], R. Ibarra[2], J. Sánchez[1], H. Stryhn[1], S. St-Hilaire[1]

[1]*Atlantic Veterinary College, University of Prince Edward Island*

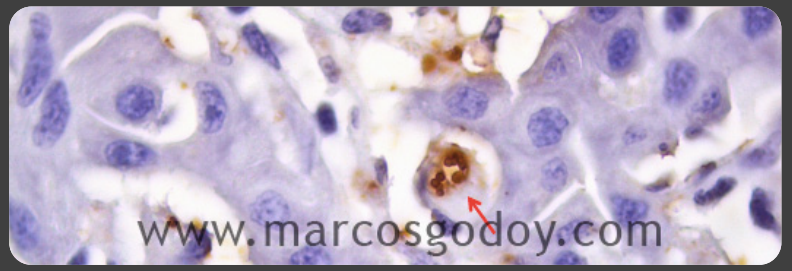
[2]*Instituto Tecnológico del Salmón, Intesal-SalmonChile*

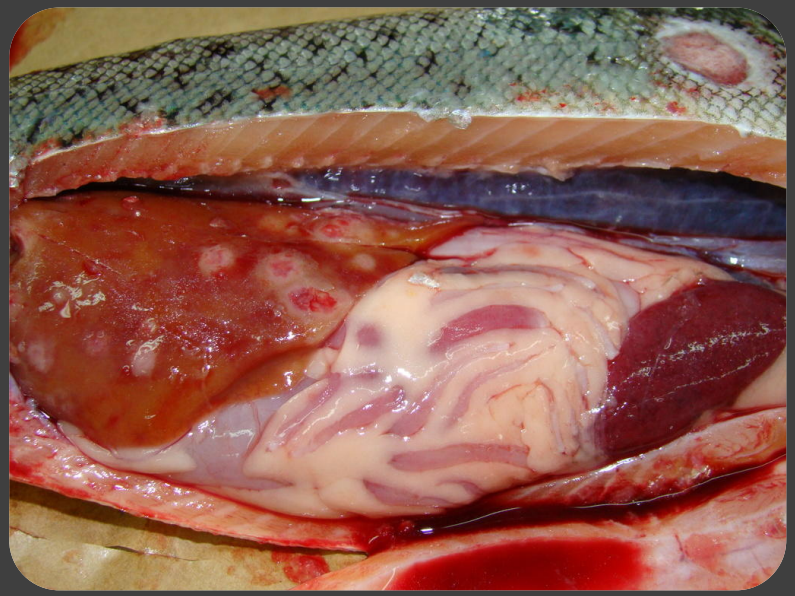
ACFFA Forum – St. Andrews, NB – Oct 2016

UNIVERSITY of Prince Edward ISLAND

Introduction

- *Piscirickettsia salmonis*
  - Gram-negative
  - Facultative Intracellular
- Chronic disease
- All farmed salmonids
- Horizontal transmission
- Main cause mortality





Introduction

- Surveillance and control program since 2009
  - Encourage good husbandry practices
  - Require use of Vaccines
  - Regulate use of Antimicrobials
- 90% of antimicrobial volume
- Treatments have variable success
- Sensitivity studies report low resistance
- Treatment failure multifactorial

Objective

Is the success of an antibiotic treatment dependent on:

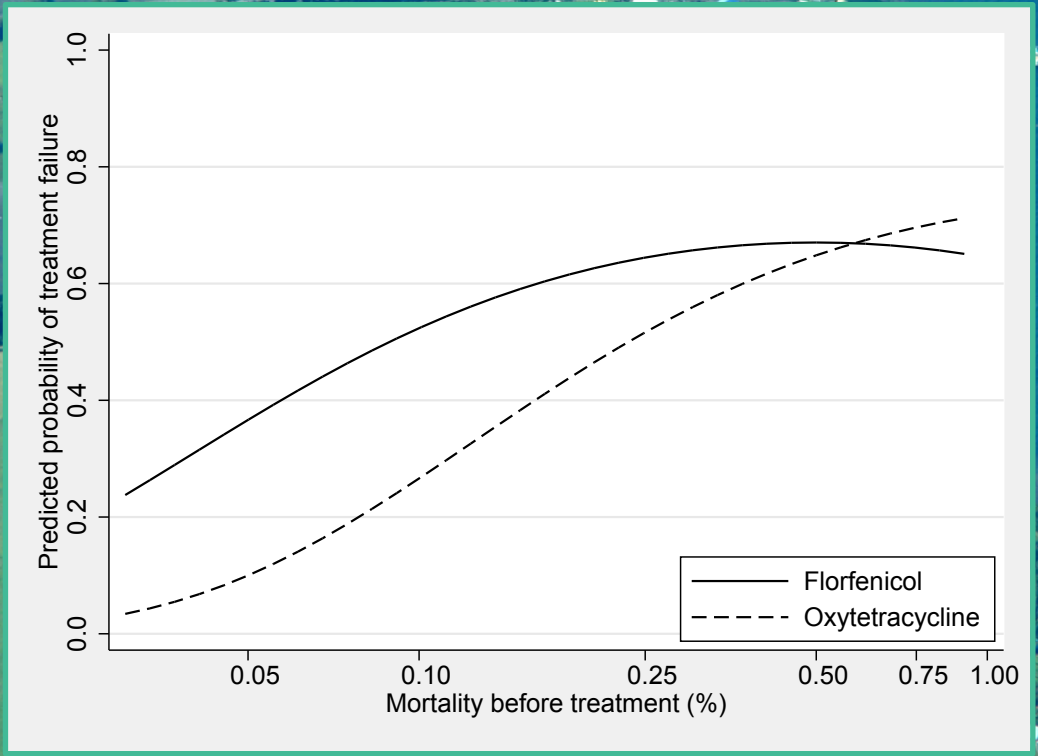
- Antibiotic product used
- Water temperature
- Average fish weight
- Pre-treatment mortality level

Material and Methods

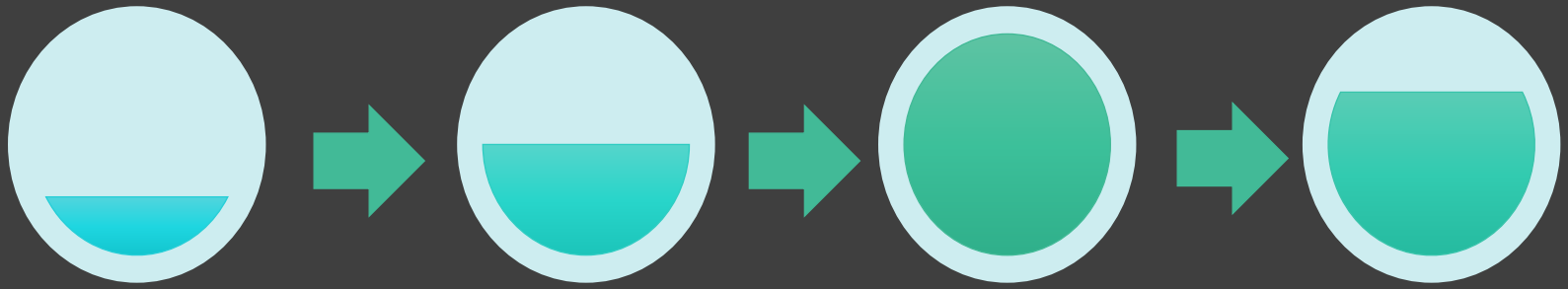
- Intesal-Salmonchile database:
  - 2014 pens on 118 farms
  - 14 Companies
  - First treatment
- Outcome: treatment failure
  - *Weekly mortality above normal level (0.1%)*
  - *~45% failure*
- Mixed logistic model

$$logit(p_i) = \beta_0 + \beta X_i + A_{company_i} + B_{farm_i}$$

Treating early leads to higher success rates



Pre-treatment mortality is related to stage of the disease in the population



Susceptible Stage

- Exposure

Subclinical Disease

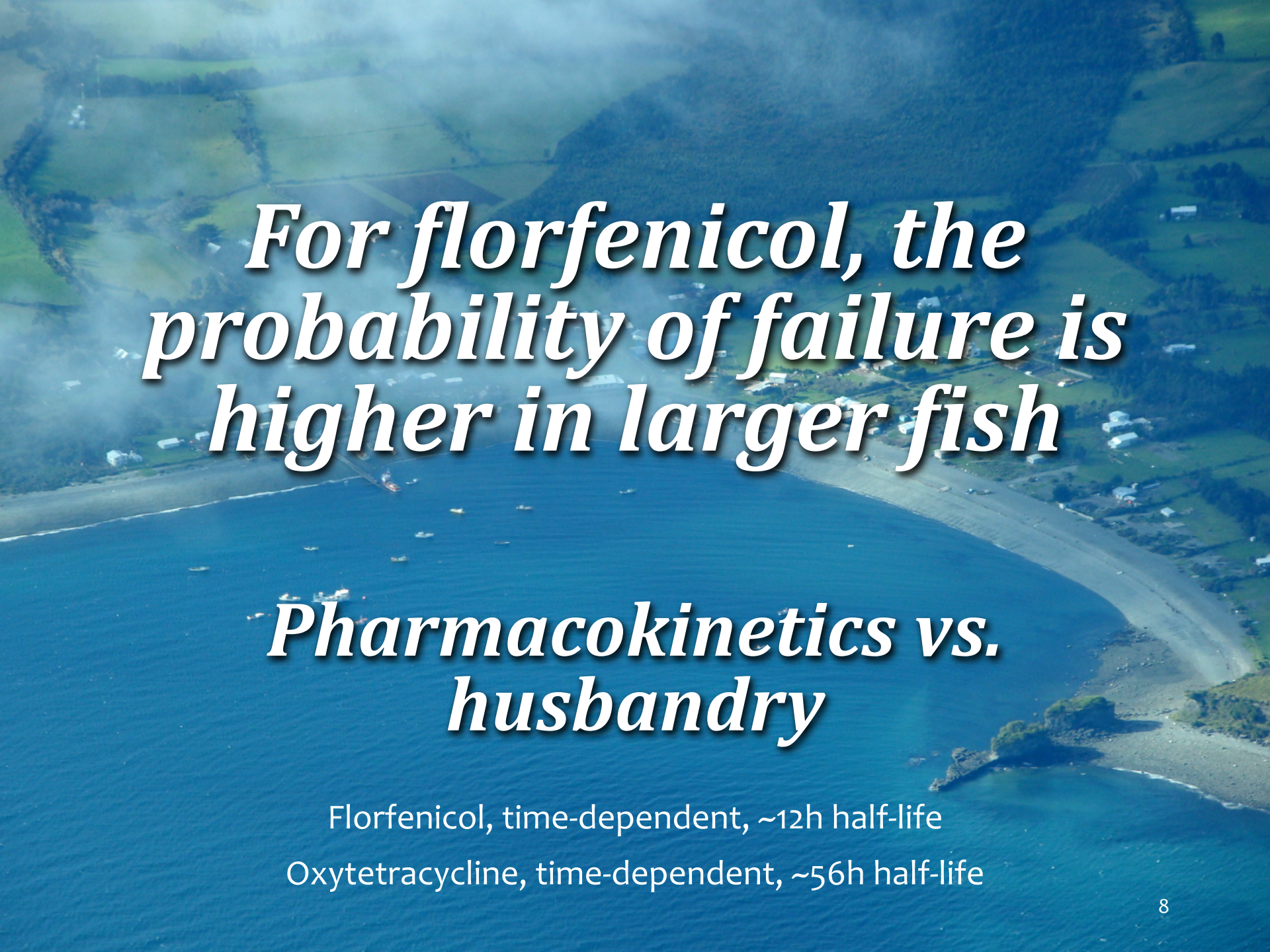
- Pathologic Changes

Clinical Disease

- Onset of Symptoms (Anorexia)
- Usual time of diagnosis
- Start of treatment

Recovery Period

- Recover
- Carriers
- Death



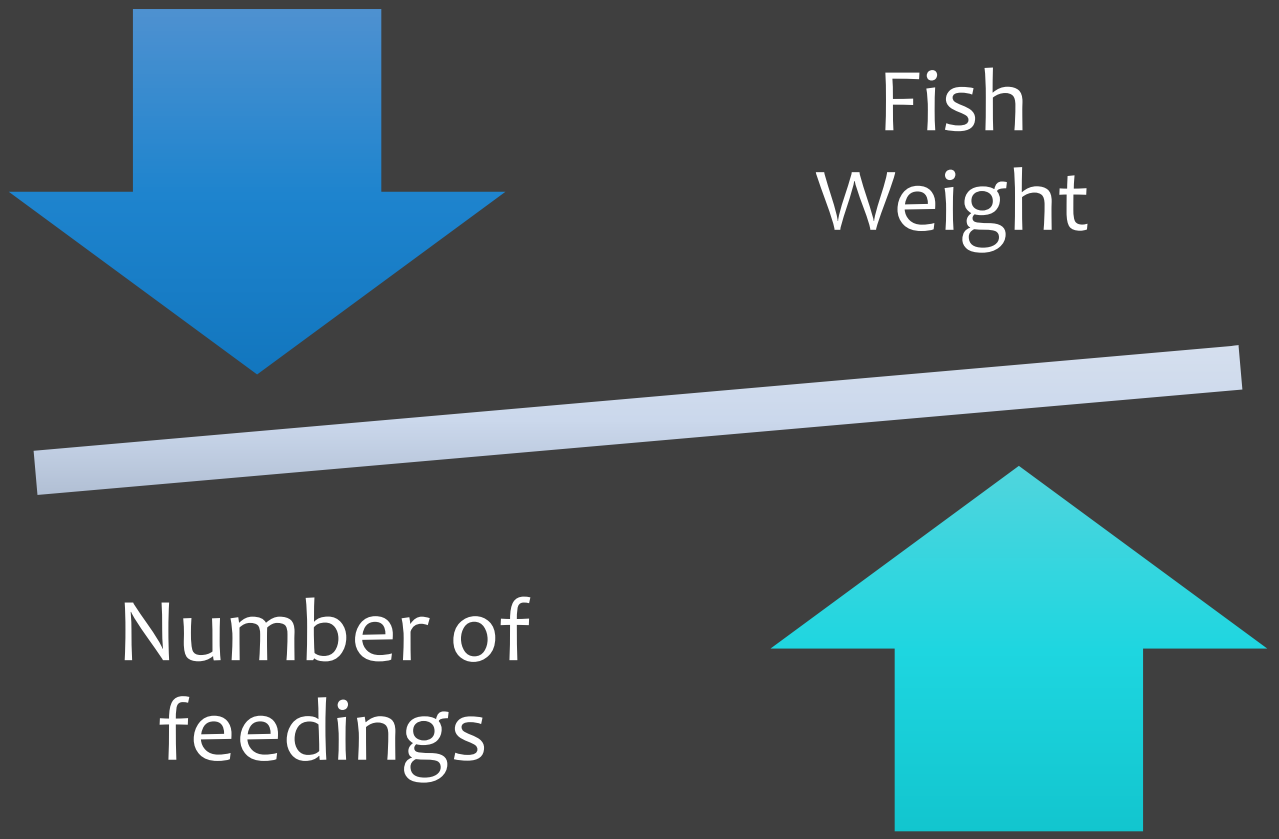
For florfenicol, the probability of failure is higher in larger fish

Pharmacokinetics vs. husbandry

Florfenicol, time-dependent, ~12h half-life

Oxytetracycline, time-dependent, ~56h half-life

Number of feeding events are reduced to minimize competition for food



Antibiotic tissue concentration during oral treatments

Preliminary findings

Objective

- Assess the level and variation of antibiotics in tissues during treatment
- Determine factors associated with “adequate” antibiotic tissue levels

Material and Methods

Outcome:

AB concentration above Epidemiological cut-off (ECOFF) values for *P. salmonis* in Chile (Henríquez et al, 2016)

- 2 µg/ml florfenicol
- 4 µg/ml oxytetracycline

Dataset:

- ~2600 tissue samples (NQC)
- ~100 treatment events
- 35 Farms

Conclusions

- Treating early may reduce treatment failure
- Wide distribution of tissue concentration
- High proportion of individuals below ECOFF
- Within a population, body condition explain variation
  - Just increasing dose may lead to extreme high values (long withdrawal)
  - Improve feed distribution

Future research

- Assess effect of feeding frequency in antibiotic tissue concentration and treatment success
- Antimicrobial sensitivity surveillance programs to assess the role of resistance

Acknowledgements

- Intesal-SalmonChile
- Canada Excellence Research Chair in Aquatic Epidemiology

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An Update on the Epidemiology of Ulcer Disease

Brett MacKinnon, DVM, MSc student

Ulcer disease in Canada

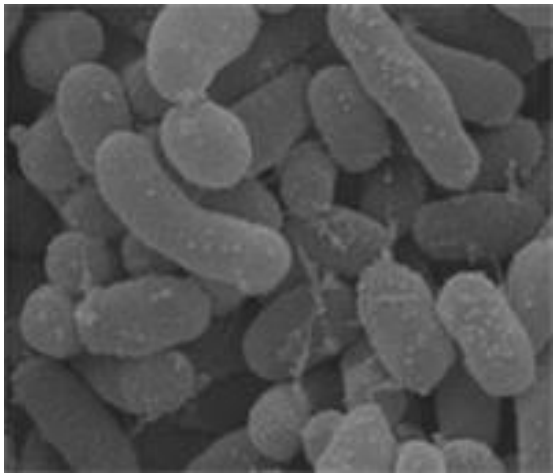
- Atlantic salmon
- Outbreaks starting in summer

http://brage.bibsys.no/xmlui/bitstream/id/252768/Masteroppgave2014Martin%20S%C3%B8rgaard.pdf

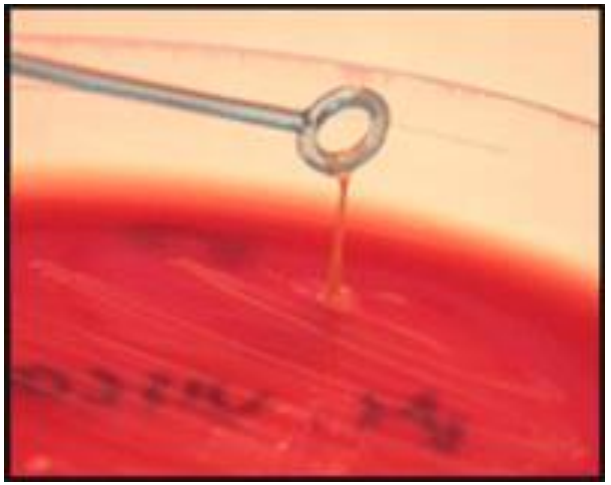
Ulcer disease in Canada

What's causing ulcer disease?

- Hypothesized to be caused by *Moritella viscosa*
  - ➢ Difficult to isolate from field samples
- Winter Ulcer Disease in Europe
- European *M. viscosa* isolates produce ECP that can cause necrosis and hemorrhages of tissue



http://www.thefishsite.com/fishnews/11002/norwegian-researcher-sheds-light-on-winter-ulcer/



http://www.thefishsite.com/fishnews/11002/norwegian-researcher-sheds-light-on-winter-ulcer/

Ulcer disease control difficulties

- Environmentally endemic
- Antimicrobial therapy - in feed
- Current vaccines do not always prevent this disease

Our research at AVC

- The epidemiology of ulcer disease of Atlantic salmon in Canada

  1) Analysis of industry data

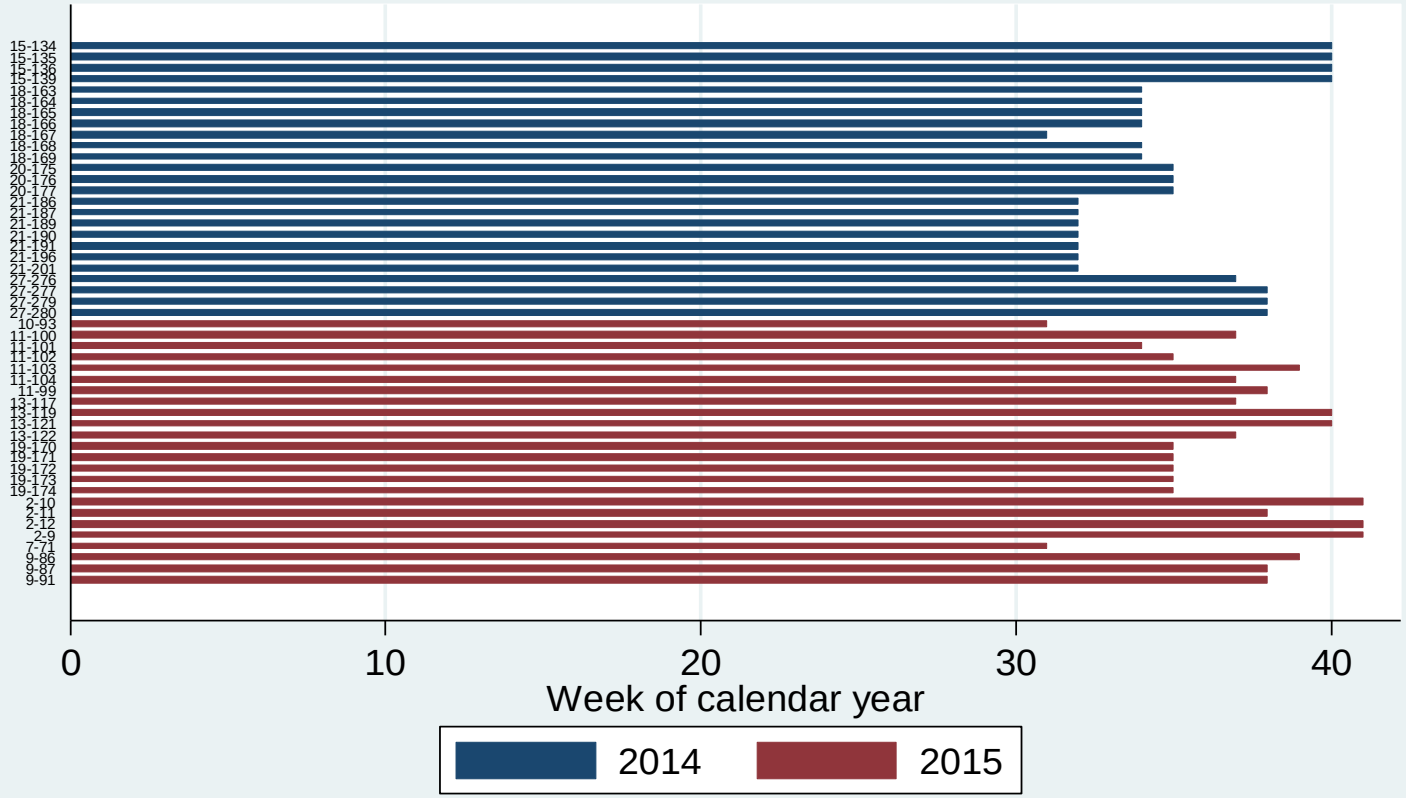
  2) Investigate ulcer disease caused by Atlantic Canadian *Moritella viscosa* isolate under laboratory conditions

  - Preliminary results

Descriptive epidemiology

- Objectives: Investigate patterns, hypothesize type of exposure
- Pen-level mortality and medical records (2014-2016)
  - Timing of outbreaks
- Cage-level & farm-level analysis of the onset, magnitude, and duration of outbreaks

Cage-level analysis: onset of outbreaks



- Outbreaks start within ~3wks of each other
- Common point exposure

Figure 1. Week of disease onset at the cage-level during 2014 and 2015.

Cage-level analysis: magnitude of outbreaks

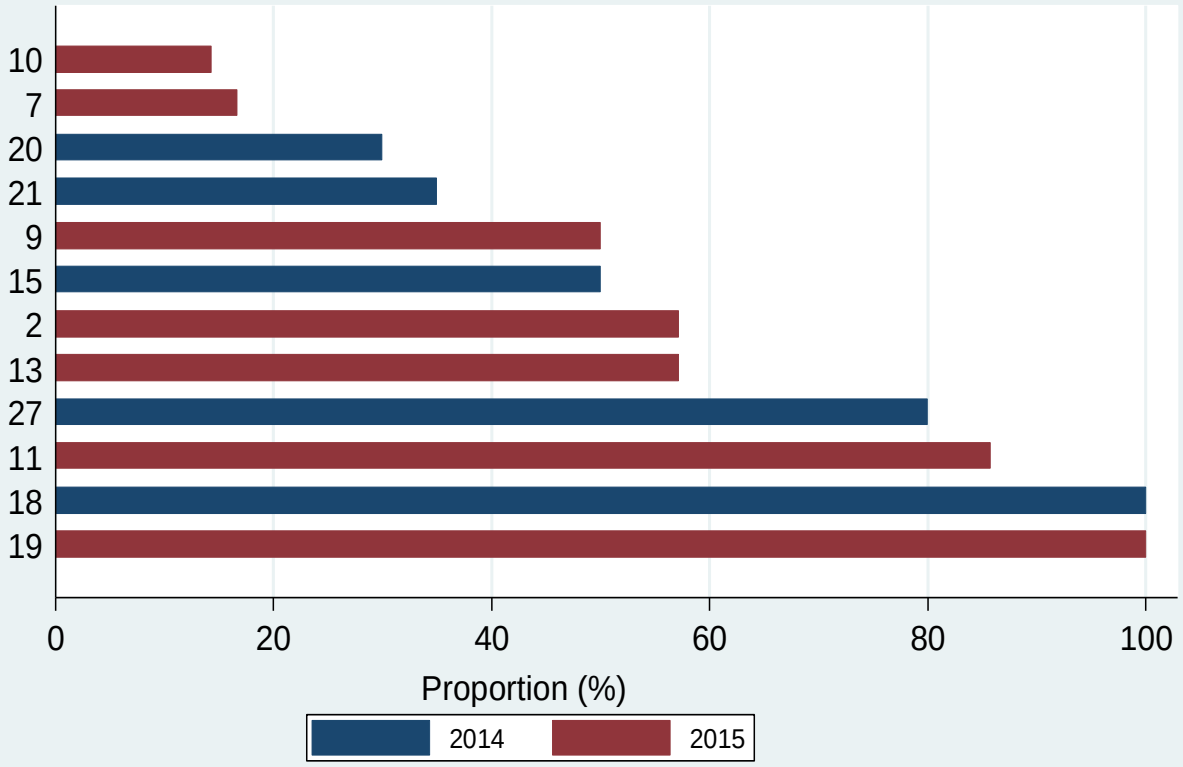


Figure 2. Proportion of cages on farms affected by ulcer disease.

- 8 to 100%
- Not consistent with cage-to-cage transmission

Cage-level analysis: magnitude of outbreaks

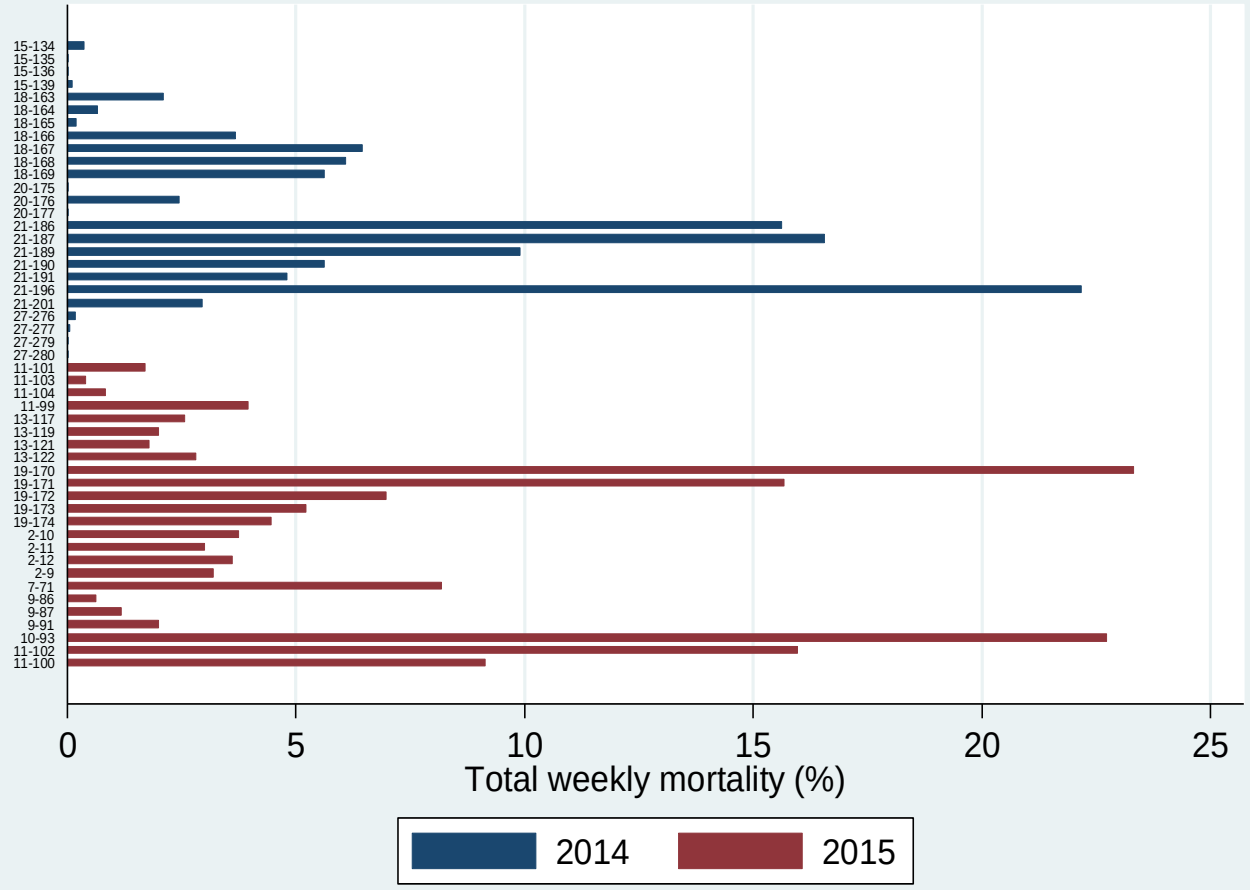


Figure 3. Total mortality during ulcer disease outbreaks at the cage-level.

- 0.006 to 23.3%

Farm-level analysis: duration of outbreaks

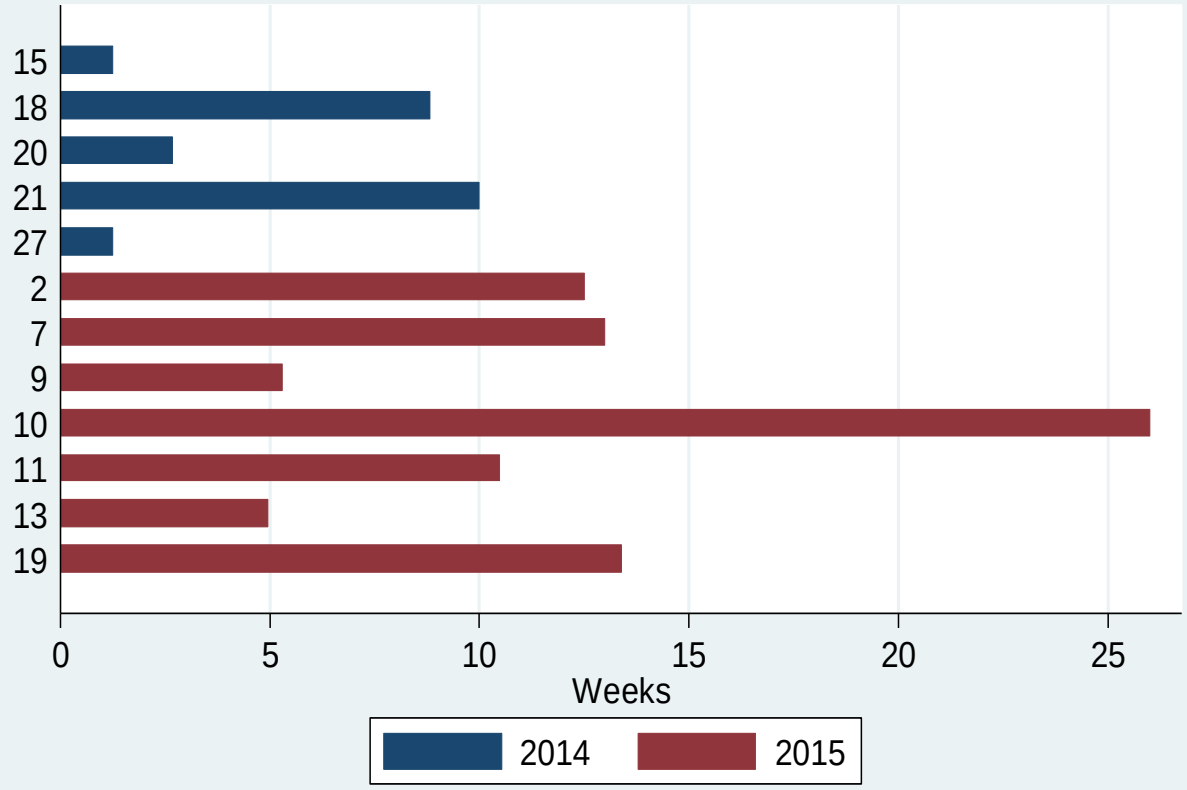


Figure 6. Average duration of ulcer disease outbreaks at the farm-level.

- 2014 – 1 to 10 wks
- 2015 – 5 to 26 wks

Variation in magnitude and duration of outbreaks

- Exposure to pathogen is not uniform on farm

  and/or

- Factors may be associated with severity of disease
  - i.e. cage density, predation stress, timing of treatments…

Farm-level analysis: onset of outbreaks

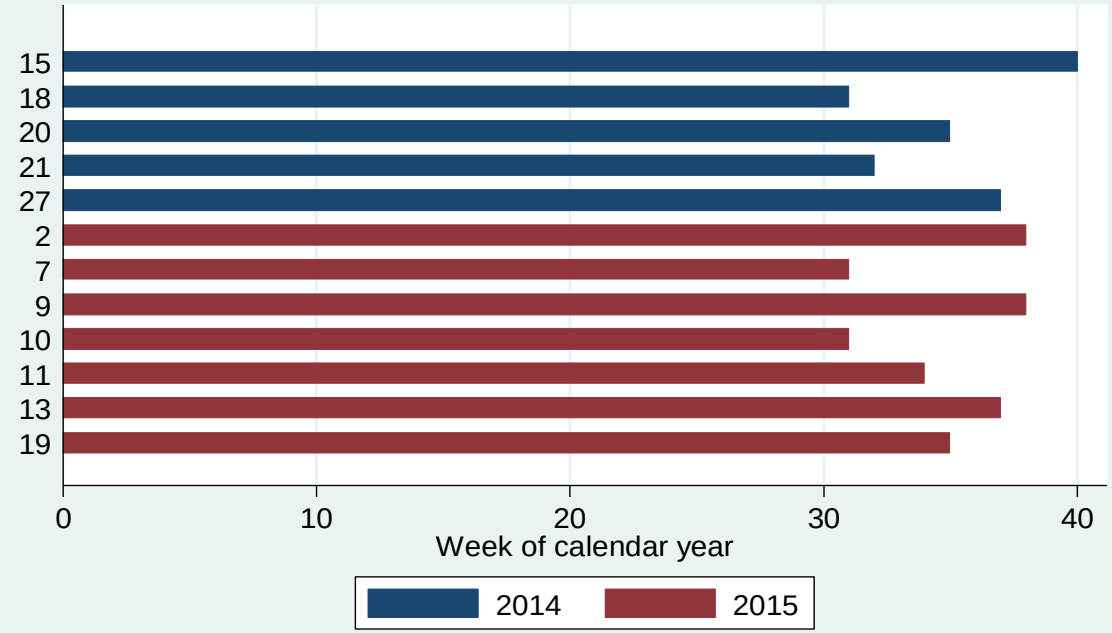


Figure 7. Week of ulcer disease onset for farms.

- 29 Farms total
- 41% +ve overall
- 2014 – 29%
- 2015 – 58%
- Similar dx pattern
  - ➢ Late July to mid-fall

- Pathogen present from late July to mid-fall

  vs

- Pathogen present but outbreaks triggered by environmental factor

Descriptive epidemiology summary

- Pathogen is widespread at specific time of year (late July to mid-Fall) and/or present during other times of year, but outbreaks triggered by environmental factor
- Common point exposure to the pathogen across farms
- Pathogen is not uniform on the farm (non-uniform exposure) and/or certain factors reduce or increase probability of disease expression and severity
- Cage-to-cage transmission may not always occur

Lab Experiment 1

- **Determine if ulcer disease can be transmitted horizontally between tanks**

- Describe the progression of ulcer disease and disease pathology
  - *Moritella viscosa* isolate from Atlantic Canada
  - Mimic field conditions (11°C)

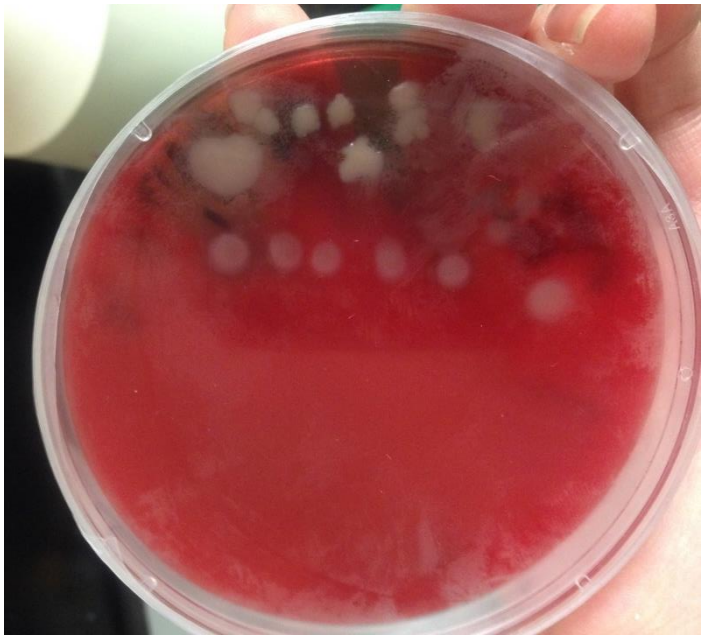
Lab Experiment 1

- Group 1 exposed to *M. viscosa* (in broth) by bath immersion
- Group 2 negative control group
- Group 3 tanks receive water from Group 1 tanks
- Group 4 positive control group

Lab Experiment 1

Lab Experiment 1

- Fish were sampled over time to follow progression of disease
  - Necropsy, bacteriology, histo, biochem, PCR, etc



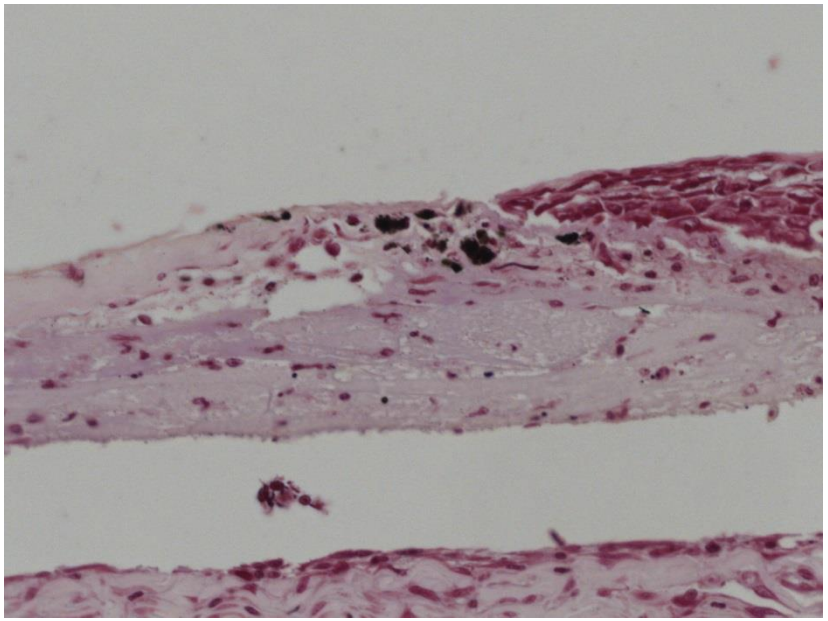
Lab Experiment 1

- Lasted for 26 days
- Typical lesions of ulcer disease for fish infected via bath immersion
  - No clinical signs in neg control tanks
  - Lesions/mortality in pos control tank
- All fish remained on feed except for a few with most severe lesions

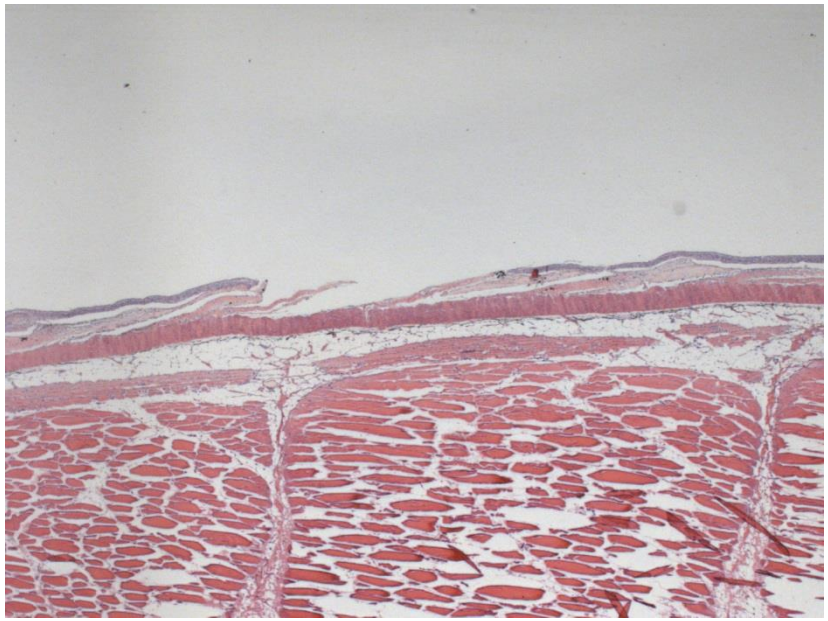
Lab Experiment 1

- No horizontal transmission of disease
  - ➢Consistent with descriptive epidemiological findings
- Recovery of fish with mild lesions

Histology of field lesion



Gram stain, 40X



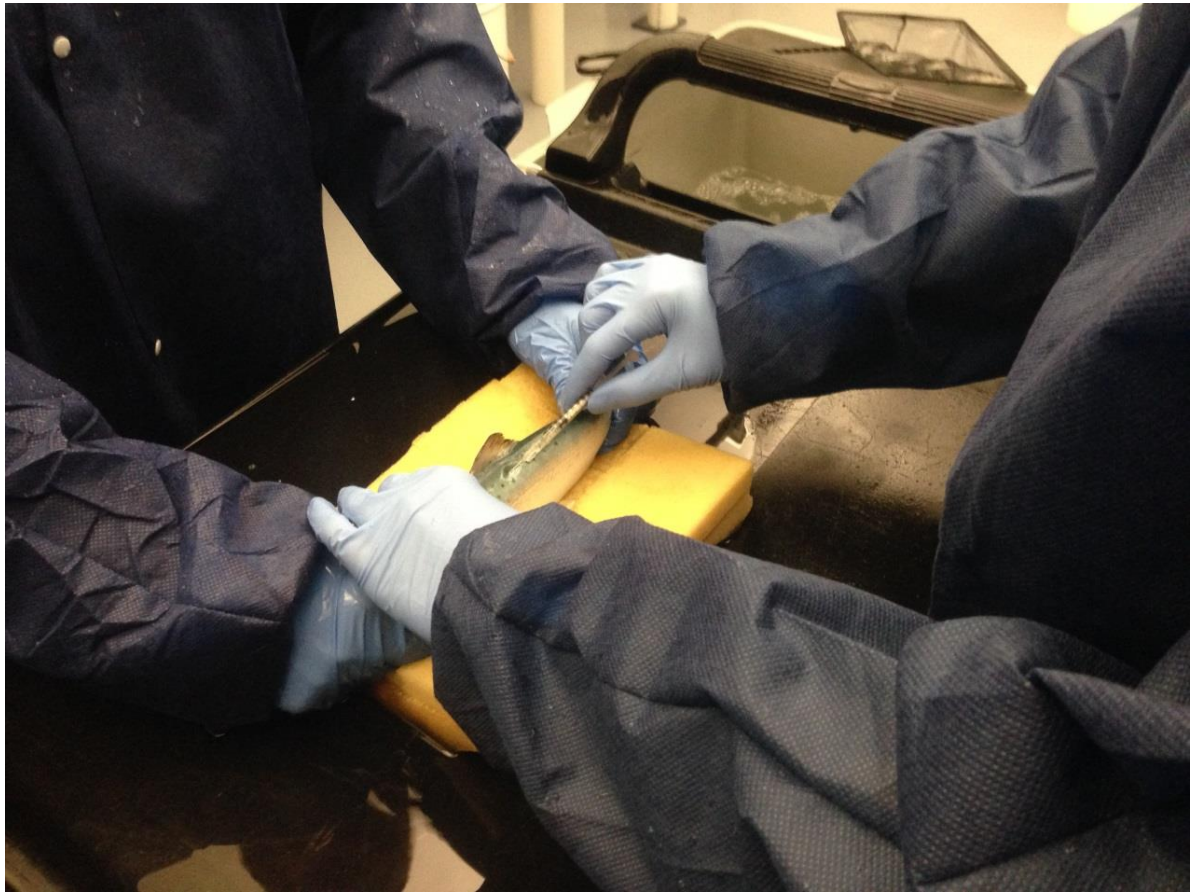
H & E stain, 10X

Lab Experiment 2

- **Determine whether ulcer disease lesions can be induced with extracellular products (ECP) produced by *M. viscosa***

Lab Experiment 2

- Group 1 injected SQ with *M. viscosa* ECP in broth
- Group 2 negative control (injected SQ with broth)
- Group 3 negative control



- Monitored fish for evidence of ulcers and sampled over time

24 hours post-injection

Day 8

Days 12 to 17

- Ulcers!

Experiments summary

- Can induce ulcer disease with Atlantic Canadian *M. viscosa* isolate at 11°C
- No horizontal transmission of *M. viscosa* under laboratory conditions
  - Consistent with descriptive epidemiological findings
- The ECP of *M. viscosa* causes necrosis/swelling/hemorrhage/ulcers of Atlantic salmon tissue under laboratory conditions

Acknowledgments

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