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ST. JOHN'S CONVENTION CENTRE
ST. JOHN'S, NEWFOUNDLAND
AND LABRADOR, CANADA



AQUACULTURE CANADA AND WAS NORTH AMERICA 2022



AQUACULTURE
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AQUACULTURE
Society



NAIA WELCOME MESSAGE

Greetings from NAIA Executive Director
Jamie Baker

Welcoming the Aquaculture World to Newfoundland and Labrador

Welcome one and all to Aquaculture Canada and WAS North America 2022 Conference and Trade Show. On behalf of the event organizers — the NL Aquaculture Industry Association (NAIA), Aquaculture Association of Canada (AAC) and the World Aquaculture Society (WAS) — it is my pleasure to welcome you all to the “Far East of the Western World”: Newfoundland and Labrador!

After three years of planning and delays due to the pandemic, the time is finally upon us to host the ocean farming world here in the province. It’s been a difficult time for us all on many levels these past few years, but we find ourselves now in what might be one of the most exciting times in our sector’s history.

This year’s conference and trade show will bring together the best and brightest from the global aquaculture community in the farming, service/supply and regulatory sectors for an event that will celebrate the achievements of the sector, look at the challenges, and continue planning a vision for the future.

If the pandemic taught us anything it is that enhancing food security is a huge challenge for our growing populations and continuing to provide healthy food responsibly and sustainably is going to be critical. Our trade show and many side events will provide a strong business networking and partnership platform that will help further these discussions and grow the sector. Our program sessions will feature some of the most influential people in the aquaculture world in talking about the challenges and opportunities ocean farming presents in a modern world. Our social events will allow the industry to interact on a personal level with key political and practical partners to help enhance the vision we are collectively building. In short, you will find out why, as we say all the time here in this province, the ocean farming sector is “Bringing so Much to the Table.”

In closing, I’d like to thank the many members of our organizing committee, committee chairs, organizations and staff for their continued hard work and perseverance in pulling together this incredible event. It’s been a tireless effort, but a worthwhile one and you are all worthy of praise for the work that’s been done.

I hope everyone enjoys our conference, trade show, many social events and presentations as well as the culture and history the city of St. John’s and the province of Newfoundland and Labrador have to offer.

Jamie Baker
Executive Director, NAIA
Steering Committee Co-Chair, Aquaculture Canada and WAS North America 2022

AAC WELCOME MESSAGE

On behalf of the Aquaculture Association of Canada, it is my pleasure to welcome AAC, WAS, and NAIA members, as well as all conference delegates and participants, to Aquaculture Canada and WAS North America 2022 Conference and Tradeshow, here in historic St. John's, Newfoundland and Labrador.

Since 2019, the three co-hosts of this event have persevered through much change and uncertainty to offer you an in-depth program, complimented by various networking events to dive back into face-to-face meetings. Coordinating an international meeting to share important aquaculture research and developments, as well as showcase new and emerging technologies, after such a long hiatus was no small feat. With a sell-out tradeshow to accompany the scientific presentations and social events, delegates will find their days (and nights) busy as we make acquaintances with new and old colleagues.

Our conference theme: *Aquaculture – The Leading Edge of Food Production*, recognizes aquaculture's increasing importance in providing a healthy, sustainable food choice. And what better place to celebrate aquaculture than St. John's, on the very edge of North America? I challenge all delegates to learn something new, make new connections, reconnect with colleagues and embrace our industry of which we can be so proud.

I would like to thank all current and past Steering Committee members for their time and energy over the past three years – particularly Dr. Kurt Gamperl, the Conference Program Chair, Dr. Jay Parsons the Steering Committee Chair, and our AAC team of Conference Manager, Joanne Burry and Office Manager, Miranda Pryor as well as past-Presidents Joanne Liutkus and Matt Liutkus. Thanks also to our Newfoundland and Labrador contingency of Dr. Jillian Westcott, Cyr Couturier, and Danny Boyce. Fellow association members John Cooksey and Dr. Michael Schwarz from WAS, Jamie Baker, Darrell Green, and Roberta Collier from NAIA, and former NAIA Executive Director Mark Lane were instrumental in making this Canada's largest ever aquaculture conference and tradeshow.

I would also like to thank you, the delegates, for your participation. The pandemic forced us into virtual meetings, the home office, and bubbles. To once again be in person with our fellow colleagues, learning about the latest in aquaculture innovations, welcoming students to the aquaculture sector, and networking late into the evening, is long overdue and is to be enjoyed and to be thankful for.

I wish you all a successful conference – please take in all the program has to offer, and enjoy yourselves in the beautiful, charismatic city of St. John's.

Kim Gill
President, Aquaculture Association of Canada

WAS WELCOME MESSAGE

On behalf of the **World Aquaculture Society**, it is my pleasure to welcome you to the Aquaculture Canada and World Aquaculture Society North America 2022 Conference and Trade Show in St. John's, Newfoundland and Labrador. This meeting is a unique collaboration among the Aquaculture Association of Canada (AAC), the Newfoundland Aquaculture Industry Association (NAIA), and the World Aquaculture Society (WAS North America).

It is absolutely wonderful to be able to meet in person once again. The challenges of organising in-person meetings over the last several years notwithstanding, the organising committees have put together a wonderful program of speakers and posters on topics of regional, national and international interest. There are eight to nine concurrent sessions each of the three days, with 300 oral presentations, 60 posters, a sold out trade show and three outstanding plenary speakers (Mr. Tavish Scott, Dr. Stefanie Colombo, and Dr. Sandra Shumway). There will be speakers, sessions and exhibitors that capture the interest of all participants.

A meeting of this size and scale does not happen on its own. The combined efforts and dedicated hours of many individuals have contributed to the development of a successful and varied science program, trade show, and social programs. A huge thanks to AAC (Kim Gill, Joanne Burry, Miranda Pryor, Jennifer Wiper), NAIA (Jamie Baker, Mark Lane, Roberta Collier, Darrell Green, Jackie Richards), WAS (John Cooksey, Michael Schwarz, Mario Stael, Noah Cooksey), MUN (Kurt Gamperl, Cyr Couturier, Jillian Westcott, Danny Boyce) and many others who helped with the local arrangements, the section chairs, and the many session organisers and chairs.

Aquaculture conferences such as the Aquaculture Canada and World Aquaculture Society North America 2022 Conference and Trade Show are important events for sharing the latest on the science, innovations, and technologies that underpin the development of a sustainable and key seafood farming industry in Canada and internationally. This meeting will truly be an opportunity to network with a broad range of participants, including researchers, students, growers, suppliers, communicators, managers, regulators, etc., in the Canadian and international aquaculture sector that is literally and figuratively on “The Leading Edge of Food Production”. Learn, share, and contribute as fully as you can!

Thank you all for attending, and enjoy your experience in this historic and culturally-unique part of Canada. Profitez de votre séjour à Terre-Neuve-et-Labrador.

G. Jay Parsons, PhD
Steering Committee Co-Chair (WAS)

TABLE OF CONTENTS

NAIA WELCOME.....	2
AAC WELCOME.....	3
WAS WELCOME.....	4
AQUACULTURE CANADA & WAS NORTH AMERICA 2022 ABSTRACTS	7

To find abstracts for a specific author or subject, use the pdf search features built into Adobe Acrobat.

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AQUACULTURE CANADA AND
WAS NORTH AMERICA 2022

ABSTRACTS

MEASURES OF MARKET STRUCTURE AND SOCIO-ECONOMIC CHARACTERISTICS OF FISH AND SHRIMPS MARKETERS IN AKWA IBOM STATE, NIGERIA

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This study assessed the measure of market structure and socio-economic characteristics of fish and shrimps marketers in Akwa Ibom State, Nigeria. Akwa-Ibom State has an official population figure of about 3.9million (NPC, 2006). It has an area of 7,081km² with its Atlantic Ocean Coastline stretching to 129km from Oron in the East to Ikot-Abasi in the West. The State is located in the Coastal Southern part of Nigeria, lying between latitudes 4°32'N and 5°33'N; and longitudes 7°25'E and 8°25'E. It is bordered on the East by Cross River State, on the West by River State and Abia State, and on the South by the Atlantic Ocean. Multistage sampling technique was used to collect primary data from 90 respondents in the study area. Descriptive statistics, Concentration Ratio were used to analyze the set objectives.

The result of the socio-economic characteristics of the respondents revealed that 53% were within the age range of 41 to 50 years. About 54% of the respondents were male while 46% represent female. The result showed that 97% of the respondents were married. Also, respondents with household size of 1 to 5 persons were recorded to be 83%. Moreover, 57% of the respondents had primary school education implying that they were literates (Table1).

From the result of the market concentration, the Gini Coefficient was 0.492 as presented in Table 2. This implies that there is inequality of shares of income in the study area. It is therefore recommended that, associations/organizations that will promote free market day without market tax to improve competitive level in seafood marketing should be encouraged. Hence, reduced oligopoly.

Table 1: Socio-economic Characteristics of Respondents

Age (Years)	Frequency	Percentage
41-50	48	53.3
others	42	46.7
Total	90	100
Gender		
Male	49	54.4
Female	41	45.6
Total	90	100.0
Marital Status		
Married	87	96.7
Others	3	3.3
Total	90	100.0
Household Size		
1-5	75	83.3
others	15	16.7
Total	90	100.0
Educational Attainment		
Primary	51	56.7
Others	39	43.3
Total	90	100.0

Table 2: Summary of Concentration Indexes

Index	Symbol	Formula Used	Value
Concentration Ratio	CR	$CR = \frac{\sum_{i=1}^i CR_i}{\sum_{i=1}^i CR_n}$	CR ₂ = 7% CR ₄ = 24% CR ₆ = 33%
Gini Coefficient	GC	$GC = A / (A+B)$	0.492

Source: Computed from Field Survey Data, 2018.

TOXICITY EFFECTS OF SMOKED AND UNSMOKED FILTERED CIGARETTE BUTTS LEACHATE ON AFRICAN CATFISH (*Clarias gariepinus*)

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In this study, the toxicity of smoked and unsmoked filtered cigarette butts of various brands was conducted on *Clarias gariepinus*. The mean bodyweight of the fish was 4.72g. Leachate of cigarette butts was prepared by soaked 24 smoked (4.21g) and unsmoked (4.82g) cigarette butts in six litres of water for 24 hours. The leachate was serially diluted to obtain concentration of 8 cigarette butts per litre (8CB/L), (6CB/L), (4CB/L), (2CB/L), (1CB/L), (0.5CB/L), (0.25CB/L) and (0CB/L). Two hundred and forty (240) *C. gariepinus* of 21 days old were randomly exposed to the 8 different concentrations with ten fishes per treatment and replicated 3 times. T1 was exposed to 0CB/L, T2 (0.25CB/L), T3 (0.5CB/L), T4 (1CB/L), T5 (2CB/L), T6 (4CB/L), T7 (6CB/L) and T8 (8CB/L). The experiment was carried out for 96 hours. The result shows that the mortality of *C. gariepinus* increased with an increase in concentrations of the cigarette butts and time of exposure. Table 1 shows that there was 100% mortality in T8, T7 and 36.7, 33.3, 16.7, 13.3, and 6.7% mortality was observed in T6, T5, T4, T3, and T2 respectively after 96 hours of exposure while there was no mortality in the control treatment (T1). Table 2 shows the results of LC_{50} and LC_{95} . LC_{50} value of 0.57 and LC_{95} value of 2.30 were obtained after 96 hours of exposure. The result of the physico-chemical water quality shows that there was no significant difference ($p>0.05$) in the mean temperature, dissolved oxygen and salinity of the leachate but there was a significant difference ($p<0.05$) in the mean pH among the treatments. This study shows that cigarette butts leachate is toxic to freshwater fish.

Table 1: Result of the percentage mortality of *C. gariepinus* response to different concentration of cigarette butts leachate

T	% Mlty at 24 hrs	% Mlty at 48 hrs	% Mlty at 72 hrs	% Mlty at 96 hrs
T1	0	0	0	0
T2	0	0	0	6.7
T3	0	0	10	13.3
T4	0	6.7	13.3	16.7
T5	6.7	16.7	23.3	33.3
T6	6.7	23.3	30.0	36.7
T7	100	100	100	100
T8	100	100	100	100

T (Treatment) Mlty (Mortality), hrs. (Hours)

Table 2: lethal concentrations of the cigarette butt leachate.

Time (Hrs)	LC_{50} (95% CL)	LC_{95} (95% CL)	Probit Equation	DF	TF
24	7.12 (3.47 - 0.90)	40.41 (2.32 - 1.40)	$Y=1.81+5.36X$	7	1.00
48	1.99 (2.1 - 0.46-)	6.76 (3.73 - 3.04)	$Y=3.37+2.19X$	7	3.58
72	0.96 (4.15 - 1.88)	3.41 (2.09 - 1.72)	$Y=2.06+4.41X$	7	7.41
96	0.57 (1.39 - 0.84)	2.81 (4.21 - 3.63)	$Y=2.91+3.80X$	7	12.49

LC (Lethal Concentration); CL (95% Confidence Limit);
DF (Degree of freedom); T.F (Toxicity factor) = LC_{50}
value of most toxic hrs/ LC_{50} value of other periods

SOME PHYSIOLOGICAL RESPONSES OF THE CATFISH, *Clarias gariepinus* (BURCHELL 1822) FED CASSAVA (*Manihot esculenta*) PEEL FORTIFIED WITH *Leucaena leucocephala* LEAF MEAL

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This study examined the growth and hematological characteristics of *Clarias gariepinus* juveniles fed diets with varying levels of fermented cassava (*Manihot esculenta*) peel (Fig.1) fortified with *Leucaena leucocephala* leaf meal (CPLM), for a period of 10 weeks in the laboratory. Fish feed accounts for at least 60% of the total cost of production. This has motivated the research for local, cheap feed material or wastes, as alternative energy feed for *Clarias gariepinus*, with the aim of reducing the cost of production, without compromising fish quality. Cassava peel as a cheap carbohydrate source has been found to be capable of supplying adequate calories to *Clarias gariepinus* fingerlings/ juvenile, with improved protein value, through fermentation with biomass from organic sources.

Six iso-caloric and iso-nitrogenous diets were formulated containing 0%, 30%, 40%, 50%, 60% and 70% CPLM replacing maize (diets D0, D30, D40, D50, D60 and D70). The juvenile *C. gariepinus* (mean average individual weight of 6.31mg \pm 0.23) fish were randomly distributed in groups of 15 fish per bowl of water, in triplicate for each treatment, in the laboratory, fed daily at 5% body weight in two rations, for a period of 8 weeks. Water temperature was maintained at between 25-27°C, pH was alkaline (7.9) while dissolved oxygen was close to air saturation (5.6mg/L). The physiological effect of the diets showed that there was significant decrease ($P < 0.05$) in the PCV, RBC, Hb of the fish respectively as the level of the CPLM level in the diet increased, while the WBC and lymphocytes increased (Table 1). The fish fed diet D50 had the highest values of MCH and MCV (47.00 pg and 160.00 fl) respectively. The sugar degrading enzyme activities from the gut of catfish also increased with increase in CPLM inclusion (Table 2).

The substitution of maize with CPLM, as energy source, beyond the optimum value of 50% elicited poor growth and physiological responses.

TABLE 1: Haematological parameters of the fish fed CPLM diets

Parameter	D 0	D 30	D 40	D 50	D 60	D 70
PCV %	32.0 _a	30.0 ^a	27.0 ^b	26.0 ^c	24.0 ^c	24.0 ^c
Hb (g/100ml)	9.2a	9.0a	8.1b	8.0 _b	7.0 _c	7.1 _c
RBC (10 ⁶ mm ⁻³)	3.6 ^c	3.2 ^a	2.0 ^b	1.9 _b	1.8 _b	1.5 _{bc}
WBC (10 ³ mm ⁻³)	7.5 ^c	7.5 ^b	7.6 ^b	7.6 _b	8.0 _a	8.0 _a

TABLE 2: Sugar degrading enzyme activity from the gut of catfish

Enzyme	D0	D30	D40	D50	D60	D70
Amylase	70.2 _a	35.1 _b	40.7 _b	39.8 _b	66.4 _a	65.3 _a
Sucrase	7.1 ^a	3.2 ^b	11.4 _a	4.9 ^a	10.0 _a	11.2 _a
Maltase	2.8 ^b	4.9 ^a	3.6 ^a	3.5 ^a	4.8 ^c	5.9 ^c

EFFECT OF *Eucalyptus globulus* ON FATTY ACID COMPOSITION OF THE GIANT AFRICAN CATFISH *Heterobranchus bidorsalis*

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H. bidorsalis is a freshwater fish that belongs to the family Clariidae. It can be readily differentiated from *Clarias* by the fact that it has the rayed dorsal fin followed by an adipose fin. The flesh contains lesser oil in comparison with that of *Clarias*. It has good taste, fast growth rate and is rarely parasitized making it well prized. They have tough skin and do not possess scales.

Susceptibility of fish to oxidation during processing and storage and subsequent loss of quality is directly related to the abundance of polyunsaturated fatty acids in fish. *Eucalyptus globulus* is a natural source of antioxidants. There is a dearth of information on the use of *E. globulus* in the diet of *Heterobranchus bidorsalis*. Therefore, the present study was carried out to evaluate the effect of *Eucalyptus globulus* on fatty acid composition of *Heterobranchus bidorsalis* fingerlings.

A 70 day feeding trial was conducted in 15 plastic tanks (40×30×35cm) with *Heterobranchus bidorsalis* fingerlings (12.05g ± 0.01 body weight) at a stocking density of ten fish per tank. Five iso-nitrogenous diets were formulated as control (CTR) along with four other diets containing four graded levels of *E. globulus* EG1 – EG4 (table 1). Each experimental diet was fed to five groups of fish between 08:00-09:00h and 18:00-19:00h GMT at the same fixed rate of 5% body weight per day. Temperature (°C), hydrogen-ion concentration (pH) and dissolved oxygen concentration (DOC) were monitored weekly.

At the end of the 70 day feeding experiment, total saturated fatty-acids (SFAs) increased progressively from the control group to EG4 (Table 2). Significant differences (p<0.05) also occurred in the Mono unsaturated fatty-acids (MUFAs) and poly unsaturated fatty acids (PUFAs) between the control and the fish fed experimental diets. The n3/n6 ratios were higher in fish fed *E. globulus* supplemented diets than that of control. The results suggests that *E. globulus* reduced PUFAs in *H. bidorsalis* and may be included in fish diet to reduce their oxidation during processing and storage.

Table 1: Composition of experimental diet (g/100g) containing dietary *Eucalyptus globulus* leaf supplementation for Giant African Catfish, *Heterobranchus bidorsalis*

Ingredients	Control	EG1	EG2	EG3	EG4
Fish meal	23.0	23.0	23.0	23.0	23.0
Soybean Meal	26.0	26.0	26.0	26.0	26.0
Groundnut Cake	28.0	28.0	28.0	28.0	28.0
Yellow Maize	11.0	11.0	11.0	11.0	11.0
Others	12.0	12.0	12.0	12.0	12.0
<i>E. globulus</i> leaf powder	0	0.5	1.0	1.5	2.0

Table 2. Fatty-acid composition of fish fed experimental diets

Fatty-acid	Control	EG1	EG2	EG3	EG4
Total SFAs	38.69	44.87	49.05	48.69	51.59
Total MUFAs	43.04	37.77	34.95	35.32	33.62
Total PUFAs	19.06	17.32	16.00	16.33	15.69
n3/n6	0.31	0.55	0.70	0.70	0.98

GROWTH, BIOCHEMICAL AND IMMUNE RESPONSES OF *Acipenser persicus* FED DIFFERENT LEVELS OF PHOSPHOLIPID

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In this study we evaluated the effect of dietary phospholipids on growth performance, biochemical and immunological parameters in Persian sturgeon *Acipenser persicus*. Fish with initial body weight of three grams were used for this study. Basal diet was supplemented with commercially easily available source of phospholipid, soy bean lecithin. Different phospholipid levels in diets were achieved by adding 0 (ontrol), 2, 4, 6, 8 and 10% soy bean lecithin at the expense of corn oil. The results indicated that fish feed supplemented with 2 and 4 percent lecithin containing 8.8 and 15.7% total phospholipids improves growth, survival, and immune responses significantly compared to control (Tables 1). It was concluded that determination of the optimal level of phospholipids in Persian sturgeon diet is of great importance.

Table 1. Growth performance in Persian sturgeon (*Acipenser persicus*) fed graded levels of PL

	Dietary soybean lecithin levels (%)					
	Control (0)	2	4	6	8	10
BW i (g)	3 ± 0.0	3 ± 0.0	3 ± 0.00	3 ± 0.00	3 ± 0.00	3 ± 0.0
BWf (g)	34.65 ± 1.0 ^a	60.69 ± 1.1 ^d	57.21 ± 0.67 ^d	53.82 ± 1.0 ^{cd}	45.35 ± 3.0 ^{bc}	44.64 ± 2.4 ^b
SGR (% day ⁻¹)	2.71 ± 0.0 ^a	3.37 ± 0.0 ^d	3.27 ± 0.0 ^d	3.20 ± 0.0 ^{cd}	3.19 ± 0.0 ^{cd}	2.99 ± 0.0 ^b
FCR	1.4 ± 0.06 ^a	0.95 ± 0.06 ^b	0.94 ± 0.01 ^b	0.92 ± 0.02 ^b	0.84 ± 0.08 ^b	0.69 ± 0.02 ^b
Survival (%)	58.82 ± 5.8 ^a	74.5 ± 6.7 ^{ab}	81.37 ± 1.6 ^c	74.50 ± 13.5 ^{ab}	65.68 ± 4.4 ^{ab}	76.47 ± 16.3 ^{ab}
Lysozyme (U mL ⁻¹)	29.14 ± 0.6 ^a	34.42 ± 1.2 ^{ab}	36.57 ± 0.0 ^b	34.56 ± 1.1 ^{ab}	34.19 ± 2.0 ^{ab}	30.85 ± 0.8 ^a
Total Ig (mg mL ⁻¹)	8.2 ± 0.3 ^a	17.26 ± 2.09 ^b	16.6 ± 1.5 ^b	16.26 ± 1.6 ^b	15.06 ± 1.9 ^{ab}	12.25 ± 1.01 ^{ab}
Phagocytic activity (%)	1.16 ± 0.1 ^a	1.86 ± 0.1 ^b	2.19 ± 0.0 ^b	2.02 ± 0.1 ^b	1.95 ± 0.1 ^b	1.18 ± 0.0 ^a
Triglyceride (mg dL ⁻¹)	1221 ± 132 ^b	620 ± 124 ^a	651 ± 93 ^a	851 ± 13 ^{ab}	771 ± 62 ^{ab}	859 ± 148 ^{ab}
Cholesterol (mg DL ⁻¹)	107.0 ± 13.5	87.3 ± 9.6	107.6 ± 13.6	120.6 ± 6.9	126.00 ± 5.6	131.3 ± 11.05
HDL (mg DL ⁻¹)	6.6 ± 0.8 ^a	16.6 ± 1.76 ^b	19.33 ± 1.76 ^b	23.66 ± 1.20 ^b	22.33 ± 2.3 ^b	21.33 ± 3.1 ^b
LDL (mg DL ⁻¹)	21.0 ± 2.8 ^a	34.0 ± 3.0 ^b	37.6 ± 3.2 ^b	45.6 ± 2.4 ^b	45.0 ± 0.57 ^b	45.6 ± 2.0 ^b

REDUCING RISK IN OPERATION OF EXPOSED HIGH VOLUME, ADVANCED FISH FARMS - HOW TO MAINTAIN SAFETY OF ASSET, PERSONNEL, MAINTAIN FISH WELFARE AND PREVENT FISH ESCAPE FOR OFFSHORE FISH FARMS

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It is a growing trend that fish farms are moved out of sheltered, near-shore areas and moved to locations further offshore. There are several advantages applying this solution, better fish welfare, less conflicts with interests in the ocean space as well as possibility to scale up output volume without compromising environmental sustainability are some of them. We need to be sure that facilities for exposed aquaculture can handle harsh environment and still contain the fish safely.

The main areas of concern when it comes to ensuring safe and reliable offshore fish farming units may be categories into: Asset integrity, personnel safety, fish welfare and prevention of fish escape.

Asset integrity includes structural strength, stability, mooring, technical arrangement, and solutions on board together with reliability of essential equipment installed.

Personnel safety is mainly addressing arrangement for emergency escape and fire safety. This included lifesaving appliances, launching equipment and similar as well as fire detection and -extinguishing. It is common to apply well know maritime codes as acceptance criteria for personnel safety. SOLAS is a good example with local flag- or shelf states interpretation of these requirements embedded in this maritime code.

Fish welfare and requirements related to this varies depending on local authorities. It is essential to verify the reliability of technology utilized to monitor environment of the fish. Instrumentation indicating oxygen level, temperature, salinity, turbidity is subject for special attention.

Fish control or prevention of escape is the main function of a fish farming unit. Structural integrity of net system and ropes together with capability of fish transfer systems are crucial items in fish control. Flexible net systems utilized in rigid high volume steel fish farming installation has proven to be exposed to fatigue and need to be attended to in particular. Wear and tear of net due to cleaning and handling is also a concern. Several of reported incidents related to fish escape happens while handling of fish – for example crowding due to de-licing or transfer. Equipment contributing to these operations needs to be specially attended to.

The four different items are considered equally important for safe and sustainable fish farming offshore. These items are also closely interconnected where integrity of one may support several others.

There is a significant potential to utilize competence from offshore and maritime to help operators of exposed fish farming units to identify operational risks by applying technical rules and requirements from classification.

Combining the well-known classification concept from maritime industry with balanced aquaculture-based requirements provides a robust and cost-efficient solutions to reducing risk in operation of offshore fish farming installations.

SNP GENOTYPING TOOLS AND THEIR APPLICATION IN AQUACULTURE BREEDING PROGRAMS: PEDIGREE MANAGEMENT TO GENOMIC SELECTION

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The last decade has seen wide adoption of incorporating genetic tools into breeding programs across all areas of agriculture. Decreasing costs coupled with advances in technology have made these tools, which were once only utilized by large scale commercial producers, accessible to mid-sized and even smaller scale producers. Modern genotyping technology provides the opportunity to address some of the unique challenges faced in aquaculture such as tracking pedigree, inbreeding avoidance, disease resistance, increasing fecundity, and improvement of growth and meat quality characteristics.

By far, the most widely used genetic marker in modern breeding programs are single nucleotide polymorphisms (SNPs). This type of genetic variant is found abundantly in all genomes, making SNP genotyping technologies a versatile genetic tool that can be designed for any species, is adaptable for various applications and is scalable, targeting anywhere from one to millions of SNPs. Generally, SNP panels fall into 1 of 4 scale categories; low density, LD (~100-500 SNPs), medium density, MD (1000-5000 SNPs), high density, HD (10,000 to 500,000 SNPs) and, more recently, ultra high density, ultraHD (10 million to 100 million SNPs). The required panel size is usually determined by the type of downstream application, with basic tasks like pedigree tracing and genetic diversity monitoring only requiring a few hundred SNPs, to more complex tasks like genomic selection and genome-wide association studies requiring 30,000+ SNPs.

Here, we demonstrate the creation and downstream application of LD, MD, HD and ultraHD genotyping panels for several well-known and lesser-known aquaculture species. The utility of these panels at accomplishing pedigree tracing, genetic diversity monitoring, QTL identification and genomic selection is discussed. We highlight the ability of SNP genotyping panels to be cost-effective tools to manage genetic diversity and guide breeding strategies to achieve both short-term and long-term breeding goals in modern aquaculture.

REARING, ONGROWING AND HARVESTING OF CARIBBEAN SPINY LOBSTER (*Panulirus argus*) IN SEA CAGES, (CUBA)

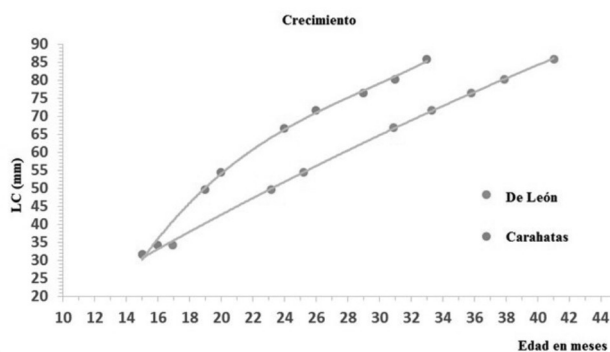
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An experimental marine farm for rearing and ongrowing of Caribbean spiny lobsters (*Panulirus argus*) was installed in Carahatas Bay, Villa Clara, Cuba, through a cooperation agreement between Vietnam and Cuba. The first operation of the farm began in April 2012, when about 30 kg of juvenile lobsters were introduced into four ongrowing cages. Thirteen months later, 289 kg of commercial-size lobsters were harvested, with an average individual weight between 400 and 525 g. An initial mortality of 10% occurred due to capture and handling stress, but survival rate throughout the ongrowing period was 90 %. The value of the food conversion factor (CF), based on dried weight, was 2.6-2.8. Growth rate (von Bertalanffy's k) was higher ($k = 0.34$) than previously reported for commercial-size lobsters ($k = 0.24$ on average), that represented 1.0-2.9 g/day. Upon harvesting, over 82 % of all lobsters had reached commercial sizes and several females exhibited spermatophores and egg masses, indicating that the quality of life for lobsters in the ongrowing cages was adequate. These results show the feasibility of the Vietnamese experience on spiny lobster ongrowing as applied to *P. argus* in Cuba. In all experimental time the total catch was 1350 kg, with 90-95 % of survival.

The lobster that growing in the cages increase their weight in 7 months in relation with the natural fisheries

Below are observed coefficients “ k ”, which represent the rate of growth of the specimens, reporting that during the whole fattening period in the cages, these are higher (0.34) than those reported for commercial lobster, which according to de León et al, (1995) and Cruz Leocadio (2008) have an average value of 0.24. These values demonstrate that the lobster from breeding farm installed in Carahatas, grow faster than the natural environment, which are subject to the effects of the fishery, and that the specimens in the cages received daily feeding and have not to expend energy to your daily foraging. The values of “ k ” growth rates showed no significant differences between all experimental time.



A STUDY ON DIGESTIVE ENZYME ACTIVITY IN *Etroplus suratensis* FINGERLINGS FED WITH DIFFERENT PERCENTAGE OF PROTEIN AND LIPID FORMULATED DIET.

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A feeding trial including diets with three protein levels (25%, 30% and 35%) and three lipid levels (3%, 6% and 9%) was conducted to investigate the digestive enzyme activity (Amylase, Lipase, Protease) in pearl spot fingerlings (average initial weight: 12.76 ± 0.04 g). Fish were randomly distributed into 18 glass tanks (100 litre capacity). The fishes were fed thrice daily at 3% of their body weight. Every fortnightly all the fishes were weighted and redefine the feed ration. During the trial the digestive enzyme activity like amylase, lipase, Protease was analyzed. Amylase, protease found to be increased by increasing protein levels was (12.1 ± 0.56 μg of maltose liberated mg^{-1} tissue protein h^{-1} at 37°C), (22.1 ± 0.62 μg of leucine liberated mg^{-1} tissue protein h^{-1} at 37°C), but no significant ($P > 0.05$) difference was observed. And lipase enzyme activity was found to be higher in fishes was (47.8 ± 0.42 μg of α -naphthol reduced mg^{-1} tissue protein h^{-1} at 37°C) fed with 9% lipid was significantly ($P < 0.01$) higher than that of the other groups. Feed with higher inclusion level of protein (35%) has higher protease activity than other treatments and higher lipase activity was noticed with feed with 9% lipid level, resulted in increased stress response in experimental fishes. The results of this study indicated that Amylase, Protease enzyme activity was found to be higher with higher inclusion level of protein in feed. And higher Lipase activity was found in higher lipid inclusion feed.

IMPACT OF WATER QUALITY ON GROWTH PATTERN AND TISSUES OF A CULTURABLE CICHLID - *Tilapia guineensis* COLLECTED FROM SOME AQUATIC ECOSYSTEMS (TOPO, GBAJI, AGBOJU AND AJEGUNLE) IN BADAGRY DIVISION, LAGOS, NIGERIA

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The study examined the impact of physico-chemical parameters of four aquatic ecosystems in Badagry division, on morphometric distances (total length, standard length and body weight) and tissues (gill, muscle and small intestine) of *Tilapia guineensis*. Samples of water were collected for 18 months, while twenty wild adult *Tilapia guineensis* from each station were examined for histopathology using standard procedures.

Length- weight relationships of *T. guineensis* obtained from all the creeks showed that *T. guineensis* had negative allometry ($b < 3$). Only samples from Ajegunle had condition factor (k) value less than 1.

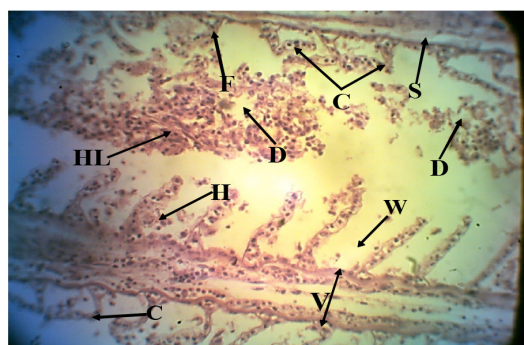


Figure 1: Gill structure of *T. guineensis* from Ajegunle creek showed severe degeneration (D) and hyperplasia (HL) of secondary gill lamellae (D), separation of gill fill mate from basement membrane (S), and curling of secondary lamellae (C) with unclear water channel (W). It is also characterized by hypertrophy of lamellae (H). There is also evidence of partial fusion of lamellae (F) and dilation of gill filament (V)

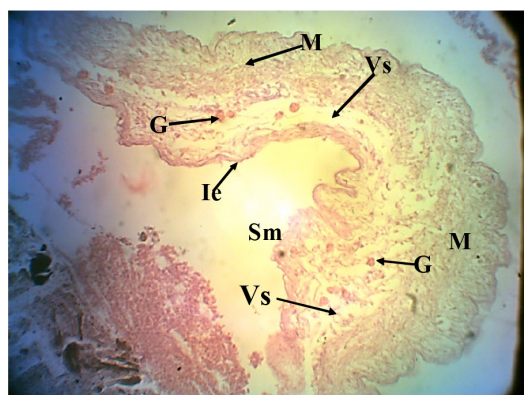


Figure 2 At Ajegunle, cross section of the small intestine showed the intestinal fold (I), Outer muscularis (Om) and inner muscularis layer (Im). There is slight degeneration and vacuolization in Submucosa connective layer (D). However, glandular cell (Gc) are adequately seen. It also showed mild lesion in epithelia lining Serosa(S).

Table 1: Water Parameters and Morphometric Features of *T.guineensis* from the stations

Parameter	Agboju	Gbaji	Topo	Ajegunle
TL (cm)	10.57±1.12 ^a	10.34±1.14 ^a	10.46±1.32 ^a	8.55±0.85 ^{ab}
BW(g)	29.13±8.34 ^a	28.85±8.56 ^a	29.44±7.85 ^a	26.47±5.65 ^{ab}
SL(cm)	8.34±1.25 ^a	8.64±1.22 ^a	8.79±1.29 ^a	6.53±1.25 ^{ab}
b value	2.143 ^a	2.132 ^a	2.002 ^a	0.385 ^{ab}
K value	1.98±0.83 ^a	2.59±0.44 ^a	2.34±0.98 ^a	0.99±0.54 ^{ab}
pH	7.14±0.54 ^a	7.33±0.48 ^a	7.43±0.52 ^a	7.56±0.63 ^a
Temp.(°c)	28.03±2.42 ^a	27.49±2.99 ^a	27.44±1.99 ^a	28.45±2.35 ^a
BOD(mg/L)	248.80±18.57 ^{ab}	209.68±21.68 ^a	215.46±24.19 ^b	247.98±28.37 ^{ab}
Conductivity (µs/cm)	254.27±42.12 ^a	220.27±30.97 ^{ab}	236.60±40.48 ^{ac}	268.30±45.41 ^{ad}
TDS (mg/L)	116.61±8.78 ^a	135.89±28.74 ^{ab}	131.78±21.12 ^{ac}	124.97±44.38 ^{ad}
COD(mg/L)	175.90±21.65 ^a	166.21±20.93 ^{ab}	167.50±24.60 ^{ac}	184.46±25.29 ^{ad}
DO(mg/L)	6.20±0.71 ^a	6.53±1.88 ^a	6.76±0.99 ^a	4.77±0.91 ^{ab}
Hardness(mg/L)	128.85±14.76 ^a	191.45±10.56 ^{ab}	197.37±5.53 ^{aa}	238.25±18.91 ^{ac}
Alkalinity(mg/L)	37.98±5.63 ^a	39.44±6.96 ^a	38.86±6.49 ^a	34.75±4.21 ^b

Mean with same superscript in the row=not significantly different ($p > 0.05$)

ASPECTS OF IMPACTS OF PROPOSED BADAGRY DEEP SEA PORT ON THE ECOSYSTEM AND LIVELIHOOD OF FISHING COMMUNITIES IN BADAGRY, LAGOS STATE, NIGERIA

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Federal government of Nigeria, in collaboration with Lagos State Government proposed development of a seaport in Badagry. Badagry is within coordinates 6°25'N 2°53' / 6.42°67°N 2.88°E. It shares boundaries with Ogun State both in the North and in East and is bounded on the west by the Republic of Benin. In the South, it stretches for 180 kilometers along the coast of the Atlantic Ocean. The research examined and documented the current state of the ecosystem and livelihood of thirteen communities that will be impacted by the proposed deep-sea port. The communities that will be impacted are Gberefu, Yovoyan, Gayingbo, Agonvi, Agonrin, Hoke-daho, Kujinada, Aivoji, Asakpo, Sheik modawa, Agonvi town, Ganyingbo town and Gbaji yeke tome.

Qualitative and quantitative approaches were used for the study. Review of secondary data was used to investigate the demographic data of the community. Participatory Rural Appraisal were conducted for about 300 households in the communities. Majority of the sampled respondents were in the age range of 45 years and above with females (60%) more than males (40%) in the entire population sampled. Educational levels are relatively low among the population sampled. About 65% of the farmers in the communities have primary school education but educational level of their children is very high and 67% of them have attained tertiary level education while 33% have attained secondary level education. The majority of the sampled households are food secured as they have multiple livelihood systems that keep them engaged throughout all seasons of the year. Seventy three livelihood were encountered but capture fisheries is the major occupation in the study area and it is complemented with culture fisheries. Most commonly produced livestock are pig, cow, goat, chicken and duck. Major sources of income in the communities are fishing and farming followed by artisans, trading and paid jobs. In addition, some activities are gender and age related such as: Hawking (boys and girls), Fish Smoking(women), Mart weaving (women), Pap (eko) leaves (women), Marketing of fish(women), Vigilante groups (odua people congress), Non farming activities (youth, men and women), Farming (men and women), Livestock (men and women), Sand mining (youth).

Existing groups were not strong enough to control market or operate as pressure group to influence policies and regulate market prices, which has been identified as a major limitation in the study area. There was no regular training or capacity building, hence the groups were not operating as a business enterprise and could not expand or increase capacity. Consequently not able to contribute significantly to poverty alleviation and increase employment opportunities in their localities. This document will serves as one of the guides to government for decision-making and compensation to the communities.

Table 1: Relative impact of project on the communities

Level/pattern of impact	Communities/groups
Communities that will lose land but not be resettled	Gbaji yeke, Gayingbo town, Agonvi town etc
Communities that will be resettled and lose land	Gberefu, Yovoyan, Agonvi sea beach, Gayingbo sea beach, Hoke daho, Kujinada, Agorin town, Mudawa community
Those at the resettlement sites who will lose land for the resettlement sites	Gberefu
Not lose land and not be resettled but who may otherwise be affected	Tenants to land owners, Market associations, Producers associations, Transport associations

(Continued on next page)

Table 2 Livelihood Analysis Diagram for the Communities

Livelihood Activities	Ranking	Tools and implement	Sources of funding	Market options	Market distance
Rain fed farming		Land	Cooperatives	Agbalata	10km
Cassava	2	Water	Thrift	Seme	20km
Maize	3	Hoe	Pension	Igogazebe	50km
Sweet potato	8	Tractor	Savings	Agbara	50km
Cowpea	7	Cutlass		Lusada	60km
Yam	9	Fertilizer		Owode	50km
Melon	13	Herbicides		Agunmo	7km
Ground nut	10	Pesticides		New market	
Pineapple	15	Manure		Ajara	
Water melon		Sprayer			
Irrigation farming	4	Labour			
Tomato	4	Pumping machine			
Pepper	5				
Okro	6				
Chochorou	6				
s	6				
Ugwu	6				
Amarantus	6				
Water leave					
Bitter leave	1				
	11				
Tree crop	15				

Coconut	11				
Mango	1				
Pawpaw	12				
Cashew	12				
Palm tree					
Plantain					
Banana					
Livestock		-Pen house	Cooperatives	Agbalata	10km
Cow	1	-	Thrift	Seme	20km
Sheep	7	-	Pension	Igogazebe	50km
Goat	3	Vaccines and treatment by veterinary officer	Savings	Agbara	50km
Pig	2			Lusada	60km
Guinea fowl	4			Owode	
Duck	10			Agunmo	50km
Rabbit	7			New market	
Chicken	4			Ajara	
Snail	8				
Cat	9	-			
Dog	5	Lantern for heating			
Alegator	11				
Fishing		Net Boat/engine Ponds	Cooperatives	Farm gate	2km
Marine			Thrift	Inside the community	
Lagoon			Pension		
Creek			Savings		
Aquaculture					
Off-farm activities				Farm gate	2km
-Smoking of fish, cray fish, sea tortoise				Inside the community	
-Cassava processing to garri, apran, fufu					
-Sun drying of cowpea, melon, maize					
Marketing Activities				Farm gate	2km
-Fish catch is sold to women who sell some to traders and process the rest as				Inside the community	

(Continued on next page)

smoked fish -Crop produce is sold to women and also at farm gate. Women process and market the product -Produce by youth is sold at farm gate to traders who take it to market -Livestock are sold to traders/wo men who take it to market					
Non-farm activities Tailoring Art and printing Textile design Brick laying Culture design Aluminum Civil servants Teaching Traders Carpentry Plumbing Hair dressing Mechanic Welder Hunting Transportat ion Clergy Herbalist Sand mining Smuggling Car dealers					

AN ARTIFICIAL INTELLIGENCE-POWERED ACE AQUATEC SEAL DETECTION SYSTEM: REAL-WORLD EXPERIMENTAL DESIGN AND VALIDATION

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Ace Aquatec Ltd is at the forefront of developing innovative products that promote fish welfare and sustainability in Aquaculture. One of the many products currently in development aims to deter seals from fish farms in a humane, sustainable way. Seals are one of the most prevalent predators on fish farms, causing physiological and psychological damage to fish, as well as damaging the nets. Ace Aquatec developed and deployed acoustic deterrent devices (ADDs) across farms which effectively prevented seal attacks. However, recent policy changes in Scotland saw the removal of ADDs due to concerns over the noise pollution and harm caused to marine species. In response, Ace Aquatec has developed the first seal detection system which is designed to work in tandem with current deterrent systems. These trigger cameras accurately identify seals and instruct the deterrent systems to turn on only when necessary.

In response, Ace Aquatec has developed a first-of-its-kind artificial intelligence (AI)-driven seal detection system which is designed to work in tandem with conventional deterrent systems. The seal camera software benefits from a range of movement capabilities (see Figure 1). Using a PTZ degree camera, the system automatically identifies and tracks the movements of seals in a 125664m² circular area. The system can also operate at night by applying night vision image adjustment, ensuring that sites have 24/7 protection.

The functionality and accuracy of the system was tested under varying conditions: changes in light, distance to the camera, movement and false positives. By displaying seal images around the test area, it was found that the system successfully identifies seals under these changes and can distinguish between seals and other animals/objects. Figure 2 shows that the confidence increases to 99% as the distance is increased. In operation, this means that the system will be able to detect seals at a large distance and track its approach to the farm. Only when it has crossed the boundary into the farm will it trigger the ADDs.

Future development will include the detection of other significant species. For example, facial recognition can be added to aid with site security and the identification of other species may be of interest for the purpose of environmental/wildlife surveys being conducted in the area.

This research is supported by Innovate UK funded Knowledge Transfer Partnership (KTP) grant with Ace Aquatec (KTP grant no. 1026904).

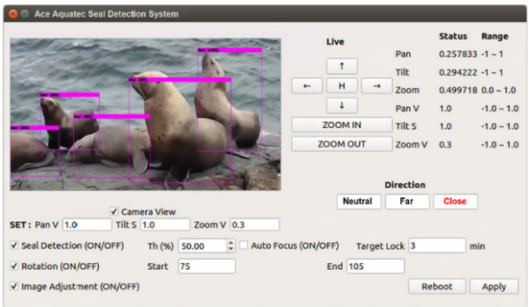


Figure 1: Ace Aquatec Seal Detection Control Panel

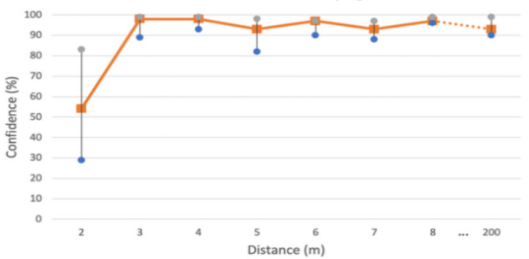


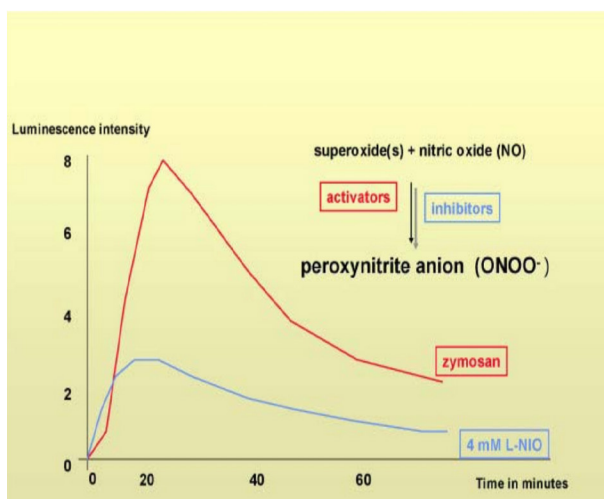
Figure 2: Seal Detection Confidence Levels

PATHOLOGY OF AQUATIC ORGANISMS AND BACTERIAL DISEASES

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Disease is a natural trait of life. Relatively little research has been done on the disease in wild aquatic animals. This article focuses primarily on cultivated animals. Aquatic animals are specifically associated with the main groups cultivated around the world: Mollusks, crustaceans, and fish. Whether the pathogen causes the disease depends on it Not only from the pathogenicity of the pathogen, but also from the immune system of the infected animal. Both sides of this relationship are influenced by the environmental conditions and stressors that can impair the immune system. It disrupts the interaction between the host and the pathogen, causing illness. Currently known, it outlines the immune mechanisms of mollusks, crustaceans, and fish. Approach to Disease prevention, diagnosis, and management are some of the technologies under development and future requirements.



IDENTIFICATION OF *Vibrio kanaloa* IN THE *Oyster crassostrea gigas* BY FLUORESCENT IN SITU HYBRIDIZATION

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Background

An ideal model for studying *Vibrio* and host and their interaction in disease dynamics is *Crassostrea Gigas* in the North Sea, exposing invasive sources to pathogens, especially (*vibrio kanaloae*).

Methods

This study used molecular fluorescent in situ hybridization (FISH) techniques to rapidly identify the diversity of bacteria in the oyster *Crassostrea Gigas* from Sylt Island and 16SrRNA gene sequence validated all strains' identification. Oysters were collected and exposed with *vibrio kanaloae* strains on different tissues were examined utilizing culture-independent methodologies.

Results

the digestive glands, gill and Muscle in *vibrio Kanaloa* were identified as metabolically active by the FISH technique, one of the advantages of the FISH technique is that it is accurate and very easy to use.

Conclusions

This technique allows rapid assessment of *Vibrio* in oysters and seafood.

IN VITRO AND IN VIVO EVALUATION OF INDUCED *Vibrio splendidus* TOWARD IMMUNE RESPONSE AND CROSS PROTECTION IN *Oyster grassostera gigas*

*Noushin Arratahery, Professor Jens Rolff, Mathias Wegner

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Vibrio splendidus is an oyster pathogen associated with the summer mortalities affecting the production of *Crassostrea gigas* oysters worldwide. The increasing experimental evidence suggests that there are some forms of specific acquired immunity in invertebrates, but the underlying mechanism is not fully understood. In the present study, Pacific oyster (*Crassostrea gigas*) stimulated primarily by exposure to 10^5 *Vibrio splendidus* displayed stronger immune responses levels when they encountered the secondary challenge of 10^8 *V. splendidus*. we tested *Vibrio* primed by exposure to colonized in oysters to investigate their resistance against hemolymph in vivo and in vitro experiments. we investigated the immunological responses in oysters and whether this colonization results in elevated mortality. The selected strains showed higher survival in seconde challenge in oyster model and cross- protection against *Vibrio splendidus* in vitro. A population of oyster selected for resistance to the *Vibrio splendidus* showed increased in vivo survival, in addition, *V. Splendidus* reduction of pathogen shedding. In conclusion, our results suggest that the primary stimulation of *V. splendidus* led to immune priming in oysters when encountering the secondary challenge with *V. splendidus*, in addition, the increased survival of oysters almost certainly by stimulating the immune system in hosts led to prevent vibriosis in the oyster.

CHARACTERIZATION OF TOXIN GENES ANTIMICROBIAL SUSCEPTIBILITY OF *Staphylococcus aureus* ISOLATES IN FISHERY PRODUCTS IN IRAN

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Introduction:

Staphylococcus aureus is one of the most common causes of seafood-borne diseases worldwide, which are attributable to the contamination of food by preformed enterotoxins.

Purpose:

The final aim of this study was to investigate the contamination of fishery products before their purchase and consumption.

Methods:

In this study, a total of 206 (34.3%) *Staphylococcus aureus* strains were obtained from 600 fish and shrimp samples and were tested by PCR for their antimicrobial susceptibility. We assessed the prevalence of the genes responsible for the staphylococcal enterotoxins (SEA, SEB) and toxic shock syndrome toxin 1 (TSST-1) genes.

Results:

The results indicated that 34% of aqua food samples were contaminated with *S. aureus*, and 23.8% of these isolates were mec-A-positive. Sixty-four percent of the strains isolated from contaminated seafood was enterotoxigenic *S. aureus*, and 28.2% of SEs were MRSA-positive. The most prevalent genotype was characterized by the presence of the sea gene (45.2%), followed by the seb gene (18.5%), and the tst gene encoding TSST-1 was found in eight strains (3.9%). Of the 206 *S. aureus* isolates, 189 strains (84.9%) were resistant to at least one antibiotic.

Significance:

Given the frequent outbreaks of enterotoxigenic MRSA, it is necessary to make revisions to mandatory programmes to facilitate improved hygiene practices during fishing, aquaculture, processing, and sales to prevent the contamination of fishery products in Iran

ANTIBACTERIAL ACTIVITY OF THE SEA CUCUMBER *Holothuria leucospilota* WHOLE BODY EXTRACT AGAINST METHICILIN RESISTANT AND ENTEROTOXIN PRODUCING *Staphylococcus aureus* STRAINS

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Introduction:

Preformed enterotoxins of *Staphylococcus aureus* are one of the most common causes of seafood-borne food poisoning worldwide. Aquatic organisms, including those used as seafood, are also a source of organic compounds of potential medical importance.

Purpose:

Within the framework of an antimicrobial activity study of marine macro-organisms from the Persian Gulf, bioactive compounds of the sea cucumber *Holothuria leucospilota* were extracted from whole sea cucumber bodies using chloroform or methanol. The extracts were evaluated for their antibacterial effects against methicillin-resistant *Staphylococcus aureus* (MRSA) and enterotoxin producing *Staphylococcus aureus* strains (SEASA, SEBSA).

Methods:

Antimicrobial activities were determined using three methods: disk diffusion tests, minimum bactericidal concentration (MBC), and minimum inhibitory concentration (MIC)

Results:

The results demonstrate that methanol and chloroform extracts have an inhibitory effect on the growth of all strains tested at MIC concentrations up to 100 mg/ml. Also, the chloroform extract demonstrated bactericidal activity against SEBSA in concentrations of about 100 mg/ml. The extract below 100 mg/ml concentrations also shows bactericidal effects against MRSA and SEBSA. The highest antibacterial activity was found in the methanol extract.

Significance:

Therefore, sea cucumber extract is an interesting candidate for the identification of new antimicrobials, however, comprehensive investigations are needed to separate and identify the active compounds from *Holothuria leucospilota* from the Persian Gulf.

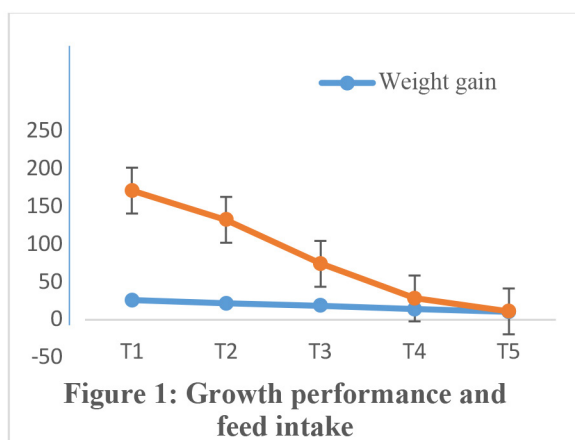
IMPACTS OF INGESTED MICRPLASTICS ON THE GROWTH PERFORMANCE AND RATE OF FEED CONSUMPTION OF AFRICAN CATFISH (*Clarias gariepinus*)

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The use of plastics for various objects has tremendously increased over the years. This result to vast numbers of plastics debris in the environment. Plastics wastes are the most common sea debris constitute over 70% of all ocean debris.

The effects of ingested microplastics on the growth rate and the rate of feed consumption of *C. gariepinus* was tested in this study. (150) juvenile *C. gariepinus* with average weight of 27.4g were exposed to microplastics and 2mm compounded feed at different levels. Five treatments (T) replicated in triplicate were stocked with ten fish. The fish in each treatment were fed 5% of their body weight 2 times per day for 8 weeks with various level of prepared microplastics (MP) and 14g of compounded feed (CF). T1, T2, T3, T4 and T5 were fed with 0, 4, 6, 8, 10g MP respectively. Highest weight gain of 26.07 ± 6.8 g was observed in T1 followed by T2, T3, T4 and T5 with a mean weight gain of 21.83 ± 2.0 , 18.53 ± 1.5 , 13.82 ± 8.0 and 10.54 ± 5.7 g respectively. The result of the feed intake shows that fish in T1 consumed the highest feed though, not exposed to microplastics while fish in T2, T3, T4, and T5 consumed 132.40 ± 6.9 , 74.09 ± 5.1 , 28.26 ± 1.7 and 11.10 ± 3.8 g respectively. There was a significant difference ($p < 0.05$) in the weight gain and in the feed intake of the fish among the treatments. Across the treatments, the fish mortality ranged from 20 to 100%. 20% mortality was recorded in T1 (control). 50% mortality was recorded in T2 while 100% mortality was recorded in T3, T4 and T5. The results from this research show that ingested microplastics have significant effects on the growth performance and the rate of feed consumption of *C. gariepinus*



BANGLADESH AQUACULTURE AND FISHERIES CHALLENGES WITH OPPORTUNITIES

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Bangladesh is overpopulated and disaster-prone developing country where Fish culture is the main income generation now a days. The data and information were sourced from the publication of the Department of Fisheries and related non-published grey literature. Bangladesh is predominantly an agrarian economy and is naturally endowed with a huge sweet water resources and the world's longest continuous sea beach. With the world's largest flooded wetland, the third largest aquatic biodiversity in Asia behind only to China and India, Bangladesh is considered as one the most suitable region for aquaculture and fisheries in the world. The country has an inland water area of about 45,000 km² and about 710 km long coastal belt. Given this extensive water resource, it is evident that fisheries play an important role in the economy and the diet of the population. Fish and fish products supply 60 percent of animal protein and around three percent of total export earnings. In recent years, however, the fisheries sector is confronted with challenges posed by numerous natural and anthropogenic causes such as climate change, natural disasters, unbalanced urbanization and industrialization, overfishing and environmental pollution. The combined effect of these factors is posing significant threat to the income and food security of the population and urges for immediate actions by government and policymakers. Government of the people's republic of Bangladesh, NGO's and Fishermen are combinedly trying to making the sustainable developed the of the Fisheries sector.

LINKING *vgll3* GENOTYPE AND AGGRESSIVE BEHAVIOUR IN JUVENILE ATLANTIC SALMON (*Salmo salar*)

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Aggressiveness can affect social hierarchies and result in the unequal distribution of resources, with aggressive individuals monopolizing access to food influencing growth. In Atlantic salmon, aggression has been found to have a genetic component, and growth also influences maturation timing. Maturation timing associates with a large-effect locus around *vgll3*, which is also linked to growth and condition, with *vgll3**EE (early maturation) individuals having higher condition factor than *vgll3**LL (late maturation). Here, we examine the possibility that aggressiveness may play a role in juvenile *vgll3**EE individuals having higher condition factor by having increased food intake due to higher aggression promoting increased food access. This prediction was tested under a social context: aggressiveness and feeding activity in four size-matched individual salmon, two from each genotype, were observed over 95 trials. Contrary to our prediction, *vgll3**LL individuals, not *vgll3**EE, were more aggressive. Increased aggression of *vgll3**LL individuals was independent of their sex and size, and genotypes did not differ in their condition factor nor feeding activity. These results imply that aggressiveness may have an energetic cost impairing growth and condition, especially when food cannot be monopolized. This may have critical implications for individual fitness and aquaculture practices.

NUTRITIONAL VALUE OF DIFFERENTLY PROCESSED KENAF *Hibiscus cannabinus* SEED MEAL AND THEIR APPARENT NUTRIENT DIGESTIBILITY COEFFICIENTS FOR AFRICAN CATFISH *Clarias gariepinus*

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Feed constitutes more than 60% of operational cost in fish farming. Increasing cost of conventional feedstuffs due to competition between livestock and human necessitates research into cheaper and readily available unconventional feedstuffs for profitable fish farming. Kenaf (*Hibiscus cannabinus*) Seed Meal (KSM) has been reported to be a nutrient-rich feedstuff for aquaculture. However, there is limited information on the nutritional value and nutrient digestibility of KSM in the diet of *Clarias gariepinus*.

The chemical components of differently processed KSM; Roasted (RoKSM), Soaked (SoKSM), Sprouted (SpKSM) and Cooked (CoKSM), as well as Raw (RaKSM) were determined using standard methods. A reference diet (RD) and five (5) experimental diets of 70% RD and 30% of each of the raw and processed kenaf seed meal constituted the experimental diets used for the digestibility trial. Chromic oxide was incorporated in the reference diet at 0.5% as an external marker. Using a completely Randomised Experimental design, the experimental diets were administered to *C. gariepinus* (n=180, 8.19±0.17g) in triplicate. Apparent Digestibility Coefficient (ADC) of Crude protein, ether extract and gross energy of the meals were calculated for *Clarias gariepinus*.

Relative to the RaKSM which had a crude protein of 21.17%, other processing method enhanced the crude protein in KSM. The ash content, Na/K ratio and Ca/P ratio were highest significantly ($p < .05$) for RoKSM. While soaking was not effective in reducing the content of tannin, saponin and oxalate in KSM, other processing techniques significantly reduced ($p < 0.05$) the levels of these anti-nutritional factors. Methionine was the limiting amino acid in the RaKSM and other processed KSMs. However, the Amino Acid Score (AAS) of the processed KSMs were higher than RaKSM. The apparent crude protein, ether extract and gross energy digestibility coefficient of kenaf seed for *C. gariepinus* significantly increased ($p < 0.05$) when processed (Table 1). It can therefore be concluded that kenaf seed meal stands the potential of being used as ingredient of protein source in the feed of *Clarias gariepinus* feed.

Table 1: Apparent digestibility coefficient (%) for crude protein, ether extract and gross energy of raw and processed kenaf (*Hibiscus cannabinus*) seed meal (KSM) for *C. gariepinus*

	RaKSM	RoKSM	SokSM	SpKSM	CoKSM
Crude protein	51.45±10.83 ^b	77.54±7.07 ^a	94.76±2.28 ^a	98.29±0.59 ^a	85.13±4.14 ^a
Ether extract	88.65±1.15 ^b	93.61±2.06 ^a	97.48±1.25 ^a	97.10±0.32 ^a	97.48±0.61 ^a
Gross energy	49.12±10.04 ^b	67.19±9.50 ^b	97.09±2.34 ^a	97.37±1.01 ^a	91.75±1.04 ^a

Mean ±SE (n = 3) value in the same row with different superscript are significantly different ($p < 0.05$).

HIGH THROUGHPUT MONITORING OF WATER SAMPLES FOR AUTOMATED DETECTION OF HARMFUL ALGAE AND SEA LICE

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Aquaculture is the fastest growing food production sector globally and is projected to increase to 109 million tons by 2030. This significant growth is due in part to the global demand driven by recognition of the significant nutritional value of seafood and higher revenue potential for farmers and producers. Aquaculture farming is also generally less detrimental on the environment than traditional livestock production. At the same time, in comparison with other livestock industries, aquaculture is significantly behind in terms of technology innovation. However, one of the most significant challenge is to mitigate the effects of diseases that are leading to significant production losses. Most diseases are caused by parasites (such as sea lice), harmful algae, etc. Currently, the water samples are collected manually at the farm and then analyzed by trained technicians in a laboratory, often centralized, using conventional lens-based microscopes. The current approach is expensive, laborious, time consuming and limiting scalability of operations. Furthermore, due to the delays associated with conventional sample analysis, even if a disease agent is detected, it is often too late to apply risk mitigation strategies and the response shifts towards using one of the treatment options mentioned above or pre-mature harvesting.

With support from the National Science Foundation, Lucendi is responding to this urgent and unmet market need by developing an automated and cost-effective Aqusens platform. Using deep learning, Aqusens provides persistent monitoring and early identification of pathogenic microorganisms, such as sea lice and dangerous algae at a throughput of >100 mL/h (1-5 L/h for larger objects, like sea lice). The device does not require any sample pre-processing or labeling, and can detect and image ~99% of all objects (sized 4 μ m - 1 mm) in water samples. This will empower aquaculture experts to apply risk mitigation and protection strategies to avoid crop loss. Fig. 1A shows the current version of the device that is portable and can be used in-field. Fig. 1B shows Aqusens images of harmful algae and of a zooplankton used as fish feed. In experimental trials we have demonstrated classification accuracy of >97% in identifying harmful microorganisms and are currently working with partners on field trials. We are interested in new partnerships and collaborations and open for trials and demonstration opportunities.

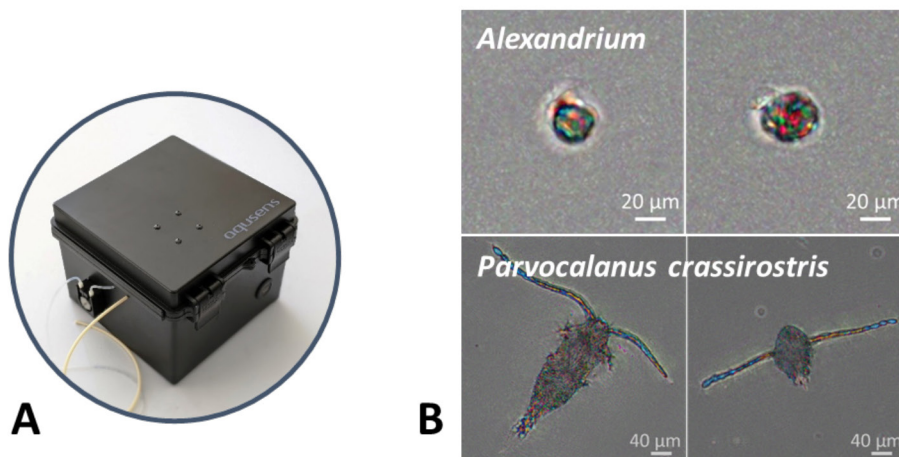


Figure 1: Aqusens platform and initial results. A. Photo of the device prototype prepared for in-field trials. Device is portable with dimensions of 8 × 8 × 6 in, weighing < 3 lbs. **B.** Images of harmful algae and zooplankton captured by the device at a flowrate of 100 mL/h. Currently Aqusens can identify selected harmful microorganisms with >97% accuracy.

ANNUAL STUDY OF FISH CATCH AND IDENTIFICATION OF THE MOST CAPTURED SPECIES, THE MORE CONSUMED IN KISANGANI AND ITS SURROUNDINGS IN THE DRC (YALOKOMBE CASE)

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An annual socio-economic study of fish catch and determination of the most caught, most consumed species was conducted in Kisangani and its surroundings, Yalokombe case, with a sample of 120 fishermen.

The aim of this study was to identify participants, along the value chain, to analyse the dynamics of production, packaging and marketing, to make a technical analysis at all strategic levels of the fishing industry for understand me organization operation and ultimately develop an intervention strategy to help the sector and improve their performance.

The study was conducted in YALOKOMBE, a village at kilometer 52, Kisangani Road to word Yangambi, Democratic Republic of Congo, following a wide range of different gear and techniques, thus allowing the capture of multiple species of fish. The catch structure at Yalokombe has a larger number of species in total. Ichthyological surveys show a fauna of 13 species divided into 8 families.

The specific abundance fish not sold by Yalokombe fishermen was of 5 species belonging to 4 families. Several species of fish are requested and consumed by consumers. With a total of 8 species identified and divided into 6 families.

Fish catches have decreased on the Congo River, with 100% of respondents unanimously saying the decline.

There are conflicts between the fishermen of Yalokombe, there are also several difficulties of various origins experienced by fishermen of the Congo River.

The results showed that in Yalokombe village, 70% of fishermen are in the 18-40 years old and 30% age group over the age of 40; 86,66% de *D.antonii* ; followed by *Hydrocinus goliath* with 66,66% ; higher than *Labeo lineatus* with 65% ; higher than *Lates niloticus* with 53,33% ; followed by *D.fasciatus* with 38,33.

There are several productive activities in Yalokombe, of which fishing ranks first as the main activity with 95%. The second activity is agriculture with 16.66%.

And there are strategies that can be adopted to develop fishing activity in Yalokombe.

Table1 : Tranche d'âge des pêcheurs de YALOKOMBE

Age	18-40years	>40years
Summy	84	36
%	70	30

Table2 : La structure des espèces de poissons les plus demandées et les plus consommées

N°	Espèces	Fréquence	%
1	<i>D.antonii</i>	104	86,66
2	<i>Hydrocinus goliath</i>	80	66,66
3	<i>Labeo lineatus</i>	78	65
4	<i>Lates niloticus</i>	64	53,33
5	<i>D.fasciatus</i>	46	38,33

USING INNOVATIVE e-DNA BASED APPROACH TO TACKLE CHALLENGES AND EVALUATE THE FOOTPRINT OF NORWEGIAN SALMON AQUACULTURE

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In Norway, the aquaculture industry has a significant socioeconomic value. The planned intensification of fish production from aquaculture requires improvement of production sustainability to respond to increased environmental pressure. Today, parasitism and diseases causing significant fish production loss, are mitigated by methods causing welfare and environmental concerns. Production in larger offshore farms is envisioned, but the consequences for the offshore marine life will need documentation. Also, the methods used to monitor the footprint for smaller production sites may need a revision for offshore production. Overall, there is a need to document effects of and tolerance limits for impact from present and future aquaculture using timely methods that can address these important issues. Because of versatile applications, environmental DNA (eDNA) monitoring is seen as one way to inform about critical occurrence of pathogens to the farm operators and to monitor the footprint on sediment and water close to aquaculture installations. eDNA monitoring, particularly in marine environments, requires quite extensive logistics that increases the sampling cost. It is therefore an expressed interest in developing automated concepts for aquatic eDNA monitoring. In parallel, both manual and automated eDNA concepts need to prove equally robust as standard taxonomy-based methods for monitoring of biodiversity and disease pathogens. The Environmental Sampling Processor (2G-ESP, developed at MBARI, USA. See illustration), is a fully automated sampling and processing platform for *in-situ* detection and quantification of several agents in water. Sample archiving is also possible. The results can be remotely communicated to decision-makers within 1-day. ESP was tested at 6 m depth, downstream a fish farm in western Norway for 3 weeks. Target organisms were the salmon louse (*Lepeophtheirus salmonis*) and the amoebic gill disease (AGD) pathogen (*Paramoeba perurans*). eDNA from salmon fish in the pens was used as a “positive control” for the ESP and to infer about current direction to the ESP. Overall, both *in situ* detection and quantification of eDNA fluctuated over the period: AGD and fish eDNA were detected in some samples, salmon lice was not. Several variables may explain this discrepancy. The archived samples are now being processed as well. New compact and portable devices such as the Biomeme qPCR device and the Oxford Nanopore mini-sequencer will now be evaluated for specific biological indicators, disease-agents, and marker gene biodiversity of taxonomic groups for footprint community assessment around farms. eDNA -based monitoring and semi- to full automatization of procedures may change the way monitoring of Norwegian salmon farm to support more sustainable production in the future.



***Aeromonas hydrophila*; FISH DISEASE AND CONTROL STRATEGY IN SOUTH SUDAN**

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This paper focused on infectious diseases among fish in South Sudan aquaculture and their impact on fish and human life, as well as the various interventions that have been used to prevent and control these diseases. Regardless of the culture system used, it is imperative that the culturist maintains an environment conducive to good fish health. However, fish farming conditions are often conducive to the spread of disease. The objectives of control measures for infectious diseases are to:

- ✓ Reduce or eliminate the source of infection
- ✓ Break the connection between the source of infection and susceptibility of fish.
- ✓ Reduce the susceptibility of fish to infection.

Important fish disease in fresh water and disease cause by *Aeromonas hydrophila* is also Known as motile *Aeromonas* Septicemia, therefore the clinical sign can cause the dropsy Ascites/Fluid and also can cause serious damage in internal organ, also damage in Kidney. As can be seen from the above review, bacteria are responsible for many diseases and heavy mortality in cultured fish. Pathogen isolated in characters of *Aeromonas hydrophila* incubating on gram negative and gram positive.

PREDICTING AND IMPROVING THERMAL TOLERANCE WITHIN A COMMERCIAL ATLANTIC SALMON *Salmo salar* BREEDING PROGRAM

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The marine (post-smolt) phase of the salmon production cycle typically exposes fish to seasonal temperature fluctuations that are potentially harmful, a concern that is exacerbated by anthropogenic climate change (i.e., global warming). Selection for thermal tolerance could present a successful long-term mitigation strategy. To this end, we examined individuals from 105 families from a single year class from an Atlantic salmon breeding program collaboration (Mowi Canada East and Huntsman Marine Science Centre) to: (1) develop a suitable method for explicitly testing climate change thermal tolerance and (2) determine the level of variability and heritability for relevant traits, and correlations between them, to allow selection.

It is imperative that broodstock programs collect relevant data to measure performance for the specific traits of interest. Our inclination was to test temperature tolerance using the critical thermal maximum challenge (CTmax) given its relatively quick and non-lethal biological endpoint. However, this approach does not use real-world rates of temperature increase as experienced in marine farm operations. Therefore, our initial efforts had to compare individual fish CTmax performance with that from the long-term and lethal incremental thermal maximum challenge (ITmax).

We tested 1,503 individually tagged Atlantic salmon over 126 CTmax trials using an increase of 0.4 °C/min to loss of equilibrium. Following recovery, 765 of these fish and 127 naïve fish were entered into a single ITmax challenge with a more gradual temperature increase of 1.0 °C/day to 16 °C and then 0.3 °C/day to reach morbidity/mortality of the entire population. Individual fish performance from the CTmax challenges had no predictive value for individual performance in the more realistic ITmax challenge (linear regression: $p = 0.22$, $r^2 = 0.002$). These results were consistent when assessed using pedigree information, with genetic and phenotypic correlations of -0.01 ± 0.16 and 0.07 ± 0.05 , respectively (\pm SE). Heritability estimates were high for both challenges: 0.47 ± 0.08 and 0.40 ± 0.08 , respectively ($h^2 \pm$ SE).

Our climate change tolerance research conclusions to date are that:

1. Completing ITmax challenges are required in favour of CTmax trials to measure the effects of climate change on farmed Atlantic salmon populations;
2. Estimated heritabilities are quite high thereby allowing effective selection to increase seawater temperature tolerance due to climate change; and,
3. Genetic and phenotypic correlations between several measured traits indicate that both increases in seawater weight (growth) and increasing seawater temperature tolerance can be selected and improved simultaneously.

Aspects of this presentation were recently published <https://doi.org/10.1016/j.cbpa.2022.111143>.

AN INTRODUCTION TO THE PRODUCTION AND USE OF TRIPLOIDS IN AQUACULTURE

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The ‘blueprint of life’ is encoded by DNA. Most of the DNA that makes up an animal’s genome is contained within chromosomes that are located within the nucleus of every cell. Although chromosome number varies among species, they are typically found in pairs (‘homologues’), with one homologue of each pair inherited from each parent. Such animals are therefore referred to as ‘diploid’ because each nucleus contains two complete sets of homologous chromosomes. Adding a third set makes the animal ‘triploid’ and generally results in sterility. Whether produced through triploidy or some other means, such sterile animals have clear advantages for aquaculture whenever reproduction affects valuable production traits (e.g., growth, appearance, meat quality, or immunocompetence) or poses risks to the environment (e.g., escapees interbreeding with wild stocks or establishing feral populations). Triploidy has been assessed for practical application in many aquaculture species, including fish, bivalve molluscs, and shrimp, but there are few examples of successful adoption for commercial production. This presentation will introduce the non-specialist to the methods used for producing triploid populations and a summary of their pros and cons for aquaculture.

IT MUST BE SOMETHING IN THE WATER: THE IMPACT OF THE OYSTER PARASITE *Haplosporidium nelsoni* TO OYSTERS AND PEOPLE OF THE BRAS D'OR LAKE CAPE BRETON, NOVA SCOTIA AND THEIR COLLECTIVE EFFORTS FOR SURVIVAL

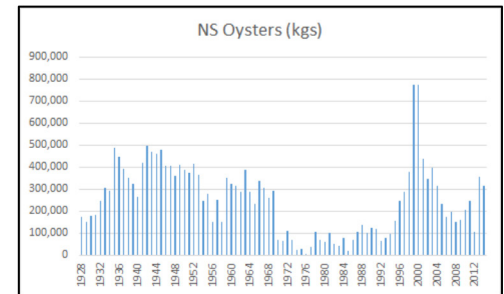
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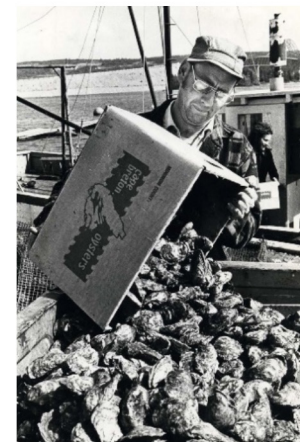
In 2001-02 the Bras d'Or Lake, Cape Breton, Nova Scotia oyster industry suffered catastrophic losses from the lethal oyster parasite, *Haplosporidium nelsoni* (MSX). This parasite has caused devastating losses along the United States eastern seaboard for decades and oyster populations are slow to recover. The presence of *H. nelsoni* in the Bras d'Or Lake caused an 80% reduction in oyster production for Nova Scotia. Over the past years, there have been small-scale research projects in the Bras d'Or Lake focused on possible measures for mitigation strategies and gaining a better understanding of the parasite. Collectively, these projects provided insight into rebuilding this industry. In 2018, funding was obtained for a 3-year project involving oyster leaseholders from across the Bras d'Or Lake to monitor growth, mortality, and disease assessment. This industry-focused project utilizes suspension culture technology in various locations throughout the Bras d'Or Lake while collecting information about the parasite, host, and environmental parameters to determine if environmental conditions can be used to mitigate losses from the parasite.

A fundamental aspect of this research effort is the perseverance and cooperation of oyster leaseholders from across the Bras d'Or Lake. Many are from families that have grown oysters for generations. Their collective passion for this animal, industry, and Island has lead to new initiatives and energy to revive this industry. Despite almost 20 years of loss, these champions of the Bras d'Or Lake continue to recognize the value oysters play historically, ecologically, economically, and culturally. Their relationship to oysters can only be described using their own words, so we will share some of their stories of survival.

The final part of the presentation will describe the science project itself in some detail, and will emphasize that a project of this scale and complexity could not have been undertaken without the support and commitment of the individuals who make up this industry live on this island, and have such a strong connection to the oyster.



Nova Scotia Oyster Landings – 1928-2014



Oyster farming in Eskasoni, ca. 1980

SUSTAINABLE AQUACULTURE – NEXT GENERATION OF AQUAFEEDS AND WASTE STREAM VALORIZATION

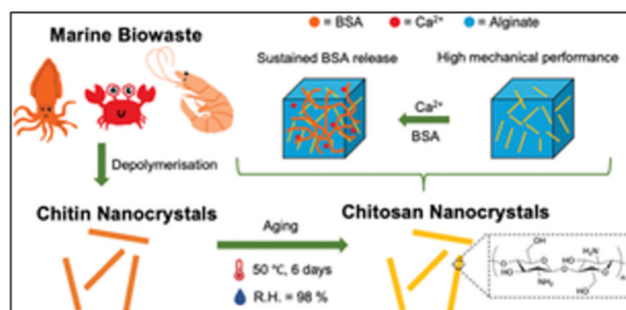
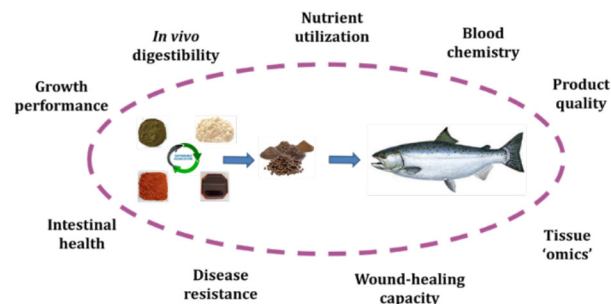
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The National Research Council of Canada (NRCC) has a long history in marine biosciences, relying on unique infrastructure including the marine research station (MRS) in Ketch Harbour and expertise in algal research, bioprocessing, and the development of natural products with commercial applications. More recently, in response to the creation of the federal Ocean Supercluster (OSC), NRC launched in 2020 its NRC Ocean Program “Ocean Wealth – Ocean Health” to provide R&D support to OSC members and the scientific ocean community, largely to help strengthen a sustainable blue economy in Canada. As one of the fastest growing agri-food sectors, aquaculture is playing a critical role in global food production, but innovation and equity are required to ensure inclusive and sustainable growth of the industry. In that context, NRC has been investing and focusing its efforts on two research areas to 1) address the needs for novel and more sustainable sources of proteins, lipids and carotenoids for the development of the next generation of aquafeed as well as 2) develop innovative solutions to transform and convert waste streams into value-added products.

fter an overview of the NRC’s Ocean program and introduce opportunities for collaborations, this presentation will describe three flagship projects representing NRC’s research in:

- 1) the development of low-trophic microbial ingredients for aquaculture feeds,
- 2) the conversion of marine waste into functionalized chitin and chitosan nanocrystal with potential biomedical applications, and
- 3) the isolation and application of cold-adapted proteases for the bioprocessing and valorization of marine waste.



SUPPLEMENTING RAINBOW TROUT BROODSTOCK DIETS WITH CHOLINE AND METHIONINE IMPROVES OFFSPRING GROWTH

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The objective of this study was to determine whether supplementing female rainbow trout broodstock diets with the epigenetic modifiers choline, methionine, or a combination affects offspring growth performance. Four treatment diets were produced by top-dressing a commercially available finfish broodstock diet (38% protein, 10% fat) with 1) choline (final 7,400 ppm), 2) DL-methionine (final 12,000 ppm), 3) choline + DL-methionine, 4) distilled water (served as the control). Six families of female rainbow trout were fed treatment diets at or just below satiation beginning 18 months post-hatch through spawning (n=3 tanks per diet). Eggs were collected and fertilized using milt from a single family of unrelated males. Offspring families were reared individually until they were PIT-tagged at approximately 10 g and comingled in three triplicate tanks. Offspring consumed a single commercially available diet (42% protein, 16% fat) and lengths and weights were recorded every 2 months.

Broodstock diet did not affect maternal body weight, body condition, egg size, or egg yield at spawning. However, there was an effect of broodstock diet on offspring growth performance that was age dependent (Table 1). Offspring fry from choline-treated broodstock (diet #1) were smaller than fry from other treatment groups through 146 days post hatch (dph, ~11g). However, by 259 dph, offspring from broodstock that received choline (diets #1 and #3) exhibited greater body weights than offspring from broodstock that received the control feed (diet #4). By 377 dph body weights of offspring from methionine-treated broodstock (diet #2) also exceeded controls. Overall, at final harvest (439 dph) offspring from control broodstock weighed 12-18% less than offspring from broodstock consuming supplemental diets, indicating that supplementing broodstock diets with methionine and choline causes a nutritional programming effect that benefits growth in offspring.

Broodstock Diet	days post-hatch						
	90	146	193	259	316	377	439
1. Choline	3.29 ^c	10.9 ^b	43.7 ^b	133.7	329.1 ^a	579.9 ^a	938.6 ^a
2. Methionine	3.70 ^b	11.3 ^{ab}	43.8 ^b	131.4	321.2 ^{bc}	573.1 ^{ab}	926.5 ^a
3. Choline+Met	3.93 ^a	11.7 ^a	45.8 ^a	138.3	342.1 ^a	603.6 ^a	971.7 ^a
4. Control	3.59 ^b	11.8 ^a	43.8 ^b	128.8	307.3 ^b	540.4 ^b	807.2 ^b
Pooled SEM	0.08	0.30	1.0	3.4	7.5	13.1	27.2

Table 1. Effects of broodstock diet on body weight (g) of offspring throughout development. Different letters indicate a significant difference between mean body weights within the same column, $P < 0.05$.

SENSITIVITY OF A LAMP ASSAY FOR DETECTION OF THE DINOFLAGELLATE *Amyloodinium ocellatum* IN SIMULATED FIELD CONDITIONS

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Amyloodinium ocellatum is a cosmopolitan, ectoparasitic dinoflagellate that infects a wide variety of marine and brackish-water fishes. The life cycle of the parasite is direct and consists of three stages: the trophont which attaches to and feeds on the host, the tomont which is free-living and reproductive, and the dinospore which hatches from the tomont and infects new hosts. Each tomont can produce up to 256 dinospores. Because of its tolerance to a wide range of temperatures and salinities, its lack of host specificity, and its high reproductive capacity it is considered one of the most troublesome parasites in warm water marine aquaculture.

Early diagnosis of infections is critical to the effective management of outbreaks considering the prolificacy of the parasite. The current gold standard for diagnosis is microscopic examination of gill tissue, but light infections may be missed, leading to false-negative outcomes and/or other organisms may be misdiagnosed as *Amyloodinium*, leading to false-positive ones. Moreover, microscopy cannot detect the parasite in the water prior to establishment of the infection. Both a conventional PCR and a LAMP assay are available. The LAMP is technically easier, quicker, highly specific, and more sensitive than PCR, but sensitivity was not assessed for detection of the parasite in either tissue or water beyond the benchtop scale. The objective of this study is to compare the sensitivity of light microscopy to that of the LAMP assay for detection of *Amyloodinium ocellatum* in gill tissue and water in simulated field conditions.

Forty-two 10-gallon aquariums were filled with 20 L of 25 psu artificial seawater and inoculated with either 0, 1, 10, 100, 1000, 5000, or 10,000 laboratory-grown dinospores (six tanks per dose), resulting in concentrations of 0, 0.05, 0.5, 5, 50, 250, and 500 dinospores/L, respectively. Ten 1-L aliquots from each of 21 tanks (3 tanks per dose) were removed and filtered on a 0.22 µm 47 mm nitrocellulose filter membrane and stored at -80 °C. The other 21 tanks (3 per dosage) received four naïve spotted seatrout (*Cynoscion nebulosus*) reared from captive broodstock spawns in recirculating systems. After 7 days or upon fish death, 4 gill arches per fish (2 from each side of the fish) were examined via microscopy then preserved in 95% ethanol for DNA extraction and analysis with the LAMP assay.

The lowest dinospore concentration that could be detected by the LAMP assay in water samples was 0.5 dinospores/L, while positive detections occurred only at 5 dinospores/L or higher when using LAMP or microscopy on gill samples. Thus, LAMP of water samples is a superior diagnostic tool. Simulations of replicated assays indicated that a 95% probability of detection was achieved with 10 replicated assays of water samples when the dinospore concentration is as low as 1 dinospore/L.

SUSTAINABLE DEVELOPMENT IN BIOTECHNOLOGY AND AQUACULTURE

Bolaji Opeyemi*, Olatunji Anuoluwapo

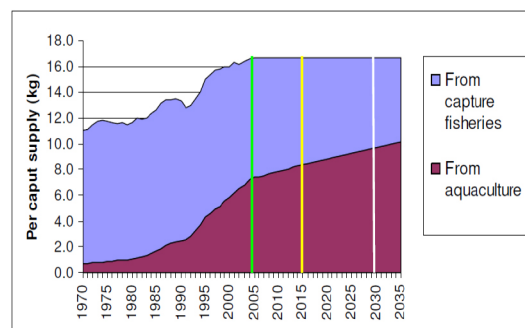
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Aquaculture is the quickest developing meal zone globally with its growing position in the financial system and secure meal method for countries. Due to the persevering depletion of the fish shares, farming of aquatic organisms including fish, crustaceans, molluscs, and aquatic plants, is now a good-sized international enterprise providing a wide percentage of the aquatic merchandise consumed. Shortage in meal delivery and excessive costs are the viable essential dangers in the future, and aquatic merchandise are the precious assets of protein and critical nutrient additives for international meal safety and doing away with malnutrition.

Aquaculture additionally performs an essential position in rural economies via the introduction of recent employment. In those cases, aquaculture outputs will want to be improved numerous fold so one can meet the growing needs for fish and different aquatic merchandise in the coming years.

Biotechnology alternatively appears to be the suitable capability for growing aquaculture productivity, meal safety, and environmental exceptions worldwide. Biotechnology is imparting precious alternatives including protein expression, microsatellite, gene mapping and genomic, DNA vaccines, DNA chips, proteomics, transgenic era, and embryonic stem molecular era. This era gives genetic manipulations, molecular cloning, embryo manipulation, genetically-engineered diagnostics, and immunoprophylactic agents. All of those programs may want to assist enhance the selective breeding, hybridization, productivity, health, growth, nutrition, cryopreservation, and conservation of genetic assets in aquaculture shares for the gain of mankind.

However, there's a want for the law of biotechnology sports in phrases of the capability detrimental influences on the surroundings and human health. There is likewise growing difficulty approximately the effect of biotechnology on sustainable improvement in numerous fields. The fundamental environmental protection trouble of those programs is the outcomes of genetically changed organisms (GMOs) on biodiversity and gene switches inside the surroundings. Therefore, this overview mentioned the combination of biotechnology and biosafety in aquaculture and rules for the environmentally sound use and control of aquaculture biotechnology in sustainable improvement.



SUBLETHAL EFFECTS OF LOW-DOSE EXPOSURE TO IMIDACLOPRID ON THE AMERICAN LOBSTER *Homarus americanus*

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Atlantic salmon farms face the ongoing challenge of managing sea lice infestations due to reduced sensitivity to extensively used chemotherapeutants. Imidacloprid is a neonicotinoid insecticide that has been implemented as a novel chemical to mitigate sea lice infestations on European Atlantic salmon farms. Prior to its consideration for use in the Northwest Atlantic, we tested the effects of these imidacloprid waste-water and dispersal concentrations on the economically valuable nontarget species, the American lobster, *Homarus americanus*. Behavior and hemolymph biochemistry of sub-adult female lobsters were assessed following a 120-minute exposure to the imidacloprid concentrations 0, 0.3 or 30 ppb. Observations were repeated after a five-day recovery period to evaluate chronic effects. Defensive behaviors were found to be significantly reduced for lobsters exposed to the 30 ppb imidacloprid concentration, and some remained significantly reduced five days after exposure. Interestingly, overall hemolymph endpoints indicative of stress (L-lactate, crustacean hyperglycemic hormone, and total protein) were not significantly different across treatments. This highlights the importance of behavioral endpoint observations, as limited upstream endpoints may not always capture the full impact downstream. These findings suggest that lobsters exposed to low environmental concentrations of imidacloprid may have impaired behaviors of ecological and economic importance.

TENACIBACULOSIS IN ATLANTIC SALMON (*Salmo salar* L.)

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The aetiological agent of Tenacibaculosis (or yellow mouth) *Tenacibaculum* spp. infect multiple fin fish species globally and is a major pathogen of sea bass (*Dicentrarchus labrax*), sea bream (*Sparus aurata*), turbot (*Psetta maxima*), and Atlantic salmon (*Salmo salar* L.) aquaculture. In Atlantic salmon, the disease and associated mortalities occur both in smolts soon after transfer to sea, and in larger adults. While the disease exhibits variable pathological manifestations, yellow mouth lesions, fin erosion, and skin lesions are characteristic of the disease. In Canada, the disease causes significant losses to the industry and is a priority issue with annual cost of outbreaks for one Canadian company cited as \$1.6 million. Moreover, instances of outbreaks in other geographical regions, e.g. Chile, Norway, and Scotland have increased in recent years. At current, there is no commercially available vaccines for use in salmonids and a reliance on therapeutic use of antibiotics has led to a strong need for the industry to establish efficacious alternatives.

Multiple isolates of *T. maritimum*, *T. finnmarkense*, and *T. dicentrarchi*, and one potentially novel species of *Tenacibaculum* originating from Western Canada (BC) were utilised in these studies. Investigations focussed on understanding the virulence, and compound factors that influence virulence, of the different isolates and species. Furthermore, the clinical presentation of the isolates, and understanding how challenge methods can be manipulated for the testing of different technologies and treatments are explored.

Isolates, within and between, species of *Tenacibaculum* have significantly different virulence and clinical presentations. Furthermore, it has been identified that while most environmental, and other compound factors, influence infection and disease outcomes some are unique. The development of these disease models is an important step in the development of technologies to mitigate its impacts on the culture of Atlantic salmon. However, Tenacibaculosis is a complex disease, and often is not exclusive to the presence/ infection of fish by a single species. Therefore, current developments are focussed on multiple species infection models for use in the industry.

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GENOMIC EVIDENCE OF RECENT EUROPEAN INTROGRESSION INTO NORTH AMERICAN FARMED AND WILD ATLANTIC SALMON

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Escape events from Atlantic Salmon, *Salmo salar*, net-pen aquaculture occur regularly and evidence from across the North Atlantic has repeatedly demonstrated interbreeding and subsequent introgression between wild Atlantic Salmon and escaped domestic individuals. The genetic impacts of gene flow from domestic into wild salmon populations depends both on the degree of domestication and the original source of the domesticated population. Atlantic salmon used in North American aquaculture are ostensibly of North American origin. However, evidence of European introgression into North American aquaculture salmon has accumulated in recent decades, yet the full extent as well as the potential impacts on wild salmon in the Northwest Atlantic remains uncertain.

Here, we extend previous work comparing North American and European wild salmon (n=5799) using a 220K SNP array to quantify levels of recent European introgression into samples of aquaculture salmon, aquaculture escapees, and wild salmon collected throughout Atlantic Canada. Analysis of North American farmed salmon (n=403) and escapees (n=289) displayed significantly elevated levels of European ancestry by comparison with wild North American individuals ($p < 0.001$). Of North American farmed salmon sampled between 2011 and 2018, ~17% had more than 10% European ancestry and several individuals exceeded 40% European ancestry. Samples of escaped farmed salmon similarly displayed elevated levels of European ancestry, with two individuals classified as 100% European. Analysis of juvenile salmon collected in rivers proximate to aquaculture locations also revealed evidence of elevated European ancestry and larger admixture tract in comparison to individuals collected at distance from aquaculture.

Overall, our results demonstrate that individuals of full and partial European ancestry have been in use over the last decade and that some of these individuals have escaped and hybridized with wild salmon throughout Atlantic Canada. Ultimately, the presence of trans-Atlantic hybridization in aquaculture salmon in Atlantic Canada elevates the potential impact and uncertainty regarding the risk posed by escapees to wild and often at-risk salmon populations.

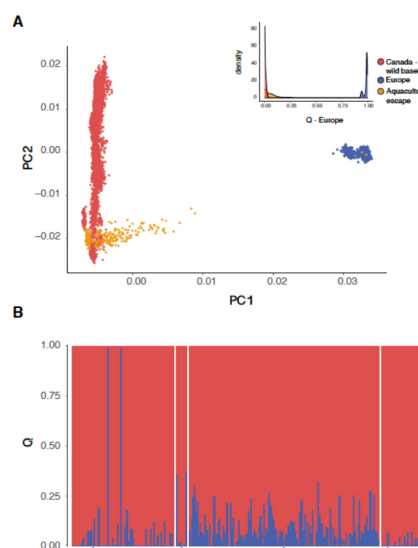


Figure 1. Genomic evidence of European ancestry in escaped farmed salmon from southern Newfoundland

**OVERWINTERING SURVIVAL IN JUVENILE OYSTERS: UNDERSTANDING THE ROLE OF
ENERGETIC PHYSIOLOGY FOR *Crassostrea virginica* SEED**

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The American oyster *Crassostrea virginica* is an important cultivated oyster species, providing every year several millions of dollars to fishery and aquaculture economies. This species expands over 4 000 kms on the East American coast, from the Gulf of Mexico to the Gulf of St-Lawrence where oysters experience harsh winter conditions such as sub-zero temperatures during 3-4 months. In response to this temperature constraint, adult oysters decrease the unsaturation index of their polar lipids during the fall acclimation period to adjust the fluidity of their cell membranes and progressively reduce their metabolism to a near standstill during winter. Throughout this dormancy phase, *Crassostrea virginica* stops feeding and mobilize its energetic reserves (lipids and to a lesser extent proteins) to maintain vital physiological functions. However, the metabolic strategies of oyster spat during winter are still understudied, although they represent a crucial stage determining the production of adult oysters and economic performances of hatcheries.

This study aims to investigate the link between survival rates of hatchery and wild juvenile oysters in relation to their size and energetic reserves. An 8-months experiment is being conducted from September 2021 to May 2022 to monitor energetic reserves and lipid dynamics of *Crassostrea virginica* spat from the fall acclimation to the end of winter dormancy. Juveniles from two origins (i.e. hatchery vs wild) and two sizes (i.e. 4 vs 6 mm) are reared both in hatchery under ambient condition (experimental group) and in the wild (control group). Total energy content is estimated through total lipids, proteins and carbohydrates analyses. In addition, lipid classes and fatty acid composition of neutral and polar lipids are analyzed to examine the dynamic of storage lipids and track seasonal metabolic adjustments in oyster spat.

According to first preliminary results, oyster spat have already low energetic reserves during the fall acclimation period (Fig. 1). However, no differences are observed between hatchery and wild juveniles, suggesting that hatchery seeds used in aquaculture have a similar physiological performance than those collected in the wild. All the results obtained at the end of the experiment will allow to better understand the energetic physiology of oyster spat during winter. This study will also provide new insights to oyster producers to predict the overwintering success of juvenile oysters and maximize winter survival by improving their diet in hatchery before release into the wild.

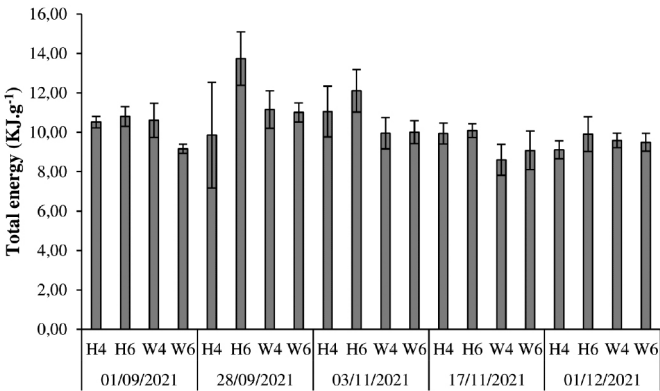


Fig. 1: Total energy content of 4 and 6 mm juvenile oysters from hatchery- (H4 and H6, respectively) and wild-origin (W4 and W6, respectively).

THE SIGNIFICANCE OF INTRODUCING A DESIGN CODE TO THE AQUACULTURE INDUSTRY IN NORWAY AND THE EFFECT ON FISH ESCAPE

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Introduction

From the initiation in the 70s to the end of the 90s there was an accelerating development in the growth of the aquaculture industry in Norway. It was observed that the increase in activity led to an increased number of incidents of structural failures leading to fish escape. This led to the development of regulations to make the aquaculture systems more reliable.

Regulations and design code

In 2003 the first revision of the NYTEK-regulations and the relating standard NS9415 was ready to be implemented. Further with the introduction of the aquaculture act in 2005 the authorities got tools that helped improve the robustness of the aquaculture plants and decrease the risk for fish escape. The NYTEK-regulation and NS9415 has been through revisions and is currently being revised.

Effects on fish escape

The regulations design code has proven to be effective as the number of fish escapes have plummeted in a period when the production volume has nearly tripled. The number of escapes due to system collapses are close to none and the few fish escape that happen is related to operations and human error.

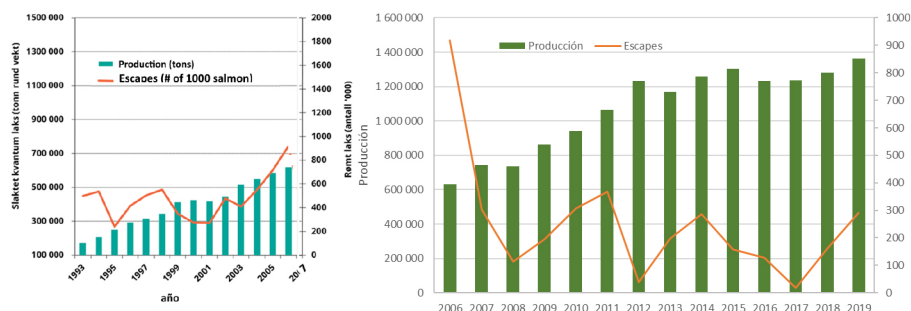


Figure 1: Historical relation between production and fish escape

FEEDING CURIOSITY: ENGAGING CONSUMERS THROUGH STORIES OF INNOVATION AND SUSTAINABILITY

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The Canada Agriculture and Food Museum is one of three national museums of science and innovation. With a mandate to foster scientific literacy and a mission to be a catalyst for curiosity, the museum has an ongoing commitment to exploring one of the key issues of our time: food security. With the Decade of Ocean Science underway and with the United Nations General Assembly declaring 2022 as the International Year of Artisanal Fisheries and Aquaculture, the museum has recently undertaken a six-year initiative to bring greater awareness and understanding to a branch of agriculture that is often overlooked and misunderstood by consumers – Aquaculture.

With support from Fisheries and Oceans Canada, a national advisory council, and producers, researchers, and communities throughout the country, this topic will be in the spotlight for audiences of all ages and backgrounds. Engaging consumers in conversations about a topic as complex and important as aquaculture could be a challenge given the diversity of opinions surrounding protein sources, environmental impacts, and cultural connections. To this end, the museum and its partners have agreed that the guiding principle for this initiative will be that innovation (past, present and future) will help secure a sustainable future for aquaculture in Canada. In addition, the Aquaculture Initiative will use the lens of the United Nations Sustainable Development Goals to ensure that the content is both inclusive for and relevant to all visitors. To this end, the museum and partners will explore this theme through a long-term exhibition at the national museum, a travelling exhibition that will be on loan to communities from coast to coast to coast and public programming to reach local, domestic and international audiences.

Farming the Waters will take visitors on a journey to learn how aquaculture grows a variety of organisms for food and products in Canada – fish, shellfish, and seaweed. From Indigenous clam gardens to the latest recirculating water systems, visitors will appreciate that aquaculture is a pursuit that requires deep knowledge about our waters and the life within them. Visitors will also learn that scientists, Indigenous communities, and other aquaculture operators are leading Canadian innovation in this global industry, providing livelihoods and income for communities throughout the country. Innovation is making this industry even more sustainable as consumer demand for healthy seafood, grown in environmentally sound ways, increases.

This presentation will include results from formative surveys, consultations, and testing with a broad-spectrum audience and will share an informal science learning approach that is already resonating with visitors and sparking dialogue about an industry that is both centuries-old and constantly evolving.

REAL-TIME DISSOLVED OXYGEN MONITORING IN A NOVA SCOTIA ATLANTIC SALMON FARM

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As the global population continues to grow and fisheries collapse, the sustainable growth of aquaculture is more important now than ever. Traditional land farming has had centuries to develop new technologies especially through the “Green Revolution” of the mid-20th century; the next logical step is for a “Blue Revolution” to match that. Recently, an important area of focus for technological advances has been on water quality monitoring to improve aquaculture management. For example, farmers can now employ dissolved oxygen sensors that report in real-time to monitor their farms with precision; however, their use is still in its infancy and further research is needed to fully operationalize them. In this study, 60 InnovaSea, real-time dissolved oxygen and temperature sensors were distributed throughout 19 cages at a salmon farm located within Shelburne Bay, Nova Scotia (Canada). The dense deployment allowed for insight into the spatial and temporal variability of dissolved oxygen throughout a farm. Tide was determined to have the most significant impact on dissolved oxygen levels, with its influence varying depending on cage location within the farm. As highly oxygenated waters flow from one end of the farm to the other, driven by tidal advection, fish respiration and flow restriction from cage infrastructure reduce oxygen levels. This results in oxygen concentration out of phase in opposite sides of the farm, with high oxygen levels at one end of the farm and low oxygen levels at the other (Figure 1). Since farmers use low oxygen as an indicator as to whether to feed, it is important to understand oxygen dynamics through the farm and across cages to make informed decisions that impact fish growth and welfare.

USE OF MICROALGAE AS AN ASTAXANTHIN SOURCE IN ATLANTIC SALMON (*Salmo salar*) FEED

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Cultured salmon (*Salmo salar*) is an important carnivorous fish with an estimated 2.5 million metric tons in 2018. The pink to red color of the fillet is one of the most important quality criteria for salmon. This pigmentation is due to deposition of carotenoids (mainly astaxanthin) in the muscle of salmon, and to a lesser degree canthaxanthin, in farmed salmon. Since salmon is unable to synthesize astaxanthin, the carotenoids originate from micro-algae in the wild, or are synthetically produced for inclusion in the diets of cultured fish.

Two 30-day stability tests were conducted with microalgae (*Haematococcus pluvialis*) containing high levels of astaxanthin. Microalgae was dried or encapsulated in 1, 2, or 3% sodium alginate at a concentration of 1, 2 or 3%. The samples were then either kept in the dark in a cool room where feed is normally stored or heated for 5 min at 150°C and then stored as described. The encapsulated samples that contained the highest levels of microalgae and lowest levels of alginate had the greatest astaxanthin degradation (samples 6, 8, and 9) compared to the lowest levels of microalgae and highest levels of alginate (samples 1, 2, and 4) and the dried algae alone (sample 10). An *in vivo* and *in vitro* study will be conducted to determine if Atlantic salmon can access the astaxanthin from the encapsulated algae.

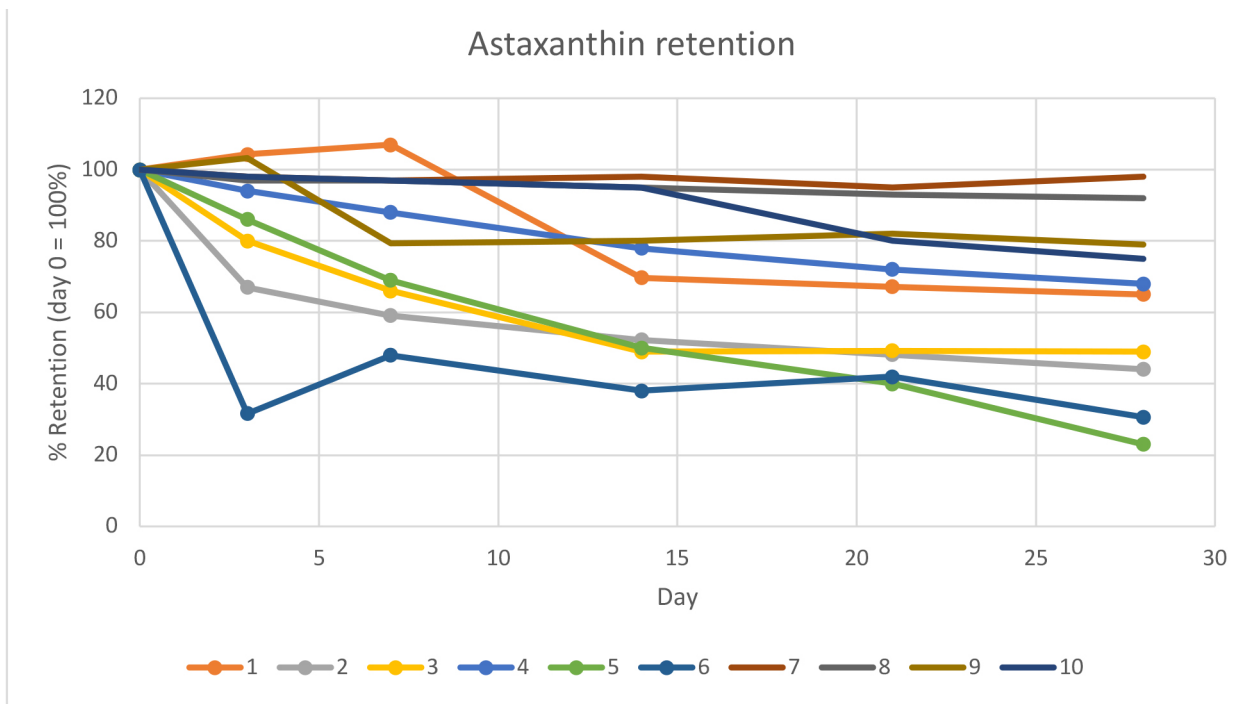


Figure 1. Astaxanthin retention, compared to the original amount of astaxanthin, in samples after heating.

DIETARY IMMUNOSTIMULATION: A POWERFUL TOOL AGAINST *Caligus rogercresseyi* AND *Piscirickettsia salmonis* CO-INFECTION IN FARMED ATLANTIC SALMON

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The co-infection of *Caligus rogercresseyi* –a parasitic copepod– and *Piscirickettsia salmonis* –the causative bacterial agent for salmonid rickettsial septicemia (SRS)– is a recurrent problem in Chilean salmon aquaculture. Clinical diets are valuable in preventing disease in farmed fish and may reduce the use of less sustainable and more aggressive fish health management procedures (e.g., antibiotic and delousing treatments).

We tested the immunostimulatory activity of EWOS-BACTER (Cargill Inc.) in salmon smolts exposed first to *P. salmonis* (via cohabitation) and subsequently to *C. rogercresseyi*. On day 41 post-*P. salmonis* introduction, and day 12 post-*C. rogercresseyi* exposure, BACTER diet had improved survival, and weight gain in co-infected salmon compared with co-infected salmon fed a control grower diet (CONTROL). Co-infected salmon fed BACTER showed less severe clinical signs (e.g., petechial skin, ascites) and reduced lice load than their CONTROL-fed counterparts.

The head kidney transcriptome response to co-infection and its dietary manipulation were investigated via RNA-Seq. Co-infection caused the differential regulation of 13,823 genes (False Discovery Rate <5%, fold-change >1.5) in the CONTROL-fed salmon. BACTER modulated the transcript levels of 1,857 genes within the co-infected salmon. The conjoint analysis of all differentially expressed genes identified two main clusters: one with 9,679 infection-repressed genes representing primarily tissue development and transforming growth factor beta-mediated processes (Gene Ontology enrichment analysis), another comprising 8,344 infection-induced genes, most of them putatively involved in inflammatory and cytotoxic immune processes. Based on this classification, the head kidney transcriptome of BACTER-fed co-infected salmon appeared more polarized towards tissue restoration than the CONTROL-fed co-infected salmon.

These results stress the importance of clinical feeds to control co-infection in Atlantic salmon farming operations. We estimate this research will improve our understanding of the molecular changes experienced by Atlantic salmon co-infected with *P. salmonis* and *C. rogercresseyi* and will provide biomarker genes and pathways leverageable for improving fish health management practices.

CERTIFICATION IN AQUACULTURE – LESSONS LEARNED AND FUTURE TRENDS

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Aquaculture Certification has developed over the last 20 years to give assurances on a range of topics such as Food Safety, Animal Welfare, Environmental, Social Responsibility and Traceability. With certified production growing at an average rate of 76% annually in the last decade, third-party aquaculture certification and eco-labels have made strides in gaining influence and market place recognition, despite only 6% by volume of global aquaculture production being certified. Compared to other countries, Canada has a high percentage of certified product, but how does this compare with other regions? Are third party certification standards fit for purpose and does their complexity offer greater assurance to the buying public, or is it all about market access? Recent developments using technology to assist with the certification process should be adopted to reduce time and costs for the Industry. Third party accredited certification is just one form of assurance and in the last 2 years, other assurance models have evolved and need to be developed to offer greater volumes of assured global aquaculture production.

ISOLATION AND CHARACTERIZATION OF AN ANTIGEN OF OUTER MEMBRANE VESICLES FROM THE FISH PATHOGEN *Moritella viscosa*

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Outer membrane vesicles (OMVs) are nano-sized proteoliposomes shed from the cell envelope of all Gram-negative species. OMVs are now recognized as a generalized secretion pathway that provides a means to transfer cargo to other bacterial cells and eukaryotic cells. OMVs play an important role in pathogenesis, delivering virulence factors to the host cells, including toxins, adhesins, and immunomodulatory molecules. *Moritella viscosa* is a Gram-negative pathogen that causes winter ulcer disease in several marine fish species. Here, we develop an infection model using lumpfish (*Cyclopterus lumpus*) and evaluate the toxicity and utilization of OMVs as vaccines. We isolated and characterized *M. viscosa* OMVs isolated from iron-rich and iron-limited growing conditions. OMVs were characterized by transmission electron microscopy and protein identification. Lumpfish injected intraperitoneally (i.p.) with 10^7 colony-forming units (CFU) fish⁻¹ of *M. viscosa* had rapid mortality and clinical signs after 3 days post-infection. *M. viscosa* OMVs in both conditions showed a spheric shape (39.8–370 nm diameter) that contains small RNA and DNA. The main OMVs proteins have a molecular size of 45, 30 and 20 kDa. OMVs isolated from iron-limited condition harbor an additional protein of approximately 60 kDa which is absent in OMVs isolate from bacteria grown under iron-rich conditions. The protein band of 60 kDa has several proteins, including a metal-dependent carboxypeptidase, glucose-6-phosphate isomerase, glucose-6-phosphate isomerase, peptide ABC transporter, extracellular solute-binding protein, oligopeptide transport system, and permease protein B. Current studies are being conducted to characterize *M. viscosa* OMVs cargo, toxicity, and utility as vaccine candidates.

ISOLATION AND CHARACTERIZATION OF OUTER MEMBRANE VESICLES FROM *Moritella viscosa* AND EVALUATED ITS IMMUNIGENICITY IN LUMPFISH (*Cyclopterus lumpus*)

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Outer membrane vesicles (OMVs) are nano-sized proteoliposomes shed from the cell envelope of all Gram-negative bacteria. OMVs are now recognized as a generalized secretion pathway which provides a means to transfer cargo to other bacterial, and eukaryotic cells. OMVs play an important role in pathogenesis, delivering virulence factors to the host cells, including toxins, adhesins, and immunomodulatory molecules. *Moritella viscosa* is a Gram-negative pathogen and the causative agent of winter ulcer disease in several marine fish species. *M. viscosa* OMVs have not been characterized and their role in the infection process and utilization as potential vaccine is unknown. Here, we develop a *M. viscosa* infection model in lumpfish, characterized the *M. viscosa* OMVs, and evaluate its toxicity and immunogenicity in in vivo lumpfish model.

We determined that the lethal dose (LD50) of *M. viscosa* in lumpfish was 8.1×10^7 colony-forming units (CFU) fish⁻¹. Lumpfish were intraperitoneally (i.p.) injected with 3.1×10^8 CFU fish⁻¹ showed a rapid mortality and typical clinical signs of winter ulcer disease after 5 days post-infection. Iron is an essential nutrient for microbes and influence pathogenesis. To characterize *M. viscosa* OMVs, the bacterial strain was cultivated under iron-rich and iron-limited conditions. Purified *M. viscosa* OMVs were characterized by transmission electron microscopy and protein analysis. *M. viscosa* OMVs from both growing conditions showed spheres of 39.8–370 nm diameter that contains RNA and DNA. The most abundant proteins in *M. viscosa* OMVs have molecular sizes of 45, 30 and 20 kDa. OMVs isolated from iron-limited condition harbor an additional protein of approximately 60 kDa which is absent in OMVs isolated from bacteria grown under iron-rich conditions. Protein identification analyses of the 60 kDa protein band indicated the presence of enzyme of Metal-dependent carboxypeptidase, Glucose-6-phosphate isomerase, Glucose-6-phosphate isomerase, and transport systems including the peptide ABC transporter, extracellular solute-binding protein, and oligopeptide transport system, and permease protein B.

The results strongly suggested that *M. viscosa* could induce the ulcer disease in lumpfish. *M. viscosa* OMVs contain various bacterial components, including proteins, DNA/RNA, and did not show toxicity in lumpfish. These OMV products may partially explain that they play an important role in the pathogenesis of *M. viscosa*.

STRATEGIES USING PROBIOTICS TO MITIGATE MICROBIAL DISEASE PRESSURE IN SHRIMP AND FINFISH AQUACULTURE PRODUCTION

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Disease pressure in aquaculture production continues to be a growing issue for the industry. Bacterial pathogens, such as species of *Vibrio*, *Aeromonas hydrophila*, and *Edwardsiella ictaluri* are known pathogens that infect shrimp and catfish, respectively for example. Pathogenicity is considered to be connected to their ability to concentrate in key organs, and once reaching critical levels, become virulent. Antibiotics have been used historically to try and control these effects. While it is debatable whether antibiotic use applied to pond systems is an effective treatment, there is growing recognition that their benefits are outweighed by concerns to eliminate their use.

Aquaculture production cannot be conducted in a sterile environment. Pathogens along with beneficial and innocuous microorganisms are always present in the microbiome, though not always virulent. Developing production strategies that mitigate the opportunistic growth of pathogenic bacteria through more holistic microbial management may present farmers with additional and, more sustainable, tools to suppress disease pressure and preserving production.

Probiotics are being broadly evaluated as an alternative management practice to antibiotic use. Research is now showing that the use of select probiotics in aquaculture production can suppress *Vibrio* growth in pond water, reduce their accumulation in the gut tract and exoskeleton of shrimp, and prevent problematic species from reaching critical virulent concentrations. Laboratory testing using proprietary blends of lactic acid bacteria and bacilli show that select strains of these species can broadly suppress *Vibrio* growth, as well as *Aeromonas hydrophila*, and *Edwardsiella ictaluri*. Growth experiments also show a lack of accumulation of *Vibrio* in shrimp during hatchery and nursery growth, as well as preliminary data for other pathogens in catfish. Field studies are now in progress to validate and extend these results from well controlled research studies to practical, cost effective tools for commercial farmers and production managers to use in improving aquaculture production.

BRAZILIAN AQUACULTURE: TOWARDS IMPROVED SURVEILLANCE, REGULATION AND MANAGEMENT OF WORKER HEALTH AND SAFETY

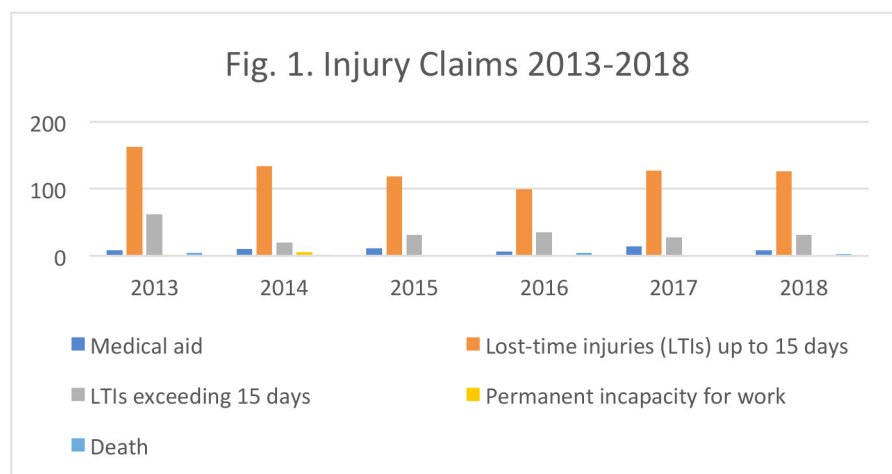
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In Brazil, all work accidents or illnesses related to work are supposed to be reported to the government for the purpose of guaranteeing access to benefits and protecting labor rights. Based on these reports, between 2013 and 2018, there were 1053 injury-claim in Brazilian aquaculture, of which 57 were for medical aid; 768 for lost-time injuries (LTIs) up to 15 days; 206 for LTIs exceeding 15 days and 9 permanent incapacity for work (fig. 1). The total number of fatal injuries between 2013 and 2018 was 13. As is the situation elsewhere, there is likely underreporting in the official statistical data for aquaculture. Brazilian workers who have an Employment and Social Security Record Booklet and contribute to social security are entitled to benefits related to an injury or an occupational illness. This benefit is granted by the government to those who are injured or made ill by their work and to the families of those killed at work, with an accepted claim for those who are unable to work for more than 15 consecutive days. In the first 15 days away from work, the company is responsible for paying the salary.

Social charges and taxes continue to be paid by the employer during the entire period of absence due to an injury at work. Currently, Brazilian regulatory standards are contained in texts designed to cover other rural sectors and pay little explicit attention to occupational health in aquaculture. This has a major impact on the effectiveness of worker protection measures, as well as on the ability to collect and monitor statistical data on injuries, fatalities and occupational diseases. This is because, contrary to what happens in regulations for other sectors, aquaculture does not have regulations that provide clear rules regarding worker safety and prevention. Brazil needs a greater commitment to OSH to improve and implement regulations. This requires an integrated system prioritizing prevention and using data on injuries and illnesses to help identify the most effective preventative measures available in the country. This could be done by a) establishing specific public policies and regulations for aquaculture OSH and b) providing training to collect accurate information on AOSH, as well as making that information available to regulators and others. Other alternatives would be c) to implement management systems and certifications that will ensure better OSH. External and internal audits, as well as inspections to promote the education and awareness of employers and workers, based on continuous quality assurance would also help. Taken together this collection of initiatives would support the development of a management model based on the One Health approach.



EFFECT OF A PROPRIETARY BLEND OF ESSENTIAL OIL BIOACTIVES ON STRESS RESPONSES OF *Salmo salar*

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Most salmon farming systems involve practices that expose animals to acute handling stressors, which have negative consequences on behavior, growth, performance and survivability. Sedatives or anesthetics are commonly administered to fish to mitigate these consequences (Aydin & Barbas, 2020). Nevertheless, these products can cause mortality and damage to fish. The present experiment aimed to study the effect of an antistress functional feed additive (AFFA) mainly based on proprietary blend of essential oil bioactives, on the stress induced by handling and crowding subsequent stages.

500 Atlantic salmon (*Salmo salar*) with an average weight of 196.74 ± 2.88 grams were equally divided into 5 tanks at a stocking density of 30.27 kg/m^3 . The fish received a standard pelleted diet and after a 14-day acclimatation period, each tank labeled from A to E was exposed to the following in-water concentration of AFFA; A-0 ppm (negative control), B-20 ppm, C-40 ppm, D-60 ppm and E-20 ppm at time 0 min + 20 ppm at 2 hrs. The fish were transferred to external containers 35 min after the addition of the AFFA and crowded to a density of 85 Kg/m^3 to reproduce typical conditions of salmon transportation. These containers contained the same concentration of AFFA as the original tanks. Fish stress responses were measured through: behavioral observations, cortisol levels at 5 different times following the AFFA addition (0 min, 35 min, 2 hrs., 4 hrs. and 48 hrs.) and time to reach a 30% oxygen depletion after transfer. Finally, feed resumption was measured from the transfer on each batch. Data were analyzed using ANOVA (General Linear Model) when appropriate.

Regardless of the essential oil bioactives concentration, salmon's body orientation wasn't affected through the study and was similar to control fish. However, at 4 hours, fish in treatments C and D were excited while a decrease of swimming activity was observed in treatments B and E, compared to the control one. The oxygen depletion time was significantly the longest for fish in treatment E and the lowest for control fish (17.4 vs. 11 min; $p=0.054$). One day after the transfer, treated fish from treatment E displayed a quicker resumption of feed intake compared to the control fish (2.05 g vs. 0.69 g). The cortisol level observed in each group was increased from 35 min ($P<0.001$), which indicates a stress response to the experimental transfer model. It became significantly lower ($P<0.05$) in treated groups 4 hours post-treatment, and once again Group E stood out displaying the lowest cortisol level at this time compared to the control fish (204 vs. 547 nmol/l; $P<0.01$).

These results suggest the potential of using an AFFA for fish sedation at 20-ppm concentration repeated every 2 hours.

***Aeromonas salmonicida* CAUSES LETHAL INFECTION TO LUMPFISH (*Cyclopterus lumpus*) BY IMMUNE SUPPRESSION AND CELL CYCLE ARREST**

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Lumpfish (*Cyclopterus lumpus*), a native fish of the North Atlantic Ocean, has been utilized as cleaner fish to biocontrol sea-lice (*Lepeophtheirus salmonis*) infestations in Atlantic salmon (*Salmo salar*) farms in the North Atlantic region. *Aeromonas salmonicida* subspecies *salmonicida* is a Gram-negative facultative intracellular pathogen that causes lethal furunculosis in several fish species, including lumpfish. The molecular immune response of lumpfish to *A. salmonicida* infection is unknown. In this study, we established an *A. salmonicida* systemic infection model in lumpfish, and examined the transcriptome profile of central and peripheral lymphoid tissues after infection. Groups of lumpfish (50±7 g; n=60 per group) were intraperitoneally (ip) injected with different doses of *A. salmonicida* to calculate the median lethal dose (LD₅₀). Samples of blood, head kidney, spleen, and liver were collected at different time points to determine the infection kinetics. We determined that *A. salmonicida* lethal dose 50 (LD₅₀) is 10² bacterial cells per dose. The infection kinetic analysis indicated that the head-kidney is the primary target organ for *A. salmonicida* infection. Triplicate biological samples were collected from infected head kidney, spleen, and liver at 3- and 10-days post infection (dpi) and compared to non-infected organs. Transcriptome sequencing was performed in Nova-Seq illumina platform, generating total 1,034 million pair-end reads. Gene enrichment analyses showed that *A. salmonicida* induces lethal infection in lumpfish by uncontrolled and detrimental blood coagulation, complement activation, and inflammation. Such responses lead to hypoxia, internal organs hemorrhages, suppression of adaptive immune system, and impairment of DNA repair system, which results in cell cycle arrest, and death. These results concordance with the fish behavior before disease onset and mortality and coincident with *A. salmonicida* described virulence factors. This study has provided a valuable resource and novel insight into the interaction of fish-pathogens.

INACTIVATED *Aeromonas salmonicida* SUPPRESS ADAPTIVE IMMUNITY IN LUMPFISH (*Cyclopterus lumpus*)

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In the North Atlantic region, Lumpfish (*Cyclopterus lumpus*) have been utilized as cleaner fish to biocontrol sea-lice (*Lepeophtheirus salmonis*) infestations in Atlantic salmon (*Salmo salar*) farms. Previous studies showed that naïve lumpfish are highly susceptible to *Aeromonas salmonicida* infection and commercial vaccines are ineffective against *A. salmonicida* J233 hyper-virulent strain. Here, we evaluate the immune protective effect of *A. salmonicida* outer membrane proteins (OMPs) and bacterins in lumpfish with and without the Vap A layer (A+/A-). Triplicated groups of pit-tagged lumpfish (11 ± 2 g; $n=40$ per group) were intraperitoneally (ip) injected with outer membrane proteins from A+ and A- strains of *A. salmonicida* grown under iron-rich (OMPs) and iron-limited condition (IROMPs), and formalin-killed A+ and A- *A. salmonicida* bacterins. CARBIGEN™, a terminally-sterilized, carbomer-based (Carbopol 934P) adjuvant was used with the vaccine formulation. Blood samples were collected every two-week post-injection for the enzyme-linked immune sorbent assay (ELISA), and spleen samples were collected at six-week post-injection for a qPCR assay. Immunized fish were ip challenged with *A. salmonicida* J223 with 10^4 cells per dose at eight weeks post-immunization. All immunized fish had a similar mortality tendency to the non-immunized control fish. Most of the control and vaccinated fish died within two weeks post-challenge. Therefore, our results demonstrate that OMPs, IROMPs, and formalin killed bacterial cells of A+/A- *A. salmonicida* does not confer immune protection to lumpfish against *A. salmonicida* J223. These findings lead to the hypothesis that the inactivated *A. salmonicida* J223 does not trigger memory immune responses and confer protection to lumpfish. To further investigate, ELISA and qPCR analyses were conducted. Interestingly, ELISA results demonstrate that formalin killed *A. salmonicida* J223 bacterins either in presence or absence of Vap A layer does not increase IgM titers. Instead, it downregulates genes encoding IgM, MHC-II, and CD4, which indicated immune suppression. Therefore, our results support the hypothesis and give us a demonstration of the ineffectivity of many commercial and in-house vaccine against *A. salmonicida* J223. This study has provided a valuable resource and novel insight into the immune suppression effect caused by the *A. salmonicida* J223 strain.

IMPROVING LUMPFISH (*Cyclopterus lumpus*) GROWTH AND ROBUSTNESS: EFFECTS OF REDUCED NUTRIENT DENSITY AND FUNCTIONAL ADDITIVES

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Sea lice remains one of the largest challenges of the salmon aquaculture industry, and there is an increasing demand for de-licing strategies that do not negatively affect the health of the fish. The use of cleaner fish as a method of controlling infection has become well established in the European salmon aquaculture industry in recent years, and farmers indicate good efficiency, reducing the need for medical or mechanical treatments.

Lumpfish (*Cyclopterus lumpus*) are well adapted to colder temperatures and are less complicated to farm than other lice eaters such as Ballan wrasse (*Labrus bergylta*). Nutrient-dense feed is a factor believed to influence rapid growth, development and health of lumpfish. However there is a request from the market to reduce the growth rate to comply with vaccination guidelines from manufacturers (i.e. 500 degree days from vaccination to sea transfer) and still have efficient lice eaters in the cages.

In this RFF Nord-funded project in collaboration with Nofima (#267738) a feeding trial was carried out at Tromsø Aquaculture Research Station, with fish delivered from Center of Marine Aquaculture. Triplicate groups of lumpfish juveniles (average initial weight 2 g) were fed 3 diets with 2 different gross energy and protein levels for 12 weeks. A functional additive pack including krill, immunostimulatory components, prebiotics, and increased levels of vitamins was present in all diets, however the third diet contained a slightly modified additive pack.

Survival was high throughout the growth period, with average survival above 93% for all diets and an average final weight of 57g. No difference in feed intake or growth performance was observed between the 3 tested diets. Cataracts presence was evaluated after 12 weeks of feeding with clear differences between diets. Liver pigmentation was also enhanced by the diets. At the end of the feeding trial, randomly selected fish from the two best performing diets were labelled and transferred to duplicate challenge tanks for atypical *Aeromonas salmonicida* bath challenge. After 41 days, survival was improved with 30% between diets. These results indicate that a well-balanced reduced nutrient diet has a positive impact on various health parameters, however an impact on growth was not identified.

AQUACULTURE CERTIFICATION AS A MEANS TO MINIMISE SOCIAL AND ENVIRONMENTAL IMPACTS, WHILST HELPING TO PROVIDE TRANSPARENCY, TRACEABILITY AND SOCIAL LICENCE

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The rapid growth of global aquaculture production in recent decades led to concerns regarding the industry's social and environmental impacts, which independent third-party certification schemes such as the Aquaculture Stewardship Council were deemed effective tools to minimise. As the ASC celebrates in 2020 its 10th anniversary, this presentation will share highlights of its achievements to-date, and of a major survey of consumer's attitude towards seafood across seven countries in Europe, Asia, and North America (USA, Canada, Germany, France, The Netherlands, China and Japan). Some of the latest developments regarding traceability, product assurance (notably the Key Data Elements, and Trace Elements Fingerprinting projects) and of the ongoing Fish Welfare project will also be presented.

AQUACULTURE OPPORTUNITY AREAS: A NEW APPROACH TO U.S. AQUACULTURE DEVELOPMENT

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With a new vision and strategic plan to support aquaculture development, NOAA is leading a shift-change in U.S. aquaculture development through the identification of Aquaculture Opportunity Areas (AOAs). The search for AOAs in U.S. waters is a planning process, not a regulatory process, to identify defined geographic areas that are environmentally, socially, and economically appropriate for commercial aquaculture. By pursuing AOAs, we increase the confidence of both entrepreneurs and regulators via proactive planning measures that inform the permitting and environmental review processes for projects proposed to be sited within an AOA.

The process to identify potential AOA siting options and complete a Programmatic Environmental Impact Statement (PEIS) for each AOA will rely on the best-available scientific information and involve extensive public engagement. AOAs are intended to minimize interactions of aquaculture development with other ocean uses and important environmental resources, maintaining NOAA's commitment to ocean stewardship. NOAA will provide updates on our progress towards identifying the first two geographic areas containing locations suitable for commercial aquaculture, drafting the PEIS for each of those two areas, and the opportunities for stakeholder input into the process.

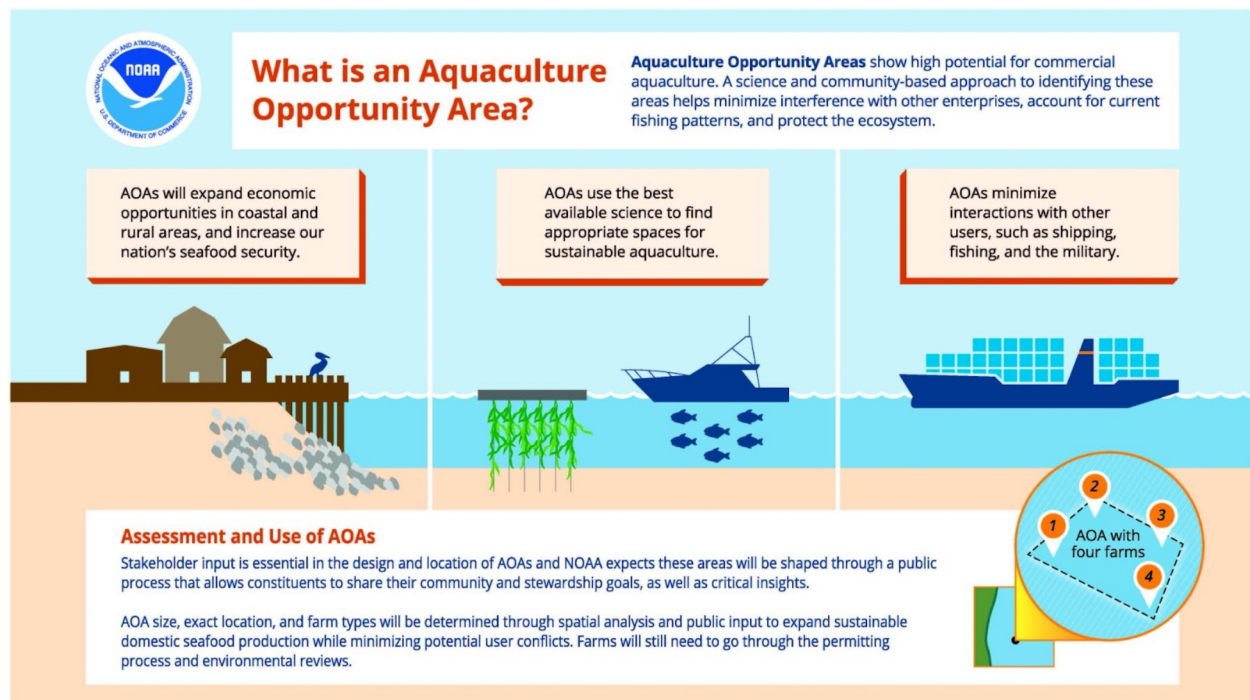


Figure 1. What is an Aquaculture Opportunity Area?

EFFECT OF OXIDIZED LIGNITE COAL AS A MICROBIAL CONTROL AGENT DURING THE INCUBATION OF RAINBOW TROUT *Oncorhynchus mykiss* EGGS

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Fungal infections primarily caused by water mold (*Saprolegnia* sp.) and bacterial infections caused by common aquatic pathogens result in annual losses in the hatchery production of rainbow trout and other freshwater fishes. The management of fungal and bacterial infections has historically relied on the use of chemical products which may have negative effects on human and environmental health. This study aimed to test the effect of AquaVate+ (MTS Environmental, Exeter, Ontario) which is derived from oxidized lignite coal, as an alternative treatment to reduce fungal and bacterial infections during the incubation of rainbow trout eggs.

Fertilized eggs were incubated in stacked incubator trays at 9°C for 42 days and AquaVate+ was added to the influent water in the treatment incubator at a rate of 0.005 mg/L. Every 10 days from fertilization to hatch, 1L water samples were collected from the trays for a total of 5 time points (0, 10, 20, 30 and 40 days post fertilization) for extraction, amplification and next generation sequencing of 16S rDNA. Fertilized eggs were enumerated at the eyed stage and at hatch to assess survival to the eyed egg stage and survival to hatch.

At 24 days post fertilization (dpf) (eye-up) survival among treated eggs was significantly higher than the control group ($P = < 0.001$). At 35 dpf, the control incubator showed evidence of fungus growth whereas no signs of fungus were observed in the treatment incubator. At 42 dpf (hatch), survival among eggs treated with AquaVate+ was significantly improved compared to the control incubators ($P = 0.001$).

The diversity of bacteria was significantly different between treatment groups ($P = 0.011$) and over time ($P < 0.001$). Specifically, alpha diversity decreased over time between samples collected on day 0 compared to days 20, 30 and 40 ($P < 0.01$). The composition of bacteria (beta diversity) was different between treatments ($P = < 0.001$), specifically the abundance of *Flavobacterium* and *Aeromonas* species, which are common bacterial pathogens.

The results of the current study have identified major findings that are relevant to the incubation of rainbow trout eggs in a hatchery environment. Specifically, the application of AquaVate+ at a rate of 0.005 mg/L improved survival, prevented the growth of fungus, reduced the abundance of pathogenic bacteria and significantly altered the microbial community of the water in which the eggs were incubated.

CO-INFECTION OF LUMPFISH WITH *Aeromonas salmonicida* AND *Vibrio anguillarum*

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Co-infection is a major challenge affecting aquaculture globally. Marine finfish have high tendencies to be affected by more than one pathogen concurrently in nature. Unfortunately, not enough attention is given to the studies of coinfection in aquaculture. Many commercial vaccines are developed to prevent single infection with little or no effort made towards the development of vaccines to combat co-infection. This can be because there is a lack of information on the infection patterns of different pathogens in several fish species. The susceptibility of fish to different pathogens or the severity of an infection caused by a pathogen can be altered during co-infections. There is need to understand how various pathogens affect fish in the presence of another pathogen. In this study, we investigated the co-infection of Lumpfish (*Cyclopterus lumpus*), a sea-lice biocleaner fish used in Atlantic salmon (*Salmo salar*) aquaculture with frequent marine bacterial pathogens *Aeromonas salmonicida* and *Vibrio anguillarum*. Three groups of fish were intraperitoneally injected with either PBS (control group) or sub-lethal doses of 10^1 and 10^4 or 10^2 and 10^5 CFU/ml of *A. salmonicida* and *V. anguillarum*, respectively. This experiment was conducted in duplicates and results showed regular clinical signs including petechial hemorrhage, skin lesions and exophthalmos. Survival rate ranged from 0 to 30%. Bacterial colonization revealed that sub-lethal doses of *A. salmonicida* could not be recovered on Tryptic Soy Agar (TSA) plates supplemented with the appropriate antibiotic marker because of the low doses. This was further elucidated in an *in-vitro* sea water analysis using 10^5 and 10^5 CFU/ml *A. salmonicida* and *V. anguillarum* respectively. The co-infection of lumpfish with *A. salmonicida* and *V. anguillarum* may be responsible for the increased mortality and severe clinical sign observed in this experiment even at sub-lethal doses suggesting a synergistic interaction between these bacteria.

SCREENING OF SOME FREQUENT PATHOGENS OF MARINE FINFISH AQUACULTURE FOR COMMON ANTIGENS AND EPITOPES TO BE USED IN UNIVERSAL VACCINE DEVELOPMENT

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Marine finfish aquaculture is affected by diverse infectious diseases, and they commonly occur as co-infection. Some of the most frequent and prevalent Gram-negative bacterial pathogens of the finfish aquaculture include *Piscirickettsia salmonis*, *Aeromonas salmonicida*, *Yersinia ruckeri*, *Vibrio anguillarum*, and *Moritella viscosa*. To prevent co-infections in aquaculture, polyvalent or universal vaccines would be ideal. Commercial polyvalent vaccines against some of these pathogens are based on whole inactivated microbes and their efficacy is controversial. Identification of common antigens can contribute to the development of effective universal or polyvalent vaccines. In this study, we identified common and unique antigens of *P. salmonis*, *A. salmonicida*, *Y. ruckeri*, *V. anguillarum*, and *M. viscosa* based on a reverse vaccinology pipeline. We screened the proteome of several strains using complete available genomes and identified a total of 154 potential antigens, 74 of these identified antigens corresponded to secreted proteins, and 80 corresponded to exposed outer membrane proteins (OMPs). Further analysis revealed the outer membrane antigens TonB-dependent siderophore receptor, OMP assembly factor BamA, the LPS assembly protein LptD and secreted antigens flagellar hook assembly protein FlgD and flagellar basal body rod protein FlgG are present in all pathogens used in this study. Sequence and structural alignment of these antigens showed relatively low percentage sequence identity, but good structural homology. Common domains harbouring several B-cells and T-cell epitopes binding to major histocompatibility (MHC) class I and II were identified. Selected peptides were evaluated for docking with Atlantic salmon (*Salmo salar*) and Lumpfish MHC class II. Interaction of common peptide-MHC class II showed good *in-silico* binding affinities and dissociation constants between -10.3 to -6.5 kcal mol⁻¹ and 5.10E^{-9} to 9.4E^{-6} M. This study provided the first list of antigens that can be used for the development of polyvalent or universal vaccines against these Gram-negative bacterial pathogens affecting finfish aquaculture.

BEYOND NANOBUBBLES: RAPID ADVANCES IN AQUACULTURE OXYGENATION TECHNOLOGIES

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The benefits of oxygenation for marine aquaculture are well known, but the technologies available for large-scale oxygenation of today's industrial-scale farming operations have – until recently – been poorly suited to the challenge. Poor efficiency, energy intensive, unreliable and difficult to integrate in to modern operations, most of the technologies available to marine farmers have been unable to deliver attractive returns on investment, resulting in lost growth, higher mortalities and lower revenues for farmers. In this presentation, Matt Clarke, P.Eng., CEO and Co-Founder of Poseidon Ocean Systems will detail significant new developments in marine oxygenation technologies for aquaculture, and provide a breakdown of the key metrics operators can use to evaluate which oxygenation technologies meet their requirements and will deliver the greatest return for their investment.

Matt Clarke, P. Eng. is the CEO and Co-founder of Poseidon Ocean Systems Ltd. He has a degree in Integrated Engineering from the University of British Columbia, and has worked to develop numerous new technologies for the marine and aquaculture industries. Recently Poseidon Ocean Systems was awarded one of Innovate BC's Aquaculture Innovation Awards for their Flowpressor technology, developed specifically for the aquaculture industry.

COMPENSATORY RESPONSE OF THE SOMATOTROPIC AXIS FROM IGFBP-2B GENE EDITING IN RAINBOW TROUT *Oncorhynchus mykiss*

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Insulin-like growth factor (IGF) is recognized as a central regulator of anabolic growth in vertebrates, largely due to its positive effects on muscle and bone growth. However, the majority of IGF-I is not free in circulation; approximately 99% of IGF-I is bound to IGF binding proteins (IGFBP). In addition to extending the half-life of IGF-I in serum, IGFBPs sequester IGF-I to peripheral tissues and can promote or inhibit ligand binding to surface receptors. Characterizing the functions of the IGFBPs is essential to understand the role of these proteins as regulators of IGF signaling. We used gene editing technology to edit IGFBP-2b, the most abundant IGFBP in serum that binds an estimated 80% of free IGF-I. The objective of this study is to determine how components of the IGF/IGFBP system respond to a reduction in serum IGFBP-2b abundance.

Rainbow trout with gene editing-induced reductions in serum IGFBP-2b (Mutant) exhibit similar growth performance compared to fish without IGFBP-2b gene disruption (Control). Editing the IGFBP-2b genes resulted in an 83% decrease in serum IGFBP-2b in mutants. This resulted in a 35% reduction in serum IGF-I (Figure 1c), which was offset by reduced expression of hepatic *igfbp-1a2* and increased muscle *igfr-1a* (Figure 1a,b); these responses suggest that an increased IGF-I signaling capacity offset reductions in serum IGF-I. During feed deprivation, the differential expression of *igfbp* genes supports the attenuation of the growth inhibitory response, likely due to the further reduction in serum IGF-I that alleviated the need for an IGF-inhibitory response. Unique *igfbp* expression patterns occurred during refeeding, suggesting an enhanced IGF-I signaling capacity in controls. Collectively, these findings support that the role of IGFBP-2b is to regulate serum IGF-I concentrations. The compensatory regulation of IGF/IGFBP system genes indicates that adjustments in other IGFBP, both circulating and at the local level, maintain IGF-I signaling at a level appropriate for the nutritional state of the fish.

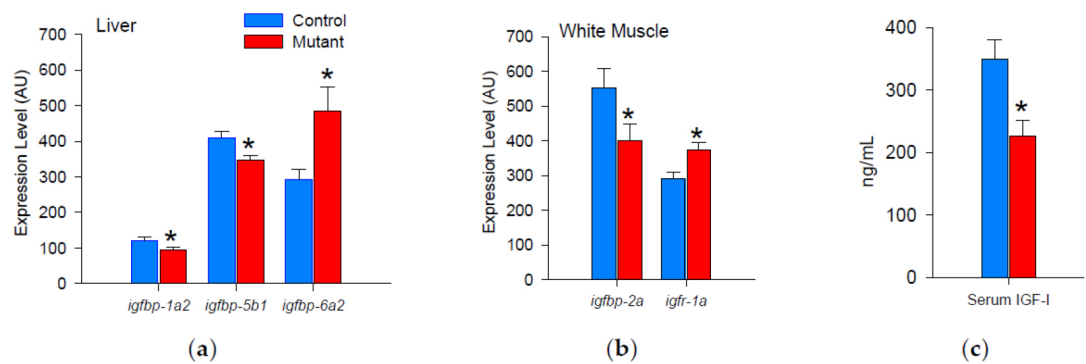


Figure 1. Differential regulation of somatotrophic axis components in a) liver, b) white muscle, and c) serum. Asterisks indicate significantly different means, $p < 0.05$.

IMPACT OF THERMAL DELOUSING ON ATLANTIC SALMON, *Salmo salar*, GILL HEALTH

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The overall cost of salmon louse (*Lepeophtheirus salmonis*) infestations on the Atlantic salmon (*Salmo salar*) industry has been estimated to be around £700 million annually. This high cost has driven a concerted effort to develop, examine, understand, and implement a variety of methods for sea lice control. Husbandry interventions utilising high temperature or pressurised water have been highly successful in complementing traditional chemotherapeutants where efficiency has waned in recent years.

In this project, we sought to examine the impact of elevated water temperatures (thermal treatment) on gill health in two commercial sites with divergent historical husbandry and treatment interventions immediately prior to and 7 days after thermal treatment. Methods to characterise gill health and the response to thermal treatment included a detailed examination of the gill surface for microparasites using both histology and qPCR and targeted expression analysis of cellular markers of immune cells, most notably antigen presenting cells (MHC II), proinflammatory cytokines (IL-1 β and TNF- α) and inhibitory cytokines (TGF- β and IL-10). Additionally, we scanned the bacterial communities present on the gill surface using a non-invasive approach. Microbiome analysis was performed through the development of a novel quantification protocol that included a titration step prior to building 16S rRNA libraries. This improved method facilitated a detailed observation of the microbial communities present in a challenging tissue.

Data obtained from these trials showed a minimal impact on gill microparasite prevalence in response to the thermal treatment. Expression of immune markers showed a significant decrease across all sites (t-test, $p < 0.05$) after treatment. Intriguingly, marked differences on gill surface bacterial communities in response to treatment between the two sites were clearly observed. This could be due to the notable differences in the husbandry history and health status of the fish at the two sites prior the thermal treatment (Figure 1). Our data suggest that microbiome diversity is an informative indicator of fish gill health and could be used to define appropriate interventions when treating for sea lice.

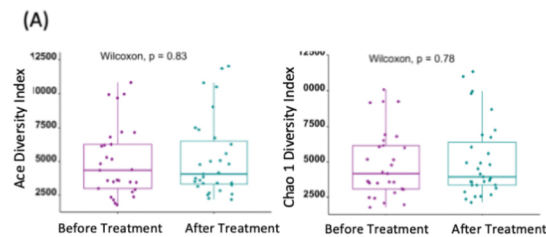
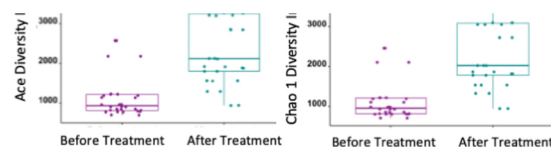


Figure 1. Comparison of thermal treatments on a previously untreated site (A) and a site with an extensive history of previous lice treatments. Data shows contradictory result based upon husbandry interventions.



THE PLACE FOR THERAPEUTIC BATH TREATMENTS IN THE MODERN INTEGRATED PEST MANAGEMENT TOOLBOX

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Sea lice are one of the greatest challenges the salmon aquaculture industry faces. The problem affects not only production, leading to substantial economic loss for the industry, but also – and arguably more importantly – the welfare of both farmed and wild fish. Highly efficacious, sustainable, and welfare-friendly medicinal bath treatments are a key tool to controlling sea lice as part of overall integrated pest management (IPM) plan, together with biological and non-medicinal treatments.

Benchmark® Animal Health has strong expertise and a depth of knowledge in providing solutions for sea lice management. Since Benchmark® Animal Health was established, we have supported farmers throughout the world to manage sea lice utilizing our recognized gentle and effective azamethiphos treatment, Salmosan® Vet.

Following over ten years of R&D, Benchmark® Animal Health, Norway received Marketing Authorization for Ectosan® Vet on 2nd July 2021. The first medicinal bath treatment to have been introduced to the Norwegian salmon market in over a decade. Ectosan® Vet used together with the water purification system, CleanTreat® – which removes medicines from treatment water before returning purified water into the sea – has now been used in Norway since August 2021.

This talk will discuss how to maximize the benefits of efficacious and welfare-focused treatments within the wider framework of an effective IPM strategy. All whilst having a focus on the importance of sustainability.

FRESHWATER, LANDLOCKED GRAND LAKE STRAIN OF ATLANTIC SALMON (*Salmo salar* L.) AS A POTENTIAL GENETIC SOURCE OF LONG CHAIN POLYUNSATURATED FATTY ACIDS SYNTHESIS

Stefanie M. Colombo*, Mohamed Emam, Brian C. Peterson, Jennifer R. Hall, Gary Burr, Zeyu Zhang and Matthew L. Rise

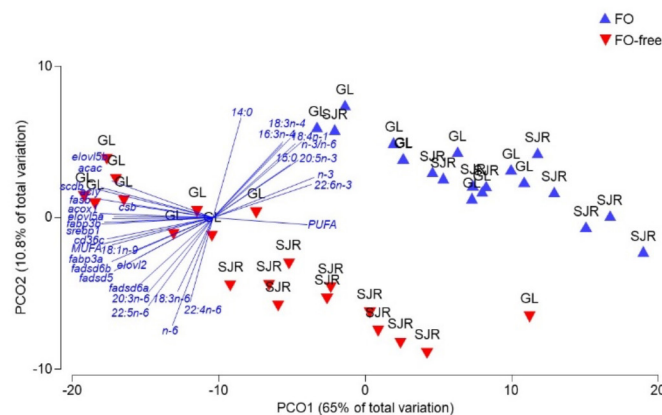
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Selection efforts focused on adaptation to plant-based diets, particularly the ability to synthesize polyunsaturated fatty acids (PUFA), are now emerging in aquaculture. Landlocked salmon (Grand Lake population; GL) may differ from the commercial Saint John River (SJR) strain in terms of PUFA metabolism. The objective of this study was to determine if landlocked GL strain have a better capacity to synthesize and store n-3 LC-PUFA, in the absence of dietary fish oil, in comparison to the SJR strain.

A 2x2 factorial trial was designed using two feeds containing fish oil (FO) or plant-based oil (FO-free) and was tested over 16 weeks in the SJR and GL strain (58 g/fish). Transcript levels of lipid metabolism-relevant genes, FA content of liver and muscle, and fish growth were determined.

GL salmon fed the FO diet showed reduced growth compared to SJR salmon (fed either diet); however, for GL salmon fed the FO-free diet, growth was not different compared to any group. In liver, SJR salmon fed the FO-free diet had higher levels of n-6 PUFAs (21.9%) compared to GL fed the same diet (15.9%); while GL salmon fed the FO-free diet had higher levels of monounsaturated FAs (48.9%) compared with SJR salmon fed the same diet (35.7%). 20:5n-3 and 22:6n-3 were the same in GL and SJR liver and muscle fed the FO-free diet. In liver, the GL strain fed the FO-free diet appeared to show higher expression levels of transcripts involved in *de novo* FA synthesis (*acac* and *acly*). The two strains also showed different strategies to balance n-3 and n-6 FA to control inflammation. However, synthesis and storage of EPA and DHA in the muscle tissue was not different between strains when they were fed the FO-free diet.

Our study clearly indicates that both the SJR strain and the GL strain responded differently to the FO-free diet compared to the FO diet. These results suggest there is a genetic basis behind the potential for GL salmon to utilize FO-free diets more efficiently than SJR salmon, with regards to FA metabolism.



DIFFERENCES IN LIPID AND FATTY ACID ACQUISITION AND STORAGE OF FARM, WILD, AND HYBRID ATLANTIC SALMON *Salmo salar* COMPETING IN THE WILD

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Farmed Atlantic salmon have experienced multiple generations of both intentional and unintentional domestication selection, generally reducing their fitness in the wild. When farmed salmon escape and interbreed with wild populations, these differences can lower wild population productivity and alter key life history traits. Understanding the genetic basis of fitness-related traits of farm, wild, and hybrid salmon is critical to predicting population impacts from farm escapees. To date, only a few European studies have considered all cross-types under common conditions in the wild, and none have examined lipid and fatty acid content, which are key influencers of fish health and performance. As such, this study used lipid and fatty acid analyses to investigate differences in energy acquisition and storage among experimentally derived Atlantic salmon fry from four cross-types (wild, farmed, and reciprocal F1 hybrids) during the first summer of growth in a southern Newfoundland river.

Wild-mother hybrids and pure wild fish had the highest recapture numbers, followed by pure farm fish, and then farm-mother hybrids. There was little effect of cross type on total lipid content at both release and recapture (Figure 1), however overall lipid and fatty acid profiles differed among cross types at both time points. Farm fish were the most differentiated cross type at recapture, with low levels of triacylglycerols (TAG) (Figure 2) and certain fatty acids indicative of freshwater prey suggesting a feeding disadvantage in the wild. Overall, the data indicate potential differences in foraging ability and/or energy storage among cross types, thereby improving our understanding of differential growth and survival.

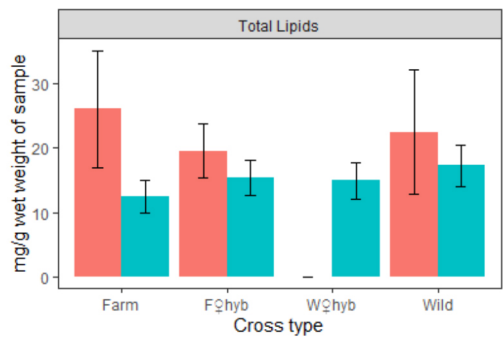


Figure 1. Mean total lipid content among cross types at release and recapture (note: W♀hyb samples not available at release). W♀hyb= wild mother hybrid; F♀hyb= farm mother hybrid

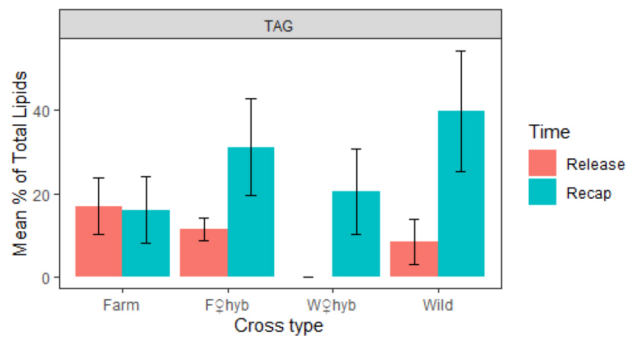


Figure 2. Mean TAG percentage among cross types at release and recapture (note: W♀hyb samples not available at release). W♀hyb= wild mother hybrid; F♀hyb= farm mother hybrid

SEMI-CLOSED CONTAINMENT SYSTEMS FOR FINFISH AQUACULTURE: ASSESSING MULTIPLE PARAMETERS FROM A COMMERCIAL SCALE OPERATON

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Semi-closed Containment Systems (SCCS), where an impermeable bag or structure separates farmed fish from the external marine environment, are gaining traction as a viable farming production system of salmonids. Early trials of enclosed floating aquaculture systems occurred from the early 1990s using a 450 m³ PVC bag. Since then, through development and optimisation phases, we now see this technology being upscaled to commercial level productions with systems utilising 30,000 m³ bags.

The main aim of adopting these systems is to limit or exclude challenges that impact fish welfare and the environment recognised in traditional open-net farming such as sea lice, predator interactions, fish escapes and waste capture. FiiZK is a producer of SCCS (Fig. 1) where we have developed a system that closes the production volume using a PVC bag and monitors the cage environment through a sensor network.

In this particular case, we provide welfare, sea lice, mortality and water quality data from a commercial scale production, where two-hundred thousand fish were stocked in six Certus 15000 SCCS in 2021, to highlight the potential these systems have for growing post-smolt from 100g to 2kg.

Welfare on 20 fish from each of the SCCS was assessed weekly by trained staff using morphological Operational Welfare Indicator's and results ranged from 0.01 to 0.87, where 0 is little or no evidence of this OWI and 1 is minor evidence of this OWI. Sea lice counts were carried out weekly and average scores showed that the number of lice ranged from zero lice to 0.13 and no sea lice treatments were carried out over the 7-month cycle. Accumulated mortality in the enclosures was between 0.6% and 3.9%. In one enclosure, elevated mortality was recorded because of wounds that occurred from handling and transferring fish from the open-net pens into the SCCS. This progressed into an acute secondary bacterial infection in the SCCS. Water quality remained within ranges that are acceptable for growing Atlantic Salmon and was highly correlated with seasonal variation in the fjord.



Fig 1. Schematic of FiiZK's SCCS

USE OF DESALINATION PLANT EFFLUENT AND SALTY GROUND WATER FOR CULTURE OF ARTEMIA

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Artemia, the brine shrimp, is an excellent live food for cultivable aquatic species. It is in great demand for use in shrimp and fish hatcheries. This tiny creature is a cosmopolitan taxon that has typically adapted to live in the stressful environmental conditions of higher saline habitats, such as salt lakes, lagoons and solar salt works all over the world. Many countries who do not have natural sources of *Artemia* have successfully set up artificial culture of *Artemia* using sea water. In this study we cultured *Artemia franciscana* and *Artemia urmiana* using salty effluent from a desalination plant and salty ground water respectively at 60 and 120 ppt salinities. The results showed that the survival of both *A. franciscana* and *A. urmiana* were significantly higher at 120 ppt compared to 60 ppt (Figure 1: A and B). However, the growth in both salinities did not show any significant differences (Figure 1: C and D). It was concluded that unconventional salty waters such as desalination plants effluent and ground salty waters may be comfortably used for artificial culture of *Artemia*. Both *Artemia* species prefer higher salinity for better performance. However, total lengths of adult *A. franciscana* and *A. urmiana* on day 23 were almost similar in both salinities which could be due to little number of survived animals in 60 ppt who had lesser food competition.

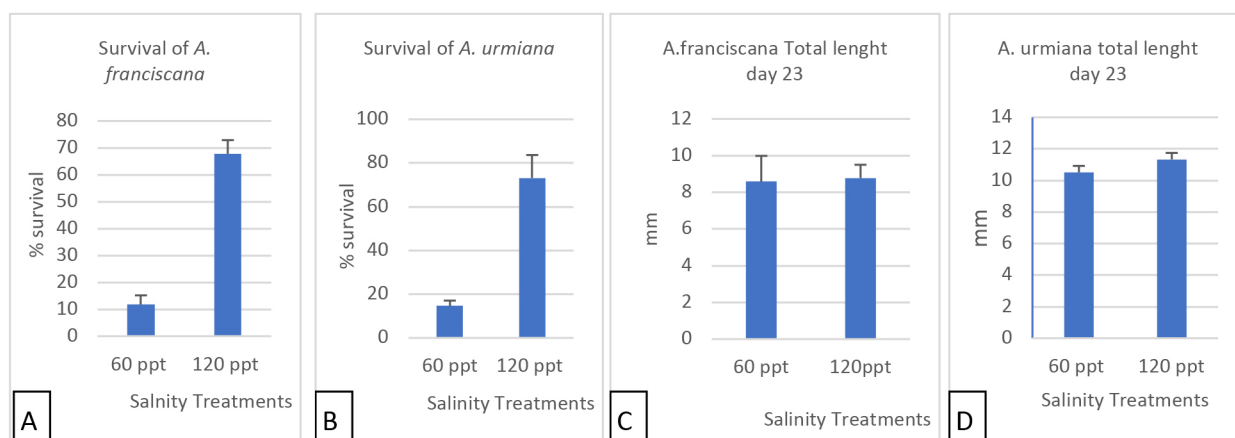


Figure 1. A: Survival of *A. franciscana*, B: Survival of *A. urmiana*, C: Total length of adult *A. franciscana*, D: Total length of adult *A. urmiana*

IMMUNE RESPONSE OF LUMPFISH (*Cyclopterus lumpus L.*) LARVAE TO ORAL IMMUNIZATION

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Vibrio anguillarum is a common pathogen that cause severe vibriosis disease in lumpfish (*Cyclopterus lumpus*). In this study, we developed a method to bio-encapsulate *V. anguillarum* bacterin in *Artemia salina* live-feed in order to orally immunize lumpfish larvae. We found that the highest amount of bacterin was detected at 3 hours *A. salina* post-inoculation. Bio-encapsulation was stable for 7 days at 4°C. We found that the bio-encapsulated bacterins were successfully delivered to the lumpfish larvae gut 6 hours after vaccination. Oral immunization of lumpfish larval using bio-encapsulated bacterins resulted in an increased gene immune response. However, the innate and adaptive gene expression was low, suggesting that vaccination of lumpfish larvae need booster immunization.

We orally immunized lumpfish larvae with bio-encapsulated *V. anguillarum* bacterin. One group was oral boosted after 3 months post primary immunization. A second group was oral and i.p boosted after 3 months and 4 months post primary immunization, respectively. A control group was mock immunized with the vaccine vehicle. The fish were challenged with *V. anguillarum* (10⁶ CFU/fish) after 6 months post primary immunization.

Oral boosted fish showed a mortality delay in contrast to the control group. The oral and i.p. boosted group showed a relative percent survival (RPS) of 75.5% ($p < 0.0001$).

In conclusion, immunization of lumpfish larvae is not recommendable since the immune system of the fish is not mature enough to mount a full protective immune response. Also, oral immunization seems to be not effective to protect the fish against *V. anguillarum* challenge. Finally, i.p. immunization trigger a protective immune response in lumpfish against *V. anguillarum*. Oral immunization perhaps required the utilization of live attenuated vaccines since inactive vaccines do not trigger a protective immune response in larvae or mature fish.

COMPARATIVE SUSCEPTIBILITY AND IMMUNE RESPONSE OF ATLANTIC SALMON (*Salmo salar* L.) FARMED, WILD AND HYBRID CROSSES TO *Aeromonas salmonicida* subsp. *salmonicida* INFECTION

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Aeromonas salmonicida subsp. *salmonicida* is the causative agent of furunculosis in several fish species including Atlantic salmon (*Salmo salar*). Although genetic susceptibility and resistance to *A. salmonicida* have been documented in salmon, the mechanisms are not well understood. Here, we compared naïve and immunized Atlantic salmon from farmed, wild and hybrid crosses to *A. salmonicida* susceptibility and immune response. Naïve fish were intraperitoneally injected with 2.5×10^4 CFU/100g and the immunized fish were challenged with 4×10^6 CFU/100g at 10 weeks post-primary immunization. Samples of tissues were taken at different time points. We found that naïve wild salmon were significantly more susceptible to *A. salmonicida* infection than the farmed and hybrid crosses ($p < 0.001$). Colonization of lymphoid tissues by *A. salmonicida* correlated with fish susceptibility. Expression of *il-1 β* , *il-10*, and *tlr5* were up-regulated after 10 days post-infection with higher expression in farmed fish in contrast to wild fish. Expression of *tnfa*, *igM* were up-regulated in farmed fish, but down-regulated in wild fish. In contrast to naïve fish, no differences in susceptibility were found between vaccinated fish challenged with *A. salmonicida*. This study provides new insights into Atlantic salmon immune response and susceptibility to *A. salmonicida* infection and immunization.

A WELFARE ANALYSIS OF NORWAY'S EXPORT PROMOTION PROGRAM FOR WHITEFISH

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Farm groups have a long history of supporting generic advertising and other activities designed to strengthen the demand for their products in domestic and foreign markets (Forker and Ward, 1993). A large empirical literature has developed on the costs and benefits of these programs (for examples see Piggott et al. 1995, Kaiser et al. (2005), and Kinnucan and Cai (2011); for a review, see Williams et al. (2018)). This study contributes to the literature of generic advertising in two ways. First, using Norway's export promotion program for whitefish as a case study we show that the benefit-cost ratio (BCR) from the producer perspective, defined as the increase in producer surplus associated with the advertising divided by the decrease in producer surplus associated with the levy used to finance the advertising, is invariant to the supply elasticity. This result is implied by Kinnucan and Myrland's (2000) analysis of the optimal levy and is demonstrated in empirical studies by Alston et al. (1998, 2005). Nonetheless, it has been largely ignored in the BCR literature. An important implication of the invariance property is that the BCR can be approximated using strictly demand-side information. If the goal is to obtain an estimate of the producer benefits of the advertising in relation to producer costs, estimates of the own-price and advertising elasticities of demand suffice; there is no need to estimate the supply side of the market. Second, we show that the BCR from the consumer perspective, defined as the increase in consumer surplus associated with the advertising divided by the decrease in consumer surplus associated with the levy, is identical to the BCR from the producer perspective, which is new. Our study considers Norwegian whitefish as a case study.

Norway recently increased the levy used to fund its export promotion program for whitefish from 0.50% to 0.75%. Study results suggest the intensified program is welfare increasing. The net social gain, defined as difference between the increase in economic surplus associated with the increased advertising and the decrease in economic surplus associated with the increased levy, is estimated at between \$81 million and \$174 million per year. The associated benefit-cost ratio (BCR), defined as the ratio of the aforementioned changes in economic surplus, is between 16 and 34 (Table 1). The BCR is invariant to the supply elasticity. The invariance property is useful as it implies the BCR can be estimated using strictly demand-side information; there is no need to estimate the supply side of the market. This result is generalizable to any advertising program funded by a levy on industry output or sales.

Table 1. Benefit-cost ratios associated with the expanded promotion program

Supply Elasticity	Foreign Consumer			Domestic Producer		
	ΔCS_A	$-\Delta CS_T$	BCR	ΔPS_A	$-\Delta PS_T$	BCR
----- Demand elasticity = - 0.416 -----						
0	0	0	undefined	179	5.33	33.5
0.5	98	2.90	33.8	81	2.41	33.6
1.0	127	3.75	33.8	52	1.56	33.6
1.5	140	4.15	33.8	39	1.15	33.6
2.0	149	4.39	33.8	31	0.91	33.6
----- Demand elasticity = - 0.863 -----						
0	0	0	Undefined	86	5.33	16.1
0.5	32	1.95	16.3	54	3.37	16.2
1.0	46	2.85	16.3	40	2.46	16.2
1.5	55	3.37	16.3	31	1.94	16.2
2.0	60	3.71	16.3	26	1.60	16.2

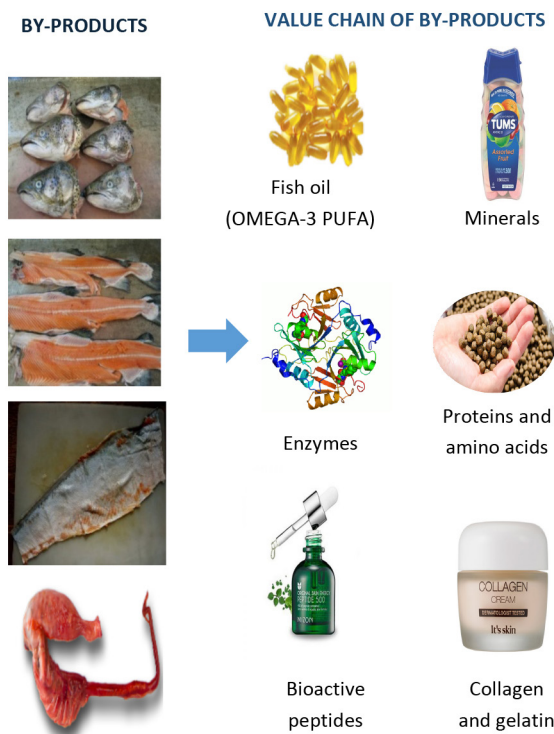
BEYOND PROCESSING: FULL UTILIZATION OF ATLANTIC SALMON BYPRODUCTS

Dr. Deepika Dave

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The Province of Newfoundland and Labrador is home to an aquaculture industry that is focused on the production of salmon and mussels. The province produced 10,620 tonnes of salmonids worth \$57.6 million in 2020. A large portion of the salmon (80%) harvested in Canada is further processed. Atlantic salmon processing removes the fillet portion of the salmon, but leaves the rest (head, skin, frame and trimmings: 45-50% of the body weight) as waste or for low value uses. Aquaculture firms dedicate a great deal of funds and resources to grow the entire salmon and when this non-fillet portion is not used effectively, this part of the investment is lost. Moreover, additional costs can be incurred in efforts to handle this potential resource and find a suitable means of disposal (transportation costs, tipping fees for dumping). In province, the issue of lost financial potential of unutilized salmon resources is worsened by the logistical and environmental issues posed by limited disposal options (composting, mink feed, landfilling). These salmon resources are a rich source of essential high value nutraceuticals and pharmaceuticals including collagen, gelatin, protein, amino acids, bioactive peptides, omega-3, oil and enzymes. The non-fillet portion of salmon is typically comprised of 15-19% proteins, 22-25% fat and 53-60% moisture in addition to minerals.

We investigated the efficient and economic pretreatment methods including grinding, tumbling and antioxidant addition for preservation of salmon by-products (head, frame and gut) followed by extracting salmon oil from different parts using enzymatic hydrolysis and total lipid content and the omega-3 fatty acid profile in the oil was analyzed. Besides fish oil, studies about collagen in marine byproducts have also been carried out. Proximate composition and hydroxyproline estimation of byproducts from Atlantic Cod, Atlantic salmon, and lumpfish have been performed. The content of moisture, ash, lipid, protein, and collagen were measured, and the results indicated that collagen is most abundant in salmon skin. Currently we are investigating the protein and amino acid compositions and extraction and isolation and purification of bioactive peptides from salmon byproducts.



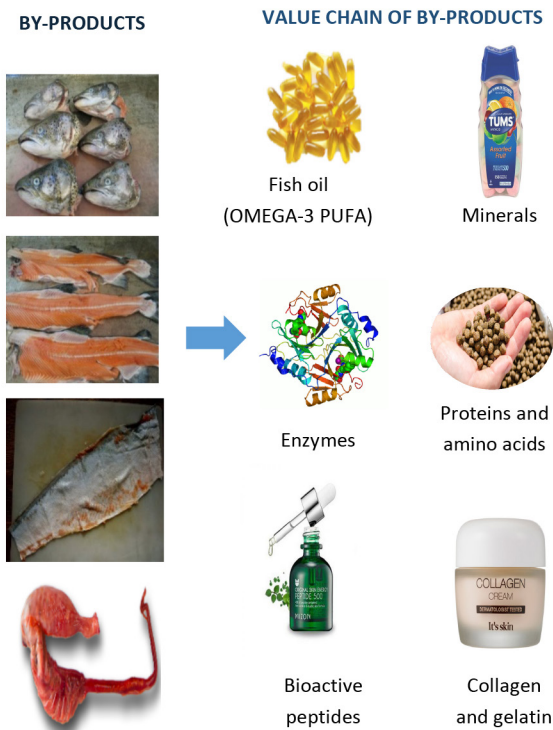
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SEAFOOD IN A TIKTOK ERA - MARKETING TO MILLENNIALS & GEN Z

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The seafood industry is changing. Today, millennials and Gen Z have the largest buying power in the market. This younger demographic is environmentally conscious, tech savvy, and convenience minded. They want information about their seafood that hasn't always been available to them and they want it on platforms where we haven't always made it available.

If we want to reach this important group of seafood customers, then we need to meet them where they're at - social media.

Today, TikTok and Instagram aren't just for pre-teens doing trendy dances. 47% of TikTok's 1 billion users are between 20 and 40 years old - which also happens to be the fastest growing demographic of seafood consumers. And more and more people are turning to these social media platforms for credible information.

There is a need for reliable information on these platforms. Social media content is widely unregulated, making it a breeding ground for fake news and misinformation. The only way to combat misinformation online is to fight back with credible information. If scientists and industry leaders in the seafood space don't flood timelines with the real story, the only narrative that will dominate online is one that isn't true.

The food industry is currently experiencing a once in a generation transition where consumers are actively shifting to their eating habits towards lower impact diets. Right now, for many, a lower impact diet means going vegan or plant based. Most consumers skip right over seafood because they don't even know it's an option. Because we haven't told them it is.

The prior conventional wisdom that consumers thought of fish and seafood as the "healthy choice" is not as prevalent among younger consumers. We need to change that, but this window of opportunity won't be open forever, so we need to act now.

If we can intercept these young consumers during their transition to lower impact eating, we can grow our sustainable blue food system, support ocean literacy, and transform diets for good.

In this session, I'll talk about how seafood marketing has to change to respond to the growing demand from young consumers and share effective strategies for communicating your seafood stories online.

AUTOMATIC ANALYSIS OF HARMFUL ORGANISMS IN WATER: A BIOLOGY AND MACHINE LEARNING POWERED APPROACH TO MONITORING WATER QUALITY

Dr. Jason Deglint*, Dr. Katie Thomas

Blue Lion Labs
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Under the impact of global climate change and human activities, harmful organisms such as sea lice and algae, are costing billions in damages and losses for the aquaculture industry worldwide. Aquaculture operators have to make decisions every day about the health and wellbeing of their stock, including dealing with environmental hazards outside of their control. When a harmful event occurs, farm operators have to decide how best to mitigate loss of fish stock. These mitigation strategies are costly, and the tools currently available to the industry don't provide the much needed information in a timely and cost-effective manner. In partnership with key players within the market (e.g. AcuaNativa, Universidad Austral de Chile, OTAQ), Blue Lion Labs is developing cost-effective solutions that will enable farm operators to continuously monitor organisms such as sea lice larvae and algae. We are building a solution that will replace manual, time consuming processes with automated equipment and machine learning software that will automatically process a water sample, analyze it and report the results to the farm operator in a consistent and reliable manner. With this proactive solution, operators will be able to trigger equipment such as netting or bubble curtains, or monitor the effectiveness of feed supplements in order to mitigate harmful events and prevent the loss of fish stock. All this combined, will lead to healthier and more resilient oceans around the world, as well as more sustainable aquaculture farming practices.

PATHOGENS AND DISEASE EXPERIENCES IN THE AQUACULTURE OF THE YUCATAN PENINSULA, MEXICO

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México's current aquaculture growth is 17%, an indicator above many other countries developing aquaculture. Main commodities are shrimp and tilapia, although marine finfish aquaculture has been developing since the 90's resulting in commercial aquaculture venues in the Pacific side. Some freshwater species like basa and trout are cultured in northern and center states while tilapia is cultured all over the country (32 states). In the Yucatan peninsula tilapia is the most important cultured organism, followed by shrimp and marine finfish in the Gulf of Mexico waters. Still, a common problem facing all aquaculture ventures are pathogens and disease which at some points have caused disastrous impacts; in 2013 a red drum farm off-the coast of Campeche lost 80% of their production due to an undiagnosed outbreak of nocardiosis (Figs. 1&2). During mid 2000's shrimp farms in Yucatan experienced the effects of ill-combined infections of Taura, white-spot disease and necrotising hepatopancreatitis which depleted the still incipient crustacean farming in the area. For over ten years there was no operative shrimp farm establishments in the whole peninsula.

Streptococcosis is detected with relative frequency y tilapia culture in the area, although no final identification has been issued by any government or dedicated lab. The id of the streptococcus strains is still to be determined. Another common finding are monogenoids parasites; at least 12 different species has been discerned, all of them of African origin. Two of them are very common, and although they seem not to be the cause of direct mortalities, the negative effects of these parasites on the condition factor and biomass loss in cultured tilapia in Yucatan has been recently issued by one of the authors (Figs. 3, 4, 5 & 6).

Mexico possesses laboratories with the standard and advanced tests for the detection of OIE-listed diseases. However, major losses in aquaculture is being caused by non-listed pathogens. Another major conceivable problem is the scarce amount of fish pathology professionals, which increase the vulnerability of aquaculture in the area. Although most curricula for veterinary studies include aquaculture teaching nowadays, pathology as a subject with sound microbiology knowledge needs to be reinforced. If the subject is not deal with proper attention, diseases will remain as the main deterrent of aquaculture sustainability.



Fig. 1. Ulcerative lesions in red drum *Sciaenopus ocellatus*

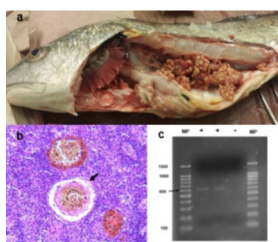


Fig. 2. Granulomatous necrotic and histological lesion in *S. ocellatus* (a and b); positive PCR for *Nocardia senoviae* (c).



Fig. 5. Skin haemorrhages in tilapia due streptococcosis

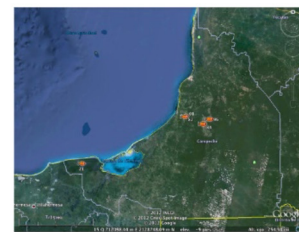


Fig. 6. Streptococcosis distribution in tilapia farms in Campeche.

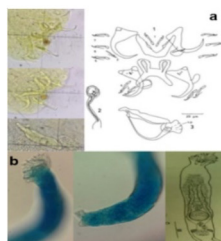


Fig. 3. Most common monogenean of tilapia in the Yucatan Peninsula. *Cichlidogyrus sclerosus* (a) and *Gyrodactylus* spp. (b).

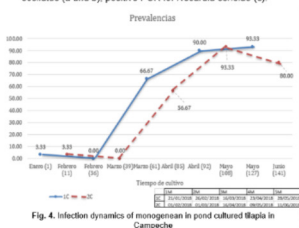


Fig. 4. Infection dynamics of monogenean in pond cultured tilapia in Campeche

ESTIMATING THE NUMBER OF LOCI REQUIRED IN A LOW-DENSITY PANEL FOR IMPUTATION OF GENOTYPES IN OYSTER GENOMIC SELECTION PROGRAMS

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Genomic selection utilizes high-density genotype information for both phenotyped individuals and selection candidates to improve genetic gains in a selective breeding program. To reduce genotyping costs, key individuals can be genotyped with a high-density panel while other individuals are genotyped with a low-density panel. Genotypes for the loci absent from the low-density panel can then be inferred through imputation so that the genomic estimated breeding values (GEBVs) can be calculated using high-density genotypes from all individuals. When designing such a program, one must know the number of loci required in the low-density panel to achieve sufficient imputation accuracy for GEBV calculations.

The number of loci in a low-density panel necessary for imputation depends on several factors including allele frequencies, distribution of loci throughout the genome, size of the genome, and the extent of linkage disequilibrium in the target population. Several of these factors are species specific. We therefore designed a series of simulations to investigate the number of loci that would be required for eastern oysters *Crassostrea virginica* and Pacific oysters *C. gigas*.

Our simulated breeding program consisted of 100 crosses per generation with 50 offspring per cross. All broodstock were genotyped with a high-density panel and all offspring were genotyped with a low-density panel. We simulated three generations and recorded both imputation accuracy and GEBV accuracy in each generation.

In the first simulation, founder genomes were generated based on demographic histories representative of wild oyster populations. Three additional simulations were run with high-density genotypes from either wild oysters or a Pacific oyster breeding program being used to define founder genotypes. These simulations demonstrated that imputation accuracy with a low-density panel of 250-500 loci was sufficient for GEBV accuracy to be comparable to that obtained with high-density genotypes for all individuals when genotypes for three generations (grandparents, parents, offspring) were available. This occurred beginning in the second generation of selection candidates. Panels of this size are achievable using currently available amplicon sequencing methodologies, making this approach immediately available for cost-effective, genomic selection in oysters.

INTERACTIONS AND BEHAVIORS OF LUMPFISH *Cyclopterus lumpus* AND STEELHEAD TROUT *Oncorhynchus mykiss* IN EXPERIMENTAL AQUACULTURE CAGES

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Using cleanerfish for sea lice biocontrol is becoming a common practice in salmonid aquaculture in the North Atlantic, however, there is still much to learn. At the University of New Hampshire, research focuses on understanding cleaning behaviors and cleanerfish-salmonid interactions in sea cages. Currently, lumpfish cleaning behaviors are being assessed with steelhead trout, a salmonid important to the future of New Hampshire's finfish aquaculture industry and one that hasn't been previously evaluated with cleanerfish. Lumpfish-trout interactions are recorded through a combination of underwater video, diver observations, and through monitoring the movements of fish implanted with transmitters (Vemco V7P tags) yielding 3-dimensional positions. Along with the collection of behavioral observations and movement tracking, sea lice infestations are monitored through routine sampling to determine the effectiveness of lumpfish treatments. This novel study goes beyond just mapping lumpfish movements, but provides greater insight into how the two species interact with each other which will illuminate best practices for utilizing cleanerfish in salmonid aquaculture.

AN ASSESSMENT OF SEA LICE IN COASTAL NEW HAMPSHIRE AND EXPLORING THE USE OF LUMPFISH *Cyclopterus lumpus* AS CLEANERFISH OF STEELHEAD TROUT *Oncorhynchus mykiss* IN EXPERIMENTAL AQUACULTURE CAGES

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Using cleanerfish for sea lice biocontrol is becoming a common practice in salmonid aquaculture in the North Atlantic, however, it has yet to be implemented in the US and there is still much to learn. At the University of New Hampshire (UNH), lumpfish (*Cyclopterus lumpus*) cleaning behaviors are being assessed with steelhead trout (*Oncorhynchus mykiss*), a salmonid important to the future of New Hampshire's finfish aquaculture industry.

In 2020 and 2021, we utilized an experimental steelhead trout aquaculture pen off the NH coast, operated by NH Sea Grant and UNH. Weekly from November to June, we assessed sea lice populations collected from a subsample of steelhead trout and documented lice species, life history stage, sex, and abundance. Lice loads (mean lice per fish) peaked in January 2020 at 3.60 lice per fish, and the dominant species observed was *Caligus elongatus* (99%; n=930) though some individuals of *Caligus curtus* were observed (n=9). Female lice and adult lice made up 74% and 87% of the lice population, respectively, throughout the assessment. The lice loads of gravid females peaked in February 2021 at 2.20 gravid lice per fish.

Using small microcosm cages stocked with steelhead trout, we also evaluated whether lumpfish presence and hide design (fake kelp, PVC panels) affected sea lice loads. Two 10-week trials were conducted from October to December 2020, with all fish sampled biweekly for lice. At the end of each trial, all lumpfish were euthanized, and gut contents analyzed. Hide design affected mean lice loads on trout in both trials, with lower lice loads in cages containing kelp hides (Trial 1: GLM, p=0.01; Trial 2: GLM, p=0.02). Lice loads were lower in cages containing lumpfish versus no cleanerfish (GLM, p=0.04), but only in one trial. There was no evidence of sea lice within any lumpfish stomachs. Water temperature and lumpfish size differed between the two trials suggesting that cleanerfish size, hide design, and water temperature are key variables for steelhead farmers to consider for effective sea lice control.

These foundational studies contribute towards developing best practices of lumpfish use for sea lice mitigation, leading towards the goal of increasing the sustainability and production of steelhead trout aquaculture in NH waters.

AQUACULTURE STEWARDSHIP COUNCIL (ASC) CERTIFICATION: EXPANDING THE SCOPE

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The Aquaculture Stewardship Council is the world's leading certification scheme for farmed seafood. The ASC uses market-based approaches to incentivise farmers to achieve strict standards regarding environmental and social performance. Currently, the ASC standards cover 17 species groups under 11 standards, plus a joint ASC-MSC standard for seaweed. Standards development to date have been multi-year dialogue processes involving many stakeholders from academia, producers, ENGOs, and others. While these processes have resulted in robust standards, they are lengthy and do not allow for a quick response to new or emerging species in the market. To respond to requests more quickly for new species standards, the ASC has outlined a niche species process.

Developing in concurrence with our aligned Farm Standard, the niche species project recognizes that many impacts of aquaculture are based on the production system, rather than only being species dependent. The Farm Standard will include a core set of indicators and will monitor ecosystem impact consistently across species and regions and set species-specific limits where necessary (e.g., mortality levels, feed conversion, etc.). Therefore, this is the perfect time to create a framework outlining necessary metrics and indicators required to more quickly add new species.

The niche species development itself begins with an analysis of market demand and emerging species in aquaculture production. Priority species are then entered into a risk matrix, which identifies areas of environmental or social concern. For example, species that are highly invasive, require wild caught broodstock, or which present concerns from a human welfare perspective will be flagged. While not necessarily meant to eliminate these species from consideration, the risk matrix will highlight areas which may require further investigation or consultation. A list of potential species for inclusion will then be reviewed by the ASC Supervisory Board, who provide the final decision on which species shall be developed.

The ASC has partnered with catfish and pikeperch producers in Europe to gather data and develop metric limits for these species. Trials will be conducted with these farms to test the process, after which a formal addition to the Standards can be released.

In combination, the Farm Standard and niche species projects will improve the ASC in several ways. First, the volume of aquaculture production able to be certified will increase with each additional species added. Second, a consistent set of standards across species will allow for us to better determine impact. Further, the development of improved reporting templates outlining metrics requirements across species will allow for trend analysis and robust impact reporting.

FORTUNE BAY SUMMER 2019 DIE-OFF: ‘OUTLIER’, ‘NEW NORM’ or ‘NORMAL VARIABILITY’?

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In September 2019, the largest die-off in the history of Newfoundland's finfish aquaculture industry occurred in Fortune Bay, a large embayment located on the south coast of the island. This die-off was quickly attributed to unusually high water temperatures that would have occurred over a 'prolonged period' and would, consequently, have reduced dissolved oxygen availability to the farmed fish. Using the results of recent oceanographic investigations, we will show how the physical environment of Fortune Bay varies from timescales of hours to decades and what we see as the most important dynamics which can affect farmed fish well-being and aquaculture activities more generally. We will investigate whether the summer of 2019 can be qualified as an 'outlier' or 'the new norm' or whether it was within the 'normal variability' of the system. We will conclude to offer avenues of other factors that may have played a role in this massive die-off.

TRACKING AQUACULTURE IMPACTS ON BENTHIC ENVIRONMENTS USING MICROBIOMES AND eDNA BIOMARKERS

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At salmon farms, the release of organic and inorganic wastes may alter benthic environments, and appropriate impact assessment tools are needed to help improve the sustainability of the industry. Our work explores novel approaches for robustly assessing aquaculture impacts at salmon aquaculture sites in Newfoundland through the use of next-generation eDNA sequencing technology and machine learning, which are compared and validated with classical approaches such as visual and chemical indicators.

Using marker-gene sequencing, we found that aquaculture operations rapidly alter existing benthic microbial communities, generating distinct shifts that can be used to identify and grade the level of impact and remediation during fallow cycles. Through cluster analysis, four bacterial community groups were identified and classified as representing low, intermediate, recent and high impact conditions. The bacterial groups correlated with the concentration of organic matter and of abiotic markers of aquaculture such as zinc and copper. Marker-gene data was also found to be suitable as input for machine-learning and can be used to build highly accurate models for predicting benthic impact, opening up avenues to automating impact assessment.

Additionally, using an inter-provincial study design that compared hard-bottom and soft-bottom aquaculture sites, we confirmed that microbial shifts are similar in nature across different geographical regions and substrate types, and that a select repertoire of microbial taxa are at the core of aquaculture induced shifts. Therefore, certain bacterial taxa could be used as biomarkers within future studies (Fig. 1) and could be targeted for the development of rapid testing solutions.

Our work gives proof-of-concept approaches based on eDNA assessment that could be integrated at the industry level to better understand and manage the lifecycle of waste at aquaculture sites.

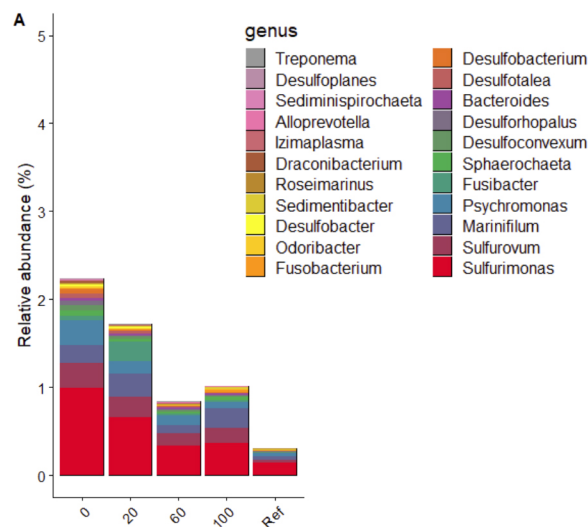


Fig. 1. Relative abundance of representative biomarkers at different distances from cages.

NUTRITIONAL EVALUATION OF AN AQUEOUS-PROCESSED OILSEED IN DIETS OF POST-SMOLT ATLANTIC SALMON *Salmo salar*

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The nutritional value of graded inclusion levels of canola protein concentrate (CPC; 75.8% crude protein, 4.4% crude lipid) obtained through aqueous processing was assessed in post-smolt Atlantic salmon over a three-month study. Eight experimental diets containing 0 (Diets A, E), 10% (Diets B, F), 15% (Diets C, G) and 20% (Diets D, H) CPC were randomly allocated to 24 750-liter tanks at 33 fish (228.0 ± 4.9 g) per tank. Diets A through D were formulated to mimic commercial salmon feeds in geographies where processed animal proteins (PAP) are used (Americas), whereas diets E through H included no PAP (Europe).

Overall, growth performances, measured using the thermal-unit growth coefficient (TGC), varied between 0.143 and 0.160, and were not significantly different among treatments ($P > 0.05$) (Table 1). Feed conversion ratios (FCR) were ≤ 1.08 and, although there was a significant CPC effect ($P = 0.003$), differences were marginal. Over the 168-day study, the best TGC and FCR were obtained with salmon fed 10% CPC, regardless of formula type. However, 20% CPC was optimal for growth and FCR during the first 28 days of this salmon study. Weight gain, TGC and FCR between days 0 and 28 improved in a linear manner ($P < 0.01$) in response to 10-20% CPC. Results on whole-body composition, nutrient deposition and distal intestine histology will be included in the presentation. The apparent digestibility coefficients were 91.1% for protein, 88.8% for lipid and 87.7-99.9% for essential amino acid in CPC. Based on these results, CPC stood as a safe and nutritious protein alternative up to 20% inclusion in salmon diet. The effect of higher inclusion level of CPC in nutrient-balanced diets deserve further studies.

Table 1: Initial body weight (IBW), final body weight (FBW), weight gain (WG), feed intake (FI), feed conversion (FCR), and thermal growth-coefficient (TGC) of Atlantic salmon fed experimental diets containing graded levels of canola protein concentrate (CPC) using either American (A) or European (E) formula. Data are means \pm SEM. Means within a column with no superscript in common differ significantly ($P < 0.05$) based on two-way ANOVA followed by Tukey test.

Treatment		IBW (g fish ⁻¹)	FBW (g fish ⁻¹)	WG (g fish ⁻¹)	FI (g fish ⁻¹)	FCR	TGC [g ^{1/3} (°C d ⁻¹)]	Survival (%)
Formula	CPC							
Day 0-168								
Americas	0	228.2 (3.2)	972.4 (29.6)	744.2 (26.5)	779.8 (26.0)	1.05 (0.00) ^{ab}	0.160 (0.004)	93.9 (0.0)
Americas	10	223.6 (2.0)	961.8 (48.5)	738.1 (46.6)	757.2 (42.3)	1.03 (0.01) ^b	0.160 (0.006)	100.0 (0.0)
Americas	15	228.3 (1.6)	916.7 (44.6)	688.4 (43.0)	744.0 (46.0)	1.08 (0.01) ^a	0.152 (0.006)	100.0 (0.0)
Americas	20	228.5 (0.9)	920.7 (30.2)	692.2 (31.1)	744.6 (24.6)	1.08 (0.02) ^a	0.152 (0.004)	100.0 (0.0)
Europe	0	230.1 (6.6)	863.8 (44.9)	633.7 (38.5)	668.1 (46.5)	1.05 (0.00) ^{ab}	0.143 (0.005)	89.9 (6.1)
Europe	10	227.5 (2.7)	946.5 (16.0)	718.9 (13.9)	753.1 (15.5)	1.05 (0.01) ^{ab}	0.156 (0.002)	99.0 (1.0)
Europe	15	230.3 (2.0)	913.6 (19.0)	683.3 (17.6)	734.4 (14.4)	1.08 (0.01) ^a	0.151 (0.002)	100.0 (0.0)
Europe	20	227.7 (1.6)	908.6 (16.8)	680.9 (16.3)	728.6 (12.2)	1.07 (0.01) ^{ab}	0.151 (0.002)	100.0 (0.0)
Two-way ANOVA (P-value)								
Formula		0.4357	0.1630	0.1205	0.1321	0.7335	0.0604	-
CPC		0.6104	0.5950	0.4777	0.8028	0.0033	0.1769	-
Formula x CPC		0.8912	0.3839	0.3260	0.3035	0.4297	0.0769	-

UPAQUACULTURE MOVING FORWARD, SUSTAINABILITY AND MODERN TECHNOLOGY HAND IN HAND

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The Placentia Bay Atlantic Salmon Aquaculture Project has two primary components: a land-based Recirculating Aquaculture System (RAS) Hatchery located in the Marystown and sea cage sites located in the northern portion of Placentia Bay that will be used to grow salmon to market size. At the RAS Hatchery, smolt will be grown to sizes ranging from 350–1,400 g and then transferred to a well boat and delivered directly to sea cage sites.

Technology: Grieg NL will use state-of-the-art technology in both its RAS Hatchery and sea cage sites. Grieg NL will use the adoption/adaption of new and innovative proven technologies including; vaccination machine, sea cages, RAS system, hybrid barges, remote monitoring in both aspects of operations. Grieg NL is also committed to acquiring accreditation and implementing Best Aquaculture Practices (BAP) through an international and proven third-party aquaculture certification program.

All Female Sterile Atlantic Salmon: After careful consideration of current research and development Grieg NL concluded that the use of sterile triploid all-female Atlantic salmon was the ideal choice for its proposed operations in Placentia Bay. The main factors for moving into full scale production of sterile triploid all-female Atlantic salmon in Placentia Bay for Grieg NL include; performance, environmental conditions, availability, and feed. All female sterile Atlantic salmon are means to sustainably provide healthy and nutritious seafood while minimizing environmental impacts, supporting economic growth and contributing to the development of local communities.

Water Use and Large Smolt: RAS Hatchery can be supplied by available groundwater sources, filtered to eliminate any entry of diseases or parasites, heated/chilled if needed, and only uses 300 L of water per minute. Also, smolt will be reared well above the industry standard before entering the sea. This increased size greatly reduces the production time in the sea. The approach reduces the time required for fish to reach a harvestable size, while reducing stress and exposure to pathogens such as disease or sea lice. A shorter production time at sea can also decrease the organic deposits that are associated with sea-based culture of fish.

Environmental Protection : Grieg NL proposes to use a number of mitigation measures that go beyond the common industry standard. These mitigations include approaches such as the utilization of sterile triploid all-female Atlantic salmon to minimize effects on wild salmon, the use of lumpfish to control sea lice, and following protocols that exceed government requirements.

CANOLA PROTEIN CONCENTRATE: CREATING A NOVEL, SCALABLE PROTEIN INGREDIENT FOR GLOBAL AQUACULTURE

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Canola (oilseed rape) is the second largest oilseed crop grown globally, with primary centers of production in Canada, northern Europe, and Australia. Canadian canola contains 4.5M mt of protein (with a similar amount in northern Europe) that is under-utilized and available for upgrading into high quality and cost-effective aquaculture feed protein.

Botaneco Inc. has developed a novel oilseed processing strategy to produce high quality protein concentrates for aquaculture feed. Canola has a well-balanced amino-acid profile but hasn't been utilized in high-value aquaculture diets due to traditional processing methods and the presence of anti-nutritional factors. Recent innovations in plant-breeding and processing strategies will enable the safe use of canola protein at high inclusion rates in salmon and shrimp diets.

These innovations present an opportunity to advance a new sustainable, large-scale source of protein without the need for further deforestation, develop shorter supply routes with a reduced carbon footprint and create new regional value-chains.

CAPTURE OF YOUNG-OF-THE-YEAR LUMPFISH *Cyclopterus lumpus* FOR CAPTIVE BREEDING IN MAINE, USA

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Maine's salmonid industry is eager to deploy lumpfish *Cyclopterus lumpus* into salmon net pens for biological control of sea lice. However, health certification requirements make it challenging and costly to source lumpfish from outside Maine territorial waters. Recent research activity has focused on establishing a lumpfish breeding colony in Maine and identifying factors that optimize captive breeding. In 2018 the University of Maine Center for Cooperative Aquaculture Research (CCAR) initiated a broodstock development program whereby wild, young-of-the-year (YOY) juveniles are captured from the Gulf of Maine to be acclimated and reared to reproductive maturity in CCAR recirculating aquaculture systems (RAS).

Juvenile lumpfish associate with free-floating mats of seaweed (*Ascophyllum nodosum* and *Fucus spp.*), which can be netted onto a skiff and sorted to capture the YOY. Fishing occurs from July – September when surface seawater temperatures range from 12-18°C. Captured fish are typically 5-15mm length and <1g, but specimens as large as 15cm have been captured in this fashion. In 2018 158 YOY fish were captured; in 2019 429 YOY fish; and in 2020 716 YOY fish. Post-capture survival was poor the first year (~20%), but in following years was improved to about 50% by co-feeding the smallest of the captured juveniles live prey (*Artemia*) along with specialty formulated post-larval diets (Otohime). It's also important to size grade the fish, as larger fish are aggressive and will cannibalize smaller individuals. Once acclimated to captivity the fish grow rapidly, reaching reproductive maturity in their 2nd year. This approach has proven to be successful but ultimately needs to be transitioned to a self-sustaining captive breeding program to avoid further impacting the wild resource.



Figure 1 Capture vessel and crew (L) and newly captured juvenile (R)

EVALUATION OF SOME PHYSICOCHEMICAL PROPERTIES OF UREJE RESERVOIR, ADO-EKITI, EKITI STATE, NIGERIA

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This study was designed to investigate the environmental factors in terms of the physicochemical parameters influencing the water chemistry of Ureje reservoir, Ado Ekiti, Ekiti State, Nigeria. Monthly variations in the physicochemical parameters were studied for a period of six months (July – December, 2018) using standard procedures of APHA (1998). Results of the Physicochemical analysis showed mean values of pH, temperature, biological oxygen demand, dissolved oxygen, total solids, total dissolved solids, total suspended solids, alkalinity, zinc, iron, cadmium, phosphate, sulphate and nitrate to be 7.65 ± 0.16 , $21.85^\circ\text{C} \pm 0.44$, $2.18\text{mg/L} \pm 0.06$, $4.94\text{mg/L} \pm 0.12$, $26.06\text{mg/L} \pm 3.35$, $11.73\text{mg/L} \pm 1.367$, $14.83\text{mg/L} \pm 2.96$, $4.32\text{mg/L} \pm 1.77$, $0.0007\text{mg/L} \pm 0.01$, $0.007\text{mg/L} \pm 0.009$ and $0.0\text{mg/L} \pm 0.0$, $0.10\text{mg/L} \pm 0.11$, $19.61\text{mg/L} \pm 4.95$, $6.56\text{mg/L} \pm 2.33$ respectively. The results obtained in this study were found to be within the recommended value stipulated by World Health Organization (WHO) for drinking and agricultural purposes thus, the water can be classified as a good, stable and healthy aquatic ecosystem. It is however imperative to intensify efforts in monitoring the water quality of the reservoir to prevent degradation due to pollution from municipal and agricultural runoffs.

Fresh water is essential to human health, agriculture, natural ecosystem and industry. Fresh water bodies are also used for stocking fish, domestic use, drinking and also for irrigation. Rapid growth of population, increasing living standard in urban areas and industrialization have resulted in greater demands of quality water (Miller, 2002). As a result of this, reservoirs have been constructed in many areas to cater for the increasing water demands. Lakes and surface water reservoirs are the planet's most important freshwater resources and provide innumerable benefits. Reservoirs may be created in river valleys by the construction of dams on a large expanse of water or may be built by excavation in the ground or by conventional construction techniques such as brickwork or cast concrete. The utilization of reservoir for fish production besides other primary uses has become common practice in many countries, especially industrialized ones, where a number of reservoirs are managed for the production of annual harvestable fish crops.

The physico-chemical parameters are very important to get exact idea about the quality of water, protect the natural ecosystem and understand the distribution and productivity level of aquatic ecosystems (Patil *et al.*, 2012). Studies on water quality monitoring is very important in this part of the world because it provides data on determination of current conditions and long-term trends for effective management of this natural resource. The supply of safe water has a significant impact on the anticipation of water transmissible diseases. Therefore, it is necessary to frequently monitor water quality, used for drinking purposes.

THE THERMAL DEPENDENT BIOLOGY OF LUMPFISH: DIFFERENCES DUE TO TEMPERATURES DURING EARLY LIFE AND HOW TOLERANCE IS MEASURED

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The use of lumpfish (*Cyclopterus lumpus*) as a “cleaner fish” in Atlantic salmon aquaculture has been very successful in mitigating the effects of sea lice on cage-site production. However, during the summer, there have been reports of lumpfish mortalities at some cage-sites in Atlantic Canada. Thus, we measured the upper thermal limits and metabolic physiology of lumpfish, with the goal of better understanding their physiology. Specifically, whether alterations in incubation or rearing temperature might improve their tolerance of high temperatures, and thus, welfare.

The specific egg incubation / larval & juvenile rearing temperature combinations were 6°C/9°C (standard production temperatures), 8.5°C/9°C, 6-11°C/9°C, 8.5°C/9-11°C and 6-11°C/9-11°C; with a range of temperatures indicating that the fish were exposed to stochastic temperature changes. Hatching success was lower in the stochastic-incubated group (~21%) as compared to the groups incubated under constant temperature (e.g., 6°C, ~61%), and larval survival in the 6-11°C/9-11°C group for the first 275 degree days (~89%) was lower than for all other groups (range 92.5 – 95%). As shown in Figure 1A, at this point, a temperature anomaly occurred and rearing temperature became variable and ranged from ~ 10 to 15°C for approx. 7 weeks. The fish incubated using standard industry protocols (6°C incubation / 9°C rearing) had the best survival during this period (~70%), whereas those exposed to stochastic incubation and rearing temperatures fared the worst (survival at the end of this period only ~30%). When the groups reached ~50 g critical thermal maximum (CT_{Max} , at 2°C h⁻¹) and incremental temperature maximum (IT_{Max} , at 1°C per week) tests were performed on all groups. Not surprisingly, (given the temperature anomaly during rearing), there was no difference between groups for these metrics of thermal tolerance (mean values 22.9 ± 0.1 °C and 20.6 ± 0.02 °C, respectively) or in metabolic parameters measured during the CT_{Max} test (values for standard and maximum metabolic rate, and aerobic scope, 34.5 ± 1.6 , 280.1 ± 9.6 and 245.6 ± 9.3 mg O₂ kg h⁻¹, respectively). Finally, we measured plasma cortisol levels in the fish used in the IT_{Max} experiment at 10, 16, 18 and 20°C. Cortisol levels were 0.70 ± 0.18 ng mL⁻¹ at 10°C and 1.57 ± 0.34 ng mL⁻¹ at 16°C ($P > 0.05$), but ~2.0 ng mL⁻¹ at both 18 and 20°C ($P < 0.05$). These data suggest that lumpfish in sea-cages become stressed by 18°C. Further, they agree with the findings of other work in our lab. Specifically, while the CT_{Max} and IT_{Max} of lumpfish are approx. 3°C lower than for Atlantic salmon, temperature alone does not explain their losses at cage-sites during the summer months.

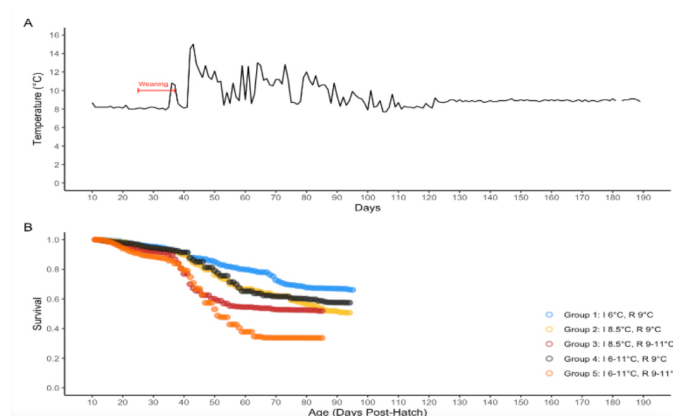


Figure 1. Temperature in the rearing tanks for the first 200 days, and the effects of different combinations of incubation and rearing temperature on survival. N = 2 tanks per group. *Note:* the lumpfish were graded at 900 dd, but remained in these tanks until moved into new tanks for the temperature tolerance tests.

STRIPED RED MULLET (*Mullus surmuletus*, L. 1758): STUDIES ON ITS AQUACULTURE POTENTIAL

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This study was undertaken with the aim of studying the reproduction in captivity and the rearing of the first larval stages of the Striped red mullet in order to attempt a first evaluation of its susceptibility, as a candidate species for aquaculture.

Striped red mullet is a demersal fish occurring along the Atlantic coasts of Europe and Africa. In Morocco, striped red mullet is distributed along the entire Moroccan coastline. This species is considered to be among the most valuable and highly priced fish species because its high nutritional proprieties. Muscle is the main lipid storage organ. The lipid content varies between 2.60% and 22.27% of the dry weight of the muscle. The reproductive season extended from April to June. Sexual maturity occurs at 2 years of age. The mean fecundity is 757699,44±530274 oocytes. The prey items identified are Crustacean (Amphipods and Decapods) Polychaeta and Molluscs. Crustacean constituted more important group (%IRI = 70, 8).

To study the reproduction in captivity of striped red mullet, a batch of broodstock were captured from the wild and kept in an indoor rectangular tank of 1m³ under natural conditions of temperature (14-19.3°C), dissolved oxygen (5.9-7.4 mg/l), salinity (34-36 ‰). The red mullet spawn naturally in captivity when the water temperature is between 9,5 and 16°C. The spawning can also obtained by hormonal induction using 20 µg/kg LHRHa. The viability rate was over 70%. The incubation realized at 14± 1°C. The hatching rate was 43.23 %. Larvae length at hatching was 2,89 ± 0,12 m, the yolk sac larvae had a total length of 1.15 ± 0.09 mm. With time, it decreased until disappeared completely around the fifth day. Larviculture was with green water. Rotifers were offered as first food 3 days after hatching. The larvae completed metamorphosis within a week after hatching.

The results of this study show that striped red mullet is an easy species for acclimatization to conditions of captivity, and it can even spawn during the same year of capture. Fecundity in captivity can reach 750,000 eggs per kilogram per year. After hatching the larvae are of good quality and present a good aptitude for rearing. These preliminary results are encouraging and merit further study to demonstrate the feasibility of producing striped red mullet fingerlings for stock enhancement or grow-out purposes.

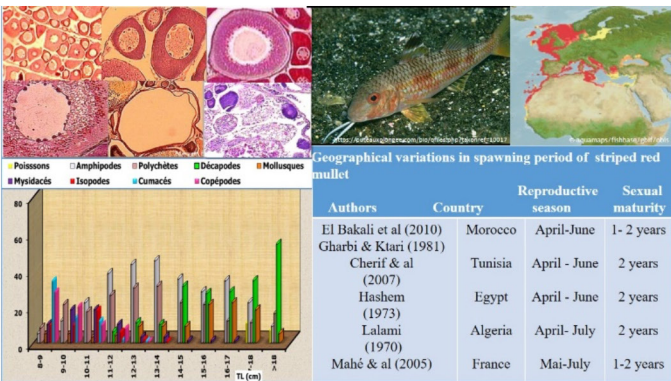


Figure 1. Biology of striped red mullet

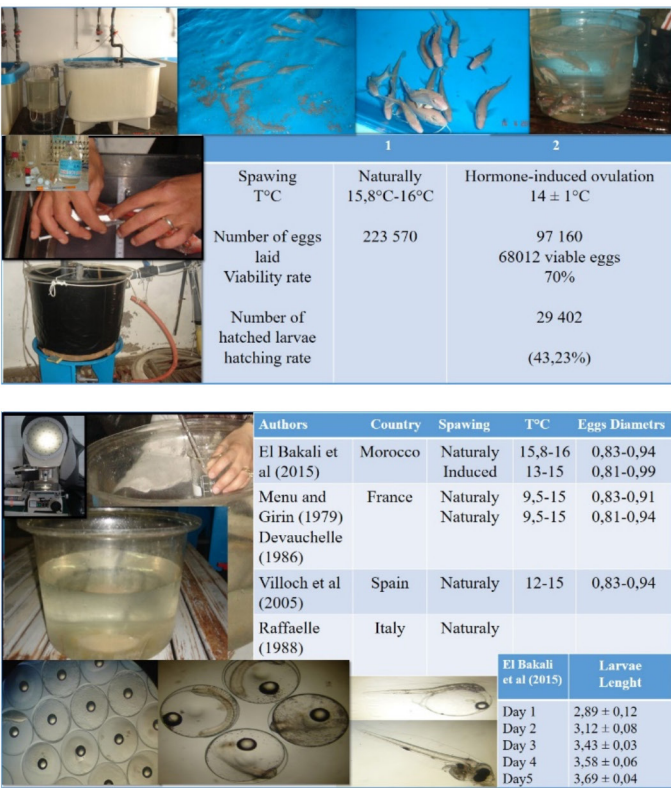


Figure 2a,b. Study of the reproduction in captivity of striped red mulle

AQUAPONICS SYSTEMS FOR REUSE WATER IN CONTINENTAL FISH FARMS : PRELIMINARY STUDY OF THE FEASIBILITY OF ITS INTRODUCTION IN MOROCCO

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The aquaculture sector continues to grow rapidly across the globe but its environmental impacts are growing as well. Considering the risks carried by classical aquaculture systems, it is necessary to develop responsible aquaculture. This is achievable by reducing the needs for water and nutrients as well as limiting waste and effluents. To this end, a shift to integrated agri aquaculture systems is a prerequisite.

Aquaponics has proven to be an effective means of recirculating aquaculture and horticulture with a decreased environmental impact. The advantages it presents earned it a growing interest worldwide as environmentally friendly expedient to achieving a larger resource productive growth.

In this context, a preliminary study of the feasibility of the introduction of aquaponic systems in continental fish farms was conducted. Thus an aquaponic system was installed. Effluents from the Silver carp (*Hypophthalmichthys molitrix*) tank are redirected to plants (lettuce, pepper, mint), instead of being discharged into the environment. Fish waste is transformed by bacteria into nutrients that can be absorbed by plants. Purified water is then returned to the fish tank. Every week, the height of each plant was measured in centimeters. Dissolved Oxygen, pH, water temperature, NO₃⁻, NO₂⁻ and NH₃ were also measured. The growth of fish was determined by measuring body weight and total length. Also, for each sampling, Feed Conversion Ratio (FCR) and Specific Growth Rate were calculated. Variation in the measurements was expressed as the mean \pm S.E.

The preliminary results are encouraging. Fish and plant growth rates were remarkable: no pathological abnormality was observed in fish. The mean growth of Silver carp had significantly improved during days of the experiment at ($\alpha \leq 0,05$). Comparing the average initial length ($15,12 \pm 1,50$ cm) and the average initial weight ($60,68 \pm 19,15$ g), to the final experiment results, the average final length was $18,25 \pm 1,89$ cm, and the average weight was $85,06 \pm 20,10$ g. Plants grew fast with no pesticide nor fertilizer input (The height was between 2,11 cm and 4,05 cm). Furthermore, the water quality was very good (water temperature (19-22°C), PH (6-7,3).

This study showed that the application of the aquaponic system has significant influence at nitrogen reduction (NO₂⁻ (12,6%), NH₃ (19,04%) and NO₃⁻ (15,1%). In light of these results, recommendations were issued aiming to nurture the development of aquaponics in Morocco in the future.

NUTRITIONAL IMMUNOMODULATION OF ATLANTIC SALMON RESPONSE TO *Renibacterium salmoninarum* BACTERIN

Mohamed Emam*, Khalil Eslamloo, Albert Caballero-Solares, Evandro Kleber Lorenz, Xi Xue, Navaneethaiyer Umasuthan, Hajarrooba Gnanagobal, Javier Santander, Richard G. Taylor, Rachel Balder, Christopher C. Parrish, and Matthew L. Rise

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We investigated the immunomodulatory effect of varying levels of dietary $\omega 6/\omega 3$ fatty acids (FA) on Atlantic salmon (*Salmo salar*) antibacterial response. Two groups were fed either high-18:3 $\omega 3$ or high-18:2 $\omega 6$ FA diets for 8-weeks, and a third group was fed for 4-weeks on the high-18:2 $\omega 6$ diet followed by 4-weeks on the high-18:3 $\omega 3$ diet and termed “Switched-diet”. Following the second 4-weeks of feeding (i.e., at 8-weeks) all groups were sampled for FA analysis. Then fish were intraperitoneally injected with either a formalin-killed *Renibacterium salmoninarum* bacterin (5×10^7 cells mL⁻¹) or phosphate-buffered saline (PBS control), and head kidney tissues were sampled at 24 h post-injection for gene expression analysis. The FA analysis showed that the head kidney profile reflected the dietary FA, especially for C₁₈ FAs. Also, this analysis suggested the role of FA ratios representing the balance between anti-inflammatory FAs and pro-inflammatory FAs (e.g., EPA/ARA in high-18:3 $\omega 3$ and DGLA/ARA in high-18:2 $\omega 6$ group). The qPCR analyses of twenty-three genes showed that both high- $\omega 6$ and high- $\omega 3$ groups had similar bacterin-dependent induction of some transcripts involved in lipid metabolism (*ch25ha* and *lipe*), pathogen recognition (*clec12b* and *tlr5*), and immune effectors (*znrf1* and *cish*). In contrast, these transcripts did not significantly respond to the bacterin in the “Switched-diet” group. Concurrently, biomarkers encoding proteins with putative roles in biotic inflammatory response (*mfrsf6b*) and dendritic cell maturation (*ccl13*) were upregulated, and a chemokine receptor (*cxcrl*) was downregulated with the bacterin injection regardless of the experimental diets. On the other hand, an inflammatory regulator biomarker, *bcl3* was only significantly upregulated in the high- $\omega 3$ fed group and a C-type lectin family member (*clec3a*) was only significantly downregulated in the Switched-diet group with the bacterin injection (compared with diet-matched PBS injected controls). Transcript fold-change (bacterin/PBS) and FA associations highlighted the role of DGLA (20:3 $\omega 6$; anti-inflammatory) and/or EPA (20:5 $\omega 3$; anti-inflammatory) versus ARA (20:4 $\omega 6$; pro-inflammatory), as representative of the anti-inflammatory/pro-inflammatory balance between eicosanoid precursors. Also, the correlations revealed associations of FA percent and ratios with several eicosanoid and immune receptor biomarkers (e.g., DGLA/ARA significant positive correlation with *pgds*, *5loxa*, *5loxb*, *tlr5*, and *cxcrl*). In summary, dietary FA profiles and/or regimen modulated the expression of some immune-relevant genes in Atlantic salmon injected with *R. salmoninarum* bacterin. The modulation of Atlantic salmon responses to bacterial pathogens and their associated antigens using high- $\omega 6$ /high- $\omega 3$ diets warrants further investigation.

MOLECULE TO ECOSYSTEM: ENVIRONMENTAL DNA AS NEXUS OF COASTAL ECOSYSTEM SUSTAINABILITY FOR MAINE

David Emerson^{1*}, Mike Kinnison², Kate Beard², Heather Leslie^{2,3}, and Kody Varahramyan²

¹Bigelow Laboratory for Ocean Sciences

²University of Maine, Orono; ³Darling Marine Center

In 2019 the National Science Foundation in the US funded a major research, education, and outreach program focused on the development of eDNA science as a means to aid coastal ecosystem monitoring in the state of Maine. The Maine eDNA project has three research themes: 1) Sustainable Fisheries, focused on alewife ecology and restoration, and ‘the larval black box’ -- revealing early life ecology of larval of mussels, scallops, and kelp; 2) Harmful and Shifting Species, focused on identifying community and biogeochemical processes associated with harmful marine and freshwater algal blooms, and monitoring of range changes of kelp beds and associated ecological impacts along the Maine Coast. The third theme, Macrosystem eDNA Integration, includes a large, systematic eDNA water sampling effort spanning several nearshore coastal to inland freshwater ecosystems; evaluating the role of microbes in blue carbon sequestration from kelp aquaculture, and a science of team science component focused on communication factors related to this large transdisciplinary science project. Maine eDNA includes a large educational component focused around a newly recruited cohort of graduate students, undergraduates, and postdocs, as well as K-12 outreach programs. Additionally, there is substantial engagement with local state agencies responsible for marine and freshwater fisheries, and coastal management, as well as conservation organizations interested in water quality issues, and professional aquaculture growers concerned about resource sustainability. Some highlights from this effort, thus far, are development and validation of numerous eDNA assays for coastal marine and freshwater species, including those of interest to aquaculturists, bioinformatic pipelines for unified assessing of metabarcoding data, and hosting a workshop on the ethics of eDNA. In terms of new partnerships, a new graduate eDNA course integrated student projects directly with partners, both for-profit and non-profit, that can benefit from eDNA science; the Maine eDNA project has also led efforts to develop biocultural labels with Wabanaki Tribal Nations related to eDNA documentation in native ancestral lands, among other activities. This presentation will provide an overview of these topics, with an emphasis on those related to aquaculture, in hopes that we can share lessons learned, as well as expand partnerships with the goal of further developing eDNA science to integrate across scientific disciplines, and aid in the sustainable management of coastal ecosystems.

PROFILING THE GLOBAL GENE EXPRESSION RESPONSE OF ATLANTIC SALMON (*Salmo salar*) TO *Renibacterium salmoninarum* INFECTION

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Bacterial Kidney Disease (BKD), which is caused by a Gram-positive, intracellular bacterial pathogen (*Renibacterium salmoninarum*), affects salmonids including Atlantic salmon (*Salmo salar*). However, the transcriptome response of Atlantic salmon to BKD remained uncharacterised. A 44K salmonid microarray platform was used to improve our understanding of the transcriptome response of Atlantic salmon to BKD. Fish (54 ± 6 g) were infected with *R. salmoninarum* and, after 13 days post-infection, head kidney samples were collected. Via the TaqMan-based estimation of the pathogen load in those samples, fish ($n=5$) with no (Control), a high (H-BKD) or a low (L-BKD) infection level were selected for microarray analyses. There were 6,766 and 7,729 differentially expressed probes (ANOVA: $p < 0.01$) in the H-BKD and L-BKD groups, respectively, compared with non-infected control group. We identified 357 probes responsive to the infection level (H-BKD vs. L-BKD). Our Gene Ontology (GO) term enrichment analysis showed dysregulation of several adaptive and innate immune processes in BKD-infected Atlantic salmon. Adaptive immune pathways related with lymphocyte differentiation and activation (e.g. lymphocyte chemotaxis, T-cell cytokine production, T-cell activation and immunoglobulin secretion), as well as antigen-presenting cell functions (e.g. MHC class I-related processes), were found to be dysregulated in response to BKD. The infection level-responsive transcripts were associated to several mechanisms including the JAK-STAT signalling pathway, B-cell differentiation and interleukin-1 responses. Sixty-five microarray-identified transcripts were used for qPCR validation, and all of the transcripts showed the same fold-change direction as microarray results. The present study revealed diverse immune mechanisms dysregulated by BKD in Atlantic salmon, and enhanced the current understanding of Atlantic salmon response to bacterial pathogens. The identified biomarker genes can be used in the development of therapeutic diets and other strategies to improve Atlantic salmon resistance to BKD.

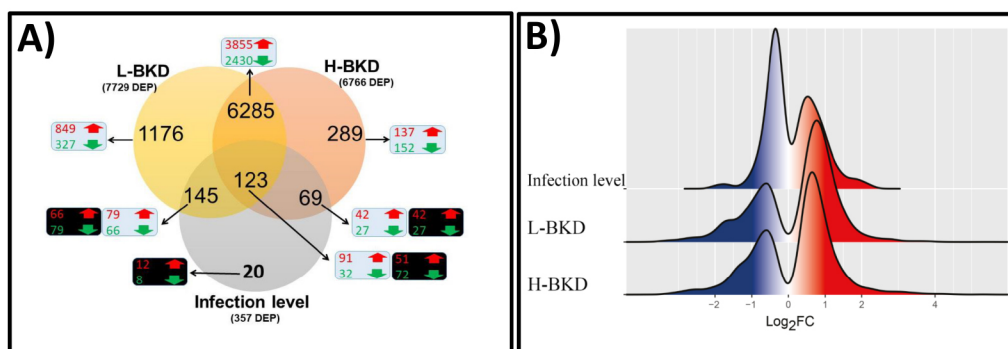


Fig. 1. A) Overview of microarray results. Differentially expressed probes (DEP) identified by ANOVA ($p < 0.01$). The number of up-regulated (red) and down-regulated (green) probes in BKD- and infection level-responsive lists are shown in blue and black boxes, respectively. **B)** Histogram of gene expression profiles of DEP in different comparisons. FC: fold-change.

OceanDNA: INFORMING SUSTAINABLE OCEAN MANAGEMENT AND ACTIVITY THROUGH ENVIRONMENTAL GENOMICS

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Conventional methods for ecological assessment based on catch-and-look approaches are expensive, often imprecise, and can harm species or sites they intend to monitor. The OceanDNA project, supported by the Canadian Ocean Supercluster, aims to revolutionize how we assess, monitor, and characterize the ocean by sampling DNA from the environment (eDNA) to identify what lives there, from bacteria to marine mammals, with a single unified sampling approach.

The OceanDNA program encompasses five major research projects aimed at advancing the use of eDNA in ocean monitoring. (1) We developed an eDNA sampling protocol with our partners from Nunavut Fisheries Association and Fisheries and Oceans Canada that can be widely deployed on any vessel to sample marine environments at depth. (2) We designed and compared two types of eDNA analyses for target species detection for monitoring commercial fish species and by-catch. (3) We designed a new quantitative sequencing method for eDNA analysis that is comparable to qPCR. (4) We used genome sequencing to develop new assays for monitoring specific genetic variants or populations within two commercial species using eDNA. (5) We integrated whole community biodiversity data from eDNA with complementary biodiversity from conventional surveys and environmental data to form new, enhanced ecosystem models. These combined models offer greater explanatory and predictive power than models based on eDNA or conventional biodiversity surveys alone.

This talk will include an overview of the OceanDNA project and presentation of key results in field sampling method development, quantitative analysis advances, and ecosystem modeling innovations. Environmental genomics offers unparalleled levels of detail about biodiversity and is an affordable solution to sustainable ocean management.

CLEANERFISH AQUACULTURE IN THE UNITED STATES, WITH AN EMPHASIS ON LUMPFISH RESEARCH IN NEW HAMPSHIRE

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American aquaculture researchers are making steady progress in transferring lumpfish, *Cyclopterus lumpus*, rearing technology from their Canadian and European counterparts to their own facilities to catalyze the use of cleanerfish for Atlantic salmon and steelhead trout ocean farms in the United States, specifically in Maine and New Hampshire. Since 2019, lumpfish have been reared from egg to adult at the University of New Hampshire's Coastal Marine Laboratory. In Maine, at the University of Maine's Center for Cooperative Aquaculture Research and at the United States Department of Agriculture's National Cold Water Marine Aquaculture Center, lumpfish culture and research also occur. Further, the US Lumpfish Consortium, made up of additional research institutions and aquaculture businesses, is working collectively to address some of the barriers that limit cleanerfish use, in general, and to transfer all known technology to the US aquaculture sector. An overview of ongoing lumpfish research in the US, with an emphasis on NH-based studies, including a variety of projects focused on lumpfish hatchery needs, using lumpfish in salmonid farms, and wild lumpfish population dynamics, will be presented. The hatchery studies focus on juvenile nutrition and feeding; tank modifications and fish behavior; understanding causes of and reducing lumpfish cannibalism and aggressive behaviors; and controlling timing and duration of broodstock spawning through photothermal manipulation and hormonal implants. Farm studies focus on the effectiveness of using lumpfish to control sea lice on steelhead trout. Wild lumpfish studies focus on mapping lumpfish distribution throughout the US Gulf of Maine and determining how populations have changed over time, especially in relation to water temperature; providing scientific guidance to resource managers to ensure that lumpfish are harvested sustainably for use in emergent lumpfish aquaculture facilities; and understanding lumpfish occurrence patterns so that emergent US lumpfish hatcheries are able to utilize the most local populations to maintain the local genetic integrity of fish slated for release into net pen salmonid farms. These studies are funded by the National Oceanic and Atmospheric Administration's Saltonstall-Kennedy Program, New Hampshire Agricultural Experimental Station, New Hampshire Sea Grant, Northeastern Regional Aquaculture Center, and the United States Department of Agriculture's National Institute of Food and Agriculture.

SWOT METHOD AND MATRIX IN A COMPARATIVE MODEL OF TRADITIONAL AND MODERN SYSTEMS IN STURGEON FARMS

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In recent years, due to the sharp decline in sturgeon stocks in the Caspian Sea, the development of sturgeon farms has increased sharply. In this study, according to the statistics and information collected from sturgeon farms using the Swot method to evaluate the strengths and weaknesses of existing farms as well as opportunities and threats from development.

According to the existing development outcome to appropriate policies to achieve development. Stable Options are offered as a suggestion, the material obtained in the square swot model can guide us to move this activity and its direction towards stability or its instability.

In the Swot model, solutions to any issues and problems are suggested as a solution. This model is descriptive and the sum of its results can be presented for proper planning in the medium term. This model is one of the most famous simple models available to provide the basic conditions for development. The suggestions provided by this model are practical and can be implemented.

MATHEMATICS ECOLOGY MODELING METHOD OF NATURAL LAKES ECOSYSTEM INLAND WATERS IN IRAN

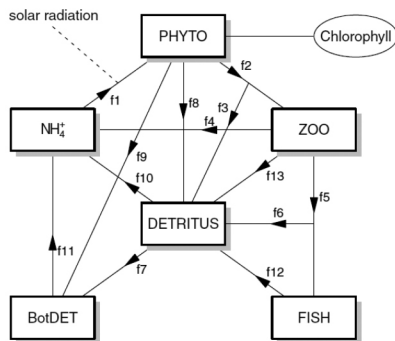
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Simulation is usually applied to study different subject's especially ecologic matters but that is very important that most of ecologic matters are qualitative. If we want to study them, it is essential that these qualitative subjects be verified to quantitative subjects. There are some kinds of methods for changing the qualitative matters but mathematic method is as important methods. Computer programs can always compile graph or solve the mathematic matters by software. Some environmental specialized soft wares include Mathematic, MAPLE, and MATLAB and also different languages of programing can help us for this case. So we find the relation between FORTURN and R in solving ecologic matters. All indicators are seen based on their relations and also in time function in mathematic model. So modelling application is more extensive than this subject that ecologic relation can be stated mathematical method. Therefore, if we want to use mathematic model for energy relation in different indicators of this lake, we can state this matter as follows it is suggested that mathematic and figuring models evaluate inland watery ecosystems. The future views of watery ecosystems and continuation of present process are studies by mathematic model. Ecologic matters and problems of natural lakes are studying modelling quantitatively that we can solve some of them. If it is necessary that incoming damages into watery ecosystems be studied, besides mathematic model, economic or ecologic- economic models can be applied.

$$\begin{aligned}\frac{dPHYTOC}{dt} &= f1 - f2 - f8 - f9 \\ \frac{dZOOC}{dt} &= f2 - f3 - f4 - f5 - f13 \\ \frac{dDETRITUSC}{dt} &= f3 + f8 + f6 + f12 + f13 - f7 - f10 \\ \frac{dFISHC}{dt} &= f5 - f6 - f12 - f14 \\ \frac{dBOTTOMDETRITUSC}{dt} &= f7 + f9 - f11 \\ &= -f4 - f10 - f11 - f14 + f1\end{aligned}$$

$$\begin{aligned}\frac{d[PHYTOC + ZOOC + DETRITUSC + FISHC + BOTTOMDETRITUSC]}{dt} &= \frac{dPHYTOC}{dt} + \frac{dZOOC}{dt} + \frac{dDETRITUSC}{dt} + \frac{dFISHC}{dt} + \frac{dBOTTOMDETRITUSC}{dt} \\ &= f1 - f2 - f8 - f9 + f2 - f3 - f4 - f5 - f13 + f3 + f8 + f6 + f12 + f13 - f7 - f10 + \\ &\quad + f5 - f6 - f12 + f7 + f9 - f11\end{aligned}$$



$$\text{NitrateUptake} = rmax \cdot \frac{NO3}{NO3 + ks_{NO3}} \exp(-InhibitionCt \cdot NH3) \cdot ALGAE$$

Figure 1: model status of relation among ecological indices in small lakes

ASSESSING THE EVOLUTION OF VIRULENCE OF THE SALMON LOUSE (*Lepeophtheirus salmonis*) IN THE BAY OF FUNDY

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An unintended consequence of fish farming is the production of an environment which may be more conducive for the evolution of virulence of pathogens to the host in production. Rearing conditions used in salmon aquaculture such as high densities, compressed rearing cycles, limited genetic diversity of broodstock as well as the practices applied to control infectious disease (e.g., vaccination, breeding for disease resistance, therapeutants), may all lead to an increased selection pressure for the evolution of virulence in parasites and pathogens. These selection pressures may be occurring without the balancing of negative fitness costs of virulence since a new supply of hosts enter the system every 18-24 months. The emergence of highly virulent strains of pathogens in salmon aquaculture may not only cause concern for further economic impacts to the industry but may also result in elevated disease spillover and subsequent risk for wild populations. To our knowledge, changes in the virulence of Canadian louse populations have not yet been assessed, despite having been identified as a crucial knowledge gap to understand the interactions between farmed and wild hosts as well as the impact of increased disease prevalence at sites which harbour highly virulent strains.

For this project, we developed a virulence challenge model by rearing lice originating from wild and farm sources for 7 generations in a common garden experiment and compared virulence on their host, testing the hypothesis that lice originating from farms were more virulent than those from wild sources. RNA sequencing was completed for copepodid, chalimus II and adult stages of the louse as well as their respective attachment sites on the host to determine responses at the host-parasite interface. Additionally, high resolution photos were taken of attachment sites and scored based on lesion size and colouration. Lastly, additional skin, louse and gill tissues were taken for further RT-qPCR analysis and RNA sequencing validation.



Figure 1. Generation 7 adult female *Lepeophtheirus salmonis* recovered from surface of a host Atlantic salmon (*Salmo salar*).

CHANGES IN TEMPERATURE OVERRIDES LIGHT INTENSITY EFFECTS AND TRIGGERS EARLY MATURATION IN PRE-SMOLT ATLANTIC SALMON

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Introduction:

Timing of maturation in salmon is a highly plastic process. The resultant life cycle depends on an interaction between the genetic makeup of the fish and environmental conditions (Swanson *et al.*, 2011). It's known, that there are some factors as growth and body fat levels that influence the onset of puberty in critical seasonal periods, however at commercial scale is not so clear how this model work to explain the high rates of mini-jacks observed. According to Bioled® gonadal ultrasound monitoring, it has been identified a series of freshwater facilities with a high prevalence of minijacks before the seawater transfer

Materials and methods:

The study was carried out in RAS experimental facility ATC Patagonia, in Puerto Montt, Chile. 4 experimental groups were formed, each group in duplicate, with different regimes of light and temperature as described in the scheme of fig 1 (Fig. N°1).

Results:

Only in the gr

oup 2 and 3 were observed effect on the entrainment in maturity in different degrees Fig 2.

Conclusions:

This is the first study where the effect of extreme changes in temperature and decrease in light intensity to very low levels are studied in Atlantic salmon. The obtained results indicate that temperature can act as an enabling factor in the early stages of sexual maturation in salmon during 8:16 and 24:0 photoperiod in pre-smolt Atlantic salmon. Light intensity seems to have a minor role as an environmental cue at this stage in the salmon life cycle to trigger maturation and the mechanism involved is still unknown. This may be of particular importance for FW salmon producers in flow through systems in warm regions such as northern salmon producing regions in Chile and Tasmania, also for production in RAS closed-containment where high temperatures are frequently observed.

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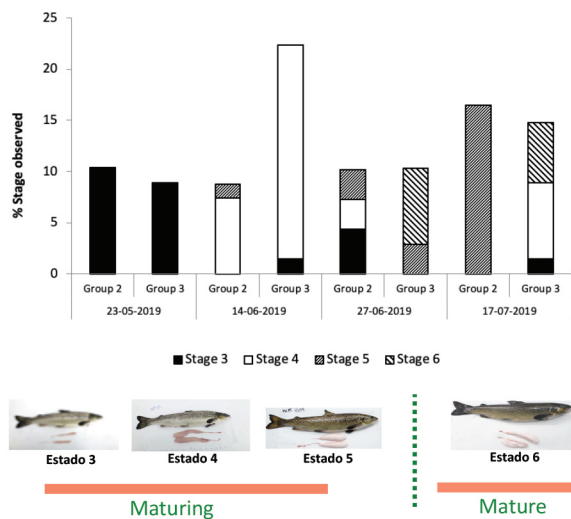


Fig. N° 2. Representation of the gonadal development by histology examination in groups 2 and 3 from May 25 until July 17. Bars show the percentage of each stage of gonadal development observed in the different treatment groups.

Table 1 - Number of observations (*n*) refers to fish with or without sexual maturation according to histological analysis in Atlantic salmon exposed to different light intensities and temperatures.

Maturity	Control group	Group 1	Group 2	Group 3	Total
Inmature	79	77 ns	57***	50***	263
Mature	1	3 ns	23***	30***	57
Total	80	80	80	80	320

Asterisks denote significant differences when compared with Control group (ns = $p > 0.05$, *** = $p > 0.001$).

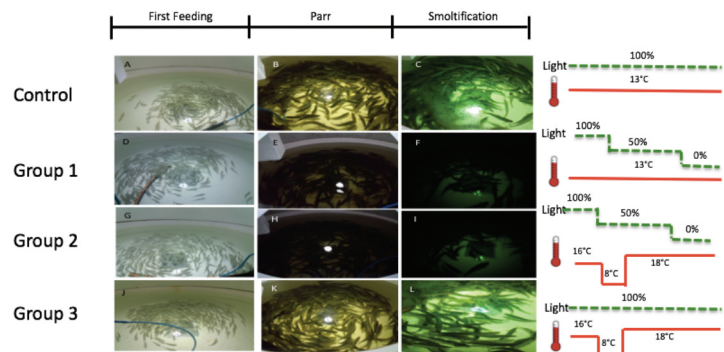


Fig. N° 1. Experimental design scheme describing different light and temperature regimes .

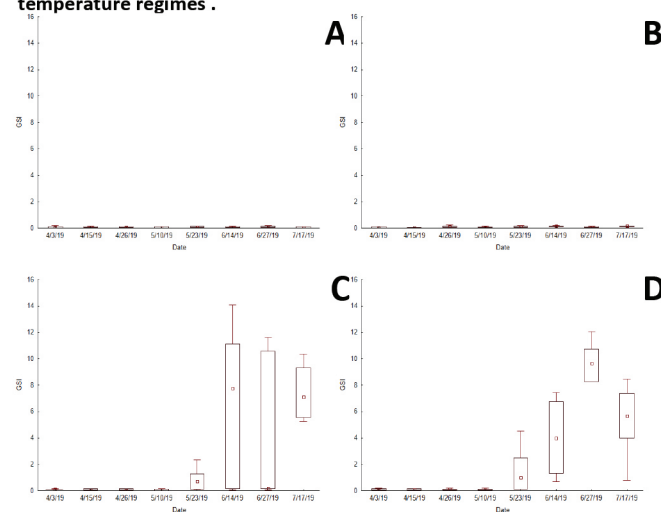


Fig. N° 3. Gonadosomatic index (GSI) of male of male pre-smolt Atlantic salmon during the progression of the spermatogenesis in the different sampling points. A) control group, B) group 1, C) group 2, D) group 3

Table 2 Relatives frequencies for the genotypes EE, EL y LL in each tank

Tank id	EE	EL	LL
Tk 4	0.1	0.7	0.2
Tk 5	0.1	0.5	0.4
Tk 6	0.2	0.4	0.3
Tk 7	0.2	0.5	0.3
Tk 14	0	0.5	0.5
Tk 15	0.4	0.4	0.2
Tk 16	0	0.5	0.5
Tk 17	0.2	0.5	0.3

LOW-COST DENITRIFICATION IN BRACKISH WATER AQUACULTURE PRODUCTION

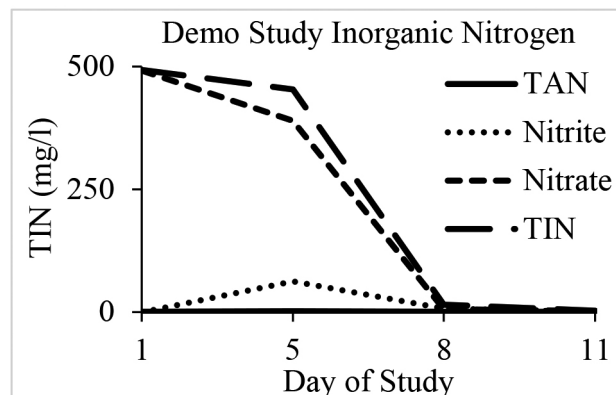
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Nitrate accumulation is a primary limiting factor of water reuse in many recirculating aquaculture systems (RAS). Water reuse is critical in inland marine RAS, as expensive artificial sea salts are often used to create salt water. Denitrification, a process in which facilitative anaerobic microbes reduce nitrate to dinitrogen gas, may allow producers to reuse water for extended periods of time. This may result in significant cost savings for producers and may have positive environmental impacts by reducing wastewater discharge. This presentation will cover several past and ongoing trials investigating simple, low-cost denitrification methods in brackish water aquaculture systems.

Three studies and a demonstration project were conducted to examine different denitrification strategies. The first study examined the effectiveness of simple, external denitrification chambers containing different media types. The treatments included plastic bio-media (BM), expanded clay pellets (EC), woodchips (WC), and a control (CO) using no media. A second study examined the addition of ethanol at a 2:1 carbon: nitrogen ratio to 4 different treatments filled with water from an active shrimp production system. The treatments were aerated with ethanol, aerated with no ethanol, no aeration with ethanol, and no aeration with no ethanol (AWE, ANE, NAW, and NANE). The third project examined denitrification in used aquaculture water using different carbon: nitrogen ratios (1:1, 3:1, 5:1, 0:1) created through ethanol additions in systems with no aeration. The demonstration project tested a denitrification method using ethanol at an active shrimp farm in a 17 m³ shrimp tank.

The first study showed that all media types and the control (no media) can denitrify to some extent, which led to further investigations using low oxygen/low ORP environments in bodies of water without an external denitrification chamber. The second study showed a significant amount of partial denitrification in the NAW treatment, but this treatment also had a large amount of nitrite accumulation, indicating insufficient carbon or insufficient time. The third study showed over a 95% reduction of nitrate in the 1:1 and 3:1 treatments over the course of 27 days. The demonstration study showed a significantly faster rate of denitrification, likely due to temperature. Nitrate fell from 495 mg/l to 2.9 mg/l in 11 days and only required 35 L of ethanol to denitrify 17 m³ of water. This has important implications for shrimp producers, as a rapid and low-cost denitrification method may significantly increase the longevity of water reuse and greatly reduce water discharge and salt use.



INTERACTIONS AND CONSEQUENCES OF HYBRIDIZATION OF FARMED ATLANTIC SALMON OF NORTH AMERICAN AND EUROPEAN ORIGIN WITH WILD SALMON OF NEWFOUNDLAND

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Escapes of farmed Atlantic salmon from net pens can lead to interactions and hybridization with wild salmon populations. An understanding of the outcomes and consequences of this, however, remain limited, particularly in North American waters. A single, large escape event in Newfoundland, resulted in wide-spread interbreeding (27.1% hybrid or feral juveniles, 17 of 18 sampled rivers). The presence of domestic alleles from this cohort decreased annually, consistent with selection against domestic genotypes in the wild, though persistent introgression involving backcrosses to wild fish was evident. In Newfoundland, the current farmed stock derives from the St. John River, over 1,000 km distant, and there has been recent site-specific permission to farm European origin (EO) salmon as triploids, which are highly genetically divergent from North American (NA) salmon. To assess the potential interactions and consequences of hybridization among these farm strains and wild Newfoundland (NF) salmon, we created a series of controlled crosses. The offspring from these crosses were then used to quantify trait and performance differences.

We undertook experiments to compare the early-life traits of NA and EO farm strains relative to their wild conspecifics and hybrids, including early development, gene expression profiles as late yolk sac fry, juvenile behaviour in different contexts and competition and growth in semi-natural stream and tank environments. While there were clear differences in early development, most of these were driven by population of origin and maternal effects associated with egg size. The gene expression study indicated greater transcriptome differences of the wild population from the EO than the NA farmed strain. Moreover, the largest differences in global gene expression were between the two farmed strains, with few significantly differentially expressed transcripts between F_1 hybrids and domesticated/wild maternal strains. These findings indicated that most of the differences seen were driven by geographic origin, with little evidence of common differences due to domestication. By contrast, the juvenile behaviour study and the competition and growth study indicated clear differences related to effects of domestication, and less so geographic origin. Both NA and EO farm fish were similarly more explorative, bold, aggressive and dominant than wild fish and related hybrids. Furthermore, both farm strains outgrew wild conspecifics in tank environments, but only EO farmed fish outgrew wild fish in semi-natural environments. These results indicate domestication may be an important driver of behavioural, growth and competitive differences, while geographical origin is a key driver of transcriptome differences, with the latter implicating greater concern related to the escaped EO than NA farmed salmon for the productivity and viability of wild NF populations.

CO-CULTURE AND COMBINATION OF PASSIVE DUCK REPELLENT TO REDUCE DIVING DUCK FORAGING ON BLUE MUSSELS (*Mytilus edulis*) FARMS

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In Europe and Canada, the economic losses in blue mussels (*Mytilus edulis*) farms due to duck predation represent a major problem. Many deterrents tried in the past generally either focused on protecting mussels by isolating them in nets or cages, or on using active repelling techniques (sound, light, etc.). Those repellents are often expensive, stressful to duck population, only effective for a short period of time and do not take into consideration drifting ice.

The last type of diving duck repellent is classified as passive. Those repellents do not intend to induce stress in ducks, and generally work by hiding the mussels. To test if they can produce a synergistic protective effect on the mussel harvest, three passive deterrents - knotted rope, flexible duck fence and sugar kelp co-culture - have been used in different combinations for this study since summer 2019. The studied variables are the amount of mussel per linear meter, the biomass per linear meter (both mussel and kelp), the average mussel shell length at the end of spring 2020, as well as duck's behavioral response observed during duck migration in spring 2020.

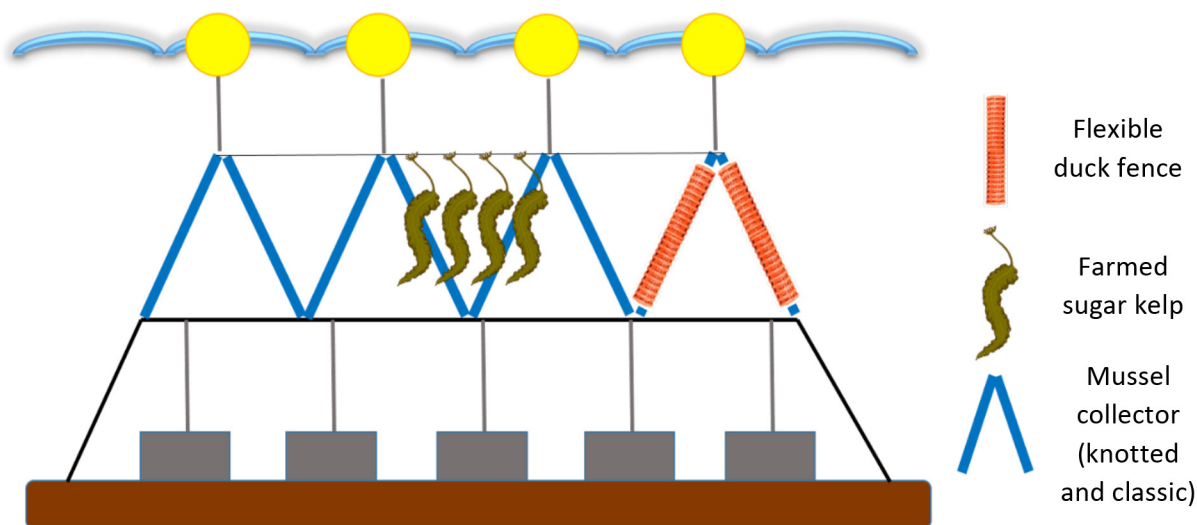


Figure 1 Floating mussel line design with the three passive repellents

VALORIZATION OF AQUACULTURE BYPRODUCTS : MARINE INVASIVE BIOFOULING ORGANISM (*Caprella mutica* or *linearis*) AS FOOD COMPLEMENT

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Each year, Quebec marine farmers produce grossly 350 tons, for a total worth of 2 millions CAD\$. Like many other marine farmers, they have to conduct their activities notwithstanding the many invasive biofouling organisms that colonize every year their structures. As of today, these community of organisms were solely viewed as a loss of gross profit due to cleaning costs and competition on their collectors. This project, ending in winter 2020, aimed to explore potential avenues of valorization for two of those biofouling species : blue mussel spat and skeleton shrimp. The project also shed light on the potential of a third group of organisms, the hydrozoan. This potential was later studied on three angles : the ease of collection for the marine farmer, the density found on the farming line and finally the nutritionnal content.

As space is the most limiting factor to implement new pieces of machinery on mussel farming boat, the experiment team developed the most efficient way to collect them without further handicap the limited space. The biofouling densities and species composition was not found to be consistent on the experimental mussel farming line and did not show significant correlation between the depth onto the collector and the biomasses found. Great potential have been observed in the nutritive content of the three groups of organisms, with high concentration of proteins and calcium, which we found could be of great interest to incorporate in livestock feed.

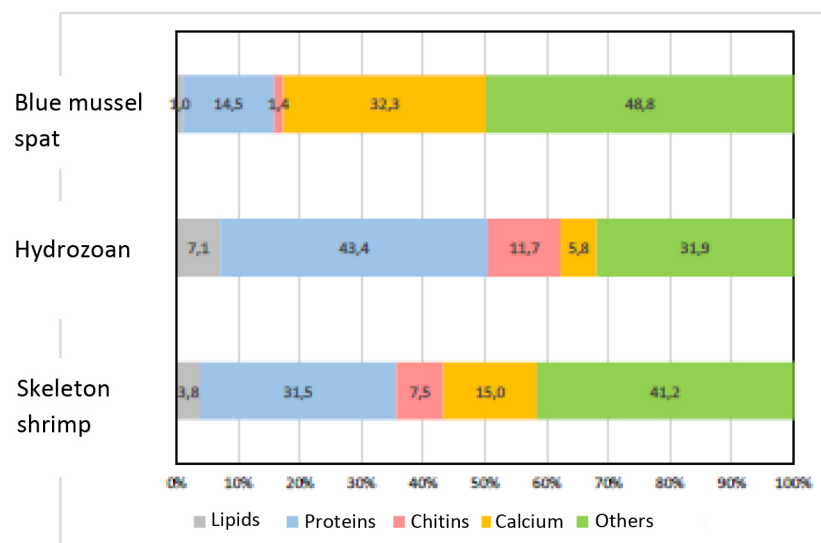


Figure 1 : Nutritionnal content (%) of skeleton shrimp, hydrozoan and blue mussel spat dry weight. Other ashes include all the ashes exept calcium.

3.

DEVELOPMENT OF VALUE-ADDED QUEBEC BY-PRODUCT MARINE GENERALIST FISH FEED

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Many native marine fish species have been targeted to stimulate the Quebec fish farming sector. During previous projects, the need for a fish feed specially developed for Quebec native marine fish species nutrient requirements was highlighted. It is well known that quality fish feed is the base for all successful growth and disease control. Currently, there is no specialised fish feed being produced in Quebec for those species. Therefore, feed developed for other species are currently used and may be linked to the reduction of egg quality and overall survival rates. This is suspected to be related to a deficiency of specific nutrients that would normally be found in natural prey. Coincidentally, Quebec legislation is being planned to forbid all landfill of organic matter in the future. Each year, many metric tons of organic matter are produced by marine production processing plants. Of those currently unused by-products, many have already been characterised and show great potential as fish meal alternative (crab, lobster, and shrimp meal). To further reduce the ecological impact of fish feed production, other by-products from other types of Quebec food production are also being studied. Thus, microbrewery grain by-products and maple syrup sugar by-product potentials as ecofriendly substitutes will also be explored during this project. Started at the end of summer 2021, the main goal of this project is to develop four different types of fish feed for Quebec marine carnivorous species. Those recipes would have to show different buoyancy profiles, maintain structural integrity during the time underwater, and show compatible nutritive contents.

To accomplish these objectives, two classes of students enrolled to become aquaculture technicians have been directly involved in the research process and physical testing. The first class was tasked to characterise the type of feed that would fulfill the biological and ecological needs of the targeted species. The second class visited the research facility and participated in the feed manufacturing process and laboratory physical experiments.

Full biochemical and microbiological tests will be undertaken during summer by the college's intern and Merinov's lab team. Ultimately, the food security and the nutritive content will be published to demonstrate the physical, chemical, and microbiological potential of those new ingredients.

YELLOWTAIL KINGFISH (*Seriola lalandi*) IN RAS TO MARKET – ARE WE MAD? THE NEW ZEALAND EXPERIENCE

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Recirculating aquaculture systems (RAS) have been used as a false flag to frustrate aquaculture developments – notably in Canada. Ingenuine actors without meaningful investment in aquaculture have insisted that RAS is a commercially viable system for whole of life production of Atlantic salmon. The contention that Atlantic salmon in RAS could be commercially competitive with conventional marine farming systems has not been proven. Marine based farmed salmon supply chains enjoy technical maturity and scale which culminates in exceptional efficiency.

Production of species such as Yellowtail kingfish have not achieved the scale or maturity which has been realised for Atlantic salmon. It is our view that the least-risk opportunity for RAS production rests with emerging species as the commercial entrants are not confronted with the possibility of stranding their existing conventional marine farming assets.

With our development partner, Northland Regional Council, we are completing construction of a RAS system designed to produce an estimated 600 tonnes p.a. of market sized Yellowtail kingfish. In the coming year, we will test our hypothesis that Yellowtail kingfish farming in RAS is a viable and preferred commercial proposition for New Zealand.

The key biological and technological findings from our existing RAS units which have informed our design will be presented. We will also review the economic, environmental, and social drivers which have influenced our investment decision. The challenges of realisation of a significant capital project in the face of supply chain disruptions will also be explored.



A NORWEGIAN PERSPECTIVE ON THE SUITABILITY OF TRIPLOID ATLANTIC SALMON FOR AQUACULTURE

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Preventing the interbreeding of wild and escaped farmed Atlantic salmon (*Salmo salar*) is an essential requirement of the Norwegian aquaculture industry if it is to be allowed to continue to expand. As such, the Institute of Marine Research (IMR) began to assess and develop production strategies back in the late 2000s to alleviate industry concerns surrounding the historic use of triploids, namely skeletal malformations and the development of cataracts. Subsequently, industrial green licenses were permitted for full scale trials throughout Norway starting in 2014. Here, we give a brief overview of the activity at IMR together with research and industrial partners in Norway as well as a summary of the main findings. For example, earlier laboratory studies found that compared to diploids, triploids have lower optimal rearing temperatures throughout the life-cycle whereas dietary requirements for phosphorus and histidine are higher in order to reduce issues with skeletal deformities and cataracts, respectively. During semi-commercial productions, triploid growth performance is generally equal to or better than diploids during freshwater rearing, but is more inconsistent in seawater with several large scale studies showing poor growth during seawater grow-out in comparison to diploids whereas other studies have shown equal or superior growth. For the industrial trials between 2014-2017, which includes data from 24 production groups of triploids, it was concluded that triploids showed promise as spring smolts, but less so as autumn smolts due to issues with wound development during the winter period. Analysis of the commercial trials post 2017 is ongoing, as well as work into smoltification and post smolt performance under different environmental conditions.

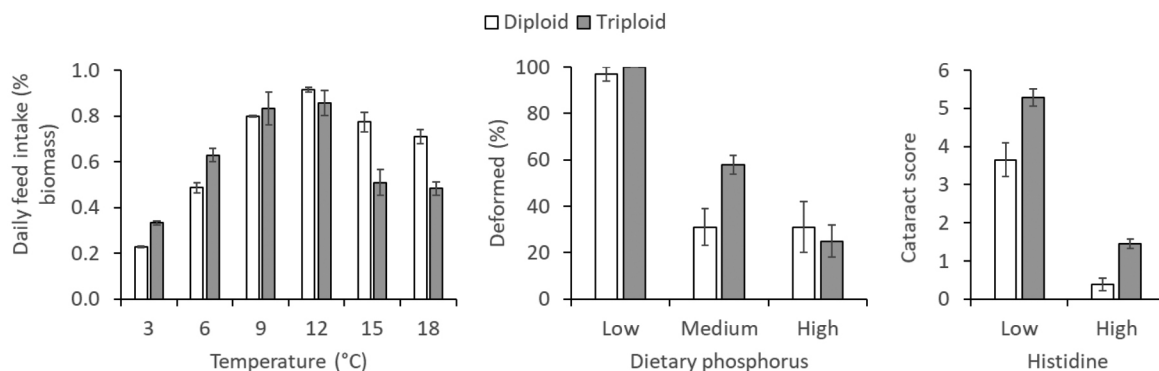


Figure 1. (A) Daily feed intake in large (2.5 kg) diploid and triploid salmon relating to rearing temperature. (B) The % of fish with one or more radiologically deformed vertebra in large (2.0 – 3.5 kg) diploid and triploid Atlantic salmon fed either a low (total phosphorus, 7.1 g/kg), medium (9.4 g/kg), or high (16.3 g/kg) phosphorus diet. (C) Cataract score in diploid and triploid post-smolts (approx. 400 g) fed either a low (10.4 g/kg) or high (13.1 g/kg) histidine diet. Data are means \pm SE.

VGLL3 IS ASSOCIATED WITH THE AGE OF PUBERTY IN FARMED ALL MALE ATLANTIC SALMON

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Atlantic salmon (*Salmo salar*) display a large degree of phenotypic plasticity regarding the age at puberty and this has been linked to the *vgll3* locus. In the current study, we aimed to establish the extent to which the *vgll3* alleles explain the likelihood of male salmon to enter puberty following environmental regimes that stimulate parr (i.e. dwarf males) and/or post-smolt (i.e. jacks) maturation. Initially we established an all-male line of Atlantic salmon using sex reversal to produce YY males that were used to fertilize XX eggs. We then conducted two experiments. Firstly, two YY males heterozygous for *vgll3* were crossed with four females, two of which were homozygous for the early *vgll3* maturation genotype and two were homozygous for the late *vgll3* maturation genotype. Fish were reared on a photoperiod and temperature regime known to induce post-smolt maturation. In the second experiment, one YY male heterozygous for *vgll3* was crossed with three females heterozygous for *vgll3*. These fish were reared on a regime that induced both parr then post-smolt maturation. In both parr and post-smolts studies, males with the early *vgll3* maturation genotype were significantly more likely to enter puberty as parr and/or post-smolts than those with the late maturation genotype, whereas those progeny heterozygous for *vgll3* were intermediate between the early and late genotypes. In addition, we also found some phenotypic females within our all male line (40/1384), of which 5% were genetically male.

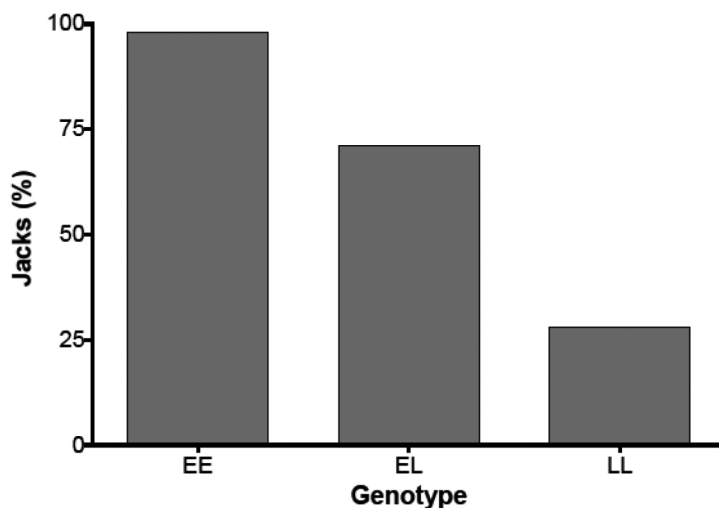


Figure 1. The % of mature males (jacks) in an all-male population of Atlantic salmon (approx. 290 g) with different *vgll3* genotypes. These fish were exposed to 16°C and constant light for 8 weeks after the winter signal cue ($n = 1312$, 289-619/genotype). EE = homozygous for the *vgll3* early maturation allele, LL = homozygous for the *vgll3* late maturation allele, EL = heterozygous for the *vgll3* alleles.

SALINITY DEPENDENT PERFORMANCE OF TRIPLOID ATLANTIC SALMON POST-SMOLTS

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Sterile triploids have been assessed for use in Norwegian Atlantic salmon farms, but their performance in seawater remains variable compared to diploid salmon. Here, we compare the performance of diploid and triploid post-smolts for 12 weeks following the expected time of smoltification in order to determine whether triploids have similar salinity optima as diploids. Underyearling diploid and triploid smolts were either maintained on freshwater (0 ppt) or had the water inflow changed to 11, 23, or 35 ppt ($n = 142$ / ploidy / salinity) and reared under continuous light at 12°C for 12 weeks. We assessed growth and smoltification markers as well as vertebral deformities and cataracts for which triploids are known to be more prone to than diploids. At the start of the experiment, the triploids were larger than diploids (mean weight (g); 88 ± 0.5 vs 72 ± 0.5). Molecular smoltification markers suggested triploids developed seawater readiness slightly earlier than diploids whereas growth at the different salinities may indicate differences in salinity optima. For example, final body weights were positively associated with salinity in diploids (final mean weight (g) \pm SE; 260 ± 5 , 305 ± 5 , 314 ± 6 , 337 ± 6 for 0, 11, 23, and 35 ppt, respectively), but not in triploids (final mean weight (g) \pm SE; 329 ± 6 , 358 ± 6 , 423 ± 8 , 413 ± 7 for 0, 11, 23, and 35 ppt, respectively). Cataract scores were not related to salinity, but were generally higher in triploids compared to diploids (mean \pm 95% CI; 2.8 ± 0.2 vs 1.9 ± 0.2 , respectively). Plasma ions and vertebral deformities are currently being assessed. In brief, early indications are that ploidy effects salinity optima that may impact on long-term seawater grow-out.

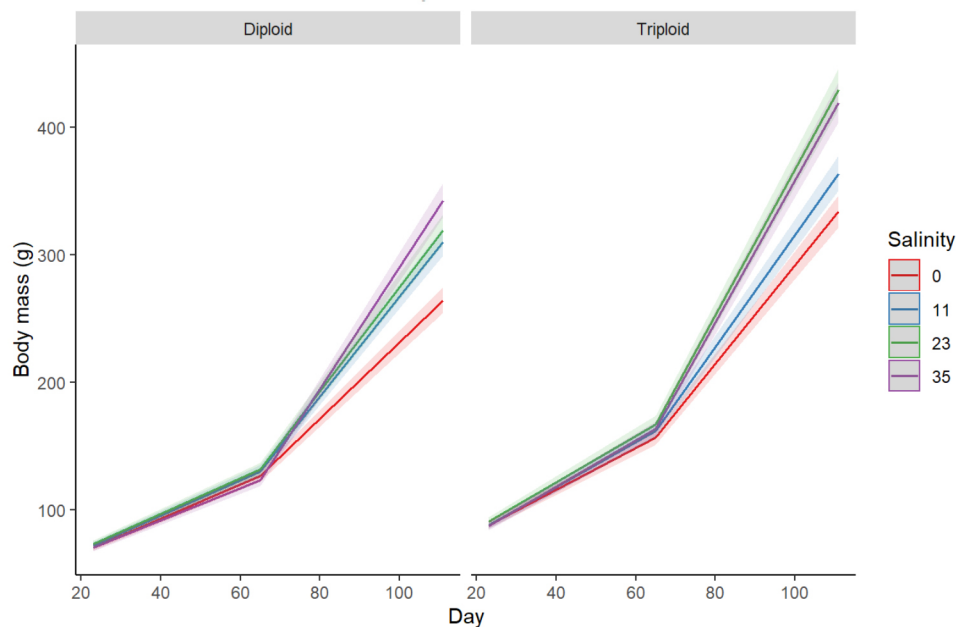


Figure 1. Body mass over time in diploid and triploid post-smolt Atlantic salmon maintained on 0, 11, 23, or 35 ppt for 12 weeks. Data are means \pm 95% CI.

CHALLENGES WITH SEA LICE MANAGEMENT IN BRITISH COLUMBIA

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Sea lice management in British Columbia, Canada is a significant challenge for Atlantic salmon farmers. Sea lice pressure has become more intense due to environmental factors exacerbated by climate change. Sea lice management in BC is regulated by the Department of Fisheries and Oceans Canada through the Marine Finfish Conditions of License and are based on keeping lice levels low during the outmigration of wild juvenile salmonids during the March to June period. As a result, sea lice are managed to a precautionary threshold to prevent risk of harm to wild juvenile salmonids. Preventative measures to reduce lice infesting farmed salmon, such as sea lice tarps are being investigated but can be difficult to implement due to environmental conditions. Control measures are limited with only one medicinal treatment, emamectin benzoate, that has a prolonged action against re-infestation. On top of this, *Lepeophtheirus salmonis* in BC are showing reduced sensitivity to the drug and the industry has no alternatives to protect their fish against re-infestation. All other forms of sea lice control require handling of the fish, cannot treat the whole farm or area at once, have seasonal limitations based on water quality and have no residual effect on sea lice re-infestation. Many of these difficulties stem from the regulatory confines resulting from the Conditions of License from DFO, the lack of new drugs being approved by Health Canada and the need to have Pesticides use Permits from the BC Ministry of Environment for using pesticides such as hydrogen peroxide. As a result of all these factors, it is challenging to practice effective integrative pest management.

DIET AND TEMPERATURE AFFECT LIVER LIPIDS AND MEMBRANE PROPERTIES IN STEELHEAD TROUT *Oncorhynchus mykiss*

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Fish can regulate cellular membrane fluidity in response to temperature changes by restructuring membrane lipid composition. The present study simultaneously evaluated the effect of diet and temperature on liver membrane fluidity in steelhead trout, an important aquaculture species. Adult steelhead trout (*Oncorhynchus mykiss*) were fed three commercial diets with different levels (30-40% of total fatty acids) of PUFA (lower ω 3- L- ω 3; medium ω 3- M- ω 3; higher ω 3- H- ω 3) from different sources (marine, terrestrial and vegetable oil); the effect of temperature changes (from 13.5°C to 18.0°C and back to 13.5°C) on liver cell membrane fluidity was measured.

Fish fed the H- ω 3 diet had the most linear response in Raman spectroscopy; this indicated that H- ω 3 fed fish can quickly adapt to changes in environmental temperature with the least affect on liver membrane physical properties, due to the higher polyunsaturate:saturate ratio in the diet counteracting the rigidifying influence of low temperature. L- ω 3 fed fish presented increased membrane fluidity at both temperatures (13.5 and 18.0°C), highlighting the influence of terrestrial fatty acids on the physical properties of the membrane. These results underscore changes in sterol: phospholipid ratios as a key response for membrane adaptability to environmental changes, as well as the necessity to include environmental variables when testing new diet formulations. Substitution of fish oil with vegetable oils may compromise sterol: phospholipid ratios, affecting the membrane adaptability to temperature. This study provides evidence of changes at the cellular level in liver tissue for fish fed different diets and subjected to different water temperatures.

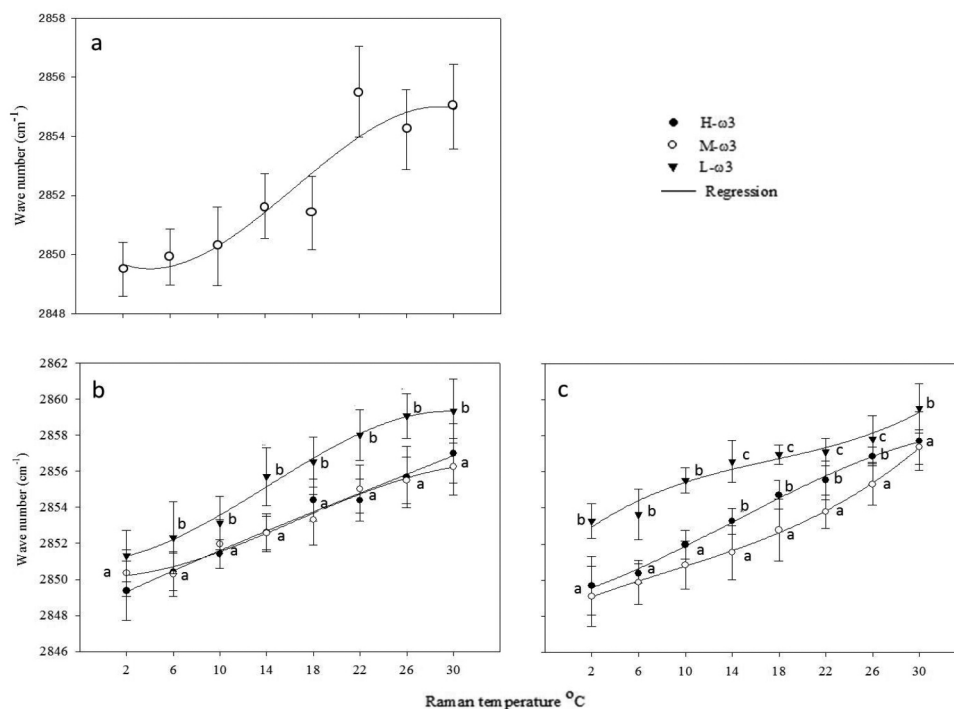


Figure 1: Raman spectroscopy results for trout fed (a) M- ω 3 feed (beginning of the experiment-13.5°C); (b) H- ω 3, M- ω 3 & L- ω 3 feed at 18.0°C; (c) H- ω 3, M- ω 3 & L- ω 3 feed (end of experiment-13.5°C).

TO WHAT EXTENT DO ‘HIGH’ WATER TEMPERATURES AFFECT THE PHYSIOLOGY OF ATLANTIC SALMON?

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Increases in average water temperatures and decreases in water oxygen levels (hypoxia), and more frequent and extreme warming events (i.e., heat waves), are occurring with climate change. Thus, there is an urgent need to understand the effects of prolonged and short-term warming on the physiology of cultured fishes, including Atlantic salmon (*Salmo salar*). This is particularly true with regard to Tasmania and Atlantic Canada where water temperatures have approached or exceeded 20°C, in combination with hypoxia, and negative effects on production and fish health (including large-scale mortalities) have been reported.

Over the past few years, we have used a multilevel (e.g., whole animal, biochemical, genomic, epigenetic) approach, and several experimental paradigms, to understand how high temperatures alone, and when combined with moderate hypoxia (60-70% air sat.), impact salmon physiology and production characteristics under realistic temperature scenarios (e.g., using an IT_{MAX} test; a 1°C increase week⁻¹ from 10°C).

In this presentation, we will show that while stress gene expression in salmon begins to be affected at 16°C, and feeding decreases dramatically as temperatures approach 20°C, there is little/no evidence that this temperature, even when prolonged or combined with moderate hypoxia, results in mortalities. The salmon’s capacity to mount an innate immune response is not compromised at these temperatures and plasma cortisol levels (indicative of a secondary stress response) do not increase until 21-22°C. Finally, in lab-based experiments, mortalities only begin when the fish reached 21°C, and even at 23°C mortalities are only ~ 30%.

Through this research, we have also been able to identify epigenetic and genomic markers of temperature and hypoxia tolerance in salmon (e.g. **Figure 1**), and have identified populations/families of salmon that have critical thermal maximum (CT_{MAX}) and IT_{MAX} values of ~28°C and 25°C, respectively. This should assist in the development genetic and other markers for use in selecting fish that are more tolerant of these conditions and for evaluating fish health.

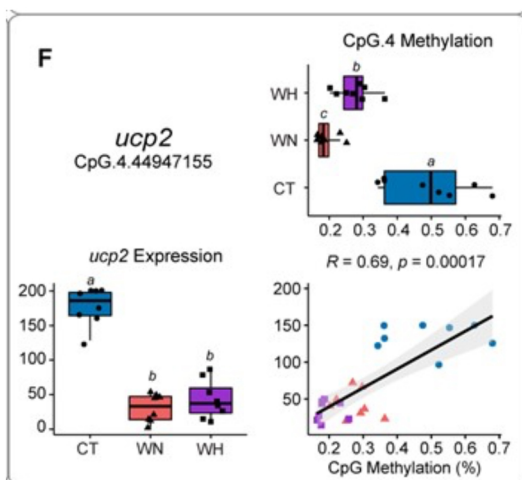
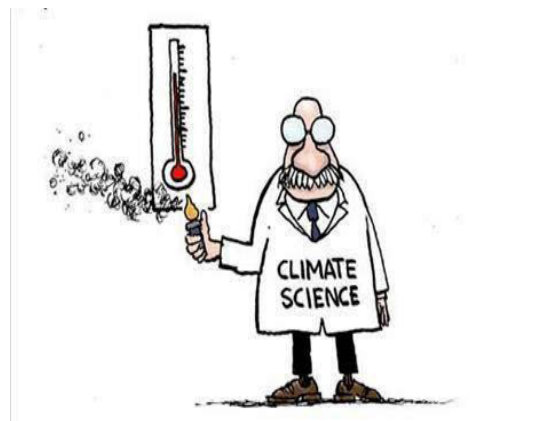


Figure 1: Correlation analysis between CpG methylation percentages (%) and corresponding gene expression (RQ) for mitochondrial uncoupling Protein 2 from Atlantic salmon that were exposed to control conditions (CT: 12°C, 100% air sat.), high temperature with normoxia (WN: 20°C, 100% air sat.), or high temperature with hypoxia (WH: 20°C, 70% air sat.) for 3-days or 4-weeks (n=8, N=48).



ABIOTIC AND BIOTIC CHALLENGES FACING SALMON AQUACULTURE IN THE ERA OF CLIMATE CHANGE: UNDERSTANDING AND MANAGING INTERACTIVE EFFECTS THAT RESULT IN MORTALITY

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Climate change is resulting in increases in average water temperatures and decreases in water oxygen levels (hypoxia), and more frequent and extreme weather events (i.e., storms, heat waves etc.). Given that salmon are predominantly reared in coastal marine sea-cages, this has led to a significant amount of research on the effects of high temperatures on the production, health and welfare of this species. However, recent research suggests that high temperatures (i.e., up to 20°C) and moderately low oxygen levels (e.g., down to 60-70% air saturation), by themselves, are unlikely to result in significant cage-site mortalities.

This was the impetus for us to take a comprehensive look at the abiotic and biotic factors that impact salmon at aquaculture cage-sites, their potential interaction with regard to causing mortalities, and management practices that might alleviate/mitigate, or exacerbate, the loss of fish under culture conditions. The model we have developed has many points of intersection/interaction, and includes factors such as algal blooms, nutrition, environmental variables, pathogens, the presence of 'nuisance' species (e.g., jellyfish), and management decisions and practices.

In this talk, I will provide some background information on the causes of salmon cage-site mortalities, and introduce an interactive model that details the interdependencies and causal relationships between the above and other factors. It is hoped that this model will: 1) improve our understanding of factors that influence salmon welfare and mortalities; 2) prove useful in mitigating these issues going forward; and 3) assist the industry and the research community in selecting directions/priorities for future R&D targeted at making salmon aquaculture more sustainable and profitable. In the future, we aim to transform this interactive model into a dynamic systems model that will predict the interaction between abiotic and biotic factors over time, and forewarn producers of conditions/events that might result in the loss of fish.



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PROVINCIAL-LEVEL MARINE AQUACULTURE HEALTH AND SAFETY REGULATION AND MANAGEMENT IN CANADA: FINDINGS FROM A COMPARISON OF COMPENSATION CLAIMS DATA, INSPECTION OUTCOMES AND HEALTH AND SAFETY OPTIONS IN PROVINCIAL REGULATIONS

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There are diverse and significant occupational health and safety (OHS) hazards in Canada and globally, as well as high rates of injuries relative to national/provincial averages. In Canada, occupational health and safety is regulated primarily at the provincial/territorial level and is broadly based on an internal responsibility system that requires the development of company-level health and safety management plans and encompasses workers' right to know, to participate in health and safety decision making and to refuse unsafe work. Most injured workers have access to workers compensation benefits funded by compensation premiums paid by employers, which also pay for inspection and prevention programs. Workplace deficiencies identified in inspections can result in orders and potential financial and legal penalties including, in rare cases, criminal prosecution. One way to explore potential strengths and weaknesses in this system is to compare source and type of injury for the marine aquaculture sector documented in compensation claims data with targeted deficiencies reported by inspectors and the main regulations cited by inspectors relative to the full envelope of regulatory options available to them. This presentation discusses findings from an analysis of claims data, inspection results and health and safety regulations in three Canadian provinces with marine aquaculture industries: Newfoundland and Labrador and New Brunswick on Canada's east coast and British Columbia on the west coast.

Objective and research questions: The objective of this research is to identify overlaps and gaps between each of these elements of the aquaculture health and safety system in order to identify potential ways to improve the effectiveness of provincial health and safety systems in detecting hazards and reducing injury risk. The research questions are:

- How do deficiencies identified by inspectors in visits to aquaculture operations map onto regulations and onto source and type of injury data found in compensation claims data across these three provinces.
- How might findings be used to help prevent future injuries in the sector?

Methods: A desktop review of health and safety regulations in Newfoundland and Labrador, New Brunswick and British Columbia; analysis and comparison of inspection results across the three provinces with: a) the regulatory scope available to inspectors to identify overlapping areas of focus and gaps; and b) information in provincial injury compensation claims on source and nature of injury data to explore the fit between injury patterns and inspection outcomes.

Findings: Compensation claims data for marine aquaculture indicate higher than average injury rates across all three provinces, along with similarities in the nature and source of injury. Information on source of injury is, however, unspecified in some of these data (i.e. New Brunswick) for up to one-third of injuries. There are both similarities and differences in inspection outcomes across the three provinces that may partly reflect the different histories of the industries. Inspections and deficiency reports tend to focus largely on deficiencies in physical infrastructure, training and in meeting formal health and safety system regulatory requirements. Relative to sources of injury in claims data, inspection reports have relatively little to say about deficiencies in on the water operations and about hazards associated with working on the water and hazards that might contribute to the high rate of soft tissue injuries in the sector. Compensation claims data potentially under-report hazards and injuries related to diving and some other activities captured in inspection reports.

INCIDENCE OF DEFORMITIES IN ATLANTIC SALMON: IMPACTS OF GENETICS AND ENVIRONMENT OVER TWO GENERATIONS

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Commercial broodstock programs are sometimes not able to effectively use genotyping to determine relatedness amongst individuals communally reared with all families in a year class. In such cases, families are held in individual tanks until such time that siblings are assessed and PIT tagged for identification before pooling within communal rearing tanks. This was the case for an Atlantic salmon breeding program for Northern Harvest Sea Farms, now Mowi Canada East, which maintained families in individual tanks at the Huntsman Marine Science Centre for the 2010-2018 year classes.

Over this period, every effort was made to standardize environmental conditions amongst individual family tanks and recirculating systems, with a focus on density, feed/feeding, temperature, oxygen saturation, mg per liter of oxygen, pH, CO₂, alkalinity, total ammonia, nitrite, nitrate, and total gas pressure. It is imperative to ensure that the culture environment was not providing a performance advantage to specific families that might otherwise be attributed to the genetic capacity of the family siblings. However, conditions and parameters can vary over time and the effect on prevalence of deformities within tanks must be quantified. A comprehensive deformity/abnormality code was developed to collect robust data on 240,733 individual fish (ranging from 15,747-35,799 per year class) for 765 families (482 sires, 504 dams) within each of the nine year classes.

Progeny from the families of each year class were randomly assessed and a portion PIT tagged when individuals reached a minimum size to make up the controlled challenge groups and future breeding nucleus. Atlantic salmon having any type of deformity or extreme abnormality were assessed but not tagged. Skeletal deformities assessed included: short opercula, jaw, head and spinal curvature. Abnormalities or irregularities, such as eye abrasions, fin erosion etc., were also recorded but, in some cases, these individuals were still PIT tagged (e.g., slight fin erosion). These abnormalities were recorded as potential environmental effects that could be attributed to tank density or other factors and might impact prevalence or severity of deformity.

This presentation will discuss the deformity/abnormality coding used (e.g., effect of technician calling severity), significance of environmental variables with attribution to differences in location of deformities, variation or lack thereof in percentages of deformities between year classes and over a generation(s), and estimated heritabilities of various deformities with discussion of the fixed and random effects ($h^2 \pm SE = \text{negligible to } 0.23 \pm 0.03$ across all year classes evaluated).

SYNTHESIZABLE AMINO ACIDS OF IMPORTANCE IN FISH NUTRITION

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In recent years it has become increasingly apparent that some amino acids previously classified as “dispensable” or “non-essential” can have significant impacts on fish when supplemented in diets. Included in this group are glutamine, glycine, hydroxyproline and proline. Other amino compounds or derivatives of amino acids such as taurine and creatine, which are abundant in fishmeal, also have gained increased attention as efforts continue to identify feedstuffs to potentially replace some portion of fishmeal in the diets of aquatic species to increase the sustainability and cost-effectiveness of aquaculture. However, many alternative protein feedstuffs are deficient in one or more indispensable as well as dispensable amino acids. Therefore, this laboratory intensified its efforts in recent years to evaluate dietary requirements for various amino compounds with red drum (*Sciaenops ocellatus*), hybrid striped bass (*Morone chrysops* x *M. saxatilis*) and channel catfish (*Ictalurus punctatus*).

Results from several different studies will be presented to provide detailed information about the synthesis of the amino acids traditionally classified as dispensable and how dietary supplementation may improve various metabolic responses. In particular, the presentation will review studies in which dietary glutamine supplementation was evaluated in terms of gastrointestinal health and immunological responses of the species noted above. Supplementation of glutamine at 1 to 2% of diet had beneficial effects on intestinal health and various immunological responses including enhanced vaccination efficiency. Additional studies were conducted with hydroxyproline nutrition of red drum because that amino acid is a vital component of collagen, and growth of other fish species has benefited with its supplementation. However, in our studies with red drum, we were unable to confirm beneficial effects of crystalline hydroxyproline supplementation in regards to growth of fish fed diets high in plant-protein feedstuffs. Changes in hydroxyproline content were found in plasma and tissues at higher inclusion levels, which could have some effect on the texture of fish flesh. Several investigations in this laboratory have established the dietary essentiality of taurine to red drum as well as its potential sparing effect on methionine in other fish species. The positive effects of dietary creatine supplementation on weight gain, feed efficiency, and reduced circulating homocysteine levels of red drum also has been observed in multiple trials. Supplemental creatine also was demonstrated to have osmoregulatory benefits in hybrid striped bass when cultured in brackish water. The beneficial effects of dietary creatine supplementation on channel catfish fry also was noted when reared in brackish (10 ppt) water. Results from these various studies emphasize the potential importance of a number of synthesizable amino acids and other nitrogenous compounds in the nutrition and metabolism of various fish species.

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FROM ATLANTIC SALMON TO STRIPED BASS CULTIVATION: INSIGHTS FROM A CONSERVATION AND FISHERIES MANAGEMENT PERSPECTIVE

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The province of Québec cumulates more than 140 years of restocking and expertise aimed at wild indigenous salmonid species such as the anadromous Atlantic salmon (*Salmo salar*). Annually, between 500 000 and 1 000 000 fishes are produced by Quebec public hatcheries to support recreational fishing and restore wild freshwater fish populations of nine species (Figure 1). Recently, large-scale re-introduction efforts through artificial breeding plans were initiated for walleye (*Sander vitreum*), striped bass (*Morone saxatilis*) and an endemic catostomid, the copper redhorse (*Moxostoma hubbsi*) for which a severe decline in its abundance and its distribution range has been documented.

Located in southern Quebec, the Baldwin-Coaticook public hatchery is a major player in intensive cold and coolwater fish production activities from broodfish conditioning to ongrowing purposes with its 12 state-of-the-art independent rearing systems. Successful yearly fingerlings production was achieved through the accelerated establishment of reproduction and natural rearing protocols. Significant contributions to the rebuilding of these populations are already observed in the wild.

Developing unique expertise and dealing with important intensive production challenges arising from our exclusive wild-caught broodstocks, the development of natural rearing protocols, the use of recirculating aquaculture technologies and the environmental regulations make Quebec public hatcheries contributing significantly in recovery action plans that have diversified and yield important incomes. Wishing to extend our facilities and expertise over the coming years, opportunities for further research and development partnerships will be sought and promoted.

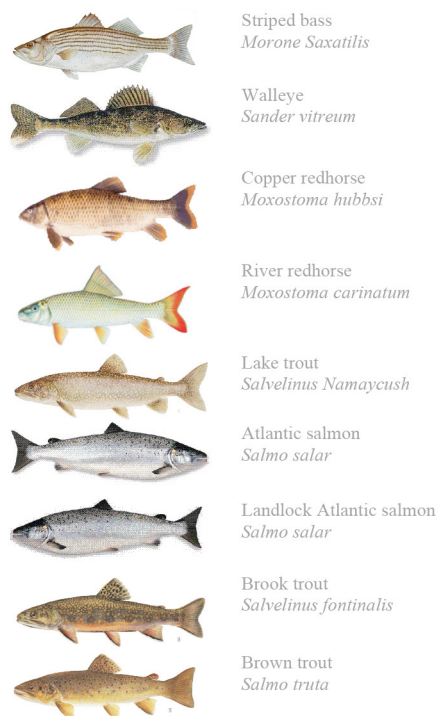


FIGURE 1. Fish species farmed by the public hatcheries of Quebec

EMERGING VARIANTS OF *Moritella viscosa*

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Winter ulcer disease caused by *M. viscosa* is a major problem for salmon farming industry as it causes animal welfare challenges and significant economic losses due to mortality and downgrade at slaughter. All commercial basis vaccines with a winter ulcer disease component currently available across markets contain a classic/typical strain of this pathogen.

During the past 5-6 years, it has become evident that vaccines with antigen based on classic *M. viscosa* isolates may not protect the fish sufficiently from disease caused by emerging atypical strains of this bacterium. A summary of the field situation in Norway as well as Canada shows a complex disease picture.

In this study we have compared the efficacy of experimental vaccines against classic- as well as variant *M. viscosa* challenge. Further, we have characterized a number of field isolates using gyrB- and OMP sequencing as well as antibody-based methods.

Our results show that there are likely several serogroups of *M. viscosa*. Experimental vaccines formulated using variant *M. viscosa* isolates show promising results against challenge by the emerging *M. viscosa* strains.

COMPARATIVE GENOMIC OF A NEW PATHOGENIC STRAIN ISOLATED FROM WINTER ULCER DISEASE IN ATLANTIC SALMON (*Salmo salar*) IN NEWFOUNDLAND

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Winter ulcer disease (WUD) is a recalcitrant issue in the salmon farming industry, usually caused by *Moritella viscosa*. The salmon farming industry has faced detrimental impacts from this disease. The mortality rate caused by WUD is around 10 % during an outbreak. Fish mostly survive with WUD, but as a consequence of large skin ulcers, and low quality of the filet, significant economic losses can be seen in the industry. Although different types of polyvalent vaccines are used against this pathogen, WUD outbreaks are continuously reported in Atlantic Canada. Currently, it is unknown whether undescribed bacterial pathogens are also causing WUD in vaccinated farmed Atlantic salmon (*Salmo salar*), or the effectiveness of current vaccines is not sufficient. In this study, we described the phenotype and genomic characteristics of a new pathogen (*Vibrio* sp J383) isolated from internal organs of Atlantic salmon displaying clinical signs of WUD.

Infection assays conducted in vaccinated Atlantic salmon revealed that *Vibrio* sp J383 can cause low mortalities when is intraperitoneally administered in high concentration (10^7 - 10^8 CFU/dose). *Vibrio* sp J383 persisted in the blood of infected fish at 10 and 12°C, clinical signs increased at 12°C, and no mortality and bacteremia were observed at 16°C. *Vibrio* sp J383 genome is composed of two chromosomes with a total genome size of ~5.9 Mb with ~3.6 Mb and ~2.0 Mb in chromosomes one and two, respectively. Phylogenetic and comparative analyses indicated that *Vibrio* sp J383 is closely related to *Vibrio splendidus*, with 93% identity. However, phenotypic studies showed significant differences between them. For instance, the optimal growing temperature for most of the *Vibrio* species is 28°C. In contrast, *Vibrio* sp J383 does not grow well at 28°C and displays an optimal growth between 15-18°C and substantial growth at 4°C. Also, *Vibrio* sp J383 displays hemolytic activity and siderophore synthesis at 15°C. Our finding suggests that *Vibrio* sp J383 may be adapted to cold temperatures and its distinctive characteristics compared to other *Vibrio* species suggest a novel species. This study introduced a new strain that causes winter ulcer disease in Atlantic salmon and highlighted the role of unknown pathogenic strains in this disease.

Table 1. phenotypic characteristics of *Vibrio* sp. J383

Characteristics	<i>Vibrio</i> sp. J383
Gram stain	-
Capsule stain	+
Hemolysin in Salmon blood agar (15°C)	+
Hemolysin in Sheep blood agar (15°C)	+
Hemolysin in Salmon blood agar(28°C)	-
Hemolysin in Sheep blood agar (28°C)	-
Type 1 fimbria	-
Growth in LB 0% NaCl (15°C)	-
Growth in LB 0.5% NaCl (15°C)	-
Growth in LB 2% NaCl (15°C)	+++
Growth in TSB 2% NaCl (4°C)	++
Growth in TSB 2% NaCl (15°C)	+++
Growth in TSB 2% NaCl (28°C)	+
Growth in TSB 2% NaCl (37°C)	-
Motility Test	+
Catalase	+
Oxidase	+

PLANT-BASED V CELL-BASED LABELING AND MARKETING CHALLENGES

Gavin Gibbons

Vice President, Communications
National Fisheries Institute

The rise of seafood amalgams in the marketplace, designed to compete with and or replace traditional seafood products, is growing. Meanwhile, the plant-based variety and cell-cultured variety have chosen to take dramatically different marketing and labeling approaches. The differing tactics have created a communications conundrum that sees one product line choosing an adversarial posture and the other an inclusive approach. How these different products have chosen to introduce themselves, to a market that features the oldest and healthiest animal protein on the planet, may determine their success or failure. Eating is only part of the equation when we're reminded to heed the warning, "watch your mouth."

PROFILING THE TRANSCRIPTOME RESPONSE OF LUMPFISH HEAD KIDNEY TO *Renibacterium salmoninarum* AT EARLY AND CHRONIC INFECTION STAGES

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Renibacterium salmoninarum is a Gram-positive pathogen that causes Bacterial Kidney Disease (BKD) in wild and cultured marine fish including salmonids, and non-salmonids like lumpfish (*Cyclopterus lumpus*) which is utilized as a living pest remover to biocontrol the sea lice (*Lepeophtheirus salmonis*) infestations in Atlantic salmon (*Salmo salar*) sea cages. Lumpfish susceptibility to *R. salmoninarum* has been reported recently. However, the transcriptome response of lumpfish to this immune-suppressive pathogen remains unknown. To profile the global gene expression response of lumpfish to *R. salmoninarum* at early and chronic infection stages, fish were intraperitoneally injected with either a high dose *R. salmoninarum* (1×10^9 cells dose⁻¹) or PBS (control), and head kidney samples were collected at 28 and 98 days post infection (dpi) for RNA sequencing. In addition, blood samples taken at 14, 28, 42, 56, 70, and 98 days post-infection were used for the lysozyme and indirect ELISA assays. Transcriptomic profiling identified 2019 and 147 differentially expressed genes (DEGs) in the 28 and 98 dpi, respectively. Pathogen recognition, inflammatory cytokines, immunoregulatory response, iron regulation, and interferon-induced effectors related genes were differentially regulated at 28 dpi, whereas cell-mediated immunity-related genes showed differential regulation at 98 dpi. The pathway enrichment analyses reveal several innate and adaptive immune pathways (i.e., NF κ B, MAPK, and JAK-STAT signaling pathways, TNF- α dependent killing pathways, MHC-dependent pathways) that were dysregulated in response to *R. salmoninarum* infection at early and chronic infection stages. Lysozyme activity and pathogen specific IgM titers of the infected fish coincided with the immune suppressive nature of the *R. salmoninarum* infection. In summary, *R. salmoninarum* causes immune suppression at early infection, whereas lumpfish induce cell-mediated immune response at chronic infection. The present study provides a more complete depiction of diverse immune mechanisms dysregulated by *R. salmoninarum* in lumpfish and open avenue to develop immune prophylactic tools to combat BKD.

ROLE OF RIBOFLAVIN BIOSYNTHETIC PATHWAY PARALOGS, AND RIBOFLAVIN TRANSPORTER (*ribN*) IN *Aeromonas salmonicida* VIRULENCE IN LUMPFISH

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Biologically active flavins from riboflavin (vitamin B2) are essential for intracellular redox reactions and extracellular bacterial physiology. Most bacterial pathogens can either synthesize riboflavin *de novo* or scavenge riboflavin from the host tissues through high-affinity transporters.

Aeromonas salmonicida is a Gram-negative pathogen of fresh and marine water fish and the etiological agent of furunculosis and its riboflavin supply pathways have not been studied. Here, we characterized the riboflavin provision pathways in *A. salmonicida* using *in-silico* and experimental approaches. We found that *A. salmonicida* possesses a riboflavin biosynthetic pathway (RBP) and a transporter *ribN*. Transcriptional orchestration analysis of riboflavin supply genes by RT-PCR revealed that the main RBP operon consists of *ribD*, *ribE1*, *ribBA*, and *ribH* genes, together with *nrdR* and *nusB* genes. *ribE1* and bifunctional *ribBA* from the main RBP operon have duplicated paralogs outside the main operon; *ribE2* (*ribE1*), *ribA*, and *ribB* (*ribBA*). Regulation analysis using RNA-Seq, and qPCR showed that the *ribB* transcriptional unit, conserving a putative FMN riboswitch, is negatively regulated by riboflavin. To study the role of paralogs found in Riboflavin Biosynthetic Pathway (RBP) and riboflavin transporter (*ribN*) in *A. salmonicida* pathogenesis and physiology, mutants of *ribE1*, *ribE2*, *ribBA*, *ribA*, *ribB*, *ribN*, and *ribA-ribE1* were constructed, characterized, and evaluated in the marine teleost, lumpfish (*Cyclopterus lumpus*). Groups of 60 fish were intraperitoneally injected with 0.1 ml (10^4 CFU/dose) of the respective *A. salmonicida* wild type and mutant strains. Tissue samples were collected at different time points to determine bacterial colonization. Mortality was recorded until 30 days post-infection (dpi). All fish died within 10 dpi from the Δ *ribE2*, Δ *ribBA*, *ribN*, and wild type infected groups, whereas 100% of the fish infected with Δ *ribE1*, Δ *ribA*, Δ *ribB*, and *ribA-ribE1* survived. Surviving fish were challenged with 10^3 CFU/dose (10 LD₅₀) of *A. salmonicida* wild type to determine the mutants' utility as live attenuated vaccine. The attenuated mutants (Δ *ribE1*, Δ *ribA*, Δ *ribB*, and *ribA-ribE1*) conferred protection with a low relative percentage of survival (10-20%). In summary, *ribE1*, *ribA*, and *ribB* of RBP play an essential role in riboflavin provision to *A. salmonicida* during host colonization and impacting bacterial virulence.

DIFFERENT APPROACHES OF DRYING ATLANTIC SEA CUCUMBERS (*Cucumaria frondosa*): PROS AND CORN

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Sea cucumbers (SC) are popular as a luxury, exotic and tonic seafood in oriental countries. Their high nutritional and functional food properties can combat several infectious and degenerative diseases. Atlantic SCs play an important role in Newfoundland and Labrador's economy. Live or fresh SCs are challenging to store and transport due to the presence of a vast quantity (>40%) of free water in their body cavity, autolysis properties, and the absence of the protective exoskeleton. As a result, more than 80% of SCs are processed to produce dehydrated products to prevent autolysis and microorganisms and facilitate their secure storage and easy transportation to the consumers. The nutritional and morphometric qualities of the dried products are affected by the handling, boiling treatment and drying technology used. The dehydration process also depends on the SC size, species, producing region and customer preference. Due to a lack of proper handling, storing, and drying, the product quality spoiled, resulting in a loss of revenue. SC body walls are transformed into cylindrical shapes by folding themselves longitudinally when they are boiled. Non-boiled ones become flattened upon drying in a hot air dryer. Upon boiling, SCs without flowers have curled better compared to those with flowers (Figure 1). The live SCs have energetic muscle bands and curled crosswise when those are slit to remove viscera. Some of them are so strong that they keep their cross curling even after boiling and drying. The drying parameters should be considered to develop a good quality SC product. The different drying processes for the Atlantic SCs, their pros and corn will be discussed.

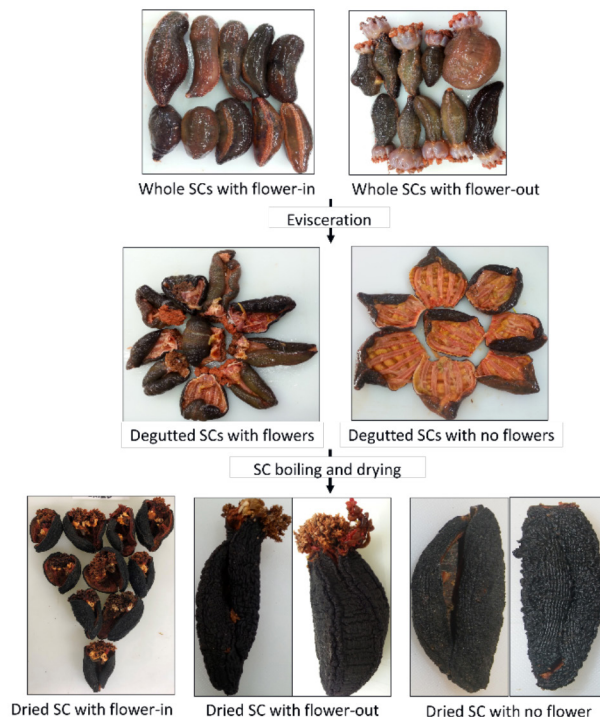


Figure 1. Drying steps for sea cucumbers (SCs) with and without flowers

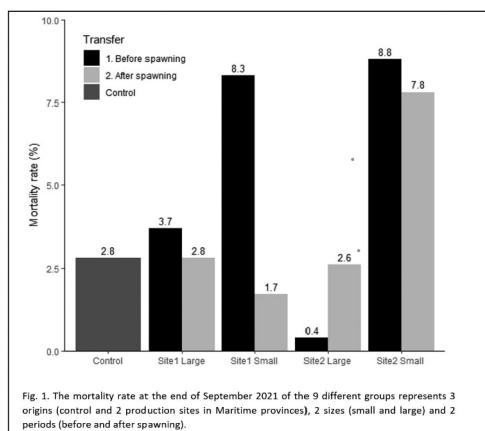
ANALYSIS OF STRESS ASSOCIATED TO INTER-SITES TRANSPORT ON THE AMERICAN OYSTER PRODUCTION

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The American oyster (*Crassostrea virginica*) benefits a value-added and well-established market in Canada. In Québec, most of the production is realized in the Magdalen Islands. The environmental conditions associated with climate change, particularly the warmer seawater observed in the lagoons stimulate the production of this tolerant species. The northern limit distribution of American oysters is observed in the Gulf of St. Lawrence in New Brunswick (47°N) explaining the natural absence of this species in Magdalen Islands. Thus, the production is supported by importation from the Maritime provinces since 2012, as natural reproduction does not support important recruitment success and spat collection in Magdalen Islands. However, since the start of this activity, episodes of mortality have been observed in the year following the transfer. This mortality seem not associated with pathogens, as no disease has been identified. We suggest that mortality is associated to transport and we try to develop physiological tools to quantify stress levels submitted for the development of best management practices.

Before and after spawning events in 2021, we received 300 large-sized oysters and 300 small-sized oysters from two production sites in Maritime provinces related to different transport distance. We used oysters implanted in Magdalen Island for two years after their interprovincial transfer as control. Thus, 9 oysters' groups constituted for 3 origins (control and 2 production sites), 2 sizes (small and large) and 2 periods (before and after spawning) were compared. To characterize the oyster condition and estimate their vitality level, different physiological indicators were used. Valvometry (the continuous measurement of the opening and closing valves), condition index and the level of resistance to air exposure were tested to estimate their vitality. Direct physiological measures of oxygen consumption and filtration rates were estimated, as well as the global accumulation of energy reserves (glycogen, lipids, and proteins) to assess their bioenergetic status and susceptibility to stress. All these indicators were related to the growth rate and mortality of each oyster groups from the beginning of the transfer until the end of the first wintering. Mortality and growth differences between oyster groups were observed with values from 0.4 to 8.8% mortality before the winter and growth twice more important in some groups. All this variability will be presented in relation to the viability and energetic status of oysters group following interprovincial transport.



INBREEDING AVOIDANCE AT THE GAMETE LEVEL IN ATLANTIC SALMON *Salmo salar*

Marco Graziano*, Monica Solberg, Martin Taylor, Kevin Glover, Anne Grete Eide Sørvik, David Murray
& Simone Immler & Matthew J.G. Gage

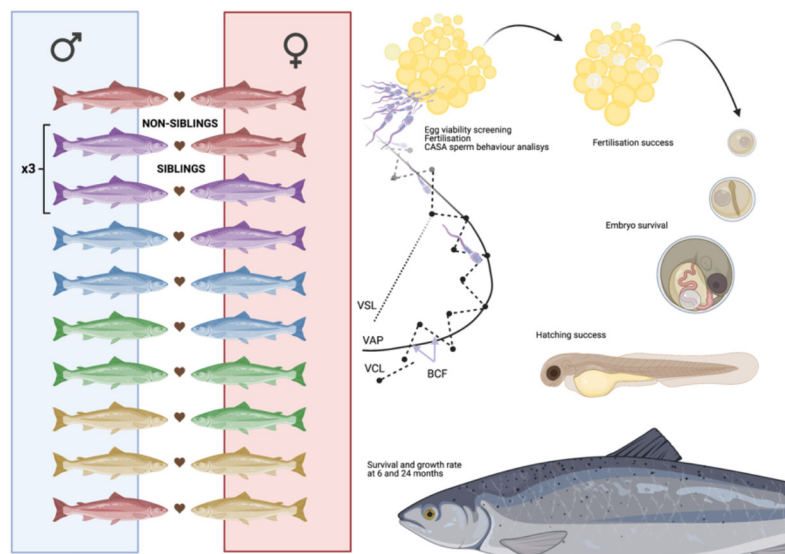
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Inbreeding can lead to fitness depression and reduce adaptive responses, and as consequence a range of mechanisms to limit reproduction between close genetic relatives have evolved. Here, we examine whether inbreeding avoidance exists between sperm and egg in Atlantic salmon, in a post-copulatory selection process where mechanisms are still to be unveiled. Philopatric salmon usually return to their natal streams to spawn, presenting a risk of breeding between genetic relatives, and reduced potential for females to control paternity under external fertilisation.

In a paired breeding design, we compared sperm motility parameters between sibling and non-sibling ovarian fluid, fertilisation and hatching success, growth rates and paternity outcomes following sperm competition trials between sibling and non-sibling males using microsatellites to assign paternity.

We found that sperm behaviour was significantly different in sibling vs. non-sibling reproductive fluid, with sperm activated in sibling fluid showing reduced motility and swimming trajectories coherent with a minor fertilisation potential. Males also suffered an average 18% reduction in fertilisation rates when crossed with eggs from sibling females, and sibling's offspring attained lower bodyweights and lengths before the onset of sexual maturation, but similar survival rates were observed despite the grade of relatedness. Our sperm competition trials revealed no difference in relative paternity success when simultaneously competing over the eggs of a sibling female and following this sibling hatched offspring showed consistently higher degrees of heterozygosity. We discuss possible reasons for this result in a context of balance between inbreeding avoidance mechanisms and preservation of local adaptation consistent with homing behaviour.

These findings indicate that post-copulatory inbreeding avoidance mechanisms have evolved at the gamete level in salmon, likely through interaction between ovarian fluid and sperm swimming, but also that sibling mates can equally contribute to overall paternity after proper 'filtering'. Our results find relevant applications in conservation aquaculture and in improving salmon farming sustainability due to the high degree of inbreeding reported in this sector and to the risk of detrimental genetic interactions in the wild as a result of farmed salmon escapes.



SELECTION OF MARINE WORMS FROM THE ST. LAWRENCE WITH POTENTIAL FOR PRODUCTION AND DEVELOPMENT

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Quebec mariculturists are dealing with a universal claim: the diversity of the species cultivated. One choice would be the produce and assessment of marine worms (polychaete annelids nereids), a process developing in other states. Although the management and gain potential remains to be established for Quebec, the establishment of a new production that matches one or more commercial needs is an original and promising avenue to explore. It was envisaged that the development of marine worms could answer to four industrial issues: check the breeding potential, improve the fish feed, reduce the waste management in saltwater production and produce molecules of interest for nutraceuticals and therapeutics. The research project thus desires to demonstrate the capacity of a development of marine worms and its assimilation into the field of marine productions.

The samples were dominated by two species of nereids: *Allitta virens* and *Hediste diversicolor*. The latter having the greatest biomass harvested; it was then selected for the suit of the experiment. The individuals were measured and weighed (avg. 0.3 g) and thus confined in tanks with sterilized natural sediment maintained at 16 ° C and a continuous seawater flow (29.5 ppt) of 2.0 L / min. Two types of feed were tested, TetraMin®, and fish rejects from the produce of spotted wolves (*Anarhichas minor*) in Merinov.

The 33-day experiment was carried out based on four tanks per analysis and the feeding rate adopted was 4% of individual weight / day. Overall, remarkably great survival rates were noted during the experiment, with an average rate of 96%, all methods combined. To fatten, the batches fed with TetraMin® showed much stronger results than those fed with fish debris, with on average conversion rates of 0.9 and 3.0 and weight increases of 20.3 and 6.3 g respectively. With the TetraMin®, the worms fed actively in the subsurface as soon as the flakes landed on the substrate. As for the feeding behavior of worms fed on fish waste, a significant drop in movement was noticed. Chemically, the diet seems to affect the percentage of fat in worms. Regarding proteins, a percentage of about 50% is observed, which makes worms an ingredient of choice in the feed preparation. Finally, *H. diversicolor* appears to be the species with the best returns in terms of growth, nutritional levels and pharmaceutical interest. It is then admirably fit for further in-depth investigations to be published.

OYSTERGRO – A PROFITABLE, TRUSTED AND PROVEN SYSTEM FOR OYSTER AQUACULTURE

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Delivering farmers, a ruggedly-engineered system designed for efficiency and high yield is what the OysterGro system is all about. Developed over the course of two decades, this trusted oyster farming method is now in service with hundreds of farms worldwide, growing high-quality, restaurant-ready oysters for clientele with discerning palettes who are looking for that exceptional oyster experience. A proven building block for coastal communities, this tried and tested method has built a reputation for delivering profitable results for business success, while at the same time working in constant harmony with Mother Nature.

Not only is oyster farming environmentally friendly, it actually has positive benefits as well. Oysters are considered nature's ecosystem engineers and have the reputation of transforming a water system's quality and clarity. One single adult oyster can filter up to 50 gallons of water a day. These bivalves have the ability to remove certain pollutants, such as nitrogen, restoring the water system to its natural state. In addition, OysterGro equipment has been engineered and designed to be "storm-ready" with industry-leading longevity to drastically reduce the possibility of marine debris.

The OysterGro aquafarming system is a trusted farming method and a proven performer, delivering convenience, strength and durability in even the harshest of weather conditions. It has been successfully tested and used around the world, in varying water depths and a multitude of climatic conditions.

The OysterGro method is a proven business model with predictable investment, performance and results. For farmers looking to run a viable aquaculture business with attractive profitability, the freedom to choose their working hours regardless of tides and the reliability they demand, the OysterGro method is the clear choice.



“PUMP FISH WITHOUT CROWDING” -TRANSFER FISH AT A HIGH RATE WHILE MAINTAINING LOW BIOMASS DENSITY

Joel Halse *

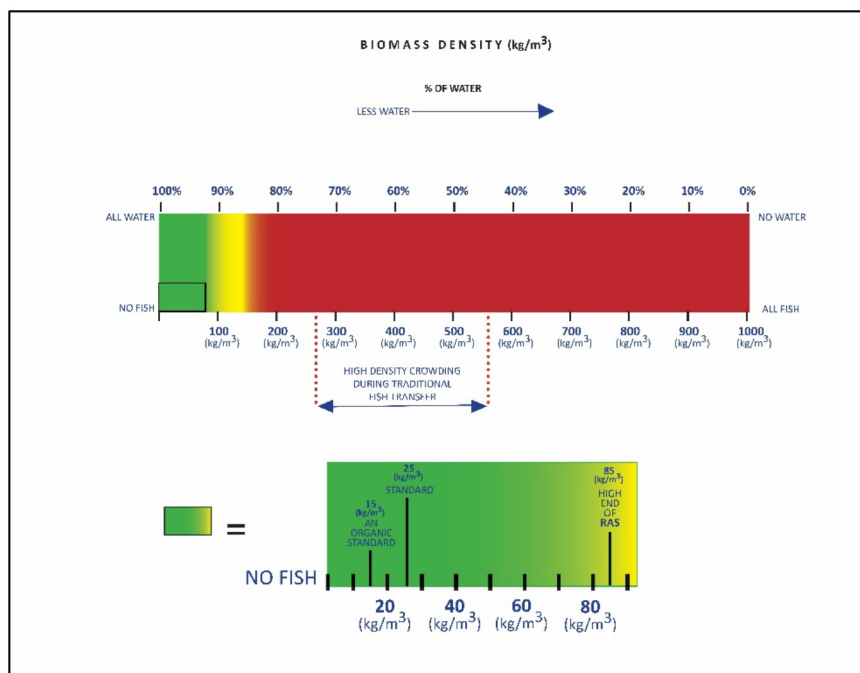
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Fish transfer worldwide is accomplished by bringing fish into a high-density situation before they can be sucked into a pump – and the more you crowd the fish, the better the transfer rate. This relationship is unsatisfactory because high density crowding is unhealthy for any population of fish. It creates stress, affects fish health and survivability and delays feeding post transfer.

During the farming process, we maintain biomass density (how much fish per unit volume of water) at a very low rate so there’s lots of water and not too many fish. There are very specific thresholds to maintain to keep the fish happy and healthy. The only time that we bring biomass density into an unsafe zone, one that is not sustainable and will eventually cause a negative outcome for the fish, is during fish transfer. The entire industry needs a solution where one can achieve a good transfer rate and keep the waiting fish population in a low biomass density situation – a safe zone – with lots of water for every animal that is waiting its turn to go through the fish pump.

We have designed a system that works with nature, that works with the fish’s natural behavioural instincts, that invites them to voluntarily participate in the transfer, to swim into the fish suction, instead of away from it.

The Voluntary Swim-In (VSI) technology is about better survivability, better health outcomes, and more feeding days. It will keep your fish happy, during and after the transfer, and help your bottom line.



INNOVATIVE CONTAINMENT TECHNOLOGY-NET PEN BIOFOULING CONTROL, THE ULTRASONIC METHODOLOGY

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Keeping subsea nets clean has been a challenge since the first fish farmer began operations. Cleaning can damage the aquatic environment as well as pose high costs for fish farmers. Current antifouling and net cleaning solutions may pose unnecessary risk and potential harm to fish health, general husbandry, and most importantly, the receiving environment. The focus is now on green technologies (new and proven) to reduce risk to the aquatic environment.

Using a first principles approach to biofouling, eliminating growth from starting on the subsea structure is the objective. Combining leading edge material science with proven ultrasonic technologies in the marine shipping/vessel industry is a new way forward for controlling growth on net pens. By transmitting ultrasonic through a specialty net fibre construction ultrasound produces a pattern of increasing and decreasing pressure on the surface of the net which causes a process called non-inertial cavitation. This causes microscopic bubbles to be created by the reduced pressure cycle implode as the pressure increases. The microscopic agitation has a cleansing effect which destroys surface algae. By disrupting this first link in the food chain, the surface is kept clean and makes it much less hospitable for larger organisms that feed on the algae.

Badinotti Combi Net combines traditional textile material blended with steel which provides additional inherent benefit of heightened Predator Protection. Raschel construction ensures 100% consistent and equal coverage of both materials and provides lower total weight and flat face as compared to heavier knotted nets. Badinotti Net provides premier conductivity as compared to knotted equivalent and has superior weight and breaking load ratio with the lowest weight fibre within the aquaculture industry. Consistent and stable high quality raw material supply chain of Dynema in all global markets indicates that cage control methods and technologies are on the verge of disrupting the market. The biggest impact is anticipated in the salmon farming market where fouled containment structures pose the greatest risks of fish health, environmental impact, cost, and social pressure.

Key Measurables and Observations:

<ul style="list-style-type: none">• 0% reduction in biofouling	<ul style="list-style-type: none">• Remote monitoring ability
<ul style="list-style-type: none">• 97.5 % reduction in energy consumption compared to biofouling removal	<ul style="list-style-type: none">• Integrates with current site infrastructure
<ul style="list-style-type: none">• No decomposition products in receiving environment	<ul style="list-style-type: none">• 40% reduction in biofouling management cost vs biofouling removal

SPECIES SENSITIVITY DISTRIBUTIONS FOR TWO WIDELY USED ANTI SEA-LICE CHEMOTHERAPEUTANTS IN THE SALMON INDUSTRY

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As part of the design of risk-based post-deposit monitoring to manage the usage of authorized aquaculture drugs and pesticides, applicable thresholds and/or environmental quality standards (EQS) need to be proposed. These values are useful for determining thresholds and/or guidelines depending on the selected regulatory usage. One approach for EQS determination is to use interspecies variation in toxicity of a compound to generate a species sensitivity distribution (SSD) for a given endpoint from which an HC_5 value (i.e. the hazardous concentration for 5% of species) can be extrapolated. Here, we construct and discuss aquatic SSDs for two widely used aquaculture anti-sea lice pesticides azamethiphos (Aza) and hydrogen peroxide. Literature was searched for available acute toxicity data (LC_{50} , EC_{50} , NOEC) for aquatic biota, and studies were assessed for quality and relevance. Data were further refined to include measurements per species (and life-stage where applicable). The focus of the study is on marine (SW) species however freshwater (FW) species were used to enhance sample sizes when sensitivities were not significantly different. Acute HC_5 values are proposed and discussed in term of limitations and applicability.

Table- HC_5 values determined by using a maximum likelihood-weighted averaging approach by fitting multiple distributions to the species sensitivity data and calculating a bootstrapped weighted average HC_5 estimate with asymmetric lower (LCL) and upper confidence limits (UCL).

Dataset		Ecosystem	Number of species	Species	HC_5 ($\mu\text{g/L}$ for Aza; mg/L for H_2O_2)	LCL ($\mu\text{g/L}$ for Aza; mg/L for H_2O_2)	UCL ($\mu\text{g/L}$ for Aza; mg/L for H_2O_2)
Aza	LC50	SW	20	1 algae; 1 annelid ; 10 crustaceans, 1 mollusc; 7 fish	0.40	0.09	2.94
	EC50	SW	11	1 bacteria; 4 crustaceans; 4 molluscs; 2 echinoderms	0.18	0.04	2.33
	NOEC /LOEC	SW	11	2 algae; 5 crustaceans; 3 molluscs; 1 fish	0.41	0.16	1.90
H_2O_2	LC50	SW & FW	39	1 insect; 2 annelids; 20 crustaceans; 1 mollusc; 15 fish	0.88	0.29	3.02
	EC50	SW & FW	21	4 bacteria; 6 algae; 8 crustaceans; 3 molluscs	0.77	0.42	1.83
	NOEC /LOEC	SW & FW	25	2 algae; 7 crustaceans; 1 mollusc; 15 fish	0.60	0.18	2.79

BIOECONOMIC ANALYSIS OF IN-POND RACEWAY SYSTEM GROWING STOCKER AND FOODSIZE HYBRID CATFISH PLUS TILAPIA

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In Pond Raceway systems consist of floating or fixed floor rectangular raceways (RW) assembled or placed into existing earthen ponds. This case study evaluated the performance and economics of producing foodsize hybrid Catfish and stocker sized fingerlings (Channel Catfish *Ictalurus punctatus* ♀ x Blue Catfish, *I. furcatus* ♂) in In-Pond Raceway Systems (IPRS) and tilapia (*Oreochromis niloticus*) grown in cages, placed into four 0.4 ha ponds. Growout raceways in ponds 1 and 2 were 63 m³, and 45 m³ in ponds 3 and 4. Each pond had one (14 m³) stocker unit raceway plus a 36 m³ tilapia cage placed into ponds 2 and 4. Each pond had 5.0 HP of aeration that maintained adequate DO levels. Catfish were fed a 32% CP commercial diet twice a day; tilapia were allowed only to graze the phytoplankton flow generated by the growout and stocker IPRS units. The combined production from growout (186 days) and stocker (142 days) resulted in 19,712 kg/ha in pond 1, 19,302 kg/ha in pond 2, 19,426 kg/ha in pond 3, and 16,555 kg/ha in pond 4. The same number of tilapia were stocked into the two cages and resulted in yields of 2,167 kg/ha (pond 2) and 2,160 kg/ha (pond 4); when combined with catfish production resulted in total gross production of 21,469 kg/ha in pond 2 and 18,715 kg/ha in pond 4. Foodsize average harvest weights ranged from 670 to 894 g, with survival rates ranging from 86 to 98 %, and having weight gain per day (WGD) of 1.74 to 3.22 g/day. Raceway stocker unit achieved harvest weights ranging from 173 g to 186 g, with survival rates ranging from 75 to 90% and having WGD from 1.02 to 1.37 g. In general, efficient FCRs were achieved in all growout and stocker hybrid Catfish IPRS units. Hybrid Catfish raised in IPRS demonstrated excellent fish inventory control, promoted uniform hybrid Catfish production, with 90 to 95% of the foodfish harvested in the preferred premium size range. Production strategies for inclusion of co-cultured tilapia along with the catfish IPRS systems were achieved with little investment and operating costs, resulting in overall positive net returns. Ponds housing IPRS catfish units plus a tilapia cage had reduced investment payback periods, increased net present value and higher internal rates of return (Table 1).

Table 1. Financial measures of profitability for producing foodsize and stocker hybrid Catfish, Channel Catfish *Ictalurus punctatus* ♀ x Blue Catfish, *I. furcatus* ♂, in In-Pond Raceway Systems (IPRS), and tilapia (*Oreochromis niloticus*) raised in cages (Ponds 2 and 4 only) placed immediately downstream of hybrid Catfish IPRS production cells in 0.4 ha ponds, U.S.\$, 2019.

Item	Pond 1	Pond 2	Pond 3	Pond 4
Financial net return, U.S.\$	-5,679	9,562	-4,090	2,298
Investment cost, U.S.\$	39,996	40,796	21,196	21,996
Payback period, year	-7.0	4.2	-2.5	4.9
Net present value, U.S.\$	-78,691	29,444	-38,823	7,024
Internal rate of return, %	-	16.5	-	20.7

*Using a discount rate of 5%.

EXPRESSION ANALYSIS OF THE IMMUNOMODULATORY GENE, CATENIN β UNDER MICROBIAL CHALLENGES IN *Amphiprion clarkii*

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Catenin beta or β catenin is a protein involved in important cellular functions such as regulation and coordination of cellular adhesions and gene transcription. It is a major protein function in the Wnt/ β catenin signaling pathway which plays a critical role in immune cell function. The current study has focused on identifying a particular sequence of β catenin from a transcriptomic database of *Amphiprion clarkii* analyzing its expression pattern in normal physiological conditions and under different immune challenges, to identify its participation in immune cell functions and modulating a particular immune response.

The sequence of *A. clarkii* was identified from the cDNA library followed by the *in-silico* analysis of the protein using appropriate bioinformatics tools and software. cDNA samples synthesized from twelve different tissues were subjected to quantitative real-time PCR in determining the tissue-specific expression of the gene. cDNA was synthesized from RNA purified from selected tissues at certain time points after challenging the fish with lipopolysaccharide (LPS), poly I:C, *Vibrio harveyi* and Phosphate buffered saline (PBS/control) and subjected to quantitative real-time PCR to determine the gene expression patterns under the microbial challenges.

The complete ORF of *A. clarkii* catenin β consists of 2307 bp encoding 768 amino acids. The highest expression of the gene could be observed in the head kidney followed by blood tissue of fish, see-through the upregulated expression of the gene in important immune organs of fish and thereby reveals its relativity in immune responses. According to the immune challenge data, stimulating fish with all the three immune challenges have upregulated catenin β expression in both the tested tissues. The viral analog Poly I:C has even a significant upregulation of the gene expression compared to other stimulants in early hours in both the tissues. These results may suggest the involvement of catenin β in the antiviral activity of the fish immune system, which will be validated further by other specific *in-vitro* antiviral assays.

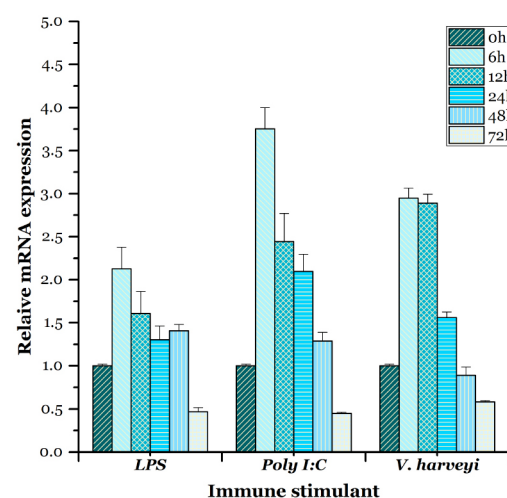


Fig.A. Relative mRNA expression of *A. clarkii* catenin β in blood after immune challenges

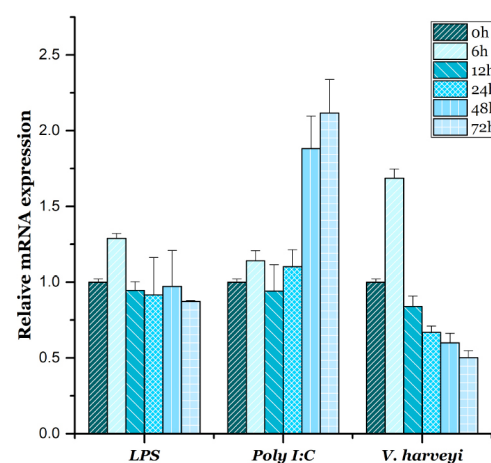


Fig.B. Relative mRNA expression of *A. clarkii* catenin β in head kidney after immune challenges

VALORIZATION OF SHELLFISH RESIDUES TO VALUE ADDED PRODUCTS FROM BIOACTIVES TO BIOMATERIALS

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The nature of shellfish processing residues is a function of the processing method, product, and season. The by-product composition varies as a function of the species, for instance shrimp residues are rich in antioxidants and chitin, while crab bodies are predominantly calcium carbonate. The diversity of the feedstock translates to a number of potential bioproducts. However, the processes to valorize these products from the residues are similar and only require small changes in operating conditions. In this presentation we will give an overview of the type of shellfish residues we are working with, characterization of residues, green processes we are using to produce the bioproducts. The focus of our work is to fully utilize the residues to bioproducts through development of processes that are environmentally sustainable and produce products that benefit the region and community.

MOLECULAR CHARACTERIZATION AND IMMUNE RESPONSE OF THIOREDOXIN DOMAIN-CONTAINING PROTEIN 17 FROM *Amphiprion clarkii*

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Thioredoxin domain-containing protein 17 (*Trdx17*) is one of the unique members of the thioredoxin (TRX)-like superfamily, consisted with TRX fold. *Trdx17* is a small (~14 kDa) cytosolic protein mainly involve in cellular homeostasis through the disulfide reductase activity and denitrosylation of nitrosylated proteins and thiols.

In this study, we have isolated the *Trdx17* from pre-constructed *Amphiprion clarkii* (*AcTrdx17*) database and characterized. The full-length nucleotide sequence of *AcTrdx17* contained 955bp and open reading frame consisted (ORF) with 372bp. Putative amino acid sequence contained 123aa and theoretical isoelectric point and molecular mass of the *AcTrdx17* were 5.75 and 14.1kDa respectively. ORF consisted with CPDC motif and TRX-related 14 family domain. Moreover, pairwise homology sequence analysis revealed that our gene had higher identity (99.2%) and similarity (100%) with *Acanthochromis polyacanthus*. Phylogenetic analysis revealed that *AcTrdx17* was clade with orthologs of various teleost species, suggested that could be diverged from common TRX ancestor. In addition, the antioxidant activity of *AcTrdx17* was analysed with recombinant protein by using the insulin disulphide reductase assay and findings confirmed the antioxidant activity of the *AcTrdx17*. In addition, tissue specific distribution of *AcTrdx17* in twelve different tissues (blood, head kidney, spleen, liver, gill, intestine, kidney, brain, muscle, skin, heart and stomach) of healthy fish were examined using quantitative Realtime PCR (qPCR) and highest expression was observed in brain, blood and head kidney tissues respectively. Moreover, *AcTrdx17* mRNA was analysed against the challenge fish group (*Vibrio harveyi*, Poly I:C and LPS) to analyse the immune response of the *AcTrdx17*. According to the transcriptional analysis, significant transcriptional modulation was observed in *Trdx17* against the bacteria and immune stimulants. Collectively, our findings suggested the putative role of *AcTrdx17* in anti-oxidant activity and may have defence responses against the invading pathogens.

CONTINUED GENETIC INTROGRESSION OF DOMESTIC ATLANTIC SALMON ALLELES FIVE YEARS AFTER A LARGE ESCAPE EVENT IN SOUTHERN NEWFOUNDLAND

Melissa Holborn*, Steve Duffy, Amber Messmer, Tony Kess, Brian Dempson, Marco Castellani, Brendan Wringe, Paul Bentzen, and Ian Bradbury

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The escape of domesticated Atlantic salmon from aquaculture facilities represents a significant genetic and demographic threat to wild salmon stocks, and throughout North America, escaped farm origin salmon have been shown to hybridize with wild conspecifics. In 2013, a large escape event of farmed Atlantic salmon occurred in southern Newfoundland and previous work has documented extensive hybridization with wild fish. However, the long-term genetic impacts of this and other escape events for wild salmon in the region are unknown. It is hypothesized that introgression may be fast-tracked by precocial maturation of male F1 hybrids, as these individuals would mature quickly and in greater numbers due to the absence of mortality at sea. In southern Newfoundland, high levels of precocial maturation in the wild may predispose salmon in the region to introgression due to interbreeding with escapees. In this study, we examined the evidence for long-term introgression in 19 southern Newfoundland rivers since 2013 and explored the role of various rates of precocial maturation in introgression by simulating genetic and demographic changes during escapee invasion.

Building on previous work, we estimated the proportions of different hybrid classes present in young-of-the-year parr sampled annually since 2014. We observed an increase in pure wild individuals over the time period sampled consistent with reductions in hybridization over time. Within the identified hybrids, the proportion of F1 individuals was initially high then decreased annually. The proportion of backcross wild hybrids increased over time, consistent with F1 hybrids successfully mating with wild fish. Simulations using IBSEM indicated rivers with high precocial maturation have greater genetic changes, greater population declines, and reduced recovery after cessation of escapee invasion. Overall, the results support the hypothesis that high rates of precocial maturation, such as those found in southern Newfoundland rivers, can fast-track genetic changes in wild populations due to escaped farmed Atlantic salmon.

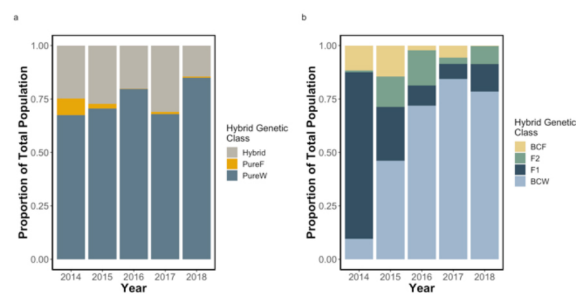


Figure 1 Total proportion of genetic classes for 19 rivers in southern Newfoundland over five years. Plot a shows proportions of pure wild, pure feral, and all hybrid categories combined. Plot b breaks down the combined hybrid category into backcross farmed/feral, F2, F1, and backcross wild.

INDRODUCING THE RASWAY – A VERY EFFICIENT SYSTEM THAT HAS LOW ENERGY, LOWER CONSTRUCTION COSTS ALL RESULTING IN A LOWER COP

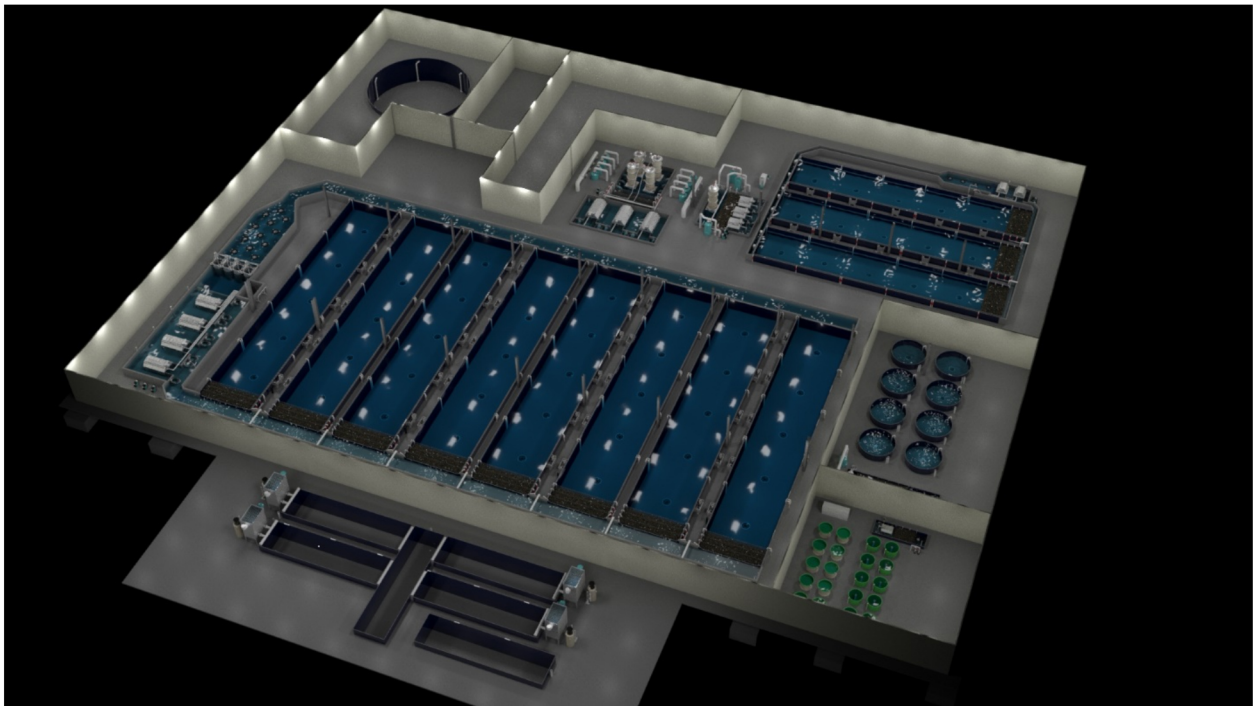
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The RASWay™ system is a very efficient way to produce fish in a RAS. The energy consumed is relatively low compared to other conventional systems for being a raceway system most of the equipment is on the same plain this reducing the cost of pumping water.

In this presentation it will be demonstrated how the farmer can keep his costs down to achieve a lower COP and thus a higher return on investment.

The RASWay™ can be used in all types of water – saltwater, brackish and freshwater as well as a large range of temperatures for 8°C to over 30°C. The species list is quite substantial as well.



In the presentation energy use will be discussed as well as fluid hydraulics on how the system works.

INNOVATING FOR SUCCESSFUL RAS OPERATIONS

May-Helen Holme

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RAS comes with a unique set of challenges, and a wholistic approach is needed to ensure problem free and profitable production. Great opportunities lie in development of new technical tools, digital applications and use of artificial intelligence and machine learning. In addition, large amount of data gathered from RC systems around the world give whole new possibilities for monitoring and predictions. BUT, there is a long way from data gathering to predictive models and improved methods that can be applied at the farmers. In this presentation we will dig more into some of the solutions that are on its way.

A main requirement for successful operation is consistent water quality. Many parameters play in, but high and consistent feed quality is a vital part. This means that feed producers must implement a whole new approach to physical quality and develop monitoring systems that ensures that excellent pellet quality can be guaranteed for every batch. This, together with feed formulations optimized for the specific farm conditions can mean a big difference to the biomass capacity and the farmers profitability. Dust and fat seepage must be kept to a minimum, digestibility of the pellet needs to be high and nutrient composition must be precise in order to minimize economic waste and build up of nutrients in the circulating water. Mass balance calculation and efficient waste handling is also farm specific and can be optimized through the feed and controlled through good digital models. This is why feed for RC systems need to be different and developed together with the users to meet each systems specific needs.

A good feed will only reach it's potential if the operation is running smoothly, and it is recommended to have a monitoring program for both fish health and pigmentation. A range of rapid methods are being developed to allow faster diagnostics and early correction, to minimize the impact on harvest quality. If needed, feed composition can be altered to fit the changing needs of the fish accommodating the different growth conditions. Again, this requires a strong collaboration between the farmers that knows the system and the fish, and the scientist and feed production specialists in the feed producing companies.

The consumer market is also developing, creating opportunities for farmers to differentiate their product. Within the space of novel ingredients and reduced footprint, many see an opportunity to improve the sustainability of their farming and meeting local and global ESG requirements. As the largest CO2 footprint in fish production comes from the feed, there is another incentive for the farmers and the producers to work closely to develop solutions in accordance with the farmers strategy and local governance. There is room for niche products, supply chain collaboration and footprint reduction that will benefit the whole aquaculture market. The feed producers need good and dedicated partners so we together can achieve progress.

REGULATORY FRAMEWORK CONDITIONS AND RISK MANAGEMENT IN NORWEGIAN FISH FARMING

Ingunn M. Holmen* and Trine Thorvaldsen

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Norway is the leading producer of Atlantic salmon worldwide. The development of the salmon fish farming industry has been profitable, but not free of safety challenges for fish, workers and the environment. The reputation of the industry is challenged by salmon escape incidents from net pens and the diseases they might carry are a threat to the wild salmon stocks.

Further growth of the produced biomass in Norwegian fish farming is at present coupled to the combat of a parasite called the “salmon louse”, through the so-called “traffic light system” introduced by the Norwegian government in 2017. Based on reports from each fish farm, the authorities monitor average louse levels in 13 geographical production regions for salmon and trout farming. Each year the Fisheries Directorate decides if growth in production will be allowed or not. Hence, development of management systems and technology for delousing when the levels are too high are prioritized by the companies. Preventing fish escapes is also paramount at the fish farms, and workers may challenge their own safety to prevent escapes. Since regulatory requirements instruct companies to manage lice levels and to implement measures to reduce the risk of fish escapes, the workers and operational managers might experience conflicting objectives from the point of view of their own health and safety.

The regulatory requirements for risk management in the Norwegian fish farming industry are supervised by five authorities. Risk assessments cover several risk dimensions, including fish health and welfare, fish escape prevention, reliability and safety of technical structures and vessels, external environment, food safety and OHS. It is required that they involve the workers at the fish farms and vessels in the hazard identification and risk analysis. Companies also aim to satisfy sustainability requirements and certification according to ASC, Global G.A.P and other international standards to ensure market access. Each national regulatory authority or NGO will conduct audits to check that the fish farmer’s management systems, risk assessments and operational practices comply with the part of the requirements for which they are responsible. However, in order for the fish farmers to optimize and rationalize their safety work, studies suggest that they should treat the different risk dimensions in a holistic manner. This presentation will give examples of how formal requirements are handled today and provide suggestions for improved practice at both regulatory and operational levels.

A REVIEW OF MATRIX EFFECTS DURING THE ENZYMATIC HYDROLYSIS OF FISHERIES BY-PRODUCTS

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By-products from fish processing plants can vary from 30-70 weight percent of landed material depending on the species and the product-dependent degree of processing. These by-products, taking the form of fish frames, viscera, and shell material, result in an environmental burden in terms of treatment and disposal, generating both high organic load liquid wastes and emission of greenhouse gases in degradation. However, these streams contain commodities such as industrial chemicals, as well as nutritional and medicinal compounds. Extraction of these compounds would not only decrease the environmental burden from processing but also provide a revenue stream. Thermochemical, biochemical, and mechanical processes can be used in product extraction. However enzymatic processes offer the advantage of preserving the integrity of the extract and do not require intensive processing conditions, which could include high temperatures, toxic chemicals etc. Proteolytic enzyme hydrolysis, which uses protein-digesting enzymes to catalyze the decomposition of substrate proteins into smaller, more soluble peptides, can be used as a treatment to separate proteins from lipids, carbohydrates, and minerals, or as a post treatment after lipids and/or bioactive chemicals are extracted. The end goal is the same, a protein rich hydrolysate, however the process route impacts the chemical matrix of the feedstock. These matrix effects are poorly understood, despite their vast importance in reaction kinetics and process scale-up and require further review and research.

In this presentation, the impact of feedstock properties on the enzyme hydrolysis reaction are discussed, along with observations and potential correlations between feedstock properties, effects of substrate pre-treatments, and the resultant kinetics and maximum degree of hydrolysis achieved in the reaction. Limitations of current research due to the use of destructive techniques in pre-treatment are then discussed, and non-destructive methodologies for study of matrix effects in the enzyme hydrolysis reaction based on results from past literature are introduced. Lastly, conclusions are made, providing an outlook for research in this field, including recommendations for future analysis.

A FULLY PHASED GENOME ASSEMBLY FOR *Mytilus edulis* UNVEILS A HIGH DEGREE OF PRESENCE-ABSENCE VARIANCE BETWEEN MUSSEL POPULATIONS

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Mussels belonging to the *Mytilus* species complex are cultivated worldwide, and Prince Edward Island produces 80% of the mussels sold in North America. Bivalve genomes are complex and contain many paralogous regions that can confound the separation of all types of variants in a genome. In addition, the mussel genome is highly repetitive and heterozygous.

To overcome the challenges imposed by these characteristics, we used a hybrid assembly approach combining PacBio CLR sequencing, Dovetail Omni-seq scaffolding, PacBio Hi-Fi sequencing and PacBio IsoSeq. We present a fully-phased chromosome level assembly of the mussel's genome that enabled the genome-wide evaluation of presence-absence variance in *Mytilus edulis*.

Length and contiguity metrics were: number of scaffolds = 347; N50 = 105 Mb, NG50 = 150 Mb, Total Length = 1.58 Gb. Quality Values and completeness generated using Merqury indicated that each haplotype individually only contains ~65% of the kmers present in the raw HiFi reads, but combined both haplotypes contain ~99% of the kmers present in the raw reads. Our haplotype collapsed assembly available on GenBank only contains 75% of the kmer present in the raw reads. That indicated that up to 25% of the polymorphism variation may be lost in a collapsed assembly.

BUSCO analysis shows that the primary assembly contains 98% of the eukaryotic conserved orthologs in odb10. PAV analysis indicates that up to 13% of shotgun reads from different individuals do not map to the reference genome. Ab-Initio annotation produced 41,319 gene models. Contigs from assembled unmapped reads contained 65,996 putative transcripts while the reference transcriptome generated with Isoseq clustering identified 216,434 putative transcripts, indicating that up to 30% of the mussel pan-transcriptome could be disposable.

In conclusion, we presented a road map to producing high-quality chromosome level phased assemblies for mussels. We also demonstrated the value of haplotype resolved assemblies for genomic analysis in blue mussels and showed evidence of significant PAV among different mussel individuals.

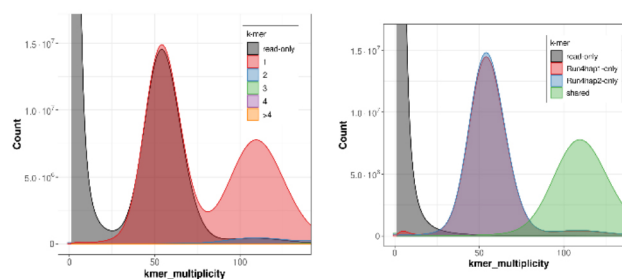


Figure 1: Kmer multiplicity in one haplotype (left) and both haplotypes (right)

INTERACTIONS BETWEEN AMERICAN LOBSTER (*Homarus americanus*) AND SALMONID AQUACULTURE IN THE CANADIAN MARITIMES

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Salmonid aquaculture occurs in coastal Atlantic waters around the Canadian Maritimes and can overlap with the American lobster fishery, the most profitable fishery in the region. There has been debate around whether there is potential for salmonid aquaculture to negatively affect the fishery that has been heated in both the scientific community and public news media. This review and resultant commentary explore the research approaches used in the Canadian Maritimes to examine known and inferred interactions between these two important industries. We re-examine some inferences of previous research and identify low oxygen environments and improper use of therapeutants as having the greatest potential to adversely affect lobster, although there are knowledge gaps. We further discuss the implications of whether localized lobster displacement from a farm area, would have any measurable impact on the lobster fishing industry as a whole, using examples from Nova Scotia, Canada. In most instances, existing regulatory compliance should be adequate to address the drivers which have the largest potential to adversely impact lobster near open net-pen salmonid aquaculture sites.

COMPARISON OF DRONE AND VESSEL-BASED COLLECTION OF MICROBIOLOGICAL WATER SAMPLES IN MARINE ENVIRONMENTS

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Many water quality metrics cannot be measured *in situ* and require collection of a physical sample for laboratory analysis. This includes microbiological samples for detection of faecal coliform bacteria in marine and freshwater systems which are a critical component of food safety programs for human consumption of bivalve shellfish worldwide. Water sample collection programs are typically vessel-based which can be time and resource intensive. In Canada, the Canadian Shellfish Sanitation Program (CSSP) aims to avoid consumption of contaminated molluscan bivalves by monitoring fecal coliform bacteria through vessel-based water sample collection. Uncrewed aerial vehicles or drones are becoming more commonly used for water sample collection given their relatively low cost but are rarely used to support microbiological analyses. A prerequisite for the acceptance of a new collection method for a regulatory program is to determine if a novel method of sample collection affects results. To assess this potential, we designed, developed, and tested a sampling device attached to the underside of a drone, to collect water samples for bacteriological analysis. Drone and vessel-based samples were collected in the same location, at 20 cm depth, within a minute apart, at ten geographic locations in coastal Nova Scotia waters, to compare fecal coliform counts. Bacterial count estimates obtained from drone-collected samples were not significantly different than estimates obtained from vessel-collected samples ($p < 0.5$). Results from this study suggest novel water sampling techniques using drones could supplement or replace traditional vessel-based sampling methods.

ROLE OF CspA PROTEINS IN *Aeromonas salmonicida* subsp. *salmonicida* in VIRULENCE AND PHYSIOLOGICAL PROCESS

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Cold Shock Proteins are known as DNA/RNA binding proteins, that play an important role in the regulation of the bacterial virulence and physiology process. In *Aeromonas salmonicida*, three cold shock proteins CspA, CspB, and CspD are found. Among these three cold shock proteins, CspB and CspD are DNA binding proteins and CspA is RNA binding protein. In this study, we evaluated the role of the RNA binding protein (CspA) on *A. salmonicida* growth, survival at low temperatures, physiology, and virulence in lumpfish (*Cyclopterus lumpus*). *A. salmonicida* *DcspA* mutant was constructed using suicide vector technology. *A. salmonicida* *DcspA* showed no differences in LPS, *vapA* modification after heat-shock, growth at 15°C and 28°C. However, a reduction in survival at 4°C and 0°C was observed in contrast to the wild type. Additionally, a delay mortality was noticed after injecting *A. salmonicida* *DcspA* in lumpfish in contrast to the wild type. To get an in-depth insight into the function of *DcspA*, mutant additional genome-wide transcriptome analysis was carried out. *A. salmonicida* *DcspA* showed 632 differential expressed genes (Log_2 fold-change 1; FDR_{pv} 0.05), including 413 upregulated genes, and 219 downregulated genes. In conclusion, we noticed that CspA plays a role in *A. salmonicida* survival at cold temperatures and influence virulence in lumpfish.

PHENOLIC COMPOUNDS AND THEIR BIOACTIVE PROPERTIES OF ATLANTIC SEA CUCUMBER PROCESSING DISCARD

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The global consumption of sea cucumber has increased rapidly due its impressive nutritional profiles with multiple medicinal properties. Sea cucumber contains numerous essential high-value nutraceuticals and pharmaceuticals, which show unique biological properties such as antithrombotic, anticoagulant, anticancer, anti-inflammatory, and antidiabetic activities. For example, orange-footed sea cucumber (*Cucumaria frondosa*) is abundant in the northwest Atlantic, which has been harvested in recent years for commercial purposes. This species is comprised of numerous compounds, namely lipids (PUFAs), proteins (collagen and peptides), vitamins, polysaccharides (chondroitin sulfate), saponins, and phenolics. The body wall is the major marketable portion (~50%) of this echinoderm. However, during the processing of sea cucumber, several visceral by-products are produced, including gonads, respiratory tracts, and intestines, that are ultimately discarded as waste. These discards represent up to 50% of the sea cucumber biomass and are a rich source of fatty acids and amino acids as well as bioactive compounds, mainly carotenoids and phenolics. Therefore, utilization of these discards is crucial in order to address both the environmental concerns and economic sustainability of sea cucumber industries.

The aim of this work was to examine the free, esterified, and insoluble-bound phenolics of sea cucumber processing discards for the first time in any species of sea cucumber. The contents of total phenolics and flavonoids were determined, and antioxidant potential, including ABTS (2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid), DPPH (2,2-diphenyl-1-picrylhydrazyl), and hydroxyl radical scavenging activities, was assessed. The metal chelating ability of different phenolic fractions was also monitored. Furthermore, bioactivities such as LDL-cholesterol oxidation inhibition, inhibition of peroxyl and hydroxyl radical-induced supercoiled DNA strand scission, anti-tyrosinase activity, anti-glycation activity, and α -glucosidase inhibitory activity were investigated. Moreover, the antioxidant activity of the phenolic extracts was also evaluated in a fish model system using thiobarbituric acid reactive substance (TBARS) assay.

It was found that the sea cucumber processing discards are a good source of phenolic compounds. The highest amount of phenolics, antioxidant activity, and bioactivity was observed in the free phenolic fraction. Moreover, phenolic extracts improved the shelf-life of Atlantic salmon up to 9 days during refrigerated storage by inhibiting lipid oxidation. A strong positive correlation was found between total phenolics and various *in vitro* antioxidant assays, suggesting that sea cucumber phenolics showed antioxidant activity. Additionally, 18, 14, and 16 compounds were identified using UHPLC-QTOF/MS-MS from the free, esterified, and insoluble-bound fractions, respectively. Phenolic acids, mainly *p*-coumaric acid, protocatechuic acid, hydroxygallic acid, and chlorogenic acid, and flavonoids, including quercetin and catechin, were the major compounds found in the free phenolic fraction. Therefore, sea cucumber processing discards may serve as a viable source of functional food ingredients with protective antioxidant properties.

AQUACULTURE AND EELGRASS (*Zostera marina*) INTERACTIONS IN TEMPERATE ECOSYSTEMS

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As the global aquaculture industry expands, there is more potential for it to overlap and interact with eelgrass (*Zostera marina*) in coastal waters. Shellfish aquaculture can have positive, neutral, and negative effects on eelgrass. Positive interactions can be generated by the filtering activity of cultured bivalves, which may improve water quality and reduce epiphyte loads. Shellfish faeces and pseudofaeces may also provide more nutrients to eelgrass, thereby enhancing growth. However, such positive interactions have only been observed within an aquaculture setting a handful of times. Negative interactions are more common and can be a result of shading and sedimentation. Generally, these negative effects tend to occur directly under and immediately surrounding shellfish farms, and rapidly diminish with increasing distance.

In contrast, only one field study has investigated the effects of finfish aquaculture on eelgrass in a temperate setting, and the results were inconclusive. Nonetheless, many studies have investigated the effects of Mediterranean finfish farms on two other species of seagrass (*Posidonia oceanica* and *Cymodocea nodosa*). These studies report clear and negative interactions. However, it is unknown if these studies are relevant for temperate regions due to differences in environmental conditions and the seagrass species being investigated. Thus, further study in a temperate setting is clearly warranted.

We conclude by highlighting key research gaps, and go on to make some general management recommendations that may help minimize potential negative interactions between aquaculture and eelgrass in temperate ecosystems.

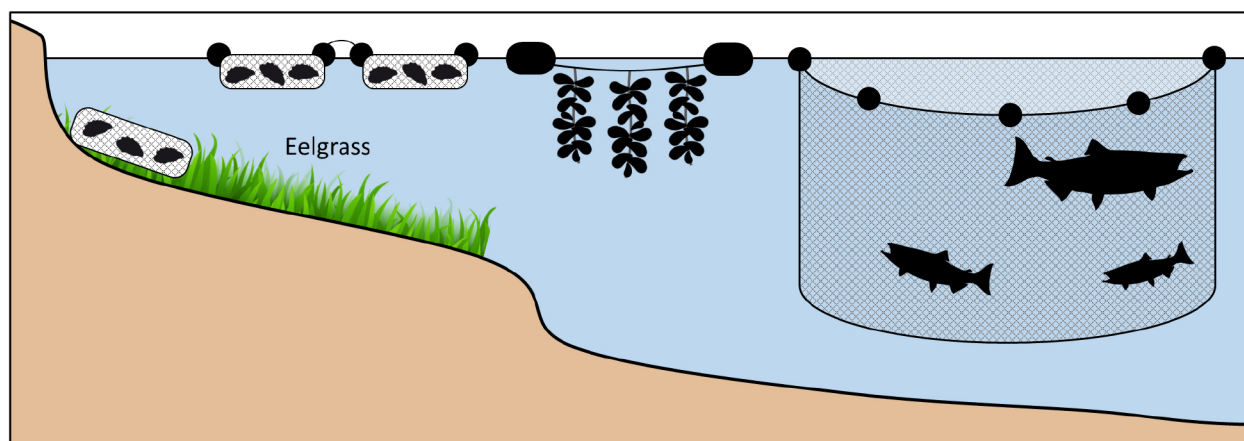


Figure (above). Some forms of aquaculture (e.g. intertidal shellfish aquaculture) have greater potential to overlap and directly interact with eelgrass.

THE POTENTIAL OPPORTUNITIES AND BARRIERS FACING OFFSHORE AQUACULTURE IN ATLANTIC CANADA

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The development of offshore aquaculture is rapidly gaining momentum in many countries around the world. This is because offshore aquaculture may potentially provide higher production levels while avoiding some of the environmental issues and conflicts currently facing the coastal aquaculture industry. In this presentation, we review the current global status of offshore aquaculture, its potential opportunities for expansion, and its potential barriers.

The term ‘offshore aquaculture’ currently lacks a standard definition and is used to describe a variety of operations in a wide range of depths and distances from the coast. When experimental offshore farms are excluded, there are only 10 operations around the globe that are in full commercial production and located further than 3 km from the coast (Figure 1).

Irrespective of the technological challenges of offshore commercial aquaculture, we find that regulatory uncertainty is likely a major barrier to the development and expansion of offshore aquaculture in several countries around the world. This is particularly true for countries like Canada and the USA, where there is ongoing: development of federal offshore aquaculture policies; confusion between multiple Federal and State / Provincial Governments; and ambiguity in jurisdictional borders. Such regulatory uncertainty may act as a deterrent to any investors considering developing offshore aquaculture in these countries.

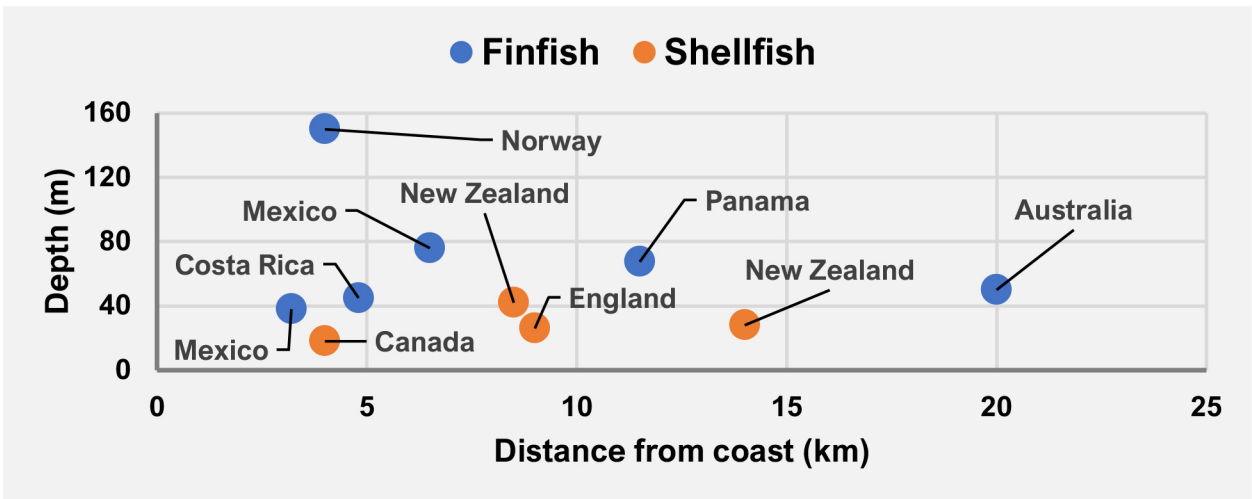


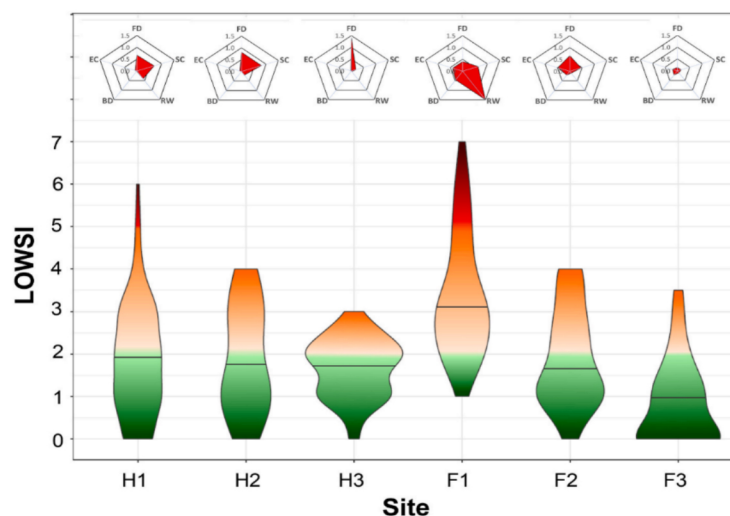
Figure 1. The depths, distance, and country of offshore aquaculture facilities (> 3 km from the coast) that are currently in full commercial operation. Colour of dots represent whether they produce finfish or shellfish.

DEVELOPMENT, VALIDATION AND TESTING OF AN OPERATIONAL WELFARE SCORE INDEX FOR FARMED LUMPFISH *Cyclopterus lumpus* L

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Lumpfish (*Cyclopterus lumpus* L.) are widely used for controlling sea lice in salmon farming, but their welfare is often challenged by poor husbandry, stress, and disease outbreaks, which compromise their ability to delouse salmon and cause public concern. It is hence important to identify when the welfare of lumpfish is being compromised in a simple and effective manner so that remedial actions can be taken. We developed, validated and tested a Lumpfish Operational Welfare Score Index (LOWSI) based on a visual assessment of skin and fin damage, eye condition, sucker deformities and relative weight, operational welfare indicators that fish farmers considered to be the most informative and were validated against cortisol measurements. We also present percentile length-weight charts to enable fish farmers to detect underweight and emaciated lumpfish at different stages of development. The lumpfish welfare score index was quick and easy to score and was highly repeatable (intra class correlation coefficient = 0.83 ± 0.05). Most lumpfish (71%) displayed good welfare, but significant differences were found between six commercial sites and 28% of lumpfish had lower than normal weights for their length, and 10% were emaciated. The most common welfare problems were sucker deformities and fin damage in hatcheries, and poor eye condition and body damage in sea cages, conditions that may increase the risk of emaciation. Being able to score the welfare of lumpfish quickly and accurately will help improve their welfare, reduce stress-related mortalities, and improve the sustainability of the salmon farming industry.



Variation in the Lumpfish Operational Welfare Score Index (LOWSI) at six commercial sites (three hatcheries – H1-H3, and three sea farms, F1- F3; n = 245). The proportion of fish falling into each of the three welfare classes (A-C) is shown by the violin plots in different colours (class A, green: welfare unlikely to be compromised; class B, orange: moderately compromised welfare; class C, red: poor welfare).

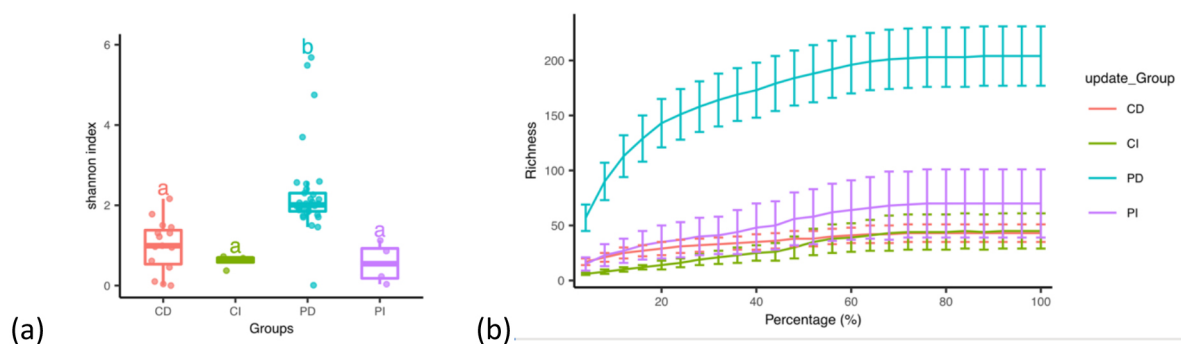
EFFECTS OF A MULTI-STRAIN PROBIOTIC (PRIMALAC®) ON THE GUT MICROBIOME OF THE MARINE FISH RED DRUM, *Sciaenops ocellatus*

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The gut microbiome of the commercially important marine fish, red drum (*Sciaenops ocellatus*), has important implications for commercial and recreational fishing, as well as aquaculture, because of the importance of the microbiome on host health and disease susceptibility. In aquaculture, while the aquatic environment provides a continuous source of microbes to the host along with the feed, supplementation in the form of prebiotics and/or probiotics, is becoming more popular. Yet little is known about the effects of probiotic supplementation on the native species richness and/or diversity of the gut microbial communities, and their importance to fish health. To begin to fill this gap in knowledge, we raised juvenile red drum for 56 days in three treatments: 1) control (no probiotics), 2) a water-soluble multi-strain probiotic (PrimaLac®, (*Lactobacillus acidophilus*, *Lactobacillus casei*, *Bifidobacterium bifidum*, and *Enterococcus faecium*) was added to tank water and, 3) PrimaLac® was commercially added to the feed in an enhanced starter feed (PESF). Following harvesting the intestine for histology and weighing the digesta, the intestine (autochthonous bacteria) and digesta (allochthonous bacteria) were profiled for microbial composition using 16S rRNA sequencing. Bioinformatic analysis included calculation of alpha diversity using the Shannon index, which measures the number of species and relative species abundances, and beta diversity using the Jaccard distance, which accounts of presence and absence of species. Preliminary results revealed robust taxonomic shifts in the Shannon index and richness of the gut microbiome in the probiotic treatments compared to controls (Fig. 1a, b). Interestingly, in the probiotic treatments, microbial species diversity and richness was significantly higher in the digesta than in the intestine. For controls, no differences existed between digesta and intestine. In addition, species diversity and richness of the intestinal samples from the probiotic treatment did not differ from those of either the control digesta, or the intestine (Fig. 1a, b). Further analysis will determine the relative taxonomic identity, as well as the presence or absence of pathogenic bacteria, among treatments and microbial sources (digesta vs intestine) and will provide valuable data of the value of probiotic supplementation for improving the quality of health for this fish species in aquaculture settings.

Figure 1. Shannon index (a) and species richness (b) for juvenile red drum raised under control and probiotic treatments for 56 days. control intestine (CI), control digesta (CD), probiotic intestine (PI) and probiotic (PD).



DIETARY EFFECTS ON THE GUT MICROBIOME OF SALMONID FISHES: A REVIEW

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In the past decade, there has been an explosion of microbiome studies focused on the fish gut using next-generation sequencing (NGS) methods. Advances in the efficiency and affordability of DNA sequencing technologies have made it possible for researchers to identify over a thousand-fold higher number of microbes that would not be seen with previous culture dependent methods. There has been a particularly high number of NGS studies on the gut microbiome of farmed salmonid species, such as rainbow trout and Atlantic salmon, due to their importance to the aquaculture industry. Many studies have characterized the gut microbiome and have found certain bacteria phyla that dominate, such as Firmicutes, Proteobacteria and Actinobacteria, which commonly include beneficial bacteria *Lactobacillus*, *Streptococcus* and *Corynebacterium* spp. These bacteria and the larger community in the gut have been considered to be an “additional organ” due to their importance and ability to break down dietary fibre, produce short-chain fatty acids, synthesize vitamins, antagonize pathogens and interact with the immune system. Research is lacking on characterizing the gut microbiome of less commonly farmed salmonids, such as Arctic charr and coho salmon, although this may change in the future as they become more valuable to the industry. In addition, we are understanding more and more about environmental effects, such as temperature and rearing system, on the communities of gut bacteria in farmed fish.

There has been a recent shift from mapping the bacteria present in the gut of farmed fish to the specific affect of diet on these microbial communities, especially alternative protein and lipid sources used to replace the unsustainable inclusion of fishmeal and fish oil in the diet. Increased inclusion of plants (e.g. soy, corn and wheat), microbes (e.g. yeast, bacteria and microalgae) and insects (e.g. black soldier fly) have been found to influence the gut microbiome with dramatic changes at both the lower genus level and higher phyla level (Firmicutes:Proteobacteria ratio). Lower dietary inclusions or supplements of probiotics, prebiotics, organic acids and essential oils have also been found to modify the diversity and composition of the gut microbiome of farmed salmonids, albeit at a lower level (specific order or genus). I will present recent findings from several NGS studies that investigated dietary effects on the gut microbiome of salmonid fishes at the University of Guelph (Guelph, Canada), University of Stirling (Stirling, UK) and Swedish University of Agricultural Sciences (Uppsala, Sweden). The effects of these functional feed ingredients and increased replacement of fishmeal and fish oil with plants, microbes and insects on the gut microbiome will need to be optimized in the future to improve the nutrition, health and welfare of farmed salmonids and maintain the growth of the aquaculture industry.

REDUCING THE USE AND IMPACT OF ANTIBIOTICS IN AQUACULTURE. THE COLLABORATIVE EXPERIENCE OF THE CHILEAN SALMON INDUSTRY

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With exports of over 550,000 tons in 2018, salmon farming in Chile is one of the most important economic activities, accounting for 7% of the country's total exports. The rapid growth of the industry during the 1980s allowed the consolidation in international markets, with Chile becoming the world's second-largest supplier of farmed salmon.

Despite continuous remodeling and reorganization of the salmon production system, during and after the pandemic ISA virus crisis, health and environmental challenges remain. The bacterial disease, Salmon rickettsia septicemia (SRS), results in annual losses, environmental impacts, and market externalities. Given the complex biology of the bacteria, there are few preventive measures and tools to control outbreaks. The primary tool for the control of an SRS outbreak is antibiotic treatment.

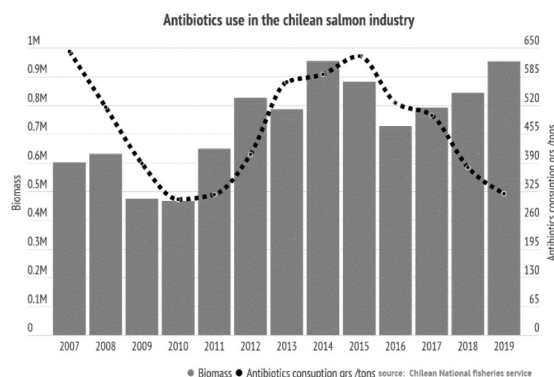
Several complexities exist regarding antibiotic use in aquaculture, including, efficient use, logistics, and operational use. In addition, concerns and uncertainties exist about antibiotic impacts on the water column and seabed, despite its heavily regulated use in Chilean

During 2019, SalmonChile and the Monterey Bay Aquarium Seafood Watch (SFW) signed a collaboration memorandum to improve the environmental sustainability of the Chilean salmon industry. One of the key objectives of this partnership is a 50% reduction or antibiotic use by 2025. This program was launched as the Chilean salmon antibiotic reduction program (CSARP).

As a result, the program has identified indicators, recommendations, and research gaps to meet the objective of the program. In addition, the programs technical advisors have identified specific metrics and actions for monitoring, reporting, and transparency of the process.

CSARP is one of the most important private and voluntary initiatives for reducing the use of antibiotics in animal production, including aquaculture. The program members include 16 salmon farming companies, equivalent to 100% of Atlantic salmon production, the Chilean salmon farmer association, the Chilean salmon marketing council, and the Monterey Bay Aquarium Seafood Watch Program.

Fig.1: Antibiotics consumption (g/tons) 2007-2019 periods.



TEMPERATURE-MEDIATED EFFECTS ON THE HEPATIC TRANSCRIPTOME OF GROWTH HORMONE TRANSGENIC FEMALE TRIPLOID ATLANTIC SALMON (*Salmo salar*)

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This study examined the impact of rearing temperature (10.5, 13.5 or 16.5 °C) on the hepatic transcriptome of AquAdvantage Salmon (growth hormone transgenic female triploid Atlantic salmon) at an average weight of 800 g. Six stranded PE libraries were Illumina-sequenced from each temperature group, resulting in an average of over 100M raw reads per individual fish. RNA-sequencing (RNA-seq) results showed the greatest difference in the number of differentially expressed transcripts (1750 DETs), as revealed by both DESeq2 and edgeR ($q < 0.05$; fold-change > 1.5), was between the 10.5 and 16.5 °C temperature groups. In contrast, 172 and 52 DETs were found in the 10.5 vs. 13.5 °C and the 13.5 vs. 16.5 °C comparisons, respectively. Considering the DETs between the 10.5 and 16.5 °C groups, 282 enriched gene ontology (GO) terms were identified ($q < 0.05$), including 'response to stress', 'immune system process', 'lipid metabolic process', 'oxidation-reduction process' and 'cholesterol metabolic process', suggesting elevated temperature elicited broad effects on multiple biological systems. Pathway analysis using ClueGO showed additional impacts on amino acid and lipid metabolism. There was a significant positive correlation between RNA-seq and real-time quantitative polymerase chain reaction (RT-qPCR) results for 8 of 9 metabolic-related transcripts tested. RT-qPCR results also correlated to changes in fillet tissue composition previously reported in these salmon (e.g., methionine and lysine concentrations positively correlated with *hsp90ab1* transcript expression), suggesting that rearing temperature played a significant role in mediating metabolic/biosynthetic pathways of AquAdvantage Salmon. Many transcripts related to lipid/fatty acid metabolism (e.g., *elovl2*, *fabpi*, *hacd2*, *mgll*, *s27a2*, *thrsp*) were downregulated at 16.5 °C compared to both other temperature groups. Additionally, enrichment of stress-, apoptosis- and catabolism-relevant GO terms at 16.5 °C suggests that this temperature may not be ideal for commercial production when using freshwater recirculating aquaculture systems (RAS). This study relates phenotypic responses to transcript-specific findings and therefore aids in the determination of an optimal rearing temperature for AquAdvantage Salmon.

IMPACT OF TEMPERATURE AND SUPPLEMENTAL DIETARY CHOLESTEROL ON TISSUE LIPID COMPOSITION AND HEPATIC TRANSCRIPT EXPRESSION OF FEMALE TRIPLOID ATLANTIC SALMON

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With rising ocean temperatures due to climate change, the Atlantic salmon aquaculture industry must adapt farming practices to mitigate risk and maintain product quality. In this study, female triploid Atlantic salmon initially acclimated to 12°C were subjected to an incremental thermal challenge, wherein temperature was raised by 0.2°C day⁻¹ to mimic summer sea-cage culture conditions; with temperature held at 16 and 18°C for several weeks to examine the effects of prolonged exposure to elevated temperatures. From 16°C onwards, the fish were fed either a control diet, or one of two nutritionally equivalent diets containing supplemental cholesterol [+1.30%, experimental diet #1 (ED1); or +1.76%, experimental diet #2 (ED2)]. Liver samples from a subset of fish were collected following an initial two weeks spent at 12°C, as the temperature reached 16°C, and at 18°C after the salmon had spent 65 days at elevated temperatures [i.e., 3 weeks at 16°C, followed by an increase at 0.2°C day⁻¹ to 18°C (10 days), then 5 weeks at 18°C].

Temperature significantly impacted the level (mg g⁻¹ wet weight) of several lipid classes (e.g., triacylglycerols, sterols, phospholipids) and fatty acids [e.g., arachidonic acid (ARA), eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA)] in the liver of fish fed the control diet at 12, 16 and 18°C. With the exception of sterols, the above lipids/fatty acids were lower at 18°C compared with at 12 and/or 16°C. Interestingly, at 18°C phospholipid concentrations were higher, and DHA and omega-3 fatty acids (i.e., ω 3) were greater in salmon fed ED2 as compared to the control diet. Transcript levels of 13 cholesterol/lipid metabolism-relevant genes were also measured using real-time qPCR. The expression of several transcripts (i.e., *apoA1a*, *hmgcr*, *srebp2*, *elovl2*, *helz2a*) was downregulated in the control group at 18°C compared to at 12 and/or 16°C. However, dietary treatment did not affect the expression of these transcripts at 18°C. Although analysis of fillet lipid/fatty acid composition is still ongoing, the present results indicate that temperature plays a greater role in regulating lipid metabolism than dietary cholesterol level.

Ultimately, this research shows that chronic exposure to high temperatures has consequences on the production of female triploid Atlantic salmon, as shown by the dysregulation of metabolic pathways and changes in lipid composition. Future studies should examine whether other diet formulations / functional feed ingredients can more effectively mitigate the potentially detrimental effects of high temperatures.

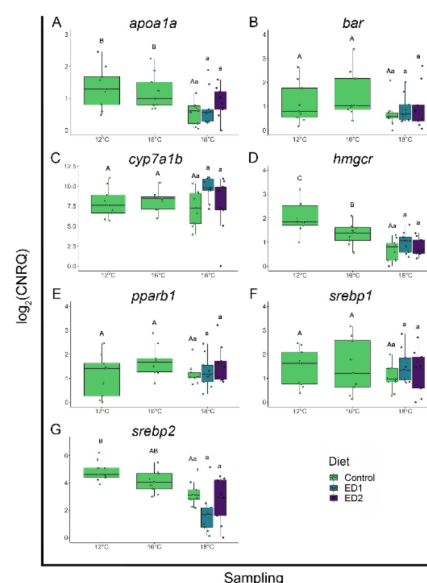


Figure 1. Expression level of 7 transcripts with roles in cholesterol metabolism.

DEVELOPING eDNA AS TOOL FOR MONITORING CULTURED AND WILD POPULATIONS OF SEA SCALLOPS *Placopecten magellanicus* IN MAINE, USA

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The Maine eDNA program aims to use molecular tools to better understand and promote sustainable use of wild and cultured resources. Here I will talk about two applications of eDNA tools for evaluating sea scallop aquaculture efforts along the coast of Maine, in the field, farm, and lab. One goal is to groundtruth eDNA assays for use in aquaculture in hatchery and field conditions. Quantitative eDNA assays for sea scallops have been developed and calibrated for sperm and dockside conditions, but this assay has not been calibrated for eggs or larvae, nor has it been tested in the field over wild beds and on functioning aquaculture farms. Here we explore whether eDNA from adults vs gametes and larvae can be distinguished by sampling at different depths and points in time in cultured and wild populations of sea scallops along the coast of Maine. In concert with traditional techniques like gonadal somatic indices (GSIs) (Figure 1), water samples were collected at three scallop aquaculture farms and three sites distant from farms in Penobscot Bay, ME, to assess the relationship between eDNA and spawning over time and magnitude.

In our next application we’re exploring how microbiomes (collections of microbes associated with scallops) vary over life stage in wild and cultured scallops and how this might influence hatchery success. Microbiomes of wild veligers from Southern Maine and cultured veligers from the Downeast Institute were surveyed using 16S metabarcoding and compared to microbiomes from tank water and biofilms. Bacterial communities from these different sources were compared to identify potentially pathogenic versus healthy bacteria to better understand the high failure rate of hatchery-reared scallop larvae (Figure 2).

Both of these studies highlight how eDNA can be applied to improve aquaculture development, operations and management. We’ll discuss the results of our studies and also the general process of eDNA assay development and processing for commercial applications.

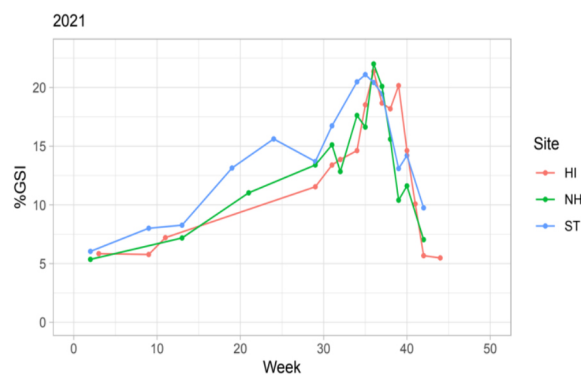


Figure 1: GSI monitoring in concert with eDNA sampling efforts.

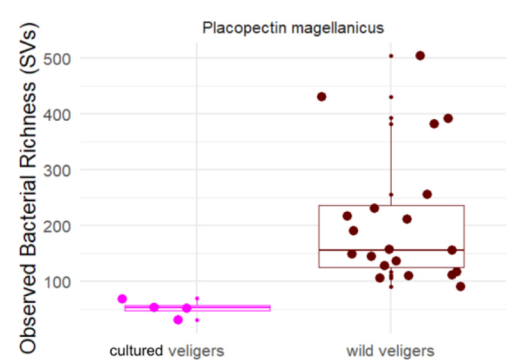


Figure 2: Bacterial richness in cultured and wild veliger samples

AN INTERNATIONAL OVERVIEW OF CURRENT WORLD SEA CUCUMBER RESOURCES AND UTILISATION

Guangling Jiao*

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Sea cucumber is a well-known traditional medicine and luxury nourishing food in Asian countries for thousands of years, and has recently gained increasing interests in western countries as well. The demand of commercial sea cucumber is growing in worldwide. This presentation overviews on the holothurian resources of the main sea cucumber origins in the Pacific and the Atlantic Oceans, focusing on the sustainability, processing and quality assurance, and principal markets for the sea cucumber products. It reviews the current status and issues raised of the global sea cucumber resources management and utilization. At last, several recommendations are designed to help build a sustainable and competitive sea cucumber industry.

BIOFOULING GROWTH ON ATLANTIC SALMON *Salmo salar* FARM NETS: EXPLORING LINKS WITH ENVIRONMENTAL FACTORS AND FISH HEALTH

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Farm-raised salmon is the highest valued seafood product in British Columbia (BC) and is the province's top agricultural export. In 2019, the salmon farming industry generated over \$1.6 billion in total revenue and supported over 6,000 jobs. Recently, however, salmon aquaculture companies in BC have experienced fish mortalities due to gill disorders and mouth lesions that have cost the companies substantial revenue. Many potential causes such as viruses, bacteria, parasites, and harmful-algae blooms have been examined extensively and ruled out as causative agents. The present hypothesis is that the removal of biofouling communities on the salmon nets via power-washing creates suspended particulate matter that impacts fish-gill health. Hydrozoans, with their stinging cells, are the most likely problematic species. They have been shown to cause gill disorders and mouth lesions in previous European laboratory-based studies and are known to occur in biofouling communities at BC salmon farms.

This study examined the development of biofouling communities on nets at two commercial Atlantic salmon (*Salmo salar*) farms on Vancouver Island (BC), with a focus on identification and quantification of the various species. Five 30 x 30-cm fish-net panels were deployed at each of five depths (1, 5, 10, 15, and 20 m), totalling 25 panels per site. These panels were collected bi-weekly, with fresh panels being re-deployed, for approximately six months. Their collection occurred just before commercial net cleaning. Species growing on the nets were identified and quantified, with an emphasis on hydrozoan identification. Key environmental parameters (e.g. temperature, salinity, dissolved oxygen, pH, nitrates, ammonia, phosphates, iron, and silica) were measured daily. The gill health of fish contained within the study pens was monitored weekly for the duration of the study. The relationship between the environmental parameters, the biofouling community, and fish-gill health will be examined by various correlative statistical techniques.

Results show seasonal/site patterns of the environmental parameters, seasonal/site/depth variations in the biomass of various species in the biofouling community, and seasonal trends in individual gill scores of the fish sampled. Specific results will be discussed. These data will be used to explore interactions between the environmental variables and the biofouling community and between the biofouling community and gill health.

MOOSE CREE FIRST NATION AQUAPONICS

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The Moose Cree First Nation is developing a commercial aquaculture and hydroponics venture to generate meaningful employment and prosperity in the community. Located immediately south of James Bay in Northern Ontario, the geographic isolation of the community results in an extraordinarily high cost-of-living. Fresh fish and vegetables are particularly expensive.

An opportunity to develop an integrated aquaculture and hydroponics operation (i.e. aquaponics) for the production of rainbow trout and leafy vegetables (e.g. leaf and romaine lettuces, basil, kale) has been developed. The facility will yield 12 tonnes of rainbow trout and approximately 65,000 heads of leafy greens per year.

The initiative offers numerous benefits to the community, including food security, more affordable fresh animal protein and vegetables, employment and sustainable economic development. The initiative also offers a unique opportunity for youth engagement. Development of local expertise in aquaculture, hydroponics and aquaponics is an essential aspect of this initiative. As part of a comprehensive training program, a demonstration system was installed to provide hands-on learning and skills development. The demonstration unit has a capacity to produce 340 kg of trout as well as 3,500 heads of vegetables per year. The design and costs of the demonstration system and the full-scale facility will be presented.

FEED A GROWING WORLD POPULATION AND TREAT ANIMALS WITH RESPECT: AN OVERVIEW OF FARMED SALMON WELFARE REGULATIONS AND BEST PRACTICES, AND HOW THE INDUSTRY IS RETHINKING HARVEST AND PROCESSING

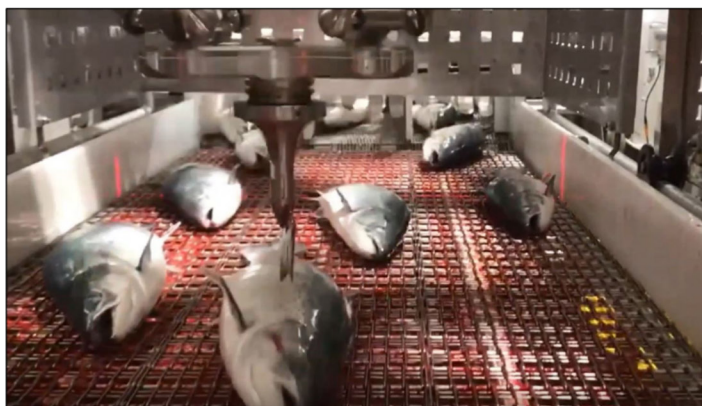
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Addressing the need to feed an ever-increasing worldwide population, the aquaculture industry is poised to help. At the same time, consumers of aquaculture products are demanding that producers proactively take measures to ensure the health and welfare of the animals grown for food consumption. While Norwegian and EU regulations require the humane treatment of animals, it is the practices and products that flow from the regulations that have actual impact. This discussion will center on current industry best practices within farmed salmon, why industry is willing to work with regulatory agencies and scientific organizations to create methods/equipment that focus on animal welfare, and how these best practices and methods can/should be adopted for other species.

Specifically, we will address how the industry is focusing on the issue of sea lice, why a lower-stress transport alternative to a vacuum pump was created, and what equipment is now considered standard for humane sedation. Additionally, we will discuss the changing dynamic of how farmed salmon is harvested and initially processed and the resulting positive effects for the environment and the bottom line.



Robotic Bleeder



Harvest, Lice Treatment, Grading - Vessels

IDENTIFICATION, MOLECULAR CHARACTERISATION OF INTERLEUKIN-1 β (IL-1 β) HOMOLOG FROM YELLOWTAIL CLOWNFISH (*Amphiprion clarkii*)

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Interleukin -1 β or IL-1 β is a cytokine that is a member of the β -trefoil cytokine family. It is produced as inactive proprotein and activated through cleavage by various enzymes. The mature peptide exerts its effect by binding with type 1 IL-1 receptors and thereby initiating the signaling. The IL-1 β gene is basically considered as a pro-inflammatory cytokine and produced by many cell types.

IL-1 β sequence was identified from the constructed cDNA library of Clark's anemonefish. Bioinformatics software and online tools were employed to characterize the identified nucleotide sequence and predicted the amino acid sequence. Healthy fish were dissected and 12 tissues including peripheral blood leucocytes, head kidney, kidney, liver, gill, and spleen were harvested to isolate mRNA and synthesize cDNA. The cDNA was used to evaluate the transcriptional difference of IL-1 β in different tissues in *A. clarkii*. Healthy fish were challenged with IP injections of LPS, Poly I:C and live *Vibrio harveyi*. Gills were dissected at 0, 6, 12, 24, 48 and 72 hour time points after the challenge. mRNA isolation and cDNA synthesis were performed and the qPCR was used to investigate the fold change of IL-1 β mRNA transcription.

A. clarkii, IL-1 β gene is composed of 741 bp ORF, 83bp 5' UTR and 397bp 3' UTR. The predicted amino acid sequence composed of 246AA. Predicted MW is 28.1kDa. Phylogenetic analysis show cladding with known fish counterparts. The multiple sequence analysis shows the IL-1 β sequence has conserved IL-1B superfamily domain within known fish IL-1 β sequences. In mRNA expression analysis shows the highest expression in gills and spleen. In the immune challenge experiment, IL-1 β was more responsive to Poly I:C treatment. However, the poly I:C treatment shows late phase induction of IL-1 β this might due to the activation of secondary immune stimulants. The IL-1 β was immediately upregulated in live *Vibrio harveyi* challenge but with low intensity compared to Poly I:C. These results suggest that IL-1 β playing an important role against viral and bacterial pathogens in *A. clarkii*.

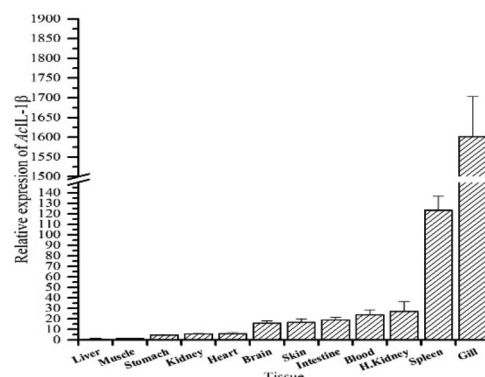


Fig.1 Tissue specific mRNA expression of the *A. clarkii*, IL-1 β

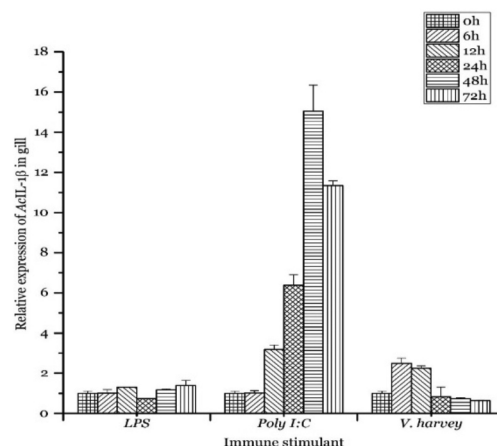


Fig.2 Relative expression of *A. clarkii*, IL-1 β in gill tissue at certain time points after different immune challenges

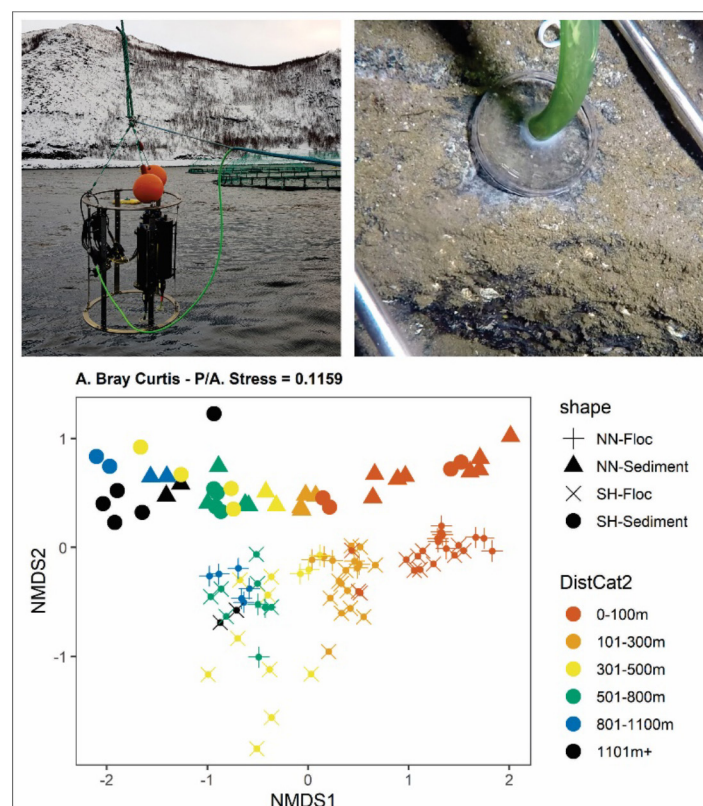
INTRODUCING SIBS: 'SUBSTRATE INDEPENDENT BENTHIC SAMPLER' FOR HARD & MIXED-BOTTOM HABITATS: A PROOF OF CONCEPT STUDY

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As is the case in most countries, the Norwegian standard for monitoring of organic impacts of salmon farming is centered around the analysis of chemical and biological parameters of soft-sediments habitats. However, many fish farms are situated in areas with hard or mixed substrates and this is likely to increase given the anticipated shift to more physically dynamic sites. Although analogous changes are probably occurring on these mixed-bottoms habitats, our knowledge of any potential interactions is severely lacking, and more critically, are going largely unmonitored due to the absence of a suitable sampling method. There are however emerging alternative biological indicators that may provide a solution. Recently, several studies have demonstrated the potential of using genetic material from micro-organisms (e.g. bacteria, foraminifera etc.), assessed through rapid, high-throughput sequencing techniques, to discern benthic enrichment. To date, these have been necessarily restricted to sampleable soft-sediment habitats. The small, ubiquitous, highly abundant and diverse nature of these organisms means that they are likely to be present in the biofilms and/or flocculent layers that overly most hard- and mixed-bottom substrates.

Here we present the first set of results using eDNA to describe the microbial communities of samples collected using a newly developed 'Substrate Independent Benthic Sampler' device. Samples were obtained from sand, shellsand, gravel, and bedrock using a low-cost surface operated suction device and are compared to more conventional microbial eDNA samples from soft-sediments and to macrofauna samples, where feasible. Minor differences were observed between sample types and farms, but in general, very good congruence was observed between the datasets with respect to distance from farm and anticipated benthic effects. Sampling of flocculent material overlying all substrates provides a potentially vital monitoring tool for the management of fish farm effects in the future.



CHROMOSOME LEVEL REFERENCE OF CUNNER (*Tautoglabrus Adpersus*) AND LOW-DEPTH RESEQUENCING REVEAL EVIDENCE OF ECOLOGICAL ADAPTATION THROUGHOUT ATLANTIC CANADA

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Cunner (*Tautoglabrus adpersus*), the northernmost ranging species of *Labridae*, are used as a cleaner fish in salmonid aquaculture. Despite their commercial importance, cunner genomics and population structure remain uncharacterized, and potential consequences of aquaculture escapees on the fitness and demography of wild populations are undefined.

To understand the existing genetic variation and evolutionary history of cunner, a 0.72Gbp reference genome spanning 24 chromosomes was constructed. Comparison to other *Labridae* reference genomes revealed 24 homologous chromosomes, suggesting a conserved genomic architecture. Low-depth resequencing of 749 individuals from 19 locations throughout Atlantic Canada identified 11 million genetic variants. Principal component analysis revealed four subpopulations in Atlantic Canada that were strongly associated with geography: Nova Scotia (NS), Northwest Newfoundland (NWNL), Northeast Newfoundland (NENL), and Southeast Newfoundland (SENL) (Figure 1A, 1B). Pairwise comparison of subpopulations via F_{ST} revealed strong signals of differentiation at specific regions in the genome and these regions overlapped with identified signatures of selection.

Genomic variation was also significantly correlated with environmental variation further suggesting that the spatial structure of cunner populations has partially stemmed from local adaptation. Observed patterns of genetic variation suggest that local adaptation to environmental differences across Atlantic Canada has resulted in genetically distinct and adaptively unique cunner subpopulations across the sampled range, these findings can inform future use of cunner within salmon aquaculture in Atlantic Canada.

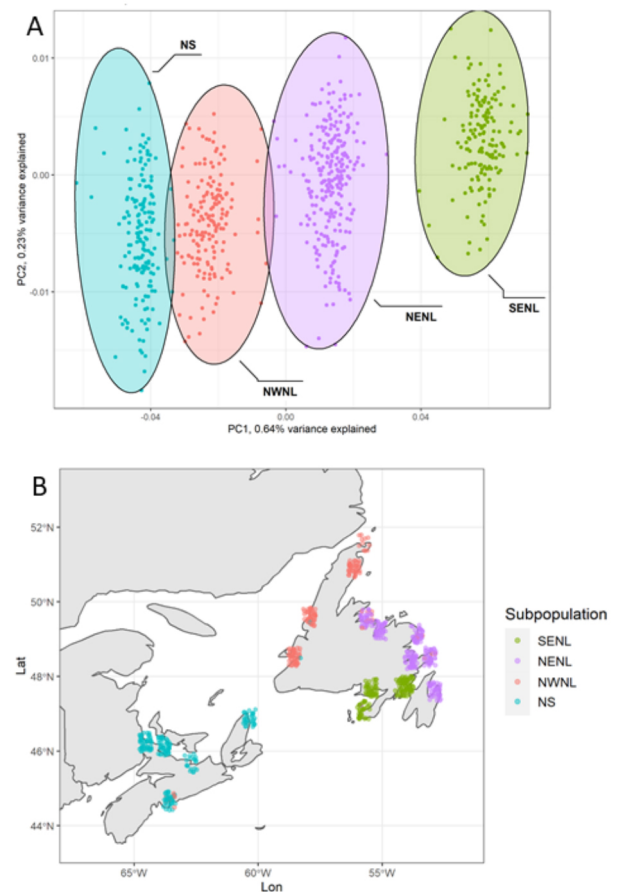


Figure 1. A) Scatter plot of Principal Components (PCs) of genetic variation for 749 cunner individuals from Atlantic Canada. The coloured ellipses denote the genetic subpopulations identified through *k*-means clustering. B) Map of sampling locations where the colour-coordinated points represent individuals and the subpopulation to which they were assigned.

DEVELOPMENT OF A CHROMOSOME-SCALE REFERENCE GENOME AND POPULATION GENOMIC DATA TO CHARACTERIZE SIGNATURES OF SEX AND EARLY DOMESTICATION IN LUMPFISH (*Cyclopterus Lumpus*)

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Parasite infections in Atlantic Salmon aquaculture pose a significant challenge to salmon aquaculture and cleaner-fish species and have become an integral part of sea lice management in salmon aquaculture. The Common Lumpfish (*Cyclopterus lumpus*) are the most frequently used cleaner-fish species and increasingly the subject of targeted breeding programs to meet a growing industry demand. However, genomic resources for this species have been lacking, limiting genomics-informed improvement, and restricting the conservation of wild Lumpfish, currently at risk in the core of their Canadian range off southern Newfoundland. Here, we develop the first chromosome-scale reference genome for Lumpfish in the Northwest Atlantic, and in conjunction with whole genome resequencing and SNP array data, explore the basis of sex determination and signatures of early domestication in a Newfoundland population.

Our genome assembly resolves 25 chromosomes, and in conjunction with population level genomic and transcriptomic data, we identify a putative male sex determining locus (i.e., anti-Müllerian hormone (AMH) on chromosome 13 (Figure 1, top), providing the potential for genetic based sex identification in future. In addition, whole genome comparisons of wild and domesticated Lumpfish from Newfoundland revealed extensive and year-class specific genomic differentiation despite only a few generations in captivity (Figure 1, bottom).

Genomic regions divergent between wild and domestic Lumpfish include regions associated with climatic variation in Newfoundland populations, and highlight the potential for future maladaptation of wild populations impacted by interbreeding with escaped domestic individuals. Together, these results reveal the utility of genomic resources in identifying genomic signatures of sex and early domestication differences within Lumpfish and will aid in future improvement and management of this species.

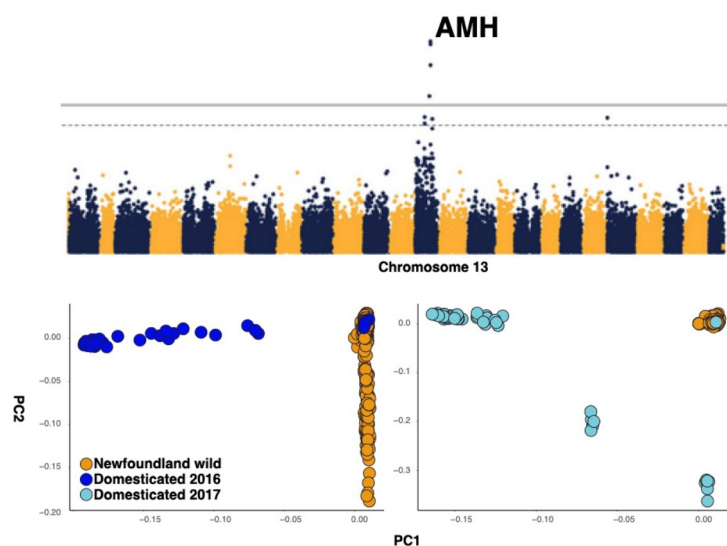


Figure 1. Identification of a sex determining locus at AMH (top) and genetic divergence between wild and domesticated populations (bottom) from Common Lumpfish population genomic data

DETERMINATION FACTORS OF FINANCIAL DECISION AND ITS IMPACT ON ECONOMIC GROWTH IN SINDH PROVINCE

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The monetary development of the areas/urban communities in Sindh is moderately lower than in different districts. This exploration demonstrated that budgetary choices majorly affect monetary development. The metropolitan spending plan decides financial development. While the nearby government spending plan centers around current spending as opposed to improvement spending, financial development will decrease. The examination additionally discovered that the monetary condition, the lawful condition, the political condition and the social condition decide the money related choices in a neighborhood government. What's more, the examination infers that the political condition impacts financial development while the monetary, lawful and social condition does not influence monetary development.

1. INTRODUCTION

Maintainable improvement stresses the parity of the three mainstays of social, financial and ecological viewpoints. Provincial advancement procedures are intended to address local issues so as to improve their jobs and capacities in overseeing social, monetary, social, instructive and social prosperity. Territorial advancement is a vital piece of the improvement of a district, particularly in country territories, which are exceedingly defenseless and are confronting real changes even with worldwide change. This change must consider parts of manageable improvement. Financial development is a marker of local advancement. Financial development is additionally a procedure that builds the capacity of a nation's economy to create products, for example, merchandise and ventures, as territorial wages.

The monetary development of an area is emphatically affected by the money related choices of the local government. As indicated by the International City/County Management Association (ICMA), there are a few factors that impact nearby government budgetary choices, including:

(1) political condition, (2) monetary condition, (3) social condition and (4) lawful condition. What's more, provincial income majorly affects the money related choices of local governments. For the districts, charge income is one of the fundamental wellsprings of income, Peacock and Wiseman (1993), which expresses that the open has an expense resilience on which the open can demonstrate the measure of assessment gathering government to back government spending. The hypothesis of Peacock and Wiseman recommends that financial advancement will prompt an expansion in duty accumulation, regardless of whether the assessment rate does not change, and expanding charge incomes will build government spending. The districts need to deal with their family units autonomously, so the administration needs to improve its open administrations. Consequently, the territorial spending plan is counter-intuitive when the extent of the financial plan for routine costs is higher. Advancement spending is a drive by territorial governments to expand open trust so as to improve provincial monetary development. In view of the past explanation, it is essential to direct more inside and out research and investigation so as to discover monetary choice equations that expand financial development.

2. LITERATURE REVIEW

2.1 Financial plan for provincial income and local consumption as a component of the usage of decentralization.

Financial development is the procedure of constantly changing the monetary states of a nation. Elements that impact monetary development incorporate HR, characteristic asset elements, science and innovation and culture components, and nearby government money related choices. As noted in the Guidelines for getting ready spending making arrangements for nearby networks, budgetary choices in the areas are ending up progressively perplexing. Current territorial money related choices are exceptionally delicate to their political, monetary, social and legitimate condition. As referenced above, ICMA expressed that there are 4 primary factors that impact the territorial government's money related choices. What's more, territorial income, particularly nearby income, assumes a job in the money related choices of local governments. Territorial Budget and Expenditure (APBD) structure the premise of local money related administration in a budgetary year. APBD is the financial plan for provincial income and consumption identified with the usage of decentralization in a given money related year. The gathering of all territorial income is gone for accomplishing the goals set out in the APBD. Additionally, all local uses and duties that load the area as a feature of the usage of decentralization will

be completed as per the figures and goals characterized in the APBD. As the APBD is the establishment of provincial money related administration, the APBD is additionally the reason for territorial budgetary control, investigation and oversight exercises. APBD is readied utilizing an exhibition approach, which is a planning framework that organizes endeavors to accomplish work or yield from arranged cost assignments or set information sources. The dimension of pay evaluated in the APBD is a sensibly estimated gauge that can be acquired for each wellspring of pay.

2.2 Political Condition

Political interests can be clarified based on spending things in the financial plan, for example, routine spending things: there are government interests, government workers. Barrier and security spending have the interests of the administration, military, police, etc. Each intrigue enters the political framework through ideological groups that can battle for the interests of each gathering. In this way, the spending banter has limited to the discussion at the dimension of gathering elites in the administration or in parliament. This issue cannot be isolated from the comprehension among business and governmental issues. Each financial, political, social, and so forth issue is inseparably connected to the issues of different divisions. With the goal that the examination of critical thinking requires interdisciplinary information. No gathering can develop without solid money related help. Accounts are expected to unite associations, enlist individuals, track objectives, and make pictures, crusades, and that's just the beginning. In the first place, every money related need of the ideological groups was secured by enrollment charges. Solid ideological connections between ideological groups and individuals make it troublesome for ideological groups to fund-raise from individuals. In any case, given the adjustments in the social structure of society and the plan of an undeniably mind-boggling popularity-based arrangement of administration, there are no ideological groups that live completely on participation expenses (Supriyanto and Lia, 2012).

Backing for ideological groups has been presented in Indonesia, in spite of the fact that the figure is as yet thought about excessively low. Thus, party administrators look for budgetary assets by performing degenerate practices. Deviations emerging from budgetary arrangements require better approaches for figuring and overseeing spending plans to give administrations to the network. Unilateralism, the battle for gatherings and their very own establishments are conspiracy to general society. The political position and the region of the spending plan have dependably been a discussion among political specialists. Spending issues are viewed as issues of government, organizations, administration, power, philosophy and legislative issues and markets, just as socio-social issues and transient political economy. The meaning of the degree and cutoff points of the political economy of the family unit is regularly viewed as indistinct and can be found all over the place. When all is said in done, nonetheless, financial strategy is the space of the job of the state, as it is viewed as an examination of political economy ponders. Hence, political power is an imperative factor in the arrangement and arranging of spending plans. The enormous issue of the political economy of the monetary allowance must be translated as an apparatus to battle against the privileges of residents as opposed to oppressing individuals. For this situation, the job of the state has the option to reformulate its job and capacity to support the general population (Wildavsky and Caiden, 2004).

2.3 Monetary Condition

Monetary elements impact money related choices in regions. Monetary conditions or conditions, for example, business cycles, expansion and loan costs, unequivocally impact neighborhood government choices on territorial spending. The financial downturn influenced provincial spending plans in two different ways. To start with, pay may decrease, particularly pay, for example, deals or pay charge, which are progressively recurrent. Furthermore, amid the retreat, state and bureaucratic incomes were regularly cruel, with the outcome that between state backing to neighborhood governments declined.

Swelling makes vulnerability in assessments of nearby government income and consumption. High swelling strongly affects spending conveyance choices. Indonesia, as a creating nation, is regularly gone up against with high swelling, which is as yet a macroeconomic issue that stresses government and society. Expansion is characterized as the inclination to build the cost of products for merchandise and enterprises as rule and proceeds. The Government's strategy on raising the cost of fuel oil (BBM) is one of the elements driving the high rate of expansion in Indonesia, as the ascent in fuel costs straightforwardly influences the general value climb, particularly for sustenance and other fundamental needs. High and precarious swelling will disturb financial strength, make business arranging progressively troublesome, diminish enthusiasm for network venture, hamper government advancement plans, and impact the spending structure set toward the start of the monetary year. Changes in financing costs can likewise influence the financial plan, in spite of the fact that the effect at nearby dimension isn't huge yet will be powerful at national dimension as the state needs on-going advances.

(Continued on next page)

2.4 Social Condition

The social condition, for example, populace changes, age dispersion and huge individual salary in the local spending plan. Expanding the extent of the populace, while keeping family unit costs unaltered, will partition the cost dimension by the quantity of relatives, bringing about a lower monetary scale, i.e. the expense per individual in the family will be lower. As money related responsibilities don't diminish in extent to populace misfortune, networks with declining populaces face challenges in lessening use. Training spending, open security is the financial plan most influenced by the age conveyance of the populace. A few past investigations have reliably demonstrated that the development of individual pay essentially influences the measure of metropolitan family units. High-salary families frequently request more and better administrations from the legislature, in spite of the fact that these family units will in general be increasingly constrained at the state level.

2.5 Lawful Condition

Lawful components that impact budgetary choices are the spending balance, the order of the focal government and nearby assessment guidelines. While allotting provincial use, account must be taken of the family unit balance. The focal government requests that the regions spend at any rate 20% of the all-out spending plan. Both focal government and commonplace governments offer commands to nearby governments. This represents an issue if the focal government does not give the full assets to the execution of the command. The neighborhood specialists must lower the other administration costs. The districts will experience the ill effects of the subsidence, which runs connected at the hip with the Center's order and reserve funds in local spending.

The presence of confinements on area/city governments to tap specific wellsprings of pay, limit charge climbs, make a parity, and request explicit administrations and dimensions of administration is a mix of variables that have caused gigantic financial worry at neighborhood government level for three decades.

2.6 City Income

The financial improvement in Sindh covers all parts of the economy of the network, both the life of country networks and urban networks, with the primary target of improving and improving the expectations for everyday comforts of the general population of Sindh. Financial advancement is done by concentrating on endeavors to develop the monetary segment, making utilization of the maximum capacity claimed by both the capability of normal assets and HR.

2.7 Theory

In view of the foundation of the issue, the definition of the issue and the examination structure and the speculation of this investigation are:

1. The political condition, the monetary condition, the social condition, the legitimate condition and the nearby income impact the budgetary choices in the territory of Sindh.

2. Monetary choices influence financial development in Sindh territory.

3. RESEARCH PROCEDURE

The populace in this investigation is the area/city work unit in Sindh territory. Research factors comprise of a variable political condition, monetary condition, political condition, social condition, lawful condition, neighborhood income, money related choices and financial development. The information in investigation utilized in this examination is the incomplete least squares (PLS) approach. PLS is a model of basic condition displaying (SEM) in light of segments or variations.

4. RESULTS AND DISCUSSION

4.1 Result

4.1.1. Test Results of the Measurement Model (Outer Model)

The external model test begins by evaluating or assessing parameters, in particular, figuring the PLS calculation. In view of the yield examination, the estimation display (external model) can be assessed by checking the concurrent legitimacy, discriminant legitimacy, and unwavering quality. All focuses are substantial and dependable inquiries.

(Continued on next page)

4.1.2. Way Coefficient Examination Results

The auxiliary model in PLS is assessed utilizing the reliant variable R square and the free factor way coefficient esteem, which at that point assesses their hugeness dependent on the t-measurement estimation of every way. The PLS result program calculation for assessing the estimation of way and Rsquared coefficients is appeared Table 1 underneath:

Table 1 Path Coefficient Value

Table 1 Path Coefficient Value

Variable	Financial Decision (Y1)	Economic Growth
Financial Decision		0.799
Political Environment (X1)	0.305	0.175
Economic Environment (X2)	0.486	-0.015
Social Environment (X3)	0.130	-0.012
Legal Environment (X4)	0.127	-0.008
Local Government Revenue (X5)	0.172	0.092

Sources: PLS Results (2019).

4.2 Discourse

4.2.1. Effect of the StrategyCondition on Money Related Choices

The consequences of the investigation presumed that the political condition spoken to by the inclusion of the network in the planning procedure had a positive and huge effect on the money related choices taken by the neighborhood government. In light of the past portrayal, political interests impact the dispersion of consumption contained in the state spending plan. Each issue has interests, both the interests of the legislature and the network and political interests. From focal government spending, for example, routine consumptions, these shopping centers are in light of a legitimate concern* for government employees. The discussion in setting up the financial backing was for the discussion at the dimension of the gathering elites in the EU

Government or Parliament. The technique for participating in political interests should be possible through studies in the network when ideological groups make breaks or amid the muscling. The incorporation of political interests in planning will influence the nature of the financial backing in help of territorial improvement. The conveyance of spending is firmly affected by political interests. The administration's budgetary choices in the dissemination of foundation spending, monetary spending, and socio-social spending rely upon the exchanges between the assembly and the official. Political interests are not generally in accordance with the interests of territorial advancement, which regularly prompts clashes in planning. This issue cannot be isolated from the comprehension among business and governmental issues. Each financial, political, social, and so on issue is inseparably connected to the issues of different areas. With the goal that the examination of critical thinking requires interdisciplinary information.

4.2.2. Effect of the Monetary Condition on Money Related Choices

Monetary factors emphatically impact the money related choices in the districts. The consequences of the examination demonstrate that the monetary condition impacts the money related choices of the regions. This implies neighborhood governments in Sindh Province are thinking about financial factors in deciding the appropriation of foundation spending, monetary consumption and socio-social use, outstandingly swelling, loan fees and financial cycles in the area. The monetary conditions or conditions as business cycles, expansion and loan costs unequivocally impact the choices of regions in the assignment of territorial consumption.

The high expansion firmly impacts the choices about the dispersion of the consumption. Indonesia, as a creating nation, is frequently gone up against with high swelling, which is as yet a macroeconomic issue that stresses government and society. High and unsteady swelling will upset the steadiness of the economy, make business arranging increasingly troublesome, decrease the enthusiasm of open speculation, hamper the administration's advancement plans, and impact the financial structure built up toward the start of the monetary year. Changes in loan costs can likewise influence the financial plan as it is one of the fundamental suppositions or presumptions in planning. Be that as it may, the effect at nearby dimension isn't substantial, yet will have a noteworthy effect at national dimension as these suppositions change from their unique esteem. This influences the dimension of the pay part, both on government income and on government use and spending shortages, which lead to changes in the dimension of spending financing.

(Continued on next page)

4.2.3. The Impact of the Social Condition on Money Related Choices

The social condition significantly affects the money related choices of regions, particularly in the readiness of territorial income and use spending plans. For state funded instruction, open wellbeing is impacted by the age conveyance of individuals in the region of each territorial government. Changes in the populace in a territory will influence provincial spending choices, in light of the fact that the more individuals live in one zone, the broader assets will be made accessible to the administration. A marker of the measure of the General Allocation Fund is the all-out populace and territory. What's more, the populace likewise impacts local spending, as the substantial populace in a territory of development organizers is viewed as the capital of advancement and, in the meantime, the weight of improvement. Vast populaces can likewise be considered as territorial resources if network individuals can improve their quality and aptitude or abilities to build national creation.

The age conveyance of the occupants of a zone will impact the monetary choices of the district. One part of spending that is affected by age dissemination is instruction spending. The more school-age occupants, the higher the spending on training.

A few past examinations reliably demonstrated that the development of individual pay essentially influenced the dimensions of city spending plans. High-salary families regularly request more and better administrations from the legislature, despite the fact that these families will in general be progressively constrained at the state level.

4.2.4. The Impact Ecological Factors on the Monetary Choice

Legitimate components with spending asset reports, changes in spending plans and incorporated guidelines have a positive and noteworthy effect on the monetary choices of regions. While allotting local use, account must be taken of the family unit balance. Harmony between provincial income and consumption. Spending changes additionally bigly affect monetary choices. On account of a salary change, acquiring must likewise be balanced. For instance, if the nearby government sets an inn charge focus of IDR 100 million for one year in the year being referred to, incidentally, the primary semester outflanked the financial plan for changes in the territorial burning through segment. Neighborhood governments can expand the quantity of exercises that will be changed in accordance with the evaluated pay rate that is typically accomplished for the current year.

4.2.5. Effect of Local Introductory Pay on Money Related Choices

Nearby income is a factor that influences the financial plan. The budgetary choices of neighborhood government depend intensely on the measure of nearby salary. The higher the nearby income, the higher the local spending. Territorial self-rule obliges locales to back local needs with wellsprings of salary that can be collected by the provincial government. The degree of the area's capacity to build its local starting income is impacted by wellsprings of income, strategies for gathering civil income, and observing metropolitan income.

The pay wellspring of the area/city has arrangements. The dissemination of focal, commonplace and regime/city income has been represented by government guidelines, and the measure of income that can be created by the territorial government has likewise been directed by law. The strategy for gathering the incomes of the regions unequivocally impacts the dimension of territorial salary.

4.2.6. Effect of Monetary Choices on Financial Development

Financial development, destitution and joblessness are positively inseparably connected to government contribution as strategy producers and the ability to utilize the monetary allowance to animate the economy through the local income and use spending plan, which is required to quicken the economy. Because of this affirmation, the improvement of a territory with pointers of monetary development and success of the network unequivocally relies upon the money related choices of the districts. With the quick turnaround of the economy, new monetary open doors are normal which will unquestionably pull in specialists who are attempting to diminish joblessness and improve individuals' expectations for everyday comforts as opposed to neediness. The aftereffects of this investigation mean to perceive how the APBD government is affecting development, diminishing joblessness and destitution in Sindh.

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5G ENABLED AQUACULTURE MONITORING SYSTEM

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EU R&D consortium 5G-HEART (5G HEalth AquacultuRe and Transport validation trials, www.5gheart.org) develops and executes large scale healthcare, aquaculture and transport trial activities on actual testbed in EU. The use case for the aquaculture vertical will comprise of two pilot sites, showcasing the applicability of the solution developed in different locations. The first site in Greece will utilize the 5G-EVE node and the second pilot in Norway will use the Norwegian node of 5G-VINNI to access the network. The European Commission is focusing on the development and application of new and emerging technologies to aid the sustainable development of European aquaculture through the introduction of 'smart farming'. Sharing farm data and integrating Big Data, the Internet of Things, Artificial Intelligence and Deep Learning into husbandry practices will contribute to standardisation and the possibility to benchmark the best production conditions. This will lead to better growth predictability and feeding regimes, more precise welfare and health monitoring, autonomous operations and improved safety conditions for aquaculture workers. Monitoring of the fish stocks, environment and the farm infrastructure are key areas for maintaining optimal production and minimizing environmental impacts of aquaculture. Significant advances have been made in recent years through the development of technologies for autonomous data acquisition and communication in the areas of monitoring fish behaviour and feeding within the net pens on marine farms, monitoring water quality parameters to determine environmental impacts and the integrity of the farm structures to minimize the potential of fish escaping. These technologies need to be supported by an appropriate communication and data processing infrastructure, capable of supporting offshore systems. 5G connectivity offers the opportunity to connect a wide range of sensors directly with applications, enable real-time video monitoring with negligible latency and supports remote monitoring, thus providing a ubiquitous communication infrastructure solution for aquaculture.

In this presentation, 5G enabled aquaculture vertical trials performed by EU R&D consortium 5G-HEART are introduced.



Figure 1 Panorama overview of the 5G-HEART Norwegian fish farm

OPERATIONAL CHALLENGES AND SUCCESSES OF BUILDING A LARGE INDIGENOUS OWNED VERTICALLY INTEGRATED SCALLOP CULTURE ENTERPRISE IN NORTHERN BRITISH COLUMBIA

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The Metlakatla First Nation is developing a shellfish aquaculture industry on the North Coast of British Columbia through Coastal Shellfish LP (CSLP). After almost two decades of trials, efforts and investment, CSLP has achieved proof of concept and is expanding farming operations to handle increased seed with a goal of harvesting > 6 million Japanese scallops (*Patinopecten yessoensis*) per annum. Commercialization in a region with no established aquaculture is requiring simultaneous efforts and investments at all steps of the value chain. Hatchery operations are focusing on improving productivity and efficiencies. Expansion of marine operations includes new farm sites and associated infrastructure and obtaining CSSP classification. Processing operations have required developing and licensing a bivalve processing facility. All phases have required recruiting and developing operational teams. An overview of the development of the project, challenges and successes experienced from an operational aspect will be presented.

CHANGES IN THE EXPRESSION OF MESSENGER RNA ENCODING TWO AMP KINASE SUBUNITS IN RESPONSE TO FOOD INTAKE IN THE MUSCLE AND LIVER OF ATLANTIC SALMON

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The ability of the tissue to sense available nutrients alters tissue nutrient metabolism and tissue growth and possibly influences food intake. The AMP protein kinase (AMPK) is an evolutionarily conserved nutrient sensor critical for proper growth and development. In our previous study in channel catfish, expression of AMPK mRNA was influenced in a subunit-specific fashion by food intake in the liver but not in the brain or muscle tissues. Whether changes in the expression observed in channel catfish will be similar in other fish species is unclear. In the current study, we hypothesized that changes in food intake affect the AMPK mRNA expression in Atlantic salmon. The objective of this study was to examine muscle and liver expression of AMPK mRNA in response to changes in food intake. 72 Juvenile Atlantic salmon (6 fish per tank) were cultured in 12 flow-through tanks during a 14-day feeding study (). On day 0 of the study, we randomly assigned tanks to one of three feeding groups (n=4 tanks per group). One group received feed twice daily (fed, control), whereas the fasted group did not receive food for 14 days. The third group did not receive food for the first seven days of the study and was fed twice daily for the subsequent seven days (Refed). Muscle and liver were collected on day 14, and the expression of AMPK alpha 1 and beta 1 subunit mRNA was measured using qRT-PCR. The AMPK alpha 1 and beta 1 mRNA expression in the liver and muscle was similar among the three feeding groups. Given the preliminary nature of the current study, we plan to investigate to clarify the role of AMPK regulates growth, food intake, and nutrient metabolism in Atlantic salmon.

CHARACTERIZING THE SHELL MICROBIOME OF AMERICAN LOBSTER *Homarus americanus* FROM ATLANTIC CANADA

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Rising temperatures and changing environmental conditions in our oceans can lead to increased physiological stress upon marine invertebrates and the emergence and spread of infectious diseases. The epizootic shell disease (ESD) in American lobsters *H. americanus* is characterized by fast spreading cuticle lesions that can invade underlying tissue which can increase mortality and lower reproduction, especially in larger specimen and ovigerous females. Outbreaks of ESD have been associated with warmer summer temperatures and were likely a factor in the collapse of the Southern New England lobster stock in the 1990s. A negative shift in the shell microbial community together with environmental stressors has been proposed to cause ESD proliferation in lobster populations.

Due to current knowledge gaps regarding the shell microbial community associated with *H. americanus* in Atlantic Canada, this study aims to describe and analyze the shell microbiome of lobsters sampled from several locations in New Brunswick, Nova Scotia and Prince Edward Island.

Long-read, next-generation 16S rDNA amplicon sequencing (PacBio) of cuticle samples followed by bioinformatic analyses will identify the shell associated bacteria to species level. Diversity indices will assess the microbial composition and diversity while network analyses explore bacterial interactions within the microbiome. Furthermore, multivariate analyses will detect any patterns in microbial species' abundances, composition or distribution based on biotic and abiotic factors.

It is expected that spatial, temporal, and environmental variables as well as lobster characteristics such as sex, size, or molt stage to some extent influence microbial profiles on lobster cuticles. Bacterial taxa that have been associated with ESD are likely present in lobsters from Atlantic Canada as they are ubiquitous in the marine environment but may play a role in ESD proliferation.

This study will help to assess and predict the risk of ESD outbreaks in Atlantic Canada and in turn will encourage the development of suitable fisheries management strategies in the future.

AQUACULTURE FOOD SAFETY AND DISEASE PREVENTION PLANS

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Aquaculture is the primary component of the seafood trade which makes up the largest traded food group in the world. More than all grains, banana, cocoa, rice, and all other meats combined. Aquaculture products are also the primary source of protein for one fifth of the world's population, mainly in the developing world. This is extremely important for food security because the world's population is expected to grow from 6.8 to 9.2 billion in 2050, causing the demand for food to increase 110%. This will result in a huge demand...an opportunity... for aquaculture products. However, there are serious food safety concerns with aquacultured products. These concerns include the presence of unsafe residues, pathogens, and the potential to contribute to antimicrobial resistance. When food safety problems are found there are a range of negative impacts. These include significant costs to producers, processors, buyers, importers, and the regulatory agencies, market disruptions, an increase in the negative perception of aquaculture, lost markets, etc. This talk will describe a new way of approaching aquaculture food safety linking it to disease prevention and minimizing the risk of trade interruptions due to food safety concerns, and an approach to obtaining more customer confidence in aquaculture.

BEHAVIORAL STRESS RESPONSE OF FARMED ATLANTIC SALMON (*Salmo salar*) TO UNFAVORABLE WATER QUALITY AND FARM OPERATIONS

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Atlantic salmon (*Salmo salar*) are a commercially important species across the globe and specifically in Atlantic Canada. There has been rapid expansion in production rates over the last few decades to meet an increasing demand for healthy, sustainable protein. The welfare of these animals has been of increasing interest to consumers, producers and regulating bodies. As the industry continues to expand, the welfare of fish in aquaculture must be prioritized to ensure the viability of scalable operations. It is imperative to define a welfare standard using a variety of indicators that can be implemented as standard operating procedures.

Advancements in technology have allowed for the increased ability to monitor animals *in situ*. Acoustic fish tags and bio-loggers can be used to monitor a variety of parameters including heartrate, temperature, acceleration, and depth of fish as they experience regular aquaculture practices and adverse water quality conditions. Environmental sensors that monitor dissolved oxygen, temperature and depth can be used to capture fine-scale changes in water quality within individual pens on a farm. The combination of these sensors and a consistent log of daily farm activities can be used to identify when stressful conditions on a farm persist, and the bio-loggers can measure the response of fish to these conditions. Seventeen Atlantic Salmon (*Salmo salar*) were each tagged with two tags measuring heart rate, acceleration, temperature, and depth. Five sensors monitoring temperature and oxygen on a profile line were also deployed in the pen. Data collection is ongoing, but some data is available in real-time and has been analyzed to date. The full dataset of both environmental and fish tag data will be available when the fish are harvested in June 2022.

This study aims to correlate and grade the severity of the physiological and behavioral response to stressful water quality conditions and regular farm operations to gain a better understanding of how these events impact livestock. The findings of this research will provide further insights to farmers on the impact of standard farm operations to enable them to make better decisions when caring for their fish.

FISHERIES AND OCEANS CANADA PACIFIC REGION BIVALVE TRACEABILITY INITIATIVE

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The Fisheries and Oceans Canada (DFO) Pacific Region Bivalve Traceability Initiative (BTI) was struck in May 2019, in response to growing concerns related to illegal and non-compliant harvest and lack of traceability of bivalve shellfish product in British Columbia, which threatened the integrity food safety system. Maintaining traceability (or control of harvest) is a core responsibility for DFO under the Canadian Shellfish Sanitation Program (CSSP) and an important element in proper management of a fishery. The BTI was co-led by DFO Fisheries Management (wild Resource Management and Aquaculture Management Division) and Conservation and Protection staff, with support of National Headquarters colleagues. A senior management committee, led by the Regional Director General Pacific Region with participation of the BC Government, Environment and Climate Change Canada and the Canadian Food Inspection Agency, was also struck to explore options to collaboratively tackle these issues.

Actions conducted under the Bivalve Traceability Initiative have focused on:

- Compliance Enforcement;
- Communications and Outreach;
- Governance and Engagement; and
- Sector Oversight and Policy

Over the past three years, DFO resource managers (wild and aquaculture) and fishery officers actively conducted ongoing compliance promotion activities with licence holders and stakeholders. Administrative condition of licence verification, patrols, and inspections were conducted on both the aquaculture and commercial fisheries, focusing on the key elements of traceability (tagging, record keeping, reporting, etc.). Regulatory changes were made to address traceability gaps in the shellfish aquaculture, wild commercial clam, and wild commercial oyster fisheries and communications tools were developed and employed to increase licence holder awareness of traceability requirements.

This presentation will give an overview of the last three years of the Bivalve Traceability Initiative, as it pertains to shellfish aquaculture management, including: its successes and opportunities, key findings, and recommendations for moving forward.

ORAL DELIVERY OF A VACCINE AGAINST STREPTOCOCCOSIS IN NILE TILAPIA

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The injection route is the most common method used for the administration of commercial vaccines but it presents some drawbacks; it is time consuming, requires qualified personnel and may induce local reactions at the injection site. Mucosal vaccination is a good alternative and allows to develop protective local immunity in order to struggle against infectious diseases. Oral administration of vaccines is particularly suitable for protection of breedings with high density of animals like in fish farming. The main challenge linked with this route of administration is to preserve the integrity of the antigen from the harsh environment created by the digestive system to reach gut associated lymphoid tissue (GALT) in order to induce a potent local immune response. This issue could be gotten around by using an adapted adjuvant.

The Gastro-Resistant Adjuvant (GRA) tested here, is an oily adjuvant containing a gastro-resistant matrix to protect antigen from gastric degradation. Indeed, an *in vitro* experiment showed GRA formulated with BSA was able to retain the antigen in acidic media and release it under neutral pH conditions. Then, a study in tilapia was performed on a *Streptococcus agalactiae* vaccine formulated with GRA and mixed at 2% or 20% with feeding pellets. The tilapia have been orally vaccinated under 2 periods one week apart as follows : first, during 4 consecutive days, then during 3 other days. Each vaccine group was constituted with 3 replicates of 40 tilapia. At D49, the tilapia were challenged with *S. agalactiae* serotype III by intraperitoneal route. The protective rate of GRA groups has reached 91.1% and 88.9% for vaccines formulated at 2% and 20% respectively while it was only 73.3% in the control group (vaccination with non-formulated antigen).

Taken together, these results show that GRA is well adapted for the oral administration of vaccines in tilapia with a good palatability, safety and efficacy. However, more data is needed especially in a large-scale trial and also for cold water fishes to confirm these first observations before considering other animal species.

THE GOOD CONCEPT

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The GOOD concept provides optimal conditions for cleaner fish. It is important to acknowledge the cleaner fish's biology and behavioral patterns to protect it from danger and give it the opportunity to return to safe grazing areas. Correct protection, adapted feeding and a clean environment provides the cleaner fish with the best prerequisites for effective lice grazing. The GOOD concept is a collaboration of four different products from OK Marine consisting of the cleanGOOD, feedGOOD, liveGOOD and leadGOOD. **cleanGOOD** is the main hide which provides the cleaner fish with the best possible environment and is an effective meeting place for the salmon and the cleaner fish. **feedGOOD** is a reliable and robust cleaner fish feeder designed to provide a minimum of maintenance at all conditions. **leadGOOD** will lead the cleaner fish from the bottom of the pen and back for the survival of wrasse in winter, as the wrasse swims deeper at colder temperatures. Without a hiding place at the bottom of the pen, the wrasse does not have anywhere it can hide and hibernate. **liveGOOD** provides the lumpfish with a safe resting place when it is led to the net by currents, thus increasing the survival of cleaner fish in exposed conditions. The concept is designed to take care of all the cleaner fish's needs and to reduce mortality. Every component works together and compliments each other to make the cleaner fish as efficient as possible. The GOOD concept has been developed in close collaboration with breeders and is based on solid competence and long experience. It is easy to clean or replace the equipment.

Several fish farmers have experienced that during the crowding of fish the sealice separates from the fish and moves freely in the water. This causes an infection risk for the farm, farms close by and for wild fish. **CatchLICE** is a harvesting net designed to keep the lice inside the net.

It is easy to use and is gentle on the salmon. The sealice are bigger than the openings of the mesh, and prevents lice from re-spreading by pumping lice on board the treatment vessels to gather in the filters.

IDENTIFYING ISOFORM SWITCHES IN ATLANTIC SALMON (*Salmo salar*) INFECTED WITH *Moritella viscosa*

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Alternative splicing is a regulatory process during gene expression that results in a single gene coding for multiple proteins. A vast majority of eukaryotic genes use alternative transcript isoforms. The alternative isoforms are generated through alternative transcription start sites (aTSS), alternative transcription termination sites (aTTS), and alternative splicing (AS). Gene isoforms are predominantly tissue-specific and may alter the stability, cellular localization and function of the gene. A switch is defined as a case where the relative contribution of the isoforms to the parent gene expression changes significantly between conditions (e.g. healthy vs. diseased). In this study, the skin transcriptome using RNA-seq of Atlantic salmon infected with *Moritella viscosa* (a Gram-negative bacterium that causes winter ulcer disease) was used to examine the isoform usage of immune genes in healthy vs. infection samples. The skin samples were collected from control and infected fish [at lesion (at) and away from lesion (adj) sites].

HISAT and StringTie software performed alignment and quantification of the skin transcriptome. IsoformSwitchAnalyzeR Bioconductor R package was used to analyze isoform usage, providing a platform to integrate many sources of predicted annotation such as open reading frame, protein domain (via Pfam), signal peptides (via SignalP), intrinsically disordered regions (IDR, via NetSurfP-2), coding potential (via CPAT or CPC2) and sensitivity to non-sense mediated decay (NMD). Differential isoform usage analysis was performed using DEXSeq and DRIMseq.

In total, 615 switches from 853 isoforms (486 genes) were identified. The comparison at lesion (at) and adjacent to lesion (adj) of infected fish with control (non-infected skin samples) resulted in 470 and 103 switches from 675 isoforms (383 genes) and 136 isoforms (986 genes), respectively. The genome-wide analysis of alternative splicing was enriched in intron-retention (IR gain), alternative transcription termination sites (ATTS gain) and alternative 5'end donor sites (A5 gain) at false discovery rate (FDR) < 0.05 (Fig. 1a). Some of the examples of enriched alternative isoform usage events were observed in *cadherin-13*, *g0/g1 switch protein 2*, and *c type lectin receptor b*. Signalling triggered by *C-type lectin receptors* (CLRs) plays crucial roles in activation of immune response against infections. CLRs have diverse immunoregulatory functions associated with phagocytic, anti-microbial, and inflammatory responses (Fig. 1b). Isoform switches identified in the present study may be employed as functional biomarkers to predict functions that impact vital biological processes.

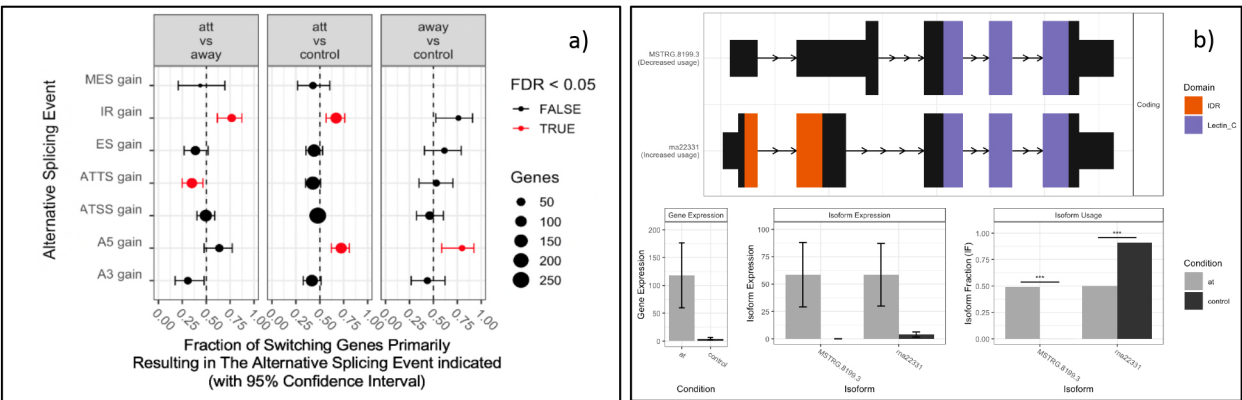


Fig 1. a) Enriched alternative splicing event at FDR < 0.05; b) The Isoform switch in *c-type lectin receptors* (CLR). MES (Multiple Exon Skipping), IR (Intron Retention), ES (Exon Skipping), ATSS (Alternative Transcription Start Sites), ATTS (Alternative Transcription Termination Sites), A5 (Alternative 5'end donor sites), A3 (Alternative 3'end acceptor sites).

INNATE IMMUNE RESPONSE IN HEAD KIDNEY, SPLEEN AND LIVER OF CLEANER FISH (*Cyclopterus lumpus*) DURING *Aeromonas salmonicida* INFECTION

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Lumpfish are cleaner fish widely used for parasite (sea-lice) control in large scale Atlantic salmon (*Salmo salar*) farming. *Aeromonas salmonicida* is a prevalent Gram-negative bacterial pathogen that directly impacts lumpfish survival. In this study, the whole transcriptome-wide response in lymphoid tissue such as head kidney, spleen and liver were studied after administering a lethal dose of *A. salmonicida*. The samples were collected at zero, three, and ten days post-infection (dpi). RNA sequencing (RNA-seq) based transcriptomic profiling revealed several transcripts and genes to understand the immune mechanisms in fish during pathogenic infection.

Trinity and Trinotate software performed *de novo* assembly and annotation of the transcriptome. RSEM software was used to quantify transcript expression before identifying differentially expressed transcripts (DETs) using DESeq2 and edgeR. The samples collected at three dpi is a critical time point to study the crosstalk between both the innate and adaptive immune response. The most DE transcripts (|FC| > 2 and adj. P-value ≤ 0.05) between three dpi vs zero dpi is 1789 (720 up-regulated and 1069 down-regulated). We further identified 450, 1049 and 224 unique DETs in Head Kidney (234 up-regulated and 216 down-regulated), Spleen (370 up-regulated and 679 down-regulated), and liver (105 up-regulated and 119 down-regulated) respectively between three dpi vs zero dpi.

Antimicrobial peptides (innate immunity), such as hepcidin (*hamp*), hemopexin (*hpx*), and haptoglobin (*hp*) have a significant role in regulating inflammatory response during bacterial infection. We identified several significant transcripts of *hamp*, *hpx*, and *hp* overexpressed in all the three central lymphoid tissue. We also identified heparanase (*hpse*) transcript (down-regulated) that is critical to effective wound healing process. The complement system defence mechanism is diverse, and it triggers inflammation, attract phagocytes to the infection site and plays a role in the activation of naive B-lymphocytes. Interestingly, we observed complement system 3 and 6 up-regulated in head kidney and spleen, respectively, while complement system 5 down-regulated in liver. We also found several DETs involved in pathways of complement activation. We further catalogued innate immune genes such as *hck*, *malt1*, *aimp1*, *ccl13*, *il1b*, *stat3*, *acod1*, *pik3cg*, and *lbp* response. These results provided with novel insights of *A. salmonicida* infection and how the fish innate immune system is hijacked during infection.

L'ALIMENTATION DES POISSONS, UN FACTEUR LIMITANT POUR LA PERFORMANCE « GAIN EN POIDS » DES SOUCHES DE TILAPIA, *Oreochromis niloticus*, ELEVEES EN AFRIQUE SUB-SAHARIENNE. CAS DE LA PISCICULTURE PAYSANNE A L'OUEST DE LA RDC

Étude comparative des performances de gain de poids de deux souches de tilapia dans l'ouest de la République démocratique du Congo

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Le tilapia est l'un des principaux poissons d'élevage en Afrique. Cependant, malgré le potentiel d'exploitation qu'offre cette pisciculture, l'élevage ne semble pas encore maîtrisé en Afrique sub-saharienne. A l'Ouest de la RDC plus précisément dans les provinces du Congo-Central, du Kwango et du Kwilu, le tilapia est la principale espèce d'élevage. Pour répondre à la question de savoir si le faible poids du tilapia dans l'Ouest de la RDC était dû à une dégénérescence génétique ou simplement à un mauvais système d'élevage, nous avons introduit une nouvelle souche de tilapia que nous avons comparée à la souche locale. Les deux souches ont été nourries sur base de l'alimentation interne avec un complément de 25 kg/ha d'aliment concentré (18% DP) et 50 kg/ha de déjections de poulets tous les 10 jours.

Le poids moyen de 85,94 g; 150,18 g et 182,28 g ont été obtenus avec la souche locale respectivement à 3, 6 et 9 mois. Alors qu'avec la souche introduite, le poids moyen de 83,34 g; 149,12 g et 218,22 g ont été observés. Les Analyses statistiques des variances des taux de croissances observés aux différents stades (âges) ont montrées que les différences observées entre les deux souches de tilapia ne sont pas significatives. Au terme de notre étude, il est clair que le très faible poids des tilapias produits par les paysans de l'Ouest de la RDC sont dus à l'utilisation de pratiques nutritionnelles inadéquates. La nutrition est actuellement le facteur limitant des performances de l'élevage du tilapia dans l'Ouest de la RDC. Cette étude ouvre des perspectives de recherche des solutions adéquate au développement de la pisciculture à l'Ouest de la RDC.



Photos1 : Deux tilapia des deux souches différentes récoltés à 9 mois d'élevage à la Station de Gimbi. Gimbi est l'un des sites expérimentaux de l'Institut National pour l'Etude et la Recherche Agronomique situé dans le District du Bas-fleuve à l'Ouest de la RDC.

MONITORING BAY-SCALE ECOSYSTEM CHANGES USING EDNA IN WATER SAMPLES

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Aquaculture is intimately linked to the functioning of the receiving ecosystem through various direct and indirect interactions and feedbacks. To date, aquaculture research has predominantly focused on near-field benthic effects. Limited research has documented bay-scale ecosystem impacts, including the potential impacts at higher trophic level. Monitoring bay-scale aquaculture ecosystem interactions requires a method and a sampling design able to detect changes in ecosystem structures over time. With its power to increase coastal survey coverage, environmental DNA (eDNA) is increasingly employed to monitor ecosystems and support decision-making. However, little is currently known about how the detection rate of coastal eDNA changes seasonally and regionally in relation to biological and physical factors. As a result, there is no guidance on optimal sampling periods to design eDNA studies for most species. Better understanding how factors alter eDNA presence are not only fundamental to optimizing eDNA project planning and avoiding false negatives, but also to interpreting temporal trends in eDNA detections within and between ecoregions, including better understanding the discrepancies between eDNA and specimen-based surveys results. We will present a new DFO initiative aiming to develop an online tool to report and visualize trends in spatio-temporal eDNA distributions of Canadian species. The latter will fill important knowledge gaps on how genomic tools can differentiate impacted and non-impacted ecosystems associated with human activities, such as aquaculture.

ASTAXANTHIN METABOLISM AND FLESH PIGMENTATION IN ATLANTIC SALMON: GAPS IN KNOWLEDGE

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The role of astaxanthin (Ax) and canthaxanthin (Cx), in flesh pigmentation of salmonids is widely recognized. The retention of these carotenoids in flesh is low (5-20%) and is affected by genetic factors, the efficiency of absorption, transport, tissue uptake, stages of growth, development, metabolism and excretion. Although nutritional strategies have been developed to effectively utilize synthetic and natural sources of Ax and the application of carotenoid dynamic models for different strains and species of salmonids by the aquaculture industry, the progress in research to better understand the molecular basis of carotenoid metabolism in fish has been slow. In recent years, research has been focused on the biochemical mechanisms involved in Ax absorption, transport, cellular uptake and metabolism, muscle binding with protein and the molecular basis of Ax and Cx breakdown by carotenoid cleaving enzymes in Atlantic and Pacific salmon. Beyond flesh pigmentation, Ax and Cx have also been shown to have other physiological functions e.g., provitamin A, antioxidant, reproduction, immunity and health. Astaxanthin is considered an effective biological antioxidant that prevents stress disorders, which are linked to the primary cause of certain diseases in humans and experimental animals.

In this presentation, a brief review of current knowledge of Ax metabolism and the bottlenecks involved in its low absorption and retention will be discussed. The progress made to date in molecular mechanisms involved in carotenoid metabolism in other vertebrates may be useful to examine the problems associated with absorption and to better define the role of Ax in oxidative stress related diseases and fish health.

DISTINCT SPATIAL GENOMIC STRUCTURE OF LUMPFISH *Cycloterus lumpus* ACROSS THE NORTHWEST ATLANTIC

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Lumpfish, *Cycloterus lumpus*, have historically been harvested throughout Atlantic Canada but are also increasingly in demand as an alternative solution to controlling sea lice in North Atlantic salmon farms - a process which involves both the domestication and the transfer of individuals between geographic regions. However, little is known regarding population structure of wild Lumpfish, restricting attempts to assess the potential impacts of escaped Lumpfish used in salmon aquaculture on wild populations. Here, our goal was to characterize the spatial structure of wild populations of Lumpfish throughout the Northwest Atlantic, with a specific focus on the waters around Newfoundland.

We obtained both 70K SNP array data and whole genome sequence data (WGS) for samples from Newfoundland (N=874, NL), the Gulf of Saint Lawrence (N=107, Gulf), Nova Scotia (N=13, NS), and Maine, USA (N=70). Our results suggest that at broad spatial scales, the southern populations (Maine and NS) were highly differentiated from more northern populations (NL and the Gulf; Fig 1a), and that this break was environmentally associated and consistent with adaptive differences. At finer spatial scales, our results suggest two distinct groups in the north, one that encompassed the majority of NL and all the Gulf, and the other which encompassed predominately juveniles caught inshore NL (some adults were present as well, Fig 1a). There was also evidence of subtle structure within the NL regional group, with significant genetic isolation by distance (fig 1b) and environmentally associated structure detected.

This study represents the first description of population structure within Lumpfish in Atlantic Canada, revealing evidence of broad and fine geographic scale genomic and likely adaptive diversity in this species. These results directly inform the use of Lumpfish as a cleaner fish in Atlantic Salmon aquaculture and the conservation of this at-risk species throughout Atlantic Canada.

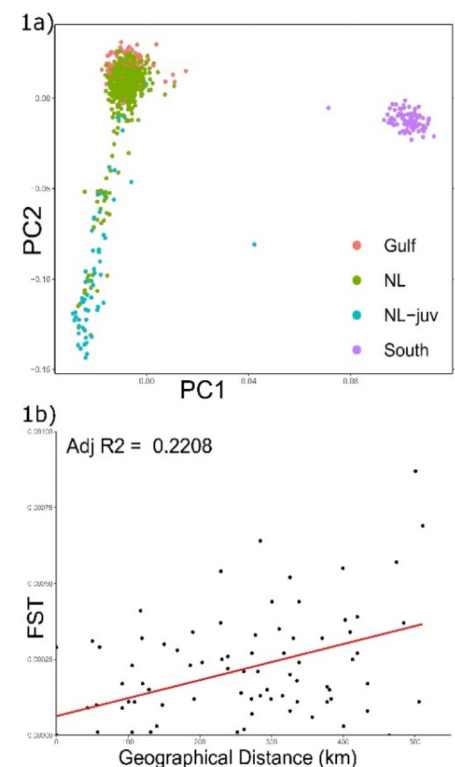


Figure 1. Population structure of Lumpfish throughout Atlantic Canada (A) and isolation by distance within Newfoundland sample locations (B).

BEYOND TAXONOMY: VALIDATING FUNCTIONAL INFERENCE APPROACHES BASED ON EDNA METABARCODING DATA IN THE CONTEXT OF FISH-FARM IMPACT ASSESSMENT

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In biomonitoring and impact assessments, the community's metabolic capability may in fact be of more interest than conventional taxonomic identity. Taxonomic and functional profiles can respond differently to biogeography, abiotic environmental changes (e.g. organic content, metal concentration) and community processes and interactions, and exhibit different level of stochasticity and temporality. Having comprehension of both aspects can increase our understanding of community turnover and of its resilience to further changes via the computation of functional redundancy within the community (also referred to as contributinal diversity).

Metagenomics has recieved considerable attention in the last few years for its ability to simultaneously determine which organisms are present, but also what metabolic capabilities they possess. However, it's mainstream use is still limited by it's relatively low sample throughput, cost-efficiency, and heavy computational and data management requirements. To circumvent those issues, several programs have been developed to infer functional profiles from 16S eDNA metabarcoding (e.g. PICRUSt2). While not as accurate as metagenomics functional analyses, these methods can provide more complete functional profiles as they do not require high sequencing depth to assign functions, and can be particularly useful in situations where metagenomics would be prohibitively expensive, such as broad microbial routine monitoring surveys.

In this study, we aim to assess the performance of three of the most popular metabolic inference methods, namely paprica, Picrust2 and Tax4Fun2, against metagenomics and environmenntal data, in the context of salmon farm benthic surveys. In particular, we are interested in 1) comparing the taxonomic and functional diversity recovered from both eDNA metabarcoding and metagenomics, 2) assessing their sensitivity towards fish farm activities in terms of microbial turnover and correlation with environmental data, 3) comparing the accuracy of functional inference methods against metagenomics data, and 4) identifying poorly assigned functions and taxonomic groups with the aim to improve future surveys. While we anticipate the the presence/absence and relative abundance of the inferred gene families to approximate those observed from metagenomics, we expect taxonomic and functional diversity of the metabarcoding data to be substantially higher due to its sensitivity towards less abundant taxa, and therefore be at least as responsive as metagenomics to detect anthropogenic impacts.

TRACKING CHANGES IN MICROBIAL AND MEIOFAUNAL COMMUNITIES AROUND COASTAL FINFISH AQUACULTURE SITES USING ENVIRONMENTAL DNA METABARCODING AND METAGENOMICS

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Finfish aquaculture has increased globally in recent years, raising concerns about the effect on the surrounding environment. Traditional biological monitoring techniques such as polychaete counts and percent coverage of the sulfur-oxidizing *Beggiatoa sp.* are often time-consuming and expensive, therefore eDNA is being investigated as an alternative method for monitoring the biological communities around aquaculture farms. A combined metabarcoding and metagenomic approach was used here to characterize the microbiota and meiofauna in sediments and water column samples collected around a fish farm off the coast of Nova Scotia. To provide a complete overview of the benthic and pelagic environments, changes in both sediment and water column communities were analyzed and compared to determine the main trends of the fallowing process at this specific finfish farm. Sampling for this study took place intermittently from July 2018 – November 2019 at the Cooke Aquaculture facility's McNutt's site outside of Shelburne, NS, which houses between 19-20 cages of Atlantic salmon (*Salmo salar*). During this time the farm was fallowed from approximately November 2018 – June 2019, allowing a separation of the samples into three categories: production cycle 1, mid-fallowing, and production cycle 2.

Results from metabarcoding showed that the water column community was influenced more by seasonal trends in temperature and water column mixing as opposed to the farm production cycle. nMDS plots showed that samples clustered tightly by sampling day regardless of the state of the farm, demonstrating a seasonality to both the prokaryotic and eukaryotic microbial communities. Given the farm site location, close to the open ocean, the strong seasonal cycle occurring in the water column was similar to what is observed for the microbial communities of the coastal North Atlantic Ocean. Unlike the water column, the beta diversity of benthic communities displayed significant differences between the 3 sample types (cage, buoy, barge), which showed a pattern connected to farm production as well as the location on the farm. Both the metabarcoding and metagenomics approaches tracked changes in the microbial and meiofaunal communities as the farm transitioned from production to fallowing, and into the subsequent production cycle. Potential indicator species of the sediment organic loading status were: Monhysterid nematode worms and sulfur metabolizing bacteria *Sulfurovum* and *Desulfobacteraceae*.

THE MONITORING AND EVALUATION OF STOCK HEALTH AND WELFARE IN AQUACULTURE USING SUBMERSIBLE REMOTELY OPERATED VEHICLES

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With the total annual aquaculture production contributing to 45% of the world's aquatic product consumption, the importance of aquaculture on the global food market cannot be overstated. As with any food resource, sustainability and animal welfare is a top priority. Keeping infrastructure and equipment in proper operational conditions and ensuring compliance with hygiene and health standards can have critical challenges.

The use of submersible remotely operated vehicles (ROVs) is changing how fisheries are monitoring and maintaining their farms. With an ROV, operators can get eyes underwater quickly to easily inspect and monitor both the farm and the stock.

There are numerous ways in which an ROV can work to monitor the health and welfare of fish on farms. Net health is absolutely imperative to the overall health of farms and stock. With an ROV users can quickly and accurately inspect nets without needing to schedule costly divers. With consistent inspection operators can ensure that their pens are structurally sound and catch minor issues early.

Observing fish behaviour is important as farmers can spot unusual behaviour quickly. By catching odd behavioural patterns early, operators can nip issues in the bud quickly to minimize negative effects. Feeding processes can also be observed and assessed using an ROV. Monitoring feeding processes allows operators to confirm that their processes are as effective as they can possibly be. Retrieving morts quickly allows operators to get effective insight as to how and why fish are being lost and at what life stage. Sampling tools allow users to retrieve water and sediment samplers to ensure quality parameters are being met.

In short, the use of ROVs provides operators with an efficient, safe and effective way to ensure stock health and wellness.

AN OVERVIEW OF GROWTH RATE, BONE DEVELOPMENT AND MINERAL COMPOSITION OF THE CLEANER FISH BALLAN WRASSE *Labrus bergylta*

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The increasing demands for cleaner fish in salmon and trout farming have resulted in intensified production of ballan wrasse (*Labrus bergylta*) juveniles. Understanding ontogeny, feeding biology, and nutrient requirements of this species is essential to provide appropriate feeds to support fish performance and improve the efficiency of wrasse juvenile production. Farming of ballan wrasse has been hampered by big variations in growth rates, high prevalence of skeletal deformities and other welfare related pathologies. It is known that the larval period has fast growth rates to obtain larger-sized bodies which have higher chances of survival. The growth rates then reduce in the juvenile stage. Only few studies report growth rates of the larval and juvenile wrasse. Furthermore, these studies do not give a complete picture of how the growth rates vary according to the developmental stages of the fish. There is one report describing the development of the skeletal system in the early stages (15 – 61 dph) of ballan wrasse. Although the head skeletal system seems to be fully developed at 0.4 – 0.5 g, this structure will still change along with the increase in body weight afterwards. Understanding how minerals related to bone formation vary through early life stage, is a fundamental step in studying mineral requirements and, thus, allows production of appropriate feeds to minimize the incidence of skeletal deformities in commercial wrasse production. This presentation provides an overview of growth rates and mineral composition of wrasse from 0.1 to 50 g, and anatomy of the head skeletal system from 0.9 to 15 g wrasse. This work is an attempt to fill some of the major gaps in the knowledge of growth rate, bone development, and body composition at different ontogenetic stages of ballan wrasse.

THE MEASUREMENT OF STEROID HORMONES IN NORTH AMERICAN ATLANTIC SALMON *Salmo salar* SKIN MUCUS USING LIQUID CHROMATOGRAPHY TANDEM MASS SPECTROMETRY

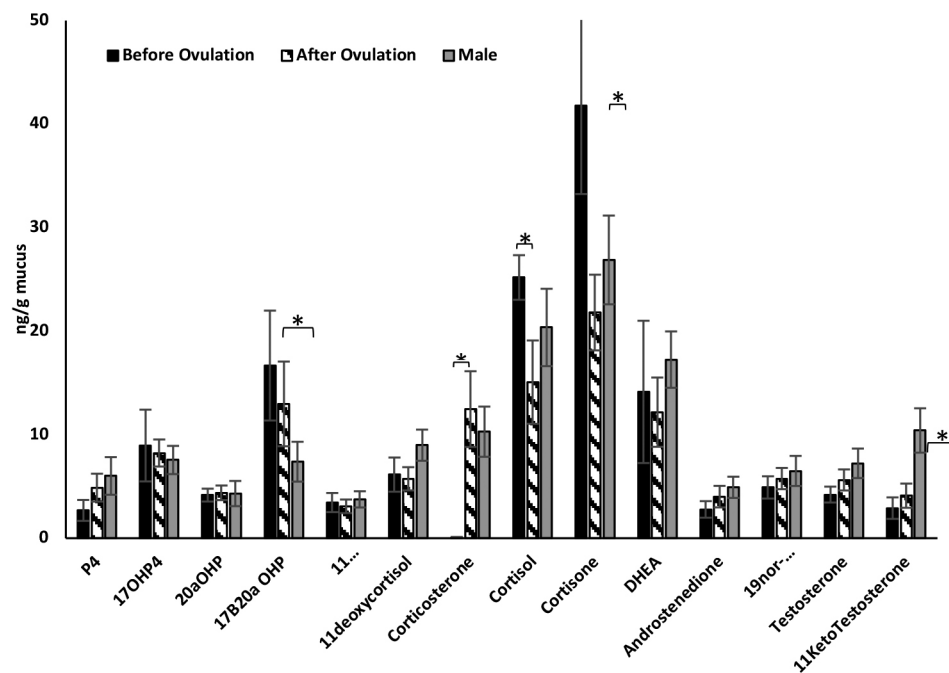
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The use of steroid hormones as biomarkers provides a snapshot of physiologies indicative of stress or reproductive status. However, in fish, sampling tends to be invasive, utilizing blood or tissue samples that are difficult to collect and stressful for all involved. Fish mucus, a complex material that contains a broad range of small molecules, is an option for steroid hormone measurements. The primary goal of this study is to define mucus as an appropriate matrix for the measurement of steroid hormones. Mucus was collected from both male (n = 13) and female sexually mature North American (NA) Atlantic salmon through absorption onto filter paper. Females were sampled before ovulation (n= 5) and after ovulation (n = 7). All samples were subjected to a methanol crash and analyzed with a liquid chromatography tandem mass spectrometry (LC-MS/MS) method.

Male fish had significantly more 11-keto testosterone (p ≤ 0.05) while female fish from both groups had significantly more 17, 20 hydroxyprogesterone (17,20 OHP, p ≤ 0.05). Glucocorticoid concentrations were significantly different between the two female groups sampled. Females before ovulation had significantly more cortisol and cortisone (p ≤ 0.05) concentrations when compared with females after ovulation. However, corticosterone concentrations for females before ovulation were significantly less than females after ovulation (p ≤ 0.05).

These data validate the use of skin mucus as an appropriate matrix for steroid hormone analysis. They also suggest an important role for glucocorticoids during ovulation in NA Atlantic salmon. These initial studies provide a basis for the future analysis of fish reproductive endocrinology using skin mucus and LC-MS/MS technologies.



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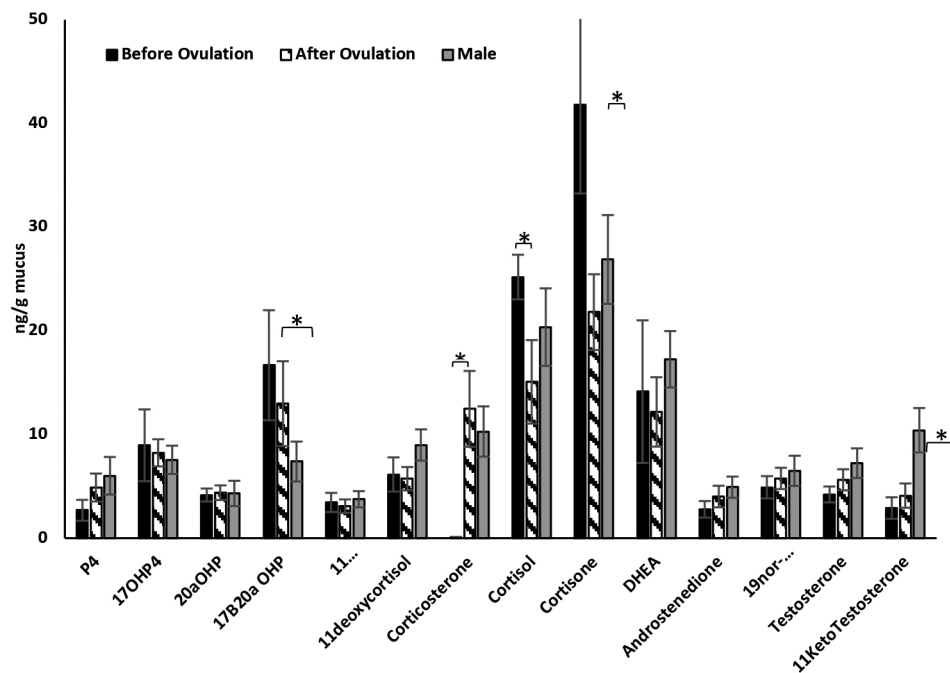
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COLLABORATIVE AQUACULTURE AND COASTAL SUSTAINABILITY RESEARCH: SUCCESSIONS, LESSONS LEARNED, AND CONSIDERATIONS FOR FUTURE ADVANCEMENT

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Limitations in science capacity needed to support sustainable aquaculture development in Nova Scotia resulted in the formation of the Centre for Marine Applied Research (CMAR) in 2017. One of the most pressing data gaps was the lack of coastal water quality data, that was needed to support site decisions. This was the genesis for the development of CMAR's Coastal Monitoring Program which began collecting high resolution, water quality data at biologically relevant scales. To ensure data access for all, CMAR applied FAIR data principles (Findable, Interoperable, Accessible, and Reusable), to ensure our marine data is freely available on the Nova Scotia Open Data Portal, the Canadian Integrated Ocean Observing System, and in summary reports on our website. Beyond aquaculture, these data have been useful to other marine users and CMAR now supports a diverse range of fisheries and environmental research, such as climate change vulnerability assessments, technology development, and carrying capacity modelling. An essential ingredient for developing these research projects and others, has been collaboration. The complexity and resource demands of modern-day aquaculture research, makes it highly challenging for any one entity to operate in isolation. Coastal sustainability is a priority for governance, academia, industry, First Nations, and non-for-profit and CMAR has been fortunate to collaborate with all these stakeholders. This presentation discusses our experience with successful collaborative research approaches, lessons learned, and considerations for future advancement.

USING eDNA METABARCODING TO CHARACTERIZE MARINE FISH ASSEMBLAGES AND MICROBIAL COMMUNITIES

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eDNA metabarcoding, designed to characterize complex communities, is being applied to answer a wide range of resource management questions by the NOAA Northeast Fisheries Science Center. Finfish communities were described over 3 years using eDNA from sampled water near oyster farms in Long Island Sound (Fig 1, doi:10.3389/fmars.2019.00674). Microorganisms growing on aquacultured sugar kelp were compared with those in surrounding seawater, and indicator species important to sugar kelp ecology and health were identified (Fig 2, doi:10.1016/j.algal.2022.102654). A pilot project analyzed eDNA in seal feces to identify fish consumed by grey seal and harbor seal. A NOAA Fisheries Strategic Initiative is using ongoing eDNA sampling over the northeastern continental shelf to advance this technology for living marine resource assessment. These projects are summarized and synthesized with respect to the potential benefits and challenges of using eDNA for the management of living marine resources.

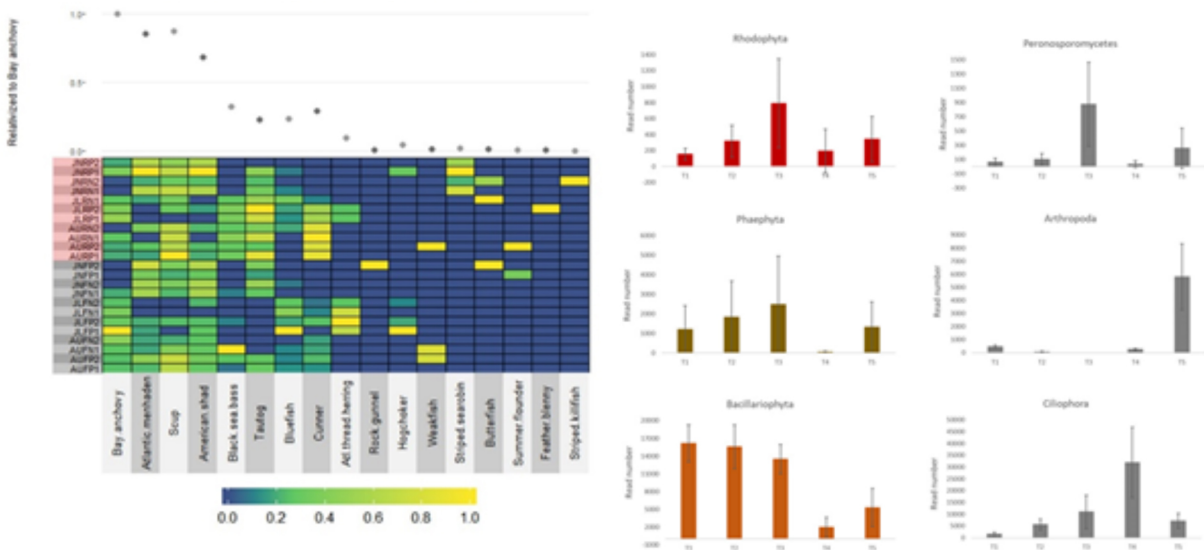


Fig 1 (left) Relative abundance of 17 fish taxa at a natural reef site (upper half highlighted in pink) and an nearby oyster farm with bottom cages (lower half highlighted in grey).

Fig 2 (right) Read numbers of blade indicator epiphytes (left: Rhodophyta, Phaeophyta, and Bacillariophyta), a potential kelp pathogen (right: Peronosporomycetes), and grazers (right: Arthropoda and Ciliophora).

EXPLORATION OF LENGTH-WEIGHT RELATIONSHIPS IN SCALLOPS

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A fishery stock assessment provides decision makers with the information necessary to make reasoned choices. (eg. maximum sustainable yield)

Length weight relationships (LWRs) are an essential component of fishery stock assessments, since they provide information about the growth, their general well being, and fitness in marine habitats. Many researchers have shown that LWRs differ among fish species based on physiological factors (eg. maturity) and biological factor (eg.sex).

Scallops play an important role in fishery markets in Atlantic Canada. They contain a huge amount of nutritive protein and other essential nutrients, such as Omega-3. Many health magazines claim that scallops can be used as a treatment for conditions like diabetes and indigestion.

However, in order to maintain the sustainability of fishery, industries need to avoid catching too many immature small scallops at the same time and reduce times spending in using trawl gear to dig into the sea floor. Also, the growth overfishing also can cause economic losses for fishery industries

Here, we want to explore how the various factors will affect LWRs in scallops in Nova Scotia. Our goal is to propose a statistical model that can predict the scallop meat weight of a given size scallop based on.

MOLECULAR POTRAIL AND IMMUNE MODULATIONS OF IL17C DUE TO PAMPS AND BACTERIAL CHALLENGES FROM *Amphiprion clarkii*

D.S. Liyanage, Sukkyoung Lee, Tae Hyug Jeong, Hyerim Yang, W.K.M. Omeke, Chaehyeon Lim, Kishanthini Nadarajapillai, H.M.V. Udayantha, W.M. Gayashani Sandamalika, and Jehee Lee

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Interleukin 17C is a cytokine that plays a vital role in the innate immunity of the epithelium, including intestinal bacterial pathogens, in an autocrine manner. It stimulates the production of antibacterial peptides and pro-inflammatory molecules for host defense by signaling through the NF κ B and MAPK pathways. Acts synergically with interleukin-22 in inducing the expression of antibacterial peptides. Synergy is also observed with TNF and IL-1 β . Depending on the type of insult, it may have both protective and pathogenic properties, either by maintaining epithelial homeostasis after an inflammatory challenge or by promoting inflammatory phenotype. Enhanced IL17C/IL17RE signaling may also lead to greater susceptibility to autoimmune diseases.

In this study, we performed the bioinformatics analysis, spatial and temporal expression analysis of the *IL17C* from the *A. clarkii*. In brief, the bioinformatic analysis done with several bioinformatics tools and the spatial expression analysis performed using the healthy *A. clarkii*'s using 12 different tissues. The temporal expression analysis experiment conducted with the poly I:C, LPS, and *Vibrio harveyi* and collected the blood at 0, 6, 12, 24, 48, and 72 post-injection intervals.

In-silico analysis revealed the identified sequence containing 492 bp ORF and 163 amino acid residues. The molecular weight and the theoretical isoelectric point are 17.9 kDa and 8.83 pI, respectively. The IL17C sequence containing a signal peptide. Considering the domain structure of the IL17C protein, it includes the primary IL17 domain. According to the identity and similarity analysis, the *A. clarkii* showed a closed relationship with the *Amphiprion ocellaris* with 81.6% identity and 79.8% similarity.

The tissue-specific analysis of *IL17C* was performed under normal physiological conditions. According to that, the gills, skin showed the highest mRNA expression where the liver showed the lowest mRNA expression from the selected tissues. Further, the immune-modulatory transcription analysis performed using blood with the PAMPs and bacteria challenges. According to that, the *IL17C* showed upregulated expression for all stimuli at 6 h and Poly I:C and LPS at 24h.

qPCR results showed the *IL17C* transcript in the blood was upregulated at early time points, and it may be due to host defence mechanisms and innate immune responses of the *A. clarkii* IL17C. Further, the *IL17C* can be up-regulated by bacterial and PAMPs stimuli. This up-regulation may be mediated by Toll-like receptors TLR2 and TLR5. Also, they were up-regulated by various proinflammatory cytokines, including TNF, IL1B, and IL17F. Therefore, it may be concluded that IL17C can effectively participate in immune-modulatory responses and host defence mechanisms by activating several pathways.

REFERED MATERIALS IN ARTIFICIAL HIDES FOR LUMPFISH

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Research report

HDPVC – Gathers more lumpfish

Lumpfish prefer thicker kelp materials.

The experiment was carried out in two farming sites with autumn smolt (October release 2018, average weight: 125 grams, 150,000 salmon in each pen) at the Bjørøya site Kraakholmen.

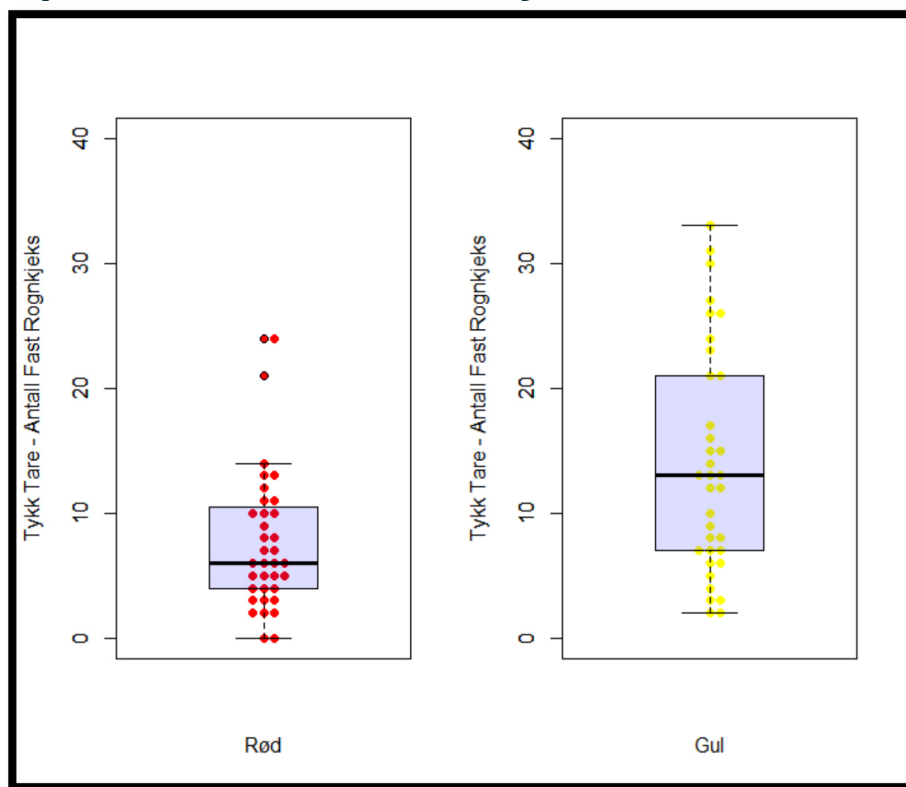
Key findings:

- Crane Kelps that did not follow the wave dynamics housed most resting lumpfish.
- Preferred thicker kelp shelters.
- Lumpfish was distributed evenly down through the kelp shed.

Weather (from almost windless to full force gale) had little effect on ammount of lumpfish in the kelps.

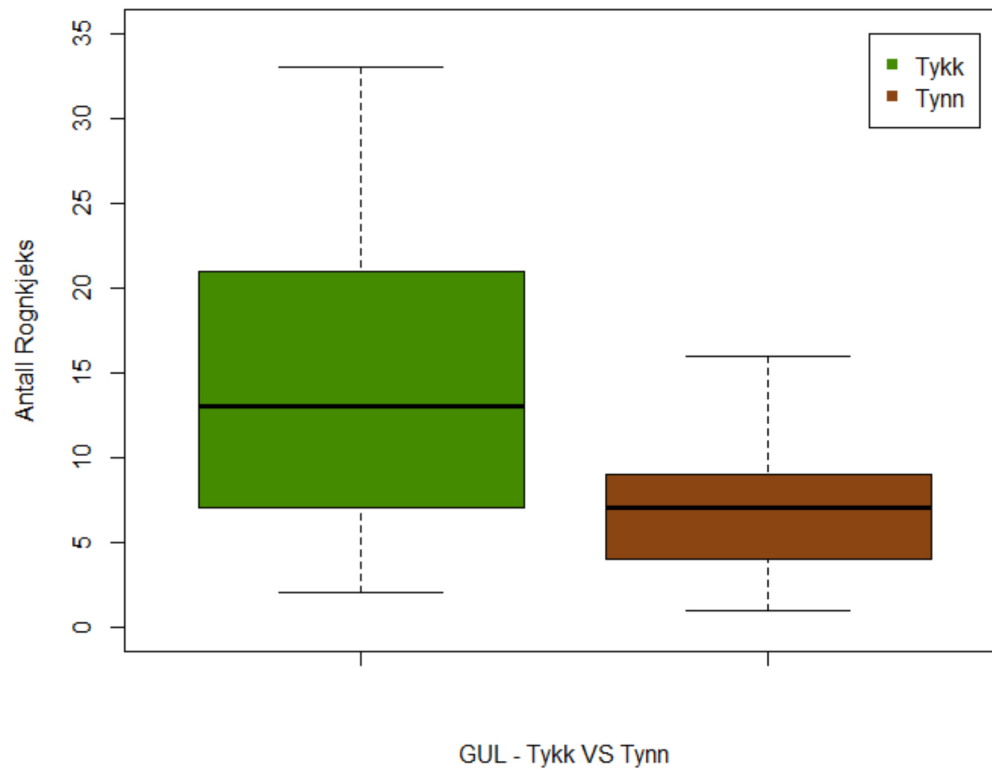
RESULTS

1. Kelp that follows the waves with a air filled floater (red) gets less lumpfish than cork floats (yellow).



(Continued on next page)

2. Lumpfish prefer thick kelps over thin ones.



DIET-INDUCED TAURINE DEFICIENCY INCREASES THERMAL TOLERANCE AND HYPOXIA SENSITIVITY IN BROOK TROUT: CARDIOVASCULAR MECHANISMS

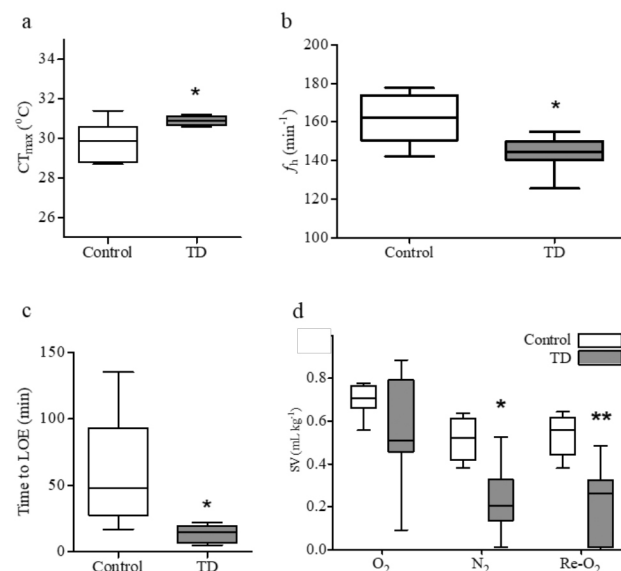
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Taurine is a beta amino acid found in high concentrations inside vertebrate heart cells where it functions primarily as an osmolyte. Mammals tightly regulate cellular taurine levels and deficiency is associated with cardiomyopathy and disruptions in energy metabolism. Taurine levels are far more dynamic in freshwater fish but the impact of taurine fluctuations have not been assessed. We developed a taurine-deficient (TD) brook trout model system using an experimental diet rich in a natural competitive inhibitor of taurine uptake. Cardiovascular function limits environmental stress tolerance in fish so we hypothesized that TD animals would exhibit impaired cardiac function and increased sensitivity to acute thermal and hypoxic environmental stress.

Heart taurine concentration decreased by 21% after 4 weeks with the experimental feed. TD fish were less sensitive to acute thermal stress (Fig 1a) despite having a lower maximum heart rate at high temperatures. TD fish were more sensitive to acute hypoxia *in vivo*, and this was mirrored *in vitro*, as cardiac stroke volume collapsed in anoxic perfused hearts. Mitochondria from TD hearts were relatively uncoupled, complex IV function was impaired, and the expression of its COX3 subunit was reduced. High concentrations of intracellular taurine appear necessary to support maximum cardiac function and aerobic metabolism in brook trout. We have demonstrated that a diet additive can uncouple acute thermal tolerance from hypoxia tolerance via changes in cardiac taurine content. Using the mechanisms identified here, we are now designing additive mixtures to rapidly (within days) enhance both thermal and hypoxia tolerance to improve survivorship under intensive culture conditions.

FIGURE 1. Effects of taurine deficiency (TD) on (a) critical thermal maximum (CT_{max}), (b) maximum heart rate (f_h) under thermal stress, (c) time to loss of equilibrium (LOE) under hypoxia, and (d) cardiac stroke volume under anoxia (N_2) and reoxygenation ($Re-O_2$).



AQUACULTURE EDUCATION: CHALLENGES, GAPS, AND IMPORTANCE IN SUCCESS OF SMALL SCALE AQUACULTURE AND AQUAPONICS IN EAST AFRICA

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Small-scale aquaculture and aquaponics farming is growing steadily in East Africa. However, its success and sustainability are short-lived, considering that most do not break even or shut down a few months after starting. The study examined the status of aquaculture and aquaponics education in East Africa, focusing on its role in the success or failure of small-scale farming, gaps, and the resultant challenges. The study was quantitative. The research disclosed a significant aquaculture and aquaponics education gap. Inadequate and poor education is primarily to blame for the high failure rates. Most respondents confused aquaponics with hydroponics. Also, the knowledge and training are limited to tilapia, sardines (omena), salmon, and catfish farming. The results revealed four categories of education gaps and challenges.

The first category is the lack of requisite education on the infrastructural set-up of aquaculture and aquaponics production systems. As a result, farmers set up wrong systems or use inappropriate procedures and materials. For example, farmers had little knowledge of integrated recycling systems and semi-closed systems. The majority adopted floating cage systems because they did not understand available alternatives. Secondly, farmers lacked education on efficient aquaculture farm management. Professionally set up fish farms deteriorated in months after starting operations due to poor management. Lack of or inadequate training on the suitable fish to rear, feeding, harvesting, hatchery, safety, and water management contributed to fish deaths and low yields. Thirdly, farmers lacked entrepreneurship education and skills to profitably run the aquaculture/aquaponics farm. Aquaculture farmers in East Africa lack adequate training in supply chain management, costing, budgeting, supply and demand planning, and marketing. This education is essential in running an efficient, market-driven, and sustainably profitable aquaculture/aquaponics farm. Lastly, there is inadequate education among consumers on the nutritional value of fish, thus affecting consumer demand.

Overall, the study revealed a high demand for specific fish types such as catfish and tilapia and low demand for sardines due to overproduction. Generally, farmers lacked the critical education needed for profitable and sustainable aquaculture. Farmers had little knowledge of aquaponics. Also, most education institutions with aquaculture courses were mainly undergraduate and postgraduate degrees, thus, revealing a significant gap in diplomas certificates and apprenticeship training for interested farmers who do not meet university degree requirements. For instance, Kenya had one non-degree aquaculture academy under Food Tech Africa Consortium in partnership with two fish farms offering non-accredited practical aquaculture training. There lacked a higher education institution providing aquaponics as a course in East Africa. The study disclosed that education is critical for aquaculture and aquaponics farming and recommended introducing certificate and diploma courses in East Africa to support small-scale aquaculture/aquaponics. However, private and public sector collaboration is crucial to bridge the high education gap through offering small-scale education to farmers on aquaponics and aquaculture trends, considering that most of these courses are provided by private non-college organizations.

CHARACTERIZATION AND COMPARATIVE GENOMIC ANALYSIS OF *Vibrio anguillarum* REVEALS SIGNATURE PATHWAYS OF THE O-ANTIGEN BIOSYNTHESIS IN SEROTYPES O1, O2 AND O3

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Vibrio anguillarum serotypes O1, O2 and O3 are the most frequent pathogens of fish world-wide. Genetics differences between serotype that could give light on the evolution of this marine pathogen are unknown. Here, we fully sequenced and phenotypically characterized a *V. anguillarum* O1 isolated from a winter Steelhead trout (*Oncorhynchus mykiss irideus*) in British Colombia, Canada and compared it to a serotype O2 isolate. *V. anguillarum* O1 strain caused vibriosis and 100% mortality in naïve lumpfish (*Cyclopterus lumpus*). Its genome contains 2 chromosomes (3,13 Mb and 1,03 Mb) and 2 typical serotype O1 pJM1-like plasmids (65,573 and 76,959 bp). Furthermore, *V. anguillarum* O1 displayed resistance to Colistin sulphate which differs from serotype O2 that could be attributed to the presence of the *ugd* gene in serotype O1.

Comparative genomic analysis among the serotypes showed that genetic heterogeneity in the O-antigen biosynthesis gene cluster is characterized by gene absence and presence of unique genes. Serotype O1 is composed of 22 O-antigen biosynthesis genes (*rfbA-H*, *galU*, *ugd*, *galE*, *gae*, *wecA-C*, *wecE*, *manA-B*, *glmU*, *wbpM*, *wbpV*, *wbpI*), whilst serotype O2 is composed of 13 O-antigen biosynthesis genes (*galU*, *galE*, *manA-B*, *wecB-C*, *glmU*, *wbpE*, *wbpI*, *wbpM*, *wbpO*, *wbpA*, *wbD*) and serotype O3 is composed of 13 O-antigen biosynthesis genes (*galU*, *galE*, *rfbA-B*, *manA-B*, *wecB-C*, *glmU*, *wbpM*, *wbpO*, *wbpP*, *wbpV*).

Serotype O1 utilizes the Wzx/Wzy-dependent pathway for O-antigen translocation and polymerization, while the serotypes O2 and O3 employs an alternative pathway. Insertion sequences play a role in the evolution of the O-antigen gene cluster via recombination events. This heterogeneity possibly results in differences in immune evasion mechanisms employed by the respective serotypes. Our work will contribute towards understanding pathogenesis of *V. anguillarum*, impacting disease control and vaccine development against the pathogen.

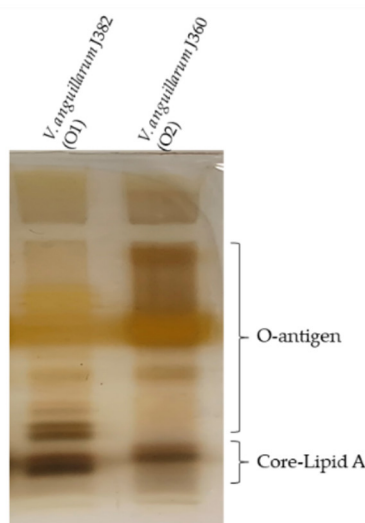


Figure 4. Comparison of lipopolysaccharide proteins in *V. anguillarum* J382 (serotype O1) and *V. anguillarum* J360 (serotype O2). A volume of 8µL of sample was loaded on 15% gel and proteins were separated at 120V for 120 mins.

IMPACTS OF SEAWATER PH BUFFERING ON LARVAL PACIFIC OYSTERS AND REPERCUSSIONS FOR SUSCEPTIBILITY OF LATER LIFE STAGES TO SUMMER MORTALITY STRESSORS

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Ocean acidification and the resulting lowered aragonite saturation state of seawater have been linked to high levels of larval mortality in commercial Pacific oyster hatcheries. In response, many major oyster-seed producers along North America's west coast have resorted to seawater pH buffering via the addition of soda ash. Whilst this may improve survival during larval stages, the repercussions of larval rearing under soda ash conditions for later life stages, particularly under stressor conditions, remain unknown. At the same time, shellfish growers who rely on seed from these hatcheries continue to observe high levels of summer mortality in commercial oyster stocks despite concentrated research efforts to pinpoint a cause and develop solutions.

In order to investigate the potential impacts of soda ash buffering on larvae and knock-on effects for later life stages, in terms of susceptibility to summer mortality stressors, Pacific oysters were reared under soda ash pH-buffered or ambient pH seawater conditions for the first 24 hours of development. At one day post-fertilization, all larvae were placed under ambient pH conditions for the remainder of the larval developmental period. Larval survival, growth, microbiome, and immune status were assessed across development. Spat (3-month) and adult (18-month) oysters arising from the larval run were used in laboratory-based challenge experiments to investigate carry-over effects to later life stages during stress events. Survival and immune status were assessed over 3–7-day exposures to the pathogenic bacterium *Vibrio aestuarianus* under high temperature (24°C).

Buffering resulted in an altered larval microbiome and animals reared under soda ash conditions as larvae showed increased mortality, as both spat (Fig. 1) and adults, compared to those reared in non-buffered seawater. Results suggest that impacts to the larval microbiome may compromise later life stages. We propose that while soda ash buffering may serve as a short-term solution for hatcheries, it may also be contributing to summer mortality of Pacific oysters.

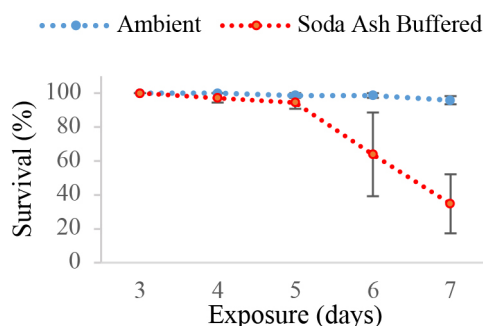


Fig. 1. Mean \pm SD survival (%) under *Vibrio aestuarianus* exposure and high temperature (24°C) of Pacific oyster spat following larval rearing under ambient (blue dots) or soda ash buffered (red dots)

CRAB BODY PYROLYSIS: CHARACTERIZATION AND APPLICATIONS OF CRAB BIOCHAR “A CRABBY SOLUTION”

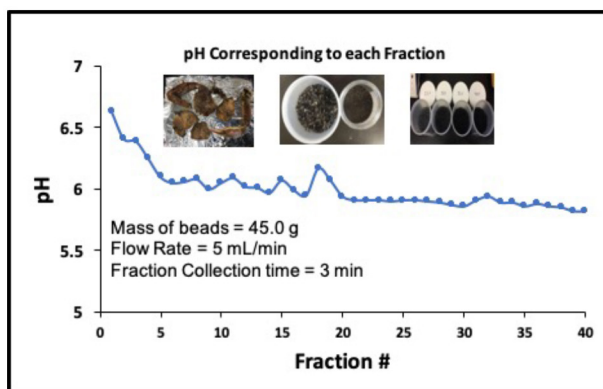
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Nova Scotia crab harvesters sell over 5 million lbs of Snow Crab (*Chionoecetes opilio*) annually. The commercially desired product are the legs and shoulders generating resultant waste streams from bodies of the snow crabs (approximately 1/3 of the crab). Currently this waste is landfilled which is costly and fossil fuel intensive. There is a desire to find a more environmentally sustainable practice to divert this organic animal waste from NS landfills. In a landfill, snow crab residues will decompose and generate some small amount of fixed carbon, however much of the carbon is released into the environment as CO₂ during decomposition and aside from some microbial benefits none of the remaining interesting chemicals are utilized during landfill decomposition. The chemical composition of the snow crab includes a high content of protein (34.2% dw) and essential amino acids; they also have fat (17.1% dw), with a high proportion of ω3 polyunsaturated fatty acids and approximately 28.5% dw minerals (calcium, phosphorous, and magnesium) making this waste stream very intriguing as a starting biomass for the generation of biochar. In this paper we have determined the optimal pyrolysis conditions and highest yield for the char generated from the crab body waste stream. The chars have been fully characterized and we have investigated several applications ranging from neutralization material for acidic waters to concrete additives and catalysis.



MODERATE EXPRESSION OF B-CELL LYMPHOMA-2 (BCL-2) AGAINST DIFFERENT IMMUNE STIMULANTS IN *Amphiprion clarkii*

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Amphiprion clarkii is being increasingly used as a prominent ornamental fish in Korea. However, recently it has been greatly hindered by frequent pond stresses like destructive diseases. Bcl-2, an apoptosis regulatory gene that is involved in immune responses has not well investigated in anemonefish including *A. clarkii*. As apoptosis is a universal response to all stressors, the present study annotates the Bcl-2 gene in *A. clarkii* genome and examined its roles in response to disease infections. *In silico* study of *A. clarkii* Bcl-2 (AcBcl-2) highlights that this 23 kDa protein contains 208 amino acids with p.I of 5.04. Unchallenged tissue expression analysis with qPCR was shown a remarkable increase in the expression of AcBcl-2 in the brain. In the immune challenge results, the expressions of the AcBcl-2 were significantly upregulated after exposure to three different stimuli; Poly I:C, LPS and *Vibrio harveyi*. Late expression of AcBcl-2 was observed in both blood and spleen of *V. harveyi* challenged samples. LPS and the *V. harveyi* had a similar expression pattern. Obtained expression patterns of AcBcl-2 indicate its involvement in the stress response and apoptosis regulation. The significant upregulation of AcBcl-2 at 24 post-injection in the blood of poly I:C challenge samples may be declaring probable antiviral nature. The above collected data shows the moderate expression of AcBcl-2 upon different challenging and tissue conditions, especially significant expression with the presence of Poly I: C. The pattern of the expression of AcBcl-2 after the immune challenge varied in the case of extent and time-span of up-regulation, indicating there may be distinct apoptotic signalling in response to different forms of cell injury caused by these stimuli.

IMPLEMENTING STRATEGIES FOR SUSTAINABLE DEVELOPMENT OF COASTAL AQUACULTURE IN IRAN

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Due to the climatic conditions of Iran and the division of this country as an arid and semi-arid region (except coastal areas), it reminds us that most aquatic development activities in the country's inland waters due to the great need for fresh water can practically not last long and stable in inland waters. In fact, scarcity of fresh water resources, increase in population and increasing need for fresh water for human and industrial use, as well as water sales with emphasis on economic competition of consumers, are all issues that greatly increase the risk of aquatic development in Iranian inland waters.

Increasing in technological advancement and economization of new methods of coastal aquaculture, as well as breeding in cages and pen systems, can all be considered as a sustainable and long-term development if they have feasibility studies and observe all environmental points (Eco-friendly).

To produce aquatic products and help provide part of the country's food security are of particular importance. Unfortunately, despite the hundreds of kilometers of northern shores of the Caspian Sea, as well as the coastal region of the Persian Gulf and the Sea of Oman and the southern islands, this strategy is still not of particular importance. Focus on coastal development should be considered as the main strategy for sustainable aquaculture development.

FEASIBILITY OF STURGEON CULTURE IN CAGES FOR STOCKS RECOVERY AND STURGEON PRODUCTION

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Nowadays entrance types of pollutions into basin Caspian Sea have created numerous problems for fish stocks such as sturgeon fish. Based upon worldly statistics and accomplished researches, sturgeon fish are in danger of extinction.

In spite of activities and efforts in fisheries organization, unfortunately the crisis of sturgeon fish still has remained. Besides entering kinds of pollutions into Caspian Sea, other various factors like sturgeon fish contraband, irregular fishing and lack of rules for accurate execution have been caused stocks reduction of sturgeon fish in Caspian Sea.

In this search is engaged about sturgeon culture in cage. Whereas aquaculture in cage has improved in the world at last decade. So it is essential to survey different dimensions of this culture new industry. cage culture is a new method for sturgeon fish production in fishponds by the exploiting of the traditional fish ponds with intensive fish production in cages makes possible for the traditional sturgeon farmers to increase their production capacity, diversify the cultured species and recycle the nutrients within the production systems.

This search intends to study other dimensions of this matter like disadvantages and advantages. This present search suggests application of cage culture modern systems for purpose of sturgeon conservation and recovery.

GREENHOUSE THEORY AND CLIMATE CHANGE BASED ON WELTENMEIER THEORY AND MATHEMATICAL MATRIX AND DETERMINATION OF THE DAMAGE PERSPECTIVE ON THE COASTAL STRUCTURE OF MARINE BIOLOGICAL RESOURCES

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The effects of climate change on the distribution of marine organisms and marine ecology can be estimated as follows. In the approximate mathematical matrix of the status of different indicators based on the current situation, the short-term perspective includes a period of 1 to 3 years and the long-term perspective includes 5 to 10 years. Quantitative determination of the index can be calculated based on the numerical range of +2 to -2. In this numbering, the range of changes based on the amount of damage to the environment is examined.

According to the results obtained in this study, it can be pointed out that the status of greenhouse effects can cause severe damage to the marine ecology over time, and if these changes do not change from negative to positive, it can Cause disruption of the living conditions of the region. In the matrix method, it can be seen that this range of changes in the current situation, which includes nearly two decades of changes, is +7, but in the short run, this amount has reached 5, which indicates an increase in the rate of harmful effects in the region. is. Also, with the continuation of these changes in the long run, it has reached -15, which is defined -20 as the maximum loss of the entire marine ecology.

It is proposed to take measures to reduce the changes in the greenhouse phenomenon in the country in relation to the following: Establish an organization or center to measure and receive recordable information on greenhouse changes in the country in water resources. These changes should be evaluated in the short term and by analyzing it, a picture of the current situation should be prepared at different times. Examine all activities that can reduce the amount of negative damage in the aquatic ecology and take action as much as possible to reduce the rate of change.

USING BIOSENSORES TO DETERMINE THE EFFECTS OF WATER POLLUTION IN COASTAL AREAS (CASE STUDIES OF COASTAL ZONE ASSALUYEH)

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In Assaluyeh region, 3 sampling stations have been designated which are located at a distance of 5 km from each other and include West Assaluyeh station, Central Assaluyeh station and East Assaluyeh sampling station. The recorded statistics are related to a 10-year period in three stations during 2001, 2007 and 2011.

According to the 7 tables presented in the results section, it can be seen that based on the sampling of Tables 1, 2, 3 and 4 performed during these years, the number of samples has decreased under the same sampling conditions and time. Also, according to the results of Tables 5 and 6, the gender composition has been disturbed in the balance of the number of males and females during the last 10 years, so that the number of genders, which usually follows the law of 50% male and 50% female in nature, this number gradually Time has become more chaotic.

This shows that unfortunately not much activity has been done to reduce the pollution load during the mentioned years and every year this amount of pollution changes affects the habitat of crabs that are located in the coastal tidal zone and this region to The title is one of the best areas to recognize the increase or decrease of pollution. More research should be done on the type and extent of pollution in the coastal areas of the country, especially in the vicinity of developed industrial sites, in order to better investigate the pollution load status. The oil and gas industry and others need to adhere more closely to environmental standards. Further training should be provided by the Environmental Protection Agency to comply with environmental standards in order to reduce pollution by transferring technical knowledge to personnel stationed in industrial locations.

SUSTAINABLE ECONOMIC VIABILITY OF SHRIMP FARM PRODUCTION IN MEXICO

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Aquaculture has increased its social and economic impact in the world through food production, income generation, as well as the supply of offspring for the repopulation of threatened or overexploited aquatic species (Medina, 2015). According to CONAPESCA (2017). In 2017, Mexico was the seventh largest producer of farmed shrimp in the world. In Mexico, Sinaloa is the leader in national production (37%). Thus, the purpose of the research was to analyze the economic viability of Production Representative Units (URP) of shrimp in the municipality of Ahome, Sinaloa.

The information was obtained in the summer period of the year 2022 in the municipality of Ahome, Sinaloa, through the producer panel technique (Sagarnaga-Villegas et al., 2018). This technique allows obtaining the economic, financial and flow costs net of cash or disbursed. A 70-hectare URP was built for shrimp farming. Detailed information was collected on costs, classified as disbursed, financial and economic, prices, income and performance.

Table 2 shows the costs and income obtained by the URP; the most probable price and yield were considered, \$62 per kg and 1 t ha⁻¹, respectively. In this way, considering the economic costs, the shrimp is produced at a cost of \$58.77 per kg, so that at a price of \$62 per kg, all costs are covered and a profit of 3.23 pesos per kg is obtained.

Table 2. Costs and income generated in the UPR (70 hectares) of shrimp in Ahome, Sinaloa.

URP	Variable	Economic (\$)	Financial (\$)	Cash Flow (\$)
SICMC70	Total income	62,000.00	62,000.00	62,000.00
	Total cost	58,767.28	51,332.46	48,048.57
	Cost per-kg	58.77	51.59	51.33
	Net income	226,240	756,727	976,579
	Income per hectare	3,232.72	10,667.54	13,951.43
	Income kg	3.23	10.41	10.67

Source: Own elaboration from field information, 2022.

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WIKWEMIKONG – OPEN FOR BUSINESS

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Wikwemikong is interested in exploring opportunities in the aquaculture sector. As an Unceded Indian Reserve, Wikwemikong exerts jurisdiction over territorial waters. This presentation outlines the due diligence process, the regulatory authority and steps the First Nation took to foster proposed opportunities for private sector to work with the First Nation.

For Wikwemikong, with 220 kms of shoreline, geographically located within an area of a number successful fish farms, it would make sense to explore further sites. Wikwemikong is open for business, and is open to companies that may wish to take an interest in locating to eastern peninsula of Manitoulin Island.

Wikwemikong aspires for participation in the economic sector and any development that occurs mirrors sustainability principles of concern for not only the financial viability of the business, but society and environment. Our primary driver is for employment creation and exploring opportunities in the supply chain. We have a site, the labour pool and predevelopment activities that may interest private sector businesses in aquaculture.

The studies and projections done to date suggest the recommended site has capacity for a large scale commercial development. With existing farms in the area, it logically resolved that there could be further exploration at opportunities in the entire supply chain.

The community has positioned itself to enter into meaningful discussions with private sector companies wishing to expand, invest, and locate within our area, authorized by the First Nation. To date, the community has completed a shoreline assessment, collection of water temperature data over 2 summer fall seasons, a feasibility study and business plan. The community is in the process of completing capacity development and training plan of members to conclude the environmental baseline study. An environmental impact assessment on the site is close to completion. This community is positioned to initiate meaningful discussions with industry looking to work with the First Nation.

PHENOTYPIC CHARACTERIZATION OF LEPTIN RECEPTOR DEFICIENT RAINBOW TROUT *Oncorhynchus mykiss*

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Leptin is a pleiotropic hormone that is known for regulating appetite and metabolism, and it has been implicated in many other facets of vertebrate physiology. While mammalian leptin elicits anorexigenic effects and acts as an adipostat, research on teleost leptins show varying responses to feed consumption and the hormone is primarily produced in the liver. Most fishes have only one leptin receptor (LepR/LepRA1), however, paralogs have recently been documented in a few species. We revealed a second leptin receptor (LepRA2) in rainbow trout that is 77% similar to trout *lepra1*, and the levels of these receptors are differentially expressed across tissues and during fasting, suggesting there are paralog- and tissue-specific functions.

To further understand how leptin and its receptors might function to regulate growth and energy utilization in rainbow trout, we used CRISPR/Cas9 genome editing to disrupt the leptin receptor genes. Triplicate tanks 150-L tanks (3 WT and 3 Mutant tanks) were stocked with 8-10 rainbow trout (145.9 ± 3.1 g mean body weight, BW). Fish were provided 2/3 ration of feed at 1.25% BW with automatic feeders, after which the fish were hand fed to satiation daily and were sampled at three and six weeks. LepR mutants exhibited a hyperphagic phenotype, which led to heavier body weight, faster specific growth rate, increased viscero- and hepatosomatic indices, and greater condition factor (Figure 1). Muscle glycogen, plasma leptin, and leptin transcripts (*lepa1*) were elevated in LepR mutant fish. A range of hypothalamic genes involved in feed regulation were measured (*agrp*, *npv*, *orexin*, *cart-1*, *cart-2*, *pomc-a1*, *pomc-b*). No differences were detected between WT and mutants except for *pomc-b*, where levels were over 7-fold higher in LepR mutant fish at 3 weeks. This suggests that leptin signaling in the brain is likely mediated in part through *pomc-b*. All detectable fatty acids (FA) were higher in muscle of fed fish mutant fish compared to WT, albeit not significant. However, fasted mutants exhibited significantly lower muscle FA for virtually all FAs, suggesting an increase in FA mobilization during fasting in LepR mutants. These data demonstrate a key role for leptin signaling in lipid and energy mobilization in a teleost fish. Leptin clearly plays a key role in food intake in rainbow trout, however, additional studies are needed on adipose- and glucoregulatory pathways to help better understand the phenotype of the LepR deficiency.

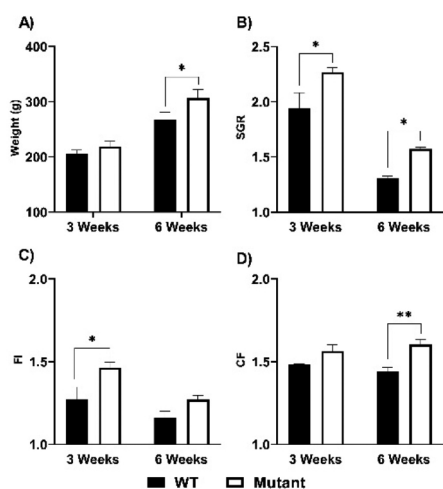


Figure 1. LepR CRISPR/Cas9 edited rainbow trout fed to satiation. WT and LepR mutant fish were sampled at 3 and 6 weeks for: A) body weight (g), B) specific growth rate (SGR), C) feed intake (FI), and D) condition factor (CF).

EFFECT OF TEMPERATURE AND SALINITY ON A TURBOT PARASITE – APPROACHING CLIMATE CHANGE IMPACTS

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Turbot (*Psetta maxima*) production in marine Recirculating Aquaculture Systems (RAS) is relevant to the global aquaculture market (~60 kT in 2016). However, its intensive culture promotes the proliferation of opportunistic pathogens and occurrence of severe diseases like Scuticociliatosis, which is responsible for fish outbreaks. The evolution of such disease may be tailored by abiotic stress factors, which tend to be aggravated under climate change scenarios (CCS). In this context, the previewed water temperature raise can boost pathogens development while simultaneously suppressing the immune system of fish, thereby worsening the already existing problem. On the other hand, salinity levels are also expected to raise in the Atlantic Ocean, thereby reinforcing the need to assess its impacts on scuticociliates reproduction. Hence, this work aims to identify the influence of temperature and salinity variations, under CCS, on the proliferation of *Philasterides dicentrarchi* isolated from a turbot RAS.

The tests were performed in 24-well plates with an initial cell density of 2×10^4 cells mL⁻¹ of *P. dicentrarchi* isolated from turbot in a local saltwater aquaculture unit. A range of temperatures (13 – 21°C) and salinities (10 - 35) were tested in triplicate, both individually and combined. The growth of *P. dicentrarchi* was daily monitored during 8 days of exposure. For that, *P. dicentrarchi* cells were count in a Neubauer chamber, being plotted as average cell densities in function of the exposure time (*i.e.*, growth curves; Fig. 1) or as growth rate.

In particular, higher temperatures enhanced the proliferation of *P. dicentrarchi* during the exponential growth phase, whereas increased salinity levels reduced cell densities. Hence, previewed CCS for seawater temperature represents a threat to turbot aquaculture production if fish infected with *P. dicentrarchi*. However, the use of higher salinities may help to counteract this effect, potentially without significant impacts on turbot development given the wide range of culture conditions supported by this flatfish species.

Acknowledgments: This work was funded by PT2020 (Mar2020 Programme) and the European Maritime and Fisheries Fund (EMFF), under the project MAXIAQUA (MAR-02.01.01-FEAMP-0019). Thanks are due to CESAM (UID/AMB/50017/2019) and to FCT/MEC through national funds, and the co-funding by the FEDER, within the PT2020 Partnership Agreement and Compete 2020. Ana P.L. Costa was supported by a PhD grant (PD/BD/127809/2016) funded by FCT. Catarina R. Marques is funded by national funds through FCT in the scope of the Decree-Law 57/2016, changed by Law 57/2017. We acknowledge the support of Stolt Sea Farm, S.A.

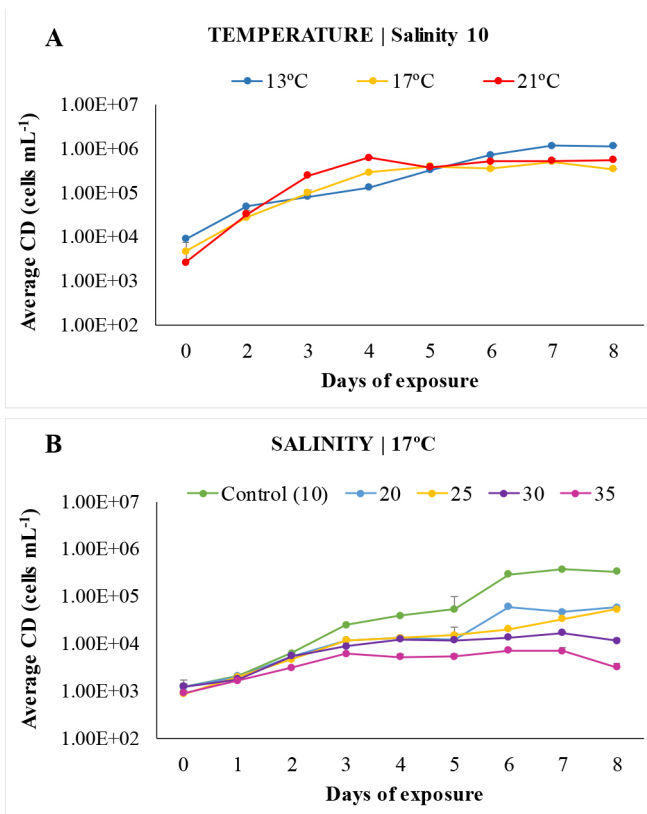


Fig. 1 – Average cell density (CD) of *P. dicentrarchi* exposed to a range of temperatures (°C; A) and salinities (B), along 8 days of exposure. Error bars represent standard deviation.

USING GENE EXPRESSION TO UNDERSTAND FATTY ACID REGULATION IN STRIPED BASS *Morone saxatilis* UNDER DIFFERENT SALINITY AND DIETS

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Following the disappearance of the St. Lawrence population of striped bass *Morone saxatilis*, the Government of Quebec has undertaken in 2002 a major program to reintroduce the species in former habitats. However, to meet the challenges posed by the restocking efforts, a better knowledge of the biology of this species is required, particularly their physiology related to development. We focussed on the effects of diet content in polyunsaturated fatty acids (PUFA) considering their fundamental role in early development. PUFA are almost exclusively synthesized by plants, and although animals can move from one form of PUFA to another using elongation and desaturation processes, very few species can synthesize PUFA *de novo*. PUFAs play an important role in regulating the properties of the cell membrane, serve as precursors for important animal hormones and are therefore essential to maintain animals healthy.

We tested the hypothesis that variable salinity and food source (estuarine vs marine sources) would markedly impact the physiology of striped bass juveniles. Juveniles were raised in winter conditions at two salinities (0 and 14) and fed with a reference diet used as a proxy of an estuarine trophic network (0.73% n-3 PUFA), and a higher n-3 PUFA diet (1.65%) used as a proxy of a marine trophic network. Preliminary results showed that salinity effects were less pronounced compared to those of dietary regimes on total liver and muscle neutral fatty acid (FA) profiles, while muscle polar FA were slightly affected. Also, nutritional essential FA (EPA and DHA respectively) needs were higher when fish were grown in fresh water (FW). These results suggest that FW represented a stressful condition for juvenile striped bass, and this was corroborated by the higher hematocrit value and the lower survival observed in FW. Further work is in progress to investigate how the fatty acid regulation is achieved through the study of the expression of some genes used as a proxy of the fatty acid synthesis (fatty acid synthetase complex *fas* and delta 6-desaturase gene *fads2*), lipid homeostatic processes (peroxisome proliferative activated receptors *ppars*), elongases and desaturases activity (sterol regulatory element binding protein-1 *srebp-1*), and the stress response (molecular chaperones heat shock proteins *hsp70*, *hsp90* and *hsf1* interacting with members of several signal transduction pathways).

BEYOND CERTIFICATION TO QUALITY – THE NEW PARADIGM?

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Certification Standards have been useful tools for the Aquaculture Industry. They have provided a roadmap for all participants to create a level of quality that is acceptable to global markets. Has the time come to move away from certification to a quality centred culture? Is the industry ready for the next stage in this evolution?

In this presentation we will explore certification within the Aquaculture industry, in particular reference to Canada and ask what next? What is the driver for food safety, that ensures the industry spends millions each year on this service.

We will step outside the current compliance framework and challenge whether compliance is enough. If everyone is compliance to a standard, what is left to differentiate a supplier?

We hope you enjoy our vision for the 2020's and how we see the journey developing for the industry, in an interactive, light-hearted review of certification and the future.



COOKE AQUACULTURE CLEANERFISH PROGRAM DEVELOPMENT: LAND-BASED GROWOUT AND TRANSPORTATION

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Lumpfish (*Cyclopterus lumpus*) production continues to expand in Atlantic Canada to meet the growing demand for cleanerfish as part of an integrated pest management strategy. Cooke Aquaculture, as an industry supporter on the Atlantic Fisheries Fund for commercial cleanerfish development, receives juvenile lumpfish from the OSC and continues to grow them out to cage-appropriate size at various facilities. For the 2019-2020 production cycle Cooke Aquaculture had 3 such facilities-the Belleoram facility in Newfoundland, Dalhousie Aquatron and ProNova Marine Services, both in Nova Scotia. Additionally, due to the large geographic distances involved in production there are inherent unique transportation & logistical challenges which will be covered as well. While continuing to expand use and development in NFLD, future plans include expanding cleanerfish use in New Brunswick and Nova Scotia while supporting industry development and cleanerfish deployment in Maine and New Hampshire.

INVESTIGATING HYPOXIA TOLERANCE OF TRIPLOID BROOK CHARR *Salvelinus fontinalis*

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Triploid salmonids are used in aquaculture and stocking programs for recreational fishing because they are reproductively sterile and therefore cannot breed in the wild. However, they appear to be more sensitive than diploids to hypoxia and high temperature stress. The objective of my MSc is to develop a better understanding of the biological requirements of triploids (specifically hypoxia tolerance), with the potential to improve triploid performance.

I compared the acute hypoxia tolerance of sibling diploid and triploid brook charr that were acclimated to either moderate hypoxia (dissolved oxygen at 70% air saturation) or normoxia (100% air saturation) for 7 weeks. Fish then underwent acute hypoxia trials, by slowly injecting nitrogen gas to displace oxygen and using loss of equilibrium (LOE) as the endpoint. Dissolved oxygen levels at LOE (later converted to oxygen tension; kPa), and time to LOE were recorded for each individual fish. Following LOE, all fish were measured (fork length and body mass) and a blood sample was taken. Fish were then euthanized and dissected to remove the whole heart, the ventricle, the liver, the gills, and the gonads. I used the body measurements, blood, and tissue samples to determine condition factor, hematocrit, whole blood glucose and lactate levels, plasma ion concentrations and osmolality, interlamellar cell mass size, relative ventricular mass, compact myocardium thickness, hepatosomatic index and gonadosomatic index. Hypoxia-acclimated fish had a higher tolerance for hypoxic conditions, as indicated by a longer time to LOE and a lower PO_2 at LOE compared to fish acclimated to normoxia, but there was no effect of ploidy on hypoxia tolerance.

Funding acknowledgement: NSERC (Discovery Grant), Mitacs (Research Training Award), NBIF (Research Assistantships Initiative), and UNB (graduate research assistantships).

IDENTIFYING REGULATORY INSTRUMENTS FOR STRENGTHENING CANADIAN AQUACULTURE OCCUPATIONAL SAFETY AND HEALTH (OSH). A COMPARATIVE ANALYSIS OF AQUACULTURE OSH REGULATORY REGIMES IN NORWAY AND CANADA

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Abstract: Globally, significantly less attention has been devoted to documenting and regulating working conditions and occupational safety and health (OSH) in aquaculture, than to regulating food safety and environmental threats in the sector. There are diverse and significant OSH hazards, as well as high rates of injuries and fatalities in aquaculture relative to national/provincial averages. This presentation discusses findings from a review and comparative analysis of OSH aquaculture regulatory instruments and operational standards within Atlantic Canada and Norway. Norway exports salmonid aquaculture research, technologies, organizational approaches and indeed aquaculture corporate ownership worldwide including into Canada. Canada's salmonid aquaculture industry is newer and smaller but poised for rapid expansion. The hazard profiles in Canada and Norway are similar despite the different scale and history of salmon aquaculture in Norway and Canada, but the regulatory regimes for OSH are quite different.

Research Question and Methods: The core research question is: could OSH standards and other tools from Norway be adapted for use in Canada to rapidly and effectively ramp up OSH regulatory practice and capacity given differences in industry scale and regulatory regimes? We explore the question through a review and comparison of the various regulations and standards as well as industry organization and OSH infrastructures (regulatory regimes) in the two countries using available standards and results from key informant interviews.

Findings: Canada's aquaculture industry is much smaller than that in Norway and concentrated in British Columbia and on the Atlantic coast – in some cases in small, remote communities. It is also more mixed with larger shares of shellfish aquaculture than in Norway. Norway has a unitary government, with all operators governed by the same set of regulations and held to the same standards. Norway also has a highly developed OSH regulatory framework that differs in significant ways from the Canadian OSH framework including in its extensive reliance on company-level risk assessment processes supported by substantial and in some cases specialized inspectorate capacity. Norway has developed detailed industry-specific OSH regulations inclusive of requirements for risk assessment of work environments and hazardous activities, standards on the design and installation of net pens, support vessel design, training of personnel and for aquaculture-related diving. Canada has a federal system of government and a complex regulatory regime consisting of both federal and provincial levels of governance for activities that bridge land and marine sites, and variances in both regulations and enforcement agencies between provinces. In Canada, it is difficult to access reasonably accurate data on injuries and fatalities at a national or regional level and there has been very limited research on injury rates, exposures and on prevention. Companies have a general duty to protect the OSH of their workers and to have a safety plan. Atlantic Canadian provinces are very limited in both their scope and capacity to identify, control or eliminate exposures and to monitor the effectiveness of interventions in terms of hazard and injury/fatality reduction. They lack the kind of industry-specific standards and requirements for aquaculture OSH found in Norway. Given the smaller size of the industry and the different regulatory frameworks for OSH, the Canadian marine finfish aquaculture industry might be able to benefit from the rapid transfer and adaptation of some Norwegian tools and practices.

USE OF ACOUSTIC TELEMETRY TO EVALUATE EFFECTS OF AQUACULTURE ON CRUSTACEAN BEHAVIOUR

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Aquaculture production is increasing world-wide, as are concerns about its interactions with the natural environment. Organic loading from farms may attract a variety of animals, including important fisheries species. Little is known about how such species interact with aquaculture farms, although there have been public concerns about potential negative effects on these animals. We provide an overview of acoustic telemetry studies examining the movement of American lobsters (*Homarus americanus*) and Atlantic rock crabs (*Cancer irroratus*) within and around blue mussel (*Mytilus edulis*) and Atlantic salmon (*Salmo salar*) farm sites in eastern Canada.

Lobster movements were evaluated around three mussel farms. Although more abundant within mussel farms than outside of them, lobsters showed no affinity to farms, often leaving the farm areas over short time periods post-tagging. However, lobster movement differed between farm and non-farm areas as movements were slower within farms, perhaps indicating increased foraging behaviour. In contrast, crabs showed a high affinity to mussel farms, rarely leaving farm areas and staying directly below mussel lines.

Lobster movements around salmon farms showed variable results. While lobsters in some soft-bottom sites showed little affinity to farms and left study areas within a couple of days, they seemed ambivalent in other hard-bottom areas and set up territories below and directly adjacent to farms. Where territories were established below or adjacent to farms, lobster home ranges were reduced relative to those established further from farms. Although crabs were more abundant below than away from farms, they seemed to have a somewhat mostly ambivalent relationship to them and showed no attraction or repulsion.

Two experiments were done to assess the impact of acoustic telemetry tagging methods on lobster movement. Lobsters were tagged on the carapace following trapping and released from the surface or *in situ* on the bottom using scuba to attach tags to their “wrists”. Tagging method did not impact behaviour. In general, lobster behaviour seems more a function of bottom type than proximity to farms with animals being transient on soft sediments, but resident on hard and mixed bottoms.



FIGURE 1. Lobster fitted with acoustic tag

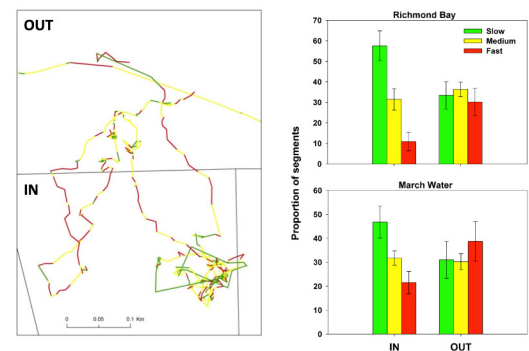


FIGURE 2. Lobster movement below and outside of mussel farms.

FISH TO FISH VARIABILITY IN RESISTANCE To *Saprolegnia parasitica* INFECTION IN ATLANTIC SALMON *Salmo salar*

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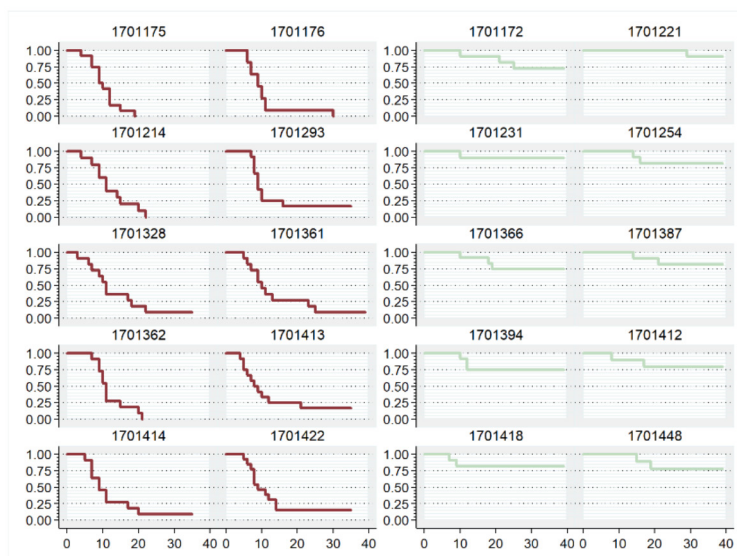
Infections by the Oomycete *Saprolegnia sp.* are problematic to freshwater salmon hatcheries worldwide. Banning use of malachite green oxalate, which was an effective treatment of choice but has carcinogenic and teratogenic properties on humans, with more recent actions to ban formalin use creates a new challenge to control *Saprolegnia* in affected hatcheries. The search to identify new bath treatment options also begs the question as to whether selective breeding might be useful in decreasing susceptibility and mitigate impact on salmon hatcheries. The overall purpose of this research was to phenotypically characterize the variability in resistance/susceptibility amongst families of one year class of Atlantic salmon.

In this study, a controlled *Saprolegnia parasitica* infection model was used to challenge 1158 fish representing 105 pedigreed Atlantic salmon families. Fish were infected in five study tanks and observed for 40 days post-infection to assess a novel lesion score and survival.

Survival analysis of the top and bottom ten resistant families indicated that the hazard of dying following *Saprolegnia* infection was 1509% higher in susceptible families. In all fish, a 10 g increase in weight correlated with a 7.8% increase in the hazard of dying while sex did not affect mortality.

Resistance to *Saprolegnia* infection was estimated to have a heritability of 0.25 ± 0.06 or higher, indicating that selection is possible. The genetic or phenotypic correlations between resistance traits to weight was $\leq 0.29 \pm 0.14$ indicating both freshwater growth and *Saprolegnia* resistance could be selected for simultaneously.

In addition, a novel 11-point fungal lesion scoring method was developed for use on all mortalities and survivors. This scoring method had genetic or phenotypic correlations of $\geq -0.87 \pm 0.01$ with both survival and mortality indicating that the scoring method may be helpful to assess lesion development in studies where mortality is not the primary biological endpoint.



Kaplan–Meier survival curves top (right columns) and bottom (left columns) ten resistant Atlantic salmon families in a *Saprolegnia parasitica* challenge.

ENGAGING THE ANTI-AQUACULTURE CONTINGENT ON TWITTER - AS A PROFESSIONAL: WHY DO IT AND WHAT CAN WE LEARN?

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In today's society, Twitter is a powerful force (along with other forms of social media such as Facebook and Youtube). About 1 in 5 US adults are on Twitter and this has remained consistent over the past several years. The "left" is more likely to use Twitter than the right (32% to 17%). The top 25% of users produce 97% of the tweets. Around 1 in 5 say they have experienced harassing or abusive behavior. 7 in 10 adult Twitter users get news on site and of these 57% say it has increased their understanding of current events.

The anti-aquaculture movement, especially out of British Columbia, has effectively used Twitter and other forms of social media to help mobilize support, resulting in the banning of salmon net pens in certain areas in that province. Several site leases have been cancelled and at the time of this writing, the entire industry is under threat. There is also a halo effect from these efforts that has tainted all of fish farming nationally and internationally. The assault on net pen salmon farming on Twitter has been unrelenting and vicious. There is never a discussion as to quantifying concerns and approaches to mitigating these concerns. All dissent appears to have the singular purpose of cancellation with a scattergun assault of issues seemingly designed to smother any dissenting point of view.

Fearmongering supposed threats that nets pens pose to the environment is standard practice by anti-aquaculturists. It is common to suggest that there is significant disease risk to wild salmon (without any risk quantification) and to promulgate these unsubstantiated claims as headlines in social media. As a veterinarian who has worked with wild and farmed fish diseases for over 30 years, the ignorance of the disease process and the blatant exploitation of the specter of disease is both irritating and concerning. This is further exasperated by peer-reviewed publications in non-medical journals that are elaborate sophistries of unvalidated disease models. These models neglect to incorporate basic variables of disease epidemiology and are merely untested biased hypotheses. However, suiting the purposes of the "anti's", their speculations are paraded in the "Twitterverse" and to politicians as being fact.

This speaker originally took to Twitter to try and learn this platform with a goal to be more of a social media "thought leader" on fish farming, science, and fish veterinary medicine. It has been an interesting journey. Some of these experiences will be presented, including the occasional provoked lapse into some unprofessional utterances. It will conclude with some thoughts on why a professional would bother to engage in such an abusive environment; what can be learned from interactions with the anti-aquaculturists; how social dogma can set in; and speculations on what some of the driving forces might be behind the anti-aquaculture movement.

ASSESSMENT OF TECHNICAL AND ECONOMIC FEASIBILITY OF A PORTABLE AQUAPONIC SYSTEM

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An experiment was conducted in the green house at the Lilongwe University of Agriculture and Natural Resources, Bunda Campus, Malawi, to assess the technical and economic feasibility of a portable aquaponic system made from locally available materials. The experiment used a shallow wooden box (25cm deep and 100cm² area) as a grow bed. Fine gravel was filled into the box as a growing medium. Two plastic buckets (60 litres each) that held water were used as fish tanks. *Oreochromis shiranus* was used as a fish species and lettuce (*Lactuca sativa*) as a crop. The tanks were stocked with 25 fish each with an average initial weight of 8.4g. The fish were fed with formulated pelleted feed containing 30% crude protein. Twenty-one days old lettuce seedlings were transplanted on to hydroponic component. Water was partially (30% of total volume) being pumped twice daily to the hydroponic component by syphoning.

The system sustained life of the fish with a survival rate of 96%. The mean weight gain was 19g and 204g for the fish and lettuce respectively in 42 days. The system showed a positive Gross Profit Margin (52%) for both fish and lettuce. Aquaponics seems to be more commercial and more complex in terms of its technicality but the portable aquaponic system from locally available materials proved to be feasible.



Figure 1 Lettuce grown in a portable aquaponic system

CLEANER FISH RESEARCH AND PRODUCTION IN SUPPORT OF THE ATLANTIC CANADIAN SALMON AQUACULTURE INDUSTRY

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In 2018, the Newfoundland Aquaculture Industry Association (NAIA) through industry consultations identified Integrated Pest Management Strategies for Sea Lice Control as the number one industry priority for Atlantic Canada's finfish aquaculture sector. Potential losses in market value and resistance concerns related to prolonged reliance on any single therapeutant are key drivers that have prompted interest in the utilization of local cleaner fish species such as lumpfish, *Cyclopterus lumpus* and cunner, *Tautoglabrus adspersus*. A multifaceted research team at the Department of Ocean Sciences at Memorial University is conducting research on broodstock domestication and vaccine development as well as standardizing production protocols dedicated to developing this technology for industry use to assist in mitigating and controlling sea lice on Atlantic salmon.

Cleaner fish are fish that provide a service to other species by removing ectoparasites. The feeding behaviour of the cleaner fish is harnessed to create a natural defense for the farm. The success of this strategy depends upon the availability of high quality cleaner fish in sufficient numbers. Salmon farms could possibly require upwards of 10% or more of the salmon population per cage site. Culturing cleaner fish enables a continuous supply of high quality juveniles as demand requires. Cultured cleaner fish also have the added advantage of health screening and vaccination prior to deployment to reduce risk associated with disease transfer.

As the use of cleaner fish continues to produce favourable results globally the demand for these fish increases. Powell et al. (2013) estimates that up to 50 million lumpfish are required globally. The demand for these fish has increased substantially in Atlantic Canada over the past few years (Figure 1) and protocols have been developed and modified in an attempt to meet the demand.

This talk will highlight some of the protocols and technology developed over the past few years in an attempt to meet the demand for cleaner fish in Atlantic Canada.

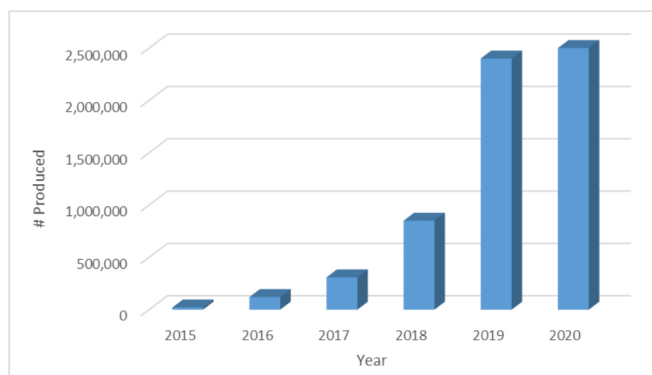


Figure 1: The number of lumpfish produced yearly at the Dr. Joe Brown Aquatic Research Building from 2015-2020.

BIOLOGICAL BIOFOULING CONTROL OF SALMON NET PENS USING THE GIANT RED SEA CUCUMBER *Apostichopus californicus*

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Biofouling of aquaculture infrastructure is a universal challenge faced by both shellfish and finfish growers. Biofouling organisms such as algae, tunicates, sponges, hydroids, and mussels can add significant weight to nets and cages, which must be regularly cleaned to maintain optimal water flow and integrity of these structures. The need to regularly clean biofouling is a time consuming and often expensive process that can be a constraint to industry. Modern cleaning techniques are highly variable and range from direct infrastructure swapping to industrialized power washing that uses high-pressure water to remove the biofouling. The latter is a popular technique in the finfish industry due to its efficacy of removing biofouling, but it is time consuming, costly, and may have negative fish-gill-health consequences that are just being recognized. Development of alternative practices to prevent and control biofouling in finfish net pens is therefore warranted to help offset the time, financial, and potential fish-health costs of current cleaning technologies.

Sea cucumbers like *Apostichopus californicus* have long been recognized for their co-culture potential due to their natural ability to recycle organic-rich sediments into nutrient poor faeces, but little research has examined their ability to control biofouling. We tested the ability of *A. californicus* to consume biofouling material on nets at two organic Chinook (*Oncorhynchus tshawytscha*) farm sites using four treatments: pens with (1) fish and sea cucumbers (SC); (2) fish, but no SC; (3) no fish, but SC; and (4) no fish and no SC. Sea cucumbers were observed feeding on biofouling organisms on the nets in treatment 3 and preferentially consuming excess feed / salmon wastes in treatment 1. Gut-content analysis and biochemical profiles of the sea cucumbers were conducted for treatments 1 and 3. No negative interactions (*e.g.* health and/or behavioural) were detected between the salmon and the sea cucumbers suggesting that this is a good partnership to explore further.



Fig. 1. Sea cucumbers feeding on biofouled nets.

LARVAL SALMON LICE (*Lepeophtheirus salmonis*) EXHIBIT BEHAVIORAL RESPONSES TO PRE-ADULT AND ADULT CONSPECIFIC CUES

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In the larval stage of the parasitic copepod *Lepeophtheirus salmonis*, the free living copepodid must locate and settle on a salmonid host. Chemosensory mechanisms appear to play a role in identifying potential sexually mature mates, yet the full suite of chemical cues and resulting behavioral mechanisms used for conspecific aggregation are unknown. After maturing, evidence has shown pre-adult females and adult male sea lice emit sex pheromones while gravid females reduce their production of these cues. Typically, sex specific cues are used for reproduction and rarely have they been shown to be utilized by larval conspecifics. The aim of this study was to investigate the potential that cues from pre-adult female and adult male sea lice influence copepodid behavior. Behavioral bioassays were conducted with copepodids exposed to water conditioned with three stages of conspecific lice (pre-adult female, adult male, and gravid female), and Atlantic salmon (*Salmo salar*, L.) conditioned water. Experiments demonstrated that copepodids exposed to water conditioned with the salmon host, pre-adult female or adult male sea lice elicited behaviors characteristic of arrestment, whereas sea lice exposed to gravid female conditioned water did not. These results may suggest that *L. salmonis* larvae respond to the cues of lice stages known to produce sex pheromones, and we conjecture that they may serve to aggregate conspecifics and amplify infestations.

COMPARISON OF BIOCHEMICAL PROFILES IN TWO LATIDINALLY SEPARATE POPULATIONS OF *Cucumaria frondosa*

Rachel Morrison, Jean-François Hamel, Jiamin Sun, Annie Mercier

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Extensive research has been conducted on various species of holothuroid (sea cucumber) around the world due to their ecological roles and high economic value as a food product. However, the focus has been on temperate and tropical populations, leaving those located at higher latitudes, such as in the Canadian Arctic, understudied. In specific, the biochemical composition of a species, which can be utilized to understand the value an organism has for human health, is partly determined by the prevailing environmental conditions, which vary latitudinally. Here, various biochemical metrics of the sea cucumber *Cucumaria frondosa*, collected in the Arctic (Nunavut), were assessed and compared to those currently harvested in the Northwest Atlantic (Newfoundland and Labrador). Metrics included, proximate composition, lipids and fatty acids, amino acids, lipid-soluble vitamins, and astaxanthin. As anticipated, the biochemical profiles of the two sampled populations differed substantially. Nunavut-caught sea cucumbers had similar ash and moisture content, but approximately 50% and 17% less total lipids and proteins respectively than Newfoundland sea cucumbers. Nonetheless, the composition and concentration of the different fatty acids present in Nunavut samples highlighted greater proportions of essential fatty acids like DHA and EPA. Nunavut samples also had twice as much proline and vitamin A in their muscle bands and strikingly more astaxanthin specifically in female individuals. Understanding biochemical variability across latitudes is of economic and ecological significance, particularly in the face of climate-related shifts. From an applied perspective, data on the nutritional value of Arctic sea cucumbers can help enhance food security through the development of traditional and commercial fisheries in northern communities.

USE OF CITIZEN SCIENTISTS TO MONITOR HARMFUL ALGAL BLOOMS AND CHANGES IN ENVIRONMENTAL CONDITIONS

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Harmful algal blooms (HABs) occur when species of phytoplankton grow very quickly forming blooms resulting in water discolorations sometimes referred to as red tides. These blooms have been observed in every state resulting in over \$1 billion in losses over the last several decades to communities that rely on recreation, tourism and seafood harvesting. The direct economic impact of HABs does not include the socioeconomic impact from loss of subsistence harvest activities, disruption of cultural practices, water insecurity, food insecurity, and social interaction tied to coastal resource use. The aquaculture industry both shellfish and finfish have experienced direct adverse effects of harmful algal blooms, both toxin producing species and non-toxin producing species. For the individual aquaculture farm, blooms of certain non-toxic phytoplankton are of paramount concern since they are known to cause mortality of shellfish and finfish worldwide.

The National Phytoplankton Monitoring Network (PMN) is a community-based network of volunteers monitoring marine and freshwater phytoplankton and harmful algal blooms. Formed in 2001, PMN enhances the Nation's ability to respond to and manage the growing threat posed by HABs by collecting important data including phytoplankton species composition and distribution as well as environmental conditions. The goal of the network would provide aquaculture farms advanced warning of these shellfish toxic and ichthyotoxic blooms to empower growers to mitigate the effects of these blooms.

This citizen science approach to monitor HABs was able to grow into a national monitoring program by use of various web-based tools such as an interactive web site and a geographic information system tool for data visualization and searchable database. This presentation will outline the use of these technologies and highlight the use of volunteer data in aquaculture settings.

AQUACULTURE SUPPLY AND DEMAND DISRUPTIONS DURING THE COVID-19 PANDEMIC: A GLOBAL ECONOMIC ANALYSIS OF CHALLENGES AND SOLUTIONS

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The Covid-19 pandemic had devastating economic implications whose ripple effect cascaded from the production to the consumer in the supply chain. The movement restrictions adversely disrupted supply and demand chains. Although these disruptions were the primary impact of the pandemic, they had a negative ripple effect on aquaculture because labor immobility affected production. Subsequently, product shortages led to hyperinflation. Aquaculture economics has two overarching mainly profit optimization and cost minimization. The economic principles advocating for cost reduction and growth optimization are paramount for the thriving aquaculture industry. Although there are myriad approaches for achieving the two goals above, such as technology infusion into aquaculture and innovation, the pandemic-related disruptions adversely affected aquaculture supply chains making it challenging for aquaculture entrepreneurs to balance the aquaculture economic principles of cost minimization and gain maximization.

The acute product shortages made aquaculture farmers settle with the quality and prices available in the market, which occasionally compromised the quality and quantities of the aquaculture yield. The global economic disruptions affected the markets as aquaculture supply chains' resilience, agility and sustainability were tested to the limit. Optimizing food conversion, water, food, power, processing and storage cost is a fundamental consideration in aquaculture economics discourses. Although aquaculture has consistently risen as a solution to food security challenges, economic challenges spanning environmental, financial and technological aspects adversely affect the sustainability of this nascent yet rapidly evolving food security trend. The cost consideration in making aquaculture decisions is paramount because entrepreneurs must balance the quality/quantity of the fish produced and the profit gained. The subject is complex due to the multidimensionality of the aquaculture economic concept affecting contemporary aquaculture practices amid the ongoing Covid-19 pandemic.

Achieving the two overarching economic goals was challenging due to the Covid-19 disruptions. This research explores aquaculture economics to demystify how aquaculture farmers can optimize gains and reduce costs by evaluating the pandemic's effects on aquaculture supply and demand and key economic solutions for flourishing sustainable aquaculture. The study used a quantitative design. Twenty structured questionnaires on the effects of covid-19 supply and demand disruptions on aquaculture from an economic perspective were administered. This paper will demystify challenges associated with the Covid-19 disruptions to the aquaculture supply and demand chains from an economic perspective. The outcomes in this paper offer solutions and recommendations on how aquaculture farmers can deploy sound, economically-driven concepts to maximize gains, reduce costs and make aquaculture more sustainable, resilient, lean and agile to cope with similar future global disruptions.

NAEMO – NORTH ATLANTIC & EUROPEAN MUSSEL ORGANISATION

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Background

Mussels (*Mytilus spp.*) are key stone species providing valuable ecosystems services. Over the past years, mussel beds in Europe and North America have been reported to be in regression. Due to the lack of long-term monitoring, especially of unexploited mussel beds, there is no baseline for estimation of the severity of this decrease. Moreover, relatively little is known about potential threats to the welfare of mussels or if these threats are general or vary geographically.

Network objectives and roadmap

We established the North Atlantic and European Mussel Organization (NAEMO) to expand international collaboration between government, academia, industry and NGOs with the aim of providing knowledge-based support for improved management of mussels by increasing our understanding of the global and local processes affecting development of wild mussel populations and their interactions with farmed mussels. Outcomes from the initial workshop identified key questions for the network, specifically (1) evidencing and monitoring mussel bed decline, (2) identification of the causes for such decline, (3) interactions between wild mussel beds and farmed mussel populations and (4) improvements in communication between stakeholders. The roadmap (Figure 1) illustrates the future steps necessary to establish the network and to achieve the objectives, aims and goals of NAEMO. For more information or to join the network, please contact Åsa Strand asa.strand@ivl.se



Figure 1. Roadmap for achieving the NAEMO objectives and aims.

SPATIAL EXPRESSION PATTERNING OF SELECT GENES ASSOCIATED WITH EARLY SPERMATOGENESIS/SPERMIOGENESIS IN *Mytilus edulis*

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The spatial expression patterns of three genes (kelch-like protein homolog 10 (*klhl10*); armadillo repeat-containing protein 4 isoform X2 (*armc4*); mitochondria cytochrome oxidase I (*mt-co1*)) important during spermatogenesis/spermiogenesis were localized to cells within the acini of male *Mytilus edulis* using nonisotopic *in situ* hybridization and ribo-probes based on differentially expressed sequences derived from mantle RNA-seq libraries.

Hybridization signals for all three genes were associated with a layer of cells near the outer wall of the testicular acini. This cell layer is known to contain developing spermatogonia, spermatocytes, and spermatids with mature spermatozoa at the center of the acini.

Klhl10 was detected in immature spermatogonia/spermatocytes localized near the periphery of the acini with little or no expression in adjacent populations of spermatids or mature spermatozoa (Fig.1A). This may support an early role in ubiquitination and cytoplasmic reorganization.

Armc4 was also detected in cells near the inner periphery of the outer acinar wall. In contrast to *klhl10*, expression of *armc4* appeared to be spatially shifted toward the inner edge of the cell layer and toward the center of the acini. Cells expressing *armc4* transcripts here suggest a role in spermatid morphogenesis and maturation (Fig.1B).

Mt-Co1 was localized to cells through the inner periphery of the acini wall but not in mature spermatozoa at the center of the acini, reflecting the intense cellular activity and associated energy demands in the region (Fig. 1C).

This work provides a first view to the molecular mechanisms behind spermatogenesis/spermiogenesis in *M. edulis*.

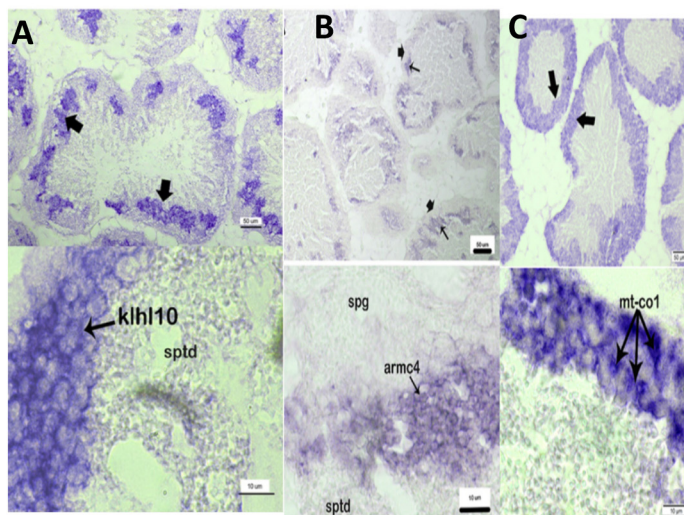


Figure 1. Expression of *klhl10*, *armc4*, and *mt-co1* within mantle testicular acini.

MOLECULAR PERSPECTIVES, EXPRESSION PROFILING AND FUNCTIONAL CHARACTERIZATION OF THIOREDOXIN MITOCHONDRIAL - LIKE PROTEIN (Trx-2) FROM BIG-BELLY SEAHORSE *Hippocampus abdominalis*

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Thioredoxin protein is a ubiquitous small redox protein (12 kDa) with characteristic conserved WCGPC active site that is found in organisms ranging from prokaryotes to eukaryotes. A distinctive thioredoxin folding motif composed of four-stranded β -sheets encompassed by three α -helices and an extra α -helix and β -sheet at the N-terminus. According to the subcellular localization, thioredoxin is divided into two isoforms. One is predominantly expressed in the cytosol (Trx-1) another one mitochondrial (Trx-2) one. Trx is involved in various biological functions for instance regulation of growth and gene expression, control of apoptosis, protein folding and scavenging of ROS.

In the present study, Trx-2 was identified from big-belly seahorse (*HaTrx-2*) cDNA library and *in-silico* analysis was carried out by using bioinformatic tools. The qPCR was performed to determine the tissue-specific distribution of *HaTrx-2*. The abiotic and biotic challenges were conducted with poly I.C, LPS, *Edwardsiella tarda*, and *Streptococcus iniae* to analyze the temporal expression of *HaTrx2* in the kidney tissue. Further, the Trx-2 was recombinantly expressed in *Escherichia coli* BL21(DE3) and the antioxidant property of recombinant Trx-2 (rHaTrx-2) was analyzed by performed the DPPH radical scavenging activity.

The HaTrx-2 has 172 amino acids which encoded by 519 bp ORF with a molecular weight of 18.8 kDa. Its calculated isoelectric pI is 7.80. The phylogenetic tree unveiled that the HaTrx-2 showed the evolutionary conserveness with other teleost fish Trx-2 counterparts especially with *Fundulus heteroclitus*. The highest value of the identity (78.7%) and similarity (86.2%) was observed with *Fundulus heteroclitus* Trx-2 in pairwise alignment results. The qPCR results revealed that *HaTrx-2* transcript ubiquitously expresses in all examined tissues with the highest expression found to be in the ovary. The immune challenges qPCR showed significant upregulated and downregulated *HaTrx-2* mRNA expression with all stimulants at certain time post immune challenges in the kidney. The DPPH radical scavenging activity elucidated the HaTrx-2 antioxidant property in a dose-dependent manner. Collectively, our data suggested that *HaTrx-2* may play an indispensable role in host oxidative protection and immune response upon pathogen attacks.

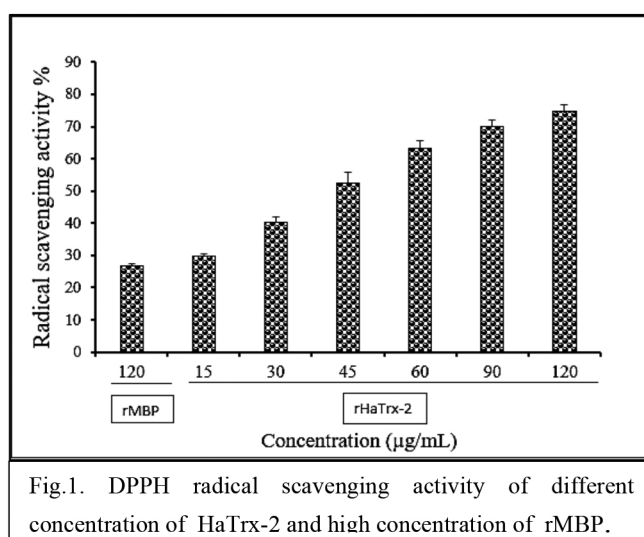


Fig.1. DPPH radical scavenging activity of different concentration of HaTrx-2 and high concentration of rMBP.

MOLECULAR CHARACTERIZATION OF CAVEOLIN-1 (Cav-1) FROM CLOWNFISH (*Amphiprion clarkii*) AND TRANSCRIPTIONAL MODULATION UPON IMMUNE CHALLENGES

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Caveolin-1 (Cav-1) is the main member of the caveolin protein family. It is a membrane-bound scaffolding protein implicated in caveolae formation that interacts with and controls the activity of a large number of proteins involved in signaling pathways relevant to growth, survival, and proliferation in different cell types. Cav-1 is expressed in two isoforms (Cav-1 α and Cav-1 β). Cav-1 is involved in numerous cellular processes, such as endocytosis, lipid transport, directional cell motility, and cell cycle regulation. In this present study, we examined molecular features and mRNA expression of Cav-1 from *Amphiprion clarkii* (*AcCav-1*).

The cDNA & protein sequence of *AcCav-1* was analyzed molecular wise by using bioinformatics software. The qPCR was performed to determine the tissue-specific Cav-1 mRNA expression of twelve different tissues collected from healthy *Amphiprion clarkii*. Also, the mRNA expression profile of *AcCav-1* upon immune challenged with poly I.C, LPS, and *Vibrio harveyi* was investigated.

The cDNA sequence of *AcCav-1* consists of 546 bp of the open reading frame (ORF) which encoding 181 amino acids with a molecular weight of 20.73 kDa and calculated pI of 5.48. According to the phylogenetic tree analysis, *AcCav-1* is close to its ortholog from *Amphiprion ocellaris*. The qPCR results revealed *AcCav-1* transcripts ubiquitously expressed in all examined tissues with high expression in skin. The mRNA expression levels in the spleen showed significant down-regulation with Poly I.C for all time and significant up-regulation with LPS at 12h and 48h and with *V.harveyi* at 6h and 48h time post immune challenge. On the whole, our data indicated *AcCav-1* may play an important role in the host immune response.

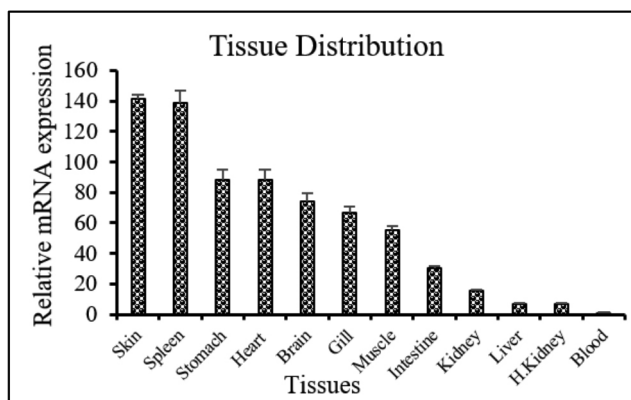


Fig.1 Tissue specific expression of the clownfish Cav-1

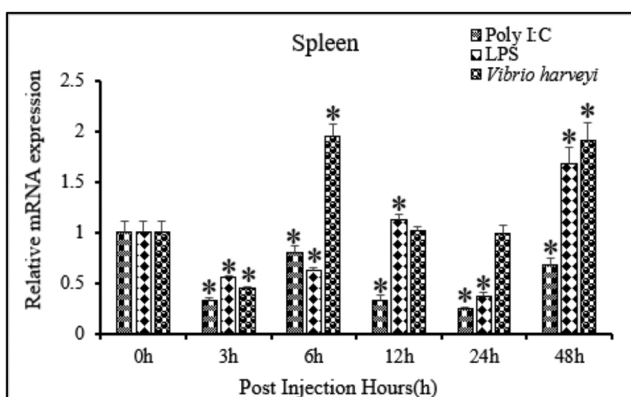


Fig.2 Relative expression of clownfish Cav-1 in spleen tissue at certain time points after different immune

ENABLING EASE OF ACCESS TO COMPLEX SPATIAL DATA: DEVELOPMENT OF AN INTERACTIVE GIS MAP TO SIMPLY AQUACULTURE DECISION MAKING

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The development of novel aquaculture operations requires extensive information on the local environment, infrastructure, and available support services. Compiling and sorting this information is an important prerequisite for supporting sound regulatory and business decision making. Despite the availability of various high-quality applicable data, the large quantity and disparate sources of different data sets requires extensive effort to collect and evaluate in a coherent manner. To help address this problem the Centre for Marine Applied Research partnered with the Atlantic Canada Opportunities Agency and provincial departments to create a single-GIS platform that shows important infrastructure, environmental data and services to support aquaculture development in Nova Scotia. An online interactive GIS map was developed in an ArcGIS Platform and posted on the CMAR website. Over fifty spatial layers make available a wide range of spatial data important to net-pen, shellfish and land-based aquaculture. This presentation reviews the tool functionality, available data layers and examples of how this tool can support pragmatic decision making for aquaculture.

ROS PRODUCTION MAY LIMIT THERMAL TOLERANCE AND CARDIAC PERFORMANCE AT HIGH TEMPERATURES IN ATLANTIC SALMON

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Given the impacts of climate change, it is important to understand what factors determine thermal tolerance and affect cardiac performance in fishes, and to be able to select for these important indices within populations used in aquaculture. Mitochondrial function [e.g., respiration and reactive oxygen species (ROS) production] is negatively impacted by high temperatures and may limit fish cardiac performance and upper thermal tolerance. Thus, we challenged twenty families of cultured Atlantic salmon to an incremental ($+0.2^{\circ}\text{C day}^{-1}$) thermal maximum (IT_{Max}) test. IT_{Max} varied significantly between the 20 families (range 25.0 to 23.3°C). Remaining fish from the two most (F19 and F4) and least (F1 and F6) tolerant families (which had been held at 10°C) were then warmed to 18°C , acclimated at this temperature for ≥ 2 weeks, and had their cardiac mitochondrial function assessed at test temperatures of $20, 24, 26$ and 28°C ($n=7-9$). With regard to mitochondrial respiration, there was no clear separation between the temperature tolerant and intolerant families. However, a number of mitochondrial parameters were significantly different between Family 1 and the two tolerant families. For example, values for respiration and in particular absolute ROS production were consistently or often higher for Family 1 at the various test temperatures (Figure 1A). These data suggest that ROS production, not respiratory capacity, limits a fish's ability to tolerate long-term exposure to high temperatures.

In a second experiment, we acclimated salmon to 10 and 20°C for ≥ 2 weeks and then: used strips from half of the heart's ventricle to simultaneously measure muscle work and O_2 consumption (and thus efficiency) at 20 and 26°C ; and used the other half to obtain myocardial homogenates and isolated mitochondria so that mitochondrial function could be measured at the same temperatures. Total CI+CIII ROS production was negatively correlated with the efficiency of contraction in 20°C acclimated salmon, whereas this relationship was not evident for 10°C acclimated fish (Figure 1B).

In conclusion, it appears that ROS production at high temperatures contributes to family-based differences in upper temperature tolerance, and may limit myocardial contraction efficiency (and thus heart function) at high temperatures. These latter results are consistent with previous data showing that warm acclimation improves cardiac mitochondrial function at high temperatures.

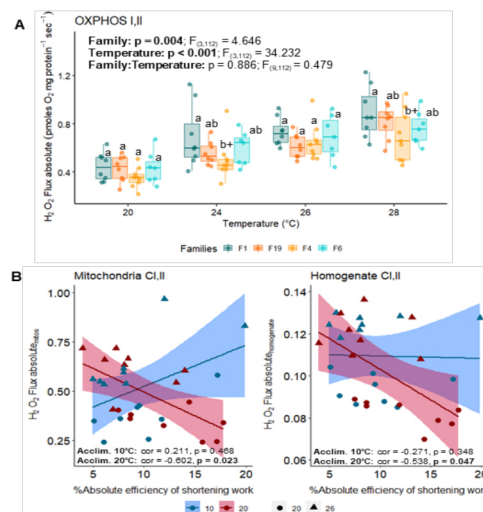


Figure 1: (A) Absolute ROS production in different salmon families (F1, F19, F4 and F6) at test temperatures of $20, 24, 26$ and 28°C . (B) relationship between absolute mitochondrial ROS production and absolute efficiency of shortening work (%) in hearts from 10 and 20°C acclimated fish

ATLANTIC SALMON HEAD KIDNEY GENE EXPRESSION RESPONSE DURING SINGLE (BACTERIAL) AND CO (BACTERIAL AND SEA LICE) INFECTIONS

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Caligidosis and Piscirickettsiosis, caused by *Caligus rogercresseyi* and *Piscirickettsia salmonis*, are the two of the most economically important diseases affecting the salmon industry. Evidence has shown interaction between these two conditions (1), however, no efforts at molecular levels have been reported. In this study, we infected *Salmo salar* with two different lice densities (15 and 50 copepod/fish) followed by *P. salmonis* injection [isolate LF89; at 16 days post-infection (dpi) with copepod]. There were two no-lice control groups that received either only LF89 or mock injection. Head kidney tissue sampled at 27 dpi was used in targeted expression profiling of sixteen selected transcripts using real-time quantitative PCR (qPCR). Lice count on low- and high-density copepod groups was 6.9 ± 1.3 (mean \pm standard deviation) and 18.3 ± 5.0 , respectively. A few bacterial (e.g., *camp-b*, *hamp-a*, *saa5*, *il1b*, *il8-a* and *lect2-a*), lice (e.g., *mmp13-c* and *timp2-b*) and viral (e.g., *mx-a* and *rsad2-a*) responsive biomarkers were induced by single (LF89 alone) and co (lice and LF89) infections. Copepod density proportionally increased the magnitude of inductive response of the majority of examined transcripts. Compared to non-infected controls, expression of *selenop-b* and *pgds-c* was down-regulated in all different infection groups. Trends in gene expression in this study were comparable with our recent study (2) except for antiviral transcripts. Collectively, these preliminary results suggest that *C. rogercresseyi* and *P. salmonis* may exert a synergistic host immune response, which could further be dependent on *C. rogercresseyi* load.

1. Arriagada G, Hamilton-West C, Nekouei O, Foerster C, Müller A, Lara M, et al. *Caligus rogercresseyi* infestation is associated with *Piscirickettsia salmonis*-attributed mortalities in farmed salmonids in Chile. Preventive Veterinary Medicine. 2019;171:104771.
2. Xue X, Caballero-Solares A, Hall JR, Umasuthan N, Kumar S, Jakob E, et al. Transcriptome Profiling of Atlantic Salmon (*Salmo salar*) Parr With Higher and Lower Pathogen Loads Following *Piscirickettsia salmonis* Infection. Frontiers in Immunology. 2021;12.

GLOBAL REGULATION OF AQUACULTURE OCCUPATIONAL HEALTH AND SAFETY: AN OVERVIEW

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This presentation introduces a session on regulation of aquaculture occupational health and safety (AOSH) in different countries. It presents an overview of findings related to AOSH regulation drawn from a forthcoming scoping report on AOSH commissioned by the Food and Agriculture Association co-authored by Watterson, Neis and others. An expert panel was asked to examine the often neglected and under-researched subject of AOSH and its immediate supply chains by gathering, reviewing and synthesizing existing scientific and gray literature in order to produce a series of national and regional profiles of AOSH. The focus included hazards and risks and opportunities/constraints for addressing these including through regulation. The FAO report offers a framework for examining AOSH across diverse settings and types of activities and provides a tentative analysis of the main production activities at global, regional and national levels and how these may impact on AOSH and nearby communities, both negatively and positively. In 2016, there were over 19 million aquaculture workers in the world – including large concentrations in China and South-east Asia. In some marine settings, aquaculture is one of the most hazardous industries in the world along with fishing and aquaculture. Much of the attention on aquaculture has focused on employment, food security, food safety risks and concerns about environmental sustainability. Less attention has been paid to AOSH, including to regulatory OSH frameworks and effectiveness. The scoping report that AOSH continues to be of marginal interest for many agencies and hence regulation may be neglected. In most contexts, aquaculture falls under agriculture and fisheries which, in turn, have specific laws and regulations that rarely specifically cover aquaculture except for regulations, codes and official guidance on such things as fish cages and manual handling. In mariculture, jurisdiction may be split between departments and agencies responsible for navigation, fishing and seafaring safety and those responsible for the occupational health of workers.

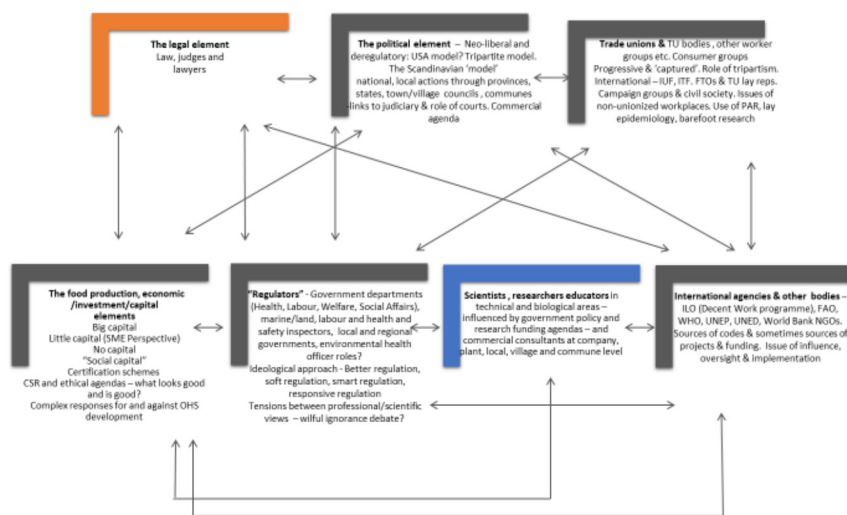


Figure 1: Industry, policy, regulatory and worker interactions in OSH

PRACTICAL OBSERVATIONS IN REFRIGERATION FOR AQUACULTURE CHILLER AND HEAT PUMP SYSTEMS USING CARBON DIOXIDE AS A REFRIGERANT IN PLACE OF REFRIGERANTS WITH HIGHER GLOBAL WARMING POTENTIAL

Philip A Nickerson, P.Eng*

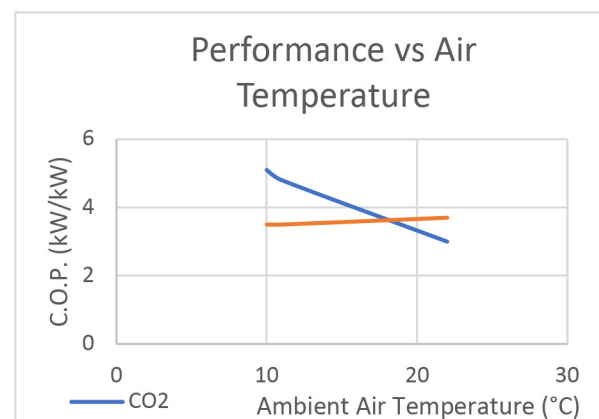
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Refrigeration to heat or cool water for land-based aquaculture often accounts for a significant portion of the capital expense, and the electrical energy operating expense. In recent years, refrigeration has also been flagged as a significant portion of the environmental footprint. This real-world case study was conducted to compare the performance and market reception of CO₂ chillers alongside one of the most efficient conventional refrigerant chillers available. The findings, notes, and observations are presented here in an effort to help those who design and/or buy refrigeration equipment for aquaculture to make informed choices when evaluating refrigeration options for aquaculture operations.

Two of each chiller were constructed and installed in a live shellfish holding operation. Each chiller was supplied by an identical pump that solely serviced that chiller. An ultrasonic flow meter was used to measure flow rate. Water temperature was measured before and after each chiller always ensuring that the inlet and outlet temperature differential was calibrated to zero before the chiller was turned on.

What are believed to be the first chillers in the world to use CO₂ in a live seafood system were designed and built by a company in Nova Scotia, and commissioned in Prince Edward Island in 2022. Data shows that below the transcritical point the efficiency of the CO₂ refrigerant is higher than conventional refrigerants. Above the transcritical point, the efficiency is reduced below the point of conventional refrigerants. Therefore, the verdict on efficiency of CO₂ as compared with conventional refrigerants rests on the ambient temperature or the cooling water temperature which drive the 'condensing temperature' or compression ratio.

There are also market advantages to using CO₂ as a refrigerant that are presented herein.



CHROMOSOME LEVEL REFERENCE GENOME OF CUNNER (*Tautogolabrus Adspersus*) AND WHOLE GENOME RESEQUENCING REVEAL EVIDENCE OF ECOLOGICAL ADAPTATION THROUGHOUT ATLANTIC CANADA

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Cunner (*Tautogolabrus adspersus*), a northern ranging species of *Labridae*, are being considered for use as a cleaner fish in North American salmonid aquaculture. Cunner genomics and population structure remain uncharacterized, and potential consequences of aquaculture escapees on the fitness and demography of wild populations are undefined.

To understand the existing genetic variation and evolutionary history of cunner, a 0.72Gbp reference genome spanning 24 chromosomes was constructed. Comparison to other *Labridae* reference genomes revealed 24 homologous chromosomes, suggesting a conserved genomic architecture. Whole genome resequencing of 749 individuals from 19 locations throughout Atlantic Canada identified 11 million genetic variants. Principal component analysis revealed four subpopulations in Atlantic Canada that were strongly associated with geography: Nova Scotia (NS), Northwest Newfoundland (NWNL), Northeast Newfoundland (NENL), and Southeast Newfoundland (SENL) (Figure 1A, 1B). Pairwise comparison of subpopulations via F_{ST} revealed strong signals of differentiation at specific regions in the genome and these regions overlapped with identified signatures of selection.

Genomic variation was also significantly correlated with environmental variation further suggesting that the spatial structure of cunner populations has partially stemmed from local adaptation. Observed patterns of genetic variation suggest that local adaptation to environmental differences across Atlantic Canada has resulted in genetically distinct and adaptively unique cunner subpopulations across the sampled range, these findings can inform future use of cunner within salmon aquaculture in Atlantic Canada.

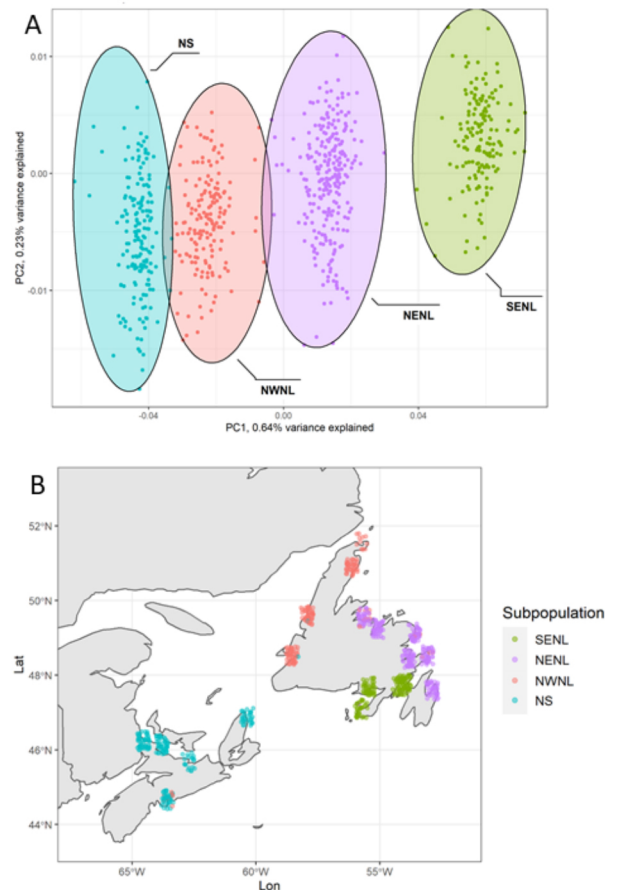


Figure 1. A) Scatter plot of Principal Components (PCs) of genetic variation for 749 cunner individuals from Atlantic Canada. The coloured ellipses denote the genetic subpopulations identified through *k*-means clustering. **B)** Map of sampling locations where the colour-coordinated points represent individuals and the subpopulation to which they were assigned.

EFFECT OF DIETARY PROTEIN ON SPAWNING PERMORMANCE OF THE ENDANGERED *Oreochromis esculentus* -KENYA

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Singidia tilapia (*Oreochromis esculentus*) is endemic to Lake Victoria and was once the most important fish species in the lake. Despite this, *O. esculentus* is now enlisted in the World Conservation Union Red book (IUCN) of endangered species as critically endangered. To rescue this highly valued fish species from total extinction, urgent conservational measures are required. This study investigated *Oreochromis esculentus* feed requirement for artificial breeding. Three feeds containing 20%CP, 25%CP and 30%CP were tested on 12 broodfish groups stocked in 12 nylon hapas (2x2x1m³) installed in earthen ponds. Females fed with 30%CP and 25%CP produced 40.1% and 36.3% more seed (eggs and yolk sac fry) respectively than females fed with 20%CP (Table 1 and 2). This study showed that the use of 25% crude protein diet is nutritionally adequate in *O. esculentus* seed production and that high protein diets were not beneficial in terms of seed output. The study recommends the use of 25% crude protein level feed when producing seeds in *O. esculentus*.

Table 1: Absolute and relative fecundity of *O. esculentus* as affected by broodstock feed

Feed CP	Female Initial weight	Female Final weight	Female Mean weight	Clutch size	Seeds/g female	Seed/female/day
20%	114.3± 3.47 ^a	216.8± 11.2 ^c	166.2± 9.33 ^c	503.0± 35.7 ^b	12.9± 0.33 ^a	16.8± 0.25 ^b
25%	114.5± 3.18 ^a	285.7 ±8.22 ^b	207.3± 15.2 ^b	685.8± 41.9 ^a	11.0± 0.19 ^b	22.9± 0.29 ^a
30%	113.8 ±5.65 ^a	314.5 ±6.06 ^a	251.1± 22.1 ^a	704.8±48.3 ^a	13.4± 0.16 ^a	23.5± 0.33 ^a

Means in the same column having different letters are significantly different (P<0.05)

Table 2: Reproductive performance of *O. esculentus* broodfish subsequently fed with varying levels of crude protein during the study period of 120 days

Broodfish feed and reproductive parameters	20%CP	25%CP	30%CP
Total feed consumed (kg hapa ⁻¹)	6.78 ^c	8.01 ^b	9.85 ^a
Clutch size (no.clutch ⁻¹)	503.0 ^b	685.8 ^a	704.8 ^a
Seed/female day ⁻¹	16.77 ^b	22.86 ^a	23.49 ^a
Seed g female	12.11 ^a	13.21 ^a	11.14 ^a
Seed yield (no.m ⁻² day ⁻¹)	25.15 ^b	34.29 ^a	35.24 ^a
Seed output (no. g ⁻¹ feed)	1.78 ^b	4.14 ^a	3.46 ^a
Seed output(no. g ⁻¹ protein)	8.9 ^b	20.7 ^a	17.29 ^a
Protein Efficiency Ratio (PER)	12.25 ^a	8.11 ^b	12.43 ^a

Means in the same column having different letters are significantly different (P<0.05)

ANTIMICROBIAL RESISTANT BACTERIA AND OCCUPATIONAL HEALTH IN AQUACULTURE: REGULATION OF ANTIBIOTIC USE ACROSS FOUR COUNTRIES

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Marine aquaculture workers face hazardous work environments. Industry use of antibiotics is a workplace hazard with potential consequences for adverse health effects on unprotected workers through exposure to antimicrobial resistant (AMR) bacteria. The growing literature on AMR bacteria in aquaculture has found evidence of elevated incidences of resistance stemming from antibiotic (AB) use in sediments under cages, and in salmonid gut and feces, but potential occupational health impacts are poorly studied. This presentation synthesizes evidence and identifies knowledge gaps on potential occupational exposures to AMR bacteria in aquaculture. We discuss existing data on trends in antibiotic use, reporting mechanisms, and regulation of antibiotic use across Norway, Canada, Scotland, and Chile. In Norway, prescription sales of ABs have hovered around 1.5 mg of active AB ingredients/kg of farmed fish since 2007; Scotland reported 6.5 mg AB/kg of salmon (2018); Canada reported 37 mg AB/kg salmon (2016) and Chile reported 530 mg AB/kg salmon (2016). Types and quantity of AB use vary across countries and within regions. The rate of AB use appears to be trending downwards in ¾ countries with Chile trending upwards between 2010 and 2016 and with some volatility. Aquaculture operators from all four countries are required to identify type and quantity of antibiotic prescribed or applied and in some cases application frequency is also reported. There are a number of weaknesses in the reporting and regulation systems for antibiotic use in aquaculture in Scotland, Canada, and Chile relative to Norway. National reporting and regulatory protocol differences make it difficult to precisely compare rates. From a regulatory perspective, all use of ABs in Norwegian, Scottish, Canadian, and Chilean aquaculture is regulated by law; only veterinarians and aquamedicine biologists can prescribe antibiotics and only for treatment of clinically diagnosed diseases. In Norway, the Veterinary Medicines Register established in 2011 by the Norwegian Food Safety facilitated the collection of information on each prescription for ABs for use in farmed fish. Data on AB use in Scotland is not easily accessed. In Canada, application of ABs requires authorization by Health Canada, and is regulated through the Food and Drug Act but public reporting only started in 2016. In Chile, antimicrobial treatments should be reported monthly through the Aquaculture Inspection System (SIFA). ABs for use in farmed fish generally must be dispensed through pharmacies. Chilean aquaculture's high dependency on ABs has stimulated research exploring AMR from a public health perspective. Researchers have documented elevated enrichment of AMR bacteria in patients from regions with intensive salmon aquaculture, but there is limited similar research in the other jurisdictions. Agencies responsible for regulating and monitoring occupational health and safety do not appear to be addressing potential AMR bacteria hazards including identifying potential exposure hotspots.

COMPARATIVE ANALYSES OF DRYING PROFILES AND ECONOMIC OUTCOMES OF SMOKE-DRIED CATFISH (*Clarias gariepinus*) USING TRADITIONAL AND STANDARDIZED ECO-FRIENDLY KILNS IN LAGOS, NIGERIA

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Fish is a major source of animal protein in a developing country like Nigeria and its shelf life is most commonly lengthened by smoke-drying using Traditional Drum Kiln (TDK). Two major challenges of using TDK however are subjective processing process and paucity of documented economic data involved with its use. The aim of this study was to standardize TDK, construction of Eco-Friendly Kiln (EFK), identification of market characteristics, assessing of total costs and estimation of revenue generation from smoke-drying catfish (*Clarias gariepinus*) in eight selected fish markets in Lagos, Nigeria.

TDK was constructed as used in the selected fish markets measuring 72 cm height and 55 cm diameter while EFK was constructed starting with TDK and then fortifying it with refractory lagging material comprising of clay, sawdust and silicon carbide in ratios 4:2:0.5 to form the drying chamber where the fresh fish were arranged for the smoke-drying process. An aperture with dimensions 60 x 20 cm was cut at the front and a flame chamber with dimensions 93.1 x 77.5 x 85.2 cm was built with red bricks and coated in and out with the refractory was assembled in front of the drying chamber. Smoke filters of sizes 0.1 and 0.3 cm were fitted in the aperture to control the amount of smoke particles entering the drying chamber from the flame chamber. A smaller aperture with dimensions 29.6 x 16.8 cm was cut at the bottom middle to accommodate the burning fuel (dried red mangrove) (Plate 1). Market parameters, total costs and revenue generation in eight fishing villages in Lagos State were investigated and smoke-drying was done at a stretch of an average of 24 ± 3 hours. The standardization of TDK and EFK was done by maintaining the smoke-drying temperature in both kilns between 60 °C and 80 °C with the use of temperature sensor, audio smart alarm and Light Emitting Diode.

EFK fitted with two layers of 0.3 smoke filters was found to have the highest quality smoke-dried from the drying profile. The market characteristics indicated that smoke-drying process was predominantly carried out by women with little formal education and there were differences in the mechanics of fresh fish procurement, processing methods and marketing channels. The cost and revenue estimation using both kilns showed that the use of EFK had significantly greater turnover and would generate about ₦ 2,715,752 as compared with ₦ 1,127,752 with the use of TDK over an estimated two-year lifespan of EFK.



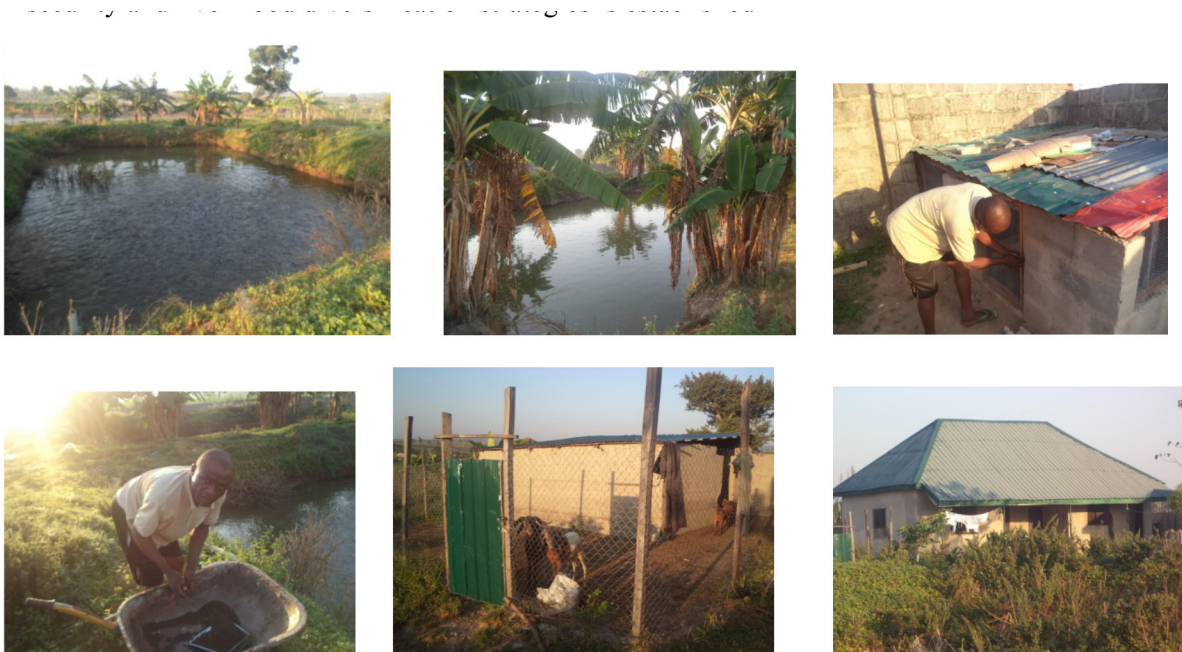
Plate 1: Constructed Eco Fish Kiln (EFK)

THE LIVELIHOOD DIVERSIFICATION STRATEGIES, LANDSCAPING OF A TYPICAL FISH FARMS IN GWAGWALADA ABUJA NIGERIA

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The study examined the different livelihood diversification strategies, landscaping of a typical Fish farms which is located in Gwagwalada (Compensation Layout Old Kutunku Abuja Nigeria). In order to realize the objectives of the study, primary data were obtained from farm workers, buyers, the owners of the fish farm, with the aid of structured questionnaire, many farm visit were made, pictures were taken and oral interviewed. We do observe the consumption rate of feeding of the fish that is in the morning and the late in the evenings, the persons that take care spread the fish feeds to the ponds and, the dead fish floats on the water and it is consume by the living fish. We also seen that banana are being planted too produce shield for the fish farm and we have the fishing net, when the fish fingers are being newly introduce too protect it from the dangerous animals and birds being feed. There is also a farm steads, where the farmers live and there are planted of grasses too check the soil erosion and make the pond too be strong and we also notice some of the feeding aspect of the goats and caging of the goats, but some of them were being sold up to stop the eating of the planted crops which could have destroy the farms. Thus, a significant relationship between households' food insecurity and livelihood diversification strategies is established.



ELUCIDATION OF PROINFLAMMATORY ROLE OF IL12 α FROM *Amphiprion clarkii*

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Cytokines play a critical role in the immune system of the fish. Interleukin 12 family consisted of five cytokines such as IL12, IL23, IL27, IL35 and IL39. The diversity of IL12 family is achieved by combining different α (p35, p19, p28) and β (p40, EBI3). IL12 or p35: p40 is well known as a proinflammatory cytokine. It can induce STAT4 and IFN γ which exert proinflammatory activities. In this study, we investigated the molecular, transcriptional and proinflammatory aspects of IL12 α /p35 subunit from *Amphiprion clarkii* (Acp35). For that, we conducted *insilico* analysis, tissue specific expression analysis, immune challenge experiments and down-stream gene expression analysis of IL12 pathway.

Phylogenetic analysis and multiple sequence analysis showed that Acp35 sequence clustered with fish p35b orthologs distinctly. Further, Acp35 shared the highest identity and similarity with *A. ocellaris*. Tissue-specific expression profile demonstrated that Acp35 highly expressed in gills. According to the temporal expression in gills, Acp35 exhibited significant transcript modulation against polyinosinic:polycytidylic acid, lipopolysaccharides and *V.harveyi*. Further, recombinant Acp35 stimulated p40, STAT4 and IFN γ expression significantly in FHM cells.

Hence, these results indicate that Acp35 could be stimulated by PAMP stimuli and pathogens with different expression profiles. Further, it could induce downstream genes in IL12 pathway. Altogether, results in this study suggest that Acp35 might act as a proinflammatory cytokine.

SAFETY AND EFFICACY EVALUATION OF AN AUTOGENOUS *Vibrio anguillarum* VACCINE IN LUMPFISH (*Cyclopterus lumpus*)

Oluwatoyin Onireti*, Trung Cao, Hajarrooba Gnanagobal, Joy Chukwu- Osazuwa, Ignacio Vasquez, Ahmed Hossain, Vimbai I. Machimbirike, Steve Hill, Zihyou Chen, Andre Khoury, Yenney Hernandez-Reyes, Antoine Khoury, Nicole O'Brian, Andrew K. Swanson, Jennifer Monk, Jillian Porter, Danny Boyce, Javier Santander

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Lumpfish (*Cyclopterus lumpus*) has become the predominant cleaner fish species used in North American salmon aquaculture for sea lice (e.g., *Lepeophtheirus salmonis*) biocontrol. Lumpfish utilization has contributed significantly towards eliminating the utilization of chemotherapeutants by effectively controlling the abundance of this damaging pest of Atlantic salmon (*Salmo salar*) aquaculture. *Vibrio anguillarum* is a frequent pathogen of lumpfish in Atlantic Canada. Here, several vaccine trials against *V. anguillarum* were conducted. Fish health, including effective vaccine design, and vaccination programs have been identified as a high priority. In this study, the safety and efficiency of a *V. anguillarum* autogenous vaccine were evaluated. Five treatments with three replicates were used in this study. They were PBS (negative control), in-house vaccine (positive control), autogenous IP (intraperitoneally), autogenous deep and autogenous deep, and IP boost. Length and weight were taken, gross pathology was conducted. These treatments were bath challenged with *V. anguillarum* J360 (serotype O2) after 10 weeks post-vaccination, and the survival was bath re-challenged after 9 weeks of the first challenge with *V. anguillarum* J360 (serotype O2) and *V. anguillarum* J382 (serotype O1). Tissue samples were collected for tissue colonization count. No mortality after vaccination signified that the vaccine is safe. The gross pathology score indicated a normal immune response. The in-house IP, autogenous IP, and autogenous deep and IP boost had a relative percentage survival (RPS) of 76.12%, 72.12%, and 68.75% respectively in the first challenge. The vaccine only protects against *V. anguillarum* J360 (serotype O2) but does not confer protection against *V. anguillarum* J382 (serotype O1).

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SELECTION OF LUMPFISH (*Cyclopterus lumpus*) RESISTANT TO BACTERIAL INFECTION

Oluwatoyin Onireti*, Joy Chukwu-Osazuwa, Trung Cao, My Dang, Steve Hill, Zihyou Chen, Denise Tucker, Jillian Porter, Andrew Swanson, Danny Boyce, Javier Santander

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The selection of broodstocks resistant to infectious diseases is a proven sustainable and accessible measurement to improve fish health. In this study, lumpfish that survived the intraperitoneal challenge with *Aeromonas salmonicida* or *Vibrio anguillarum* was selected. These fish were quarantined and grown until they reached sexual maturity. Resistant males were crossed with resistant females and domestic females. Similarly, resistant females were crossed with domestic males. A few individuals of the F1 generation from the resistant cross showed an eye deformity. After the F1 generation reached an average weight of 73g, they were bath challenged with *V. anguillarum* (1×10^4 CFU/mL) for 30 minutes. At 42 days post-challenge, the relative percentage survival (RPS) of the resistant cross was 43.05%. The RPS for the resistant female crossed with the domestic male and the resistant male crossed with the domestic female was 6.77 % and 0%, respectively. These results suggest that the lumpfish resistance to *V. anguillarum* could be conferred by a recessive set of genes.

EVALUATING THE GROWTH OF BULL KELP *Nereocystis luetkeana* FROM HATCHERY TO LONG LINE CULTIVATION IN HUMBOLDT BAY

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The cultivation, restoration, and conservation of macroalgae are emerging mariculture practices in the United States. In spring of 2019, Cal Poly Humboldt partnered with GreenWave to establish one of the first small-scale commercial seaweed farm, ProvidenSea, in California. Since then, the farm has been home to several research projects that focus on regenerative and related initiatives in Humboldt Bay. The purpose of this proposal is to evaluate the cultivation of bull kelp (*Nereocystis luetkeana*) in a hatchery at the Cal Poly marine lab in Trinidad, CA, and successfully integrate them to grow out in open water in Humboldt Bay.

This project will consist of 8 aquaria replicates containing spools (pvc wrapped with speeding string). We will evaluate the growth and morphology of bull kelp using a variety of seeding strings commonly used in different regions for open water cultivation of macroalgae. Once the spools reach the optimal development stage and length (juvenile sporophyte at 3 mm in length), they will be transported to Cal Poly Humboldt ProvidenSea seaweed farm, where they will be out planted in the longline for further assessment of growth.

Nutrients in the seaweed's tissues and surrounding water will be analyzed to determine the extractive properties of macroalgae grown in the bay.

Presently, this is one of the first attempts to grow bull kelp from a hatchery to an ocean farm setting on the northern Pacific coast of California. Results from this study will help in expanding the under-developed research in bull kelp cultivation practices, early gametophyte settlement and provide a foundation for future farmers in regenerative seaweed farming practices within the California northern pacific coast.



Fig 1. Remote sensing imaging of Trinidad Bay wild bull kelp canopies where sori are collected. Image captured and processed by Hannah Joss

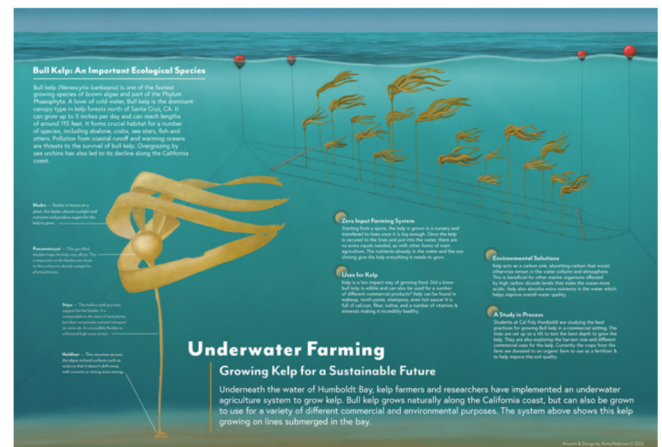


Fig 2. ProvidenSea multi-line system array. Illustration designed by Anna Pederson.

USING MICROBIAL GENOMICS TO MONITOR ECOSYSTEM RESPONSES TO ANTHROPOGENIC PERTURBATIONS: OIL SPILLS AS A CASE STUDY

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Microbial communities form the base of the food webs in the ocean, with phytoplankton fixing CO₂ to produce biomass, microzooplankton consuming this biomass and repackaging it for larger consumers and Bacteria and Archaea, “Prokaryotes”, recycling and repackaging waste products. These small organisms have short generation times, are abundant and contribute to a wide range of biogeochemical cycles. These characteristics make microbes ideal organisms for monitoring ecosystem responses to anthropogenic perturbations in the oceans.

Over the last several decades, advances in genomics technologies have led to the wide-spread use of amplicon sequencing in characterizing and monitoring microbial community composition and structure. Using primers designed to target a broad range (‘universal’) of organisms, or to specifically target a small taxonomic group or functional genes, amplicon sequencing can provide detailed information about which organisms are present or which metabolic pathways or biogeochemical transformations are possible. While DNA is the most common biomolecule targeted in amplicon surveys, RNA can also be targeted, providing information about active microbes and pathways.

Microbes have long been known to be active players in the recovery of marine ecosystems following an oil spill. Several genera of Bacteria, some fungi and a few phytoplankton have been identified as being able to degrade hydrocarbons in oil. Other groups may respond positively or negatively to oil, with responses occurring within hours or a few days. This rapid response may be useful in monitoring for hydrocarbon pollution but can also provide information about the fate and likely impacts of oil following a spill. Using a series of mesocosm experiments, ranging from 12 to 215 L, we have quantified the response of microbial communities to diluted bitumen, diesel and a medium crude using 16S rRNA amplicon sequencing. The microbial communities respond rapidly to hydrocarbons (Figure 1), with similarities across oil types. Seasonal and geographical differences in the responses are generally a function of the initial microbial community, which reflects the local environmental conditions. Generally, microbes are capable of contributing to oil spill response, aiding recovery of ecosystems after a spill.

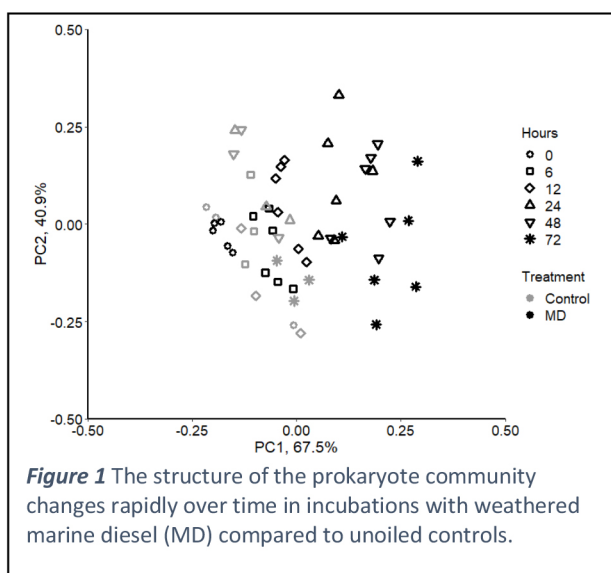


Figure 1 The structure of the prokaryote community changes rapidly over time in incubations with weathered marine diesel (MD) compared to un-oiled controls.

COMPARATIVE EFFICIENCY OF TWO COMMERCIAL FEEDS FED TO *Clarias gariepinus* JUVENILES UNDER LABORATORY CONDITIONS

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An 8-week feeding trial was conducted to compare the efficiency of two commercial feeds fed to *Clarias gariepinus* juveniles under laboratory conditions. One hundred and forty (140) juveniles were used for the study. The experimental diets used in this study were Top feed (indigenous feed) and Aller Aqua feed (foreign feed). The experiment was a complete randomized design with two experimental diets in six replicates of ten fish per bowl for a period of eight weeks. The growth performance (weight gain in kg), percentage growth rate (PGR), specific growth rate (SGR) and mean growth rate (MGR)) and nutrient utilization indices of fish were significantly different ($P<0.05$) in all fish fed the two experimental diets. The carcass traits (moisture, protein, fat, ash, carbohydrate) of the fish fed the experimental diets were significantly different ($P<0.05$) from one another.

Results from this study therefore showed that foreign fish feed performed better than local fish feed. The implication of the result is that Aller Aqua feed can be used for raising *Clarias gariepinus* juveniles in a culture system instead of Top feed without compromising their growth performances. However, Top feed is more cost effective for fish farmers than Aller Aqua which is twice more expensive due to the fact that Aller Aqua is an imported fish feed. Both are however recommended for *C. gariepinus* production but more preferably Top feed on the basis of affordability without compromising growth performance of the fish.

TRIPLOID MUSSELS FOR AQUACULTURE COULD LEAD TO BETTER PRODUCTION YIELD WITH HIGHER SURVIVAL RATE

Kevin Osterheld*, John Davidson, Luc A. Comeau, Tiago Hori, Isabelle Marcotte, Christian Pellerin, Jose MF Babarro, and Réjean Tremblay

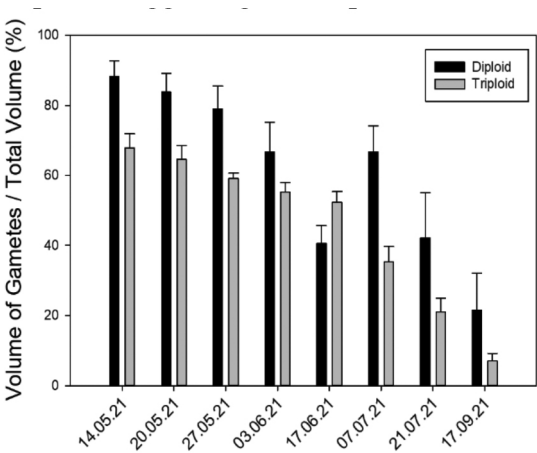
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Mussel farming is an important economic resource for Canada, producing 26 101 tonnes of mussels in 2019 worth more than 43 million Canadian dollars. A major cause of loss of production for mussel farmers originates from the fall-off of mussels during their growth. This phenomenon is particularly important during the reproduction season, as mussels invest most of their energy in the reproduction and have less energy for byssogenesis. This leads to byssal weakening, which can ultimately end with the fall-off of the mussels. Recently, there has been an increasing interest in the development of mussel hatcheries and more specifically on the production of triploid mussels. Indeed, it is known that triploidy often reduces the fertility of bivalves and can even produce sterile bivalves that have more energy to invest in their growth and other vital tissues (e.g. byssus).

In this study, we compared the production and mechanical properties of byssal threads of diploid and triploid mussels in relation to their energy budget and reproductive investment. To determine the effect of triploidy on byssal threads, diploid and triploid mussels were placed in a recirculating flume to induce production of byssal threads, then their scope for growth were estimated. The threads were counted and collected for tensile testing. Results show that triploid mussels produced up to 40% more threads than diploid mussels, with triploid threads eliciting better mechanical properties.

From May to September, during the reproductive season, diploid and triploid mussels were harvested in Georgetown Harbour, Prince Edward Island. Gonadosomatic index (GSI), gonad volume, gonad maturation, and sex ratio were determined for both groups. Results showed that while triploid mussels do invest a part of their energy in reproduction, they had lower GSI and gonad volume than the diploids. Also, for all tested groups, triploid gonads were less mature than those of diploids, and their sex ratio was significantly altered with absence of females in triploids. Furthermore, triploid gonads showed signs of gamete resorption, suggesting that triploid mussels could use their gametes for energetic storage.

This study shows that triploid mussels have lower energetic investment in reproduction and suggests that this energy could be used for a higher byssal production with better performance. Thus the creation and production of triploid mussels are of great interest for mussel farmers as they are less likely to fall-off during production, and have higher energy available after spawning, which could result in increased survival and production.



GREEN & BLUE: FOREST BY-PRODUCTS A PROMISING FEED RESOURCE FOR ATLANTIC SALMON

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Biotechnology processing of lignocellulosic biomass such as Norwegian spruce trees and chicken hydrolysates to produce high-quality microbial ingredients such as yeast can help address the protein deficit, reduce environmental footprint and improve fish health and welfare. The yeast obtained contains about 50-55% crude protein with a favorable amino acid composition and bioactive components like β -glucans, mannoproteins and nucleic acids, which can modulate the immune response of the fish.

In general, our studies with Atlantic salmon showed that fish perform well when fed yeast-based diets. Dietary yeast has also shown to have positive health effects in salmon, including improved gut barrier function, immunity, and gut microbiota composition. Documenting value-added effects beyond the nutritional value such as effect on health is important for the overall profitability of yeast, as yeast cannot compete with the cheap prices of plant proteins such as soy protein concentrate. However, yield, nutritional value and health effects of yeast depend on species, fermentation and downstream processing (DSP) conditions.

We evaluated the structure and physicochemical properties of the cell wall of three non-saccharomyces yeast species (*Cyberlindnera jadinii* (CJ), *Blastobotrys adeninivorans* and *Wickerhamomyces anomalus* (WA)) produced from spruce tree sugars and chicken hydrolysates and then exposed to DSP by autolysis. The yeasts were evaluated in diets for Atlantic salmon fry. Fish were fed a fish meal-based control diet or a challenge diet with 40% soybean meal (SBM), and 6 test diets with 40% SBM and 5% of the three whole yeast species or their autolysates. Adding yeast to a challenge SBM-based diet did not affect growth performance, but CJ and WA reduced the severity of enteritis in the distal intestine (DI) and modulated gut microbiota composition in DI and immune responses in DI and spleen. Best effects were observed with autolyzed WA. Processing by autolysis affected the length, accessibility and binding properties of the cell wall components β -glucans and mannoproteins, which improved the health effects of yeast.

CJ was also evaluated in a salmon trial during sea water transfer. Adding 25% CJ to a commercial-like diet increased feed intake and growth rate, reduced secretion of cytokines in DI (IFN γ , TNF- α , IL-1 β and IL-8) on both a transcriptional and a protein level, prevented morphological changes, and maintained numbers of CD3 cells in DI after seawater transfer, suggesting reduced inflammatory processes in yeast fed fish. Thus, yeast, depending on the species and processing conditions, is a promising protein source with health-beneficial properties during critical life stages.

The fermentation technology has now been scaled up and a field trial with Atlantic salmon in sea cages is being performed. This will provide important information on growth performance, health and sustainability and economical aspects of yeast as a novel feed resource for Atlantic salmon.

SELECTION FOR UTILIZATION OF PLANT-BASED DIETS LEADS TO NON-SPECIFIC PATHOGEN RESISTANCE IN RAINBOW TROUT

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Utilizing RNA-seq, this study compared the transcriptomic responses of three improved strains (VSEL, PSEL, and CSEL) of rainbow trout fry during acute stages of challenge with infectious hematopoietic necrosis virus (IHNV). The VSEL strain has been selected for resistance against the specific strain of IHNV used in our challenge, PSEL has undergone selection for utilization of plant-protein based feeds and previously has shown elevated non-specific disease resistance despite no disease related selection pressures, and the final strain, CSEL, is a commercial strain that has been domesticated for several years but has not been selected for specific viral disease resistance. Following a 21-day IHNV challenge, Kaplan-Meier survival estimator curves and cumulative percent mortality (CPM) showed significant differences in IHNV resistance across strains: VSEL - $19.3 \pm 5.0\%$, PSEL - $67. \pm 3.03\%$, CSEL - $94.6 \pm 4.1\%$ CPM. To evaluate acute responses to IHNV infection, whole blood, as well as samples from the kidney, liver, and intestine, were collected at 0, 4, 12, 24, and 48 hours post infection (hpi). Serum lysozyme activity, a marker of non-specific innate immunity, showed strain and temporal effects during the acute infection phase with PSEL showing the highest activity at 0 and 48 hpi. Differential gene expression responses were detected, with varying degrees, in all tissues, both between strains, as well as across acute timepoints within strains. The VSEL strain showed upregulation for a particular subset of viral recognition genes during early infection timepoints and rather limited upregulation of immune genes later, while maintaining and reactivating metabolic pathways. The CSEL strain showed a downregulation of metabolic related genes and a limited upregulation of immune genes, while the PSEL strain showed similar downregulation of metabolic genes during acute infection, yet when compared to the CSEL strain, showed a more robust innate immune response. Evaluation of upregulated immune response genes, as well as interferon-related genes showed the PSEL strain to have the greatest number of uniquely upregulated immune genes in both the kidney and intestine, with CSEL and PSEL showing a similar number of such genes upregulated in liver. A moderate number of immune response genes were shared between PSEL and CSEL in all tissues, though both PSEL and VSEL showed a high number of uniquely overexpressed immune response genes in the kidney, and PSEL showed the highest number of uniquely upregulated interferon related genes in the intestine. Overall, the VSEL response was unique from the CSEL with very little overlap in activated immune responses. Findings from this study highlight the disparity in IHNV resistance among genetic strains of rainbow trout, while identifying molecular mechanisms underlying differences in disease phenotypes. Furthermore, our results on trout strains with distinct selection backgrounds yields comparative insights into the adaptive gains brought about by selection programs for pathogen-specific disease resistance, as well as the non-specific immune enhancement associated with selection for utilization of plant-based diets.

SHAWANAGA FIRST NATION WALLEYE *Sander Vitreus* HATCHERY

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Shawanaga First Nation (SFN) is located approximately 30 kilometres northwest of Parry Sound in south-central Ontario. The Shawanaga First Nation's involvement in walleye culture and restoration dates back to the early 1970s. With the construction of a hatchery in 1996, the SFN was able to expand its efforts and now produces approximately 3 to 5 million walleye fry annually. The SFN fish culture program benefits from long-standing community support and is recognized as a valued opportunity to engage youth in various stages of the walleye collection, spawning and incubation processes, providing them with valuable experiences in fish culture and environmental management. It is evident that the Shawanaga First Nation walleye program has been well developed and is operated by knowledgeable and dedicated staff. Importantly, the First Nation leadership is supportive of the initiative.

As Anishinabe people, we have traditionally looked after the lands and waters since time immemorial practicing natural law, and from these practices we developed a way to balance traditional harvesting and modern conservation techniques that have proven to be successful as evidenced by the letter of support from the MNR/F which states that our work is precedence setting.

Since the beginning, the efforts towards operation of the Shawanaga Walleye Hatchery has been paid for by community finances, reflecting the strong commitment from the community towards this initiative. Under full direction from Leadership, strong fishery management practices & techniques have been put in to place to ensure the well being of this stock will be maintained for generations to come. Community River Watchers are deployed to protect the Spawning Walleye and the river during the most critical time of the year to compliment the dedication shown by hatchery workers, volunteers and the community.

With the soon to be completed expansion of their current facility, the community will be working towards improving on the numbers of Walleye being stocked to 8 to 10 million fry, countless fertilized eggs and nearly 200,000 advanced fingerlings.

USE OF MASS SPECTROMETRY IN AQUACULTURE NUTRITION STUDIES OF LIPIDS

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Of the four principal classes of biomolecules, i.e. carbohydrates, proteins, lipids, and nucleic acids, lipids stand out among the various metabolites in terms of the sheer number of distinct chemical entities. The diversity in lipid structures leads to a diversity in chemical and physical properties and then to a wide variety of biological functions ranging from energy storage to the formation of biological membranes, to cell signaling, inflammation, and immunity.

Lipids differ from the other major groups of biomolecules in that they are not polymers of relatively small numbers of different building blocks. They exist in highly interconnected metabolic pathways leading to various combinations of fatty acyl chains, backbone structures, and polar headgroups. The variety of polar headgroups gives rise to different phospholipids which make up most of the total lipids in membranes. These membranes form barriers for cells and organelles and act as a solvent within which membrane proteins fold and function. Each specialised membrane has a unique composition.

Until recently, the comprehensive analysis of lipids generally involved separation into simpler categories, according to their chemical nature. Lipids would be quantified as bulk species and/or as individual components. Advances in chromatography and mass spectrometry permitting quantitation of lipid molecular species is greatly propelling our ability to study metabolic pathways and networks.

Lipidomics is the complete analysis of lipid molecular species, including their quantitation, in order to study metabolic pathways and networks. The large number and structural diversity of natural lipids and their modifications make lipidomic analyses challenging. It has been estimated that there may be as many as 200,000 individual lipid structures.

The method of choice for sensitive detection and quantitation of lipid molecular species is mass spectrometry, especially with electrospray ionization, either by direct infusion (shotgun lipidomics) or coupled with liquid chromatography. A specialization of mass spectrometry, isotope-ratio mass spectrometry, is used to measure the relative abundance of isotopes in a sample. Compound-specific stable isotope analysis can be used to determine the extent of long-chain polyunsaturated fatty acid synthesis or the uptake and movement of organic nutrients from farm outputs to the surrounding ecosystem.

SHIP-SOURCE OIL POLLUTION COMPENSATION FOR DAMAGES TO FISHERS, AQUACULTURISTS, AND ALL INVOLVED IN RELATED ACTIVITIES

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Did you know that there is a federal Fund that compensates those who suffer damages caused by any type of oil, from any type of ship or boat, anywhere in Canadian waters? All involved in the “fishing sectors”¹, from catch or harvest to consumption or use can benefit from the Ship-source Oil Pollution Fund (the Fund).

Over the past 30 years, the Fund has received very few claims from the fishing sectors: **less than 1%** of the total amount paid from the Fund. We are therefore increasing our outreach efforts!

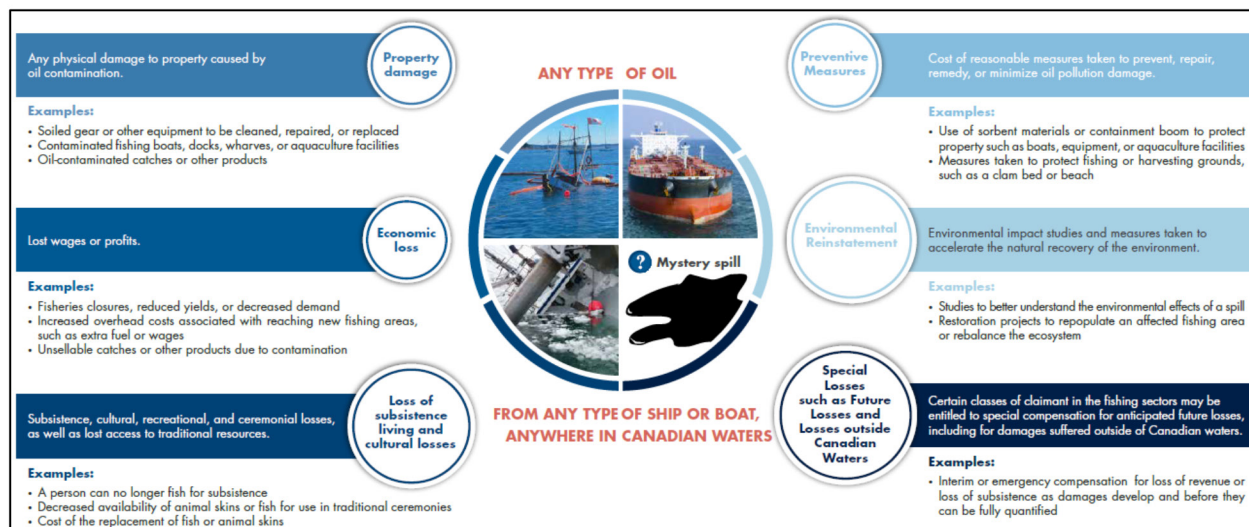
This presentation, based on the Fund’s Handbook, is a starting point for a broad audience across the fishing sectors, including workers and business-owners, Indigenous communities, and those pursuing commercial and non-commercial activities, both in fresh and salt water.

This presentation will be useful to all engaged in any of the following activities:

- Fishing
- Aquaculture and Mariculture
- Harvesting
- Hunting of animals
- Processing, preparation, packaging, and distribution
- Tourism and hospitality
- Management of ports, harbours, or marinas

While those in the fishing sectors can benefit from the Fund as claimants, ships used by the sectors can also cause oil pollution. For instance, fishing vessels have been responsible for one in four claims submitted to the Fund over the last 30 years. Once we pay a claimant, we take all reasonable measures to recover from the responsible person — this is the “polluter pays principle”.

The term “fishing sectors” applies to commercial and non-commercial activities in both fresh and salt water. These activities include fishing, aquaculture, harvesting, hunting, and processing. More broadly, all those who sell or otherwise use or depend upon fish, shellfish, seafood, seaweed and marine plants, crustaceans, molluscs, and other animals are also covered.



DETERMINATION FACTORS OF FINANCIAL DECISION AND ITS IMPACT ON ECONOMIC GROWTH IN SINDH PROVINCE: A CASE STUDY

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The monetary development of the areas/urban communities in Sindh is moderately lower than in different districts. This exploration demonstrated that budgetary choices majorly affect monetary development. The metropolitan spending plan decides financial development. While the nearby government spending plan centers around current spending as opposed to improvement spending, financial development will decrease. The examination additionally discovered that the monetary condition, the lawful condition, the political condition and the social condition decide the money related choices in a neighborhood government. What's more, the examination infers that the political condition impacts financial development while the monetary, lawful and social condition does not influence monetary development.

1. INTRODUCTION

Maintainable improvement stresses the parity of the three mainstays of social, financial and ecological viewpoints. Provincial advancement procedures are intended to address local issues so as to improve their jobs and capacities in overseeing social, monetary, social, instructive and social prosperity. Territorial advancement is a vital piece of the improvement of a district, particularly in country territories, which are exceedingly defenseless and are confronting real changes even with worldwide change. This change must consider parts of manageable improvement. Financial development is a marker of local advancement. Financial development is additionally a procedure that builds the capacity of a nation's economy to create products, for example, merchandise and ventures, as territorial wages.

The monetary development of an area is emphatically affected by the money related choices of the local government. As indicated by the International City/County Management Association (ICMA), there are a few factors that impact nearby government budgetary choices, including:

(1) political condition, (2) monetary condition, (3) social condition and (4) lawful condition. What's more, provincial income majorly affects the money related choices of local governments. For the districts, charge income is one of the fundamental wellsprings of income, Peacock and Wiseman (1993)), which expresses that the open has an expense resilience on which the open can demonstrate the measure of assessment gathering government to back government spending. The hypothesis of Peacock and Wiseman recommends that financial advancement will prompt an expansion in duty accumulation, regardless of whether the assessment rate does not change, and expanding charge incomes will build government spending. The districts need to deal with their family units autonomously, so the administration needs to improve its open administrations. Consequently, the territorial spending plan is counter-intuitive when the extent of the financial plan for routine costs is higher. Advancement spending is a drive by territorial governments to expand open trust so as to improve provincial monetary development. In view of the past explanation, it is essential to direct more inside and out research and investigation so as to discover monetary choice equations that expansion financial development.

Table 1 Path Coefficient Value

Variable	Financial Decision (Y1)	Economic Growth
Financial Decision		0.799
Political Environment (X1)	0.305	0.175
Economic Environment (X2)	0.486	-0.015
Social Environment (X3)	0.130	-0.012
Legal Environment (X4)	0.127	-0.008
Local Government Revenue (X5)	0.172	0.092

Sources: PLS Results (2019).

(Continued on next page)

2. LITERATURE REVIEW

2.1. Financial plan for provincial income and local consumption as a component of the usage of decentralization

Financial development is the procedure of constantly changing the monetary states of a nation. Elements that impact monetary development incorporate HR, characteristic asset elements, science and innovation and culture components, and nearby government money related choices. As noted in the Guidelines for getting ready spending making arrangements for nearby networks, budgetary choices in the areas are ending up progressively perplexing. Current territorial money related choices are exceptionally delicate to their political, monetary, social and legitimate condition. As referenced above, ICMA expressed that there are 4 primary factors that impact the territorial government's money related choices. What's more, territorial income, particularly nearby income, assumes a job in the money related choices of local governments. Territorial Budget and Expenditure (APBD) structure the premise of local money related administration in a budgetary year. APBD is the financial plan for provincial income and consumption identified with the usage of decentralization in a given money related year. The gathering of all territorial income is gone for accomplishing the goals set out in the APBD. Additionally, all local uses and duties that load the area as a feature of the usage of decentralization will be completed as per the figures and goals characterized in the APBD. As the APBD is the establishment of provincial money related administration, the APBD is additionally the reason for territorial budgetary control, investigation and oversight exercises. APBD is readied utilizing an exhibition approach, which is a planning framework that organizes endeavors to accomplish work or yield from arranged cost assignments or set information sources. The dimension of pay evaluated in the APBD is a sensibly estimated gauge that can be acquired for each wellspring of pay.

2.2. Political Condition

Political interests can be clarified based on spending things in the financial plan, for example, routine spending things: there are government interests, government workers. Barrier and security spending have the interests of the administration, military, police, etc. Each intrigue enters the political framework through ideological groups that can battle for the interests of each gathering. In this way, the spending banter has limited to the discussion at the dimension of gathering elites in the administration or in parliament. This issue cannot be isolated from the comprehension among business and governmental issues. Each financial, political, social, and so forth issue is inseparably connected to the issues of different divisions. With the goal that the examination of critical thinking requires interdisciplinary information. No gathering can develop without solid money related help. Accounts are expected to unite associations, enlist individuals, track objectives, and make pictures, crusades, and that's just the beginning. In the first place, every money related need of the ideological groups was secured by enrollment charges. Solid ideological connections between ideological groups and individuals make it troublesome for ideological groups to fund-raise from individuals. In any case, given the adjustments in the social structure of society and the plan of an undeniably mind-boggling popularity-based arrangement of administration, there are no ideological groups that live completely on participation expenses (Supriyanto and Lia, 2012).

Backing for ideological groups has been presented in Indonesia, in spite of the fact that the figure is as yet thought about excessively low. Thus, party administrators look for budgetary assets by performing degenerate practices. Deviations emerging from budgetary arrangements require better approaches for figuring and overseeing spending plans to give administrations to the network. Unilateralism, the battle for gatherings and their very own establishments are conspiracy to general society. The political position and the region of the spending plan have dependably been a discussion among political specialists. Spending issues are viewed as issues of government, organizations, administration, power, philosophy and legislative issues and markets, just as socio-social issues and transient political economy. The meaning of the degree and cutoff points of the political economy of the family unit is regularly viewed as indistinct and can be found all over the place. When all is said in done, nonetheless, financial strategy is the space of the job of the state, as it is viewed as an examination of political economy ponders. Hence, political power is an imperative factor in the arrangement and arranging of spending plans. The enormous issue of the political economy of the monetary allowance must be translated as an apparatus to battle against the privileges of residents as opposed to oppressing individuals. For this situation, the job of the state has the option to reformulate its job and capacity to support the general population (Wildavsky and Caiden, 2004).

2.3. Monetary Condition

Monetary elements impact money related choices in regions. Monetary conditions or conditions, for example, business cycles, expansion and loan costs, unequivocally impact neighborhood government choices on territorial spending. The financial downturn influenced provincial spending plans in two different ways. To start with, pay may decrease, particularly pay, for example, deals or pay charge, which are progressively recurrent. Furthermore, amid the retreat, state and bureaucratic incomes were regularly cruel, with the outcome that between state backing to neighborhood governments declined.

(Continued on next page)

Swelling makes vulnerability in assessments of nearby government income and consumption. High swelling strongly affects spending conveyance choices. Indonesia, as a creating nation, is regularly gone up against with high swelling, which is as yet a macroeconomic issue that stresses government and society. Expansion is characterized as the inclination to build the cost of products for merchandise and enterprises as rule and proceeds. The Government's strategy on raising the cost of fuel oil (BBM) is one of the elements driving the high rate of expansion in Indonesia, as the ascent in fuel costs straightforwardly influences the general value climb, particularly for sustenance and other fundamental needs. High and precarious swelling will disturb financial strength, make business arranging progressively troublesome, diminish enthusiasm for network venture, hamper government advancement plans, and impact the spending structure set toward the start of the monetary year. Changes in financing costs can likewise influence the financial plan, in spite of the fact that the effect at nearby dimension isn't huge yet will be powerful at national dimension as the state needs on-going advances.

2.4. Social Condition

The social condition, for example, populace changes, age dispersion and huge individual salary in the local spending plan. Expanding the extent of the populace, while keeping family unit costs unaltered, will partition the cost dimension by the quantity of relatives, bringing about a lower monetary scale, ie the expense per individual in the family will be lower. As money related responsibilities don't diminish in extent to populace misfortune, networks with declining populaces face challenges in lessening use. Training spending, open security is the financial plan most influenced by the age conveyance of the populace. A few past investigations have reliably demonstrated that the development of individual pay essentially influences the measure of metropolitan family units. High-salary families frequently request more and better administrations from the legislature, in spite of the fact that these family units will in general be increasingly constrained at the state level.

2.5. Lawful Condition

Lawful components that impact budgetary choices are the spending balance, the order of the focal government and nearby assessment guidelines. While allotting provincial use, account must be taken of the family unit balance. The focal government requests that the regions spend at any rate 20% of the all-out spending plan. Both focal government and commonplace governments offer commands to nearby governments. This represents an issue if the focal government does not give the full assets to the execution of the command. The neighborhood specialists must lower the other administration costs. The districts will experience the ill effects of the subsidence, which runs connected at the hip with the Center's order and reserve funds in local spending.

The presence of confinements on area/city governments to tap specific wellsprings of pay, limit charge climbs, make a parity, and request explicit administrations and dimensions of administration is a mix of variables that have caused gigantic financial worry at neighborhood government level for three decades ,

2.6. City Income

The financial improvement in Sindh covers all parts of the economy of the network, both the life of country networks and urban networks, with the primary target of improving and improving the expectations for everyday comforts of the general population of Sindh. Financial advancement is done by concentrating on endeavors to develop the monetary segment, making utilization of the maximum capacity claimed by both the capability of normal assets and HR.

2.7. Theory

In view of the foundation of the issue, the definition of the issue and the examination structure and the speculation of this investigation are:

1. The political condition, the monetary condition, the social condition, the legitimate condition and the nearby income impact the budgetary choices in the territory of Sindh.

2. Monetary choices influence financial development in Sindh territory.

(Continued on next page)

3. RESEARCH PROCEDURE

The populace in this investigation is the area/city work unit in Sindh territory. Research factors comprise of a variable political condition, monetary condition, political condition, social condition, lawful condition, neighborhood income, money related choices and financial development. The information investigation utilized in this examination is the incomplete least squares (PLS) approach. PLS is a model of basic condition displaying (SEM) in light of segments or variations.

4. RESULTS AND DISCUSSION

4.1. Result

4.1.1. Test Results of the Measurement Model (Outer Model)

The external model test begins by evaluating or assessing parameters, in particular, figuring the PLS calculation. In view of the yield examination, the estimation display (external model) can be assessed by checking the concurrent legitimacy, discriminant legitimacy, and unwavering quality. All focuses are substantial and dependable inquiries.

4.1.2. Way Coefficient Examination Results

The auxiliary model in PLS is assessed utilizing the reliant variable R square and the free factor way coefficient esteem, which at that point assesses their hugeness dependent on the t-measurement estimation of every way. The PLS result program calculation for assessing the estimation of way and R squared coefficients is appeared Table 1 underneath:

4.2. Discourse

4.2.1. Effect of the Strategy Condition on Money Related Choices

The consequences of the investigation presumed that the political condition spoken to by the inclusion of the network in the planning procedure had a positive and huge effect on the money related choices taken by the neighborhood government. In light of the past portrayal, political interests impact the dispersion of consumption contained in the state spending plan. Each issue has interests, both the interests of the legislature and the network and political interests. From focal government spending, for example, routine consumptions, these shopping centers are in light of a legitimate concern* for government employees. The discussion in setting up the financial backing was for the discussion at the dimension of the gathering elites in the EU

Government or Parliament. The technique for participating in political interests should be possible through studies in the network when ideological groups make breaks or amid the muscling. The incorporation of political interests in planning will influence the nature of the financial backing in help of territorial improvement. The conveyance of spending is firmly affected by political interests. The administration's budgetary choices in the dissemination of foundation spending, monetary spending, and socio-social spending rely upon the exchanges between the assembly and the official. Political interests are not generally in accordance with the interests of territorial advancement, which regularly prompts clashes in planning. This issue cannot be isolated from the comprehension among business and governmental issues. Each financial, political, social, and so on issue is inseparably connected to the issues of different areas. With the goal that the examination of critical thinking requires interdisciplinary information.

4.2.2. Effect of the Monetary Condition on Money Related Choices

Monetary factors emphatically impact the money related choices in the districts. The consequences of the examination demonstrate that the monetary condition impacts the money related choices of the regions. This implies neighborhood governments in Sindh Province are thinking about financial factors in deciding the appropriation of foundation spending, monetary consumption and socio-social use, outstandingly swelling, loan fees and financial cycles in the area. The monetary conditions or conditions as business cycles, expansion and loan costs unequivocally impact the choices of regions in the assignment of territorial consumption.

(Continued on next page)

The high expansion firmly impacts the choices about the dispersion of the consumption. Indonesia, as a creating nation, is frequently gone up against with high swelling, which is as yet a macroeconomic issue that stresses government and society. High and unsteady swelling will upset the steadiness of the economy, make business arranging increasingly troublesome, decrease the enthusiasm of open speculation, hamper the administration's advancement plans, and impact the financial structure built up toward the start of the monetary year. Changes in loan costs can likewise influence the financial plan as it is one of the fundamental suppositions or presumptions in planning. Be that as it may, the effect at nearby dimension isn't substantial, yet will have a noteworthy effect at national dimension as these suppositions change from their unique esteem. This influences the dimension of the pay part, both on government income and on government use and spending shortages, which lead to changes in the dimension of spending financing.

4.2.3. The Impact of the Social Condition on Money Related Choices

The social condition significantly affects the money related choices of regions, particularly in the readiness of territorial income and use spending plans. For state funded instruction, open wellbeing is impacted by the age conveyance of individuals in the region of each territorial government. Changes in the populace in a territory will influence provincial spending choices, in light of the fact that the more individuals live in one zone, the broader assets will be made accessible to the administration. A marker of the measure of the General Allocation Fund is the all-out populace and territory. What's more, the populace likewise impacts local spending, as the substantial populace in a territory of development organizers is viewed as the capital of advancement and, in the meantime, the weight of improvement. Vast populaces can likewise be considered as territorial resources if network individuals can improve their quality and aptitude or abilities to build national creation.

The age conveyance of the occupants of a zone will impact the monetary choices of the district. One part of spending that is affected by age dissemination is instruction spending. The more school-age occupants, the higher the spending on training.

A few past examinations reliably demonstrated that the development of individual pay essentially influenced the dimensions of city spending plans. High-salary families regularly request more and better administrations from the legislature, despite the fact that these families will in general be progressively constrained at the state level.

4.2.4. Impact of Ecological Factors on the Monetary Choice

Legitimate components with spending asset reports, changes in spending plans and incorporated guidelines have a positive and noteworthy effect on the monetary choices of regions. While allotting local use, account must be taken of the family unit balance. Harmony between provincial income and consumption. Spending changes additionally bigly affect monetary choices. On account of a salary change, acquiring must likewise be balanced. For instance, if the nearby government sets an inn charge focus of IDR 100 million for one year in the year being referred to, incidentally, the primary semester outflanked the financial plan for changes in the territorial burning through segment. Neighborhood governments can expand the quantity of exercises that will be changed in accordance with the evaluated pay rate that is typically accomplished for the current year.

4.2.5. Effect of Local Introductory Pay on Money Related Choices

Nearby income is a factor that influences the financial plan. The budgetary choices of neighborhood government depend intensely on the measure of nearby salary. The higher the nearby income, the higher the local spending. Territorial self-rule obliges locales to back local needs with wellsprings of salary that can be collected by the provincial government. The degree of the area's capacity to build its local starting income is impacted by wellsprings of income, strategies for gathering civil income, and observing metropolitan income.

The pay wellspring of the area/city has arrangements. The dissemination of focal, commonplace and regime/city income has been represented by government guidelines, and the measure of income that can be created by the territorial government has likewise been directed by law. The strategy for gathering the incomes of the regions unequivocally impacts the dimension of territorial salary.

4.2.6. Effect of Monetary Choices on Financial Development

Financial development, destitution and joblessness are positively inseparably connected to government contribution as strategy producers and the ability to utilize the monetary allowance to animate the economy through the local income and use spending plan, which is required to quicken the economy. Because of this affirmation, the improvement of a territory with pointers of monetary development and success of the network unequivocally relies upon the money related choices of the districts. With the quick turnaround of the economy, new monetary open doors are normal which will unquestionably pull in specialists who are attempting to diminish joblessness and improve individuals' expectations for everyday comforts as opposed to neediness. The aftereffects of this investigation mean to perceive how the APBD government is affecting development, diminishing joblessness and destitution in Sindh.

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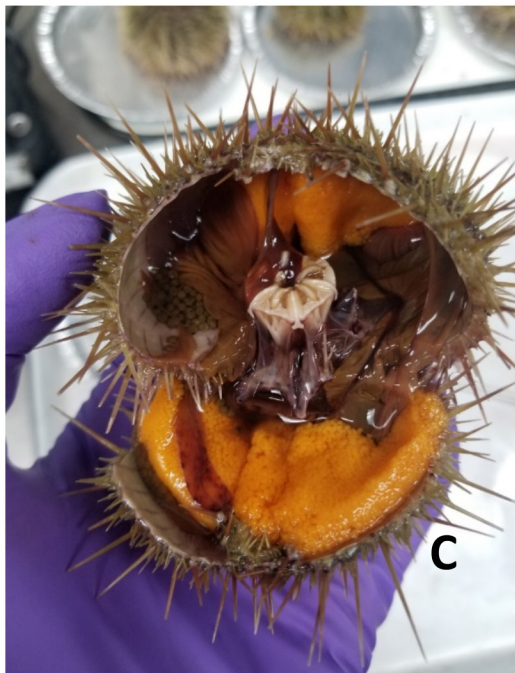
FEED AND TEMPERATURE EFFECTS ON LAND-BASED GONAD ENHANCEMENT OF SEA URCHINS IN BRITISH COLUMBIA, CANADA

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When sea urchin population growth is uncontrolled, they can over-graze and decimate macroalgal beds, creating areas termed “urchin barrens”. Sea urchin gonad enhancement is a proposed method to remove sea urchins from barren grounds, promote the re-growth of macroalgal beds, and produce a highly-valued marketable product. Prepared feeds need to be developed that not only increase gonad yield, but also impart appropriate market colour and flavour. This project assessed the effects of two prepared diets (V10.1.9 and V10.1.10) and a natural feed (bull kelp, *Nereocystis luetkeana*) and three seawater temperatures (8, 12, 16°C) on gonad yield and quality for both the green (*Strongylocentrotus droebachiensis*) and red (*Mesocentrotus franciscanus*) sea urchins held under laboratory conditions for 9–12 weeks. Green urchins fed the prepared diets had overall higher gonad yields and better colour than the green urchins fed kelp at all three temperature treatments, with V10.1.9 producing the highest yields for all three temperatures and V10.1.10 producing the best colour at 16°C (Fig. 1). Red urchins fed V10.1.10 at 12°C had the highest gonad yield out of all the treatments, while the red urchins fed kelp at 8°C had the lowest. The gonad colour was better overall with the red urchins fed V10.1.10, with the best colour again obtained at 16°C, which is the same trend as seen in the green urchins. The prepared feeds tested produced high gonad yields and appropriate market colour in both sea urchin species.

A



B

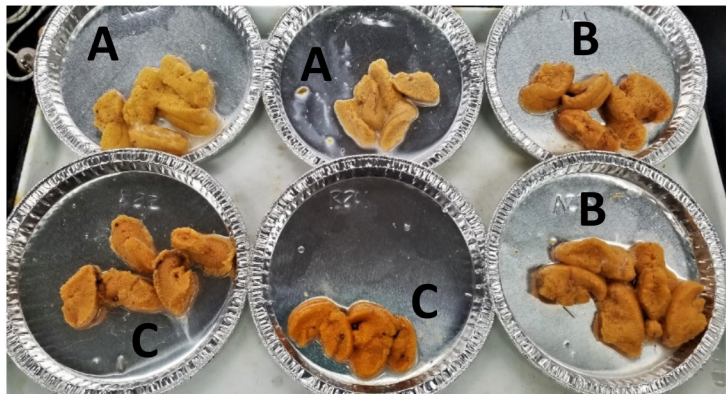


Fig. 1. (A) Green sea urchin split open and showing gonad. (B) Representative gonads taken from green urchins fed different diets. A: kelp (*Nereocystis luetkeana*), B: prepared feed V10.1.9, and C: prepared feed V10.1.10.

HISTAMINE AND PROTEOLYTIC BACTERIA LEVELS DURING FERMENTATION OF *Carcinus maenas*

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Carcinus maenas, commonly known as green crab, is an aggressive invasive species found on the east coast of North America. The proliferation of this species has led to significant habitat loss and predation of economically significant bivalves such as soft shell clam. Mitigation efforts have focused on finding marketable uses for green crabs, including the recent development of a soft shell fishery. Although this pathway holds significant potential to decrease green crab levels while increasing revenue for fishers, the financial viability of a soft shell market relies on the valorization of catch unsuitable for this use. Investigations of alternative uses for excess crab have included development of value-added products such as broth and jerky products. The purpose of this study was to assess the feasibility of producing a fish sauce-style condiment from invasive green crab.

Fermented fish sauce is a staple of Southeast Asian cuisine, consisting in its simplest form of a spontaneous fermentation of salted, whole fish. Whole crabs were caught off the coast of Maine and frozen until use. Crabs were finely chopped in a bowl cutter and combined with Kosher salt (10, 20 or 30% w/w). All treatments were prepared in triplicate and were fermented at 24°C. The product was sampled after 60, 90, and 120 days of fermentation. Prototype product, as well as several commercial fish sauces, were assessed for characteristics including pH, moisture content, water activity, browning index, total volatile base nitrogen, amine nitrogen, presence of biogenic amines (determined by HPLC using a Waters AccQ·Tag) and level of proteolytic microorganisms. Experimental sample data was analyzed by MANOVA in R studio followed by Tukey's HSD post hoc test to determine the effects of fermentation time and salt level.

No significant differences ($p > 0.05$) in histamine content at different salt concentrations or time points were observed, the average histamine content in experimental samples being $67.1 \pm 16.4 \mu\text{g/mL}$. The population of proteolytic microbes was significantly lower in high salt treatments, compared to 10% salt samples. The majority of tests conducted, including amine nitrogen, a marker for protein degradation characteristic of the fish sauce fermentation, were unchanged between days 60 and 120, indicating that the majority of proteolysis occurred early on in the fermentation. Data from this study suggest that a fermented condiment resembling commercial fish sauce can be prepared from green crab. Additional characterization of sensory qualities of such a product should be completed to assess market potential.

EFFECTS OF MINIMAL PROCESSING ON THE MICROBIAL SAFETY OF KELP

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Recent increases in production of aquacultured kelps in the United States has driven interest in value-added processes that can be used to extend the shelf life of this highly perishable crop. Although Maine leads the US in production of kelp, processing infrastructure remains decentralized and largely inaccessible to growers. A number of low-input processes, including freezing, salting and fermentation, have been suggested as ways for growers to minimize spoilage of unheated kelps during storage and distribution, but the potential survival of foodborne pathogens in these products is unknown.

In this study, the effects of fermentation and dry salting of seaweed on the survival of bacterial foodborne pathogens *Salmonella enterica*, *Listeria monocytogenes*, *Staphylococcus aureus* and *Vibrio vulnificus* during refrigerated storage was assessed. A sauerkraut-style seaweed was prepared from varying ratios (25, 50 or 75% wt/wt) of sugar kelp or winged kelp (*Saccharina latissima*, *Alaria esculenta*) and cabbage. Mixtures were inoculated with *Lactobacillus plantarum* and *Leuconostoc mesenteroides* as well as relevant pathogens. Pathogen population was monitored using cultural methods during fermentation and sixty days of refrigerated storage. Additionally, winged kelp was combined with various levels of salt (2-20% wt/wt) and inoculated with pathogens. Pathogen survival was monitored for up to 90 days during refrigerated storage.

All four pathogens investigated were inactivated within 14 days in fermented sugar kelp regardless of the ratio of kelp utilized. Pathogens were equally susceptible in fermented winged kelp with the exception of *Salmonella* in the 75% kelp sample, which survived for 49 days. Decreases in pathogen population (from an initial level of approximately 5.0 log CFU/g) roughly mirrored the decrease in pH caused by fermentation. Conversely, *Salmonella*, *L. monocytogenes* and *S. aureus* survived in salted winged kelp for a minimum of 23 days regardless of salt level applied. *V. vulnificus* was the most susceptible to salting treatments, becoming undetectable after enrichment between days 7 and 14 of storage in all treatments.

Although prior research has documented the acceptable quality of the seaweed products investigated in this study, data generated from this work suggest significantly lesser risk of foodborne illness from consumption of a fermented, as opposed to salted, product. The pH decrease resulting from fermentation inactivated all inoculated pathogens within two weeks of processing. Salting, however, regardless of salt level, was inadequate to mitigate microbial risks. Investigators suggest that processors pursuing this approach adopt a validated microbial kill step.

GC/MS ANALYSIS OF PHYTOSTEROL CONTENT OF SALMON *Salmo salar* AND THEIR DIETS IN A MICROBIAL OIL FEEDING EXPERIMENT

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Fishmeal and fish oil have been considered the most digestible and nutritious ingredients for farmed fish, but there is a shift towards more sustainable alternatives. In this study, fish oil was replaced with microbial oil based equivalents. Atlantic salmon (*Salmo salar*) were fed four different diets, two included microbial oil at low and high levels of inclusion, while one contained fish oil as well as canola oil, the fourth was a control diet containing traditional fish oil. Sterols were Identified and quantified in salmon muscle tissue after feeding trials with different levels of microbial oil, fish oil, and canola oil using gas chromatography/mass spectrometry (GC/MS) and gas chromatography/flame ionization detection (GC-FID). Results indicate that cholesterol content decreased in the microbial diets as compared to the fish oil diet. There were phytosterols identified in the microbial oil diets that were not present in the fish oil diet. In the salmon muscle, cholesterol content decreased in fish fed the fish oil/canola oil diet, but did not change in salmon fed the microbial oil diets.

THE EFFECTS OF FISH SIZE, STOCKING DENSITY, AND PHOTOPERIOD ON JUVENILE LUMPFISH AGGRESSION

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Due to the high demand for lumpfish as cleanerfish in salmonid ocean farming operations, increasing hatchery production and rearing efficiency is of great importance. As juveniles, lumpfish are cannibalistic which is controlled, to some extent, though frequent size grading of the fish, however, cannibalism still occurs. Understanding and mitigating for factors that exacerbate aggressive behaviors in juvenile lumpfish, which can lead to cannibalism, would help achieve the goal of increasing juvenile production in the hatchery. We hypothesize that lumpfish cannibalism is linked to a specific ontogenetic period related to fish size and can be exacerbated by various stressors such as stocking density and photoperiod. To test this hypothesis, we subjected two different size classes of juvenile lumpfish (5g and 10g) to varying densities (40g/L, 65g/L, or 90g/L) under different photoperiod regimes (ambient, constant low light, or constant bright light) for a 10-week duration in winter 2022. Fish growth, survival, and aggression were measured biweekly, and stocking densities adjusted to baseline levels biweekly by removing any necessary fish. Final results, including daily growth rates, overall mean percent growth, survival, and occurrence and severity of fish aggression, as well as recommendations for lumpfish facilities, will be shared with session participants.

MICROBIAL AQUAFEED PRODUCTION FROM LIGNOCELLULOSIC SIDESTREAMS VIA AN AEROTOLERANT CELLULOLYTIC CONSORTIUM

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Lignocellulosic biomass is globally abundant, and can be found in food manufacturing side streams and post-harvest residues, representing a promising feedstock for microbial bioconversion to food and feed ingredients, and this is of particular interest for the international aquaculture industry.

However, recalcitrance of these materials has limited their application, and pretreatment, sterilization, and enzymatic hydrolysis are bottlenecks to economic feasibility. Through consolidated bioprocessing, aerotolerant thermophilic cellulolytic consortia can be applied to overcome these challenges, achieving rapid and complete cellulose conversion into short-chain fatty acids (SCFAs) in less than 48 hours (Fig 1A-B).

These SCFA cocktails have now been confirmed to be an excellent substrate for the growth of food grade strains such as *Candida utilis*, *Yarrowia lipolytica*, and *Corynebacterium glutamicum*. *C. utilis* has been shown to produce up to 30 g/L cell dry weight when cultivated on SCFAs in bioreactors, with 43% protein and 18% lipids containing up to 50% polyunsaturated fatty acids. (Figure 1C)

Importantly, the selective thermophilic and anaerobic conditions of this mixed culture process can be performed without sterilization, which could significantly reduce processing cost. The cellulose-derived microbial biomass demonstrated here represents a previously untapped marine-free aquafeed ingredient, and can contribute to increased food security and a circular bioeconomy.

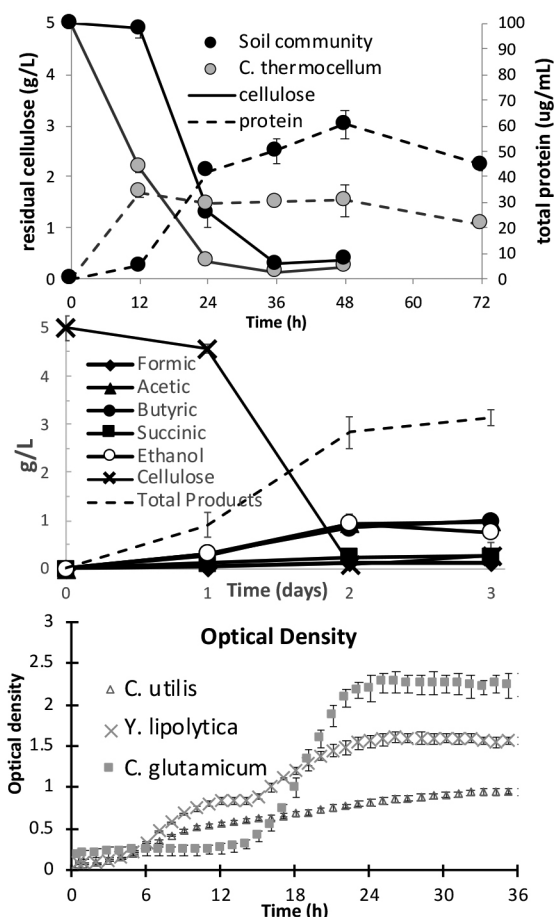


Figure 1A) Anaerobic cellulose utilization by both *Clostridium thermocellum* and an enriched consortium. B) Production of short chain fatty acids (SCFA) and ethanol by *C. thermocellum* and an enriched consortium C) Cultivation of food grade microorganisms on cellulose-derived biogenic SCFAs

FISH WEAR THEIR IMMUNE SYSTEMS ON THE OUTSIDE: WHAT THIS SLIME MEANS AND HOW TO PRACTICALLY MEASURE IT

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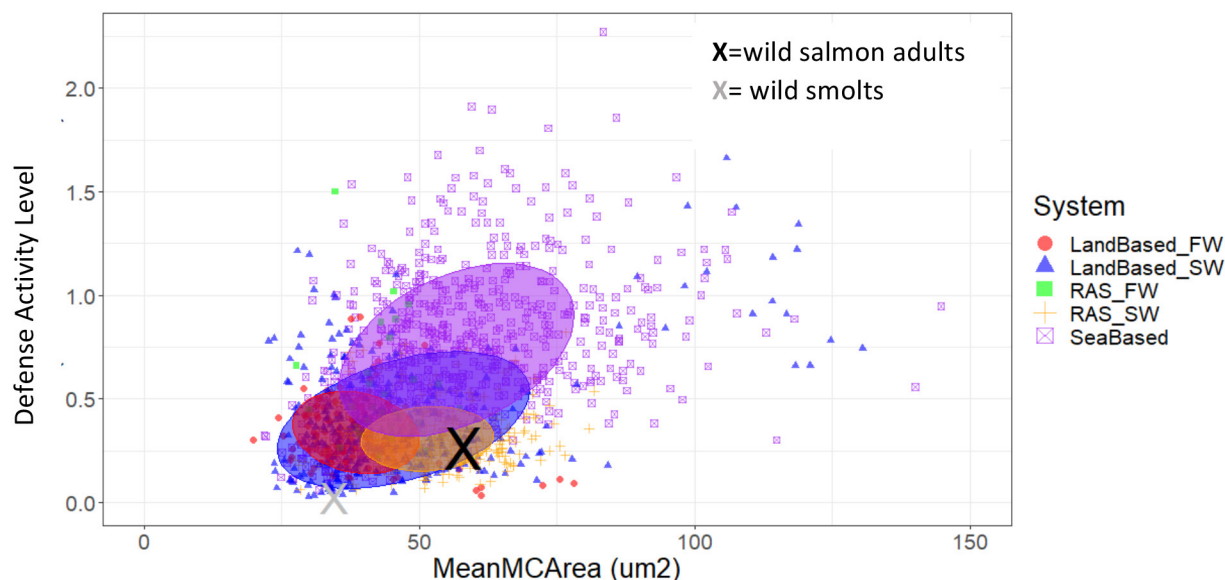
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The mucosal immune system of fish is active on the external surfaces of skin and gills and on the internal surface of the gut, employing naturally produced antiviral, antifungal, antiparasitic and antibacterial substances to maintain homeostasis. The histological architecture of these three tissues is essentially the same, with changes in the mucous cell sizes and number and changes in the epithelial cells typical of a broad range of gill diseases, skin responses to external challenges and dietary composition.

It has been possible since 2010 to quantify these mucosal responses, which show clinically important results. The trademarked Veribarr™ method has been applied in 9 farmed species, in 7 countries and 3 tissues in over 60 lab scale and commercial scale trials and monitoring. Quantidoc AS now has a unique and large database about healthy farmed and wild salmon in 7 different systems gathered over 10 years. This comprises RAS FW, RAS SW, landbased FW and SW, open sea cages, closed seabased systems and natural fjords, and salmon sizes from less than 100 g to over 6 kg giving tens of thousands of objective, quantitative, statistically robust data points and metadata.

The natural barriers of gills, skin and gastrointestinal tract of the salmon are the most relevant and sensitive health probes possible. Differences from healthy barriers can be detected early and with statistical robustness. Veribarr™ Grid allows early diagnosis of changes to give new systems time to bring fish back to a healthy status. The figure below shows the lamellar mucosa of the gills which represent 50% of the surface area of the fish and are constantly reacting to waterborne substances. The talk will include standardized sampling and tissue handling protocols.

Mucous Cell Size and Defense activity in Gill Lamellae, n=1464, CI=50%



REPRODUCTIVE STERILITY IS FOUNDATIONAL TO SUCCESSFUL COMMERCIALIZATION OF GENOME EDITING FOR AQUATIC SPECIES

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Aquaculture continues to grow to meet the consumption of an increasing world population and aquaculture breeding must move quickly to help producers to meet the expectations of consumers and retailers in quality, health and welfare. Genome editing offers the opportunity to make targeted changes in the genetic sequence, introducing variation that will result in rapid and substantial improvements in performance, health and welfare. Such variation exists naturally at low levels in the population, but genome editing allows for the fast and effective introduction of favorable variants into the farmed lines making them better adapted to cultivation in captivity. Where favourable legislation and consumer demand exists, aquaculture breeders seek to identify genes and variation that improve health, welfare and performance, and have the opportunity to employ genome editing to develop strains that facilitate more sustainable production. Traits of interest for which research programs are currently active include disease resistance, growth and yield.

To responsibly introduce animals which are the product of genome editing into commercial productions systems, it is imperative to ensure that the animals in production are infertile. Research is underway to use genome editing to deliver better control of sex and fertility to ensure biological containment of any additional introduced traits and reproductive sterility has several benefits for production on its own. Our work to deliver sterile animals as a product of genome editing at a commercial scale will ensure that additional traits of interest can be introduced into an infertile background quickly, while simultaneously addressing concerns associated with any risk of unintentional gene flow to wild populations.

REVIEW OF NON-TOXIC AND MECHANICAL PRE-TREATMENT PROCESSES TO PREPARE NATIVE CHITIN FOR HIGH-STRENGTH NANOMATERIAL APPLICATIONS

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Chitin is a polymer chain of N-acetylglucosamine and glucosamine. It provides structure and stability to the exoskeleton of crustaceans, insects and fungal cell walls. Intermolecular hydrogen bonds between polymer chains create a very tight crystalline fibre network with excellent mechanical properties. Chitins' antimicrobial properties and high biocompatibility make it an attractive polymer source to produce nano-materials such as gels, fibers, or films

To date, commercial utilization of native chitin has been limited due to its insolubility in aqueous solutions and most common solvents. Instead, chemical modifications such as deacetylation (conversion to chitosan) or depolymerization are used to make chitin soluble and easier to work with but consequently, chitin loses some of its mechanical stability. Other approaches for chitin dissolution employ toxic chemicals, potentially preventing its use in biomedical and food applications. Studies in recent years have produced several environmentally-friendly and non-toxic pre-treatment processes which can reduce chitin's crystallinity and tight macrostructure without destroying or changing the native polymer chain. This review (oral or poster) will begin with a description of the macrostructure of crustacean cuticles, followed by a summary of non-toxic and mechanical pre-treatment processes which can be applied to prepare the fiber for nanomaterial applications. The effects of macrostructure on pre-treatment are discussed.

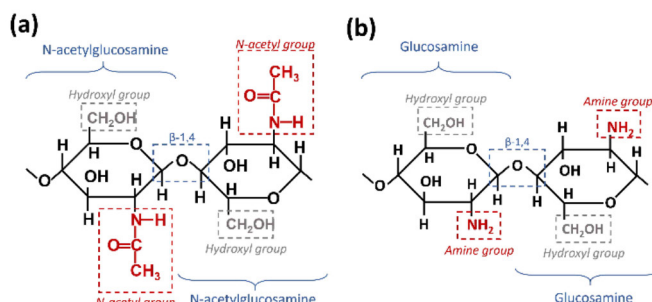


Figure 1: Chemical structure of the two building blocks of chitin - N-Acetylglucosamine (a) and glucosamine (b)

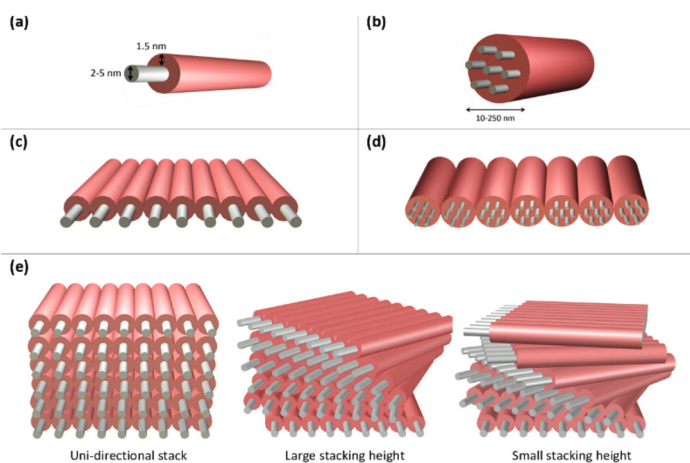


Figure 2: Macrostructure of crustacean cuticles. (a) Chitin nanofibril consisting of 18-24 individual chitin polymer chains, surrounded by a protein coat made up of chitin-binding proteins (CBP); (b) Chitin nanofiber consisting of several nanofibrils (c) Chitin nanofibril sheet, found in many arthropod species; (d) Chitin nanofiber sheet, found e.g., in crab; (e) Nanofibril or nanofiber sheets stacked in layers with varying degrees of twist, forming the cuticle.

PREDICTING ATLANTIC SALMON (*Salmo salar* L.) SMOLTIFICATION READINESS USING GILL GENE EXPRESSION PROFILES

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Smoltification of anadromous salmonids is a complex physiological process required for migration from freshwater to seawater environments. This process includes changes in fish condition factor, body color, behavior, and molecular profiles of muscle, brain, gill, and plasma, among other tissues. For wild fish, the transition to higher salinity environments is often gradual, with various amounts of time spent in estuaries prior to full seawater migration. However, smolts produced for aquaculture in land-based, freshwater hatcheries are transferred directly to seawater without brackish acclimation. This transfer also includes additional stressors such as crowding, transport, and handling. Thus, seawater transfer of salmonids in aquaculture is a key event that, if done sub-optimally, can lead to compromised immunity, lower osmoregulatory capacity, stunted growth, and increased rates of mortality. Several biochemical tests based on Na^+/K^+ ATPase (NKA) activity and mRNA abundance of NKA isoforms and related cotransporters are used by the industry to predict the readiness of smolts for seawater introduction. In the present study, a novel mRNA target for predicting Atlantic salmon smolt readiness is reported and benchmarked against industry standard markers. Multiple studies examining the expression of these genes from non-lethally sampled gill lamellae using RT-qPCR show that the predicative power in determining the life stage, smolt readiness, and response to environmental factors influencing smoltification (e.g. photomanipulation, functional feeds, etc.) are improved using the novel smolt gene. Furthermore, introduction of smolts to different salinities resulted in dose-response expression curves of the novel gene, with 100- and 300-fold increases in expression three days and three weeks post-seawater (25 ppt) transfer. This work demonstrates improvements to traditional smolt test markers and suggests that the use of several genes may offer improved insights on Atlantic salmon transfer to seawater.

CARDIORESPIRATORY FUNCTION AND SWIMMING CAPACITY OF ATLANTIC SALMON (*Salmo salar*) AT COLD TEMPERATURES

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The severity and frequency of challenging environmental conditions, such as marine heat waves, storms, hypoxia and extreme reductions in water temperature, continue to increase as a result of climate change. However, most literature on the *in vivo* physiological impacts of climate change on cultured fish species, including Atlantic salmon, is limited to the warm end of temperatures that this species can be exposed to in culture (i.e., 18-23°C). Temperatures at cage-sites in Atlantic Canada can also be 0-2°C for days to months at a time in the winter. Thus, it is crucial to understand how these temperatures affect the cardiorespiratory function, bioenergetics and swimming performance of salmon.

Mixed sex salmon (~ 675 g) were acclimated to 8, 4 or 1°C in seawater, or acutely cooled (from 8-1°C overnight), and changes in aerobic metabolism and cardiac function were measured during a critical swim speed (U_{crit}) test. Swimming capacity (U_{crit}) decreased by ~ 40%, and maximum metabolic rate (MMR) and aerobic scope [AS; MMR – standard metabolic rate (SMR)] were ~50% lower, for fish tested at 1 vs. 8°C (with values for 4°C acclimated fish intermediate between these values). Interestingly, fish acclimated to 1°C had a lower U_{crit} value than fish acutely exposed to this temperature, despite having statistically similar values for MMR, AS and anaerobic capacity (measured as post-exercise lactate levels). This suggests that factors independent of oxygen delivery, such as muscle contractile performance, may limit the swimming capacity of salmon when chronically exposed to temperatures approaching their lower thermal limit.

Fish tested at 8 and 4°C had the capacity to increase cardiac output (Q) and tissue oxygen extraction (MO_2 / Q) to meet the metabolic demand of increased performance. However, fish tested at 1°C were only capable of increasing one of these parameters. Fish acclimated to 1°C had a high resting MO_2 / Q and low Q (i.e., a high scope for Q), whereas fish acutely exposed to 1°C had a high resting Q and low MO_2 / Q (i.e., a high scope for MO_2 / Q). These data suggest that salmon do not have the ability to increase both parameters when exposed to 1°C and are interesting with respect to how acute vs. chronic exposure to cold temperatures affect the salmon's phenotypic plasticity in the context of supporting oxygen delivery during exercise.

Overall, these data show that the Atlantic salmon's metabolic and swimming capacity decrease considerably as water temperature is lowered to near 0°C, and suggest that this may make it difficult for them survive if challenged with other suboptimal conditions or stressful events.

Table 1. Maximum swim speed (U_{crit} : BL s⁻¹), MMR (mg O₂ kg⁻¹ hr⁻¹), SMR (mg O₂ kg⁻¹ hr⁻¹), AS, factorial scope for Q , and factorial scope for MO_2 / Q for all groups used in these experiments. Values without a letter in common are significantly different.

	8°C	4°C	1°C	8 to 1°C
U_{crit}	2.08 ^a	1.69 ^b	1.27 ^c	1.44 ^d
SMR	46.35 ^a	36.12 ^{ab}	41.70 ^{ab}	27.57 ^b
MMR	352.08 ^a	272.43 ^b	222.09 ^{bc}	184.83 ^c
Absolute AS	305.72 ^a	236.31 ^b	185.97 ^{bc}	147.26 ^c
Q (FS)	1.98 ^{ac}	2.42 ^{ab}	3.11 ^b	1.55 ^c
MO_2 / Q (FS)	3.42 ^a	2.86 ^a	1.58 ^b	2.90 ^a

COMPARATIVE PROXIMATE ANALYSIS OF WILD AND CAPTIVE LUMPFISH *Cyclopterus lumpus* EGGS SHOW DEFICIENCIES IN CAPTIVE EGGS AND POSSIBLE EGG QUALITY DETERMINANTS

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There is increased commercial interest in the production of Lumpfish (*Cyclopterus lumpus*, Linnaeus, 1758) as a biological control for sea lice infections in Atlantic salmon (*Salmo salar*). To ensure sustainability, reliable captive broodstock management is required however many aspects are still unknown. A key component in closing the life cycle of any species is improving survival within the egg. Thus determining the components that affect egg quality is a key step in improving the sustainability of any captive breeding program.

The present study attempts to assess the current difference in egg quality between wild and captive broodstocks. Egg samples and quality data have been collected from Norwegian wild (n=40) and Captive Scottish (n=44) stocks. There was a significant difference between wild and captive egg performances at all stages (fertilization, eyeing and hatch rates). There was high variation in hatching success in wild batches (0%-75%), whereas the majority of captive eggs did not reach hatching (6/44), those that did, hatching rates were significantly lower than wild eggs.

There was significant compositional difference between wild and captive stocks at the point of stripping, in lipids, fatty acids, minerals and pigments. The captive eggs were deficient in EFA's such as EPA and DHA as well as overall lipid levels 1/3rd lower than the wild stocks. The captive eggs were deficient in 8/12 measured minerals and overall pigment levels were 1/3rd of those in the wild stocks. Lumpfish eggs are characterized by a high level of neutral lipid fraction similar to those found in other species which exhibit lipid globules within the egg, and high levels of pigmentation were found within the eggs, with large levels of variation of individual pigments, which appear to reflect the observed natural variation found in wild caught fisheries. A total of 49 nutritional components were identified as having significant positive or negative relationships with hatching rate. EFA's such as EPA and DHA and Total N- 3 PUFA showed strong significant positive relationships with hatching rate as well as levels of saturated FA's such as 16:0. High levels of N-6 PUFAS especially ARA were strongly associated with poor egg quality in this species.

The present study is the first to address egg quality in Lumpfish, currently underperforming eggs are a substantial factor in the creation of a commercially viable broodstock. the present study has confirmed significantly poorer egg quality in captive derived broodstock, than the wild caught stocks, displaying significantly lower fertilization, eyeing and hatching rates. Poor egg quality is a major hurdle in successful commercial production of this new species. This work can provide key information to inform the creation of broodstock diets for lumpfish, as well as an important baseline in further egg quality work within the species.

CHANGES IN LUMPFISH *Cyclopterus lumpus* EGG COMPOSITION OVER EMBRYONIC DEVELOPMENT SUGGEST POTENTIAL EGG QUALITY DETERMINANTS

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The Lumpfish (*Cyclopterus lumpus*) is the most widely used cleanerfish in Atlantic Salmon aquaculture. To ensure sustainability, reliable captive broodstock management is required however many aspects are still unknown. The sustainable captive production of lumpfish is reliant on good egg quality from captive broodstock. A key component in closing the life cycle of any species is improving survival within the egg. Thus, determining the components that affect egg quality is a key step in improving the sustainability of any captive breeding program. At present all production is currently being met by wild caught eggs, which is unsustainable moving forward and opens up the species for genetic selection for desirable traits.

The present study assessed the compositional changes of fatty acids, minerals, and pigments in wild lumpfish eggs over the course of embryonic development in order to identify potential egg quality determinants. Samples were taken at 6 different time points, stripping, 24 hours post fertilisation (8DD), 60, 150, 200-degree days and hatched larvae (350DD). There was a significant decrease in levels of total Monosaturates, Saturates, Total N-3 and N-6 PUFAS over the course of embryonic development. However, changes in levels of specific FFA's varied. Levels of DHA and EPA decreased significantly between stripping and hatching. 18:4 N-3 and 20:4 N-3 also decreased significantly between stripping, early development and hatching.

Levels of sodium, magnesium and a number of minerals increased over development suggesting import into the egg over the developmental process. As was a number of changes throughout the developmental progress in mineral content. These findings suggest that some minerals may not be as a limiting factor in embryonic development for this species. Lumpfish eggs are well known for their diversity in egg colourations. With significant increases of Beta Carotene, as well as significant decreases in Canthaxanthin and astacene between stripping a hatch. Suggesting total pigment, not individual pigments being factors in egg quality for lumpfish

The present study sought to further address the issue of egg quality in Lumpfish. Currently underperforming eggs are a substantial limitation in the creation of a commercially viable broodstock. Areas of significant changes over development suggest potential egg quality determinates in lumpfish as well as an important baseline in further egg quality work within the species. This work can practically provide key information to inform the creation of broodstock diets for lumpfish, working to improve egg and juvenile quality to ultimately close the life cycle of this species.

POST OVULATORY AGING OF LUMPFISH *Cyclopterus lumpus* EGGS SUGGESTS SHORT OVERRIPENING WINDOW, WITH POTENTIAL OVARIAN FLUID BIOMARKERS

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There is increased commercial interest in the production of Lumpfish (*Cyclopterus lumpus*) as a biological control for sea lice infections in Atlantic salmon farming. In the early stages of domestication for new species, ambiguity in the spawning windows can have detrimental effects on egg quality. A lack of clarity in the lumpfish's spawning season can lead to a-synchronous development and long spawning windows. Under these conditions, eggs can be left in the body cavity for long periods of time and result in overripening of eggs and a reduction in egg quality.

The present study aimed to investigate the length of the overripening window, through documenting the effect of post ovulatory aging for lumpfish. It also aimed to characterise the protein composition of the ovarian fluid and identify potential biomarkers for egg quality. Eggs were stripped from wild and captive broodstock and maintained in their ovarian fluid at 4 °C until fertilised. Batches of eggs were fertilised at 4, 12, 24, 48, 72, 96 and 120 hours post stripping. Ovarian fluid was collected at the point of stripping and analysed using TMT isobaric tagging to identify proteins present.

All three measured quality parameters decreased significantly with increased ageing. The quality of captive eggs decreased faster than the wild eggs. Hatching success was significantly reduced (> 50 %) after just 24 hours post ovulatory aging for both groups. The data suggests a short overripening window for this species. A total of 304 proteins were identified as being present in the lumpfish ovarian fluid, with no proteins unique to either wild or captive ovarian fluid. 21 proteins were identified as potential biomarkers associated with egg quality which warrant further investigation, with 1 protein (VPS33B late endosome and lysosome associated) being a significant marker of quality for this species.

Poor egg quality is a major hurdle in successful production of any new marine species and there appeared to be a significant difference in the performance of the wild and captive eggs. The findings of this study suggest that even if egg composition improves within the species, there is a small window that these benefits can be lost because of mismanagement of ovulation and stripping in the species. This study clearly shows that intensive management of broodstock is required in order to identify ovulating individuals for stripping. With a short window (24hr) post ovulation until a significant reduction in quality. Future work needs to clarify the role of many of the proteins identified in this study. It would also aim to build on the data in the current study to develop individual biomarkers for poor quality within the species. Development of such can allow rapid on farm testing of ovarian fluid to improve broodstock management, identify good quality individuals and reduce hatchery effort in poor quality eggs.

SURVEY OF HOBBYIST FISHKEEPERS INDICATES SIGNIFICANT LEVEL OF ORNAMENTAL FISH PRODUCTION UNDOCUMENTED BY CURRENT METHODS

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Ornamental fish are one of the most popular pets in the world, with a massive number of thriving hobbyist groups both online and off. The global trade of Ornamental fish, both marine and freshwater is growing rapidly. However, documentation of the production of fish is limited, due to the complexity, origin, number of species, and limited reporting. Despite this, there is a considerable desire to document this production due to Animal welfare, invasive species, and conservation concerns where a lack of information limits stock management, future research and encourages detrimental practices and isolationism between the industry and the scientific community.

This study aimed to document the number of hobbyists who are involved in the production of ornamental fish. A Google forms survey was distributed using the snowball sampling method within Facebook fishkeeping groups and two online forums. A total of 3304 valid responses were collected, representing a 12.7 % response rate.

This study identified that 66.29 % of hobbyists were actively engaged with breeding fish. With another 23.57 % indicating that they had experienced fish breeding in their aquarium accidentally. There were clear increases in active breeding effort with experience in freshwater hobbyists, increasing from 47.11 % to 97.75 % of those with over 50-year experience in the hobby. There was less effort on the marine side with only 4.38 % overall actively breeding fish, with no difference in experience groups.

There was a high level of dissatisfaction with vet care from hobbyists with 90.99 % indicating that they felt there was not enough access to vet care for hobbyists. Additionally, a number of respondents highlighted a significant issue in, deformity, microbial issues and egg quality issues within the production of their fish. This number also correlated with 68.8 % indicating that greater access to primary information would benefit their production, with a further 79.8 % indicating they would like to see species guidelines produced similar to aquaculture species.

In conclusion, this study has identified that there is a significant level of ornamental fish production by the hobbyists' community. Despite any potential for error in the data of this survey, there is significant evidence of meaningful levels of fish production by hobbyists. This effort towards breeding fish appears to be a natural progression for many within the hobby. There appears to be a higher level of effort engaged in the freshwater side of the hobby, than the marine most likely due to the perceived complexity of marine species reproduction. As such it sets a baseline for future engagement with the hobby and identifies that a large number of those in the hobby are also eager for such engagement.

PHENOTYPIC AND TRANSCRIPTOMIC CHARACTERIZATION OF DERMO TOLERANT EASTERN OYSTER FAMILIES

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Disease tolerance occurs when pathogens and parasites have minimal effect on host fitness. The degree of tolerance is typically quantified by the relationship between pathogen load and host survival or reproduction. Dermo disease, caused by the protozoan parasite *Perkinsus marinus*, negatively impacts survival in both wild and cultured eastern oyster (*Crassostrea virginica*) populations. Several studies have demonstrated breeding oysters that survive in Dermo-endemic environments leads to higher population-level survival in subsequent generations; however, it is unclear whether the increased survival is due to evolved resistance or tolerance to the disease. Here we describe a disease challenge experiment designed to 1) test for evolved tolerance among selectively-bred eastern oyster families and 2) characterize phenotypic and transcriptomic responses to the parasite. Eastern oyster families exhibiting a range of field survival phenotypes were obtained from a well-established breeding program and each family was divided into four groups. The groups were exposed to distinct doses of *P. marinus* (0, 10^6 , 10^7 , and 10^8 parasite spores per gram wet weight) via injection in the adductor muscle and monitored for survival every day for 50 days. Individuals ($n = 3 - 6$) from each family/dose combination were censored seven days post exposure and mantle tissues were collected to confirm parasite load (via qPCR) and generate global gene expression profiles (via RNAseq). mRNA samples from two families at each extreme of the dose-response tolerance spectrum were ultimately selected for short-read, paired-end Illumina sequencing. On average, 93M reads per sample were generated and 93% of paired reads from each sample mapped to the eastern oyster genome. Counts of reads mapped to each feature (transcript) of the nuclear genome were used to quantify gene expression. Principal component analysis of gene expression profiles showed significant clustering of samples by treatment (control vs. injected) along PC1 and by phenotype (tolerant vs. susceptible) along PC4. To assess the effects of family, phenotype, and dose on the transcriptomic response to Dermo, comparisons between control and treated sample expression were made at multiple levels (e.g. each family and dose separately, families grouped by phenotype at each dose, and all doses combined for each family). Depending on the comparison, hundreds to thousands of differentially expressed transcripts were detected. Analysis of overlapping differentially expressed transcripts found a higher percentage of overlapping transcripts among doses within each family than among families within a single dose, suggesting that the host genotype/phenotype may be more important than parasite load in driving the transcriptomic response to Dermo. More detailed analyses of differentially expressed transcripts, functional annotation and enrichment are underway. Taken together, the results of this study should provide valuable insights to this host/parasite interaction and the mechanisms underlying Dermo tolerance in the eastern oyster.

ENVIRONMENTAL IMPACTS OF OYSTERS PRODUCTION FROM AQUACULTURE IN PORTUGAL

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Seafood has a relevant role in meeting food demand while also contributing to climate change mitigation as it typically presents lower greenhouse gases emissions than most land-based foods. Oysters have progressively increased their role in the aquaculture sector in Portugal and still have great potential for growth. In Portugal, it represents about 25% (by volume) of the aquaculture production. In Ria de Aveiro, a lagoon in northern region of Central Portugal, oyster fattening is performed through an extensive system where the feed is exclusively natural.

Life cycle assessment (LCA) is a methodology that can be used to answer many questions that are traditionally not considered in seafood sustainability schemes. Therefore, the aim of this work is to apply life cycle assessment (LCA) to evaluate the environmental impacts associated with the oysters produced from two aquaculture farms in Ria de Aveiro (Table 1) to identify hotspots and support different policy makers involved in the value chains of seafood production to opt for the most environmentally friendly option.

Oyster fattening is performed through an extensive system where the feed is exclusively natural. The functional unit is the production of 1 t of fresh oysters (with shell) ready to the consumer market. System boundaries include the transport of juveniles from France and the fattening stage (Fig. 1). The hatchery and nursing stage are excluded from the system boundaries. The carbon sequestration due to oyster shells growth was also included.

The characterisation factors used in this study are those suggested for conducting a Product Environmental Footprint (PEF). The fattening stage was particularly relevant for freshwater eutrophication and resource use (mineral and metals). The impacts in this stage resulted mainly from the end-of-life at landfill of several materials used in the infrastructure, such as oyster iron racks, steel hooks and rubber anchor bands. Therefore, policies that encourage alternative end-of-life management (e.g. reuse or recycling) could improve the sustainability of oyster production.

TABLE 1. Oysters aquaculture farms characteristics.

	Farms	
	1	2
Species	<i>Crassostrea gigas</i>	
Spats weight (g/spat)	0.35	0.20
Oyster grow-out period (months)	12	14
Mortality rate (%)	67	15
Final oyster weight (g/oyster)	60	85
Average oyster production (t/yr)	40	25

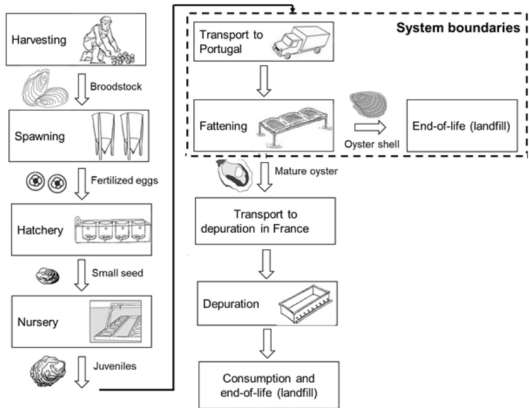


Fig. 1 System boundaries.

KEY THERMOCHEMICAL PROCESSES IN THE AQUACULTURE INDUSTRY AS A TOOL FOR CLIMATE CHANGE MITIGATION

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In response to accelerated climate change, global agreements have been signed to implement and establish the use of renewable energy with low or zero GHG emissions,. As a result, the global bioeconomy is moving towards processes that can offer not only traditional products, but also new advanced renewable fuels, chemicals and other value-added products. Owing to their low cost and easy availability, thermochemical conversion processes, such as pyrolysis and hydrothermal carbonization (HTC), are attractive methods for producing bio-oils and bio/hydro-chars that can be considered as second generation biofuels [1]. The properties and yields of these products and energy requirements depend on operating conditions. These thermochemical processes use a variety of raw waste materials from agriculture, forestry, and non-traditional sources, such as wet animal manure, human waste, sewage sludge, municipal solid waste, as well as aquaculture and algae residues. Fast pyrolysis result in two products, biochar and bio-oil, while HTC is mainly used to obtain hydrochar. However, HTC has the advantage of using raw material with a moisture content greater than 50% [2], thus there is no need for a drying pre-treatment step before the thermochemical process, which opens up the possibility of using aquaculture residues. The fish processing industry generates enormous amounts of waste, approximately 45% of the live weight of fish, which could be used for HTC processing. Several studies have shown that the hydrochar quality of fish waste is largely comparable to conventional hydrochar [3, 4]. Both biofuels, bio-oil and hydrochar have great potential to mitigate GHG emissions and, at the same time, have other economic and environmental benefits. Bio-oil can be used as fuel in existing industrial burners for heating applications and for chemical extraction with high economic value. The advantage of using bio-oil is that it produces significantly lower emissions than fossil fuels. Hydrochar can be used in different applications, such as feedstock in gasification processes [5], as soil amendment [2], as adsorbent, as raw material for carbon production [6], hydrogen storage or electrochemical energy storage [7]. The use or storing of biochar in soils is a way of mitigating climate change through carbon sequestration, while providing energy and increasing crop yields.

This presentation will show that the use of waste generated by the aquaculture industry by either pyrolysis or HTC processes represents a sustainable option for biofuel production. It also provides the possibility of developing new lines of products and extra income from the sale of waste to companies that are dedicated to the production of biofuels.

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SHELLFISH MONITORING ON PRINCE EDWARD ISLAND: BUILDING, MAINTAINING AND ADAPTING PROGRAMS TO CHANGING NEEDS

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The Prince Edward Island Aquaculture Division operates two shellfish monitoring programs: the Mussel Monitoring Program and the Oyster Monitoring Program. The main goal of both programs is to provide the aquaculture industries with technical information to support growers with the management of their farms. Both programs were initiated to gather information to optimize the successful collection of mussel and oyster seed stocks. Since that time, the monitoring programs have evolved to address changing industry needs.

In addition to the Monitoring Programs, the Aquaculture Division added a much-required Productivity Program. Several years ago, there was an unexplained and widespread decline in shellfish productivity. At the time, several theories were considered including unusual weather patterns, changes in water quality and changes in stocking densities within growing areas. Stocking density was ruled out as the cause due to the widespread (Island wide) nature of the decline. Historical weather data from Environment and Climate Change Canada was investigated and anomalies were highlighted as being worthy of additional investigation; however, there was very little data available on water quality. As a result, several multi parameter water quality instruments were acquired and deployed in major shellfish production areas. The goal of the monitoring is to enable the comparison of water quality parameters between growing areas and to also highlight differences between years and determine if these trends align with changes in productivity at the bay level.

A significant amount of data has been collected over the past number of years through these programs. The data has enabled trends to be observed and highlighted; however, the vast amounts of data being collected do not come without challenges and limitations. Some considerations include quality assurance and quality control, data management and access, deployment methods and standardization and of course, ongoing costs to long-term monitoring.

LENNOX ISLAND FIRST NATION – DEVELOPMENT OF A COMMERCIAL SHELLFISH HATCHERY IN PEI

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The rapid growth in oyster culture cannot be sustained by reliance on wild seed, further demonstrated by inconsistent wild spat collection observed over the past two years. There is a recognition by the industry that hatchery produced seed is necessary to ensure the sustainability and security of the industry into the future.

After surveying the oyster growing industry and finding that a need for consistent seed supply was real, Lennox Island, in 2016 began the process of retrofitting an existing shore based building into a shellfish hatchery. The retrofit process involved upgrading the water supply system, adding water supply treatment and UV disinfection, building an algae lab and algae production room, building and equipping a larval room and seed nursery and building FLUPSY (floating upweller systems) for the on-growing of the seed in the ambient environment.

Staff were trained through a variety of means including mentoring at the Eskasoni Shellfish Hatchery and both on-site training and off-site training at facilities such as Valores and the Truro Agricultural College.

The operation now employs five full time staff, three of which are First Nation.

Since its inception in 2016, the hatchery has produced over 15 million seed including 1 million seed that have gone to the Band's new off-bottom lease which offers another opportunity for additional First Nation employment.

Mike Randall, Executive Director of the Lennox Island Development Corporation will discuss how the Band has developed a highly advanced commercial shellfish hatchery in just under four years, starting virtually from scratch.

WATER STRUCTURE IN THE COAST OF BAYS OF NEWFOUNDLAND AND ITS EFFECT ON MODELING WASTE DEPOSITION ASSOCIATED TO AQUACULTURE ACTIVITY

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Measures to ensure sustainable development of marine aquaculture operations include a good management and minimization of the impact of aquaculture activity to the benthic environment. For that, it is necessary to understand deposition around aquaculture sites through monitoring as well as modeling of waste deposition from the operation.

The distribution of such deposition is controlled by the ocean currents at the site. These currents are affected by the water structure with stratification playing a great role on their vertical distribution and their climate variability. Stratification tends to decouple the surface currents from those at depths leading to ocean currents that are variable in the vertical. We present how the structure of the water in the coast of Bays area varies depending on the season and how the structure of the currents also varies accordingly.

When modeling the deposition of waste from aquaculture farms, waste particles are transported by the ocean currents at each depth while they are falling from the cages to the bottom of the ocean. Particle tracking model stipulates the use of high frequency currents (frequency in time and in space) to ensure information is not lost during calculation. We present comparison of outputs of a deposition model driven by ocean currents at different depths with the objective of providing a guideline.

In the Coast of Bays area, understanding the water structure is necessary as the structure informs at what depths the ocean currents need to be considered when modeling waste deposition from aquaculture cages.

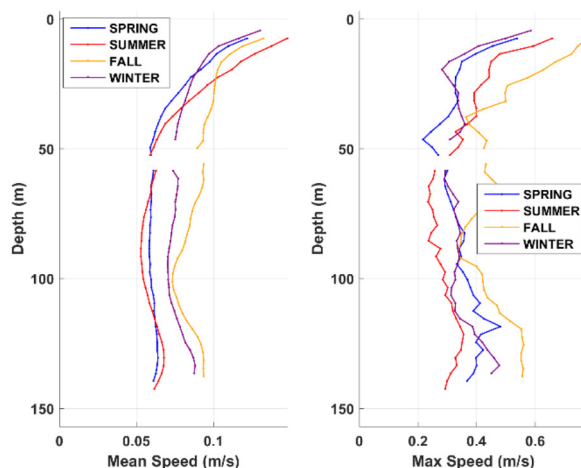


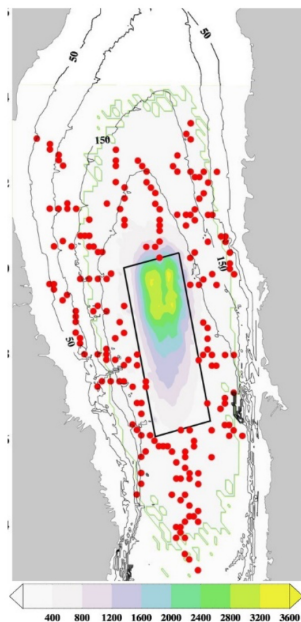
Figure 1: Seasonal mean current speed (left panel) and maximum current speed (right panel) on the western side of Belle Bay in Spring 2015 - Spring 2016.

ASSESSMENT OF BENTHIC IMPACTS OF SALMON FARM ACTIVITIES ON A DEEP SITE IN NEWFOUNDLAND (NL)

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Assessing the changes in benthic organic loading due to particle wastes is challenging in environments such as in NL with deep sites dominated by hard substrates. Here, we analyzed video surveys and generated outputs of a waste deposition model using oceanographic and feeding rate data provided by industry in one NL site. Changes, at different time points during production (over 15 months) in the density of epifauna including habitat building species such as corals and sponges were evaluated. The area was classified in different habitats based on depth, substrate type and taxa clustering using baseline information (Table below- Habitats characterization following multivariate analyses of baseline videos using PERMANOVA+). Still images from drop camera taken during three surveys were also analyzed to identify taxa present at different times (4, 9, 14 months of production). Changes in benthic communities were found to be correlated to distance from cage array and cumulative deposition calculated using DEPOMOD model (V 2.2) with extrapolated 1.5-month ocean current measurement and real time feeding rates. Modelled solid and carbon deposition decreases away from the site as expected (Figure: Map of cumulative carbon deposition (g C/m²) after 15 months of production. Red dots represent locations of video surveys). The analysis shows a decrease in abundances and richness up to a distance of 270 m away from the cages which corresponds to a modelled solid deposition with an upper value of 190 g C/m². In habitats dominated by Ball like sponges or seapen, PERMANOVA revealed an effect of deposition, distance to cage and time of sampling with decreasing abundances 270 m from net pens. No performance-based indicators (Bacterial mats, Polychaetes) were observed during the surveys.



Dominant taxa (Sand- n=722 images)	% stations (depth in m)	Dominant taxa (Rockwall- n=471)	% stations (depth in m)	Dominant taxa (Boulders- n=279)	% stations (depth in m)
Ball-like sponge	19 (100-240)	Ball-like sponge	77 (20-220)	Ball-like sponge	72 (20-240)
Anemones	1 (180-220)	Anemones	13 (40-220)	Anemones	27 (20-260)
Sponge	1 (200-240)	Encrusting sponges	9 (20-220)	Encrusting sponges	1 (40-160)
Seapen	76 (180-240)	Metridium sp	1 (0-20)		

UV LED DISINFECTION: A NOVEL TREATMENT FOR COMMON SALMON AQUACULTURE PATHOGENS

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Both enteric red mouth disease and furunculosis (caused by *Aeromonas salmonicida* and *Yersinia ruckeri* respectively) significantly impact the aquaculture industry. Although vaccination is the most widely accepted mode of protection against these organisms, vaccination efforts are not always reliable. Traditional mercury-based UV disinfection systems have been used to effectively inactivate both species; however, these systems are fragile and energy intensive, making them incompatible for use in challenging environments. Alternatively, UV light emitting diodes (UV LEDs) are durable, mercury-free point-sources of UV radiation that can be designed for output at any wavelength in the UV region. These unique features provide the necessary robustness and design flexibility to implement UV treatment within more challenging environments like open marine waters.

The objective of this project was to explore the effects of UV LED wavelength and particulate loading on the inactivation kinetics of *A. salmonicida* and *Y. ruckeri*. Batch treatments were conducted using a UV LED collimated beam apparatus at 255, 267 and 279 nm for both species suspended in either phosphate buffered solution or sterile municipal wastewater effluent. Species were enumerated using standard plate culturing techniques.

Both pathogens were highly susceptible to UV inactivation, and kinetics were species, wavelength, and matrix dependant. Table 1 highlights differences in key kinetics parameters k_{linear} (inactivation rate constant) and N_{res} (upper limit of disinfection). For *A. salmonicida*, 267 and 280 nm treatments resulted in optimal disinfection in both matrices and was negatively impacted by particulate loading. *Y. ruckeri* responded best to the 267 nm treatment and was slightly less susceptible to UV when compared to *A. salmonicida*. Furthermore, *Y. ruckeri* was not impacted by additional particulate loading.

This study demonstrates that UV LEDs are significantly more effective for inactivation of these pathogens when compared to the traditional UV disinfection wavelength at 255 nm, and highlights the potential for use of this technology in novel disease control in a variety of aquaculture settings.

Table 1. Key Inactivation Parameters for *A. salmonicida* and *Y. ruckeri*

Species	Matrix	UV LED (nm)	N _{res}	k _{linear}
<i>A. salmonicida</i>	Pure Water	255	5.85	0.885
		267	6.72	1.274
		280	6.67	1.130
<i>Y. ruckeri</i>		255	4.39	0.645
		267	6.34	0.817
		280	6.36	0.751
<i>A. salmonicida</i>	Municipal Wastewater	255	4.51	0.712
		267	5.09	1.079
		280	4.88	1.076
<i>Y. ruckeri</i>		255	4.41	0.627
		267	6.24	0.907
		280	6.36	0.824

NON-HALOPHYTE PLANT SPECIES IN BRACKISH-WATER AQUAPONICS

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Brackish-water aquaculture producers often face challenges with nitrate accumulation to the point it becomes toxic to animals. Disposing of high-nitrate water wastes both water and salt, can be toxic to terrestrial crops, and often cannot be discharged into municipal sewer systems. Using aquaponics to reduce waste discharge while producing marketable plants is gaining popularity globally; however, brackish water producers are typically thought to be limited to halophytes, or salt-tolerant plants only. Although halophytes grow well and remove nitrogen in brackish water systems, the plants are often unknown to consumers, or are not palatable. Finding non-halophyte species that are tolerant of salt and better known to consumers may allow producers to utilize aquaponics and reduce water and salt use.

Over 10 different plants have been tested at KSU to determine their suitability for brackish water aquaponics. Each plant is evaluated for salinity tolerance by growing plants in 0, 5, 10, 15, and 20 salinity. Plants that show high survival and growth at elevated salinities are used in further trials that examine plant performance in situations representative of shrimp farming conditions. Variables such as varying nutrient concentrations, plant age, acclimation strategies, decoupled versus coupled aquaponics with shrimp, and the effects of supplemental iron on chloride tolerance have been tested.

Kale has been a standout performer with 100% survival even at 20 salinity. Mustard has also shown high survival while only showing moderate growth reduction at higher salinities. A primary finding is the importance of an acclimation period. This acclimation process involves germinating plants in freshwater, then increasing salinity over a two-week period. Many plants show survival at 5, 10, and even 15 salinity when using this acclimation period. Changes in nutrient uptake and tissue concentrations have been noted, particularly replacement of Ca and K by Na, while Mg seems to be unaffected. Some plants appear to store excess Na in leaf tissue, while other plants (notably kale), appear to have lower amounts of Na in the leaves and can reduce Na uptake at the roots or have a Na transport/evacuation ability. Nitrate uptake rates tended to decrease as salinity increases across all plants, however significant amounts of nitrogen were removed from the water, even at 15 and 20 salinity. Other findings are the increased necessity of dosing certain nutrients at high levels compared to freshwater, particularly iron. This line of research has opened potential opportunities for brackish-water aquaculture producers. Further research will include human sensory profiles of the plants and consumer acceptance studies.

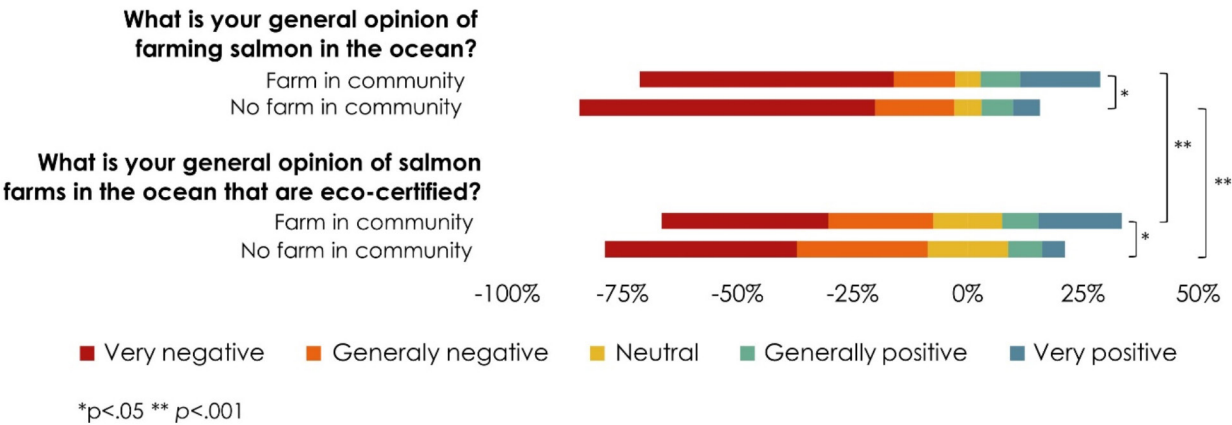
ECO-CERTIFICATION AND SALMON FARMING IMPACTS OF CONCERN IN COMMUNITIES WITH AND WITHOUT FARMS

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The sustainability criteria used to eco-certify aquaculture production emphasizes the prevention of negative local impacts; however, eco-certification addresses social legitimacy at a global scale by providing consumers and retailers with a mechanism to influence aquaculture activities through the creation of a market for sustainable seafood. Further, although eco-certification can improve market access for producers, reputational benefits of eco-certification for producers are unclear. The role of eco-certification in moderating attitudes towards marine salmon aquaculture in British Columbia and Nova Scotia, in communities with and without salmon farms, was investigated using an online survey.

Communities with farms had a more positive opinion of salmon farming and were more concerned about local and provincial economic impacts of salmon farming than communities without farms. Communities without farms were more concerned about local environmental impacts of salmon farming than communities with farms. Communities with farms also reported a higher level of trust in eco-certification than communities without farms. Despite these differences, statistically significant increases in opinion from salmon farming to eco-certified salmon farming suggests that eco-certification could improve the reputation of salmon farming in both communities with and without farms.



MEANS VERSUS MEDIANS: DOES IT MAKE A DIFFERENCE FOR AQUACULTURE REGULATORY COMPLIANCE OF BENTHIC SULFIDES?

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Benthic sulfide concentration is a common proxy for dissolved oxygen. It is often used to classify benthic health as Oxidic, Hypoxic, or Anoxic for regulatory compliance of marine finfish aquaculture. This site classification can dictate if mitigative action is required, such as changes in farm management practices or production levels.

Extensive variability is common with environmental physicochemical metrics such as sulfides. Accurate measurements necessitate replicate sample collection to determine measures of central tendency (*i.e.*, mean or median). Scientific research typically aims to confirm or refute a hypothesis through inferential statistics based on an acceptable level of confidence. However, most regulatory environmental monitoring does not achieve the statistical power necessary for valid inferential comparisons. This is often because, determining if there is an impact is not in question and measures are used to scale an effect. It could be that a small number of samples are collected in an exploratory nature with results used to trigger a larger sampling regimen, if needed. It may also be that the ideal number of replicates is cost prohibitive or impractical to achieve. Sulfide measurements for benthic classification arguably meet all these criteria, being notoriously difficult to measure, highly variable, and expensive. Consequently, descriptive statistical measures such as means are common sulfide reporting metrics applied for regulatory compliance of marine finfish aquaculture in many jurisdictions. However, sulfide data distributions are also often skewed. This begs the question: would sulfide medians be a more accurate measure for regulatory compliance given data distribution potential for skewness? If so, would medians favour more stringent or more lenient regulatory compliance?

In Nova Scotia, Canada, benthic sulfide data has been collected at marine finfish aquaculture sites annually since 2002. In this jurisdiction, means are applied to replicates to determine sulfide values at individual sample locations, and the mean of sample locations are used to classify farm site condition. A total of 193 historical sulfide site assessments were reanalyzed, applying medians to sample values and sample locations. An R dashboard was created to enable ease of plot generation for individual years and sites, and to track changes in classification, upon the application of medians. If medians were applied to only sample values alone, 3% of sites transitioned to a less impacted classification. If medians were applied to both sample values and sample locations, almost 10% of site classifications changed, with most transitioning to a less impacted classification. Implications on environmental monitoring programs of finfish aquaculture are discussed in the context of traditional sulfide monitoring and new more accurate methods being proposed.

RECONCILING CLIMATE CHANGE AND AQUACULTURE: VULNERABILITY ASSESSMENTS AND EVOLVING RESEARCH NEEDS

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Climate change threatens our ability to ensure global food security, eradicate poverty, and achieve sustainable development. Aquaculture is well positioned to help meet the world's future seafood needs, but heavy reliance of most global aquaculture on the ambient environment and ecosystem services suggests inherent vulnerability to climate change effects. While mitigation of climate change is essential, it remains elusive. Adaptation then becomes the only pragmatic option. Reconciling climate change and aquaculture to support adaptation is a huge challenge as decision making typically needs to consider large amounts of information encompassing multiple disciplines. This results in increasing demands for addressing knowledge gaps and accountability for aquaculture research. A climate change vulnerability assessment is one method that triages threats to prioritize adaptation efforts and this approach has recently begun to see application in aquaculture. This presentation explores potential roles for climate change vulnerability assessments and how they could compliment the evolving needs of climate change and aquaculture research, which is crucial to support planned adaptation.

INSTITUTIONAL ANALYSIS OF BRAZILIAN TILAPIA VALUE CHAIN

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Fish farming corresponds to approximately 90% of the Brazilian aquaculture production, which in 2020 reached 643,279 t. Tilapia production alone accounts for approximately 50% of this production and 40% of the commercialized value (IBGE, 2020). Despite being one of the largest aquaculture producers in the world (FAO, 2020), export rates of the country's production are still very small. In the case of tilapia, historically (2013 to 2020), Brazil exported less than 1.1% of its total production (CIAQUI, 2022).

In this context, this research aims to discuss the institutional environment of tilapia production in Brazil. For this purpose, the institutional environment indexes were estimated using fuzzy modeling in three perspectives (Economic, Social and Local). The research data was collected in a region with high capacity to lead value chain globalization in Brazil.

The results indicate the spatial specialization may be a factor leading to a higher offer of credit to production units. However, access to this credit for units located in the production zone may be more difficult due to the environmental and operational laws requirements in that area.

In this context, we argue that the main bottlenecks of the chain (credit and legal compliance) may create a harmful feedback effect, especially for small farmers. Additionally, we point out the importance of reinforcement in the institutional framework for less developed aquaculture chains, as in the case of the Global South. Finally, we highlight the relevance of a research agenda that analyze from different perspectives and moments (e.g.: ex-ante and ex-post) the introduction of Southern firms in globalized chains.

Table 1. Institutional environment fuzzy indexes

Group	Averages of the contextual indexes		
	FEI	FSI	FLI
Ilha Solteira	66.9	54.5	19.2*
Others	62.8	51.6	37.2*

Note: FEI is fuzzy economic index; FSI is fuzzy social index; FLI is fuzzy local index. *Indicates statistical significance at 1% between groups.

ECONOMIC ANALYSIS OF SMALL-SCALE AQUAPONICS PRODUCTION IN NORTHERN BRAZIL

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Aquaponics production integrates in a symbiotic way aquaculture and hydroponics. In this model, it is possible to reach high productivity of fish and plants adding environmental value to the productive system. However, research that analyzes the economic viability of these systems in Brazil is scarce, especially with native fish in the North of the country. In this context, this research aims to analyze the economic viability of lettuce (*Lactuca sativa*) and tambaqui (*Colossoma macropomum*) production in a small-scale aquaponic system.

The research will use a structure installed in the Federal Institute of Tocantins (IFTO) with an estimated production of 360 kilograms of adult tambaqui and 2,250 kilograms of vegetables. The project will estimate the costs of implementing the system, as well as collect data on operating costs and sales of the products on the local market. For the purpose of economic feasibility analysis, the indicators Net Present Value (NPV), Internal Rate of Return (IRR) and payback will be used. Additionally, the project will simulate different scenarios for feasibility analysis, including larger production scales, alternative juvenile tambaqui cultivation, and variations in costs and sales prices.

As results, the project expects to generate technical information on the economic viability of aquaponics, relevant to both the productive sector and academia. Additionally, it is expected that the dissemination of the system itself, as well as the results of the research can also encourage the use of the aquaponics system as an alternative strategy for environmental education and food security in one of the poorest regions of the country.



US COASTAL RESTAURANT SURVEY HIGHLIGHTS MARKETING OPPORTUNITIES FOR LOCAL AQUACULTURE AND SEAFOOD PRODUCERS

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Over four hundred (n=424) restaurant procurers in the coastal United States were surveyed in June 2021. For comparison purposes, 25% of restaurant respondents were from the northeastern, southeastern, northwestern, and southwestern U.S., respectively. The survey sought respondents that work in the restaurant industry and make food purchasing decisions for their restaurants. The questionnaire consisted of general questions about restaurant characteristics, seafood purchasing preferences and willingness to pay, and barriers to buying or buying more local seafood. Survey results show that restaurant procurers look for specific traits in local seafood and aquaculture products. The top three desired traits were: Marine Stewardship Council (MSC), locally certified (through state programs), and Aquaculture Stewardship Council (ASC) certified. Cited barriers to purchasing more local seafood include inconsistent quality, high prices, limited availability, and further processing needs.

Variables significant for restaurants purchasing local seafood (versus those that do not) are fewer seats, higher entrée cost, and more years in business. Variables significant for restaurants’ willingness to purchase local seafood found that restaurant type, serving alcohol, and catering to residents (versus tourists) were associated with restaurants willing to pay more. Summarizing restaurants’ willingness to pay for local seafood shows the most frequent response as a 1% to 24% premium (Figure 1).

This research helps local aquaculture producers target their sales efforts while identifying marketing barriers. It also appears that certifications for seafood are important to restaurant procurers.

Figure 1. Restaurant Willingness to Pay Responses

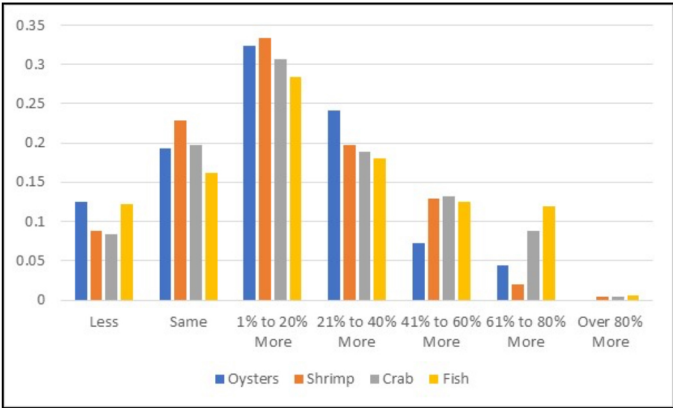


Table 1. Local Seafood Traits Desired by Restaurants

Traits	Rank
MSC Certified	1
Certified {your state}	2
ASC Certified	3
Farm Raised	4
Knowing fisher/producer	5

Table 2. Cited Barriers to Purchasing Local Seafood

Barriers	Frequency
Quality of the product is inconsistent	42.5%
Price point is too high	41.9%
Limited availability/difficulty sourcing	40.7%
Quality does not meet specifications	32.5%
Seafood needs to be processed	26.6%
I do not know where to source	22.8%
Customers do not value local seafood	7.7%

MICROBIAL 'OMICS SHED LIGHT ON COMMUNITY DYNAMICS IN AQUACULTURE SYSTEMS

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Microbial communities influence the growth, development, and welfare of animals farmed in recirculating aquaculture systems (RAS). Despite this well-established fact, the composition of microbial communities within RAS is largely unknown. Establishing accurate, standardized, and user-friendly methods to study microbiota in RAS may lead to better designed RAS, improved management practices, and biological methods to prevent disease outbreaks, thus ensuring food security. We combined various sequencing methods to characterize microbial communities in two geographically separated, commercial-size RAS fish farms. First, we used Illumina short-read amplicon sequencing to compare the performance of three commonly used primer pairs targeting different hypervariable regions of the 16S rRNA gene: v1-3 (27F/534R), v3-4 (341F/805R; MiSeq), and v4 (515F/806R; Earth). Then a subset of samples was subjected to Pac-Bio long-read amplicon (16S-ITS-23S rRNA regions) and Illumina metagenomics sequencing. The short-read amplicon data revealed that samples amplified with the Earth primers yielded higher read quality and diversity but could not assign as many taxa at the genus level as the MiSeq samples. However, both primer pairs could detect similar spatiotemporal patterns between samples types, farms, and biofilm succession and would be adequate for future spatiotemporal studies. The long-read amplicon data could further assign species, allowing for a deeper understanding of functional roles and pathogen detection. Metagenomics results could detect organisms across the tree of life, thus offering a more in-depth view of the community dynamics. In conclusion, different sequencing approaches should not be considered alternatives but complementary strategies. Our approach constitutes a practical and cost-effective strategy for combining multiple sequencing methods to obtain the maximum amount of information on microbial dynamics in aquaculture.

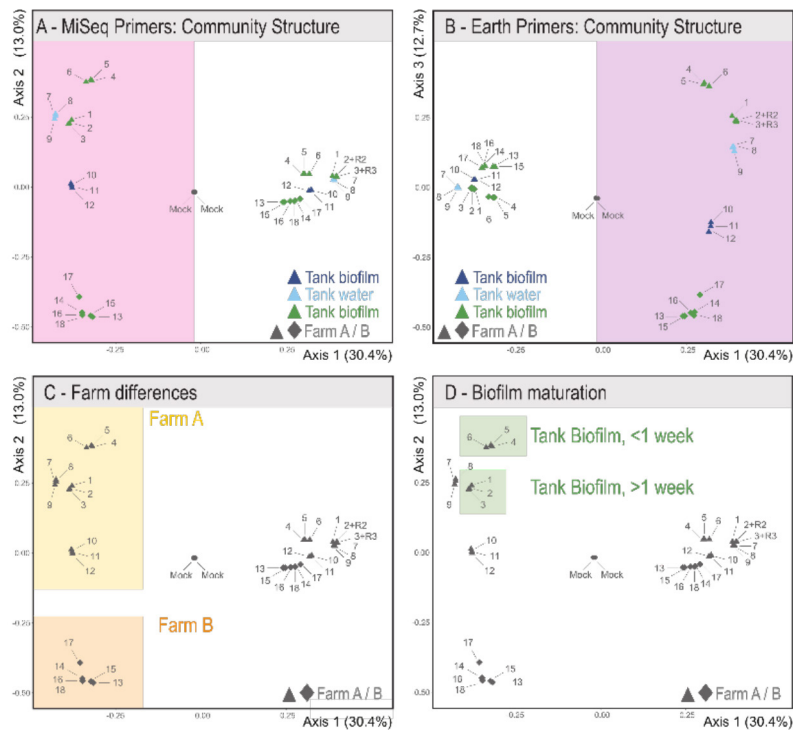


Fig. 1: Biological differences between primers and sample types (A & B), farms (C), and production cycles (D).

AQUACULTURE NUTRIGENOMICS AND IMMUNOGENOMICS RESEARCH: TOWARD DEVELOPMENT OF SUSTAINABLE AND THERAPEUTIC DIETS FOR SALMON

Matthew L. Rise*, Albert Caballero-Solares, Xi Xue, Wenlong Cai, Jennifer R. Hall, Umasuthan Navaneethaiyer, Khalil Eslamloo, Surendra Kumar, Nicole C. Smith, Richard G. Taylor, Rachel Balder, Rune Andreassen, Christopher C. Parrish, and Mark D. Fast

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Immune-relevant components of two Genome Canada supported projects (entitled “Biomarker Platform for Commercial Aquaculture Feed Development”, and “Integrated Pathogen Management of Co-infection in Atlantic Salmon”) will be presented. These projects involved close collaborations between academic (e.g. Memorial, UPEI) and industry (Cargill) researchers using transcriptomics, lipid biochemistry, and complementary techniques to identify and validate molecular biomarkers of Atlantic salmon responses to diet ingredients and immunogenic stimuli [e.g. exposure to pathogens or pathogen-associated molecular patterns (PAMPs)]. Experimental diets were designed and formulated by Cargill, and feeding and immune challenge trials were run in Memorial, UPEI, and Cargill facilities. In addition to *in vivo* analyses, *in vitro* methods were developed to study the impact of feed ingredients on salmon macrophage function. 44K microarrays, mRNA and miRNA sequencing, multiplex PCR, and real-time quantitative PCR (qPCR) were used to identify transcripts that respond to diet (e.g. varying DHA+EPA levels; with and without immunostimulant such as CpG) and/or immune challenges [e.g. co-infection with sea lice and ISAV; or exposure to PAMPs such as poly(I:C) or bacterin]. Selected publications from these projects (see below) will be discussed. Applications of biomarkers (e.g. mRNA, miRNA, lipid/fatty acid) identified in these research projects include the development of novel, sustainable diets that improve Atlantic salmon resistance to infections and co-infections.

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- Cai W, Kumar S, Navaneethaiyer U, Caballero-Solares A, Carvalho LA, Whyte SK, Purcell SI, Gagne N, Hori TS, Allen M, Taylor RG, Balder R, Parrish CC, Rise ML and Fast MD. 2022. Transcriptome analysis of Atlantic salmon (*Salmo salar*) skin in response to sea lice and infectious salmon anemia virus co-infection under different experimental functional diets. *Frontiers in Immunology* 12, 787033.
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- Xue X, Hall JR, Caballero-Solares A, Eslamloo K, Taylor RG, Parrish CC, and Rise ML. 2020. Liver transcriptome profiling reveals that dietary DHA and EPA levels influence suites of genes involved in metabolism, redox homeostasis, and immune function in the Atlantic salmon (*Salmo salar*). *Marine Biotechnology* 22(2), 263-284.
- Xue X, Woldemariam NT, Caballero-Solares A, Umasuthan N, Fast MD, Taylor RG, Rise ML, and Andreassen R. 2019. Dietary immunostimulant CpG modulates microRNA biomarkers associated with immune responses in Atlantic salmon (*Salmo salar*). *Cells* 8(12), 1592.

TRANSCRIPTOMICS OF ATLANTIC SALMON RESPONSES TO BACTERIAL PATHOGENS

Matthew L. Rise*, Khalil Eslamloo, Xi Xue, Albert Caballero-Solares, Umasuthan Navaneethaiyer, Surendra Kumar, Jennifer R. Hall, Mohamed Emam, Eva Jakob, Stanko Skugor, Javier Santander, Richard G. Taylor, and Mark D. Fast

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The bacterial pathogens *Renibacterium salmoninarum*, *Moritella viscosa*, and *Piscirickettsia salmonis* cause infectious diseases [Bacterial Kidney Disease (BKD), Winter Ulcer Disease (WUD), and Salmonid Rickettsial Septicemia (SRS), respectively] that are serious threats to global Atlantic salmon aquaculture. Genomics approaches [i.e. 44K microarrays, RNA sequencing, and real-time quantitative PCR (qPCR)] were used to identify and validate head kidney transcripts that respond to *R. salmoninarum* and *P. salmonis*, and skin transcripts that respond to *M. viscosa*. Thousands of genes were identified as significantly responsive to each pathogen, and qPCR assays were developed for over 100 of these immune-relevant biomarker genes. Gene ontology (GO) term and pathway enrichment analyses of transcriptomic data (i.e. pathogen-responsive genes) provided insights into molecular mechanisms involved in the infections. For example, GO terms enriched in *M. viscosa*-induced skin transcripts included several related to phagocyte function (e.g. macrophage migration, regulation of macrophage activation, phagosome maturation, respiratory burst, Fc-gamma receptor involved in phagocytosis). These transcriptomic studies identified and validated molecular biomarker genes that can be used at the transcript expression level (e.g. qPCR) for aquaculture Research and Development (e.g. diagnostics, vaccine efficacy testing, and studies of the influence of dietary components and feeding regimens on Atlantic salmon responses to these pathogens). In addition, our studies provide comprehensive trait-relevant gene lists (e.g. genes up-regulated and down-regulated by BKD, WUD, or SRS) that can be mined to identify candidate genes for SNP discovery and association analyses, potentially accelerating the development of tools for marker assisted selection of disease resistant Atlantic salmon broodstock.

Eslamloo K, Caballero-Solares A, Inkpen SM, Emam M, Kumar S, Bouniot C, Avendaño-Herrera R, Jakob E, and Rise ML. (2020).

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Xue X, Caballero-Solares A, Hall JR, Umasuthan N, Kumar S, Jakob E, Skugor S, Hawes C, Santander J, Taylor RG, and Rise ML. (2021). Transcriptome profiling of Atlantic salmon (*Salmo salar*) parr with higher and lower pathogen loads following *Piscirickettsia salmonis* infection. *Frontiers in Immunology* 12: 789465.

FRAMING THE NARRATIVE: HOW HEADLINES QUICKLY BECOME FACT

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In today's fast-paced social media world, readers rarely dive deep below a headline. "*Farmed tilapia is less nutritious than a donut*" and "*Study confirms farmed salmon more toxic than wild fish*" are headlines that have been shared around the world, with effect.

This presentation will review two case studies that, while foundationally baseless, have undoubtedly influenced a negative perception. How do we go from a study that confirms all salmon species (farmed and wild) are a fraction of national guidelines for contaminants of concern, to a headline that claims "toxic"?

The presentation will end with best tips on preventing shock headlines, interrupting negative stories, and driving for positive headlines.

EFFECT OF THE TEMPERATURE AND THE SALINITY ON EMBRYONIC DEVELOPMENT OF THE OCTOPUS *Octopus insularis* Leite & Haimovici 2008

Fernando A. Rodríguez-Reyes*, Roberto González-Gómez, Maite Mascaró, César Meiners-Mandujano, Piedad S. Morillo-Velarde

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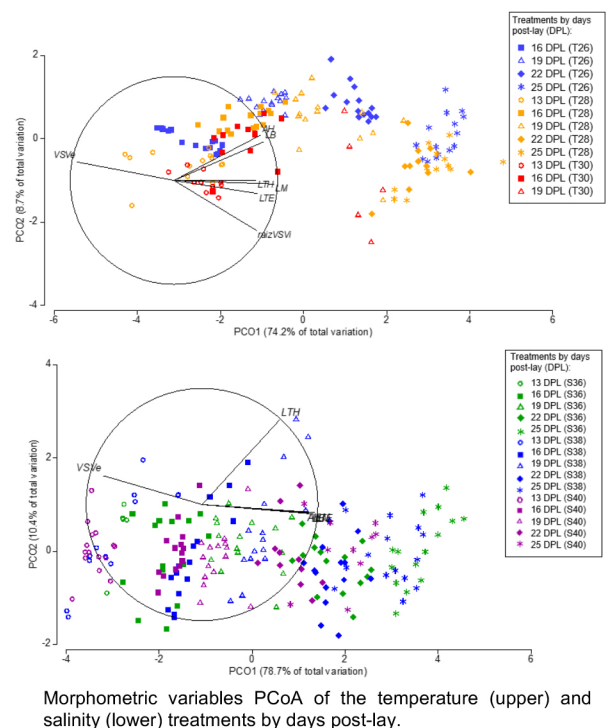
Octopus insularis has recently been recognized as the dominant species in the southwestern Gulf of Mexico fisheries, presenting a new octopus culture opportunity for the region. The development of cephalopods is influenced by environmental variables such as temperature and salinity, however, effects of these factors on the early stages of embryonic development of *O. insularis* are still unknown.

Fifty-four *O. insularis* egg cords were artificially incubated at temperatures of 26 (T26), 28 (T28) y 30°C (T30) and salinities of 36 (S36), 38 (S38) y 40 (S40). In each treatment the Naef development stage, embryonic morfometry and embryonic development time were determined. The variation of these factors was evaluated by a Principle Coordinate Analysis (PCoA) and a PERMANOVA test using the PRIMER-e program.

The temperature and embrionic development length varied inversely proportional. T26 was six days longer than T28, this last one being the shortest time (31 days). S36 and S38 were 33 days development time long. While T30 and S40 generated a deleterious effect. At T30, S38 and S40 treatments, malformations were observed in the embryos.

The first principle coordinate PCoA about morphometry in eggs exposed to differents temperatures explained 74.2% of the observed variability, with an important contribution of egg width, mantle length, arm length and yolk sac volumen in the form as samples were ordered along the horizontal axis. In thermal treatments a significant effect ($p < 0.05$) on the speed of morphometric changes was found. T30 treatment delayed the increase in all the measured variables, especially in terms of the arm length and the egg width.

The first principle coordinate PCoA about morphometry in eggs exposed to differents salinities explained 78.67% of the variability, meanly by the egg width, followed by the embryo total length, mantle length, arm length and yolk sac volumen. The morphometry changed over time, but it did so in the same way for all salinity treatments. In general, morphology of S40 embryos differed from the rest troughout the experiment ($p < 0.05$).



The temperature effect on embryonic development time has been previously studied in other species (*O. vulgaris*, *O. maya*, *O. mimus*, *O. tehueltchus*, etc.). In addition, temperatures outside the optimal range of each species have been related to low fecundities, hatching rates and development interruption. High salinities confirm the interruption of the embryonic development and generation of malformations, but no effect on the development time. While similar effects have been reported in cuttlefish and squids, this is the first time it has been reported for octopus.

BRITISH COLUMBIA FISHERIES AND AQUACULTURE OCEAN ACIDIFICATION AND HYPOXIA ACTION PLAN

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In 2019, the Province of British Columbia (BC) completed a Preliminary Strategic Climate Risk Assessment as part of BC's Climate Preparedness and Adaptation Strategy (CPAS) to better understand climate-related risks. The risk assessment identified ocean acidification as a significant climate risk for BC. However, due to a lack of information the assessment gave a lower consequence rating due to a lack of regionally relevant information.

Notwithstanding the ecological and cultural value of BC's coastal marine resources, the risk from ocean acidification and hypoxia to BC's seafood sector is significant. BC's ocean economy contributes \$21.5 billion to the provincial GDP or 8%, directly employing 131,000 people. Of this, the seafood sector contributes \$1.5 billion or 0.6% to the provincial GDP. BC's seafood sector consists of both wild capture & aquaculture and provides thousands of jobs to British Columbians, employing 9,722 people in 2020. Seafood is also an integral cultural resource for many First Nations. Fishing and aquaculture are key factors contributing to the well-being of many First Nations communities across the province. Many First Nations in BC have identified fisheries and aquaculture as an important part of reconciliation, economic self determination, and food security and sovereignty.

To better understand the state of knowledge related to ocean acidification and hypoxia in BC's coastal waters, the Province of BC initiated the development of an Ocean Acidification and Hypoxia Action Plan to inform recommendations for mitigation and adaptation strategies to support the long-term sustainability of the fisheries and aquaculture sectors. Development of the plan was led by an advisory committee consisting of science experts, industry, First Nations, Provincial, Local and Federal Government representatives. The advisory committee convened 4 virtual workshops between November 2021 and March 2022 to assess: 1. state of the science; 2. seafood harvester and producer perspectives; 3. coastal community perspectives; and 4. policy and governance considerations. The workshops attracted 242 participants (172 unique) from 88 unique organizations.

This presentation will provide a summary of the key findings from the workshop discussions along with the goals, objectives, and actions recommended.

CURRENT STATUS OF THE BRITISH COLUMBIA AQUACULTURE SECTOR: WHERE DO WE GO FROM HERE?

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British Columbia's coastline, which extends over 25,000 km, and unique geography provide for a significant seafood industry. In 2020, BC harvested 272,000 tonnes of seafood, amounting to a wholesale value of \$1.62 billion. The aquaculture and fisheries sectors provide thousands of jobs to British Columbians, employing 9,722 people in 2020. The aquaculture sector grows finfish, shellfish and marine plants for harvest. In 2020, aquaculture had a wholesale value of \$822 million, 51% of the total seafood value. The most significant aquaculture species in BC is Atlantic salmon, with a wholesale value of \$710 million in 2020, 86% of the aquaculture value and 44% of the total seafood value. Farmed Atlantic salmon is also BC's most valuable Agrifood export (\$566 million in 2020). Shellfish aquaculture is another significant part of BC's aquaculture sector with a wholesale value of almost \$33 million in 2020.

First Nations have been engaged in shellfish aquaculture in BC since time immemorial and more recently supporting growing involvement in contemporary aquaculture. 78% of B.C. salmon production is carried out under partnerships with First Nations who hold approximately 20% of the jobs in B.C. associated with salmon farming. First Nations are also active participants in contemporary shellfish aquaculture and have a keen interest in the emerging seaweed farming sector. Many First Nations in BC have identified aquaculture as an important part of reconciliation, economic self determination, and food security and sovereignty.

While aquaculture has grown significantly since the 1980s, production in both finfish and shellfish has plateaued in recent years. The reasons for this are multifactorial, ranging from public perception issues, regulatory uncertainty leading to waning investment and, more recently, existential threats from climate change. This presentation will review the current state of the sector, factors that have influenced sector growth, and options and considerations for moving the industry forward.

eDNA METABARCODING OF STOMACHS FROM TWO CANADIAN CLEANER FISHES: WILD CUNNER WRASSES AND LUMPFISH FROM ATLANTIC SALMON SEACAGES

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Lumpfish (*Cyclopterus lumpus*) and cunner wrasse (*Tautoglabrus adspersus*) are two species of cleaner fish used in sea cages by Canadian Atlantic salmon aquaculture for the biological control of the ectoparasitic salmon louse (*Lepeophtheirus salmonis*). Lumpfish are the predominant species used in Canadian salmon aquaculture, whereas cunner are the only member of the cleaner wrasse family that inhabit Atlantic waters. To better characterize the diets of wild cunner populations and cultured lumpfish from industrial-scale sea cages (lumpfish) we combined traditional morphological diet assessments with eDNA metabarcoding. The primary goal of this research was to provide insight into how natural diet variation and prey availability influence the propensity of cleaner fish to feed on salmon lice in an industrial aquaculture setting.

We first performed traditional morphological stomach analysis following eDNA protocols and then isolated DNA from 122 lumpfish stomachs from Canadian Atlantic salmon cages (Cooke Aquaculture Inc.) in southern Newfoundland¹ and 110 cunner gastrointestinal tracts from four regions along coastal Atlantic Canada². Diet assessments and taxonomic identifications were completed using morphological techniques (biomass; prey abundance), and further characterized by molecular metabarcoding for targeted identification at higher taxonomic resolutions (read abundance: READS; sequence diversity: ESVs).

Wild cunners, fed primarily on benthic prey including invasive ascidians (southern regions), barnacles, bivalves, and malacostracans (Figure 1). Lumpfish diets were largely characterized by pelagic wild crustaceans, and industrial feed components (fishmeal and poultry), indicating they fed predominately in the water column (Figure 2). Most notably, metabarcoding was the only technique able to identify the salmon louse in ethanol-preserved lumpfish or cunner stomachs, and document when cleaning activity had occurred. For both cleaners, traditional diet assessments produced low resolution, semi-quantitative prey abundance at the order or family level, whereas metabarcoding could identify prey that were present at the species level. This analysis indicated a clear difference in diet between the two species which can be used to make decisions on their use within Canadian sea cages.

Figure 1²

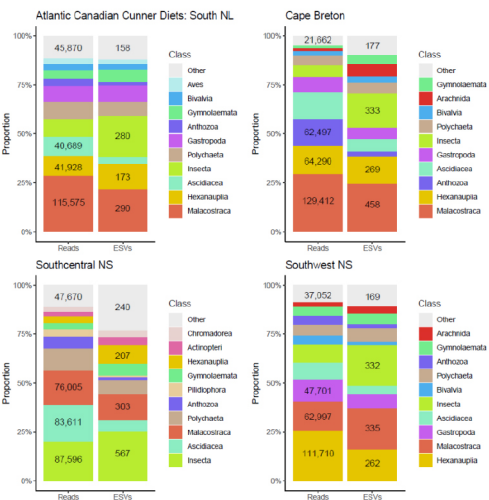
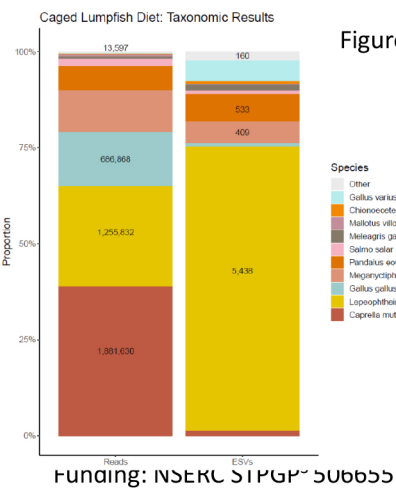


Figure 2¹



Funding: INSEK STPGP SUB655

GENOMIC SELECTION FOR MUSCLE GROWTH AND FILLET QUALITY TRAITS IN RAINBOW TROUT

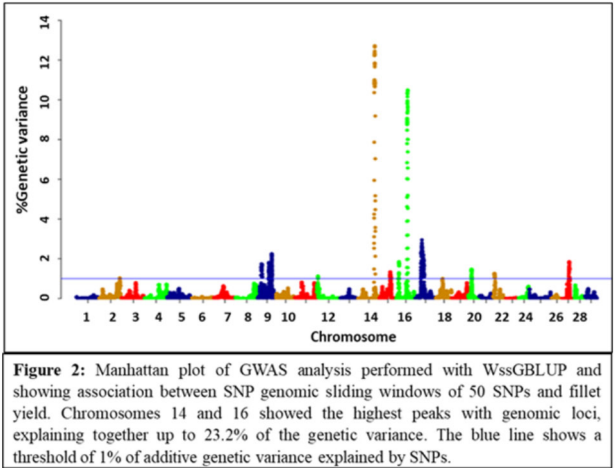
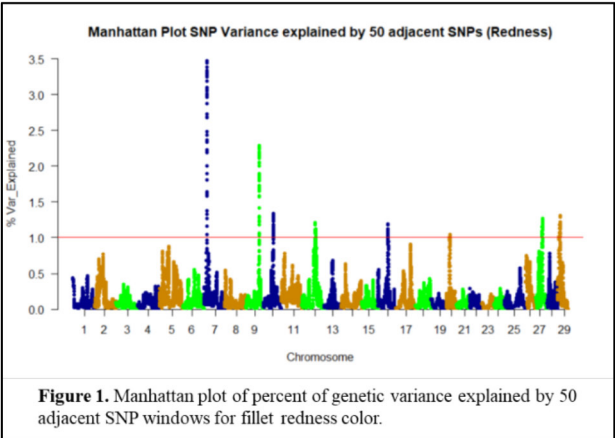
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Rainbow trout is the most cultivated cold-water fish in the U.S., and muscle fillet is its most valuable product. Increasing fillet yield and reducing fillet downgrading will improve production efficiency, which can have substantial economic benefits.

Genomic selection has been widely used to increase the efficiency of genetic improvement programs in livestock but has yet to be widely adopted in aquaculture. Classical breeding approaches can improve fillet yield but are time-consuming because of long generation intervals. A more efficient approach is to use genomic selection, where thousands of SNP are used as additional information to increase the accuracy of breeding values and reduce generation interval, consequently increasing genetic gain.

The industry’s immense interest is to implement genomic selection in their breeding populations. Using a “functional” 50K SNP-chip developed in our lab, we identified QTL for fillet yield, firmness, color, protein, and fat contents. We demonstrated the utility of the genomic selection in rainbow trout. Our studies show that the genomic predictions can improve the estimated breeding values by 33.3% for the fillet yield and 53.1% for fillet firmness. The genomic predictions outperformed the traditional breeding by 35% for fillet yield and 42% for fillet firmness. Additionally, reducing SNP panel densities indicated that using ~1000 SNPs in genomic predictions still provides predictive abilities higher than traditional breeding. These results suggest that genomic evaluation is a feasible strategy to identify and select fish with superior genetic merit within rainbow trout families, even with low-density SNP panels.



AQUACULTURE DIVERSIFICATION: OPTIMIZATION OF GROWTH PERFORMANCE AND TECHNICO-ECONOMIC ASSESSMENTS FOR OYSTER AQUACULTURE DEVELOPMENT IN MAGDALEN ISLANDS, QUEBEC

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Shellfish aquaculture diversification is essential to ensure rentability and sustainability of shellfish farming activities worldwide, and more specifically for small-scale industries developed in local coastal areas such as Magdalen Islands (QC). Magdalen Islands have a subarctic oceanic climate and are composed of multiple shallow coastal areas and lagoons where the seawater temperature is expected to increase with global warming. As such, local farmers, farmers that were predominantly growing mussels, have recently taken up oyster farming of the American oyster (*Crassostrea virginica*) due to its high potential market value and positive outlook. However, one of the short-term challenges faced by the new oyster farmers is the adaptation of methods specific to their environment. This includes which type of structure, oyster density and antifouling techniques to use, but also how to evaluate growth performance and its economic value as a new activity. This study had 3 main objectives: the first objective aimed to identify the best method for oyster growth and fouling management. To answer this objective, oysters were placed for 2 years (2018-2019) in 2 structure types: a combination of pearl nets and lanterns, and Australian baskets. Structures were then deployed in Merinov's site (lagoon) and 3 industrial partners' sites (2 in lagoon, one in a more exposed area). During this period, various antifouling methods were used: air exposition and seawater power washing were applied at two frequencies (monthly and bimonthly), while oyster growth, mortality, and biofouling coverage were followed. The second objective aimed to identify what combination of oyster density, seeding size, and structure type was the most performant. Here, growth and mortality were followed over 2 years on individuals placed in 2 structure types (Australian baskets and pearl net/lanterns), at 3 initial densities and at 2 seeding sizes on Merinov's site (lagoon). Interestingly, the bi-monthly air exposition technique was the most efficient on biofouling but also had the largest impact on growth. Finally, the last objective was to develop a techno-economic model for oyster farming production and its application within the Magdalen Islands context. The advantages and drawbacks of each growing and management method as well as economic factors will be further discussed in the presentation.

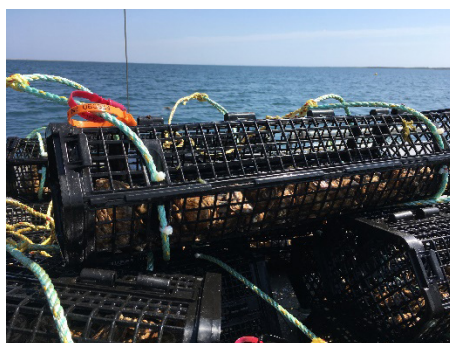


Figure 1: Oysters in Australian baskets prior deployment.

A BIOTECHNOLOGY PLATFORM FOR AQUACULTURE

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As aquaculture production must double in the next 30 years to meet the increased demand in fish consumption, our industry needs to offer innovative technologies to unlock key issues while being sustainable, environmentally conscious and appealing to both the feed manufacturer and the farmer. The aquaculture industry has higher demands for its product than ever, while production inefficiencies due to mortalities and constraints on existing infrastructure potentially constricts future growth. Creating functional products for aquafeed that support healthy fish while at the same time offering the highest quality products consumers expect is one of the challenges that KnipBio is tackling.

KnipBio's single cell protein (SCP) technology platform enables a versatile, functional protein effective in practical formulations for juvenile stages of growth and RAS-based settings. Application examples include off-flavor remediation, juvenile health, and pigmentation. The lack of antinutrient factors, as well as the presence of antioxidant carotenoids and prebiotics with immune-enhancing properties have the potential to lead to healthier fish and shrimp. By targeting specific issues encountered in established production systems, KnipBio Meal (KBM) products significantly decrease production costs while increasing the survival and quality of aquatic animals on the farm, which make operations more profitable.

KnipBio will present an update on its technical progress including recent strides in its R&D platform and animal trials, its recent CFIA approval for salmon feeds and other product characteristics.

ECOLOGICAL AND GENETIC IMPACTS OF FARMED ATLANTIC SALMON (*Salmo salar*) INTROGRESSION INTO WILD POPULATIONS IN SOUTHERN NEWFOUNDLAND

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Atlantic Salmon (*Salmo salar*) are one of the most heavily cultured fish in North America, as this practice presents an alternative to supply the growing global demand for marine food resources. However, this process has resulted in highly domesticated salmon, with notable genetic and phenotypic differences relative to their wild counterparts. Due to this divergence, interbreeding of farmed Atlantic salmon with wild salmon may result in the expression of maladaptive traits for life in the wild, affecting local adaptation. Therefore, it is essential to understand how genetic interactions with farm escapees affect wild populations at local scales. Yet, the few studies that have quantified survival and phenotypic differences among wild, farmed, and hybrid individuals in nature have been done in Europe or at limited scales. In this study, survival, growth, and the genetic impacts of farmed Atlantic Salmon introgression on a wild population in a natural environment in southern Newfoundland were measured.

A release experiment was conducted in three sites in the Garnish River system, located near an area of intensive aquaculture in the Burin Peninsula. Four cross types were produced in 2017 and released in July of 2018 (pure wild, pure farmed, farm-mother hybrids [F♀hyb], and wild-mother hybrids [W♀hyb]). Recapture occurred in the fall of 2018, 2019, and 2020, where samples were weighed, fork length measured, and caudal fin tissue collected for later use in parentage analysis. A panel of 25 microsatellite loci with a total of 277 alleles was used to assign individuals back to their cross type and family. With this, the proportion of wild, farmed, and hybrid individuals at recapture over two years was quantified and their differences in survival and growth were analyzed. Furthermore, the differences in survival over time among families of each cross type were studied. This builds on the previous survival and growth analyses done the first summer after release, adding two further years of data, as well as the family composition during this time.

Survival, measured as odds of recapture, differed significantly among cross types in certain sites, with farmed and W♀hyb having consistently higher odds both years. However, the sites where this difference was evident differed over time. Size at recapture differed significantly among cross types in sites 1 and 3 one year after release (mean length: $p < 0.001$; mean weight: $p < 0.001$). In both sites, farmed and W♀hyb were significantly larger than wild and F♀hyb, with no notable differences between one another. The family specific analyses demonstrated that the composition of families remained relatively consistent from 0+ to 1+ and then to 2+. These results indicate that direct genetic interactions between farmed and wild Atlantic Salmon can impact the survival and fitness of wild populations, contributing to their decline over the last decades.

TWO PENTRAXIN-LIKE PROTEINS IDENTIFIED FROM *Amphiprion clarkii*: MOLECULAR CHARACTERIZATION AND TRANSCRIPTIONAL EXPRESSION ANALYSIS

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Pentraxins (PTXs) are a superfamily of evolutionary conserved proteins in the humoral arm of the innate immunity. PTXs can be divided into two subclasses, the classical short-chain PTXs including C-reactive protein (CRP) and serum amyloid P (SAP), and the long-chain PTXs, which have an unrelated amino-terminal region coupled to a carboxyterminal PTX-like domain. Fish CRP/SAP are widely expressed in the tissues and cells of the innate immune system. Upon infection via a pathogen, their expressions increase dramatically and immediately.

CRP and SAP were identified from *Amphiprion clarkii* database and referred as *AcCRP* and *AcSAP*, respectively. *AcCRP* encoded polypeptide with 220 amino acids with 25 kDa of molecular weight. *AcSAP* encoded polypeptide with 223 amino acids with 25.5 kDa of molecular weight. A characteristic 16 residue signal peptides were identified at the N- terminal of the sequences confirming their secretory properties. The phylogenetic analysis and the pairwise sequence alignment showed their close relatedness with *Amphiprion ocellaris* with 95.8 % of sequence identity. Moreover, in the phylogenetic tree, *AcCRP* and *AcSAP* clustered together with other fish short pentraxins, showing their closest relatedness together with teleost fish. The tissue distribution analysis showed their highest expression in Liver. By the induction of poly (I:C), LPS and *Vibrio harveyi*, *AcCRP* and *AcSAP* showed their significant regulations in a time course manner revealing their acute phase immune responses. Together with that this study provides an experimental insight in to the molecular and transcriptional characteristics of two major evolutionary conserved acute- phase proteins involving in the innate immunity while allowing us to propose possible functions of CRP and SAP in the innate immune system of *Amphiprion clarkii*.

USING EDUCATION NETWORKS TO BUILD PUBLIC TRUST AND ENHANCE AQUACULTURE ACCEPTANCE

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Aquaculture industry expansion, particularly in countries like the United States and Canada, is at the leading edge of Blue Economy development. With growing potential to improve food security, restore habitat and at-risk species, and enhance economic activities, the aquaculture sector sits at the intersection of science, industry, and policy. Through combining mariculture techniques with Blue Economy goals, we are presented with a unique opportunity for transdisciplinary learning and development across sectors. However, domestic aquaculture expansion also generates a wide variety of potential barriers, often fueled by negative public perceptions. Negative perceptions and misperceptions can impact public acceptance of local or regional aquaculture efforts and jeopardize public trust for aquaculture products and their associated technologies. Aquaculture education strategies that help consumers navigate, and take ownership of their decisions can reduce public misperceptions of aquaculture and paint a more holistic picture of what the sector looks like in their region.

Here, we discuss NOAA's approach to aquaculture outreach and education. First, we outline existing NOAA partnerships that recognize the value of education in connecting the public to the aquaculture stakeholders. Next, we look ahead to NOAA's effort to build a community of practice for aquaculture education. We discuss collaborative actions taking place to disseminate accurate information about marine aquaculture more consistently across diverse stakeholder groups. We hear from our partners in this effort and also open the floor to facilitate a discussion of how to embrace regional variation in community needs to expand domestic aquaculture in North America. Given the regional specificity of public's perception on aquaculture, if North American domestic aquaculture expansion is to truly succeed then regionally and locally relevant education strategies must be collaboratively developed by educators and aquaculture experts. This will ensure that such strategies result in programming that is effective, credible, and widely accepted. No one sector can accomplish this alone and the time is now to discuss how we can help each other build coalitions for aquaculture literacy.

THE ENVIRONMENTAL TOLERANCES OF ATLANTIC SALMON (*Salmo salar*): WHAT MEASURES ARE THE MOST REVELANT?

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Given the predicted increase in average ocean temperatures, and in the frequency and severity of heat waves, cold events and hypoxic episodes, due to climate change there is significant focus on determining the temperature and oxygen (hypoxia) limits of many fish species. Methods used to determine acute temperature (CT_{Max} and CT_{Min}) and hypoxia tolerance vary in the literature, and there is concern about how lab-based measures relate to those in free-swimming fish. Further, longer term (incremental temperature, IT_{Max}) tests have now been conducted on a number of species as they better reflect the temporal nature of seasonal changes in water temperature in temperate regions (including at aquaculture cage-sites), but there is limited data on how CT_{Max} and IT_{Max} values differ.

To investigate how different methods of determining acute upper temperature and hypoxia tolerance affect the values obtained, three widely used protocols that utilize heart rate-temperature relationships and other indices to estimate a species' tolerances were compared using adult (~ 800 g) Atlantic salmon from the same population. Salmon were: **i)** implanted with data loggers, given a month to recover, and tested (2°C h^{-1} ; an ecologically-relevant rate of heating) while free-swimming in a tank with conspecifics; **ii)** fitted with Doppler flow probes and exposed to a 2°C h^{-1} temperature increase in a respirometry chamber until CT_{Max} ; and **iii)** anaesthetized, implanted with a data logger, and pharmacologically stimulated prior to a temperature increase of $10^{\circ}\text{C h}^{-1}$ (i.e., using the 'ramp protocol'). Fish in respirometers and those free-swimming in the tank were also exposed to a stepwise decrease in water oxygen level (from 100 to 30% air. sat.) and the oxygen level at which bradycardia occurred was determined.

Resting heart rate (f_H) was significantly lower for the free-swimming fish as compared to those in respirometers (~49 vs. 69 min^{-1}), and this was reflected in their scope for f_H (~70 vs. 105 min^{-1}) and their CT_{Max} values (27.7 vs. 25.9°C). Further, the Arrhenius breakpoint temperatures and the temperature at maximum f_H for the free-swimming fish were much greater than for the other two groups, respectively (~18.4, 18.1 and 14.6°C; 26.5, 23.2 and 20°C). Finally, the oxygen level at which bradycardia occurred was significantly higher in free-swimming salmon as compared to those in respirometers (62 vs. 52% air sat.). Interestingly, the IT_{Max} for salmon (determined at $0.2^{\circ}\text{C day}^{-1}$) was 25.2°C, ~3-4°C lower than their CT_{Max} (28.5°C; both measured in free-swimming fish), and this difference was much greater than previously determined in cod (21.8 vs. 22.5°C).

Overall, these studies question the relevance of CT_{Max} measurements (particularly those not performed on free-swimming fish or at ecologically relevant rates of heating) with regard to making predictions of how climate-related changes in water temperature and oxygen levels will impact salmon (fish) physiology and survival in culture.

ATLANTIC SALMON (*Salmo salar*) CAGE-SITE DISTRIBUTION, BEHAVIOUR AND PHYSIOLOGY MEASURED DURING A NEWFOUNDLAND HEAT WAVE USING DATA LOGGERS

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Increasing water temperatures and reduced oxygen levels due to climate change are a challenge for fish reared in cage-sites in coastal regions. In addition, the frequency and severity of storms, heat waves and 'cold events' can increase temperature variability, and pose significant additional challenges. The Atlantic salmon industry must adapt to these challenges to grow sustainability, and thus, a better understanding of how sea-cage conditions affect fish physiology and behaviour under such conditions is needed.

Salmon (~2.5 kg) in a sea-cage on the south coast of Newfoundland were fitted with Star-Oddi milli-HRT ACT and milli-TD data loggers in the summer of 2019. These tags continuously recorded the fish's 3D acceleration (i.e., activity/behaviour), electrocardiograms (used to determine heart rate and heart rate variability), depth and temperature from early July to mid-October.

Surface water temperatures ranged from ~10-12 to 18-19.5°C over the summer/ early fall, and then abruptly fell to 8-10°C after hurricane Dorian passed over the region. The fish typically selected temperatures of 14-18°C during the warmest weeks of the summer (the maximum temperatures experienced by the fish $19.3 \pm 0.05^\circ\text{C}$), but used the full range of depths available during this period. Heart rate and depth were greater during the day, and activity and the proportion of time spent non-steady swimming increased when feeding was stopped at high temperatures.

These data show that data loggers can be used effectively to map environmental conditions throughout salmon sea-cages, and to determine how conditions and management decisions impact the behaviour and physiology of fish. Thus, they can provide important insights into fish health and welfare, and in combination with lab-based studies, valuable information about what conditions are a concern with regard to the increased risk of mortalities / mortality events.

Further advancements in this technology, including the development of tags with extended battery life and acoustic transmission capabilities, could make it suitable for integration into industry cage-site monitoring programs.

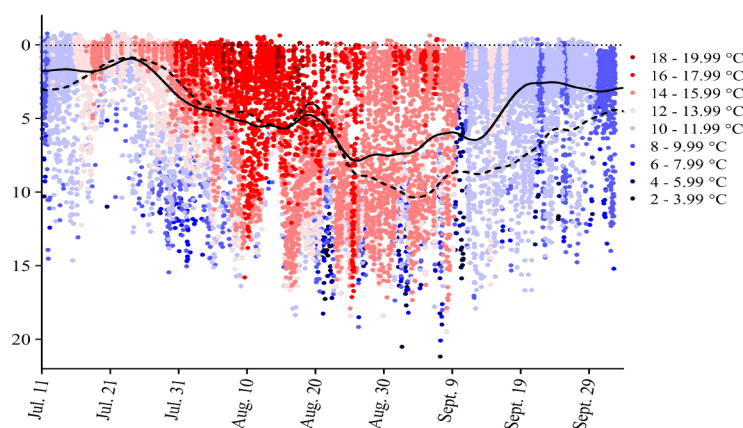


Figure 1. Cage-site temperature data at various depths over the duration of the study. The Lowness curves show the mean depth that the fish (n=7) occupied during the day (broken line) and night (solid line). Hurricane Dorian Sept. 9th.

IRON ACQUISITION MECHANISMS OF *Moritella viscosa*, IMPLICATIONS FOR PATHOGENESIS AND FISH VACCINOLOGY

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Iron is an essential element for life and plays a pivotal role in bacterial pathogens. It is required for the respiratory chain and as a co-factor. However, iron overload is toxic, causing a dysregulated oxidative response, thus iron uptake is tightly regulated. Bacterial pathogens can sequester iron from the host protein, and outside of the host, like in marine environments, bacteria have to scavenge for this limited and essential nutrient. *Moritella viscosa* is a marine psychrophilic Gram-negative pathogen that causes winter ulcerative disease in several fish species. Here, we describe for the first time the *M. viscosa* iron uptake mechanism and the regulatory network in response to iron limited conditions.

Outer membrane protein profiles of *M. viscosa* grown under iron-rich and iron-limited conditions revealed specific genes related to iron-limitation response. Transcriptome analyses showed that 77 genes were dysregulated. Heme, ferrous iron, maganase, tugsten, hydroxamate siderophores transporters, and genes related to oxidative stress were up-regulated. These findings provide a comprehensive analysis of the transcriptional response of *M. viscosa*, adding relevant understanding of the gene regulatory network of this pathogen and revealing target genes that can aid to the development of effective therapeutic strategies against this important fish pathogen.

LUMPFISH (*Cyclopterus lumpus*) IMMUNITY TO *Vibrio anguillarum* VACCINES AND TRANSCRIPTOMIC PROFILE OF SYSTEMIC IMMUNE RESPONSE

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Lumpfish (*Cyclopterus lumpus*) has become the predominant cleaner fish species used in North American salmon aquaculture for sea lice (e.g., *Lepeophtheirus salmonis*) biocontrol. Lumpfish utilization has contributed significantly towards eliminating the utilization of chemotherapeutants by effectively controlling the abundance of this damaging pest of Atlantic salmon (*Salmo salar*) aquaculture. *Vibrio anguillarum* is a frequent pathogen of lumpfish in Atlantic Canada. Here, several vaccine trials against *V. anguillarum* were conducted. We determined that the *V. anguillarum* growth conditions are essential for expressing protective antigens in the vaccine formulation. Generic commercial vaccines provide between 2-55% protection against *V. anguillarum*, not adequately protecting the lumpfish. Generic or autogenous vaccines delivered by mucosal routes (e.g., dip or bath) stimulate the naïve fish. However, only systemic immunization regimes (e.g., intraperitoneal injection) induced significant protective immunity against the lethal *V. anguillarum* systemic or mucosal challenge. Mucosal immunization conferred an evident immune stimulation but not immune protection. Transcriptome analysis of immunized fish revealed an evident difference in the transcriptome profile of the spleen and the head kidney. A modest number of DEG was detected in the head kidney, spleen from single immunized animals, and head kidney from boosted fish. A more significant number of DEGs was detected in the spleen of boosted animals. Common DEGs were observed between the boosted organs but not in single immunized organs. Gene ontology analysis in the boosted spleen revealed enrichment for humoral immune response, plasma lipoprotein particle, vitamin transport, metallopeptidase activity, among other immune-related gene groups. Hepcidin and c6 complement encoding genes were consistently upregulated in all organs studied and might be valuable biomarkers of effective vaccination.

Piscirickettsia salmonis* INFECTION IN *Cyclopterus lumpus

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Lumpfish (*Cyclopterus lumpus*) is the most utilized cleaner fish in the North Atlantic salmon (*Salmo salar*) aquaculture industry. Lumpfish preys on the ectoparasite sea lice (e.g., *Lepeophtheirus salmonis*), one of the major threats to salmon farming worldwide. Lumpfish perform well in cold environments, and it has been commercially utilized as sea lice biocontrol since 2013 in Norway and since 2017 in Atlantic Canada. A healthy population of lumpfish could be used during two production cycles of Atlantic salmon. Bacterial infectious diseases are the most frequent health issues of lumpfish that impact its performance and extended utilization. We have developed several bacterial infectious disease models in lumpfish for vaccines and bacterial pathogenesis studies. Here, lumpfish immunity and infection to *Piscirickettsia salmonis* are examined and discussed. *P. salmonis* is a facultative intracellular Gram-negative bacterium, which is the etiological agent of salmonid rickettsial septicemia (SRS), a disease that causes significant losses in the salmon farming industry and infect lumpfish. Nevertheless, this pathogen is not limited to only salmonid species, since the first report of SRS in lumpfish, no more information is available about the pathogenesis of *P. salmonis* in lumpfish. To unveil *P. salmonis* infection kinetics and disease on lumpfish, an appropriate infection model needs to be studied. Here, we develop a model infection for lumpfish. Lumpfish were infected intraperitoneally with three doses of *P. salmonis* (low, mid, and high doses), monitored for 90 days, samples of head kidney and spleen were taken for RNA-sequencing and histology. *P. salmonis* infection in lumpfish showed over 98 % mortality, and differences between doses only showed a delay in mortality. A total of 3047 differential expressed genes (DEG) were identified at 21 days post-infection (dpi) in the head kidney and 1551 in the spleen, at 35 dpi showed 910 DEG in the head kidney and 3761 in the spleen. Gene ontology analysis showed enrichment in negative regulation of antimicrobial peptide production, nucleate erythrocyte maturation, natural killer cell differentiation involved in immune response, protection from natural killer mediated cytotoxicity, regulation of neutrophil differentiation, positive regulation of type IV hypersensitivity, and positive regulation of plasma cell differentiation at 21 dpi, at 35 dpi interestingly enrichment analysis showed the same enriched pathways, but except negative regulation of antimicrobial peptide production, probably the genes involved in this pathway could be essential to overcome *P. salmonis* infection.

GROWTH AND FEED INTAKE OF GST *Oreochromis niloticus* AT DIFFERENT TEMPERATURES

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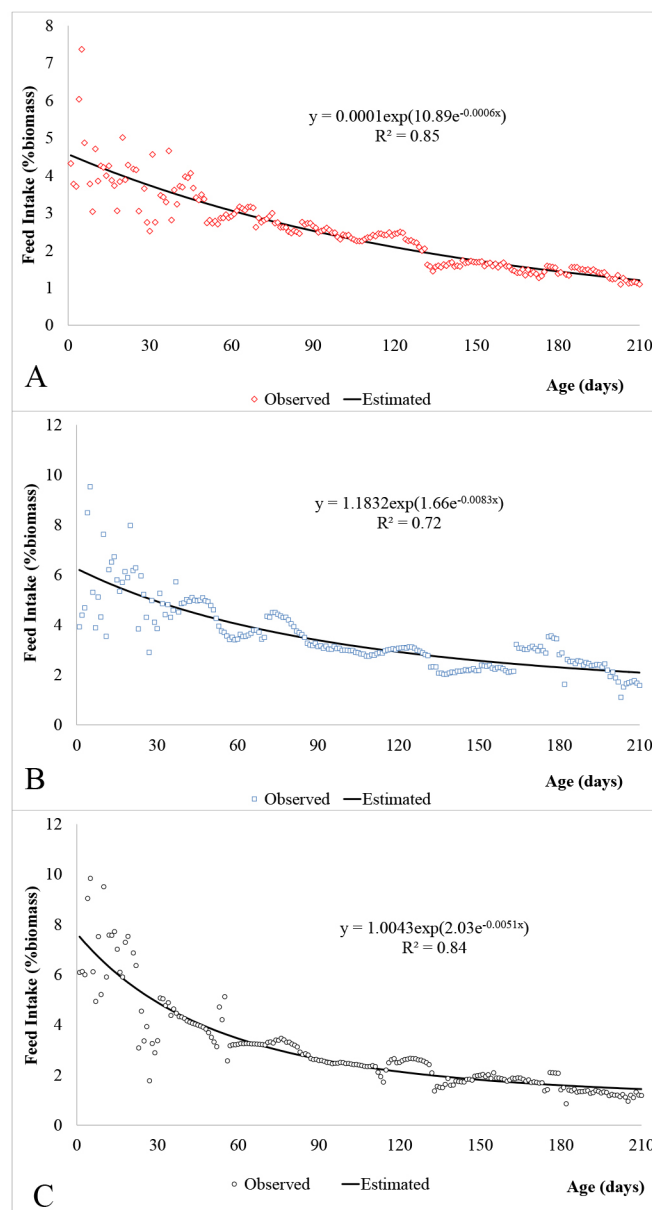
The growth and feed intake of Genomar Supreme Tilapia (GST) cultivated at different temperatures were determined. Tilapia fingerlings weighing approximately 8 g were grown in water recirculation systems with 0.5 m³ tanks (4 replicates) at a density of 80 fish m⁻³. The water temperatures were 22, 26 and 30°C. The fish were fed ad libitum three times a day with the same feed for each growth phase. After 30 min of feeding, leftover feed was collected, dried in an oven and weighed. Total feed supplied was calculated as %Biomass = (feed supplied – leftover feed) (tank biomass)⁻¹. Ten fish from each tank were weighed each 30 days until day 210th to determine the tank biomass. The nonlinear Gompertz models were fit by generalized least squares.

Fish reared at 30 and 26°C showed a higher specific growth rate (2.29 and 2.22 g day⁻¹, respectively) than those reared at 22°C (2.02 g day⁻¹). Feed intake increased along with temperature. Feed conversion was poorest in fish reared at 26°C (P<0.05). Final fish weight estimates at day 210 were 597.84, 819.26 and 1079.39 g for 22, 26 and 30°C, respectively.

Figure 1 shows the feed intake of GST reared at different temperatures. At 22°C, tilapia started to ingest 4.54% of biomass, and at day 210, intake was 1.20%. At 26°C, the variation was from 6.20% at the beginning to 2.10% at the end. At 30 °C, the intake was 7.51% at day 1 and 1.44% at day 210.

Figure 1. Estimation of feed intake (%biomass) for Genomar Supreme Tilapia reared at different temperatures. A) 22°C. B) 26°C. C) 30°C. Each point represents the mean intake of four tanks.

Acknowledgments: Financial support was given by Fapesp.



MUSCLE GROWTH OF GST *Oreochromis niloticus* AT DIFFERENT TEMPERATURES

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The objective of this study was to evaluate the muscle growth in Genomar Supreme Tilapia, generation 25, assessing the morphology of the white fibers. Tilapia fingerlings weighing approximately 8 g were grown in water recirculation systems in temperatures 22 and 26 °C. The fish are fed three times a day, with the same feed. The histology of muscle and subcutaneous ventral adipose tissue were evaluated at day 30th and 45th of cultivation to quantify cells diameter and density. Diameter cells were separated in different diameter class and plotted in a histogram. It was proceeded ANOVA considering $\alpha=0.05$ for significance.

At 30 days of cultivation, a similar fish weight of 36.00 (3.61) g and 41.25 (4.27) g was observed when grown at 22° or 26°C, respectively ($P>0.05$). No differences in fiber diameter and density (fibers / mm²) were found in fish grown at 26°C compared to 22 °C (Table 1).

At 45 days of cultivation, it was found that the weight of tilapia grown at 26°C was 65.66 g (1.89) and those raised at 22°C was 52.73 g (12.95) ($P<0.05$). The fiber density in tilapia at 26°C was higher compared to those raised at 22°C. This can also be seen in the greater fiber diameter of fish raised at 22°C compared to those raised at 26°C. In Figure 1, the presence of a higher frequency of fibers of 20-40 μm and lower frequency of fibers in the classes of 40-60 and 60-80 μm can be verified in fish reared at 26°C when compared to those cultivated at 22°C ($P<0.05$). This characterizes the process of hypertrophy of dorsal muscle fibers in tilapia raised at lower temperatures.

Acknowledgments: Financial support was given by Fapesp.

Table 1. Mean and standard deviation of muscle fiber diameter of tilapia reared in different temperatures.

Day	Temp. °C	Diameter (μm)	Density (fibers mm ⁻²)
30 th	22	43.09 (4.06)a	496.72 (71.19)a
	26	40.96 (5.06)a	431.13 (27.92)a
45 th	22	48.11 (3.35)a	473.89 (49.46)b
	26	36.71 (3.28)b	594.62 (100.83)a

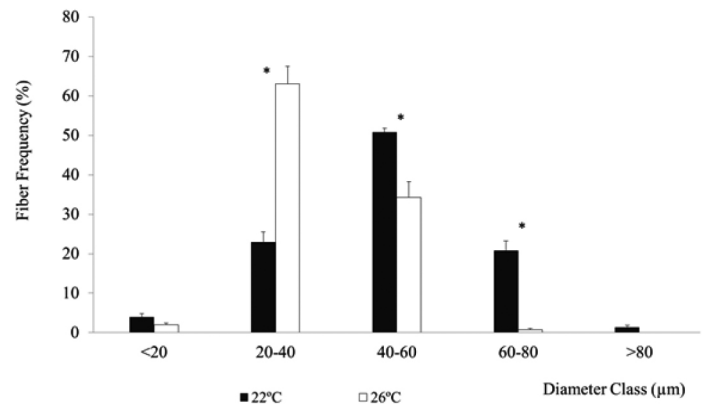


Figure 1. Frequency distribution of muscle fibers in different classes of diameters of tilapia grown at different temperatures at day 45th. *Significant differences between temperatures ($P < 0.05$).

SOLUBLE MANNO-OLIGOSACCHARIDES FOR PATHOGEN CONTROL IN AQUACULTURE

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Infections caused by *Vibrio spp.* and *Tenacibaculum spp.* have led to billions of dollars in losses in the aquaculture industry, yet there are few options to prevent or manage these infections in shrimp, salmon, and other aquaculture species.

Manno-oligosaccharides (MOS) are widely recognized for their ability to control pathogens such as *Salmonella spp.* and *E. coli* in the poultry and livestock industries, but are little used in aquaculture. GreenSage Prebiotics (GSP) has developed a high purity, soluble MOS targeted at key pathogens impacting aquaculture, animal, and human health.

GSP has conducted *in vitro* and *in vivo* trials to evaluate its novel MOS product (GSPMOS).

In vitro trials to determine the minimum inhibitory concentration (MIC) and minimum lethal concentration (MLC) of GSP-MOS versus *Vibrio parahaemolyticus* and *Tenacibaculum maritimum* demonstrated the effectiveness of GSP-MOS and the superiority of GSP-MOS versus low purity, insoluble yeast-based MOS products (Table 1).

Additional *in vitro* studies have demonstrated efficacy of GSP-MOS versus *Salmonella enteritidis* and *E. coli*, and have shown enhanced growth of beneficial microbes, including probiotics such as *Lactobacillus spp.*, which can collectively support the digestive and immune systems.

An *in vivo* *Vibrio parahaemolyticus* challenge trial was completed in shrimp, following two weeks of consumption of feed containing GSP-MOS. Shrimp consuming 0.25 wt% GSP-MOS had 100% survival following a *Vibrio* challenge, compared to 42% survival in shrimp that did not receive MOS (Figure 1).

This presentation will discuss the benefits of high purity, soluble MOS as a feed additive for control of pathogens, and the corresponding impacts on aquaculture and animal health.

Pathogen	MIC (mg/mL)		MLC (mg/mL)	
	GSP MOS	Yeast MOS	GSP MOS	Yeast MOS
<i>Vibrio parahaemolyticus</i>	5.55	ND	50	ND
<i>Tenacibaculum maritimum</i>	16.67	50	50	50

ND : No detectable effect

Table 1: MIC and MLC of MOS Products

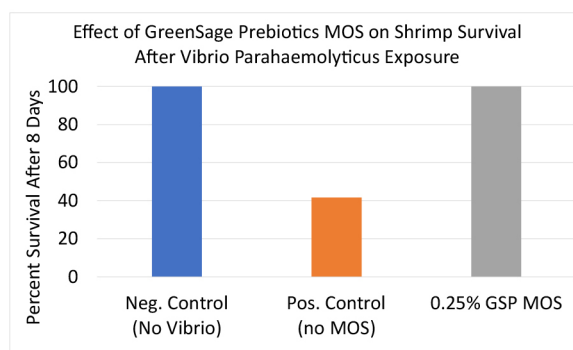


Figure 1: Impact of GSP-MOS on Survival After Vibrio Challenge

EXPERIMENTAL APPROACH AND METHOD FOR ISOTOPE LABELING OF MICROALGAE BIOMASS AND RELEVANT BIOMOLECULES FOR INFLAMMATORY AND METABOLOMIC STUDIES

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Stable isotope labeling is a powerful tracer approach with a wide range of applications in domains including biology, ecology, trophic dynamics, biomedical, pharmacology, drug design, nutritional, metabolomics and quantitative proteomics. There is a strong demand for biomass and biomolecules labeled with 13-carbon (^{13}C), 15-nitrogen (^{15}N) and (^2H) deuterium. Stable isotope-labeled compounds can either be chemically synthesized or produced through metabolic incorporation using living organisms. Microalgae are well suited for this task because they can easily be cultured, have a high metabolic plasticity to environmental changes, and are able to convert inorganic compounds into organic compounds through photosynthesis. High purity isotope-labeled substrates (>95%) are needed for biomedical, pharmacology and analytical purposes but are still expensive to produce. Therefore, the optimization of microalgae culture productivity and biomass and biomolecules quality labelling (>95%) is mandatory.

Exposure to deuterium is generally associated with negative impacts on microalgae cell survival, growth and division. This project puts forth an isotope labeling method for biomass enrichment of *Scenedesmus obliquus* with deuterium (^2H). After an acclimation period of several weeks at low concentrations of deuterium (10%, v/v), consecutive subcultures of *S. obliquus* were realized while gradually increasing (+10%) deuterium concentrations from 10% to 100% (v/v) in culture media. Cell survival, density and growth rate were observed and compared to a control culture. Exposed microalgae cells survived the increasing concentration of deuterium that followed the acclimation period but showed a delayed growth when compared to the control culture.

This project is part of a bigger study which aims to provide stable isotope-labeled biomolecules extracted from microalgae biomass. The isolated and purified lipids will be used to supplement human lung cell cultures in inflammatory challenge trials. Stable isotope tagging coupled with nuclear magnetic resonance (NMR) and mass spectrometry (MS) will help to understand the mobilization of active compounds during inflammatory processes symptomatic of Covid-19.



FIGURE 1. Picture of a *Scenedesmus obliquus* culture.

UPDATES ON DEVELOPMENT OF A RECIRCULATING AQUACULTURE SYSTEM FOR BIVALVE LARVAL CULTURE

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Climate change continues to challenge coastal aquaculture, particularly sectors heavily reliant upon ambient coastal water resources. Typical bivalve larval hatchery practices rely on pumping near-shore coastal waters to support flow through larval production. In such applications, suboptimal ambient water quality can cause impaired hatchery/larviculture performance. The application of recirculating aquaculture systems (RAS), has been investigated to allow reuse of larval production water, thereby minimizing impacts from fluctuating ambient coastal waters. This presentation provides updates from ongoing efforts in development of such a system in Virginia, integrating programming from the Virginia Institute of Marine Science Eastern Shore Lab, the Virginia Tech – Virginia Seafood Agriculture Research and Extension Center, Virginia Sea Grant, and Oyster Seed Holdings, LLC.

LESSONS LEARNED FROM OPEN OCEAN FARMS AROUND THE WORLD

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The extensive coastline of the east coast of Canada and the United States creates a large diversity of environments which each present a unique set of challenges and opportunities for aquaculture installations and operations. The coast has both sheltered sites for which traditional finfish farming equipment is suitable as well as exposed open water environments which require equipment and operating procedures that are specifically designed for such high energy sites. The viability of an open-ocean finfish farming industry is assessed in the context of the challenges and successes of existing farms in other countries and locales.

Fully exposed sites create engineering and operational challenges, which can be amplified by the high cost of labor in North America. However, the industry is transitioning towards more digital and automated operations making farms more viable and competitive. Local infrastructure within the region is also more amenable to operating a complex, technologically advanced farm than more remote sites. Furthermore, basing production closer to major markets and the ability to market a locally produced fish makes for a more compelling and profitable model. Examples of active open-ocean farms operating in foreign countries and remote indicate that an open-ocean industry in the Northeast North America could be viable.

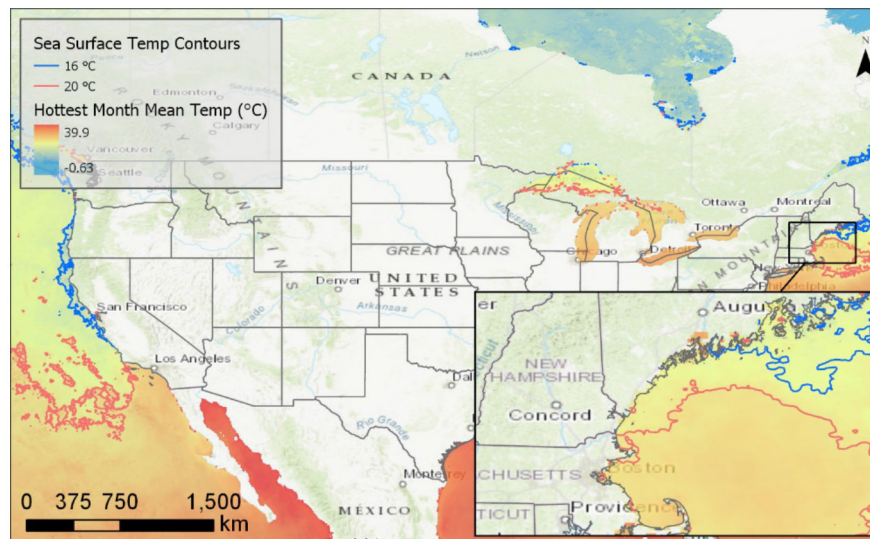


Figure 1: A simple model showing the potential expansion of suitable areas for salmonid production by submerging pens below a 4°C thermocline.

APPLYING LIVE DATA TO NET PEN FARM OPERATIONS

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Net pen aquaculture systems are intimately connected to the local environment. Significant benefits from ecosystem services are obtained such as removal of wastes, provision of well oxygenated water, and maintenance of a relatively stable temperature, but these are natural processes and they are subject to fluctuations. It is critical for farmers to monitor environmental conditions to ensure the health of their fish and to make sure operations are optimized for current conditions. Tools that provide live data streams wirelessly are the most suitable to create the spatial and temporal resolution necessary to apply the data and get the most value from it.

Temperature does not usually vary significantly over the spatial scale of a typical net pen farm, but it can change significantly on a temporal scale relevant to farm operations. A temperature change as small as 2°C has a significant impact on total feed intake and feed intake rate making it important to have live temperature data streams recorded hourly or faster to optimize feeding operations. Adjusting feeding rates and target feed amounts based on temperature can reduce feed wastage to the environment, particularly in low visibility environments when cameras are less useful. Additionally, extreme temperature events can be stressful for fish, compromising immunocompetence and even causing mass mortality.

Dissolved oxygen levels can vary significantly on very small spatial and temporal scales. During feeding or bathing, dissolved oxygen levels can drop by over 1% per minute. Regular measurements are essential to ensure that fish are maintained in safe conditions. Aside from farm operations, there are environmental events that can reduce dissolved oxygen content to sub-lethal levels within hours.

An environmental monitoring system that populates data onto a cloud allows for automated analysis and reactionary steps to be taken. These can be as simple as automated emails or text messages that are sent out when a parameter crosses a certain threshold, or entire systems can be automated to react to a changing environment. For example, an air diffuser system can be turned on automatically in response to low dissolved oxygen readings or high temperatures. This level of automation can improve fish performance and avoid critical issues which can lead to mass mortality events.

Live, uninterrupted data streams that report at a spatial and temporal resolution appropriate for the parameter that they measure, should be considered essential aquaculture tools. They enable farmers to achieve higher levels of efficiency and can also reduce the risk of critical issues leading to mass mortality events.

THE CHALLENGES AND OPPORTUNITIES FOR FARMING SALMON IN SUBMERGED PENS

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Open ocean aquaculture can offer both economic and environmental advantages over other culture methods. Oceanographic conditions in large areas of many countries, Canada and the United States included, are unsuitable for surface pens, making submerge options an interesting consideration for many farmers. The heavy waves at exposed sites can damage pens that are not designed for this level of energy causing excessive repairs costs, stress to fish, and escapes.

The development of submerged pen systems has the potential to enhance the feasibility and range of offshore production by keeping infrastructure out of surface waters and away from damaging wave energy. Submerged systems can offer further benefits by accessing sites that can support higher stocking densities, keeping fish out of extreme temperatures, and away from parasites near the surface. Submerged pens are being used successfully to grow several species around the world.

Salmonids and other physostomal species (fish with “open” swim bladders), cannot be maintained without access to air for extended periods. They require an air-water interface to gulp air and maintain an inflated swim bladder and neutral buoyancy. When held for too long without surface access, they start to modify their swimming to compensate negative buoyancy, and eventually start to experience stress and can suffer from injury from bumping into or rubbing on the pen net. FCR will increase as fish swim less efficiently and suffer injury, and mortality can result from the stress.

Both operational and engineering solutions are being developed to make submersible pens compatible for salmonids. Periodic emergence and submerged air reservoirs are two solutions being considered for successful production.

This talk will review the benefits of farming salmon in submerged pens, with a focus on the US and Canadian east coast and explore some of the solutions being considered for swim bladder inflation.

LEADING THE NARRATIVE IN SCOTLAND

Tavish Scott

CEO Salmon Scotland

- Salmon Scotland is the voice of Scottish farmed salmon, the UK's biggest food export.
- We are a source of trusted information and represent our members' interests in Scotland, across the UK and internationally.
- We raise our voice in the published media, on social media, and through political and stakeholder engagement.

SCOTLAND'S AQUACULTURE STORY – INFORMING AND EDUCATING GOVERNMENT, COMMUNITIES AND OUR STAKEHOLDERS

Tavish Scott

Salmon Scotland is the trade body for the UK's No 1 food export. Our sector dispatches 100,000 tonnes of Scottish salmon to 50 countries around the world. We have achieved 36% growth in value in 2021. We are an export success story for Scotland and UK plc despite the Covid pandemic and the UK leaving the European Union's single trading market on January 1st 2021. We sell into the UK retail market based on Scottish salmon's nutritional, healthy advantages and a significantly lower carbon footprint than beef, lamb or even free range chicken.

We use these facts with government and our wider stakeholders in building the case for sustainable growth – greater biomass grown in Scotland based on solid animal husbandry, farming innovation, acute regulatory oversight and international certification of our businesses. Science, evidence and data underpin Salmon Scotland's advocacy for our members.

World population growth, the UN's desire for protein production to be increasingly marine based and the ever altering political environment within the UK, create opportunities and challenges.

Our fundamental approach is to have government, communities and stakeholders understand what we do, why and how we involve them. We show and tell. We use school career days, supply chain events and farm open initiatives. The message is simple – we welcome people who want to learn, ask questions and probe how we look after fish. Be that a Prime Minister, a local councillor or the Head Teacher of the local community school. Never lose the politicians. Keep local people actively interested. And front up to the media, on social channels and with opinion formers. We have so much to be proud of as a sector both in Scotland and around the globe.

CYSTATIN C FROM *Amphiprion clarkii*; GROWTH INHIBITION OF *Escherichia coli* AND EXPRESSION LEVEL ANALYSIS UPON IMMUNE CHALLENGE

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Cystatins are cysteine protease inhibitors present in a wide range of organisms and involved in various physiological processes, such as protein degradation, arthritis, tumor invasion, metastasis and antigen presentation. Cystatin C is an extracellular polypeptide belongs to type 2 subfamily of cystatins. In present study, cystatin C from *Amphiprion clarkii* (AcCST3) was identified and structural and immunological role were studied.

The structural analysis of AcCST3 was performed using relevant bioinformatic web tools. The AcCST3 sequence contains cystatin superfamily domain and closely related to the CST3 sequence from *Amphiprion ocellaris*. The real-time qPCR analysis revealed the AcCST3 expression in all the analyzed tissues (blood, head kidney, spleen, liver, gill, intestine, kidney, brain, muscle, skin, heart and stomach). The immune challenge experiment followed by qPCR showed significant expression modulation of AcCST3 in spleen tissue with three of immune stimulants (*Vibrio harveyi*, polyinosinic: polycytidylic, and lipopolysaccharide) through the experimental period (6 h, 12 h, 24 h, 48 h, and 72 h post stimulation). Moreover, the growth inhibition effect of AcCST3 against *Escherichia coli* was observed by radial diffusion assay using recombinant AcCST3 protein (rAcCST3). The observed significant modulations in AcCST3 mRNA level upon immune challenge and the anti-bacterial activity of rAcCST3 suggested that the AcCST3 might have a potential role in innate immunity of *Amphiprion clarkii*.

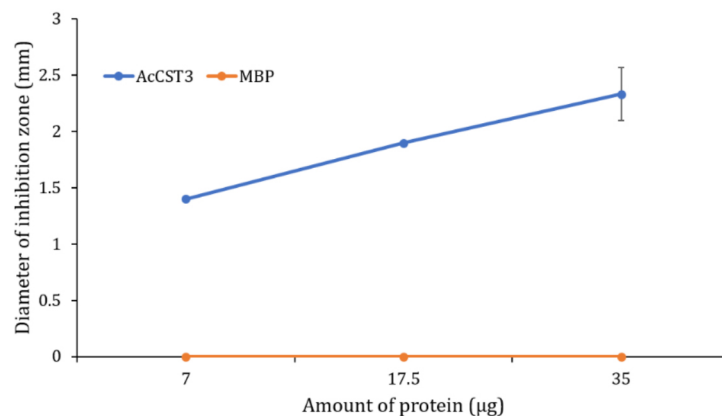


Figure: Growth inhibition effect of rAcCST3 against *E. coli* by radial diffusion assay. Wells were bored into underlay gel (1% agarose in 0.01 tryptic soy broth, 10 mM NaPB, pH 7.4) impregnated with bacteria. Proteins were introduced into wells (7-35 µg/well), and the plates were overlaid with 1% agarose in 2 × tryptic soy broth (6% wt/vol) and incubated overnight at 37°C.

CALCIUM-BASED BIOCHAR DERIVED FROM SNOW CRAB WASTE AS CATALYSIS FOR TRANSESTERIFICATION

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Waste from aquaculture and fishing is a major issue in coastal areas. The global fishing industry is estimated to produce 179 million tonnes each year, of which 156 million tonnes are used to produce food for human consumption. The remaining aquaculture is converted into low-value products like animal feed and fertilizers. With a harvest of more than around 5 million pounds, snow crab constitutes one of the largest harvested species in Nova Scotia. Crab legs and shoulders are the only pieces of the crab that are commercially valued, leaving around one-third of the crab untouched and going into waste composting or landfilling facilities, contributing to methane and CO₂ emissions. This waste, however, contains high-value organic and inorganic materials which can be reprocessed and used in a variety of higher-end products. The thermochemical conversation is a technique used to degrade biomasses thermally in a deoxygenated environment. This process yields two main products, known as biochar and biooil. The biochar generated from the pyrolysis of crab body is rich in Calcium which can be used in various applications including catalysis.

As we are moving away from fossil fuels, the demand for biodiesel as an alternative energy source is increasing. Consequently, the production of biodiesel from waste triglycerides such as vegetable, fish, and animal oils has also increased, resulting in the production of substantial quantities of crude glycerol of economic value as well as fuel. We have demonstrated that the calcium-based crab biochar can be implemented as a catalyst for transesterification of vegetable and fish oil generating a renewable biodiesel from two under-utilized waste streams.

REVISITING OCCUPATIONAL HEALTH AND SAFETY GOVERNANCE IN THE CANADIAN AQUACULTURE INDUSTRY

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The aquaculture industry has long been recognized as requiring shared governance of federal and provincial governments. Aquaculture cuts across several powers constitutionally allocated to the respective governments while ironically aquaculture itself is not expressly constitutionally assigned. Appellate caselaw in British Columbia suggests aquaculture is a federal responsibility, although the two levels of government appear to have adopted a pragmatic approach to enable the industry to develop. In a spirit of 'cooperative federalism' federal-provincial memoranda of agreement to coordinate responsibilities have been adopted and some provinces – namely New Brunswick, Newfoundland and Labrador, Nova Scotia and Quebec – are regulating the industry. Differently, the federal government regulates aquaculture in British Columbia but the province is responsible for leasing, whereas in Prince Edward Island the governance of the industry is through federal leasing and industry co-management.

This paper revisits the regulation of aquaculture in Canada and focuses on occupational health and safety jurisdiction. The labour process in the aquaculture industry involves both land-based hatchery work, fish feeding and harvesting work at sea. The work at sea aspects expose workers to occupational hazards concerning not only the aquaculture activity itself but also navigational and vessel operations.

The terrestrial-maritime dimension of the labour process raises questions for standard-setting and risk distribution in occupational health and safety regulation. In *Jail Island Aquaculture Ltd. v. R.* (2000), the shipowner challenged the provincial jurisdiction over a workplace death accident occurring on a fish harvesting barge at sea, but the New Brunswick Court of Queen's Bench left the jurisdictional question over aquaculture OHS unanswered. More recently the appellate courts of British Columbia and Nova Scotia considered OHS matters with respect to fishing vessels and had no difficulty in asserting provincial jurisdiction over the matter. The Supreme Court of Canada further considered workers compensation in a Newfoundland case and explored a possible way forward in clarifying federal and provincial jurisdiction over the claims of dependents. However, the two decisions raised concerns over the desirability of uniformity when an industry straddles provincial economies.

The paper analyses the jurisdictional questions and attempts to clarify the contemporary state of understanding concerning aquaculture OHS jurisdiction in Canada.

ABILITY OF AKIRIN-2 TO REQUITE INTO NF- κ B TRANSCRIPTION FACTOR COMPLEX WITH THE PRESENCE OF RNA STIMULUS; A STUDY OF ORNAMENTAL FISH *Amphiprion clarkii*

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Animal defence toward viruses are distinct compared to the bacterial or other pathogenic defences. Understanding of molecular mechanisms behind these defence mechanisms holds paramount importance when improving solutions for the virus diseases. This study describes the transcription factor Akirin-2 and its involvement in the virus defence which was identified from the ornamental fish *Amphiprion clarkii*. An immune challenge was conducted to disclose potential immune stimulants and responses for Akirin-2. Involvement in the NF- κ B transcription factor complex was analysed with a luciferase promoter assay and an antiviral gene expression assay.

Bioinformatics analysis revealed that the *A. clarkii* Akirin-2 (AcAk-2) as a highly conserved protein with two nuclear localization signals. Localization analysis with florescent tagged AcAk-2 was observed in the nucleus regardless of poly I:C stimulation. Expression analysis revealed the strongest expression with the presence of poly I:C. Overexpression of AcAk-2 solely unable to enhance the luciferase activity, but overexpression in the presence of poly I:C significantly enhanced the luciferase activity. *In-vitro viperin* expression was significantly enhanced after 24 h post-transfection of AcAk-2 gene. These results provide evidence for the role of AcAk-2 plays during the RNA virus infections.

USING EDUCATION NETWORKS TO BUILD PUBLIC TRUST AND ENHANCE AQUACULTURE LITERACY

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Public awareness of aquaculture plays an important role in unlocking its full potential to support healthy people, a healthy planet, and a healthy economy. Still, negative public perceptions of aquaculture are often fueled by lack of familiarity with, or misinformation about, the sector. This low public aquaculture literacy creates significant barriers to sustainable aquaculture expansion in North America. Environmental educators and informal learning institutions provide the necessary platforms through which to build trusting relationships with, and provide credible aquaculture information to, diverse public audience groups. Aquaculture education, outreach, and communication strategies that help consumers confidently navigate their decisions can improve aquaculture literacy and paint a more holistic picture of each community's unique relationship with the industry.

Here, we discuss how NOAA is pairing with the North American Association for Environmental Education (NAAEE), aquariums, the aquaculture industry, and other outreach groups to develop a Community of Practice for Aquaculture Literacy (CoPAL). Join this interactive discussion to learn about ten cross-sectoral partnerships that are improving public awareness of local aquaculture industries and connections to working waterfronts. You will also have a chance to participate in an open discussion about how to build stronger knowledge-sharing relationships with working waterfronts, including how to ensure equitable partnerships across diverse stakeholder groups. No one sector can accomplish this alone and the time is now to discuss how we can help each other build coalitions for aquaculture literacy.

AN OVERVIEW OF THE AREA-BASED AQUACULTURE MANAGEMENT INITIATIVE IN BRITISH COLUMBIA

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Area-Based Aquaculture Management (ABAM) is a new initiative in British Columbia (BC) which aims to have governments, stakeholders, and industry working together to spatially plan, manage, monitor, and improve aquaculture activities. This will occur within distinct geographical areas so the unique jurisdictional, ecological, social, cultural, and economic values can be considered.

In 2018, the former Minister of Fisheries and Oceans Canada, announced that Canada would “work in partnership with the provinces and territories, industry, Indigenous partners, environmental groups and other stakeholders to ensure an economical and environmentally sustainable path forward” for aquaculture in BC.

To support this path forward, a proposed framework for ABAM was developed. The framework recommends concrete actions for ABAM to enhance the sustainability and introduce collaborative governance structures to aquaculture management in BC.

Enhancing coordination and collaboration between the Federal Government, the Province and Indigenous groups is a key focus of ABAM. Through collaboration, DFO hopes to advance reconciliation, strengthen the role of Indigenous groups in aquaculture planning and management, improve transparency and decision-making, adopt an ecosystem-based management approach, and improve social and economic benefits for communities in BC.

In Budget, 2021, DFO was granted \$3M over two years (2022-2024) to pilot ABAM in BC. Area-based management approaches will be piloted at both a regional and area level. In Fall 2021, DFO invited interested partners to submit an expression of interest application for consideration for the initial pilot area(s) in BC. A decision on the pilot area(s) is anticipated in Spring 2022.

DFO will work with partners to develop a regional committee which will provide guidance from a province-wide perspective and the area committee(s) which will collaborate on tools and materials to support the pilot area and development of an area management plan.

During the ABAM Pilot, DFO will work with partners to demonstrate successes of area-based management approaches, build tools and materials, identify challenges, and share lessons learned to be carried forward in aquaculture management in BC.



SAXITOXIN ACCUMULATION AND DEPURATION IN ROCK SCALLOPS *Crassadoma gigantea*: A PREREQUISITE FOR CULTURE ON THE WEST COAST OF NORTH AMERICA

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The rock scallop *Crassadoma gigantea* can achieve market size in 2-4 years and shows promise for commercial culture along its native range, from Baja California to southeastern Alaska. One serious unresolved issue, however, is the lack of information on accumulation and retention of algal toxins that can cause paralytic shellfish poisoning (PSP) in humans. This is critical because toxins associated PSP, e.g. saxitoxin (STX) and its derivatives, are currently the most widely reported toxins in bivalves along the west coast of North America. To address this issue, hatchery-reared rock scallops were deployed at three locations prone to PSP closures in Washington State, and sampled when a closure occurred and weekly thereafter until the closure was lifted. Induced toxicity under controlled conditions of algal cell density, temperature and salinity were also conducted at the NOAA Northwest Fisheries Science Laboratory in Manchester, WA. This work represents the first attempt to induce toxicity in *C. gigantea*.

The overarching goal of this research was to address the needs of public health agencies and shellfish producers by investigating where biotoxins accumulate in scallops, and how long the toxins remain. Temporal field exposure trials and lab experiments examined bioaccumulation and subsequent detoxification of STX in the adductor muscle and viscera of individual scallops. Both field and lab studies demonstrated that rock scallops can attain very high toxin loads for long periods of time, but toxicity was generally confined to the viscera (gut). Mean toxicity levels in rock scallop adductor muscles (the part that is consumed) were below the regulatory limit for STX (80µg/100g tissue). Induced toxicity experiments revealed a pattern of toxification and detoxification similar to other studies of bivalves fed toxic dinoflagellates, with rock scallops increasing toxin loads in digestive gland tissues at significantly higher rates than for other tissues (Bricelj and Shumway 1998). It is not clear from any of our datasets, field exposure or laboratory, how long rock scallops take to completely depurate STX from all tissues. This aspect of research will need to be further explored with future monitoring efforts to identify complete depuration rates. This data gap aside, it is clear that very high, persistent levels of STX in scallop viscera will likely preclude this species from safe, whole product consumption. The potential for a shucked, adductor only market will also require careful scrutiny due to persistent toxicity and variability of toxicity among individual rock scallops. This work was supported by NOAA-OARSG-2016-2004807.

MICROPLASTICS, MOLLUSCS, AND THE MISINFORMATION HIGHWAY

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Microplastics are a contaminant of global concern and, as such, there has been a rush to action and publication. Over the past decade, this haste has resulted in a chaotic and cluttered literature that is rife with inappropriate methodologies, unrealistic experimental protocols, misinterpreted results, and overstated significance. A comprehensive critical assessment of the current literature on interactions between particle-feeding molluscs and microplastics and their purported impacts (> 600 publications) is underway. It is not surprising that microplastics have been noted in shellfish guts globally. What is surprising is the extremely low level of particles routinely recorded (see Table 1 and references therein). The data to date clearly demonstrate extremely low numbers of microplastics in bivalve molluscs globally. There are no unequivocal data demonstrating that their presence in filter-feeding bivalve molluscs is a serious risk to human health and few data to demonstrate negative impacts on the shellfish. Many studies on suspension-feeding bivalve molluscs and other invertebrates are weak. There is a recurring presence in the published literature of misunderstanding of the feeding processes, capabilities for particle selection and rejection, and species-specific differences that all leads to misinformation, misinterpretation, and incorrect assumptions regarding potential impacts. There are major shortcomings to many laboratory studies that examined uptake and accumulation of microplastics by bivalves and their subsequent effects. In most cases, the issues can be traced to poor experimental procedures and animal husbandry, and lack of knowledge of the literature. They are compounded by a misunderstanding of the basic biology and physiology of molluscs. The shortcomings have led to a seriously flawed literature based upon the interactions and impacts of microplastic on these animals. Bivalves and other particle-feeding molluscs are complex living organisms with extraordinary capabilities for the control of selective capture, ingestion, and egestion of particulate material. They should be recognized and treated as such in any attempt to describe impacts of stressors, including different particle types, on their feeding and ability to accumulate materials. Any future experimental studies need to be focused carefully, based upon clear questions, use standardized analytical procedures, demonstrate a knowledge of the animals being studied, and an understanding of the literature extant. The hype needs to be curtailed and scientists should not imply impacts or potential impacts when there are no data to support the suppositions. Editors of scientific journals must make a stronger effort to engage qualified peer-reviewers and stop the flow of poorly done studies and superficial reviews that do nothing more than confuse the literature and reinforce prior inadequate studies and potential reviews. Much of this review is excoriating and that is by design – the current state of the science and literature demands it.

IMMUNO-EFFICACY OF IMIDAZOLE ANTIFUNGAL DRUG IN CONTROLLING OOMYCETES INFECTION IN *Labeo rohita* FINGERLINGS

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The present study evaluated the effect of sub-lethal doses of antifungal drug miconazole nitrate (MCZ) on immunological responses including immune related gene expression and its role as a prophylactic drug against *S. parasitica* in *Labeo rohita* fingerlings. Fish were fed with sub lethal doses of MCZ i.e., T1- 6.30 mg MCZ kgBW⁻¹, T2- 12.61 mg MCZ kgBW⁻¹ and T3- 25.22 mg MCZ kgBW⁻¹ and sampling was done at different time intervals for 240 h. Immunological parameters viz. lysozyme activity, oxygen radical production and plasma anti-protease activity showed significant enhancement ($p < 0.05$) in fish fed with T2 and T3 doses. Significant reduction in plasma protein content was observed in all the dietary groups as compared to control. Expression of immune relevant genes like TLR-22 and β 2-M showed significantly higher expression at 6 h and 24 h of sampling in both liver and head-kidney. However, these genes showed a down-regulation after 120 h of sampling in both the tissues. Preventive efficacy study showed that single dose of MCZ provides protection against oomycetes up to fourth day of infection. Significantly higher mortality was observed in control diet fed fish as compared to fish fed with MCZ medicated diet. Thus, from the study, it can be concluded that the MCZ can act as a potent antifungal agent for preventing oomycetes infection as well as to enhance immune response.

ASSESSING THE EFFECTS OF INTRODUCING NEW REGULATIONS OVER SEA LICE MANAGEMENT AND CONTROL IN ATLANTIC SALMON FARMS IN BRITISH COLUMBIA, CANADA

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Farmed Atlantic salmon are frequently infected with sea lice (*Lepeophtheirus salmonis*), which can increase the exposure of wild out-migrating salmon to this parasite. To minimise this, in 2020, Fisheries and Oceans Canada introduced several changes to marine finfish licences.

Our objectives were to describe current and past sea lice management and control practices and to assess the effects of the 2020 changes to conditions of licence.

Datasets with mitigation events and industry-reported sea lice counts were used to determine sea lice abundance for each event from two weeks before the intervention to eight weeks after. Exploratory data analysis was used to describe changes in sea lice abundance over this 11-week period while a Cox proportional hazards model was used to compare how long it took to reduce sea lice abundance below regulatory thresholds.

Our analysis suggests that when an intervention was implemented, the abundance of sea lice was reduced. However, the rate and duration of this reduction depended on the type of mitigation used. Additionally, we determined that abundance at the start of in-feed medication was associated with the time needed to reduce sea lice under threshold. Compared to previous years, interventions after the introduction of new regulation favor the use of mechanical and bath treatments and are initiated at lower abundance. Using our cox model, we found that the time needed to reduce sea lice has also reduced significantly. While in previous years about 50% of the mitigation events managed to reduce lice below threshold in the first month, almost 90% of the interventions currently achieve that objective.

In conclusion, as farmers have incorporated new mitigation alternatives in response to challenges such as the appearance of resistance to emamectin benzoate and the implementation of new regulation, we have observed their ability to manage and control sea lice infestations has improved. Although these changes cannot be attributed to regulation alone, our findings suggest availability of multiple therapeutical tools and active management of sea lice to mandatory thresholds have played an important role in the farmer's ability to control sea lice.

RISK OF GENETIC INTROGRESSION OF DOMESTICATED ESCAPED SALMON INTO WILD ATLANTIC SALMON POPULATIONS

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The Norwegian risk assessment of aquaculture uses Bayesian network structures for visualizing risk, including risk of further genetic introgression of domesticated salmon escapees in wild populations. Every year thousands of farmed Atlantic salmon (*Salmo salar* L.) enters the wild environment, and thus far introgression have been documented in two thirds of the populations investigated. Here we present the results of the 2022 risk assessment executed across 13 large-scale production areas, and the ongoing work towards executing risk assessment at a higher resolution: geographical dispersal of escapees, rivers-specific attractiveness of escapees and population-specific vulnerability towards introgression.

There are five factors included in the current risk assessment for further introgression of domesticated escapees. Together these factors represent the numbers and proportions of domesticated escapees in spawning populations and the resilience of the wild population; (1) the number of escaped fish reported to the authorities; (2) the observed proportions of escapees in a river as reported by the national monitoring program; (3) the removal of farmed escapees pre-spawning; (4) the demographic status of the wild populations (spawning target status) and (5) the genetic status (level of introgression) of the wild populations.

Aquaculture production is regulated through 13 production areas. The 2022 risk assessment shows that only the two southernmost (production area 1 and 2), as well as the northeastern production area (area 13) are considered to have a low risk of further genetic change (further introgression) as a result of escaped farmed salmon. Three of the other production areas (5, 6 and 12) are considered to have a moderate risk of further genetic change, while seven production areas (3, 4 and 7–11) are considered to have a high risk of further genetic change as a result of escaped salmon.

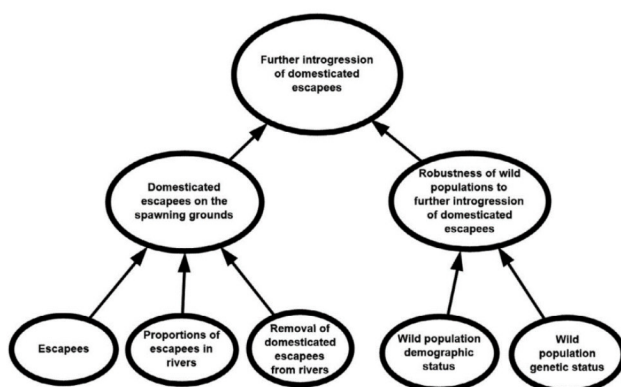


Figure 1: Factors included in the risk assessment for further genetic introgression of domesticated escapees

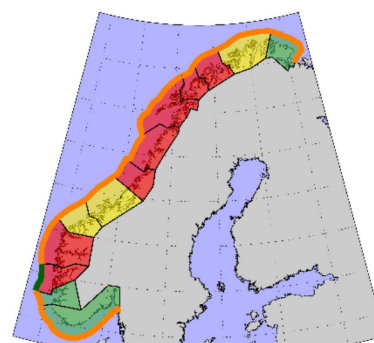


Figure 2: Risk of further introgression

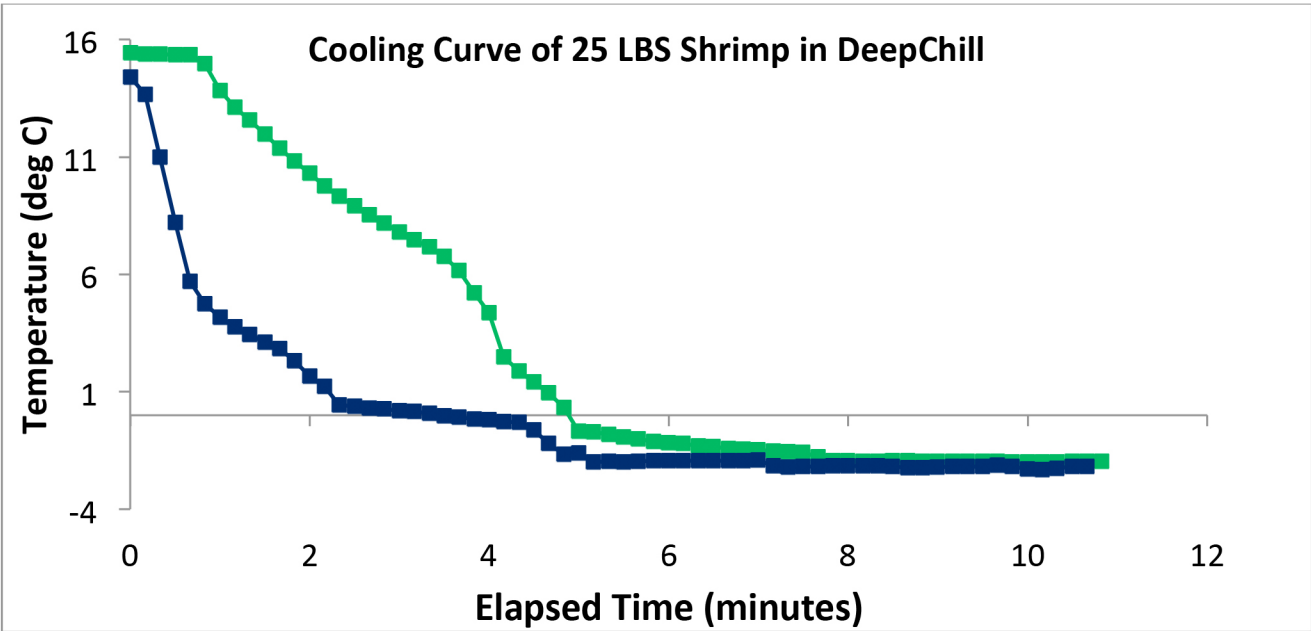
ABSTRACT: COMPREHENSIVE BENEFITS OF DEEPCHILL ICE SLURRY FOR SUPERIOR PRESERVATION AND YIELD GAIN IN SHRIMPS

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Effective and rapid seafood preservation is an important concern for aqua culturists. With better seafood preservation comes higher quality to consumers and higher premium to farmers. The use of flake ice, refrigerated sea water (RSW) or tube ice offers cooling that is fairly laborious, does not provide effective cooling below subzero temperatures, and involves fairly significant drip losses. In addition, handling of any form of crushed ice needs manual intervention thereby increasing the risk of contamination

DeepChill® is a proprietary ice slurry technology that is be able to address these concerns. As a true variable state ice slurry, it is pump able and the amount of DeepChill crystals in the slurry can be varied depending on heat transfer requirement. This paper presents the DeepChill® advantages for cooling farm raised shrimps. It must be noted that DeepChill is also effective for salmon, mussels and many other aqua cultured species.



OPTIMIZATION OF JUVENILE LUMPFISH REARING: STOCKING DENSITY AND TANK MODIFICATIONS

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The use of lumpfish as cleanerfish for salmonid aquaculture is a novelty and as a result, many aspects of lumpfish culture have yet to be perfected. Studies addressing juvenile stocking density and tank modifications to optimize growth and functionality, areas that may improve fish production, are foci of on-going research at the University of New Hampshire Coastal Marine Laboratory.

Juvenile lumpfish can be cultured at high densities compared to many other species, however, the optimal stocking density is unknown. Because lumpfish have ventral suckers and adhere to any smooth surface, growers often use available surface area as a way to calculate the amount of fish to stock into a system. However, lumpfish are cannibalistic so overstocking a tank may prove counterproductive for operations. Using common garden experiments with 3-liter tanks as experimental units, we are testing various densities (10-40 g/L) with age-0 fish. Growth rate, survival, and aggressive behaviors are the metrics being used to identify best stocking densities for small lumpfish.

Lumpfish will stick to any available smooth surface, including the tank bottom, making routine tank cleaning and waste removal difficult, often resulting in many fish being siphoned off of the bottom inadvertently. Further, while live feed is not necessary from a fish developmental standpoint and, on one hand, *Artemia* replacement diets may simplify larval rearing, their use requires increased tank cleaning during a fragile ontogenetic period. With those limitations in mind, a series of tank modifications are being evaluated to improve lumpfish rearing tanks using colors, textures, lighting, and drainage options to increase fish survival yet aid in waste removal. Growth and mortality rates as well as behavioral metrics (tank location preference) and scalability are being used to identify which tank modifications show the most promise for commercial production of lumpfish.

The outcome from these experiments will result in greater and more profitable lumpfish production in hatcheries and grow-out facilities to support cleanerfish use in salmonid farms.

NUTRITION AND DENSITY DEPENDENCE AS PROXIES FOR MANIPULATING JUVENILE LUMPFISH (*Cyclopterus lumpus*) GROWTH

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Lumpfish are of interest to the aquaculture industry because of their usefulness as biological controls for sea lice infestations in salmonid ocean farms. Despite millions of Lumpfish produced each year for this purpose, Lumpfish rearing protocols can be improved by understanding the nutritional needs of and density dependent effects on juvenile fish growth. During 2020-21, a series of experiments evaluating the effects of nutrition and stocking density on the growth, survival, and aggression of juvenile Lumpfish were conducted at the University of New Hampshire's Coastal Marine Laboratory.

The effects of varying protein (50, 55 %) and lipid concentrations (10, 15, 20 %), as well as plant versus fish meal-based protein sources, on the growth, survival, and aggression of juvenile Lumpfish were evaluated using six experimental diets and two commercial diets. In general, varying protein and lipid concentrations did not negatively affect growth, survival, or fish aggression of juvenile Lumpfish; overall mean percent growth ranged from 633 to 781 %. The use of plant-based protein (and a diet formulated for salmonids), however, suppressed juvenile Lumpfish growth; overall mean percent growth was only 394 %.

Juvenile Lumpfish are aggressive and will bite each other's fins resulting in severe caudal damage and ultimately death. To determine if this behavior is ontogenetic and density dependent, fish growth, survival, and aggression of two different fish sizes (2 g and 13 g) at four different constant rearing densities (40, 60, 70, and 90 g/L) were measured. For both size classes of Lumpfish, stocking density was negatively correlated with fish growth, but stocking density did not affect either fish survival or fish aggression. Overall mean percent growth of small juveniles ranged from 167 to 307 % (Figure 1), and from 286 to 471 % for large juveniles.

Lumpfish hatchery managers can use this information to manipulate lumpfish growth rates to better match hatchery production to salmonid farms' cleanerfish needs.

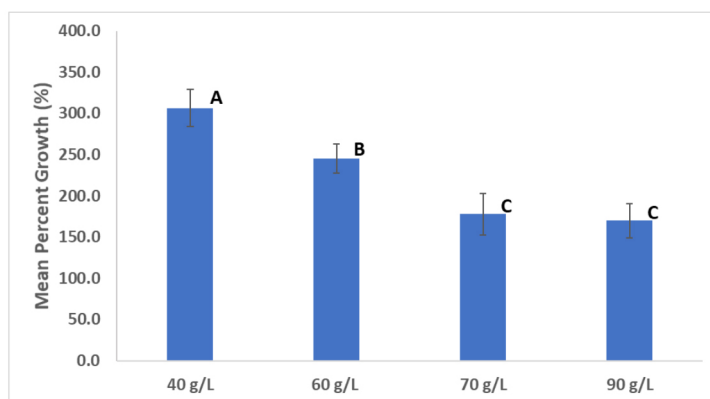


Figure 1. Overall mean percent growth (\pm one standard deviation) of small juvenile Lumpfish reared under varying densities for 10 weeks.

REMOTE AUTOMATED AERATION: THE BENEFITS OF CONTROLLED AERATION IN AQUACULTURE

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Fish farming suffers from a number of environmental factors that affect productivity and mortality rates. On the West Coast of the Americas, between British Columbia down to Chile, Harmful Algal Blooms (HAB) and Plankton is a large cause of fish stock mortality on an annual basis. In the Atlantic, from Newfoundland to Scotland, Iceland and Norway, extreme ocean temperatures exist (high in the summer and low in the winter) causing fish stocks to dive deeper resulting in lower dissolved oxygen (DO) levels causing mass mortalities. Internationally, fish farming suffers from sea lice issues where skirts and tarps are dropped around the pens, which causes stagnant water inside the pens and a corresponding decrease in DO. Furthermore, the increased frequency of in-situ net cleaning is driving more debris into fish gills, that can be a contributor to gill health disease and poor environmental conditions. Controlled Aeration can be a tool to mitigate a number of these issues, and Remote Automated Aeration can be a cost-effective measure to optimize the life support operations for the fish farming industry.

Aeration systems are an important defensive tool-in-the-toolbox for the fish farm operators to utilize in the pens in order to adjust and take control of environmental parameters such as DO, temperature, salinity, and HAB cell counts. Controlled Aeration has been effective at increasing DO levels with pens utilizing Sea Lice Skirts and HAB Tarps, allowing farms to run deeper skirts and tarps to better protect the fish stock. CPI has developed an Automated Controlled Aeration platform, called the *ODiN TITAN*, which allows operators and farm managers to remotely control the amount of air and oxygen being dispersed in the pens from the feed barge or at a centralized head office. When coupled with in-pen environmental monitoring sensors, the Automated Controlled Aeration platform can adjust and react to poor in-pen environmental conditions, and then increase or decrease the air or oxygen flow in order to push out HABs and debris, up-well water from depth to mix high or low surface water temperatures, circulate stagnant pen water, and increase DO saturation levels. In addition, the *ODiN TITAN* can collect, store and profile historical data which is synchronized to local weather, wind, tidal and current conditions in order to develop analytical models that allow fish farming companies to optimize their operations and reduce mortalities on a cyclical annual basis. The *ODiN TITAN* provides a verification method to head office and insurance companies to ensure that in the case of a mortality event, that it can be proven that the operators did all they could with the Life Support tools available.



PROTEIN INDUSTRIES CANADA – POSITIONING CANADA AS A GLOBAL SOURCE OF HIGH-QUALITY PLANT PROTEIN FOR USE IN AQUACULTURE AND OTHER INDUSTRIES

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Protein Industries Canada (PIC) is an industry-led, not-for-profit organization created to position Canada as a global source of high-quality plant protein and plant-based co-products. We are one of Canada's five innovation Superclusters.

PIC works with private sector industry partners to create co-investment projects that have the potential to transform the aquaculture, agriculture, and food production sector, allowing Canada to secure our position as a global leader in the production of plant-based products and co-products.

The goal of the Supercluster is to challenge Canadian businesses to collaborate with other businesses, and post-secondary and research institutions to create projects that have the potential to transform the food processing sector in Canada, creating jobs and stimulating local economies.

PIC works with companies to gain pathways to customers, markets, and partnerships that they could not access on their own, thereby providing them with the opportunity to leverage their expertise and innovations with that of large and multinational enterprises, public sector organizations and academia.

PIC has provided funding and supported the work of companies developing ingredients and products for use in aquaculture. Three of these projects will be summarized and discussed.

PRECISION FARMING DRIVING INNOVATION AND THE BENEFITS TO ACHIEVING AND MAINTAINING MULTI-SITE CERTIFICATIONS

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Grieg Seafood BC Ltd. (Grieg) and Scoot Science of Santa Cruz, California partnered together to create an innovative ocean analytics and data management platform that provides real-time data on ocean environmental conditions to Grieg's Atlantic salmon farms in British Columbia. This platform, the SeaState Dashboard, unifies existing sensor networks on farms, collecting cover one million data points every day, with publicly available data to provide a clear window into how salmon farms react to changing ocean conditions. The SeaState platform uses predictive modelling to forecast the environmental conditions up to three days in advance with extreme accuracy. This means better predicting ocean trends, giving Grieg an opportunity to reduce its exposure to marine risks, such as harmful plankton blooms and sea lice outbreaks, improve fish welfare and efficiency and support conformity with certification standards.

However, to have operational value from data-driven decision support, effective mitigating measures must be available. Grieg is currently trialling floating, semi-closed containment systems which are the first of their kind in BC. Semi-closed technology uses impermeable barriers between the farm-raised fish and the surrounding ocean to prevent sea lice and harmful plankton from entering (or leaving) sea pens. Deep water is pumped into the farmed enclosure, bringing cooler, higher oxygenated water to the farmed fish. Plankton and sea lice are generally found in the top layers of the water column. Pumping deeper water into the pens reduces interactions with both plankton and sea lice.

The SeaState platform combined with the semi-closed containment system enables Grieg to achieve and maintain conformance with challenging certification indicators in the BC Coast, including maintaining sea lice thresholds. Grieg is also aiming to have all 16 operational sea sites included in our current ASC Multi-site Certification certificate.

This presentation will briefly review the concept of precision farming, data-driven decision-making processes, the benefits of semi-closed containment and how they support sustainable certification standards .

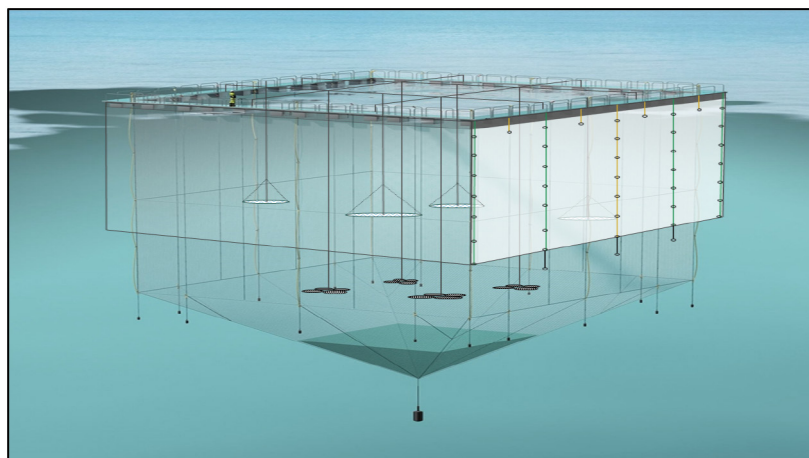


Figure - Schematic of semi-closed containment system.

DIET- AND ESCAPE-HISTORY AMONG FARMED ATLANTIC SALMON *Salmo salar* ESCAPEES CAUGHT IN NORWEGIAN RIVERS AND ON THE COAST IN THE PERIOD 2011-2021

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Each year, thousands of farmed Atlantic salmon (*Salmo salar*) escape from net pens in Norway. Although the majority don't survive until maturation, some escapees enter rivers where they may interbreed wild salmon. Farmed salmon display reduced spawning success in the wild, but how long the escapees have been on the run is likely of importance for their reproductive success. Due to their more natural life cycle, farmed salmon that escape early in the production cycle (early escapees) and survive to maturation could be better competitors and consequently contribute more to introgression than mature farmed salmon that have recently escaped (recent escapees). However, knowledge about time-since-escape of farmed salmon approaching the spawning grounds, and the variation in escape history in time and space, is limited.

Shortly after escaping, farmed escapees have a fatty acid (FA) profile which reflects the aquaculture feed. However, early escapees, which likely escaped as smolt, will, as adults, have the same FA profile as wild salmon. Thus, the terrestrial FA 18:2n-6 can be used as a marker for the aquaculture diet and separate the escapees based on time-since-escape (Fig. 1). Little is known about farmed escapees' long term diet post-escape. Using FA profiling, we are able to not only separate the farmed salmon by time-since-escape but also examine the diet of escapees and wild salmon to investigate whether the diet is the same for the escapees which successfully made the switch to marine feed. In addition, two genes (*vgl3* and *six6*) that are hypothesized to be linked to diet were examined and connected to the individual's FA profile to determine whether these genetic markers affect the FA profile and, thus, long-term dietary choices.

The overall aim of this project was to investigate the escape history and compare the diet of escaped salmon to wild salmon. This was achieved by distinguishing escapees from wild individuals using scales, fatty acids both as biomarkers and time-since-escape assignment, and genotyping of salmon caught in four rivers and marine fisheries in Norway from 2011 to 2021.

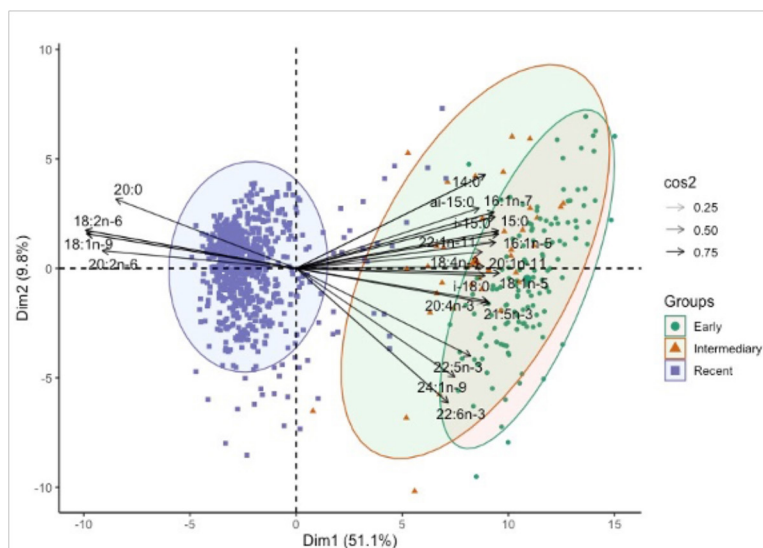


Fig. 1. Biplot over the principal component analysis of FA composition (% of total TAG) for recent, intermediate, and early farmed escapees. The 95% confidence ellipses are of pre-assigned groupings (based on 18:2n-6 values)

THE USE OF PERACETIC ACID IN AQUACULTURE

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Peracetic acid (PAA) is a promising disinfectant for biosecurity in the US aquaculture industry to prevent disease outbreaks from fish pathogens. PAA is a stabilized mixture of acetic acid, hydrogen peroxide and water that breaks down quickly to water and vinegar. PAA has greater reactivity and lipid-penetrating properties than H_2O_2 alone and is not deactivated by catalase and peroxidase (naturally occurring on organism membranes) which happens with H_2O_2 . Thus, PAA eliminates unwanted organisms easier and faster.

It is being increasingly used to replace chlorine in many industries. The U.S. Environmental Protection Agency (EPA) first registered PAA as an antimicrobial in 1985 for indoor use on hard surfaces (e.g., hospitals). Registrations have been expanded to include: sanitation in food/beverage plants, agricultural premises, wineries/breweries, greenhouse equipment, animal housing, as well as commercial laundries, prevention of bio-film formation in pulp/paper industries, and as a disinfectant for wastewater treatment. PAA is approved for use in Denmark, Germany and Norway as a water disinfectant, and our labs international collaborations have studied its effectiveness to many pathogens including *Ichthyophthirius multifiliis*, *Saprolegnia* spp., and various bacteria.

PAA is registered as a disinfectant for the US aquaculture industry for two products:

PeroxyChem's VigorOx® SP-15 Antimicrobial Agent is for: 1) Sanitizing surfaces of harvesting equipment used in the aquaculture industry, and 2) Cleaning and disinfecting fish culture tanks and raceways when water is drained and fish are not present.

AquaTactics' Aqua Des™ is for: 1) In-water use in fish ponds/raceways (remove fish from pond prior to use and test for residual PAA levels prior to restocking), 2) Use on aquaculture equipment (pumps, boots, foot bath mats, net dips, waders, dive equipment, etc.), and 3) Use on fish pond equipment (water free raceways, gratings, pipes, etc).

Aquaculture research to date will be discussed.

PRODUCTION OF TRIPLOID SUNSHINE BASS BY HEAT-SHOCK

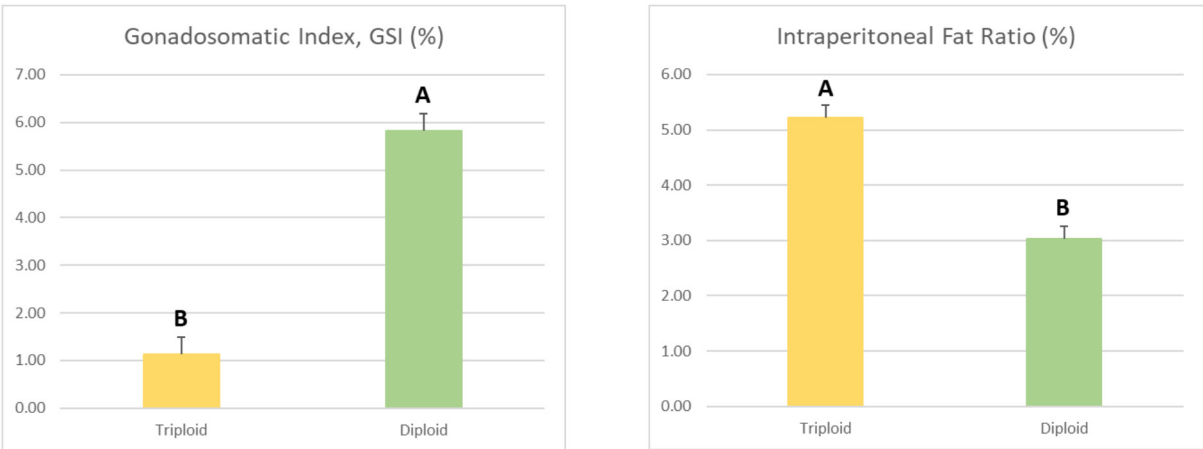
D.L. Straus*, A.M. Kelly, H.E. Quintero, S. Rawles, M. McEntire and J.W. Abernathy

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Sunshine bass are an important food fish raised in US aquaculture. Spawning of these hybrid striped bass is done by manual fertilization of female white bass eggs with male striped bass sperm. The industry grows these fish to ~1.5 to 2.5 lbs over ~1.5 to 2 years, during which time the females can become fertile and produce eggs. This is a major problem in the industry, since producers usually sell these fish whole. Loss in weight due to expulsion of eggs caused by handling stress often results in lower market appeal. Therefore, the industry needs a way to produce sterile (triploid) fish.

In this study, triploidy was induced through temperature shock applied to the embryo shortly after fertilization. We chose a range of temperatures and times post-fertilization to shock the eggs during initial trials. Larvae produced were checked for triploidy using a flow cytometer. Based on these data, we chose the highest triploid rates from the cold- and warm-shock treatments for a commercial-scale trial.

There were few survivors in the cold-shock treatment and none of these were triploid (verified using a Coulter Counter). Triploidy of the warm-shock treatment was 7% (66 fish). Triploids and an equal number of diploid controls were raised in flow-through tanks until spring spawning 2020 to verify maturation/gamete production vs diploid fish. Average growth rates were similar. Most triploids had undeveloped gonads; 5 were obvious males and had well developed testes; 9 were presumptive females and had grainy ‘ovaries’, only 1 had developed eggs (~100). Triploid condition factor, gonadosomatic index (below) and intraperitoneal fat ratios (below) were significantly different from the diploid controls. The hepatosomatic index and muscle ratio were not significantly different.



GROWTH EVALUATION OF *Clarias gariepinus* FED ENZYME TREATED PIG HOOF MEAL BASED DIET

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Pig Hoof Meal (PHM) is an inedible slaughter waste rich in keratin protein and its usage as feed ingredient feedstuff is limited due to processing method needed to break its strong helical cysteine disulphide bond. The advent of enzyme biotechnology in material utilisation has made keratin product to be utilised in livestock and aquaculture nutrition with variable results. This research was conducted to determine the inclusion level of PHM in diet of catfish fingerlings.

Feeding trial was conducted for 8 weeks with catfish total initial weight 1.79g stocked at the rate of 30 fish/plastic tank in replicate. PHM processed with enzyme Valkerase® (CibenzaIND900) was used as protein source in dietary feed at 40% crude protein at 25%, 50%, 75% and 100% inclusion of PHM with fishmeal as control diet (Table 1).

At the end of experiment (Fig.1) catfish fed diet without fishmeal (100%) had significantly ($P<0.05$) lower final weight (FW)/ mean weight gain (MWG), percentage weight gain (PWG) and specific growth rate (SGR) with highest feed conversion ratio (FCR) compared to all other dietary treatments (Table 2). Result showed that enzyme treated PHM can be used for optimal fish growth between 25%-75% without implication on fish survival.

Table 1: Nutrient composition of PHM diets.

Ingredients	0%	25%	50%	75%	100%
Fish meal	23.49	18.9	14.33	9.75	-----
Pig hoof meal	-----	4.58	9.16	13.74	23.49
Soya bean meal	23.49	23.49	23.49	23.49	23.49
Groundnut cake	23.49	23.49	23.49	23.49	23.49
Maize	26.53	26.53	26.53	26.53	26.53
Bone	0.5	0.5	0.5	0.5	0.5
Salt	1.0	1.0	1.0	01.0	1.0
Premix	1.0	1.0	1.0	1.0	1.0
Limestone	0.5	0.5	0.5	0.5	0.5

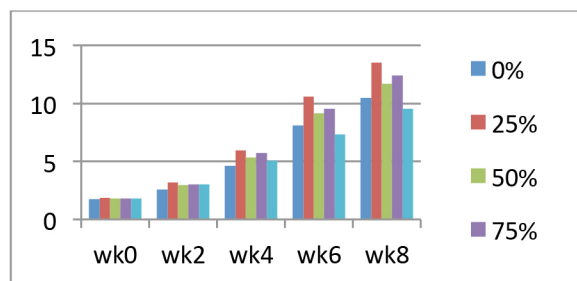


Figure 1: Weight gain of *Clarias gariepinus* fed PHM.

Table 2: Growth of *Clarias gariepinus* fed PHM diets. Row means with different superscripts are significantly different ($P<0.05$).

Parameters	0%	25%	50%	75%	100%
IW (g)	1.74 ^a	1.85 ^a	1.78 ^a	1.78 ^a	1.81 ^a
FW (g)	10.49 ^c	13.49 ^a	11.69 ^b	12.42 ^{ab}	9.54 ^c
MWG (g)	8.75 ^c	11.65 ^a	9.91 ^b	10.64 ^{ab}	7.73 ^c
PWG (%)	636.01 ^{bc}	759.35 ^a	704.14 ^{ab}	734.17 ^{ab}	618.89 ^c
Survival (%)	83.33 ^a	81.11 ^a	78.89 ^a	88.89 ^a	81.11 ^a
SGR%/g/day	2.34 ^{cd}	2.59 ^a	2.45 ^{bc}	2.51 ^{ab}	2.24 ^d
FCR	1.27 ^{ab}	1.21 ^a	1.26 ^{ab}	1.23 ^a	1.48 ^b

GROWTH PERFORMANCE AND DIGESTIBILITY OF PROCESSED CATTLE HORN MEAL BASED DIET FED TO *Clarias gariepinus* JUVENILES

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The future of cultured fisheries is hinged on availability of quality fishmeal. The demand for fishmeal has escalated the price in domestic and international market. This necessitated the need to find alternative rich source of protein with complimentary amino acid profile that compares favourably with fishmeal. Keratin sources are adjudged to be of importance due to their nutritional composition. African cattle breeds possess long horns, constituting waste at slaughter houses needed to be valorised for production of protein ingredient. Cattle horn keratin (Chk) has found limited uses as organic fertilizer despite its abundance from slaughtered cattle at abattoirs worldwide. Valorisation of Chk was conducted and its digestibility as animal protein source assessed on African catfish juvenile for subsequent inclusion in diet on experimental basis.

Clarias gariepinus juveniles of 14.82g average weight stocked at 15 fish /tank/replicate were fed 40% protein reference (70%) and test (30%) diet at 3% body weight for 8 weeks. Test diets were incorporated with treated horn of: soda ash (Chk2), potash (Chk5), fermented (Chk8), autoclaved (Chk11) and raw horn (Chk14) with chromic oxide as inert biomarker (Table 1). Faecal collection was carried out by siphoning after 8 hours of feed administration.

At the end of the experiment growth performance of fish fed Chk2 and Chk14 showed improvement among the treatments, while reference diet was significantly different ($P<0.05$) in performance. Apparent digestibility of reference diet > Chk2 > Chk14 > Chk11 > Chk8 > Chk5; while nutrient digestibility was > Chk2 > Chk14 > Chk11 > Chk8 with least in Chk5 (Table 2).

The result indicated that at 30% inclusion of processed Chk, digestibility and weigh gain of *C. gariepinus* is achievable with Chk2 and Chk14.

Table 1: Composition of diets fed to *C. gariepinus* juveniles

Ingredient	Reference diet 70%	Test diet 30%
Fish meal	23.07	16.15
Soya bean meal	23.07	16.15
Groundnut cake	23.07	16.15
Maize	29.19	20.43
Vitamin mineral premix	0.6	0.42
Chromic oxide	1.0	1.0
Test ingredient	0.0	29.70

Table 2: Growth performance, apparent and nutrient digestibility of *C. gariepinus* fed cattle horn diets.

Parameters	R. diet	Chk2	Chk5	Chk8	Chk11	Chk14
IW (g)	13.59 ^a	15.64 ^a	14.29 ^a	14.86 ^a	15.01 ^a	15.54 ^a
FW (g)	43.82 ^a	33.76 ^b	26.57 ^b	27.27 ^b	29.55 ^b	31.79 ^b
MWG (g)	30.22 ^a	18.12 ^b	12.27 ^c	12.42 ^c	14.55 ^{bc}	16.26 ^{bc}
PWG (%)	68.95 ^a	53.67 ^b	45.51 ^{bc}	44.34 ^c	49.04 ^{bc}	51.15 ^{bc}
SGR%/g/day	1.62 ^a	1.51 ^b	1.39 ^b	1.41 ^b	1.45 ^b	1.48 ^b
FCR	3.22 ^a	4.45 ^b	5.74 ^{bc}	6.54 ^d	5.29 ^{bc}	4.88 ^{bc}
<u>Apparent digestibility</u>						
Protein	88.26 ^a	82.06 ^b	43.59 ^f	62.19 ^e	72.12 ^d	74.46 ^c
Fat	97.91 ^a	88.90 ^b	86.43 ^b	89.32 ^b	79.22 ^c	82.29 ^c
Dry matter	90.94 ^a	81.92 ^b	46.86 ^c	64.29 ^d	65.69 ^c	67.26 ^c
Energy	90.66 ^a	81.05 ^b	47.22 ^f	68.37 ^e	69.10 ^d	71.28 ^c
<u>Nutrient digestibility</u>						
Protein		71.11 ^a	-34.59 ^e	16.37 ^d	45.36 ^c	51.44 ^b
Fat		72.95 ^{ab}	66.31 ^b	74.22 ^a	48.11 ^d	56.22 ^c
Dry matter		61.36 ^a	-35.15 ^e	22.64 ^d	28.01 ^c	36.01 ^b
Energy		64.04 ^a	-28.85 ^e	29.16 ^d	33.23 ^c	38.95 ^b

Row means with different superscripts are significantly different ($P<0.05$).

AQUACULTURE CANADA AND WAS NORTH AMERICA 2020 SUBMISSION

Takeda

Proposal for Session “Material Management, Processing and Utilization”

Urchinomics B.V. would like to propose a 20-minute presentation about the utilization of overgrazing, empty sea urchins as an input, and turning them into premium seafood using aquaculture techniques.

Suggest Contents

Below are the topics I believe I can cover in the talk:

- Problem Statement
 - Overfishing, climate change and pollution has set the stage for sea urchins to explode in population, overgrazing entire kelp forests in Canada, USA, Japan, Norway and Australia (to name a few)
 - After having eaten all the kelp around them, the urchins become empty and valueless to fishers and predators, resulting in desert-like barren conditions to persist for decades or centuries
- Solution
 - Removing urchins and reducing grazing pressure will bring kelp forests back
 - Restored kelp forests contribute to improved marine biomass, marine biodiversity, carbon binding and sequestration, absorption of nitrogen and phosphorus run off, protect from wave related erosion and ocean acidification.
 - Removed urchins can also then be ranched using aquaculture techniques to turn into premium, exportable seafood (uni)
- Methodology
 - Fishing, ranching, and exporting
 - Drill down on the ranching part to show how the combination of aquaculture systems, feed and well handled urchins make the business case
- Results so far
 - Show kelp restoration in practice in California and Northern Norway
 - Highlight results from our pilot ranching operations in Japan, USA, Norway and Canada
 - Highlight efforts in Newfoundland, New Brunswick and Quebec
 - Opportunities and Challenges
 - Highlight what makes this kind of utilization model possible in some parts of the world, and less so in others

Additional Angle

- Perhaps we can also arrange a tasting event of our ranched, Canadian urchins at some point in time during the conference.
- Perhaps we can also use this event as an opportunity to announce the first shipment of commercially ranched urchins from Newfoundland to NYC.

SEMEN CHARACTERISTICS AND OPTIMIZATION OF SPERM CRYOPRESERVATION PROTOCOL OF *Labeo gonius* (Hamilton, 1822)

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The study describe the semen characteristics and experimental results to determine suitable protocol for sperm cryopreservation of minor carp, *Labeo gonius* (Kuria Labeo), which can be used in seed production. The mean pH of seminal plasma was 7.6 ± 0.19 , mean osmolality of milt was 276 ± 15 mOsmol kg^{-1} and sperm count in milt samples from different males ($n=9$) were found in the range of 25.4×10^9 to 34.2×10^9 cells ml^{-1} . To determine the suitable extenders for cryopreservation a total of 9 different extenders were screened. Finally, three extenders (8, 9B and 9C) having mean sperm activation with water above 70% and mean spermatozoa survival above time (40 seconds) were selected for cryopreservation work. Milt to diluent ratio of 1:6 was kept for cryopreservation work. DMSO (10% v/v) was added in extenders as cryoprotectant to defend from stress on cells during cooling and freezing and diluted milt was then filled in 0.25 ml capacity straw and stored in liquid nitrogen after 20 minutes equilibration time. The feasibility of selected extender and cryoprotectant combinations was assessed by the ability of cryopreserved sperm to fertilize the *Labeo gonius* eggs and hatching percentage. Fertilized eggs were incubated in flow-through incubation system with a water flow rate of 2-3 litre per minute. Recorded hatching rates and survival rates in extender 8, 9B and 9C were $61.1 \pm 0.23\%$, $81.3 \pm 0.19\%$, $83.7 \pm 2.7\%$ and $44.6 \pm 1.2\%$, $45.0 \pm 2.64\%$ and $46.7 \pm 1.52\%$, respectively. Control group (having fertilized from fresh sperms) yielded $84.99 \pm 0.88\%$ hatching rate and $48.6 \pm 1.52\%$ survival rate. Statistical analysis indicated that the extender 9C and 9B with 10% DMSO gave higher hatching and survival rate in comparison to extender 8 which might be due to the presence of sugar in 9C and 9B. The fertilization capacity of cryopreserved sperms and control group (having fresh milt) were compared and significant difference was not found in terms of hatching rate ($p \geq 0.05$) as well as for survival rate ($p \geq 0.05$) with control except for extender 8 which was even higher than 50 percent. Therefore, extender 8, 9B and 9C were recommended for cryopreservation of milt of *L. gonius* which would be helpful in *ex-situ* conservation of the species.

EFFECTS OF ANTARCTIC KRILL PRODUCTS ON FEED INTAKE, GROWTH PERFORMANCE, FILLET QUALITY AND HEALTH IN SALMONIDS

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The interest for krill-based ingredients for aquaculture feed applications has increased steadily in recent years. For decades there has been a heavy reliance on the limited sources of fishmeal and fish oil in the salmonid aquaculture industry. Further growth in farming of carnivorous fish is dependent on new feed resources becoming available. The only unexploited marine resources of significant biomass are found at lower trophic levels, of which the Antarctic krill has a high potential. Antarctic krill (*Euphausia superba*) is one of the most abundant species on earth, with an estimated biomass of around 500 million tonnes. Krill plays a key role in the marine food chain in the Antarctic Ocean, and hence krill harvesting is highly regulated by the Commission for the Conservation of Antarctic Marine Resources (CCAMLR). These strict regulations of Antarctic krill harvesting have led to an increase in its biomass over the years, from 60.3 million tonnes measured in 2000 to 62.6 million in 2018/19 according to the findings from CCAMLR.

Among the several products produced from Antarctic krill for the aquaculture feed formulations, krill meal is the most vastly used. Krill meal is a sustainable source of protein, n-3 phospholipids, feed attractants (nucleotides, free amino acids, TMAO) and astaxanthin. It has a proximate composition that is similar to fish meal with an almost identical amino acid profile. The lipid fraction of krill meal contains a high proportion of polar lipids, as well as a high content of polyunsaturated fatty acids (PUFAs), sterols (mainly cholesterol) and astaxanthin esters. The present article provides an overview on the documented benefits of krill meal, focusing on feed intake, growth performance, fillet quality, slaughter yield, and health benefits in terms of reducing fat accumulation and inflammation in liver and intestinal tissues in salmonids.

GUT MICROBIOTA IN ATLANTIC SALMON (*Salmo salar*) AT DIFFERENT DEVELOPMENTAL STAGES

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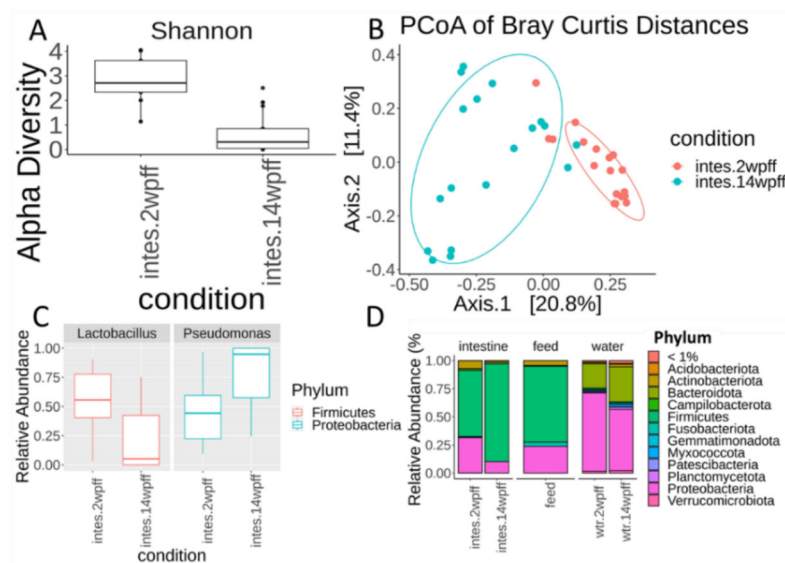
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Understanding the interplay between gut microbiota and gut function in farmed fish is important for designing the aquaculture feeds that maximise fish health and performance from first feeding to commercial size. 16S rRNA V3-V4 amplicon sequencing was utilised to investigate the microbial communities in the gut of Atlantic salmon at two weeks and 14 weeks post-first feeding (wpff). Fish were raised in triplicate tanks and fed marine-rich diet from first feeding to the end of experiment. Intestinal samples were collected at two and 14 wpff from 6 fish per tank (36 samples in total). Water and feed samples were also collected. Sequences were analysed using DADA2 to identify the microbial community as Amplicon Sequence Variants (ASVs). Richness and diversity of gut microbiota significantly decreased across fish development (Chao1 $P < 0.05$; Shannon $P < 0.0001$) (Figure 1A). Beta-diversity analysis showed clear separation between fish developmental stages (Figure 1B). The dominant bacterial taxa changed significantly with time (Figure 1C, D). Core microbes were detected at $\geq 0.1\%$ in 80% of the fish at each developmental stage. At two wpff, 14 core ASVs were identified compared to a single ASV (unclassified genus of Family Ruminococcaceae) at 14 wpff. The fish gut microbiota was impacted by the environmental conditions (e.g., Firmicutes from feed and Proteobacteria from water) and potentially by changes in selectivity in the fish during development. These findings are important for understanding the interaction between fish gut and gut microbiota in the developmental context.

The work was funded by BBSRC grant NUTRIPROG (BB/R018812/1) and MT was funded by Newton-Mosharafa PhD Scholarship.



FULL-FILLING THE PROMISE OF SABLEFISH *Anoplopoma fimbria* GENOMICS: SEX-TESTING, PARENTAGE ANALYSES, WHOLE-GENOME SEQUENCING AND GENOME PREDICTION

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At the Golden Eagle Sablefish (GES) hatchery on Saltspring Island (British Columbia) eggs and sperm from wild-caught and hatchery-reared captive brood stock are used to generate offspring that spend approximately 1 year in the hatchery and 1 year in ocean pens at Kyuquot Sound (Vancouver Island). Embryo- and larval-stage mortality is highly variable and beyond 2 months of age growth rate varies with fish ranging from <2 kg to almost 5 kg at the time of processing. Efforts to support slow-growers by separating them from larger fish and enhancing their diet have been largely unsuccessful, thus a proportion of the population (referred to as ‘drop-outs’ by GES staff) fails to convert food into flesh after reaching about 1.8 kgs.

The research discussed here is a collaboration involving the University of Victoria, GES, and the KA:’YU:’K’T’H’ and CHE:K:TLES7ET’H’ First Nations. We have developed, or are currently developing, molecular tools for sexing sablefish, for parentage analyses, and for Genome-Wide Association Studies and Genome Prediction in an effort to assess the influence of genetics on the phenotypic variation briefly described above.

Genetic and genomic data have played a major role in the improvement of long-domesticated species (e.g., dairy cattle) and in the development of new domestic lineages (e.g., Atlantic salmon). In the case of sablefish, these new tools will allow us to i) test the hypothesis that males make up a large proportion of the ‘drop-out’ population, ii) determine whether or not a new ‘neomale’ breeding program is producing all-female broods, iii) identify crosses that contribute disproportionately to fast or slow growers, iv) enhance the deployment of the largely wild-caught brood stock population, and v) recruit brood stock from the captive population that are more-likely to produce progeny with high survival, growth rate, and health metrics. Our long-term goal is to develop an exceptional domestic population. Progress made during the first six months of this collaboration will be presented.



OPTIMISATION OF EGG TO TABLE GROW OUT of Atlantic salmon IN A FULL FRESHWATER RECIRCULATION AQUACULTURE SYSTEM

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RAS (Recirculating Aquaculture System) technology is a proven solution for providing sustainable, high-quality protein to meet the increasing demand of the continuously growing population. This presentation discusses how significant advances in growth performance (Fig 1.) from egg to 4.5kg market size Atlantic salmon (*Salmo salar*) have been achieved at both technological and biological level in an entirely freshwater RAS technology over the last 5 years.

Innovative design of AquaMaof RAS facility has developed a tailor-made solids settler, a unique biofilter, oxygen-dissolving systems with low head requirements, a depuration capability, and a built-in denitrification system that enables the recycling of water and significantly reduces water consumption. This technology has significantly improved carbon dioxide stripping efficiency and elimination of hydrogen sulphide to support optimal growth and welfare. Further, through optimised adjustments in water chemistry and thermal regimes, in conjunction with the application of enhanced nutrition in each production phase, we have reduced operational grow-out by 7-8 weeks. At present we have achieved a head on gutted weight of 4.2kg within 19 months of first feeding, whilst maintaining high survival and welfare standards, providing a high-quality market-size salmon utilising a sustainable aquaculture production system.

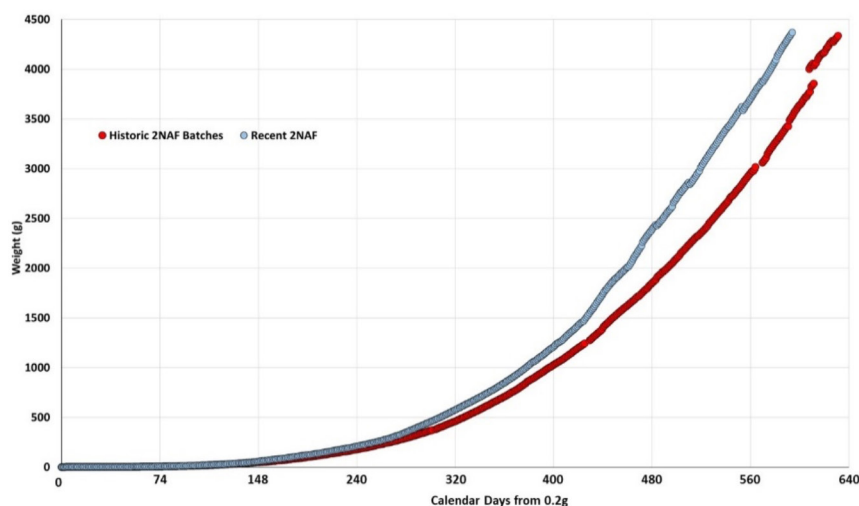


Figure 1. Advances in egg to table growth performance of current (blue) and historic (red) diploid all-female salmon.

WHY AND HOW AQUARIUMS AND FISHBASE CAN WORK TOGETHER TO PROMOTE FISH KNOWLEDGE AND CONSERVATION? ASSESSEMENT OF A FOUR-YEAR PROJECT

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Aquariums have evolved over the last decades from exhibiting animals primarily for public enjoyment to conservation organizations whose missions are to inspire and contribute significantly to wildlife conservation. In France, the Association of Aquarium Curators (UCA) groups today 29 active members working in 26 aquariums. Despite significant implications in many conservation and research programmes, UCA's aquarium contributions to science remains low. One possible solution to highlight the contribution of UCA members to the knowledge and conservation of fish could be to develop a strong and sustainable partnership with FishBase. The idea is to upload the data generated by UCA aquariums directly on FishBase so that they would be freely available on-line (Figure 1).

The main goal of this article is to describe how this collaboration was developed in the past four years. Based on a survey conducted in 2019, 18 curators agreed to spend some time on this project and shared their list of species and pictures. Fifteen agreed to appoint a FishBase representative, whose main missions would be to collect and curate data in each aquarium before sending them to FishBase. We also found that UCA members have successfully bred over 100 species. Overall, 851 species are hosted within the seven aquariums for which we obtained the list of species. This study also highlighted that less than 10% of species hosted in aquariums are well known in FishBase. The increasing number of species maintained and reproduced in aquariums allows new opportunities for fruitful research collaborations and contributions of a wide array of disciplines. First and foremost, UCA aquariums have the potential to become “conservations centers” where real conservation and research work is done. Beyond conservation biology, aquariums could contribute to basic and applied research on wildlife husbandry, nutrition, disease, physiology, and reproduction. Data generated from aquarium could also generate large-scale comparison of life stage traits that would be otherwise impossible to perform. The sustainable development of this new partnership will rely chiefly on the implication of the FishBase representatives, associated with a dedicated research focus and adequate institutional support through research funding and staffing, and dedicated personnel in FishBase. To be successful in the long term, this new partnership will require also strengthening collaborations among aquariums, as well as collaborations between aquariums and academia, and hobbyists. It is anticipated that new aquariums could join this project.

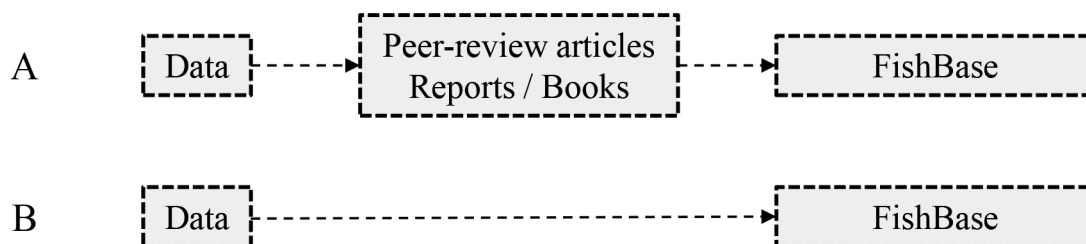


Figure 1. Classical path for getting data published and then available on-line on FishBase (A) and alternative way proposed in the present project (B).

FISH DOMESTICATION AND GENETICS: A BRIEF OVERVIEW

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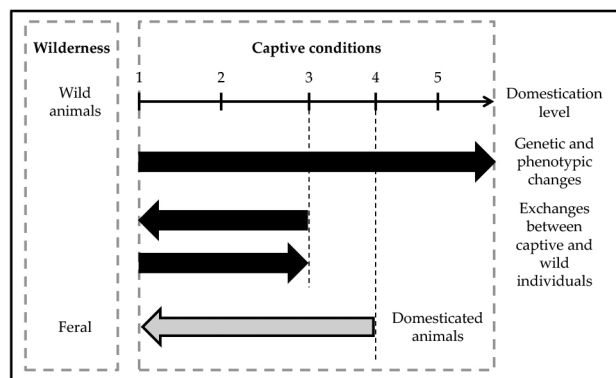
During the domestication of fish, the animals will gradually diverge from their wild ancestors, so that they will no longer be considered captive wild animals, or even tame, but domesticated. Nevertheless, it is important to emphasize that the distinction between wild and domesticated remains elusive and one should not consider that wild/domestic represents a dichotomy such as black/white or true/false. On the contrary, there is a continuum between these two extremes, and considering a species as domesticated is somewhat arbitrary and questionable for fishes in which domestication is relatively recent, except for a handful of species.

To get around this problem, we proposed the concept of domestication levels, including five successive levels (Figure 1).

Figure 1. Evolution of a wild fish species throughout the process of domestication. As soon as wild animals are transferred to captivity (level 1), the process starts. The numbers correspond to the domestication levels: initial rearing trials (level 1), control of a portion of the life cycle in captivity (level 2), control of the entire life cycle in captivity (level 3), control of the life cycle without input from the wild (level 4), and implementation of breeding programs (level 5). During domestication, wild animals will evolve both genetically and phenotypically, particularly when exchanges with wild congeners do no longer exist (level 4). Domesticated animals can return to the wild and are then known as feral.

Once species have reached level 4, breeding programs (level 5) can be implemented. Historically, these programs have focused on growth enhancement, then feed conversion efficiency, age of sexual maturity, improved resistance to bacterial and viral diseases, and a number of product quality-related traits have been progressively included. The considerable improvement in fish performance has been made possible by the tremendous progress in genetic selection techniques and concepts, which have been applied to an increasing number of species.

In conclusion, domestication is an ongoing process that adapts fish to both captive conditions and humans. New breeding programs will need to balance market and non-market values (fish welfare) while maintaining sufficient genetic variability to ensure that fish are productive and robust enough to cope with environmental changes. The sustainable future of aquaculture will depend firstly on the continuous improvement of already domesticated fish species and secondly on our willingness and ability to diversify farmed fish species, preferably indigenous ones, to promote a more resilient and balanced aquaculture production at the global level.



WHY AND HOW TO PROMOTE AQUAPONICS IN HIGH SCHOOL? ASSESSEMENT OF A FIVE-YEAR PROJECT.

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Within the framework of a multi-year agreement between the Territorial Council of Saint-Pierre and Miquelon (SPM) and the University of Lorraine (2018-2023), several innovative educational actions have been carried out with the central objective of introducing students to various soil-less production methods. Indeed, given the climate and soil conditions of the archipelago, these methods could represent a real opportunity to significantly increase local agricultural production.

Two projects were conducted in SPM: the first at the Emile Letournel high school in Saint-Pierre for 3 years (2018-2021), and the second at the Ecole du Socle in Miquelon for one year (2021), as well as in Nancy (Figures 1,2).

Thanks to a European project (<http://perciponie.eu/>), five new schools in Lorraine get involved this year (Figure 3).

In conclusion, more than 30 students from the IUT Nancy-Brabois (University of Lorraine) were involved to help about 20 teachers to develop new activities or solve technical problems. In total, 300 students from the archipelago and metropolitan France, from kindergarten to high school, have been made aware of these new production methods. At the same time, this project has allowed the development of a real network of schools between the archipelago and metropolitan France, as well as Canada. This project will be pursued at least during the next three years.

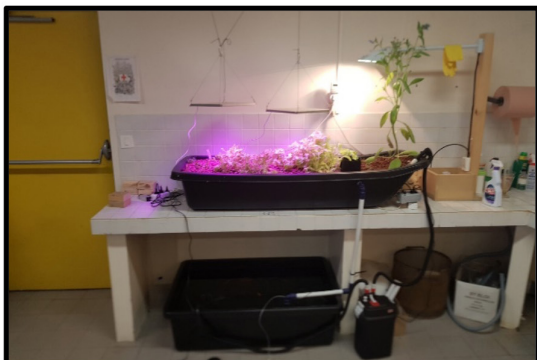


Figure 1. Example of a small aquaponic system set in the high school in Saint-Pierre (SPM).



Figure 3. Example of a small aquaponic system set in one school in Lorraine.



Figure 2. Aquaponic system built by students and set in the Botanical garden in Nancy.

DESCRIPTION OF PATTERNS OF INFECTIOUS SALMON ANAEMIA VIRUS OUTBREAKS IN MARINE SALMON FARMS IN NEWFOUNDLAND AND LABRADOR

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Since its first detection in 1996, infectious salmon anemia has been a viral disease of significant concern for farmed salmon in the Atlantic provinces of Canada. Several epidemiological aspects of ISA outbreaks in Atlantic Canada remain incompletely understood. The objectives of this study were to describe the spatio-temporal patterns of infectious salmon anemia virus (ISAv) outbreaks during marine phase of production of farmed Atlantic salmon in Newfoundland and Labrador (NL) and explore risk factors associated with the timing of outbreak detection and depopulation at affected sites. ISAv surveillance data (2012- 2020) were provided by the Aquatic Animal Health Surveillance Program led by NL Department of Fisheries, Forestry, and Agriculture. Final analyses included fish and site-level data consisting of 94 sampling events and 20 Atlantic salmon production sites. ISAv detection was based on RT-PCR with genotyping for moribund and harvested fish samples. Linear regression models were used to investigate factors influencing time-to-detection (days from stocking to first ISAv detection) and time-to-depopulation (days from the first detection to site depopulation). During the study period, a total of 28 unique outbreaks were reported. Site-level annual incidence risk of ISAv detection was 3-33% and fish-level annual detection in sampled fish was 2-45%. European (EU) ISAv genotype detections were more common compared to North American (NA) genotypes. The variables, ISAv genotype, neighboring farm outbreaks, and year of the outbreak were not associated with the time to first ISAV detection. Outbreaks with NA genotypes had significantly shorter ($p=0.02$) time to depopulation compared to EU genotypes. Time to depopulation was also significantly shorter ($p=0.0004$) if ISAV was first detected at a site from 2016-2018, compared to 2012-2015. The annual risk of ISAv detection in marine salmon farming sites in NL varied over years at both fish and site levels. Time to depopulation was inversely associated with ISAV's North American genotype and the year of the first detection.

MOVING PUBLIC OPINION ON AQUACULTURE: THE MESSAGE MATTERS

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Expansion of the aquaculture industry hinges on the backing of local residents and other stakeholders capable of withholding necessary social, political, or financial support. Regional and individual variations in level of enthusiasm for marine aquaculture have prompted several public opinion surveys as a way of assessing overall support for, or resistance to, the expansion of aquaculture in the US. These studies generally find that although the majority of the public do not have strong objections to the sector, there is consistently a smaller, but vocal, group that opposes marine aquaculture. A second common finding is a broad lack of awareness of marine aquaculture in the US. Two important unanswered questions concern 1) how malleable public opinion about aquaculture is; and 2) what specific benefits of aquaculture in general and seaweed farming in particular most influence support for the industry.

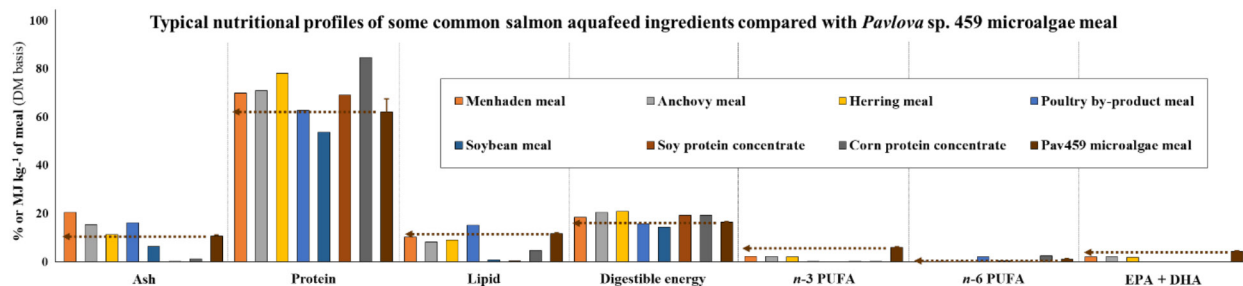
In 2021, we conducted a deep dive synthesis into two surveys conducted as part of a series of aquaculture perceptions projects managed by the Aquarium of the Pacific's Seafood for the Future program in Long Beach, California to answer some of these questions. The synthesis was part of a broader portfolio of perceptions work funded by World Wildlife Fund and USC Sea Grant. We will present some of the highlights of this portfolio, with a focus on the findings from our synthesis. Some key findings we will share include a before-and-after assessment of how different thematic messages (i.e., environmental, economic, social) resonated across survey participants of different sociodemographic attributes (i.e., income, education, ethnicity, state of residency). We will also speak to how baseline familiarity with marine aquaculture affects malleability of participant opinion about the sector. Altogether, this work provides critical insight to aquaculture literacy efforts which aim to equip community members with the knowledge needed to engage in decision making processes for marine aquaculture development in their 'ocean neighborhoods'.

NUTRITIONAL EVALUATION OF A PROMISING MARINE PRYMNESIOPHYTE MICROALGAE *Pavlova* sp. CCMP459 GROWN IN PHOTOBIOREACTORS AS A POTENTIAL LOW-TROPHIC FEED RESOURCE FOR ATLANTIC SALMON *Salmo salar* L.

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The aquaculture industry, consumers of farmed salmon products, and our environment would all benefit from greater access to more sustainable feed inputs. Novel ingredients must have high nutritional quality, be safe for consumption, and must be capable of cost-effective sustainable production at large scale. Ideal candidates should come from low-trophic natural resources, require little post-harvest processing, should not compete as human food, and have the ability to restore *n*-3:*n*-6 fatty acid ratios (particularly essential *n*-3 LC-PUFAs) in aquafeeds and final consumer products. Several *Pavlova* sp. CCMP459 microalgae meals (*Pav*459) produced in land-based photobioreactors were similar to good quality fish meals; with high crude protein (~60%), crude lipid (~12%) and gross energy (~22 MJ kg⁻¹) with low ash (~10%) and carbohydrate (~13%). Unlike many current fish meal alternatives, they contained high *n*-3 PUFA (~6%); comprised mainly of the essential *n*-3 LC-PUFAs EPA (~3%) and DHA (~1%). As this was concomitant with low *n*-6 PUFA (~1%), they had high *n*-3:*n*-6 fatty acid ratios (~5); which far exceed terrestrial ingredients commonly used in today's modern aquafeeds (e.g., less than 2). *Pav*459 meals had excellent essential amino acid (EAA) profiles; demonstrated by high EAA:non-EAA ratios (~1) and EAA indices (>0.9) relative to egg albumin, premium fish meal and soy protein. *Pav*459 meals were rich in essential minerals, trace elements and carotenoids; with negligible contaminating heavy metals or antinutritional factors (ANFs). *In vitro* 2-phase gastric/pancreatic protein digestion was high (82%) for *Pav*459 meals; irrespective of cell-rupture which indicates a cell wall of low recalcitrance and little requirement for costly and energy-intensive downstream processing. An *in vivo* substitution digestion assay with juvenile (~25 g) Atlantic salmon demonstrated high digestibility of nutrients in *Pav*459 meals for protein (83%), lipid (91%), carbohydrate (72%), energy (71%), EAAs (92 to 97%), SFA+MUFA (76 to 80%), PUFA (97%), EPA+DHA (99%) and high DIAAS values for all EAAs (1 to 4). A feeding study with larger (~170 g) Atlantic salmon demonstrated that *Pav*459 meal can be included at 20% of the diet (in partial or total displacement of fish meal and fish oil) with little effects on feed intake, growth performance or nutrient utilization of the fish over a 12-week feeding period. In addition, there were no significant differences observed in total crude protein (74 to 77%), total crude lipid (63 to 70 mg g⁻¹) or total EPA+DHA levels (5 to 6 mg g⁻¹) in the fillets of salmon fed diets containing up to 20% *Pav*459 meal; concomitant with greatly reduced or fully displaced dietary fish meal and fish oil.



UTILITY OF RESCIRCULATING AQUACULTURE ORGANIC WASTE FOR INTENSIFICATION OF BOREAL AGRICULTURE ON PODZOLIC SOILS, AN OPPORTUNITY FOR SUSTAINABLE NUTRIENT MANAGEMENT

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Simultaneous expansion of aquaculture and agriculture in Newfoundland and Labrador (NL) creates opportunities for integrated waste stream utilization. Agriculture expansion in NL involves the land use conversion (LUC) of boreal podzols. Podzols offer a distinct layering of chemical properties with agronomic potential decreasing with soil depth. Utilizing aquaculture organic waste could be an effective way of capturing resources while enhancing agriculture activity on boreal podzols. To explore the agricultural use of recirculating aquaculture system (RAS) organic waste a study was conducted at the Centre for Agriculture and Forestry Development. The study was a 12 week greenhouse pot experiment in a completely randomized block design. Treatment factors included organic and mineral fertiliser application to topsoil (0-15cm) and subsoil (15-30cm) collected from a white spruce (*Pinus strobus*) plantation. The utility of RAS organic solids and supernatant were compared against dairy manure and also against mineral fertilizer as positive controls. Tall fescue (*Festuca arundinacea*) a regionally common forage was the test crop and was fertilized at rates equivalent to 200 kg/ha of N and 110 kg/ha of P. Agronomic performance was measured as growth rate, biomass accumulation, and nutrient acquisition (Table 1). While initial growth rates were accelerated for the subsoil treatments they were not sustained through the length of the experiment, a behaviour distinct from the topsoil. Topsoil treatments where slower but consistent growth was maintained for the entire experiment. This suggests variable nutrient retention and availability between soil depths. Final yields for the same fertilizers where generally comparable between soil layers except for the RAS treatments. The distinctly higher productivity with RAS in the topsoil was not matched in the subsoil. Furthermore, RAS amended subsoils led to a reduced plant P acquisition, an indication of the subsoils' capacity to retain P. This highlights the impact that the interaction between soil layer and fertilizer type had on the nutrient availability profiles. When aware of the nature of the soil, organic waste from RAS could be a sustainable nutrient source in the agriculture sector.

Table 1. Mean growth response parameters of tall fescue in two boreal podzol depths treated with RAS and dairy organic waste, along with mineral fertilization.

Treatment [#]	Dry matter yield (Mg Ha ⁻¹)			Height (cm)	Leaf width (mm)	Acquisition % ^{##}	
	Shoot	Root	Shoot:Root ratio			N	P
<i>Topsoil</i> _{NEG}	1.53 d	2.26 a	0.69 a	18.64 c	1.00 e	68 c	45 a,b
<i>Topsoil</i> _{POS}	3.28 c	3.57 a	1.02 a	34.96 b	3.47 b	55 c	17c
<i>Topsoil</i> _{DMN}	4.87 b	8.72 a	0.86 a	35.52 a,b	3.60 b	87 c	31 b,c
<i>Topsoil</i> _{RAS}	7.13 a	6.28 a	1.52 a	38.80 a	4.93 a	164 b	58 a
<i>Topsoil</i> _{SUP}	4.81 b	4.64 a	1.20 a	34.76 b	2.80 c,d,e	56 c	21 c
<i>Subsoil</i> _{NEG}	0.96 d	1.53 a	0.72a	16.36 c	1.00 e	244 a	29 b,c
<i>Subsoil</i> _{POS}	4.70 b,c	4.87 a	1.20 a	36.16 a,b	3.33 b,c	78 c	24 b,c
<i>Subsoil</i> _{DMN}	5.66 b	7.42 a	1.08 a	35.28 a,b	3.46 b	78 c	20 c
<i>Subsoil</i> _{RAS}	4.70 b,c	5.32 a	1.40 a	33.00 b	1.86 d,e	77 c	17 c
<i>Subsoil</i> _{SUP}	5.10 b	4.08 a	1.46 a	35.48 a,b	2.86 b,c,d	71 c	21 c

In each column, means followed by same letter indicate that they are statistically similar

[#] NEG (negative control), POS (mineral fertilizer), DMN (dairy manure), RAS (RAS organic solids), SUP (RAS waste supernatant)

^{##} Acquisition % calculated as nutrient present in plant tissue/initial available nutrient provided to soils

SUPPORTING AQUACULTURE THROUGH COASTAL OCEAN MONITORING AT BIOLOGICALLY RELEVANT SCALES

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Coastal ocean monitoring is critical for informing aquaculture management decisions such as, site selection, engineering specifications, and carrying capacity. Dedicated nearshore monitoring is required to measure timeseries of complex coastal dynamics at multiple depths, which cannot be captured by broad-scale or offshore monitoring. Long-term data sets are also becoming increasingly valuable to prepare for and understand climate change influenced events such as low oxygen occurrences at net pens.

The Centre for Marine Applied Research (CMAR) in Nova Scotia, Canada coordinates an extensive Coastal Monitoring Program to address this fundamental data need. CMAR maintains a network of nearly 70 oceanographic moorings that measure Essential Ocean Variables (e.g., temperature, dissolved oxygen, salinity, sea state, currents), typically within 1 km of Nova Scotia's coast.

To our knowledge, the resulting oceanographic data products are unique in Canadian coastal waters for their static depth profiles, time series length, and high spatial and temporal resolution. CMAR ensures these data are freely available on several platforms and in multiple formats (e.g., summary reports, processed data) to ensure maximum access. Aquaculture and coastal stakeholders are now using this coastal data for diverse applications. Implications and future plans for CMAR's Coastal Monitoring Program are discussed.

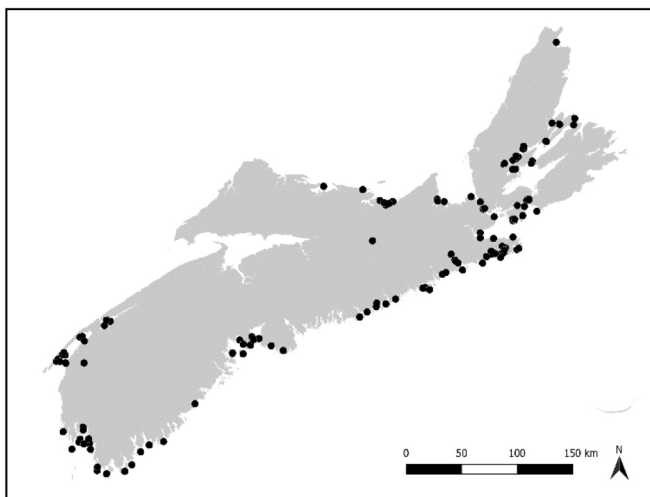


Figure 1: Deployment locations for autonomous sensors collecting temperature, dissolved oxygen, and/or salinity data.

AQUACULTURE RESEARCH INSTITUTE'S AQUACULTURE WORKFORCE DEVELOPMENT PROGRAMMING

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The Aquaculture Research Institute (ARI) at the University of Maine has developed several new aquaculture workforce development programs with the core objective to promote careers in the aquaculture industry to workers of all types (i.e. professional development and degree seeking students). UMaine is offering a new micro-credentialing program designed to take a learner from foundational knowledge and rigorous training, to application in a real-world work-setting. ARI in collaboration with Cooperative Extension developed an aquaculture micro-credentialing pathway for youth and adults. This pathway includes ARI's new hands-on skills development courses in Aquatic Animal Husbandry, Aquatic Animal Health, Recirculating Aquaculture Systems, and more. These hybrid courses allow learners to gain foundational knowledge online that will be implemented with further skills development during the week-long laboratory sessions. Skills gained in these courses will include skills sets identified in Aquaculture Occupational Competencies approved by the Maine Aquaculture Association. Laboratory sessions take full advantage of the aquaculture facilities across UMS giving students experience with industry sized facilities and cutting-edge research. ARI also offers an Industry Partnered Internship Program allowing learners to demonstrate and reinforce their skills within an aquaculture setting. This internship program matches students with industry hosts allowing students to gain experience in the aquaculture industry while conducting projects/research lead by the industry partner.

***Piscine orthoreovirus* SUBGROUP 1 (PRV-1) IN WILD SALMON IN THE NORTHEAST PACIFIC**

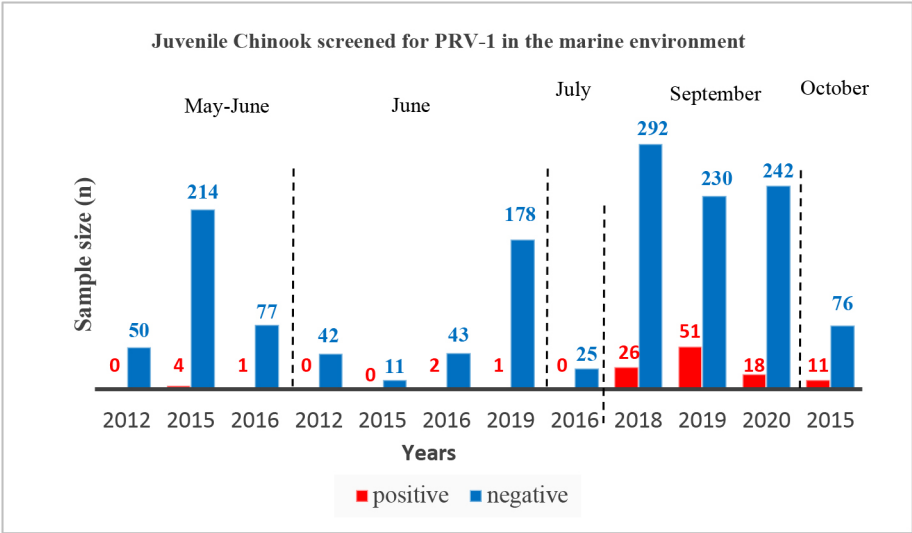
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Piscine orthoreovirus (PRV-1) is a segmented RNA virus, which is commonly found in wild and farmed salmonids in the Atlantic and Pacific Oceans. Although PRV-1 has been found in all species of Pacific salmon, infections are most commonly reported for Chinook and Coho Salmon (Polinski et al. 2020). We are examining the prevalence and genomic diversity of PRV-1 in wild and farmed salmonids in the Northeast Pacific with the goal of using these data to examine virus transmission routes. As part of this work, we used a two-step RT-qPCR protocol for the L1 segment to conduct surveillance for PRV-1 in 4,031 wild and enhanced Pacific salmon collected in May through October of 2011-2020.

PRV-1 was not detected in freshwater Chinook (n = 263), Coho (n = 200) or Sockeye (n = 344) Salmon or in juvenile Chum (n = 135), Sockeye (n = 373) and Pink (n = 70) Salmon collected in marine waters (Strait of Georgia and Johnstone Strait) in May – June. The overall prevalence of PRV-1 was low in juvenile Coho (1.7%, n = 595) and Chinook (7.4%, n = 1594) Salmon from marine waters, and adults (Coho 3.3%, n = 215; Chinook 2.9%, n = 242) collected at spawning. In September 2018 and 2019 the majority of PRV-1 positive Chinook Salmon were collected in the Discovery Islands, an area with Atlantic Salmon farms, whereas in 2020 the majority of positive fish were collected in areas without farms.

In this presentation we will discuss why differences in the ecology and migratory behaviors between species of salmon, as well as differences in stock composition within species, need to be considered when interpreting the spatial and temporal distribution PRV-1 in wild juvenile salmon in the Northeast Pacific.



ASPECTS OF REPRODUCTIVE BIOLOGY OF THE RETICULATE KNIFE FISH *Papyrocranus afer* (GUNTHER, 1868) IN LEKKI LAGOON, NIGERIA

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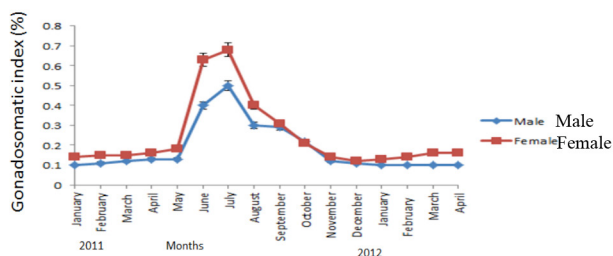
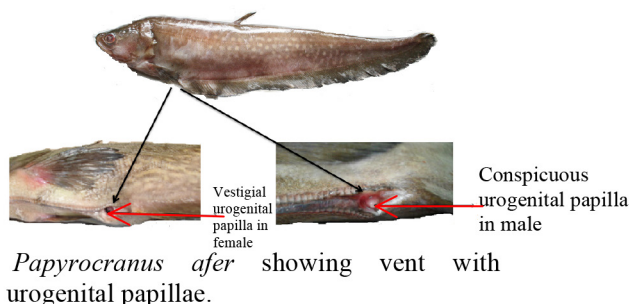
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Sizes at maturity (L_{m50}), fecundity, sex ratio and gonadosomatic indices (GSI) of the reticulate knife fish, *Papyrocranus afer*, collected from Lekki Lagoon, were investigated. A Total of 1154 specimens with standard lengths of 5.2-75.9 cm (mean = 34.86 ± 17.2 cm) and body weight of 7.9-1,958.8g (mean = 249.12 ± 28.56 g) were collected by means of artisanal passive and active gears (traps, long lines and nets) and examined. Sexes of fish specimen were determined macroscopically and microscopically after dissection. The length at which 50% of the fish population reached sexual maturity (L_{m50}) was considered as length at sexual maturity. Fecundity was determined by total counts of eggs; sex ratio by proportion of males to females, while GSI was determined as gonad weight expressed as percentage of total body weight.

Results showed that the most frequently caught fish size was 34.5cm; while sizes at maturity were 49.1cm (males) and 53.4cm (females). Matured specimens had characteristic urinogenital papillae prominent in males but vestigial in females.

Sex ratio (1: 0.6; Male: female) was significantly different ($X^2(1) = 32.21$, $p < 0.0001$). Fecundity was low (mean 49 ± 17 eggs for a fish of 52.86 ± 7.53 cm); it increased with fish size ($r = 0.71$). Higher GSI during rainy season with peak in July (female: 0.44 ± 0.14 %; male: 0.22 ± 0.01 %) indicated seasonal/annual spawning.

Low fecundity and annual spawning underlined the need for sustainable management of this species in Lekki Lagoon.



Monthly gonadosomatic indices of *Papyrocranus afer*.

LIVER DYSFUNCTION, STRESS AND HIGH MORTALITY IN ATLANTIC SALMON (*Salmo salar*) HELD AT 3°C

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Atlantic salmon reared in sea-cages in Atlantic Canada and Iceland experience temperatures as low as 1°C during the winter months, and such conditions have been associated with large-scale mortalities in both countries in recent years. However, most of our knowledge of this species' biology/physiology is from fish reared at warmer temperatures (10-25°C), and there is very limited data and information on the cause of these winter mortalities.

Commercially reared Atlantic salmon (~300 g) of St. John River (New Brunswick, Canada) origin were exposed to a gradual decrease in temperature from 8 to 3°C, and held at the latter temperature for 6 weeks. By the end of this period, mortalities had reached 25%, with moribund animals all displaying the same symptoms. These fish were lethargic and swimming vertically near the water's surface. They suffered from significant damage to/erosion of their snout and jaws, and there was often damage to the fins (caudal, pectoral, and anal) and body wall (mostly under the pectoral fins). Finally, these fish had a hepatosomatic index much greater (by ~ 2-fold), and pale livers, as compared to asymptomatic individuals.

To better understand the physiological changes associated with the development of this condition, blood (plasma) samples from symptomatic and asymptomatic fish were analyzed (n=9) for enzymes associated with tissue damage, as well as ion, metabolite and cortisol levels. The symptomatic fish exhibited increased plasma ion levels (Na⁺, Cl⁻) and osmolality that was likely due to the skin and fin erosion, and they possessed high circulating cortisol (~100 ng mL⁻¹) levels indicative of severe stress. Further, indices of tissue damage (plasma creatine kinase, aspartate aminotransferase and lactate dehydrogenase) were all elevated for the individuals in the later stages of the condition.

The enlarged pale liver, erratic swimming/lethargy, and changes in enzymatic biomarkers of tissue damage are very typical of what is observed in sea bream and yellow drum experiencing 'Winter Syndrome', which is associated with 'fatty liver disease'. Further, the dermal erosion and sores observed in our symptomatic fish were similar to that recently reported for salmon experiencing low temperatures (4-6°C) at cage-sites in Norway, and possibly related to *Tenacibaculum* sp. infection. Better understanding of the etiology of both of these pathologies associated with cold temperatures, and how to prevent them, should be a priority for the Atlantic salmon industry. This would allow for better health management and fish welfare in regions that experience prolonged low water temperatures.

AT WHAT TEMPERATURE DO CULTURED ATLANTIC SALMON (*Salmo salar*) EXPERIENCE COLD-INDUCED PHYSIOLOGICAL DISTURBANCES?

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Salmon reared in sea-cages in Atlantic Canada experience a large range of temperatures during grow out (0-19°C), and the extremes of this temperature range may be stressful for the fish. The effects of high temperatures on Atlantic salmon are being actively researched given the current, and predicted, effects of climate change. However, climate change is also resulting in more 'cold shocks', and there are sparse data on the effects of cold temperatures (< 2°C) on salmon physiology, health and welfare.

Commercially reared smolts (~175 g) of St. John River (New Brunswick, Canada) origin were separated into 8 tanks (2 groups x 4 tanks; 35 fish per tank). The first group was the control group, and was kept at 8°C. The second (experimental) group was exposed to a decline in temperature of 1°C week⁻¹ from 8 to 1°C, and then held at this temperature for an additional week (see Fig. 1). This temperature regimen mimicked the typical seasonal decrease in temperatures at sea-cage sites in Newfoundland. Photoperiod was 12h light: 12 h dark, and the fish were offered feed twice daily until apparent satiation. Plasma and liver samples were taken from 2 fish per tank after 1 week at 8, 6, 5, 4, 3, 2 and 1°C, and after 2 weeks at 1°C. Fish morphometrics (incl. HSI; hepatosomatic index) and feed consumption (FC) were measured over the course of the experiment, and blood (plasma) samples were analyzed for enzymes associated with tissue damage, ion and metabolite levels, and for stress indicators.

The salmon fed less starting at 6°C, and FC was only 10% of that at 8°C at 1-2°C. During the 2 weeks at 1°C, 5% of the fish died, and many of the fish had elevated values of HSI (1.57 ± 0.05 compared to 1.16 ± 0.04). However, there were signs that fish health/welfare was impacted before this point. An ionoregulatory disturbance was apparent at 4°C, with cold-exposed fish having lower levels of K⁺ and higher levels of Na⁺ and Cl⁻. However, an increase in plasma cortisol levels was only observed 1°C (18 ng mL⁻¹), there was no increase in plasma lactate (i.e., no indication of the onset of anaerobic metabolism), and none of the plasma levels of enzymes indicative of tissue damage (creatine kinase, aspartate aminotransferase, or lactate dehydrogenase) were elevated. Thus, Atlantic salmon show changes in appetite and physiology beginning at 6 and 4°C, respectively, and stress, mortality and other signs of compromised health are apparent with even short-term exposure to cold temperatures. Clearly, even short periods of exposure to temperatures $\leq 1^\circ\text{C}$ pose a unique challenge to growers of Atlantic salmon in regions that experience these low water temperatures.

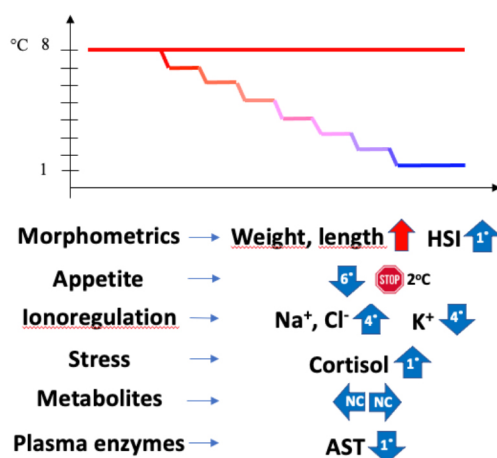


Figure 1. Impact of a seasonal decline in temperature on Atlantic salmon.

***Aeromonas salmonicida* INFECTION KINETICS AND AUTOGENOUS VACCINE EVALUATION IN SABLEFISH (*Anoplopoma fimbria*)**

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Cultured sablefish (*Anoplopoma fimbria*), is one of the most valuable cultured fish species in Canada's Pacific coast. Effective vaccine programs against *A. salmonicida* are high priority for sablefish production. In this study, an infection model in cultured sablefish was established, a vaccine challenge model and the evaluation of the immune protection provided by an *A. salmonicida* autogenous vaccine preparation compared to two commercial vaccines (Forte and Alpha Ject). Atypical *A. salmonicida* J410 lethal dose 50 (LD₅₀) was estimated to be 3×10^5 CFU/dose in sablefish. Immune protection for the different vaccine preparations was evaluated in a common garden experiment. Blood samples were taken bi-weekly to evaluate IgM titers. After 10 weeks post-immunization the fish were intraperitoneally challenged with 100 times the LD₅₀ dose (10^7 CFU ml⁻¹). Thirty days post-challenge the relative percent survival (RPS), compared to the control, was calculated for each vaccine. The RPS for the bacterin mix was 63.7%, 54.57% for the Forte vaccine, and 27.28% for the Alpha Ject vaccine. *A. salmonicida* tissue colonization 10 days post-challenge negatively correlated with the RPS. Additionally, ELISA assays indicated differences in IgM titers between vaccines at 6-8 weeks post-challenge. It was determined that *A. salmonicida* A-layer binds to immunoglobulins F(ab)' in a non-specific fashion. These study results indicate that vaccine design influences sablefish immunity and provides a guide for future sablefish vaccine programs.

UPDATE OF THE TRANSCRIPTOMIC PROFILE OF ATLANTIC SALMON (*S. salar*) PRIMARY MACROPHAGES RESPONSE TO *Aeromonas salmonicida* subsp. *salmonicida* INFECTION

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A. salmonicida is one of the oldest known marine pathogens with an increasing host-range from freshwater to marine fish species, mainly affecting salmonids, and currently emergent cultured fish species such as lumpfish, Atlantic cod, sablefish. *A. salmonicida* infection causes furunculosis, resulting in a systemic shock, organ failure and ultimately death. Understanding *A. salmonicida* infection development at early stages is critical for the disease output, including intracellular infection, immune response elusion, and propagation to lymphoid organs. Here, we performed a semi-dual RNA-seq based technology to identify relevant genes for *A. salmonicida* early infection in real-time in Atlantic salmon primary macrophages. RNA samples were taken after 1 h post-infection and 2 h post-infection, mimicking cellular attachment and intracellular infection. We identified a total of 871 differential expressed genes (DEGs) at 1 hpi and a total of 1,683 DEGs were identified at 2 hpi. Gene ontology (GO) of these DEGs during the attachment of *A. salmonicida* to the host cell showed a negative impact on the expression of genes related to cellular stimulus and biological processes, ion binding and integral components of the membrane. In contrast, during an early infection process, expression of genes associated with cytoskeleton organization and actin polymerization, phospholipid binding, plasma membrane organization, cytokine activity, inflammatory response, wound healing, calcium transport and RNA polymerase II transcription factor activity are modulated. The enrichment of KEGG pathways showed that *A. salmonicida* hijacks pathways related to innate immune response, lipids synthesis, gene transcription, cellular arrest, and apoptosis, mainly affecting cytokine-cytokine receptor interaction and protein processing endoplasmic reticulum, toll-like receptor signaling, RIG-I-like signaling, p53, and necroptosis pathways. These indicate that *A. salmonicida* induces and modulates phagocytosis and apoptosis to survive in an intracellular environment during the early stages of furunculosis.

IMPACT OF SOUND INTENSITY RELATED TO MARITIME TRANSPORT ON THE EMBRYOGENESIS AND METAMORPHOSIS OF THE BLUE MUSSEL *Mytilus edulis*

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The blue mussel *Mytilus edulis* is a sessile organism with a major role in the development, the functioning and the sustainability of coastal environments but also reared in shellfish farms located in coastal areas under high anthropogenic influence. In recent years, maritime traffic activity has considerably increased with a serious concern about the effects of sounds generated by ships, considered as an emerging pollutant. Impact of ship underwater noise on the larval development and the subsequent recruitment success of the blue mussels is the objective of this study. We present for the first time the impact of a cargo ship sound on both embryos and competent larval stages of blue mussels at different levels of stress, by correlating the complementary metabolomic, lipodomic, behavioral, growth and survival responses obtained.

A month-long experiment was conducted in *Larvosonic* mesocosms, created to emit sounds with low reverberation and resonance. Thus, we exposed developmental stages to contrasting sound treatments including a control (ambient sound) and three increasing levels (low, medium and high; from 132 to 150 dB re 1 μ Pa) of a previously in situ recorded sound sequence of a 120 m cargo ship. We suggest that critical windows to stress could occur during larval development, and thus tested several ontogenetic phases as embryogenesis (embryo to larva-D) and metamorphosis (pediveliger to post-larvae). Several analyzes were carried out to explore the primary, secondary and tertiary stress responses to sound including i) prostaglandins and oxidative metabolism, ii) energy metabolism and analysis of lipid profiles and iii) larval growth through shell measurements (Prodissoconch I, II and Dissoconch, Figure 1). Preliminary results show that larvae subjected to high sound intensity settle faster and have greater post-larval growth compared to other conditions. However, when they are subjected to lower sound intensities then a delay in metamorphosis seems to be observed.

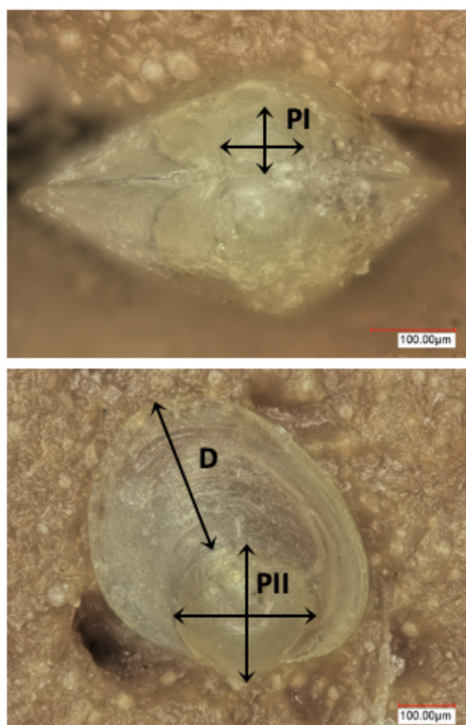


FIGURE 1. Picture of a *M. edulis* recruit showing the measures acquired on the shell.

MOLECULAR, FUNCTIONAL AND TRANSCRIPTIONAL ANALYSIS OF SEAHORSE (*Hippocampus abdominalis*) B-CELL LYMPHOMA-2 ASSOCIATED X PROTEIN AND ITS POTENTIAL IMMUNOLOGICAL ROLE

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Apoptosis is the best-studied form of programmed cell death, which is important for the development and maintenance of homeostasis within multicellular organisms. Bcl-2 family members are major apoptosis regulations in mammalian cells. B-cell lymphoma-2 associated X protein (Bax) is a well-known proapoptotic gene that belongs to the Bcl-2 family.

Seahorse complete cDNA of *HaBax* got from seahorse cDNA transcriptomic database. *HaBax* protein characteristic and structural features were analyzed by several bioinformatics tools. The expression level of mRNA in seahorse was analyzed by qPCR technique. Conserved domain parts of the *HaBax* were determined by using ClustalW. The phylogenetic tree analysis was constructed according to the Neighbor-joining (NJ) method by MEGA 5.0. The 3D structure was constructed by I-TASSER server. To measure the immune response of *HaBax* on stimulant or pathogens, each group of seahorses was injected with lipopolysaccharide, *Edwardsiella tarda*, *Streptococcus iniae* and polyinosinic: polycytidylic acid. Then total RNA was extracted from each tissue and did the qPCR assay. The Bcl-2 like domain and transmembrane domain in the *HaBax* open reading frame (ORF) were identified by using online software tools. The *HaBax* mRNA was highly expressed in ovary tissue compared with others. The phylogenetic analysis represented that the *Hippocampus comes* Bax and *HaBax* have a closer relationship with each other. In response to the infection with Poly: IC, LPS administration, *E. tarda* infection, and *S. iniae* significantly increased *HaBax* expression respectively. *HaBax* is a protein composed of 204 aa. *HaBax* represent high aa sequence identity with fish homologs, but low identity with reptiles, mammals and bird's homolog. We find the evolutionary stage of that gene in vertebrate evolution. We identified the *HaBax* mRNA content in various tissues and assessed that was upregulated with the certain immune stimulants and pathogens. Findings of the current study represent the overall informative information related to the *HaBax* gene of the seahorse.

CLIMATE CHANGE VULNERABILITY ASSESSMENTS: CONSIDERING AQUACULTURE

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The global population is increasing along with the demand for secure and sustainable seafood. Aquaculture production is growing to meet this demand, but the heavy reliance of most culture systems on the ambient environment and ecosystem services suggests an inherent vulnerability to climate change. In order to design and implement effective climate change adaptation, policymakers and growers need to understand where their industry is vulnerable and how climate change stressors may affect their sector. Climate change vulnerability assessments are an internationally recognized assessment process that can help growers and decision-makers plan for climate change. This form of assessment is relatively new to aquaculture but has begun to see application to regional culture systems such as salmon in Chile, multi-species in Korea, and oyster culture in Australia.

Vulnerability assessments are generally based on three indices: ‘*Exposure*’, ‘*Adaptive capacity*’, and ‘*Sensitivity*’, which are combined to form an overall vulnerability index. Data inputs for these indices can be quantitative, qualitative, or a combination of both (semiquantitative), and can be challenging to combine, given different data types and collection methods. For example, quantitative inputs such as wind and wave exposure may have magnitude and frequency data generated from climatic models, whereas qualitative inputs such as farmers’ perceptions of risk are assessed through questionnaires and interviews. The result is typically a semi-quantitative model, that translates different data types into a final understandable value. The availability of data or scale may also be a challenge, as some regions or species may have more available data than others. In this case, a more qualitative approach, such as expert opinion, may be sought.

Despite its limitations, climate change vulnerability assessments are an important tool for guiding decision-makers and prioritizing adaptation efforts. Here we present current practices and limitations of climate change vulnerability assessments in consideration of application to aquaculture.

EFFECT OF DIET AND TEMPERATURE ON GONAD ENHANCEMENT AND WASTE DEPOSITION FOR THE RED SEA URCHIN (*Mesocentrotus franciscanus*)

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When sea urchin population growth is uncontrolled, they can over-graze and decimate macroalgal beds, creating areas termed “urchin barrens”. Sea urchin gonad enhancement is a proposed method to remove sea urchins from barren grounds, promote the re-growth of macroalgal beds, and produce a highly-valued marketable product. Prepared feeds need to be developed that not only increase gonad yield, but also impart appropriate market colour and flavour. For sea-based enhancement, there may be benthic impacts due to organic enrichment from uneaten feed and faeces, but this has not been examined before. This project assessed the effects of two prepared diets (V10.1.9 and V10.1.10) and a natural feed (bull kelp, *Nereocystis luetkeana*) and three seawater temperatures (8, 12, and 16°C) on gonad yield and quality for the red (*Mesocentrotus franciscanus*) sea urchin held under laboratory conditions for 12 weeks. Red urchins fed V10.1.10 at 12 °C had the highest gonad yields out of all the treatments, while the red urchins fed kelp at 8°C had the lowest yields. The gonad colour was better overall with the red urchins fed V10.1.10, with the best colour obtained at 16°C. Ingestion rate, and faecal pellet size and settling velocity were also measured and will subsequently be used in a waste deposition model (along with various oceanographic variables of a proposed site) to predict the potential environmental impact a commercial-scale sea urchin farm could have.

FISH SLUDGE – A PROBLEM OR A RESOURCE?

Steinar Wasmuth*

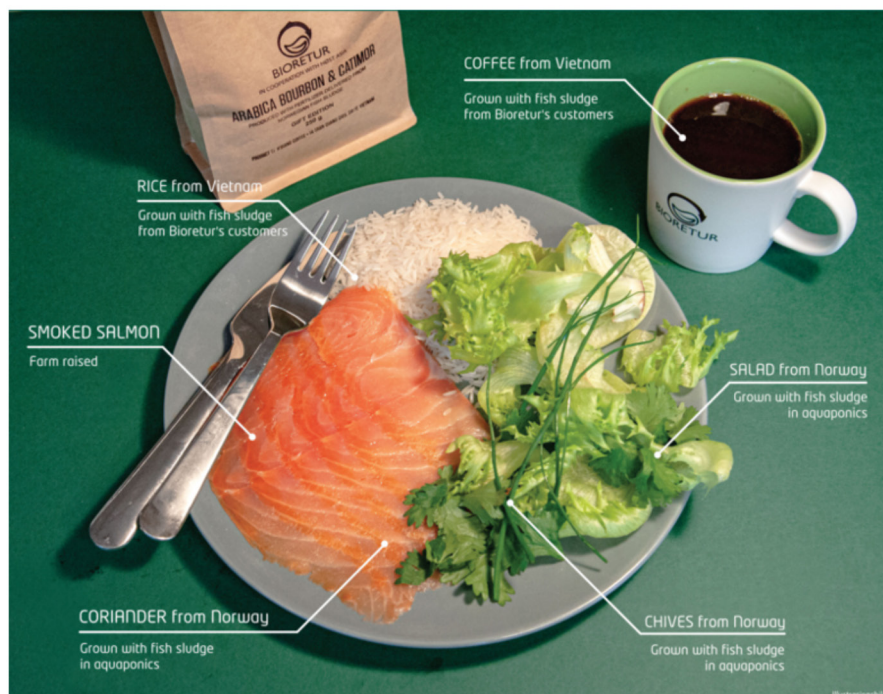
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Fish sludge from land-based aquaculture facilities can be considered The Elephant in the Room, if this problem is not addressed and handled it can be a barrier for new land based project and further growth in marine food production. Fish sludge consists of feces and food waste from the production in the aquaculture facility, and is high on phosphates and other important minerals that is essential for sustaining life on the planet. Fish sludge also contains heavy metals such as zinc (Zn) and cadmium (Cd) and can thus not be used as is for fertilizer without special treatment and regulations. To be able to reach the climate goals, reuse of minerals such as phosphorus in a circular economy is essential, and we can simply not continue to waste thousands of tons of fish sludge every year.

Treatment of sludge in land-based aquaculture often consists of mechanical dewatering from 0,1 DS (dry solids) to around 15 % DS before it is transported to a biogas facility. Because the aquaculture sites often are far away from the biogas plants, this means transporting enormous amounts of water every year. This is highly expensive and not sustainable because it results in thousands of transport mileages. The use of fish sludge in production of biogas also results an high volumes of bio rest materials, with concentrated heavy metals.

The Norwegian company Bioretur offers a solution to the fish sludge problem and gives fish farmers access to the best available technology for sludge treatment for the aquaculture industry. At the same time they get a partner who ensures that the sludge is transformed from a problem to a sustainable resource. Bioretur are experts on sludge, and their value proposition is that for a fixed monthly fee they build, operate and maintain the technology of sludge treatment, while the client do what they do best: Produce high quality fish and seafood!

For the fist time, Steinar Wasmuth will present the Bioretur concept in Canada.



ESTABLISHING A STAKEHOLDER GROUP TO COMBAT VANDALISM OF DATA BUOYS AND MARINE SCIENTIFIC EQUIPMENT IN CANADIAN WATERS

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Advanced data collection buoys for marine environments are becoming common to inform a variety of marine users, including aquaculture producers. Despite improving access and ease of environmental data collection, there are a number of buoy deployment and maintenance challenges that require resolution. Vandalism of deployed marine equipment is a serious issue affecting multiple stakeholders. Vandalism may be intentional, such as theft, or removal from moorings, or unintentional, such as through fishing gear entanglement or wildlife interactions. Regardless of cause, this consequence can be devastating for research programs or decision support. Vandalism can result in the loss of expensive equipment, data, or interruptions in the continuity of time series. Recovering equipment often requires the use of contractors to find the gear, repair damages, and redeploy, all of which requires additional resources. To help resolve some of these issues the Centre for Marine Applied Research has implemented a stakeholder group which includes government, industry, and academia stakeholders to examine successes and failures in marine equipment deployment, with an aim to resolve vandalism mitigation strategies. This presentation explores challenges and solutions to address vandalism of marine equipment with a focus on data buoys.

NOAA OPTIONS TO SUPPORT THE U.S. RAS SECTOR

Chuck Weirich* and Clete Otoshi

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1315 East-West Highway, Room 11735
Silver Spring, Maryland 20910

Efforts are underway to inform a decision on if and how NOAA could support the emerging U.S. recirculating aquaculture system (RAS) sector. This presentation will include:

- An overview of NOAA's support of RAS research to date,
- Current status of RAS commercial projects in the U.S.,
- Industry research needs and potential applications of RAS technologies, and
- Potential avenues that NOAA can support RAS as a viable aquaculture production system in the US and recommendations under current mechanisms and funding levels.

CONCERNS, CONFLICTS AND CONTROVERSY: UNDERSTANDING DRIVERS OF OPINION AND OPPOSITION TO SALMON FARMING IN NOVA SCOTIA, CANADA

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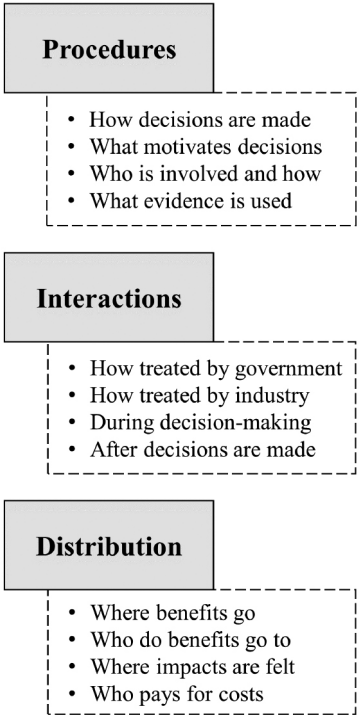
Salmon aquaculture is a publicly controversial issue in Canada, with strong opposition being mobilized by certain groups such as environmental NGOs, fishing groups, and residents. Recently, social aspects are increasingly being recognized as important for more sustainable aquaculture management, fostering public trust and creating a positive environment for growth of the industry. The present study was conducted to better understand the motivations of opposition and identify factors influencing societal attitudes to salmon farming, using Nova Scotia as a case study.

This work is supported by two approaches. First, an online public survey aimed to capture views and opinions of 495 Nova Scotians towards salmon farming. Rural and urban respondents were significantly different in their perceptions and opinions of aquaculture, highlighting the need to incorporate both perspectives in understanding conflicts and fostering acceptance. Statistical models identified five major drivers best predicting negative views of aquaculture, including: age, environmental value system, salmon consumption patterns, trust in government, and perception of risks. Aquaculture knowledge and exposure had little influence on opinion but may still help people who are undecided make decisions.

Second, interviews with diverse representatives within three salmon-farming communities in Nova Scotia provided a comparative analysis of how locals have experienced aquaculture and identifies common dimensions of conflict and controversy. Three main dimensions emerge from conflict with both industry and government and include aspects regarding decision-making procedures, interactions, and distribution of outcomes (Figure 1). As a result, social acceptance of salmon aquaculture is highly tied to public trust and concerns over how industry is managed.

Drawing from this work, we find that opposition does not represent a uniformed set of experiences, concerns, or conflicts. Social concerns revolve not only around material (e.g., economic) well-being, but highlight the specific importance of relational well-being, being the relationships with family, community, and their environment. Still, findings reveal that opinion is nuanced across demographic and geographic contexts. Responding to opposition will require understanding variability in perceptions and considering the underlying personal, social, and geographic contexts in which they are embedded.

FIGURE 1. Main dimensions of conflicts around salmon farming.



eDNA FOREECOSYSTEM AND AQUACULTURE MONITORING TO SUPPORT IMPLEMENTATION OF THE US NOAA 'OMICS STRATEGIC PLAN

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Omics is one of six US National Oceanic and Atmospheric Administration (NOAA) Science and Technology focus areas that were introduced in 2020, resulting in the implementation of the NOAA 'Omics 2021-2025 Strategic Plan (<https://tinyurl.com/ycxt6ekx>). Investment in 'omics is motivated by a need to increase operational efficiency for a broad suite of activities under NOAA's mission, including ecosystem and fisheries assessments, understanding the impact of climate change, and contributions to the US Blue Economy, including aquaculture. The overarching goals of the Omics Strategic Plan are focused on enhancing infrastructure for the collection and analysis of 'omics data; targeting research in marine forensics, sustainable aquaculture, biosurveillance, bioprospecting, and for protected resources; accelerating transitions of 'omics into regular operations; expanding partnerships and funding for 'omics work; and supporting a diverse and inclusive workforce for 'omics work. One of the goals listed in the strategic plan is to foster the development of aquaculture by using 'omics to optimize animal health, yield, and product characteristics while supporting safe and sustainable farming practices. The utility of environmental DNA (eDNA) in the NOAA 'Omics Strategic plan and across ongoing research within the agency is diverse, and ranges from evaluating biodiversity and abundance of marine microbial, invertebrate, and vertebrate communities in regular assessment and exploration surveys, to evaluating ocean change and human impacts on local and regional scales. These and other activities are relevant to a number of international and bilateral activities, including the All-Atlantic Declaration, UN Decade of Ocean Science Endorsed Programs, and Collaborations with DFO

We present herein an overview of these activities that use eDNA as a tool within the NOAA 'Omics community, in particular focusing on aquaculture applications. NOAA uses 'omics tools to address a broad range of mission applications, including development of eDNA for monitoring microbes through marine mammals. For example, We are using eDNA analysis to understand the impacts of netpen and other types of in situ aquaculture on ecosystem diversity, fate & transport of eDNA, and the microbiome characteristics related to the health of aquacultured plants and animals. Efforts include development of autonomous methods to collect eDNA samples and transition to operations.

AN ANALYTIC APPROACH TO INCREASING SUCCESS AND A POSITIVE PRESENCE IN AQUAPONICS

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Aquaponics has been an emerging industry with steady growth for quite some time. Its development has shown a strong increase in academic papers alongside a unique set of foci and considerations particular to the practice. However, despite the progress made in the industry, aquaponics is still taken to be a hobbyist practice, with trends treating the practice as a poor analogue to hydroponics. Data analyses ran through Google Scholar, keyword searches, Google Trends, and sentiment analyses indicate that considerations for aquaponics on the industry level do not coincide with a consumer basis and show little overlap with the research community. In order to maintain appeal and marketability for aquaponics, it is necessary to bridge the gap between consumer and industry expectations through an analytic approach through its online presence.

A multifaceted approach was used to gather analytics. Python was used to scrape Google Scholar for the keyword “aquaponics” and found a steady increase in scholarly articles, with trends and associated keywords similar to hydroponics. Google Ads and keyword planning showed very little technical terms and a focus on “how-to” and instructional. Very few specifics were associated with keyword “aquaponics” and the only prominent brands were at-home, hobbyist builds under 10 gallons. Google Trends indicate that aquaponics is still treated as a counterpoint to hydroponics, with searches in decline. Curiously, organic appeal was not a concern. Hydroponics and aquaponics seemed to have respective location “territories”, where hydroponics had a heavier sense of brand trust. Finally, using R software to datamine social media and perform sentiment analyses showed a neutral sentiment towards aquaponics, with primary keywords focusing on at-home hobbyist levels.

Following as an extension of the research presented at the World Aquaculture Society 2020 Hawaii conference (*Keeping Interest in Aquaponics Alive: a Composite Look at Trends, Keywords and Sentiment Analyses in the Industry*), this presentation further details the limitations of aquaponics set forth from the consumer perspective and seeks further measures to translate the mitigated focus from the online community into a positive presence that could help increase the success of current operators. Aquaponics has a decisive market in US and global locations, but lacks brand awareness, consumer understanding, and little knowledge of large-scale operators and nuances that could positively influence purchase decisions. It is necessary to focus on outreach and community-driven strategies to capitalize on opportunities to shift the public opinion.

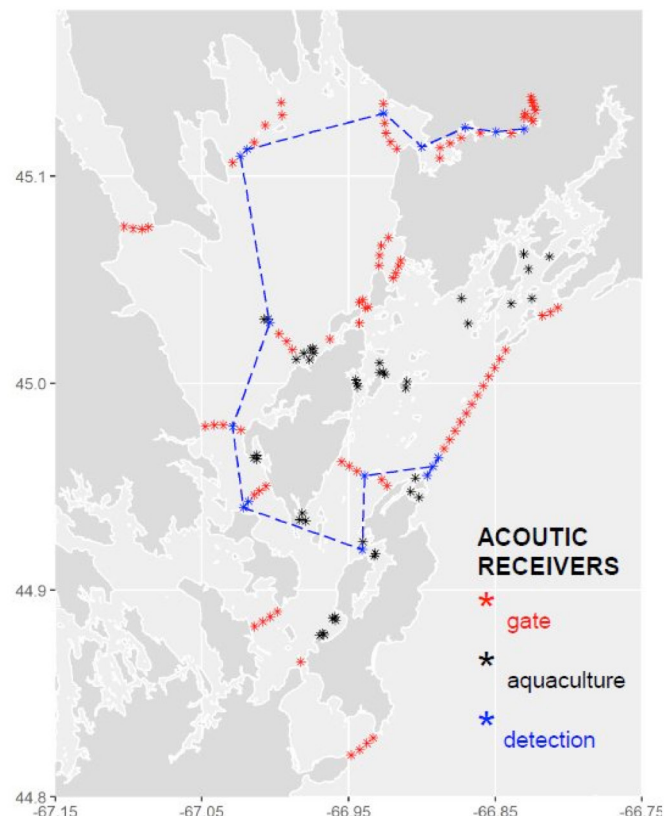
ASSESSING THE EFFECT OF MULTIPLE STRESSORS ON THE ESTUARINE AND EARLY MARINE SURVIVAL OF ATLANTIC SALMON POST-SMOLTS

Brent Wilson*, Marc Trudel, Claire Rycroft, Jon Carr, Jason Daniels, Dave Hardie, Jim Hawkes, Chris McKindsey, Fred Page, Shawn Robinson, Émilie Simard, Fred Whoriskey, and Glenn Crossin

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Atlantic salmon have been extirpated from several of their native rivers and often fail to meet conservation requirements, particularly populations in the Bay of Fundy, Canada and northeastern United States. Although multiple factors likely contributed to these declines, such as habitat destruction, overfishing, and climate change, the cumulative effects of multiple stressors on the survival of Atlantic salmon is poorly understood. Here, we present the results of an acoustic telemetry project to assess the population response of Atlantic salmon to multiple stressors during a critical period of their life cycle, including passage through a hydropower system and bycatch mortality associated with in-river fisheries. We also assessed their migration routes and survival rates during their early marine life to determine whether there are any interactions with aquaculture (Figure 1). For this project, a total of 160 smolts were tagged with pressure and temperature sensor acoustic tags and released above and below a hydropower dam, before and during a commercial alewife fishery. A total of 129 acoustic receivers were placed in the river and estuary of the Magaguadavic River (New Brunswick, Canada), at all passages in Passamaquoddy Bay, as well as at twenty-three salmon aquaculture sites located in three Bay Management Areas (stocked with year 1 or year 2 fish, or in fallow) (Figure 1). Generally, there was little evidence for direct or latent effects of dam and fishery passage with relatively low cumulative residence demonstrated at salmon aquaculture sites. It is anticipated that this research will provide information necessary to develop effective mitigation strategies to limit associated threats, and aid in the recovery of wild salmon populations.

Figure 1: A map of the acoustic receiver array and release site in Passamaquoddy Bay, with the migration route of a tagged salmon exemplified.

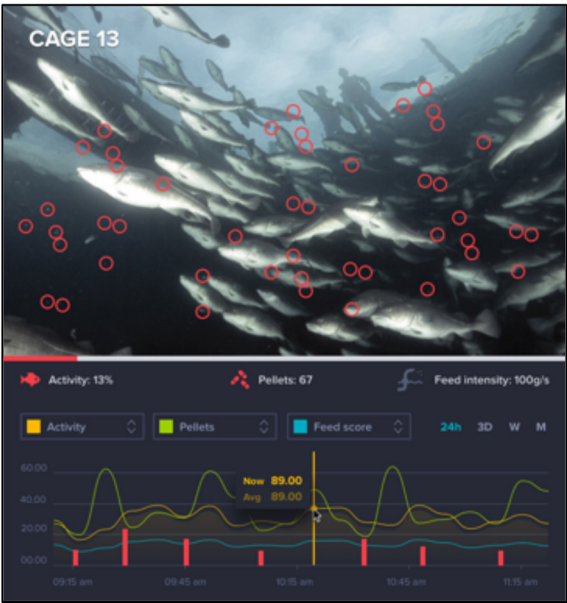
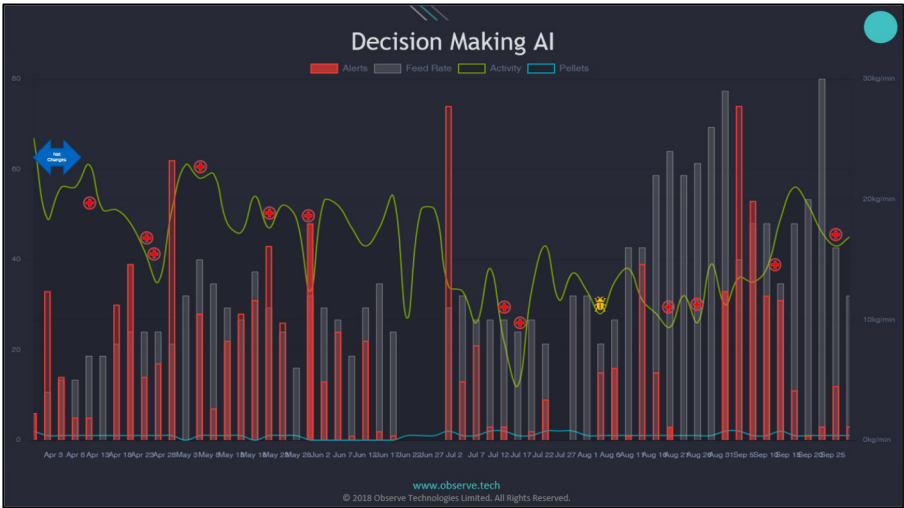


DIGITALIZATION AND ARTIFICIAL INTELLIGENCE FEEDING INNOVATIONS IMPLEMENTED IN INDUSTRY APPLICATION

Laura Windatt

Imagine an industry empowered by rich real time data visualization, accurate and efficient operations and a future where every fish is accounted and cared for. Digitalization and artificial intelligence are the instruments to drive these values. Fish welfare, safety, operational control and efficiency will be right at your fingertips. Every physical condition monitored. Every feeding automated. Every environmental change, environmental threat, behavioral irregularity and growth condition monitored, analyzed, visualized and adhered to. These instruments will be used as a foundation for improved operations, knowledge sharing and provide sustainable results.

Since January 2019 AKVA group has been providing the intelligent feeding assistant for farmers around the world. Discover industry experience with different case studies that captured the importance of real time feeding data that has never been recorded before and has aided in maximizing fish performance.



DETECTION OF CYANOTOXIN MICROCYSTIN-LR (MC-LR) WITH PORTABLE OPTICAL SENSOR

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Harmful algae blooms (HABs) are excessive growth of certain types of algae or cyanobacteria that cause grave danger to humans, wildlife, and the ecosystem. HABs are also an increasing threat to aquaculture not only by reducing dissolved oxygen but also by poisoning fish species with their secondary metabolites cyanotoxins. The acute toxicity of these secondary metabolites is triggering mass mortality in various fish species and harvesting bans on toxin accumulated shellfish. The economic loss due to HABs in aquaculture is reaching a few billion dollars per year globally. Moreover, the incapability of the current algae detecting microscopic method to recognize cyanotoxins is enhancing their adverse health and economic impact.

Our research has focused on the development of portable sensors that can deliver real-time, accurate information on cyanotoxin concentration. The sensor consists of three main components: a recognition unit, a response unit and a detection unit. The recognition unit is made of a synthetic receptor molecularly imprinted polymer (MIP) that provides high target selectivity and long-term stability; the sensing unit is comprised of fluorescent material quantum dots (QDs) that offer high sensitivity and visualizable sensor signal; the detection unit consists of a simple light detector that imparts sensor with portability. In the first stage, the sensor was tested using model cyanotoxin microcystin-LR (MC-LR). The sensor was able to show an increasing sensor response to elevating MC-LR concentration, high selectivity against other common chemicals in water and a limit of detection of 1 $\mu\text{g/L}$.

On account of the synthetic nature of the MIP receptor and the wide-range applicability of the fluorescence materials, the developed sensing materials can be easily modified to detect other families of cyanotoxins to offer comprehensive monitoring of HABs. Therefore, the development of this sensor provides a fast and cost-effective method of monitoring cyanotoxin in real-time and a chance to take immediate action to mitigate the effect of HABs on aquaculture.

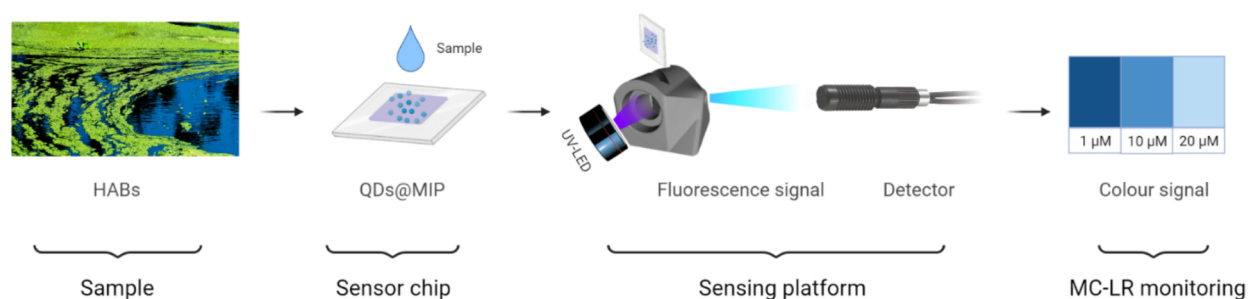


Figure 1: Graphical abstract of the cyanotoxin sensing platform. The sensing material formed with QDs and MIP was coated on quartz to generate sensor chips. The fluorescence signal of the sensor was induced by UV Light Emitting Diode (LED) and measured by a portable detector. Finally, the intensity of fluorescence signals and the colour signal were used to analyze cyanotoxin concentration.

HIGH-THROUGHPUT *de novo* MITOCHONDRIAL GENOME ASSEMBLY PIPELINE SUPPORTS THE OPTIMAL ENVIRONMENTAL DNA ASSAY DESIGNS IN A GLOBAL CLIMATE CHANGE ERA

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Canada is a country with highly rich biodiversity stretching from the Atlantic Ocean to the Pacific Ocean. Biodiversity loss is currently at an unprecedented rate within Canada due to climate change, overfishing, habitat degradation, etc. Development of new environmental monitoring tools for targeted coastal and inland ecosystems is highly necessary. Environmental DNA (eDNA) methods using the genetic material shed by organisms have been applied to monitor at-risk and invasive species across many agriculture and ecology fields. Common eDNA assays include environmental sample collection, followed by DNA extraction and quantitative PCR (qPCR) analysis. This eDNA-based rapid, non-destructive and cost-effective approach helps detect the presence of invasive, at-risk, culturally and economically important species. However, many previously established eDNA qPCR assays lack high-quality primers and this has historically led to poor assay sensitivity and high false positives.

To optimize the eDNA assays, taxa-specific genomic regions need to be characterized. Mitochondrial DNA (mtDNA) is an ideal target for unique genomic sequence identifications as mtDNA have hundreds to thousands of copies in each eukaryotic cell, making it easier to be extracted from the whole-genome shotgun sequencing data through bioinformatics approaches. Due to the high cost for sequencing in the past, the availability of complete mitogenomes in public databases such as National Center for Biotechnology Information (NCBI) is limited and the intra-species variations were not well represented. Hence, we aim to develop a high-throughput *de novo* mtDNA assembly pipeline that allows the generation of large quantities of standardized high-quality mtDNA genomes which can be later used in capturing unique mtDNA sequences of target taxa. The key steps of the proposed bioinformatics pipeline includes *de novo* assembly, gap filling/polishing and start-site/strand standardization (Figure 1).

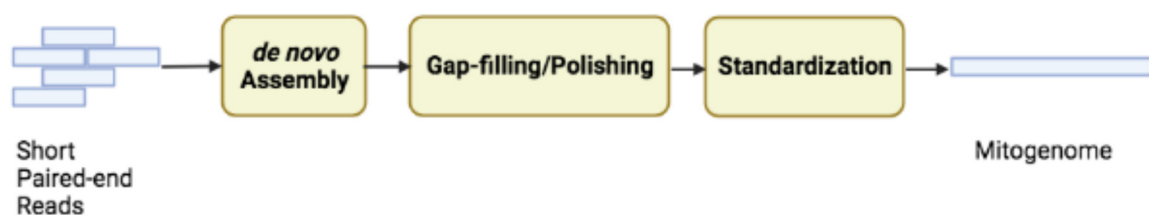


Figure 1. Overview of the mitogenome *de novo* assembly pipeline.

EVALUATION OF PHOSPHOLIPID AND EICOSANOID PROFILES IN ATLANTIC SALMON (*Salmo salar* L.) MUSCLE AFTER FEEDING MICROBIAL OIL USING ELECTROSPRAY IONIZATION (ESI) TRIPLE QUADRUPOLE MASS SPECTROMETRY

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Changes in phospholipid and eicosanoid profiles of Atlantic salmon muscle after feeding diets with different levels of microbial oil, fish oil and canola oil were investigated using electrospray ionization (ESI)-MS-MS in a triple quadrupole mass spectrometer. A total of 43 PL species with various fatty acyl chain combinations was identified in the four dietary groups of salmon. The group fed with microbial oil, which contains a high level of DHA, showed an increased level of DHA in the structure of PC (phosphatidylcholine) and PS (phosphatidylserine) in muscle tissue compared to salmon fed with fish oil and the mixture of fish oil and canola oil. This indicates that DHA present in the diets (High and low microbial oil) was efficiently converted/synthesized into phospholipids, followed by accumulation in salmon muscle tissue. In addition, two eicosanoids, prostaglandin E₂ and F3 α , were also identified for the first time in salmon muscle tissue and their quantitative changes upon feeding four different diets will be discussed in detail. This study suggests that the shotgun lipidomic approach along with neutral loss and precursor ion scanning modes is a useful means for the lipidomic analysis of fish tissues.

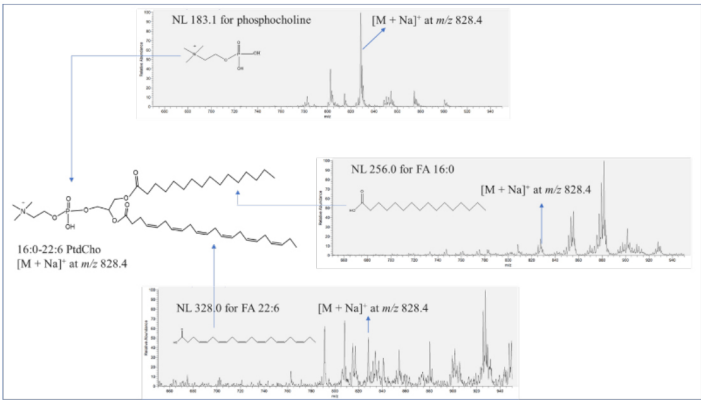


Figure 1. Identification of phospholipids using ESI-MS-MS with multiple neutral loss scanning mode.

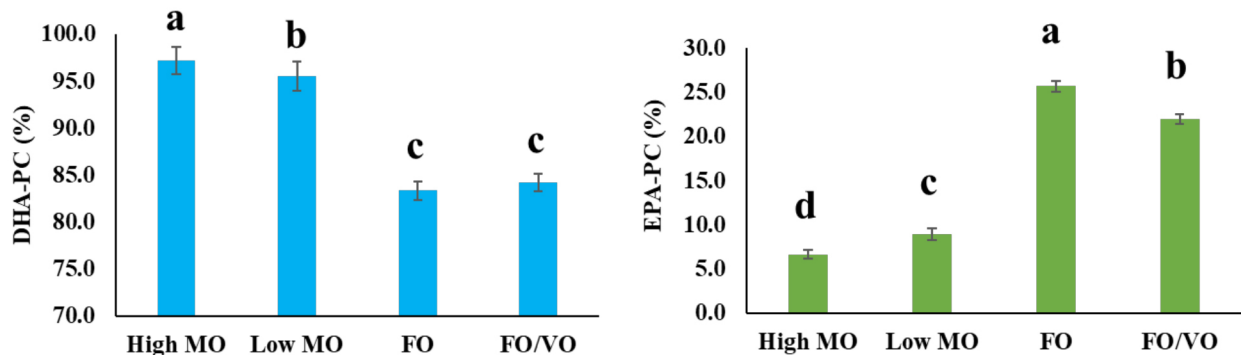


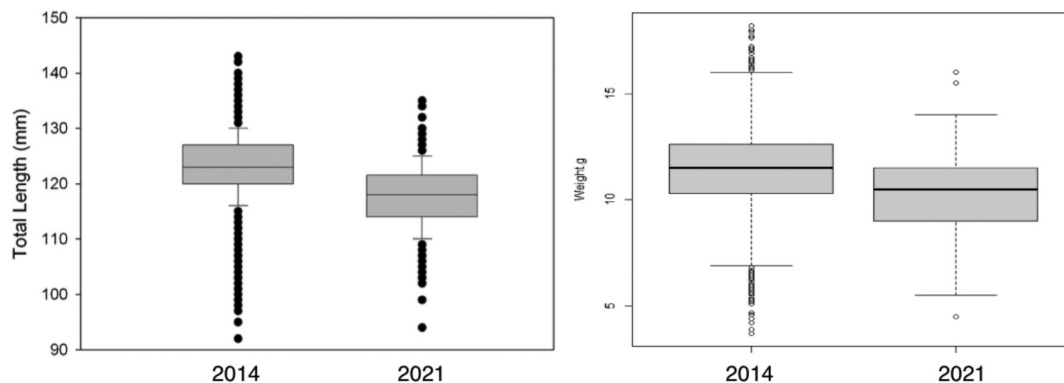
Figure 2. DHA containing PC (DHA-PC) and EPA containing PC (EPA-PC) in salmon muscle tissue after feeding High MO (High Microbial oil), Low MO, FO (Fish Oil), FO/CO (Mixture of Fish Oil and Canola Oil) for 16 weeks.

CO-MANAGEMENT AND STATUS OF NIGHT SMELT *Spirinchus starksi* IN NORTHERN CALIFORNIA IN 2021

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Night smelt (*Spirinchus starksi*) occur from Central California to South East Alaska and spawn on the shore of sandy beach surf zones. This species is a vital part of regional food webs and in California, is fished commercially, recreationally and for subsistence, primarily in Humboldt and Del Norte counties. Night smelt are known as ch'uy-xee-ni in the Tolowaa Dee-ni' Nations' language. These fish are seen as an imperative part of the food resources for Indigenous tribal nations and have been documented as culturally significant to thirteen sovereign nations in the occupied north coast of California (Tolowa Dee-ni' Nation, 2017). This species has been fished for subsistence and cultural purposes for time immemorial (Palmer et al. 2018). Despite the ecological, commercial and cultural importance of this species, very little is known about its biology and ecology. Consequently, there are currently no criteria to identify when this fishery is overfished or in decline. To study the status of these populations we collected adult night smelt with an A-frame net at six beaches in Humboldt and Del Norte counties once a month from March to September 2021. We then statistically compared length (mm, Total Length), weight (g) and sex ratio with data collected at the same beaches in 2014. We caught 529 fish on 9 of 35 days sampled in 2021. Length and weight averaged (\pm Standard Deviation) 11.7 ± 0.8 cm TL and 10.4 ± 1.9 g, and were significantly lower than 2014 values (12.2 ± 6.0 cm TL, 11.5 ± 2.5 g, respectively, $p < 0.01$). Most of the fish were males (96%), which was slightly higher than during 2014 (93%, $p < 0.05$). Our study suggests that at present Humboldt and Del Norte night smelt are smaller, lighter and more of them are males than almost a decade ago. These results suggest that these populations are in decline, potentially due to overfishing or unfavorable oceanographic conditions, and may require stricter regulations such as seasonal or annual closures.



**PISCINE ORTHOREOVIRUS (PRV) IN SALMON IN BRITISH COLUMBIA:
RISK VS CONSEQUENCES**

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Our measured physiological consequences of injecting concentrated, BC-strain PRV into juvenile sockeye and Atlantic salmon were incompatible with a previous suggestion of sub-lethal cardio-respiratory impacts. Peak PCR response at 4-6 weeks post-injection and a sustained PRV infection, at or beyond that seen in wild and farmed salmon, produced no mortality and body condition was maintained. Well-established respirometry techniques for individual fish compared time-matched measurements of routine (RMR), maximum (MMR) and hypoxic respiratory performance with sham-injected salmon. While blood hemoglobin concentration and hematocrit decreased modestly at week 4 post-infection in sockeye and at weeks 1 and 9 in Atlantic salmon, these modest changes were not sustained at week 9 in sockeye or at week 18 in Atlantic salmon. However, neither RMR, MMR nor hypoxia tolerance of PRV-infected salmon were significantly different from their time-matched controls, except MMR was significantly higher in infected Atlantic salmon at week 10.

