WA '21

World Aquaculture 2021

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May 24 -27, 2022

Mérida, Mexico

Centro Internacional de Congresos de Yucatán, CIC

Annual global meeting of the World Aquaculture Society

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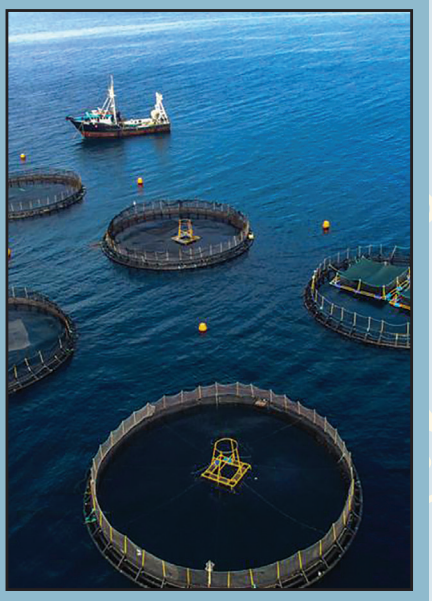

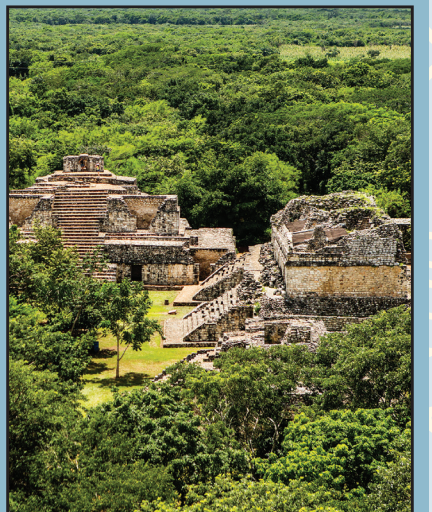

WELCOME MESSAGE

Dear all:

Welcome to the World Aquaculture 2021 Conference. It is an honor for me to receive you in the beautiful city of Mérida, Yucatán, México. I am sure that during the following days we will have the opportunity to share many experiences, meet old friends, start new businesses, and contribute to the development of aquaculture in the region. Mérida is a place that will be providing all of us with the best amenities, incredible cultural and outdoor activities, and outstanding gastronomy that distinguishes her as the most beautiful city in México.

Bringing a global event to the Latin American and the Caribbean Region is something that confirms the importance to WAS of continuing to be the most important aquaculture professionals' association in the world; where our diversity is one of our bigger strengths. This diversity is confirmed in our Board of Directors, in which every single continent is represented, and where all ideas and points of view are welcome. I invite you to get involved and keep expanding our mission.

Our trade show, as well as our scientific conferences, constitute the back bone of our events. I will like to thank all our sponsors and expositors for being part of the WAS family and making this event a success. As you will see, WA21 will offer some new options in our Conferences, where we will include high level panel sessions that will add to our traditional format. We are looking forward to listen your opinion about it.

Finally, I would like to thank the Government of Yucatan for all the support received through the SEPASY and the Bureau of Tourism, and to COMEPESCA, CANAINPESCA, INAPESCA and SOMEXACUA for their partnership. Also, I would like to congratulate again our Home Office, and the Steering, Program and National Committees for their hard work in making WA21 what I am sure will be an unforgettable event.

Enjoy World Aquaculture 2021!

Antonio Garza de Yta
World Aquaculture Society
President

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World Aquaculture 2021

ABSTRACTS

LESSONS FROM CHILE'S ARTISANAL FISHERS AND SMALL-SCALE AQUACULTURE FARMERS AS THEY CONFRONT CLIMATE CHANGE

Dr. José Aguilar-Manjarrez

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FAO Regional Office for Latin America and the Caribbean

Climate change is a reality for the entire planet, and Chile is no exception. The country has a high degree of vulnerability to climate change, and while many productive sectors face profoundly impacted conditions, artisanal fishers and small-scale fish farmers are among the most severely affected because of both their geographical locations and their economic status. Thus, actions that support and promote the adaptation of this sector are necessary to address climate change and other related issues.

With funding from the Global Environment Facility (GEF), the Food and Agriculture Organization of the United Nations (FAO), Chile's Undersecretariat of Fisheries and Aquaculture and the Ministry of the Environment, conducted a project, from 2017 to 2021 entitled, "Strengthening the Adaptive Capacity to Climate Change in the Fisheries and Aquaculture Sector", to help reduce the vulnerability of four coves and their communities.

Primary results and innovative aspects of the project are presented in keeping with its three main components: strengthening of public and private institutional capacities; improvement of the adaptive capacity of artisanal fisheries and small-scale aquaculture; and strengthening knowledge and awareness about climate change in fishing and aquaculture communities. It also details the lessons learned during the project, with the goal of providing the authorities and communities involved in the fisheries and aquaculture sector with the capacity and tools required to adapt to future climate scenarios. Recommendations are made for scaling-up adaptation actions in the pilot coves and making this initiative sustainable beyond the end of the project.

EFFECT OF WATER SALINITY ON THE OXIDATIVE SYSTEM OF JUVENILES OF THE NORTH ATLANTIC WHITE SHRIMP *Litopenaeus setiferus* REARED IN BIOFLOC TECHNOLOGY

Diana Aguilera-Rivera*, Manuel Valenzuela-Jiménez, Claudia Durruty-Lagunes, Gerard Cuzon, Eduardo Pacheco, Miguel Arévalo, Wilson Wasielesky, Gabriela Rodríguez-Fuentes, Álvaro Barreto, and Gabriela Gaxiola

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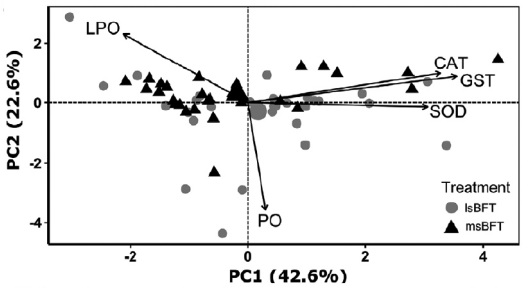
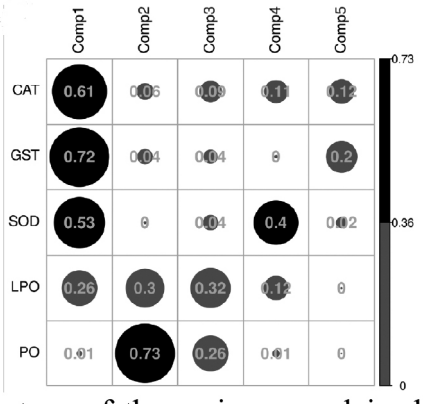
Litopenaeus setiferus is a native species of the North Atlantic; it shows good survival in seawater or in low salinity in clear water systems. However, its reduction in the natural population puts it at risk from an ecological viewpoint not only for its capture but also for its conservation. Biofloc have proven to be a sustainable alternative for other native shrimp species (Emerenciano et al., 2012; Magaña-Gallegos et al., 2018). Therefore, the aim of this study was to evaluate the effect of biofloc at high and low salinity on the antioxidant activity, and oxidative damage of *L. setiferus*.

L. setiferus postlarvae were separated into two groups: low and high salinity (5 and 35 psu, respectively). Shrimp were distributed into six fiberglass tanks and acclimated for 5 days. For biofloc stimulation in the experimental tanks, a volume of 1,000 L biofloc inoculum was used and filled at full capacity with filtered clear water. Three replicates were established and monitored for 90 days. At the end of the experiment, 15 shrimp for each treatment were dissected to obtain a portion of muscle tissue for further antioxidant (CAT, SOD, GST) and oxidative damage (PO, LPO) analysis. The contribution of salinity conditions on these parameters were identified by PCA analysis, supporting the tendency obtained by UPGMC to visualize the grouping for each treatment.

Results suggest that biofloc can stabilize and improve conditions for *L. setiferus*. During the experimental phase, oxidative stress was stimulated, which increased cell damage at low salinity due to the high oxygen consumption under fasting and postprandial conditions.



PCA for antioxidant enzyme activity (PC1) and oxidative damage (PC2) in muscle tissue of *L. setiferus* postlarvae.



Percentage of the variance explained by each component for both treatments (lsBFT and hsBFT).

A VIBRIOSIS OUTBREAK IN THE PACIFIC WHITE SHRIMP, *Litopenaeus vannamei* REARED IN BIOFLOC AND CLEAR SEAWATER

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Biofloc possess a probiotic effect and has a role on mechanisms related to the immune response in shrimp (Aguilera Rivera et al., 2018, 2014;). We reported a vibriosis outbreak in a biofloc and clear seawater rearing system. To treat it, oxytetracycline was administered only in clear seawater tanks, but the results were not successful. To understand the mechanisms related to the susceptibility or resistance to *Vibrio* of *L. vannamei* juveniles reared in biofloc or clear seawater during a vibriosis outbreak, we isolated and identified *Vibrio* species, and expression of pathogenicity and oxytetracycline resistance-related genes of *Vibrio* strains.

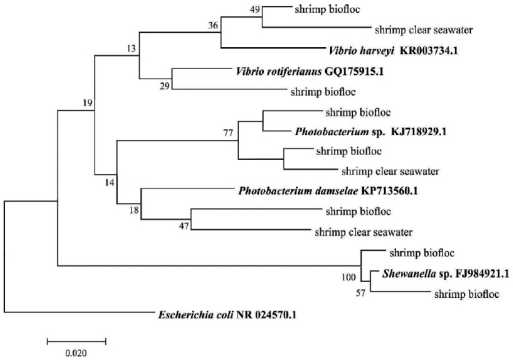
Juvenile *L. vannamei* previously reared in biofloc (13.98 ± 2.94 g) and clear seawater (11.34 ± 1.54 g) presented mortalities (25 and 80%, respectively). Moderate and severe signs of disease were observed in shrimp reared in clear seawater, where a treatment with oxytetracycline was applied in the feed with only occasionally positive results. Shrimp reared in biofloc were observed to have less multifocal melanization in the exoskeleton and redness in appendages. These signs disappeared without the addition of antibiotics to treat the possible infection.

Bacterial isolation and molecular identification of strains were performed. *V. harveyi, V. rotiferianus, Shewanella* sp., *Photobacterium* sp. and P. *damselae* were identified for both rearing systems. Only *Shewanella* sp. was identified in shrimp reared under biofloc conditions.

Presence of pathogenicity (toxR) and oxytetracycline resistance (tet(A), tet(B), tet(C), tet(D), tet(E), tet(G)) genes were identified in five strains previously isolated from hepatopancreas of shrimp *L. vannamei* reared in biofloc and clear seawater system.

Although the occurrence of vibriosis in shrimp reared in biofloc is possible, the microbial dynamics in the system help to control the development of bacterial diseases produced by *Vibrio*. In comparison to the clear seawater rearing system, the presence of *Shewanella* sp. only in biofloc could be related to less severe lesions in shrimp. Although oxytetracycline was used for the treatment of vibriosis in the clear seawater system, expression of tet(B), tet(C) and tet(D) genes confirmed the inefficiency of the antibiotic we observed by the presence of disease signs and mortality in shrimp.

Bacterial strains isolated from hepatopancreas of shrimp *L. vannamei* reared in biofloc and clear seawater system.



CO-ENCAPSULATION OF *Bacillus subtilis* AND β-GLUCAN IMPROVED PROBIOTIC VIABILITY UNDER SIMULATED TILAPIA GASTROINTESTINAL CONDITIONS

Angélica P. C. Alves*, Amanda C. Alves, Brunno S. Cerozi, Rodney A. F. Rodrigues, José Eurico P. Cyrino

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The use of probiotic bacteria in fish feed is of increasing interest and microencapsulation technology can be used to maintain the viability of probiotic bacteria during processing and storage. In this study oat β-glucan (70%) was used for co-encapsulating probiotic *Bacillus subtilis* on the alginate matrix. The spray drying technique was used to produce the microcapsules. Simulated gastric fluids (SGF) of Nile tilapia were prepared using saline solution, adjusted pH to 1.5, using HCl solution. Exactly 0.5 g of *B. subtilis* microcapsules and 0.5 mL of free *B. subtilis* were placed into separated test tubes containing 4.5 mL of SGF. One mL of each sample was taken after incubation at 25 °C in a water bath for 0, 1, 2, and 3 h and enumerated for the cell counts on LB agar using a pour- plate method (Figure 1). For bile tolerance determination, 0.5 g of B. subtilis microcapsules and 0.5 mL of B. subtilis free cells were added into 4.5 mL of SGF at pH 1.5 and incubated at 25 °C for 1 h. After the incubation, the samples were then centrifuged. Then, SGF was removed and replaced with 4.5 mL of tilapia bile salt (10%). One mL of each sample was taken after further incubations of 1, 2, and 3 h at 25 °C and enumerated on LB agar using a pour-plate method to determine for the number of viable cells (Figure 2). Results were subjected to ANOVA and Tukey's test ($p<0.05$).

The combination of β-glucan with sodium alginate improved viability of *Bacillus subtilis* under simulated tilapia gastrointestinal conditions. The spray dried *B. subtilis* with β-glucan could be incorporated into the feed ingredients to be pelleted or after feed pellet grading through a coating process, providing valuable effects to the fish.

Fig. 1. Cell viability of free and encapsulated B. subtilis under simulated gastric condition.

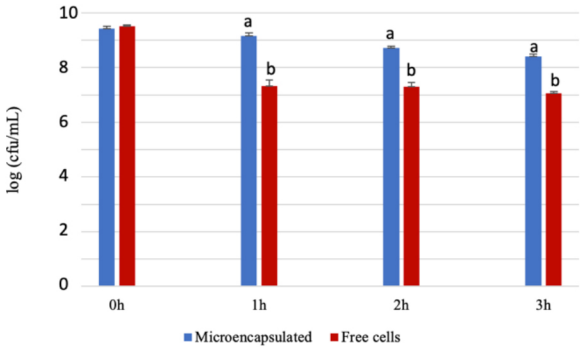
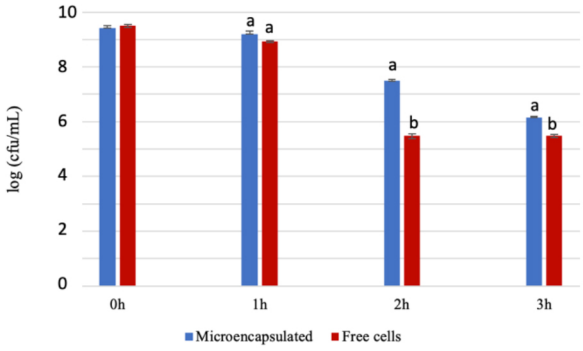


Fig. 2. Cell viability of free and encapsulated B. subtilis in tilapia bile.



ASSOCIATIVITY TO INNOVATE IN AQUACULTURE'S PRESENT AND FUTURE CHALLENGES. THE CHILE CASE

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The aquaculture industry in Chile is widely dominated by salmon farming, which is currently producing more than 1 million t and 5,000 million USD export value, representing around 90% of total Chilean aquaculture production. The salmon industry was a macro innovation in the south of the country, taking advantage of a market opportunity, comparative advantages, and a rapid process of technology transfer and adaptation. Rapidly, a cluster formed around the original core of producers to replace costly imports of goods and to develop national services as well as R+D+I capacities (Fig.1). Producers, suppliers, academia, and government agencies all shared a common origin and location, which facilitated active communication and interaction among them, bolstering innovation.

Challenges and crises also contributed to the development of national capacities and solutions, with the ISA crisis between 2007 and 2010 reshaping the production model, reinforcing R+D+I, and emphasizing the importance of collaboration in facing current and future challenges. Technology bloomed as well as R+D centers and the number of technology - based suppliers increased. Following that trend, at the end of 2019, the aquaculture innovation club was created to reinforce individual and collaborative innovation as a tool to generate solutions to complex present and future challenges in the industry.

At present, several initiatives in the aquaculture innovation ecosystem have been developed, contributing to the shared objective of building a competitive as well as sustainable industry as well as actively exporting goods and services to other countries.

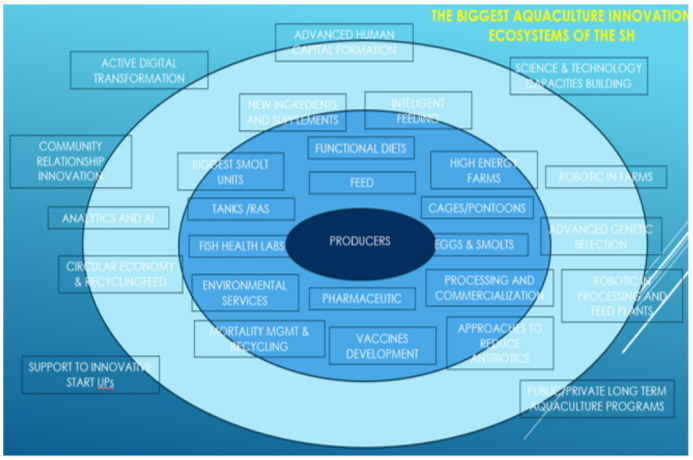

Figure 1. The aquaculture innovation ecosystem in Chile.

EVALUATION OF GROUNDWATER IN THE COASTAL PORTION OF GUASAVE, SINALOA FOR WHITE SHRIMP FARMING (*Penaeus vannamei*) THROUGH VES, CHEMICAL COMPOSITION, AND SURVIVAL TESTS

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We studied the potential of white shrimp farming with groundwater from the coastal aquifer of Guasave, Sinaloa. The use of water from the aquifer in aquaculture represents challenges due variability in quality of groundwater. We did twenty-three vertical electrical soundings (VES), performed to guide the search for continental groundwater, obtaining the resistivity of the saturated formation (Ro) and a relationship with resistivity of aquifer water of Rw=0.4478 Ro + 0.8371. We obtained ionic content and nutrients from shrimp farming water. Also, positive corre-lation was found between the electrical conductivity of the aquifer water (inverse of Rw) with chlorine, sodium, magnesium and calcium ions in 34 water samples. The analysis of ions and ammonia-nitrogen, nitrates, phosphates, potassium, manganese and calcium were used to select suitable sites to perform in two shorts bioassays: natural aquifer water and adding KCl and Mg2Cl to simulate diluted seawater. In most natural waters, survival of larvae was higher than 60% and in simulated seawater survival improved only in two sites. Building Rw-Ro relation allowed to infer the quality and suitability of water and positioning the most suitable place for drilling. Final-ly, Rw relation with dissolved ions, allows to estimate aquifer water quality and reduce uncertainty.

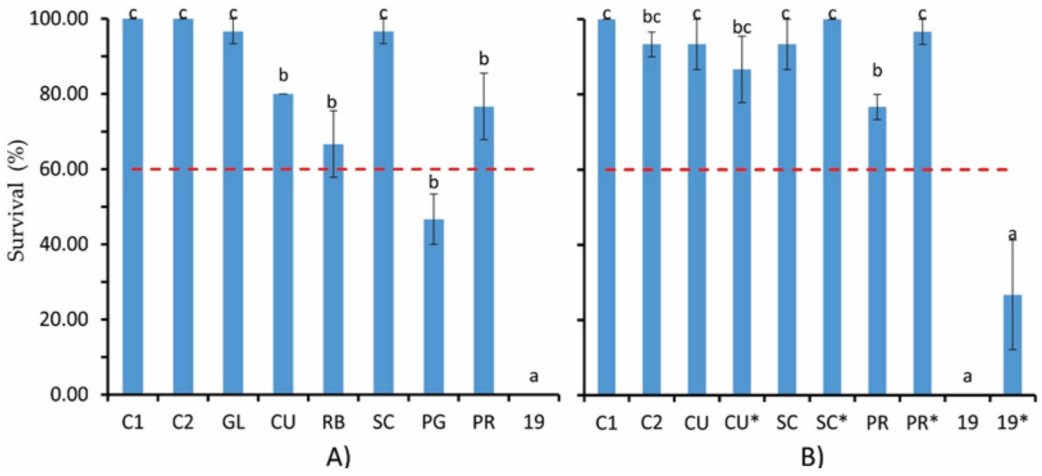

Figure 1. Survival of P. vannamei during the first 48 hours, the letters indicate a significant differ-ence p <0.05 (* indicates that salts were added) A) Survival of P. vannamei PL20 B) Survival of P. vannamei (30.40 + 2.42 mg).

"EVALUATION OF THE BIOLOGICAL-PRODUCTIVE AND ECONOMIC PERFORMANCE OF WHITE SHRIMP *Penaeus Vannamei* IN FLOATING CAGES IN AN INTENSIVE SYSTEM"

Juan Pablo Apún-Molina*, Jorge Herrera Olmos, Juan Carlos Sainz Hernandez Apolinar Santamaria Miranda and Leonardo R. Palacios

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The evaluation of the biological-productive and economic performance of two varieties the white shrimp Penaeus vannamei in floating cages in an intensive system". Similarly, in this system no electrical energy and/or fuels are used during cultivation; In addition, the use of antibiotics is completely eliminated, these and other advantages compared to traditional crops. The present work shows that, in financial and economic terms, a floating cage system requires less investment, reduces operating costs and, together with the commercialization of juvenile P. vannamei as bait, becomes an innovative, sustainable and sustainable business.

With this production strategy, no investment is made in pumping water, since this resource is renewed daily with the movement of water through the network caused by the flow of the tides; at the same time, small organisms enter the cage and are ingested by the cultured shrimp, which contributes to lowering the feed conversion factor.

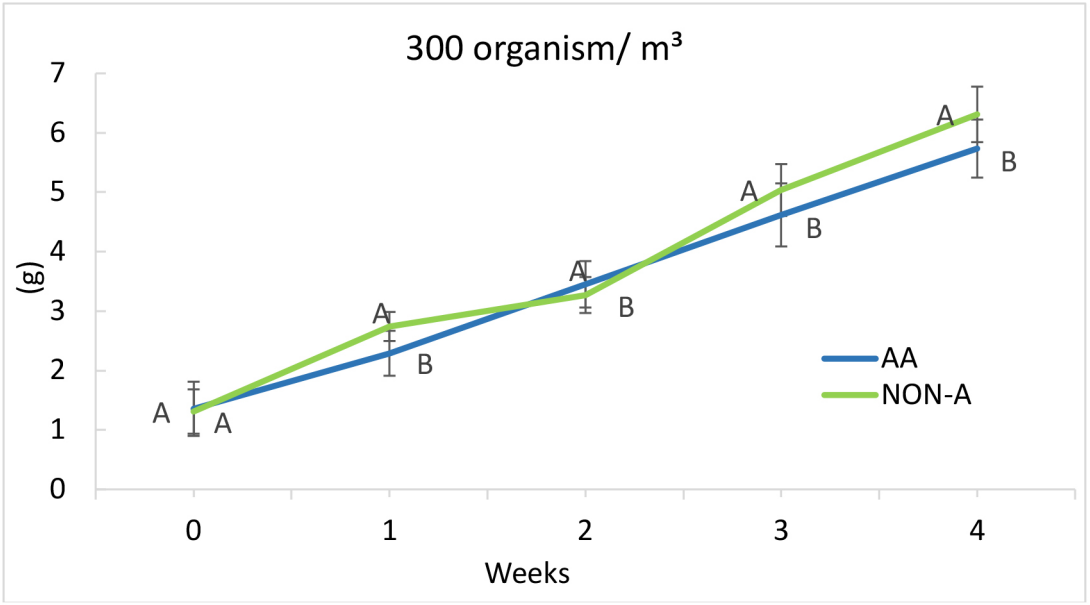

Fig.-1Weekly growth

IN-POND RACEWAYS SYSTEM, (IPRS) EXPERIENCES IN LATAM

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As the world population increase, and overall steady world fisheries captures, the urgent demand of aquaculture production is eminent. Year increase of World aquaculture products (FAO 2020) decline from more than 6% on years (2001-2005) to 4% (2016-2018). However, countries like Mexico and Colombia show an increase of 9% on the latest years. Unite States catfish industry has decline since 2005 (Hanson and Site,2015) due to imports of pangasius to US markets. IPRS technology born over the necessity to increase production, economically efficient. IPRS is not new, it shows up on the late 90 under direction of Mike Masser, but with a low-key technology. Dr. Chappell redesigned this technology, creating a water flow average 10 cm/s. this create a river inside a pond exposing nitrogen derivates to nitrification bacteria and designing a solid settling area where solid can be taken out of the aquatic environment. This latest feature not only improves water quality but also increase carrying capacity as well as using same water for years. Production data collected over several countries in LATAM shows that fish producer yield an average of 45,000 kg/Ha/Year in two crops. IPRS technology can increase yield at an average of 96,946 kg/Ha/year in three crops, and in many cases more than that with an average ROI % of 39.2

Tilapia Production Resumen on IPRS in America Latina

Granja	Produccion Kg/Ha/año	FCR	Gr/Dia	Kg/m³	Rw/m³	Alimento kg/Ha/Dia	Sobrevivencia %	ROI %
Campeche, Mexico	60,756	1.34	4.40	50	150	253	81	38.6
Chetumal, QR, Mexico	132,676	1.20	5.43	72	163	388	90	65.0
		1.30		96	163			
Veracruz, Mexico[3]	139,700	1.28	3.82	60	275	800	82	
Huila, Colombia[3]	76,000	1.30	3.78	39	275	683	88	
Cartagena, Col	75,600	1.29	4.34	64	154	450	83	30.0
Promedio	96,946	1.29	4.35	63			85	39.2
Santa Rosa de Copan, Honduras[1]	119,267	1.41	2.62	87	14.5	1075	87	44.0
Tilapia Blukalsa, Honduras[1]	40,783	1.53		28	175		84	32.9
Raceways Flotantes, Auburn, AL[2]	16,236	1.60	2.58	102	63.4		80	24.4
	13,720		2.69	120	45.4		90	

1= Tilapia Roja
2= Bagre de Canal
3= Publicacion en progreso

EFECTOS DEL PROPIONATO DE SODIO DIETARIO EN EL CRECIMIENTO, ACTIVIDAD ENZIMÁTICA DIGESTIVA Y EXPRESIÓN DE GENES DEL SISTEMA INMUNE EN JUVENILES DE PEJELAGARTO *Atractosteus tropicus*

Jesús G. Arellano-Carrasco *, Rafael Martínez-García, Alberto Asiain-Hoyos, Juan L. Reta-Mendiola, Pablo Díaz-Rivera, Susana A. Frías-Gómez, Talhia Martínez-Burguete, Reyes González-Salvador, Emyr S. Peña-Marín, Gloria Asencio-Alcudia, Carlos A. Álvarez-González

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Introducción. El pejelagarto es una especie con una marcada importancia económica, ecológica y cultural en el sureste de México. La correcta alimentación en el cultivo de especies acuáticas tiene un papel importante en la capacidad de asimilación de nutrientes en los organismos, lo que determina el estado de salud de los peces y con ello, la capacidad de respuesta ante posibles enfermedades. Se ha estudiado en el pejelagarto los efectos de diversos aditivos, prebióticos y probióticos en su capacidad digestiva. El presente estudio buscó analizar los efectos del propionato de sodio sobre diferentes variables adicionado a dietas balanceadas para juveniles de *A. tropicus*.

Materiales y método. Se evaluaron 4 tratamientos con la inclusión de Propionato de Sodio (PS) en diferentes concentraciones (0.5, 1.0, 1.5 y 2%) y una dieta control (0%). Los peces fueron alimentados 4 veces al día, y se tomaron datos biométricos cada 15 días. Al final del experimento se realizaron homogenados de estómago e intestino por separado para determinar la actividad enzimática digestiva. Se realizó un análisis de expresión de genes en intestino e hígado mediante qPCR, los genes analizados fueron ocludin (ocln), dominio de oligomerización por unión de nucleótidos (nod2) y mucina (mucc2). Una vez comprobados los supuestos de normalidad y homocedasticidad se realizó un ANOVA de una vía con una significancia de 0.05, utilizando el software GraphPad Prism v.8.0.2.

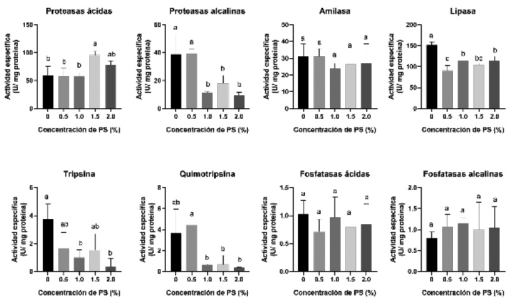

Figura 1. Actividad enzimática digestiva de juveniles de A. tropicus. Letras diferentes muestran diferencias significativas (p<0.05).

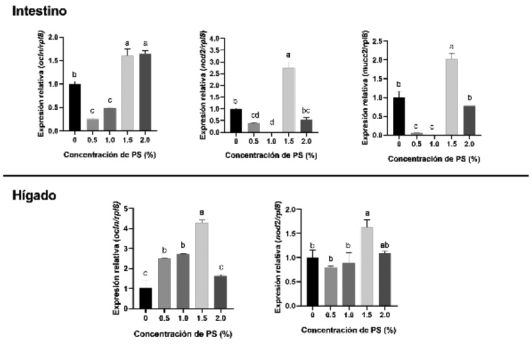

Figura 2. Expresión relativa de ocln, nod2 y mucc2 en juveniles de A. tropicus en diferentes órganos. Letras diferentes muestran diferencias significativas (p<0.05).

Resultados. Los peces alimentados con 0.5% y 1.5% de PS mostraron el mejor promedio en cuanto al peso (25.79 ± 8.73 g) y talla final (17.83 ± 2.42 cm) respectivamente. La mayor sobrevivencia ocurrió en los tratamientos 1.5 y 2 % de PS ambas con 92%. El tratamiento 1.5% PS mostró mayor actividad de la enzima proteasa ácida, mientas que el tratamiento 0.5% tuvo mayor actividad para proteasas alcalinas y quimotripsina, mostrando diferencias significativas respecto a los demás tratamientos (Figura 1). La inclusión de 1.5% PS promovió un incremento en la expresión de los genes *ocln, nod2 y mucc2* en intestino, y *ocln* y *nod2 en hígado* (Figura 2).

Conclusión. La inclusión de propionato de sodio en las dietas balanceadas para pejelagarto no es completamente determinante en el crecimiento y la actividad enzimática digestiva. Sin embargo, la mayor supervivencia se encontró en las concentraciones más altas, sugiriendo que el PS fortalece la barrera intestinal de dicha especie mejorando su sistema inmune.

RESTORATION OF THE BIODIVERSITY OF CORAL REEF FISH ASSEMBLAGES IN THE MEXICAN CARIBBEAN

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Given the current effects of global environmental change on marine ecosystems, tools to support coral reef restoration have been substantially diversified. Restoration efforts have focused primarily on the recovery of coral species using different techniques. Harnessing restoration to achieve ecological rehabilitation of the ecosystem needs innovative methods that include the restoration of coral reef fish assemblages, contributing to the conservation of biodiversity and ecosystem functions and services.

Here we highlight a multidisciplinary engage approach necessary to establish a strong partnership among the science, public and private sector. We conducted field studies in the northern part of the State of Quintana Roo, Mexican Caribbean to analyze for the first-time possibility of using the capture and aquarium-culture of post-larvae fish species and releasing the juveniles as a tool for the potential recovery of reef biodiversity resilience. We tested the potential of post-larvae capture using night light traps. We collected 748 post-larvae reef fishes from 8 orders, 20 families, and 40 species. Acanthuridae, Pomacentridae, Monacanthidae, and Tetraodontidae comprised the highest species number of post-larvae families. We also explored pilot release experiment *Stegastes partitus* and a landscape analysis across the Mexican Caribbean as a potential tool to determine appropriate reef sites to release the cultured juveniles and for adding an ecological planning. The results of the pilot release experiment with *S. partitus*, showed that there is a positive effect in survivorship during the capture and release procedures into suitable habitat. Both technique approaches allow sustainable capture and culture of post-larvae fish species and can be combined with different techniques of conditioning and release to supply target reef sites. Using this method in fish-scarce coral reefs with vulnerable food webs may be helpful to test the feasibility and potential ecological, fisheries and aquaculture application.

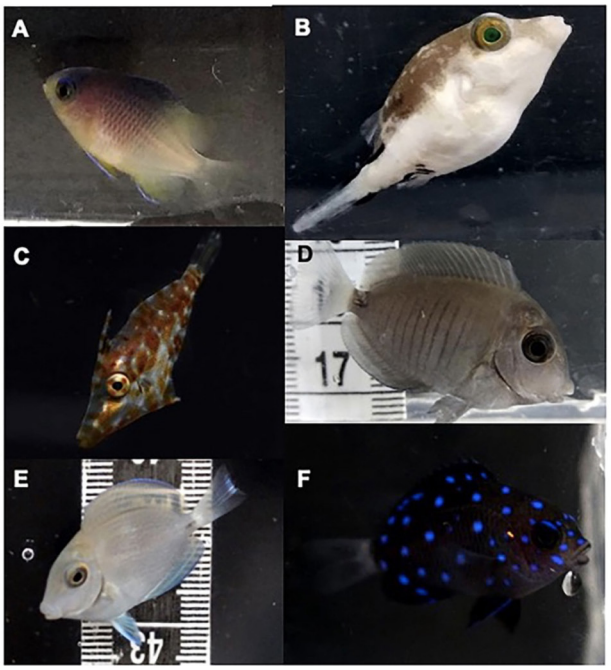

Figure. Some of the most abundant species recorded in the area include the following: **(A)***Stegastes partitus*; **(B)** *Canthigaster rostrata;* **(C)** *Monacanthus tuckeri*, **(D)** *Acanthurus chirurgus*, **(E)** *Acanthurus tractus*, and **(F)** *Mycrospathodon chrysurus*.

THERAPEUTIC TREATMENT AGAINST OPPORTUNISTIC COPEPODS AFFECTING CULTURED SEA CUCUMBER *Isostichopus badionotus*

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During the culture of *Isostichopus badionotus* in tanks at Telchac, Yucatán, we found high abundance of harpacticoid copepods producing skin lesions. The wounds were colonized by opportunistic pathogens such as bacteria, which in turn led to Skin Ulceration Disease (SUD). Organophosphate pesticides such as trichlorfon have been used in concentrations between 1-5 ppm for up to 12 h, to control copepods in sea cucumber aquaculture, followed by a comprehensive water exchange to eliminate the compound from the farming system. However, trichlorfon has been banned in Mexico since 2019.

An alternative compound could be Temephos, an organophosphate endorsed by the World Health Organization (WHO) to control the freshwater copepod *Cyclops vernalis*, an intermediate host of the nematode *Dracunculus medinensis*. Therefore, our aims were to determine the Lethal Dose 50 (LD_{50}) and 99 (LD_{99}) of the organophosphate temephos to control the opportunistic harpacticoid copepods as well as to calculate the therapeutic margin of this compound for *I. badionotus*.

We have identified five different copepod morphotypes and the taxonomic specific identification is on its way. The toxic effect of temephos was evaluated through 96h static bioassays using stage 5 copepodites and adult copepods. After performing a range detection test, 0.062, 0.125, 0.250, 0.500, 0.100, and 2.000 mg/L were determined as the final bioassay doses and were evaluated for 24 and 48 h of exposition. Preliminary results suggest that temephos LD_{50} and LD_{99} at 48 h exposure are 0.312 and 0.715 mg/L respectively. Tests are still in progress to obtain the LD_{50} and LD_{99} of temephos at 24 h, and to calculate the therapeutic margin of this compound for *I. badionotus*.

DIATOM *Odontella aurita,* AN ALTERNATIVE FOOD SUPPLEMENT FOR THE PREVENTION OF CARDIOVASCULAR DISEASES AND OBESITY IN THE MEXICAN POPULATION

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Modern human lifestyle is based on high calorie and excessive red meat consumption, and limited or no physical activity that put their health at risk. In 2018, 75.2% of the Mexican adult population was either overweight or obese. Between 2000 and 2015, deaths associated to this problem increased: cancer (34.4%), osteoarthritis (39%), cardiovascular diseases (70%) and diabetes mellitus (111.3%). The diatom *Odontella aurita*, native to Baja California Sur (BCS), identified and curated at the Northwest Biological Research Center (CIBNOR) Microalgae Collection, is been proposed as a food supplement to help reduce this diseases. *O. aurita* shows high fucoxanthin and eicosapentaenoic acid content, both of which have demonstrated a synergic effect against metabolic syndrome, hyperlipidemia, oxidative stress and inhibition of cellular cancer lines. From 2019, the international market for microalgae has grown at a 6% rate with expectations of that trend to continue to 2027. This represents $56.5 billion dollars. CIBNOR has significant experience in the production of consistent quality marine microalgae and the technology transfer process. The biotechnological potential of *O. aurita* is based on the high quality nutritious compounds that have direct benefit for the human health, resulting in a compelling product for a market in need of sustainable alternatives. Production is ideal in the climate and geography of Baja California Sur. A strategic alliance through a licensing model for the technology transfer is proposed, where technical assistance is tailored to the needs of the client.

DIATOMEA *Odontella aurita*, SUPLEMENTO ALIMENTICIO COMO ALTERNATIVA PARA LA PREVENCIÓN DE ENFERMEDADES CARDIOVASCULARES Y OBESIDAD EN LA POBLACIÓN MEXICANA

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El estilo de vida del hombre está basado en un alto consumo de calorías, excesiva ingesta de carnes rojas, baja o nula actividad física, poniendo en riesgo su salud. En el 2018, el 75.2% de la población de adultos en México, presentaron sobrepeso y obesidad. Entre el 2000 y 2015, se incrementaron las muertes asociadas a este problema: cáncer (34.4%), osteoartritis (39%), cardiovasculares (70%) y diabetes mellitus (111.3%). La diatomea *Odontella aurita*, nativa en Baja California Sur (BCS), depositada en la Colección de Microalgas del CIBNOR, se propone como suplemento alimenticio para estas enfermedades, entre otras. *O. aurita* presenta alto contenido de fucoxantina y de ácido eicosapentaenoico, que han demostrado un efecto sinérgico contra el síndrome metabólico, hiperlipidemia, estrés oxidativo e inhibición de líneas celulares de cáncer. Desde el 2019, el mercado internacional de microalgas ha crecido anualmente 6%, y se espera que siga así hasta el 2027 representado por $56.5 mil mdd. El CIBNOR tiene amplia experiencia en la producción de microalgas marinas y en transferencia de tecnología. El potencial biotecnológico de *O. aurita* radica en los compuestos de alta calidad nutritiva que tienen beneficios en la salud del hombre, esto constituye una alternativa sostenible y viable de comercialización ante los retos de desarrollo para el país. Su producción en las condiciones climáticas y geográficas de BCS, son idóneas para impulsar una industria biotecnológica de biorrefinería de microalgas, aún incipiente a nivel nacional. Se propone un licenciamiento, sobre el método de producción y asistencia técnica mediante una alianza estratégica.

GENETIC OVERVIEW TO REVEAL HOW THE CAPTIVE POPULATION WAS, IS, AND SHOULD BE MANAGED

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Genetic Overview (GO) is a compelling service to characterize populations in captivity that are usually managed for commercial purposes. Using a set of software tools and calculations the GO report describes the levels of diversity, consanguinity, and the genetic structure of the sampled population. These proprietary techniques of the Center of Aquaculture Technologies make use of all types of Single Nucleotide Polymorphisms (SNP) arrays: Low Density (LD), Medium Density (MD) and High Density (HD). These arrays are now available for a variety of aquatic species, including shrimp (*Litopenaeus vannamei*), oysters (*Crassostrea gigas* and *Crassostrea virginica*), and fish (salmon; (*Salmo salar),* Red snapper (*Lutjanus peru),* cobia (*Rachycentron canadum),* yellowtail (*Seriola rivoliana, Seriola lalandi),* halibut (*Hippoglossus hippoglossus),* sable fish (*Anoplopoma fimbria*), among others. The option of design and customization of genomic tools for non-traditional aquaculture species is also available.

The present work describes the parameters of GO used to characterize these cultured stocks and its biological meaning. To protect the confidentiality, synthetic (simulated) data is used for this purpose. Four fictitious breeding lines; AA, BB, CC, and DD are compared to show the meaning of diversity parameters such as the percent of polymorphic loci, the heterozygosity (observed and expected) and the Nei's diversity index. The extent of consanguinity is assessed by pairwise comparisons of the relatedness coefficient (r). Finally, the structure of the breeding lines based on their differentiation or similarity is explained using the interspecific variation coefficient (Fst), the Principal Coordinates Analysis (PCoA) and the Structure plots.

Some of the applications of this type of analysis will be further summarized. It is particularly powerful in monitoring genetic health over time in breeding programs based on mass selection but is broadly applicable to all programs. In general terms, the Genetic Overview is a snapshot of the current state of the population genetic characteristics. It is highly recommended at the beginning of the breeding programs of all types to understand the level of diversity and relatedness, and the history that the DNA reveals about previous management and origin of the lines, this helping to device sound breeding strategies. Adopting GO into the hatchery practice will help to evaluate the impact of selection and assess the genetic quality of the lines, guide the mating decisions to maximize diversity and decrease the rate of inbreeding/relatedness, and consequently provide an evaluation of the genetic management through the production process.

MICROBIOLOGICAL DIVERSITY AND COMPOSITION OF WATER AND SEDIMENTS IN THREE FARMS FROM ECUADOR

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It is understood that there is an association between productivity and microbial diversity in aquaculture systems. For example, changes in bacterial microbiomes are implicated in animal performance, in disease development associated with both bacterial and viral origin, and in dysbiosis (disequilibrium in microbial communities) triggered by environmental stressors or diet choice. With the increasing development of novel DNA sequencing technologies, new concepts and cost-effective applications have emerged to better understand the role of microbial communities in the growth and health of farmed vertebrates and invertebrates. Evidence of any of the three main mechanisms that lead to dysbiosis, such as the decrease in diversity, the loss of beneficial bacteria and the expansion of pathogens or potentially harmful microorganisms, can be used as an indicator tool for productivity monitoring/forecasting purposes. This work establishes a methodology and a pipeline for microbiome characterization in three commercial shrimp ponds in Ecuador. Also, we compare the merit of two types of samples (water and sediment) and detect patterns that allow inference on water quality in the ponds and evaluation of productive performance of the shrimp.

Briefly, the samples were filtered to concentrate, remove debris, and capture microbes present. DNA was extracted from the concentrated microbes sequenced to look at the type of bacteria present and the relative amount of each type. This was successful, with clear profiles produced along with some information on pathogen presence. Alpha diversity is a measurement of the microbial diversity of each sample. All sediment samples from the three ponds were very diverse with alpha diversity scores of over 600. Water samples were markedly less diverse with alpha diversity scores generally less than 250. For one of the ponds, alpha diversity was relatively high and over 250. For another pond alpha diversity was less than 100, and a single bacterial species made up over 50% of the bacteria detected in all water samples. In addition, for some water samples potentially pathogenic Vibrio species were identified.

Overall, this information reveals that monitoring the microbiome of production ponds over a grow-out cycle has the real chance to deliver data meaningful for pond management and pond performance. Possible application of this technology/analysis are 1) assessing genotype by environment effects in shrimp performance, 2) understanding of the impact of the microbes in the growth, development, and survival of shrimp, 3) evaluation of the effectiveness of probiotics or diets, and 4) predicting pond performance by evaluation of variations in microbial composition at different timepoints in the production cycle.

EL CULTIVO DE LANGOSTINO MALAYO EN MÉXICO: UNA SEGUNDA OPORTUNIDAD PARA *Macrobrachium rosenbergii*

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El langostino malayo *Macrobrachium rosenbergii* se introdujo a México a principios de la década de los setentas y durante varias décadas se promovió su cultivo a través de diferentes dependencias gubernamentales, como una manera de diversificar la oferta acuícola nacional. Aunque la academia y el sector privado se involucraron, las cifras oficiales muestran que únicamente se registró producción durante el periodo que va de 1995 a 2016. Durante esas dos décadas, los volúmenes de producción reportados nunca lograron superar las 300 toneladas anuales. Pero algo ocurrió después y la especie dejó de producirse. La evidencia sugiere que los pocos criaderos del gobierno y privados dejaron de producir y ofertar postlarvas, lo que ocasionó el colapso de la incipiente industria.

El escenario nacional ha cambiado. Las especies nativas de *Macrobrachium* que tradicionalmente soportaban pesquerías artesanales en México (*M. americanum* y *M. tenellum*, en el litoral del Pacífico; *M. carcinus* y *M. acanthurus*, en el litoral del Atlántico) han disminuido drásticamente, principalmente por sobreexplotación y destrucción de su hábitat. Actualmente, los precios de estos crustáceos dulceacuícolas en el mercado nacional son muy elevados y la creciente demanda está siendo atendida por producto importado, principalmente de Asia.

Con la intención de rescatar el cultivo del langostino malayo en México, se colectó material biológico de algunas granjas en el sureste del país, remanente de los lotes que fueron diseminados para su engorda por los últimos laboratorios que estuvieron en funcionamiento. Los organismos fueron trasladados a Veracruz para seleccionar reproductores. A partir de las técnicas convencionales de producción de postlarvas a nivel internacional, se adaptaron y evaluaron sistemas de producción que fueran fácilmente adoptados por pequeños productores. Mediante alianzas público – privadas, se ha promovido el establecimiento de nuevos laboratorios de producción de postlarvas, principalmente en el sureste del país, que han comenzado a diseminar la semilla entre productores interesados. En la actualidad existe en ese territorio una enorme cantidad de infraestructura para cultivo de tilapia, que tiene el potencial de ser utilizada como policultivo con langostino, para gradualmente volver a posicionar la especie como una alternativa rentable, que al mismo tiempo ayude a disminuir la presión sobre los recursos nativos.

TROPICAL SEAWEED CULTIVATION IN RÍA LAGARTOS, YUCATÁN, MEXICO: ALTERNATIVES FOR THE FISHERIES SECTOR

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Seaweed farming is a global business that is carried out in at least 56 countries around the world, although more than 99% of production is in Asia. In 2018, OECD countries accounted for more than 60% of the total export value (US$1 billion) of seaweed traded for direct human consumption or as raw material. European countries accounted for almost half of the value of global imports (USD 1.3 billion) of seaweed-based thickeners. Currently, algal biomass produced by aquaculture has expanded with a production of up to 32.4 million tons (FAO, 2020). In Mexico, algal exploitation has developed mainly in Baja California from natural populations, and recent aquaculture efforts have been successful. In contrast, on the Yucatan coast, even with the diversity of algae found, there is no current exploitation. Preliminary experiences showed that the Yucatan peninsula is ideal for algal culture; during 2002 the pilot culture of *Kappaphycus alvarezii* was successful in Dzilam de Bravo, with growth rates of up to 6.5 ± 1 % day^{-1} (Robledo et al., 2013). Recent experiences under Integrated Multitrophic Aquaculture cultivation have shown great potential for *Solieria filiformis* and *Halymenia floresii*.

Currently, SEPASY- Secretaría de Pesca y Acuacultura Sustentable de Yucatán is supporting the fisheries sector through the development of aquaculture, including seaweed culture. This study is part of that effort, in which an estuarine area was evaluated for the cultivation of algae of potential interest in Ría Lagartos, in collaboration with the fishermen's cooperative "Pepineros de Río Lagartos". Vegetative specimens of native red algae, selected from wild populations and acclimatized strains, were cultivated in an experimental oyster farm (Table 1, Fig. 1). Outdoor culture systems use low-cost infrastructure under natural conditions; growth rates of each cultivar were determined, and environmental parameters were monitored during the study (2021-2022). The algae were subject to strong environmental changes, especially salinity and temperature, therefore growth rates were highly variable throughout the study period. Some species acclimated to estuarine conditions with positive growth rates (*Rhodymenia pseudopalmata*, *Gracilaria cervicornis* and *Crassiphycus corneus*), which varied by season and species. *Rhodymenia pseudopalmata* showed a growth rate of up to 8.0 ± 1 % day^{-1} during the summer of 2021, demonstrating a high potential for cultivation. Acclimatization and cultivar selection is crucial for successful cultivation and to evaluate different coastal zones to optimize cultivation.

Table 1. Species under evaluation in Ría Lagartos, Yucatán.

Species	Use
Crassiphycus corneus[1]	Agar; Aquaculture feeds
Eucheumatopsis isiformis[1]	Iota-carrageenan; Human consumption
Gracilaria cervicornis[1]	Agar; Aquaculture feeds
Gracilaria blodgettii[1]	Agar; Human consumption
Rhodymenia pseudoplamata[2]	Human consumption; Bioactives
Solieria filiformis[2]	Iota-carrageenan; Bioactives; Aquaculture feed

[1]Wild population; [2]Acclimated strains from outdoor tank cultivation.

Figure 1. Experimental cultivation site.

PROTECTIVE EFFECT OF THE ADRIATIC SEA MACROALGA *Codium adhaerens* AGAINST HYDROGEN PEROXIDE-INDUCED OXIDATIVE STRESS IN ZEBRAFISH *Danio rerio* EMBRYOS

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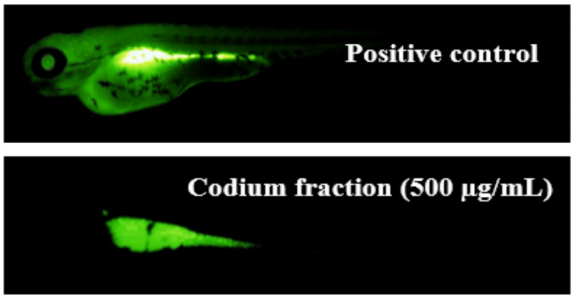
Marine macroalgae are an extremely valuable source of bioactive molecules with unique properties and multiple potential applications in the cosmeceutical, pharmaceutical, and food industries. Adriatic Sea is an extremely harsh environment characterized by high salinity, seasonal oscillations in temperature and relatively shallow depth which indicates increased UV radiation. It can be assumed that such conditions have forced macroalgae to evolutionary develop molecules which could through activation of anti-stress mechanisms enable surviving in such a hostile environment. With such a hypothesis, *Codium adhaerens* was sampled from the Adriatic Sea, extracted with methanol:dichloromethane and further fractionated by solid phase extraction to obtain less polar fractions (F3, F4). Chemical composition was determined by UHPLC-ESI-HRMS and the antioxidant potential was comprehensively evaluated using colorimetric assays (Folin-Ciocalteu, ABTS, DPPH, FRAP) and *in vivo* experiments on zebrafish *Danio rerio*.

Zebrafish *D. rerio* embryos were employed for this research, as currently one of the most valuable laboratory model organisms for bioactivity determination of natural and synthetic molecules. The results obtained from *in vivo* experiments on zebrafish embryos demonstrated protective effects of tested *C. adhaerens* fractions against H_2O_2 induced mortality (23% and 27% declined mortality in 500 µg/mL of F3 and F4 fractions), along with the significant decrease of reactive oxygen species (ROS) generation. F3 and F4 in concentration of 500 µg/mL significantly decreased ($p<0.001$) ROS generation (60.5 and 51.9 % decreased in comparison to H_2O_2 treatment group). The results obtained by *in vivo* assay on zebrafish embryos correlate well with the ones obtained by colorimetric methods since both *C. adhaerens* fractions (F3 and F4) exerted similar antioxidant activity responses. This can be explained by the presence of bioactive molecules (UHPLC-ESI-HRMS analysis) with already proven antioxidant potential (e.g., carotenoid - fucoxanthin, chlorophyll derivatives - pheophytin *a* and pheophorbide *a*). It should be mentioned that the antioxidant activity cannot be related to the particular compound, since various bioactive molecules could work in synergy. Such findings point out *C. adhaerens* as an inexhaustible source of natural antioxidants that could be used in research of oxidative stress-related diseases such as cancer, cardiovascular and neurodegenerative diseases. This study is published in Pharmaceuticals (doi: 10.3390/ph14090944).



BROWN SEAWEED ALGINATE AS CARRIER FOR *Myrtus communis* ESSENTIAL OIL STABILIZATION BY ELECTROSTATIC EXTRUSION

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Since bioactive molecules are extremely sensitive to a number of external factors and are often unstable, they must be protected and stabilized by encapsulation methods such as spray drying and electrostatic extrusion. Myrtle (*Myrtus communis* L.) has long been known for its beneficial therapeutic effects on disorders of various organic systems. Its essential oil is obtained by distillation of the leaves and contains various terpenoids, with 1,8-cineole, myrtenyl acetate, and α-pinene predominating. Alginates isolated from brown seaweed are often used as carriers for encapsulation due to their gel forming property in the presence of divalent cations such as calcium where alginate solution containing bioactive compounds is first dispersed into small droplets and then solidified by gelation. Therefore, the aim of this study was to investigate the possibility of stabilization of myrtle essential oil by the formation of brown algae alginate beads.

Myrtle essential oil beads were prepared by electrostatic extrusion encapsulation and the effect of the concentrations of sodium alginate as carrier (0.5, 1 and 1.5%) and calcium chloride as recipient (3 i 5%) on the yield and physical properties of the beads was studied. Myrtle essential oil (5%) was added to sodium alginate solutions together with 0.5% Tween 20 as emulsifier and homogenized with a mechanical stirrer at 10,000 rpm for 4 min. Droplets were formed by extrusion on Büchi encapsulator by applying electrostatic potential of 500 V, frequency 80 Hz with 1 mm needle tip. Yield (Figure 1) was calculated as percentage of wet beads weight and showed significant differences between formulations. The lowest yields were obtained for the samples with the lowest concentration of Na-alginate (0.5%). For both $CaCl_2$ concentrations an increase in encapsulation yield was observed with increasing Na-alginate concentration with the highest value determined in the sample prepared with 3% $CaCl_2$ solution. Largest beads (Table 1) were produced with 1.5% alginate and 5% $CaCl_3$ while 3% $CaCl_3$ caused less variation between samples. Sphericity improved greatly with the addition of higher levels of Na-alginate.

FIGURE 1. MEO electrostatic extrusion encapsulation yield (%). Different letters indicate significant differenced ($p<0.05$)

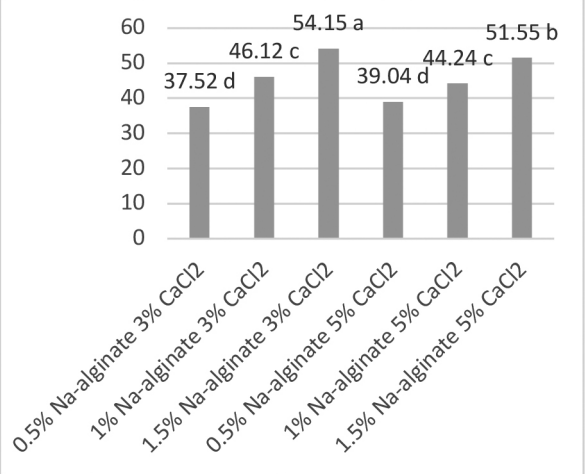

TABLE 1. Largest (d_{max}) and smallest (d_{min}) dimension and sphericity factor (SF) of MEO/alginate beads. Different letters indicate significant differenced ($p<0.05$)

Formulation		d_{min}	d_{max}	SF
Na-alginate (%)	$CaCl_2$ (%)	(mm)	(mm)	
0.5	3	$2.46^{b,c}$	$3.12^{a,b}$	$0.12^{a,b}$
1	3	$2.67^{a,b}$	$3.02^{a,b}$	$0.06^{a,b}$
1.5	3	$2.69^{a,b}$	2.98^{b}	0.05^{b}
0.5	5	2.21^{c}	2.43^{c}	0.14^{a}
1	5	$2.46^{b,c}$	$3.21^{a,b}$	0.05^{b}
1.5	5	2.92_{a}	3.41^{a}	$0.08^{a,b}$

GENE EXPRESSION IN THE PACIFIC WHITE SHRIMP *Penaeus vannamei* UNDER COMPENSATORY GROWTH PREVIOUSLY EXPOSED TO TEMPERATURE CHANGES AND TEMPORARY FASTING

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The Pacific white shrimp, *Penaeus vannamei*, is the most cultivated species worldwide, however, during cultivation, environmental changes can occur that generate stress and alter the normal growth of organisms. When stress is temporary and favorable conditions for growth are restored, shrimp may exhibit an accelerated growth rate known as compensatory growth (CG), which allows them to reach partially or fully the weight of non-stressed organisms. However, if stress is extreme, the CG response capacity may be limited and reach a no-return point. The aim of this study was to evaluate the effect of temperature and temporary fasting periods on the CG response of *P. vannamei* and to determine gene expression during this process.

Shrimp *P. vannamei* with an average weight of 0.75 g were exposed to different stress periods with fasting for 3, 7, and 14 days. Simultaneously, each period was analyzed under three temperatures: 22°C, 26°C, and 30°C. Each period was followed by a recovery stage in which feeding and temperature were restored *ad libitum* and 30°C, respectively, for the remaining time of the 5 experimental weeks. Treatments were evaluated in triplicate and compared with a control group fed to satiety and at 30°C throughout the experiment. Specific growth rate (SGR, % day^{-1}), weight gain (WG), survival (S), food intake (FI), and feed conversion ratio (FCR) were weekly determined and muscle was sampled from all treatments for differential gene expression (DGE) analysis.

Organisms exposed to 3 days of fasting achieved the same weight as those in the control group, which indicated a complete CG, whereas organisms exposed to 14 days of fasting reached the no-return point, being unable to compensate growth and a reduction in WG (>50%) was observed. The SGR and FI were higher in shrimp exposed to 3 days of fasting, however, within this group, FI and FCR were lower at 26°C, suggesting shrimp were more efficient to use the nutrients at this temperature. The DGE with respect to the control group showed over-expressed genes related to the use of stored energy and the molting process during CG (Table 1), whereas genes related to cell damage repair were overexpressed in organisms that reached the no-return point (Table 2).

Penaeus vannamei exposed to 3-day fasting were able to fully compensate their growth and overexpressed genes related to the use of stored energy and the molting process, whereas an extended fasting of 14 days affected their CG response reaching the point of no-return and induced over-expression of genes related to cell damage repair.

TABLE 1. Overexpressed genes during compensatory growth of *P. vannamei* exposed to 3 days fasting and 30°C.

Gene name	Function
Heat shock protein 67B2	Stress response
MTS domain	DNA methylation
Phosphorylase b kinase	Glycogen metabolism
Cuticular proteins	Cuticle constituents

TABLE 2. Overexpressed genes associated with the no-return point of *P. vannamei* exposed to 14 days fasting and 30°C.

Gene name	Function
Centromere protein X	DNA repair
QM protein	Ribosome constituent

ANTIMICROBIALIN VITROACTIVITY OF PHYTOEXTRACTS *Allium sativum*, *Origanumvulgare*, *Curcumalonga* AND *Cinnamoniumverum* AGAINST *Streptococcus agalactiae* Ib ISOLATED FROM TILAPIA (*Oreochromis* sp.) WITH STREPTOCOCCOSIS

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Streptococcosis is the most important bacterial disease in tilapia (*Oreochromis* sp.) production worldwide.Itiscausedmainlyby*Strepto coccus*agalactiae(GBS)serotypesIa,Ib,III,IV,andIX in tilapia. Antibiotic resistance has been described for GBS in aquaculture, for this reason, it has an urgent need to discover new alternative therapies to control bacterial diseases. Phytoextracts have shown efficient activity against the most important bacterial pathogens in aquaculture. ALLIUM-OR®is a product composed of *Allium sativum*, *Origanum vulgare,* and ascorbic acid. TUR-AQUA®has *Curcuma longa*, *Cinnamonium verum* and choline chloride. We evaluated in vitrodifferentdosesofbothproductsagainst*S.agalactiae*Ib.*Streptococcusagalactiae*(GBS)Ib wasaerobicallyculturedfor24hat30°Cuntilthe exponentialgrowthphasewasreachedat5,7x10^{4}ufc/ml. Four doses of each product were incubated with GBS at 30ºC for 24 to 48h. All were tested in triplicate. ALLIUM-OR®completely inhibited the growth of *S. agalactiae* Ib in the different doses evaluated. TUR-AQUA did not show an inhibitory effect on *Streptococcus agalactiae* Ib growing at any concentration tested.

Oilsandextractsof *Allium sativum*havebeen**T**evaluatedpreviouslyfortheireffectagainst*S.mutants*,*S.pyogenes*inhumans,and*S.iniae*and*S. agalactiae* in vitro and in vivo in tilapia and trout with similar results to our study. *Origanum vulgare* had been evaluated against *S. agalactiae* Ib and our results showed that in combination with *Allium sativum* inhibited the growth of GBS Ib.*Curcuma longa* and *Cinnamonium verum* didnothaveaneffecttoreducethegrowthofthisbacterium.ALLIUM-OR®with*Alliumsativum*,

Table1. In vitro effect of ALLIUM-OR® and TUR-AQUA® against *Streptococcus agalactiae* Ib strain

Name of product	Concentration	Growth		
		Replicate 1	Replicate 2	Replicate 3
ALLIUM-OR®	8g/Kg of feed	not growth	not growth	not growth
	4g/Kg of feed	not growth	not growth	not growth
	2g/Kg of feed	not growth	not growth	not growth
	1g/Kg of feed	not growth	not growth	not growth
TUR-AQUA®	8g/Kg of feed	INC	6 x10^{7}UFC/ml	7 x10^{7}UFC/ml
	4g/Kg of feed	2 x10^{8}UFC/ml	5 x10^{6}UFC/ml	1,4 x10^{8}UFC/ml
	2g/Kg of feed	1 x10^{8}UFC/ml	8 x10^{8}UFC/ml	2,5 x10^{7}UFC/ml
	1g/Kg of feed	6 x10^{8}UFC/ml	4 x10^{8}UFC/ml	4 x10^{7}UFC/ml
FLORCAP® 5% (Florfenicol)	10 mg/Kg of biomass	not growth	not growth	not growth
Positive control (GBS)	5,7 x10^{4}UFC/ml	6 x10^{4}UFC/ml	3 x10^{4}UFC/ml	5 x10^{4}UFC/ml

Aeromonas hydropila; 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 conductive 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 Asciles/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.

METODOLOGÍA PARA MEDIR LA RESILIENCIA DE LAS GRANJAS ACUÍCOLAS ANTE EL CAMBIO CLIMÁTICO

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Nunca hemos estado tan sujetos a cambios climatológicos en el periodo de la era moderna como en nuestros tiempos. Desde el año 2005 se han suscitado cambios relevantes en materia de variación climática como altas temperaturas y huracanes simultáneos a nivel mundial. Esto ha comprometido seriamente la seguridad alimentaria exigiendo una visión de futuro en los sistemas de producción acuícola, priorizando la necesidad a ser más resiliente a dichos cambios, lo que deriva en una búsqueda de indicadores de resiliencia acuícola a nivel ambiental y socio-económico en las unidades de producción, para garantizar la capacidad de recuperación después de un *shock*, ya sea de tipo antropogénico o natural. En términos ecológicos, la resiliencia está referida a la capacidad que tiene un sistema para enfrentar y acomodarse a factores perturbadores manteniendo sus funciones pese a las alteraciones.

En esta investigación se evaluaron 10 unidades de producción acuícola, en un periodo espacio temporal de diez años, considerando 5 indicadores de resiliencia ambiental que se involucran en la producción, determinado en base a ellos dos categorías: nivel de resiliencia y factor de riesgo ambiental. Ambos son vitales para la acuicultura de futuro y para sobreponerse a los efectos inminentes del cambio climático.

Basadados en el modelo PER (Presión, Estado, Respuesta), mediante herramientas de teledetección, en base a los siguientes indicadores: 1.-Ubicación geográfica de la unidad productiva, 2.- Altura sobre el nivel del mar, 3.-Distancia más próxima a un cuerpo de agua, 4.-Nivel de producción en toneladas anuales, 5.- Perímetro total de la unidad productiva (autoconsumo/ empresarial). Con base a estos indicadores, se determinó la categoría de resiliencia que ostenta cada unidad de producción, las cuales están divididas en: (Factor I Nivel de Resiliencia I-A), (Factor II Nivel de Resiliencia II-B), (Factor III Nivel de Resiliencia III-C). En conclusión, la resiliencia es vital para los agroecosistemas, pues las alteraciones en el medio ambiente siempre están presentes, por lo cual debemos adaptar la producción acuícola a la aplicación de una visión más holística, donde se gesta la base de una acuicultura resiliente de fututo y se refuerza la toma de conciencia a la adaptación frente a escenarios adversos.

EFFECT OF SIZE HETEROGENEITY OF NILE TILAPIA *Oreochromis niloticus* ON THE OPTIMAL HARVEST TIME: A BIOECONOMIC APPROACH

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A critical problem in the production of Nile tilapia *Oreochromis niloticus* in intensive and hyperintensive systems is the heterogeneity of body sizes as it influences the final production and economic yield. The objective of this study was to calculate the bioeconomic effect of size heterogeneity on the production of Nile tilapia at a commercial level and to determine the optimum harvest time (OHT) considering four minimum marketable sizes target (MMS = 350, 400, 450, 500 g).

A trial of 330 days was conducted in a hatchery of a commercial farm, Nile tilapia fingerlings were reared at the same stocking density (40 ind m^{-3}) with different fish sizes: homogeneous size (HM) and heterogeneous size (HT). The initial mean weight ± standard deviation (SD) of the organisms was HM = 100.17 ± 5.91g and HT = 96.55 ± 24.58 g, with three replicates for treatment. The initial variance of HM and HT was σ^2 34.91 and σ^2 604.19, respectively.

The optimal harvest time (OHT) corresponds to the time at which the quasi-profit of the variable costs (understood as the profits obtained after paying for the variable costs of production) are maximized. The large variation in the size observed in the tilapia of the same cohort illustrates a high propensity of the organisms to develop and grow at different rates. The biometric data of discrete 15-day periods, for the 330 day study period was used for the bioeconomic analysis.

The seeding of homogenous organisms is highly recommended to the farmers, because the heterogeneity at seeding negatively affects the system performance by lowering biomass, in addition to reducing the quasi-profits of the variable costs.

Table 3. Performance of Nile Tilapia rearing at homogenous (HM) and heterogeneous (HT) seeding strategies

Parameters	HM	HT
N_0 initial number	180	180
Initial weight (g)	100.17 ± 5.91 [a]	96.55 ± 24.58 [b]
Final weight (g)	894.35 ± 198.03 [a]	860.74 ± 230.95 [a]
Initial biomass (g)	18,030.00 [a]	17,379.00 [a]
Final biomass (g)	160,983.23 [a]	154,932.52 [a]
AGR (g day^{-1})	1.78 [a]	1.77 [a]
Survival (%)	96.11 [a]	97.22 [a]
CV_0 (%)	5.90 [a]	25.50 [b]
CV_{330} (%)	22.10 [a]	26.80 [a]
S_{kB0}	- 0.13 [a]	- 0.01 [b]
S_{kB180}	- 0.11 [a]	- 0.38 [b]
S_{kB330}	- 0.23 [a]	- 0.12 [b]
K	0.02068 [a]	0.02065 [a]
Total food (kg)	365.46 [a]	345.89 [a]
FCR	2.35 [a]	2.33 [a]

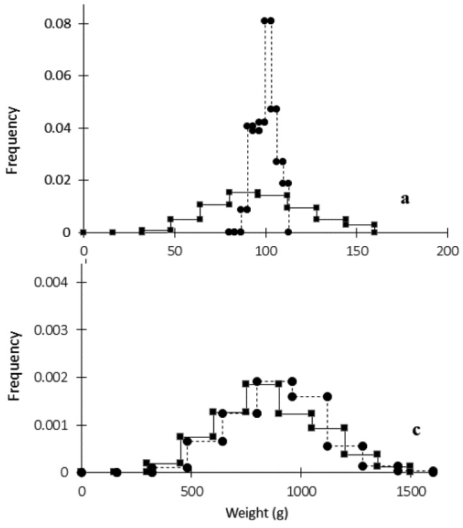

Figure 1. Size distribution in the homogenous (HM = •) and heterogeneous (HT = □) groups of Nile Tilapia during the growth stage. (a) Rearing time = 0 and (c) Rearing time =330 days.

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.

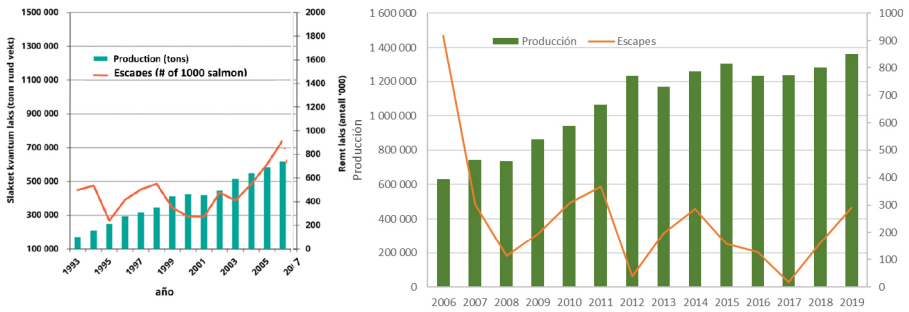

Figure 1: Historical relation between production and fish escape

EVALUACIÓN DE TRES PROGRAMAS DE ALIMENTACIÓN SOBRE EL COMPORTAMIENTO PRODUCTIVO DE LA GAMITANA *Colossoma macropomum*

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La gamitana (Colossoma macropomum) es una de las especies de mayor cultivo en la amazonía peruana, sin embargo, se necesita de estrategias que contribuyan con la sostenibilidad y rentabilidad de la producción. Una de estas estrategias involucra el manejo adecuado de la nutrición y alimentación de la especie. Los requerimientos nutricionales van disminuyendo a medida que los peces aumentan su edad y tamaño, por lo que es importante conocer el momento óptimo para realizar el cambio de alimento de una etapa temprana a otra más tardía. Para ello, se utilizan programas de alimentación utilizando alimentos balanceados de inicio, crecimiento y acabado, con diferentes tamaños de partícula, que se suministran en cantidades adecuadas de acuerdo a la etapa de cultivo (periodo óptimo de cambio de dieta). De esta manera se asegura una alimentación para promover el máximo potencial de crecimiento.

La evaluación consiste en tres programas de alimentación, utilizando los resultados de requerimientos nutricionales para gamitana obtenidos en el LINAPC (Laboratorio de Investigación en Nutrición y Alimentación de Peces y Crustáceos). El periodo experimental de 180 días, utilizando las instalaciones de la Universidad Nacional del Centro del Perú, Satipo-Junín, ubicada en la selva alta del Perú. Alevines de gamitana de 27g fueron distribuidos en 9 estanques de tierra (79 peces por unidad experimental), bajo el diseño estadístico DCA. Los peces del programa I con alimento de inicio durante 90 días, alimento de crecimiento por 60 días y alimento de acabado durante 30 días; el programa II con el alimento de inicio durante 60 días, alimento de crecimiento 60 días y alimento de acabado 60 días y las gamitanas del programa III utilizando el alimento de inicio durante 30 días, alimento de crecimiento 60 días y alimento de acabado durante 90 días.

Los resultados preliminares durante 90 días de evaluación, se muestran en el Cuadro 1. Se observa que no existen diferencias significativas en los parámetros de consumo de alimento diario y ganancia de peso. Se observan diferencias significativas en la conversión alimenticia a favor de los programas II y III.

Cuadro 1. Efecto del programa de alimentación sobre el comportamiento productivo de la gamitana

Programa	Consumo de alimento diario (g)	Ganancia de peso diario (g)	Conversión alimenticia
I	4.55a	3.37a	1.35b
II	4.43a	3.45a	1.28a
III	4.78a	3.70a	1.29a

a, b letras diferentes en la misma columna muestran diferencias estadísticas (α<0.05)

FIRST RECORD OF THE INVASIVE CLAM *Corbicula fluminea* IN ENSENADA, BAJA CALIFORNIA, COULD BE A CANDIDATE FOR AQUACULTURE?

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Back in the 1970´s an invasive freshwater clam was reported in Mexicali, Baja California Mexico. Since then, no additional data on this clam has been published to accurately measure the extent of colonization of the freshwater sources in the state by this invasive species, nor a corroboration of the identity has been conducted. During the dry season of 2021, many shells of dead freshwater clams were observed along the shore of the dam "Emilio López Zamora" in city of Ensenada, Baja California, Mexico. A sampling of these shells was carried out in the dam to perform morphological and molecular identification of the invasive species.

Morphological identification was performed using conventional malacological techniques. Molecular identification was carried out by using DNA extracted from the shells and periostracum for whole genome amplification (WGA) in combination with PCR and sequencing.

The absence of live organisms during the sampling limited the study to using the shells for the identification. It is unknown if a mortality event had occurred that decimated the freshwater clam populations. Both classic malacological and molecular identification, independently, indicated that the clam is *Corbicula fluminea*, synonymous of *C. malianensis*. This species is characterized by a medium-sized bivalve shell, sub-triangular, subequilateral of moderate thickness, black to dark brown near the edge of the shell and yellowish brown on the umbo. Genus specific primers for the cytochrome oxidase subunit 1 gene of *Corbicula* spp. yielded 401 bp (Fig 1) fragment with a sequence identity of 100% to *C. fluminea* (GenBank: KT893369.1) from the Yangtze River Basin.

This is the first record of this invasive species in Ensenada, Baja California confirmed by conventional taxonomy and molecular methods. The molecular identification using the WGA approach as was performed in this study could prove useful for the identification of dead mollusks were limited nucleic acids are available. An interesting potential application that arises is use of this species to aid in biomonitoring and bioremediation of the environment as filter feeder. In addition, it could also be used as a source of food, as is currently being exploring in Malaysia.

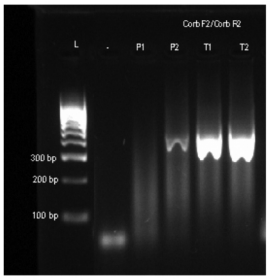

Fig 1. Amplification of a fragment of the cytochrome oxidase subunit 1 gene of *Corbicula* spp. from the periostracum (P1 and P2) and shell (T1 and T2) of the dead clams.

EFFECT OF MEDICATED DIETS WITH FLORFENICOL ON INTESTINAL VILLI IN ATLANTIC SALMON

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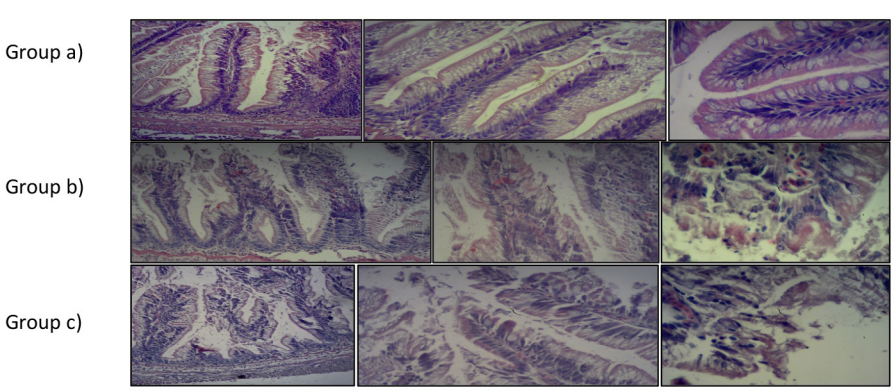
In 2021, 426,309 tons of florfenicol (FCL) were used to control outbreaks of the bacterial pathogen *Piscirickettsia salmonis*. This antibiotic is administered through medicated food as a metaphylaxis strategy. To date, the effect of this drug on the intestinal epithelia has not been reported in healthy fish. With the objective to study the effect of FCL medicated food at a histological level on the intestinal epithelium, a biological assay with healthy fish was performed. 240 Atlantic Salmon with an average weight of 120 g were divided in three groups were treated by administering feed with different concentrations of FCL. Group 1 (no antibiotic, control); group 2 (20mg/kg FCL); and group 3 (30mg/kg). After transportation to the experimental facilities, the organisms were acclimatized for 30 days. Following the acclimation period, the organism were feed with medicated diets following a prophylactic scheme that is identical to those used in commercial production systems. The applied scheme was the following: 15 days of antibiotic treatment; 10 days off; 15 days of antibiotic treatment; and finally, two following periods of 10 days. At the end of each stage, nine fish from each of the groups were euthanized with an overdose of MS-222. Each specimen was dissected, and a portion of the intestine was taken, fixed with bouin solution and stained with hematoxylin and eosin. We confirmed that fish subjected to the antibiotic treatment had shorter villi and damage in the structure and conformation of the intestinal villi compared to the fish of the control group. These results underscore the importance of optimize antibiotic treatment schemes to mitigate the negative effects associated with current metaphylaxis strategies that undoubtedly affect fish health and welfare.



Distal intestine of Atlantic Salmon (H&E, 10X, 20X and 40X) where it is expected to observe intestinal villi with a mucosal layer formed by columnar epithelium. The presence of a moderate number of mucosal cells and a thin lamina propria in the villi is also considered normal. In group a) intestinal villi with a simple columnar epithelium in good condition and the presence of mucous cells are observed. In group b) the villi present structural damage, some mucous cells are observed. In group c) the damage to the intestinal villi is more severe and detachment/fragmentation in the apical area of the villi with very few mucous cells is observed.

DNA EXTRACTION FROM A HISTOLOGICAL SECTION OF THE DISTAL INTESTINE OF *Salmo Salar* FIXED IN BOUIN SOLUTION AND EMBEDDED IN PARAFFIN

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Most of the analysis to determine the composition and diversity of the intestinal microbiota in living beings is carried out from stool samples or directly from the gut. However, the applications of these methods are limited by the need for fresh, unfixed tissues that allow high-quality DNA extraction. Fixed, paraffin-embedded tissue is an invaluable resource for retrospective molecular genetic studies, but high-quality genomic DNA extraction can be problematic[1] . Bouin's solution, a compound fixative, is used in the histological evaluation of cells, due to the nuclear morphological preservation obtained1. Fixation facilitates obtaining images of cell localization and morphology[2]. However, it is unknown how bouin fixation affects the quality of DNA from the gut distal of Atlantic Salmon. To tap into this potentially huge resource, it is imperative to develop methods to isolate high-quality, abundant DNA from fixed, paraffin-embedded tissue sections[3].

DNA extraction from Salmo salar gut distal high-quality DNA. samples embedded in paraffin, fixed with Bouin's solution, was evaluated. For this, we use a DNA extraction kit that includes the combination of proteinase K digestion and column purification. By means of PCR using primers, we amplified the 16s and 23s gene. Amplified fragments of the expected sizes for the 16s gene were obtained, indicating that this method is suitable for the isolation of

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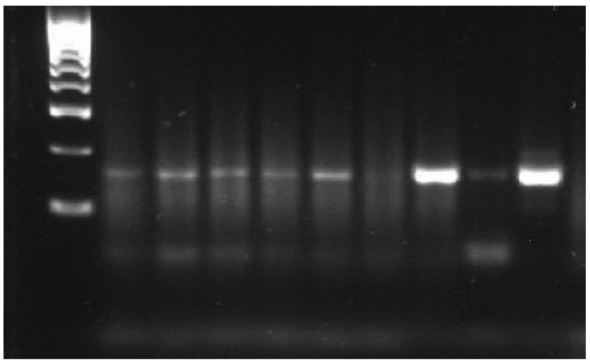

SPERM CRYOPRESERVATION OF GREEN ABALONE *Haliotis fulgens* FOR CONSERVATION AQUACULTURE

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Green abalone, *Haliotis fulgens,* is one of the five species along the coasts of Baja California, Mexico, that has become economically important. Nonetheless, nonregulated fishing activities and anthropogenic impacts have contributed to the decline of its populations, increasing the interest in abalone conservation; therefore, habitat restoration and stocking of native species need to be considered.

Cryopreservation of sperm of aquatic species can be helpful in such programs to protect endangered species in natural disasters or accidents that can affect wild populations. Additionally, the aquaculture industry can also benefit from artificial insemination, reducing the risk of disease transmission, creating new lines with favorable traits, conserving stocks, and transporting good quality gametes between farms. Developing a cryopreservation protocol for green abalone would aid in such restoration efforts, providing an alternative breeding option for this aquaculture industry that is not entirely established.

This work aimed to develop cryopreservation protocols for sperm of *H. fulgens*. In this study, three commonly used cryoprotectants (CPAs) were chosen: Dimethyl sulfoxide (DMSO), glycerol, and methanol. Four different CPAs concentrations (5%, 10%, 15%, and 20 %) and four equilibration times (5, 10, 15, and 20 min) were evaluated in a manually controlled cooling Styrofoam chamber.

Thawed sperm quality was evaluated by estimating the percentages of sperm motility and plasma membrane integrity (PMI) using a dual staining technique. Results suggested that 15% DMSO was the best among the CPAs evaluated, resulting in higher motility (48%) and PMI (58%) (Fig. 1).

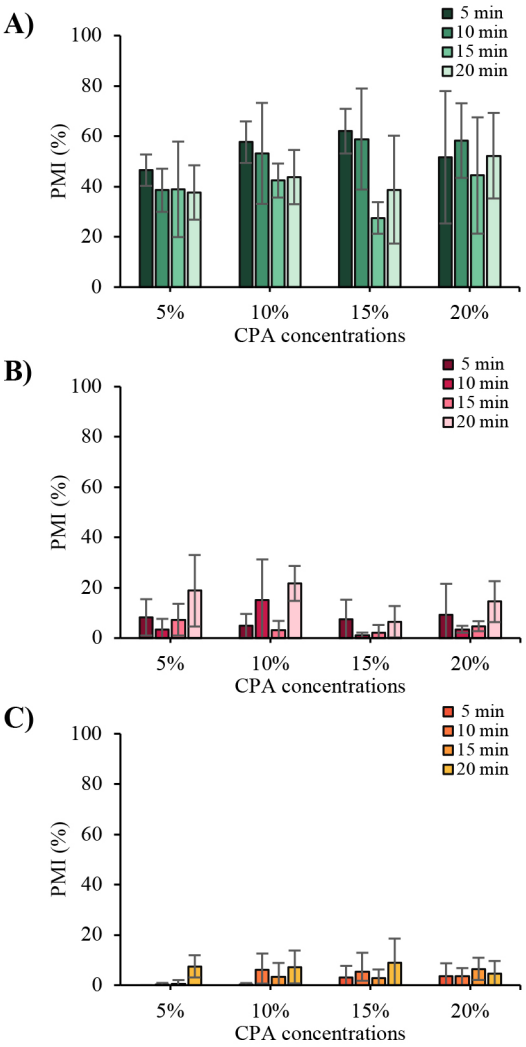

Fig. 1. Percentages of PMI after exposure to A) DMSO, B) Glycerol, and C) Methanol at different times and concentrations.

EVALUATION OF PRODUCTIVE PERFORMANCE AND BODY CHEMICAL COMPOSITION OF THE NILE GRAY TILAPIA *Oreochromis niloticus* DURING THE FATTENING PERIOD UNDER TRADITIONAL CULTURE SYSTEMS AND BIOFLOC TECHNOLOGY

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The Nile gray tilapia is one of the most consumed species in the world, it has characteristics that allow its cultivation in sustainable systems with little or no water exchange, such as the biofloc system, which provides a supplement in the nutrition of cultivated organisms. To determine the effect of the biofloc system on the water quality, the productive performance and the body chemical composition of the Nile gray tilapia *Oreochromis niloticus*, an experiment was carried out for 120 days, with two treatments and three repetitions: traditional system (TS) and heterotrophic biofloc system (BFT) (**Table 1**). Male monosex organisms with initial average weight of 109 ± 39.29 g were used at a stocking density of 100 fish m^{-3}, fed with extruded pellets with 32% CP. The final body weight was significantly higher in the traditional system (279.93 ± 78.51 g) than in the biofloc system (183.49 ± 38.57 g) (**Figure 1**). Water consumption was significantly lower in the biofloc system (1.38 ± 0.15 m^3) compared to the traditional system (48.4 ± 0.0 m^3) (**Figure 2**). The percentage of crude protein in the fillet of the organisms grown in the traditional system was significantly higher (17.69 ± 0.42%) in relation to the biofloc system (16.56 ± 0.07%) (**Figure 3**). The results showed that Nile gray tilapia juveniles raised in the traditional system exhibited the higher growth and body chemical composition, however, the biofloc technology was efficient in water consumption, demonstrating the sustainability of the system.

Table 1. Productive performance and body chemical composition of the Nile gray tilapia (*Oreochromis niloticus*) in traditional systems and with biofloc technology.

Variable	Traditional system			Biofloc system		
	Min	Max	Mean ± sd	Min	Max	Mean ± sd
Performance						
Initial body weight (g)	102	124	110.92±12.05^a	96	120	108.85±12.59^a
Final body weight (g)	116	458	279.93±78.51^b	100	280	183.49±38.57^a
Survival (%)	55.05	100	83.35±24.64^a	82.54	95.02	90.01±6.59^a
Water consumption (m^3)	48.4	48.4	48.4±0.0^a	1.30	1.56	1.38±0.15^b
Fillet proximate composition (%)						
Moisture	77.84	78.9	78.37±0.74^a	78.71	79.37	79.04±0.46^a
Crude protein	17.39	17.99	17.69±0.42^a	16.51	16.62	16.56±0.07^b
Crude fat	1.16	2.33	1.74±0.82^a	1.11	1.33	1.22±0.15^a
Ash	1.20	1.21	1.20±0.00^a	1.15	1.18	1.16±0.02^a

Different superscript letters indicate significant differences among treatments (P<0.05).

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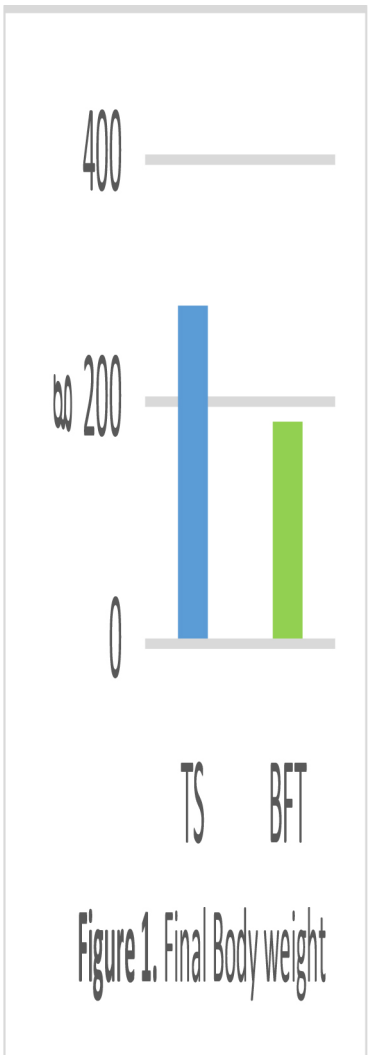

Figure 1. Final Body weight

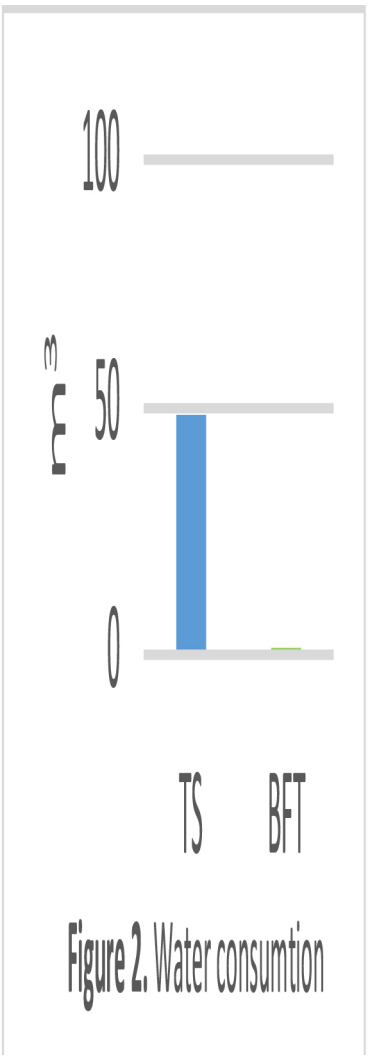

Figure 2. Water consumtion

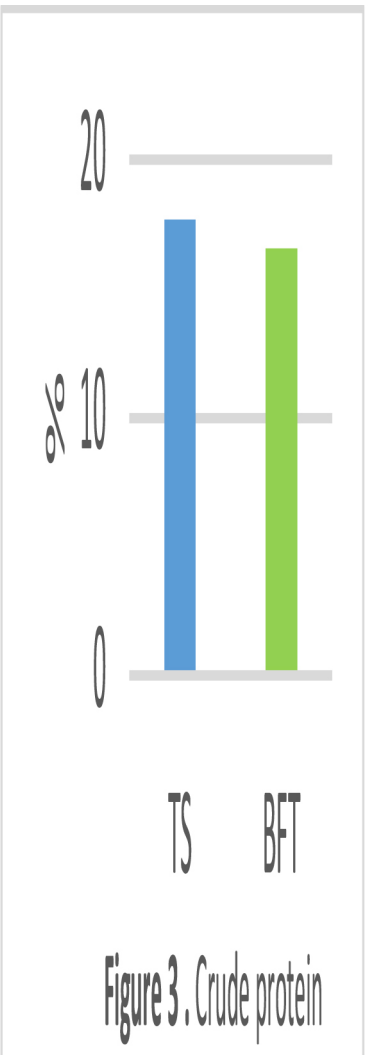

Figure 3 . Crude protein

EVALUATION OF A NEUROSENSORIAL FEED ADDITIVE ON *Litopeneaus vannamei* SHRIMP STRESS REDUCTION

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The marine shrimp aquaculture industry, in particular the whiteleg *shrimp Litopeneaus vannamei*, has experienced exponential technological development, with different production systems worldwide. Shrimp farming is made up of several stages and transitions that represent stressful events for the animals, which have a direct effect on reducing appetite and feed intake. Appetite is regulated by various factors as shrimp has a highly developed and sensitive sensory system, and appetite messages are known to reduce stress messages. In this context, modulating shrimp appetite can have a positive effect on stress reduction. A trial was conducted to evaluate the effect of a neurosensorial feed additive (NFA) on shrimp stress reduction through feed intake.

During 8 weeks, 900 Pacific white shrimp (*Litopeneaus vannamei*) with an average initial weight of 0.5g were divided into 18 ponds of 220L in brackish water with 15 ppt salinity. Faecal matter was siphoned daily, and 60% of the water was changed every third day. The balanced feed used had a protein content of 35% and 7% lipids. The feed was distributed to the shrimp 4 times/day. The NFA was applicated at a 3kg/T rate. Each treatment was replicated 4 times. Feed intake rate, live weight, survival rate, and total biomass were measured at the end of the experiment (week 8). To measure stress reduction, an acute stress induction protocol was started at week 4.

The use of the NFA led to a global improvement of survival rate, from 44 to 62% (Fig.1). After the stress induction protocol, it was observed that immobility time was reduced from 25 to 5 seconds with the NFA. These results suggest that a neurosensorial product have a beneficial effect on stress and mortality reduction in shrimp.

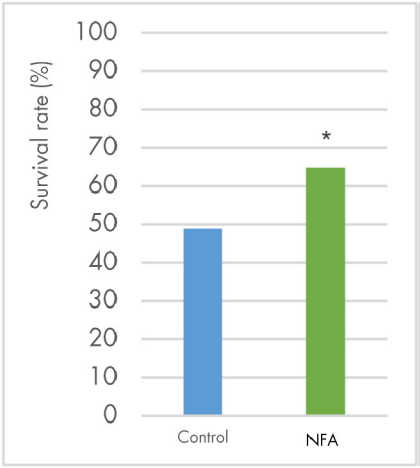

Fig.1 Final survival rate at week 8 *p<0,05

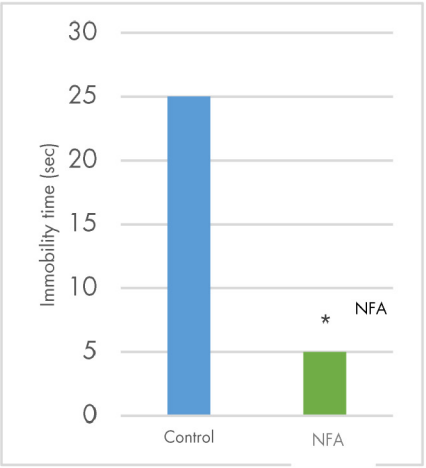

Fig.2 Average immobility time after acute stress in week 4 *p <0,05

EVALUATION OF A NEUROSENSORIAL FEED ADDITIVE ON *Litopeneaus vannamei* SHRIMP STRESS REDUCTION

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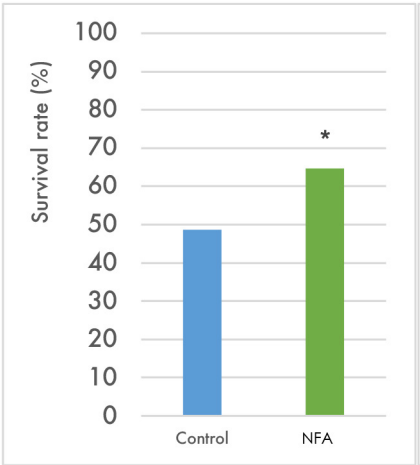

Fig.1 Final survival rate at week 8 *p<0,05

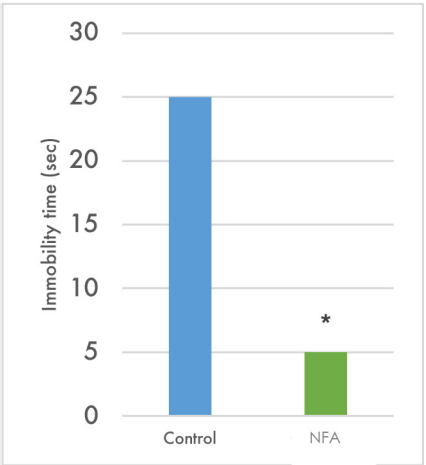

Fig.2 Average immobility time after acute stress in week 4 *p

EXPOSURE TO THE HEAT: THE CONSEQUENCES ON THE PROGENIE OF: *O. maya*

MVZ EPA María de la Luz Chavacán Ávila*
MVZ Norma Alejandra Plata Díaz
Dr. Carlos Rosas Vázquez

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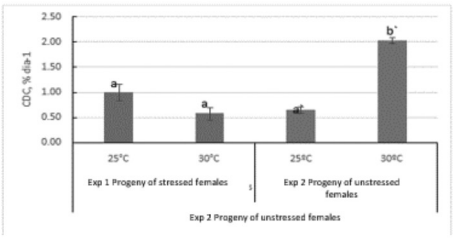
In recent years, various studies have been carried out with *O. maya* with the purpose of evaluating thermal preferences by exposing juvenile and adult octopuses to multiple temperatures and experimental tests to know the thermal resistance and determine physiological events.

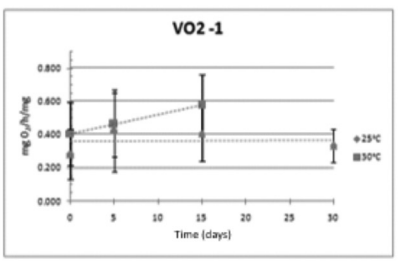
In the present study, the consequences of the exposure of adult female octopus to high temperatures on the physiological state of juveniles maintained in different thermal regimes were evaluated. Juveniles from stressed (30ºC) and unstressed (24ºC) females were kept at 25°C and 30ºC for a period of 20 to 30 days. To which the routine oxygen consumption and the maximum activity metabolism, the activity of the antioxidant defense mechanisms (ANTIOX) and some indicators of the damage produced by free radicals (ROS) were measured. It was observed that octopuses from stressed females kept at 30ºC were unable to survive for more than 15 days. In contrast, the octopuses from unstressed females kept at 30ºC for up to 20 days showed an ability, although limited, to regulate the presence of (ROS).

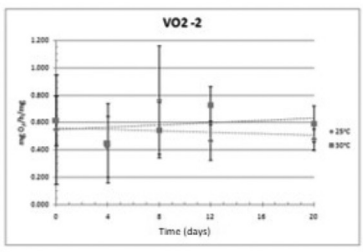
Growth Coefficient (CDC): In experiment 1, temperature did not affect growth. However, there is a tendency to obtain higher growth in specimens exposed to 25°C. In contrast, in experiment 2 it was observed that temperature exerted a significant effect on growth ($P < 0.05$) with values 3.1 times higher in the specimens exposed to 30°C compared to 25°C.

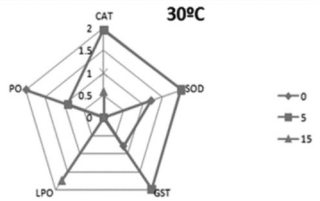
In evaluations of oxygen consumption for both juveniles from stressed and non-stressed females, the specimens exposed to 30°C had an oxygen consumption that increased with exposure time, while oxygen consumption was maintained at 25°C constant.

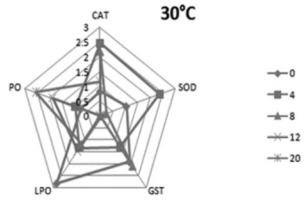
In the octopuses kept at 30°C in experiment 1, it was identified that after 15 days of exposure, the enzymes of the antioxidant system collapsed, allowing the accumulation of peroxidized lipids (LPO). In the octopuses subjected to 30°C of experiment 2, it could be seen that they were able to control (ROS) levels from the first day, managing to maintain this until day 12 of exposure. By day 20, a reduction in the activity of Superoxide dismutase (SOD) and Catalase (CAT) was observed, anticipating a progressive increase in Protein Carboxylation (PO) and peroxidized lipids (LPO).











EVALUACIÓN DE GENES CONSTITUTIVOS Y REPRODUCTIVOS PARA RT-PCR CUANTITATIVA EN *Macrobrachium americanum*

María A. Cisneros Geraldo*, Miriam V. Martín Manzo, Edilmar Cortes Jacinto, Rosa M. Morelos Castro, Adrián Munguía Vega, Manuel A. Vargas Ceballos

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El género *Macrobrachium*, también conocido por el nombre común de langostino, camarón del río, camarón de agua dulce, entre otros, según la región, se distribuye en zonas tropicales, desde la Península de Baja California hasta el noroeste de Perú. *Macrobrachium*, es una de las especies nativas con alto potencial de cultivo, y de las más grandes y robustas, con un peso de hasta 500g. Es por ello, que se encuentra sobreexplotada, sin regulación, ni conocimiento de sus poblaciones naturales, por lo que existe la necesidad de poner énfasis en aspectos reproductivos. Martín, *et al*., (2021) realizaron el ensamble del transcriptoma y análisis diferencial entre testículos, conductos deferentes y ámpula terminal de *M. americanum*, y se obtuvo que cerca de 70 genes se relacionaron con determinación de sexo, espermatogénesis, embriogénesis, entre otros procesos reproductivos. Con ello, surge la necesidad de verificar y evaluar la expresión de genes de interés relacionados con la reproducción del *M. americanum*, por medio de las técnicas PCR y qPCR.

La validación de la expresión de los genes (factor de elongación 1α, gliceraldehído 3-fosfato deshidrogenasa, β-actina, hormona de la glándula androgénica parecida a la insulina y sperm gelatinasa) comenzó con el diseño de *primers* de dichos genes, utilizando plataformas como BLAST-NCBI, Primer3, Primer-BLAST y Oligo analyzer, evaluando así los parámetros como contenido de GC, longitud y temperaturas de fusión. Una vez diseñados los *primers*, se procedió a extraer ARN de muestras de vasos deferentes, testículos y ámpula terminal, utilizando como reactivo TriReagent, y evaluando la integridad del ARN por medio de un gel de agarosa 1%. Posteriormente, se trataron las muestras con DNAsas y se evaluó la contaminación por medio de PCR punto final, utilizando como gen de referencia β-actina. Una vez que se corroboró que no se tenía presencia de contaminantes ni ADN y evaluar la integridad del ARN por medio de gel de agarosa 1%, se procedió a realizar homogenizados con base en el transcriptoma realizado por Martín, *et al*., (2021) y se realizó la cuantificación de ácido nucleico presente en cada una. Seguidamente, se sintetizó cDNA de las muestras a condiciones específicas, y se cuantificó nuevamente el cDNA en NanoDrop, los resultados obtenidos mostraron que las muestras E, F, G y H presentaron mayor concentración. Finalmente, para estandarizar los genes previamente mencionados, se realizaron gradientes de temperatura con base en las temperaturas de fusión de los primers diseñados, utilizando solo las muestras que resultaron con más concentración de cDNA. Una vez determinada la temperatura ideal, se procedió a realizar PCR punto final para el resto de las muestras.

Finalmente, los genes de interés relacionados con la reproducción del langostino del río, se expresan en los diferentes tejidos del tracto reproductor masculino, amplificando para factor de elongación 1α a una temperatura de 62°C, gliceraldehído 3-fosfato deshidrogenasa 58°C, β-actina 59°C, hormona de la glándula androgénica parecida a la insulina 64°C, y sperm gelatinasa 63°C. Así pues, se recomienda concluir con la evaluación de la expresión por medio de qPCR y compararla entre los diferentes tejidos.

CREATING BETTER EDUCATIONAL VIDEOS FOR EXTENSION AND OUTREACH

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Welcome to the video age. The spread of high speed internet access and simple video editing software makes the creation and delivery of online educational videos easier than ever. YouTube videos provide an excellent mechanism for Extension specialists to share information and reach a large potential audience. Putting a Power Point presentation in video format does not make the most of this medium. The creation of good video content requires careful planning and practice. The average attention span of most adults is 15 minutes and students approximately 7-10 minutes. Most lecture presentations, however, last 30 minutes to an hour. How can you get your point across as quickly as possible? We must learn to provide entertainment in addition to education. YouTube suggests that the first 15 seconds of a video are critical for engagement and retention of the audience.

A central figure or "hero" and a story with a beginning, middle and end will go a long way towards retaining the attention of the audience. Join us for a discussion these and other simple tips and techniques that can improve your ability to create watchable or perhaps even enjoyable educational Extension videos.

FEMINIZACIÓN DEL ROBALO *Centropomus undecimalis* MEDIANTE IMPLANTES DE ESTRADIOL

María de Jesús Contreras-García[12]*, Wilfrido Miguel Contreras-Sánchez[1], Manuel Mendoza-Carranza[2], Alejandro Mcdonal-Vera[1], Leonardo Cruz Rosado[1] y Felipe Ramón Zapata[1]

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El robalo blanco, *Centropomus undecimalis* es un pez hermafrodita de importancia comercial en las costas tabasqueñas de México. En estos peces, el momento en que ocurre el cambio de sexo y los factores que lo inducen aún se desconocen, lo cual dificulta su manejo en cautiverio. En este trabajo se presentan los resultados de la inducción del cambio de sexo mediante el uso de estradiol (E2), así como el potencial efecto de la manipulación del fotoperiodo para acelerar la maduración. Se emplearon 80 machos jóvenes producidos en laboratorio en un diseño completamente aleatorizado, distribuyéndose aleatoriamente en grupos de 20 peces bajo los siguientes tratamientos: (T1) implantes con E2 en fotoperiodo natural; (T2) implantes sin estradiol en fotoperiodo natural; (T3) implantes con E2 en fotoperiodo controlado; y (T4) implantes sin E2 en fotoperiodo controlado. Los datos de calidad del agua estuvieron dentro de los rangos considerados como aceptable para cultivos marinos (Tabla 1). Los resultados de canulación e histología indican un 100% de feminización de los peces tratados con E2 (Fig. 1). Las hembras obtenidas son funcionales.

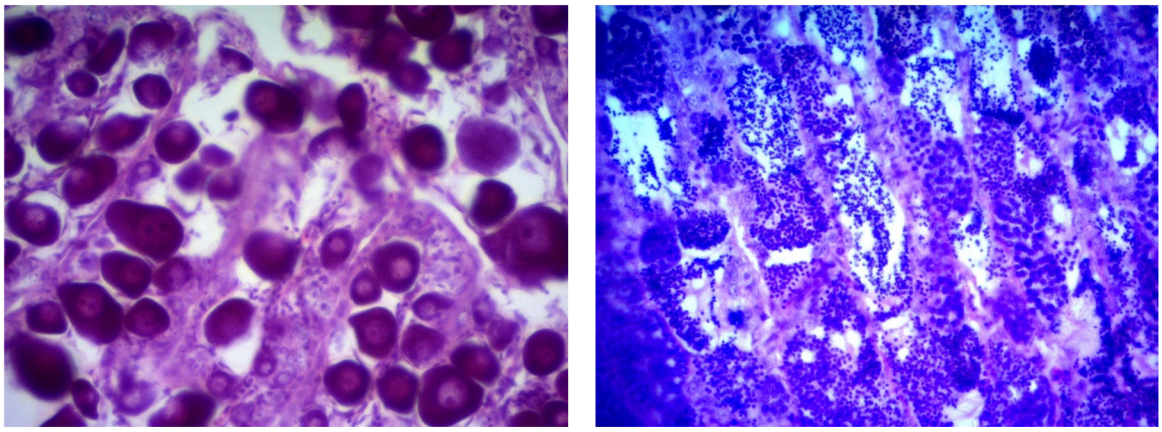

Figura 1. (A) micrografía de una hembra (B) y micrografía de un macho (C)

Tabla1. Valores promedio de calidad del agua en los tanques de cada tratamiento.

Tratamiento	OD (mg/L)	Temp (ºC)	pH (UI)	Salinidad (UPS)	Amonio (mg/L)	Nitratos (mg/L)	Nitritos (mg/L)
T1	4.38 ± 1.15	26.26 ± 1.87	8.56 ± 0.63	34.22 ± 098	0.21 ± 0.22	1.86 ± 1.38	0.11 ±0.13
T2	4.39 ± 1.20	26.29 ± 1.91	8.52 ± 0.66	34.22 ± 098	0.53 ± 0.39	1.84 ± 1.39	0.08 ± 0.11
T3	4.20 ± 1.18	27.75 ± 1.65	8.79 ± 2.31	34.22 ± 098	0.60 ± 0.44	2.22 ± 1.92	0.21 ± 0.20
T4	4.25 ± 1.16	27.70 ± 1.66	8.48 ± 0.73	34.22 ± 098	0.52 ± 0.40	2.60 ± 2.14	0.37 ± 0.40

AVANCES Y CONTRIBUCIONES PARA LA CONSERVACIÓN DE LANGOSTINOS DEL GENERO *Macrobrachium* EN MEXICO

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Las especies de langostinos han sido sobreexplotadas por los pescadores artesanales en diversas cuencas en la que se distribuyen las especies del genero *Macrobrachium*, sea por autoconsumo o venta como alimentos en mercados locales. El cultivo experimental y comercial de los langostinos, aún no ha sido implementado debido a la falta de investigaciones relacionadas a la biología básica de la especie. Centros de investigaciones e Instituciones de Educación Superior (IES) de México (CIBNOR; CIIDIR-IPN-Oax, U de G. UNAM-FES Iztacala), en colaboración UBA (Argentina), Sao Paulo State University (Brasil), entre otras IES han logrado producir larvas, juveniles y cultivar experimentalmente el langostino, desarrollando estudios acerca del crecimiento; fisiología, requerimientos nutricionales, alimentación y reproducción. Los estudios de nutrición, alimentación y reproducción realizados pretenden contribuir en las bases para el cultivo sustentable de especie, y su conservación; para lograr esto se considera importante perfeccionar la tecnología de acondicionamiento de reproductores, juveniles y cultivo larvario. La producción constante de las larvas será el paso inicial para la domesticación de los langostinos, teniendo éstas que presentar desempeños adecuados en los cultivos, índices productivos elevados (alta supervivencia y tasas de crecimiento). Se han logrado avances en su biología básica (sitios de reproducción en BCS, Gro, Jal, y Estado de Mex., captura de pies de crías; movilidad, aclimatación de reproductores; desarrollo y aplicación de dietas; manejo y alimentación); estos temas y resultados de investigación fueron obtenidos por alumnos y alumnas de pregrado, posgrado; y posdoctorado, mismos que se presentaran en obra en proceso.

ACCELERATING THE DEVELOPMENT OF MARINE AQUACULTURE IN ENERGETIC OCEANS: OFFSHORE INNOVATIONS IN SCALING UP CATAYLZE NEARSHORE INNOVATIONS TO SCALE OUT

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Nearshore oceans with their high primary and secondary productivities, logistical support systems, and close proximity to markets are excellent places to develop marine aquaculture. However, as common property resources, the number and diversity of user conflicts increase with proximity to shore. Conflicts result in the overuse of the precautionary approach by ocean governance systems which create too many obstacles to increasing the number and sizes of marine aquaculture operations nearshore. Sustainable system change to expand marine aquaculture nearshore would consider fully ocean engineering advances made offshore that would open up large areas of highly energetic nearshore oceans that have fewer user conflicts. For example, in this context, offshore would be defined as a straight line drawn just along the outer edges of the state of Maine, USA complex shore of islands/bays a distance about 360 km to Canada, extending about 4 km from the shore. This area is approximately 1700 km^2 of theoretically available space in energetic nearshore oceans. Improved development policies would include access in energetic sites nearshore to scale "out", and connect these to offshore leasing for operations to scale "up". The case of the state of Maine, USA and its contiguous sea, the Gulf of Maine (GoM) illustrates the potential for a seamless nearshore-offshore aquaculture governance system. The GoM is a productive marine basin with potential for development of both fed and non-fed aquaculture, located in a bioregion that has an historical affinity for seafoods. Markets are abundant as consumers have a higher rate of seafood consumption than national averages but the region meets its seafood needs from imports. Nearshore energetic ocean spaces are underutilized aquaculture opportunity areas that can serve as well-planned testing platforms for offshore enterprises. Scaling up of nearshore ocean space for aquaculture is currently regulated though an ocean-leasing structure tiered by size. Sizes of permitted offshore aquaculture systems have been derived less by applied science than by social/political factors derived from nearshore experiences, e.g. the "social license to operate". Maine has a unique nearshore aquaculture research and development permit system for "scaling out" of nearshore marine aquaculture development, the limited permit access (LPA) system, which has attracted over 700 entrants. We call for the establishment of aquaculture platforms in nearshore, energetic oceans at scale that will inform the scale of offshore operations needed for financial success. Platforms will incorporate the best available science and participatory tools for improving knowledge exchange from industry to civil society. Sizing permits for offshore aquaculture should be based more on science than the precautionary principle, which has been overused to govern and permit offshore aquaculture.

COMPARISON OF TWO DICTYOTA SPECIES (BROWN SEAWEEDS) BASED ON THEIR ANTIOXIDANT ACTIVITY

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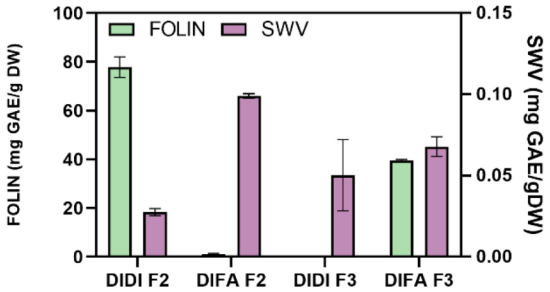
Marine macroalgae or seaweeds are in a continuous focus of the scientific community, but even in the focus of a wider public as they are acknowledged as a valuable source of different bioactive compounds such as polyphenols, peptides, pigments, and polysaccharides that are associated with several health benefits and biological activities. Brown algae are known for their accumulation of specific metabolites with great antioxidant potential. In this study, we evaluated the antioxidant activity of different fractions from two brown macroalgae, *Dichtyota dichotoma,* and *Dichtyota fasciola*. The primarily used method was square-wave voltammetry (SWV), followed by the reduction of the radical cation (ABTS), the 2,2-diphenyl-1-picryl-hydrazyl (DPPH) assay, the Folin–Ciocalteu, and the oxygen radical absorbance capacity (ORAC) assays. To correlate obtained results for antioxidant activity, Pearson's correlation coefficient was used. Toxicity evaluation of obtained fractions revealed the safety of their usage and further implementation.

In this research, the dry residues of extracts from brown macroalgae samples were immobilized on the surface of GCE and immersed into a 0.1 mol/L phosphate buffer solution. To evaluate their activity and optimal electrochemical response, a change in the pH values, ranging from 3 to 11, for all obtained extracts was tested. To assess antioxidant activity using voltammetric analysis, the area under the curve (AUC) was integrated, the value of which represents an estimate of the total antioxidant activity of the extracts. The highest antioxidant activity based on the AUC value was obtained for *D. fasciola* F2 (0.099±0.001 mg/g GAE) followed by *D. fasciola* F3 (0.068±0.006 mg/g GAE) > *D. dichotoma* F3 (0.050±0.002 mg/g GAE) > *D. dichotoma* F2 (0.028±0.002mg/g GAE). No significant difference in antioxidant activity was observed between fractions of two Dictyota species. Other employed spectrophotometric methods also revealed similar behavior of two Dictyota species, however, somewhat higher activity was observed for both *D. dichotoma* fractions. The Pearson's correlation coefficient for SWV *versus* Folin-Ciocalteu method was a negative value of -0.99 (p<0.05). For other spectrophotometric methods, there was no statistically significant correlation obtained with SWV. Also, a high negative correlation between DPPH and Folin-Ciocalteu assay was observed with Pearson's correlation coefficient of -0.81. Correlations indicate that the synergistic effect of different compounds extracted from the samples impacted their antioxidant response. All three researched brown macroalgae have shown to be a potent source of natural antioxidants that could further be used in the research of oxidative stress-related diseases.



USING FORMALIN-FIXED PARAFFIN-EMBEDDED TISSUES AS A SOURCE OF NUCLEIC ACIDS FOR MICROBIOME ANALYSIS IN RAINBOW TROUT *(Oncorhynchus mykiss)*

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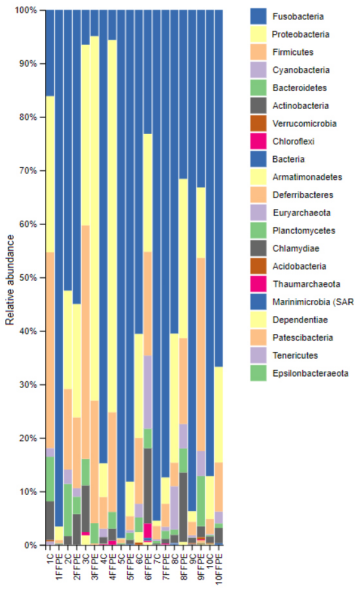
In recent years, the study of the gut microbiota of cultured teleost fish has gained relevance as cornerstone studies that provide important information on the gut microbial status and their impact on the fish health and welfare. Considering the role of the microbiome in animal health, it is fundamental to explore the utility of microbiota studies using histological samples. These studies could provide valuable insights on how the gut microbiome is modulated upon dietary and environmental changes under which fish are being reared currently compared to rearing conditions in the past.

In this study, we analyzed 20 microbiomes targeting the V3-V4 hypervariable region of the 16S rRNA gene derived from Illumina® amplicon sequencing of the rainbow trout (*Oncorhynchus mykiss*) acclimated at two temperatures (16˚ and 22˚C) for five weeks. At the end of the experiment trout were euthanized and the intestine was aseptically removed from each fish. Part of the intestine was flash frozen for molecular biology and the other section was fixed for histological processing. DNA was extracted from both samples, fresh frozen (FF) and formalin-fixed paraffin-embedded (FFPE), and subjected to 16S rRNA amplicon sequencing.

Amplicon sequencing of the V3-V4 region of the 16S rRNA resulted in an average of 17,335 reads for the FF samples and 5,032 reads for the FFPE samples, corresponding to a total of 735 OTUs. The microbiome of the intestine of the rainbow trout acclimate at 16˚C showed a higher diversity of species (α-diversity) (p-value = 0.04) than organism at 22˚C when analyzed using FF samples of the intestine. The difference in the diversity was not captured when parallel but FFPE samples were used (p-value = 0.34). The *O. mykiss* microbiome was dominated by the bacterial taxonomic phylum Fusobacteria. This phylum constituted a high, but variable, sequence proportion at both temperatures and for both types of samples (FF and FFPE) (Fig. 1). This data reveals that microbiome can be successfully determined using FFPE tissue opening a new horizon in studying intestinal microbiota.

Fig 1. Rainbow trout microbiome at a phylum level at both temperatures and fixation types. “C” indicates fresh frozen and “FFPE” indicates formalin-fixed paraffin-embedded.



REARING TILAPIA (*Oreochromis niloticus)* IN PONDS FROM A RIVER SPRING: ECOLOGICAL IMPLICATIONS

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Oreochromis niloticus is the most cultivated fish species in Brazil, representing 60.6% of fish production in our country. Tilapia is mainly produced in net cages, however, excavated or natural ponds have also been used. We surveyed parasites in tilapias collected in ponds from a river spring and discuss its ecological implications.

During an inventory of fishes and parasites conducted in 2020/2021, 32 specimens of *O. niloticus* specimens were collected from a spring river area of Pardo River, Botucatu, São Paulo state, Brazil (23°0'21.21"S, 48°22'31.99"W) and screened for monogenean, crustacean, and myxozoan parasites.

The analyzed tilapias were only infested with monogeneans (*Cichlidogyrus halli*, *C. mbirizei*, *C. thurstone*, *C. tilapiae*, *C. philander*, and *Scutogyrus longicornis*) (Tab. 1, Fig. 1) and copepods (*Lamproglena* sp., *Rhinergasilus piranhus*, and *Tropocyclops* sp.) (Fig. 2).

Many farmers have lakes on their properties and, for several reasons and without proper technical knowledge, try to produce tilapia and end up introducing them and their parasites as alien species. Studies have been demonstrated the negative impact of the introduction of tilapias on the population of native fauna.

Tilapias were infested with six monogenean species. However, these parasites have not been detected in native fishes so far. On the other hand, we found three native crustaceans infesting some tilapias.

We recommend a management plan to control this alien species avoiding the extinction of native species and future parasite-host switch among fishes, mainly in river springs.

Table 1. Ecological data of monogeneans found in gills of *Oreochromis niloticus* in ponds from a river spring in Brazil. P – prevalence; A, abundance; MII, mean intensity of infestation; SE, standard error.

Monogenean	P	A ± SE	MII ± SE
C. halli	9.4	0.094 ± 0.052	1.000 ± 0.000
C. mbrizei	3.1	0.125 ± 0.125	4
C. thurstone	9.4	0.250 ± 0.191	2.667 ± 1.667
C. tilapiae	15.6	0.656 ± 0.344	4.200 ± 1.463
C. philander	15.6	0.313 ± 0.171	2.000 ± 0.775
S. longicornis	3.1	0.031 ± 0.031	1
Total	**28.1**	**1.469 ± 0.716**	**5.222 ± 2.146**

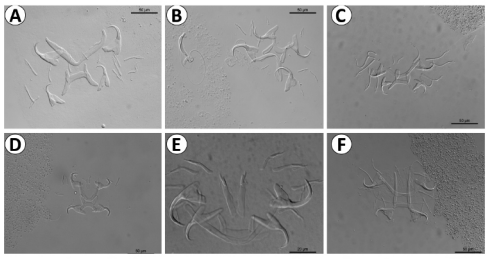

Figure 1. Monogeneans found in gills of *Oreochromis niloticus* in ponds from a river spring in Brazil. A) *C. halli*; B) *C. mbirizei*; C) *C. thurstone*; D) *C. tilapiae*, E) *C. philander*; F) *S. longicornis*.

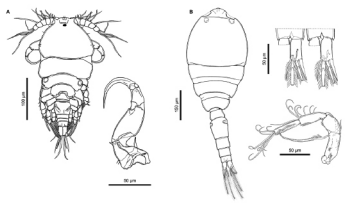

Figure 2. Copepods found in gills of *Oreochromis niloticus* from a river spring in Brazil. A) *R. piranhus*; B) *Tropocyclops* sp.

SUSTAINABILITY OF URBAN AQUAPONICS FARMS: AN EMERGY POINT OF VIEW

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Aquaponics is a food production system that aims higher sustainability by integrating advantages gained from aquaculture and hydroponic production. Aquaponics aims to mimic the biological process that happens in the natural environment in a controlled production system. As it can be applied to small scales, aquaponics is considered an important alternative for urban regions, which have low availability of agricultural land and water resources. Furthermore, the advantage is that it is located close to final consumers.

Aquaponics has been labeled as an environmentally friendly food production system, but its demand for energy and materials cast doubt on its sustainability. A systemic understanding of aquaponics production systems is needed to determine the magnitude and balance between its potentialities and constraints, in which emergy synthesis appears as a powerful tool for this purpose. This study applies emergy synthesis to assess the sustainability of two different (scale and marketable products) urban aquaponics farms in Brazil, but differently from other emergy studies, ecosystem services and disservices are included in the analysis as an attempt to represent the system performance holistically. Results show that the type of materials used in aquaponics infrastructures has the highest influence on total emergy demand. Surprisingly, electricity and fish feed showed a low influence on the total emergy, reinforcing the idea that aquaponics systems have a more efficiency feeding management than traditional aquaculture systems. Besides producing vegetables and fish, the inclusion of ecosystem services highlights the importance of aquaponics for educational and tourism purposes. Finally, the obtained indicators from modeling scenarios revealed that replacing the water source and some materials deserves priority attention to increase the sustainability of urban aquaponics farms.

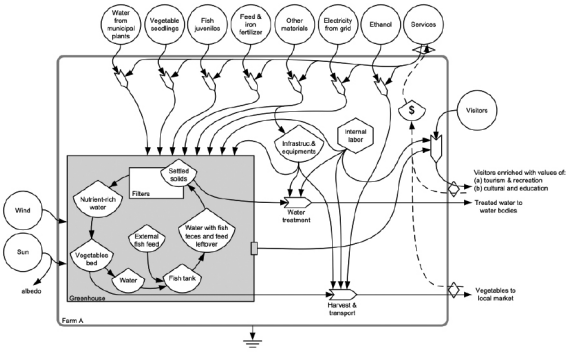

Figure 1. Energy diagram of the aquaponics system in Farm A.

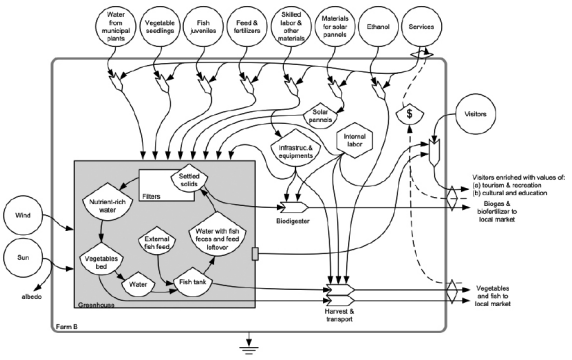

Figure 2. Energy diagram of the aquaponics system in Farm B.

ENZYMATIC RESPONSES TO DIETS WITH DIFFERENT PROTEIN LEVELS IN LARVAL AND JUVENILE STAGES OF *Brycon moorei* (STEINDACHNER 1878), DORADA DEL RIO MAGDALENA, COLOMBIA

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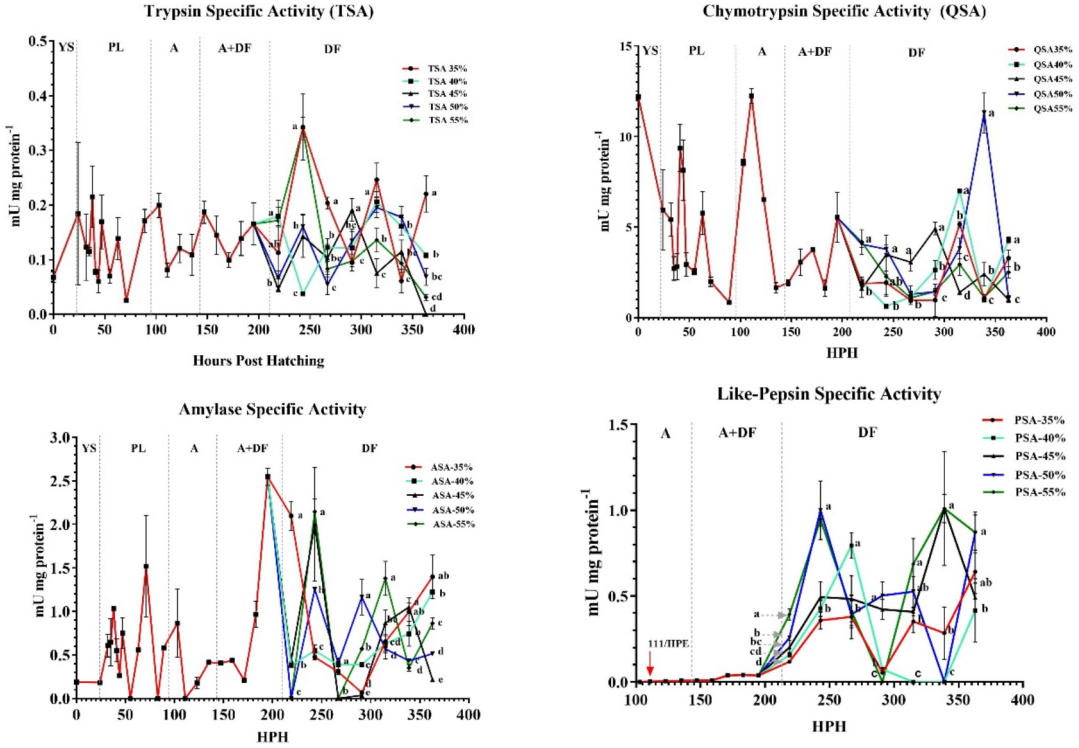
Of the *Brycon moorei* endemic species of Colombia, it is not known about the enzymatic activity and its modulation concerning diets formulated in the early stages. In a recirculating set-up and with a completely randomized quintuplicate design, an ad libitum feeding protocol was applied, which included, among others, balanced feed with inclusion levels from 35% to 55% of crude protein (CP). For this, larvae 22 hours post-hatching (HPH), with an average initial weight and length of 3.07±0.69 mg and 6.069±0.22 mm, respectively, were seeded at a density of 17 larvae L-1 in tanks with an effective volume of 60 L. The enzymatic activity was analyzed about the experimental diets. Specific activity (SA) of serum enzymes (Figure 1, 2, and 3) was found from the beginning of hatching, with fluctuating values significantly related to the feeding protocol and time in HPH and the pepsin-type SA began from the 111 HPH, marking the time in which the use of dry diet can be started (Figure 4).



Figures 1, 2, 3, and 4. SA trypsin, chymotrypsin, amylase and pepsin (mU mg of protein^{-1}) about the inclusion levels of CP in the experimental diets, at HPH and to the food item: yolk-sac larvae (YS); forage larva (PL); nauplii of Artemia salina (A); A+dry diet (DF) and dry diet (DF). Different letters show significant differences ($p < 0,05$).

INFLUENCE OF DIETARY PROTEIN LEVELS ON ZOTECHNICAL PARAMETERS IN LARVAL AND JUVENILE STATES OF *Brycon moorei* (STEINDACHNER 1878), DORADA DEL RIO MAGDALENA, COLOMBIA

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Brycon moorei is known to be an opportunistic omnivorous species. It is not known about their nutritional requirements and zootechnical responses concerning formulated diets at any stage of development in this species. In a recirculating aquaculture system (RAS) and with a completely randomized quintuplicate design, an *ad libitum* feeding protocol was applied, which included forage larvae of Prochilodus magdalenae, Artemia salina, and balanced feed with inclusion levels from 35% to 55%. of crude protein (CP). For this, larvae 22 hours after hatching (HPE), with an average initial weight and length of 3.07±0.69 mg and 6.069±0.22 mm, respectively, were seeded at a density of 17 larvae L-1 in tanks with an effective volume of 60 L. The main zootechnical parameters were analyzed about the experimental diets (Weight gain (WG-mg); daily weight gain (DWG-mg); length gain (LG-mm). Daily gain in length (DGL-mm), feed conversion (FC), consumption (C, mg/day/larva), specific growth rate (SGR), condition factor (K) and survival (S%)). The results of the zootechnical parameters indicate that the best diet was significantly 45% CP when compared to the other CP levels used (Table 1). In the mean comparison tests between weight gain (GP) and % CP, the polynomial regression model determines that the probable protein requirement for this stage corresponds to 48.5% CP (Figure 1).

Table 1. Effect of protein levels on zootechnical parameters in B. moorei (n=105 per CP level). Different letters show significant differences ($p< 0.05$)

	PROTEIN LEVELS (%)									
VARIABLES	**35**		**40**		**45**		**50**		**55**	
WG	62,82±31,48	**bc**	61,5±20,4	**c**	176,2±61,3	**a**	69,3±27,7	**bc**	114,05±50,02	**bc**
DGW	4,18±2,09	**bc**	4,09±1,35	**c**	11,75±4,08	**a**	4,62±1,84	**bc**	7,6±3,35	**bc**
LG	0,4±0,22	**b**	0,46±0,22	**b**	0,72±0,19	**a**	0,53±0,12	**ab**	0,34±0,1	**b**
DGL	0,027±0,014	**b**	0,031±0,015	**b**	0,048±0,012	**a**	0,035±0,008	**ab**	0,023±0,008	**b**
FC	0,88±0,06	**b**	0,88±0,04	**b**	0,57±0,03	**a**	1,2±0,06	**c**	0,49±0,07	**a**
C	5,57±0,37	**c**	5,43±0,25	**c**	10,21±0,54	**a**	8,35±0,41	**bc**	5,69±1,65	**c**
SGR	7,4±3,2	**b**	6,6±2,3	**b**	13,0±1,75	**a**	7,42±1,86	**b**	9,8±3,9	**ab**
K	1,08±0,19		1,00,379±		1,14±0,06		1,1±0,23		1,1±0,17	
S	43,7		42,8		45,6		39,9		43,5	

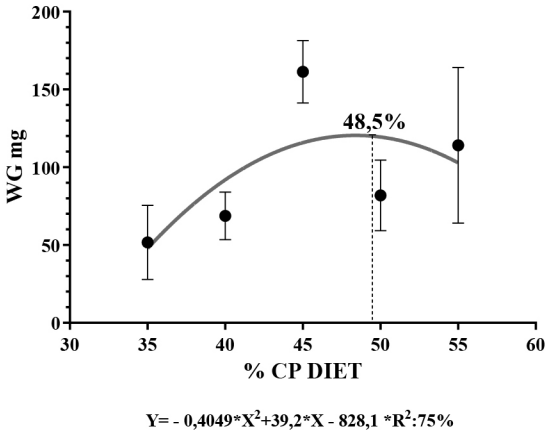

Figure 1. Comparison test of means, using the polynomial regression model for the different PC levels (35 to 55% PC).

FEED EFFECTORS IN SOY OPTIMIZED DIETS: PASSIVE ACOUSTICS FROM THE LABORATORY TO THE POND

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Passive acoustics monitoring (PAM) can contribute to improve feed formulations by providing information on shrimp feeding behavior or it can be used as a tool to manage feed inputs. Under laboratory conditions, we have utilized PAM to identify feed modifiers that act as chemoattractants, feeding incitants, and stimulants in plant-based feed formulations. Based on laboratory results we formulated an "all plant" basal diet (AP) and three AP diets supplemented with 2% krill meal (AP+KM), 2% squid meal (AP+Sq), or 4% fish hydrolysate (AP+FH). These diets were then tested in the laboratory to evaluate the response of the shrimp, in research ponds using automatic feeders equipped with acoustic feed management (AQ1) as well as outdoor tanks where feed inputs were controlled. In the laboratory, the acoustic feeding activity was monitored by hydrophones connected to a multitrack recorder (16 bit/192 kHz), while feed consumption (FC) (g) was measured as the amount of food eaten by the shrimp (n=10) in each aquarium over a 30 min period. The clicks produced by shrimp during feeding activity were automatically detected and quantified using specific acoustic targets. These data were also used to calculate the relationships between the number of clicks and FC. An overall improvement was observed in acoustic feeding activity and FC for all-plant diets with attractants in comparison with the basal diet, confirming an improvement in feeding response in the laboratory. To confirm the efficacy in the field, the same feeds were offered to shrimp (30 shrimp/m^2) reared in 0.1 ha research ponds each equipped with AQ1 acoustic feeders which allowed for demand-style feeding. The growth and general health of shrimp were monitored via weekly sampling and final production values were determined after fully harvesting each pond. Harvest was delayed due to a hurricane, which in combination with multiple blue green algae blooms, led to shrimp mortalities. As a result of low survival, some ponds were excluded from the final data set. No statistically significant differences were found between the treatments for any major production parameter apart from feed input and feed cost (Table 1). Significantly more of the AP+FH diet was offered to shrimp in the respective ponds than that of the basal diet, suggesting that fish hydrolysate increases the intensity of the feed response in Pacific white shrimp in semi-intensive pond culture.

Table 1. Pacific White Shrimp response to four soy-based diets with varying attractants from 13-week trial in ponds.

Treatment	g/week	Weight (g)	Feed Input (Kg/ha)	Feed Cost ($/ha)	Survival (%)	Yield (kg/ha)	FCR
All Plant[1]	1.41	19.40	6636[b]	8,732[b]	69.90	4044.6	1.65
Krill[2]	1.47	20.37	7556[ab]	10,845[ab]	77.38	4778.3	1.63
Squid[3]	1.53	20.99	8145[ab]	11,597[a]	76.56	4831.5	1.69
Fish Hydro[4]	1.50	20.42	9147[a]	12,333[a]	83.17	5155.5	1.79
P-value	0.7194	0.8128	0.0237	0.0143	0.5155	0.2139	0.9492
PSE[5]	0.0612	0.9099	329.142	451.64	4.9987	286.971	0.1552

[1]n=4 ; [2]n=3 : [3]n=1 : [4]n=2
[5]PSE: Pooled Standard Error

TEXTURE PROFILE ANALYSIS OF COLLAGEN EXTRACT FROM *Octopus maya*

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Mayan octopus (*Octopus maya*) is an endemic species of high commercial value to the Yucatan Peninsula. This cephalopodred in color and has a pair of spots under its eyes, it reaches up to 100 cm with a short lifespan of approximately a year. In 2019, almost 12,000 tons were caught, with annual revenues of 360 million pesos which represented 15,000 direct jobs. *O. maya* is commercialized without its mantle (MA) and tentacle's ends (TE), which are removed to obtain visually attractive products. However, research is being carried out to create products of high value from these wastes, ensuring the sustainable use and exploitation of *O. maya*. This is the case of obtaining collagen from mayan octopus wastes (MA and TE) which can be used in the food, cosmetic and pharmaceutical industries.

Collagen is a structural protein considered the most abundant, it's found in tissues such as skin and tendons, but also in bones and gristles. Due to its high protein content and its functional properties, collagen is highly demanded in the food industry as a gelling agent, emulsifier in food industry and drug delivery systems, with a wide range of applications such as cell scaffolding, moisturizer agent, etc. In southeast unit of CIATEJ a collagen extract has been obtained from *O. maya* which is being characterized. The determination of texture profile analysis (TPA) will allow to infer its quality as a biomaterial and gelling agent. TPA in collagen samples (moisture=97.65 %) was performed with a texturometer (EZ-SX, Shimadzu). A penetration test was carried out with a 15 mm plate in a cylindric container with a diameter of 25 mm. A downward compression polarity speed of 2 mm/s with a displacement limit of 15 mm/s was used in both compression cycles and determining the parameters of hardness, adhesiveness, cohesiveness, adhesive force and gumminess. Samples showed a slight hardness of 0.31 N in collagen extracts from *O. maya* (Table 1), which is less than collagen from pork (14.6 N) 6.67% (w/v), also the samples exhibited slight adhesive force and gumminess with no adhesiveness (0 N), while extracted collagen from pig reported 13.17 N (Fig. 1) this may be due to a higher moisture content in the octopus' samples. It's recommended to increase collagen concentration in the samples as well as to continue evaluating its textural but also its viscoelastic and flow behavior in to infer potential applications in the industry.

Table 1 Results of TPA in collagen extract.

Name	*Hardness (H)*	*Cohesiveness (C)*	*Adhesive Force (AdF)*	*Gumminess (G)*
Unit	N		N	N
TPI	0.31 ± 0.13	0.45 ± 0.26	-0.06 ± 0.04	0.14 ± 0.08

H: Maximum force at any time during the first compression cycle, C: total area of 1st cycle / total area of 2nd compression cycle, AdF: force required to detach the sample from surface and G=H*C.

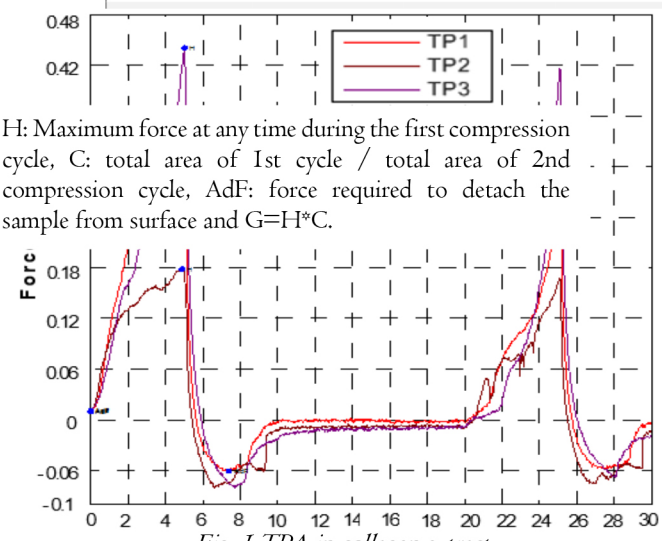

Fig. 1 TPA in collagen extract.

FEASIBILITY STUDY OF AN AQUAPONIC POLYCULTURE TILAPIA-CATFISH AND LETTUCE-CHARD

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Aquaponics is positioned as a food production system that approaches sustainability. However, there are still some challenges to overcome, these derived from the complexity of integrating its biological components; which still limits crop productivity. Likewise, studies of aquaponic systems have focused on establishing the relationships between fish and plants that allow the highest growth rates; however, the option of linking more than one aquatic and plant species in each production cycle has not been explored. Therefore, the objective of this work is to determine the feasibility of implementing an aquaponic polyculture with tilapia-catfish and lettuce-chard.

The feasibility of implementing a coupled aquaponic polyculture was determined; evaluating the growth of specimens of tilapia, catfish, lettuce and chard. The polyculture worked for 60 days with three treatments: T1 (system with 40 tilapia, 20 lettuce and 20 chard), T2 (system with 40 catfish, 20 lettuce and 20 chard) and T3 (system with 20 tilapia, 20 catfish, 20 lettuce and 20 chard).

The water was monitored in the variables of temperature, dissolved oxygen; as well as the concentrations of phosphate, nitrates, nitrites and non-ionized ammonia. Likewise, biometric data monitoring of fish and plants was carried out. During the 60 days of experimentation, the variables monitored in the water were within the ranges required for tilapia and catfish farming (Table 1). Discarding the influence of water quality as a limiting factor in fish growth. However, the differences between treatments place T3 with the highest concentration of nitrates; which is related to the greater growth of the plants in said treatment.

The fish with the highest growth and survival rate were those present in T1 (Table 2); however, this is not related to the growth of the plants, which in this treatment ended with low growth rates. On the contrary, T3 with a fish growth rate lower than T1 presents the highest growth rates for lettuce (Table 3) and Swiss chard (Table 4).

The study provides information that allows considering the implementation of aquaponic systems coupled with polyculture modality; this with the objective not only of generating food but also diversifying its production.

Table 1. Water quality in the different treatments. Values are presented as mean ± standard deviation of samples collected during the experimental period. Values with different superscripts present significant differences (P<0.05)

Variable	Values of culture	T_1	T_2	T_3
Temperature (ºC)	20 - 32	23.1 ± 1.1^a	23.5 ± 0.7^a	23.9 ± 1.0^a
Dissolved oxygen (mg L^{-1})	4 - 9	7.36 ± 0.48^a	7.45 ± 0.46^a	7.94 ± 0.51^a
Nitrates (mg L^{-1})	<300	129.12 ± 3.05^b	124.25 ± 4.18^b	140.93 ± 3.66^a
Nitrites (mg L^{-1})	<5	0.57 ± 0.12^a	0.52 ± 0.11^a	0.64 ± 0.12^a
Non-ionized ammonia (mg L^{-1})	<2	0.29 ± 0.08^a	0.23 ± 0.09^{ab}	0.12 ± 0.06^b
Phosphate (mg L^{-1})	<1.4	0.10 ± 0.02^a	0.10 ± 0.03^a	0.11 ± 0.03^a

Table 2. Productive behavior that presents the means ± standard deviation of the growth rate (GR) and survival rate (SR) of the fish at the end of the 60 days of experimentation. Values with different superscripts present significant differences (P<0.05)

Variable	T_1	T_2	T_3	
	Tilapia	Catfish	Tilapia	Catfish
Initial weight (g)	2.85 ± 0.08	2.93 ± 0.06	2.83 ± 0.05	2.91 ± 0.06
Final weight (g)	45.09 ± 2.09	39.92 ± 1.38	40.14 ± 1.75	36.73 ± 1.75
GR (g)	42.24 ± 1.83^a	36.99 ± 1.27^b	37.31 ± 1.39^b	33.82 ± 1.56^c
SR (%)	95 ± 2 ^{a}	93 ± 3^a	87 ± 2^b	86 ± 3^b

Table 3. Productive behavior that presents the means ± standard deviation of the growth rate (GR) and survival rate (SR) of the lettuce plants at the end of the 60 days of experimentation. Values with different superscripts present significant differences (P<0.05)

Variable	T_1	T_2	T_3
		Lettuce	
Initial weight (g)	5.74 ± 0.21	6.15 ± 0.28	5.98 ± 0.19
Final weight (g)	120.76 ± 2.87	112.83 ± 2.21	136.95 ± 2.87
GR (g)	115.02 ± 1.49^b	106.68 ± 1.25^c	130.97 ± 1.84^a
SR (%)	89 ± 2 ^{a}	87 ± 4^a	75 ± 2^b

Table 4. Productive behavior that presents the means ± standard deviation of the growth rate (GR) and survival rate (SR) of the chard plants at the end of the 60 days of experimentation. Values with different superscripts present significant differences (P<0.05)

Variable	T_1	T_2	T_3
		Chard	
Initial weight (g)	7.78 ± 0.16	7.93 ± 0.15	8.08 ± 0.18
Final weight (g)	136.43 ± 3.22	139.55 ± 3.89	148.67 ± 3.01
GR (g)	131.65 ± 2.09^b	133.62 ± 2.34^b	140.59 ± 2.38^a
SR (%)	92 ± 2 ^{a}	90 ± 3^a	82 ± 3^b

THE COMPLETE MITOGENOME OF LION'S PAW SCALLOP *Nodipecten subnodosus* AN SPECIES WITH AQUACULTURAL POTENTIAL

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Nodipecten subnodosus (G. B. Sowerby I, 1835) belongs to the Pectinidae family and is a species with a high aquacultural potential in Mexico, where it is known as "Mano de León" or lion's paw. As with any other aquacultural species, there is interest in generating genetic markers for aquaculture purposes. The objective of this study was to obtain the mitochondrial genome (mitogenome) using next generation sequencing (NGS). Total DNA was extracted (QIAGEN, Hilden, Germany) from a gill sample, cleaned and sent for NGS at the University of Georgia using a high-throughput sequencer TruSeq NanoTM kit (Illumina,San Diego, CA).

After, reads were cleaned and assembled with CLC Genomics Workbench (10.1), Novoplasty and Geneius software packages. The largest contig corresponded to the mitogenome. However, there was a size difference among contigs (~300 bp) from the different software packages, which is been corroborated. Annotation was carried out with Mitos (software) and checked with other Pectinidae mitogenomes downloaded from the NCBI.

The mitogenome of *N. subnodosus* had a length of ~16,396 bp (35%A, 25%T, 20% G and 20% C) (Fig. 1). When compared with other Pectinidae. mitogenomes, *N. subnodosus* mitogenome had a novel gene arrangement, which will be shown and discussed further together with the phylogenetic relationships.

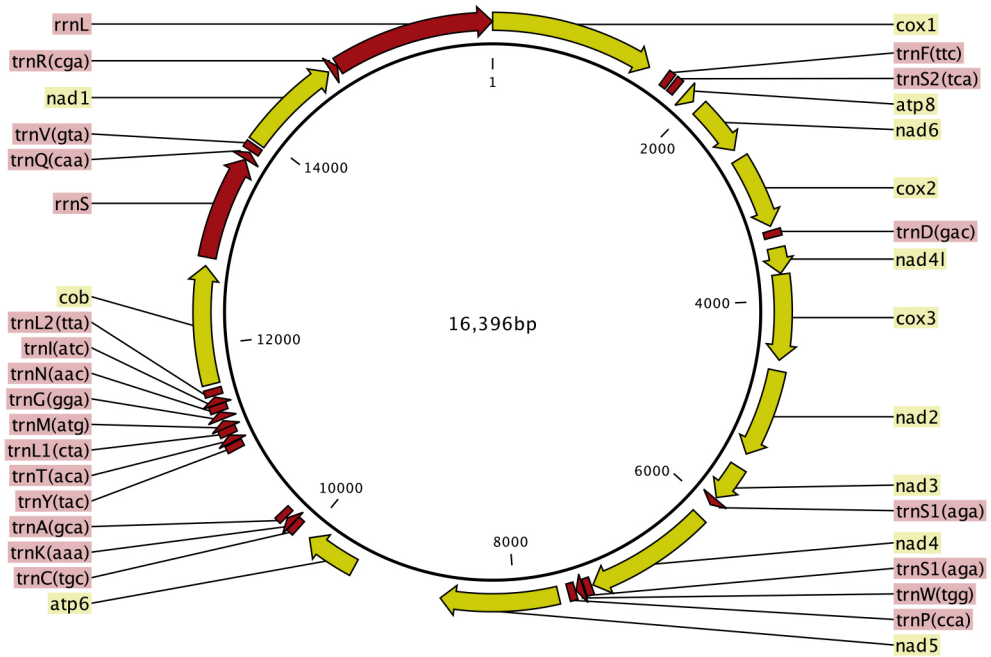

Fig. 1. *Nodipecten subnodosus* mitochondrion genome.

THE MEXICAN AQUATIC GENETIC RESOURCE NETWORK: WHERE DOES IT STAND AND WHERE IS IT GOING?

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The Network for the Aquatic Genetic Resources (NAGR) in Mexico is an inter-institutional and interdisciplinary network mainly composed of researchers to support the National Subsystem for Aquatic Genetic Resources (SUBNARGENA, whose name is Spanish) belonging to the Ministry of Agriculture and Rural Development (SADER) of the government of Mexico. The SUBNARGENA's main objective is to increase the knowledge and the conservation and sustainable use of Mexican biodiversity, primarily of that used as a food resource. The NAGR was founded in 2010, and one of its objectives is to genetically characterize key aquatic species. At that time, mitochondrial genomes and microsatellites were considered as main molecular markers used for genetic characterization. Next-generation sequencing (NGS) has been used to obtain complete mitochondrial genomes (mitogenomes) from several fish, elasmobranchs, octopuses, and geoducks (Table 1). Microsatellites were also identified and sets of specific markers were developed and standardized for some species.

As the NGS tools have increased and the third-generation sequencing is widely available, the NAGR using new markers such as SNPs under different platforms for genetic characterization. In this talk, more options for the NAGR will be proposed for the genetical characterization of aquatic species submitted to the SUBNARGENA and few examples will be given.

Table 1. Mitogenomes obtained from different important aquatic species.

Scientific name Teleost fish	Mexican common name	English common name
Anoplopoma fimbria	Bacalao negro	Sablefish
Atractosteus tropicus	Pejelagarto	Tropical Gar
Chirostoma humboldtianum	Pez blanco	Shortfin Silverside
Cyprinodon macularius	Perrito del desierto	Desert Pupfish
Herichthys minckleyi	Mojarra de Cuatro Ciénegas	Minkley's Cichlid
Petenia splendida	Tenguayaca-mojarra	Bay Snook
Pterois volitans	Pez león	Lionfish
Stegastes flavilatus	Damisela	Beaubrummel Damselfish
Xenotoca variata	Pintada	Jeweled Splitfin
Elasmobranch		
Carcharodon carcharias	Tiburón blando	Great White Shark
Carcharhinus falciformis	Tiburón sedoso	Silky Shark
Carcharhinus leucas	Tiburón toro	Bull Shark
Narcine entemedor	Raya eléctrica gigante	Giant Electric Ray
Zapteryx exasperata	Raya guitarra	Banded Guitarfish
Mollusks		
"Octopus" fitchi	Pulpo pigmeo del Pacífico	Pigmy Octopus
Octopus mimus	Pulpo de Gould	Gould Octopus Or Changos Octopus
Panopea generosa	Almeja generosa	Pacific Geoduck Clam
Panopea globosa	Almeja generosa	Cortés Geoduck Clam
Coral		
Porites panamensis	Coral de dedos	Eastern Pacific Coral

EFFECT OF MICROENCAPSULATED *Bacillus licheniformis* ON THE GROWTH, SURVIVAL AND IMMUNE SYSTEM OF WHITE SHRIMP (*Litopenaeus vannamei*) CHALLENGED WITH *Vibrio parahaemolyticus* AT LABORATORY LEVEL

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Shrimp farming is considered a fundamental activity in food security programs in many countries. White shrimp (*Litopenaeus vannamei*) is one of the most cultivated species worldwide. One of the alternatives to reduce the problem of diseases caused by vibrios and increase sustainability in the aquaculture sector is the use of probiotic bacteria.

Therefore, the objective of the present investigation was to evaluate the growth, survival and immune system challenged with *Vibrio parahaemolyticus* IPNGVE 16 in *L. vannamei*. 50 µL of *B. licheniformis* were inoculated in 100 mL of trypticasein soy broth at 2.5% NaCl and the culture was incubated for 72h at 32ºC. The microcapsules of *B. licheniformis* were obtained by the spray-drying method using maltodextrin (15%) as an enveloping material, which were added to commercial feed as an additive. Shrimp (*L. vannamei*) were collected in commercial postlarvae laboratories in the state of Sinaloa.

The shrimp were transported to the CIIDIR-IPN Aquaculture Laboratory. Shrimp of 40-60 mg were used in tubs (20 L) with 10 L of filtered seawater (20 µm), at 30 ‰ salinity, constant aeration and fed twice a day. The bioassay lasted 36 d and consisted of 4 treatments (triplicate), I): Negative control, Commercial feed (AC); II): AC + microencapsulated *B. licheniformis* (1.0 g/kg of food), III): AC + microencapsulated *B. licheniformis* (2.0 g/kg of food), IV): AC + microencapsulated *B. licheniformis* (3.0 g/kg feed). For the expression of the immune system, the activity of prophenoloxidase, intracellular superoxide anion concentration and hemocyte count were determined.

The LD50 was 780,000 CFU/mL. Regarding growth, it was observed that the TII (2.08 ± 0.083g) presented the highest growth with respect to the control (1.94 ±0.083 g), there were no significant differences ($P > 0.05$). Survival after challenge with *V. parahaemolitycus* was: Control (52.38%), TII (47.61%), TIII (76.19%) and TIV (76.19%). Being the TIII and TIV where the highest survival was presented (Fig. 1).

For the hemocyte count, TI (positive control) was presented with $2.039 \times 10^6 \pm 0.31 \times 10^6$ hemocytes/mL, T II with $1.84 \times 10^6 \pm 0.17 \times 10^6$ hemocytes/mL, T III with $1.72 \times 10^6 \pm 0.22 \times 10^6$ hemocytes/mL, T IV with $1.49 \times 10^6 \pm 0.30 \times 10^6$ hemocytes/mL.

The generation of intracellular superoxide anion, the activity of prophenoloxidase and phenoloxidase in the treatments did not present significant differences between the treatments with respect to the control ($P > 0.05$).

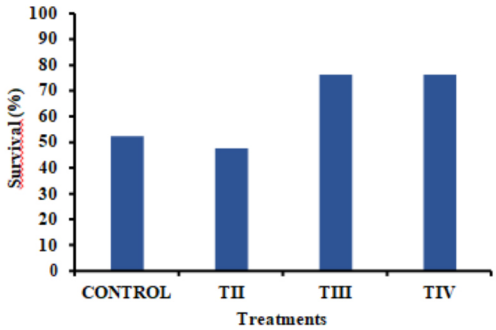

Figure 1. Survival of *Litopenaeus vannamei* after challenge with *V. parahaemolitycus* under laboratory conditions

EFFECT OF OXYTETRACYCLINE ON THE INTESTINAL MICROBIOTA OF *L. vannamei* UNDER PRODUCTIVE CONDITIONS

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\In a *L. vannamei* farm located located in the coastal area of the Gulf of California, 109 km from Hermosillo (Sonora, Mexico), two groups of ponds geographically separated were selected, during a semi-intensive production cycle of approximately 6 months. After around 70 days, an infectious outbreak affected the production, which was diagnosed as AHPND(+) for one of the ponds studied. Oxytetracycline was applied through feed throughout the farm (5kg/ton), which was carried out in each group according to their culture days. Those sown first and whose desired harvest size was larger (green) received the antibiotic before those who were sown later and whose target harvest size was smaller (yellow) (Fig. 1).

At four times, intestinal samples were collected, which were subjected to analysis of the V3-V4 region of the 16s rRNA gene. The results were as follows: the infection period coincides with the decrease in *Proteobacteria* and the increase in *Tenericutes*, being represented mainly by *Vibrio* and *Candidatus* Bacilloplasma, respectively. The statistically significant decrease in the latter was possibly due to treatment with Oxytetracycline, while *Vibrio* had no major changes. The abundance of *Fusobacteria* increased significantly in the group that was sampled three days after the end of antibiotic treatment, and its level was negatively related to the presence of *Tenericutes*. However, at this sampling time only two genera of *Fusobacteria* were detected, and these did not exceed 0.5% relative abundance. These results raise new questions regarding the dynamics of bacterial groups that cannot yet be identified with next generation sequencing.

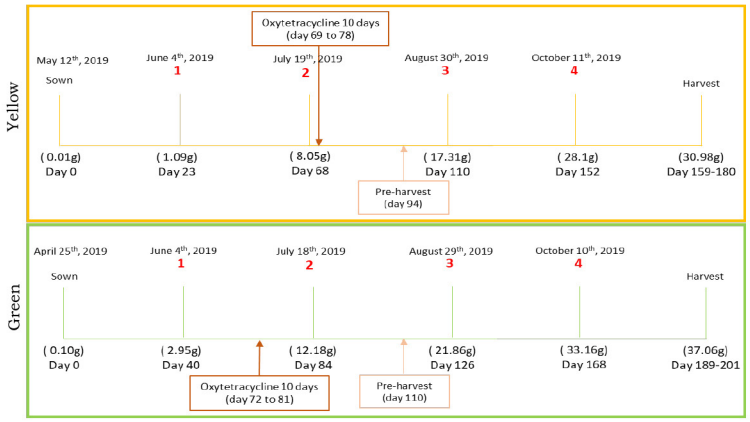

Fig. 1: Sampling times

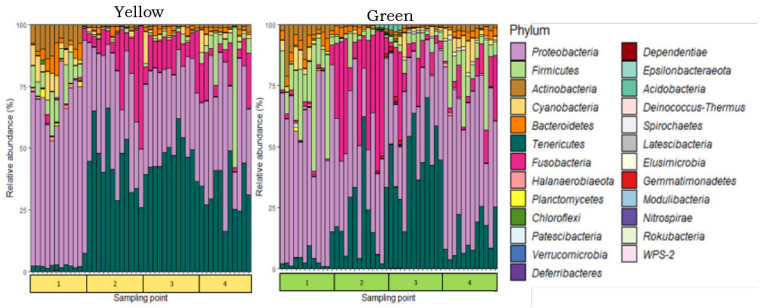

Fig. 2: Phylum relative abundances

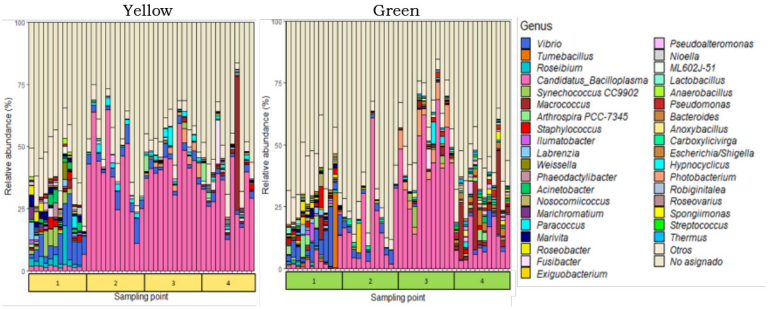

Fig. 3: Genus relative abundances

CHARACTERIZATION OF BACTERIAL MICROBIOTA ASSOCIATED WITH HISTAMINE-CONTAMINATED SARDINES USING NEXT GENERATION SEQUENCIG (NGS)

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Every year hundreds of tons of sardines are discarded due to the presence of histamine, a biogenic amine with the potential to cause intoxication in humans, which is formed by the degradation of fish musculature by microbial agents. There are numerous studies identifying microbes that form histamine in fish meet by plating methods, however, to our knowledge, no study has demonstrated which bacterial group(s) are of greatest importance for the formation of histamine in sardines by NGS so far. Thus, this study aimed to characterize the bacterial microbiota associated with histamine-contaminated sardines using NGS, as an attempt to identify the main causes behind it and help the industry to develop strategies to avoid it.

The regulations of the Brazilian Ministry of Agriculture determines that batches of fresh fish with histamine concentration >100 ppm must be discarded. Therefore, this study separated samples in two groups: contaminated, batches with histamine ≥ 200ppm; and non-contaminated, batches free of histamine (< 1ppm). These batches were from the same supplier, fishing areas and fishing/processing dates. Sample collection took place at the company GDC S/A, located in Itajaí-SC, Brazil. Five samples (10g) of each batch group were collected and submitted to NGS (quintuplicate). The sequences were processed and grouped into operational taxonomic units (OTUs) for identification and, finally, used to determine the relative abundance and for alpha and beta diversity analyses. Generalized univariate linear models were used to identify differences in the OTUs relative abundances ($p<0.05$) between contaminated and non-contaminated samples.

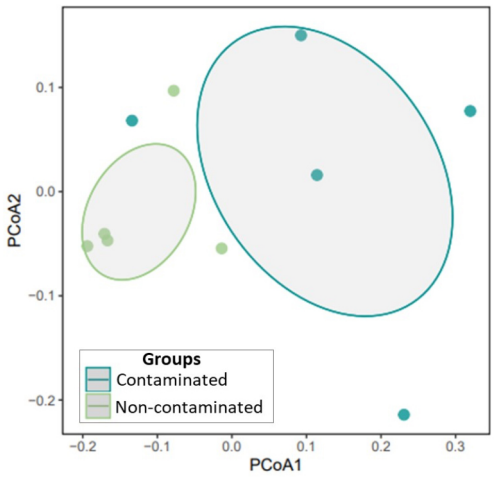

Figure 1. Principal coordinates analysis (PcoA), based on the Bray-Curtis dissimilarity matrix. A beta diversity index used to evaluate the microbiota composition of samples contaminated (>200ppm) and non-contaminated (<1ppm) with histamine.

Eighteen OTUs were identified in total. Alpha diversity analysis revealed that contaminated samples showed higher bacterial richness but lower diversity than non-contamined. Beta diversity analysis confirmed that both groups showed different microbial composition (Figure 1). Bacteria with optimal growth under mesophilia and commonly associated with fish decomposition, such as *Shewanella* ssp. and *Photobacterium* ssp., were more abundant in the contaminated samples than in the non-contaminated ones ($p<0.05$), while psychotropic bacteria unrelated to spoilage, such as *Psychrobacter* ssp. and *Pseudoalteromonas* ssp. had higher relative abundance in samples without histamine ($p<0.05$). Therefore, the analyses of the microbiota structure and composition of both samples suggest that samples with histamine concentration ≥ 200ppm was handled differently from those without histamine, probably the cold chain was not correctly applied from the time of fishing to freezing and sending to the slaughter plant.

Funding: GDC Alimentos S/A

IMPROVING MANAGEMENT OF SIZE HETEROGENEITY AND MINIMUM MARKET SIZES IN TILAPIA CULTURE

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Size heterogeneity and minimum market sizes are two aspects that can improve the economics of a farm. A bioeconomic model has been used to understand the effect of both aspects in tilapia farming and considering a homogeneous size (HM) and heterogeneous size (HT) planting strategy. Results were compared using a mean growth model and quantile regressions, respectively.

Growth data from a pilot tilapia farm in the state of Yucatan were used. Parameterization of the growth models was performed in R, using the nls and quantreg packages. The quantiles = 0.025, 0.05, 0.10, 0.15, 0.25, 0.50, 0.75, 0.85, 0.95 and 0.975 were considered. Days to harvest were considered as 180, 195, 210 and 225, while the minimum market sizes (MMS) were 350, 400, 450 and 500 g.

The economic results using an average growth model and under a fixed price of USD$ 2.24 per kg, regardless of the MMR, is negative in both HT and HM planting strategies. With variable price, the highest net profit in HM was generated in the harvest strategy (day 225,500g) with USD$ 5,061.91 and USD$ 5,220.53 in HT. While the lowest net profit in HM was found in the harvest strategy (day 180, 450g) with USD$ 1637.53 and USD$ 1,714.54 in HT, respectively.

Under quantile regression analysis, and lowered a fixed price, the harvest strategy (day 225, 350g) showed higher yields in HM and HT with USD$ 4,323.51 and USD$ 4,190.90, while the lowest, for both planting strategies in that order, was in (day 210, 500g) with USD$ 708.38 and USD$ 1,175.56, respectively.

Considering a variable price, the harvest strategy (day 225,450g) showed higher yields in HM and HT with USD$ 8,634.03 and USD$ 8,983.26, while the lowest, for both planting strategies in that order, was in (day 210, 350g) with USD$ 3,788.77 and USD$ 3,580.18, respectively.

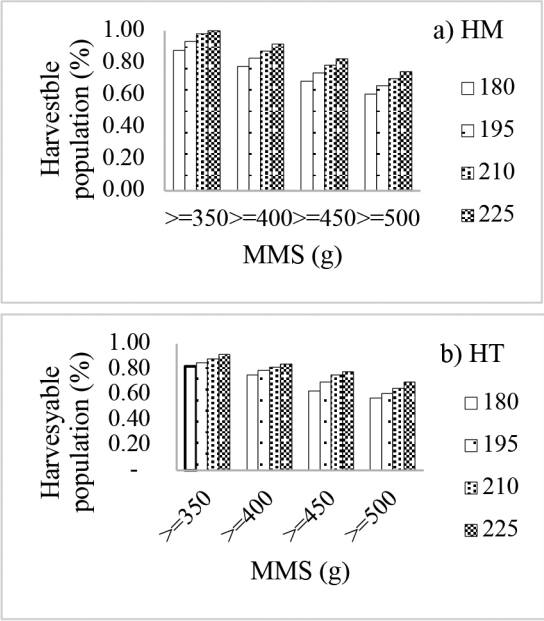

Figura 1. Población total cosechable considerando todos los cuantiles de acuerdo con las TMM, en los días de cosecha en cada estrategia de siembra HM y HT.

Nannochloropsis sp. AS FEED ADDITIVE FOR THE PACIFIC WHITE SHRIMP: EFFECT ON MIDGUT MICROBIOLOGY, THERMAL SHOCK RESISTANCE AND IMMUNOLOGY

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This work aimed to evaluate *Nannochloropsis* spp. as feed additive in the diet of Pacific white shrimp (*Litopenaeus vannamei*) for their effect on midgut microbiology, thermal shock resistance and immunological parameters. Initially, the digestibility of the microalgae meal was assessed, and the apparent digestibility coefficient (APC) was determined. The APC was, in general, high in lipids (78.88%) and eicosapentaenoic fatty acid (73.86%). Then, *Nannochloropsis* spp. were included in diets at four levels (0, 0.5, 1 and 2% inclusion). The shrimp were reared in 500 L clear water tanks containing 20 shrimp per tank with initial weight of 6.05 ± 0.06 g and fed four times a day. Shrimp fed with supplemented diets containing *Nannochloropsis* spp. (0.5 and 2%) presented higher resistance to thermal shock when compared to the non-supplemented group (Figure 1). Shrimp fed with 1 and 2% of algae inclusion had higher production of reactive oxygen species (ROS) when compared to other treatments. No statistical difference was observed in immunological parameters and microbiology of the intestinal tract. Thus, the inclusion of *Nannochloropsis* spp. in shrimp diets at 0.5% and 2% levels increase resistance to thermal shock and ROS production in shrimp.

Financial support: CAPES (Financing code 001) and by the Aquavitae project (Horizon 2020, number 818173

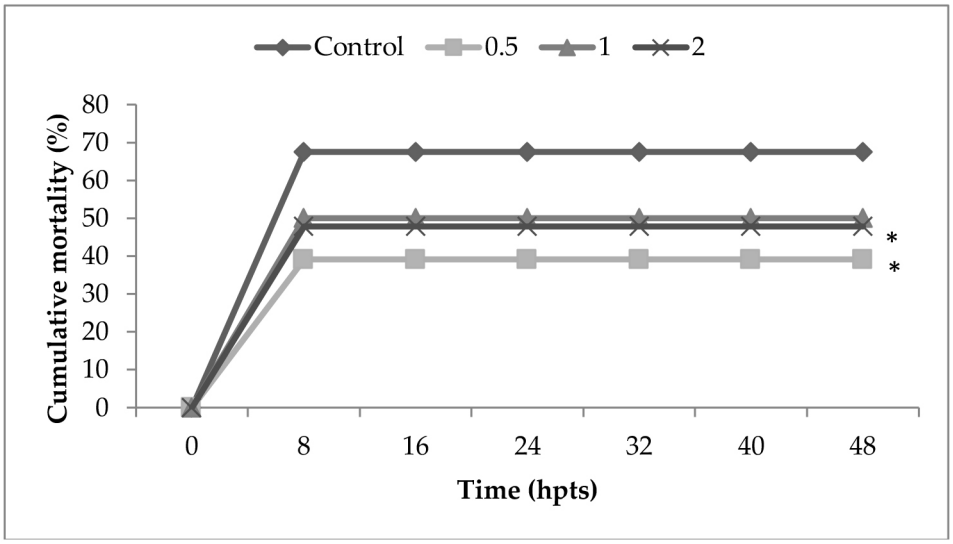

Figure 1: Cumulative mortality of shrimp fed during 15 days with diets containing 0.5, 1 and 2% of *Nannochloropsis* spp. and control diet (0% inclusion). *corresponds to statistical difference in comparison to control ($p<0.05$).

INFLUENCE OF CONVENTIONAL AND ADVANCED EXTRACTION TECHNIQUES ON POLYSACCHARIDE YIELD, CHEMICAL STRUCTURE, MOLECULAR PROPERTIES AND ANTIOXIDANT CAPACITY FROM ALGAE *Fucus virsoides* AND *Cystoseira barbata*

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Fucoidan, a sulfated polysaccharide from brown algae, exhibits a wide range of biological activities that depend on its chemical structure and are closely correlated with the extraction technique used. The conventional extraction technique (CE) is time consuming and requires high temperatures, so the use of advanced extraction techniques is a promising alternative. The aim of this study was to extract polysaccharides from the brown algae *Fucus virsoides* and *Cystoseira barbata* using 5 different extraction techniques (CE, microwave-assisted extraction (MAE), pressurised liquid extraction (PLE), ultrasound-assisted extraction (UAE), and non-thermal plasma (NTP)) under previously defined optimal conditions. The influence of these techniques on polysaccharide yield (%PS), chemical structure (fucose, sulfate groups, uronic acids), molecular properties (weight average molecular weight (M_w), number average molecular weight (M_n), polydispersity index (PDI)) and antioxidant capacity (ORAC) was studied.

The highest %PS for both algae was obtained with PLE and the lowest with NTP. The highest antioxidant capacity was obtained with MAE, while UAE had the highest M_w and, together with NTP, the highest sulfate group content and the lowest concentration of uronic acids. The application of PLE and MAE reduced the extraction time from 3 hours to 30 minutes.

Table 1. Influence of algal species and extraction technique on polysaccharide yield (%PS) and their chemical composition

		%PS	Fucose (%)	Sulfate group (%)	Uronic acid (%)
		$p \leq .05$	$p \leq .05$	p=0.43	$p \leq .05^*$
F. virsoides	CE	18.53±0.43^{e,f}	41.54±0.82^{g}	28.46±2.11^{a}	20.06±0.36^{g}
	MAE	20.42±0.43^{f}	48.48±0.82^{h}	37.13±2.11^{a,b}	15.93±0.36^{f}
	PLE	24.22±0.43^{g}	60.08±0.82^{i}	51.82±2.11^{c,d}	5.32±0.36^{c,d}
	UAE	12.14±0.43^{c}	14.75±0.82^{c}	83.37±2.11^{e}	1.77±0.36^{a,b}
	NTP	6.10±0.43^{a}	9.49±0.82^{b}	88.31±2.11^{e}	3.68±0.36^{b,c}
C. barbata	CE	16.29±0.43^{d,e}	22.53±0.82^{d}	35.53±2.11^{a,b}	15.72±0.36^{f}
	MAE	15.27±0.43^{d}	26.61±0.82^{d,e}	45.56±2.11^{b,c}	12.52±0.36^{e}
	PLE	18.77±0.43^{f}	28.06±0.82^{e,f}	57.58±2.11^{d}	7.15±0.36^{d}
	UAE	11.80±0.43^{b,c}	31.70±0.82^{f}	90.44±2.11^{e}	1.16±0.36^{a}
	NTP	9.64±0.43^{b}	2.83±0.82^{a}	88.86±2.11^{e}	0.46±0.36^{a}

Results are expressed as mean±SE. Values with different letters within column are statistically different at $p \leq 0.05$.

Table 2. Influence of algal species and extraction technique on molecular properties and antioxidant capacity

		M_w (kDa)	M_n (kDa)	PDI (M_w/M_n)	ORAC (µmol TE g^{-1})
		$p \leq .05^*$	$p \leq .05^*$	$p \leq .05^*$	$p \leq .05^*$
F. virsoides	CE	693.43±25.88^{c}	264.42±11.77^{c}	2.62±0.06^{d}	37.14±0.24^{c}
	MAE	891.25±25.88^{d,e}	332.14±11.77^{d}	2.68±0.06^{d}	42.22±0.24^{e}
	PLE	521.72±25.88^{b}	149.64±11.77^{b}	3.49±0.06^{e}	21.07±0.24^{a}
	UAE	1133.78±25.88^{f,g}	500.16±11.77^{f}	2.27±0.06^{c}	19.97±0.24^{a}
	NTP	16.38±25.88^{a}	16.38±11.77^{a}	1.00±0.06^{a}	20.21±0.24^{a}
C. barbata	CE	766.00±25.88^{c,d}	322.87±11.77^{c,d}	2.37±0.06^{c,d}	34.75±0.24^{b}
	MAE	1252.19±25.88^{g}	681.34±11.77^{g}	1.84±0.06^{b}	38.62±0.24^{d}
	PLE	1031.94±25.88^{e,f}	415.75±11.77^{e}	2.48±0.06^{c,d}	20.25±0.24^{a}
	UAE	1651.52±25.88^{h}	616.65±11.77^{g}	2.67±0.06^{d}	34.35±0.24^{b}
	NTP	930.83±25.88^{e}	492.50±11.77^{f}	1.89±0.06^{b}	34.51±0.24^{b}

Results are expressed as mean±SE. Values with different letters within column are statistically different at $p \leq 0.05$.

BFT SYSTEM IMPROVES GOLDFISH *Carassius auratus* SKIN PIGMENTATION

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The objective of the present study was to evaluate the effect of biofloc system (BFT) and the carotenoid suplementation in the zootechnical performance and skin pigmentation of goldfish (*Carassius auratus*). For this purpose, the effects of two production systems (clear water and BFT) and two diets (with and without astaxanthin supplementation) were evaluated in a completely randomized 2X2 factorial design.

Goldfish juveniles (12.18±0.27 g) were divided into 24 experimental aquariums (15 L). Fish were fed twice a day (2% of body weight per day) with isoproteic (40 %) and isoenergetic (4250kcal.kg^{-1}) diets for 56 days. Asthaxanthin was used as a carotenoid source (80 mg.kg^{-1}). At the end of the experiment zootechnical performance indexes and pigment intensity (portable colorimeter) of the skin were evaluated.

There was no interaction between the systems and the diets (P>0.05) for any of the performance or skin pigmentation variables. No effect (P>0.05) on goldfish performance was observed (Table I). The cultivation in BFT system improved (P<0,05) the pigmentation of goldfish skin in relation to the clear water system (Table II). Carotenoid supplementation intensified fish pigmentation regardless of the culture system used.

Cultivation in the BFT system improves the goldfish skin pigmentation when compared to the clear water system. These results prove that the BFT system is an effective and promising technology for goldfish production. The BFT system promotes pigmentation but it can be intensified with exogenous sources.

Table I. Zootechnical performance indexes of goldfish (mean±standard deviation: n=12) produced in BFT and clear water systems, with and without carotenoid supplementation in the diet for 56 days.

	System (S)		Diet (D)	
	Clear water	BFT	Control	Astaxanthin
Weight gain (g)	3.96±0.50	4.03±0.51	4.05±0.39	3.94±0.59
Food conversion rate	3.79±0.42	3.76±0.42	3.72±0.35	3.83±0.48
Survival (%)	98.33±5.77	98.33±5.77	100±0.00	96.66±7.78

Different letters in the lines represent statistical difference by the Tukey test (P<0.05).

Table II. Color indexes in the skin of goldfish (mean±standard deviation: n=36) produced in BFT and clear water systems, with and without carotenoid supplementation in the diet for 56 days.

	System (S)		Diet (D)	
	Clear water	BFT	Control	Astaxanthin
L	73.89±2,43a	68.97±2.74b	71.73±3.97	71.13±3.26
A	3.49±1,85b	7.71±1.72a	4.74±2.49b	6.46±2.88a
B	37.63±3,24b	40.47±3.58a	37.38±2.44b	40.72±3.96a

Different letters in the lines represent statistical difference by the Tukey test (P<0.05). L: lightness (-100 black and +100 white), a (-100 green and +100 red), b (-100 blue and +100 yellow).

MIXED CULTURE MICRORGANISMS FERMENTATION AFFECTS SOYBEAN MEAL NUTRITIONAL CARACTERISTICS AND IMPROVES THE INTESTINAL MORPHOMETRY OF NILE TILAPIA *Oreochromis niloticus* JUVENILES REARED IN BFT SYSTEM RECEIVING PLANT-BASED DIETS

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The objective of this study was to evaluate the effect of plant-based diets containing different inclusion levels of a fermented soybean meal (FSBM) prepared with a mixed microbial culture on the intestinal morphometry of Nile tilapia (*Oreochromis niloticus*) juveniles reared in a BFT system. FSBM was evaluated at four inclusion levels (7%, 14%, 21% and 28%) and compared to a positive control diet with fish meal and a negative control diet with no animal protein nor FSBM.

Soybean meal was fermented with a mixed microbial culture containing *Lactobacillus acidophilus, Saccharomyces cerevisiae* and *Enterococcus faecium*. All diets were isoprotein (33% CP) and isoenergetic (4300 kcal GE/kg). The design was completely randomized with four replications. Nile tilapia juveniles (1.635±0.198 g) were distributed in 24 tanks (70 L) at a density of seven animals per experimental unit. The tanks were connected to a recirculation system equipped with heating and aeration. Water quality parameters were periodically monitored. After 54 days of the experiment the intestinal morphometry was evaluated.

The soybean meal fermentation increased lactic acid bacteria and yeast counts (Table I). A reduction in pH was observed in the FSBM and there was also an increase in soluble protein. In animals fed 21% and 28% FSBM diets, higher ($P<0.0,5$) villi height were observed compared to the other treatments (Figure I). In the inclusion of 28% of FSBM, the goblet cell count did not differ from the positive control, but it was higher compared to the other treatments.

The utilization of a mixed microbial culture increased the probiotic microorganisms count and improved nutritional characteristics of FSBM. In the inclusions above 21% of FSBM, there was also evidence of improvement in the intestinal health of Nile tilapia juveniles.

Table I. FSBM chacteristics and composition

	Soybean meal	FSBM
Dry matter (%)	88.35	90.82
Gross energy (kcal kg^{-1})	4,259.09	4,383.85
Crude protein (%)	44.63	47.09
Soluble protein (mg g^{-1})	19.90	40.80
pH	6.64	6.06
LAB (log CFU g^{-1})	0	6.47
Yeast (log CFU g^{-1})	0	7.43

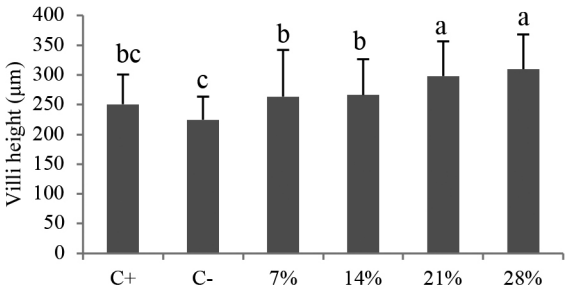

Figure I. Villi height of Nile tilapia juveniles fed FSBM.
$y=-0.038x^2 + 3.994x + 227.99$; $R^2=0.95$

FERMENTATION OF SOYBEAN MEAL WITH *Lactobacillus acidophillus* ALLOWS GREATER INCLUSION OF PLANT PROTEIN AND REDUCES *Vibrionacea* BACTERIA IN THE INTESTINE OF SOUTH AMERICAN CATFISH *Rhamdia quelen*

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The objective of this study was to evaluate the effect of diets containing different inclusion levels (0, 7, 14, 21 and 28%) of soybean meal fermented by *Lactobacillus acidophilus* (SMFL) on the zootechnical performance and intestinal bacteria count of South American catfish juveniles (*Rhamdia quelen*). The experimental design was completely randomized with five treatments and four replications and lasted 56 days.

Five isoproteic (39% crude protein) and isoenergetic (4,300 kcal of gross energy kg^{-1}) diets were formulated where SMFL was included in replacement of fish meal. 240 South American catfish juveniles (3.0±0.5 g) were distributed in 20 tanks (70 liters) connected in a recirculation aquaculture system.

At the end of the experiment the inclusion of SMFL up to 21% in replacement of fish meal did not affect ($P>0.05$) the weight gain (Figure I.A) and also decreased ($P<0.05$) the concentration of Vibrionaceae bacteria present in the intestine compared to the control group (Figure I.B). The amount of total lactic and heterotrophic bacteria did not differ between dietary treatments.

The results demonstrate that fermentation with *Lactobacillus acidophillus* enables greater inclusion of soybean protein in South American catfish diets and promotes the control of intestinal pathogenic bacteria.

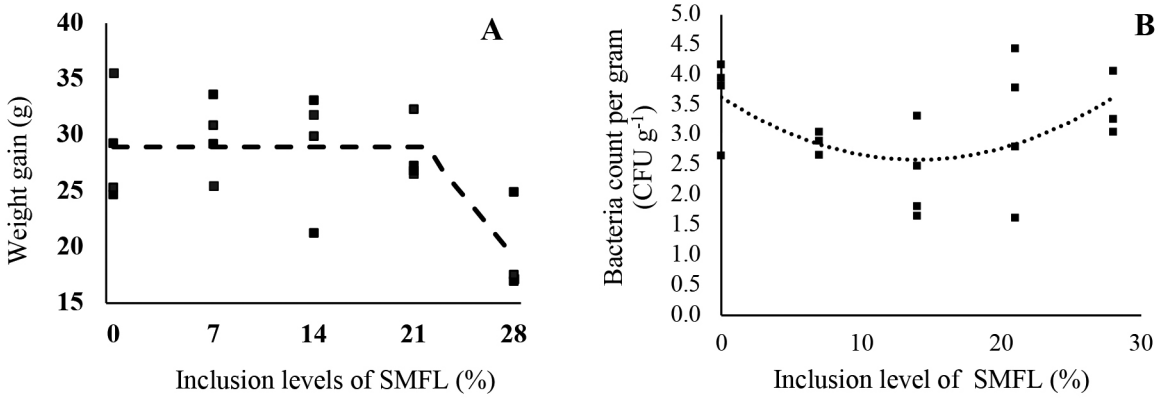

Figure I. Weight gain (A) an Vibrionacea intestinal bacterial count (B) of South American catfish fed with diets containing different levels of soybean meal fermented by *Lactobacillus acidophilus* (SMFL). Weight gain: $Y = 28.9506 - 1.5489(X - 21.6785)$, $Y = 28.9506$ if $X < 21.6785$. The breakpoint of the broken-line is 21.6% ($P = 0.0002$); $Y = 0.0053x^2 - 0.1484 + 3.6279$, $R^2 = 0.2755$, $P = 0.0316$. Vibrionacea count: $Y = 0.0053x^2 - 0.1484 + 3.6279$, $R^2 = 0.2755$, $P = 0.0316$.

ENZYMATIC COMPLEX IMPROVES THE PERFOMANCE OF NILE TILAPIA *Oreochromis niloticus* JUVENILES FED WITH DIETS CONTAINING DIFFERENT PROTEIN SOURCES

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The aim of this study was to evaluate the effect of diets containing two protein sources (plant and animal) and different levels of an enzymatic complex (0, 50 and 75g/ton) in the zootechnical performance of Nile tilapia *Oreochromis niloticus* juveniles. The experimental design was completely randomized in a 2x3 factorial scheme with four replications.

The enzyme complex consists of a combination of amylase, xylanase, beta glucanase, beta mannanase and protease. All diets were isoprotein (32% CP) and isoenergetic (4100 kcal GE/kg). Fish meal were used as a animal protein source, and soybean meal was the main ingredient in the plant based diet. Nile tilapia juveniles (approximately 5,25±0,22 of initial weight) were distributed in 24 tanks (70 L) at a density of ten fish per tank. The tanks were connected to a recirculation system equipped with heating and aeration. After 90 days of the experiment the zootechnical performance was evaluated.

No interaction ($P>0.05$) was found between the factors for any evaluated parameter. Regarding the source of protein, diets containing animal protein improved ($P<0.05$) the growth, feed ingestion and feed efficiency compared evaluated when compared to plant based diets. The higher inclusion of the enzymatic complex increased ($P<0.05$) the growth and feed ingestion, Survival was not affected($P>0.05$) by the treatments.

From the results it can be concluded that the protein sources affects the zootecnical performance of Nile tilapia juveniles. Supplementation with enzymatic complex improved growth and feed intake. The weight gain response was proportional to the level of enzyme supplementation.

Table 1. Mean values (± standard deviation; n= x) of the performance variables.

	protein source		enzymatic complex (mg kg^{-1})		
	AP	VP	0	50	75
FBW (g)	142,72±18,33a	104,94±11,48b	113,60±20,09b	124,64±18,00ab	133,25±31,76a
WG (g)	137,47±18.32a	99,66±11,47b	108,40±20,07b	119,39±17,89ab	127,91±31,90a
SGR	1,49±0,06a	1,36±0,04b	1,40±0,07	1,42±0,07	1,46±0,1
FI (g)	131,12±18,15a	114,16±10,06b	113,80±9,57b	122,73±10,26ab	131,39±23,45a
FCR (g)	0,95±0,05a	1,15±0,1b	1,07±0,13	1,04±0,11	1,05±0,14
S (%)	90±12,8	90±9,53	91,25±6,40	87,5±12,81	91,25±13,56

Different letters on the same line indicate a significant difference ($P<0.05$) within the factors (protein source and enzyme level) by Tukey's test. FBW – final body weight; WG – weight gain; SGR– specific growth rate; FI – individual feed ingestion; FCR – feed conversion ratio; S: survival.

GELLING TEMPERATURE OF ENDEMIC MAYAN OCTOPUS (Octopus maya) MUSCLE OF THREE LOCATIONS OF YUCATAN

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Octopus maya is an endemic species of the Yucatan peninsula of great sociocultural and commercial importance, being its capture and trade two of the main economic activities in the region. Even though Octopus vulgaris is the most common octopus' species worldwide, 74 % of the octopuses captured on the Yucatan peninsula are O. maya. One of the most important aspects for handling octopus is the gelling temperature that is directly related to its cooking temperature since, when cooked at the wrong conditions, their meat becomes tough, compromising the consumer's acceptance. The cooking temperature of mollusks ranges from 55-60 °C, but there is scarce information about the one of octopuses. Therefore, the object of this work was to determine the gelling temperature of O. maya muscle and study the changes on its structure based on its viscosity and location.

Samples were made by blending three octopuses into a paste, each from different ports of Yucatan (Coloradas CL, Rio Lagartos RL and Cuyo CU). The rheological behavior of the octopus' paste was characterized using a rheometer (DHR2, TA Instruments; EE.UU.) with a plate-plate geometry (40 mm) and a 1,050 mm gap. The samples were allowed to rest at the rheometer for 5 min/25 °C before the analysis; a vapor trap was attached to the geometry to reduce water evaporation. Temperature ramps were run on the samples over the range 25 - 90 °C (rate= 5 ºC/min; shear rate= 2 s^{-1}).

The temperature ramps showed that all three samples had a similar temperature at which their viscosity started to raise (Figure 1 Table 1) with no statistically significant difference, as well as the final temperature. The viscosity of the samples varied (CL 2562, RL 729 and CU 491 Pa·s), being the octopus from CL statistically significant different from the others. This could be caused by the location, for the environment feed or for the lack of control on the age of capture of the octopuses. By the change of the samples viscosity, it can be inferred that internal structure modifications, that can be associated to its cooking temperature, take place. Figure 1. Temperature ramp for octopuses´ samples.

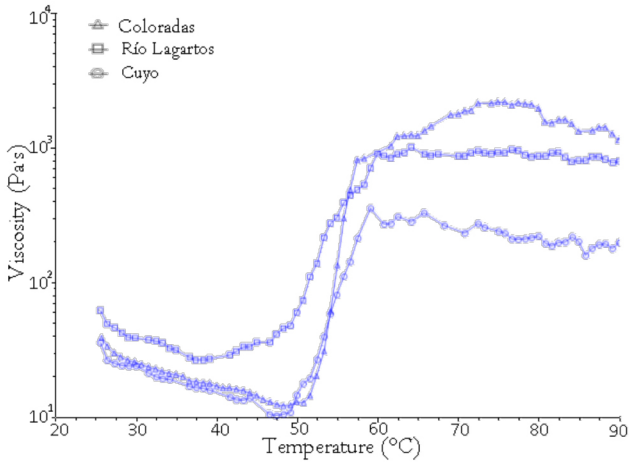

Figure 1. Temperature ramp for octopuses´ samples.

Table 1. Temperature ramp data for octopuses´ samples.

Port	Initial temp	Final temp
Río Lagartos	49.941 ± 1.16	66.631 ± 3.53
Coloradas	50.780 ± 1.17	70.814 ± 2.35
Cuyo	52.44 ± 1.15	69.098 ± 4.72
Average	51.054 ± 1.45	68.848 ± 3.41

The range of gelling temperatures obtained (51-68.8 °C) were slightly different to the cooking temperature range reported for mollusks (55-60 °C). O. maya needed higher temperatures to reach their maximum viscosity (8.8 °C). This information is of vital importance in heated processes as cooking or making industrial products with octopus.

EFFECT OF EARLY CO-FEEDING ON THE PERFORMANCE AND DEVELOPMENT OF DIGESTIVE SYSTEM OF TOTOABA (*Totoaba macdonaldi*) LARVAE AND JUVENILES

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Introduction. The production of marine fish larvae depends on the use of live food such as rotifers and *Artemia*, among others. This increases the difficulty and complexity for fish production. For these reasons, acceptable growth rates cannot be maintained using solely live food due to the low nutrient content (1). Also, acceptable growth rates cannot be maintained using live feeds exclusively due to the low nutrient content and restricted intake of these feeds (1). On the other hand, the use of microdiets in larviculture can improve the nutrition of the larvae especially in terms of amino acids profile and protein quality, therefore, implementing co-feeding is expected improved the nutritional status of the larvae and could facilitate the transition to only microdiets (2). The objective of the present study was to evaluate co-feeding during the early development of totoaba (*Totoaba macdonaldi*) larvae under culture conditions.

Method. Twelve 100-L fiberglass tanks in a recirculating system were supply at a density of 150 larvae/L. Four feeding treatments were performed (T1, T2, T3 and T4): T1 consisted of using the feeding protocol proposed by (3) and T2 doubled the live feed ration according to this protocol. T3 consisted of co-feeding with rotifer-microdiet from Day Post Hatch (DPH) 4, and T4 was to start co-feeding at DPH 12 with nauplii of *Artemia*-microdiet (Figure 1). Larvae growth (g and cm) and survival were evaluated and the development of the digestive system.

Results and Discussions. No significant differences (P>0.05) were observed on growth, length, and survival. The best growth (g and cm) and survival results were observed in T1 and T2 compared to T3 and T4 (Table 1), however, regarding the ontogenic development, a better development, larvae quality and conformation of the digestive system was observed in the T3 and T4, therefore, based on these results, we could suggest implementing co-feeding from DPH 12, as an early weaning protocol for totoaba larvae, contributing with a promising strategy for the production of this species.

Conclusions. Based on the results of the present study, totoaba larvae could be fed with a co-feeding (*Artemia*-microdiet) from DPH 12, having growth rates similar to those reported in previous protocols.

Table 1. Biological parameters analyzed at DPH 14 and 28 of totoaba larvae fed with different feeding protocols.

Parameters	Treatment 1	Treatment 2	Treatment 3	Treatment 4
Growth (mg) dph 14	17.9 ± 4.6	22.3 ± 8.2	11.2 ± 6.1	12.6 ± 1.2
Growth (mg) dph 28	108.7 ± 35.7	90.9 ± 7.5	85.6 ± 1.6	81.4 ± 2.7
Lenght (mm) dph 14	6.6 ± 0.8 [a]	5 ± 0.0 [b]	4.8 ± 0.5 [b]	4.7 ± 0.5 [b]
Lenght (mm) dph 28	12.7 ± 5.7	9.9 ± 1.8	7.8 ± 1.1	6.8 ± 1
Survival (%)	13.3 ± 8.7	10.6 ± 8.7	4.31 ± 0.6	3.93 ± 0.5

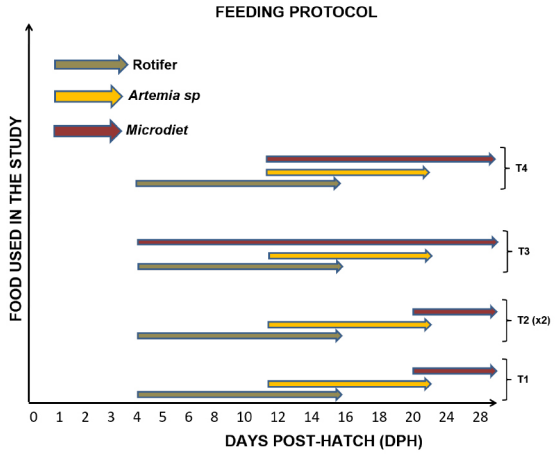

Figure 1. Feeding protocol during the study

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DESARROLLO Y EVALUACIÓN *IN VITRO* DE UN IMPLANTE HORMONAL DE LIBERACIÓN PROLONGADA PARA LA INDUCCIÓN DEL DESOVE EN PECES

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La acuicultura de peces marinos es una actividad productiva que ha adquirido gran importancia durante las últimas décadas; sin embargo, su desarrollo enfrenta diversas problemáticas. Dentro de las más importantes se encuentran las alteraciones en la reproducción durante el cautiverio, sobre todo en las hembras. Para resolver estos problemas, desde hace varias décadas se ha recurrido al uso de terapias hormonales en diversas especies. En la actualidad se utilizan análogos de la hormona liberadora de las gonadotropinas (GnRHa) dado su resistencia a la degradación enzimática en comparación con la GnRH nativa. Sin embargo, aunque existen implantes para la liberación de GnRHa en peces, estos no son útiles para todas las especies de importancia comercial y los protocolos de preparación no son claros para su replicación en campo, lo que hace necesario el desarrollo de nuevos protocolos claramente descritos. El presente estudio tuvo como objetivo describir y comparar el perfil de liberación de GnRHa de implantes preparados con diferentes excipientes en pruebas *in vitro*: 1) 95% colesterol m/m, 5% celulosa m/m; 2) 1.5% alginato de sodio m/v-- metil éter metacrilato de polietilénglicol (PEGMA) v/v cubierto con etilvinilacetato (EVAc) m/m-inulina m/m, y 3) EVAc m/m. La cuantificación de la GnRHa liberada se hizo con el método del ácido bicinconínico (BCA). No se encontraron diferencias significativas en la magnitud de liberación tipo estallido entre los implantes de alginato y los de colesterol-celulosa, sin embargo, esta liberación se registró a las 6 y 48 h, respectivamente. El porcentaje acumulado liberado fue significativamente mayor (62.1%, p=0.003) en los implantes de alginato. Los implantes de colesterol-celulosa liberaron por 192 h, mientras que los de alginato liberaron por 72 h (Figura 1). La cuantificación de GnRHa liberada de los implantes de EVAc no fue precisa debido a que el método no era específico para GnRHa, sin embargo, se detectaron las proteínas liberadas hasta las 336 h después del inicio del ensayo (Figura 2). Finalmente, los implantes de 95% colesterol y EVAc resultaron con perfiles de liberación de GnRHa más prolongados comparados con el perfil de liberación de implantes de alginato, sin embargo, dado el potencial del alginato como excipiente, es conveniente continuar su evaluación incorporando otros componentes y coberturas para retrasar la liberación de compuestos activos.

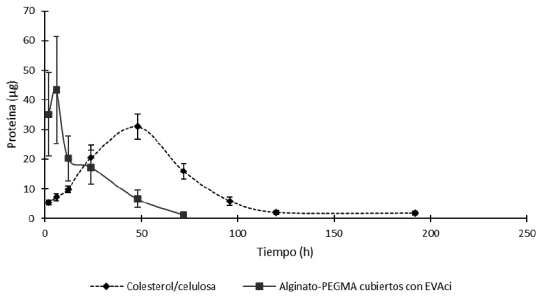

Figura 1. Perfil de liberación de proteína de los implantes de colesterol/celulosa (95 y 5%) e implantes con 1.5% de alginato de sodio-PEGMA cubiertos de EVAc/inulina. Cada implante contenía 200 µg de GnRHa. Promedio ± desviación estándar.

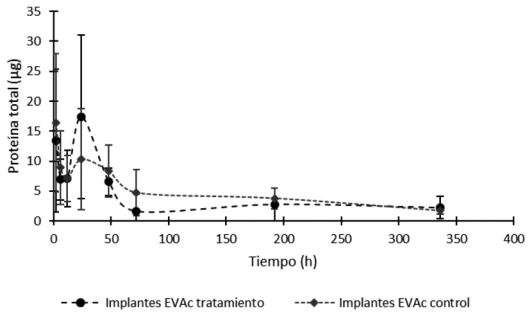

Figura 2. Perfil de liberación de proteína de implantes de EVAc. Cada implante contenía 171 µg de GnRHa. Promedio ± desviación estándar.

SEASONAL VARIATION OF AMINO ACIDS AND FATTY ACID COMPOSITION OF A POLYCHAETA Abarenicola pusilla AND ITS POSSIBLE USE AS AN ADDITIVE IN AQUACULTURE FEEDS

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This study aims to provide a quantitative analysis of amino acids (AA) and fatty acids (FA) on samples of the *Abarenicola pusilla* Polychaeta, taken at different seasons along the year, and under three types of processing (eviscerated carcass, pressed cake, and the liquid remaining).

The results showed a clear seasonal difference, where essential nutrients significantly decreased in the winter. Regarding the essential fatty acids, the eicosapentaenoic acid (EPA) and arachidonic acid (ARA) resulted in a relationship close to 1. In comparison, docosahexaenoic acid (DHA) was absent in some extracts during winter. At the same time, the highest protein values were observed in the spring and summer, while crude lipid values were the same, indicating that *A. pusilla* uses AAs more efficiently as energy source than lipids. Also, a significantly higher amount of free amino acids, essential (EAA) and non-essential (NEAA), were presented in winter.

The Polychaeta press cake was the product with the highest quality in terms of protein content and AA and FA composition, being a good source of functional amino acids LYS, GLU, GLY, ALA, and HIS. Also, the Polychaeta press cake would be a good source of EFA, such as EPA and ARA.

Finally, the conditions under which Polychaeta could be harvest and the type of extract recommended to produce a valuable additive for marine aquafeeds are also discussed. It is also proposed to study the culture possibilities into the integrated multitrophic aquaculture providing them the best possible conditions as spring and summer when the highest and most valuable nutrients are encountered.

EFFECT OF CO-SUPPLY WILD JUVENILE CRABS TO ENRICHED *Artemia* AS FOOD FOR THE PATAGONIAN RED OCTOPUS (*Enteroctopus megalocyathus*) PARALARVAE FROM DAY 15 TO 50 AFTER HATCHING UNDER CONTROLLED CULTURE CONDITIONS

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One of the most sensitive problems in the Patagonian red octopus (*Enteroctopus megalocyathus*) aquaculture, is the high mortality of paralarvae. Paralarvae are active hunters being highly selective until they reach the juvenile stage, around 90 to 120 days after hatching (DAH). *Artemia* is the food provided to paralarvae, with enrichment practices to provide better nutrients like essential amino acids and fatty acids.

The aim of the present study was to investigate the mortality preventive effect of diet in paralarvae between 15 and 50 DAH (i.e. from the maternal reserves have been depleted until the time before the benthic settlement) both through using juvenile crabs to co-feeding and varying diet of the previous feeding period. The growth, biochemical composition, and the expression of genes related with fatty acid biosynthesis, antioxidant defense system and stress response in paralarvae were evaluated after a experimental period of 35 days. To obtain paralarvae of 15 DAH two diets were used: only enriched *Artemia* (A), or enriched *Artemia* plus crab (AC), when paralarvae reached 15 DAH, each group was divided in two new groups that were fed until 50 DAH with enriched *Artemia* with or without co-feeding with crabs. Treatment names indicated the diet before and after 15 DAH, as follows: A-A, A-AC, AC-A, and AC-AC.

No differences were detected in weight growth, whereas those paralarvae fed A-A and AC-A, resulted in longer length. On the contrary, the highest survival was observed with both groups fed with crab juveniles at initial diet, with 40.5% for AC-AC, followed by AC-A with 34.8%, far higher than those A-A and A-AC (9.5 and 16.2%, respectively). Also, a trend towards a higher value in AL/ML ratio, highest values of 18:0, and the absence of 20:4n-6, were observed in paralarvae fed A-AC and AC-AC, without effect of the initial diet and without interaction between the diets of both periods. Of the genes analyzed, only the expression of the stress protein HsP70 showed a trend to higher values in paralarvae fed in the A-A the same with the highest mortality. It is discussed how co-feeding with juvenile crab resulting in an improvement compared to the use of pure enriched *Artemia*.

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SEA CUCUMBER AQUACULTURE IN LATIN AMERICA AND CARIBBEAN (LAC), PROBLEMS, PROSPECTS, AND FUTURE

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Fueled by high demand and dwindling stocks, sea cucumber (SC) aquaculture is growing worldwide. This increases the importance of its development and diversification. The LAC region has the potential for a substantial expansion in SC farming. To this end, some efforts have been made with limited success. This work explores recent advances with the leading commercial local species (*Isostichopus badionotus and Holothuria sp*.), compiling regional experiences for more than ten years, with historical, societal, institutional, and private background, identifying the main problems and providing options to solve them, suggesting future goals, objectives, and guidance.

With the interest from buyers in *I. badionotus, H. mexicana* and H. floridana fisheries, aquaculture research started in the Yucatan peninsula and expanded to countries like Colombia, Bermuda, Belize, Panama, and other. The private sector followed the institutions and startup ventures in Belize and Panama initiated. At each level, various problems have arisen, getting most projects to a halt.

Biological and environmental: Proper SC broodstock for aquaculture is scarce. It is challenging to keep them in captivity for a long time thereby limiting their reproductive potential for culture purposes. For these species, there are substantial information gaps about the nutritional requirements, diseases, parasites, and improving farming technologies for their growth and survival. Additionally, they face fishing and environmental pressures in the wild. More R&D and sustainable production methods like integrated multitrophic aquaculture (IMTA) are required.

Social: SC aquaculture faces the same social misconceptions as other aquaculture species: competition for coastal space, the perception of pollution, and conflicts with the fishery. There is a scarce specialized workforce for these activities, and, in some countries, security is a concern. New production efforts require community training and generation of simple low-tech for remote areas.

Policy: In many LAC countries, there is little to no institutional support for aquaculture, and yet, a highly complicated bureaucracy or no regulatory framework. It is critical to educate key decision-makers about sustainable mariculture, including IMTA approaches with sea cucumbers as the basis and their integration in the blue economy.

Private sector: Many of the stakeholders ignore SC aquaculture profitability potential. At the same time, some have hired consultants from other regions without the required knowledge of local species and ecosystems, generating the perception of high risk, making them prefer other investments. Promising, simple, cheap technologies and procedures should generate more interest.

Conclusion

Sea cucumber aquaculture could be one of the main economic and societal drivers for many LAC countries. However, a groundwork multi-disciplinary approach is needed involving every stakeholder in an organized and conscientious manner.

EFFECT OF TEMPERATURE AND CULTURE DENSITY ON THE GROWTH AND NUTRITIONAL QUALITY OF TILAPIA (*Oreochromis niloticus*) IN RAS

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The development of sustainable production systems, that uses less water and energy consumption, are become important in recent years. Among these options based on aquaculture, it can be mentioned recirculating aquaculture systems (RAS). Studies regarding stocking densities of tilapia crops have been carried out in RAS; those studies shows that in intensive crops, physical variables such as water temperature influence the growth of the fish, but also affected the quality of the meat produced. Previous research have not reported the effect of these variables on intensive crops, considering the maximum cultivation capacity for tilapia without sacrificing production or product quality. Thus, the aim of this study is to determine the optimal conditions of stocking density and water temperature to obtain the highest levels of growth and nutritional meet quality of tilapia in a recirculating aquaculture system.

Optimal conditions to obtain the highest growth levels and nutritional quality on tilapia (*Oreochromis niloticus*) in a recirculating aquaculture system were evaluated. During 60 days, fish were cultivated considering three cultivation densities (67, 135 and 270 fishes/m^3) and water with and without temperature control (28 °C and environmental temperature). Water quality was measured through the determination of temperature, pH and dissolved oxygen. Biometric data were registered throughout the growth of fish, along with the determination of moisture, total ash, total lipids and protein on tilapia fillet. Results showed that temperature and stocking density did not impact on survival rate of tilapia fish; however, specific growth rate for the farmed fish was higher at 28 °C (Table 1). In addition, higher moisture percentage values were observed in tilapia fillet cultivated at 28 °C; while, those fillet obtained from cultivation at room temperature showed percentage higher values of ash and total fat. There was no significant difference between the treatments respect to percentage of total ashes and protein (Table 2). Treatments with the lowest density, with and without temperature control, presented the greatest significant difference in percentage of total fats. Overall, according to the results a culture with a density of 270 fish/m^3 (equivalent to 80 kg/m^3) to harvest of tilapia can be carried out without affecting the growth and quality of product, as long as there was a control system for physical conditions of the fish culture, such as temperature at 28 °C and oxygenation, main

Table 1.
Growth performance parameters of *O. niloticus* for 6 treatments of stocking density and temperature during 60 days (mean ± DS)[z]

Treatment						
Performance parameters	T1	T4	T2	T5	T3	T6
	TC	WTC	TC	WTC	TC	WTC
Inicial number (n)	20	20	40	40	80	80
Final mean number (n)	20^{c}	20^{c}	39 ± 0.5^{b}	$39\pm.10^{b}$	79 ± 1.2^{a}	78 ± 1.5^{a}
Initial total weight (g)	33.3 ± 1.38^{a}	34.4 ± 0.77^{a}	33.93 ± 3.53^{a}	33.9 ± 3.81^{a}	37.71 ± 4.25^{a}	36.05 ± 2.60^{a}
Final total weight (g)	574.8 ± 109.4^{a}	283.9 ± 44.4^{bc}	600.5 ± 107.8^{a}	288.5 ± 20.1^{bc}	438.0 ± 48.5^{ab}	196.4 ± 28.5^{c}
Initial individual mean weight (g)	1.66 ± 0.48^{a}	1.72 ± 0.38^{a}	1.70 ± 0.38^{a}	1.70 ± 0.39^{a}	1.89 ± 0.49^{a}	1.80 ± 0.43^{a}
Final individual mean weight (g)	28.74 ± 9.31^{a}	14.49 ± 4.71^{c}	30.02 ± 12.07^{a}	14.43 ± 5.33^{c}	21.90 ± 7.63^{b}	9.82 ± 2.61^{c}
Weight gain (g)	541.6 ± 108.3^{a}	255.4 ± 43.2^{bc}	566.6 ± 105.9^{a}	254.6 ± 19.4^{bc}	400.3 ± 44.4^{ab}	160.4 ± 26.9^{c}
Specific growth rate (%day)	4.73 ± 0.26^{a}	3.54 ± 0.28^{b}	4.78 ± 0.25^{a}	3.57 ± 0.18^{b}	4.09 ± 0.04^{b}	2.82 ± 0.18^{c}
Survival rate (%)	100.0 ± 0.0^{a}	100.00 ± 0.0^{a}	98.33 ± 1.44^{a}	97.5 ± 2.5^{a}	98.33 ± 1.44^{a}	97.92 ± 1.91^{a}

**Average values for each treatment followed by a superscript indicates that there is a significant difference (P <0.05).
** For the average final number the data was rounded.
*** TC (Temperature control), WTC (Without temperature control).

Tabla 2.
Results of bromatological analysis of *O. niloticus* for 6 treatments of stocking density and temperature during 60 days (media ± DS)[z]

Treatment						
Performance parameters	T1	T4	T2	T5	T3	T6
	TC	WTC	TC	WTC	TC	WTC
Inicial number (N)	20	20	40	40	80	80
Humidity (%)	78.72 ± 0.25^{ab}	77.86 ± 0.25^{b}	79.04 ± 0.20^{a}	78.2 ± 0.26^{ab}	78.63 ± 0.54^{ab}	78.19 ± 0.34^{ab}
Ash (%)	1.32 ± 0.004^{a}	1.37 ± 0.13^{a}	1.29 ± 0.02^{a}	1.31 ± 0.14^{a}	1.23 ± 0.104^{a}	1.41 ± 0.07^{a}
Total lipids (%)	4.50 ± 0.26^{d}	5.38 ± 0.38^{bc}	4.83 ± 0.13^{cd}	6.33 ± 0.13^{a}	5.51 ± 0.05^{b}	5.52 ± 0.09^{b}
Protein (%)	28.71 ± 1.16^{a}	26.87 ± 0.25^{a}	26.55 ± 1.36^{a}	25.48 ± 3.62^{a}	27.01 ± 3.17^{a}	25.32 ± 0.81^{a}

**Average values for each treatment followed by a superscript indicates that there is a significant difference (P <0.05).
** TC (Temperature control), WTC (Without temperature control).

CIRCULATING ncRNAs IN BLOOD PLASMA FROM RAINBOW TROUT (*Oncorhynchus mykiss*): INTEGRATIVE BIOMARKERS IN FISH NUTRITION.

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Among other factors, nutrition is one of the most determining factors in fish farming, representing 40–70% of the production costs and determining survival, growth potential and quality of fish. The proper development of nutritionally balanced diets relies on multifactorial and multidisciplinary studies to fulfill species- and stage-specific nutritional requirements. However, since each nutrient might have different roles (and requirements) depending on the tissue and biological process considered, the use of integrative biomarkers as hall mark of the global condition of farmed fish might be a powerful tool to improve feed formulation and fish physiology monitoring. Here, the use of circulating non-coding RNAs (ncRNAs) found in blood plasma from rainbow trout (*O. mykiss*) as integrative biomarkers was explored.

We studied the population of miRNAs as biomarkers of fish physiology when they were fed with diets formulated base on different alternative raw materials during > 60 days or under feeding/fasting conditions during 10 days or (Experiment 2).

In Exp. 1, miRNAs were isolated from 12 (6 from fish fed on diets containing fish meal (FM) and 6 from FM devoid diets) samples, while in Exp. 2 we studied 8 (4 from fed and 4 from unfed fish) samples, using the miRNeasy Serum/Plasma Kit. Library preparation was performed using < 100 ng of ncRNA, normalized and sequenced in an Illumina platform (single-end, 51 cycles).

Raw reads were bioinformatically processed and mapped/annotated against *Salmo salar* known miRNAs from miRBase v22.1/RNAcentral 14.0 databases. Novel miRNAs were predicted with miRDeep2 using USDA_OmykA_1.1 genome as reference. SnoRNAs, rRNAs and tRNAs were also found. An example of the output from Exp. 1 is presented in Table 1.

We will list the identified miRNAs being found differentially expressed in both experiments and the potential molecular pathways post-transcriptionally regulated by them. Using this approach, we were able get further insights on how diet and feeding regime might alter the expression of these miRNAs in blood plasma, and the potential physiological consequences.

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TABLE 1. Summary of the sequenced miRNAs in circulation in rainbow trout fed FM or FM devoid diets.

Sample	Trimmed reads (M)	Q30 (%)	Mapped reads*	Identified miRNAs
1-10C	12.5	95.3	151,753	294
1-2A	12.1	95.3	138,485	290
1-2B	11.7	95.4	149,491	287
1-2C	11.0	95.5	48,244	265
1-6B	11.0	95.5	157,593	289
1-9B	8.9	95.3	136,808	295
1-9C	9.3	95.4	122,959	291
2-10C	11.6	95.7	183,007	314
2-1B	12.4	95.4	144,622	306
2-7A	6.6	95.7	40,643	236
2-7C	14.1	95.4	265,300	318
2-9C	8.4	95.5	50,970	265

*Denotes mapped reads as miRNAs against miRbase; M, millions; Q30(%), ratio of bases that have phred quality score greater than or equal to 30.

THE USE OF NARBONNE VETCH *Vicia narbonensis* AS A LOCALLY PRODUCED CROP FOR RAINBOW TROUT *Oncorhynchus mykiss* DIETS

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Aquaculture´s sustainability relies on the identification and implementation of alternative raw materials to replace fish meal and oil. European countries (among others) also depends on the production of soybean meal (SBM) from third countries. Here we will review the research works conducted to explore the potential use of Narbonne vetch *(Vicia narbonensis)* meal (NVM) as a substitute of SBM in rainbow trout (*Oncorhynchus mykiss*) diets.

The evaluation of NVM in rainbow trout diets was developed in 3 research phases. In phase 1, the effects of the inclusion of NVM at 0%, 10% and 30% (C, A10 and A30, respectively) in diets (isoproteic 43%, isolipidic 18%) was explored. In phase 2, the substitution of SBM with 0% 33% and 66% of NVM was evaluated under pre-treated conditions with the exogenous enzyme rovabio® phytase or not in diets (C, A33, A33E, A66, and A66E, respectively; isoprotein 42%, isolipidic 18%). Both experiments were carried out in RAS units with 500 L tanks, with a daily feed intake of 3% and during a 63-days trial. In phase 3, the underlying mechanisms of the effects of SBM substitution by NVM were explored through the assessment of digestive enzyme activities (proteases, α-amylase, alkaline phosphatase), blood biochemistry (glucose, triglycerides, cholesterol, among others) and the quantification of the expression of genes involved in glucose metabolism.

We first reported that NVM can be safely included at 10% in rainbow trout diets (Fig. 1A); since 30% inclusion induced growth reduction and impaired status on the proximal intestine (Fig. 1B). Afterwards, we demonstrated that a 33% of SBM can be replaced by NVM when is pre-treated with the exogenous enzyme (Fig. 1C).

Altogether suggest that a 33% substitution of SBM by NVM can be used for rainbow trout diets when it was pre-treated with an exogenous enzyme.

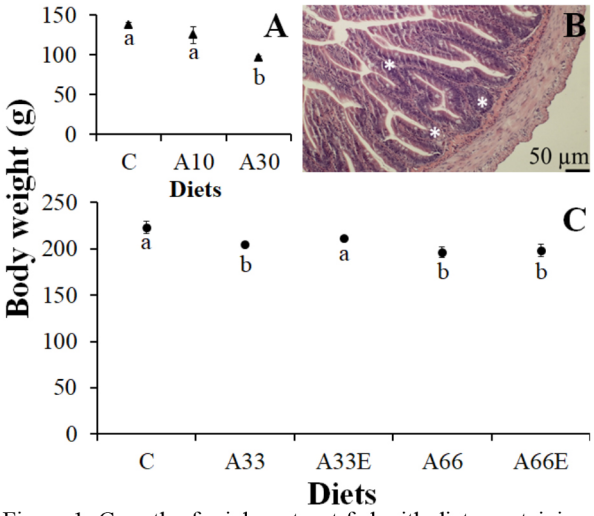

Figure 1. Growth of rainbow trout fed with diets containing Narbonne vetch meal (NVM) and histopathology of the proximal intestine. Final body weight (a) and histopathology of the proximal intestine showing high number of villi fusions when fish were fed with high NVM content in diets. Final body weight of fish fed diets were soybean meal (SBM) was partially replaced by NVM pre-treated or not with exogenous enzyme rovabio®phytase. Different letters denote significant differences among experimental groups (ANOVA, $p < 0.05$; $n = 3$).

Acknowledgments: This work was partially funded by "Optimización integral de los sistemas productivos en acuicultura: revalorización de materias primas locales en piensos y en cría de especies en potencial desarrollo (OPTI-ACUA)" project from the European Regional Development Funds (ERDF). F.J. T-S. acknowledges the National Council for Science and Technology (CONACYT) for the post-doctoral fellowship No. 2019–000012-01EXTV-00292. I.F. acknowledges the Ramón y Cajal (Ref. RYC2018-025337-I) contract from MICIU and the European Social Fund, "The European Social Fund invests in your future". Authors also thanks the support from the network LARVAplus (117RT0521) funded by the CYTED.

MEAL FROM *Lathyrus cicera L.* AS A SUSTAINABLE EUROPEAN CROP TO REPLACE SOYBEAN MEAL IN RAINBOW TROUT *Oncorhynchus mykiss* DIETS

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Aquaculture´s sustainability deeply relies on the identification and implementation of alternative raw materials (ARMs) to replace fish meal and oil. One of the main ARMs currently used in fish diets is soybean meal (SBM) that in some regions (e.g. Europe) has to be imported. Thus, the identification of locally produced crops to substitute SBM is an urgent need to reduce not only the dependency of SBM from third countries, but also to reduce the carbon footprint. Here a nutritional dose-response trial was performed to explore the use of red vetchling (*Lathyrus cicera*) meal (RVM) in rainbow trout (*Oncorhynchus mykiss*) diets.

A 90-day feeding trial was conducted to evaluate the effects of different substitution levels (0 %, 8%, 16%, 33%, 66% and 100%, named as C, T8, T16, T33, T66 and T100, respectively; isonitrogenous 42% and isolipidic 18%, diets) of SBM with RVM on growth, and feed utilization of juvenile rainbow trout. A total of 360 juvenile fish of 10 ± 0.5 g maintained under controlled parameters (temperature of 15 ° C, dissolved oxygen > 7 mg/L, etc.) in 500 L tanks connected to a RAS unit (20 fish/tank) were hand-fed at a daily ration of 3% body weight.

TABLE 1. Growth performance in rainbow trout fed with experimental diets.

	C	T8	T16	T33	T66	T100
IBW	10.4 ± 0.1	10.3 ± 0.1	10.4 ± 0.1	10.3 ± 0.2	10.4 ± 0.1	10.3 ± 0.1
IFL	9.5 ± 0.1	9.4 ± 0.1	9.5 ± 0.1	9.4 ± 0.1	9.4 ± 0.1	9.4 ± 0.1
BW	36.3 ± 1.1^{a}	35.9 ± 1.7^{a}	36.6 ± 1.2^{a}	36.8 ± 1.9^{a}	36.2 ± 0.5^{a}	31.4 ± 1.5^{b}
FL	14.6 ± 0.2	14.5 ± 0.3	14.5 ± 0.2	15.0 ± 1.3	14.5 ± 0.1	13.9 ± 0.2
SGR	3.0 ± 0.1^{a}	3.0 ± 0.1^{a}	3.0 ± 0.1^{a}	3.0 ± 0.1^{a}	3.0 ± 0.1^{a}	2.6 ± 0.1^{b}
WG	250 ± 10^{a}	250 ± 18^{a}	253 ± 12^{a}	256 ± 13^{a}	250 ± 9.0^{a}	204 ± 16^{b}
FCR	0.76 ± 0.03^{a}	0.76 ± 0.04^{a}	0.76 ± 0.03^{a}	0.75 ± 0.03^{a}	0.76 ± 0.02^{a}	0.89 ± 0.06^{b}

Values are expressed as mean ± standard deviation. Different superscript letters within each row denote significant differences among experimental groups (ANOVA, $p < 0.05$; n = 3). IBW, initial body weight (g); ITL, initial furcal length (cm); BW, body weight (g); FL, furcal length (cm); SGR, specific growth rate (% per d); WG, weight gain (%); FCR, feed conversion ratio.

Preliminary results presented here, obtained after 42 days feeding, indicate significant lower values for wet weight, WG, SGR and FCR only in those fish fed on the highest substitution (ANOVA, $p > 0.05$; Table 1). Although growth performances, as well as other parameters like the histopathological status of the digestive system (liver, proximal and distal intestines), blood biochemistry (glucose and triglyceride's plasma content) and muscle quality (proximal composition, amino acid and fatty acids profiles) will be assessed again at the end of the experimental period.

These results suggest that SBM could be partially replaced up to 66% by red vetchling meal in rainbow trout feeds.

Acknowledgments: This work was partially funded by "Optimización integral de los sistemas productivos en acuicultura: revalorización de materias primas locales en piensos y en cría de especies en potencial desarrollo (OPTI-ACUA)" project from the European Regional Development Funds (ERDF). F.J. T-S. acknowledges CONACYT for the post-doc fellowship No. 2019–000012-01EXTV-00292. I.F. acknowledges Ramón y Cajal (Ref. RYC2018-025337-I) contract from MICIU and the European Social Fund, "The European Social Fund invests in your future". Authors also thanks the support from the network LARVAplus (117RT0521) funded by the CYTED.

DECOUPLED AQUAPONICS SYSTEM USING PHOTOAUTOTROPHIC BIOFLOC

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Decoupled aquaponics is a strategy that allows combining the culture of animals and plants in a disruptive way. These systems present several advantages in using water, space, and residuals. In the same line, Biofloc technology (BFT) has shown to be a beneficial technique implemented in this production activity. It represents low-cost equipment and a versatile way to transform aquaculture waste into valuable products used in horticulture. Despite the favorable attributes of BFT, the typical combination was heterotrophic BFT and aquaponic, but the description of the strategies for integrated aquaponics and photoautotrophic BFT with *Chlorella* microalgae has not been completely explained. Diversifying the production system is a necessity in developing countries, which is why this investigation focus on the characterization of residuals from *O. niloticus* BFT in a photoautotrophic, heterotrophic, and control, and their relationship and strategies implemented to integrate these residuals with the NFT hydroponic horticulture using five green leaf plant species: lettuce (*Lactuca sativa*), pak-choi (*Brassica rapa* subsp. *chinensis*), rocket (*Eruca sativa*), spinach (*Spinacia oleracea*), and basil (*Ocium basilicum*). This experiment was performed in five weeks; it evaluated plant growth, production, parameters, the size, elements content in biofloc, and the diverse strategies to integrate these residuals (mineralization, flocculation, and filtration). During the experimental period, the photoautotrophic treatments showed the highest levels of micro and macronutrients among all treatments. According to growth percentage, lettuce and pak-choi did not show significant differences ($p > 0.05$), while for basil and spinach, the best results were obtained with photoautotrophic treatment ($p < 0.05$). The best combination was with spinach and/or pak-choi and/or lettuce. The relationship between N:P was suitable in the photoautotrophic treatments compared with hydroponic solutions. The ideal integration process was the combination of flocculation and filtration before adding the liquid fraction from BFT to NFT system. The aerobic-anaerobic reactor allows the attaining of essential micronutrients.

Therefore, *O. niloticus* BFT rearing in the photoautotrophic mode and its integration with decoupled aquaponics system is beneficial in arid coastal zones where water is scarce. It is necessary to improve aquaculture performance, reuse water, BFT nutrients, and increase profits.

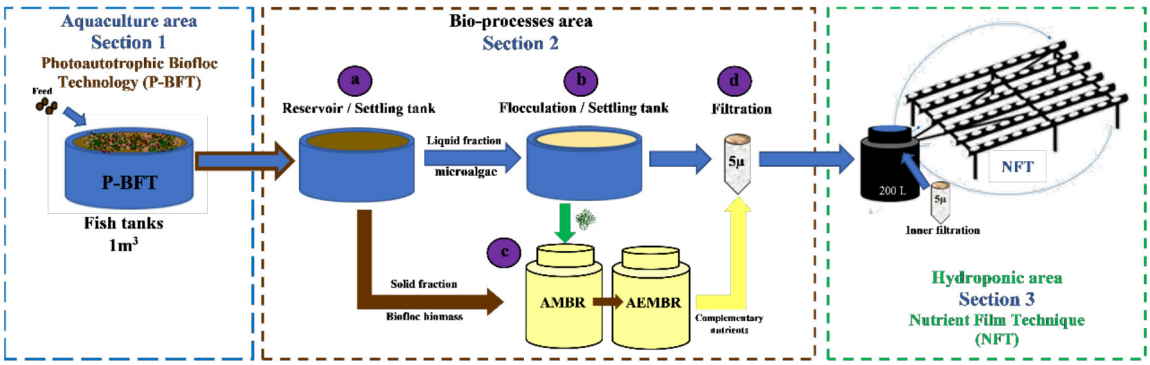

Figure 1. Bioprocesses to integrated BFT and aquaponics

COUPLING TECHNOLOGIES IN TILAPIA FARMING AT CIBNOR´S NAYARIT UNIT

Fimbres-Acedo, Y.E., Garza-Torres, R., Telles, A., Carrera-Alvarado, G. & Maeda-Martínez, A.N.

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Unidad Nayarit del Centro de Investigaciones Biológicas del Noroeste (UNCIBNOR) (Fig. 1) continues to consolidate itself as a center for technological development and innovation in Tilapia. Its facilities have been in continuous operation for more than three and a half years. In the period that has elapsed, the Unit continues to have a significant influence on the Tilapia value chain in Western Mexico by being depositories and distributors of one of the best genetic lines of Tilapia in the world, with operations in the facilities of our laboratory, dedicated to the production of offspring and juveniles of this species with Biofloc technology (BFT).

With this, it has been possible to provide the raw material required by aquaculturists, producers, and fishers organized in cooperative societies and rural production societies in western Mexico (Nayarit, Jalisco, Michoacán), Guerrero, Campeche, Hidalgo, Sinaloa, Zacatecas, and Baja California Sur, where the demand for our product has been increasing. Since the beginning of the Unit's operations, it has managed to sell approximately a little more than 15 million male tilapia hatchlings. Supporting more than one hundred producers, considering that UNCIBNOR continues to consolidate itself in this area and is the key to the development of the tilapia production industry in our country.

In addition, the advisory and technical assistance actions demanded by the producers have continued to increase, as well as the development of demonstration projects of the different existing technological alternatives for the production of tilapia. In the same way, in this period, the task of training students from regional higher education institutions has been increasing to contribute to the training of professional and technical staff required by aquaculture and fishing in our country. The personnel commissioned by Conacyt to UNCIBNOR have been working on integrating research proposals focused mainly on the optimization of farming techniques, the development of assisted tilapia fisheries in natural and artificial reservoirs, and the transfer of farming technologies to vulnerable groups in Mexico.

Figura 1. Photoreactors in Unidad Nayarit del Centro de Investigaciones Biológicas del Noroeste (UNCIBNOR).

THE CURRENT SITUATION AND CHALLENGES OF THE ABALONE FARMING INDUSTRY IN MEXICO

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Abalone are one of the most luxurious and expensive seafood products around the world, with high demand specifically in the Asian countries because of the cultural, traditional, and medicinal qualities associated with abalone. Abalone aquaculture over the last decade has increased its share of the world abalone market with the decline of abalone fisheries. Abalone culture technology is now well established in several countries, and the industry can be considered to be in its maturation phase.

The total volume of worldwide abalone fisheries has declined since the 1970's, but over the same period, farm production has increased from negligible quantities in the 1970s to more than 100,000 tons, the vast majority being produced in China and South Korea.

Other relevant abalone producing countries are South Africa, Chile, Australia, USA and Mexico.

In Mexico, the annual abalone cultured production has been stable, less than 100 tons. The number of new entrants also have been stable for the past two decades.

This presentation gives an overview of the development of the Mexican abalone industry. It discusses options and challenges for expanding and to sustain the abalone industry in Mexico.

CRYOPRESERVATION OF GONADAL TISSUE OF WHITE SNOOK *Centropomus viridis*

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The white snook *Centropomus viridis* is one of the main artisanal fisheries in the Gulf of Mexico with high farming potential. In 2018, the farming of white snook was successfully achieved in Mazatlan, Mexico. However, its aquaculture production is still limited due to a lack of biological and ecological information. Therefore, the cryopreservation of germ cells will contribute to developing assisted reproduction techniques to optimize white snook farming and production. Also, germ cells' cryopreservation will help conserve it since *C. viridis* is considered on the International Union for Conservation of Nature's red list as a species of minor concern.

Thus, the objective of this work was to develop a cryopreservation protocol for gonadal tissue of white snook. Pieces of gonadal tissue were suspended in Ethylene glycol (1.5 M or 2 M), Dimethyl sulfoxide (1.5 M or 2 M), or the combination of Dimethyl sulfoxide 1.5 M with the addition of lactose (0.1 M or 0.2 M), and 10% egg yolk. Then, samples were cooled at -80°C for 10, 15, or 30 minutes and plunged into liquid nitrogen (-196°C). Gonadal tissue was thawed, enzymatically dissociated with 0.25% trypsin, and germ cells enriched using a Percoll density gradient (40% and 10%). Cell viability will be determined with double staining of fluorescein diacetate and propidium iodide. The identification of germ cells will be made by immunohistochemistry technique using the protein VASA as a molecular marker. The results will be presented at the meeting.

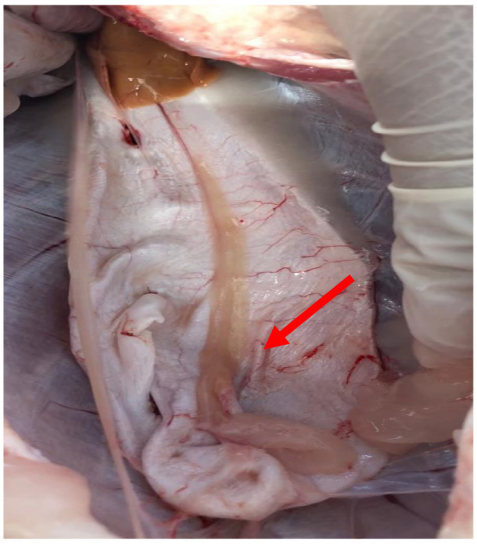

Fig. 1: Gonadal tissue of white snook *Centropomus viridis*

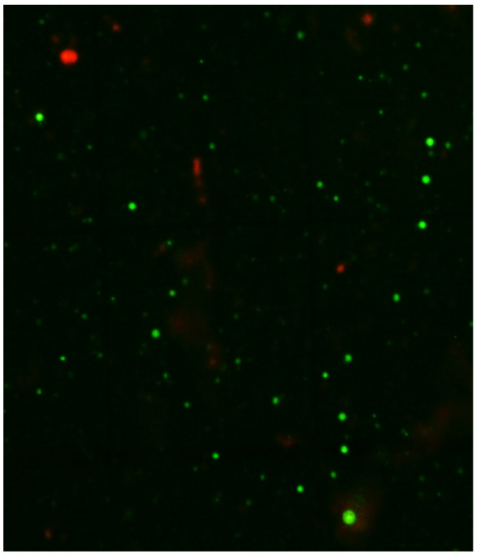

Fig. 2: Gonadal tissue cells stained with fluorescein diacetate (viable green cells) and propidium iodide (nonviable red cells)

EFFECT OF DIETARY PROTEIN, LIPID AND CARBOHYDRATE RATIO ON GROWTH, DIGESTIVE AND ANTIOXIDANT ENZYME ACTIVITIES OF FRESHWATER PRAWN *Macrobrachium acanthurus* JUVENILES

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Freshwater prawn *Macrobrachium acanthurus* is an American native species suitable for aquaculture. The present investigation was conducted to evaluate the effect of two levels of protein and three lipid to carbohydrate ratios on growth, digestive and antioxidant enzyme activities and thus contribute to the formulation of feeds that improve health and growth of juveniles.

180 juveniles (initial weight of 70.4 mg) were randomly stocked in 18 tanks with a water recirculation system. Six treatments were formulated with two levels of protein (35 and 40%) and three combinations of lipids (L) and carbohydrates (C) to a ratio of 2:1 (L= 15% and C= 8%), 1:2 (L= 10% and C= 20%) and 1:6 (L= 5% and C= 30%) in a 2x3 factorial design. Treatments were tested by triplicate and prawns were fed twice a day for 8 weeks. Length and weight were measured bimonthly for growth calculations. Homogenates of juveniles cephalotorax were prepared at the end of the feeding trial for assaying digestive and antioxidant enzyme activities. Data was tested for normality and homoscedasticity before running a 2 way ANOVA using GraphPad Prism v.8.0.2 software.

Juveniles fed with 35% of protein and a 1:6 L:C ratio attained the highest weight (330.4 mg). Protease activity was higher with 40% of dietary protein associated to the increased amount of substrate (Figure 1). Amilase activity decreased significantly with the 1:6 L:C ratio, suggesting that 30% exceeded carbohydrate requirements affecting digestibility. Total antioxidant capacity was highest with P35 and L:C ratio 1:2, and a ratio of 1:6 increased phenoloxidase, prophenoloxidase and catalase to a peak (Figure 2), suggesting oxidative stress. Results evidence that a diet containing 35% of crude protein, 10% of lipids and 20% of carbohydrates (L:C ratio 1:2) helps to improve health and growth in juveniles of *M. acanthurus*.

Figure 1. Digestive enzyme activity of *M. acanthurus* juveniles. Results are mean of triplicates ± std. dev. Different upper case literals indicate significant differences (p <0.05) between group fed 35% of protein (light blue). Upper case literals (dark blue) represent group fed with 40% crude protein.

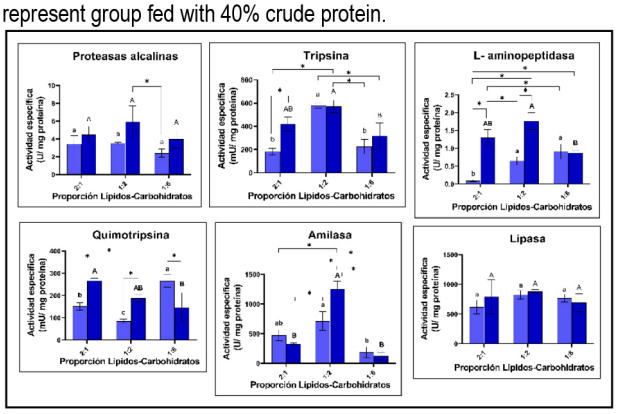
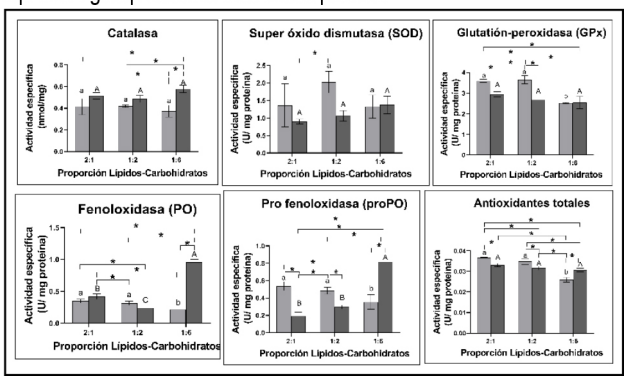


Figure 2. Antioxidant enzyme activity of *M. acanthurus* juveniles. Results are mean of triplicates ± std. dev. Different upper case literals indicate significant differences (p <0.05) between group fed 35% of protein (light gray). Upper case literals (dark gray) represent group fed with 40% crude protein.



ASSESSMENT OF PROBIOTIC BACTERIA FROM MARINE COASTS AGAINST ACUTE HEPATOPANCREATIC NECROSIS DISEASE (AHPND)

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Outbreaks caused by *Vibrio parahaemolyticus* (VP_{AHPND}) emerged in Mexico in 2013, causing a 65% drop in shrimp tonnage produced and reaching 95% mortality. This work aimed to isolate the best bacterial strains from Mexican marine ecosystems to assess the microbiota antagonism against VP_{AHPND} and to choose strains with protective roles against this pathogen for an environmentally friendly aquaculture treatment.

Bacterial marine agar and trypticase soy agar isolates were obtained from invertebrates and different Sonora coast ecosystems. Sampled and reisolated pathogens were analysed by qPCR. Among 258 bacterial isolates, only 17 showed antagonist activity *in vitro*, but only five, 32a, 43, H-A, Y119, and Y100, were chosen based on cell-free supernatant assays (Fig. 1). Bioassay results with white shrimp highlighted *Vibrio alginolyticus*-32a with a 4.76% cumulative mortality, followed by *Bacillus pumilus*-Y100 (12.54%) and *Vibrio campbellii*-H-A (16%); *Bacillus pumilus*-Y119 and *Bacillus pumilus*-43 did not show any protection or beneficial effect against VP_{AHPND} (Fig. 2). Pathogen reisolation on CHROMagar Vibrio and qPCR were performed only in the positive control and H-A; however, 32a, Y100, Y119, 43, and a negative control yielded creamy pink and green-blue colonies based on the absence of VP_{AHPND}, which was confirmed with a negative qPCR (Fig. 3). This result suggested that they were able to inhibit the pathogen with a microbicidal effect.

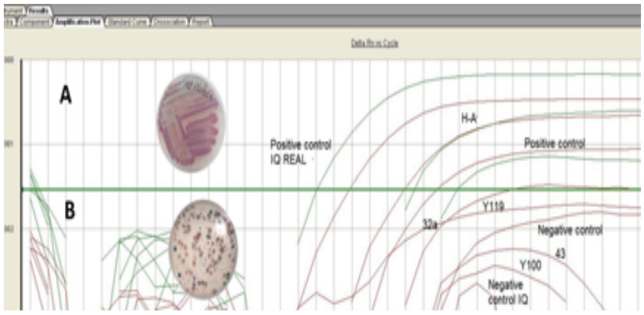

Figure 1. Antagonism assay by the well diffusion test. Averages of triplicate results for bacteria with antagonistic activity. The results are expressed by the length of the inhibitory diameter, which were analysed with Dunnett's multiple comparisons test (95% CI; $P < 0.05$). Error bars at the top of each column indicate the standard deviation of the mean.

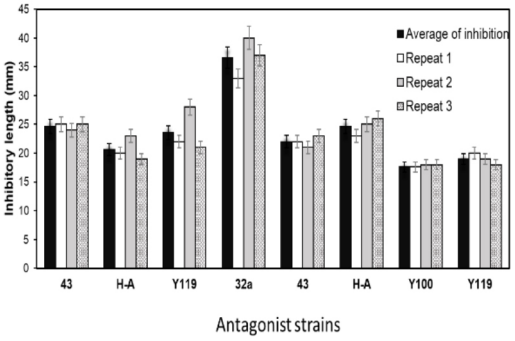

Figure 2. Trials of inhibitory strains against VP_{AHPND} in shrimp (*Litopenaeus vannamei*). Only the 32a and Y100 strains showed a protective role as good probiotics.

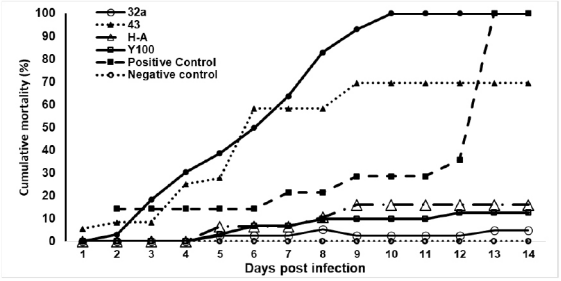

Figure 3. CHROMagar Vibrio and qPCR results. Only H-A showed the presence of VPAHPND with a positive qPCR result, while the probiotic strains 43, 32a, Y100, and Y119 demonstrated a microbicidal effect with a qPCR negative result.

SILAGE FISH WASTE AS A CULTURE MEDIUM FOR THE PRODUCTION OF THE MICROALGAE *Tetraselmis chuii*

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The microalgae *Tetraselmis chuii* is part of the live foods used in the larval rearing of fish, crustaceans and molluscs. Its nutritional quality is closely related to culture media, with F/2 Guillard being the most widely used in aquaculture production centers. Fish waste silage can be an important alternative to commercial culture media as it contains high nutrient contents. In this study, the effect of partial and total replacement of the F/2 Guillard by chemical ensilage of fish waste on the growth of the microalgae *T. chuii* was evaluated.

Fish waste from the "canané" snapper (*Ocyurus chrysurus*) obtained from riverside fishermen from the port of Sisal, Yucatan, Mexico, was used. After being homogenized, formic acid was added at a ratio of 20 ml kg (v/p) and the initial pH and during the 30-day silage was recorded (Gallardo et al, 2012). The silage was carried out in triplicate using containers of 2 liters capacity.

Two bioassays of partial and total replacement of the F/2 Guillard medium were carried out, evaluating the population growth of *T.chuii* for 10 days under controlled conditions with 500 ml Erlenmeyer flasks. In the first experiment, seawater was fertilized at the beginning of the bioassay prior to inoculation. In the second bioassay, the silage was added on both day 0 and day 3 (Table 1).

A maximum value of concentration of *T. chuii* was observed on day 5 with treatments CT, A and B whose average value of 1.3 x 10^{-6} was much higher than that obtained when only the silage was used (0.5 x 106) (Fig. 1a). The results of experiment 2 show that the silage was beneficial for the growth of microalgae, allowing concentrations similar to those obtained with the commercial medium (Fig.1b).

Table 1.- Design of experiments 1 and 2 of the partial and total replacement of the F/2 Guillard culture medium for the growth of the microalgae *Tetraselmis chuii*

Treatment	Description	n
Control	F/2 Guillard (Sol. A + Sol B)	3
A	Sol A + 50% Sol B + 50% silage	3
B	Sol A + silage	3
C	Silage	3

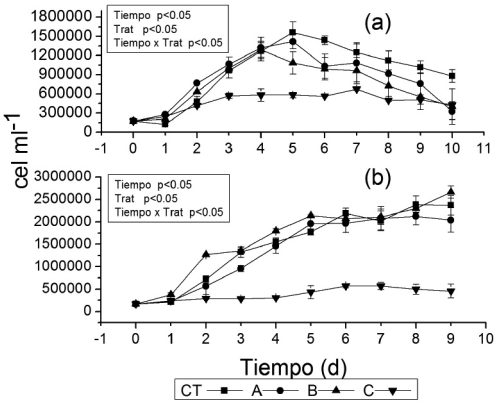

Fig. 1.- Growth (cell ml^{-1}) of the microalgae *Tetraselmis chuii* cultivated with fish waste silage. (a) Exp. 1 and (b) experiment 2. Mean ± S.E. n=3.

Literature

Gallardo P., …., and Sotelo A. 2012. Nutritive value of diets containing fish silage for juvenile Litopenaeus vannamei (Bonne, 1931). *Journal of the Science of Food and Agriculture* (DOI 10.1002/jsfa.5632). 2320 -2325

Acknowledgments

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ADVANCES IN THE DEVELOPMENT OF PELLED DIETS FOR THE FEEDING AND NUTRITION OF THE *Octopus maya*

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Octopus maya being a species with direct development, high growth rate and adaptation to captive conditions make it a potential and suitable species for cultivation. In recent years, the use of semi-moist and dry feeds have allowed high survival and growth rates (Martínez et al 2014, Gallardo et al, 2020). The selection of inputs, as well as the conditions of the food preparation process have been decisive for obtaining the pelleted feed, for which in this work the results obtained from 3 experiments carried out with different diets elaborated with fish discards (*Haemulon plumieri*). A control diet based on a mixture of squid and crab (Gallardo et al, 2020) was used.

For each experiment, newly hatched organisms from a single spawn (0.1 g ww) were used. The fish discards were homogenized and incorporated into a mixture with vitamins, minerals and gelatin as a binder. Three different protein hydrolysates made with discarded fish ("chac chi", "canane" *Ocyurus chrysurus*) and crab (*Callinectes spp*) were tested, which were incorporated into the diets for each of the experiments. The pellets (0.5 cm) were fed (3 times day-1) for 90 days.

The survival and growth rates obtained in the three experiments were similar to those observed with the control diet (mean 65 % and 4.5 % day-1 respectively) ($p>0.05$). With the weight values obtained up to 90 days (average of 16.5 g), a growth projection was made, which shows that the octopuses can reach a size greater than 60 g in 150 days after hatching (Fig. 1). The results obtained indicate that fish discards added with the different proteic hydrolysates can be a good alternative for the development of food for octopus farming.

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Martínez R., … and Carlos Rosas. 2014. Growth, survival and physiological condition of Octopus maya when fed a successful formulated diet. *Aquaculture* 426-427: 310 – 317.

Acknowledgments

This work was carried out with the support of the DGAPA-UNAM, Project PAPIIT IT 201611 under the responsibility of Dr. Pedro Gallardo

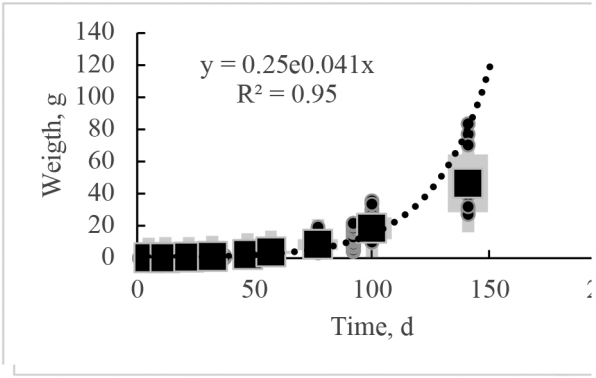

Fig. 1.- Growth curve of *Octopus maya* juveniles fed with diets made with discarded fish and protein hydrolyzate.

ESTUDIO HEMATOLOGICO DE *Petenia splendida* (GÜNTER, 1862) CÍCLIDO NATIVO DE IMPORTANCIA COMERCIAL EN EL ESTADO DE TABASCO, MÉXICO

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Los parámetros hematológicos son herramientas en la determinación del estado de salud y equilibrio metabólico en los peces, tanto de vida silvestre como en los cultivos intensivos. Entre las especies nativas más importantes del estado de Tabasco que poseen un gran potencial para la acuacultura, se encuentran el cíclido conocido localmente como mojarra "tenguayaca". El objetivo del trabajo fue determinar los valores hematológicos de poblaciones de *Petenia splendida*

Los peces fueron adquiridos en granjas acuícolas y trasladados vivos en neveras con aireación al laboratorio de Acuacultura; fueron anestesiados y se les tomaron los datos de peso y longitud total. La sangre fue obtenida del pedúnculo caudal con jeringas heparinizadas y colocada en tubos vacutainer con anticoagulantes. Los parámetros sanguíneos se midieron empleando técnicas hematológicas descritas para peces Los resultados fueron analizados mediante estadística descriptiva (Programa spss).

Un total de 120 peces fueron muestreados, los rangos de pesos oscilaron entre 110 y 420 g y longitudes entre 21 y 28 cm. Los resultados obtenidos se muestran en la tabla 1. Cabe mencionar que para la especie estudiada no hay trabajos hematológicos reportados, pero los resultados obtenidos están dentro de los rangos encontrados en las Tilapias y otros peces teleósteos.

Tabla 1. Valores hematológicos medios de la tenguayaca (*Petenia splendida*)

Parámetro	Unidades de Producción					
	A	B	C	D	E	F
Microhematocrito (%)	24± 4.6	19.7±3 .4	26.3 ±4.3	23.5 ±7.9	31.3 ±3.2	30.2 ±3.8
Hemoglobina (g/100ml)	9± 1.6	8.0 ±1.5	10.8 ±.1. 4	6.8± . 2.1	7.06 ± 1.7	8.74 ±1.4
Proteína total del plasma (g/dl)	5.48 ±0.8	3.9± 1.5	6.3± 0.3	6.4± 0.9	6.7± 0.5	7.2± 0.4
Recuento de eritrocitos (cel/mm^3)	2.5 $X10^6$	1.4 $X10^6$	1.5 $X10^6$	1.52 $X10^6$	1.1 $X10^6$	1.8 $X10^6$
Recuento de leucocitos (cel/mm^3)	53.9 $X10^3$	39.9X 10^3	31.3 $X10^3$	48.1 $X10^3$	41.5 $X10^3$	36.5 $X10^3$

TILAPIA FARMING BASED ON PERIPHYTON AS A NATURAL FOOD SOURCE

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Nutrient-rich effluents from aquaculture can be reused to enhance the development of natural food to feed fish in periphyton-based systems. A periphyton-based system is a strategy to increase fish farming efficiency, reducing feed use and effluent disposal.

This study aimed to evaluate different types of feeding restriction management in periphyton-based systems to produce Nile tilapia in ponds. To do this, two new production models were compared: (P100-0) Nile tilapia fed 100% of the recommended amount of feed sharing effluent with a tilapia production in a periphyton-based system with no feed input, and (P50-0) Nile tilapia fed 50% of feed plus periphyton sharing effluent with a tilapia production in a periphyton-based system with no feed input.

Biomass gain and productivity were significantly higher in P100-0 than in P50-0, and no differences in the feed conversion ratio and survival were found. For both production models, fish fed with feed and periphyton-based fed corresponded, respectively, to 75% and 25% of the total productivity of each model. The tilapia growth performance indicates that the proposed production models are promising strategies for using natural food in a periphyton-based system and reusing effluents from monocultures, especially when inputting a high amount of nutrients as in P100-0.

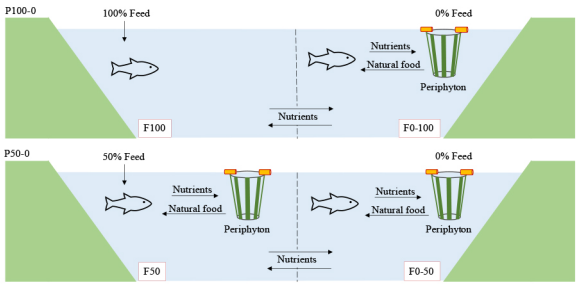

Figure 1. Feeding management adopted during the trial. P100-0: on the left side, fish were fed only feed (F100) sharing effluent with the right side where fish were periphyton-based fed with no feed input (F0-100). P50-0: on the left side, fish were fed feed every other day (50% of feed) plus periphyton (F50), sharing the effluent with the right side in which fish were fed at a periphyton-based system (F0-50).

Table 2. Productive parameters (mean ± SD) found for each production model after 112 days of the experimental period.

Parameter	Production Model					
	P100-0			P50-0		
FCR	1.37	± 0.20		1.46	± 0.39	
BG (kg)	47.18	± 7.40	a	20.15	± 5.86	b
Survival (%)	99.14	± 0.29		99.33	± 1.15	
PROD (kg/m^2)	0.48	± 0.04	a	0.30	± 0.04	b

FCR: Feed conversion ratio; BG: Biomass gain; PROD: Productivity.

FISH FEED DEVELOPMENT USING BLACK SOLDIER FLY AND MICROALGAE AS TOTAL FISHMEAL REPLACEMENT FOR TILAPIA CULTURE

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Fishmeal is consider the main protein source in fish feed, that is why the high demand has affected its production and costs for aquafeeds development, questioning meal supply for the next years. Currently, there is a need to find new alternative inputs to replace fishmeal without affecting the benefits that the latter provides to fish nutrition. Between the most relevant options are black soldier fly larvae (BSFL) and microalgae due to its protein, lipids, vitamins, minerals and antioxidant content; they had been used as partial replacements of fishmeal with favorable results on fish growth performance, however, they had not been used successfully as total fishmeal replacement and neither combined in the same formulation. Therefore, the objective of this study was to develop a balanced fish feed with BSFL and microalgae according to the nutritional requirements of Nile tilapia fry.

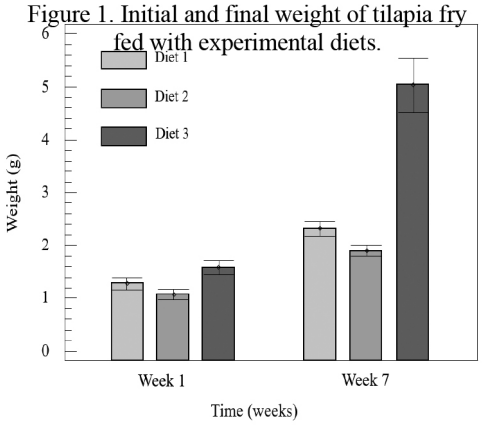
Three isoproteic (45%) diets were formulated with 5, 10 y 15% of BSFL and 5% of microalgae *Nannochloropsis limnetica*; additionally, experimental diets were balanced with animal and vegetal meals as corn, soybean, wheat gluten, wheat bran and meat and bone meal (Table 1). All diets were mixed, pelletized and dried in an oven at 60°C for 24 hours. Feeding trials were conducted for 7 weeks in a recirculation system conformed with tanks of 12 liters, controlled culture conditions of oxygen (5 mg/L), temperature (28°C) and water quality recirculated through an external filter with chemical and mechanical agents. Each tank contained 30 fish of the specie *Oreochromis niloticus* (1±5 g) and each experimental diet was assigned to triplicate tanks. The experimental diets were compared with a commercial feed.

After 45 days, all experimental diets were well accepted by the fish. Figure 1 shows initial and final weight obtained during the experiment. Fish fed with Diet 3, which contained 15% of BSFL, resulted in a weight increased with significant difference (p<0.05) compared with the other alternative diets. Based on these results, the combination of BSFL at 15% of the diet with 5% of microalgae can be used as a total fishmeal replacement without causing unfavorable effects on fish growth.

Table 1. Formulation of the experimental diets

	Diets		
	1	**2**	**3**
Microalgae	5	5	5
Black soldier fly larvae	5	10	15
Meat and bone meal	32	27	25.5
Soybean	30	30	26.5
Wheat gluten	10	10	10
Wheat bran	10	10	10
Corn	5	5	5
Black soldier fly oil	3	3	3

Figure 1. Initial and final weight of tilapia fry fed with experimental diets.



VARIATIONS ON DIGESTIVE ENZYMATIC ACTIVITY OF THE LONGARM RIVER PRAWN *Macrobrachium tenellum* ADAPTED FROM THE WILD TO CULTURE WITH PREPARED MEALS

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Macrobrachium tenellum is an omnivorous freshwater prawn living in rivers near the coast and in coastal lagoons with potential to be cultured. Since they are capable of living in rivers, estuaries and coastal lagoons, in each of these they have access to foods with different nutritional content. In this work, it has been tested if this has an immediate effect on their digestive enzymatic activity once they are brought to laboratory-controlled conditions. Wild prawns were captured in a river and in a coastal lagoon. After acclimation, they were fed with three different dietary regimes consisting on 1) commercial pelletized food (Camaronina Purina®) with 35% protein, 2) prepared pelletized food with 35% protein and 3) mixture of fresh tilapia and squid flesh.

Total alkaline protease, trypsin, chymotrypsin, lipase and α amylase were measured from wild specimens and from specimens kept 15 and 30 days with the experimental diets.

Prawns collected from the wild had more intense enzymatic activity in comparison with those maintained in the laboratory with different diets on all enzymes. However, the proportion in which these enzymes are expressed in the prawns from the different treatments remains proportional to what was observed in wild specimens. We consider that the study of this phenomenon can contribute to a better understanding of the nutritional needs of this species if a species-specific diet needs to be prepared.

EL POLICULTIVO LANGOSTINO-TILAPIA, UNA OPCIÓN VIABLE PARA LA PRODUCCIÓN DE *Macrobrachium rosenbergii* EN MÉXICO

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Las características físicas, naturales y sociales que presentan las zonas tropicales y subtropicales de México son aptas para la producción acuícola en policultivo, específicamente con las especies de *Macrobrachium rosenbergii* y *Oreochromis* spp., ya que el conjuntar los dos cultivos permite usar una misma infraestructura, incrementando la productividad, presentándose como una opción con mayores beneficios económicos para el productor acuícola.

Desde el arribo del langostino malayo a México en 1973, el cultivo de este se promocionó principalmente a través del monocultivo, obteniendo resultados productivos buenos, pero no los esperados. Sin embargo, es una especie estudiada y domesticada que se cultiva con gran éxito en diversas partes del mundo, por lo que el desarrollo del cultivo es una alternativa para satisfacer la demanda y disminuir la presión que la pesca ejerce sobre los recursos naturales silvestres. Por ello, es el momento oportuno para promover una vez más la producción del langostino malayo, ahora en policultivo con tilapia, en las instalaciones creadas para el cultivo de ésta.

El objetivo del trabajo fue utilizar de manera eficiente los recursos disponibles en el estanque, ya que el policultivo de langostino-tilapia tiene un rendimiento neto superior al aprovechamiento de estas mismas especies producidas en monocultivo. De esta forma, el rendimiento es mayor por unidad de superficie sin incrementar significativamente los costos de operación ni la infraestructura, ya que, con la misma mano de obra y los mismos requerimientos energéticos en el uso de equipos de aireación, se logra incrementar la rentabilidad de la actividad acuícola en la producción del policultivo.

Figura 1. Utilidad del policultivo tilapia-langostino con diferentes densidades de siembra.

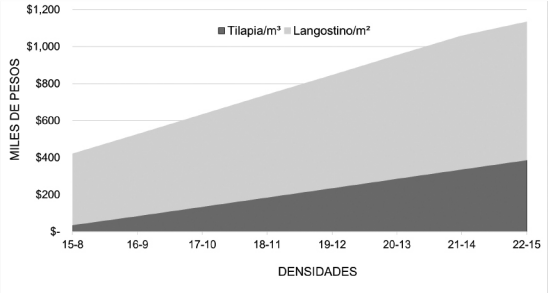
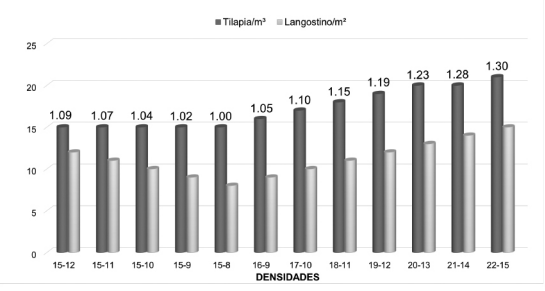


Figura 2. Relación beneficio costo en el policultivo tilapia-langostino con diferentes densidades de siembra.



EFFECT OF MACROALGAE AS A FUNCTIONAL INGREDIENT IN GROW-OUT DIETS ON THE BIOLOGICAL PERFORMANCE OF TOTOABA, *Totoaba macdonaldi*

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Currently soybean meal (SBM) and other plant ingredients are used to partially replace fishmeal in diets of carnivorous fish to reduce costs and the extraction of fishery resources. However, a high SBM content causes enteritis, the non-contagious subacute inflammation of the intestine, which also result in immune and physiological changes that in turn alter the absorption of nutrients and growth of some fish, such as totoaba. The totoaba is an endangered endemic fish from the Gulf of Baja California with a high potential for aquaculture.

On the other hand, prebiotics can prevent enteritis caused by SBM. These ingredients have the characteristic to be resistant to digestion and stimulate gut microbiota. Macroalgae contain polysaccharides that meet the characteristics of prebiotics, however, their effects are species-specific, so the aim of this work was to evaluate the effects of macroalgae in grow-out diets on the growth performance of juvenile totoaba, feed utilization, digestibility of the diets, body composition, enzyme activities (trypsin, lipase, amylase), immune responses (respiratory burst and leukocyte count), histological changes and gene expression (*il8*, *aqp8*, *IgM*). Four isoproteic (48%) and isolipidic (13%) diets were tested. Two commercial diets with local macroalgae meal (Alg1 and 2) and two experimental diets, both with 26% of SBM and one including a macroalgae prebiotic mix (SBM and SBM+Pre).

At the end of the trial fish fed with SBM and SBM+Pre diets resulted a significant higher growth and utilization of nutrients compared to the diets with macroalgae. Also, significant differences were found between the treatments in the histological samples of the distal intestine. There was expression of genes related to the immune system such *Igm* and *il8*, and cell structure such as *aqp8*, but only SMB+Pre had a lower level than the other treatments.

These results reveal that the commercial diets negatively affected the gut health of totoabas even more than diets that included SBM, however more research is needed to elucidate which compounds of these diets altered the gut and make clear if there is a prebiotic effect of the macroalgae on other species.

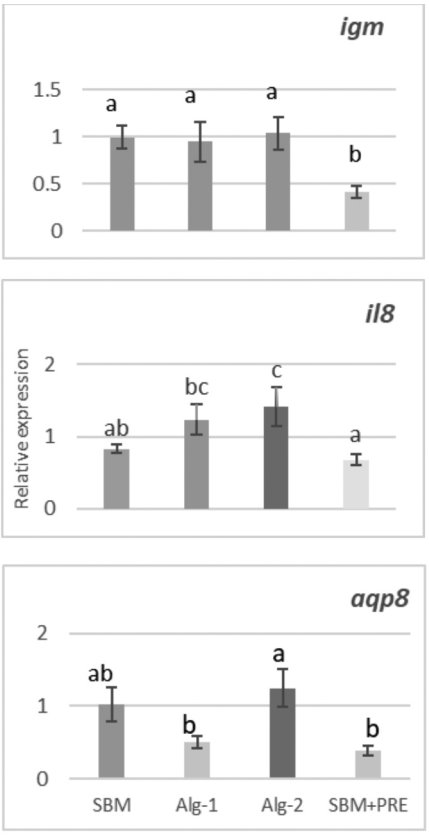

Figure 1. Immunoglobulin (*igm*), interleukin (*il8*) and aquaporin (*aqp8*) relative expression in *Totoaba macdonaldi* distal intestine fed with commercial and experimental diets. Different letters represent significantly different values ($p<0.05$)

EFFECT OF LOW AND HIGH SALINITY ON *IN VITRO* AND *IN VIVO* PROTEIN DIGESTIBILITY, GROWTH PERFORMANCE, FEED UTILIZATION AND SURVIVAL OF PRACTICAL FEED INGREDIENTS FOR WHITELEG SHRIMP *Litopenaeus vannamei* JUVENILES

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The aim of this study was to determine the effect of salinity on *in vitro-in vivo* digestibility, growth performance, feed utilization and survival of practical ingredients available in the industry, for whiteleg shrimp *L. vannamei* juvenile (5.0 ± 1.8 g). The ingredients used for the study were: fish meal (FM), squid meal (SQM), soybean meal (SBM), canola meal (CM), poultry by-product meal (PBM), meat and bone meal (MBM), horn and hoof meal (HHM), blood meal (BM) and casein (CA), and a reference diet (RD). *In vitro* and *in vivo* studies were conducted using the methods of the pH-stat with shrimp hepatopancreas enzymes, and zeolite was included as a marker, respectively. Results from both methods showed that the RD diet was the highest apparent protein digestibility. In low salinity, they were PBM and MBM diets and in high salinity, SBM, CM, PBM and MBM diets. The most significant relationship between apparent digestibility coefficients and growth performance was with the thermal growth coefficient parameter. There was greater growth in all diets in high salinity than in low salinity.

PARTIAL SUBSTITUTION OF FROZEN-FRESH FOOD BY AN EXPERIMENTAL DIET IN *Litopenaeus vannamei* (Boone, 1931) (Decapoda: Dendrobranchiata: Penaeidae) BROODSTOCK

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Declarations of interest: none

The aim of this study was to determine the effect of completely or partially replacing fresh foods from the broodstock diet with an experimental diet. During a 40-day period, three dietary treatments were tested on *Litopeaneus vannamei* broodstock. As part of the first dietary treatment, denoted as "FF", broodstock shrimp were fed only fresh frozen food (squid, polychaete, mussel and *Artemia* biomass). The second treatment denoted as "ED" was 100 % an artificial experimental diet. The third treatment, denoted as MD, comprised both the experimental diet and the fresh-frozen food (only squid and mussel were used). In terms of fertile spawns, females with ≥ 1 spawn, females with ≥ 2 spawn, and fecundity, the MD treatment did not differ significantly from the FF treatment. Fecundity was lowest among females receiving the ED treatment. MD treatment demonstrated equivalent fertility in females, and sperm rate in males to that of the FF treatment. The highest normal sperm rate was found in the ED and MD treatments. As a result, a combination of fresh food and the experimental diet resulted in a more balanced reproduction performance.

DOES UNILATERAL EYESTALK ABLATION AND CULTURE SYSTEM ORIGIN AFFECT PENAEID SHRIMP REPRODUCTIVE PERFORMANCE AND PROGENY QUALITY?

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For almost five decades, the unilateral eyestalk ablation (hereafter named ablation) has been used to accelerate the gonadal maturation of broodstock females from several species of penaeid shrimp (e.g., *Penaeus, Litopenaeus* and *Farfantepenaeus* species). And why not, if the ablation results in regular peaks of maturity and spawning, making it easier to plan production schedules in hatchery facilities. However, there is growing public and animal welfare concern about this procedure, which may result in its prohibition in the future. As a result, there is a need to induce a transition and paradigm shift regarding the employment of this technique. Despite efforts in the literature dating back to the 1980s to leave the shrimp to mature naturally, the findings were contradictory. However, broodstock management has evolved over time, and significant strides in broodstock output have been made (e.g. diet, pre-maturation culture systems and maturation management). As a result, there is a rising interest in investigating how ablation and broodstock origin - *alternative culture procedures* - affect reproductive performance and offspring quality. Over the last five years, on a series of experiments, our laboratory has investigated the effects of ablation and the biofloc system (*vs* clear-water) on a native penaeid shrimp species, *F. brasiliensis*, and an exotic species, *L. vannamei*. According to our findings, ablation increased female mortality in both species. When both species came from clear-water, the quantity of eggs produced by ablated and non-ablated females was comparable, however evidence suggests that *L. vannamei* produced more eggs when it was collected from biofloc. Furthermore, there was substantial evidence that non-ablated females generated higher quality offspring, and this was especially true for eggs produced by *L. vannamei* from biofloc origin. All these findings suggest that both ablation and origin may affect the reproductive performance and offspring quality of penaeid shrimp species, and that the lower number of spawning and predictability from non-ablated females can be remedied by increasing the number of reproductive females, which can live longer and produce higher-quality offspring.

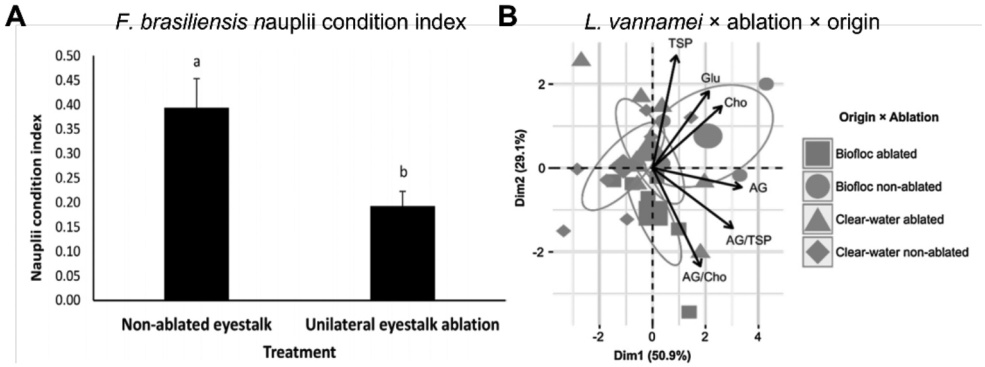

Figure 1 (A) F. brasiliensis nauplii condition index [(nauplii AG × % viable nauplii × nauplii lenght)/100] and (B) PCA of L. vannamei egg reserves.

FUNDAMENTAL CRITERIA FOR THE SELECTION OF INGREDIENTS FOR FEED FORMULATION

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Unidad Multidisplinaria de Docencia de Sisal, Facultad de Ciencias, Universidad Nacional Autónma de Mexico

To formulate balanced feed for aquatic organisms, it is necessary not only to know the proximal analysis, or the biochemical composition of the ingredients, but also the digestion potential, using enzyme extracts from the digestive tract (in vitro digestibility), its absorption capacity (apparent digestibility), its assimilation capacity (using direct methods, such as stable isotope of C and N) and evaluation methods of energy distribution. In this conference, different results obtained by the Aquaculture Nutrition Program of the UMDI Sisal, of the Faculty of Sciences of the UNAM, will be presented.

INCREMENTO DE LA CALIDAD BIOQUÍMICA DE LA HARINA DE PESCADO FERMENTADA CON EL HONGO *Aspergillus oryzae*

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La harina de pescado es una importante fuente proteica en la fabricación de alimentos destinados a la producción acuícola (NRC, 2011), esto gracias a su aportación nutricional y características organolépticas. Sin embargo, la disposición es cada día más crítica principalmente por su alto costo (Ayisi *et al*., 2017). El método de Fermentación en Medio Sólido (FMS), podría ser una alternativa para el aprovechamiento de subproductos de pescado o pescados enteros en la elaboración de harina de pescado (Pizardi, 1999). La harina de pescado fermentada representa una harina de bajo costos, nutrientes potencializados y un bajo contenido de compuestos tóxicos. En este estudio se evaluó la calidad bioquímica de la harina de pescado fermentada mediante el hongo *Aspergillus oryzae,* como fuente de proteína alternativa en la industria de nutrición acuícola.

Se comparó harina de pescado fermentada (HF) mediante el método de Alquicira (2003), contra una harina de pescado control sin fermentación (BCO), donde se analizó su composición nutrimental: proteína cruda (PC), energía bruta (EB), digestible (DE), metabolizable (ME) y neta (NE) mediante AMINONIR® NRG, así como el extracto etéreo (EE), ceniza (CZ) y fósforo (FS) mediante AMINONIR® PROX. El análisis de aminoácidos (AA) se llevó a cabo por el método AMINONIR®.

Figura 1. Acondicionamiento y fermentación del tratamiento HF.

El proceso de fermentación incremento de manera significativa en HF las variables nutrimentales medidas obteniendo un incremento de 33.1% de NE, 31.5 de DE y ME, 28.8% de EE, 24.7% PC, 20.5% de EB, 20.0% de FS (Tabla 1), asimismo, se obtuvo el incremento de aminoácidos esenciales y no esenciales donde resaltan el triptófano (39.7%), histidina (27.3%), serina (21.5%), metionina-cisteína (19.4%), luecina (18.4%), alanina (16.8%) y fenilalanina (16.6%) (Figura 1).

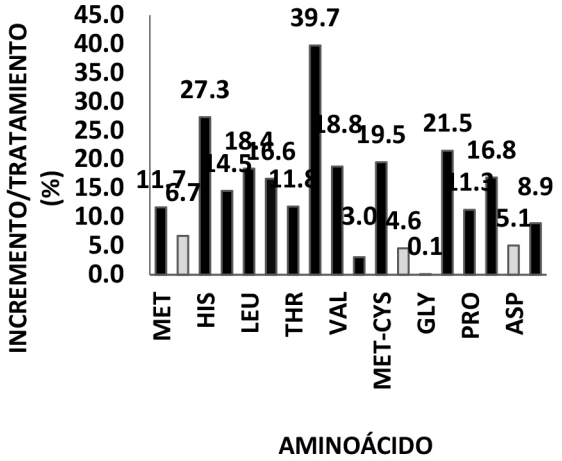

Figura 2. Incremento (%) de aminoácidos (AA) del Tratamiento HF respecto a la Prueba control

Se puede concluir la fermentación con *Aspergillus oryzae* sobre la harina de pescado (HF) puede incrementar significativamente los nutrientes de dicho ingrediente básico en la formulación de dietas acuícolas, lo anterior tiene un gran impacto en esta industria ya que incrementa la funcionabilidad de la harina de pescado desde un punto de vista económico y sustentable.

Tabla 1. Resultados de análisis bioquímicos proximales de los tratamientos nutricionales

ANÁLISIS	(BCO)	(HF)	Diferencia (%)
PC (%)	50.8±0.9[a]	63.3±1.9[b]	24.7
EE (%)	5.7±0.2[a]	7.3±0.3[b]	28.8
CZ (%)	21.2 ± 0.9[a]	18.6±1.4[b]	12.2
FS (g/Kg)	26.3±0.3[a]	21.0±1.0[b]	20.0
EB (MJ/kg)	15.3±0.2[a]	18.4±0.3[b]	20.5
DE (MJ/kg)	11.6±0.4[a]	15.3±0.4[b]	31.5
ME (MJ/kg)	10.5±0.3[a]	13.9±0.4[b]	31.5
NE (MJ/kg)	6.5±0.3[a]	8.7±0.2[b]	33.1

USO DE CONCENTRTADOS PROTEICOS DERIVADOS DE GRANOS DE DESCARTE PARA LA INCLUSIÓN DIETÉTICA EN CAMARÓN BLANCO DEL PACÍFICO *P. vannamei*

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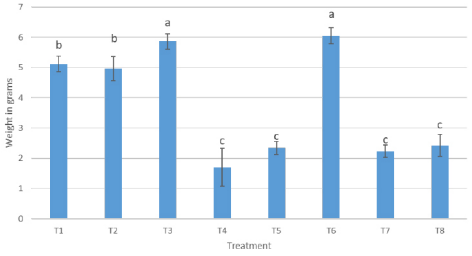
INTRODUCCIÓN. La búsqueda de remplazos para la harina de pescado en la la nutrición acuícola, es un tema de interés sustentable y económico, el cual, implican el uso y aprovechamiento de toda materia prima, que puedan cumplir con los requerimientos nutricionales para dietas animales y que, no representen una competencia directa para la alimentación humana (Corredor & Pérez, 2018). Los subproductos generados por el sector agrícola, en especial los granos de descarte. Se ha comprobado su valor nutricional en otros rubros, como la nutrición acuícola. Es por ello, que la presente investigación busca evaluar diferentes mezclas de concentrados proteicos derivados de tres granos de descarte como fuente de proteína vegetal.

METODOLOGÍA. Se obtuvieron concentrados proteicos de tres granos de descarte (C1, C2 y C3), los cuáles se obtuvo la caracterización proteica (análisis bromatológicos, perfiles de aminoácidos y antinutricionales). Evaluando en variables productivas de camarón blanco la adición en un 15% de distintas mezclas de concentrados proteicos. Además, se obtuvo la capacidad de recepción de granos descarte de una empresa receptora de granos de la región, para estimar la capacidad de producción de concentrados de proteínas de tres granos, según los rendimientos de concentrados obtenidos en una planta alimentaria industrial.

RESULTADOS Y CONCLUSIÓN
La inclusión de C1 en un 15% no compromete el crecimiento de *P. vannamei*, ya que no hay diferencias significativas con el tratamiento de control. Las inclusiones de concentrados de C2 por sí solo, y en combinación con C1 que reemplaza el 15% de la harina de pescado en dietas de camarón blanco, generaron un mayor peso final en organismos en comparación con el tratamiento de control.

La empresa receptora de granos tiene la capacidad de producción de 56.34 ton de C1, 123.14 ton C2 y 711.19 ton C3 por año.

Figura 1: Crecimiento de camarón blanco *P. vannamei* en bioensayo de crecimiento de 60 días



Dónde: T1: control, T2: C1, T3: C2, T4: C3, T5: C1, C2 y C3, T6: C1 y C2, T7: C2 y C3, T8: C1 y C3.

ESTADO GLOBAL DE LA CADENA PRODUCTIVA DE *Macrobrachium rosenbergii*: UNA REVISIÓN

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Macrobrachium rosenbergii es una especie nativa de países de la región Indo-Pacífico y de acuerdo con la FAO, su introducción como cultivo acuícola se identifica en al menos 40 países. La producción mundial en 2018 fue de 234.4 mil toneladas, representando 2.5 % de los productos totales de crustáceos. Es importante garantizar que la acuicultura sea una actividad responsable en los procesos de su cadena productiva, la cual es un sistema constituido por actores interrelacionados y una sucesión de procesos de producción, transformación y comercialización de un producto. En particular el cultivo de *M. rosenbergii* tiene un bajo impacto ambiental y ofrece oportunidades socioeconómicas a los países en desarrollo. Por ello, el objetivo de este trabajo fue revisar el estado actual de la cadena productiva de *Macrobrachium rosenbergii* a nivel internacional. Se realizó una búsqueda de literatura enfocada en la cadena productiva de *M. rosenbergii*. Se consultaron las siguientes bases de datos: Web of Science, EBSCO, Scopus, Scielo, Redalyc, Dimensions y Google Scholar con las palabras clave "Macrobrachium rosenbergii", "production chain" y "Freshwater prawn" con una selección de años del 2001 al 2022. Los principales eslabones de la cadena productiva de *M. rosenbergii*, identificados son: producción de postlarva (PL), engorda, transformación y comercialización. En la producción manejan la compra o la generación de esta, si bien en países con experiencia en el cultivo se desarrolla llegando a generar PL monosexo, es importante divulgar tecnologías de producción adecuadas para incorporar otras regiones emergentes a la producción mundial. En la engorda, los tipos de cultivo son principalmente intensivos, semi intensivos y extensivos; el cultivo monosexo y la selección por tallas se utilizan para mejorar la homogeneidad. Se emplean tecnologías para mantener la calidad del agua para el cultivo como sistemas de recirculación de agua, y acuicultura simbiótica. Se maneja monocultivo y policultivo especialmente con *Oreochromis niloticus*. En la transformación, se encuentra congelado, pelado, cola congelada, sin cabeza y entero que es apreciado por la vista en el mercado Gourmet, siendo un carácter distintivo. La comercialización se realiza en el mercado local, además de su exportación; siendo los principales exportadores China, Tailandia, India y Bangladesh este último importa a Estados Unidos, Reino Unido, Bélgica, Alemania y Japón. En América se produce en regiones tropicales y subtropicales en países como Brasil, Costa Rica, Perú y México. Su consumo puede ser directamente en restaurantes, también se implementa en festivales gastronómicos como estrategia de mercadotecnia. Se concluye que, para mejorar la cadena productiva de *M. rosenbergii*, se debe garantizar el suministro de PL, desarrollar mejores prácticas de monocultivo y la diversificación del cultivo con otras especies, así como la organización de la cadena productiva y los actores directos e indirectos que la integran.

PHYSIOLOGICAL AND IMMULOGICAL RESPONSE OF SEA CUCUMBERS: THE EFFECTS OF TEMPERATURE AND pH

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Sea cucumbers have adopted strategies that lead to the development of hypometabolism and involuntary non-pathological response. The first occurs by reduction of aerobic scope, which in turn determines physiological responses that generate dormancy and aestivation. Involuntary non-pathological responses, such as skin ulceration and evisceration, occurs as extreme control mechanisms to seek to reset organismal physiological condition at the expense of anatomical modification. In these organisms, pH and temperature represent two of the most important key factors that produces these responses. Evisceration and autotomy, reinforces the hypothesis that the mechanism involved in temperature perception remains similar throughout the life cycle. The detachment of the sensory fibres of collagen from the body walls suggest that coelomocytes play a greater role in perception. A plausible mechanism that explains how coelomocytes might respond to temperature could involve the adjustment of their affinity to Ca^{+2}, which activates transient receptor proteins, an important kind of membrane protein that constitutes a primary mechanism for detecting heat. With regard to pH, this affect antioxidant activity, especially when it is accompanied by changes in temperature. Temperature and pH produce molecular and physiological response that generate stress and might influence survival. As seen during aestivation, these parameters increased the presence of reactive oxygen species (ROS) and produce and imbalance in the proportion of antioxidants, which causes oxidative stress. The response might be regulated through adjustment of electron transport in mitochondria, with production of superoxide anions ($O_2^{-\bullet}$) as minor by-products. As high concentration of ROS modifies protein structure, antioxidant enzyme activity regulates their concentration in mitochondria and cytosol. Enzymes such as superoxide (SOD), glutathione peroxidase (GPx), catalases (CAT), and thioredoxins-peroxiredoxin (Trx-Prx) participate in the elimination of $O_2^{-\bullet}$ by converting it into hydrogen peroxide and water. Some experiment performed with sea cucumbers showed that constant increase of temperature increased the initial SOD activity of the body wall tissues from 45.2 to 128.2 U mg^{-1} of protein, while the increase of the exposure time to 25°C for 72h and 168h did not produce significant changes in antioxidant activity. Furthermore, in these organisms, the reduction of pH from 8.1 to 7.7 increase GPx activity of the coelomic fluid. Although the effect of temperature and pH are important in oxidative stress, few studies have addressed their interaction. From the publications, the evidence shows that at least in the short term, pH rather than temperature appears to be more important when antioxidant activity is considered. Here we present a hypothesis that explained the interaction of these factors and resume the main results of the articles that have been addressed for the topic.

SOCIAL CHALLENGES AND RESISTANCE TO CHANGE OF COMMUNITY MEMBERS IN THE NORTHERN ZONE OF CHIAPAS AND HOW THE INDUSTRY (AQUACULTURE) HAS DEVELOPED THE LOCAL ECONOMY

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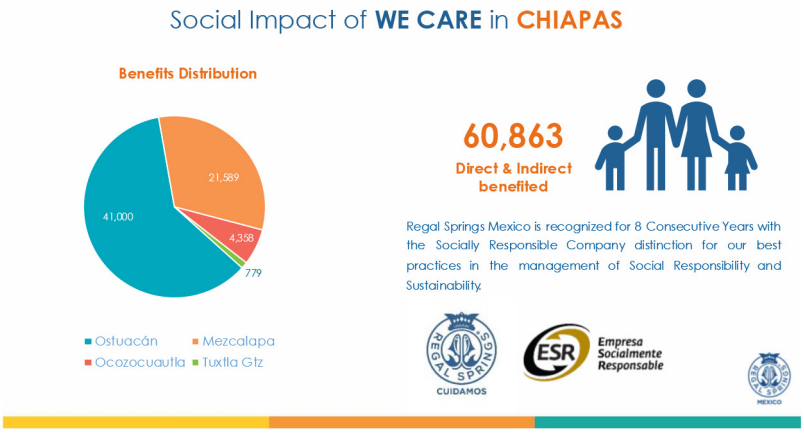
Chiapas is the poorest state in the country, it is characterized by having an infinity of social problems in all its regions, but it is also rich in cultural aspects, nature, climates, water and more; this being one of the reasons why the Regal Springs group decided to start operations 12 years ago, having the potential to be the main producer of tilapia at the national level, and even in LATAM.

The initial strategy was to identify the main interest groups around the municipalities where we operate (Ostuacán and Mezcalapa), identify the primary needs, and later create dialogue committees with the communities, fishing cooperatives and key stakeholders. At Regal Springs, we also work with all stakeholders in this way so that our operations and the tilapia we produce meet regulatory, sustainability, customer requirements and consumer expectations.

In 2018 Regal Springs WE CARE program was born, it aims to produce Naturally Better Tilapia by putting consumer needs first and without compromising sustainability. It is a commitment that is good for the consumer and the communities and economies in the countries in which we operate, while respecting the environment and natural resources. Our Regal Springs WE CARE Integrated Sustainability Program covers three sustainable development pillars: Environment, Social and Economic.

Annually our programs benefit more than 60,000 of Mexicans, we have got the Socially Responsible Company Distinctive, which is granted by the Mexican Center for Philanthropy (CEMEFI), the most important in social issues in LATAM.

We still have some challenges on the environmental, social and cultural side, we mitigate with our 5 CUIDAMOS pillars: education, health, reforestation, lakes & water and infrastructure.



MORPHOLOGIC AND GENETIC CHARACTERIZATION OF THE POLYDORID SPECIES THAN AFFECTS *Magallana gigas* CULTURED IN THE SAN QUINTIN BAY

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One of recurrent health problems that effect the culture of the Japanese oyster, *Magallana gigas*, which is one of the bivalve mollusks with the highest aquaculture production at a global scale. Is the infestation by burrowing worms of the family Spionidae that have the ability to settle on calcareous shells piercing them and forming tunnels where they take refuge. The hosts become affected when the infestation is high and the worms reach the inner face of the shell. This ultimately triggers a reaction from the host who expedites energy into depositing additional nacreous layers to isolate the polychaete to stop it from reaching the soft tissues forming blisters. In such cases that the worm reaches the soft tissues of the hosts, it becomes to be more susceptible to bacterial infections. Additionally, severe infestations result in a brittle and unpleasant aspect of the shell for marketing. In Mexico the production of M. gigas is concentrated in San Quintin Bay and since 1997, the presences of these burrowing worms and the damage that produces in the oyster has been documented: however, his identity has not been confirmed.

In this study we carry out a broad sampling of oysters from the Bay and found three different phenotypes of polydorids, which differ in the pigmentation of the palps. The Whole Genome Sequencing analysis of the most prevalent phenotypes shows that the 18S rRNA gene has a similarity of 99.5% whit *Boccardiella hamata* while mitochondrial genes show similarities of ranging from 83-89% (Table 1). Conventional taxonomy analysis and ultrastructural studies for describing discrete morphological features are underway in addition to further next generation sequencing and sanger sequencing studies. Similar analysis will be conducted to the other two phenotypes. This research will help clarify the taxonomic affiliation of these burrowing worms and their role in the effect on the shell. Additionally, will aid in the development of diagnostic tools to differentiate among possible different species and to provide base line element that contribute to mitigate their negative effect on Mexican oyster culture.

Table 1. Similarity of three mitochondrial genes of the novel Polidorid with *B. hamata*

Gen	Size (pb)	% Similarity
COX1	1547	83.27
16S	1218	87.78
12S	815	89.07

THE EFFECT OF DISTAL-END TRIMMING ON *Saccharina latissima* MORPHOLOGY, COMPOSITION, AND PRODUCTIVITY

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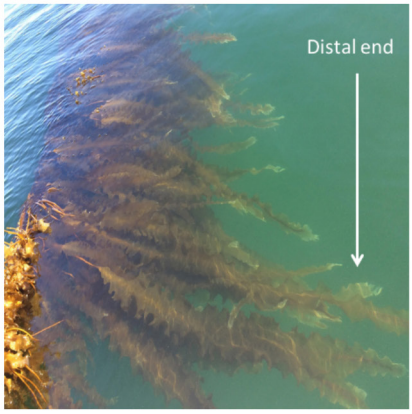
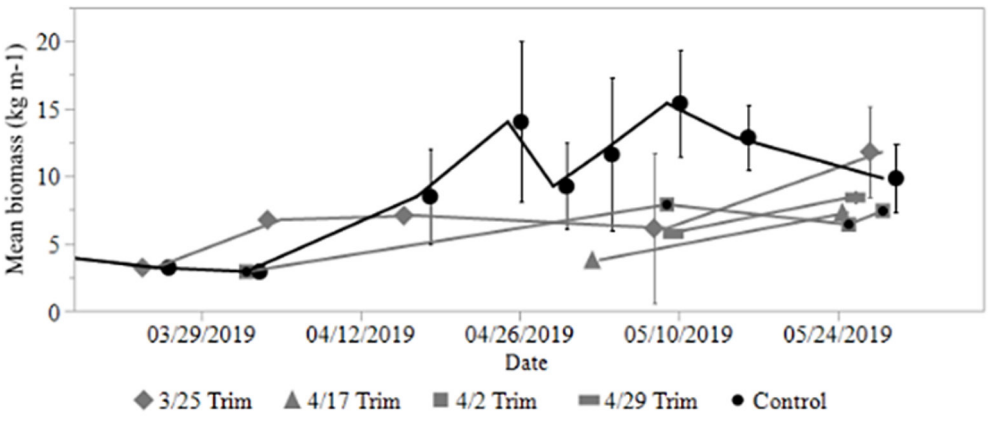
As kelp cultivation increases around the world, so does the need for farm management strategies that produce specific crop characteristics, optimize yield, widen harvesting windows, and prevent biomass loss. Distal-end trimming of macroalgae has been recommended as a farm management method addressing these needs. In this study, we trimmed cultivated *Saccharina latissima* sporophytes grown in the Western Gulf of Maine to 60 cm above the stipe-blade interface. We characterized the effect of trimming on the morphology, tissue nutrient content, stable isotope ratio, and nitrate reductase activity of the kelp. We also evaluated the economic trade-offs of trimming using a simple production model.

The results suggest that trimming the blade to 60 cm may have minimal biological consequences. Additionally, the trimming appears to benefit "short" kelp blades in proximity to the trimmed blades. Daily yield (% increase in weight day^{-1}) after trimming was initially lower than the control, but late-season daily yields and crop-retention following storms were markedly improved. Ultimately, we conclude that growers could use trimming to acquire kelp biomass earlier in the season, retain late-season biomass, and potentially increase the total revenue gained from kelp farming if price premiums can be exacted for this biomass.





GENETIC CHARACTERIZATION AND EPIZOOTIOLOGY OF RICKETTSIALES-LIKE ORGANISM ASSOCIATED WITH TISSUE INCLUSIONS OF THE OYSTER *Crassostrea corteziensis* IN THE STATE OF NAYARIT

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In numerous histopathological studies carried out on several species of bivalve mollusks of commercial aquaculture importance around the world, the presence of characteristic tissue inclusions has been related to Rickettsiales-like organism (RLOs). In some records, these RLOs were associated with mass mortalities and tissue damage but not in others. Thus, the type of symbiotic association between the oyster and the bacterium have not been clearly defined. Typically, the studies on RLOs are limited to ultrastructural and histological descriptions which are inadequate for a robust taxonomic classification. These RLOs have been observed on the west coast of Mexico in routine health surveys of the pleasure oyster *Crassostrea corteziensis* (Figure 1). In addition to RLOs being the causative agents of the inclusion bodies observed in bivalves, the genus *Endozoicomonas* has been recently found to cause very similar inclusion bodies in a wide range of bivalve hosts. The genus *Endozoicomonas* has been reported on the west coast of Mexico suggesting the presence of these bacteria in *C. corteziensis*.

Employing histological methodologies in combination with transmission electron microscopy (TEM), *in situ* hybridization and Next generation sequencing (NGS) from DNA extracted from formalin-fixed paraffin-embedded (FFPE) tissues we attempt to identify and characterize the bacteria observed and causing the inclusion bodies in *C. corteziensis*. The NGS was performed from 10 years old FFPE tissues. The sequences were compared using BLASTn and mapped to the partial sequence of 16S rRNA for *Endozoicomonas* sp. In addition, microbiome analysis of the bacterial community associate with *C. corteziensis* (table 1) showing the tissue inclusions of the RLOs are underway. Histologically, the intracytoplasmic vacuoles (Ivs) (figure 1) presented sizes of 15.70-15.24 μm in length and 15.42-14.95 μm in width. The prevalence of RLOs in the study period was 6% while the infection intensity of the RLOs was grade one in 89% of the infected oysters.

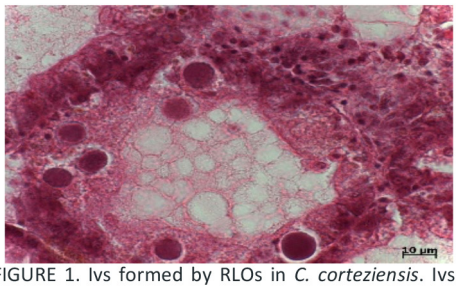

FIGURE 1. Ivs formed by RLOs in *C. corteziensis*. Ivs were observed in secondary ducts of the digestive gland of *C. corteziensis.*

TABLE 1. Microbiome analysis of the bacterial community

specie	Per. Ident	Accession
Aeromonas sp.	97.75%	KC665708.
P. lundensis	98.01%	CP075180.1
P. otitidis	99.30%	AP022642.1
P. alcaliphila	99.54%	CP016162.1
P. pseudoalcaligenes	98.63%	HG916826.1
Pseudomonas sp.	99.54%	CP031606.1
P. aeruginosa	97.06%	CP052759.1

USO DE SORGO BICOLOR EXTRUIDO E INTEGRAL COMO ALTERNATIVA PARA ALIMENTO PARA TILAPIA DEL NILO *Oreochromis niloticus*, MEDIANTE VARIABLES PRODUCTIVAS Y PLAN DE NEGOCIOS

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Palabras clave: Evaluación técnico-económica, Sorghum bicolor, Extruido, Oreochromis nilóticos, variables productivas y plan de negocios

Introducción. La acuicultura es una de las actividades primarias de gran importancia para la producción de alimentos con proteína de alta calidad. En las últimas décadas, ha ido creciendo más rápido que otros sectores. En el año 2018, la producción mundial de acuicultura incluidas plantas acuáticas aportaron 114.5 millones de toneladas de la producción de alimento en el mundo, estimadas en un valor de 263, 600 millones de dólares.

El éxito del cultivo de tilapia del Nilo *O. niloticus* se debe principalmente a su buen desempeño en crecimiento, mayor rendimiento en alimentación, adaptación a diversas fuentes de proteína, entre otras (CONAPESCA, 2011; Fimbres, 2019). Sin embargo, la producción no alcanza a cubrir la demanda del mercado nacional. De lo anterior, es importante mencionar que, de todos los organismos provenientes de la acuicultura tan solo el 47.25% están representados por los peces cultivados a nivel mundial, por lo que estos organismos cumplen un papel importante de la seguridad alimentaria en todo el mundo.

Por otra parte, el elevado costo de producción los sistemas de producción piscícola derivado del costo de alimentación pueden ser superior al 50% debido a uso de harina de pescado, al igual que carne, sangre y la pasta de soya (Perea-Román et al., 2018). Las materias primas antes mencionadas tienen un alto valor nutritivo, buena digestibilidad y palatabilidad, sin embrago, son caros y de baja disponibilidad (Civera et al., 2010). Por lo anterior, es necesario buscar fuentes alternativas proteicas regionales de bajo costo, pero alto contenido nutricional, con la finalidad de disminuir los costos de producción (Perea-Román et al., 2018).

Dentro de la especie de S. bicolor, la variedad Gavatero-203 de grano rojo tiene mejor calidad bromatológica que los híbridos comerciales en el forraje, además de 77.3% y 66.4 % de digestibilidad.

Tabla 1. Parámetros productivos de *O. niloticus* alimentada de HSINE Y HSIE (35 %) durante los primeros 90 días de cultivo. (Datos expresados con media ± desviación estándar).

Tratamientos	C2 (H. Soya & Trigo)	T3 (HSINE 35%)	T6 (HSIE 35%)
Peso Inicial (g)	34.00 ± 0.63 [a]	35.30 ± 0.83 [a]	34.00 ± 0.63 [a]
Peso final (g)	230.56 ± 4.43 [b]	287.39 ± 17.31 [a]	258.95 ± 15.50 [ab]
GP (g)	195.68 ± 5.01 [b]	252.22 ± 17.52 [a]	224.94 ± 15.36 [ab]
TCA (g/día)	1.49 ± 1.22 [a]	0.95 ± 1.48 [a]	0.95 ± 1.45 [a]
TCE (%/día)	1.59 ± 0.18 [a]	1.50 ± 0.21 [a]	1.53 ± 0.18 [a]
FCA	1.85 ± 0.04 [b]	1.46 ± 0.03 [a]	1.67 ± 0.15 [ab]
S (%)	76 ± 4.00 [b]	96 ± 6.9 [a]	72.00 ± 8.32 [ab]

[a, b] Medidas con letras diferentes entre tratamientos indican diferencias estadísticas significativas ($p < 0.05$). C2: dieta control con harina de pescado, soya y trigo, T3: inclusión de HSINE 35 y T6: inclusión de HSIE 35%, GP: ganancia de peso, TCA: tasa de crecimiento absoluta, TCE: tasa de crecimiento especifica, FCA: factor de conversión alimenticia, S: sobrevivencia.

(Continued on next page)

El realizar un estudio del efecto de incluir harina de sorgo integral extruida y no extruida por otras fuentes alternativas nutricionales para obtener alimento balanceado que ayude a reducir costos de producción.

Objetivo general. Determinar la factibilidad técnico-económica de la inclusión de harina de sorgo bicolor (S. bicolor) integral, extruida y no extruida en la dieta de la tilapia del Nilo *O. niloticus*, mediante indicadores productivos y un plan de negocios.

Estrategia metodológica. El experimento se realizó en el invernadero del Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional (CIIDIR-SINALOA), el cual consistió en dos fases: Fase I: determinar el porcentaje de inclusión de harina de sorgo extruida e integral (15%, 25% y 35%); se elaboraron 8 dietas experimentales con distintos porcentajes de inclusión de harina de sorgo extruida e integral, así como dos dietas control, la duración total de este bioensayo es de 60 días, haciendo biometrías cada 15 días, y Fase II: realizar una prueba comercial para evaluar costos de elaboración del alimento; se elegirán los tratamientos con mejor rendimiento en cuanto a peso y crecimiento (Muñoz, 2016). Los datos experimentales se obtuvieron en los bioensayos, se analizaron mediante un análisis de varianza (ANOVA) con un nivel de 95% de confianza, y con una P=0.05, se procesarán en el programa Statistica 7.0. Para el desarrollo del plan de negocios, se atenderá en función a la propuesta estructurada para empresas de nueva creación Seigel, Ford y Bornstein, 1993.

Resultados

En la Tabla 1 se muestran los parámetros productivos obtenidos durante los primeros 90 días de cultivo del bioensayo comercial. En cuanto a los pesos finales promedios, se presentaron diferencias estadísticas entre tratamientos ($p< 0.05$), en el T3 se encontraron los mayores pesos finales promedios con un valor de (287.39 ± 17.31 g), sin embargo, este no fue diferente del T6 (224.94 ± 15.36 g).

Tabla 1. Parámetros productivos de *O. niloticus* alimentada de HSINE Y HSIE (35 %) durante los primeros 90 días de cultivo. (Datos expresados con media ± desviación estándar).

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EFFECT OF *Litopenaeus vannamei* CONTACT FEMALE SEX PHEROMONES ON MALE COURTSHIP BEHAVIOR

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Litopenaeus vannaemei is one of the most important aquaculture species at international level, so it is necessary to continue with the development of biotechnologies that allow optimization of larval production in an efficient and sustainable way. Within the lines of research that can contribute to this objective, sexual pheromones are one of those with the greatest potential; however, although the existence of two types of female sex pheromones in this organism (distance and contact) is hypothesized, to date their existence has not been proven. In this work, the detection of the contact female pheromone involved in the reproductive process of this important specie was carried out for the first time.

For this purpose, the behavior presented by sexually mature males against "artificial females" previously treated with liposoluble cuticular extracts of mature and immature females was observed and classified. The treatments tested were: (1) ventral exoskeleton of immature female (EVHI), (2) dorsolateral exoskeleton of immature female (EDHI), (3) ventral exoskeleton of mature female (EVHM) and (4) dorsolateral exoskeleton of mature female (EDHM). The experiment was divided into two bioassays: 1) extracts from immature females and 2) extracts from mature females; each with two treatments and one control (hexane), evaluated in 15 replicates per treatment. The behavior was classified according to the following grades: 0 = no response; 1 = contact; 2 = push and 3 = prolonged contact (≥ 10 s). The association between each behavior presented and between the different treatments was determined using Barnard's exact test; if any behavior was presented more than once in at least one replicate, a one-way ANOVA was performed to determine if there were differences in the number of times each behavior occurred. In addition, the total bioactivity presented was evaluated with the Kruskal-Wallis test.

Figures 1 and 2 show the results obtained in terms of total bioactivity, finding a significant association between the reproductive behavior of *L. vannamei* and the different extracts tested. The results show sufficient evidence to determine the existence of a contact female sex pheromone with sexual recognition function

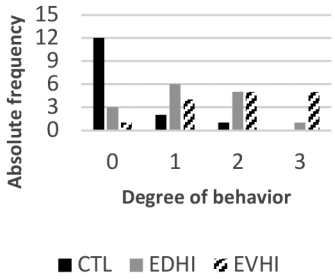

Figure 1. Degree of bioactivity presented by *Litopenaeus vannamei* males when exposed to cuticular extracts from immature females (n= 15 replicates/treatment). Grade 0: no response; grade 1: contact; grade 2: push; and grade 3: prolonged contact (≥ 10 s). Abbreviations: CTL = control; EDHI = dorsolateral exoskeleton of immature female; EVHI = ventral exoskeleton of immature female.

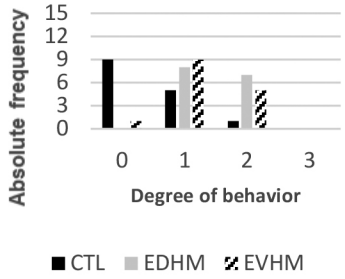

Figure 2. Degree of bioactivity presented by *Litopenaeus vannamei* males when exposed to cuticular extracts from mature females (n= 15 replicates/treatment). Grade 0: no response; grade 1: contact; grade 2: push; and grade 3: prolonged contact (≥ 10 s). Abbreviations: CTL = control; EDHM = dorsolateral exoskeleton of mature female; EVHM = ventral exoskeleton of mature female.

PRACTICAL CHALLENGES WHEN INJECTING MILLIONS OF FISH WITH VACCINES

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Injection vaccines are quickly becoming the method of choice for farmed fish species kept in high densities, mainly due to the long-term protection they provide for the fish. In order to ensure proper immunization, animal welfare and sustainability, it is important that the vaccinations are done accurately and swiftly.

Almost all farmed Atlantic salmon is currently injected with up to three different vaccines in the abdominal cavity, and sometimes also intramuscularly before outset to sea. The majority of fish are vaccinated by hand, but vaccination by machine is becoming increasingly more common in salmon, trout and sea bass farming. The first automatic vaccinations of tilapia were performed earlier this year. Current manual injectors can inject one or two vaccines intraperitoneally or muscularly, but it is not possible to inject in two different locations simultaneously. Machines offer a greater vaccination rate, accuracy, flexibility, and better biosecurity due to a minimum of people needing to visit the fish farming location. Innovations within machine vision and injection systems allows the machines to accurately inject the fish with several vaccines simultaneously, and the site and depth of injection is adapted to the individual fish. Injured or deformed fish can be excluded automatically, and populations can be graded by size during vaccination.

We will present the use of injection tools to enable vaccinations of large numbers of fish, while increasing production efficiency, improving sustainability and maintaining a high level of fish health.

AGAVIN REDUCES HIGH-DENSITY STRESS ON GROWTH PERFORMANCE, CORTISOL AND GLUCOSE LEVELS ON NILE TILAPIA (*Oreochromis niloticus*)

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Nile tilapia (*O. niloticus*) is one of the most popular aquaculture species in the world, however, factors like high-density of culture cause poor growth performance and blood parameter alterations, such as increment of cortisol and glucose levels, and oxidative stress[1]. The use of agavin, as a functional additive, can exert health benefits due to its prebiotic properties. Agavin is a highly branched fructan that can be obtained from the waste of the tequila industry [2,3]. Studies about agavin in aquatic organisms are scarce, however, the addition of dietary fructans might improve the growth performance, maintain cortisol and glucose levels, and promote fish wellness.[3,4] Therefore, this study aimed to evaluate the effects of high-density stress on growth performance and cortisol and glucose levels of Nile tilapia fed dietary agavin.

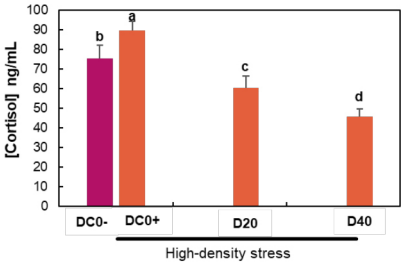
Methods: Fish were fed a base diet (D0), and two diets with agavin inclusion, 20 (D20), and 40 g/Kg (D40). Masculinized Nile tilapia (1.04±0.01 g) were stocked into 350 L fiberglass tanks, assigned to four treatments: a control, without high-density stress challenge (DC0-) and three groups challenged with high-density stress (DC0+, D20, and D40). Each treatment had five replicates, an initial density of 6 kg/L, and feeding to 10% of body weight. At 90 days, DC0+, D20, and D40 were subjected to high-density stress (600 kg/L) for additional 20 days, by reducing water level[3], after which growth performance and plasma cortisol and serum glucose levels were evaluated.

Results and Discussion: The D20 diet reduced the negative effect of high-density stress on final weight (FW) ($p<0.05$) (Table 1). Regarding cortisol and glucose levels, both agavin inclusions (20 and 40 g/kg) reduced cortisol levels, while the glucose level was only maintained by the D20 diet during the challenge (Figure 1). Previous studies have shown that the addition of fructans can promote growth performance and reduce oxidative stress by acting as anti-stress additives, preventing the increase in cortisol and glucose levels[2,3,4]. This is because agavin can regulate the cortisol hormone by modulating the hypothalamic-pituitary-adrenal axis, and, stimulating the vagus nerve, in addition to modulating pathways related to gluconeogenesis, which reduces glucose and cortisol levels in stress conditions.[3, 6, 7] However, factors such as agavin dosage, chemical structure, stress conditions, as well as life stage can affect its prebiotic potential.[3,4]

Table 1. Growth performance parameters in high-density of Nile tilapia.

	DC0-	DC0+	D20	D40
FW (g)	235.6 ± 22. 8 [a]	159.6 ±27.5 [b]	219.8 ±24.5 [a]	193.3±16.6[b]
GW (g)	123.0 ±13.7	89.7 ±14.2a	110.9 ±14.4a	99.3 ±24.4
FCR	0.94±0.1	1.0±0.06	1.0 ±0.09	1.05 ±0. 1
K	1.9±0.1	1.7±0.1	1.7±0.05	1.7±0.1
S (%)	97±0[b]	97±0 [b]	94±0[b]	100±0[a]

Final weight (FW), weight gain (WG), feed conversion ratio (FCR), Condition factor (K), survival (S). Different letters in the same file indicate significant differences (Tukey, p≤0.05). DC0-: without stress control, DC0+: stress subjected to high-density; D20: 20 g/kg agavin; D40: 40 g/kg agavin.



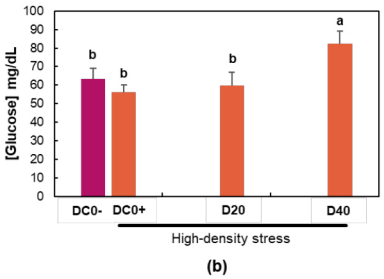


Figure 1. The effect of agavin on (a) cortisol and (b) glucose levels of Nile tilapia after 20 days in high-density stress conditions (600 g/Kg). DC0-, control without high-density stress challenged; DC0+, control in high-density stress challenged; D20, an experimental diet with 20g/Kg agavin in high-density stress challenged; D40, an experimental diet with 40g/Kg agavin in high-density stress challenged. Data obtained from five biological repeats are shown as mean ± SD values. Different letters correspond to significant differences ($p<0.05$) to according Tukey's-HSD test (n=5).

(Continued on next page)

Conclusion: The agavin inclusion (20 and 40 g/kg) reduces plasma cortisol levels during the high-density stress challenge (600 kg/L), while the inclusion with 20 g/kg of agavin also maintains serum glucose levels and reduces the negative effects in FW under stress conditions.

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DESARROLLO DE ALIMENTOS FUNCIONALES PARA ACUACULTURA

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Los sistemas de cultivo intensivo de peces y camarón generan estrés oxidativo relacionado al daño oxidativo y mortalidad de los organismos. Los residuos agroindustriales y pesqueros son fuentes potenciales de antioxidantes como, polifenoles y péptidos, los cuales potencian la respuesta antioxidante y previenen del daño oxidativo. Por lo tanto, el objetivo del Laboratorio de Nutrición de CIAD es desarrollar alimentos funcionales que modulen la respuesta antioxidante de organismos acuícolas. Se evaluó el efecto de extractos polifenólicos de bagazo de café contra el estrés oxidativo inducido en células de cerebro de robalo *C. viridis*, así como el efecto de las inclusiones dietéticas de polifenoles de harina de hoja de mazorca de maíz y extracto de cáscara de mango, y péptidos bioactivos de residuos de pescado sobre la respuesta antioxidante enzimática (superóxido dismutasa, catalasa, glutatión peroxidasa y prevención de la peroxidación lipídica) en tilapia (*Oreochromis niloticus*), pez cebra (*Danio rerio*) y camarón blanco (*Penaeus vanammei*), respectivamente. Los extractos polifenólicos de bagazo de café (230 y 460 µg/mL) incrementaron la actividad de superóxido dismutasa y redujeron la peroxidación lipídica en células de cerebro de robalo con estrés oxidativo inducido. Los polifenoles de la cáscara de mango incrementaron la actividad de catalasa hepática y redujeron la peroxidación lipídica en el músculo del pez cebra. La hoja de mazorca de maíz (25g/kg de alimento) como fuente de polifenoles incrementó la actividad de catalasa hepática en tilapias expuestas a estrés por hipoxia (1.5 ± 0.2 mg/L oxígeno disuelto) durante 5 h. Los péptidos de bajo peso molecular de músculo oscuro de atún incrementaron la actividad de superóxido dismutasa en camarones blancos sometidos a estrés térmico (33°C) por 6 h. Los residuos vegetales y pesqueros tienen potencial para usarse como aditivos antioxidantes en el desarrollo de alimentos funcionales para especies acuícolas de interés comercial.

ECLOSIONADOR AUTOMÁTICO DE ARTEMIA SALINA PARA ALIMENTACIÓN DE LARVA DE LANGOSTINO MALAYO (*Macrobrachium rosenbergii*)

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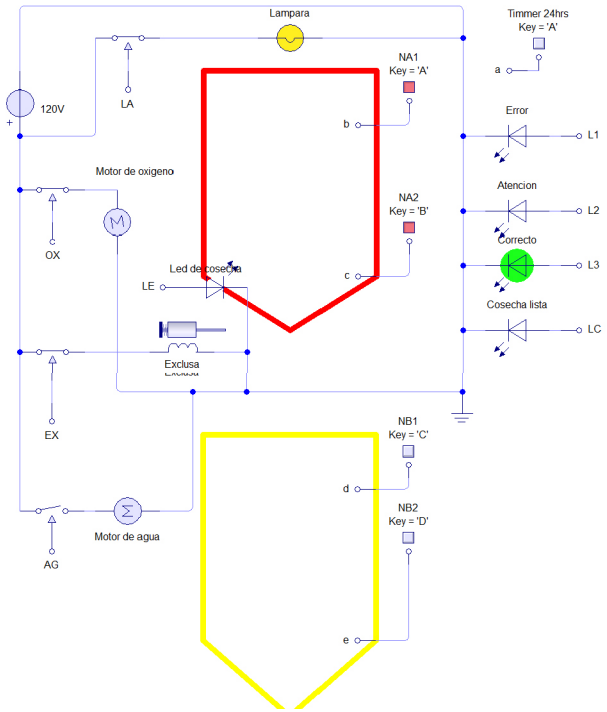
El presente proyecto se llevó a cabo en el laboratorio de acuacultura del Colegio de Postgraduados Campus Veracruz, ubicado en el municipio de Manlio Fabio Altamirano Veracruz, con la finalidad de diseñar un prototipo que permita incubar el alimento vivo (Artemia salina), para alimentar a las larvas de langostino malayo (Macrobrachium rosenbergii). Se utilizó un microcontrolador ATMega328p, dos contenedores de 4 litros con dos sensores de agua integrados, una bomba de aire, una bomba de agua, un led ultra brillante, una lámpara, un temporizador, cable calibre 22 y un software de programación Arduino IDE 1.6.10. Los dispositivos fueron conectados en sus respectivos pines para su alimentación, de igual manera fueron conectados al microcontrolador para su control.

Al iniciar el proceso, se verificó que el primer contenedor tuviera suficiente agua; posteriormente se activó la aireación y se encendió una lámpara para mantener a los quistes de Artemia en incubación durante 23 h. Al finalizar se abrió el paso de agua con el servomotor para dejar libre a los nauplios hacia un colador donde quedan atrapados en una malla y quedan listos para suministrarlos a las larvas de langostino.

Se realizó una evaluación del prototipo para realizar un control de eficiencia y control de operación, lo que proporcionará en un futuro un mejor control sobre la alimentación durante las fases larvales permitiendo un aumento en la tasa de supervivencia, al traspaso de postlarvas y un incremento en la productividad del sistema de producción de langostino.

El avance tecnológico propuesto tiene la particularidad de adaptarse a distintos tamaños para la cosecha de Artemia, permite mantener evitar el desperdicio de agua y es amigable con el medio en donde se encuentra. Gracias a ello se logró quitar la carga de trabajo para el productor, el cual se utilizó para distintas actividades en el área de maternidad, debido a que son muchas las actividades a realizar para lograr tener una buena producción.

Figura 1 Diagrama electrónico de funcionamiento del eclosionador de Artemia salina



Vibrio areninigrae AS A PATHOGENIC BACTERIUM IN A CRUSTACEAN

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The occurrence of infectious diseases poses a significant threat to the aquaculture industry worldwide. Therefore, characterization of potentially harmful pathogens is one of the most important strategies to control disease outbreaks. In the present study, we investigated for the first time the pathogenicity of two *Vibrio* species, *Vibrio metschnikovii*, a foodborne pathogen that causes fatalities in humans, and *Vibrio areninigrae*, a bacteria isolated from black sand in Korea, using a crustacean model, the signal crayfish *Pacifastacus leniusculus*. Mortality challenges indicated that injection of *V. metschnikovii* (10^8 CFU/crayfish) has a mortality percentage of 22% in crayfish. In contrast, injection of *P. leniusculus* with 10^8 or 10^7 CFU of *V. areninigrae* resulted in 100% mortality within one and two days post-injection, respectively. *V. areninigrae* was successfully re-isolated from hepatopancreas of infected crayfish and caused 100% mortality when reinjected into new healthy crayfish. As a consequence of this infection, histopathological analysis revealed nodule formation in crayfish hepatopancreas, heart, and gills, as well as sloughed cells inside hepatopancreatic tubules and atrophy. Moreover, extracellular crude products (ECP's) were obtained from *V. areninigrae* in order to investigate putative virulence factors. In vivo challenges with ECP's caused >90% mortalities within the first 24 h. *In vitro* challenges with ECP's of hemocytes induced cytotoxicity of hemocytes within the first hour of exposure. These findings represent the first report that *V. areninigrae* is a highly pathogenic bacterium that can cause disease in crustaceans. On the contrary, *V. metschnikovii* could not represent a threat for freshwater crayfish.

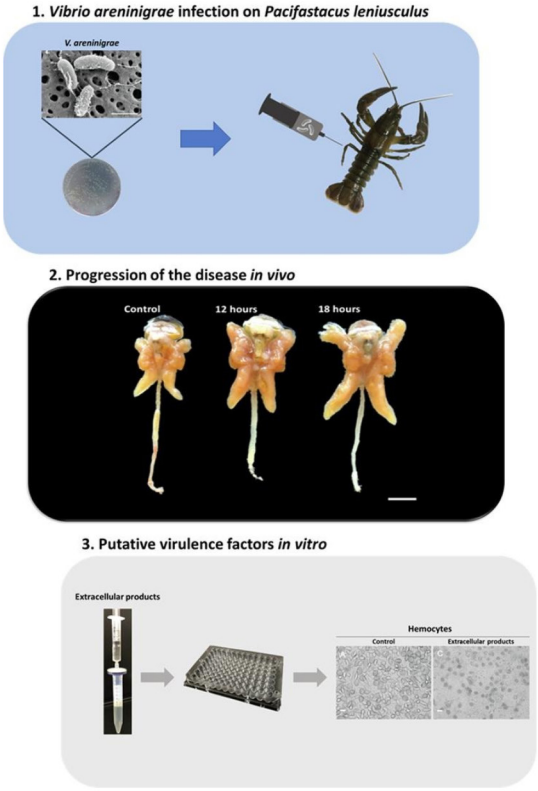

Figure 1. Mortality assay with *Vibrio areninigrae*. Fig 2. Photographs of the gastro-intestinal tracts (stomach, hepatopancreas and intestine) from *P. leniusculus* injected with 0.9% NaCl (Control), 4.6×10^8 CFU of *V. areninigrae* at 12 hpi and 18 hpi. Bar scale=1 cm. Fig 3. Citotoxicity assay of extracellular crude products in hemocytes culture.

IS AQUAPONICS SYSTEM A STRATEGY TO IMPROVE FISH HEALTH AND CONTROL DISEASE PRESENTATION?

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Aquaponics biotechnology integrates aquaculture and hydroponics. In this sense, it is accepted that these systems result in the improvement of water quality, higher nutrient availability, and high quality production of a second product, the plants. However, information regarding a beneficial or detrimental effect upon aquatic organisms at physiological level, and particularly in reducing disease presentation, is still scarce.

Up to date, the most common physiological parameters, or "health indicators", evaluated in organisms from aquaponics systems are those related to production performance aspects, including growth and feed conversion ratio[1,2,3]. Although valuable in terms of productivity, these parameters aren't indicators of the immunological features of the organisms, and their capacity to overcome disease presentation, making it necessary to include a more comprehensive approach. Likewise, recently, some efforts have been made to understand, from an integrative perspective, the role of disease presentation in aquaponics systems[4,5]. However, differences in the design of systems, the lack of direct comparison of aquaponics technology with traditionally closed-systems, species, water, sizes and stocking densities of fish, and composition of feed and feeding rates, still make difficult to fully identify a beneficial interaction between aquaponics and aquatic species to improve health and control disease presentation.

Elucidating this important issue in a complex system, such as aquaponics, requires the implementation of comprehensive strategies, including: i) *in vivo* infection challenges, ii) multi-omics approach, and iii) functional studies. This information will help for a better understanding of the relationships between aquatic organisms to aquaponics during adverse events like disease.

Table 1. Study cases of aquaponics effect in the physiological response of some commercial fish.

Species	Parameter evaluated	Physiological effect
Qihe crucian carp (*Carassius auratus*)[1]	Production performance (inclusion of lettuces)	A trend towards higher growth performance with *L. aquatica*, *L. sativa*, and *C. demersum*.
Common carp (*Cyprinus carpio* L.)[2,3]	Production performance (inclusion of synbiotics)	Positive effect of symbiotic on growth and feed utilization.
	Density (high vs. low)	Growth and feed conversion were negatively influenced by higher stocking density.
Siberian sturgeon (*Acipenser baerii*)[4]	Production performance (inclusion of insects in food)	Negative effect in growth and survival, promotes fasting and stress, and reduces energy reserves.
European sea bass (*Dicentrarchus labrax*)[5]	Salinity and disease presentation	Salinity causes behavior modification and limited growth during disease. Reduced fish growth was a consequence of the decrease in appetite stimulus.

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EFFECT OF FOOD RESTRICTION / REFEEDING ON THE SURVIVAL, GROWTH AND ENZYMATIC ACTIVITY IN HEPATOPANCREAS AND INTESTINE OF MALAYSIAN PRAWN POSTLARVAE CULTURED IN BIOFLOC

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In recent years, different strategies have been implemented to optimize the activity and make it intensive and sustainable, so biofloc technology (BFT) has been implemented in production, reducing the volume of daily water replacement and providing complementary food to organisms during culture. In addition to biofloc, strategies for efficient use of food in aquaculture have begun to be developed, such as intermittent feeding and compensatory growth, especially in detritus-consuming species, a technology that to date has not been evaluated in the Malayan prawn (*Macrobrachium rosenbergii*) in conjunction with biofloc culture. Therefore, the objective was to evaluate the effect of food restriction on the survival, growth and enzyme activity of Malayan prawn (*M. rosenbergii*) postlarvae during their culture in biofloc.

The experimental design was completely randomized with six treatments and three replicates. Three treatment were kept in clear water (W) and three in biofloc (B) with 3 days restriction / 3 days refeeding (3) and 3 days restriction / 6 days refeeding (6). Controls (BC) and (CW) received food daily. 15 prawns of 0.5 g were used and the study lasted 75 days. The parameters of water quality, productive performance, as well as proximal composition and digestive enzyme activity in hepatopancreas and intestine were evaluated. Data were analyzed with an ANOVA and Tukey test at $p<0.05$. The physicochemical variables in water were within the tolerance range for the PL´s. The survival at the end of the study was similar in the treatments and greater than 80%. The most efficient growth was obtained in BC, which was significantly higher than the rest of the treatments. Partial compensatory growth of treatments B3 and B6 was observed vs BC and total compensatory growth was obtained when compared B3 and B6 vs clear water control (WC), indicating that the biofloc allows maintaining the growth of the organisms even in the absence of commercial food. (Table 1). The nutritional composition of the prawns subjected to the treatment BC is significantly higher compared to the other treatments. Variations in enzymatic activity were observed in the hepatopancreas and in the intestine as a response to adapt to changes in food restriction and use of the biofloc.

Table 1. Mean values (± standard deviation) for productive performance variables of *Macrobrachium rosenbergii* reared in biofloc system during restriction and refeeding phases

	Treatment					
Variable	BC	B3	B6	WC	W3	W6
S (%)	91.11±3.85	86.86±6.67	84.44±3.85	88.89±3.85	80.00±6.67	84.44±3.85
FW (g)	4.71±0.65a	3.04±1.54b	3.13±1.07b	3.08±1.11b	2.05±0.94b	2.41±0.95b
WG (%)	1278.90±59a	697.83±46b	570.01±42b	495.36±35b	344.22±24b	388.33±18b
FCR	2.78±0.37cd	2.53±0.05abc	2.68±0.03bcd	2.84±0.37d	2.28±0.12a	2.49±0.25ab

Different letters mean significant differences at $p<0.05$.

EVALUATION OF *Vieja bifasciata* CICHLID FOR AQUACULTURE: LARVICULTURE AND MASCULINIZATION

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The twoband cichlid *Vieja bifasciata* is an indigenous fish species from the tropical areas of Central America. This specie is considered as candidate for aquaculture due the high demand for traditional cuisine reaching 5 US/kg in regional market, and ornamental production is considered promissory for international sales due to coloration. Twoband cichlid larvae production is feasible using experimental RAS using commercial diets, and actually fry is mainly destinated for restocking traditional fishing areas. In the wild, adults weight commonly show differences by sex like another cichlid species, but absence of data on age do not allows to define a differential grow hypothesis. In order to determine if *Vieja* monosexual population show differential growth, an experimental protocol for fry production and masculinization was developed.

Fourteen, one year old *Viaja bisfaciata* adults (180-460 g) reared in laboratory were stocked in 1.7 m^3 tanks for larvae production during reproductive season (April). Adults were fed twice a day using commercial diet and water quality maintained by weekly 5% water renewal. A triplicated masculinization trial was conducted using first feeding larvae. Larvae were fed by 28, 45 and 60 days using a diet containing 60 mg/kg of the steroid (17α-MT), and a control treatment. At the end of the treatment, fry was transferred to a RAS for grow-out for 120 days and masculinization evaluation.

Spawning's occur the second day of adult stocking, but cannibalistic behavior is commonly observed and repeated fight. Larvae production ranged from 450 to 1700 larvae by spawn. First feeding occur 5-6 days post hatching at 26-28°C. Results on masculinization indicate that males tend to increasing according to treatment time (P=0.00). Sex proportion data indicate a minimal time of 45 days using oral administration of MT is required to increase male proportion production in the twoband cichlid. New experimental protocol is necessary to improve result on male production.

Table 1. Sex proportion in the twoband cichlid using oral administration of MT steroid.

Treatment	Males	Females	Intersex
CT	67.3	32.8	--
28D	67.2	24.4	8.3
45D	86.7	13.3	0.0
60D	83.0	10.4	6.7

ADVANTAGES OF THE IMPLEMENTATION OF GENETIC TECHNOLOGIES FORT HE PRODUCTION IMPROVEMENT OF JAPANESE OYSTER *Crassostrea gigas* CULTURE

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Context / Problem: Japanese oyster culture is done in Mexico (in Sonora, Sinaloa, Baja California and Baja California Sur) from the 80´s. However, grow out is plagued by low yields. This can be associated with endogamy as there are no natural populations of the species to maintain adequate population sizes, since it is a species form Asia. Additionally, the use of diploid seed for culture is also a problem, as they are fertile and grow significantly less than triploids, frequently spawning resulting in flaccid individuals.

To solve this problem, biotechnology alternatives were incorporated to improve production of the Japanese oyster in Northwest Mexico. Among them, the use of certified triploid organism. The triploid oyster marketed worldwide is known as a "biological triploid" derived from mating a tetraploid and a diploid. The competitive advantages of production, market value and financial projections were analyzed when the hatchery applies our technology package. This package includes two manuals and technical support tailored to the needs of the client.

Main competitive advantages of a selection program and triploid production implementation represent a 10% increase in final weight or height per generation based on selection, and up to 70% weight increase when comparing triploid to diploid organisms, particularly in stress environments. (see Ibarra AM, Ascencio-Michel R, Ramírez JL, *et al*. 2017, J. Shellfish Res. 36, 119-139). Integrating selection and biological triploid production significant results can be obtained after three generations (time varies depending of culture site and water temperature).

FED-BATCH CULTIVATION OF MICROALGA *Picocholorum* sp. AND HARVESTING OF BIOMASS BY BIO-FLOCCULATION

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Microalgae are promising sustainable feedstock for the production of biofuels, food, feed, chemicals, nutraceuticals and pharmaceuticals. Green marine microalga *Picochlorum* displays a high growth rate, broad salinity tolerance and high-temperature tolerance, making this genus suitable for outdoor cultivation. One of the major bottlenecks in the commercialization of microalgal biomass production is energy-intensive harvesting methods. Bio-flocculation is an efficient, low-cost method for harvesting microalgal biomass. Conventional chemical flocculants, commonly used to improve the harvesting efficiency, contaminate growth medium and make biomass unsuitable for food and feed production. Bio-flocculation uses microorganisms as bio-flocculants, especially fungi, which could also improve the quality of isolated biomass.

To investigate the growth productivity, *Picochlorum* sp. was cultivated phototrophically in a 35 L bubble column bioreactor. Cultivation was conducted in two phases using f/2 medium without pH and temperature control. During the first cultivation phase, a high growth rate was maintained by feeding the culture with nitrogen and phosphate sources. When cells entered the stationary phase on the 16th day of cultivation, nitrogen and phosphate sources were depleted. In the second phase of cultivation, the culture was fed with an inorganic carbon source (1 g L^{-1}), sodium bicarbonate. The culture was also fed on the 21st and 23rd days with 1 and 0.5 g L^{-1} of sodium bicarbonate, respectively. Maximal cell number and dry cell weight concentrations were 3.46×10^8 cells per millilitre and 1.68 g L^{-1}, respectively. Biomass productivity was 0.0646 g L^{-1} d^{-1}. Cell biomass was analyzed for macromolecular composition (carbohydrates, lipids and proteins), fatty acid composition and chlorophyll content. Proteins were the most abundant macromolecule in cell biomass, followed by carbohydrates as the main source of reserve energy with a maximal 37.7 % in dry cell weight. *Picochlorum* sp. had a low capacity for lipid accumulation; at the end of cultivation, the lipid content was only 8 % in dry cell weight. The most dominant fatty acid was linoleic (C18:2), followed by heptadecanoic (C17:0) and palmitic (C16:0). These fatty acids comprised more than 70 % of all fatty acids. During the second cultivation phase, cells also accumulated a significant amount of linolenic (C18:3) acid.

Microalgal biomass was harvested by bio-flocculation under optimized conditions using pellets of fungus *Mortierella isabellina.* This fungus is known for its ability to accumulate a significant quantity of lipids abundant in unsaturated fatty acids. The recovery efficiency of microalgal biomass was more than 80 % (g g^{-1}) after 150 min. The developed bio-flocculation system is a suitable method for chemical-free harvesting of microalgae that could be used as feedstock for feed or food production.

ORGANIC ACID IN BFT WATER FOR NILE

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The goal of this work was to evaluate the use of organic acid (Eurotec Nutrition, Brazil) in the water of biofloc technology to reared Nile tilapia (*Oreochromis niloticus*). The work was divided into two steps: 1st) *in vitro* assays, inhibition test and dose-response; and 2nd) *in vivo* assay with Nile tilapia juveniles in BFT.

For the *in vitro* assays, minimal inhibitory concentration (MIC) assays were performed against seven pathogenic bacteria (two *Aeromonas hidrophyla*, *A. veronii*, *Staphylococcus aureus*, *Citrobacter freundii*, *Vibrio parahaemolyticus*, and *V. alginolyticus*). For the dose-response test, 2.5 ppm, 5.0 ppm, 10.0 ppm and 20.0 ppm (doses defined from the product's user manual) were evaluated in triplicate. Fifteen bottle units with a capacity of 1 liter were used, all bottles received a constant and independent aeration system, which entered through the lid. For *in vivo* assay, in experimental units (8 retangular tanks, 0.72 m^2 x 0.35 m) with 250 L, water fertilization was carried out with a carbon source (sugar) and powdered diet to keep the carbon:nitrogen (C:N) ratio 10:1, resulting in an initial solids concentration of 100.0 mg L^{-1}. The experimental units were divided into two groups (organic acid and control) in quadruplicate, each tank with 20 fish. Fish were fed three times per day (8:00, 12:00 and 16:00), with commercial diet, with 3% of the fish biomass. Dissolved oxygen and temperature were performed measured twice a day. Total suspended solids (TSS), settleable solids, pH, hardness and alkalinity were monitored twice a week. Ammonia (total ammonia nitrogen – TAN), nitrite-N, and nitrate-N were also monitored twice a week. Growth performance was determined after six weeks.

Organic acid had an inhibitory effect against all pathogenic bacteria tested (Table 1). The largest inhibition halo (mm) was obtained against the bacterium *S. aureus* (60.00 ± 5.00), and the smallest halos were obtained against *A. hidrophyla* (42.00 ± 3.46) and *C. freundii* (41.00 ± 1.73). The 2.5 ppm dose presented the most interesting results for the *in vivo* assay, as it did not significantly reduce the pH and alkalinity of the water. Nile tilapia shown FCR closed 1.0 in both treatments, as well, SGR above 3.4%.day^{-1} acid reduced the nitrogen compounds orthophosphate, the other parameters did not diverge (Table 1).

The organic acid in the water in BFT, at a dosage of 2.5ppm, proved to be favorable against the reduction of toxic ammonia, nitrite and orthophosphate, improving the Nile tilapia culture environment.

Table 1. BFT water quality parameters

	$^2NH_3$ (mg. L^{-1})	$^3NO_2$ (mg. L^{-1})	$^4NO_3^{-2}$ (mg. L^{-1})	$^5PO_4^{3-}$ (mg. L^{-1})
Control	0,05 ± 0,01*	13,58 ± 0,92*	51,30 ± 65,38	2,70 ± 0,37*
Organic Acid	0,02 ± 0,02*	8,06 ± 2,41*	26,98 ± 20,50	1,67 ± 0,32*

* It indicates a significant difference ($p<0.05$) between treatments in the T test.

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Pichia guilliermondii ENHANCES SHRIMP IMMUNITY AND GROWTH

François Jégou*[1]
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Sarah Cooper[2], Yoav Rosen[1], Delphine Weissman[1]

Pathogens, environmental stressors and opportunistic diseases are ubiquitous in aquaculture. Achieving efficient and profitable production, especially with the increasing global threat of antimicrobial resistance, is critical for producers. Helping aquaculture species to better perform with infectious and physiological pressures is therefore of high interest and a focus of research.

Cardozo *et al.*, 2018[3] compared *in vitro* bacterial aggregation capacity of inactivated *Pichia guilliermondii* (*Pg*) and *Saccharomyces cerevisiae* (*Sc*), with Mannose as the positive control. *Pg* demonstrated twice the aggregation capacity at all commercial dosages compared to that of *Sc* (Fig 1). Pathogen aggregation is critical in innate immunity and homeostasis.

To assess the bacterial aggregation capacity of *Pg in vivo*, a bacterial clearance study was performed: *Litopenaeus vannamei* shrimp were injected with *Vibrio harveyi*. Three hours post-injection, *V. harveyi* concentration in hemolymph in the *Pg* fed shrimp was more than 3 times lower compared to control-fed shrimp (Fig 2). It was also shown that the granular hemocyte concentration in hemolymph was 56% higher after 4 weeks in *Pg* fed shrimp compared to that in control shrimp. (Fig 3) . Granular hemocytes are key for an effective and rapid immune response in shrimp, and represent an ideal parameter in monitoring shrimp immune potency.

In order to confirm the potential benefit of *Pg* in shrimp culture conditions, 2 challenge studies were performed in *L. vannamei* shrimp supplemented with 0.1% *Pg*[4]*:* Survival of shrimp 5 weeks after White Spot Syndrome Virus challenge increased by 84% compared to unsupplemented control shrimp, whereas it was 76% higher 5 weeks after *V. parahaemolyticus* challenge. In field conditions without specific pathogenic challenge, *vannamei* shrimp supplemented with 0.1% *Pg* showed significant 10% increased growth compared to control at the end of a 7 week trial.

Inactivated whole yeast *Pichia guilliermondii* feed supplementation offers unique traits for use in shrimp culture and daily management, improving growth while significantly helping the shrimp to better cope with physiological and disease stress.

Fig 1

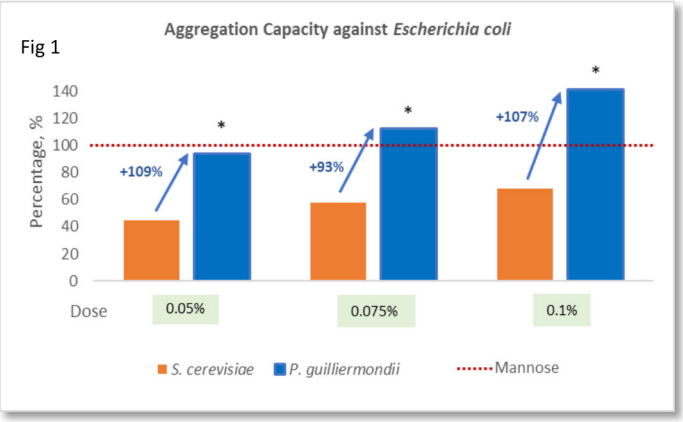
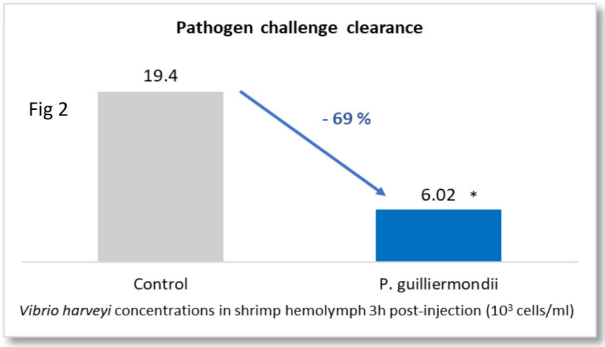
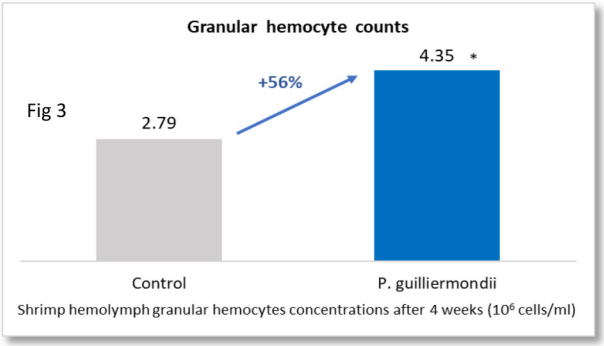


Fig 2



Vibrio harveyi concentrations in shrimp hemolymph 3h post-injection (10^3 cells/ml)

Fig 3



Shrimp hemolymph granular hemocytes concentrations after 4 weeks (10^6 cells/ml)

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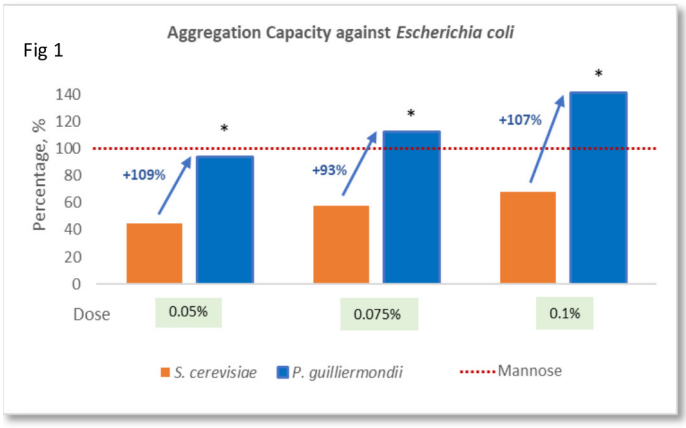

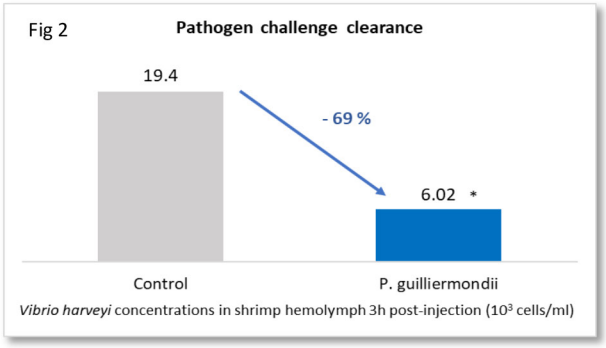

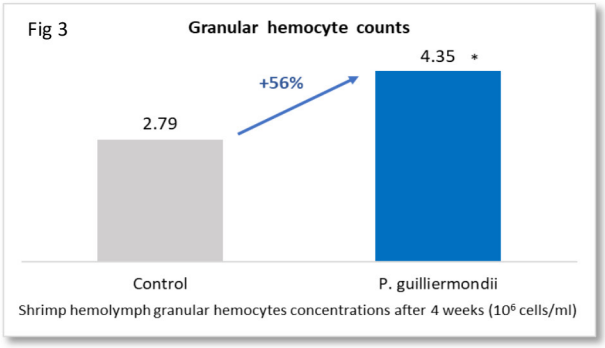

EVALUATION OF AN INTEGRATED MULTITROPHIC AQUACULTURE SYSTEM (IMTA) FOR CHILI PRODUCTION

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In aquaculture, to increase production and mitigate competition for water resources, multiple technologies have been implemented, such as AMTI, which consists of the culture of two or more species of different trophic levels, so that the waste from one is used by another (González-Henríquez, 2015). The objective of the present research was to develop an integrated multitrophic aquaculture system (IMTA) by tilapia (*Oreochromis niloticus*), Malaysian prawn (*Macrobrachium rosenbergii*) for the production of two species of chili plants.

The experimental design was completely randomized with two treatments (IMTA and control) in triplicate. Each experimental unit consisted of 85 tilapia m^3, 20 prawn m^2, 20 chili plants (serrano and jalapeño) per m^2. The control consisted of the individual cultivation of each species (plants in hydroponic system). The study lasted 180 days; the parameters of water quality, productive performance of the animals and plants in culture, as well as nutrients (K, Na, Ca and P) were evaluated both in the water culture and in the sap of the plants. Data were analyzed with a Student's t-test at $p<0.05$.

The physicochemical variables in water showed similar concentrations in IMTA and the control, except for NO_3 which presented significantly higher values in the water plants. Electrical conductivity (Ce) was higher in organisms cultured in IMTA. Plants in the control group had higher values of K, while Na and Ca concentrations were higher in organisms cultured in IMTA, except between tilapia in IMTA and control, when the values were similar. Values of P were high and similar among organisms cultured in IMTA and control plants. The productive performance in tilapia was higher in the IMTA, and the prawns in the control group. Chili plants had higher survival and growth in the IMTA and only jalapeño plants produced fruit in the IMTA (Table 1).

Table 1. Physicochemical and productive variables evaluated in water and culture organisms in the IMTA and control.

Variable	IMTA				CONTROL			
Physicoquemical	**Tilapia**	**Prawn**	**Plants**		**Tilapia**	**Prawn**	**Plants (hydroponics)**	
T (°C)	28.05±1.42^a	28.05±1.42^a	28.05±1.42^a		28.05±1.42^a	28.05±1.42^a	28.05±1.42^a	
OD (%)	96.54±4.42^a	96.54±4.42^a	96.54±4.42^a		96.54±4.42^a	96.54±4.42^a	96.54±4.42^a	
NH_4 (mg L^{-1})	0.91±3.23^b	0.88±0.27^b	1.78±0.24^a		1.24±0.25^a	0.36±0.04^c	0.35±0^c	
NO_2 (mg L^{-1})	1.60±1.18^b	1.48±1.07^b	0.75±0.25^c		2.54±0.99^a	0.90±0.06^c	0.91±0.94^c	
NO_3 (mg L^{-1})	187.11±18.46^c	184.27±16.39^c	298.57±46.57^b		152.78±15.58^c	45.54±8.96^d	485.1±201.35^a	
pH	6.33±0.05^b	6.34±0.11^b	6.01±0.11^c		6.30±0.15^b	7.12±0.13^a	6.60±0.27^b	
DG (mg L^{-1})	41.40±1.22^a	40.74±1.28^a	45.71±0^a		40.74±1.28^a	43.50±2.43^a	41.33±2.30^a	
Ce (S/cm)	0.861±0.083^a	0.823±0.092^a	0.813±0.150^a		0.530±0.083^b	0.315±0.049^d	0.412±0.079^c	
Nutrient								
K (mg L^{-1})	57.90±4.79^c	55.1±8.35^c	60.66 ± 11.35^c		26.67±3.37^d	10.7±2.42^e	177.29 ± 82.53^a	
Na (mg L^{-1})	36.27±15.62^a	24.06±1.95^b	26.90 ± 3.62^b		16.9±1.15^c	9.56±0.66^d	12.73 ± 1.62^c	
Ca (mg L^{-1})	97.53±16.33^c	142.8±16.28^a	136.61 ± 15.01^a		99.56±19.92^c	66.4±1.66^d	126.83 ± 20.69^a	
P (mg L^{-1})	1500±0	1500 ± 0	1500±0		823.33±165.03	326.67±97.13	1500±0	
Productive			**Serrano**	**Jalapeño**			**Serrano**	**Jalapeño**
Survival (%)	93.33±8.82	11.67±2.89	100	100	92.22±8.39	43.33±20.82	80	33.33±41.63
L initial (cm)	18.65±0.56	4.24±0.29	18.26±3.42	18.03±1.10	19.01±0.44	4.11±0.34	19.6±0.77	16.06±0.77
L final (cm)	26.3±0.87	10.02±1.15	36.60±10.88	44.73±20.60	25.76±0.24	11.54±0.89	20.6±0.84	20.6±0.84
Biomass (kg)	24.37±1.17	0.056±0.015			21.77±.49	0.33±0.04		
# fruits			0	5			0	0

Different letters mean significant differences at $p<0.05$. Sap nutrients in plants are not shown.

FATTY ACID PROFILE OF MACROALGA *Gongolaria barbata* FROM THE ADRIATIC SEA

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The aim of present research was to contribute to *G. barbata* chemodiversity and to the knowledge of specially protected *Cystoseria* areas. The fatty acid content was analyzed by gas chromatography with a flame ionization detector (GC-FID). The major saturated fatty acid (SFA) in *G. barbata* was palmitic acid (33.16 ± 0.41%) followed by stearic (8.22 ± 0.61%) and myristic acids (4.90 ± 0.07%). Besides oleic acid isomers (17.72 ± 0.18%), a significant amounts of arachidonic acid (ARA) were found (11.96 ± 0.19%) among polyunsaturated FAs. ARA is an important biomembranes constituent, a precursor of prostlandins and other eicosanoids.

From nutritional aspect, FAs are very important components that provide normal functioning of the human metabolism, health and prevention of diseases. The composition and content of fatty acids in macroalgae samples was determined using gas chromatography with flame-ionization detection after total lipid extraction was obtained by Folch method. Nutritional value of FAs is evaluated and expressed as nutritional indices and most widely used indices are: PUFA/SFA ratio, index of atherogenicity (IA), index of thrombogenicity(IT), unsaturated index (UI), hypocholesterolemic/hypercholesterolemic ratio (HH). IA and IT are indices correlated with potential effect of FA on cardiovascular health (CHV). IA and IT values for *G. barbata* obtained in this study were 1.11 and 1.32. When comparing IA and IT values of brown macroalgae to other foods, the values are higher than in crops, fish and meat but lower than in dairy products. UI indicated the degree of unsaturation in lipids and is commonly used for evaluation of PUFAs content and quality in macroalgae. UI for *G. barbata* calculated in this study was 101.49.

TABLE 1. Fatty acid profile of *Gongolaria barbata*

No.	Fatty acid	Av ± SD (%)
1.	Tetradecanoic acid (Myristic acid) (C14:0)	4.90 ± 0.07
2.	Pentanoic acid (C15:0)	0.43 ± 0.01
3.	Hexadecanoic acid (Palmitic acid) (C16:0)	33.16 ± 0.41
4.	Heptadecanoic acid (Margaric acid) (C17:0)	0.40 ± 0.01
5.	Octadecanoic acid (Stearic acid) (C18:0)	8.22 ± 0.61
6.	Eicosanoic acid (Arachidic acid) (C20:0)	0.61 ± 0.02
7.	Heneicosanoic acid (C21:0)	2.14 ± 0.02
8.	Docosanoic acid (Behenic acid) (C22:0)	0.58 ± 0.01
9.	Tricosanoic acid (C23:0)	0.46 ± 0.03
10.	Tetracosanoic acid (Lignoceric acid) (C24:0)	1.38 ± 0.02
Total saturated fatty acids (SFA)		52.28
11.	(*Z*)-Hexadec-9-enoic acid (Palmitoleic acid) (C16:1)	6.88 ± 0.12
12.	(*Z*)-Octadec-9-enoic acid+(*E*)-Octadec-9-enoic acid (*cis*-Oleic acid+*trans*-Oleic acid) (C18:1n-9c+t)	17.72 ± 0.18
13.	(*Z*)-Docos-13-enoic acid (Erucic acid) (C22:1n-9)	0.48 ± 0.06
Total monounsaturated fatty acids (MUFA)		25.08
14.	(*Z,Z*)-Octadeca-9,12-dienoic acid (*cis*-Linoleic acid) (C18:2n6-c)	5.11 ± 0.02
15.	(*Z,Z,Z*)-Octadeca-9,12,15-trienoic acid (α-Linolenic acid) (C18:3n-3)	2.62 ± 0.03
16.	(*Z,Z*)- Icosa-11,14-dienoic acid (Eicosadienoic acid) (C20:2n-6)	1.18 ± 0.02
17.	(*Z,Z,Z*)-Icosa-11,14,17-trienoic acid (Eicosatrienoic acid) (C20:3n-3)	0.66 ± 0.01
18.	(*Z,Z,Z,Z*)-Icosa-5,8,11,14-tetraenoic acid (Arachidonic acid) (C20:4n-6)	11.96 ± 0.19
19.	(*Z,Z,Z,Z,Z*)-Icosa-5,8,11,14,17-pentanoic acid (Eicosapentaenoic acid) (C20:5n-3)	0.48 ± 0.01
20.	(*Z,Z,Z,Z,Z,Z*)-docosa-4,7,10,13,16,19-hexaenoic acid (Docosahexaenoic acid) (C22:6n-3)	0.62 ± 0.00
Total polyunsaturated fatty acids (PUFA)		22.63
Total n-3 fatty acids (n-3 PUFA)		4.38
Total n-6 fatty acids (n-6 PUFA)		18.25
Nutritional indices		
PUFA/SFA		0.43
Index of Atherogenicity (IA)		1.11
Index of Thrombogenicity (IT)		1.32
Hypocholesterolemic/Hypecholesterolemic ratio (HH)		1.06
Unsaturation index (UI)		101.49

SUSTAINABLE INTENSIFICATION OF SHRIMP FARMING: I. GENERAL PRINCIPLES

Lorenzo M. Juarez Mabarak*

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Intensification is the application of additional resources to increase shrimp farm productivity. Such resources include management, labor, land, water, feeds, postlarvae, energy, aeration, and agrochemicals, among others. Ideally, intensification results in those additional resources getting amortized over the additional production in such a way that less of each resource ends up been used by unit of shrimp produced. Typically, intensification implies higher total investment and operation costs, but lower cost of production per kg of shrimp. As intensity increases biological and business risks also increase.

The concept of sustainability is subjective and can have different meanings under different disciplines. It normally encompasses environmental, economic, and social components with a fourth dimension of corporate governance. In this presentation I will mostly use a utilitarian definition based on the quantifying of resources used by unit of shrimp produced.

There are two main reasons to intensify shrimp farms: (1) to make more efficient use of resources and (2) to improve enterprise profitability and competitiveness. A recent study showed that 94% of all farmed shrimp production comes from only 58% of the farming area, or conversely that only 6% comes from the remaining 42% of the area. A large portion of the global pond area is highly inefficient and hides large environmental costs.

Intensive systems can be better for the environment because they use less land; especially pristine or highly sensible land, exchange less water and contaminants, and make generally more efficient use of resources such as feeds.

Examples of intensive systems, and of areas or countries that have intensified, are presented, and reasons for resistance to intensification are shown together with possible solutions and opportunities.

ENVIRONMENTAL SUSTAINABILITY AND INTENSIFICATION IN AQUACULTURE: II. EFFICIENCY IS THE NAME OF THE GAME

Lorenzo M. Juarez Mabarak*

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Properly executed intensification helps the planet and helps the enterprise by making more efficient use of resources and producing products that cost less to produce per unit. This paper begins with a bird's eye look at the state of the planet in relation to the major environmental problems, including climate change, ocean acidification, plastic residues, rates of extinction, and the state of world's fisheries. Sustainability is defined according to different disciplines and a practical definition is presented in terms of resource use efficiency. The main recognized environmental impacts of aquaculture are reviewed with emphasis on their management and minimization. We end by discussing the future of aquaculture sustainability and the technologies that will make it more friendly to the environment

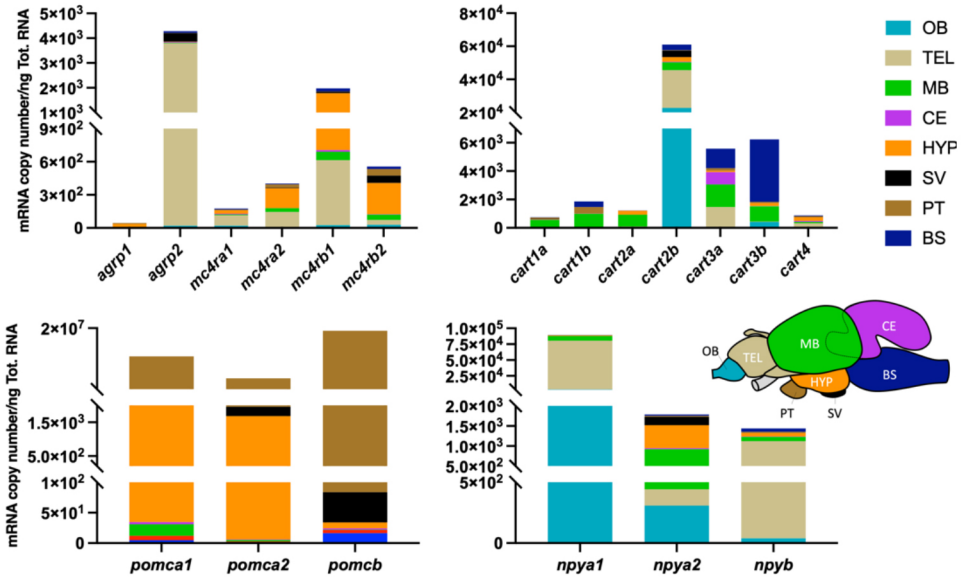

Figure 1. The mRNA expression of *agrp*, *mc4r*, *cart, pomc* and *npy* paralogs in eight brain regions (Insert: illustration of brain dissection) in Atlantic salmon. The genes were grouped according to their expression level to fit in the graphs.

FREE AMINO-ACIDS MIX MADE OF POULTRY KERATIN IMPROVES SURVIVAL OF WHITELEG SHRIMP POST LARVAE *Litopenaeus vannamei* CHALLENGED WITH ACUTE HEPATOPANCREATIC NECROSIS DISEASE AND WHITE SPOT SYNDROME VIRUS

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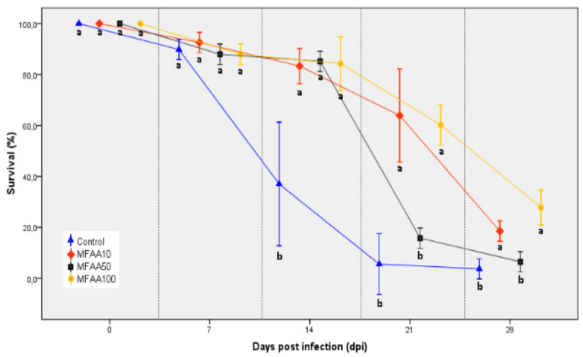
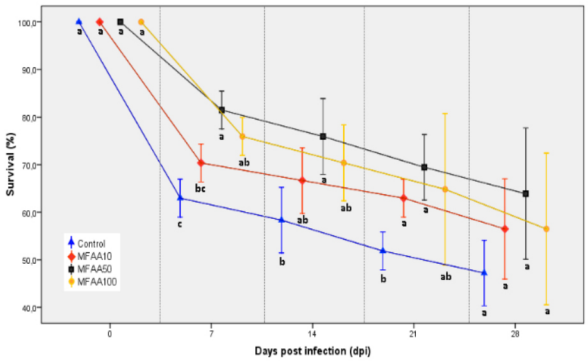
With their low molecular weight and high level of assimilation, mixes of free amino acids (MFAA) obtained from poultry keratin extensive hydrolysis are potential interesting candidates for aquaculture feeds dedicated to first development stages. In addition to previous results underlining MFAA positive effect on shrimp zootechnical performances, further investigations were conducted in partnership with Incabiotec/Concepto Azul, Tumbes, Peru, to evaluate potential of MFAA as new efficient solutions to improve immune response of white shrimp, *L.vannamei*, in case of bacteriological and viral challenges. Two trials, I and II, were conducted to evaluate the effects of MFAA on whiteleg shrimp *Litopenaeus vannamei* post larvae (PL). Both trials included a growth phase and experimental challenges. In these two trials, PL were fed four diets (control; control+1% MFAA; control+5% MFAA; control+10% MFAA) respectively for 28 days (trial I) and 21 days (trial II) for growth phases. Following this growth phases, animals were either experimentally infected with white spot syndrome virus (WSSV group) or Vibrio parahaemolyticus, with a toxin gene-bearing plasmid responsible for acute hepatopancreatic necrosis disease (AHPND group), or mock infected (non-infected control) considering four diets treatments (control; control+1% MFAA; control+5% MFAA; control+10% MFAA) during 28 days for trials I and II.

In these two trials, survival and biomass reached higher rates in WSSV infection groups, for PL fed with MFAA (Graph 1). For AHPND infection group in trial II, survival and biomass were also higher for PL fed with MFAA (Graph 2). Those results show the potential of MFAA to enhance shrimp PL performance and their application as shrimp feeding ingredients with functional benefits on animal survival in case of immune challenge.

Graph 1. Evolution of Survival week by week after WSSV infection in Trial I (95% CI of averages and Duncan test by week)



Graph 2. Evolution of Survival week by week after AHPND infection in Trial II (95% CI of averages and Duncan test by week)



DE NOVO ASSEMBLY AND ANNOTATION OF THE TRANSCRIPTOME OF THE NARROW-CLAWED CRAYFISH *Pontastacus leptodactylus* IN CASPIAN SEA

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The narrow-clawed crayfish *Pontastacus leptodactylus* (Eschscholtz, 1823) is an important cultured crayfish species in Iran. For further genetic improvement, however, our current knowledge of genetic resources in this species remains very limited. We conducted comparative analysis of the gills transcriptomes of *P. leptodactylus* from two different environments (Caspian Sea and Aras reservoir) based on Illumina RNA-seq technology. The transcriptome sequencing of cDNA libraries generated 419.2 million clean reads, the N50 and mean length of the identified 78895 unigenes were 1648and 864bp, respectively. A total of 3,759 differentially expressed genes (DEGs) were obtained between the two environments, of which 1,443 predicted to be up-regulated in Aras reservoir and 2,316 in Caspian Sea samples. A total of six differential genes related to the osmoregulation were selected. The qPCR analysis showed that the expression changes of these six differential genes were consistent with the results of transcriptome sequencing. Total number of filtered SNPs for Caspian Sea and Aras reservoir samples were 259 and 193, respectively. In addition, total number of identified single sequence repeats (SSRs) were 14,235. The putative DEGs between the gills provide an important source of information for further study of the osmoregulation related genes in *P. leptodactylus* as well as development of molecular markers.

ABNORMAL BEHAVIOR IN ROCK BREAM *Oplegnathus fasciatus* DETECTED USING DEEP LEARNING-BASED IMAGE ANALYSIS

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Fish production by the aquaculture industry has steadily increased to provide alternative fish resources. To overcome difficulties associated with a manual labor-intensive farming technologies and create a more systematic aquaculture management system that is compatible with the oncoming fourth industrial evolution, it is important to automate some aspects of the aquaculture industry such as water quality detection, automatic feeding systems, and real-time underwater monitoring. Despite of technologies have already been adapted to monitor physical conditions, those for automatic monitoring of fish behavior are still needed for real-time fish condition assessment. In this study, we applied the YOLO deep learning algorithm to detect abnormal swimming behavior of rock bream *Oplegnathus fasciatus*, based on fish movement data. We recorded images of rock bream before and after adding an anesthetic (MS-222) or the replacement of seawater with fresh water and then evaluated the ability of algorithm to detect fish displaying abnormal behavior. The proposed algorithm showed a high accuracy (88.1%) in discriminating normal and abnormal rock bream behavior. We conclude that artificial intelligence-based detection of abnormal behavior can be applied to develop an automatic biomanagement system for use in the aquaculture industry.

The collected data included 10,110 rock bream images obtained from the video recording. Among these, 210 images were randomly selected and divided these into a training dataset containing 168 images and a test dataset containing 42 images. Images of rock bream swimming upright and lying on their side were considered to exhibit normal and abnormal swimming, respectively.

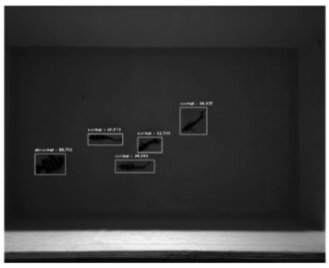

FIG 1. Detection of fish status (normal or abnormal) using YOLO v3 algorithm

TABLE 1. Confusion matrix used to evaluate the ability of deep learning algorithm.

Abnormal class		Actual behavior	
		Abnormal	Normal
Predicted behavior	Abnormal	25 (True Positive)	2 (False Positive)
	Normal	0 (False Negative)	78 (True Negative)

TABLE 2. Performance evaluation for abnormal object detection of rock bream.

Indicator	Performance
Total accuracy	**0.981**
$P_{abnormal}$	0.926
$R_{abnormal}$	1.000
$F1_{abnormal}$	0.962

GOOD AQUACULTURE PRACTICES; THE COLLABORATIVE USE AND BENEFITS

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The Joint Institute for Food Safety and Applied Nutrition (JIFSAN) is a collaboration between the United States Food and Drug Administration (FDA) and the University of Maryland (UM). JIFSAN has developed an aquaculture program that links food safety and disease prevention. It's called Good Aquaculture Practices (GAqPs). GAqPs are widely used and implemented around the world. This presentation is a short synopsis on GAqPs and a request for to help take GAqPs to the next level where it can be universally used and recognized for a wide variety of purposes. GAqPs could be used or integrated into new or existing programs to show food safety and disease controls are in place by the following:

- Individual aquaculture farms for their buyers;
- Governments for their farm and or processor registration and certification programs;
- Private third-party certification programs to verify food safety and disease prevention;
- Processors in their HACCP or Preventative Control programs to act as their food safety controls for aquaculture related food safety hazards;
- Academia to train future aquaculture professionals in food safety and disease prevention;
- The aquaculture industry and others to demonstrate to the general public that aquaculture products are safe, sustainable, and free of hazardous residues and pathogens.

POTENCIAL DE LA ACUACULTURA ORGÁNICA EN MÉXICO

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La acuacultura como producción orgánica de alimentos puede lograr la seguridad alimentaria de los países sin comprometer los recursos naturales de los mismos, ya que ha logrado satisfacer las demandas de la sociedad. Esta práctica tiene como finalidad establecer sistemas ecológicos que no dañen al ambiente y que consideren el bienestar animal, mientras se producen alimentos saludables y de alta calidad. La acuacultura orgánica, evita el uso de sustancias tóxicas que afecten al ambiente o a la salud humana de manera negativa, además de ofrecer soluciones y beneficios a los mercados involucrados. Es así, como el objetivo del presente trabajo fue analizar el potencial de la acuacultura orgánica en México. Debido a la actitud de las personas por dejar de consumir alimentos industrializados, se ha incrementado la demanda de alimentos más nutritivos e inocuos, el aumento del consumo per cápita de productos orgánicos que ha ocasionado este cambio de actitud provoca que la acuacultura orgánica sea vista como una práctica económica rentable. Por ello, se estima que la acuacultura orgánica comenzará a tomar relevancia en el mundo y en el país, aunado a la concientización de las personas por el agotamiento de los recursos naturales y por el daño a la salud humana que causan de manera directa o indirecta los sistemas de producción industrializados; De este modo la acuacultura orgánica se ha empezado a considerar como una opción que puede impulsar el desarrollo del mercado interno. Es así como a través de la acuacultura orgánica pueden elaborarse políticas públicas orientadas al desarrollo e implementación de programas económicos y/o sociales que beneficien a todos los involucrados, mientras se fomenta la producción de alimentos orgánicos orientados al desarrollo sustentable. Es importante mencionar que el etiquetado orgánico de los alimentos producidos en los sistemas acuícolas hace que los consumidores opten por adquirir dichos productos por sus implicaciones ambientales y productivas (carne de mejor calidad), ya que garantizan ser alimentos saludables e inocuos, producidos de manera amigable con el medio ambiente. El presente trabajo consistió en revisar los protocolos de la producción orgánica a nivel mundial y nacional, y su aplicación en la acuacultura mexicana, especialmente en *Oreochromis niloticus* que es la especie que más se produce en el país y que tiene el mayor potencial de producirse orgánicamente por su calidad de omnívora. La mayor limitación es el alimento orgánico, pero este se puede producir *in situ* a través de varias tecnologías como Biofloc, Predigeridos y/o Fermentados, entre otros, incluso se bajarían los costos de producción al no consumir alimento balanceado comercial. Se concluye que la acuacultura orgánica tiene el potencial de atender problemas ambientales, económicos y sociales en el país.

INTENSIVE PRODUCTION SYSTEM FOR PACIFIC WHITE SHRIMP, *L. vannamei*

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We have created a range of scaled, modular aquaculture production systems based on larger-scale RAS systems – the Mini Indoor Aquaculture Facilities. The systems provide optimal indoor growing conditions for smaller sized production with production sizes of 25, 50, 100 and multiples of 100 metric tons per year.

- Minimizes exploitation: Indoor, efficient RAS production minimizes the need for exploitation of marine resources.
- Minimal environmental impact: RAS avoids water pollution from feed, feces and chemical waste.
- Geographically unconstrained: Allowing for near market production and low carbon footprint.
- Disease control: Complete bio-security significantly diminishes risk of disease (removing the need for antibiotics).
- Traceability & quality: Full control of growth parameters allows for full traceability and improved product quality.
- Fast growth / continuous harvest: RAS can allow for faster growth and stable, continuous production across the year.

Our RAS facilities provide full-scale solutions for commercial, intensive and sustainable shrimp production, including sophisticated water treatment systems to recirculate water in a closed-loop system, requiring zero chemicals or antibiotics.

TRANSCRIPTOMIC PROFILING OF JUVENILE AND ADULT STARRY FLOUNDER *Platichthys stellatus* EXPOSED TO HEAT STRESS

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Starry flounder (*Platichthys stellatus*) is a promising aquaculture fish farmed near the east coast of South Korea and China. The optimum temperature for growth of starry flounder is ranging 15°C to 19°C and it maintains feeding and growth under 18°C. Above 24°C, it got stressed with osmoregulatory disturbances. Since 2015, east coast of Korea, land base flounder farm is continually harmed with temperatures climbing to 28°C or higher, which induces long-term stress responses, thus lowering immunity and causing mass mortality. However, during high temperature in the recent summer, 1st year juvenile survived, and only 2nd year adult fish died in mass mortality in farms. We hypothesized that there is a difference in the heat resistance of starry flounder by age in heat environment. This study, we investigated the different response of juveniles and adult starry flounder to heat stress. This study compares the physiological and molecular consequences of exposure in starry flounder to increasing water temperature (2°C/day) from 16°C until the target temperature, 28.5±0.5 °C. We conducted a comparative transcriptome analysis of starry flounder in response to heat stress.

A total of 79,908,211 raw reads were obtained from the three libraries from the kidney, and clean reads were 77,872,948. After removing the redundant data, 127,460 unigenes were assembled. The GO functional analysis showed that 41,580 unigenes were successfully assigned into 50 categories of three major categories: biological process, cellular component, and molecular function. A total of 5,384 unigenes were annotated to 25 categories from the KOG annotation in which the cluster of general functional prediction only (R) represented the largest group (687, 12.8%), followed by function unknown (S) (651, 12.1%) and signal transduction mechanisms (T) (539, 9.8%).

TABLE 1. Summary of the sequencing data and assembled contigs and mapping statistics.

Items		16℃		28℃	
		juvenile	adult	juvenile	adult
Raw data	Number of paired-end reads	18,924,455	21,142,721	18,868,394	20,972,641
	Over Q30 Bases (%)	86.47	86.28	86.02	85.58
	Quality Score	34.2	34.13	34.1	34.02
	Number of bases (bp)	5,677,336,600	6,342,816,400	5,660,518,200	6,291,792,300
Raw sequences after trimming	Number of paired-end reads	18,487,499	20,621,036	18,381,233	20,383,180
	Number of paired-end reads (%)	97.7	97.3	97.33	97.17
	Number of bases	5,440,710,722	6,044,816,563	5,399,382,651	5,986,890,935
	Number of bases (%)	95.67	95.33	95.47	95.17
Mapping	Mapping rate into reference seq. (%)	65.5	67.41	59.43	61.56

BIO-INTEGRATED AQUAPONICS: AN INTENSIVE COMMERCIAL PRODUCTION OF TILAPIA IN AN INTEGRATED AQUAPONICS SYSTEM USING ALTERNATIVE FISH FEED

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Aquaponics 5.0 is a circular production Biosystem that is based on the integration of FIVE production components to make more efficient use of FIVE natural resources with FIVE principles that promote FIVE human values for the integration of FIVE business areas. This model of Aquaponics 5.0 has been able to be implemented from a home system or a rural system in which, in addition to obtaining Healthy Food, Education and Agrotourism are obtained, to a commercial system where the same integration produces by-products of high added value that increase profitability. of the company.

Aquaponics 5.0 is an example of the cyclical model of nature, which is based on the use of resources where the reduction of elements prevails. Aquaponics 5.0 uses Biomass conversion components that generate raw materials to convert them into insect or worm protein through Permaculture techniques, which are in turn used by Aquaculture crops that produce nutrients in the water for the development of plants that They rid the water of toxic waste. All this with the help of renewable energy that gives movement to the system and the water that is stored from rain and condensation.

A commercial system of Aquaponics 5.0 not only seeks a productive profit, but also a commercial one. In production, techniques of recirculation, biofloc, microalgae, mineralization, filtration, energy efficiency, generation of raw materials, among others, are implemented so that components such as aquaculture, hydroponics and livestock are integrated into a productive circle where outputs are inputs. to another production line. And with regard to its commercial advantage, today there is a greater number of consumers and markets that seek or demand different techniques and certifications that demand Healthy and Sustainable food, so this productive-commercial integration is really the future of the Sustainable Food.

PROYECTO CERO: SISTEMA HIBRIDO DE PRODUCCIÓN DE TILAPIA/ROBALO CON CAMARON EN SISTEMA DE RECIRCULACION DESACOPLADO CON ACUAPONIA

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La acuacultura, al igual que la agricultura, requiere de la explotación de 5 recursos necesarios para que ésta actividad crezca y se intensifique. Se requiere de un **ESPACIO** para colocar las unidades de cultivo; **AGUA** para como hábitat de los organismos acuáticos; **MATERIA PRIMA** para alimentar el cultivo; **ENERGÍA** para mantener o intensificar la producción; y finalmente se eliminan **DESECHOS** producto de la engorda.

Los sistemas de RECIRCULACION con **acuaponia**, se aprovechan los desechos de la acuacultura para integrarla en un sistema de producción de plantas sin suelo (hidroponia), en el cual, el 100% del agua es reutilizada y no existe una generación de desechos ya que todo se convierte en planta de alto valor comercial. Los Sistemas de Acuaponia involucrán la presencia de microorganismos que consumen y/o convierten los desechos de los peces, en elementos más simples y que de esta manera las plantas lo tomán fácilmente.

Los sistemas Híbridos-Integrados de Acuaponia, están diseñados para el aprovechamiento tanto del agua, aire, desechos, energía, materia prima como el alimento y fertilizantes. Esto es debido a que su sistema es completamente cerrado y toda el agua que circula a través de los peces es canalizada hacia las plantas, microalgas y cultivo de camarón alterno en biofloc. El dióxido de carbono producido por los peces y bacterias heterótrofas, es dirigido hacia plantas y microalgas para su crecimiento, y estas a su vez incorporan oxígeno que es extraído para su incorporación en los peces y camarones.

Este proyecto presenta una alternativa de producción sustentable de Tilapia-Robalo y Camarón, con el uso de desechos para producir plantas en sistema de Acuaponia a nivel comercial y de gran escala. Se presenta una analisis financiero y comercial.

ACUAPONIA COMO SISTEMA AGROTURÍSTICO PARA DAR VALOR A LA PRODUCCIÓN Y A LA EDUCACIÓN/AQUAPONICS AS A AGRO-TOURISTIC INVESTMENT

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Síntesis

Los sistemas de Acuaponia son muy conocidos por su alta sustentabilidad en el uso de recursos naturales, así como por la producción de peces y plantas de alto valor económico. Sin embargo, existe otro valor que cobra gran importancia debido al interés y curiosidad de la gente por conocer este sistema, y esto es un valor Agroturístico.

La acuaponia puede tener varias arístas en su implementación, es decir, producir peces, plantas, y otros subproductos. El valor agroturístico y la integración social son otros adeptos que pueden ganarse si se hace el diseño adecuado y adaptado a los requerimientos del lugar donde se alocará. Es decir, depende si es un restaurante, un hotel, una comunidad o una escuela, deberá buscarse el diseño adecuado que permita generar valor para que los clientes, ademas de poder consumir los productos, tambien sea un motivo por el cual asistan a ese lugar en específico.

En esta presentación se muestran una serie de experiencias a pequeña y gran escala, donde se ha podido observar el valor agroturístico que genera la acuaponia.

EVALUATION OF MARINE PROBIOTIC VS PROBIOTIC HGS-7, VITAMIN DSM USED IN POST LARVAL STAGE OF WHITE SHRIMP *Litopenaeus vannamei*, ATISA, TUMBES, PERU

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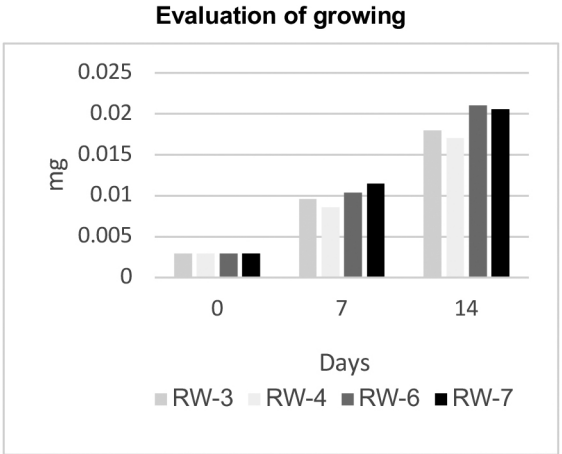
Probiotics are live microorganisms that, when administered in quantities adequate, are beneficial to the host´s health (FAO 2005). Have been consolidated as one of the natural alternatives of the use of antibiotics, because they do not generate effects colateral. Probiotics can supplant growth promoters and in adequate amounts reduce mortality and increase feed conversión (Gutierrez et 2013).

The experiment was carried out in September 2019, at the Atisa company, Puerto Pizarro, Tumbes, with a duration of 14 days, in which 6,560,000 postlarvae of white shrimp Litopenaeus vannamei were used, originating from the Arrecife laboratory, Ecuador. The larvae arrived in bags with oxygen inside a cardboard box, these were randomly distributed in 4 concrete raceways, each having a 1640000 with a density of 21 L / m3. A versus was carried out in which the RW-6 and RW-7 were added the marine probiotic in the food and for the RW-3 and RW-4 the probiotic HGS-7, vitamin DSM and glue were added in the white shrimp feed. The initial weight of the larvae was 310 larvae x gram, the initial feeding rate was 30% of the biomass, of the AQUATECH brand. About the quality of water (dissolved oxygen, pH, temperature and salinity) were evaluated every 2 hours with a multiparameter of the brand, while the chemical parameters of water (NH4, NO2, NO3) were evaluated with a spectrophotometer every 2 days during the experiment.

No significant difference was found at the end of evaluation in the productive indicators, such as specific and absolute growth rate, the same in the water quality parameters. However, the use of marine probiotic would save work time and costs.



Final weight, specific growth rate in the final evaluation

Treatment	RW-6	RW-7	RW-3	RW-4
FW (mg)	0.021	0.021	0.018	0.017
SGR (%)	14.1	14	13.2	12.8

USE OF IMMUNOMODULATORS FOR THE CONTROL OF THE IMMUNE AND INFLAMMATORY REPSONSE IN FISH FARMING.

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The increasing demand for seafood leads to the significant growth of aquaculture, however the reduction of land-water availability ratio, requires intensifying the culture system. This scenario needs to face certain technical problems as oxygen depletion, generation of high amount of nitrogen waste, large concentrations of fish in confined environment. These factors act as a stressors to the animals, and affect in negative ways physiological and zootechnical parameters like growth ratio, immune system response, and the risk of disease outbreaks.

Is understood by adaptogen a nontoxic substance and especially a plant extract that is held to increase the body's ability to resist the damaging effects of stress and promote or restore normal physiological functioning. Botanicals are nowadays considered to be safe additives that can be included in diets to improve the health and performance of animal as well as fish (Le et al., 2016; Fawole et al., 2016).

The objective of this study, carried out in Nile tilapia (*Oreochromis niloticus*), was to evaluate the effect of three nutraceutical compounds on growth, blood parameters, immune response, and liver health before and after the challenge with the bacteria *Aeromona Hydrophila*.

The results obtained during the study, show that the addition of nutraceuticals PIM1 and PIM 3 enhanced the zootechnical parameters, improved Nile tilapia survival against *Aeromona hydrophila* infection by increasing innate immunity and antioxidant response. The liver histology and lower CRP value revealed that PIM 1 and PIM 3 have the capacity to confer certain level of protection on the hepatic tissue of Nile Tilapia against pathogen aggression.

	Parameters					
	SOD (U/L)		**CAT (umol/ml/mins)**		**GPx (U/L)**	
Dietary group	Pre-challenge	Post-challenge	Pre-challenge	Post-challenge	Pre-challenge	Post-challenge
Control	56.60±7.35	1.24^{b}±0.17	0.63^{C}±0.06	39.93^{b}±1.87	278.33±39.42	26.26^{c}±3.09
PIM 1	57.81±0.52	**2.41^{a}±0.52**	**6.01^{A}±0.05**	**52.15^{a}±0.4**	**392.18±27.8**	**42.20a±2.27**
PIM 2	60.07±6.71	1.91^{ab}±0.03	**4.43^{AB}±0.94**	39.78^{b}±2.80	348.48±36.95	36.01^{ab}±2.81
PIM 3	60.07±6.08	**2.64^{a}±0.19**	**3.23^{B}±0.64**	40.22^{b}±0.73	277.58±36.02	29.82^{bc}±0.56
p-value	0.972	0.026*	0.005*	0.012*	0.130	0.016*

SURVEY FOR VIRAL PATHOGENS IN INVASIVE SPECIES *Penaeus monodon* CAUGHT IN THE GULF OF MEXICO COAST IN 2019 AND 2020

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Viral pathogens such as white spot (WSSV), Hepatopancreatitis Parvo virus (HPV), Penstyldensovirus (PstDV1), Taura syndrome (TSV) and Infectious myonecrosis virus (IMNV) were studied in 31 organism (10 juveniles and 21 adults) of *Penaeus monodon* captured on the coast of the Gulf of Mexico during 2019 (26 organisms) and 2020 (5 organisms). The average size and weight of specimens was 198 mm TL, 35 mm CL and 92.8 W (Table 1), where 55% were female and 45% male Stage I and II gonadal development stages were found in females

The fixation of branchial lamellae and tests were carried out individually, using the polymerase chain reaction (PCR) technique. Commercial kit IQ2000 ™ was used for each virus. The criteria for the interpretation of the results was according to the kit manufacturer's instructions. The 2% agarose gel results show us that no sample of the tiger shrimp caught in the Mexican coasts of Gulf de Mexico wasn´t positive for the fourth viruses (IMNV, TSV, HPV and WSSV) (Fig. 1) while PstDV1 was detected in two specimens collected from Veracruz and Tabasco. It is important to continue monitoring wild populations in the Gulf of Mexico, in order to establish a relationship between the health status of wild shrimp populations and invasive species *Penaeus monodon*.

Table 1 Biometry of specimens of *Penaeus monodon*

	LT (mm)	LC (mm)	PESO (gr)
Average	198	35	92.8
Maximum	330	75	326
Minimum	76	6	6

* LT from tip rostrum to tip telson

* LC Rostrum tip to posterior dorsal end of cephalothorax

* ♂ male and ♀ female

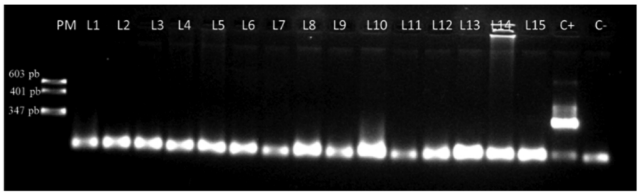

Figure 2. Results of the Polymerase Chain Reaction (PCR, end point) for the presence of White spot (WSSV, 630 bp), of the samples of the tiger shrimp *Penaeus monodon*. PM = molecular weight, L1 = Tamaulipas 1, L2 = Tamaulipas 2, L3 = Tamaulipas 3, L4 = Veracruz 1, L5 = Veracruz 2, L6 = Veracruz 3, L7 = Tabasco 1, L8 = Tabasco 2, L9 = Tabasco 3, L10 = Campeche 1, L11 = Campeche 2, L12 = Campeche 3, L13 = Yucatán 1, L14 = Yucatán 2, Yucatán 3 C + = positive control, C- = negative control

EVALUATION OF INLAND SALINE GROUNDWATER FOR *Penaeus brasiliensis* CULTURE IN YUCATAN

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For many decades, shrimp aquaculture has relied on the culture of a few penaeid species, with a preference on *Penaeus monodon* and *P. vannamei*. These species have achieved such success that they are being raised outside their natural regions of distribution, raising concern about potential escapes and the ecological harm they could cause. As a result, the culture of native species is of considerable importance around the world, and significant efforts must be done in this area. *Penaeus brasiliensis* is a native species in the Yucatan peninsula with a great potential for culture. This species has been shown to thrive and survive at quite high densities and salinities above its isosmotic point (> 25 ppt). Given the karstic structure of the Yucatan peninsula's ground and the saline intrusion tens of kilometers inland, inland culture of this native species could be a possibility. The aim of this work was to evaluate the suitability of the culture of *P. brasiliensis* in saline groundwater. Shrimp weighing 1.1 g were cultivated outdoors for 45 days in biofloc at 95 ind m^2 in 3.14 m^2 tanks (depth = 0.8 m) in triplicate in either saline groundwater collected from a well (60 m depth, salinity = 30 ppt) or natural seawater (salinity = 35 ppt). Shrimps were fed five times per day. Salinity was corrected with freshwater, aeration was provided with a 1 HP blower and diffusers, and each tank had an artificial substrate. Temperature and salinity decreased over time due to climatological conditions, which may be associated to a decrease in growth rate of shrimp at the end of the experiment. Nutrient levels (NH_4, NO_2, and NO_3) were optimal for this species culture, and alkalinity decreased over time because of the nitrification process (no corrections were made). Shrimp performance was evaluated at the end of the experiment in terms of final weight, survival, growth rate, and final biomass. There were no significant differences in shrimp production between saline groundwater and natural seawater (Table 1), suggesting that inland shrimp culture is a possibility.

Table 1 *Penaeus brasiliensis* growth performance (mean ± SD) grown for 45 days in either saline groundwater or natural seawater (N = 3 tanks per treatment). There were no significant differences between treatments in any variable (Student's t tests, $\alpha = 0.05$).).

Variable	Natural seawater	Saline groundwater
Initial weight (g)	1.1	1.1
Final weight (g)	4.8 ± 0.0	4.8 ± 0.2
Grams per week	0.41 ± 0.2	0.45 ± 0.1
Survival (%)	75.8 ± 5.4	74.6 ± 7.5
Final biomass (kg $tank^{-1}$)	1.0 ± 0.1	1.0 ± 0.1

EFFICIENT MICROALGAE PRODUCTION SYSTEM

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One of the important limitations in aquaculture of filter-feeding mollusks, shrimp and fish in their early stages of development is the availability of high-quality microalgae in sufficient quantities. Since the 1960s, technologies have been developed to mass-produce them monospecifically and without pathogenic bacteria for the organism to be fed, and it is not yet possible to replace them with inert foods. In most cases, the production of this live food in hatcheries is one of the phases with the highest cost and represents a risk for crops if a safe and financially viable technology is not available. This proposal presents an alternative developed at CIBNOR for the massive and continuous production of microalgae, with moderate investment and low production costs. This system has been tested and evaluated in a commercial mollusk hatchery, using pasteurized water and LED lighting as an alternative to lower production costs. Typical species used in aquaculture were produced in this system including: *Tisochrysis luthea, Pavolova lutheri, Chaetoceros gracilis, C. calcitrans and Thalassiosira pseudonana*. The technical and financial results will be presented in the runway of innovative technological developments at the World Aquaculture 2021 conference, as well as some business alternatives that investors could consider to cover the demand for live feed in mollusk, shrimp and fish hatcheries.

THE EFFECTS OF SALINITY REDUCTION RATES DURING *Litopenaeus vannamei* POSTLARVAE ACCLIMATION ON SURVIVAL AND GILLS AND HEPATOPANCREAS HISTOPATHOLOGY

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Although *Litopenaeus vannamei* can tolerate a wide range of salinity (1 to 50 g/L), adequate post-larvae (PLs) acclimation is crucial for its successful farming at low salinities. The aim of this work was to evaluate the effects of different rates of salinity reduction on the survival and gill and hepatopancreas histopathology of PLs during the acclimation period.

Shrimp (21-old-day PL; n=360) were distributed in nine 5 L tanks, where they were acclimated to low salinity via its decreasing from 35 to 1 g/L within 24 hours according two salinity reduction rate treatments: A1, constant rate of 1.45 g/L/h; A2, using 0.46%/h from 35 to 5 g/L, 0.25g/L/h from 5 to 2 g/L, and 0.13 g/L/h from 2 to 1 g/L. Also, a control group was maintained at 35 g/L but with a saltwater exchange rate of 40%/h. All groups were evaluated in triplicate. At the end of the acclimation (24h) and 24 hours later (24h+), survival was determined, and PLs were collected for further histological analysis of gill and hepatopancreas following hemocyanin and eosin method.

No differences were observed for survival at 24h. On the other hand, it was significantly lower in A1 at 24h+ compared to A2 and control (Table1). Despite no difference was observed for survival between A2 and control, the former exhibited several histopathological damages compared to control. For instance, in the epithelial structure of the gill filaments was observed hemocytic infiltration and a hypertrophy at 24h and 24h+ (Fig.1B and D). Hepatopancreas of the analyzed samples, in 70% it was not possible to identify the cells (R, B, F) from A2 (Fig.1F and H). While, at 24h+, 60% from A2 exhibited ruptured epithelial cells, abnormal appearance in the structure and reduction of B and R cells (Fig. 2D). In conclusion, salinity reduction during PLs acclimation may not reduce survival but causes several histopathological damages in gills and hepatopancreas.

TABLE 1: *L. vannamei* postlarvae's survival exposed to different rates of salinity reduction during acclimation

	Control	A1	A2	P
24 h	88.67 ± 4.42	83.60 ± 4.45	84.07 ± 5.21	0.058
24h+	83.33 ± 4.16[a]	65.47 ± 6.27[b]	81.07 ± 4.58[a]	0.026

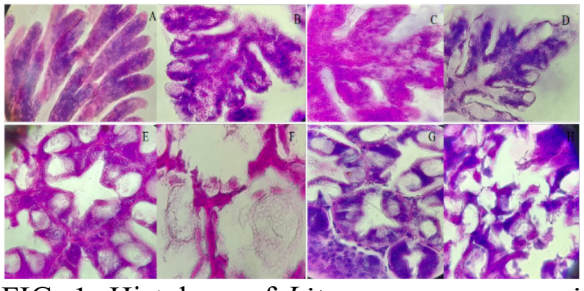

FIG. 1: Histology of *Litopenaeus vannamei* postlarvae gills and hepatopancreas in response to different rates of salinity reduction: Gill: control (A) and A2 (B) at 24h; and control (C) and A2 (D) at 24h+. Hepatopancreas: control (E) and A2 (F) at 24h; and control (G) and A2 (H) at 24h+. x100.

THE ROLE OF PREVENTATIVE FISH HEALTH MANAGEMENT IN SUSTAINABLE AQUACULTURE PRODUCTION (oral presentation)

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Global aquaculture of finfish has seen significant growth over the last decades, driven by static wild capture, increased consumer demand and expansion of global trade. Despite industrialization, consolidation and rapidly improving technologies, the aquaculture industry faces significant challenges to meet the sustainability criteria required for a healthy growth. There is a strong focus on animal welfare, food safety and environmental impact among authorities, consumers and the general public, and the increasing occurrence of antimicrobial resistance caused by use of antibiotics is a major concern to both animal- and human health.

A shift towards preventative fish health management is required to build a healthy and sustainable industry. Key factors are diagnostic tools, biosecurity, regulations, collaboration between stakeholders, introduction of vaccines and knowledge of fish health and welfare. A good example of successful preventative fish health management is salmonid farming in Norway, where more than 1.4 million tonnes are produced annually with minimal use of antibiotics. Vaccination is mandatory, and multivalent vaccines protecting against up to seven different pathogens are commonly used.

The most appropriate interventions depend on the level of intensification and industrialization. A large part of global aquaculture production is still done by small farms who supply local markets that do not necessarily recognize and reward environmentally sound and sustainable practices. When intensive monoculture evolves into industrialized production, introduction of advanced technology such as vaccines becomes feasible. Infrastructure, tools and procedures for husbandry and fish health management are then in place, and profit margins allow farmers to invest in new technologies. Among species being produced industrially today are salmonids, sea bass, sea bream, tilapia, pangasius, catfish, barramundi and groupers.

Important success factors when developing vaccines are knowing the species and having a proper understanding of the disease situation through appropriate diagnostics. The epidemiology is often complex and protection against several pathogens may be required in order to make a significant impact. Establishing good procedures for vaccination, such as health checks, proper equipment, training of personnel and quality control, is also key to ensure an optimal result and profitability.

We will present our experiences from fish health management and development and introduction of vaccines in established markets, and highlight challenges and success factors for emerging species.

IMPROVED GROWTH PERFORMANCE OF WHITELEG SHRIMP *Penaeus vannamei* USING A PHYTOGENIC FEED ADDITIVE

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Crustacean production in 2018 reached 9.4 million tonnes, being marine shrimps dominating the production of crustaceans typically farmed in coastal aquaculture. Besides, they are an important source of earnings for a number of developing countries in Asia and Latin America. Whiteleg shrimp (Penaeus vannamei) is the largest produced species with 4.9 million tonnes, accounting for the 52.9% of total crusteans produced. Optimizing shrimp production performance is crucial to the success of the sector. Efficient diet formulation together with successful health and welfare management determines higher production output. As part of a proactive approach to production health and performance, the use of functional and sustainable additives, such as phytogenics, has demonstrated to be an effective tool to boost shrimp performance.

Two different studies were conducted to determine the efficacy of a new phytogenic feed additive (PFA) mixture (saponins, spices and essential oils) on whiteleg shrimp growth performance and feed efficiency. In the first one, six hundred SPF shrimps of average initial weight of 1.9±0.26 g were allocated into three groups using with four replications for each treatment in 350 L tanks. In the second one, two hundred and forty shrimps of average initial weight of 1±0.01 g were randomly allocated into three groups with four replications for each treatment in 20 L baskets. In both studies, shrimps were fed for 6 weeks the different treatments: one control group (basal diet) and two groups receiving Syrena® Boost included at 200 & 400 mg/kg of feed, respectively.

After 6 weeks of feeding, results in the first study showed that shrimp survival did not differ between treatments being above 80%. In terms of growth performance, shrimps fed Syrena® Boost at both inclusion rates showed an increase of weight gain of 12.5% and 13.3%, respectively, with an increase in average daily growth (ADG) of 12.9% and 13.5%, respectively. Feed conversion ratio (FCR) was similar between treatments. In the second study, again shrimp survival did not differ between treatments being above 94% in all treatments. Shrimps fed Syrena® Boost at both inclusion rates showed again an increase of weight gain of 10.1% and 7.9%, respectively, being statistically significant ($p \leq 0.1$) at the lowest dose. ADG was also improved by 9.7% and 7.9%, respectively, again being statistically significant ($p \leq 0.1$) at the lowest dose. FCR was reduced by 3.7% and 1.7%, respectively. Altogether, these results indicate the positive growth performance effects of this new PFA in whiteleg shrimp.

EFFECT OF PELLED DIETS BASED ON FISH WASTE ON THE NUTRITION OF *Octopus maya* JUVENILES

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Octopus maya has been recognized as having a high potential for commercial cultivation. Various studies have allowed the development of a food formulation based on squid and crab meat for this species (Martínez et al., 2014; Gallardo et al, 2020). However, this diet can become expensive, which is why the present work seeks to replace these inputs with fish waste. For this, 3 diets (in triplicate) of partial and total replacement of squid and crab with fish waste were tested: CT) Control (freeze-dried squid and crab); partial replacement (DPR) and total replacement (DTR).

Octopuses 75 days old (DPH, 5g) from a single spawning were used, maintained with the control diet. Chac chi (*Haemulon plumieri*) fish waste was homogenized and mixed with vitamins and minerals, gelatin and crab protein hydrolyzate. The experiment lasted 43 d in rectangular tanks of 2 x 1.5 x 0.4 m with recirculating seawater and controlled conditions. 1" (5 cm) PVC pipes were used as shelters and fed 3 times a day.

The control diet allowed good growth and survival, whose values were higher than those observed in the octopuses maintained with the other diets (Table 1). However, both the growth and the survival of the octopuses subjected to the diets with the waste showed a great variation, which influenced that there were no differences between treatments with the applied statistic (ANOVA) (Table 1).

The results show that the diets formulated with "Chac chi" fish waste do not exceed the nutritional quality of the control diet whose inputs are freeze-dried, however, it could be a good alternative by modifying the feeding frequency in future experiments.

References

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Martínez R., … and Carlos Rosas. 2014. Growth, survival and physiological condition of Octopus maya when fed a successful formulated diet. *Aquaculture* 426-427: 310 – 317.

Table 1.- Growth and survival of *Octopus maya* juveniles fed pelleted diets. Mean ± S.D., n=3.

	CT	DPR	DTR	p-value
Gain weight	6.3 ± 2.8	3.1 ± 1	4.7 ± 2.2	0.27
SGR	2.5 ± 0.7	1.5 ± 0.3	2.1 ± 0.8	0.22
Survival	89 ± 19	44 ± 9.6	44 ± 34	0.06

REQUERIMIENTO DE PROTEÍNA Y ENERGÍA EN GAMITANA *Colossoma macropomum*

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La gamitana representa la segunda especie nativa más cultivada y consumida en la Amazonía peruana. Existen avances tecnológicos en la reproducción, producción de alevines, manejo y calidad de la carne; que han convertido a la gamitana en una de las especies nativas de mayor demanda por los mercados locales y regionales. Sin embargo, para lograr una actividad rentable para los piscicultores, se requiere el desarrollo de paquetes tecnológicos relacionados a la alimentación con el propósito de reducir su costo. Para el cultivo exitoso de los peces es necesario el desarrollo de alimentos balanceados con niveles adecuados de nutrientes para alcanzar el comportamiento productivo de acuerdo a su potencial genético.

El objetivo de la presente investigación es la determinación del requerimiento de proteína y energía digestible de la gamitana (Colossoma macropomum), utilizando la técnica de dosis-respuesta. Se diseñaron dos experimentos, en el primero para determinar el requerimiento de proteína, se formularon cinco dietas isoenergéticas (3.6 Mcal ED/Kg) con niveles crecientes de Proteína (30,32,34,36 y 38 %). En el segundo experimento, se formularon cinco dietas isoproteicas (35%) con niveles crecientes de ED (3.0,3.2, 3.4,3.6,3.8 Mcal ED/Kg). Cada dieta se administró a 3 grupos (repeticiones) durante 28 días. Los resultados obtenidos utilizando el análisis de regresión cuadrática (Figura 1 y 2) indican el requerimiento de proteína de 35.08% y el requerimiento de energía digestible de 3.43 Mcal ED/kg, para optimizar la conversión alimenticia de la gamitana.

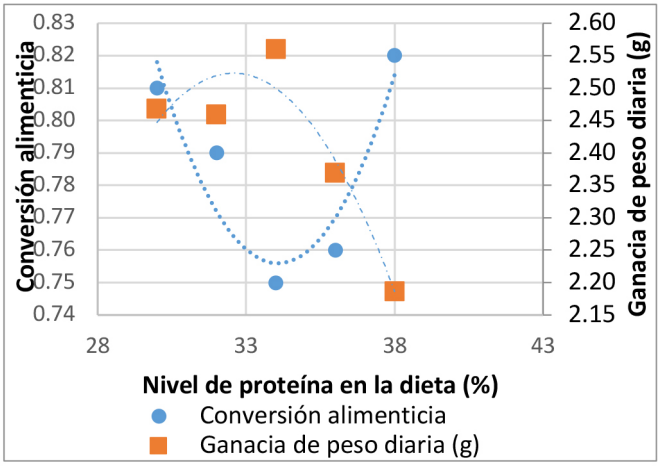

Figura 1. Relación entre la ganancia de peso diaria y la conversión alimenticia con el nivel de proteína dietario en base al análisis de regresión cuadrático

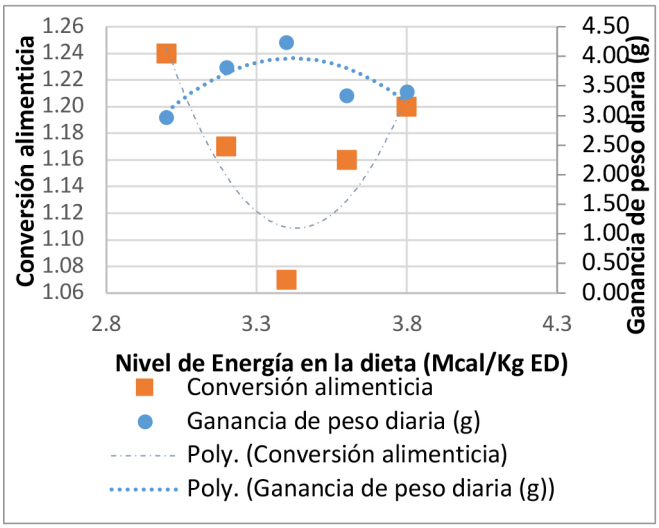

Figura 2. Relación entre la ganancia de peso diaria y la conversión alimenticia con el nivel de energía dietario en base al análisis de regresión cuadrático

SEASCAPE GENOMICS OF PINK ABALONE *Haliotis corrugata* AND BINATIONAL MANAGEMENT IMPLICATIONS

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Studies in seascape genomics have detected levels of adaptive divergence in marine species with high gene flow and homogeneous populations. This has highlighted the relevance of geographic and environmental factors as drivers for local adaptations in fishery resources. The pink abalone (*Haliotis corrugata*) is distributed from California, USA to Baja California Sur, Mexico; exposed to an environmental latitudinal gradient, influenced mainly by the California Current System (CC). The CC carries warm waters from California to the south, while the California Countercurrent carries warm waters, seasonally supplied by the North Equatorial Current to the north. The management of the pink abalone transcends the international border, while Mexico has an active fishery, organized in four administrative areas, the United States keeps the fishery in permanent closure. Both contrasting management strategies lack genetic information that supports such spatial arrangement. Recently, evidence of genomic-population structure was obtained in Mexico; however, the effect of the environment on genetic variation along the species distribution range remains unknown. The objective of the study is to define the biocomplexity of *Haliotis corrugata*, in terms of neutral and adaptive genomic structure and its underlying causal effects.

A total of 203 samples from 13 locations (Fig. 1a) were processed, covering the species' distribution, using ddRADseq. Overall, 2,242 SNP's were obtained, 2,231 neutral and 9 potentially adaptive. The neutral structure identified two groups: 1) California locations (PV, SCnaI, SCteI), and 2) Baja California Peninsula locations (GI, SJI, PCS, FS, CI, NI, PE, PuE, BA, SJ) (Fig 1b). In contrast, the adaptive structure separated a north group, from PV to CI, and a south group, from NI to SJ (Fig. 1c). The adaptive structure matches the grouping of localities based on PCA of environmental variables, where temperature and dissolved oxygen concentration distinguished a north group from PV to FSJ, and a south group from CI to SJ.

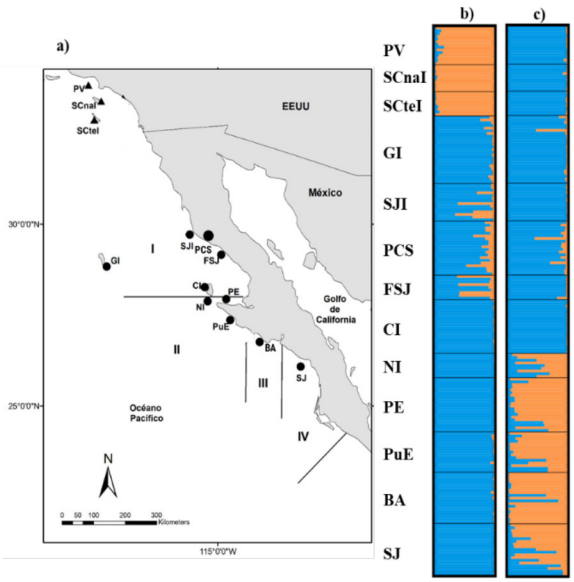

Fig. 1. Population genomic structure of the pink abalone (*H. corrugata*): a) Sampling localities; b) Neutral Structure and c) Adaptive Structure.

MICROALGAE-SUPPLEMENTED DIET IMPROVES THE SKELETAL HEALTH OF GILTHEAD SEABREAM *Sparus aurata* JUVENILES

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Fish cultured in intensive farming conditions may develop severe skeletal deformities that affect both their external morphology and welfare. In the case of commercially important fish species, such as the gilthead seabream (*Sparus aurata*), this is a matter of great concern and a major factor that affects production costs, downgrades hatcheries' production and fish market value. Among the solutions proposed to improve the skeletal deformities of aquaculture fish, the supplementation of their diet with natural compounds or extracts that stimulate skeletogenesis is increasingly seen as an economically sound approach to improve the competitiveness of the aquaculture industry and fish health.

In this regard, we supplemented a commercial fish diet with two ethanol extracts prepared from *Skeletonema* sp. and Tetraselmis sp. and containing bone anabolic compounds, and used them to feed gilthead seabream (*Sparus aurata*) juveniles of 4.8±0.7 g until they tripled their weight. Fish were housed at the Estação Piloto de Piscicultura de Olhão (EPPO/IPMA) in 250-L rectangular tanks with water renewal of approximately 350 L per hour and a temperature of 25.3±1.0ºC, and fed *ad libitum* 3 to 4 times a day.

Considering fish growth performance indicators, dietary treatments had no impact on feed conversion rate (FCR), although a slight increase was observed for fish fed with the diet supplemented with *Skeletonema* sp. extract. The total length was not significantly altered in fish fed supplemented versus control diets, but a positive effect was observed on the weight of the fish fed the diet supplemented with *Tetraselmis* sp., which consequently show a moderate increased condition factor. The analysis of several haematological parameters revealed that food supplementation with microalgae extracts does not influence fish physiological status or stress levels. In terms of bone parameters, the vertebrae of fish fed with the diet containing *Skeletonema* sp. extract showed a higher phosphorous content, although the optimal balance between calcium and phosphorous was maintained in the vertebrae of fish fed supplemented diets. The analysis of the type and severity of the skeletal deformities revealed that most of them are associated with the caudal fin complex, independently of the treatment, but that the incidence is reduced when fish are fed with the diets containing extract of *Skeletonema* sp. (deformity charge (df) = 1) or *Tetraselmis* sp. (df = 1.17) when compared to the control diet (df = 1.36). The expression of several bone marker genes was also altered upon dietary treatment, as it is the case of the osteoblast markers *sp7* and *col1a1*, that are, respectively, up and downregulated for fish treated with *Skeletonema* sp., and osteoclast marker *acp5*, that is upregulated for fish fed with *Tetraselmis* sp. Interestingly, the expression of the glutathione peroxidase *gpx1* was increased in the vertebrae of fish treated with both extracts, suggesting an activation of antioxidant mechanisms by both experimental diets. Overall, the data gathered in this study points out to a general improvement of fish skeletal status upon dietary treatment with microalgae extracts containing bone anabolic compounds, without compromising, and possibly improving, fish growth performance indicators.

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DEVELOPMENT AND IMPLEMENTATION OF EARLY AND FAST DIAGNOSTIC METHODS TO PREVENT MARINE FISH DISEASE OUTBREAKS IN AQUACULTURE

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As 40% of all aquaculture production is lost to disease, sanitary control of the farmed species and their environment is extremely relevant. Thus, an early and efficient detection of the classical and emerging pathologies (including virus, bacteria, parasites) is essential.

The molecular methodologies (commonly known as "omic tools") have gained much importance as invaluable tools to assess the welfare and health of farmed organisms. These "omic tools" can be used to develop databases with the signature of the major pathogens to allow the detection, identification and/or quantification of pathogenic agents in a faster and more reliable way, complementing the classical biochemical methodologies, directly impacting on the success of an aquaculture production.

Taking this into account, the strategy outlines in this work was to provide pathogen detection and identification by using molecular tools, such as PCR. Sequence data available in public databases was used to designed oligonucleotide primers that could specifically detect several bacteria species found in aquacultures in the Mediterranean, such as *Photobacterium damselae*, *Tenacibaculum maritimum, Aeromonas salmonicida, Aeromonas hydrophila,* and several subspecies of Vibrios, *namely V. harveyi, V. anguillarum, V. pelagius, V. vulnificus, V. splendidus, and V. fischeri*.

For a fastest detection of Vibrio sub-species, a multiplex PCR was used in which a combination of primers allowed for the specific detection of each vibrio in different samples. This reflects the usefulness of these protocols in the detection of the correct sub-specie while being time saving.

Altogether, our results show that a faster and more reliable detection of different pathogens in aquaculture can be performed for several bacteria and parasite species. Our results can also lead to the development of novel preventive and treatment measures, as well as mitigation tools, with the objective of control fish diseases, with subsequent reduction of economic losses for the aquaculture industry.

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S2AQUAcoLAB: A JOINT VENTURE THAT BRIDGES RESEARCH AND INDUSTRY TO FOSTER AQUACULTURE'S DEVELOPMENT

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The S2AQUAcoLAB is a private non-profit institution, founded in 2021, whose mission is to elevate aquaculture to a new level by playing an interface role between academia, research and industry. The coLAB performs research in production optimization, identification of health and welfare markers, climate change adaptations and development of new products for market diversification. Its members include 1 state laboratory (IPMA), 2 higher education institutions (UAlg and IPL), 1 R&D centre (CCMAR), 1 municipality (CMO), 1 producer's association (Formosa) and 9 private companies (ACUINOVA, P. Vale da Lama, SPAROS, NECTON, Atlantik Fish, P. Espargueira, Oceano Fresco, Riasearch, Docapesca). The S2AQUAcoLAB has 49 associated researchers with proven experience and countless scientific publications in the area and hired, until now, 15 highly qualified human resources which are involved in three ongoing research projects (REALM - HorizonEU; Blue Route – EEA Grants; Fosteam@south - PRR2021), international networks (Aquaexcel 3.0; Scar Fish; European Food Safety Authority) and applications in European (EEA Grants; Horizon Europe) and national projects (9 PRR projects). The S2AQUAcoLAB and its network of partners have the infrastructure, equipment and means to pursue scientific research of excellence whilst is exceptionally well-positioned to establish the link between scientific outputs and stakeholders.

The main goal of the S2AQUAcoLAB is to carry out R&D activities with a view to innovation for sustainable and intelligent aquaculture It aims to play an active role in the transfer of knowledge and technology, provide services that increase food safety, and diversify aquaculture products.

Acknowledgments: Project ALG-05-3559-FSE-000021.

SEASONAL VARIATIONS IN CHEMICAL AND BIOLOGICAL DIVERSITY IN BROWN MACROALGA *Halopteris scoparia*

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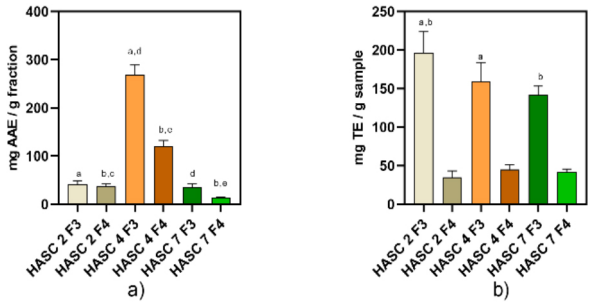
Brown macroalgae or seaweeds have recently been in the focus of many researches due to their composition and the accumulation of specific metabolites with great antioxidant potential. They represent a valuable source of bioactive compounds: polyphenols, pigments, peptides and polysaccharides that are associated with several health benefits and biological activities. Food, cosmetics and pharmaceutical industries are constantly working to find natural, non-toxic compounds that can be used in their products.

In this study, we evaluated the antioxidant activity of two (methanolic F3 and dichlormethane F4) different fractions from macroalga *Halopteris scoparia* in order to determine differences between harvesting season. Total protein content, chlorophyll and carotenoid content were determined. Also, several methods were used for evaluation of antioxidant activity. The semipurified fractions were also tested for antimicrobial activity using broth microdilution method for quantitative assessment and the toxicity of samples was determined using the zebrafish embryotoxicity test up to 96 h of embryonal development in order to reveal the safety of their usage and further implementation. Pearson's correlation coefficient was used to correlate obtained results for antioxidant activity.

All methanolic fractions showed higher antioxidant activity than the dichloromethane fractions of same season by implementing 3 different antioxidants assays, namely reduction of the radical cation (ABTS), the 2,2-diphenyl-1-picryl-hydrazyl (DPPH) and the oxygen radical absorbance capacity (ORAC) assays. April sample of *H. scoparia* (HASC) methanolic fraction (F3) showed the highest activity (268.6±20.6 mg AAE/g fraction) followed by HASC F4 April and HASC F3 February samples by DPPH method (Fig.1a). By using ABTS assay, the highest activity was observed for HASC F3 February followed by F3 April and F3 July fractions (Fig.1b). The highest protein value was observed for *Halopteris scoparia* April sample (20.5%) followed by HASC July (19.7%) and February (14.3%) samples. Chloropyll a and its derivate pheophytin a are dominant pigments in all three samples and the highest pigment content (chloropyll a and b, pheophytine a and b and carotenoids) was observed for HASC February sample.

Obtained results indicate that harvesting season has a crutial role in obtaining samples with best properties that have a potential to be used as a source of natural antioxidants in food, pharmaceutical and cosmetic industry.



TRANSCRIPTOME ANALYSIS AND DIFFERENTIAL EXPRESSION OF REPRODUCTIVE TRACT TISSUES OF MALE RIVER PRAWN *Macrobrachium americanum*

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The river prawn, *Macrobrachium americanum (M. americanum)*, is a large amphidromous species distributed along the Pacific coast of America. This prawn has commercial value due to its size and taste, making it a good option for aquaculture production. Sustainable prawn production requires a constant supply of high-quality hatchery and laboratory-reared larvae. To raise prawn larvae, the quality of adult reproduction is critical. The basic concepts of *M. americanum* reproduction have begun to be unraveled, but our knowledge of the mechanism that regulates reproduction at the molecular level is limited. To address this issue, we performed transcriptome assembly and differential expression analysis among testes, vas deferens, and terminal ampulla of *M. americanum.*

Adult males of *M. americanum* were captured in an estuary and a river located in Tomatlán, Jalisco and Puerto Vallarta, Jalisco, respectively. Total RNA extraction was performed using the TRIsure reagent following the manufacturer´s guidelines with modifications. Approximately one µg of high-quality RNA was used to prepare cDNA libraries, using the Zymo-Seq RiboFree™ Total RNA Library kit. The libraries were sequenced on the HiSeq 2500 platform (2 x 150 bp) by genomic service provider Novogene. The sequences were evaluated to determine their quality with FastQC software, and the adapters were removed using the SeqPrep software to obtain clean readings. A combined transcriptome with the three tissues was constructed using Trinity software. Sequences were compared against Swissprot and nonredundant Macrobrachium sp. databases and Gene Ontology (GO) databases using BLASTx and Blast2GO. Differential expression analysis among tissues was executed using EdgeR

The transcriptome assembly generated 1,059,447 unigenes, of which 7,222 genes had significant hits (e-value < $1x10^{-5}$) when compared against the Swissprot database. Based on Swissprot annotation, a total of 6,271 unigenes were annotated in GO, which were classified into three functional categories: cellular components (6,741), molecular functions (8,625), and biological process (40,628). About 20% of unigenes annotated in GO were involved in a reproductive process or reproduction within the category of biological processes. Based on Gene Ontology classification and comparison with data from other publications, about 70 genes were related to sex determination, testis development, spermatogenesis, spermiogenesis, fertilization, maturation of testicular cells, neuropeptides, hormones, hormone receptors, and/or embryogenesis. Differential expression analysis showed 179 transcripts differentially expressed (FDR < 0.05, Log2FC >1) among the tissues.

These results provide new molecular information about *M. americanum* reproduction, representing a reference point for further genetic studies of this species.

HYPER INTENSIVE SHRIMP FARM IN TAMAULIPAS, MÉXICO

Nowadays, there is a great business opportunity in the shrimp aquaculture in Mexico, due to the important growth of its demand y and prices both in Mexico and the USA. This initiative comes from a team of experts in shrimp aquaculture, with ongoing companies that have mastered the hyper intensive technology with BIOFLOC (BT).

The team that leads of this business consist of four experts in aquaculture with international prestige that add up to one hundred years of experience in the sector and one financial specialist with two decades of management experience.

The team of experts have used the hyper intensive technology for more than 7 years, obtaining a production of forty-two metric tons per hectare per year compared to less than 4 metric tons that results with the use of traditional semi-intensive technology.

The business proposal for investors consists in the sale of preferred shares. The minimum investment is USD$70,000 and all the private placements of preferred shares must be in multiples of this amount. The Internal Rate of Return (IRR) for the investor for an investment period of 10 years is 35% and the Payback is less than 5 years.

POVERTY IMPACTS OF TILAPIA AQUACULTURE AT SMALL SCALE IN RURAL HOUSEHOLDS IN SOUTH PACIFIC STATES IN MEXICO

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Tilapia aquaculture socioeconomic benefits in Mexico have been clear since the introduction of the species in the country in the 1960´s. In rural territories and their economies, they contribute at household level impacting both its poverty conditions and the nutrition of the house members. Results are shown from an ongoing study in the States of Oaxaca, Guerrero, and Chiapas, which have the highest levels of poverty in Mexico. Measured as multidimensional poverty, there are fewer tilapia farmers in extreme or moderate poverty, compared with the general population values.

These results reinforce the relevance of public policy developed at territorial level, differentiated based on particular characteristics of the economic agents in rural areas, its relationships and externalities like market and climate change vulnerability. Prosperous and inclusive rural societies has been identified as one of the three priorities for Latin American and Caribbean countries, during the recently finished Regional Conference of FAO. In the necessary agrosystems transformation, there is no doubt that aquaculture consolidation for AREL and SMEs is critical, particularly tilapia aquaculture carried out in all the production systems found in these territories.

INCOMEDIVERSIFICATIONINRURALHOUSEHOLDSDEPENDENTONAQUACULTURE IN THE SOUTH PACIFIC OF MEXICO, AS A RESILIENCE STRATEGY

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Rural non-farm employment has been the subject of study in the economic theory for several decades. For the case of tilapia farms at AREL and SMEs scale in the States of Oaxaca, Guerrero, and Chiapas in Mexico, we present results of an ongoing project, related to the rural economies and the relevance of analyzing production activities per territory.

The results and further discussion call the attention on how relevant the tilapia project is for climate change, inflation and other external shocks´ resilience strategies. For national, regional or local policies focused on Food Security and Food Sovereignty, identifying these elements is critical. In the Agrifood systems transformation, ways to support these farmers and their communities must be reinforced.

EVALUATION OF A FLOCPONIC SYSTEM FOR THE CULTURE OF TILAPIA *Oreochromis niloticus*, JALAPEÑO PEPPER AND MINI BELL PEPPER *Capsicum annum*

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The term FLOCPONIC is used to name an integrated system of aquaponics with bioflocs and has been successfully proven for the production of diverse animal and vegetal species, showing to be a sustainable and economically feasible practice. In the present study, a preliminary assay was conducted to evaluate the feasibility of a flocponic system for the production of tilapia (*Oreochromis niloticus*), jalapeño pepper and mini bell pepper (*Capsicum annum*) with promising results at least for tilapia and mini bell pepper.

The study was conducted during eight weeks (mid-September to mid-November) in the facilities of the Departamento de Agricultura y Ganadería, Universidad de Sonora at Hermosillo, Sonora, México. Tilapia was farmed in tanks (500 L), with and without bioflocs incorporated. The bioflocs were produced in a transparent tank (500 L) inoculated with the microalgae *Scenedesmus oblicuos* and wheat brand. The effluents were passed to PVC tubes for the hydroponic culture of jalapeño pepper (*Capsicum annum*). The final effluent was used to a fertigation culture of mini bell pepper (*Capsicum annum*). The production response of tilapia in terms of weight gain, survival, biomass and FCR, was little better in tanks with bioflocs (Table 1). The jalapeño pepper had not a good performance in the hydroponics system in terms of plan height, survival, number of fruits per plant and days for flowering (Table 2). Contrarily, mini bell pepper showed a better performance (Table 3).

We concluded that our experimental flocponics system could be more efficient, but some adjustments need to be done.

Table 1. Production parameters of tilapia

	Growth (g)	Survival (%)	Biomass (g/tank)	FCR
W	120 ± 26a	83 ± 2^a	2969 ± 182^b	1.1^a
WO	90 ± 19a	80 ± 2^a	2160 ± 103^a	1.4^b

W = with bioflocs; WO = without bioflocs

Table 2. Production parameters jalapeño

	Height (cm)	Survival (%)	Fruits per plant	Days x flowering
J	26.4 ± 2a	63 ± 5^a	1.6 ± 0.3^a	16 ± 1^a
MB	27.1 ± 2a	66 ± 7^a	1.7 ± 0.3^a	16 ± 1^b

J = jalapeño; MB = mini bell

Table 3. Production parameters of mini bell

	Height (cm)	Survival (%)	Fruits per plant	Days x flowering
J	39 ± 4a	93 ± 3^a	12 ± 2^a	12 ± 1^a
MB	40 ± 5a	100 ± 0b	13 ± 3^a	12 ± 1^b

THE EFFECT OF REARING TEMPERATURE ON MEAGRE (*Argyrosomus regius*) GROWTH PERFORMANCE DURING LARVAL AND EARLY JUVENILE STAGES

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Meagre (*Argyrosomus regius*) is considered an interesting species for the diversification of Mediterranean aquaculture and, mainly due, to its fast growing capacity and to its processing potential, it is known by many as the "salmon of the south". Additionally, meagre is a species easily adapted to captivity and well-known by the consumers, presenting a high commercial value, making it an interesting species for aquaculture production.

At the Aquaculture Research Station of Olhão (EPPO), an aquaculture facility from the Portuguese Institute for the Ocean and Atmosphere (IPMA) dedicated to perform pre-industrial studies, several trials have been conducted to address questions related to meagre rearing and along the years one of the most common question raised was which is the best rearing temperature for meagre. Although the normal spawning temperature rounds the 20ºC, the optimal rearing temperature, to enhance larvae and juvenile growth, seems to be situated a couple degrees above, which makes meagre a suitable species to grow in recirculation systems (RAS), at higher and controlled temperatures. This issue is particularly important in the context of the foundation of the S2AQUAcoLAB (in 2021), a collaborative laboratory, that brings together research and industry partners, with the main objective of fostering the development of the aquaculture sector in Portugal and which has, among several other areas of research, interest in the implementation and optimization of recirculation culture systems.

In this work we present the results of several trials performed to determine the better growing temperature for meagre larvae and juveniles, which for larvae is around 22 and for early juveniles is located between 24 and 28ºC. Fish presented a higher size dispersion when grown at higher temperatures, but the incidence of skeletal malformations decreased when compared with the lower temperatures. In order to have more detailed information on the physiological and biochemical performance of the fish, methodologies such as histology to determine the onset of main organs and gene expression to evaluate growth-related genes were performed.

This study had the support of the project DIVERSIAQUA II (Mar2020-P02M01-0656P)

LIMNOLOGICAL STUDY OF EARTHPONDS OF TILAPIA CULTURE

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In the present study, some limnological factors of earthponds of monosex and mixed tilapia culture were investigated during the breeding season of 2016. Comparison of the length and weight of the cultured tilapia and FCR between two treatments in the earthponds indicate a more favorable growth performance in monosex tilapias, as expected.

Tilapias are the world's second largest farmed fish and their production in the world has sharply increased, especially in recent decades. Nile tilapia, *Oreochromis niloticus* production consisted more than 70% of total farmed tilapia in the last decade. Tilapia farming is now done in more than 135 countries and its' global production has exeed than 5.6 million tons (Fitzsimmons, 2016; FAO, 2014).

Nile tilapia *O. niloticus* samples were entered to Iran for research purposes at December 2008 for the first time. Studies about the feasibility of introduction of tilapia to inland aquaculture in Iran were begun in National Research Center of Saline Water Aquatics in Yazd province at the center of Iran (Rajabipour, 2013). Different aspects of reproduction, culture, feeding, monosex production, processing are studied (Bitaraf, 2012; Mashaii et al., 2016; Mohammadi et al., 2014; Morady et al., 2012; Rajabipour, 2013 & 2016; Sarsangi et al., 2012).

In the present study, some limnological factors of earthponds of monosex and mixed tilapia culture were investigated during the breeding season of 2016. The minimum and maximum of water temperature ranges were 16.6-27.5°C and 20.5-31°C, air temperature 2-27°C and 30-48°C, dissolved oxygen in the morning and afternoon 2.11-10.19mg/l and 7.06-14.64mg/l, pH in the morning and afternoon 8.97-10.9 and 9.04-13, the depth of transparency 40-105cm, salinity 7.94-8.82ppt and ammonium density less than 0.05mg/l to 0.4mg/l. The most abundant phytoplankton and zooplankton were Chlorophytes, diatoms and rotifers, and dominant macrobenthos were chironomids. There was significant positive correlation between pH value with ammonium density and body length and weight, water temperature with total length, and negative correlation between dissolved oxygen and temperature and pH ($p<0.05$). Tilapia culture in earthponds has advantages especially for using water biomass by fish and lower costs of the structures. Comparison of the length and weight of the cultured tilapia and FCR between two treatments in the earthponds indicate a more favorable growth performance in monosex tilapias, as expected.

TILAPIA CULTURE IN TNAK INDOOR SYSTEM

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Tilapia culture in earthponds is a traditional method of breeding this fish, but water consumption is high in this ponds. So, considering limited water resources in many areas prone to tilapia culture, it is preferred to planning culture in indoor ponds. In the present study, brackish water quality factors of indoor tilapia culture, also growth performance of the cultured tilapia were investigated in indoor tanks.

Tilapia studies in Iran were begun since 2008 and different aspects of tilapia aquaculture are researched. In the recent years, some central areas of the country are permitted to culture tilapia. In the present study, brackish water quality factors of indoor tilapia culture, also growth performance of the cultured tilapia were investigated in indoor tanks. Monosexed red tilapia *Oreochromis* sp. were stocked. Water temperature, dissolved oxygen, pH, salinity and ammonium concentration were measured. Pearson 2-tailed correlation between the amounts of factors including temperature, salinity, dissolved oxygen, pH and ammonium with length and weight of fish in ponds showed a direct significant correlation between the length and weight of the fish body with each other, the amount of salinity against pH, salinity against total length and body weight. Also, there was a reversed significant correlation between the amount of dissolved oxygen against water temperature, pH and the total length of the fish. Tilapia culture in earthponds is a traditional method of breeding this fish, but water consumption is high in this ponds. So, considering limited water resources in many areas prone to tilapia culture, it is preferred to planning culture in indoor ponds. Indoor tilapia culture systems such as tank culture, biofloc and aquaponic systems are more suitable for tilapia production, especially in areas with limited water resources and high evaporation. These methods makes possibility of producing tilapia with higher density and low water consumption with environmental considerations.

CAMBIOS MORFOLÓGICOS EN EL DESARROLLO EMBRIONARIO DEL LANGOSTINO DE RIO *Macrobrachium tenellum*

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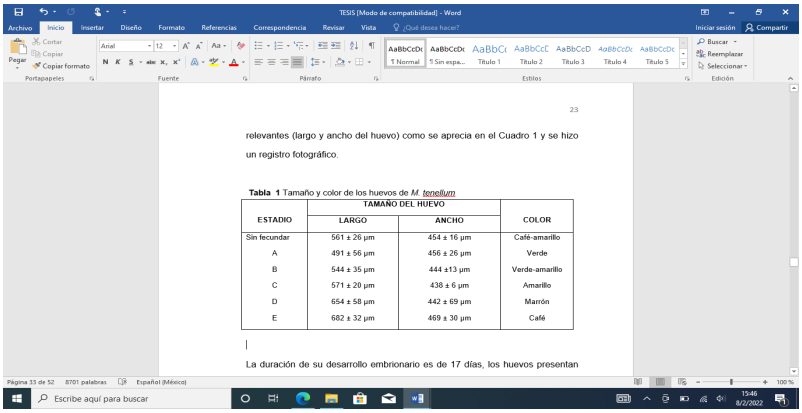
El langostino de río *Macrobrachium tenellum* es una especie que habita en las costas del pacífico mexicano, actualmente existen reportes relacionados a su ecología, técnicas de cultivo, y reproducción, sin embargo, a pesar de esta información no se ha logrado completar con éxito el principio de su ciclo de vida. El presente trabajo tuvo como objetivo caracterizar los cambios morfológicos que presentan los huevos de *Macrobrachium tenellum* durante su desarrollo embrionario.

Para este estudio se colectaron organismos de *M. tenellum* en muestreos periódicos en el Rio Colotepec situado en el municipio de Santa Maria Colotepec en la Costa de Oaxaca, los organismos se mantuvieron en condiciones de cautiverio en el Centro Interdisciplinario de Investigación para el Desarrollo Integral Regional Unidad Oaxaca (CIIDIR). La identificación de la presencia de la masa ovigera en las hembras se hizo mediante la observación e inspección diaria del organismo, las hembras ovadas fueron transportadas al Laboratorio de Bioquímica de proteínas y Glicoproteínas de la Coordinación de Posgrado e investigación, Facultad de Medicina y Cirugía de la UABJO. Para la toma de muestra se hizo un lavando del área abdominal de las hembras con agua destilada, se retiraron loshuevos y fueron fijados con Paraformaldehído al 4%, posteriormente fueron observados en el microscopio invertido (Leica con 18 cámara CCD acoplado) a 10x, en donde se anotaron las características relevantes que se presentaba en los huevos en cada estadio de su desarrollo embrionario, realizando un registro fotográfico.



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relevantes (largo y ancho del huevo) como se aprecia en el Cuadro 1 y se hizo un registro fotográfico.

Tabla 1 Tamaño y color de los huevos de *M. tenellum*

ESTADIO	TAMAÑO DEL HUEVO		COLOR
	LARGO	ANCHO	
Sin fecundar	561 ± 26 µm	454 ± 16 µm	Café-amarillo
A	491 ± 56 µm	456 ± 26 µm	Verde
B	544 ± 35 µm	444 ±13 µm	Verde-amarillo
C	571 ± 20 µm	438 ± 6 µm	Amarillo
D	654 ± 58 µm	442 ± 69 µm	Marrón
E	682 ± 32 µm	469 ± 30 µm	Café

La duración de su desarrollo embrionario es de 17 días, los huevos presentan

Se describieron 6 estadios embrionarios en base a la coloración, tamaño del huevo (largo y ancho) y características morfológicas del embrión observadas mediante microscopia; el estadio A se puede observar la forma elíptica de los huevecillos con vitelo distribuido, en estadio B se aprecia el cambio de coloración durante el crecimiento del embrión; así como las primeras estructuras de este. En estadio C se observa la segmentación del abdomen en estadio D se observa la presencia de ojos y telson y en el estadio E se observa con claridad la presencia de ojos pigmentados, pleòpodos y telson. Se determinó que la duración de su desarrollo embrionario es de 17 días, los huevos presentan una forma esférica con un diámetro de 618 ± 73 μm a 451 ± 42 μm como se muestra en la tabla 1. En conclusión, se establecieron 6 estadios del desarrollo embrionario basados a la coloración del huevo, el diámetro del embrión y de la mancha ocular cuyos parámetros varían dependiendo de las necesidades del embrión por lo que estos datos pueden ser utilizados como parámetros de crecimiento de este langostino.

MANEJO REPRODUCTIVO DE ROBALOS Y PARGOS EN LA ESTACIÓN DE ACUICULTURA MARINA

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La Estación de Acuicultura Marina (EAM) está ubicada en la localidad de Jalapita, Centla, Tabasco y forma parte de la División Académica de Ciencias Biológicas (DACBiol) de la Universidad Juárez Autónoma de Tabasco (UJAT). En la EAM, se encuentra el Laboratorio de producción de crías de peces marinos. En La EAM se desarrollan investigaciones para la producción de semilla de las especies de peces marinos nativos de la región, donde se incluyen tres de las especies del género *Centropomus* (*C. Undecimalis, C. Poeyi* y *C. Parallelus*) que son consideradas las más importantes para las comunidades de pescadores que explotan el recurso en los hábitats dulceacuícola y marino, siendo el soporte económico de muchas comunidades durante el año. Este laboratorio fortalece y complementa la infraestructura del Laboratorio de Acuicultura Tropical UJAT al permitir la investigación sobre aspectos de la biología de estos organismos para la producción masiva de crías de peces marinos nativos. En el Laboratorio también se lleva a cabo la producción masiva de alimento vivo para el desarrollo de protocolos de alimentación de larvas de peces marinos. Además, se han incorporado especies como *Archosargus probatocephalus*, *Lujtajus cyanopterus* y *Lutjanus griseus* para en el futuro a largo plazo desarrollar sistemas de producción destinados a la repoblación de áreas sobreexplotadas y el desarrollo de la acuicultura, ambas actividades con su impacto social y de mitigación sobre la presión de captura de los recursos silvestres. Dentro de los resultados que se han obtenido en inducción a desove, destaca que para *C. undecimalis* se han obtenido en promedio 71.30 +/- 31.34 % de fertilización, 56.07 +/- 42.53 % de eclosión, para c. poeyi 53.89+/-40.16 % de fertilización y 55.33% de eclosión y en *C. parallelus* 92.35 +/- 5.11% de fertilización y 68.08 +/- 34.86 % de eclosión, Aunque hembras individuales han proporcionado eventos de 100 % de fertilización. En *L. griseus* la fertilización ha sido de 78 % y la eclosión de 1.4 %. En nuestras poblaciones de peces se ha obtenido desoves a partir de ≥350 μm de diámetro de ovocitos por lo que los implantes hormonales inducen a la maduración y desove efectivamente. El promedio de larvas producidas en *C. undecimalis* fue 1, 829 999.50 larvas, 1, 543 260.55 para *C. poeyi* y 9, 846.50 para *C. parallelus* y 10, 600.00 para *L. Lutjanus*.

ESTUDIO DE ASOCIACIÓN GENÓMICA PARA RESISTENCIA AL VIRUS DEL SÍNDROME DE MANCHA BLANCA EN CAMARÓN BLANCO DEL PACÍFICO

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El desarrollo de microarreglos con miles de Nucleótidos de Polimorfismo Simple (SNPs) para *Litopenaeus vannamei*, ha permitido profundizar en la búsqueda de variantes genéticas asociadas a mayor resistencia al virus del Síndrome de la mancha blanca (VSMB) y así evaluar la incorporación de selección asistida por marcadores (MAS) y de la selección genómica (SG) en los programas de selección. El objetivo de este trabajo fue identificar variantes genómicas asociadas con la resistencia al VSMB, utilizando un panel SNP de alta densidad (50K) en una población de *L. vannamei* en un desafío controlado.

Se usaron 1,685 camarones de 176 familias de *L. vannamei*, con edad promedio (D.E.) de 78.4 (3.7) días post-eclosión y un peso de 1.85 g (0.45 g), distribuidos en dos tinas de un sistema cerrado e inoculados vía *per os* con músculo macerado de camarón con una concentración de 10^6 partículas virales /g en dos tomas. De las 24 horas y hasta las 144 horas se recolectaron los camarones muertos cada hora (Tiempo de muerte = TM) y se consideraron vivos aquellos que se mantuvieron hastas las 144 horas (1=vivo, 0= muerto). El genotipado se realizó con el panel AquaArray HD (50 K) vannamei® (Neogen®). En el control de calidad se usó una tasa de llamado para SNPs de 80%, frecuencia de alelo mayor menor a 0.01 y equilibrio de Hardy-Weinberg con p-value < $1x\ 10^6$. Se conservaron 33,537 SNPs para el análisis. La estimación de la varianza genética explicada por los SNPs, se realizó con el programa GCTA, se consideraron como efectos fijos el peso y la tina de siembra. Se consideró un SNP con significancia genómica cuando su valor de p < *0.05/= número total de SNP utilizados*. Se utilizaron las secuencias nucleotídicas correspondientes a los SNPs significativos para la búsqueda de genes candidatos.

La media para TM y SB fueron de 110.6 (35.10) horas y 0.40 (0.01). En el cromosoma 1 se localizó un SNP significativo para TM y SB, mientras que otro SNP en el cromosoma 21 solo fue significativo para SB, en ninguno caso el aporte a la varianza genetica fue mayor al 1%. El SNP del cromosoma 1 esta relacionado con genes asociados con la respuesta inmune al VSMB. Lo anterior sugiere una arquitectura poligénica para SB y TM por lo que la SG sería la mejor forma de incorporar este tipo de información en el proceso de selección para la resistencia al VSMB en *L.vannamei*.

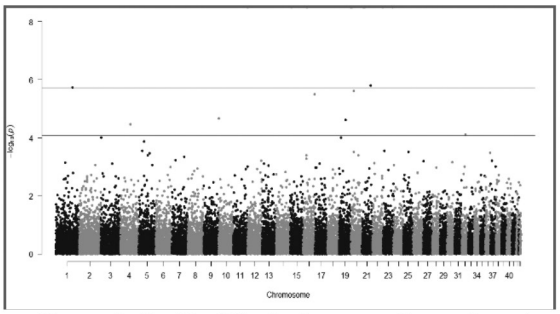

Figura 1. Grafico Manhattan para Supervivencia

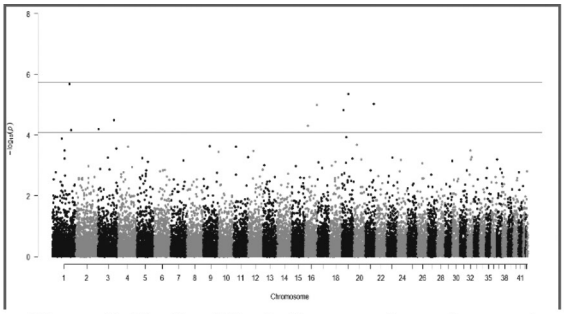

Figura 2. Grafico Manhattan para hora de muerte

EFECTIVIDAD DE PROFILÁCTICOS COMERCIALES PARA CONTROLAR *VIBRIOS* PATÓGENOS EN LARVICULTURA DE CAMARÓN EN ECUADOR

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Las patologías bacterianas en la producción larvaria de la Península de Santa Elena, han producido mortalidades superiores al 70%, teniendo un mayor impacto en la zona de Mar Bravo. Se ha calificado a las bacterias del género *Vibrio,* como las responsables de enfermedades tales como Síndrome de bolitas, Síndrome de zoea y Síndrome de la Mortalidad temprana (EMS), siendo los responsables de dichas patologías: *V. Harveyi, V. Vulnificus y V. Parahaemolitycus* respectivamente. Se detectó además, que las bacterias del tipo *Pseudomonas sp*. *Pseudomonas fluorescens* presentes en las larviculturas fueron igual responsables de altas mortalidades. Ante estas infecciones bacterianas, los antibióticos han sido utilizados mundialmente como terapeúticos y algunas veces como profilácticos contra estos patógenos, sin embargo, el uso de estos conlleva importantes desventajas, entre ellos presencia de residuos en productos de la acuicultura, desarrollo y propagación de resistencia entre patógenos, incluidos patógenos humanos,bajo este contexto se hace necesario implementar estrategias alternativas de control, tales como el uso de productos naturales como ácidos orgánicos, Aceites escenciales, Probióticos para el control de dichas bacterias.

El presente trabajo monitoreó diferentes laboratorios de la Peninsula de Santa Elena,
y seleccionó cepas bacterianas de larvas en diferentes estadios, donde se determinaron las dosificaciones de Concentración Mínima Inhibitoria (MIC) empleando productos comerciales,.De esta manera se buscaba determinar con exactitud la dosis bactericida ha ser empleada en la producción de larvas. Los principales productos incluidos en los protocolos de larvas de camarón, han sido los ácidos orgánicos, aceites esenciales y probióticos, los que en diferentes presentaciones y concentraciones han dado respuestas equivalentes superiores a los 2500 ppm, sin embargo se pudo evidenciar un efecto sinérgico cuando se mezclaban diferentes productos en menor dosificación, potencializando su acción para eliminar a las cepas bacterianas analizadas.

Se indica que algúnos de estos productos, no deben ser usados en simultáneo cuando se aplican probióticos, sin embargo, se pudo encontrar concentraciones que permitieron trabajar de manera sinérgica en una mezcla unica con todos los profilacticos mencionados.

Las bacterias fueron caracterizadas bioquimicamente y molecularmente, para tener una secuencia de estudios que se vienen realizando muy de la mano con los diferentes protocolos de bioseguridad y control en los diferentes laboratorios de larva, para asegurar corridas productivas exitosas.

Palabras clave:
Concentración mínima inhibitoria; bacterias patógenas, Larvas, *Litopenaeus vannamei*.

BREEEDING AND CULTURE OF MEXICAN FRESHWATER NATIVE ESPECIES: A GROWING OPPORTUNITY

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Mexico has a huge diversity of freshwater fishes, with at least 160 species, including 50 genders and 30 families, mainly located in the south eastern part of the country. Of these, just a few of them have been studied and developed for aquaculture, however, not even this very few have been fully utilised for regional aquaculture advance, despite the fact that they could be grown using the already existing Tilapia facilities, hence, could be used as a mean to increase, diversify and improve production and benefits for many fish farmers.

Aquaculture research of native species with potential in aquaculture in Mexico began in the earlys 80´s, involving many institutions and personnel, this led to the organization of national and international Seminars in 1988, 1994 and 2006. Results showed that there was aquaculture technology for some of the big cichlids and the gar, as well as the many ecological and socioeconomical benefits of their exploitation. Market was also a matter, as prices were always higher than tilapia and demand was often unsatisfied, for instance.

Today, all the reasons that were argued as beneficial for native fish culture not only remain but have been increased, e.g., fisheries supply is shorter, prices continue to rise as well as the consumer demand. In addition, aquaculture as a sector has also grown in the region, so we have more and better feeds, technicians, services and growing facilities.

This paper presents regional examples were the culture of Castarrica *Cichlasoma urophthalmus*, Paleta *Vieja sinspyla* and Gar *Atractosteus tropicus* as well as fisheries restocking programs including this species and tilapia can be a sustainable and very profitable aquaculture alternative for the region. Special emphasis is given to point out some of the many technological, marketing and economical strengths of the culture of this native fishes.

EFFECTIVENESS OF AN RECIRCULATION AQUACULTURE SYSTEM (RAS) IN THE QUARANTINE AND MATURATION OF WHITE LEG SHRIMP BROODSTOCK *Litopenaeus vannamei* Boone, 1931

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A specially designed recirculation aquaculture system (RAS) was used to maintain a batch of Litopenaeus vannamei white leg shrimp broodstock in quarantine, maturation, reproduction and spawning. The objective was to determine the effectiveness of the RAS by monitoring the physical and chemical parameters of seawater quality and recording the biological production parameters.

The supply of shrimp broodstock from a genetic nucleus of lines tolerant to specific viral diseases and with improvements in growth is a necessary biosecurity measure to respond to the requirements of shrimp producers regarding the supply of healthy shrimp postlarvae. Their reproductive performance was evaluated in an operation period of 144 days, since the import of a batch of 450 reproducers from a commercial genetic nucleus of 2 lines of shrimp reproducers with an average weight of 30 grams: SPARTAN -tolerance to diseases- (234 organisms: 142 females and 92 males) and FURIOS -improved growth- (216 organisms: 144 females and 72 males).

The variables were the physicochemical parameters of seawater: temperature, hydrogen potential, ammonia nitrogen and alkalinity. The biological parameters were recorded: Survival, percentage of copulations, fertilization and hatching. The recirculation percentage was 95% with 5% replacement. The physical parameters described a stable behavior and within the requirements of the species; without presenting a significant difference ($p > 0.05$) between the RAS and the production tanks, but with respect to seawater. The recorded measurements of the chemical parameters did show a significant difference ($p < 0.05$) between them, with better results in the RAS designed for this purpose. The data obtained from the biological parameters of production showed how the SPARTANs are significantly better ($p < 0.05$) in their performance compared to the FURIOS (Fig. 1).

It was possible to establish the effectiveness of the system to maintain the quality of the water during the period of quarantine, maturation, reproduction and spawning of shrimp, at commercially acceptable levels; with an adapted equipment, reducing the investment and in a vinculation Institute:Company for the formation of human capital.

This level of reduction allows the supply of nauplii destined for the biosafety production of shrimp postlarvae, without the use of large quantities of seawater; thereby reducing the coastal environmental impact, preventing the occurrence of the introduction-dispersal of exotic diseases to the environment and the probability of the entry of vectors from the environment to the production facilities.

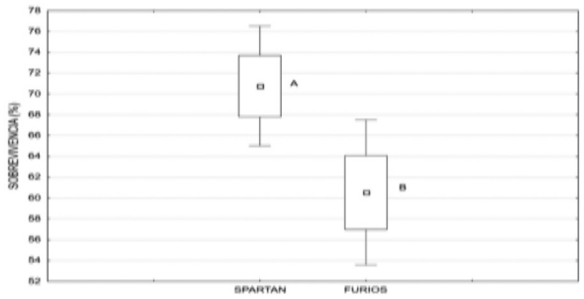

Figure 1. Survival by genetic line. Source: Own elaboration, 2020.

DETERMINATION OF THE SURVIVAL AND GROWTH OF OYSTER SEED *Crassostrea virginica* IN THE SOTO LA MARINA RIVER

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The American oyster *Crassostrea virginica* is a mollusk that is naturally distributed along the Atlantic coast, from Canada to the state of Campeche, Mexico; it plays an important biological role and has excellent nutritional properties for human consumption. It has been pointed out that excessive extraction and anthropogenic activities have decreased mollusk production in different regions of the world and aquaculture represents an alternative to reduce these impacts. The present study determined the survival and growth rates of oyster seed in floating bags at 6 sites of the Soto La Marina river (SLM), associated with environmental parameters.

Seeds of *C. virginica* of 2 cm in length (Lt) collected in the SLM river were sown in floating plastic mesh bags (8 x 16 mm mesh size, 48 cm wide x 53 cm long), at a density of 30 seeds per bag, placing 3 replicates in 6 stations, from the beach to the most distant area upriver where the presence of the species was observed. Every 3 weeks, the bags were cleaned; the length, height, width (cm), and individual weight (g) of oysters were measured for 12 weeks. At the end of the study, the average survival and growth of the organisms were determined; turbidity, salinity (‰) (Fig 1), oxygen (mg/l), temperature (°C), and pH were measured at each station.

The highest survival was found at the Cortadura station with 85%; Vista H had 78%, La Pesca and Contadero 72%, and the lowest survival occurred in La Playa and Carricitos with 51%. The growth in total Lt (Mean ± SD) was higher at the Vista H station with 2.2 ± 0.3 cm, with an average salinity of 16 ‰; the lowest growth was observed at La Playa with 0.6 ± 0.3, and a salinity of 35 ‰ (Fig 2).

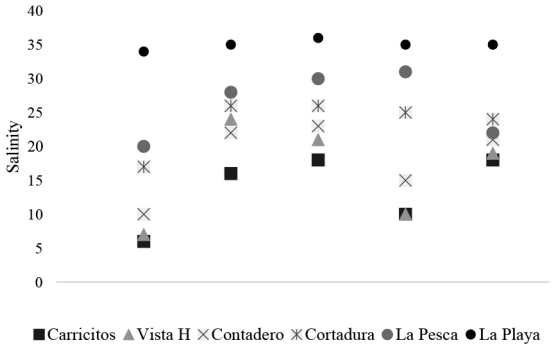

Fig. 1. Monthly variation of salinity (‰) in the water of the SLM river in each sampling station.

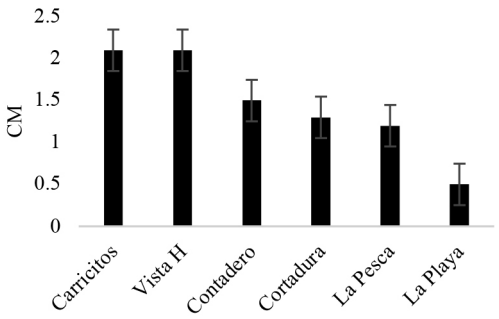

Fig. 2. Growth in Lt (cm) (mean ± SD) of *C. virginica* seed in different sites during the experiment.

ANÁLISIS BIOECONÓMICO PARA LA SELECCIÓN DE TRES ESPECIES DE ROBALOS DEL GÉNERO *Centropomus* CON POTENCIAL ACUÍCOLA PARA EL DESARROLLO DE LA MARICULTURA EN EL PACÍFICO MEXICANO

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Palabras clave: C.viridis, C.nigrescens, C.medius, Análisis bioeconómico, VPN, TIR.

Introducción. La bioeconomía constituye la primera oportunidad de implementar una economía global realmente sostenible basada en recursos biológicos, que, gracias a las biotecnologías se convierten en renovables.

Uno de los grandes desafíos en la acuicultura, es la selección de especies de peces marinos candidatas para esta actividad. El robalo representa uno de los recursos más importantes de la pesca ribereña multiespecífica en los sistemas lagunares. Sin embargo, su producción no satisface la demanda del mercado nacional. La solución es implementar sistemas de producción para determinar el desempeño de la especie y con ello, evaluar la rentabilidad de la misma.

Desde los años ochenta la acuicultura se ha convertido en una actividad de rápido crecimiento a nivel mundial. Esta actividad contribuye al rápido aumento de la oferta de pescado, pretendiendo compensar el agotamiento de las poblaciones de peces naturales del mundo. Sin embargo, no ha podido contribuir del todo, ya que no ha podido frenar el colapso que enfrenta más del 50% de las pesquerías existentes en el Pacifico mexicano. (Beltrán Meza, 2017).

México se proyecta a mediano plazo, como potencia mundial en la actividad por acuicultura, debido a que en los últimos años la tasa media de crecimiento ha sido del 15% anual, alcanzando en 2016, cifras históricas de 337 mil toneladas de producción por acuicultura, representando esto el 22% en la actividad pesquera del país.

Sin embargo, a pesar de contar con las condiciones necesarias y el potencial suficiente para hacer de esta actividad una fortaleza, su crecimiento ha sido moderado y no ha logrado adquirir la suficiente relevancia como en los países industrializados (INAES, 2018).

Es por ello que el análisis bioeconómico en la acuicultura surge como respuesta a la necesidad de integrar factores económicos, biológicos, técnicos y ambientales para mejorar la eficiencia y toma de decisiones a nivel empresarial.

Debido a la complejidad de los sistemas de producción acuícola y los numerosos desafíos impuestos por el rápido crecimiento de esta industria, son necesarios implementar y desarrollar modelos integrales, con el objetivo de proporcionar información tecnológica y alternativas a los productores.

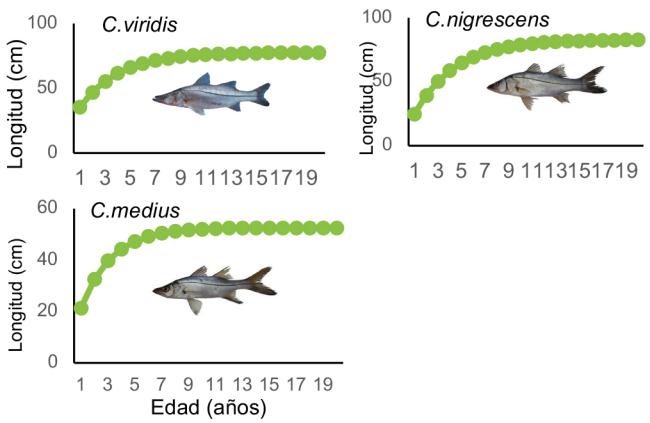

Figura. 1 Curva de Crecimiento Von Bertalanffy de tres especies de robalos (*C.viridis, C.nigrescens, C.medius*).

(Continued on next page)

El desarrollo de sistemas de producción acuícola es una solución, para suministrar alimento, contribuir a la economía en la generación de empleo e ingresos. Para ello, se utilizan modelos matemáticos, los cuales, son técnicas usadas en el desempeño de sistemas de producción biológica sujetos a restricciones económicas, biológicas y técnicos en los mercados.

Objetivo general. Evaluar el potencial bioeconómico de las tres especies de robalo en el Pacífico mexicano (*C.nigrescens, C.viridis, C.medius*) mediante análisis pesqueros y modelos financieros.

Estrategia metodológica. El área de muestreo se localiza en el Norte del Estado de Sinaloa.
Para el análisis biológico, se realizaron muestreos biológicos de 30 peces capturados por mes con chinchorro por la flota artesanal.
Cada organismo se midió en longitud y peso. La identificación de las especies se realizó siguiendo las claves convencionales de la FAO considerando los aspectos morfológicos y de color.

Con la información de longitudes de las tres especies, éstas se integraron al programa FISAT II para obtener los parámetros de crecimiento y ser ajustados al modelo de Von Bertalanffy (K, $L\infty$ y t_0) (Fig.1); pudiendo determinar con ello, cual especie presenta los mayores crecimientos y cuál es la más factible de cultivar.

Con los parámetros obtenidos, se realizó una corrida técnica en hojas de cálculo, la cual, se llevó a cabo comparando las tres especies con un cultivo de robalo previamente diseñado.

Para la optimización de las ganancias totales y por semana de un posible cultivo potencial para las tres especies de robalos, se aplicaron tres tratamientos distintos (ración 3,5 y 7%). Posteriormente se utilizaron modelos financieros (VPN y TIR) para determinar la rentabilidad.

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AFTER COVID COMMERCIAL PRODUCTION STATUS OF *Seriola rivoliana* BY KING KAMPACHI

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In the Americas there is an incredible emerging mariculture industry, which comes out due to the fact that there are different marine species for which it has been necessary to develop knowledge for each one of them and learn the specific management or "know how" for each species in different specific sites with a high range of temperatures and several environmental interactions.

When I talk about an "incredible industry" I am talking about an industry with fertile soil for all enthusiasts who love aquaculture, particularly marine fish and that we have had the opportunity to try new farming methods, develop new technical protocols and challenge the premises that are held in the Salmon, Sea Bream and Sea Bass industries that are available in the Mediterranean to compete in the global market with species of high nutritional and economic value.

In the King Kampachi hatchery in La Paz, B.C.S., 3 years after launching, we have produced under controlled conditions more than one million juveniles that have been stocked into offshore cages. To do this, we had to solve several bottleneck issues such as:

- Broodstock management to obtain high quality year-round without hormones;
- Egg incubation crashes;
- Post-hatch sinking larvae;
- First feeding mortalities;
- High quality rotifers in massive production;
- Improvement in deformity rates;
- High intake water temperature management (up to 30 Celsius);
- Specific pathogens with high temperature environments; and
- Fish welfare principles applied to *Seriola rivoliana*.

The marine aquaculture industry (mariculture) in Mexico has been an individual effort for all of the companies involved, the lack of peripherical services related to the offshore operations, and hatchery supplies has been a bottle neck that got worse during and after COVID shut down where the fragility of the chain supply was evidenced. Besides the lost of costumers during this period, research and development was set to stand by, we had to adapt to the new reality, looking for a way to get directly to the consumer and being able to be more efficient in the production operations with what we have with the minimum technical staff available.

One of our greatest milestones was the use of a RAS system for larvae culture in the hot weather seasons and has been now the way we have outcome covid shutdown with juvenile production before transferring fish to offshore.

After covid hatchery improvement opportunities.

During the 2020 covid lockdown, we had to leave on stand by all the non-essential research activities, when the production status was at the lowest point. Now we are starting to assess priorities with the following improvement points:

- Qrill fish meal and oil to enhance immune system in juveniles before transferring to offshore.
- AI technology development for small juvenile sorting.
- Genetic breeding program for broodstock management improvement.
- BAP certifications process.
- Whole hatchery scale up phase.

ASSESSMENT OF ANTIBIOTICS RESIDUES IN AFRICAN CATFISH *Clarias gariepinus* FROM AQUAFARMS IN IBADAN, NIGERIA

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Inadvertent consumption of drug residues in fish and fish products is a major factor contributing to incidence of drug resistant strains of micro-organisms in consumers. This study was designed to investigate the prevalence and concentrations of frequently used antibiotics (Enrofloxacin, Gentamicin and Tetracycline) by farmers in catfish grow-out production in Ibadan, a major hub of catfish cultivation in Nigeria.

Simple random sampling technique was used to select three (3) Local Government Areas (LGAs) namely, Iddo, Ibadan Northwest and Akinyele. Proportionate sampling technique was employed to select forty (40) fish farms from where eighty (80) samples of marketable African Catfish *C. gariepinus* (475±7.58g) were collected for laboratory analysis. Antibiotic residues in the fish muscles were determined by high performance liquid chromatography (HPLC).

All the fish samples (100%) contained residues of Gentamicin and Tetracycline while 76% of the samples had residues of Enrofloxacin. Further, the average concentrations of Enrofloxacin, Gentamicin and Tetracycline in the fish muscles were 17.27±1.72 µg/kg, 40.52±2.95 µg/kg and 198.40±9.3 µg/kg, respectively. None of the catfish samples had concentrations of Enrofloxacin and Gentamicin above the allowable limits (100 µg/kg), whereas there was preponderance of Tetracycline concentrations, and above allowable limit (200 µg/kg).

The high prevalence and concentrations of the considered antibiotic residues in the sampled fish give room for food safety concerns because of the long-term public health effects. It is imperative that the use of antimicrobial agents in fish production be regulated, and that programs for responsible aquaculture be developed.

Table 1: Concentrations of Tetracycline in fish samples.

LGAs	Concentrations	
	≤ 200 µg/kg	>200 µg/kg
Iddo	10 (35.71)	18 (64.29)
Ibadan Northwest	9 (37.5)	15 (62.5)
Akinyele	7 (25.0)	21 (75.0)

Frequency (percentage)

DETERMINATION OF A PREDICTIVE GROWTH MODEL FOR CULTIVATED AFRICAN CATFISH *Clarias gariepinus* (Burchell, 1882)

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In recent years, aquaculture of the African catfish *Clarias gariepinus* (Burchell 1882) has experienced rapid growth in Africa, Asia and Europe. In order to improve its rearing systems, it is imperative to determine the individual growth patterns of the species. Individual fish growth is relevant when evaluating commercial viability, planning the time of harvest or projecting profits.

In this study, we analysed the growth data of African catfish juveniles reared in an indoor freshwater recirculation aquaculture system and fed five different commercial aquafeeds over a period of 111 days. We fitted five nonlinear growth models—von Bertalanffy, Gompertz, Brody, Richards and Logistic—to the weight data and applied the Akaike information criterion, the sum of squares and the maximum likelihood to determine the model that best describes the growth data of the species under the controlled culture conditions.

According to the goodness-of- fit criteria, the Gompertz model was consistently the one that best described the growth of African catfish. The F-test showed significant differences ($p = 0.05$) for one of the experimental feeds (T1,2) that produced the superior growth in terms of rate and biomass.

TA B L E 3 Parameters and goodness of fit of the five growth models for *C. gariepinus* in captivity

Parameter	Gompertz	BVTF	Logistic	Brody	Richards
W_o	30	29	N/A	0	30
W_{max}	1,016	2,060	623	2,471	896
K	0.017	0.008	0.048	0.002	0.021
M	N/A	N/A	2.89	N/A	0.12
SSQ	90,008	95,386	113,992	186,215	90,350

TABLE 4 Results of the Akaike information criterion analysis for the five growth models

Model	*K*	*AIC*	*AICc*	*ΔAICc*	*AICW*	*Cum W*	*-LL*
Gompertz	**3**	**2.1**	**2.2**	**0.000**	**0.25**	**0.25**	**-91.07**
VBTF	**3**	**2.1**	**2.2**	**0.003**	**0.25**	**0.50**	**-90.71**
Logistic	**3**	**2.3**	**2.4**	**0.195**	**0.23**	**0.73**	**-72.75**
Brody	**3**	**2.7**	**2.8**	**0.585**	**0.19**	**0.91**	**-46.43**
Richards	**4**	**4.1**	**4.4**	**2.154**	**0.09**	**1.00**	**-89.89**

APPLICATIONS OF BIOINFORMATICS TOOLS ANALYSIS FOR GENE REGULATORY NETWORKS INVOLVED IN SEXUAL MATURATION OF RAINBOW TROUT (*Onchorhynchus mykiss*)

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In the present study, we used bioinformatics approach and compare gene regulatory networks using microarray data to identify gene expression differences in sexual maturity traits of rainbow trout (*Onchorhynchus mykiss*). The gene expression profiles of sexual maturity were investigated and compared based on microarray gene analysis. We identified putative related signaling pathway and interaction networks associated with sexual maturation. Differentially expressed genes (DEGs) and Gene Ontology analysis were applied to identify protein encoded and then signaling pathway involvement using and gene functions were clustered using WEBGSAL, Bnlearn, Cytohubba and Cytoscape softwares. A total of 174 significant genes were differentially expressed at more than a two-fold change in fish were treated without feeding as compared to regular feeding. Of these, 94 upregulated genes and 80 downregulated genes were confirmed. The results showed that the genes BRCA2, DCT, EDC3, GJA1B, CFL2, CFD, MELTF, ZNF33B, SEC24D and PIPE had significant effects on sexual maturation and related to reproductive process. The results of the present study indicate that the bioinformatics microarray analysis could provide important data regarding molecular mechanisms of reproductive process for future experimental investigations.

META-ANALYSIS OF QUANTITATIVE TRAIT LOCI (QTLS) REVEALS THE GENETIC CONTROL OF GROWTH-RELATED TRAITS IN FARMED RAINBOW TROUT (*Onchorhynchus mykiss*)

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The field of association mapping studies has recently received major attention for genetic studies of quantitative traits in many important aquatic species. Access to next generation sequencing technologies, high phenotypic data and a variety of sophisticated statistical tools have enabled association mapping studies in aquatic species to be successful in identifying gene loci controlling quantitative traits. Due to the importance of association mapping method in mapping studies of the quantitative traits, the present project was prepared to explain the association mapping method and its use in rainbow trout breeding and also to perform a meta-analysis of these QTL to identify regions of the rainbow trout genome that are consistently associated with growth traits. To identify Meta-QTL (MQTL), a QTL database was developed from 1400 QTL targeted at growth traits. This project also provides some information about statistical software packages used in association mapping and then the opportunities and challenges of association mapping and post-genome wide association studies at the whole genome level discussed. For QTL mapped relative to a single marker, nucleotide sequence of the marker was retrieved from the relevant marker database. For QTL mapped relative to two flanking markers, sequences for both flanking markers were retrieved from the database. The positions of individual QTL were projected onto a consensus genetic map based on the presence of common molecular markers and a 95% confidence interval (CI) was calculated for each QTL. After positioning the individual QTL, the software 'Biomercator v2.1' was used to predict the location and CI of MQTL based on maximum likelihood. In total, 854 QTL were reported for 80 growth traits. This included 280 for average daily gain (ADG), 16 for body weight (BW), 9 for Condition factor (CF) and 7 for fork length (FL) trait QTL in rainbow trout genome. In total, 27 QTLs were detected on four linkage groups for the studied traits. That from 2 to 23/8% of the phenotypic variation (LOD) were justified. Most QTLs were detected on 13 linkage groups. In this study, for the body height traits not detected a QTL. Results revealed the existence of co-localized QTLs for studied traits, which enhance the efficiency of marker-assisted selection and developing rainbow trout breeding programs

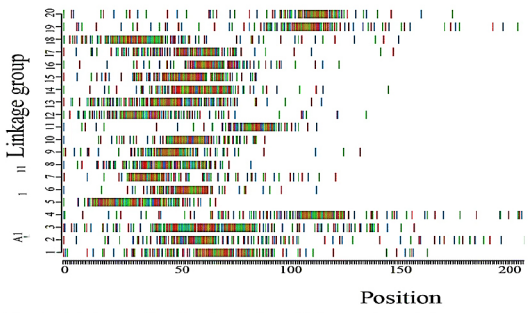

Fig. 1. Linkage distribution of multi-locus for rainbow

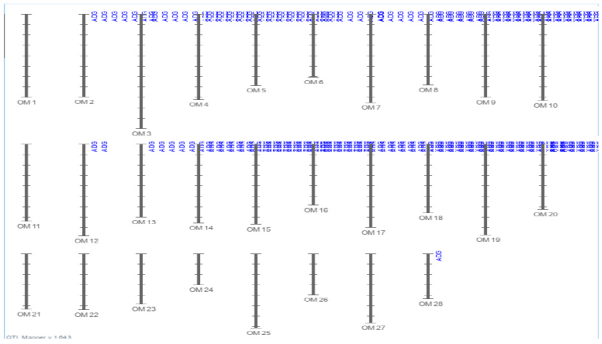

Fig. 2. Distribution of QTLs for body weight trait for the rainbow trout

IN VITRO STUDY OF GREEN SYNTHESIZE NANOPARTICLE FROM *Terminalia catappa* AS CANDIDATE ANTIBACTERIAL PATHOGEN IN AQUACULTURE FIELD

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Green synthesis of nanoparticle using silver (Ag+) and plant extract as a bioreductor is gaining a momentum. *Terminalia catappa leaves* has a great potential as an antibacterial pathogen in aquaculture field. Current study purposed was to evaluate in vitro antibacterial assay of silver nanoparticle (AgNPs) synthesized using Silver nitrate and *T. catappa* ethanolic leaves extract against *Aeromonas hydrophila* and *Pseudomonas fluorescens*. Scanning Electron Microscope (SEM) was performed to confirm the resulting nanoparticle. Meanwhile, in vitro assay of antibacterial activity of the AgNPs was done by agar well diffusion. Fourier-transform Infrared Spectroscopy was used to evaluate active compound in the AgNPs which may be responsible as antibacteria. The results stated that AgNPs had a cubic form and size range between 59-87 nm (Figure 1). In vitro antibacterial activity of AgNPs found optimum value at 20%, showing inhibition index at. 8.12± 0.29 mm against *A. hydrophila* and 3.13±0.15 mm for *P. fluorescens* (Table 1). Though Inhibition index of AgNPs is lower than Chloramphenicol (Control +), the AgNPs biosynthesized using *T. catappa* ethanolic leaves extract is a potential to be a candidate antibacteria. Further, the FTIR analyses found different functional groups some group active compounds which might be responsible to antibacterial activity (Figure 2).

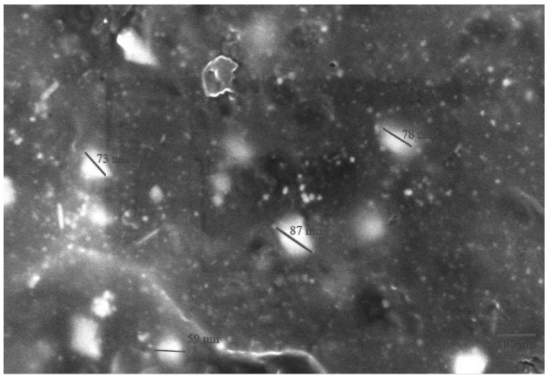

Figure 1. SEM analysis of AgNPs using green synthesize from ethanolic leaves extract of *Terminalia catappa*.

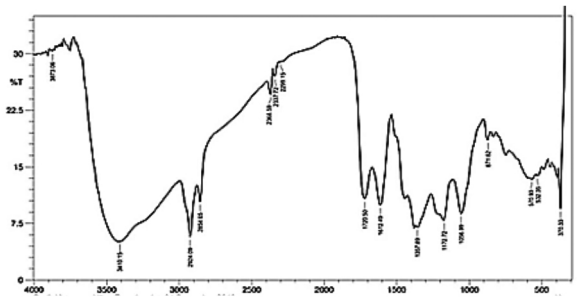

Figure 2. FTIR Spectrum silver nanoparticle synthesized using *Terminalia catappa* ethanolic leaves extract

Table 1. Inhibition index (mm) comparison between ethanolic extract of *Terminalia catappa leaves* and AgNPs against *Aeromonas hydrophila* and *Pseudomonas fluorescens*

Groups	Inhibition index (mm)	
	Aeromonas hydrophila	*Pseudomonas fluorescens*
Control (-)	0	0
Control (+)	9.78± 0.23	11.67± 0.23
10%	3.43± 0.47	1.39±0.40
20%	8.12± 0.29	3.13±0.15
40%	4.66± 0.15	3.03±0.66

Control (-) = Aquadest, Control (+) = Chloramphenicol 0.1%. *Aeromonas hydrophila* and *Pseudomonas fluorescens* were isolated from infected *Clarias gariepinus*. AgNPs is the nanoparticle which was synthesized using silver nitrat and *Terminalia catappa* leaves ethanolic extract

GENOME-WIDE IDENTIFICATION AND EXPRESSION ANALYSIS OF SIX INSULIN-LIKE PEPTIDES WITHIN *Haliotis discus hannai*

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Insulin is one of the essential hormones that make up the body metabolic system by converting glucose into glycogen. In animals, known as insulin-like peptides (ILPs), they are involved in growth, reproduction, and endocrine secretion. In the case of abalone, there is no report on the insulin-related system, and it will play an important role in elucidating the endocrine system of *Haliotis discus hannai*. This study aims to identify six Abalone insulin-like peptides of northern abalone through transcriptome sequencing and to confirm changes after feeding through the expression levels of these peptides. Furthermore, RNA-Seq data was utilized to further explore the expression patterns of AIPs, which may help uncover vital functions of Pacific abalone. Based on the above analysis, six insulin like-peptides were analyzed each hour after feeding. this study showed a relative increase in AIP1-5 expression in hepatopancreas 3 h after feeding compared to other tissues. In most of the experimental groups, the expression levels before feeding were restored after 6 h. However, AIP6 showed the highest relative expression level after 1 h, and then decreased after 3 h of feeding. This is the first report showing the uptake of feed influence on the expression of insulin-like peptide in abalone species and it is assumed that a physiological regulatory mechanism using insulin-like peptides may exist in *H. discus hannai* endocrine system.

TENDENCIAS FENOTÍPICAS PARA SUPERVIVENCIA Y PESO A LA COSECHA EN UNA POBLACIÓN DE CAMARÓN BLANCO DEL PACÍFICO *Litopenaeus vannamei* BAJO SELECCIÓN

Ordoñez Ramírez José Manuel, Alejandra Caballero-Zamora*, Thania Medrano-Mendoza, Psique V. Rivero-Martínez, Juan C. Quintana-Casares, Gabriel R. Campos-Montes

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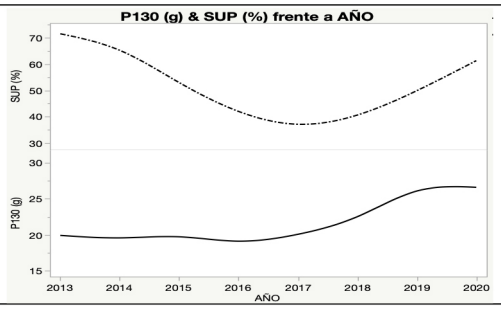
Un programa de mejoramiento genético (PMG) permite aprovechar la variación genética de las especies y así poder maximizar su producción. Para la producción de camarón, los PMG se centran en reducir el tiempo de crecimiento, aumentando el peso a la cosecha, así como en mejorar la resistencia a enfermedades teniendo una mayor supervivencia. Una forma de determinar el éxito en los PMG de camarón es a través de la evaluación de tendencias fenotípicas para el peso y supervivencia entre generaciones sucesivas. En esta idea, el presente estudio tuvo como objetivo evaluar los efectos de un programa de mejoramiento genético sobre la producción de camarón en dos granjas comerciales.

Se utilizó la información del peso y supervivencia a la cosecha en estanques comerciales, en Bahía de Kino en Sonora México, de la empresa Maricultura del Pacífico de los años de 2013 a 2020. A partir de 2013 se inició la suititución de una línea especializada en crecimiento por otra con antecedentes de resistencia al Síndrome de Mancha Blanca, manteniendose el criterio de selección para peso corporal y la sustitución final ocurrió en el ciclo 2016. Para obtener la tendencia fenotípica, se utilizó un modelo mixto que incluyó en el modelo además de la media poblacional para P130 y S, respectivamente, el tipo de ciclo de engorda (corto, largo), época de cultivo (primavera, verano), densidad y año de cultivo como efectos fijos; así como grupo genético y residual como efectos aleatorios.

Se detectaron diferencias significativas para tipo de ciclo y densidad de siembra, siendo el tipo de ciclo el factor con mayor efecto tanto para la supervivencia como al peso a la cosecha. En el peso promedio a la cosecha hay diferencias significativas ($P<0.05$) a través de los años, siendo 2016 el año con menor valor (16.70 g ± 0.71) y 2019 el de valor más alto (30.97 ± 0.91). Por otro lado, en supervivencia también hay diferencias significativas ($P<0.05$) en el tiempo, donde las supervivencias más bajas fueron en el periodo de 2015 a 2018. Es posible observar que el cambio de línea genética tuvo el efecto deseado en supervivencia, así como el efecto de la selección en el peso.

Medias mínimas cuadráticas ± error estándar y tendencias fenotipicas, para peso y supervivencia a la cosecha de *Litopenaeus vannamei* en condiciones comerciales del periodo 2013 a 2020

Año	*Peso (g)*	*Supervivencia (%)*
2013	20.40 ± 0.98	68.0 ± 3.8
2014	17.85 ± 0.78	73.5 ± 3.4
2015	22.75 ± 0.87	49.5 ± 3.1
2016	16.70 ± 0.71	42.2 ± 2.7
2017	21.44 ± 0.64	34.3 ± 2.3
2018	18.87 ± 0.84	40.6 ± 3.1
2019	30.97 ± 0.91	50.7 ± 3.4
2020	24.66 ± 0.91	62.0 ± 3.4



RELACIÓN ENTRE ESTIMADORES DE CONSANGUINIDAD OBTENIDOS A PARTIR DE INFORMACIÓN GENÓMICA Y DE PEDIGRÍ EN *Litopenaeus vannamei*

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En la camaronicultura la consanguinidad (F_x) influye sobre características de interés económico, por lo que es importante evaluarla. Generalmente el cálculo de la F_x se realiza utilizando el método descrito por Wright (1922), que requiere de conocer la genealogía de la población y con la desventaja de ser susceptible a errores de registro, derivando en paternidades erróneas e imprecisiones en su estimación. El uso de Polimorfismos de Nucleótido Simple (SNPs), permite estimar la homocigosidad de cada individuo y por lo tanto estimar la $F_{x;}$ sin embargo, su implementación es más costosa que la metodología tradicional. Por lo anterior, el objetivo del estudio fue evaluar la relación entre los estimadores de consanguinidad obtenidos a partir de información genómica y de pedigrí en una población de *L. vannamei.*

Se utilizó la información de la población de la empresa Maricultura del Pacífico SA de CV. Considerando la información de 1944 individuos que cuentan con un pedigrí de 19 generaciones de profundidad. Los genotipos fueron obtenidos con el panel AquaArray HD (50K) vannamei® (Neogen®), que considera 50,811 SNPs. Para el control de calidad (QC), se utilizó la frecuencia de alelo menor inferior al 1%, una tasa de llamado para SNPs de 80%, el equilibro de Hardy-Weinberg con un valor de significancia menor a $p < 10^{-6}$ y una exclusión de individuos por tasa de llamado < 75%, resultando en 36,171 SNPs y 1,930 individuos. La estimación de la F_x por pedigrí se estimó con profundidades de pedigrí de 5 y 19 generaciones (G5 y G19) utilizando el software Endog (v4.8), mientras que para la consanguinidad genómica se calculó del índice de consanguinidad poblacional (FIS) que considera la relación de la diferencia entre la heterocigosidad esperada y observada y la heterocigosidad esperada calculada a partir del equilibrio de Hardy Weinberg. Se estimaron las correlaciones de Pearson entre las consanguinidades individuales considerando G5, G19 y FIS.

La F_x promedio (D.E.) para G5 y G19 fueron de 1.49 (1.7) y 2.25 (1.8) y la correlación entre ellas fue cercana a 1, lo que indicaría que la profundidad del pedigrí no afecta de manera importante la estimación de F_x con el método tradicional, mientras que las correlaciones de G5 y G19 con FIS fue relativamente baja. Lo que podría estar relacionada a que la información de pedigrí considera que los individuos de la primera generación no están relacionados entre si, ni considera los genes idénticos por ascendencia, por lo que es posible que las estimaciones con el método tradicional estén subestimadas a pesar de la profundidad del pedigrí.

Cuadro 1. Correlaciones de Pearson para índices de consanguinidad con genealogías con profundidad de 5 y 19 generaciones e índice de consanguinidad (FIS).

	G5	G19	FIS
G5	**1**	0.9865	0.3461
G19		**1**	0.3513
FIS			**1**

G5 y G19: corresponden a información de pedigrí de 5 y 19 generaciones de profundidad, **FIS:** el índice de consanguinidad.

AMINOACID ANALYSIS IN FILLET OF THE NILE GRAY TILAPIA *Oreochromis niloticus* RAISED IN TRADITIONAL CULTURE SYSTEMS AND WITH BIOFLOC TECHNOLOGY AFTER THE FATTENING PERIOD

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Fish is considered a meat of important biological value as it represents a valuable source of high quality animal protein; this protein is highly digestible and provides all the essential amino acids required by humans. The amino acids are the fundamental constituents of the protein, and those that are present in the food are the decisive ones to determine the quality of the protein of the tilapia fillet. Of the 20 existing amino acids, 10 can be synthesized by the body (non-essential), and the rest need to be supplied to the body through the diet (essential). The Nile gray tilapia is one of the most consumed species in the world, it has characteristics that allow its cultivation in sustainable systems with little or no water exchange, such as the biofloc system, which provides a supplement in the nutrition of cultivated organisms. The biofloc not only provides complementary food *in situ*, but also produces extracellular enzymes that break down proteins, carbohydrates and other nutritional ingredients, facilitating the digestibility and absorption of the food. The type of diet offered to farmed fish has a significant effect on the nutrient composition of the meat, and can be adapted to produce a desirable nutrient profile in the farmed species. To determine the effect of the biofloc system on the amino acid in fillet of the Nile gray tilapia *Oreochromis niloticus*, an experiment was carried out for 120 days, with two treatments and three repetitions: traditional system (TS) and heterotrophic biofloc system (BFT). Male monosex organisms with initial average weight of 109 ± 39.29 g were used at a stocking density of 100 fish m^{-3}, fed with extruded pellets with 32% CP. In the present work, nine of the ten essential amino acids were analyzed in the tilapia fillet; where six of these (lysine, threonine, methionine, isoleucine, leucine and phenylalanine) were present in a higher proportion in the tilapia fillet grown in the biofloc system (**Table 1**). With the results obtained in the present work, it is concluded that the biofloc system has influence in the protein quality of the meat due to the consume of the microrganisms presents contributes a constant nutritional supplement compared with the traditional system.

Table 1. Amino acids present in the fillet of Nile gray tilapia *Oreochromis niloticus* in traditional culture systems and with biofloc technology.

Amino acids	Traditional system	Biofloc system
Lysine	1.97	2.30
Histidine	1.55	0.31
Threonine	1.19	2.33
Arginine	1.53	0.43
Valine	1.00	0.90
Methionine	0.92	0.99
Isoleucine	0.75	1.12
Leucine	0.78	1.42
Phenylalanine	1.63	8.09

BIOTECHNOLOGICAL POTENTIAL OF MARINE BACTERIA ISOLATED FROM EUTROPHIC SEA PIT IN THE ADRIATIC SEA

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Extreme marine habitats are inhabited by diverse microorganisms which have developed a wide variety of adaptations to help them succeed in these challenging environments. Microorganisms isolated from harsh environments are a rich source of new and stable enzymes with unique properties that could improve industrial processes and make them more economical and environmentally friendly. In this study, we isolated and examined bacteria capable of surviving in eutrophic sea pit. We further examined the microbial capacity to synthesize hydrolytic enzymes and antimicrobial compounds of biotechnological or medical interest.

We investigated microbial diversity in an underwater sea pit piled with ~15 m high *Posidonia oceanica* waste biomass that has accumulated for over a century. The pit is located in the central Eastern Adriatic; the entrance to the pit is located 7 m below the sea surface and the bottom of the pit is at 32 m below the surface level. The degree of biomass decomposition increases towards the bottom of the pit (Fig. 1A). The samples of the degrading biomass were collected at 19 m and 29 m depth and incubated at 16 °C on 13 different solid media to recover culturable microbes adapted to this niche. Isolates were identified using either MALDI-TOF MS or 16S rDNA and were tested for antibacterial and antifungal activity. Different extracellular enzymatic activities of isolates were assessed with specific substrates for proteases, esterases, lipases, amylases, cellulases, and chitinases. Out of 298 cultivable isolates across 13 media, 67 isolates exhibited 99% 16S rRNA gene sequence identity to *Alcanivorax borkumensis* (γ-proteobacteria). Analysis of *rpo*B sequences from four *Alcanivorax* isolates displayed 90% similarity to *A. borkumensis* suggesting so far unreported species. This genus is known to thrive on petroleum and is therefore useful for biodegradation of oil spills. Our isolates showed strong esterase and lipase activities. This microorganism can tolerate up to 10% salt in medium supplemented with 1% of oil and form specific mold-like hydrophobic clumps at the liquid medium surface. Confocal time-lapse microscopy has shown that this bacterium can colonize oil droplets more rapidly than *A. borkumensis* (Fig. 1B). Different assays revealed capacity of this bacterium to degrade various hydrocarbon substrates. The currently ongoing whole genome sequencing will reveal a more complete arsenal of enzymes associated with oil bioremediation and other metabolic processes.

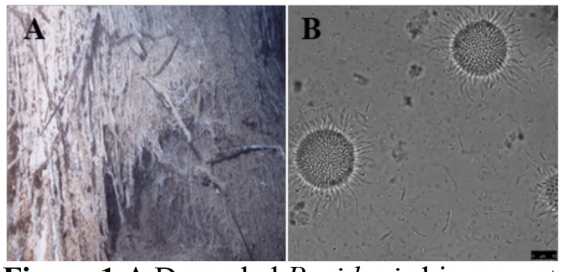

Figure 1.**A** Degraded *Posidonia* biomass at 29 m depth in marine pit; **B**. Colonization of oil droplets by *Alcanivorax* sp.

NEGATIVE EFFECT OF ECTOPARASITE BURDENS ON THE CONDITION FACTOR FROM FARMED TILAPIA *Oreochromis niloticus* IN THE YUCATAN, MEXICO

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Nile tilapia (*Oreochromis niloticus* Linnaeus, 1758) is one of the most important aquaculture species in the world, and, when introduced, the ectoparasites of Nile tilapia have followed. Currently, farmers worldwide consider these ectoparasites harmless, but intensities can reach up to 1,000 individuals per fish in tropical regions. In this cross-sectional study, we used the condition factor to estimate the potential effects of low (45 ± 31 ectoparasites per fish) and high (295 ± 191) ectoparasitic burdens across 28 tilapia farms and included the analysis of the effects of 44 management and environmental variables from the farms. A stepwise procedure in a multiple linear regression analysis retained the variables that explained the most variance, which was the ectoparasitic burden (57%). We found significantly higher values of the condition factor in Nile tilapia with low ectoparasitic burden than in those with high ectoparasitic burden. Additionally, Nile tilapia with a high ectoparasitic burden weighed less than half than those with a low burden (102 ± 105 g versus 230 ± 128 g, respectively). We also found a significant non-linear, negative relationship between the ectoparasitic burden and the relative condition factor values per fish, which was most likely due to an increase in gill mucus caused by the ectoparasitic burden that depleted energy in the Nile tilapia. The economic consequence of the ectoparasitic burden translated roughly into a loss up to 12-15% in profit margin per ton of fish, based on the price of Nile tilapia in the Mexican market.

WEIGHT-LENGHT RELATIONSHIP OF *Ictalurus punctatus* AND *Pangasius hypophthalmus* CATFISHES IN COMMERCIAL EXPERIMENTAL CONDITIONS

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Channel catfish (Ictalurus punctatus, Rafinesque, 1818) is one of the most important species for aquaculture in Mexico. *Pangasius hypopthalmus, known* as Basa, is not officially recognized, but already cultured in different states. Weight Length relationship (WLR) is an useful that can be used for converting lengths into biomass, determining the fish condition, comparing fish growth among areas, and complement to species-specific reproduction and feeding studies. An experiment was conducted submmiting fishes to growth-out conditions during 40 days using 1 m^3 floating cages (three repetitions by population) at a density of 250 fishes/m^3. Using final weights and furcal lengths, double logarithmic regression models were fitted to estimate a and b parameters. Fulton condition factor (K) was estimated.

The Figure 1, shows the logarithmic plots of data from Local Channel Catfish and Basa. The line of prediction estimated a b=3.02 (R^2>0.95) indicating isometric growth. Basa population WLR had a slightly lower b parameter (b= 2.92, R^2>0.95) that suggest a relative negative allometric of hipo-allometric, and can be interpreted as fishes that weigh less than their length predicts or, related to the better feeding condition of small organisms at the time of sampling. K for Channel Catfish and Basa catfish populations, were 1.84±0.127 and 1.65±0.179, respectively, with no statistical differences between populations (P>0.05). The results indicates good and similar growth potential under commercial conditions and estimates can be used as reference information or prediction of biomass under commercial conditions.

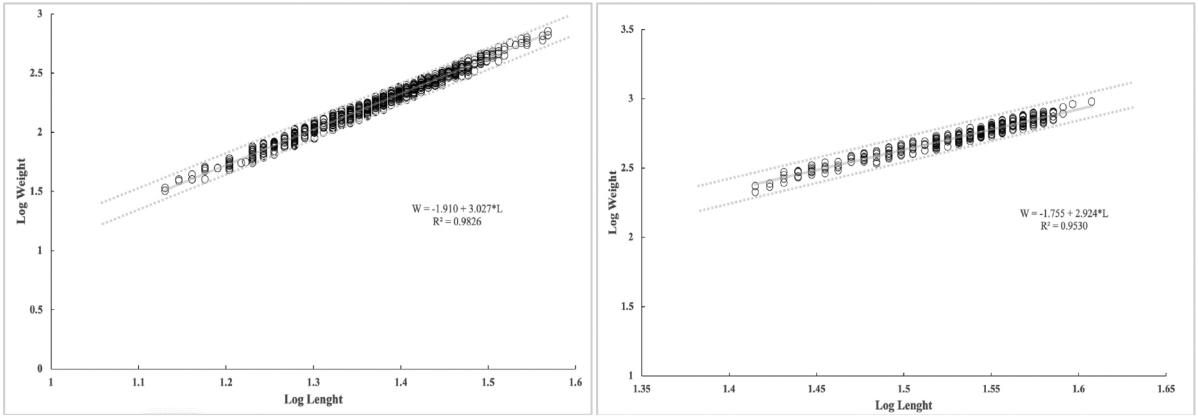

Figure 1. Double-logarithmic plot of data of local channel catfish (Ictalurus punctatus) and Basa catfish (Pangasiodon hypopthalmus) weight and length growth under commercial experimental conditions. Channel catfish: n= 868. Coefficient of variation= 1.98%. 95% CL α: -2.098-1.723; 95% CL b: 3.012-3.039. Basa catfish: n= 351. Coefficient of variation= 0.97%. CL α: -1.808-1.702; 95% CL b: 2.890-2.958.

COMPARATIVE STUDY OF ANALYTICAL TECHNIQUES IN BLOOD SAMPLES OF COMMON SNOOK, *Centropomus undecimalis*

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The white sea bass, *Centropomus undecimalis*, is a species of fishing importance that inhabits coastal lagoons and river mouths during its juvenile stage. It is a species with high potential for cultivation, for which it is essential to monitor its nutritional and health status. In this work, clinical diagnostic analytical techniques were compared using blood to characterize nutritional status and physiological status.

36 organisms cultivated in the marine fish area of UNAM, Sisal Unit, were used. The fish were anesthetized (eugenol), and the blood sample was obtained by puncture in the caudal vein. Plasma metabolites were evaluated using clinical diagnostic kits, Accutrend Plus test and refractive index to compare the concentration of total protein, glucose, lactate, cholesterol, and acyl glycerides. Immunological status was also determined by hematocrit, hemoglobin concentration, and hemagglutinating activity. A paired analysis was performed to compare analytical techniques (diagnostic kits and test strips), and the effect of sample preservation time (fresh and 24 hours).

The results indicate that there is a significant effect by the type of evaluation, and also by the preservation time of the sample ($p < 0.05$). Thus, the results indicate a decrease in the concentration of metabolites and hemagglutinating activity in samples preserved in refrigeration, indicating that comparative studies should consider the analytical technique used, and also the preservation time of the sample in refrigeration.

The data obtained agree with values reported in wild organisms of the same species, indicating the possibility of using blood components as indicators of physiological status for nutritional studies and to monitor wild organisms.

Acknowledgments
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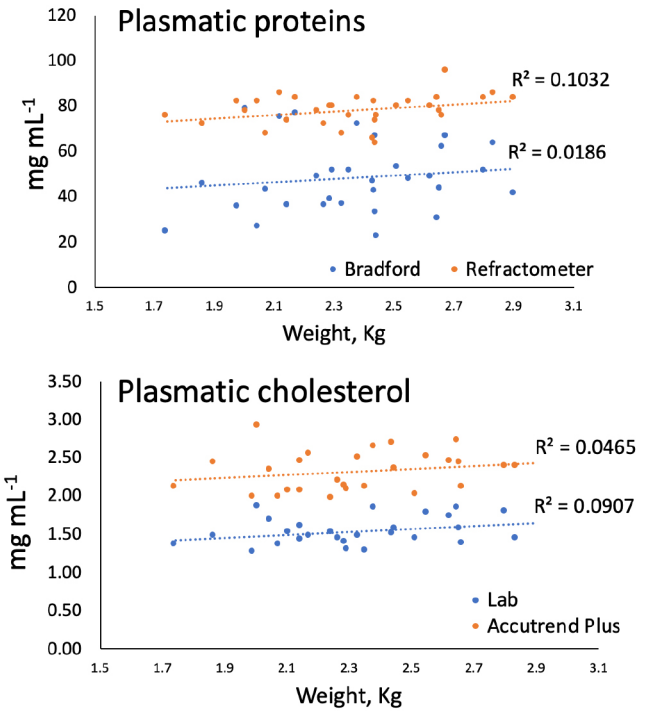

EFECTO DEL HONGO *Ganoderma lucidum* SOBRE LA SALUD DEL CAMARÓN *Litopenaeus vannamei* CULTIVADO EN ECUADOR

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El Ecuador se convirtió en el primer productor de camarones a nivel mundial en el 2021, con un total de 1847,73 millones de libras exportadas, lo que significó un incremento del 24% en comparación a las exportaciones del año anterior.

Aun bajo este escenario la producción camaronera en Ecuador enfrenta constantemente grandes desafíos patológicos. Es así que los registros de mortalidades desde maduración pasando por larvicultura hasta la fase de engorde, mayormente son causadas por bacterias del género Vibrio, cuyo nivel de impacto está relacionado con las condiciones del agua y del suelo, incremento de densidades en el cultivo donde hay limitaciones de infraestructura, así como posibles factores ambientales que se pueden dar a lo largo del año. Este estrés se convierte en un enemigo silencioso de las producciones, por lo que como herramientas de manejo se han empleado diferentes productos para salud que potencializan y activan el sistema inmunológico, permitiendo a los camarones responder ante los posibles detonantes de enfermedades.

Productos inmunomoduladores como los extractos obtenidos del hongo *Ganoderma lucidum*, cuenta con más de 200 fitonutrientes , de los cuales 150 son poderosos antioxidantes, metabolitos funcionales como los ácidos ganodericos . 1,3/1,6 beta glucanos, proteína LZ8, así como alcaloides, tripertenoides, ganoderanos, germanio orgánico polisacáridos con amplias propiedades anti fúngicas, antibacterianas y anti inflamatorias, han demostrado tener un efecto en el mejoramiento de la capacidad inmuno moduladora, convirtiéndolo en un profiláctico de interés en la acuacultura. Mediante cromatografía liquida de alta resolución (HPLC), se ha podido purificar, extraer, identificar y caracterizar doce de los cientos de ácidos ganodericos presentes en el hongo para estudiar individualmente su efectividad.

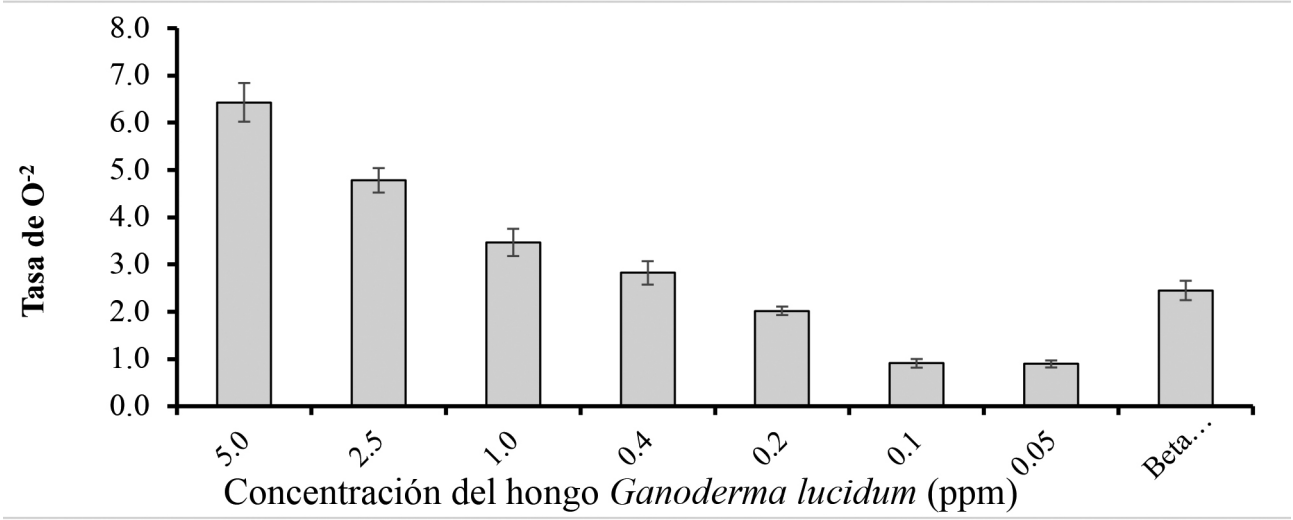

Figura 1. Tasa de producción de anión superóxido en cultivos primarios de hemocitos de camarón a diferentes concentraciones del hongo *Ganoderma Lucidum*

(Continued on next page)

A la fecha no hay reportes del uso de hongo *G. lucidum* en camaronicultura por lo que el presente trabajo es el primero que describe los resultados obtenidos tanto *in vitro* como *in vivo* en camarón *Litopenaeus vannamei*. Un estudio de efectividad *in vitro*, contra cultivos primarios de hemocitos de camarón fue desarrollado a diferentes concentraciones del hongo *G. lucidum* desde 0.05 ppm hasta 5 ppm (Fig. 1), donde por medio de la técnica de reducción del nitra blue tetrasolium (NBT), se pudo determinar que la producción de anión superóxido se incrementaba constantemente a medida que se aumentaba la concentración del hongo a partir de 0.2 ppm. Los resultados muestran una estimulación de los hemocitos a la concentración de 0,2 ppm, mostrando tasas de O_2^- superiores a 1.5, considerada como la base promedio de activación de los mismos. A esta concentración de *G. lucidum* resulto ser del 10% al 20 % menor a las empleadas por otros productos imunoestimulantes basados en levadura.

La concentración mínima inhibitoria letal también fue ensayada en cinco cepas bacterianas que son conocidas como patógenas para camarones. Los resultados al término de 48h mostraron que una concentración de1000 ppm fue suficiente para inhibir el crecimiento *Vibrio parahaemolytcus, Pseudomonas flourescen, Pseudomonas sp*. En tanto que se requirió 3000 ppm para matar al *Vibrio vulnificus y Vibrio harveyi*.

Pruebas realizadas *in vivo* en larvicultura durante 3 ciclos de cultivo y en cuadruplicado en cada ciclo fue suministrado *G. lucidum* al alimento en una de las raciones de alimentación diaria 4g/kg y al agua a razón de 5g/TM una sola vez al día. Al término de los ciclos de larvicultura que duraron 18 días se observó en promedio un 17% más de supervivencia a favor de los tanques que recibieron *G. lucidum* junto con un 95% de uniformidad en tallas de las larvas. Mientras que los tanques control terminaron con una supervivencia promedio de 73% y una uniformidad de tallas a la cosecha del 83%. Estos resultados son consecuencia del mejor estado de salud lo cual fue evidenciado por análisis histopatológicos, donde se observó larvas con menor carga bacteriana y estructuras definidas en sus órganos internos (resultados a presentarse)**.** En las maduraciones para contra restar las mortalidades causadas por el transporte se evaluó la aplicación de 20 g /TM de agua. Los resultados al final del trasporte desde camaronera hasta los laboratorios de maduración mostraron una reducción de las tasas de mortalidades de 15 % a 1,5 %, demostrando efectividad en la reducción perdida de reproductores, camarones de gran importancia para los programas de mejoramiento genético.

En la fase de precria el hongo *G. Lucidum* fue evaluado en una dosis de 5 g/kg de alimento en piscinas de tierra con una densidad de 4 millones de post larvas por ha, reporto una supervivencia superior al 70%, proporcionando una diferencia promedio a favor del producto del 40%, más de supervivencia con respecto a las precrias que no lo emplearon, demostrando su inmunomodulación y mejora en la salud de los animales.

Este trabajo demuestra la capacidad inmunomodular el sistema inmune del camarón y de inhibir el crecimiento de bacterias patógenas que tiene el hongo *G. Lucidum,* lo cual se corrobora con la mayor supervivencia reportada en larvicultura y la precria del camarón. Adicionalmente fue evidenciado una menor disparidad de tallas en larvas que es un problema comúnmente asociado entre una de esas a condicionantes de estrés.

Este estudio evidencia que el uso del hongo *G. Lucidum* es una alternativa para la prevención de estrés y control frente a cepas bacterianas, principales responsables de pérdidas en la producción ecuatoriana.

Palabras claves. Hongo, *Ganoderma lucidum*, inmunomodulador, camarones, sistema inmune, concentración mínima inhibitoria, larvicultura, precria

EFFICIENCY OF BROWN ALGAE (*Fucus virsoides*) POLYSACCHARIDES IN RETENTION OF LIPOPHILIC BIOACTIVES DURING SEA BUCKTHORN (*Hippophaë rhamnoides* L.) OIL SPRAY DRYING

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Plant species such as sea buckthorn (*Hippophaë rhamnoides* L.), (SB) and brown algae (BA) are rich in numerous lipophilic and hydrophilic bioactive molecules (BAM) but their potential as functional food (FF) ingredients are not yet used adequately. The most valuable part of SB is berry oil due to high content of carotenoids, tocopherols, fatty acids, sterols, omega-3, -6 and -7 fatty acids while BA are valuable source of polyphenols, pigments, lipids, proteins and polysaccharides which possess different biological activities such as antioxidant and anti-inflammatory etc (Shah et al., 2021; Quitério et al., 2021). BA polysaccharides are already used in food industry as gelling, thickening and emulsifying agents. High content of structurally different BAM in SB oil and BA making them an excellent choice for design of FFs. However, carotenoids and tocopherols present in SB oil are potent antioxidants but highly unstable and susceptible during processing and storage. The encapsulation is an effective approach to prevent their degradation and control release in food products. The most common method for the effective encapsulation of BAM is spray drying (SD) but the quality of the final product considerably depends on proper carrier selection (Eun et al. 2019). Therefore, the aim of this study was to determine carotenoid and tocopherol content as well as antioxidant capacity (AC) in SB oil powders produced by SD using different carriers [β-cyclodextrin (CD) and gum arabic (GA)] and oil to carrier ratio (1.2; 1:4) with addition of polysaccharides (0, 15 and 30%) extracted from BA (*Fucus virsoides*) at drying temperature of 150 °C. Results showed the highest retention of carotenoids and tocopherols and AC in powders produced using CD carrier and oil to carrier ratio 1:4 with addition of 30% of polysaccharides. The addition of polysaccharides significantly affected retention of carotenoids.

Table 1. Content of lipohilic BAM and AC in spray dried SB oil

Carrier	Oil to carrier ratio	Addition of polysaccharides (%)	α-tocopherols (mg/100 g oil)	Total carotenoids (mg/100 g oil)	ORAC (umol TE/100 g oil)
GA	1:2	0	41.17±1.41	14.09±0.22	239.87±0.42
GA	1:4	0	44.50±1.06	7.41±0.35	299.06±0.52
GA	1:2	15	43.15±1.14	15.74±0.41	246.15±1.05
GA	1:4	15	46.85±0.38	9.50±0.52	301.94±0.74
GA	1:2	30	48.91±0.48	22.26±075	252.01±1.41
GA	1:4	30	51.10±0.45	12.12±0.54	302.93±0.52
CD	1:2	0	51.02±1.25	16.39±0.56	217.96±0.41
CD	1:4	0	67.05±1.45	24.52±0.41	323.12±0.21
CD	1:2	15	51.86±0.56	20.96±0.52	232.05±0.74
CD	1:4	15	68.62±0.88	26.12±0.18	330.75±0.19
CD	1:2	30	55.57±0.45	28.87±0.41	245.84±0.42
CD	1:4	30	70.97±0.22	30.96±0.35	335.65±0.61

Results are expressed as mean±SD.

Table 2. Influence of SD parameters on lipophilic BAM content and AC of spray dried SB oil

Source of variation		á-tokoferol (mg/100 g oil)	Ukarotenoidi (mg/100 g oil)	ORAC (umol TE/100 g oil)
		$p \leq .05^{*}$	$p \leq .05^{*}$	$p \leq .05^{*}$
Carrier	GA	45.45±2.25^{a}	13.62±1.54^{a}	270.95±17.06^{a}
	CD	59.17±2.25^{b}	23.05±1.54^{b}	278.18±17.06^{a}
Oil to carrier ratio	1:2	51.63±2.09^{a}	17.89±1.43^{a}	272.15±15.80^{a}
	1:4	53.51±2.09^{a}	19.14±1.43^{a}	277.08±15.80^{a}
Addition of polysaccharides (%)	0	49.31±2.86^{a}	14.76±1.95^{a}	262.59±19.35^{a}
	15	52.34±2.86^{a}	17.83±1.95^{ab}	276.70±19.35^{a}
	30	55.76±2.86^{a}	22.56±1.95^{b}	283.36±19.35^{a}

Results are expressed as mean±SE. Values with different letters within column are statistically different at p≤0.05.

FRESHWATER PRAWN, *Macrobrachium rosenbergii* CULTURE IN QUINTANA ROO, MEXICO

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Worldwide demand for freshwater prawns has been increasing significantly in the last few years. The production of freshwater prawn *M. rosenbergii* in México is close to zero. The main cause is the lack of hatchery post larvae production. In the Indo-Pacific region production commenced based on government supported PL production. In the 70s and 80s the Mexican government produced PLs that were distributed to growers with little support for production and, thus, little success. Or solution was to produce PLs to demonstrate that commercial rearing is a viable business. This will bring investors into the nascent industry.

Conditions in the State of Quintana Roo for *M. rosenbergii* culture are ideal in terms of climate, water supply, available land, market, connectivity to national and international markets and security.

We started a pilot Project of PL production for grow out at our farm. First year production was 600,000 Pls and the hatchery is being modified to double production. At present our infrastructure consists of 6 20-m circular HDPE tanks for Pl nursery, 8 1,800 m^2 earthen ponds and 6 1 ha ponds for grow out and process, selection and cool storage rooms (under construction). Our personnel is highly skilled with experience in all aspects of production. PL production cycles require 30 days, the nursery phase is 2 months and grow out is a 5-6 month process.

Production results are similar to reports from Asia with consistent yields over the last 3 cycles. Production costs are discussed, representing 50% of sale price for a market that is, currently, being supplied from imports. The main product for national and international markets is frozen with prawns of around 80 grams.

Production of freshwater prawns in Quintana Roo represent a significant opportunity to develop a sustainable, socially responsible and economically viable business.

AVANCES EN EL CULTIVO DE LANGOSTINO MALAYO *Macrobrachium rosenbergii* EN QUINTANA ROO MEXICO

Jorge Ignacio Peláez Pier * José Rafael Reyes Campos, Erika Lolany Camacho Chuy, Marily Cruz Ramírez

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Comenzamos a trabajar con langostino *Macrobrachium rosenbergii* en el mes de octubre de 2020 con Postlarvas compradas al Dr. Juan Reta, se desarrollaron y de ahí se seleccionaron reproductores con características fenotípicas y de desarrollo.

En febrero 2020 montamos nuestro laboratorio para producción de postlarvas consiguiendo en el primer año 600,000 Pls, mismas que pasaron a una fase de crecimiento en tanques circulares de 20 m de diámetro, cosechándose de 2 a 3 gramos en un tiempo de 78 días promedio, sembrando para el ciclo de engorda en tanques de 1,800 m^2.

Las densidades de siembra fueron diferentes, buscando conocer las tasas de crecimiento a esas densidades y buscando conocer también las variables en desarrollo por efecto de temperatura.

Las primeras cosechas en estanques chicos (1,800 m^2) tuvieron resultados similares a los números de producciones de otros países sobre todo en el área Indo-pacifico que tienen muchos años produciendo, lo que nos lleva a considerar que, si todavía no alcanzamos la eficiencia deseada, vamos por buen camino.

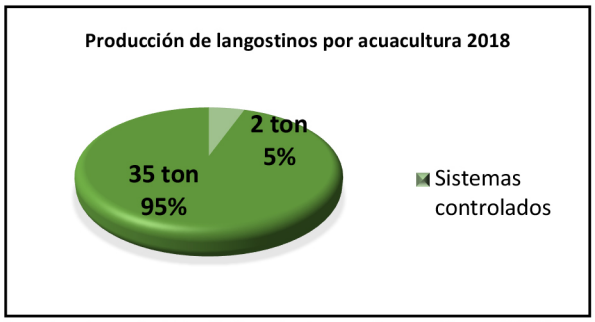
Siembra para desarrollo Pls 140/m^2

Siembra para engorda directa buscando tamaño jumbo 5/m^2, pudiendo tener variaciones estos tiempos dependiendo de las temperaturas.

El mercado en México esta abastecido 100 % por importaciones del área Indo-pacifico. El producto tamaño jumbo alcanza precios premium en el mercado internacional y mexicano. La presentación mayormente es producto congelado. La demanda en el mercado mundial se incrementa anualmente de forma importante.

La oportunidad para México como productor de langostinos malayo es excelente.

fuente: Anuario Estadístico de Acuacultura y Pesca 2018.



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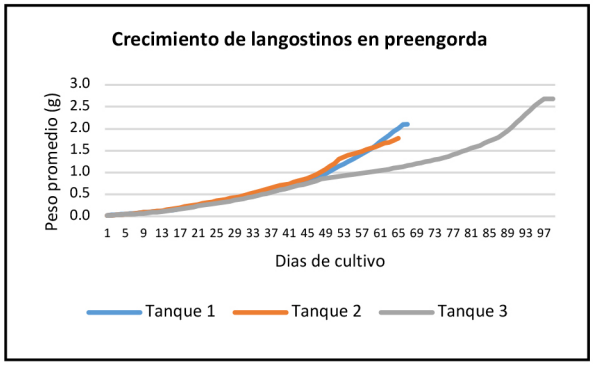

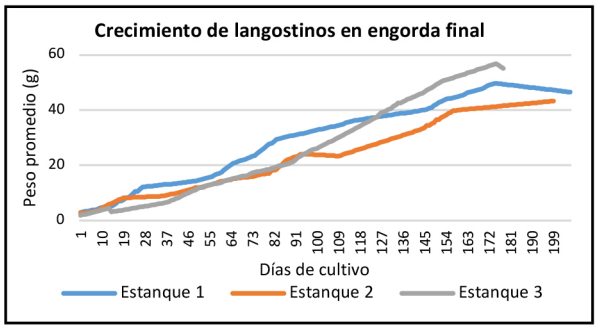

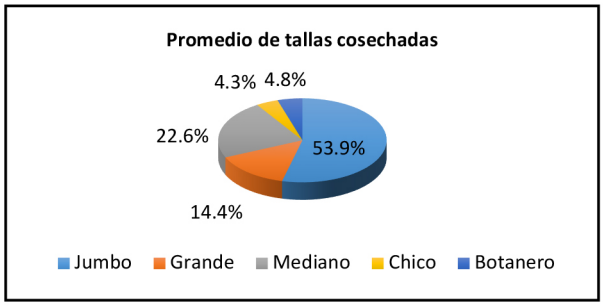

EVALUACION DEL SISTEMA DE PRODUCCION IN POND RACEWAYS EN UN PERIODO DE 3 AÑOS CON TILAPIA *Oreochromis niloticus*

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Iniciamos operaciones en canales rápidos en el mes de febrero de 2019 con la intención de reducir costos de producción para poder ser competitivos en el mercado.

En el primer año fueron resultados excelentes en todos los aspectos, decreciendo los números en el segundo año debido a 2 problemas.

1.- La materia orgánica generadas por las excretas de las tilapias, no fue posible extraer del estanque por el gran porcentaje de flotación de estos y el sistema de sifoneo con bomba de extracción de solidos operada manualmente, que no tuvo la capacidad de realizar correctamente el trabajo debido a que implicaba operarlo 24 horas, lo que no nos fue posible realizar.

2.- La reproducción de la tilapia, si se lleva a cabo en los canales rápidos, debido a la baja velocidad en el flujo de corriente en la parte pegada al piso del canal, ocasionando una importante población de tilapia en estanques. Con una reversión normal de 95%, implica que cada canal de 20,000 organismos tuviera 1000 hembras que al 2° mes en canales se están reproduciendo, si consideramos 3 ciclos al año por canal son miles de tilapias que se están reproduciendo en el estanque ocasionando un consumo de oxígeno y generación de materia orgánica.

La combinación de estos 2 problemas nos ocasiono fuerte deterioro en la calidad del agua, reduciendo los niveles de oxígeno que afecto en niveles de desarrollo y supervivencia.

Los 2 problemas tienen solución, en el caso de extracción de sólidos, el uso de bombas viajeras automáticas programables. En la reproducción de tilapias, siembras en el estanque de peces carnívoros que no se reproduzcan en el mismo. **CORRIGIENDO LOS 2 PROBLEMAS LOS CANALES OPERARAN ADECUADAMENTE.**

ESTANQUE 3

SIEMBRA				DATOS DE COSECHA TOTAL											
Fecha	Org. (#)	Bio. (kg)	Prom. (g)	Fecha	Dias de cultivo	Cosecha total	Org. (#)	Org. (g)	Bio. Obtenida en canales	Alim.	FCA	g/dia	org/m3	Bajas registradas	% sobrevivencia
03-mar-19	26,409	2,165	82	28-jun-19	118	15,652	25,587	611	13,484	17,690	1.31	4.5	158	628	96.8
01-sep-20	22,456	2,425	108	31-dic-20	122	10,337	19,541	529	7,912	12,896	1.63	3.5	120	1,943	87.0
14-jun-21	17,430	1,714	98	08-oct-21	117	10,540	16,844	625	8,826	12,310	1.4	4.5	104	411	96.6

ESTANQUE 5

SIEMBRA				DATOS DE COSECHA TOTAL											
Fecha	Org. (#)	Bio. (kg)	Prom. (g)	Fecha	Dias de cultivo	Cosecha total	Org. (#)	Org. (g)	Bio. Obtenida en canales	Alim.	FCA	g/dia	org/m3	Bajas registradas	% sobrevivencia
11-nov-19	24,589	1,974	91	03-mar-20	113	16,471	24,139	682	12,388	16,575	1.33	5	130	367	98.1
30-nov-20	19,860	1,638	82	03-abr-21	129	9,054	17,448	518	7,416	11,577	1.6	3.3	93	2,635	87.8
17-may-21	17,778	2,102	118	03-sep-21	110	8,834	16,606	531	6,732	10,025	1.48	3.8	89	952	93.5

PREVALENCIA DE LA NECROSIS AGUDA DEL HEPATOPÁNCREAS EN CAMARONERAS DEL GOLFO DE NICOYA Y PACÍFICO CENTRAL DE COSTA RICA

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La enfermedad de la necrosis aguda del hepatopáncreas (AHPND, por sus siglas en inglés) es ocasionada principalmente por bacterias *Vibrio parahaemolyticus* que contienen un plásmido (pVA-1), que codifica los genes de las toxinas *pir* A y *pir* B. El AHPND se caracteriza por la atrofia y descoloración del hepatopáncreas, con descamación masiva de células epiteliales en ese órgano, ocasionando anorexia, mostrado por intestino vacío, y llevando a la muerte de los camarones.

El objetivo del presente trabajo fue determinar la presencia de AHPND en fincas de camarones que cultivan *Penaeus vannamei* en Costa Rica. Durante el 2019, se recolectaron camarones y agua de estanques (33 estanques muestreados) de 31 camaroneras localizadas en el Golfo de Nicoya y Pacífico Central (Figura 1). Las muestras se analizaron mediante la técnica de reacción en cadena de la polimerasa (PCR), utilizando diferentes protocolos e iniciadores para detectar los genes de las toxinas *pir* A y *pir* B. El 93.9% de los estanques (31 de 33) muestreados fueron positivos para *Vibrio parahaemolyticus* causante de AHPND (Figura 2).

Los resultados confirman la presencia de AHPND en agua y hepatopáncreas de camarones en casi todas las fincas camaroneras muestreadas en la zona costera del Pacífico de Costa Rica.

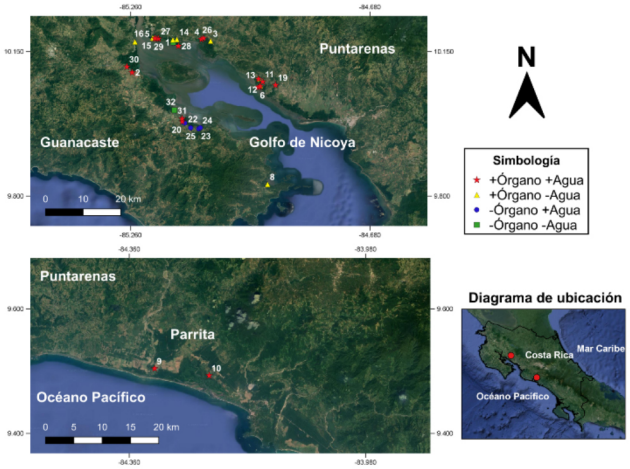

Figura 1. Distribución geográfica de las fincas productivas de camarón negativas y positivas a genes de las toxinas que ocasionan AHPND en Costa Rica, 2019. A. Fincas camaroneras ubicadas en el Golfo de Nicoya. B. Fincas camaroneras ubicadas en Parrita.

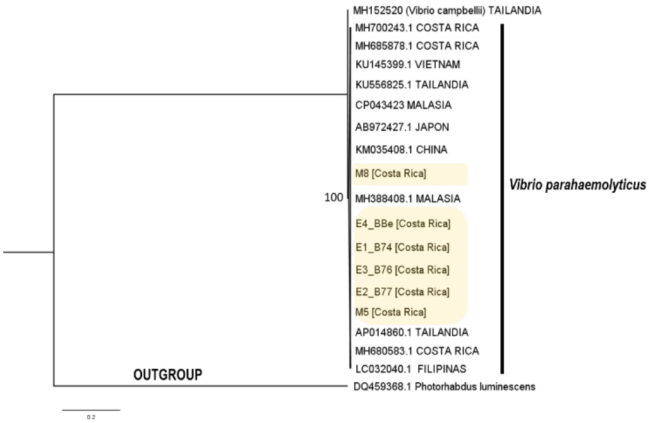

Figura 2. Topología de identificación molecular mediante máxima verosimilitud (ML), basado en secuencias parciales del gen de la toxina pirA de *Vibrio parahaemolyticus* (M5, M8, B74, B76, B77, BBe) y obtenidas del Genbank. El número en los nodos indica el valor de soporte bootstrap (porcentaje derivado de 5000 repeticiones). *Photorhabdus luminicens* está posicionado como Outgroup en el árbol filogenético.

BUILDING CONFIDENCE IN INNOVATION: A NOVEL APPROACH TO VALUATING NEW TECHNOLOGIES IN AQUACULTURE

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Implementing new technologies in the market is one of the main strategies used by firms to increase their competitiveness. A recurring problem that hinders the presence of new technologies in the market is to reach agreements that align the interests of technology developers with those of producers. In the case of aquaculture, an added difficulty arises in evaluating new technologies, since working with living organisms implies non-linearity of the productive factors. This study describes and shows the application of two complementary bioeconomic indicators, which allow facilitating negotiation and reducing technology transfer risks. The indicators proposed are "*minimum coverage*" – defined as the minimum values of one or more factors that constitute the production function, justifying investment in a technology and "*maximum price*" – defined as the maximum value that a new technology can reach in a way that justifies the investment made by the buyer (Figure 1). These metrics are exemplified by their assessment in a hypothetical case study applied to the use of genetic improvement programs in salmon production.

The application of contracts with pre-established guarantees is a significant argument for trust generation, which is especially relevant when dealing with new technologies associated with a significant investment since in this manner the risk is shared. The concepts developed in this study are aimed at obtaining analytical values that allow maximizing trust between parties, facilitating the development and transfer of new technologies, which has a crucial role to play in achieving sustainable aquaculture growth.

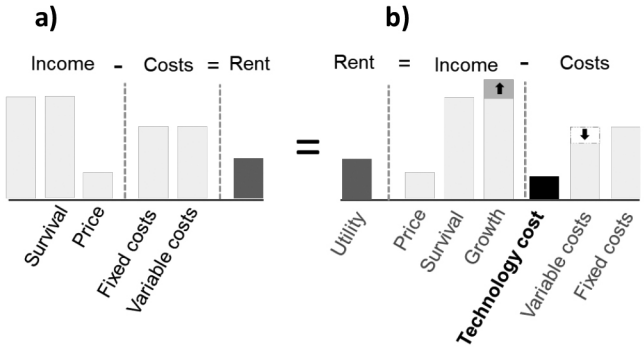

Figure 1. Exemplification of estimating minimum coverage and maximum price indicator with a technology that reduces variable costs and increases growth. The indicator conception is the same regardless of the factors impacted by the new technology. (a) Corresponds to utility function of the productive unit under normal conditions: (b) Is the new utility function when incorporating a new technology. The minimum coverage corresponds to minimum changes in cost and income functions required to cover the technology cost while the maximum price describes the maximum value a technology may have for a company derived from changes in productive factors of its utility function.

IS THERE ONLY ONE SPECIES OF *Hepatozoon* INFECTING BRAZILIAN CAIMANS? MOLECULAR ANALYSIS BRINGS NEW INSIGHTS INTO HAEMOGREGARINE DIVERSITY

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The population of Brazilian caimans is considered the largest in the world. Brazil is the second-largest exporter of fur and Brazilian alligator farming is a potential aquaculture production in agribusiness and an alternative to reduce the disorderly and illegal extraction of these wild animals. Therefore, a strict sanitary scheme is necessary for the breeding sites to avoid the spread of diseases. Among the infections that affect these animals, Brazilian caimans are hosts of a great number of parasites, including hemoparasites.

In 2018-2020, eleven *Caiman crocodilus* from Mato Grosso State were collected and screened for haemogregarine parasites. Through morphological analysis, a species of the genus *Hepatozoon* (Adeleorina: Hepatozoidae) was identified with free gamonts [Fig.1a], gamonts with cytoplasmic vacuoles [Fig.1b] and mature gamonts [Fig.1c] in the blood smears, and meronts with merozoites [Fig.1d-f] in the liver of these animals.

The eleven isolates amplified targeting the 18S gene have shown 100% similarity among them, and 98.4% - 98.8% in comparison with other isolates of *Hepatozoon caimani*, the only species known to infect Brazilian caimans so far. Complementary to this, the phylogeny revealed a main clade of isolates from Brazilian caimans, divided into two subclades, one of this study and the other with *Hepatozoon caimani* isolates only [Fig. 2]. This result brings new insights on *Hepatozon* diversity infecting the caimans, with a possible new species reported in this study and generates important information for diseases in alligator farming in Brazil.

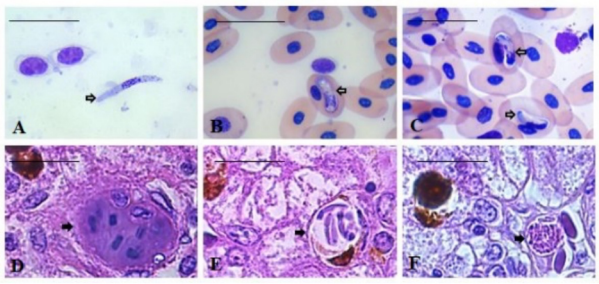

Figure 1: *Hepatozoon* sp. observed in the blood smears and histological slides of *Caiman crocodilus* specimens.

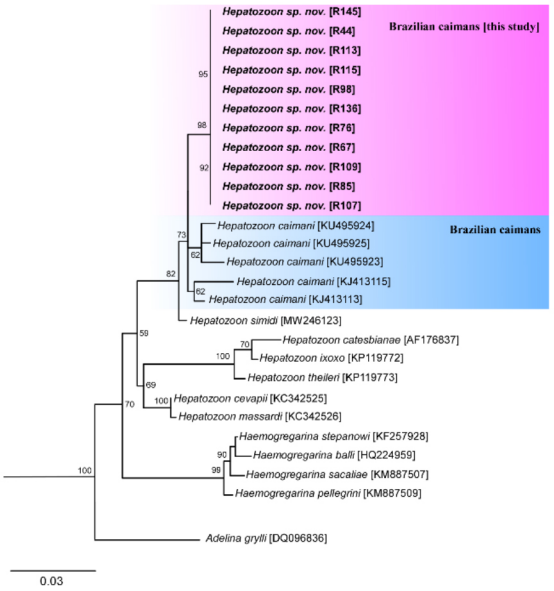

Figure 2: Phylogenetic analysis [Maximum Likelihood method] of haemogregarines based on 18S rDNA sequences.

FORO DE INVERSIÓN Y FINANCIAMIENTO EN ACUICULTURA

Sesión Especial del Congreso Mundial de Acuacultura WA2021
World Aquaculture Society (WAS)
Ciudad de Mérida, Yucatán, 24 a 27 de mayo de 2022

Genetic characterization of aquaculture populations

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One of the most important components in larvae and seed production in aquaculture is related to the genetic make-up of the breeding stocks. The genetic characterization is fundamental for crossbreeding strategies design within a genetic nucleus for its long-term maintenance and to support the obtaining of improved genetic lines either for growth, pathogen resistance or reproductive capacity, among other commercial important traits. This characterization consist in the genetic profiling of the breeding stocks to estimate, among other parameters, the levels of genetic diversity, inbreeding, kinship, and genetic differentiation among stocks.

At the laboratory of Aquaculture Genetics at CIBNOR, we offer an innovative scientific-based service for the genetic characterization of breeding stocks supported by the use of low-cost genotyping platforms that can be affordable for medium to low-sized producers. This service goes from sample collection to genetic data analysis, which is delivered to the hatchery owners and technicians by a technical report thoroughly explained in a dedicated workshop.

For the short-term future, we are also developing new genotyping strategies as well as implementing more sophisticated tools to include genomic data. All these with the goal of promoting a more productive and sustainable Mexican aquaculture.

Table. Relative genetic diversity grades based on number of alleles per locus (Na) and heterozygosity (He) for a hypothetical hatchery. Percentages represent the propotion of hatchery's broodstocks within each class. Colors represent management risk levels.

	He	Low	Medium	High
Na	Range	< 0.25	0.25 - 0.37	0.37 - 0.5
Low	< 1.6	2%	-	-
Medium	1.6 - 1.8	7%	34%	-
High	1.8 - 2.0	-	57%	-

A TECHNOLOGY FOR THE CULTURE OF AMBERJACK, *Seriola rivoliana*, IN MEXICO

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The need to increase production of foods with better nutritional quality is evident. Fisheries is more and more unsustainable and aquaculture will need to take a major role in providing nourishment if we want to contribute to improve human health in the years to come. Aquaculture has been growing steadily over the years and provides more than 50% of seafood for human consumption.

The amberjack, *Seriola rivoliana*, is considered an excellent candidate for sustainable aquaculture because is highly adaptable to captivity and intensification, breeds routinely, readily consumes extruded balanced rations, has fast growth rates to commercial size (2-3 kg in 18 months) and is well received in the market at prices that range from US$10 to 20/kg. The Northwest Biological Research Center has developed a relevant knowledge base for processes like broodstock capture, transport and maintenance, reproduction, larval culture, nursery rearing and grow out, while understanding disease interactions and the efficient use of feeds. In association with commercial producers, an innovative technology for the culture of the species has been developed to competitively enter the market. Information on initial investment for infrastructure, working capital, income and financials (IRR, NPV, B/C) is discussed.

PRINCIPLES AND PRACTICES OF SHRIMP FARMING IN ECUADOR AND WAY FORWARD

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Ecuadorian shrimp industry has experienced sustained growth over the last 20 years (Fig. 1). In 2021, shrimp exports exceeded 840,000 MT (a growth of 25% compared to 2020), positioning the country among the world's leading producers of *Penaeus vannamei*, the main supplier of shrimp to China and the European Union and one of the major suppliers to the United States. It is estimated that by 2027 Ecuador will double current production, reaching 2 million MT of shrimp.

Despite this notable and sustained increase, Ecuador's shrimp production model differs significantly from that used by its main competitors. To get out of the white spot crisis, which reduced production by 70% between 1998 and 2000, Ecuador opted to use selected breeders from among the survivors of ponds affected by white spot. At first using the mass selection method (choosing the largest survivors) and currently with breeding programs that use the most modern tools for this purpose.

While shrimp farming has experienced sustained growth, driven primarily by growth in global demand for seafood, it is no less true that much of this growth is attributed to the new and best practices that the entire Ecuadorian shrimp production chain has developed and implemented in recent years.

This presentation explores the evolution of the Ecuadorian shrimp industry since its first stages, the main changes experienced over time, as well as the challenges they have had to face and the plans for the immediate future.

Figure 1. Evolution of Ecuadorian shrimp exports between 1979 and 2021

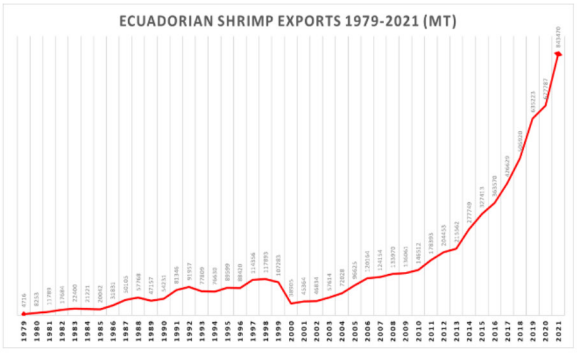

Table 1. Evolution of Ecuadorian shrimp culture conditions. Adapted from Piedrahita, Arcentales and Velasco (2007)

Period	1970-1980	1981-1992 before TSV outbreak	1993-2002 between TSV and WSSV	2002-2006 WSSV Recovering	2006- 2015	2015-present New tecnologies
Stocked species	*P. vannamei*	*P. vannamei* (wild/hatcheries source), *P. stilyrostris* (hatcheries)	*P. vannamei* (wild/hatcheries, imported SPF), *Cherax quadricarinatus*	*P. vannamei* (wild/hatcheries/genetic programs), tilapia	*P. vannamei* (only from local genetic improvement programs)	*P. vannamei* (only from local genetic improvement programs)
Kind of culture	Extensive	Semi- extensive	Semi-intensive	Extensive to semi-intensive	Semi-intensive	Semi intensive-intensive
Stocking densities (ind/m2)	2-5	5-15	10-30	6-12	10-25	15-25
Stocking way	Direct	Direct	Direct, nurseries	Direct, nurseries	Raceways, nurseries (2-3 phases)	Raceways, nurseries (2-3 phases), greenhouses
Feed/ feeding management	Natural primary production	Natural primary production and formulated feed/ direct	Formulated feed (direct, 2 times/day	Formulated feed (feed trays, 2-5 times/day	Functional feeds, automatic feeders, probiotic	Functional feeds, probiotics, automatic feeders, AI
Health controls	None	Protozoans, bacteria (microscopic cheks)	Protozoans, bacteria, virus (microscopy, agar culture)	Protozoans, bacteria, virus (microscopy, agar culture, PCR), Use of probiotics	Protozoans, bacteria, virus (microscopy agar culture, PCR), Use of probiotics	Protozoans, bacteria, virus (microscopy agar culture, PCR), Use of probiotics
Water/soil controls	None	Oxygen, pH, turbiduty	Oxygen, pH, turbiduty, nutrients, organic matter	Oxygen, pH, turbiduty, nutrients, organic matter	Oxygen, pH, turbiduty, nutrients, organic matter	Oxygen, pH, turbiduty, nutrients, organic matter. Monitoring sensors
Biosecurity	None	None	Water fitration, desinfection, seed analysis, vectors control	Water fitration, desinfection, seed analysis, vectors control	Water fitration, desinfection, seed analysis, vectors control	Fitration, desinfection, pathogen control, recirculation systems
Average new pond size (ha.)	15-30	10-15	5-10	5-10	1-5	1-5

RESULTS OF A COMMERCIAL VACCINATION PROGRAM BY INJECTION USING AN AUTOGENOUS VACCINE FOR *Streptococcus agalactiae* BIOTYPE 1a IN A TILAPIA CAGE FARM OPERATION IN HAITI

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Taino Aqua Ferme SA is a private company located in Lac Azuei, Fond Parisien Haiti. The company was founded in 2015 and produces whole fresh tilapia for the local Haitian market, currently producing 300 metric tons per annum. Tilapia are raised in 15-meter diameter HDPE cages inside Lac Azuei, an inland saline lake (10 ppt salinity) close to the border with the Dominican Republic on the island of Hispaniola.

Since the beginning of grow-out operations, mortality caused by Streptococcus led to low recoveries (51% on average). An approach of formulating an autogenous vaccine against this pathogen was implemented in November of 2020. To date, seventeen cages have undergone a full production cycle after having been vaccinated by injection at 30 grams ABW.

Results comparing these vaccinated cages vs control cages demonstrate an improvement of 12.5 % cumulative mortality throughout the cage farming growth cycle. Prior to implementation of the vaccination program, three day fasting periods and treatments using antibiotics approved for tilapia farming, were strategies used to mitigate acute mortality spikes. The program, which assures all the population in the farm is vaccinated, has led to a minimizing both fasting and medication, shortening culture cycle and dependency on antibiotic use.

DECOUPLED FLOCPONICS SYSTEMS AS AN ALTERNATIVE APPROACH TO REDUCE THE PROTEIN LEVEL OF TILAPIA JUVENILES' DIET IN INTEGRATED AGRI-AQUACULTURE PRODUCTION

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FLOCponics (DFP) is a promising aquaponics approach which takes advantage of the nutritional benefits of biofloc technology (BFT). Enabling the use of less protein in the fish diets is one of the benefits of BFT. The effect of the reduction of protein content, and consequently the input of nitrogen, on fish and plant production in DFP systems has not yet been investigated. This study was designed to investigate and evaluate the production of lettuce and tilapia juveniles in a DFP system using different levels of crude protein (CP) in the fish diets.

The zootechnical performance of tilapia juveniles and lettuce growth in the DFP system were evaluated, using different diets containing 24, 28, 32, and 36% CP. Fish production in DFP systems was compared to those reared in traditional decoupled aquaponics systems (DAPS) and in biofloc-based systems (BFT), both fed with 32% CP diet. The experimental period of tilapia juvenile production lasted 56 days. Lettuce production in two cycles was also performed in DFP systems with different CP levels and their growth was compared to those in DAPS and hydroponics systems, as control treatments. In *Cycle 1*, the seedling phase was evaluated in a 14-day trial. In *Cycle 2*, the final production phase was performed for 21 days until harvest. High mortality of fish occurred in DFP-36 in the middle of the experiment, thus this treatment was discontinued.

The results showed that tilapia reared in DFP and fed with 24 and 28% CP grew similarly to those in DAPS fed with 32% CP diet. Fish in DFP-32 and BFT-32 grew similarly and above the other treatments. Additionally, plant growth results showed no differences in both cycles among all treatments. The results obtained in this study indicate that using less CP in fish diets to produce lettuce and tilapia juveniles is technically possible and feasible in a decoupled FLOCponics system.

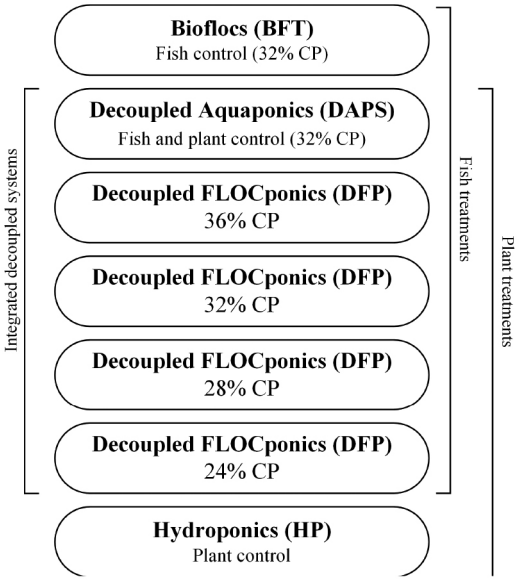

Figure 1. Schematic illustration of the experiment design. Three replicates of each fish treatment and six of each plant treatment were run. CP: crude protein

Funding: FAPESP (grants 2017/50431-9 and 2018/13235-0), NWO (grant 438-17-402), Belmont Forum (grant 726744), and CNPq (grant 311108/2017).

PARENTAL THERMAL STRESS AFFECTS THE PHYSIOLOGICAL CONDITION OF THE NEXT GENERATION OF JUVENILE *Octopus maya*

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There are records that *Octopus maya* adults could experience extreme temperatures during reproductive maturation. So far, there is no data to indicate what kind of consequences an offspring produced by thermally stressed parents might experience. The present study was dedicated to evaluating the effect of thermal stress in female *O. maya* at the end of their reproductive season on aerobic performance (AS) and antioxidant defense mechanisms (ADM) of juveniles exposed to extreme temperatures. For this, two groups of wild females were managed, some acclimated to 24°C and others to 30°C until spawning, and the juveniles of each group of females were divided into two groups, the first group exposed to 25°C and the other group at 30°C.

In the evaluation of the respiratory metabolism (Oxygen consumption in routine, as well as in the metabolic rate induced by the maximum temperature), it was observed that the juveniles from stressed females had higher metabolic rates than those observed from non-stressed females, mainly in animals at 30°C, suggesting a transgenerational effect of temperature on the energetic physiology of this animal (Fig. 1).

Aerobic scope was evaluated every 5 days until day 20 along with ADM and two indicators of oxidative damage (OD). The results obtained indicate that juveniles from thermally stressed females had limited physiological capacities to compensate for OD, being very sensitive to 30°C. In contrast, juveniles from non-thermally stressed females showed an ADM that, at least for a time, was able to eliminate the OD. These results are discussed in light of the possible consequences of warming between generations and its relationship with the time of exposure to high temperatures (Fig. 2).

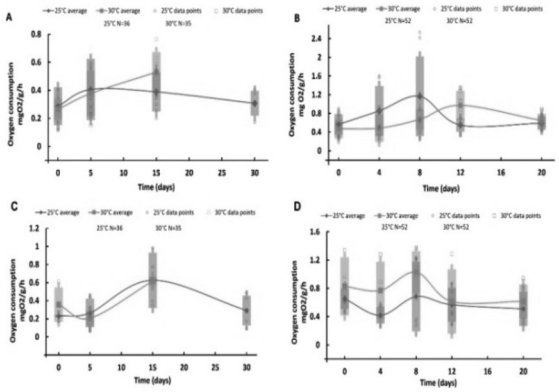

Figure 1. Routine (A and B) and high metabolic rate (TIMR max: C and D) of *O. maya* juveniles from un-stressed females (A and C) and thermally stressed females (B and D). Values as mean + SD.

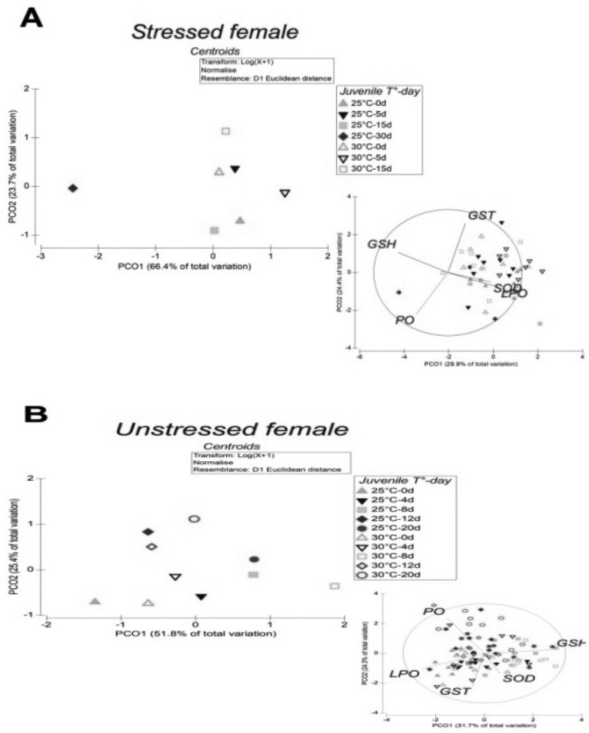

Figure 2. Changes of antioxidan defence enzymes and oxidant damage of *O. maya* juveniles during the experiments. Animals from (A) thermally stressed females, (B) non-stressed females.

PROSPECTIVE OF PANGASIUS PRODUCTION IN SOUTHWEST MEXICO

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Pangasius Aquaculture in Mexico is an activity that can be strategically promoted to create jobs and food security for low-income families in states with potential; especially in cases that present problems of malnutrition and socioeconomic lags. This is strategic to fight against hunger and, due to the demand for inputs, it stimulates the internal market, therefore, it has a great social impact, due to its wide range of technologies and systems from backyard family units to highly technical and intensive commercials farms. Pangasius Aquaculture is profitable and a good rural business under certain conditions. Mexico, due to its physical, natural and social characteristics, as well as its geographical position has all necessary and sufficient conditions to have a world leadership in this sector. This activity shows the highest economic growth in the primary sector worldwide during the last 10 years. The objective of this essay is to analyze the impact, areas of opportunity that aquaculture has and its potential in Mexico.

THE PORTUGUESE EXPERIENCE ON SARDINE'S *Sardina pilchardus* AQUACULTURE: ZOOTECHNICAL AND NUTRITIONAL ASPECTS

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The European sardine (*Sardina pilchardus*) is an important fish species for Portugal and the Mediterranean countries due to its cultural, economic and gastronomic importance. Also, by having an important value for the Portuguese and Spanish canning industry, sardines from the fisheries are traditionally absorbed by this sector. Besides, the stock of sardine populations in the Mediterranean Sea reached very low levels that lead to a strict stock management, which raised questions on the viability of the production of the species in captivity. At the Aquaculture Research Station of Olhão (EPPO), an aquaculture facility from the Portuguese Institute from the Ocean and Atmosphere (IPMA), which has the capacity for the conduction of trials at a pre-industrial scale, it was possible to successful adapt three lots of wild sardine's breeders which are currently on the third year of natural spawns.

The zootechnical parameters of the different stages of sardine culture in captivity (e.g. broodstock management, larval rearing and juvenile fattening) have been studied in order to obtain the maximum information on the aquaculture production of this species. Fish adapted well to captivity and presented a continuous and voracious feeding behavior. The comparison with wild individuals showed a higher content of lipids in the muscle in captive fish with adequate and not significantly different levels of EPA and DHA.

As nutrition is an extremely important factor for the success and quality of broodstock eggs, larvae and juvenile growth, nutrition trials have been carried out to assess the appropriate levels of protein in diets which was shown to be situated around 35% with higher values of protein in the diet favoring gonad maturation.

The results on sardine's production led us to consider that sardine's production in aquaculture might be promising in a near future, to ensure the demand for the fresh fish market and mainly to guarantee a constant supply for the canning industry. In order to establish an optimal nutritional protocol for sardine production, further and more detailed studies using metabolomics and molecular biology approaches are being conducted.

This study had the support of the project DIVERSIAQUA II (Mar2020-P02M01-0656P)

AQUACULTURE RESEARCH STATION OF OLHÃO: FACILITIES AND RESEARCH LINES

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The Portuguese Institute for the Ocean and Atmosphere (IPMA, I.P.) is a public research institute and act as a counselor to the national authorities on the sea and atmosphere. IPMA, I.P., possesses a strong cluster of competences for the ocean and marine resources related to research, carried out by different groups, particularly dedicated to aquaculture and fisheries.

The Aquaculture Research Station of Olhão (EPPO, figure 1) stands out for the unique experimental conditions on aquaculture at the national and international levels. This marine core facility is equipped to carry out production studies at every scale from bench-top laboratory work to a much larger semi-industrial level. EPPO has an area of about 7ha with more than 200 tanks, including an hatchery fully equipped for research and experimental production with different rearing circuits (for broodstock, larvae, juvenile production and research with live animals), a support building (with rooms for trophic chain production, daily routines and biological sampling), several analytical laboratories (biochemical, histological, molecular, microbiological and fish pathology), an unit for seafood packing, an area for pre-fattening (for earthen ponds and sea cages production) and 17 earthen ponds. It holds breeders of several marine fish species (e.g. meagre, gilthead seabream, seabass, Senegalese sole and sardine among others), microalgae and invertebrates as well as the know-how on the production of these species.

Production of new species, nutrition, welfare, environmentally friendly production systems and assessment of onshore and offshore and production systems for fish grow-out are some of research lines developed at EPPO (figure 2).

Acknowledgments: The research was funded by DIVERSIAQUA II (Mar2020-P02M01-0656P) project.

Figure 1 - Aerial view of the EPPO

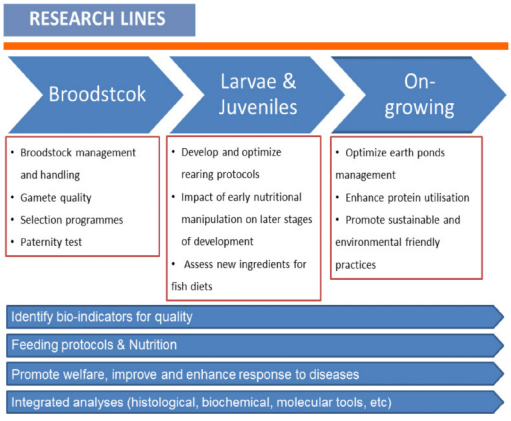

Figure 2 - Research lines developed at EPPO

microalgae and invertebrates as well as

MODELING JUVENILE TAMBAQUI (*Colossoma macropomum*) GROWTH IN AQUAPONICS SYSTEMS UNDER DIFFERENT STOCKING AND FEEDING RATES

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In 2018 Brazilian native fish culture accounted for 38% of the national aquaculture production being the Amazonian fish tambaqui the most produced species. Since aquaponics is a globally growing aquaculture technology, studies that evaluate the performance of tambaqui in aquaponics systems are needed. Growth models are an option to formally describe fish growth and consequently culture performance all along the culture period. These models serve also as a tool to make managerial decisions. A 34-day trial culture of tambaqui and lettuce in permanently coupled aquaponic systems was carried out during the summer in Brazil. The research facility comprises 16 experimental aquaponic units, each one containing one fish tank (0.38m^{-3}), 3 plant beds (0.42 m^2 of surface and volume of 60 L each), and filtering systems. Four fish stocking rates (50, 100, 150 and 200 fish m^{-3}) were combined with four feeding rates. The base feeding rate (E) was adapted from the recommended by the Brazilian Agriculture Research Corporation (EMBRAPA) and a value of 1.5% was added (E+1.5%) or the values of 1.5 or 3% were subtracted (E-1.5%, E-3% respectively) to stablish the other feed rates, totaling sixteen combinations. Fish with initial average weight of 16.88 g were introduced in the systems according to the corresponding stocking rate and at least 10% of the fish of each system were weighted weekly to adjust the feeding rates. With the weight (g) data, and after outlier elimination, the Von Bertalanffy, Champan-Richards, Gompertz, monomolecular and logistic growth models were tested. The model that best adjusted to tambaqui growth in the aquaponics systems cultured with lettuce was the logistic one, , were *W* is the weight in the respective culture day (*t*), and ,, the growth model parameters (Table 2). The models were validated through the Theil index which was minor than 0.2 in each case.

These growth models may be used to compare the tambaqui performance in aquaponics in other seasons or with other production technologies. Likewise, it may be useful as input for analysis of stocking and feeding rate optimization, bioeconomic analysis or system modelling.

Financial support: FAPESP (2018/23658-5, 2018/23605-9, 2017/50431-9), CNPq (311108/2017-2).

Tab 1. Juvenile tambaqui growth model parameters for the different treatments

Feeding rate	Parameter	Stocking rate 50	100	150	200
E +1.5%	α	133.1960	170.4289	124.8303	84.9018
	β	7.95050	10.23880	7.84030	5.97497
	γ	0.0812	0.0741	0.0935	0.1115
E	α	90.4348	105.5434	97.2417	71.6188
	β	5.04609	5.97760	5.62207	4.27411
	γ	0.09028	0.0672	0.08347	0.09580
E -1.5%	α	139.3820	86.07296	122.1549	89.20792
	β	7.07650	4.44960	6.84990	4.56759
	γ	0.0556	0.0779	0.0612	0.0626
E -3%	α	71.5052	66.7082	123.5800	154.1200
	β	3.37186	3.47166	7.32619	8.73665
	γ	0.0702	0.0594	0.0468	0.0406

ANTIOXIDANT ENZYME ACTIVITIES OF WHITE SHRIMP *Penaeus vannamei* UNDER COMPENSATORY GROWTH AND ABIOTIC STRESS

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The Pacific white shrimp, *Penaeus vannamei,* is one of the most economically valuable aquaculture species and the most widely farmed crustacean in the world. Expansion of aquaculture industry requires the implementation of strategies to reduce production costs. Among these, feed restriction is gaining interest due to the unusual acceleration of growth after a feed deprivation period, known as compensatory growth. While compensatory growth may allow reaching the weight of organisms never subjected to dietary restriction, the associated physiological costs are poorly studied, particularly in crustaceans.

A 52-day trial was conducted on juvenile *P. vannamei* (0.5 ± 0.02 g avg. wt.), exposed to three different dietary regimes: three cycles of 3 days of feed restriction and 3 of feed *ad libitum* (T3:3), four cycles of 3 days of feed restriction and 9 of feed *ad libitum* (T3:9), and an unrestricted treatment as control. Each treatment consisted of 3 replicates of 95 shrimp in 600-L tanks. Following the cyclic feed restrictions, all shrimp were fed *ad libitum*. Intensity of feed restriction was calculated to allow a 20% saving of feed over the experimental period for both restricted treatments. On day 35 (during compensatory response), hepatopancreas and muscle samples were collected. On the same day, 16 shrimp from each replicate were extracted and subjected to two abiotic challenges. Eight shrimp were subjected to 24-hour hyperthermia (32°C). The other 8 shrimp were subjected to an increase in ammonium concentration for 24 hours (40 mg L^{-1}). At the end of the abiotic challenges, hepatopancreas and muscle samples were collected.

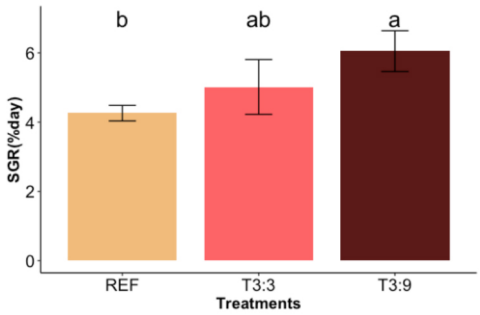

Figure 1. Specific growth rate between days 28-35 of experiment

After feeding restriction periods, T3:3 and T3:9, shrimp exhibited a lower weight than control organisms. On the final day, T3:3 reached a weight not significantly different from the control, showing full compensatory growth. T3:9 organisms were unable to reach the weight of the control organisms, however, they exhibited a significantly higher specific growth rate than the control during refeeding, indicating partial compensatory growth (Figure 1).

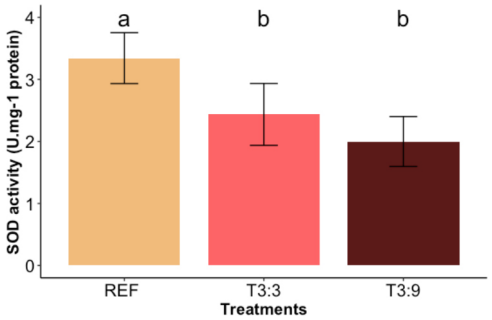

Figure 2. SOD activity in the hepatopancreas after hyperthermal challenge

At day 35, shrimp under compensatory growth showed lower hepatopancreas superoxide dismutase (SOD) activity than the control. Organisms' survival during the abiotic challenges was not affected by the different treatments. After the abiotic challenge, shrimp in compensatory growth showed a significantly lower hepatopancreas SOD activity than the control (Figure 2). Compensatory growth was related to altered antioxidant activity in *P.vannamei*, however, lipid peroxidation, the main consequence of oxidative stress, was not significantly affected between treatments.

IDENTIFICAÇÃO DE INDIVÍDUOS DE PIRAPITINGA (*Piaractus brachypomus*) SEM ESPINHAS INTRAMUSCULARES (SEIM) NA REGIÃO SUDESTE DA COLÔMBIA

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A pirapitinga (*Piaractus brachypomus*), originária da bacia do rio Amazonas, é considerada a principal espécie piscícola nativa produzida na Colômbia. Sua produção e comercialização têm sido limitadas devido à grande presença de espinhas intramusculares (EIM) em forma de Y na região dorsal e na região ventro-caudal, dificultando o processamento e consumo do filé. Em estudos de caracterização, conformação do sistema esquelético e confirmação da presença de EIM nos peixes têm se utilizado ferramentas de imagens diagnósticas como Raios-x e ultrassom. No entanto, ao ter maior sensibilidade e especificidade na detecção das espinhas, a Tomografia Axial Computadorizada (TAC) mostra um aporte complementário na identificação da presença ou ausência de EIM.

Posterior à indagação em campo, foi localizado na região sudeste da Colômbia um núcleo de pirapitingas de cultivo, sem espinhas intramusculares (SEIM) ou com reduzido número de espinhas intramusculares (RNEIM) e contando com a aprovação e participação do proprietário, se desenvolveu um protocolo de identificação e confirmação destes indivíduos.

De uma população de 80 exemplares da espécie *P. brachypomus*, com pesos entre 1.500-2.600 gramas, 33 indivíduos foram selecionados e marcados com microchip na base da nadadeira torácica. Foi feita biometria e se registraram características morfológicas específicas. Para a identificação inicial em campo de indivíduos SEIM e RNEIM, utilizou-se um equipo portátil de ultrassom em 24 exemplares. Deste grupo se selecionaram dois animais SEIM por ultrassom para confirmação através de Raios-X e TAC. Foi confirmada a existência de espécimenes SEIM, com RNEIM e com EIM (CEIM) na população analisada. O ultrassom é uma ferramenta de imagens diagnósticas não invasiva para realização em campo. Os Raios-X e TAC são estudos que permitem a confirmação e verificação da presença de EIM. Este é o primeiro reporte científico da existência de peixes SEIM e com RNEIM em populações de pirapitinga de cultivo.

Nosso grupo de pesquisa encontra-se trabalhando na caracterização morfológica, genética e de qualidade dos filés de indivíduos SEIM.

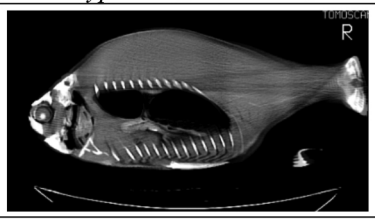
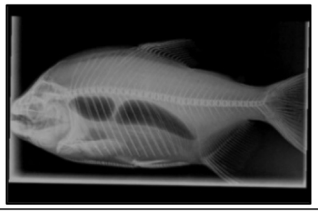
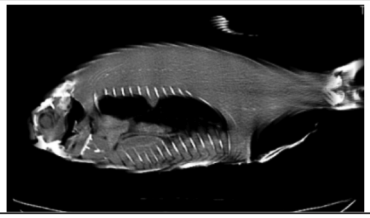
P. brachypomus SEIM – Ultrassom	*P. brachypomus* SEIM - TAC	*P. brachypomus* SEIM – Raios-X
		
P. brachypomus CEIM – Ultrassom	*P. brachypomus* CEIM - TAC	
		

Figura 1. Imagens diagnósticas realizadas em *P. brachypomus* CEIM e SEIM

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 LC50 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

TILAPIA PRODUCTION IN BIOFLOC SYSTEM

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Intensive culture of Tilapia has developed globally in different systems. The biofloc method is one of the effective responses to the environmental and nutritional issues in aquaculture industry. The system consists of a liner ponds enclosed with enclosed space to prevent evaporation and maintain water temperature and prevent temperature shocks with the central drainage. Aeration causes the flow of water and the discharge of the sewage from the center. Tilapia production in biofloc system has been activated in the desert central region of Iran and has led to the intensive tilapia production in indoor system, recently.

Tilapia was the secound fish cultured in the world during the last decade and it has the first stage of fish culture, now. Due to the special characteristics of this fish, including high adaptability and high resistance, its culture has development in various systems. The development of Tilapia breeding around the world, along with the lack of freshwater for agricultural and urban use, has gradually fueled tilapia from semi-intensive to intensive systems.

Despite the limitation of agriculture and aquaculture activities in the deserted areas due to limited agricultural soil and water resources, technology can response for the proper exploitation of these are sources for food and protein production. Intensive culture of Tilapia has developed globally in pools, tanks, raceways, cage, return and aquaponic systems. Water quality and nutrition management are the most important factors for success of intensive tilapia culture.

Tilapia is a filter feeder fish, so, fertility of pond has an important role in the culture. The biofloc method is one of the effective responses to the environmental and nutritional issues in aquaculture industry (Zhang & Luo, 2014). Biofloc technology is the use of a microbial community to produce a high-protein supplementary microbial source that is used by the filter feeder species. Aeration and proper mixing lead to water quality control. The microbial community, which is mainly heterotrophic bacteria in water, acts as a biofilter. The function of these bacteria accelerates the absorption of nitrogen and reduces ammonia levels and is more efficient than the nitrification process. Increasing hydrocarbons leads to fixing N and non-organic C producing proteins (Azim & Little, 2008; Day, 2015; Avnimelech 2012 & 2007).

In the biofloc systems water change is very low, the amount of feed consumed in these systems is minimized, the nutrient excreted from the breeding system is significantly reduced and nutrients are recycled regularly. Besides, using brackish water in the system leads to intensive tilapia culture.

Our implanted system consists of a liner ponds enclosed with enclosed space to prevent evaporation and maintain water temperature and prevent temperature shocks with the central drainage. Aeration causes the flow of water and the discharge of the sewage from the center.

Tilapia production in biofloc system is used in some parts of the world. In Iran, it has been activated in the desert central region of Iran and has led to the intensive tilapia production in indoor system, recently.

MAKING OFFSHORE AQUACULTURE VIABLE: ADVANCEMENTS IN SUBMERSIBLE NET PENS

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Interest in submersible or more rugged “offshore” (open ocean, high energy) pens has been increasing for the past several years because of the many benefits they bring to operator and the expansion potential they bring to the industry. However, they also can come with operational challenges like the risk of barotrauma in certain species due to cumbersome ascent/descent systems or the need to incorporate new feeding strategies when the pens are submersed. Innovasea’s designs incorporate features and capabilities to address many of these challenges.

ACUICULTURA VIABLE EN ALTA MAR: AVANCES EN JAULAS SUBMERGIBLES

El interés en las jaulas sumergibles o más robustas para operaciones “offshore” (mar abierto, alta energía) ha aumentado durante los últimos años debido a los muchos beneficios que brindan al operador y el potencial de expansión que brindan a la industria. Sin embargo, también pueden presentar desafíos operativos como el riesgo de baro trauma en ciertas especies debido a los engorrosos sistemas de ascenso/descenso o la necesidad de incorporar nuevas estrategias de alimentación cuando los corrales están sumergidos. Los diseños de Innovasea incorporan características y capacidades para superar muchos de estos desafíos.

EVALUACIÓN DE LA PATOGENICIDAD DE *Vibrio parahaemolyticus*, CAUSANTE DE AHPND, AISLADOS DE CAMARONERAS DEL GOLFO DE NICOYA EN COSTA RICA

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En Costa Rica la enfermedad de la necrosis aguda del hepatopáncreas es ocasionada por la bacteria *Vibrio parahaemolyticus* la cual posee el plásmido (pVA-1), que codifica los genes de expresión de las toxinas *pir* A y *pir* B. La presencia de esta bacteria en las camaroneras ocasiona mortalidades y pérdidas económicas, aun se desconoce la concentración mínima de bacterias necesarias para que se produzcan estas mortalidades.

El objetivo de este estudio fue determinar la patogenicidad de una cepa de *V. parahaemolyticus* aislada de tres estanques camarones del Golfo de Nicoya de una finca ubicada en Abangares en el Golfo de Nicoya. Se tomaron muestras de camarón con signos clínicos visibles, se recolectaron los hepatopáncreas en tubos con agar Cary-Blair, las muestras se rayaron en agar chromogénico para detección de *V. parahaemolyticus* (sigma aldrich). Las cepas azul verdosas resultantes (Figura 1), se analizaron mediante la técnica de reacción en cadena de la polimerasa (PCR) para determinar la presencia de las toxinas *pir* A y *pir* B.

Una vez confirmadas se rayaron en agar TSA con 2% de sal e incubaron 24h a 30°C, trascurrido el tiempo se tomó una colonia e inoculó en 50mL de caldo TSB con 2% NaCl, que se agitó a 120rpm y 30°C por 24h, se tomaron alícuotas para hacer las diluciones bacterianas de trabajo (10^3 hasta 10^7). Para el desafío se colocaron 18 recipientes con 3 L de agua a 20ppm y con 3 camarones de 4g. Se trabajó por triplicado cada concentración bacteriana y se registraron durante 10 días las muertes. Tanto en las muestras de la finca como en las muestras obtenidas de bioensayo, se les realizó PCR. Se concluye que la una densidad de bacterias de 10^6 causa mortalidades en menos de 24 horas, mientras que la dosis letal media fue de 10^5, luego de 10 días de realizado el ensayo.

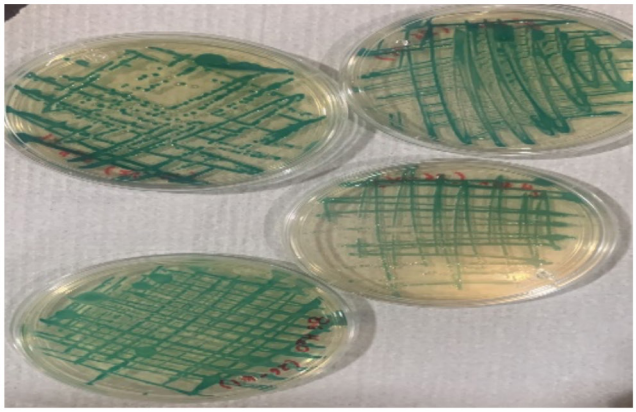

Figura 1. Colonias verdeazuladas obtenidas a partir de las muestras.

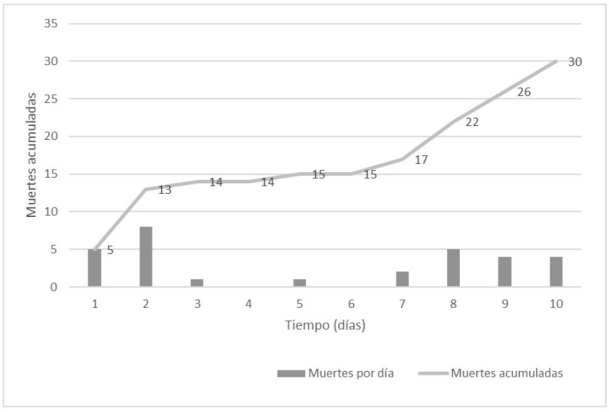

Figura 2. Muertes diarias y acumuladas registradas en los tratamientos.

ACCIÓN ANTIBACTERIANA DEL EXTRACTO HIDROALCOHOLICO DE *Salix babylonica* SOBRE BACTERIAS QUE AFECTAN LA PRODUCCION ACUICOLA DE TRUCHA ARCOÍRIS *Oncorhynchus mykiss* Y TILAPIA *Oreochromis sp.*

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Introducción.
La acuacultura es una actividad agropecuaria con potencial en México, la trucha arcoíris y tilapia son especies que se cultivan, teniendo una producción de 67,192 toneladas para el 2018. La producción en las unidades acuícolas, con altas densidades de cultivo, sin embargo, se ha determinado que los peces son más susceptibles a los agentes infecciosos. Por otra parte, la OIE menciona perdidas en la acuacultura por 6 mil millones de dólares al año, debido a enfermedades, además se espera el brote de nuevas enfermedades. Para controlar y tratar a los peces afectados por bacterias, se ha recurrido al uso de los antibióticos, sin embargo, se ha reportado que diversas especies de bacterias han adquirido resistencia a múltiples antimicrobianos. Las bacterias que afectan al cultivo de trucha arcoíris y tilapia son *Aeromonas hydrophila*, *Listonella anguillarum*, *Edwarsiella tarda* y *Streptococcus iniae*. Se ha buscado nuevas alternativas para combatir agentes bacterianos como los extractos de plantas, los cuales, contienen diversos compuestos secundarios como alcaloides, terpenos, taninos, saponinas, glucósidos y flavonoides, que han demostrado tener una actividad antibacteriana en estudios *in vitro*. El objetivo del presente estudio fue evaluar la actividad antibacteriana del extracto hidroalcohólico de *Salix babylonica* (EHSb) sobre *Aeromonas hydrophila*, *Listonella anguillarum*, *Edwardsiella tarda* y *Streptococcus iniae*, las cuales provocan alta mortalidad en la producción de trucha arcoíris y tilapia.

Material y Métodos: Para la obtención del EHSb se utilizaron 250 g de las hojas previamente secadas a la sombra, posteriormente se realizó una maceración con 1000 ml de una solución hidroalcohólica (70 % agua y 30% metanol).
Las cepas utilizadas fueron *A. hydrophila* CAIM^{347}, *L. anguillarum* CAIM^{763}, *E. tarda* CAIM^{1875} y *S. iniae* CAIM^{527}, obtenidas de la Colección de Microorganismos de Importancia Acuática (CAIM) del Centro de Investigación en Alimentación y Desarrollo de Mazatlán, Sinaloa, México (CIAD).

La actividad antibacteriana del EHSb se determinó por medio de la Concentración Mínima Inhibitoria (CMI) y la Concentración Mínima Bactericida (CMB).

Los datos obtenidos de la CMI y CMB fueron analizados mediante un análisis de varianza (ANOVA) y para determinar diferencias entre tratamientos se realizó una prueba de Tukey ($P < 0.05$), en el paquete estadístico SAS versión de 9.0.

Tabla. Concentración Mínima Inhibitoria y Concentración Mínima Bactericida del EHSb.

Bacteria	CMI EHSb (mg/mL)	CMB EHSb (mg/mL)
A. hydrophila	25^{c}	100c
L. anguillarum	1.56^{a}	3.12^{a}
E. tarda	3.12^{b}	25^{b}
S. iniae	25^{c}	25^{b}
Valor de P	0.0001	0.0001

EHSb: extracto hidroalcohólico de *Salix babylonica*, diferentes literales ^{a,b,c} en las columnas indican diferencias estadísticas significativas ($P \leq 0.05$).

Resultados: El EHSb tuvo la capacidad de inhibir el crecimiento de las cepas evaluadas. Existió diferencias estadísticas significativas (p=0.0001) entre los tratamientos. La mejor actividad inhibitoria del EHSb se presentó sobre *L. anguillarum* (1.56 mg/mL), seguida de *E. tarda* (3.12 mg/mL) y con menor actividad inhibitoria sobre *A. hydrophila* y *S. iniae* (25 mg/mL) (Tabla 1).

Los resultados obtenidos de la CMB indican que el EHSb tiene actividad bactericida contra las bacterias evaluadas. La mejor CMB se observó sobre *L. anguillarum* (3.12 mg/mL), seguido de *E. tarda* y *S. iniae* (25 mg/mL), la menor actividad sobre *A. hydrophila* (100 mg/mL) (Tabla 1). La relación entra la CMB y CMI indica que el EHsb presenta un efecto bactericida sobre las cepas evaluadas.

Conclusión: El EHSb presenta actividad antibacteriana sobre las cepas evaluadas, por lo tanto, podría ser una alternativa para el control y tratamiento de las bacterias evaluadas las cuales afectan al cultivo de tilapia y trucha arcoíris.

RECONCILING CLIMATE CHANGE AND AQUACULTURE

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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 facilitating research. Climate change vulnerability assessments is one method that triages threats needed to prioritize adaptation efforts. This approach has recently begun to see application in aquaculture. This presentation explores potential roles for climate change vulnerability assessments and the evolving needs of climate change and aquaculture research, which is crucial to support planned adaptation.

ALGINATE-BASED BEADS: ENCAPSULATION OF SAGE ESSENTIAL OIL BY ELECTROSTATIC EXTRUSION

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Recently, various techniques have been developed for encapsulating bioactive compounds to convert them into a more stable form. One of them is electrostatic extrusion, which is highly recommended for hydrophobic bioactive compounds such as essential oils (EOs). Among the various polymers, alginate is most commonly used as a core material. This is a naturally occurring polysaccharide found in brown algae and is characterized by high viscosity, gelling properties, high stability, low cost and non-toxicity. Sage (*Salvia officinalis* L.) is a perennial aromatic and medicinal plant of the Lamiaceae family. In addition to various phytochemicals, it also contains EOs which are found to have antioxidant, antimicrobial, antifungal, insecticidal and antiproliferative properties. Therefore, this study investigated the development of alginate-based beads containing sage EO prepared by electrostatic extrusion.

For the preparation of beads, the content of alginate (0.5, 1 and 1.5%, *w/v*) and calcium chloride ($CaCl_2$) (3 and 5%, *w/v*) was varied. Alginic acid sodium salt (low viscosity), previously dissolved in distilled water, sage EO (5%, *w/v*) and Tween 20 (0.5%, *w/v*) were homogenized at 10 000 rpm for 4 min. Beads were prepared with Büchi encapsulator B-390 (Flawil Switzerland) using a 1 mm stainless steel needle at a frequency of 80 Hz, a pressure of 1000 mbar, an amplitude of 6 and an electrostatic potential of 500 V. After collection in $CaCl_2$ solution, the beads were washed in distilled water, drained and used for further analysis. Encapsulation yield (%) was expressed as the ratio between the amount of beads obtained and the amount of emulsion used for the encapsulation. The size of the beads was measured using a micrometer screw gauge and the average d_{max}, d_{min} and sphericity factor (SF) were calculated.

All analyzed parameters were significantly affected by alginate and $CaCl_2$ content, except for SF (Table 1). The highest encapsulation yield and bead size were obtained at the highest concentrations of alginate and $CaCl_2$. In addition, the beads generally maintained their roundness. Although not significant, most spherical beads were produced with 1.5% alginate, while a lower $CaCl_2$ content resulted in slightly elongated shape of the beads.

Table 1. Influence of alginate and calcium chloride content (%) on encapsulation yield and beads size

Source of variation	Yield (%)	d_{min} (mm)	d_{max} (mm)	Sphericity factor
Alginate content (%)	p<0.001*	p=0.031*	p=0.023*	p=0.606
0.5	60.44±1.16^a	2.75±0.10^a	3.11±0.09^a	0.06±0.01^a
1	64.97±1.86^b	2.84±0.12^{ab}	3.27±0.11^{ab}	0.07±0.01^a
1.5	69.96±1.39^c	3.07±0.17^b	3.38±0.09^b	0.05±0.02^a
$CaCl_2$ content (%)	p<0.001*	p<0.001*	p<0.001*	p=0.071
3	62.66±1.69^a	2.64±0.06^a	3.08±0.07^a	0.08±0.01^a
5	67.58±1.86^b	3.13±0.09^b	3.43±0.05^b	0.05±0.01^a

Results are expressed as mean±standard error. Means in column with different letter were significantly different at p≤0.05.

Figure 1. Beads of sage EO prepared with 1% alginate and 5% calcium chloride

BUSINESS MODEL FOR THE MALASIAN PROWN *Macrobrachium rosenbergii* POSTLARVAE PRODUCTION IN SOTAVENTO, VERACRUZ, MEXICO

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In Sotavento and Grandes montañas regions of Veracruz, Mexico, the production of Malayan prawn *M. rosenbergii* has decreased due to the closure of the postlarvae production units, which has resulted in the loss of productivity, importation of the product and the increase in prices in local markets.

The recovery of the production chain lies in the supply of postlarvae, so the objective of this work is to determine an optimal business model to activate innovation in the production of Malaysian shrimp postlarvae.

The methodology used includes the identification of factors that have influenced the decrease in postlarvae production, the identification of the characteristics of the economic agent for optimal performance, the development of a business model.

As outstanding results, it was found that the causes of the decline in production are related to ineffective innovation programs, territorial lag, arbitrary financial support, low economic movement, low knowledge of how to operate production systems.

A proposal of the economic agent and a business model including productive technology and its respective financial analysis are presented.

AQUACULTURE AND FISHERIES FINANCE IN MEXICO. EXPERIENCES, PROJECTIONS AND PROPOSALS TO FINANCE AQUACULTURE

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This presentation covers 35 years of experience in the financial sector, with emphasis in the aquaculture and fisheries industries of Mexico. It provides information on who provides finance and analyses the investment risks, based on historic results, support from the government, private and development banking sectors, and the capacity to respond to the client´s needs.

Information on the industry development is provided, production volume, value and a review of the commercial balance. A brief discussion on some of the factors that have limited development of aquaculture in Mexico is attempted before offering some ideas to drive the industry, like the differentiation on criteria and policies needed for fisheries and aquaculture management; the dissemination and implementation of sustainable knowledge-based technologies; the implementation of strict certification processes; and the support of better academia-industry-government-finance interactions.

ALGINATE MICROCAPSULES AS DELIVERY AND PROTECTIVE SYSTEMS OF *Bacillus licheniformis* IN A SIMULATED SHRIMP'S DIGESTIVE TRACT

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In Mexico, the farming of shrimp and other marine species represents an important activity that provides profits for producers, is a good source of food with high nutritional value, and gives a job opportunity for skilled and unskilled workers. Therefore, new strategies that improve biosafety protocols are necessary today, to ensure an adequate control of diseases that limit shrimp production. The implementation of non-antibiotic agents, such as probiotics, has emerged as a promising strategy to enhance shrimp farming.

The evaluation of its controlled and targeted release within a simulated shrimp digestive tract (DT). The encapsulation process was carried out using the ionic gelation technique. Both free and bacteria-loaded AMPs were physicochemically characterized by size, morphology, surface electrical charge, the survival, and the number of encapsulated bacteria after the encapsulation process, and the bacterial survival after 40-days of storage (at 4 ◦C and 25 ◦C). The *in vitro* release and survival studies of the bacteria were carried out using a protocol developed in our laboratory by implementing buffers of dissected organs from shrimp's DT.

Results indicated that microparticles with an average size of 172–185 μm and negatively charged (- 16.77 and - 17.66 mV, respectively) were obtained after using the ionic gelation method (Fig. 1). The bacterial survival and encapsulation efficiency showed high cell viability and yield above 99%. Stability studies showed that the best storage temperature was 4 ◦C, in which it remained almost 100% of the bacteria viable for 15 days; however, cell viability declined to 55% survival after 30 days of storage at this temperature.

Regardless of the cell viability reduction after 30 days, there are enough viable bacteria cells to be considered as a probiotic product. Release and survival studies showed that alginate particles had a protective effect on bacteria by keeping *ca.* 51.29% of viable probiotic within the shrimp intestine; in contrast, free bacteria only reached the shrimp intestine *ca.* 27.16% viable (Fig. 2).

Our results suggest that microparticles can be produced by a low-cost method that could ultimately benefit shrimp farming in a near future.

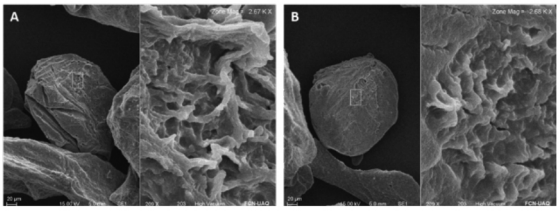

Figure 1. Scanning electron micrographs of free (A) and *Bacillus licheniformis*-loaded (B) alginate microparticles.

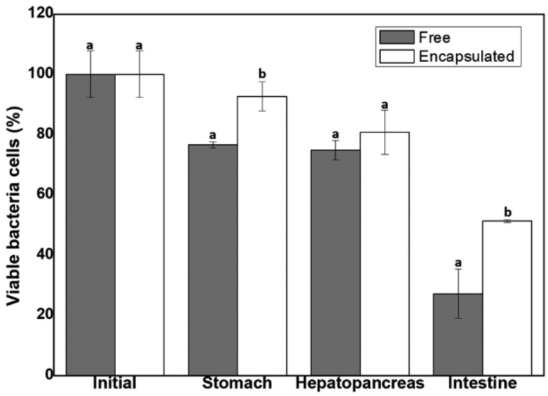

Figure 2. Percentage of viable bacteria (%, free or encapsulated in alginate microparticles) during their passage through a simulated shrimp's digestive tract. Results are expressed as the average ± standard error of at least 2 individual experiments with 3 repetitions. Different letters indicate significant difference ($p < 0.05$) between samples in each stage of the digestive tract analyzed.

SPATIAL PLANNING FORECASTS OPPORTUNITY FOR OFFSHORE AQUACULTURE

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The planning and siting of Aquaculture Opportunity Areas (AOA) in the U.S. Gulf of Mexico and the Southern California Bight is the first ever application of marine spatial planning in U.S. waters for the development of offshore aquaculture at this scale. The results of these analyses provide compelling evidence for the opportunities as well as challenges of siting offshore aquaculture in the coastal ocean within a reasonable range of the working waterfront.

Geospatial analysis for identification of AOA options was based on a categorical framework to ensure relevant, comprehensive data acquisition and characterization for spatial suitability modeling. An authoritative spatial data inventory was developed that included data layers relevant to administrative boundaries, national security (i.e., military), navigation and transportation, energy and industry infrastructure, commercial and recreational fishing, natural and cultural resources, and oceanography. With over 200 data layers included in each analysis, the maps, models, and descriptions provide the most comprehensive marine spatial modeling in the U.S. federal waters to date. Our methods and models can be used to significantly improve the next generation of marine spatial planning, contributing support far beyond aquaculture development by unleashing the power of spatial analytics for offshore energy exploration, shipping and navigation, national security and military strategy, identification of marine protected areas, and burgeoning sectors of the ocean economy.

PARÁMETROS GENÉTICOS PARA PESO CORPORAL Y RESISTENCIA AL SÍDROME DE MANCHA BLANCA EN CAMARÓN BLANCO DEL PACÍFICO UTILIZANDO INFORMACIÓN GENÓMICA

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La resistencia al virus del Síndrome de la Mancha Blanca (VSMB) y el peso corporal son criterios importantes en las evaluaciones genéticas en camaronicultura. La disponibilidad de microarreglos con miles de Nucleótidos de Polimorfismo Simple (SNPs) para *Litopenaeus vannamei* abre la oportunidad de mejorar las estimaciones de los parámetros genéticos al considerar la información genómica al mismo tiempo que la genealogía disponible; sin embargo, es importante evaluar su uso en la estimación de parámetros genéticos. El objetivo de este trabajo fue estimar los parámetros genéticos para la resistencia al VSMB y el peso a los 120 días en *Litopenaeus vannamei,* utilizando información genómica.

Se utilizó la información 176 familias de dos ciclos de la población de la empresa Maricultura del Pacífico ubicada en el noroeste de México. En 2020 se generó un desafío controlado para evaluar la resistencia al VSMB (inóculo *per os*, 10^6 partículas virales /g) en camarones de 75 días de edad, se obtuvo la supervivencia binaria (sSMB) de 6108 camarones a las 144 horas post inoculación y de ellos se genotiparon 1684. En 2021 se obtuvo el genotipo y peso a los 120 días (P120) de 824 individuos que crecieron en instalaciones controladas. Se incluyó la información del genotipo de los 429 progenitores de ambas generaciones. Los genotipos fueron obtenidos con el panel AquaArray HD (50 K) vannamei® (Neogen®), que contiene 50,811 SNPs. En el control de calidad se usó una tasa de llamado para SNP de 80%, frecuencia de alelo mayor menor a 0.01 y equilibrio de Hardy-Weinberg con p-value < $1x\ 10^{-6}$. Quedando 35,258 SNPs y 3,000 individuos. Para la estimación de heredabilidad de P120 días y sSMB y la correlación genética entre ellas, se usaron don enfoques: Un modelo animal tradicional (BLUP) y otro incluyendo la matriz de información genómica (ssGBLUP). En ambos se consideraron como efectos fijos para la sSMB la tina de prueba y el peso al registro, mientras que para P120 el sexo y la edad, además del efecto común de familia. Se utilizó el programa AIREMLF90.

Como se observa en el Cuadro 1, con el enfoque de ssGBLUP las estimaciones de heredabilidad se reducen los errores estándar en ambas características, lo que mejora su precisión. En la r_G fue consistentemente negativa y no significativa en ambos modelos, como se ha observado en otros estudios entre peso corporal e indicadores de resistencia a SMB.

Cuadro 1. Heredabilidades, correlaciones genéticas y efecto de familia para peso a los 120 días y supervivencia a Síndrome de Mancha Blanca en *Litopenaeus vannamei*

Modelo	Característica	P120	sSMB	f^2
BLUP	**P120**	**0.28±0.12**	-0.38±0.76	0.05±0.05
ssGBLUP		**0.20±0.07**	-0.37±0.41	0.08±0.04
BLUP	**sSMB**		**0.07±0.04**	0.04±0.01
ssGBLUP			**0.10±0.03**	0.08±0.04

Heredabilidades en la diagonal, las correlaciones genéticas sobre la diagonal. **BLUP**= Modelo con pedigrí, **ssGBLUP=** Modelo con pedigrí e información genómica, f^2 = efectos comunes de familia

PARÁMETROS GENÉTICOS DE CARACTERÍSTICAS MORFOMÉTRICAS ASOCIADAS A LA BELLEZA DEL BARBO SUMATRANO *(Puntius tetrazona)*

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En la acuicultura ornamental el objetivo de los programas de mejoramiento genético es generar biomasa con buena calidad estética. Existe poca información sobre parámetros genéticos de características morfométricas asociados a la belleza en peces ornamentales. Por lo que, el poder disponer de parámetros genéticos de este tipo de características permitirá definir con mayor precisión los criterios de selección asociados a la calidad estética en el diseño de programas de mejoramiento genético en *Puntius tetrazona*. El objetivo de este estudio fue estimar parámetros genéticos de características morfométricas asociadas a la belleza en *Puntius tetrazona*.

Se formaron 77 familias de 3 variedades de *P. tetrazona*, se mantuvo la identificación familiar. A partir de fotografías digitales de 296 organismos, se obtuvieron las medidas en función de una líneas de referencia trazadas sobre la imagen de cada individuo (Figura 1). Los componentes de varianza se estimaron usando un modelo animal univariado, donde se consideraron como efectos fijos la densidad, el largo patrón y la variedad y como aleatorios el efecto de animal y acuario.

Las medias (D.E.) para LP y PC fueron 18.9 (4.8) mm 3.0 (0.8) mm; para el LVC= 7.2 (2.0) mm; y para LPA y LDA, 2.6 (0.9) mm y 3.1 (1.2) mm, respectivamente. Los valores de h^2 de las medidas corporales son altos (Cuadro 1) como en peces de consumo. El valor de h^2 de las aletas puede relacionarse a que la heterogeneidad en el ambiente de crecimiento aporta variación en el tamaño de las aletas. La correlacion de valores geneticos aditivos (r_{VGA}) entre LP y PC está dentro del rango de los valores de correlaciones genéticas entre características de crecimiento de otras especies. La asociación de estas características con las de aletas fue negativa, posiblemente relacionado a que los organismos tengan que compensar el largo del PC con aletas de mayor tamaño. Existe variación genética aditiva en las características analizadas en este estudio y asociaciones entre características. Sin embargo, es importante considerar las preferencias del mercado para establecer la dirección de los criterios de selección asociados a la belleza de *P. tetrazona*.

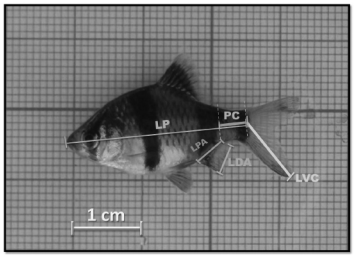

Figura 1 PC= Longitud del pedúnculo caudal; LVC= L. ventral de la aleta caudal; LDA= L. distal de la aleta anal; LPA= L. proximal de la aleta anal.

Cuadro 1. Heredabilidades (h^2), proporción de efecto del acuario (a^2), correlaciones de valores genéticos aditivos predichos y fenotípicas (± error estándar) para características asociadas a la belleza de Puntius tetrazona

Característica	*LP*	*PC*	*LVC*	*LPA*	*LDA*	a^2
LP	**0.73± 0.23**	0.76± 0.02	-0.02± 0.05	-0.03± 0.05	-0.04± 0.05	0.16± 0.09
PC	0.77± 0.02	**0.58± 0. 24**	-0.33± 0.04	-0.38± 0.04	-0.12± 0.05	0.37± 0.12
LVC	0.94± 0.01	0.63± 0.03	**0.14± 0.20**	0.40±0.04	0.43± 0.04	0.28± 0.11
LPA	0.82± 0.02	0.50± 0.04	0.81± 0.02	**0.06± 0.14**	0.67± 0.02	0.16± 0.09
LDA	0.66± 0.03	0.53± 0.04	0.66± 0.03	0.78± 0.02	**0.11± 0.12**	0.08± 0.08

Heredabilidades en la diagonal, las correlaciones de valores genéticos aditivos predichos sobre la diagonal y las correlaciones fenotípicas debajo de la diagonal.

RESEARCH AND DEVELOPMENT OF TROPICAL SEAWEED AQUACULTURE TECHNIQUES AND BEST MANAGEMENT PRACTICES ACROSS MULTIPLE SITES IN THE CARIBBEAN

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The Caribbean's small countries and island nations are experiencing a loss of resources due to climate change, nutrient pollution, ocean acidification, seagrass bed habitat loss, fishing pressure, and lost tourism revenues due to COVID-19. Well-managed development and growth of tropical seaweed aquaculture in the region may help to assuage these issues while also providing a new source of seaweed biomass for the existing carrageenan, new food and textile and possibly future biofuel markets. We are exploring the opportunities for expanded seaweed aquaculture in the Caribbean and the Gulf of Mexico in collaboration with partners at 15 institutions and research sites in Puerto Rico, Florida, and Belize. Together we are prototyping cultivation systems that allow farms to be deployed in offshore areas, creating tools to mechanize seeding and harvesting, assessing the environmental impacts of these farm systems, characterizing the growth and composition of tropical algae in near and offshore environments, and conducting economic and life cycle analyses of macroalgal aquaculture systems in this region. Findings from these efforts will be tailored to the conditions in the Caribbean and The Gulf of Mexico and be adaptable for other locations with similar environmental threats or needs for alternative marine livelihoods.

In this presentation we will explain how insight from our field research is being combined with consultation from local stakeholders to guide the development of Ecological Best Management Practices (BMP) for seaweed aquaculture in the Gulf of Mexico and the Caribbean Sea. Our intention is that these ecological BMPs will support prospective seaweed farmers, resource managers, and buyers through ecologically and socially responsible decision-making around seaweed aquaculture. Fostering sustainable expansion of Caribbean and Gulf of Mexico macroalgal cultivation will encourage production in these regions to expand beyond the existing small-scale farms producing seaweed for artisanal beverage and cosmeceutical markets. Establishing larger-scale farms in a responsible manner will enable production of algal biomass for additional local and global markets and allow resource managers to consider seaweed farming as an extractive component of an integrated water quality management strategy.

One of our candidate species, *Eucheumatopsis isiformis*, which is native to the region.

RELATIONSHIP BETWEEN ALPHA DIVERSITY AND RHODOBACTERACEAE VIBRIONALES RATIO AS AN INDICATOR OF GUT HEALTH OF WHITELEG SHRIMP *Litopenaeus vannamei*

Ana Rodiles[1]*, Florian Geay[2], Eric Leclercq[1], Stephane Ralite[1] and Mathieu Castex[1]

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The use of the 16S rRNA marker genes is gaining *momentum* as a practical tool to appraise the role of intestinal microbiota in shrimp culture. High-throughput sequencing produces an enormous amount of data and results are often expressed by the relative abundance of bacteria taxa, however this approach introduces a bias in the absence of microbial load quantification. Latest advance in managing 16S sequencing data contemplates the log ratio between bacteria, which automatically cancels this bias, by comparing the ratios of different taxa between samples instead of the relative abundance of individual taxa (Morton et al., 2019). We applied this approach to shrimp microbiota data sets by looking at the relationship between alpha diversity and the ratio between two important taxa in the shrimp intestinal microbiota: the family Rhodobacteraceae and the class Vibrionales. Indeed Rhodobacteraceae includes many beneficial bacteria species while Vibrionales contains well-known opportunist and pathogenic species, suggesting the Rhodobacteraceae/Vibrionales (R/V) ratio as a potential biomarker of gut microbial health and welfare in shrimp.

Data were compiled from 4 independent pond trials performed in Vietnam over the grows-out phase, hence covering a variety of dietary, environmental and sanitary conditions. A total of 196 hindgut samples were included with the same molecular approach applied for DNA extraction, PCR primers and bioinformatics processing.

High-throughput sequencing of the 16S rRNA V3-V4 region was analysed with DADA2 independently per trial, then Shannon diversity index and ratio Rhodobacteraceae/Vibrionales values were computed to test a potential correlation on QIIME2 (Bolyen et al., 2019), a python-based microbiome platform.

Results showed a highly significant correlation between the R/V log ratio and Shannon alpha diversity index ($r = 0.76$; $P = 0.000$; Fig. 1). R/V ratio showed a wider range of 15 (-9 to 6; min max) showed a wider range of 15 (-9 to 6; min max) compared to 7 for shannon (1 to 8). Interestingly,

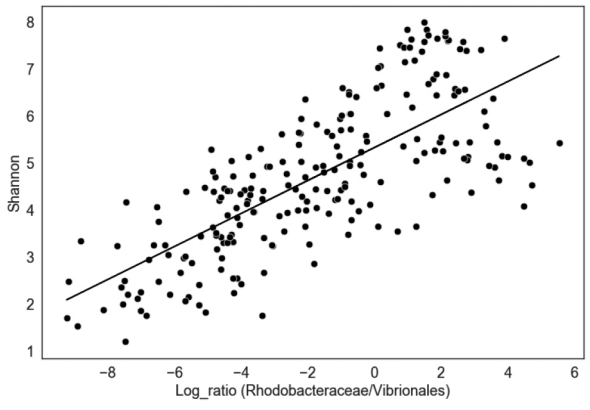

Fig. 1. Spearman correlation of Shannon vs R/V log ratio in shrimp hindgut samples across the all datasets Each dot represents 1 sample.

EFECTO DE LA TEMPERATURA Y SALINIDAD EN EL DESARROLLO EMBRIONARIO DEL PULPO *Octopus insularis* Leite & Haimovici 2008

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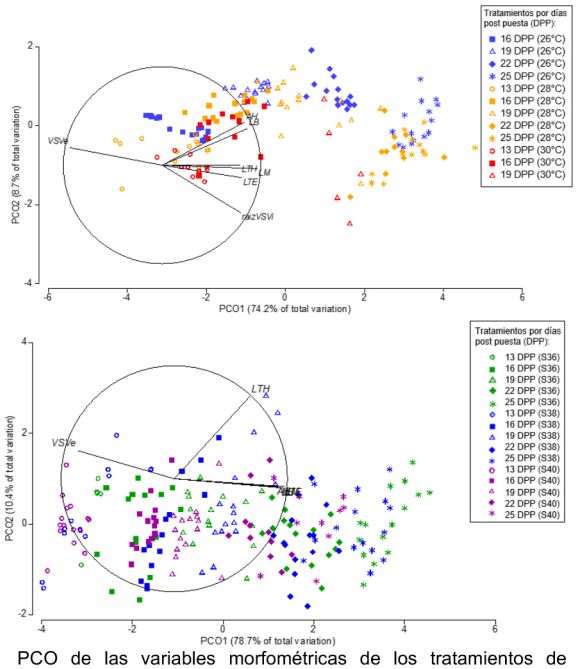
El desarrollo de los cefalópodos se encuentra influenciado por variables ambientales como la temperatura y salinidad. *Octopus insularis* es un pulpo de reciente descripción que presenta gran abundancia en las pesquerías del suroeste del Golfo de México, por lo que, presenta una nueva oportunidad de cultivo de pulpo para la región, sin embargo, se desconocen los efectos que tiene la temperatura y salinidad en las primeras etapas del desarrollo embrionario.

Cordones de huevos de *O. insularis* fueron incubados artificialmente a temperaturas de 26, 28 y 30°C y salinidades de 36, 38 y 40. En cada tratamiento, el estadio de desarrollo Naef (1928), morfometría embrionaria y tiempo de desarrollo embrionario fueron medidos. Se realizó un gráfico PCO y un PERMANOVA con ayuda del programa estadístico PRIMER-e.

La temperatura afectó de manera inversamente proporcional al tiempo de desarrollo embrionario. T26 fue 6 días más largo que T28, siendo este el tiempo más corto (31 días). S36 y S38 obtuvieron un tiempo de desarrollo de 33 días. Mientras que, T30 y S40 generaron un efecto deletéreo. Malformaciones en embriones fueron observadas en T30 y, S38 y S40.

El PCO1 del gráfico de temperatura explicó el 74.2% de la variación, principalmente por el ancho del huevo, longitud del manto, longitud del brazo y volumen del saco vitelino. El cambió en la morfometría fue diferente a lo largo del tiempo dependiendo del tratamiento térmico (p<0.05). T30 modificó el cambio en la morfometría de los embriones, retrasando el aumento en todas las variables medidas, en especial el largo del brazo y el ancho del huevo.

El PCO1 de salinidad explicó el 78.67% de la variación, principalmente por el ancho del huevo, longitud total del embrión, longitud del manto, longitud del brazo y volumen del saco vitelino. La morfometría cambió a lo largo del tiempo, pero lo hizo de la misma manera para todos los tratamientos de salinidad. En general, los embriones de S40 tuvieron una morfometría diferente a lo largo del experimento en comparación al resto. (p<0.05).



PCO de las variables morfométricas de los tratamientos de temperatura (superior) y salinidad (inferior) por días post puesta.

PERMANOVA de los tratamientos de temperatura.						
Source	**df**	**SS**	**MS**	**Pseudo-F**	**P(perm)**	**Unique perms**
Tr	2	91.545	45.773	10.553	0.0001	9942
Di	4	705.99	176.5	39.839	0.0001	9961
TrxDi	5	58.735	11.747	2.7769	0.0006	9915

PERMANOVA de los tratamientos de salinidad						
Source	**df**	**SS**	**MS**	**Pseudo-F**	**P(perm)**	**Unique perms**
Tr	2	84.349	42.174	10.266	0.0001	9954
Di	4	839.19	209.8	51.204	0.0001	9950
TrxDi	8	27.314	3.4143	0.84803	0.6571	9905

Modificaciones en el tiempo del desarrollo por temperatura han sido observados en otros pulpos (*O. vulgaris, maya, mimus, tehuelchus,* etc.). Así mismo, temperaturas fuera del rango óptimo de cada especie han reportado bajas fecundidades, tasas de eclosión e interrupción del desarrollo. Elevadas salinidades confirman la interrupción del desarrollo embrionario y generación de malformaciones, pero no efecto en el tiempo de desarrollo. Efectos similares han sido reportadas en sepias y calamares, pero no en pulpos.

DISMINUCIÓN DE LA EFICIENCIA DE MASCULINIZACIÓN EN CRÍAS TILAPIA DEL NILO *Oreochromis niloticus* CON 17 α-METILTESTOSTERONA EN COMBINACIÓN CON EXTRACTOS ACUOSOS DE LA MACROALGA *Padina durvillaei*

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Esta descrito que la exposición al agente andrógeno utilizado durante el proceso de producción de crías de tilapia 100% masculinizadas influye negativamente en la respuesta inmune no específica del pez, por lo que es relevante buscar compuestos inmunoestimulantes factibles de utilizar durante este proceso. El presente trabajo muestra los resultados obtenidos de un bioensayo del proceso de masculinización de crías de *Oreochromis niloticus*, utilizando un alimento hormonado (60 mg/kg de 17 α-metiltestosterona) suplementado con extracto del alga *Padina durvillaei*. Se evaluó el crecimiento, la supervivencia, los parámetros productivos y el porcentaje final de machos de los peces sometidos a tres tratamientos con una inclusión del extracto del 0.5% (E1), 1.5% (E2) y 3% (E3), un control positivo (C+, alimento hormonado sin extracto) y un control negativo (C-, alimento sin hormona y extracto), durante un periodo de 28 días. No se encontraron diferencias significativas en el crecimiento (peso y longitud), y supervivencia entre los distintos tratamientos y los controles. No obstante, los valores obtenidos en el porcentaje final de machos presentaron diferencias significativas, siendo el control positivo (alimento con hormona sin extracto) en el que se obtuvo la masculinización del total de los organismos de prueba, observando una disminución del porcentaje de machos del 11% al 13%, por efecto de la suplementación con el extracto de alga, siendo el alimento suplementado al 3%, el que presentó la menor masculinización (87%). Esta disminución en la tasa de masculinización podría deberse a una competencia por el receptor esteroideo, entre el andrógeno y algunos de los compuestos fitoquímicos (fenoles y flavonoides) presentes en el extracto con afinidad por dicho receptor. Se concluyó que la inclusión del extracto no causa alteración en la supervivencia final, pero si disminuye la tasa de masculinización, por lo que se requiere profundizar sobre el efecto como protector de disruptores endocrinos por parte de compuestos presentes en esta macroalga.

EFECTOS POSITIVOS DE LA APLICACIÓN CONTINUA DE PROBIÓTICOS COMERCIALES PARA EL MANEJO DE TILAPIA ROJA CULTIVADA EN AGUA DE MAR CON BIOFLOC

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Las tilapias rojas (*Oreochromis* sp.) presentan las mejores características favorables de adaptación a las altas salinidades (0 a 35 ‰) pero se conoce poco de su respuesta fisiológica y de crecimiento durante su producción en sistemas marinos con biofloc inoculados con probióticos comerciales. Para tal fin se desarrollaron dos experimentos incluyendo la evaluación de la respuesta fisiológica digestiva y crecimiento de juveniles híbridos de tilapia roja cultivadas en agua de mar a dos densidades de cultivo (D1, 120 peces/m3; D2, 240 peces/m3) y sistemas biofloc con o sin inóculos bacterianos y la evaluación de la respuesta metabólica hepática y de la respuesta antioxidante con biofloc y alimentada con 25%, 35% y 40% de proteína cruda (PC) en dietas comerciales (P25, P35 y P40, respectivamente) a una densidad de 80 peces/m3. Se presentarán los resultados en cuanto a parámetros zootécnicos y la información recabada sobre las diferencias en la actividad de enzimas digestivas (actividad proteolítica alcalina total, tripsina, quimotripsina, lipasa y amilasa) así como también cambios histológicos en las vellosidades intestinales y conteo de células caliciformes para la primera prueba. Para el segundo experimento se presentarán los datos sobre la respuesta de la actividad de enzimas clave del metabolismo hepático (glucoquinasa, alanina aminotransferasa, fructosa-1,6-bifosfatasa, glucosa-6-fosfato deshidrogenasa (G6PDH) y ácido graso sintasas), de la actividad de enzimas antioxidantes en hígado y músculo, en particular de la catalasa y G6PDH, así como de los daños en esos tejidos por lipoperoxidación a través de los valores de malonaldehído.

APPLICATION OF SIZE EXCLUSION CHROMATOGRAPHY IN POLYSACCHARIDES ANALYSIS OF ADRIATIC SEA MACROALGAE *Acetabularia acetabulum AND Padina pavonica*

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Bioactive polysaccharides are valuable molecules that have been highly used in biomedical and pharmaceutical applications. Extensive research has been done to ensure their efficient extraction from marine algae using specific techniques to isolate and purify them. Marine macroalgae contain various polysaccharides such as frame polysaccharides, mucopolysaccharides, and storage polysaccharides.

In our study we used common extraction methods, which include various steps of pretreatment, using solvents for extraction, precipitation, and size exclusion chromatography (SEC). Pretreatments are necessary to remove chlorophylls, mannitol, salts, and other small compounds. Dry algal powder was suspended in ethanol, and the mixture was continuously stirred with a mechanical stirrer at room temperature. After filtration the same procedures were applied using acetone and dichloromethane. The residual material was then extracted with 0.1 M HCl (60 °C, 2 hours), centrifuged at 4500 rpm for 40 minutes, and the supernatant precipitated with ethanol (overnight, 2-8 °C). The precipitated polysaccharides were filtered-off, washed with ethanol and acetone, air-dried and analysed.

The molecular weights were determined by HPLC on Agilent 1260 Infinity II instrument, equipped with a refractive indeks detector G7162A. Analysis was performed using a PL aquagel-OH size-exclusion column (8 μm, 300 x 7,5 mm) and a PL Aquagel-OH Guard (8 μm, 50 x 7,5 mm) from Agilent Technologies. The column was calibrated with pullulan polysaccharide calibration kit (MW 180-700 kDa, Agilent) and a standard curve was then established.

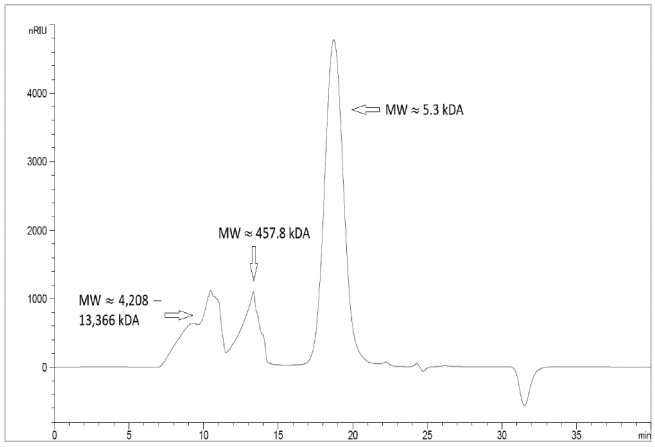

Figure 1. Chromatogram of *Acetabularia acetabulum*

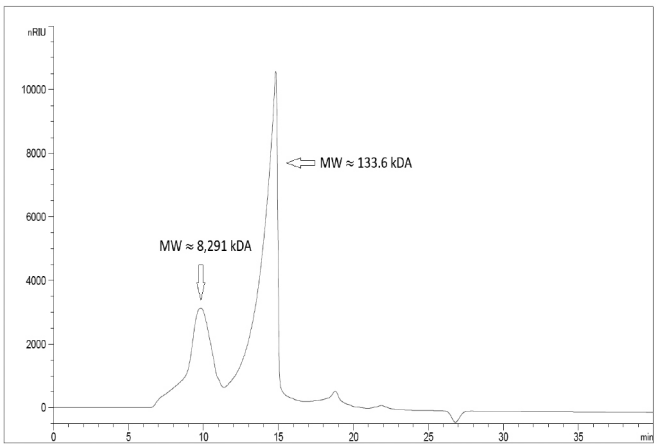

Figure 2. Chromatogram of *Padina pavonica*

SHORT-TERM MOVEMENTS AND USE OF ARTIFICIAL SHELTERS BY SEA CUCUMBER *Isostichopus badionotus* (SELENKA, 1867)

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The sea cucumber *Isostichopus badionotus* has been overfished in the Yucatan Peninsula. Due to this, population restoration actions have been proposed, such as the translocation of wild individuals or the release of captive-bred juveniles in reserve or non-fishing areas. To determine the optimal size of the reserve areas, it is necessary to know the potential dispersal of individuals throughout their life cycle, which is generally based on studies of the movement patterns and short-term dispersal rates. The objective of this work was to estimate the movement patterns and dispersal rates of *I. badionotus* to provide key knowledge for the design of protection zones for the species.

A set of experiments were carried out under semi-controlled conditions, in external ponds with 50% sun protection and natural fluctuation of temperature and solar radiation. Fifty-five organisms classified as small, medium and large were used to determine the pattern and magnitude of movement in daily cycles, as well as its relationship with temperature and solar radiation. The preference for the use of colonized and non-colonized artificial refuges was also evaluated. The movement rates were obtained using a camera located in a zenithal position above the experimental tank. Differences in displacement were identified between large and small cucumbers (ANOVA, $p = 0.0252$, Tukey HSD). The average daily movement for the size class was: 518 cm for small, 615 cm for medium sizes and 688 cm for large sizes (Table 1). The displacement pattern was characterized by a very low displacement in the mornings (07:00 to 14:00), a maximum in the afternoon (16:00 to 22:00) and intermediate at night (23:00 to 5:00). A positive linear relationship was determined between organism size (estimated length: eL) and total daily displacement (D) ($D = 2.88\ eL + 22.27$ cm). Positive correlations (Spearman: r_s) were obtained between the displacement of large and medium organisms with temperature (rs: 0.48, 0.89) while, negative r_s values were observed between displacement and light intensity in small organisms (r_s: - 0.44, - 0.74). Higher percentage of use of refuge by small organisms (53% of the daily time) and greater affinity for colonized refuges (80% of refuge use) were observed. Medium and large organisms did not presented shelter preferences (<20% of the daily time). This demonstrates the importance of shelters. Daily movements found in this work are similar to other sea cucumbers species, being most active in the afternoon and at night, and provides specific data for long dispersion models for *I. badionotus.*

Table 1. Statistic summary of total daily displacement, measures in centimeters.

Class	n	Mean	Sd	Median	Range	Se
Small	15	518	210	513	648	54.2
Medium	15	615	138	630	531	35.7
Large	15	688	185	655	779	47.8

n number of data, Sd standard deviation, Se standard error.

INTENSIFIED CULTIVATION OF THE MACROPHYTIC RED ALGA *Gracilaria vermiculophylla* ON POROUS MESH PANELS IN RACEWAY PONDS

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New approaches are needed to automate and intensify the cultivation of commercially important red macroalgae. Using cell and tissue culture techniques, we have developed clonal cultures for the macrophytic red alga *Gracilaria vermiculophylla* that can be vegetatively propagated, mechanically blended into fragments, and then fluidically injected onto a mesh support. This alga has high carbohydrate content and exhibits broad temperature, salinity, and pH tolerance. Panels of *G. vermiculophylla* clonal tissue were stacked upright in dense array within a circular raceway pond, where fluid flowed over stationary mats of tissue (*Fig. 1*). Since the macroalgae grew as a dense, porous mat (*Fig. 2*), biomass loading was densified within the cultivation control volume, leading to enhanced areal productivity. Cultivation performance was assessed in Microbio Engineering 100 L (0.5RWi) raceways in both laboratory controlled and greenhouse environments under nutrient replete conditions. Laboratory raceway cultivations were carried out at 22 ^{o}C, 8000 ppm CO_2 in air at 0.010 L air L^{-1}culture min^{-1}, and 800 mmol photons $m^{-2}s^{-1}$ (16:8 photoperiod) with 8 cm panel spacing and 15 cm/s fluid velocity. At these conditions, the *G. vermculophylla* tissue fragments proliferated over the panel surface, ultimately creating a loose mat of tissue nearly 8-10 cm in thickness with biomass loading exceeding 10 kg FW per m^2 of panel surface over the 24 day grow out period. The cumulative areal productivity was 58 ± 9 g AFDW m^{-2} day^{-1} and final biomass density of 7 g AFDW L^{-1} (*Fig. 3*). Overall, dense arrays of stationary *G. vermiculophylla* panels cultivated under defined current flow in open raceways offer future potential for high productivity and modular deployment for aquaculture applications in land-based or open-water environments.

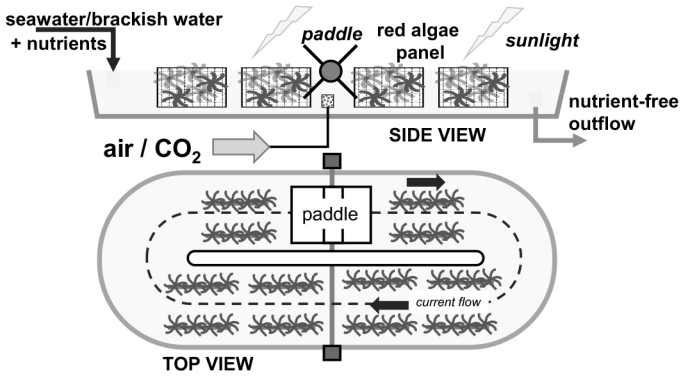

Fig. 1. Raceway with panel arrays.

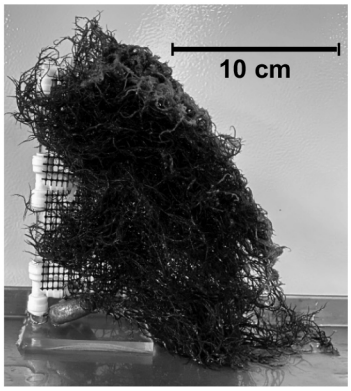

Fig. 2. G. vermculophylla panel.

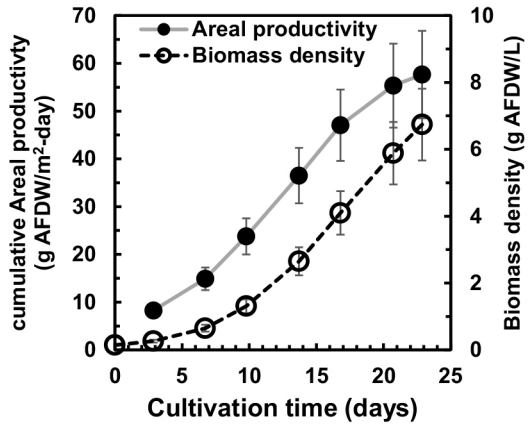

Fig 3. Biomass productivity.

EMBRYO QUALITY IN THE VOLUNTARY SPAWNING OF FIRST-GENERATION (F1) BARRED SAND BASS *Paralabrax nebulifer* KEPT IN CAPTIVITY

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Closing the cycle of commercially important fish in a recirculating aquaculture system (RAS) is growing in importance for stablishing culture protocols. From wild broodstock, it is possible to lab-produce juveniles. Subsequently, the juveniles are taken systems for grow-out and gonadal conditioning is implemented along with a controlled environmental, which facilitates the process of maturation, ovulation and voluntary spawning in captivity. In this investigation, a broodstock was established from juveniles produced in the laboratory (F1) of barred sand bass *Paralabrax nebulifer* with the use of RAS and voluntary spawning quality obtained under controlled conditions was evaluated.

The juveniles grow-out and gonadal conditioning is implemented with the photoperiod and temperature control (13:11 L:D; 22 ºC) in a RAS, which facilities the voluntary spawning in captivity. The broodstock group was composed of 6 females:4 males. To assess the quality of spawnings, the following parameters were determinated: volume of floating and non-floating embryos (Figure 1), embryo diameter, oil globule diameter, notochordal length (NL), yolk sac volume, hatching rate and survival percentage. The embryos/larvae were observed daily from day 0 to 3 days after hatching (DAH). The total number of spawns obtained each month was compared by means of a one-way analysis of variance. Statistical analysis was performed with the software Statistic 10.0.

The highest volumes of floating embryos were observed during April to June. In the embryos, the mean diameter was 0.88 ± 0.02 mm. The hatching percentage between the evaluation months did not show significant differences and had a mean value of 73.4 %, while the average survival percentage was 32.4 %. In the eleutheroembryos and apterolarvae, there were not significant differences in the mean NL at 1 and 3 DAH and it was 1.64 ± 0.24 mm and 2.16 ± 0.19 mm respectively.

Although larval survival is still a factor to consider, being able to conclude the life cycle and have reproductive availability in the captivity offers a significant step for to achieve the cultivation of this species.

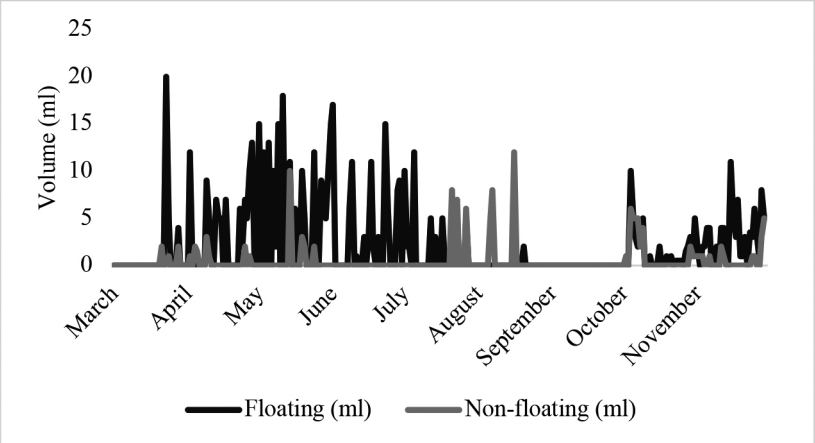

Figure 1. Volume of floating and non-floating embryos of broodstock F1 barred sand bass *Paralabrax nebulifer* subject to photoperiod and temperature control from March to November 2021.

EFFECT OF WATER TEMPERATURE ON EMBRYONIC DEVELOPMENT AND HATCHING TIME OF THE BARRED SAND BASS *Paralabrax nebulifer* (PERCIFORMES: SERRANIDAE)

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In recent decades, aquaculture has been one of the highest growing food production sectors from an animal source. The barred sand bass *Paralabrax nebulifer*, is one of the most important sources of small-scale commercial fisheries which represents the largest economic stream for cooperatives in Baja California Sur. Being a species of great fishery importance, it can be said that this species can also be of aquacultural importance. Being temperature a primary factor in the survival and growth of many aquatic organisms, the intention is to evaluate and determine an adequate temperature at which a better survival of embryos during incubation of the barred sand bass can be observed.

Eggs were obtained by voluntary spawning, the incubation of embryos were carried out at controlled water temperatures (23, 25, 27, and 29 ºC). To determine the optimal incubation temperature a system was constructed consisting of 4 incubators each at different temperatures. Four steel inserts were used to attain the water temperature of 23 °C, this was regulated through the control of ambient temperature with the help of an air-conditioning mini-Split in automatic mode. We used resistance immersion heaters to increase the temperature to 25, 27, and 29 °C. The incubators were maintained in constant aeration for a better dispersion of temperature throughout the water. The embryos were randomly selected and transferred in 24-well microtiter plates (one embryo per well). Each well was filled with 3.0 ml UV-sterilized sea water and were taken to the incubation area, each temperature treatment was replicated three times. During the incubation, the photoperiod (13:11 L:O) was kept up with neon lights. To ensure accurate measurements of the time intervals, the microtiter plates were examined every 6 hours to determine the hatching status and health of the embryos and larvae, for which a dissecting microscope was used. To obtain the LD50 (average lethal dose), after which 50% of the organisms died, a binary logistic regression "probit" was applied using the Minitab program.

According to survival probality program the LD50 was 22.4 °C (Figure). This indicates that 50% of the embryos survived to this temperature. The incubation period is particularly sensitive to extreme temperatures within the tested temperature range.

From a probabilistic perspective, the binary logistic model revels that at 22.4 degrees, an organism has a 50% probability of survival.

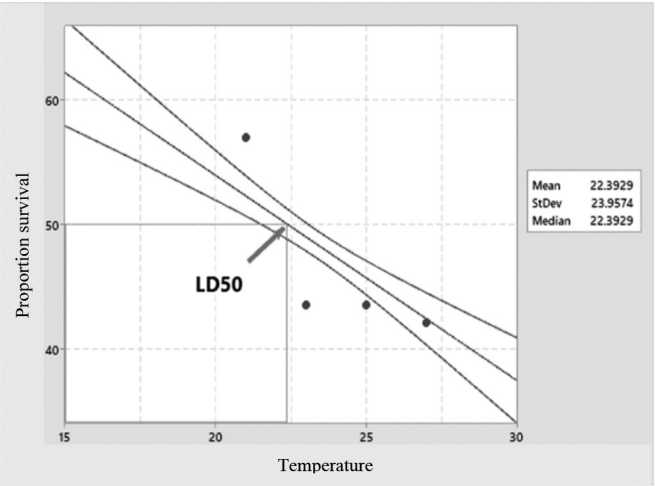

Figure. Effect of acclimation on mortality response to temperature for barred sand bass. Proportion survival and 95% confidence interval estimated using the Survival probability model.

LA ACUACULTURA EN YUCATÁN: UNA PROPUESTA QUE ARTICULA A LOS SECTORES SOCIALES, ACADEMICOS Y GUBERNAMENTALES

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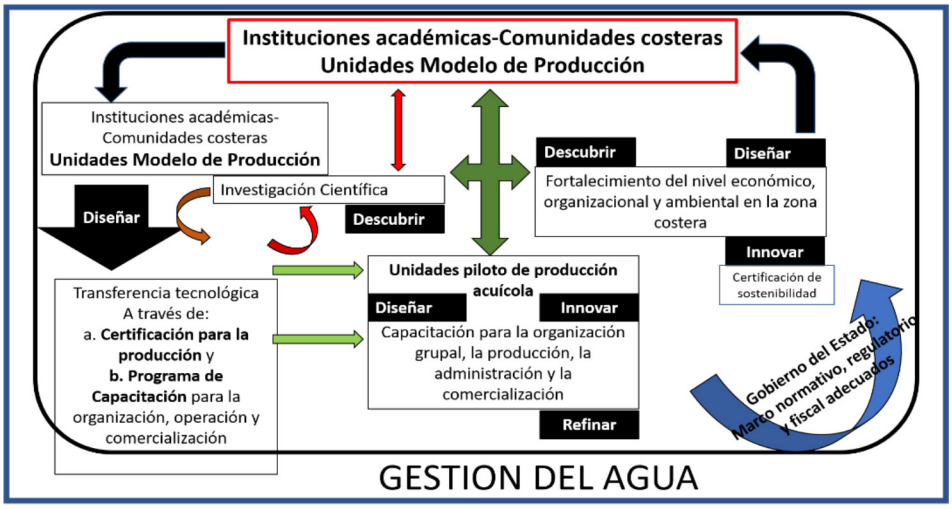
A partir de la articulación del sector académico, gubernamental y social, se desarrolló un proyecto para impulsar la actividad acuícola en el estado de Yucatán. A través de conocer la percepción de las comunidades costeras del estado se diseñó un esquema que aprovecha la infraestructura que se encuentra en las instituciones de investigación en el que las comunidades costeras y las instituciones académicas entienden que a través de la articulación se pueden desarrollar proyectos productivos con base en la transferencia tecnológica y la capacitación, ambos en un esquema participativo. Las Instituciones académicas cuentan con las Unidades Modelo de Producción en donde las personas interesadas en la producción acuícola se certifican, al mismo tiempo que se capacitan en otros aspectos complementarios a la organización, administración y mercadeo de los productos acuícolas. Como resultado, se generan proyectos piloto-productivos en donde se generan nuevas preguntas para la investigación científica, la cual permite descubrir, diseñar, refinar e innovar, tanto en las unidades modelo de producción, en el sector académico y en las comunidades costeras cuyos saberes se fortalecen en ese circuito de enseñanza aprendizaje-innovación. El Gobierno impulsa un marco normativo, regulatorio y fiscal que estimula la producción acuícola y facilita la generación de proyectos sustentables, al mismo tiempo que las unidades piloto se insertan en una correcta gestión del agua.



Octopus maya AQUACULTURE: LOOKING FOR PRODUCTS FOR GOURMET MARKET

Carlos Rosas Vázquez, Daniel Méndez, Antonio Cob Reyes, Adriano Cob, Geni Huicab, Juana de la Cruz Ek, Julio Sierra and Silvia Canul Pardenilla and Angel Yunes

UMDI-Sisal, Faculty of Sciences, UNAM and the Mayab Mollusc Fishery Production Cooperative Society Yucatan

Pre-commercial Scaling up for production

Project description: In recent decades, particular emphasis has been placed on the diversification of aquaculture, especially considering the species that have aquaculture potential in each region of the world. In the case of Mexico and particularly in the Yucatan Peninsula, the Octopus maya is a species with a high commercial value whose biological characteristics favour the development of farming methods. Although females spawn only once in their lives, they produce between 800 and 1,500 eggs per spawning, with a hatching rate of between 75 and 80%. One of the advantages of this species is that the newly hatched juveniles are benthic, which feed on live prey, dead, fresh, frozen, and even balanced food.

The Faculty of Sciences (UNAM-Sisal Yucatán) has the capacity to the production of 20 thousand embryos/month, artificially incubated. The juveniles are placed in outside ponds at a density of 10 octopuses/m^2, where they are fed at 100% of their biomass until they reach 2 to 3 g. Subsequently, the octopuses are separated by size to avoid cannibalism and fed at a rate of 50% of their weight. Survival of 50% can be obtained with growth rates ranging between 4 and 6% day^{-1}.

At present, the Science Faculty of the UNAM has established a collaboration agreement with the SCPP Molluscos del Mayab to start a farm for the production of O. maya juveniles with a maximum weight of 150g. This farm will be the first of its kind, with the primary objective of conducting a bio-economic study to establish production costs in this pre-commercial stage. Soon, the Cooperative will need to be injected with capital to build the reproduction, incubation, and pre-growth modules. With that capital, it will be possible to reach a pre-commercial octopus farm. At the same time, the installation of a food production plant is planned that serves to feed the octopuses and takes advantage of the waste from the fishing industry in the region.

MITIGATION OF SOYBEAN MEAL-INDUCED ENTERITIS IN RAINBOW TROUT *Oncorhynchus mykiss* USING BILE SALT DIETARY SUPPLEMENTATION

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The replacement of fish meal (FM) in aquafeeds by alternative sources of protein such as plant ingredients like soybean meal (SBM) has typically encountered a variety of challenges at high levels of inclusion, especially in carnivorous species like rainbow trout (*Oncorhynchus mykiss*). The development of intestinal inflammation, in this context referred to as soybean meal-induced enteritis, is a noteworthy sign of reduced tolerance to SBM, and there is an array of approaches that have been explored to alleviate inflammation. Bile salts (BS) have recently been suggested to have a role in immune signaling regulation, and we have therefore hypothesized their potential as feed supplements towards SBM diets improvement.

The goal of this study was to evaluate the potential mechanisms by which BS might exert a protective effect on reducing inflammation and restore barrier function in rainbow trout over a long-term experimental period. A total of 2,000 fish initially weighing ~ 40.0 ± 1.0 g, were randomly distributed into 20 350-L tanks (100 fish/tank). Five experimental diets (isonitrogenous: 42% crude protein and isolipidic: 20% lipid) including a FM diet (control), a SBM30 diet (30% inclusion level), a SBM40 diet (40% inclusion level) and two SBM-BS supplemented diets (1.5% BS supplemented to each SBM diet) were fed to apparent satiation for 18 weeks. During the trial fish were sampled three times at 6, 12, and 18 weeks.

Feed intake was recorded daily, and fish weight was measured at each sampling point. Samples were collected from the distal intestine for gene expression analyses of the inflammatory markers: TNF-α, NF-κB, IL-8, IL-10; barrier function markers: MLCK, occludin and claudins; as well as the bile brush border transporter TGR5 and basolateral transporter OSTα. Similarly, liver gene expression analysis was done on the bile acid synthesis enzyme. Digesta was taken from proximal and distal intestine 12 hrs. after feeding for bile acid quantification and resorption calculation. Distal intestine and liver samples were also collected for histology analysis.

No significant differences ($p > 0.05$) in weight gain were observed, but fish fed the SBM40 diet showed slightly higher weight gain in the BS supplemented diet compared to the non-supplemented one (Fig. 1), and this was supported by significantly higher ($p < 0.05$) feed conversion ratio (FCR) at 18 weeks. Histology analyses in liver and distal intestine also indicate a mitigating effect of BS supplementation in diets containing up to 40% SBM.

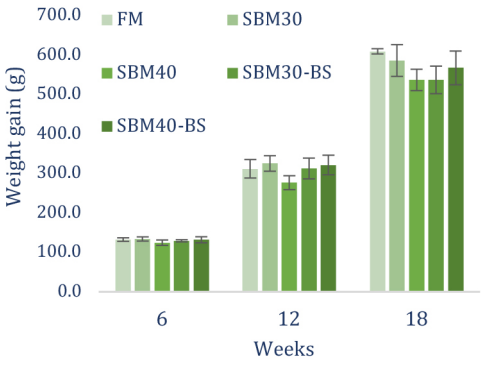

Fig. 1. Weight gain (g) of each dietary group at each sampling point.

PRODUCTION AND COMMERCIALIZATION OF JAPANESE OYSTERS IN BAJA CALIFORNIA SUR FOR THE NATIONAL AND EXPORT MARKET

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The Japanese Oyster *Crassostrea gigas* originates from the seas and estuaries of Japan and Korea: The species is considered as the most relevant bivalve mollusk in commercial aquaculture. In Mexico, the species was first introduced in Baja California in 1973.

Our commercial operation is located in a 53 ha coastal property with a 4 ha production concession at Estero El Chisguete, in Comondu, Baja California Sur, approximately 270 kms from La Paz, where conditions are ideal for the development of the species. We stock high quality seed from hatcheries in La Paz every 4 months for a carrying capacity of 1 million oysters per hectare. Commercial size (8-10 cm) is obtained using long-lines after 8-10 months at a weight of 80-120 grams. Annual production is 480,000 pieces with monthly sales of 40,000 units sold mainly in La Paz and Los Cabos for local consumption and tourism demand.

Our midterm objective is to increase production to 1.5 million oysters/year for the export market. We are looking for $5 million pesos (US$250,000) to improve infrastructure and for additional culture equipment and materials.

CULTIVATION OF MICROALGAE *Tetraselmis chuii* AND *Chaetoceros muelleri* USING SILAGE FROM FISH WASTE

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Live food is an essential part of the nutrition of the larval stages of different aquaculture species. Its quality is closely related to the means of fertilization used for its cultivation, being the F/2 Guillard the most used. Chemical silage from fish waste, with high nitrogen and carbon content, can be an alternative. In this study, the effect of chemical silage from fish waste as a supplement, complement and total substitute for the commercial culture medium F/2 Guillard on the growth of the microalgae *Tetraselmis chuii* and *Chaetoceros muelleri* was evaluated.

The silage was made with waste from the "Chac-chi" fish (*Haemulon plumierii*) according to Gallardo et al (2012) from the catch of riverside fishermen from the port of Sisal, Yucatan, Mexico. The pH and soluble protein content during silage was recorded (31 days).

Two experiments were carried out to evaluate the effect of partial and total replacement of the F/2 Guillard medium on the population growth of *T. chuii* (Exp. 1) and *Ch. muelleri* (Exp. 2) (Table 1) during 10 and 8 days respectively. The cultures of both microalgae were carried out in 500 ml Erlenmeyer flasks.

Table 1. Experimental design of experiment 1 and 2.

Treatments	Description	n
Control (TCT)	Sol A + sol B	3
A (TA)	Sol A + Sol B + silage*	3
B (TB)	Sol A + silage*	3
C (TC)	Silage*	3

** The silage was supplied on day 0 and day3. Metasilicate was added for Exp. 2 with Ch. muelleri*

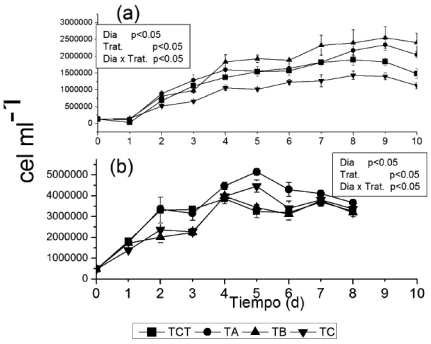

Fig. 1.- Effect of silage on the growth (cel ml^{-1}) of *Tetraselmis chuii* (a) and *Chaetoceros muelleri* (b). Mean ± S.E. n = 3.

Silage as a complement and supplement to F/2 Guillard allowed higher concentrations (cell ml^{-1}) of *T. chuii* with TB and TA on day 9 of culture, respectively (Fig. 1a). In the case of diatoms, the highest concentration was obtained on day 5 of culture with the TA (5.15 x 10-6 cel ml^{-1}) and where only the silage (TC) was used (4.4 x 10-6 cel ml- 1) (Fig. 1b).

The results obtained indicate that the chemical silage of fish waste with a high content of nitrogen and other elements, provided nutrients that were beneficial for the growth of microalgae and that it can be used as an enrichment or partial replacement of the F/2 Guillard medium for *T. chuii*. In the case of *Ch. muelleri*, the results indicate that the silage allowed a total replacement of the commercial medium F/2 Guillard.

Literature

Gallardo P., …., and Sotelo A. 2012. Nutritive value of diets containing fish silage for juvenile *Litopenaeus vannamei* (Bonne, 1931). ***Journal of the Science of Food and Agriculture*** (DOI 10.1002/jsfa.5632)**. 2320 -2325**

Acknowledgments

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Epinephelus morio A HOLOBIONT VISION

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Red grouper (*Epinephelus morio)* is a species of great economic importance in Yucatán, where it is the third species most captured, with a production of 5,550.36 tons in live weight, representing the 60.89% of the total national live weight of grouper fishing, with a value of 335.74 million Mexican pesos (CONAPESCA, 2020). Microbiota, the range of microorganisms that can be found in a multicellular organism can be affected by environmental factors such as pH, temperatura, and salinity with an influence in metabolism, growth, and development among other vital processes. The relationship between the microbiota and the host is known as the holobiont system. The gut microbiota has been extensively studied in several species of fish, however information regarding *Epinephelus morio* remains largely elusive. The objective of this study was to characterize the cultivable gut microbiota of *E. morio* in addition to implementing a new protocol for gut microbiota sampling *in vivo*.

For this purpose, 3 wild red grouper adults captured off the Yucatán Coast and taken to the facilities of the UMDI-Sisal of the Faculty of Sciences, UNAM were studied. They were anesthetized with clove oil, then a medical probe and a syringe were used to aspirate gut content from the cloaca. Samples were serially diluted in sterile physiological solution, cultured in TSA medium and marine agar plates in triplicates and incubated for 7 days at 37 °C. A total of 21 colonies were selected for further isolation in TSA medium plates incubated for 24 h at 37 °C. Isolates (n=20) were characterized using Gram stain, the BD Crystal system and 16S rRNA gene sequencing.

The isolates were identified as *Staphylococcus simulans* (5), *Staphylococcus sciuri* (1), *Staphylococcus haemolyticus* (1), *Staphylococcus saprophyticus* (1), *Corynebacterium diphteriae* (4), *Corynebacterium* sp. (1) and *Photobacterium damselae (7)*. Further studies will be performed to shed light on the roles of the *E. morio* gut microbiota in vital functions and health.

Fig. 1 *Epinephelus morio* adult individual.

Fig. 2 *Corynebacterium diphtheriae* isolate in TSA medium.

PHYSICOCHEMICAL AND METABOLIC VARIABLES OF WHITE SNOOK *Centropomus viridis*, CULTURE IN FLOATING CAGES IN NORTHWESTERN MEXICO

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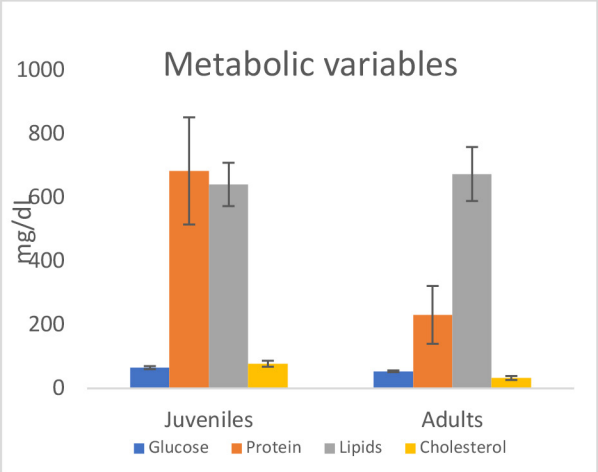
Snook is considered to be one of the most important fisheries in Mexico is considered one of the most important fish species because of its commercial and nutritional value. Blood biochemical parameters are frequently used to determine the health status of cultured fish. The objective of this study was to evaluate the metabolic variables of the white snook *Centropomus viridis* culture in 10m diameter and 3m deep cages (235m^3), 60 (30 juveniles and 30 adults) organisms were stacked during the experiment. The physicochemical parameters such as dissolved oxygen, temperature, pH and salinity as well as ammonia, nitrites, nitrates and metabolic variables such as glucose, protein, lipids and cholesterol were closely monitored, and recorded. Performance variables such as specific growth rate, and survival were evaluated. The results of our performance variables were a daily growth rate of 2.17 ± .06, 2.35 ± .05 and 2.49 ± .06 g, and 1.67, a total survival of 27% for the nursery stage and 60% for the grow out stage; the metabolic variables the protein and cholesterol had significant differences. Correlation between physicochemical parameters and growth was determined through a correlation analysis and a direct correlation was found between temperature and growth.



EFFECT OF EXPERIMENTAL VIRAL INFECTION (PaV1) ON THE IMMUNE SYSTEM OF THE CARIBBEAN SPINY LOBSTER (P*anulirus argus*)

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Immunological challenges are widely used to corroborate the success of breeding programs for lines resistant to specific pathogens, and to investigate immune-additives that improve immunity of cultured animals. The Caribbean spiny lobster, *Panulirus argus*, supports valuable commercial fisheries through the wider Caribbean region. Since the last decade, this species has been affected by a highly pathogenic virus known as *Panulirus argus* Virus 1 (PaV1). The present study analyzes the immune response of lobsters under an experimental viral infection.

Ninety juvenile *P. argus* were distributed in three tanks with an approximate capacity of 150 liters of water connected to a recirculation system with 300% daily replacement. Subsequently, one organism with signs of PaV1 infection was introduced into each pond for 120 days.

Hemolymph was extracted to determine the activity of the immuno-physiological components of the peripheral system. Diagnosis of infection was made by qPCR using samples from the hepatopancreas. Immunological evaluations were also performed: hemagglutination activity and total phenoloxidase activity (PO).

Statistical analysis (ANOVA), showed a decrease in the levels of hemocytes, hemagglutinating activity, and hemo-cyanin concentration. On the other hand, the phenoloxidase system did not show significant differences. However, the components of this system showed a decrease in plasma total phenoloxidase and an increase in hemocyte granule phenoloxidase. The results suggest that experimental viral infection produce a defense response similar to natural pathways of contagion, and provides the bases to validate an immunological challenge protocol for crustaceans.

Acknowledgments
This work was carried out with the support of the UNAM (PAPIIT IN217322, IT201621).

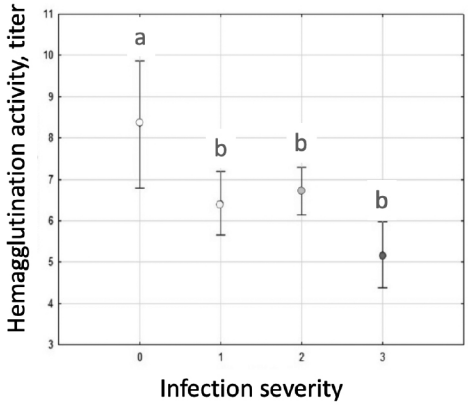

Figure 2. Lectin recognition by hemagglutination activity in *P. argus* infected with PaV1 through water. The degrees of severity correspond to organisms without infection, and with light, medium and advanced infection (0, 1, 2 and 3, respectively), which was determined on the basis of qPCR. Mean values ± interquartile range. Different letters denote a significant difference ($p<0.002$), with a confidence level of 95%.

EXTRUSION PROCESSING OF FISH FEED PELLETS: INFLUENCE OF SILICA AND OIL CONTENT ON THE PRODUCT PROPERTIES

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Over the last several decades, aquaculture has gained importance since wild capture can no longer meet the global demand for fish. Therefore, this market has grown exponentially, and with it, the demand for fish feed. In particular feed with high amounts of fat has gained relevance, since it allows for many species (e.g. salmon, trout) the most efficient growth and, thus, farming.

Extrusion processing is often used to produce fish feed pellets. Within the heated barrel containing rotating screws, the raw materials are mixed, heated, and sheared along the extruder. The combined thermal and mechanical stresses applied to the material lead to its plasticization and cooking. Due to the temperature and pressure gradients between the melt and the room conditions, the material can expand after it leaves the die and is pelletized through a rotating knife. Although addition of oil is desired, it has a negative influence on the cooking process during extrusion, as its lubricating properties reduce the specific thermal and mechanical energy input and, consequently, the degree of cook. In general, a lower degree of cook has a negative influence on the product quality of the feed pellets, i.e. decreased water stability and hardness. Therefore, to cook the material sufficiently and to achieve the aimed product properties, manufacturers avoid adding high oil levels during extrusion. Instead, manufacturers add oil in a separate coating step post-extrusion. This additional step makes the production of aquatic feed more complicated and expensive. Additionally, if fat does not bind properly to the pellets fat leakage can occur, whereby the fat drains out of the pellet, consequently, leading to increased fat oxidation. The addition of additives, especially silica, would serve to resolve these issues while achieving high fat levels in high quality fish feed pellets without a coating step. Since the increase of fat leads to a decrease in the matrix viscosity, the mechanical energy input is expected to decrease leading to a lower degree of cook. As silica is known for its water and oil binding capacity as well as for its ability to control rheology and therefore to increase viscosity, its addition is expected to counteract this effect leading to an enhanced gelatinization during the extrusion process. This strategy will be introduced and the relationship between process conditions, addition of different oil and silica concentrations and resulting product properties (among others pellet hardness, pellet abrasion and fat content) will be evaluated.

FROM LA PAZ TO MEXICO AND THE WORLD, WITH LOVE, THE KAMPACHI COMPANY: A RIPPLE EFFECT ANALYSIS

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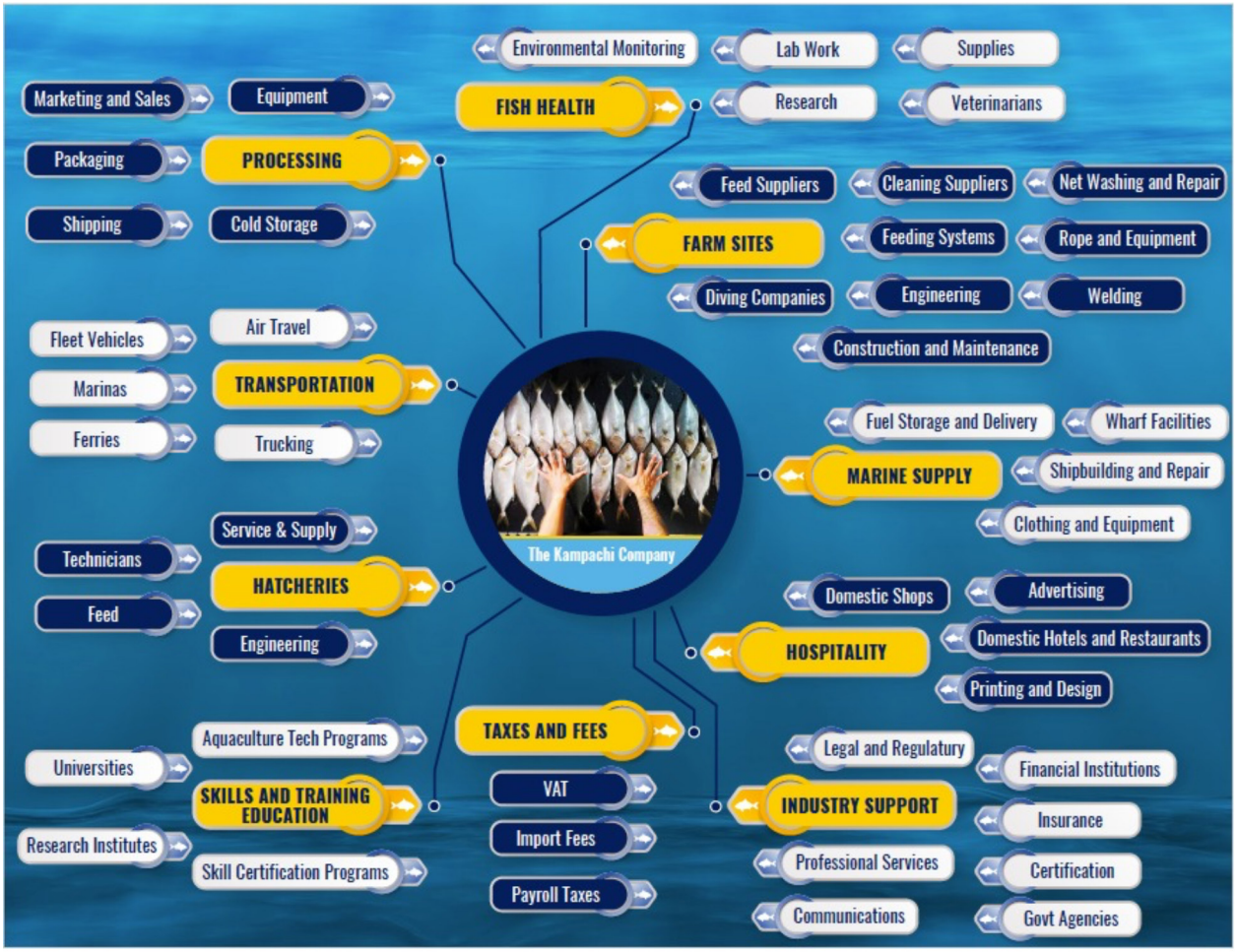
Launched in October 2017, in and about La Paz, B.C.S. México, the Kampachi Company's Mexican operations built from scratch the infrastructure required to raise *Seriola rivoliana* from broodstock to commercial harvest size. As of the end of April, 2021, it had produced close to 1,200 metric tons of King Kampachi® and Kampachito®, and sold this production mainly in Mexico and the United States. This presentation will give an overview of the project's direct and indirect impact, in economic, social and academic dimensions.



FROM ZERO TO ONE THOUSAND PLUS

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Aquaculture represents the most significant potential for mass production of high quality protein. New species and new sites are needed to fulfil the world expectations of sustainable food for 2050. The road to success from start up to successful production is discussed for a new knowledge-based enterprise.

In four years after securing investment, including two full years of a full pandemic, the King Kampachi project in La Paz, Baja California Sur went from zero to more than one thousand tons of *Seriola rivoli*ana fish ready for the Mexican and export markets. Along the way, many significant goals were achieved. Among them, a domestic company (Nutec) became the high quality feed supplier for the company, all the processes and permits required for fertilized egg export were obtained, and a new product, *Kampachito* (little Kampachi), was developed. All this while complying with strict internal regulatory collaboration and certification processes.

BENEFITS OF ADJUSTING TRACE MINERAL PREMIX IN SHRIMP FEED– A MEXICAN FIELD STUDY

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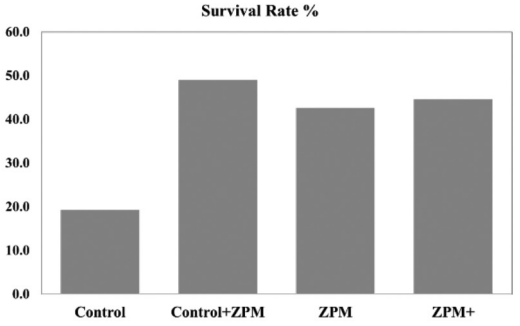
Replacement of inorganic mineral premix (sulfates) with complete metal-AA complexes (i.e. Zinpro Performance Minerals®, ZPM) at 0.5x rate of inorganic, resulted in numerically higher body weight, 8.3% reduced FCR, and reduced mortality of shrimp challenged with *Vibrio harveyi* (Jintasataporn et al., 2015). Metal-amino acid complexes have an absorption advantage over inorganic minerals, because they are minimally antagonized by dietary components such as phytic acid and utilize the AA transporters instead of common metal transporters. Davis et al. (1993) reported that *L. vanammei* required 33 ppm Zn to maintain normal growth and maximize Zn concentration in hepatopancreas, whereas 200 ppm inorganic Zn was required to overcome depressed Zn bioavailability caused by phytates. More recently, Yuan and co-authors reported improved growth performance and health of shrimp when replacing 120 ppm zinc sulphate with 60 ppm Zn-AA complex (Yuan et al. 2020) and 30 ppm Cu sulphate with 15 ppm Cu-AA complex (Yuan et al., 2019a,b). Importantly, increasing Zn-AA complex from 60 to 80 ppm further improved shrimp health-related parameters (Shi et al., 2021). The present study aimed to evaluate benefits of replacing inorganic trace mineral premix with metal-AA complexes (ZPM) on shrimp performance and survival rate under commercial conditions. Shrimp of 0.5 g initial body weight were grown for ~100 days (cycle varying from 95-130 days) in earthen ponds at a stocking density of 100 shrimp per m^2 and 5-6 ppt. Feeds were produced at a commercial feed plant in Mexico, following ingredient and nutrient composition of a typical commercial shrimp feed. Four feeds were produced, varying in their trace mineral premix, as described in embedded table. Inorganic trace mineral premix used in the Control diet was combined with ZPM premix (Control+ZPM) or completely replaced with ZPM (ZPM diet). An additional diet was produced to include fortified ZPM premix (ZPM+), by giving special attention to trace minerals critical in the modulation of shrimp immune system (Zn, Se, Cu, and Cr). Replacement of inorganic premix with ZPM doubled shrimp survival rate. Fortifying trace mineral premix by combining inorganic with ZPM (higher total levels) or using ZPM+, tended to further increase survival rates. Regardless of the treatment, adjustment of inorganic trace mineral premix with ZPM increased final biomass and significantly reduced FCR, translating in positive return on investment (ROI, measured by additional income over feed cost) under commercial conditions. Best ROI was found with ZPM+ treatment (>150%).

	Supplemented Trace Mineral, ppm feed			
TM Source	Control	Control+ZPM	ZPM	ZPM+
Inorganic Zn	60.5	60.5		
Availa Zn		60.5	60.5	80
Inorganic Se	0.24	0.24		
Availa Se		0.24	0.24	0.4
Inorganic Cu	21	21		
Availa Cu		21	21	40
Inorganic Mn	47.2	47.2		
Availa Mn		47.2	47.2	40
Inorganic Fe	48.6	48.6		
Availa Fe		48.6	48.6	50
Availa Cr				0.4



ESTIMATION OF CHOLINE REQUIREMENT AND EFFECTS OF DIETARY LIPID LEVEL AND ENVIRONMENTAL TEMPERATURE ON THE SEVERITY OF LIPID ACCUMULATION IN ATLANTIC SALMON (*Salmo Salar L.*)

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Background
The dietary shift of farmed Atlantic salmon (*Salmo salar L.*) from fish-based to plant-based protein has revealed new challenges concerning the dietary requirements necessary to ensure fish health. Among the nutrients lacking in plant feeds, choline deficiency can lead to the presence of excessive unabsorbed fat within the intestinal enterocytes, a condition known as Lipid Malabsorption Syndrome (LMS). However, knowledge over the minimum dietary choline requirement in Atlantic salmon is still limited. It is also likely that the amount may vary across a salmon's life, reflecting growth demands and changes in diet. Here we present part of two experiments conducted to establish the choline requirement in post-smolt Atlantic salmon and determine whether this requirement may be affected by different dietary lipid levels and water temperatures.

Materials and Methods
An initial dose-response experiment was performed to achieve a first estimation of minimum choline requirement. A fish-based control diet and eight plant-based diets supplemented with different levels of choline chloride were made and fed to post-smolt Atlantic salmon. In a subsequent experiment, four choline-deficient plant diets with differing lipid concentrations were provided to fish raised at 8 and 15 ˚C to assess the influence of lipid level and water temperature. At the end of each feeding trial, fish were sacrificed, and body measurements were taken. The fish were then opened, and the organ package removed. The intestinal tract was detached from the other organs, cleaned of external fat and divided into the pyloric (PI), mid (MI) and distal (DI) sections which were then weighed and sampled. Tissue samples from all the fish were processed to perform gene expression analyses and histological assessments.

Results
Biomarkers of various indicators of lipid assembly, storage and transport showed an inverse relationship with the increasing choline level in the diet. These elements were selected as main factors to determine choline requirement and indicated that a dietary inclusion of 3.4 g/kg is necessary to prevent LMS symptoms in post-smolt Atlantic salmon kept under similar conditions. The results showed a clear dose response relationship between fat percentage and lipid accumulation both at a histological and molecular level. On the other hand, water temperature only had a significant effect when interacting with dietary lipid concentration.

LAMBARI *Astyanax lacustris* AND MUSTARD MICROGREENS *Brassica juncea* PRODUCTION IN SLIGHTLY SALINE AQUAPONIC SYSTEMS

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In freshwater aquaculture, a more isosmotic environment achieved through salinity control may favor fish growth since less energy would be spent for the osmoregulatory process. A 57-day experimental trial was conducted to study aquaponics on-demand coupled system using lambari (*Astyanax lacustris*) and mustard (*Brassica juncea*), species reported as tolerant to slightly saline conditions. The mustard was cultured as microgreens given their high sales price, short culture cycle and low nutrient demand. Two treatments (1.0 and 2.0 g NaCl L^{-1}) with four replicates each, were established. An aquaponics on-demand control without salt addition (0) and a hydroponic control (MHy) for the microgreens were also settled, each one with four replicates. During the trial, one lambari and five microgreen culture cycles (9 day each) were completed. At the end of the trial, the lambari (Table 1) and all the microgreen cycles (Table 2 presents only cycles with the highest and lowest microgreen harvests) performances in the 1.0 and 2.0 treatments were statistically similar to the controls (ANOVA, $p>0.05$). However, lambari asymptotic weight was achieved earlier in the 2.0 treatment compared with the control and the 1.0 treatment (Fig.1).

In conclusion, the use of slightly saline water (2g NaCl L^{-1}) in aquaponics on-demand coupled systems may accelerate lambari growth without compromising mustard microgreen production if compared with the performance on RAS (0g NaCl L^{-1}) and hydroponic systems, respectively.

Funding agencies: FAPESP, grants 2018/23658-5, 2018/23605-9 and 2017/50431-9, cnpq, grants 139130/2020 and 311108/2017-2.

Table 1. Average ± sd values for lambari performance

Treatment	Initial weight (g)	Final weight (g)	SGR (%/day)	FCR
0	3.04 ± 0.10	7.72 ± 0.23	1.63 ± 0.09	1.54 ± 0.15
1.0	3.15 ± 0.20	7.63 ± 1.32	1.53 ± 0.23	1.78 ± 0.71
2.0	3.15 ± 0.11	7.74 ± 0.39	1.58 ± 0.11	1.67 ± 0.25

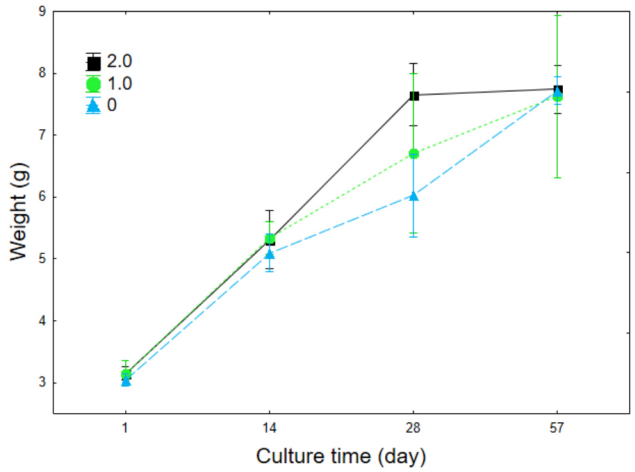

Figure 1. Lambari growth performance (± sd) during the trial in treatments 2.0, 1.0 and 0.

Table 2. Average ± sd values of productivity indexes in the cycles with the lowest (Lw) and highest (Hg) microgreen harvests (HL)

HL/ cycle	Treatment	Productivity (Kg m^{-2})	Weekly Productivity (Kg m^{-2})
Lw/1	**0**	0.58 ± 0.08	0.45 ± 0.06
	1.0	0.68 ± 0.09	0.53 ± 0.07
	2.0	0.62 ± 0.05	0.48 ± 0.04
	MHy	0.60 ± 0.07	0.46 ± 0.05
Hg/3	**0**	0.87 ± 0.11	0.67 ± 0.08
	1.0	0.94 ± 0.05	0.73 ± 0.04
	2.0	0.96 ± 0.06	0.75 ± 0.05
	MHy	0.90 ± 0.10	0.70 ± 0.07

IS ESCHERICIA COLI A GOOD INDICATOR OF POSSIBLE FECAL CONTAMINATION OF CATFISH FILLETS?

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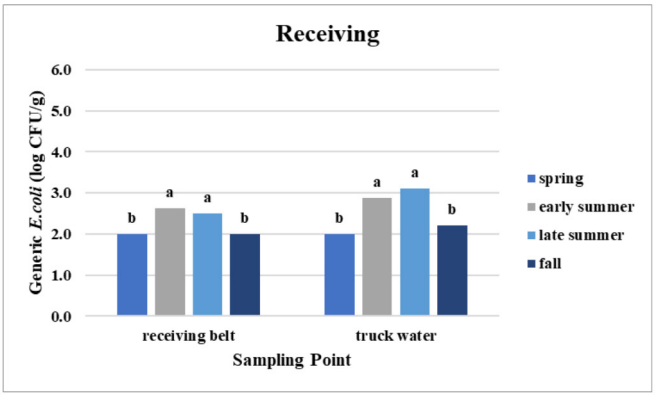
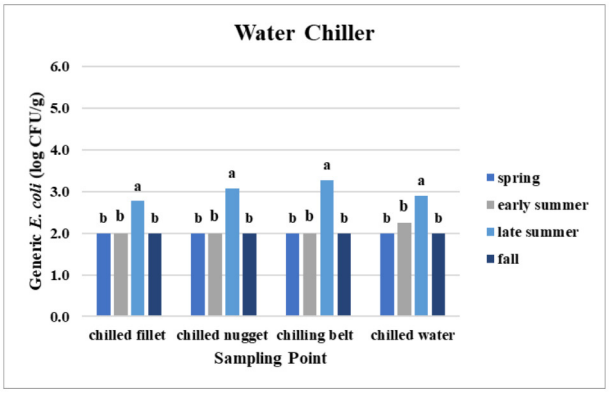
In 2017, USDA-FSIS compliance guidelines for Siluriformes, implemented the testing of generic *Escherichia coli* to ensure product wholesomeness and safety. The BAP standards for processed fish call for rejection of sample if from n= 5 subsamples, 3 or more subsamples exceed 4 MPN per gram or any subsample exceed 40 MPN per gram. Others have placed a limit of 1 CFU/g. *E. coli* and other indicator organisms in catfish, liquid and environment of processing facilities were assessed by season and time of day. Environmental, fish, and liquid samples were collected in 3MTM Swab-Sampler (with 10 ml of Buffered Peptone Water) and Whirl-pak® sterilized stomacher bags respectively from two different catfish processing plants. (site) located in the state of Mississippi, USA. Replications were defined by the number of visits. Each two visits represented a season and for each visit, 25 samples were collected. All samples were plated on the following corresponding media: aerobic plate (APC), psychotropic plate (PPC), and indicator organisms: generic *E. coli*, total coliforms (TCC), and Enterobacter following FDA-BAM and AOAC methods. *Escherichia coli* counts were similar ($P \leq 0.05$) for all sampling points with slightly higher incidence in the late Summer than the other seasons. Within the types of sampling, liquid and fish part sample had higher counts than ($p \leq 0.05$) environmental samples. Liquid samples consisting of truck water (5.3 log CFU/ml) and chilled water (5.1 log CFU/ml) resulted in higher counts than ($p \leq 0.05$) offal water and injector solution (4.4,4.7 log CFU/g, respectively). It is possible that the presence of indicator organisms evaluated varies according to the sampling season, considering that such microorganisms have been found to have a relationship with pathogens, *E. coli* could be part of the natural microflora of the catfish ponds, thus not a good fecal indicator.





πBIOACTIVITY POTENTIAL AND DRUGLIKENESS OF NATURAL PRODUCTS FROM MARINE ORGANISMS

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Natural products (NP) are enourmous, still largely unexplored resource of molecules with potentially therapeutic properties. More than 90 % of Earth species live in the marine environments, providing vast structural diversity and bioactivity potential of NPs associated with marine organisms. By investigating these organisms new compounds with applications in agriculture, medicine and various biotechnological processes may be discovered. While plants and terrestrial microorganisms have been more explored for new chemical entities so far, the increasing number of new drug candidates and drugs from marine organisms is showing that this resource will be of major interest in the future. Computational approaches offer a scalable, early approach for evaluating of bioactivity potential of large chemical space of natural products. We provide here a large scale, qualitative breakdown of properties of small molecules (less than 800 Da) stemming from different organisms and compare them against current drugs and nutraceuticals. We used COCONUT [1] (natural products) and DrugBank [2] (drugs and nutraceuticals) as sources of compounds, PASS software [3] to provide bioactivity estimates and Classyfire WEB service [4] to structuraly classify compounds. Table 1. depicts major structure-bioactivity profiles of compounds from different origins, with their rough druglikenes assessment. Using more rigorous computational (AI, machine learning based) approaches we are working to provide deeper structural-biaoctivity assessment of the compounds originating in marine organisms.

Table 1. Breakdown of major structural, bioactivity and drug-like properties of compounds from marine organisms, microalgae, plants compared to drugs and nutraceuticals on the market.

Marine organisms												
Structures			Druglikeness			Bioactivity						
Structure superclass	Number	%	MW-failures %	logP failures %	Average LoR failures	Bioactive* %	Antibacterial %	Antiinflammatory %	Chemopreventive %	Radioprotector %	Antidiabetic %
Lipids and lipid-like molecules	9191	49.50%	27.4%	46.0%	0.88	93.0%	57.5%	73.2%	40.1%	17.0%	25.1%
Organoheterocyclic compounds	3849	20.70%	14.6%	21.2%	0.41	89.4%	35.8%	36.4%	12.3%	4.9%	9.2%
Organic acids and derivatives	1715	9.20%	40.3%	8.9%	0.75	87.7%	34.6%	28.2%	8.3%	12.4%	10.2%
Benzenoids	1424	7.60%	22.3%	22.0%	0.51	90.3%	30.1%	31.0%	12.7%	6.7%	15.5%
Others	2379	12.80%	21.1%	24.3%	0.63	91.8%	42.6%	44.5%	15.3%	10.4%	13.5%
Microalgae											
Organic acids and derivatives	256	32.50%	73.4%	14.5%	1.16	93.4%	40.6%	9.0%	2.0%	15.2%	4.7%
Lipids and lipid-like molecules	236	30.00%	28.0%	50.9%	0.88	98.7%	63.1%	86.4%	51.7%	30.1%	42.8%
Organoheterocyclic compounds	80	10.20%	20.0%	17.5%	0.59	97.5%	43.8%	25.0%	3.8%	8.8%	13.8%
Phenylpropanoids and polyketides	62	7.90%	54.8%	30.7%	1.26	100.0%	72.6%	75.8%	33.9%	19.4%	30.7%
Others	152	19.30%	27.6%	40.8%	0.89	94.7%	43.4%	39.5%	11.2%	23.0%	22.4%
Plants											
Lipids and lipid-like molecules	10895	36.00%	48.5%	38.4%	1.17	91.5%	51.9%	77.6%	44.0%	9.7%	30.3%
Phenylpropanoids and polyketides	5979	19.80%	57.1%	28.9%	1.54	72.7%	36.2%	50.4%	45.2%	22.7%	28.7%
Organoheterocyclic compounds	4587	15.20%	41.1%	26.1%	0.89	69.2%	19.3%	32.1%	14.2%	5.9%	8.4%
Benzenoids	4218	13.90%	55.5%	42.8%	1.37	80.1%	22.9%	34.5%	17.2%	10.3%	13.1%
Others	4565	15.10%	41.8%	17.1%	1.11	85.3%	44.3%	51.7%	26.9%	19.0%	31.7%
Nutraceuticals											
Organic acids and derivatives	37	37.00%	0.0%	0.0%	0.08						
Lipids and lipid-like molecules	29	29.00%	31.0%	69.0%	1.24						
Organoheterocyclic compounds	20	20.00%	5.0%	0.0%	0.25						
Nucleosides, nucleotides, and analogues	5	5.00%	60.0%	0.0%	2.00						
Others	9	9.00%	11.1%	0.0%	0.33						
Drugs											
Organoheterocyclic compounds	768	30.70%	10.6%	9.1%	0.25						
Benzenoids	646	25.80%	10.7%	13.8%	0.29						
Lipids and lipid-like molecules	309	12.30%	18.5%	28.2%	0.53						
Organic acids and derivatives	302	12.10%	29.5%	3.3%	0.66						
Others	477	19.00%	26.0%	10.5%	0.69						

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DEVELOPMENT OF IMPROVED COMMERCIAL FEEDS FOR COMMERCIAL NILE TILAPIA *(Oreochromis niloticus)*. A LABORATORY STUDY ON BALANCING COST, PERFORMANCE, AND SUSTAINABILITY

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The state of static production volume coupled with highly volatile market conditions leaves farmers with few options for increasing the profitability of their businesses. Considering that intensive aquaculture feed expenses generally account for 50-70% of the total variable costs, its reduction constitutes a priority for the economical sustainability of aquaculture operations and a prime subject for investigation. Thus, research devoted to the development of nutritious, environmentally friendly, and cost-effective diets is of utmost importance to ensure the success and sustainability of aquaculture enterprises. Formulated aquaculture feeds are among the most expensive animal feeds on the market. In this 12-week study, two commercial tilapia formulations were fed to Nile tilapia in an indoor RAS at the University of Idaho's Aquaculture Research Institute, Moscow, ID utilizing two feeding strategies. Fish were fed either a single feed throughout the experiment; Diet A (36% CP 6% Lipid), Diet B (40% CP, 10% Lipid) or switched from Diet B to Diet A after 6-weeks for finishing. Results indicated that the fish fed Diet A had significantly lower daily gain and significantly higher FCR than fish fed solely Diet B or the treatment fish fed Diet B for 6 weeks followed by Diet A for the final six weeks. The unit cost of Diet A is lower than Diet B, however we will discuss the impact on final profit due to the feed and production cost differentials.

Treatment	Av. Final wt. (g)	Survival (%)	FCR	Gain (g/d/fish)
36-6	568.66 +/- 34.22	99.05 +/- 0.95	1.30 +/- 0.04 c	3.78 +/-0.43 c
40-10 to 36-6	631.33 +/- 31.68	99.05 +/- 0.95	1.24 +/- 0.02 ab	4.59 +/- 0.42 ab
40-10	653.00 +/- 39.10	100.00 +/- 0.0	1.21 +/- 0.03 ab	4.86 +/- 0.51 ab

THE NUTRIMO PROJECT: DEVELOPMENT OF HANDMADE DIETS FOR TILAPIA NUTRITION IN COMMUNITY-BASED AQUACULTURE IN CABO DELGADO, MOZAMBIQUE

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The project NutriMo (Development of handmade diets for tilapia nutrition in community-based aquaculture in Mozambique) aims to improve the development of sustainable aquaculture in Mozambique, namely the community-based tilapia aquaculture in inland villages of Cabo Delgado province. The overall objective is to substantially improve the community aquaculture practiced in Mozambique inland waters, thus contributing to improving the quality of life of the villages, enabling the production of food in an efficient and sustainable way and reducing the levels of poverty in the villages of Mozambique.

Developing countries in Eastern Africa, like Mozambique, are overdependent on natural resources, which causes ecosystems degradation. Community-based aquaculture can increase income and locally available food resources in villages with limited access to animal protein. However, to ensure the success of this activity, it is extremely important to develop adequate diets, based on local ingredients, that can be produced at the community level.

Tilapias have a high potential to be farmed under simplified conditions due to their characteristics, such as omnivory, fast growth and resistance to stress and diseases. Here, an experimental diet, based on local ingredients from the North of Mozambique, was developed and tested against a commercial diet, in tilapia fingerlings, during 2 months. Growth performance and resistance to thermal stress were evaluated. Tilapia fed with the experimental diet presented lower growth rates, lower DNA damage, and higher neurophysiological and antioxidant activity leading to increased oxidative stress.

Overall, we show that the dietary inclusion of local ingredients could be viable for community-based aquaculture in earthen ponds.

IMPACTO DEL SER HUMANO EN EL LANGOSTINO DE RÍO *Macrobrachium americanum* EN LAS POBLACIONES SILVESTRES DE SAN PEDRO DE LA PRESA (BCS) Y AGUAS BLANCAS (GRO)

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El langostino de río *Macrobrachium americanum* es una especie que puede alcanzar una talla de 460 g en el medio silvestre, por lo que tiene una gran importancia económica y ecológica. Desde hace más de una década, las especies de langostino han sido explotadas por parte de pescadores ribereños, para proveer de producto a los mercados locales o bien, para autoconsumo (New, 2009). El deterioro constante del hábitat en el cual se desarrollan las especies de langostinos, aunado con el incremento en la demanda de producto por los mercados locales, ha puesto en riesgo a las poblaciones naturales de estos crustáceos.

Por ello, se realizó un estudio de conservación de la especie en San Pedro de la Presa y puntos circunvecinos, Baja California Sur y en el poblado de aguas blancas, Municipio de Coyuca de Benítez, Guerrero. Se llevó a cabo por medio del método cuantitativo para la recolección de datos primarios (encuestas). La información obtenida a través de las encuestas indicó que la captura de *M. americanum* se realiza generalmente durante la temporada de lluvias, cuando tiene lugar la reproducción de la especie. No existe la aplicación estricta de su reglamentación en su captura, lo cual ha ocasionado un descenso considerable en sus poblaciones, en ambas localidades (Fig. 1). Debido a que los volúmenes de pesca durante cada temporada no son suficientes para el mantenimiento de las familias de los pescadores, estos tienen que realizar otras actividades, no se dedican exclusivamente a la pesca. Existe poca información sobre el grado de explotación pesquera de las especies, por lo que se desconoce el estado real de las poblaciones silvestres. Se recomienda que es necesario una evaluación permanente del recurso, con el objetivo de implementar medidas que garanticen su conservación y manejo sustentable.

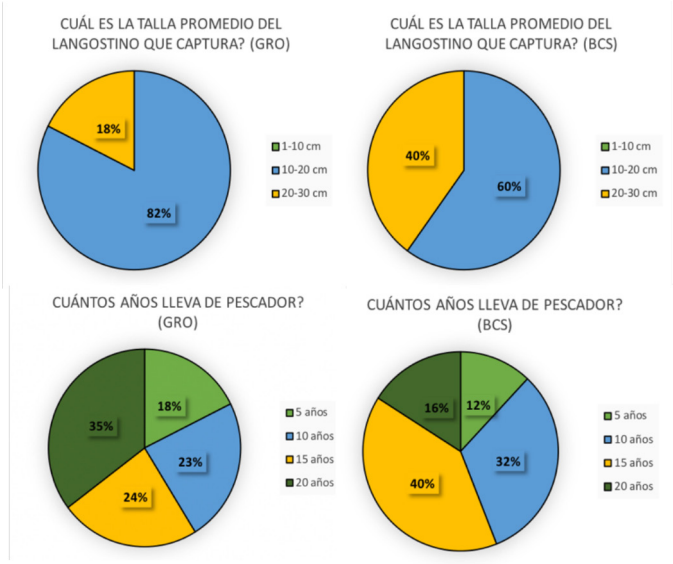

Figura 1. Captura del langostino de río

UN MÉTODO SIMPLE PARA ESTIMAR EL BIENESTAR ANIMAL EN GRANJAS ACUÍCOLAS

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El bienestar animal en acuacultura se relaciona generalmente con aspectos éticos, morales y de inocuidad. El concepto de bienestar animal es un tema de interés social, que está poco estudiado en las cadenas de producción acuícolas a nivel mundial, principalmente en países en desarrollo, además de ser un término incipiente y con varias connotaciones dependiendo de los ángulos en los que se analice. El bienestar animal en la acuacultura es una característica que se le adjudica a los organismos con pocas o nulas experiencias negativas en las distintas fases productivas y confiere a las especies un valor agregado, pues la calidad del producto mejora significativamente. En la mayoría de las unidades de producción acuícola del sureste de México, principalmente de tilapia y trucha, el tema de bienestar animal se sigue considerando como "tabú" y prácticamente no existe información sobre las percepciones de productores, comercializadores y consumidores ante este asunto. Para tratar de entender el estado actual del bienestar animal en la acuacultura continental del sureste de México, se diseñó un instrumento para evaluarlo de una manera rápida y objetiva. El instrumento se basa en la escala visual análoga (EVA) Wong-Baker FACES®, que es una escala que trabaja con intervalos. La escala toma valores de 0 a 10, que se traducen a una escala numérica de bienestar animal. Valores tendientes a 0 sugieren bajo nivel de sufrimiento para los organismos, mientras que valores tendientes a 10 corresponden a niveles elevados de sufrimiento. El instrumento se aplica dos etapas de las cadenas productivas: durante el proceso de cultivo, y durante la cosecha y sacrificio. Los criterios para asignar los diferentes intervalos corresponden a las Buenas Prácticas de Manejo, aceptadas convencionalmente en la mayoría de los sistemas de certificación. Resultados preliminares al utilizar este instrumento indican que la mayoría de las unidades de producción del sureste de México, principalmente las pequeñas granjas y los acuacultores de recursos limitados, cultivan, cosechan y sacrifican los organismos sin considerar y manejar adecuadamente el bienestar animal. La capacitación podría ayudar a revertir esta tendencia.

LA ACUACULTURA INTEGRADA MULTITRÓFICA COMO ESTRATEGIA PARA EL CULTIVO EN TANQUES DE PARGO CANANÉ *Ocyurus chrysurus,* EXPERIENCIAS PRELIMINARES

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En la península de Yucatán varias especies se encuentran sometidas a intensas presiones extractivas, entre las que se encuentra el pargo canané (*Ocyurus chrysurus*) integrante del "complejo rubia" y de la fauna acompañante del mero, sosteniendo actualmente la pesquería de escama en Yucatán ante la disminución en la abundancia del mero. Por esta razón, se plantea su cultivo como una alternativa para su producción en cautiverio, mediante la aplicación de un sistema integrado multitrófico (SIM) incluyendo pepino de mar (*Isostichopus badionotus*), macroalgas (*Solieria filiformis*) y halófitas (*Salicornia bigelovii, Rhizophora mangle*).

Se realizó un experimento durante 10 meses utilizando un SIM en sistema cerrado recirculante con los peces como especie principal. Se sembraron en un tanque de 1 m^3 54 juveniles de pargo canané con un peso promedio inicial de 37.53 g. Fueron alimentados ad libitum con alimento de engorda para pargo (Skretting Marine MX®; Tootle, UT, USA). El peso de los organismos se registró cada mes.

A los 10 meses se obtuvo una supervivencia del 96%, donde el 67% de los organismos alcanzó al menos 400 g de peso mínimo, indicando elevado potencial para cultivos comerciales.

El cultivo de pepino de mar se dividio en dos etapas. En la primera se evaluó por triplicado la densidad de 14 juveniles por m^3 en tanques de fibra de vidrio acoplados al SIM. Se alimentaron con una mezcla 1:1:1 de macroalgas (*Ulva* sp., *Sargassum* sp., *Macrocystis* sp.). Debido a las altas tasas de mortalidad, en una segunda etapa se redujo la densidad a 10 juveniles por m^3 y se enriqueció la alimentación con microdietas. Al final de esta etapa la supervivencia fue del 100% demostrando que *I. badionotus* tiene un elevado potencial para su cultivo en tanques en un SIM.

El efecto de las macroalgas (*S. filiformis*) y halófitas (*Salicornia bigelovii, Rizophora mangle*) en la concentración de metabolitos disueltos en el agua se evaluó mediante el cálculo de la tasa de retención (TR), para lo cual se llevó un registro de la concentración de NH4, NO2, NO3 y PO43 en diversos puntos del sistema.

S. filiformis y *S. bigelovii* resultaron adecuados como filtros biológicos, reteniendo cantidades significativas de ortofosfatos y metabolitos nitrogenados (NH4 y NO3) disueltos en el agua, manteniendo niveles adecuados para el crecimiento y supervivencia de los peces y los pepinos de mar.

Se concluye que el cultivo de pargo canané, pepino de mar, macroalgas y plantas halófitas en sistemas integrados multitróficos, es una alternativa factible para la producción acuícola sustentable de estos organismos, siendo éste el primer reporte de cultivo de *O. chrysurus* en tanques en tierra firme.

FORMULACIÓN DE CONCENTRADOS PROTEICOS A PARTIR DE SUBPRODUCTOS PARA NUTRICIÓN ACUÍCOLA

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INTRODUCCIÓN: La industria acuícola utiliza, en su mayoría, alimentos de origen animal como lo es la harina de pescado. Sin embargo, el alimento comercial representa alrededor del 50% de costo total de producción. Para reducir dicho costo es importante buscar alternativas sostenibles con subproductos para sustituir la harina de pescado (Faillace et al., 2016). METODOLOGÍA: Se elaboraron concentrados proteicos a partir de subproductos de origen vegetal y animal por extracción y precipitación de proteínas. Se formularon dietas isoproteicas para sustituir 7.5% y 15% la harina de pescado con los concentrados proteicos. La caracterización química proximal de los ingredientes y dietas se determino de acuerdo a la metodología recomendada por la AOAC (2010). RESULTADOS: Los análisis químicos proximales se encuentran en proceso y se expondrán los resultados en la presentación.

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BETTER PLANNING AND MANAGEMENT OF AQUACULTURE AS FIRST STEPS TOWARDS CLIMATE CHANGE ADAPTATION

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Climatic variability and climate change affect aquaculture and the sector has begun to develop standard and general adaptation measures often including improved access to information on climate change, livelihood diversification, access to credits, improved and more resilient infrastructure etc. However, often general adaptation measures are planned and implemented without a good understanding of the specific hazards and risks they will be addressing. Often as well the role of aquaculture planning and better management of production have not been acknowledged as a relevant first step towards adaptation. However, many types of impacts could cause effects whose magnitude would be farm-density dependent.

Following the IPCC risk framework here we examine the existence/implementation of a process to address and reduce risks including*: Information and climatic projections, assessing risks, reduce risks through best adaptation options and, the role of non-regrets actions* such as better planning and management. This was done by an analysis of more than 400 published papers and reports on the topic during the past 10 years. We also used 2 case studies from Chile; salmon farming and mussel farming.

Although the understanding about climate change impacts on the sector is increasing worldwide, in most cases, especially in developing countries, the lack of spatially and temporally relevant climate change risk assessment may be an obstacle to more targeted adaptation. Only about 8% of the publications involve field risk assessments leading to specific adaptation measures. Within this set of publications those dealing specifically with better planning and improved management as non-regret actions represents 5%.

Chilean aquaculture is globally prominent by its salmon and mussel production. Both sectors are expose to climate change and a national research initiative, ARCLIM, allowed to develop risk maps that showed areas at higher risk due to an increasing drought which will reduce freshwater inputs to fjords and coastal zones which may increase diseases in salmon and HABs affecting both. The assessment allowed to examine ways to reduce risks including spatially reallocating production to reduce both sensitivity and exposure in certain areas. In the case of mussel farming the collection of wild spats may be at high risk and the risk reduction measures involved better management of parental mussel beds as well as better management and spatial allocation of collection systems.

The analysis of the literature and the case studies allow to underscore the need to understand the role of better planning and management as non-regret actions and first steps towards adaptation. They are also actions that give more responsibility and ownership of the adaptation process to farmers.

MORPHOLOGICAL DEVELOPMENT IN CULTURED BARRED SAND BASS *Paralabrax nebulifer* LARVAE

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The study of the early life phases of marine fish allows us to know the morphological changes that occur throughout development and make it possible to improve biotechnologies for culturing species of interest. The barred sand bass *Paralabrax nebulifer* is the most important small-scale commercial fishery along the Pacific coast of Baja California, Mexico making is a considerable species with aquacultural potential. The aim of this work is to contribute to the recognition of main morphological changes during larval development until transformation to juvenile of barred sand bass, through the description of morphometry, meristic, pigmentation and squamation patterns.

During culture, the larvae were fed for 32 days on three diets (rotifers, *Artemia* nauplii, and microdiets). For the morphometric description, the phases of ontogenetic development were divided into apterolarval phase, pterolarval phase and juvenile, based on the criteria of saltatory ontogeny. The conventional measurements were taken from 219 specimens, corresponding to 112 apterolarval phase, 88 pterolarval phase, and 19 juveniles. To detect allometric growth, the morphometric relationships of each developmental phase were used by the growth coefficient. A series of developments was revealed for the study of meristics. The meristic, pigmentation and squamation patterns were outlined. As for results, the following developmental phases were observed: apterolarval phase, from 1.82-3.79 mm notochordal length (NL) (3-19 days post hatching [DPH]); pterolarval phase (10-25 DPH), with the presence of fin primordia from 3.80-8.05 mm NL; juveniles (23-30 DPH), with full finray complement from 8.06-11.27 mm standard length (SL). The growth of the body parts analyzed throughout the development of the apterolarval phase to the juvenile, behaved with positive or negative allometry, except in the case of the depth of the body, since isometry is present in the apterolarva phase. In the meristic study, it was observed that from the pterolarval phase, the presence of the fin elements were detected starting at 5.05 mm NL, along with the anal fin spine and the vertebral centers; later the rest of the fins were completed, until completing the count in 8.06 mm SL in the juvenile. In addition, the pigmentation and squamation patterns are described until transformation to juvenile. Finally, a feeding schedule from larval development to juvenile of barred sand bass based on their morphological changes is proposed (figure 1).

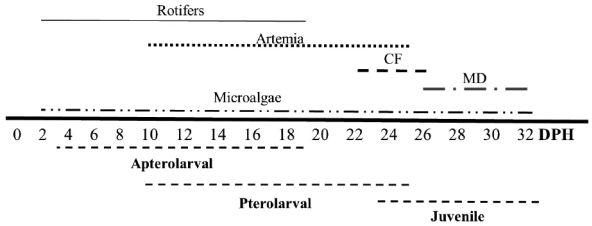

Figure 1. Proposed feeding schedule for larval rearing of barred sand bass until transformation of juvenile. The type of food, live food (rotifers, and *Artemia* nauplii), Co-feeding (CF), Microdiets (MD) and microalgae by DPH of each phase are shown.

BIOPROSPECTING PELOIDS –THE BORDERLINE BETWEEN LAND AND AQUATIC ENVIRONMENT

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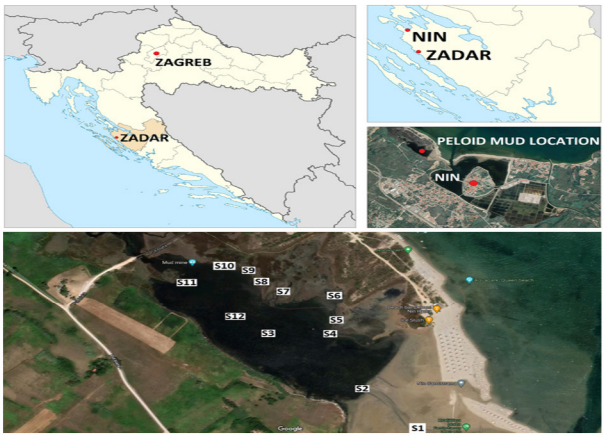
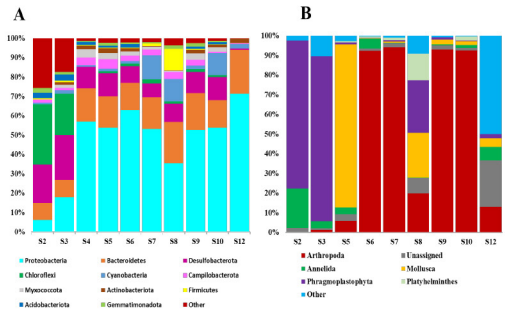
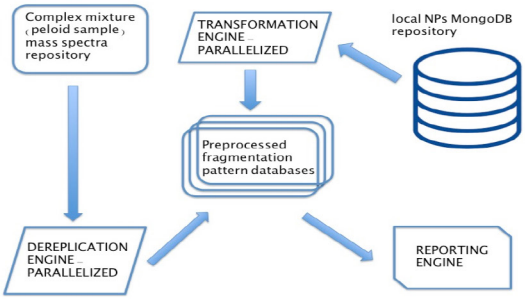
Seawater is an environment in which numerous organisms evolve, some with great potential for biotechnology. In recent years, however, many scientists have moved away from the assumption that the origin of life was in pools of water, and instead propose life on Earth probably originated in accumulations of warm, nutrient-rich mud. This mud, also called a peloid, has formed over many years through various processes and is a rich source of living organisms that, due to their adaptation to this unique environment, produce a wide variety of primary and secondary metabolites with numerous and diverse activities, including anti-cancer, anti-inflammatory, antiviral, and immunomodulatory ones.

This research aimed to characterize both prokaryotic and eukaryotic contribution to peloid. It also reports the living microorganism part of the peloid attributed healing properties by screening for bioactive compounds produced by microorganisms making up the peloid ecosystem. This was accomplished by high-throughput mass spectrometry coupled with bioinformatics pipeline based on DEREPLICATOR+ (Mohimani, H. et al. 2018) allowing for screening of thousands of available natural products from COCONUT database (Sorokina et al. 2021).

The results of research have been accepted for publication in Frontiers in Marine Science, under the title: “Bioprospecting for microorganisms in peloids - extreme environment known for its healing properties” by Vadlja et al. 2022.







BACTERIOPHAGE BASED CONTROL OF VIBRIOSIS IN AQUACULTURE

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The shrimp industry ranks as the highest foreign exchange earner among our marine product exports. Asian countries like Indonesia, Thailand and India have emerged as global leaders in shrimp production. Aquaculture is the most rapidly expanding sector in food production, yet the most vulnerable to losses caused by disease. Among the groups of pathogenic microorganisms, vibriosis is the well-known cause of severe economic losses and responsible for massive mortality of cultured shrimp, fish and shellfish. An intensive mode of culture with high stocking density has resulted in increase in the incidence of diseases. To maintain productivity of such intensive aquaculture massive use of antibiotics has been employed. The spread of antibiotic resistance from aquaculture settings to the natural environment is increasing. About 70% of the Vibrio isolated from aquaculture settings is multi-drug resistant.

Another concern associated with the use of antibiotics is the problem of residues, which has resulted in rejection by seafood importing countries of shrimp containing traces of antibiotics. Alternative strategies must be developed to control diseases in aquaculture. These strategies should reduce the risk of developing and spreading microbial resistance and more environment friendly.

Today, unless concerted action is taken to curb antibiotic overuse and misuse, in humans and animals, the world may be heading for a post-antibiotic age.

Bacteriophage Therapy
Bacteriophage therapy is currently considered as a viable alternative to antibiotics for the treatment of bacterial infections in aquaculture systems. The use of bacteriophages to control bacterial infections in aquatic food production system has the promising potential to address the twin problem of controlling bacterial infections and at the same time avoiding residue contamination.

The application of phages in aquaculture has good advantages over the use of antibiotics.
- This includes the targeted attack of vibrios by phages, phages are abundant in nature, no serious side effect; On the contrary, antibiotics target both pathogen and normal micro-flora, which have multiple side effects;
- Self-replicating therapeutic
- Self-limiting -Phages thrive in the presence of bacteria and die out in their absence.
- “Green Natural Alternative” to antibiotics
- No damage to normal (beneficial) flora

The present study is aimed at assessing the potential advantages of using bacteriophages against vibrios in shrimp culture.

Field Trials:
- Several Field trials showed that the larval survivability in the hatcheries increased markedly with the bacteriophage therapy.
- Use of phage therapy in grow-out culture: Phage cocktail against vibriosis was also mixed with feed and fed to animals which showed that there is a decrease in the vibrio counts in the gut of animals, decrease in running mortality, White faecal disease, increase in survival and increase in Average daily weight gain.
- Bacteriophage-based formulation against vibrios resulted in better growth, survival, FCR and quality of marine shrimp *Penaeus Vannamei*.
    - The trials have demonstrated the potential of vibrio-specific phages to significantly reduce the impacts of vibrios, with a resulting positive effect on shrimp survival. Therefore, phage therapy may be a realistic alternative approach for controlling pathogenic bacteria in aquaculture owing to its several advantages over the conventional antibiotics and other methods against pathogenic multiple drug resistant (MDR) bacteria.

CARACTERIZACIÓN DE EXTRACTOS LIPÍDICOS DERIVADOS DE SUBPRODUCTOS DE CABEZA DE CAMARÓN OBTENIDOS MEDIANTE LA TECNOLOGÍA DE ULTRASONIDO

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Palabras clave: Subproductos, lípidos, rendimiento, ultrasonidos

Introducción. La acuacultura de crustáceos es el sector de producción de productos del mar más grande del mundo y proporciona un suministro de alimentos ricos en proteínas. Los camarones y sus productos se consumen ampliamente en todo el mundo y su demanda aumenta cada año debido a su delicadeza y valor nutricional. La producción mundial de camarón llega a los 5,03 millones de toneladas en 2020 y se espera que aumente hasta los 7,28 millones de toneladas para 2025 con una tasa de crecimiento anual compuesta del 6,1% de 2020 a 2025 .

Generalmente, los camarones una vez de ser capturados de su hábitat se almacenan y en ocasiones se exportan congelados con o sin cáscara, dependiendo de la demanda del mercado. Por lo tanto, durante el procesamiento del camarón, aproximadamente del 50 al 60% de los subproductos del procesamiento del camarón (Senphan y Benjakul, 2012). Además, el proceso de lavado y cocción también genera contaminación de aguas residuales , alrededor de 1 galón por toneladas de camarón cocido (Nirmal y col., 2020). Este subproducto constituye alrededor del 45-50% de la captura y causa contaminación ambiental y problemas de eliminación debido a descargas no reguladas (Sila y col., 2015). Una pequeña cantidad de desechos de camarón se utiliza como alimento para animales y como ingrediente en la formulación de alimentos para la acuacultura. Sin embargo, se están desperdiciando grandes cantidades de este subproducto y a su vez generando un aumento en la contaminación ambiental, lo que resulta en la pérdida de valiosos componentes ricos en compuestos lipídicos.

Hoy en día, la extracción asistida por ultrasonido (EAU) se ha utilizado para extraer lípidos del cefalotórax de camarón con un mayor rendimiento (Gulzar y Benjakul, 2018) y la EAU en condiciones óptimas podrían ser una forma útil de extraer lípidos del residuo sólido del cefalotórax (RSC).

Dado que los RSC puede ser una fuente alternativa de lípidos, el cefalotórax de camarón podría utilizarse plenamente con el concepto de "desperdicio cero". Para mejorar la eficacia de la extracción de lípidos de RSC, se podrían utilizar EAU con las condiciones adecuadas. Por lo que, en esta investigación, se estudia el efecto de EAU (tiempo ultrasónico) sobre el rendimiento de extracción y composición bioactiva de lípidos de RSC de camarón blanco del Pacífico.

Objetivo general. Evaluar y caracterizar el rendimiento de extractos lipídicos derivados de subproductos de cabeza de camarón obtenidos mediante la tecnología de ultrasonido..

Estrategia metodológica. Para el estudio, se propuso una estrategia experimental dividida en tres etapas.
En la primera etapa se estudió el efecto del tiempo (0-50 min) en la EAU sobre el rendimiento de extracción de lípidos a diferentes relaciones de muestra/solvente (1:2, 1:4). Los resultados obtenidos,

Cuadro 1. Contenido de rendimiento lipídico extraídos de RSC por EAU con diferentes relaciones de muestra/disolvente.

	Tiempo ultrasónico (min)					
m/s	0 (control)	10	20	30	40	50
1:2	7.69 ± 0.11 ^{c}	7.93 ± 0.17 ^{c}	8.41 ± 0.16 ^{b}	8.90 ± 0.09 ^{a}	8.94 ± 0.09 ^{a}	8.93 ± 0.08 ^{a}
1:4	7.87 ± 0.05 ^{c}	8.50 ± 0.06 ^{b}	8.88 ± 0.03 ^{a}	8.88 ± 0.16 ^{a}	9.02 ± 0.15 ^{a}	9.12 ± 0.20 ^{a}

Los valores se muestran como media + desviación estándar (n=3). m/s = denota la relación entre muestra y disolvente.
Diferentes letras minúsculas dentro de la misma fila indica diferencias significativas ($p<0.05$).

(Continued on next page)

indicaron que el mayor rendimiento (8.88 g de lípido/100 g de RSC) se podía encontrar a los 20 min en una relación muestra / solvente 1:4. La segunda etapa del estudio consistió en determinar el contenido total de carotenoides (CTC) de los extractos lipídicos obtenidos en la etapa anterior. Los resultados obtenidos, indicaron que el máximo rendimiento (8.25 mg de carotenoides/g lípido) del CTC se encontró a los 20 min en una relación muestra/solvente 1:4.

Finalmente, con el objetivo de evaluar los compuestos asociados a los extractos lipídicos obtenidos de los RSC provenientes de subproductos de camarón, se evaluó el contenido total de fenólicos y capacidad antioxidante. Los resultados obtenidos mostraron que el mayor rendimiento de ambas variables de respuesta medidas se encontró generalmente a los 20 min en una relación muestra/solvente 1:4. Por lo tanto, la EAU con las condiciones adecuadas (20 min en una relación muestra/solvente 1:4) podrían usarse como un método potencial para la extracción de lípidos de los RSC.

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CHARACTERIZATION OF THE OSSIFICATION PROCESS IN THE SILVERSIDE *Chirostoma estor* UNDER CULTURE CONDITIONS

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The silverside *Chirostoma estor* is an endangered fish species native to the Pátzcuaro's Lake, located in the Central Part of Mexico. In this study the ossification process of *C. estor* was evaluated in specimens under culture conditions. Results show that the ossification of the first skeletal structures begin between 30 to 45 days after hatching (dah), and the process concludes between 60 to 100 dah. Therefore, the first two months of life of the species must be closely supervised, since the ossification process is highly susceptible to the influence of biotic, abiotic, and physicochemical factors that may produce skeletal deformities.

For this study, the double staining protocol (Darias *et al*., 2010) was used. Incubation times and temperatures were adjusted accordingly to fish size and morphological characteristics, to achieve optimal stain of cartilaginous and bony tissues.

Fish were obtained from the hatchery at the Instituto de Investigaciones Forestales y Agropecuarias (IIAF), Universidad Michoacana de San Nicolás de Hidalgo (UMSNH).

Fifteen fish were sampled from the following intervals of age: 10 to 30, 30 to 45, 45 to 60 and 60 to 100 days after hatching (dah). Fish were fed according to the protocol described by Martínez-Palacios *et al*. (2002). For newly hatched larvae, the photoperiod applied was 24 h light: 0 h dark. For larvae since 10 dah and juveniles, natural photoperiod (12L:12D) was applied.

The total body length for each of the four sampled groups of age was the following:

Age (days after hatching)	10-30	30-45	45-60	60-100
Total lenght (mm)	5.34-7.58	12.76-22.48	19.19-36.76	41.64-59.30

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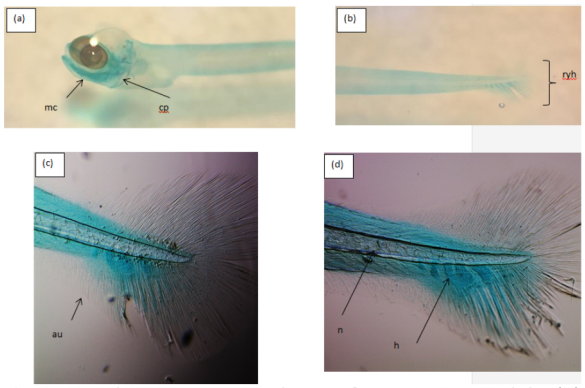

Figura 1. *Chirostoma estor* larvae from 10 to 30 dah. (a) Alcian blue stained cartilaginous tissues from the skull, cl: cleinthrum; mc: Meckel's cartilage (b) caudal fin, ryh: hypural fin rays; (c) larvae in pre-flexion, au: unitary fin; (d) larvae in flexion; n: notochord; h: hypurals.

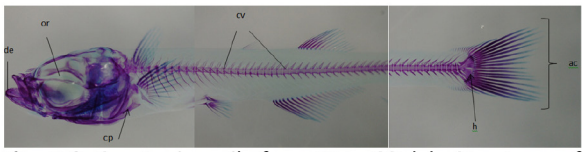

Figura 2. *C. estor* juvenile from 45 to 60 dah. Structures of the axial and appendicular skeleton are almost completely ossified. de: dentary; cp: pelvic girdle; cv: vertebral column; al: cauda fin; h: hypural.

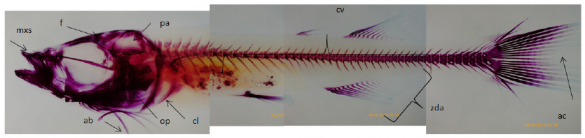

Figura 3. *C. estor* juvenile from 60 to 100 dah. The structures of the axial and appendicular skeleton are now completely ossified. mxs: superior maxilla; op: operculum; pa: parietal; ab: gill arches; cv: vertebral column; zda: distal part of the anal fin; ac: caudal fin.

GREEN ORMER (*Haliotis tuberculata*) TISSUE RESPONSES TO SEASONAL VARIATIONS IN THE NORTHERN ADRIATIC SEA

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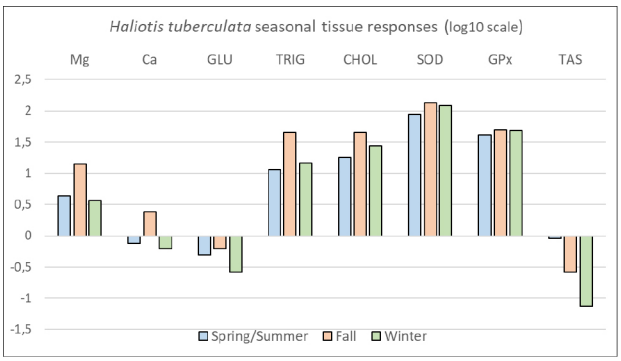
In the Northern Adriatic Sea, tuberculate abalone (green ormer) (*Haliotis tuberculata* L.) are particularly threatened by bottom trawling and seafood disturbance, and their populations face a decline. A low average depth of 30 m, and a weak bathymetric gradient characterizes the Northern Adriatic. The most important oceanographic factors in the Adriatic are seasonal salinity, the concentration of nutrients, light intensity, currents and temperature. Seasonal changes of oceanographic factors, including temperature, algae and wave motion, greatly affect the growth and physiology of green ormers. For this reason, the Northern Adriatic green ormer tissues were studied over the main seasonal periods for their microanatomical structure, biochemical and antioxidant defense properties.

Tissue concentrations of glucose (GLU), triglyceride (TRIG) and cholesterol (CHOL) were determined as the main energy sources for metabolic demands. The activities of superoxide dismutase (SOD), glutathione peroxidase (GPx), total antioxidant status (TAS) were evaluated as parameters for antioxidant capacity and lipid peroxidation. Tissue magnesium (Mg) and calcium (Ca) were investigated as formative shell elements, crucial for ionic homeostasis and reproduction control. Significant differences were found between spring/summer and winter samples for TAS, with peaks in June. GPx had the highest activities in fall samples, just as SOD, CHOL, GLU, TRIG, Mg and Ca parameters. Differences in green ormer tissue biochemical and antioxidant defense properties over seasons are shown in the figure as a log10-based scale. Data were grouped tightly in ormer tissues in spring/summer and winter, having unsymmetrical interquartile ranges. Long-term temperatures between 8.5 and 9.0°C are the lethal limit for ormers, and at low temperatures, they are limited in their ability to absorb food and grow. Winter low temperatures thus contributed to lower TAS and SOD concentrations. Not only antioxidant capacity, but also tissue Mg, Ca and GLU were on the decreased levels in ormers in winter. However, in fall a number of parameters had their yearly peaks, such as Mg, Ca, SOD, GPx, TRIG, CHOL and GLU. Interestingly, in this work ormers had elevated total antioxidant status in the warmer period, which can be attributed to the availability of food containing antioxidant compounds. During the spring/summer season, gonads were at the mid-maturation stage, and in fall, the ripe and spawned stages were recorded.



DISTRIBUTION OF MARINE MUSSELS SPECIES OF THE GENUS *Mytilus* ALONG THE CHILEAN COAST

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The chilean native blue mussel *Mytilus chilensis*, is an economically important marine resource in Chile. The *M. chilensis* aquaculture began in 1943 in Chiloé Island, southern Chile, and the aquaculture production increased rapidly from 3,864 t in 1993 to 398,274 t in 2018, equivalent of 29,3% of the total Chilean aquaculture production. Therefore, and because the relative recent description of two species of mussels *Mytilus galloprovincialis* and *Mytilus edulis* in chilean waters, there is a need to carry out a surveys to establish the actual distribution of these two exotic mussels along the Chilean coast and evaluate their actual and potential effects on the biodiversity along the coast and on the fasting growing mussel (*M. chilensis*) aquaculture industry. Once the evidence that these mussel species arrived to a coastal location it is important to study their distribution and urgently to investigate their origin and its potential for bioinvasion along the chilean coast.

Samples of 30 adult mussels from 36 locations along almost 2,500 km of Chilean coast were analyzed using two nuclear markers (*Me 15-16 and Myti*) and two mitochondrial markers (*16S and COIXba*). The results indicate that from the 1030 mussels analyzed 76% belongs to the Chilean native *M. chilensis*; 17% to the mediterranean mussel *M galloprovincialis*, 4% to the northern hemisphere *M. edulis* and only 2% to hybrids between these species of mussels. The highest number of hybrids were detected between *M. edulis* and the native mussel *M. chilensis*. Also samples of cultured mussels from 8 aquaculture production centers were analyzed, within the X Region (southern Chile), where over 95% of the Chilean mussel aquaculture production is obtained. Among these mussel aquaculture centers, the results indicated that 96% of the mussels were the native Chilean mussel *M. chilensis*, 3% *M. galloprovincialis* and 1% of hybrids. Interesting is that among these hybrids, there were *Mytilus trossulus* alleles detected (a mussel species that inhabits the cold waters in the northern hemisphere), as described earlier by other authors. Also a high plastic variability among mussels shell (different morphology and coloration) within pure species was observed which precludes the use of these phenotypic traits in taxonomy. Two of the mussels detected can be detrimental for the chilean mussel aquaculture. *M. galloprovincialis* has been declared one of the world´s 100 worst invasive species and *M. trossulus* have lower rates of somatic tissue and shell weight.

Research founded by FONDECYT 1170194.

REPLACEMENT OF FISH MEAL BY ANTARCTIC KRILL MEAL IN DIETS FOR EUROPEAN SEA BASS *Dicentrarchus labrax*: EFFECTS ON GROWTH PERFORMANCE, FEED UTIZATION AND LIVER METABOLISM

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A sustainable growth of the aquaculture sector implies the use of sustainable novel raw materials as replacers of the traditional fishmeal (FM) and fish oil (FO) ingredients. This fact has led to the development of sustainable and functional diets as part of a management strategy to reduce the effects on fish growth performance and health derived from low FM/FO dietary contents. In this sense, Antarctic krill (*Euphausia superba*) is considered a potential candidate in dietary inclusions to potentiate fish growth and health status.

In this study, European sea bass (*Dicentrarchus labrax*) were fed a practical diet with either a 15% fishmeal content (KM0; control diet) or the same diet substituted by 30% (KM5; 50 g KM/kg diet) or 50% (KM7.5; 75g KM/kg diet) Antarctic krill meal (KM) for 12 weeks in triplicates. At the end of the feeding trial, growth performance, liver morphology, liver proximate composition, lipid classes and fatty acid profiles, as well as the expression of hepatic genes related with lipid metabolism were evaluated.

Fish fed KM-based diets presented higher ($p<0.05$) final weight, protein and lipid efficiency ratios, specific growth rate (SGR) and improved feed conversion ratio (FCR), irrespective of the KM dietary level. Whole body and muscle proximate composition and fatty acid profiles were similar among dietary groups. Livers of European sea bass fed the experimental diets presented similar ($p>0.05$) biochemical composition and fatty acid profile. However, smaller hepatocellular area and lower grade of cytoplasm vacuolization as well as a better alignment around sinusoidal spaces were found. The analyses of liver lipid classes revealed a positive correlation between the level of dietary KM and the pigmented material such as astaxanthin and free fatty acid content, as well as a negative correlation with the cholesterol levels. The expression of hepatic genes studied demonstrated a downregulation of *3*-hydroxy-3-methylglutaryl-coenzyme A reductase *(hmgr)* and delta-6-desaturase (*fads2*) expression levels in fish fed KM-based diets. Besides, gene expression levels of fatty acid binding protein 7 (*fabp7*) and lipoprotein lipase (*lpl*) were significantly correlated with KM dietary levels. Altogether, these results profile KM as a potential promoter of growth and liver health in European sea bass fed low fish meal and oil diets.

PROINMUNOIL PLUS: FUNCTIONAL FEED ADDITIVES IN LOW FISH MEAL AND FISH OIL-BASED DIETS FOR EUROPEAN SEA BASS *Dicentrarchus labrax*: EFFECTS ON MUCOSAL HEALTH

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An effective replacement for fish meal (FM) and fish oil (FO) based on plant-based raw materials in the feed of marine fish species is necessary for the sustainability of the aquaculture sector. One of the main objectives of PROINMUNOIL PLUS project was to promote European sea bass (*Dicentrarchus labrax*) mucosal health through the use of feed additives in low FM and FO diets. To achieve this objective, the effect of several feed additives included in low fish meal (10%FM) and fish oil (6%FO)-based diets, on gill and gut mucosal health as well as in disease and stress resistance of European sea bass were evaluated through a series of experiments. Three experimental diets were tested along PROINMUNOIL PLUS project, one was void of the test ingredients (control diet), while the remaining two contained 5,000 ppm of galactomannan oligosaccharides (GMOS; Delacon, Austria) and 200 ppm of a blend of garlic and labiatae-plant oils (PHYTO; Delacon, Austria), respectively. Diets were supplemented for 63 days before exposing the fish to an intestinal *Vibrio anguillarum* infection combined with crowding stress. In order to evaluate functional diets efficacy in mucosal health maintenance, structural, cellular, and immune mucosal tissues status were evaluated by optical and electron microscopy, gene expression and microbiota analyses.

The main results of the project concluded the specific role of each additive in maintaining mucosal health or promoting disease or stress resistance for this species. Even more, the effects detected were not only tissue specific but also region specific within the same tissue. Feeding with functional diets did not affect growth performance; however, feed additives reduced European sea bass susceptibility to V. anguillarum and acute stress cortisol levels. Functional additives did not affect gut folds morphometry but reduced the posterior gut fold area covered by goblet cells. PHYTO reduced posterior gut goblet cell size and increased rodlet cell density, whereas dietary GMOS reduced submucosal thickness and increased rodlet cell density in rectum. Structural TEM analyses revealed increased rectum microvilli length after feeding GMOS, whereas the use of PHYTO increased *Ocln*, *N-Cad* and *Cad-17* posterior gut gene expression. In terms of microbiota populations, the main beneficial effects of dietary GMOS and PHYTO were the reduction of coliforms and Vibrionales bacteria and the enrichment of gut microbiota composition with butyrate producer taxa. In gills, GMOS and PHYTO downregulated the expression of *sod*, *gpx*, cat and *gr* compared to gills of fish fed the control diet. The morphological, histopathological, immunohistochemical, and biochemical parameters of the fish gills were mostly unaffected by GMOS and PHYTO. However, the PHYTO group the lowest incidence of lamellar fusion, whereas GMOS reduced gill goblet cell size. Both additives significantly reduced the concentration of PCNA+ cells in gills epithelia.

REFUGE PREFERENCE AND ITS RELATIONSHIP WITH GROWTH AND SURVIVAL RATES OF JUVENILE *Octopus maya* IN CULTURE

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The *Octopus maya* octopus is endemic to the Yucatan Peninsula and is one of the most important fishing resources in the area. At the UMDI Sisal of the UNAM (Yucatán, Mexico) various aspects of the species aimed at its cultivation have been investigated. Octopuses are solitary animals that shelter in shelters mainly to avoid being preyed upon. In this study, the preference of *O. maya* juveniles for a refuge was evaluated, a preference ranking was carried out and in a second bioassay growth and survival were evaluated by comparing the preferred refuge and the least chosen refuge.

Post-hatching of *Octopus maya* from a single spawning were used. In experiment 1, an individual was placed in the center of an experimental arena containing five different types of shelters (gastropod shell, artificial raffia grass, PVC tube, PVC elbow, and artificial cave) of two different sizes. After 24 h, the refuge in which the octopus was found was recorded and this was the preferred refuge. The selected shelter was removed from the arena and the octopus was later placed back in the center of the experimental arena for 24 h for a new choice. The same procedure was performed to determine an order of preference.

In experiment 2, the growth and survival rates of octopuses kept under two different treatments were compared: octopuses with their preferred refuge and octopuses last selected refuge.

Table 1. Relationship of the refuge preference sequence (n=13 octopuses). The dots (octopuses) show the number of times each shelter was chosen as the first to fifth choice when all five types of shelters were presented simultaneously.

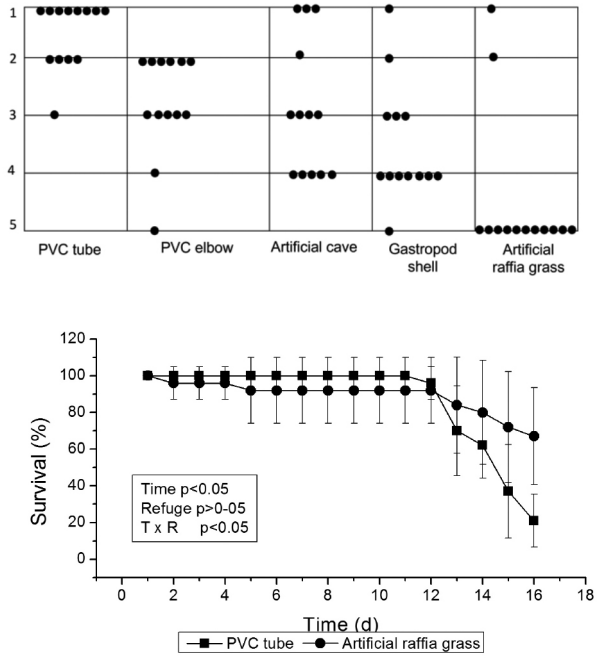

Figure 1. Survival (%) of *O. maya* juveniles from day 1 to day 17 in each treatment (tube and raffia). (Mean + SD) n = 3.

The octopuses chose PVC tube as their preferred shelter, while they chose raffia artificial grass as the last option (Tabla 1).

In the 2nd bioassay, a similar growth rate was obtained (average 3.5% day-1) ($p>0.05$).

Survival at day 16 was greater in the raffia treatment (65.5%) than in the PVC tube treatment (18%). However, the statistical analysis showed that there is no relationship between the refuges and the survival of the individuals. Regarding growth, no differences were observed between the two treatments ($p>0.05$).

Acknowledgments
This work was carried out with the support of the DGAPA-UNAM, Project PAPIIT IT 201621 under the responsibility of Dr. Pedro Gallardo

ECTOPARASITES IN *Seriola rivoliana* PRODUCTION IN THE GULF OF CALIFORNIA: CHALLENGES AND AREAS OF OPPORTUNITY

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Mariculture of *Seriola rivoliana* in the Gulf of California, México is subject, like other marine finfish species, to parasitic infestations. Managing them is a central issue for health and welfare. The main parasite species affecting *S. rivoliana's* growth cycle is *Neobendenia* spp. The Gulf of California is a natural habitat of *S. rivoliana* and is therefore ideal for its production. However, given the large number of native fish species in the area (about 911 documented species) the probability of encountering parasite vectors is correspondingly high. In the specific case of *Neobenedenia* spp, it has been reported to infect about 100 species of 30 different families, and 5 different orders of fish in the world. For this reason, total prevention of parasitic infestations in this circumstance is practically impossible. Moreover, *Neobenedenia* spp is a parasite with a direct life cycle. The adult affixes to its host and lays filamentous eggs into the water. These hatch into ciliated larvae (oncomiracidia) that can re-infect rapidly the same host, and the filaments of the eggs allow them to entangle to any available surface. Typical management methods used to control monogeneans parasites infecting fish are freshwater baths, oral administration of the anthelmintic praziquantel, and hydrogen peroxide or formalin baths. In this work we present results of different treatments applied in the course of two and half years. We also evaluate different management strategies for the specific conditions of our production site. The main goal is to determinate the best strategy that will balance animal welfare, certification standards, production costs and also minimizing environmental impact. We share the challenges and areas of opportunity that we have identified throughout this process.

FUNCTIONAL GROWER SHRIMP FEED IN ECUADOR AND MEXICO INCREASED SURVIVAL IN *Litopenaeus vannamei* DURING 2020 – 2021

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Enteric disease in whiteleg shrimp *Litopenaeus vannamei* are presenting during the whole year and distributed worldwide, especially in Ecuador and Mexico shrimp farming facilities. Generic pathogen *Vibrio* spp. can cause mortality and poor performance of farmed shrimp, affecting economically shrimp industry. Although there is not an economical estimate only for bacterial diseases, in general the impact of diseases caused in the industry is estimated in about 22% production losses annually (Flegel *et al*., 2008).

Cargill developed a functional feed concept called SmartShield to tackle enteric diseases in *L. vannamei* for starters feeds which is commercialized in both countries, Ecuador and Mexico since 2019. Latest developments of postbiotics and phytogenics brought a new SmartShield concept for grower feeds which was assessed commercially with strategic customers between 2020-2021.

The blend of additives has a total inclusion level of 6.0% in feed and in is based in a high nutritional density feed. This feed was assessed in Ecuador and Mexico in field trials with records of bacterial diseases during different cycles of production. First trial was performed in Ecuador, where shrimps were allocated in 13 ponds (~10 Ha/pond) with a stocking density of 15 shrimp/m^2, distributed in 7 ponds for regular feed and 6 ponds for SmartShield feed. Shrimps were fed with regular and functional feeds, respectively, since 1.5 g/shrimp until 95 days before harvest.

Second trial was done in Mexico, in Sonora state, where shrimps were allocated in 8 ponds (5 Ha/pond) with a stocking density of 20 shrimp/m^2. Each group of shrimps were fed with a regular feed or functional feed since 3 g/shrimp until 61 days before harvest.

Results showed that additional survival of animals fed with SmartShield were 5% and 8%, in Ecuador and Mexico respectively, both with statistically significant differences ($p<0.05$).

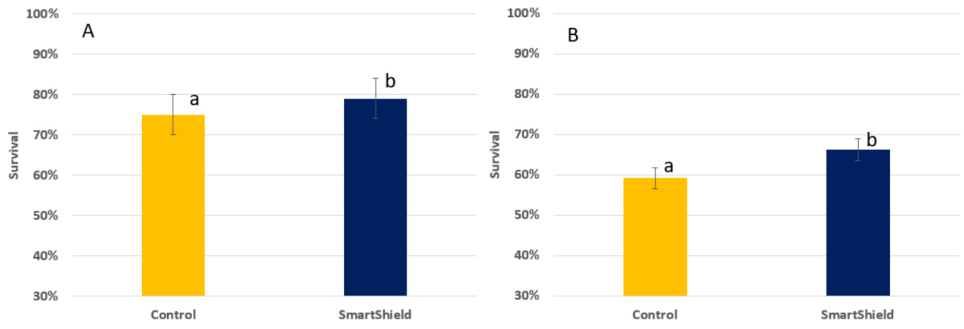

Figure 1. Shrimp survival obtained in trials done in Ecuador (A) and Mexico (B) when animals fed SmartShield.

TILAPIA LOVE MILK OF MAGNESIA: STRONG, GENTLE SUPPLEMENT FOR CLOSED-LOOP AQUACULTURE SYSTEMS

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A land-based closed-loop tilapia farm in British Columbia, Canada was struggling with fish health due to insufficient pH control and accumulation of nitrogen-based contaminants in the water system. To address the problem the farm installed a membrane bioreactor (MBR) system to maintain low levels of contaminants.

They used hydrated lime ($Ca(OH)_2$) for pH control, which was very labor intensive, caused plugging of feed lines, and did not effectively control pH – especially when the fish were being fed. As a result, nitrate levels in the water were reaching a danger point for the fish. In addition, they were needing to supplement with table salt (NaCl) in order to keep a healthy protective slime layer on the fish bodies.

This study provides data on how the replacement of lime with a magnesium hydroxide-based product resulted in strong pH control and nitrogen reduction, along with dramatic improvements in water clarity and fish health. The nonhazardous liquid product, called AMALGAM-30, did not require operator maintenance, providing reliable, steady feed for optimum pH and alkalinity control. After three weeks of operation the nitrate levels in the water system were reduced by 89% and the water clarity (turbidity) was dramatically improved. They stopped supplementing with sodium chloride.

Perhaps the most dramatic observation from this study was the overall improvement in the health of the fish. The protective slime layer on the tilapia improved greatly when the feed of lime and table salt was replaced with AMALGAM-30, with noticeably less cuts and abrasions on the fish. The improvement in health was so noticeable that they received numerous comments from buyers about the improved fish quality and dramatically improved water clarity. As a result, the farm has completely discontinued the use of lime and salt, and has installed a permanent AMALGAM-30 storage and feed system.

GENETIC AND GENOMIC TOOLS FOR ABALONE SPECIES IDENTIFICATION FROM THE BAJA CALIFORNIA PENINSULA

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In last decades, genomics has allowed the development of a wide range of tools to identify and improve the number of informative SNP markers, mainly in non-model species. These molecular markers have been extensively used in species identification, parental assignment, characterization of genetic variability, forensic analysis, traceability, and other applications. However, the use of many markers for species identification can become impractical for routine applications. Otherwise, using a single molecular marker is not always sufficient and can sometimes lead to erroneous conclusions. In the present study, we searched for the most informative SNPs that enable discrimination among six abalone species: green (*H. fulgens*), pink (*H. corrugata*), red (*H. rufescens*), black (*H. cracherodii*), white (*H. sorenseni*), and pinto/threaded abalone (*H. kamtschatkana assimilis*). These abalone species inhabit the northeast (NE) Pacific coast and have high commercial and ecological importance in the Baja California peninsula, Mexico. SNP markers were identified by RADseq. Unique SNPs for each species were evaluated and selected for their potential use in species assignment, as a rapid identification tool for routine monitoring of fishery products. Applying these species-specific SNPs in a standardized assay permit the traceability of these abalone species. This assay will contribute to the management and conservation of this natural resource. Ultimately this work will be patented and developed as a commercially available kit to identify abalone species from the NE Pacific region.

PHYSIOLOGICAL CHARACTERIZATION OF *Euchuematopsis isiformis* UNDER LABORATORY CONDITIONS: STEPS FOR SCALING UP OUTDOOR CULTURES

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The demand for carrageenan, a sulfated polysaccharide obtained from Eucheumatoids species (*Kappaphycus*/ *Eucheuma*) widely used as a food thickener, has increased in recent years. Efforts to cultivate these carrageenan-producing species in the Americas have been successful in Brazil and some Caribbean islands with the introduced species *K. alvarezii* (Hayashi et al., 2017). In Mexico, *Eucheumatopsis isiformis* (formerly *Eucheuma isiforme*) a native species found in the southern Gulf of Mexico and the Caribbean produces a high-quality iota carrageenan suitable to be considered a viable source of carrageenan with yields up to 40% of its dry weight (Freile-Pelegrín and Robledo, 2006). Under the support of the MARINER ARPA-e DoE program, the "Tropical Seaweed Farm" project aims to design and develop a cultivation system for large-scale production of this tropical seaweed. To achieve this, it is necessary to select and domesticate cultivars of *E. isiformis* and basic physiological knowledge is crucial to understand the conditions for maximum growth and biomass production. In this study, morphotypes of *E. isiformis* were collected from a subtidal population in Yucatan (Dzilam de Bravo) and acclimatized to laboratory conditions (23°C + 80 µmol quanta m^{-2} s^{-1}). Physiological responses (photosynthesis and growth) were determined at different light intensities: low, LL (80 µmol quanta m^{-2} s^{-1}); medium, ML (250 µmol quanta m^{-2} s^{-1}) and high, HL (550 µmol quanta m^{-2} s^{-1}). Preliminary results have shown that one to two weeks are sufficient to acclimate the photosynthetic apparatus to each of the light conditions and that light levels had a strong effect on the photosynthetic parameters analyzed (maximum photosynthesis, respiration rates and saturation irradiance). Specimens under ML and HL showed thallus elongation and development of numerous lateral branches. The results suggest that *E. isiformis* is able to tolerate higher irradiances under cultivation conditions, up to twice its photosynthetic saturation irradiance (Ek), without nutrient limitation. Preliminary chemical analyses of *E. isiformis* morphs revealed differences, including carrageenan content, so understanding the factors controlling their changes should be a priority. Further experiments are underway to extend these cultivars to outdoor conditions.

Funding: ARPA-e DE-AR0000912 Project Cinvestav Subward 54336.

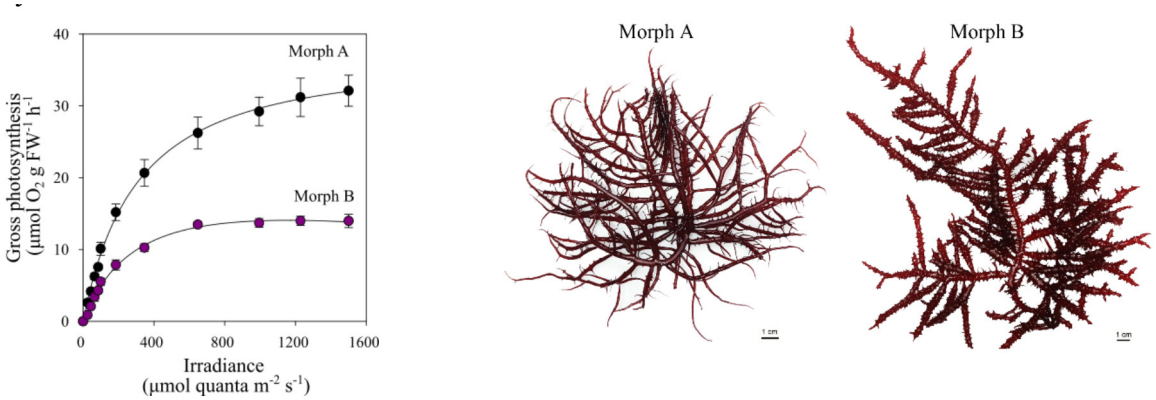

Fig. 1. Photosynthesis to light response curves of *E. isiformis* morphs.

EFFECT OF METHIONINE SOURCES AND *Bacillus amyloliquefaciens CECT 5940* ON GROWTH PERFORMANCE OF SHRIMP FED REDUCED FISHMEAL DIETS

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Limited availability of fishmeal (FM) and increasing disease pressure are two key challenges in the sustainable growth of shrimp farming. To successfully reduce FM, an appropriate balance of amino acids (AA) and all essential nutrients are required. Methionine (Met) is typically the first limiting AA which is commonly met with the use of supplemental Met sources. A dipeptide of DL-Met (DL-Methionyl-DL-Methionine) or in short 'Met-Met', was shown to be the most effective Met source for shrimps due to its very low water solubility/ leaching and better utilization. As an alternative to antibiotics, an in-feed probiotic based on *Bacillus amyloliquefaciens* CECT 5940 (Ecobiol®) have shown to improve the health and performance of shrimps. However, the combination of these strategies has not been fully explored. A trial was conducted to determine the effects of Met-Met and Ecobiol® on growth performance of whiteleg shrimp (*Litopenaeus vannamei)* fed reduced FM diets. A total of 480 shrimps (initial BW = 0.30 ± 0.04 g) were randomly distributed to 24 tanks (150 L each) and fed 6 dietary treatments (4 replicates) for 54 days. Treatments in pellet form were fed 3 times per day including: D1) positive control diet (PC) with 20% FM and no supplemental Met, D2) reduced FM diet with 10% FM (RFM) & 0.13% DL-Met, D3) RFM with 0.06% Met-Met, D4) RFM with 0.19% Met-Met, D5) RFM & 0.13% DL-Met + 0.1% Ecobiol®, and D6) RFM with 0.06% Met-Met + 0.1% Ecobiol®. Diets were isoenergetic and to the same level of essential AA as in the PC (total Lys = 2.11%). Met-Met was used with a Met bioefficacy (BE) of 200% and all diets had the same Met:Lys and Met+Cys:Lys ratios achieved in D1 (33 and 54% respectively) except for D4 that had a greater Met:Lys and Met+Cys:Lys ratios (45 and 66% respectively). Data were analyzed by ANOVA and pre-planned contrast statements. Reduction of FM from 20 to 10% with an adequate balance of Met and other nutrients did not affect ($P > 0.05$) any of the growth performance parameters (D1 vs. D2 and D3). Replacing DL-Met with 50% Met-Met (D2 and D5 vs. D3 and D6), improved final BW and biomass gain ($P < 0.05$) and tended ($P = 0.06$) to reduce FCR suggesting that the BE of Met-Met could be greater than 200%. Increasing the Met+Cys:Lys from 44 to 66% (D3 vs. D4) resulted in numerical greater final BW and reduced FCR. Ecobiol® supplementation (D5 and D6 vs. D2 and D3) improved ($P < 0.05$) final BW and biomass gain regardless of the Met source used. These results demonstrate that FM levels can be reduced while maintaining an appropriate balance of AA and other nutrients. Increased Met+Cys:Lys resulted on improved performance and survivability. Met-Met is a superior Met source compared to DL-Met with a BE of at least 200% while Ecobiol® supplementation improved final BW and biomass gain. Both strategies (Met-Met and Ecobiol® supplementations) added to a reduced FM diet resulted on improved growth performance compared to a diet with high FM level in shrimps.

Table 1. Growth performance of shrimps fed experimental diets with reduced fishmeal (RFM) supplemented with DL-Met, Met-Met and Ecobiol® for 54 days

	D1	D2	D3	D4	D5	D6
Parameters	**PC**	**RFM + 0.13% DL-Met**	**RFM + 0.06% Met-Met**	**RFM + 0.19% Met-Met**	**RFM + 0.13% DL-Met + Ecobiol**	**RFM + 0.06% Met-Met + Ecobiol**
Final BW (g)	5.00^{ab}	4.60^{a}	5.34^{bc}	5.84^{c}	5.27^{abc}	5.98^{c}
Biomass gain (g)	72.1^{ab}	61.8^{a}	87.4^{bc}	104.6^{c}	82.7^{abc}	102.5^{c}
Feed intake (g/shrimp)	7.82^{ab}	7.01^{a}	8.33^{abc}	9.43^{bc}	8.56^{abc}	9.81^{c}
FCR	2.02^{ab}	2.14^{a}	1.87^{ab}	1.79^{b}	1.99^{ab}	1.86^{b}
Survival	77.5^{ab}	73.7^{a}	86.2^{ab}	95.0^{b}	83.7^{ab}	91.2^{ab}

REFERENCE POINTS FOR COMMERCIAL GROW OUT FOR RED DRUM (*S. ocellatus*) IN YUCATAN, MEXICO

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High protein food products that can be offered consistently and the generation of jobs are pillars for the development of Mexican aquaculture. The project aims to produce high quality seafood based on reference points for commercial red drum grow out in Yucatan, characterized by a production unit for 132 tons/year of fresh and eviscerated 1 kg product at harvest, for the regional or international market.

At present, there are no commercial operations that can undertake the production of red drum. The business plans comes from a join effort from the Government of Yucatan in alliance with the Universidad Marista, which provides technical support and the production of high quality seed. The objective is to promote aquaculture as an investment option for suitable development areas in the northern region of the State.

An initial investment of $9,428,531.04 (US$460,000) for infrastructure and $11,750,865.65 (US$573,000) for working capital generates a quasi-utility of $2,777,570.61 (US$135,500) with an IRR of 14% and a 5-year NPV of $583,989.38 (US$28,500).

EVALUATION OF THE SANITIZING EFFECTIVENESS OF A COMMERCIAL QUATERNARY AMMONIUM-GLUTARALDEHYDE (PECDESIN® 4G) APPLIED TO SHRIMP´S POND WATER BY ADOPTING AN ATTRIBUTE SAMPLING

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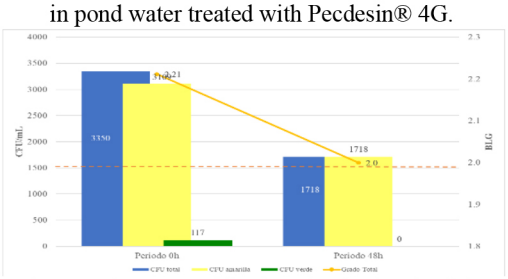
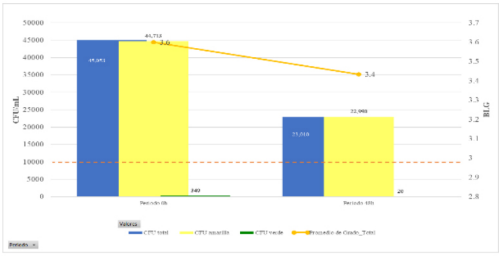
World shrimp production reached a record of 114.5 million tons of live weight in 2018, however, this issue is a concerned down to health on shrimp's health. Strains of *Vibrio* spp. continued as an important pathogen for the penaeid shrimp causing illness and mortality (i.e. hepatopancreatic necrosis syndrome). Disinfection had been a tool for prevent and control these bacterial diseases. Combination of quaternary ammonium salts (QAC) and glutaraldehyde (GLU) are two of the most widely used disinfectants on shrimp's ponds to maintain low overgrowth of *Vibrio* spp. This study evaluated the sanitizing effectiveness of an applied QAC-GLU combination on water ponds by adopting an attribute sampling and analytical criteria.

The study was done in a shrimp fattening farm located at the south of Sonora (25 ponds distributed in 75 ha with common estuary). During June to August 2021, all ponds were treated with a commercial QAC-GLU (Pecdesin® 4G) applying 1 liter/ha[3] (23 ppm) as previously recommended[1,2,3,] then, three ponds were selected for analysis through 20 water samples per pond (100 mL). In addition, hepatopancreas from 15 shrimp were sampled in Whirl-Pak® bags; both samples were collected twice at 0 and 48 h post treatment for pH, salinity, QAC residual and counting of native *Vibrio* spp (green and yellow ones). Bacterial counts were converted to $\log_{10}$ and then adjusted to a score scale (0 to 5+) while only physicochemical values were adjusted to score scale (0 to 5+). The analytical criteria method considered as satisfactory (for overgrowth control) if more than 90% samples achieve up to 2.0+ and 3.0+ bacterial load grade on both water and hepatopancreas, respectively, as well as satisfactory (for product application) if more than 50% samples achieve up to 1.0+ QAC score scale on water. A comparison between consecutive cycles were done to analyze productive performance using the sanitization mentioned. Graph 1 and 2 shows the mean count as bars (CFU/ml) and bacterial load grade as slope (BLG) obtained in water (w) and hepatopancreas (hp).

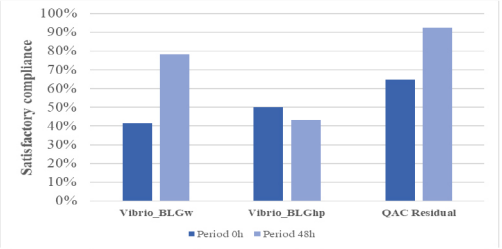
Graph 1. Total counts and bacterial load grade of *Vibrio* spp. in pond water treated with Pecdesin® 4G.



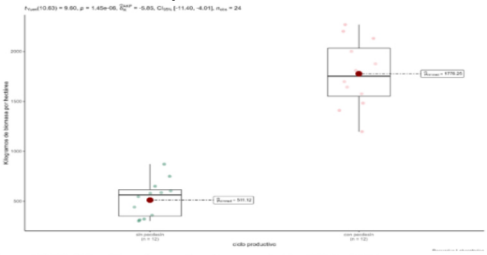
Graph 2. Total counts and load grade of *Vibrio* in shrimp's hepatopancreas of ponds treated with Pecdesin®4G.



Graph 3. Compliance percentage achieved during treatment with Pecdesin® 4G.



Graph 4. Productive performance during treatment and non-treatment cycles with Pecdesin® 4G.



1 Lightner 1993. Handbook mariculture vol. 1, CRC: 432.
2 Sung *et al.,* 2003. Aquaculture 219 (1-4): 123-133.
3 Avunje&Lalitha 2018. Proc.Water Soil Treat.Aquac.ICAR: 55-57.

IN VITRO EVALUATION OF THE RESIDUALITY AND SANITIZING EFFECT WITH DIFFERENT CONCENTRATIONS OF A QUATERNARY AMMONIUM-GLUTARALDEHYDE (PECDESIN® 4G) PRODUCT IN MARINE WATER

Venegas, J. Ana*[1], Chavez, P.J.[2,3], Pantoja-Nuñez, G.[2], Luevano, A.J.[2] and Acuña, A.M.[2]

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Bacterial strains of genus *Vibrio* spp. can cause severe mortality as in early death syndrome or acute hepatopancreatic necrosis syndrome. Disinfection is an important and necessary tool for the management and control of diseases in aquaculture farms dedicated to shrimp fattening. Combination of quaternary ammonium salts (QAC) and glutaraldehyde (GLU) are two of the most widely used disinfectants on shrimp's ponds to maintain low overgrowth of *Vibrio* spp. The objective of the study was known the *in vitro* residuality and sanitizing effect of an artificial challenge with *Vibrio parahaemolyticus* in seawater using different concentrations of a quaternary ammonium and glutaraldehyde commercial combination (QAC+GLU).

Through an experimental study, were compare four treatments with repetitions and measurements of the CFU/mL reduction at different time intervals using an artificial challenge as previously described. QAC+GLU solutions in 1L of sterile water were prepared separately in Erlenmeyer flasks with different concentrations of the commercial product being the low concentration 117 ppm (T1), the medium concentration 334 ppm (T2) and the high concentration 668 ppm (T3) while a nontreated 0 ppm (T0) was include a control. Then a pure culture of an overnight *Vibrio parahaemolyticus* (VPH) strain SO-11 (AHPND) (donated by Dr. Ricardo Sánchez-Díaz, ITSON) was collected to prepare a bacterial suspension adjusting its turbidity to the 0.5 McFarland scale. Then, serial dilution of this suspension was done and 100 μL of each dilution was inoculated onto Petri dishes with TCBS agar. All treatments were challenged at a concentration of 2.82×10^6 CFU/mL of VPH. Finally, 100 μl was seeded in each flask and counting for viable bacteria on agar plate at of 0, 24, 48, 72 and 192 hours (h). Variation in QAC concentration was carried out using a colorimetric technique with reactive strip (MQuant™ Merck KGaA). The concentrations were annotated according to the colorimetric concentration scale and then converted it to ordinal scale score as previously described; the scale was adjusted to values of 0, 1, 2, 3, 4, 5 and 6, having as quaternary ammonium concentration of 0 ppm, 10 ppm, 25 ppm, 50 ppm, 100 ppm, 250 ppm and 500 ppm respectively. The pH of each treatment was measured with the help of a potentiometer and the salinity with a refractometer.

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Table 1. Concentration of quaternary ammonium (ppm) in each treatment at different periods.

	TIME				
	0 h	**24 h**	**48 h**	**72 h**	**192 h**
Nontreated	0	0	0	0	0
117 ppm	61.11	31.25	26.39	21.67	21.79
334 ppm	141.66	137.5	44.44	44.44	50
668 ppm	347.22	312.5	191.67	175	196.43

EVALUATION OF CALIFORNIA YELLOWTAIL *Seriola dorsalis* FED PRACTICAL DIETS FROM PARTIAL TO TOTAL FISH OIL AND FISH MEAL REPLACEMENT

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From the environmental point of view, aquaculture diets must be, avoiding the use of marine resources, increasing diet efficiency to improve production management. For this to happened, it is crucial the increase of unused by-products. For this reason, it is essential the fish oil (FO) and fish meal (FM) replacement by using protein and oil sources from recycle by-products such as terrestrial proteins and fats from husbandry animals. California yellowtail Seriola dorsalis, is a promising fish species to be culture. In fact, several *Seriola spp*. fish species are already produced in around the world with great success. However, being a carnivore species, a high demand of FM is needed. Therefore, the study to replace FM is crucial.

This study aimed to evaluate the effect of partial to total replacement of fish meal (FM) and fish oil (FO) from diets formulated to yellowtail (*Seriola dorsalis*) using poultry by-product meal (PBM) using beef tallow supplemented with DHA. Four experimental diets were formulated to be isoproteic and isolipidic, based on the yellowtail nutritional requirements. Cholesterol was added to compensate its content in the FO. *S. dorsalis* juveniles (14.54g ± 0.19 g, mean ± SE) were randomly distributed into 12 tanks, with 500L each, connected into a recirculation system. After 48 days experimentation procedure, no significant differences were observed in performance for the PBM in replacement of FM. Significant differences were observed in the fatty acid profile of muscle tissue among treatments. Saturated fatty acids were less retained indicating a major use as energy source. It was observed a lower DHA level in fish from T-Low compared those from T-Med.

It is concluded that PBM can efficiently replace FM and FO in diets for *Seriola dorsalis* without any negative impact. However, more experiments are needed to find better fatty acid combination to improve the protein sparing effect.

Table 1. Ingredient composition (%) and proximate analysis, of diets formulated to contain 45% crude protein (CP) and 12% crude fat (CF), to replace FM by PBM and bovine tallow enriched with DHA. Diets were fed to juvenile *Seriola dorsalis*, for 48 days.

	TREATMENTS			
Ingredients	Control	PBM-Low	PBM-Med	PBM-Total
Fish meal	21	14	7	0
Poultry by product meal	22	29.3	36.6	44
Beef tallow	0	2.7	2.8	2.9
DHA (24% DHA)	1	2.8	3.4	4
Fish oil	6	1.8	0.9	0
Others [r]	50	49.4	49.3	49.1
Proximal composition (% dry matter)				
Crude protein (%)	45.37	45.32	45.31	45.30
Crude fat (%)	12.08	12.10	12.09	12.11
Ash (%)	2.5	2.6	2.7	2.7

Table 2. Biological indices of *Seriola dorsalis* fed for 48 days four different diets from partial to total substitution of fish meal and fish oil. Mean values and standard deviation (n=3) are given. Values in the same row with a different superscript were significantly different (P <0.05).

Biological indices	TREATMENTS				
	Control	PBM-Low	PBM-Med	PBM-Total	*P Value*
Initial weight (g)	14.51±0.02	14.30±0.14	14.77±0.26	14.55±0.18	0.122
SGR (%/day)	2.28±0.06	2.02±0.13	2.21±0.22	2.05±0.11	0.170
FI (% day $^{-1}$)	2.95±0.11	3.09±0.02	2.99±0.10	2.95±0.29	0.723
FCR	1.39±0.06	1.61±0.08	1.45±0.16	1.52±0.10	0.177
PER	1.59±0.07	1.37±0.07	1.53±0.18	1.46±0.10	0.203
Survival (%)	100	100	100	100	

AN EVALUATION OF AERATION EQUIPMENT TYPE AND TIME ON THE ECONOMIC IMPACT OF INTENSIVE PHOTOHETEROTROPHIC SYSTEMS WITHOUT WATER EXCHANGE FOR THE WHITE SHRIMP *L. vannamei*

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The drop in shrimp production in Mexico has been associated mainly to diseases, outdated infrastructure and the use of obsolete technologies. To counter this, bio secure and economically efficient systems are needed. The photoheterotrophic culture is a hybrid system that mixes heterotrophic, chemotrophic and autotrophic microorganisms where a balance is struck in the ecosystem between nutrients, available dissolved oxygen and a capacity for particle transportation, through aeration. Aeration is fundamental to stabilize the system, minimize water exchange so that culture is optimized. However, the cost for its use can be significate. The object of the present research was to characterize the impact of different aeration equipment for the intensive photoheterotrophic production of shrimp, establish the effect of a reduction in aeration time and characterize the impact on the physiological response of the organism. A spring-summer culture cycle was conducted to generate information on the continuous use of different aeration equipment on water quality, survival, food conversion ratio and yield. A second production cycle tested the reduction of aeration from 24 to 12 hours per day. Average yields of 18.10 t/ha were obtained for 18.58 g shrimp. Survivals over 80% were not significantly different among treatments. Production costs, gross income and net income per kilo were US$3.90, US$7.36 and US$3.45, respectively, for optimum systems. A 45% reduction in final weight occurs when aeration is reduced from 24 to 12 hours per day, without statistical differences in survival (av. 81.3%). This is reflected at the physiological level in the organism where individual oxygen consumption tests showed that more energy expenditure for routine metabolism is needed. It is concluded that the intensive photoheterotrophic culture is a good alternative for the shrimp industry, as it allows for high yields to be obtained in limited space, reduce the costs for feeding an energy for water pumping, maintaining stable culture conditions and minimizing potential pathogen entries to the system.

A SUSTAINABLE ZERO-WATER-EXCHANGE, FULL-WATER-REUTILIZATION SYSTEM FOR THE INTENSIVE CULTURE OF FRESHWATER REDCLAW CRAYFISH, *Cherax quadricarinatus*

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Freshwater crayfish demand has increased over 8 times in the last 15 years. They represent the 6th most valuable aquaculture group with production over 2 million tons and an unmet demand of more than 200,000 ton/year in worldwide markets, including Asia, Europe, the U.S. and Latin-America. The Redclaw crayfish adapts to diverse culinary options for a unique organoleptic experience, giving it a distinct competitive advantage. The increase in crayfish demand worldwide requires state-of-the-art technologies to fully incorporate new species for commercial production. The zero-water exchange, full water reutilization system for the intensive culture of redclaw crayfish, is a knowledge-based technology developed over the last 30 years. The photoheterotrophic culture is sustainable and bio secure, allowing for efficient feed use while self-cleaning organic wastes in the pond. It has been tested and validated at the commercial level, increasing yields significantly over traditional technologies.

A three phase (reproduction, nursery and grow out) 20-ha modular system produces over 200 ton of redclaw/year for continuous product delivery to the market. The modules can be replicated in locations where demand is identified. With an investment of US$4.6 million for infrastructure development (80 0.25-ha ponds), US$ 1 million for operation to first-harvest (month 18) and a sale price of US$15/kg, annual gross income is US$2.9 million, with production and commercialization costs of US$4.5/kg and a benefit/cost of 4.3. A 10-year income statement shows: IRR= 38%, NPV= US$5.2 million, ROI= 28 and payback time of 3.1 years.

HYPER INTENSIVE ZERO-WATER EXCHANGE CULTURE OF WHITE SHRIMP *L. vannamei*

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Shrimp production in Mexico has been increasing over the years but has been affected by diseases, cost increases, price variations, the use of outdated infrastructure and obsolete technologies. This has impeded consolidation and expansion. To counter this, sustainable, bio secure and economically efficient systems are needed. The photoheterotrophic culture is a hybrid system that mixes heterotrophic, chemotrophic and autotrophic microorganisms where a balance is struck in the ecosystem between nutrients, available dissolved oxygen and a capacity for particle transportation, through aeration. We have developed a three-phase hyper intensive technology for year round production of shrimp in the desert region of Northwest Mexico.

A three phase (conditioning, nursery, grow out) 6.6 ha modular system produces 6 cycles of 18 gram shrimp and 30 ton/ha with 60-day grow out. With an investment of US$5.8 million for infrastructure development, US$ 1.24 million for operation and a sale price of US$5.4/kg a benefit/cost of 1.4 is obtained. A 10-year income statement shows: IRR= 23%, NPV= US$3.5 million and payback time of 3.1 years.

The intensive photoheterotrophic culture is a good alternative for the shrimp industry, as it allows for high yields to be obtained.

BIOECONOMIC ANALYSIS OF THE ANNUAL INTENSIVE PRODUCTION OF PACIFIC WHITE SHRIMP (*Litopenaeus vannamei*) IN BAJA CALIFORNIA SUR, MEXICO

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In the future, aquaculture production will play a major role in meeting the nutritional demands of the population, contributing to food security worldwide. Over the last ten years, aquaculture has grown at a rate slightly over 5%/year, which is not enough to meet protein demands for 2050. This means that sustainable and economically viable intensified systems are necessary. A high-production scenario for aquaculture will decrease seafood prices significantly (around 25%) and increase their consumption. This also requires incorporation of new production areas, as Asia generates over 90% of total output, while there is enormous potential in other regios. In Northwest Mexico, shrimp farms have been using semi-intensive systems based on water exchange over the last 20 years. The industry faces severe challenges due to the presence of diseases (WSSV, EMS), increases in production costs and lack of environmental sustainability.

CIBNOR has been working with producers in Baja California Sur, a state located in the desert belt of Northwest Mexico where, production to 2013 averaged 20 tons/ha/cycle in 2 cycles/year, using 30 hp/ha of aeration and 25% water exchange. Nevertheless, disease problems (WSSV, AHPND) have limited production since. To improve biosecurity, and reduce water exchange costs, we evaluated changes in water quality, annual yields and production costs in two photoheterotrophic rearing cycles (spring-summer, summer-fall). We then established economic viability of the production strategy. We used six 1,000 m^2 PVC lined ponds at Biohelis®, the Innovation and Technology Park operated by CIBNOR, with 24 h aeration, that increased, depending on O_2 demand, from 20 to 40 hp/ha. There was no water exchange, but filtered seawater was incorporated in weekly pulses, for an equivalent of less than 2%/day, to maintain water level. A 35% crude protein diet was fed 2 times/day. Stocking rate was 120 Pl/m^2. Spring-summer yields of *L. vannamei* after 105 days reached 13,400 $\pm$ 233 kg/ha/cycle with a mean final weight of 13.33 $\pm$ 0.35 g. Yields for the summer-fall cycle reached 17,900 $\pm$ 530 kg/ha, in 90 days. Mean final shrimp weight was 18.60 $\pm$ 1.07 g. A stochastic model was fitted to the growth, mortality and food conversion curves, production costs were incorporated and sale prices determined income. Economic variables and financial indicators for each cycle are presented. Implications for the consolidation of intensive shrimp culture in semiarid areas are discussed.

BROODSTOCK CONDITIONING OF PACIFIC OYSTER IN RAS: EFFECT OF THE WATER QUALITY, CO_2-CARBONATE SYSTEM, AND *Polydora* sp.

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The Pacific oyster, *Magallana gigas* (formerly *Crassostrea gigas*), is one of the most cultured bivalves in Mexico. Nevertheless, the Pacific oyster production has a significant challenge: continuous seed production to meet the current demand. For seed production, the hatcheries frequently use broodstock from the farming zones exposed to natural conditions that cannot be controlled, like those related to the oysters' reproductive cycle or pollutants and parasites as *Polydora* sp. This has resulted in a constant variation of oysters' condition and unpredictable time required to produce mature organisms. For more accurate control of the factors involved in the reproductive cycle of oysters, the use of recirculating aquaculture systems (RAS) may be significant. Little information on the reproductive cycle on bivalves cultured in RAS has been registered, especially the consideration of other interactions as the CO_2-carbonate system (pH, alkalinity, CO_2, carbonates, bicarbonates, calcite, and aragonite) and the water quality (temperature, salinity, nitrogen compounds, and dissolved oxygen). Thus, this work aimed to characterize and evaluate the effect of the water quality and the CO_2-carbonate system and *Polydora* sp. on the reproductive cycle of *M. gigas* cultured in a RAS. The broodstock was conditioning in four different temperatures (18°C, 20°C, 22°C, and 24°C) for ten weeks. After the ten weeks of conditioning, the water quality factors and the CO_2-carbonate system were within the ranges reported as favorable for bivalves. However, no mature organism was observed, and high variability in the condition of the oysters was detected (Fig. 1.). The results suggest a negative effect of the *Polydora* sp. on the reproductive cycle of *M. gigas* cultured in a RAS (Fig. 2).

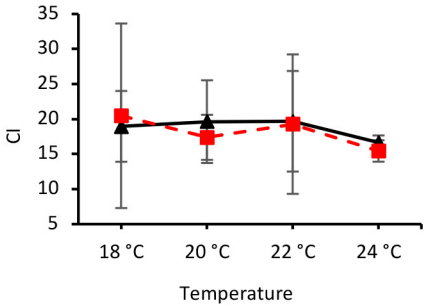

Fig. 1. Condition index of *M. gigas* during the broodstock conditioning in a RAS.

Fig. 2. Mud blister produced by *Polydora* sp. in the left valve of *M. gigas*

SUSTAINABLE COMMUNITY AQUACULTURE: OYSTER FARMING IN THE MEXICAN SOUTH PACIFIC.

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Climate Change is the most important threat to the survival of the human species and all species on planet earth. The stable climate that has served for the development of civilization is changing at a dizzying pace and puts all human activities at risk. The risk of extreme events, sea level rise, increase in terrestrial temperature and alteration to the balances of climate systems and ecosystems are already a reality throughout the planet (Bruno et al., 2018; Gattuso et al., 2018; Esperón-Rodríguez et al., 2019; Chapman et al., 2020).

However, the rapid growth of aquaculture had led to concerns regarding its ecological sustainability. Key issues include conversion of ecologically critical freshwater and coastal ecosystems to farms, pollution and eutrophica-tion of aquatic and marine ecosystems, heightened trans-mission of parasites and pathogens, overutilization of scarce fish meal and oil in aquafeeds, introductions of non-native species and interbreeding of escaped aquaculture stocks with locally adapted populations of wild relatives (Nie y Hallerman 2021).

This is because during the 50 years that aquaculture has been growing, it has done so by imitating more and more the intensive practices of industrialized livestock and agriculture. These intensive practices are due to the need of aquaculture companies to integrate into a market following a capitalist logic. This logic, which is basically to obtain the maximum (economic) benefit at the lowest cost, has brought significant profits for the owners of the companies, but has devastating environmental consequences, and does not allow local people to have a real benefit that reduces their poverty.

Using as a model the cultivation of native oysters in the marine portion of the state of Oaxaca, Mexico, we propose an aquaculture based on cooperative enterprises with a horizontal structure, where human development, resource utilization, environmental care and equitable benefits are at least at the same level of priority as economic gains. Since 2000, technical and biological feasibility studies have been carried out in the region, it is necessary to develop an education and training program for local coastal dwellers, constituted in Cooperative Societies, in order to achieve the change of vision necessary for aquaculture to become a strong drive to achieve several of the Sustainable Development Goals of the UN's 2030 agenda.

ENVIRONMENTAL SUSTAINABILITY OF MARICULTURE SYSTEMS WITH ALTERNATIVE FUELS – LCA CONSIDERATIONS

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Aquaculture is expanding sector on the global level resulting in higher concerns for its sustainability. Generally, sustainability can be defined as a process of meeting the needs of the present without compromising the ability of future generations to meet their needs, and it is considered to be constituted of three pillars: the economy, society and environment. This work considers environmental pillar of sustainability of mariculture systems, with a goal to investigate effect of alternative powering options in mariculture on its lifetime environmental footprint. In this sense the alternative mariculture system from Korican at al. (2022) has been considered. This work, as a result of INTEL-MARIC project, conducted at UNIZAG FSB considers set of alternative fuels (LNG, Methanol, Biodiesel) and associated emissions (CO_2, NOx, SO, and PM) of the mariculture system. The life-cycle assessment (LCA) indicates that significant environmental benefits are possible in case of alternative mariculture sysrem layouts and by use of alternative fuels like for instance methanol or biodiesel.

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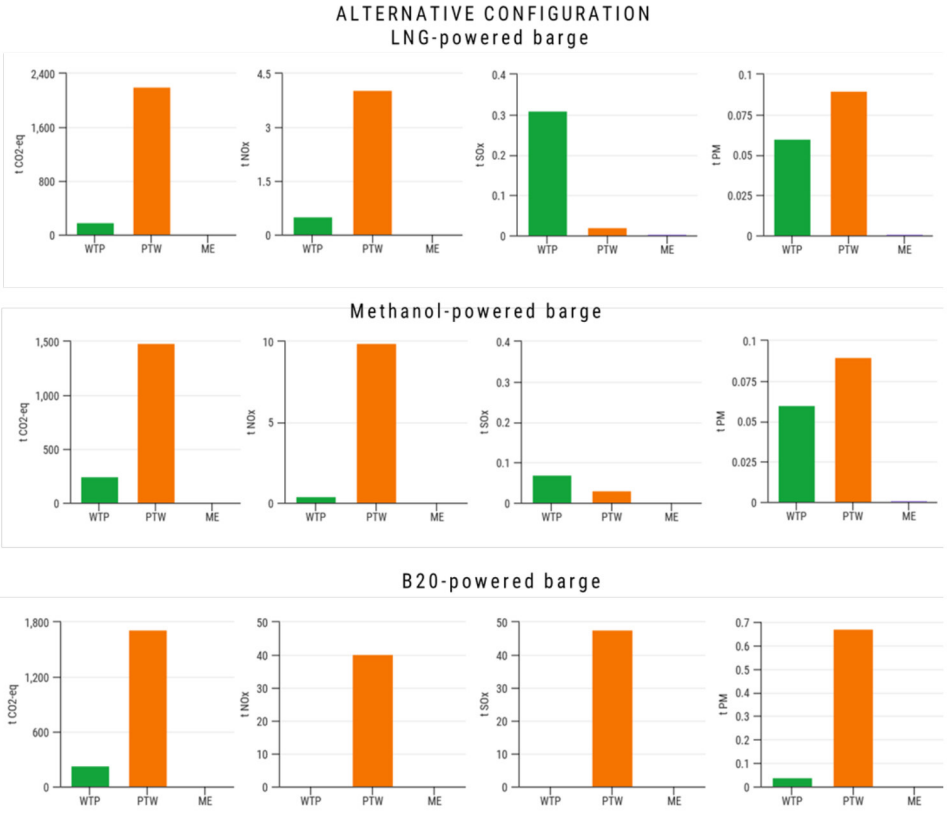

SUSTAINABLE DEVELOPMENT OF AQUACULTURE THAT SUPPORTS INDIGENOUS FOOD SECURITY, SOVEREIGNTY AND SUSTAINABILITY

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This paper considers the the Indigenous approaches to ensuring *Evechinus chloroticus* sea urchin, *Jasus edwardsii* spiny red crayfish or red rock lobster, *Haliotis iris* black foot abalone, *Perna canaliculus* green lipped mussel, *Anguilla dieffenbachii* long-finned eel, *Anguilla australis* short finned eel and various seaweeds and algae remain secure for Indigenous, coastal tribes of the East Coast of the North Island have secure food stocks that are culturally important for their customary, cultural and commerical wellbeing, now and into the future.

We examine the challenges for these tribes in their efforts to explore data-based decision-making to that supports self-determination in their marine environment, both life underwater and life on-land.

We also consider the role Indigenous Knowledge, and or Traditional Knowledge has to play in policy and legislation.

NGA KAITIAKI O PUREHUA – AN INDIGENOUS MARINE BIOSECURITY PROJECT

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This summer project came about to support Indigenous whanau (families of the tribe) as cutomary food gatherers to grow their knowledge of the biosecurity system in an effort to ensure the protection and sustainability of their kapata kai (food cupboards) in their rohe moana (marine environment).

Questions included:

- What is biosecurity?
- What are marine pests?
- What do we have in our kapata kai and rohe moana?
- What is biodiversity?
- What does kaitiakitanga mean and look like for whanau at place?
- What does it mean for others who want to work *with* us and alongside us?

Visual data collection was done before wananga and shared with whanau using their already present biodiversity index at their marae, Taharora.

COMMERCIAL SEAWEED CULTIVATION IN MEXICO

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Mexico´s seaweed industry started in the late 50´s, in the Pacific northeast coast of the Baja California peninsula, with the commercial harvesting of *Macrocystis pyrifera*, *Gelidium robustum* and *Chondracanthus canaliculatus*, as sources of phycocolloids. Presently, the use of seaweed in Mexico has diversify to uses such as seaweed extracts for agriculture, balance feed for marine cultivated organisms (abalone and fish) and even small amounts for human consumption. Other species have been harvested to produce phycocolloids (*Gracilaria-Gracilariopsis*, *C. squarrulosus*) and for new applications (*Eisenia*, *Laminaria*, *Pyropia*). Practically all the seaweed use for these products is harvested from natural beds which have decreased in a combination of higher demand and climate change. The efforts to develop the necessary studies for seaweed cultivation started in the mid-eighties.

In the early nineties and following the success of the commercial cultivation of *Gracilaria* in Chile, Mexico was part of a Latin-American initiative, supported by FAO, to promote the interest on seaweed cultivation in the region. By the mid-nineties, the first papers in methods for seaweed cultivation appeared. Many studies on the ecology and reproductive biology on the species of commercial interest were published simultaneously.

At the end of the 90's, a program for the cultivation of the Atlantic algae *Chondrus crispus* cultivation in ponds was tested for the first time and cultivation in the sea was developed using plastic mesh tubes. Both culture methods were tested with another red algae species such as *C. squarrulosus*, *G lemaneiformis*, *Pyropia perforata*. Following the basic methodologies proposed in the literature for the cultivation of kelps, in a program focused on the development of integrated aquaculture, experimental cultures studies for *Macrocystis pyrifera* and *E. arborea* were tested in several sites along the temperate Pacific coast of Baja California. These studies evidenced the possibility to develop the commercial cultivation of kelps in this region. However, it was not until 2016 that the cultivation of Ulva spp, for human consumption, at a commercial pilot level in ponds, justified the establishment of the first commercial cultivation farm Currently, about 120 wet tons per year are produced commercially. Production is continuous throughout the year. On the other hand, commercial trails for kelp cultivation in the ocean have initiated. Although the production of seaweeds in Mexico through aquaculture is still small, its possibility of growth, both onshore and offshore, are promising due to optimal oceanographic, geographic, and economic conditions.

MICROCYSTIN-LR INDUCING FERROPTOSIS IN INTESTINE OF COMMON CARP (*Cyprinus carpio*) AND INFLUENCES INTERACTIONS BETWEEN INTESTINAL MICROBIOTA AND ENVIRONMENTAL FACTORS

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Previous studies provide comprehensive evidence of the environmental hazards and intestinal toxicity of microcystin-LR (MC-LR) exposure. However, the interaction between the aquacultural environment and aquatic organisms, and underlying injury mechanisms in the intestine in response to MC-LR require further clarification. In the present study, juvenile common carp (*Cyprinus carpio*) were exposed to MC-LR (0 and 10 μg/L) for 14 days. The results suggest that organic anion-transporting polypeptides 3a1, 4a1, 2b1, and 1d1 mediate MC-LR entry into intestinal tissues. Lesion morphological features (vacuolization, deformation and dilation of the endoplasmic reticulum [ER], absence of mitochondrial cristae), up-regulated mRNA expressions of ER stress, iron accumulation, and down-regulated activity of antioxidant enzymes and glutathione (GSH) content were all typical characteristics of ferroptosis in intestinal tissue following MC-LR exposure. *Verrucomicrobia* and *Bdellovibrionota* were significantly associated with water quality (total nitrogen and total phosphorus) and GSH, and thus are suitable biomarkers of aquaculture and even natural environments in response to MC-LR exposure. These findings provide new perspectives of the ferroptosis mechanism of MC-LR-induced intestinal injury in the common carp, clarify the interactions between organisms and aquatic environments, and lay a foundation for biosafety assessment.

OPTIMIZATION OF ENCAPSULATION BIOLOGICALLY ACTIVE MOLECULES OBTAINED FROM MIXED *Halopteris scoparia* AND *Pistacia lentiscus* EXTRACTS

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Due to the rich source of biologically active molecules (BAM), extracts from algae and plants are an excellent potential for the production of functional foods with added value. Despite the fact that today there are many researches on different plant species, their application in the production of functional products is not sufficient. Studies have shown that the brown alga *Halopteris scoparia* is a source of various BAM's such as amino acids, polysaccharides, vitamins, sterols, minerals, dietary fibers, fatty acids and pigments (El-Din & Alagawany, 2019., Mišurcová et al., 2014), which have enormous functional and nutraceutical potential, while on the other hand, the analysis of mastic tree (*Pistacia lentiscus* L.) leaves, stems, fruits and roots revealed significantly higher concentration of the main groups of secondary metabolites (flavonoids, phenolic acids and tannins), indicating that the plant has great biological potential (Dragović et al., 2020). Due to the different composition of BAM, these two remarkable natural sources show a greater synergistic effect in the production of functional products by mixing the extracts. Given the particular sensitivity to degradation of BAM by temperature, pH, enzymes, and oxygen, encapsulation has proven to be an effective way to protect these compounds, especially in the production of functional foods. Therefore, the aim of this work was to optimize the spray-drying conditions of mixed extracts from the algae *H. scoparia* and *P. lentiscus* using two different carriers (maltodextrin DE 13-17 and gum arabica) on the phenolic and pigment content and physical properties of the obtained powder at spray-drying temperature of 120, 150 and 180 °C and wall-to-core ratio of 1:20.

The powders prepared with MD had lower product yield but better other physical properties and retention of BAC. The obtained results showed a strong synergistic effect of phenols from Mediterranean plants and algal pigments and their encapsulation by spray drying, which can be an excellent basis for further research and production of new value-added functional foods.

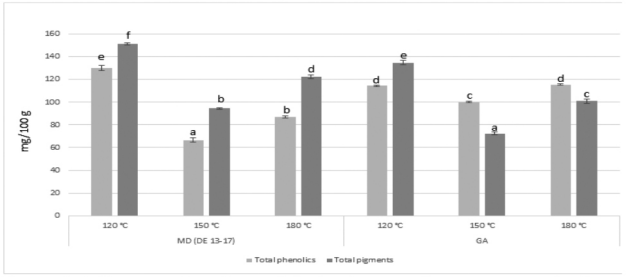

Figure 1. Influence of carrier and drying temperature on concentration of total pigments and phenolics

Table 1. Influence of carrier and drying temperature on physical properties obtained powders

Carrier	Temperature (°C)	Yield (%)	Solubility (%)	Moisture (%)	Bulk density (mg/mL)
MD (13-17 DE)	120	47.80±0.25	85.56±0.86	4.45±0.02	0.40±0.00
	150	50.12±0.20	84.04±0.20	4.03±0.01	0.37±0 01
	180	48.34±0.47	83.37±0.61	4.03±0.04	0.33±0.01
GA	120	54.88±0.92	75.83±0.67	6.24±0.02	0.31±0.01
	150	52.06±0.37	65.28±0.93	4.86±0.01	0.83±0.01
	170	53.22±0.45	79.46±0.59	3.63±0.01	0.39±0.01

Results are expressed as mean ± st.dev.