ACES JOINT MASTERS | AQUACULTURE | ENVIRONMENT | SOCIETY

Wednesday 5th August 2020

STUDENT CONFERENCE 2018-2020

Presentations from our FOURTH cohort of ACES students

BOOK OF ABSTRACTS

Erasmus Mundus

University of the Highlands and Islands
Oilthigh na Gàidhealtachd agus nan Eilean

UNIVERSITÉ DE NANTES

SAMS

Wednesday 5th August

10:00 – 10:25 Welcome and Introduction from Professor Liz Cottier-Cook, SAMS-UHI & Professor Michalis Pavlidis, University of Crete

10:25 – 11:40

Gerald Misol: Biological and genomic characterization of a novel jumbo bacteriophage, vB_pir03 with broad host lytic activity against *Vibrio harveyi*

Vibrio harveyi is a Gram-negative marine bacterium that causes major disease outbreaks and economic losses in aquaculture. Due to the crisis of antibiotic resistance, phage therapy is a potential alternative to antibiotics however, candidate bacteriophages require comprehensive characterizations for a safe and practical phage therapy. In this work, a lytic novel jumbo bacteriophage, vB_pir03 was isolated and characterized against *V. harveyi* type strain DSM19623. The bacteriophage was determined to have a similar morphology to the *Myoviridae* family and a broad host lytic activity against 31 antibiotic-resistant strains of *V. harveyi, V. alginolyticus, V. campbelli* and *V. owensii.* The adsorption rate of vB_pir03 was determined to be at 6 minutes and latent-phase at 40 minutes with a burst-size of 85 pfu/mL. In-vitro analysis also showed that vB_pir03 was able to lyse several host strains at multiplicity-of-infections (MOI) 0.1 to 10. The genome of vB_pir03 consists of 286,284 base pairs with a GC content of 43.6 % and 334 predicted ORFs. However, only 27 % of the predicted ORFs of vB_pir03 displayed homologies in existing databases while no virulence encoded-genes were detected. Whole-genome alignment and phylogenetic analysis with the large terminase subunit showed that vB_pir03 is a novel bacteriophage that showed the highest similarity with another jumbo phage, vB_BONAISHI infecting *Vibrio coralliilyticus*. Experimental phage therapy trial on brine shrimp, *Artemia sp.* infected with *V. harveyi* demonstrated that vB-pir03 was able to significantly reduce mortality 24 hours post infection at MOI 0.1 which suggests that vB_pir03 is a good candidate for phage therapy.

Joshua Superio: Parentage contribution of European sea bass (*Dicentrarchus labrax*) broodstocks, and correlation with fertilization success and larval survival

Increasing parentage contribution in aquaculture broodstocks is important in order to take full advantage of the available genotypes of selected fish. Two European sea bass (*Dicentrarchus labrax*) broodstocks (G1 and G3) were monitored during two consecutive reproductive seasons. In 2019, both broodstocks consisted of 14 males and 11C were allowed to spawn naturally. In 2020, G1 was induced to spawn using controlled-release implants loaded with gonadotropin-releasing hormone agonist (GnRHa), while G3 was again allowed to spawn naturally. Eggs were collected at every spawn for quality evaluation and parentage analysis. Relative fecundity in G1 increased in 2020, although the fertilization success decreased (P=0.002), perhaps in response to the observed lower sperm motility (P=0.001) of the males. There was no difference in the viability and hatching when G1 was induced to spawn in 2020, but larval survival improved significantly (P=0.041). Conversely, relative fecundity in G3 remained unchanged in two years, but egg viability decreased (P=0.029). Fourteen microsatellite loci were used to infer parentage of the collected egg batches based on broodstocks genotypes. In G1,

the parental assignment rate was 95.2% in 2019 distributed to 44 families and 77.5% in 2020 assigned to 37 families, respectively. In G3, the assignment rate was 94.8% in 2019 and 92.3% in 2020 with offspring belonging to 43 and 30 families, respectively. For parental contribution, 2-8 males and 1-4 females participated in each spawn in the naturally spawning broodstocks for both years, with 1-3 dominant males and females. On the other hand, in the GnRHa-induced group, 3-7 males and 3-6 females participated in each spwan with 3-4 dominant males and females. These results suggest that GnRHa induction increased both the number of broodstocks participating in each spawn and the relative fecundity and larval survival. Thus, GnRHa treatment can be employed in breeding selection programs to ensure wide genetic variation in progenies.

Raghi Rappai: Effects of lipid-rich diets and exercise in zebrafish

Aquaculture is a flourishing food industry swhich is crucial for meeting the food demands and nutritional balance of growing global population. Continuous improvements in technology and aquaculture science is necessary for developing an efficient system. The objective of the experiment is to study the effects of lipid-rich diets and exercise in zebrafish. The video samples of behavioural tests (novel tank and novel object recognition tests) of the study done by the master's research student for her dissertation were collected and analysed using tracking software ZebTrack. In this experiment, three tanks (35 zebrafish at 60 dph per tank)were randomly assigned to each of the four experimental groups (Control Diet – Control Exercise; Control Diet – Exercised; Algae Diet – Control Exercise; Algae Diet – Exercised). For three months, fish were fed their respective experimental diet (ZebraFeed® / 80%ZebraFeed+20% *Spirulina platensis*) twice daily and fish in the appropriate experimental group were either exercised or not, through increasing the flow rate within the tank to 3.6cm/S for a set period of time per day.15 fish per experimental group (5 fish per replicate) used for the behavioural tests and their videos recorded for 18-20 minutes. The results from the analysis of trial video samples showed that the lipid-rich diets and exercise has significant role in improving the boldness, memory, cognition and reducing anxiety and stress which is advantageous to easily adapt in the habituation and conditioning process of Aquaculture farming and to improve immunity, thus reducing production loss, which encourages aquaculture farmers to invest in expensive technology for a profitable efficient aquaculture system.

12:05 – 13:20

Emily Purvis: The effects of cryogenically preserved sperm on the fertilisation, embryonic development and hatching success of Lumpfish (*Cyclopterus lumpus)*

Lumpfish (*Cyclopterus lumpus*), are used as cleaner fish in the Atlantic salmon farming industry to remove parasitic sea lice. At present, wild lumpfish broodstock are used which puts strain on wild populations. By successfully cryopreserving lumpfish sperm, the number of wild males required will be reduced and it enables the long-term storage of sperm for use in breeding programmes and on-going research into their lice picking behaviour. The present study compared the use of fresh sperm and sperm which was cryogenically frozen for 24 hours to test whether it is a viable method of preservation. The fresh and frozen sperm from 5 males was used (in equal volumes) to fertilise eggs pooled from 5 females and the difference between fertilisation success, percentage of eggs which reached the eyed stage, and the hatching success was measured. A group of 100 hatched larvae were on grown for two weeks to test whether there was a difference weight between treatment groups. The results of the trial showed that fresh sperm produced a higher percentage of fertilised eggs (fresh **92.6** ± 0.8 %, frozen **77.9** ±1.8 %, mean ± SEM, n=5 sperm samples from different males), a higher percentage of eggs surviving to the eyed stage (fresh **93.9** ± 0.5 % and frozen **80.8** ± 1.4%) and had a

more successful hatch rate (fresh **72.3** ± 6.6 % and frozen **63.6** ± 5.0%,). There was a small difference in the weight of the fish between treatments (fresh **0.63g** ± 0.024, frozen **0.59g** ± 0.028). In conclusion, this study showed that the same volume of cryogenically preserved lumpfish sperm, produced fewer viable lumpfish larvae than fresh sperm. There is potential for further study into developing the application of cryogenically preserved Lumpfish sperm, including testing whether a larger volume of sperm would produce a more successful result.

Raneesha De Fonseka: Effects of Salinity on Growth and Welfare of Triploid Atlantic

The inferior saltwater performance of artificially induced triploid Atlantic salmon (*Salmo salar* L. 1758) has been identified as one of the greatest challenges in commercial-scale application. The purpose of this study was to determine the salinity optima of triploids compared to diploids in order to explore the possible relationships between growth and salinity tolerance. In freshwater, triploid and diploid underyearling smolts were assessed for smoltification, transferred to different salinities; 0, 11, 23 and 35 ppt (*n*=142 /ploidy /salinity) at 12°C and 24 hours continuous light for 12 weeks for grow-out. During this period, fish growth, plasma physiology, vertebral deformities and cataracts were investigated. According to gene expression results, triploids smoltified two weeks earlier than diploids although peak Na^+K^+-ATPase activity was similar between both ploidies. The weight advantage of triploids was significant throughout the experiment, but was effected by salinity treatment (final mean weight (g) ± SE), 322.9± 9.7, 361.7 ± 10.7, 425.9 ± 12.1, 415.2 ± 12.2 for triploids and 255.2 ± 7.4, 303.9 ± 9, 313.9 ± 9 and 342.4 ± 12 for diploids at 0, 11, 23, 35 ppt respectively. Therefore, triploid post-smolts grew best at 23 and 35 ppt whereas diploids performed best at 35 ppt only. Plasma physiology did not show any consistent ploidy effects at 23 ppt that would relate the growth advantage of triploids with the salinity. In addition, cataract score and vertebral deformities were not associated with salinity. Triploids had significantly more cataracts (mean cataract score ± SE, 2.78± 0.1) than diploids (1.93 ± 0.1) whereas vertebral deformities did not show any ploidy or salinity effect (less than 22% of ≤1 deformed vertebrae). Overall, triploid post-smolts can perform relatively better at lower salinities compared to diploids, although the reasons why require further clarification.

Sarah Jane Pugh: Recovery of head deformities in Gilthead seabream (*Sparus aurata L.*) during the on-growing period

One of the main loss of profitability of Mediterranean finfish aquaculture is the processing, selection and discarding of juveniles that deviate from the typical external morphology, prior to on-growing. Skeletal aberrations, in this case deformities in the head, are frequent abnormalities that arise during the hatchery rearing phase of Gilthead seabream (*Sparus aurata L.*) due to sensitivity to conditions. Although the degree of severity and occurrences are high, recent studies suggest that skeletal deformities (haemal lordosis) recover during the on-growing period in sea cages. Post-hatchery, a pool of 200 individuals from 1700 electronically tagged Gilthead seabream were examined for the presence of cranium and jaw deformities. The morphology of the phenotypes was investigated periodically until the end of the on-growing period, via geometric morphometric analysis and observational categorisation scoring; 0-Normal, 1- Uncertain, and 2-Abnormal. The prevalence of fish with head abnormalities decreased during the experimental period by ~15%. While 3 individuals completely recovered from an abnormal to normal phenotype, the highest recovery rate is seen in those individuals with initial light or uncertain deformities. Geometric morphometric analysis showed significant differences in head shape between the categorisations (0,1, and 2). Morphometric regression analysis is used as evidence to support natural phenotypic variations in growth throughout the on-growing period and to confirm that time of recovery varies between individuals. The results, along with past evidence of haemal lordosis recovery, provides useful information to alter the

processes of juvenile separation prior to on-growing and thus, presents the possibility to reduce significant production costs in the future.

14:20 – 15:35

Aaliya Malla: Investigating the Effects of Light on the Broodstock Conditioning of the European Flat Oyster *Ostrea edulis* (Linnaeus, 1758)

Controlling the reproductive cycle in hatcheries is crucial for the development and sustainability of the aquaculture industry. *Ostrea edulis* not only has been economically important since the 1800s, but its reefs provide important ecosystem services. Challenges occurring within broodstock facilities are a result of *O. edulis'* protandrous sequential hermaphroditic nature; lack of synchronicity and an uneven sex ratio hinder the intensive production required for developments in gametogenesis. In the industry, there is lack of available tools and guides for the assessment of gonadal development regarding *O. edulis*. Three objective were stated for this study: 1) compile a review on the practical quantitative and qualitative staging methods used for bivalves like *O.* edulis, 2) design an experimental set-up to investigate the effects of light on the reproductive cycle of *O.* edulis, and 3) conduct a preliminary experiment that investigated whether photoperiod and wavelength have an effect on reproduction. Oysters were exposed to five treatments: white light with natural photoperiod at 14°C (C-NP), white light with 24L:0D at 14°C (C-LL), red light with 18L:6D at 14°C (Red), blue light with 18L:6D at 14°C (Blue) and white light with 18L:6D at ambient water temperature (C-AMB). Our preliminary data showed that the experimental design is effective in providing the conditions for successful gametogenesis and spawning, out with the oysters natural spawning season. Nevertheless, no significant correlation was found between photoperiod and wavelength affecting the biometric parameters, suggesting that these may only 'fine-tune' gametogenesis. Therefore, further studies should investigate whether endogenous modulators are activated when exposed to variations in photoperiod and wavelength.

Rodrigo Mendes: Teleost fish mucosal microbiota: a microscopic solution to achieve future sustainability in aquaculture

Aquaculture has been expanding and intensifying in recent years to meet the feeding demand of the world population. However, the industry's development possesses an elevated risk for fish disease outbreaks and deterioration of the water quality. Antibiotics and vaccines represent the traditional approaches to mitigate diseases, however these strategies can develop antibiotic resistance genes, and their efficiency is only moderate. Moreover, feed absorption by the fish is low, which represents a significant source of nutrient input that leads to water eutrophication. In this sense, it is mandatory to discover efficient techniques to mitigate these problems and ensure future aquaculture sustainability. Microbiota is the assemblage of microorganisms present in the major mucosal surfaces of the gut, skin, and gill that establishes a symbiotic relationship with the fish being involved in host immunity and metabolism. Microbiota composition, diversity and consequently, physiological roles are affected by aquaculture practices but can also be directly manipulated via the administration of probiotics, prebiotics, synbiotics and immunostimulants given as dietary supplements to cultured teleosts. These products provide many benefits to the fish and farmers. They enhance the fish immune system through the regulation of immune-related genes, which increases disease resistance and survival rate, while decreasing the use of the traditional methods to mitigate pathogens. Moreover, the supplements allow the feed to be more efficiently absorbed in the gut by modifying the gut morphology and enhancing the activities of digestive enzymes, which minimises the amount of excreted metabolic waste, decreasing the risk of deteriorating water quality. Although several

research gaps need to be overtaken, the manipulation of microbiota using dietary supplements offer a promising and feasible solution to improve worldwide aquaculture sustainability in the future.

Grace Medina Madariaga: GIS-based site selection for offshore seaweed aquaculture in a Northern Atlantic case study (France)

Macroalgal aquaculture production has increased exponentially during the last decades, and the demand for diverse uses and industries increases every year. In Europe, seaweed aquaculture is not well established and there is a need for planning the development of this activity. The suitability for the offshore cultivation of *Laminaria digitata*, *Saccharina latissima and Palmaria palmata* in a study site located along the French Atlantic coast was determined with a spatial multi-criteria evaluation process based on a Geographical Information System (GIS) workflow. Variables such as nutrients, temperature, irradiance, light attenuation, current speed and wave height, were selected due to their influence on macroalgal growth, and adapted to each species requirements. All forcing data were obtained from satellite remote sensing (METEOSAT, MERIS and MODIS-AQUA) and model (ECOMARS 3D and WAVEWATCHER III) outputs. In addition, space competition with other socio-economical activities was included in the SMCE to consider potential conflicts,and integrated in a global suitability index. All the selected species showed high potential for offshore aquaculture, especially *L. digitata* and *S. latissima*. The seasonal variability of the suitability index was also taken into account, with March being the best month. A sensitivity analysis showed high sensitivity of the suitability index to changes in temperature, followed by nitrate, attenuation coefficient, and lastly phosphate. This study showed that large areas (up to 2671 km²) are available and suitable for offshore seaweed aquaculture within 25 nautical miles from the coast. Further validation based on experimental growth assessment is a perspective considered by the regional aquaculture industry.

15:35 – 16:00 EMJMD ACES+ – Student Award and Closing Remarks