



JOINT MASTERS | AQUACULTURE |  
ENVIRONMENT | SOCIETY

Tuesday 9th and Wednesday 10th July 2019

**STUDENT CONFERENCE 2017-2019**

*Presentations from our THIRD cohort of ACES students*

*At the Lecture Room,  
Natural History Museum,  
Heraklion, Crete*

**BOOK OF ABSTRACTS**



University of the  
Highlands and Islands  
Oilthigh na Gàidhe:  
agus nan Eilean



UNIVERSITÉ DE NANTES



## Tuesday 9<sup>th</sup> July

**09:00 – 09:20      Arrival**

**09:20 – 09:25      Welcome from Professor Jiannis Karakassis, Vice-Rector of Academic Affairs, University of Crete**

**09:25 – 09:35      Introduction to the Student Conference – Professor Liz Cottier-Cook, SAMS-UHI**

**09:35 – 10:50**

**Jennefe Cabarubias: Disease and Vaccination Challenge for *Streptococcus iniae* in Brown Marbled Grouper, *Epinephelus fuscoguttatus* (Forsskål, 1775)**

*Streptococcus iniae* is a hemolytic, Gram-positive bacterium that causes cellulitis in humans and meningitis and other lesions in various fish species. This bacterium causes significant disease problems in both freshwater and marine fish species including high-valued fish species such as brown-marbled grouper (*Epinephelus fuscoguttatus*). However, there has been few studies conducted on this economically important pathogen infecting this high-valued species and with no commercial vaccine yet in the market. In this study, two types of disease challenges were conducted for *Streptococcus iniae* in juvenile brown-marbled grouper which aims to obtain >70% mortality rate (>LD<sub>70</sub>). Experimental fish ( $n = 640$ , 85 g) were challenged through intraperitoneal (IP) injection and cohabitation. The IP injection of four (4) different *S. iniae* doses:  $1 \times 10^6$ ,  $1 \times 10^7$ ,  $1 \times 10^8$ ,  $1 \times 10^9$  resulted to a cumulative mortality of 78.8%, 88.8%, 93.8% and 90.0%, respectively within 14-day post-challenge. While the cohabitation with 6 or 8 shedders injected with  $10^7$  or  $10^8$  *S. iniae* concentrations:  $6(1 \times 10^7)$ ,  $6(1 \times 10^8)$ ,  $8(1 \times 10^7)$ ,  $8(1 \times 10^8)$  resulted to a mortality of 28.8%, 33.8%, 35.0%, and 36.3%, respectively within 13-day post-challenge. An exponential function from the results of IP injection disease challenge revealed that a  $3.4 \times 10^4$  *S. iniae* concentration should be given to attain LD<sub>70</sub>.

The laboratory efficacy of unadjuvanted, formalin-inactivated antigens were evaluated by IP injection and IP challenge to a new set of experimental brown-marbled grouper ( $n = 560$ , 40g). The five vaccine candidates with *S. iniae* concentrations of  $2 \times 10^5$ ,  $2 \times 10^6$ ,  $2 \times 10^7$ ,  $2 \times 10^8$ ,  $2 \times 10^9$  gave a relative percentage survival (RPS) values of: 73.9, 93.7, 72.4, 79.0, and 100%, respectively, when challenged with  $3.4 \times 10^4$  *S. iniae* concentration which produces a cumulative mortality of 20% in the control challenged group. The vaccine concentration of  $2 \times 10^9$  (FVG-UK 6 *S. iniae*  $10^9$ ) presented a significant result ( $p = 0.02$ ) of providing the highest protection against *S. iniae* in juvenile brown-marbled grouper.

**Ishrat Anka: Zebrafish as a model for aquaculture vaccines**

Disease incidence and reduced survival caused by *Aeromonas veronii* bv. *sobria* have been reported recently in farmed European seabass (*Dicentrarchus labrax*). Vaccination has been proposed as an

effective prevention to overcome this problem. However, research on vaccine efficacy in seabass under laboratory conditions has several limitations. Therefore, zebrafish (*Danio rerio*) was selected in the current study as a model organism to assess the efficacy of three developed autogenous vaccines of *A. veronii* - Vaccine 1 (NS), Vaccine 2 (PDB) and Vaccine 3 (5.28.6), comprising adjuvant (montanide 763). Intraperitoneal injection was performed at a dose of 10 µL/fish (N = 50) for the three vaccines and control-adjuvant groups. Vaccine efficacy was assessed in subsequent challenge tests (N = 12,10 µL/fish) using NS, PDB, 50 A and NS 2 bacterial strains (total count- 10<sup>7</sup> cfu mL<sup>-1</sup> each). Vaccinated fish groups were compared with non-vaccinated (control-adjuvant and control-naive) groups, in terms of survival and immunity. All three tested vaccines showed high efficacy against different bacterial strains. Vaccine 3 exhibited high survival in NS (100%), PDB (100%) and NS 2 (91.67%). However, the 50 A strain was classified as non-virulent for zebrafish. Cumulative mortality from challenged control-naïve groups was found to be higher than that of vaccines in NS (66.67%), PDB (41.67%) and NS 2 (58.33%). From challenge assessments, vaccine 3 showed the highest (100%) relative percentage survival (RPS) in NS, PDB and 50 A. Both vaccine 1 and vaccine 3 revealed high RPS (85.71%) in NS 2 challenge, whereas the lowest RPS (14.29%) was observed in the control-adjuvant group. Vaccine efficacy testing was found to be a successful screening tool, using zebrafish as a model organism for aquaculture. The present study-findings would be very useful to conduct further research on the impact of the tested vaccines and adjuvant in fish immunity.

**Mark Lisher: Determination of parental contribution to offspring of European flounder *Platichthys flesus* stock enhancement program in the Limfjord, Denmark.**

Stock enhancement programs using hatchery reared juveniles are implemented as fisheries and ecological tools towards addressing depleted stocks. In Denmark, stock enhancement of the European flounder into the Limfjord has resulted in 1.5 million juveniles being released since 1996 over a 23-year period. Culture technologies and production have been well studied within stock enhancement programs, with less attention focused on genetics and possible associated risks. Substantial genetic risk may occur from stock enhancement if contributing brood-stock sizes are too small, leading to loss of genetic diversity. This study aimed to determine the parentage contribution to offspring of European flounder in the Limfjord stock enhancement program. Six crosses from two batches containing different parent ratios (2♂:1♀ and 3♂:2♀) were analyzed for the 2019 reproductive season. Parentage analysis was undertaken using two multiplexes containing three and five microsatellites respectively (eight in total) for 24 brood-stock and 135 newly hatched larvae. Results are comparable to those of similar parental analysis studies in that the contribution of parents to offspring is skewed with a small number of contributing brood-stock accounting for majority of the offspring. Suggestions and recommendations to improve hatchery practices and potentially improve parental contribution of the stock enhancement program are also provided.

**11:20 – 13:00**

**Andrew Richardson: Exploring the Effects of a Commercial Insect Meal on *Dicentrarchus labrax* Juveniles**

The current growth of the aquaculture industry globally has created a race to find suitable, sustainable protein to support the growing demand. Insects have long been considered a viable option within the scientific literature, and following the publication of an EU Whitepaper, there has been a proliferation

of companies producing various insect-based products. Black Soldier Fly (*Hermetia illucens*) has been shown to have a similar amino acid profile to fish meal and is already utilised by some areas of the northern European aquaculture industry. In this study we investigate the effects of commercially relevant inclusion rates (10% & 5%) of a commercial Black Soldier Fly larvae meal on European Sea Bass (*Dicentrarchus labrax*) juveniles. We employ molecular, physiological and zootechnological indicators to assess more fully the effects and resultant interactions induced by insect protein inclusion. We show that there is no effect on any commonly used performance index we investigated, supporting proponents of the industry. However, there is a cause for further investigating potential effects at the haematological level. Inclusion at first sampling was shown to effect haemoglobin, glucose and cortisol levels ( $p < 0.05$ ). These indicate that there is a possible early effect of hyperglycemia. This has dissipated by the final sampling, with no significance found across the plasma metabolites. Furthermore, we show that there is no effect on the relative expression of hepatic Insulin-like Growth Factor -1 (IGF-1) across diets ( $p > 0.5$ ), whilst supporting the previous literature that reports a positive correlation between weight and IGF-1 expression. This work supports the usage of BSFM as part of a varied solution to solving the sustainability of the Mediterranean industry.

#### **Guilherme Sansao: Water Temperature as an Early Life Programming Factor for Gilthead Seabream (*Sparus aurata*) Stress Response**

In the present study the effect of early life water temperature on the stress response at subsequent stages of development was evaluated in gilthead sea bream (*Sparus aurata*). To this end, fish were reared at three different temperatures (17°C, 20°C and 23°C) from epiboly to first feeding (EL-17, EL-20, and EL-23, respectively), and then at 20°C until the end of the larval rearing period. All groups were challenged with a transportation stress at the stage of full formation of all fins. Samples from the three groups were collected at the stages of embryos, first feeding, flexion, full formation of all fins (before and after stress), for whole-body cortisol levels and gene expression (Hypothalamus-pituitary-interrenal and somatotrophic axes) analysis. Here, we also developed and evaluated for the first time a new method for whole body homogenization and further cortisol extraction of pre-larvae and larvae at the individual level. Both hormonal and molecular parameters revealed significant differences ( $p < 0.05$ ) between the different groups based on early life history. A distinctively U-shaped pattern of basal cortisol (supported by the molecular data) was observed at first feeding, flexion and all fins stages, with lower cortisol levels at EL-20 group compared to the EL-17 and EL-23 groups. Additionally, while at the EL-20 group a peak was observed at 30 min post-stress, at EL-17 the peak was prolonged (30 min and 1 h) and at EL-23, values remained constantly high until 4 h post-stress. Thus, providing supporting evidence that early life temperature rearing, from epiboly to first feeding, has imprinting effects on later life stages. Furthermore, it can be inferred that an early life temperature of 20°C results in optimal physiological outcomes when compared to 17°C and 23°C, regarding *S. aurata* welfare.

#### **Mary Arnaldo: Purification and characterization of antioxidant and antiproliferative endogenous peptides from red seaweeds *Chondrus crispus* and *Gracilaria gracilis***

Rhodophytes, among other seaweeds, contain high protein levels comparable to conventional protein-rich terrestrial foods. Recently, the role of seaweed proteins and peptides expanded beyond nutrition, along with the search for compounds that provide additional physiological functions. Dietary peptides received huge interest and increasing applications in the pharmaceutical and nutraceutical industry due to their expressed bioactivities (e.g., antioxidant, antihypertensive, antiproliferative). Hence, red seaweeds are inestimable reservoirs of peptides that could be excellent raw materials for

potentially novel bioactive compounds. Although the bioactivities of seaweed peptides were reported, most studies focus on protein-derived or exogenous peptides. The structure and biological activities of endogenous peptides, on the other hand, remain to be scarce and/or poorly undocumented. In this study, hydrosoluble proteins and endogenous peptides were extracted from *Chondrus crispus* and *Gracilaria gracilis*, two rhodophytes with markedly higher protein content. Extraction yield was determined, and peptides were ultrafiltered for bioactivity tests. The crude peptidome was screened for antioxidant activity using the DPPH assay, and for antiproliferative effect on human osteosarcoma cell line (KHOS) using the crystal violet staining assay. Results indicated higher hydrosoluble protein yield from *G. gracilis* ( $7.76 \text{ mg}\cdot\text{g}^{-1}$  dry weight) than *C. crispus* ( $4.39 \text{ mg}\cdot\text{g}^{-1}$  dry weight). Endogenous peptide concentration was also higher for *G. gracilis* ( $3.11\pm 0.15 \text{ mg}\cdot\text{g}^{-1}$  dry weight) than *C. crispus* ( $3.02\pm 0.87 \text{ mg}\cdot\text{g}^{-1}$  dry weight). Antioxidant activity existed for both *G. gracilis* and *C. crispus* endogenous peptides, but *G. gracilis* demonstrated higher scavenging activity ( $15.98\pm 0.36\%$ ) than *C. crispus* ( $8.45\pm 0.42\%$ ) at  $500 \mu\text{g}\cdot\text{mL}^{-1}$ . Both endogenous peptides crude extracts exhibited dose-dependent inhibitory effect against KHOS, with strongest inhibition ( $79.35\pm 1.23\%$ ) observed at  $200 \mu\text{g}\cdot\text{mL}^{-1}$  of *C. crispus*. The peptide extracts are further fractionated by HPLC to purify and identify target peptides. This study validates *C. crispus* and *G. gracilis* endogenous peptides as antioxidants, and as antiproliferative agents against KHOS human osteosarcoma cell line.

#### **Sofia Tretiak: Optimization of the antioxidant activity of *Gracilaria vermiculophylla* for use in an edible film for food packaging**

Single-use food packaging contributes towards almost one-third of the global plastic waste. In an attempt to minimize environmental impacts, sustainable alternatives need to be developed. Seaweed is a renewable resource that can be utilized for packaging development for plastic replacement. *Gracilaria* is one of the most widely-cultivated algae, used in food and pharmaceutical industries as a valuable source of agar. Studies have succeeded in developing biopolymer plastics from *Gracilaria* containing high levels of antioxidants. Antioxidant properties are essential in biopackaging as they are beneficial to packaged food by limiting lipid oxidation, which is one of the main causes of the food spoilage. This study focuses on the cultivation of *Gracilaria vermiculophylla* and optimizing its antioxidant properties for further use as an edible film for food packaging. Antioxidant compounds are synthesized in seaweeds as a response to oxidative stress that often occurs due to harsh environmental fluctuations. Therefore, antioxidant activity of the seaweed can be modified through exposure to different stress conditions. Three consecutive experiments were conducted in this study to better understanding on how different levels of light irradiance, differences in light : dark cycles, exposure to UVA radiation, hypersalinity, desiccation and the duration of exposure can affect the antioxidant profile of *Gracilaria vermiculophylla*. High light intensity increased the antioxidant levels of *G. vermiculophylla*, reaching a maximum of 33.45%. Moreover, the combination of the high light irradiances and UVA enhanced the antioxidant activity and had a positive survival effect on *G. vermiculophylla*. The study showed that increased antioxidant activity could be achieved after 3 days of exposure to different cultivating conditions. These findings may be beneficial to industrial scale seaweed cultivation, where favorable antioxidant levels could be reached shortly prior to harvesting.

**14:25 – 15:15**

**Caroline Schkeeper: Aquaculture Higher Education and Training in Cambodia: A Q-methodology Study and Curriculum Comparison**

Although Cambodia has historically relied on its inland freshwater capture fisheries, it is predicted that these have plateaued, highlighting the importance of developing sustainable aquaculture in this region. In recent years, Cambodia has enjoyed growth in the aquaculture sector, but it has not yet developed into a commercial industry like its powerhouse neighbors. The discrepancy in the rate of development is likely due to several factors, but it could possibly be explained by the undeveloped nature of aquaculture higher education and training in Cambodia. While the issues facing the education sector in general are well-documented, the perceptions and opinions of relevant aquaculture stakeholders on issues facing the development of aquaculture education in Cambodia are not well-known. This study used q-methodology to assess the perceptions and opinions of nineteen relevant stakeholders on the issues and areas of improvement of aquaculture education and training in terms of industry development. The analysis revealed four well-defined factors and certain areas of consensus which identified both major barriers to educational and industry development and possible ways forward. The major barriers identified were: inadequate technology, low student motivation, inefficient development programs, low quality of educators, and lack of university programs. In conjunction with the q-methodology analysis, a curriculum analysis was performed in order to identify possible gaps or strengths in Cambodian aquaculture bachelor programs compared to neighboring countries with stronger aquaculture education. This analysis revealed that aquaculture curriculum varies from country to country and there is no level of standardization across the region. However, across the four factors there was general agreement to design aquaculture curriculum in Cambodia according to stakeholder and industry needs. Overall, it is recommended that Cambodian universities and relevant stakeholders strengthen international collaboration in order to advance both aquaculture education and the industry.

**Ron Tardiff: Integrated Mariculture in China - An iterative assessment**

Chinese integrated mariculture (InMar) encompasses a broad array of systems, species, and methodologies. While China has practiced integrated aquaculture in freshwater for more than 1000 years, Chinese InMar practices developed rapidly beginning in the 1980s. The rest of the world has likewise grown interested in the space and resource efficiency offered by these systems. However, little is known about the composition, drivers, and extent of InMar practices throughout China. Through an analysis of Chinese and English literature, we assess the state of published knowledge on Chinese InMar from the last two decades. A vast amount is known about InMar practices in Sanggou Bay, Shandong Province (44% of the literature). The remaining InMar cases (n=31) were analyzed separately. The literature contains a large body of evidence supporting the nutrient, carbon, and space efficiencies of InMar, but limited evidence supporting the proposed socio-economic benefits. Further, we conducted informal discussions with nearly three-dozen stakeholders ranging from farm workers to high-level administrators in three provinces. Stakeholders indicated high uncertainty regarding the new wave of marine functional zoning (MFZ), highly flexible species choice, acute sensitivity to market price, problems related to rising labor costs, and good relationships with researchers. The third element of this work aimed to assemble data on the extent and scale of InMar practices. We collected data on >3.2 Million tons of production in 5 areas, with an average trophic level of 1.6. Thus, Chinese mariculture remains primarily extractive. These investigations compose the first attempt to

characterize the full suite of information, drivers, and extent of InMar practices in China. The continued development of InMar practices will require focused attention through MFZ, emphasizing carrying capacity limits based on trophic level. Meanwhile, market development for organic or “eco-friendly” fisheries products produced by InMar practices is also recommended.

## **15:45 – 16:10**

### **Fiona Mackecknie: Comparison of growth performances and lipid production of marine benthic diatoms using two type of photobioreactor: airlift and biofilm**

Currently, world algal biomass production remains low and within specialized areas dealing with high value products (aquaculture, cosmetics, pharmaceuticals, nutraceuticals) due to issues with growth and high costs associated with growth, harvest, and contamination. Specific types such as benthic diatoms can be difficult to cultivate under conventional airlift methods, as it does not reflect their natural habitat conditions. Recent developments in attached microalgal cultures have proved successful in improving the growth, by reducing light attenuation and increasing exposure to CO<sub>2</sub>, as well as reducing energy needed to separate water from biomass. Four species of microalgae from the Nantes Culture Collection (*Nitzschia alexandrina* NCC 33, *Nitzschia laevis* NCC 39, *Amphora sp. 2* NCC 169, and *Opephora sp. 1* NCC 366) were grown to determine if there were differences in growth rates between stirred and unstirred cultures. A porous substrate bioreactor (PSBR) was developed to compare growth between airlifts and attached bioreactors. One species of algae, *Nitzschia alexandrina* NCC 33, was chosen to compare growth between airlift and PSBR, by standardizing the nutrients per cell ratio and the lighted surface area available. Three trials were run, and growth were measured via pulse amplitude modulation (PAM, Fv/Fm), normalized difference vegetation index (NDVI), cell counts, and gravimetrically. The crude lipids from the final biomass was then extracted from each trial to determine differences crude lipids between trials. There was no difference found between the specific growth rates on sampling days (0, 3, 6, 10, 14, 18) of the stirred and unstirred cultures or the final cell counts (t-test of unequal variances). The Fv/Fm, NDVI, gravimetric and cell counts were used to determine growth rates and compared across trials.

## Wednesday 10<sup>th</sup> July

**09:10 – 09:35      Arrival and Set Up**

**09:35 – 10:50**

**Stephanie Acevedo Carvajal: Evaluation of astaxanthin dietary levels in red seabream for the optimization of skin colour**

The porgies (*Pagrus spp.*) enjoy a high wild-capture demand with a significant global market price. Under aquaculture conditions, however, fish presents hypermelanosis and loss of the natural skin hue and chroma impacting negatively on consumer's acceptance and market appeal. Currently, the inclusion of astaxanthin, mainly esters, in feed fish is used to overcome this bottleneck, leading to an increment in production cost. Therefore, the objective of this study is to determine the optimum dose and administration time of astaxanthin from natural resources on red seabream. Juveniles of red seabream (*Pagrus major*) ( $32 \pm 3.0$  g; mean  $\pm$  SD) were fed for 60 days on four diets supplemented with different natural astaxanthin concentrations (25, 60, 80, 100 mg Kg<sup>-1</sup>) and a control with no astaxanthin addition by triplicates. Zootechnological, physiological parameters and color attributes were measured on fed fish at 30 and 60 days. Growth performance, antioxidant capacity, cortisol and biochemical indices like glucose and cholesterol did not differ significantly among the experimental diets. Carotenoid-diet did not impact significantly the melanophores-covered area and skin melanin content, however, lightness in the dorsal area had a significance increment since day 30 in all astaxanthin concentrations. Furthermore, the concentrations of 60, 80 and 100 mg kg<sup>-1</sup>, improved significantly the presence of erytrophores/xanthophores and the values for hue and chroma in fish skin since day 30, compared with the 25 mg kg<sup>-1</sup> diet and controls. In conclusion, the supplementation of astaxanthin in fish feed for red seabream is already optimal in a concentration of 60 mg kg<sup>-1</sup> for 60 days, at a water temperature of  $20.8 \pm 1.3$  °C and is enough to reduce the darkness and achieve a close to natural aspect redness in porgies skin.

**Risha Alleyne: Development of super intensive, biofloc-dominated systems for indoor production of the Pacific White shrimp, *Litopenaeus vannamei*.**

Pacific White shrimp (*Litopenaeus vannamei*) is the most cultivated crustacean species (53% in 2016), with Asia and Latin America dominating production. In France, the cultivation of shrimp is low (40 tonnes of *Litopenaeus japonicus*). Consumer preferences have created opportunities to culture chemical-free, good quality, fresh shrimp in Europe. To exploit such opportunities, research was conducted at LISAQUA, France, to cultivate post larvae (PL) Pacific White shrimp with favourable zootechnical performance in an indoor, super-intensive, recirculating, biofloc system. Several feeding trials to determine the preferential feed quantity and ideal feed brand were performed over a four-month period (February – May 2019). Some feed quantities and brands exhibited lower feed conversion ratios (FCR) and better growth rates. The ideal stocking density of PL100 shrimp was evaluated over four weeks in three 210 L experimental units with stocking densities of 3, 5 and 7kg/m<sup>3</sup>. The density of 3kg/m<sup>3</sup> achieved the highest survival rate. Survival rates, FCR and growth performance were calculated for each experiment. Water quality parameters were analysed for all experiments. Investigations were done to pinpoint sustainable benchmark feed and their suppliers to optimise shrimp growth. Furthermore, resistant pathogens are plaguing the industry due to mismanaged

application of antibiotics. Reviews were conducted to identify probiotics products and beneficial microbes to boost shrimp resistance, quality and feeding efficiency. Melanosis is a major problem in the post-mortem stages of shellfish, adversely affecting the aesthetic appeal to consumers thereby reducing shrimp market value. For decades, melanosis has been controlled by the use sulphite-based products which cause many negative health and environmental effects as allergens and carcinogens. Hence, there is a need for sulphite-free alternatives to address consumer demands. A bibliographic survey to source sulphite-free products was conducted. Three potential alternatives were recommended.

**Diego Ruiz Orta: Effect of different post-fragmentation rearing methods and light spectra on the survival, self-attachment and photobiology of the soft corals *Sarcophyton sp.* and *Sinularia sp.***

Corals are one of the main organisms traded within the aquarium industry, as well as a promising source for bioprospection for marine natural products (MNP). Hence wild populations encounter a series of threats such as habitat degradation and overharvesting. Aquaculture has the potential to help mitigate those impacts. The present study aimed to describe the response of the soft coral species *Sinularia sp.* and *Sarcophyton sp.*, subjected to different *ex situ* experimental conditions. Two responses were tested: 1) the attachment of coral fragments to an artificial substrate, comparing different post-fragmentation methods (cyanoacrylate glue, rubber band and impaling), and efficiency of different commercial cyanoacrylate-base glues; and 2) the photobiological response of corals reared with two different techniques (attached to a substrate vs suspended in the water column). For both tests the coral fragments were subjected to three different light spectra: mixed light, blue light, and white light; delivered by 80 W T5 fluorescence lamps. The Photosynthetically Active Radiation (PAR) for the three light spectra was  $175 \mu\text{mol quanta m}^{-2} \text{s}^{-1}$ . After 44 days, fragments of both species that were glued to a substrate had higher survival (86%) and self-attachment success (69%) than the other methods. A  $\chi^2$  association test showed that neither the attachment method, type of glue nor the light spectra affects the self-attachment. Pulse Amplitude Modulation fluorometry (PAM) was used to measure the maximum quantum yield of PSII. The mean values of  $F_v/F_m$  obtained from the fragments suspended in the water column presented significantly higher values than the attached to a substrate for the three light spectra. The effect of the light on the  $F_v/F_m$  was only present on the fragments attached to a substrate. The hanging method provides a promising new approach on the culture of coral species, with potential for its use in the bioprospection of MNP.

**11:20 – 12:10**

**Janel McNab: Microphytobenthos biofilms on sea urchin *Paracentrotus lividus* recruitment: the effects on juvenile metamorphosis, growth and survival.**

Recruitment of competent larvae is crucial in sea urchin seed production, with the growth and survival relying heavily on the settlement success and on the grazing substrate. The objective of this study was to evaluate local microphytobenthos species, *Nitzschia laevis*, on *Paracentrotus lividus* metamorphosis induction, growth and post-larval survival, serving as an initial grazing substrate for a cost-effective production cycle. In the metamorphosis induction assay, 750 competent larvae were inoculated into five replicates of 80 mm diameter dishes conditioned with four different treatments: (1) *N. laevis*, (2) *N. laevis* and antibiotics, (3) oyster shells particles and (4) natural biofilm obtained from an oyster culture site. Larvae on these treatments were compared to larvae on 100 ml filtered sea water (negative control) for an extended period of eleven days. Survival and growth of juveniles

(test diameter) were monitored for 44 days post-metamorphosis on the treatments of *N. laevis*, *N. laevis* + antibiotics and natural biofilm, with 0.50 g of *Palmaria palmata* thalli added to each replicate as first food for larvae. Maximum metamorphosis values were obtained from *N. laevis* biofilm, with 67% metamorphosis rate, and a significant difference being detected only between *N. laevis* biofilm and the control. Although this treatment had the highest metamorphosis rate, the natural biofilm treatment was the first to had recordings of metamorphosis at 72h post-inoculation with a metamorphosis rate of 3% whiles, *N. laevis* biofilm recorded metamorphosis at 96h post-inoculation with a metamorphosis rate of only 1%. Regarding survival and test diameter, the higher values were observed for *Nitzschia laevis* + antibiotic (93% and 354.08  $\mu\text{m}$  respectively). In general, the results showed that the natural biofilm provides an adequate environment for recruitment while the *N. laevis* biofilms with or without antibiotics provided excellent environment for the recruitment, growth and survival of the *P. lividus* juveniles.

#### **Leovigildo Rey Alaban: Investigation of fungal halogenated metabolism: comparison between terrestrial and marine *Penicillium canescens* strains**

The marine environment is a vast frontier for natural product discovery. Marine derived organisms are known to produce compounds which are important in the medical field – such as the halogenated secondary metabolites. The presence of a vanadium – dependent haloperoxidase (v HPO) gene in a marine strain of *Penicillium canescens* (MMS194), which produces this group of metabolites, has recently been identified. This study proposes to investigate the role of this enzyme by comparing the metabolome of this marine strain with a terrestrial strain (MMS1620) of the same species, which does not possess this gene using untargeted metabolomics approach. Mono and co-cultures in solid media – Wickerham (WKH) or Kohlmeyer (KMS) – which were either enriched with halogen (SX) or not (DW) were carried out. Microextraction was performed using ethyl acetate and extracts were suspended in methanol (HPLC grade) prior to analysis by HPLC – ESI – HRMS (High Performance Liquid Chromatography – Electron Spray Ionization – High Resolution Mass Spectrometry). MZmine 2<sup>®</sup> was used to generate the data matrix of mz-RT (mass/charge - Retention Time) features which was analysed with MetaboAnalyst<sup>®</sup>. Chemometrics analyses indicated that the type of culture media and enrichment can substantially alter *P. canescens* metabolic profile. PCA (Principal Component Analysis) and PLS DA (Partial Least Square Discriminant Analysis) showed distinct clusters particularly for the terrestrial strain (MMS 1620). Further, co-culture of both strains did not induce production of any unique feature. However, a clear distinction between the marine and the terrestrial strains was observed, leading to hypotheses on the role of the v HPO on metabolite production. A possible novel brominated compound was isolated (using normal flash chromatography) and is currently being purified using semi preparative reverse phase HPLC. These results confirm the value of marine fungi for the discovery of new promising metabolites, as well as for biotechnological applications.

**12:10 – 13:00 Meeting with ACES External Auditor, Prof Gavin Burnell – All students**

**14:00 – 14:25 EMJMD ACES – Student Social Media Award and Closing Remarks**

**20:30 til late Farewell Party**