closely related to tetrapods than to bony fishes, and therefore could be considered as the living sister-group of tetrapods.

* Corresponding author.
E-mail address: fbuono@unitus.it (F. Buonocore)

O-361.

Eel immune response to *Vibrio vulnificus* infection. Host-pathogen relationship

A. Callol [1,2,], D. Pajuelo [1], S. MacKenzie [2,3], C. Amaro [1].*

[1] Department of Microbiology and Ecology, Faculty of Biology, University of Valencia, Burjassot, Valencia, Spain;
[2] Institute of Biotechnology and Biomedicine, University Autonomous of Barcelona, Bellaterra, Barcelona, Spain;
[3] Institute of Aquaculture, School of Naural Sciences, University of Stirling, Stirling, Scotland, UK

Abstract
The European eel (*Anguilla anguilla*), has experienced a dangerous decline in recruitment, yield and stock over the last 30 years and this decline is likely to continue into the future. Several major threats are responsible for this situation, including overfishing of glass eels for consumption, new infections by introduced pathogens, dams and blocking of migration routes… The lack of knowledge concerning the biology of this species represents a handicap to the conservation and recovery of the population. Nevertheless, the eel farming industry is increasing, therefore it could be considered one of the major risks for wild eel stock perpetuation, which is threatened principally by the lack of knowledge of the complete reproductive cycle of the European eel and by emerging bacterial, viral and parasite diseases.

Vibrio vulnificus is the aetiological agent of warm-water vibriosis, a disease that constitutes the main threat to eels under culture conditions, provoking important outbreaks and can be an opportunistic pathogen for humans. *V. vulnificus* uses a novel characterized virulence and survival system named MARTX (multifunctional repeat in toxin) that it is supposed to trigger a cytokine storm when it is produced in blood during the infection. It is also involved in the colonization of the gills, to invade internal organs and causes death by septicemia.
We sequenced an eel immune-enriched transcriptome with Roche 454, and the data has been used to create new molecular tools for further research. We have also designed a custom eel-specific microarray (4x44K, Agilent).
Our research has been focused on eel-vibrio interaction, how the host responds to the challenge and the importance of MARTX system of the bacteria for the colonization. Different challenges were performed using wild type strain (CECT4999) and Rtx double mutant strain (CT285) to evaluate mucosal immunity in the gills, the principal portal of entry. We also studied the early immune response of circulating leukocytes and the role of the erythrocytes, as one of the target cells of the lytic activity of this bacterium. Results obtained by RT-qPCR and ISH on gills demonstrate the immunocompetent function of this tissue, based on the expression and localization of immune-relates genes such as, TLRs, cytokines, chemokines and signalling molecules. Microarray studies describe key genes expressed after vibrio infection analyzed across 12h post-challenge on gills and blood (leukocytes and erythrocytes separately), specific host response against vibrio Rtx toxin, as well as, a possible function of erythrocytes in the immune response.

* Corresponding author.
E-mail address: agnes.callol@uab.cat (A. Callol)

O-241.

Interactions between hemolymph components of *Mytilus galloprovincialis* and different *Vibrio* species

L. Canesi, T. Balbi, R. Fabbri, A. Smerilli, E. Pezzati, C. Grande, M. Stauder, L. Vezzulli, C. Pruzzo.*

DISTAV, Dipartimento di Scienze della Terra, dell'Ambiente e della Vita, Università di Genova, Italy

Abstract
Marine bivalves are filter-feeding invertebrates that can accumulate large numbers of bacteria, in particular *Vibrio* species particularly abundant in coastal waters. Persistence of different vibrios in bivalve tissues largely depends on their sensitivity to the bactericidal activity of the hemolymph, resulting from complex interactions between bacteria, circulating hemocytes, and soluble hemolymph components. Host-pathogen interactions have been increasingly investigated in different bivalve species, with the aim of understanding the pathogenesis of diseases in species susceptible to infection by certain *Vibrio* spp. and strains. *V. splendidus* LPG32 strains have been associated with the 'summer mortalities' syndrome of juvenile oysters. On the other hand, the mussel *Mytilus* is considered to be particularly resistant to *Vibrio* infection. In this work, in order to explore the susceptibility of mussels to different *Vibrio* species, the interactions between hemolymph components of *Mytilus galloprovincialis* and *V. aestuarianus* and *V. splendidus* were investigated. *In vitro*, the bactericidal activity of whole hemolymph towards different vibrios, the capacity of bacteria to adhere to hemocyte monolayers both in the presence and in the absence of different sugars, as well as their effects on hemocyte lysosomal membrane stability (LMS) were evaluated. In *in vivo* experiments, mussels were injected with live *V. aestuarianus* and *V. splendidus* and bactericidal activity of whole hemolymph, bacterial concentration in mussel tissues, hemocyte LMS and serum lysozyme activity were evaluated at 6, 24 and 96 h post-injection. Moreover, LMS was evaluated as a biomarker of general stress in the digestive gland. Overall, the results indicate distinct surface interactions between mussel hemolymph components and the vibrio strains tested: these interactions lead to killing of *V. aestuarianus*, whereas *V. splendidus* was actually able to grow within the hemolymph. Moreover, *V. splendidus* induced severe decreases in LMS in both circulating hemocytes and digestive gland. The results demonstrate that, although *V. splendidus* LPG32 is considered not pathogenic to *Mytilus*, this vibrio strain is able to grow within the mussel host and to cause disruption of the lysosomal function at the cellular and tissue level, thus leading to significant stressful conditions and alteration of mussel health.
This work was supported by the EU project BIVALIFE (FP7-KBBE-2010-4).

* Corresponding author.
E-mail address: Laura.Canesi@unige.it (L. Canesi)

O-185.

Studies of antimicrobial peptides in rainbow trout

E. Casadei [1,], J. Zou [1], S. Wadsworth [2], J.L. González Vecino [2], C.J. Secombes [1].*

[1] SFIRC, Institute of Biological and Environmental Sciences, University of Aberdeen, Tillydrone Avenue, Aberdeen AB24 2TZ, UK;
[2] EWOS Innovation A.S., Dirdal N-4335, Norway

Abstract
In fish, the innate immune system is the first line of defence against disease and is represented by physical barriers such as the scales, skin, epithelium, mucus and other body fluids. These structures not only provide protection against external agents, they also contain humoral components (complement factors, lysozyme, antimicrobial peptides, immunoglobulin) able to augment the defence of the organism. In particular, antimicrobial peptides (AMPs) can directly interact with the cell wall components of invading microbes, disrupting the membrane integrity of these pathogens. In addition in mammals these AMPs are known to activate the acquired immune response required for a more efficient and long-lasting protection. AMPs may also have value as markers of immunostimulant treatment or as adjuvants to generate more potent vaccines. The purpose of this research was two-fold. Firstly, to study the expression of the known repertoire of rainbow trout AMPs after administration of a novel functional feed containing peptidoglycan, to assess which AMPs are induced and when/where this occurs. Secondly, initial attempts were undertaken to produce the four known trout beta-defensins (omDB-1, omDB-2, omDB-3, and omDB-4), using transfection of RTG-2 and establishment of cell lines expressing these AMPs. Our findings shed light on the multiple roles of AMPs in fish not only as a first