spleen. In addition, vaccinated fish showed PCNA +/IgM + and PCNA+/IgT+ B cells in the mass of vaccine and cells (CVM) associated with the spleen, which may indicate an importance of the CVM during vaccination, which goes beyond a mere place of phagocytosis and exchange of material.
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**keywords:** Turbot, Spleen, Vaccination, B lymphocytes, Immunoglobulins

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**P-032.**

**Field validation of immunotoxic responses in the carpet shell clam (*Ruditapes decussatus*) from contaminated sites in the South Lagoon of Tunis (Tunisia)**

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Abstract
The aim of this study was to validate immunological alterations as ecotoxicological biomarkers to detect and monitor the biological effects of anthropogenic pollution in the South Lagoon of Tunis (Tunisia). Carpet shell clams (*Ruditapes decussatus*) were collected during summer and Winter from four locations: three of them within the polluted lagoon of Tunis (S1, S2 and S3) and from a clean site on the Mediterranean coast (Louza, Tunisia). To study the immunity of clams, the phenoloxidase, lysozyme, alkaline phosphatase, esterase, peroxidase, protease, antiprotease and bactericidal activities were measured in the haemolymph. Phenoloxidase activity was significantly lower in clams sampled from the three contaminated areas of the lagoon (S1, S2 and S3) than in those from the control point in both summer and winter. Lysozyme, esterase, protease and antiprotease activities were higher in the clams from site S3 than in the clams collected from the control site during winter. No significant variations were detected in the alkaline phosphatase, peroxidase and bactericidal activities of the clams collected from the four experimental sites. A significant seasonal effect was observed in clam immune status in winter. The results clearly showed that the affected biomarkers (mainly phenoloxidase) could be useful tools for biomonitoring clams in the study area.
This study was supported by the Ministry of Scientific Research and Technology, the University of Monastir (Tunisia), the Spanish Ministry of Economy and Competitiveness co-funded with Fondos Europeos de Desarrollo Regional/European Regional Development Funds (Grant no. AGL2014- 51839-C5-1-R) and Fundación Séneca de la Región de Murcia (Grupo de Excelencia. Grant no. 19883/GERM/15).
**keywords:** Biomarkers; Biomonitoring; Seasonality; Innate immunity; carpet shell clam (*Ruditapes decussatus*).

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**P-033.**

**Hepcidin, an antimicrobial and iron-regulated peptide that provides an ability to prevent bacterial diseases in grass carp (*Ctenopharyngodon idella*).**

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Abstract
Hepcidin is an antimicrobial peptide and a regulator of iron homeostasis which has three isoforms: -20, -22 and -25. While hepcidin-25 has been studied extensively, the physiological significance of other isoforms remains poorly understood. Herein, we focused on the analysis of the differences in antibacterial and iron regulatory functions of hepcidin-20 and hepcidin-25, looking for a derivative of hepcidin as a preventive drug for bacterial diseases. In this study, we examined the antimicrobial potentials of the two hepcidins in the form of synthesized peptides, hepcidin-25 and hepcidin-20. We found that hepcidin-25 and hepcidin-20 exhibited apparent bactericidal activities against both Gram-positive and Gram-negative bacteria in a dose-dependent manner. *In vitro*, the hepcidin-20 had better antibacterial activity than the hepidin-25. However, the antimicrobial activity on the cellular level has the opposite effect. We suspected that the iron-regulating function of hepcidin limits the available iron content of extracellular bacteria to enhance its bactericidal activity. Further tests indicated that only hepcidin-25 can block iron release from liver cell line L8824 via internalization and degradation of cellular iron exporter ferroportin, and restrain the use of iron in extracellular bacteria. This result also confirms our hypothesis. *In vivo*, recombinant *Ctenopharyngodon idella* hepcidin improved the survival rate of *C. idella* challenged with *Flavobacterium columnare*. In addition, the fish fed diet containing recombinant *C. idella* hepcidin had a higher survival rate than other pretreatment groups. The study showed that recombinant *C. idella* hepcidin regulated iron metabolism, causing iron redistribution, decreasing serum iron levels and increasing iron accumulation in the hepatopancreas. Immune-related genes were also evaluated, showing higher expression in the groups pretreated with recombinant *C. idella* hepcidin at an early stage of infection. In general, *C. idella* hepcidin not only has a direct killing effect on bacteria, but also reduces the available iron content of bacteria to inhibit bacterial growth. Our findings revealed a new role for hepcidin in fighting against bacterial infections and indicate a potential in controlling the bacterial infection in aquaculture.
**keywords:** *Ctenopharyngodon idella*; hepcidin; antibacterial;

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**P-034.**

**A novel *CQTRIM32* from red claw crayfish *Cherax quadricarinatus* inhibits white spot syndrome virus infection**

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Abstract
Tripartite motif-containing (TRIM) proteins are highly conserved molecules that participate in a variety of biological processes such as regulation of development, apoptosis, and innate immunity in vertebrates. In this report, we identified a *TRIM32* homolog (named *CqTRIM32*) in red claw