been focused on the molecular characterization, expression analysis by real-time PCR, both at basal condition and after *in vivo* challenges, and 3D structural studies on the g-type lysozyme from sea bass (*Dicentrarchus labrax*). Moreover, a recombinant sea bass lysozyme has been produced in *E. coli* and utilized to investigate the activity of the enzyme at different pH and temperatures and to perform antibacterial assays against typical fish pathogens. The cloned sea bass cDNA for g-type lysozyme (accession number FN667957) consists of 742 bp and translates for a putative protein of 188 amino acids. The molecular weight is 20251.40 Da with a theoretical pI of 9.53, two cysteine residues along the sequence and no putative signal peptide. These features of the enzyme are in agreement with the expected characteristics of a proper g-type lysozyme, except for the cysteine residues that in fish are quite variable in number. An alignment between known g-type lysozyme sequences evidences that the amino acid residues thought to be involved in the enzyme catalysis (Glu71, Asp84 and Asp95 in sea bass) are quite well conserved between mammalian, avian and fish sequences. Modelling of 3D structure has been performed on the template structure of g-type lysozyme from Atlantic cod. The catalytic site appears well conserved when compared with known structures of fish g-type lysozymes (cod and salmon). The basal expression of lysozyme transcripts is highest in gills, followed by head kidney and peripheral blood leukocytes. The lysozyme expression is up-regulated in head kidney leukocytes both after challenge with the fish bacterial pathogen *Photobacterium damselae* subsp. *piscicida* and with the viral pathogen nervous necrosis virus. The lysozyme activity, determined using as substrate *Micrococcus lysodeikticus*, was optimal at pH 5.5 and at a temperature of 30 °C. In conclusion, these results suggest that the identified g-type lysozyme is involved in innate immune responses of sea bass.

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P-412.

Purification and molecular characterization of the rhamnose binding lectin from sea bass (*Dicentrarchus labrax*) that agglutinate Gram positive and negative bacteria

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Abstract
Lectins are a multifamily of proteins present in almost all living organisms and due to the carbohydrate binding ability are involved in several biological processes, including immune responses. Biochemical and structural aspects of rhamnose-binding lectins from invertebrate and fishes led to the identification of a novel lectin family mainly localized to eggs and recently characterized by a unique sequence motif and a characteristic structural fold. Here we describe the purification, biochemical and molecular properties of a rhamnose binding lectin (DlRBL) isolated from sea bass (*Dicentrarchus labrax*) serum. DlRBL exhibits a subunit with two covalently arranged carbohydrate-recognition domains with an apparent MW of 24 kDa under reducing condition and a tetramer of 100 kDa under not reducing condition. The complete DlRBL sequence revealed that this lectin, like the other fish RBLs, possesses two tandemly arrayed CRDs.
The DlRBL sequence consists of an open reading frame encoding 212 aminoacid residues including 18 residue signal sequence at the N-terminal. The deduced size of 24.1 kDa for the mature protein is in good agreement with subunit size of the isolated lectin.
The DlRBL is strongly expressed in the serum after bacteria stimulation. That molecules are able to agglutinate Gram positive and Gram negative bacteria suggesting that DlFBL may play a role in host pathogen interaction.

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P-348.

Seabream (*Sparus aurata*) hierarchy among *alfa* and *beta* subordinates and dominant interplay affects stress responses and phagocytic activity by peritoneal cavity cells

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Abstract
The fish are affected by environmental conditions that can cause stress leading to changes in the innate immune system and plasma parameters increasing their susceptibility to disease. We examined the social stress of gilthead seabream (*Sparus aurata*) group. Social hierarchies ("*dominant*", subordinate: "*alfa*" and "*beta*") were characterised by behavioural changes, such as "aggressiveness" and "feeding order"; social hierarchy was established after an hour of exposure to social stress. The experimental models used were two: in the first the fish have been inserted simultaneously, in the second in a sequential manner.
To characterise physiological stress, we measured plasma levels of cortisol, glucose, lactate, and osmolarity and we observed that the levels of these stress markers were higher in subordinate individuals "*beta*" than in "*dominant*" and subordinate "*alfa*". Moreover, we examined the relationship between the social rank and the effects on the innate immunity. In particular the activity of the peritoneal cavity cells (PCC) was estimated using a phagocytosis assay employing yeast (*Saccharomyces cerevisiae*) and a respiratory burst activities following to *stimulus* with zymosan. After 24 h we revealed an increase of reactive oxygen intermediate showing that that social stress appeared to affect the cellular innate immune response of the subordinate specimens. In particular, the social stress mainly interested in the beta specimens. These results are according to Cammarata *et al.* (2012) which showed the same modulation response of PCC in subordinate individuals in paired experimental model.

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P-350.

Comparative cytomorphological features and PCNA expression pattern in haemic neoplasia from mediterranean mussels (*Mytilus galloprovincialis*) and Galician common cockles (*Cerastoderma edule*)

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Abstract
Haemic neoplasia (HN) is a pathologic conditions reported in several bivalve species in different geographic areas. In this study we describe the cytomorphological features and the proliferative behaviour, assessed by the PCNA (*Proliferating nuclear Antigen*) of the above disease in common cockle *Cerastoderma edule* and mediterranean mussel *M. galloprovicialis*. In mussels the presence of at least five types of atypical haemocytes was detected, including A and B type cells, previously described in *M. edulis* and *Mytilus sp.*, with predominance of A cells in early phase of the disease and B cells in more advanced stage. The PCNA immunostaining revealed positivity of 97–100% of the neoplastic cells, with both cytoplasmic (A cells) and nuclear pattern (B cells). Differently, in *C. edule* there wasn't distinctive morphological cell sub-population, recording atypical haemocytes with PCNA (range from 93–100%) showing nuclear expression in early phases of disease, and cytoplasmic in more advanced stages. The above findings suggest distinct histo-pathogenetic pathways for HN in mussels and common cockles, respectively.

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