seeds in the diets showed significant influence on the non-specific and specific immune system of catla. Respiratory burst, nitric oxide synthase and serum lysozyme were significantly (P<0.05) higher in fish fed with diet D2 compared to other two groups. Highest bactericidal activity was recorded in D1 diet fed catla compared to D2 and D3 diets fed fish. The antibody response was significantly (P<0.05) higher in D1 diet fed fish compared to D3. There was no significant (P > 0.05) difference between D1 and D2 diets fed groups. In kidney, TNF-α gene expression was up-regulated in seed supplemented diets fed groups (23-36% higher) compared to the control one. It was significantly (P<0.05) higher in D2 diet fed fish compared to others. There was up-regulation of lysozyme c and g expression in seed supiplanted diets fed catla compared to the control group. A direct relationship was found between the dose of seed in the diet and the expression of lysozyme c and g in kidney. In the gill, TNF-α expression was significantly (P<0.05) higher in D1 diet fed group compared to others. There was no expression of IL-10 in D1 in this tissue. Lysozyme c and g expression were significantly (P<0.05) higher in D2 diet fed group compared to other groups. There was no expression of TNF-α in the hepatopancreas of catla. IL-10 expression was up-regulated by the supplementation of seed in the diet. Lysozyme c and g expressions were significantly (P<0.05) higher in seed supplemented diet fed fish compared to the control one. This study showed tissue-specific gene expression pattern in catla. An inverse relationship was also found between the expression of TNF-α and IL-10 in various treatments.

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**O-250.** Characterisation of a functional intracellular type I interferon system in rainbow trout *Oncorhynchus mykiss*

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Abstract

Type I interferons (IFNs) are encoded by intronless genes in amnion cells and by intron containing genes in amphibians and fish. IFNs are known to activate a cellular antiviral response through cell surface receptors. In teleosts, multiple transcript variants have been reported and shown to be transcribed from alternative splicing of introns. Whether these transcript variants are translated into functional proteins is unclear and if so, their physiological importance requires to be determined. In this report, two alternatively spliced IFN transcripts were identified in rainbow trout *Oncorhynchus mykiss* and predicted to encode proteins lacking a signal peptide. Expression studies have shown they are induced in cells after stimulation with poly(I:C) and in head kidney of fish infected with virus. The proteins have been confirmed to be made from the IFN transcripts and are located within the cytoplasm. Overexpression of intracellular IFN (iIFN) proteins in cells or stimulation of cells with bacteria-derived recombinant proteins significantly up-regulates expression of antiviral genes and enhances cell resistance to viral infection. Fed fish. The two intracellular IFN receptor chains have been identified and found to be co-localised with the IFN ligands in the cytoplasm and are required to facilitate phosphorylation of STAT1 and STAT2. Taken together, our data demonstrate for the first time a functional intracellular IFN system in teleosts.

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**O-387.** Melanization cascade of shrimp and its importance for white spot syndrome virus infection

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Abstract

Melanization cascade, which is activated by the phenoloxidase (proPO) system, is one of a potent effective defense mechanism that plays a key role in the production of cytotoxic intermediates as well as melanin products for microbial sequestration in invertebrates. In the present study, the effect of melanization cascade of the black tiger shrimp *Penaeus monodon* on viral infection was investigated. Analysis of the effect of melanization on white spot syndrome virus (WSSV) infection showed that melanization reaction of shrimp significantly reduced the viral replication, whilst the survival rate of shrimp injected with melanization reaction-treated WSSV was found to be significantly higher than that of the control shrimp injected with untreated-WSSV. The dsRNA-mediated gene silencing of shrimp PmproPO gene followed by WSSV infection resulted in a significant increase in the cumulative mortality of the PmproPO silenced shrimp. In addition, injection of WSSV significantly reduced the hemolymph phenoloxidase (PO) activity of the infected shrimp at day 2 and 3 post
infection compared to the uninfected shrimp and the reduction of PO activity was compromised after an exogenous trypsin was added to the re-action. This result suggests that WSSV probably affect the proteolytic activity of some proteases in the proPO cascade of shrimp. Using yeast two-hybrid screening, a viral protein named WSSV453 was identified as a protein that capable to interact with a proPO-activating enzyme 2 (PmPPAE2) of P. monodon. WSSV453 is an uncharacterized protein with no putative domains and its sequence consists of 306 bp encoding for a predicted 101 amino acid protein with a calculated molecular mass and an estimated pf of 11.92 kDa and 9.95, respectively. Recombinant protein of WSSV453 and PmPPAE2 were then produced and co-immunoprecipitation assay indicated that WSSV453 interacts directly with the PmPPAE2 protein. Additionally, in vivo gene silencing of WSSV453 showed a significant increase in the hemolymph PO activity of the WSSV-infected shrimp when compared to the control shrimp. These results suggest the important role of shrimp proPO system in the defence against WSSV infection and also demonstrate the mechanism on inhibition of shrimp proPO cascade by the protease inhibitory activity of the viral protein.

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O-188.
The peroxiredoxin genes and their functions in the antivirus immunity of Kuruma shrimp, Marsupenaeus japonicus

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Abstract
White spot syndrome virus (WSSV) is one of the major pathogenic viruses that cause extensive mortalities in aquaculture. WSSV infection can bring about oxidative stress through the release of reactive oxygen species (ROS) which are deleterious to the cells. Peroxiredoxin (Prx) is a protein that protects the organisms against various oxidative stresses and functions in the intracellular signal transduction. To investigate the composition and the function of peroxiredoxin in shrimp, one of the model animals for aquatic, we cloned five peroxiredoxin genes from Kuruma Shrimp, Marsupenaeus japonicus (M. japonicus). Phylogenetic analysis and multiple alignment showed the genes we cloned belong to the Prx I, Prx II, Prx IV, Prx V and Prx VI subfamily and named as MjPrx I, MjPrx II, MjPrx IV, MjPrx V and MjPrx VI separately. RT-PCR analysis of the tissue distribution of peroxiredoxin showed the transcript of MjPrx IV, MjPrx V and MjPrx VI can be detected in all the tissues tested (he-mocytes, heart, stomach, gills, hepatopancreas, ovaries, spermary and intestine) while the MjPrx II was mainly detected in gonad and MjPrx I expressed mainly in stomach, intestine, testis and ovaries. The expression profiles of peroxiredoxin transcripts in gonad upon WSSV infection was investigated by Real-time PCR and showed different patterns. In spermary, the transcript of MjPrx II and MjPrx IV were upregulated after the 6 h WSSV challenge while MjPrx I and MjPrx VI were downregulated at the same time. In ovary, the transcripts of all MjPrxs were upregulated after the 6 h WSSV challenge. The MjPrx IV was selected for the further function study. The mature peptide of MjPrxIV was recombinant expressed in Escherichia coli system and showed peroxiredoxase activity in vitro. Furthermore, suppression of MjPrxIV by dsRNA resulted in the increase replication of WSSV in shrimp while injection of rMjPrx IV into shrimp could decreased the replication of WSSV in vivo. To our knowledge, this is the first integrative research about the Prxs and their function in the antivirus immunity of shrimp.

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O-437.
Survival of Edwardsiella tarda in fish serum relates to baterial surface LPS

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Abstract
Edwardsiella tarda is the etiological agent of edwardsiellosis, a systematic disease that affects a wide variety of marine and freshwater fish worldwide. Survival in fish serum is an important feature for virulent strains of E. tarda to escape from host immune surveillance and plays essential roles for the bacterial establishment and infection in fish. In the study, we investigated the complement activity in turbot serum after incubating with virulent E. tarda. Our results indicated that over 80% of bacteria can survive in fish serum as the complement molecules in the serum could not be activated by the E. tarda bacteria. Further study suggested that the intact LPS layer plays important roles for virulent E. tarda against complement activation in fish serum. The detailed mechanisms were still under investigation.

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O-393.
Trout transcriptome induced by the non-virion (NV) protein of VHSV. Identification of NV targets

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Abstract
Viral haemorrhagic septicemia virus (VHSV) infects many species of freshwater and marine fish, leading to important economic losses mainly for aquacultured salmonids. VHSV belongs to Novirhabdovirus genus, which also includes Hirame rhabdovirus, infectious haematopoietic necrosis virus (IHNV), and snakehead virus. Novirhabdoviruses are characterized by using probes from GenBank immune-related genes corresponding to interferons, VHSV-induced, macrophage-related, complement components, toll-like receptors, tumour necrosis factors, chemokines, interleukins, antimicrobial peptides, and cluster differentiation antigens. The results suggested that to favour VHSV infection, NV or VHSV exposition, might help to better understand the functional mechanism of NV protein as well as improving VHSV immune response pathways in trout that could be extrapolated to other affected species. Some of these newly discovered downregulated trout genes, after NV or VHSV exposition, might help to better understand the functional mechanism of NV protein as well as improving fish vaccines.

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