crayfish *Cherax quadricarinatus*. *CqTRIM32* was widely distributed in the tissues selected, with the highest expression in muscle, relatively abundant in haematopoietic tissue (Hpt) and the lowest presence in eyestalk. Multiple amino acid alignment showed that *CqTRIM32* contained a conserved RING-finger domain but without B-BOX domain and coiled-coil region, which was different from the traditional TRIMs family. Interestingly, the expression of *CqTRIM32* was significantly up-regulated at both 24 h and 48 h after white spot syndrome virus (WSSV) challenge in *vivo* in crayfish Hpt tissue. Meanwhile, the expression of *CqTRIM32* was significantly up-regulated at both 12 h and 24 h after WSSV challenge in *vitro* in Hpt cells. The quantity of WSSV was increased in red claw crayfish Hpt cell cultures after gene knockdown of *CqTRIM32* post WSSV infection, in which the transcription of both an immediate early gene *ie1* and a late envelope protein gene *vp28* of WSSV were clearly up-regulated. Taken together, our data provide the first evidence that *CqTRIM32* exerts the antiviral activity in a crustacean.

keywords: Tripartite motif-containing (TRIM); Antiviral; White spot syndrome virus (WSSV); Haematopoietic tissue (Hpt); *Cherax quadricarinatus*.

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P-035.

Lipid deposits and foamy macrophage-like cells in focal red and melanised muscle changes in Atlantic salmon (*Salmo salar*)

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Abstract

Focal melanised changes or “black spots” in farmed Atlantic salmon (*Salmo salar*) fillet is a common quality problem seen at slaughter. The changes develop during the seawater phase, starting as acute focal hemorrhages or “red changes” which progress into chronic inflammatory changes with melanisation. Regeneration in most changes remains ongoing without proper healing; a process that has been associated with the chronic persistence and replication of *Piscine orthoreovirus* (PRV).

Another chronically persistent feature in this condition is the histopathological presence of what appears as fat (seen as empty vacuoles) in both focal red and melanised changes. Previous studies have described vacuoles of various sizes assumed to be fat-containing, but as most studies have been carried out on formalin fixed and paraffin-embedded tissues, the content in such vacuoles has diminished during processing and histological investigations of lipids have hitherto been inconclusive.

Here, we use glutaraldehyde-fixed and frozen material, thus preserving the fat. Sections from both acute red and chronic melanised changes were stained with two different special stains (Sudan Black and Oil Red O) for detection of lipids. We show that most vacuoles indeed contain fat and that these are highly prevalent in the acute manifestations in areas of necrosis, haemorrhage and inflammation. We also show fat-containing vacuoles in chronic changes with melanisation, though with a different appearance; often in association with melano-macrophages. In addition, cells though to be foamy macrophages are identified and investigated by transmission electron microscopy. Based on our results, we discuss the potential role of fat in the development of focal melanised changes.

keywords: Inflammation; lipids; Macrophage; Melano-macrophage; Myositis

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P-036.

Immune response in turbot exposed to the ciliate parasite *Philasterides dicentrarchi*

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Abstract

Philasterides dicentrarchi is a marine scuticociliate that causes scuticociliatosis in farmed fish worldwide and is currently considered one of the most important pathogens of cultured flatfish. Although there is abundant information about the infections caused by *P. dicentrarchi* in fish and about how the ciliates and fish immune cells interact in vitro, little is known about the interaction between this ciliate and the fish immune system in vivo. In the present study, turbot (*Scophthalmus maximus*) were exposed twice to the parasite (on days 1 and 21). Immersion infection was performed by adding ciliates to tanks of seawater (18 ºC) to yield a final concentration of 4.5 x 104 ciliates/mL. Fish were exposed to the ciliates by immersion in the seawater for 20 min and were then transferred to tanks of clean seawater for 60 days. Control fish were immersed in seawater with no ciliates, and were subjected to the same conditions as the experimental fish. Four fish died of scuticociliatosis during the experiment. Fish (eight per group) were sampled on days 3, 7, 21 after the first exposure to *P. dicentrarchi* and on days 3, 7 and 40 after the second exposure. The presence of ciliates on the skin and gills was evaluated by qPCR. The IgM, IgT and IgD levels were measured in serum on days 3, 7 and 40 and in mucus on day 40 after the second exposure. Changes in gene expression of immunoglobulins, MHCII and other immune-related genes were determined by qPCR, in gills, skin, and spleen at all sampling times. There were no significant differences in serum IgM, IgD and IgT levels between experimental and control groups at any of the sampling times; however, there was a significant increase in mucus IgT levels 40 days after administration of the second exposure. The results of the qPCR analysis showed few changes of the immunoglobulin expression in the analyzed organs and a mild inflammatory response with the current infective dose.

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keywords: Turbot, *Philasterides dicentrarchi*, immunoglobulins, immune response, infection

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P-037.

Development of a reverse genetics system for snakehead vesiculovirus

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