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Involvment of mucous cells in the protective response of fish against enteric parasites

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Abstract
Fish immunology receives considerable attention due to the breeding of several species for human consumption. The pathogens of fish attach to mucosal membranes of skin, gills, and gut and can cause a significant loss of affected species, that result in economic constrains. In the intestinal mucosa, the presence of parasites causes a local inflammatory reaction with recruitment of several immune cell types at the sites of infection, such as mast cells, neutrophils, macrophages and rodlet cells. Moreover, parasites as intestinal helminths disrupt the mucosal layer at their attachment site and could facilitate entry of pathogenic bacteria or viruses. In the intestine, mucous cells produce and secrete mucins, different high molecular weight glycosylated protein, which first hydrate, lubricate and protect the intestinal epithelium from the mechanical injuries due to the transition of digesta. Mucous intestinal cells are considered elements of the innate immune system, as they secrete lectins, toxins, immunoglobulins, and anti-microbial peptides as well. Most studies show the hyperplasia and hypertrophy of mucous cells in the area of parasite attachment, and their high production and discharge of mucus at the epithelial surface and in the lumen. In the point of helminth attachment, intestinal mucus shows the prevalence of the high viscosity acid mucins, and worms can appear surrounded by an adherent mucus layer or blanket. Acid mucins can also envelope and eliminate other infected microorganisms and their possible role in the stimulation of the immune system has been previously reported. In the parasitized intestine, mucous epitelial cells often are close to endocrine cells of the diffuse endocrine system from which they possibly receive or send local signals. Additionally, mast cells that infiltrate the intestinal epithelium are observed near mucous cells. The current study reports quantitative data on the density of the different mucous cell types and the mucus composition in perch, Perca fluviatilis, and mullet, *Liza ramada*, infected with *Acantocephalus lucii* and *Neoechinorhynchus agilis* (Acanthocephala), respectively. Mucous cell types are discriminated by the histochemical reaction with Alcian Blue pH2.5 and Periodic Acid Schiff, and by lectin histochemistry. Eight lectins were used to characterize the differences in glycoconjugate composition of mucous cells in infected/uninfected fish. The results between these two fish-helminth systems are discussed and compared with the data we obtained previously in seven different fish-helminth systems.
keywords: Mucous cells; mucins; intestinal helminths; perch; mullet

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Cellular inflammatory response on marbled rockfish *Sebastiscus marmoratus* experimentally infected with *Ochroconis humicola*

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Abstract
The fungal infection with *Ochroconis humicola* was previously reported caused mortality in young stages of cultured marine fish in Japan, including devil stinger *Inimicus japonicus and* marbled rockfish *Sebastiscus marmoratus*. Occurrence of this infection is depending on the body size of the fish, adult fish appeared to be more resistant to this infection. This study compared the histopathological features of inflammatory response among the experimentally infected marbled rockfish of different body sizes. Fish were divided into small (25-35mm), medium (50-60mm) and large (70-80mm), then intraperitoneally injected with the conidia of *O. humicola* NJM1503 at concentration 1.105 per fish The fish in each group were kept in aquaria containing 40L artificial seawater and the temperature was maintained at 22ºC. The dead fish were collected accordingly and survived fish was sacrificed by using over dose FA100 after 30 days post infection (d.p.i). Spleen, kidney and liver of the fish were fixed and routinely embedded in paraffin and sectioned at 5μm. The serial sections were stained with hematoxylin and eosin (H&E), periodic acid-Schiff (PAS) reaction and Schmorl method. First mortality of infected small and medium fish was recorded 7 d.p.i and 15 d.p.i respectively. Mortality was not observed in large fish. Histopathologically, severe mycotic necrosis with large number of hyphae was observed in the infected small fish. Infected medium fish showed granulomatous inflammation in infected organs. This features was also observed in infected small fish dead from 20 d.p.i onwards. All large fish survived and showed more evident granuloma, including appearance of epithelioid cells and the hyphae was found inside the granuloma. The hyphae in large fish was encapsulated inside the granuloma and prevent further penetration. Therefore, it is suggested that the granulomas formation in infected large fish may suppress the fungal growth and showing resistant to this infection.
keywords: *Ochroconis*, granuloma, inflammation, epithelioid cells, fungal infection

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The inducible MicroRNA-21 negatively modulates the inflammatory response in teleost fish via targeting IRAK4

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Abstract
Eradication of bacterial infection requires timely and appropriate immune and inflammatory responses, but excessive induction of inflammatory cytokines can cause acute or chronic inflammatory disorders. Thus, various layers of negative mechanisms and regulators are needed to control the homeostasis of the immune system. miRNAs are a family of small non-coding RNAs that emerged as significant and versatile regulators involved in immune response. Recently, the molecular mechanisms of miRNA in host-pathogen interaction networks have been extensively studied in mammals, whereas the underlying regulatory mechanisms in fish are still poorly understood. In this study, we identify miR-21 as a negative regulator involved in regulating teleost inflammatory response. We found that lipopolysaccharide and *Vibrio anguillarum* significantly upregulated the expression of fish miR-21. Upregulated miR-21 suppresses LPS-induced inflammatory cytokine expression by targeting IL-1 receptor-associated kinase 4 (IRAK4), thereby avoiding excessive inflammatory responses. Furthermore, we demonstrated that miR-21 regulates inflammatory responses through NF-κB signaling pathways. The collective findings indicate that miR-21 plays a regulatory role in host-pathogen interactions through IRAK4-mediated NF-κB singling pathway.
keywords: miR-21; inflammation; IRAK4; NF-κB signaling

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