this work a recombinant VNAR antibody library was generated from a nurse shark (*Ginglymostoma cirratum*) host immunised with a combination of inactive SAV and recombinant SAV E2 protein. This library was panned using phage display technology to identify SAV-specific VNARs for use as immunological detection reagents. Of the novel VNAR clones identified three bound viable SAV with a high degree of sensitivity. These VNARs were shown to detect SAV subtypes 1, 2, 3, and 5 by ELISA. Two of the VNARs have specificity to SAV E2 glycoprotein. All of the VNARs showed characteristically high resistance to irreversible thermal denaturation. The subtype cross- reactivity and demonstrable robustness of these VNAR domains should enhance their utility as diagnostic reagents in the field.

**Keywords:** Shark IgNAR; VNAR antibody; Pancreas disease; Salmonid Alphavirus; Phage display

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## O-016.

### Influence of hypoxia stress on the immune response of pikeperch (*Sander lucioperca* L., 1758)

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Abstract
Animal welfare is a main issue in today's aquaculture. Pikeperch (*Sander lucioperca* L., 1758) is a native food fish of the northern hemisphere and very attractive for European aquaculture. It is susceptible to typical stress parameters of ongoing domestication. Unsuitable water temperatures and the lack of oxygen influence the well-being of fish and evoke to stress responses. Oxygen saturations lower than 40% are considered unfavourable for aquaculture facilities. These can be caused by insufficient water circulation, high stocking densities or high water temperatures. Temperatures above 20°C are optimal for pikeperch farming. Above 25°C, the oxygen concentration in water and the growth rate in pikeperch are reduced. These conditions are thus considered almost critical. Acute stress in fish modulates the immune system, including the release of a distinct set of cytokines and acute phase proteins. Chronic stress suppresses the immune system impairing the defense against pathogens. Hypoxia inducible factor (HIF)-1α is the most prominent regulator of hypoxic conditions in vertebrates, including Teleostei. It is expressed in nearly all immune cells and an interesting candidate as possible biomarker for hypoxic stress conditions. Little information has been published on the stress physiology of pikeperch. For successful breeding in aquaculture, details on effects of the stress parameters temperature and hypoxia will be important. We demonstrated recently, that a rise from 15°C to 25°C not only changes the expression pattern of heat shock induced genes (e.g. *HSP90AA1* and *SERPINH1*), but also of the hypoxia induced gene *HMOX1*, in liver and gills. The present study investigates the influence of oxygen deficiency on the early immune response of pikeperch. After peritoneal stimulation with inactivated *Aeromonas hydrophila* cells, we evaluated the transcript levels of possible biomarker genes, cell composition in certain tissues and immune cell activity. Preliminary data show that persistent lymphocytes of the peritoneum decrease in number after stimulation. Myeloid cells invade the site of infection to clear the pathogens. We suggest that hypoxia stress decreases the number of myeloid cells entering the peritoneum and these cells seem to originate from the head kidney. This work of the Campus bioFISH M-V was financed by the European Maritime and Fisheries Fund (EMFF) and the Ministry of Agriculture and the Environment Mecklenburg-Western Pomerania, Germany (Grant #: MV-II.1-RM-001).

**Keywords:** Pikeperch, hipoxia, welfare, immune System, stress Physiology

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## O-017.

### Anti-inflammatory and antioxidant properties of ethanol extract from Verbenaceae plant *Clerodendrum cyrthophyllum* turz in copper sulfate induced inflammation in zebrafish

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Abstract
Oxidative stress and inflammation are commonly present in several chronic diseases. Interestingly, these responses are closely related to pathophysiological processes. The inflammatory process can induce oxidative stress and *vice-versa* through activation of multiple pathways. Therefore, agents with antioxidant and/or anti-inflammatory activities are very useful in the treatment of many pathologies. *Clerodendrum cyrthophyllum* Turcz, a plant belonging to the Verbenaceae family, is used in Vietnamese traditional medicine for treating migraine, hypertension, inflammation of the throat, rheumatic arthritis. Despite its usefulness, studies on its biological properties are still limited. In this study, antioxidant and anti-inflammatory properties of an ethanol extract from leaves of *C.cyrtophyllum* (CCEE) were evaluated. In an *in vivo* anti-antioxidant test, 3 day-post fertilization (dpf) zebrafish larvae were treated with CCEE at 5, 20 and 40 μg mL-1 for 1 h and then exposed to 10 μM CuSO4 during 20 min to induce oxidative stress. Fluorescent probes were used to detect and quantify oxidative stress by measuring the fluorescent intensity (FI) in larvae. At 5 and 20 μg mL-1, the CCEE displayed a significant reduction of FI when compared with control group, indicating that it had profound antioxidant effects, reducing or preventing oxidative stress from CuSO4. Moreover, an experiment on 3 dpf zebrafish larvae treated with CCEE at 5, 20 and 40 μg mL-1 for 1 h and then exposed to 10 μM CuSO4 for 4 h showed that CuSO4 elicited a general stress response by the upregulation of *hsp70* and *gadd45bb,* involved in inducible DNA damage repair. But, the co-administration of CCEE protected zebrafish larvae against oxidative damage of CuSO4 through a down-regulation of *hsp70* expression and the upregulation of glutathione S-transferase genes *gstp1* and *gstp2*. To evaluate the anti-inflammatory properties of CCEE, a similar experiment was designed, using 10 μM CuSO4 to stimulate inflammation reaction. After 4 and 24 h of CuSO4 exposure, the expression of genes related to inflammatory process was analyzed in zebrafish larvae. Due to the copper accumulation in zebrafish tissues, the damage and oxidative stress were exacerbated overtime, resulting in the upregulation of genes related to inflammatory process such as *COX-2, PLA2*, *C3a*, pro- and anti-inflammatory cytokines (*IL-1, TNF-α; IL-10* respectively). However, the association of CuSO4 with CCEE reduced significantly *COX-2, PLA2, C3a, IL-1*. Taken together, the results suggested that CCEE has potent anti-oxidant and anti-inflammatory activities and may be useful in the treatment of various inflammatory diseases.

**Keywords:** Anti-inflammation, oxidative stress, anti-oxidant, CUSO4, zebrafish larvae, *Clerodendrum cyrthophyllum*

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