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TargetFish brings together leading European research groups that are experts on the fish immune system and enterprises from the Biotech and Veterinary sectors that aim to commercialize fish vaccines for European fish farming. By developing a targeted vaccination strategy, TargetFish will prevent important fish diseases in European aquaculture industry.
This highlight is part of monthly progress updates by the TargetFish consortium.

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Characterization of a g-type lysozyme from European sea bass

The first line of defence in fish is represented by the innate immune system with lysozyme being one of the molecules involved. The animal kigdom shows the presence of three types of lysozymes; c-type (chicken or conventional), g-type (goose-type) and i-type (invertebrate type) lysozyme, which have been widely studied and identified as hydrolytic enzymes with the capability to cleave the beta-(1,4)-glycosidic bond between N-acetylmuramic acid (NAM) and N-acetylglucosamine (NAG) residues of peptidoglycan, a polymer found in the bacterial cell wall.
Researchers from the laboratory of Animal Biotechnology of the University of Tuscia in Viterbo (Italy), part of the TargetFish consortium, have studied lysozyme

of sea bass (*Dicentrarchus labrax* L.), as an important fish species for Mediterranean aquaculture.

The work started with a molecular characterization and associated gene expression analysis by real-time PCR, both in unstimulated fish and after *in vivo* challenge (Molecular Immunology, 2014, 62: 10-18). Similar to most g-type lysozymes found in fish, sea bass g-type lysozyme has no predicted signal peptide, suggesting that the enzyme may not be secreted from cells. Gene transcription was up-regulated in head kidney leukocytes both after *in vitro* stimulation with LPS, and after in vivo infection with the bacterial pathogen *P. damselae*. This suggests this enzyme is indeed involved in the innate immune responses of sea bass.

A bacterial recombinant sea bass lysozyme was produced to investigate the biological activity at different pH and temperatures and to perform antibacterial assays against typical fish pathogens. Lytic activity, as determined using *Micrococcus lysodeikticus* bacteria as substrate, was optimal at pH 5.5 and at a temperature of 30 ◦C. Antibacterial activity was tested against the bacterial pathogens *Vibrio anguillarum* (two strains) and *Photobacterium damselae* subsp. *Piscicida,* often found to infect sea bass.

From a practical aspect, these results will not only help to understand sea bass immune responses, but could also lead to the use of lysozyme activity measurements as simple indicators of immune function able to interfere with the growth of bacterial pathogens important to sea bass aquaculture.

For more information, please visit targetfish.eu or contact the consortium via targetfish.cbi@wur.nl

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