

Immunodetection of rainbow trout IL-8 cleaved-peptide: Tissue bioavailability and potential antibacterial activity in a bacterial infection context

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Abstract

Chemokines such as IL-8 are part of an important group of proinflammatory response molecules, as well as cell recruitment. However, it has been described in both higher vertebrates and fish that IL-8 has an additional functional role by acting as an antimicrobial effector, either directly or by cleavage of a peptide derived from its C-terminal end. Nevertheless, it is still unknown whether this fragment is released in the context of infection by bacterial pathogens and if it could be immunodetected in tissues of infected salmonids. Therefore, the objective of this research was to demonstrate that the C-terminal end of IL-8 from *Oncorhynchus mykiss* is cleaved, retaining its antibacterial properties, and that is detectable in tissues of infected rainbow trout. SDS-PAGE and mass spectrometry demonstrated the cleavage of a fragment of about 2 kDa when the recombinant IL-8 was subjected to acidic conditions. By chemical synthesis, it was possible to synthesize this fragment called omIL-8 α_{80-97} peptide, which has antibacterial activity against Gram-negative and Gram-positive bacteria at concentrations over 10 μ M. Besides, by fluorescence microscopy, it was possible to locate the omIL-8 α_{80-97} peptide both on the cell surface and in the cytoplasm of the bacteria, as well as inside the monocyte/macrophage-like cell. Finally, by indirect ELISA, Western blot, and mass spectrometry, the presence of the fragment derived from the C-terminal end of IL-8 was detected in the spleen of trout infected with *Piscirickettsia salmonis*. The results reported in this work present the first evidence about the immunodetection of an antibacterial, and probably cell-penetrating peptide cleaved from the C-terminal end of IL-8 in monocyte/macrophage-like cell and tissue of infected rainbow trout. © 2021 Elsevier Ltd

Author keywords

Antimicrobial peptides; Cell-penetrating peptide; IL-8 chemokine; Innate immune response; Salmonids