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Effects of in vitro altered microflora on immunolocalization of ladderlectin and intelectin in trout intestine

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Lectins are carbohydrate-binding proteins or glycoproteins present in all types of organisms. They serve different biological functions including the roles in the innate immune system by recognizing carbohydrates that are found on the surface of potential pathogens. Soluble lectins have been identified from the plasma and mucus of various fish species. Rainbow trout ladderlectin (RTLL) and intelectin (RTInt) are two plasma proteins implicated in innate immune surveillance and pathogen elimination based upon their ability to bind Gram-negative bacteria and chitin.

The present work, by using an in vitro model, was aimed to evaluate the expression of RTLL and RTInt, as visualized by specific antibodies, in the trout intestine exposed to distinct experimental conditions. The removed intestines were separately exposed to a mixture of the probiotic bacteria Lactobacillus rhamnosus, e Lactobacillus paracasei (group 1), a suspension of the pathogen bacteria Vibrio anguillarum (group 2), a mixture of the probiotics above, followed by the pathogens V. anguillarum (group 3). In the control group (group 4), the intestines were exposed to sterile saline solution and TSBgs in the same conditions, as above.

Following exposure to V. anguillarum, a higher lectin reactivity was found, as compared with the controls, at the apical cell membrane of the epithelium, probably as a consequence of a stronger secretion induced by the pathogen. Consistently, the intestinal goblet cells, which in the controls proved to be the main site of the lectin expression, in the group 2 appeared unstained and nearly devoid of their contents. An increased immunostaining was observed also within inflammatory leucocytes, club cells and SP-positive cells. Notably, in the group 3, both the immunohistochemical pattern and the Western blotting analysis indicated that exposure of intestine to probiotics prior to V.anguillarum affects positively the RTInt expression, by reducing the pathogen-induced effect. These preliminary findings support a role for both RTLL and RTInt as putative innate defence molecules on intestinal mucosal surfaces. Further studies are required to confirm a possible involvement of probiotics in the RTInt-mediated immunomodulation.

Keywords: Trout ladderlectin, trout intelectin, probiotics, immunohistochemistry.