New research tool reveals mucosal interaction and dietary influence in fighting sea lice infections

By Dr. Karin Pittman, University of Bergen, Norway

As the world's leading producer of Atlantic-farmed salmon, Norwegian fish farmers, governmental regulatory bodies and NGO's are keen to promote sustainable practices and ensure that both wild and farmed fish are healthy and robust. Much controversy surrounds the presence of naturally occurring sea lice, their interaction with wild stocks and the role that farms play as a reservoir. Stringent reporting procedures are required and monitored by external authorities to ensure that sea lice in farm populations are monitored and controlled and that therapeutic intervention is coordinated on an area basis so as to maximize their effectiveness.

The primary defense mechanisms of fish against any pathogens and parasites are the skin, gills and gastrointestinal tract. These tissues are covered by a fine mucous layer which possesses strong anti-microbial and parasitic defenses (lysozymes, immunoglobins, lectins, crinotoxins and antibacterial peptides) inherent in the immune system of the fish. As sea lice attach themselves to the skin this mucosal barrier is the first natural line of defence with which the fish defends itself. The rate of renewal of this mucous barrier, from the underlying goblet cells that generate it, is an important factor in the effective-ness of this defence mechanism. Until recently this mechanism has been poorly understood given the key role it plays in fish health.



Trials at the University of Bergen in Norway to find a nonchemotherapeutic solution to the sea lice challenge

Recently two animal health companies, Produs Aqua and Alltech, teamed up with the University of Bergen in Norway to try to find a nonchemotherapeutic solution to the sea lice challenge and focused on the salmon's skin and its natural defense system to help keep fish skin healthy and fight against these parasites.

A series of trials was conducted and a new technology developed. This novel designbased stereological method (Pittman et al 2011, 2013) has enabled the quantification of developing mucous cells, their density and location on the salmon's skin. This allows comparative studies to be undertaken for the first time and has shown the differences in this system between male and female salmon, density of mucosal cells at different locations on the salmon and even the influence of genetic stock in terms of mucous productivity. This technique has now demonstrated differences in mucosal production associated with different feeds and their components and allows comparisons between fish stocks.

Trials were conducted by incorporating a feed additive (Aquate[™] from Alltech) to assist with the production of more protective mucous and thus support the salmons' defense system against sea lice. Four field trials with several hundred thousand salmon were carried out and the results are unique.

Through these studies it was demonstrated that mucous cells the are much larger on the dorsal skin of the salmon than on the head and upper tail where parasites prefer to attach themselves (Figure 1). The density of mucous cells is higher in the gills than in the skin, is higher on the dorsal and the mid-ventral area and generally increases with fish size (Figure 2). This method also showed the trend to differences between the mucosal tissues of males and females (Figure 3). Cell densities almost tripled in one trial period and mean cell sizes doubled. Interestingly when the Aquate diet was discontinued a decline in both of these characteristics was ob-







Figure 1



Significantly denser mucous cells on the dorsal (p<0.05)

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served over a short period of time in that trial.

In a subsequent trial involving several hundred thousand fish over a four month period with four diets (one control and three test diet formulations) regular mucous cell growth over time was observed. However when the fish were subjected to a natural sea lice challenge from the environment the control group had the highest number of sea lice, whereas the test groups had significantly lower lice numbers. All the fish on the



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test diets showed in response to the sea lice reduced mucous cell sizes, while maintaining their density. Immediately after the occurrence of the sea lice they also had the highest cell area/cell density ratio. In contrast the average cell size in the control group continued to increase.

This suggests that the ability to change the mucosal tissue's cellular response may be a key element of pathogen resistance. It further indicates that diet composition is integral in the ability of the tissue to exhibit that response, perhaps by providing essential building blocks for mucosal cell turnover.

This new quantitative methodology is robust and enables comparative studies to be undertaken that evaluate skin mucosal response to sea lice infestations and dietary influences. The strengthening of the innate immune system at the mucosal level plays an important role in the natural defence mechanisms of the fish and may enable the reduction in chemotherapeutic intervention in sea lice control so benefitting farmers and environmentalist alike.

References

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These fascinating new tools have been made available to the industry through the establishment of Quantidoc SA, a commercial entity under the technology transfer system of the University of Bergen and more details can be obtained from <u>Dr. Karin Pittman, University of Bergen</u>.



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