Immunization of rainbow trout against Flavobacteriosis

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Abstract

Flavobacteriosis is primarily caused by *Flavobacterium psychrophilum* and *F. columnare*. Together they are the major bacterial pathogen contributors to losses in the rainbow trout (*Oncorhynchus mykiss* Walbaum) farming industry in Idaho, USA and freshwater salmonid farming around the world. Outbreaks of Flavobacteriosis can result in significant mortality but further losses due to deformities and reduced growth in survivors contribute to additional and potentially greater economic impacts for the farmer. Disease prevention is essential to the continued development of aquaculture around the world and remains a primary strategy for avoiding Flavobacteriosis with vaccination being the most effective method of preventing or minimizing disease. Killed, attenuated, subunit and a variety of other vaccine formulations against Flavobacteriosis have been developed and tested over the years and despite a number of publications there are still no commercially available vaccine(s) for rainbow trout.

Equally important to the development of Flavobacteriosis vaccines is the development of mass immunization strategies. While injection is clearly the most effective vaccination method, the limitations of this technique require that different methods continue to be investigated. As such it may be possible to improve upon the efficacy of injection delivery. The highly efficacious DNA vaccines against the fish Novirhabdoviruses require intramuscular (i.m.) delivery to be effective, all other potential delivery strategies do not elicit the level of protection seen when these vaccines are delivered i.m.. It appears that the injection site within the muscle is not limited to the epaxial muscle around the dorsal fin as three alternative intramuscular delivery sites have also been shown to be equally efficacious [1]. Additionally, an attenuated IHNV vaccine was found to be equally efficacious as the IHNV DNA vaccine when delivered intramuscularly [2]. This suggests that the muscle site may provide enhanced efficacy for other types of vaccines as well and may warrant further investigation.

One slightly different, alternative “vaccine” we have evaluated is an *Enterobacter* sp. as a surrogate vaccine against *F. psychrophilum*. Originally developed as a probiotic [3] it shows promise, at least for short term protection when delivered orally [4] and by i.m. injection.

Groups of 350 rainbow trout (mean weight, 2 g) were either left unhandled or i.m. injected with 25 µl of a suspension of *Enterobacter* (1.0 optical density at a wavelength of 525 nm) from either a 48 h or 72 h culture grown in tryptic soy broth on a stir plate at room temperature. At 7, 28 and 56 days post-injection duplicate 25-fish groups were challenged with a virulent strain of *Flavobacterium psychrophilum* (isolate 259-93) at two different doses using a standardized subcutaneous injection challenge methodology. Mortality was monitored daily for 21–28 d and *F. psychrophilum* was reisolated from at least 20% of the fish that died each day. Comparisons were made for cumulative percent mortality and relative percent protection for each treatment at each time point and for each challenge dose.

Table 1. Cumulative percent mortality in rainbow trout intramuscularly injected with an *Enterobacter* probiotic after 48 or 72 h of growth or injected with saline. Challenge evaluations with *Flavobacterium psychrophilum* (isolate 259-93) were conducted at two different doses using a standardized subcutaneous injection challenge methodology at different durations after the fish were injected with the *Enterobacter* probiotic.

		Cumulative Percent Mortality		
Time After Injection	Challenge Dose	Unhandled	48 h Culture	72 h Culture
7 d	0.2 0.4	94 94	32 12	26 22
28 d	0.2 0.4	71 67	36 39	33 33
56 d	0.2 0.4	82 70	67 62	70 70

Table 2. Relative percent protection in rainbow trout intramuscularly injected with an *Enterobacter* probiotic after 48 or 72 h of growth or injected with saline. Challenge evaluations with *Flavobacterium psychrophilum* (isolate 259-93) were conducted at two different doses using a standardized subcutaneous injection challenge methodology at different durations after the fish were injected with the *Enterobacter* probiotic.

		Relative Percent Protection		
Time After Injection	Challenge Dose	Unhandled	48 h Culture	72 h Culture
7 d	0.2 0.4	-	66 87	72 77
28 d	0.2 0.4	-	49 42	53 51
56 d	0.2 0.4	-	18 11	15 0

Again, equally important to the development of Flavobacteriosis vaccines is the development of mass immunization strategies. An efficacious vaccine is only of use to producers if it can be delivered cost effectively to small fish. With this in mind, a rifampicin attenuated *F. psychrophilum* vaccine has been developed and field tests suggest it can be effective when delivered by immersion [5]. Similarly a live attenuated *F. columnare* vaccine licensed for use in channel catfish is also being evaluated for use in rainbow trout.

References

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