# DETECTION OF STRESS INDUCIBLE FURUNCULOSIS IN SALMONIDS VACCINATED WITH WATER AND OIL-BASED FURUNCULOSIS VACCINES

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This note presents the results of stress testing for the detection of covert *Aeromonas salmonicida* infection in two species of salmonids, Brown trout, *Salmo trutta* and Atlantic salmon, *Salmo salar* L., which had previously been vaccinated against furunculosis. Stress testing of fish was carried out by the protocol of McCarthy (1977). This protocol requires holding of fish in batches, thus mortality figures given here cannot be used to generate quantitative estimations of the incidence of *A. salmonicida* infection in the test groups. A portion of the head kidney of all mortalities incurred during the test and survivors of the test were examined bacteriologically for the presence of A. *salmonicida* on Tryptone Soya Agar (TSA)(Oxoid, UK). The ability of TSA batches to support growth and pigmentation of *A. salmonicida* was confirmed, using a positive control strain, prior to use. Plates were incubated at 22°C for 72 h and any putative *A. salmonicida* colonies were confirmed by microscopic examination and with an *A. Salmonicida* specific latex agglutination test (Drinan, 1985).

The Brown trout had been injection vaccinated as yearlings in late October-November with a water-based vaccine according to the manufacturers recommendations. The water temperature at the time of vaccination was 10-11°C and the fish were in the weight range 100-200 g. Two batches of 50 vaccinated Brown trout were sampled in February when the water at the hatchery was 6°C and the fish were in the weight range 200-300 g. Following two days at ambient temperature (4.5-5.5°C) in the fish holding facilities one batch of fish were stress tested. Mortalities occurred on day 8 (n=6) and day 9 (n=5) of the stress test. Bacterial colonies isolatcd from the kidneys of these mortalities were confirmed as *A. salmonicida.* Following the 14 days of thc test the 39 remaining fish were culled and *A. salmonicida* was isolated from the kidneys of 20 of these fish. The second batch of 50 vaccinated Brown trout were held at ambient temperature (5-7°C) in the fish holding facility for the duration of the test but were not subjected to stress testing. The kidneys of this non-stressed group were also examined bacteriologically for the presence of A. *salmonicida* but the organism could not be cultured from any sample. The failure to isolate A. *salmonicida* from the kidneys of unstressed fish was not supprising and underlines the inefficiency of this method for detecting covert intections, when compared to stress testing (Bullock and Stuckey, 1975; Jensen; 1977; Scallan, 1983; Rose *et al.,* 1989; Hiney, 1994; Hiney *et al.,* 1994).

The Atlantic salmon had been injection vaccinated in mid-November with an oil-based vaccine according to the recommendations of the manufacturer. The water temperature at the time of vaccination was 8-9°C and the fish were in the weight range 10-15 g. A batch of 150 salmon were sampled in March as pre-smolts when the watcr temperature at the hatchery was 7°C and the fish were in the weight range 35-50 g. Following two days at ambient (temperature (6-7°C) in the fish holding facilities the fish were stress tested. Mortalities occurred on day 5 (n=12) and day 6 (n=45) of the stress test and A. *salmonicida* was isolated from the kidneys of all mortalities. The test was aborted at day 7, all remaining fish were culled and their kidneys examined bacteriologically. A. *salmonicida* was isolated from the kidneys of 41/92.



Available data on the prcvious disease histories of the hatcheries from which these fish were sampled was incomplete and precludes detailed analysis of the background to the events reported here. However, the limited data presented demonstrates that covcrt *A. salmonicida* infection could be detected in Brown trout and Atlantic salmon that had been vaccinated with a water-based and oil -based vaccine preparation, respectively. Thus, in thc case of the groups dcscribed, vaccination either failed to eliminate covert infections present in the fish prior to vaccination or failed to prevent establishment of covert infections in these fish following vaccination. These results imply that at least two events must have occurred. *A. salmonicida* must have been prcsent in or on the vaccinated fish, and following the stress resulting from the performance of thc test the extent of protection provided by vaccination was insufficient to prevent clinical disease.

Experience in Galway has shown that the stress test has validity in predicting incidences of furunculosis consequent on commercial transfer of Atlanlic salmon smolts to sea. This validation of the test was, however, obtained using data from studies of unvaccinated salmonids. The extent to which the results of a stress test can be used to predict the future disease events in vaccinated fish following the stress of sea transfer is unknown. Similarly the extent to which vaccination of covertly infected fish will prevent these fish from acting as vectors of disease is unknown. The finding that the stress test allowed the detection of covert *A. salmonicida* infections in vaccinated fish is noteworthy but raises a number of qucstions that have not been addressed in this work.

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*References*

Bullock, G. L. and Stuckey, H. M. ( 1975). *Aeromonas salmonicida:* detection of asymptomatically infected trout. *Prog. Fish-Cult.,* **37**, 237-239

Drinan, E. (1985). *Studies on pathogenisis of furunculosis in salmonids.* PhD Thesis, National University of Ireland.

Hiney, M. P. (1994). *Development and validlalion of detection techniques for the fish pathogen* Aeromonas salmonicida. PhD Thesis, National University of Ireland.

Hiney, M. P., Kilmartin, J. J.. and Smith, P. R. (1994). Detection of *Aeromonas salmonicida* in Atlantic salmon with asymptomatic furunculosis infections. *Dis. Aquat. Org.,* **19**,161-167.

Jensen, N. J. (1977). The diagnosis of Furunculosis in salmonids. *Bull. Off. Int. Epizoot.,* **87**, 469-473.

McCarthy, D. H (1977). Some ecological aspects of the bacterial fish pathogen *Aeromonas salmonicida.* In: F. A. Skinner and J. M. Shewan (eds), *Aquatic Microbiology.* Academic Press, London, pp. 299-324

Rose, A. S., Ellis, A. E., and Adams, A. (1989). An assessment of routine *Aeromonas salmonicida* carrier detection by ELISA. *Bull. Eur. Ass. Fish Pathol.,* **9**, 65-67.

Scallan, A. (1983). *Investigations into asymptomatic carriers of furunculosis.* PhD Thesis, National University of Ireland.