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Alternative control measures to antibiotics: *Flavobacterium psychrophilum* in rainbow trout fry and the effect of salt and warm temperatures.

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<u>Introduction</u>: *Flavobacterium psychrophilum* is a worldwide bacterial pathogen affecting salmonid aquaculture [Rainbow Trout Fry Syndrome (RTFS) and Bacterial Coldwater Disease (BCWD)]. Due to the unavailability of a commercial vaccine and the rise of AMR, alternative measures are of ecological and economic interest.

<u>Methodology</u>: In this study, we investigated the effect of salt water (1%) and warm temperature (18±1°C) in the control of *F. psychrophilum* in rainbow trout fry. Fish experimental trials were set up. In the first part of the experiment, fish (0.7 g) were at first infected by bath challenge and then exposed to either salt water (1 dpi – 12°C) or to increased water temperature (1 dpi or at onset of mortalities). Water parameters were changed gradually over 24 hours. Negative infection controls were included (freshwater and salt water at 12°C; warm freshwater at 18°C) and fish survival was followed over time. In the second part of the experiment, a cohabitation challenge was established and the effect of salt water (1%) on disease transmission evaluated (1.5-2 g/fish). Fish survival was followed over time.

<u>Results</u>: Following bath challenge, the salt treatment delayed the appearance of clinical disease while in the warm temperature groups fish survival decreased more rapidly than the positive control. In the second part of the experiment, we performed a co-habitation challenge. Ip injected fish reached 0% survival within two weeks in both groups. Cohabitant fish swimming in salt water had a significant increase in survival (42.6%) compared to the positive controls (17.9%). Infected dead and moribund fish were confirmed positive for *F. psychrophilum*.

<u>Conclusions</u>: Increasing water salinity delayed and partly prevented RTFS. The delay can be an advantage, as it gives time to achieve AMR test results and initiate treatment before reaching heavy mortalities in a fish batch. Further studies should evaluate the robustness of the preventive effect of this approach, its effect on the microbial communities (fish and farm environment), and whether it could be combined with other measures like e.g. phage therapy. The effect of the warm water was surprising, as *F. psychrophilum* favours colder temperatures and grow slower at higher temperatures.

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