COLORADO PARKS & WILDLIFE Dual Disease Resistance in Rainbow Trout

EVAUATING RESISTANCE TO BACTERIAL COLDWATER DISEASE AND WHIRLING DISEASE IN COLORADO'S TROUT





Bacterial coldwater disease (BCWD) is caused by the

bacterium *Flavobacterium psychrophilum*. Found worldwide, BCWD causes significant complications and death in hatchery trout populations. Outbreaks typically occur at temperatures between 39 and 50°F. Infected fish show a broad range of clinical disease signs including lesions, spiral swimming, "black tail", spinal deformities, and pale or necrotic gills. Mortality can be high if left untreated, and antibiotics are commonly used to treat BCWD. As an alternative, the

USDA National Center for Cool and Cold Water Aquaculture (NCCWA) developed a Rainbow Trout strain that is resistant to *F. psychrophilum*. With the help of Utah Division of Wildlife Resources, *psychrophilum*-resistant Rainbow Trout (PRR) were incorporated into the CPW hatchery system to help manage BCWD outbreaks.

Whirling Disease (WD)

Whirling disease (WD) is caused by the parasite *Myxobolus cerebralis*. Signs of infection include skeletal deformities, "black tail", and "whirling" or spiral swimming. WD cannot be treated, and susceptible fish typically die within their first year. *M. cerebralis* has a complex multi-stage life cycle, making it extremely difficult to remove from aquatic environments. One option for management is to use *M. cerebralis*-resistant fish. The Hofer strain is genetically resistant to *M. cerebralis*, however, it is domesticated and shows reduced survival in the wild. To increase survival, CPW crossed the Hofer with wild Rainbow Trout strains. The resulting crosses (HxC, Hofer by Colorado



River Rainbow; HxH, Hofer by Harrison Lake Rainbow) are resistant to *M. cerebralis*, and survive and reproduce in the wild. Stocking *M. cerebralis*-resistant Rainbow Trout has helped reduce WD in aquatic systems throughout Colorado.

Evaluating Dual Resistance via Dual Exposure to BCWD and WD

CPW uses the PRR to reduce mortality in the hatchery due to *F. psychrophilum* outbreaks. However, it is unknown if the PRR are resistant to *M. cerebralis*. Stocking PRRs with no resistance to *M. cerebralis* could result in high losses from WD, as well as increased *M. cerebralis* prevalence. Conversely, although resistant to *M. cerebralis*, the HxH shows increased mortality in the hatchery during BCWD outbreaks. This study examined if crossing the PRR with the HxH resulted in fish that are genetically resistant to both *F. psychrophilum* and *M. cerebralis*.

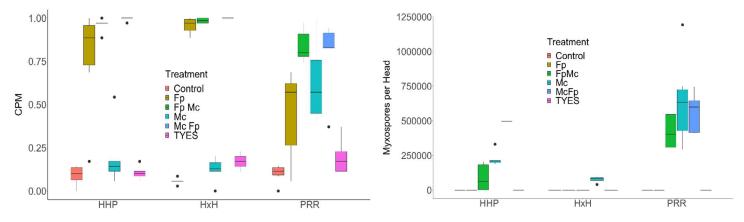


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The PRR, HxH, and HHP, the first generation cross between the PRR and HxH, were used for the dual exposure experiment. To test genetic resistance to both pathogens, fish were exposed to *F. psychrophilum*, *M. cerebralis*, or both. Fish were exposed to *F. psychrophilum* using injections under the skin, and to *M. cerebralis* using bath exposure to triactinomyxons, the waterborne infectious stage of the parasite. Mortality from *F. psychrophilum* occurs within 28 days of exposure, and was the first measureable endpoint of the experiment. Fish were then reared for six months to allow development of myxospores, the countable form of *M. cerebralis* in fish, and disease signs and myxospore counts were obtained from the fish remaining at the end of the experiment.

The PRR experienced the lowest cumulative mortality when exposed to *F. psychrophilum*, showing that it was more resistant to *F. psychrophilum* than either the HxH or HHP. However, with higher myxospore counts than either the HxH or HHP, the PRR did not show any resistance to *M. cerebralis*. The HxH had much lower myxospore counts and was more resistant to *M. cerebralis* than the PRR. However, as had been observed in the hatchery, the HxH did not exhibit any resistance to *F. psychrophilum*. The myxospore counts for the HHP were intermediate to those of the HxH and PRR, and the HHP experienced high cumulative mortality when exposed to *F. psychrophilum*, showing that it had not gained resistance to *F. psychrophilum* from the PRR. Coinfection with *F. psychrophilum* and *M. cerebralis* increased mortality in the PRR, HxH, and HHP compared to single-pathogen exposure.



Left: Cumulative percent mortality (CPM) for the HHP, HxH, and PRR across six treatments, 1) control (no pathogen exposure), 2) *F. psychrohilum* only (Fp), 3) exposure to *F. psychrohilum* followed by exposure to *M. cerebralis* (Fp Mc), 4) *M. cerebralis* only (Mc), 5) exposure to *M. cerebralis* followed by exposure to *F. psychrophilum* (Mc Fp), and 6) mock injection with TYES media (TYES). Right: Myxospores per fish head as a measure of *M. cerebralis* infection for the HHP, HxH, and PRR in each of the six treatments at the end of the experiment.

Management Implications

The results of this experiment suggest that it was not possible to create fish that are resistant to both *F*. *psychrophilum* and *M*. *cerebralis* using the HxH and PRR. In a follow up experiment, we exposed pure strains and their crosses to *F*. *psychrophilum* and found that the first generation cross between the Harrison Lake Rainbow Trout and the PRR showed reduced mortality and resistance to *F*. *psychrophilum*. Therefore, it may be possible to produce Rainbow Trout that are resistant to both pathogens, though their resistance to *M*. *cerebralis* still needs to be evaluated. More research is needed to determine if other strains not included in these experiments can be used to create fish resistant to both pathogens. Until then, hatchery outbreaks of BCWD in susceptible fish such as the HxH can be reduced by maintaining high water quality, flows, and reduced densities to prevent stressful rearing conditions. Using PRRs in hatcheries where *F*. *psychrophilum* outbreaks are common will help reduce mortality from BCWD on the unit, but due to their susceptibility to WD, these fish should not be stocked in aquatic systems in which *M*. *cerebralis* is established.

Associated Literature

Fetherman, E. R., B. Neuschwanger, B. W. Avila, and T. B. Riepe. 2020. Sport Fish Research Studies. Annual Report. Colorado Parks and Wildlife, Aquatic Research Section. Fort Collins, Colorado.