

THE SAFETY OF CHLORAMINE-T USE ON LAKE TROUT *Salvelinus namaycush*

Study Protocol Number: BFTC-99-CHLT-TAS

Experiment Number LKT-01: Lake trout fingerlings tested at 12°C

Experiment start date: July 17, 2001

Experiment end date: August 8, 2001

Study Director

Jim Bowker
U.S. Fish and Wildlife Service
Bozeman Fish Technology Center
National INAD Office

ORIGINAL

Investigator

Tina M. Miller
123 Onley Road
Salisbury University
Salisbury, MD 21804
410-341-4401

Co-Investigator

Daniel Carty
U.S. Fish and Wildlife Service
Bozeman Fish Technology Center
National INAD Office

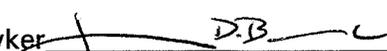
Testing Facility

U.S. Fish and Wildlife Service
Bozeman Fish Technology Center
National Investigational New Animal Drug Office
4050 Bridger Canyon Road
Bozeman, MT 59715
Phone: 406-587-9265
FAX: 406-582-0242

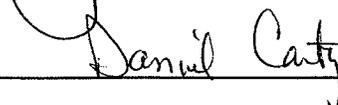
Author

Molly Poehling
U.S. Fish and Wildlife Service
Bozeman Fish Technology Center
National INAD Office

James D. Bowker

 D.B. 9/27/01

Daniel Carty

 Daniel Carty 9/27/01

Abstract

The United States Fish and Wildlife Service's National Investigational New Animal Drug Office designed and conducted a target animal safety study (Study Protocol Number BFTC-99-CHLT-TAS) to generate data needed to obtain U.S. Food and Drug Administration approval for the use of chloramine-T to control mortality in hatchery-reared salmonids diagnosed with bacterial gill disease or other external flavobacterial infections. Under the study protocol, a series of 10 separate experiments were conducted in which groups of healthy fry, fingerling, or juvenile rainbow trout *Oncorhynchus mykiss* were acclimated to a water temperature of $\approx 8^{\circ}\text{C}$ or $\approx 14^{\circ}\text{C}$ and then exposed to chloramine-T three times at concentrations ranging from 0 to 100 mg/L. An additional experiment, also conducted under this study protocol, was recently completed in which lake trout *Salvelinus namaycush* were exposed to chloramine-T to evaluate the target animal safety. This report presents results from this near-pivotal experiment (study number BFTC-99-CHLT-TAS-LKT-01), in which groups of fingerling lake trout were exposed three times to chloramine-T at concentrations of 0, 50, 100, 150, 200, or 300 mg/L (i.e., 0x, 2.5x, 5x, 7.5x, 10x, or 15x the proposed maximum therapeutic treatment concentration of 20 mg/L). The chloramine-T exposures were administered on three consecutive days. Exposures were administered as static-bath treatments that lasted 3 h each (i.e., three times longer than the proposed standard therapeutic treatment duration of 1 h). Each of the six exposure concentrations was tested in

duplicate; consequently, 12 test tanks of test fish were used in the experiment. Completely randomized design procedures were used to allocate fish to test tanks and assign exposure concentrations to test tanks. Although blinding techniques were employed to ensure that study participants involved in day-to-day data collection did not know which test tanks received which exposure concentrations of chloramine-T, on occasion, non-blinded study participants were involved in day-to-day data collection. The experiment lasted 23 d and consisted of a 6-d pre-exposure phase (experiment days 1 - 6), a 3-d exposure phase (experiment days 7 - 9), and a 14-d post-exposure phase (experiment days 10 - 23). In the test tanks, groups of fingerling lake trout ($n = 20$ fish per test tank) were acclimated to $\approx 12^{\circ}\text{C}$ water during experiment days 1 - 6 and then exposed to chloramine-T on experiment days 7, 8, and 9. Mortality that occurred during the exposure and post-exposure phases of the experiment was the primary response variable, and "total mortality" for each test tank was calculated by adding the number of dead fish removed from the test tank to the number of fish missing and unaccounted for when test fish were counted out of the test tank on the last day of the experiment. Mean total mortality in the 0-, 50-, 100-, 150-, 200-, and 300-mg/L chloramine-T exposure groups was 0.0, 0.0, 2.9, 8.8, 79.4, and 100.0%, respectively. However, no statistically significant difference (P - (estimated) = 0.123; P - (exact) = 0.067) existed in mean total mortality among the 100-, 150-, and 200-mg/L exposure groups. A non-statistical comparison of the mortality data suggested that mean total mortality was "low" in the 0-, 50-, and 100-mg/L exposure groups,

"moderate" in the 150-mg/L exposure group, and "high" in the 200- and 300-mg/L exposure group. Of the total number of mortalities recorded (65 dead fish + 0 fish missing and unaccounted for = 65 mortalities) 57 (87.7%) were fish that died during or within approximately 20 h of the end of the first chloramine-T exposure period. Results from this experiment (1) demonstrate that the proposed maximum therapeutic treatment concentration of 20-mg/L chloramine-T, when administered as a static-bath treatment on three consecutive days, is safe for use on fingerling lake trout being reared at a water temperature of $\approx 12^{\circ}\text{C}$; (2) indicate that the margin of safety for the consecutive-day, static-bath therapeutic treatment of fingerling lake trout being reared at $\approx 12^{\circ}\text{C}$ is greater than 100 mg/L, but less than 150 mg/L chloramine-T; and (3) suggest that fingerling lake trout are most susceptible to the toxic effects of relatively high concentrations (≥ 150 mg/L) of chloramine-T the first time they are exposed to it.