

# EFFECT OF ACUTE EXPOSURE TO CHLORINE, COPPER SULFATE, AND HEAT ON SURVIVAL OF NEW ZEALAND MUD SNAILS

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## ABSTRACT

The New Zealand mud snail (*Potamopyrgus antipodarum*) is a recent invader to aquatic systems in North America. The biology, ecology, and contemporary distribution of New Zealand mud snails in Europe and Australia suggest that the snail will spread rapidly in North America. Because the species can seal the opening of the shell with its operculum and survive out of water in a moist environment for long periods, improperly cleaned fishing and other gear may facilitate their dispersal. A proposed solution is to provide cleaning stations for sterilization of equipment at public access sites along snail-infested waters. We assessed the acute effectiveness of several commonly used biocides on New Zealand mud snails. Lethal exposure tests were conducted over short durations using chlorine (Cl) and copper sulfate ( $\text{CuSO}_4$ ) solutions, as well as heated water. Mortality of snails exposed to Cl at levels ranging from 500 to 3000 mg/L at all exposure durations rarely exceeded 30 percent. We observed similar results for dilute  $\text{CuSO}_4$ . Effectiveness increased with concentrations of  $\text{CuSO}_4$  with mortality generally exceeding 60 percent for 100 and 1000 mg/L at all durations. Exposure to hot water at temperatures of 45 °C for 60 sec or 50 °C for 15 sec killed most of the snails, which suggested this treatment offered the best option for sterilizing field equipment of those tested.

**Key words:** acute toxicity, chlorine, copper sulfate, heat, New Zealand mud snail, *Potamopyrgus antipodarum*

## INTRODUCTION

The New Zealand mud snail (*Potamopyrgus antipodarum*) (Gray 1843) is an exotic species recently introduced to North America (Zaranko et al. 1997). The history of its spread in Europe and Australia, where it was introduced in the 19th century and is now found across these continents (Bondesen and Kaiser 1949, Ponder 1988), suggests that the mud snail potentially could spread widely in North America. The mud snail was first found in North America in 1987 in the Snake River of eastern Idaho where it now occupies at least 640 km of river (Bowler 1991, Zaranko et al. 1997). Since then it has been

found in streams of the Greater Yellowstone Ecosystem, the Great Lakes, the mouth of the Columbia River, the Owens River in California, the Colorado River in the Grand Canyon, and several other streams in western North America (Zaranko et al. 1997, Gangloff 1998, D. L. Gustafson, Montana State University, personal communication).

Curtailing the spread of mud snails is important because invasive populations without natural predators or control mechanisms are often very successful and may ultimately negatively affect native invertebrate fauna and their food resources. Invasive populations of mud snails often increase rapidly because they are ovoviparous (Winterbourn 1970), parthenogenetic (Wallace 1992), and can tolerate a broad range of environmental

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conditions (Jacobsen and Forbes 1997). In the United States and Europe, mud snail densities have been reported to exceed 100,000 individuals/m<sup>2</sup> (Dorgelo 1987, Gangloff et al. 1999). Mud snails now are the numerically dominant invertebrates in the Middle Snake River, Idaho, and in several streams in the Upper Madison River in Yellowstone National Park (Bowler 1991, Gangloff et al. 1999, Richards et al. 2001, Vanderloop and Hall 2001). However, direct evidence of the effect of mud snails on native macroinvertebrate assemblages is rare. In a spring creek of the Madison River drainage in fall, periphyton (the algal food resource of mud snails and many macroinvertebrates) biomass and density, and biomass of macroinvertebrate functional groups, e.g., grazers and gatherers, were lower where mud snail abundance was moderate (~30,000/m<sup>2</sup>) than where mud snail abundance was low (~0-100/m<sup>2</sup>) (Cada and Kerans 2002). On the other hand, Schreiber et al. (2002) showed that the mud snail facilitated colonization of native fauna in an Australian stream. Clearly, the effects of invasive mud snails on native macroinvertebrates are complex.

The mud snail is an excellent colonist. Fish and floating aquatic vegetation have been implicated as possible within-system dispersal vectors (Haynes et al. 1985, Ribi 1986). Long-range and between-system dispersal of mud snails are facilitated by their ability to use an operculum to seal the shell opening (Burch 1989), and tolerate significant periods out of water (25 days) in moist and shaded environments (Winterbourn 1970). Moreover, the mud snail's small size, i.e., typically <10 mm, makes it difficult to detect in vegetation, gravel, or other debris thought to be likely mediums of transport. Thus, fishing and outdoor equipment, e.g., neoprene waders and wetsuits, wading boots, aquatic sandals, canoes, kayaks, bait buckets, and tackle boxes, if improperly cleaned and dried, can possibly facilitate dispersal of this exotic snail.

Providing stations for anglers and boaters to sterilize potentially contaminated

equipment has been proposed as a possible way to minimize human-caused risk of mud snail dispersal beyond its present range. Sterilization methods such as chlorine solution (Cl), copper sulfate solution (CuSO<sub>4</sub>), and hot water have been proposed. To be useful a molluscicide needs to be 1) effective with short duration application, 2) cost-effective, and 3) environmentally benign. Chlorine solutions are inexpensive, become inert quickly in the environment, and are widely used to minimize transmission of water-born fish pathogens (Warren 1991). Copper sulfate is a widely used algicide but is also toxic to fish and invertebrates (Watson and Yanong 1989). Dr. Robert McMahon (Department of Biology, Texas A&M University) suggested that hot water (~ 50 °C) would effectively kill snails and is the least environmentally harmful option.

Our objective was to evaluate how well three recommended treatments—chlorine (Cl) and copper sulfate (CuSO<sub>4</sub>) solutions and high water temperature—killed mud snails. We designed trials to simulate concentrations and durations that might be realistically used at equipment sterilization sites, e.g., short duration and rapid application, in the field.

## MATERIALS AND METHODS

We collected mud snails used in these experiments from the Madison River near West Yellowstone and held them in aquaria at room temperature (~17 °C) at the Wild Trout Laboratory, Montana State University, Bozeman. These snails in the Greater Yellowstone Ecosystem are genetically identical to the Snake River population.

We exposed mud snails to multiple concentrations of chlorine and copper sulfate solutions and temperatures of water over a series of time intervals to determine the most effective treatment as measured by mortality. Clorox® (5.25% active ingredient) and de-chlorinated tap water were used to make a stock Cl solution that we diluted to five exposure concentrations of 250, 500, 1000, 2000, and 3000 mg/L (verified using a Hach® Chlorine Test Kit).

Snails were initially exposed to the three lower chlorine concentrations for 15, 30, and 60 sec. However, mortality was extremely low so we conducted a second series of tests using concentrations of 2000 and 3000 mg/L for 30, 60, and 90 sec. We made copper sulfate stock solution by dissolving 10 g of hydrated copper sulfate ( $\text{CuSO}_4 \times 5\text{H}_2\text{O}$ ) in 1 L of de-chlorinated tap water. The stock solution was then diluted to three concentrations (10, 100, or 1000 mg/L) and mud snails were exposed for three time intervals (30, 60, and 90 sec) at each concentration. Snails were also exposed to heated dechlorinated water at 40, 45 and 50 °C for 15, 30, or 60 sec. For each series of tests we included an appropriate control of either clean water or water at room temperature (~17 °C) (see Tables 1, 2, 3).

For each concentration and exposure interval, we conducted three replicates. A replicate consisted of five snails (in a few cases <5 snails were tested) that were placed in a strainer and then dipped into a beaker containing 1 L of the treatment solution. After exposure, snails were rinsed in plain de-chlorinated tap water and placed in a Petri dish with aquarium water. Because the treated mud snails held their opercula tightly closed for some time after exposure, survival was assessed 24 hrs after treatment. However, in some cases in which snails were exposed to copper sulfate, it was difficult to tell if the snails were dead at 24 hrs. A final determination of mortality was made at 72 hrs post exposure. We examined the snails in the Petri dish for movement by prodding with a probe or forceps. Dead snails exhibited no visible movement of the foot, head, tentacle, or operculum (in closed snails) after contact. We calculated mortality by combining all replicates for each treatment level ( $n = 15$  typically) and dividing the total deaths by the total number of mud snails in each treatment level.

## RESULTS

Chlorine treatments were moderately effective in killing mud snails (Table 1), and

increasing the concentration and duration of chlorine exposures had little effect. All mud snails survived when exposed to 250-500 mg/L Cl for 15, 30, and 60 sec. Further, the mortality rates for the 1000 mg/L exposure were 0, 6.6, and 26.6 percent at 15, 30, and 60 sec, respectively. At 2000 mg/L, we observed a mortality rate of 13.3 percent for all exposures (30, 60, and 90 sec) and at 3000 mg/L we obtained mortality rates of 13.3, 33.3 and 20 percent, respectively (Table 1).

Copper sulfate was more effective than chlorine and snail mortality rates increased with  $\text{CuSO}_4$  concentration (Table 2). Mortality increased from 0 percent in the control to 15, 73, and 93.4 percent after 90-sec exposures to 10, 100, and, 1000 mg/L respectively (Table 2).

Snails exposed to 40 °C water showed an increase in mortality rate with exposure time with a maximum of 28-percent mortality at 60 sec of exposure (Table 3). At water temperatures of 45 °C we attained 100-percent mortality after 60 sec of exposure. Water at 50 °C was lethal to snails at all exposure intervals (Table 3).

## DISCUSSION

New Zealand mud snails proved remarkably resistant to chlorine treatments. Based on the results of these trials we suspect that most equipment contaminated with snails and field treated with chlorine at the levels and time intervals tested could still transport viable individuals to other locations. Increasing chlorine concentrations could improve effectiveness but might become irritating to mucus membranes of humans using the treatment and harmful to sensitive materials in recreational or research equipment, i.e., membranes, scales, fabrics.

Copper sulfate was more effective than chlorine but did not achieve 100-percent mortality at the levels and time intervals tested. An additional risk is its toxicity to a wide variety of non-target organisms at or below the levels tested, making it an undesirable solution in environmentally sensitive areas (Pimentel 1971). Even at

**Table 1.** Acute chlorine toxicity data for New Zealand mud snails showing treatment concentration, exposure time, total number living and dead snails in three replicates (24-hr post exposure), range (lowest and highest number of dead snails in a replicate), and percent mortality.

Concentration mg/L	Exposure (sec)	Number of snails Total/Dead	Range of dead snails per treatment	Mean % Mortality
0 (control)	15	15/0	0	0
0 (control)	30	15/0	0	0
0 (control)	60	15/0	0	0
250	15	15/0	0	0
250	30	15/0	0	0
250	60	14/1	0-1	7.1
500	15	15/0	0	0
500	30	15/0	0	0
500	60	13/0	0	0
1000	15	15/0	0	0
1000	30	15/1	0-1	6.7
1000	60	15/4	1-2	26.7
2000	30	15/2	0-2	13.3
2000	60	15/2	0-2	13.3
2000	90	15/2	0-2	13.3
3000	30	15/1	0-1	6.7
3000	60	15/5	1-2	33.3
3000	90	16/3	0-2	18.8

<sup>1</sup>Occasionally the number of snails in a replicated differed slightly from five.

**Table 2.** Acute copper sulfate solution ( $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ ) toxicity data for New Zealand mud snails showing treatment concentration, exposure time, total number living and dead snails in three replicates (72-hr post exposure), range (lowest and highest number of dead snails in a replicate), and percent mortality.

Concentration mg/L	Exposure (sec)	Number of snails Total/Dead	Range of dead snails per treatment	Mean % Mortality
0 (control)	30	15/0	0	0
0 (control)	60	15/0	0	0
0 (control)	90	15/0	0	0
10	30	15/3	0-2	20.0
10	60	15/1	0-1	6.7
10	90	15/2	0-2	13.3
100	30	15/9	2-4	60.0
100	60	15/7	1-3	46.7
100	90	15/11	2-5	73.3
1000	30	15/12	2-5	80
1000	60	10/8	4	80
1000	90	15/14	4-5	93.3

<sup>1</sup>Occasionally the number of snails in a replicated differed slightly from five.

**Table 3.** Acute temperature exposure data for New Zealand mud snails showing treatment temperature, exposure time, total number living and dead snails in three replicates (24-hr post exposure), range (lowest and highest number of dead snails in a replicate), and percent mortality.

Temperature °C	Exposure (sec)	Number of snails Total/Dead	Range of dead snails per treatment	Mean % Mortality
17 (control)	15	15/0	0	0
17 (control)	30	15/0	0	0
17 (control)	60	15/0	0	0
40	15	14/0	0	0
40	30	14/2	0-2	14.3
40	60	14/5	0-3	35.7
45	15	15/3	0-3	20.0
45	30	15/7	0-4	46.7
45	60	15/15	5	100
50	15	15/15	5	100
50	30	15/15	5	100
50	60	15/15	5	100

<sup>1</sup> Occasionally the number of snails in a replicated differed slightly from five.

recommended rates of application for algae control (1-2 mg/L), it may be toxic to trout, especially in low alkalinity waters. However, its toxicity to fish generally decreases as water hardness increases (Gangstad 1986). In addition, copper sulfate is toxic to aquatic invertebrates; the 96-hr LC50 for pond snails is 0.39 mg/L at 20 °C (U.S. National Library of Medicine, Hazardous Substances Databank 1995). Whereas the toxicity information is interesting, we note that a 96-hr LC 50 is not directly comparable to mortality in acute toxicity tests with exposures of 90 sec.

Heated water appears to be the most simple and effective way of killing mud snails. Hot tap water, which is usually about 49 °C (120 °F), may be the least likely of the tested sterilizing agents to cause environmental harm and would be relatively harmless to most types of equipment. Results of this experiment suggest that simply washing gear in hot water can effectively kill snails. Cleaning stations similar to those used to control the spread of the zebra mussel could be established at fishing access areas and would undoubtedly help contain New Zealand mud snails and other nuisance

aquatic species. But ultimately, educating fishery workers, anglers, and boaters will be critical to stopping the spread of this and other exotic species. Anyone using or working in infested waters can and should take steps to disinfect their equipment.

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