Upper Clark Fork River Fisheries Monitoring Study: 2013 Annual Report

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DEQ Contract No.

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Cover Photograph: First day of 2014 cage checks at Silver Bow site located in Warm Springs, Montana in April 2014.

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Introduction

Mines and mills were operated in the Upper Clark Fork River Basin for roughly 100 years, from the 1880s to 1980s. These mining and mill operations were relatively large-scale operations that produced large amounts of product by processing large quantities of material. As a result of these operations, material wastes were discharged, released, or deposited directly into the Clark Fork River System (MultiTech 1987). It was later discovered that these wastes consisted of high concentrations of metals and which were hazardous to the environment (Copeland 2002).

Fish are known to accumulate metals through water borne exposure and their diet and these accumulations are known as tissue metal burdens. Erickson et al. (2008) stated that fish absorb metals through their gills and skin through water, as well as through ingestion. Marr et al. (1995a, b) exposed trout to water with similar metal concentrations to those found in the Clark Fork River, and fish were found to accumulate metals. Studies by Farag et al. (1994) and Louma et al. (2008) showed metals accumulation occurred when fish were fed invertebrates from the Clark Fork River.

Upper Clark River Basin fish have been adversely affected by metals pollution from historic mining and mill operations. Studies have shown both acute and chronic effects including reduced survival and growth (Marr et al. 1995a, b), and cell damage (Woodward et al. 1995a). Factors such as water discharge and water temperature may also exacerbate these effects as a greater number of fish mortalities have been found during high spring discharges and also during the descending limb of the hydrograph as temperatures rise (Mayfield and McMahon 2010, 2011). In addition, tolerance to metals pollution varies by species, with more tolerant species having increased metallothionein (a metals binding protein that protects against metals toxicity). Farag et al. (1995) found that brown trout (*Salmo trutta*) from the Clark Fork River possessed elevated levels of metallothionein and were more tolerant to metals pollution than rainbow trout (*Oncorhynchus mykiss*), and this has in turn created low trout species diversity and brown trout dominance throughout much of the Upper Clark Fork River Basin. Fish also avoid areas with elevated metal levels even if they are able to tolerate them, meaning that not all available habitat is used when metals are high (Woodward et al. 1995b; Louma et al. 2008). Metals pollution may be the main factor affecting fish population numbers in the Upper Clark Fork River Basin.

Previous studies have utilized fish cages and have shown variation in fish mortality based on metals, space, and time. A fish cage study was conducted by Phillips and Spoon (1990) in the Clark Fork River from 1986 to 1989 and another fish cage study was conducted by Richards et al. (2013) in the Upper Clark Fork River in 2011 and 2012. Philips and Spoon (1990) found that mortality was high at Beavertail, consistently low at Clinton below Rock Creek with mortality varying both in space and time. Metals pollution was implicated as contributing to poor fish survival in the Clark Fork River. Richards et al. (2013) found similar results with high mortality at upstream sites (Galen and Warm Springs), and with mortality elsewhere varying in time and space, unrelated to metals pollution, except for one site (Turah) below Rock Creek which displayed high mortality. Richards et al. (2013) suggest that mine wastes and potentially elevated pH in the Upper Clark Fork River Basin had negative effects on fish populations within the Upper Clark Fork River Basin.

Although research has suggested negative effects from mine wastes in the Upper Clark Fork River Basin, more research is needed to further address this issue. Metals concentrations (including copper) continue to exceed acute and chronic aquatic toxicity criteria in the Upper

Clark Fork River (PBSJ 2010) and other conditions have changed. Remediation work on Silver Bow Creek, and possibly other factors, may currently affect mortality rates at sites in the Upper Clark Fork River. Assessment of potential confounding factors that may mask the response of trout populations to metals cleanup and cause high mortality in the mainstem is warranted. A more current and complete understanding of mortality rates would aid in planning and monitoring Clark Fork River remediation efforts.

In 2013, Montana Fish, Wildlife and Parks (FWP) received funding from Montana Department of Environmental Quality (MTDEQ) to complete a similar caged fish study to that completed by Richards et al. (2013) in addition to collecting fish population information on the mainstem Clark Fork River. The focus of this project is to assess the effects of current levels of metals contamination in the Upper Clark Fork River on the mortality of fishes along with trout population monitoring as pre-remediation monitoring data for future assessment of remediation efforts. Another objective of the study was to assess impacts ongoing in-stream remediation efforts are having on Clark Fork River fish populations.

Objectives

1. Determine mortality rates of age 0 brown trout in the upper Clark Fork River at nine sites (from Warm Springs Ponds to Turah, Montana), two control streams, and one handling control site.

2. Identify water quality factors affecting the mortality rates of young trout, including non-metal stressors.

3. In terms of metals tissue burdens, draw comparisons between: 1. control and treatment sites, 2. sites upstream and downstream of the construction area in Warm Springs, Montana, 3. upper river, middle river, and lower river sites, and 4. live versus dead fish.

4. Explore possible trends between data collected in previous years and the current year.

5. Provide information to remediation project managers that will aid in the planning and implementation of cleanup efforts.

Methods

Trout Population Monitoring

Population estimates were calculated for the following sample reaches of the Upper Clark Fork River in 2013: Bearmouth, Flint Creek Mouth, Phosphate, Williams-Tavenner, Below Sager Lane, and pH Shack. Field methods were conducted in the same manner as Lindstrom (2011). Trout populations were monitored with electrofishing completed in April of 2013. Fish were collected with the use of a 14 ft long aluminum drift boat with a mounted electrofishing unit and two front boom anodes. The system was powered by a 5,000-watt generator and current was modified with a Coffelt VVP-15 or Smith-Root VVP-15B rectifying unit. Estimates were made using two mark passes and two recapture passes of which recapture passes were completed roughly one week later. All captured trout were identified to species, weighed (grams) and measured (mm), and given a small fin clip unique to the sampling section and day. Resulting data was analyzed by sample reach and species and was summarized by the population estimate (if available; standardized to number of fish per mile), 95% confidence interval with upper and lower bounds, capture efficiencies, number of fish handled, mean length, length range, and percent of species composition. Population estimates were generated using the Chapman modification (Chapman 1951) of the Petersen method provided in Montana Fish, Wildlife and Park's Fisheries Information System database. Estimates and capture efficiencies were calculated for trout species that had a minimum of 4 marked fish that were recaptured (B. Liermann, Montana, Fish, Wildlife, and Parks, personal communication, 2014). Due to low numbers and/or poor capture efficiency of smaller size classes, only estimates for fish greater than 175 mm (~7 in) in length were reported.

Estimates from previous years (2008-2012) were included in this report for completeness as they are part of the long-term dataset required for this study. A Chapman modification of the Petersen method, as described above, was used to generate estimates in the Fisheries Information System for data from 2011, 2012, two sample reaches from 2010 (Bearmouth and Flint Creek Mouth), and two sample reaches from 2009 (Bearmouth and Flint Creek Mouth). Estimates from 2008, remaining sample reaches in 2009 (pH Shack, Below Sager Lane, Williams-Tavenner, and Phosphate), and remaining sample reaches in 2010 (pH Shack, Below Sager Lane, Williams-Tavenner, and Phosphate) were generated using a Chapman estimator for the Peterson method provided in Montana Fish, Wildlife and Park's Fisheries Analysis Plus (FA+) software package, and are presented here as originally reported in Lindstrom 2011. Both programs produce identical population estimates, but confidence intervals around the estimates are calculated differently, with FA+ assuming sample data is normally distributed and the Fisheries Information System assuming sample data is binomially distributed (see Ogle 2010 for details).

Cage Construction

Thirty-six wooden cages constructed for a previous study were used for this 2013 study. The cages resembled those used by FWP on the Middle Clark Fork River, but were 34% larger to accommodate the brown trout used in this study (Figure 1). The internal volume of the cages was 0.75 ft^3 (actual volume of water available). Knotless nylon seine material (1/16 inch bar mesh) was used for the netting on the sides and bottom of the cages. Cages were also fitted with floats to provide buoyancy.

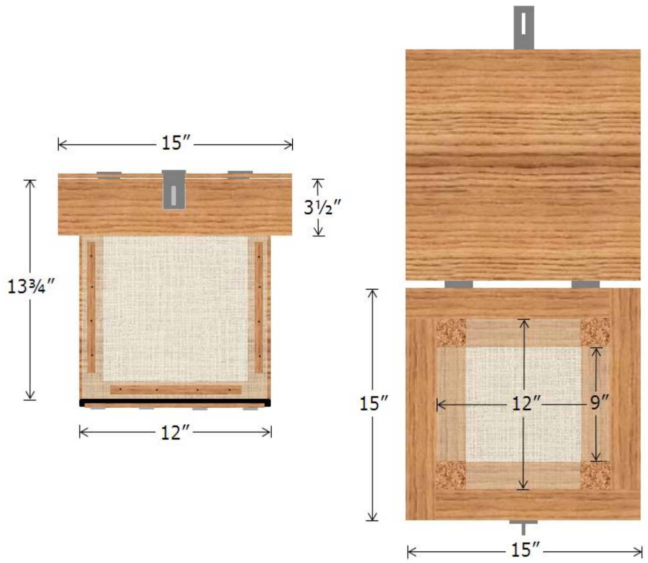

Figure 1. Dimensions of the cages constructed for the study.

Study Sites

Cages were deployed at twelve locations in the Upper Clark Fork River Drainage in early April 2013 (Figure 2). Nine treatment sites were located at the following locations:

1) Mill Willow Bypass at Warm Springs, Montana (Mill Willow)
2) Pond 2 Outlet at Warm Springs, Montana (Pond 2)
3) Silver Bow Creek at Warm Springs, Montana (Silver Bow)
4) Warm Springs Creek at Warm Springs, Montana (Warm Springs)
5) Galen, Montana – River Left (Galen Left)
6) Galen, Montana – River Right (Galen Right)
7) Deer Lodge, Montana (Deer Lodge)
8) Upstream of the Little Blackfoot River (U/S Lil Black)
9) Turah, Montana (Turah)

Two control sites were located on tributaries:

10) Lower Little Blackfoot River (Lil Black)
11) Lower Flint Creek (Flint)

One handling control site was located in a spring-fed channel:

12) Clinton, Montana (Clinton Spring)

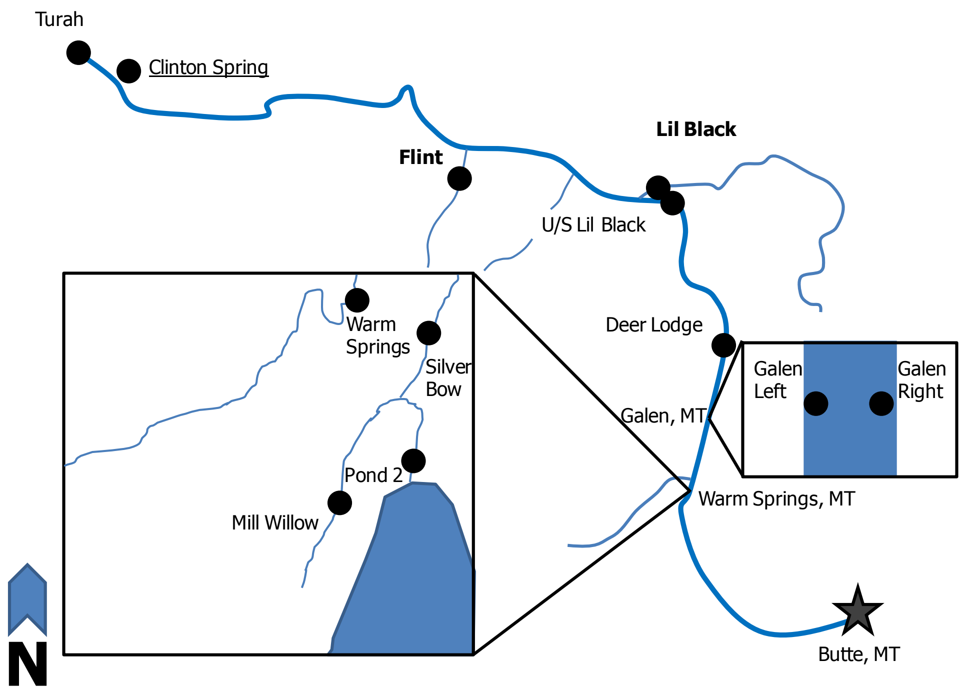

Figure 2. Distribution of the eleven study sites in the Upper Clark Fork River drainage. Control sites are shown in bold and the handling control is underlined.

All sites except the Mill Willow Bypass in Warm Springs, Montana, the Pond 2 Outlet in Warm Springs, Montana, and the spring channel near Clinton, Montana (handling control), were located near U.S. Geological Survey (USGS) gauging stations equipped to measure discharge four times per hour. The handling control served as a reference to adjust mortality rates if cage checks (e.g., cleaning and relocating) or stress from initial fish delivery to the cages negatively impacted survival, independent of water quality.

Cage Deployment

Exact locations of the cages were dependent on the availability of low velocity habitats with access to refuge during periods of high runoff. Cages were positioned in velocities less than 0.75 ft/s. Three cages were deployed at each site. Two served as treatment cages (i.e., one replicate) and the third held fish for replacement of individuals in the treatment cages. The study began with 25 brown trout per cage and these densities were maintained in the treatment cages as long as possible by replacing them with individuals from the replacement cage. Cages were secured with sections of reinforcing bar (rebar) driven into the substrate, as well as sash weights and tether lines (Figure 3). The sash weights provided additional anchoring during rising water levels, and tether ropes insured the cages were not completely lost should a flood event occur.

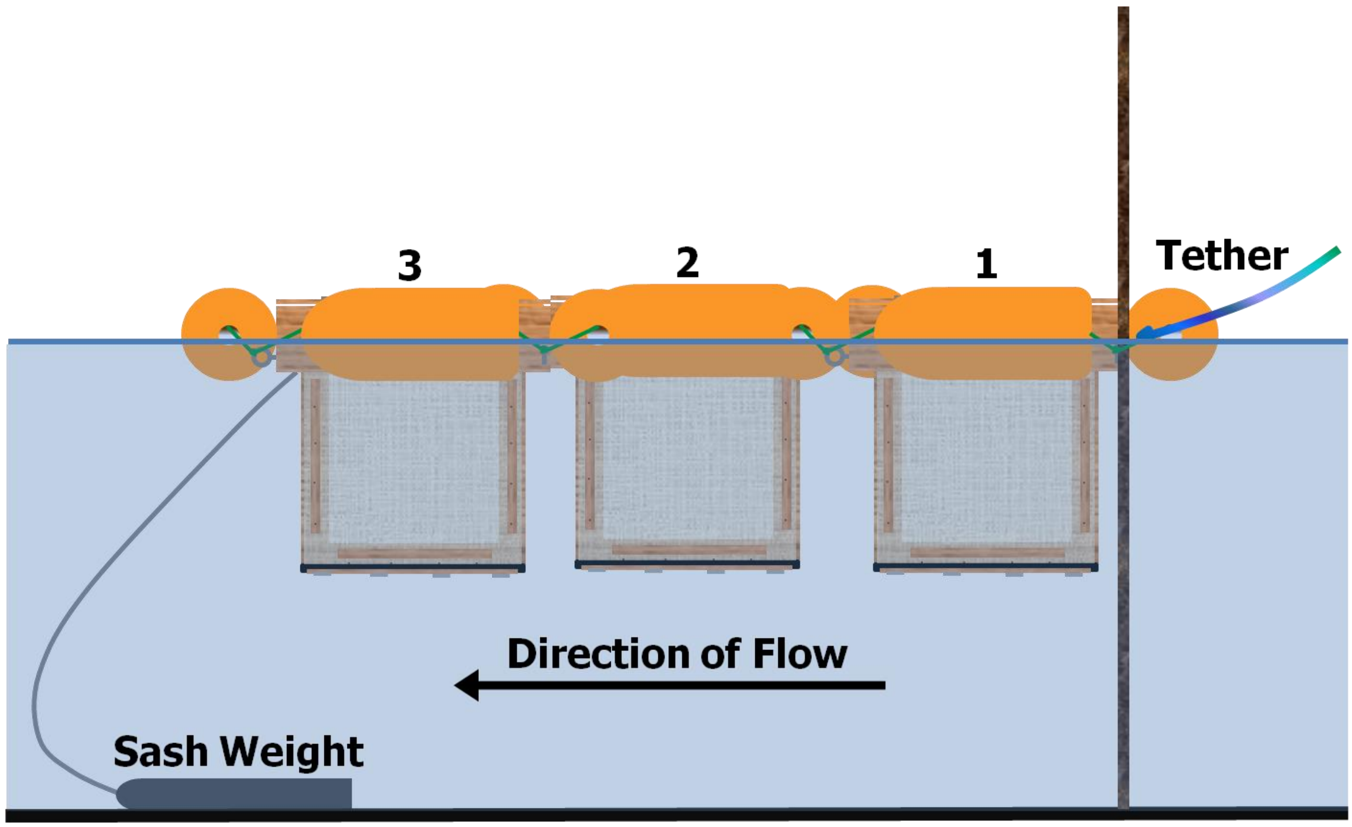

Figure 3. Representation of cage deployment (arrangement of cages differed by site, and cages often drifted together).

Brown trout were selected for this study given their dominance in the Upper Clark Fork River. Due to low densities of young trout in the upper river, study specimens were obtained from a state hatchery. The fingerlings ranged from 51-85 mm and were feed-trained on pellet feed upon delivery.

In late March approximately 900 fingerling brown trout were obtained from Big Springs Hatchery in Lewistown, Montana. The trout were transported from the hatchery to Helena, Montana in a hauling truck and from Helena to the sites in an aerated cooler. At each site trout were anesthetized with clove oil, measured to total length, and divided into one of the three cages. Prior to being anesthetized, fish were acclimated to the water temperature at each site with the addition of onsite water. In 2013 at the first site stocked, the hatchery water was 6.7 °C and water temperatures at the sites varied from 4.4 °C to 12.2 °C. Mean length of trout stocked in cages was 71.3 mm (SD = 5.8 mm) in 2013.

Mortality Monitoring

Beginning the first week of April each year, trout mortality was monitored twice per week. At each visit the trout in each cage were fed one tablespoon of pellet feed. During the first three months trout were fed 1.0 mm sinking feed (Silver Cup Extruded Salmon). The remaining months, trout were fed slightly larger No. 3 sinking feed (Silver Cup Crumbled

Salmon/Trout). Cages were repositioned to seams and eddies with reduced velocities as discharge varied at each site. Velocities around the cages were measured periodically to ensure velocities did not exceed 0.75 ft/s. The exterior of the cages were brushed clean as needed to provide for exchange of water between the cage and the site. At each visit mortalities were removed from the treatment cages (cages 1 and 2) and were replaced with individuals from the replacement cage (cage 3). All mortalities were measured to total length in millimeters and archived in a freezer at the Region 2 FWP headquarters.

Statistical analyses of trout survival at the nine treatment sites consisted of chi-square comparisons between observed and expected survival and mortality in 2013 with $\alpha = 0.05$. Yates's correction for continuity was applied to all chi-square tests as the degrees of freedom for each test was one (Yates 1934). Expected mortality for each year was determined by using the mean mortality at the two control sites located in the Little Blackfoot River and Flint Creek (mean mortality = 20.5). Expected survival at each site was set to 50 as this was the number of live fish maintained in cages one and two combined. Mortalities during the first week of April each year and mortalities after the end of July were not included in the analyses because any mortalities occurring at the treatment sites during this period may have been due to fish being held in cages based on previous data from the Clinton Spring handling control.

Growth

A subsample of all specimens placed in cages was taken for each site at the beginning of each field season and those fish were measured to the nearest millimeter and a one-way analysis of variance (ANOVA) was used to determine if initial lengths of fish placed in treatment cages differed among sites. Initial lengths did not differ significantly among sites in 2013 (ANOVA: $F_{11, 348} = 0.9021$, $P = 0.5385$); however, a subsample of 30 fish (15 surviving fish randomly selected from both cages 1 and 2) per site was measured at the completion of the field season to remain consistent with analyses in previous years. If there were less than 30 surviving fish at site (Pond 2 and Flint) at the end of the field season all surviving fish were sampled. Subsamples were also used to evaluate growth by calculating change in mean total length by site.

Tissue Metals Burdens

Tissue metals burdens in fish can be used as a measure of exposure and can be correlated to histopathological effects (Hansen et al. 2004). Upon completion of the study, all mortalities from the treatment cages from April through August each year were submitted to the Montana Department of Health and Human Services Environmental Laboratory in Helena for analysis of tissue metals burdens. Mortalities from each site during each month of each year were submitted as individual samples for tissue analysis. In addition, 14 fish surviving at the conclusion of the field season at the end of August at each site were randomly selected and were submitted as individual samples. Due to close proximity, similar results in other analyses, and budget constraints, Galen Left fish were submitted for tissue metals burdens analysis and Galen Right fish were not submitted for tissue metals burdens analysis.

Samples were blended to a powder to ensure homogeneity, and then the samples were weighed, dried, and reweighed to determine moisture content. The dried samples were then crushed and dissolved with nitric acid, diluted with deionized water, and analyzed for copper and zinc with inductively coupled plasma optical emission spectrometry (ICP-OES) using the U.S.

Environmental Protection Agency (USEPA) Method 200.7 (USEPA 2001). The samples were also analyzed with inductively coupled plasma mass spectrometry (ICP-MS) for contaminants that have a lower detection limit including arsenic, cadmium, lead and selenium using USEPA Method 200.8 (USEPA 1999). All results were reported as µg/g dry weight.

Analyses of tissue metals burdens data was completed both graphically and statistically. Graphical comparisons were made between tissue metals burdens (copper and zinc) and each of the following variables: month, mortalities, site location, and site type (control vs. treatment, upstream construction vs. downstream construction, upper river vs. middle river vs. lower river, and live fish vs. dead fish). Fish from the hatchery were sacrificed prior to stocking fish cages in order to determine baseline tissue metals burdens. Comparisons to the previous years of this study (2011 and 2012) were included for discussion. In addition, statistically significant differences between copper tissue burden and fate (live versus dead) were tested for using a logistic regression. Zinc tissue burdens were not used for statistical analyses because zinc tissue burden results were always above minimum effect thresholds. Lastly, copper tissue metals burdens of dead fish from treatment sites in the main area of concern (Pond 2 downstream to U/S Lil Black) were analyzed graphically against the maximum water temperature (°C) experienced within the previous 5 days prior to death to try and discern causes of mortality. Four quadrants were created based on the minimum effect threshold of copper (Colt et al. 1979) and the upper critical temperature threshold for brown trout (Elliot 1994). Quadrant one (Q1) contained fish mortalities due to water temperature, quadrant two (Q2) contained fish mortalities due to a combination of water temperature and copper tissue metals burdens, quadrant three (Q3) contained fish mortalities due to copper tissue metals burdens, and quadrant 4 (Q4) contained fish mortalities due to unknown causes. Equal sample sizes were assumed (12 for each quadrant due to total sample size of 46) and differences from this were considered evidence for a relationship.

Water Contaminants

Water samples were collected three times at each of the twelve sites (known as Main Events), with the exception of the Little Blackfoot River site from which samples were only collected twice due to obstructed access. Collections roughly coincided with low-elevation runoff (ascending limb of the hydrograph), peak runoff, and the descending limb of the hydrograph (Figure 4). Grab samples were collected for the caged fish study using the techniques outlined by the MTDEQ Field Procedures Manual for Water Quality Assessment Monitoring (MTDEQ 2012a). Samples were collected on May 28, June 14, and July 18 in 2013. All samples were delivered to Energy Laboratories Inc. in Helena, Montana and were analyzed for dissolved and total recoverable metals including copper, arsenic, lead, cadmium, and zinc, as well as calcium, magnesium, and total ammonia nitrogen (NH_3-N). Atkins collected additional water data under a contract for MTDEQ during the quarterly monitoring of the Clark Fork River Operating Unit (CFROU) (Figure 4). The Atkins report detailing the 2013 data was not yet available at the time of preparation of this manuscript. In addition, site-specific water samples were collected during suspected rain events (known as Rain Events) throughout the 2013 field season (Figure 4).

Performance standards have been identified for contaminants in the upper Clark Fork River (USEPA 2004; Atkins 2012) and are defined as the more stringent of the freshwater aquatic life standards (ALS) published by the MTDEQ (2012b). Because the chronic ALS is the most stringent and since this study focuses on chronic effects, the chronic ALS was used to evaluate contaminant data. Freshwater ALS are a function of total water hardness and are evaluated on the basis of total recoverable metals concentrations (Atkins 2012; MTDEQ 2012b). Chronic freshwater ALS values were obtained from the table of standards for Montana waters or calculated using the hardness relationships described by MTDEQ (2012b). The chronic ALS values were calculated as:

$$\text{Chronic} = exp.\{mc[ln(hardness)]+bc\}$$

where *mc* and *bc* = values listed by MTDEQ (2012b). Chronic ALS compliance ratios were calculated by dividing the measured contaminant values by the calculated chronic ALS values, and were plotted for each site and sampling period. Compliance ratio values <1 indicate contaminant levels below the chronic ALS, while values >1 indicate contaminant levels above the chronic ALS.

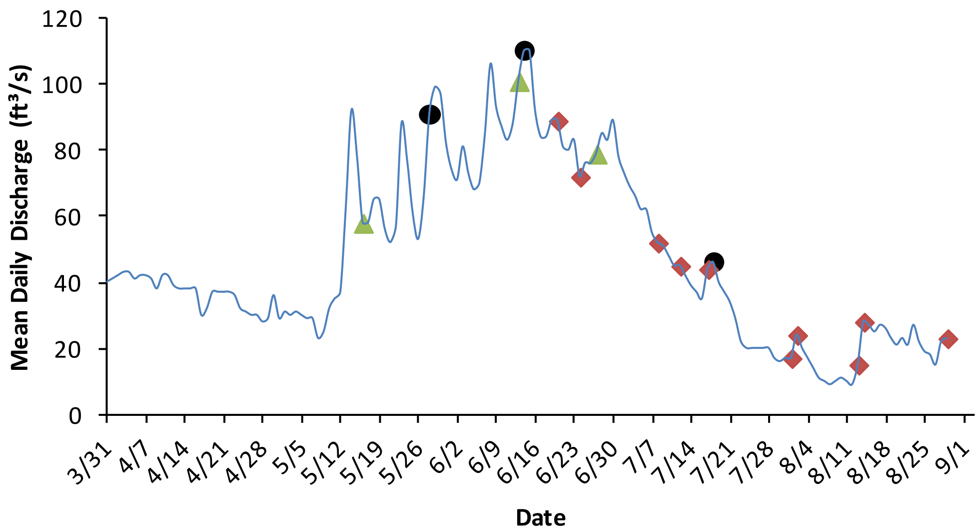

Figure 4. Clark Fork River hydrograph for 2013 at the Warm Springs gauging station in Warm Springs, Montana. Dots represent FWP, triangles represent Atkins, and diamonds represent rain event water collection dates.

Discharge and Water Temperature

Discharge data presented in this report were obtained from USGS gauge stations recording measurements four times per hour. Estimates of mean daily discharge were downloaded from the USGS National Water Information System: Web Interface. It is important to note that not all estimates presented in this report have been reviewed and approved for publication. All gaps in datasets during the 2013 field season were the result of equipment malfunctions. No station existed at the Mill Willow, Pond 2, or Clinton Spring sites.

Maximum daily water temperatures were obtained for each site with water temperature data loggers (HOBO ® U22 Pro v2). Loggers were attached to the rebar securing the cages in the channel and the units were most often set 6-12 inches above the substrate. Due to logger malfunctions temperature data may contain gaps at some sites; when available, this data was substituted with data from the appropriate USGS station.

Water Quality

Water quality parameters were recorded in the Clark Fork River at five sites in 2013 with continuously recording multiparameter water quality probes (Hydrolab ® MS5). Cross referencing of data collected with continuously recording multiparameter water quality probes was achieved by sampling intermittently at the nine treatment and three control sites using a handheld multiprobe (YSI ® 556 MPS). Hydrolab and YSI probes were calibrated at regular intervals during each field season. Probes were deployed at Mill Willow, Pond 2, Silver Bow, Galen, and U/S Lil Black in 2013. Water quality parameters recorded include temperature, pH, specific conductivity, and luminescent dissolved oxygen (LDO) at all sites, with the addition of total ammonia ($NH_4 + NH_3$) at Pond 2, Silver Bow, and at Galen. Toxicity of total ammonia is dependent on other water parameters including water temperature and pH (Emerson et al. 1975; MTDEQ 2012b). The increased toxicity is due to the conversion of the generally inert form (NH_4) to the highly toxic form (NH_3) through the process of de-ionization (Barton 1996). Acute freshwater ALS for total ammonia based on hourly average measurements and chronic ALS based on a 30 day average were calculated based on equations published by MTDEQ (2012). The acute ALS values were calculated as:

$$\text{Acute} = (0.275/(1+10^{7.204-pH})) + (39.0/(1+10^{7.204-pH}))$$

and the chronic ALS were calculated as:

$$\text{Chronic} = ((0.0577/(1+10^{7.688-pH})) + (2.487/(1+10^{pH4-7.688}))) \text{ x MIN}(2.85,1.45 \text{ x } 10^{0.028 \text{ x } (25-T)})$$

where T = temperature (°C). Thirty day averages for comparison to chronic ALS values were calculated around peaks in total ammonia measurements.

Results

Trout Population Estimates

Trout population estimates were calculated for brown trout from 2011-2013 for every stream reach and 2009-2010 for the Bearmouth and Flint Creek Mouth sections (Tables 1-6). 2008-2010 population estimates in the Below Sager Lane, Williams-Tavenner, and Phosphate electrofishing sections from Lindstrom (2011) are included an appendix to this report (Appendix I). Figure 5 displays all brown trout population estimates by sample reach from 2008-2013, including population estimates already reported in Lindstrom (2011). The pH shack Section consistently had the highest brown trout population estimates, with a population estimate of 1878 fish/mile in 2013. Conversely, the Bearmouth Section consistently had the lowest brown trout population estimates, with a population estimate of 60 fish/mile in 2013. Flint Creek Mouth, Below Sager Lane, Williams-Tavenner, and Phosphate sections had 2013 brown trout population estimates of 197, 462, 532, and 506 fish/mile respectively. All catch statistics and other trout species population estimates are displayed in Tables 1-6.

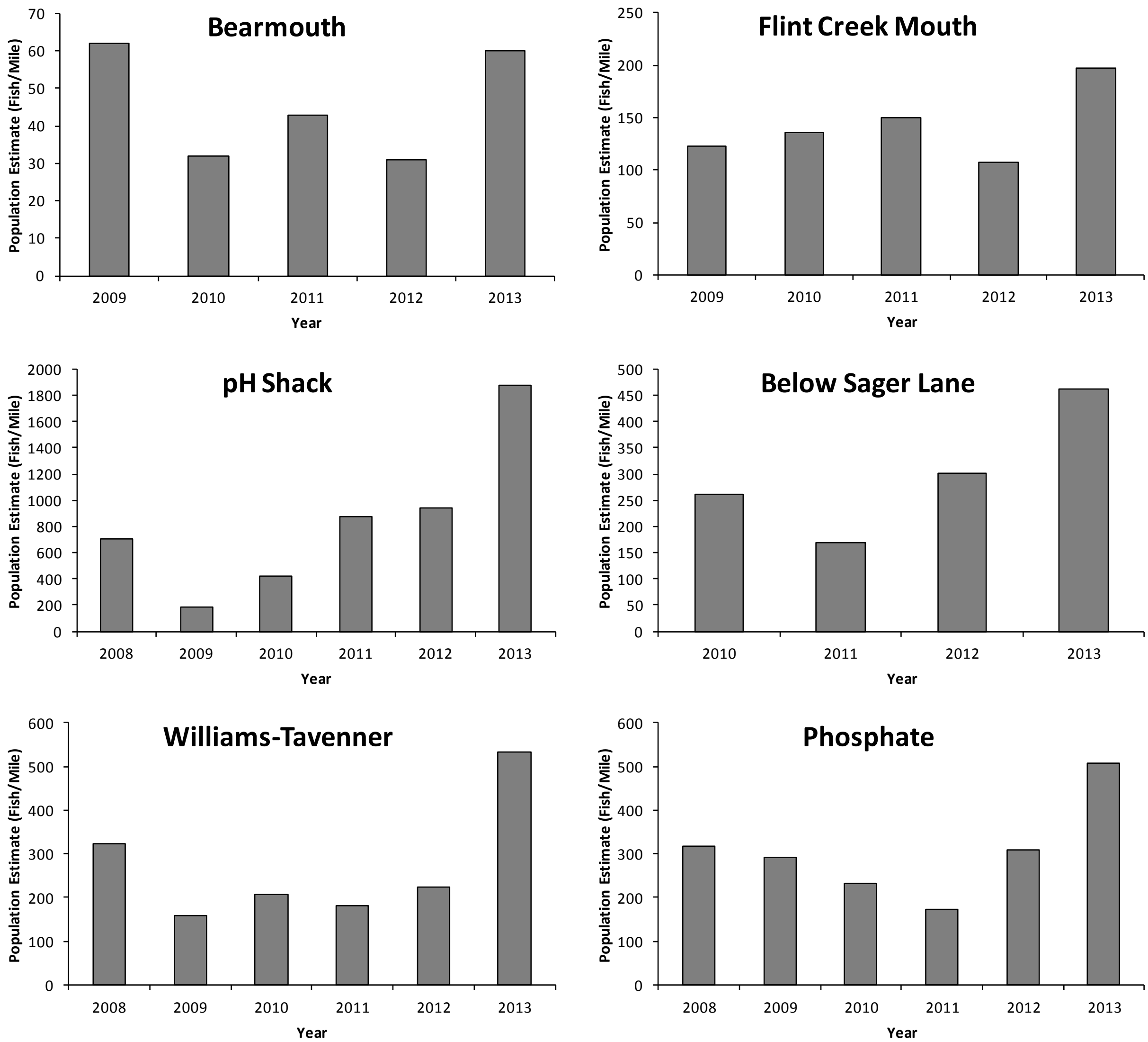

Figure 5. Clark Fork River brown trout population estimates from 2008-2013 by sample reach. Please note that x-axis and y-axis values are not the same for every sample reach.

Table 1. Electrofishing data collected on the Upper Clark Fork River at the Bearmouth Section from 2009-2013. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Numbers following the population estimate (in parentheses) represent the 95 % confidence interval. Cutt x Rbow represents a phenotypic hybrid between a cutthroat and rainbow trout.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2009*	Brown	62 (38,102)	13	134	358	119-528	69
	Cutthroat	7 (4,14)	27	26	314	152-410	13
2010	Brown	32 (23,49)	35	106	362	157-525	45
	Rainbow	-	-	13	345	242-442	6
	Cutthroat	6 (4,11)	42	27	308	100-400	12
	Bull	-	-	2	321	297-345	1
	Cutt x Rbow	-	-	8	371	320-458	3
2011	Brown	43 (30,65)	27	123	342	152-523	27
	Rainbow	7 (4,13)	38	28	342	152-479	6
	Cutthroat	13 (9,20)	38	54	309	182-414	12
	Bull	-	-	2	424	362-486	< 1
2012	Brown	31 (21,47)	29	95	326	177-502	21
	Rainbow	21 (14,34)	31	69	285	178-467	16
	Cutthroat	41 (30,59)	27	134	290	168-434	30
	Bull	-	-	2	266	260-272	< 1
2013	Brown	60 (43,87)	21	169	339	191-476	32
	Rainbow	19 (11,35)	24	49	344	230-455	9
	Cutthroat	45 (32,66)	27	134	321	175-426	26
	Bull	-	-	3	379	337-400	1

* In 2009, entire Upper Clark Fork River was sampled and as a result the Bearmouth Section is roughly a tenth of a mile longer than in other years.

Table 2. Electrofishing data collected on the Upper Clark Fork River at the Flint Creek Mouth Section from 2009-2013. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Numbers following the population estimate (in parentheses) represent the 95 % confidence interval. Cutt x Rbow represents a phenotypic hybrid between a cutthroat and rainbow trout. Brook x Bull represents a phenotypic hybrid between an eastern brook and bull trout.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2009*	Brown	123 (88,177)	18	273	369	97-550	95
2010	Brown	136 (105,181)	20	377	345	115-535	94
	Rainbow	-	-	4	389	326-421	1
	Cutthroat	-	-	16	284	227-355	4
	Cutt x Rbow	-	-	4	332	305-352	1
2011	Brown	150 (122,187)	25	481	311	110-509	89
	Rainbow	-	-	3	441	425-468	1
	Cutthroat	14 (8,24)	20	54	275	195-390	10
	Brook	-	-	1	287	-	< 1
	Brook x Bull	-	-	1	393	-	< 1
2012	Brown	107 (82,141)	19	334	293	124-515	87
	Rainbow	-	-	6	352	232-468	2
	Cutthroat	-	-	42	289	186-445	11
	Bull	-	-	2	374	373-375	1
2013	Brown	197 (161,245)	20	572	315	195-502	96
	Cutthroat	6 (3,11)	21	25	326	220-378	4
	Bull	-	-	1	273	-	< 1

* In 2009 entire Upper Clark Fork River was sampled and as a result the Flint Creek Mouth Section is roughly half a mile longer than in other years.

Table 3. Electrofishing data collected on the Upper Clark Fork River at the pH Shack Section from 2011-2013. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Numbers following the population estimate (in parentheses) represent the 95 % confidence interval. Cutt x Rbow represents a phenotypic hybrid between a cutthroat and rainbow trout.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2011	Brown	878 (531,1476)	13	265	311	89-498	98
	Rainbow	-	-	2	531	472-590	1
	Cutthroat	-	-	3	350	292-424	1
	Cutt x Rbow	-	-	1	423	-	< 1
2012	Brown	943 (686, 1322)	17	403	293	105-473	98
	Rainbow	-	-	7	369	256-540	2
	Cutthroat	-	-	2	306	292-319	< 1
	Cutt x Rbow	-	-	1	323	-	< 1
2013	Brown	1878 (1595,2223)	19	1056	296	156-630	98
	Rainbow	-	-	13	447	314-610	1
	Cutthroat	-	-	6	327	271-352	1
	Cutt x Rbow	-	-	1	282	-	< 1

Table 4. Electrofishing data collected on the Upper Clark Fork River at the Below Sager Lane Section from 2011-2013. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Numbers following the population estimate (in parentheses) represent the 95 % confidence interval.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# Fish Handled	Mean Length (mm)	Length Range (mm)	Species Compositio n (%)
2011	Brown	170 (119,251)	20	205	313	103-495	98
	Cutthroat	-	-	4	335	280-392	2
	Brook	-	-	1	202	-	< 1
2012	Brown	302 (232,397)	17	533	240	90-595	96
	Cutthroat	-	-	6	314	277-347	1
	Brook	-	-	15	216	134-273	3
2013	Brown	462 (390,553)	25	655	308	139-497	99
	Rainbow	-	-	1	324	-	< 1
	Cutthroat	-	-	2	323	308-337	< 1
	Brook	-	-	6	245	194-275	1

Table 5. Electrofishing data collected on the Upper Clark Fork River at the Williams-Tavenner Section from 2011-2013. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Numbers following the population estimate (in parentheses) represent the 95 % confidence interval.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# Fish Handled	Mean Length (mm)	Length Range (mm)	Species Compositi on (%)
2011	Brown	182 (140,244)	26	247	311	108-514	90
	Cutthroat	15 (9,28)	29	24	275	213-328	9
	Brook	-	-	2	203	196-209	1
2012	Brown	224 (180,285)	29	351	266	109-497	88
	Cutthroat	23 (18,34)	46	48	301	170-373	12
	Brook	-	-	1	221	-	< 1
2013	Brown	532 (453,632)	26	636	317	129-507	93
	Cutthroat	33 (22,56)	32	47	295	193-383	7
	Brook	-	-	1	320	-	< 1

Table 6. Electrofishing data collected on the Upper Clark Fork River at the Phosphate Section from 2011-2013. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Numbers following the population estimate (in parentheses) represent the 95 % confidence interval. Cutt x Rbow represents a phenotypic hybrid between a cutthroat and rainbow trout.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2011	Brown	171 (140,215)	41	239	300	104-474	97
	Cutthroat	-	-	7	294	207-378	3
	Cutt x Rbow	-	-	1	367	-	< 1
2012	Brown	308 (231,419)	21	282	270	111-464	92
	Rainbow	-	-	2	423	215-630	1
	Cutthroat	-	-	23	267	187-364	7
	Brook	-	-	1	305	-	< 1
2013	Brown	506 (393,664)	22	387	301	120-461	96
	Cutthroat	-	-	14	305	255-357	3
	Cutt x Rbow	-	-	1	389	-	< 1

Caged Fish Mortality, Discharge, and Water Temperature

Table 7 contains the results of chi-square comparisons between observed and expected survival and mortality, and Figures 6-17 depict total mortalities between cages one and two combined, maximum daily water temperatures, and mean daily discharges at cage sites in 2013. The solid red horizontal line in each figure represents the upper critical temperature threshold for brown trout of 19.0 °C (Elliot 1994). At temperatures above this critical threshold, significant disturbances to normal brown trout behavior may occur, including cessation of feeding and growth and ultimately death (Elliot 1994). The dashed red horizontal line in each figure represents the upper incipient lethal temperature for brown trout of 24.7 °C, above which thermal stress is lethal with mortality a function of exposure time (Elliot 1994).

In 2013, over half of the cage sites displayed bimodal mortality with some mortality occurring early in the study season on the ascending limb of the hydrograph, and some mortality on the descending limb as water temperatures approached and/or exceeded 19 °C (Figures 6-17). Sites deviating from this trend include Silver Bow, Galen Left, Galen Right, Deer Lodge, and Turah, all of which exhibited either consistent mortality throughout the field season (Silverbow, Galen Left, Galen Right, and Deer Lodge) or mortality after the peak of the hydrograph as water temperatures approached or exceeded 19.0 °C (Turah). Mean daily discharge, maximum daily water temperatures, and timing of mortalities at each site are outlined below in order from upstream to downstream.

<u>Mill Willow</u>

There is no discharge data available for Mill Willow in 2013 because there is not a USGS station present at this site. Peak maximum daily water temperature at Mill Willow in 2013 was 26.0 °C on July 26 (Figure 6). Maximum daily water temperature in 2013 exceeded 19.0 °C for 63 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for 6 days (Figure 6). Mill Willow experienced significantly lower mortality than expected (Table 7), with bimodal mortality occurring early in the study season and later in the study season (Figure 6).

<u>Pond 2</u>

There is no discharge data available for Pond 2 in 2013 because there is not a USGS station present at this site. Peak maximum daily water temperature at Pond 2 in 2013 was 24.9 °C on July 17 (Figure 7). Maximum daily water temperature in 2013 exceeded 19.0 °C for 69 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for two days (Figure 7). Pond 2 experienced significantly higher mortality than expected (Table 7), with bimodal mortality occurring early in the study season and later in the study season (Figure 7).

<u>Silver Bow</u>

Peak mean daily discharge at Silver Bow in 2013 was 192 ft^3/s on May 30. In 2013 peak maximum daily water temperature at Silver Bow was 25.6 °C on July 17 (Figure 8). Maximum daily water temperature in 2013 exceeded 19.0 °C for 66 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for four days (Figure 8). Silver Bow experienced significantly lower mortality than expected (Table 7), with consistent mortality throughout the field season (Figure 8).

Warm Springs

Peak mean daily discharge at Warm Springs in 2013 was 110 ft^3/s on June 14 and 15. In 2013 peak maximum daily water temperature at Warm Springs was 22.1 °C on July 26 (Figure 9). Maximum daily water temperature in 2013 exceeded 19.0 °C for 34 days and the upper incipient lethal temperature for brown trout of 24.7 °C was never exceeded (Figure 9). Warm Springs experienced significantly lower mortality than expected (Table 7), with bimodal mortality occurring early in the study season on the ascending limb of the hydrograph, as well as on the descending limb as water temperatures exceeded 19.0 °C (Figure 9).

Galen Left

Peak mean daily discharge at Galen Left in 2013 was 293 ft^3/s on May 30. In 2013 peak maximum daily water temperature at Galen Left was 23.8 °C on July 1 (Figure 10). Maximum daily water temperature in 2013 exceeded 19.0 °C for 62 days and the upper incipient lethal temperature for brown trout of 24.7 °C was never exceeded (Figure 10). Galen Left experienced significantly lower mortality than expected (Table 7), with consistent mortality throughout the field season (Figure 10).

Galen Right

Peak mean daily discharge at Galen Right in 2013 was 293 ft^3/s on May 30. In 2013 peak maximum daily water temperature at Galen Right was 23.7 °C on July 26 (Figure 11). Maximum daily water temperature in 2013 exceeded 19.0 °C for 61 days and the upper incipient lethal temperature for brown trout of 24.7 °C was never exceeded (Figure 11). Galen Right displayed consistent mortality throughout the field season (Figure 11).

Deer Lodge

Peak mean daily discharge at Deer Lodge in 2013 was 349 ft^3/s on May 30. In 2013 peak maximum daily water temperature at Deer Lodge was 25.8 °C on July 1 (Figure 12). Maximum daily water temperature in 2013 exceeded 19.0 °C for 70 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for four days (Figure 12). Deer Lodge experienced significantly lower mortality than expected (Table 7), with consistent mortality throughout the field season (Figure 12).

U/S Lil Black

Peak mean daily discharge at U/S Lil Black in 2013 was 398 ft^3/s on May 30 and 31. In 2013 peak maximum daily water temperature at U/S Lil Black was 27.0 °C on July 1 (Figure 13). Maximum daily water temperature in 2013 exceeded 19.0 °C for 79 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for eight days (Figure 13). U/S Lil Black experienced significantly lower mortality than expected (Table 7), with bimodal mortality occurring early in the study season on the ascending limb of the hydrograph, as well as on the descending limb as water temperatures exceeded 19.0 °C (Figure 13).

Lil Black (Control)

Peak mean daily discharge at Lil Black in 2013 was 497 ft^3/s on June 4. In 2013 peak maximum daily water temperature at Lil Black was 25.0 °C on July 26 (Figure 14). Maximum daily water temperature in 2013 exceeded 19.0 °C for 61 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for one day (Figure 14). Lil Black displayed bimodal mortality with some mortality occurring early in the study season on the ascending limb of the hydrograph, and some mortality on the descending limb as water temperatures exceeded 19 °C (Figure 14).

Flint (Control)

Peak mean daily discharge at Flint in 2013 was 169 ft^3/s on June 3. In 2013 peak maximum daily water temperature at Flint was 25.0 °C on July 1 (Figure 15). Maximum daily water temperature in 2013 exceeded 19.0 °C for 72 days and exceeded the upper incipient lethal temperature for brown trout of 24.7 °C for two days (Figure 15). Flint displayed bimodal mortality with some mortality occurring early in the study season on the ascending limb of the hydrograph, and some mortality on the descending limb as water temperatures approached or exceeded 19 °C (Figure 15). Extremely low flow conditions were also experienced at the Flint Creek site in 2013 with flows as low as 8 cfs experienced in May and 15 cfs in late July/early August.

Clinton Spring (Handling Control)

There is no discharge data available for Clinton Spring in 2013 because there is not a USGS station present at this site. In 2013 peak maximum daily water temperature at Clinton Spring was 16.5 °C on August 24 (Figure 16). Maximum daily water temperature never exceeded 19.0 °C and the upper incipient lethal temperature for brown trout of 24.7 °C was never exceeded in 2013 (Figure 16). Clinton Spring displayed bimodal mortality with some mortality occurring early in the study season and some mortality later in the study season (Figure 16).

Turah

Peak mean daily discharge at Turah in 2013 was 2,764 ft^3/s on May 14. In 2013 peak maximum daily water temperature at Turah was 24.0 °C on July 2 (Figure 17). Maximum daily water temperature in 2013 exceeded 19.0 °C for 62 days and the upper incipient lethal temperature for brown trout of 24.7 °C was never exceeded (Figure 17). Turah experienced significantly lower mortality than expected (Table 7), with mortality occurring after the peak of the hydrograph as water temperatures approached and exceeded 19.0 °C (Figure 17).

Table 7. Results of χ^2 tests between expected and observed survival and mortality for 2013, with Yates's correction for continuity applied; df = 1 for all tests. Red asterisks denote significantly higher than expected mortality at α = 0.05; black asterisks denote significantly lower than expected mortality.

	Year
Site	2013
Mill Willow	*P* = 0.004*
Pond 2	*P* = 0.0127*
Silver Bow	*P* = 0.0483*
Warm Springs	*P* = 0.0483*
Galen Left	*P* = 0.0483*
Galen Right	*P* = 0.1221
Deer Lodge	*P* = 0.004*
Upstream of Little Blackfoot	*P* = 0.0008*
Turah	*P* = 0.0003*

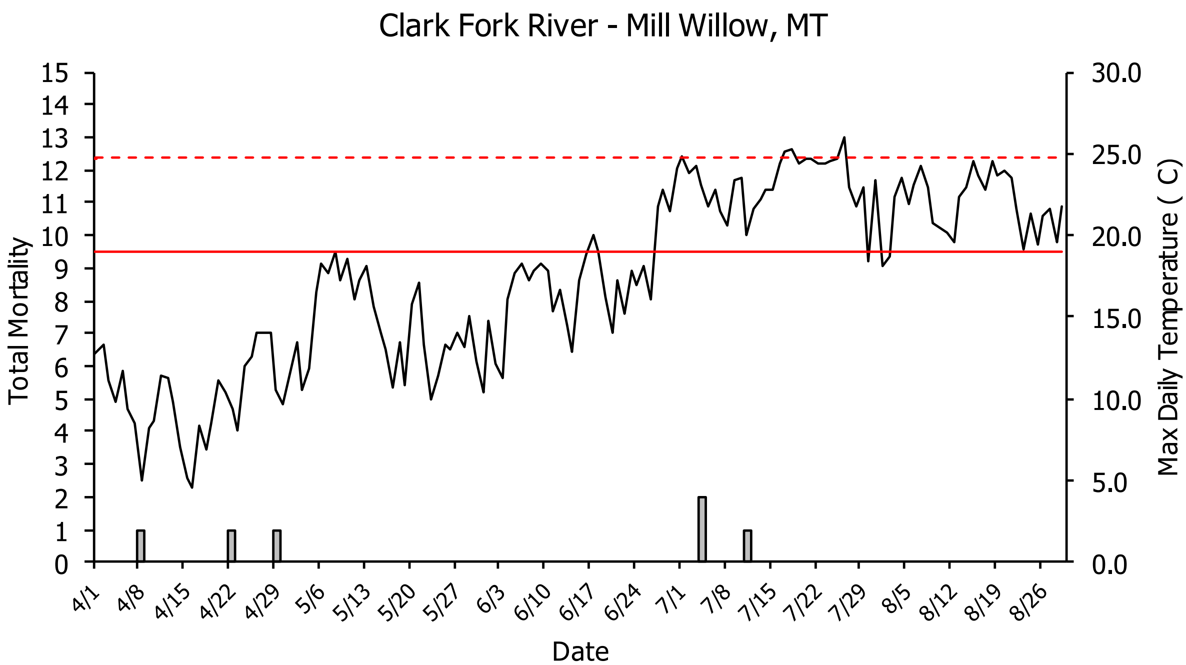

Figure 6. Total mortalities between cages one and two combined (gray bars) and maximum daily water temperature (black line) for 2013 in the Clark Fork River at the Mill Willow site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

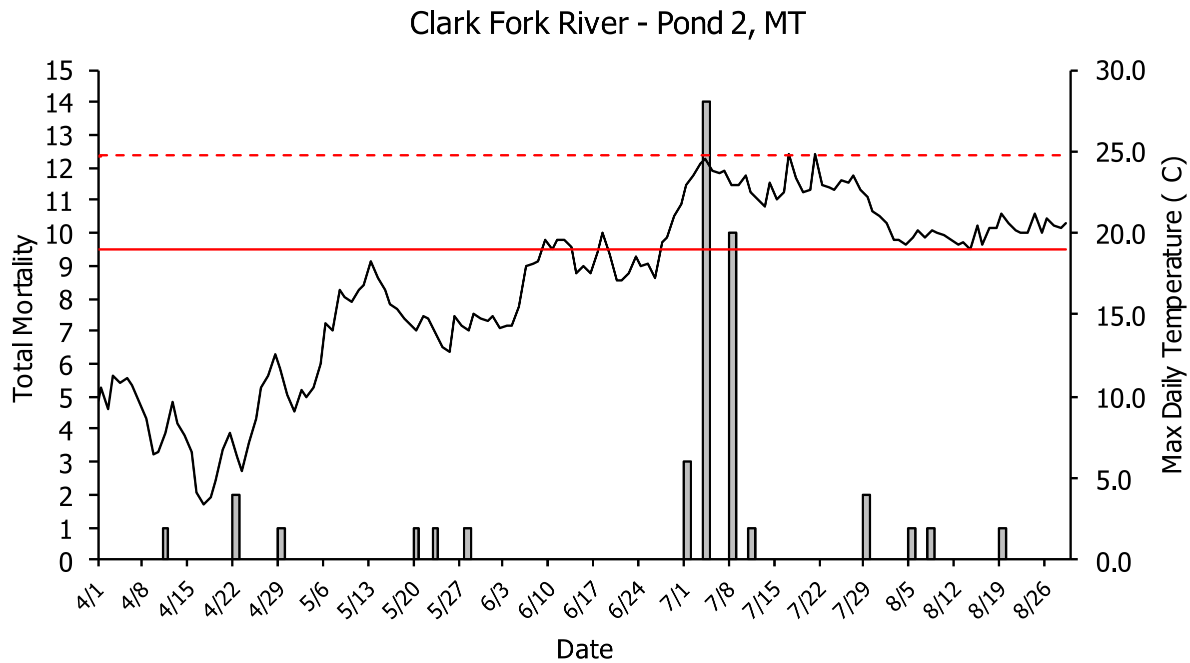

Figure 7. Total mortalities between cages one and two combined (gray bars) and maximum daily water temperature (black line) for 2013 in the Clark Fork River at the Pond 2 site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

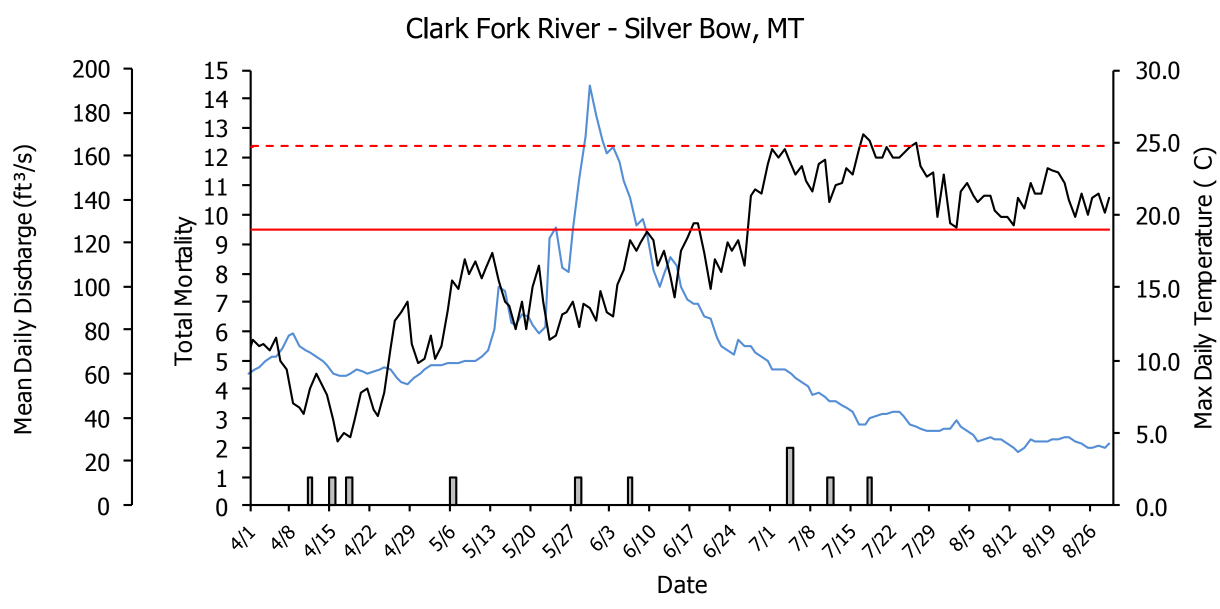

Figure 8. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River at the Silver Bow site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

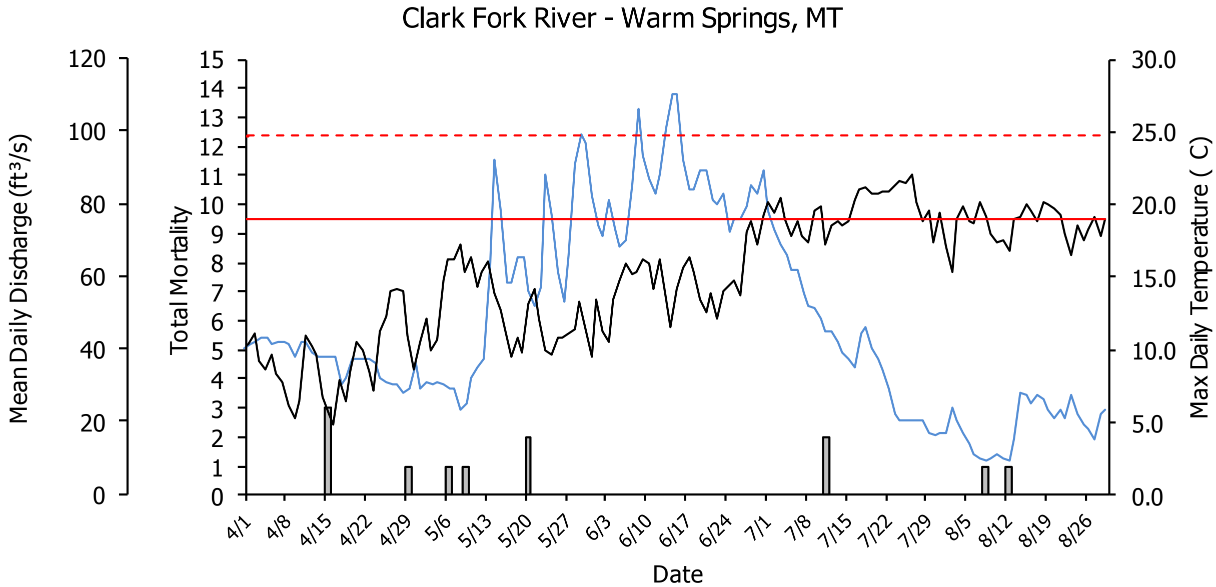

Figure 9. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River at the Warm Springs site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

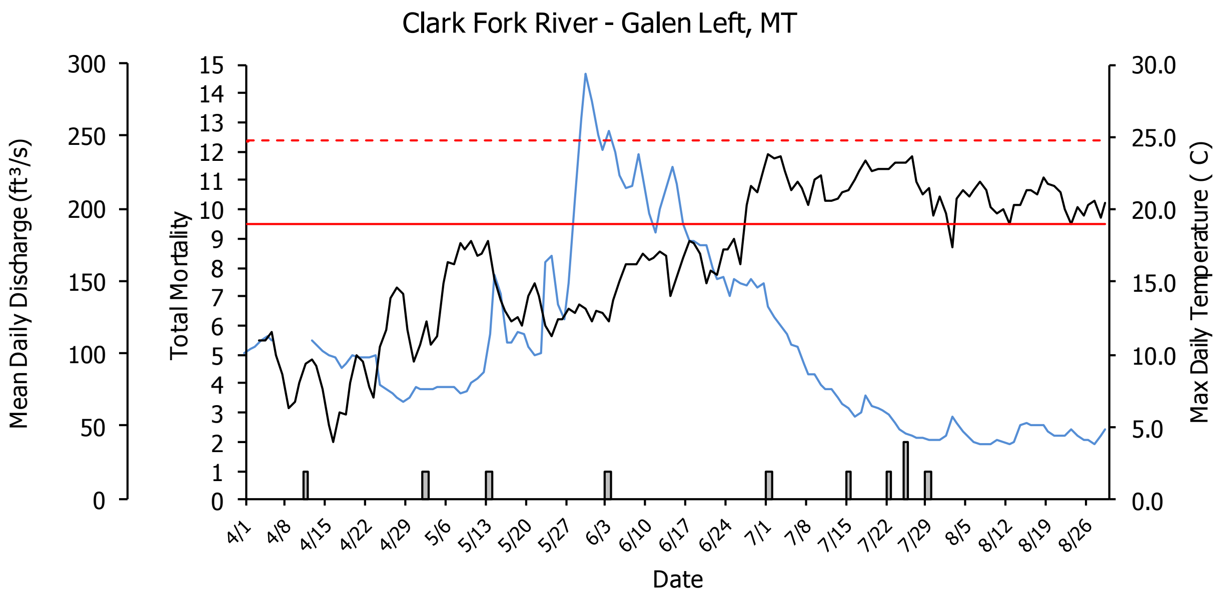

Figure 10. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River at the Galen Left site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

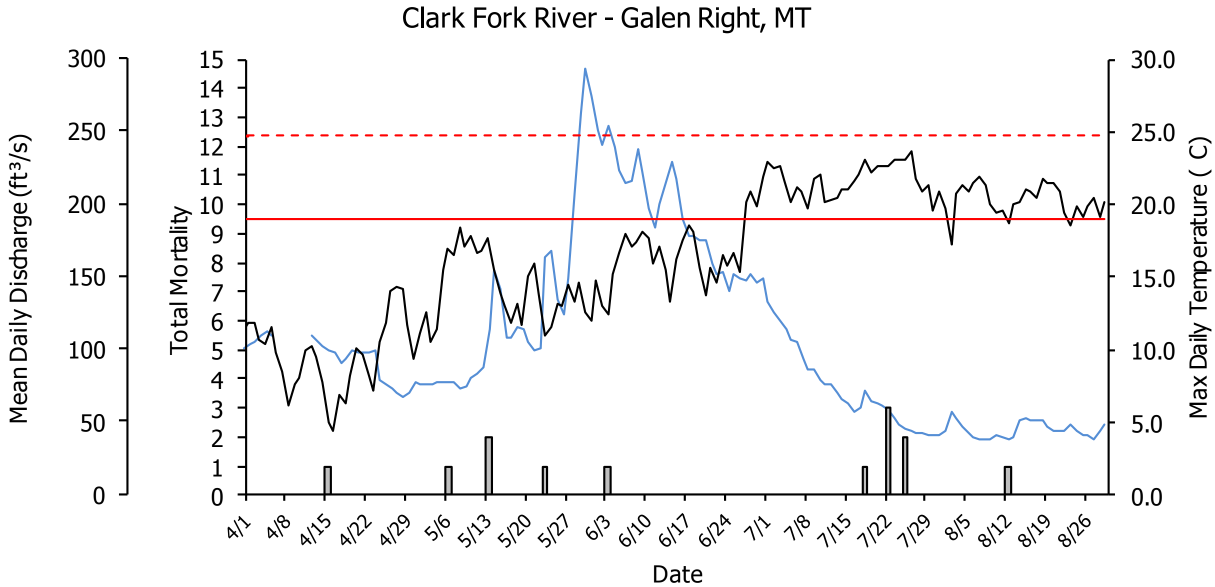

Figure 11. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River at the Galen Right site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

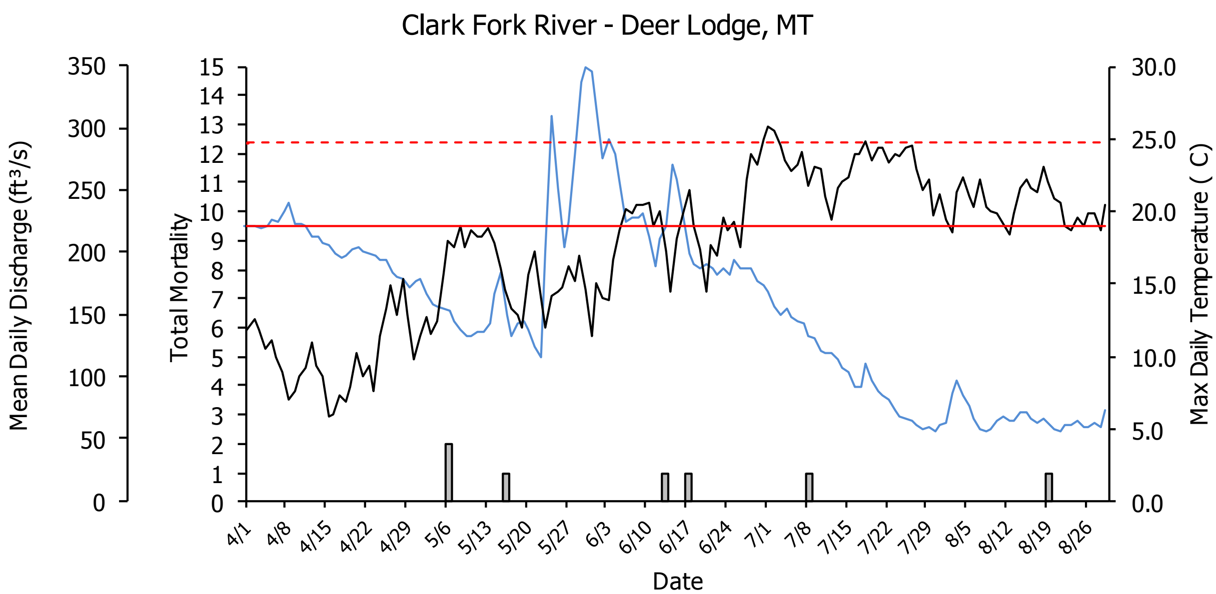

Figure 12. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River at the Deer Lodge site. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

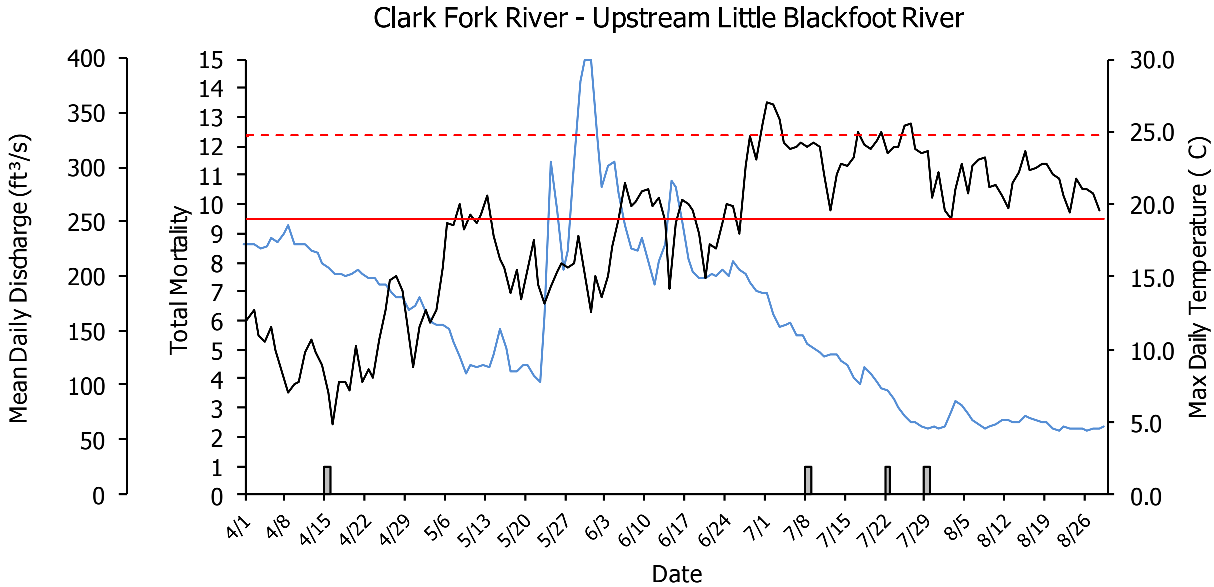

Figure 13. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River at the site upstream of the Little Blackfoot River. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

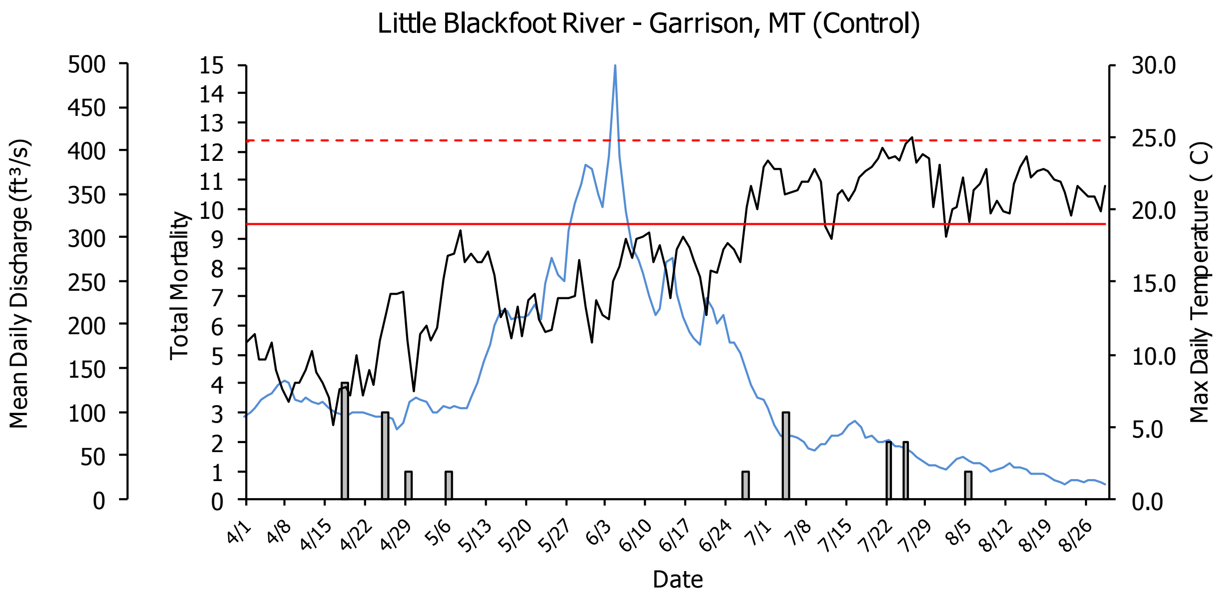

Figure 14. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 at the control site in Little Blackfoot River. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

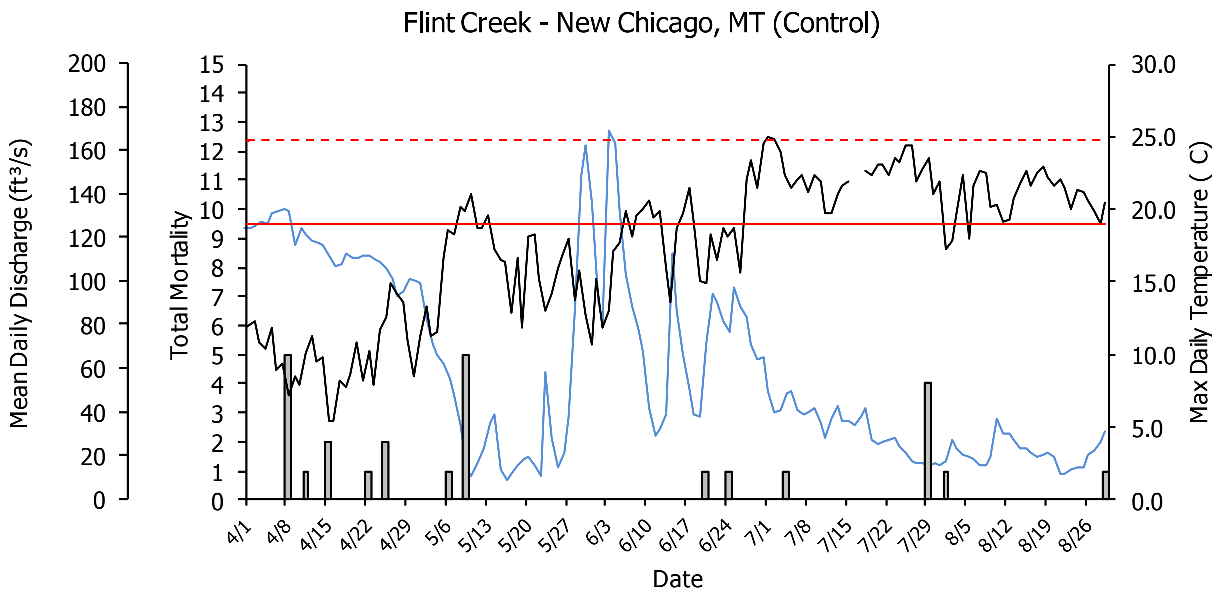

Figure 15. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 at the control site in Flint Creek. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

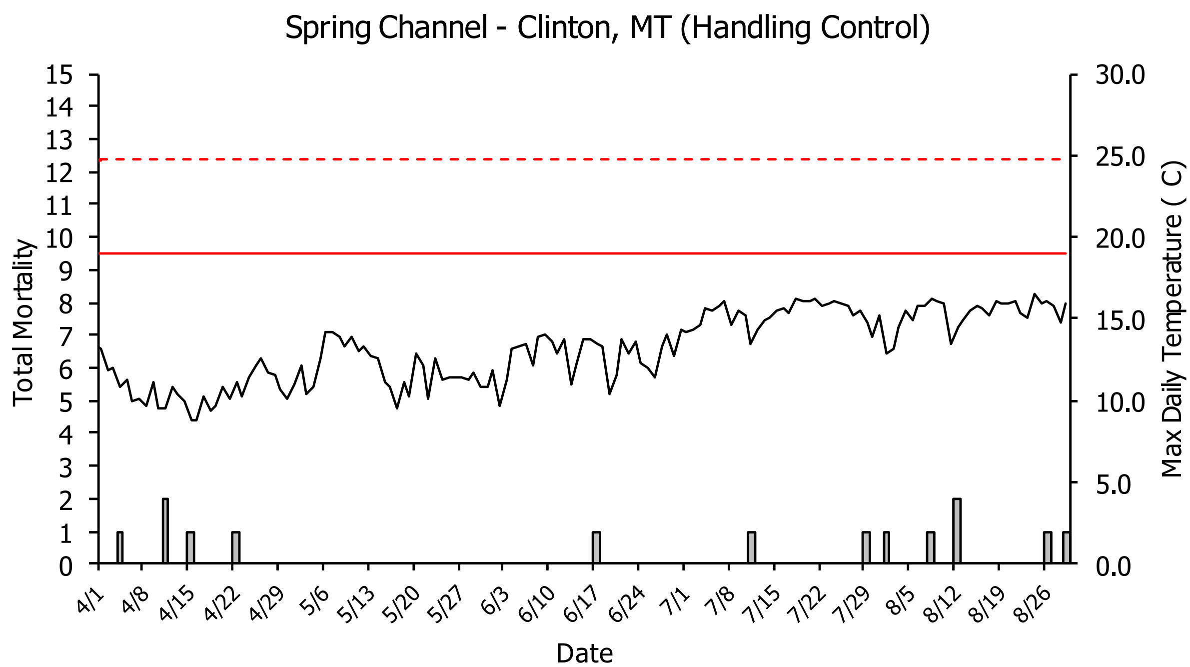

Figure 16. Total mortalities between cages one and two combined (gray bars) and maximum daily water temperature (black line) for 2013 at the control site in the spring channel near Clinton, Montana. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

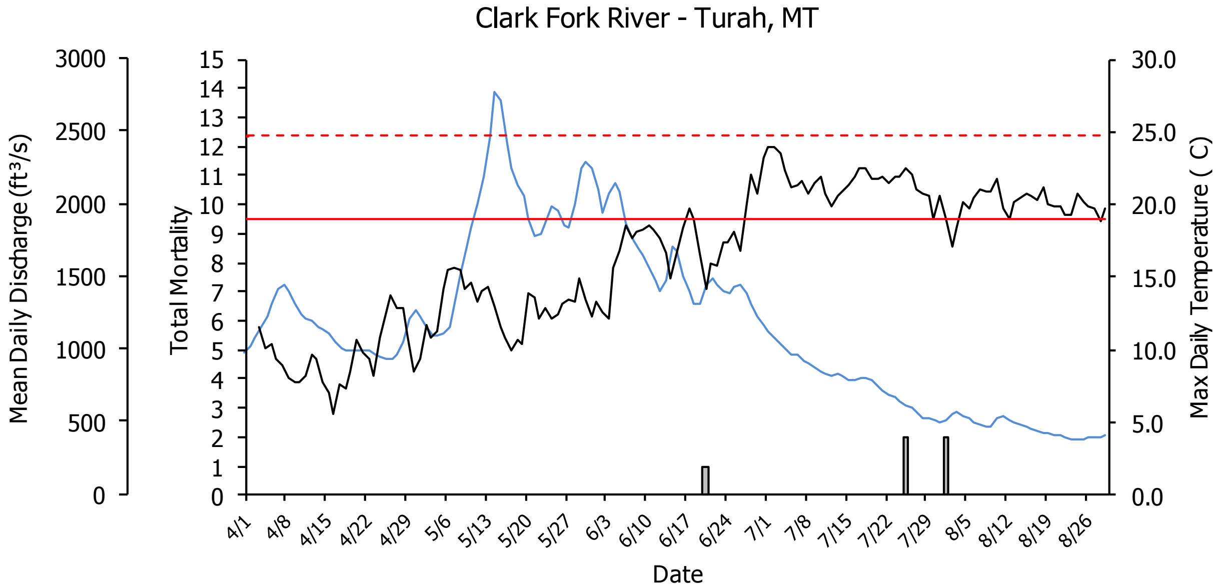

Figure 17. Total mortalities between cages one and two combined (gray bars), maximum daily water temperature (black line), and mean daily discharge (blue line) for 2013 in the Clark Fork River near Turah, Montana. The solid red line indicates the upper critical temperature threshold and the dashed red line represents the upper incipient lethal temperature for brown trout.

Spatial Distribution of Brown Trout Survival

Cumulative survival (%) at each site was calculated by dividing the number of live fish at the end of the analysis period in cages one and two combined by the total number of fish placed in both cages over the entire season. Cumulative survival in 2013 (April 8 to July 31) from the Mill Willow site downstream was as follows; Warm Springs 89 %, Pond 2 58 %, Silver Bow 83 %, Warm Springs 83 %, Galen Left 83 %, Galen Right 81 %, Deer Lodge 89 %, U/S Lil Black 93 %, Lil Black 75 %, Flint 68 %, Clinton Spring 88 %, and Turah 94 % (Figure 18).

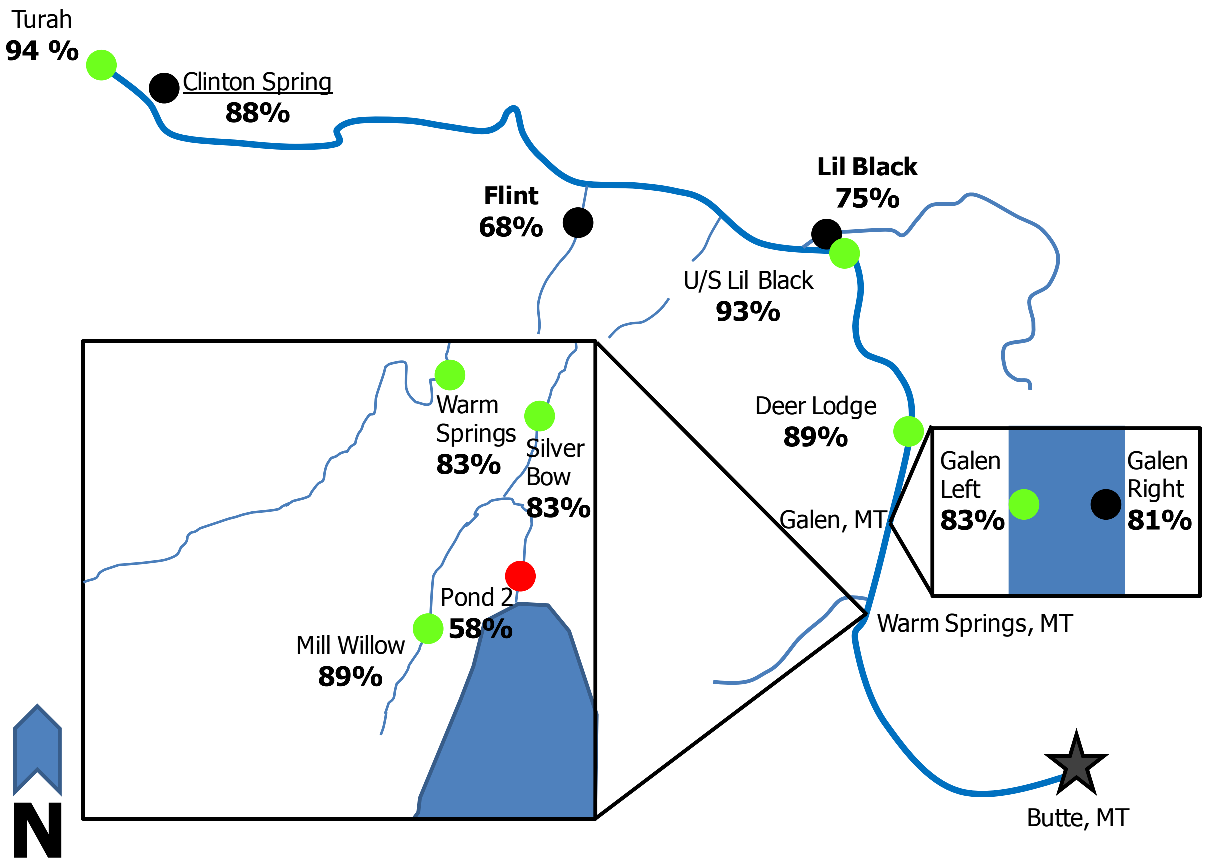

Figure 18. Cumulative brown trout survival calculated from April 8th to July 31st across sites for 2013 respectively. Control sites are shown in bold and the handling control is underlined. Red dots denote sites with significantly lower than expected survival; green dots denote sites with significantly higher than expected survival.

Survival in the control tributaries was low, with 75% survival in Lil Black and 68% in Flint. Clinton Spring (the handling control), showed 88% survival in 2013 indicating that mortalities observed at the experimental mainstem sites were not likely due to conditions inside the cages. Mill Willow, Silver Bow, Warm Springs, Galen Left, Deer Lodge, U/S Lil Black, and Turah exhibited significantly higher survival than expected. Pond 2 was the only cage site that exhibited significantly lower survival than expected. It is important to note that mortality was higher at control sites in 2013 (72% average survival) than in previous years of this study documented by Richards et al. (2013) (89% average survival in 2011 and 2012).

Growth

Growth was higher at Clinton Spring (handling control) and at Lil Black (control) than at adjacent mainstem sites; however, growth at Flint Creek (the other control site), was lower than at the adjacent mainstem sites (Figure 19). In 2013, growth was higher at the sites with the lowest water temperatures and/or least days that exceeded 19 °C (Warm Springs, Lil Black, and Clinton Spring) indicating that high temperature in 2013 had an effect on growth at the remaining sites. The high growth rates at Warm Springs, Galen Left, and Galen Right are expected as there is a "tail water" effect of the upstream ponds resulting in increased nutrients and additional food sources including freshwater shrimp and isopods.

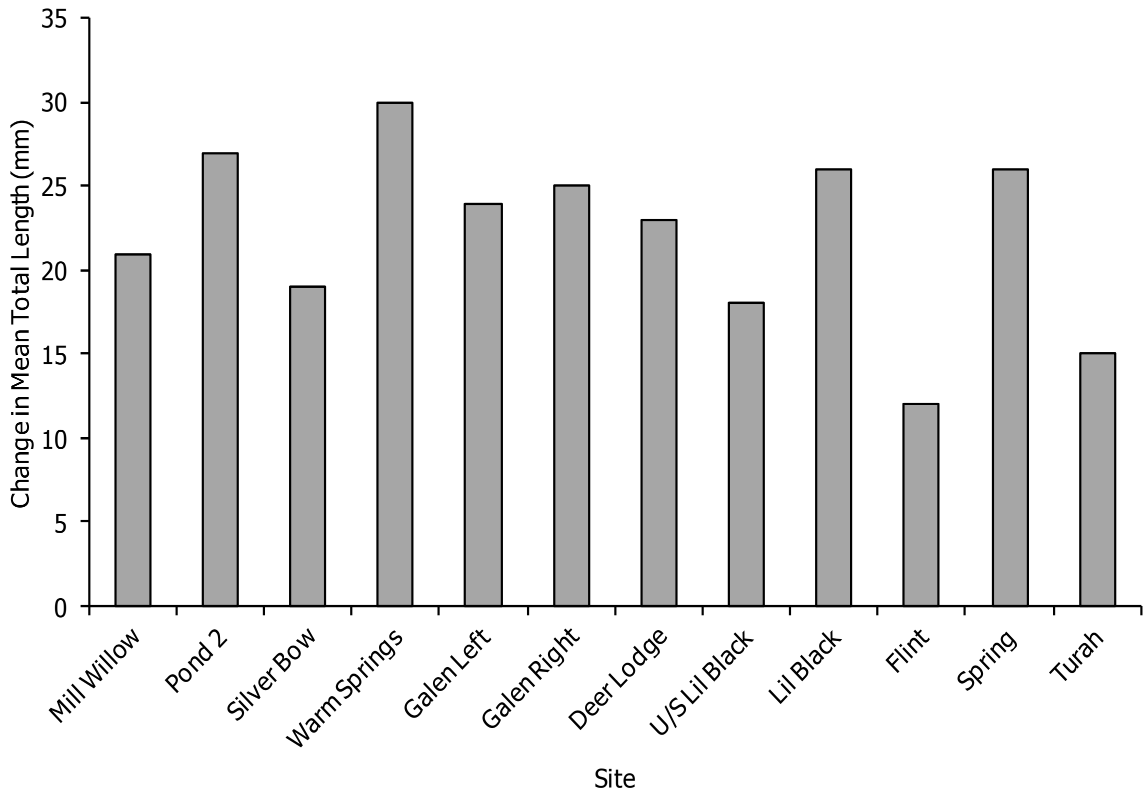

Figure 19. Change in mean total length by site for juvenile brown trout held in cages by site in 2013, arranged from upstream to downstream.

Tissue Metals Burdens

Tissue metals burdens from fish held at cage sites were compared to reported values from previous studies assessing growth or mortality effects using whole body burdens in salmonids. Studies reporting effects on rainbow trout from whole body burdens of copper and zinc were found. Table 8 displays reported minimum effect thresholds for trout and their associated studies.

Table 8. Summary of studies relating whole body metals burdens to growth or mortality effects in salmonids. All values reported were the minimum concentrations causing an effect.

Species	Metal	Metal concentration (µg/g dry weight)	Reference
Rainbow trout	Copper	8.57	Marr et al. 1996
Rainbow trout	Zinc	105.09*	Gundogdu and Erdem 2008

* Zinc value presented represents a level that may cause impaired fish health and was used as a threshold minimum effect threshold for this report.

Copper and zinc consistently exceeded the aforementioned minimum effect thresholds by month. It is important to note that fish were fed a feed diet before (at hatchery) and during the course of this study that is likely high in zinc and copper (T. Selch, Montana, Fish, Wildlife, and Parks, personal communication, 2014). Hatchery samples showed copper tissue burden levels below minimum effect thresholds and zinc tissue burden levels above minimum effect thresholds (Figure 20). All tissues metals burdens for copper and zinc were depicted for individual samples graphically by site (Figures 21-31). Generally, tissues metals burdens were higher at mainstem sites than at control sites. Galen Left had the highest sample value of copper with a sample value of 63.2 µg/g (Figures 21-31). Pond 2 had the highest sample value of zinc with a sample value of 574.00 µg/g (Figures 21-31). Copper and zinc burdens typically peaked in May from Mill Willow to Galen Left, with the exceptions of Pond 2 (Zinc peaked in August) and Silver Bow (peaked mid-season). Conversely, copper and zinc burdens typically peaked in July from Lil Black to Turah (Figures 21-31).

Sample values of copper and zinc were higher in dead fish than in live fish. All tissues metals burdens for copper and zinc were depicted for live versus dead fish graphically by site cages (Figures 32-42). Generally, tissues metals burdens were higher at mainstem sites than at control sites. Most sites experienced tissue metals burdens that did not exceed minimum effect thresholds for copper in live fish and that did exceed minimum effect thresholds for copper in dead fish (Figures 32-42). Exceptions to this were Mill Willow, Deer Lodge, U/S Lil Black, and Lil Black, which had tissue metals burdens that exceeded minimum effect thresholds for copper in both live and dead fish. Every site had all sample values of tissues metals burdens that exceeded minimum effect thresholds for zinc (Figures 32-42).

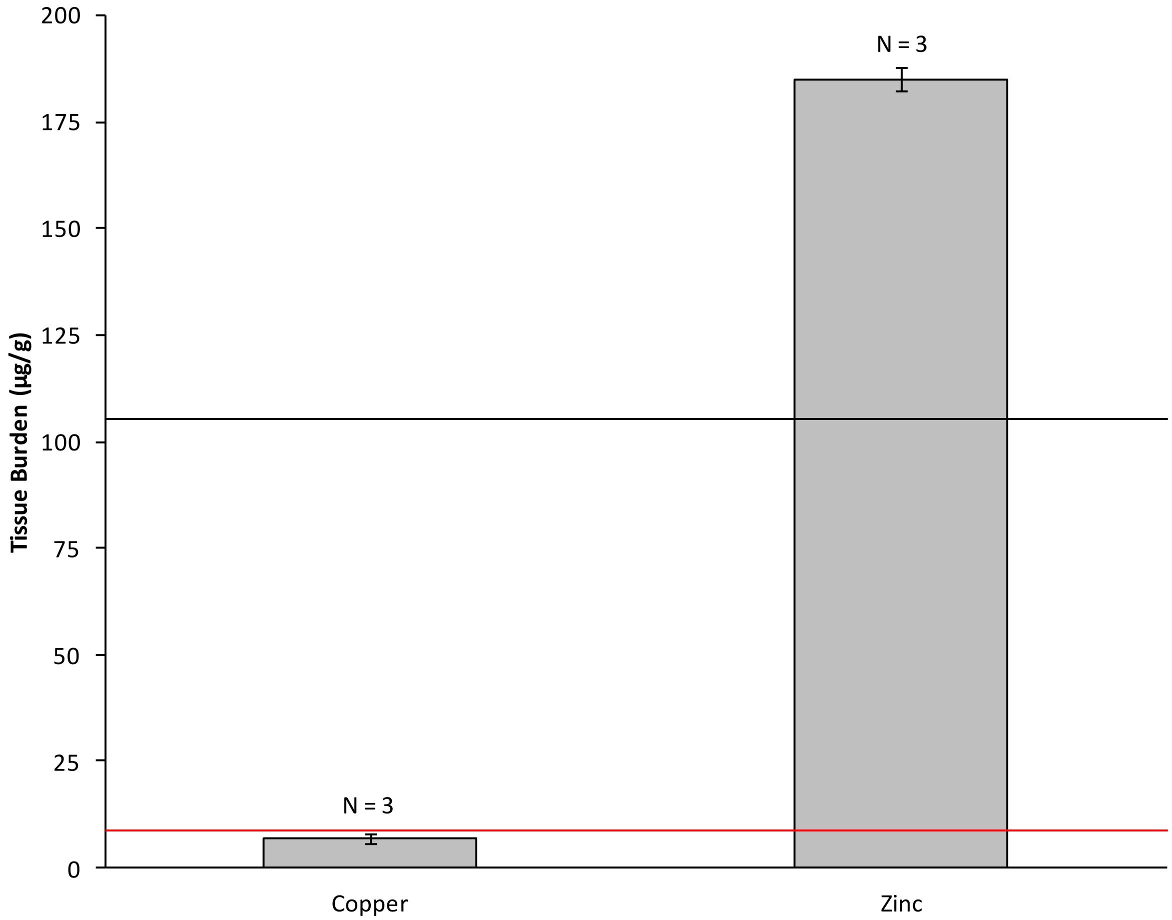

Figure 20. Copper and zinc tissue burdens for hatchery fish not placed in cages. Individual samples are shown with 95% confidence intervals. The red line in each panel indicates the copper minimum effect threshold identified for salmonids. The black line represents the zinc minimum effect threshold identified for salmonids. Sample sizes represent the number of fish never placed in cages submitted for analysis.

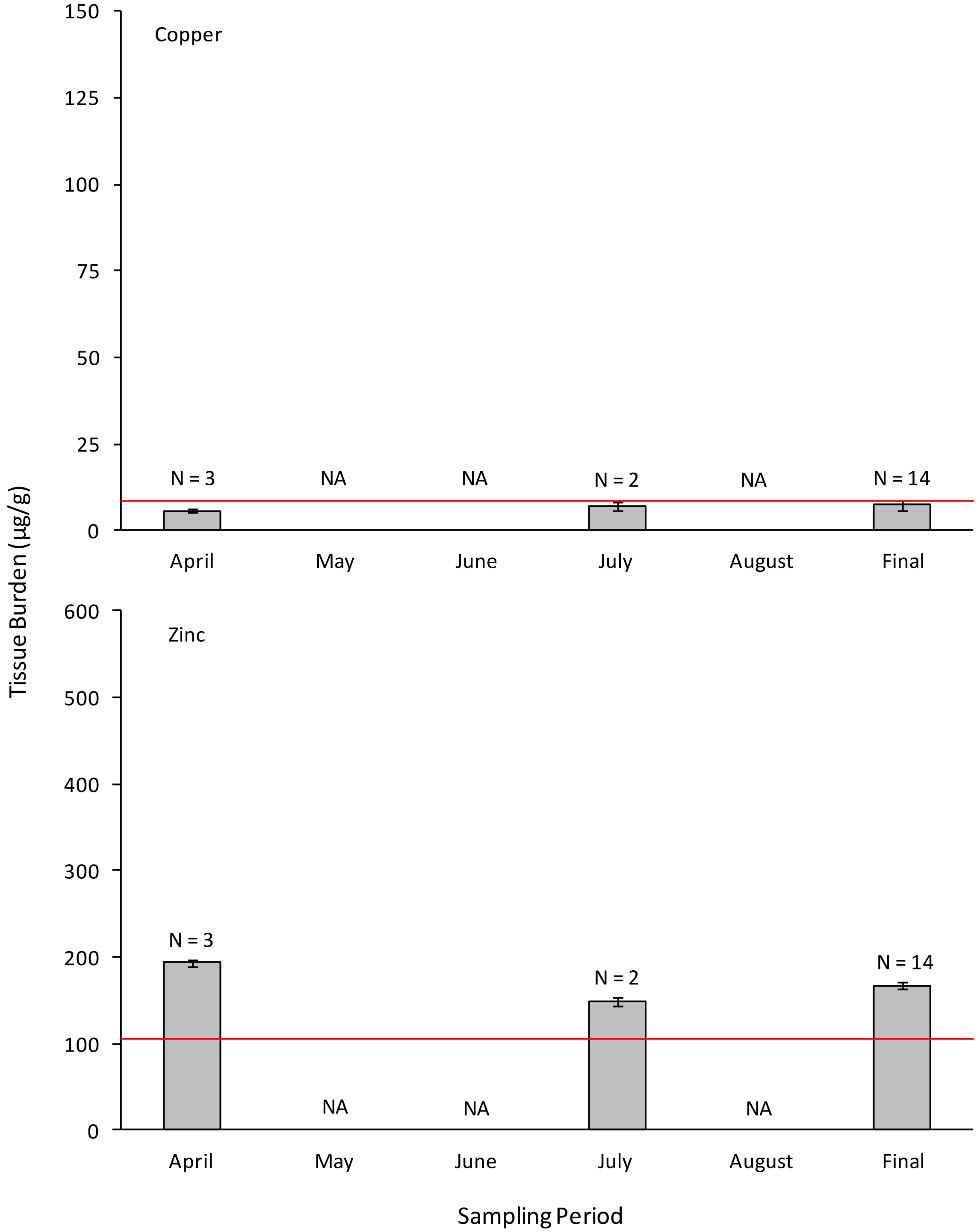

Figure 21. Copper and zinc tissue burdens by month at Mill Willow in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

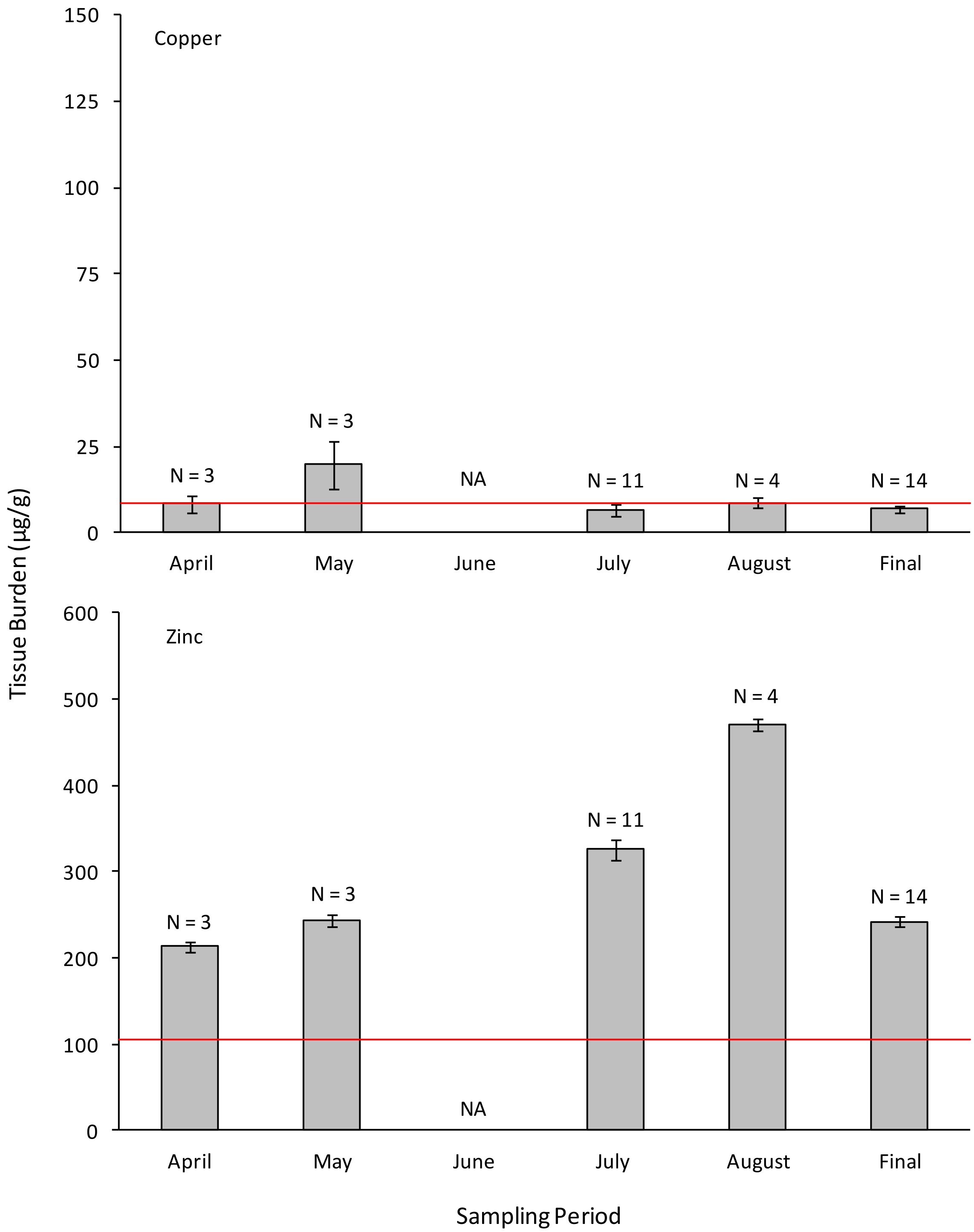

Figure 22. Copper and zinc tissue burdens by month at Pond 2 in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

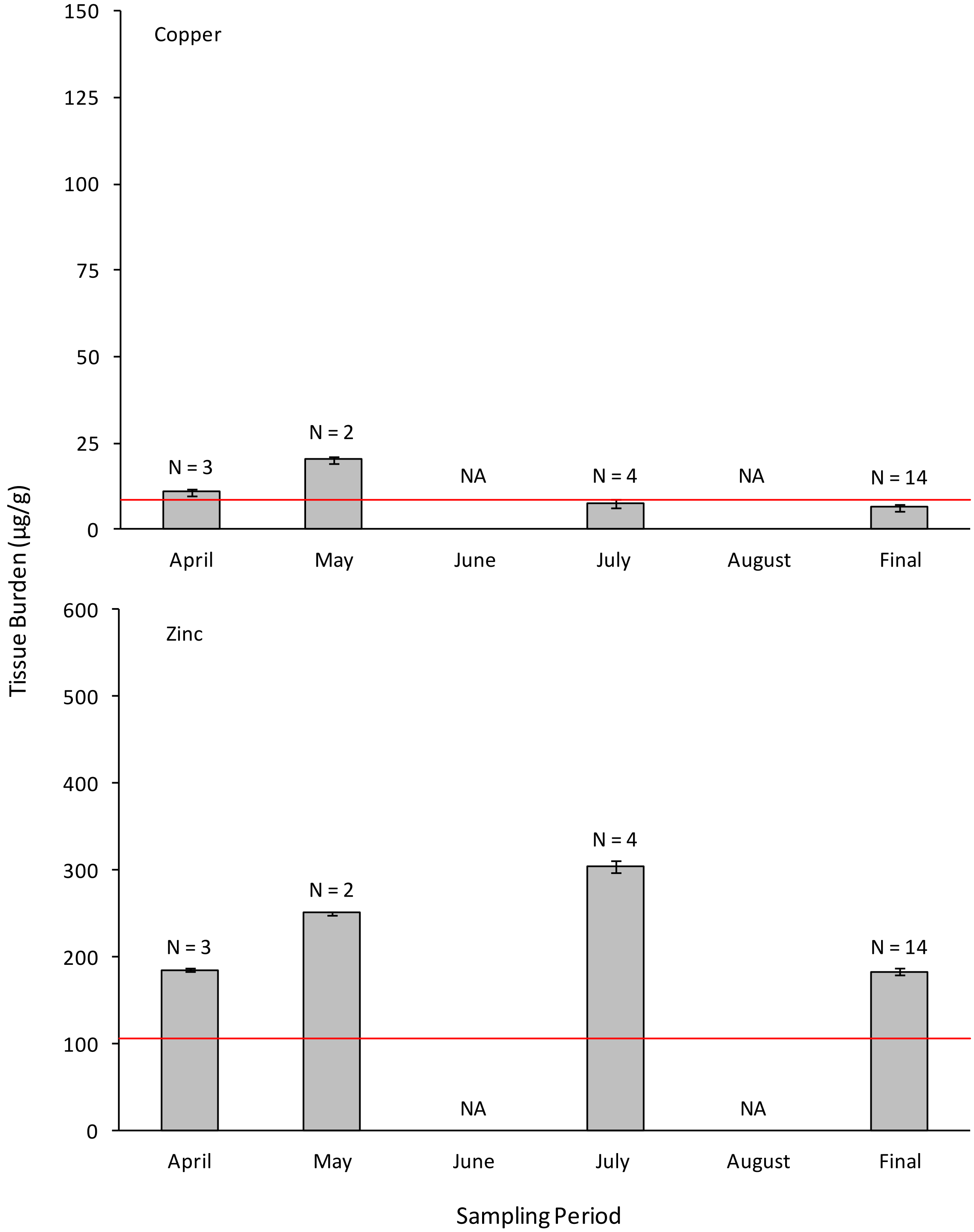

Figure 23. Copper and zinc tissue burdens by month at Silver Bow in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

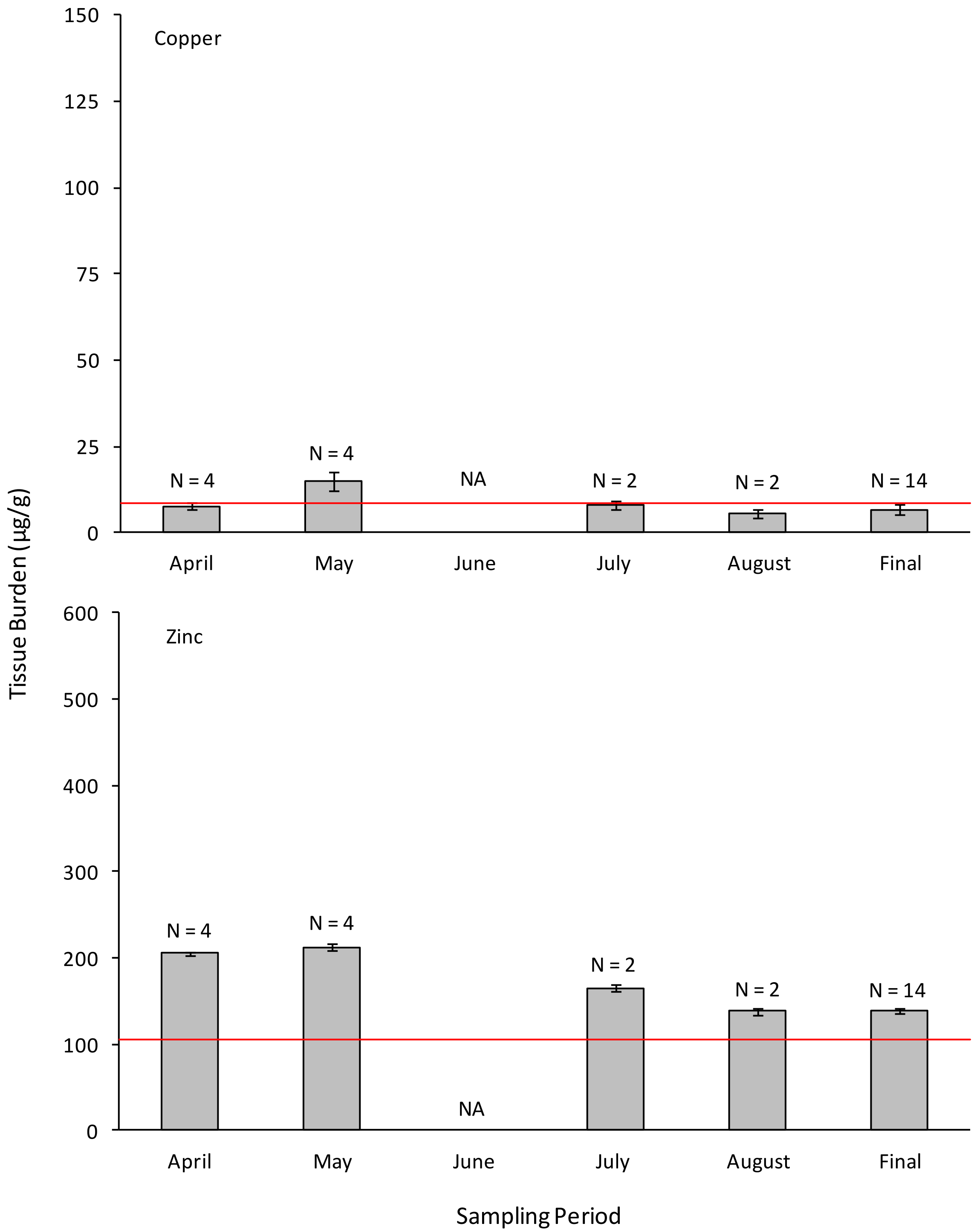

Figure 24. Copper and zinc tissue burdens by month at Warm Springs in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

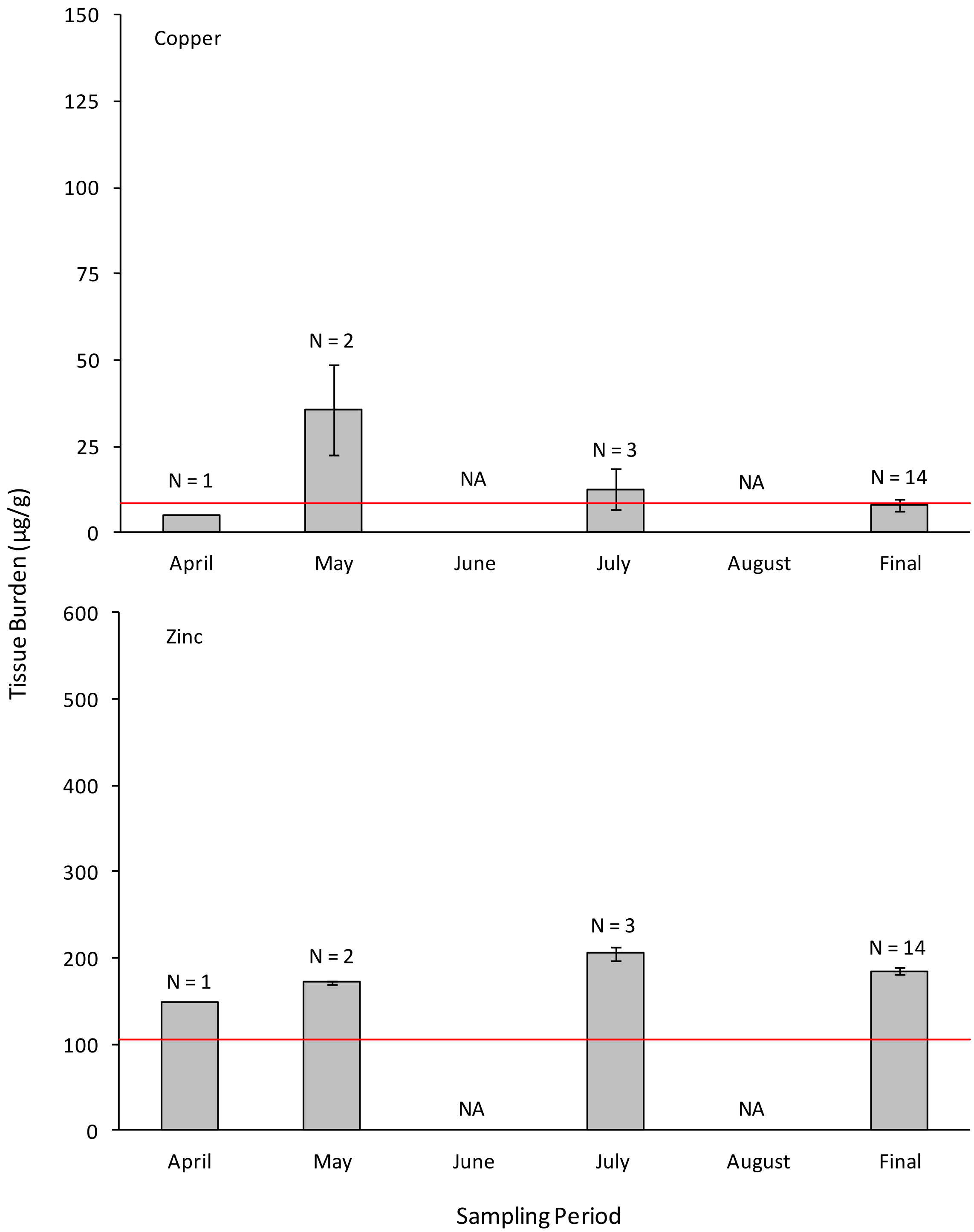

Figure 25. Copper and zinc tissue burdens by month at Galen Left in Galen, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

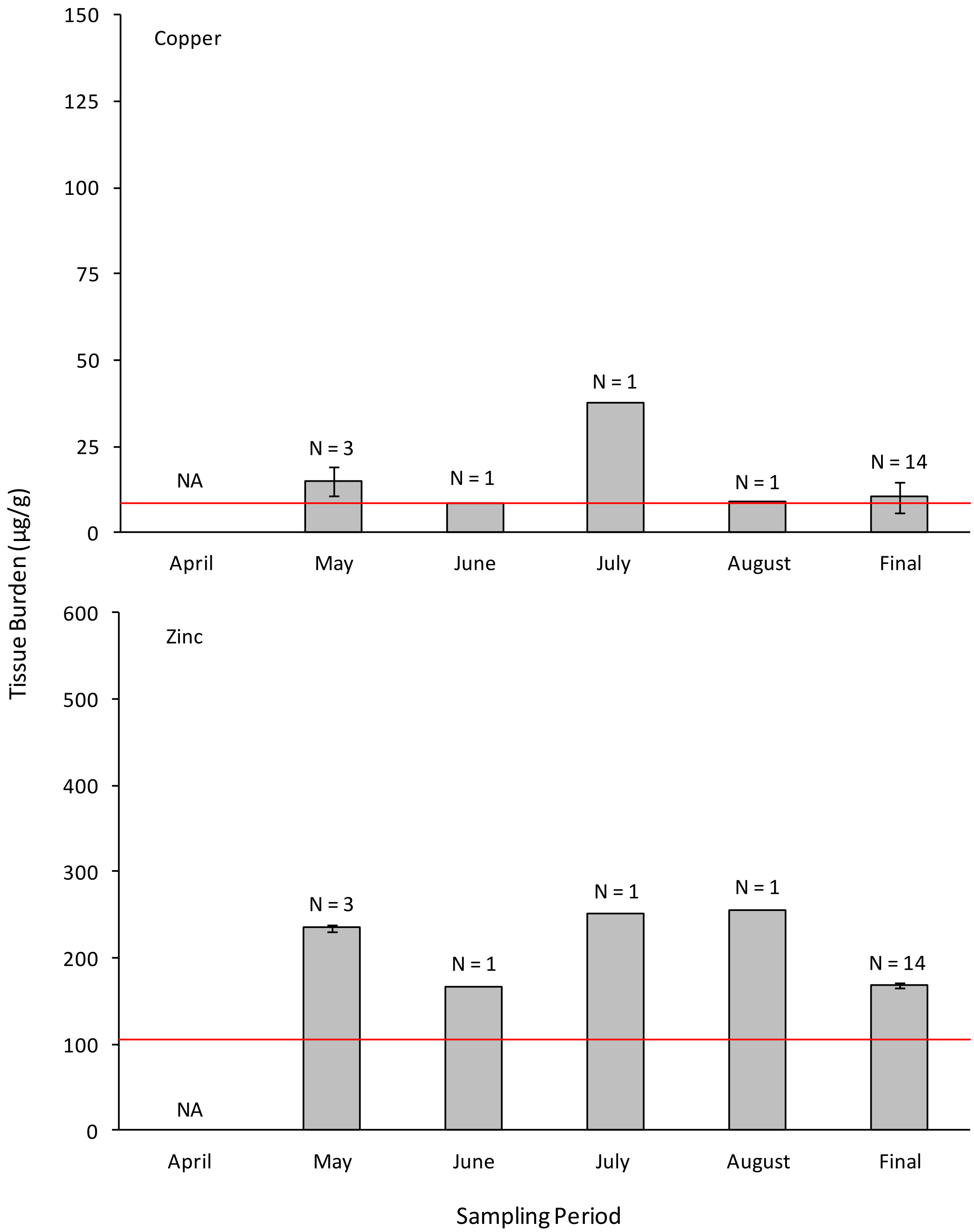

Figure 26. Copper and zinc tissue burdens by month at Deer Lodge in Deer Lodge, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

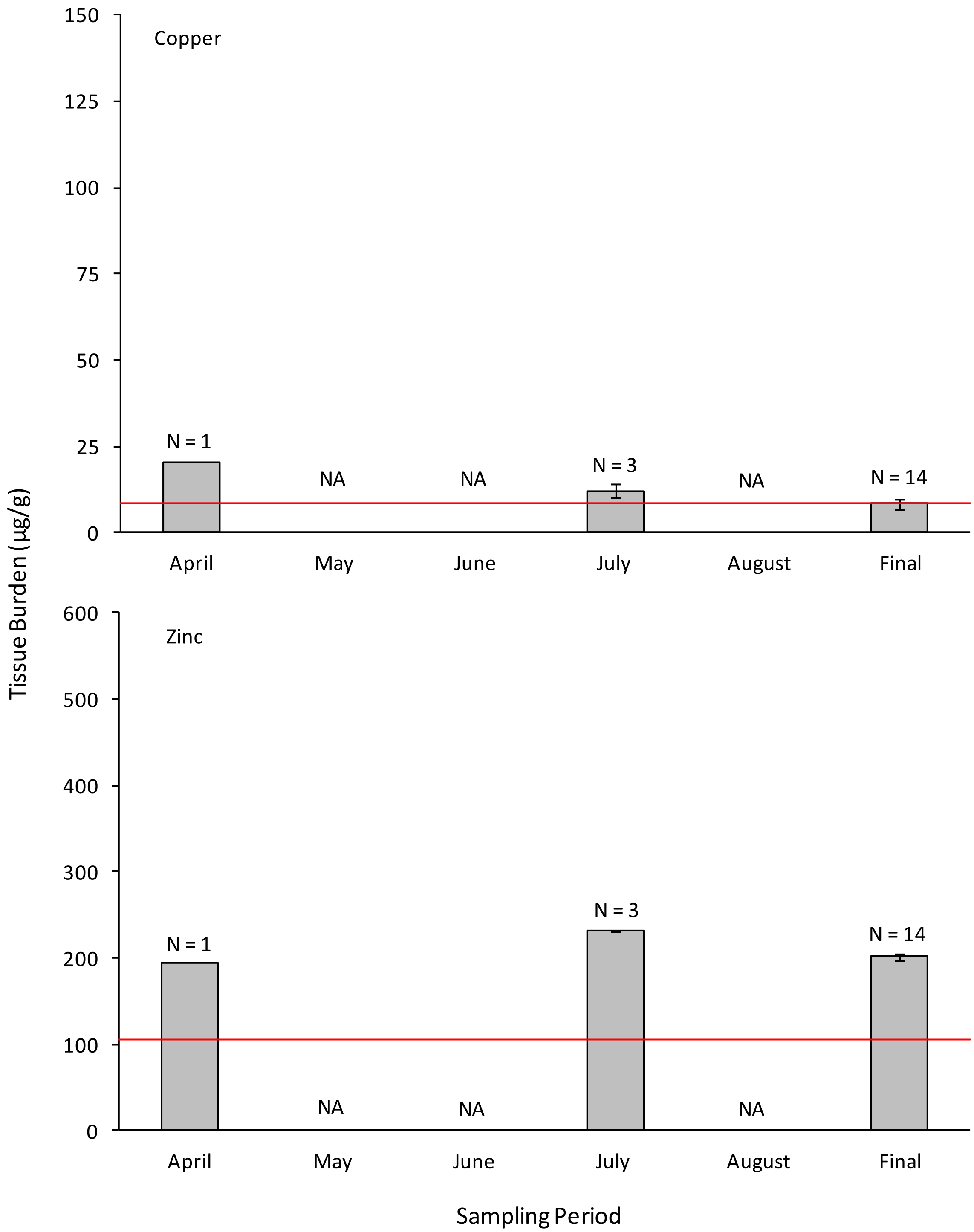

Figure 27. Copper and zinc tissue burdens by month at U/S Lil Black in Garrison, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

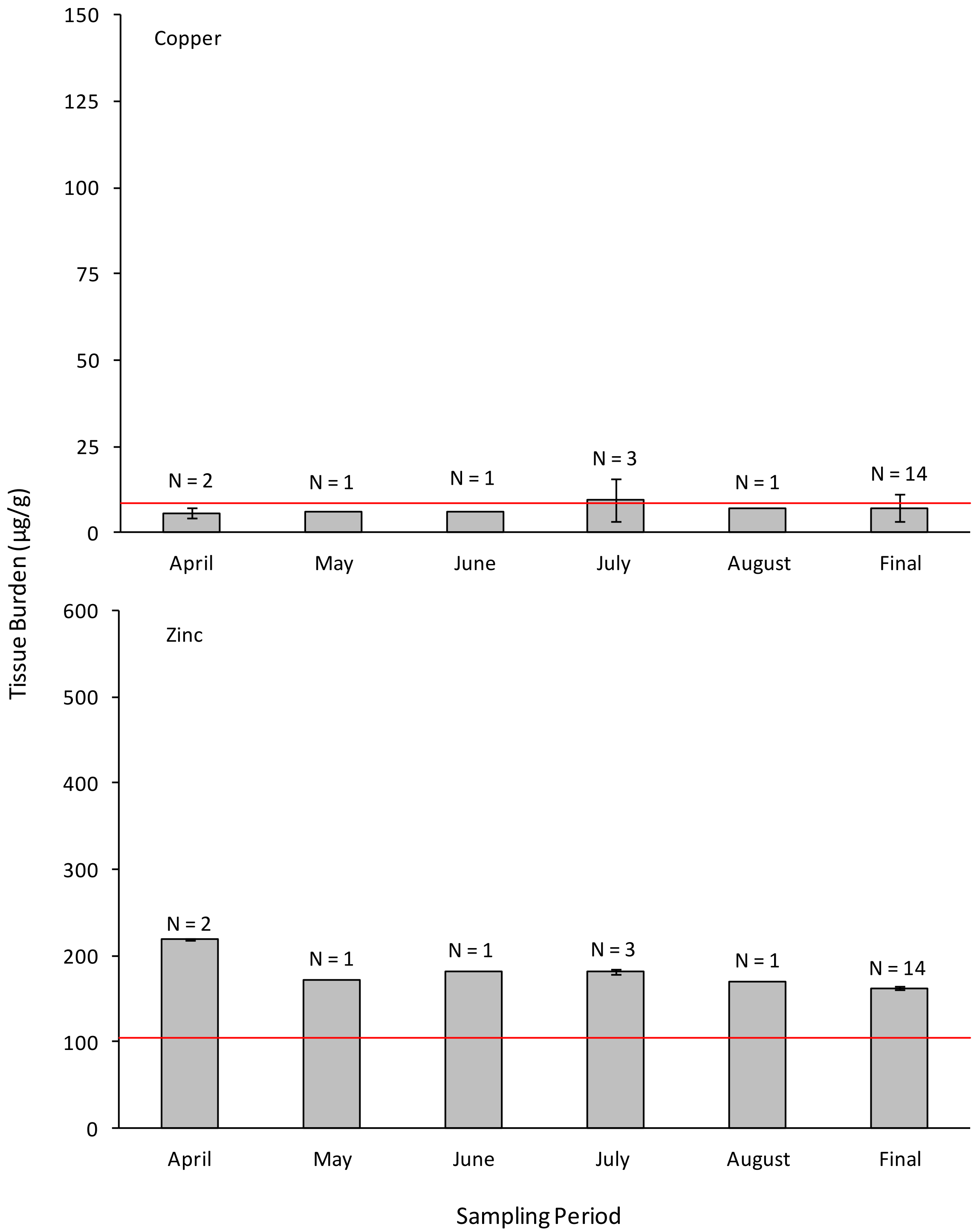

Figure 28. Copper and zinc tissue burdens by month at Lil Black in Garrison, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

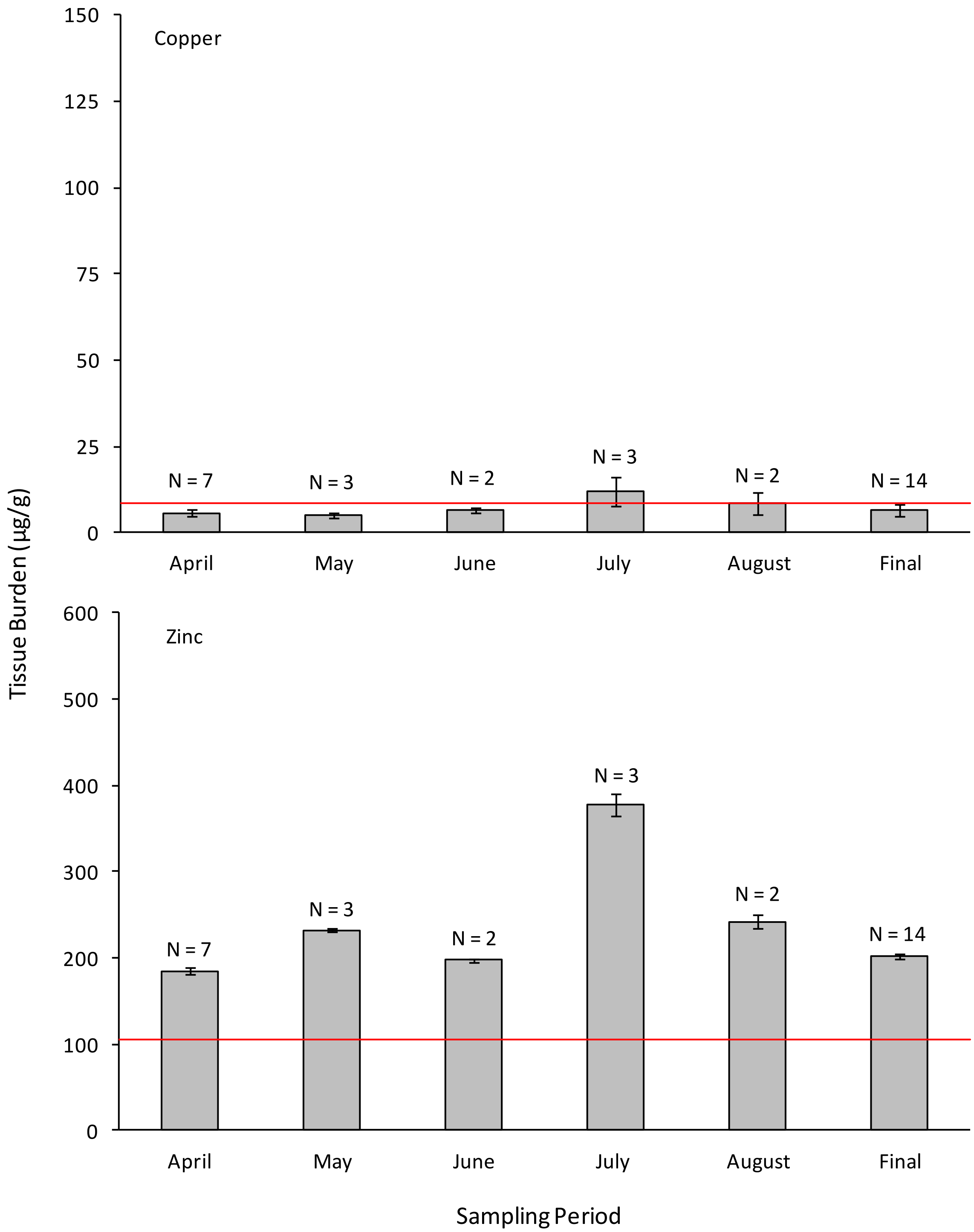

Figure 29. Copper and zinc tissue burdens by month at Flint in Drummond, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

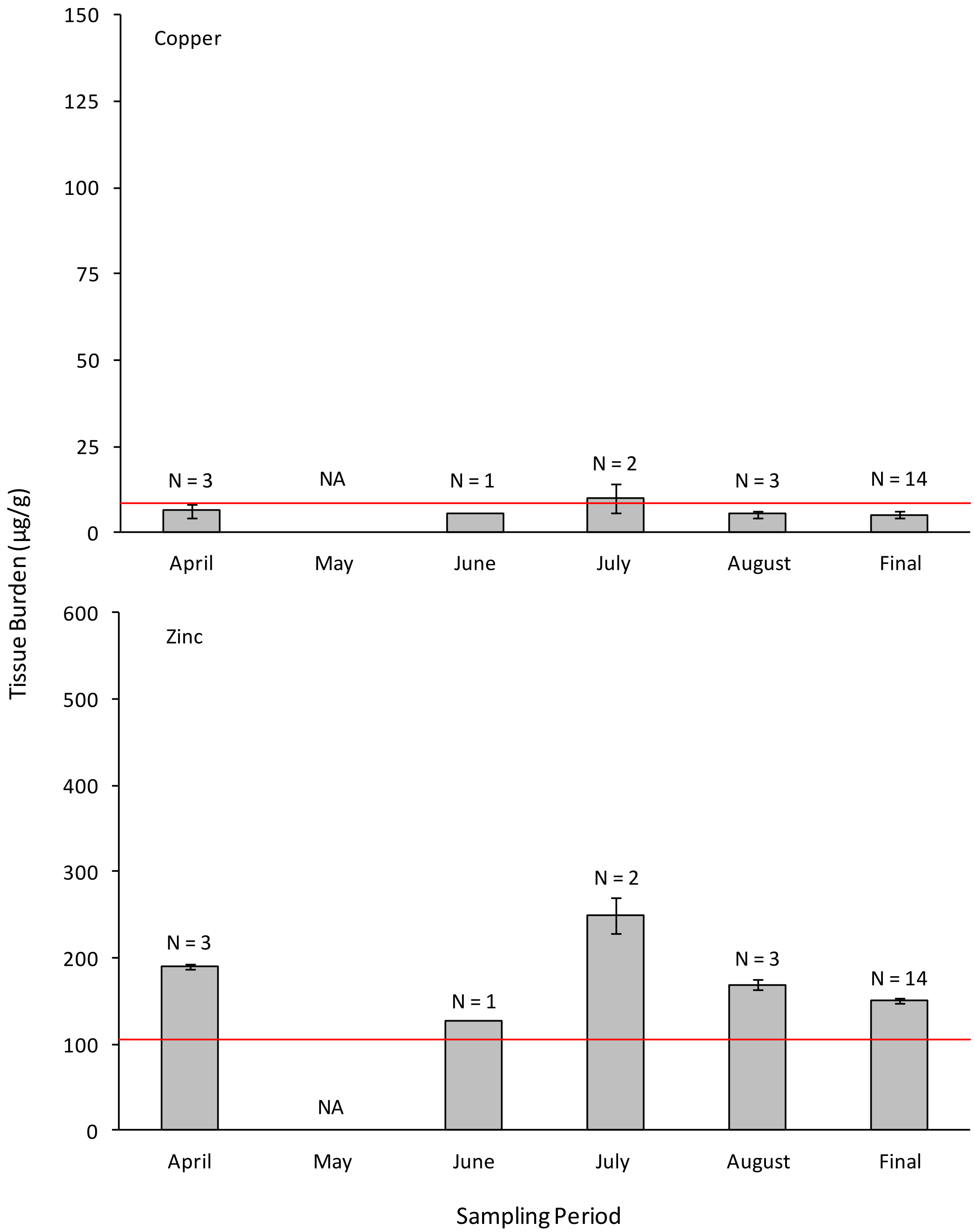

Figure 30. Copper and zinc tissue burdens by month at Clinton Spring in Clinton, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

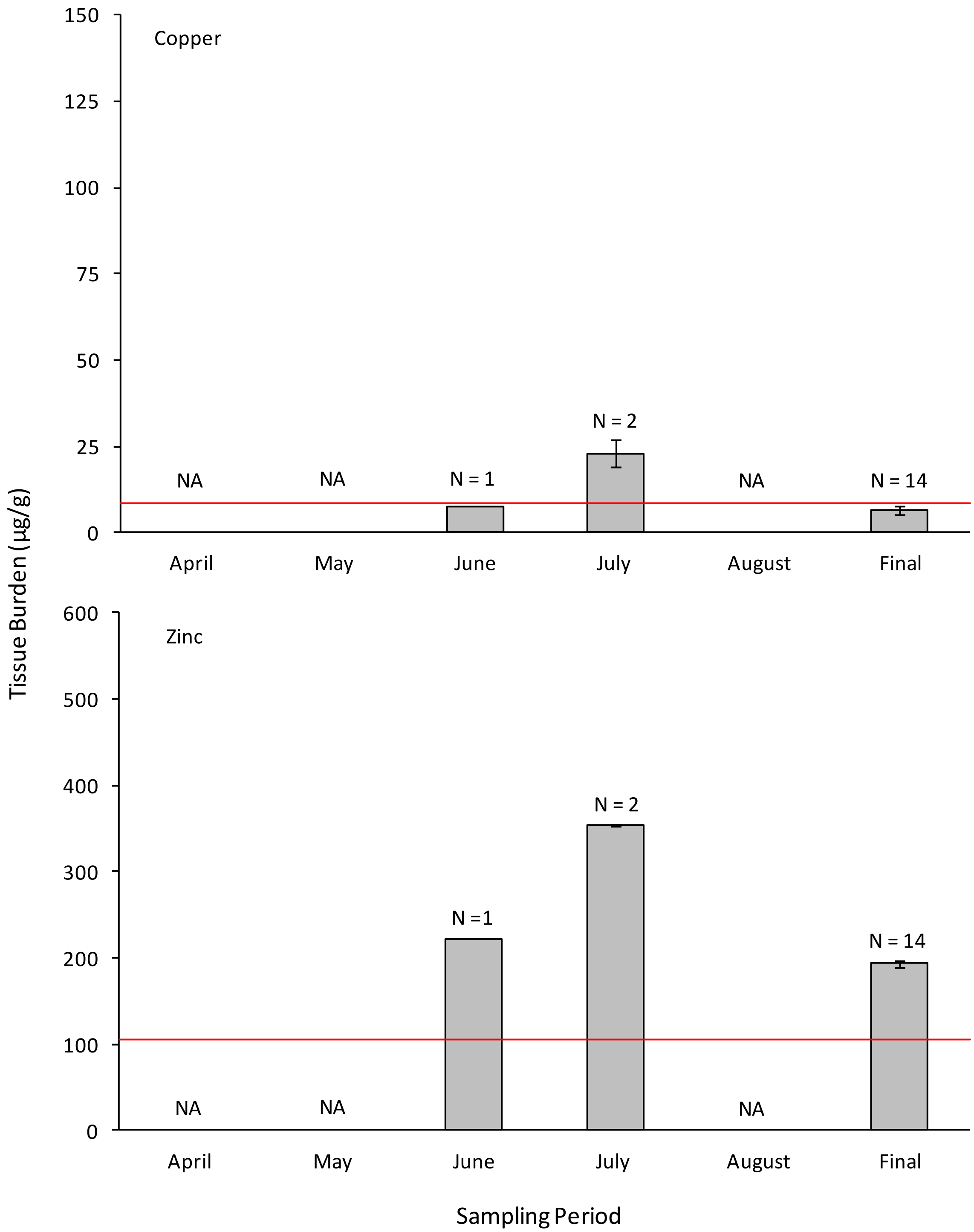

Figure 31. Copper and zinc tissue burdens by month at Turah in Turah, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

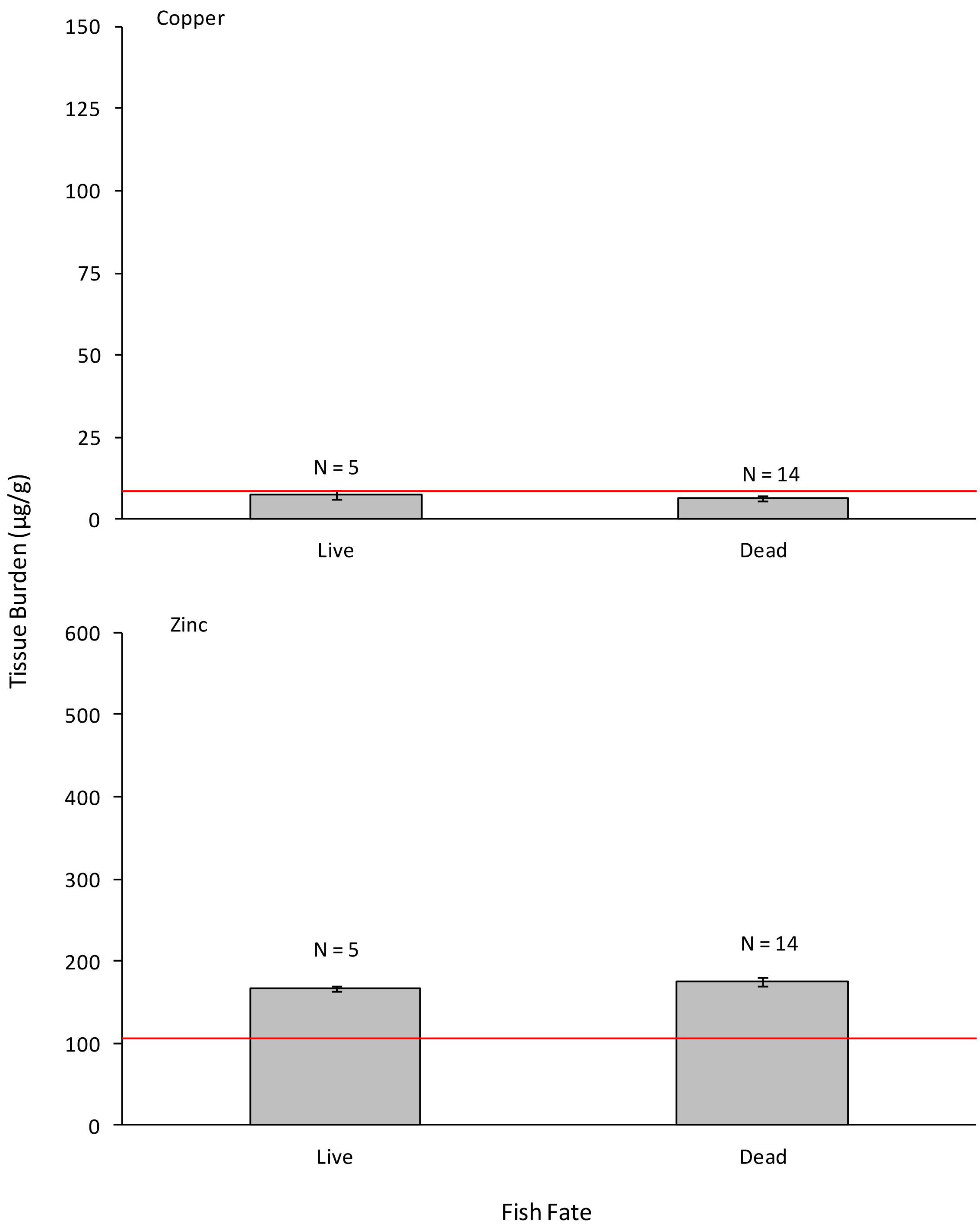

Figure 32. Copper and zinc tissue burdens for live fish versus dead fish at Mill Willow in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

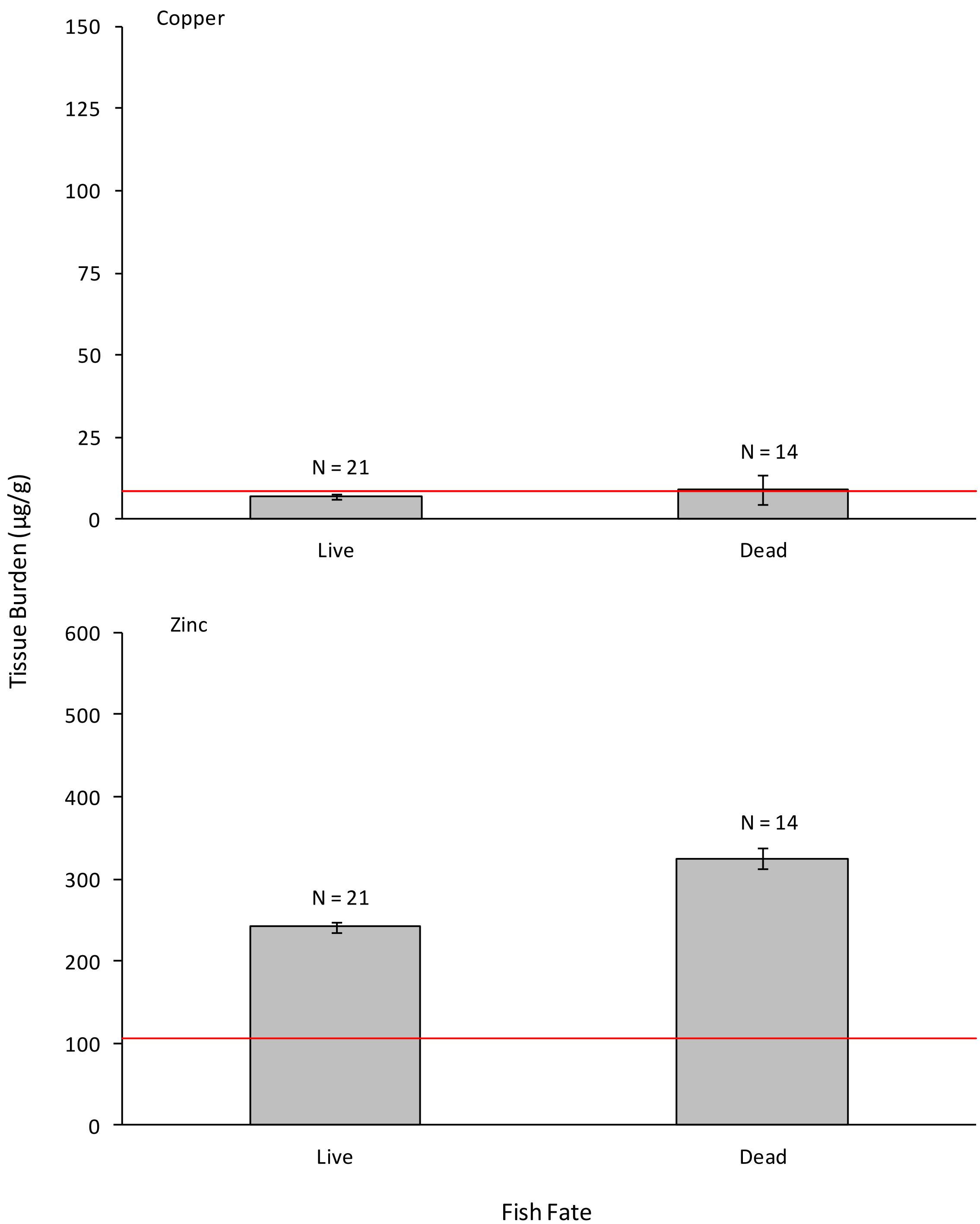

Figure 33. Copper and zinc tissue burdens for live fish versus dead fish at Pond 2 in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

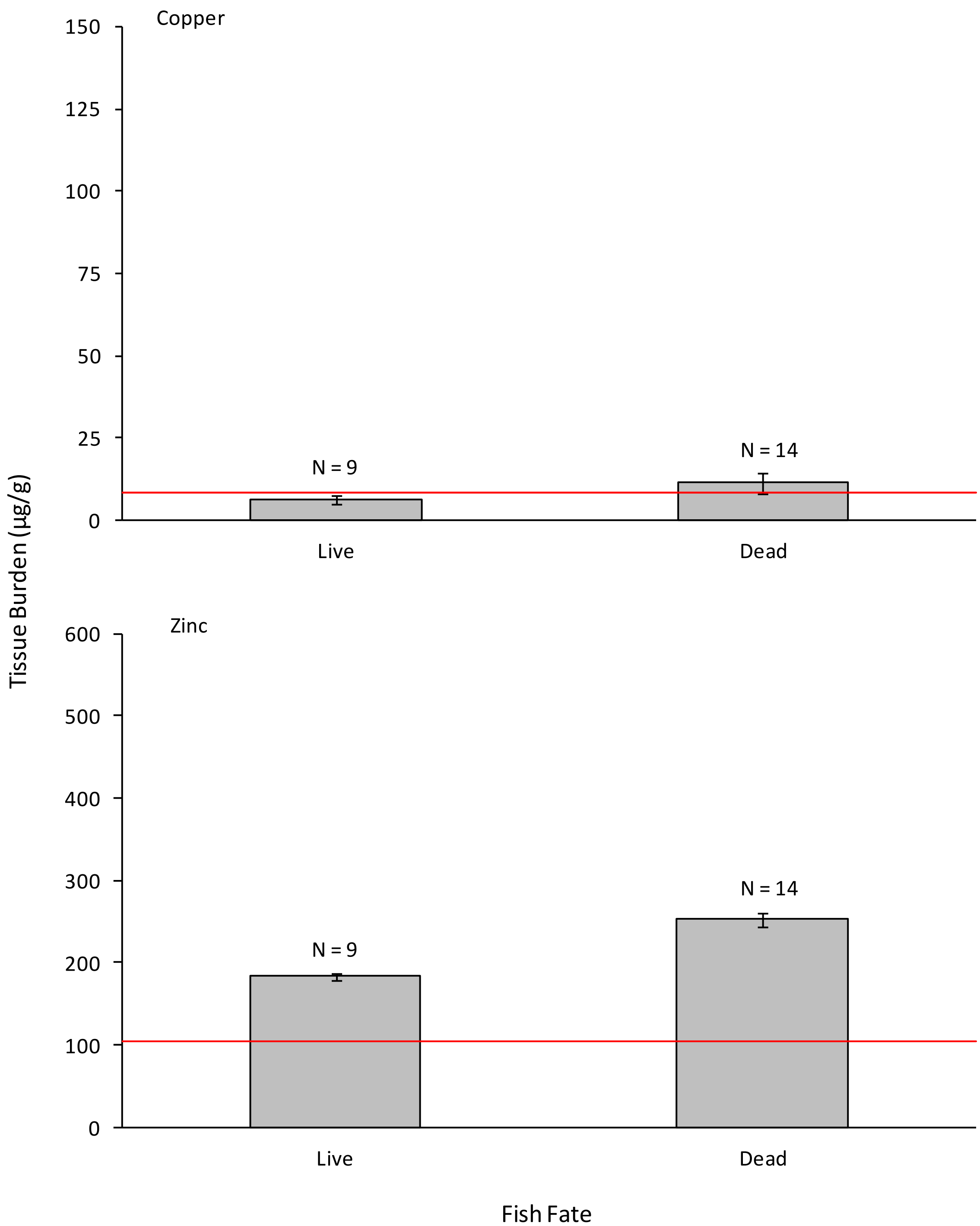

Figure 34. Copper and zinc tissue burdens for live fish versus dead fish at Silver Bow in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

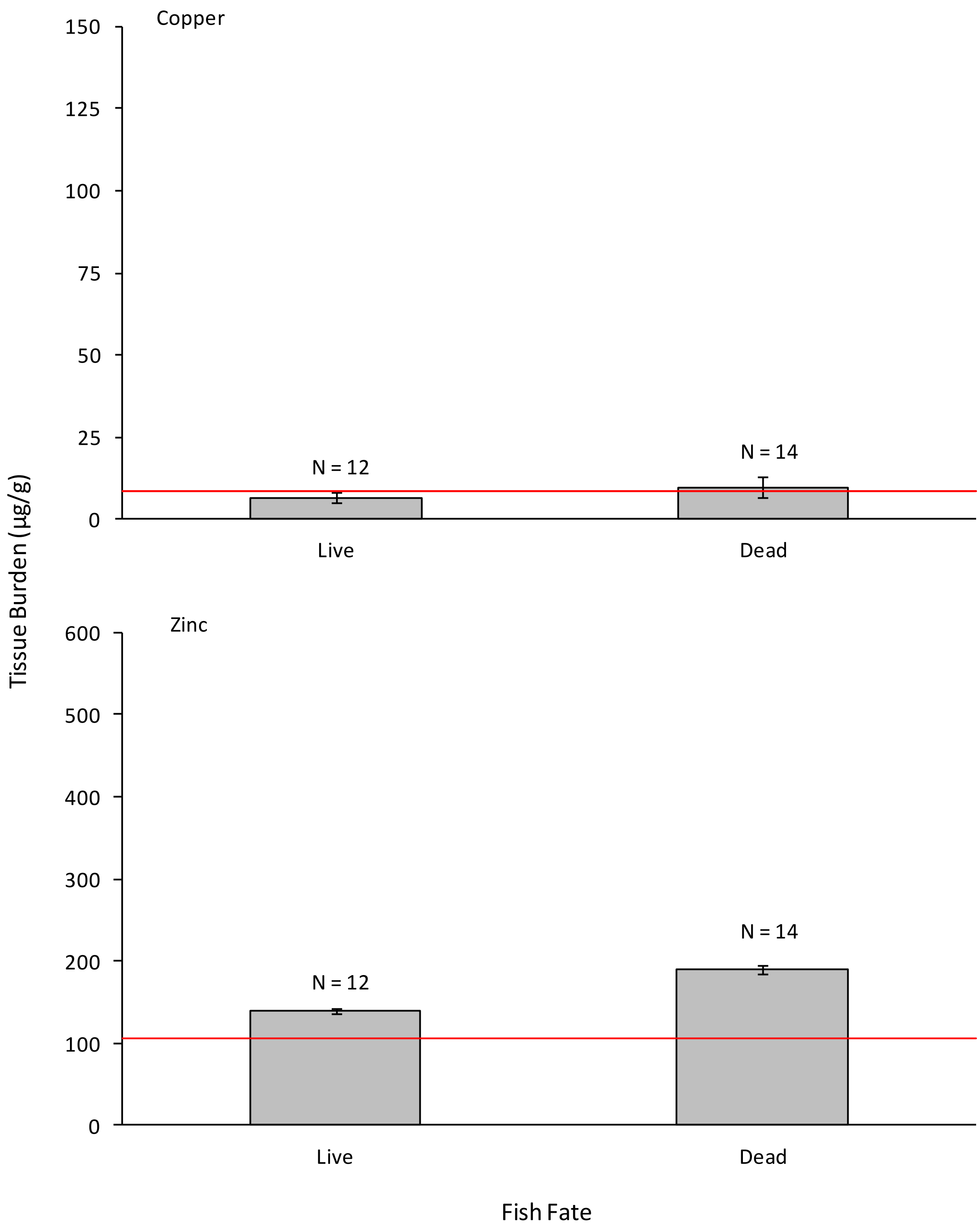

Figure 35. Copper and zinc tissue burdens for live fish versus dead fish at Warm Springs in Warm Springs, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

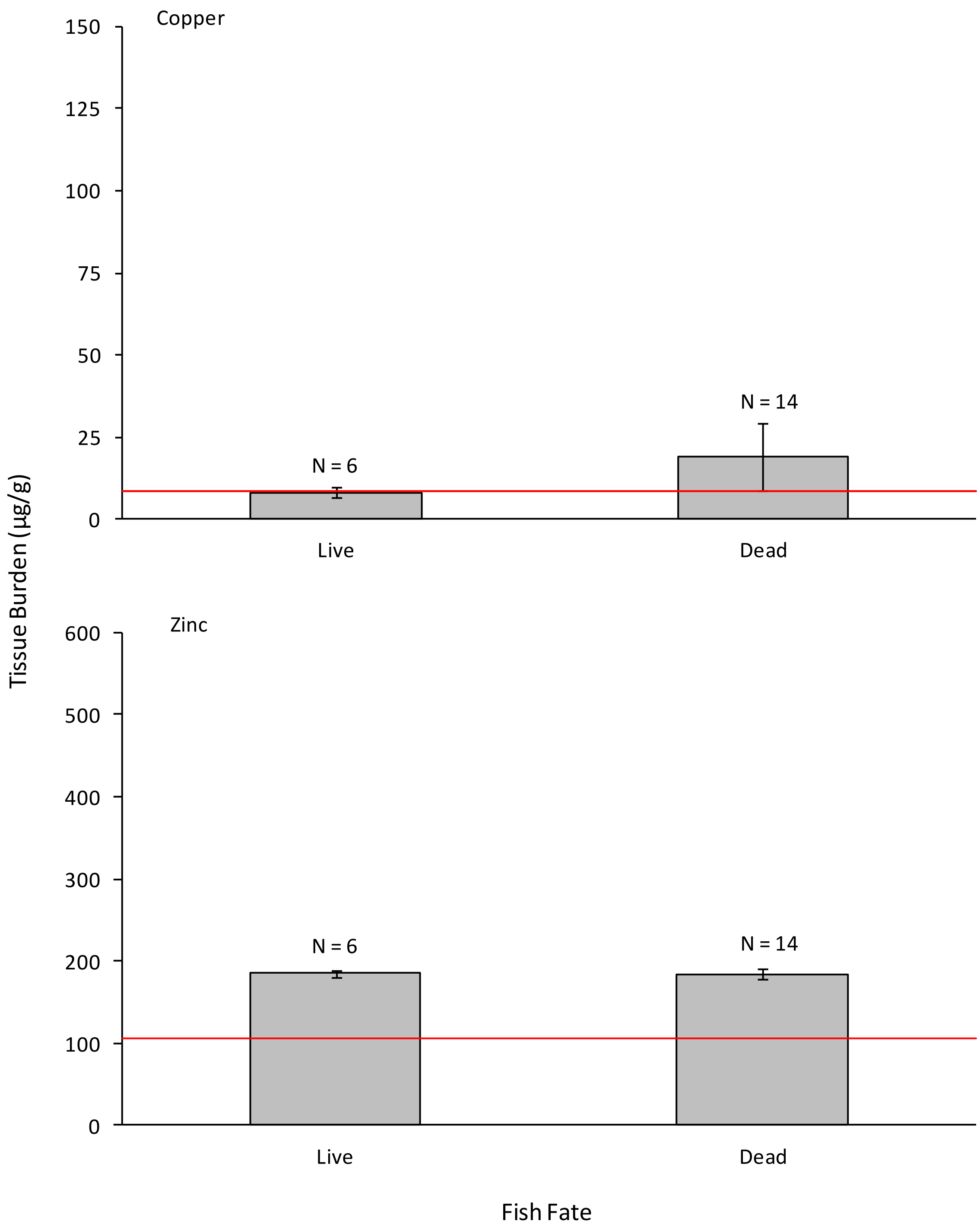

Figure 36. Copper and zinc tissue burdens for live fish versus dead fish at Galen Left in Galen, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

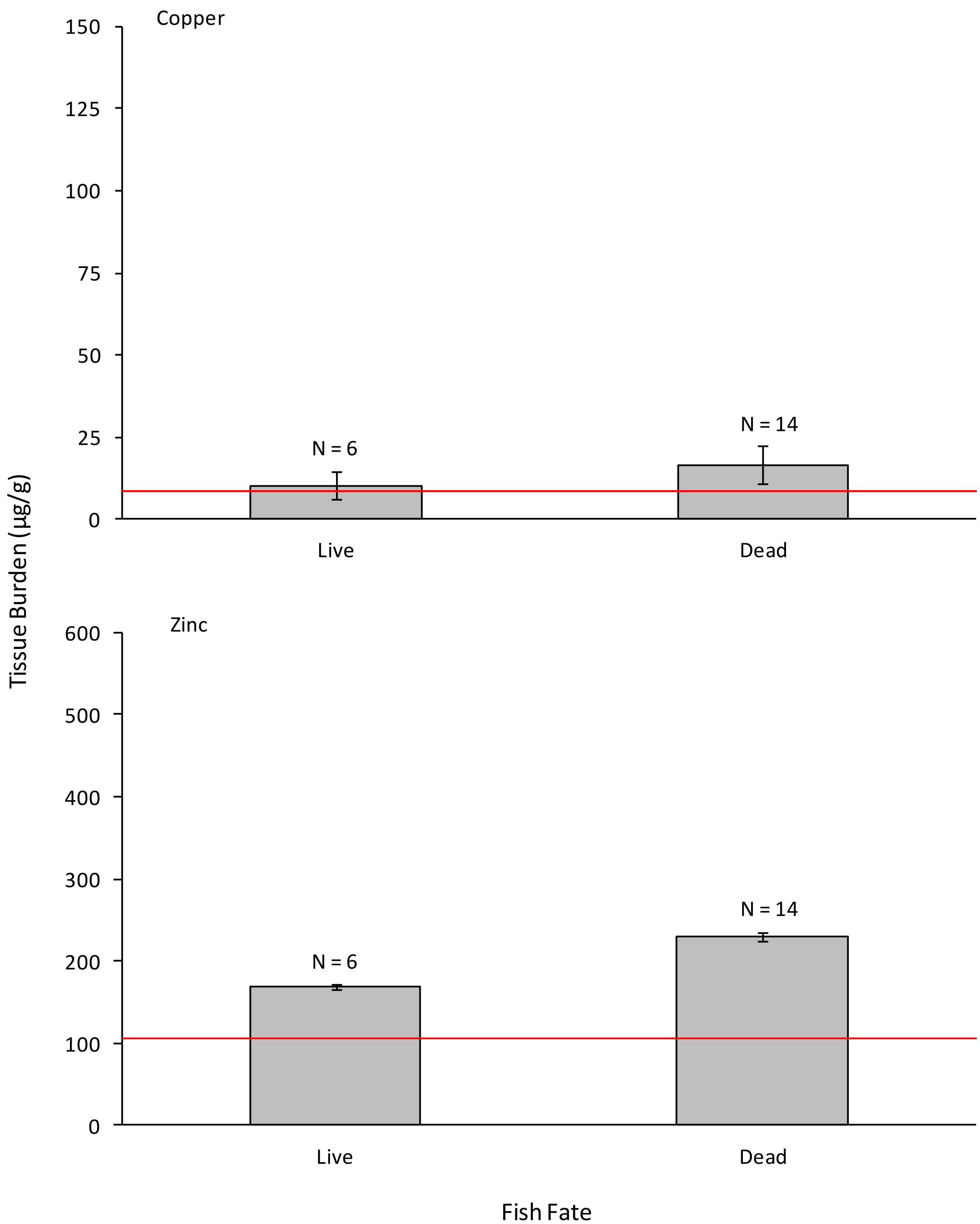

Figure 37. Copper and zinc tissue burdens for live fish versus dead fish at Deer Lodge in Deer Lodge, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

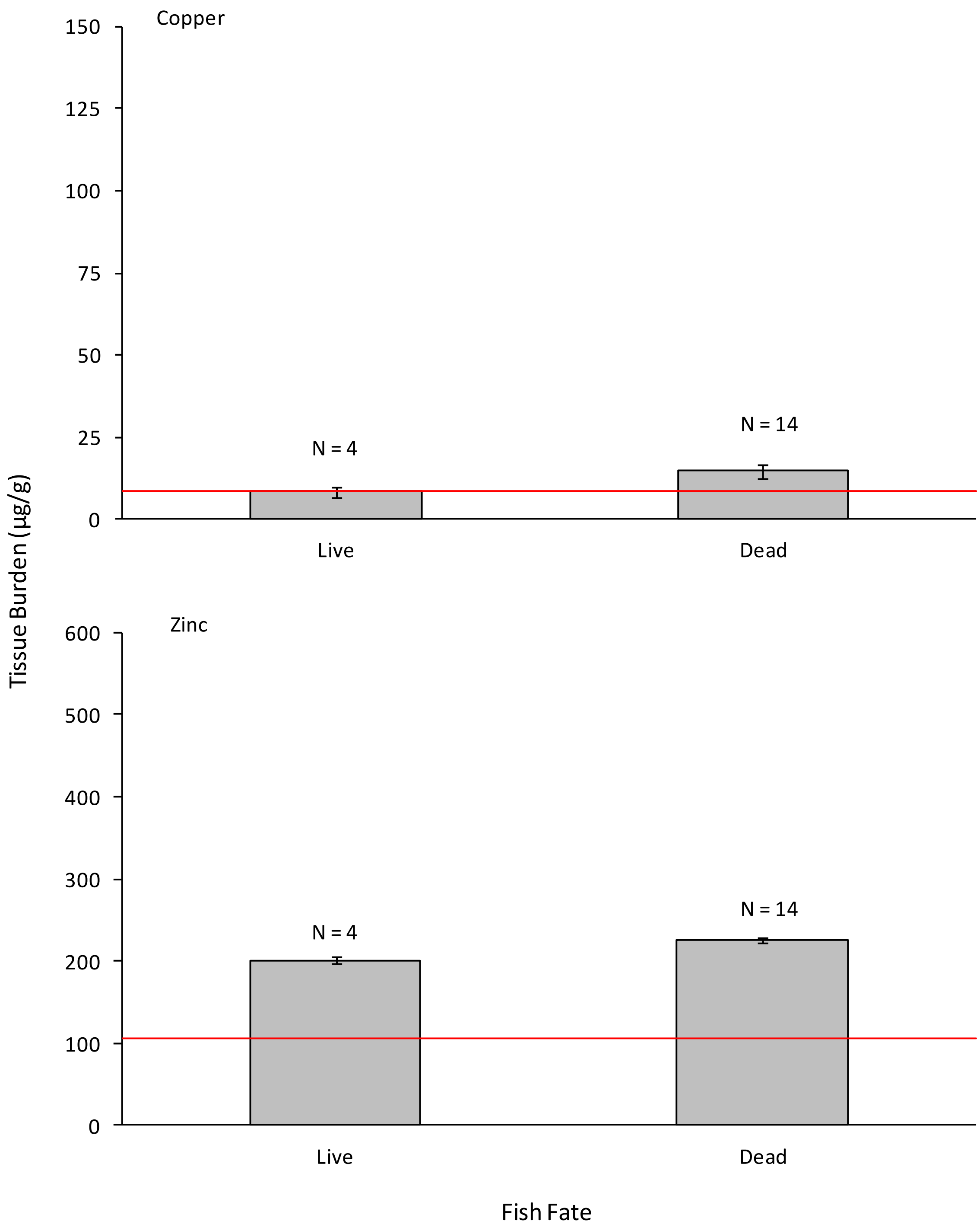

Figure 38. Copper and zinc tissue burdens for live fish versus dead fish at U/S Lil Black in Garrison, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

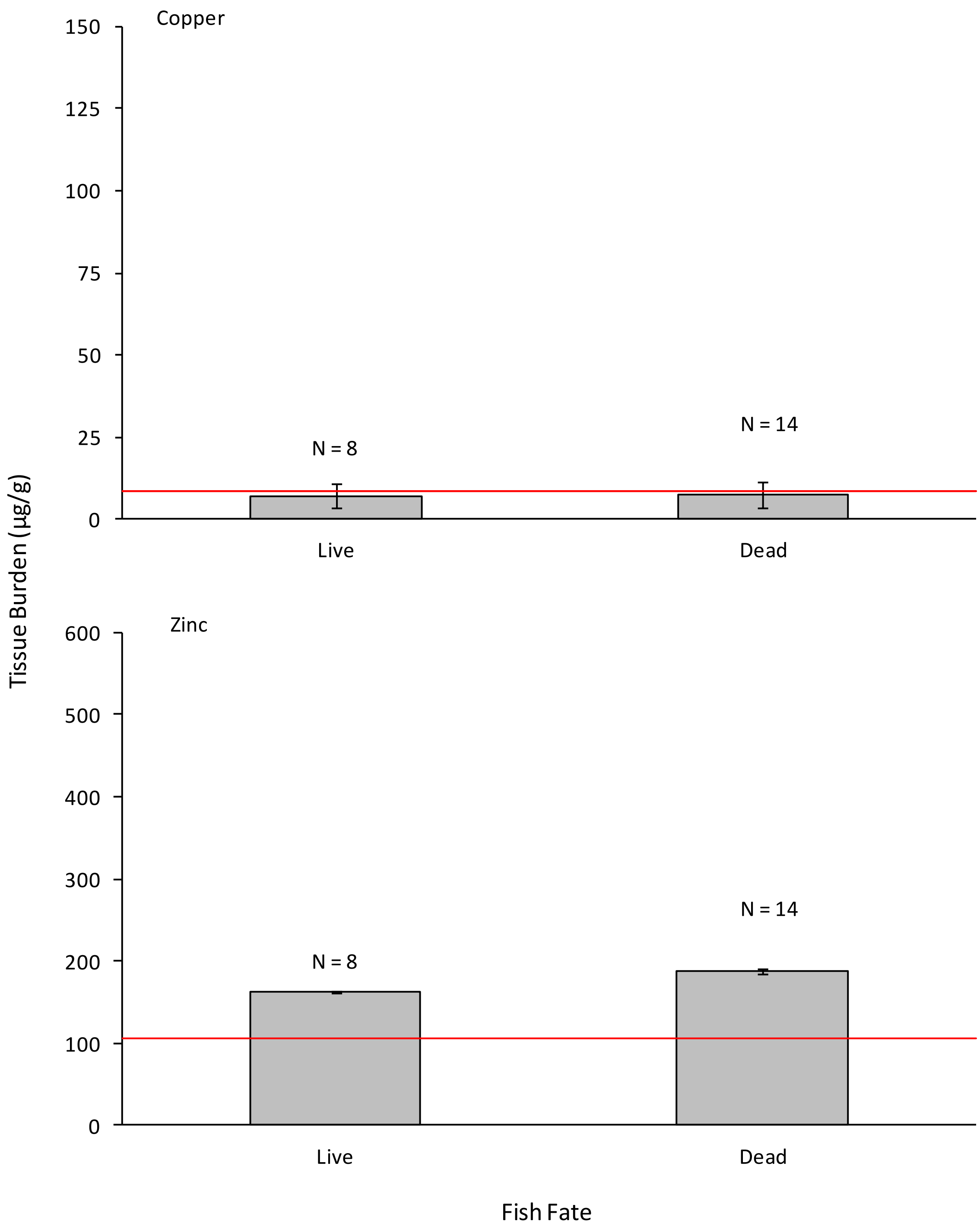

Figure 39. Copper and zinc tissue burdens for live fish versus dead fish at Lil Black in Garrison, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

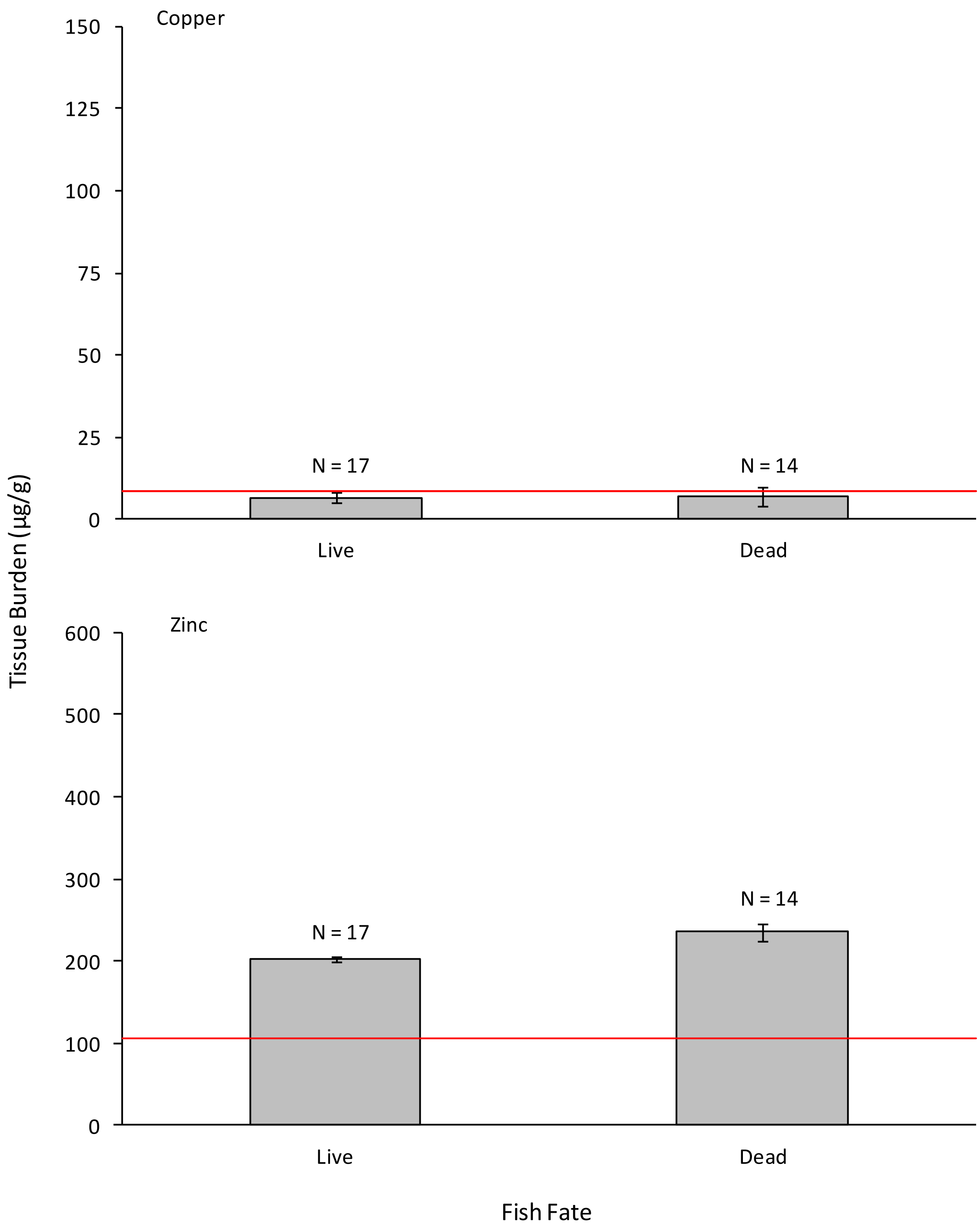

Figure 40. Copper and zinc tissue burdens for live fish versus dead fish at Flint in Drummond, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

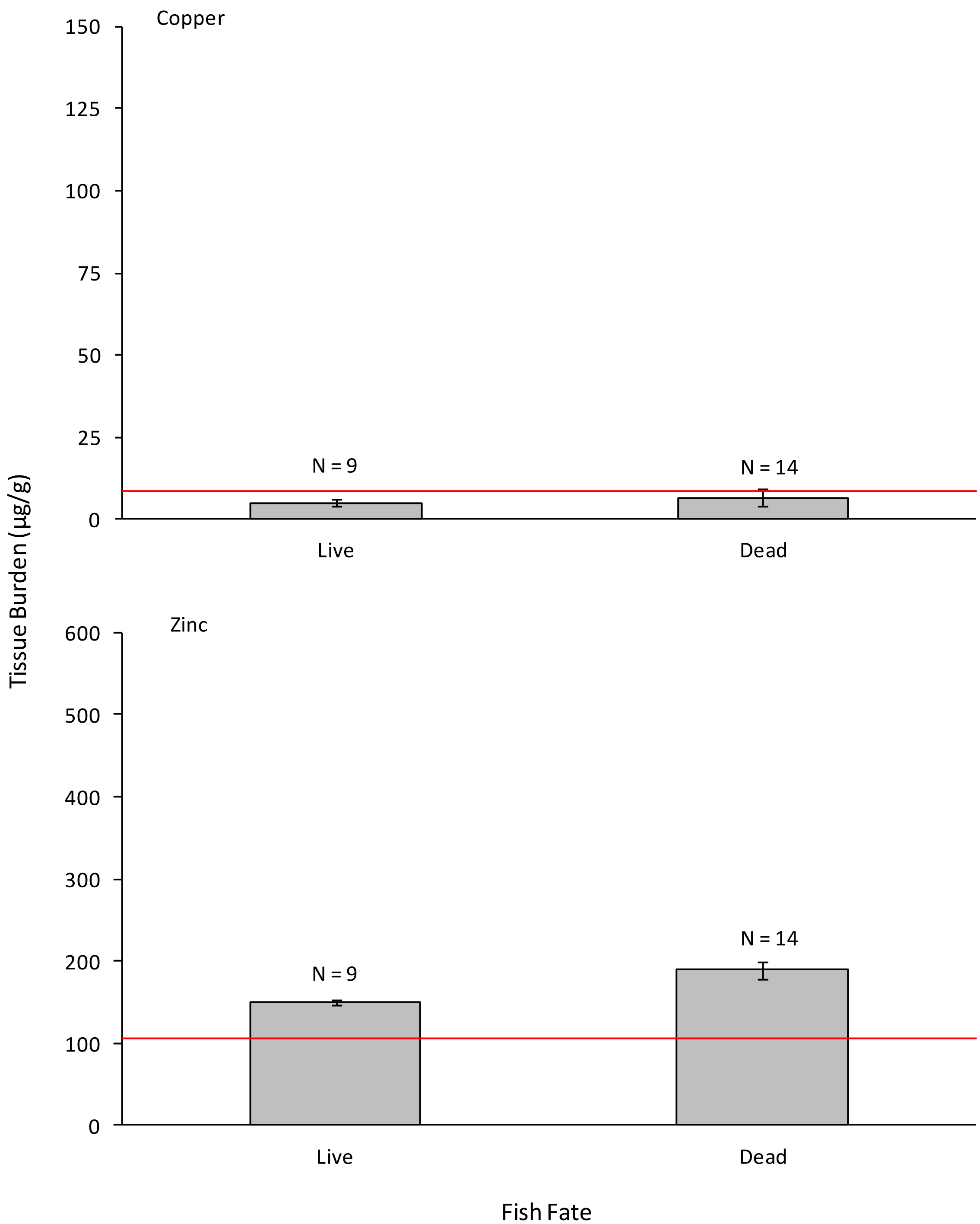

Figure 41. Copper and zinc tissue burdens for live fish versus dead fish at Clinton Spring in Clinton, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

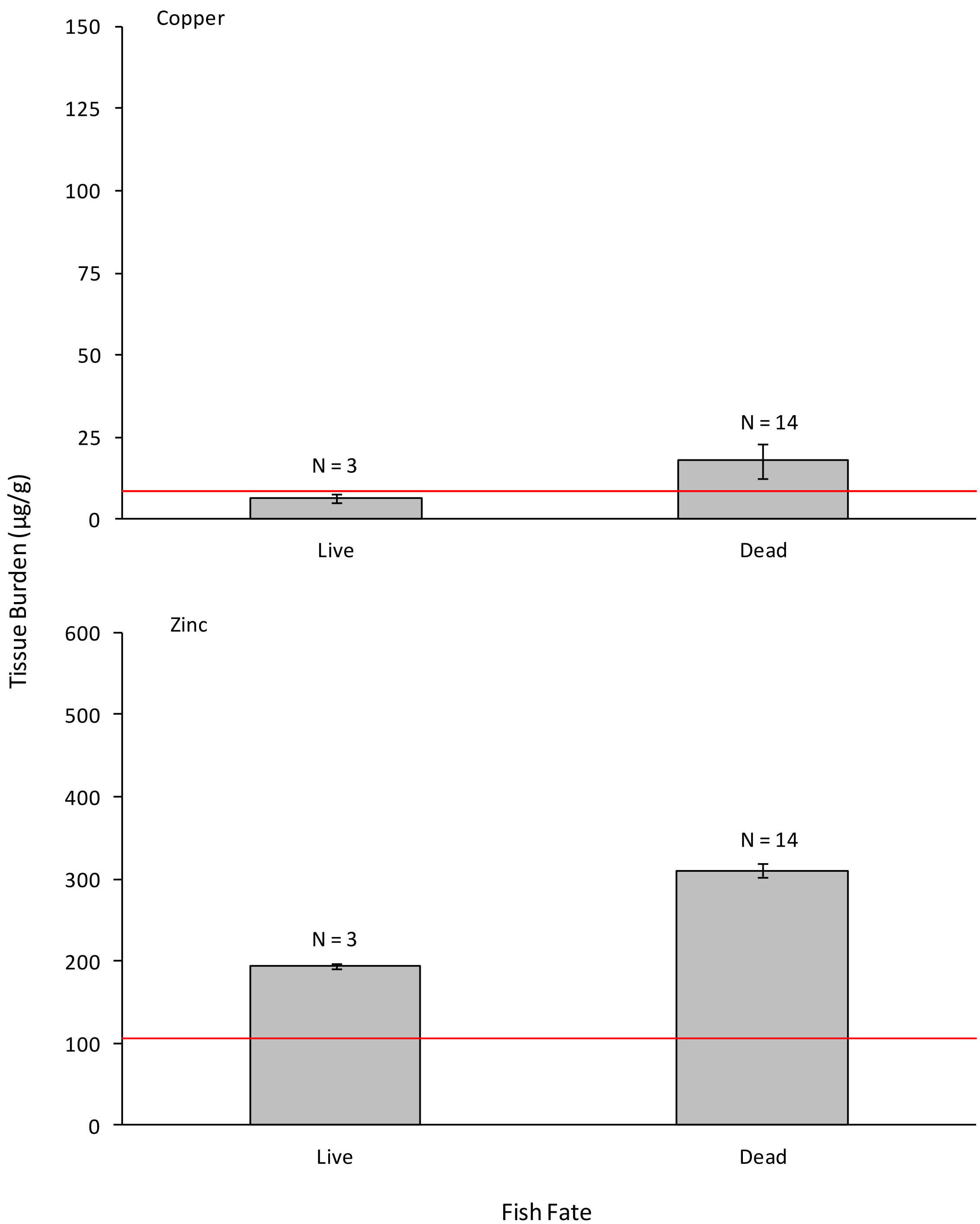

Figure 42. Copper and zinc tissue burdens for live fish versus dead fish at Turah in Turah, Montana. Both panels display individual samples (copper in the top panel and zinc in the bottom panel) with 95% confidence intervals. The red line in each panel indicates the minimum effect threshold identified for salmonids. The sample sizes in each panel represent the number of mortalities that were individually sampled (NA = 0) for April through August. The final sample sizes represent 14 individual samples of fish alive at the end of the field season.

Comparisons

Comparisons were conducted graphically between control sites and treatment sites, between upstream of construction sites and downstream of construction sites, and between upper river sites, middle river sites, and lower river sites regarding tissues metals burdens and number of mortalities for 2013 data. Previous years' data (2011 and 2012) are included as an appendix for this report (Appendix II) so that possible trends may be discussed. It is important to note that tissue metals burdens data collected in 2013 consisted of individual samples that allowed for variance calculations whereas in the past (2011 and 2012) tissue metals burdens data collected consisted of composite samples that did not allow for variance calculations.

<u>Control vs Treatment</u>

For the purposes of the analysis between control and treatment sites, Clinton Spring was not included as a control site and Mill Willow and Warm Springs were not considered treatment sites. Generally control sites were found to have lower tissue burdens than treatment sites (Figure 43). There were greater differences in copper tissue burdens between control sites and treatment sites than zinc tissue burdens between control sites and treatment sites (Figure 43). The largest differences in tissue metals burdens values between control and treatment sites appeared to occur during the months of May and July. These differences in tissue metals burdens in May and July appeared to correspond with increased mortality observed during these months (Figure 43).

<u>Upstream Construction vs Downstream Construction</u>

For the purposes of the analysis, sites located above and below the Phase 1 construction area near Warm Springs, Montana were compared. The Warm Springs site was considered above the construction area and the Galen site was considered downstream of the construction area for 2011 and 2012 and the Silver Bow site was considered above the construction area and the Galen Left site was considered downstream of the construction area for 2013. The control sites were analyzed separately. Generally upstream sites were found to have lower copper tissue burdens than downstream sites and the opposite was true for zinc burdens (Figure 44). There were greater differences in copper tissue burdens between upstream sites and downstream sites than zinc tissue burdens (Figure 44). The greatest differences in tissue metals burdens values between upstream construction and downstream construction sites occurred during the months of May, June, and July. Mortality did not necessarily correspond with these time periods (Figure 44).

<u>Upper River vs Middle River vs Lower River</u>

For the purposes of the analysis between upper river, middle river, and lower river sites, Turah was considered a lower river site, U/S Lil Black and Deer Lodge were considered middle river sites, and Galen Left, Silver Bow, and Pond 2 were considered upper river sites. Controls were analyzed by themselves. Mill Willow and Warm Springs were not included in this analysis. Generally, upper sites were found to have lower copper tissue burdens than middle sites and lower sites, with middle sites having the highest copper tissue burdens (Figure 45).

Zinc tissue burdens were more variable in terms of which sites had the highest or lowest values (Figure 45). There were greater differences in copper tissue burdens between upper sites and lower sites than zinc tissue burdens (Figure 45). The largest differences in tissue metals burdens values between upper, middle, and lower sites appeared to occur during the months of May and July for copper and August for zinc. These differences in tissue metals burdens did not necessarily correspond with increased mortality with the highest mortality being observed at the upper sites in 2013 (Figure 45).

Live Fish vs Dead Fish

Overall, there appeared to be differences between live fish and dead fish in terms of metals tissue burdens (Figures 32-42). It is important to note that Lil Black was likely the best control site in terms of tissue metals burdens. Tissue metals burdens values stayed similar throughout the entire study at Lil Black as we would expect because fish were fed the same food in the hatchery as they were during the study. The results of the logistic regression performed from tissue metals burdens of sampled fish showed a statically significant difference between the amount of copper tissue burdens live fish contained and the amount of copper tissue burdens dead fish contained ($p = < 0.001$). Figure 46 displays the results of the logistic regression graphically.

Temperature and Metals Burdens Influence

We also compared the influence of both temperature and metals tissue burdens to assess the effects of both of these variables (Figure 47). Graphically, there were different sample sizes in each quadrant, with Q1 (temperature-related mortality) having the most samples (N = 18) and Q4 (unexplained mortality) having the least samples (N=5) (Figure 47). Q2 (combined temperature and tissue burden-related mortality had a sample size of 10 and Q3 (tissue burden-related mortality) had a sample size of 13 (Figure 47). Pond 2 had 12 mortalities in Q1, 3 mortalities in Q2, 4 mortalities in Q3, and 2 mortalities in Q4. Silver Bow had 3 mortalities in Q1, 1 mortality in Q2, 5 mortalities in Q3, and 0 mortalities in Q4. Galen Left had 2 mortalities in Q1, 1 mortality in Q2, 1 mortality in Q3, and 2 mortalities in Q4. Deer Lodge had 1 mortality in Q1, 2 mortalities in Q2, 2 mortalities in Q3, and 1 mortality in Q4. U/S Lil Black had 0 mortalities in Q1, 3 mortalities in Q2, 1 mortality in Q3, and 0 mortalities in Q4. This graph appears to indicate that both temperature and metals burdens likely affect the caged fish mortality observed in 2013.

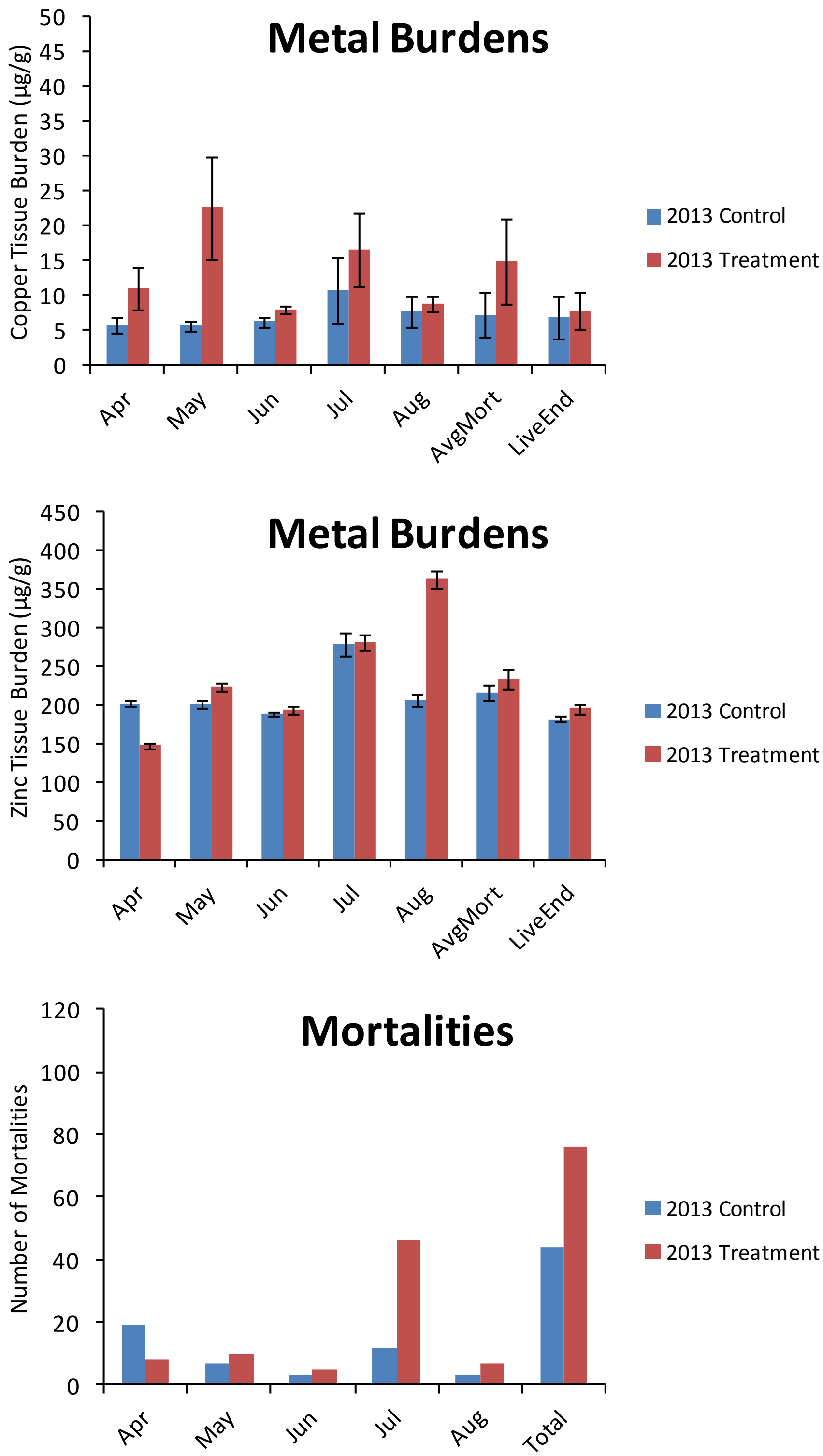

Figure 43. Comparisons between control and treatment sites' tissue metals burdens and number of mortalities by month. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2013 field season and the last column of the mortalities figure represents the total number of mortalities during the 2013 field season. Metals burdens figures display 95% confidence intervals.

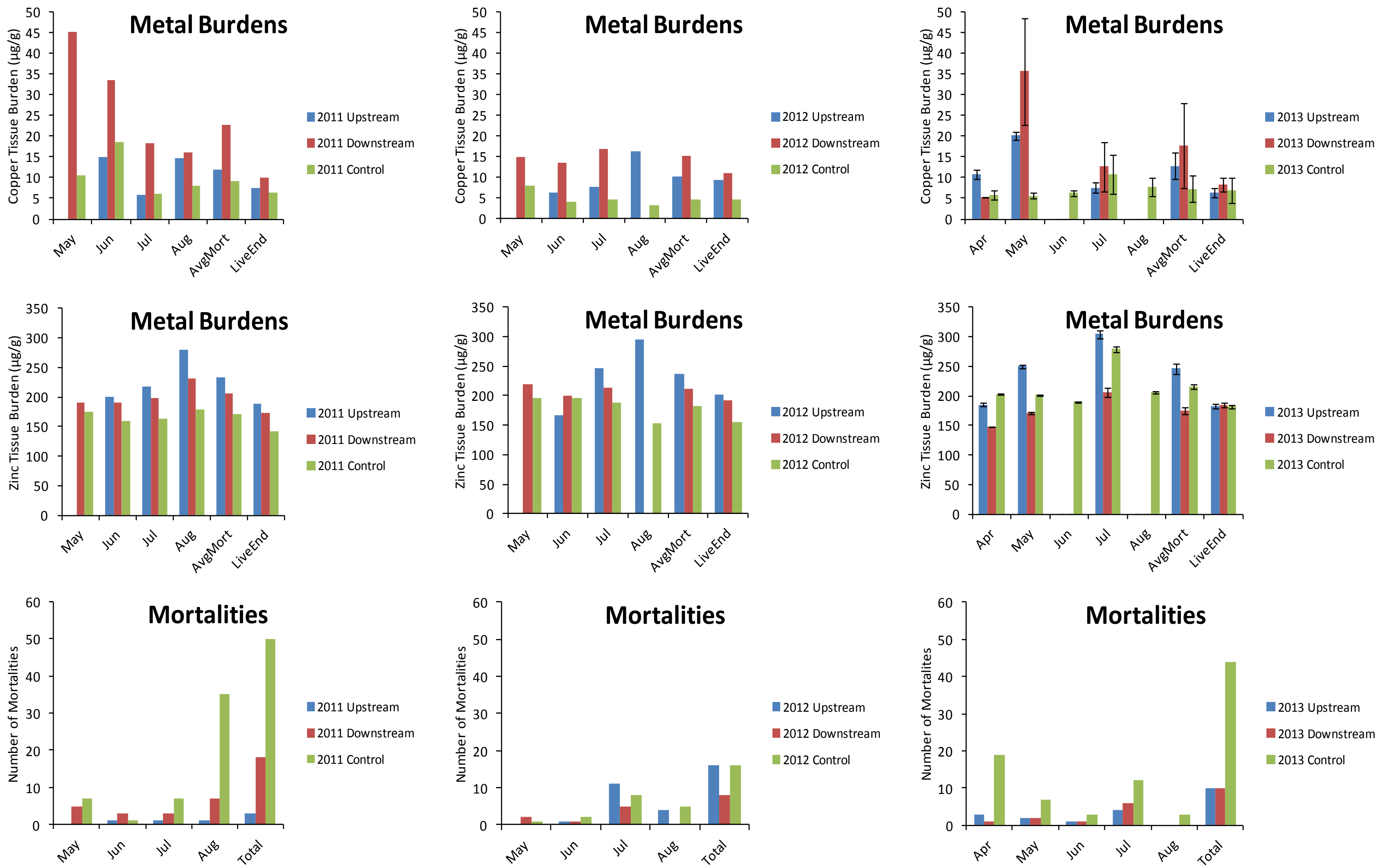

Figure 44. Comparisons between upstream construction and downstream construction sites' tissue metals burdens and number of mortalities by month and year. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2013 field season and the last column of the mortalities figure represents the total number of mortalities during the 2011-2013 field seasons. Metals burdens figures for 2013 display 95% confidence intervals.

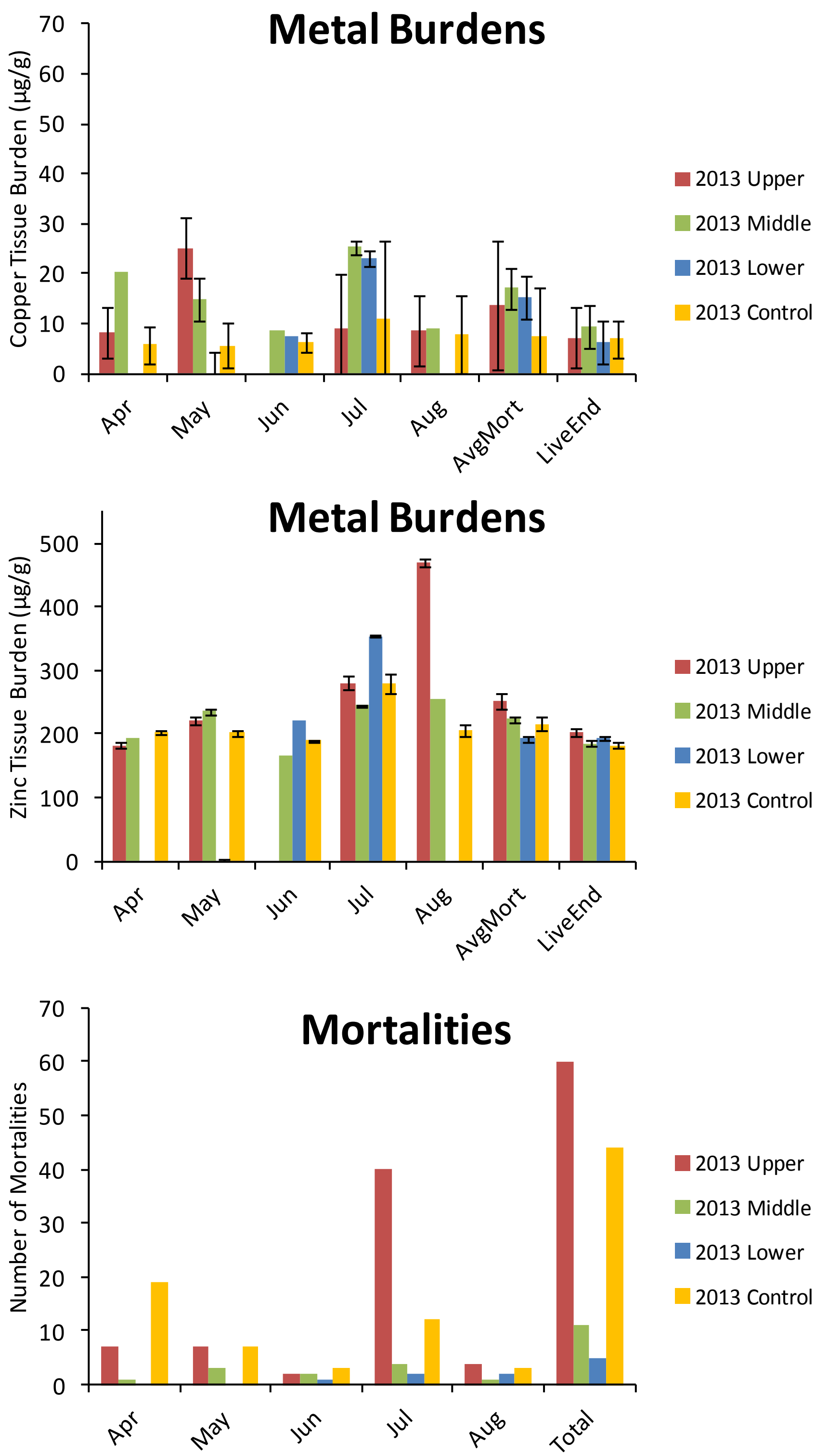

Figure 45. Comparisons between upper, middle, and lower sites' tissue metals burdens and number of mortalities by month. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2013 field season and the last column of the mortalities figure represents the total number of mortalities during the 2013 field season. Metals burdens figures display 95% confidence intervals.

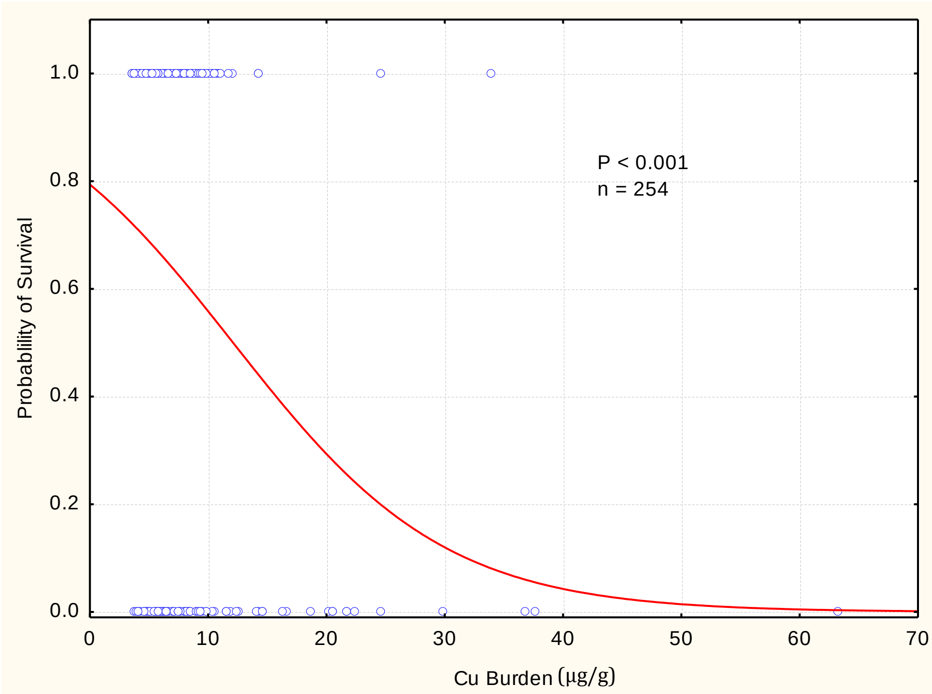

Figure 46. Results of logistic regression performed on probability of survival given copper tissue burden on fish sampled from the 2013 field season. Live fish are represented by 1 and dead fish are represented by 0.

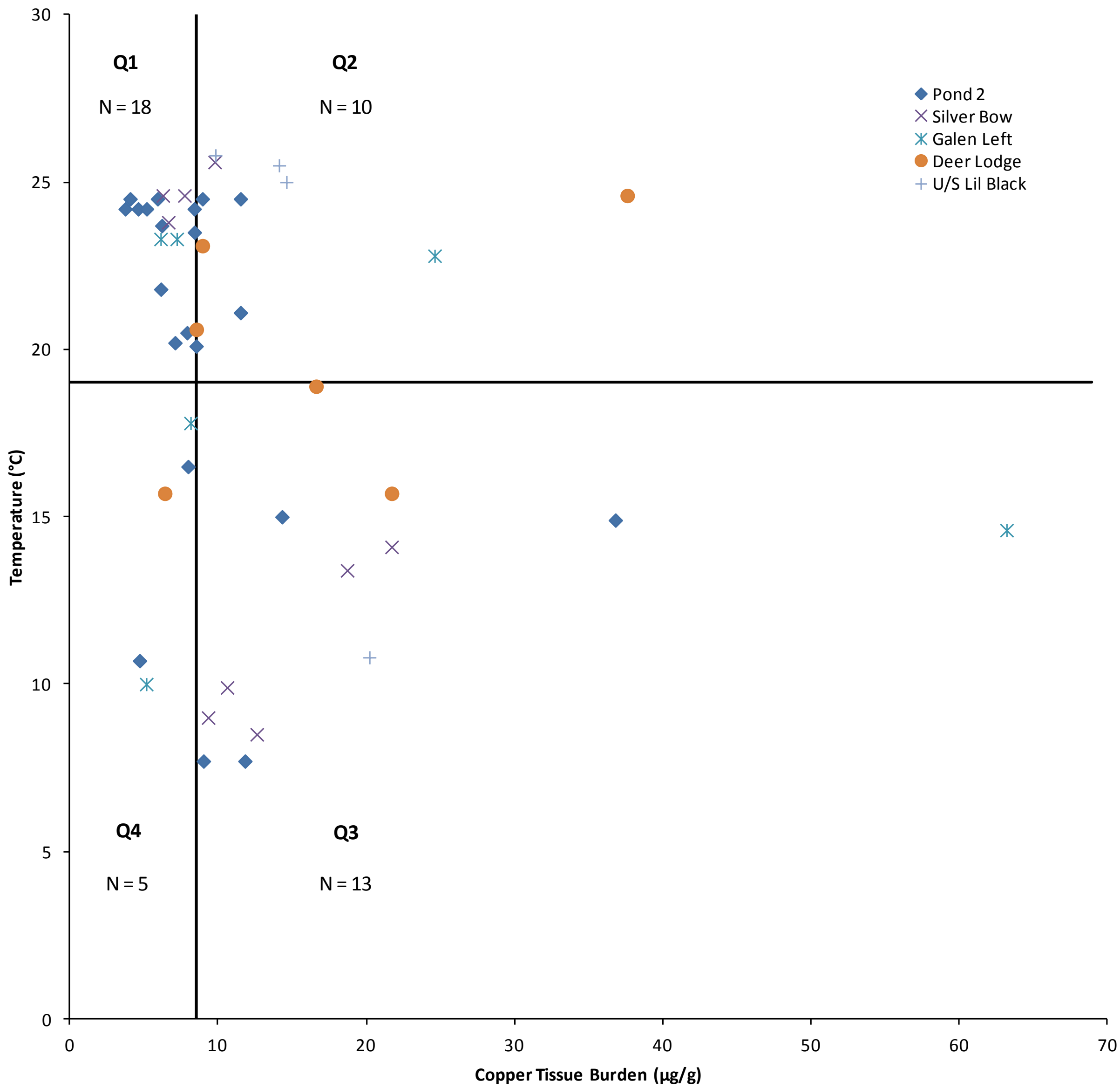

Figure 47. Quadrant analysis of fish mortalities from treatment sites in main area of concern (treatment sites from Pond 2 downstream to U/S Lil Black). The vertical line represents copper minimum effect threshold. The horizontal line represents the upper critical temperature threshold for brown trout. Quadrant one (Q1) contains fish mortalities related to water temperature, quadrant two (Q2) contains fish mortalities related to a combination of water temperature and copper tissue metals burdens, quadrant three (Q3) contains fish mortalities related to copper tissue metals burdens, and quadrant 4 (Q4) contains fish mortalities related to unknown causes. Sample sizes are indicated in each quadrant by N =.

Water Contaminants

Chronic freshwater ALS values for metals in surface water are evaluated based upon the analysis of samples following a total recoverable method (Atkins 2012; MTDEQ 2012); therefore discussion of water sampling results will focus on total recoverable levels. Dissolved metals concentrations generally followed the same trends as total recoverable concentrations. Ammonia nitrogen (NH_3-N) was never detected at any of the cage sites in 2013 with these sampling methods.

<u>Main Events</u>

In 2013, total recoverable arsenic concentration was lower at control sites (mean = 0.011 mg/L; SD = 0.005) than at mainstem treatment sites (mean = 0.022 mg/L; SD = 0.009). Total recoverable arsenic did not exceed the chronic ALS values (Figure 48). Overall, total recoverable arsenic concentrations at the mainstem sites were similar at all sites, with the exception of Warm Springs and Turah. Warm Springs and Turah showed lower arsenic concentrations than other mainstem treatment sites.

In 2013, total recoverable cadmium concentration was higher at control sites (mean = 0.00038 mg/L; SD = 0.00044) than at mainstem treatment sites (mean = 0.00023 mg/L; SD = 0.00011). Total recoverable cadmium concentrations in 2013 exceeded chronic ALS values at least once at one control site (Clinton Spring), and at least once at one mainstem treatment site (U/S Lil Black) (Figure 48). Total recoverable cadmium concentration at the mainstem treatment sites in 2013 was highest at U/S Lil Black and decreased at sites upstream and downstream from this site, with the exception of the samples collected on July 18 where the only detectable cadmium concentration at the treatment sites was observed at Deer Lodge.

In 2013, total recoverable copper concentration was lower at control sites (mean = 0.00275 mg/L; SD = 0.00128) than at mainstem treatment sites (mean = 0.02633 mg/L; SD = 0.02465). Total recoverable copper exceeded the chronic ALS at least once at eight of nine mainstem treatment sites (Pond 2 was exception) and total recoverable copper never exceeded the chronic ALS at a control site (Figure 50). In 2013, overall total recoverable copper at the mainstem sites was highest at Deer Lodge and decreased at sites upstream and downstream of this site. However, on May 28, 2013 the highest total recoverable copper was observed at U/S Lil Black and decreased at sites upstream and downstream of that site.

In 2013, total recoverable lead concentration was higher at control sites (mean = 0.0047 mg/L; SD = 0.0043) than at mainstem treatment sites (mean = 0.0037 mg/L; SD = 0.0036). It is important to note that control sites were high in lead due to Flint having some of the highest lead readings of any site. Total recoverable lead concentrations in 2013 exceeded the chronic ALS value at four mainstem treatment sites (Warm Springs, Galen Left, Deer Lodge, and U/S Lil Black) and one tributary control site (Flint) (Figure 51). On May 28, 2013 total recoverable lead at the mainstem sites was highest at U/S Lil Black and decreased at sites upstream and downstream of this site. On June 14, 2013 total recoverable lead at the mainstem sites was highest at Galen Left and decreased at sites upstream and downstream of this site, and on July 18, 2013 total recoverable lead at the mainstem sites was highest at Deer Lodge and decreased at sites upstream and downstream of this site.

In 2013, total recoverable zinc concentration was lower at control sites (mean = 0.024 mg/L; SD = 0.01342) than at mainstem treatment sites (mean = 0.03688 mg/L; SD = 0.02301).

Total recoverable zinc concentration in 2013 did not exceed the chronic ALS value at any of the treatment or control sites (Figure 52). On May 28, 2013, total recoverable zinc at the mainstem sites was highest at U/S Lil Black and decreased at sites upstream and downstream of this site. On June 14, 2013 total recoverable zinc at the mainstem sites was highest at Galen Left, Deer Lodge, and U/S Lil Black and decreased at sites upstream and downstream of these sites, and on July 18, 2013 total recoverable lead at the mainstem sites was highest at Deer Lodge and decreased at sites upstream and downstream of this site.

Rain Events

In 2013, rain event samples did not have total recoverable concentrations of arsenic, cadmium, and zinc that exceeded chronic ALS values. Total recoverable concentrations of copper exceeded chronic ALS values at three sites: Turah (June 20), U/S Lil Black (June 20), and Deer Lodge (June 24, August 1, and August 29) in 2013. The highest total recoverable concentrations of copper were 0.057 mg/L at Deer Lodge on August 1 and 0.038 mg/L at Turah on June 20. Total recoverable concentrations of lead exceeded chronic ALS values at Turah on June 20, 2013. The highest total recoverable concentration of lead was 0.0074 mg/L at Turah on June 20. Figures displaying metals compliance ratios for rain events were completed and are included as an appendix for this report (Appendix III).

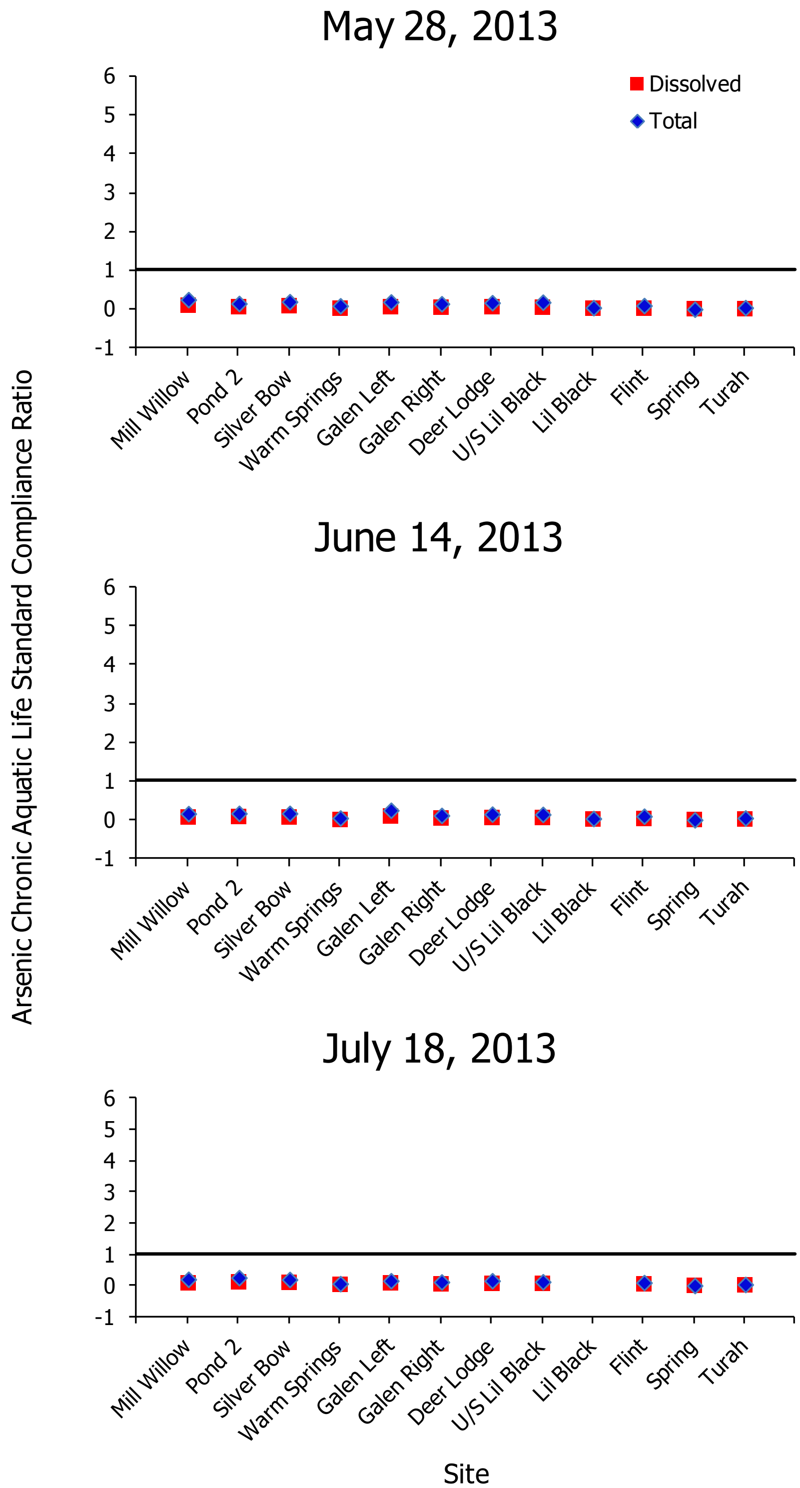

Figure 48. Arsenic compliance ratios at the cage sites in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing arsenic concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate arsenic levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

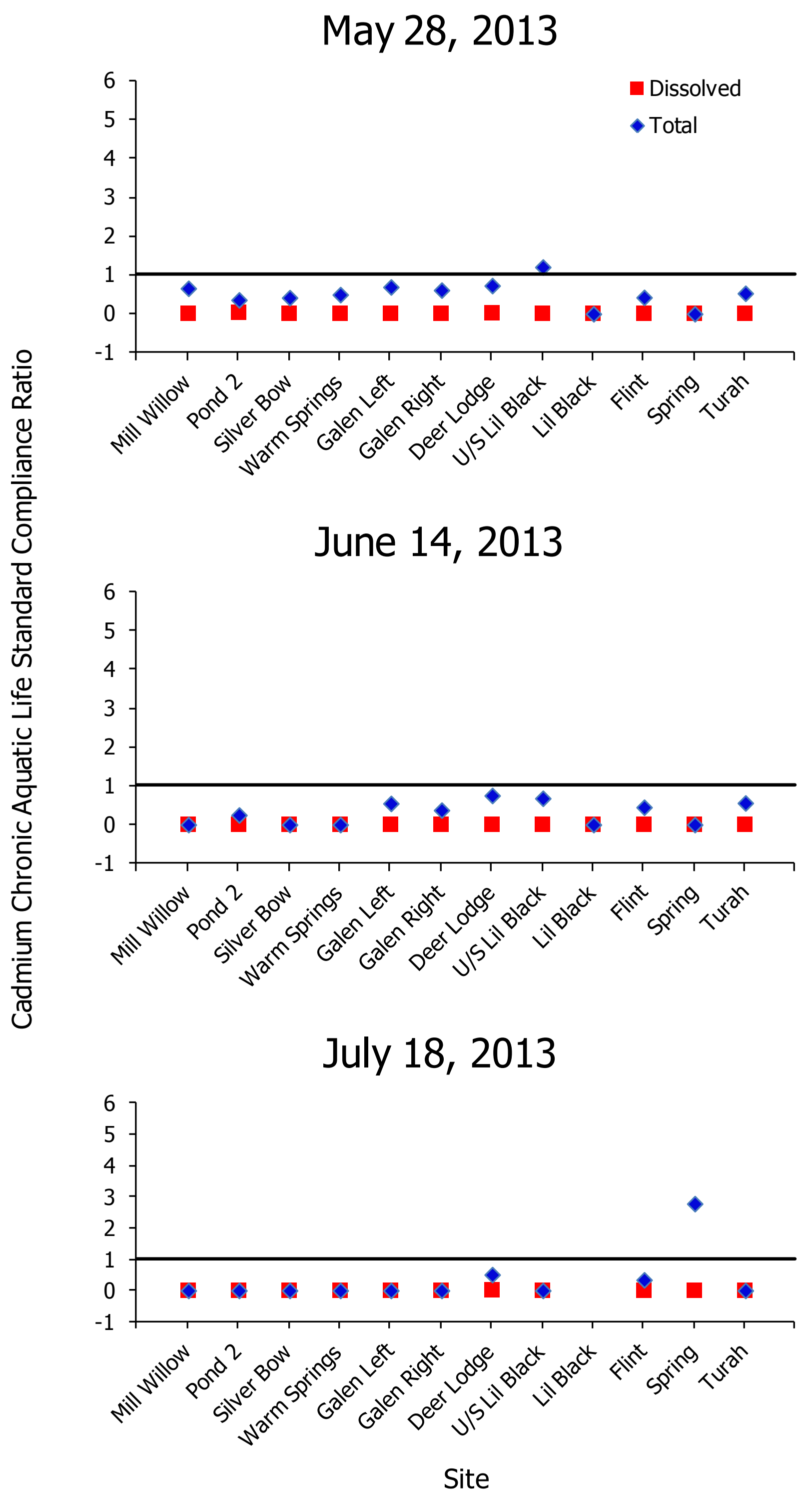

Figure 49. Cadmium compliance ratios at the cage sites in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing cadmium concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate cadmium levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

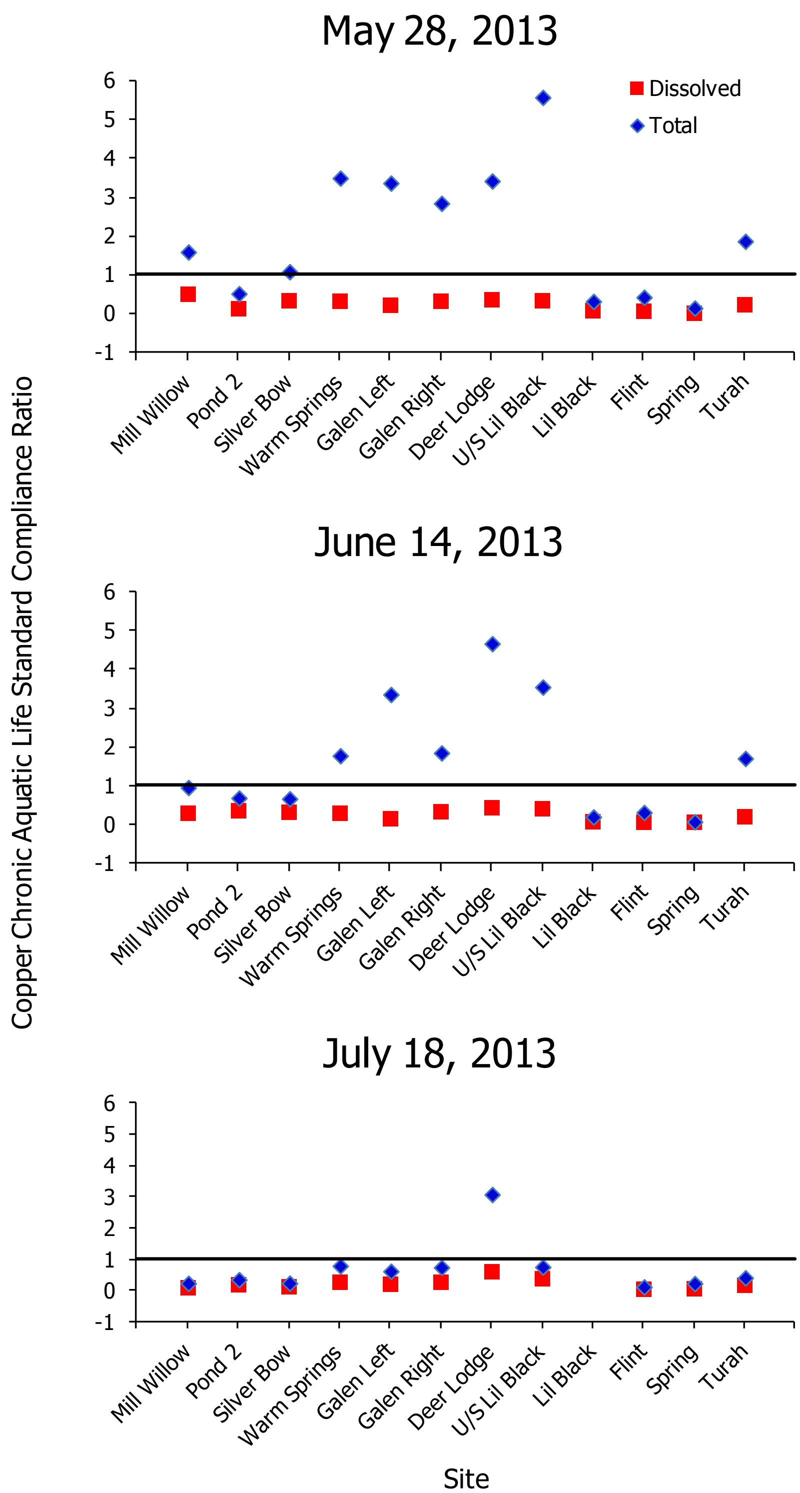

Figure 50. Copper compliance ratios at the cage sites in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing copper concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate copper levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

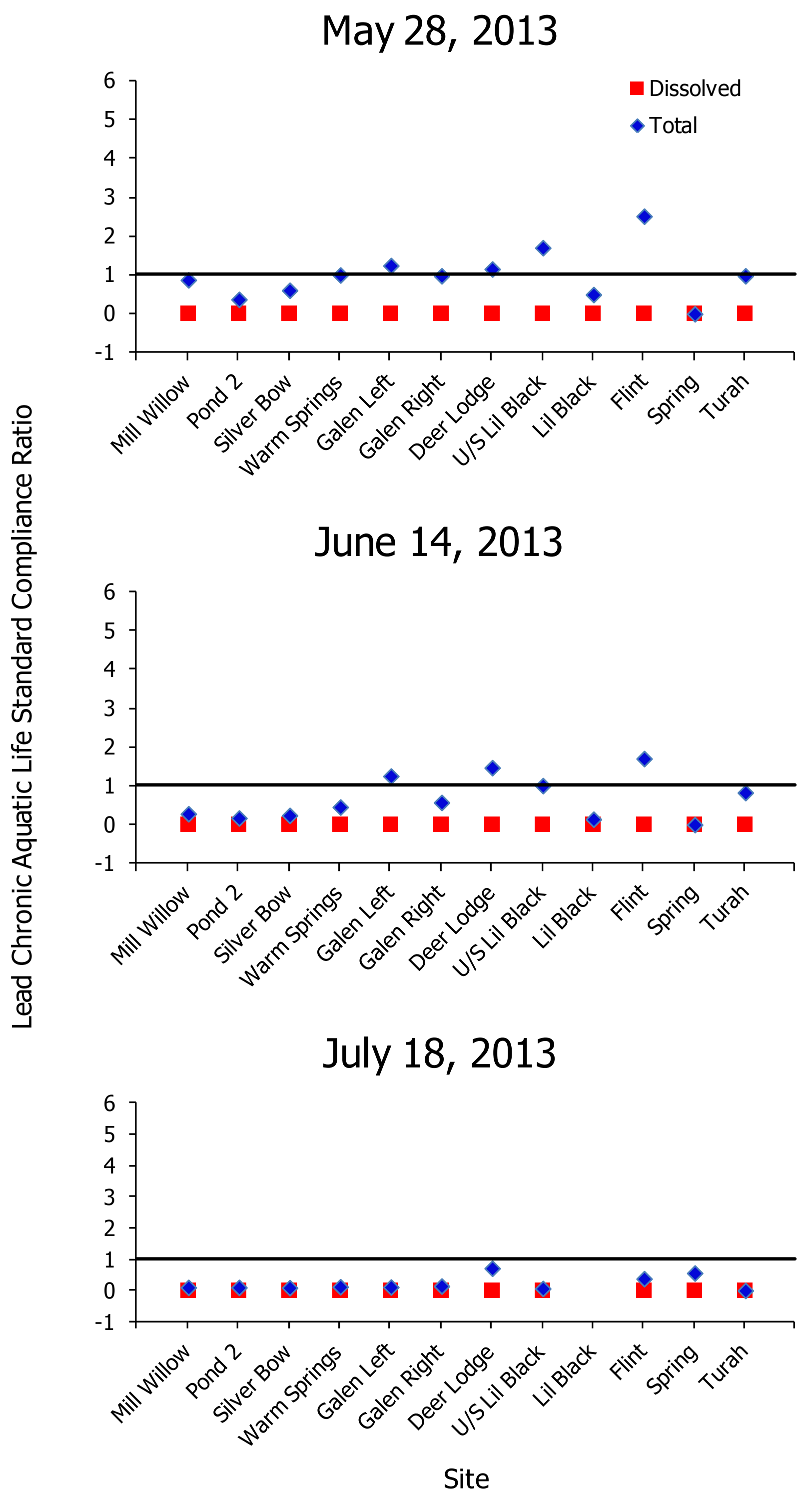

Figure 51. Lead compliance ratios at the cage sites in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing lead concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate lead levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

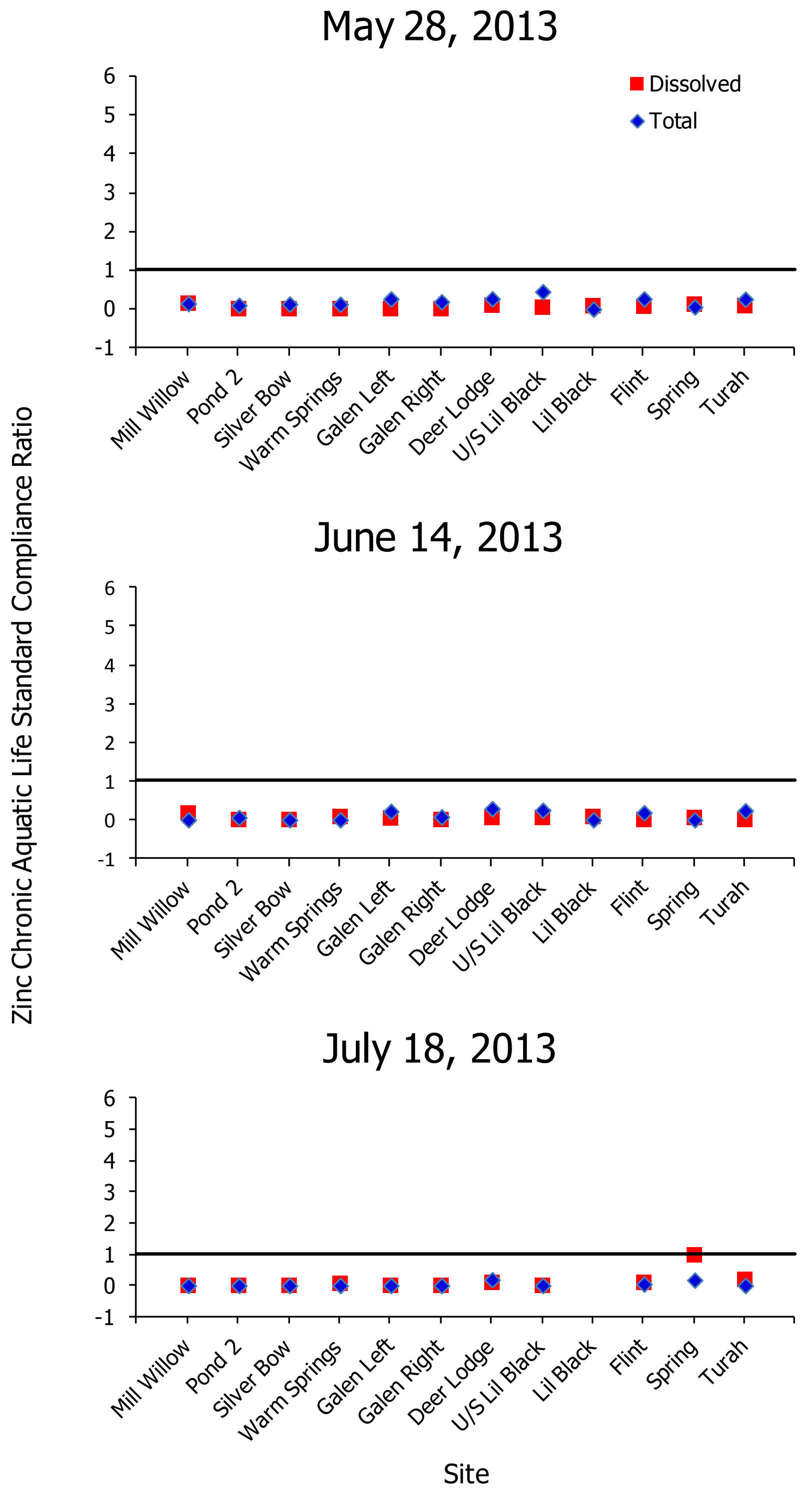

Figure 52. Zinc compliance ratios at the cage sites in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing zinc concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate zinc levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

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Water Quality

Water quality parameters were recorded on continuously recording Hydrolab ® MS5 water quality probes at Mill Willow, at Pond 2, at Silver Bow, at Galen, and at U/S Lil Black and the results are outlined in the following sections.

pH

Elevated pH was observed at the Pond 2 and at Silver Bow sites (Figure 53). Extended exposure to pH > 9 may be harmful to trout (Colt et al. 1979). Mean daily values for pH exceeded 9 in early April, late May, June, July, and August at Pond 2, and at Silver Bow in early and late June, middle and late July, and early August 2013. In contrast, mean daily pH at the remaining mainstem sites with probes deployed did not exceed 9 and generally varied from 7.5 to 8.5 (Figure 53), which is considered within the ranges suitable for trout (Colt et al. 1979). In comparison, pH was measured with a handheld probe at the control sites with the mean ranging from 7.5 to 8.5 (Figure 53). In addition to the high pH values measured later in the season, Pond 2 mean daily values for pH were approximately 7 for the first week of May which were the lowest recorded pH values observed throughout the study area.

Specific Conductivity

Specific conductivity is a measure of the ability of water to conduct electricity and can be used as a relative measure of water quality. Specific conductivity typically varies from 10 to 1000 µS/cm, but may exceed 1000 µS/cm in polluted waters or waters receiving large quantities of land runoff (Chapman 1996). Mean daily specific conductivity at all sites was within normal ranges in 2013 (Figure 54). All sites experienced specific conductivities ranging from 150 to 600 µS/cm, with the exception of U/S Lil Black which experienced a specific conductivity range from 300 to 650 µS/cm. All sites appeared to follow the same trends with regards to specific conductivity and these results were also seen with the handheld probe, with the exception of U/S Lil Black (Figure 54).

Luminescent Dissolved Oxygen

The freshwater ALS one day minimum for dissolved oxygen for fish > 30 days post-hatch in the Clark Fork River is 4.0 mg/L (MTDEQ 2012b). Mean daily dissolved oxygen levels were below this threshold for two days in late June/early July 2013 at U/S Lil Black. Reduced dissolved oxygen levels in late June/early July at U/S Lil Black in 2013 likely represent actual oxygen depression rather than probe failure as the oxygen level rebounds after dropping whereas probe failure generally results in dropping oxygen levels that do not rebound. None of the depressed oxygen levels coincided with increased mortality. The overall trend in mean daily dissolved oxygen levels was values > 10.0 mg/L at all sites at the beginning of each field season that gradually decreased to values of approximately 6 to 8 mg/L by the end of the year (Figure 55). Handheld probe data displayed the same overall trend in mean daily dissolved oxygen levels with dissolved oxygen levels gradually decreasing throughout the field season, but values began at > 11.0 mg/L at all sites and gradually decreased to values of approximately 8.5 to 12 mg/L (Figure 55). It is important to note that the dissolved oxygen sensor on the handheld probe used relies on moving water in order to properly calculate dissolved oxygen levels and improper deployment can cause inaccurate readings.

Total Ammonia

Total ammonia ($NH_4 + NH_3$) was measured at Pond 2, Silver Bow, and Galen in 2013. However, total ammonia levels recorded were not consistent with data collected in previous years. The precision with which the Hydrolab ® MS5 records total ammonia levels has been questionable in the past (T. Selch, Montana, Fish, Wildlife, and Parks, personal communication, 2014). Water samples collected during the 2013 field season by both the Montana Department of Environmental Quality and the Montana Fish, Wildlife, and Parks closely resembled one another and the previous years' recorded levels but were quite different than those recorded by the Hydrolabs in 2013. For these reasons total ammonia levels recorded in 2013 from the Hydrolab ® MS5 were not presented or included in this report.

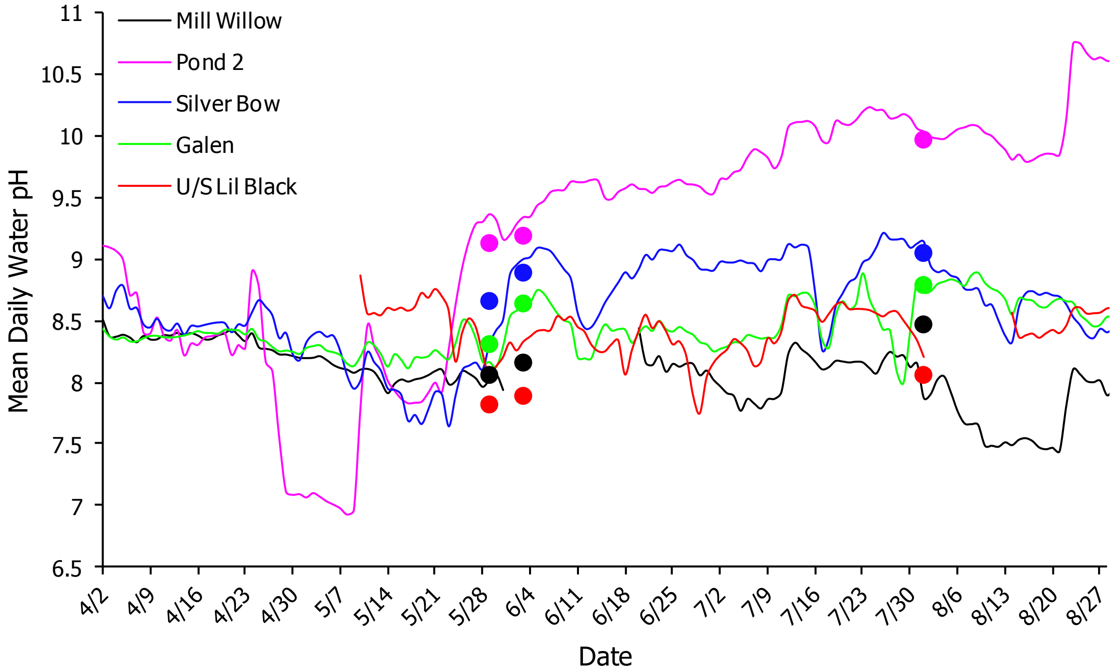

Figure 53. Mean daily water pH at sites with probes deployed in 2013. Lines represent hydrolab data and circles represent handheld multiprobe data.

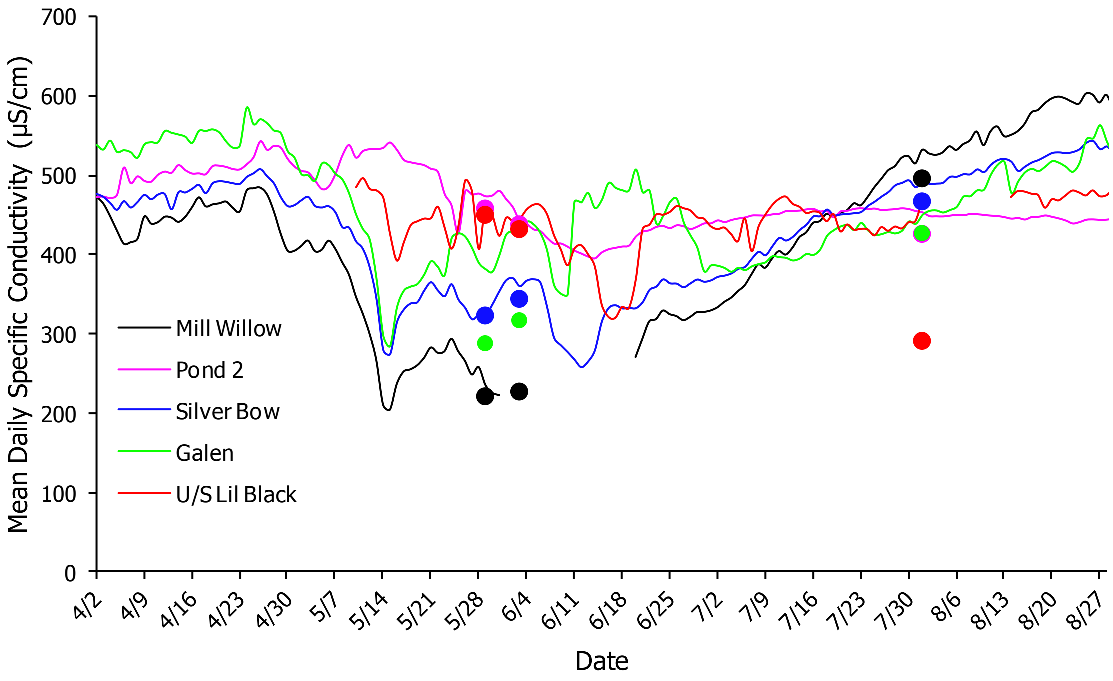

Figure 54. Mean daily specific conductivity at sites with probes deployed in 2013. Lines represent hydrolab data and circles represent handheld multiprobe data.

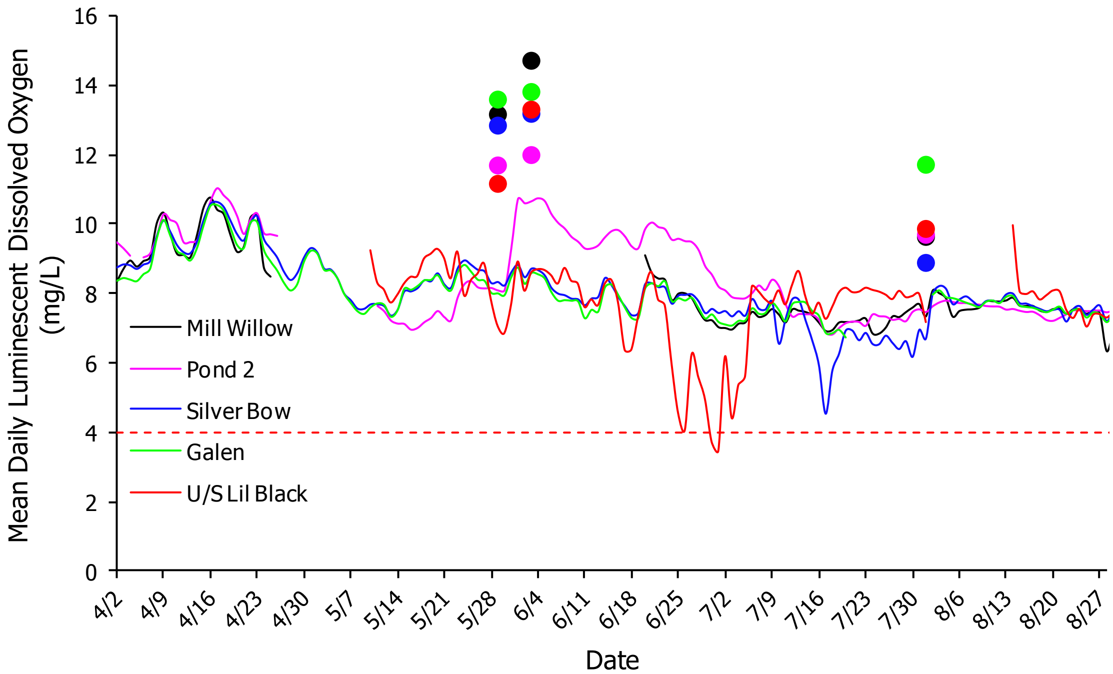

Figure 55. Mean daily luminescent dissolved oxygen at sites with probes deployed in 2013. Lines represent hydrolab data and circles represent handheld multiprobe data. The red dashed horizontal line denotes the freshwater ALS one day minimum.

Discussion

Trout Population Monitoring

Trout population estimates have generally increased in recent years compared to estimates published in Lindstrom (2011) and previous years' estimates included in this report. In particular, brown trout densities appeared to increase throughout the Upper Clark Fork River in 2013. This trend was observed for all sections although the Bearmouth section was not substantially higher than observed in 2009. Interestingly, westslope cutthroat trout densities also improved in the Bearmouth section from 2009 through 2013. Sampling has been conducted during the same times of year and in the same sections of river, so these variables should not be confounding. Therefore, it is possible that previous better than average water years may be responsible for the improvement in trout densities in these reaches. In addition to providing current trend data, these population estimates will also serve as a baseline to compare fish populations pre- and post- remediation of the Upper Clark Fork River.

Caged Fish Study

The majority of mortality observed during the 2013 field season occurred during April, May, and July. The largest portion occurred in July as water temperatures approached or exceeded the upper critical temperature threshold for brown trout of 19.0 °C (Elliot 1994). Water temperatures exceeded the upper incipient lethal temperature for brown trout of 24.7 °C (Elliot 1994) at seven sites: Mill Willow, Pond 2, Silver Bow, Deer Lodge, U/S Lil Black, Lil Black, and Flint in 2013. This is in contrast to previous years reported by Richards et al. (2013) where only two sites exceeded the upper incipient lethal temperature for brown trout. Elevated water temperatures can stress fish to the point of influencing feeding and growth (Elliot 1994; Elliot and Hurley 2001) and also make fish more susceptible to other environmental stressors and diseases (Wahli et al. 2002; Hari et al. 2006; Jonsson and Jonsson 2009). High water temperatures also increase susceptibility to metals exposure through increased respiration (Sorensen 1991).

Tissue metals burdens have been shown to compromise trout health (Woodward et al. 1995a; Farag et al. 1999, 2003). Results of tissue metals burden analyses indicate effects from copper in the Clark Fork River. Whole body copper burdens were found to have an effect on rainbow trout at 8.57 µg/g after exposure for 60 days (Marr et al. 1996). In 2013, whole body copper burdens of dead fish exceeded this value at all mainstem sites but never at the control sites. This value was also exceeded by all mainstem sites and at least one control site in 2011 and 2012. However, whole body copper burdens of live fish did not exceed the minimum effect threshold for copper, with the exception of Deer Lodge. Contrary to copper, results of zinc tissue burdens analyses indicate no effects from the Clark Fork River. This is likely because fish supplied for this study were above concentrations shown to reduce growth in young salmonids prior to cage placement. In addition, difficulty in respiration, decreased feeding, and decreased growth were observed in rainbow trout with average whole body zinc burdens varying from 105.09 to 178.66 µg/g after experimental exposure and fish supplied for this study exceeded these values prior to cage placement (Gundogdu and Erdem 2008). The lower value in the range for whole body zinc burden from that study was always surpassed at all sites whether the fish were alive or dead in 2013. In previous years, the minimum effect threshold for zinc was surpassed at all sites with the exception of Rock Creek in 2011 (Richards et al. 2013). Studies have suggested that dietary exposure to high zinc levels increases zinc tolerance (e.g., Wekell et al. 1983). This may explain why there were never large mortality events across sites even though fish used in this study were high in zinc when delivered from the hatchery and continued to feed on hatchery food high in zinc throughout the study (T. Selch, Montana, Fish, Wildlife, and Parks, personal communication, 2014).

pH exceeded values tolerable to trout at Pond 2 and Silver Bow (Colt et al. 1979) in 2013. Of these two sites, only Pond 2 experienced these values for an extended period of time. However Pond 2 also experienced the highest number of mortalities during the 2013 season. Similar trends were observed in 2011 and 2012 with both the Silver Bow and Galen sites experiencing pH levels that exceeded tolerable levels to trout. In addition to direct adverse effects of high pH, these conditions may also influence toxicity of metals (Couture and Pyle 2012). Studies have shown that copper and zinc are more toxic to steelhead trout (*Salmo gairdneri*) at high pH values (Cusimano et al. 1986). In addition to influencing the toxicity of metals, high pH results in a higher concentration of toxic NH_3 in the total ammonia makeup (Emerson et al. 1975). Average daily pH was above the tolerable level from May 26 to the end of the study in 2013 at Pond 2. Mill Willow is located upstream of Pond 2 and displayed lower pH than Pond 2. pH generally decreased at sites downstream of Pond 2 and the decrease was more apparent the further downstream a site was from Pond 2. This is likely due to the addition of lime to the settling ponds upstream of these sites in attempt to decrease water acidity and reduce metals toxicity leaving Warm Springs Ponds, as well as the large amount of photosynthesis occurring in the ponds. An unintended consequence to the addition of lime to the settling ponds may be an increase in metals toxicity if too much lime is added to the system. This when combined with other stressors such as high water temperature, may be resulting in increased mortality at the upstream sites.

Comparisons between this study and results observed in Richards et al. (2013) yielded similar results. Generally control sites yielded lower amounts of tissue metals burdens than treatment sites for fish that died and control sites also yielded roughly the same amounts of tissue metals burdens for fish that died and fish that lived through the sample period (fish that lived are those that were sampled at the end of the study period). The actual amounts of tissue metals burdens for both control sites and treatment sites do not appear to have changed significantly over time.

In comparing sites upstream of construction and sites downstream of construction in Warm Springs, Montana, upstream construction sites generally yielded lower amounts of tissue metals burdens than downstream construction sites for fish that died. Tissue metals burdens were similar between upstream construction sites and downstream construction sites for fish that lived. Sites upstream of construction had less mortality than sites downstream of construction in 2011, but in 2012 and 2013 upstream construction sites had more mortality than downstream construction sites. The actual amounts of tissue metals burdens and number of mortalities for both upstream construction sites and downstream construction sites do not appear to have changed significantly from 2011 to 2013 suggesting that construction did not appear to negatively impact the amount of metals tissue burdens in trout residing downstream of the construction site.

Comparisons between upper river sites, middle river sites, and lower river sites yielded higher amounts of tissue metals burdens in middle river sites than lower river sites and upper river sites for dead fish in 2011 and 2012, but these differences were not as evident in 2013.

Live fish showed similar levels of tissue metals burdens between upper river, middle river, and lower river sites. Upper river sites had more mortalities than lower river sites in each year except 2011, which may be due to higher pH and temperature stressors not present at lower river sites. The actual amounts of tissue metals burdens and number of mortalities for upper river sites, middle river sites, and lower river sites do not appear to have changed significantly over time. It is not surprising that metals levels have not decreased yet as 2013 was the first year of significant remediation in the Upper Clark Fork River drainage. The lack of reduction in tissue metals burdens by both site and year may explain the consistent number of mortalities observed each year given that tissue metals burdens likely do directly impact mortality.

The observation that copper tissue burdens appear to increase between the upper and middle sites of our study area is consistent with copper concentrations observed in the Upper Clark Fork River by other researchers (Sando et al. 2013). Sando et al. (2013) concluded that suspended sediment and copper concentrations are reduced below Warm Springs Ponds by settling that occurs in the ponds. They also identified the reach from Galen to Deer Lodge as a major source of additional copper and suspended sediment to the Clark Fork River (Sando et al. 2013). Our study indicates that less copper is being taken up by fish at sites directly below the ponds likely due to a reduction in metals in this reach, but that copper uptake by fish increases at the middle sites of our study area (Deer Lodge and Upstream of the Little Blackfoot River). This increase in copper tissue burdens appears to correlate with the increased copper concentrations observed by Sando et al. (2013) between Galen and Deer Lodge. While the Warm Springs Ponds do appear to reduce copper concentrations in the section of the Clark Fork River directly downstream, we do still question whether the pond system is overall beneficial to the fishery of the Upper Clark Fork River due to other potential negative factors observed in that reach that may be caused by the pond system, such as high pH (see further discussion on page 81).

The highest mortality rates did not necessarily occur at sites with the highest water temperatures, waterborne metals concentrations, or tissue metals burdens, but rather at sites exhibiting a combination of these factors. This is likely due to a cumulative effect of environmental stressors (Kiser et al. 2010) as seen in previous years of this study (e.g., Richards et al. 2013). For example, in 2013, the chronic freshwater ALS values for copper and zinc were exceeded in water samples collected during the study, suggesting that waterborne metals may have influenced mortality in fish cages. However, metals concentrations were highest on the ascending limb and peak of the hydrograph while the majority of mortalities were on the descending limb as discharges approached or achieved base flow. Also, quadrant analysis performed in 2013 showed less unexplained fish mortality and more fish mortality corresponding to either high water temperatures, high copper tissue metals burdens, or both, with the high water temperature having more fish mortalities than any other quadrant. For example, Pond 2 experienced the most mortalities of any site, experienced the third greatest number of days above the upper critical temperature threshold for brown trout, and over half of the observations occurred in Q1 indicating temperature-related mortality. Conversely, Silver Bow experienced the second greatest number of days above the upper critical temperature threshold for brown trout and over half the mortalities occurred in Q3 indicating tissue burden-related mortality. Water temperatures experienced in western Montana during the summer of 2013 were higher than average and in combination with low water levels led to Clark Fork River fishing closures (Chaney 2013). Studies have shown fish are affected more by other stressors as temperatures rise above minimum effect thresholds (e.g., Lobón-Cerviá and Rincón 1998) suggesting that temperature that may lead to mortality when combined with other stressors, such as elevated metals concentrations. In the Clark Fork River, we feel that chronic effects of waterborne metals were exacerbated by environmental conditions such as elevated water temperature and pH, ultimately causing mortality in all years. Interestingly, low dissolved oxygen levels were not observed at most sites in 2013, with the exception of U/S Lil Black. In previous years, low dissolved oxygen levels were detected and possible synergistic effects of pH, metals, high water temperature and periods of low dissolved oxygen were suggested as reasons for the observed mortality (Richards et al. 2013).

Results indicate that mortality was statistically significantly lower than expected at all treatment sites other than Pond 2 which had significantly higher mortality and Galen Right which showed no difference in mortality. It is important to note that 2013 showed higher mortality at control sites than in previous years which affected the expected mortality values in the chi square analysis. We feel that mortality was excessively high at the control sites in 2013 and this data likely does not provide an accurate representation of a good control mortality rate. A possible explanation for the very high mortality rates observed at Flint Creek and the Little Blackfoot River are the extremely low flow conditions observed at these sites in 2013. In Flint Creek, flows reached as low as 8 cfs in both May and late July/early August and in the Little Blackfoot River, flows reached as low as 16 cfs in late April/early May, July, and August. The precise mechanisms that led to the high mortality at these sites are not completely understood, but may be due to very high water temperatures and potentially low dissolved oxygen conditions caused by these extremely low flow conditions.

Overall, we offer the same explanation to these results as Richards et al. (2013). At all of the treatment sites and during all years, mortality was likely the result of the cumulative effect of many environmental stressors including increased metals concentrations and high water temperatures, although further work is needed to better understand this relationship. The cumulative effect of environmental stressors such as unsuitable pH, dissolved oxygen, and water temperature in addition to metals exposure likely explain the high mortality seen at Galen in 2011, Warm Springs in 2012, and Pond 2 in 2013 (although DO was not low at Pond 2 in 2013), and could potentially indicate a point source of poor water quality. Water quality appears to improve downstream of these sites, potentially due to inflow of clean water from tributaries, with lower than expected mortality observed downstream of Gold Creek and at Bearmouth in 2011 and 2012 and at Turah 2013.

In conclusion, multiple factors appear to be affecting survival of brown trout at some sites throughout all years of this study conducted thus far. Upstream and downstream differences in water quality may explain the spatial distribution of brown trout mortality. Thus studies such as this are vital in determining the effect of mining contamination on trout populations in the upper Clark Fork River. This study further documented impairment of trout habitat in the upper Clark Fork River and should be continued into the future to both bolster the dataset and allow for further analysis. Moving forward, this study will be collecting live fish every month from every site in order to test the assumption that surviving fish have lower tissue metals burdens than those that die. Testing this assumption may allow for further evidence of impacts of tissue metals burdens on fish in the upper Clark Fork River. Also, we will be moving the locations of our control sites to locations further upstream that should be less impacted by low flows during drought years. In addition, future years of this study will collect weights on fish when they are received from the hatchery, when they are found dead in cages, when they are collected alive, and when they are pulled at the end of the field season so that condition factor can be analyzed and possibly provide more information on the impacts of tissue metals burdens on these fish.

We also recognize the need to better understand the processes occurring in the Warm Springs Ponds and their impacts on fish through the release of the ponds' water into the Upper Clark Fork River. More specifically, further research is necessary to understand the effects the ponds (including water chemistry within the ponds) and the liming station have on water temperatures, pH, metals and general water quality of the Clark Fork River and overall whether their impact is positive or negative to the fishery of the Clark Fork River. Our findings of high pH below the ponds is of particular interest, as these values were above levels that are known to increase metals toxicity (Couture and Pyle 2012) and are above levels that trout species generally can tolerate (Colt et al. 1979). In summary, many of the detrimental environmental conditions observed are more likely to result in sub-lethal effects alone (Blazer et al. 1987; Molony 2001). However, conditions often interact synergistically or cumulatively to influence fish growth, survival, and ultimately populations (Driedzic and Hochachka 1978; Hellawell 1986; Boyd and Tucker 1998; Molony 2001; Kiser et al. 2010; Richards et al. 2013).

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Appendices

Appendix I: Published Electrofishing Data from Lindstrom (2011)

Table A1-1. Electrofishing data collected on the Upper Clark Fork River at the pH Shack Section from 2008 through 2010. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Number following the population estimate (in parentheses) represents the 95 % confidence interval. Cutt x Rbow represents a phenotypic hybrid between a cutthroat and rainbow trout.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# of Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2008	Brown	708 (+/- 102)	26	567	318	88-461	99
	Rainbow	-	-	5	388	296-502	< 1
	Cutthroat	-	-	3	365	355-381	< 1
2009	Brown	185 (+/- 73)	22	116	357	96-500	95
	Rainbow	-	-	5	362	302-560	4
	Cutthroat	-	-	1	383	-	1
2010	Brown	421 (+/- 149)	15	232	300	111-615	95
	Rainbow	-	-	5	478	312-565	2
	Cutthroat	-	-	3	260	252-276	1
	Cutt x Rbow	-	-	3	357	338-392	1

Table A1-2. Electrofishing data collected on the Upper Clark Fork River at the Below Sager Lane Section in 2010. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Number following the population estimate (in parentheses) represents the 95 % confidence interval.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# of Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2010	Brown	262 (+/- 85)	14	383	293	93-525	99
	Brook	-	-	3	232	125-293	< 1
	Rainbow	-	-	1	645	-	< 1

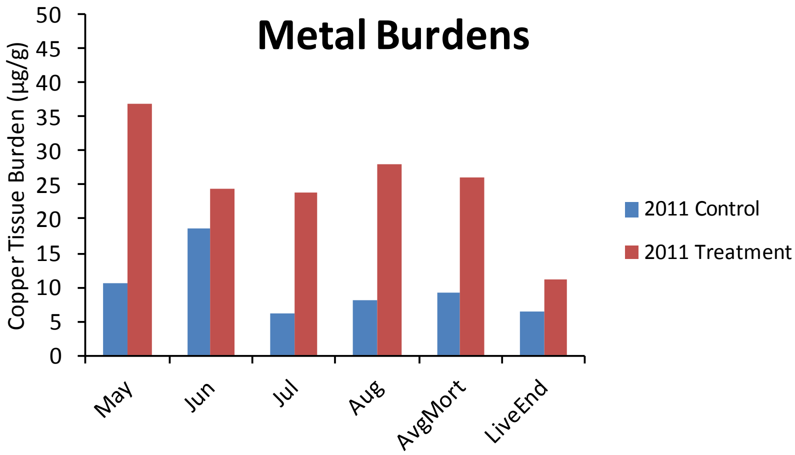
Table A1-3. Electrofishing data collected on the Upper Clark Fork River at the original Williams-Tavenner Section from 2008 through 2010. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Number following the population estimate (in parentheses) represents the 95 % confidence interval.

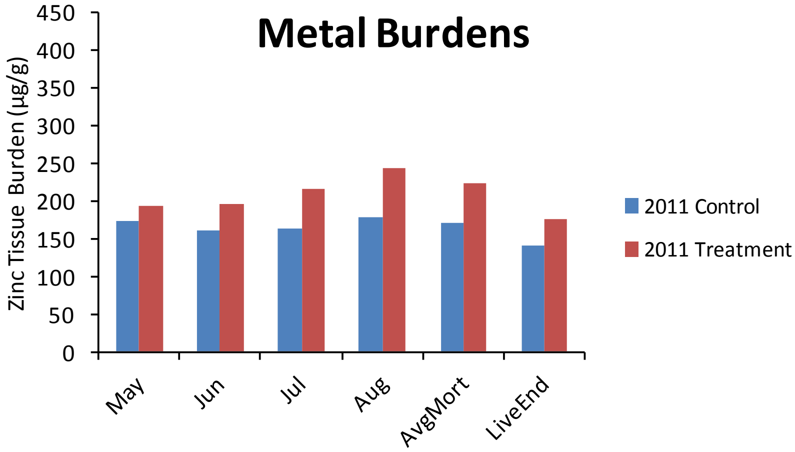
Year	Trout Species	Population Estimate -fish/mile-	Capture Efficiency -%-	# of Fish Handled	Mean Length -mm-	Length Range -mm-	Species Composition -%-
2008	Brown	324 (+/- 84)	28	194	349	118-524	100
2009	Brown	158 (+/- 77)	19	77	341	132-527	99
	Cutthroat	-	-	1	279	-	1
2010	Brown	206 (+/- 59)	27	146	332	114-509	99
	Cutthroat	-	-	1	285	-	< 1
	Brook	-	-	1	145	-	< 1

Table A1-4. Electrofishing data collected on the Upper Clark Fork River at the Phosphate Section from 2008 through 2010. Population estimates and capture efficiencies are for brown trout greater than 175 mm (~7") in total length. Number following the population estimate (in parentheses) represents the 95 % confidence interval.

Year	Trout Species	Population Estimate (fish/mile)	Capture Efficiency (%)	# of Fish Handled	Mean Length (mm)	Length Range (mm)	Species Composition (%)
2008	Brown	316 (+/- 58)	31	343	333	97-468	99
	Cutthroat	-	-	3	325	256-380	1
2009	Brown	292 (+/- 143)	13	159	334	125-465	99
	Cutthroat	-	-	1	274	-	1
2010	Brown	233 (+/- 46)	35	279	308	97-478	99
	Cutthroat	-	-	3	291	242-345	1

Appendix II: Previous Year's Comparisons





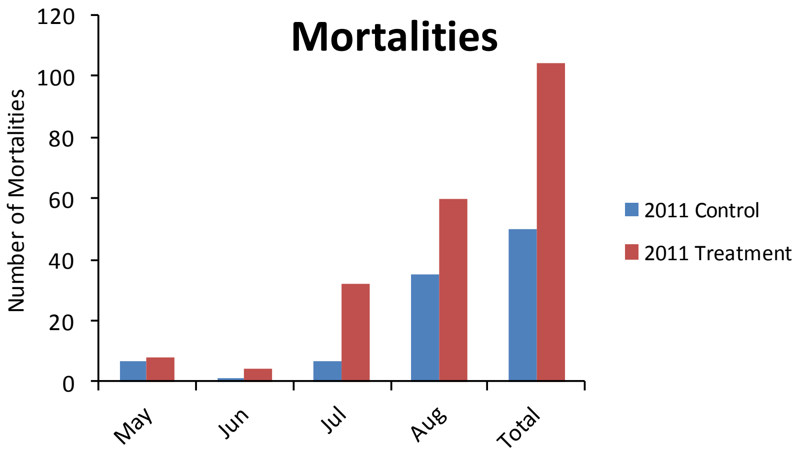


Figure A2-1. Comparisons between control and treatment sites' tissue metals burdens and number of mortalities by month in 2011. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2011 field season and the last column of the mortalities figure represents the total number of mortalities during the 2011 field season.

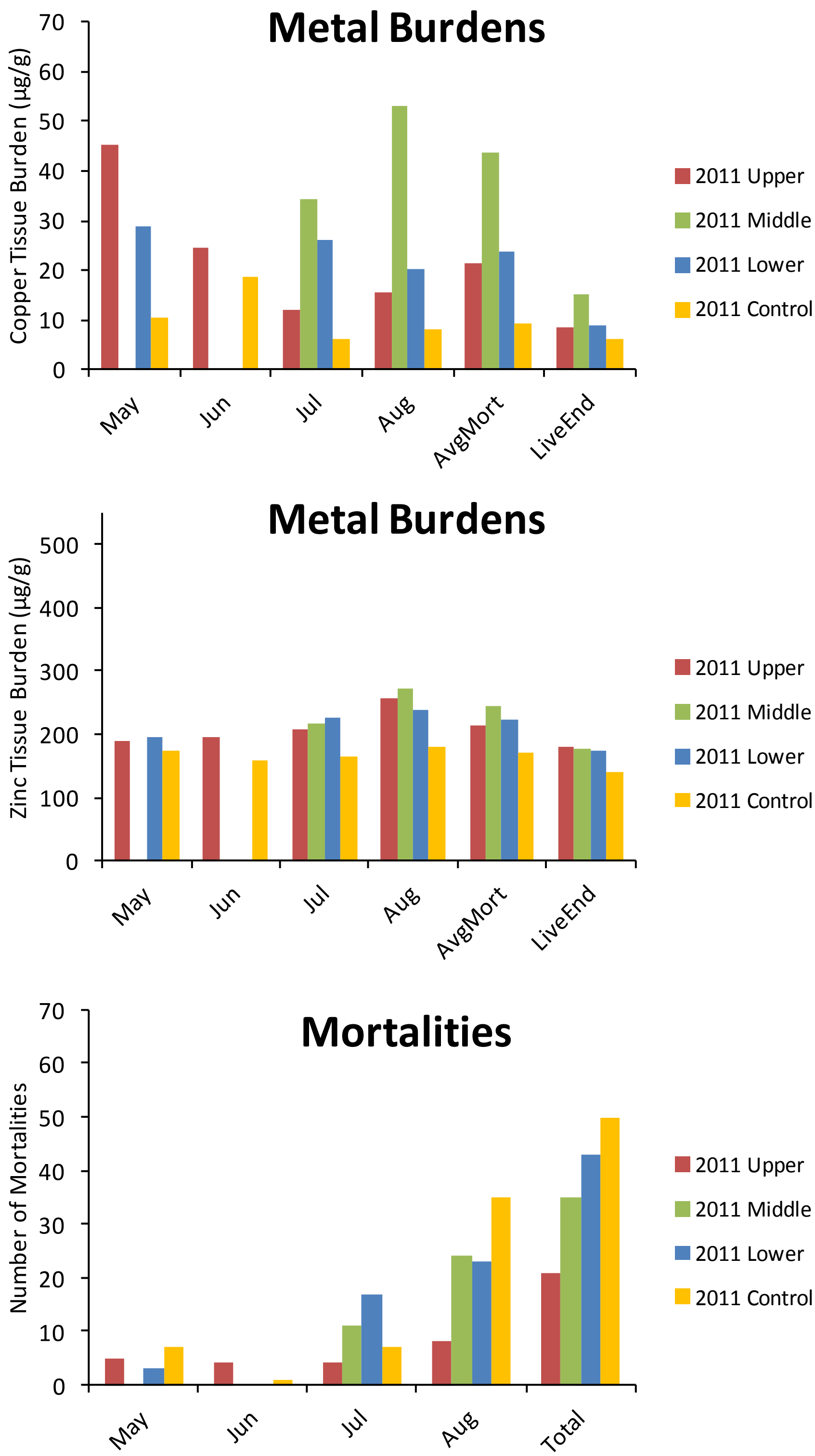


Figure A2-2. Comparisons between upper and lower sites' tissue metals burdens and number of mortalities by month in 2011. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2011 field season and the last column of the mortalities figure represents the total number of mortalities during the 2011 field season.

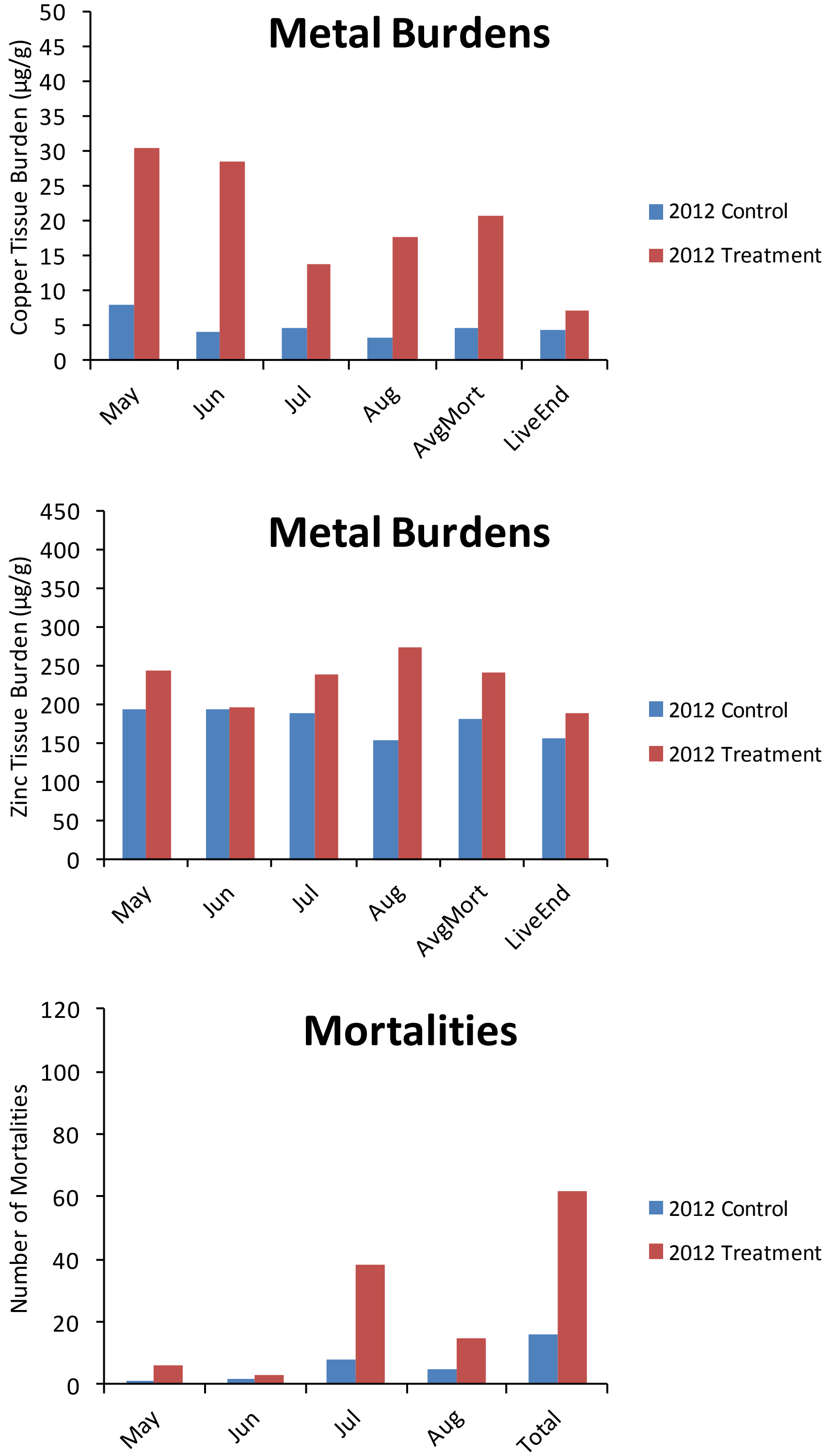


Figure A2-3. Comparisons between control and treatment sites' tissue metals burdens and number of mortalities by month in 2012. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2012 field season and the last column of the mortalities figure represents the total number of mortalities during the 2012 field season.

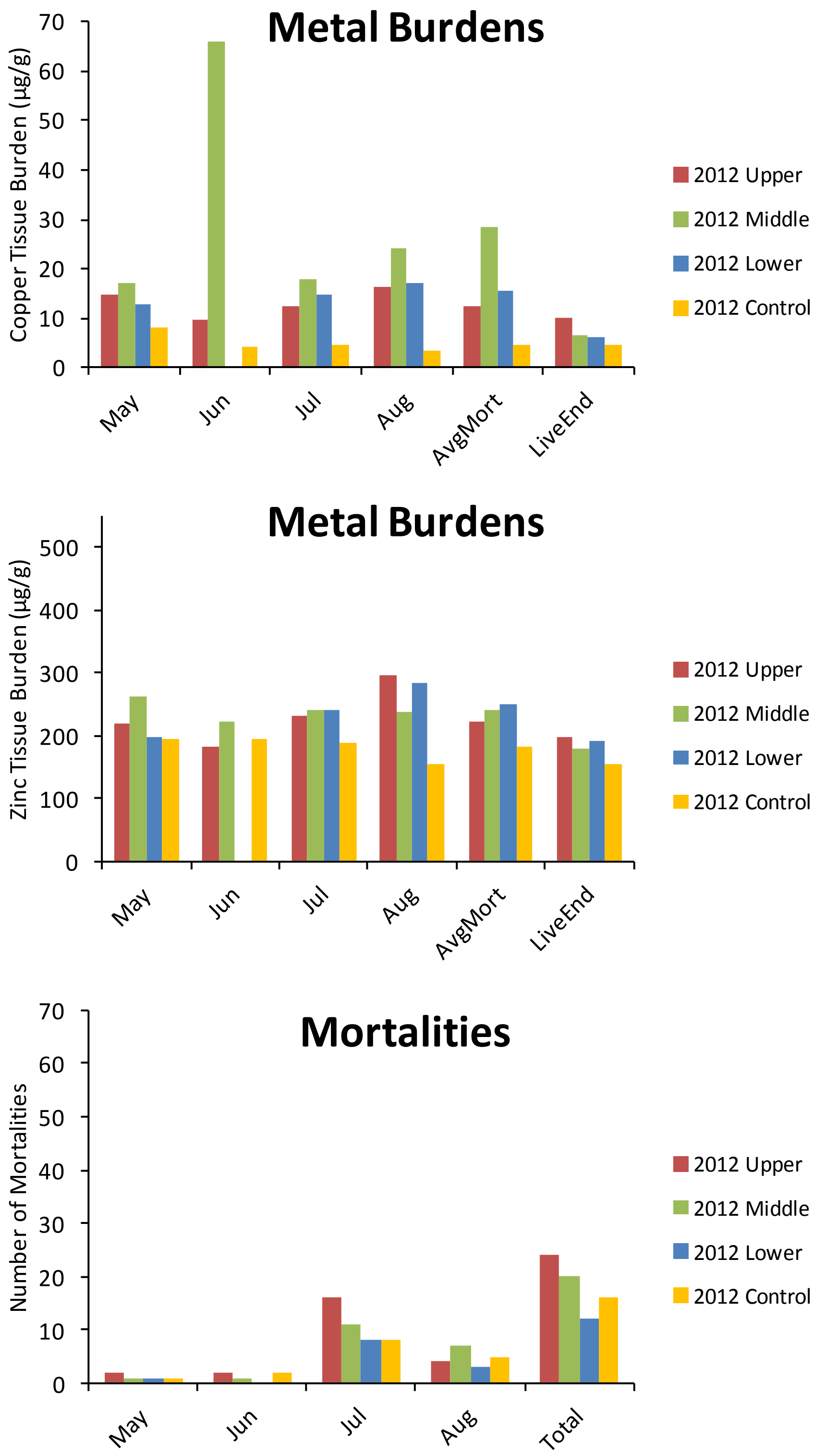


Figure A2-4. Comparisons between upper, middle, and lower sites' tissue metals burdens and number of mortalities by month in 2012. The last column of each metals burdens figure represents the values of the fish sampled that lived to the end of the 2012 field season and the last column of the mortalities figure represents the total number of mortalities during the 2012 field season.

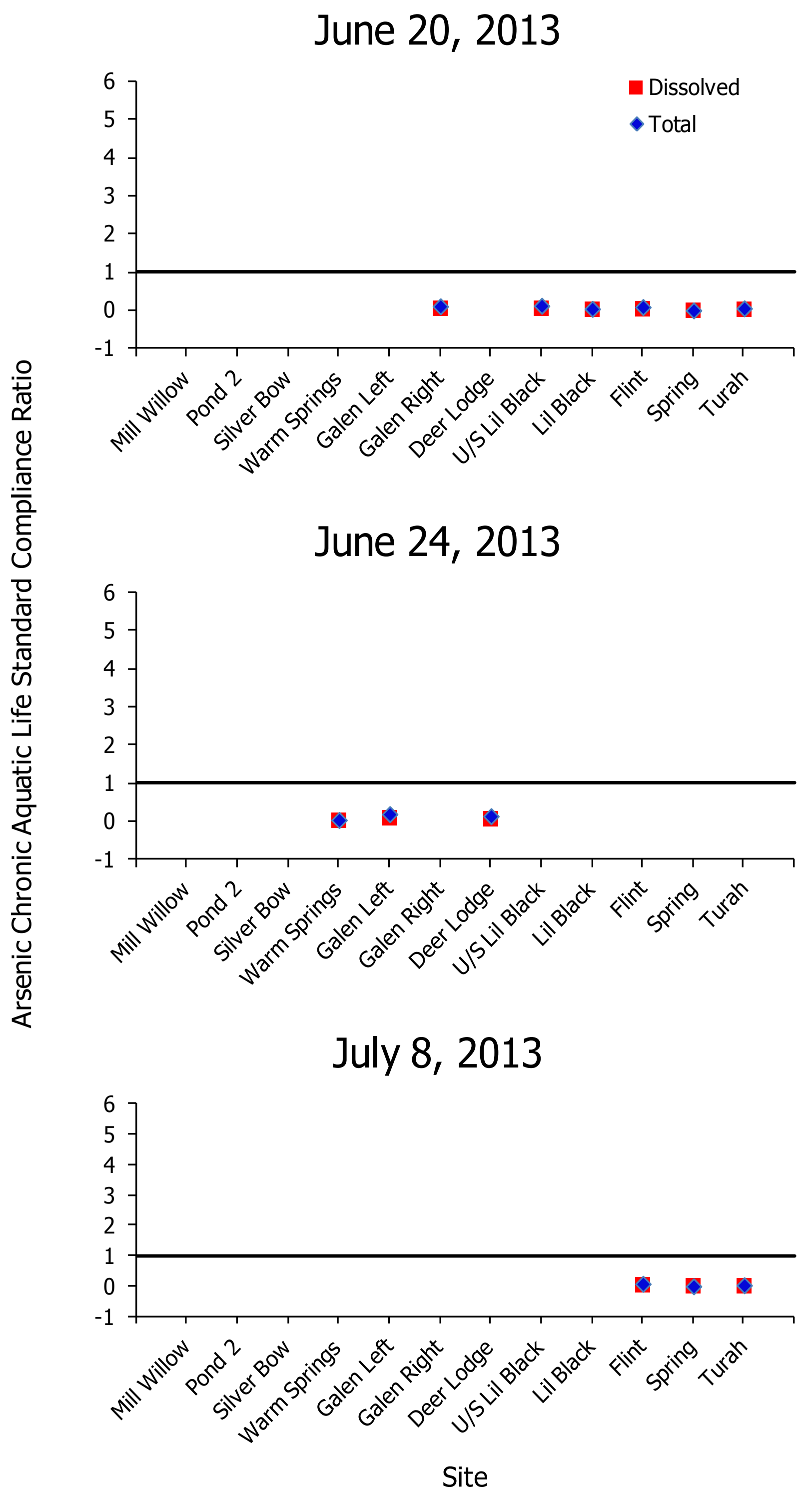


Figure A3-1. Arsenic compliance ratios at the cage sites from rain events on June 20, June 24, and July 8 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing arsenic concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate arsenic levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

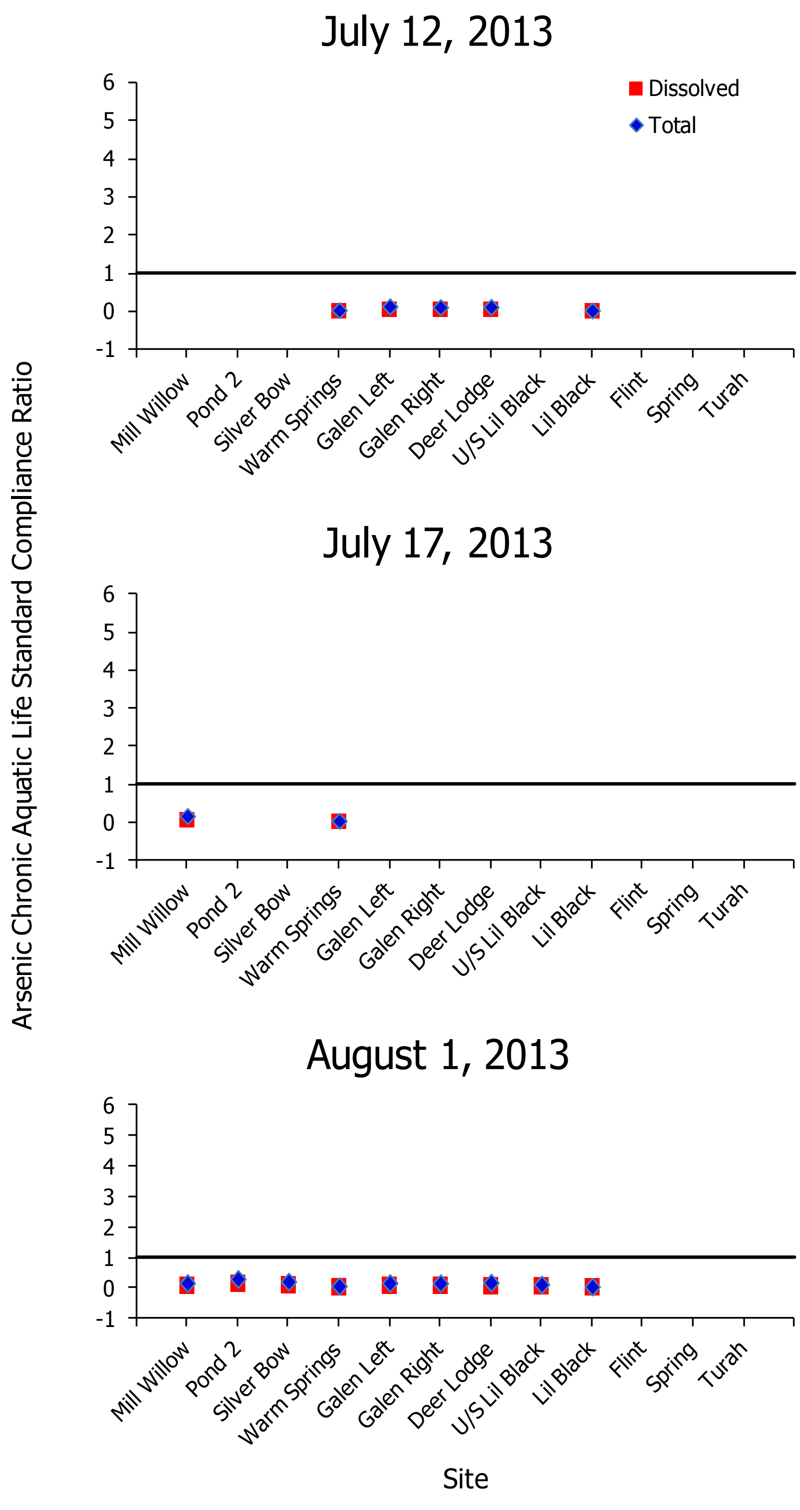


Figure A3-2. Arsenic compliance ratios at the cage sites from rain events on July 12, July 17, and August 1 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing arsenic concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate arsenic levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

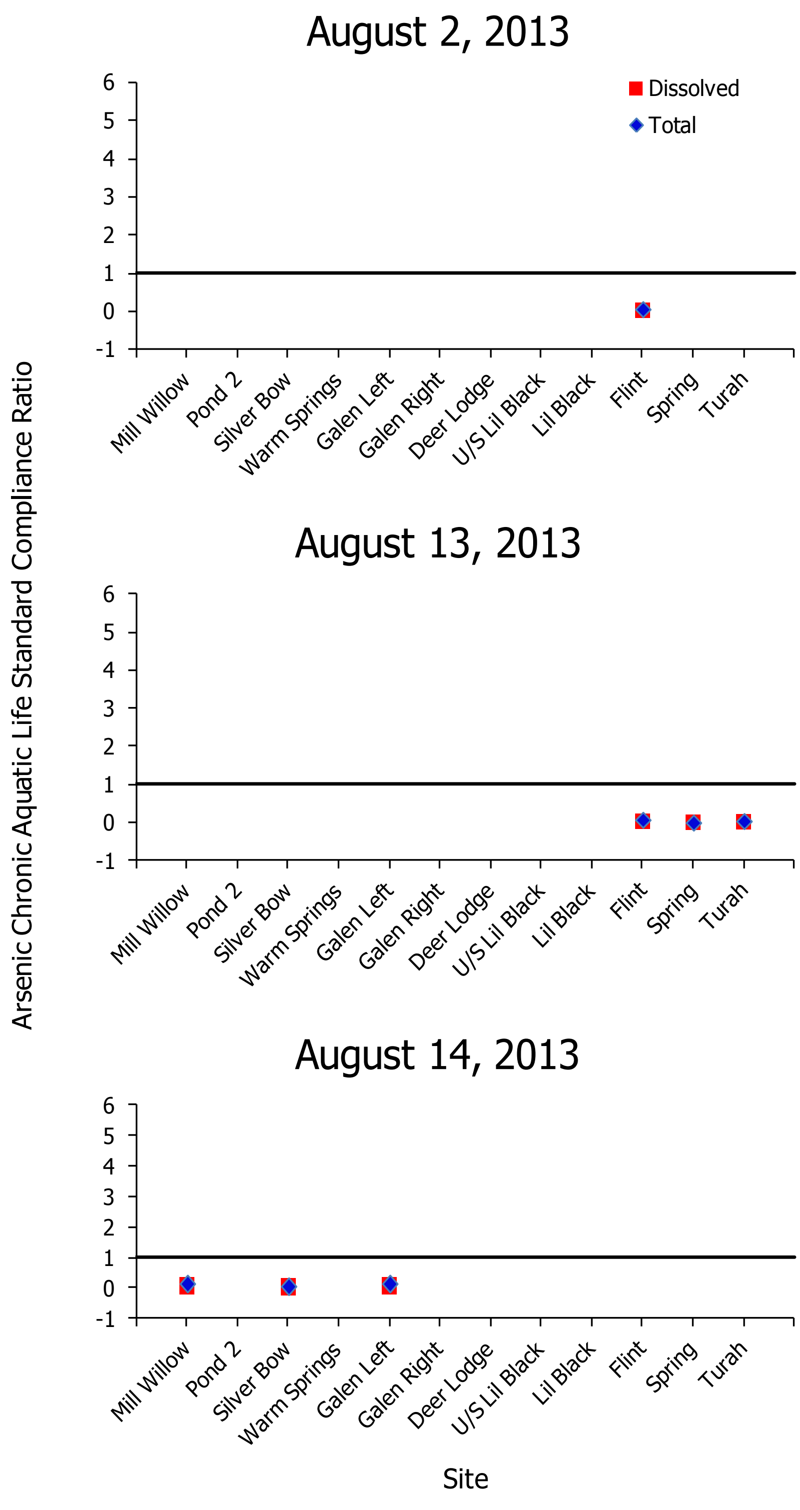


Figure A3-3. Arsenic compliance ratios at the cage sites from rain events on August 2, August 13, and August 14 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing arsenic concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate arsenic levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

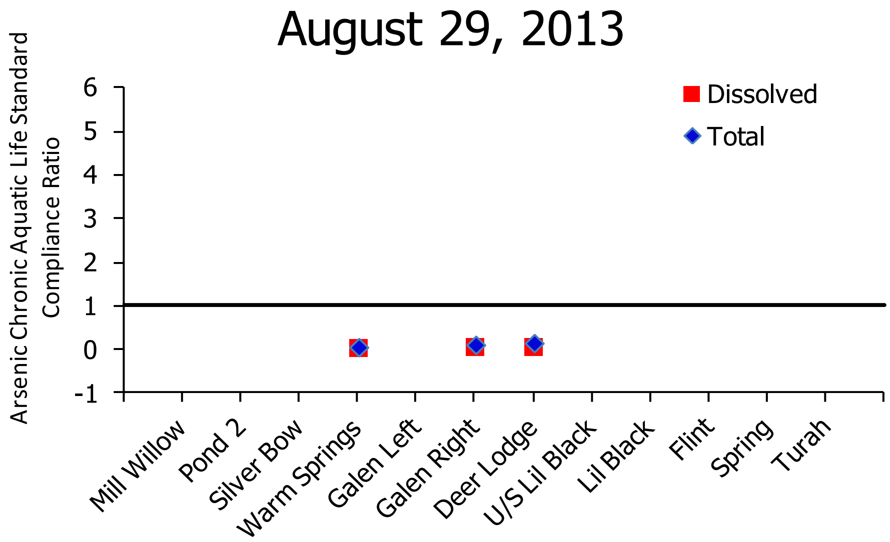


Figure A3-4. Arsenic compliance ratios at the cage sites from the rain event on August 29, 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing arsenic concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate arsenic levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

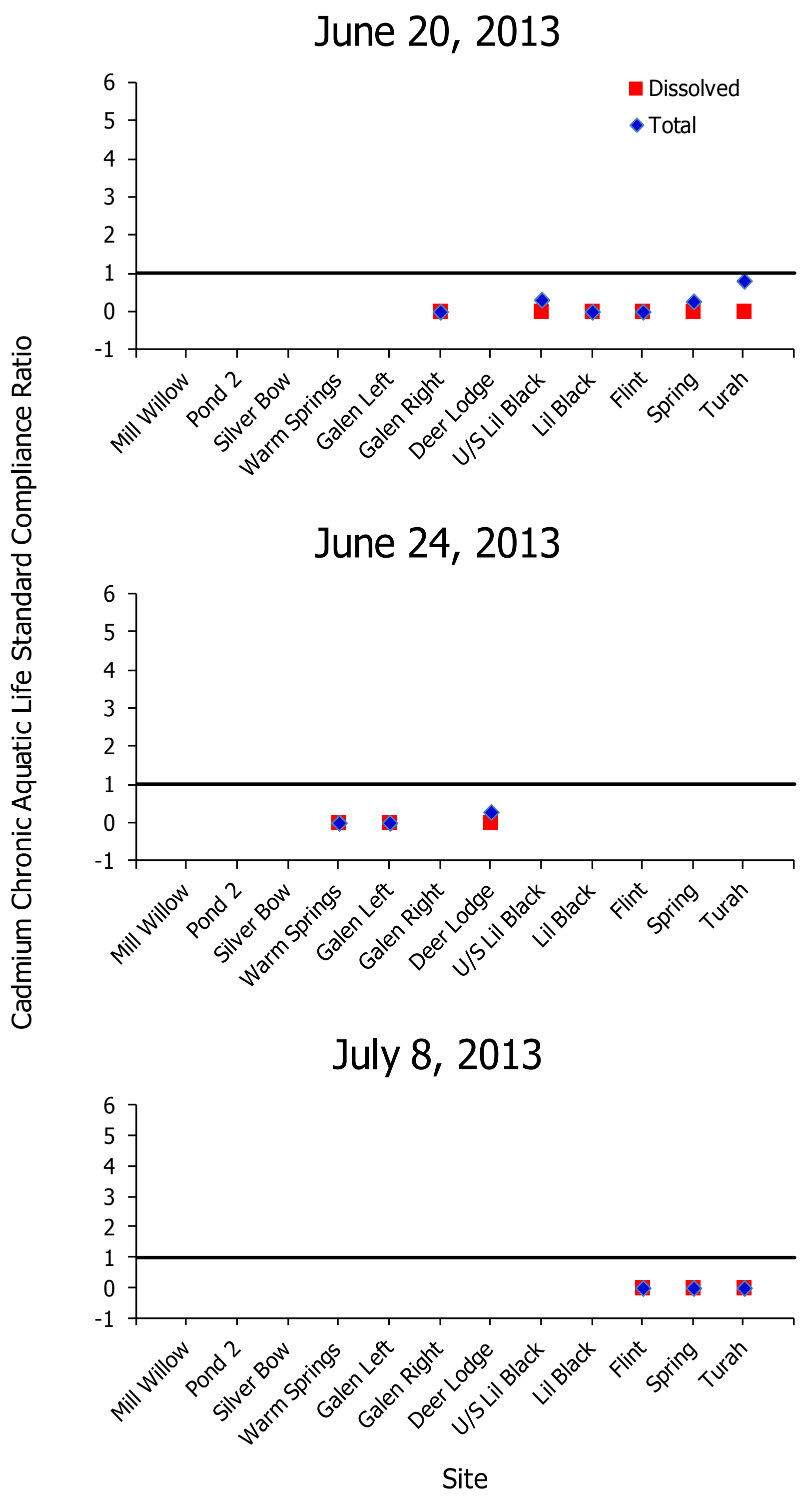


Figure A3-5. Cadmium compliance ratios at the cage sites from rain events on June 20, June 24, and July 8 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing cadmium concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate cadmium levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

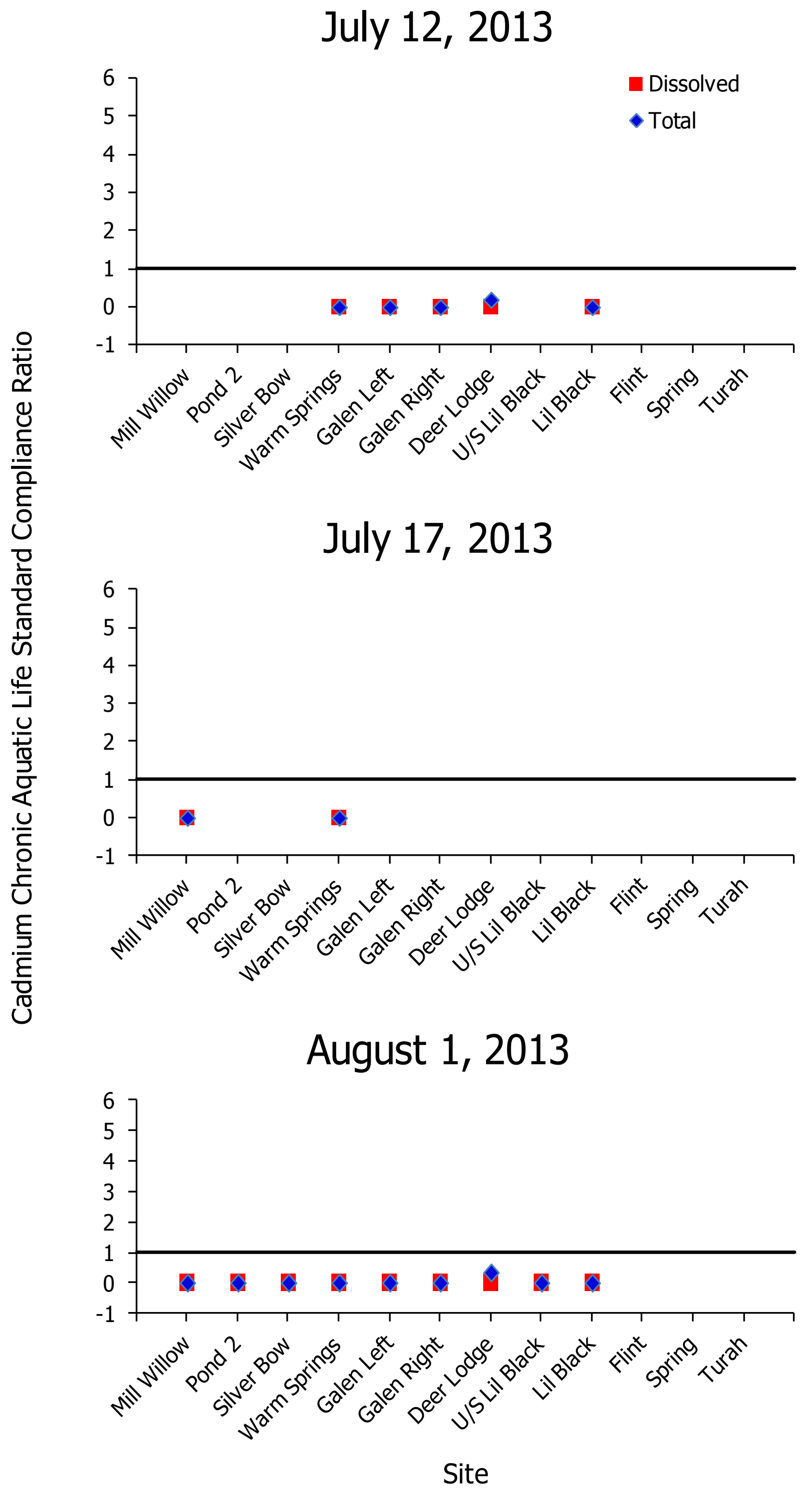


Figure A3-6. Cadmium compliance ratios at the cage sites from rain events on July 12, July 17, and August 1 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing cadmium concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate cadmium levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

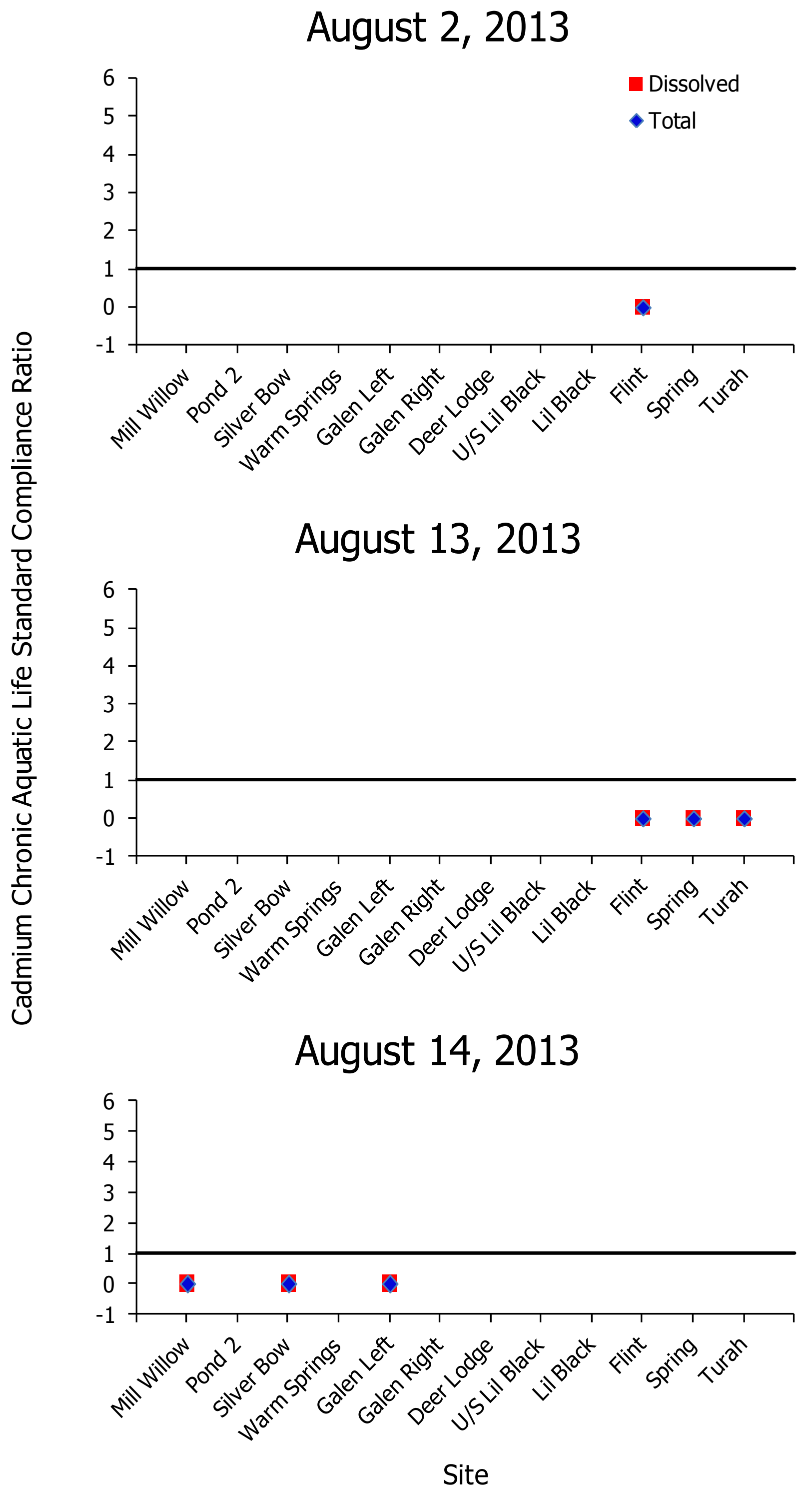


Figure A3-7. Cadmium compliance ratios at the cage sites from rain events on August 2, August 13, and August 14 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing cadmium concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate cadmium levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

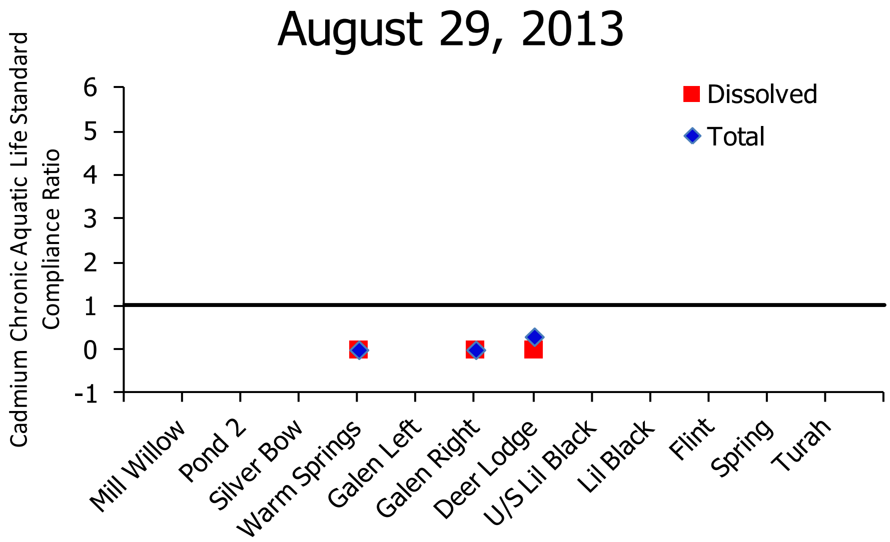


Figure A3-8. Cadmium compliance ratios at the cage sites from the rain event on August 29, 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing cadmium concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate cadmium levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

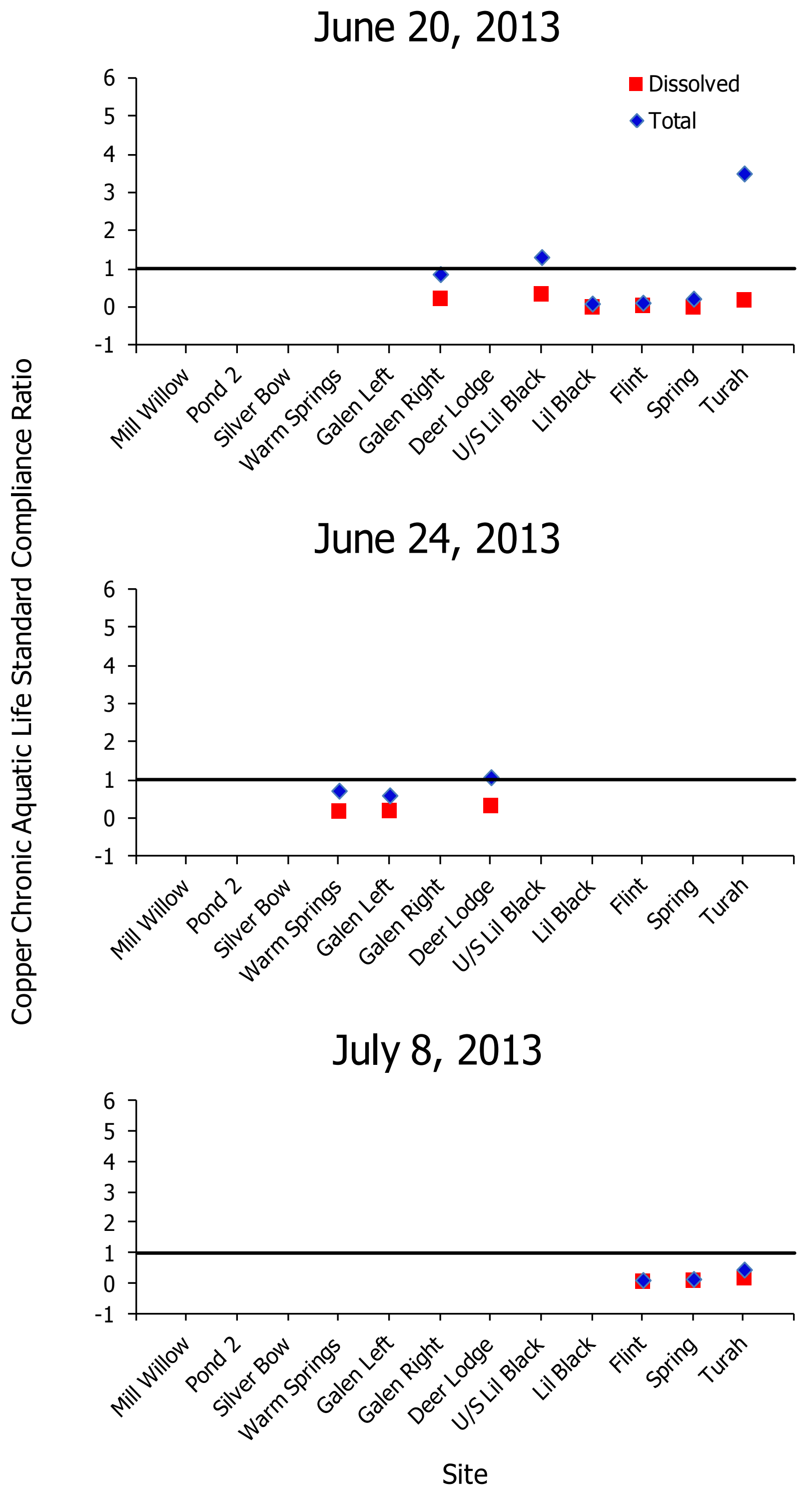


Figure A3-9. Copper compliance ratios at the cage sites from rain events on June 20, June 24, and July 8 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing copper concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate copper levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

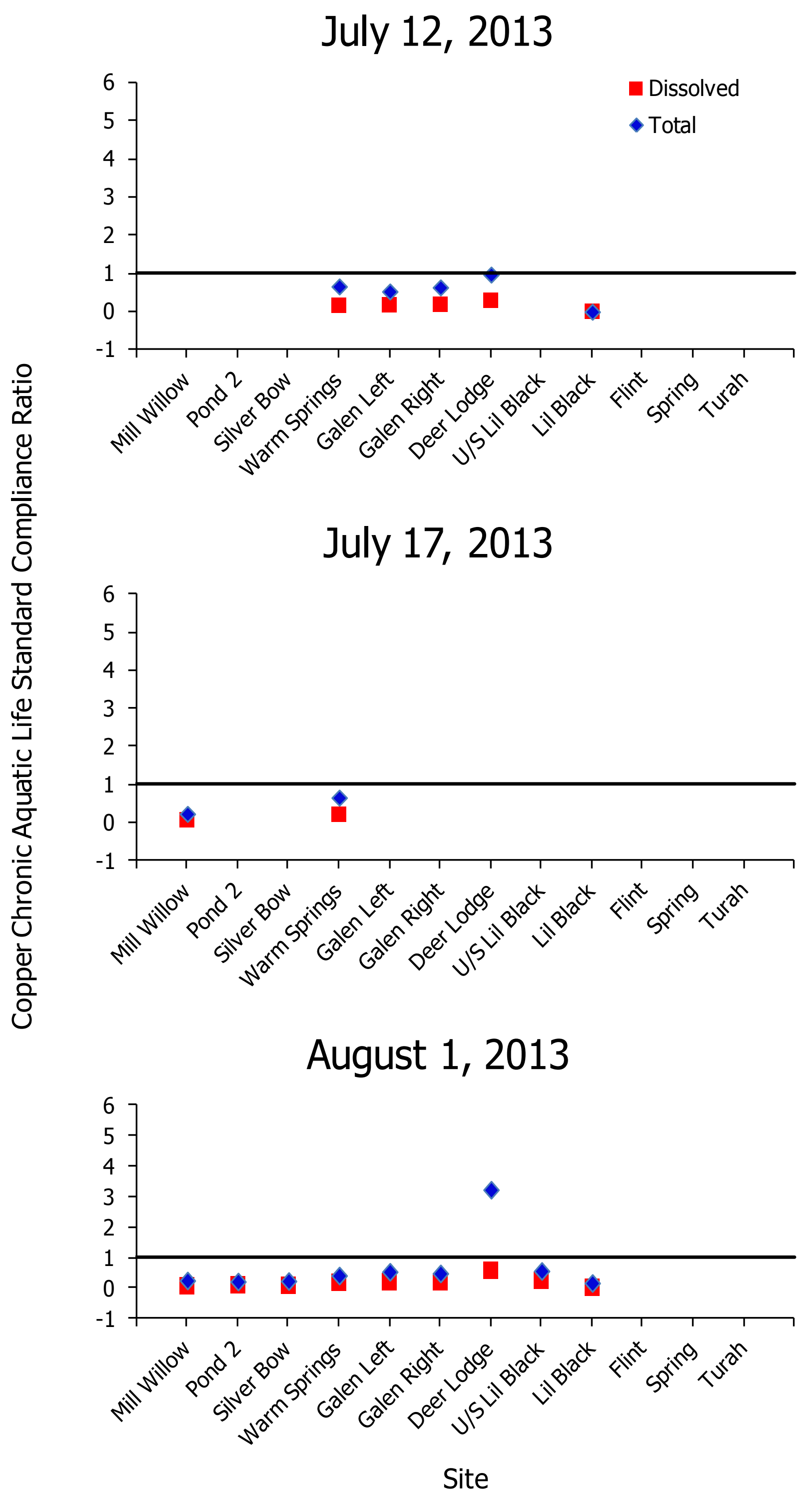


Figure A3-10. Copper compliance ratios at the cage sites from rain events on July 12, July 17, and August 1 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing copper concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate copper levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

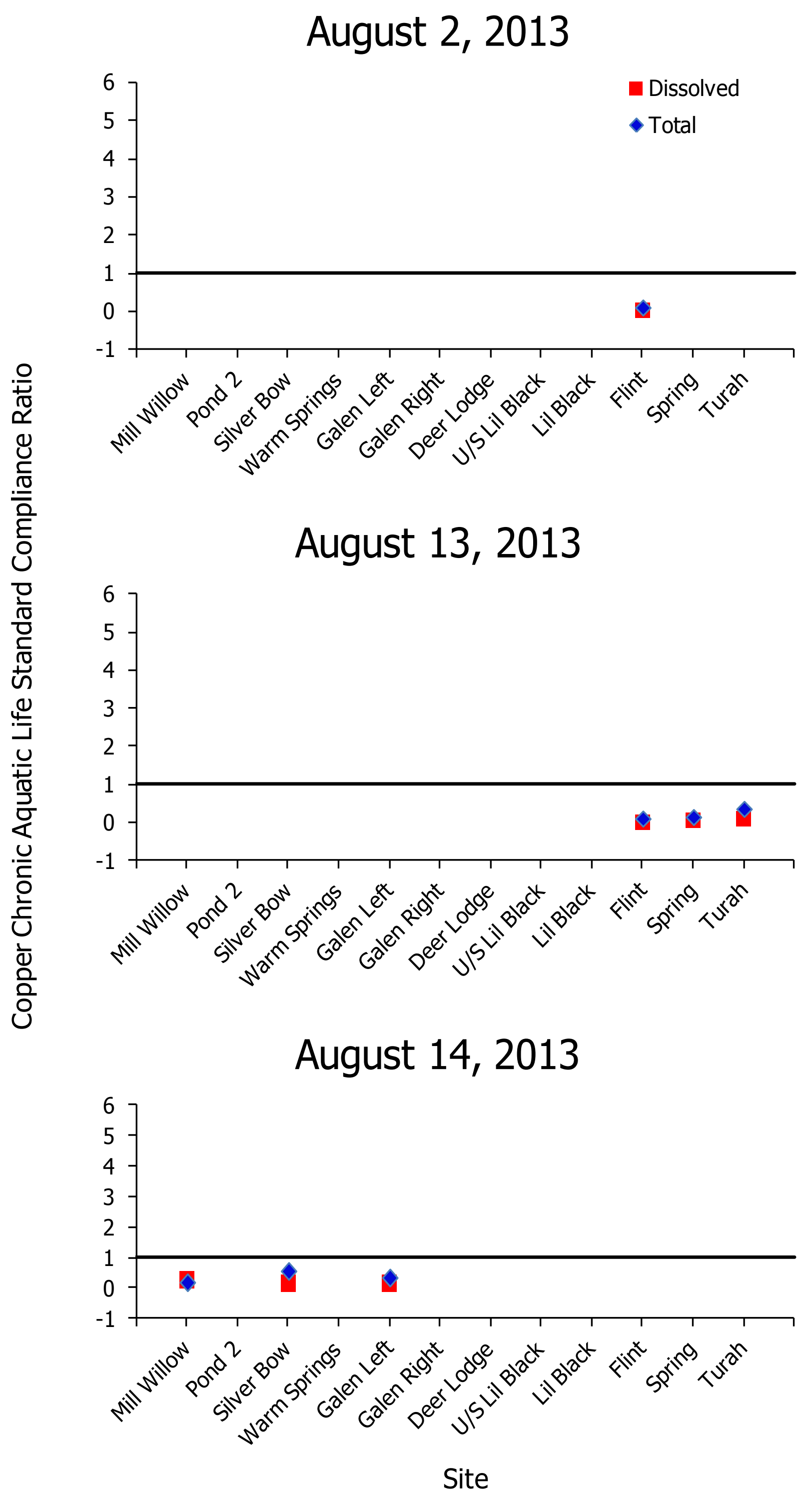


Figure A3-11. Copper compliance ratios at the cage sites from rain events on August 2, August 13, and August 14 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing copper concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate copper levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

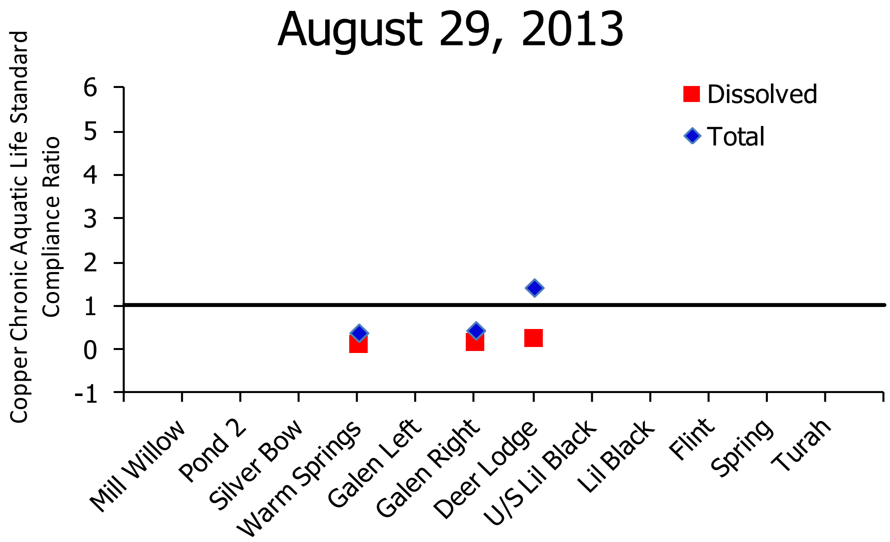


Figure A3-12. Copper compliance ratios at the cage sites from the rain event on August 29, 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing copper concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate copper levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

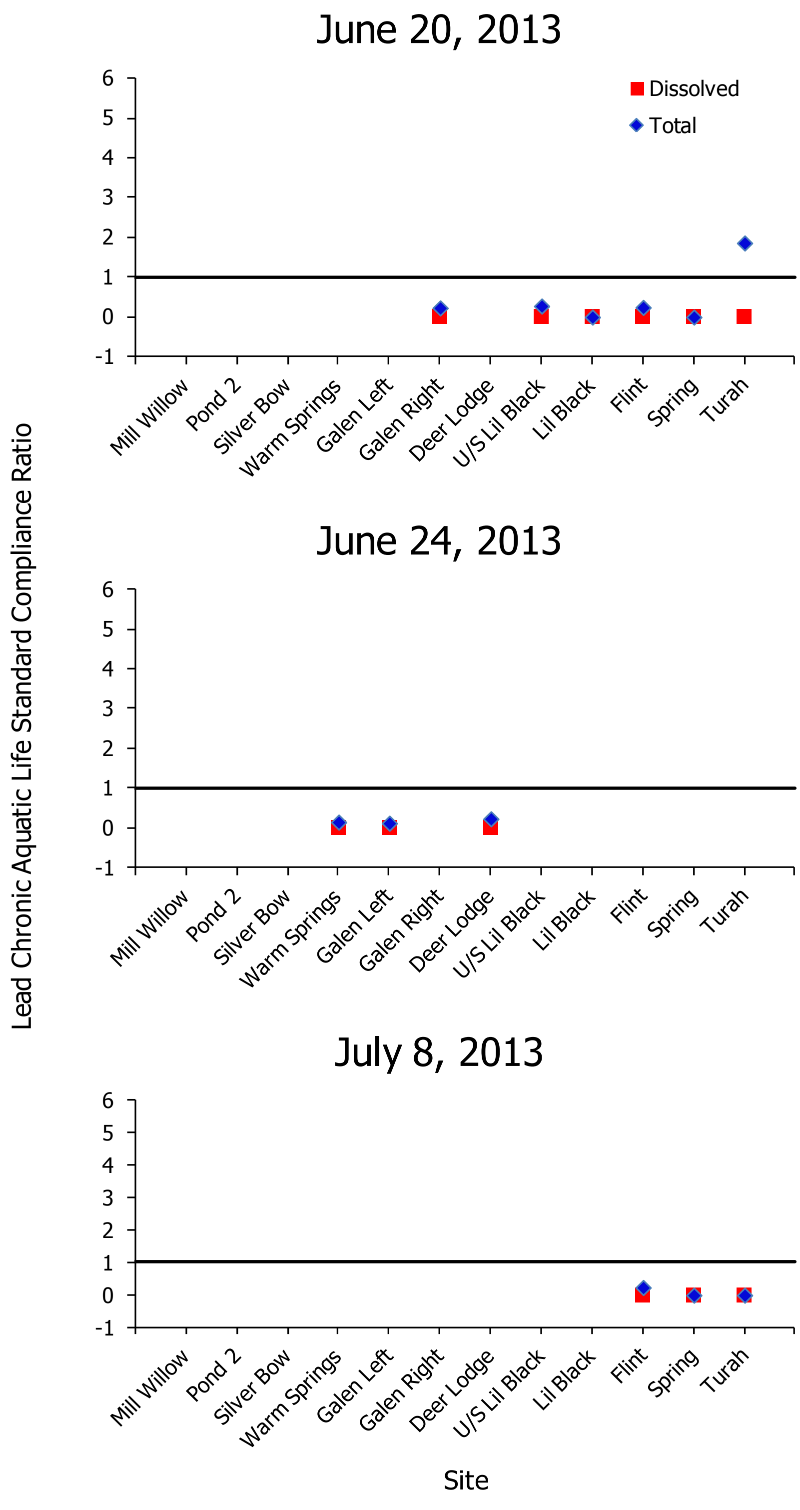


Figure A3-13. Lead compliance ratios at the cage sites from rain events on June 20, June 24, and July 8 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing lead concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate lead levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

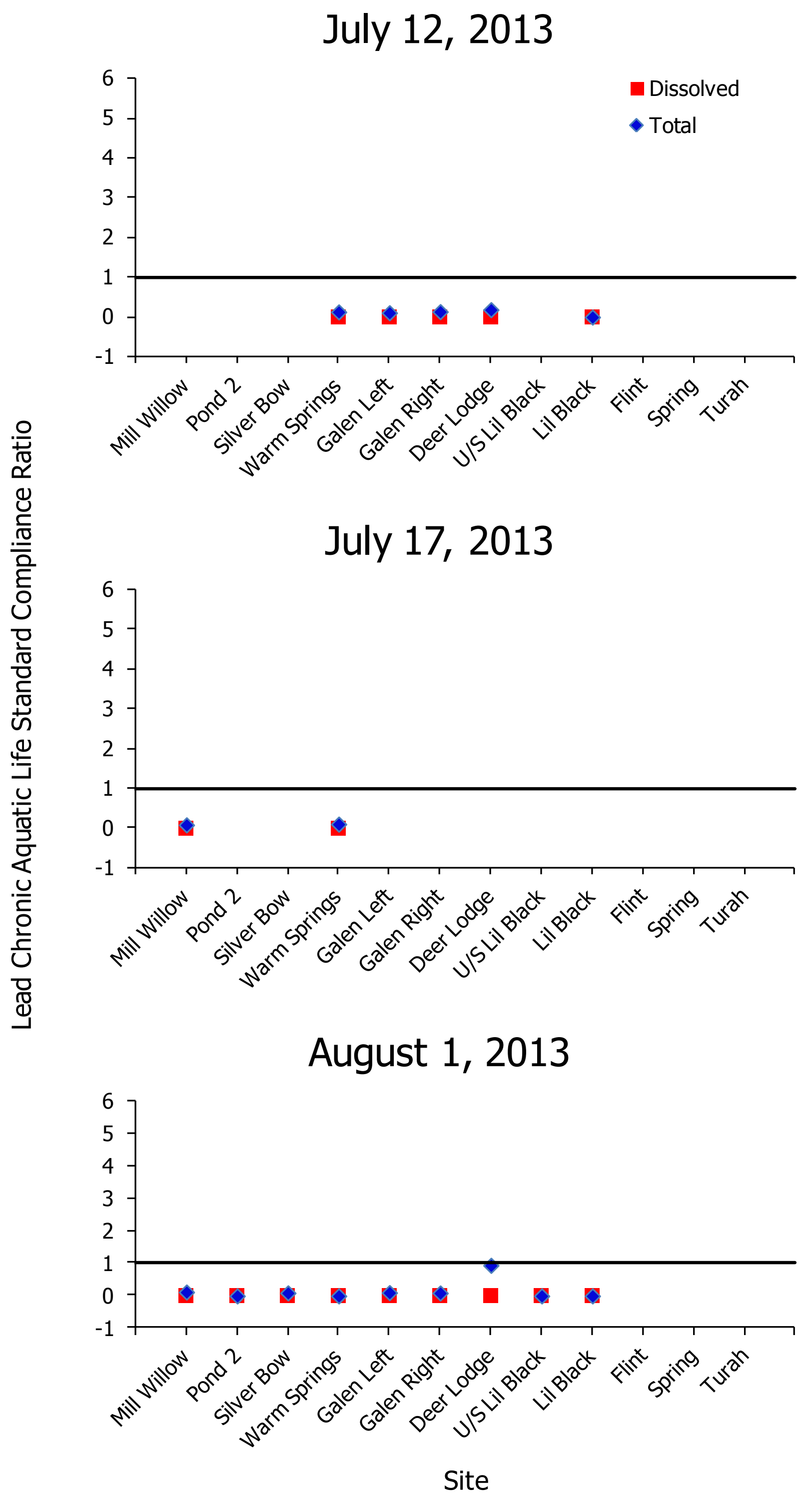


Figure A3-14. Lead compliance ratios at the cage sites from rain events on July 12, July 17, and August 1 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing lead concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate lead levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

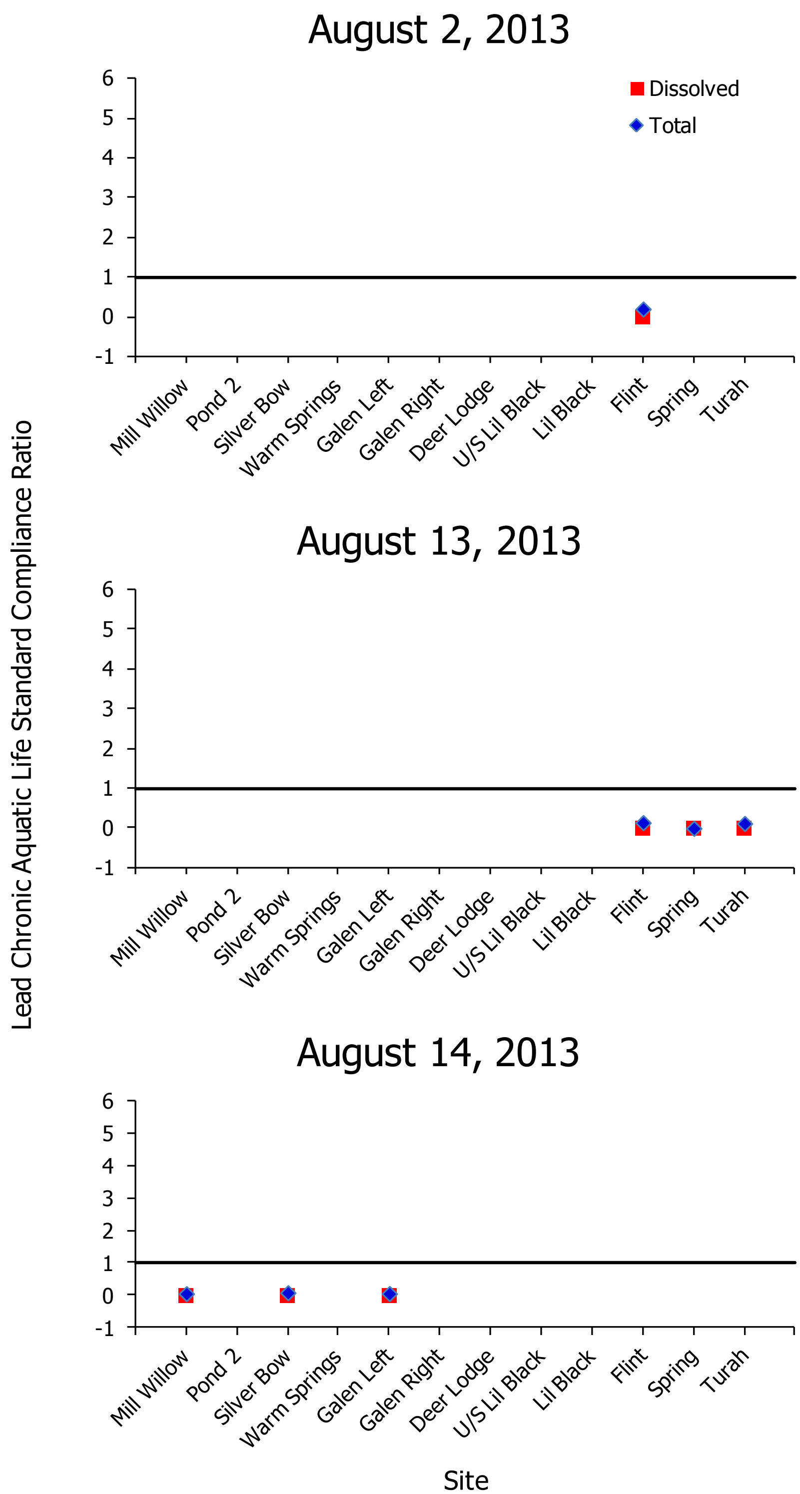


Figure A3-15. Lead compliance ratios at the cage sites from rain events on August 2, August 13, and August 14 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing lead concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate lead levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

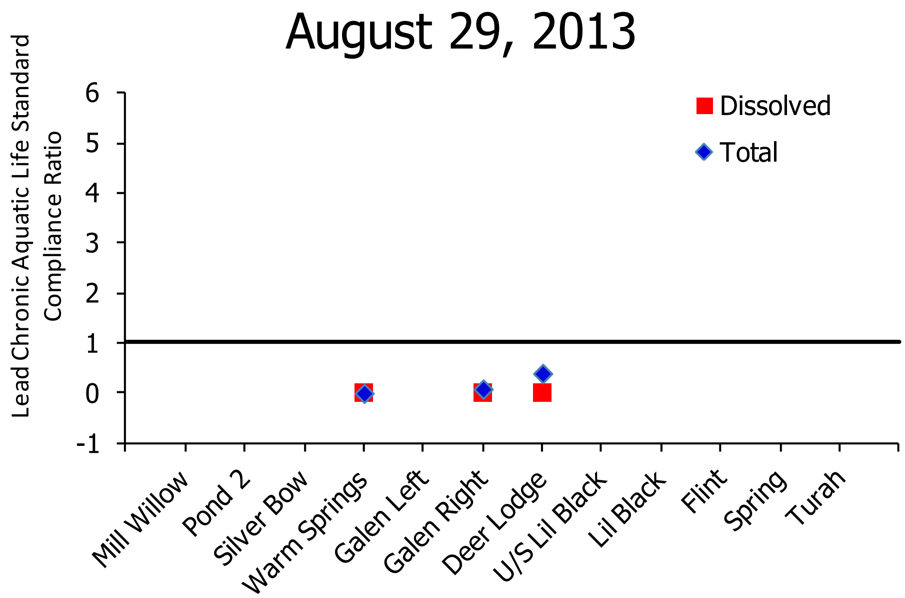


Figure A3-16. Lead compliance ratios at the cage sites from the rain event on August 29, 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing lead concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate lead levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

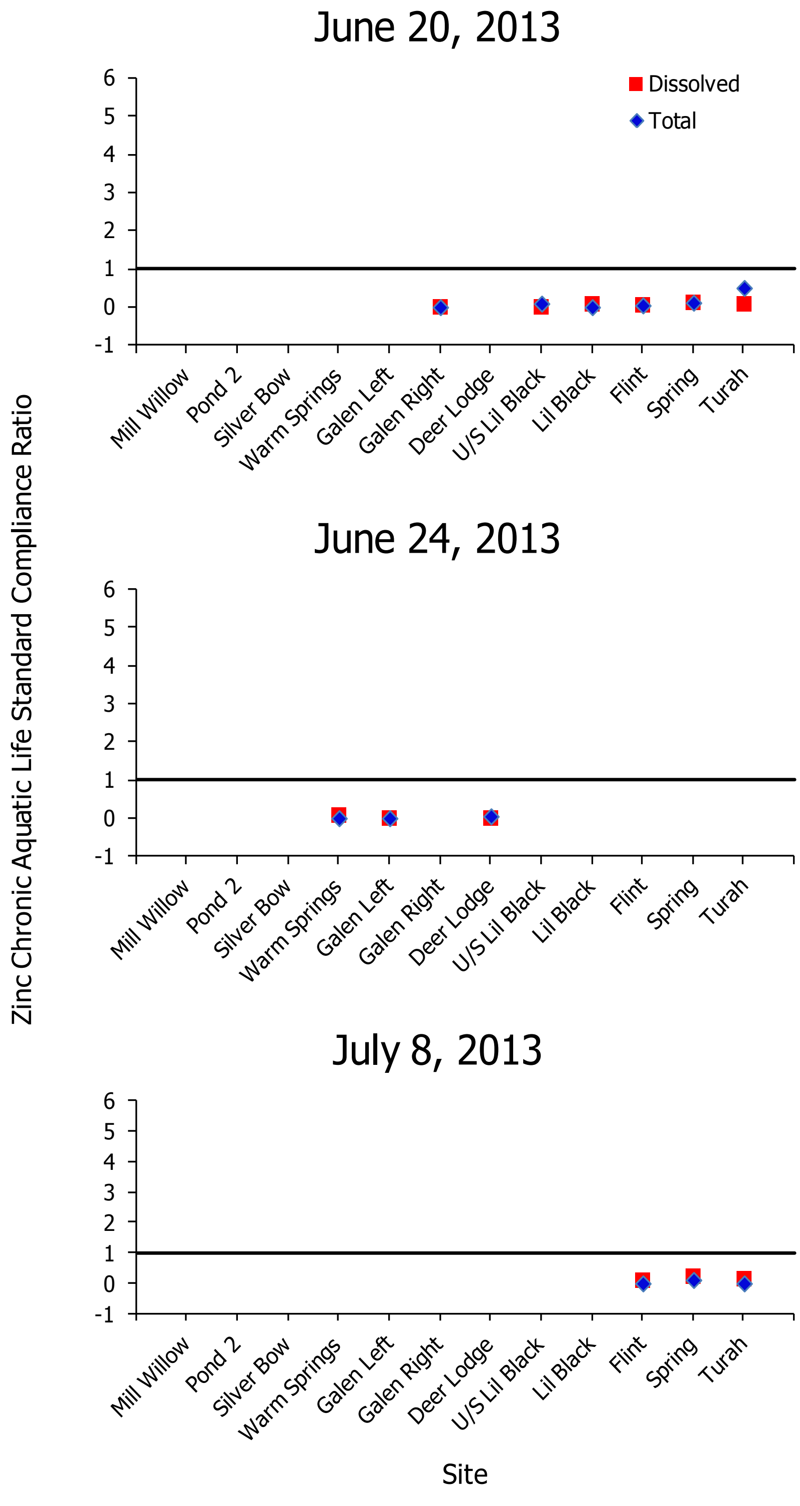


Figure A3-17. Zinc compliance ratios at the cage sites from rain events on June 20, June 24, and July 8 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing zinc concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate zinc levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

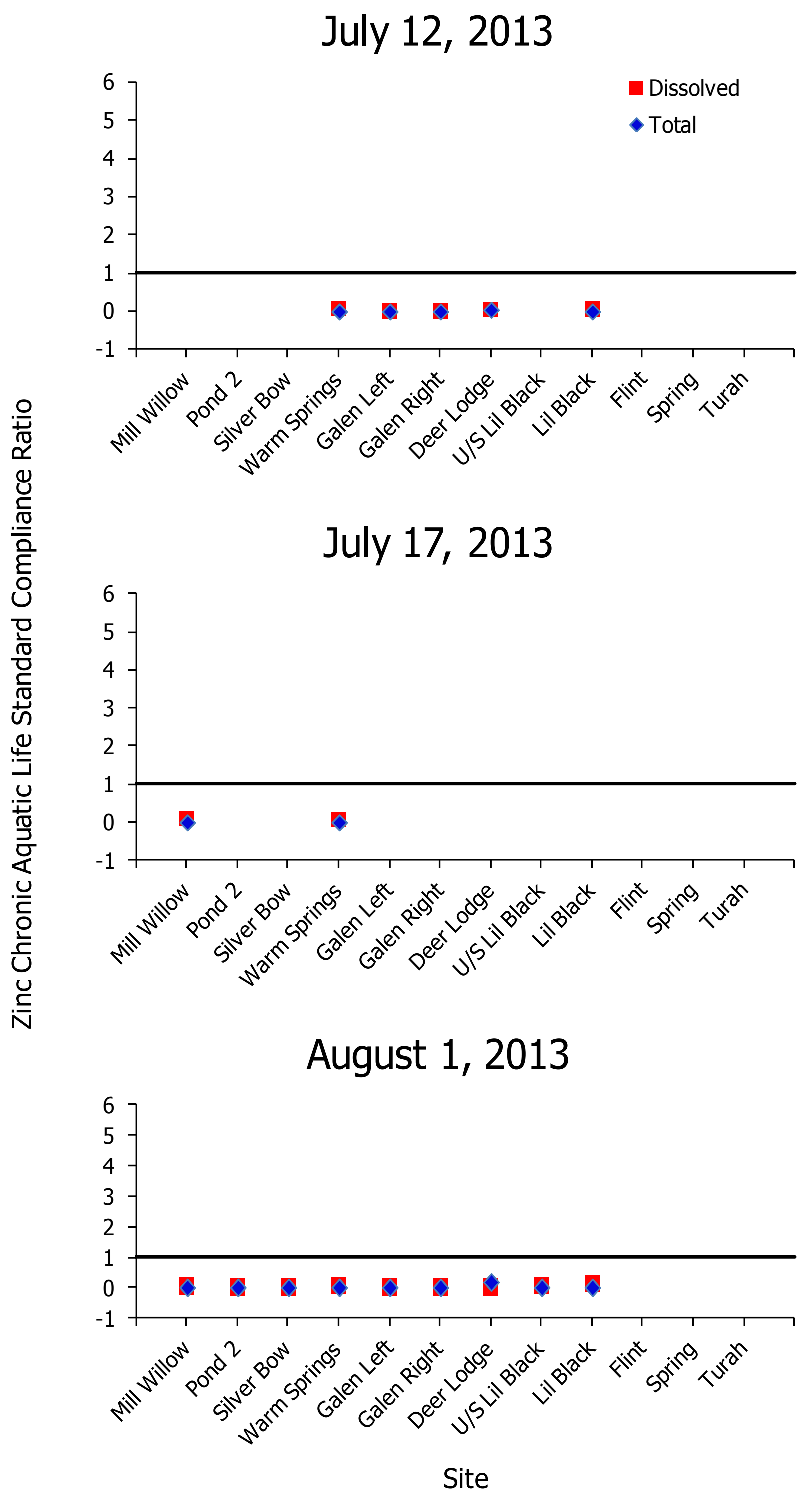


Figure A3-18. Zinc compliance ratios at the cage sites from rain events on July 12, July 17, and August 1 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing zinc concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate zinc levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

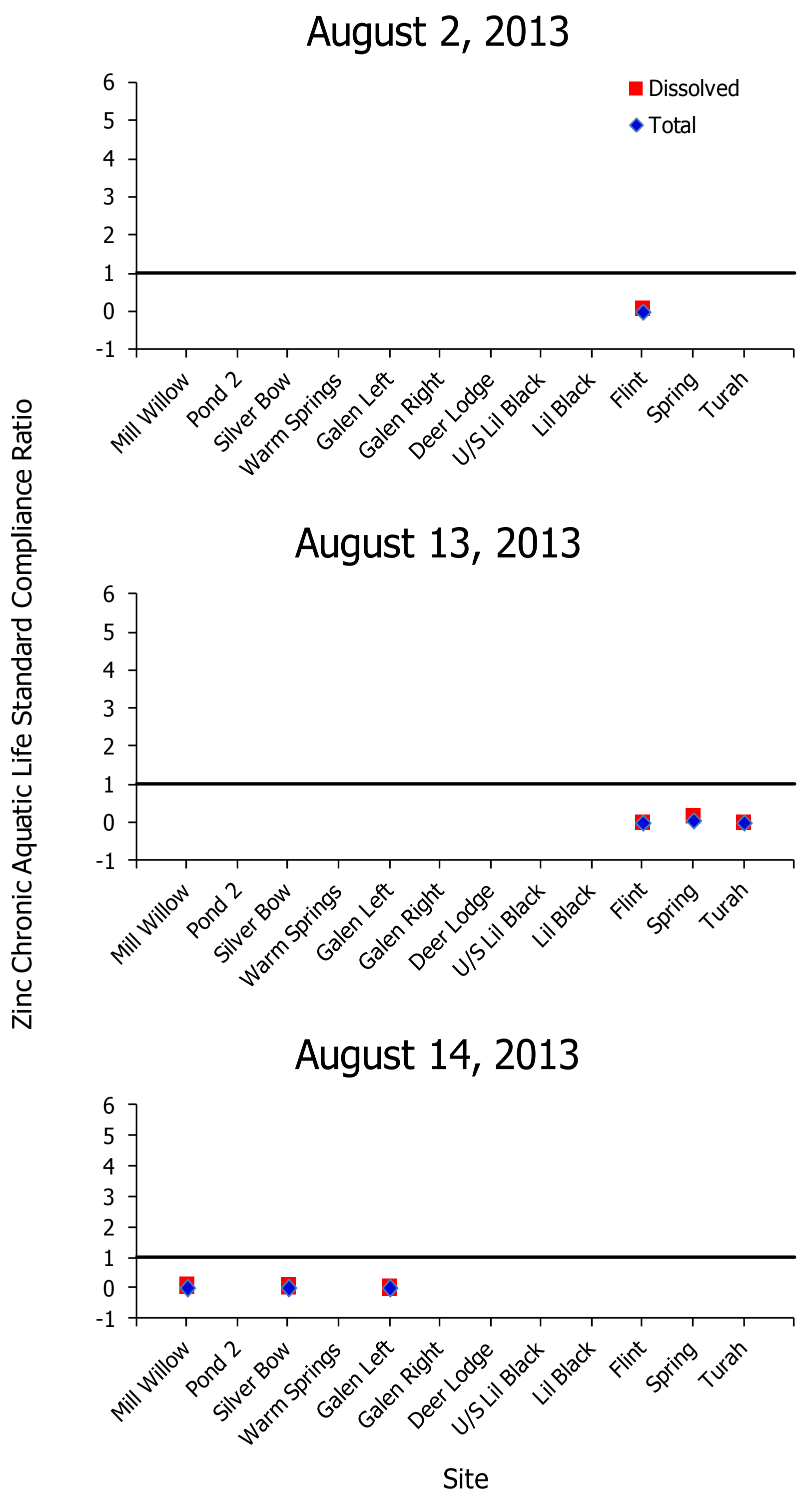


Figure A3-19. Zinc compliance ratios at the cage sites from rain events on August 2, August 13, and August 14 in 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing zinc concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate zinc levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).

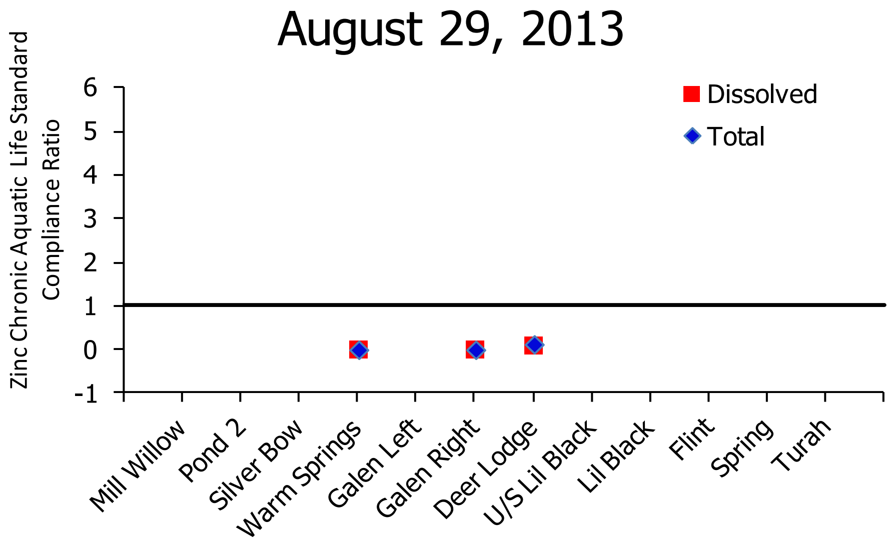


Figure A3-20. Zinc compliance ratios at the cage sites from the rain event on August 29, 2013 arranged from upstream to downstream. Compliance ratios were calculated by dividing zinc concentrations by the calculated chronic aquatic life standards. Compliance ratio values <1 indicate zinc levels below the aquatic life standard (compliance) while values >1 indicate levels above the standard (non-compliance).