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Ambient Toxicity Assessments of Clark Fork River Water-Toxicity Tests and Metals Residues in Brown Trout Organs

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Abstract

Trout population densities decline dramatically in the Clark Fork River from nearly 2,000 catchable brown trout per mile just downstream of mine waste settling ponds to 50 trout Instream toxicity tests (1986-89), and analyses of metals infish organs (1989) were conducted in various river reaches to try to understand how metals influence trout density patterns.

Instream toxicity tests with swim-up stage rainbow trout fry demonstrate that river water induces chronic mortality during spring runoff when metals concentration, particularly copper, exceed chronic criteria for protection of aquatic life. Concentration of copper in livers of adult brown trout (salmo trutta) are higher than those in laboratory fish populations exposed for several generations to chronically toxic concentrations of copper. Both acute and chronic stress from metals, particularly copper, are implicated as contributing to poor fish production in the Clark Fork.

Introduction

Fish kills have occurred frequently in the upper Clark Fork River over the last several years. Six were documented between 1983 and 1989-some killing several thousand fish. Kills occur during high intensity summer thunderstorms that flush metals sails accumulated on the surface of streamside mine tailings into the river. Tailings are rich in cadmium, copper, zinc, arsenic, iron and other metals.

During late winter and spring, concentrations of metals (particularly copper) in the Clark Fork frequently exceed criteria for protection of freshwater aquatic life (USEPA 1985). Fish kills have not been documented during these events but they may chronically stress trout populations by causing toxicity to sensitive early life stages -occurrences that could cagily go unnoticed.

To evaluate effects on early life stages, we conducted instream toxicity tests using fry and fingerling rainbow trout (1986-89) and measured concentrations of metals in brown trout organs (1989). Data were collected in the Clark Fork drainage between Anaconda and Clinton to gain a better understanding of factors influencing the distribution and abundance of brown trout in this segment of the river.

Methods and Materials

Toxicity Tests Beginning in 1986, in stream toxicity tests were conducted in the Clark Fork River for four consecutive years. Tests included at least six sites and up Lo nine sites including one or two control sites (Figure 1). Fingerling rainbow trout were tested during 1986 and 1987 and swim-up stage fry were tested from 1987-1989. All test fish were the progeny of Eagle Lake brood stock obtained from the Creston National Fish Hatchery. U.S. Fish and Wildlife Service. Creston. Montana.

Tests began in early to mid-April and continued through spring runoff - terminating in early July fish were kept in cube-shaped vessels, approximately 30 cm on a side, constructed with a wooden frame

covered with 3 mesh nylon netting. Vessels were suspended with floats, tethered to a fence post and positioned in low velocity areas near the edge of the current -typical rearing areas for young fish. The fry test containers were equipped with an automatic feeding device (Sweeney Enterprises Inc. Bourne, TX 78006) that included a timer, rheostat and battery.

Equipment was calibrated so that fry were fed commercial trout food four times daily; fingerlings were not fed during the tests. Tests included 40 fingerlings or 100 fry per vessel. Two vessels per site were used during fingerling tests.

Mortality was monitored three times weekly and alkalinity, hardness and pH were measured in the field each week (Table 1). Additionally, water samples were collected three Limes each week and analyzed for total recoverable, copper and zinc. A pair wise multiple comparison technique was used to determine if mortality in the mainstem was statistically different from the control site (Fleiss 1981).

Metals in fish organ

During April 1989, brown trout were sampled from five locations along the Clark Fork River and one location in Rock Creek. Sampling sites included reaches near Warm Springs, Sager Lane, Deer Lodge, Gold Creek and Beavertail. Rock Creek was sampled approximately two miles upstream from its mouth as a control site (Figure 1).

Approximately 30 adult brown trout were sampled from each site. Trout were weighed, measured and sacrificed in the field and liver, kidney and gill tissue were dissected, placed in glass containers and iced; tissues were frozen later the same day. At a given site, fish were intentionally selected to include a range of sizes. Mean lengths (mm) of fish sampled were: Warm Springs -328, Sager Lane -334, Deer Lodge -358, Gold Creek -324, Beaverk1il -331 and Rock Creek 372.

Prior to analyses, frozen tissues were vacuum freezed dried in a lyophilizer. Freeze dried tissues were microwave digested in concentrated nitric acid and analyzed for copper, cadmium and zinc using either induction coupled "plasma spectrophotometry or graphite furnace atomic absorption spectrophotometry. Quality control measures included analyses of duplicates, spikes, and reference materials (EPA fish samples and NBS bovine liver). The instrument was calibrated initially and recalibrated after every twenty samples using three reference standards and a blank. Each calibration was cross-checked with an EPA reference water sample.

Results and Discussion

During all four years of testing, mortality was observed In Silver Bow Creek and at several Clark Fork River sites that was significantly higher ($P<0.05$) than at control sites (Table 2). Mortality in Silver Bow Creek was at or near 100 percent during all four years of testing and was associated with extremely high concentrations of copper and zinc.

In some instances high mortality corresponded with low fish population densities. For example, the Beavertail section, where significant mortality occurred each year, supports less than fifty catchable brown trout per mile -the lowest trout density in the Clark Fork River (Johnson and Schmidt 1988). Similarly the Silver Bow Creek site, where mortality was nearly complete is unable to support trout. Comparatively, mortality at the Clinton site, downstream of the confluence with Rock Creek, was not

statistically different from the control- reflective of lower metals concentrations downstream of Rock Creek (Table 2).

High mortality in the Gold Creek section observed during 1987 was associated with a local thunderstorm that apparently washed mine wastes into the river. Most of the mortality occurred the day of the thunderstorm and coincided with a local fish kill in the table 2rnble 2briver. Water monitoring conducted the day after the storm was too late to detect elevated concentrations of metals.

Concentrations of copper at all sites except Clinton and zinc in Silver Bow Creek were in a range shown to cause chronic toxicity to laboratory fish populations (USEP A 1985). But the relationship between rates of mortality at some of the sites and concentrations of metals (measured as total recoverable) was not well defined. For example, in a given year measured concentrations of both copper and zinc were often highest at Warm Springs, yet rates of mortality were higher at downstream sites where metal concentrations were lower. Perhaps there are unmeasured variables that influence metals speciation and toxicity. Sampling may also have been too infrequent to measure acute events such as occurred near Gold Creek during a thunderstorm. Our observations point out the difficulty of assessing toxicity potential using only periodic water sampling.

Table 3) and have observed mortality. Bionomics (1979) tested the toxicity of water discharged from Warm Springs Pond-2 to early life stages of rainbow trout (eggs and fry) and to Daphnia middendorffiana -a native daphnid. Rainbow trout survival and hatchability were not reduced by exposure to pond water but all fry, including those exposed to dilutions of 50 and 75% pond water, experienced reduced growth. Copper and zinc concentrations in a 50% dilution of pond-2 watertable 3 averaged 25 and 65 ug/l, respectively. Additionally, Daphnia middendorffiana reproduction was significantly impaired by exposure to 100% pond water but not by exposure to 50% pond water (Bionomics 1979). Copper and zinc concentrations in 100% pond water were 33 and 77 ug/l, respectively. Identical tests with Daphnia magna produced a similar result (Bionomics 1978); numbers of young per female were reduced by exposure to 27 ug Cull and 31 ug Zn/l (measured as total recoverable).

Janik and Melancon (1982), during a site-specific water quality assessment of Silver Bow Creek and the upper Clark Fork, completed a few bioassay tests with Daphnia and bluegill. In these tests Daphnia were not adversely affected by Clark Fork River water nor was ventilation rate in bluegill. However, bluegill in Clark Fork River water showed evidence of acetylcholinesterase inhibition. Total and dissolved copper and zinc concentrations during the survey averaged 30 and 22 ug Cu/1 and 101 and91 ug Zn/l; the report did not include specific information on metal concentrations that were present in the bioassay water.

Parrish and Rodriguez (1985) tested the chronic toxicity of Clark Fork River water in the Deer Lodge vicinity to early life stages of rainbow trout including separate tests using green eggs, eyed eggs, and fingerlings. Tests were conducted in May and early June to coincide with run off, however, unusually dry spring conditions resulted in lower than normal streamflows and concomitantly low metals concentrations. Percent mortality of both eyed eggs and fingerlings were higher in 100% Clark Fork River water that in various dilutions of Clark Fork River water but results were not conclusive. During the test, acid soluble copper concentrations ranged from 10- 78 ug/l. For the water hardnesses that were present, EPA chronic and acute criteria for copper were calculated to be approximately 20 and 31 ug/1 respectively. It is noteworthy that most of the mortality occurred during the last week of the tests when

copper concentrations exceeded the acute criteria (when the weekly average concentration reached 7R ug Cu/l).

Concentrations of copper and zinc measured in previous tests where mortality occurred are similar to those measured during our tests (Table 2). Plausibly, copper is present in much of the Clark Fork during most years at concentrations that affect larval survival but not adults. Unpolluted tributaries and possibly springs that upwell in the mainstem may be necessary for recruitment to the population. Additional work is needed to identify spawning areas and spawning success in the Clark Fork.

Metals in Fish Organs

Cadmium, copper and zinc residues found in kidney, liver and gill of adult brown trout collected from various reaches of the Clark Fork River and Rock Creek are summarized in Figures 2-4. All metals concentrations are expressed on a dry weight basis.

Cadmium

Laboratory toxicity tests show that brook trout accumulate cadmium in liver, kidney, and gill and that residues increase with exposure concentration (Benoit et al. 1976). All tissues reached an equilibrium after approximately 20 weeks of exposure to cadmium.

The concentration of cadmium in organs of Clark Fork brown trout decreased from upstream to downstream. Cadmium concentrations in liver and kidney were higher than published reports of background concentrations (Benoit et al. 1976) but lower than those measured in brook trout exposed in the laboratory for 39 weeks (Benoit et al. 1976) to cadmium concentrations that were just above the chronic toxicity threshold of 3.4 ug Cd/1. Cadmium in gill was near or below reported background concentrations at all sites. Data suggest that Clark Fork River brown trout are exposed to cadmium concentrations that are above background but lower than required to elicit chronic toxicity. Concentrations of cadmium measured in the gills of Clark Fork fish which died during fish kills also indicate that cadmium is not as important as copper and zinc during acute toxicity events in the Clark Fork (Phillips 1985).

Zinc

Zinc concentrations in organs of Clark Fork River fish were similar at all sites sampled including the control site in Rock Creek (Figure 3). Concentrations in kidney and gill were higher and in liver similar to concentrations reported as baseline for rainbow trout (Goettl et al. 1971). Mount (1964) showed that the concentration of zinc in gill and kidney increased with exposure concentration but liver did not accumulate zinc. This may account for zinc concentrations in livers of Clark Fork fish being near reported background concentrations.

Water monitoring of Clark Fork River water conducted by the Water Quality Bureau (Gary Ingman, unpublished data) indicate that water quality criteria for zinc are only occasionally exceeded. Although zinc is present at high concentrations during fish kills and may contribute to acute mortality, residues of zinc in organs are not indicative of exposure to concentrations of zinc that would be cxpcc1cd 10 be chronically toxic.

Copper

Benoit (1975) observed that fish begin to accumulate copper in organs when exposed to concentrations at and above the chronic toxicity threshold, suggesting that any accumulation of copper may be indicative of exposure Lo chronically toxic concentrations. Liver has a tendency to retain accumulated copper (Solbe and Cooper 1976) thus liver residues of copper may be an index of chronic exposure to low concentrations.

Copper concentrations in liver and gill of Clark Fork fish were highest near Warm Springs and decreased downstream (Figure 4). Kidney copper concentrations were similar at all sites including the Rock Creek control site. Concentrations in liver were extremely high at all Clark Fork sites with average concentrations as high as 1663 ug/g at Warm Springs and] 517 ug/g at Sager Lane. These concentrations are much higher than those reported as background (McKim and Benoit 1974) and exceed by several-fold concentrations found in laboratory fish populations experiencing chronic toxicity due to copper (Benoit 1975). Rainbow trout exposed to 59ug Cu/l for 107 weeks accumulated a maximum concentration in Jiverof738 ug Cu/g (Goettl et al.)(1974). Bluegill exposed for 24 months to f62 ug Cu/l in water averaged 480 ug Cu/g in their livers. This concentration in water caused reduced survival, growth, and spawning success in adult bluegill. Concentrations in water as low 3S 40 ug Cu/l caused reduced surviva1 in larval bluegill.

Concentrations of copper in gill also exceeded those in gills of fish suffering from chronic copper toxicity (Benoit 1975). Solbe and Cooper (1976) found that fish eliminate copper rapidly from gill after return to copper free water. The high concentration of copper in gill from trout collected from the Clark Fork near Warm Springs indicate that they were being exposed to copper at the time they were collected. Indeed, copper concentrations in water were high near Warm Springs immediately before trout were collected for organ testing (Gary Ingman, unpublished data).

Results indicate that Clark Fork brown trout are exposed to large amounts of bioavailable copper. While there is no direct evidence that adult trout were adversely affected, residues of copper in liver are indicative of exposure to concentrations that have proven to be chronically toxic to sensitive life stages of fishes including salmonids.

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Figure 1: Map of the upper Clark Fork River basin showing sites for bioassays, fish organ sampling and habitat inventory work.

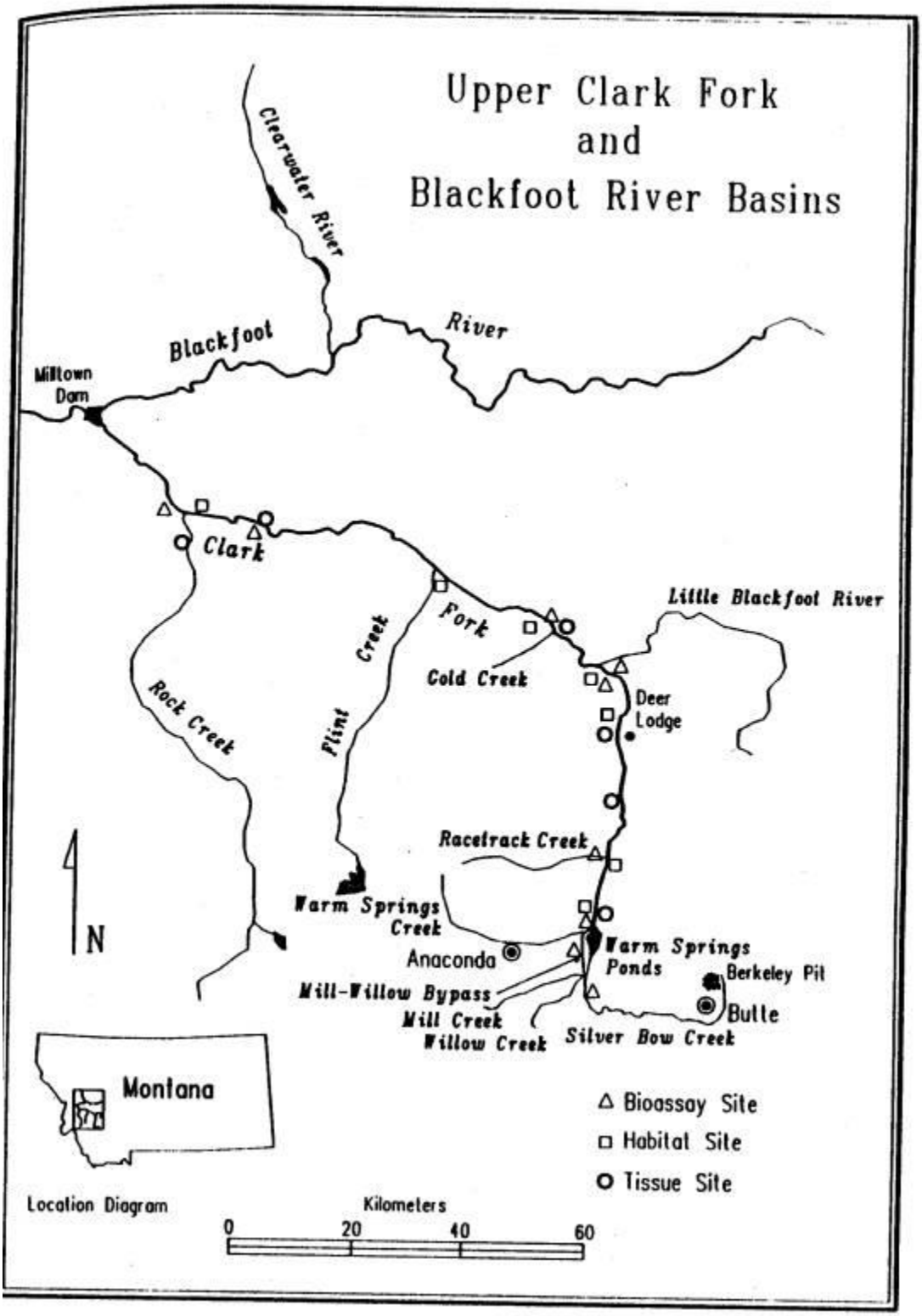

Table 1

Table 1. Alkalinity, hardness, and pH at nine locations on the Clark Fork River and tributaries during bioassays conducted from 1986 through 1988.

Location	n	Total alkalinity mg/l (as $CaCO_3$)			Hardness mg/l (as $CaCO_3$)			pH		
		mean	range	SD	mean	range	SD	mean	range	SD
Silverbow Creek	26	88	64-110	10	118	64-154	18	7.7	7.1-8.2	0.3
Mill-Willow Bypass	11	82	44-132	25	153	80-262	55	7.9	7.5-8.2	0.2
Clark Fork at Warm Springs	24	81	58-100	12	155	80-216	32	8.0	7.4-8.4	0.3
Racetrack Creek	26	103	28-140	25	97	28-122	23	7.5	6.9-7.7	0.2
Clark Fork at Deer Lodge	22	136	80-228	30	200	96-252	34	7.8	7.5-8.1	0.2
Little Blackfoot	11	107	86-136	17	104	80-130	16	7.8	7.7-7.9	0.1
Clark Fork at Gold Creek	26	127	76-184	23	167	94-216	28	7.7	7.2-8.1	0.3
Clark Fork at Beavertail	26	141	96-172	26	186	100-230	34	7.8	7.5-8.1	0.2
Clark Fork at Clinton	22	91	62-128	18	109	64-148	22	7.7	7.5-8.0	0.2

Table 2

Table 2. Results of instream toxicity tests in the Clark Fork drainage using fry and fingerling trout.

Location	Copper (ug/l) mean	Copper (ug/l) range	Zinc (ug/l) mean	Zinc (ug/l) range	Hardness (mg/l as $CaCO_3$) mean	Hardness (mg/l as $CaCO_3$) range	Cumulative % mortality fry	Cumulative % mortality fingerling
				1986				
Racetrack Creek (control)	5	(5-10)	7	(3-15)	83	(28-116)	--	0
Silver Bow Creek	201	(90-690)	381	(154-770)	84	(64-96)	--	89[a]
Clark Fork at Warm Springs	48	(5-160)	141	(35-693)	128	(80-200)	--	25[a]
Clark Fork at Deer Lodge	59	(20-140)	67	(24-130)	177	(96-224)	--	15[a]
Clark Fork at Gold Creek	55	(10-160)	60	(19-163)	145	(94-170)	--	7
Clark Fork at Beavertail	55	(5-170)	83	(24-223)	155	(110-184)	--	21[a]
Clark Fork at Clinton	28	(5-70)	44	(9-105)	103	(70-124)	--	3
				1987				
Racetrack Creek (control)	6	(5-10)	12	(6-24)	110	(98-122)	8	2
Silverbow Creek	219	(70-520)	478	(32-994)	116	(104-124)	92[a]	88[a]
Clark Fork at Warm Springs	28	(10-50)	99	(27-430)	169	(140-216)	18	7
Clark Fork at Gold Creek	14	(5-30)	35	(19-57)	172	(124-204)	36[a]	24[a]
Clark Fork at Beavertail	15	(5-40)	31	(4-54)	192	(100-228)	55[a]	12[a]
Clark Fork at Clinton	8	(5-20)	17	(2-30)	113	(64-148)	10	8

[a] Significantly different from the control at the 95% confidence level.

Table 2 (continued) Results of instream toxicity tests in the Clark Fork drainage using fry and fingerling Rainbow trout.

Location	Copper (ug/l) mean	Copper (ug/l) range	Zinc (ug/l) mean	Zinc (ug/l) range	Hardness (mg/l as $CaCO_3$) mean	Hardness (mg/l as $CaCO_3$) range	Cumulative % mortality fry	Cumulative % mortality fingerling
				1988				
Racetrack Creek (control)	1	(1-5)	8	(5-24)	95	(38-110)	0	--
Little Blackfoot River (control)	1	(1-2)	5	(5-10)	104	(80-130)	6	--
Silver Bow Creek	254	(17-2200)	605	(286-3740)	129	(102-154)	100[a]	--
Hill Willow Bypass	39	(5-220)	52	(9-284)	153	(80-262)	22[a]	--
Clark Fork at Warm Springs	30	(10-68)	57	(21-165)	154	(114-202)	7	--
Clark Fork at Beck Hill	32	(10-94)	44	(10-127)	209	(166-252)	6	--
Clark Fork at Gold Creek	24	(7-63)	34	(7-81)	175	(126-216)	5	--
Clark Fork at Beavertail	25	(3-71)	42	(5-109)	199	(160-230)	16[a]	--
Clark Fork at Clinton	17	(6-34)	36	(5-105)	107	(84-142)	0	--
				1989				
Racetrack Creek (control)	1	(1-3)	6	(5-22)	95	(42-118)	2	--
Silverbow Creek	--		--		--		68[a,b]	--
Clark Fork at Beck Hill	27	(10-70)	44	(19-83)	196	(160-236)	25[a]	--
Clark Fork at Gold Creek	18	(8-40)	28	(12-53)	157	(116-206)	33[a]	--
Clark Fork at Beavertail	19	(4-60)	37	(8-97)	180	(128-228)	64[a]	--
Clark Fork at Clinton	10	(3-20)	20	(6-38)	128	(98-224)	6	--

[a] Significantly different from the control at the 95% confidence level.

[b] 68% mortality occurred the first day of the test; the site was vandalized the second day.

Table 3

Table 3. Summary of bioassay results in the Clark Fork River drainage.

Date	Location	Test species and life stage	Mean metal conc. (ug/l)		Mean Hardness mg/l as $CaCO_3$	Response observed	Author(s)
			Cu	Zn			
August 26 - October 6, 1977	Pond-2 discharge	Daphnia magna, life cycle	27	31	624	fewer young/female	Bioanalysis (1978)
July 21 - October 21, 1977	Pond-2 discharge	rainbow trout, eggs and fry	29	36	642	no response	Bioanalysis (1978)
May 16 - July 4, 1979	Pond-2 discharge 50% dilution	rainbow trout, eggs and fry	25	65	270	reduced growth fry	Bioanalysis (1979)
May 16 - July 4, 1979	Pond-2 discharge	Daphnia middendorffiana, life cycle	33	77	310	reduced survival offspring	Bioanalysis (1979)
1982	Clark Fork, near ponds	bluegill	30	101	--	acetylcholinesterase inhibition	Janik and Halaneon (1982)
May 7 - June 6, 1985	Clark Fork, Deer Lodge	rainbow trout, green eggs	28	33	179	no response	Parrish and Rodriquez (1986)
May 7 - June 6, 1985	Clark Fork, Deer Lodge	rainbow trout, eyed eggs	28	33	179	14.5% mortality (control 5.5%)	Parrish and Rodriquez (1986)
May 16-22, 1985	SBC[a], near Colorado Tailings 18% dilution	Ceriodaphnia, life cycle	22	132	138	LOEL	Lazorchak (1986)
May 16-22, 1985	SBC[a], near Ramsey Flats 32% dilution	Ceriodaphnia, life cycle	45	220	138	LOEL	Lazorchak (1986)
May 16-22, 1985	SBC[a], above ponds 75% dilution	Ceriodaphnia, life cycle	55	197	115	LOEL	Lazorchak (1986)
May 16-22, 1985	Pond-2 discharge	Ceriodaphnia, life cycle	21	58	185	no response	Lazorchak (1986)
May 24- June 6, 1985	Clark Fork, Deer Lodge	rainbow trout, fingerling	39	43	172	20% mortality (control 7%)	Parrish and Rodriquez (1986)
May 25-June 1, 1987	SBC, above ponds 50% dilution	Ceriodaphnia, life cycle	195	390	--	LOEL	Himes (1987)

[a] SBC indicates Silver Bow Creek.
[b] LOEL indicates lowest observable effect level.

Figure 2: Cadmium concentrations (and 95% confidence intervals) in liver, kidney and gill from Brown trout taken from various reaches of the Clark Fork Basin and from Rock Creek.

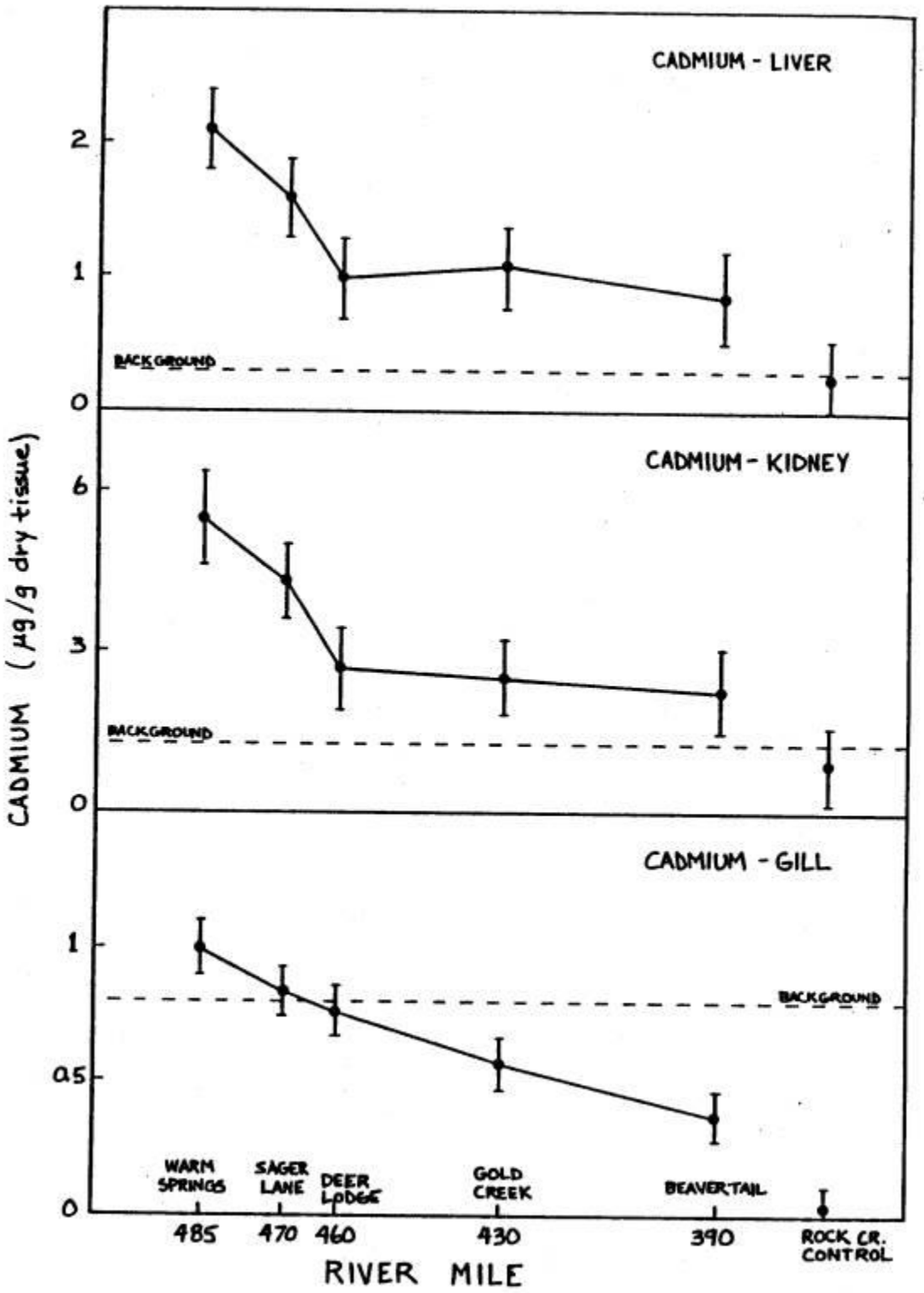

Figure 3: Zinc concentrations (and 95% confidence intervals) in liver, kidney and gill from Brown trout taken from various reaches of the Clark Fork Basin and from Rock Creek.

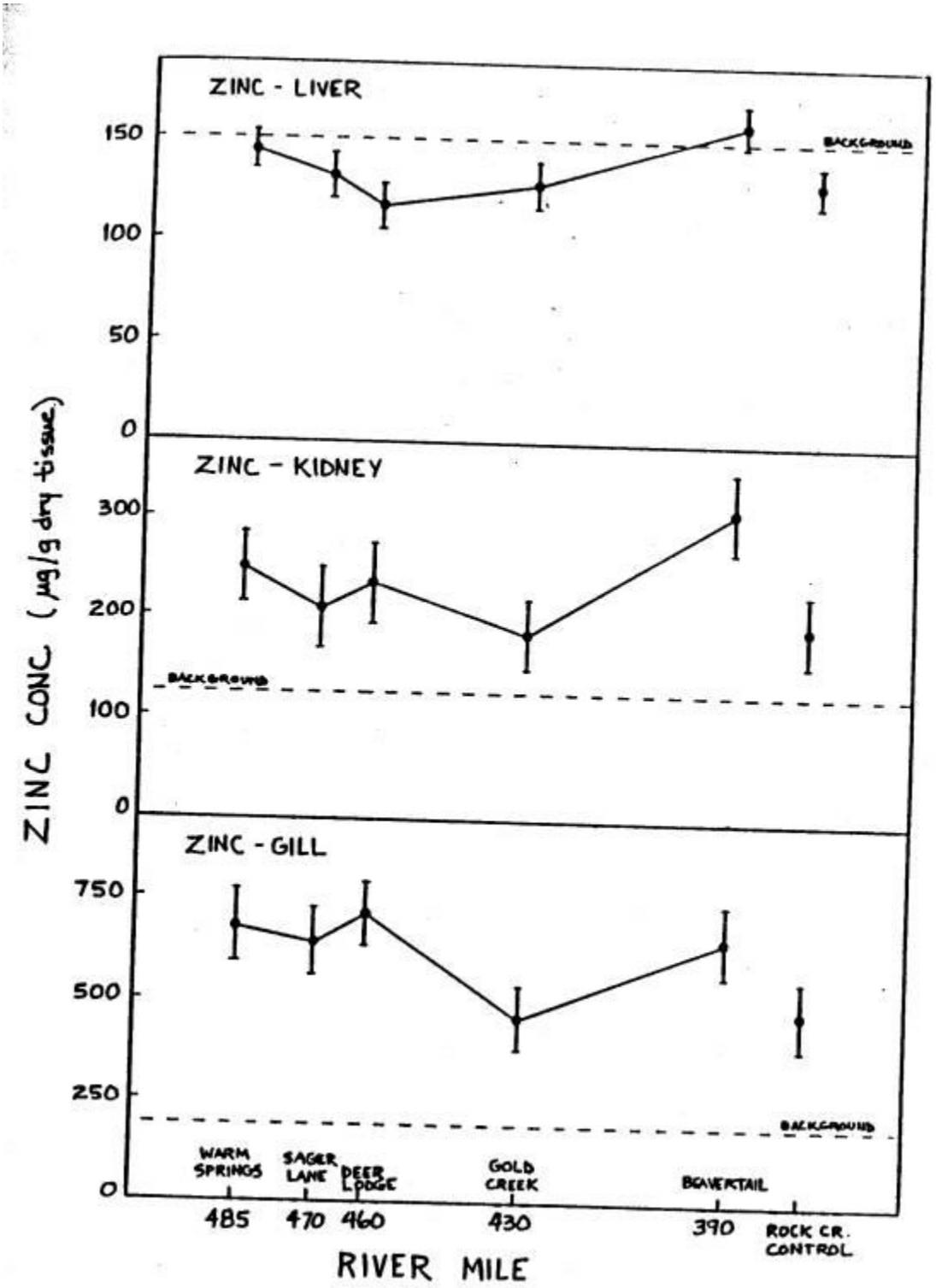

Figure 4: Copper concentrations (and 95% confidence intervals) in liver, kidney and gill from Brown trout taken from various reaches of the Clark Fork Basin and from Rock Creek.