AN EVALUATION OF THE POSSIBLE IMPACTS OF HEAVY METAL POLLUTION ON THE BROWN TROUT POPULATION OF THE UPPER ARKANSAS RIVER

Colorado Division of Wildlife
February 18, 1986

R. Barry Nehring

An evaluation of the relationship between chronic heavy metal (lead, zinc, copper, and cadmium) pollution of the upper Arkansas River and the brown trout population occurring there was completed. The study reveals lead and zinc bioaccumulation in liver and kidney tissues of brown trout does not occur to any significant degree. Brown trout liver tissues accumulate copper with both increasing exposure levels and exposure time. The liver copper bioaccumulation appears to be occurring via uptake through the food chain rather than osmotically across the gills or other absorptive tissues. Long-term exposure to extremely low levels of cadmium appear to pose the most significant detrimental impact on the brown trout population in the upper Arkansas River. Cadmium bioaccumulates in both liver and kidney tissue with both increasing exposure time and exposure levels. Statistically significant relationships are demonstrated between cadmium levels in the water over a 2-year period and depressed brown trout population densities over that same time period. Statistically significant relationships are demonstrated between brown trout population densities and liver and kidney cadmium bioaccumulation in 2- and 3-year-old brown trout. Statistically significant relationships exist between cadmium levels in the water and liver cadmium accumulation levels.

Background

Over the past decade many studies have been conducted on the aquatic life of the Arkansas River (LaBounty et al. 1975; Roline and Boehmke 1981; and Nesler 1983). These studies concentrated on the physical, chemical, and biological aspects of the upper Arkansas River, delving into the role of severe chronic heavy metal pollution and its impacts on the upper Arkansas River from above Leadville, Colorado, downstream to the confluence with Lake Creek.

The Colorado Division of Wildlife implemented a quality trout management program on sections of the Arkansas River from Salida downstream for more than 30 miles in 1981. Evaluations of the brown trout population in this reach of the Arkansas River indicated at Age 1 and 2, these trout were among the fastest growing, stream dwelling trout in Colorado (Nehring and Anderson 1981). It was believed the Arkansas River held excellent potential for providing a dense population of quality size (<14 in. and larger) and age brown trout under the proper management regime. Despite the implementation of a two-trout bag limit with a minimum size limit of 16 inches, the management program has not produced any increase in the numbers of quality size (<14 in.) brown trout since 1981. Evaluations have continued on an annual basis since 1981 (Nehring and Anderson 1981, 1982, 1983, and 1985; Nehring, Anderson, and Winters 1984) that have clearly documented the poor survival of brown trout in the Arkansas River beyond 3 or 4 years of age. Habitat degradation from siltation, a poor food supply, and heavy metal pollution have all been considered as contributing to the poor survival of trout in the Arkansas River. However, many streams with much more severe living conditions for trout with equally poor food supplies have not resulted in the near elimination of trout 4 years of age and older to the degree observed in the Arkansas River. Annual electroshocking surveys each March have revealed the majority of trout in the 12-16 inch size range were in very emaciated condition.

This perplexing problem, i.e. very rapid growth and good survival for the first 2 years of life, followed by a steep decline in annual growth rate, and poor survival after attaining sexual maturity by the brown trout, led us to examine the role of heavy metal pollution in the Arkansas River and to study the potential bioaccumulation of heavy metals by the brown trout. What follows is a synthesis of all of the data from the studies cited above, an intensive review of the pertinent literature on heavy metal toxicology as it pertains to salmonids, and a detailed analysis of the heavy metal (zinc, copper, lead, and cadmium) bioaccumulation in brown trout liver and kidney tissues. The purpose of this evaluation is two-fold. First, is it possible to determine if there are significant correlations between: (1) the heavy metal content in the Arkansas River; (2) the heavy metal content of the liver and kidney tissues of the Arkansas River brown trout; and (3) the brown trout population density throughout the Arkansas River? Secondly, if correlations do exist, is it possible to determine which metal(s) are the most cause for concern.

Results

Table 1 (attached appendices) presents the results of the heavy metal analyses for brown trout liver and kidney tissue samples collected at six

different sites from Salida upstream beyond Leadville. Table 2 describes each of the six sample sites, as well as the additional collections of rainbow and brown trout listed in the last five subsections of Table 1. The data in Table 1 is arranged from an upstream to downstream fashion. This data has been subjected to numerous two-sample t-tests to evaluate statistical significance in heavy metal bioaccumulation between stations (but within a single tissue and age group), as well as within stations (between age groups within the same tissue). The tests accomplish two things. First, if bioaccumulation occurs within a given tissue and age group between stations, is this related to increasing levels of population? Second, do certain metals bioaccumulate at a given station with increasing exposure time, i.e. as the fish get older?

Lead, zinc, copper, and cadmium are the four most abundant metal contaminants that are highly toxic to salmonids in the upper Arkansas River. Each of these metals will be discussed individually with respect to the level of bioaccumulation and potential for damage to the trout population in the Arkansas River.

Lead

Examination of the bioaccumulation data for lead in the liver and kidney tissues of brown trout (Table 1) reveals very low levels of lead in either tissue. Furthermore, there is no consistent pattern of lead accumulation, either within a tissue and sample site with increasing age, or between sample sites as one proceeds downstream into the most severely polluted areas. Goettl and Davies (1978) worked with rainbow trout in the laboratory over a complete life cycle and found lead accumulation in the kidneys averaged about 50 µg Pb/g after 48 weeks exposure to about 5-13 µg Pb/l (ppb or parts per billion). Liver accumulation of lead at that exposure level was only 5 µg Pb/g tissue dry weight (ppm or parts per million). At an exposure level of 55 µg Pb/l for 48 weeks, liver Pb accumulation averaged 40 µg Pb/g and kidney lead levels ranged up to 1,500 µg Pb/g on an average. Clearly, these levels of lead bioaccumulation in the laboratory studies are in the range of 100-300 times higher than our data from the Arkansas River at all stations. The water quality data of Roline and Boehmke (1981) clearly indicates total lead levels at most stations are high enough to warrant concern for serious bioaccumulation and potential toxic effects from lead to the trout. However, their liver analyses in 1979 also showed very little bioaccumulation with either exposure level or exposure time. Apparently, the lead is in a particulate suspended or complexed state such that it is not taken up osmotically (absorbed through the gills) nor taken in through the food chain. Ionic lead (Pb^{++}) is known to readily form insoluble precipitates with carbonate and bicarbonate ions that occur in all natural waters. Water flowing in a stream would, through turbulence, facilitate this precipitation process.

My conclusions are that the chronic lead pollution of the upper Arkansas River does not pose a serious threat to either the trout or the aquatic invertebrate life in this section of the river. Aquatic insects as a group are very tolerant of lead pollution (Nehring 1979).

Zinc

The bimonthly water sampling program of Roline and Boehmke (1981) from March 1979 through September 1980 clearly indicates that zinc is probably the most consistently prevalent of the four toxic heavy metals examined in this study. The data of LaBoundy et al. (1975) indicates zinc was the most prevalent highly toxic heavy metal in the early 1970's as well. Both of these studies revealed zinc levels in the 0.3-1.0 mg/l range or higher. Nehring and Goettl (1974) demonstrated that zinc was acutely toxic to rainbow, brown, cutthroat, and brook trout at 0.41, 0.64, 0.67, and 0.96 mg Zn/l in 14-day toxicity tests. Clearly, water in the upper Arkansas River carries zinc loads that run into the acute toxicity range for all four species of trout commonly found in streams in Colorado, especially for unacclimated fish. Trout eggs incubated in water containing zinc result in fingerling trout that are more resistant to zinc than fingerling hatched from non-zinc exposed eggs (Goettl, Sinley, and Davies 1974). This undoubtedly explains the ability of brown trout to exist in the upper reaches of the Arkansas River where zinc levels may exceed 1 mg Zn/l on a short-term basis.

Goettl et al. (1973) demonstrated that kidney zinc levels averaged 120-190 µg Zn/g from exposure levels ranging from 0.2 to 2.2 mg Zn/l. They found no correlations between liver zinc bioaccumulation with fish length, age, or exposure time. The liver and kidney zinc bioaccumulation data in this study (Table 1) confirms their conclusions. There is no apparent bioaccumulation of zinc in either liver or kidney tissue for given age fish between stations or for different age fish within a given station. Thus, neither exposure time nor exposure level appear to impact the level of zinc bioaccumulation in these tissues of brown trout in the Arkansas River. However, because of the high levels of zinc entering the Arkansas River from both the Leadville Drain and the Yak Tunnel (via California Gulch) the threat of acutely toxic levels of zinc is a threat to the trout population of the upper Arkansas River, upstream of the Lake Creek confluence.

My conclusion is that high chronic levels of zinc pollution in the upper Arkansas River pose a serious threat to the trout population on a short-term acute toxicity basis and should not be taken lightly.

Copper

Goettl and Davies (1976) recommended that the maximum safe level for copper in cold water was 10 µg Cu/l. Water quality analyses of Roline and Boehmke (1981) demonstrated that the copper levels in the Arkansas River between the inflow from California Gulch and the confluence with Lake Creek routinely ranged from 10-40 µg Cu/l at all stations from March 1979 through September 1980. Goettl, Sinley, and Davies (1974) demonstrated that copper bioaccumulates in the liver tissue of rainbow trout chronically exposed to copper levels of 3-37 µg Cu/l over a 48-week period. The maximum level of bioaccumulation of copper in the liver was 738 µg Cu/g at an exposure level of 59 µg Cu/l over a 107-week period. McKim and Benoit (1974) demonstrated there was no correlation between bioaccumulation of copper in brook trout's liver, kidney, gill, or muscle tissue at exposure levels below 10 µg Cu/l. Liver copper levels ranged between 208-239 µg Cu/g at an exposure level of 9 µg/l.

The bioaccumulation data on liver copper levels in Table 1 clearly demonstrates that copper does bioaccumulate at increasing levels of exposure between stations as one proceeds downstream to the more severely polluted collecting sites and within each individual collection site with increasing exposure times (older age trout). More importantly, the levels of copper bioaccumulation in the liver tissue of brown trout in the Arkansas River are 5-10 times as high as the levels observed in rainbow trout at comparable levels of exposure to waterborne copper pollution. The data in Table 1 indicates average copper bioaccumulation levels in the liver of 2- and 3-year-old trout in the 600-1,400 µg Cu/g range. Many individual samples at all stations were in the 2,000-4,000 µg Cu/g range.

Statistical comparisons between brown trout collected from the Arkansas River near the Mt. Shavano Rearing Unit with yearling rainbow trout from the hatchery and yearling rainbows stocked in the Arkansas River at Salida for 6 months clearly lead to the conclusion that the copper bioaccumulation is occurring through the food chain and not through osmotic uptake across the gills. Sixty to 75% of the water utilized in rearing rainbow trout on the Mt. Shavano Rearing Unit comes directly from the Arkansas River. Thus, if copper bioaccumulation were occurring through osmotic uptake, then the liver copper levels in the rainbow trout should not be much different from that of similar age brown trout collected in the Arkansas River near the hatchery. While there was a slight increase in the liver copper levels of rainbows held in the live-box at the time the Yak Tunnel spill passed the Mt. Shavano Hatchery compared to those rainbows sacrificed prior to the spill, the liver copper levels of yearling brown trout from the river contained more than three times as much copper as the yearling brown trout from the hatchery. Similarly, the Eagle Lake yearling rainbows stocked in the Arkansas River in May-June 1985 and sacrificed in late October 1985 had significantly higher liver copper levels compared to the hatchery rainbows. The hatchery rainbows had an average liver copper accumulation of 218 µg Cu/g compared to an average of 491 µg Cu/g in the liver tissue of rainbow trout that lived in the Arkansas River for approximately 6 months. These statistics clearly indicate the majority of the liver copper burden is apparently accumulating through the food chain.

A statistical comparison between yearling brown trout from the Arkansas River with yearling rainbows stocked in the Arkansas River indicated no significant difference in copper accumulation in the liver tissue, further substantiating the conclusion that copper accumulation in the liver is occuring primarily through the food chain.

Due to the obvious bioaccumulation of copper in brown and rainbow trout liver tissue to extremely high levels, and the chronic copper levels in the upper Arkansas River (from the Lake Fork to the Lake Creek confluence) running near the acute toxicity range, it appears that copper pollution is a more serious threat to the trout community because of the dual threat from both an acute and chronic exposure standpoint. On this basis, copper is probably a more serious pollution problem than zinc, and certainly more so than lead.

Cadmium

Cadmium is perhaps the most highly toxic heavy metal known to occur in the aquatic environment, at least as far as the toxicity to fish and aquatic insects is concerned. While the "safe" levels for aquatic life for both cadmium and silver are measured in the parts-per-trillion (nanograms/liter), cadmium, by virtue of its much greater solubility in cold water than most naturally occurring silver compounds, poses a much more serious threat to the aquatic ecosystem. It is also one of the most highly toxic heavy metals to humans.

Examination of the data in Table 1 clearly indicated that cadmium bioaccumulates in both liver and kidney tissue of brown trout within a given station over time and also between stations as one proceeds downstream into the more severly polluted sections of the Arkansas River. Statistical analyses indicated that kidney cadmium accumulation levels increased significantly at each succeeding station moving downstream through the first three sample sites (EF 1, EF 2, and Big Union Creek station). The Leadville Drain joins the East Fork of the Arkansas River above the EF 2 sample site, and California Gulch joins the Arkansas River upstream of the confluence with the Big Union Creek site. High levels of liver and kidney cadmium accumulation persist in the brown trout as far downstream as Salida and beyond. Even liver and kidney samples at Coaldale (almost 100 river miles downstream of Leadville) have easily detectible levels of cadmium (see Figures 3 and 4 in the Appendices).

Goettl and Davies (1978) found liver cadmium levels in rainbow trout averaged 13 µg Cd/g after three years of exposure to 0.5 µg Cd/l. Liver cadmium levels ranged up to 130 µg Cd/g and kidney cadmium levels ranged up to 180 µg Cd/g after two years of exposure to 3.5-7.1 µg/l. The water quality data of Roline and Boehmke (1981) show average cadmium levels in the Arkansas River in the 2-4 µg Cd/l range for the entire period from March 1979 through September 1980 from the confluence with California Gulch downstream to the Lake Creek confluence. Mean liver and kidney cadmium accumulation levels in the brown trout from the Arkansas River collection sites for Age 1-, 2-, and 3-year-old trout were plotted over the accumulation and exposure levels for rainbow trout tested in the laboratory by Goettl and Davies (1978), as shown in Figures 1 and 2 in the Appendices. Not surprisingly, with the exceptions of the Ossman Ranch and Coaldale collection sites, virtually all of the brown trout cadmium accumulation data falls exactly within the ranges for age and exposure levels of the rainbow trout in the laboratory studies. The brown trout data is even more closely correlated with the exposure levels of about 1.7-3.5 µg Cd/l in the laboratory studies, almost exactly within the average range of cadmium levels in the Arkansas River over an 18-month period as shown by Roline and Boehmke (1981). Long-term studies on the toxicity and bioaccumulation of cadmium in brook trout (Benoit et al. 1976) very closely approximate the results of Goettl and Davies (1978) with rainbow trout. These similarities strongly indicate that cadmium acts very much the same in all three species (brown, brook, and rainbow) of trout.

There is one instance where the response of Arkansas River brown trout exposed to cadmium does not follow the responses seen in the laboratory experiments with rainbow and brook trout at similar exposure levels. In every

instance where we were able to find a 4-year-old brown trout, they had significantly less cadmium accumulation in the liver and kidney tissues. This indicates that only those trout with a significantly lower cadmium burden in the liver and kidneys survive beyond age four.

DISCUSSION

Our electroshocking studies of brown trout in the Arkansas River reveal that we are losing large numbers of brown trout between the third and fourth year of life, the first year at which most of the trout in the population are attaining sexual maturity. Brown trout, being fall spawners, generally cease feeding early in the fall with the onset of sexual maturity. Males begin to ripen in September, while females ripen in October, with spawning usually commencing in late October-early November in much of Colorado. The onset of winter brings near freezing water temperatures and very low metabolic rates for the trout from November through March of the next year. Our "hypothesis" is that the additional stress from excessive heavy metal accumulations in vital body organs such as the liver and kidney is the one additional stress (in addition to starvation through the spawning period, a long winter of cold water temperatures, a poor forage base, and low body fat and energy reserves) that the trout cannot survive. The question is, does the literature on laboratory studies in heavy metal toxicology contain evidence that lends credence to our excessive stress "hypothesis"?

There is considerable evidence from the literature that points to the stressful response in a wide variety of fish upou exposure to extremely low levels of cadmium in water. Cearley and Coleman (1974) found largemouth bass exhibited very erratic behavior patterns after 12 weeks of exposure to only 8 µg Cd/l. They began swimming erratically, had muscle spasms, convulsions, loss of equilibrium, with alternating periods of quiescence and paralysis. Hiltebran (1971) demonstrated the depressing effects of cadmium on oxygen and phosphate metabolism in bluegill liver mitochondria, ultimately depressing energy production. Rowe and Massaro (1974) demonstrated that cadmium chloride introduced into the digestive tract of white catfish was ultimately accumulated in liver and kidney tissue. Mount and Stephan (1967) reported that cadmium bioaccumulated in the liver tissue of bluegills and brown bullheads. Eaton (1974) demonstrated that chronic toxicity of cadmium in bluegills began after 8 months exposure to 80 µg Cd/l and occurred concomitant with the onset of spawning activity. The liver tissues from bluegills that died during spawning showed liver bioaccumulations of 200-500 µg Cd/g. Kidney cadmium accumulation ranged from 188 µg/g at exposure levels of 31 µg/Cd/l to 218 µg Cd/g at 240 µg/l.

Pickering and Gast (1972) demonstrated that cadmium was lethal to fathead minnows at 55 µg Cd/l in a chronic (long-term) toxicity test. Jackim et al. (1970) demonstrated that cadmium was a strong inhibitor of alkaline phosphatase activity in the liver tissue of the killifish, Fundulus heteroclitus. Flagfish exposed to 16 µg Cd/l showed severe hyperactivity during spawning, with males more severely distressed than females. Pickering and Gast (1972) found fathead minnows exposed to 110 µg Cd/l exhibited severe nervous disorders with males more adversely affected than females. Benoit et al. (1976) found that all male brook trout died during spawning after exposure to only 3.4 µg Cd/l over two different generations. Both liver and kidney tissues of brook trout revealed signifiant levels of cadmium accumulation from exposure to 3.4 µg Cd/l for a period of less than one year, with the average

liver accumulation of 10 µg/g and the average kidney accumulation of 50 g Cd/g. Measurable levels of liver and kidney cadmium bioaccumulation were found at exposure levels of 60 µg Cd/l (60 parts per trillion).

Goettl and Davies (1976) demonstrated measurable toxic effects of cadmium on rainbow trout over longer-term exposure to 7.1 µg Cd/l for rainbows that were incubated in cadmium containing water. The "effect - no effect" range for rainbows hatched from unexposed eggs was between 1.5 and 0.7 µg Cd/l. Studies have shown that in brook and rainbow trout, cadmium bioaccumulation in liver and kidney tissue is an irreversible process. Removal of cadmium-contaminated fish to a cadmium-free water supply does nothing to reduce the cadmium burden in renal and hepatic tissue, even after months in cadmium-free water.

All of the above clearly demonstrates the extremely toxic effects of cadmium to a wide variety of fish at extremely low levels of exposure. Many of the above studies demonstrated that exposure to very low levels of cadmium manifested itself in death during the onset of spawning activity, especially among the males, giving credence to the hypothesis that the brown trout in the Arkansas River may be succumbing to the excessive liver and kidney cadmium accumulation during the post-spawning winter stress period when body fat and energy reserves are most depleted.

Having adequately demonstrated that: 1) cadmium bioaccumulates in trout liver and kidney tissue over a long period of time from very low levels of exposure; and 2) that it is toxic to trout at the chronic exposure levels known to exist in the upper Arkansas River documented by Roline and Boehmke (1981), it appears worthwhile to see if any statistical relationships exist between cadmium in the water, cadmium in the liver and kidney tissue of brown trout, and (most importantly) if either of these two sets of data show any correlation with the observed brown trout populations in the upper Arkansas River. The data used in these statistical regression analyses is presented in Table 3.

In Section A of Table 3, the average cadmium content of the water from March 1979-September 1980 (from Roline and Boehmke 1981) is the X variable and is regressed against the brown trout population estimates (Y variable) at six different sample locations that correspond exactly or very closely to the water sample stations. Since the relationship between X and Y is curvilinear, the population variable (Y) was transformed to natural logarithms. The correlation coefficient (r) was -0.9261 and the coefficient of determination (r^2) was 0.86. These two statistical measures indicate that there is: 1) a highly negative relationship between the cadmium level in the water in 1979 and 1980, and the brown trout population estimates taken from August 1977 through April 1980; and 2) that 86% of the variation in the population estimates is explained by the variation in the cadmium measured in the water.

In Section B of Table 3, cadmium accumulation in the liver tissue of 2- and 3-year-old brown trout (X variable) is regressed against the brown trout population estimates completed from October 23-November 5, 1985 (Y variable). Logarithmic regression analyses (natural logarithm transformation on the population estimate) resulted in a correlation coefficient (r) of -0.8911 and -0.7496, for age two and three brown trout, respectively.

Similarly, a logarithmic regression analysis of the cadmium accumulation in 2- and 3-year-old brown trout kidney tissue regressed against the population estimates for October-November 1985 resulted in correlation coefficients (r) of -0.9443 and -0.9695, respectively. The coefficient of determination (r^2) between kidney cadmium accumulation and brown trout population density was 0.89 and 0.94, for 2- and 3-year-old trout, respectively. In other words, 89% of the variation in the brown trout population density was accounted for by the variation in the kidney cadmium accumulation in 2-year-old brown trout and 94% of the variation in the population density was accounted for by the variation in the kidney cadmium accumulation in 3-year-old brown trout.

Finally, is the cadmium accumulation in the liver tissue correlated with the cadmium content of the water in the upper Arkansas River? Although we do not have simultaneous long-term water quality data available at enough stations in the upper Arkansas drainage with which to correlate liver cadmium accumulation data, it is not unreasonable to assume (after examining the averages and range of fluctuations observed over 18 months in 1979-80 by Roline and Boehmke) annual average cadmium levels in the water do not vary that much from one year to the next at each station. Thus, we correlated the cadmium content of the 2-year-old brown trout liver tissues (Table 3-C) from October-November 1985, with the water quality data on cadmium levels form the corresponding sampling stations in 1979-80. Not too surprisingly, the correlation coefficient was +0.9961 and the coefficient of determination was 0.992, indicating that 99% of the variation in cadmium content of the liver tissue for 2-year-old brown trout was accounted for by the variation in the cadmium content of the water over the five sample stations from the Ossman Ranch to the Otero Pump Plant.

Regression analyses were run on the brown trout population data versus the lead, copper, and zinc bioaccumulation in liver and kidney tissues. None of the correlation coefficients of determination explained more than 35% of the variation in the brown trout population density, at best. Most of the r^2 values were in the 0.008 to 0.24 range.

The data analyses above indicate very strongly that the long-term exposure of brown trout in the Arkansas River to very low levels of cadmium is having a very detrimental impact on the brown trout population density. It also indicates very strongly that the impact is being felt the most on the older spawning age fish.

Table 2. Description of sampling sites and their relationship to study sites of LaBounty et al. (1975) and Roline and Boehmke (1981).

DOW Station Name and approximate location	Description and Other pertinent data
Ossmann Ranch (1)*	The least polluted collection site (from a heavy metal standpoint). About 4 miles NE of Leadville above the confluence with the Leadville Drain, just downstream of the Hwy. 24 Bridge at the Ossmann Ranch (west of highway). Corresponds to EF 1 of Roline and Boehmke 1981.
East Fork of the Arkansas River downstream of the Leadville Drain (2)	Corresponds very closely to EF 2 of Roline and Boehmke (1981). This station is downstream of the Leadville Drain (an important source of heavy metal pollutants to the Arkansas River) but upstream of the confluence with Tennessee Creek.
Arkansas River at Big Union Creek (3)	This collection site lies between AR 5 and AR6 of Roline and Boehmke (1981), downstream of the confluence with the Lake Fork of the Arkansas but several miles upstream of the Lake Creek confluence.
Arkansas River 1/2 mi. above the Lake Creek confluence (4)	Corresponds very closely to AR 8 of Roline and Boehmke (1981) upstream of the confluence with Lake Creek (Twin Lakes outlet).
Arkansas River 1/2 mi. below the Otero Pump Station (5)	Approximately 12 miles downstream from the confluence with Lake Creek, 1/2 mile downstream of the Bureau of Reclamation Otero Pump Plant.
Arkansas River at Salida--catch-and-release section (6)	This collection site begins at the Stockyard Bridge, approximately 1 mile east of Salida on U.S. Hwy. 50.
Pre-Spill, Arkansas River at Salida 10/23/85	Brown trout were collected from the Arkansas River at the Mt. Shavano State Fish Rearing Unit and put into a live box to document any mortality of trout in the river as the Yak Tunnel spill of October 1985 occurred.
Post-Spill (from live-box) Arkansas River at Salida 10/28/85	Brown trout collected prior to the spill and placed in the live box were sacrificed for kidney and liver heavy metal accumulation after the Yak Tunnel spill of 10/85 had passed.

Table 2. Description of sampling sites and their relationship to study sites of LaBounty et al. (1975) and Roline and Boehmke (1981) (continued).

Station Name and approximate location	Description and Other pertinent data
Hatchery rainbows - Pre-spill from hatchery, 10/22/85	Catchable size (yearling) rainbow trout sacrificed from the Mt. Shavano Fish Rearing Unit for analysis of heavy metal accumulation in the liver and kidney tissue prior to the passing of the Yak Tunnel spill of 10/85.
Hatchery rainbows from live-box post-spill, 10/28/85	Hatchery rainbows placed in a live box in the Arkansas River prior to the arrival of the Yak Tunnel spill of 10/85 and sacrificed for comparative heavy metal content after the spill had passed.
Eagle Lake rainbows - stocked in Arkansas River for about six months	Rainbow trout from the Mt. Shavano Hatchery stocked in the Arkansas River in the catch-and-release section in May or June 1985. Sacrificed for comparative evaluations of liver and kigney bioaccumulation of heavy metals.
Coaldale (7)	Brown trout collected from the Arkansas River downstream of Coaldale, Colorado in March 1984 for analysis of liver and kidney tissues for heavy metal accumulation.
Controls	Brown trout collected from the South Fork of the Rio Grande in April 1984 for comparative analyses of liver and kidney heavy metal accumulations. This tributary of the Rio Grande near South Fork, Colorado is not impacted by base metal mining activities.

*The numbers in parentheses indicate the number of the stations as given in Figures 3, 4, and 5 in the appendices.

Table 3. Regression analyses of the relationships between: 1) cadmium content of the water in the upper Arkansas River; 2) brown trout population densities; and 3) cadmium bioaccumulation in the liver and kidney tissues of brown trout in the upper Arkansas River.

Section A

Station	Mean cadmium content in water (79/80) (µg Cd/1)	Population sampling date month/year	Brown trout population estimate No./hectare
EF 1	0.73	10/79	4,407
EF 2	3.02	10/79	421
AR 1	2.24	8/77	1,269
AR 2	1.32	10/79	1,011
AR 7	3.53	8/77	276
AR 9	2.92	4/80	320

Section B

Station	Brown trout Population Estimate	Age 2 liver Cd (µg Cd/g)	Age 3 liver Cd (µg Cd/g)	Age 2 Kidney Cd (µg Cd/g)	Age 3 Kidney Cd (µg Cd/g)
EF 1	2,184	9.96	7.60	20.8	11.2
EF 2	919	21.8	35.0	47.0	54.7
AR 8	152	25.7	33.6	156	183
AR 9	198	22.1	27.1	108	106
Salida	393	17.1	14.1	65.4	68.3
Log r (correlation coefficient)		-0.8911	-0.7496	-0.9443	-0.9695
r^2 (coefficient of determ.)		0.794	0.562	0.892	0.940

Section C

Station	Mean cadmium content in water (79/80)	Age 2 liver cadmium (µg Cd/g)
EF 1	0.73	9.96
EF 2	3.02	21.8
AR 6	3.60	26.7
AR 8	3.59	25.7
AR 9	2.92	22.1

Correlation coefficient - $r = +0.9961$
Coefficient of determination - $r^2 = 0.99$

TABLE 1. HEAVY METAL ACCUMULATION IN LIVER AND KIDNEY TISSUE FROM TROUT IN THE ARKANSAS RIVER, 1985

Tissue	Age	Copper (µg/g) N	Copper (µg/g) X	Copper (µg/g) S.D.	Cadmium (µg/g) N	Cadmium (µg/g) X	Cadmium (µg/g) S.D.	Lead (µg/g) N	Lead (µg/g) X	Lead (µg/g) S.D.	Zinc (µg/g) N	Zinc (µg/g) X	Zinc (µg/g) S.D.
At Ossmann Ranch													
Liver	1	17	240	129	14	5.72	5.60	12	12.0	19.9	17	335	108
Liver	2	24	998	978	24	9.96	7.01	20	1.58	1.24	23	235	83.9
Liver	3	2	566.4	61.9	2	7.60	1.70	1	0	—	2	320	14.5
Kidney	1	13	43.2	20.5	17	11.7	14.2	13	9.07	5.97	17	363	229
Kidney	2	24	34.2	18.2	24	20.8	13.0	19	4.66	3.88	23	269	163
Kidney	3	2	29.1	2.10	2	11.2	7.99	2	6.45	2.62	2	278	113
East Fork of Arkansas River Downstream of Leadville Drain													
Liver	1	32	301	237	31	14.7	7.68	9	4.57	3.71	32	572	290
Liver	2	23	460	276	23	21.8	11.0	14	1.56	1.05	23	438	154
Liver	3	2	862	489	2	35.0	24.1	2	0.650	0.495	2	427	58.1
Kidney	1	32	90.6	85.5	32	30.7	15.8	8	6.13	3.37	32	464	219
Kidney	2	23	34.2	16.7	23	47.0	30.3	18	3.64	1.67	22	352	260
Kidney	3	2	30.7	5.94	2	54.7	33.0	2	2.05	0.919	2	214	62.4
Arkansas River at Big Union Creek Confluence Below Lake Fork Confluence													
Liver	1	3	521	106	3	28.0	14.6	3	1.43	0.929	3	277	81.5
Liver	2	14	670	245	14	26.7	12.5	11	2.14	1.33	13	442	372
Liver	3	1	814	—	1	43.8	—	1	4.90	—	1	321	—
Kidney	1	3	487	640	3	134.6	91.0	3	4.53	3.49	3	316	48.0
Kidney	2	13	63.1	37.2	13	156.0	65.8	13	8.81	2.72	13	350	99.9
Kidney	3	1	83.5	—	1	183	—	1	11.8	—	1	515	—
Arkansas River One-Half Mile Above Lake Creek Confluence													
Liver	1	6	416	160	6	12.8	6.56	5	3.08	2.48	5	580	528
Liver	2	29	661	593	29	25.7	21.8	28	2.56	1.76	27	416	220
Liver	3	10	1224	860	10	33.6	27.1	10	4.70	3.14	10	336	171
Liver	4	—	—	—	—	—	—	—	—	—	—	—	—
Kidney	1	6	63.7	15.8	6	51.8	25.9	6	9.45	9.01	6	451	154
Kidney	2	30	45.7	24.3	30	66.2	38.8	30	12.6	13.0	30	553	442
Kidney	3	12	54.2	34.6	12	81.7	37.1	12	15.4	13.8	12	693	776
Kidney	4	1	38.8	—	1	50.4	—	1	4.6		1	233	—
Arkansas River One-Half Mile Below the Otero Pump Station													
Liver	1	1	376	—	1	18.3	--	1	0	--	1	318	—
Liver	2	8	649	690	8	29.3	21.2	8	3.21	3.88	8	253	67.1
Liver	3	7	620	340	7	27.1	7.28	7	2.23	1.00	7	467	475
Liver	4	1	1877	—	1	43.3	—	1	1.00	—	1	312	—
Kidney	1	1	56.5	—	1	88.2	—	1	11.3	—	1	350	—
Kidney	2	7	26.6	8	7	108.0	57.1	7	9.3	10.2	7	459	348
Kidney	3	6	26.8	13.4	6	105.8	41.8	6	12.7	9.87	5	590	682
Kidney	4	1	19.0	—	1	73.1	—	1	22.1	—	1	522	—
Arkansas River At Salida Catch/Release Section													
Liver	1	3	337	186	3	10.3	0.61	2	2.60	1.70	3	1710	1296
Liver	2	26	1307	893	26	26.3	15.7	24	2.69	1.90	26	433	303
Liver	3	17	1381	998	17	28.9	20.8	15	3.03	2.00	17	748	1257
Kidney	1	3	37.8	23.9	3	20.9	8.14	3	3.23	1.46	3	229	33.4
Kidney	2	26	26.4	14.0	26	65.3	43.2	24	9.16	6.13	26	288	105
Kidney	3	17	24.2	7.84	17	68.4	41.9	17	9.41	5.68	16	293	148
Pre-Spill, Arkansas River at Salida, 10/23/85													
Liver	1	4	673	137	3	16.8	12.4	4	3.07	1.77	—	—	—
Liver	2	14	788	261	14	17.4	6.37	9	2.49	2.60	—	—	--
Liver	3	4	1140	397	4	16.2	3.28	3	1.10	0.721	—	—	—
Kidney	1	4	35.7	31.9	4	74.9	74.6	3	4.07	3.44	—	—	—
Kidney	2	12	78.8	14.1	14	65.4	34.6	12	6.72	4.99	—	—	--
Kidney	3	4	60.2	36.4	4	60.3	16.0	4	6.47	2.64	—	—	—
Post-Spill (From Livebox), Arkansas River At Salida, 10/28/85													
Liver	1	1	1268	—	1	42.3	--	1	6.50	--	—	--	—
Liver	2	12	879	222	12	17.1	11.6	12	2.22	1.42	—	—	--
Liver	3	2	789	11.0	2	9.05	4.60	1	0.6	--	--	--	--
Liver	4	2	675	573	1	0	—	1	0.3	—	—	—	—
Kidney	1	1	45.9	--	1	82.7	--	--	--	—	--	--	--
Kidney	2	12	26.6	21.9	12	57.2	32.5	12	5.37	3.08	--	—	--
Kidney	3	2	19.7	11.7	2	43.2	40.6	2	3.75	2.62	—	—	--
Kidney	4	2	6.4	0.28	1	0	—	2	1.05	0.636	--	—	--
Hatchery Rainbows - Pre-Spill From Hatchery, 10/22/85													
Liver	1	17	218	88.2	17	0.410	0.113	--	--	—	—	—	—
Kidney	1	17	15.4	4.24	17	1.45	0.408	--	--	—	--	—	—
Hatchery Rainbows from Livebox Post-Spill, 10/28/84													
Liver	1	16	278	77.7	16	0.675	0.710	--	--	—	—	--	—
Kidney	1	16	13.7	9.77	16	2.51	3.98	—	—	—	--	—	—
Eagle Lake Rainbows Stocked in Arkansas River for 6 Months													
Liver	1	15	491	161	15	3.50	0.835	8	2.32	2.16	15	449	143
Kidney	1	15	36.0	16.7	15	13.0	9.17	8	4.12	2.49	15	364	204

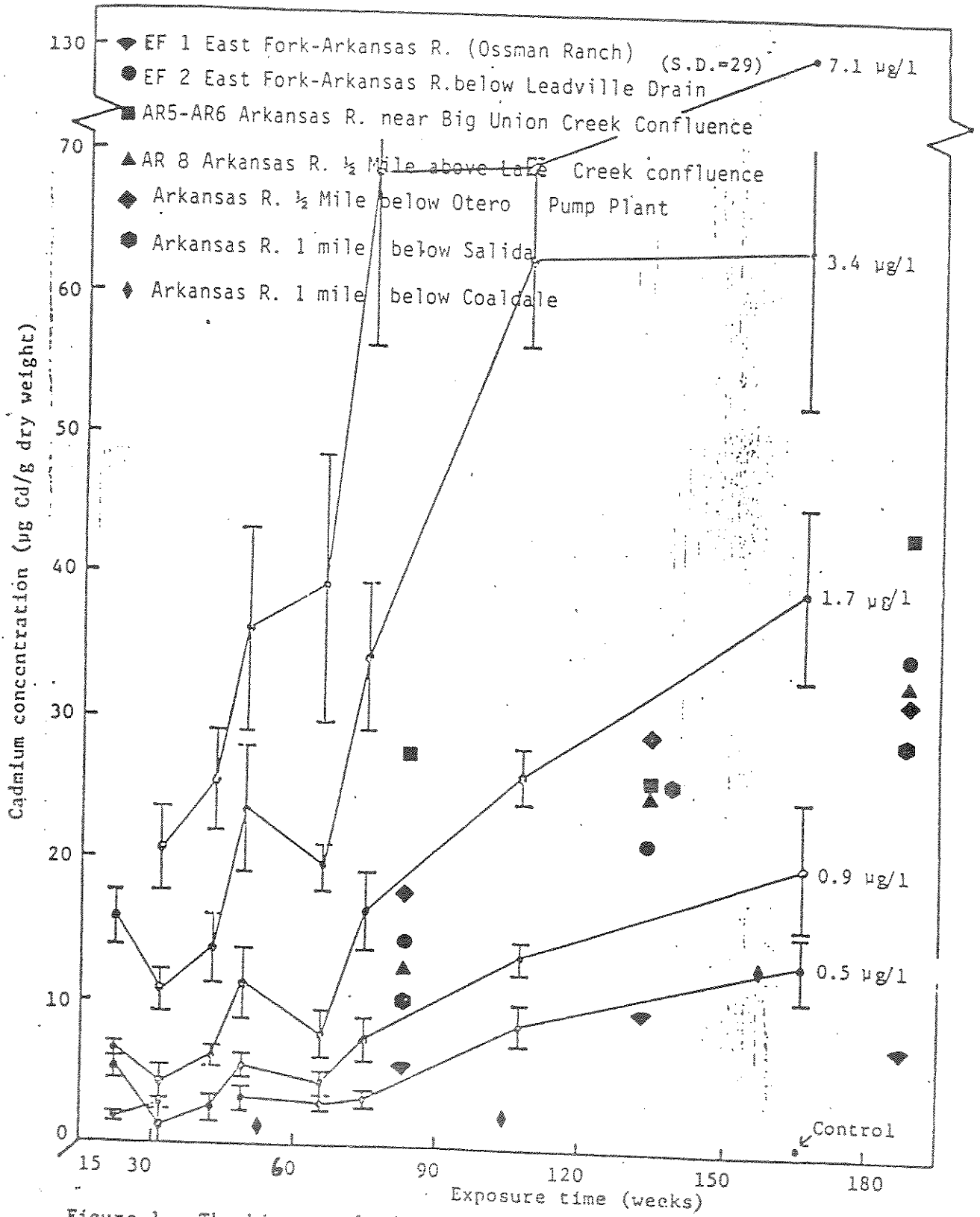


Figure 1. The bioaccumulation of cadmium in the livers of rainbow trout exposed to cadmium for periods up to 166 weeks. Each bracketed point represents the mean ±0.5 standard deviation (S.D.) of the mean. (With Arkansas R. brown trout liver cadmium bioaccumulation over time).

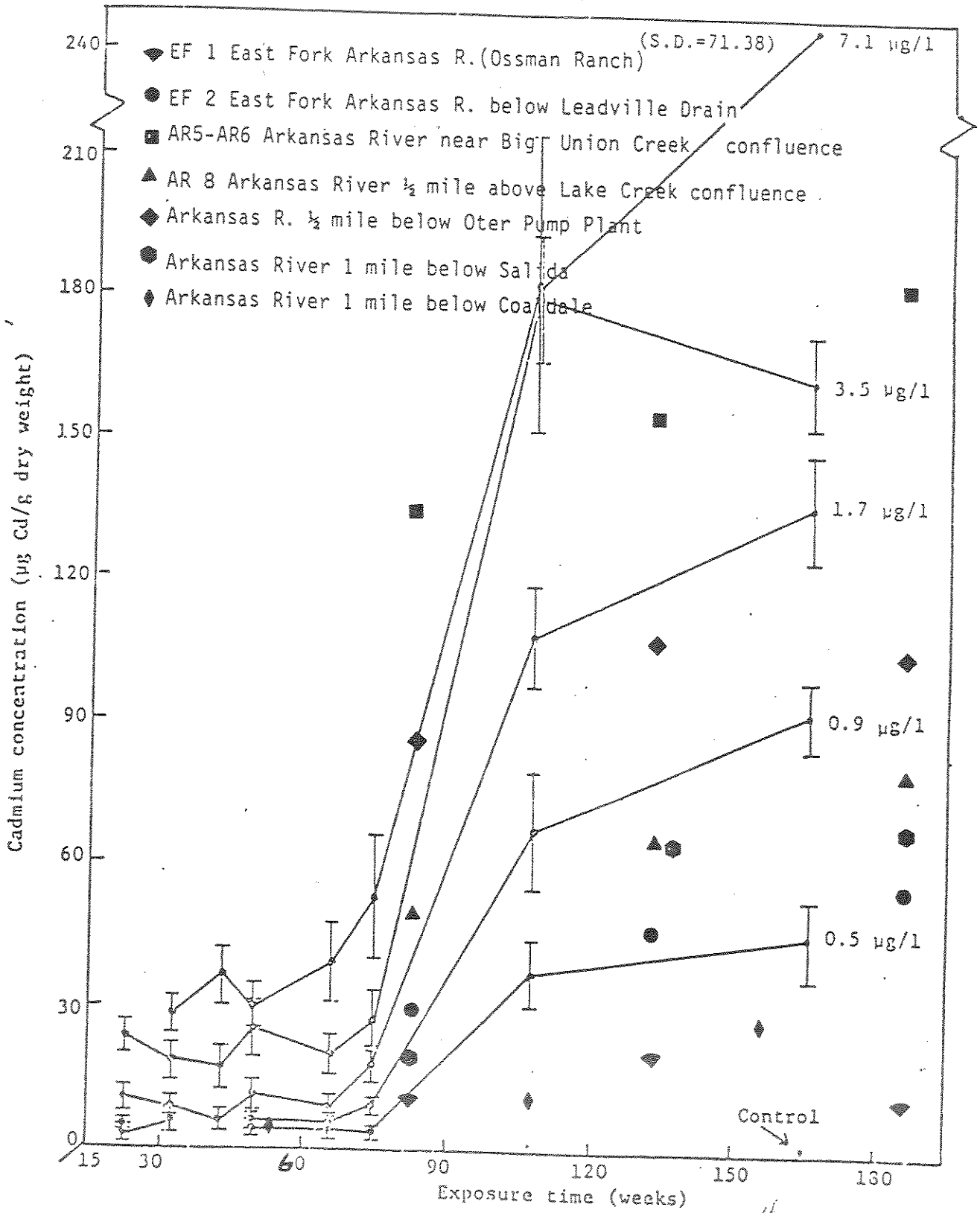


Figure 2. The bioaccumulation of cadmium in the kidneys of rainbow trout exposed to cadmium for periods up to 166 weeks. Each bracketed point represents the mean ±0.5 standard deviation (S.D.) of the mean.(With Arkansas R. brown trout kidney cadmium bioaccumulation over time).

FIGURE 3. BROWN TROUT (SALMO TRUTTA) LIVER CADMIUM ACCUMULATION OVER TIME (FISH AGE) AND DISTANCE (NUMBER FROM UPSTREAM TO DOWNSTREAM) IN THE ARKANSAS RIVER FROM ABOVE LEADVILLE TO COALDALE.

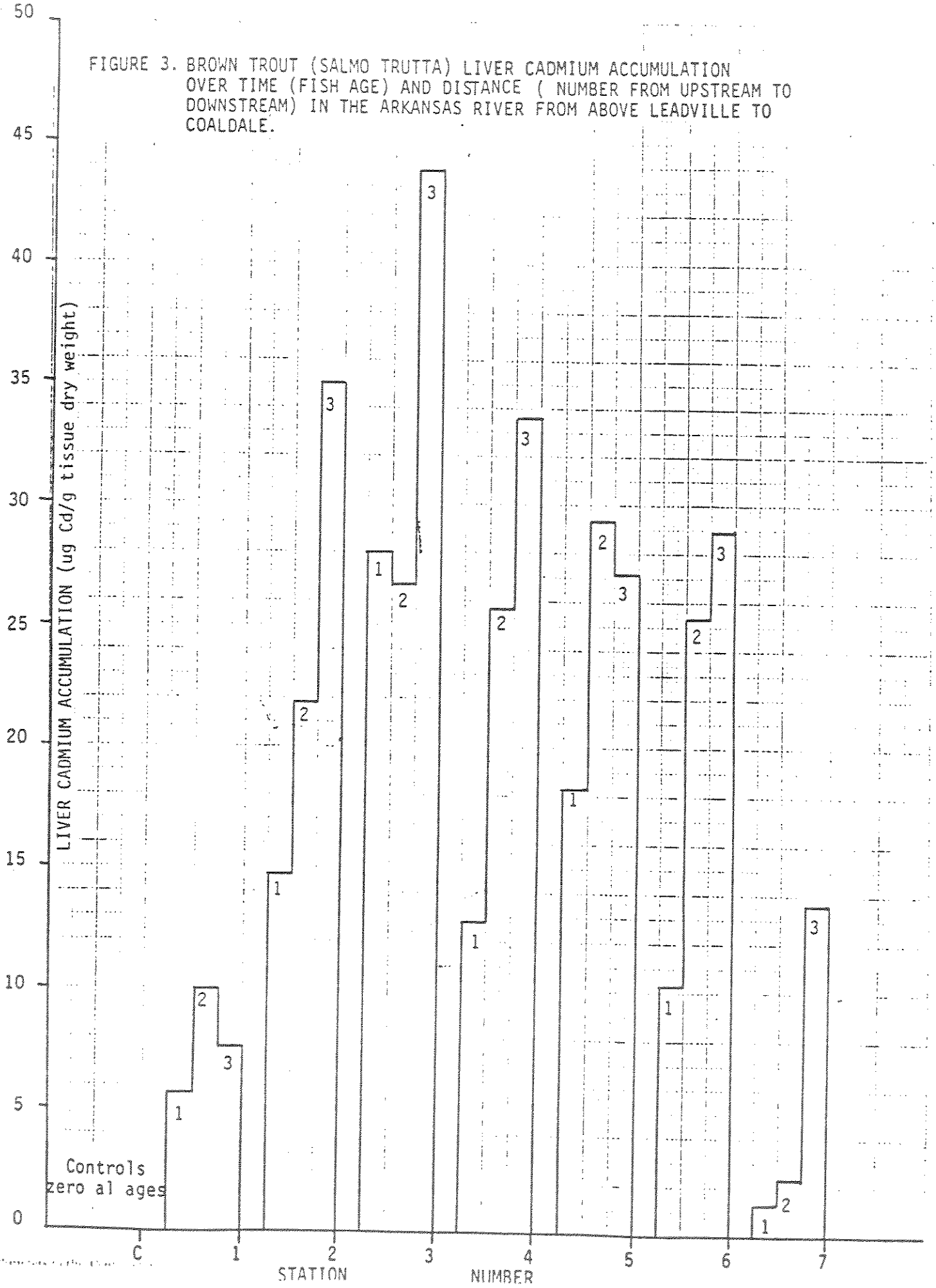
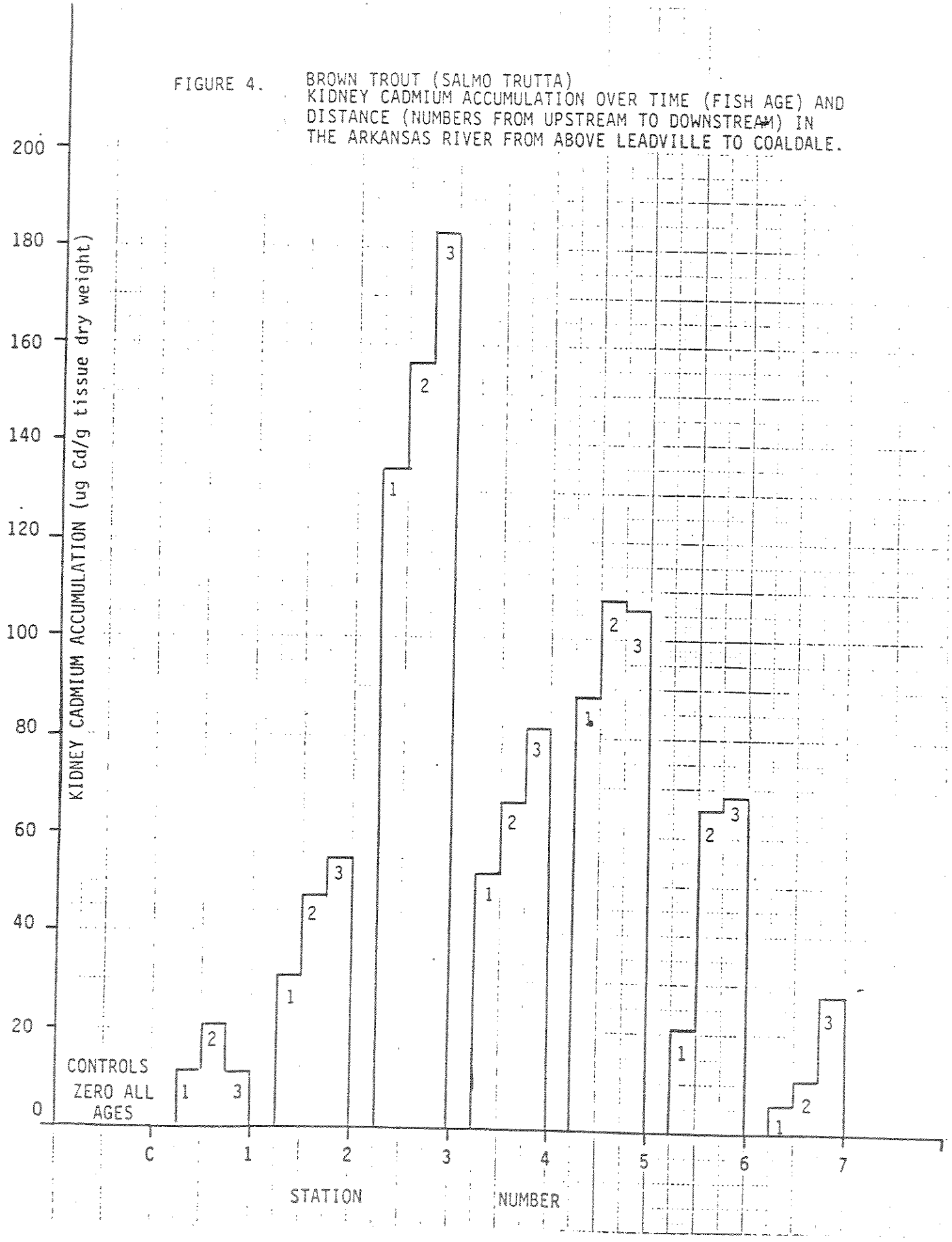


FIGURE 4. BROWN TROUT (SALMO TRUTTA) KIDNEY CADMIUM ACCUMULATION OVER TIME (FISH AGE) AND DISTANCE (NUMBERS FROM UPSTREAM TO DOWNSTREAM) IN THE ARKANSAS RIVER FROM ABOVE LEADVILLE TO COALDALE.



REFERENCES AND LITERATURE CITED

Anderson, R.M., and R.B. Nehring. 1974. Stream fisheries investigations. Colo. Div. Wildl. Job Prog. Rep. Fed. Aid Proj. F-51-R-10. 171 p.

Benoit, D.A., E.N. Leonard, G.M. Christensen, and J.T. Fiandt. 1976. Toxic effects of cadmium on three generations of brook trout (Salvelinus fontinalis). Trans. Amer. Fish. Soc. 105(4):550-560.

Cearley, J.E. and R.L. Coleman. 1974. Cadmium toxicity and bioconcentration in largemouth bass and bluegill. Bull. Env. Cont. & Toxic. 11(2):146-151.

Eaton, J.G. 1974. Chronic cadmium toxicity to the bluegill (Lepomis macrochirus Rafinesque). Trans. Amer. Fish. Soc. 103(4):729-735.

Goettl, J.P., J.R. Sinley, and P.H. Davies. 1974. Water pollution studies. Colo. Div. Wildl. Job. Prog. Rep. Fed. Aid Proj. F-33-R-11. 58 p.

_____, _____, and _____. 1973. Water pollution studies. Colo. Div. Wildl. Job Prog. Rep. Fed. Aid Proj. F-33-R-8. 123 p.

_____ and P.H. Davies. 1978. Water pollution studies. Colo. Div. Wildl. Job Prog. Rep. Fed. Aid Proj. F-33-R-13. 45 p.

_____, and _____. 1976. Water pollution studies. Colo. Div. Wildl. Job Prog. Rep. Fed. Aid Proj. F-33-R-11. 58 p.

Hiltebran, R.C. 1971. Effects of cadmium, zinc, mangenese, and calcium on oxygen and phosphate metabolism of bluegill liver mitrochondria. J. Water Poll. Cont. Fed. 43(5):818-823.

Jackim, E., J.M. Hamlin, and S. Sonis. 1970. Effects of metal poisoning on five liver enzymes in the killifish (Fundulus heteroclitus). J. Fish. Res. Bd. Can. 27:383-390.

LaBounty, J.F., J.J. Sartoris, L.D. Klein, E.F. Monk, and H.A. Salman. 1975. Assessment of heavy metals pollution in the upper Arkansas River of Colorado. Bur. Rec. Rep. No. REC-ERC-75-5. 120 p.

McKim, J.M. and D.A. Benoit. 1974. Duration of toxicity tests for establishing "no effects" concentrations for copper with brook trout (Salvelinus fontinalis). J. Wildl. Man. 31(4):449-452.

Mount, D.I. and C.E. Stephan. 1967. A method of detecting cadmium poisoning in fish. J. Wildl. Man. 31(1):168-172.

Nehring, R.B. and R. Anderson. 1985. Stream fisheries investigations. Colo. Div. Wildl. Job. Prog. Rep. Fed. Aid Proj. F-51-R-8. 171 p.

_____, _____, and D. Winters. 1983. Stream fisheries investigations. Colo. Div. Wildl. Job Prog. Rep. Fed. Aid Proj. F-51-R-8. 188 p.

_____ and _____. 1982. Stream fisheries investigations. Colo. Div. Wildl. Job. Prog. Rep. Fed. Aid Proj. F-51-R-7. 185 p.

_____ and _____. 1981. Stream fisheries investigation. Colo. Div. Wildl. Job Prog. Rep. Fed. Aid Proj. F-51-R-6. 161 p.

_____. 1976. Aquatic insects as biological monitors of heavy metal pollution. Bull. Env. Cont. & Toxic. 15(2):147-154.

_____ and J.P. Goettl. 1974. Acute toxicity of a zinc-polluted stream to four species of salmonids. Bull. Env. Cont. & Toxic. 12(4):464-469.

_____. 1973. Heavy metal toxicity in two species of aquatic insects. M.S. Thesis, Colo. State Univ., Fort Collins.

Nesler, T.P. 1982. Fish populations and fishery of the upper Arkansas River, 1977-80. Fryingpan-Arkansas Fish Research Investigation. Colo. Div. Wildl. Final Rep. 42 p.

Pickering, Q.H. and M.H. Gast. 1972. Acute and chronic toxicity of cadmium to the fathead minnow (Pimephales promelas). J. Fish. Res. Bd. Can. 29(8):1099-1106.

Roline, R.A. and J.R. Boehmke. 1981. Heavy metals pollution of the upper Arkansas River, Colorado, and its effects on the distribution of the aquatic macrofauna. Bur. Rec. Rep. No. REC-ERC-81-15, Denver, Colo. 71 p.

Rowe, D.W. and E.J. Massaro. 1974. Cadmium uptake and time dependent alterations in tissue levels in the white catfish Ictalurus catus (Pisces: Ictaluridae). Bull. Env. Cont. & Toxic. 11(3):244-249.