Fish Population Dynamics and Concentrations of Selected Trace-Elements in Salmonid Tissues and Aquatic Invertebrates, Lower Bryant Creek and East Fork Carson River, Douglas County, Nevada, 2001

U.S. Fish and Wildlife Service
Division of Environmental Quality
Nevada Fish and Wildlife Office
Reno, Nevada

A COORDINATED EFFORT

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Preliminary Assessment of Fish Community Dynamics and Trace-Element Exposures to Aquatic Invertebrates and Salmonids, Lower Bryant Creek and East Fork Carson River, Douglas County, Nevada, 2001

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U.S. FISH AND WILDLIFE SERVICE
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1.0 INTRODUCTION

Leviathan Mine is an inactive mine site contributing trace-element-enriched wastes to the Bryant Creek watershed of California and Nevada. The actual mine property, currently owned by the State of California, Lahontan Regional Water Quality Control Board (LRWQB), covers 656 acres in Alpine County, California, on the eastern flank of the central Sierra Nevada, roughly 6 miles east of Markleeville, California (Figure 1). Metal-enriched wastes are released directly from the mine property into both Leviathan Creek and Aspen Creek. Leviathan Creek joins with Mountaineer Creek approximately 2½ miles downstream of the mine property to form Bryant Creek. Bryant Creek crosses the Nevada state border and empties into the East Fork of the Carson River approximately 7 miles downstream from the confluence of Leviathan and Mountaineer.

Underground mining of copper and sulfur occurred at the Leviathan Mine intermittently from 1863 to 1941, and open pit mining of sulfur occurred from 1951 to 1962. Infiltration of precipitation into and through the open pit and overburden piles created acid mine drainage (AMD), which discharged directly into Leviathan Creek. Contact with waste piles deposited directly into the creek also contaminated Leviathan Creek. Releases of AMD from the site resulted in fish kills in Leviathan and Bryant creeks and the East Fork Carson River (Trelease 1959). The first reported fish kill as a result of Leviathan Mine came in 1952 when a large release of AMD killed fish in Bryant Creek and the East Fork Carson River. A fish survey in Leviathan and Bryant creeks in 1957 failed to detect fish or aquatic organisms (California Regional Water Quality Control Board 1975). In 1959, a fish kill of 10,000 to 20,000 fish was reported along 10 miles of the East Fork Carson River; the fish kill resulted from a failed dike that released approximately 5 million gallons of AMD (California Regional Water Quality Control Board 1975). In 1961, another dike failure released millions of gallons of AMD and caused a fish kill that extended ten miles down the East Fork Carson River (California Regional Water Quality Control Board 1995).

In January 1984, the State of California acquired ownership of the mine property. Jurisdiction over the mine property was transferred to the State Water Resources Control Board. From 1983 (before California's acquisition of the site) to 1985, the LRWQCB built the Leviathan Mine Pollution Abatement Project in an attempt to deal with the AMD generated by past mining operations (Schoen et al. 1995).

In 1998, the Washoe Tribe of Nevada and California, Atlantic Richfield Company, California Department of Fish and Game, Bureau of Indian Affairs, the U.S. Forest Service, and the U.S. Fish and Wildlife Service (Service) initiated cooperative Natural Resource Damage Assessment and Restoration (NRDAR) activities to evaluate injury to natural resources resulting from release of contaminants from the Leviathan Mine. Investigations conducted under NRDAR assessment activities have documented elevated concentrations of several metals and other inorganic contaminants in drainage from the mine (ENSR 1999, Thomas and Lico 2000, Leviathan Mine Natural Resource Trustees 2003). Concentrations of some constituents, including arsenic, cadmium, copper, nickel, and zinc, remained elevated in water in Leviathan, Aspen, and/or Bryant creeks down gradient of the mine site. Similarly, stream sediment collected down gradient of the site contained elevated concentrations of a variety of metals. The occurrence of elevated concentrations of these constituents in aquatic invertebrates collected from Bryant Creek

demonstrated the availability of metals to aquatic organisms (ENSR 1999, Thompson and Welsh 1999, Leviathan Mine Natural Resource Trustees 2003). However, accumulation by aquatic invertebrates in the East Fork Carson River was less apparent. Additionally, the occurrence, extent, and severity of sediment contamination in the East Fork Carson River is largely unknown

With respect to releases of AMD from Leviathan Mine impacting biota in the East Fork Carson River and lower Bryant Creek, the objectives of this investigation were to:

- describe the status of the fish community,
- determine the health and condition of salmonids,
- determine trace-element concentrations in aquatic invertebrate tissues and salmonids, and
- assess the potential adverse effects to fish from trace-element exposure.

Low pH causes system-wide effects in fish communities such as elimination of sensitive species and proliferation of tolerant species; reductions in abundance and diversity of aquatic systems; increasing degrees of abnormal behavior by affected organisms; and reproductive, emergence, and rearing failure (Short et al. 1990, Nelson et al. 1991, Rutherford and Mellow 1994, Saiki et al. 1995). Fish communities have long been viewed as potentially useful bioindicators of stream water quality (Fausch et al. 1990). For example, the Index of Biotic Integrity (IBI; Karr, 1991), a multimetric approach, evaluates habitat impairment at a site by comparing elements of fish species composition, abundance and physical condition to expectations based on ‘‘reference’’ fish communities at unimpacted sites in the region. Developed initially for small, midwestern warm-water streams, the IBI has now been modified for use in other regions (Hughes and Gammon 1987, Miller et al.1988, Steedman 1988, Hughes et al. 2006), and coldwater streams (Leonard and Orth 1986, Lyons et al. 1996). Observations of presence/absence of fish species in this study and comparison of IBI scores are intended to determine the degree to which discharges from Leviathan Mine may be impacting fish communities in the East Fork Carson River.

Environmental stress can affect growth rate and general condition of fish. Condition factors, such as Fulton’s condition factor, provide a relative measure of nutritional state or “well being” of individual fish and populations (Anderson and Gutreuter 1983). Such factors may also be used to compare relative condition of populations and to monitor environmental change over time (Ney 1993). Additionally, degraded environmental conditions and a variety of environmental contaminants have been associated with effects to fish health. Such effects may include increased susceptibility to disease, increased parasitism, and teratogenic deformities (Tuttle et al. 2003). Determinations of health and condition of salmonids by this study is intended to assess the extent of stress salmonids may be exposed to in AMD-influenced drainage of the East Fork Carson River near Leviathan Mine.

Aquatic insects accumulate metals and trace-elements from water, detritus, food, and sediment pathways, and contributions to their whole-body concentrations include tissue incorporations, gut contents, and external adsorptions. Whole-body concentrations serve as indicators of exposure, not direct determinations of the dose of a metal or trace-element to which an aquatic invertebrate has been exposed (Luoma and Carter 1991).

Whole body concentrations determined in this study are intended to indicate whether biota are accumulating excessive amounts of metals and trace-elements discharged from Leviathan Mine, and identify which constituents may be bioaccumulating in fish tissue.

If appreciable accumulations of metals and trace-elements occur in aquatic insects, potentially hazardous exposure of contaminants may occur to insectivorous fish through the dietary pathway. Invertebrate diets of fish contaminated by metals and trace-elements have been associated with adverse effects in trout such as reduced growth and increased mortality, as well as histopathological and physiological effects (Woodward et al. 1994, 1995, Farag et al. 1994, Elderkin 1997). Aquatic insects are important foods of the salmonid fishes that currently occupy or historically occupied the streams of the eastern Sierra Nevada (Moyle 2002). Historical range of the Lahontan cutthroat trout (*Oncorhynchus clarki henshawi*), a species listed as threatened under the Endangered Species Act, includes the surface waters near the Leviathan Mine (U.S. Fish and Wildlife Service 1995). Concentrations of trace-elements in aquatic insects can be evaluated to indicate whether fish occupying these habitats are likely to be harmed by dietary exposure.

The trace-element concentrations in fish tissues indicate whether fish have been exposed to or are accumulating contaminants from water, sediment, or dietary pathways. The gill is the primary site of ionoregulation, acid–base balance, nitrogenous waste excretion and gas exchange; these processes, in particular ionoregulation, are especially sensitive to metal toxicity (Wood 1992). Effects at the level of the gills may take place before the metals are distributed to other parts of the body. Due to the detoxification role of the liver, this organ is often a major site of accumulation of metals and trace-elements in fish (Sorenson 1991). Skeletal muscle tissue concentrations of certain metals are useful in human health and ecological risk assessments, as these tissues comprise much of the fish biomass that is consumed. Determinations of whole body concentrations of trace-elements are important for assessing whole body burdens and accumulations relative to uptake by other tissues. In addition, whole body concentrations can assist in determining potential dietary risks to Native Americans that culturally utilize the entire carcass as opposed to only the fillet.

2.0 METHODS

2.1 FIELD METHODS

Field work was conducted during 15-19 October 2001 at three sites within a 15-km reach of the East Fork Carson River and one site within Bryant Creek (Table 1). Within the East Fork Carson River, a reference site was located upstream from the mouth of Bryant Creek near the California State line, whereas two sites were located downstream from the mouth of Bryant Creek. The lowermost site was situated just above Ruhenstroth Dam near Gardnerville, Nevada. Locations of sites were similar to those sampled by Lehr (2000), Thomas and Lico (2000), Thompson and Welsh (1999, 2000), and Markin and Yee (2006).

Table 1. Locations of three sampling sites on the East Fork Carson River and one sampling site on Bryant Creek.

SITE[a]	STREAM NAME	LOCATION	COORDINATES (D=DOWNSTREAM; U= UPSTREAM)		SITE LENGTH (METERS)
EFC1	East Fork Carson River	~250 meters upstream of Ruhenstroth Dam (located downstream from EFC2)	D	38º 52' 20.81" 119º 41' 32.32"	378
			U	38º 52' 10.40" 119º 41' 32.60"	
EFC2	East Fork Carson River	~1,850 meters downstream from the confluence with Bryant Creek	D	38º 48' 51.37" 119º 41' 28.93"	686
			U	38º 48' 48.27" 119º 41' 46.86"	
EFC3	East Fork Carson River	~520 meters upstream from the confluence with Bryant Creek	D	38º 47' 59.42" 119º 41' 40.56"	584
			U	38º 47' 40.77" 119º 41' 37.22"	
BRY	Bryant Creek	~170 meters upstream from the mouth (confluence with East Fork Carson River)	D	38º 48' 13.05" 119º 41' 46.34"	418
			U	38º 48' 13.90" 119º 41' 29.85"	

[a] Site locations are shown in Figure 1.

2.1.1 Fish Community and Health Assessment

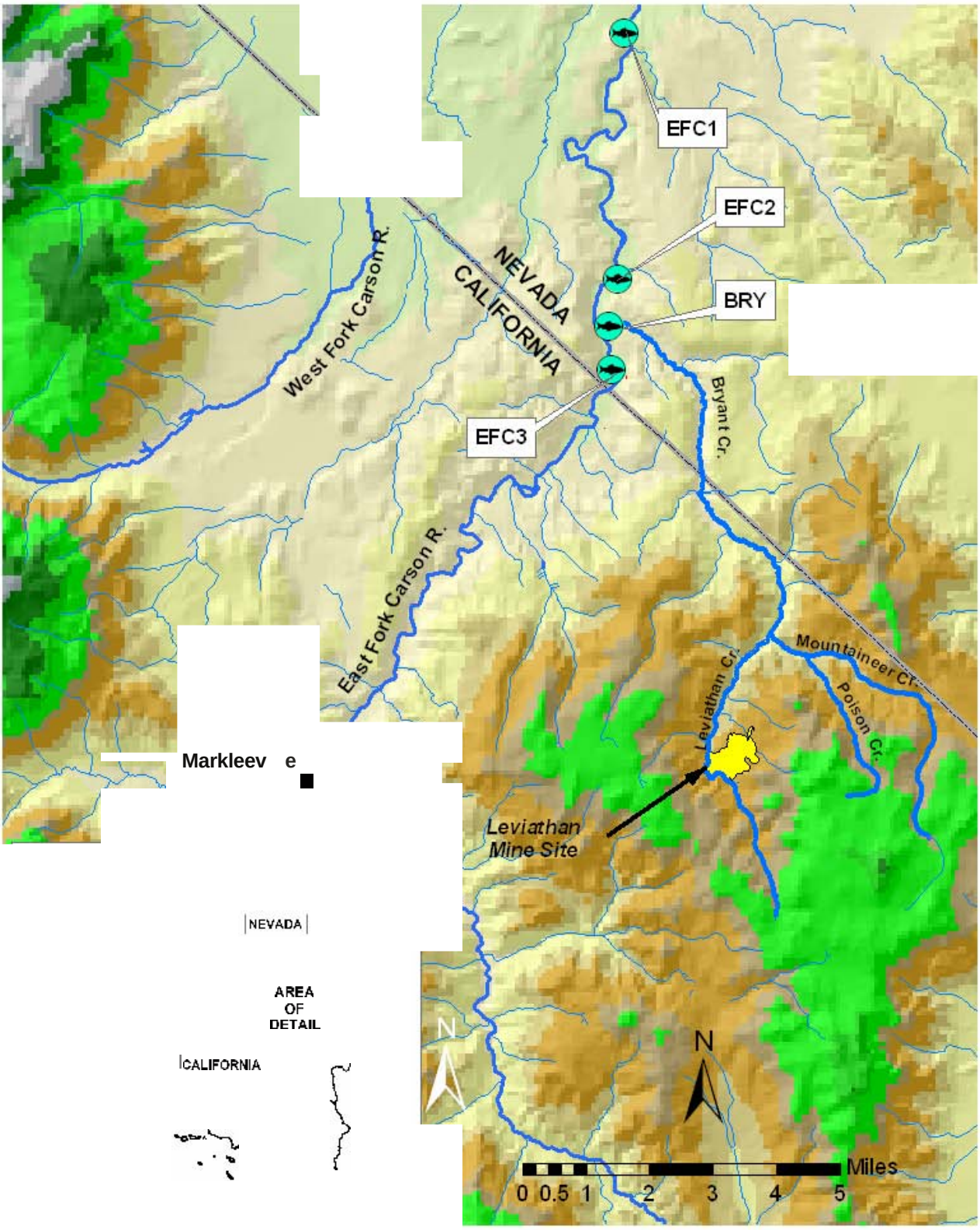
A combination of electrofishing and snorkel observation was used to assess fish species assemblages at each sampling site (Table 1). Each site included at least two sequences of three primary habitat types (e.g., runs, riffles, and pools) as prescribed by Meador et al. (1993) for non-wadeable streams. Downstream boundaries of each sampling reach ended with a pool that allowed observers to assess escapement or recruitment of fish into the study area. Fluorescent orange painted re-bar stakes were driven into the ground on the right bank (as viewed downstream) at the upstream and downstream boundaries of the sample site and locations were recorded with a Global Positioning System (GPS) Unit.

Snorkel surveys followed methods described by Thurow (1994) for one upstream pass followed by a downstream float. Five snorkelers simultaneously conducted an upstream pass within relatively equal corridors. Snorkelers counted all fish in one direction between themselves and the adjacent snorkeler. Snorkelers nearest the shore also counted all fish between themselves and the nearest bank. Fish species and size class were counted by each observer and recorded on a polyvinylchloride (PVC) cuff to be later transferred to a data sheet. After completing counts within a habitat unit (pool, run, or riffle), snorkelers conveyed the information to observers on shore. A second snorkeling pass floating downstream through the survey reach was conducted to offset potential bias in observations resulting from fish avoiding snorkelers moving upstream. PVC pipes, one-inch in diameter, were used to maintain a straight counting line among groups of two or three snorkelers floating downstream.

Within one to three days after snorkeling surveys, sample reaches were electro-fished. Methods presented in Kolz et al. (1995) were followed during electrofishing. The downstream most reach was sampled first, followed by the next reach upstream. Representative reaches of stream were isolated with block nets and systematically electroshocked with a pulsed DC electrical current to collect fish. Sample sites in the East Fork Carson River utilized a two-pass survey using a cataraft towed barge electrofishing unit in the middle portion of the stream and two backpack electrofisher units deployed along the left and right stream bank areas. Due to differences in stream morphology, a two-pass survey utilizing one backpack electro-fisher unit was conducted at the Bryant Creek site. The barge was composed of a pair of Northwest River Supply, 12-foot long by 22-inch diameter inflatable "Surf Cat" cataraft tubes with a 66-inch wide by 50-inch long aluminum pipe rowers frame. A Smith-Root Generator-Powered Pulsator (GPP) electrofisher unit was mounted on a plywood platform secured to the cataraft rower's frame. Electrical output was provided by a five horsepower, 2.5 kilowatt gasoline Honda generator. The electrical field on the barge was generated by two anode probes containing 9-inch diameter rings and two cathode cables mounted along the flanks of the cataraft tubes. Operation required one electrofisher operator who pushed the barge upstream, two anode probe operators, and several netters who flanked probe operators. The backpack electro-fisher units were Smith-Root Type VII models and consisted of an operator with an anode probe and two netters flanking the probe to capture stunned fish. Netters collected stunned fish and transported them in 5-gallon buckets to a streamside live car where personnel identified, enumerated, and processed fish for tissue samples.

To assess fish health and general condition, up to 50 randomly selected fish of each salmonid species were measured, weighed, and assessed for indicators of disease, parasites, and external anomalies from each sample site. Fish were weighed with an Acculab scale (Model VI-1200; 0.1g accuracy). Examinations of external condition of fish were adapted from procedures provided in Meyer and Barclay (1990), Foott (1990), and Goede and Barton (1987). Fish from each site were also examined for deformities using methods adapted from Lemly (1997). Each individual was examined for: 1) lordosis, 2) scoliosis, 3) kyphosis, 4) missing or deformed fins, 5) missing or deformed gills or gill covers, 6) abnormally shaped heads, 7) missing or deformed eyes, and 8) deformed mouth. All fish, with the exception of salmonids collected for chemical analyses, were released back to the stream from which they were collected.

Figure 1. Map depicting the location of Leviathan Mine Site and stream locations that were sampled October 15-19, 2001.



2.1.2 Trace-Element Sample Collection and Preparation for Analysis

On October 15–19, 2001, stoneflies from the Plecoptera order and salmonid fish (trout) were collected from four stream locations in the East Fork Carson River and lower Bryant Creek (Table 1) to obtain tissue for determination of trace-element concentrations.

2.1.2.1 Aquatic Invertebrates

Insect samples were collected from each site using a modification of methods described in Cuffney et al. (1993). Three insect samples were collected at each site from a downstream to upstream order targeting riffle and run habitats within the stream. At each site, a Wildco Aquatic Kick Net (800 µm mesh; 20 x 46 cm opening) was held perpendicular against the stream bottom while substrates upstream were disturbed to dislodge insects. Insects were carried into the net by the current and placed in a clean stainless steel tray. *Skwala* spp. individuals were selected from other insects in the tray using stainless steel forceps and placed in certified clean 60 ml-jars with teflon-lined closures in the field. Instruments and working surfaces were cleaned with a brush and Citranox® detergent, rinsed with a dilute nitric acid solution, and triple rinsed with de-ionized water prior to use at each sample location. All collecting equipment between each sampling site was also washed with a brush using Citranox® detergent, triple rinsed with de-ionized water, and submerged in site water for at least one minute prior to use. Invertebrate samples were stored on blue ice in the field and transferred to a freezer within 8-hours of collection. Samples remained frozen before and during shipment to the laboratory for analyses.

2.1.2.2 Salmonid Tissues

Samples of gill, liver, and muscle from up to five individual salmonids of appropriate size (150 to 250 mm total length (?)) were collected from each site. Up to five additional fish of appropriate size from each site were collected for analysis of trace-elements in whole body. Instruments and working surfaces used for tissue collection were cleaned with a brush and Citranox® detergent, rinsed with a dilute nitric acid solution, and triple rinsed with deionized water prior to use on each fish.

Gill and liver samples were composites of fish collected at each site. However, due to the limited number of adult-sized salmonids captured at EFC1 and BRY, gill, liver, and muscle samples were analyzed from sites EFC2 and EFC3 only. The muscle sample extracted from each fish consisted of one fillet with the skin attached. Whole body samples were weighed and measured for length and put into labeled Ziploc bags. All tissue samples were removed in the field with pre-cleaned stainless steel instruments and placed in certified clean 60 to 250 ml glass jars with Teflon-lined closures. Instruments and working surfaces were cleaned with a brush and Citranox® detergent, rinsed with a dilute nitric acid solution, and triple rinsed with de-ionized water prior to use at each sample location. Tissue samples and whole bodies were stored on ice in the field and transferred to a freezer upon return to the Service's Nevada Fish and Wildlife Office (NFWO). Samples remained frozen before and during shipment to the laboratory for analyses.

2.2 LABORATORY METHODS

2.2.1 Sample Preparation and Analysis

Laboratory analyses for determination of trace-elements were performed by Trace-element Research Laboratory, Texas A&M Research Foundation, College Station, Texas. Percent moisture was determined for all samples. Samples were then homogenized, wet digested with nitric acid, and converted into acidic digest solutions for analysis. Analyses included determination of mercury by cold vapor atomic adsorption spectroscopy, arsenic by graphite furnace atomic adsorption, selenium by atomic fluorescence spectroscopy, cadmium and lead by inductively coupled plasma-mass spectroscopy, and the remaining elements by inductively coupled plasma optical emission spectroscopy.

2.2.2 Data Quality Assessment

Laboratory analytical quality-assurance and quality-control (QA/QC) procedures were ensured by the Patuxent Analytical Control Facility (PACF) as described in their reference manual (PACF 1990). QA/QC procedures included the use of procedural blanks, duplicate samples, spiked samples, and reference materials. All samples met QA/QC requirements and were within standards with the following exceptions:

- limits of detection for cobalt and titanium in several samples of fish tissues were higher than PACF default but within acceptable limits;
- variability in manganese and titanium concentrations from a fish muscle sample were high at EFC3 and an invertebrate sample at BRY respectively. However this variability had no effect on data interpretation;
- recovery of silicon from a spiked sample on a whole body analysis of brown trout from site EFC3 was high. This could be due to analytical bias or non-homogeneity of the sample; and
- recoveries of iron and nickel from dogfish muscle tissues used for standard reference material were slightly low. However, this did not have an effect on the interpretation of the data.

2.3 DATA ANALYSIS

2.3.1 Fish Community and Health Assessment

Fish assemblages were examined for relative abundance and species presence or absence. To describe and assess fish community structure, abundance, species evenness values, and Index of Biotic Integrity (IBI) scores were determined and compared for sampling sites. Although equal numbers of habitat sequences were included in electrofishing surveys for each site, transect lengths were variable among sites and could bias abundance data. Therefore, densities of fish species for each site were calculated by taking the number of individuals caught in electrofishing surveys and dividing them by the transect length.

Length and weight data were used to calculate Fulton's condition factor for each salmonid fish using methods described in Anderson and Neuman (1996). Median length, weight, and mean condition factors for salmonid species captured at sampling sites were determined along with measurements of central location, dispersion, skewness, and potential outliers. Community structure of fish for each sample site was determined using indices provided in Newman (1995) that describe species heterogeneity (Shannon's index), and species evenness (Pielou's J). Differences in mean Fulton factors and community structure indices were detected among sites using non-parametric Kruskal-Wallis tests ($p \leq 0.05$). Mann-Whitney tests ($p \leq 0.05$) were used to identify specific matrices that were significantly different.

A modification of the IBI metrics developed by Hughes et al. (2006) specifically for the Truckee River, which is another Eastern Sierra river, was utilized in this study. IBI methodologies provide a reproducible method to evaluate and rank relative condition of stream fish communities on a geographic scale and to assess changes over time (Miller et al. 1988; Plafkin et al. 1989). The IBI was first proposed by Karr (1991) to synthesize various aspects of fish assemblage condition into a single, easily understood number. Fish IBIs typically sum standardized scores of multiple metrics representing species richness; general tolerance; relative abundance; anomalies; and trophic, habitat, and reproductive guilds. Each metric had a value continuously from zero to one as recommended by Ganasan and Hughes (1998), Hughes et al. (1998), and Minns et al. (1994). Mountain and Tahoe sucker species (*Catostomus platyrhynchus* and *Catostomus tahoensis*) were combined for analyses since they occupy the same ecological niche and physiology is mostly identical. The 11 metrics and their scoring criteria are provided in Table 2. IBI's were not computed for BRY because habitat and hydrological conditions differed from those at the EFC sites.

Age compositions for salmonid species were determined using length-frequency analysis. Age class of younger fish (<3 years old) can be determined through length-frequency analysis and time of fish emergence from gravels. Rainbow trout (*Oncorhynchus mykiss*) spawn in the spring, whereas brown trout (*Salmo trutta*) and mountain whitefish (*Prosopium williamsoni*) spawn in the fall. Dependent upon water temperature, egg incubation time is between 30 to 90 days (Moyle 2002), therefore when size classes are plotted in a histogram, the smallest fish should be the young of the year (usually <75 mm total length (?)) and each subsequent modal class is a year class. Due to increases in growth of gonadal tissues combined with reductions in growth of somatic tissues, age classes become indistinguishable after the third modal class (Moyle and Cech 1988).

Table 2. Index of Biotic Integrity (IBI) metrics for fish, as modified from Hughes et al. (2006).

METRIC[a]	SCORING CRITERIA	TOTAL METRIC VALUE
1. Number of native fish species	0 to ≥5 species present (0.2 points for each species)	0 to 1
2. Percent of individuals that are mountain whitefish	0 to ≥10% (0.1 points for each percent)	0 to 1
3. Evidence of reproduction by mountain whitefish and/or presence of Paiute sculpin	0.5 added for each fish species	0 to 1
4. Percent of individuals that are Lahontan cuttthroat trout	0 to ≥10% (0.1 points for each percent)	0 to 1
5. Percent of individuals that are sensitive species	0 to ≥20% (0.05 points for each percent)	0 to 1
6. Percent of individuals that are mountain/Tahoe sucker	≥20 to 0% (-0.05 points for each percent)	1 to 0
7. Percent of individuals that are as adults	≥20 to 0% (-0.05 points for each percent)	1 to 0
8. Percent of individuals that are generally tolerant	≥5 to 0% (-0.2 points for each percent)	1 to 0
9. Percent of individuals that are alien to Carson River	≥25 to 0% (-0.04 points for each percent)	1 to 0
10. Percent of individuals that have external anomalies	0 to ≥5% (-0.2 points for each percent)	1 to 0

Note: [a] See Table 3 for scientific names of fish

2.3.2 Trace-Element Occurrence and Relative Concentrations

Due to the limited nature of available habitat for salmonids in this portion of the East Fork Carson River, sample sizes precluded some statistical evaluations for examining relationships among the different matrices. Therefore, subjective comparisons of these data were conducted for some trace-elements. Trace-element concentrations were subjected to Lilliefor's test, which is a modification of the Kolmogorov-Smirnov goodness-of-fit test that is effective for testing the normality assumption of data. Data for most of the trace-elements did not meet assumptions of normality, therefore non-parametric Kruskal-Wallis tests were conducted on trace-element data to determine if differences occurred among sites. Mann-Whitney tests ($p \leq 0.1$) were utilized to identify potential differences in median concentrations of constituents in tissues among sites. Statistical analyses were conducted using SYSTAT version 10 (SYSTAT 2004) where sample sizes and detection limits permitted. Non-detections (below the minimum reporting value) for derivation of geometric means were set to half the detection limit. Analytes that had non-detections in greater than 50 percent of observations within matrices were excluded from additional statistical analyses.

To assess potential dietary accumulation of trace-elements for the study area, mean concentrations of salmonid tissue were compared to invertebrate concentrations from each site to derive bioaccumulation ratios. Trace-element concentrations in invertebrates, rainbow trout liver tissue, and trout whole bodies were compared to adverse effect values determined or estimated from published studies. Gill concentrations were compared to estimated saturation levels determined by McGeer at al. (2000). Muscle concentrations were compared to single value guidelines for select trace-elements developed by U.S. Environmental Protection Agency (USEPA) for edible tissue (USEPA 2000).

3.0 RESULTS AND DISCUSSION

3.1 FISH COMMUNITY AND HEALTH ASSESSMENT

3.1.1 Abundance, Density, and Community Indices

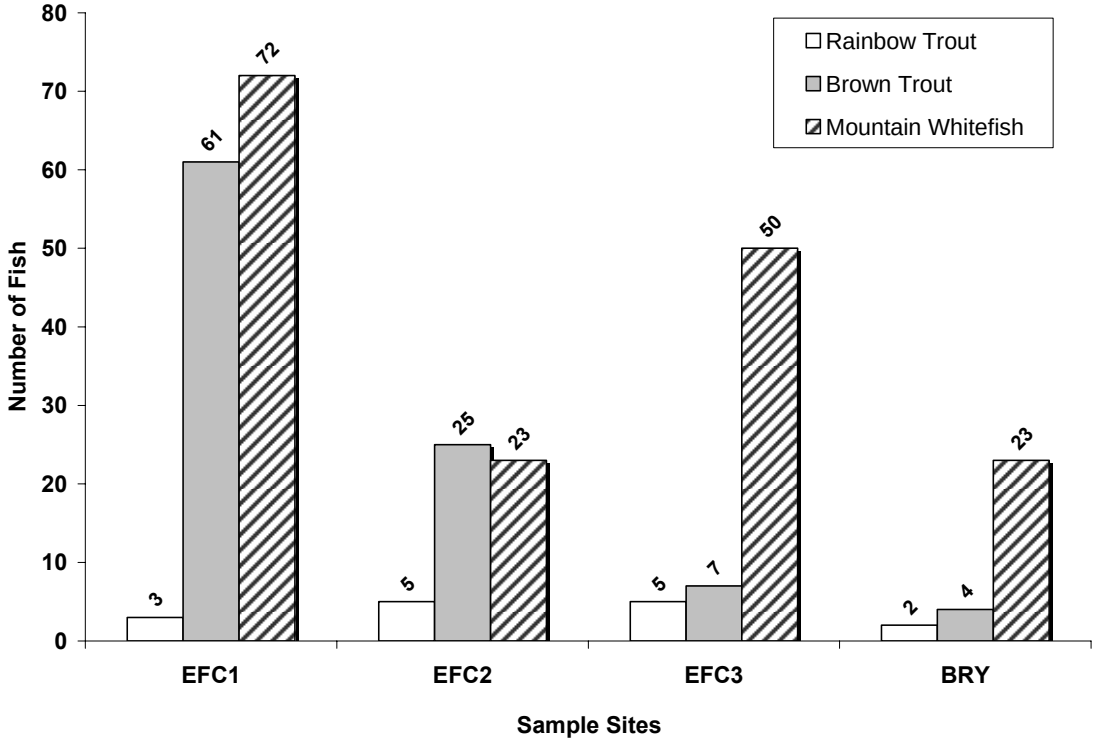
Overall, seven species of fish were identified (Table 3). The number of species at each site ranged from 5 (EFC2) to 7 (EFC1 and EFC3). Species common to all sites were brown trout, mountain/Tahoe sucker, mountain whitefish, rainbow trout, and speckled dace. Paiute sculpin were not captured or observed from BRY and EFC2. Lahontan redside shiners were absent from EFC2. Total abundance for sites ranged from 130 (BRY) to 877 (EFC1). Mountain whitefish was the most abundant salmonid at all sites except at EFC2 where brown trout was similar (Figure 2). Speckled dace was the most abundant non-salmonid fish at EFC1 and BRY. Mountain/Tahoe sucker was most abundant at EFC2 and EFC3 (Figure 3). Abundance of Paiute sculpin was not determined because none were captured by electrofishing surveys at any site, however, they were observed in snorkel surveys at EFC1 and EFC3 (Table 3).

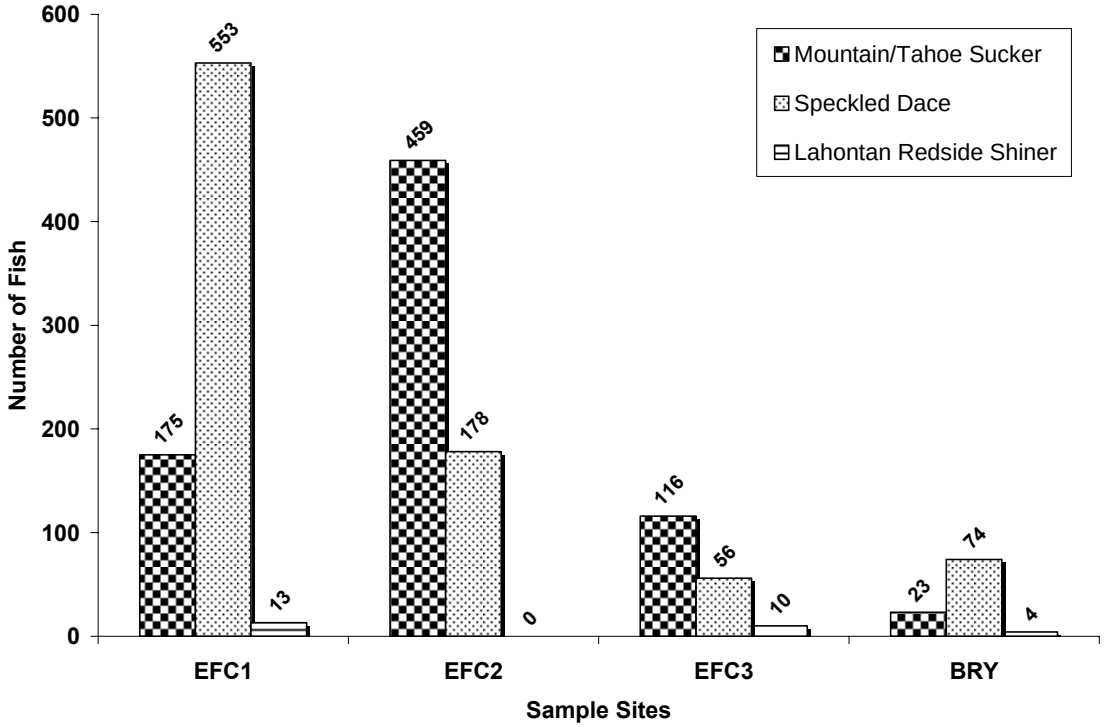
Table 3. Geographic origin, trophic group, and pollution tolerance of fish observed or collected from sample sites in the East Fork Carson River drainage, October 2001 (modified from Chandler et al. 1993 and Barbour et al. 1999).

COMMON NAME	SCIENTIFIC NAME	FAMILY	ORIGIN [a]	TROPHIC GROUP OF ADULT [b]	TOL-ERANCE [c]	SITES OF OCCURRENCE
Mountain/Tahoe sucker	*Catostomus spp.*	Catostomidae	N	H	T	EFC1, EFC2, EFC3, BRY
Paiute sculpin	*Cottus beldingi*	Cottidae	N	I	I	EFC1, EFC3
Rainbow trout	*Oncorhynchus mykiss*	Salmonidae	I	P	M	EFC1, EFC2, EFC3, BRY
Mountain whitefish	*Prosopium williamsoni*	Salmonidae	N	I	M	EFC1, EFC2, EFC3, BRY
Lahontan redside shiner	*Richardsonius egregius*	Cyprinidae	N	I	M	EFC1, EFC3, BRY
Lahontan speckled dace	*Rhinichthys osculus robustus*	Cyprinidae	N	O	M	EFC1, EFC2, EFC3, BRY
Brown trout	*Salmo trutta*	Salmonidae	I	P	T	EFC1, EFC2, EFC3, BRY

Note: [a] N= native; I= introduced; [b] H= herbivore; O= omnivore; I= insectivore; P= piscivore; [c] I= intolerant; M= intermediate; T= tolerant

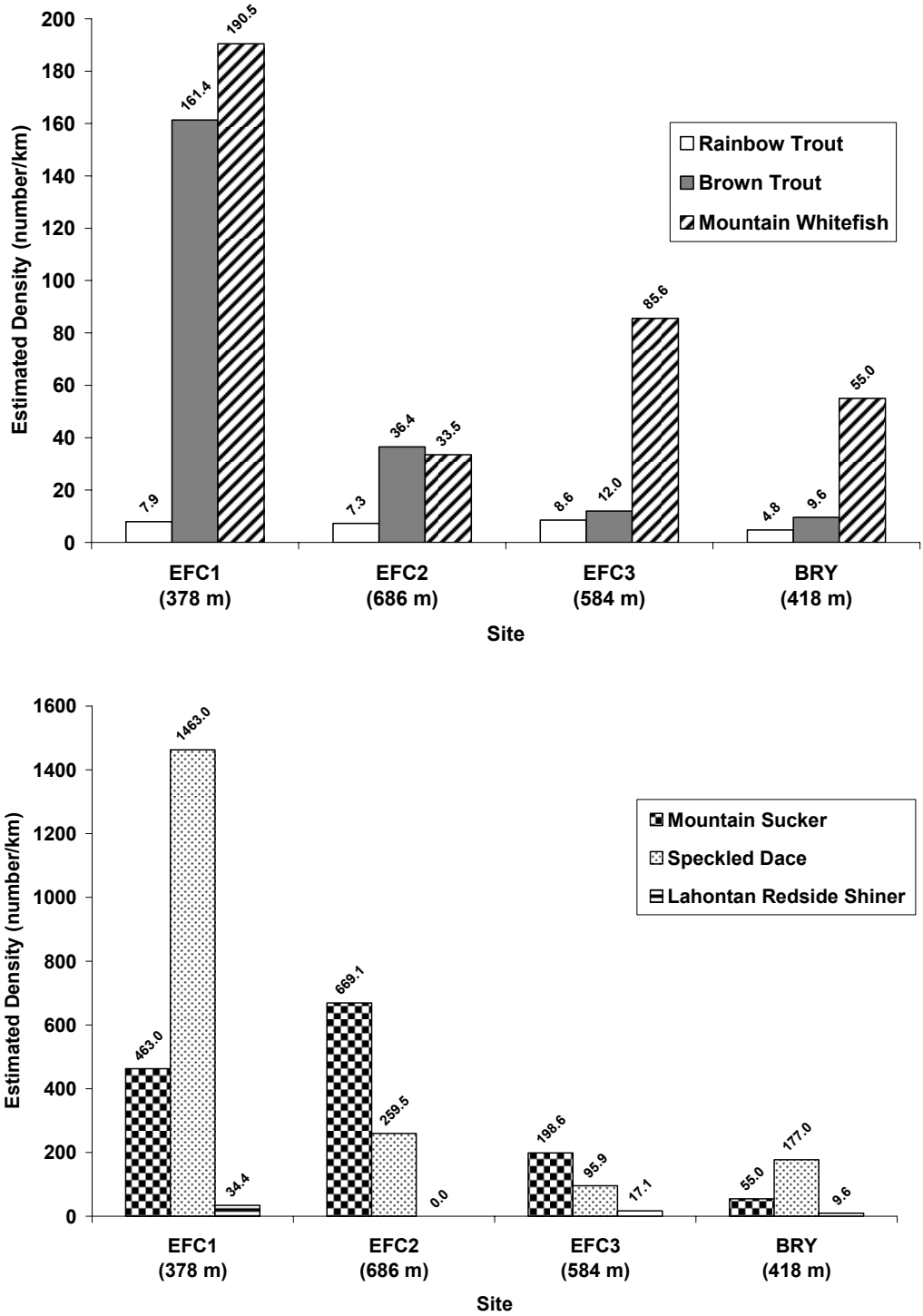
Figure 2. Abundance of salmonid fish species collected from electrofishing surveys conducted at sample sites on the East Fork Carson River and lower Bryant Creek, Nevada, October, 2001. See Table 1 and Figure 1 for locations.





Despite potential bias of abundance data due to varying transect lengths, density of fish species had similar characteristics to abundance data (Figure 3).

Figure 3. Density of salmonid and non-salmonid fish collected from electrofishing surveys conducted at sample sites on the East Fork Carson River and lower Bryant Creek, Nevada, October, 2001. See Table 1 and Figure 1 for locations.



Fish community indices that describe species heterogeneity and evenness for each sample site are presented in Figure 4. Shannon's index describing heterogeneity ranged from 0.89 (EFC2) to 1.33 (EFC3). Pielou's index for species evenness ranged from 0.55 (EFC2) to 0.74 (EFC3). However, heterogeneity and evenness values were not significantly different suggesting no differences in species diversity between sites.

Interpretation of abundance in salmonid and non-salmonid species in stream habitats throughout the Great Basin is difficult due to high variability in hydrologic conditions from year to year and naturally low diversities of fish assemblages. The fewest number of species observed in this study (i.e., 5) occurred just below the Bryant Creek confluence (EFC2). A maximum of seven species were observed at EFC1 and EFC3. At EFC2, there was an absence of the intermediately tolerant Lahontan redside shiner and intolerant Paiute sculpin and a marked increase of tolerant mountain sucker abundance. These two species are fairly localized and do not migrate to the extent of salmonid species and represent localized conditions. The absence of the intermediate to intolerant species combined with marked increase of tolerant species at EFC2 is likely the result of influences on water or habitat quality in East Fork Carson River from Bryant Creek. Although the heterogeneity and evenness is of fish communities were not significantly different among sites, the higher values generally observed in the East Fork Carson River above Bryant Creek suggest impairment from Bryant Creek and below its confluence with the East Fork Carson River. These observations are consistent from other studies that assessed impacts from AMD-influenced drainages (Platts et al. 1979, Farag et al. 2003).

While interpretation of fish species abundance is complicated, comparison of abundance among sites reflects the quality of aquatic habitat at a given point in time. For most species, abundance decreased from EFC1 upstream to EFC3, with the exception of mountain whitefish which was markedly greater at EFC3 upstream of the confluence with Bryant Creek. The greater abundance suggests better habitat and/or food quality for the species.

IBI scores for each sampling site consistently declined in a downstream fashion and ranged from a value of 7.60 at EFC3 to 6.13 and 6.75 at EFC2 and EFC1, respectively (Table 4). Because IBI data from minimally-disturbed or natural reference sites are not available, quantitative estimates of reference conditions for the East Fork Carson River River are lacking. However, following interpretations of median IBI scores provided by Bozzetti and Schulz (2004), Hughes (1994), and Hughes et al. (1998), interpretation of the IBI scores for the EFC1 and EFC2 sampling sites are considered marginally impaired compared to EFC3 located upstream of the Bryant Creek confluence which is considered acceptable. The reductions in IBI scores for down-gradient sites could be the result of cumulative effects of AMD-influenced drainage from Bryant Creek or elevation-influenced differences between some locations.

Figure 4. Species diversity (Shannon's index) and evenness (Pielou's J) indices calculated from electro-fishing survey data, East Fork Carson River drainage, Nevada, October, 2001. See Table 1 and Figure 1 for locations.

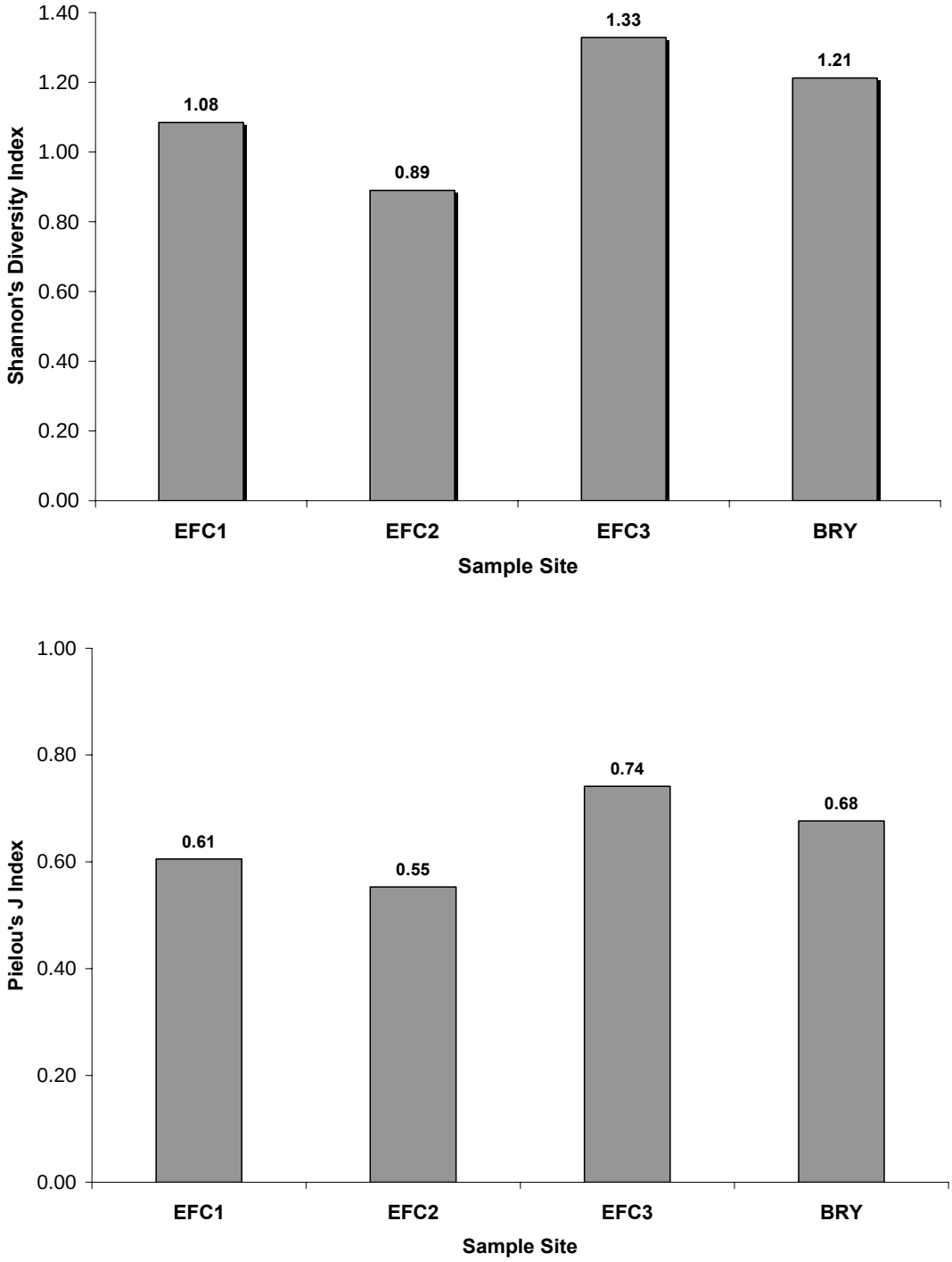


Table 4. Index of Biotic Integrity scores for fish communities at sample sites on the East Fork Carson River, Nevada, October 2001 (adapted from Hughes et al. 2006).

METRIC	SITE					
	EFC1		EFC2		EFC3	
	METRIC VALUE	SCORE	METRIC VALUE	SCORE	METRIC VALUE	SCORE
1. Number of native fish species	4.00	0.80	3.00	0.60	4.00	0.80
2. Percent of individuals that are mountain whitefish	8.21	0.82	3.46	1.00	21.10	2.11
3. Evidence of reproduction by mountain whitefish and/or presence of Paiute sculpin	yes/yes	1.00	yes/no	0.50	yes/yes	0.50
4. Percent of individuals that are Lahontan cuttthroat trout	0.00	0.00	0.00	0.00	0.00	0.00
5. Percent of individuals that are sensitive species	8.55	0.43	4.21	0.21	23.21	1.00
6. Percent of individuals that are mountain/Tahoe sucker	19.95	1.00	69.02	1.00	48.95	1.00
7. Percent of individuals that are omnivorous as adults	83.01	1.00	95.79	1.00	72.57	1.00
8. Percent of individuals that are generally tolerant	19.95	0.00	69.02	0.00	48.95	0.00
9. Percent of individuals that are alien to Carson River	7.30	0.71	4.51	0.82	5.06	0.80
10. Percent of individuals that have external anomalies	0.005	1.00	0.003	1.00	0.00	1.00
TOTAL	**6.75**		**6.13**		**7.60**	

3.1.2 Fish Health and Condition

A summary of external anomalies observed in fish captured at sample sites is presented in Table 5. Only two types of external anomalies were observed among salmonids: eroded or missing fins, and one observation of a tumor growth. No external anomalies were observed in any salmonids at EFC3 and BRY. No external anomalies were observed in mountain whitefish at any site. Salmonids that were selected for extraction of tissues for trace-element analyses were observed internally for any abnormalities such as lesions or parasitic infestations, however, no internal abnormalities were observed.

Table 5. Summary of external anomalies observed in salmonids captured from sampling sites on the East Fork Carson River and lower Bryant Creek, Nevada, October 2001.

SITE	FISH SPECIES	ANOMALY	# OF OBSERVATIONS	N
EFC1	rainbow trout	Missing pectoral fin	1	136
		Eroded or clubbed caudal fin	1	
	brown trout	Eroded or clubbed caudal fin	1	
		Tumor on mouth	1	
EFC2	brown trout	Eroded or clubbed caudal fin	2	53
EFC3	-	None observed	0	62
BRY	-	None observed	0	29

Median length, weight, and mean condition factors for salmonid species captured at sampling sites are presented in Table 6 and measurements of central location, dispersion, skewness, and potential outliers appear in Figure 5. Mean Fulton's condition factors for brown and rainbow trout did not differ among sites or species. Fulton condition factors for mountain whitefish were highest at EFC2 when compared to all other sites. Upon further examination, relative weights of mountain whitefish, utilizing a standard weight equation provided by Rogers et al. (1996), revealed the majority of mountain whitefish were below the estimated physiological optimum with the exception of smaller-sized individuals at EFC2 (Figure 6). Relative weights of mountain whitefish are of interest to fishery managers because the species is a sensitive indicator of poor water quality and other salmonid stressors (Bergstedt 1994). These data also show physiological state of individuals decrease as length (or age) increases which may be the result of chronic long-term effects from contaminant exposures. In addition, a potential effect on invertebrate densities and, indirectly, fish condition, could be occurring as a result from AMD from Leviathan, which has been demonstrated to be toxic to invertebrates at some distance downstream from the mine (Thompson and Welsh 2000).

Table 6. Median total length (millimeters) and weight (grams), and mean condition (Fulton's factor) for salmonid species captured at sampling sites in the East Fork Carson River watershed, October 2001.

	BROWN TROUT				RAINBOW TROUT				MOUNTAIN WHITEFISH			
SITE	**EFC1**	**EFC2**	**EFC3**	**BRY**	**EFC1**	**EFC2**	**EFC3**	**BRY**	**EFC1**	**EFC2**	**EFC3**	**BRY**
N	61	25	7	4	3	5	5	2	72	23	50	23
Length Range (mm)	85 – 295	108 –352	179 – 312	105 – 126	212 – 287	238 – 315	268- 420	247 – 249	95 – 180	122 – 305	97 – 265	105 – 166
Median length (mm)	158	152	303	106	267	261	297	248	134.5	165	131	130
Weight Range (g)	10 – 222	10 – 444	55 – 318	10 – 18	84 – 240	159 – 316	208 – 767	138 – 196	6 – 41	19 –244	6 – 180	4 – 35
Median weight (g)	38.7	32.7	260.4	12.25	226.2	179.8	268.7	167.05	17.05	48.8	14.2	14.4
Mean Condition	0.99	1.10	0.98	0.97	1.03	1.07	1.02	1.09	0.73	1.07	0.72	0.62

Figure 5. Fulton's condition factors for salmonids captured at sampling sites within East Fork Carson River drainage, Nevada, October, 2001.

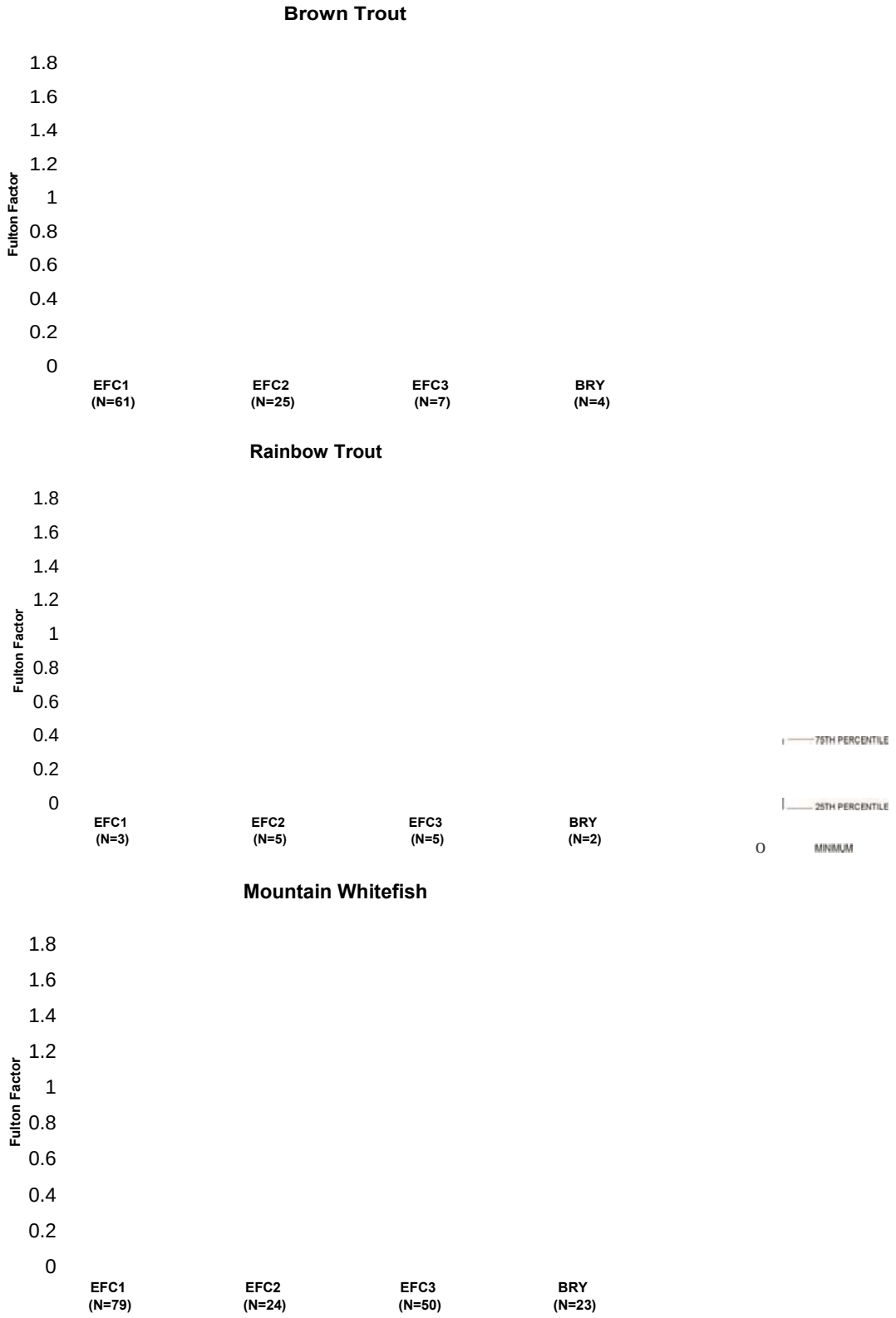
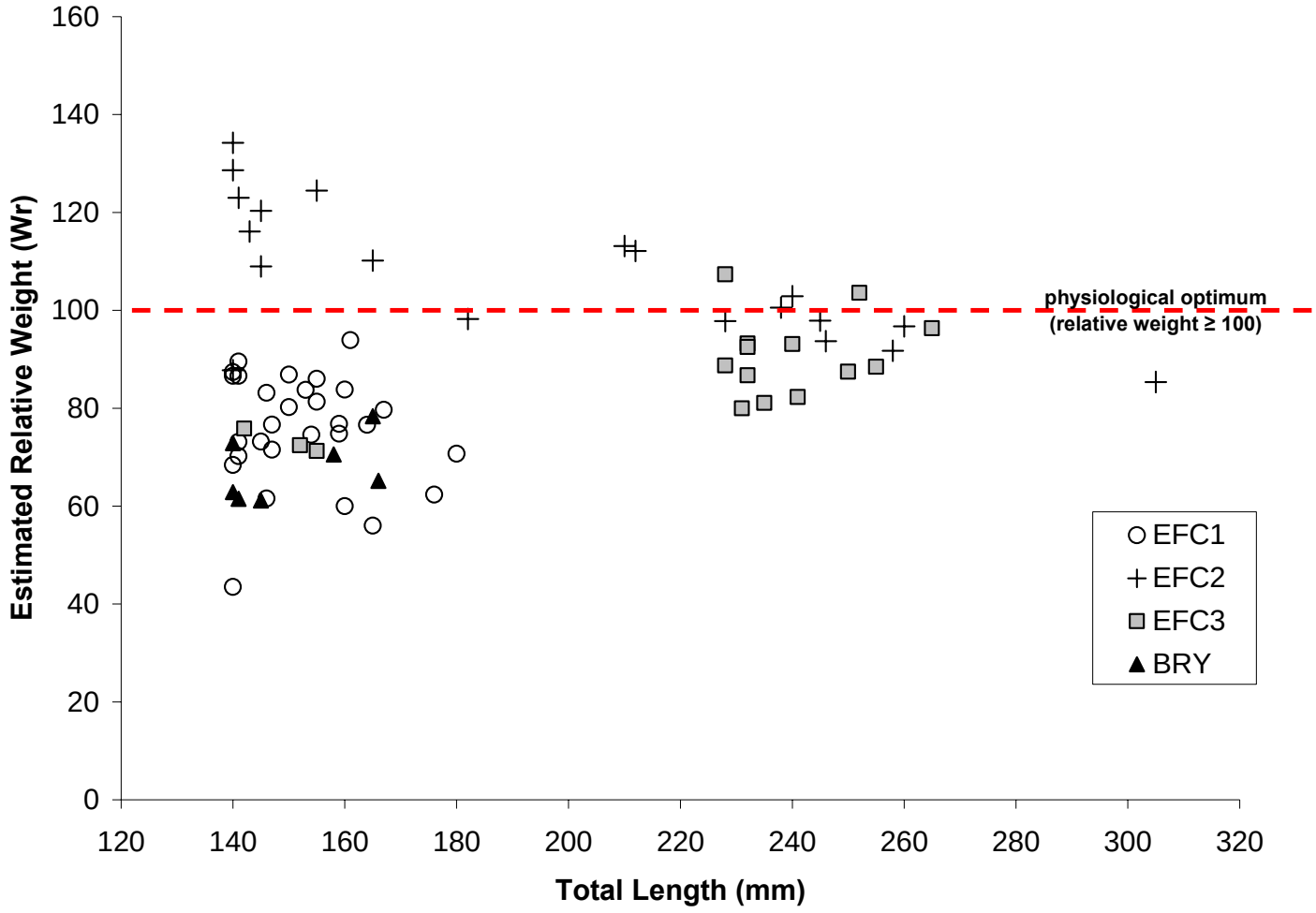


Figure 6. Comparison of physiological optimum to relative weight by length of mountain whitefish (*Prosopium williamsoni*) collected in the East Fork Carson River and lower Bryant Creek, October, 2001.



3.1.3 Length-Frequency Histograms of Salmonids

Length-frequency histograms for brown trout, rainbow trout, and mountain whitefish are presented in Figures 7, 8, and 9, respectively. A robust age-class structure for brown trout occurred at EFC1 with several adult and sub-adult individuals of various sizes that included juveniles that were young from the previous year. EFC2 had at least three age-classes represented, however, numbers in each age-class were reduced compared to EFC1. The numbers of brown trout at EFC3 were reduced compared to the two sites downstream and consisted of only 7 adult individuals of various sizes, half of which were ≥ 300 mm in total length. Brown trout captured at BRY consisted of 4 individuals ranging between 100 and 130 mm in total length, representing only 1 or 2 juvenile age classes. Total number of rainbow trout captured at each site ranged from 2 (BRY) to 5 (EFC2 and EFC3). All rainbow trout captured were adults that ranged from 212 to 420 mm in total length. Mountain whitefish at all sites contained robust age-class structures with several adult and sub-adult individuals of various sizes and juveniles that were young from the previous year. However, the number of individuals at EFC2 and BRY was smaller compared to other sites.

Length-frequency distributions reflect interactions of the rates of reproduction, recruitment, growth, and mortality of age-groups present. These distributions can help our understanding of the dynamics of populations and identify problems with year-class failures or low recruitment, slow growth, or excessive annual mortality (Anderson and Neuman 1996). Comparison of lengths for brown trout at all sites revealed an absence of individuals in the 200 to 260 mm length range (Figure 7). In addition, no juvenile or sub-adult rainbow trout were observed or captured for the reaches sampled indicating a lack or absence of reproduction and/or recruitment (Figure 8). Data for mountain whitefish indicated adequate reproduction and recruitment at all sites but comparison of lengths also revealed a depression or absence of fish between 160 and 200 mm (Figure 9).

The lack of individuals in the various size ranges could be caused by several factors such as low-level fish stocking, high angler pressure, or adverse exposure to contaminants. The California portion of the East Fork Carson River from Hangman's Bridge (near Markleeville) to the California-Nevada State line is a designated Wild Trout fishery by the California Department of Fish and Game (CDFG), has a zero-limit for salmonids, and is not artificially stocked with salmonids (pers. comm., Stafford Lehr, Fisheries Biologist, CDFG, 2005). However, the Nevada Department of Wildlife (NDOW) regularly stocks the East Fork Carson River with brown and rainbow trout on a semi-annual basis. NDOW stocking data from 1995 to 2001 is provided in Table 7. No disease outbreaks were known to have occurred among salmonids in the East Fork Carson River during the same time period and angling pressure on trout was managed effectively through state regulations (pers. comm., Patrick Sollberger, Fisheries Biologist, NDOW, 2005). Therefore, degraded water quality and/or habitat due to contaminants discharged from the Mine are likely contributing to conditions leading to inadequate spawning and rearing habitat or reduced reproductive fitness in individuals. Depauperate numbers of individuals and missing age classes were also observed by Lehr (2000) in Bryant Creek in 1998. These observations suggest adverse impacts to reproduction and recruitment due to elevated contaminant exposures associated with AMD from Leviathan Mine.

Figure 7. Length frequency histograms of brown trout collected in East Fork Carson River and lower Bryant Creek, Nevada, October, 2001.

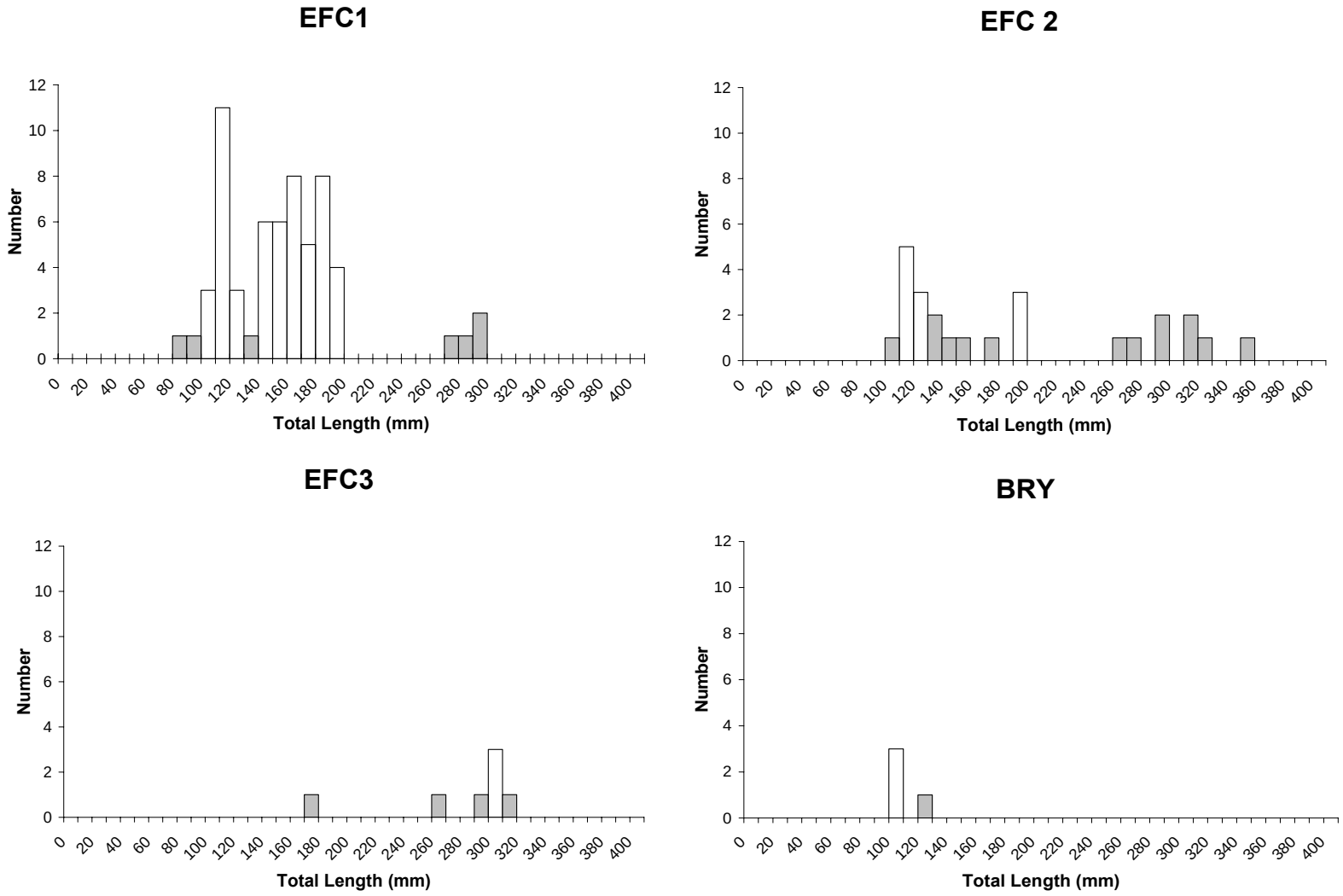


Figure 8. Length frequency histogram s of rainbow trout collected in East Fork Carson River and lower Bryant Creek, Nevada, October, 2001.

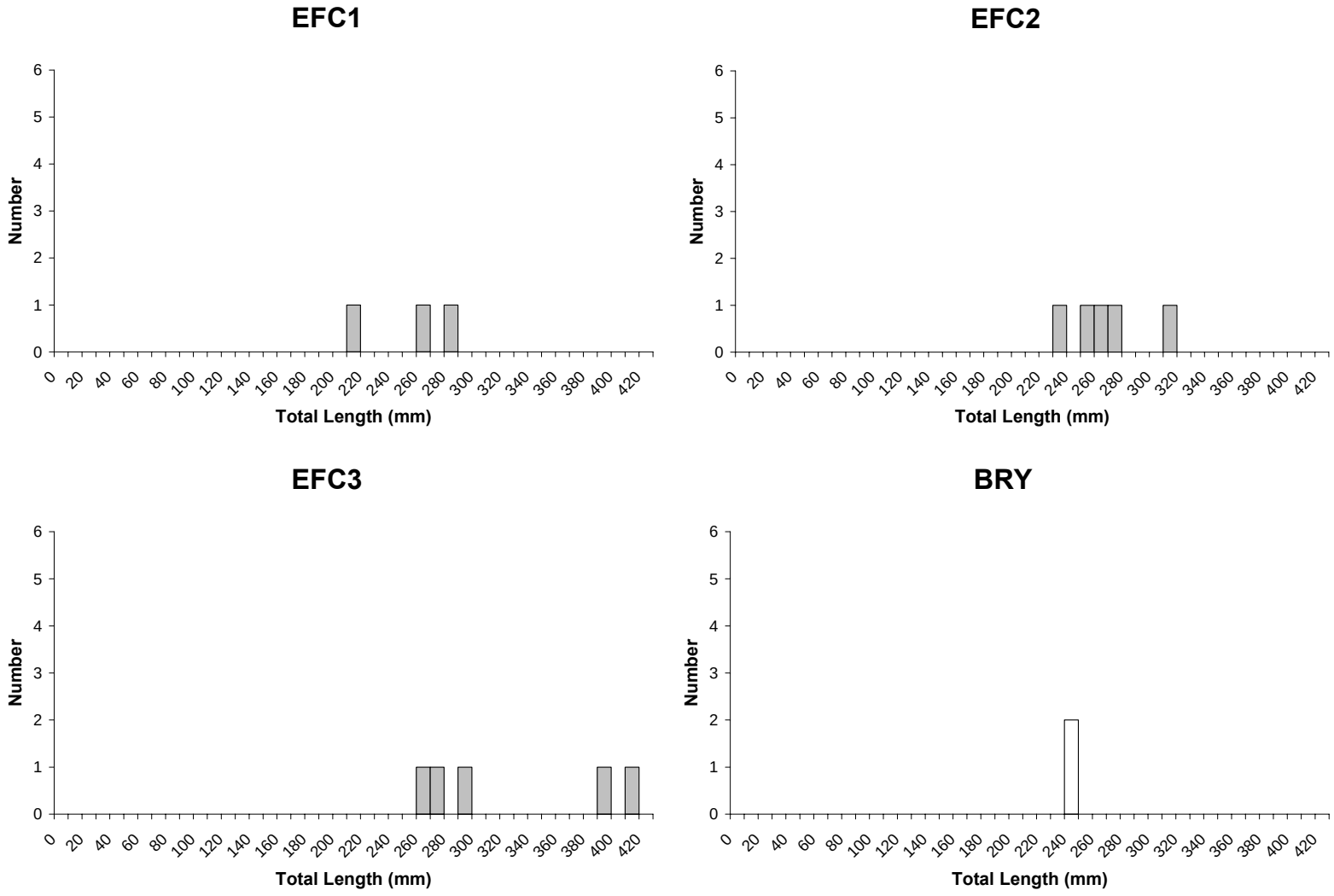


Figure 9. Length frequency histogram s of mountain whitefish collected in East Fork Carson River and lower Bryant Creek, Nevada, October, 2001.

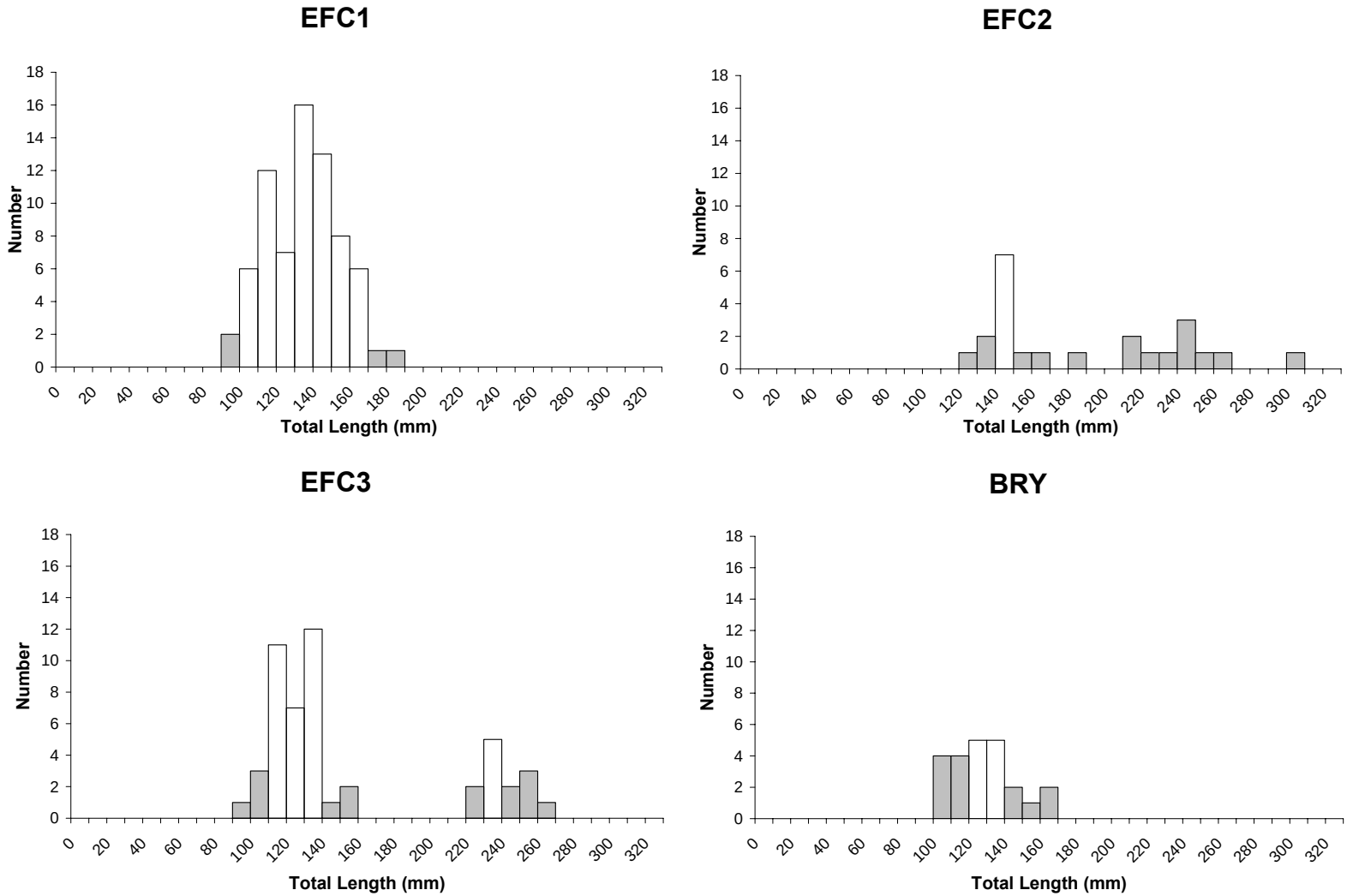


Table 7. Summary of stocking histories for brown and rainbow trout in the East Fork Carson River from Ruhenstroth Dam to the Nevada-California Stateline, 1995-2001 (taken from data provided by Patrick Sollberger, fisheries biologist, Nevada Department of Wildlife).

SPECIES AND SIZE CLASS	YEAR						
	1995	1996	1997	1998	1999	2000	2001
Brown Trout-Fingerling (1/2" to 2" in length)	9,910	15,940	7,300	16,599	9,500	27,421	17,670
Brown Trout-Catchable (≥ 8 inches in length)	1,330	0	575	700	0	0	0
Rainbow Trout-Catchable (≥ 8 inches in length)	4,673	1,828	0	2,860	400	0	750

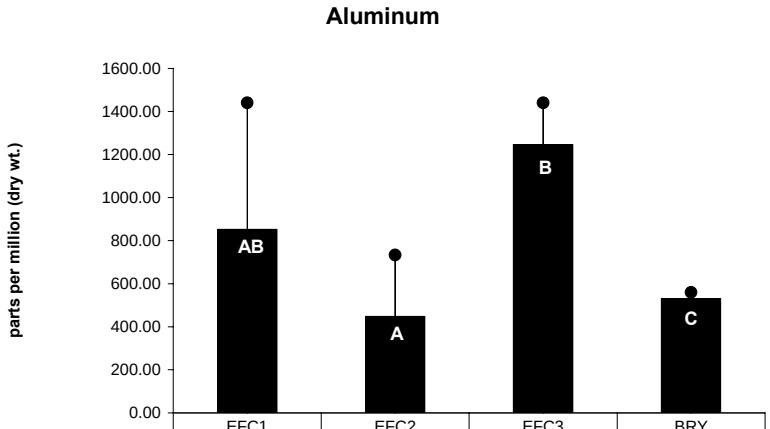
3.2 TRACE-ELEMENT EXPOSURE IN BIOTA AND RELATIONS TO ADVERSE EFFECTS AND POTENTIAL RISKS

Aquatic invertebrates and fish tissue were analyzed for a total of 27 trace elements. Descriptions of the samples collected, including sample number; location collected (from Fig. 1); type of sample collected, type of species collected, weight of sample, and percent moisture of sample, are provided in Appendix 1. A complete list of all samples with results of trace-element concentrations and detection limits for each sample is provided in Appendix 2.

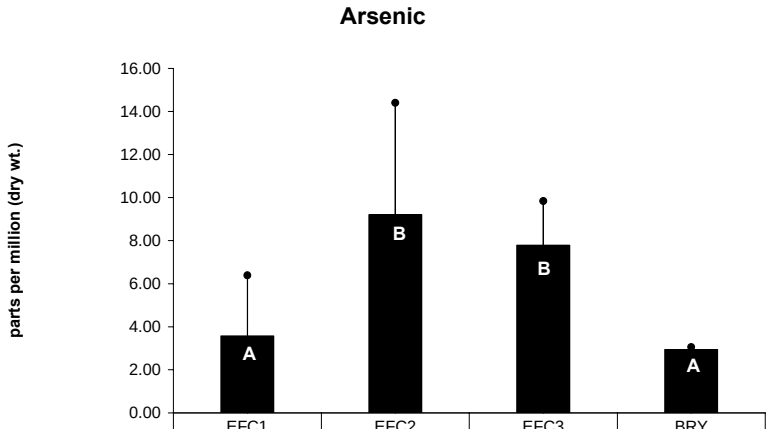
3.2.1 Aquatic Invertebrate Exposure

Concentrations of trace-elements for stonefly larvae samples are provided in Appendix 2. Differences existed among sites for arsenic, calcium, cadmium, cobalt, manganese, mercury, nickel, selenium, titanium, and vanadium. Mean and maximum concentrations of these select trace-elements along with designations of differences between sites are presented in Figure 10.

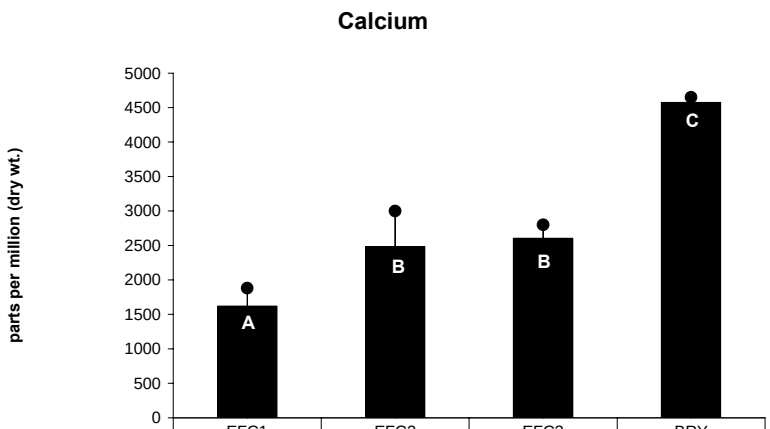
Figure 10. Mean and maximum concentrations along with statistical differences ($p \leq 0.1$) of select metals and trace-elements in stonefly (Plecoptera) larvae collected from sampling sites in East Fork Carson River and lower Bryant Creek, Nevada, October 2001. (letters indicate significant differences among sites)



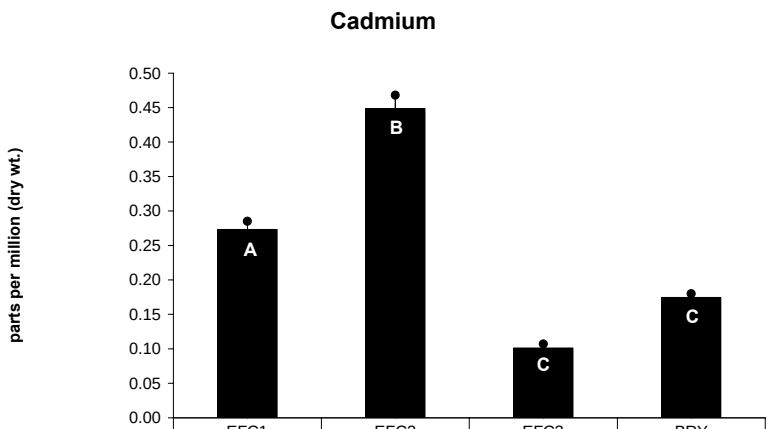
	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	852.66	447.56	1246.24	529.68
● Maximum	1440.00	733.00	1440.00	560.00



	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	3.58	9.20	7.79	2.94
● Maximum	6.39	14.40	9.84	3.06

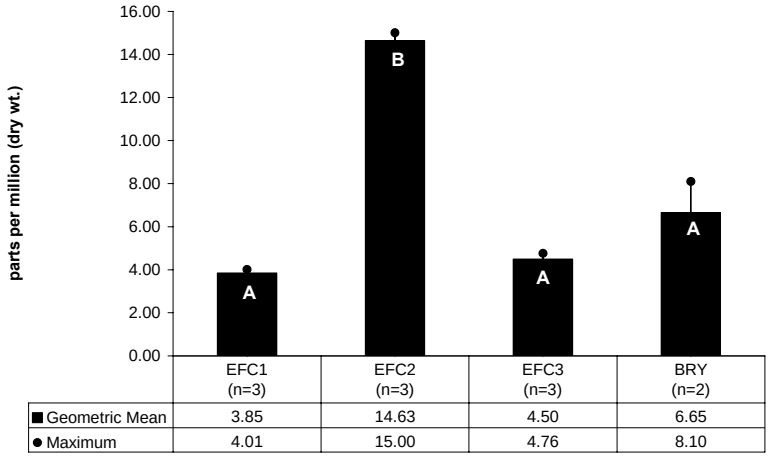


	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	1619.510089	2483.37232	2602.669841	4574.385205
● Maximum	1880	3000	2800	4650

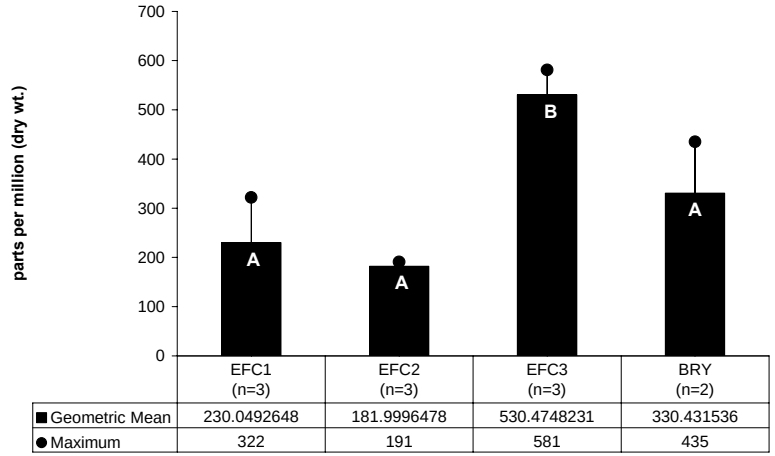


	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	0.27	0.45	0.10	0.17
● Maximum	0.29	0.47	0.11	0.18

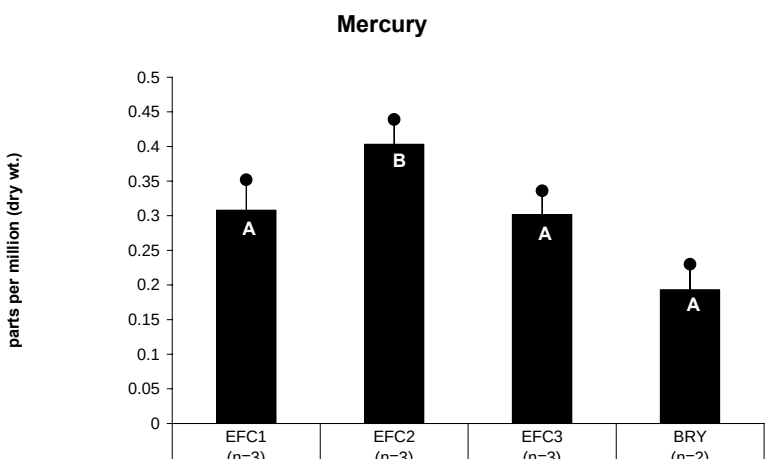
Figure 10 (cont). Mean and maximum concentrations along with statistical differences ($p \leq 0.1$) of select metals and trace-elements in stonefly (Plecoptera) larvae collected from sampling sites in East Fork Carson River and lower Bryant Creek, Nevada, October, 2001. (letters indicate significant differences among sites)



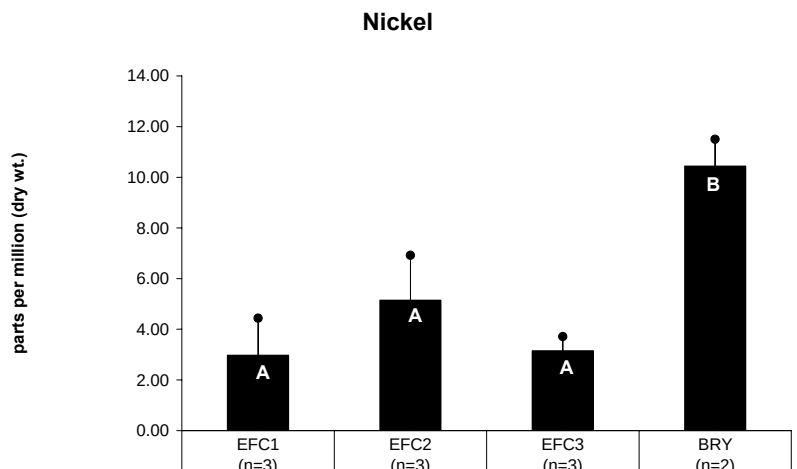
	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	3.85	14.63	4.50	6.65
● Maximum	4.01	15.00	4.76	8.10



	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	230.0492648	181.9996478	530.4748231	330.431536
● Maximum	322	191	581	435

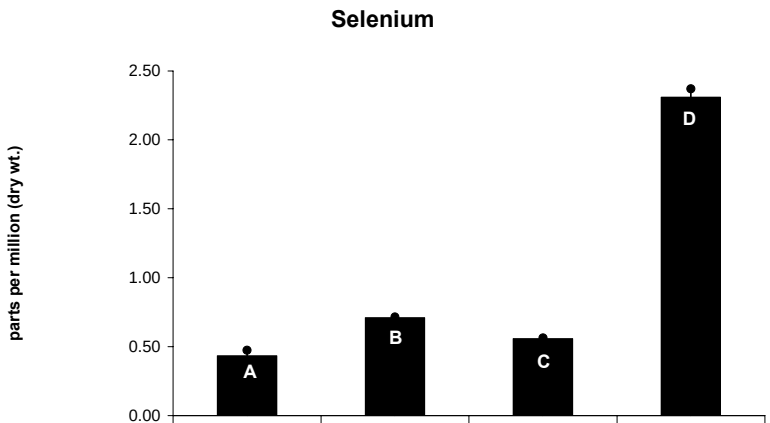


	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	0.307918346	0.403228079	0.301701267	0.193028495
● Maximum	0.35	0.44	0.34	0.23

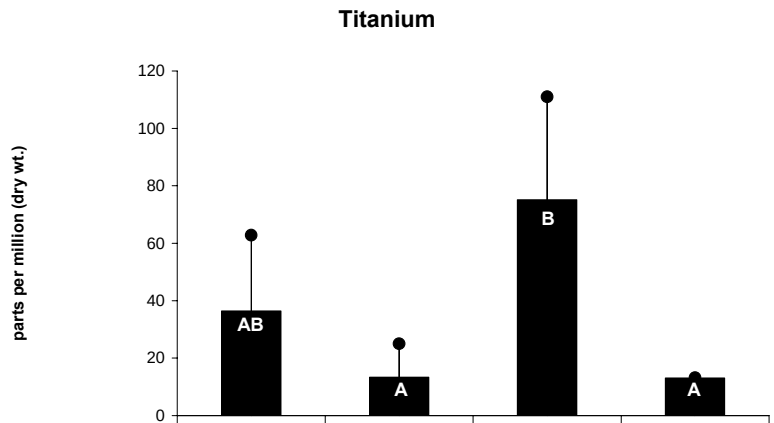


	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	2.98	5.14	3.15	10.44
● Maximum	4.44	6.92	3.71	11.50

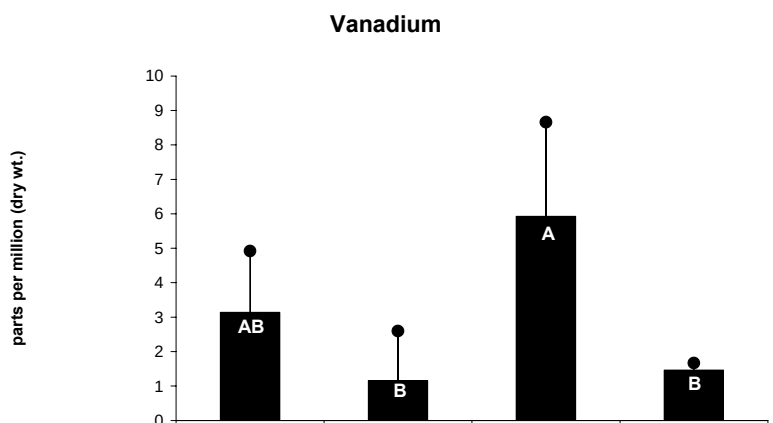
Figure 10 (cont). Mean and maximum concentrations along with statistical differences ($p \leq 0.1$) of select metals and trace-elements in stonefly (Plecoptera) larvae collected from sampling sites in East Fork Carson River and lower Bryant Creek, Nevada, October, 2001. (letters indicate significant differences among sites)



	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	0.43	0.71	0.56	2.31
● Maximum	0.47	0.72	0.56	2.37



	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	36.44212809	13.3777253	75.11129078	13.0491379
● Maximum	62.80	25.00	111.00	13.20



	EFC1 (n=3)	EFC2 (n=3)	EFC3 (n=3)	BRY (n=2)
■ Geometric Mean	3.135867961	1.16286253	5.923892068	1.462053351
● Maximum	4.92	2.6	8.66	1.67

Aquatic invertebrates are fairly limited in their movement and reflect more localized exposures to trace-elements. Metal accumulation strategies among invertebrate species can be explained by different life history habits, feeding strategies, and physiological factors affecting accumulation and depuration rates (Moore and Rainbow 1987; Phillips and Rainbow 1988). However, variability observed in invertebrate tissues in this investigation may be the result of inherent variability, not accountable by environmental or physiological factors or by man-induced alterations of hydrologic conditions previously unaccounted for. During the agricultural growing season, water in Bryant Creek is regularly diverted for irrigation purposes to River Ranch. This diversion begins 7.8 miles downstream of Leviathan Mine (California Regional Water Quality Control Board 1995). Unused water from this diversion may flow into the East Fork Carson River upstream of the confluence with Bryant Creek. Frequently during the growing season, the entire flow of Bryant Creek is diverted to the River Ranch (California Regional Water Quality Control Board 1995). However, flows in Bryant Creek during periods of diversion resume below this diversion from seepage through the diversion canal and from sub-surface flows in the creek. Another diversion from Bryant Creek located about one-quarter of a mile above the confluence of Bryant Creek with the East Fork Carson River is used to irrigate pastures north of Bryant Creek on the River Ranch and downstream of the confluence of Bryant Creek with the East Fork Carson River (California Regional Water Quality Control Board 1975). Site BRY was located below both diversions whereas site EFC3 could be impacted by return flows from the upper diversion.

3.2.2 Salmonid Tissue Exposure

The quantity of adult-sized trout captured at EFC1 and BRY was insufficient for obtaining gill, liver, and muscle tissues from either salmonid species. Therefore, samples of gill, liver, and muscle at EFC2 and EFC3 were collected only from rainbow trout. Brown trout were submitted for whole body analyses for all sites except BRY. Rainbow trout were submitted for whole body analyses from BRY because no brown trout adults were captured at this site. Analytical results for fish tissues and comparisons to potential adverse effect concentrations are provided in the sections below.

3.2.2.1 Gills

The initial site of impact of waterborne metals in freshwater fish is the gill and accumulations of metal on or in the gill are assumed to be causative of the initial damage. According to Nieboer and Richardson (1980), the relative binding affinity of a metal for biological ligands is a function of the tendency of a metal to form ionic vs. covalent bonds, as well as the chemistry of the particular ligand. Therefore, metal binding to gill surfaces could be dependent upon the ionic interactions with the epithelial tissue (Reid and MacDonald 1991). Twenty-three of 27 elements were detected in gill samples (Table 8). Comparisons of these concentrations among sites are subjective because composite samples of gill tissue from RBT were not replicated (N=1) from EFC2 and EFC3. No additional gill tissues were collected for other fishes and sites. Boron, beryllium, molybdenum, and vanadium were below detection limits. Cadmium and cobalt were detected at EFC2 but not at EFC3. Chromium was above detection limits at EFC3 but not at EFC2.

Subjective comparisons between EFC2 and EFC3 revealed concentrations of arsenic, cadmium, cobalt, iron, and zinc were higher at EFC2. Aluminum, barium, chromium, manganese, and titanium were higher at EFC3. Of these constituents, arsenic, iron, and zinc were nearly two-fold or greater at EFC2 than EFC3 (EFC2/EFC3 ratio ~ 2 or greater; Table 8). Barium and manganese were nearly two-fold or greater at EFC3 than EFC2 (EFC2/EFC3 ratio ~ 0.5 or less; Table 8).

Low levels of arsenic exist naturally in most aquatic organisms and their environments. In water, arsenic primarily exists as two species, arsenic (III) and arsenic (V). The majority of arsenic in water occurs in a soluble form with some adsorption to suspended organic matter (CCME 1992). The increased arsenic concentration observed in gill tissue at EFC2 suggests that arsenic solubility is greater in the East Fork Carson River downstream of Bryant Creek and may be influenced by AMD. Arsenic concentrations at both sites from this study were higher when compared to median brown trout gill tissues at AMD-influenced sites from the Clark Fork River, Montana (3.15 and 1.65 ppm dry weight, respectively vs. ~ 1.4 ppm; Farag et al. 1995). The elevated iron concentrations observed at EFC2 could also be the result of precipitation dynamics at the gill surface as a result of AMD-influenced discharges from the Bryant Creek drainage. Insoluble salts of iron, such as ferric hydroxide and some of its humic complexes, can be toxic by means of their tendency to precipitate (Tipping et al. 1981, Jones et al. 1993, Vegas Vilarrubia 1994) on the surface of biological membranes such as the gill. Lower pH levels can also increase the uptake and binding of zinc in gill tissues of salmonids through the dissolved fraction of water. Other studies have found similar uptake interactions between zinc and pH in salmonids (Pagenkopf 1983, Bradley and Sprague 1985, Campbell and Stokes 1985, Grippo and Dunson 1996).

The primary source of naturally occurring barium in water results from the leaching and eroding of sedimentary rocks into groundwater (Kojola et al. 1978). Although barium occurs naturally in most surface water bodies (i.e., approximately 99% of those examined) (Kopp and Kroner 1967), releases of barium to surface waters from natural sources are much lower than those to groundwater (Kojola et al. 1978). Therefore, increased barium uptake observed at EFC3 may be the result of groundwater inputs (i.e., springs) to the East Fork Carson River upstream of the Bryant Creek confluence. However, detailed hydrologic evaluations would be needed to determine the extent of those inputs, as well as their chemistry. In terms of manganese toxicity from water exposures (and potential uptake from the gill), manganese concentrations in water at EFC3 from 1998 to 1999 (ENSR 1999; Thomas and Lico 2000; Thompson and Welsh 2000) and from 2002 to 2003 (Markin and Yee 2006) were below effect levels determined by Stubblefield et al. (1997), which estimated an effect level (interpolated concentration causing adverse effects to 25 percent of a test population; IC_{25}) of 5.59 ppm and 8.68 ppm at medium to high water hardness (150 and 450 mg/L respectively as $CaCO_3$) for brown trout (*Salmo trutta*).

To maintain constant ion levels in the blood relative to the ion-poor environment, freshwater fish must take up essential electrolytes (Na^+, Ca^{2+}, Cl^-) from the water by active transport through binding sites on the gill epithelium. Cationic metal ions like Cu^+, Ag^+, Cd^{2+}, and Zn^{2+} will compete selectively with Na^+ and/or Ca^{2+} for these specific binding sites at the gills (see Wood 2001 for additional details). At acutely toxic concentrations, these cationic metals will severely interfere with the ability of the fish to perform normal ion uptake processes across the gill, and therefore passive ion losses will exceed active uptake, leading to eventual ionoregulatory failure and death (Spry and

Wood 1985; McDonald et al. 1989; Wood 1992; Wood et al. 1996, 1999). Therefore, concentrations of copper, cadmium, and zinc in gill tissues from this study were compared to predicted saturation concentrations determined by McGeer at al. (2000) for exposures to pre-determined water concentrations (Table 9). Only zinc exceeded the predicted saturation concentration for gill tissues at an aqueous exposure concentration of 250 µg/l and suggests salmonids are at a greater risk from zinc uptake in water immediately below the Bryant Creek confluence.

Table 8. Trace-element ratios between sites for gill tissues of rainbow trout (*O. mykiss*) collected from East Fork Carson River, Nevada, October 2001.

ELEMENT	EFC2/EFC3 RATIO	ELEMENT	EFC2/EFC3 RATIO
Aluminum	0.62	Mercury	1.27
Arsenic	1.91	Nickel	1.28
Barium	0.29	Phosphorus	0.90
Calcium	0.83	Potassium	1.08
Cadmium	-	Selenium	0.93
Cobalt	-	Silicon	0.85
Chromium	-	Sodium	1.11
Copper	0.84	Strontium	0.95
Iron	1.97	Sulfur	1.00
Lead	0.79	Titanium	0.58
Magnesium	0.70	Zinc	5.06
Manganese	0.51		

Total zinc concentrations in water from lower Bryant Creek have exceeded other sites on the East Fork Carson River four to eight-fold (Thomas and Lico 2000, Thompson and Welsh 2000). Zinc has been shown to inhibit calcium uptake even at levels that are considerably less than the 96-h LC50 values (Hogstrand et al. 1994, 1995). The inhibition of calcium uptake creates an imbalance in the uptake and loss rates, leading to a net loss of calcium. The gradual net loss of calcium can result in decreased plasma calcium concentrations, and eventually to hypocalcemia (Santore et al. 2002). This condition may be lethal if sufficiently high levels of zinc are sustained for an extended period of time. Impaired reproduction has been also been reported for rainbow trout exposed to as little as 50 µg/l zinc and may be an indirect effect due to inhibition of calcium uptake

(Hogstrand and Wood 1996). These effects are likely due to the fact that oocyte production requires the synthesis of vitellogenin, and this requires considerable amounts of calcium (Santore et al. 2002). The elevated exposure of zinc by salmonids may in part explain the reduction and/or absence of size classes previously observed by Lehr (2000) and the current study.

Table 9. Concentrations (ppm, wet wt) of cadmium, copper, and zinc in gill tissues of rainbow trout collected in the East Fork Carson River relative to predicted saturation concentration ranges in rainbow trout gills sub-chronically exposed to either 3 µg/l cadmium or 75 µg/l copper or 250 µg/l zinc[a] (McGeer et al. 2000).

ELEMENT	AQUEOUS EXPOSURE (µg/l)	PREDICTED SATURATION CONCENTRATION RANGE	SITE	
			EFC2	EFC3
Cadmium	3	3.0 - 3.9	0.0425	bdl [b]
Copper	75	1.6 - 2.2	0.342	0.374
Zinc	250	51.5 ± 2.3	105	19.1

Notes: [a] For cadmium and copper, prediction variables are calculated for two different exposure experiments being 65 and 100 days in length; [b] bdl = below detection limits

3.2.2.2 Liver

The liver is typically important for metal accumulation and storage in fish, especially in metal contaminated environments (Sorenson 1991, Giguére et al. 2004). In addition, the response time of the liver to short-term fluctuations in ambient metal concentrations is slower than for organs in direct contact with the external environment, such as the gills and gut (Kraemer et al. 2005), and would reflect more chronic long-term exposures. Twenty-two of 27 elements were detected in liver tissues collected from sites EFC2 and EFC3 (Table 10). Boron, beryllium, nickel, titanium, and vanadium were below detection limits in samples from both sites. Arsenic and chromium were below detection limits at EFC2. Cobalt was below detection limits at EFC3.

Comparisons of these concentrations among sites are subjective because composite samples of liver tissue from RBT were not replicated (N=1) from EFC2 and EFC3. No additional liver tissues were collected for other fishes and sites. Subjective comparisons of trace-element concentration ratios between EFC2 and EFC3 showed cadmium, cobalt and copper were higher at EFC2; whereas aluminum, arsenic, chromium, iron, manganese and silicon were higher at EFC3 (Table 10). Of these constituents, copper was over four times higher at EFC2 (EFC2/EFC3 ratio ≥ 2) and iron was nearly three times higher at EFC3 (EFC2/EFC3 ratio ≤ 0.5).

Table 10. Trace-element ratios between sites for liver samples collected from rainbow trout (*Oncorhynchus mykiss*) on the East Fork Carson River, Nevada, October 2001.

ELEMENT	EFC2/EFC3 RATIO	ELEMENT	EFC2/EFC3 RATIO
Aluminum	0.52	Manganese	0.59
Arsenic	-	Mercury	1.00
Barium	0.58	Molybdenum	0.92
Calcium	1.09	Phosphorus	0.99
Cadmium	1.79	Potassium	1.05
Cobalt	-	Selenium	1.06
Chromium	-	Silicon	0.51
Copper	4.66	Sodium	0.81
Iron	0.34	Strontium	1.06
Lead	1.09	Sulfur	1.02
Magnesium	0.98	Zinc	1.23

Comparisons of trace-element concentrations in liver to known effect concentrations determined from published studies revealed no exceedance of known adverse effect concentrations, with the exception of copper and zinc (Table 11). Copper concentrations exceeded a concentration associated with reduced survival in rainbow trout (Handy 1992). Concentrations of zinc exceeded concentrations associated with reduced reproduction in brook trout (Holcombe et al. 1979).

Copper concentrations in this study were comparable to concentrations in predatory trout collected by Moore et al. (1991) that examined a river gradient affected by upstream mine waste inputs. At the most contaminated station examined by Moore et al. (1991), the mean concentration of copper in the livers of brook trout was 311 ppm (dry weight) compared to 554 ppm, as determined by this study. Copper concentrations in water upstream of Bryant Creek have exceeded acute and/or chronic toxicity thresholds for fish in Leviathan Creek below the Mine to the confluence of Aspen Creek (Markin and Yee 2006). Copper concentrations in sediment samples collected by Thomas and Lico (2000) at the same locations observed in this study have exceeded the consensus-based threshold effect level of 31.6 determined by MacDonald et al. (2000), below which harmful effects are unlikely to be observed. Du Preez and Steyn (1992), and Seymore (1994) found the concentration of copper to be much higher in the gut than in the gills. Others have shown that increased concentrations of copper in the liver and gills were due to increased copper in the gut (Knox et al. 1982; Gatlin et al. 1989). According to Miller et al. (1993), exposure via the gut appeared to be the dominant source of copper to rainbow trout, particularly in the liver. Therefore, elevated copper concentrations in liver tissues are

likely attributed to chronic exposure and uptake from contact with and/or ingestion of contaminated sediments and potentially diet, particularly within AMD-influenced areas (EFC2 and BRY).

Table 11. Trace-element concentrations (ppm, dry wt.) in rainbow trout liver samples relative to toxicological effects concentrations for salmonids.

Element	SITE		Effect Concentration (ppm)	Type of Effect	Reference
	EFC2	EFC3			
Arsenic	bdl	1.19	1	No observable effect	Schmidt and Brumbaugh 1990
Cadmium	0.296	0.165	5.8	Reduced reproduction	Brown and Parsons 1978
Chromium	bdl	0.624	1.98	No observable effect	Calamari et al. 1982
Copper	554	119	72	Reduced (63%) survival in rainbow trout	Handy 1992
Lead	0.166	0.152	26.8	Reduced reproduction in brook trout	Holcombe et al. 1976
Mercury	1.39	1.39	8.3	No effect in brook trout	McKim et al. 1976
Nickel	bdl	bdl	2.92	No observable effect	Calamari et al. 1982
Selenium	4.75	4.5	8.84	Reduced growth in *O. mykiss*	Hilton et al. 1982
Zinc	113	91.9	66.3	Reduced reproduction in brook trout	Holcombe et al. 1979

Note: bdl = below detection limit

The concentrations of zinc suggest chronic exposure and uptake that may lead to potential reproductive impacts. Both precipitated zinc in sediments and dissolved zinc in overlying waters are bioavailable to benthic organisms (Bryan and Langston 1992). The adsorption of zinc to particles therefore does not decrease the bioavailability to aquatic invertebrates and fish, since these particles are taken up as food (Enserink et al. 1991). Zinc concentrations in sediment determined by Thomas and Lico (2000) showed elevated concentrations in lower Bryant Creek compared to East Fork Carson River sites and exceeded a threshold effect level of 121 ppm determined by MacDonald et al. (2000). Therefore, in addition to exposures previously described via the gill, elevated concentrations in liver tissues are also likely attributed to chronic exposure and uptake from contact with and/or ingestion of contaminated sediments and potentially diet.

3.2.2.3 Muscle

There is generally a correlation between levels in muscle tissue and those in other internal tissues in fish (Denton and Burdon-Jones 1996) and can provide information on potential risk to the fish themselves and to consumers of those fish. Mean concentrations of trace-elements collected from rainbow trout at EFC2 and EFC3 are presented in Table 12. Twenty of 27 elements were detected in muscle tissues. Trace-elements below detections limits for both sites included arsenic, boron, beryllium, cadmium, chromium, cobalt, molybdenum, and vanadium. Lead, manganese, potassium, selenium, and sulfur were significantly different between sites. Lead, potassium and sulfur were higher at EFC2, whereas manganese and selenium were higher at EFC3.

Table 12. Geometric means (range) of trace-element concentrations (ppm, dry wt.) in rainbow trout (*Oncorhynchus mykiss*) muscle collected from East Fork Carson River, Nevada, October 2001.

ELEMENT	SITE		EFC2/EFC3 RATIO
	EFC2 *(n=5)*	**EFC3** *(n=3)*	
Aluminum	7.25 (< 2.6 – 22.2)	7.37 (< 2.08 – 25.2)	0.98
Barium	0.16 (< 0.54 – 0.294)	0.45 (0.224 – 0.707)	0.36
Calcium	1072 (582 – 3390)	2207 (896 – 4240)	0.49
Copper	1.93 (1.39 – 2.25)	1.48 (1.26 – 1.61)	1.30
Iron	22.20 (16.6 – 46.5)	29.68 (21.9 – 36.4)	0.75
Lead*	0.24 (0.117 – 1.96)	0.09 (< 0.45 – 0.12)	2.67
Magnesium	1254 (1240 – 1270)	1196 (1150 – 1230)	1.05
Manganese*	1.14 (0.69 – 1.45)	2.07 (0.94 – 3.34)	0.55
Mercury	0.97 (0.801 – 1.29)	1.01 (0.915 – 1.23)	0.96
Nickel	bdl (< 0.27 – 0.61)	0.47 (< 0.25 – 0.65)	-
Phosphorus	11654 (11300 – 12400)	11879 (11000 – 12700)	0.98
Potassium*	18278 (17800 – 18500)	16526 (16000 – 17200)	1.11
Selenium*	0.54 (0.429 – 0.622)	0.85 (0.641 – 1.07)	0.64
Silicon	35.42 (27.5 (54.2)	19.48 (4.15 – 58.4)	1.82
Sodium	1134 (1040 – 1220)	988 (854 – 1170)	1.15
Strontium	3.03 (1.41 – 10.2)	5.73 (2.11 – 11.9)	0.53
Sulfur*	8974 (8460 – 9670)	7782 (7400 – 8390)	1.15
Zinc	17.95 (15.7 – 25.1)	15.24 (12.8 – 17.3)	1.18

Notes: bdl = greater than 50 percent of samples collected were below detection limits; * indicates statistically significant difference among sites

Trace-element concentrations in muscle were compared to USEPA screening values (SV) to assess potential human exposure risks. USEPA SV's are defined as concentrations of target analytes in fish tissue that are of potential public health concern and that are used as threshold values against which levels of contamination in similar tissue collected from the ambient environment can be compared (USEPA 2000). Of the calculated SV's established for trace-elements, only mercury exceeded the SV for both recreational and subsistence fishers (Table 13).

Table 13. Comparison of USEPA Screening Values (SV) to geometric mean concentrations (parts per million, dry weight) of rainbow trout muscle from the East Fork Carson River, October 2001. (bdl= > 50% of samples were below detection limits)

ELEMENT	Screening Value (USEPA 2000)	Combined Geometric Mean
Arsenic	0.147[a] – 1.2[b]	<0.3
Cadmium	0.491[a] – 4.0 [b]	<0.04
Mercury	0.049[a] – 0.4 [b]	0.99
Selenium	2.457[a] – 20.0 [b]	0.64

Notes: [a] Based on a subsistence fish consumption rate of 142.4 g/d, 70kg body weight; [b] Based on a recreational fish consumption rate of 17.5 g/d, 70kg body weight.

Previous data indicated mercury water concentrations in this reach of the East Fork Carson River were low at <0.2 ppb (Thomas and Lico 2000; Thompson and Welsh 2000). However, data from Thomas and Lico (2000) revealed mean total mercury concentrations in sediment at BRY were six to 13 times greater than other sites on the East Fork Carson River. Benthic invertebrates may take up mercury from sediments, making it available to other aquatic animals through the food chain and to vertebrates that consume emergent aquatic insects (Hildebrand et al. 1980; Wren and Stephenson 1991; Dukerschein et al. 1992; Saouter et al. 1993; Suchanek et al. 1993; Tremblay et al. 1996). However, invertebrates collected by this investigation and others (see Table 16) were lower in total mercury at BRY compared to other sites suggesting that mercury in lower Bryant Creek is not as bio-available as it is in the East Fork Carson River. Sources of mercury in the East Fork Carson River could include geologic formations, geothermal springs, up-gradient anthropogenic sources, and atmospheric deposition (Fischer and Gustin 2002). However, more intensive site-specific monitoring and/or evaluation should be conducted to determine bioavailability in the East Fork Carson River.

3.2.2.4 Whole Body

Geometric mean and range of concentrations in salmonid whole bodies are presented in Table 14. Twenty-four of 27 elements were detected. Boron, beryllium, and molybdenum were below detection limits. Only cobalt and mercury concentrations were significantly different among sites. Mercury was the most elevated at EFC1 whereas cobalt was the most elevated at EFC2.

Trace-element concentrations were compared to known effect concentrations determined from published studies to identify trace-elements that may be affecting fish in the East Fork Carson River and lower Bryant Creek (Table 15). All sample concentrations for arsenic, cadmium, mercury, and selenium were below known effect levels. One sample concentration for copper from EFC2 exceeded an effect level associated with reduced weight gain and increased mortality.

All samples from the East Fork Carson River sites (EFC1, EFC2, EFC3) had zinc concentrations that exceeded estimated normal background concentrations for fish. Based upon observations of elevated zinc concentrations in gill and liver tissues combined with previous determinations of elevated water and sediment, zinc uptake and distribution throughout salmonids are occurring.

Table 14. Geometric mean (range) of trace-element concentrations (ppm, dry wt.) in salmonid whole bodies collected from East Fork Carson River, Nevada, October 2001.

	SITE			
	EFC1	**EFC2**	**EFC3**	**BRY**
Species	(*Salmo trutta*)	(*Salmo trutta*)	(*Salmo trutta*)	(*Oncorhynchus mykiss*)
No. of fish	4	5	4	2
Avg. fish size (g)	217.6	149.8	256.1	155.6
Aluminum	18.21 (9.16 - 46.2)	50.68 (7.38 – 353)	39.13 (15.5 - 59.8)	77.29 (37.1 – 161)
Arsenic	Bdl (<0.298 - <0.324)	1.02 (<0.312 - 2.58)	0.92 (<0.31 - 2.27)	0.49 (<0.32 - 1.5)
Barium	1.28 (0.93 - 1.69)	1.94 (0.614 - 3.88)	2.47 (1.19 - 5.23)	2.8 (2.52 - 3.11)
Cadmium	Bdl (<0.039 - <0.051)	Bdl (<0.034 - 0.055)	Bdl (<0.03 - 0.11)	0.04 (<0.05 - 0.066)
Calcium	12937 (8860 – 26300)	13090 (5540 – 46700)	13536 (9500 – 19300)	8541 (5790 – 12600)
Chromium	1.39 (0.821 - 3.68)	2.3 (0.535 - 11.1)	0.64 (0.557 - 0.683)	0.55 (<0.51 - 1.19)
Cobalt*	Bdl (<0.497 - <0.541)	1.3 (0.925 - 2.53)	Bdl (<0.51 - <0.53)	0.73 (0.59 - 0.914)
Copper	8.76 (5.98 - 11.1)	7.83 (4.15 - 19.8)	6.77 (6.07 - 8.66)	3.9 (<3.67 - 4.15)
Iron	65 (38.5 - 93.8)	156.3 (82.1 – 521)	64.7 (49.3 - 78.7)	173.8 (148 – 204)
Lead	Bdl (<0.078 - <0.102)	Bdl (<0.069 - 0.453)	0.10 (<0.09 - 0.165)	0.13 (0.13 - 0.137)
Magnesium	1022.9 (896 - 1290)	999.4 (811 – 1260)	1000 (820 – 1280)	993.4 (958 – 1030)
Manganese	6.3 (4.67 - 8.2)	11.33 (2.94 - 21.4)	13.9 (6.35 - 23.7)	26.9 (17.7 - 40.8)
Mercury*	1.023 (0.85 – 1.17)	0.749 (0.55 – 1.01)	0.95 (0.75 – 1.24)	0.34 (0.20 – 0.56)
Nickel	0.37 (<0.5 - 1.08)	0.87 (<0.515 - 3.42)	Bdl (<0.51 - <0.53)	0.86 (0.75 - 1.0)
Potassium	12149 (11900 - 12400)	11600 (10700 – 13100)	11101 (10400 – 12200)	11184 (10600 - 11800)
Selenium	0.86 (0.687 - 1.08)	0.82 (0.724 - 0.905)	0.76 (0.628 - 0.925)	1.9 (1.44 - 2.5)
Silicon	49.5 (33.6 - 111)	81.2 (31.3 – 156)	82.6 (47.2 – 118)	114.3 (90.7 – 144)
Sodium	3048 (2730 - 3250)	2773 (2410 – 3020)	2532 (2250 - 2800)	2549 (2110 – 3080)
Strontium	43.5 (29.9 - 89.1)	45.4 (18.9 – 153)	47 (34.6 - 65.2)	24.1 (18.6 - 31.2)
Sulfur	7737 (7220 - 8230)	7377 (6630 – 8150)	6688 (6290 - 6950)	6985 (6940 - 7030)
Titanium	1.14 (0.709 - 2.56)	2.18 (<0.521 - 12.6)	2.70 (1.23 - 4.54)	5.65 (1.32 - 24.2)
Vanadium	Bdl (<0.99 - <1.08)	Bdl (<0.991 - <1.04)	Bdl (<1.01 - <1.07)	1.00 (<1.02 - 1.98)
Zinc	140.8 (122 - 161)	148.7 (115 – 196)	153.4 (125 – 197)	74.0 (68.9 - 79.5)

Note: * indicates statistically significant difference among sites; *bdl* indicates >50 percent of samples in matrix were below detection limits).

Table 15. Comparison of trace-element concentrations (parts per million, dry weight) in whole bodies of brown trout (*Salmo trutta*) and rainbow trout (*Oncorhynchus mykiss*) to published adverse effect levels.

ELEMENT	SITE AND SPECIES				EFFECT CONCEN-TRATION	TYPE OF EFFECT	REFERENCE
	EFC1 (*S. trutta*) n=4	EFC2 (*S. trutta*) n=5	EFC3 (*S. trutta*) n=4	BRY (*O. mykiss*) n=2			
Arsenic	< 0.149 - < 0.162	< 0.156 - 2.58	< 0.156 - 2.27	< 0.158 - 1.5	3.1	Reduced growth of *O. mykiss*	Cockrell and Hilton 1988
Cadmium	< 0.02 - < 0.025	< 0.017 - 0.055	< 0.017 - 0.11	< 0.025 - 0.066	1.6	Reduced growth of *O. mykiss*	Kumada et al. 1973
Copper	5.98 - 11.1	4.15 - 19.8	6.07 - 8.66	3.67 - 4.15	13.3	Significant decrease in weight gain and increase in mortality of *O. mykiss*	Julshamn et al. 1988
Mercury (wet weight)	(0.216) - (0.316)	(0.141) - (0.29)	(0.21) - (0.33)	(0.056) - (0.156)	10 (wet weight)	Sub-lethal or lethal toxic effects in *O. mykiss*	Wiener and Spry 1996
Selenium	0.687 - 1.08	0.724 - 0.905	0.628 - 0.925	1. 4 - 2.45	4	Health and reproductive effects to freshwater and anadromous fish	Lemly 1996
Zinc	122 - 161	115 - 196	125 - 197	68.9 - 79.5	88	Normal background level among fish	Schmitt and Brumbaugh 1990

3.2.3 Potential Salmonid Dietary Exposure and Assessment of Effects

Trace-element concentrations in invertebrate samples collected from this investigation and by the Fish and Wildlife Service in 1998 for the same sites (Thompson and Welsh 1999) were compared to known dietary effect levels determined for salmonids from published literature in order to assess risks to salmonids in the East Fork Carson River and lower Bryant Creek (Table 16). Cadmium, selenium and vanadium were the only trace-elements identified to have concentrations that would be of concern for dietary risks to trout. Cadmium concentrations in stoneflies collected by this study did not exceed any known adverse dietary effect level for salmonids. However, mayflies collected by Thompson and Welsh (1999) did exceed a dietary effect level associated with reduced survival and growth in rainbow trout fry. Selenium in invertebrates collected by this study at site BRY approached but did not exceed dietary effect levels associated with impaired survival or development of trout. Vanadium concentrations in invertebrate samples collected by Thompson and Welsh (1999) approached or exceeded a dietary effect level associated with reduced growth and feeding response in trout from the three sites on the East Fork Carson River.

A comparison of the ratios of mean trace-element concentrations in fish tissue (liver, muscle, and whole body) to aquatic invertebrates at the same sites provides indication of the potential for bioaccumulation through a dietary item (Figure 11). Cadmium, copper, mercury, and selenium showed accumulations in liver tissue. Cadmium showed only slight accumulation in the liver and is consistent with previous studies that have shown that it does not accumulate significantly in the muscle (Harrison and Klaverkamp 1989; Sorensen 1991; Szebedinszky et al. 2001; Chowdhury et al. 2003, 2004). The ratio of copper in liver to invertebrate tissue was the highest of the trace-element ratios examined. As discussed previously, elevated copper concentrations in liver tissues are likely attributed to chronic exposure and uptake from contact with and/or ingestion of contaminated sediments and potentially diet.

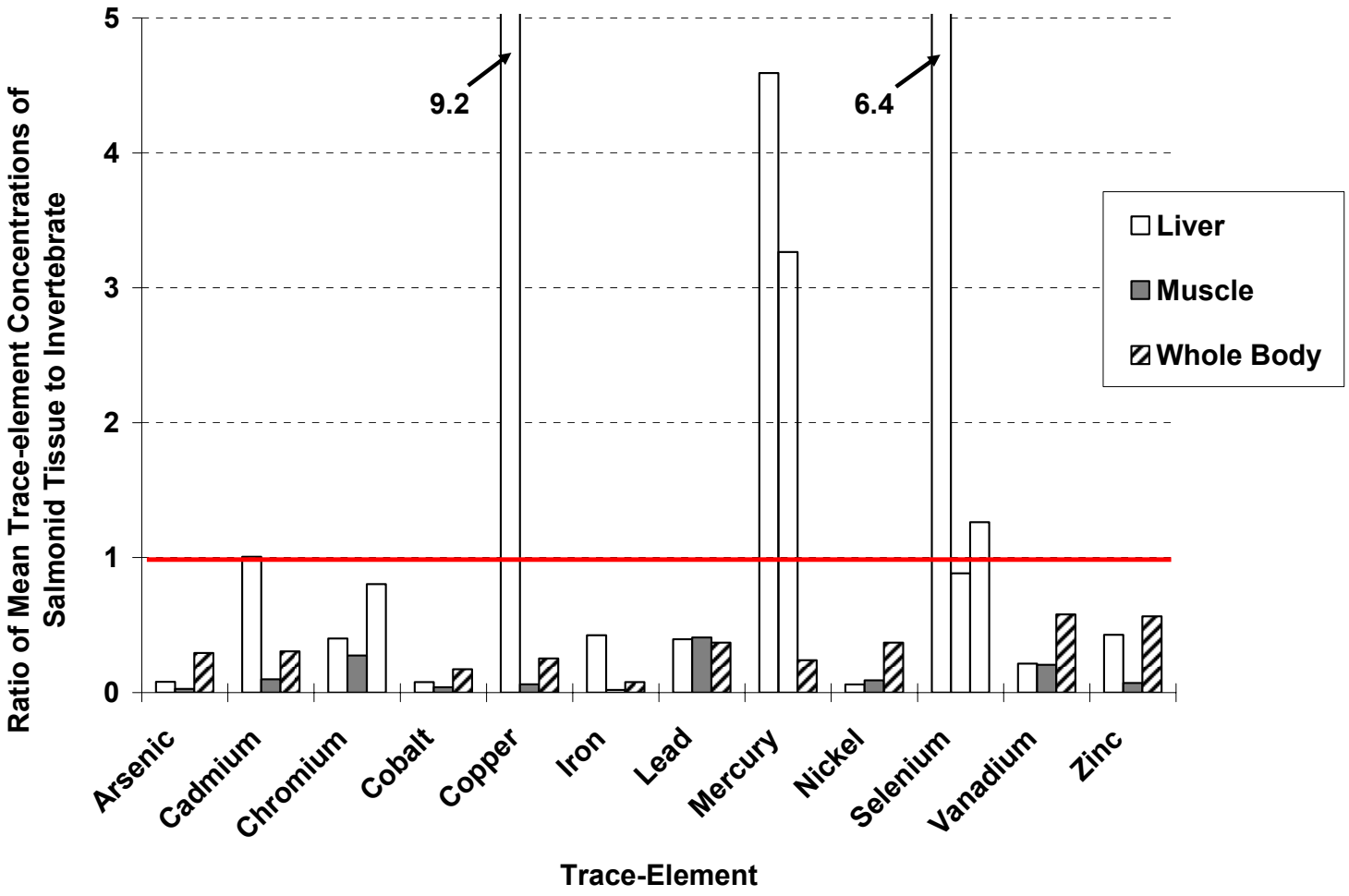
Mercury, lead, and selenium showed accumulations in muscle tissue. Mercury and selenium also accumulated in whole body as shown by the ratios of whole body to invertebrates for all sites combined.

Fish exposed to methylmercury in laboratory tests at first had greater concentrations in the blood, spleen, kidney, and liver than in muscle (McKim et al. 1976). Harrison and Klaverkamp (1990) found that after exposure was discontinued, the mercury concentration increased in muscle while decreasing in other organs and tissues. Based upon those observations and the current bioaccumulation data from this study, mercury appears to be accumulating in salmonids at a constant rate and retaining the mercury for redistribution throughout the body (liver avg. = 1.39 ppm; muscle avg. = 1.01 ppm; whole body avg. = 0.82 ppm). For selenium, muscle generally contains less concentration than whole body due to the relatively high amounts of selenium found in spleen, liver, kidney, heart, and other tissues (Hilton et al. 1982; Kleinow and Brooks 1986; Lemly and Smith 1987; Hermanutz et al. 1992).

Table 16. Summary of concentrations (in parts per million, dry weight; ppm) of select contaminants of concern from aquatic invertebrate samples collected in the East Fork Carson River and lower Bryant Creek with comparisons to known dietary effects to fish identified in published literature [SF= geometric mean of stoneflies from this study; CF= caddisflies from Thompson and Welsh (1999); MF= mayflies from Thompson and Welsh (1999)].

Trace-element	EFC1			EFC2			EFC3			BRY			Effect concen-tration (ppm)	Type of Effect	Reference
	SF *n=3*	CF *n=1*	MF *n=1*	SF *n=3*	CF *n=1*	MF *n=1*	SF *n=3*	CF *n=1*	MF *n=1*	SF *n=2*	CF *n=1*	MF *n=1*			
As	3.58	5.05	4.53	9.2	6.28	7.39	7.79	5.77	5.45	2.94	9.8	13.4	90	Initial negative growth in rainbow trout gave way to slow positive growth over time	Cockell and Hilton 1988
Cd	0.27	0.59	5.75	.045	0.381	2.19	0.10	0.380	0.386	0.17	0.978	5.6	2.39	Observed reduction in survival and growth of rainbow trout fry	Woodward et al 1994
Cu	28.4	17.6	24.0	27.6	23.2	18.0	27.6	9.55	17.7	27.8	39.5	18.9	660	Reduced survival in rainbow trout fry	Mount et al. 1994
Hg	0.31	0.161	0.135	0.40	0.249	0.149	0.30	0.249	0.047	0.19	0.0872	0.0735	10	Pathological brain damage, significantly reduced neural enzyme activity, and reduced overall post-feeding activity behavior in Atlantic salmon (*Salmo salar*)	Berntssen et al. 2003
Se	0.43	<0.216	<0.54	0.71	<0.50	<0.62	0.56	<0.332	<1.09	2.31	1.84	0.858	3 to 8	Impairment of juvenile survival and/or development in salmonids; reproductive failure in fish	Hamilton et al. 1990; Lemly 1996
V	3.1	10.3	9.08	1.2	9.08	11.4	5.9	9.74	27.1	1.5	3.52	9.14	10.2	Reduced growth and feeding response in rainbow trout	Hilton and Bettger 1988
Zn	227	123	172	257	135	138	216	125	42.2	263	138	374	1,500	No observable adverse effect in rainbow trout	Mount et al. 1994

Figure 11. Ratios of mean trace-element concentrations of fish tissue (liver, muscle, and whole body) to mean aquatic invertebrate (stoneflies) trace-element concentrations from the East Fork Carson River, Nevada. (Solid red line represents equal concentrations for fish tissue and invertebrates).



4.0 CONCLUSIONS

Data collected by this investigation indicated that fish communities are impaired in the East Fork Carson River. The fewest number of species observed in this study were below the Bryant Creek confluence. The presence/absence of sensitive species (Paiute sculpin) combined with observations of marked increases of abundance/density of tolerant species (mountain sucker) in the East Fork Carson River immediately below the Bryant Creek confluence may be the result of influences on water or habitat quality from AMD. These observations are consistent from other studies that assessed impacts from AMD-influenced drainages (Platts et al. 1979, Farag et al. 2003). IBI scores for each sampling site also declined on the East Fork Carson River downstream of the Bryant Creek confluence and were considered marginally impaired compared to upstream of the Bryant Creek confluence which was considered acceptable. However, additional data on fish population dynamics would need collected on an annual basis and in different seasons to confirm these observations.

Although the Nevada Department of Wildlife artificially stocks brown and rainbow trout within the study area, length-frequency data appears to show that brown trout and mountain whitefish have one or more age-classes missing. In addition, rainbow trout reproduction and recruitment is absent. The missing age-classes may be the result of exposure to contaminants or a response to slow growth patterns from other environmental factors. Conditions of brown and rainbow trout were within expected ranges of what is considered healthy. However, mountain whitefish had reduced condition and further analysis showed individuals were much below their expected relative weight. The observations of relative abundance, community composition, and fish condition indicate some impairment, particularly in lower Bryant Creek and the East Fork Carson River immediately below the Bryant Creek confluence. However, as mentioned previously, additional data would need collected on an annual basis and in different seasons to confirm these observations.

Concentrations of trace-elements in aquatic invertebrates were variable among sites. The variability observed in invertebrate tissues in this investigation may be the result of inherent variability or from man-induced alterations of hydrologic conditions since water is diverted from Bryant Creek and returned to the East Fork Carson River above its confluence. However, invertebrates in lower Bryant Creek and immediately below its confluence with the East Fork Carson River were elevated in cadmium, cobalt, mercury, nickel, and selenium.

With respect to salmonids, this investigation assessed 1) potential trace-element exposure; 2) potential risks by comparing trace-element data in tissues to adverse effect concentrations determined from published literature; and 3) potential bioaccumulation and risks from dietary uptake. While assessment of exposure and uptake by salmonids from trace-element concentrations in water or sediment pathways was outside the scope of this investigation, limited information was available on trace-elements in water and sediment from previous studies and was included as appropriate to assist in the assessments for adverse effects to salmonids. Based upon the trace-element concentrations observed in tissues, salmonids were exposed to elevated sources of arsenic, cobalt, copper, iron, lead, mercury, and zinc.

The elevated exposures of arsenic and copper are consistent with environments impacted by AMD-influenced drainage. Arsenic concentrations in gill tissues were

higher than those compared to median brown trout gill tissues at AMD-influenced sites from the Clark Fork River, Montana. Copper concentrations in both liver and whole body tissues exceeded concentrations associated with reduced survival and increased mortality in salmonids and were comparable to another investigation that observed trout affected by AMD in the Blackfoot River, Montana.

Cobalt concentrations in aquatic invertebrates were significantly higher in the East Fork Carson River immediately below the Bryant Creek confluence compared to other sites. Cobalt was detected in gill and liver tissues of rainbow trout in the East Fork Carson River immediately below Bryant Creek but not above it. Cobalt was also detected in whole fish only in Bryant Creek and the East Fork Carson River immediately below Bryant Creek. However, concentrations in invertebrates were higher than those found in fish tissue indicating a low hazard for dietary bioaccumulation in fish. Information on adverse effect concentrations in invertebrates and fish tissues is lacking and due to the limited toxicological data for cobalt, no current national water quality criteria have been established in the United States. Therefore, assessing toxicological risks to salmonids from cobalt is difficult.

Mercury concentrations in salmonid muscle tissues from all sites exceeded USEPA SV's which indicates a potential public health concern for consumption, especially for individuals that may subsist on fish diets in the East Fork Carson River.

Zinc concentrations in salmonid gill tissues from the East Fork Carson River immediately below the confluence of Bryant Creek exceeded predicted saturation limits and suggested uptake from the water environment. Previous data collected by other investigators found zinc concentrations in water were elevated in lower Bryant Creek and exceeded other sites on the East Fork Carson River four to eight-fold. Other investigators also found zinc concentrations in sediment in lower Bryant Creek to exceed a threshold effect concentration below which harmful effects are unlikely to be observed. Therefore, the uptake of zinc in salmonids likely is from both water and sediments and would explain the elevated levels observed in liver tissue at concentrations that exceeded effect levels associated with reduced survival and/or reproduction. The elevated concentrations of zinc observed in whole body samples also support the likely uptake from water/sediment/diet that lead to concentrations that exceed normal background concentrations.

Trace-elements that had the potential for elevated bioaccumulation in salmonids tissues included copper, mercury, and selenium. The high concentrations of copper observed in liver tissue contributed to the observed high bioaccumulation potential from dietary sources. However, from a dietary perspective, concentrations of copper in invertebrates from this investigation and others did not exceed known adverse effects. Previous data on sediment concentrations for copper within the study area have exceeded the consensus-based threshold effect concentration below which harmful effects are unlikely to be observed. Therefore, elevated copper concentrations in liver tissues are likely attributed to chronic exposure and uptake from contact with and/or ingestion of contaminated sediments and potentially diet, particularly within AMD-influenced areas. Mercury also showed an expected bioaccumulation potential through dietary sources. However, based on data from this investigation and others, mercury in lower Bryant Creek is not as bioavailable as it is in the East Fork Carson River and its source may be from diffuse sources other than AMD. However, more intensive site-specific evaluations need to be conducted to determine mercury sources in the East Fork Carson River. Selenium concentrations in invertebrates from BRY in this investigation were

significantly higher than other sites and began to approach a toxic effect threshold of 3.0 ppm identified by Lemly (1996). Selenium is strongly bioaccumulated in aquatic organisms (Lemly 1996) and relatively low concentrations in water can quickly become concentrated to potentially toxic levels in aquatic organisms. Selenium appeared to accumulate in whole bodies of trout in lower Bryant Creek most likely from the dietary pathway. The body burdens for selenium were not elevated enough to exceed an identified adverse effect level. Waterborne concentrations in the low part per billion range can lead to reproductive failure in adult fish with little or no additional symptoms of selenium poisoning in the environment (Lemly 1996). Maier and Knight (1994) summarized seven recent studies that provided estimates of waterborne thresholds for food chain-mediated toxicity to fish and wildlife. All reports concluded that toxic thresholds were less than or equal to 0.003 ppm in water. Detection limits for water data collected previously by other investigators (Thomas and Lico 2000; Thompson and Welsh 2000; Markin and Yee 2006) within the current study area were above the described threshold values and prevented determination of potential adverse effects from water concentrations. However, sediment concentrations collected by Thomas and Lico (2000) in lower Bryant Creek exceeded a biological effect concentration (EC_{10}) of 2.5 ppm identified in Skorupa (1998). However, due to selenium's bioaccumulative properties, further information should be collected to identify risks to higher trophic levels of the food chain within the environment of lower Bryant Creek.

The observations of fish community impairment combined with elevated concentrations of trace-elements in aquatic invertebrate and salmonid tissues, some of which exceeded concentrations associated with adverse effects, provide evidence that AMD drainage from Leviathan Mine may be having negative impacts on aquatic biota in the East Fork Carson River. These impacts would likely occur from chronic long-term exposures to a variety of trace-elements. The magnitude of exposures to trace-elements is temporally and spatially variable and likely affected by irrigation diversions of Bryant Creek. Further investigation is needed to determine the transport and fate of trace-elements in lower Bryant Creek and its influence on the East Fork Carson River, especially during spring snow-melt and irrigation diversions. Furthermore, additional data on trace-element exposures in aquatic biota needs to be collected in areas further upstream of the Bryant Creek confluence to determine reference or background concentrations that may be influenced by naturally occurring sources.

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Appendix 1. Meta-data for samples collected for trace-element analysis which includes: sample number; location collected (from fig. 1); type of sample collected, type of species collected, weight of sample, and percent moisture of sample.

Sample #	Site	Sample Type	Scientific Name	Common Name	Sample Weight	% Moisture
01LMIV11	EFC1	invertebrate-composite	*Skwala spp.*	Stonefly	12.824	82.9
01LMIV12	EFC1	invertebrate-composite	*Skwala spp.*	Stonefly	14.956	85.8
01LMIV13	EFC1	invertebrate-composite	*Skwala spp.*	Stonefly	22.401	84.7
01LMIV21	EFC2	invertebrate-composite	*Skwala spp.*	Stonefly	16.043	87.2
01LMIV22	EFC2	invertebrate-composite	*Skwala spp.*	Stonefly	18.144	85.5
01LMIV23	EFC2	invertebrate-composite	*Skwala spp.*	Stonefly	11.259	86.8
01LMIV31	EFC3	invertebrate-composite	*Skwala spp.*	Stonefly	17.815	83.5
01LMIV32	EFC3	invertebrate-composite	*Skwala spp.*	Stonefly	12.835	87.1
01LMIV33	EFC3	invertebrate-composite	*Skwala spp.*	Stonefly	22.01	87.3
01LMIV42	BRY	invertebrate-composite	*Skwala spp.*	Stonefly	10.715	85.5
01LMIV43	BRY	invertebrate-composite	*Skwala spp.*	Stonefly	20.995	85.9
01LMFG20	EFC2	gill-composite	*Oncorhynchus mykiss*	Rainbow trout	19.631	76.4
01LMFG30	EFC3	gill-composite	*Oncorhynchus mykiss*	Rainbow trout	25.099	78.4
01LMFL20	EFC2	liver-composite	*Oncorhynchus mykiss*	Rainbow trout	17.223	75.6
01LMFL30	EFC3	liver-composite	*Oncorhynchus mykiss*	Rainbow trout	18.818	76.4
01LMFM21	EFC2	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	51.543	79.1
01LMFM22	EFC2	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	70.744	78.3
01LMFM23	EFC2	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	65.379	76.7
01LMFM24	EFC2	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	90.511	77.8
01LMFM25	EFC2	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	59.893	76.7

Appendix 1 (cont). Meta-data for samples collected for trace-element analysis which includes: sample number; location collected (from fig. 1); type of sample collected, type of species collected, weight of sample, and percent moisture of sample.

Sample #	Site	Sample Type	Scientific Name	Common Name	Sample Weight	% Moisture
01LMFM31	EFC3	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	86.85	75.3
01LMFM32	EFC3	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	78.373	76.5
01LMFM33	EFC3	muscle-individual	*Oncorhynchus mykiss*	Rainbow trout	54.103	78.4
01LMFW11	EFC1	whole body-individual	*Salmo trutta*	Brown trout	241.307	72.8
01LMFW12	EFC1	whole body-individual	*Salmo trutta*	Brown trout	221.098	72
01LMFW13	EFC1	whole body-individual	*Salmo trutta*	Brown trout	212.776	73
01LMFW14	EFC1	whole body-individual	*Salmo trutta*	Brown trout	195.272	74.5
01LMFW21	EFC2	whole body-individual	*Salmo trutta*	Brown trout	69.41	74.9
01LMFW22	EFC2	whole body-individual	*Salmo trutta*	Brown trout	38.814	74.2
01LMFW23	EFC2	whole body-individual	*Salmo trutta*	Brown trout	250.452	72.2
01LMFW24	EFC2	whole body-individual	*Salmo trutta*	Brown trout	177.81	71.3
01LMFW25	EFC2	whole body-individual	*Salmo trutta*	Brown trout	212.512	71.6
01LMFW31	EFC3	whole body-individual	*Salmo trutta*	Brown trout	253.962	73.1
01LMFW32	EFC3	whole body-individual	*Salmo trutta*	Brown trout	291.529	72.4
01LMFW33	EFC3	whole body-individual	*Salmo trutta*	Brown trout	181.22	72.1
01LMFW34	EFC3	whole body-individual	*Salmo trutta*	Brown trout	297.824	69.6
01LMFW41	BRY	whole body-individual	*Oncorhynchus mykiss*	Rainbow trout	181.205	72.5
01LMFW42	BRY	whole body-individual	*Oncorhynchus mykiss*	Rainbow trout	130.085	72

Appendix 2. Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	Al	As	B	Ba	Be	Ca	Cd	Co	Cr
01LMIV11	EFC1	invertebrate-composite	1440	6.39	9.53	23.4	0.103	1880	0.256	4.01	1.31
01LMIV12	EFC1	invertebrate-composite	679	2.84	3.17	12.2	0.056	1580	0.279	3.93	0.712
01LMIV13	EFC1	invertebrate-composite	634	2.52	3.06	11.4	<0.0552	1430	0.285	3.62	2.08
01LMIV21	EFC2	invertebrate-composite	312	6.84	4.25	15.4	<0.0538	2310	0.468	14.1	<0.538
01LMIV22	EFC2	invertebrate-composite	392	7.91	4.06	16.2	<0.0548	2210	0.427	14.8	<0.548
01LMIV23	EFC2	invertebrate-composite	733	14.4	5.78	17.7	0.0847	3000	0.452	15	0.66
01LMIV31	EFC3	invertebrate-composite	1440	6.18	6.48	32.5	0.104	2570	0.0916	4.62	11.5
01LMIV32	EFC3	invertebrate-composite	1380	7.77	11.1	33.7	0.121	2800	0.105	4.76	1.38
01LMIV33	EFC3	invertebrate-composite	974	9.84	9.38	35.3	0.117	2450	0.107	4.13	1.42
01LMIV42	BRY	invertebrate-composite	560	3.06	<1.05	14.2	0.0829	4650	0.18	5.46	0.594
01LMIV43	BRY	invertebrate-composite	501	2.83	<1.11	20	0.0877	4500	0.169	8.1	1.11
01LMFG20	EFC2	gill-composite	50.5	3.15	<1.05	5.46	<0.0526	33300	0.18	1.42	<0.526
01LMFG30	EFC3	gill-composite	81.9	1.65	<1.02	18.6	<0.0508	40100	<0.043	<0.508	0.68

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	Al	As	B	Ba	Be	Ca	Cd	Co	Cr
01LMFL20	EFC2	liver-composite	7.17	<0.328	<1.09	0.109	<0.0546	292	0.296	0.939	<0.546
01LMFL30	EFC3	liver-composite	13.8	1.19	<1.04	0.188	<0.0522	268	0.165	<0.522	0.624
01LMFM21	EFC2	muscle-individual	6.31	<0.315	<1.05	0.294	<0.0525	3390	<0.0501	<0.525	<0.525
01LMFM22	EFC2	muscle-individual	<5.2	<0	<1.04	0.208	<0.052	918	<0.0487	<0.52	<0.52
01LMFM23	EFC2	muscle-individual	22.2	<0.312	<1.07	0.277	<0.0533	808	<0.0365	<0.533	<0.533
01LMFM24	EFC2	muscle-individual	6.34	<0.327	<1.09	0.131	<0.0545	967	<0.0437	<0.545	<0.545
01LMFM25	EFC2	muscle-individual	8.7	<0.323	<1.08	<0.108	<0.0539	582	<0.0432	<0.539	<0.539
01LMFM31	EFC3	muscle-individual	7.64	<0.305	<1.02	0.224	<0.0508	896	<0.0447	<0.508	<0.508
01LMFM32	EFC3	muscle-individual	<4.16	<0.25	<0.833	0.591	<0.0416	4240	<0.0383	<0.416	<0.416
01LMFM33	EFC3	muscle-individual	25.2	<0.295	<0.982	0.707	<0.0491	2830	<0.0453	<0.491	0.598
01LMFW11	EFC1	whole body-individual	46.2	<0.298	<0.994	1.65	<0.0497	26300	<0.0390	<0.497	3.68
01LMFW12	EFC1	whole body-individual	12.2	<0.300	<1.00	0.93	<0.0500	8860	<0.0506	<0.500	1.13
01LMFW13	EFC1	whole body-individual	21.3	<0.324	<1.08	1.04	<0.0541	12600	<0.0511	<0.541	0.821
01LMFW14	EFC1	whole body-individual	9.16	<0.306	<1.02	1.69	<0.0510	9540	<0.0511	<0.510	1.08

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	Al	As	B	Ba	Be	Ca	Cd	Co	Cr
01LMFW21	EFC2	whole body-individual	169	1.81	<1.00	3.18	<0.0502	5660	0.0545	2.53	5.61
01LMFW22	EFC2	whole body-individual	353	1.14	<1.04	3.88	<0.0521	46700	0.0513	1.52	11.1
01LMFW23	EFC2	whole body-individual	43.4	2.58	<0.991	1.66	<0.0496	14500	<0.0463	1.11	0.623
01LMFW24	EFC2	whole body-individual	7.38	0.156	<1.04	0.614	<0.0521	5540	<0.0393	0.925	3.07
01LMFW25	EFC2	whole body-individual	17.5	1.3	<1.03	2.18	<0.0515	18100	<0.0344	0.926	0.535
01LMFW31	EFC3	whole body-individual	59.8	2.27	<1.07	5.23	<0.0533	9500	<0.0433	<0.533	0.671
01LMFW32	EFC3	whole body-individual	51	1.23	<1.01	1.95	<0.0507	19300	<0.0479	<0.507	0.557
01LMFW33	EFC3	whole body-individual	49.6	1.63	<1.03	3.08	<0.0515	10900	0.11	<0.515	0.638
01LMFW34	EFC3	whole body-individual	15.5	0.1555	<1.04	1.19	<0.0518	16800	<0.0347	<0.518	0.683
01LMFW41	BRY	whole body-individual	37.1	1.5	<1.02	3.11	<0.0509	12600	0.066	0.914	<0.509
01LMFW42	BRY	whole body-individual	161	0.1575	<1.05	2.52	<0.0525	5790	<0.0493	0.591	1.19

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	Cu	Fe	Pb	Mg	Mn	Hg	Mo	Ni	P
01LMIV11	EFC1	invertebrate-composite	30.7	2680	0.647	1620	322	0.286	<1.1	4.44	7490
01LMIV12	EFC1	invertebrate-composite	27.3	570	0.307	1100	199	0.352	<1.12	2.9	7280
01LMIV13	EFC1	invertebrate-composite	27.3	1100	0.284	1420	190	0.29	<1.1	2.05	7130
01LMIV21	EFC2	invertebrate-composite	28.2	718	0.239	1150	167	0.439	<1.08	4.09	7920
01LMIV22	EFC2	invertebrate-composite	25.8	804	0.286	1120	189	0.416	<1.1	4.81	8000
01LMIV23	EFC2	invertebrate-composite	29	1840	0.468	1430	191	0.359	<1.09	6.92	8220
01LMIV31	EFC3	invertebrate-composite	27.5	2730	0.564	1630	549	0.336	1.54	3.21	7510
01LMIV32	EFC3	invertebrate-composite	28.3	2750	0.782	1510	581	0.294	<1.08	3.71	7530
01LMIV33	EFC3	invertebrate-composite	27	1600	0.709	1350	468	0.278	<1.11	2.63	7570
01LMIV42	BRY	invertebrate-composite	27.4	870	0.4	1620	251	0.23	<1.05	9.47	9960
01LMIV43	BRY	invertebrate-composite	28.2	758	0.196	1560	435	0.162	<1.11	11.5	9940
01LMFG20	EFC2	gill-composite	1.45	481	0.164	1140	18.4	0.724	<1.05	1.03	24000
01LMFG30	EFC3	gill-composite	1.73	244	0.207	1640	35.9	0.571	<1.02	0.806	26600

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	Cu	Fe	Pb	Mg	Mn	Hg	Mo	Ni	P
01LMFL20	EFC2	liver-composite	554	313	0.166	754	6.63	1.39	1.15	<0.546	15100
01LMFL30	EFC3	liver-composite	119	930	0.152	771	11.3	1.39	1.25	<0.522	15200
01LMFM21	EFC2	muscle-individual	2.15	16.6	0.163	1240	1.45	0.802	<1.05	<0.525	12400
01LMFM22	EFC2	muscle-individual	2.01	17.1	0.157	1260	1.39	1.27	<1.04	0.531	11700
01LMFM23	EFC2	muscle-individual	2.25	46.5	1.96	1270	1.42	0.83	<1.07	0.61	11400
01LMFM24	EFC2	muscle-individual	1.98	22.3	0.142	1260	0.981	1.29	<1.09	<0.545	11300
01LMFM25	EFC2	muscle-individual	1.39	18.3	0.117	1240	0.69	0.801	<1.08	<0.539	11500
01LMFM31	EFC3	muscle-individual	1.26	21.9	0.12	1150	0.935	1.23	<1.02	<0.508	11000
01LMFM32	EFC3	muscle-individual	1.61	32.8	0.116	1210	2.84	0.925	<0.833	0.651	12700
01LMFM33	EFC3	muscle-individual	1.59	36.4	<0.0907	1230	3.34	0.915	<0.982	0.62	12000
01LMFW11	EFC1	whole body-individual	9.43	91.2	<0.0780	1000	5.87	0.997	<0.994	1.08	21400
01LMFW12	EFC1	whole body-individual	5.98	38.5	<0.101	947	7.01	1.11	<1.00	<0.5	12700
01LMFW13	EFC1	whole body-individual	11.1	93.8	<0.102	896	4.67	1.17	<1.08	<0.541	14300
01LMFW14	EFC1	whole body-individual	9.39	54.1	<0.102	1290	8.2	0.845	<1.02	<0.51	14100

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	Cu	Fe	Pb	Mg	Mn	Hg	Mo	Ni	P
01LMFW21	EFC2	whole body-individual	4.15	197	0.0942	1140	39	0.814	<1.00	2.06	11700
01LMFW22	EFC2	whole body-individual	6.95	521	0.453	1260	21.4	0.545	<1.04	3.42	30800
01LMFW23	EFC2	whole body-individual	9.36	116	<0.0926	811	6.92	0.773	<0.991	0.519	15200
01LMFW24	EFC2	whole body-individual	19.8	82.1	<0.0787	815	2.94	1.01	<1.04	0.524	10700
01LMFW25	EFC2	whole body-individual	5.51	95.5	<0.0688	1050	11	0.679	<1.03	<0.515	16400
01LMFW31	EFC3	whole body-individual	6.51	72.5	0.165	1280	23.7	1.24	<1.07	<0.533	13500
01LMFW32	EFC3	whole body-individual	8.66	62.3	<0.0959	1000	14.1	0.984	<1.01	<0.507	17900
01LMFW33	EFC3	whole body-individual	6.14	78.7	0.121	954	17.6	0.753	<1.03	<0.515	13600
01LMFW34	EFC3	whole body-individual	6.07	49.3	0.107	820	6.35	0.89	<1.04	<0.518	16200
01LMFW41	BRY	whole body-individual	3.67	148	0.128	958	40.8	0.202	<1.02	1	14600
01LMFW42	BRY	whole body-individual	4.15	204	0.137	1030	17.7	0.557	<1.05	0.746	11200

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	K	Se	Si	Na	S	Sr	Ti	V	Zn
01LMIV11	EFC1	invertebrate-composite	7380	0.404	266	4010	5420	19.7	62.8	4.92	200
01LMIV12	EFC1	invertebrate-composite	6340	0.474	307	3250	5300	16.5	33.8	2.69	241
01LMIV13	EFC1	invertebrate-composite	6010	0.425	218	3290	5330	14	22.8	2.33	243
01LMIV21	EFC2	invertebrate-composite	6430	0.716	207	3560	5550	25.1	8.95	<1.08	292
01LMIV22	EFC2	invertebrate-composite	6680	0.713	246	3580	5370	24.8	10.7	1.12	252
01LMIV23	EFC2	invertebrate-composite	7710	0.699	238	4680	5540	36.9	25	2.6	230
01LMIV31	EFC3	invertebrate-composite	6270	0.563	205	3570	5080	28.8	111	8.66	223
01LMIV32	EFC3	invertebrate-composite	6970	0.547	462	4220	5180	33.2	76.2	4.97	225
01LMIV33	EFC3	invertebrate-composite	6380	0.564	248	3760	5350	30.4	50.1	4.83	202
01LMIV42	BRY	invertebrate-composite	7770	2.37	259	3530	5540	26.6	12.9	1.28	231
01LMIV43	BRY	invertebrate-composite	6830	2.25	285	3120	5510	27.5	13.2	1.67	301
01LMFG20	EFC2	gill-composite	7080	1.35	100	6840	7180	139	2.71	<1.05	447

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	K	Se	Si	Na	S	Sr	Ti	V	Zn
01LMFG30	EFC3	gill-composite	6530	1.45	117	6150	7160	147	4.65	<1.02	88.3
01LMFL20	EFC2	liver-composite	13100	4.75	36.5	3650	9440	0.972	<0.546	<1.09	113
01LMFL30	EFC3	liver-composite	12500	4.5	70.9	4500	9260	0.921	<0.522	<1.04	91.9
01LMFM21	EFC2	muscle-individual	17800	0.429	36.5	1070	8460	10.2	0.745	<1.05	17.4
01LMFM22	EFC2	muscle-individual	18500	0.521	30.4	1180	9240	2.73	<0.52	<1.04	15.7
01LMFM23	EFC2	muscle-individual	18200	0.59	54.2	1170	9100	2.42	1.97	<1.07	25.1
01LMFM24	EFC2	muscle-individual	18400	0.622	27.5	1220	9670	2.67	<0.545	<1.09	15.8
01LMFM25	EFC2	muscle-individual	18500	0.541	33.7	1040	8460	1.41	<0.539	<1.08	17.2
01LMFM31	EFC3	muscle-individual	16000	1.07	30.5	854	7400	2.11	<0.508	<1.02	12.8
01LMFM32	EFC3	muscle-individual	17200	0.887	<8.3	965	7590	11.9	<0.416	<0.833	16
01LMFM33	EFC3	muscle-individual	16400	0.641	58.4	1170	8390	7.5	1.25	<0.982	17.3
01LMFW11	EFC1	whole body-individual	11900	0.727	111	3250	7220	89.1	2.56	<0.994	161
01LMFW12	EFC1	whole body-individual	12100	0.687	37.7	2730	7520	30	0.709	<1.00	122
01LMFW13	EFC1	whole body-individual	12400	1.08	42.6	3020	8020	44.7	1.04	<1.08	135

Appendix 2 (cont.). Trace-element concentrations determined in aquatic invertebrates and salmonid tissues collected from lower Bryant Creek and the East Fork Carson River, Douglas County, Nevada, October, 2001.

SAMPLE #	SITE	SAMPLE TYPE	K	Se	Si	Na	S	Sr	Ti	V	Zn
01LMFW14	EFC1	whole body-individual	12200	1.01	33.6	3220	8230	29.9	0.897	<1.02	148
01LMFW21	EFC2	whole body-individual	13100	0.905	126	3010	8150	21.1	7.7	<1.00	196
01LMFW22	EFC2	whole body-individual	12500	0.807	156	3020	7840	153	12.6	1.41	147
01LMFW23	EFC2	whole body-individual	10700	0.838	106	2910	7540	51.4	2.28	<0.991	115
01LMFW24	EFC2	whole body-individual	10800	0.84	31.3	2410	6630	18.9	<0.521	<1.04	177
01LMFW25	EFC2	whole body-individual	11100	0.724	54	2570	6840	61.7	0.845	<1.03	124
01LMFW31	EFC3	whole body-individual	12200	0.862	78.7	2800	6950	34.6	3.8	<1.07	145
01LMFW32	EFC3	whole body-individual	10500	0.653	118	2490	6670	65.2	4.54	<1.01	197
01LMFW33	EFC3	whole body-individual	11400	0.925	106	2620	6860	39.6	2.52	<1.03	155
01LMFW34	EFC3	whole body-individual	10400	0.628	47.2	2250	6290	54.5	1.23	<1.04	125
01LMFW41	BRY	whole body-individual	10600	2.5	90.7	3080	7030	31.2	1.32	<1.02	68.9
01LMFW42	BRY	whole body-individual	11800	1.44	144	2110	6940	18.6	24.2	1.98	79.5