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Annual Review

CHARACTERIZATION OF ECOLOGICAL RISKS AT THE MILLTOWN RESERVOIR–CLARK FORK RIVER SEDIMENTS SUPERFUND SITE, MONTANA

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Abstract—A comprehensive field and laboratory approach to the ecological risk assessment for the Milltown Reservoir–Clark Fork River Sediments Site, a Superfund site in the Rocky Mountains of Montana, has been described in the preceding reports of this series. The risk assessment addresses concerns over the ecological impacts of upstream releases of mining wastes to fisheries of the upper Clark Fork River (CFR) and the benthic and terrestrial habitats further downstream in Milltown Reservoir. The risk characterization component of the process integrated results from a triad of information sources: (a) chemistry studies of environmental media to identify and quantify exposures of terrestrial and aquatic organisms to site-related contaminants; (b) ecological or population studies of terrestrial vegetation, birds, benthic communities, and fish; and (c) in situ and laboratory toxicity studies with terrestrial and aquatic invertebrates and plants, small mammals, amphibians, and fish exposed to contaminated surface water, sediments, wetland soils, and food sources. Trophic transfer studies were performed on waterfowl, mammals, and predatory birds using field measurement data on metals concentrations in environmental media and lower trophic food sources. Studies with sediment exposures were incorporated into the Sediment Quality Triad approach to evaluate risks to benthic ecology. Overall results of the wetland and terrestrial studies suggested that acute adverse biological effects were largely absent from the wetland; however, adverse effects to reproductive, growth, and physiological end points of various terrestrial and aquatic species were related to metals exposures in more highly contaminated depositional areas. Feeding studies with contaminated diet collected from the upper CFR indicated that trout are at high risk from elevated metals concentrations in surface water, sediment, and aquatic invertebrates. Integration of chemical analyses with toxicological and ecological evaluations of metal effects on the wetland and fishery has provided an important foundation for environmental decisions at this site.

Keywords—Ecological risk assessment Metals Wetland Clark Fork River Mining wastes

INTRODUCTION

In the Clark Fork River (CFR) basin of Montana, an ecological risk assessment was performed to determine baseline risks to the aquatic and terrestrial habitats of the upper CFR and Milltown Reservoir. This article is the last in the present series that describes the risk assessment for the Milltown Reservoir–CFR Sediments Site, a National Priority List (NPL) "Superfund" site. Detailed descriptions of the site, its history, the sources and extent of metals contamination in the upper CFR and Milltown Reservoir, ecological concerns due to metals contamination, and the approach selected for the comprehensive ecological risk assessment program at the site are presented in the first report of this series [1]. In general, the upper CFR is contaminated with waste metals from upstream releases of mine tailings to the CFR headwaters. The tailings are enriched in As, Cd, Cu, Pb, and Zn, and result from historical copper mining activities in the Butte and Anaconda areas of Montana (see [1] for area map).

In the methodology developed for assessing ecological risks at hazardous waste sites, the U.S. Environmental Protection Agency (EPA) identifies the final step as risk characterization [2,3]. In the present article, we focus on this step and present the characterization of ecological risks to terrestrial and aquatic habitats at the Milltown Reservoir–CFR site. Information for the risk characterization was generated from the risk assessment studies described in the preceding reports in this series, and in technical studies prepared for the EPA. These studies characterize the ecology of the site [4]; describe the biological evaluation of soil contamination [4] and a preliminary food chain contamination study for the terrestrial habitat of the wetland operable unit [5]; characterize sediment chemical contamination [6], sediment toxicity to

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aquatic invertebrates [7], benthic community impacts [8], and bioaccumulation of metals by sediment macroinvertebrates in both the reservoir and upper CFR operable units [9]; and describe the impacts of a diet of contaminated aquatic invertebrates collected from the upper CFR, and an artificially contaminated live diet, on early life stages of rainbow and brown trout [10–13].

APPROACH AND METHODOLOGY

Ecological risk assessments are seldom purely probabilistic in nature, in that the probability of an adverse ecological effect is difficult to quantify. Hence, the risk component of the assessment is usually defined by (a) a measured effect in the field, or (b) a qualitative or semiquantitative estimate of the likelihood of an effect based on extrapolation from field and laboratory measurements or the scientific literature. Field and laboratory measurements of impacts at the Milltown Reservoir–CFR site provided site-specific information designed to minimize the uncertainty inherent in extrapolations of impacts from other habitats, similar geographic areas, or other time periods.

Given the complex informational base generated for this site, the EPA's Risk Assessment Forum guidelines on methodology for performing ecological risk assessments were used to characterize ecological risks [3]:

Integration of exposure profiles and stressor–response relationships—This step describes contaminant measurements in environmental media and biota, stressor–response relationships, and results of ecological surveys for the terrestrial and aquatic habitats. For the Milltown Reservoir–CFR site, the stressor was considered to be the concentration of metal or dose of metal exposure [2]. The studies described in this series and elsewhere provided the exposure profiles and stressor–response relationships for the lower trophic receptors identified for the site [1]. Exposure estimates and chemical dose–response relationships for higher trophic organisms were taken from the technical risk assessment report [5].

Ecological risk summary—In this step, exposure estimates and metals concentrations or dose–response relationships were integrated to characterize risks. As the establishment of causality is often difficult in ecological risk assessments, a weight-of-evidence approach was used to characterize risks that relied on the combination of empirical observations and inferences founded in reasonable scientific judgment. As defined in the EPA guidance for evaluating ecological risks [3,14,15], unless the risk assessment can be strictly limited to comparisons with existing quality criteria, the characterization of risk should consist of such a weight-of-evidence approach. A similar approach was advocated for assessing lotic ecosystems where impacts were expected to be subtle or where ambient concentrations of contaminants might show temporal variability [16]. Both of these conditions characterize the upper CFR basin [1].

Interpretation of ecological significance—This step places the risk estimates in context of the types and extent of anticipated effects. The significance of the risk estimates for terrestrial and aquatic ecosystems of the Milltown Reservoir–CFR site was determined by the relative-ranking approach described by Duinker and Beanlands [17].

INTEGRATION OF EXPOSURE AND STRESSOR–RESPONSE PROFILES

Ambient concentrations and exposure measurements, results of laboratory and in situ toxicity tests, and ecological surveys for terrestrial and aquatic habitats are qualitatively summarized below under the headings Exposure Assessment, Biological Effects Assessment, and Ecological Assessment, respectively. Quantitative evaluations are provided in the next step.

Exposure assessment

Terrestrial/wetland habitat. Exposures of wetland and terrestrial organisms to soil and sediment-derived metals are described in reports presented in this series [4] and elsewhere [5,18]. In summary, the exposure assessment for the terrestrial habitat indicated that target metals (As, Cd, Cu, and Zn) in soils and biota (i.e., terrestrial and wetland plants, small mammals) are elevated across the site. The site was divided into 12 sample units based on the historical metals distribution and physiographic observations [1]. The distribution of metals in soils is heterogeneous, with the highest concentrations occurring at depositional areas. Although variation across the site was substantial, variation within defined sample units was much less so. Major depositional areas were identified in the problem formulation step [1] as the old river channel (ORC) and railroad slough (RRS).

Primary receptors of concern were identified as predatory birds, waterfowl, muskrat, and deer [1]. Exposure measurements generated site-specific information on contaminant levels in lower trophic biota, focusing on plants and macroinvertebrates, which was used to evaluate potential risks to primary consumers and higher trophic organisms.

Bioavailability of the metals in soil was found to be limiting and may be as low as 0.1% for small mammals [18]. Bioavailability of metals to soil invertebrates was found to vary across the wetland, and although a weak association was noted with cation exchange capacity, metals bioavailability was not significantly related to any particular soil variable [5].

Aquatic habitat. A number of studies have investigated the contamination of sediments, surface water, and biota in the CFR from upstream releases of metals [9,19–26]. Because few of the studies prior to this risk assessment program focused on Milltown Reservoir, intensive studies were performed to assess exposures to reservoir sediment metals and the factors governing those exposures. Studies on both the reservoir and upper CFR, including sample station locations, are described in the preceding reports of this series [6–9]. Trout exposures were studied in the upper CFR operable unit [10–12], whereas aquatic vegetation and amphibian studies were performed in the reservoir operable unit only [4].

In summary, the exposure assessment for the aquatic habitat concluded that sediment concentrations of metals are greatly elevated in the CFR headwaters (e.g., Silver Bow Creek), near the historical upstream sources of mining wastes. A decreasing trend in metals concentrations downriver from these sources to segments of the river beyond Milltown Reservoir is apparent [19,20,23,24]. An anomaly in the downriver trend is noticeable in elevated sediment concentrations of metals in depositional areas of Milltown Reservoir, with total Cu concentration as high as 1,200 $\mu g/g$ [5], as compared to more typical concentrations below 500 $\mu g/g$ found at immediate upstream and downstream stations [1].

Acid-volatile sulfide (AVS) was found to be a major factor governing concentrations of dissolved Cu, Cd, and Zn in sediment porewaters, although other unidentified factors likely play a role [6]. No relation was noted between dissolved pore-water metals and total organic carbon. Variabilities of simultaneously extracted metals (SEM) Cu, SEM Zn, and AVS concentrations were high among replicate cores sampled within a station [6], suggesting that biological responses in sediments could be highly localized.

Sediments stations CF-01, MR-11, and MR-19 [1] had greater percentages of oxide-bound Cu, Cd, and Zn than other forms of the metals, compared to the other sediment stations, and AVS was relatively low [6]. These findings are suggestive of oxidizing conditions at these stations, and are not unexpected given the shallow water or fast flow characteristic of these stations.

Ammonia was elevated in pore-water samples of stations CF-01, MR-01, MR-07, and MR-17; and PCBs were elevated in station MR-01 [6]. Elevated ammonia concentrations at stations MR-07 and MR-17 may be related to their locations just downstream of heavily grazed wetland soils [5].

Concentrations of metals are elevated in sediment macroinvertebrates collected from depositional areas of the reservoir and riffle and depositional areas of the upper CFR [9,23,25,27]. The pattern of metal accumulation between laboratory-exposed *Hyalella azteca* and field-collected animals was similar [9]. These findings indicate that sediment is a significant source of metals to invertebrates in the CFR.

A diet of invertebrates collected from sediments from the upper CFR was found to increase body burden of metals in rainbow trout [10,11] to levels observed in trout collected from the reservoir and the CFR [5,25]. Body burdens of metals are elevated in trout from both the reservoir [28,29] and the upper CFR [25], with metals concentrations in upper CFR trout collected near Warm Springs Ponds greater than concentrations in trout from Turah Bridge upstream of Milltown Reservoir [30].

Biological effects assessment

Terrestrial/wetland habitat. Site-specific biological effects for terrestrial macroinvertebrates and plants were investigated using both laboratory and field methods [4]. In addition, toxicology information on higher trophic organisms was compiled from the literature.

In summary, review of the literature on the receptors of concern indicated that Cu and Zn were potentially the most toxic metal contaminants in the wetland. Exposures to these metals subsequently showed the strongest correlation with biological test results [4]. However, no overt toxicity (i.e., acute toxicity or overwhelming responses of chronic end points) was observed in any laboratory or field test organisms.

Subtle effects were observed in toxicity tests of a variety of terrestrial/wetland species, including laboratory and field earthworm tests [4], the seed germination test [31], and root elongation test [5]. Results of sublethal toxicity tests with earthworms and terrestrial vegetation generally were consistent within sample units [4]; however, results varied highly among sample units. Minor variations among stations within a unit were attributable to variations in metal concentrations. Because of limitations in the tests performed within each unit (e.g., limited replication and low sample size), the low variation in results among the different test types could not be statistically confirmed.

Aquatic habitat. Sediment toxicity tests evaluated exposures of a wide variety of invertebrate and plant species as well as rainbow trout [7,10,32]. Amphibian toxicity tests were performed on surface water samples during preliminary studies of the reservoir [33].

In summary, the biological effects assessment found that whole-sediment samples were toxic to aquatic species on the order of *H. azteca* > *Chironomus riparius* > rainbow trout > *Daphnia magna* [8]. Whole-sediment samples were likely less toxic to rainbow trout or *Daphnia* because of their more limited exposures. Relative sensitivity of the toxicity end points measured in the *H. azteca* test was length > sexual maturation > survival. Pore-water toxicity test results were inconclusive because of changes in pore-water characteristics during holding [8].

Amphipod toxicity tests identified sediment from six of seven reservoir stations and two of six CFR stations as toxic (MR-01, MR-02, MR-07, MR-11, MR-17, MR-19, CF-01, CF-03) [8]. Toxicity of sediments in laboratory tests to *H. azteca* was evaluated by a no-effects concentration (NEC) approach, with NEC values assigned to the treatment that did not significantly reduce amphipod body length or maturation [8]. Reduced amphipod body lengths were correlated to SEM Cu concentrations and SEM Cu normalized to AVS concentrations (SEM Cu/AVS). Similar stations exceeded the sum SEM/AVS and Zn SEM NECs. This correspondence was not unexpected since Cu and Zn accounted for >95% of the extractable metals on a molar basis [6].

Effects of exposures of aquatic plants to split reservoir sediments from the benthic toxicity tests showed no effects on survival or root growth, either in situ or in the laboratory [4,32]. The observed effects to *Potamogeton pectinatus*, *Hydrilla verticillata*, and indigenous *Elodea* species were stimulation of peroxidase activity and shoot growth following exposures to sediments of stations MR-07 and MR-17. Of the stations observed to affect benthic macroinvertebrates in toxicity tests, station MR-11 was not evaluated in aquatic plant tests, and no effect was observed with aquatic plants from exposures to sediments from station MR-19.

Limited toxicity bioassays on amphibians using surface waters near unit 4 demonstrated no acute effects on survival or growth [33]. Morphological effects were noted in specimens exposed to samples with elevated metals concentrations.

Feeding studies using invertebrates collected from various segments of the upper CFR demonstrated adverse effects to early-life-stage rainbow trout with increasing concentrations of metals in food sources [10–12]. Rainbow trout were more sensitive than brown trout to metals-related effects. Effects included reduced survival, feeding activity, and growth; constipation; gut impaction; cell membrane damage; decrease in digestive enzyme production; and sloughing of intestinal cells.

Trout collected from the upper CFR near Warm Springs Ponds demonstrated a higher incidence of histopathological liver damage, copper inclusions in hepatocyte cytoplasm, and elevated lipid peroxidation products [30]. In contrast to the feeding studies with CFR invertebrates, rainbow trout fed a diet of brine shrimp enriched with As, Cu, Cd, Pb, and Zn at concentrations similar to those in CFR invertebrates were unaffected in 60-d flow-through exposures [13].

Ecological assessment

Terrestrial habitat. Information on the ecological component for the terrestrial habitat at the reservoir operable unit comes from the wetland delineation study and wildlife survey, both performed under the direction of the U.S. Fish and Wildlife Service (FWS) [4,27,34]. The wetland delineation study provided quantitative data on plant dominance at stations within the terrestrial sampling units, including those units with the highest metals contamination in the wetland. Although only certain components were considered statistically rigorous, the wildlife survey provided an indication of whether ecological impacts were observable at the site [4,5].

In summary, terrestrial and wetland wildlife were found to be diverse and apparently healthy. No overt indications of adverse effects of metals contamination to waterfowl, amphibians, earthworms, or vegetative cover were observable. Species compositions of migratory bird, small mammal, and terrestrial invertebrates were typical of riparian sites in western Montana. Although game mammals were not inventoried, visual observations suggested that deer and predatory bird populations were apparently healthy.

Canada geese breeding was observed to be successful in the reservoir wetland. A high number of breeding songbirds were observed with species diversity quantitatively similar (Shannon–Weiner index 2.8) to uncontaminated wildlife refuge wetlands in nearby watersheds (Shannon–Weiner index 2.3).

Although overt ecological impacts in the terrestrial habitat at the site were not observable, soil microbial and invertebrate communities were not evaluated quantitatively, partly due to unavailable standardized methods at the time of the studies. In addition, focused ecological studies on vegetation in areas where subtle biological effects were expressed in toxicity tests were also not performed.

Aquatic habitat. Ecological studies for the aquatic environment consisted of the benthic community structure analysis performed by the FWS [8], observations of aquatic plants and amphibians from the wildlife survey [4,27,34], and studies of fish populations performed on the upper CFR and the reservoir [30,35]. Fish population studies were not performed as part of the risk assessment program described in the present series of articles; available information is summarized below.

In summary, results of aquatic ecological studies on the upper CFR and Milltown Reservoir have shown that benthic organisms are present at all depositional stations in the reservoir and at depositional and riffle areas of the upper CFR [8,10]. However, total abundance of organisms in the depositional samples did not follow a consistent spatial pattern in either operable unit relative to concentrations of metals.

Oligochaeta and Chironomidae accounted for over 96% of the benthic invertebrate community abundance at all stations in the reservoir, and 70 to 99% of abundance at CFR stations [8]. The Oligochaeta community in both operable units was dominated by pollution-tolerant species; no intolerant species of Oligochaeta were collected at any stations. The most tolerant species occurred at Stations MR-11 and MR-19. Higher numbers of Chironomidae genera were present at stations with higher concentrations of metals in sediment samples (e.g., MR-11, MR-19, CF-01, CF-03). The Chironomidae community consisted primarily of metals-tolerant species [8].

Limited visual observations of aquatic plant communities suggested relatively high biomass and species diversity of emergent plants in the reservoir [4,34]. Limited visual observations of amphibians suggested good species abundance [33].

Overall populations of fish in the CFR are considered to be below the carrying capacity of the river [36], and are only partially recovered from historical impacted levels [37]. Review by the State of Montana [36] concluded that a major factor limiting fish populations in the upper CFR is limited use of available spawning habitat. Both recruitment and mortality of trout in the upper CFR have been suggested to be due to elevated metals concentrations [38,39] and limited habitat [40]. Elevated metals concentrations have also been implicated in fish impacts in the Blackfoot River [41]. Brown trout are the dominant species in the more contaminated upper CFR, whereas rainbow trout are predominant in lesser contaminated downstream areas [36].

Numerous fish kills in the upper CFR have been documented over the past decades [36]. Fish kills have been associated with summer thunderstorms and are believed to be the result of metals entering the river following rainfall on streamside mine tailings. In addition to low pH, physical mixing of the benthic substrate can result in mobilization of both particulate and dissolved forms of metals. Metals have been recorded at their highest concentrations in CFR waters during the early spring runoff [1,26], although there is no documentation of fish kills during that time. As recently as summer 1989, some 5,000 fish were lost in the upper CFR from acidic runoff from tailings wastes [42].

In general, fish populations in the upper CFR increase immediately below the confluence with tributary rivers [36]. Trout density in the upper CFR near Rock Creek averages about 300 per mile [43]. This is considered sparse when compared to populations in tributaries where 2,500 per mile are reported for some sections of the Blackfoot River, 1,500 per mile for Warm Springs Creek, Gold Creek, and Rock Creek, and 1,000 per mile for Flint Creek and the Little Blackfoot River [44]. A comparison of upper CFR fish populations with those of the Yellowstone River suggested that the CFR above Milltown Dam should support about 900 trout per mile [43]. Studies conducted by ARCO in Milltown Reservoir in 1991 [5,28] showed that the fish population is apparently healthy, but is dominated by large-scale sucker, longnose sucker, and northern squawfish.

Montana Power Company [43] estimated that Milltown Dam is responsible for 50% of the reduced trout population in the CFR from Rock Creek to 40 miles downstream of the dam, including local portions of the Blackfoot River. Primary impacts were identified as year-round blockage of access to upstream tributaries for spawning and rearing, and reduction of the riverine habitat with resultant increased predation of juvenile trout by squawfish residing in the reservoir. In addition to the blockage of migration and recruitment of fish by the dam, other habitat-related factors, such as few riffle areas and general lack of bank cover and woody debris, many contribute to low trout populations in Milltown Reservoir. Low trout populations downstream of the reservoir have been suggested to be related to chronic effects of releases of contaminated water and sediments from Milltown Reservoir [38,45].

RISK SUMMARY

Terrestrial/wetland habitat

Risks to lower trophic components of the wetland ecosystem were assessed by integration of the results of the site-specific exposure, biological effects, and ecological assessments that were summarized above. Higher trophic organisms were evaluated through a food-chain model.

Soil macroinvertebrates. Sublethal effects to earthworms during exposures to soil metals were observable with both laboratory and field exposures [4]. Sublethal effects were limited to a varicty of malformations, including segmented swelling, segmental constriction, coiling, rigidity, flaccidity, cutaneous lesions, and behavioral patterns.

The data on exposure and toxicity to earthworms were the most complete relative to other studies of the terrestrial habitat. These data were therefore analyzed by a variety of statistical measures to evaluate potential relationships among the metals concentrations, the observed biological effects, and the associated units [46]. In the absence of widespread acute effects, only the sublethal effects noted in earthworms were appropriate for statistical analysis.

Although total and extractable Cu concentrations showed the strongest correlations with morphological effects in earthworms [4], clear concentration–response relationships were not observed. The relationships between earthworm exposures and responses were therefore evaluated by a ranking procedure. Summaries of the average soil concentration and earthworm response for each sampling unit are presented in Table 1. The top-ranked units based on morphological endpoint measurements were units 2, 8, 7, and 4. The top units ranked by total Cu concentration were units 7, 6, 8, and 2. Qualitative comparison of the ranking results suggested a fairly good correlation by unit of sublethal effects and total metals or total Cu concentrations, with the exceptions of units 4 and 6.

Figure 1 shows the graphical ranking of sublethal effects in earthworms with total Cu concentrations in soil for all stations sampled during the preliminary and Tier I studies. Control responses with on-site reference soils ranged from 0 to 6.67%. Results of the ranking suggest an association of ele-

Table 1. Summary results of terrestrial bioassays

Sample unit	Soils[a] (μg/g dry wt)			Malformations in earthworm bioassay[b] (%)		Rhizosphere water[c] (μg/ml)		Soil eluates[d] (μg/ml)			Length in root elongation bioassay (% of control)	
	Cu	Cd	Zn	On-site	Lab	Cd	Zn	Cu	Cd	Zn	Rhizosphere water	Soil eluates[d]
Control				1	1						(35.5 mm)	
2	734	9.7	3,580	12	15	NT	NT	0.130	<0.001	0.400	NT	43
3	529	5.5	1,131	3	11	NT	NT	0.182	<0.001	0.435	NT	45
4	244	5.3	711	5	33	NT	NT	0.109	<0.001	0.220	NT	>75
6	870	12.1	2,476	0	10	NT	NT	0.082	<0.001	0.280	NT	>75
7	975	13.4	3,918	8	4	0.015	4.2	<0.036	0.007	1.700	42	77
8	761	4.8	1,266	23	9	0.048	34.8	<0.036	0.005	0.933	24	73
9	595	6.8	1,932	1	7	<0.001	1.3	<0.036	<0.001	0.167	90	77
10	654	9.7	2,400	1	3	0.002	0.6	<0.036	<0.001	0.200	81	82
11	585	4.5	1,390	1	0	<0.001	0.5	<0.036	<0.001	0.500	98	84
12	470	5.3	2,049	1	0	<0.002	0.8	<0.036	<0.001	0.081	98	76

NT, not tested.
[a]Values are mean of 3 to 11 samples [4,5].
[b]Morphological changes included segmented swelling, segmental constriction, coiling, rigidity, flaccidity, cutaneous lesions, or behavioral patterns [4].
[c]Values are mean of 2 to 3 samples. All Cu values were less than detection limit (0.03 μg/ml) [46].
[d]Values are mean of 3 to 4 samples. Units 2 to 6 were tested during preliminary studies [4,31]; units 7 to 12 during Tier I studies [46].

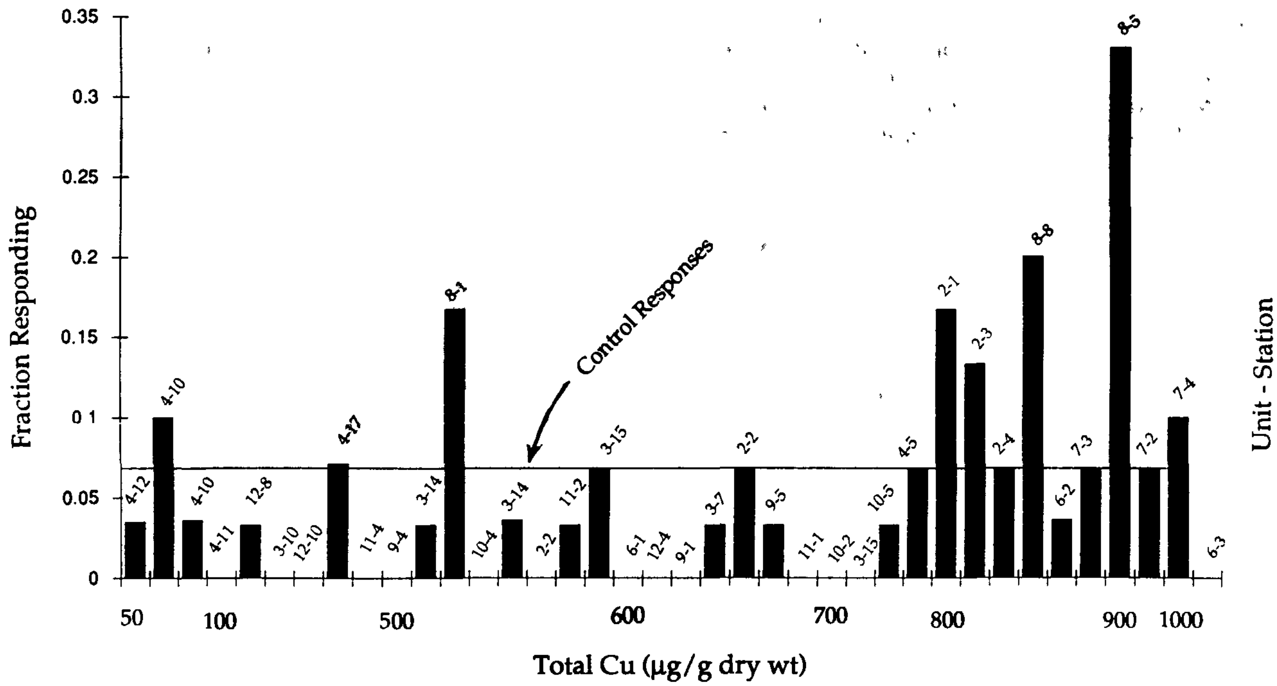

Fig. 1. In situ earthworm responses vs. total Cu concentrations in soils. Observed responses consisted of changes in behavioral patterns and physical malformations, including swelling, segmental constriction, coiling, rigidity, flaccidity, and cutaneous lesions. "Unit-station" refers to the wetland sampling unit described in [1] and the individual sampling station within that unit.

vated earthworm responses with soil Cu concentrations exceeding 800 μg/g in units 2, 7, and 8.

To understand the factors that govern bioavailability of metals in Milltown Reservoir soils, further statistical analyses compared concentration–response relationships with physical and chemical characteristics of the soils. Regression analyses and ANOVA tests consistently demonstrated a lack of correlation between sublethal effects in earthworms and cation exchange capacity (CEC), percent organic material, and percent clay of soils [4,5]. Normalization of metal concentrations to these variables failed to show correlations to sublethal effects that were any stronger than with total Cu alone. Despite these poor correlations, the marked differences in soil characteristics between units 4 and 6, for which biological responses differed greatly, suggest that unexamined properties of the soils of these units likely govern metals bioavailability, and hence biological responses in wetland soils.

Terrestrial vegetation. The root-zone study at the wetland operable unit demonstrated subtle biological effects from exposure to rhizosphere water and soil eluates in certain sampling units [4,5]. Acute effects were not observed, but significant inhibition of root growth was observed in multiple species with root-zone water from units 7 and 8 when compared to control samples and to samples from units 9, 10, 11, and 12 (Table 1). Because of the preliminary nature of these studies, data on soil characteristics from these stations were unavailable to evaluate potential factors governing bioavailability of metals in the rooting zone.

Graphical presentation of the individual sample results in Figure 2 suggests possible threshold ranges for inhibitory effects on root growth at concentrations of Cd at 3 to 4 μg/L and of Zn at 1,500 to 9,000 μg/L in rooting-zone water. Amounts that exceeded these ranges were associated with root-growth inhibition in samples from units 7 and 8. In preliminary studies, root elongation also was inhibited in plants exposed to eluates of soil from units 2 and 3 (Table 1). In general, the results depicted in Table 1 suggest that the higher concentrations of metals in soils and rooting-zone water from units 2, 3, 7 and 8 are related to root-growth inhibition under laboratory exposures.

Risks to primary consumers and higher trophic organisms. Evaluation of risks to primary consumers and higher trophic organisms in the wetland habitat was performed by a food-chain evaluation. Exposures, expressed as daily intake, were compared to dose–response information (i.e., no-observed-adverse-effects levels, NOAELs, in mg/kg-d) from the scientific literature [3,47]. The lack of toxicity information on most metals for many of the exposed species necessitated the use of dose–response data from rodents as surrogate toxicity criteria. Exposures were modeled from the results of field measurements in vegetation and lower trophic biota as well as sediments, soils, and water [5].

Because the limited data set for metals concentrations in vegetation and invertebrates at the site did not show correlations among concentrations and geographical locations on a unit basis [5], site-wide average concentrations of metals in biota were used to model their trophic transfer. Receptors were assumed to be exposed to site-wide contaminants on a

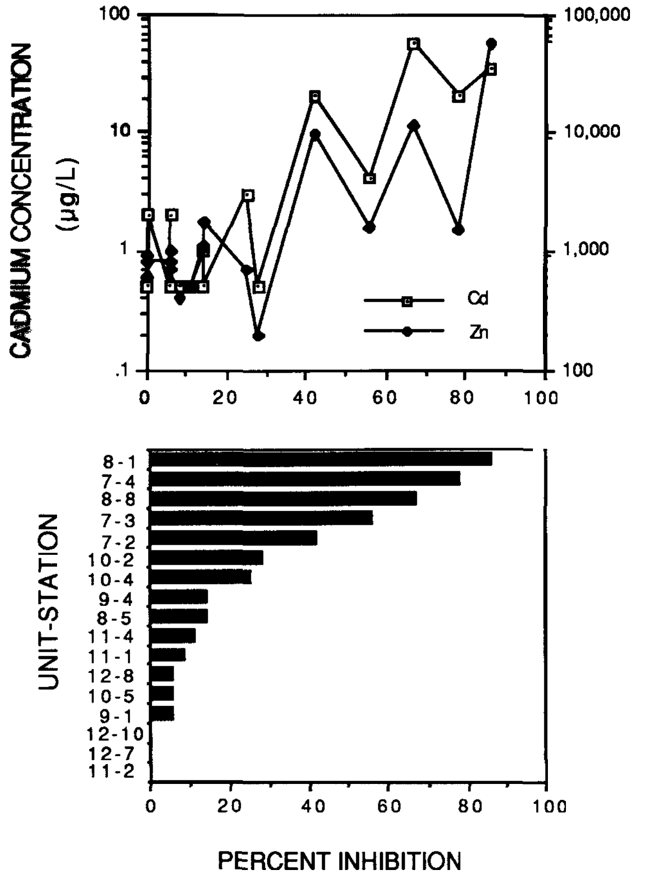

Fig. 2. Relationship between metal concentrations in root-zone water and percent inhibition of root growth. Top panel, Cd and Zn concentrations in root-zone water from each sample station; bottom panel, response per sample station.

random basis. Given the limited habitat in upland areas of the site and the presence of the dam at the downstream end, it was assumed that all exposures, except for migrating species, occurred at the site. The receptors of concern are listed in the first article of this series [1].

Results of the food-chain analysis are taken from the risk assessment technical document [5]. Site-specific data on body burden measurements of As, Cd, Cu, Pb, and Zn in the meadow vole (*Microtus pennsylvanicus*) and deer mouse (*Peromyscus maniculatus*) suggested that soil metals bioavailability was limited at <0.1% at the wetland [18]. A conservative estimate of 10% was assumed for soil/sediment metals absorption by deer, waterfowl, and predatory birds.

For all small mammals, regardless of modeled territory size, the highest estimated daily doses for As, Cd, Cu, and Zn were below the surrogate toxicity criteria [5]. However, for muskrat (*Ondatra zibethicus*), consumption of aquatic vegetation from the depositional areas of the wetland (i.e., RRS or ORC in [1]), where metals concentrations were two- to tenfold greater than site-wide concentrations, the calculated Pb intake rate (0.23 mg/kg-d) exceeded the toxicity criterion. This was largely influenced by the preference of muskrat for cattails as the primary food source, which had elevated tissue concentrations of Pb in these areas.

Intake rates of metals calculated for waterfowl listed in [1] were below surrogate toxicity criteria [5]. Similarly, intake rates of metals calculated for deer (*Odocoileus hemionus*), assuming partial forage time at the wetland, did not exceed surrogate toxicity criteria [5].

Summary of terrestrial risk characterization. A relative-ranking scheme [48] is used to summarize and integrate exposures and biological effects for the terrestrial habitat in Table 2. In this procedure, where consistent statistical differences in biological effects for the sample stations within each of the 12 sample units are observed, the unit is graded as a plus (+), inconsistent but positive responses as a plus/minus (+/−), and negative responses as a minus (−). (Unit 4 was scored as [++] for the earthworm laboratory test because of the exceptionally high responses relative to other samples.) Units are then ranked by the sum of ranks for the biological responses and by the concentrations of total Cu. For this ranking, a rating of (++) depicts the highest-ranking units, (+) the medium-ranked units, and (−) the lowest-ranked units. This comparison demonstrates the apparent association between total Cu concentrations and a variety of biological effects across the site, with units 2, 7, and 8 standing out as having highest rankings for both chemistry and biological responses. For units 4 and 6, normalizing for soil

Table 2. Summary of terrestrial studies at Milltown Reservoir

	Sample unit[a]									
	7	6	8	2	10	9	11	3	12	4
Biological test[b]										
Earthworm — Field	+	−	+	+	−	−	−	+/−	−	+/−
— Lab	+/−	+	+	+	+/−	+/−	−	+	−	++
Root elongation	+		+	+	+/−	−	−	+	−	−
Seed germination				+						+
Biomass	+			+						
Sum biological[c]	++	+	++	++	+	+	−	+	−	++
Cu concentration (μg/g)[d]	975	870	761	734	654	595	585	529	470	244
Sum chemistry[e]	++	++	++	++	+	+	+	+	−	−

Conclusions: Units 2, 7, 8—Biological effects likely associated with metal contamination; units include major depositional areas of wetland.
Units 3, 9, 10—Moderate biological responses may be related to low metals contamination.
Unit 4—Biological effects likely due to other than metals.
Units 6—Inconsistent results in biological tests. Likely low metals bioavailability; however, poor association was observed between biological effects and metals concentrations normalized to cation exchange capacity, organic material, or percent clay [4].

[a]Units are rank-ordered by concentration of total Cu.
[b]Consistent statistical differences in biological effects for the various sample stations within each unit are graded as plus (+), inconsistent but positive responses as plus/minus (+/−), and negative responses as minus (−). Unit 4 was scored (++) for the earthworm laboratory test because of the exceptionally high responses relative to other samples [4,46].
[c]Relative ranking of overall biological test results. A value of (++) was assigned to the highest ranking units, (+) for medium ranked units, and (−) for low ranked units.
[d]Mean concentration of total Cu in soil samples [4,5].
[e]Relative ranking of concentrations of Cu, Zn, and Cd in soils [4,5]; concentrations above 500 mg/kg were scored (+); concentrations above 700 μg/g were scored (++).

physical and chemical characteristics could not readily explain the observed inconsistencies between chemistry and biological response rankings.

Results of this comparison are depicted geographically on a map of the wetland sampling units in Figure 3. Concentrations of total Cu and the sum of biological responses are shaded for moderate (+) and high (++) values from Table 2 (i.e., + is shaded with single lines and ++ with double lines, with the chemistry ranking depicted by vertical lines and the biological ranking by horizontal lines; the triangular representations are described below for sediment ranking). The graphical depiction in Figure 3 allows a better feel for the spatial correlation of biological responses with metals concentrations in the reservoir wetland soils. This type of presentation also more clearly demonstrates the higher concentrations of Cu and greater biological effects in sediments that have accumulated directly behind the dam in the old river channel (ORC) (i.e., unit 2) and behind the railway berm (i.e., units 7 and 8).

The food-chain study indicated that minimal risks to the health of primary consumers and higher trophic organisms are expected from the metals present in the wetland soils, vegetation, and lower trophic biota at the site. Any potential risks predicted from the site-specific toxicity tests do not appear to be expressed in ecological impacts, as suggested by the results of the wildlife survey and vegetation studies during the wetland delineation. Thus, although potential risks to the health of individual ecological receptors in the wetland habitats due to elevated metals concentrations may be suggested by toxicity studies, risks to wetland populations and communities are likely minimal. The likelihood of limited metals bioavailability at the site, as suggested by results of small mammal studies [18] and soil chemistry studies described above, further supports lower ecological effects than predicted by soil contaminant data alone.

In summary, integration of information from the exposure, biological effects, and ecological assessments suggests that the soils of the ORC near the interstate highway and the depositional area just upstream of the abandoned railroad berm pose the greatest risk for biological effects to terrestrial receptors.

Aquatic habitat

Sediment benthos. Sediment benthic data were evaluated by Canfield et al. [8] by a ranking procedure, using data from both the Milltown Reservoir and CFR operable units. The ranking analysis produced a scaled value between 1 and 100 for the triad of sediment chemistry, toxicity, and benthic community structure [49]. The criterion for elevated chemistry was a concentration of Cu SEM or Cu SEM/AVS in whole sediment exceeding the respective NEC. As shown in Table 3, this criterion was met by stations CF-01, CF-03, MR-07, MR-11, and MR-19. The criterion for elevated toxicity was a significant decrease in either amphipod length or sexual maturation relative to the control, and was met by the

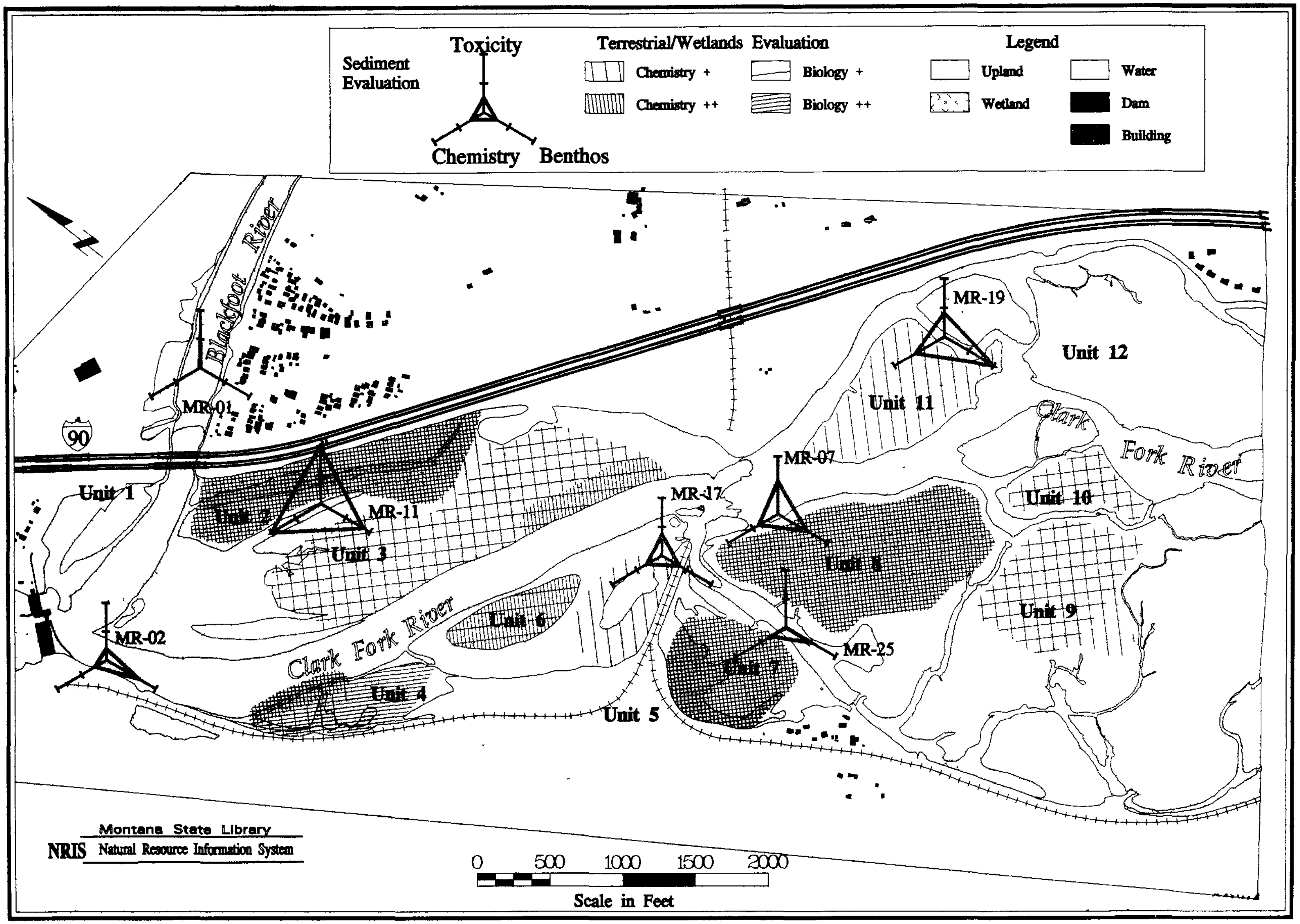

Fig. 3. Cartographic ranking of soil and sediment responses in the Milltown Reservoir operable unit. The sum of chemistry ranking (Cu concentration) is depicted by vertical lines and the sum of biological ranking by horizontal lines for the various units across the wetland; moderate rankings (+) are shaded with single lines and high rankings (++) with double lines. The rankings are taken from Table 2. The Sediment Quality Triad ranking results are depicted as triangles located at the sediment stations in the wetland, with relative values for chemistry (C), toxicity (T), and benthos responses (B) along the three axes of the triangle. Rankings are taken from Table 4.

Table 3. Summary of Sediment Quality Triad data

	Total Cu (μg/g)	Pore-water dissolved Cu (μg/L)	Pore-water dissolved Zn (μg/L)	SEM Cu (μg/g)	Cu SEM/AVS[a]	Sum SEM/AVS[b]	Number of Chironomidae genera
Milltown Reservoir							
MR-01 (reference)	30	2	<10	11	0.28	0.89	3
MR-02	266	4	11	141	0.15	0.68	12
MR-07	411	10	29	233	0.37[c]	1.46	7
MR-11	878[c]	90[c]	187[c]	607[c]	3.89[c]	24.3[c]	15[d]
MR-17	459	15	51	185	0.13	0.57	8
MR-19	364	102[c]	183[c]	354[c]	3.72[c]	11.8[c]	16[d]
MR-25	410	15	135	178	0.22	1.54	11
Clark Fork River							
CF-01	7,820[c]	79[c]	2,630[c]	6,971[c]	423[c]	960[c]	14[d]
CF-02	583	36	166	325	0.27	0.85	9
CF-03	480	16	40	287	0.88[c]	2.15[c]	16[d]
CF-04	478	9	28	251	0.30	0.99	6
CF-05	128	9	20	77	0.15	0.75	11
CF-06 (reference)	16	2	2	12	0.03	0.07	7
No-effect concentration (NEC)							
Amphipod length	583	36	166	325	0.30	1.54	NA
Amphipod maturation	583	102	183	354	3.72	11.8	NA
Sediment-quality criteria[e]							
AET	390						
ER-L	34						
ER-M	270						
Water-quality criteria							
Acute	NA	18	120				
Chronic	NA	12	110				

Data from Brumbaugh et al. [6], Kemble et al. [7], Canfield et al. [8].
NA, not applicable.
[a]Molar concentration of SEM Cu divided by molar concentration of AVS.
[b]Sum molar concentration of SEM Cd, Cu, Hg, Ni, Pb, and Zn divided by molar concentration of AVS.
[c]Exceeded NEC for amphipod length [7].
[d]Significantly different from reference stations for number of genera [8].
[e]Sediment-quality criteria from Barrick et al. [54] and Long et al. [55].

same stations (Table 3). The criterion for impacted benthos was a Chironomidae genera number that was significantly different from both the MR-01 and CF-06 reference stations, and was met by stations CF-01, CF-03, MR-11, and MR-19 (Table 3).

Increased numbers of Chironomidae genera have been reported in response to metal or organic contamination [50,51]. Canfield et al. [8] discuss how increased richness of Chironomidae genera may be explained by the intermittent disturbance hypothesis [52] and the life-history strategies in Chironomidae. For example, Connell [52] contends that continually changing conditions in the local environment may keep a community at nonequilibrium conditions, thereby allowing an increased genera richness as dominant species become less competitive. Metals contamination of sediment and surface water may be the disturbance resulting in community changes in Milltown Reservoir and CFR depositional areas. Elevated metals concentrations in surface waters may exacerbate the nonequilibrium conditions. The areal dispersion method of colonizing areas may explain why Chironomidae and not other taxa are the first to recolonize disturbed areas in the CFR [8].

Results of the ranking procedure for elevated chemistry, toxicity, and benthic impacts are integrated in Table 4, and stations are identified for likely metals-induced degradation of sediment benthos. Stations CF-01 and CF-03 in the upper CFR, and stations MR-11 and MR-19 in the reservoir showed evidence of metal-induced degradation. Samples from the remaining stations in the upper CFR and station MR-25 in the reservoir were classified as no evidence of metal-induced degradation. None of the stations with low chemistry and low toxicity were classified as having impacted benthos. Toxicity in samples from stations MR-01, MR-02, and MR-17 may be related to something other than metals. Because of the elevated chemistry and toxicity in the absence of an impacted benthos, metals may be stressing the system at station MR-07.

Amounts in excess of the toxicity criterion for *H. azteca* found with samples from stations CF-01, MR-01, MR-07, and MR-17 can be associated with total ammonia (Table 3). For stations MR-07 and MR-17, elevated ammonia concentrations may be due to proximity to grazing of wetland areas immediately upstream [4]. In addition to ammonia and PCBs [8], sediments from station MR-01 may also have other unmeasured contaminants because of its location downstream from known chemical dumping areas [5].

Table 4. Comparison of Sediment Quality Triad analysis with copper concentrations

Station	SEM Cu/AVS[a]	Sum SEM/AVS[b]	Pore-water Cu (μg/L)	SQT[c] C/T/B	Conclusion
CF-01	423	960	80	+/+/+	Evidence of metal-induced degradation
MR-11	3.89	24.3	90	+/+/+	
MR-19	3.72	11.8	102	+/+/+	
CF-03	0.88	2.15	16	+/+/+	
MR-07	0.37	1.46	10	+/+/−	Metals may be stressing the system
MR-01	0.28	0.89	2	−/+/−	Other chemicals or conditions exist with potential to cause degradation
MR-02	0.15	0.68	4	−/+/−	
MR-17	0.13	0.57	15	−/+/−	
MR-25	0.22	1.54	15	−/−/−	No evidence of metal-induced degradation
CF-02	0.27	0.85	36	−/−/−	
CF-04	0.30	0.99	9	−/−/−	
CF-05	0.15	0.75	9	−/−/−	
CF-06	0.03	0.07	2	−/−/−	

Adapted from Canfield et al. [8]; chemistry data from Brumbaugh et al. [6] and Kemble et al. [7].
[a]SEM Cu/AVS, simultaneously extracted Cu normalized to AVS concentrations.
[b]Sum SEM/AVS, sum of simultaneously extracted metals concentrations normalized to AVS concentrations.
[c]A plus for chemistry (C) indicates a concentration of Cu that exceeded either the Cu SEM NEC or the Cu SEM/AVS NEC. A plus for toxicity (T) indicates a significant decrease relative to the control in either amphipod length or sexual maturation. A plus for benthos (B) indicates significant difference from the reference stations [8].

The scaled ranking for the chemistry, toxicity, and benthos variables are plotted on maps of the reservoir operable unit as triaxial graphs in Figure 3, as per Chapman et al. [53]. Plots of triaxial graphs for the CFR operable unit can be found in Canfield et al. [8]. The triaxial graphs help to visualize both geographic trends and the magnitude in the differences among station responses. A symmetrical triangle indicates similar chemistry, toxicity, and benthos responses; a larger triangle indicates a more severely degraded station. As an example of the utility of this type of presentation, stations CF-01 [8] and MR-11 (Fig. 3) have large and relatively symmetrical triangles, representative of the elevated chemistry, toxicity, and impacted benthos (i.e., evidence of metal-induced degradation). Triangles for stations CF-03 [8] and MR-19 are also large, though less symmetrical.

Comparison with available sediment criteria. Freshwater sediment-quality criteria have not been established to the degree of marine and estuarine criteria. Concentrations of Cu in whole sediments and the Cu NEC values for stations in Milltown Reservoir and the upper CFR are compared with available sediment criteria in Table 3. Apparent-effects threshold (AET) values were developed for estuarine sediments, and are defined as the maximum concentration of a chemical in a sediment that does not produce a significant effect in the most sensitive of four end points [54]. Effect range-median (ER-M) values are concentrations of a chemical in sediment (pooled values of freshwater, marine, and estuarine) above which effects are observed with greater than 84% of the data [55]. Effect range-low (ER-L) values are lower concentrations where effects have been observed with about 10% of the data [55].

The NEC for SEM Cu of 325 μg/g sediment for amphipod length is similar to the AET value of 390 μg/g for total Cu, and somewhat greater than the ER-M value of 270 μg/g for total Cu. Although the proposed sediment criteria might not be directly applicable to the CFR, this comparison supports the likelihood of a metals-associated impact on at least those stations exceeding the SEM Cu NEC (i.e., stations CF-01, MR-11, and MR-19). The NEC for SEM Zn for amphipod effects was well above the ER-M value of 410 μg/g, whereas the NECs for SEM As, Cd, and Pb were similar to the ER-M values [7]. The elevated concentrations of these other metals in the sediments of stations CF-01, CF-03, MR-11, and MR-19 may have contributed to the observed toxicity.

Found in sediments from station MR-01 were PCBs at 730 μg/kg sediment [6], which greatly exceeded the ER-M value of 180 μg/kg sediment [55]. This finding suggests that the toxicity of sediments from MR-01 may have been due to the presence of PCBs or other unmeasured organic contaminants co-occurring with PCBs. The toxicity observed with sediments from MR-02, however, could not be directly related to the presence of any chemicals, including metals, PCBs, or PAHs [7], but may be related to unknown contamination resulting from Blackfoot River input [5].

Ambient water-quality criteria for Cu and Zn in freshwater are also provided in Table 3. Acute and/or chronic water-quality criteria were exceeded with pore-water dissolved Cu and Zn concentrations in stations MR-11, MR-17, MR-19, MR-25, CF-01, CF-02, and CF-03. The highest pore-water concentrations of Cu and Zn generally were observed at stations with highest SEM Cu concentrations. Pore-water dissolved Cu and Zn concentrations in some stations (e.g., stations MR-11, MR-19, and CF-01) exceeded both the respective NECs for amphipod length and ambient water-quality criteria (Table 3). However, dissolved Cu concentrations in pore water of a number of stations exceeded the chronic ambient water-quality criterion, yet were not associated with amphipod toxicity (e.g., stations MR-17, MR-25, CF-02, and CF-03). This finding is consistent with recently calculated water-effects ratios for dissolved Cu in the upper CFR from acute toxicity studies that were consistently greater than 1, and as high as 3.3 (W. Stubblefield, personal communication).

Aquatic vegetation. Responses of emergent macrophytes to whole-sediment exposures were significant for stations MR-07 and MR-17 [32], which were characterized by the highest concentrations of SEM Cu and exceeded the toxicity criterion in the benthic analyses. Stations MR-07 and MR-17 were also the two stations for which amphipod toxicity was associated with ammonia concentrations exceeding the NEC.

The ecological component of the aquatic plant evaluations was based on qualitative observations during the wildlife survey and other field activities [4,5]. Although community structure analyses were not performed, visual observations suggested that aquatic plant communities were healthy and widespread throughout the reservoir. Obvious impacts to emergent plant communities at stations MR-07 and MR-17 were not apparent [32], suggesting that elevated peroxidase activity of aquatic plants is not associated with readily apparent risks to the aquatic ecosystem at Milltown Reservoir.

Amphibians. The sublethal responses observed in amphibians during preliminary studies at Milltown Reservoir were insufficient for statistical analysis [5,33]. However, a possible association is observable between an increasing incidence of malformations from laboratory exposures to reservoir surface waters with increasing metals concentrations. For example, no-effect concentrations of metals in surface water, at <6% effects, were associated with concentrations of Cd and Zn at 1 and 6 μg/L, respectively, and NECs in whole sediments associated with these water samples ranged from 300 to 450 μg/g for Cu and 650 to 1,060 μg/g for Zn. Above these ranges, amphibian malformations in laboratory studies were greater than 20%. The NECs for amphibian responses are similar to the NEC values for SEM Cu at 325 μg/g and Zn at 1,064 μg/g based on amphipod responses [7].

Fish. Laboratory and field studies performed in support of this risk assessment [10–12] and the natural resource damage assessment for the CFR [30] are summarized in Table 5. Feeding studies indicate that a diet of dried, metals-contaminated invertebrates from the upper CFR can decrease growth and survival—and adversely impact the health—of early-life-stage rainbow and brown trout. As summarized in Table 5, tissue concentrations of Cu in natural populations of trout in the upper CFR, at an average of 6.4 μg/g tissue, are similar to the 7.1 μg/g found in the feeding studies. Also similar to the dietary exposed trout, trout collected from the upper CFR near Warm Springs demonstrated a higher incidence of histopathological liver damage, copper inclusions in hepatocyte cytoplasm, and elevated lipid peroxidation products (Table 5).

Feeding studies using diet comprised of invertebrates collected near Turah Bridge just upstream of Milltown Reservoir suggest that the lower metals concentrations typical of the middle CFR may not directly impact fish health, as only minor liver degeneration was observed in these fish (Table 5).

Table 5. Comparison of impacts to trout collected from the upper CFR with dietary exposures in the laboratory

	Cu concentration (μg/g wet wt.)		
	Invertebrates	Trout whole body	Trout health indices
Feeding studies[a]			
Milltown Reservoir[b]	103 (4)/87 (2.2)	1.6 (0.23)/3.5 (0.25)	None
Upper CFR[c]	390 (11)/174 (1.9)	7.1 (0.97)/8.2 (1.5)	Reduced survival Reduced growth Scale loss Degenerative liver Lipid peroxidation Avoidance behavior
Field studies[d]			
Reference	NA	3.0 (0.5)	None
Milltown Reservoir	103 (4)/87 (2.2)	4.3 (0.6)	Minor liver degeneration
Upper CFR	390 (11)/174 (1.9)	6.4 (0.7)	Scale loss Degenerative liver Lipid peroxidation

Data from [10–12, 30]. All values are arithmetic means with standard deviations in parentheses.
NA, not applicable.
[a]Values are presented from two studies in the format "1st study/2nd study" [10,11].
[b]Reference invertebrate samples were collected from near Turah Bridge immediately upstream of Milltown Reservoir (first value [10]), or from the Snake River (second value [11]), and used in feeding studies. Invertebrate measurements represent dried, pelletized diets. Trout body burdens represent specimens fed the first/second respective reference invertebrate diet.
[c]Invertebrates were collected from the upper CFR just below Warm Springs Ponds and used as diet in the trout toxicity tests [10,11]. Invertebrate measurements represent dried, pelletized diets. Trout body burdens represent specimens fed the first/second respective reference invertebrate diet.
[d]Values representative of native invertebrates and trout collected from Turah Bridge immediately upstream of Milltown Reservoir and the upper CFR near Warm Springs Ponds. Invertebrates are those used in the feeding studies. Reference trout were collected from Rock Creek and the Big Hole River.

However, elevated metals in benthic invertebrates from the middle CFR (i.e., in the vicinity of Milltown Reservoir) likely add further stress to trout populations impacted by the presence of Milltown Dam. The near absence of the more metals-sensitive rainbow trout, compared to brown trout, from the upper CFR reaches immediately upstream of Milltown Dam supports this possibility.

Additional studies of rainbow trout that were fed metals-enriched brine shrimp (*Artemia* sp.) with simultaneous exposures to soluble metals at concentrations similar to those in the upper CFR found no adverse health effects [13]. These results do not support the conclusions of dietary studies with invertebrates collected from the upper CFR. However, discrepancies in results between the studies may be due to differences in the nutritional values of the two diet sources and the availability of the dietary metals. Results of Woodward et al. [10,11] at least suggest that elevated metals in the upper CFR could add further stress to trout populations that are at minimal nutritional adequacy.

Metals concentrations in surface waters in the upper CFR occasionally exceed water-quality criteria for the protection of freshwater organisms [26], particularly during summer thunderstorms with high precipitation. However, comparisons of dissolved metals in surface waters with water-quality criteria show far fewer instances of these values being exceeded [26]. The high concentrations of metals in the upper CFR water during the few weeks of spring snowmelt and summer thunderstorms [1] are likely to impart additional stress to trout populations exposed to elevated metals in dietary sources from the river.

Summary of aquatic risk characterization. The sediment benthos was the most studied of habitats in Milltown Reservoir. The weight of evidence from the triad of chemistry, ecology, and toxicity studies suggests that benthic invertebrate communities in the reservoir are at risk in sediments where metal concentrations are elevated and AVS concentrations are low. This combination of elevated metals in oxidizing environments is found in sediments in the ORC (unit 2) and upstream of the railroad berm (unit 7) in the Milltown Reservoir wetland (Fig. 3). Oxidizing environments at risk in the reservoir and upper CFR have been observed in shallow or fast-moving waters in depositional areas where metal concentrations are elevated. From Table 4, a range for the association of Cu concentrations and adverse effects to benthos can be estimated for SEM Cu/AVS values between 0.4 and 0.9, which is equivalent to a range of the sum of SEM/AVS values of 1.5 to 2.2, or extractable Cu of about 250 to 350 $\mu g/g$ in an oxidizing environment. Note that sediment toxicity was generally observed where molar SEM Cu/AVS ratios exceeded unity, consistent with observations of others [56].

Available evidence suggests that aquatic plants are not at risk for community or ecosystem impacts at moderately elevated metals concentrations; however, the station with the highest metal concentrations was not evaluated (i.e., station MR-11). Because of the limited number of stations, observed biochemical effects may be due to the presence of elevated ammonia, metals, or other chemicals in the sediments. The correlation of aquatic plant effects and amphipod toxicity in the same sediments suggests that emergent plants are good indicators of chemical contamination in the wetland at Milltown Reservoir.

Results of preliminary studies on amphibian responses to surface-water exposures were consistent with those of the sediment benthic analyses. Concentrations of metals in sediments at stations that were toxic to amphibians were in the range of concentrations where benthic invertebrate toxicity was observed (i.e., 300 to 450 $\mu g/g$ for total Cu and 650 to 1,060 $\mu g/g$ for Zn). Although normalization of extractable metal concentrations to AVS was unavailable, the results of the amphibian studies support the cstimated thresholds for risks to sediment biota.

The integration of fish exposure, toxicity, and population studies indicate that the health of the trout fishery of the upper CFR is at continued risk from metals contamination of surface water, sediments, and subsequently invertebrates in the riffle environment of the river. Both acute and chronic risks are apparent from the metals runoff during thunderstorms and spring snowmelt and the continued presence of metals in trout food sources.

ECOLOGICAL SIGNIFICANCE OF RISK ASSESSMENT FINDINGS

The ecological significance of the risk assessment results is essential in evaluating the overall hazards to the wetland ecosystem. Risk managers depend on an adequate description of the significance of the risk findings in making decisions about managing a contaminated ecosystem or watershed.

Answering the question of ecological significance of the findings summarized in this report depends on defining the relationships among differing types of "significance." In general, the better quality and the higher quantity of data allow an easier determination of statistical significance. However, as pointed out by Barnthouse et al. [57], very precise data can result in statistical significance even with biologically inconsequential results, or low biological importance. Thus, biologically important impacts are defined as those that affect populations, communities, and ecosystems, rather than individuals as evaluated in toxicity bioassays. This definition is based on the understanding that measurement of impacts to individuals does not necessarily indicate impacts tc the ecosystem. The biologically important results are the focus in setting remedial goals for the site.

The definition of ecological significance can be further addressed by a relative ranking of impacts, as outlined by Duinker and Beanlands [17]:

Major impact: Affects an entire population or species in sufficient magnitude to cause a decline in abundance and/or change in distribution beyond which natural recruitment (reproduction, immigration from unaffected areas) would not return that population or species, or any population or species dependent upon it, to its natural level.

Moderate impact: Affects a portion of a population and may bring about a change in abundance and/or distribution over one or more generations, but does not threaten the integrity of that population or any population dependent upon it.

Minor impact: Affects a specific group of localized individuals within a population over a short period of time, but does not affect other trophic levels or the population itself.

Negligible impact: Any impacts below the minor category are considered negligible.

For the Milltown Reservoir–CFR risk assessment, the inclusion of quantitative studies on chemical exposures, biological effects assessment, and ecological studies was designed to help determine the ecological significance of the results by providing a link among these three legs of the risk assessment triad [48].

Terrestrial habitat findings

For the lower trophic organisms, the sublethal effects observed in the toxicity studies suggest subtle ecological impacts. The study of rooting-zone impacts serves to illustrate the potential significance of sublethal effects measured in biological test systems. The rooting-zone study demonstrated the inhibition of root growth in plants exposed to rhizosphere water from areas of high metals concentrations (i.e., units 2, 7, and 8). An inhibition in root elongation in this bioassay suggests that lateral root growth may be inhibited in certain plant types in those units. Such inhibition could result in decreased absorption of nutrients, including iron, and an imbalance in such nutrients as Cu and Zn. These alterations in turn could affect biomass above ground, as was demonstrated during the greenhouse study with soils from units 2 and 7 [5], and result in iron-deficiency chlorosis. Thus, the risks to wetland vegetation in depositional areas of the reservoir operable unit can be considered minor to moderate.

For the terrestrial habitat, the findings of the exposure and biological effects assessments need to be placed in context with those of the ecological assessment to identify the significance of ecological risk. For example, results of the wildlife survey and wetland delineation study suggested a healthy wetland ecosystem on a site-wide basis. Thus, integration of the results of the ecological studies with the subtle effects observed in the bioassays suggests that the wetland ecosystem on a site-wide basis does not show substantial adverse ecological effects despite possible local impacts. Because community structure analyses of soil invertebrates were not performed, the link connecting the toxicity studies to the apparently healthy wildlife on a site-wide basis is missing. Also, the lack of community studies on smaller discrete areas of the wetland, such as those units identified as exhibiting biological effects, precludes their identification as ecologically impacted. In other words, whether the subtle effects observed in the earthworm and vegetation studies may be observable at the ecosystem level in smaller subunits of units 2, 7, and 8 has not been studied.

The food-web analysis demonstrated that primary consumers and higher trophic organisms like predatory birds, waterfowl, and deer are not at high risk from exposures to site-related contaminants. This conclusion was supported by the finding of low bioavailability of soils metals. The only potential concern was the effect of the modeled intake of metals on muskrat. Given the uncertainties in the modeled intake and the lack of toxicity data on muskrat, it is unlikely that the exposures would be expressed in population or ecosystem impacts. In addition, the waterfowl survey at the wetland suggested that the populations of Canada geese and various duck species are healthy and reproducing. No evidence was observed of contaminated sediment invertebrates affecting the health of waterfowl in the wetland. Similarly, the food-chain transfer study did not suggest that metals in the invertebrates or aquatic plants would impact muskrats, beaver, or mink in the wetland.

In summary, the bioassessment studies and food-chain analysis suggested a lack of overt acute toxic effects from the terrestrial or wetland soils exposures, and that the sublethal effects observed would not likely increase ecosystem risks. From this, risks to the terrestrial habitat can best be characterized as minor.

Aquatic habitat findings

For the aquatic habitat, elevated exposures to sediment metals correlated with toxicity to benthic invertebrates and impacted benthic community structure. The finding of increased number of genera of Chironomidae in sediments exhibiting toxicity to benthic invertebrates is suggestive of an imbalance in benthic community due to metals contamination. Overt effects, such as the decreased abundance or loss of taxa observed in major groups of biota in contaminated Great Lakes sediments [58], are not apparent in Milltown Reservoir or CFR sediments. However, as described in Canfield et al. [8], the type of imbalance seen in benthic community structure at the site has been observed in other studies of sediments with high metals concentrations [50,51], and can be explained by a disturbance in population competition dynamics.

The significance of the risk assessment findings for the sediment benthic habitat can be described as moderate for the communities in oxidizing environments. Although sediment ecology is imbalanced at contaminated stations under oxidizing conditions, it is unknown if other ecological parameters, such as predator–prey relationships, are subsequently affected by this imbalance. The impacted benthic invertebrates in both the upper CFR and the reservoir operable units are undoubtedly of greater significance to trout populations through the transfer of metals during feeding.

A number of lines of evidence indicate severe impacts of contaminant metals on fishery of the upper CFR. For example, concentrations of various metals in surface water of the upper CFR have exceeded water-quality criteria; metals concentrations are elevated in invertebrates from riffle and depositional areas; toxicity tests have demonstrated that fish health is adversely affected following exposure to water and diet at metals concentrations typical of the upper CFR; and trout behavior in toxicity tests suggests avoidance of elevated metals concentrations. Fish with reduced growth rate and health could be eliminated from natural populations where metal stresses are present, and their potential avoidance of CFR waters may prevent adequate access to suitable habitat and food sources. The weight of evidence from fishery studies indicates that the health of trout populations and the fishery ecology of the upper CFR are at major risk from metal contamination.

Finally, as discussed in the first article of this series [1], one of the driving concerns for evaluating the site ecology was the potential for chronic impacts from releases of metals during such episodic events as spring snowmelt and summer thunderstorms Because the ecological and in situ biological assessments were performed during summer and fall, results of the studies could be representative of conditions after the spring snowmelt Also, as spring flows of the upper CFR prior to the risk assessment sampling program were high enough to increase metals concentrations in surface waters, the study results may be reflective of conditions of both chronic exposures and a recovery period from acute impacts

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