Technical Report No. 02-03

Aquatic Biomonitoring At Greens Creek Mine, 2001

by Phyllis Weber Scannell, ADF&G
and Steve Paustian, USFS

May 2002

Alaska Department of Fish and Game — Habitat and Restoration Division

Aquatic Biomonitoring at Greens Creek Mine

Technical Report No. 02-03

By

Phyllis Weber Scannell, ADF&G
and Steve Paustian, USFS

Chip Dennerlein
Director
Habitat and Restoration Division
Alaska Department of Fish and Game
Juneau, AK

Table of Contents

List of Tables

List of Figures

Acknowledgements

Acknowledgements

We thank Kennicott Greens Creek Mining Inc. for the financial and logistical support that allowed us to conduct this biomonitoring project. In particular, we acknowledge the support given by Mr. Bill Oelklaus and Mr. Steve Hutson of Kennecott Greens Creek Mining. We also thank Ms. Deborah Rudis of USFWS, Mr. David Gregovich of ADF&G Region I, and Ms. Laura Jacobs, Pete Schneider and Lance Lerum (USFS) for their help collecting samples. Mr. Jason Harris, Mr. Bruce McIntosh, and Ms. Laura Jacobs of ADF&G conducted laboratory analysis of invertebrate and chlorophyll samples. We thank Dr. Larry Duffy and Mr. Dan Scannell of University of Alaska for conducting Microtox analysis. Ms. Lisa Moen, ADF&G, provided administrative assistance in bringing the report to final form. The University of Alaska, Cooperative Fish and Wildlife Research Unit allowed us to use their laboratory facilities for chlorophyll analysis.

Executive Summary

On 2001, the Alaska Department of Fish and Game and the US Forest Service, in cooperation with the US Fish and Wildlife Service, conducted aquatic biomonitoring in Greens Creek and Tributary Creek. The purpose of the current biomonitoring project was twofold: to document the continued health of the aquatic communities and to establish productivity and taxonomic richness of existing aquatic habitats so comparisons could be made with future conditions of these sites. The 2001 biomonitoring program included periphyton abundance, aquatic invertebrate richness and abundance, fish abundance and distribution, concentrations of select elements in fish tissues, and toxicity testing.

Three sites in Greens Creek and the site in Tributary Creek had complex, diverse aquatic macroinvertebrate communities with high densities. The prevalence of Ephemeroptera and Plecoptera were signs of excellent water quality. Periphyton communities were well

established in each site, especially in Tributary Creek where stream flows are low and scouring flood events are rare.

Fish populations were well established in each site, although populations of coho salmon were lower than expected in the Greens Creek sites, possibly due to high water events in the previous year. Site 48 contained only resident Dolly Varden because access by anadromous fish is restricted. Dolly Varden in Greens Creek at Sites 6 and 54 were a mixture of anadromous and resident fish, and all salmonid fish in Tributary Creek were anadromous.

The concentration of Ag in juvenile Dolly Varden was highest in Tributary Creek. Cd concentrations were highest in fish from Site 6 and Site 48. Cu concentrations were similar in fish from all sample sites. The median Pb concentration was slightly higher in fish from Site 6 than from the other Greens Creek sites. Concentrations of Se and Zn were similar among all sites.

We found no indication of either chronic or acute toxicity (determined with Microtox analysis) in water from all sites. Because there was no toxic response, the IC-20 value for each site was >100%.

Overall, the four sites sampled, three in Greens Creek and one in Tributary Creek, contained productive, diverse communities of algae, invertebrates, and fish.

Introduction

In 2000, an interagency regulatory team made up of representatives from Kennecott Greens Creek Mining Company (KGCMC), Alaska Department of Natural Resources (DNR), Environmental Protection Agency (EPA), United States Forest Service (USFS), United States Fish and Wildlife Service (USFWS), Alaska Department of Fish and Game (ADF&G), State Attorney Generals Office (AGO), and Alaska Department of Environmental Conservation (DEC) were invited by KGCMC to conduct an environmental review of the Greens Creek Mining operation within the Admiralty Island National Monument. From the findings of that review the KGCMC Fresh Water Monitoring plan was updated, including specifications for biomonitoring in areas adjacent to the KGCMC surface facilities associated with the Mine and Mill. This document presents results of the first year (2001) of this biological monitoring at the Greens Creek Mining operation.

The role of biological monitoring is to determine in those stream reaches near the KGCMC surface facilities, the continued use of Greens Creek and its tributaries by fish and other aquatic species, and to document the continued health of the aquatic community. Biomonitoring will detect early changes to the aquatic community that may result from changes in water chemistry, either through surface or groundwater inputs to the system.

Results from biomonitoring usually are compared to baseline conditions, or if baseline data are unavailable, to a reference site that is unaffected by the mine. Few baseline biomonitoring studies as intensive as this current program were conducted before development of the Greens Creek mine using current state-of-the-art protocols. The existing biomonitoring program is designed to compare present conditions to future conditions, with consideration given to any previous monitoring. All biological monitoring follows standard protocols acceptable to USEPA, ADEC, USFS, ADF&G (1998), and Standard Methods (APHA 1992).

Purpose

The objective of the biomonitoring program is to establish existing conditions of the biological communities in selected reaches of Greens Creek and Tributary Creek near to the KGCMC surface facilities. Future sampling during the mine life or during reclamation and closure can be compared to the conditions defined under the current biomonitoring program to detect any changes that may have occurred in aquatic communities.

Elements of the Biological Monitoring Program

The biological monitoring program for the Greens Creek mining and milling operations addresses the following factors:

1. Distribution and abundance of juvenile fish;
2. Whole body concentrations of Cd, Cu, Pb, Se, Ag, and Zn in juvenile fish;
3. Periphyton biomass, estimated by chlorophyll concentrations;
4. Abundance and community structure of benthic macroinvertebrate populations;
5. Standardized laboratory toxicity testing.

Location and Schedule of Monitoring

Three sites were selected for routine biomonitoring: Site 48 (Upper Greens Creek), Site 54 (Greens Creek below Pond D), and Site 9 (Tributary Creek). Site 6 (Middle Greens Creek) will be sampled one time (2001) to provide information on baseline conditions (or conditions at the beginning of the biomonitoring program). KGCMC also routinely monitors the ambient water quality at these sites on a monthly basis. Water quality samples will be collected at each of the biomonitoring sites within the month of that biomonitoring effort. Table 1 summarizes the biomonitorng factors that were sampled at each site, Figure 1 shows the location of the Greens Creek Mine.

Table 1. Summary of Biomonitoring Sites

Site Name	Monitoring Objective	Compare to:	Frequency	Factors	Time to Sample
Upper Greens Creek (Site 48)	Routine, control		Annually for 5 years, then review	FA, FM, P, MI, TOX	mid-late July
Middle Greens Creek (Site 6)	Baseline		Baseline Sample on 5 year schedule, unless indication of WQ exceedence	FA, FM, P, MI, TOX	mid-late July
Greens Creek below Pond D (Site 54)	Routine, treatment	Control	Annually for 5 years, then review	FA, FM, P, MI, TOX	mid-late July
Tributary Creek (Site 9)	Baseline	Change over time	Annually for 5 years, then review	FA, FM, P, MI, TOX	mid-late July

KEY:
WQ water quality – collected monthly a each Greens Creek site, and with each biomonitoring sampling effort
FA fish abundance and distribution
FM fish metals content
P periphyton biomass
MI macroinvertebrate abundance, community
TOX micro-toxicity tests

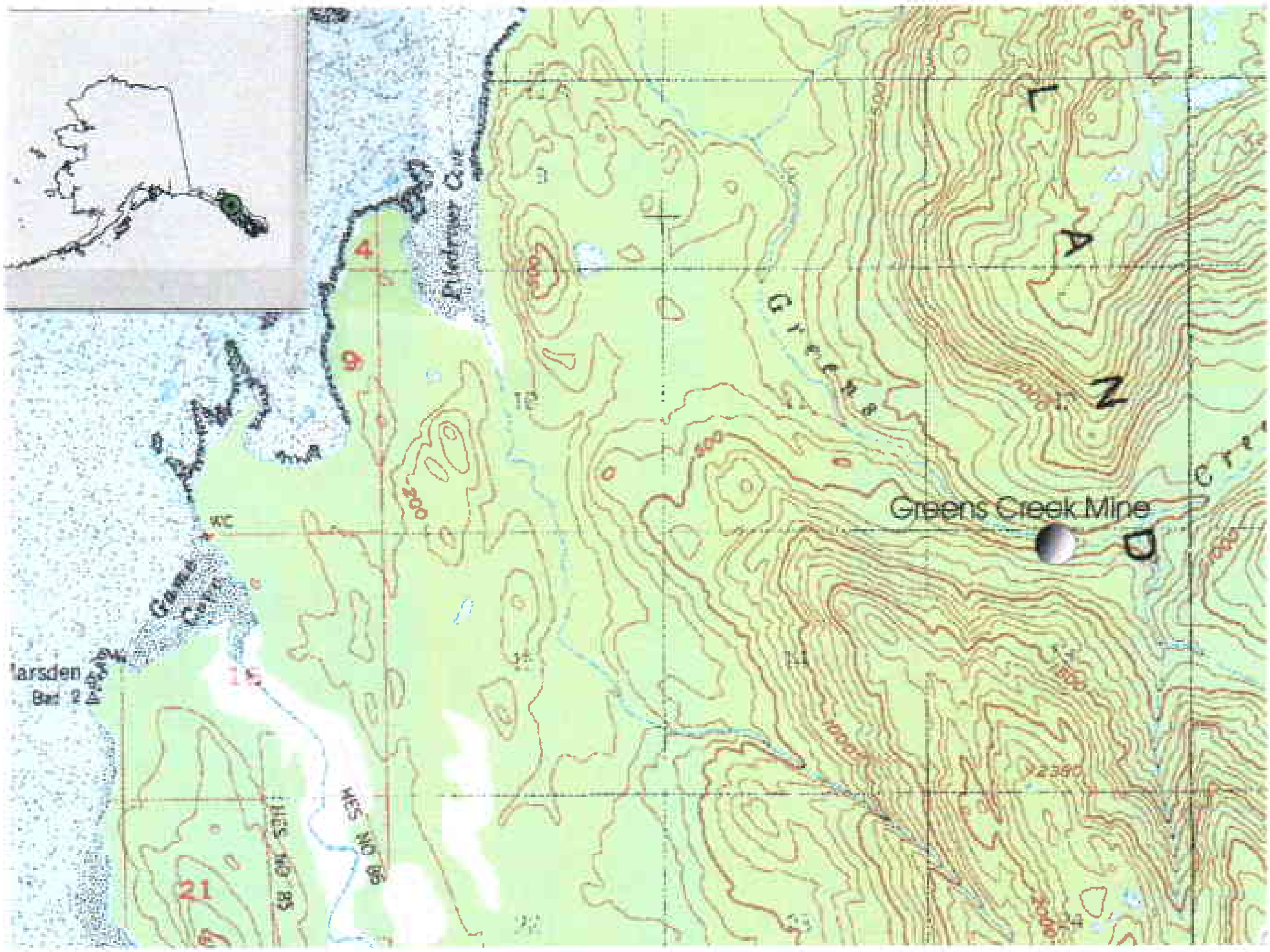

Figure 1. Location of Greens Creek Mine, Adrmiralty Island, AK.

Methods

Periphyton Biomass

Rationale

Periphyton, or attached algae, is sensitive to changes in water quality. Their abundance confirms that productivity is occurring at specific locations within a water body.

Sample Collection and Analysis

The protocol for collecting stream periphyton follows the protocol from the Alaska Department of Fish and Game (1998) and Barbour et al (1997). Periphyton were sampled during a period of stable flow. Ten rocks were collected from the stream benthos in each study reach. A 5-cm x 5-cm square of high-density foam was placed on the rock. Using a small toothbrush, all material around the foam square was removed and rinsed away with clean water. The foam was removed from the rock and the rock was brushed with a clean toothbrush and rinsed onto a 0.45 μm glass fiber filter, attached to a hand vacuum pump. After extracting as much water as possible, approximately 1 ml saturated $MgCO_3$ was added to the filter to prevent acidification and conversion of chlorophyll to phaeophytin. The filter was wrapped in a large filter (to absorb any additional water), labeled, placed in a sealable plastic bag, and packed over desiccant. Filters were frozen on site in a lightproof container with desiccant, then transported to Fairbanks where they were kept frozen until laboratory analysis.

In Tributary Creek, we sampled stream periphyton directly from the substrate and from glass slides placed in the stream. Three screens, each containing 15 glass slides, were placed on the stream bottom and anchored in place. After a 3-week colonization period, the slides were removed, placed in labeled slide boxes, and frozen.

Methods for extraction and measurement of chlorophyll followed USEPA protocol (EPA 1997). Filters were removed from the freezer, cut into small pieces, and placed in a centrifuge tube with 10 ml of 90% buffered acetone. Centrifuge tubes were placed in a metal rack, covered with aluminum foil, and held in a dark refrigerator for 24 hrs. After extraction, samples were centrifuged for 20 minutes at 1600 rpm, and then read on a Shimadzu Spectrophotometer UV-601 at optical density (OD) 664 nm, OD 647 nm, and OD 630 nm and at OD 750 nm to correct for turbidity. An acetone blank was used to correct for the solvent. Samples were then treated with 0.1 ml of 0.1 N hydrochloric acid to convert chlorophyll to phaeophytin, and read at OD 665 nm and OD 750 nm. Chlorophyll-a, -b, and –c and phaeophytin were determined according to Standard Methods (APHA 1992).

Benthic Macroinvertebrates

Rationale

The primary objective of sampling benthic macroinvertebrates was to collect sufficiently quantitative samples to characterize structure and abundance of benthic macroinvertebrate communities. Benthic macroinvertebrate species abundance and taxonomic richness is a useful measure of stream health.

Field Collection and Analysis

Five benthic samples were collected with a modified Hess sampler from each sample site. We used a stratified random sample design, modified from Barbour et al. (1997) by limiting the randomly placed samples to riffle areas. This sample design eliminated variability from sampling pool or other marginal habitats where macroinvertebrates are unlikely to occur. For each sample, the substrate was first manually disturbed, then rocks were brushed and removed. After the larger substrate was removed, the fine gravels were disturbed to a depth of approximately 15-20 cm. Macroinvertebrates disturbed from the substrate were collected in a 1-meter, 300 *u*m mesh net, attached to the sampler. The sample was removed, placed in pre-labeled whirl-pack bags, and preserved in 70% ETOH.

Invertebrate samples were later sorted from all debris and identified to the lowest practical taxonomic level. Larger samples were sub-sampled with a gridded tray subsampler. Randomly selected grids were sorted and identified until a minimum of 300 macroinvertebrates was found. Four samples were re-sorted to determine the accuracy in recovering macroinvertebrates. We found that recovery was approximately 98%.

Abundance of Rearing Fish

Rationale

Rationale

The purpose of juvenile fish monitoring is to determine potential trends in the numbers of Dolly Varden (*Salvelinus malma*) and coho salmon (*Onchorynchus kistuch*) in stream segments near the surface mine facilities in the Greens Creek and Tributary Creek drainages. The sampling protocol was derived from Aho (2000). Sample design and methods followed procedures in the 2000 Greens Creek Fresh Water Monitoring Plan. Precise GPS coordinates were measured at the upstream end of four stream reaches (28 m to 135 m in length). A complete set of digital photos was taken to document site conditions at each survey reach

Sample Collection and Analysis

Fish population estimates were made with a three-pass removal method, using ¼ in mesh minnow traps baited with salmon eggs, treated with Betadyne. At each site, a sample reach was identified and marked with aluminum tree tags and metal stakes driven into the streambank. Approximately 25 minnow traps were deployed for each sampling event. Sample reaches varied in length among sites because of the limited availability of suitable habitat to set traps. In Greens Creek at Site 48, we sampled a 75-meter reach, in Greens Creek at Site 6, a 135-meter reach; in Greens Creek at Site 54, a 28-meter reach, and in Tributary Creek at Site 9, a 44-meter reach.

Traps were placed throughout the sample section focusing on pools, undercut backs, bank alcoves, and under root-wads or logjams. Where possible, natural obstructions, like shallow riffles or small waterfalls over log steps, defined upper and lower section boundaries to minimize fish movement into the sample section during sampling.

Minnow traps were set for about 1.5 hours at which time all captured fish were transferred to plastic buckets with holes drilled in the sides. Buckets were placed in the stream for water exchange to keep the fish aerated. The traps were re-baited and reset for another 1.5- hr period. While the second set was fishing, fish captured during the first set were identified to species, counted, and measured to fork length.

A subset of the fish population sample was retained for whole body analysis for metals accumulation. Fish not retained for the metals bioassay were returned to the stream immediately after sampling was completed.

Metals Concentrations in Whole Body Juvenile Fish

Rationale

The response time for juvenile fish to accumulate metals is rapid; for example, ADFG has documented metals accumulation in juvenile Dolly Varden within five to six weeks after dispersing from their overwintering grounds to mineralized and unmineralized tributaries (Weber Scannell and Ott 2001). Should changes occur at the Greens Creek mine that result in higher concentrations of metals in the creek, tissue sampling of juvenile fish should reflect these changes.

Sample Collection and Analysis

Six juvenile Dolly Varden were caught in baited minnow traps at each sample site and measured to fork length. The fish were individually packed in clean, pre-labeled bags, placed in an acid-washed cooler, and frozen on site until transport to Fairbanks. We followed the techniques of Crawford and Luoma (1993) for minimizing contamination of the samples. In Fairbanks, the fish were weighed without removal from the bags (we corrected for the weight of the sample bag). The fish were submitted to a private analytical laboratory, where they were digested, dried, and analyzed for Ag, Cd, Cu, Pb, Se, and Zn on a dry-weight basis, with percent moisture reported. In 2001, all fish retained for metals analysis were Dolly Varden, although samples from Sites 48, 6, and 54 contained a mixture of resident and anadromous forms. In 2000, samples from Site 54 and Tributary Creek both contained a mixture of coho salmon and Dolly Varden.

Samples were numbered following the convention used by ADF&G:

Date/Stream Code/Species Code/Age Code/Sample Number

An example fish label would read: 071201GC54DVJ01, Where 070201 represents July 2, 2001; GC54 represents Greens Creek, Site 54; DV represents Dolly Varden; J represents juvenile; and 01 represents sample replicate #1.

Quality Control / Quality Assurance of Laboratory Analysis

The analytical laboratory provided quality assurance/quality control information for each analyte, including matrix spikes, standard reference materials, laboratory calibration data, sample blanks, and sample duplicates.

Toxicity Testing

Rationale

Toxicity tests measure the combined toxic effects of all constituents in any particular sample. They measure toxicity of multiple components that may not be able to be measured using standard analytical techniques because some substances can be toxic in amounts that are below detection limits. This is especially true when multiple toxic components synergistically cause toxicity, although each component may be below a detection limit. A commonly available test is the Microtox test, which uses the luminescent bacteria *Vibrio fischeri*. When grown under optimum conditions, the bacteria produce light as a by-product of their cellular respiration. Bacterial bioluminescence is directly related to cell respiration, and any inhibition of cellular activity results in a decreased rate of respiration and a corresponding decrease in the rate of luminescence (Azur 1999).

Sample Collection and Analysis

The Microtox test (Azur 1999) requires approximately 100 ml of sample water per test replicate. Water samples were collected at the same time other biomonitoring sampling was done. Samples were kept refrigerated until they were analyzed for both chronic (24-hr) and acute (1-hr) Microtox toxicity. Each analysis consisted of 4 sample controls, 4 positive controls, and 4 replicates for each of 6 dilutions. The acute test used solutions of 0% to 45% of the test (creek) water mixed with reagent water. The chronic test used solutions of 0% to 100% of the creek water. IC-20 concentrations were calculated from each site's chronic toxicity data. The IC-20 represents the estimated toxicant concentration that would cause a 20 percent reduction in a non-lethal biological measurement of the test organism. In the case of the tests conducted in this study , the non-lethal biological measurement was the growth of the test species, *Vibrio fisherii.*

Results and Discussion

Samples collected in summer 2001 represent the first year of a long-term biomonitoring program. We included 2000 results on fish tissues for comparison. No comparisons are made among sites except to show relative productivity.

Upper Greens Creek, Site 48

Upper Greens Creek at Site 48 (Figure 2) was selected as an upstream, control site for comparison to "treatment" sites adjacent to and downstream from the KGCMC facilities. This site lies approximately ½ mile upstream of the weir which blocks access to anadromous fish. Therefore, the only salmonid species at this site is resident Dolly Varden.

Figure 1. Greens Creek Site 48.

Periphyton Biomass

Periphyton communities in upper Greens Creek were similar to those in Greens Creek at Site 54: chlorophyll-a concentrations ranged from 0.3 to 5.6 mg/m^2 (Figure 3). Forty percent of the chlorophyll b samples were less than the limit of detection (0.005 mg/m^2) and the remaining samples were less than 0.15 mg/m^2. Chlorophyll-c also was low; all samples contained concentrations less than 0.7 mg/m^2.

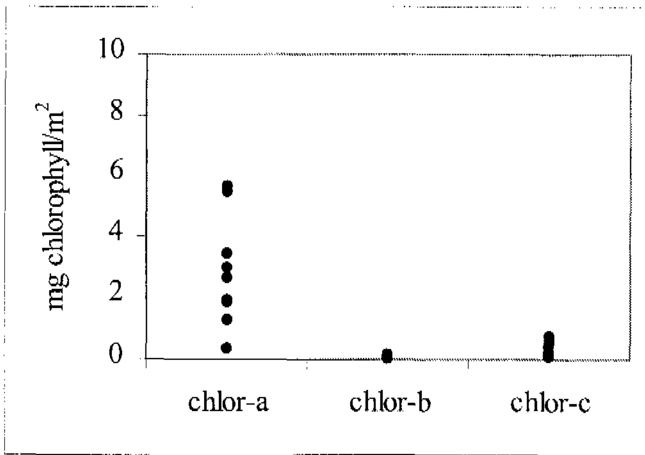

Figure 2. Concentrations of chlorophyll a, b, and c, Site 48, 2001.

Benthic Macroinvertebrate Community

Invertebrate density and taxonomic richness were high in Greens Creek at Site 48, as at Site 54 (Appendix 1). In 2001, we found an average macroinvertebrate density of 2,368/m^2 and a total of 25 individual taxa (usually genus level). Ephemeroptera and Plecoptera were the dominant groups (97.% of the total organisms collected) with 0.5% Trichoptera. The Heptageniidae: *Epeorus* was the dominant taxon (38% of the community). *Epeorus* is considered a sign of excellent water quality (Merritt and Cummins 1996, McCafferty 1998).

Juvenile Fish Community

Upper Greens Creek, Site 48, lies approximately ½ mile upstream of the weir which blocks access to anadromous fish. Therefore, only resident Dolly Varden occur at this site. The Greens Creek Site 48 sample reach was an MM2 Channel Type (Paustian et al. 1999) with an average width of 10 meters and gradient 2 to 4 percent. Cobble is the dominant substrate and large woody material is a key factor in pool formation and fish habitat cover. The 2001 fish population survey conducted within a 72-meter reach, collected a total of 48 Dolly Varden with an estimated population density of 0.20 fish/m^2. The fork length of the fish ranged from 55 mm to 140 mm (Figure 4). Because growth rates of resident fish are highly variable, it is not possible to estimate the number of age classes represented by these fish.

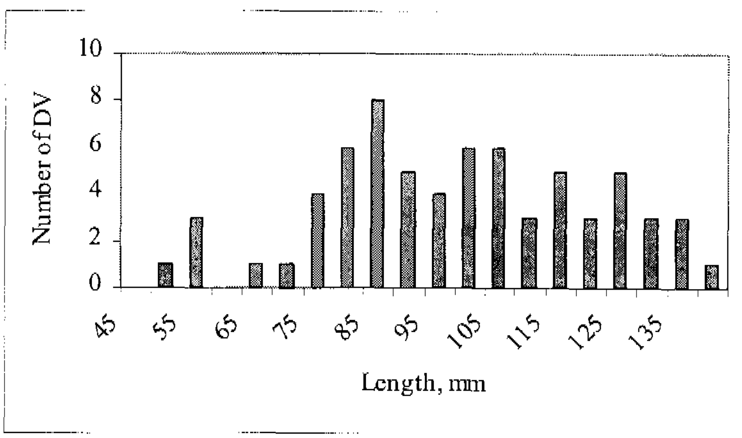

Figure 3. Size distribution of Dolly Varden, Site 48, 2001.

Metals in Juvenile Fish

Only resident Dolly Varden juvenile fish were found at Site 48; therefore, the fish collected and analyzed for metals concentrations might be considerably than fish from other sites. All whole body fish tissue data are reported as dry weight basis (Table 2). The complete laboratory results are presented in Appendix 2.

Table 2. Concentrations of metals in juvenile fish, Site 48, 2001.

	Median	Maximum	Minimum
Ag, mg/kg	0.02	0.03	0.02
Cd, mg/kg	1.41	2.27	0.89
Cu, mg/kg	7.05	8.3	4.7
Pb, mg/kg	0.185	0.23	0.1
Se, mg/kg	5.5	6.2	4.6
Zn, mg/kg	168.5	189	138

Toxicity Testing

We did not detect any toxicity in any of the dilutions of Site 48 water with either the chronic or acute Microtox toxicity tests. All sample replicates showed growth of *Vibrio fischeri* similar to the control the calculated IC-20 was >100%.

Middle Greens Creek, Site 6

Middle Fork Greens Creek at Site 6 (below the confluence of Bruin Creek, Figure 5) has been monitored continuously under the FWMP since 1978. The site was located to detect potential effects on Greens Creek from activities in the KGCMC mine, mill, and shop areas. Access of anadromous fish to this stream reach was created by KGCMC in 1989 by installing a fish pass in a waterfall about 3 miles downstream. This site is near the upper limit of anadromous fish, defined by a weir located about ½ mile upstream. Both Dolly Varden and coho salmon have been found in this reach. Biomonitoring information from this site will be used to detect possible changes in aquatic communities that may occur from natural causes or as a result of mine activities.

Figure 4. Greens Creek Site 6.

Periphyton Biomass

Chlorophyll-a concentrations in Greens Creek at Site 6 ranged from 0.37 to 7.15 mg/m2 in late July 2001 (Figure 6). Concentrations of chlorophyll b were near detection (0.005 mg/m2), and concentrations of chlorophyll-c ranged from 0.16 to 1.9 mg/L. The presence of chlorophyll-c suggests the importance of diatoms to the periphyton community.

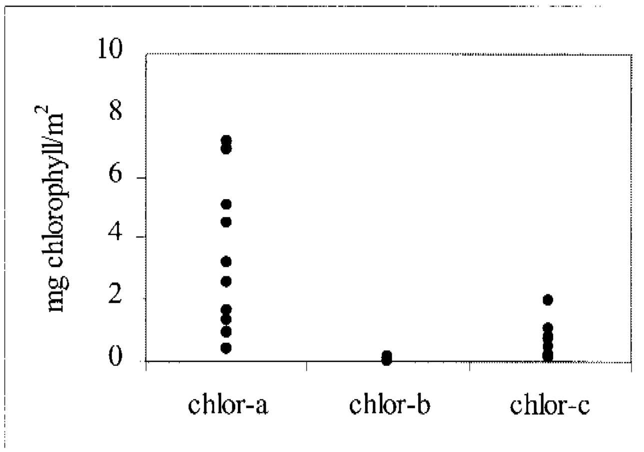

Figure 5. Concentration of chlorophylls a, b, and c, Site 6, 2001.

Benthic Macroinvertebrate Community

Invertebrate density and taxonomic richness were high in Greens Creek at Site 6, as in Sites 54 and 48 (Appendix 1). In 2001, we found an average macroinvertebrate density of 1,996/m^2 and a total of 20 individual taxa (usually genus level). Ephemeroptera and Plecoptera were the dominant groups (95% of the total organisms collected) with 0.2% Trichoptera. The Heptageniidae: *Epeorus* was the dominant taxon (41% of the community). *Epeorus* is considered a sign of excellent water quality (Merritt and Cummins 1996, McCafferty 1998).

Juvenile Fish Community

In July 2001, the USFS surveyed a 135-meter stream reach at Site 6 to estimate the fish population. Both Dolly Varden and coho salmon have been previously documented in this reach; however, in 2001, only Dolly Varden were found. The size distribution of Dolly Varden was similar to Greens Creek at Site 54, with the majority of fish in the 80 to 110 mm range (Figure 7). A few fish were evenly distributed from 120 to 170 mm. A total of 175 Dolly Varden were collected in this sampling effort and the estimated density of Dolly Varden was 0.13 fish/m^2.

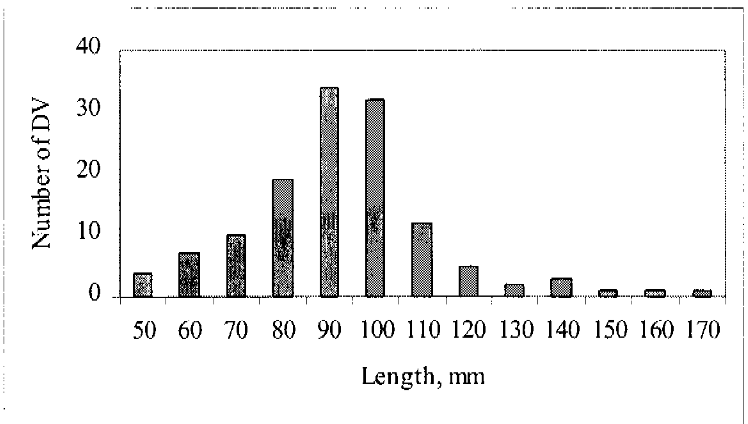

Figure 6. Size distribution of Dolly Varden, Site 6, 2001.

Metals in Juvenile Fish

Six resident Dolly Varden juvenile fish (from 109 to 168 mm fork length) were collected and analyzed for whole body concentrations of Ag, Cd, Cu, Pb, Se, and Zn (Table 3). All data are reported as dry weight. The complete data are presented in Appendix 2.

Table 3. Concentrations of metals in juvenile fish, Site 6, 2001.

	Median	Maximum	Minimum
Ag, mg/kg	0.03	0.04	0.02
Cd. mg/kg	0.865	1.94	0.73
Cu, mg/kg	6.7	16.7	4.6
Pb, mg/kg	1.51	1.96	0.33
Se, mg/kg	4.55	5.3	4.3
Zn, mg/kg	170	215	126

Toxicity Testing

We did not detect any toxicity in any of the dilutions of middle Greens Creek at Site 6 water with either the chronic or acute Microtox toxicity tests. All sample replicates showed growth of *Vibrio fischeri* similar to the control and the calculated IC-20 was >100%.

Greens Creek Below D-Pond, Site 54

Greens Creek at Site 54 (Figure 8) is located about ¼ mile downstream of Site 6, about ¾ mile downstream of the weir, and near the upper limit of anadromous fish. Anadromous fish access to this stream reach was created by KGCMC in 1989 by installing a fish pass in a waterfall area some 3 miles downstream. Both Dolly Varden and coho salmon have been previously documented in this reach.

Figure 7. Greens Creek Site 54.

Periphyton Biomass

Stream primary productivity was estimated from samples of periphyton biomass, measured as chlorophyll concentrations. The concentration of chlorophyll-a ranged from 1.4 to 4.9 mg/m^2 in July 2001 (Figure 9). Eighty percent of the samples contained concentrations of chlorophyll b that were below the limit of detection (0.005 mg/m^2). Concentrations of chlorophyll-c were above detection, but all samples contained less than 0.5 mg/m^2.

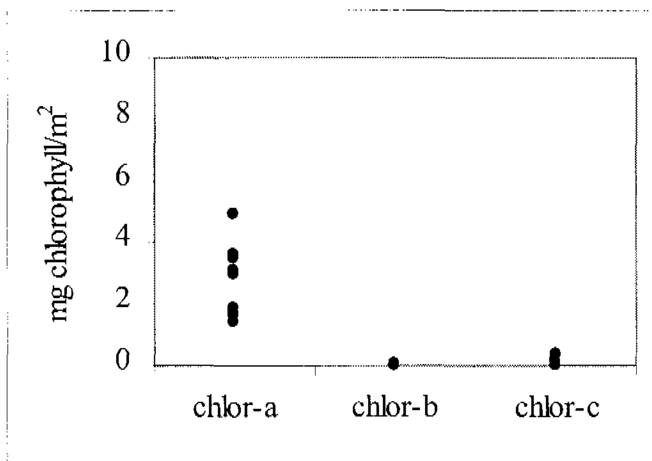

Figure 8. Concentration of chlorophylls a, b, and c, Site 54, 2001.

Benthic Macroinvertebrate Community

In 2001, we found an average macroinvertebrate density of 3,564 /m^2 and a total of 29 individual taxa (usually genus level, Appendix 1). Ephemeroptera and Plecoptera were the dominant orders (96% of the total organisms collected) with 0.4% Trichoptera. The Heptageniidae: Epeorus was the dominant taxon (52.5% of the community). This mayfly species is considered a sign of excellent water quality (Merritt and Cummins 1996, McCafferty 1998).

Both the taxonomic richness and macroinvertebrate density were higher at this site than at any of the other sites sampled in 2001.

Juvenile Fish Community

The Greens Creek sample reaches at Site 54 is of the MM2 Channel Type (Paustian et al. 1999), with an average channel width of 10 meters and stream gradient from 2 to 4 percent. Cobble is the dominant substrate material and large woody debris (fallen logs, roots, etc.) is integral to pool formation and fish habitat cover. In 2001, the USFS reported few juvenile coho salmon in Greens Creek at Site 54, which were not measured. However, Dolly Varden juveniles were abundant, with a total of 138 fish collected in the 28-m reach that was sampled. The USFS estimated a fish density of 0.58 Dolly Varden per m^2 of suitable habitat. The majority of Dolly Varden fork lengths ranged from 95 to 110 mm (Figure 10). The size distribution from 30 mm to 165 mm suggests the presence of four age classes, from age-0 to age-3 plus a possible age-4 fish at 165 mm.

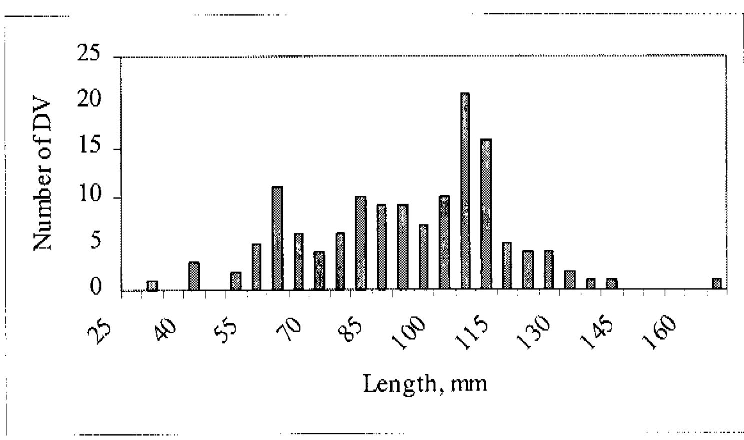

Figure 9. Size distribution of Dolly Varden, Site 54, 2001.

Metals in Juvenile Fish

In 2000, we collected 6 coho salmon for tissue analysis from Greens Creek at Site 54 (we were unable to collect a full sample of 6 Dolly Varden). In 2001, we collected 6 Dolly Varden. Most of the fish collected in 2001 were anadromous forms (DeCicco, pers. comm. 2001). Both median and maximum concentrations of Ag, Cd, Cu, Pb, and Zn were higher in 2000 coho salmon than in 2001 Dolly Varden (Figures 11 to 14 and 16). Median and maximum concentrations of Se were slightly higher in 2001 than in 2000 (median for 2000 = 4.7 mg/kg and 2001 = 5.1 mg/kg, Figure 15). Differences between fish collected in 2000 and 2001 may be due to differences in the species composition of the samples. All data from fish tissues is provided in Appendix 2.

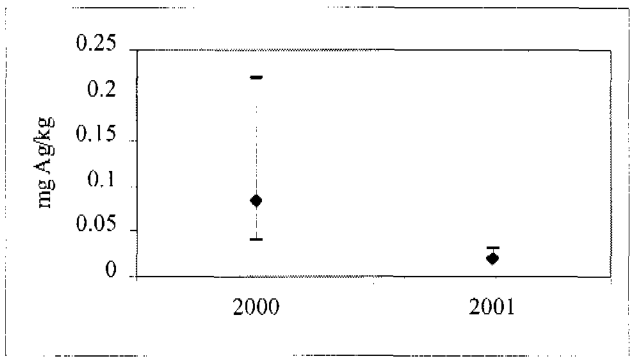

Figure 10. Concentration of Ag in whole body juvenile fish, Greens Creek at Site 54 (median, maximum, and minimum).

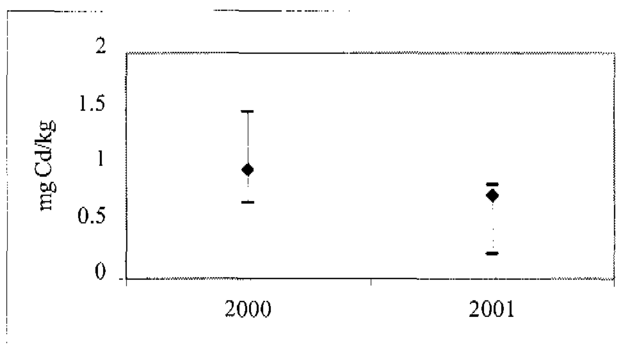

Figure 11. Concentration of Cd in whole body juvenile fish, Greens Creek at Site 54 (median, maximum, and minimum).

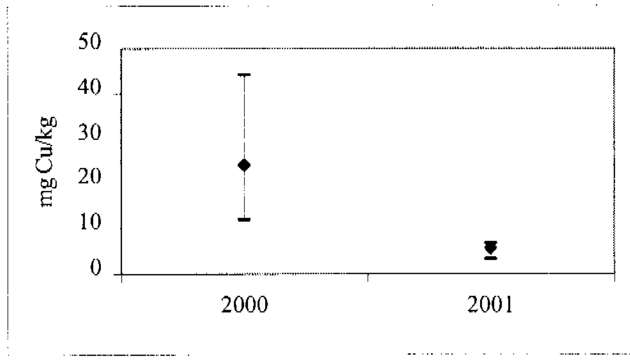

Figure 12. Concentration of Cu in whole body juvenile fish, Site 54 (median, maximum, and minimum).

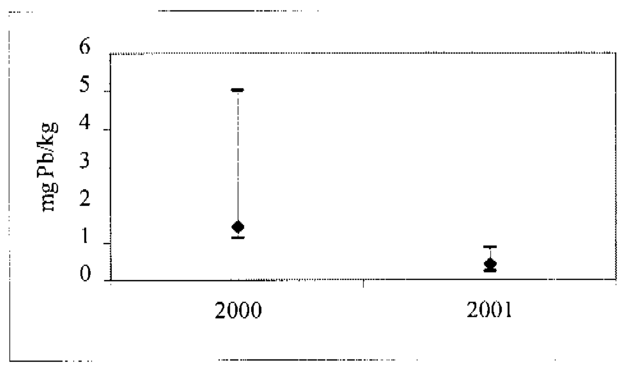

Figure 13. Concentration of Pb in whole body juvenile fish, Site 54 (median, maximum, and minimum).

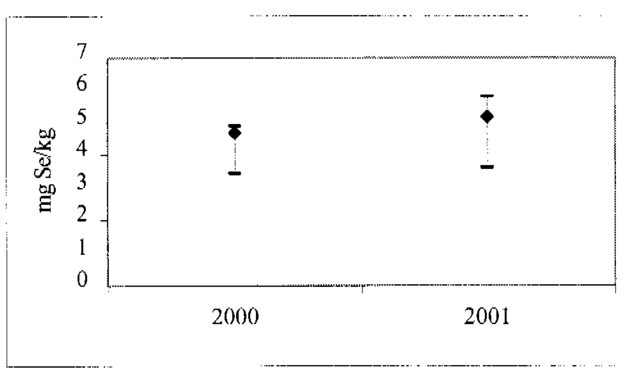

Figure 14. Concentration of Se in whole body juvenile fish, Site 54 (median, maximum, and minimum).

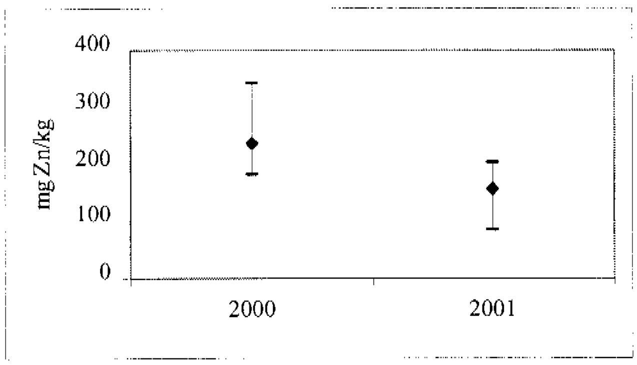

Figure 15. Concentration of Zn in whole body juvenile fish, Site 54 (median, maximum, and minimum).

Toxicity Testing

We did not detect any toxicity in any of the dilutions of Site 54 water with either the chronic or acute Microtox toxicity tests. All sample replicates showed growth of *Vibrio fischeri* similar to the control and the calculated IC-20 values were >100%.

Tributary Creek. Site 9

Tributary Creek is a small tributary with a dense canopy (Figure 17). This site was previously monitored under the FWMP from 1981 through 1993 and is included in the current biomonitoring program because it is located downstream from the KGCMC dry tailings placement facilities. This creek has populations of pink (*Onchorynchus gorbuscha*), chum (*O. keta*), and coho salmon, and Dolly Varden char.

Figure 16. Tributary Creek.

Periphyton Biomass

One of the screens containing glass slides in Tributary Creek was no longer submerged when the water level dropped. The slides were left dry for in unknown time. Extraction tests for phaeophytin showed that more than 25% the chlorophyll from this group of slides had degraded to phaeophytin. Therefore, we did not include this group in the results. The remaining the two screens of slides that had remained submerged contained less than 7% phaeophytin.

Samples collected directly from the stream substrate contained higher concentrations of chlorophyll-a than the glass slide (Figure 18). However, the within sample variability of substrate samples was greater than for the slides.

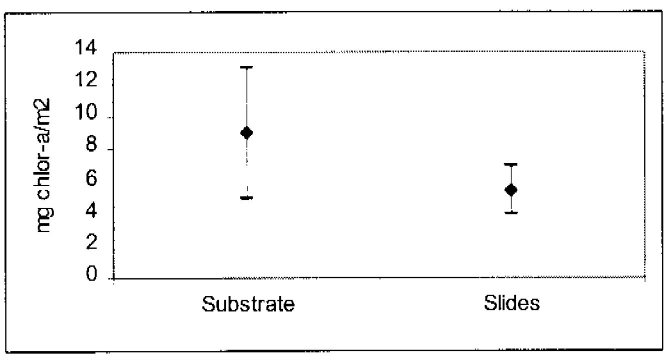

Figure 17. Chlorophyll-a concentrations from Tributary Creek, 2001. Samples from substrate and glass slides, mean and 95% confidence interval.

The concentration of chlorophyll-a (combined substrate and slide samples) in Tributary Creek ranged from 0.16 to 16.6 mg/m^2 of stream substrate (Figure 19). The concentration of chlorophyll b was low and most samples were at our limit of detection (0.005 mg/m^2). Low concentrations of chlorophyll b indicate a periphyton community with little filamentous green algae or blue-green bacteria. The higher concentration of chlorophyll-c found at this site indicates a community dominated by diatoms.

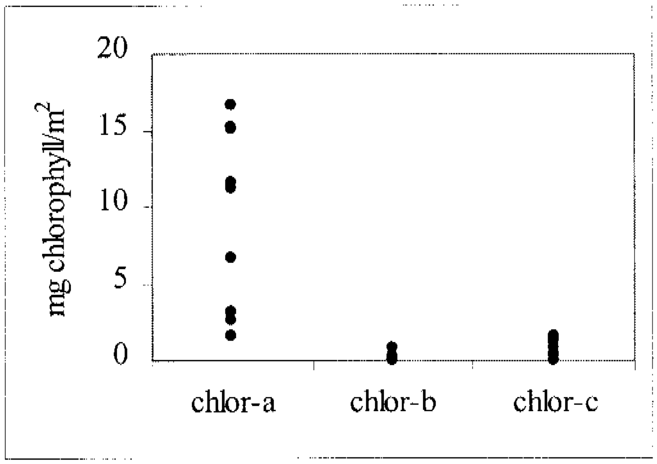

Figure 18. Concentrations of chlorophylls a, -b, and –c in Tributary Creek, 2001. Samples from slides and substrate combined.

Benthic Macroinvertebrate Community

The average density of aquatic macroinvertebrates at this site was 1018 macroinvertebrates/m^2 of stream bottom and 21 different taxa (usually identified to genus level) (Appendix 1). Of the macroinvertebrates collected, 54% were Ephemeroptera, Plecoptera, and Trichoptera (EPT) taxa. The dominant taxa at Site 9 were Ostracoda (18% of the total collected) and Ephemeroptera Heptageniidae *Cinygmula* (17 %) Both Ostracoda and *Cinygmula* are regarded as signs of excellent water quality (Merritt and Cummins 1996, McCafferty 1998).

Juvenile Fish Community

The Tributary Creek reach (FP3 Channel Type, Paustian 1999) is 2 meters wide with a 1% stream gradient. Fine gravel is the dominant substrate. A variety of fish rear in Tributary Creek, including coho salmon, Dolly Varden, cutthroat trout (*O. clarki)* and sculpin (species not given). Coho, pink and chum salmon spawn in this creek. Cutthroat and sculpin (4 total collected) are minor components of the fish community in the

Tributary Creek reach. Juvenile coho salmon is the most commonly occurring rearing fish species (total caught = 120 in the 44 m reach surveyed) followed by Dolly Varden (total caught =81).

The juvenile coho salmon at this site are likely a mixture of age classes, the lower range being age 0 and the higher range a mixture of age 2 and 3 (Figure 20). Morrow (1980) states that coho salmon spend 1 to 2 years in freshwater before going to sea, although the time period to smolt can be as long as 3, and rarely, 4 years.

According to the age-length distributions presented by Armstrong and Morrow (1980), the Dolly Varden caught in Tributary Creek are likely age 1 and age 2-3, the length frequency distribution (Figure 21) suggests there are no age 0 fish. Armstrong and Morrow (1980) provide average ranges of 25 to 33 mm for age 0 fish, 51 to 75 mm for age 1 fish, 61 to 98 mm for age 2 fish, and 65 to 137 mm for age 3 fish).

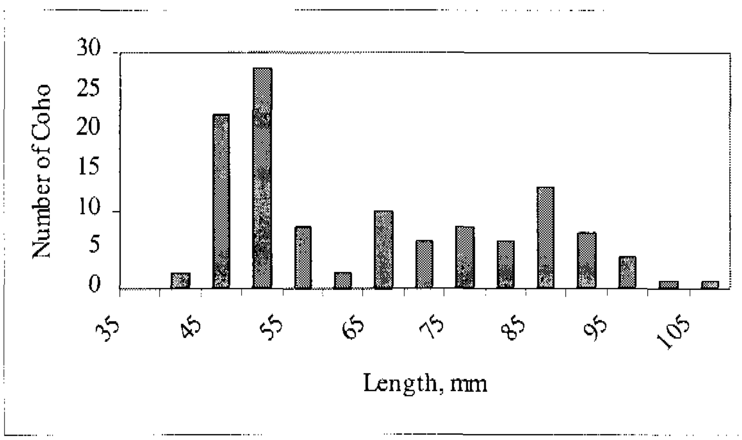

Figure 19. Size distribution of coho salmon, Tributary Creek at Site 9, 2001.

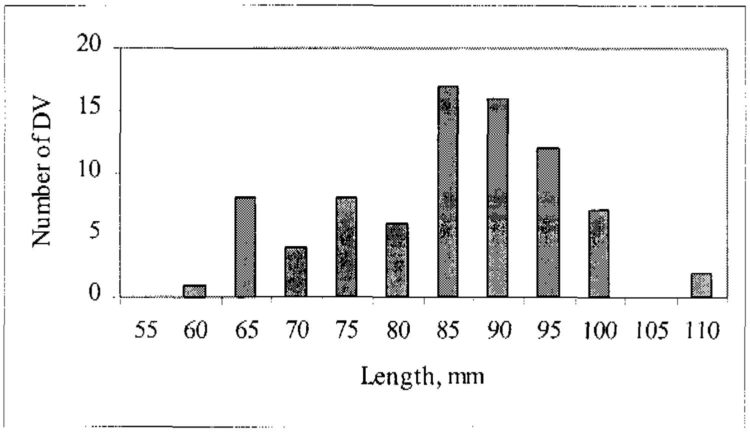

Figure 20. Size distribution of Dolly Varden, Tributary Creek at Site 9, 2001.

Tributary Creek (Site 9) has high densities of both Dolly Varden and coho salmon juveniles, the USFS estimated fish densities of 0.92 Dolly Varden and 0.80 coho salmon juveniles per m^2. These densities are considerably higher than regional averages of 0.26 coho salmon parr per m^2 and 0.30 Dolly Varden parr per m^2 for this channel type (USFS FP3 Channel Type, Paustian et al. 1999). The coho salmon parr density estimate for Site 9 is based on fish greater than 55 mm fork length, based on the assumption that fish under 55mm are young of the year (age 0) fry (Bryant 2000). Tributary Creek at Site 9 is located low in the Greens Creek watershed where recruitment of spawning adults is expected to be high. The watershed above this site in influenced by palustrine wetland inflow buffered by small ponds that enhance the habitat for rearing fish. This site has relatively warm water temperature, a stable flow regime and high influx of organic nutrients when compared to similar streams of this size and type. We believe that the location of this site within the watershed and the habitat features contribute to the high densities of fish found in this first year of sampling.

Metals in Juvenile Fish

Concentrations of Ag, Cd, and Zn were similar in anadromous Dolly Varden tested in 2001 and juvenile coho salmon and anadromous Dolly Varden tested in 2000 (Figures 22, 23, and 27). Concentrations of Ag are near the Method Reporting Limit and should be regarded as semi-quantitative. Median concentrations of Cu and Pb were lower in 2001 than 2000; however the 2001 sample contained one fish with higher Pb concentrations (Figures 24 and 25). Concentrations of Se were slightly higher in 2001 (median = 6.8 mg/kg) than in 2000 (median = 3.65 mg/kg) (Figure 26). All data from fish tissues is provided in Appendix 2.

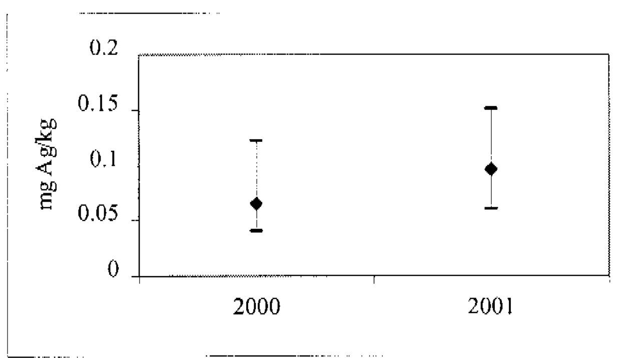

Figure 21. Concentration of Ag in whole body juvenile fish, Tributary Creek. (median, maximum, and minimum).

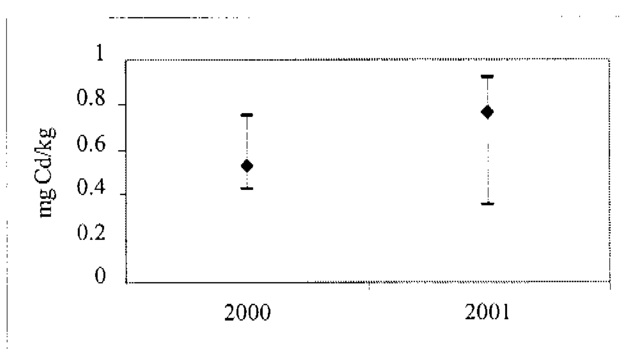

Figure 22. Concentration of Cd in whole body juvenile fish, Tributary Creek. (median, maximum, and minimum).

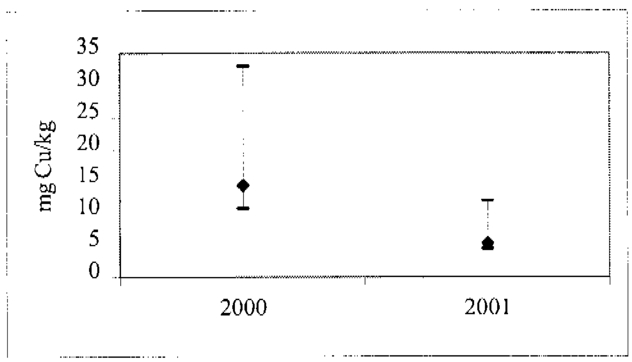

Figure 23. Concentration of Cu in whole body juvenile fish, Tributary Creek. (median, maximum, and minimum).

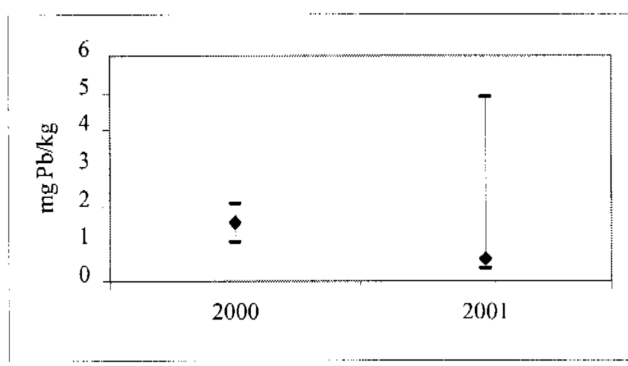

Figure 24. Concentration of Pb in whole body juvenile fish, Tributary Creek. (median, maximum, and minimum).

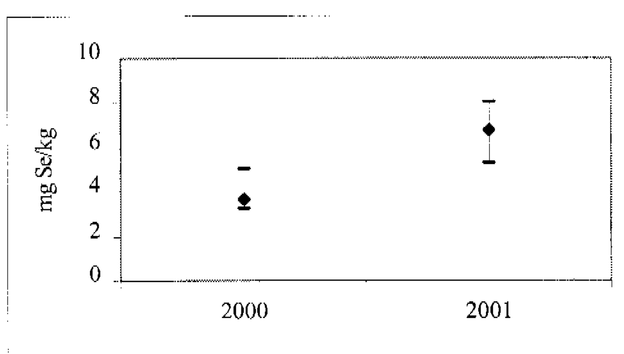

Figure 25. Concentration of Se in whole body juvenile fish, Tributary Creek. (median, maximum, and minimum).

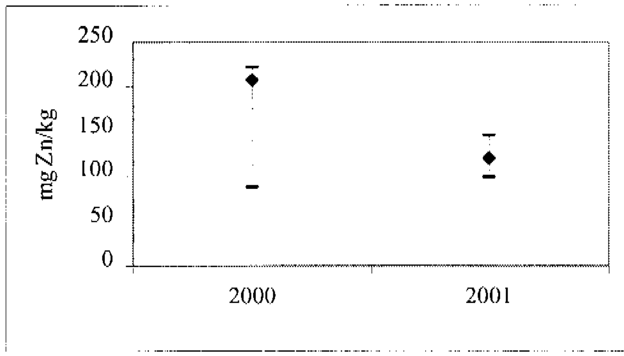

Figure 26. Concentration of Zn in whole body juvenile fish, Tributary Creek. (median, maximum, and minimum).

Toxicity Testing

We did not find any toxicity in any of the dilutions of Tributary Creek water with either the chronic or acute Microtox toxicity tests. The growth of the test species, *Vibrio fisherii*, was the same in controls and all test dilutions (100% Tributary Creek water for chronic tests and 45% for acute tests). Therefore, the calculated IC-20 value was >100%.

Comparisons Among Sites

Periphyton Biomass

The abundance of periphyton, estimated by the concentrations of chlorophyll-a, was highest in Tributary Creek at Site 9 (Figure 28). The small size of this creek, combined with limited scouring flows, is more conducive to the establishment and growth of attached algae. The high flows in Greens Creek (all sites) likely limit the establishment of a dense algal community in the riffle areas that were sampled.

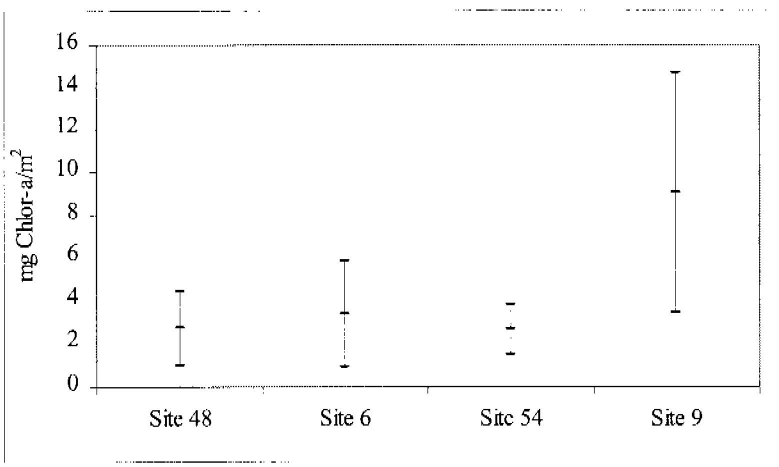

Figure 27. Periphyton biomass, expressed as concentration of chlorophyll-a (mean $\pm$ 1 standard deviation), all sites, 2001.

Benthic Macroinvertebrates

The density of aquatic macroinvertebrates was highest in Greens Creek at Site 54 and all Greens Creek sites had higher densities than Tributary Creek (Figure 29). The taxonomic richness (measured by total number of taxa) also was highest in Greens Creek at Site 54 (Figure 30). Taxonomic richness was similar in Greens Creek at Site 6 and Tributary Creek Site 9, (Figure 31), with Greens Creek Sites 48 and 54 producing higher numbers of taxa.. The proportion of EPT taxa was high at all sites, although Tributary Creek contained fewer EPT organisms (Figure 32). Percent dominant taxa is frequently used to signal a stressed community where sensitive species are reduced or missing and pollution-tolerant species are most common. Ephemeroptera and Ostracoda, both known to be sensitive to many pollutants, were most common in the Greens Creek and Tributary Creek sites. The high proportion of these two taxa, combined with high taxonomic richness at each site (20 to 29 distinct forms) indicates a diverse and complex macroinvertebrate community, not a community that is stressed.

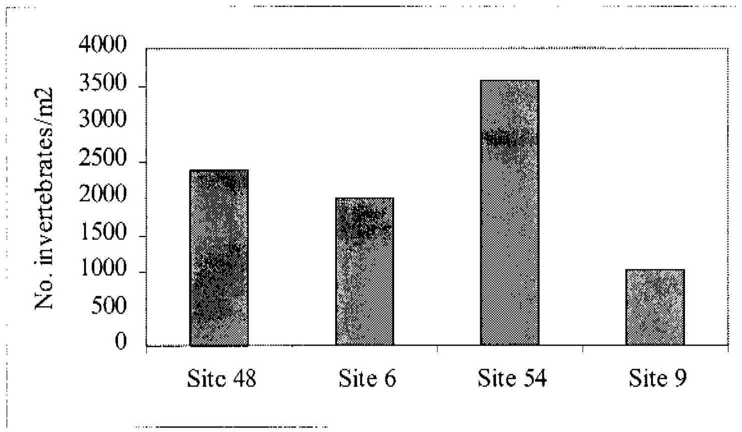

Figure 28. Benthic macroinvertebrate density at each site, 2001.

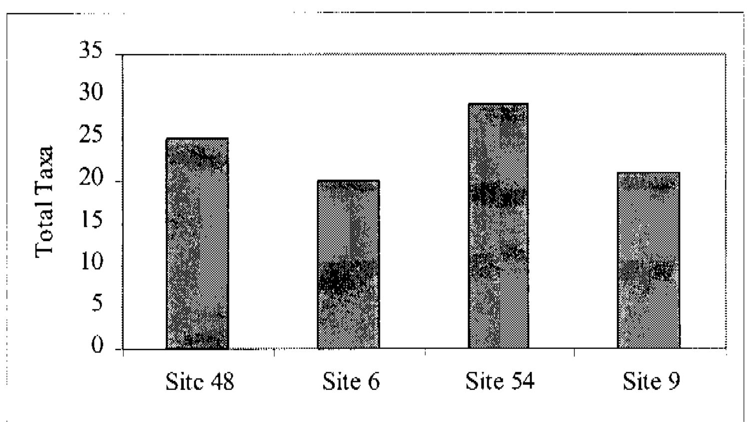

Figure 29. Taxonomic richness, all sites, 2001.

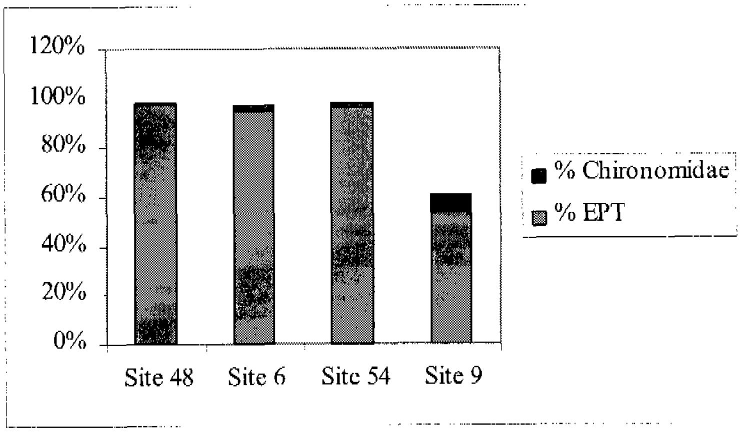

Figure 30. Percent Chironomidae and percent EPT taxa, all sites, 2001.

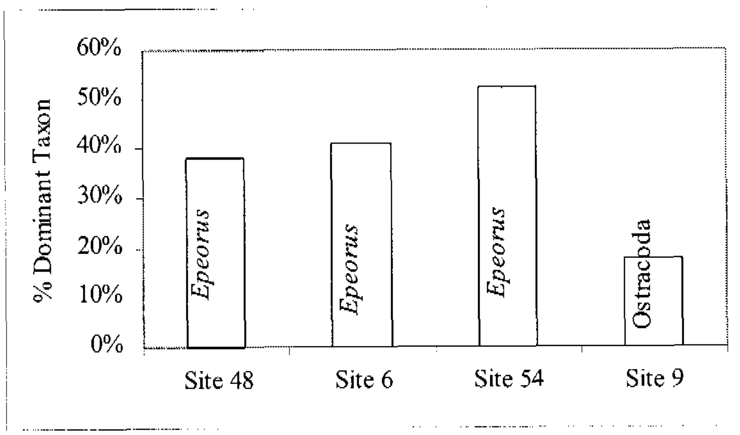

Figure 31. Percent dominant taxon, all sites, 2001. Dominant taxon in Greens Creek at sites 6, 48, and 54 was Heptageniidae: *Epeorus* and in Tributary Creek at Site 9, Ostracoda.

Juvenile Fish Community

Population estimates for Dolly Varden juveniles were made at the three Greens Creek biomonitoring sites (Table 4). Density of fish (estimated from total reach length and average channel width) ranged from 0.13 per m^2 at Greens Creek Site 48 to 0.58 per m^2

at Greens Creek Site 54. The estimated fish density at Greens Creek Site 54 is higher than the average regional density of 0.42 for this channel type (Paustian et. al 1999).

Table 4. Fish population estimates for length of reach sampled. (Data from USFS)

Location	Fish	Pop. Est.	Standard Error	95% confidence interval
Upper Greens Ck (48)	DV	144	74.76	84 --> 448
Middle Greens CK (6)	DV	175	21.67	149 --> 240
Greens Ck below D pond (54)	DV	164	12.32	150 --> 200
Tributary Ck (9)	DV	81	0.78	81 --> 81
Tributary Ck (9)	CO	120	2.14	119 --> 128
Tributary Ck (9)	SC	4	0.21	4 --> 4

Dolly Varden populations appear healthy in all of the Greens Creek sample reaches. Numbers of coho salmon juveniles, however, were lower in Greens Creek in 2001 than expected. Pilot sampling last summer indicated a greater abundance of coho salmon juveniles than were observed this year. The apparent decrease in juvenile coho salmon may be the result of fall flow conditions that restricted movement of adult spawners through the Middle Greens Creek fish pass, or low run strength. Because of the apparently high year-to-year natural variability in juvenile coho salmon populations, we recommend focusing fish population studies on juvenile Dolly Varden.

We found little differences in Dolly Varden length distributions among the Greens Creek biomonitoring sites. However, few Dolly Varden in the larger size classes (greater than 100mm) were observed in Tributary Creek. Their presence in this system is likely limited by the smaller channel and low flows.

For future fish population monitoring, we recommend concentrating the traps in shorter 30 to 40 meter sample reaches at Sites 48 and 6 to increase the confidence level of the population estimates. We further recommend that future samples be compared among years to evaluate possible changes in the length distribution of Dolly Varden.

Metals in Juvenile Fish Tissue

The concentration of Ag in juvenile Dolly Varden was similar among all fish collected in Greens Creek (Sites 48, 6, and 54) and highest in Tributary Creek (Figure 33). Cd was highest in Site 6 and Site 48 samples, concentrations from Site 54 were lower and similar to concentrations from Site 9 (Figure 34). Median Cu concentrations were similar in fish from all sample sites (Figure 35). The median Pb concentration was slightly higher in fish from Site 6 than from the other Greens Creek sites (Figure 36). Fish from Tributary Creek, Site 9, contained two fish with higher Pb concentrations (2.19 and 4.88 mg/kg). These values were checked by the analytical laboratory and confirmed. Concentrations of Se and Zn were similar among all sites (Figures 37 and 38).

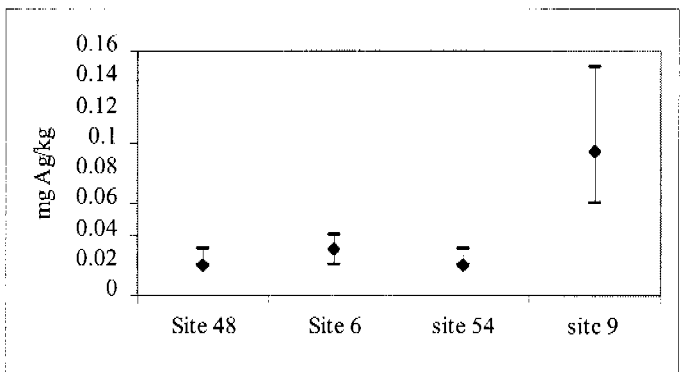

Figure 32. Concentration of Ag in juvenile Dolly Varden, 2001.

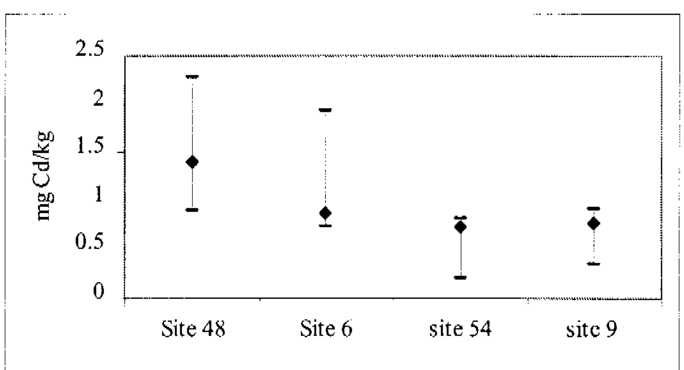

Figure 33. Concentration of Cd in juvenile Dolly Varden, 2001.

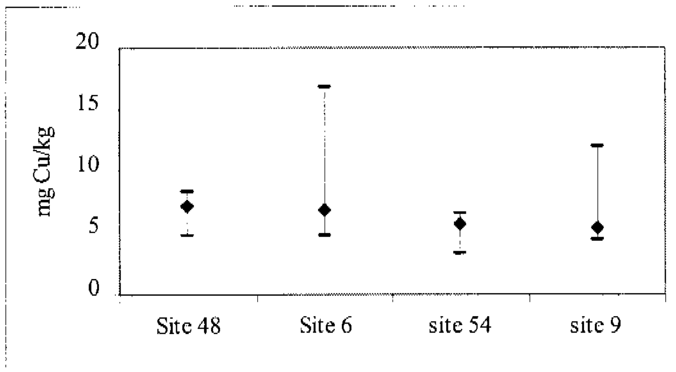

Figure 34. Concentration of Cu in juvenile Dolly Varden, 2001.

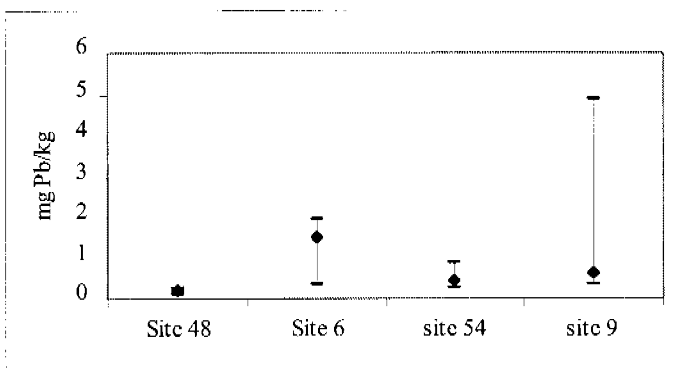

Figure 35. Concentration of Pb in juvenile Dolly Varden, 2001.

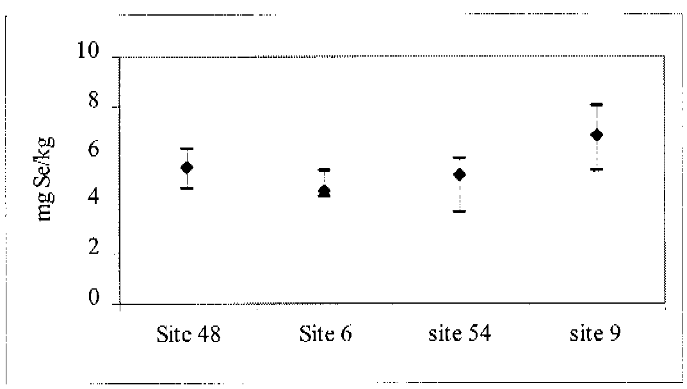

Figure 36. Concentration of Se in juvenile Dolly Varden, 2001.

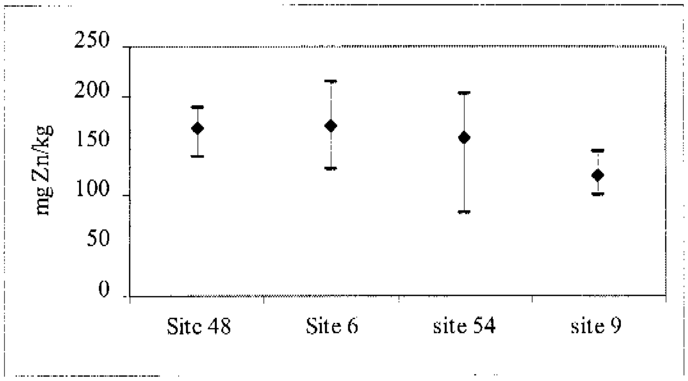

Figure 37. Concentration of Zn in juvenile Dolly Varden, 2001.

Toxicity Testing

We conducted Microtox acute (1-hour) and chronic (24-hour) toxicity tests with water from all sites. None of the tests detected toxicity, and growth of *Vibrio fischeri* was similar to the control for all dilutions. Because there was no toxic response, the IC-20 value for each site is >100%.

Conclusion

Three sites in Greens Creek and the site in Tributary Creek had complex, diverse aquatic macroinvertebrate communities with high densities. The prevalence of Ephemeroptera and Plecoptera are signs of excellent water quality. Periphyton communities (estimated by algal standing crop) are well established in each site, especially in Tributary Creek where stream flows are low and scouring flood events are rare. We found no indication of either chronic or acute toxicity in water from all sites. Fish populations are well established in each site, although populations of coho salmon were lower than expected in the Greens Creek sites. We believe that low numbers may have resulted from high water events in the previous year. Site 48 contained resident Dolly Varden, access by anadromous fish is restricted by a downstream weir. Dolly Varden in Greens Creek at Sites 6 and 54 were a mixture of anadromous and resident fish, and all salmonid fish in Tributary Creek were anadromous.

References

ADF&G. 1998. Methods for aquatic life monitoring to satisfy requirements under NPDES permit. NPDES AK-003865-2, Red Dog Mine Site. AK Dept. of Fish and Game. 23 pp.

Aho, Richard 2000. Monitoring Plan for Determining Trends in Populations and Habitat for Resident Dolly Varden and Cutthroat Trout--February 28, 2000. Unpublished report. Available at: Tongass National Forest Supervisors Office, Petersburg AK. 12 pages.

APHA. 1992. Standard Methods for the Examination of Water and Wastewater. 17th Edition. American Public Health Association, Washington DC

Armstrong, R.H. and J.E. Morrow. 1980. The Dolly Varden charr, Salvelinus malma. Chapter 2 *in* Charrs, Salmonid Fishes of the Genus Salvelinus. Eugene K. Balon (ed.). Dr. W. Junk publ. The Hague, Netherlands.

Azur Environmental. 1999. Microtox Test Manual, 3rd Revision. Azur Environmental Document, available in electronic format by request from the company.

Barbour, M.T., J. Gerritsen, B.D. Snyder, and J.B. Stribling. 1997. Revision to Rapid Bioassessment Protocols For Use in Streams and Rivers: Periphyton, Benthic, Macroinvertebrates, and Fish. EPA-841-D-97-002.

Bryant, M.D. 2000. Estimating Fish Populations by Removal Methods with Minnow Traps in Southeast Alaska Streams. North American Journal of Fisheries Management: 20: 923-930.

Crawford, J.K. and S.N. Luoma. 1993. Guidelines for studies of contaminants in biological tissues for the National Water-Quality Assessment Program. U.S. Geological Survey Open File Report 92-494. Lemoyne, Pa. 69 pp.

DeCicco, A. 2002. Alaska Department of Fish and Game, Char Fish Biologist. Pers. Comm.

McCafferty, W.P. 1998. Aquatic Entomology, the fishermen's and ecologists' illustrated guide to insects and their relatives. Jones and Bartlett Publ., Boston. 448 pp.

Morrow, J.E. 1980. The freshwater fishes of Alaska. Alaska Northwest Publishing Company. Anchorage. AK. 248 pp.

Paustian S.J., Murphy M.L., Kessler S.J., and V.J. Starostka. 1990. Coho Salmon and Dolly Varden Char Habitat Capability for the Tongass National For the Tongass National Forest, Alaska. Prepared for Tongass National Forest Management Plan

Revision , Analysis of the Management Situation (R10-MB-89). Sitka AK. 14 pages.

U. S. EPA. 1997. Method 446.0. In Vitro Determination of Chlorophylls a, b, c1 + c2 and Pheopigments in Marine and Freshwater Algae by Visible Spectrophotometry. Adapted by Elizabeth J. Arar. Revision 1.2, September 1997. National Exposure Research Laboratory, Office of Research and Development, U. S. Environmental Protection Agency, Cincinnati, Ohio."

Weber Scannell, P. and A.G. Ott. 2001. Aquatic biomonitoring at Red Dog Mine, 2000. National Pollution Discharge Elimination System Permit No. AK-003865-2. Technical Report No. 01-04. AK Dept. of Fish and Game, Habitat and Restoration Division. 163 pp.

Appendix 1. Macroinvertebrate Data

Tributary Creek, Site 9, 2001.

			Total Collected
Insecta			
Ephemeroptera	Baetidae	*Baetis*	41
		Procloeon	5
	Ephemerellidae	*Caudatella*	3
	Heptageniidae	*Cinygma*	1
		Cinygmula	89
	Leptophlebiidae	*Paraleptophlebia*	66
	Ameletidae	*Ameletus*	
Plecoptera	Capniidae	*Suwallia*	34
	Leuctridae	*Despaxia*	3
		Paraleuctra	7
	Nemouridae	*Zapada*	23
	Perlodidae	*Isoperla*	1
Coleoptera	Elmidae	*Narpus*	2
Diptera	Ceratopogonidae	*Probezzia*	3
	Chironomidae		35
	Empididae	*Oreogeton*	4
	Psychodidae	*Psychoda*	
	Simuliidae	*Simulium*	40
	Tipulidae	*Tipula*	4
Miscellaneous			
Acarina			15
Oligochaeta			40
Gastropoda			1
Ostracoda			92

Summary of macroinvertebrate data, Tributary Creek, Site 9, 2001.

Abundance	
Average aq. invertebrates/m2	1018.00
% Sample aquatic	100.0%
% Sample terrestrial	0.0%
Community	
Total aquatic taxa	21
Tot. Ephemeroptera	41
Tot. Plecoptera	14
Tot. Trichop.	0
Total Aq. Diptera	17
Misc.Aq.sp	30
% other	25.7%
% Ephemeroptera	42.5%
% Plecoptera	14.8%
% Trichoptera	0.0%
% Aq. Diptera	17.0%
Total Chironomidae	
% EPT	53.6%
% Chironomidae	6.9%
Dominant taxon	
% Dominant Taxon	26.1%

Greens Creek below Pond D, 2001.

Insecta			Total Counted
Ephemeroptera	Baetidae	*Baetis*	248
	Ephemercllidae	*Ephemerella*	2
		Drunella #1	100
		Drunella #2	18
	Heptageniidae	*Cinygmula*	319
		Epeorus	935
	Leptophlebiidae	*Paraleptophlebia*	1
	Amcletidae	*Ameletus*	4
Plecoptera	Capniidae	*Capnia*	3
	Chloroperlidae	*Paraperla*	2
		Suwallia	6
	Leuctridae	*Paraleuctra*	13
	Nemouridae	*Zapada*	52
	Perlodidae	*Diura*	1
		Isoperla	3
Trichoptera	Limnephilidae	*Psychoglypha*	1
	Rhyacophilidae	*Rhyacophila*	6
Coleoptera	Staphylinidae		1
Diptera	Chironomidae		33
	Dolichopodidae		2
	Empididae	*Chelifera*	2
		Oreogeton	10
	Simuliidae	*Simulium*	3
	Tipulidae	*Antocha*	1
		Dicranota	2
		Tipula	
Miscellaneous			
Acarina			9
Oligochaeta			3
Gastropoda			1
Ostracoda			1

Summary of macroinvertebrate samples, Greens Creek, Site 54, 2001.

Abundance	
Average aq. invertebrates/m^2	3564.00
% Sample aquatic	100.0%
% Sample terrestrial	0.0%
Community	
Total aquatic taxa	30
Tot. Ephemeroptera	325
Tot. Plecoptera	16
Tot. Trichop.	1
Total Aq. Diptera	11
Misc.Aq.sp	3
% other	0.9%
% Ephemeroptera	91.5%
% Plecoptera	4.4%
% Trichoptera	0.4%
% Aq. Diptera	2.9%
% EPT	96.2%
% Chironomidae	1.9%
% Dominant Taxon	52.5%

Invertebrates collected from Upper Greens Creek, Site 48, 2001.

Insecta			Total Counted
Ephemeroptera	Baetidae	*Baetis*	309
	Ephemerellidae	*Caudatella*	2
		Drunella #1	47
	Heptageniidae	*Cinygmula*	99
		Epeorus	444
		Rhithrogena	193
Plecoptera			
	Chloroperlidae	*Alloperla*	1
		Plumiperla	5
		Suwallia	8
		Sweltsa	1
	Leuctridae	*Paraleuctra*	4
	Nemouridae	*Podmosta*	7
		Zapada	23
Trichoptera	Hydropsychidae	*Arctopsyche*	2
	Rhyacophilidae	*Rhyacophila*	5
Coleoptera	Staphylinidae		1
Diptera	Chironomidae		14
	Deuterophlebiidae	*Deuterophlebia*	2
	Empididae	*Chelifera*	1
		Oreogeton	3
	Psychodidae	*Psychoda*	1
	Simuliidae	*Parasimulium*	2
		Prosimulium	2
		Simulium	6
Miscellaneous			
Acarina			2

Summary of macroinvertebrate samples, Site 48, 2001.

Abundance	
Average aq. invertebrates/m2	2368.00
% Sample aquatic	100.0%
% Sample terrestrial	0.0%
Community	
Total aquatic taxa	25.0
Tot. Ephemeroptera	219
Tot. Plecoptera	10
Tot. Trichop.	1
Total Aq. Diptera	6
Misc.Aq.sp	1
% other	0.4%
% Ephemeroptera	93.1%
% Plecoptera	3.3%
% Trichoptera	0.5%
% Aq. Diptera	2.7%
% EPT	97.1%
% Chironomidae	1.2%
% Dominant Taxon	41.0%

Invertebrates collected in Green Creek, Site 6, 2001.

Insecta			Total Counted
Ephemeroptera	Baetidae	*Baetis*	153
	Ephemerellidae	*Drunella #1*	45
		Drunella #2	7
	Heptageniidae	*Cinygmula*	303
		Epeorus	408
Plecoptera	Chloroperlidae	*Suwallia*	2
	Leuctridae	*Paraleuctra*	7
	Nemouridae	*Zapada*	16
	Perlodidae	*Isoperla*	7
Trichoptera	Rhyacophilidae	*Rhyacophila*	1
Coleoptera	Staphylinidae		1
Diptera	Chironomidae		19
	Deuterophlebiidae	*Deuterophlebia*	1
	Dolichopodidae		1
	Empididae	*Chelifera*	1
		Oreogeton	3
Miscellaneous			
Arachnida			1
Acarina			4
Oligochaeta			15
Ostracoda			3

Summary of macroinvertebrate samples, Greens Creek, Site 6, 2001.

Abundance

Average aq. invertebrates/m2 water	1996.00
% Sample aquatic	100.0%
% Sample terrestrial	0.0%

Community

Total aquatic taxa	20
Tot. Ephemeroptera	183
Tot. Plecoptera	6
Tot. Trichop.	0
Total Aq. Diptera	5
Misc.Aq.sp	5
% other	3.1%
% Ephemeroptera	91.6%
% Plecoptera	2.5%
% Trichoptera	0.2%
% Aq. Diptera	2.6%
% EPT	95.1%
% Chironomidae	1.9%
% Dominant Taxon	52.9%

Appendix 2. Juvenile Fish Tissue Data.

Sample catalogue number, date collected, location, site number, species, weight, and length.

Sample Number	Date, collected	Creek	Site	Fish spp.	Weight, grams	Length, mm (fl)
062100GCCOJ01	21-Jun-00	Greens Cr.	Site 54	Coho	4.4	72
062100GCCOJ02	21-Jun-00	Greens Cr.	Site 54	Coho	6.1	82
062100GCCOJ03	21-Jun-00	Greens Cr.	Site 54	Coho	4.9	73
062100GCCOJ04	21-Jun-00	Greens Cr.	Site 54	Coho	3.4	68
062100GCCOJ05	21-Jun-00	Greens Cr.	Site 54	Coho	5.9	73
062100GCCOJ06	21-Jun-00	Greens Cr.	Site 54	Coho	6	75
072301GC54DVJ01	23-Jul-01	Greens Cr.	Site 54	DV	121	21.5
072301GC54DVJ02	23-Jul-01	Greens Cr.	Site 54	DV	119	19.32
072301GC54DVJ03	23-Jul-01	Greens Cr.	Site 54	DV	107	15.73
072301GC54DVJ04	23-Jul-01	Greens Cr.	Site 54	DV	109	13.64
072301GC54DVJ05	23-Jul-01	Greens Cr.	Site 54	DV	105	13.52
072301GC54DVJ06	23-Jul-01	Greens Cr.	Site 54	DV	138	27.54
062100TRCOJ01	21-Jun-00	Tributary Cr.	Site 9	Coho	9.7	102
062100TRCOJ02	21-Jun-00	Tributary Cr.	Site 9	Coho	5.3	75
062100TRCOJ03	21-Jun-00	Tributary Cr.	Site 9	DV	12.8	112
062100TRDVJ04	21-Jun-00	Tributary Cr.	Site 9	DV	13.8	105
062100TRDVJ05	21-Jun-00	Tributary Cr.	Site 9	DV	13.4	105
062100TRDVJ06	21-Jun-00	Tributary Cr.	Site 9	DV	11.3	100

Sample Number	Datc, collected	Creek	Site	Fish spp.	Weight, grams
072301TR09DVJ01	23-Jul-01	Tributary Cr.	Site 9	DV	97
072301TR09DVJ02	23-Jul-01	Tributary Cr.	Site 9	DV	97
072301TR09DVJ03	23-Jul-01	Tributary Cr.	Site 9	DV	97
072301TR09DVJ04	23-Jul-01	Tributary Cr.	Site 9	DV	98
072301TR09DVJ05	23-Jul-01	Tributary Cr.	Site 9	DV	86
072301TR09DVJ06	23-Jul-01	Tributary Cr.	Site 9	DV	93
072301GC06DVJ01	23-Jul-01	Greens Cr.	Site 6	DV	139
072301GC06DVJ02	23-Jul-01	Greens Cr.	Site 6	DV	140
072301GC06DVJ03	23-Jul-01	Greens Cr.	Site 6	DV	167
072301GC06DVJ04	23-Jul-01	Greens Cr.	Site 6	DV	155
072301GC06DVJ05	23-Jul-01	Greens Cr.	Site 6	DV	109
072301GC06DVJ06	23-Jul-01	Greens Cr.	Site 6	DV	168
072301GC48DVJ01	23-Jul-01	Greens Cr.	Site 48	DV	131
072301GC48DVJ02	23-Jul-01	Greens Cr.	Site 48	DV	137
072301GC48DVJ03	23-Jul-01	Greens Cr.	Site 48	DV	119
072301GC48DVJ04	23-Jul-01	Greens Cr.	Site 48	DV	121
072301GC48DVJ05	23-Jul-01	Greens Cr.	Site 48	DV	111
072301GC48DVJ06	23-Jul-01	Greens Cr.	Site 48	DV	121

Concentration of select elements in juvenile fish.

Sample Number	Ag	Cd	Cu	Pb	Se	Zn	%
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	Solids
	0.02(MRL)	0.02(MRL)	0.1(MRL)	0.02(MRL)	1(MRL)	0.5(MRL)	
062100GCCOJ01	0.04	0.95	15.3	1.4	4.9	251	20.5
062100GCCOJ02	0.09	0.66	11.7	1.21	4.7	224	20.2
062100GCCOJ03	0.22	1.07	24.2	1.4	3.4	206	20.4
062100GCCOJ04	0.1	0.97	24	1.12	3.5	181	21.4
062100GCCOJ05	0.05	0.96	44	1.53	4.9	304	20.7
062100GCCOJ06	0.08	1.47	36.1	5.02	4.7	340	20.2
072301GC54DVJ01	0.03	0.46	4.3	0.33	5.7	126	22.6
072301GC54DVJ02	0.02	0.21	3.2	0.22	3.6	82	26.1
072301GC54DVJ03	0.03	0.73	6.3	0.59	4.7	144	23.5
072301GC54DVJ04	0.02	0.82	5.4	0.86	4.9	172	21.1
072301GC54DVJ05	<0.02	0.79	6.5	0.45	5.8	203	22.8
072301GC54DVJ06	<0.02	0.74	5.8	0.4	5.4	171	22.1
062100TRCOJ01	0.04	0.42	16.2	1.03	3.2	213	22.9
062100TRCOJ02	0.07	0.5	16.5	2.01	3.7	220	22.5
062100TRCOJ03	0.12	0.75	11.2	1.63	3.8	194	23.1
062100TRDVJ04	0.07	0.56	10.6	1.53	3.6	87.9	22.2
062100TRDVJ05	0.06	0.58	12.8	1.59	3.5	204	22.1
062100TRDVJ06	0.05	0.45	32.8	1.57	5	213	23
072301TR09DVJ01	0.09	0.35	4.3	0.56	6.8	127	22.1
072301TR09DVJ02	0.1	0.77	5.2	0.67	8	118	21.3
072301TR09DVJ03	0.15	0.92	5.4	4.88	5.3	144	22.2
072301TR09DVJ04	0.15	0.86	6.7	2.19		99.1	22.6
072301TR09DVJ05	0.08	0.76	4.9	0.33	6.2	106	22.2
072301TR09DVJ06	0.06	0.37	12	0.38	6.8	122	20.6

Sample Number	Ag	Cd	Cu	Pb	Se	Zn	%
	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	mg/kg	Solids
	0.02(MRL)	0.02(MRL)	0.1(MRL)	0.02(MRL)	1(MRL)	0.5(MRL)	
072301GC06DVJ01	0.04	1.94	16.7	1.24	5	173	20.8
072301GC06DVJ02	0.03	0.84	4.6	1	4.5	167	22.8
072301GC06DVJ03	0.03	0.82	5.3	1.94	4.3	171	21.7
072301GC06DVJ04	0.03	1.52	5.4	1.78	4.5	215	21.6
072301GC06DVJ05	0.02	0.89	11.1	0.33	5.3	126	22.2
072301GC06DVJ06	0.04	0.73	8	1.96	4.6	169	21.9
072301GC48DVJ01	0.02	1.76	8.3	0.2	6.1	180	21.6
072301GC48DVJ02	0.03	0.89	7.2	0.17	4.6	146	23.7
072301GC48DVJ03	0.02	2.27	5.7	0.2	6.2	189	20.7
072301GC48DVJ04	0.02	1.56	6.9	0.17	5.2	182	22.8
072301GC48DVJ05	0.03	0.89	4.7	0.23	5.4	138	21.8
072301GC48DVJ06	<0.02	1.26	7.4	0.1	5.6	157	20.3