Impacts of acid mine drainage on juvenile salmonids in an estuary near Britannia Beach in Howe Sound, British Columbia

Karen L. Barry, Jeffrey A. Grout, Colin D. Levings, Bruce H. Nidle, and G. Elizabeth Piercey

Abstract: The abandoned copper mine at Britannia Beach, British Columbia, has been releasing acid mine drainage (AMD) into Howe Sound for many years. To assess the impacts of AMD on juvenile salmonids in the Britannia Creek estuary, we compared fish abundance, distribution, and survival at contaminated sites near the creek with uncontaminated areas in Howe Sound. Water quality near Britannia Creek was poor, particularly in spring when dissolved Cu exceeded 1.0 $mg \cdot L^{-1}$ and pH was less than 6. Beach seine surveys conducted during April–August 1997 and March–May 1998 showed that chum salmon (*Oncorhynchus keta*) fry abundance was significantly lower near Britannia Creek mouth ($0–1.2 \cdot 100\ m^{-2}$) than in reference areas ($11.5–31.4 \cdot 100\ m^{-2}$). Laboratory bioassays confirmed that AMD from Britannia Mine was toxic to juvenile chinook (*Oncorhynchus tshawytscha*) and chum salmon (96-h LC_{50} = 0.7–29.7% in freshwater and 12.6–62.2% in 10 ppt water). Chinook salmon smolts transplanted to surface cages near Britannia Creek experienced 100% mortality within 2 days. These results demonstrated that juvenile salmonids are vulnerable to AMD from Britannia Creek: their abundance peaks during spring when Cu concentrations are highest and toxicity is greatest in surface freshwater, which matches their preferred vertical distribution.

Résumé : La mine de cuivre abandonnée de Britannia Beach, en Colombie-Britannique, rejette depuis des années du drainage minier acide (DMA) dans la baie Howe. Pour évaluer l'impact du DMA sur les salmonidés juvéniles dans l'estuaire du crique Britannia, nous avons comparé l'abondance, la distribution et la survie des poissons capturés sur des sites contaminés proches du crique à celles observées dans des zones non polluées de la baie Howe. Près du crique Britannia, la qualité de l'eau était mauvaise, particulièrement au printemps, le cuivre dissous dépassant alors 1,0 $mg \cdot L^{-1}$, et le pH étant inférieur à 6. Des relevés à la senne de plage effectués d'avril à août 1997 et de mars à mai 1998 ont révélé que l'abondance des alevins de kéta (*Oncorhynchus keta*) était nettement plus basse près de l'embouchure du crique Britannia ($0–1{,}2 \cdot 100\ m^{-2}$) que dans les zones témoins ($11{,}5–31{,}4 \cdot 100\ m^{-2}$). Des bioessais menés en laboratoire ont confirmé que le DMA de la mine Britannia était toxique pour les juvéniles de quinnat (*O. tshawytscha*) et de kéta (CL_{50} à 96 h = 0,7–29,7% en eau douce; 12,6–62,2% dans de l'eau à 10 ppt). Les smolts de quinnat installés dans des cages de surface près du crique Britannia ont présenté une mortalité de 100% dans l'espace de deux jours. Ces résultats ont démontré que les salmonidés juvéniles sont vulnérables au DMA du crique Britannia, car leur abondance connaît un maximum au printemps, moment où les concentrations de cuivre sont les plus hautes et où la toxicité est la plus forte dans l'eau douce de surface, ce qui correspond à leur distribution verticale préférée.

[Traduit par la Rédaction]

Introduction

The abandoned Britannia copper mine, located in the community of Britannia Beach approximately 50 km north of Vancouver, operated from 1902 until 1974. During operation, most mining activity took place underground, but several surface pits were also excavated. In addition to mine tailings disposed of in Howe Sound, another byproduct of mining has been the continued production of acid mine drainage (AMD), which is characterized by low pH and high levels of certain metals. AMD results when sulphide- containing rocks are exposed to air and undergo an oxidation reaction, often accelerated by the bacterium *Ferrobacillus ferrooxidans*. This produces sulphuric acid, which then dis-

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solves metal sulphides from the rock, and these metal ions are transported by water passing through the mine. An extremely large flow of AMD can be generated from the Britannia Mine because the surface pits trap rain and snow, funneling large amounts of water through the extensive system (80 km) of underground tunnels (McCandless 1995).

Britannia Mine is considered to be the worst point source of pollution of any mine in British Columbia (McCandless 1995), and AMD from Britannia Mine is the largest source of dissolved metals into Howe Sound (Dunn et al. 1992). The predominant metal discharged into Howe Sound from Britannia Creek is Cu (R. McCandless, Environment Canada, 224 Esplanade, North Vancouver, BC V7M 3H7, Canada, personal communication). In 1996, total Cu loadings from Britannia Creek ranged from lows of 10 $kg \cdot day^{-1}$ in summer to maximum loadings of 425 $kg \cdot day^{-1}$ in spring due to snowmelt. Zinc, the second most abundant metal discharged from Britannia Creek, had loading rates of 10–160 $kg \cdot day^{-1}$, while Cd loadings ranged from 0.2 to 1.4 $kg \cdot day^{-1}$. Britannia Creek water can be extremely acidic, especially in spring. For example, from 4 to 27 May 1998, surface pH ranged from 4.6 to 5.1 approximately 50 m upstream from the creek mouth (Grout et al. 1999).

There are two sources of AMD to the Britannia Creek estuary: (*i*) the 4100 portal (an entrance to the mine located 4100 ft from the top of the mountain, 67 m above sea level), which transports AMD via a pipeline, releasing it directly into the Sound from an outfall at 30 m in depth approximately 70 m from shore, and (*ii*) the 2200 portal (700 m above sea level), which drains directly into Britannia Creek and then into Howe Sound. Both point sources have the potential to contaminate the foreshore of Britannia Creek estuary, but because the 4100 portal discharge tends to remain trapped below the pycnocline during the summer (Chretien 1997), the most important source of contamination to the nearshore zone is likely to be AMD from Britannia Creek. Hence, AMD from this source was the focus of our study.

Although AMD from Britannia Mine has been recognized as a potentially serious hazard to aquatic organisms (Van Aggelen and Moore 1986; McCandless 1995), no studies have investigated the effects of AMD on salmonids and their habitats in Howe Sound. Previous research has found elevated concentrations of Cu and Zn in water, sediment, algae, and invertebrates near Britannia Beach (Dunn et al. 1992). Copper is an essential element that occurs naturally in the environment, yet it can be highly toxic to fish in freshwater at relatively low concentrations, i.e., 1–10 $\mu g \cdot L^{-1}$ (Eisler 1997). Effects of Cu on chum salmon (*Oncorhynchus keta*) fry have been studied in mesocosm experiments in Saanich Inlet, British Columbia (Thompson and Paton 1976), but there are no published field studies of the effects of Cu on salmonids in estuaries. There are several reports of other mines discharging high levels of dissolved Cu into some salmon-bearing estuaries (e.g., Foster et al. 1978; Featherstone and O'Grady 1997), but the biological effects have not been investigated, even though estuaries are key rearing grounds for many salmonid species.

The foreshore areas of Howe Sound provide important habitat for juvenile chum salmon and chinook salmon (*Oncorhynchus tshawytscha*) as they migrate to the Pacific Ocean (Levings and Riddell 1992). Estuaries are essential rearing grounds because they provide productive feeding habitat with reduced risk of predation where young salmon can begin the process of seawater adaptation (e.g., Iwata and Komatsu 1984; Macdonald et al. 1988; Levings 1994). During smoltification, juvenile salmon shift from freshwater to salt water habitats, which involves numerous morphological and biochemical changes, often associated with increased energetic demands and high metabolic rates (Healey 1982). Thus, juvenile chum and chinook salmon may be particularly sensitive to AMD from Britannia Creek during estuarine residency because this is a critical transition period when young salmon are physiologically stressed. Previous research has shown that exposure to sublethal Cu concentrations impairs seawater adaptation and reduces the ability of juvenile salmon to migrate successfully (Lorz and McPherson 1976). In addition, exposure to sublethal levels of Cu can cause many other effects on young salmon such as reduced swimming performance, lower growth rates, impaired sensory mechanisms, and reduced immunity (e.g., Lorz and McPherson 1976; Wilson and Taylor 1993).

We hypothesized that juvenile salmonids would be vulnerable to the toxic effects of AMD from Britannia Creek because salmon fry are most abundant during spring when Cu levels are high and also because AMD is concentrated in surface freshwater that bathes the nearshore habitat in contaminated water. To evaluate the impacts of AMD on juvenile salmonids, we compared differences in fish abundance, distribution, and survival at contaminated sites near the mouth of Britannia Creek and lesser impacted areas further away. We used a unique approach combining laboratory tests with in situ field measurements to assess the biological impact of AMD on juvenile salmonids in estuaries.

Methods

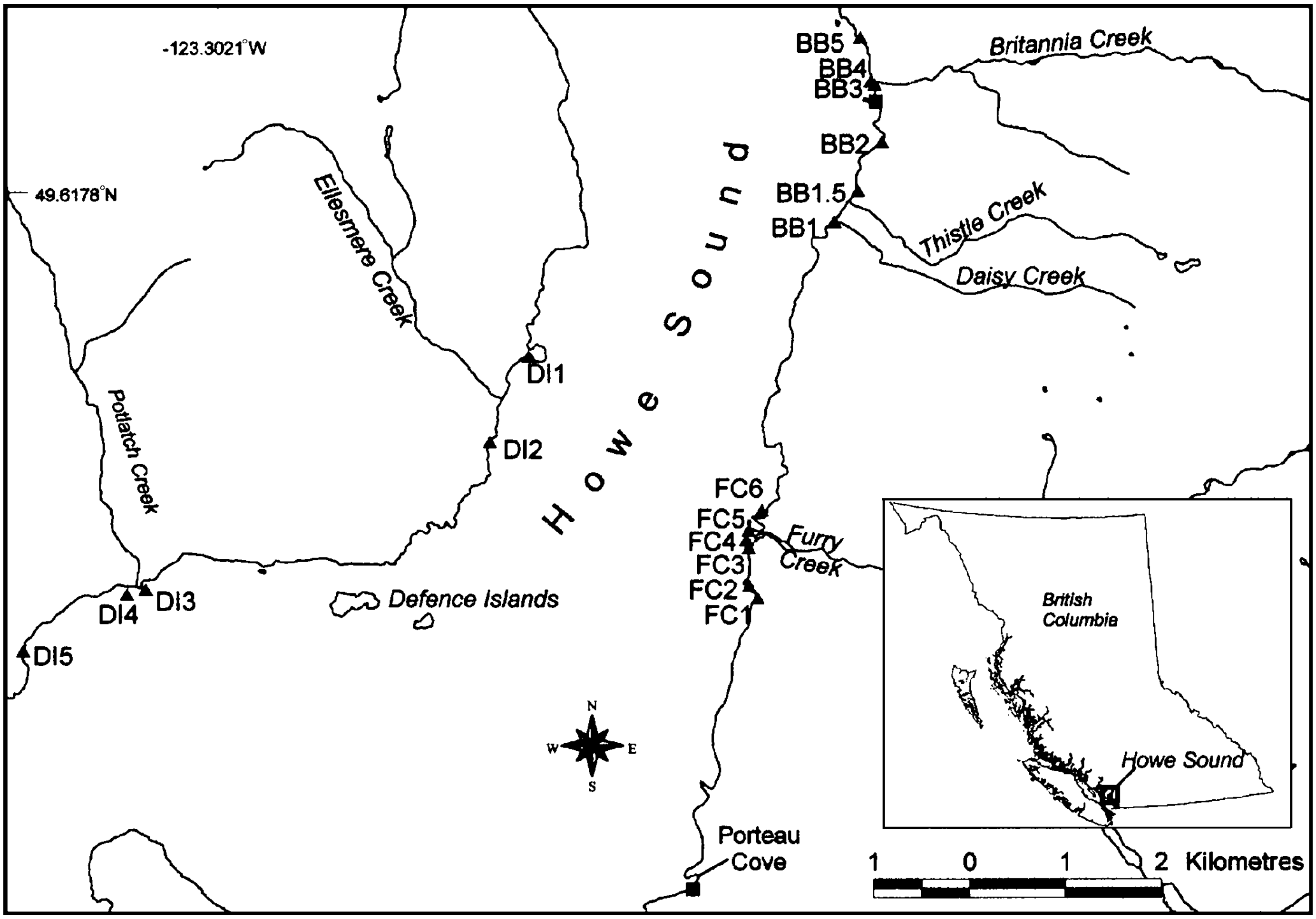
Study area

Several small creeks drain into Howe Sound, but the largest freshwater input is from the Squamish River (mean annual discharge 273 $m^3 \cdot s^{-1}$) located at the north end of the Sound. The Squamish River supports stocks of chum salmon, chinook salmon, coho salmon (*Oncorhynchus kisutch*), pink salmon (*Oncorhynchus gorbuscha*), and steelhead trout (*Oncorhynchus mykiss*). Chum salmon are known to occur in several creeks in Howe Sound, but fewer than 100 fish commonly spawn in these smaller creeks (Levings and Riddell 1992).

The surface circulation patterns of Howe Sound are driven by a major outflow of freshwater from the Squamish River during the spring freshet. In 1997, maximum flow exceeded 500 $m^3 \cdot s^{-1}$ between 15 May and 22 July (L. Campo, Water Survey of Canada, 120-1200 W. 73 Ave., Vancouver, BC V6P 6H9, Canada, personal communication). The freshet causes pronounced thermal and salinity stratification in upper Howe Sound during the spring and summer months, June–September (Thomson 1981).

Britannia Creek, located approximately 10 km south of the mouth of the Squamish River (Fig. 1), is roughly 9 km long with a watershed area of 28.5 km^2 and a mean discharge of 8.2 $m^3 \cdot s^{-1}$. Seasonal freshets occur during spring from snowmelt and during fall and winter from heavy rainfall. Estuarine conditions (i.e., low surface salinity) exist at the mouth of Britannia Creek and are most pronounced during periods when the Squamish River has low flow. A counterclockwise gyre frequently occurs near Britannia Beach (BB) (Thomson 1981). Sampling sites were positioned near the

Fig. 1. Locations of sampling sites in Howe Sound at the study area, Britannia Beach (BB), and the reference areas, Furry Creek (FC), Defence Islands (DI), and Porteau Cove. Symbols represent water quality and beach seining sites (▲) and locations used in the caged salmon experiment (■). BB1 was located approximately 1600 m south of the mouth of Britannia Creek, BB2 was 630 m south, BB3 was 25 m south, BB4 was 25 m north, and BB5 was 500 m north of the creek mouth.



creek mouth, with BB1, BB2, and BB3 south of the creek mouth and BB4 and BB5 north of the creek mouth (Fig. 1).

The principal reference area for this study was the Furry Creek estuary located 6 km south of Britannia Creek. Furry Creek is similar to Britannia Creek in size (12 km in length, 54-km^2 watershed) and flow patterns. Sampling sites were located as at Britannia Creek, with FC1, FC2, and FC3 south of the creek mouth and FC4, FC5, and FC6 north of the creek mouth. Additional reference areas included Defence Islands (DI), located about 8 km southwest of Britannia Beach, and Porteau Cove (PC), located approximately 3 km south of Furry Creek.

Water quality at Britannia Creek estuary and reference areas

Temperature, salinity, pH, and dissolved Cu were measured in surface water at Britannia Creek estuary (BB1–BB5) and reference areas (FC1–FC6, DI1–DI5) approximately every other week (4 April – 18 August 1997, 20 March – 27 May 1998). Temperature was measured with a hand thermometer, salinity with a refractometer (Argent Chemical Laboratories), and pH with an Oakton WD-35615 series pH/mV/temperature meter.

To measure surface dissolved Cu concentrations, 100 mL of water was collected with a syringe, filtered through a 0.45-μm sodium acetate Sartorius filter, and preserved with 1 mL of analytical-grade concentrated nitric acid. Periodically, samples of deionized water and nitric acid were also submitted for chemical analysis to ensure quality assurance/quality control. Copper concentrations were measured using graphite furnace – atomic absorption spectrometry, which has low detection limits for Cu (0.005 mg·L^{-1}) in estuarine water. At each site, three replicate samples were obtained in 1997, but only one sample was obtained in 1998.

Nearshore fish sampling

Abundance and distribution of juvenile salmonids were assessed with biweekly beach seine surveys using a 15 × 2.5 m beach seine (0.3-cm mesh). In 1997, sampling took place from 4 April to 18 August at Britannia Beach, Furry Creek, and Defence Islands and the net was deployed from shore. In 1998, sampling was conducted at Britannia Beach sites and two sites at Furry Creek (FC1, FC4) during the months corresponding to salmon fry out-migration (20 March – 27 May) and the net was set using a boat. At each site, three sets were completed. All captured fish were enumerated by species and held in separate buckets until the final set was completed at each site to prevent recapture. The area seined was visually estimated and catch per unit effort (CPUE) was calculated as the number of fish per 100 m^2.

Caged chinook salmon experiment

Chinook salmon smolts, originating from the Squamish River, were obtained from a net pen rearing facility operated by the Tenderfoot Hatchery at Porteau Cove. Fish were transported to experimental cages located 30 m southwest of the mouth of Britannia Creek and to the reference site at Porteau Cove. All fish were subject to the same transportation process to control for handling and transport stress. After transport to each cage site, fish appeared to be in good condition and no mortalities were observed.

Three cages (60 × 40 × 90 cm) constructed of a stainless steel frame covered with 1-cm nylon mesh were deployed at each site. The first transplant experiment took place from 13 to 19 May 1998. In this experiment, all cages were deployed at the surface. A second experiment was completed from 20 May to 4 June 1998 in which one cage was suspended at the surface (0 m), another at 2 m depth, and a third at 3 m depth at each site. During both experiments, 25 fish were placed into each cage for a total of 150 fish per test. The fish were not fed for the duration of the study. Cages were checked for mortalities every second day. Surface water quality (temperature, salinity, pH, dissolved metals) was assessed as previously described.

Laboratory bioassays

Bioassay water was collected from Britannia Creek, 200 m upstream from the creek mouth (above the salt wedge intrusion zone), and from the 4100 portal. Surface water samples were collected during May, June, and July 1997 and February 1998. Prior to each bioassay, the concentration of dissolved metals was measured in three replicate samples using ICP methods (inductively coupled argon plasma atomic emission spectrometry).

Bioassays were conducted with juvenile chum and chinook salmon obtained from the Tenderfoot Hatchery in Squamish, B.C. Chum bioassays took place during May, June, and July 1997, while chinook bioassays were conducted in February 1998. All tests were completed in environmentally controlled rooms (15°C, 16 h light : 8 h dark photoperiod). Chum toxicity tests were run at 0, 5, and 10 ppt by adjusting the water with 90 ppt hypersaline brine derived from seawater. Chinook salmon toxicity tests were run at 0 ppt. The pH of the water was adjusted to between 7 and 8.

Fish were exposed to various concentrations of Britannia Creek or 4100 portal water diluted with sand-filtered seawater. For each concentration, 10 juvenile fish were placed in 40-L glass aquaria. The concentration causing 50% lethality (LC_{50}) in 96 h was determined as a percent concentration of the original sample water. Acute toxicity was also expressed as the concentration of Cu resulting in 50% mortality by multiplying the original Cu concentration by the 96-h LC_{50} (percent).

Statistical analyses

Because previous studies showed that Cu concentrations were extremely high near the mouth of Britannia Creek and lower elsewhere in Howe Sound (Van Aggelen and Moore 1986; Dunn et al. 1992), we hypothesized that a two-tiered effect level may exist. That is, there may be a "nearfield" effect as well as a "farfield" effect. The statistics used in this study were designed to assess any nearfield effect by testing results among sites at Britannia Beach. To assess the farfield effect, results from the most contaminated sites near the creek mouth (i.e., BB3, BB4) were compared with the results at the more distant reference areas.

All statistical tests were conducted using SYSTAT (version 7.0) at a significance level of 0.05. Data were first tested for normality with a normal probability plot of residuals, and homogeneity of variance was verified with Levene's test. One-way analyses of variance (F) were used to assess whether any significant differences existed in environmental conditions (pH, dissolved Cu) among Britannia Beach sites. Tukey pairwise tests were used to identify which sites were significantly different. To compare environmental conditions at the creek mouth (BB3, BB4) with pooled data from the reference sites, t tests were conducted.

Chum salmon abundance data violated the homogeneity of variance assumption; therefore, these data were $\log(x + 1)$ transformed prior to analyses. The nonparametric Kruskal–Wallis test (H) was used to assess whether any significant differences existed among Britannia Beach sites followed by Tukey pairwise comparisons. To compare abundance at the creek mouth (BB3, BB4) with pooled data from the reference sites, nonparametric Mann–Whitney tests (U) were used.

Results

Water quality at Britannia Creek estuary and reference areas

Temporal patterns in temperature and salinity were largely controlled by the seasonal discharge patterns of the Squamish River. In 1997, spring freshet from the Squamish River occurred over a broad period, from mid-May to late July. Surface water temperature and salinity were similar between Britannia Beach sites and the reference areas. Mean temperature was higher during the freshet than during the prefreshet period (Fig. 2*a*). Before freshet, temperatures ranged from 7 to 13°C at Britannia Beach and from 7.0 to 10.0°C at the reference areas. During freshet, temperatures were 9–18.5°C at Britannia Beach and 9.5–19°C at the reference areas. Mean surface salinity was lower during freshet than before freshet (Fig. 2*b*). Before freshet, salinity ranged from 3 to 23 ppt at Britannia Beach and from 3 from 20 ppt at the reference areas. During freshet, salinity was 1–13 ppt at Britannia Beach and 1–12 ppt at the reference areas. Surface temperature and salinity during 20 March – 27 May 1998 followed trends similar to those of 1997.

At all sites, except BB3, the seasonal pattern in mean pH was characterized by typical values for marine water (7.6–8.5) before the onset of spring freshet followed by a decrease to 6.5–7.3 during freshet (Fig. 2*c*). Site BB3, which is close to the mouth of Britannia Creek, had the lowest mean pH (6.5) with a minimum value of 5.8. During 1998, a similar trend occurred in which pH decreased near Britannia Creek mouth during spring (4.8 at BB4) and there was little seasonal variation at the reference areas.

There were no significant differences in pH among Britannia Beach sites during 1997; however, pH at BB3 was significantly lower than at the reference sites (two-sample t test: $t = -3.511$, df = 12, $P = 0.004$). In 1998, pH was lowest at sites nearest the creek mouth, but no significant differences were found among Britannia Beach sites or between sites at the creek mouth and the reference area.

During both 1997 and 1998, surface concentrations of dissolved Cu were highest during spring near the mouth of Britannia Creek at BB3 and BB4 (Fig. 3). There was a significant relationship between dissolved Cu and distance from the creek mouth ($r = 0.79$, df = 40, $P < 0.0005$), with dissolved Cu decreasing exponentially with increasing distance from the mouth. Dissolved Cu at BB3 in May 1997 ($2.67\ mg \cdot L^{-1}$) was significantly higher than at other Britannia Beach sites (one-way analysis of variance: $F = 9.98$, df = 4, $P < 0.0001$). In 1998, dissolved Cu was measured from 5 May to 4 June only, and the highest concentrations were observed once again in May but at BB4 ($1.79\ mg \cdot L^{-1}$). This was lower than in the previous year, and no significant differences were found among Britannia Beach sites or between creek sites and reference areas for 1998, probably due

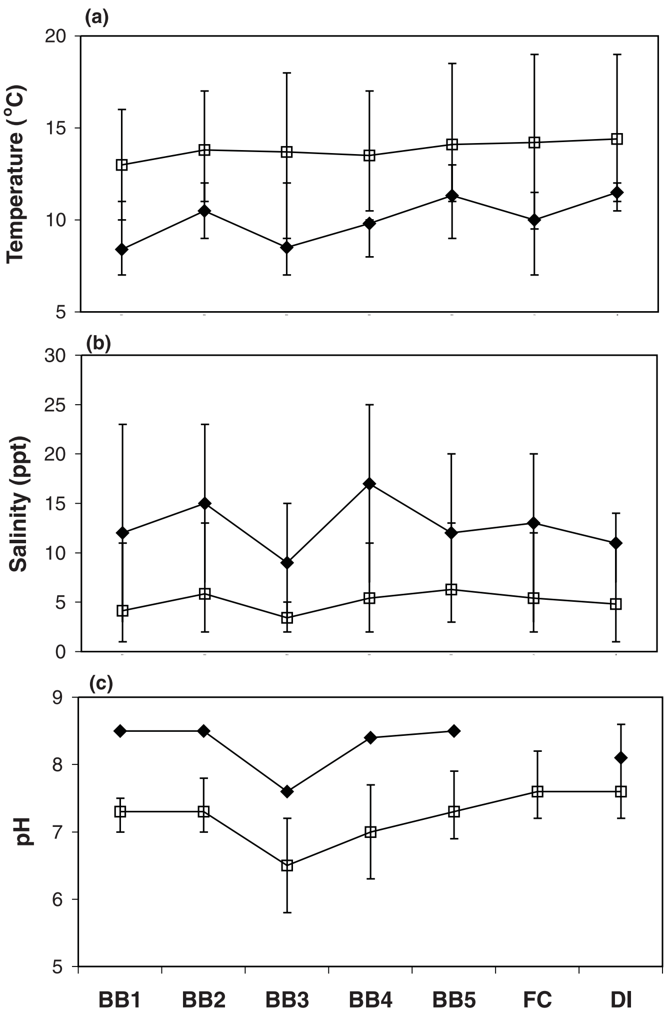


Fig. 2. Ranges of and mean surface water (*a*) temperature, (*b*) salinity, and (*c*) pH for the prefreshet period (◆, 1 April – 15 May) and freshet period (□, 16 May – 31 August) at Britannia Beach sites and reference areas from 4 April to 14 August 1997.

to the low sample size. Standard errors were calculated for 1997 data but are not plotted in Fig. 3 because of the high variability. Standard error was highest for BB3 (0.1465) and BB4 (0.0377) because these sites were located in the creek mixing zone where the AMD plume varied according to discharge, tides, and surface currents. Standard error at the other Britannia Beach sites was comparatively low (0.008–0.01).

Dissolved Cu decreased logarithmically with increasing salinity at the Britannia Creek estuary. This relationship was significant in 1998 (r = 0.63, df = 18, P = 0.0016) but not in 1997, probably because of the extremely high Cu level at BB3 in May, which acted as an outlier. Dissolved Cu exhibited a significant log negative relationship with pH in 1997 (r = 0.65, df = 28, P < 0.001) and in 1998 (r = 0.45, df = 18, P < 0.024).

Abundance and distribution of juvenile chum and chinook salmon

Chum salmon (fry stage) was the most abundant species caught in beach seines (Table 1), representing 35.2–36.9% of the total catch at Britannia Beach and 47.0–81.5% at Furry Creek. Juvenile chinook salmon (fry and smolt stages) made up a small proportion of the catch at Britannia Beach (0.9–4.0%) and at the reference areas (5–7.1%).

CPUE of chum salmon fry showed a seasonal trend with highest numbers during the spring and fewer numbers by mid-June. Mean catch of chum salmon fry at Britannia Beach was $10.9 \cdot 100\ m^{-2}$ in April, $16.5 \cdot 100\ m^{-2}$ in May, and less than $2 \cdot 100\ m^{-2}$ for the remainder of the summer. CPUE of chum salmon fry was highest in early spring (11 April) at the most northern Britannia Beach sites ($8.9 \cdot 100\ m^{-2}$ at BB4, $58.2 \cdot 100\ m^{-2}$ at BB5) whereas peak CPUE occurred slightly later at southern Britannia Beach sites ($45.1 \cdot 100\ m^{-2}$ at BB2 on 6 May, $111.6 \cdot 100\ m^{-2}$ at BB1 on 21 May). This trend probably reflects the seaward movement of chum salmon fry through the Britannia Beach area. A similar seasonal pattern in chum salmon fry CPUE was observed for Furry Creek and Defence Islands.

In 1997, mean CPUE of chum salmon fry was lowest ($<2 \cdot 100\ m^{-2}$) near the mouth of Britannia Creek at BB3 and BB4 (Fig. 4). However, no significant differences were found among Britannia Beach sites, probably due to the high variability of the catch data. At BB1, BB2, BB3, and BB4, a large proportion of the seine hauls (61–91%) did not catch any chum salmon fry, and the greatest number of zero catches was at BB3 and BB4. Compared with the reference sites where an average of 10 chum salmon fry$\cdot 100\ m^{-2}$ were caught, CPUE was significantly less at BB3 (Mann–Whitney two-sample nonparametric test: U = 19.0, df = 1, P = 0.015) and at BB4 (Mann–Whitney two-sample nonparametric test: U = 23.5, df = 1, P = 0.041).

In 1998, chum salmon fry CPUE was significantly different among Britannia Beach sites (Kruskal–Wallis nonparametric test: H = 16.03, df = 4, P = 0.003), with chum fry essentially absent near Britannia Creek mouth ($<1 \cdot 100\ m^{-2}$). CPUE of chum salmon fry at BB3 (P = 0.047) and BB4 (P = 0.040) was significantly lower than at other Britannia Beach sites. At the reference areas, CPUE of chum salmon fry exceeded $30 \cdot 100\ m^{-2}$, which was significantly greater than abundance at BB3 (Mann–Whitney two-sample nonparametric test: U = 2.5, df = 1, P = 0.013) or at BB4 (Mann–Whitney two-sample nonparametric test: U = 3.5, df = 1, P = 0.013).

Chum salmon fry catch was lowest at those sites with highest dissolved Cu levels, i.e., BB3 and BB4. In 1997, mean Cu was $0.918\ mg \cdot L^{-1}$ at BB3, $0.166\ mg \cdot L^{-1}$ at BB4, and only $0.009\ mg \cdot L^{-1}$ at the reference sites. In 1998, mean Cu was $0.483\ mg \cdot L^{-1}$ at BB3, $0.451\ mg \cdot L^{-1}$ at BB4, and $0.021\ mg \cdot L^{-1}$ at the reference sites. The relatively higher value at the reference sites in 1998 is due to an anomalous value from FC1 on 15 April ($0.068\ mg \cdot L^{-1}$) (see Fig. 3). Omitting this outlier results in a mean value of $0.009\ mg \cdot L^{-1}$ at the reference sites, which is similar to our 1997 results. The negative log relationship between chum CPUE and dissolved Cu was significant in 1998 (r = 0.64, df = 16, P = 0.0022) but not in 1997, probably due to the variability in

Fig. 3. Seasonal patterns in mean concentration of dissolved Cu in surface water at beach seine sites at Britannia Beach and Furry Creek during 1997 and 1998.

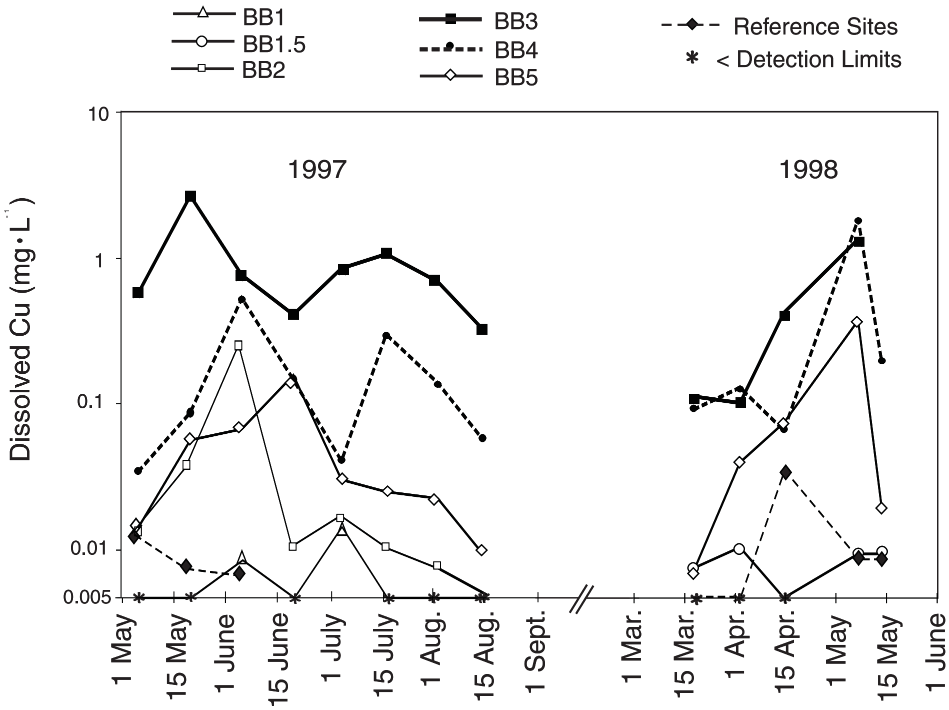


Table 1. Salmonid abundance (CPUE, no.·100 m^{-2}) and percent composition from beach seine surveys at Britannia Beach and reference areas (all sites combined) during 1997 (April–August) and 1998 (March–May).

		Britannia Beach						Reference sites					
		1997 (*n* = 33)			1998 (*n* = 21)			1997 (*n* = 297)			1998 (*n* = 36)		
Species	Stage	CPUE	SE	%	CPUE	SE	%	CPUE	SE	%	CPUE	SE	%
Chum salmon	Fry	5.5	2.4	35.2	6.9	3.1	36.9	11.5	5.2	47.0	22.6	15.6	81.5
Chinook salmon	Smolt	0.6	0.2	4.0	0.2	0.1	0.9	1.7	0.4	7.0	1.4	1.1	5.0
Chinook salmon	Fry	0.4	0.2	2.4	0.0	—	—	1.8	0.9	7.1	0.0	—	—

Note: The number of hauls is provided in parentheses. A dash indicates no value.

the catch data. No clear trend was evident between chum salmon fry abundance and surface water pH.

Although relatively more juvenile chinook salmon were captured at the reference areas than at Britannia Beach, their numbers were too low to permit statistical analyses. In 1997, CPUE of chinook salmon fry was 0.4·100 m^{-2} at Britannia Beach and 1.8·100 m^{-2} at the reference sites. No chinook fry were caught at any site in 1998. CPUE of chinook salmon smolts at Britannia Beach was 0.6·100 m^{-2} in 1997 and 0.2·100 m^{-2} in 1998 and at the reference sites was 1.7·100 m^{-2} in 1997 and 1.4·100 m^{-2} in 1998.

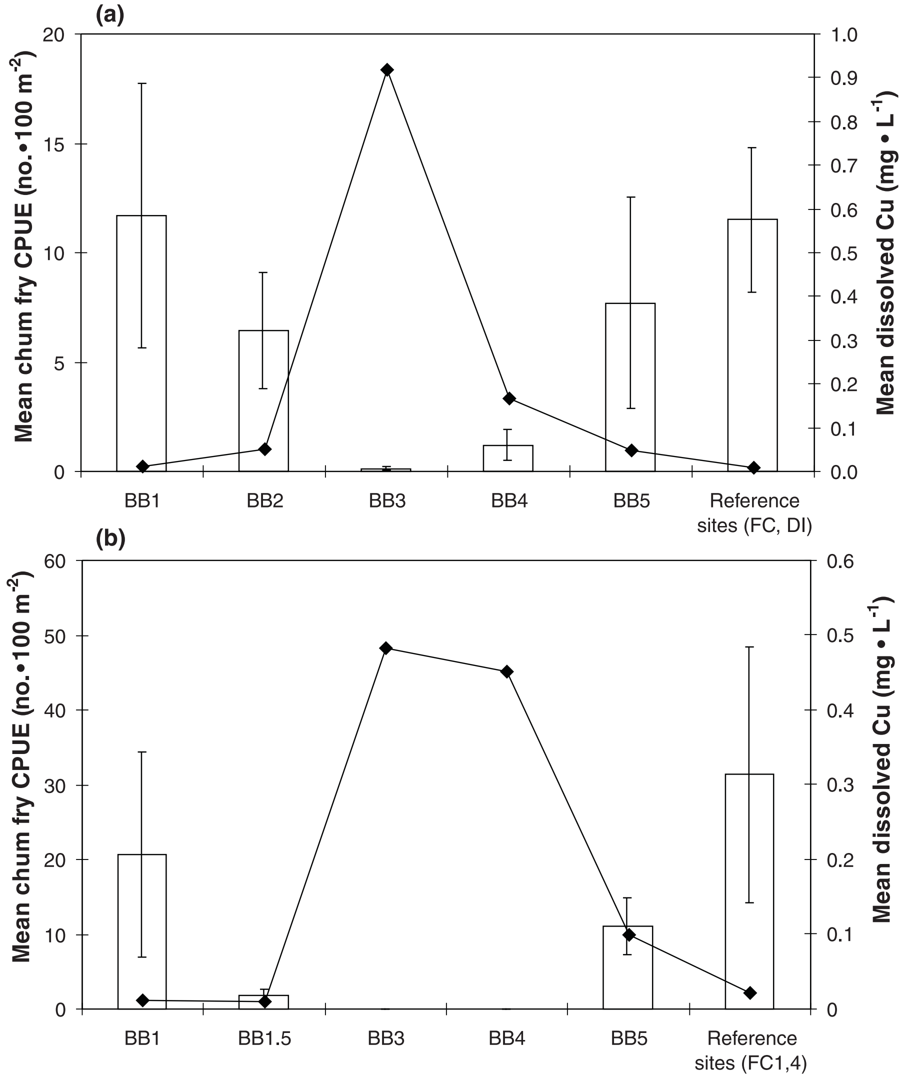
Laboratory bioassays

Britannia Creek water had lower concentrations of dissolved metals and lower hardness than water from 4100 portal (Table 2). Of all the metals present, Cu occurred in the highest concentration in Britannia Creek. Seasonally, concentrations of Al, Cu, and Zn were highest in May, while Cd concentrations were similar between months, often below detection limits. For 4100 portal water, the highest Cd and Zn occurred in May, while Al and Cu were highest in June.

Bioassay results confirmed that water from both the 4100 portal and Britannia Creek was acutely toxic to young salmon (Table 3). In freshwater, very low concentrations of 4100 portal water were lethal to chum salmon fry (0.7% LC_{50}) and to chinook salmon smolts (3.4% LC_{50}). Exposure to Britannia Creek water, at 0 ppt, resulted in LC_{50} values of 18% for chum salmon fry and 29.7% for chinook salmon smolts, indicating that 4100 portal water was more toxic than creek water. Toxicity was somewhat mitigated by increased salinity. At a salinity of 10 ppt, toxicity of Britannia Creek and 4100 portal water to chum salmon fry was reduced to 62.2 and 12.6%, respectively.

The initial dissolved Cu concentrations in sample water varied because water was collected at different times of the year and subject to seasonal differences in metal concentrations. In freshwater, LC_{50} results ranged from 9.4 mg dissolved Cu·L^{-1} in Britannia Creek water to 10.4 mg·L^{-1} in 4100 portal water. At higher salinity (10 ppt), LC_{50} occurred

Fig. 4. Mean CPUE of chum salmon fry in relation to mean dissolved Cu at beach seine sites at Britannia Beach and reference sites during (*a*) 1997 and (*b*) 1998. Bars represent 1 SE of the mean.



at higher Cu concentrations, 103.3 mg·L^{-1} in Britannia Creek water and 209.2 mg·L^{-1} in 4100 portal water.

Caged chinook salmon experiment

Temperature and salinity were similar at both fish cage sites with mean values of 7.6°C and 4.8 ppt at Britannia Beach and 9.9°C and 6.6 ppt at Porteau Cove. Surface water pH was lower at Britannia Beach (5.8) than at Porteau Cove (7.3), and surface concentrations of dissolved Cu were much higher at Britannia Beach (1.0–1.2 mg·L^{-1}) than at Porteau Cove (<0.01 mg·L^{-1}).

Survival results showed that surface water near the mouth of Britannia Creek was more toxic to juvenile chinook salmon than water more than 1 m deep in the same area. In the first experiment where all cages were placed at the surface, chinook salmon smolts near Britannia Creek experienced 100% mortality after 2 days, while caged fish at Porteau Cove experienced only 4% mortality after 6 days (Fig. 5*a*). In the second experiment where cages were placed at 0, 2, and 3 m, all chinook salmon smolts in the surface cage at Britannia Beach died within 2 days whereas fish in cages at 2 or 3 m at Britannia Beach had 100% survival after 5 days (Fig. 5*b*). At Porteau Cove, chinook salmon smolts experienced over 90% survival at all depths for 5 days. During the second experiment, chinook salmon smolts at Porteau Cove experienced some mortality, but this was probably due to poor water circulation in the cages as a result of fouling by algae. At Britannia Beach, no algae colonized the cages.

Discussion

Effects of AMD on juvenile salmonids

Results from this study demonstrated that juvenile salmon are negatively impacted by AMD from Britannia Creek, as seen by reduced abundance of chum salmon fry near the mouth of the creek and high mortality of chinook salmon smolts transplanted to surface cages near Britannia Creek. Laboratory bioassays showed that AMD from Britannia Mine was toxic to juvenile chum and chinook salmon. Concentrations of dissolved Al, Cd, Cu, and Zn in Britannia Creek and 4100 portal water used in the bioassays exceeded

Table 2. Mean concentration of selected dissolved metals and hardness for Britannia Creek (n = 3) and 4100 portal (n = 3) water used for the salmon bioassays.

	May 1997				June 1997				July 1997				February 1998			
	Britannia Creek[a]		4100 portal[a]		Britannia Creek[a]		4100 portal[a]		Britannia Creek[a]		4100 portal[b]		Britannia Creek[a]		4100 portal[a]	
Concentration ($mg \cdot L^{-1}$)	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Al	1.76	0.019	21.5	0.219	1.06	0.354	22.7	1.300	0.073	0.009	16.8	7.42	0.1	—	17.7	1.054
Cd	0.006	0.001	0.128	0.004	0.006	0.0003	0.100	0.006	<0.005[c]	—	0.083	0.003	0.006	—	0.059	0.005
Cu	2.55	0.029	15.4	0.153	1.66	0.247	16.6	0.977	0.522	0.009	14.9	0.907	0.464	0.001	11.6	0.689
Zn	1.26	0.012	28.2	0.318	1.03	0.340	22.3	1.089	0.362	0.015	14.8	0.921	0.375	0.002	14.2	0.649
Total hardness	41.9	0.400	1537	20.3	44.6	20.3	1493	93.9	14.3	0.033	1088	67.9	17.4	0.100	1147	60.1

Note: A dash indicates that the standard error was not calculated because only one sample was above detection limits.
[a]Detection limits ($mg \cdot L^{-1}$): A1 = 0.05, Cd = 0.005, Cu = 0.005, Zn = 0.002.
[b]Detection limits ($mg \cdot L^{-1}$): A1 = 0.5, Cd = 0.05, Cu = 0.05, Zn = 0.02.
[c]Below detection limits.

current working and approved water quality criteria (B.C. Ministry of Environment, Lands and Parks 1998) for the health of aquatic life (criteria: Al = 0.05 $mg \cdot L^{-1}$, Cd = 0.1 $\mu g \cdot L^{-1}$, Cu = 3 $\mu g \cdot L^{-1}$, Zn = 0.086 $mg \cdot L^{-1}$). Copper was the most likely component of Britannia Mine AMD to cause mortality due to its extremely high concentration and its known toxicity to fish. Although Zn concentrations also exceeded water quality criteria, juvenile salmonids are generally more tolerant of Zn than of Cu (Chapman 1978). Therefore, dissolved Cu from AMD poses the greatest risk to juvenile salmonids in the Britannia Creek estuary. This corresponds to other research, which reported that most toxic effects of AMD to Atlantic salmon (*Salmo salar*) are associated with increases in the proportion of Cu, not Zn (Gray 1998).

Copper toxicity to fish is directly related to the activity of the free cupric ion (Cu^{2+}), which is a function of the proportion of dissolved Cu in the environment (Engel et al. 1981). The primary target of Cu toxicity is the gill, where it affects membrane permeability, thus disrupting ionic and osmotic balance (Eisler 1997). Recent work has suggested that exposure to dissolved Cu in excess of 5.6 $\mu g \cdot L^{-1}$ presents an ecological risk to marine organisms (Hall and Anderson 1999), and the current U.S. Environmental Protection Agency (1995) criterion for Cu exposure in salt water is 4.8 $\mu g \cdot L^{-1}$. These criteria correspond well to the detection limits used in our analysis of Cu in water from Britannia Creek estuary. Dissolved Cu concentrations during 1997 and 1998 were higher than detection limits at most sites, except BB1. Near the mouth of Britannia Creek, dissolved Cu concentrations were as much as 100 times greater than typical background levels for surface seawater, 0.06–0.21 $\mu g \cdot L^{-1}$ (Eisler 1997). These results are consistent with previous measurements obtained near Britannia Creek in 1986 (Van Aggelen and Moore 1986), 1992 (Dunn et al. 1992), and 1997 (Chretien 1997). Therefore, surface Cu concentrations near Britannia Creek appear to have remained consistently high for the last 15 years. In an earlier study in Howe Sound, Thompson and Paton (1978) found relatively low concentrations of dissolved Cu near Britannia Beach (0.73–1.04 $\mu g \cdot L^{-1}$), but this is most likely because their sampling locations were located offshore approximately 500 m away from the mouth of Britannia Creek and measurements were obtained at 1 m below the surface.

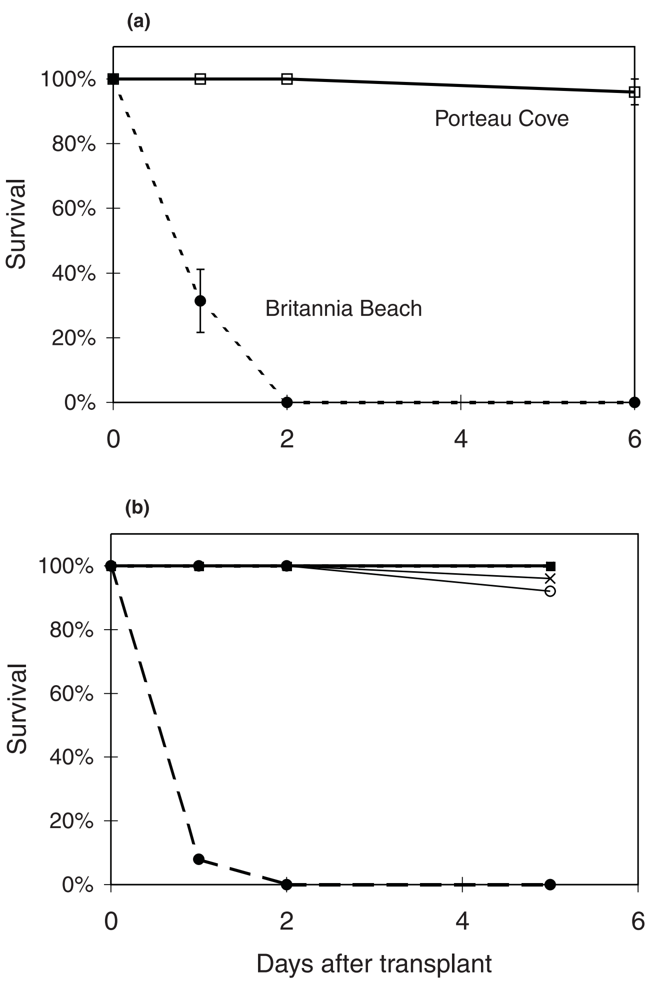
Our pH data indicate that surface waters near the mouth of Britannia Creek were sufficiently acidic to cause toxic effects in juvenile salmon. Marine waters normally have a pH of 7.8–8.2, while a pH of less than 6 is considered harmful to fish (Fromm 1980; B.C. Ministry of Environment, Lands and Parks 1998). Near the mouth of Britannia Creek, pH was less than 6 in spring during both 1997 and 1998. Exposure to low pH can cause acute lethality as well as many sublethal effects in fish, such as disrupted ionic and respiratory exchanges, reduced growth, reproductive failure, and skeletal deformities (Fromm 1980). The extent to which low pH affects fish is often species specific, but early developmental stages tend to be less tolerant of acidic conditions. In salmonids, the alevin and fry stages are most susceptible to low pH (Fromm 1980).

Results from our laboratory bioassays indicate that acute lethality occurred at 9.4–13.7 mg $Cu \cdot L^{-1}$ in fish exposed to

Table 3. Laboratory bioassay results (96-h LC_{50}) for chum salmon fry and chinook salmon smolts exposed to water from Britannia Creek and the 4100 portal.

					Britannia Creek		4100 portal	
Species	Bioassay date	Length range (cm)	Weight range (g)	Salinity (ppt)	96-h LC_{50} (%)	96-h LC_{50} (mg $Cu \cdot L^{-1}$)	96-h LC_{50} (%)	96-h LC_{50} (mg $Cu \cdot L^{-1}$)
Chum salmon	1 July 1997	6.0–9.7	1.6–7.1	0	18.0	9.4	0.7	10.4
Chum salmon	1 May 1997	4.9–6.4	0.9–1.8	5	4.7	12.0	0.9	13.9
Chum salmon	1 June 1997	4.1–5.6	1.0–3.1	10	62.2	103.3	12.6	209.2
Chinook salmon	1 February 1998	4.8–5.8	1.0–1.8	0	29.7	13.7	3.4	39.4

Fig. 5. Survival of chinook salmon smolts (length: mean = 88 mm, SD = 6 mm; weight: mean = 7.6 g, SD = 1.4 g) in (*a*) surface cages placed at Britannia Beach (BB, ●) and Porteau Cove (PC, □) and (*b*) cages placed at BB surface (●), PC surface (○), and PC 3 m (×). Results from three cages (BB 2 m, BB 3 m, and PC 2 m) overlap on the 100% survival line, which is shown by (■). Results from the second experiment (*b*) are reported for the first 5 days only, before the cages at the reference area were substantially fouled with algae.



Britannia Creek water at 0 ppt. This Cu concentration is higher than that observed in the field, yet in situ experiments showed that surface water near Britannia Creek was highly toxic. This discrepancy is likely because the water used in the bioassays was adjusted to normal pH but caged fish were exposed to high dissolved Cu and low pH.

Previous toxicity tests conducted with Britannia 4100 portal water determined that the 96-h LC_{50} in seawater (19 ppt) was 16% for chum salmon and 17.8% for stickleback (Van Aggelen and Moore 1986). These results correspond well to our chum salmon bioassay results in 4100 portal water at 10 ppt (LC_{50} = 12.6%). Differences between our results and other single-metal bioassays are not unexpected, since mixtures of metals typically found in AMD often have a more-than-additive toxic effect on fish (Eisler 1997). The slightly lower toxicity found by Van Aggelen and Moore (1986) could also have been because their tests were conducted at higher salinity, which decreased the toxic effects of Cu.

The physiological effects of Cu are not well documented in saltwater fish, but toxicity seems to be reduced at higher salinities (Anderson et al. 1995), as indicated in our bioassays with chum salmon. Toxicity of Britannia Creek and 4100 portal water to juvenile chum salmon was considerably lower at 10 ppt compared with freshwater. Wilson and Taylor (1993) found a similar result in that rainbow trout exposed to 0.4 mg $Cu \cdot L^{-1}$ did not suffer any noticeable effects after they were adapted to full-strength seawater. In another study where juvenile chum salmon were placed in enclosures in seawater and exposed to sublethal Cu levels (2.5–5.0 $\mu g \cdot L^{-1}$), no physiological or growth effects were observed (Thompson and Paton 1976). The protective function of salt water to Cu toxicity is thought to be related to two factors. Firstly, the abundant Ca and Mg ions in seawater outcompete Cu ions for access to gill epithelia. Secondly, Cu complexes and precipitates with Ca, Mg, and other ions present in seawater, which decreases the concentration of bioavailable Cu (Engel et al. 1981).

Toxicity of AMD from Britannia Creek is influenced by many factors besides salinity, including pH, water hardness, and the amount of organic matter. The low pH typical of AMD enhances the toxicity of heavy metals because acidic conditions allow more metal ions to be solubilized and therefore become bioavailable. Copper toxicity is inversely related to pH, with highest toxicity occurring at around pH 5 because at this pH, Cu exists mostly in the ionic form (Campbell and Stokes 1985). The inherently soft waters in and near Britannia Creek also exacerbate the toxicity of AMD to fish because there is no natural buffering capacity against acidity and the toxic action of Cu tends to be higher in soft water (Howarth and Sprague 1978). The presence of organic matter has been recently shown to reduce toxicity of AMD by preventing Cu ions from binding to gill tissue (Richards et al. 1999). Organic material is transported by the Squamish River during freshet periods (May–July), but this may not lessen the toxicity of AMD from Britannia Creek to juvenile salmonids, since the majority of chum salmon fry have already migrated through the area by the time the Squamish River reaches peak discharge levels.

Susceptibility of juvenile salmonids to AMD from Britannia Creek

Our results confirm that juvenile salmonids are susceptible to the adverse effects of AMD from Britannia Creek because the timing of salmon fry out-migration coincides with the period when potential Cu toxicity is greatest. Chum salmon fry were most numerous during spring (April–May), which agrees with previous research in Howe Sound (Levings and Riddell 1992). Spring also corresponds to the period when metal loading rates from Britannia Creek are greatest as a result of rain and melting snow rapidly flushing contaminated water from the mine. This seasonal pattern has been observed in other AMD-impacted systems (Johnson and Thornton 1987; Gray 1998).

Juvenile salmon are especially vulnerable to the toxic plume from Britannia Creek because AMD is concentrated in the freshwater surface layer and salmon fry preferentially occupy upper depths during estuarine residency. Other research in Howe Sound has shown that chum salmon fry will occupy the upper 2 m of the water column even when these waters are contaminated with pulp mill effluent (Birtwell and Kruzynski 1989). Results from our caged fish experiments demonstrated that surface water near Britannia Creek is more toxic than deeper water. Since surface water quality was similar between Britannia Beach and Porteau Cove, except for dissolved Cu, which was more than 100 times greater at Britannia Beach, mortality was probably due to elevated Cu concentrations and, to a lesser extent, low pH. Additional data collected in April 1999 (Varela et al. 2000) showed that dissolved Cu at the Britannia Beach cage site was much higher at the surface (0.13 $mg\cdot L^{-1}$) than at 6 m depth (0.013 $mg\cdot L^{-1}$), which further indicates that mortality was related to high levels of dissolved Cu.

A similar caged fish experiment was conducted at Britannia Beach in April 1972 (Dent 1972). In this study, young coho salmon were placed in cages at five sites on the eastern shore of Howe Sound, from approximately 700 m north of Britannia Creek to about 1 km north of Furry Creek. After 8 days, none of the fish had died; however, this was probably because the cages were placed at 2–3 m depth, which according to our caged salmon experiment would be sufficiently deep to minimize exposure to the toxic AMD in surface water.

Extent of AMD contamination near Britannia Creek

There are several possible sources of dissolved Cu to Howe Sound from Britannia Mine, including submerged mine tailings, AMD from the 4100 portal, and AMD from Britannia Creek. It is unlikely that our observed Cu levels originated from mine tailings, since other research has shown that these tailings do not release significant amounts of Cu. Trace metals present in the tailings are geochemically unreactive because natural sediments have been deposited over the tailings and bacterial decomposition has resulted in an anoxic environment, which inhibits dissolution of metals (Drysdale and Pederson 1992). In 1978, Cu concentrations in interstitial water of these tailings were only 3.74 $\mu g\cdot L^{-1}$ (Thompson and Paton 1978), which is much lower than surface concentrations near Britannia Creek. It is also unlikely that our observed dissolved Cu concentrations were due to AMD from the 4100 portal, since previous research has shown that this discharge remained confined at depth during the summer (Chretien 1997). Furthermore, our vertical profiles indicated that dissolved Cu concentrations were five times higher at the surface than at depth, which is consistent with Britannia Creek being the main source of AMD to the nearshore zone.

The AMD plume from Britannia Creek was highly spatially and temporally variable, depending on oceanographic conditions, metal loading rates, current patterns, and geochemical removal processes. Regression analyses indicated that dissolved Cu was greatest in freshwater with low pH, suggesting that the main source of Cu to the estuarine environment is AMD from Britannia Creek. Because of the counterclockwise gyre at Britannia Beach and the presence of a large jetty immediately south of the creek mouth, it is unlikely that highly contaminated water extended as far south as Furry Creek. Sites BB2 and BB5 were approximately equidistant from the mouth of Britannia Creek, yet dissolved Cu levels were consistently higher at the more northern site, BB5. This probably reflects AMD movement due to the gyre transporting water in the nearshore zone in a northward direction.

The behaviour of Cu as it enters an estuary is very complex, since the physicochemical factors that affect Cu speciation are highly variable in estuaries (Engel et al. 1981). In the Britannia Creek mixing zone, Cu is extensively removed by precipitation (Chretien 1997). This was corroborated by our data, which showed that dissolved Cu concentrations in the nearshore zone decreased exponentially with increasing distance from the creek mouth. Specifically, dissolved Cu levels decreased by approximately 99% at 600 m south of the creek mouth. Dissolved Cu concentrations were indirectly related to salinity and pH, which agrees with other studies in AMD-impacted estuaries where Cu removal rates increased with higher salinity and higher pH (e.g., Foster et al. 1978; Johnson and Thornton 1987; Featherstone and O'Grady 1997). Thompson and Paton (1978) found a similar trend in Howe Sound in which dissolved Cu concentrations at 1 m depth decreased significantly with increasing salinity from 2.82 $\mu g\cdot L^{-1}$ at 2.5 ppt to 0.55 $\mu g\cdot L^{-1}$ at 29.2 ppt.

Implications of AMD contamination to salmon productivity

There are a variety of ways in which juvenile salmon may be responding to the toxic plume emanating from Britannia Creek. Firstly, they may remain near Britannia Creek and sustain high mortality due to acutely lethal conditions. Or, upon sensing poor water quality, juveniles may move quickly through the area near the creek mouth while sustaining some mortality or sublethal effects. Finally, salmon fry may avoid the area completely. Results from beach seining suggest that juvenile chum salmon near Britannia Creek are either suffering immediate mortality or avoiding the AMD plume. The most likely scenario is that chum salmon fry are avoiding the plume, since a mark–recapture study conducted in 1997 showed that marked chum salmon fry released at Britannia Creek were not recaptured shortly after release. No marked fry released at BB4 were recovered at that site more than 2 h after release, while fish released at Furry Creek remained there for up to 11 days (Emmett 1997). The response is difficult to confirm in the field because if chum salmon fry are dying immediately or suffering sublethal effects, they would be difficult to recover. According to our 1997 beach seine results, chum salmon fry were present at BB1, BB2, and BB5 for approximately 2 months but only sporadically at BB3 and BB4. Since chum salmon fry are known to reside in estuaries for extended periods (Healey 1982) and individual migration times vary, there could be a continuous flux of chum salmon fry from the Squamish River migrating past Britannia Beach from early April to mid-June each year.

The behavioural response of salmonids to Cu is not well understood. Juvenile salmonids have been reported to avoid sublethal concentrations (Sprague 1964) while exhibiting attraction to acutely lethal levels (Pedder and Maly 1985). Birtwell and Kruzinski (1989) found that some chum salmon fry remained in surface water contaminated with low levels of pulp mill effluent, but nearer the outfall, they avoided the surface layer, suggesting that their instinct to occupy surface water may override avoidance reactions. Although avoidance behaviour could minimize the toxic effects of AMD, ultimately it could prove detrimental if juvenile salmon are forced into other less optimal habitats. Previous work has indicated that juvenile chum salmon that move to deeper habitats experience higher predation, increased competition, reduced food supply, and, consequently, higher mortality (Healey 1982). Thus, poor water quality from AMD contamination could reduce the quantity and quality of suitable rearing habitat in Howe Sound. In the future, it will be important to assess the response of juvenile chum and chinook salmon to AMD from Britannia Creek to determine how behavioural modifications may affect survival and growth.

In addition to the direct effects of AMD on juvenile salmon, broader ecosystem effects are also likely at Britannia Creek estuary. Related studies have shown that rockweed (*Fucus gardneri*), a macroalgae that is normally ubiquitous in the intertidal zone of Howe Sound, was absent near Britannia Creek (Marsden 1999). Invertebrates, such as amphipods and mussels, were also directly affected by AMD from Britannia Creek (Grout et al. 1999; Grout and Levings 2000). Other salmonid life stages may also be impacted by AMD from Britannia Mine. For example, during July and August, mature chinook salmon are routinely collected for hatchery brood stock at Britannia Beach, just north of the creek mouth, at site BB4 (R. Semple, Tenderfoot Creek Hatchery, P.O. Box 477, Brackendale, BC V0N 1H0, Canada, personal communication). Although this period does not correspond to peak metal loadings from the creek, adult salmon may experience sublethal toxic effects. Furthermore, fish that use deeper habitats, such as postsmolt chinook salmon that can occur at depths of 30 m near Britannia Beach (C.D. Levings, personal observation), may be adversely affected by AMD discharged from the 4100 portal outfall.

To conclude, more than 25 years after closure of Britannia Mine, water quality near the mouth of Britannia Creek remains very poor. Results from this study demonstrate that juvenile salmonids in the Britannia Creek estuary are adversely affected by AMD discharged from Britannia Creek. Furthermore, our results confirm that juvenile salmon are particularly vulnerable to the toxic effects of AMD due to their seasonal abundance patterns and surface distribution. Although AMD is considered to be the most serious water contamination problem associated with mining because of its continuous, persistent nature and toxicity to aquatic organisms, several treatment strategies exist and some can be effective in terms of biological recovery (Deniseger et al. 1990; Gray 1997). Reducing the toxic risk to juvenile salmon will be necessary before productive capacity at the Britannia Creek estuary can be restored.

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