PERGAMON

Marine Pollution Bulletin 46 (2003) 65–73

MARINE POLLUTION BULLETIN

www.elsevier.com/locate/marpolbul

Survivorship and growth of *Fucus gardneri* after transplant to an acid mine drainage-polluted area

A. Dale Marsden [a,b], Robert E. DeWreede [a,*], Colin D. Levings [b]

[a] *Department of Botany, University of British Columbia, 3529-6270 University Blvd., Vancouver, BC, Canada V6T 1Z4*

[b] *Coastal and Marine Habitat Science Section, Department of Fisheries and Oceans, West Vancouver Laboratory, 4160 Marine Drive, West Vancouver, BC, Canada V7V 1N6*

Abstract

Acid mine drainage (AMD) from an abandoned copper mine at Britannia Beach, British Columbia, Canada, enters the marine environment through Britannia Creek. The surrounding intertidal zone is devoid of rockweed, *Fucus gardneri* Silva, a seaweed that dominates nearby shores. Rockweed plants were transplanted to the intertidal zone near Britannia Creek and monitored for changes in percent cover, survivorship, growth rate and Cu content. Autumn and winter transplants to within 100 m of Britannia Creek resulted in negative growth rates and high mortality within 57 days of exposure to AMD, with Cu levels in rockweed surpassing 2300 ppm in dry tissue. Summer transplants to sites 300–700 m from Britannia Creek showed no consistent differences between AMD-exposed rockweed and control plants, possibly because the plants were stressed by desiccation. The results are consistent with ecological effects observed in other studies, and provide strong evidence for the role of AMD in excluding rockweed from the shores near Britannia Creek.

Keywords: *Fucus gardneri*; Acid mine drainage; Heavy metals; Britannia Mine; British Columbia

1. Introduction

Copper (Cu) ore was extracted from the mine at Britannia Beach, British Columbia, Canada, from 1902 until the mine ceased operations in 1974. Rain, snowmelt and groundwater now percolate through the abandoned mine tunnels, producing an acidic solution of dissolved metals known as acid mine drainage (AMD) (Price et al., 1995). A portion of the AMD from the mine flows into Britannia Creek, which in turn flows into Howe Sound, 50 km north of Vancouver, BC (Fig. 1). Rockweed, *Fucus gardneri* Silva (Phaeophyceae, Fucales), is an intertidal seaweed that thrives within about 2 km of the mouth of Britannia Creek and throughout Howe Sound, but is absent from the shore near the Creek (between the points labelled "Z" in Fig. 1; Marsden and DeWreede, 2000). Rockweed beds provide habitat and food for a variety of benthic invertebrates (Nassichuk, 1975), some of which are important food sources for chum salmon (*Oncorhynchus keta*) fry and chinook salmon (*Oncorhynchus tshawytscha*) fry and smolts (Levings and McDaniel, 1976; Levings and Riddell, 1992). Concern about the impact of AMD on these economically important salmon and their habitat has spurred this and other investigations into the effects of AMD on the marine ecosystem near Britannia Creek (Barry et al., 2000; Marsden and DeWreede, 2000; Grout and Levings, 2001).

Chretien (1997) examined the movement of metals from Britannia Creek into Howe Sound. The peak in dissolved Cu, Cadmium (Cd) and Zinc (Zn) concentrations at the mouth of Britannia Creek from April through June results from accumulated AMD being flushed from the mine workings by spring run-off (Fig. 2A; Chretien, 1997). These high levels of dissolved metals coincide with the freshet of the nearby Squamish River, which lowers the salinity of northern Howe Sound to 3–10‰. Grout and Levings (2001) measured the dispersal of dissolved Cu from Britannia Creek into the surface waters of Howe Sound in April–June 1998 (Fig. 2B).

* Corresponding author. Tel.: +1-604-822-6785; fax: +1-604-822-6089.

E-mail address: dewreede@interchange.ubc.ca (R.E. DeWreede).

0025-326X/02/$ - see front matter © 2002 Elsevier Science Ltd. All rights reserved.
PII: S0025-326X(02)00317-X

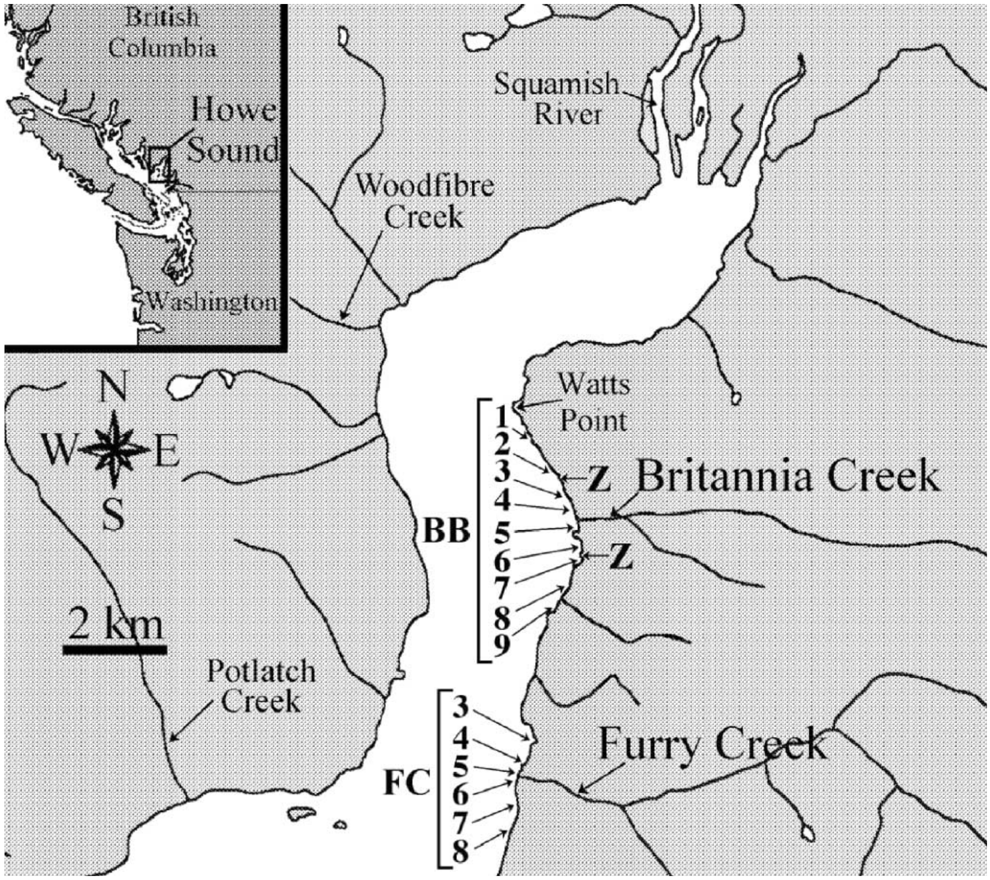

Fig. 1. Map of upper Howe Sound showing transplant sites. BB: Britannia Creek (AMD-exposed sites). FC: Furry Creek (control sites). The entire shoreline between points labelled Z is devoid of rockweed. Sites BB- and FC-5a and -5b are 50 and 100 m from their respective creek mouths, and are too close to show as separate on this map.

While dissolved Cu in seawater does not necessarily reflect the amount of bioavailable Cu at a given site or time (Gledhill et al., 1997), it does provide a useful tracer of how much AMD is present in a given sample of water; the amount of AMD in water should be proportional to the concentration of dissolved Cu. Additionally, we focused on the distribution of Cu, rather than other aspects of the AMD such as Zn, Cd and acidity, because (1) Zn is typically an order of magnitude less toxic to algae than Cu (Hargreaves and Whitton, 1976; Munda and Hudnik, 1986), (2) Cd is one to two orders of magnitude less concentrated in Britannia Creek than is Cu (Chretien, 1997), and (3) the buffering capacity of seawater appears to overwhelm the acidity of the drainage except in the immediate vicinity of the Creek mouth (Grout et al., 1998). However, any effects of AMD on biota cannot be attributed to any one component; these effects are likely caused by the cumulative effects of several parts of the effluent.

Many previous studies have examined the effects of Cu on algae. Strömgren (1979, 1980) tested the effect of Cu on five brown algal species in laboratory experiments. He found that growth rates of three species of *Fucus*, as well as two other fucoids, were decreased when exposed to 12–50 μg Cu l^{-1}. In laboratory experiments, Munda and Hudnik (1986) induced mortality in *F. vesiculosus* within 20 days of exposure to 2500 μg Cu l^{-1}. Other work has focused on the composition of intertidal communities in Cu-polluted areas. Castilla (1996) found a drastic reduction in algal and invertebrate diversity near the Salado River, Chile, where water that has drained over mine tailings flows into the Pacific Ocean. At sites within ~8 km of the river mouth, he found algal communities composed almost entirely of *Enteromorpha compressa* (L.) Greville (Chlorophyta). More distant, unpolluted sites showed a much greater richness of species (Castilla, 1996).

To examine the effects of AMD on rockweed, we conducted a series of bioassays by transplanting plants from a relatively unpolluted site to several sites near the mouth of Britannia Creek. We predicted that (1) percent cover, survivorship and growth rates of rockweed would decrease when plants were moved to this area, (2) Cu tissue burden of the plants would increase, and (3) these effects would decrease with distance from the mouth of Britannia Creek.

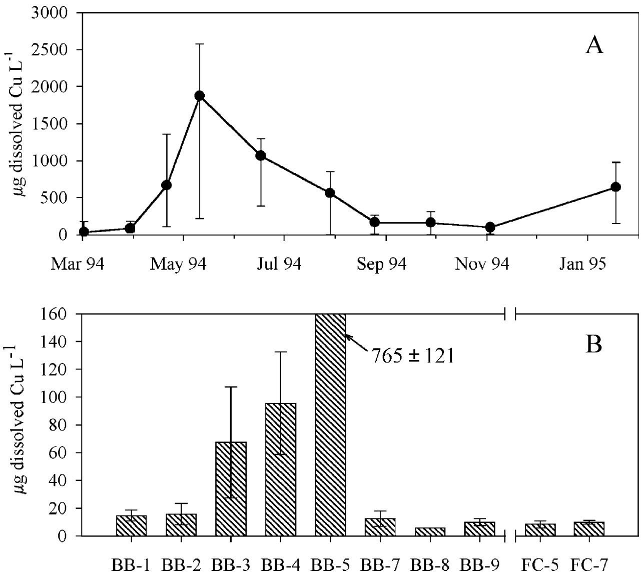

Fig. 2. Concentrations of dissolved Cu in Howe Sound near the mouth of Britannia Creek. (A) Temporal change in Cu levels from March 1994 to February 1995. Values are means of eight samples taken along a transect near the mouth of Britannia Creek. Error bars are the range of concentrations in each set of samples. Data from Chretien (1997). (B) Spatial dispersion of Cu from Britannia Creek in Howe Sound. See Fig. 1 for locations of sampling stations. Each value is a mean (±1 SE) of single measurements taken 29 April, 15 May, 28 May and 8 June 1998, except at sites BB-4 and -5, where three measurements were taken each day. Data from Grout and Levings (2001).

2. Methods

2.1. Transplant of rockweed

The beach near Furry Creek (see Fig. 1) showed abundant growth of rockweed, typical of much of Howe Sound, and was selected as a source area for plants to move to the area near Britannia Creek. It was also the recipient site for control plants, which were transplanted in the same manner as those moved to the Britannia Creek estuary. We labelled sites so that those with matching numbers were also matched in position relative to the creek mouths; for example, sites FC-5a and BB-5a were both 50 m south of their respective creek mouths.

Four transplants were conducted over a 13-month period, starting on the following dates: 2 November 1997 (to sites 5a and 5b, where 5b is 100 m south of the creek mouths; see Fig. 1 for all site locations), 15 February 1998 (to sites 5b), 11 June 1998 (to sites 6 and 7, respectively ~300 and ~600 m south of the creek mouths) and 10 August 1998 (to sites 3 and 4, respectively ~700 and ~300 m north of the creek mouths). While we refer below to plants "at" Britannia Creek or Furry Creek, no plants were actually moved into the creeks themselves; rather, we refer to the entire set of transplant sites near a creek mouth by that creek's name.

Rockweed-covered rocks (15–25 cm in diameter) were haphazardly selected for transplant based on several criteria. An attempt was made to select plants from all parts of their vertical range (typically 1.5–3.0 m above chart datum), as well as over a 100–200 m horizontal distance. Rocks were selected based on percent cover of rockweed; a reasonably high (>80%) cover was deemed necessary. Finally, rocks were selected in pairs, where each member was similar in size (±5 cm), percent cover and average plant length of rockweed (visual estimate), and within 1 m of each other in their original position on the beach. Selected rocks were tagged with numbered flagging tape and randomly assigned to treatments so that one member of each pair was moved to the appropriate control site near Furry Creek, while the other member of each pair was moved, using a boat or van, to a site near Britannia Creek. Transplants were always carried out during extremely low tides, when the plants would normally have been out of the water, and the total time the plants were in the boat or van was always less than one hour.

The selection of experimental sites was based primarily on the gradient of AMD with distance from the mouth of Britannia Creek. Once an appropriate site was chosen, the actual location on the beach was selected at the mean elevation of the plants' original positions at Furry Creek (2.0 ± 0.2 m above chart datum). All sites were gently sloping (10–25°) cobble and boulder beaches with similar wave exposure. Ice cream pails, 22 cm in diameter and 10 cm in height, were used as bases in which to place the rocks. The pails could be dug into the substratum much deeper than the rocks alone, which helped to prevent movement and loss of the transplants. Rocks and pails were placed in a group at the site with the greatest horizontal distance between any two rocks being 3 m.

2.2. Monitoring of rockweed condition

Plants were observed at varying intervals (see the *x*-axes on Figs. 3–6); a series of measurements were taken on each rock during each data collection. Percent cover of rockweed was estimated using a 25 cm × 25 cm quadrat as follows. The quadrat was pre-strung with nylon string in a grid pattern (with squares 1.5 cm × 1.5 cm), and 20 randomly selected intersection points were marked with pieces of wire. After haphazardly placing the quadrat on a rock, the object immediately under each point was recorded as rockweed or "other". Survivorship of rockweed was estimated by monitoring tagged plants; six plants were tagged on each rock during the first data collection, and their presence or absence was scored at each subsequent data collection. Growth rate of rockweed was estimated by measuring the length of the six tagged plants from the holdfast to the end of the longest frond. This provided an estimate

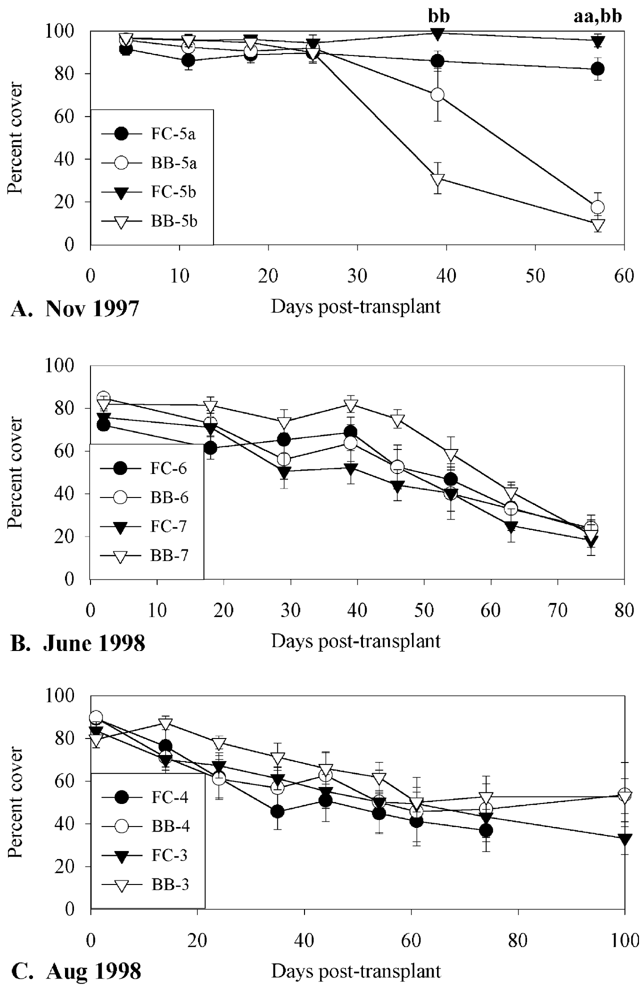

Fig. 3. Percent cover (mean ± 1 SE) of rockweed on rocks for three experiments. BB: Britannia Creek, FC: Furry Creek. See Fig. 1 for site locations. $n = 6$–8 transplanted rocks. Letters above graphs indicate significant differences: a for sites represented by circles, b for triangles; a/b indicates $p \leqslant 0.05$, aa/bb indicates $p \leqslant 0.01$. Results for the February 1998 experiment were similar to those for November 1997, and so are not shown.

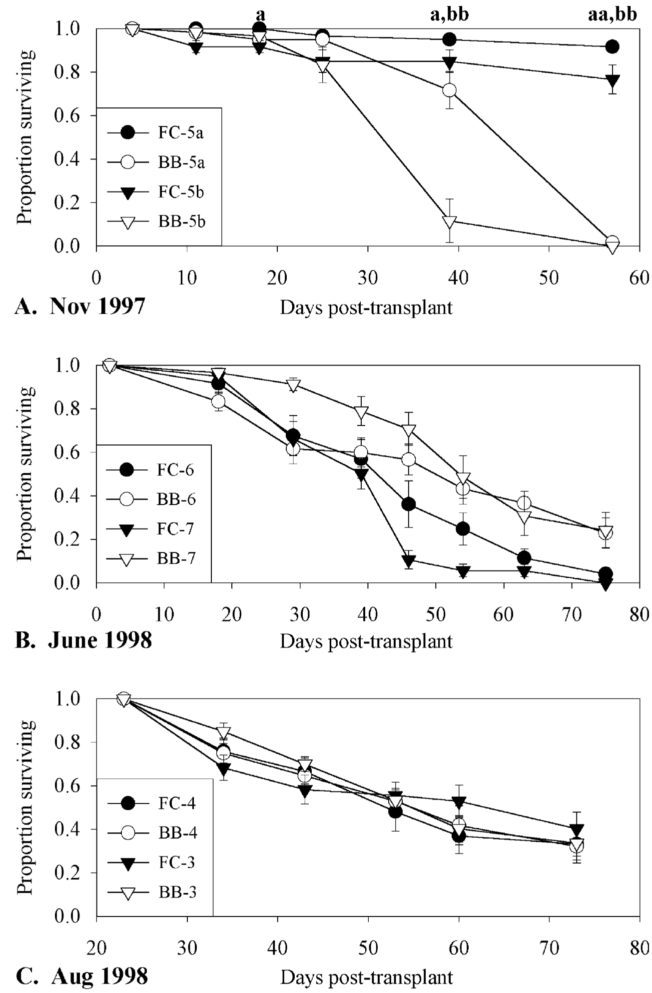

Fig. 4. Survivorship (mean ± 1 SE) of rockweed for three experiments. BB: Britannia Creek, FC: Furry Creek. See Fig. 1 for site locations. $n = 9$–10 for all transplants. See legend of Fig. 3 for meaning of letters above graphs. Results for the February 1998 experiment were similar to those for November 1997, and so are not shown.

of the average growth rate over the entire interval between measurements. Observations of plant colour and texture were also recorded at each data collection.

Cu concentration in rockweed thalli was measured in two or three plants from each site during the November 1997 experiment. The plants were removed from the rock, placed into plastic bags and returned to the laboratory. They were rinsed three times with seawater from the West Vancouver Laboratory system (~9 °C and salinity ~27) and frozen at −20 °C. When all samples had been collected during a given experiment they were dried in a fumehood at ~40 °C for ~4 h and sent to the Analytical Chemistry Laboratories of the Geological Survey of Canada (Ottawa, Ontario). There they were digested in 20 ml concentrated nitric acid and 2 ml perchloric acid, reduced to near dryness and then additionally digested with 10 ml concentrated hydrochloric acid, 10 ml concentrated hydrofluoric acid and 2 ml concentrated perchloric acid. Analysis was conducted by inductively coupled plasma mass spectrometry (ICP-MS).

The conclusion of the first three transplants was determined by mortality of rockweed plants; almost all plants at one or both of the Britannia Creek and Furry Creek sites had died and detached by the final time point shown in the results. The end point of the August 1998 transplant was determined when most of the rocks at all sites could not be located, apparently due to shifting of the substratum caused by wave action.

2.3. Statistical analysis

Survivorship scores yielded a proportion of plants surviving on each rock. Growth rate was averaged over all plants on a given rock. Individual plants could not be

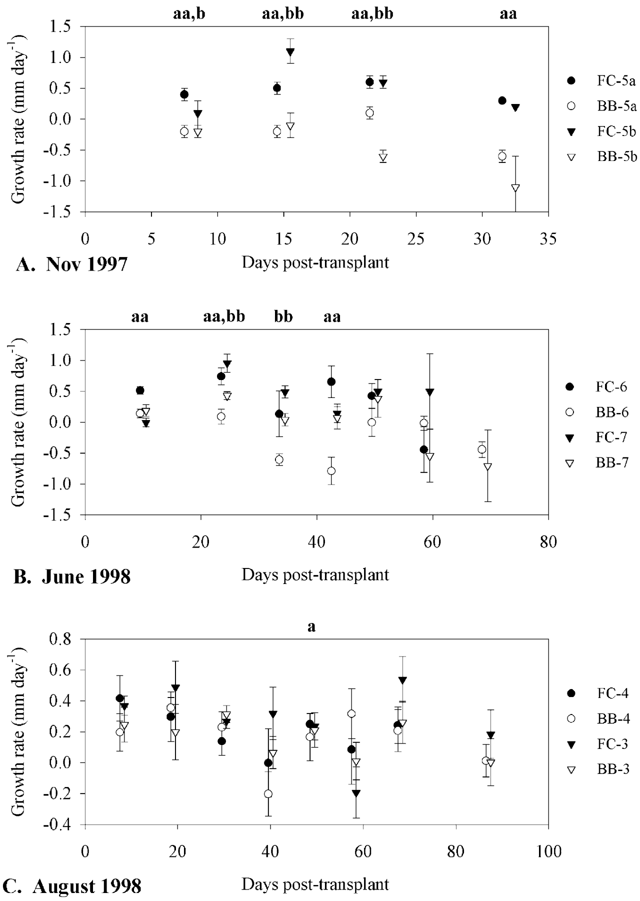

Fig. 5. Growth rates (mean ± 1 SE) of rockweed for all transplants in which measurements were taken. Time is expressed as the mid-point of the interval over which the growth rate is calculated. BB: Britannia Creek, FC: Furry Creek. See Fig. 1 for site locations. $n = 10$. See legend of Fig. 3 for meaning of letters above graphs. For clarity, circles are shifted one half day to the left and triangles are shifted one half day to the right.

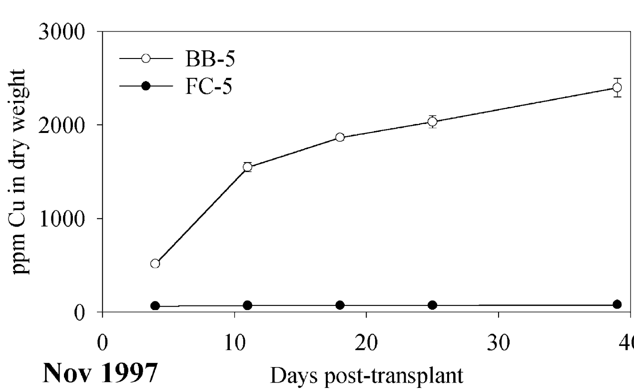

Fig. 6. Copper tissue burden (mean ± 1 SE) of rockweed on rocks transplanted November 1997 to sites BB-5 and FC-5. BB: Britannia Creek, FC Furry Creek. $p < 0.015$ in all cases. $n = 3$, except day 39, when $n = 2$.

counted as replicates for either growth or survivorship because they were not independent of each other: growth and survival of any given plant could be correlated with that of others on the same rock due to biological interactions or genetic relatedness.

Distributions of all variables were examined for normality, and non-normal variables were transformed using logarithmic or arcsine square-root transformations. Percent cover, proportion of tagged plants surviving and growth rates of Britannia Beach and Furry Creek plants were then compared for each data collection by paired or unpaired one-tailed Student's *t*-tests. Cu concentration in tissue was compared using unpaired one-tailed Student's *t*-tests. The direction of one-tailed hypotheses was determined by the predictions outlined in the introduction.

3. Results

3.1. Percent cover

November 1997 and February 1998 experiments showed similar trends. There was rapid loss of rockweed cover on rocks transplanted to Britannia Creek, while no such loss of plant cover was observed at Furry Creek (Fig. 3). In contrast, the June and August 1998 experiments showed no significant differences between cover of rockweed on rocks transplanted to Britannia Creek and that on rocks moved to Furry Creek; percent cover decreased on rocks at both sites.

3.2. Survivorship

The same general trends were seen in survivorship data as in the percent cover data (Fig. 4). Plants began to detach from rocks after approximately 25 days during the November 1997 and February 1998 experiments, while plants at the control sites maintained survivorship >75%. During the June and August 1998 experiments, both control and transplanted plants sustained heavy mortality so that survivorship to the final data collection was <25% and <40% for the June and August experiments, respectively. During the August 1998 transplant, tagging strings were poorly attached to the base of the plants and were observed to be falling from the plants. Plants were therefore retagged on 3 September (day 24) and previous data were disregarded.

3.3. Growth rate

Mean growth rates of plants moved to Britannia Creek in the November 1997 transplant were significantly lower than those of control plants during all measurement intervals (Fig. 5). In the June 1998 experiment, plants moved to site BB-6 had significantly lower mean growth rates than control plants during three of six measurement intervals, while plants moved to site BB-7 grew slower than control plants during two of six measurement intervals. Only one significant difference was detected between mean growth rates of AMD-exposed plants and controls in the August 1998 transplant. Growth rates were not measured in the February 1998 experiment.

3.4. Cu tissue concentration

Tissue concentration of Cu in rockweed increased rapidly during the first two weeks after the plants were transplanted to the mouth of Britannia Creek (sites BB-5a and -5b; Fig. 6); the mean tissue burden of Cu was significantly higher in plants at Britannia Creek during all data collections. The rate of uptake began to decrease after 11 days, but the Cu tissue burden was still increasing on day 39, after which no plants were available to be sampled. There was no change in the Cu tissue burden of plants at Furry Creek.

3.5. Changes in colour and texture

Qualitative changes in the condition of the plants became apparent when they were transplanted to the AMD-exposed sites. The most obvious change occurred in the colour and texture of plants that were moved to the Britannia Creek area: the entire thallus took on a reddish brown colour, which contrasted remarkably with the olive green colour of plants at Furry Creek and other nearby sites. The plants at Britannia Creek also produced large amounts of exudate and became very soft in texture relative to plants at Furry Creek, to the point that small bits of tissue often fell from the tips of the thalli. These changes in plant appearance and texture became apparent at different times for different experiments: 14 days post-transplant for all plants in the November 1997 experiment (sites BB-5a and -5b) and 30 days for the plants at site BB-6 for the June 1998 transplant. Plants at sites BB-7 (June 1998), BB-3 and -4 (August 1998) and all Furry Creek sites showed no such changes. During the summer experiments, many plants became severely desiccated, taking on a black and shriveled appearance. However, this change was equally prevalent in plants at Furry and Britannia Creeks, as well as in extant rockweed on the beaches in the immediate vicinity of the Furry Creek sites.

4. Discussion

4.1. Percent cover, survivorship and growth rate

The results of the percent cover, survivorship and growth rate measurements can be grouped into two overall patterns: (1) high mortality and low growth rates in plants at Britannia Creek relative to the control plants during the November 1997 and February 1998 transplants to sites BB-5, and (2) few or no differences between Britannia Creek and Furry Creek plants in terms of percent cover, survivorship or growth rate during the June and August 1998 transplants to sites BB-3, -4, -6 and -7, i.e., at greater distances from the AMD source. These results were consistent with the predictions presented in the introduction, but the lack of differences between AMD-exposed and control plants in June and August 1998 was surprising given the lack of extant plants at these sites (compare these sites with the points labelled Z in Fig. 1). We address this discrepancy below.

While direct measurements of dissolved Cu concentrations in seawater were not a part of this study, data are available from two related studies. Dissolved Cu levels in surface layers were measured by Grout and Levings (2001) at a series of sites on the shores of Howe Sound from April to June 1998 (Fig. 2B). These measurements provide an estimate of the decrease in Cu levels with increasing distance from Britannia Creek. In 1994, Chretien (1997) measured dissolved Cu levels each month along a transect extending seaward from the mouth of Britannia Creek (Fig. 2A). While these data were collected well before the present study, the Cu concentrations in Chretien (1997) were likely similar to those during our study because the mine's discharge is primarily determined by seasonal factors (Chretien, 1997). When the two data sets were combined, using linear interpolation between time points in Chretien's (1997) data, we obtained an estimate of the average Cu concentration present for the duration of each transplant (Table 1). These estimates are consistent with the results: the November and February transplants, which showed high mortality and low or negative growth rates in rockweed moved to Britannia Creek, were exposed to average dissolved Cu concentrations that were 3–60 times greater than those to which the June and August transplants were exposed.

The results of the experiments, together with the estimated dissolved Cu levels, can also be compared to published work that has examined the effects of Cu on algae in laboratory studies. Strömgren (1979) exposed *Ascophyllum nodosum* to a variety of concentrations of Cu in a flowing seawater system. He found that this species' growth rate was decreased to 80% compared to controls when exposed to 66 μg Cu l^{-1} for four days, while growth rates of plants exposed to 340 μg Cu l^{-1} were reduced to 20% of controls after four days. The same author also examined the effect of Cu on other fucoid algae (Strömgren, 1980). He observed a significant reduction of growth rate of *Pelvetia canaliculata* (L.) Decaisne & Thuret and *Fucus spiralis* (L.) at 12 μg Cu l^{-1}, of *Fucus serratus* (L.) at 25 μg Cu l^{-1}, and of *F. vesiculosus* at 50 μg Cu l^{-1}. By comparison, we found no reduction in *F. gardneri* growth rate relative to control plants at estimated dissolved Cu levels up to 16 μg Cu l^{-1}. Such comparisons should be interpreted with

Table 1
Estimated concentrations of dissolved copper in surface water at Britannia Creek sites during transplant experiments

Study dates	Site	Distance and direction from creek mouth	Est. Diss. Cu at BB (μg Cu l^{-1})	Est. Diss. Cu at FC (μg Cu l^{-1})
02 Nov 1997–08 Jan 1998	5	50–100 m South	270	2.7
15 Feb 1998–03 Apr 1998	5	50–100 m South	50	0.5
11 Jun 1998–25 Aug 1998	6	300 m South	nd	5.4
11 Jun 1998–25 Aug 1998	7	600 m South	4.6	5.4
10 Aug 1998–18 Nov 1998	4	300 m North	16	1.4
10 Aug 1998–18 Nov 1998	3	700 m North	10	1.4

Calculated using data from Chretien (1997) and Grout and Levings (2001), nd: no data.

caution, however, as differences between plant species and water chemistry in the two experiments may well exert considerable influence on the results observed.

One factor that may confound the results of the June and August 1998 experiments is the harsh conditions to which transplanted rockweed were exposed. Howe Sound has a mixed semi-diurnal tide pattern and during the summer months the lowest tides occur near midday. The summer of 1998 was hot and sunny in upper Howe Sound, resulting in severe desiccation of rockweed thalli, often on a daily basis (pers. obs.). As an index of the heat and desiccation to which the plants would have been subjected, we calculated degree–hour values for each transplant (Fig. 7). These values show that the June and August 1998 transplants were subjected to much higher heat and desiccation stress than the first two transplants. Heavy mortality of rockweed was observed at many areas in Howe Sound in the latter half of 1998 (Marsden and DeWreede, 2000). Furthermore, Thom (1983) observed a decrease in *Fucus distichus* ssp. *edentatus* (de la Pyl.) Pow. cover during summer in Puget Sound, Washington, USA, which he attributed to high temperature, heavy sun exposure, and sparse precipitation. This raises the possibility that any effects of AMD that may otherwise have been detected were overshadowed by the much greater impact of "background" mortality, which impacted both AMD-exposed and control plants.

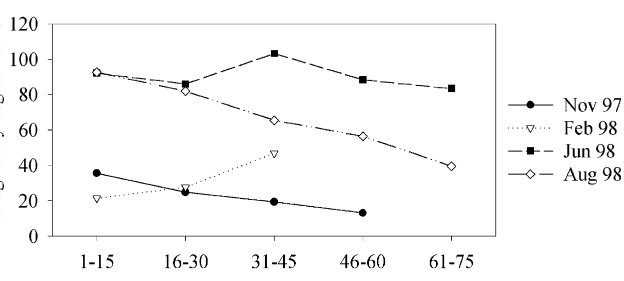

Fig. 7. Degree–hours to which transplants were exposed during experiments, expressed as daily averages for 15-day segments of each experiment. Degree–hours are calculated by multiplying the number of hours the tide was below the elevation of the plant (based on predicted tides from X-Tide, http://tbone.biol.sc.edu/tide) by the temperature that day (daily high temperature for low tides from 1200 to 1600, daily low for low tides from 2400 to 0400, daily mean for all other times; data from an Environment Canada weather station, 9 m above sea level near Furry Creek).

4.2. Cu tissue concentration

Rockweed that were transplanted to sites BB-5a and -5b had Cu levels 2–3 times those of the nearest extant plants (~500–900 ppm in dry weight at sites BB-2 and -7: Dunn et al., 1992; Marsden and DeWreede, 2000) after just 11 days. Given that the levels of Cu found in the extant plants nearest to Britannia Creek likely represents an approximate maximum that the plants can withstand in the long term, severe degradation of the plants' health at >2000 ppm Cu is expected.

Some work has been done elsewhere involving the transplantation of algae into areas with relatively high Cu concentrations and the measurement of uptake. Ho (1984) moved *F. vesiculosus* and *A. nodosum*, two fucoid algae, from an unpolluted site to an estuary which was contaminated with Zn and Cu from past mining activities. Ho (1984) observed Cu concentrations of <20 ppm in dry weight in untransplanted *F. vesiculosus*, while those moved to the polluted area contained 100, 150 and 230 ppm Cu after 15, 35 and 57 days, respectively. *A. nodosum* contained 60, 60 and 100 ppm Cu at the same time intervals, while control plants contained <35ppm Cu. These concentrations of Cu in *F. vesiculosus* are 3–10% of those we observed in our experiments, possibly due to lower concentrations of available Cu, although Ho (1984) did not report dissolved Cu concentrations. However, interspecific differences between *A. nodosum* and *F. vesiculosus* are apparent within Ho's experiment; such differences may also be important when comparing Northeast-Pacific *F. gardneri*, an intertidal plant, with Northeast-Atlantic *F. vesiculosus*, a subtidal plant. Forsberg et al. (1988) moved *F. vesiculosus* from an unpolluted site to a relatively polluted site near Stockholm, Sweden. After 12 months there was significantly more Cu in old tissues at the polluted site, but no significant change in Cu concentration in young tissues. Ambient concentrations of Cu in the two areas were not reported.

4.3. Changes in colour and texture

The change in colour and texture of the plants soon after the November 1997, February 1998 and June 1998 transplants to the mouth of Britannia Creek appeared to be an indication of the quickly declining health of the plants. The change in colour was consistent with Sorentino's (1979) finding that very high concentrations of Cu cause degradation of chlorophyll and other pigments in some algae, which can lead to a change in colour. A change in plant texture may be a result of Cu binding to cell proteins such as those that maintain the plant's structure and control the osmotic permeability of the cell (Sorentino, 1979). The increased production of exudates may be one way the plant has of detoxifying the metals (Ragan et al., 1980; Schramm, 1993). However, further work would be required to determine if the observed changes were related to exposure to AMD. The fact that plants transplanted to site BB-6 in June 1998 exhibited these changes, combined with significant decreases in growth rate in these plants (Fig. 5) suggests that there may have been some degree of sublethal AMD impact on the plants.

4.4. General discussion

The results provide strong correlative evidence for the role of AMD in excluding rockweed from the immediate vicinity of Britannia Creek. Plants moved close to the AMD source deteriorated quickly, while control plants and those moved to more distal sites showed no such putative effects. Estimated concentrations of dissolved Cu provide strong evidence of the presence of AMD at the sites where deterioration occurred. Uptake of Cu in one of the experiments to concentrations 2–3 times the highest levels seen in surrounding plants indicate that Cu likely played a strong role in the observed deterioration.

Given the lack of detectable decline in summer transplants to sites close to the creek mouth (sites BB-3, -4, -6 and -7), however, there is only partial explanation herein for the lack of extant plants at these sites. One possible explanation for the absence of rockweed at these sites would invoke different effects of AMD on adult and juvenile stages of the plants. Several authors have investigated the effects of Cu on algal propagules and reproduction, and found that it has negative effects at concentrations as low as 10 $\mu g\,l^{-1}$ (Chung and Brinkhuis, 1986; Anderson et al., 1990; Scanlan and Wilkinson, 1987). In a study on Baltic Sea *F. vesiculosus*, Andersson and Kautsky (1996) found that at salinities higher and lower than optimal, germination declined by 70–80% when exposed to 20 μg $Cu\,l^{-1}$. Plants at the optimal salinity showed no such reduction. These studies suggest that juvenile stages and reproductive processes may be more susceptible to Cu than their adult counterparts (see above discussion of Strömgren, 1979, 1980), especially at non-optimal salinites such as those to which rockweed in Howe Sound are exposed. Direct examination of this possibility, through field or laboratory studies, would shed further light on the mechanisms producing the observed rockweed distribution. Other components of the AMD, such as Zn, Cd and acidity, which were not the focus of this study, may also play a role in excluding rockweed.

An alternative explanation for the lack of rockweed at Britannia Creek relative to Furry Creek might be the higher turbidity and lower salinity the plants would encounter at the former location due to its proximity to the Squamish River, which dominates the salinity regime in upper Howe Sound during freshet. However, all other creek mouths observed in upper Howe Sound, including Furry, Potlatch and Woodfibre Creeks (see Fig. 1 for locations), are dominated by a thick cover of rockweed (pers. obs.). Extremely dense populations of rockweed were also observed to the north of Britannia Creek at Watts Point (pers. obs.). The Woodfibre Creek area encounters higher turbidity and lower salinity than Britannia Beach during freshet (Grout et al., 1999), but has rockweed cover in excess of 80% in many places (pers. obs.). Salinity and turbidity, therefore, do not appear to be viable explanations for the lack of rockweed at Britannia Beach.

While the results of this work point to AMD as the primary factor determining the distribution rockweed in the vicinity of Britannia Creek, it is not possible to attribute causation to any single component of the effluent. We have used literature values of dissolved Cu as a "tracer" of AMD, but the deterioration observed in rockweed transplants to Britannia Creek may be caused by a combination of all components of AMD: Cu, Zn, Cd and acidity. For the reasons discussed in the introduction, however, Cu is likely the most significant component of the drainage influencing the distribution of rockweed in this region of Howe Sound.

Acknowledgements

Funding for this work was provided by the Department of Fisheries and Oceans (Toxic Chemicals Program and Science Branch, Pacific Region) to ADM, and by the National Science and Engineering Research Council (grant #5-89872) to RED. Dr. Colin Dunn of the Geological Survey of Canada kindly arranged for analysis of plants for Cu tissue burden. Drs. Elanor Bell, Al Lewis and Terri Sutherland provided helpful comments on the work. Special thanks go to the many workers and volunteers who helped during the field portion of this study, especially to Christy Stesky and Annabel Forsythe, who did the majority of the data collections on the February 1998 transplant.

References

Anderson, B.S., Hunt, J.W., Turpen, S.L., Coulon, A.R., Martin, M., 1990. Copper toxicity to microscopic stages of giant kelp *Macrocystis pyrifera*: interpopulation comparisons and temporal variability. Marine Ecology Progress Series 68, 147–156.

Andersson, S., Kautsky, L., 1996. Copper effects on reproductive stages of Baltic Sea *Fucus vesiculosus*. Marine Biology 125, 171–176.

Barry, K.L., Grout, J.A., Levings, C.D., Nidle, B.H., Piercey, G.E., 2000. Impacts of acid mine drainage on juvenile salmonids in an estuary near Britannia Beach in Howe Sound, British Columbia. Canadian Journal of Fisheries and Aquatic Sciences 57, 2032–2043.

Castilla, J.C., 1996. Copper mine tailing disposal in northern Chile rocky shores: *Enteromorpha compressa* (Chlorophyta) as a sentinel species. Environmental Monitoring & Assessment 40, 171–184.

Chretien, A.R.N., 1997. Geochemical behaviour, fate and impacts of Cu, Cd and Zn from mine effluent discharges in Howe Sound. Ph.D. Thesis, University of British Columbia, Vancouver, Canada.

Chung, I.K., Brinkhuis, B.H., 1986. Copper effects in early stages of the kelp, *Laminaria saccharina*. Marine Pollution Bulletin 17, 213–218.

Dunn, C.E., Percival, J.B., Hall, G.E.M., Murdoch, A., 1992. Reconnaissance geochemical studies in the Howe Sound drainage basin. In: Levings, C.D., Turner, R.B., Ricketts, B. (Eds.), Proceedings of the Howe Sound environmental science workshop. Canadian Technical Report on Fisheries and Aquatic Science #1879, pp. 89–99.

Forsberg, A., Söderlund, S., Frank, A., Petersson, L.R., Pedersén, M., 1988. Studies on the metal content in the brown seaweed, *Fucus vesiculosus*, from the Archipelago of Stockholm. Environmental Pollution 49, 245–263.

Gledhill, M., Nimmo, M., Hill, S.J., 1997. The toxicity of copper (II) species to marine algae, with particular reference to macroalgae. Journal of Phycology 33, 2–11.

Grout, J.A., Levings, C.D., 2001. Effects of acid mine drainage from an abandoned copper mine, Britannia mines, Howe Sound, BC, on transplanted blue mussels (*Mytilus edulis*). Marine Environmental Research 51, 265–288.

Grout, J.A., Levings, C.D., Nidle, B., Piercey, B., Marsden, D., 1998. Beach seine data from near Britannia mines and in Howe Sound, British Columbia, during 1997. Canadian Data Report of Fisheries and Aquatic Sciences #1044.

Grout, J.A., Levings, C.D., Piercey, G.E., Mossop, B., 1999. Biological data from near Britannia mine and in Howe Sound, British Columbia, during 1997–1998. Canadian Data Report of Fisheries and Aquatic Sciences #1055.

Hargreaves, J.W., Whitton, B.A., 1976. Effect of pH on tolerance of *Hormidium rivulare* to zinc and copper. Oecologia 26, 235–243.

Ho, Y.B., 1984. Zn and Cu concentrations in *Ascophyllum nodosum* and *Fucus vesiculosus* (Phaeophyta, Fucales) after transplantation to an estuary contaminated with mine waters. Conservation & Recycling 7, 329–337.

Levings, C.D., McDaniel, N.G., 1976. Industrial disruption of invertebrate communities on beaches in Howe Sound, BC. Fisheries and Marine Service Technical Report #663.

Levings, C.D., Riddell, B.E., 1992. Salmonids and their habitats in Howe Sound basin: status of knowledge. In: Levings, C.D., Turner, R.B., Ricketts, B., (Eds.), Proceedings of the Howe Sound Environmental Science Workshop. Canadian Technical Report on Fisheries and Aquatic Science, #1879, pp. 65–81.

Marsden, A.D., DeWreede, R.E., 2000. Marine macroalgal community structure, metal content and reproductive function near an acid mine drainage outflow. Environmental Pollution 110, 431–440.

Munda, I.M., Hudnik, V., 1986. Growth response of *Fucus vesiculosus* to heavy metals, singly and in dual combinations, as related to accumulation. Botanica Marina 29, 401–412.

Nassichuk, M.D., 1975. Structural and interactive relationships between intertidal *Fucus* populations and associated faunal assemblages. M.Sc. Thesis, University of British Columbia, Vancouver, Canada.

Price, W.A., Schwab, T., Hutt, N., 1995. A reconnaissance study of acid mine drainage at the Britannia mine. British Columbia Ministry of Energy, Mines and Petroleum Resources, Victoria, Canada.

Ragan, M.A., Ragan, C.M., Jensen, A., 1980. Natural chelators in sea water: detoxification of Zn^{2+} by brown algal polyphenols. Journal of Experimental Marine Biology and Ecology 44, 261–267.

Scanlan, C.M., Wilkinson, M., 1987. The use of seaweeds in biocide toxicity testing. Part 1. The sensitivity of different stages in the life-history of *Fucus*, and of other algae, to certain biocides. Marine Environmental Research 21, 11–29.

Schramm, W., 1993. Investigations on the influence of organic substances produced by seaweeds on the toxicity of copper. In: Rijstenbil, J.W., Haritonidis, S., (Eds.), Macroalgae, eutrophication and trace metal cycling in estuaries and lagoons. Proceedings of the COST 48 Symposium, Thessaloniki, Greece, pp. 106–120.

Sorentino, C., 1979. The effects of heavy metals on phytoplankton—A review. Phykos 18, 149–161.

Strömgren, T., 1979. The effect of copper on length increase in *Ascophyllum nodosum* (L.) Le Jolis. Journal of Experimental Marine Biology and Ecology 37, 153–159.

Strömgren, T., 1980. The effect of dissolved copper on the increase in length of four species of intertidal fucoid algae. Marine Environmental Research 3, 5–13.

Thom, R.M., 1983. Spatial and temporal patterns of *Fucus distichus* ssp. *edentatus* (de la Pyl.) Pow. (Phaeophyceae: Fucales) in Central Puget Sound. Botanica Marina 26, 471–486.