Generation and treatment of acid mine drainage: Correlating bacterial populations with water chemistry

S. G. Benner
D. W. Blowes

Department of Earth Sciences, University of Waterloo, Waterloo, Ontario, N2L 3G1, Canada

W. D. Gould

CANMET Natural Resources Canada, Ottawa, K1A 0G1, Canada

The oxidation of sulphide minerals in mines and mine waste generates high concentrations of dissolved Fe and SO_4. These constituents often are transported downwards into the underlying groundwater system and can discharge to the surface. Along the flow path from the zone of recharge to discharge a series of oxidation reactions occur that produce acidic waters that can adversely impact the surface water ecosystem. In this study we correlated observed changes in water chemistry with populations of bacteria associated with those changes.

Tailings at the Nickel Rim Mine site have been oxidizing for 30 years. Effluent entering the underlying aquifer contains 3000 to 10,000 mg/L SO_4, 500 to 4000 mg/L Fe, and pH of between 2 and 4 and forms a groundwater plume enriched in SO_4 and Fe. Aqueous parameters vary along the flow path; the higher Fe and SO_4 concentrations and lower pH are within the unsaturated and shallow saturated zone of the tailings and solute concentrations decrease and pH increases down-gradient (Bain *et al.*, 1998). About half the groundwater emanating from the tailings discharges to the surface at the base of the tailings dam while the remainder flows within the aquifer for 300 m prior to discharge to a small lake (Fig. 1). The water along this longer flow path is now undergoing passive treatment by passage through a permeable reactive barrier installed into the aquifer about half way between the tailings dam and the second zone of discharge at the lake. The permeable reactive barrier is designed to treat mine drainage effluent by bacterially-mediated sulphate reduction.

We determined pore water chemistry and characterized populations of bacteria associated with sulphur and iron redox transformations along these two flow paths. Flow path (A) typifies de-coupled sulphide oxidation with oxidation of primarily sulphur in the recharge zone and oxidation of ferrous iron, Fe (III) hydrolysis and precipitation of ferric hydroxides at the discharge zone. Flow path (B) begins with oxidation of sulphides in the zone of recharge but then flows through the sulphate-reducing, permeable reactive barrier. Water chemistry indicates that major changes in Fe and SO_4 oxidation states and concentrations occur at redox interfaces. Corresponding bacterial activity is high in those locations where a redox interface exists and is low in those areas of the flow field where these electrochemically active species are at apparent equilibrium. Water chemistry was determined and sulphur oxidizing, sulphur reducing, and iron oxidizing bacteria were characterized and values were averaged for each of the following zones: the oxidized tailings, the tailings undergoing active oxidation, the aquifer, the zone of discharge at the base of the dam, and the permeable reactive barrier.

Methods

Monitoring wells were installed in nests along a transect roughly parallel to groundwater flow. Samples of groundwater were collected in July 1997. A detailed description of well installation, water sampling and field and laboratory analysis can be found in Benner *et al.* (1997). Sediment samples were collected using a 5 cm driven coring device. Cores were collected in July 1997 for bacterial analysis. Cores were sealed and refrigerated at 4°C until analysis.

The sulphate reducing bacteria were grown in a modified Postgate medium C in 20 mL serum bottles. Serum bottles containing medium were autoclaved and 1 gm sediment was added to each of five replicates. Inoculated samples were sequentially diluted and incubated in an anaerobic glove box for 30 days. Positive growth of SRB was indicated by precipitation of Fe-sulphides. Values are reported as Most Probable Number (MPN) determinations. *Thiobacillus ferrooxidans* were grown in an media consisting of 0.5 g of K_2HPO_4, 0.5 g of $(NH_4)_2SO_4$, 0.5 g of $MgSO_4·H_2O$, and 33.4 g of $FeSO_4·7H_2O$ per L water. The media without ferrous sulphate was adjusted to pH 3.0 and autoclaved. The ferrous sulphate solution with a pH of 3.0 was prepared, filter

sterilized, and then the two solutions were combined and pH adjusted to 2.2. A five-tube MPN method was used, the samples were incubated for 4 weeks, and a positive result was indicated by a presence of oxidized iron in the tube. *Thiobacillus thiooxidans* were grown in Medium B (ATCC medium 23) containing 0.1 g NH_4Cl, 3.0 g KH_2PO_4, 0.2 g $MgCl_2·6H_2O$, 5 g $Na_2S_2O_3$ $5H_2O$, and 0.1 g $CaCl_2$ per L water. The pH was adjusted to 4.2, a five-tube MPN method was used, and the tubes were incubated for 4 weeks. A positive result was indicated by the presence of a sulphur deposit and a pH < 4.

Results and discussion

Within the upper metre of the tailings, sulphide oxidation is nearly complete but residual pore water is still acidic (ave. pH = 3.1), and contains high concentrations of SO_4 (ave. 470 mg/L) and low concentrations of Fe (<5 mg/L) (Johnson *et al.*, 1998). Within this part of the tailings, populations of measured *Thiobacillus sp.* are low, supporting the conclusion that little active sulphide oxidation occurs within this zone.
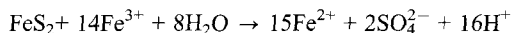
The zone of active oxidation within the tailings is located within about 20 cm of the water table. At this location, concentrations of Fe and SO_4 increase to average values of 540 mg/L and 3100 mg/L, respectively. The pH increases slightly to an average value of 3.6. The observed increases in Fe and SO_4 concentrations are the result of sulphide oxidation which can be expressed for pyrite as:

$$FeS_2 + 7/2O_2 + H_2O \rightarrow Fe^{2+} + 2SO_4^{2-} + 2H^+$$
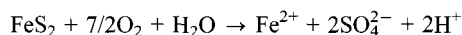
Oxidation of ferrous to ferric iron also occurs:

$$2Fe^{2+} + 1/2O_2 + 2H^+ \rightarrow 2Fe^{3+} + H_2O$$

Ferric iron can then directly oxidize sulphide pyrite by:

$$FeS_2+ 14Fe^{3+} + 8H_2O \rightarrow 15Fe^{2+} + 2SO_4^{2-} + 16H^+$$

Thiobacillus thiooxidans are highest in this zone, with populations averaging 1.3×10^3 MPN/g. Populations of *T. ferrooxidans* are also elevated to average of 7.4×10^2. *T. ferrooxidans* can oxidize ferric iron and reduced sulphide species, while *T. thiooxidans* can oxidize intermediate sulphide species. Although populations of *T. thiooxidans* are highest in this zone compared to the other sites in the aquifer, the values are low compared to those measured at other tailings impoundments (Blowes *et al.*, 1995). The high degree of saturation within the
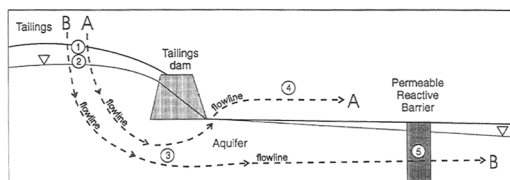
zone of active oxidation at the Nickel Rim site results in relatively low rates of sulphide oxidation compared with other, less mature, tailings impoundments (Johnson *et al.*, 1998).

Below the zone of active oxidation, concentrations of Fe and SO_4 gradually increase, reflecting the oxidative history at the site, but do not exhibit rapid changes indicative of changing redox conditions. Populations of all measured bacterial groups within this part of the aquifer are low. Low populations of the acidiphilic *Thiobacillus sp.* are expected given the moderate pH. Low SRB populations may indicate the lack of suitable organic substrates to promote sulphate reduction.

At the point of discharge, Fe concentrations decrease to an average of 360 mg/L and pH declines to an average of 2.8. Within the surface sediments at the point of discharge, populations of *T. ferrooxidans* are high (average of 9.6×10^5 MPN/g) and populations of SRB and *T. thiooxidans* are low. The declines in pH and Fe concentration are due to the oxidation of ferrous to ferric iron and the subsequent precipitation of ferric oxyhydroxides:

$$2Fe^{2+} + 1/2O_2 + 5H_2O \rightarrow 2Fe(OH)_{3(s)} + 4H^+$$
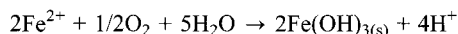
Finally, water entering the permeable reactive barrier exhibits declines in Fe (to average 84 mg/L), SO_4 (average 940 mg/L) and an increase in pH to an average of 6.7. These changes are driven by sulphate reduction and iron sulphide precipitation (Benner, *et al.*, in preparation) which can be expressed as:

$$SO_4^{2-} + 2CH_2O + 2H^+ \rightarrow H_2S + 2CO_{2(aq)} +2H_2O$$
$$Fe^{2+} +H_2S \rightarrow FeS_{(s)} + 2H^+$$
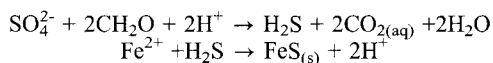
Within the permeable reactive barrier, populations of SRB are very high (average 3.7×10^7 MPN/g) while *T. ferrooxidans* and *T. thiooxidans* are low. These high SRB values suggest that active sulphate reduction is occurring within the reactive barrier.

FIG. 1. Schematic diagram showing idealized flowlines (A) and (B) and approximate locations of water chemistry and bacterial populations.