

TREATMENT OF METAL-CONTAMINATED WATER USING
BACTERIAL SULFATE REDUCTION:
RESULTS FROM PILOT-SCALE REACTORS¹

by

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Abstract. Pilot-scale biological reactor systems were installed to treat metal-contaminated water in an underground coal mine and at a smelting residues dump in Pennsylvania. The reactors consisted of barrels and tanks filled with spent mushroom compost, within which bacterial sulfate reduction became established. Concentrations of Fe, Zn, Mn, Ni, and Cd were lowered by over 95% as these metals were precipitated in the reactors. The formation of insoluble metal sulfides by reaction with bacterially generated H₂S was identified as an important metal retaining process in the reactors. This paper examines the chemistry of the reactor systems and opportunities for enhancing their metal-retaining and alkalinity-generating potentials.

Additional keywords: anaerobic reactors, heavy metal sulfides, spent mushroom compost.

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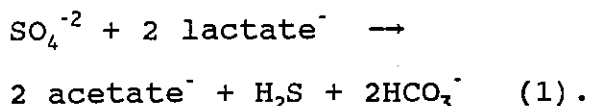
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Introduction

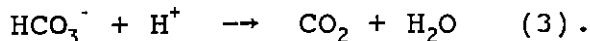
Bacterial sulfate reduction has been identified as a potentially valuable process for removing contaminant metals from coal and metal-mine drainage (Tuttle et al., 1969; Hedin et al., 1988; Wildeman and Laudon, 1989). Under anaerobic conditions, sulfate-reducing bacteria oxidize simple organic compounds with sulfate, and thereby generate hydrogen sulfide and bicarbonate ions:



Hydrogen sulfide reacts with many contaminant metals to form insoluble metal sulfides:



where M includes metals such as Fe, Zn, Ni, Cd, Cu, and Pb. Bicarbonate ions can consume protons to raise the pH of acidic water:



Hammack and Hedin (1989) and McIntire et al. (1990) experimented with simple water treatment reactors consisting of carboys and columns filled with organic matter in which sulfate reduction became established. Their reactors lowered iron, nickel, and manganese concentrations in synthetic mine drainage by 60 to 90%. These results led us to construct pilot-scale reactors of a similar design to

evaluate their potential for treating metal-contaminated water. The design of these sulfate reduction reactors required:

- 1) exclusion of oxygen,
- 2) a source of sulfate (commonly present in contaminated water),
- 3) a source of simple organic compounds to serve as a bacterial carbon source, and
- 4) a means by which to physically retain the metal sulfide precipitates.

Low pH (<5) inhibits sulfate-reducing bacteria (Postgate, 1984) and increases the solubility of metal sulfides, which required that the reactors also be designed to generate sufficient alkalinity to raise the pH of acidic inflows.

In this paper we present our initial findings on the performance and chemistry of the pilot-scale reactors, and discuss some strategies for enhancing their performance.

Methods

Two pilot-scale reactor systems were constructed in 1990. The reactors in each system consist of either capped barrels (Pittsburgh system) or covered tanks (Palmerton system), filled with loosely-packed spent mushroom compost. Spent mushroom compost consists of a composted mixture of manure, hay, straw, corn cobs, and wood chips that has been conditioned with gypsum and limestone, and used for

cultivating mushrooms. Since it decomposes readily, the compost serves as a source of organic carbon for bacterial sulfate reduction, and its bulk form serves to physically detain any precipitated solids. The different mushroom composts we used in each system contained 50-60 wt% organic matter and 10-15 wt% pulverized limestone (dry wt basis).

The Pittsburgh reactor system was installed to treat acidic, iron-contaminated drainage within the Experimental Mine at the U.S. Bureau of Mines, Pittsburgh Research Center. This system consists of three 200 L reactor barrels plumbed in series and receiving water from a 1140 L supply tank (see Figure 1[a]). Barrels were used instead of a single large reactor because of height limitations within the coal mine. The underground location of this system maintained a nearly constant temperature of 10°C, permitting operation during the winter.

The Palmerton reactor system was installed to treat metal-contaminated drainage from a smelting residues dump at the former New Jersey Zinc Company plant in Palmerton, Pennsylvania. This system consists of 2 independent, 4,500 L reactor tanks receiving water from a 3,500 L supply tank (see Figure 1[b]). The data presented for this system were collected during summer operation, when average ambient temperature ranged between 18 and 24°C.

Table 1 shows the quantity of compost contained in each system and the mean flow rates and residence times at which

each system was operated. As our primary objective was to evaluate the chemistry and water treatment potential of the reactor systems, we used residence times at which we expected significant metals removal to occur. In the Palmerton system, the flow rate through one reactor (reactor 2) was doubled after 10 weeks of operation to observe the effects of a shortened residence time.

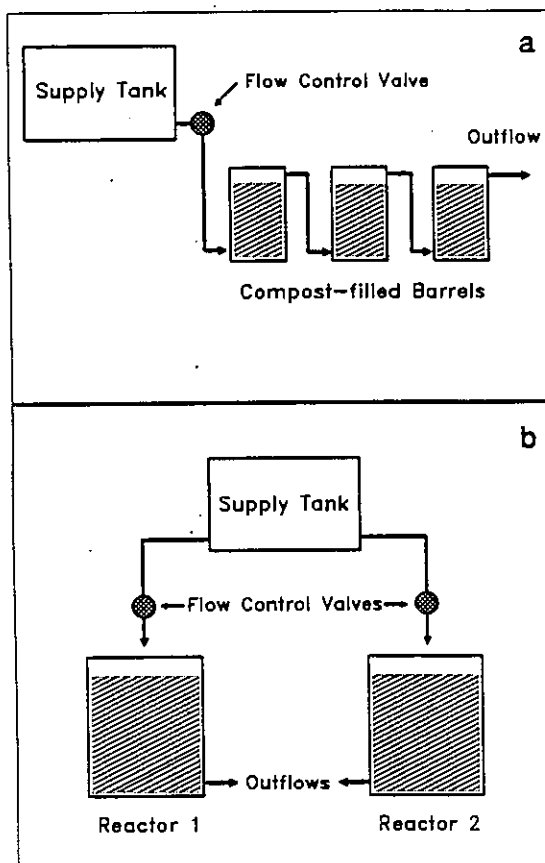


FIGURE 1

Figure 1. Schematics of the Pittsburgh (a) and Palmerton (b) Reactor Systems.

Water Sampling and Analysis

Influent, effluent, and occasional porewater samples

Table 1. Selected physical characteristics of reactor system design and operation

	Pittsburgh System	-- Palmerton System -- Reactor 1 Reactor 2	
Volume of compost, L	624	3038	3038
Dry mass of compost, kg	68	572	572
Mean flow rate, ml/min	55	70	70/131 ¹
Mean residence time, days	5	17	17/9 ¹

Flow rate increased (residence time decreased) four weeks after the leaching phase.

were collected at 1-2 week intervals. Samples for metals analysis were acidified immediately upon collection (total metals) or following filtration through a 1.2 μm glass fiber filter (dissolved metals). Metal concentrations were determined using an ICP atomic emission spectrophotometer, except for Cd (atomic absorption spectrometry) and ferrous iron (titration with $\text{K}_2\text{Cr}_2\text{O}_7$). Dissolved sulfide was determined using an ion-specific electrode on samples preserved with NaOH. Sulfate was measured using a liquid chromatograph coupled with a conductivity detector, and alkalinity was determined by titration with 0.2 N H_2SO_4 to pH 4.5. Whenever possible, dissolved sulfide, alkalinity, and pH were determined on the same day the samples were collected.

Compost Sampling and Analysis

The spent mushroom compost used in each system was sampled before being placed in the

reactors, and after 17-18 weeks of system operation. The 18-week samples from the Pittsburgh system were collected from depths of 10 cm and 60 cm in each of the 3 reactor barrels (maximum depth = 75 cm). The 17-week samples from the Palmerton system were collected from a depth of 60 cm (maximum depth = 135 cm) at 2 locations within each reactor. At each depth or location, a composite sample of 3-6 subsamples was collected by hand and immediately sealed in an argon-filled container.

The total content of inorganic cations and anions was determined by ashing subsamples in a muffle furnace at 550°C for 6 hours, and analyzing an HCl extract of the ash. A residual fraction consisting primarily of silica and clay minerals remained undissolved by this procedure. Calcium carbonate content was deduced from discrepancies in the mass and charge balances revealed by a comprehensive analysis of the ash extract. Sample moisture content was determined at 112°C.

The total iron content was expected to consist of a "fixed" fraction, tightly bound within organic matter and resistant clay minerals, and a "labile" fraction, occurring in forms, such as FeOOH, that are soluble under acidic and reducing conditions. Because of its potential solubility, the labile iron would be susceptible to leaching, and would be available for reaction with H₂S. The labile iron content of the compost samples from the Pittsburgh system was determined by extracting subsamples with 4 N HCl under an argon sparge, and analyzing the extract for iron.

A single subsample from the 10 cm depth in the first reactor barrel of the Pittsburgh system was examined by Mossbauer spectroscopy (Willard et al., 1974) to directly identify the iron minerals present.

Acid-volatile sulfide (AVS), present as monosulfides such as FeS and ZnS, and chromium-reducible sulfur (CRS), present as pyrite (FeS₂) and elemental sulfur, were determined by anoxic acid distillation (Cornwell and Morse, 1987). The H₂S was liberated with 6 N HCl, and trapped in 0.2 N NaOH.

Results

Following start-up, both systems underwent an initial phase during which Ca, Mg, Na, K, and sulfate ions were leached from the compost. By the time this phase was completed, effluent color had changed from dark brown to yellow; the effluent acquired a strong hydrogen sulfide odor;

effluent concentrations of magnesium, sodium, and potassium had fallen to influent levels; and net sulfate consumption (reduction) and metal retention became discernible. The initial leaching phase was complete for the Pittsburgh system after 3.5 weeks of operation and 3940 L of flow, and for the Palmerton system after 6 weeks of operation and 4140 L of flow.

The results presented below pertain to system performance following the initial leaching phase. For the Pittsburgh system, a 14-week period involving 6110 L of flow is considered, and for the Palmerton system, a 16-week period involving 11,000 L (reactor 1) to 19,300 L (reactor 2) of flow is considered.

Changes in Water Chemistry

Mean influent and effluent water analysis results are shown for the Pittsburgh system in Table 2, and for the Palmerton system in Table 3. Both systems lowered contaminant metal concentrations by greater than 95%, lowered sulfate concentrations by about 30%, and produced circumneutral effluents having high alkalinity. The effluents from both systems contained high concentrations of dissolved sulfide (HS⁻ and H₂S), which, coupled with the lowering of sulfate concentrations, indicates that bacterial sulfate reduction was occurring.

Both systems continued to release calcium after leaching of the other base cations had

Table 2. Chemical analysis of influent and effluent water for the Pittsburgh reactor system.

	Influent	Effluent
pH	3.7	6.9
Alkalinity, as CaCO ₃	----- 17 ¹	mg/L ----- 632
Total Fe	67	<0.2
Ferrous Fe	37	<0.2
Ferric Fe	10	nd
Suspended Fe	20	nd
Dissolved Al	7	<0.2
Ca	294	518
Sulfate	973	712
Dissolved sulfide	nd	34

Values are means of 8 influent and 11 effluent samples;
nd = not detectible;

¹ Alkalinity present only in samples with pH >4.5 (n=2).

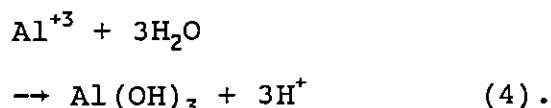
Table 3. Chemical analysis of influent and effluent water for the Palmerton reactor system

	Influent	Effluent
pH	6.3	6.9
Alkalinity, as CaCO ₃	----- 17	mg/L ----- 1102
Zn	302	0.42
Mn	24	0.50
Ni	0.85	0.03
Cd	0.29	<0.005
Ca	275	720
Sulfate	2989	2352
Dissolved sulfide	nd	109

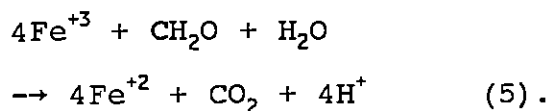
Values are means of 9 influent and 18 effluent samples;
nd = not detectible

ceased. Possible origins for this calcium include the dissolution of limestone (a constituent of the spent mushroom compost) and the decomposition of calcium-rich organic matter.

The Pittsburgh system consistently lowered aluminum concentrations to less than 0.2 mg/L. Since aluminum does not form a stable sulfide in the presence of water, this removal likely resulted from its hydrolysis to insoluble $\text{Al}(\text{OH})_3$:

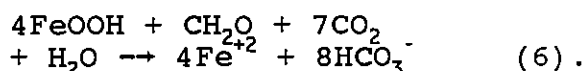


This reaction is spontaneous at the circumneutral pH prevailing within the reactor barrels (Stumm and Morgan 1981). Ferric iron also hydrolyzes at circumneutral pH, but in the anaerobic environment within the barrels, it apparently was reduced to ferrous iron, which requires a much higher pH for hydrolysis. This reduction probably occurred in conjunction with the microbial oxidation of organic compounds ("CH₂O"):



Ferric iron was never detected in water that had passed beyond 7% of the total system volume, though ferrous iron concentrations usually remained high beyond that point. Also, suspended ferric oxyhydroxide (FeOOH) did not visibly accumulate in the inflow zone of the first reactor barrel,

which suggests that influent FeOOH was similarly reduced and dissolved:



Analyses of pore water from the Pittsburgh system revealed that essentially all of the influent iron was removed by the first reactor barrel. Also, very little dissolved sulfide and alkalinity left the first barrel, thereby leaving the second and third reactor barrels to generate the high levels of dissolved sulfide and alkalinity observed in the system effluent.

Changes in Compost Composition

Table 4 shows the results of the analyses of Pittsburgh system compost. Total iron content for the second and third reactor barrels did not change significantly during the course of this study. This is consistent with the results of the water analyses, which detected very little iron leaching from the system, and which indicated that the influent iron was being removed entirely in the first barrel. Since little iron left the system, most of the original 63 μmoles of labile Fe/g compost was retained within the reactors.

Comparison of total iron contents indicates that the first reactor barrel accumulated 90 μmoles of Fe/g compost from the influent water. By adding this accumulated iron to the 63 $\mu\text{moles/g}$ of retained labile iron, a total retained iron content of 153 $\mu\text{moles/g}$ is

Table 4. Chemical analysis of the Pittsburgh system compost

	Before operation	----After 18 weeks----	
		Barrel 2-3	Barrel 1
	-----μmoles/g dry wt-----		
Total Fe	136	133	226
Labile Fe	63	nm ¹	nm ¹
AVS	<0.1	69	150
CRS	23	140 ²	140 ²
CaCO ₃	999	708	547

Values are means of 4 to 12 samples;

¹ Not measured directly--probably equal to corresponding AVS;

² System average.

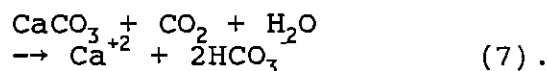
obtained. This closely matches the acid-volatile sulfide (AVS) content of 150 μmoles/g that was found in the first barrel, thus suggesting that iron was retained primarily as ferrous monosulfide (FeS). In agreement with this, Mossbauer spectroscopy identified FeS as the predominant iron form present.

Note that, despite the absence of net iron accumulation in the second and third reactor barrels, AVS still accumulated there, evidently due to the reaction of H₂S with the labile iron in those barrels.

Of the total accumulation of reduced sulfur (AVS and CRS) within the system, an average of 55% was accumulated in the chromium-reducible sulfur (CRS) fraction. Mossbauer spectroscopy failed to detect any pyrite (FeS₂) in the

compost, which suggests that elemental sulfur was the CRS form accumulated.

During system operation, the limestone content in the compost decreased by 35% for the entire system, and by 45% in the first barrel. This decrease confirmed the hypothesis that limestone dissolution generated much of the system's net effluent calcium and alkalinity:



Limestone dissolution was evident in the second and third reactor barrels, where pore-water pH was circumneutral, as well as in the first barrel, where pore-water pH was often lower. The mean rate of limestone dissolution for the system was 234 mg CaCO₃/L of flow.

Table 5. Chemical analysis of the Palmerton system compost

	Before operation	----After 17 weeks----	
		Reactor 1	Reactor 2
	-----μmoles/g dry wt-----		
Zn	26	308	545
Mn	6	38	25
Fe	98	109	87
AVS	<0.1	259	198
CRS	45	108	184

Values are means of 3 to 4 samples.

Table 5 shows the results of the analyses of Palmerton system compost. Note that, in this system, accumulated AVS accounts for only 40-80 % of the retained metals. This indicates that some of the metals were being removed by processes such as hydrolysis, in addition to being precipitated as insoluble sulfides. Chromium-reducible sulfur comprised 29 - 48 % of the total reduced sulfur (CRS + AVS) accumulated by the Palmerton reactors.

Sulfur Budget

The results of the water and compost analyses identified 3 sinks for the hydrogen sulfide produced by bacterial sulfate reduction: reaction with metal ions to form insoluble metal sulfides (AVS), conversion to CRS forms, and escape from the system in the effluent as unreacted sulfide. Figure 2 shows the mmol/L of

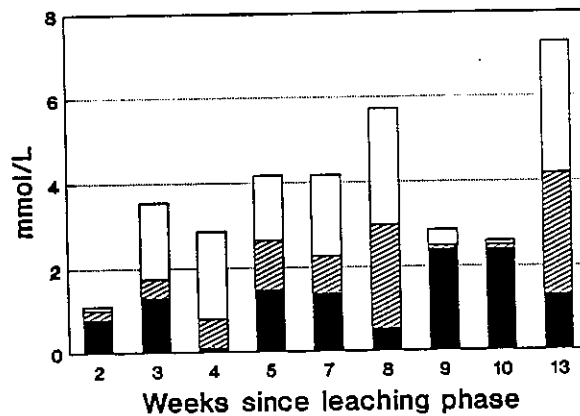


FIGURE 2

Figure 2. Sulfur budget for the Pittsburgh reactor system. Bar height represents total sulfate reduced, and is divided into portions accounted for by retained FeS (black), CRS (white), and effluent sulfide (shaded).

sulfate reduced (total bar height) over time for the Pittsburgh system, divided according to the portions accounted for by iron accumulation, effluent sulfide, and CRS (by difference). All

Table 6. Average rates of sulfate reduction, metal retention, and AVS/CRS accumulation for the reactor systems.

Process	Mean Rate (nmol/cm ³ compost/day)	
	Entire System	First barrel
---Pittsburgh system -----		
Sulfate reduction ¹	377	id
Total iron retention ²	101	170
Retention of influent Fe	31	100
Retention of labile Fe	70	70
AVS accumulation ²	107	167
CRS accumulation ²	156	id
---Palmerton system ----- Reactor 1 ----- Reactor 2 ---		
Sulfate reduction ¹	276	235
Zn, Mn, Ni, and Cd retention ¹	168	292

^{1,2} Calculated from changes in water chemistry (1) or substrate composition (2); id = insufficient data.

of the sulfate removed from the influent water was assumed to have been reduced to H₂S. Since the influent iron was completely retained by the system, the bar segment representing iron retention also represents influent iron concentration. Note that, for weeks 4 and 8, when influent iron concentrations were less than 0.5 mmol/L (28 mg/L), over 4 times as much sulfide escaped in the effluent as was retained as FeS. In contrast, during weeks 9 and 10, when influent iron concentrations were around 2.4 mmol/L (134 mg/L), nearly all of the sulfur reduced was retained as FeS, and very little effluent sulfide escaped.

Table 6 shows the mean rates of sulfate reduction, metal retention, and AVS/CRS accumulation computed for the 2 reactor systems. The iron retention and AVS/CRS accumulation rates for the Pittsburgh system were computed using compost analysis data:

$$\frac{\text{nmol change in } X}{\text{g dry compost}} \times \frac{\text{g dry compost}}{\text{cm}^3 \text{ moist compost}} \times \frac{1}{\text{days}} = \text{nmol/cm}^3/\text{day}$$

Compost analysis data were not used to compute rates for the Palmerton system because the compost sampling method was insufficiently representative.

The other rates were computed from water analysis data:

$$\frac{\text{nmol/L change in X}}{\text{cm}^3 \text{ compost in system}} \times$$

$$\frac{\text{L of flow}}{\text{day}} = \text{nmol/cm}^3/\text{day}$$

The rates of AVS accumulation almost equal those of total Fe retention for the Pittsburgh system, as would be expected if FeS was being formed. Note that the sulfate reduction rate for the Pittsburgh system exceeds the sum of the AVS and CRS accumulation rates by 114 nmol/cm³/day. This difference suggests an average effluent sulfide concentration of 37 mg/L, which is close to the measured average of 34 mg/L. By neglecting CRS formation, and similarly computing effluent sulfide from the Palmerton system rates, values of 104 and 0 mg/L sulfide are obtained for reactors 1 and 2, respectively. These concentrations are low in comparison with the measured averages of 149 (reactor 1) and 69 (reactor 2) mg/L. This indicates that more sulfide escaped the Palmerton system unreacted than the metal retention rates suggest, and further indicates that some of the metal removal observed in the Palmerton system occurred through processes other than sulfide precipitation.

Origins of Effluent Alkalinity

Both reactor systems produced effluents having high alkalinity. Equations 1, 2, and 4 - 7, describing the principal reactions consuming or producing alkalinity in the reactors, were used to derive the following equation for

predicting effluent alkalinity (meq/L) from the absolute values of measured changes in the concentrations (mmol/L) of SO₄⁻², Ca⁺², FeOOH, M⁺², Al⁺³, and Fe⁺³:

$$\begin{aligned} \text{Alkalinity} &= 2\text{SO}_4^{-2} + 2\text{Ca}^{+2} \\ &+ 2\text{FeOOH} - 2\text{M}^{+2} - 3\text{Al}^{+3} - 1\text{Fe}^{+3} \\ &+ \text{influent alkalinity} \\ &- \text{influent H}^+ \end{aligned} \quad (8).$$

The derivation of equation 8 assumes that equations 1, 2, and 4 - 7 are the sole sources of alkalinity and acidity, and that the concentration changes observed for the indicator ions are induced solely by the reactions they indicate.

Figure 3 shows the alkalinity values computed using equation 8, along with the corresponding measured alkalinity values for the reactor system effluents. Correlation of the 2 sets of values gives an r² of 0.92 (P < .01; n = 8) for the Pittsburgh system, and 0.87 (P < .01; n = 18) for the Palmerton system. This agreement between the 2 sets of values supports the hypothesized origins of effluent alkalinity.

Sulfate reduction (equation 1) and limestone dissolution (equation 7) were the dominant sources of alkalinity for both systems. Of the total computed alkalinity generated by these 2 reactions combined, limestone dissolution contributed an average of 53% for the Pittsburgh system, 58% for Palmerton reactor 1, and 71% for Palmerton reactor 2.

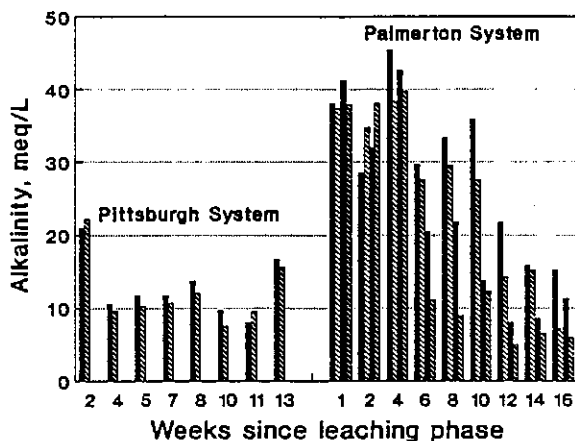


FIGURE 3

Figure 3. Alkalinity of effluents from the 2 reactor systems. Shaded bars depict measured values and solid bars depict computed values.

Discussion

The results presented in this paper confirm that contaminant metals can be precipitated as insoluble sulfides by reaction with biologically-generated hydrogen sulfide. Several strategies for enhancing the metal-retaining and alkalinity-generating effects are suggested by the results.

The maximum quantity of metal that can be precipitated in a sulfide form is dictated by the quantity of H_2S available for reaction. One means of enhancing metal retention, therefore, might be to increase the rate of bacterial sulfate reduction. McIntire et al (1990) found that when lactate, a preferred carbon source for sulfate-reducing bacteria, was added to the metal-contaminated water entering their reactors, sulfate reduction and metal retention increased. This suggests that the type or

quantity of organic compounds released by the decomposing spent mushroom compost limits the rate of sulfate reduction. Thus, the rate might be increased by filling the reactors with a different organic material, or by supplementing the compost by adding organic compounds to the influent water. Since sulfate reduction is a microbial process, its rate might also be increased by raising reactor temperature.

Another means of enhancing reactor metal retention would be to modify reactor design or operation so that all of the H_2S produced reacts completely to form metal sulfides and none is lost as effluent sulfide or CRS. For the Pittsburgh system, an inverse relationship was found between influent iron and effluent sulfide concentrations. Similarly, pore-water sulfide concentrations were low where dissolved iron was still present, and high where dissolved iron was absent. These observations demonstrate that excess sulfide, destined for release in the effluent, can be exploited for metals removal, provided the system is fed metal ions at a sufficiently high rate. For metals that are relatively toxic to bacteria, such as cadmium and copper (Postgate 1984; McIntire et al., 1990), the influent metal load might best be adjusted by modifying flow rate, whereas, for other metals, influent concentration could also be modified. No means can be suggested for exploiting the H_2S converted to CRS forms, since the pathway of this conversion is poorly understood.

If acidic water is to be treated, the reactors must generate net alkalinity to prevent the bacteria from being inhibited and metal sulfides from being dissolved. Limestone dissolution in the reactors generated slightly more alkalinity than did sulfate reduction. When the compost limestone content eventually becomes depleted, or in a reactor built without limestone, sulfate reduction would have to be relied upon as the principal alkalinity generator. Since metal sulfide formation (equation 2) consumes as many moles of alkalinity as are generated by sulfate reduction (equation 1), no net alkalinity will be gained from sulfate reduction if all of the H_2S produced is consumed for metal retention. Thus, sulfate reduction serves as an important alkalinity source only if some H_2S remains unreacted. In the Pittsburgh system, this tradeoff between alkalinity generation and H_2S consumption was observed directly, as most of the net alkalinity and excess H_2S accumulated in the second and third reactor barrels in the absence of iron removal.

This study demonstrates that sulfate reduction reactors of the simple design investigated here are capable of removing Fe, Zn, Mn, Ni, and Cd from contaminated water. Without modifying reactor design, metal retention rates can be increased by providing influent metal loads sufficiently large to completely consume the H_2S generated. The theoretical maximum metal retention rate will equal the sulfate reduction rate. If acidic

water is being treated, the reactor interior must be protected from acidification, either by incorporating limestone into the organic substrate, or by operating the reactors at a lower metal retention rate such that H_2S is produced in excess of the amount that will be consumed by metal sulfide formation. Higher rates of metal retention could also be attained by modifying reactor design to encourage higher rates of bacterial sulfate reduction.

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