TREATMENT OF ACID MINE DRAINAGE IN SULFATE REDUCING BIOREACTORS: EFFECT OF HYDRAULIC RESIDENCE TIME AND METALS LOADING RATES.

by

Robin E. Madel

ProQuest LLC.
789 East Eisenhower Parkway
P.O. Box 1346
Ann Arbor, MI 48106 – 1346

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Golden, Colorado
Date 8/25/92

Signed: Robin E. Madel
Robin E. Madel

Approved:
Dr. Ronald R. H. Cohen
Thesis Advisor

Golden, Colorado
Date 8/25/92

Dr. John Cordes
Department Head,
Environmental Science and Engineering.

ABSTRACT

This study investigated the ability of sulfate reducing bacteria in bench scale bioreactors to treat acid mine drainage at lower residence times than those previously investigated. The bioreactors were filled with a manure and hay mixture that provided sulfate reducing bacteria and a carbon source for the bacteria. Residence times of 10, 20, 50 and 100 hours were investigated.

The effectiveness of bioreactors in series was compared to bioreactors in parallel to determine if a multiple-stage system would operate as well as a single-stage system at lower residence times. The bioreactors were designed to have equivalent residence times but different average velocities, and were operated in an upflow configuration. Treatment efficiency was defined in terms of an increase in pH to 6.5-9.0 and reduction of the following metals to below Eagle Mine treatment plant discharge permit limits: cadmium, copper, iron, lead, manganese and zinc.

Statistical analyses indicate that there is no significant difference in performance of the multiple stage series system compared to the single stage system. Results indicate that at a residence time of 100 hours, both systems remove Cd, Cu, Pb, Fe and Zn to below permit limits. Removal of Mn is above permit limits. An average pH of 7.16 is observed. A residence time of 100 hours is comparable to residence times observed in previous studies.

At residence times below 100 hours, Cd, Cu, and Pb are consistently removed below permit limits. Fe, Mn and Zn are removed consistently above permit limits. Removal of Fe and Zn decreases with decreasing residence times. Effluent Mn concentrations are approximately equal to influent concentrations. Average effluent pH values are below the lower permit limit of 6.5. These results have implications for design of systems for treatment of acid mine drainage.

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Finally, I would like to thank my friends, especially Alison Deans and Judy Bolis, who provided emotional, physical and professional assistance during some rather trying times. You helped me do what I needed to.

DEDICATION

This thesis is dedicated to my mate, Robert L. Tucker, Jr.
Now it's your turn.

CHAPTER 1

INTRODUCTION

The Eagle Mine

Mining runoff adversely affects over 15,000 miles of streams and rivers in the United States (Cohen and Gorman 1991). One example of this problem is the Eagle River in Minturn, Colorado. As a result of almost a century of mining operations at the Eagle Mine, concentrations of dissolved and precipitated metals in the Eagle River have been considerably elevated over natural values.

Typical water quality parameters for Eagle Mine water are presented in Table 1. While present concentrations do not represent a significant human health risk (Williams 1991), water in the river has been discolored and aquatic life has been reduced. The Eagle River is listed as a Recreation Class II Water Supply. Dames and Moore Inc., in cooperation with Paramount Resources (determined to be the Principal Responsible Party), has established a cleanup program designed to remediate Eagle Mine water quality problems.

Standards affecting Eagle Mine drainage are found in two forms; permit discharge limits for the current treatment facility and water quality standards for the Eagle River. Both sets of standards are presented in Table 1.

One form of remediation in use at the Eagle Mine is a Passive Mine Drainage Treatment System (PMDTS). The system treats acid mine drainage predominately through bacterial sulfate reduction, which results in sulfide production. Under anaerobic conditions, metals will precipitate out of solution as metal sulfides. Other processes include adsorption and complexation of metals. Previous investigations at the Eagle Mine have demonstrated that, through the use of a PMDTS, the pH of the water can be increased and metals can be effectively removed from the mine drainage (Staub 1992).

Staub (1992) studied treatment efficiency of acid mine drainage within the Eagle Mine using one sulfate reducing bioreactor in parallel with two bioreactors arranged in series. No significant difference in treatment efficiency was found between the series system compared to the parallel system.

Table 1. Eagle Mine water quality parameters (mean and one std. dev. shown) and the standards affecting Eagle Mine drainage.

Parameter	Typical Values (mg/L)	Permit Discharge Limit (mg/L)	Stream Segment Standard (mg/L)
D.O.	N/A	N/A	6.0
pH	2.92 ±0.12	6.5-9.0	6.5-9.0
F. Coli	N/A	2000/100mL	N/A
S	N/A	0.002	N/A
NO_2	N/A	0.05	N/A
NO_3	N/A	10.0	N/A
SO_4	4314 ±432	250.0	N/A
Cd (total)	0.83 ±0.14 (dis)	0.001	0.008
Cu (total)	13.1 ±1.7 (dis)	0.014	0.15
Fe (total)	N/A	1.0	9.8
Fe (dis)	279 ±38	0.3	N/A
Pb (total)	0.63 ±0.15 (dis)	0.009	0.12
Mn (total)	N/A	1.0	31.0
Mn (dis)	190 ±20	0.05	1.2
Zn (total)	N/A	N/A	0.4
Zn (dis)	238 ±48	0.4	N/A
Eh (mV)	652.9 ±84.1	N/A	N/A
Cond (mmhos)	3837 ±945	N/A	N/A

Permit Limits from Williams (1991).
Stream Standards from Neukirchner, personal communication.
N/A = data not available.

Hypothesis and Research Objectives

The main objective of this study is to design an effective treatment of acid mine drainage using bioreactors containing sulfate reducing bacteria. Acceptable treatment is defined as an increase in pH to near neutral (6.5-9.0) and reduced metals concentrations to within discharge permit limits. It is hypothesized that, for a given short hydraulic residence time, the loading rate of metals will exceed the rate of sulfate reduction, and therefore the rate of sulfide production. Treatment efficiency will be reduced and concentration of metals in the effluent will exceed acceptable levels.

Several research objectives are investigated. The first objective is to estimate from the literature at what hydraulic residence time the metal loading rate will exceed the rate of sulfide production. The second objective is to apply this estimated residence time to a bench scale system consisting of one bioreactor in parallel with two bioreactors in series. This system is described in detail in the methods section. The third objective is to compare performance of the series system to the parallel system. The fourth objective is to apply results from the bench scale system to a pilot scale system located within the Eagle Mine. The fifth objective is to ascertain whether bench scale results are comparable to pilot scale results.

Acid Mine Drainage Formation

Acid mine drainage results from the oxidation of pyrite, FeS_2, yielding hydrogen ions, sulfate and metals. Microorganisms accelerate the process, which consists of the following steps (Singer and Stumm 1970). Step 1 is the oxidation of pyrite:

$$2FeS_2(s) + 2H_2O + 7O_2 \rightarrow 4H^+ + 4SO_4^{2-} + 2Fe^{2+} \qquad (1)$$

with concomitant production of sulfuric acid. Step 2 is the auto-oxidation of ferrous iron to ferric iron:

$$4Fe^{2+} + O_2 + 4H^+ \rightarrow 4Fe^{3+} + 2H_2O \qquad (2)$$

Step 2 occurs slowly at low pH values. Below pH 3.5, the iron oxidation is catalyzed by the iron bacterium *Thiobacillus ferrooxidans*, and in the pH range of 3.4 to 4.5 it may be catalyzed by a variety of *Metallogenium*. Other bacteria that may be involved in acid mine drainage formation are *Thiobacillus thiooxidans*.

The ferric ion further reacts with pyrite in the next step:

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

which, in conjunction with equation (2), constitutes a cycle for the dissolution of pyrite. At $pH < 3$, iron(III) precipitates as the hydrated iron(III) oxide:

$$Fe^{3+} + 3H_2O \longleftrightarrow Fe(OH)_3(s) + 3H^+ \qquad (4)$$

The beds of streams affected by acid mine drainage are often covered with "yellow boy," an unsightly deposit of amorphous, semigelatinous $Fe(OH)_3$. The most damaging and toxic component of acid mine drainage, however, is sulfuric acid (Manahan, 1990), which can cause significant ecological imbalances.

Surface water quality and aquatic life resources in and along the Eagle River have been adversely affected by heavy metal pollution from the Eagle Mine facility (Engineering Science 1985). Until recent efforts to remediate mine drainage problems along the Eagle River, the river had been subject to airborne contaminants, acid mine drainage seeps, and leaching from tailings piles (Engineering Science 1985).

Site Description and History

The Eagle Mine facility is an inactive mining and milling facility located adjacent to the Eagle River near Gilman, Colorado (Figure 1). The mine is located between the towns of Minturn and Redcliff, Colorado approximately eight miles southwest of Vail and 100 miles west of Denver (Engineering Science 1985).

Miners were first attracted to the area in the late

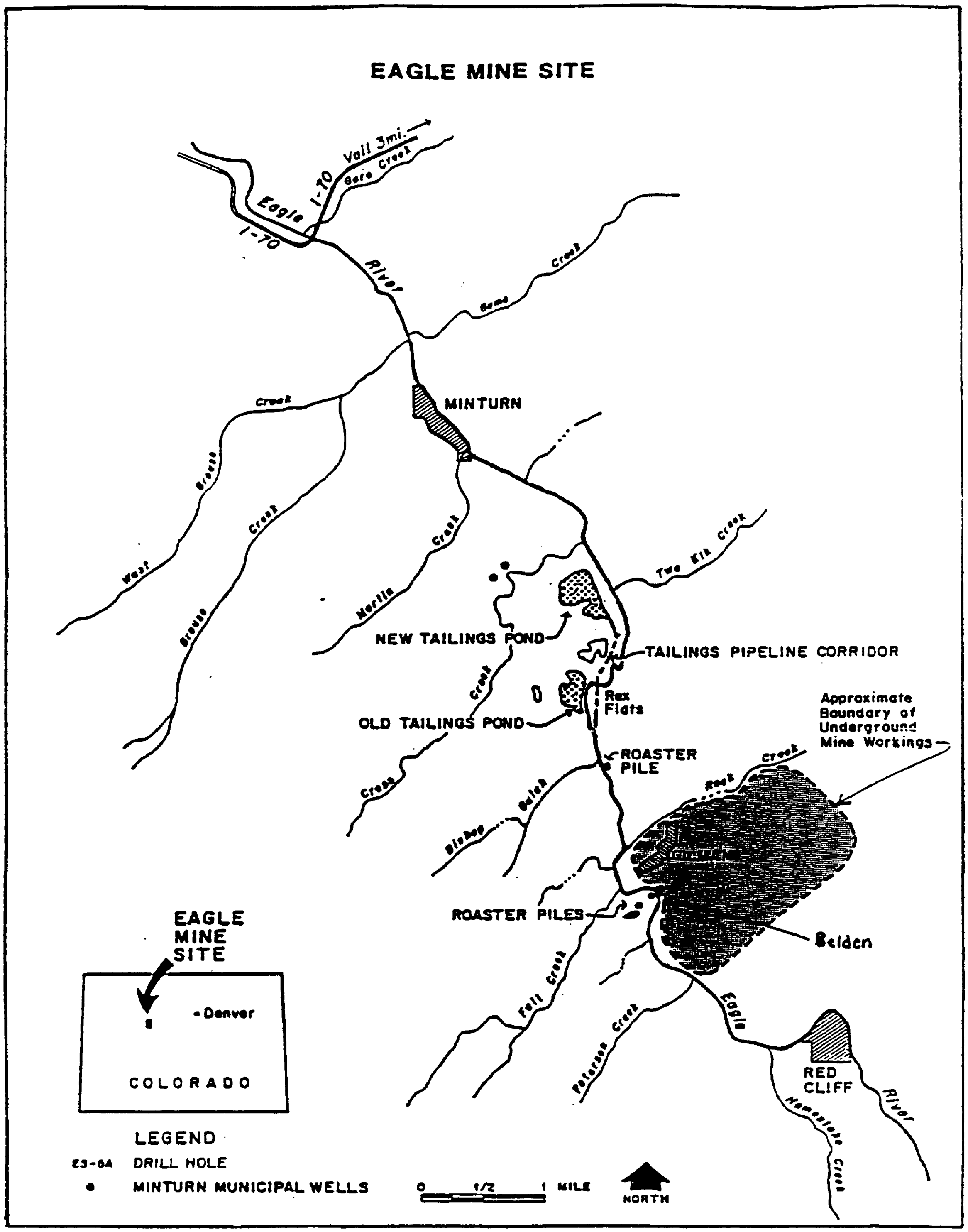

Figure 1. Map showing the Eagle River and the Eagle Mine facility (from Engineering Science 1985). Arrow shows approximate location of field system.

1800's when a series of small independent mines were established to search for gold, silver, lead and zinc. In 1912, the Empire Zinc Company (a subsidiary of New Jersey Zinc) began acquiring and consolidating many of these small mines, including the Eagle Mine.

In 1928, an underground froth flotation mill was constructed, a unique feature of this mine. The waste material from the mill was transported as a slurry through a buried pipeline from the millsite to a point approximately 4 miles away. In 1966, the New Jersey Zinc Company merged into Gulf and Western Industries, Inc. The milling area was converted into an acid mine drainage treatment facility in 1977. In 1983, the facility was sold to an individual who ran out of financing and abandoned the facility a year later (Engineering Science 1985).

To keep the mine from flooding, water had been pumped to a level approximately 300 feet above the mine workings. When the mine shut down in 1984, the electricity was turned off and the mine began to flood. Concrete bulkheads were constructed in the adits to hold back the water, which, nevertheless, eventually found its way out through fractured and faulted headrock. The water began draining into the Eagle River, at which point it was collected and piped, via the old mill slurry line, to a treatment plant approximately three miles away. Between 1988 and 1990, Paramount Resources acquired Gulf and Western. Paramount Resources was assigned responsibility for cleanup of the mine.

The current treatment process involves adding slaked lime and soda ash to the water. Alkalinity and pH are increased allowing metals to precipitate out of solution as metal hydroxides. Up to 300 gal/min are treated, producing approximately 50,000 lbs/day of metal hydroxide sludge (Thompson 1992) which is difficult to de-water. This is an expensive, multi-unit, energy intensive process, requiring trained operators. Obviously, an alternative, less demanding treatment process is preferable.

CHAPTER 2

ACID MINE DRAINAGE REMEDIATION

Constructed Wetlands

It has been known for over five decades that natural wetlands will partially treat acid mine drainage (Dollhopf et al. 1990), however, it is undesirable to contaminate natural wetlands. The focus of recent research, therefore, has been the construction of wetland systems that simulate natural wetlands. These systems are designed to optimize specific processes relevant to the removal or modification of certain contaminants from water. Low cost immobilization of metals for long periods of time is the goal of using constructed wetlands for acid mine drainage treatment (Wildeman 1990).

Figure 2 shows a model of a typical wetland. Klusman and Machemer (1991) list the following removal processes operating in a wetland:

1) Exchange of metals by an organic rich substrate.
2) Sulfate reduction with subsequent precipitation of metal sulfides.
3) Precipitation of ferric and manganese hydroxides.
4) Adsorption of metals by ferric hydroxides.

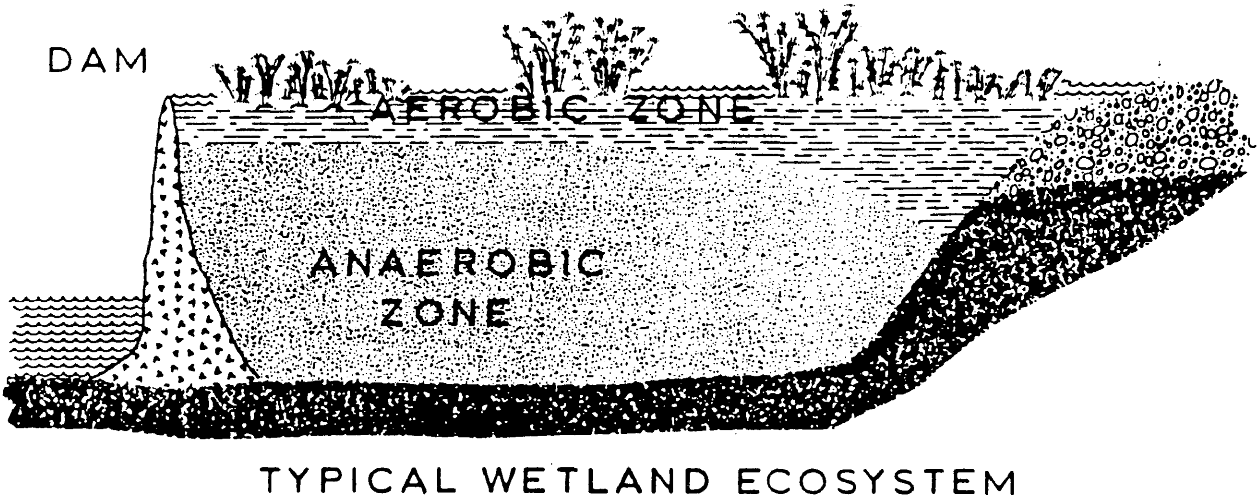

Figure 2. Model of a typical wetland (from Wildeman 1990).

5) Metal uptake by living plants.
6) Filtration of suspended and colloidal material.

Uptake of Metals by Plants

Some controversy exists as to whether plants actually accumulate metals or whether they assist in metal removal. Several studies suggested that plant uptake was an important process in metal removal (Brodie et al. 1988; Donlan 1989; Guntenspergen et al. 1989). Some studies suggested that uptake by plants may account for as little as 1-5% of metal removal (Sencidiver and Bhumbla 1988; Emerick et al. 1988).

Adsorption and Complexation of Metals

Complexation of metals onto organic substrates was studied at the Big Five constructed wetlands sight in Idaho Springs, Colorado (Machemer and Wildeman 1991). That study confirmed that there is competition for organic complexation sites between Fe, Cu, Zn and Mn according to the following order:

$$Fe = Cu >> Zn = Mn$$

They found that, during an initial period of approximately one month of mine drainage flow to the substrate, organic complexation was an important metal removal process. As sulfate reduction increased and complexation sites filled, however, the importance of organic complexation as a form of metal removal decreased.

Sulfate Reducing Bacteria

Some constructed wetland research has suggested that sulfate reduction by bacteria is the principal process for removal of metals from acid mine drainage (Dvorak et al. 1991; Hedin et al. 1988; Herlihy and Mills 1985; Herlihy et al. 1987; Machemer and Wildeman 1991; Reynolds 1991; Tuttle et al. 1969; Wieder 1988; Wildeman and Laudon 1989). Therefore, a system which optimizes conditions under which sulfate reducing bacteria function is desirable for treatment of waters high in metals and sulfate such as acid mine drainage.

Many natural anaerobic environments contain heterotrophic micro-organisms that use organic compounds for growth and as an energy source for reducing sulfate to sulfide (Laanbroek 1984; Oremland and Polcin 1982; Smith and Klug 1981; Sorenson et al. 1981). When heavy metals are present, they will precipitate as insoluble sulfides. Overall bacterial sulfate reduction and precipitation of metal sulfides can be represented by the following equation:

$$\text{Metal + sulfate + Carbon substrate} \longrightarrow$$
$$\text{MeS} + CO_2 + H_2O + \text{cell biomass + acetate} \quad (5)$$

These organisms are involved in the formation of certain sedimentary metal sulfide deposits such as the iron sulfide found in coal (Trudinger and Swaine 1979; Zajic 1969; Howarth 1979).

Sulfate reducing bacteria, or SRB, are obligate anaerobes that decompose simple organic compounds using sulfate as the terminal electron acceptor (Macaskie and Dean 1989; Grady, 1980). The result is the production of sulfide that may be given off as H_2S gas or that may react with metals to form metal sulfides.

SRB are part of a consortium of interdependent organisms, utilizing a limited variety of organic substrates. Table 2 gives some examples of sulfate reducing organisms together with their preferred substrates and products (Barnes et al. 1991a). Desulfovibrio and desulfotomaculum are the primary members of the sulfate reducers (Postgate 1979).

Temperature effects on the growth of SRB can be described by an Arrhenius type relationship for kinetic coefficients. Growth kinetics for some types of sulfate reducing organisms are described by Middleton and Lawrence

(1977). A Monod type kinetic model can be used to describe the growth of SRB. Also, SRB growth is independent of sulfate concentration beyond a certain concentration (50 mg/L at 20°C, low compared to typical concentrations found in acid mine drainage).

Table 2. Some examples of sulfate reducing bacteria, their preferred growth substrates and end products.

Organisms	Substrate	Product
Desulfovibrio vulgaris	La	Ac
Desulfomonas pigra	yeast	Ac + H_2
Desulfobulbus propionious	La, Pr, Et	Ac
Desulfococcus multivorans	La, Ac, Et, Me	CO_2
Desulfobacter postgatei	Ac, Et	CO_2
Desulfosarcina variabilis	La, Ac, Et, Me	CO_2
Desulfonema magnum	Be	CO_2
Desulfotomaculum orientis	H_2 + CO_2	(-)

La = Lactate Ac = Acetate Pr = Propionate
Be = Benzoate Me = Methanol Et = Ethanol

In order to sustain a maximum growth rate of SRB, two important environmental conditions (among others) must be satisfied (Brown 1973). First, a near neutral pH must be maintained. This occurs through precipitation and dissolution of carbonates and sulfides, which are products of organism growth. Second, a low redox potential must be maintained. This is accomplished by the presence of sulfide ions in solution and the absence of oxygen due to rapid uptake of available oxygen within the first few centimeters of solid phase substrate.

Metal Sulfide Stability

An Eh-pH diagram is shown in Figure 3 (Stumm and Morgan 1981). The sulfate ion exists over a wide range of pH values. For formation of metal sulfides to take place, Eh and pH values must be maintained where sulfide species are stable. As seen in Figure 3, this occurs under neutral, reducing conditions. These conditions can be accomplished in an anaerobic bioreactor.

Table 3 lists solubility products of metal sulfides of interest in the Eagle Mine drainage (Whitten and Gailey, 1981). Table 3 shows that Cd-, Cu-, Pb- and Zn-sulfides are less soluble than Fe- and Mn-sulfides. Therefore sulfides of Cd, Cu, Pb and Zn should precipitate out of solution more readily than sulfides of Fe and Mn. This is consistent with results of previous studies (Bolis 1991; Dvorak et al. 1991; Hurley Euler 1992; Staub 1992).

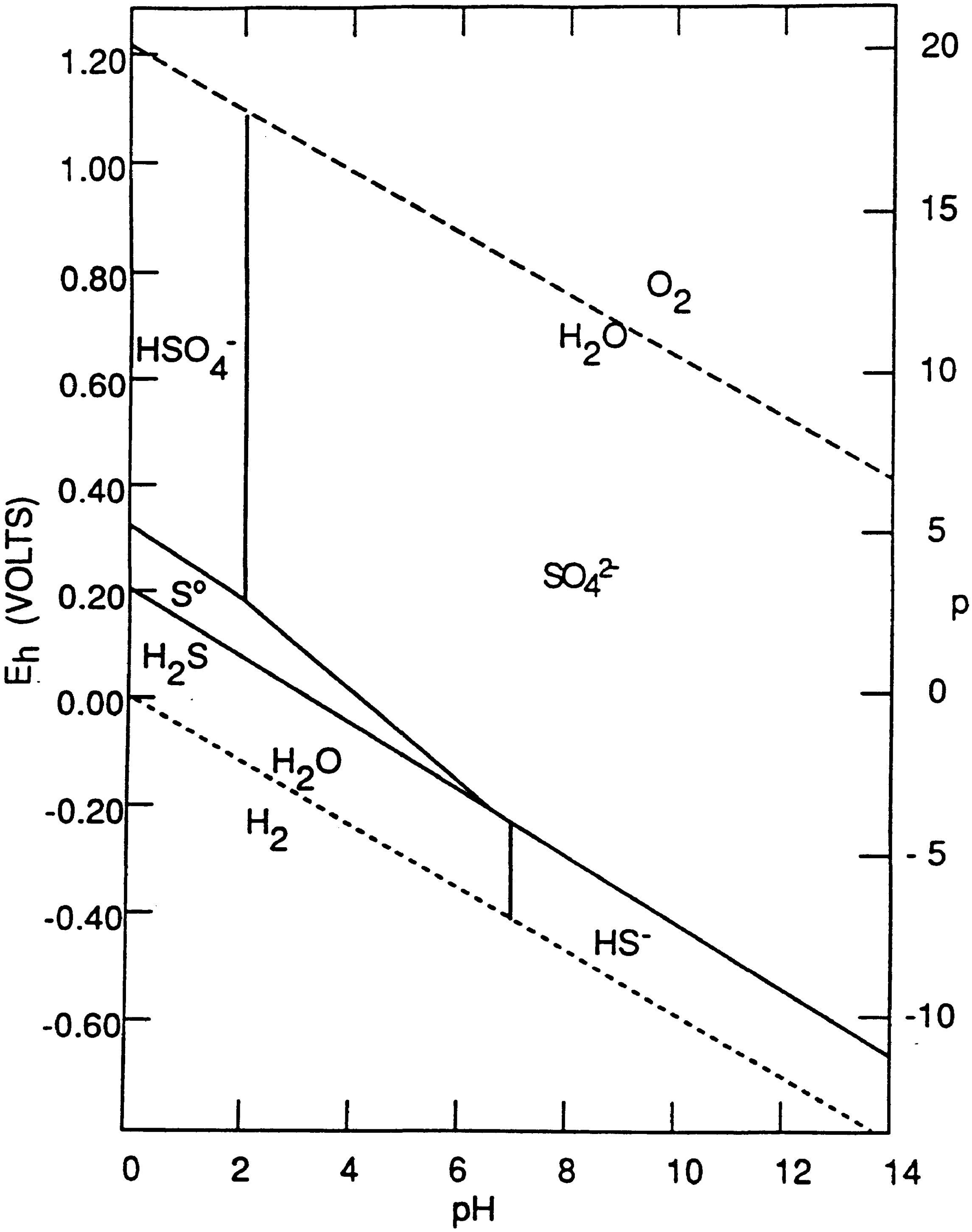

Figure 3. An Eh-pH diagram for sulfur species (from Stumm and Morgan 1981).

Table 3. Solubility product constants of metal sulfides at 25°C.

Metal Sulfide	K_{sp}
CdS	3.6×10^{-29}
CuS	8.7×10^{-36}
Fes	4.9×10^{-18}
PbS	8.4×10^{-28}
MnS	5.1×10^{-15}
ZnS	1.1×10^{-21}

Bioreactors: Design Considerations

There has been considerable research performed on sulfate reducing bacteria and their ability to remove metals and sulfate from wastewater. Several bioreactor designs and configurations based on optimizing conditions for SRB are discussed here.

Lema et al. (1991) made recommendations for engineering concepts in design and operation of anaerobic reactors. They suggested that since the energy available for bacterial growth is small, some kind of a bacterial detention device is necessary to build up the biomass. They also concluded that reaction rates are much slower than for many other biological reactions. These long reaction times should be accounted for in the design of a system. Further recommendations are to consider the stoichiometry and kinetics of the processes involved in anaerobic treatment. For example, rate constants are low compared to other biological processes and especially chemical processes, emphasizing the need for cell detention. The following articles addressed some of the issues presented by Lema et al.

Nakamura (1988) used a 180 L continuous-flow, fluidized bed anaerobic reactor. Laboratory and pilot scale systems were used to determine optimum conditions to maintain the biological and chemical reactions involved in sulfate reduction. Different kinds and amounts of growth media such as yeast and sodium lactate were examined. Temperature effects, pH, and the flow rate of raw water on rates of bacterial sulfate reduction were studied. Mine drainage with a chemical composition of Fe = 40 mg/L, Zn = 40 and pH = 4.7 was successfully treated at 12°C, sodium lactate level at 1.5 g/L and mg/L and a retention time of 6 hours. Nakamura's system produced acceptable results for metal removal at low residence times but he found his effluent COD values were high (800-1000 mg/L).

Barnes et al. (1991a) studied several bioreactor designs for treatment of extracted groundwater contaminated by zinc leachate from a processing plant. Three designs were considered for a pilot plant system.

The first design was a 1.6 L stirred (at 200 rpm) tank reactor. This reactor design offered consistently reliable and rapid equilibrium conditions, however, the high shearing forces of the stirring apparatus dislodged the microorganisms from the sludge. This design was, therefore, considered unacceptable.

The next configuration tested was a 1.8 L fixed bed reactor at controlled temperatures. This design was considered preferable to the stirred tank reactor due to the absence of high shear forces. This allowed for a higher biomass concentration, however some channelling of the sludge was observed. Perfect steady state operation was never achieved in this reactor, although some valuable information on residence time and sludge bed buffer capacity was obtained. A residence time of only four hours was achieved for complete precipitation of heavy metals.

The final design considered was a raked sludge blanket reactor. The sludge blanket consisted of metal sulfides to which the microorganisms adhered. The sludge blanket was mechanically raked or suspended by liquid recycle to prevent compaction or channelling of the sludge. This design was chosen for the pilot scale system.

A pilot scale sludge blanket reactor of 9 m^3 was then constructed. Influent values used in this study were SO_4 = 1580 mg/L, Cd = 1.8 mg/L, Cu = 1.4 mg/L, Fe = 3.5 mg/L, Mn = 5.8 mg/L and Zn = 115 mg/L. This reactor achieved high metal removal with low residence times (< seven hours) but the addition of a flocculant was necessary to settle the metal sulfides into the sludge and to prevent substantial organism washout.

Maree and Strydom (1985) examined sulfate removal in an upflow, packed bed reactor of 1 L total liquid volume, to treat mine drainage and industrial effluent. They examined several types of packing media. Influent water had the following chemical characteristics: pH = 5.0, Cd = 0.01 mg/L, Fe = 2.68 mg/L, Pb = 0.135 mg/L, Mn = 0.340 mg/L and Zn = 0.32 mg/L. In the pilot scale version of this system a residence time of 12 hours was achieved (Maree et al. 1991).

The bioreactor designs discussed above were able to achieve low residence times but were not passive in their design. They were multiple unit, energy intensive systems that required a high degree of operator skill. A passive, less demanding approach to a sulfate reducing anaerobic bioreactor has been studied extensively by students and faculty at the Colorado School of Mines and by researchers at the U.S. Bureau of Mines, Pittsburgh Research Center.

This system is based on the concepts of a wetland (see discussion above). An advantage of this system in pilot scale is that it is passive, therefore it can be located in remote areas without electricity, an important consideration at many abandoned mine sites.

An organic medium serves as the substrate, and therefore, a source of organic carbon and as the source of sulfate reducing bacteria. The substrate acts as a matrix upon which a biomass can accumulate. Typically, mushroom compost or composted livestock manure has been used. Wood chips, sawdust and sediments have also been examined. Several configurations and substrate amendments have been tested (Bolis et al. 1991; Bolis et al. 1992; Dvorak et al. 1991; Reynolds et al. 1991; Staub 1992).

Mushroom compost or composted livestock manure with an amendment of hay or hay extract seemed to produce the highest rates of sulfate reduction and metal removal (Bolis 1992; Rabenhorst et al. 1992; Reynolds et al. 1991). The manure provides a source of carbon that does not need replenishing for several years. An experimental, constructed wetland in Idaho Springs, Colorado has been operating successfully for approximately five years with the original substrate (Wildeman 1990). Initially, addition of hay provides an easily degradable carbon source to the bacteria. Reynolds et al. (1991) reported significantly increased rates of sulfide production with mushroom compost amended with hay extract. Several flow configurations were tested and an upflow system was determined to provide the most consistent hydraulic conductivity (Bolis 1992; Lemke 1989).

Metal Loading Rates

Loading rates have been recommended for systems treating acid mine drainage through bacterial sulfate reduction. Early work suggested 1 gpm per 200 ft^2 (Sims et al. 1963). Wildeman (1990) suggested a more conservative loading rate of 1/8 gpm per 100 ft^2 for a variety of substrates. These loading rates were based on areal determinations for a system of fixed depth. Higher loading rates based on volume were determined by Staub (1992) and Hurley Euler (1992) for a substrate of manure and hay. Staub reported 200 mL/min/ 1.27 m^3 substrate for effective treatment. Hurley Euler reported effective treatment at 5 mL/min/0.024 m^3 substrate.

The reactions of sulfate reduction and carbonate formation are:

$$2CH_2O + SO_4^{2-} \text{--->} S^{2-} + 2CO_2 + 2H_2O \quad (6)$$

$$S^{2-} + 2CO_2 + 2H_2O \text{--->} H_2S + 2HCO_3^- \quad (7)$$

Where acid mine drainage is the primary source of sulfate, a variety of metals are often present. The metals will complex with the S^{2-} and, under proper conditions, will form insoluble metal sulfides that precipitate from the water primarily as metal monosulfides (Reynolds 1991; Mills 1985).

It is possible to estimate a loading rate for the amount of metals that can effectively be removed as MeS using rates of sulfate reduction. As is seen in equation (6) there is a mole to mole relationship between the amount of sulfate reduced and the amount of sulfide produced. Several studies have reported rates of sulfide production (Table 4). Values range from 2 to 3440 nmol S^{2-}/cm^3 substrate/day with a mean value of 880 nmol/cm^3/day $\pm$ 1042. Using these values for a given volume of substrate, and assuming that for each mole of S^{2-} produced one mole of metal will be removed, a loading rate can be calculated for a drainage of a given composition. This information is calculated for the Eagle Mine drainage used in Chapter 3.

Table 4. Sulfide production rates reported in the literature.

Author	Substrate	SR Rate (nmol/ cm^3/d)
Herlihy and Mills 1985	Lake Sediments	800
McIntire and Edenborn 1989	Mushroom Compost	2 to 600
Reynolds et al. 1991	Mushroom Compost with Sodium Lactate Hay Extract No Amendment	 1920* 3440* 728*
Dvorak 1991	Mushroom Compost Double Stage Single Stage	 405 377
Hammack and Edenborn 1991	Mushroom Compost w/Lactate	92 40
Eger 1992	Municipal Compost	400
	mean	880±1042

* Assuming a bulk density of 0.8 g/mL from Bolis.

u = 880 ± 1042 nmol/d/cm^3

CHAPTER 3

METHODS

Laboratory

The laboratory system consisted of two parallel systems, one two stage series system and one single stage system (Figure 4). The laboratory system was located in the Environmental Science and Engineering laboratory of the Colorado School of Mines. The room was not insulated and had east-facing windows. The temperature varied from approximately 20°C to 25°C. Mine drainage was transported from the Eagle Mine water treatment plant to the laboratory and stored at room temperature for up to three weeks.

The bioreactors were manufactured by AIA Plastics, Denver, Colorado. The bioreactors were constructed from 1/2 inch wall thickness, clear plexiglass with an inner diameter of 5.5 inches and a total height of 24 inches (Figure 5). Each bioreactor was filled to an equal volume with manure and hay in a 3:1 by volume mixture, to a height of 18 inches. The total volume of substrate per bioreactor was 7008 mL or 0.007 m^3. A sheet of wire mesh (with approximately 1/2 inch grid openings) and furnace filter fabric was placed below and on top of the manure/hay

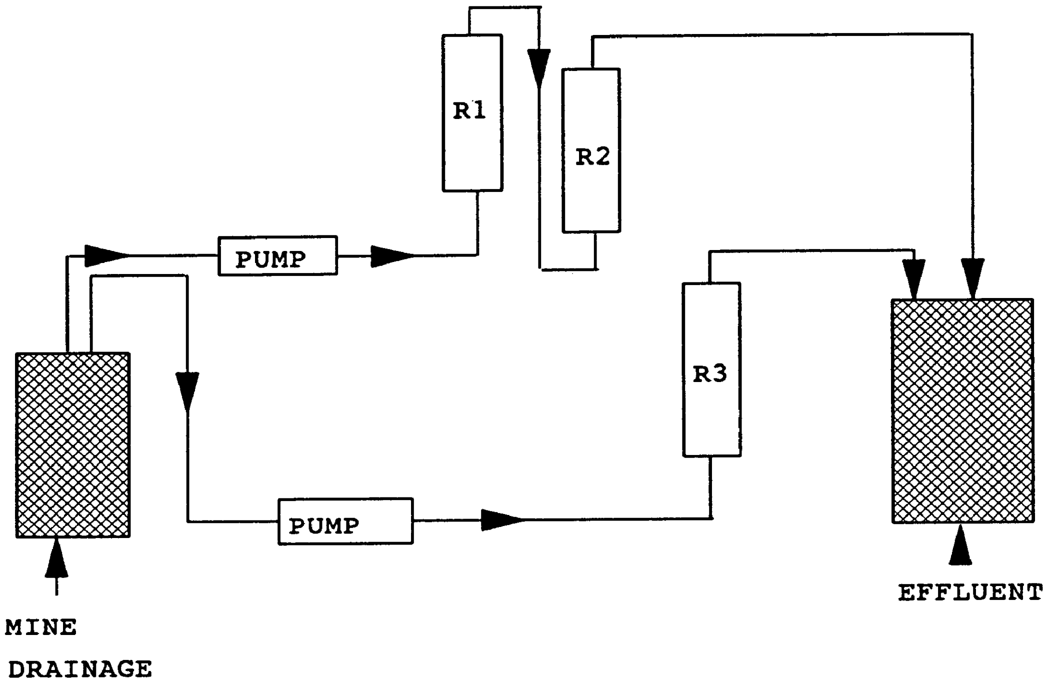

Figure 4. Plan view diagram of layout of bioreactors used in the laboratory.

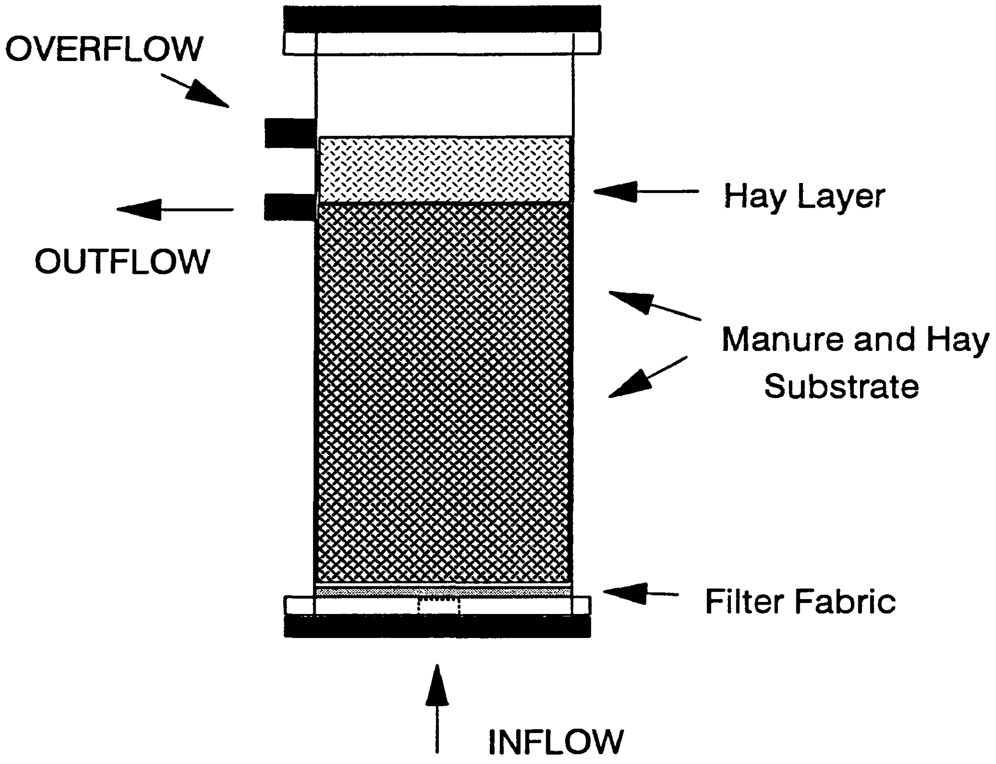

Figure 5. Schematic of 7008 mL plexiglass bioreactor used in the laboratory.

mixture. These sheets dispersed the mine drainage and prevented loss of manure and hay.

Each bioreactor contained 5128 grams of manure and 167 grams of hay. The mixture was made as homogenous as possible. The three reactors were packed in a consistent manner to achieve comparable hydraulic conductivities. Approximately 450 grams of a seed culture (a 9% ratio of inoculum to substrate) was mixed into each bioreactor to promote bacterial activity (Bolis 1991). Lema et al. (1991) suggest 10% as a reasonable ratio of inoculum to substrate as this shortens the start-up period in anaerobic reactors.

Manure was chosen as a substrate based on successful results with that substrate by Bolis (1992). Composted cow manure was found to have a high pH which aided in buffering the acidic water. Hay was chosen as a nutrient amendment (for easily assimilated carbon) because it was found to effectively increase sulfate reduction (Reynolds et al. 1991).

After the bioreactors were filled with substrate they were soaked in acid mine drainage for one week. The bioreactors were operated in an upflow configuration with a plug flow hydraulic regime. Hydraulic conductivity was measured using a falling head permeameter (Figure 6) and equation (8) (EPA 1986).

$$K = \frac{2.3\ aL}{At} \log_{10} \frac{h_0}{h_1} \qquad (8)$$

where:

a = the cross-sectional area of the standpipe

A = the cross-sectional area of the substrate

L = the length of the substrate

t = the time elapsed

Hydraulic conductivity of the laboratory bioreactors ranged from 10^{-2} cm/sec to 10^{-4} cm/sec, slightly lower than, but generally consistent with, previous research (Bolis 1992; Lemke 1989). The same manure was used in each reactor throughout the experimental period. That is, the bioreactors were not re-packed between each residence time.

The acid mine drainage used in the laboratory was collected at an access point in the slurry line approximately 500 feet upstream from the treatment plant. This water had a slightly different chemistry than did the water used in the mine system. This is because the water was aerated as it travelled from the mine, through the slurry line to the treatment plant. Chemistry of the water used in the laboratory is shown in Table 5.

Water was collected in 15 gallon Nalgene containers and transported back to the laboratory. The containers were kept

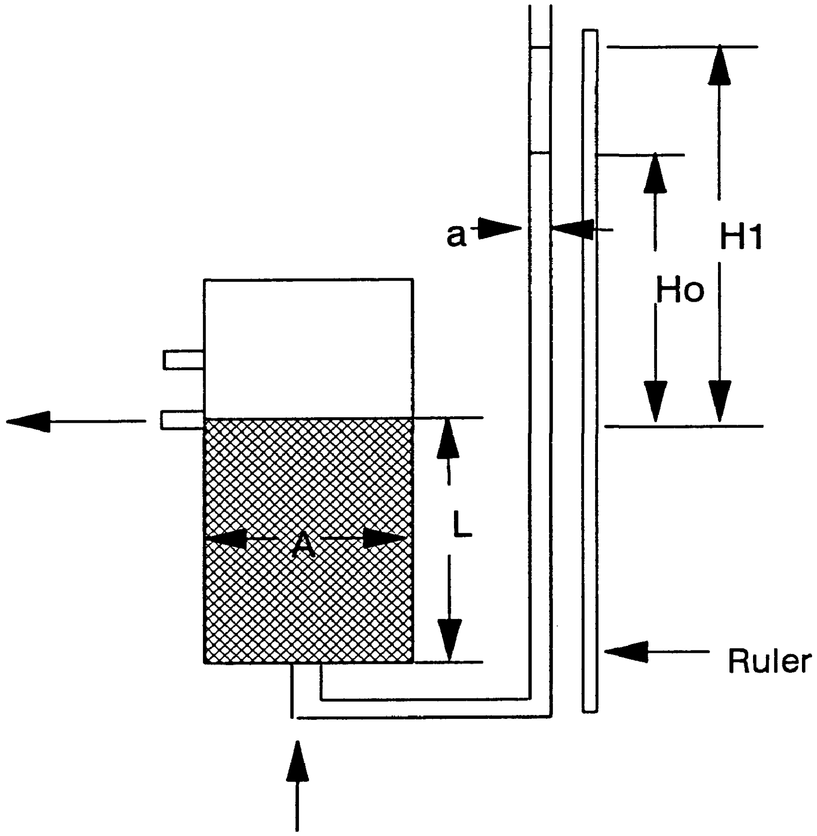

Figure 6. Schematic of falling head permeameter used to measure hydraulic conductivity of bench scale bioreactors.

Table 5. Water quality parameters of acid mine drainage used in the laboratory (concentration in mg/L). Values shown are for dissolved metals.

Cadmium	0.26±0.03
Copper	1.39±0.19
Iron	194±45
Lead	0.15±0.02
Manganese	171±22
Zinc	113±15
Sulfate	4630±316
pH	2.92±0.08
Eh (mV)	452.9±56.3
Conductivity (mmhos)	6084±300

tightly closed until use. Water was pumped into the bioreactors using a Masterflex multichannel, peristaltic, variable speed pump.

The volume of the single stage system was one-half the volume of the series system, therefore, the single stage system was operated at one-half the flow rate of the series system to maintain the same residence time in both systems. Flow rates were verified at least three times per week by measuring how much was collected over a 10 minute period.

Eagle Mine

The bioreactor used in the mine was constructed by Staub (1992). The bioreactor was constructed from a 200-gallon Nalgene HDPE tank with a packed substrate volume of 0.55 m^3, a diameter of 37 inches and a height of 31 inches. A perforated inflow pipe was embedded in six inches of gravel to distribute the mine drainage. A layer of landscaping fabric was placed over the gravel to keep the manure and gravel from mixing. Substrate in the tank consisted of a 3:1 mixture by volume of manure and hay. A layer of hay was placed above the substrate to aid in providing anaerobic conditions in the tank. An illustration of the bioreactor is shown in Figure 7.

The bioreactor was operated in an upflow configuration and was fed from a constant head reservoir tank. Figure 8 illustrates the layout of the mine system. The reservoir tank was fed from a dam constructed above the system providing a constant gravity flow to the tank. Flow into the bioreactor was controlled by a valve placed upstream from the reactor. Flow rates were calculated and reset once per week. Flow rates were calculated by measuring effluent collected for one minute.

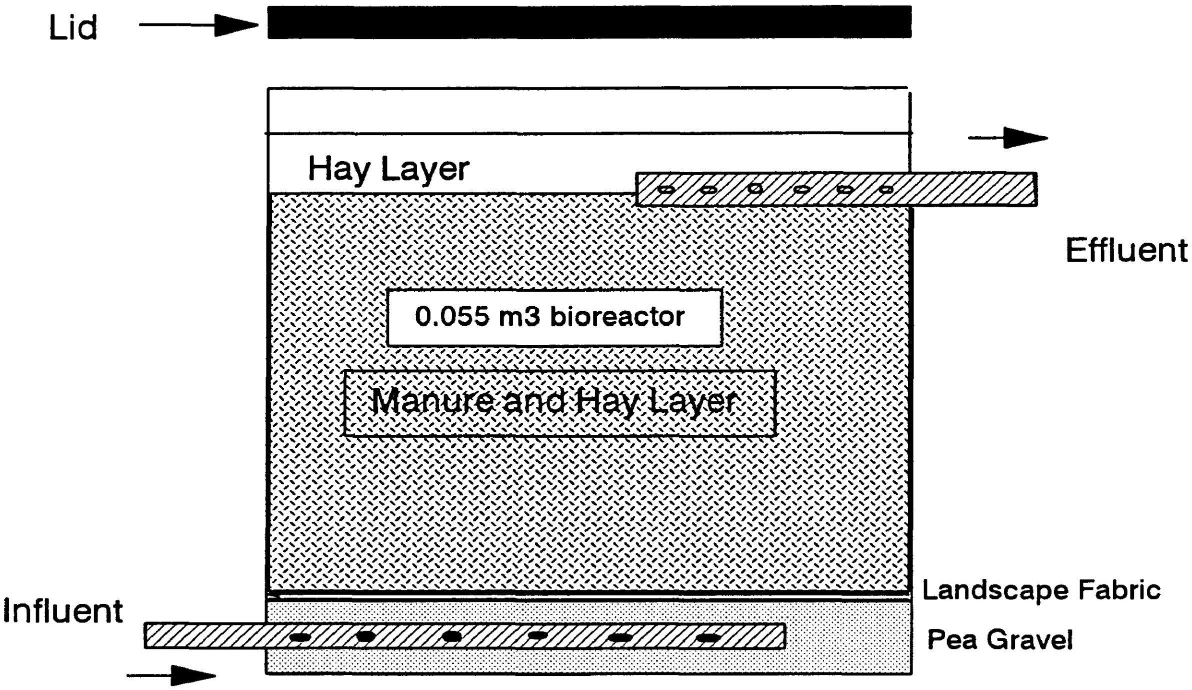

Figure 7. Schematic of 200 gallon bioreactor used in Eagle Mine field study.

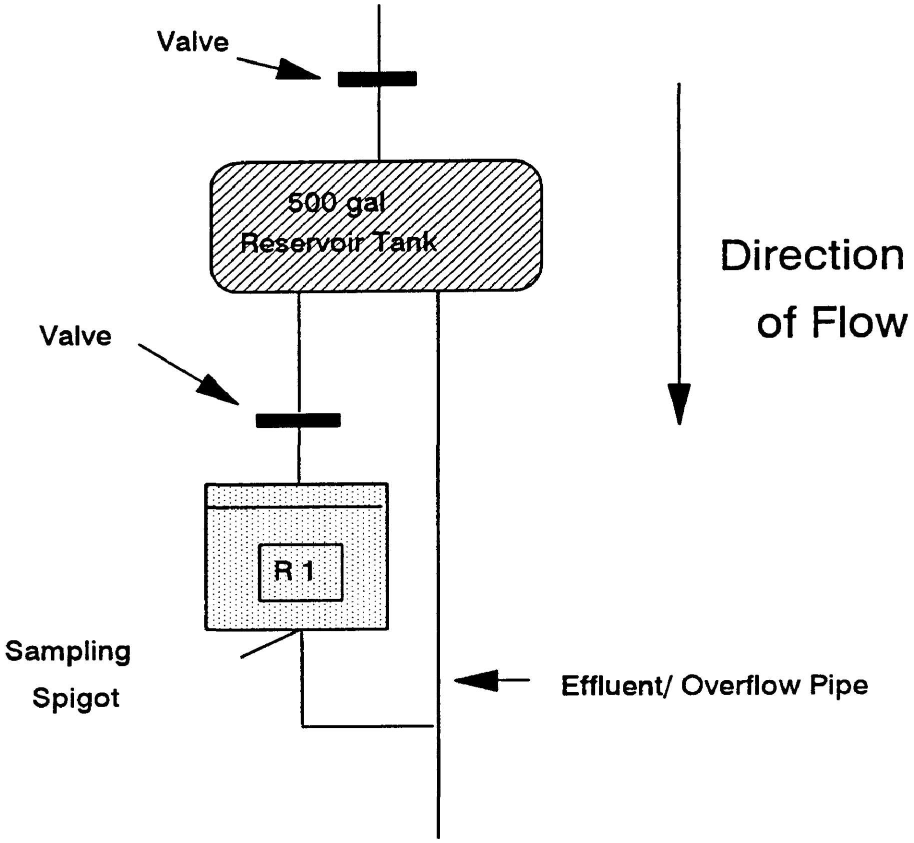

Figure 8. Plan view diagram of layout of bioreactor and constant head tank used in the Eagle Mine.

Selection of Flow Rates

Initial laboratory flow rates were selected at levels that have been shown to offer metal removal efficiencies of approximately 90% (Hurley Euler 1992; Staub 1992). A comparison of flow rate to volume ratios from previous work produced a flow rate of 2 ml/min for the volume of 0.007 m^3 used in the single stage system.

This flow rate was verified in a comparison of metal loading rate to sulfide production rates reported in the literature. The total molar concentration of metals in the mine drainage was calculated to be 8.35 mmol/L. The sulfide production rate reported for mushroom compost amended with hay was 0.00344 mmol S^{2-}/cm^3 substrate/day. Applying this rate of sulfide production to the volume of substrate in one bioreactor gave an estimated rate of sulfide production of 0.0167 mmol/min. This corresponds to a metal loading rate of 0.0167 mmol/min which correlates to a flow rate of 2.5 mL/min in the single stage system.

The series reactors, therefore, were operated initially at 2 ml/min and the parallel reactor was operated at 1 ml/min. After operation at this initial flow rate the flow rate was increased by one order of magnitude. Further increases or decreases were based on the results obtained for these flow rates. Subsequent flow rates for the series reactors were determined to be 5 mL/min, 10 mL/min and 20 mL/min. Analogous flow rates for the single stage reactor were 2.5 mL/min, 5 mL/min and 10 mL/min.

The flow rate in the mine system was based on data collected in the laboratory. A flow rate was calculated to operate the reactor in the mine at the lowest residence time observed in the laboratory. The highest flow rate run in the laboratory was 20 ml/min for the series reactors and 10 ml/min for parallel reactor. Using the laboratory flow rate to volume ratios with the volume of the bioreactor in the mine, an initial flow rate for the mine system was determined to be 800 ml/min.

Sampling

Laboratory

Influent and effluent samples were collected once per week for analysis for the following parameters: pH, Eh, conductivity, sulfate, cadmium, copper, iron, lead, manganese, and zinc. The pH, Eh, and conductivity were measured immediately after collection. Sulfate samples were collected and stored in a refrigerator for up to 48 hours. Metals samples were filtered through a 0.45 um glass filter using a 2.4 liter Geotech barrel filter to removed suspended solids. Metals samples were collected into sample bottles containing a 1 molar nitric acid preservative. All samples were taken to the laboratory for analysis within 48 hours of collection.

Eagle Mine

Influent and effluent samples were collected once per week at the mine site. Samples were transported to an office approximately three miles away from the site for processing. The procedures used for sample collection and preparation in the laboratory were used at the mine site. The same set of parameters listed in the laboratory sampling section above were determined for the mine samples.

Quality Control/Quality Assurance

A split and a duplicate sample of the mine drainage were taken every other week to determine the quality of the analyses and field variability. A blank sample using distilled water was taken each week to check for cross-contamination in the filtering device.

Analysis

The pH and Eh were measured using a Beckman pH/Eh meter. For Eh measurement a Corning Redox Combination electrode calibrated to Light's solution was used. For determination of pH a VWR Scientific Sealed Combination electrode was used. Conductivity was measured with a VWR Extended Range Digital Conductivity Meter and probe.

Analyses for metals and sulfate were performed by Vista Laboratories, Broomfield, Colorado. Copper, iron, manganese and zinc were analyzed on a Thermo Jarrell Ash 61E inductively coupled argon plasma emission spectrometer (ICAP). Cadmium and lead were analyzed on a Perkin Elmer 2380 graphite furnace atomic adsorption spectrophotometer (GFAA). Sulfate was analyzed by spectrophotometry according to EPA method 375.4.

CHAPTER 4

RESULTS AND DISCUSSION

Physical Observations: Laboratory

The most apparent changes initially observed in the system are a hydrogen sulfide odor (the smell of rotten eggs) and a color change in the effluent. Since H_2S is one of the end products formed by the sulfate reducing bacteria, the H_2S odor is a good indication that the bacteria are operating.

The color of the influent water is pale yellow and fairly clear. Initially, the effluent color turns to what can best be described as the color of a stout beer. This is due to the removal of fine particles from the manure which drastically increases suspended solids in the effluent. The high suspended solids content causes problems when the samples are filtered for metals analysis. The filters tend to clog quickly and, therefore, need to be replaced frequently. This condition clears up after about one month. The color of the effluent turns to that of iced tea and the suspended solids decrease. Filtering at this point is much quicker.

Clear plexiglass reactors allow for visual inspection of the substrate with time. Initially, the substrate is black, the color of composted manure. After about one month, the substrate begins to turn brown. This change begins at the bottom and moves up through the reactor (since it is an upflow system). This color change is probably due to iron hydroxides and iron carbonates forming in the manure. At the top of the substrate level, where the effluent leaves the reactor, an orange precipitate has formed. This is probably from iron oxy-hydroxides.

Clogging due to iron hydroxide is a problem in all parts of the system. Since the mine drainage is forced through the reactors with a peristaltic pump, the point at which the feed tube contacts the pump tends to clog. As the iron concentration increases in the effluent at the lower residence times, iron oxy-hydroxides form in the outflow tube, causing clogs to occur. Frequent cleaning of the laboratory system is necessary since the tubing used is so small (0.14 mm).

Finally, after increasing the flow rate by an order of magnitude, it was difficult to maintain a consistent flow rate. The first residence time examined is the longest residence time tested in this study (Table 6). This residence time correlates to the lowest flow rate used. The next sequential residence time examined is the lowest residence time in this study. This residence time correlates to the highest flow rate used. After the flow rate increase, the system took approximately two weeks to maintain a consistent flow rate. The first stage of the series reactors remained clogged until a tube that had been placed on the overflow outlet was removed. This seemed to vent the gas the system was then able to maintain a stable flow rate. Subsequent decreases in flow rates took were established very quickly.

Table 6. Flow rates and hydraulic residence times used in bench scale investigations.

	Series System		Single stage System	
Date	Flow rate (mL/min)	Res time (h)	Flow rate (mL/min)	Res time (h)
4/8/92	*	*	1.2	97.3
4/15/92	2.6	89.8	1.1	106.2
4/22/92	2.3	101.6	0.9	129.8
4/29/92	2.2	106.2	1.1	106.2
5/18/92	26.2	8.9	14.4	8.1
5/26/92	11.3	20.7	11.8	9.9
6/3/92	22.5	10.4	10.3	11.3
7/9/92	10.1	23.1	5.6	20.9
7/13/92	10.1	23.1	5.6	20.9
7/15/92	9.4	24.9	6.5	18.0
7/28/92	5	46.7	2.4	48.7

* data excluded-clogged reactor

Laboratory Results

Raw data from bench scale investigations are presented in Appendix A. A discussion of these data and specific points of interest follow. On 4/8/92 The series system clogged to a very low flow rate. All of the data for this date, including flow rate, residence time, metals and sulfate concentrations, pH, Eh and conductivity, were removed from the results list for the series system. The data for SO_4 on 4/15/92 was excluded due to a laboratory error which gave inaccurate readings. No other data is missing from these tables. Detection limits are listed at the bottom of the table.

The results have been analyzed to determine if a relationship exists between measured constituents and the residence time. Hydraulic residence time is calculated by equation (9):

$$T = \frac{V}{Q} \tag{9}$$

where
T = residence time, (h)
V = empty bed volume, (cm^3)
Q = flow rate, (mL/min)

Statistical analyses were done on the data obtained from each system to determine if there was a significant difference in performance between the two systems. No significant difference in performance was determined. The data were then averaged over each residence time for each system and plotted against residence time. A discussion of the statistical analyses on these averaged values is presented in a later section. Mean values are presented for the mine drainage and for effluent from each reactor in Tables 7-10. Only one data point was collected for the 50 hour residence time. Therefore, no mean or standard deviation could be presented for that residence time. Values averaged over each residence time from the series and the single stage system have been plotted against average residence times in the following graphs. Where possible, discharge permit limits for the existing Eagle Mine water treatment plant are indicated. Each constituent measured is discussed individually in the following sections.

Table 7. Experiment 1: T = 100 hours. Values are averaged over the 100 hour residence time. Concentrations are in mg/L, Eh is in mV and conductivity is in mmhos/cm.

Constituent	Influent	Effluent Series System	Effluent Single Stage
Cadmium	0.25 ± 0.02	<	<
Copper	1.55 ± 0.06	<	<
Lead	0.16 ± 0.03	<	<
Iron	225 ± 13	1.0 ± 0.15	<
Manganese	188 ± 5	111 ± 19[a]	34 ± 25[a]
Zinc	125 ± 6	<	<
Sulfate	4967 ± 351	4100 ± 283[b]	4133 ± 153[b]
pH	2.90 ± 0.04	7.03 ± 0.17	7.25 ± 0.24
Eh	449.4 ± 15.3	-117.4 ± 74[b]	-48.0 ± 199[b]
Conductivity	6438 ± 107	5967 ± 1672[b]	7072 ± 1454[b]

[a] Values that do not meet Eagle Mine treatment plant discharge permit limits.

[b] No permit limit information is available.

< Constituent not detected at or above the listed reporting limit.

Reporting Limits:

Cd	0.0005 - 0.03 mg/L	Fe	0.1 - 0.5 mg/L
Cu	0.02 - 0.1 mg/L	Mn	0.01 - 0.05 mg/L
Pb	0.003 - 0.03 mg/L	Zn	0.02 - 0.1 mg/L

Table 8. Experiment 2: T = 10 hours. Values are averaged over the 10 hour residence time. Concentrations are in mg/L, Eh is in mV and conductivity is in mmhos/cm.

Constituent	Influent	Effluent Series System	Effluent Single Stage
Cadmium	0.29 ± 0.03	<	<
Copper	1.23 ± 0.06	<	<
Lead	0.15 ± 0.02	<	<
Iron	157 ± 15	123 ± 6[a]	112 ± 24[a]
Manganese	157 ± 6	153 ± 6[a]	157 ± 15[a]
Zinc	103 ± 6	14 ± 11[a]	21 ± 15[a]
Sulfate	4467 ± 115	4267 ± 379[b]	4367 ± 208[b]
pH	2.96 ± 0.03	6.04 ± 0.13[a]	6.17 ± 0.11[a]
Eh	432.4 ± 16.4	-10.1 ± 8.7[b]	-13.1 ± 13[b]
Conductivity	5800 ± 142	5510 ± 122[b]	5827 ± 406[b]

[a] Values that do not meet Eagle Mine treatment plant discharge permit limits.

[b] No permit limit information is available.

< Constituent not detected at or above the listed reporting limit.

Reporting Limits:

Cd	0.0005 - 0.03 mg/L	Fe	0.1 - 0.5 mg/L
Cu	0.02 - 0.1 mg/L	Mn	0.01 - 0.05 mg/L
Pb	0.003 - 0.03 mg/L	Zn	0.02 - 0.1 mg/L

Table 9. Experiment 3: T = 20 hours. Values are averaged over the 20 hour residence time. Concentrations are in mg/L, Eh is in mV and conductivity is in mmhos/cm.

Constituent	Influent	Effluent Series System	Effluent Single Stage
Cadmium	0.25 ± 0.04	<	<
Copper	1.40 ± 0.26	<	<
Lead	0.12 ± 0.01	<	<
Iron	217 ± 46	98 ± 37[a]	104 ± 33[a]
Manganese	170 ± 35	143 ± 6[a]	187 ± 24[a]
Zinc	112 ± 24	3 ± 2[a]	8 ± 2[a]
Sulfate	4600 ± 100	4233 ± 58[b]	4233 ± 47[b]
pH	2.97 ± 0.12	6.06 ± 0.10[a]	5.95 ± 0.26[a]
Eh	423.8 ± 10.7	-8.4 ± 35.1[b]	67.0 ± 42.2[b]
Conductivity	5957 ± 38	5500 ± 26[b]	5450 ± 16[b]

[a] Values that do not meet Eagle Mine treatment plant discharge permit limits.

[b] No permit limit information is available.

< Constituent not detected at or above the listed reporting limit.

Reporting Limits:

Cd	0.0005 - 0.03 mg/L	Fe	0.1 - 0.5 mg/L
Cu	0.02 - 0.1 mg/L	Mn	0.01 - 0.05 mg/L
Pb	0.003 - 0.03 mg/L	Zn	0.02 - 0.1 mg/L

Table 10. Experiment 4: T = 50 hours. Only one data point per system is available. Concentrations are in mg/L, Eh is in mV and conductivity is in mmhos/cm.

Constituent	Influent	Effluent Series System	Effluent Single Stage
Cadmium	0.25	<	<
Copper	1.2	<	<
Lead	0.17	<	<
Iron	120	100[a]	54[a]
Manganese	150	150[a]	150[a]
Zinc	100	5.0[a]	2.9[a]
Sulfate	4200	4000[b]	4100[b]
pH	2.79	6.07[a]	4.87[a]
Eh	615.4	67.6[b]	221.5[b]
Conductivity	5900	5280[b]	5240[b]

[a] Values that do not meet Eagle Mine treatment plant discharge permit limits.

[b] No permit limit information is available.

< Constituent not detected at or above the listed reporting limit.

Reporting Limits:

Cd	0.0005 - 0.03 mg/L	Fe	0.1 - 0.5 mg/L
Cu	0.02 - 0.1 mg/L	Mn	0.01 - 0.05 mg/L
Pb	0.003 - 0.03 mg/L	Zn	0.02 - 0.1 mg/L

pH.

Figure 9 shows average pH values vs average residence time. At the longest residence time of around 100 hours, the average pH has risen from 2.90 to 7.03 in the series system and 7.25 in the single stage system. This is above the lower permit limit value of 6.5, indicated on the graph. At residence times below 50 hours average pH values have dropped below 6.5, therefore below permit limits. These values are centered around 6.0 except for the single stage reactor at the 48 hour residence time. This may be due to manipulation of the system to remove a clog less than 24 hours before the sample period. The flow rate was turned up in en effort to remove the clog. It took 48 hours for one empty bed volume of mine drainage to be completely replaced at a 50 hour residence time. There may not have been sufficient time to replace the low pH water that flooded the system while the clog was being worked out.

As stated earlier, the pH of the bioreactor must be near neutral for efficient metal removal. The pH data from Tables 8-10 indicate that metals removal decreases at lower residence times, however, the pH is still raised to around 6.0 in the effluent. This is indicative of the ability of the SRB to produce enough bicarbonate to neutralize their environment even though they are being flooded with highly

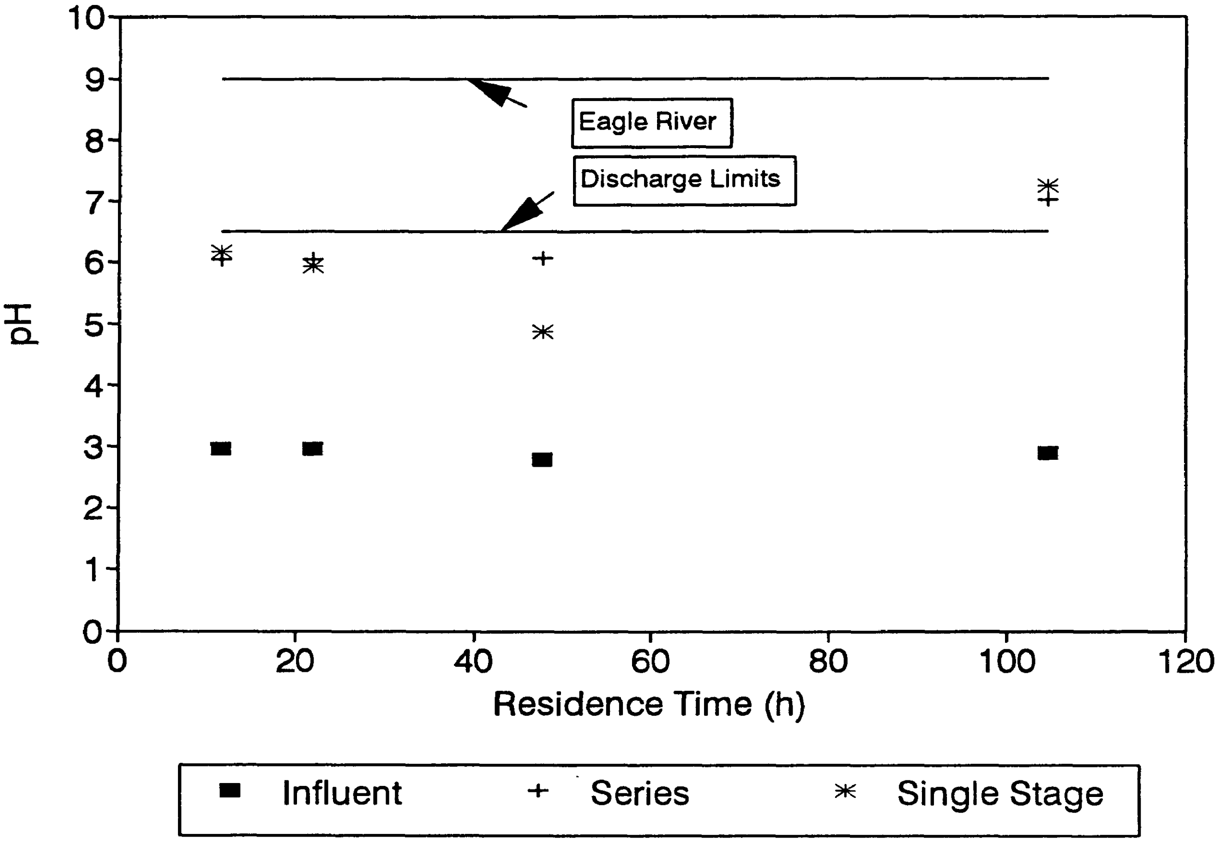

Figure 9. Graph showing average pH vs average residence time obtained from laboratory studies. Eagle Mine treatment plant discharge permit limit for pH is indicated.

acidic water.

Eh.

As stated earlier, bioreactors must maintain a reducing environment in order for SRB to reduce sulfate, and for metal sulfides to form. The Eh values listed in Tables 7 through 10 show that at a residence time of around 100 hours, the average Eh values are less than 0 mV in both systems. At residence times lower than 50 hours, the Eh is reduced from influent values but not to the extent where efficient metal sulfide precipitation can occur. The consortium of heterotrophic bacteria responsible for reduction of O_2 may lose their ability to reduce O_2. The rate of O_2 reduction may not be high enough to compensate for the mass loading of O_2. Effluent concentrations of Fe, Mn and Zn are increased, since sulfides of Fe, Mn and Zn are dependent upon highly reducing conditions. This is discussed in subsequent sections.

Conductivity.

Conductivity is a measure of the number of ions present in the water. The number of ions lost to precipitation of metal sulfides is insignificant compared to the total number of ions present in the mine drainage, as can be seen in Tables 7 through 10.

Cadmium, Copper, and Lead.

Cadmium, copper and lead are removed close to or below reporting limits in both systems and at all residence times, as is seen in Tables 7 through 10. Although the influent concentrations of cadmium, copper, and lead vary, they are present in low enough concentrations and their metal sulfides are insoluble enough that they are easily removed at all residence times. Therefore they are below Eagle Mine discharge limits at all residence times.

Iron

Tables 7 through 10 list iron concentrations. Iron is removed to below discharge limits at the 100 hour residence time. When the residence time is decreased, however, iron removal decreases to above permit limits, as can be seen in Figure 10. This may be due to a high concentration of Fe relative to other metals in the mine drainage. Or it may be due to the drop in average effluent pH to 6.0, in both systems.

Garrels and Christ (1965) discuss the stability of iron minerals in water containing sulfide. At a pH of 7.0 and under very reducing conditions, FeS is stable. However, below a pH of 6.0, Fe^{++}(aq) is most stable. When CO_2 is abundant at a pH of 6.0-8.0 and under strongly reducing conditions, the FeS field is eliminated and a field for $FeCO_3$ is formed. The concentration of carbonate must be high (10^0 *M*) for $FeCO_3$ to be stable. In the SRB system, the HCO_3^- is used up in neutralizing the low pH water, so there probably is not enough carbonate to produce a sufficiently stable field for only iron carbonates to form. It is more likely that iron sulfides as well as iron carbonates and iron hydroxides are forming.

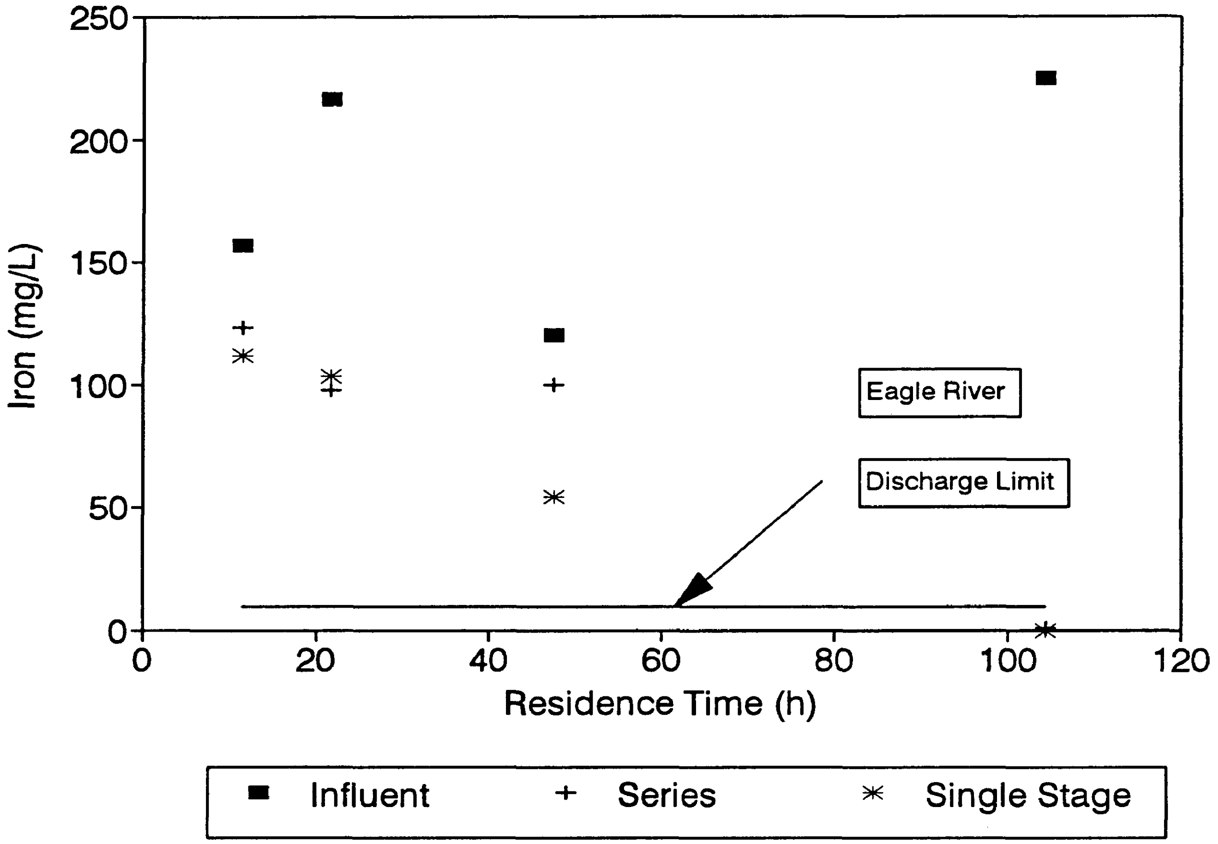

Figure 10. Graph showing average iron concentration (mg/L) vs average residence time obtained from laboratory studies. Eagle Mine treatment plant discharge permit limits for iron are indicated.

Manganese.

Figure 11 shows average manganese removal. Even at a residence time of around 100 hours, average manganese effluent concentrations are above permit discharge limits for the current treatment plant. Concentration in the single stage system has a mean value of 34 ±25 mg/L. This is considerably lower than the series system with a mean of 111 ±19 mg/L. At residence times below 50 hours, metal concentrations are approximately equal to influent concentrations. Manganese is difficult to remove at lower residence times because the pH drops below 7.0.

Manganese chemistry is similar to that of iron. In solutions with low redox potential, both iron and manganese are stable as divalent ions. The mono-sulfide of manganese, MnS, is much more soluble than FeS, however. Also, iron compounds oxidize more readily than do manganese compounds.

In an Eh-pH diagram, $MnCO_3$ is stable over a wide range of Eh and pH if dissolved carbonate is high (1*M*). MnS does not form unless sulfide is much more concentrated than carbonate (i.e. if total dissolved sulfur exceeds total carbonate by a factor of 100) (Krauskopf 1967). Even if the pH drops to 6.0, $MnCO_3$ will form as long as there is sufficient carbonate. In this system, it is more likely that MnS is forming, since the carbonate concentration is low relative to the sulfur concentration. As more Mn is loaded on to the system and the pH drops below 6.0, Mn^{++} becomes stable. This is consistent with results obtained from experiments at shorter residence times.

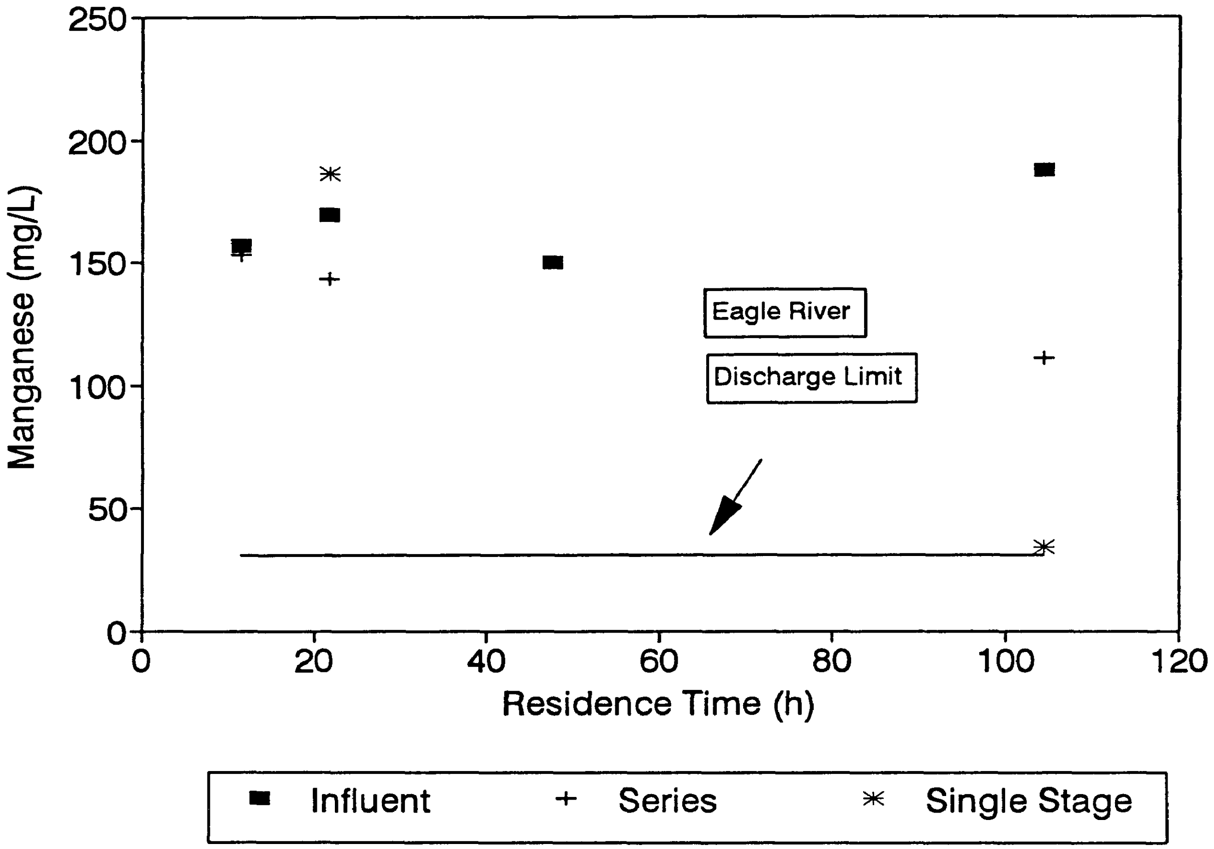

Figure 11. Graph showing average manganese concentration (mg/L) vs average residence time obtained from laboratory studies. Eagle Mine discharge permits limits for manganese are indicated.

Zinc.

At a residence time of 100 hours, zinc is removed to below detection limits in both systems, as is seen in Figure 12. At a residence time of less than 50 hours, effluent zinc concentrations increase to above permit limits, which are very low at 0.4 mg/L. As the residence time is reduced, the effluent concentration increases. Zinc sulfide formation is dependent on pH, which decreases at lower residence times. Therefore, zinc removal in both systems is most efficient at a residence time of 100 hours.

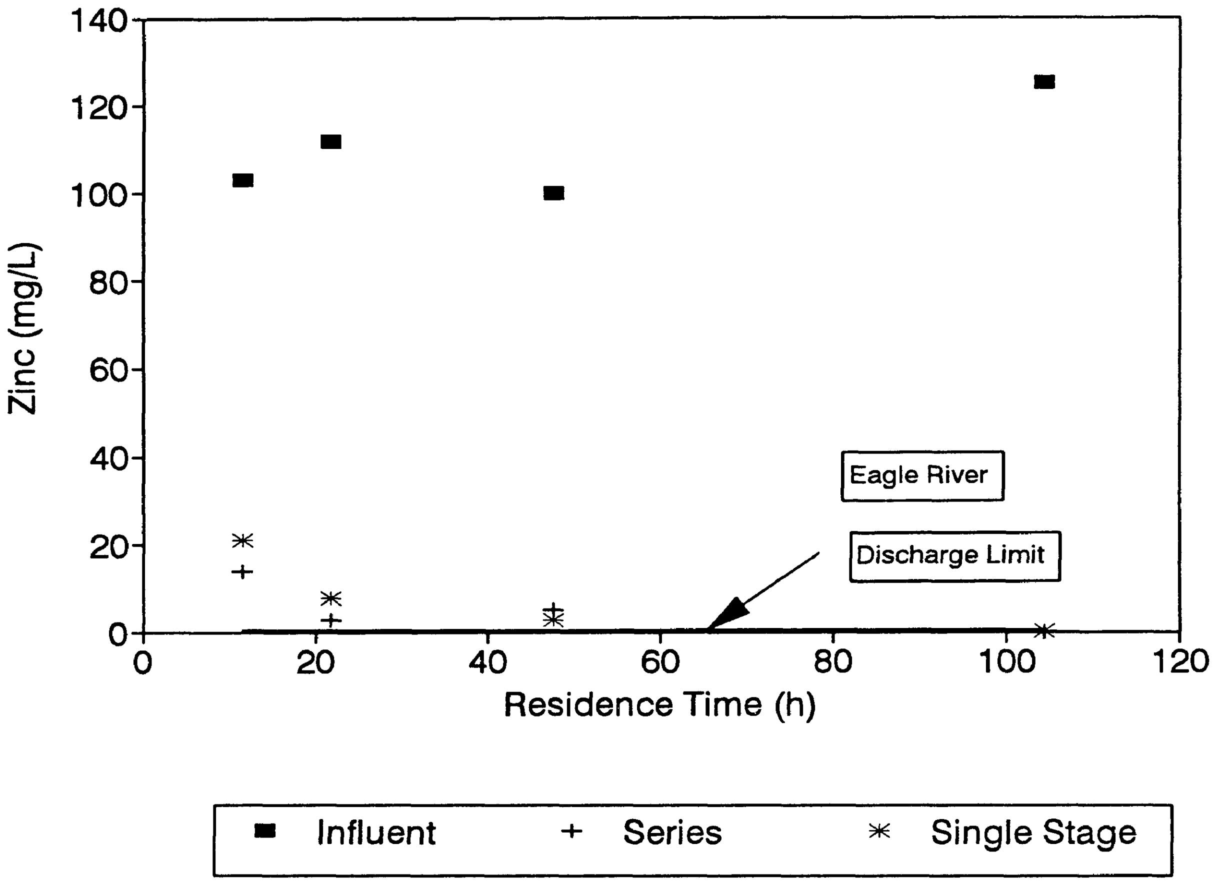

Figure 12. Graph showing average zinc concentration (mg/L) vs average residence time obtained from laboratory studies. Eagle Mine treatment plant discharge permit limits for zinc are indicated.

Iron, Manganese and Zinc.

Iron, manganese and zinc are present in high concentrations in the influent water relative to the cadmium, copper and lead (approximately 200 mg/L compared to less than 20 mg/L). Even when the residence time is reduced to 10 hours, cadmium, copper and lead are removed to almost 100%. This may be due to the fact that sulfides of Cd, Cu and Pb are insoluble and stable over a wide range of pH and Eh conditions. Nevertheless, the SRB can not produce enough sulfide, given the volume of substrate used, to completely remove all of the metals at all residence times examined. Consequently the metals of higher influent concentrations are not completely removed.

Sulfate

At a residence time of around 100 hours, the effluent sulfate concentrations decrease by an average of approximately 800 mg/L. Below the 20 hour residence time, effluent values are much closer to influent values, decreasing by only 100-300 mg/L, on average. Sulfate does not change by large numbers because the concentrations of metals in the influent water are low relative to the concentrations of influent sulfate.

Using the values calculated for metal removal in Table

11, and assuming that one mole of metal removed equals one mole of sulfide produced, rates of sulfide production can be estimated for this system. For example, at an average residence time of 99 hours in the series reactor, 7.4 mmol/L of metals are removed. A corresponding concentration of 7.4 mmol/L of S^{2-} is formed to remove the metals as metal sulfides. The sulfide production rate can be estimated for subsequent residence times using reactor volumes and average flow rates. These values are listed below.

Residence Time	Rate of Sulfide Production	
(h)	(nmol/mL/d)	
	Series	Single
10	3789	5143
20	4354	5548
50	925	1331
100	1790	1922

These estimated rates of sulfide production are generally consistent with those reported by Reynolds (1991). The substrate that gave the highest rate of sulfide production in that study was mushroom compost amended with hay extract. The experiment was performed as a batch system. It may be that a continuous-flow system produces a slightly higher sulfide production rate because the bacterial waste is washed away. It would be worthwhile in future studies to determine what component of this system actually accelerates bacterial activity.

Table 11. Concentration of sulfate removed compared to total metals removed. Values are averaged over residence times.

Res Time (h)	Removal of metals and sulfate (mmol/L)	Series Reactors	Single Reactor
10	SO_4	2.1	1.0
	Metals	2.1	2.1
20	SO_4	3.8	3.8
	Metals	4.3	4.6
50	SO_4	2.1	1.0
	Metals	1.8	2.7
100	SO_4	9.0	8.7
	Metals	7.4	8.8

Metal Removal Capacity

It is worthwhile to examine the capacity of the systems to remove metals and sulfate in terms of the rate of metals removed, for the residence times examined. This is calculated according to equation (10):

$$MRC = (C_{in} - C_{out}) * \frac{Q}{V} \tag{10}$$

where,
C_{in} = influent concentration (mg/L)
C_{out} = effluent concentration (mg/L)
Q = flow rate (mL/min)
V = reactor volume (cm^3)

This estimates mass removed per unit time per unit volume. These values are presented in Table 11. At the 20, 50 and 100 hour residence times both the series system and the single stage system perform similarly, as is illustrated in Figure 13. From 100 hours to 50 hours there is a decrease in the capacity of both systems to remove metals and sulfate. This may indicate that the bacteria are incapable of producing sufficient sulfide. High effluent Eh values may indicate that the bacteria were overwhelmed by the high flow rates observed in Experiments 2 and 3 and were not able to recover. Since only one data point was taken for each reactor at the 50 hour residence time, it is difficult to draw firm conclusions about performance of the system in Experiment 4. This situation might be resolved by repeating the experiment with fresh substrate for each change in flow rate.

Experiments 2 and 3 were run at the two lowest residence times, 10 and 20 hours. Metal removal capacity was highest of all the times at the 20 hour residence time, then it decreased slightly at the shortest residence time. The single stage system showed some variation between the amount of sulfate removed and the amount of metals removed. This was not a significant difference though.

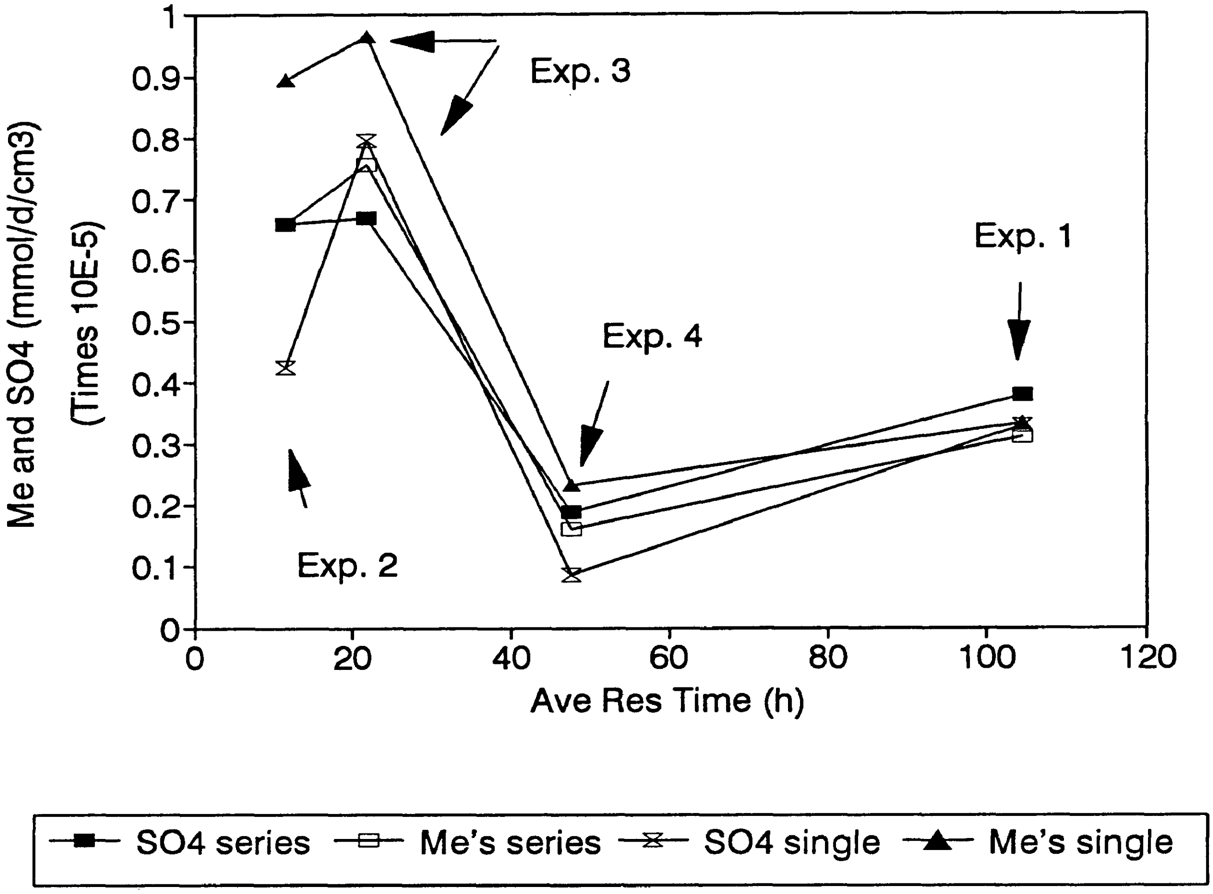

Figure 13. Graph showing total metal and sulfate removal capacity (mmol/d/cm^3) vs average residence time. Arrows indicate which experiment corresponds to each residence time.

Statistical Analysis of Laboratory Results

The laboratory data have been analyzed with twosample-t tests (Ryan et al. 1985) to determine if there is a statistically significant difference between results obtained for each system. These results are presented in Appendix B.

The means for effluent values of pH, iron and zinc from each system were compared over the endpoint point residence times examined. These parameters were the only parameters that showed a relationship between effluent values and residence time. The twosample-t tests indicated that mean values of pH, iron concentration and zinc concentration in the effluent were not significantly different between the two systems.

The other parameters did not show any relationship. Cadmium, copper and lead were removed almost completely to below reporting limits at all residence times. Eh, conductivity, and sulfate values were so scattered that it was difficult to determine a trend. Manganese values were also scattered and were never below the Eagle Mine discharge limits. Therefore, no statistical analyses were performed on these parameters.

The values for pH, iron and zinc were then combined for each system over the endpoint residence times (11.6 ±3.1 h and 104.3 ±7.8 h). Means and standard deviations were calculated and the two means compared with twosample-t tests. These results are listed in Appendix B.

pH

The mean pH values of the longest and shortest residence times, respectively, are 7.16 ±0.23 and 6.11 ±0.13. The pH from the lower residence time has a smaller standard deviation than the pH from the higher time. These means are significantly different from each other. The mean of the longer residence time is above permit limits. The mean of the shorter time is below the limits. Therefore, to keep the effluent within permit limits for pH, the system would need to be designed around the 100 hour residence time.

Fe and Zn

At the longer residence time, the mean effluent concentration of iron is 0.53 ±0.62 mg/L and of zinc is 0.01±0.03 mg/L. At the 10 hour residence time, the mean of iron is 117.5 ±16.7 mg/L and of zinc is 17.4±12.5 mg/L. Both iron and zinc are below discharge permits at the longer residence times. Likewise, both metals are above permit limits at the shorter residence times. Residence time does have a significant effect on removal of iron and zinc.

All of the metals except Mn have acceptable mean effluent concentrations (according to permit limits) at the 100 hour residence time. At the shorter residence times the bacteria show overall increased rates of sulfate reduction than at the longer residence times. Nevertheless, they do not seem to be capable of producing enough sulfide to precipitate enough of the metals present to bring effluent concentrations down to current discharge permit limits. This should be considered when designing a PMDTS.

Physical Observations: Eagle Mine

The reactor used at the mine was shut down for six months prior to this study. During this time, iron hydroxide precipitates may have formed in the piping, causing clogging to occur. Consequently, flow rates were hard to control in this reactor, and only four weeks of data were collected.

Controlling flow rates with a valve was not successful, as the valve clogged easily. The head provided by the feed tank did not seem to keep the flow rate consistent or keep the clogging down. In fact, the head was lost once the water got through the valve. If a pump is not available, systems should be designed to run by constant head with no valves. This will create the most consistent flow condition.

Eagle Mine Results

Data collected from the mine system from the four weeks it was operated are presented in Appendix C. As was stated, the flow rate could not be controlled well, however some results that were consistent with bench scale results were obtained.

The pH values from all four weeks are raised from around 3.0 to above 6.0. All four Eh readings show a decrease in Eh from oxidating values to reducing values. This indicates that, even after a period of no flow, the bacteria are able create a suitable environment for some metal sulfide formation.

Conductivity shows no trend. The values are scattered. Sulfate, on the other hand, is reduced for the first three weeks. There appears to be a limited period of re-adjustment for re-adsorption after a period of shut down.

As was the case with the laboratory reactors, cadmium, copper and lead are removed to detection limits or lower. These results indicate that cadmium, copper and lead are consistently removed regardless of the residence time and influent concentration.

Iron, manganese and zinc are significantly affected by the pH, as is seen in the field data. The first sample collected is the only one with a pH above 7.0. Consequently iron and manganese effluent concentrations are below discharge limits in that sample. Zinc is slightly above permit limits. The pH values from subsequent samples are closer to 6.0 and the effluent concentrations of Fe, Mn and Zn are all above permit limits.

Quality Control/Quality Assurance

Data gathered from QA/QC samples is presented in Appendix D. Blank, split and duplicate sample concentrations are listed. Also, one total metals sample was taken. The blank samples tend to have manganese in them, but this is usually at reporting limits. One SO_4 sample was calculated as 9500 mg/L. This was measured in error by the laboratory. The split and duplicate samples show nothing significant. The values are close to those used as actual sample values. The total metals sample showed all metals below detection limits except Fe and Mn. The Fe concentration was 0.7 mg/L (where the filtered sample showed Fe below reporting limits) and the Mn concentration was 55 mg/L (where the filtered sample showed Mn as 38 mg/L).

Nutrient Characteristics of Effluent

Several effluent samples were analyzed for nutrient concentrations. The parameters examined included BOD_5, COD,

$NO_2 + NO_3$, ammonia and total phosphorous. The results are presented in Table 12. The samples taken were random grab samples and based on what was collected here, it is not possible to present a nutrient profile for the system. Some of these nutrient concentrations are high. These constituents would most likely need to be removed from the effluent if the system were discharging into a regulated water body. An aerobic reactor, acting as a polishing step, might take out the nutrient constituents possibly to acceptable levels for discharge.

Table 12. Random sample effluent nutrient concentrations (mg/L). One sample is from a laboratory reactor and one sample is from the Eagle Mine reactor.

Constituent (mg/L)	Lab:Reactor 3 6/3/92	Mine:Reactor 1 7/1/92
BOD_5	19	30
COD	210	250
$NO_2 + NO_3$	<	*
Tot. Phos.	0.9	10
NH_3 as N	*	13

< = Analyte not detected at or above reporting limit.

* = no sample taken.

Relationship to Other
Colorado School of Mines Studies

Several studies using Passive Mine Drainage Treatment systems have been conducted at the Colorado School of Mines. Results from those studies, as they apply to the current study, are presented in Appendix E. The data analyzed were from the National Tunnel, Quartz Hill and Big Five drainages from Bolis (1992), the Big Five drainage, for reactors 1-3 from Hurley Euler (1992) and the Eagle Mine drainage, for the single stage reactor from Staub (1992). Effluent concentration data, as well as flow rate and volume information were examined. The same water quality parameters examined in this current study were plotted against residence time for the previous studies. Determining a relationship between metals and sulfate and the residence time was difficult because influent values were not always similar. When percentage removal of metals and sulfate was plotted against residence time, a relationship was more easily observed. Figures 14 through 19 illustrate the relationship between residence time and selected parameters examined.

Figure 14 illustrates the relationship between effluent pH and residence time. Above a residence time of 50 hours, the pH has a mean and standard deviation of 7.3±0.5. Below a

50 hour residence time, the mean pH drops to 5.6±0.8. At shorter residence times, bacterial producuction of HCO_3^-, NH_3 and H_2S may not be enough to remove H^+ ions or to buffer the pH of acidic influent water. The lower pH values have an effect on the formation of sulfides of manganese, zinc and especially iron, as is seen in the Fe, Mn and Zn graphs.

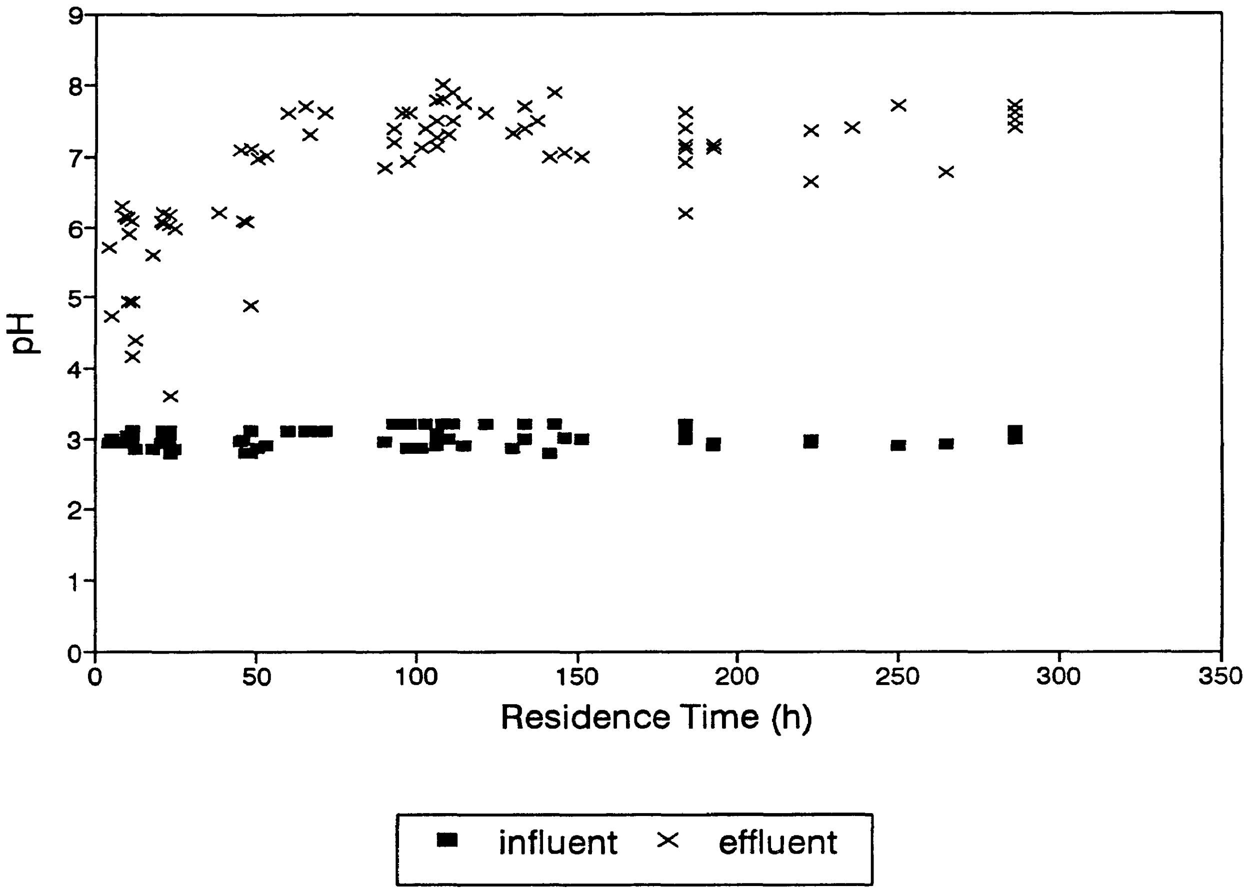

Figure 14. Graph showing composite of data from CSM studies by Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Effluent pH vs residence time.

The effluent Eh values show a lot of scatter, but generally indicate that Eh is lowered to reducing conditions by the bacteria. Above a 200 hour residence time, effluent values are well into reducing conditions. Below a 200 hour residence time, the Eh shows a lot more scatter. No statistical analysis was performed on the Eh values because there was too much scatter in the values.

Conductivity values are also scattered. Some of the effluent values are lower than the influent values and some are very close to influent values. This is expected. Some of the effluent values are higher than the influent values. This may reflect start-up periods of the reactors when there is a lot of material being washed from the substrate. This would include ions such as ammonia, humic materials and other nutrients in the substrate. These data points are so scattered, it is difficult to draw any sort of conclusions about conductivity versus residence time. Therefore, no statistical analysis was performed.

Cadmium, copper and lead are removed close to or below reporting limits at any residence time and at any influent concentration, which is consistent with results from the current study. This is probably because they are present in low molar concentrations in the mine drainage and metal sulfides of Cd, Cu and Pb are more insoluble than sulfides of Fe, Mn and Zn. No statistical analysis was performed on these metals because removal is generally close to 100%.

Percentage removal of iron from the four studies is displayed in Figure 15. Above a residence time of 50 hours removal is above 80%. Below 50 hours, percentage removal ranges from 99% to less than 0% (indicating that iron is being released into the effluent). As explained in the discussion of iron removal in the laboratory results, iron removal is dependent on pH. Below a pH of 6.0 $Fe^{++}(aq)$ is more stable than FeS. This is consistent with the results presented here.

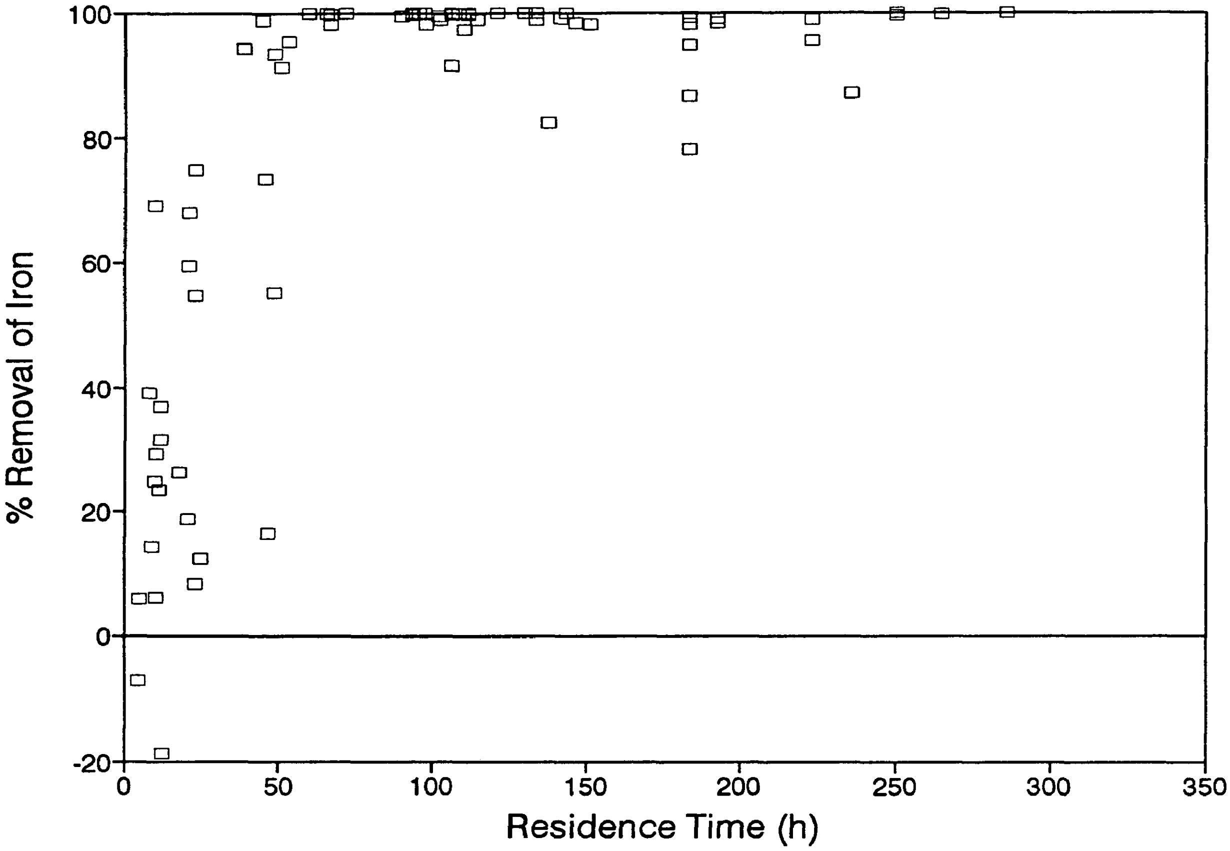

Figure 15. Graph showing composite of data from CSM studies from Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Percentage removal of iron vs residence time.

Figure 16 illustrates the relationship between percentage removal of zinc and residence time. Removal of zinc displays a similar trend to removal of iron in that at residence times above 30 hours, the removal is above 90%. At residence times below 30 hours, removal of zinc ranges from approximately 99% to 25%. This implies that a residence time of well over 30 hours is necessary to remove zinc to acceptable levels.

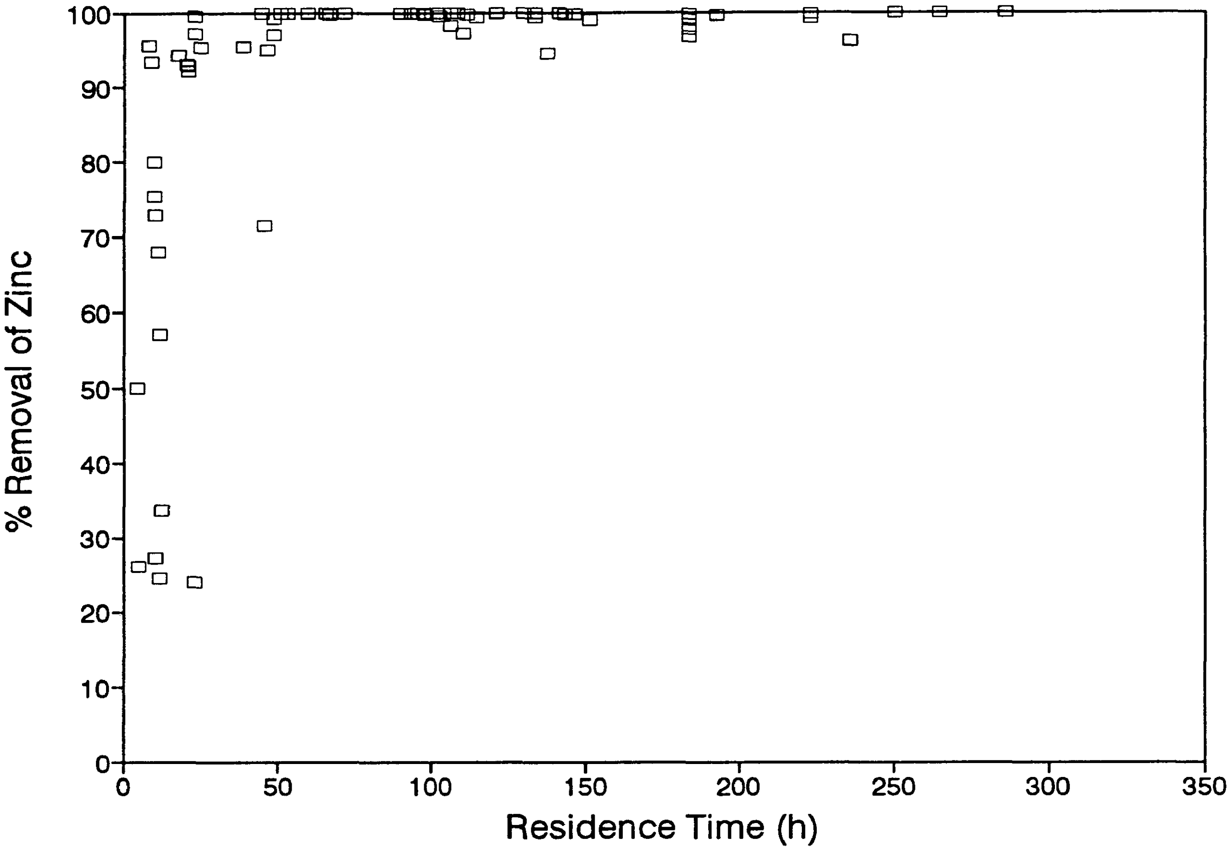

Figure 16. Graph showing composite of data from CSM studies from Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Percentage removal of zinc vs residence time.

Figure 17 displays percentage removal of manganese vs residence time. Manganese removal is scattered, but there does appear to be a general trend where at residence times above 150 hours removal is above 80%. Below 150 hours removal ranges from approximately 90% to below 0% (which indicates that manganese is being released into the effluent).

The shorter residence time produces a lower average effluent pH. Manganese removal is dependent on pH values. Below pH values of 7.0 MnS is not stable and $MnCO_3$ is stable only if there is a high carbonate concentration. The composite data show a general relationship between pH values and percent removal of manganese. Figure 18 displays this relationship. Below a pH of 6.0 removal of manganese is 20% or less. At pH values that are close to 7.0, removal of manganese ranges from less than 0% to 100%. This indicates that there is more influencing these systems for manganese precipitation than pH alone. This is consistent with the discussion of manganese chemistry in Garrels and Christ (1965).

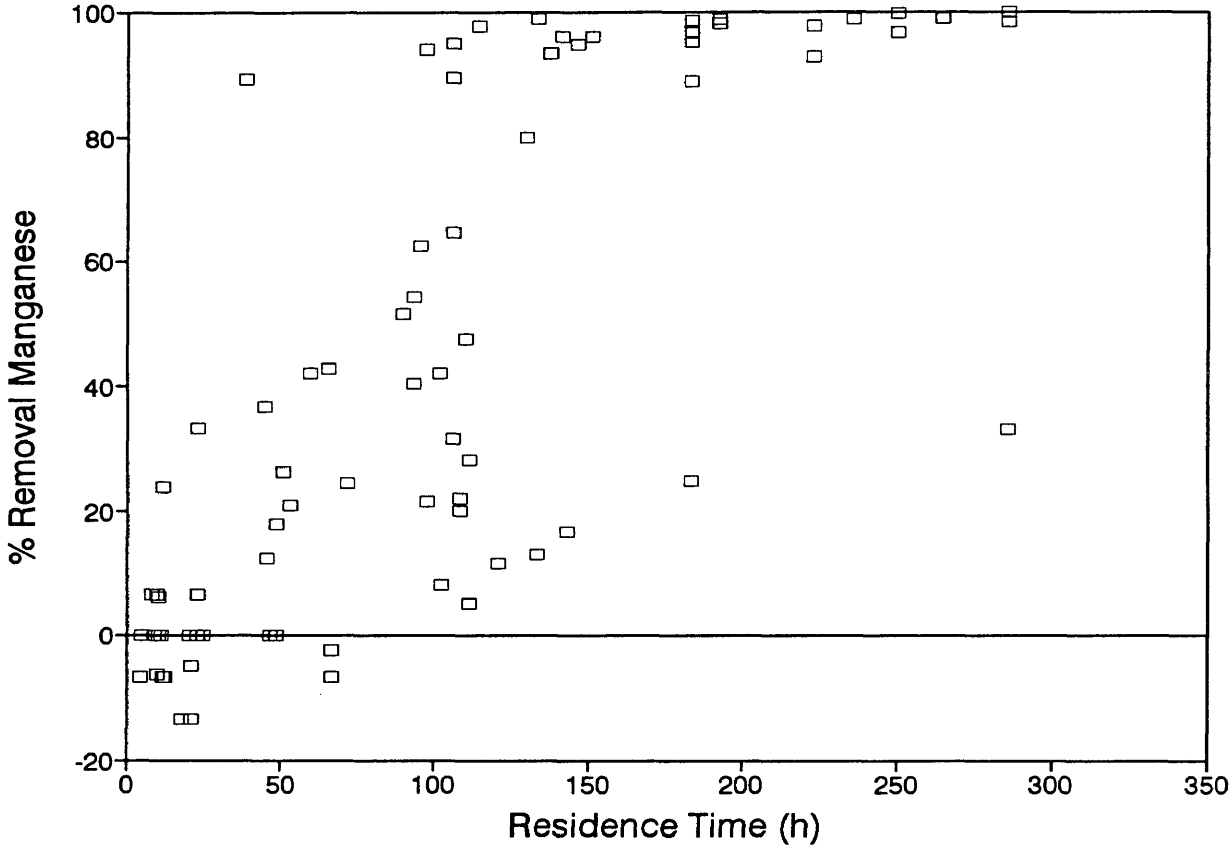

Figure 17. Graph showing composite of data from CSM studies from Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Percentage removal of manganese vs residence time.

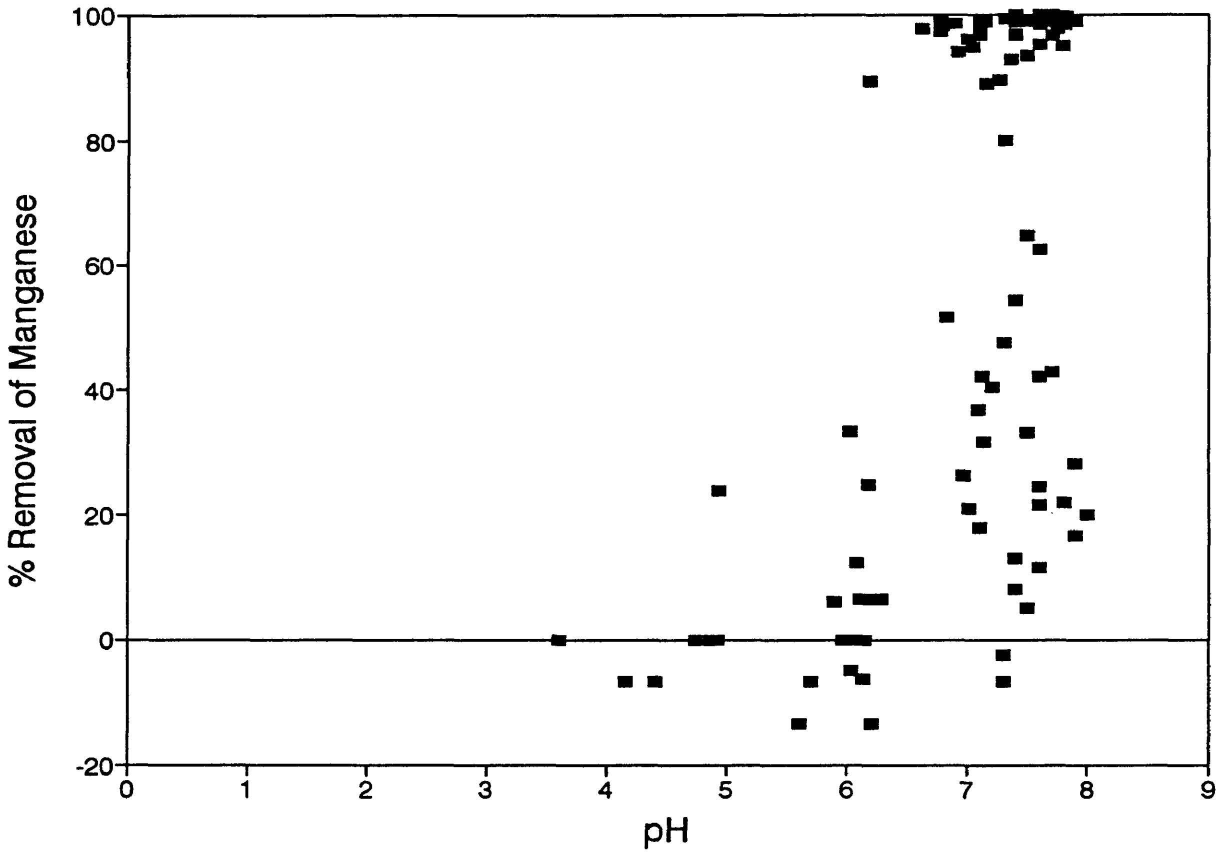

Figure 18. Graph showing composite of data from CSM studies from Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Percentage removal of manganese vs pH.

Figure 19 illustrates the relationship between percent removal of sulfate vs residence time. As the residence time is increased, the percent removal of sulfate increases. This indicates that longer residence times are necessary to produce a significant change in the sulfate concentration. In contrast, Figure 20 illustrates the relationship of mass removal capacity of sulfate vs residence time for the four studies. Sulfate mass removal capacity tends to increase with decreasing residence times.

Given that sulfate reduction increases with decreasing residence time, there is the potential to remove more metals through increased sulfide production. The pH, however, drops below 6.0 and the environmental conditions are not conducive to formation of sulfides of iron, manganese and zinc. One possible solution to this problem, and a means of potentially reducing the residence time for effective treatment, is recycling of the effluent back into the system. Lema et al. (1991) advocate recycling for fixed-bed systems as a means of increasing the upflow liquid velocity. Grady (1980) suggests that increasing the velocity of flow against the biofilm layer reduces the thickness of the layer thereby enhancing contaminant removal capacity of the bacteria. Recycling does require an energy source to overcome the head loss in feeding the effluent back into the system. An advantage of recycling is the increased buffering capacity of the medium and the reduced concentration of the influent water, in the case of high influent concentrations.

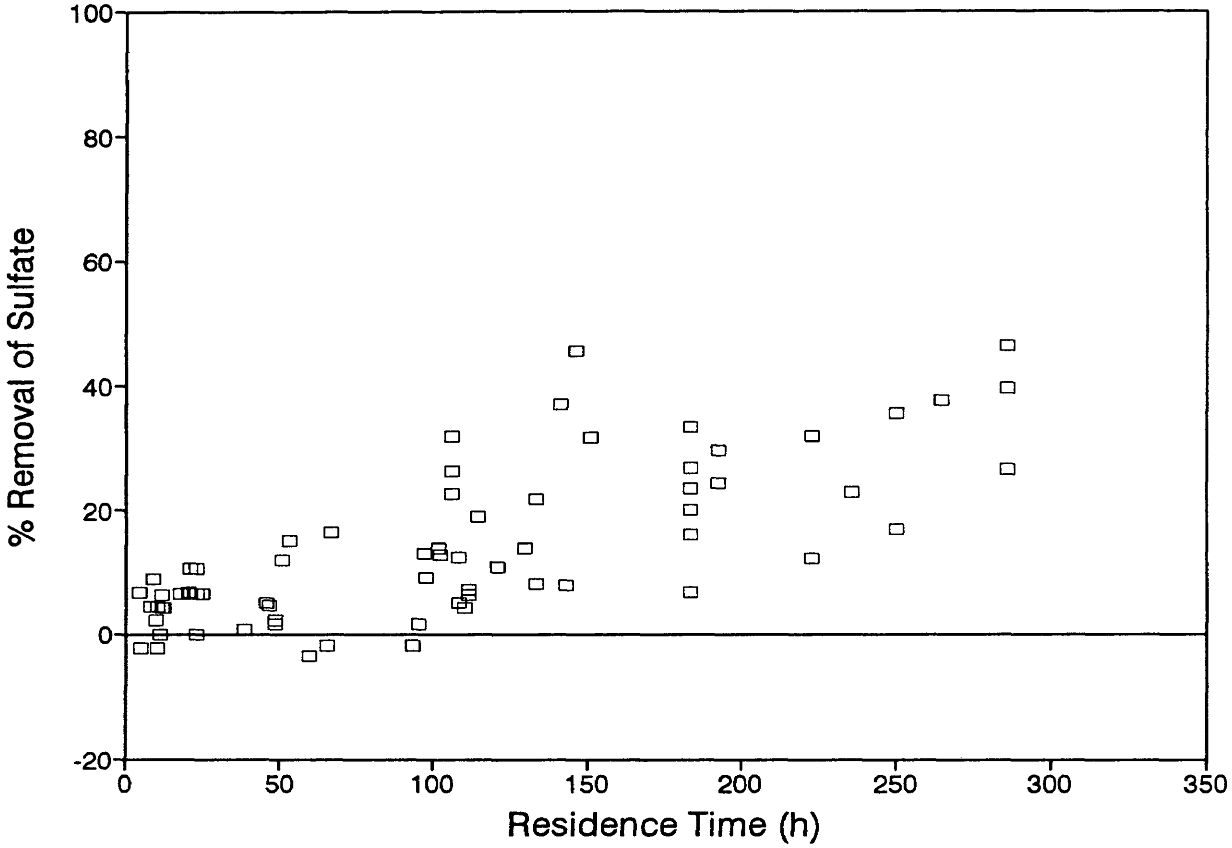

Figure 19. Graph showing composite of data from CSM studies from Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Percent removal of sulfate vs residence time.

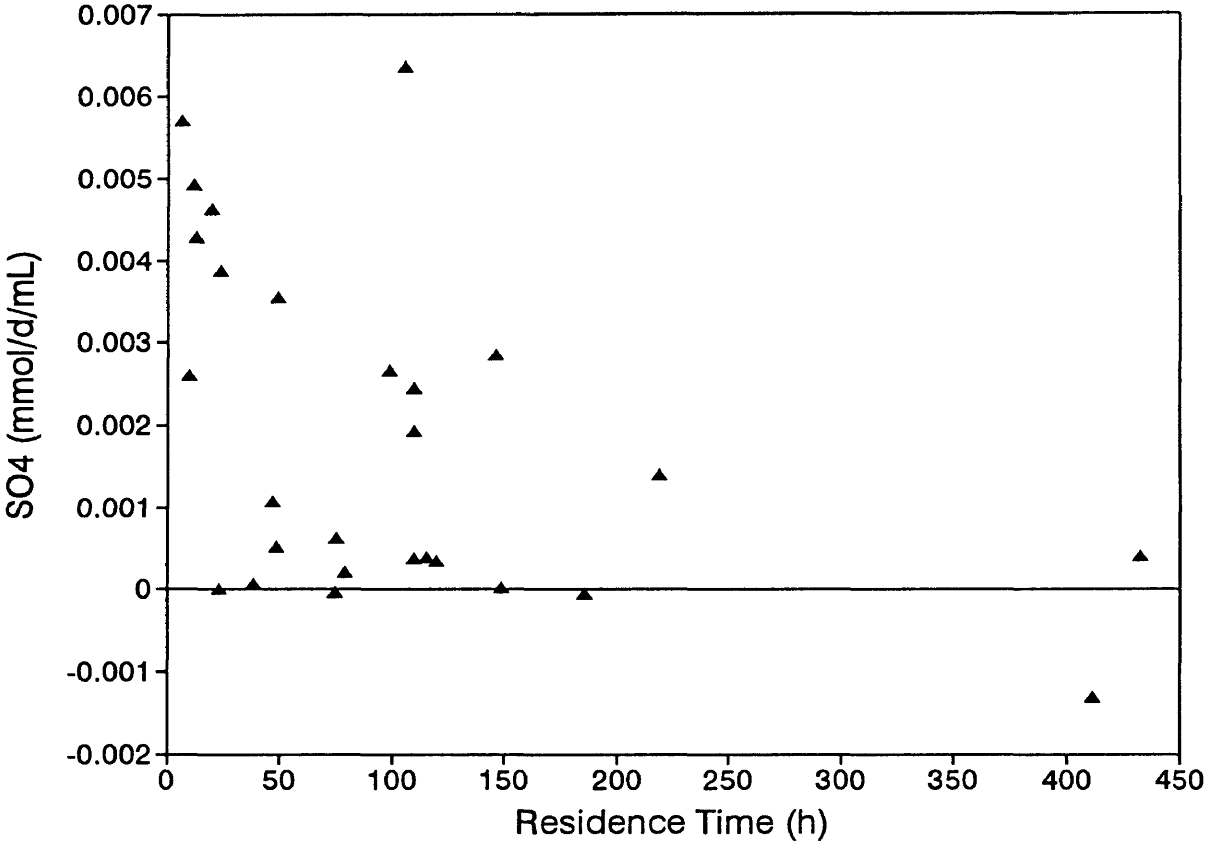

Figure 20. Graph showing composite of data from CSM studies from Bolis (1992); Hurley Euler (1992); Madel and Staub (1992): Sulfate removal capacity (mmol/d/cm^3) vs residence time.

CONCLUSIONS

The main objective of this study was to design an effective treatment of acid mine drainage using bioreactors and sulfate reducing bacteria. Statistical analyses of effluent values from the series system and the parallel system showed that there was no significant difference in treatment between the two systems over the residence times examined. Under the conditions of effective treatment established in Chapter 1, both systems provided effective treatment in series and in parallel at the 100 hour residence time. The pH was increased and all metals were decreased below Eagle Mine discharge permit limits except manganese.

Residence times below 50 hours produced mixed results. Cadmium, copper and lead were removed at all residence times. Effluent manganese concentrations were consistently above discharge permit limits. Below 50 hours of residence time effluent iron and zinc concentrations were above permit limits. Sulfate reduction was low compared to influent concentrations but generally consistent with total amounts of metals removed (in terms of molar concentrations). Eh values were scattered, but in general, Eh was always decreased to at least slightly reducing conditions. Conductivity was scattered so no consistent relationship with residence time was established. At 50 hours and below, pH was increased but not enough to meet permit requirements.

Sulfate and metal removal capacity, in terms of the mass removed per unit volume per day, in general, increased as the residence time was reduced. The removal capacity for the 50 hour residence time was lower than for the 100 hour residence time. This may reflect saturation of the substrate, since this residence time was run as the last experiment. The bacteria may no longer be able to efficiently produce sulfide. Nevertheless, overall increasing sulfate and metal removal capacities indicate that there is potential to treat the mine drainage more efficiently, but effluent concentrations are not within permit limits.

Effluent from the systems examined had increased values of COD and ammonia. Not enough data was collected to draw any conclusions about whether nutrient values diminish with time. A nutrient profile from start up of a PMDTS would be a worthwhile study. Also, further treatment with an aerobic polishing step might alleviate the problem.

It may be possible to control effluent COD by stoichiometrically determining the amount of organic carbon necessary for efficient operation of the bacteria. This would be difficult, given that a mixed consortium of bacteria are used. If a pure culture could be obtained, this amount only could be calculated and fed to the bacteria in a passive system. Such a system would require a bacterial culture and a matrix for bacterial growth.

Another method to reduce nutrients that are released initially in operation, may be substrate washing. If it were determined that nutrient concentrations diminish with time, substrate washing could remove a large amount of nutrients in the initial stages of system operation.

An attempt was made to test low end residence times observed in the laboratory in a field system. The reactor used was acquired from another study. Because the reactor had not been used for six months, it was difficult to restart it. Also, because a valve was used to control flow into the system, it was difficult to maintain a consistent flow rate. Therefore, the field data was not particularly useful for direct comparison with bench scale results. A valveless system is strongly recommended in future efforts. Field data was included in an overall review of three other studies performed by Colorado School of Mines researchers. Results from those studies are fairly consistent with results from the current study. Below a 50 hour residence time, pH is increased, but only to around 6.0. Cadmium, copper and lead are removed almost completely, regardless of residence time and loading rate. Percent removal decreases below a residence time of 50 hours for iron, manganese and zinc. Effluent concentrations of these metals are above permit limits. Manganese removal is scattered, but generally dependent upon pH. Percent removal of sulfate increases with increasing residence time. Eh and conductivity values are scattered and show no clear relationship with residence time. Overall metal and sulfate removal capacities, in terms of the mass of constituent removed per unit volume per day, increases with decreasing residence time.

It is recommended that total metals samples be used for effluent analysis. Since permit limits and standards generally are reported for total metals, it may be worthwhile and cost effective to analyze for total metals in the effluent from this type of a system. The additional cost involved in analyzing for total metals ($10 more at VISTA Laboratories) may be offset by saving time and money on filters, since total metals samples do not have to be filtered. Also, there was not a significant difference found between total metal concentrations and dissolved metal concentrations.

Finally, the conditions established in Chapter 1 for effective treatment are stringent, especially given that the mine drainage treated in this experiment is higher in metals and lower in pH than values typically reported. System design, therefore, should be based on a residence time of 30-100 hours to effectively increase the pH and reduce the metals to acceptable levels. If Fe and Zn are primary metals of concern, a residence time of 30-50 hours should produce acceptable results. If manganese is the primary metal of concern, a residence time of greater than 150 hours is necessary to provide efficient removal. A neutral effluent pH is dependent upon a residence time of approxmiately 50 hours. The actual residence time will be dependent on total metals in the influent. One way to reduce this residence time, as discussed in Chapter 4, may be recycling of effluent back into the reactor, which does, however, require addition of an energy source.

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APPENDIX A

Results from bench scale investigations.

Appendix A. Results of laboratory analyses. Influent mine drainage values, and effluent values from Reactors 1 through 3 are presented. Data for flow rates (mL/min), residence times (h), pH, Eh (mV), conductivity (mmhos/cm) and sulfate and metals concentrations (mg/L) are listed.

Date	Mine Drainage									
	pH	Eh	Cond	Cd	Cu	Fe	Pb	Mn	Zn	SO4
4/8/92	2.87	452.3	6440	0.24	1.5	230	0.13	180	120	4600
4/15/92	2.96	428.7	6560	0.26	1.5	240	0.16	190	120	
4/22/92	2.87	465.5	6300	0.26	1.6	210	0.19	190	130	5000
4/29/92	2.91	451	6450	0.23	1.6	220	0.17	190	130	5300
5/18/92	2.94	417.3	5640	0.26	1.2	140	0.14	150	100	4400
5/26/92	2.94	430.2	5910	0.32	1.2	160	0.17	160	110	4400
6/3/92	3	449.8	5850	0.28	1.3	170	0.15	160	100	4600
7/9/92	3.09	412	5930	0.3	1.7	270	0.12	210	140	4500
7/13/92	2.96	426.5	6000	0.22	1.3	190	0.13	150	98	4700
7/15/92	2.85	432.9	5940	0.23	1.2	190	0.12	150	98	4600
7/28/92	2.79	615.4	5900	0.25	1.2	120	0.17	150	100	4200

Appendix A. continued;

Reactor 1: First stage of series reactors.

Date	pH	Eh	Cond	Cd	Cu	Fe	Pb	Mn	Zn	SO4	Flow rate (mL/min)	Res time (hr)
4/8/92	*	*	*	*	*	*	*	*	*	*	*	*
4/15/92	7.09	-87.3	6570	<	<	3.4	<	120	<		2.6	44.9
4/22/92	6.97	-85.3	6330	<	<	18	<	140	<	4400	2.3	50.8
4/29/92	7.01	-106.6	6040	<	<	10	<	150	<	4500	2.2	53.1
5/18/92	5.71	9	5330	0.007	<	150	<	160	50	4100	26.2	4.5
5/26/92	4.93	167.3	5600	0.01	<	150	<	160	80	4200	11.3	10.3
6/3/92	4.74	157.5	5500	0.013	<	160	<	160	74	4700	22.5	5.2
7/9/92	4.94	197.3	5490	0.013	<	170	<	160	60	4300	10.1	11.6
7/13/92	4.15	286.4	5450	0.053	0.1	130	<	160	74	4400	10.1	11.6
7/15/92	4.4	205.1	5520	0.01	<	190	<	160	65	4400	9.4	12.4
7/28/92	3.6	312.5	5440	<	<	110	<	150	76	4200	5	23.4

Appendix A. continued;

Reactor 2: Second stage of series reactors.

Date	pH	Eh	Cond	Cd	Cu	Fe	Pb	Mn	Zn	SO4	Flow rate (mL/min)	Res time (hr)
4/8/92	*	*	*	*	*	*	*	*	*	*	*	*
4/15/92	6.83	-200.1	7510	<	<	1	<	92	<		2.6	89.8
4/22/92	7.12	-59.3	4190	<	<	0.9	<	110	<	4300	2.3	101.6
4/29/92	7.14	-92.8	6200	<	<	0.7	<	130	<	3900	2.2	106.2
5/18/92	6.15	-16.1	5370	<	<	120	<	150	6.5	4000	26.2	8.9
5/26/92	6.08	-14.1	5590	<	<	130	<	160	7.6	4100	11.3	20.7
6/3/92	5.9	-0.1	5570	<	<	120	<	150	27	4700	22.5	10.4
7/9/92	6.03	-9.8	5480	<	<	68	<	140	0.6	4200	10.1	23.1
7/13/92	6.17	-42.8	5490	<	<	86	<	140	2.8	4200	10.1	23.1
7/15/92	5.97	27.3	5530	<	<	140	<	150	4.5	4300	9.4	24.9
7/28/92	6.07	67.6	5280	<	<	100	<	150	5	4000	5	46.7

Appendix A. continued;

Reactor 3: Single stage.

Date	pH	Eh	Cond	Cd	Cu	Fe	Pb	Mn	Zn	SO4	Flow rate (mL/min)	Res time (hr)
4/8/92	6.92	-336.5	9010	<	<	<	<	11	0.1	4000	1.2	97.3
4/15/92	7.27	-25.4	7170	<	<	<	<	20	<		1.1	106.2
4/22/92	7.32	80.1	6560	<	<	<	<	38	<	4300	0.9	129.8
4/29/92	7.5	89.7	5550	<	<	<	<	67	<	4100	1.1	106.2
5/18/92	6.29	-24.1	6290	<	<	85	<	140	4.4	4200	14.4	8.1
5/26/92	6.14	-16.6	5660	<	<	120	<	170	27	4300	11.8	9.9
6/3/92	6.08	1.3	5530	<	<	130	<	160	32	4600	10.3	11.3
7/9/92	6.04	49.6	5470	<	<	110	<	220	9.8	4200	5.6	20.9
7/13/92	6.21	26.2	5430	<	<	61	<	170	7.6	4200	5.6	20.9
7/15/92	5.6	125.2	5450	<	<	140	<	170	5.5	4300	6.5	18.0
7/28/92	4.87	221.5	5240	<	<	54	<	150	2.9	4100	2.4	48.7

< Constituent not detected at or above the listed reporting limit.

* Bad data-excluded from results.

Reporting Limits:

Cd	0.0005 - 0.03 mg/L	Fe	0.1 - 0.5 mg/L
Cu	0.02 - 0.1 mg/L	Mn	0.01 - 0.05 mg/L
Pb	0.003 - 0.03 mg/L	Zn	0.02 - 0.1 mg/L

APPENDIX B

Results of twosample-t tests performed on bench scale results.

Appendix B. Results of twosample-t tests performed on laboratory results. Data from Reactor 2 (second stage of series reactors) and Reactor 3 (single stage reactor) are presented. Data from both reactors are averaged over the 104 hour residence time and the 11 hour residence time.

t-tests between the series system and the single stage system

For all tests:

H_0: $u_1 = u_2$

H_1: $u_1 \neq u_2$

alpha = 0.05

Reject H_0 if $t_{calc} > t_{alpha/2}$ or if $t_{calc} < -t_{alpha/2}$

Calculation:

$$t = \frac{(x_1 - x_2)}{(s_1^2/n_1 + s_2^2/n_2)}$$

, calculated by MINITAB

Decision: see individual results.

Appendix B. continued;

parameter	Series system mean	Single stage mean	t	alpha /2	decision
HRT-1	99.2±8.5	109.9±14	-1.25	2.776	do not reject
HRT-2	13.3±6.4	9.8±1.6	0.93	4.303	"
HRT-3	23.7±1.04	19.9±1.7	3.31	3.182	reject H_0
pH-1	7.25±0.24	7.03±0.17	1.41	2.776	do not reject
pH-2	6.17±0.11	6.04±0.13	1.30	3.182	"
pH-3	5.95±0.32	6.06±0.10	-0.56	4.303	"
Fe-1	0.0±0.0	0.87±0.15	----	----	"
Fe-2	112±24	123±6	-0.83	4.303	"
Fe-3	104±40	98±37	0.18	3.182	"
Zn-1	0.02±0.05	0.0±0.0	----	----	"
Zn-2	21.1±14.7	13.7±11.5	0.69	3.182	"
Zn-3	7.63±2.15	2.63±1.96	2.98	3.182	"

HRT = Hydraulic Residence Time (h)
Fe concentration (mg/L)
Zn concentration (mg/L)

Appendix B. continued;

parameter	HRT = 100	HRT = 10	t	alpha /2	decision
HRT	104.3±7.8	11.57±3.1	30.05	2.262	reject H_0
pH	7.16±0.23	6.11±0.13	10.32	2.262	reject H_0
Fe	0.53±0.62	117.5±16.7	-17.19	2.571	reject H_0
Zn	0.01±0.03	17.4±12.5	-3.41	2.571	reject H_0

HRT = Hydraulic Residence Time (h)
Fe concentration (mg/L)
Zn concentration (mg/L)

APPENDIX C

Results from Eagle Mine Field Work

Appendix C. Results from Eagle Mine field work.

	pH		Eh (mV)		Cond (mmhos)	
Date	influent	Reactor1	influent	Reactor1	influent	Reactor1
6/15/92	3.05	7.16	505.3	-78.3	4720	5710
6/25/92	3	6.18	523.5	-84.3	5070	5230
7/1/92	2.98	6.08	515.5	-25.2	5050	5000
7/7/92	3.03	6.12	534.8	-31.8	5030	5010

	Cadmium		Copper		Iron	
6/15/92	0.68	0.005	11	0.08	220	1.4
6/25/92	0.58	<	9.5	<	210	46
7/1/92	0.8	<	10	<	230	61
7/7/92	0.7	<	9.5	<	210	65

	Lead		Manganese		Zinc	
6/15/92	0.61	<	170	19	200	0.5
6/25/92	0.48	<	160	120	210	6.8
7/1/92	0.6	<	160	140	190	54
7/7/92	0.51	<	150	140	180	36

	Sulfate		Flow Rate (mL/min)	Res Time (h)
6/15/92	4200	2800	50	183.3
6/25/92	4700	3600	50	183.3
7/1/92	3800	3600	200	45.8
7/7/92	3700	9400	915	10.0

APPENDIX D

Results of Quality Analysis /Quality Control of Sampling

Appendix D. Results of quality analysis and quality control samples. Results for split samples, duplicate samples and blank samples are listed. All concentrations are in mg/L.

BLANKS

date	SO_4	Cd	Cu	Fe	Pb	Mn	Zn
4/8/92	*	*	*	*	*	*	*
4/15/92	19	<	<	<	<	0.19	<
4/22/92	*	<	<	<	<	0.06	0.03
4/28/92	<	<	<	<	<	0.10	<
5/18/92	<	<	<	<	<	<	<
5/26/92	<	<	<	<	<	0.03	<
6/3/92	<	<	<	<	<	<	<
6/15/92	<	<	<	<	<	0.01	0.02
6/25/92	<	<	<	<	<	0.02	<
7/1/92	<	<	<	<	<	<	<
7/7/92	9500	<	<	<	<	<	<
7/9/92	*	*	*	*	*	*	*
7/13/92	*	*	*	*	*	*	*
7/15/92	<	<	<	<	<	<	<
7/28/92	*	*	*	*	*	*	*

* = no sample taken
< = not detected at or above the listed reporting limits

Appendix D. continued;

SPLITS

date	SO_4	Cd	Cu	Fe	Pb	Mn	Zn
4/8/92	*	*	*	*	*	*	*
4/15/92	**	0.28	1.5	240	0.15	190	120
4/22/92	*	*	*	*	*	*	*
4/28/92	5000	0.23	1.6	210	0.17	190	130
5/18/92	4400	0.28	1.2	140	0.14	150	100
5/26/92	*	*	*	*	*	*	*
6/3/92	4300	0.30	1.3	170	0.17	160	100
6/15/92	4200	0.69	12	240	0.61	180	210
6/25/92	*	*	*	*	*	*	*
7/1/92	3800	0.8	10	230	0.6	160	190
7/7/92	*	*	*	*	*	*	*
7/9/92	4500	0.3	1.3	210	0.15	160	100
7/13/92	*	*	*	*	*	*	*
7/15/92	4600	0.23	1.3	190	0.12	150	100
7/28/92	*	*	*	*	*	*	*

* = no sample taken
** = bad data
< = not detected at or above the listed reporting limits

Appendix D. continued;

DUPLICATES

date	SO_4	Cd	Cu	Fe	Pb	Mn	Zn
4/8/92	*	*	*	*	*	*	*
4/15/92	*	*	*	*	*	*	*
4/22/92	5000	0.26	1.6	210	0.19	190	130
4/28/92	*	*	*	*	*	*	*
5/18/92	*	*	*	*	*	*	*
5/26/92	4400	0.31	1.2	160	0.17	160	110
6/3/92	*	*	*	*	*	*	*
6/15/92	*	*	*	*	*	*	*
6/25/92	4200	0.59	9.8	220	0.49	160	210
7/1/92	*	*	*	*	*	*	*
7/7/92	3700	0.7	9.4	200	0.52	150	180
7/9/92	*	*	*	*	*	*	*
7/13/92	*	*	*	*	*	*	*
7/15/92	*	*	*	*	*	*	*
7/28/92	*	*	*	*	*	*	*

* = no sample taken
< = not detected at or above the listed reporting limits

APPENDIX E

Results From Previous Colorado School Of Mines Studies

Appendix E. Results obtained from previous CSM studies (Bolis 1992; Hurley Euler 1992; Staub 1992).

	pH		Eh		Conductivity		Cadmium		Copper		Iron		Lead		Manganese		Zinc		Sulfate		Flow Rate	Res. Time
	influent	effluent	influent	effluent	influent	effluent	influent	effluent	influent	effluent	influent	effluent	influent	effluent	influent	effluent	influent	effluent	influent	effluent	ml/min	hours
M	2.94	5.71	417.3	9	5840	5330	0.26	0.007	1.2	<	140	150	0.14	<	150	180	100	50	4400	4100	26.2	4.3
M	3	4.74	440.8	157.5	5850	5000	0.28	0.013	1.3	<	170	160	0.15	<	180	180	100	74	4800	4700	22.5	5.2
M	2.94	4.93	430.2	167.3	5910	5800	0.32	0.01	1.2	<	160	150	0.17	<	180	180	110	80	4400	4200	11.3	10.3
M	3.08	4.84	412	197.3	5830	5480	0.3	0.013	1.7	<	270	170	0.12	<	210	180	140	80	4500	4300	10.1	11.6
M	2.96	4.15	426.5	208.4	6000	5430	0.22	0.033	1.3	0.1	190	130	0.13	<	150	180	98	74	4700	4400	10.1	11.6
M	2.85	4.4	432.9	205.1	5840	5020	0.23	0.01	1.2	<	160	180	0.12	<	150	160	98	65	4800	4400	9.4	12.4
M	2.79	3.6	613.4	312.5	5800	5440	0.25	<	1.2	<	120	110	0.17	<	130	130	100	78	4200	4200	5	23.4
M	2.96	7.09	426.7	-87.3	6580	6570	0.26	<	1.5	<	240	3.4	0.16	<	180	120	120	<			2.6	44.9
M	2.87	6.97	463.5	-85.3	6300	6330	0.26	<	1.6	<	210	18	0.19	<	190	140	130	<	5000	4400	2.3	50.6
M	2.91	7.01	451	-108.6	6450	6040	0.23	<	1.8	<	220	10	0.17	<	180	150	130	<	5300	4500	2.2	53.1
M	2.94	6.15	417.3	-18.1	5840	5370	0.26	<	1.2	<	140	120	0.14	<	150	150	100	6.5	4400	4000	26.2	8.9
M	3	5.9	440.8	-0.1	5850	5570	0.28	<	1.3	<	170	120	0.15	<	180	150	100	27	4800	4700	22.5	10.4
M	2.94	6.08	430.2	-14.1	5910	5580	0.32	<	1.2	<	160	130	0.17	<	180	180	110	7.6	4400	4100	11.3	20.7
M	3.08	6.03	412	-9.8	5830	5480	0.3	<	1.7	<	270	88	0.12	<	210	140	140	0.6	4500	4200	10.1	23.1
M	2.96	6.17	426.5	-42.6	6000	5490	0.22	<	1.3	<	190	88	0.13	<	150	140	98	2.8	4700	4200	10.1	23.1
M	2.85	5.97	432.9	27.3	5840	5030	0.23	<	1.2	<	160	140	0.12	<	150	130	98	4.5	4800	4300	9.4	24.9
M	2.79	6.87	613.4	67.6	5800	5280	0.25	<	1.2	<	120	100	0.17	<	130	130	100	5	4200	4000	5	46.7
M	2.96	6.83	426.7	-200.1	6580	7510	0.26	<	1.5	<	240	1	0.16	<	180	92	120	<			2.6	88.8
M	2.87	7.12	463.5	-30.3	6300	4190	0.26	<	1.6	<	210	0.9	0.19	<	180	110	130	<	5000	4300	2.3	101.6
M	2.91	7.14	451	-92.8	6450	6200	0.23	<	1.6	<	220	0.7	0.17	<	180	130	130	<	5300	3800	2.2	108.2
M	2.94	6.29	417.3	-24.1	5840	6280	0.26	<	1.2	<	140	85	0.14	<	150	140	100	4.4	4400	4200	14.4	8.1
M	2.94	6.14	430.2	-16.6	5910	5680	0.28	<	1.2	<	160	120	0.17	<	180	170	110	27	4400	4300	11.8	9.9
M	3	6.08	440.8	1.3	5850	5530	0.32	<	1.3	<	170	130	0.15	<	180	180	100	32	4800	4800	10.3	11.3
M	2.85	5.6	432.9	123.2	5840	5430	0.3	<	1.2	<	180	140	0.12	<	150	170	98	3.5	4800	4300	6.3	18.0
M	3.08	6.04	412	40.6	5830	5470	0.22	<	1.7	<	270	110	0.12	<	210	220	140	8.8	4500	4200	5.6	20.9
M	2.96	6.21	426.5	26.2	6000	5430	0.23	<	1.3	<	190	61	0.13	<	150	170	98	7.6	4700	4200	5.6	20.8
M	2.79	4.87	613.4	221.5	5800	5240	0.25	<	1.2	<	120	54	0.17	<	130	150	100	2.9	4200	4100	2.4	48.7
M	2.87	6.82	452.3	-336.5	6440	8010	0.24	<	1.5	<	230	<	0.13	<	180	11	120	0.1	4800	4000	1.2	97.3
M	2.91	7.5	451	88.7	6450	5550	0.26	<	1.6	<	220	<	0.17	<	190	67	130	<	5300	4100	1.1	108.2
M	2.96	7.27	426.7	-25.4	6580	7170	0.23	<	1.5	<	240	<	0.16	<	180	20	120	<			1.1	108.2
M	2.87	7.32	463.5	80.1	6300	6580	0.26	<	1.6	<	210	<	0.19	<	190	38	130	<	5000	4300	0.8	128.6
M	3.05	7.16	505.3	-78.3	4720	5710	0.88	0.005	11	0.08	220	1.4	0.61	<	170	19	200	0.5	4200	2600	30	183.3
M	3	6.18	523.5	-84.3	5070	5230	0.58	<	9.5	<	210	46	0.48	<	180	120	210	6.8	4700	3800	30	183.3
M	2.98	6.08	515.5	-25.2	5050	5000	0.8	<	10	<	230	61	0.6	<	180	140	190	34	3800	3800	200	45.8
M	3.03	6.12	534.8	-31.8	5030	5010	0.7	<	9.5	<	210	65	0.51	<	150	140	180	38	3700		915	10.0
S	3	7.63	738.2	54.6	3250	20800	0.91	<	15	0.6	270	7	0.51	0.13	230	0.9	180	1.1	4280	8500	30	423.3
S	3.01	7.81	736.5	165.8		20500	0.84	<	13	0.6	230	6	0.32	0.12	200	0.8	190	1.1	4240	6780	48	441.0
S	2.87	7.32	755.9	2.8	5070	17150	0.9	0.03	13	0.9	270	9	0.34	0.18	180	1.3	200	1	4290	7050	48	441.0
S	2.82	6.79	735.7	-20.5	3780	13160	0.8	<	14	0.6	280	14	0.47	<	170	3.4	200	1.1	3500	8140	53	398.4
S	2.85	6.77	688.1	-13.6	4580	11480	0.9	0.02	15	0.5	310	19	0.75	0.12	200	5.7	240	1.4	5400	6040	60	332.8
S	2.88	6.63	683.6	-13.8	4370	10020	0.5	0.01	15	0.2	330	15	0.84	0.03	210	4.8	370	0.8	4700	4120	95	222.8
S	2.81	7.1	720.1	-83.1	3220	8550	1	<	14	<	280	5	0.6	<	200	3.7	310	1.2	3020	3800	110	182.4
S	2.93	7.15	694.4	-76	3790	8840	1.1	<	13	<	310	3	0.46	0.03	180	2.6	280	1.1	4400	3100	110	182.4
S	2.92	6.77	698.2	-27	3770	8880	1	<	13	<	290	1	0.61	<	180	2	280	0.2	4000	2500	80	264.6
S	2.94	7.36	680	-76.3	3800	7730	0.9	<	15	<	330	4	0.6	<	210	15	290	2.2	4400	3000	95	222.8
S	2.79	7	656.3	-87.2			0.9	<	14	0.1	300	2.8	0.8	<	190	7.4	240	0.38	4800	2800	150	141.1

Appendix E. continued;

S	3	7	657		2600		0.8	<	14	0.1	300	6	0.8	<	200	6	230	2.5	4100	2600	140	151.2
S	3.01	7.05	670.8	161.1			0.8	<	13	0.07	300	5.3	0.8	<	180	10	230	0.45	4400	2400	145	146.0
S	3.08	7.79	304	132.9	2300	4000	0.7	0.02	13	0.71	310	26	0.38	<	200	8.8	240	4.3	4400	3000	200	105.8
S	2.91	7.75	571.3	81.2	2700	4820	0.8	0.01	14	0.14	280	3	0.8	<	180	4.4	220	1.3	3984	3210	185	114.4
B		7.5	307	122			*	*	0.2	0.05	41.9	7.4	*	*	22.8	1.5	9.1	0.5	722	1317	12	137.5
B		7.4	382	315			*	*	0.1	0.08	42.3	5.5	*	*	23.1	0.3	7.9	0.3	880	884	7	230.7
B		6.9	430	380			*	*	0.1	0.3	54.8	2.9	*	*	24.8	0.4	8.5	0.1	1027	732	6	163.3
B		6.2	413	30			*	*	0.09	0.03	49.2	2.9	*	*	24.4	2.6	8	0.4	1010	1001	43	38.4
B	2.6	7.5	700	330			*	*	63.5	0.08	800.5	3	*	*	62.1	0.8	467.9	0.5	3820	732	0.4	4125.0
B	2.5	7.7	730	400			*	*	57.1	0.4	682	3	*	*	76.8	0.9	130.1	0.5	3080	1777	0.8	1833.3
B	2.5	7.7	720	330			*	*	34.4	0.04	702.9	1.8	*	*	70.7	0.6	177.4	0.4	4027	2207	0.7	2357.1
B	2.3	7.1	740	360			*	*	50	0.28	633.8	1.6	*	*	85.8	1.2	180	0.1	3438	3430	1.2	1375.0
B	3.1	7.8	630	220			*	*	2.04	0.08	51.6	4.91	*	*	37.3	1.8	13	0.3	1830	1800	8	183.3
B	3.1	7.4	630	110			*	*	2.05	0.08	42.5	3.45	*	*	35.3	1.2	12.1	0.2	1840	980	1	1650.0
B	3.2	7.1	630	110			*	*	2.17	0.07	40	0.74	*	*	35.3	1.2	12.3	0.1	1800	1350	9	183.3
B	3.1	7.4	630	-10			*	*	2.28	0.04	37.5	0.71	*	*	35.2	1.1	12.4	0.1	1850	1480	8	183.3
B	3	7.3	880	40			*	*	4.29	0.03	38.8	1.1	*	*	35.5	18.6	3.6	0.1	1820	1740	15	110.0
B	3.1	7.1	840	-80			*	*	4.43	0.06	40.2	2.7	*	*	35.6	29.2	13.9	0.1	1830	1820	34	48.5
E	3	7.6	320	236.2	2140	2640	*	*	1.88	<	19.8	0.314	*	*	33.4	0.082	11.7	0.017		1375	0.3	1333.3
E	3	7.7	508	298.8			*	*	2.14	<	18.3	0.108	*	*	34.3	0.082	12.2	0.013		1222	0.9	444.4
E	3	7.7	483.8	-298.1			*	*	2.11	<	21	0.008	*	*	34.8	0.038	12.2	<	1840	1111	1.4	285.7
E	3	7.6	471.4	-316			*	*	2.88	<	23.7	0.023	*	*	37.5	0.027	13.4	0.003	1840	1350	1.4	285.7
E	2.8	7.7	480.5	-23.8			*	*	3.1	0.004	16.8	0.1	*	*	38.6	0.128	13.2	0.014	1801	1616	1.6	250.0
E	3.2	7.2	417.4	351.8	2470	2450	*	*	2.885	<	29.4	0.089	*	*	30.65	18.25	10.9	0.01	1858		4.3	93.0
E	3.1	7.3	389.1	-180.4	2380	2440	*	*	2.8	<	21.2	0.073	*	*	31.35	32.1	11.05	0.014	1782	1466	6	66.7
E	3.1	7.3	408.3	-75.5	2840	2430	*	*	2.845	0.007	19.35	0.385	*	*	30.3	32.3	10.8	0.036		1767	6	66.7
E	3.2	7.3	430.8	-132.1	2380	2380	*	*	2.76	0.007	22.05	0.066	*	*	32.25	30.8	11.25	0.028	1884	1731	3.6	111.1
E	3.2	7.4	412.5	-100.5	2830	2380	*	*	2.735	0.01	18	0.187	*	*	32	29.35	11.1	0.038	1844	1805	3.9	102.6
E	3.2	7.4	428.8	-143.5	2820	2440	*	*	2.67	0.013	16.85	0.203	*	*	31.75	27.8	11.2	0.067	1844	1882	3	133.3
E	3	7.8	320	254.7	2140	3820	*	*	1.88	0.0116	19.8	0.1380	*	*	33.4	0.072	11.7	0.0281		1375	0.9	444.4
E	3	7.7	508	-322.5			*	*	2.14	<	18.3	0.0887	*	*	34.5	0.104	12.2	0.034		1317	0.9	444.4
E	3	7.8	483.8	-325.3			*	*	2.11	<	21	0.0282	*	*	34.8	0.1475	12.2	0.003	1840	1157	0.7	571.4
E	3	7.7	471.4	-288.1			*	*	2.88	<	23.7	0.0228	*	*	37.5	0.403	13.4	0.012	1840	1438	3	133.3
E	2.8	7.8	480.5	-281.5			*	*	3.1	<	16.8	0.0024	*	*	38.6	0.423	13.2	<	1801	1302	0.7	571.4
E	3.2	7.6	417.4	301.3	2470	2380	*	*	2.885	<	29.4	0.0081	*	*	30.65	11.35	10.9	0.0038	1858	1823	4.2	95.2
E	3.1	7.7	389.1	-188.6	2380	2310	*	*	2.8	<	21.2	<	*	*	31.35	17.8	11.05	0.0038	1782	1813	6.1	65.6
E	3.1	7.5	408.3	-162.2	2840	2320	*	*	2.845	<	19.35	0.0033	*	*	30.3	20.25	10.8	0.0076		1807	1.4	285.7
E	3.2	7.9	430.8	-208.9	2380	2420	*	*	2.76	0.0002	22.05	0.0215	*	*	32.25	23.15	11.25	0.0186	1884	1745	3.6	111.1
E	3.2	7.6	412.5	-184.6	2830	2460	*	*	2.735	0.003	18	0.3215	*	*	32	25.1	11.1	0.0263	1844	1675	4.1	97.6
E	3.2	7.6	428.8	-158.9	2820	2430	*	*	2.67	0.0029	16.85	0.0035	*	*	31.75	28.05	11.2	0.0089	1844	1842	3.3	121.2
E	3	7.8	320	244.8	2140	2450	*	*	1.88	<	19.8	0.0041	*	*	33.4	0.1625	11.7	<		1385	0.4	1000.0
E	3	7.8	483.8	-382.2			*	*	2.11	<	21	<	*	*	34.8	0.38	12.2	0.0083	1840	738	0.5	800.0
E	3	7.8	471.4	-334.1			*	*	2.88	<	23.7	0.0088	*	*	37.5	0.6085	13.4	0.0077	1840	888	1.4	285.7
E	2.8	7.7	480.5	-304.8			*	*	3.1	<	16.8	0.0047	*	*	38.6	1.28	13.2	0.0081	1801	1238	1.6	250.0
E	3.2	7.4	417.4	308.1	2470	2480	*	*	2.885	<	29.4	0.0435	*	*	30.65	14	10.9	<	1858	1888	4.3	93.0
E	3.1	7.6	389.1	-174.4	2380	2380	*	*	2.8	<	21.2	<	*	*	31.35	18.15	11.05	<	1782	1844	6.7	59.7
E	3.1	7.6	408.3	-119.3	2840	2480	*	*	2.845	<	19.35	0.0054	*	*	30.3	22.8	10.8	0.0024		1768	5.6	71.4
E	3.2	8	430.8	-217.9	2380	2380	*	*	2.76	<	22.05	<	*	*	32.25	23.8	11.25	0.0025	1884	1768	3.7	108.1
E	3.2	7.8	412.5	-120.4	2830	2420	*	*	2.735	<	18	<	*	*	32	24.85	11.1	0.0071	1844	1613	3.7	108.1
E	3.2	7.9	428.8	-135.9	2820	2430	*	*	2.67	<	16.85	<	*	*	31.75	26.4	11.2	0.0235	1844	1666	2.8	142.9

B = Bolis (1992)
E = Hurley Euler (1992)
M = Madel (this study)
S = Staub (1992)

* = no data
** = bad data
< = value below detection limits