Mine Water Environ (2015) 34:442–454
DOI 10.1007/s10230-014-0289-1

TECHNICAL ARTICLE

The Use of Mussel Shells in Upward-Flow Sulfate-Reducing Bioreactors Treating Acid Mine Drainage

Benjamin Uster · Aisling D. O'Sullivan ·
Su Young Ko · Alex Evans · James Pope ·
Dave Trumm · Brian Caruso

Received: 23 January 2014 / Accepted: 14 July 2014 / Published online: 8 November 2014

Abstract In this study, sulfate-reducing bioreactors (SRBRs) efficiently treated acid mine drainage (AMD) for a contiguous period of 5 months. The AMD was sourced from an active coal mine on the South Island of New Zealand and typically had a $pH < 3$, 1,700 mg/L of sulfate, 50 mg/L of Fe, 18 mg/L of Al, 15 mg/L of Mn, 4 mg/L of Zn, and lower concentrations of other contaminants. Two alkalinity-generating materials (mussel shells and limestone) and two hydraulic retention times (HRTs) of 3 and 10 days were evaluated. Influent and effluent water quality parameters were monitored weekly. Each SRBR system successfully increased the pH ($\geq$6) and the alkalinity ($\leq$350 mg/L $CaCO_3$) of the water while removing substantial amounts of dissolved metals at both HRTs ($\geq$90 % Al, $\geq$86 % Fe, $\geq$87 % Cu, $\geq$99 % Zn). Mn removal was lower and ranged from 19 to 55 %. Increasing the HRT from 3 to 10 days significantly improved effluent water quality in terms of pH, alkalinity, and metals and sulfate removal. SRBRs using mussel shells in their reactive mixtures were more effective than those using limestone, with a higher (60–113 %) alkalinity generation and a better (3–5 %) metal removal. This study showed that mussel shells are an inexpensive and sustainable alternative to mined limestone for AMD passive treatment, and that better treatment efficiency resulted from a longer HRT.

Keywords Mine water · Passive treatment · Organic substrate · Alkalinity generation · Metal removal rates

Introduction

Acid mine drainage (AMD) is a multi-factor source of pollution that typically exhibits low pH and contains high concentrations of metals and sulfate (Blowes et al. 2003; Nordstrom and Alpers 1999; Pope et al. 2010; Younger et al. 2002). It can be severely detrimental to aquatic life and usually needs to be treated before being discharged into the environment (Byrne et al. 2012; Salomons 1994; Younger 2004). Numerous treatment methods based on mechanical and chemical processes or relying on natural (bio)geochemical processes have been developed (Hedin et al. 1994; Johnson and Hallberg 2005; Lens et al. 1998; Skousen et al. 2000; Watzlaf et al. 2004; Wildeman and Schmiermund 2004; Younger et al. 2002). Sulfate-reducing bioreactors (SRBRs) are a promising technique that has gained prevalence in the past few years because they concomitantly remove the key contaminants of acidity, metals, and sulfate from AMD (Garcia et al. 2001; Johnson and Hallberg 2005; Zagury et al. 2005). Other advantages of this treatment approach include relatively low maintenance costs, minimal energy requirements, and the possibility of using natural organic wastes in the reactive mixture (Rose 2010; URS 2003; Wildeman et al. 2006). SRBR relies on the principle of sulfidogenesis, during which sulfate-reducing bacteria (SRB) oxidize a carbon source (i.e. the organic substrate) and reduce sulfate to sulfide through a dissimilatory process (Eq. 1), where CH_2O represent a simple carbon/electron source (Hao et al. 1996; Widdel 1988). Typically, a substrate comprises a mixture of organic and alkaline materials.

B. Uster (✉) · A. D. O'Sullivan · S. Y. Ko · A. Evans · B. Caruso
Department of Civil and Natural Resources Engineering, University of Canterbury, Private Bag 4800, Christchurch 8140, New Zealand
e-mail: benjamin.uster@pg.canterbury.ac.nz

J. Pope · D. Trumm
CRL Energy Ltd, 97 Nazareth Ave, PO Box 29-415, Christchurch 8540, New Zealand

$$2CH_2O + SO_4^{2-} \rightarrow 2HCO_3^- + H_2S \quad (1)$$

In order for the SRB to effectively conduct sulfidogenesis, a pH above 5.5 and a reducing environment (≤100 mV) are required (Postgate 1979; Tang et al. 2009). Since AMD is typically acidic, it is essential to mitigate this acidity to optimize SRB activity. Because limestone is relatively inexpensive, it has been commonly used as the alkalinity generating material in AMD passive treatment (Nairn and Mercer 2000; Thomas and Romanek 2002b). In SRBRs, limestone is typically incorporated in the substrate mixture along with the organic materials to mitigate acidity through calcium carbonate dissolution. The recommended amount of limestone added in the reactive substrate is about 30 vol% (Wildeman et al. 2006).

Following acidity mitigation, highly reactive hydrogen sulfide produced during sulfidogenesis (Eq. 1) reacts with dissolved metals present in AMD to form various insoluble metal-sulfide precipitates (Stumm and Morgan 1996). Due to the complex biogeochemical interactions occurring in SRBRs, there is no consensus on the best substrate to use, but several studies have found that AMD treatment is more effective when a combination of easily biodegradable substances and more recalcitrant ones are used together (Gibert et al. 2004). Along with the biogeochemical properties of a substrate, its physical characteristics are essential to achieve an efficient long-term treatment. The substrate porosity as well as the hydraulic conductivity, which both influence the hydraulic retention time (HRT), are of particular importance in optimizing AMD treatment in engineered bioreactors (Benner et al. 2002; McCauley 2011; Neculita et al. 2007). If the HRT is too short, there will be insufficient reaction time to adequately treat the AMD. Conversely, if the HRT is too long, depletion of organic matter will rapidly occur, halting long-term treatment (Dvorak et al. 1992; Gusek 2005). Typically, an HRT of at least 40 h is required for effective treatment in full-scale SRBRs, while an HRT of approximately 4 days or more is necessary for metal sulfide precipitation to occur (Neculita et al. 2008; URS 2003). According to Younger et al. (2002), HRT optimization is one of the most important design parameters, but is also the most difficult to achieve. As the performance of the microbiological processes is critical to SRBR treatment success, the physical and chemical composition of a substrate must provide conditions conducive for the microbial community to operate over a long period of time.

Although the designs of SRBRs and their short-term efficiencies are now relatively well understood, their long-term effectiveness is not, due to the inherently complex biogeochemical processes. Carbon exhaustion and hydraulic malfunctions, such as clogging and preferential flow-paths development, are among the most frequent reasons reported for system failure (Gusek 2005). Exact metal removal mechanisms, and especially the factors affecting them, still need to be investigated in detail.

Even though several authors have successfully employed alternative alkaline materials, such as seafood wastes, steel slag, or fly ash, their use in SRBRs have received little attention overall (Batty and Younger 2004; Gitari et al. 2008; McCauley et al. 2009; Robinson-Lora and Brennan 2010; Simmons et al. 2001). Moreover, despite the fact that seafood wastes are a ubiquitous biogenic calcium carbonate source available in many countries around the world, few studies have examined their use in lab- and field-scale passive treatment systems (Crombie et al. 2011; Kohler et al. 2007; McCauley et al. 2009; Robinson-Lora and Brennan 2009; Trumm and Ball 2014). In New Zealand, where waste mussel shells (i.e. free of flesh shells) are produced in abundance by the seafood industry and are mostly disposed of in landfills, their reuse in passive treatment systems like SRBRs could potentially be an economical and sustainable alternative reuse opportunity.

Previous studies conducted with waste mussel shells either used a substrate comprised of mussel shells only (no organic materials) or were operated as downward flow systems. The present study assessed the performance of four different upward-flow SRBR systems using a mixture of organic waste materials and either mussel shells or limestone as a neutralizing material at two different HRTs.

Materials and Methods

Experimental Design and Operation

Four SRBR treatment designs were operated in duplicate. The influence of two different alkalinity sources (mussel shells and limestone) and two different hydraulic retention times (3 and 10 days) was evaluated over a 20 week period (Fig. 1). The duration of the experiments was based on the need to study the SRBRs operating at steady-state over a minimum period that was long enough to observe clear performance trends.

All reactors were 45 cm high and 31 cm wide, high-density polyethylene cylinders, with rubber-seal screw lids. AMD was fed into the bottom of each reactor using split peristaltic pumps and the treated discharge freely drained from the effluent port situated on the upper part of the reactors (Fig. 1). The upward-flow design was chosen over more conventional downward flow to abate preferential flow paths development and to minimize compaction of the substrate mixture to help maintain an adequate perme-

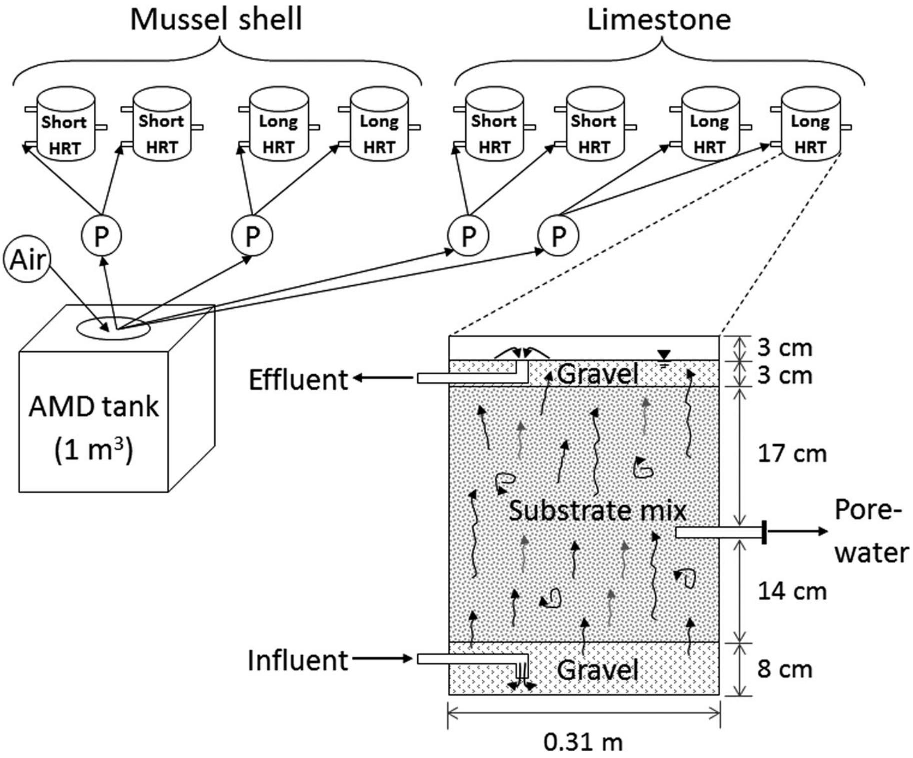


Fig. 1 Experimental set-up and design of a reactor showing water and gas flows

ability (URS 2003). One pore-water sample port was positioned 14 cm above the inlet port. Each reactor was filled with clean sub-rounded gravels (washed in distilled water) to a depth of 8 cm to allow a uniform dispersion of the influent AMD. A 31 cm layer of reactive substrate mixture was placed above this layer and a 3 cm layer of clean sub-rounded gravel was placed on top of the reactive mixture, to maintain the substrate in place. A fine mesh geotextile separated the reactive substrate from the bottom and top gravel layers to prevent washout of fine substrate materials and clogging of the inlet and outlet ports. The original experimental design included Tedlar® bags fixed on top of the reactors to capture the hydrogen sulfide gas produced by the microbial sulfate reduction (Eq. 1). Due to the nature of H_2S gas being relatively heavy and soluble, most of the gas exited the reactors through the effluent port. Even after the addition of a P-shape gas trap on the outlet, a considerable amount of gas continued to exit the reactor through the effluent port, rendering its capture and quantification unfeasible. The experiment was conducted at air room temperature (monitored at 12–19 °C).

The reactive substrate comprised a mixture of organic materials and an alkaline material (Table 1). All organic materials and limestone were obtained from a local garden supplier. The mussel shells were donated by Solid Energy New Zealand and were originally sourced from a seafood company located in Nelson, New Zealand. The AMD was sourced at an active coal mine (West Coast-South Island, New Zealand). In total, three 1 m^3 batches of AMD were used in this study (Table 2). The first batch of AMD, collected from a stream located downstream of the mine site, represented AMD diluted with in-stream surface water and was used to determine the metals present in detrimental concentrations in the immediate vicinity of the mine site. The second and third batches were collected from a settling pond collecting run-off from the waste dumps. The AMD was collected during three separated sampling trips in December 2012, April 2013, and June 2013.

Before the start of the flow-through experiment, the reactors were upward-flow saturated with AMD (from AMD batch #1, see Table 2) and operated in batch mode for a 3-week period. This acclimation stage is necessary to create a favorable reducing environment and to establish an effective and sufficiently important bacterial population capable of sustaining adequate sulfate-reduction rates. The sulfate-reducing bacteria naturally present in the compost self-inoculated the reactors (no system was purposely inoculated). The time period of 3 weeks was based both on recommended values found in the literature and on measured parameters (i.e. oxido-reduction potential, sulfate, and dissolved hydrogen sulfide concentrations) in the pore water samples (Neculita et al. 2008). Following this acclimation period, the AMD, which had been continuously aerated with an air pump to simulate the aerobic and

Table 1 Substrate mixture composition and hydraulic parameters (MS-S = mussel shells short HRT, MS-L = mussel shells long HRT, LS-S = limestone short HRT, and LS-L = limestone long HRT)

	Treatment systems			
	MS-S	MS-L	LS-S	LS-L
Substrate materials (% vol.)				
Mussel shells	30	30	0	0
Limestone	0	0	30	30
Bark (*Pinus radiata*)	30	30	30	30
Bark mulch (*Codyline banksii*)	20	20	20	20
Compost[a]	20	20	20	20
Hydraulic parameters				
Inflow rate (L/day)	4.5	1.5	4.5	1.5
Porosity	0.51	0.51	0.49	0.49
HRT (days)	3.3	10	3.2	9.8

[a] Blend of bark, animal effluents and pig manure sawdust

Table 2 Influent AMD characteristics and water quality guidelines (ANZECC 2000), all metal concentrations are in mg/L

	Trigger values	AMD #1	AMD #2	AMD #3
Period of usage (week)	–	1–5	6–13	14–20
pH	6.5–8.5[a]	4.23	2.65	2.66
ORP (mV)	N/A	220	450	470
Fe	0.3[a]	19	25	60
Al	0.2[a]	5	15	21
Mn	3.6[b]	9	13	18
Cu	0.0025[b]	0.014	0.4	0.3
Zn	0.03[b]	1.05	3.6	4.8
Ni	0.0017[b]	0.4	1	1.3
Cd	0.0008[b]	0.002	0.003	0.004
SO_4^{2-}	400[a]	760	1,560	1,800
Acidity (mg/L $CaCO_3$ eq.)	N/A	n.d.	260	460

n.d. not determined, *N/A* not applicable

[a] Guidelines for recreational purposes

[b] Trigger values for fresh water aquatic ecosystem (80 % protection)

oxidizing conditions typically encountered at an open cast mine, was pumped into the reactors following a semi-continuous upward-flow approach; ORP values varied <1 % and dissolved oxygen stayed >90 %. Four SRBRs (two of each alkaline material) were operated with a short HRT (3.2–3.3 days); four other SRBRs (two of each alkaline material) were operated with a long HRT (9.8–10 days). The corresponding net inflow rates were 4.5 and 1.5 L/day, respectively (Table 1). The actual inflow delivered to each reactor was verified every 2 weeks using a graduated cylinder and a stopwatch.

Substrate Analyses

Chemical analyses performed on the individual organic materials before the start of the flow-through experiment included total metals, total carbon, and total nitrogen (to obtain C/N ratios). Metal concentrations were determined following U.S. EPA solid digestion method 200.8 (Creed et al. 1994) and analyzed by inductively coupled plasma mass spectrophotometry (ICP-MS, Agilent 7500cx), according to Standard Method 3125B (APHA 2005). Total carbon and total nitrogen were analyzed by an IANZ-accredited laboratory following the Dumas method (combustion method) on dry and ground samples (RJ Hill Laboratories, Christchurch). Physical analyses of the substrate mixtures included saturated hydraulic conductivity (k_{sat}) and effective porosity. Saturated hydraulic conductivity was determined using the constant head method (Das 2002). Effective porosity was measured by filling the reactors with tap water, allowing them to sit covered for 48 h, and then letting them drain by gravity. The volume of water drained from each layer (e.g. gravel, substrate mixture) divided by the corresponding layer's volume gave the effective porosity (Table 1). Theoretical HRTs were then calculated using Eq. 2 derived from Darcy's Law (Younger et al. 2002). For each system, individual HRTs were calculated for the gravel layers (bottom and top) and for the substrate mixture layer, and then summed to obtain a total HRT.

$$HRT = \frac{Vn_e}{Q} \tag{2}$$

where V is the layer volume (m^3), n_e is the effective porosity of the corresponding material, and Q is the inflow (m^3/day).

Water Analyses

Influent AMD, pore-water, and treated effluent samples were collected weekly and analyzed immediately for pH (EDT RE-357Tx), ORP (YSI pH100), electric conductivity, and temperature (Thermo Orion 130A). Ferrous iron and sulfide concentrations were measured within 24 h on non-filtered samples using a spectrophotometer (HACH DR-2500). Sulfate samples were filtered (0.22 µm) and stored at 4 °C until analysis using a Dionex ion chromatograph (Dionex ICS-2000). Total and dissolved metals were collected in 120 mL polypropylene bottles, preserved with concentrated nitric acid (69 %, Fisher trace analysis grade), and stored at 4 °C until analysis via ICP-MS (Agilent 7500cx) according to Standard Method 3125B (APHA 2005). Dissolved metals were filtered immediately after collection through a 0.45 µm nylon filter, and total metals were obtained by digestion following the method established by Wicke et al. (2012). Briefly, the samples were thoroughly mixed and 25 mL were transferred into a 50 mL centrifuge tube. After the addition of 5 mL of concentrated nitric acid (69 %, Fisher trace analysis grade), the centrifuge tubes were placed into a heating block and boiled for 1 h. Cooled samples were then filtered (0.45 µm) and analyzed via ICP-MS. Alkalinity on effluent samples and acidity on influent AMD were analyzed by titration to an endpoint of pH 4.5 (alkalinity) and pH 8.3 (acidity) according to Standard Methods 2310 and 2320 (APHA 2005). Quality assurance and control (QA/QC) was achieved through the use of accredited standards, blanks, and duplicate analyses (10 %).

Statistical Analyses

Statistical analyses were performed with IBM SPSS Statistics version 20 (Kirkpatrick 2012). Statistical differences between the four treatment systems were analyzed by two-way ANOVA using the general linear model procedure. Significant differences between means at the $p < 0.05$ level were determined using Tukey HSD test.

Results and Discussion

Influent AMD

Table 2 shows the chemical signatures of the three batches of AMD used in this study as well as the New Zealand water quality guidelines (ANZECC 2000). The metal trigger values (TVs) were obtained from the ANZECC guidelines for freshwater aquatic ecosystems (80 % level of protection, hardness of 30 mg/L as $CaCO_3$) as well as from the ANZECC Recreation Guidelines (for Al, Fe, and SO_4^{2-}). These documents specify TVs of contaminants at various levels of species protection (99, 95, 90, and 80 %) where 80 % protection level is recommended for highly disturbed ecosystems. On comparing in-stream concentrations with the TVs, seven metals of concern were identified (Al, Mn, Fe, Ni, Cu, Zn, and Cd) with values ranging from 2.5 (Cd, Mn) to 235 (Ni) times the TVs. Because TVs are only applicable to concentrations of contaminants in flowing streams where dilution/mixing is occurring, the metal concentrations in AMD batches #2 and #3 cannot be compared directly to the TVs. Moreover, because of analytical uncertainties (QA/QC procedures were not applied properly), and because the first batch of AMD was considered to have too weak a signature to accurately reflect SRBR operational capacity, the results of the first 5 weeks of treatment are not considered in the following sections of this paper.

Substrate Materials

Since suspended solids, metal precipitates, and biofilms can alter the hydraulic properties of the treatment systems (i.e. reduce the permeability), thus affecting their longevity and performance, it is crucial to ensure that a K_{sat} value between 10^{-2} and 10^{-3} cm/sec is maintained through the SRBR's operation (URS 2003). In this study, K_{sat} values were 6.3×10^{-2} and 9.7×10^{-2} cm/s for the mixture containing the mussel shells and the limestone, respectively. Effective porosities for lab-scale systems should be within the range of 0.35–0.63 (Amos and Younger 2003; Neculita et al. 2008). In the present study, effective porosities were 0.51 and 0.49 for the mixture containing mussel shells and limestone, respectively, and 0.38 for the gravel. The different calculated HRTs resulted from the different measured effective porosities. Because both hydraulic conductivity and effective porosity measured in this study were in the ranges of recommended values, the risk of clogging and/or short-circuiting was estimated to be relatively low.

Metal content in the organic materials were low (<0.4 g/kg) and several orders of magnitude less than the AMD total metal loadings (over the 20 week period) for all metals of concern except for Al and Fe. Aluminum and Fe contents were 3.6 g Al/kg and 4.2 g Fe/kg (for bark), and 4.2 g Al/kg and 5.2 g Fe/kg (for compost). Nevertheless, total Al and Fe contents were still less than the AMD total loading, indicating that the materials used in this study were unlikely to be a source of contamination. C/N ratios for the compost, bark, and bark mulch were 27, 280, and 150, respectively. These values are slightly out of the recommended range of 45–120 defined by Okabe et al. (1992). Values above this threshold are reported to be too low in nitrogen to properly stimulate SRB growth, possibly indicating that SRB establishment during the acclimation period was lower than expected. However, it worth noting that Okabe et al. (1992) used lactate in their study, which is a readily available carbon source for the SRB compared to the more recalcitrant carbon sources used in the present study.

In-Situ Parameters, Sulfate Removal and Alkalinity Generation

Figure 2 shows the changes in pH, ORP, sulfate, and alkalinity concentrations throughout the experiments. While several effluent concentrations including alkalinity, sulfate, and sulfide fluctuated during the first 7 weeks of treatment, other parameters such as pH, ORP, conductivity, and temperature showed steady-state values from week one onward. These fluctuations are common in new

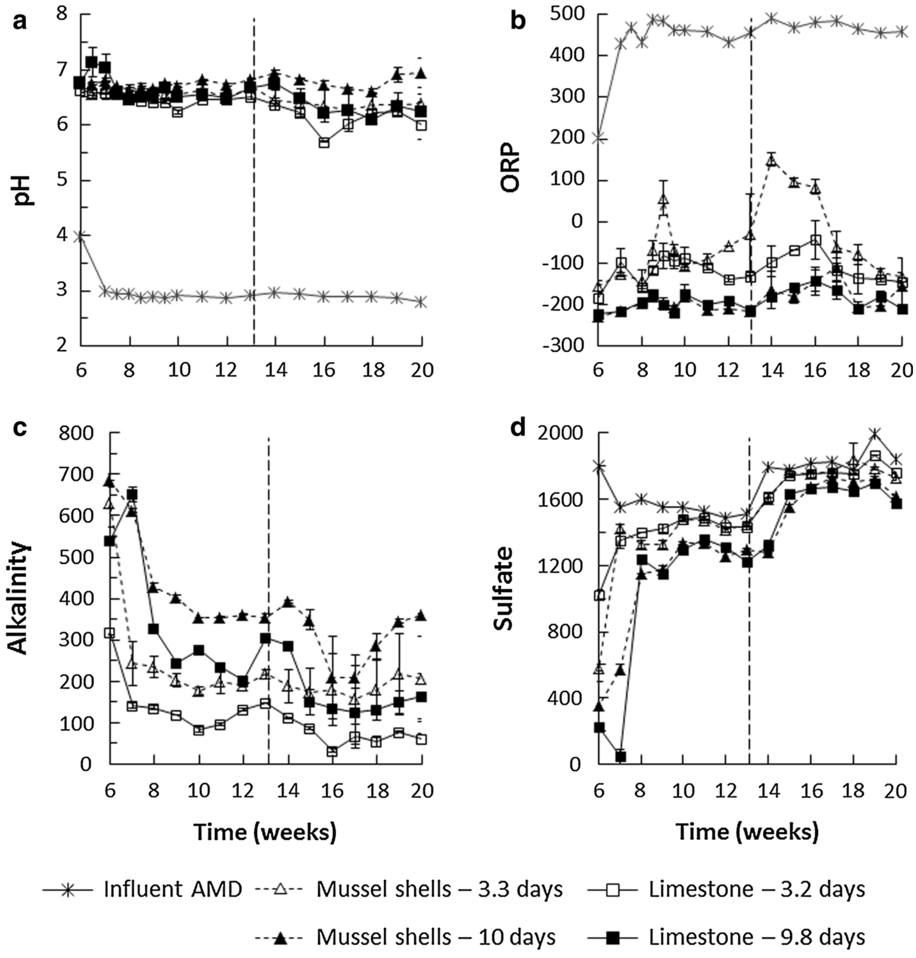


Fig. 2 Changes over time of **a** effluent pH; **b** pore-water ORP (mV); **c** effluent alkalinity (mg/L $CaCO_3$ eq.); and **d** effluent sulfate concentration (mg/L). *Dashed lines* indicate when a new batch of influent AMD is used

waterlogged systems as the biogeochemistry establishes (Dvorak et al. 1992).

Median pH increased from 2.9 in the influent to effluent values of 6.49 (MS-S), 6.79 (MS-L), 6.36 (LS-S), and 6.49 (LS-L) (Fig. 2a). Pore-water pH differed by <2 % from the effluent pH, indicating that the bulk of the neutralization occurred in the lower (first) half of the reactor beneath the pore-water effluent port (data not shown). Systems operating at a longer HRT as well as systems containing mussel shells showed a slightly higher pH compared to systems operating at a shorter HRT or those containing limestone. A Tukey post hoc statistical test showed that pH differences between short and long HRT, as well as between mussel shells and limestone systems, were both statistically significant ($p < 0.001$). ORP values in the pore-water samples were constantly negative except for system MS-S, which showed positive values during four sampling events (Fig. 2b). Three of the four events happened after the third batch of AMD was introduced, possibly indicating that the SRB were impacted by the higher metal loading. Nevertheless, treatment efficiency in terms of pH, alkalinity generation, and metal removal did not show any signs of decline during these 3 weeks. Because the HRT had a statistically significant influence ($p < 0.001$) on the ORP (a longer HRT resulted in a lower ORP), another explanation could be the formation of preferential flow paths in system MS-S.

Sulfate removal ranged from 0.14 mol/m^3/day (5 %) to 0.18 mol/m^3/day (14.8 %) (Fig. 2d). Several authors reported sulfate removal rates ranging from 0.17 to 0.44 mol/m^3/day (Dvorak et al. 1992; Gibert et al. 2004; Thomas and Romanek 2002a), and a value of 0.3 mol/m^3/day is frequently recommended as a SRBR design criteria (Gusek 2002). Despite the fact that pore-water pH and ORP were both within the desirable range (pH > 5.5 and ORP <−100 mV) for SRB to metabolize (Postgate 1979), the hypothesis of a low initial SRB establishment (due to the substrate's low nitrogen content) possibly explains the relatively low sulfate removal rate obtained in this study. Additionally, despite the 3-week acclimation period, the SRB inoculated from the compost were adapted to circumneutral pH conditions (i.e. the bacterial community was not acidophilic) and their activity may have suffered from the influent low pH. Dissolved sulfide concentrations in the effluent were overall very low (<1 mg/L). Nevertheless, a strong sulfide odor was detected during all sampling events indicating that a substantial amount of H_2S gas escaped the reactors, as mentioned earlier. This observation is in accordance with the fact that at the observed pH, temperature, and pressure ranges, most of the hydrogen sulfide is present in the water as insoluble H_2S gas (Carroll and Mather 1989). Moreover, some of the H_2S gas may have escaped from the sample bottles between the time of sampling and the analyses.

Median effluent alkalinity concentrations for each system were 194.4 mg/L $CaCO_3$ (MS-S), 354.4 mg/L $CaCO_3$ (MS-L), 91.3 mg/L $CaCO_3$ (LS-S), and 219.4 mg/L $CaCO_3$ (LS-L) (Fig. 2c). Treatment systems containing mussel shells produced greater alkalinity than systems containing limestone at the same HRT and, in general, systems operating at a longer HRT produced more alkalinity than systems running at a short HRT. After 5 months of continuous operation, alkalinity generation from mussel shells and limestone was not exhausted or obviously diminished. Statistically significant differences ($p < 0.001$) between system MS-L and the three other systems demonstrated that MS-L was constantly the most effective treatment system at generating alkalinity. Although the median alkalinity values indicated that the limestone-long (LS-L) HRT generated more alkalinity than mussel shells-short HRT (MS-S), statistical analyses revealed that the difference was not statistically significant ($p = 0.859$). Furthermore, until week 14, the two systems operating at a longer HRT (MS-L and LS-L) generated more alkalinity than systems operating at a shorter HRT (MS-S and LS-S) but from week 15 onward, system MS-S produced more alkalinity than system LS-L, indicating that alkalinity generation was driven by its source rather than influenced by its HRT. The most probable reason for the greater alkalinity generation from mussel shells compared to limestone is a faster dissolution rate. Different properties, including mineralogy, reactive surface area, grain size, calcium carbonate content, and calcium carbonate structure, may explain this difference in reactivity between mussel shells and limestone. While calcium carbonate present in the limestone is assumed to be purely calcite, mussel shells are comprised of a mixture of calcite (approx. 90 %) and aragonite (approx. 10 %) (Cubillas et al. 2005b). Cubillas et al. (2005b) also demonstrated that mussel shells' BET surface-area increase by 80 % during dissolution, compared to 30 % for pure calcite. Another study by Cubillas et al. (2005a), showed that the dissolution of aragonite was less affected by mineral armoring or coating than the dissolution of calcite. However, limestone passivation by armoring or coating of iron precipitates is unlikely to happen in reducing systems like SRBRs (Thomas and Romanek 2002a; URS 2003). Furthermore, nitrogen present within the proteins in the periostracum (outer layer of the mussel shells) may helped the microbial community and contributed to the total alkalinity generation via SRB neutralization (Eq. 1) (Abdulkarim et al. 2013; McCauley et al. 2009). However, this last hypothesis is not supported by the sulfate measurements, as very little difference in sulfate removal was observed between MS and LS systems.

Figure 3 shows the alkalinity generation from calcium carbonate dissolution and from sulfate-reducing bacteria neutralization (Eq. 1). Microbial contribution to alkalinity generation was calculated using the sulfate concentration mass balances (from week 8 to 20) by assuming that, after the initial sorption onto organic matter, the only sulfate removal mechanisms was microbial reduction, and that no gypsum was precipitating in the substrate (confirmed using PHREEQC 3.0.6 with the MinteqV4 database). $CaCO_3$ contribution to alkalinity was then obtained by subtracting the sulfate-reduction contribution to the total alkalinity generated. Even though gypsum's saturation indexes (SI) from PHREEQC modelling in pore-water and effluent samples were always negative (data not shown), it is important to consider that: (1) PHREEQC modelling does not consider microbial influence, and; (2) SI values in the open solution do not necessarily represent near-surface values (where solute concentrations and pH can be substantially higher), and therefore the presence of gypsum

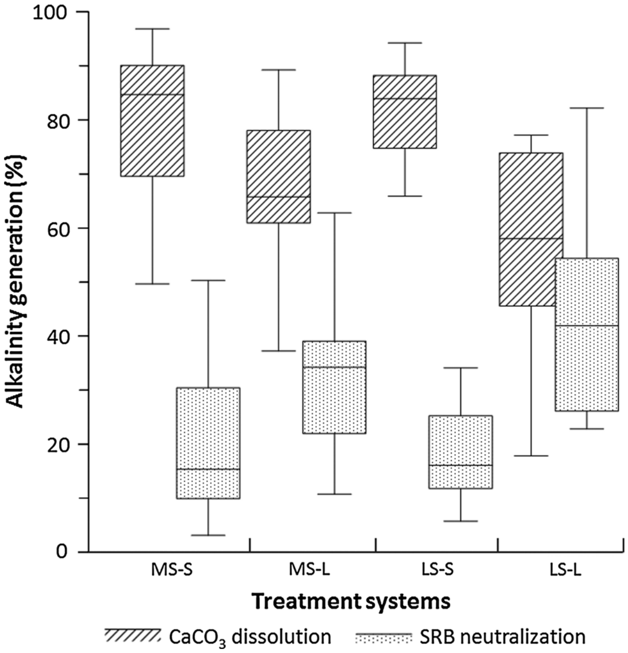
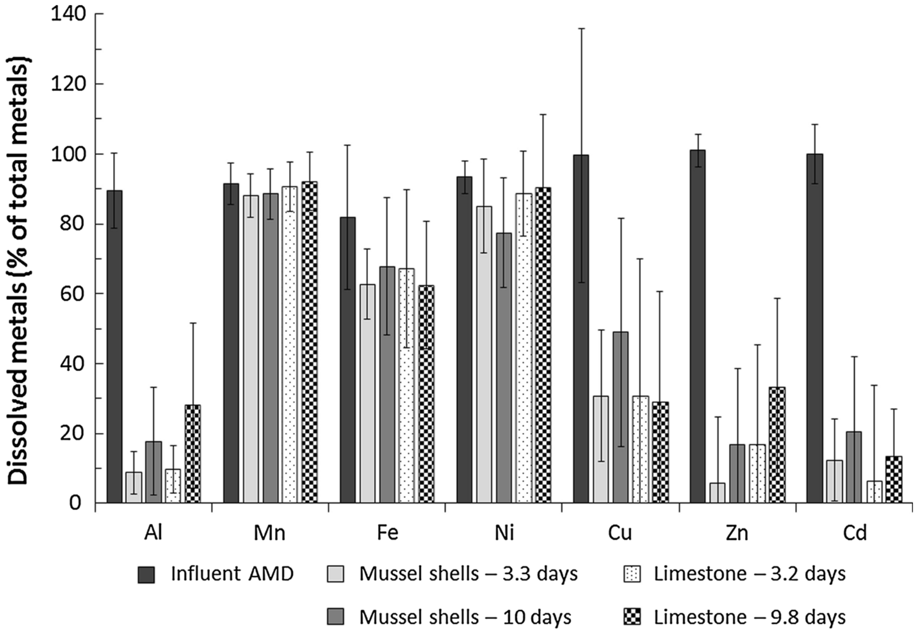


Fig. 3 Alkalinity generation from calcium carbonate dissolution and sulfate-reducing bacteria neutralization (*MS-S* mussel shells short HRT, *MS-L* mussel shells long HRT, *LS-S* limestone short HRT, and *LS-L* limestone long HRT)

Fig. 4 Percentages of dissolved metals in total metals for influent and effluent samples



cannot be entirely ruled out. Indeed, Rose et al. (2007) found small amounts of gypsum precipitating within Al precipitates and/or in patches on the limestone surface in four vertical flow ponds where PHREEQC modelling of the effluent indicated negative SI values for gypsum.

Overall, $CaCO_3$ dissolution generated substantially more alkalinity than bacterial neutralization. Additionally, the two alkalinity generation pathways exhibited distinct behaviors. Calcium carbonate dissolution was mainly a function of the amount of acidity entering the systems with its dissolution negatively influenced by HRT (short HRTs resulted in more alkalinity production). By contrast, SRB neutralization was positively correlated to HRT. Moreover, SRB neutralization was not affected by the alkalinity source as MS and LS systems were not significantly different.

Metal Removal

A comparison between total and dissolved metal concentrations showed that most metals of concern were present predominantly in their dissolved forms in the influent and in particulate forms in the effluents (Fig. 4). This change in speciation was observed for all metals apart from manganese and to a lesser extent for nickel and iron (more than 90 % Mn, 80 % Ni, and 60 % Fe stayed in their dissolved forms in the effluent). Even though this change in speciation between the influent and the effluents was clear, the overlapping error bars suggested that there was no significant difference between the four treatment systems. However, it is worth noting that the distinction between total and dissolved metals is operationally defined by filtration at 0.45 μm and that some metals might not have been truly dissolved, but present in a colloidal form.

In reducing systems, Fe is suspected to be removed mainly as iron sulfides, but depending on the redox potential and the pH, iron hydroxides and iron carbonates may precipitate as well (Younger et al. 2002). Al is assumed to be solely retained as a hydroxide, e.g. $Al(OH)_3$ (Stumm and Morgan 1996), and trace metals like Cu, Zn, Ni, and Cd are expected be removed principally via sulfide precipitation (Younger et al. 2002). Due to its high solubility, manganese sulfide was not expected to form under the present conditions (Dvorak et al. 1992; Jong and Parry 2003; Machemer and Wildeman 1992), and precipitation as oxides and hydroxides are unlikely under reducing conditions. Therefore, manganese carbonate precipitation (e.g. rhodochrosite, kutnohorite), as well as adsorption onto organic matter and iron (oxi)hydroxides are potential removal mechanisms (Bamforth et al. 2006; Trumm and Ball 2014; Younger et al. 2002).

Figure 5 shows the amount of Fe and Mn retained in the SRBR's substrates and discharged in the effluent during the 20 weeks of treatment. Clearly, there were differences in removal behavior of Fe and Mn. First, Fe removal seems to be independent of the loading (at the concentrations used in this study). While Fe loading increased three-fold at week 13 (changing from AMD batch #2 to batch #3), the Fe discharge only increased by a few mg/d, and correlation coefficients between cumulative loading and effluent discharge concentrations showed no clear relationship (R^2 ranged from 0.19 to 0.77). Moreover, the four treatment designs do not show great differences in the amount of Fe discharged, even though systems containing mussel shells

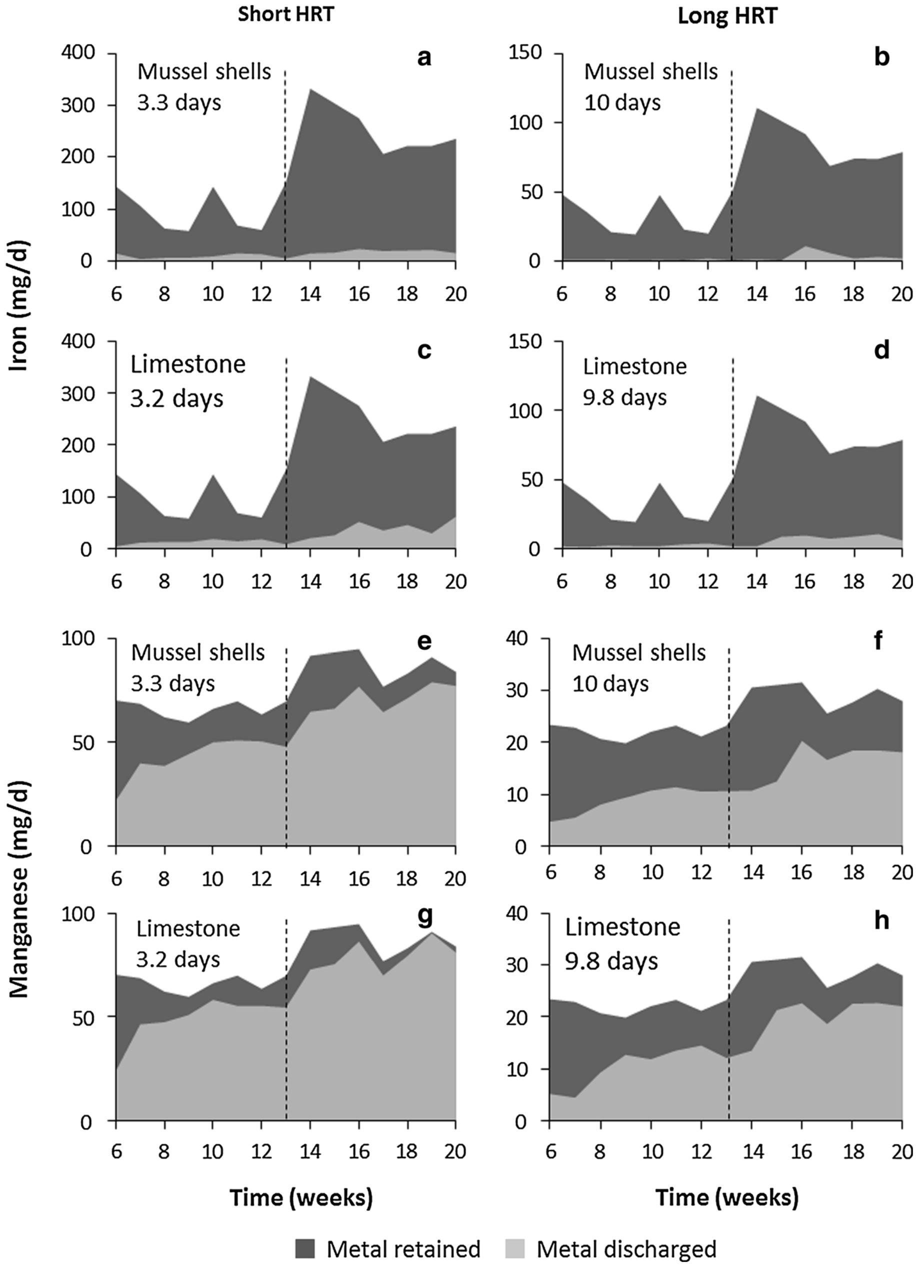


Fig. 5 Metal loading plots showing Fe and Mn retained and discharged from each system (*dashed lines* indicate new batch of influent AMD). **a** Fe MS-S, **b** Fe MS-L, **c** Fe LS-S, **d** Fe LS-L, **e** Mn MS-S, **f** Mn MS-L, **g** Mn LS-S, **h** Mn LS-L

retained more iron than systems containing limestone. This general behavior supports the hypothesis that the main Fe removal mechanism is precipitation as iron sulfides and/or iron (oxi)hydroxides. After 20 weeks of treatment, Fe removal started exhibiting signs of decreased efficiency for system LS-S only, suggesting that insufficient sulfide and/or bicarbonate were generated (Fig. 5). By contrast, Mn showed a constant decline in removal efficiency over time, indicating that adsorption might be an important retention mechanism (as adsorption sites reached capacity, less Mn was being removed). Moreover, correlation coefficients between cumulative loading and discharge were significantly higher (R^2 ranging from 0.83 to 0.88). Important differences were observed between each system. Reactors containing mussel shells and/or operating at a longer HRT retained more Mn, indicating that Mn removal is positively correlated with the higher alkalinity reported for these systems. Under the reducing conditions observed in this study, it is unlikely that Mn is being removed as an oxide or a hydroxide. Moreover, if we assume that most of the iron is removed as a sulfide, adsorption onto the organic matter and precipitation as a carbonate (e.g. rhodochrosite) are the most likely removal mechanisms. This is supported by: (1) the work of Cubillas et al. (2005b), which showed that mussel shells afford more adsorptive surface area than limestone, and; (2) PHREEQC modeling, which showed

Table 3 Treatment efficiency in terms of dissolved metal removal (%)

	Al	Fe	Mn	Cu	Zn	Ni	Cd
Mussel shells—short HRT	99.5	92.0	29.6	89.6	99.5	83.0	99.0
Mussel shells—long HRT	99.7	95.2	55.2	91.8	99.6	95.3	99.4
Limestone—short HRT	99.0	86.5	19.8	87.4	99.1	81.7	98.1
Limestone—long HRT	99.4	91.8	47.1	91.2	96.1	82.7	98.8

that rhodochrosite was slightly oversaturated in all four systems' effluents, but that important SI differences resulted from the different treatments: MS-S (0.4), MS-L (0.85), LS-S (0.03), and LS-L (0.47).

Treatment performances were also assessed using the percentage of metals removed (Table 3) and the area-adjusted removal rate (Table 4) computed using Eq. 3 as defined by Wieder (1989), and Eq. 4 as defined by Hedin et al. (1994), where C_{in} and C_{out} are inflow and outflow metal concentrations (mg/L), R_A is the area-adjusted removal rate (g/m^2/day), Q is flow rate (m^3/day), and A is area (m^2). However, in SRBR design, the depth is often a more important dimension than the length, width, or radius (in case of a column-shaped reactor). Therefore, reporting results in term of volume-adjusted removal rate (g/m^3/day) by substituting volume (m^3) to area (m^2) in Eq. 4 seemed more appropriate.

$$Treatment\ Efficiency\ (\%) = \frac{C_{in} - C_{out}}{C_{in}} \times 100 \tag{3}$$

$$R_A = \frac{Q(C_{in} - C_{out})}{A} \tag{4}$$

Metal removal was effective in all four treatment designs for all metals of concern apart from Mn (Table 3). The overall order of removal efficiency was Al > Cd > Zn > Fe > Cu > Ni ≫ Mn. Generally, systems operating at a longer HRT removed more metals than systems operating at a short HRT, with the exception of Zn, for which system LS-L did not perform as well as LS-S. Similarly, systems containing mussel shells removed more metals than systems containing limestone, concurring with the alkalinity data. MS-L was consistently the most effective system, removing ≥91.8 % of all metals (except Mn). Nevertheless, a relatively high Mn treatment efficiency (55.2 %) was achieved in system MS-L compared to the other studies (reported Mn removal efficiencies in similar SRBR systems range from 0 to 36 %) (McCauley 2011; Trumm and Ball 2014).

For each of the four treatment systems, the following order of metal removal efficiency was observed for both area- and volume-adjusted removal rates: Fe > Al > Mn > Zn > Ni > Cu > Cd (Table 4). Moreover, for Al, Fe, and Mn, system efficacy followed the same order (MS-S > LS-S > MS-L > LS-L). Overall, the short HRT systems always outperformed long HRT systems, indicating that metal loading is probably a limiting factor, and that the long HRT systems possibly performed below capacity. While Fe, Mn and Zn removal rates were slightly above typical rates reported in the literature, Cu, Ni, and Cd were well below. The low influent pH at 2.65 could explain these sub-optimal removal rates, as most of the literature values reported in Table 4 were derived from field and laboratory

Table 4 Area (g/m^2/day) and volume (g/m^3/day) adjusted dissolved metal removal rates (MS-S = mussel shells short HRT, MS-L = mussel shells long HRT, LS-S = limestone short HRT, and LS-L = limestone long HRT)

	MS-S		MS-L		LS-S		LS-L		Published area-adjusted removal rates
	Area-adjusted	Volume-adjusted	Area-adjusted	Volume-adjusted	Area-adjusted	Volume-adjusted	Area-adjusted	Volume-adjusted	
Al	0.90	2.92	0.30	0.97	0.90	2.91	0.30	0.97	0.8[a]
Fe	1.44	4.63	0.50	1.62	1.50	4.83	0.50	1.61	0.01[a]
Mn	0.26	0.84	0.16	0.50	0.17	0.54	0.13	0.41	10[b]
Cu	0.009	0.032	0.003	0.011	0.006	0.034	0.003	0.011	0.11[c]
Zn	0.250	0.805	0.084	0.272	0.239	0.771	0.083	0.269	2[d]
Ni	0.052	0.168	0.017	0.056	0.048	0.156	0.017	0.056	0.02[d]
Cd	1.65E−04	5.31E−04	5.51E−05	1.78E−04	1.65E−04	5.33E−04	5.42E−05	1.75E−04	0.02[d]

[a] Heal and Salt (1999)

[b] PIRAMID Consortium (2003)

[c] Kadlec and Knight (1996)

[d] Ettner (1999)

experiments using pH > 3 AMD. Moreover, depending on reactor design, substrate materials, microbial development as well as influent pH and metals loadings, large disparities in treatment performances have been reported (Mayes et al. 2009), thus limiting comparison of published removal rates using different operational designs.

A comparison (in mol/L) between the amount of sulfate reduced and the amount of metals retained (except for Al and Mn, which were not assumed to form sulfides under the experimental conditions) showed that sulfate removal exceeded metals removal in each of the four systems. In long HRT systems, 5.4E−04 mol of metals were retained and 2.4E−03 mol of sulfate reduced, while in short HRT systems, 5.1E−04 mol of metal were removed, but only 7.9E−04 mol of sulfate reduced. This indicated that long HRT systems are likely to remove most of the metals of concern (except Al and Mn) as sulfides, and supported the hypothesis that metal loading might actually be a limiting factor to the metal sulfide removal pathway, possibly explaining the small removal rates reported in Table 4. For the short HRT systems, even though metal sulfide formation most likely occurred, precipitation as hydroxides and carbonates may also be important removal mechanisms. Future reactor autopsies including scanning-electron microscopy and chemical sequential extractions performed on these reactors will help us to understand the exact removal mechanisms occurring in each system.

Conclusions

This study evaluated the influence of two alkalinity sources (mussel shells and limestone) and the effect of two HRTs ($\approx$3 and 10 days) on the treatment performance of eight upward-flow SRBRs. Each of the four SRBR designs (operated in duplicate) successfully treated AMD over a continuous period of 5 months. Overall, metal concentrations significantly decreased, pH and alkalinity significantly increased, and sulfate showed relatively poor removal efficiency. Median metal removal efficiencies were $\geq$99 % for Al and $\geq$86.5 % for Fe. Higher Mn removal (55.2 %) occurred in system MS-L than in the other systems. Generally, reactors operating at a long HRT and/or containing mussel shells had better metal removal and alkalinity generation than systems operating at a short HRT and/or containing limestone. Overall, this study demonstrated that: (1) waste mussel shells offer an attractive alternative to limestone for AMD passive treatment in New Zealand, and; (2) even though an increase in HRT resulted in better contaminant removal, a HRT of less than 4 days was sufficient to remove more than 80 % of all metals except Mn. Therefore, the choice of HRT must balance the need to meet a specific effluent quality while keeping the treatment time reasonably short, and an intermediate retention time might have been optimal. Longer investigations will be undertaken to evaluate if mussel shells can sustain their greater alkalinity generation and if organic depletion will occur.

Acknowledgments This research was funded by CRL Energy Ltd, the Dept of Civil and Natural Resources Engineering at the University of Canterbury (New Zealand), and the University of Lausanne (Switzerland). Technical and logistical support was provided by various university technicians including Peter McGuigan, David McPherson, Robert Stainthorpe, and Matt Cockcroft. Francis Mining Company Ltd kindly granted access to the Echo Mine site for AMD collection. Aaron Dutton from CRL Energy Ltd helped during AMD collection. Solid Energy New Zealand donated the mussel shells.

References

Abdulkarim A, Tijani IM, Abdulsalam S, Abubakar Jaju M, Alewo OA (2013) Extraction and characterisation of chitin and chitosan from mussel shell. Civ Environ Res 3(2):108–114

Amos PW, Younger PL (2003) Substrate characterisation for a subsurface reactive barrier to treat colliery spoil leachate. Water Res 37(1):108–120

ANZECC (2000) Australian and New Zealand Environment and Conservation Council (ANZECC) Water quality guidelines

APHA (2005) Standard methods for the examination of water and wastewater. American Public Health Association (APHA), 21st Edit, APHA, Washington, DC

Bamforth SM, Manning DAC, Singleton I, Younger PL, Johnson KL (2006) Manganese removal from mine waters—investigating the occurrence and importance of manganese carbonates. Appl Geochem 21(8):1274–1287

Batty LC, Younger PL (2004) The use of waste materials in the passive remediation of mine water pollution. Surv Geophys 25(1):55–67

Benner SG, Blowes DW, Ptacek CJ, Mayer KU (2002) Rates of sulfate reduction and metal sulfide precipitation in a permeable reactive barrier. Appl Geochem 17(3):301–320

Blowes DW, Ptacek CJ, Jambor JL, Weisener CG (2003) The geochemistry of acid mine drainage. In: Lollar BS (ed) Environmental geochemistry: treatise on geochemistry, vol 9. Elsevier, Amsterdam, pp 149–204

Byrne P, Wood PJ, Reid I (2012) The impairment of river systems by metal mine contamination: a review including remediation options. Crit Rev Environ Sci Technol 42(19):2017–2077

Carroll JJ, Mather AE (1989) The solubility of hydrogen sulphide in water from 0 to 90°C and pressures to 1 MPa. Geochim Cosmochim Acta 53:1163–1170

Creed JT, Brockhoff CA, Martin TD (1994) Determination of trace elements in waters and wastes by inductively coupled plasma mass spectrometry, revision 5.4, EMMC version. US Environmental Protection Agency, Washington

Crombie FM, Weber PA, Lindsay P, Thomas DG, Rutter GA, Shi P, Rossiter P, Pizey MH (2011) Passive treatment of acid mine drainage using waste mussel shell, Stockton coal mine, New Zealand. In: Bell LC, Braddock B (eds) 7th Australian acid and metalliferous drainage workshop, Darwin, Australia

Cubillas P, Köhler S, Prieto M, Causserand C, Oelkers EH (2005a) How do mineral coatings affect dissolution rates? An experimental study of coupled $CaCO_3$ dissolution–$CdCO_3$ precipitation. Geochim Cosmochim Acta 69(23):5459–5476

Cubillas P, Köhler S, Prieto M, Chaïrat C, Oelkers EH (2005b) Experimental determination of the dissolution rates of calcite, aragonite, and bivalves. Chem Geol 216(1–2):59–77

Das BM (2002) Principles of geotechnical engineering, 5th edn. Brooks Cole/Thompson Learning, Pacific Grove, CA

Dvorak DH, Hedin RS, Edenborn HM, McIntire PE (1992) Treatment of metal-contaminated water using bacterial sulfate-reduction: results from pilot-scale reactors. Biotechnol Bioeng 40(5):609–616

Ettner DC (1999) Pilot scale constructed wetland for the removal of nickel from tailings drainage, southern Norway. In: Proceedings, congress of the international mine water association, pp 207–211

Garcia C, Moreno DA, Ballester A, Blázquez ML, González F (2001) Bioremediation of an industrial acid mine water by metal-tolerant sulphate-reducing bacteria. Miner Eng 14(9):997–1008

Gibert O, de Pablo J, Cortina JL, Ayora C (2004) Chemical characterisation of natural organic substrates for biological mitigation of acid mine drainage. Water Res 38(19):4186–4196

Gitari WM, Petrik LF, Etchebers O, Key DL, Iwuoha E, Okujeni C (2008) Passive neutralisation of acid mine drainage by fly ash and its derivatives: a column leaching study. Fuel 87(8–9):1637–1650

Gusek JJ (2002) Sulphate-reducing bioreactor design and operating issues: is this the passive treatment technology for your mine drainage? In: Proceedings, National meeting of the American society of mining and reclamation (ASMR), Park City, UT, USA

Gusek JJ (2005) Design challenges for large scale sulfate-reducing bioreactors. In: Calabrese EJ, Kostecki PT, Dragun J (eds) Contaminated soils, sediments and water, vol 9. Springer, NYC, NY, USA, pp 33–44. doi:10.1007/0-387-23079-3_4

Hao OJ, Chen JM, Huang L, Buglass RL (1996) Sulfate-reducing bacteria. Crit Rev Environ Sci Technol 26(2):155–187

Heal KV, Salt CA (1999) Treatment of acidic metal-rich drainage from reclaimed ironstone mine spoil. Water Sci Technol 39(12):141–148

Hedin RS, Nairn RW, Kleinmann RLP (1994) Passive treatment of coal mine drainage. US Bureau of Mines IC 9389, US Dept of Interior, Pittsburgh, PA, USA

Johnson DB, Hallberg KB (2005) Acid mine drainage remediation options: a review. Sci Total Environ 338(1–2):3–14

Jong T, Parry DL (2003) Removal of sulfate and heavy metals by sulfate reducing bacteria in short-term bench scale upflow anaerobic packed bed reactor runs. Water Res 37(14):3379–3389

Kadlec RH, Knight RL (1996) Treatment wetlands. CRC Press, Boca Raton, Florida, p 893

Kirkpatrick LA (2012) A simple guide to IBM SPSS: for version 20.0. Cengage Learning, Stamford

Kohler SJ, Cubillas P, Rodriguez-Blanco JD, Bauer C, Prieto M (2007) Removal of cadmium from wastewaters by aragonite shells and the influence of other divalent cations. Environ Sci Technol 41(1):112–118

Lens PNL, Visser A, Janssen AJH, Pol L, Hulshoff W, Lettinga G (1998) Biotechnological treatment of sulfate-rich wastewaters. Crit Rev Environ Sci Technol 28(1):41–88

Machemer SD, Wildeman TR (1992) Adsorption compared with sulfide precipitation as metal removal processes from acid mine drainage in a constructed wetland. J Contam Hydrol 9(1–2):115–131

Mayes WM, Batty LC, Younger PL, Jarvis AP, Koiv M, Vohla C, Mander U (2009) Wetland treatment at extremes of pH: a review. Sci Total Environ 407(13):3944–3957

McCauley CA (2011) Assessment of passive treatment and biochemical reactors for ameliorating acid mine drainage at Stockton coal mine. PhD thesis, University of Canterbury, Christchurch, New Zealand

McCauley CA, O'Sullivan AD, Milke MW, Weber PA, Trumm DA (2009) Sulfate and metal removal in bioreactors treating acid mine drainage dominated with iron and aluminum. Water Res 43(4):961–970

Nairn RW, Mercer MN (2000) Alkalinity generation and metals retention in a successive alkalinity producing system. Mine Water Environ 19:124–133

Neculita CM, Zagury GJ, Bussière B (2007) Passive treatment of acid mine drainage in bioreactors using sulfate-reducing bacteria: critical review and research needs. J Environ Qual 36(1):1–16

Neculita CM, Zagury GJ, Bussière B (2008) Effectiveness of sulfate-reducing passive bioreactors for treating highly contaminated acid mine drainage: I. Effect of hydraulic retention time. Appl Geochem 23(12):3442–3451

Nordstrom DK, Alpers CN (1999) Geochemistry of acid mine waste. In: Plumlee GS, Logsdon MJ (eds) The environmental geochemistry of mineral deposits. Part A: processes, techniques and health issues. Reviews in economic geology, vol 6, pp 133–160

Okabe S, Nielsen PH, Characklis WG (1992) Factors affecting microbial sulfate reduction by *Desulfovibrio desulfuricans* in continuous culture: limiting nutrients and sulfide concentration. Biotechnol Bioeng 40(6):725–734

PIRAMID Consortium (2003) Engineering guidelines for the passive remediation of acidic and/or metalliferous mine drainage and similar wastewaters. University of Newcastle Upon Tyne, UK

Pope J, Newman N, Craw D, Trumm D, Rait R (2010) Factors that influence coal mine drainage chemistry West Coast, South Island, New Zealand. N Z J Geol Geophys 53(2):115–128

Postgate JR (1979) The sulphate-reducing bacteria, 1st edn. Cambridge University Press, Cambridge

Robinson-Lora MA, Brennan RA (2009) Efficient metal removal and neutralization of acid mine drainage by crab-shell chitin under batch and continuous-flow conditions. Bioresour Technol 100(21):5063–5071

Robinson-Lora MA, Brennan RA (2010) Chitin complex for the remediation of mine impacted water: geochemistry of metal removal and comparison with other common substrates. Appl Geochem 25(3):336–344

Rose AW (2010) Advances in passive treatment of coal mine drainage (1998–2009). Presented at the 27th national meeting, ASMR, Pittsburgh, PA, USA

Rose AW, Morrow T, Dunn M, Denholm C (2007) Mode of gypsum precipitation in vertical flow ponds. In: Barnhisel RI (ed), National meeting of the ASMR, Gillette, WY, USA

Salomons W (1994) Environmental impact of metals derived from mining activities: processes, predictions, prevention. J Geochem Explor 52:5–23

Simmons J, Ziemkiewicz P, Black DC (2001) Use of steel slag leach beds for the treatment of acid mine drainage: the McCarty highwall project. Presented at the national association of abandoned mine lands annual conference, Athens, OH, USA

Skousen JG, Sexstone A, Ziemkiewecz PF (2000) Acid mine drainage controle and treatment. In: Barnhisel RI, Darmody RG, Daniels WL (eds) Reclamation of drastically disturbed lands, American society of agronomy-crop science society of America-soil science society of America, Madison, WI, USA

Stumm W, Morgan JJ (1996) Aquatic chemistry: chemical equilibria and rates in natural waters. Wiley-Interscience, NYC

Tang K, Baskaran V, Nemati M (2009) Bacteria of the sulphur cycle: an overview of microbiology, biokinetics and their role in petroleum and mining industries. Biochem Eng J 44(1):73–94

Thomas RC, Romanek CS (2002a) Passive treatment of low-pH, ferric iron-dominated acid rock drainage in a vertical flow wetland I: acidity neutralization and alkalinity generation. Presented at the national meeting ASMR, Lexington, KY, USA

Thomas RC, Romanek CS (2002b) Passive treatment of low-pH, ferric iron-dominated acid rock drainage in a vertical flow wetland II: metal removal. Presented at the national meeting ASMR, Lexington, KY, USA

Trumm D, Ball J (2014) Use of sulfate-reducing mussel shell reactors in New Zealand for treatment of acid mine drainage. Presented at the West Virginia mine drainage task force symposium, Morgantown, WV, USA

URS (2003) Passive and semi-active treatment of acid rock drainage from metal mines-state of the practice. Prepared for US Army Corps of Engineers, Concord, MA, USA

Watzlaf GR, Schroeder KL, Kleinmann RLP, Kairies CL, Nairn RW (2004) The passive treatment of coal mine drainage National Energy Technology Laboratory, US Dept of Energy, Pittsburgh, PA, USA

Wicke D, Cochrane TA, O'Sullivan AD (2012) Build-up dynamics of heavy metals deposited on impermeable urban surfaces. J Environ Manage 113:347–354

Widdel F (1988) Microbiology and ecology of sulfate- and sulfur-reducing bacteria. In: Zehnder AJB (ed) Biology of anaerobic microorganisms. Wiley, NYC, pp 469–586

Wieder RK (1989) A survey of constructed wetlands for acid coal mine drainage treatment in the eastern United States. Wetlands 9(2):299–315

Wildeman T, Schmiermund R (2004) Mining influenced waters: their chemistry and methods of treatment. Presented at the ASMR national meeting and the 25th West Virginia surface mine drainage task force, Morgantown, WV, USA

Wildeman TR, Gusek JJ, Higgins J (2006) Passive treatment of mine influenced waters. In: Course material for the ARD treatment short course, 7th international conference on acid rock drainage (ICARD), St. Louis MO, USA

Younger PL (2004) The mine water pollution threat to water resources and its remediation in practice. IDS Water Europe

Younger PL, Banwart SA, Hedin RS (2002) Mine water: hydrology, pollution, remediation environmental pollution. Kluwer Academic, Dordrecht

Zagury GJ, Neculita CM, Bussière B (2005) Passive biological treatment of acid mine drainage: challenges of the 21st century. Presented at the 2nd symposium on mining and the environment, Rouyn-Noranda, QC, Canada