https://dspace.org/

Building blocks for better biorepositories in Africa

T Croxton; E Jonathan; K Suleiman; O Balogun; Mukthar Kader ...

https://hdl.handle.net/10539/39535
WOS

Downloaded from a DSpace repository. https://dspace.org/

! "
" # &
' " (

! ( ) " * + , - !) + !

. " " ! " !
! " " # ! " ! # $! ! % " & !
% ! " ! '! % ()*% + 0-,%1-/,)2,+,0 0 - ,(

. + "3)11"! 0! #1- 0- , 1- ,,2,+,0 0 - ,(

4 $ " !) + / 0

$! ! &!

---

5) -+,

---

6 "

---

7 8 9! " ! ! " $! "
3)11 0 " ! 0 ! 1 ! 1&! ! ! :&! 9!" ;$$

BIOREMEDIATION JOURNAL
2023, VOL. 27, NO. 4, 368–381
https://doi.org/10.1080/10889868.2022.2130873

Check for updates

Bioremediation of acid mine drainage contaminated soils using bioattenuation, wastewater and air-injection system

Ifeanyi Michael Smarte Anekwe and Yusuf Makarfi Isa

School of Chemical and Metallurgical Engineering, University of the Witwatersrand, Johannesburg, South Africa

ABSTRACT

Acid mine drainage (AMD) poses a serious challenge to the ecosystem due to its disastrous effect on soil and water resources which contributes to the pollution and reduction of the accessibility of potable water and arable land for agricultural activities. The treatment of contaminated soils using physical, chemical, or thermal methods requires some improvements to meet the required remediation purpose. The purpose of this study is to investigate the application of bioattenuation, wastewater, and air-injection for the remediation of AMD polluted soils. A microcosm comprising 1 kg of polluted soil was amended with varying loadings of domestic and brewery wastewaters for biostimulation treatments, and the bioattenuation received no amendment while the bioventing treatment was supplied with wastewaters and atmospheric air at 3 L/min for 30 mins intervals every 48 h. The investigation conducted under the ambient condition for 28 days recorded average metal removal efficiencies of 12–31% (bioattenuation), 50–66% for all heavy metals (iron, aluminum, copper and zinc) except manganese (28%) (biostimulation) and 56–60% (bioventing) while 32%, 35% and 52% sulfate removal efficiencies were recorded for bioattenuation, bioventing and biostimulation respectively. The study revealed that wastewater and bioventing systems can be efficiently utilized as an eco-friendly approach for the treatment of acid mine contaminated soils and pollution mitigation in the mining environments. In addition, from a comparative point of view, the results of the study further showed that the amendment of wastewater is more effective for sulfate removal while air injection enhanced metal removal from acid mine drainage contaminated soils. The synergetic effects have also been observed; the current study demonstrates that the combined application of biostimulation and bioventing system requires constant monitoring to ensure the provision of appropriate anoxic conditions for biosulfidogenic activity by enhancing sulfate removal while promoting an oxic environment for the oxidation process to facilitate metal removal for the effective treatment of acid mine drainage contaminated soil.

KEYWORDS
Acid mine drainage; bioattenuation; biostimulation; bioventing; wastewaters

Introduction

Industrialization has contributed immensely to the development and advancement of technologies worldwide poised to enhance the production of goods and services for the satisfaction of human wants which improves the living standard. It is indisputable that most anthropogenic, agricultural and industrial activities (mining and exploration of mineral resources) continue to play an integral part in the economic growth, development, and sustainability of most countries like South Africa and many other countries that are actively engaged in mining of mineral resources which account for their major exports and income. These activities have continued to rise with the growth of the human population and do not seem to retard in future. While these have brought a significant amount of wealth to individuals and government, it is worth noting that this "progress" has come at the price of extensive ecological damage because of little or no strategy to effectively manage the direct and indirect effects of mine waste contaminants. Despite efforts by man to move toward sustainable processes in the nearest future, it is a known fact that the very much desired transition will take place at a price. As we prepare toward ensuring the price has been met, the status quo has to be

CONTACT Yusuf Makarfi Isa Yusuf.Isa@wits.ac.za School of Chemical and Metallurgical Engineering, University of the Witwatersrand, Johannesburg 2050, South Africa.

2022 Taylor & Francis Group, LLC

explored in a very responsible manner such that any potential damage to the environment is minimal. Mine waste is among the major soil and water pollutants that contribute to the degradation and reduction of agricultural arable lands and the availability of potable water. This causes contamination of proximate water bodies and physical soil deterioration through soil compaction, sealing of pores, reduction in root penetration, low infiltration, waterlogging and runoff, and accelerated erosion (Osman 2014). The negative impact as a result of this contamination has directly and indirectly affected both aquatic and terrestrial organisms. Furthermore, most of the areas affected by these pollutants do not have immediate access to alternative solutions considering their remote locations. Notably among the pollutants from the mining sector is acid mine drainage (AMD). Although AMD is attributed to various discharges from the mining industry, it is important to note its varied composition as such it can come in various acidic strengths as well as the composition of heavy metals. The myriad nature of its composition clearly implies a very vast impact on both the area of discharge and organisms that are very much dependent on the discharge areas such as water and soils among others.

Soils are the source of primary deposits for metal pollutants in terrestrial habitats, while sediments are the main deposit for heavy metals in marine ecosystems. Heavy Metals can also be found in organic fragments, mostly in an encapsulated state, with some metal recovery taking place because of the degradation of organic materials in the soil. The aqueous process offers a mobile platform for metal transport and redistribution through soil and animals, as well as in the marine ecosystem (Warren and Haack 2001). The marine ecosystem also includes a large number of heavy metals existing in several chemical phases with most of the heavy metals settling in sediments (Warren and Haack 2001; Gilmour and Riedel 2009). While some of these elements could be detrimental to the ecosystem, it is important to note that many of them play vital roles in different sectors of the economy including agriculture among others. Some elements such as carbon (C), Hydrogen (H), Nitrogen (N) Oxygen (O), Phosphorous (P) and Sulfur (S) account for most nutrients in the plant, animal, and microbial biomass; besides, many other elements are commonly found in organisms, the majority of which have basic biochemical and physiological functions such as Potassium (K), calcium (Ca), Magnesium (Mg), Boron (B), Chlorine (Cl), Iron (Fe), Manganese (Mn), Cupper (Cu), Zinc (Zn), Molybdenum (Mo), Nickel (Ni), Cupper (Co), Selenium (Se), Sodium (Na) and Tin (Sn). Also, plants, animals, and microbes may contain approximately 90 known natural elements, including gold (Au), Arsenic (As), Mercury (Hg), cadmium (Cd) & Uranium (Ur). Any of these heavy metals can end up in food and the ecosystem as pollutants. Metal pollutants can influence the terrestrial and aquatic environment via different means such as runoff, leaching, and mobile colloid transport (Adriano 2013). The adverse effect of some of these metals could be lethal leading to losses and damage to the ecosystem, in addition, these metals could have had economic value hence, their discharge in effluent streams does not only have environmental and health impacts but also could pose economic challenges. Metalloids, organometals, and organometalloids are pollutants linked to heavy metals and other radionuclides that penetrate the ecosystem. However, metals are important inherent constituents of all soils, where they comprise a collection of possible mobile metallic ions, plant and microbe vital nutrients and solid materials (such as clays, minerals, iron and manganese, etc) that can significantly alter the biogeochemical activities of soil (Huang et al. 2004). The weathering and degradation of minerals and rocks are attributed to physical, biological, and chemical processes and for biodegradation, metal-microbe associations are often involved. Bioweathering involved microbial activities for rock and mineral degradation. The great majority of minerals contain heavy metals, and there is an enormous number of mineral forms associated with each heavy metal, with varying environmental distribution and physicochemical characteristics. For instance, Mn is present in more than 100 naturally prevalent mineral deposits, mostly as oxides, carbonates and silicates, as a main or minor constituent (Ehrlich 1998). Moreover,

heavy metals can be a standard part of several minerals such as natural or industrial impurities (silicates, sulfides, oxides, carbonates, and phosphates) that can contain possibly hazardous or other heavy metals. No matter if they are of geological or biogenic source, microorganisms can associate with minerals and influence their structural design through mechanical and biochemical processes which alter heavy metal speciation and mobility (Ehrlich 1996; Landa 2005; Gleeson, McDermott, and Clipson 2007).

Weathering of Fe-minerals on the surface (rocks, soils), and subsurface (sediments) zone is aided by bacterial and fungal activities, as well as chemical reactions (Kashefi and Lovley 2000). Sulfate-reducing bacteria contribute significantly to the production of some sulfide minerals, particularly Fe-pyrite (FeS2), in some sedimentary deposits. Bacteria such as Acidithiobacillus ferrooxidans, and Leptospirillum ferrooxidans can oxidize metal sulfides. Bacterial action can entail either a direct oxidative effect on the crystalline structure of a metal sulfide or an indirect oxidative effect through the production of acid ferric sulfate, for the chemical oxidation of the metal sulfide. In the solubilization of uraninite, for instance, the indirect process plays a crucial role. Metals are extracted from low-grade metal sulfide ores and uraninite using bacterial oxidation of metal sulfides. The oxidation of FeS_2 by these bacteria in exposed old, abandoned or active bituminous coal seams or gold mines is an environmentally hazardous process that contributes to acid mine drainage (AMD) (Ehrlich 1998; Rawlings, Dew, and du Plessis 2003; Rawlings 2005; Ehrlich, Newman, and Kappler 2015).

Acid mine drainage (AMD) is generated through pyrite oxidation which is triggered by the exposure of mine water containing pyrite to oxygen. This exposure leads to a series of chemical reactions as shown with Equations 1–5. While acid mine drainage could vary in composition based on its origin, it has been shown to exhibit high sulfate content, low pH, heavy metals, and sometimes, the presence of radioactive elements such as Uranium (Ur) and Radium (Ra) depending on the type of mine site (Diels and Vanbroekhoven 2008). Pyrite weathering, which is the initiation phase in the overall AMD production is represented using Eq. 1 where oxidation of pyrite (FeS_2) forms ferrous iron (Fe^{2+}) and sulfate. Subsequently, the ferrous iron is converted to ferric iron (Fe^{3+}) (Eq. 2); this step is facilitated by the presence of bacteria. Hydrolysis of ferric iron to form ferric hydroxide (which is regarded as the AMD) is represented in Eq. 3 of the weathering process which occurred at pH 3.5 and above and no precipitation is feasible at a lower pH while the oxidation of the additional FeS_2 with Fe^{3+} as the oxidizing agent (not O_2) continues until the depletion of either FeS_2 or Fe^{3+} (Eq. 4) (Lottermoser 2003). The summary of the pyrite weathering process for the generation of AMD is illustrated in Eq. 5

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

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

$$4Fe^{3+} + 12H_2O \rightarrow 4Fe(OH)_3 + 12H^+ \quad (3)$$

$$FeS_2 + 14Fe^{3+} + 8H_2O \rightarrow 15Fe^{2+} + 2SO_4^{2-} + 16H^+ \quad (4)$$

$$4FeS_2 + 15O_2 + 14H_2O \rightarrow 4Fe(OH)_3 + 8H_2SO_4 \quad (5)$$

Metal speciation in soil involves the chemical nature of the soil solution metal, as a free ion or complexed to a ligand, during the gaseous stage and dispersed in solid phases throughout the soil. These characteristics regulate the chemical activity of components, whether on the soil surface or the aquatic and contribute immensely to toxicity determination. The chemical and physical aspects defining metal speciation in soil regulate its interaction, reactivity, and fate in soil, and in certain cases, toxicity, including its solubility, and uptake. In environmental soil chemistry, the quantitative speciation and variation of metals over time are necessary for the study of metal concentrations in the soil. Although species have formed for thousands and millions of years in the presence of heavy metals in the natural world, it is only after industrialization that large concentrations of metals have consistently been added globally to soil ecosystems. The need to classify and reduce the concentration of those species in soils that pose a great challenge to organisms is crucial to minimizing high exposure of soil organics and

microbes to heavy metals toxicity (Roberts, Nachtegaal, and Sparks 2005; Reeder, Schoonen, and Lanzirotti 2006). Many cations are found in soil environments, and they struggle for the sorbents' surface sites. Sorption competition between two or more metal cations is critical for determining their relative affinity for a specific sorbent. According to Violante et al. (2013), competition between two or more cations in the absence or presence of anions is an area of interest. The effect of anions (such as SO_4^{2-} or CO_3^{2-}) on the sorption/desorption operations of metal cations on/from minerals with variable charge is more complex, as organic and inorganic ligands can interact directly with the surfaces of minerals with variable charge or metal ions in solution, altering metal ion sorption/desorption. This can result in the increased sorption of some polyvalent cations (Al, Ca, Fe, Mg, and Zn) on metal oxides in the presence of sulfate or ligands.

Many physical, geochemical, and biological processes influence metal behavior in the soil surface and subsurface environments. Much emphasis has been put on the mechanism that mobilizes or immobilizes metals because mobility ensures the availability of metals. A dissolved metal may be moved by fluid flow and ultimately join the water supply or be used in an ecosystem. The bioavailability of geomaterials in certain cases is equated with the phenomenon of dissolved metal. Metals that are linked to small mobile particles can have greater exposure potential. Colloids and airborne particles, for example, are mobile and all have metals connected to them (Roberts, Nachtegaal, and Sparks 2005; Reeder, Schoonen, and Lanzirotti 2006). The relationship between metals or metallic compounds with soil elements, including clay and other minerals, humic materials, soil colloidal particles, biogenic debris and foreign bodies, and microbes, decides metal bioaccessibility in biological ecosystems. Metal sorption to cells may significantly contribute to the overall interactions among microorganisms, metals and minerals (Burford, Fomina, and Gadd 2003), occurring over a long period. Most studies have shown microbial cells can display higher metal-sorption values than any clay minerals on a particular unit area basis. Biosorption processes likely contribute to metal speciation, bioaccessibility, and distribution in the ecosystem than originally understood (Krantz-Rülcker, Allard, and Schnürer 1993, 1996; Ledin and Pedersen 1996). Metal association with particular cell-surface categories may indeed facilitate or hinder metal transfer, conversion, and biomineralization activities.

The deployment of physicochemical methods for the remediation of AMD polluted soils has proven to be challenging and even criticized due to high operation cost, non-eco-friendly, inability to effectively meet the remediation purpose and the possibility of recontamination of the site in the course of reclamation. The need to find alternative treatment techniques that can address these major concerns should be explored and encouraged. However, bioremediation technology provides a cost-effective and eco-friendly method for the effective remediation of polluted soils using a biologically induced method. Bioremediation can take the form of organic or inorganic nutrient application (biostimulation), air injection into the treatment system (bioventing), natural attenuation (bioattenuation) or inoculation of genetically modified microorganisms or consortium (bioaugmentation) to provide sufficient energy source, ventilate the contaminated environment, control the environmental conditions or increase microbial load respectively to increase hydrogenase and facilitate the treatment of contaminated soil.

Previous studies by McCullough and Lund (2011), Costa and Duarte (2005), Greben et al. (2009), Sanchez-Andrea, Triana, and Sanz (2012) have shown that the application of organic waste like wastewaters, sludge, fertilizer animal waste, and inorganic substrates has been viable in the remediation of AMD contaminated environment. This bioremediation method also known as biostimulation uses relevant media to stimulate the contaminated soil environment and provide nutrients to the indigenous microorganisms to enhance the microbial degradation of contaminants. In contrast to biostimulation, bioattenuation is a passive biodegradation approach that is dependent on the natural environmental factors to revive and sustain the growth of the microbial communities to enhance the natural biodegradation of pollutants without human intervention

Table 1. Brewery and Domestic Wastewater properties.

Wastewater/Composition	pH	COD (mg/L)	Microbial Count (CFU/100mL)
BWW	8.2	750	1.7 x 10^6
DWW	7.9	704	9.2 x 10^4

aside from monitoring the degradation rate (Garbisu et al. 2017).

A bioventing system is designed to aerate the treatment environment to create the oxic condition by introducing O_2 using hydrogen peroxide and/or air and/or water. This ventilates the polluted environment to increase the rate of metabolism. Air injection triggers an oxidation reaction which in turn precipitates metals such as sulfide, carbonates, or hydroxide to enhance removal from the contaminated environment (Ibanez et al. 2008). However, the use of H_2O_2 has been criticized due to its low penetration power and the inhibitory role attributed to a high concentration above 100 mg/l according to Hinchee, Downey, and Aggarwal (1991) which limits the availability of O_2 to the microbial community in the vadose zone and reduced the rate of biodegradation respectively.

These bioremediation methods can be applied as a single method, incorporated or enhanced by genetic manipulation to improve the remediation process. However, irrespective of the success of bioremediation application, removal of these contaminants still depends on some factors such as temperature, pH, nutrient availability, nature and concentration of contaminants, availability of microbes, suitable bioremediation environment (aerobic or anaerobic) for microbial growth, toxicity and bioavailability of contaminants to degrading bacteria (Alexander 1999; Boopathy 2000). Moreover, adjustment and optimization of bioremediation conditions are necessary to mitigate most of these drawbacks and to provide effective treatment of the polluted sites (Awasthi et al. 2015). Owing to the aforementioned inefficiencies of physicochemical and other contaminant remediation methods, the need to develop a viable, environmentally friendly, readily accessible and cost-effective method for the remediation of contaminants becomes imperative for environmental sustainability. The objective of this study is to investigate the potential of bioattenuation, air-injection and wastewater systems as eco-friendly biostimulants for the treatment of acid mine drainage (AMD) polluted soils.

Materials and methods

The samples of soil used in this study were from the city of Durban, which is located in the province of KwaZulu-Natal in South Africa. After being exposed to air for twenty-four hours, the samples were then put through a conventional set of sieves, the highest one of which had a screen size of 2 mm. Based on the results of the separation, the soil sample was made up of 79.32% sand (2–0.02 mm), 14.71% silt (0.02–0.002 mm), and 5.97% clay (0.002 mm). Before beginning the bioremediation investigation, soil samples were stored in a moisture-free polyethylene bag. The South African Brewery (SAB) and the Local wastewater treatment facilities were responsible for providing the Brewery wastewater (BWW) and the Domestic wastewaters (DWW), respectively. The wastewaters were kept in High-Density Poly Ethylene (HDPE) containers and stored in the refrigerator at temperatures that would inhibit any microbial activity from occurring. Table 1 reports the results of the analysis of the carbon-oxygen demand (COD) and microbiological count of BWW and DWW.

Simulation of AMD-contaminated soil

This work has taken into account the non-uniform distribution of AMD in soils, it has also accounted for the need to ensure that the findings are not in any way influenced by variation in sample composition. It was therefore decided to produce representative soils in the laboratory. Acid mine drainage was replicated according to Anekwe and Isa (2021a) with metal constituents that were comparable to those found in central Witwatersrand (Humphries, McCarthy, and Pillay 2017): consisting of 25.70 zinc sulfate hydrate ($ZnSO_4.H_2O$); 10.70 copper sulfate pentahydrate ($CuSO_4.5H_2O$), 200.01 iron sulfate heptahydrate ($FeSO_4.7H_2O$), 20.00 manganese sulfate hydrate ($MnSO_4.H_2O$), 50.50 Aluminum sulfate octadecahydrate ($Al_2SO_4.18H_2O$) (mg/kg of soil). These metal sulfates were first dissolved in one liter of

deionized water, and then the pH was modified to 2.7 using sulfuric acid to provide the necessary acidic medium for acid mine drainage. The mixture was agitated at 200 revolutions per minute (rpm) for 60 minutes to ensure that the metal sulfates were homogenized. In order to create soil that was contaminated with acid mine drainage, simulated mine water was combined with the soil sample, and then the acid mine drainage-tainted soil was agitated at a speed of 180 rpm to ensure that the acid mine drainage solution was distributed evenly throughout the soil sample. Before the administration of bioremediation treatments, the contaminated sample was dried and allowed to sit undisturbed for forty-eight hours. This was done to allow for aging and to simulate the conditions of the actual contaminated soil environment.

Biostimulation (BST) and bioattenuation (BAT) studies for AMD-contaminated soil

Biostimulation and bioattenuation treatments for acid mine drainage were carried out in two sets of five plastic bioreactors designated as BST and BAT to represent biostimulation and bioattenuation respectively. Each bioreactor was charged with 1 kg of acid mine drainage contaminated soil samples. The five bioreactors for BST were amended with different ratios of the brewery (BWW) and domestic waste (DWW) each as shown in Table 2 The bioreactor for BAT treatment was not amended with wastewater effluents which also served as the control treatment.

Bioventing studies for AMD contaminated soil

The bioventing investigation for the treatment of AMD polluted soil was conducted in 4 plastic bioreactors with similar contents and designated as BVT. Each bioreactor contained 1 kg of acid mine drainage polluted soil. One bioreactor received simply air injection while the other three bioreactors received varied ratios of BWW, DWW, and ambient air as amendments. In order to ensure that there was sufficient air circulation all around the bioreactors, atmospheric air was pumped in via the vadose unsaturated zone into the bioreactors using an air compressor pump.

Table 2. Composition of biostimulation (BST), bioventing (BVT) and bioattenuation (BAT) treatment systems for AMD contaminated soil.

Bioreactors	BWW (g/kg)	DWW (g/kg)	ATM-Air (3L/min)	Amendments (Ratio)
BST-1	100	0	–	4:0
BST-2	0	100	–	3:1
BST-3	50	50	–	2:2
BST-4	25	75	–	1:3
BST-5	0	100	–	0:4
BVT-1	0	0		0:0
BVT-2	100	0		0:4
BVT-3	0	100		0:0
BVT-4	50	50		4:0
BAT	0	0	–	4:0

Since bioventing will be more cost-effective if the lowest flow rate and largest flow interval are taken into consideration as reported by (Thom et al. 2014), atmospheric air was provided at a flow rate of 3 liters per minute for 30 minutes every 48 hours.

Analytical procedure

The carbon-oxygen demand (COD) was quantified by the standard method utilizing a COD reactor (Hach) and Spectrophotometry (DR 3900) (APHA 1995) while an electronic pH meter was employed to determine the pH of samples. Soil pH was measured according to Peech et al. (1947): 20 g of homogenized soil sample was mixed with 20 ml of distilled water (1:1 w/v) in a 50 ml beaker and the suspension was stirred for 60 minutes. The microbiological count was determined by the vapor phase transfer procedure according to Amanchukwu, Obafemi, and Okpokwasili (1989).

Heavy metals and sulfate analyses

To avoid heavy metal degradation and loss, soil samples were dried in an oven set at 35 degrees Celsius. A sample of 5 g of dry soil was pulverized with a mortar and pestle to achieve a fine texture, and then it was allowed to run through a sieve with a mesh size of 63 micrometers in order to eliminate any rocks, pebbles, or stick particles. Subsequently, Scanning electron microscopy (SEM) and Energy-dispersive X-ray spectroscopy (EDXS) using Zeiss Sigma 300 VP equipped with a field emission gun and Oxford instruments x-act PentaFET precision respectively while the X-ray fluorescence (XRF) analysis using Spectro X-

Figure 1. (a) AMD soil Biostimulant (wastewater). (b) Control Treatment (BAT). (a) shows the formation of black ppt. on the surface of the samples amended with wastewater as a result of biosulfidogenic process while in (b) (control treatment), no black ppt. was formed due to lack of organic amendment.

Lab 2000 XRF equipment were conducted on the samples to determine the qualitative and quantitative composition of heavy metals in the soil as reported in the work of Baranowski, Rybak, and Baranowska (2002). Aqueous Extraction was used to extract residue heavy metals from the soil for sulfate analysis according to Walker (1972). 15 g of homogeneous soil samples were weighed out after which 150 mL of deionized water was added to the weighed-out sample. The mixture containing the sample was then shaken at 300 rpm for 30mins and filtered. The sample was mixed with reagent via the instrument while SO_4^{2-} ion was precipitated with $BaCl_2$ in an acidic medium and color was detected at 420 nm with a Thermo scientific gallery plus discrete analyzer.

Results and discussion

Results

BST results

The BST treatment showed yielding results from the first week with the presence of black precipitate on the surface of the samples as can be seen in Figure 1a for all the BST treatments amended with wastewaters except for bioattenuation (BAT) control treatment which received no amendment as shown in Figure 1b. Biostimulation (BST-1) treatments recorded 51.12% average removal efficiencies as the changes in Cu concentration showed significant removal (65.69%) while Mn was least removed at a value that was a little over 20% (Figure 2a) after 28 days treatment period. Interestingly enough, the treatment (BST-1) showed an increase in sulfate removal efficiency from week 1 to week 4 (13.20% to 47.10% respectively) (Figure 2g) it is worth noting that while the removal might not have been proportional to time, it increased over the four weeks of study. Also, in treatment BST-2 which recorded 52.75% average removal efficiency, Cu was mostly removed while Mn was least removed with 71.26 and 25.36% removal efficiencies respectively (Figure 2b). An average metal removal efficiency of 58.50% was recorded by BST-3, with individual metal removal efficiencies of 66.17, 59.38, 69.77, 60.53 and 36.41% for iron, aluminum, copper, zinc and manganese respectively (Figure 2c). BST-2 and BST-3 treatments resulted in a substantial decrease in the sulfate removal efficiency from week 1 to week 2 (50.90 to 30.21% and 47.42 to 32.10% respectively) but later improved appreciably to 47.91%.and 50.82% in week 4 accordingly (Figure 2g). The average metal removal efficiencies of 49.38 and 49.82% were recorded by BST-4 and BST-5 treatments respectively (Figure 2d & e). While BST-4 treatment led to an increase in sulfate removal efficiency from week 1 to week 4 with values of 37.0% to 53.4%, BST-5 treatment recorded a slight increase from week 1 (29.50%) and climax in week 3 (52.1%) with a decline to 33.65 in week 4. The BAT treatment showed the lowest metal removal efficiencies of 12-33% with the least average metal and sulfate removal efficiencies of 22.70% (Figure 2f) and 31.56% respectively.

Figure 2. (a–f) Showed metal removal efficiencies for biostimulation (BST) and bioattenuation (BAT) treatments. (g) Sulfate removal efficiency for BST treatments.

Bioventing treatment (BVT) results

The bioventing treatments (BVT) showed a reduction in metal concentration in all treatments, implying a positive impact on the investigated parameters. BVT-1 recorded 49.16% average removal efficiency after the 28 days (Figure 3a). The sulfate removal efficiency for this treatment increased from week 1 and

Figure 3. (a–d) Showed metal removal efficiencies from BVT treatments. (e) Sulfate removal efficiency for BVT treatments.

climaxed in week 3 before taking a downward trend between weeks 3 and 4, the decline recorded a change of 3.4% which translates to about 26.86% removal efficiency (Figure 3e). BVT-2 and BVT-3 recorded a general increasing pattern in removal efficiencies for all heavy metals from week 1 (59.72% and 60.67% respectively) with 65.25% and 63.56% average removal efficiencies respectively (Figure 3b & c). The sulfate removal efficiency of BVT-2 increased gradually from the first week and continued reaching 41.9% in week 4 (Figure 3e), this trend shows potential for continuous removal efficiency beyond the work treatment. The BVT-4 treatment showed an average metal removal efficiency of 70.20% with significant metal removal efficiencies of 75.78, 86.17, 93.42, 88.23, and 80.85% for Fe, Al, Cu, Zn and Mn respectively (Figure 3d). There was a reduction in weekly sulfate removal efficiencies in treatments BVT-3 (from 39.30 in the second week to 31.60% by the fourth week) and BVT-4 (from 34.90 in the second week to 26.51% by the third week), nonetheless, the change between the second and third weeks in the case of BVT-4 was not continuous as it was observed that by the fourth week, BVT-4

Figure 4. (a & b) SEM result showed cryptocrystalline coating layer of spherulitic aggregates consists of Fe, Al, O, S, microelements, and minor amounts of other metals. (from BST & BVT).

appreciated by 5% in whereas efficiency in BVT-3 slightly decreased further by 3% (Figure 3g).

Scanning electron microscopy (SEM)-energy dispersive X-ray spectroscopy (EDS) results and discussion

In order to have a comprehensive knowledge of the structures of the samples and any changes resulting from the various treatments, they were exposed to scanning electron microscopy (SEM). The surface of samples from both treatments (BST and BVT) was coated with a pervasive layer, based on SEM study (Figure 4a & b); further analyses show that the samples contain Fe, Al, S, and O, microelements and little quantity of other heavy metals as shown in the peaks of EDS spectrum (Figure 5a & b). These peaks can be attributed to the composition of the AMD and their treatment systems; this further demonstrates that Fe, Al, and other heavy metals could have been precipitated in the form of metal sulfides. It can be deduced (from the SEM-EDS results) that the earlier observed black precipitate (Figure 1a) formed on the surface of the treatment is attributed to the biosulfidogenic process similar to findings made by Pawlowska and Sadowski (Pawlowska and Sadowski 2019) which reduces sulfate to elemental sulfur to facilitate the formation of metal sulfide precipitate. (Dvorak et al. 1992; Lyew and Sheppard 1997; Gibert et al. 2003). However, metal complexation and precipitation were feasible during the treatment due to the formation of amorphous Fe-Al (oxy) hydroxide precipitate since the presence of Fe, Al, S, and O was evident as shown in Figure 4a & b which contributed to co-precipitation and removal of heavy metals from the treatments (Song et al. 2001; Gibert et al. 2005; Cravotta 2007).

Figures 5 a, b & c allow for a comparative assessment of the various treatment techniques, the figures, a, b and c show the elements detected using the energy dispersive spectroscopy (EDS). The results indicate the absence of some metals compared to the BAT treatment, indicating the effectiveness of the BST and BVT treatment systems for metal removal.

Discussions

Biostimulation study

Heavy metal removal in this study was triggered by the biosulfidogenic activity promoted by sulfate-reducing bacteria (SRB), which reduce sulfate to biogenic hydrogen sulfide while utilizing organic substrate (brewery and domestic wastewaters) as an energy source as represented in Eq. 6a. The biogenic H_2S subsequently reacts with heavy metals in the treatment system to form metal sulfides as shown in Eq. 6b (Marchioretto, Bruning, and Rulkens 2005; Lu et al. 2011; Nancucheo and Johnson 2012), by adding alkalinity in the form of HCO_3^-, while lowering dissolved SO_4^{2-} concentration and increasing the pH of the system to enhance remediation of AMD (Gibert et al. 2003; Bhagat et al. 2004). This process can only be feasible if the environment is amended with organic substrates to foster

Figure 5. (a) BST treatment system. (b) BVT treatment system. (c) BAT treatment system.

microbial activities of reducing bacteria (Hammack, Dvorak, and Edenborn 1993) which results in the occurrence of black precipitate (Figure 4a) as visible on the surface of the samples amended with wastewater (Figure 1a) after week 1 except for the bioattenuation (BAT) treatment without black precipitate on the top layer (Figure 1b) which suggests the absence of organic amendment (Bai et al. 2013). The formation of black precipitate within the first week of treatment could be attributed to the increase in metabolic activities of reducing bacteria present in the wastewater which enhanced sulfate removal and a concomitant decrease in heavy metals concentration. In the biosulfidogenic process, certain bacteria are capable of reducing SO_4^{2-} and other sulfur oxyanions to H_2S when supplied with a suitable carbon source (Postgate 1984).

$$2CH_2O + SO_4^{2-} \rightarrow H_2S + 2HCO_3^- \quad (6a)$$

$$H_2S + Me^{2+} \rightarrow MeS_{(s)} + 2H^+ \quad (6b)$$

The oxidation reaction that initially creates the AMD with heavy metals in different concentrations is reversed by the precipitation of heavy metals as sulfide, which is a recurring activity (Skousen et al. 2017). With the biosulfidogenic activity, heavy metals are reduced via precipitation as sulfides, hydroxides, or carbonates (Martins et al. 2011; Bai et al. 2013; Deng and Lin 2013; van den Berg et al. 2016). Aluminum precipitates at pH greater than 5 while Manganese removal is a complicated process

because of the variable oxidation state but is generally dependent on the oxidation number of cations at pH of > 7. Sometimes, a high pH (around 10.5) is needed for the complete removal of Mn as recorded by Balintova and Petrilakova (2011) which can be attributed to the low removal of Mn in the present investigation with an average removal efficiency of 27.92% and 12.82% from BST and BAT treatment respectively. Also, the uncatalyzed oxidation of Mn contributes to its low removal efficiency as Mn is required to be oxidized from soluble Mn(II) to insoluble Mn(IV) in order to facilitate removal as MnO_2 precipitate (Johnson and Hallberg 2005; Sanchez-Andrea, Triana, and Sanz 2012). For Al and divalent transition metals, sorption to organic materials is important, whereas sorption to precipitated iron and manganese contributes to the removal of trace metals (Zachara, Cowan, and Resch 1991). However, the removal of divalent metals (Cu, Fe, Zn) during the treatment was feasible by precipitation as sulfide minerals after microbial sulfide reduction (Stumm and Morgan 1996) as shown in Eq. 7 for Fe which also accounts for the appreciable removal of other heavy metals (> 50%) when compared to Mn. Additional processes for metal removal in the AMD treatment include co-precipitation with these precipitates (Cravotta and Trahan 1999; Song et al. 2001), biosorption into the organic substrates used for the treatment purpose (Machemer and Wildeman 1992; Songet al. 2001; Gibert et al. 2003; Willow and Cohen 2003), exchange to precipitated Fe, organic materials, and soil-like materials (Skousen et al. 2017), while Fe and Al precipitate as hydroxides (Gilbert et al. 2003).

$$Fe^{2+} + H_2S + 2HCO_3^- \rightarrow FeS + 2H_2O + 2CO_2 \quad (7)$$

The BST-1 (BWW only) treatment recorded slightly higher removal efficiency when compared to the BST-5 (DWW only) with an average metal removal efficiency of 51.13 and 49.82% respectively, however, the BST-3 (brewery + domestic wastewaters) treatment recorded an appreciable metal removal efficiency (58.45%) which can be due to the possible synergy between the two wastewater effluents in the ratio of 2:2. This suggests that an appropriate mixture of the organic amendment is effective in AMD treatment suggesting that getting the right combination of the two treatments could be more effective than the application of a single substrate (Neculita, Zagury, and Bussière 2007; McCullough and Lund 2011). It should however be noted that brewery wastewater contains a substantial amount of COD than domestic wastewater owing to the presence of organic materials in high quantity which accounts for the significant microbial loads (as shown in Table 1) and attributes to the appreciable removal efficiencies recorded with this amendment (Jaiyeola and Bwapwa 2016; Amenorfenyo et al. 2019).

In accordance with the present BST investigation which recorded appreciable average metal removal efficiencies of 59%, 50%, 66%, and 56% for iron, aluminum, copper, and zinc (with the exception of Manganese 28%) after 4 weeks with an average pH of 7.2, the study by Hughes and Gray (2013) recorded removal efficiencies of 52–84%, 47–61%, 74–86% and 58–90% for aluminum, copper, iron and zinc respectively in the co-treatment of AMD with municipal wastewater after 39 days treatment. Moreover, Strosnider et al. (2013) recorded removal efficiencies of 99%, 99% 73% and 13% for aluminum, iron, zinc and manganese respectively with final pH of 6.79 after 19 weeks using an organic substrate while Sanchez-Andrea, Triana, and Sanz (2012) recorded > 85% heavy metal removal efficiencies for Fe, Co, Cu, Zn respectively (except for Mn 13%) after 183 days treatment period. However, in contrast to the low Mn removal recorded in this investigation, the study by Silva et al. (2012) reported 99% Mn removal after 2 weeks study period achievable with carbonate ions (introduced as limestone or sodium carbonate) which provided the pH within the alkaline range preferably above 9.0 for the precipitation and removal of Mn.

Bioventing study

The removal of metal occurred through the oxidation process for the BVT study. When the bioreactor was vented, the oxidation process was activated (facilitated by native microbes in the

brewery and domestic wastewaters), which resulted in the oxidation of Fe^{2+}, the main constituent of the AMD was oxidized to Fe^{3+}. The interactions that take place between the microbes, oxygen, and AMD, in addition to the presence of alkalinity (in form of wastewater), facilitated the oxidation process in the treatment system (Berghorn and Hunzeker 2001) which accounts for significant metal removal. Ackman and Kleinmann (1984) noted that the introduction of alkalinity into the aeration system invariably increases microbial activities and the rate of oxidation through neutralization of the AMD (Vadapalli et al. 2015) which is evident in the current investigation where treatment with wastewater combined with bioventing (BST-2, 3, 4) recorded greater than 15% increase in average removal efficiency in contrast to the treatment with air-injection only (BST-1). Precipitation occurred over the range of pH for various heavy metals due to the gradual oxidation of Fe^{2+} to Fe^{3+} as a result of oxygen (O_2) injected into the system and its precipitation in the form of $Fe(OH)_3$ as shown in Eq. 8 which enhance Fe removal alongside other heavy metals such as copper, Zinc, Aluminum and Manganese via co-precipitation with iron (Ackman and Kleinmann 1984; Jones et al. 2014). However, unlike the BST treatment, Mn oxidation was feasible where soluble Mn^{2+} was oxidized to insoluble Mn^{4+} to facilitate Mn removal as MnO_2 (Eq. 9) which results in the average removal efficiency of 58.28% as against 27.92% recorded in BST treatment. This process can be sensitive and sluggish at low pH (acidic medium) as reported by Nijjer, Thonstad, and Haarberg (2000), or in the presence of Fe^{2+} which either hinders the process or triggers reverse oxidation (Skousen et al. 2017), however, bacteria and surface catalysis can accelerate the process (Stumm and Morgan 1996). Hence, the combined oxidation and precipitation process are responsible for the significant Mn removal efficiency recorded with treatments amended with wastewaters (BWW and DWW) and vented (BVT-2, BVT-3 and BVT-4) with > 60% while BVT-1 (only vented) recorded < 40% average removal efficiency. The BVT treatment showed effective Mn removal when compared to the BST treatment.

$$Fe^{2+} + 0.25O_2 + 2.5H_2O \rightarrow FeOOH_3 + 2H^+ \quad (8)$$

$$Mn^{2+} + 0.5O_2 + H_2O \rightarrow MnO_2 + 4H^+ \quad (9)$$

It is worth noting that reacting parameter always affects the possible reactions. While the reactor system was ventilated, it is worth noting that the ventilation was not continuous throughout the process. Since the bioreactor was not subjected to constant ventilation, other potential parameters that could have affected the process should be considered. Another process that enhances the removal of heavy metals is the biosulfidogenic process which can be favored in the presence of organic substrates and this case, in the form of BWW and DWW, as evident in BST treatment (Marchioretto, Bruning, and Rulkens 2005; Muyzer and Stams 2008; Lu et al. 2011; Nancucheo and Johnson 2012) which involves metal precipitation as shown in Figure 4b. However, sorption to organic materials also passively contributes to the removal of heavy metals (Zachara, Cowan, and Resch 1991). It, therefore, implies that there could have been other parameters that contributed to the reactor performance in addition to bioventing.

With the metal removal efficiencies of 60%, 57%, 69%, 66% and 58% for iron, aluminum, copper, zinc, and manganese respectively over 4 weeks which recorded a pH of 7.2, the present BVT study is in agreement with the study by Balintova and Petrilakova (2011) which recorded removal efficiencies of 97%, 95%, 93%, 89% and 89% for iron, copper, aluminum, manganese and zinc respectively after 3 days where O_2 was introduced into the system using H_2O_2 to induced oxidation of Fe^{2+} to Fe^{3+} followed by precipitation of metal hydroxides from the AMD using NaOH (which introduces alkalinity into the system). It can be seen that although the works were carried out under similar conditions, the difference in results could be due to the introduction of other parameters which were not investigated in the present work. Nonetheless, in both studies, the removal of copper showed an efficiency of more than 90%. According to the results obtained, it can be inferred that the conditions of the present experiment were not as favorable as those in the work of Balintova and Petrilakova

(2011) for the removal of iron, Aluminum and Zinc despite being conducted at a lower reaction time; this further confirms that other factors other than time could play critical roles during metal removal in bioventing. Other similar works like that of Cravotta (2007) reported increased oxidation (of Fe^{2b}) after the bioreactor was aerated with an appreciable metal removal efficiency of > 90% for iron and > 80% for aluminum and manganese after 24 hours.

Sulfate removal

The removal of sulfate in a very low level of oxygen supplies (BST treatment) facilitates the biosulfidogenic activity that reduces sulfate to sulfide to improve metal sulfide (black) precipitate formation (Chang, Shin, and Kim 2000; Bhagat et al. 2004). The modification of the treatment system with organic nutrients (BWW and DWW) aided this process by providing nutrients for the SRB to speed up the sulfate removal process, these findings were also well echoed in other works (Gibert et al. 2003; Kaksonen, Riekkola-Vanhanen, and Puhakka 2003; Bhagat et al. 2004). BST treatments were however more effective than BVT in sulfate removal, as a result of the anaerobic condition created by BST bioreactors which affirmed that the sulfate removal process is favored by anoxic conditions (Ford and Cohen 1992). This is evident in the current investigation as the same treatment BST-2 (amended with BWW) with 47% removal efficiency reduced by 5.2% when the air was supplied in BVT-2 and the average sulfate removal efficiency of 52% (BST) reduced to 35% (BVT) while bioattenuation (BAT, or control) recorded 31% removal efficiency due to lack of organic substrate. Consequently, the oxic condition caused by air injection in BVT treatment systems affects the sulfidogenic mechanism, reducing the actions of SRB, resulting in low sulfate removal in all bioventing systems when compared to BST treatments (Jørgensen 1994; Liu et al. 2015; Anekwe and Isa 2021b).

Furthermore, to substantiate the influence of the aerobic environment in sulfate removal, it can be deduced that the removal efficiencies of biostimulation treatment with brewery wastewater (BST-1), domestic wastewater (BST-5) and a combination of the two wastewaters (BST-3) recorded increased weekly removal efficiency when compared to corresponding bioventing treatments (BVT-2, BVT-3 and BVT-4 respectively). This could be due to the air-injection that provided sufficient O_2 in the bioventing system and consequently reduced SRB activities, as these bacteria that facilitate sulfate removal function better in a very low O_2 environment, as reported by Anekwe and Isa (2021a). The present investigation agrees with studies by Lukova et al. (2016), Muhammad et al. (2015), Benner et al. (2002), Chang, Shin, and Kim (2000) which recorded sulfate removal efficiency of 12–66% with the organic amendment as an energy source while the sulfate removal efficiency decreased from 68% recorded under reduced oxic conditions to 45% at a high oxygen concentration in the system, as reported by Ford and Cohen (1992).

Comparative study

The biostimulation (BST) treatment through the biosulfidogenic process recorded 51.61% (range: 27-66%) average metal removal efficiency as Fe, Al, Cu and Zn showed > 50% while Mn recorded the least average removal efficiency of 27.92% due to the uncatalyzed Mn oxidation after the 28 days treatment (Figure 6). However, the introduction of atmospheric air for the BVT treatment provides oxygen to the bioreactor which triggers the oxidation process to facilitate metal removal, while the amendment of wastewater to the BVT system enhanced the metal removal efficiency when compared to the treatment that was vented only (without wastewater amendment). BVT treatment showed average metal removal efficiency of 62.12% (range: 56-69%) with 56% removal efficiency for all heavy metals including Mn due to effective oxidation and precipitation processes (Figure 6). While the Mn removal efficiency changed among the three treatment methods, it is worth noting that in both scenarios (BST and BAT) where the removal efficiency for Mn was less than 50%, Mn was removed the least compared to other metals. The bioattenuation (BAT) showed the least average metal removal efficiency of 22.70% due to a lack of

Figure 6. Evaluation of BST, BVT and BAT for metal removal from AMD contaminated soils. Bars indicate the average of five replicates while error bars show the standard deviation.

Figure 7. Evaluation of BST, BVT and BAT for sulfate removal from AMD contaminated soils. Bars indicate the average of five replicates while error bars show the standard deviation.

biostimulants. However, the average sulfate removal efficiency for BST was 17.08% (135.52 mg/kg) greater than BVT and 20.05% (159.2 mg/kg) greater than BAT (control) treatment (Figure 7) which recorded the least sulfate removal. The biostimulation and bioventing treatments were viable in sulfate and heavy metal removal respectively. Despite the low metal removal in bioattenuation treatment, it is also worth noting that the system is effective, however like in most natural processes, the reactions here will require much longer periods as such they are likely to be most effective over very long periods. While time may be crucial in this research, we believe that bioattenuation is worth considering when sustainable processes are to be considered.

Conclusion

The challenges posed by acid mine drainage (AMD) are enormous and continuous due to the industrial growth and mining activities in many countries, these challenges have impacts on both human health and the environment. With a view to finding lasting solutions to the threats caused by AMD, it is necessary to explore viable solutions toward ensuring the safe discharge of the wastes from the mining industries. Various methods of treating contaminated soils have been considered and their performances have shown to be factors of the treatment conditions. It is interesting to note that all bioremediation methods were effective in metal removal from simulated soils. These bioremediation treatments were able to reduce metal and sulfate concentration in Acid mine drainage (AMD) contaminated soil but biostimulation treatment recorded appreciable sulfate removal with 17.08% and 20.05% higher than bioventing (34.53%) and bioattenuation (31.56%) treatments respectively. This implies that the stimulants used in this study are very promising biostimulants in soil treatment. From our findings, efforts to boost sulfate removal efficiency through air-injection were not very effective; this we suggest is a result of high oxygen concentration which hindered SRB activities. Metal removal was most favorable with BVT treatment which showed significant removal of all heavy metals with 10.89 and 39.42% efficiencies greater than BST and BAT treatments, respectively. Based on the findings, it is evident that sulfates and metals are removed differently. The results of this study showed that the amendment of wastewater is more effective for sulfate removal while air injection enhanced metal removal from AMD polluted soils. This study demonstrates that the combined application of biostimulation and bioventing system requires constant monitoring to ensure the provision of appropriate anoxic conditions for biosulfidogenic activity (to enhance sulfate removal) while promoting an oxic environment for the oxidation process (to facilitate metal removal) for the effective treatment of AMD polluted soil. While this study has been conducted on a laboratory scale, it is evident that upscaling this could tremendously assist in tacking the ever-growing challenges faced as a result of waste discharges from mines. The proposed techniques do not require sophisticated equipment or resources as such exploring the

methods will be both economically and environmentally friendly.

Disclosure statement

No potential conflict of interest was reported by the authors.

References

Ackman, T. E, and R. L. P. Kleinmann. 1984. In-line aeration and treatment of acid mine drainage. Washington DC: US Department of the Interior, Bureau of Mines.

Adriano, D. C. 2013. Trace elements in the terrestrial environment. New York: Springer Science & Business Media.

Alexander, M. 1999. Biodegradation and bioremediation. Oxford, United Kingdom: Gulf Professional Publishing.

Amanchukwu, S., A. Obafemi, and G. Okpokwasili. 1989. Hydrocarbon degradation and utilization by a palm-wine yeast isolate. FEMS Microbiology Letters 57 (2):151–4. doi: 10.1111/j.1574-6968.1989.tb03290.x

Amenorfenyo, D. K., X. Huang, Y. Zhang, Q. Zeng, N. Zhang, J. Ren, and Q. Huang. 2019. Microalgae brewery wastewater treatment: potentials, benefits and the challenges. International Journal of Environmental Research and Public Health 16 (11):1910. doi: 10.3390/ijerph16111910

Anekwe, I. M. S, and Y. M. Isa. 2021a. Comparative evaluation of wastewater and bioventing system for the treatment of acid mine drainage contaminated soil. Water-Energy Nexus 4:134–40. doi: 10.1016/j.wen.2021.08.001

Anekwe, I. M, and Y. M. Isa. 2021b. Wastewater and bioventing treatment systems for acid mine drainage contaminated soil. Soil and Sediment Contamination: An International Journal 30 (5):518–31. doi: 10.1080/15320383.2020.1863909

APHA, A. P. H. A., American Water Works Association. 1995. Standard methods for the examination of water and wastewater. In Standard methods for the examination of water and wastewater, 1000. Washington, DC: American Public Health Association.

Awasthi, M. K., A. K. Pandey, P. S. Bundela, and J. Khan. 2015. Co-composting of organic fraction of municipal solid waste mixed with different bulking waste: Characterization of physicochemical parameters and microbial enzymatic dynamic. Bioresource Technology 182:200–7. doi: 10.1016/j.biortech.2015.01.104

Bai, H., Y. Kang, H. Quan, Y. Han, J. Sun, and Y. Feng. 2013. Treatment of acid mine drainage by sulfate reducing bacteria with iron in bench scale runs. Bioresource Technology 128:818–22.

Balintova, M, and A. Petrilakova. 2011. Study of pH influence on selective precipitation of heavy metals from acid mine drainage. Chemical Engineering Transactions 25:1–6.

Baranowski, R., A. Rybak, and I. Baranowska. 2002. Speciation analysis of elements in soil samples by XRF. Polish Journal of Environmental Studies 11 (5):473–82.

Benner, S., D. Blowes, C. Ptacek, and K. Mayer. 2002. Rates of sulfate reduction and metal sulfide precipitation in a permeable reactive barrier. Applied Geochemistry 17 (3): 301–20. doi: 10.1016/S0883-2927(01)00084-1

Berghorn, G. H, and G. R. Hunzeker. 2001. Passive treatment alternatives for remediating abandoned-mine drainage. Remediation Journal 11 (3):111–27. doi: 10.1002/rem.1007

Bhagat, M., Burgess, J. E. Antunes, A. P. M. Whiteley, C. G, and Duncan, J. R. 2004. Precipitation of mixed metal residues from wastewater utilising biogenic sulphide. Minerals Engineering 17 (7–8):925–32. doi: 10.1016/j.mineng.2004.02.006

Boopathy, R. 2000. Factors limiting bioremediation technologies. Bioresource Technology 74 (1):63–7. doi: 10.1016/S0960-8524(99)00144-3

Burford, E., M. Fomina, and G. Gadd. 2003. Fungal involvement in bioweathering and biotransformation of rocks and minerals. Mineralogical Magazine 67 (6):1127–55. doi: 10.1180/0026461036760154

Chang, I. S., P. K. Shin, and B. H. Kim. 2000. Biological treatment of acid mine drainage under sulphate-reducing conditions with solid waste materials as substrate. Water Research 34 (4):1269–77. doi: 10.1016/S0043-1354(99)00268-7

Costa, M. C, and J. C. Duarte. 2005. Bioremediation of acid mine drainage using acidic soil and organic wastes for promoting sulphate-reducing bacteria activity on a column reactor. Water, Air, and Soil Pollution 165 (1-4): 325–45. doi: 10.1007/s11270-005-6914-7

Cravotta, C, and M. Trahan. 1999. Limestone drains to increase pH and remove dissolved metals from acidic mine drainage. Applied Geochemistry 14 (5):581–606. doi: 10.1016/S0883-2927(98)00066-4

Cravotta, C. A. 2007. Passive aerobic treatment of net-alkaline, iron-laden drainage from a flooded underground anthracite mine, Pennsylvania, USA. Mine Water and the Environment 26 (3):128–49. doi: 10.1007/s10230-007-0002-8

Deng, D, and L. S. Lin. 2013. Two-stage combined treatment of acid mine drainage and municipal wastewater. Water Science and Technology: A Journal of the International Association on Water Pollution Research 67 (5):1000–7. doi: 10.2166/wst.2013.653

Diels, L., and K. Vanbroekhoven. 2008. Remediation of metal and metalloid contaminated groundwater. In Methods and techniques for cleaning-up contaminated sites, NATO Science for Peace and Security Series C: Environmental Security, ed. M. D. Annable, M. Teodorescu, P. Hlavinek, and L. Diels, 1–23. Dordrecht: Springer. doi: 10.1007/978-1-4020-6875-1_1

Dvorak, D. H., R. S. Hedin, H. M. Edenborn, and P. E. McIntire. 1992. Treatment of metal-contaminated water using bacterial sulfate reduction: Results from pilot-scale

reactors. Biotechnology and Bioengineering 40 (5):609–16. doi: 10.1002/bit.260400508

Ehrlich, H. L. 1996 How microbes influence mineral growth and dissolution. Chemical Geology 132 (1-4):5–9. doi: 10.1016/S0009-2541(96)00035-6

Ehrlich, H. L. 1998 Geomicrobiology: Its significance for geology. Earth-Science Reviews 45 (1–2):45–60. doi: 10.1016/S0012-8252(98)00034-8

Ehrlich, H. L., D. K. Newman, and A. Kappler. 2015 Ehrlich's geomicrobiology. Boca Raton: CRC press.

Fründ, C, and Y. Cohen. 1992 Diurnal cycles of sulfate reduction under oxic conditions in cyanobacterial mats. Applied and Environmental Microbiology 58 (1):70–7.

Garbisu, C., O. Garaiyurrebaso, L. Epelde, E. Grohmann, and I. Alkorta. 2017 Plasmid-mediated bioaugmentation for the bioremediation of contaminated soils. Frontiers in Microbiology 8(:1966. doi: 10.3389/fmicb.2017.01966

Gibert, O., J. De Pablo, J. L. Cortina, and C. Ayora. 2005 Sorption studies of Zn (II) and Cu (II) onto vegetal compost used on reactive mixtures for in situ treatment of acid mine drainage. Water Research 39 (13):2827–38. doi: 10.1016/j.watres.2005.04.056

Gibert, O., J. de Pablo, J. Luis Cortina, and C. Ayora. 2003 Evaluation of municipal compost/limestone/iron mixtures as filling material for permeable reactive barriers for in-situ acid mine drainage treatment. Journal of Chemical Technology & Biotechnology 78 (5):489–96. doi: 10.1002/jctb.814

Gilbert, H. J., A. F. Sheehan, K. G. Dueker, and P. Molnar. 2003 Receiver functions in the western United States, with implications for upper mantle structure and dynamics. Journal of Geophysical Research: Solid Earth 108 (B5).

Gilmour, C, and G. Riedel. 2009 Biogeochemistry of trace metals and mettaloids.

Gleeson, D., F. McDermott, and N. Clipson. 2007 Understanding microbially active biogeochemical environments. Advances in Applied Microbiology 62:81–104.

Greben, H. A., J. Baloyi, J. Sigama, and S. N. Venter. 2009 Bioremediation of sulphate rich mine effluents using grass cuttings and rumen fluid microorganisms. Journal of Geochemical Exploration 100 (2-3):163–8. doi: 10.1016/j.gexplo.2008.01.004

Hammack, R. W., D. H. Dvorak, and H. M. Edenborn. 1993 The use of biogenic hydrogen sulfide to selectively recover metals from a severely contaminated mine drainage. In Proceedings of Proceedings of the International Biohydrometallurgy Symposium.

Hinchee, R. E., D. C. Downey, and P. K. Aggarwal. 1991 Use of hydrogen peroxide as an oxygen source for in situ biodegradation: Part I. Field studies. Journal of Hazardous Materials 27 (3):287–99. doi: 10.1016/0304-3894(91)80055-S

Huang, X.-D., Y. El-Alawi, D. M. Penrose, B. R. Glick, and B. M. Greenberg. 2004 A multi-process phytoremediation system for removal of polycyclic aromatic hydrocarbons from contaminated soils. Environmental Pollution (Barking, Essex : 1987) 130 (3):465–76. doi: 10.1016/j.envpol.2003.09.031

Hughes, T. A, and N. F. Gray. 2013 Co-treatment of acid mine drainage with municipal wastewater: Performance evaluation. Environmental Science and Pollution Research 20 (11):7863–77. doi: 10.1007/s11356-012-1303-4

Humphries, M. S., T. S. McCarthy, and L. Pillay. 2017 Attenuation of pollution arising from acid mine drainage by a natural wetland on the Witwatersrand, South African Journal of Science 113 (Number 1/2):1–9. doi: 10.17159/sajs.2017/20160237

Ibanez, J. G., Hernandez-Esparza, M. Doria-Serrano, C. Fregoso-Infante, A. Singh, and M. M. 2008 Air Oxidation of Metal Ions. In Environmental chemistry: Microscale laboratory experiments, ed. Ibanez, J. G., Hernandez-Esparza, M., Doria-Serrano, C., Fregoso-Infante, A. and Singh, M. M., 89–98. New York, NY: Springer New York. doi: 10.1007/978-0-387-49493-7_7

Jaiyeola, A. T, and J. K. Bwapwa. 2016 Treatment technology for brewery wastewater in a water-scarce country: A review. South African Journal of Science 112 (Number 3/4):1–8. doi: 10.17159/sajs.2016/20150069

Johnson, D. B, and K. B. Hallberg. 2005 Acid mine drainage remediation options: A review. The Science of the Total Environment 338 (1–2):3–14. doi: 10.1016/j.scitotenv.2004.09.002

Jones, A. M., P. J. Griffin, R. N. Collins, and T. D. Waite. 2014 Ferrous iron oxidation under acidic conditions—The effect of ferric oxide surfaces. Geochimica et Cosmochimica Acta 145:1–12. doi: 10.1016/j.gca.2014.09.020

Jørgensen, B. B. 1994 Sulfate reduction and thiosulfate transformations in a cyanobacterial mat during a diel oxygen cycle. FEMS Microbiology Ecology 13 (4):303–12. doi: 10.1016/0168-6496(94)90068-X

Kaksonen, A., M.-L. Riekkola-Vanhanen, and J. Puhakka. 2003 Optimization of metal sulphide precipitation in fluidized-bed treatment of acidic wastewater. Water Research 37 (2):255–66. doi: 10.1016/S0043-1354(02)00267-1

Kashefi, K, and D. R. Lovley. 2000 Reduction of Fe (III), Mn (IV), and toxic metals at 100 C by Pyrobaculum islandicum. Applied and Environmental Microbiology 66 (3):1050–6. doi: 10.1128/AEM.66.3.1050-1056.2000

Krantz-Rücker, C., B. Allard, and J. Sörbo. 1993 Interactions between a soil fungus, Trichoderma harzianum, and IIb metals—adsorption to mycelium and production of complexing metabolites. Biometals 6 (4):223–30. doi: 10.1007/BF00187759

Krantz-Rücker, C., B. Allard, and J. Sörbo. 1996 Adsorption of IIB-metals by three common soil fungi—comparison and assessment of importance for metal distribution in natural soil systems. Soil Biology and Biochemistry 28 (7):967–75. doi: 10.1016/0038-0717(95)00169-7

Landa, E. R. 2005 Microbial biogeochemistry of uranium mill tailings. Advances in Applied Microbiology 57:113–30.

Ledin, M, and K. Pedersen. 1996. The environmental impact of mine wastes—Roles of microorganisms and their significance in treatment of mine wastes. Earth-Science Reviews 41 (1-2):67–108. doi: 10.1016/0012-8252(96)00016-5

Liu, H., S. Tan, Z. Sheng, T. Yu, and Y. Li. 2015. Impact of oxygen on the coexistence of nitrification, denitrification, and sulfate reduction in oxygen-based membrane aerated biofilm. Canadian Journal of Microbiology 61 (3): 237–42. doi: 10.1139/cjm-2014-0574

Lottermoser, B. 2003. Sulfidic mine wastes. In Mine wastes: Characterization, treatment and environmental impacts, ed. Lottermoser, B., 3–32. Berlin, Heidelberg: Springer Berlin Heidelberg. doi: 10.1007/978-3-662-05133-7_2

Lu, J., T. Chen, J. Wu, P. C. Wilson, X. Hao, and J. Qian. 2011. Acid tolerance of an acid mine drainage bioremediation system based on biological sulfate reduction. Bioresource Technology 102 (22):10401–6. doi: 10.1016/j.biortech.2011.09.046

Luptakova, A., E. Macingova, I. Kotulicova, and D. Rudzanova. 2016. Sulphates removal from acid mine drainage. In Proceedings of IOP conference series: Earth and environmental science, Prague, Czech Republic, IOP Publishing, 052040.

Lyew, D, and J. D. Sheppard. 1997. Effects of physical parameters of a gravel bed on the activity of sulphate-reducing bacteria in the presence of acid mine drainage. Journal of Chemical Technology & Biotechnology 70 (3): 223–30. doi: 10.1002/(SICI)1097-4660(199711)70:3<223::AID-JCTB762>3.0.CO;2-L

Machemer, S. D, and T. R. Wildeman. 1992. Adsorption compared with sulfide precipitation as metal removal processes from acid mine drainage in a constructed wetland. Journal of Contaminant Hydrology 9 (1-2):115–31. doi: 10.1016/0169-7722(92)90054-I

Marchioretto, M. M., H. Bruning, and W. Rulkens. 2005. Heavy metals precipitation in sewage sludge. Separation Science and Technology 40 (16):3393–405. doi: 10.1080/01496390500423748

Martins, M., M. L. Faleiro, G. Silva, S. Chaves, R. Tenreiro, and M. C. Costa. 2011. Dynamics of bacterial community in up-flow anaerobic packed bed system for acid mine drainage treatment using wine wastes as carbon source. International Biodeterioration & Biodegradation 65 (1): 78–84. doi: 10.1016/j.ibiod.2010.09.005

McCullough, C. D, and M. A. Lund. 2011. Bioremediation of acidic and metalliferous drainage (AMD) through organic carbon amendment by municipal sewage and green waste. Journal of Environmental Management 92 (10):2419–26. doi: 10.1016/j.jenvman.2011.04.011

Muhammad, S. N., Kusin, F. M. Zahar, M. S. M. Halimoon, N. Yusuf, and F. M. 2015. Passive treatment of acid mine drainage using mixed substrates: Batch experiments. Procedia Environmental Sciences 30:157–61. doi: 10.1016/j.proenv.2015.10.028

Muyzer, G, and A. J. Stam. 2008. The ecology and biotechnology of sulphate-reducing bacteria. Nature Reviews. Microbiology 6 (6):441–54. doi: 10.1038/nrmicro1892

Nancucheo, I, and D. B. Johnson. 2012. Selective removal of transition metals from acidic mine waters by novel consortia of acidophilic sulfidogenic bacteria. Microbial Biotechnology 5 (1):34–44. doi: 10.1111/j.1751-7915.2011.00285.x

Neculita, C. M., G. J. Zagury, and B. Bussiere. 2007. Passive treatment of acid mine drainage in bioreactors using sulfate-reducing bacteria. Journal of Environmental Quality 36 (1):1–16. doi: 10.2134/jeq2006.0066

Nijjer, S., J. Thonstad, and G. M. Haarberg. 2000. Oxidation of manganese(II) and reduction of manganese dioxide in sulphuric acid. Electrochimica Acta 46 (2-3):395–9. doi: 10.1016/S0013-4686(00)00597-1

Osman, K. T. 2014. Physical deterioration of soil. In Soil degradation, conservation and remediation, 45–67. Dordrecht Heidelberg New York London: Springer.

Pawlowska, A, and Z. Sadowski. 2019. Bioreduction in the development of new mineral technology. In Proceedings of IOP Conference Series: Materials Science and Engineering, 012031. Bristol, England: IOP Publishing.

Peech, M., ALExANDER, L. Dean, L. Reed, and J. 1947. Methods of soil analyses for soil fertility investigations. US Department of Agriculture, Circ. N 757 (23):3–5.

Postgate, J. R. 1984. The sulphate-reducing bacteria, 2nd ed. Cambridge: University Press.

Rawlings, D. E. 2005. Characteristics and adaptability of iron-and sulfur-oxidizing microorganisms used for the recovery of metals from minerals and their concentrates. Microbial Cell Factories 4 (1):13–5. doi: 10.1186/1475-2859-4-13

Rawlings, D. E., D. Dew, and C. du Plessis. 2003. Biomineralization of metal-containing ores and concentrates. Trends in Biotechnology 21 (1):38–44. doi: 10.1016/S0167-7799(02)00004-5

Reeder, R. J., M. A. Schoonen, and A. Lanzirotti. 2006. Metal speciation and its role in bioaccessibility and bioavailability. Reviews in Mineralogy and Geochemistry 64 (1):59–113. doi: 10.2138/rmg.2006.64.3

Roberts, D., M. Nachtegaal, and D. L. Sparks. 2005. Speciation of metals in soils. Chemical Processes in Soils 8:619–54.

Sanchez-Andrea, I., D. Triana, and J. L. Sanz. 2012. Bioremediation of acid mine drainage coupled with domestic wastewater treatment. Water Science and Technology: A Journal of the International Association on Water Pollution Research 66 (11):2425–31. doi: 10.2166/wst.2012.477

Silva, A. M., E. C. Cunha, F. D. R. Silva, and V. A. de. 2012. Treatment of high-manganese mine water with limestone and sodium carbonate. Journal of Cleaner Production 29-30:11–9. doi: 10.1016/j.jclepro.2012.01.032

Skousen, J., C. E. Zipper, A. Rose, P. F. Ziemkiewicz, R. Nairn, L. M. McDonald, and R. L. Kleinmann. 2017. Review of passive systems for acid mine drainage

treatment. Mine Water and the Environment 36 (1): 133–53. doi:10.1007/s10230-016-0417-1

Song, Y., M. Fitch, J. Burken, L. Nass, S. Chilukiri, N. Gale, and C. Ross. 2001. Lead and zinc removal by laboratory-scale constructed wetlands. Water Environment Research: A Research Publication of the Water Environment Federation 73 (1):37–44. doi:10.2175/106143001x138660

Strosnider, W. H. J., B. K. Winfrey, R. A. M. Peer, and R. W. Nairn. 2013. Passive co-treatment of acid mine drainage and sewage: Anaerobic incubation reveals a regeneration technique and further treatment possibilities. Ecological Engineering 61:268–73. doi:10.1016/j.ecoleng.2013.09.037

Stumm, W, and J. Morgan. 1996. Metal ions in aqueous solution: aspects of coordination chemistry. In Aquatic chemistry: Chemical equilibria and rates in natural waters, 252–348. New York, NY: John Wiley & Sons, Inc.

Thome, A., C. Reginatto, I. Cecchin, and L. M. Colla. 2014. Bioventing in a residual clayey soil contaminated with a blend of biodiesel and diesel oil. Journal of Environmental Engineering 140 (11):06014005. doi:10.1061/(ASCE)EE.1943-7870.0000863

Vadapalli, V. R. K., J. N. Zvimba, M. Mathye, H. Fischer, and L. Bologo. 2015. Acid mine drainage neutralization in a pilot sequencing batch reactor using limestone from a paper and pulp industry. Environmental Technology 36 (19):2515–23. doi:10.1080/09593330.2015.1036786

van den Berg, M., M. Botes, E. Slabbert, and T. Cloete. 2016. Evaluating sulphate removal and identifying the bacterial community present in acid mine drainage treated with synthetic domestic wastewater sludge. Water SA 42 (3):475–82. doi:10.4314/wsa.v42i3.13

Violante, A., J. Zhu, M. Pigna, A. Jara, V. Cozzolino, and M. Mora. 2013. Role of biomolecules in influencing transformation mechanisms of metals and metalloids in soil environments. In Molecular environmental soil science, ed. J. Xu and D. L. Sparks, 1–57. Dordrecht: Springer.

Walker, D. 1972. Soil sulfate I. Extraction and measurement. Canadian Journal of Soil Science 52 (2):253–60. doi: 10.4141/cjss72-031

Warren, L. A, and E. A. Haack. 2001. Biogeochemical controls on metal behaviour in freshwater environments. Earth-Science Reviews 54 (4):261–320. doi:10.1016/S0012-8252(01)00032-0

Willow, M. A, and R. R. Cohen. 2003. pH, dissolved oxygen, and adsorption effects on metal removal in anaerobic bioreactors. Journal of Environmental Quality 32 (4): 1212–21. doi:10.2134/jeq2003.1212

Zachara, J., C. Cowan, and C. Resch. 1991. Sorption of divalent metals on calcite. Geochimica et Cosmochimica Acta 55 (6):1549–62. doi:10.1016/0016-7037(91)90127-Q