

AN ENVIRONMENT-FRIENDLY TECHNOLOGY FOR BIOLOGICAL SULPHATE AND SULPHIDE REMOVAL FROM ACID MINE DRAINAGE

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ABSTRACT

This paper presents the results of an innovative combined biological sulphate reduction and sulphide oxidation to sulphur technology. The novelty of the biological sulphate reduction system described here is the use of the degradation products of grass cellulose as the carbon and energy sources. Sulphate removal of an average of 85% was obtained operating a hybrid fermentation reactor system, using rumen fluid as the cellulose degrading microorganisms and sulphate reducing bacteria to reduce sulphate to sulphide. When the weekly grass cutting addition was reduced from 150 to 100 g an improvement in the sulphate reduction was observed. Biological sulphide oxidation was conducted at different volumes of air entering the sulphide oxidising reactor. The results showed that when using 0.2 L/min of air, most sulphide was oxidised to sulphur. The purity of sulphur varied from 17 to 81%. This paper showed that the biological operated system offers a number of advantages, the acidic pH of mine water increased, the metals precipitated as metal-sulphides, the sulphate was removed by 85% and the excess sulphide after metal-sulphide precipitation could be oxidised to relatively pure sulphur.

1. INTRODUCTION

Mineral mining generates acidic, saline, metal-rich mine waters, often referred to as acid mine drainage (AMD). Treatment of AMD and recovering saleable products during the treatment process is a necessity since water is a scarce commodity, especially in South Africa, while product recovery can contribute to recapturing part of the costs incurred during the water treatment process. Several technologies exist for the treatment of AMD e.g. a novel CSIR developed biological sulphate/sulphide removal technology, reducing the sulphate concentration to less than 500 mg/L and oxidising the sulphide generated to biologically produced sulphur. Degradation products of grass-cellulose are used as cost-effective carbon and energy sources for the sulphate reducing bacteria (SRB) (Greben et al., 2007; 2008). Due to the alkalinity and sulphide production, the pH of the AMD can be increased to neutral and the metals can precipitate as metal-sulphide, respectively. The solubility product of metal sulphide is lower than that of most metal hydroxides and thus stabilisation of metals is preferred in the form of sulphides (Gazea et al., 1996). Traditionally, AMD was neutralised with lime or another alkali to increase the pH of the AMD and to precipitate metals as hydroxides and carbonates (Santos et al., 2004). In the UK as well as in the USA the focus for AMD treatment is shifting to passive treatment systems with the advantage that a denser and more stable sludge is produced during biological passive treatment than following the chemical treatment. Neculita et al., 2007 stated that the biological treatment of AMD does not require the addition of extra chemicals, but it does need an electron donor with sulphate as electron acceptor.

Sulphide is the reduction product from sulphate, which due to its toxicity, unpleasant odour and to its high oxygen demand should not be released in the environment. Furthermore, sulphide has highly corrosive properties as can be noticed from the damage done to concrete pillars at harbours, in sewer systems and in steel pipelines (Buisman, 1989). Sulphide can be removed by physical-chemical processes, such as stripping and by chemical precipitation and oxidation. However, high energy requirements related to air stripping combined with the production of H₂S as a gas constitute important drawbacks of the stripping option. The chemical treatment option to produce sulphur is a highly intensive procedure, while the biological sulphide oxidation is a natural process, forming a part of the biological sulphur cycle: Sulphate reduction → sulphide production → sulphide oxidation → sulphur production. The sulphur produced can be retrieved and used as a saleable product, e.g. as source for the production of sulphuric acid, needed at mines for mineral processing.

The first aim of this study is to show that the degradation products of grass-cellulose, fermented by rumen microflora, can function as the carbon and energy source for the biological sulphate removal in mine effluents and the second aim is to show that the sulphide produced can biologically be oxidised to sulphur.

2. MATERIAL AND METHODS

Reactor Configuration and Sulphate-Rich Feed Water for Biological Sulphate Removal

Pre-treated, sulphate-rich mine water, consisting of one part AMD and one part reactor effluent (obtained after biological sulphate removal) was used as feed water. The AMD was obtained from a closed mine in the Witbank area, South Africa. The feed rate of 5 L/d resulted in a HRT of 2.4 d. The experimental period feeding sulphate-rich pre-treated AMD lasted 128 days during which the reactor received at first (till day 64, included) 150 gram grass cuttings (GC)/week and from day 67 to day 128, it received 100 g GC/week.

A one stage anaerobic (the dissolved oxygen concentration (DO) measured 0) reactor system, hybrid fermentation system (HFS), (Volume: 20 L and Active Volume 12 L) was operated (Figure 1). The lower part of the reactor contained ceramic rings for biofilm formation for the SRB, thus preventing microbial wash out. The SRB were obtained from a biological sulphate removal demonstration plant (Witbank, South Africa). The upper part of the reactor contained GC to which sieved (mesh 10x10 mm) rumen inoculum (RI), obtained from an abattoir (Witbank, South Africa) was added. GC were used as the source of cellulose, from which the fermentation products served as the carbon and energy source in the reactor. Kikuyu GC were obtained from the CSIR, Pretoria, Garden Service. The GC (size 10-20 mm) were collected and stored at room temperature. The mass of the GC in this report refers to air dried grass. Grass consists generally of 52% water, 14% cellulose and 28% hemicellulose. (Sonakya *et al.*, 2003). The empirical COD concentration of grass was found to be 1 g O₂/g. The GC are at first solubilised by enzymes excreted by the hydrolytic microbes, followed by the fermentation of monomers, producing volatile fatty acids (VFA) and hydrogen (H₂) to be used by the SRB as electron donors. The feed water entered HFS at the top (Figure 1). A high flow recycle stream (~600 L/day) was installed from the fermentation part to the top of the reactor, for improved reactor mixing purposes. The high return flow assists in preventing blockages, which can occur through “grass-debris”, which originate from partly degraded GC. HFS was operated at 25°C, through water recycling from a water bath (25°C) entering a water jacket surrounding the reactor. The effluent (5 L/day) left HFS at the bottom, from where it was pumped in the sulphide oxidising reactor (SOR).

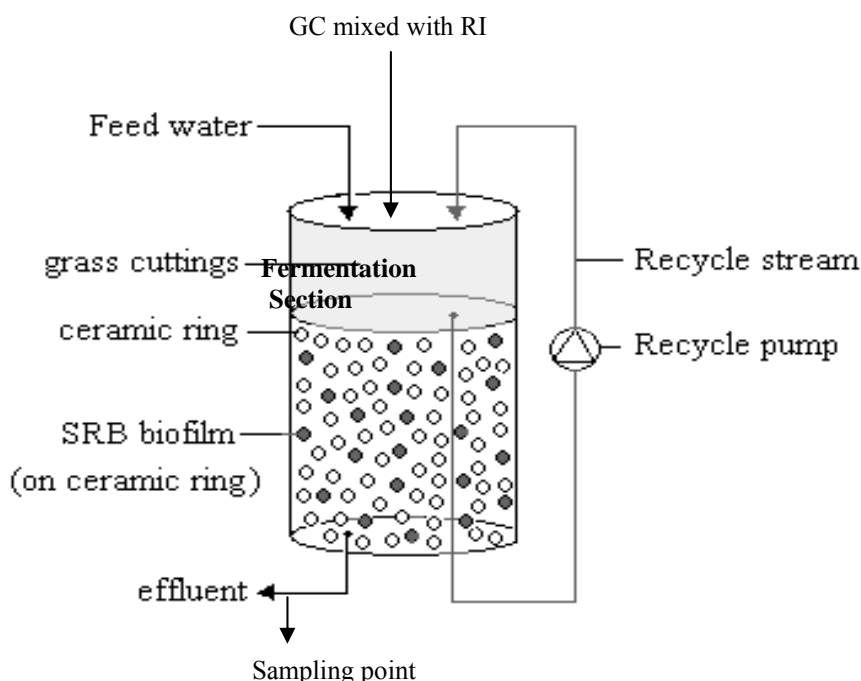


Figure 1. Schematic overview of hybrid fermentation system reactor system

Reactor Configuration and Sulphide-Rich Feed Water for Biological Sulphur Production

SOR had a volume of 4.7 L and an active volume 4.4 L and comprised a 9.1 cm diameter column, inside which another column of 5.6 cm diameter was constructed. This inner column contained plastic packing material as support medium for biofilm formation for the sulphide oxidising bacteria (SOB). Compressed air was used for the oxidation of sulphide. Excess air was recycled into the reactor, while the overflow of the gas was released through an outlet at the top of the reactor and bubbled into a Zinc Acetate solution to capture the released H₂S gas. The sulphide rich feed water as well as the air flow entered the reactor at the bottom of the 5.6 cm diameter column through two different inlet systems. The airflow rises to the top of the inner tube, during which it contacts the biofilm on the plastic support material, where biological sulphide oxidation to sulphur occurs.

The air is recycled at the top of the inner tube and re-enters the reactor at the bottom. The reactor was operated in continuous mode and the study consisted of four different periods, determined by the air supply, which varied from 0.2, 0.4, 0.6 and 0.8 L/min.

Scanning Electron Microscope Preparation

Liquid samples preparation

Samples were fixed in 3ml of 2% glutaraldehyde for 10 minutes at room temperature and washed with the buffer 3 times in a series of 15 minutes in between. Preparations were then filtered through a 0.2µm membrane filter and dehydrated in a series of between 30 to 100% ethanol ascending in 10% stages. Filters were critical point dried, coated with gold and then viewed under a JEOL-840 scanning electron microscope at an acceleration voltage of 5-15 kV.

Immobilizing material preparation

A piece of support material sample was cut and placed in a beaker, fixed in 3 ml of 2% glutaraldehyde for 15 minutes at room temperature. Preparations were then washed with buffer and dehydrated in a series of between 30 to 100% ethanol ascending in 10% stages. Samples were critical point dried, coated with gold and then viewed under a JEOL-840 scanning electron microscope at an acceleration voltage of 5-15 kV.

Analytical

Daily samples were analysed for sulphate, sulphide, alkalinity, soluble COD, pH, VFA and VSS. All analyses were carried out according to standards analytical procedures as described in Standards Methods (APHA, 1985). The nitrate (NO_3^- -N) and the phosphate (PO_4^{3-} -P) concentrations were analysed using the Hach Spectrophotometer DR/2010. The presented graphs in the following figures represent the results of the daily analyses, while the data in the tables represent the average values of the results of the daily analyses. The analyses were all carried out on filtered samples except for the COD analysis on feed water, the redox potential and the sulphide samples. Alkalinity was determined by titrating with 0.1 N HCl to a pH of 4.3. Prior to the COD measurement, the sulphide in the samples from the reactors was removed by adding a few drops of 98% sulphuric acid and flushing the sample with nitrogen. The redox potential of the samples was calculated from the mV and stabilization temperature measured with a pH/redox meter (Metrohm 744)..

3. RESULTS AND DISCUSSION

Sulphate, Sulphide and Residual Cod Concentrations in HFS, Feeding Pre-Treated AMD

When feeding pre-treated mine water, an average of 85% sulphate removal was achieved (Figure 2), during which time the average COD concentration in the reactor was 1 060 mg/L (Table 1). This relatively high COD concentration resulted in a good sulphate removal as can be observed from Figure 3. The feed SO_4^{2-} concentration was on average 2 434 mg/L (Table 1), but on d 38 it decreased to 2 000 mg/L. The lower feed SO_4^{2-} concentration coincided with a higher COD concentration in the reactor, while the SO_4^{2-} concentration in the treated water was 0 mg/L during this period. These results may indicate that due to the lower feed SO_4^{2-} concentration to HFS, less COD was needed, resulting in a higher residual COD concentration in the reactor. The high SO_4^{2-} removal efficiency resulted in a high S^{2-} production (Figure 4), except during the period around d 38, when no SO_4^{2-} was detected in the treated water and when the higher residual COD concentration was noted (Figure 3). The experimental determined $\text{S}^{2-}/\text{SO}_4^{2-}$ ratio was 0.20 as opposed to the theoretical value of 0.33. The lower experimental value can partly be ascribed to sulphide removal as H_2S gas, as the average pH in the reactor was maintained at \pm pH of 6.6-6.9 to accommodate the rumen bacteria, which require this pH range for optimal performance (Hungate 1966). Weast (1981) described that the pK_a value of the dissociation equilibrium of H_2S is 7.04 at 18 °C. Above pH 8.0-9.0 virtually all dissolved sulphide is present in its ionised form, while at neutral pH values, 20 to 50% of the dissolved sulphide is present as H_2S , depending on the reactor temperature (O'Flaherty & Colleran, 2000). The average feed water pH to HFS was maintained at just over pH of 6 (6.14), while the pH of the treated water was 6.96. The average values obtained from the daily samples from HFS are presented in Table 1. These obtained results indicated that the SO_4^{2-} removal efficiency during these two periods (d1-65 and d 69-128) was similar to the full period at 85% removal. The higher residual COD concentration when feeding less grass (100g/week) can possibly be ascribed to a well functioning cellulose degrading microbial community, which became adapted to the reactor environment with time. From empirical data resulting from previous experiments (Greben et al., 2007; 2008), it was documented that generally 0.50 gram SO_4^{2-} was reduced using 1 gram of GC. During the two experimental periods from d 1-65 and d 66-128, when 150 and 100 gram grass/week were added, respectively, these values were 0.37 and 0.72 gram SO_4 reduced from 1 gram of GC, an increase of almost 100% more efficient use of the GC. This result may indicate that the microbial population can either degrade the grass more readily when less grass is available or due to a lower amount of substrate, the microorganisms become more effective in the degradation of cellulose.

Table 1. Average results of the different parameters related to the operation of HFS, feeding pre-treated AMD over a period of 128 days

Parameter	Feed water	Treated water
COD (mg/L)		1 060
COD (mg/L) from d 1-63		986
COD (mg/L) from d 64-128		1 129
SO ₄ ²⁻ (mg/L)	2434	368
S ²⁻ (mg/L)		400
Alkalinity (mg/L)	759	2 248
g SO ₄ ²⁻ reduced/1 g grass (d 1-65)		0.34
g SO ₄ ²⁻ reduced/1 g grass (d 66-128)		0.72

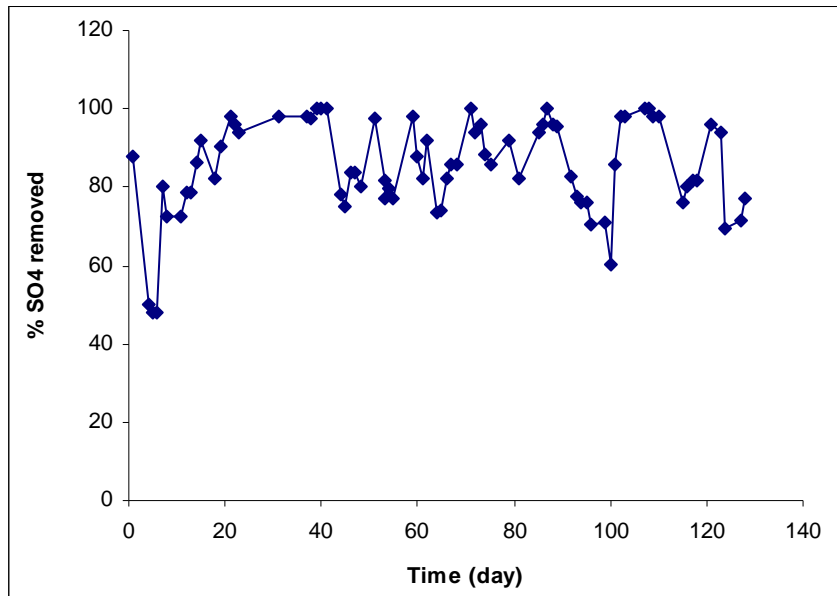


Figure 2. The % SO₄²⁻ removal in HFS, feeding pre-treated AMD during the total experimental period of 128 days

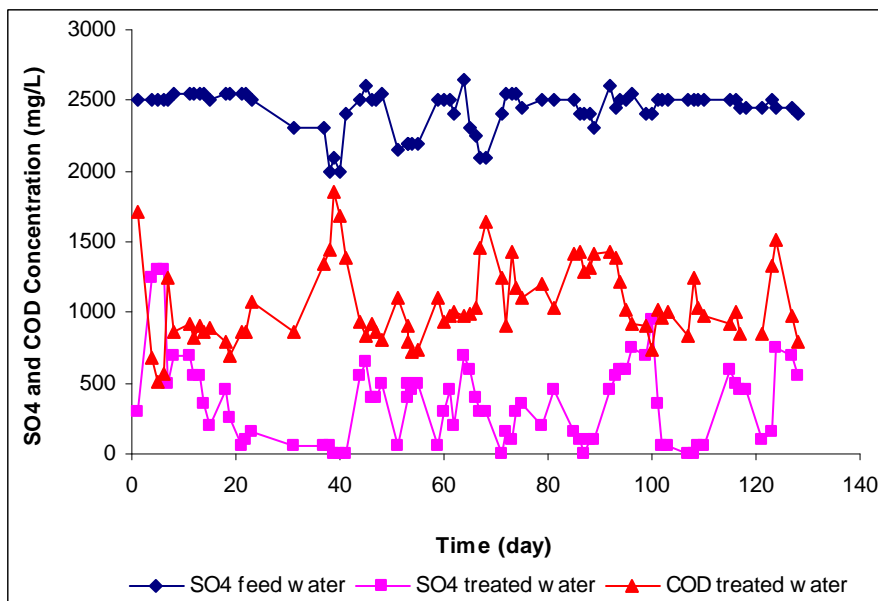


Figure 3. The SO₄²⁻ and COD concentration in the feed and treated water, respectively

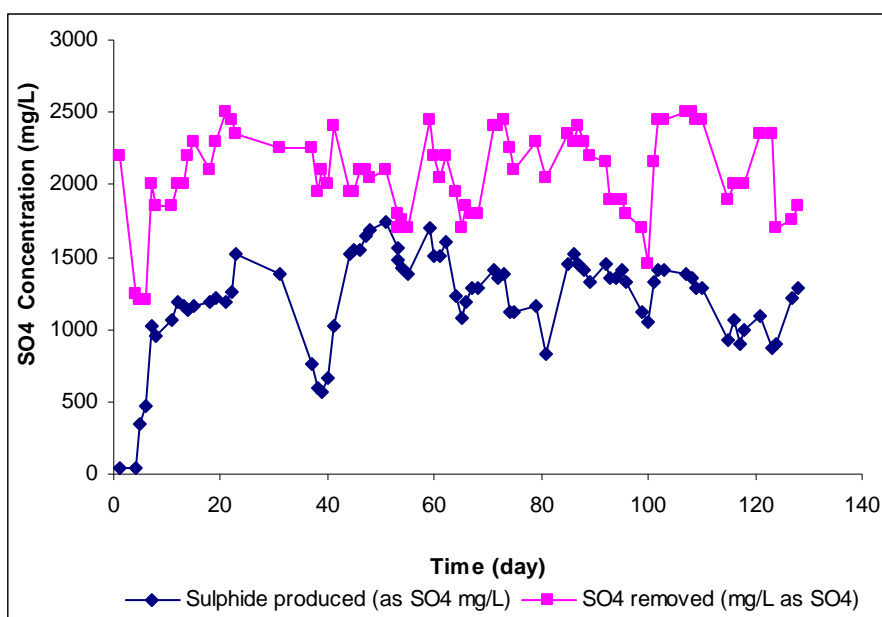


Figure 4. The sulphide produced and the sulphate removed (as mg/L SO_4^{2-})

Metal Removal from AMD and Pre-Treated AMD

During the pre-treatment of the AMD with the effluent from HFS (after biological sulphate removal), which is rich in sulphide, the metals present in the AMD can be precipitated with sulphide as is shown in Table 2. The data show that the AMD was rich in metals, especially iron, but it also showed relatively high concentrations of aluminium (Al) and manganese (Mn). Both Al and Fe could be removed to values < 1 mg/L, while the Mn was removed from 48 mg/L during pre-treatment at a pH of 6.14 and to 5.9 Mg/L at the higher pH 6.9.6. The additional metal removal inside HFS can account for the lower $\text{S}^{2-}/\text{SO}_4$ ratio as discussed earlier, since the S^{2-} is used for metal precipitation. The results in Table 2 show that the highest concentration of the metals was already removed in the pre-treatment phase. This result implies that the metal-sulphide precipitate can be removed in a settler, prior to AMD treatment. Since most of the metals have value, the metal sulphide sediment can be treated to retrieve the valuable metals during a leaching process.

Table 2. The metal concentration (mg/L) in the AMD, pre-treated AMD and in the effluent of HFS

Metal	AMD	Pre-treated AMD	Effluent HFS
Aluminium	24	14	<0.09
Chromium	<0.05	<0.05	<0.05
Iron	851	102	0.21
Lead	0.15	<0.03	<0.03
Manganese	48	27	5.9
Nickel	11	4.3	0.04
Zinc	1.4	0.94	<0.06

Scanning Electron Microscope (SEM) Images

Figure 5 shows the SEM images from the microbial population in HFS. It can be observed from the images that many different microorganisms are present in the reactor seemingly attached to the grass, assisting in the degradation process. The studies of Weimer et al., (2009) indicate that in order to attain significant fibre degradation the fibrolytic bacteria should adhere to cellulosic substrates and these authors furthermore state that the cellulose degradation rate is linear dependant on the surface area of the cellulosic material.

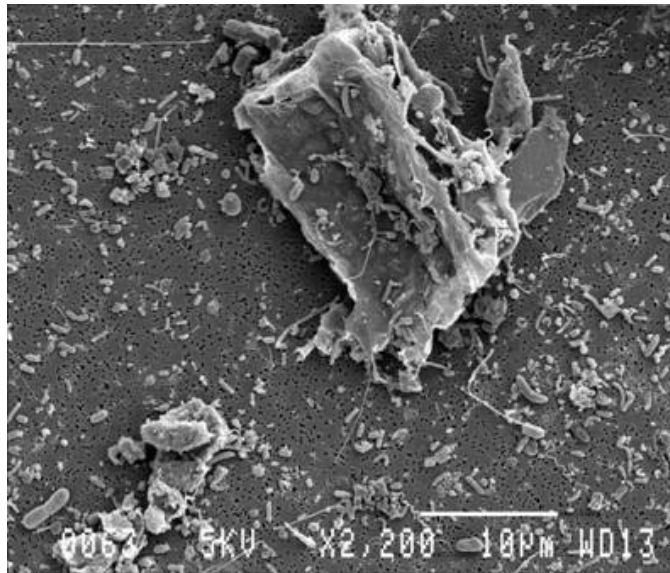


Figure 5. SEM image of the microbial population in HFS degrading grass-cuttings.

4. SULPHIDE OXIDATION

The product of biological sulphate reduction is sulphide, which can be biologically oxidised using oxygen/air (air comprises 80% nitrogen and 20% oxygen) to sulphur as the end product. In order to attain the objective, air instead of oxygen was used for the laboratory experiments, with the rationale that when conducting this oxidation process at full scale, the use of air is more cost effective than the use of oxygen. As indicated under Material and Methods, the effluent from HFS served as the feed water to SOR. This water contained S^{2-} , as the reduction product of SO_4^{2-} , as well as a residual SO_4^{2-} concentration. The different concentrations of air (0.2; 0.4; 0.6 and 0.8 L/min) as supplied to the reactor resulted in four experimental periods, namely d 29-43; d 44-59; d 62-81 and d 83-97.

Results Sulphide/Sulphate Removal in SOR

The biological produced S^{2-} as obtained following the biological SO_4^{2-} reduction in HFS needs to be removed from the HFS effluent. The average S^{2-} concentration from HFS was 400 mg/L, while the SO_4^{2-} concentration in the treated water was 368 mg/L SO_4 , which resembles a S^{2-} concentration of $368/3 = 123$ mg/L. Thus the total S^{2-} concentration entering SOR comprises the S^{2-} and the S^{2-} concentration as represented by the SO_4^{2-} concentration. The S^{2-} removed in SOR during the different periods is shown in Figure 6. These periods (demarcated by the vertical bars) were determined by the airflow as L/min (illustrated in the text boxes) to the reactor, during the different experimental periods.

When observing the sulphide concentration removed during the different experimental periods, it can be noted that highest sulphide removal was obtained during the periods when 0.2 L/min. air and 0.8 L/min. air was supplied, respectively. However the data in Table 3 show that the effluent S^{2-} concentration decreased with the increasing air flow concentration, thus the higher the airflow, the lower the S^{2-} concentration in the effluent of SOR. The sulphide removed in SOR can be linked to the sulphur produced during the same periods (Table 3). The SO_4^{2-} concentration entering and leaving SOR was measured to investigate whether the air supply was high enough for total sulphide oxidation and low enough that no sulphur to sulphate oxidation occurred. The relationship between the air supplies, the sulphide removed, sulphur produced and sulphur oxidised are presented in Table 3. The results show that the amount of sulphide removed (g/d) is much higher than the sulphur produced. This can be possible ascribed to the fact that a part of the sulphur formed is still present in the reactor water. Samples of the harvested sulphur were sent to be analysed for purity of the sulphur particles (Sanas testing Laboratories, South Africa). The results showed that the purity of the sulphur in the samples varied from 17% to 50%, to 60% to 70% with as highest 81 % pure sulphur. Further studies will be conducted to assure a more stable purity of the biologically produced sulphur.

The results in Table 3 furthermore show that with the increase of the airflow, the SO_4^{2-} concentration between the influent and effluent in the reactor increased and the more the airflow increased the higher the amount of SO_4^{2-} formed increased. Thus the airflow needs to be carefully regulated so that an increase the SO_4^{2-} concentration in the sulphide oxidising reactor can be avoided.

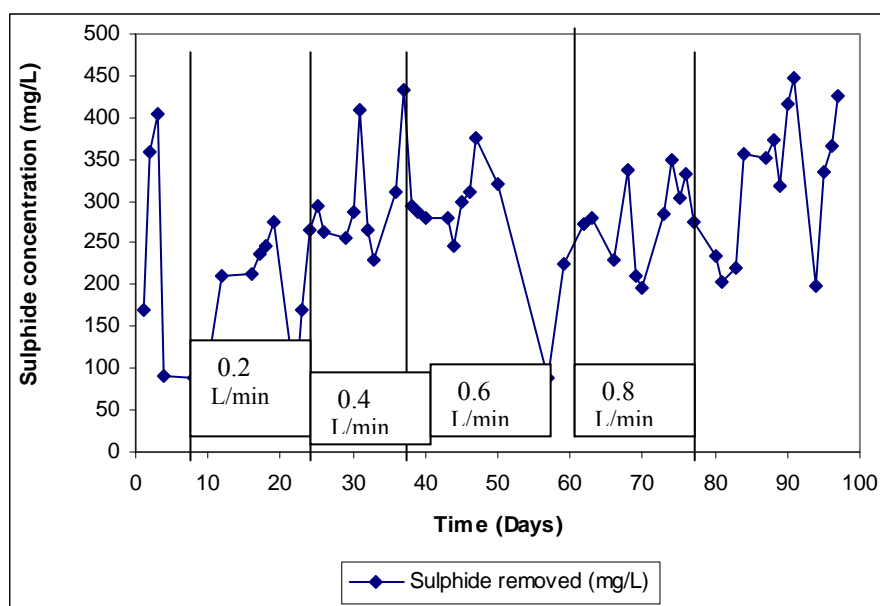


Figure 6. Total sulphide concentration (mg/L) removed in the sulphide oxidising reactor (including the sulphate concentration, expressed as sulphide)

Table 3. The relationship between the sulphide removed and sulphur formed and the sulphate increase under different concentrations of air.

Air (L/min)	S ²⁻ conc. in effluent (mg/L)	S ²⁻ removed (g/d)	S ⁰ produced (g/d)	SO ₄ ²⁻ in influent to SOR (mg/L)	SO ₄ ²⁻ in effluent from SOR (mg/L)	Increase in SO ₄ ²⁻ (%)
0.2	123	5.13	1.98	823	888	8
0.4	79	4.45	1.96	713	913	28
0.6	43	4.62	2.06	635	931	47
0.8	15	5.77	1.28	508	795	57

Janssen (1996) observed that with increasing loading rates of sulphide, the chemical sulphide oxidation became more important, which induced the formation of thiosulphate. Janssen (1996) furthermore observed that under oxygen limitation, which is at molar $(O_2/S^{2-})_{consumption}$ between 0.5 and 1.0, the system produces mainly thiosulphate and sulphur, while at molar $(O_2/S^{2-})_{consumption} > 1$, sulphate is the primary oxidation product. The results as presented in Table 3 corroborate the finding of Janssen, since when the air flow increased, making the molar $(O_2/S^{2-})_{consumption} > 1$, the sulphate concentration increased. Janssen (1996) also noted that at molar $(O_2/S^{2-})_{consumption}$ between 0.6 and 1.0, a maximal sulphur formation was obtained. This was not achieved at the stoichiometrically value of molar $(O_2/S^{2-})_{consumption} = 0.5$, due to the formation of thiosulphate. Janssen concluded that it is not possible to convert all sulphide in the influent to elemental sulphur, but that some sulphate will always be produced, either due to an excess of oxygen or to the formation of thiosulphate under oxygen limiting conditions. The results in Table 3 show that 0.2 L/min. air for continuous operation provided very little additional sulphate, while relative high sulphide removal (g/d) was observed. It must however be noted that the amount of air is immediately related to the sulphide concentration entering the reactor. It was shown in this investigation that sulphide removal was achieved during two different processes: using the sulphide-rich effluent to precipitate the metals, present in the AMD and through the biological oxidation to sulphur, using air and mobilised sulphide oxidising bacteria.

5. CONCLUSIONS

The percentage sulphate removal efficiency during the presented study was 85%, when feeding the hybrid bioreactor with sulphate-rich pre-treated AMD as feed water over the total experimental period of 128 days. The high sulphate removal seemed to be dependant on a high residual COD concentration, which was maintained by the weekly addition of GC. When decreasing the weekly addition of GC by 50 g, the g grass_{added}/g sulphate_{removed} ratio improved by more than 100% and resulted in an increase in the SO₄²⁻ removal efficiency by 3%. Metal removal was achieved both in the pre-treatment, using the sulphide rich effluent of the reactor after sulphate reduction to precipitate the metals present in the diluted AMD. The remaining metals in the feed water were precipitated during the SO₄²⁻ removal process in HFS. In

the second step of the biological sulphate/sulphide removing technology, sulphide was removed in the sulphide oxidising reactor, when combining air and HFS effluent in this reactor. The sulphide oxidising bacteria biologically oxidised the sulphide to sulphur, ideally the end product in the biological sulphur cycle. The sulphur produced was analysed for its purity and the results showed that the sulphur purity varied from 17% to 81 %. Pure sulphur can be sold for the production of sulphuric acid, thus generating money from the treatment of waste water.

6. REFERENCES

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