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Performance of Biological Sulphate Reduction at Low pH, Low Temperature and Low Hydraulic Retention Time

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Biological sulphate reduction (BSR) is a promising technology for the treatment of acidic, heavy metal-laden, and high sulphate concentration effluents from mining industries. BSR is a process that uses sulphate reducing bacteria to reduce sulphate to sulphide using substrates as nutrients under anaerobic conditions. The performance of BSR is dependent on several factors including pH, temperature and hydraulic retention time (HRT). In this study, the effect of the above-mentioned factors was investigated in lab-scale reactors that were mimicking a pilot plant that is operational at a coal mine. Packed bed reactors (packed with cellulosic organic matter) were operated in a continuous downflow mode. Cow manure and lucerne pellets were used as substrates and they were replenished once a week. Sulphate reducing bacteria were able to reduce sulphate at 10 °C although at lower efficiencies compared to 30 °C. Decreasing the pH from 6 to 4 did not impact sulphate reduction significantly as sulphate reduction was above 89 % at pH 4 (20 °C) for the duration of the experiment. A washout of substrates was observed when HRT was deceased from 7 d to 2 d. This lead to a sulphate reduction drop from above 98 % at 7 d to 37.5 % at 2 d before replenishing and above 80 % after replenishing. This study shows that BSR is a promising technology for the abatement of AMD.

1. Introduction

High sulphate concentration, heavy metal-laden and acidic effluents are a result of many industrial processes. Of all the industrial processes, mining is the major contributor to these effluents. The phenomenon associated with the formation of such effluents is known as acid mine drainage (AMD). AMD is formed by the oxidation of a sulphide bearing mineral such as pyrite in the presence of oxygen and water as shown in Eq(1) (Bwapwa et al., 2017). The oxidation process is mediated by acid-tolerant bacteria (Kuyucak, 2002).

$$8FeS_2 + 15O_2 + 14H_2O \rightarrow 4Fe(OH)_3 + 8SO_4^{2-} + 16H^+ \quad (1)$$

Sulphate concentration in AMD in South Africa is above 2000 mg/L and pH below 3. When AMD seeps into freshwater bodies, it has negative impacts on humans and aquatic organisms (Simate and Ndlovu, 2014). High sulphate concentration is known to have laxative effects in humans (Luptakova et al., 2013) and can also cause acute and chronic diseases (Kefeni et al., 2017). The recommended wastewater discharge sulphate concentration in South Africa is below 600 mg/L and the World Health Organization (WHO) recommends 250 mg/L for drinking water (Arnold et al., 2016).
The treatment and management of AMD has been investigated in laboratory scale level, pilot scale level and full field scale level (Gibert et al., 2002). However, AMD characteristics differ according to what is being mined (McCarthy, 2011) therefore treatment goals may be different from site to site for different countries due to different discharge standards (Arnold et al., 2016). As a result, studies are still ongoing to investigate the best and sustainable way to treat AMD to meet different discharge standards.
Chemical addition of a neutralizing agent such as lime and calcium hydroxide have been used to neutralize AMD and precipitate heavy metals. However, this method is costly because it requires constant addition of a neutralizing agent and it does not reduce sulphate concentration efficiently (Arnold et al., 2016). In addition, this method produces large sludge quantities that pose disposal problems. Other AMD treatment methods

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include ion exchange, reverse osmosis and electro-dialysis. Although efficient, these methods are costly as they require pre-treatment and they produce brine waste that could be difficult to dispose of (Luptakova et al., 2013).

An AMD treatment method, classified as passive or semi-passive process, known as biological sulphate reduction (BSR) has received attention over the years due to its sustainability and cost-effectiveness (Neale et al., 2017). BSR uses sulphate reducing bacteria (SRB) to reduce sulphate to sulphide in anaerobic conditions. SRB uses sulphate as a terminal electron acceptor and substrates as electron donors (Moosa et al., 2002). Ethanol and methanol are examples of the most used substrates for SRB but they are costly (Zagury et al., 2007) and as a result, studies have been leaning towards other organic sources such as animal manure, sewage sludge and food wastes (Liamleam and Annachhatre, 2007).

The performance of BSR is dependent on the activity of SRB which is affected by several factors including pH, hydraulic retention time (HRT), substrate availability, temperature, redox potential and solid support for SRB growth (Sheoran et al., 2010). For this reason, it is important to consider these factors when designing a system to treat AMD biologically.

This study focuses on the effect of pH, temperature and HRT on the performance of BSR. This work forms part of an ongoing project by Mintek. Mintek piloted their BSR process at a coal mine in eMalahleni, Mpumalanga province, South Africa. This study was done to investigate what impact low pH, low temperature and low HRT has on their process to test how robust it is. The experiments were carried out in lab scale reactors that mimicked the pilot plant. The results found will be used to improve Mintek's BSR process.

2. Materials and Methods

2.1 Acid Mine Drainage and inoculum

Raw AMD was sourced from a coal mine in eMalahleni, Mpumalanga province, South Africa. The AMD had sulphate concentration above 2500 mg/L and the pH was below 3. To inoculate the reactors, an anaerobic mixed SRB culture was collected from one of the reactors operating at Mintek's pilot plant. A mixture of mine water (70 % $v.v^{-1}$) with pH adjusted to 6.5 using hydrated lime and inoculum (30 % $v.v^{-1}$) was used to inoculate the reactors.

2.2 Experimental setup

Water jacketed packed bed reactors that were operated continuously in a downflow mode were used. During startup, the reactors were packed with woodchips, wood shavings, lucerne straw, hay and cow manure to support the SRB biofilm formation. Cow manure and lucerne pellets were used as substrates and they were replenished once a week. Each reactor had a total volume of 17.6 L and an operating volume of 8 L. To control the temperature, a PolyScience Whispercool heater/chiller was used. Hydrated lime was used for all pH adjustments.

Initially, the reactors were operated batchwise for 10 days followed by recirculation for about a week. Once sulphate was reduced sufficiently (>98 %), feeding of AMD commenced.

2.3 Analytical methods

In order to prevent interferences with suspended particles, sulphate samples were filtered using 0.22 μm syringe filters. Merck Spectroquant Prove 300 was used for all sulphate determinations. All pH measurements were done using a Metrohm pH sensor.

3. Results and discussion

3.1 The effect of temperature

Both the growth rate and activity of SRB are greatly affected by temperature (Lettinga et al., 1984) with the optimum operating temperature between 20 and 35 °C (Baskaran, 2005). Figure 1 shows the temperature effect on sulphate reduction at influent pH of 6 and HRT of 4.5 d. At 30 °C, sulphate reduction was maintained above 93 % for the duration of the experiment reaching 97.4 % at the end of the experiment. This was expected as 30 °C is within the optimum SRB temperature. However, when the temperature was decreased to 10 °C, sulphate reduction was still possible although the efficiency decreased drastically. Sulphate reduction decreased by more than 55 % and it varied between 27 % and 36 % for the duration of the experiment. This can be attributed to the decreased SRB metabolic activity.

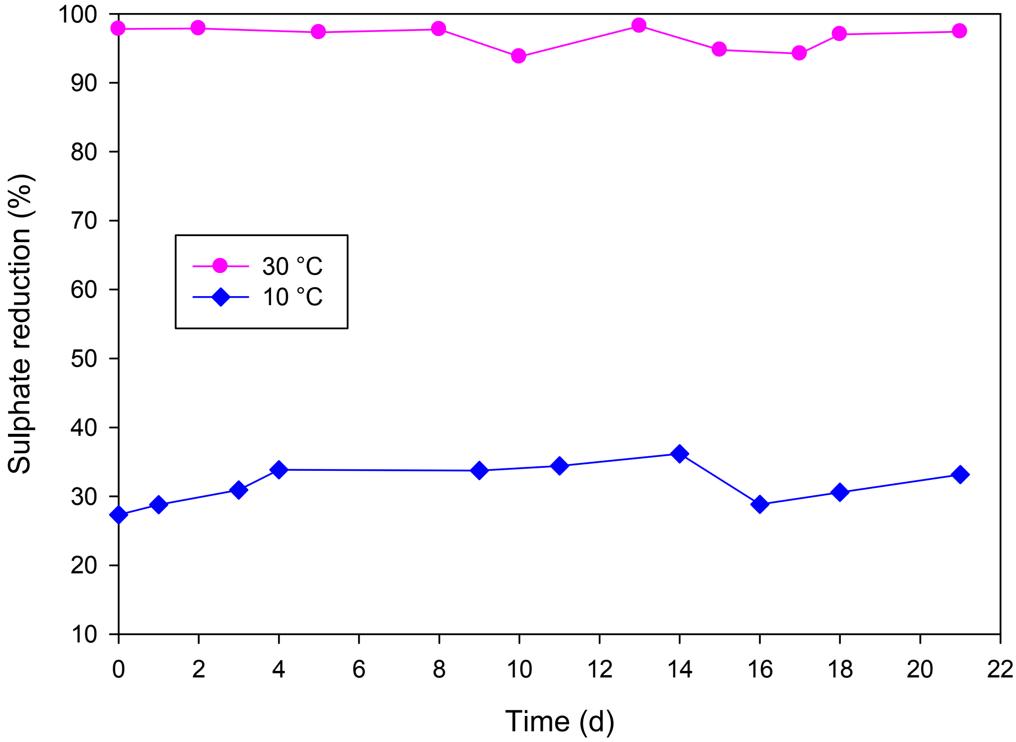

Figure 1: Temperature effect on sulphate reduction at pH 6 and HRT of 4.5 d.

3.2 The effect of pH

The effect of feed pH 4 and 6 on sulphate reduction was investigated at 20 °C and HRT of 7 days as shown in Figure 2a. Sulphate reduction was maintained above 96 % for the duration of the experiment at pH 6. Decreasing the pH from 6 to 4 resulted in a slight decrease in sulphate reduction from day 4 reaching 89 % at day 9, however, sulphate reduction started increasing from day 14 reaching 94 % at day 16 until day 21. Although there was a slight decrease in sulphate reduction when pH was decreased, sulphate reduction was maintained above 89 % at pH 4. This shows that SRB can survive at pH as low as 4 even though other studies suggest that SRB perform best at pH above 5.5 (Chaiprapat et al., 2011). In one study sulphate reduction was significantly affected dropping from above 98 % to below 65 % with a decrease in pH from 6 to 4 at 55 °C (Lopes, 2007), however, Jong and Parry (2006) have shown that sulphate reduction above 80 % is possible at 23 ± 1 °C. The results presented also show that SRB can thrive at pH 4 and sulphate reduction efficiency above 89 % at 20 °C can be achieved

A similar trend was observed when pH was decreased from 6 to 4 at 10 °C and HRT of 4.5 days shown in Figure 2b. Sulphate reduction was stable between 27 % and 36 % at pH 6. A decrease in pH lead to a decrease in sulphate reduction. However, after 10 days, SRB acclimatized to the pH and sulphate reduction was not significantly different ($p = 0.3$). This shows that given enough time, SRB can acclimatize to low pH. The same effect was observed when pH was decreased from 6 to 5 (Mukwevho et al., 2019).

Brahmacharimayum et al. (2019) suggested that a mixed SRB culture is likely to be tolerant to extreme conditions than a pure culture, which is possibly why the change in pH did not affect sulphate reduction efficiency. Sulphate reduction was not significantly affected by a decrease in pH from 6 to 4 at both temperatures.

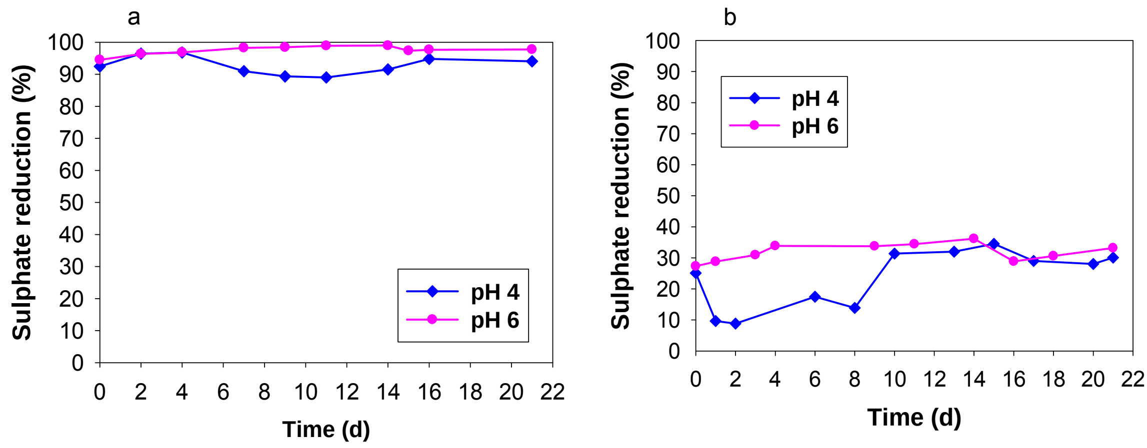

Figure 2: pH effect on sulphate reduction at (a) 20 °C and HRT of 7 d and (b) 10 °C and HRT of 4.5 d.

Figure 3 shows the effluent pH to influent pH 4 and 6 at (a) 20 °C, HRT 7 d and (b) 10 °C, HRT of 4.5 d. At 20 °C, the effluent pH was between 7 and 7.25 for influent pH of 4 and between 7.2 and 7.4 for influent pH of 6. At 10 °C, the effluent pH was between 5.96 and 6.16 for influent pH of 4 and between 6.04 and 6.44 for influent pH of 6. These results show that even at pH as low as 4, the reactors could increase the effluent pH to above 6. The increase in pH shows that SRB metabolic activity was not inhibited by low initial pH hence the system was able to consume the acidity (Castro Neto et al., 2018). This implies that lower costs need to be expended to get AMD to pH levels above 5.5 and this could have a positive impact on the process' operating expenses.

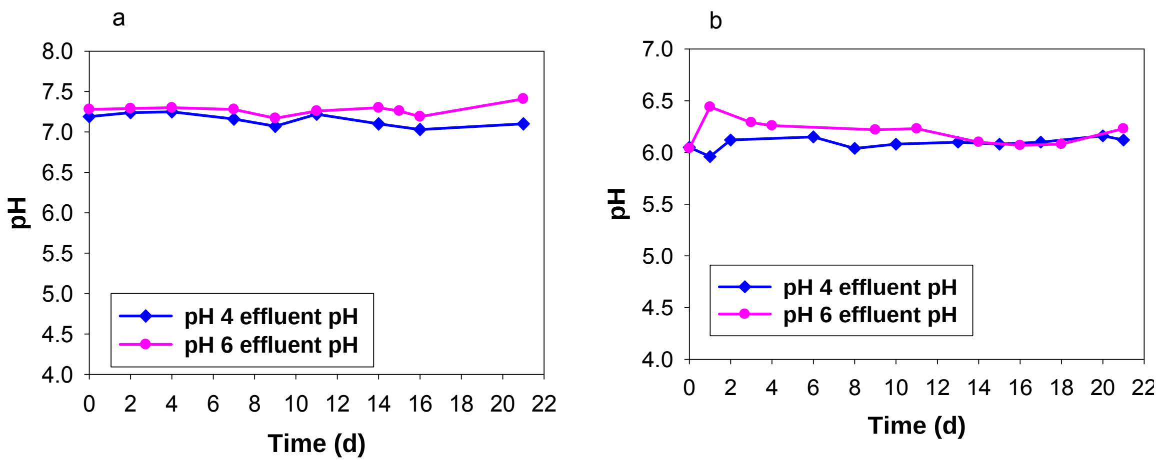

Figure 3: Effluent pH 4 and 6 at (a) 20 °C and HRT of 7 d and (b) 10 °C and HRT of 4.5 d.

3.3 The effect of HRT

HRT is one of the important factors that affect the performance of BSR. Figure 4 shows the impact HRT had on sulphate reduction when decreased from 7 d to 2 d. At HRT of 7 d, sulphate reduction was stable around 98 % throughout the experimental period. Decreasing HRT from 7 d to 4.5 d lead to a slight decrease in sulphate reduction to 93.7 % at day 4, however sulphate reduction increased and was maintained above 95 % until day 21. A further decrease in HRT to 2 d had a significant impact on sulphate reduction. A decrease in sulphate reduction to 50 % was observed when HRT was decreased to 2 d. Interestingly, after replenishing, sulphate reduction increased to 87.2 % at day 3. After 5 days of not replenishing the substrates, sulphate reduction decreased to 37.5 % (day 8). At day 8, the substrates were replenished and this lead to an increase in sulphate reduction to 82.3 % (day 11). The same trend was observed until the end of the experiment (day 21). Low HRT is known to washout SRB biomass (Sipma et al., 2007) and some studies showed that decreasing HRT leads a decrease in sulphate reduction efficiency due to SRB washout (Isa et al., 1986). However, in this study, it is presumed that low HRT resulted in the washout of substrates. This became evident when sulphate reduction improved upon replenishing.

These results show that sulphate reduction is still possible at HRT as low as 2 d, however, to maintain it above 80 % the substrates have to be replenished more often.

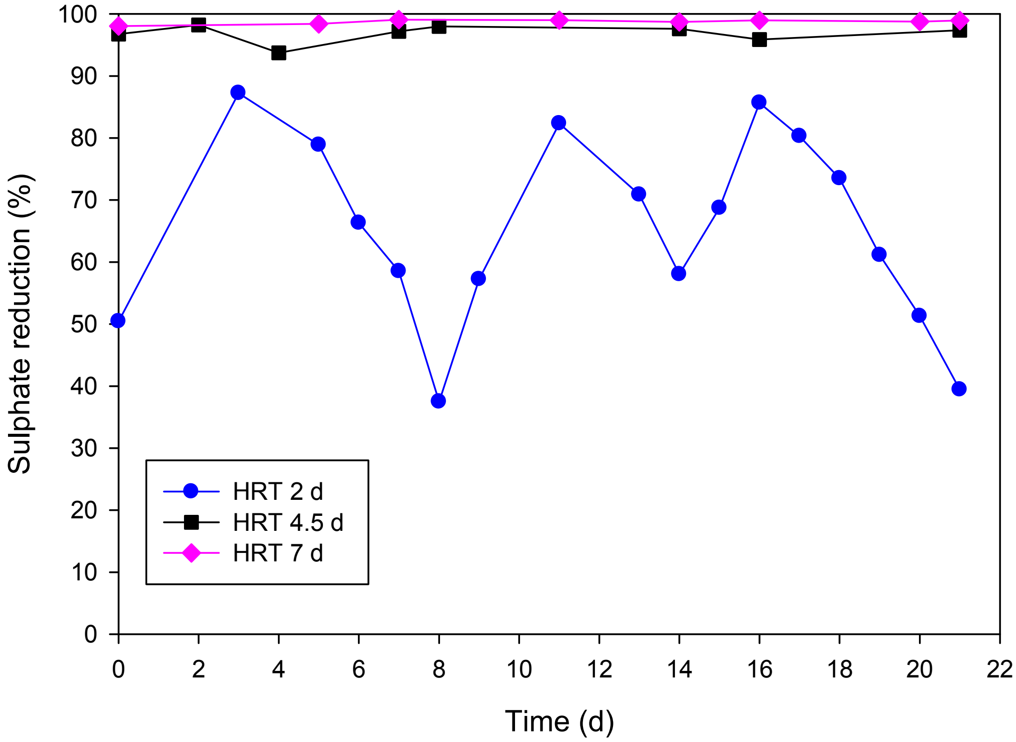

Figure 4: HRT effect on sulphate reduction at 30 °C and pH 5.

4. Conclusions

This study demonstrated that low temperature had a major impact on the metabolic activity of SRB. Sulphate reduction efficiency decreased from above 93 % at 30 °C to below 36 % a 10 °C. A decrease in pH from 6 to 4 did not have a significant impact on sulphate reduction. The efficiency of sulphate reduction was maintained above 89 % with a decrease in pH from 6 to 4 at 20 °C. The results presented also shows that there was no major impact on sulphate reduction when HRT is decreased from 7 d to 4.5 d but decreasing HRT further to 2 d had a major impact on sulphate reduction possibly due to washout of substrates. The results obtained in this study are of great importance in providing a tool for the operation of a BSR system and they provide a basis in understanding the interactions between operating conditions. The authors are therefore currently conducting further studies to produce a model that will determine the impact of sulphate reduction efficiency with changes to multiple operating conditions.

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