Sulfide removal from industrial wastewaters by lithotrophic denitrification using nitrate as an electron acceptor

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ABSTRACT

Sulfide is present in wastewaters as well as in biogas and can be removed by several physicochemical and biotechnological processes. Nitrate is a potential electron acceptor, readily available in most wastewater treatment plants and it can replace oxygen under anoxic conditions. A lab-scale reactor was operated for treatment of sulfide containing wastewater with nitrate as an electron acceptor and is used to evaluate the effects of volumetric loading rates, hydraulic retention time (HRT) and substrate concentrations on the performance of the lithotrophic denitrification process for treating industrial fermentation wastewaters. Sulfide is removed more than 90% at the loading rates between 0.055 and 2.004 kg S$_2^2$/m$^3$ d, when the influent sulfide concentration is kept around 0.163 kg/m$^3$ and the HRT decreased from 86.4 to 2 h. Nitrogen removal differed between 23 and 99% with different influent NO$_3^-$N concentration and loading rates of NO$_3^-$/S$_2^2$ ratio. The stoichiometry of sulfide oxidation with nitrate is calculated assuming different end-products based on thermodynamic approach and compared with experimental yield values. The calculated maximum volumetric and specific sulfide oxidation rates reached 0.076 kg S$_2^2$/m$^3$ h and 0.11 kg S$_2^2$/kg VSS h, respectively. The results are obtained at industrially relevant conditions and can be easily adapted to either biogas cleaning process or to sulfide containing effluent streams.

Key words | biogas cleaning, hydrogen sulfide, lithotrophic denitrification, nitrate

INTRODUCTION

In the wastewater containing high concentrations of sulfate, a significant fraction of the organic matter is degraded with sulfate as an electron acceptor in anaerobic reactors resulting in accumulation of hydrogen sulfide in the biogas (Kleerebezem & Mendez 2002). Sulfide in wastewaters has severe toxic effects on ecosystems, microorganisms and human health even at very low concentrations (Jing et al. 2007; Mahmood et al. 2007). Therefore, reduced sulfur compounds, especially hydrogen sulfide, cause environmental problems and have to be removed from gas streams, especially after the anaerobic treatment. Among those problems, high toxicity, odour, corrosive properties and high chemical oxygen demand may be listed (Manconi et al. 2007).

Many processes have been proposed and employed to remove sulfide from gas streams and sulfide rich wastewaters. Commercially applied processes normally use oxygen as electron acceptor for sulfide removal (Abatzoglou & Boivin 2008). However nitrate (and nitrite) are potential electron acceptors and are available in most wastewater treatment plants (Başpinar & Türker 2010). Chemolithotrophic denitrifying microorganisms oxidize reduced inorganic sulfur compounds by using nitrate as an electron acceptor. The process employing these denitrifying bacteria...
is generally called lithotrophic denitrification process. This process utilizes lithotrophic denitrifiers, such as *Thiobacillus denitrificans* and *Thimicrospira denitrificans*. They grow at neutral pH and reduce nitrate/nitrite to nitrogen gas. Oxidizing sulfide or other reduced sulfur compounds (S\(^-2\), S\(_2O_3\)^-2, SO_3^-2) to sulfate, eliminate the need to use organic compounds, resulting in less sludge production and disposal cost (*Moon et al. 2004; Beristain-Cardoso et al. 2007; Mahmood et al. 2007; Sierra-Alvarez et al. 2007; Campos et al. 2008*). On the other hand, sulfide is a by-product of sulfate reduction associated with methanogenesis. Sulfide sources for lithotrophic denitrification may be obtained virtually at no cost as by-products from some industrial processes (*Manconi et al. 2006*). The oxidation of sulfide by lithotrophic denitrifying bacteria can lead to the formation of elemental sulfur or sulfate depending on the stoichiometric balance and offers opportunities for the removal of this compound as the elemental sulfur. It can be easily removed from effluent for reuse since it has very low solubility in water (*Beristain-Cardoso et al. 2008*).

Previously in our group *Yavuz et al. (2007)* studied the removal of sulfide from molasses-based industrial wastewaters using oxygen and nitrate as electron acceptors in the presence of activated sludge. The effects of pH, temperature and concentration of activated sludge on specific sulfide oxidation rates were studied in laboratory-scale batch experiments. In addition, the stoichiometry of sulfide oxidation using both nitrate and oxygen were calculated assuming different end products based on thermodynamic approach and compared with experimental yield values (*Y_S/N*). Nitrite is another potential electron acceptor and readily available in most wastewater treatment plants. In continuing work, sulfide-nitrite reaction is studied in continuous culture with promising results (*Can-Doğan et al. 2010*). In this work, continuous flow stirred reactor (CFSR) is inoculated with an activated sludge from industrial wastewater treatment plant (WWTP) that receives fermentation wastewaters. The activated sludge used in this study was obtained from aerobic wastewater treatment plant and adapted to substrates for 52 days. The reactor was operated under anoxic conditions to accomplish lithotrophic denitrification. The experimental set up installed in the laboratory is schematically shown in *Figure 1*. A glass continuous flow stirred tank reactor, with a working volume of 10 L was used. The impeller speed of CFSR was maintained at 4 rpm during all runs. The CFSR was operated at 30°C by means of a thermostatic water bath circulator connected to the external double jacket of the cylindrical vessel. The sludge was recycled from the settling tank. A recycling pump was used to mix the influent substrate and sludge, hence to decrease possible substrate inhibition. The reactor pH value was controlled at 7.2 ± 0.3 by using HCl.

**MATERIALS AND METHODS**

**Experimental set up**

The experimental study for sulfide oxidation was conducted using nitrate as electron acceptor. Na\(_2\)S was used as the electron donor and prepared by considering the mass percentage of the hydrogen sulfide in biogas produced in anaerobic reactors. KNO\(_3\) was provided in cases where nitrate concentration was insufficient in the wastewater used. The wastewater was obtained from the outlet of aerobic wastewater treatment plant treating fermentation industry wastewaters. The activated sludge used in this study was obtained from aerobic wastewater treatment plant and adapted to substrates for 52 days. The reactor was operated under anoxic conditions to accomplish lithotrophic denitrification. The experimental set up installed in the laboratory is schematically shown in *Figure 1*. A glass continuous flow stirred tank reactor, with a working volume of 10 L was used. The impeller speed of CFSR was maintained at 4 rpm during all runs. The CFSR was operated at 30°C by means of a thermostatic water bath circulator connected to the external double jacket of the cylindrical vessel. The sludge was recycled from the settling tank. A recycling pump was used to mix the influent substrate and sludge, hence to decrease possible substrate inhibition. The reactor pH value was controlled at 7.2 ± 0.3 by using HCl.

![Figure 1](image-url) *The schematic diagram of experimental set up.*
Analytical methods

The samples were collected periodically for the analysis and all the determinations were performed in duplicate. Analytical methods used to determine suspended solids (SS), volatile suspended solids (VSS), alkalinity, and $S^{2-}$ were all performed according to “Standard Methods for the Examination of Water and Wastewater” (2005). The SS concentrations were determined according to the gravimetric method at 103°C. The VSS concentrations were analyzed via the gravimetric method at 550°C. Concentration of alkalinity was measured by using the titrimetric method. The $S^{2-}$ concentration was determined by the iodometric method whereas $SO_3^{2-}$ was measured by the refractometric method (RQflex Refractometer, Merck). $NO_3^-$, $NO_2^-$, COD, and $SO_4^{2-}$ values were all measured by spectrophotometric method with CADAS 30 spectrophotometer (DR. Lange). $NO_3^-$, $N O_2^-$, COD, and $SO_4^{2-}$ values were all measured by spectrophotometric method with CADAS 30 spectrophotometer (DR. Lange). TOC was analyzed at high temperature (850–900°C) with Ionics Model 1555B Carbon Analyzer. All chemicals used in the experiment were of analytical grade. Before the analysis, all samples were centrifuged for 10 minutes with Heraeus Sepatech Omnifuge 2.0 RS at 3,180 rpm.

Operating parameters

The influent concentration, hydraulic retention time, loading rate, $NO_3^-/S^{2-}$ molar ratio and removal efficiency are the operational and performance parameters of CFSR. In this study, the operational parameters of CFSR were maintained by using HRT, loading rate and influent $NO_3^-/S^{2-}$ molar ratio. The performance of the reactor during the experiment was assessed by using the removal efficiency. The lab-scale anoxic sulfide oxidation reactor (ASOR) was operated for 104 days to evaluate these parameters. The hydraulic retention times varying from 1 hour to 2 days have been used for the sulfide oxidation in the literature (Soares 2002; Reyes-Avila et al. 2004; Manconi et al. 2006; Perez et al. 2007; Mahmood et al. 2008; Li et al. 2009). In this study, the value of HRT varied between 2 and 86.4 h starting from the highest value then gradually decreased to find the optimum or minimum HRT. The HRT remained at 2 h from the day 75 to 104. Influent flow rate was gradually increased to control the HRT.

RESULTS AND DISCUSSION

Process efficiency

The process efficiency of CFSR was evaluated according to the changes of hydraulic retention time, loading rate and $NO_3^-/S^{2-}$ molar ratio. The wastewater, in which $S^{2-}$ and $NO_3^-$ were dissolved, was normally obtained from the nitrification stage of industrial wastewater treatment plant and contained almost only the inert COD. The TOC concentrations between inlet and outlet almost corresponded to each other and did not differ significantly. The influent contained only inert COD that was originated from the nature of the wastewater used in the fermentation industry. This COD left the reactor unchanged. On the other hand, the COD analysis of influent and effluent wastewater showed that an average of 35% COD removal took place in the reactor. This reduction in COD between inlet and outlet almost corresponded to each other and did not differ significantly. The influent contained only inert COD that was originated from the nature of the wastewater used in the fermentation industry. This COD left the reactor unchanged. On the other hand, the COD analysis of influent and effluent wastewater showed that an average of 35% COD removal took place in the reactor. This reduction in COD between inlet and outlet of the reactor was only due to the contribution of sulfide present in the wastewater. Therefore it can be concluded that sulfide removal taking place in the reactor was by lithotrophic denitrification and not due to organotrophic denitrification. We have also previously studied the possibility of chemical sulfide oxidation in the same wastewater. Li et al. (2009) implied that the sulfide oxidation process was rapid and a shorter HRT should be more suitable for the complete removal of sulfide from wastewater. In this study, high sulfide efficiency was also obtained for shorter HRTs. The flow rates were varied between $2.78 \times 10^{-3}$ and $120.48 \times 10^{-3}$ m$^3$/d which gave the possibility to operate at HRTs between 2 and 86.4 h. The volatile suspended solid concentration was kept at about 2.43 kg VSS/m$^3$ throughout the experiments by removing excess sludge from time to time. NaHCO$_3$ was added to the influent wastewater, so that the average alkalinity value was 1.98 kg/m$^3$, and a 28% alkalinity removal, in average, was observed in the reactor related with the inorganic carbon requirement. During the experiments the values of influent and effluent total organic carbon (TOC) and chemical oxygen demand (COD) have been monitored and analyzed in order to assess the presence of organotrophic denitrification.
of chemical sulfide removal was insignificant and dominant mechanism was biological (Yavuz et al. 2007).

The presence of nitrate and sulfide in stoichiometric amounts is an important factor for the growth of biomass and operation of the lithotrophic denitrifying system (Campos et al. 2008). The influent $S^-2$ and NO$_3$-N concentrations are given in Figures 2 and 3, respectively. The influent $S^-2$ concentrations were kept at about 0.163 kg/m$^3$ but the concentrations of NO$_3$-N in the influent were varying throughout the whole experimental period. The influent NO$_3$-N concentrations, supplied either synthetically or from nitrification plant, were between 0.047–0.760 kg/m$^3$ (Figure 3). In order to determine the optimal NO$_3$/S$^-2$ molar ratio on the process performance and to calculate the yield value, influent S$^-2$ concentration was kept constant and simultaneously influent NO$_3$-N concentration was gradually decreased from 0.435 to 0.047 kg/m$^3$ after the 74th day.

Loading rate is an imperative index to assess the potential of a bioreactor. Increased loading rates were achieved by reducing the HRT or increasing the influent concentrations. According to the previous investigations, the sulfide volumetric loading rate for biological treatment was between 0.042–0.294 kg/m$^3$d while the maximum nitrate loading rate varied between 0.175–0.594 kg/m$^3$d (Jing et al. 2007). Zhang (2004), Vaiopoulou et al. (2005) and Tanaka et al. (2007) stated that the loading rate of the nitrate should not exceed 0.3 kg N/m$^3$d to obtain the maximum nitrate removal. In this study, the S$^-2$ loading rates (SLRs) were increased from 0.055 kg/m$^3$d to 2.004 kg/m$^3$d but the SLR was kept at about 1.588 kg/m$^3$d after the 74th day while the HRT remained at 2 hour (Figure 2). In spite of the increase in SLR in relation with the decrease in HRT, the sulfide removal efficiency in the whole of the experiment exceeded 92%. Li et al. (2009) implied that shorter HRT should be more suitable for complete removal of sulfide from wastewater but in this study, hydraulic retention time, S$^-2$ loading rate and NO$_3$/S$^-2$ molar ratio did not affect the sulfide removal efficiency.

The more appropriate approach is to operate the reactor under stoichiometrically required amount of nitrate, avoiding nitrite accumulation (Manconi et al. 2007). In this work, NO$_3$-N removal efficiency is independent of HRT since NO$_3$-N loading rate (NLR) is changed by changing its concentration in the inlet besides the change in wastewater flowrate as shown in Figure 3. The nitrogen removal efficiency, as can be seen from Figures 3 and 4, changed in relation with NLR and NO$_3$/S$^-2$ molar ratio. Despite the average concentration value of 0.414 kg/m$^3$ NO$_3$-N in between days 72nd and 80th, nitrate removal efficiency is lower due to the high NLR. However, after the 80th day, the nitrate concentration and NLR were decreased to stoichiometrically required amount, then the nitrate removal efficiency increased up to 100% while maintaining highest sulfide removal. As the nitrate concentration is higher than stoichiometrically required amount, the percent nitrate removal is lower. At higher loading rates of 5.216 kg/m$^3$d and 3.839 kg/m$^3$d, nitrogen removal efficiency was between

![Figure 2](image1.png)

**Figure 2**: The effects of influent $S^-2$ concentration and $S^-2$ loading rate, SLR on the sulfide removal efficiency.

![Figure 3](image2.png)

**Figure 3**: The variation of influent NO$_3$-N concentration and NO$_3$-N loading rate, NLR as a function of time.
In the experimental studies, however, it was observed that the NO\textsubscript{3}/S\textsubscript{2} molar ratio also affected the nitrogen removal efficiency (Figure 4). When the NO\textsubscript{3}/S\textsubscript{2} molar ratio exceeded 4.5, the nitrogen removal efficiency stayed at 25% even in lower loading rates (1.039 kg/m\textsuperscript{3} d). However after the 74th day, the nitrate loading was further decreased. As the NO\textsubscript{3}/S\textsubscript{2} molar ratio decreased from 6.4 at the day 74 to 0.7, the percent nitrate removal gradually increased from 29% up to at least 98% respectively while maintaining highest sulfide removal.

Sulfur mass balance gives an indication as to the performance of the system. In order to assess the performance of sulfide oxidizing reactor, sulfur mass balance has been set up by analyzing different sulfur species such as S\textsuperscript{2−}, S\textsuperscript{0}, SO\textsubscript{3}\textsuperscript{2−}, and SO\textsubscript{4}\textsuperscript{2−} at the inlet and outlet of the reactor. But elemental sulfur could not be analyzed in this study due to the difficulty of separation of solid sulfur from biomass.

The S\textsuperscript{2−} concentration at the inlet of the reactor was kept at about 0.163 ± 0.03 kg/m\textsuperscript{3} and was almost zero at the outlet of the reactor. The SO\textsubscript{3}\textsuperscript{2−} concentrations at the inlet and outlet of the reactor were measured. It was found as insignificant (<0.020 kg/m\textsuperscript{3}), but also it was considered during the evaluation of the sulfur mass balance. The NO\textsubscript{3}/S\textsubscript{2} molar ratio affected the end products. When nitrate was limited or reactor was fed with excess amount of nitrate, the sulfide oxidation led to the formation of an unidentified intermediate and sulfate concentration decreased (Beristain-Cardoso et al. 2007). Elemental sulfur has also been reported to accumulate in chemolithotrophic denitrifying bioreactors, which treat sulfide, during periods of sulfide overloading (Reyes-Avila et al. 2004). In this work, the NO\textsubscript{3}/S\textsubscript{2} molar ratio was gradually decreased from about 7 to 0.6 throughout all the experiments. When both sulfide and nitrate were available at stoichiometric quantities, the denitrifying sludge was able to achieve the total oxidation of S\textsuperscript{2−} to SO\textsubscript{4}\textsuperscript{2−}, concomitant reduction of nitrate possibly to dinitrogen gas. Figure 5 shows mass balance with existing sulfur species. The main end product was sulfate in this study. On the other hand, under the nitrate limitations (NO\textsubscript{3}/S\textsubscript{2} = 0.7), elemental sulfur was observed as yellow solid particles in the reactor (period 101–104 days). The complete removal of nitrate and sulfide continued four days exchanging average 48 reactor volumes indicating the sustainability of the process resulting in elemental sulfur formation. Moreover, while the influent NO\textsubscript{3}/S\textsubscript{2} molar ratio was approximately 6.4 during the days 55–81, nitrite accumulation was observed in the reactor due to the presence of excessive nitrate.

![Figure 4](image1.png)  
**Figure 4** | The influence of NO\textsubscript{3}/S\textsuperscript{2} ratio (a) on sulfide and nitrate removal efficiency (b).

![Figure 5](image2.png)  
**Figure 5** | The total sulfur mass balance at the influent and effluent wastewaters.
theoretical yield values for nitrate were adopted from Yavuz et al. (2007). The experimental yield values, defined as \( Y_{S/N} \) (mol S\(^2\)/mol NO\(_3\)) are summarized in Table 2, as a function of inlet NO\(_3\)/S\(^2\) molar ratios. The yield values differ as a function of nitrate loading rates. As the nitrate loading rate decreases, the partial formation of elemental sulfur and other sulfur forms occur. When nitrate is limited, elemental sulfur is formed as an end product. Under these conditions, nitrate consumption and biomass production are relatively low compared to sulfate as an end product. As a result, elemental sulfur is the desired end product, in order to produce less biomass and to consume less energy, in the biological sulfide oxidation systems. It is known that less nitrate consumption would reduce the costs considerably. Furthermore, the elemental sulfur is non-toxic, stable, and insoluble in water so it can be recovered easily.

\[
\begin{align*}
S^2^- + 1.4272NO_3^- + 0.1872CO_2 + 1.4224H^+ \rightarrow SO_4^{2-} + 0.0568C_3H_7NO_2 + 0.5792H_2O + 0.6952N_2 \\
S^2^- + 0.3568NO_3^- + 0.0468CO_2 + 2.3556H^+ \rightarrow S^0 + 0.0092C_5H_7NO_2 + 1.1448H_2O + 0.1738N_2
\end{align*}
\]

The experimental values are in the range of theoretical limits (0.70 to 2.82 mol S\(^2\)/mol NO\(_3\)). The yield increases between lower and upper limits of the sulfide oxidation when the denitrification process is nitrate limited. The value of \( Y_{S/N} = 1.31 \) mol S\(^2\)/mol NO\(_3\) indicates that the end products of sulfide oxidation are mixture of sulfate and elemental sulfur. These results are in agreement with the recently published literature values, as shown in Table 4.

### The stoichiometry and rates of sulfide oxidation

The thermodynamically calculated yield values, for nitrate as electron acceptor, are presented in Table 1. The derivations of theoretical yield values for nitrate were adopted from Yavuz et al. (2007). The experimental yield values, defined as \( Y_{S/N} \) (mol S\(^2\)/mol NO\(_3\)) are summarized in Table 2, as a function of inlet NO\(_3\)/S\(^2\) molar ratios. The yield values differ as a function of nitrate loading rates. As the nitrate loading rate decreases, the partial formation of elemental sulfur and other sulfur forms occur. When nitrate is limited, elemental sulfur is formed as an end product. Under these conditions, nitrate consumption and biomass production

### Table 1 | Thermodynamically calculated yield values (\( Y_{S/N} \)) of sulfide oxidation when nitrate is used as the electron acceptor

<table>
<thead>
<tr>
<th>Time (d)</th>
<th>( Y_{S/N} ) (mol S(^2)/mol NO(_3))</th>
<th>( \text{Inf NO}_3^-/\text{inf S}^2^- ) (mol/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0–61</td>
<td>0.28 ± 0.08</td>
<td>7.3 ± 1.71</td>
</tr>
<tr>
<td>73–89</td>
<td>0.54 ± 0.1</td>
<td>6.4 ± 0.25</td>
</tr>
<tr>
<td>87–89</td>
<td>0.76 ± 0.12</td>
<td>2.7 ± 0.11</td>
</tr>
<tr>
<td>94–97</td>
<td>0.77 ± 0.08</td>
<td>1.4 ± 0.07</td>
</tr>
<tr>
<td>101–104</td>
<td>1.31 ± 0.13</td>
<td>0.7 ± 0.06</td>
</tr>
</tbody>
</table>

### Table 2 | Experimentally calculated yield values of sulfide oxidation when nitrate was used as the electron acceptor (\( T = 30^\circ C, \text{pH} = 7.2 \))

### Table 3 | Experimental yield values as function of end product, during the sulfide oxidation in the presence of nitrate as an electron acceptor

### Table 4 | Summary and comparison of literature and this study on the biological sulfide oxidation

<table>
<thead>
<tr>
<th>Reference</th>
<th>Electron acceptor</th>
<th>Volumetric rates (kg S(^2)/m(^3)/h)</th>
<th>Specific oxidation rates (kg S(^2)/kg VSS h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Buisman et al. (1991)</td>
<td>Oxygen</td>
<td>0.240</td>
<td>–</td>
</tr>
<tr>
<td>Buisman et al. (1991)</td>
<td>Oxygen</td>
<td>0.740</td>
<td>–</td>
</tr>
<tr>
<td>Nishimura &amp; Yoda (1997)</td>
<td>Oxygen</td>
<td>–</td>
<td>0.008</td>
</tr>
<tr>
<td>Barbosa et al. (2002)</td>
<td>Oxygen</td>
<td>–</td>
<td>0.003–0.0054</td>
</tr>
<tr>
<td>Yavuz et al. (2007)</td>
<td>Oxygen</td>
<td>2.286</td>
<td>0.062–0.234</td>
</tr>
<tr>
<td>Kleerebezem &amp; Mendez (2002)</td>
<td>Nitrate</td>
<td>0.125</td>
<td>–</td>
</tr>
<tr>
<td>Vaiopoulou et al. (2005)</td>
<td>Nitrate</td>
<td>0.0194</td>
<td>–</td>
</tr>
<tr>
<td>Manconi et al. (2006)</td>
<td>Nitrate</td>
<td>0.0063</td>
<td>–</td>
</tr>
<tr>
<td>Beristain-Cardoso et al. (2007)</td>
<td>Nitrate</td>
<td>–</td>
<td>1.25</td>
</tr>
<tr>
<td>Yavuz et al. (2007)</td>
<td>Nitrate</td>
<td>1.975</td>
<td>0.047–0.22</td>
</tr>
<tr>
<td>Jing et al. (2007)</td>
<td>Nitrate</td>
<td>0.202</td>
<td>0.003</td>
</tr>
<tr>
<td>This study</td>
<td>Nitrate</td>
<td>0.076</td>
<td>0.11</td>
</tr>
</tbody>
</table>
Table 3, as well as with theoretical predictions from the thermodynamic analysis.

Table 4 shows the results of calculated specific and volumetric sulfide oxidation rates obtained from this study, compared with those present in the literature. As it is reported, the volumetric rates and the specific oxidation rates were different from each other in all studies implying different experimental conditions. The specific oxidation rates were found higher compared to the values in the literature. As it is reported, the volumetric sulfide oxidation rates obtained from this study, reached 0.11 kg S \(/kg VSS \cdot h\). Specific sulfide oxidation rates \(q_{S}^{2}\) were calculated throughout the experimental periods and reached 0.11 kg S \(/kg VSS \cdot h\).

**CONCLUSIONS**

The oxidation of sulfide was carried out in continuous culture using nitrate as the electron acceptor in the presence of activated sludge as catalyst. The removal efficiencies of sulfide were observed more than 90% when the sulfide loading rates increased from 0.055 to 2.004 kg/m\(^3\) d. This study showed that nitrate, readily available in most wastewater treatment plants, could be used to oxidize sulfide. The end product could be either sulfate or elemental sulfur, depending on the ratio of nitrogen source to sulfide. As a result, the yield value was observed to be function nitrate/sulfide loading ratio. Therefore the end product of sulfide oxidation can be controlled by controlling inlet nitrate/sulfide ratio. The specific sulfide oxidation rates are comparable or better than those presented in the literature, but they could be further improved by retaining and enriching the sludge. Combining the anaerobic treatment, biogas production, and biogas cleaning with the aerobic treatment is possible. This approach allows the integration of sulfur and nitrogen cycles to improve sulfur emissions. The oxygen is used as the electron acceptor to control sulfide emissions in practice. On the other hand, nitrate has not so far been used in the industry. Combining sulfide removal with nitrate allows not only the control of sulfide but also improves nitrogen removal via lithotrophic denitrification without using a carbon source.

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