Short-term temperature impact on simultaneous biological nitrogen-sulphur treatment in EGSB reactor
Michal Sposob, Carlos Dinamarca and Rune Bakke

ABSTRACT

Sulphides are present in many wastewater streams; their removal is important due to corrosiveness, toxicity and unpleasant odour, and can be carried out by anaerobic biological treatment. This study focuses on the temperature effect (25–10 °C) on an expanded granular sludge bed (EGSB) reactor for sulphide removal using nitrate as electron acceptor. The reactor was run at a NO3-/HS- molar ratio of 0.35 and pH of 8.5–9.0. Samples were analysed by ion chromatography (NO3-, SO4 2- and S2O3 2-), spectrophotometry (S2-) and by scanning electron microscopy (SEM). S2- and NO3 removal was 99.74 ± 0.04 and 99.5 ± 2.9%, respectively. Sulphur (S0) was found on the outer granule surface and struvite inside the granule, by SEM. Sulphide conversion to sulphur was up to 76%. Temperature transitions and levels influenced S2O3 2- and SO4 2- concentrations.

Key words | anaerobic sulphide-nitrate removal, EGSB reactor, temperature

INTRODUCTION

Industrial wastewaters are often rich in sulphides such as petrochemical, tannery, electricity production, coal gasification and viscose rayon factories (Cai et al. 2008). Hydrogen sulphide and its ionic forms (HS-, S2-) are products of anaerobic decomposition of sulphur-rich organic substances, and reduction by sulphate-reducing bacteria (Pokorna & Zabranska 2015). Sulphides are unwanted in wastewater streams due to corrosiveness, toxicity and unpleasant odour. Anaerobic biological treatment is assumed to be a more sustainable solution than physico-chemical treatment, as it may be less complex and have less negative environmental impact (Gabriel & Deshusses 2003).

Bacterial species like Thiobacillus denitrificans are able to oxidize (anaerobically) sulphide to elemental sulphur (S0) and reduce nitrate to dinitrogen (Muyzer et al. 2013). Nitrate and nitrite as an electron acceptor are highly soluble in water and therefore favourable compared to oxygen (Cai et al. 2010). Utilization of nitrates and nitrites as electron acceptors for anaerobic sulphide oxidation have been demonstrated in several studies (Kleerebezem & Mendez 2002; Reyes-Avila et al. 2004; Mahmood et al. 2007; Dinamarca 2014).

Sulphides can be oxidized to elemental sulphur and/or oxidized to sulphate (SO4 2-) depending on quantitative presence of electron acceptor (NO3-). Thus, the ratio between electron acceptor and electron donor is an important factor for the degree of sulphide oxidation, as seen by the reaction stoichiometry (Equations (1) and (2)) with nitrate as electron acceptor (Kleerebezem & Mendez 2002).

$$3\text{HS}^- + 3.9\text{NO}_3^- + 0.2\text{NH}_4^+ + \text{HCO}_3^- + 1.7\text{H}^+ \rightarrow \text{CH}_1\text{H}_8\text{O}_5\text{N}_0\text{2} + 1.95\text{N}_2 + 3\text{SO}_4^{2-} + 2.3\text{H}_2\text{O} \hspace{1cm} (1)$$

$$14.5\text{HS}^- + 5\text{NO}_3^- + 0.2\text{NH}_4^+ + \text{HCO}_3^- + 20.3\text{H}^+ \rightarrow \text{CH}_1\text{H}_8\text{O}_5\text{N}_0\text{2} + 2.5\text{N}_2 + 14.5\text{S} + 17.4\text{H}_2\text{O} \hspace{1cm} (2)$$

Studies related to simultaneous carbon, nitrate and sulphide removal are focused on appropriate C/N/S ratio, load, pH or setup configuration adjustments (Reyes-Avila et al. 2004; Krishnakumar et al. 2005; Mahmood et al. 2007; Cai et al. 2008, 2010; Huang et al. 2016). Nonetheless, studies related to temperature impact were not performed. Biological processes are sensitive to temperature transitions and it is therefore recommended to limit variations in anaerobic digestion to less than 1 °C (Grady et al. 2011). Published research on simultaneous nitrogen-sulphur anaerobic treatment are limited to mesophilic conditions. Relevant wastewaters are often colder (psychrophilic conditions), thus, effects of lower temperatures are of interest.

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An expanded granular sludge bed (EGSB) reactor was used to obtain high biomass retention time and thereby high volumetric efficiency. The EGSB reactor principles also has the potential advantage that elemental sulphur as a product (Equation (2)) can be incorporated in the granular sludge so that sulphur surplus can be bled off as granules. The objective of the present study was to elucidate the short-term temperature impact (25–10 °C) on the reactants (nitrate, sulphide) and metabolites (elemental sulphur, sulphate and thiosulphate) at NO3−/HS− molar ratio = 0.35 (Equation (2)), alkaline pH (8.5–9.0) and invariable low-strength sulphide load.

MATERIAL AND METHODS

Inoculum and enrichment

The inoculum was taken from UASB methane reactor treating pulp and paper industry wastewater at Norske Skog Saugbrugs, Halden, Norway. The EGSB reactor was started by inoculating 0.25 L of sludge. The reactor was fed synthetic wastewater continuously at 25 °C with the same influent composition as for the trial period for one month in order to acclimatize the bacteria to the new substrates and enrich the sludge. The metabolism changed from methanogenic conditions to denitrifying lithoautotrophic conditions almost immediately, observed as no methane production and the presence of sulphur compounds in the effluent, during the acclimatization period since there were no organics in the feed but sulphide.

Synthetic wastewater

The EGSB reactor synthetic influent contained Na2S·9H2O (50 mg S/L) with NaHCO3 at concentration according to Equation (2), potassium phosphate buffer was used to adjust pH value. Nitric acid (HNO3) was used as a source of nitrate at concentration according to Equation (2), additionally nitrate feed contained following stock solutions: (A) NH4Cl (10 g/L), MgCl2·6H2O (10 g/L), CaCl2·2H2O (10 g/L); (B) K2HPO4 (300 g/L); (C) MnSO4·H2O (0.04 g/L), FeSO4·7H2O (2.7 g/L), CuSO4·5H2O (0.055 g/L), NiCl2·6H2O (0.1 g/L); ZnSO4·7H2O (0.088 g/L), CoCl2·6H2O (0.05 g/L), H3BO3 (0.05 g/L); (D) vitamin solution (Wolin et al. 1965), 10 times concentrated. HNO3, stock solutions A (10 mL/L), B (2 mL/L), C (2 mL/L) and D (1 mL/L) were dissolved in distilled water. Electron donor (as Na2S·9H2O) and acceptor (as HNO3) were fed from separate bottles to prevent contamination and reactions in the feed bottles (Figure 1).

Experimental setup

The laboratory-scale EGSB reactor was made of a polycarbonate tube with an inner diameter of 32 mm and an effective height of 620 mm, giving a working volume of 0.5 L (Figure 1). The reactor was maintained at variable temperatures (25–10 ± 0.1 °C) by cold plate cooler. Four different temperatures were tested under invariable influent composition, starting from 25 °C and then reducing temperature by 5 °C in three steps without waiting for steady state to establish at each temperature. A peristaltic pump introduced synthetic influent from a 20 L influent vessels (under nitrogen gas) to the reactor bottom at 2 L/d (hydraulic retention time, HRT = 6). Recycling pump was employed to mix the influent with sludge and expand the sludge bed (maintaining vertical velocity = 6 m/h). Reactor pH was maintained in range 8.5–9.0 and monitored by electrode on the recirculation loop.

Analytical procedures

Effluent samples were collected daily and analysed immediately. Nitrate (NO3−), sulphate (SO42−), and thiosulphate (S2O32−) in collected liquid samples (following 0.45 μm filtration) were measured by ion chromatography (Dionex ICS-5000). Sample separation and elution was performed using an IonPac AS11-HC 2 mm analytical column and potassium hydroxide (KOH) at 19 mM as the eluent. Sulphid concentration was determined by methylene blue method (APHA 1995), using spectrophotometer (Spectroquant UV/VIS Pharo 300). A pH meter (Hanna Instruments) determined the pH of liquid samples; a three-point calibration of pH meter was carried out.

Scanning electron microscopy

Sludge removed at the end of the experiment was examined microscopically. Scanning electron microscopy (SEM) and energy-dispersed X-ray (EDX) analysis were done by a scanning electron microscope (Hitachi SU3500) at 20 kV. Prior to analysis, intact granules were dehydrated with a graded series of ethanol (30, 60 and 100%) and coated with gold by magnetron sputtering. The manual sectioning was performed after the sample was dried. EDX analysis were orientated on detection of elements and their spot spatial distribution.
RESULTS AND DISCUSSION

The reactor performance was recorded daily over 36 days. \( \text{S}_2^2^- \) and \( \text{NO}_3^- \) removal remained stable at 99.74 ± 0.04 and 99.5 ± 2.9%, respectively, at sulphur load of 0.2 kg \( \text{S}/\text{m}^3\text{d} \) (Table 1). Overall process outcomes at the different temperatures are presented in Table 2. Nitrate presence in the effluent was detected only once (day 24), through the trial period. Sulphide removal rate did not exhibit any significant variability even at the lowest temperature (10 °C). Thus, temperature levels and transitions had little impact on the percentage of sulphide removal.

Collected effluent samples were visually characterized by a yellow ‘straw’ colour (before and after 0.45 μm filtration). Chen et al. (2008) and Krishnakumar et al. (2005) have observed similar phenomena. It suggests the

![Figure 1](https://iwaponline.com/wst/article-pdf/74/7/1610/458300/wst074071610.pdf)

**Table 1** Experimental conditions and effects on EGSB reactor performance given as average values for the entire test period

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>( \text{NO}_3^- /\text{HS}^- ) molar ratio</td>
<td>0.35</td>
</tr>
<tr>
<td>HRT (h)</td>
<td>6</td>
</tr>
<tr>
<td>Influent pH</td>
<td>8.5–9.0</td>
</tr>
<tr>
<td>Vertical velocity (m/h)</td>
<td>6</td>
</tr>
<tr>
<td>Effluent pH</td>
<td>8.76 ± 0.31</td>
</tr>
<tr>
<td>( \text{S}_2^2^- ) influent (mg/L)</td>
<td>50</td>
</tr>
<tr>
<td>( \text{S}_2^2^- ) removal (%)</td>
<td>99.74 ± 0.04</td>
</tr>
<tr>
<td>\text{NO}_3^- influent (mg/L)</td>
<td>33.3</td>
</tr>
<tr>
<td>\text{NO}_3^- removal (%)</td>
<td>99.5 ± 2.9</td>
</tr>
</tbody>
</table>

**Table 2** Process outcome under different temperatures

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>( \text{S}_2^2^- ) removal (%)</th>
<th>\text{NO}_3^- removal (%)</th>
<th>( \text{SO}_4^{2-} ) (mg/L)</th>
<th>( \text{SO}_3^{2-} ) (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>25</td>
<td>99.8</td>
<td>&gt;99.7(^a)</td>
<td>23.1 ± 9.9</td>
<td>44.2 ± 14.8</td>
</tr>
<tr>
<td>20</td>
<td>99.7</td>
<td>&gt;99.7</td>
<td>25.8 ± 8.6</td>
<td>49.3 ± 13.6</td>
</tr>
<tr>
<td>15</td>
<td>99.7</td>
<td>98.1</td>
<td>30.2 ± 6.1</td>
<td>40.4 ± 30.8</td>
</tr>
<tr>
<td>10</td>
<td>99.8</td>
<td>&gt;99.7</td>
<td>42.4 ± 41.0</td>
<td>81.0 ± 16.6</td>
</tr>
</tbody>
</table>

\(^a\) > 99.7 % nitrate removal implies that nitrate was not detected in the effluent samples (nitrate detection limit is 0.1 mg \( \text{NO}_3^- /L \)).
presence of unattached and freely dispersed elemental sulphur/polysulphides.

**Sulphate and thiosulphate formation under temperature transitions**

The measured reactor effluent $\text{SO}_4^{2-}$ and $\text{S}_2\text{O}_3^{2-}$ concentrations are presented in Figure 2. $\text{S}_2\text{O}_3^{2-}$ concentration (3.3–97.2 mg/L) was predominantly higher than $\text{SO}_4^{2-}$ (10.1–58.6 mg/L), except for days 22–26. No obvious explanation is found for this behaviour but the following observations may give some clues. In comparison to $\text{S}_2\text{O}_3^{2-}$, the average $\text{SO}_4^{2-}$ concentration was less variable during the different temperature levels, except for an increase at 10 °C.

The most significant changes in the process performance are $\text{S}_2\text{O}_3^{2-}$ concentration variations at 15 °C and 10 °C. $\text{S}_2\text{O}_3^{2-}$ tended to increase through the experiment but not while operated at 20 °C. Temporary $\text{S}_2\text{O}_3^{2-}$ concentration drops were observed following each temperature change: days 14–15 (20 °C), 22–26 (15 °C) and day 32 (10 °C). The lowest $\text{S}_2\text{O}_3^{2-}$ concentrations were obtained at 15 °C when it decreased below $\text{SO}_4^{2-}$ concentration until day 26. $\text{S}_2\text{O}_3^{2-}$ reached the lowest value (3.3 mg/L) on day 25.

The lower $\text{S}_2\text{O}_3^{2-}$ concentrations coincided with decreased sample colour intensity during days 22–26, suggesting reduced presence of unattached and freely dispersed elemental sulphur/polysulphides when $\text{S}_2\text{O}_3^{2-}$ was low. The utilized analytical method assumes sample elution under potassium hydroxide (eluent). The probable elemental sulphur/polysulphides present in the analysed samples can react with the eluent, resulting in $\text{S}_2\text{O}_3^{2-}$ (Tartar 1915). Thus, the measured $\text{S}_2\text{O}_3^{2-}$ may also represent the oxidized elemental sulphur/polysulphide. Given this characteristic of the performed analysis, it is troublesome to estimate which part of the detected $\text{S}_2\text{O}_3^{2-}$ represents the effect of the reaction between sample and eluent. In consequence, accurate quantification of unattached elemental sulphur/polysulphide will require another analytical approach to be developed. This analytical uncertainty is carefully considered in the interpretation of the experimental observations in sections below. It seems as if this analytical challenge has been overlooked in previously published studies on this topic. Resolving this issue by an improved analytical procedure that quantitatively distinguish between $\text{S}_2\text{O}_3^{2-}$ and oxidized elemental sulphur/polysulphide, in future studies, may lead to a deeper understanding of mechanisms involved.

The presence of $\text{SO}_4^{2-}$ and $\text{S}_2\text{O}_3^{2-}$ increased after day 26 with dominating concentration of $\text{S}_2\text{O}_3^{2-}$ over $\text{SO}_4^{2-}$. At the last experimental period (at 10 °C) both $\text{SO}_4^{2-}$ and $\text{S}_2\text{O}_3^{2-}$ started to increase but not simultaneously, $\text{S}_2\text{O}_3^{2-}$ increased until day 34 reaching 97.2 mg/L. $\text{SO}_4^{2-}$ decreased when $\text{SO}_4^{2-}$ concentration increased at the end, suggesting a
mutual dependence between these components. Several reports show that when reduced sulphur compounds are completely consumed, the elemental sulphur and \(S_2O_3^{2-}\) are oxidized to \(SO_2^{4-}\) (Shively 1974; Holkenbrink et al. 2011). By oxidation of reduced sulphur compounds (sulphide, elemental sulphur, thiosulphate) *Thiobacillus* bacteria derives energy (Schedel & Trüper 1993). It leads to \(SO_2^{4-}\) excess, \(S_2O_3^{2-}\) depletion and size reduction of sulphur globules. These phenomena seem to occur from day 33 until the end of the trial period, however, with an apparent \(S_2O_3^{2-}\) increase. This is not in accordance with above references and can perhaps be partly explained by observations by Rebac et al. (1995) who observed granule diameter decrease by erosion (as further discussed below based on microscopy).

**Effluent sulphur balance**

An indirect method to quantify sludge-incorporated sulphur based on an overall process mass balance is applied since direct quantitative measurements of incorporated sulphur in anaerobic granular sludge was not readily obtained. The sulphur balance, calculated based on the effluent concentrations of sulphate-sulphur (\(SO_4^{2-} - S\)), thiosulphate-sulphur (\(S_2O_3^{2-} - S\) and sulphide (\(S^{2-}\)), according to Equation (3) is presented in Figure 3. Due to alkaline pH maintained in the EGSB reactor, presence of \(H_2S\) in the gas phase is neglected in the balance.

\[
[S]_{\text{eff}} = [S^{2-}]_{\text{eff}} + [SO_4^{2-} - S]_{\text{eff}} + [S_2O_3^{2-} - S]_{\text{eff}}
\]

Effluent data points located below the constant \(S^{2-}\) influent concentration profile represent sulphur incorporation, while points over the influent \(S^{2-}\) level imply sulphur release from the reactor (Figure 3). The amount of sulphur incorporated varied during the trial period, ranging from 7 to 76% of inlet sulphur, reaching the highest level at 15 °C, coinciding with the lowest \(S_2O_3^{2-}\) measurements (Figure 2). Possible mechanisms leading to large variations in the incorporation of elemental sulphur into granular sludge in the EGSB reactor are discussed as follows. The highest obtained sulphur incorporation coincided with a temporary sludge bed expansion lasting no longer than 72 h. The enhanced sulphur incorporation coinciding with bed expansion did not occur immediately after the temperature transitions but with a delay of 24–48 h. The temporary sludge expansion was visually estimated as a doubling of the sludge bed height. This did not cause sludge washout since the sludge bed filled less than half the reactor height prior to the sludge bed expansion. This study, in contrast to Huang et al. (2016), who studied the regulation of elemental sulphur recovery, reveals that even with the same reactor construction, sludge concentration, sulphur load and same quantity of microbial community, enhanced sulphur incorporation is possible. In addition, in this case, \(S_2O_3^{2-}\), not \(SO_4^{2-}\), was the key variable that co-varied with sulphur incorporation. The elemental sulphur/polysulphides present, observed as yellow colour and by increased \(S_2O_3^{2-}\), seems to be available for sludge incorporation under appropriate sludge bed levels (sludge bed expansion coincided with less \(S_2O_3^{2-}\) and yellow colour). It suggests that higher vertical flow velocity to expand the sludge bed may be a method to enhance the mass transfer to obtain more
efficient sulphur incorporation. The interrelation between sludge expansion and $S_2O_3^{2-}$ concentration may play an important role in sulphur incorporation, not yet understood. 57.2% of $S_2O_3^{2-}$ molar mass is sulphur, thus better understanding of enhanced $S_2O_3^{2-}$ (which, as measured, may include elemental sulphur/polysulphides) removal is critical.

The sulphur balance exceeds a threshold on day 29, at the end of the temperature period at 15°C when effluent sulphur reached 51.9 mg S/L which is above $S_2$− influent concentration (50 mg/L). From day 29 until the end of experiment (except day 32), effluent total sulphur was exceeding this threshold, reaching a maximum value on day 34 (69.2 mg S/L). Effluent sulphur concentration exceeding influent $S_2^−$ at the end of the 15°C and during most of the 10°C periods may suggest changes in the granular sludge characteristic, which led to removal of earlier incorporated sulphur, stored as bacterial inclusion bodies (Muyzer et al. 2013). The loss of total sulphur in the effluent towards the end of the present study suggests a relatively unstable sulphur incorporation over the experimental period. Thus, any fluctuations away from optimum conditions, such as temperature changes, can result in decreased reactor performance. As observed here, such changes may be transient and the biomass may adapt to the new conditions.

**Microscopy**

Examined granular sludge had a slightly oval shape with dimensions 1.0 × 0.75 mm (length × width) and 0.95 × 0.75 mm (Figure 4). The granules are characterized by a porous outer surface and the presence of unidentified black spots. EDX analysis by SEM showed these black spots do not correspond with outer structural composition variability.

Further analysis carried out by SEM coupled with EDX performed on both whole granules and manually sectioned granules shows significant differences between external and internal surfaces. Some distinct local differences between internal and external surfaces are seen in Figure 5. Sulphur was mostly present on the outer surface of the granules where it had the high abundance. Iron and phosphorus presence was also considerable on the outer surface. Elemental sulphur evidently accumulated mainly on the outer layers without deeper penetration. It implies that sulphur incorporated during some experimental stages can be easily removed from the granular sludge surface at other stages. The excess effluent sulphur at 10°C is, therefore, probably a result of erosion type detachment from the granular sludge surfaces, such as observed by Rebac et al. (1995).

The sulphur present on the granules surface may exist in two forms. One is iron sulphide (FeS) formed as a purely chemical precipitation product, where the precipitated particles are incorporated into the granular sludge mass. Because kinetics of metal sulphides particles formation is fast, only small particles with poor settling abilities are formed (Villa-Gomez et al. 2012). In the present case, the FeS observed is, however, accumulating on granules with good settling abilities. The amount of added iron-containing compound (FeSO₄·7H₂O) in the reactor feed could lead to maximum precipitation of about 2.5% of influent $S_2^−$. Other metal sulphide particles may also be present on the granule surface, but their abundance is assumed minor. The second main form of sulphur is assumed to be a purely biologically-driven sulphur production according to Equation (2). The sulphur abundance compared to iron suggests that this is a large fraction of the elemental sulphur present on the granules’ surface.

**Figure 4** | Granules appearance by SEM: intact granules (left), cross-section (right).
The presence of elongated, crystal-like, narrow objects present in relatively high abundance embedded in extracellular polymeric substance (EPS) was observed on the internal surface of the manually sectioned granular sludge. EDX examination of these crystal-like objects manifested the qualitative presence of phosphorus, magnesium and oxygen in the highest abundances, respectively (Figure 5). Detected elements are key components of struvite (MgNH₄PO₄·6H₂O). Struvite formation can occur when the concentrations of phosphorus, magnesium, and oxygen ions exceed the solubility product for struvite (Doyle & Parsons 2002). The maintained alkaline pH in the reactor led to suppression of struvite solubility, but if pH rises above a pH of 9, the solubility of struvite would increase (Snoeyink & Jenkins 1980). The observation that struvite probably is present in the granular sludge suggests that such crystals may be part of the granular framework in the investigated process.

Granular sludge generated in a sulphide removal process must be bled off in a regular way to remove excess sludge and maintain stable operation over time. Given the high mineral and nutrient content of the granules from the sulphide-nitrate treatment, which are rich in elements like magnesium, nitrogen, potassium and sulphur (Figure 5), they appear to display exceptional fertilizer traits. Some parts are probably easily available for plant uptake while struvite is characterized by slow nutrient release and a low salt index (Latifian et al. 2012). Struvite is known to form on surfaces in many anaerobic digestion processes causing severe problems such as pipes clogging. Struvite formation inside granules that are recovered from sludge bed reactors would not cause such problems and may even be a way to avoid struvite formation on surfaces.

Further investigations to better understand what influences the formation of sulphur species in such reactors in relation to temperature and sludge bed expansion will be carried out in longer experimental trials.

CONCLUSIONS

The results of the present study show that an EGSB reactor is able to treat low strength sulphide-laden wastewater under
variable temperature conditions (25–10 °C) with 99.74 ± 0.04 and 99.5 ± 2.9% S²⁻ and NO₃⁻ removal, respectively, with little influence by temperature levels or transitions.

Temperature variations between 25–20 °C had little impact on SO₂⁻ and S₂O₃⁻ formation while increasing concentrations of S₂O₃⁻ and SO₂⁻ were observed at 15–10 °C.

The amount of sulphur incorporated in the sludge bed was up to 76% of influent sulphide.

The granular bed temporarily expanded after the imposed 5 °C temperature drops, significantly influencing S⁰ incorporation. Sulphur incorporation appeared to be enhanced by granular sludge bed expansion.

The measured S₂O₅⁻ concentration probably includes varying degrees of elemental sulphur/polysulphide due to the applied analytical procedure (KOH elution) may convert these to S₆O₇⁻.

The effluent sulphur (sulphate-sulphur (SO₄²⁻–S), thiosulphate-sulphur (S₂O₃²⁻–S) and sulphide (S²⁻)) exceed the S²⁻ influent concentration at the end of the 15 °C period and the whole 10 °C period, suggesting loss of earlier incorporated sulphur from the granular sludge bed.

Sulphur and iron, probably as FeS and elemental sulphur, on the granular sludge surface were observed by microscopic analysis.

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REFERENCES


Gabriel, D. & Deshusses, M. A. 2003 Retrofitting existing chemical scrubbers to biotrickling filters for H₂S emission control. Proceedings of the National Academy of Sciences 100 (11), 6308–6312.


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