Anaerobic treatment of sulfate-containing municipal wastewater with a fluidized bed reactor at 20 °C

B. Düppenbecker and P. Cornel

ABSTRACT

This study focuses on the anaerobic treatment of sulfate-containing municipal wastewater at 20 °C with a fluidized bed reactor. Mean influent chemical oxygen demand (COD) and sulfate concentrations were 481 and 96 mg/l. The response of the COD removal efficiency to increasing organic loading rates (OLR) was investigated. Average total COD removal was 61% at OLR between 2.7 and 13.7 kg COD/(m³·d) and did not distinctly depend on the OLR. To assess the removal efficiency in more detail the COD in- and output mass flows were balanced. The results showed that only 11–12% of the input COD was recovered as gaseous methane. About 12–13% of the input COD remained in the effluent as dissolved methane. Furthermore, a distinct amount of 12–19% of the input COD remained in the reactor as settled sludge and was not further biologically degraded. Due to the reduction by sulfate-reducing bacteria, 13–14% of the input COD was degraded. Further adverse impacts of the influent sulfate on the anaerobic treatment process are discussed as well.

B. Düppenbecker (corresponding author)  
P. Cornel  
Technische Universität Darmstadt,  
Institute IWAR,  
Franziska-Braun-Str. 7,  
64287 Darmstadt,  
Germany  
E-mail: b.dueppenbecker@iwar.tu-darmstadt.de

Key words | anaerobic treatment, fluidized bed, low temperatures, municipal wastewater, sulfate

INTRODUCTION

The anaerobic treatment of municipal wastewater still represents a challenge for wastewater engineers. At low temperatures and substrate concentrations anaerobic treatment systems should enable high biomass retention at high hydraulic loading rates as well as sufficient turbulence to overcome mass transfer limitations (Lettinga et al. 2001). Both demands can be met via the immobilization of biomass on a fluidized carrier. Hence, the fluidized bed reactor (FB) might represent a promising technology for the anaerobic treatment of municipal wastewater. The feasibility of the anaerobic FB for municipal wastewater treatment at low temperatures was shown by Jewell et al. (1981), Yoda et al. (1985) and Sanz & Fdz-Polanco (1990). Recent studies focused on the combination of the anaerobic FB and micro- or ultrafiltration (Yoo et al. 2012; Gao et al. 2014; Shin et al. 2014). However, in the presence of sulfate, the degradation of organic matter by sulfate-reducing bacteria (SRB) impairs the recovery of valuable methane gas. SRB are able to metabolize a wide variety of organic compounds, such as acetate, propionate or glucose as well as hydrogen (Hansen 1993; Muyzer & Stams 2008). Equation (1) shows the conversion of acetate by SRB.

\[ CH_3COO^- + SO_4^{2-} \rightarrow HS^- + 2HCO_3^- \] (1)

Hence, 64 g chemical oxygen demand (COD) is required to reduce 1 mole sulfate. Previous studies have shown that SRB will outcompete methanogenic archaea during the treatment of synthetic low-strength wastewater (Harada et al. 1994) and mixtures of acetate and propionate (van den Brand et al. 2014). Therefore, it can be assumed that sulfate is completely reduced during the anaerobic treatment of municipal wastewater. Table 1 shows COD and sulfate influent concentrations of anaerobically treated municipal wastewater. At a COD/sulfate ratio of 5.0 (present study), 13% of the influent COD is theoretically degraded by sulfate reduction and not available for methane production. Furthermore, at decreasing temperatures the solubility of methane rises and a significant amount of methane remains dissolved in the effluent (Souza et al. 2011). Considering these issues, the objective of the present study is the critical evaluation of the in- and output COD mass flows during
the anaerobic treatment of sulfate-containing municipal wastewater at 20 °C.

**MATERIAL AND METHODS**

The reactor consisted of an acrylic tube (height 700 mm, diameter 24 mm). The volume (317 ml) was defined as reaction zone for calculating the organic loading rate (OLR) and hydraulic retention time (HRT). Feed and recirculation inflows were introduced in a lower part (height 40 mm, diameter 24 mm). A wire cloth was fixed between both sections. Furthermore, glass beads were filled in as support layer. A cylindrical settler was attached on top of the reactor (height 150 mm, diameter 72 mm). A second cylindrical settler (height 180 mm, diameter 72 mm) was connected in series to catch carry-over carrier material. Peristaltic pumps were used for recirculating (ProMinent, Germany) and introducing feed (IDEX, USA). The recirculation rate was adjusted using a flow meter (GEMÜ, Germany). The reactor was operated at 20 °C in a climate chamber. The operating conditions are shown in Table 2. By increasing the influent flow rate, the OLR increased (and hence the HRT decreased) gradually in each mode, although the OLR varied in each mode as a result of fluctuating COD influent concentrations. Due to the high recirculation rate the reactor can be considered as a completely stirred tank reactor. The reactor was fed with municipal wastewater (160 μm screened) from the wastewater treatment plant Darmstadt Süd (Darmstadt, Germany). Wastewater parameters are shown in Table 1. The wastewater was stored in a refrigerated, stirred feed tank at about 5–10 °C and replaced weekly. Total and dissolved COD concentrations were verified three times a week and did not change significantly.

COD and sulfate concentrations were determined by cuvette tests (Hach, USA). Dissolved COD (COD\textsubscript{diss}) and sulfate samples were filtrated using a 0.45 μm syringe filter (VWR, USA). Samples were homogenized with a dispersion tool (IKA, Germany) before determining total COD (COD\textsubscript{tot}). The particulate COD (COD\textsubscript{part}) results from the difference of COD\textsubscript{tot} and COD\textsubscript{diss}. Before analyzing the effluent COD 10 μl of sulfuric acid (4 mol/l) per ml sample were added. The samples were then vibrated in open vials for 10 min on a platform shaker (Heidolph, Germany) to strip hydrogen sulfide and dissolved methane. Sulfide removal was verified by determination of residual sulfide. Extension of the pretreatment time from 10 to 30 min did not result in a distinct further decrease of the COD. Therefore, it was assumed that the dissolved methane was completely stripped during the above-mentioned pretreatment procedure. Volatile suspended solids (VSS) were determined according to *Standard Methods* (APHA 2005)

### Table 1 | COD and sulfate influent concentrations of anaerobically treated municipal wastewater

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Temperature</strong></td>
<td>(° C)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Temperature</td>
<td>20\textsuperscript{a}</td>
<td>21</td>
<td>10</td>
<td>23</td>
<td>15–25</td>
</tr>
<tr>
<td>COD\textsubscript{tot}</td>
<td>(mg/l)</td>
<td>481 (224–759)\textsuperscript{b}</td>
<td>720</td>
<td>143</td>
<td>256</td>
</tr>
<tr>
<td>COD\textsubscript{diss}</td>
<td>(mg/l)</td>
<td>148 (106–315)\textsuperscript{b}</td>
<td>84</td>
<td>390</td>
<td>52</td>
</tr>
<tr>
<td>SO\textsubscript{4}\textsuperscript{2–}</td>
<td>(mg/l)</td>
<td>96 (53–122)\textsuperscript{b}</td>
<td>112</td>
<td>200</td>
<td>41</td>
</tr>
<tr>
<td>COD/SO\textsubscript{4}\textsuperscript{2–}</td>
<td>(–)</td>
<td>5.0 (3.5–7.3)\textsuperscript{b}</td>
<td>6.4</td>
<td>3.8</td>
<td>3.5</td>
</tr>
</tbody>
</table>

\textsuperscript{a}Operating temperature of the bioreactor.

\textsuperscript{b}Minimum–maximum value.

### Table 2 | Operating conditions of the FB (arithmetic averages)

<table>
<thead>
<tr>
<th>Mode</th>
<th>Start-up</th>
<th>I</th>
<th>II</th>
<th>III</th>
</tr>
</thead>
<tbody>
<tr>
<td>Operating time</td>
<td>(d)</td>
<td>0–35</td>
<td>57–91</td>
<td>92–126</td>
</tr>
<tr>
<td>HRT (h)</td>
<td></td>
<td>5.7</td>
<td>3.0</td>
<td>1.9</td>
</tr>
<tr>
<td>Influent flow rate (l/h)</td>
<td>0.056</td>
<td>0.108</td>
<td>0.168</td>
<td>0.222</td>
</tr>
<tr>
<td>OLR (kg COD\textsubscript{tot}/(m\textsuperscript{3}·d))</td>
<td>2.1</td>
<td>3.8</td>
<td>6.1</td>
<td>8.5</td>
</tr>
<tr>
<td>Recirculation rate (l/h)</td>
<td>15</td>
<td>15</td>
<td>15</td>
<td>15</td>
</tr>
<tr>
<td>Upflow velocity</td>
<td>(m/h)</td>
<td>33</td>
<td>34</td>
<td>34</td>
</tr>
</tbody>
</table>
using glass fiber filters and ignition at 500 °C (Whatman GF 92, UK). Oxidation reduction potential and pH (Sensolyt, WTW, Germany) electrodes were installed in the settler and connected to a pH meter (pH 191, WTW, Germany).

Granular activated carbon (GAC) (8 × 30 mesh, Donau Carbon, Germany) was chosen as carrier material. GAC was sieved to sort out particles larger than 1 mm. Before starting the test phase, the biofilm on the GAC was cultivated for more than 12 months using a sulfate-containing sucrose-based synthetic medium. The settled immobilized sludge volume (SISV) of the GAC (covered with biofilm) was determined in a 100 ml graduated cylinder (diameter 27 mm) filled with tap water, after sedimentation to constant volume. In order to determine the immobilized volatile suspended solids (IVSS) the biofilm was separated carefully with a scalpel from the GAC of 5 ml SISV and suspended in deionized water. Then, the GAC was rinsed with deionized water to remove residual biofilm, and VSS of the suspension was determined. The mass of dried GAC was determined by drying the GAC at 105 °C. The volume of the GAC was calculated by assuming an apparent density of 505.5 kg/m³. Estimated SISV, IVSS and sludge loading rate (SLR) at start and end of operating time are shown in Table 3.

To determine VSS and COD of the settled particulate organic matter the sludge of both settlers was removed at the end of each operating mode. The sludge was 0.45 μm filtered (Whatman ME 25, GE, UK) using a pressure filtration equipment. The filter cake was washed by filtration using 100 ml deionized water. Afterwards, the filter cake was suspended in deionized water and COD tot was determined. The gas flow rate was measured volumetrically (MilliGascouter, Ritter, Germany). The gas composition (CH₄, CO₂, N₂, O₂) was determined using a gas chromatograph equipped with a flame ionization detector and thermal conductivity detector (Agilent Technologies, USA). For analysis a 5 ml gas sample was taken with a syringe from the gas collection system. During each operating mode the fraction of methane increased to a constant value. For calculating the methane volume flow the steady state values were chosen, assuming that they represent the composition of the produced gas. For determination of dissolved methane a glass vial (25 ml) was filled with 4 to 6 g NaCl (Merck, Germany) and sealed with a septum. Vacuum was generated in the vial and 7 to 12 ml of liquid sample were sucked from the reactor into the vial. After shaking the samples for 30 min at 20 °C, pressure was measured and a gas sample was taken from the headspace. The gas volume was determined by filling the vial with a saturated solution of NaCl.

Dissolved methane and gaseous hydrogen sulfide were calculated according to Henry's law. Henry's law constants were calculated based on data from NIST (2015). Dissolved methane concentrations were calculated by considering the above-mentioned variation of the gas composition. Concentrations of dissolved (undissociated) hydrogen sulfide (H₂S diss) were calculated by assuming total sulfate reduction and considering the dissociation of H₂S in aqueous solution as shown in Equation (2) (Garrels & Christ 1965).

$$H_2S \leftrightarrow HS^- + H^+ \quad(2)$$

Within the relevant pH range of 6.5–8.2 (Speece 1996) the second dissociation step can be neglected. Thus, the amount of H₂S diss can be calculated according to Equation (3). Here, C₀ (mol/l) is the total dissolved sulfide concentration and K is 10⁻⁷ mol/l (Garrels & Christ 1965).

$$H_2S_{\text{diss}} = C_0 \cdot [H^+] / (K + [H^+]) \quad (3)$$

### RESULTS AND DISCUSSION

**Reactor performance**

In Figure 1, the volumetric and percentage removal efficiency during Mode I, II and III (operating conditions of each mode are shown in Table 2) of total, particulate and dissolved COD are plotted against the OLR. A strong influence of OLR on the COD tot removal is not discernible. A closer look at the average COD tot removal, which was 65% in Mode I and 59% in Mode II and III, confirms this fact. The results agree well with previous studies of Jewell et al. (1981), Yoda et al. (1985), Sanz & Fdz-Polanco (1990).
and Yoo et al. (2012), who reported that increasing OLR (at decreasing HRT) did not impact the COD removal during the treatment of municipal wastewater in an anaerobic FB. However, Jewell et al. (1987) and Yoda et al. (1985) observed a strong decrease in removal efficiency at HRT lower than 1 h. In the present study the HRT was always higher than 1 h.

It should be mentioned that Jewell et al. (1987) and Sanz & Fdz-Polanco (1990) reported distinctly higher COD$_{tot}$ removal rates of about 70–80%. Considering that in the present study coarse particulate organic matter was removed by 160 μm screening, it is assumed that the reported higher removal efficiencies are attributed to accumulation of coarse particulate organic matter within the reactor. Yoo et al. (2012), for instance, reported a lower COD$_{tot}$ removal of 53% using 10 μm screened municipal wastewater at HRT of 1.5 h (comparable to HRT in Mode III of this study), though at low influent COD$_{tot}$ concentrations of about 152 mg/l and low OLR of about 2.9 kg COD$_{tot}$/m$^3$·d, respectively.

As Figure 1 shows, the COD$_{part}$ removal acted comparably to the COD$_{tot}$ removal. Nevertheless, the COD$_{part}$ removal was slightly higher. Consequently, the COD$_{diss}$ removal has to be lower and the average removal was 50% in Mode I, 40% in Mode II and 35% in Mode III. In contrast to the COD$_{tot}$ and COD$_{part}$, the removal of COD$_{diss}$ seemed to decrease with increasing OLR (and hence with decreasing HRT). Due to the fact that the particulate fraction of the COD$_{tot}$ is about 70%, the COD$_{tot}$ removal is more influenced by the COD$_{part}$ removal. Compared to the removal efficiencies of upflow anaerobic sludge blanket (UASB) reactors it can be said that the FB reaches comparable removal efficiencies at higher OLR and lower HRT, respectively. Urban et al. (2007), for instance, reported a COD$_{tot}$ removal of 60% at HRT of 10 h, feeding wastewater with comparable influent parameters. Seghezzo (2004) reported a COD$_{tot}$ removal of 55% at HRT of about 5 h. However, the influent concentrations in his study were distinctly lower (COD$_{tot}$ about 153 mg/l) than that of the present study.

**Methane production and biogas composition**

It is a well-known phenomenon that during the treatment of low-strength wastewater at low temperatures the methane fraction of the produced biogas is comparatively low (Lettinga et al. 1985; Yoda et al. 1985; Giménez et al. 2012; Yoo et al. 2012). The results of these previous studies agree well with the results of the present study, as Figure 2 (left)
shows. Methane and nitrogen contents are 66–68% and 27–29%, respectively. This is caused by the increased solubility of methane at low temperatures and the stripping of dissolved nitrogen. The evaluation of the nitrogen (N₂) balance for operating Mode III (data not shown) showed that 12.6 mg N₂/l influent (according to Henry’s law) could theoretically be stripped into the gas phase within the reactor. It should be mentioned that the nitrogen solubility in the effluent is increased due to cooling the effluent below 10 °C. In the biogas, about 8.2 mg N₂/l influent were collected. The difference might be caused by liquid–gas mass transfer limitation. The difference decreased with increasing HRT as indicated by the N₂ mass balances for Mode I and II (data not shown).

As Figure 2 (right) shows the yield of gaseous methane was distinctly lower than the theoretical value (neglecting COD demand of biomass growth) of 350 l/kg CODremoved. In detail, the yield of gaseous methane was 62 l/CODremoved in Mode I, 64 in Mode II and 54 in Mode III. Hence, only 15–18% of the theoretically produced methane could be recovered as gaseous methane. A fraction of 49–54% of the total produced methane left the reactor as dissolved methane with the effluent. The dissolved methane concentrations were estimated by Henry’s law. As Table 4 shows, the measured mean saturation was about 100%. Only during Mode II was a distinct supersaturation measured. Due to a leakage in the gas collection system, the fraction of methane in the reactor headspace was particularly low during this measurement. Therefore, it was concluded that a significant supersaturation had not to be considered in the present study. Furthermore, the poor methane yield can be attributed to the organic carbon utilization of SRB and the fact that a significant amount of particulate organic matter is only settled in the reactor instead of being degraded. In contrast, Sanz & Fdz-Polanco (1990) reported a methane recovery of 40–60% during the treatment of municipal wastewater with an anaerobic FB. Due to the fact that Sanz & Fdz-Polanco (1990) reported as well significant sulfate concentrations in the effluent, it might be that the SRB have not outcompeted the methanogens in their study.

**COD mass balance**

To assess the removal efficiency in more detail the in- and output COD mass flows were balanced (Table 5). About 35–40% of the input CODtot remained in the effluent. Moreover, the effluent contained as well 10–12% of the input CODtot as dissolved methane. The ratio of the total produced methane (gaseous and dissolved) is about 20–23% of the input CODtot. However, only 9–11% of the input CODtot could be recovered as gaseous methane. Furthermore, a distinct amount of 12–19% of the input CODtot was recovered as settled sludge in the reactor. The ignition loss of this sludge varied between 0.72 and 0.75 VSS/TSS (total suspended solids). In contrast, for sludge from a UASB reactor fed with settled municipal wastewater, Seghezzo (2004) reported an ignition loss of 0.55 volatile solids/total solids. Considering that the influent ignition loss in the present study was about 0.80 VSS/TSS, the results strongly indicate that it was only a mechanical

<table>
<thead>
<tr>
<th>Mode</th>
<th>Measured dissolved CH₄ (mmol/l)</th>
<th>Reactor headspace CH₄ (Vol%)</th>
<th>Estimateda dissolved CH₄ (mmol/l)</th>
<th>Saturation (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Start-up</td>
<td>0.77</td>
<td>60</td>
<td>0.91</td>
<td>85</td>
</tr>
<tr>
<td>I</td>
<td>0.95</td>
<td>66</td>
<td>0.99</td>
<td>96</td>
</tr>
<tr>
<td>II</td>
<td>0.96</td>
<td>43</td>
<td>0.65</td>
<td>148</td>
</tr>
<tr>
<td>II</td>
<td>0.97</td>
<td>60</td>
<td>0.90</td>
<td>108</td>
</tr>
<tr>
<td>III</td>
<td>0.84</td>
<td>68</td>
<td>1.02</td>
<td>82</td>
</tr>
<tr>
<td>Mean</td>
<td>0.90</td>
<td></td>
<td>0.89</td>
<td>104</td>
</tr>
</tbody>
</table>

*aCorresponding to the solubility of methane in pure water at 20 °C calculated by Henry’s law.*
separation that took place. It should be mentioned that Yoda et al. (1985) reported the effective entrapment of organic particulate matter in a FB by the formation of granular sludge agglomerations due to liquid flow motion. Furthermore, due to the reduction by SRB, 13–14% of the input COD<sub>tot</sub> was degraded and was therefore not available for methane production. The gap of 5–16% can be explained by the variation of the influent concentrations (and the volume flow, respectively), measuring inaccuracies and the neglect of possible methane supersaturation of the effluent.

**Adverse impacts of influent sulfate on the anaerobic treatment process**

The mean sulfate influent concentration was 96 or 32 mg/l in terms of sulfur. Assuming total sulfate reduction – neglecting other sulfide sources and precipitation – an effluent concentration of about 32 mg/l total dissolved sulfide (S<sup>2-</sup>) has to be expected at given operating conditions (pH = 7.0–7.3). The sulfur mass flow in the form of gaseous H<sub>2</sub>S should be less than 0.5% of the total sulfur input. This is attributed to the high solubility of H<sub>2</sub>S in water, which is about 70 times higher than the solubility of methane at 20°C. Due to its toxic effects, the concentration of H<sub>2</sub>S<sub>diss</sub> is a crucial parameter in anaerobic bioreactors (Speece 1996). At given operating conditions a concentration of about 13 mg/l of H<sub>2</sub>S<sub>diss</sub> has to be expected in the reactor. Previous investigations in biofilm reactors showed that H<sub>2</sub>S<sub>diss</sub> concentrations of 130 mg/l did not affect the methanogens in acetate-fed fixed-bed reactors (Maillacheruvu et al. 1993).

Furthermore, sulfide concentrations of 50–80 mg/l did not negatively affect methanogens in an acetate-fed anaerobic FB at similar pH values as in the present study (Yoda et al. 1987). An inhibiting effect of the H<sub>2</sub>S<sub>diss</sub> is therefore not expected in the present study. However, considering that 1 mole sulfide is equal to 64 mg COD, the effluent contained about 13% of the input COD<sub>tot</sub> as dissolved sulfide. The high sulfide concentrations cause odor emissions as well as corrosion problems (Visser 1993). Therefore, further treatment regarding sulfide removal might be required.

**CONCLUSIONS**

This study focuses on the anaerobic treatment of sulfate-containing municipal wastewater at 20°C with a FB. The total COD removal was on average 61% at OLR between 2.7 and 13.7 kg COD/(m<sup>3</sup>·d) and did not distinctly depend on the OLR. The reported removal efficiencies of 70–80% (Jewell et al. 1981; Sanz & Fdz-Polanco 1990) achieved with the anaerobic FB could not be observed in this study. In contrast to the total and particulate COD removal the dissolved COD removal seemed to decrease with increasing OLR and decreasing HRT, respectively. However, only 9–11% of the input total COD was recovered as gaseous methane. The corresponding yields of gaseous methane were between 54 and 64 l/COD<sub>removed</sub>. It should be emphasized that 12–19% of input total COD remained as settled sludge in the reactor and was not further biologically degraded, as ignition losses between 0.72 and 0.75 VSS/TSS indicated. Furthermore, assuming total sulfate reduction, 13–14% of the input total COD was degraded by SRB.

**ACKNOWLEDGEMENTS**

This research project is funded by the Willy-Hager-Foundation, Stuttgart, Germany.

**REFERENCES**


First received 9 September 2015; accepted in revised form 12 February 2016. Available online 26 February 2016