An Experimental Study on the Effects of Oxygen in Bio-gasification – Part 2

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Abstract. In order to investigate the impacts of partial aeration in anaerobic bio-gasification, a laboratory scale bioreactor (5.5 L working volume) was operated for more than 120 days duration at 35 °C under the organic loading rate of 0.33 kg COD/m².d and hydraulic retention time of 33 days. Different oxygenation conditions, 0, 2.5, 5.0 and 10.1 % (as % of feed COD) induced by daily air injection after daily feeding, were tested during the course of the experiment. Oxygenation, under these operating conditions, reduced the methane generation with only a trifling effect on biogas generation. The accumulation of volatile fatty acids is extensively reduced by oxygen introduction, significantly improving the digester stability. Reduction of soluble COD and VFAs at increasing aeration level, decrease of CH₄/CO₂ ratio with increased aeration, and immediate increase of CH₄ content upon reducing the aeration level at the end of the experimental duration, points out that the observed digester behaviour can be explained considering the substrate consumption by aerobic respiration activity of facultative biomass present in the reactor. Partially aerated anaerobic digestion can be a useful and a stable process for enhanced waste treatment and resource recovery.

Key words
Bio-gasification, Methane, Oxygen effects, Waste treatment

1. Introduction

Bio-gasification has a huge potential both as an alternative energy avenue [1-3] and a waste stabilization scheme [4-7] in the current global perspective of environmental sustainability. Positive impact on the greenhouse gas reduction [1, 3, 8] and the potential use of digestate as a fertilizer [1, 4, 8, 9] are added advantages. Improvement of process efficiency [10] and the maintenance of stable reactor operation [7] can greatly enhance the economy and usability of this technology. Studying different options for enhanced digestion and understanding the true nature of various possible disturbances, hence, play a key role in the current research works on bio-gasification.

Impacts of limited aeration in anaerobic bio-gasification processes have been discussed in few studies carried out before. Improved solubilisation of particulate organic matter in the presence of oxygen is noted in past studies [11-13]. Inhibitory nature of oxygen towards strict anaerobic acetate (acetogenic) and methane forming (methanogenic) organisms has also been discussed quite extensively [14-16]. Direct consumption of readily available organic matter (like monosaccharides, amino acids and volatile fatty acids (VFAs)) by aerobic respiration can also happen [17, 18]. In the digestion of sulfur containing organic wastes, partial aeration can aid removing sulfides from the system [19-22]. This may improve the process by reducing the toxic effects of sulfides on methanogenic organisms [22] and also minimizing the corrosive and odorous H₂S formation [2, 21].

As briefly pointed out above, the oxygen can have multifaceted effects in anaerobic bio-gasification. The overall impact of aeration on the methane yield of a digestion system, thus depends on the resultant of these diverse biochemical and physiochemical reactions involving oxygen. The rates of these different pathways would eventually depend on the operating conditions of the specific digestion system, such as the nature and concentrations of substrates (soluble, particulate or other specific ingredients like S), hydraulic behavior of the reaction system (hydraulic retention time (HRT) and mixing intensity), reactor feed conditions (batch, fed batch or continuously fed) and characteristics of the inoculum used (oxygen adaptation and culture tolerance).

2. Objectives

The positive overall impact of limited aeration in anaerobic digestion was demonstrated in a previous experimental study, described in the part 1 of this paper [13] using batch and semi-continuously fed bioreactors. It has been shown that aeration is less advantageous in the semi-continuous mode compared to the batch feeding mode. The maximum aeration level showing a definite positive effect on methane generation was found to be 1.5 % (as % of feed chemical oxygen demand (COD)), under the complete mixed (CSTR) semi-continuous feed mode.

The basic aim of the study described here was to further investigate the observed negative impacts in semi-continuous mode at increased aeration conditions. To
accomplish this objective, a larger volume bioreactor (total working volume of 5.5 L) facilitating extensive sampling, was operated for a longer duration (120+ days), compared to the experimental set-up used in the first study [13] where the limited reactor volume and operational time hindered more comprehensive analysis of the reactor dynamics.

3. Methodology

A. Experimental set-up and operation

A semi continuously (daily) fed, completely mixed glass bioreactor having a liquid volume of 3.3 L and a headspace volume of 2.2 L was operated for more than 120 days under strict anaerobic and various oxygen loaded conditions to evaluate the effects of oxygenation on the reactor performance. The inoculum used for this reactor was taken from an active mesophilic (35 °C) biogasification plant fed with chemically precipitated primary sludge from municipal wastewater (Porsgrunn municipality, Norway). Inoculum was initially filtered through a 0.5 mm wire mesh to remove any bulky particulates and then purged with N₂ gas until the dissolved oxygen reading of 0.3 mg/L is obtained. A synthetic sterile substrate solution (Table 1), having a COD of 10.9 g/L, was fed once a day with an organic loading rate (OLR) of 0.33 kg COD/m³.d and with the HRT of 33 days. The OLR was fixed at the above value after trialling with several higher values at the beginning (< 20 days – Fig. 1). The used OLR and HRT were suitable for maintaining a stable reactor operation which was an essential factor for studying the reactor dynamics disturbed by varying oxygenation conditions. Note that typical full scale CSTR biogas plants, the most common type [10, 23, 24] adopted in many applications, are operated at 12 – 30 days HRT [8, 10, 24] often with sludge recycle. No sludge recycle is used in the operational time hindered more comprehensive analysis of the reactor dynamics presented as the part 1 of this paper.

Table 1: Substrate composition

<table>
<thead>
<tr>
<th>Compound</th>
<th>Composition (g/L)</th>
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</thead>
<tbody>
<tr>
<td>Starch</td>
<td>3.90</td>
</tr>
<tr>
<td>Peptone</td>
<td>3.01</td>
</tr>
<tr>
<td>Yeast extract</td>
<td>3.58</td>
</tr>
<tr>
<td>KH₂PO₄</td>
<td>0.29</td>
</tr>
<tr>
<td>K₂HPO₄</td>
<td>0.29</td>
</tr>
</tbody>
</table>

A temperature controlled water bath fitted with an immersion heating circulator (Julabo® ED) was used for incubation at 35 (±0.1 °C). CSTR condition was established by using a magnetic stirrer (Ika® Ret) placed under the reactor and the water bath (rotation speed, 375 rpm). The reactor was operated under increasing oxygen loads of 0, 2.52, 5.04 and 10.07 % (as % of daily feed COD) as shown in the Figure 1. During the last stage of operation reported here, the oxygen load was reversed as shown. Oxygenation was carried out by daily air injection after daily feeding. The generated biogas was collected in an aluminium layered inflatable polymeric gas sampling bag (CaliBond®) connected to the reactor headspace.

B. Reactor content analyses

Typical anaerobic digestion parameters were measured in order to evaluate the reactor performance. Volumetric biogas generation, biogas composition, total and soluble CODs, pH and the volatile fatty acids of acetic, propionic, iso-butyrlic, butyric, iso-valeric, valeric, iso-caproic, caproic and heptanoic were analysed on the samples collected on daily basis from the reactor. Total/volatile suspended solids (TSS/VSS), total/volatile solids (TS/VS) and alkalinity were also determined at regular intervals. The used analytical and sample preparation procedures are described elsewhere [13].

4. Results

A. Biogas and methane generation

Despite a minor reduction observed at the aeration level of 10.1 %, generally the biogas generation rate under different aeration stages (0, 2.5, 5.0 and 10.1 %) was approximately constant at 541 (±67) ml/d (Fig. 2a). This unvarying biogas generation with oxygen load is consistent with the observations made before [13]. Under the increasing oxygen loads, however, methane generation was reduced (Fig. 2b and Fig. 3a). Averaged methane generation rates of 377 (±69), 294 (±24), 253 (±13) and 174 (±12) ml/d were observed at the corresponding oxygenation loads of 0, 2.5, 5.0 and 10.1 %. This reduction in methane generation is linear with the gradient of -19 ml/d per % O₂. The coefficient of determination is 0.964. The ratio of CH₄/CO₂ was decreasing through the increasing aeration (Fig. 2c and Fig. 3a) indicating a rise in CO₂ generation. Increased CO₂ generation implies increased aerobic respiration.

When the oxygen load was reduced from 10.1 % to 5.0 % at the last stage of reactor operation reported here (from day 109 onwards), an immediate increase in CH₄ generation was observed (Fig. 2b). CH₄/CO₂ ratio was also increased immediately (Fig. 2c). This fast response may indicate that the reduced methane generation at increased aeration should, mainly, have been due to the direct substrate consumption by aerobic respiration. If the methane reduction was mainly due to the oxygen induced inhibition of the anaerobic culture, the noted positive response should have been delayed longer than observed.
B. Volatile fatty acids (VFAs)

Oxygen introduction has a dramatic impact on the total VFAs (TVFA) content of the reactor (Fig. 3b and Fig. 4). The averaged TVFA contents of the reactor were 608 (±280), 118 (±56), 84 (±47) and 54 (±19) mg/L at corresponding oxygen loads of 0, 2.5, 5.0 and 10.1 % (these values represent the reactor condition just before daily feeding). An important observation is that, the variability of measured data (including VFAs and \( \text{CH}_4 \) generation) is significantly reduced at increasing oxygenation. This suggests the formation of a more stable digestion system in the presence of aeration. Probably the reduction and stabilization of VFAs is the key aspect in this regard. All measured types of VFAs are reduced in considerable amounts, due to aeration (Fig. 4).

The averaged contents of different species of VFAs at different oxygenated phases of reactor operation are shown in Table 2. When the aeration condition changes, from 0 to 2.5 %, from 2.5 to 5.0 % and from 5.0 % to 10.1 %, the percentage reductions in averaged acetic content are 81 %, 40 % and 31 %, respectively. The corresponding reductions for propionic are 83 %, 22 % and 11 %. The reduced content of propionic is especially helpful for stable digester operation [25] as propionic degrading organisms are the slowest growing and the most sensitive acetogenic organisms present in most anaerobic digester ecosystems [7].

An abrupt reduction of the soluble chemical oxygen demand (sCOD) in the reactor was noted when the aeration was introduced (Fig. 3b and Fig. 5). The variation of sCOD corresponds to the observed changes in the concentrations of VFAs (Fig. 3b). A synchronistic behaviour of TVFA and sCOD was previously observed by Mshandete et al. [18] while studying the ability of aerobic pre-treatment to enhance the mesophilic digestion of sisal pulp waste. The total COD curve shows a gradual decrease at the beginning (zero oxygen stage) and then stabilizes through the aeration stages (Fig. 5). No clear effect from the different aeration levels was noted. Increased hydrolysis and increased biomass generation due to aeration can have counterbalancing effects on tCOD. Oxidation of sCOD including VFAs by aerobic
respiration of facultative organisms can be the likely reason for the observed oxygen effects on sCOD.

![Image of graphs showing fluctuations in COD and sCOD with time](https://doi.org/10.24084/repqj08.732)

**Fig. 5:** Total and soluble COD variation during the reactor operation.

The reactor pH remained constant near neutral all the time (data not shown), with no observed impact by the aeration levels. Irrespective of the variation of VFAs and the CO₂ generation, the buffer capacity (2760 ± 76 mg CaCO₃/L) of the reactor maintained the fixed pH observed.

**C. Transient behaviour**

In order to study the transient reactor behaviour at the introduction of each new aeration level, a close monitoring of the reactor has been done for 24 hours before and after the aeration/feeding point (on days 42-44, 62-64, 79-81 and 108-110). Graphs for a few selected parameters during these transient periods are displayed in Fig. 6.

![Image of transient behaviour graphs](https://doi.org/10.24084/repqj08.732)

**Fig. 6:** Transient behaviour of biogas (a), CH₄ (b) and sCOD (c) during 24 hrs., before and after introducing each new aeration level.

After observing the maximum rates just after feeding, methane and biogas generation are slowing down through the individual feeding cycles until the next feeding point is reached. Meanwhile, each step of increased aeration level resulted in a decreased cumulative methane generation in the consecutive cycles (Fig. 6b). When the aeration level decreased back to 5.0 from 10.1 % at the last stage, the cumulative methane generation increased immediately in the next cycle. The total volumetric biogas generation is not affected by aeration (Fig. 6a), as described before.

Regular decreases of sCOD and VFAs are observed during each feeding cycle, after peak values near to the feeding point. This behaviour is expected in a stable semi-continuously fed reactor. Each step increase in aeration further caused reduced amounts of sCOD (Fig. 6c) and VFAs, while the OLR of the reactor was constant throughout the operation. However, the reduction of sCOD and VFA at increasing aeration levels were not as prominent as the dramatic reductions observed when the condition changed from complete anaerobic to aerated operation (Fig 3b). When analysing the VFA transient profiles, it can be recognized that the oxidation of both initial hydrolytic products (monosaccharides, amino acids etc.) and subsequent VFAs are plausible. The peak TVFA contents observed after feeding were 231(±62), 205(±37) and 151(±46) mg/L for the aeration stages of 2.5, 5.0 and 10.1 %, respectively. This reducing trend suggests that the formation of VFAs is suppressed by oxygen and the logical reason is the aerobic respiration on initial hydrolytic products. However, these VFA values are much higher than the just before feeding TVFA values reported in section 4B. This indicates that aerobic respiration on generated VFAs has also happened.

**5. Discussion**

**A. Rationale**

Simultaneous maintenance of both aerobic and anaerobic activities in a single bioreactor is demonstrated in this study. Methanogenic activity was sustained with no notable prolonged inhibition under the presence of oxygen in a suspended biomass system.

Decreased amounts of methane generation coupled with oxidative loss of soluble organic matter was previously observed by O’Keefe and Chynoweth [17] while experimenting on aeration effects in simulated landfill cells. Mshandete et al. [18] reported that 9 hours of aerobic pre-treatment enhanced the methane potential from mesophilic anaerobic digestion of sial pulp waste, but noted that increasing further aeration only resulted in a significant loss of methane potential due to aerobic substrate consumption.

Any increased level of hydrolytic products by aeration, as suggested before [11-13], could have been undermined by faster aerobic respiration in the presence of increased levels of oxygen. Drastic reductions of VFAs and soluble COD was noted by Charles et al. [26] too, while studying effects of pre-aeration on thermophilic anaerobic
digestion of organic fraction of municipal solid wastes. They [26], furthermore, found an increased enzymatic activity related to the cellulase and protease, (two hydrolytic enzymes), during the pre-aeration period.

B. Waste treatment

Considerably reduced soluble organic matter, observed due to the introduction of aeration, represent an enhanced effluent quality and an efficient treatment scheme. Due to the introduction of 2.5 % aeration, the effluent sCOD was reduced to 760 mg/L from 1473 mg/L without aeration, representing a 48 % improvement in effluent quality. This is at a cost of 22 % reduction in methane content. At the aeration condition of 10.1 %, the corresponding effluent sCOD was 516 mg/L representing a 65 % improvement in effluent quality, at a 54 % reduction in methane generation. A cost benefit analysis may be used to choose between enhanced energy production and improved treatment. Under normal operation of anaerobic digesters, a considerable fraction of refractory COD always exists the process resisting anaerobic biodegradation [27]. To reduce this content and enhance the treatment, various means of “exotic” pre-treatment prior to anaerobic digestion have been suggested, such as thermal, ultrasonic, chemical (ozone, H2O2 etc.) and enzymatic hydrolysis [28-32]. The investigated aerated anaerobic digestion process can be a simple alternative solution which could especially be useful in dealing with recalcitrant, persistent and eco-toxic organic wastes. Sequential aerobic-anaerobic or anaerobic – aerobic treatment has been suggested previously for such waste streams [33, 34]. Micro-aeration assisted anaerobic treatment has been introduced for the treatment of monoethanolamine wastes generated in post combustion CO2 capture [35]. Further studies must be performed for assessing the real potential of this aeration assisted anaerobic digestion approach.

C. Self heating capacity

Although not scrutinized in this study, increased self heat generation due to aerobic respiration associated with an anaerobic process [26] can be helpful in elevating the digestion temperature to thermophilic conditions; hence increasing the process efficiency and methane yield [25, 28]. Thermophilic digestion (~ 55 °C) further contributes towards reducing the effluent pathogenic organisms count by thermal inactivation [25, 36]. According to one study [36], at 55 °C, a holding time of 20 – 60 min. can achieve complete pathogen reduction in anaerobic digester effluents. The same study [36] revealed that Escherichia coli and Salmonella spp. were not damaged by mesophilic digestion while thermophilic digestion resulted in a rapid pathogen inactivation. It is probable that aerobic and anaerobic combined ecologic environment might be further helpful in breaking pathogenic propagation cycles. Sometimes, sequential anaerobic – aerobic treatment is adopted for enhanced pathogen removal [37]. WWTP operators in certain countries are legally bound to heat up anaerobic effluents for pathogen reduction [37]. In Norway, the accepted practice is to include an upstream hygienisation stage (>70 °C, > 2 hrs.) for digesters treating municipal organic wastes. In that kind of situations, a part of the energy lost due to aeration and methane reduction can in fact be recovered as heat generation by aerobic respiration, requiring reduced external heat load.

D. Sludge production

Increased sludge production by aerobic activity [38, 39] can be a disadvantage, but the low aerations levels applied give low production. Also, the use of this biological sludge as a fertilizer/soil amendment [1, 4, 8, 9] can offset this drawback. Anaerobic digestion effectively increases the plant available nutrient content of the organic wastes [1]. Aeration can in fact aid in producing a more stable and aesthetically sound (odourless - less H2S and VFAs) and safer (reduced pathogens) sludge compared to a complete anaerobic process. Oxygen can further be useful in fixing non bio-degradable contaminants like heavy metals present in the digestates [9]. Post aeration of anaerobically digested sludge for improved C and N removal has been suggested in other studies [40].

6. Conclusions

Under the operating conditions tested here aeration levels of 2.5, 5.0 and 10.1 % resulted in reduced methane generation proportional to the amounts of oxygen supplied. The total volumetric biogas generation was not significantly influenced by the aeration as reduced methane yield was replaced by increased CO2 yield.

Aeration has a significant impact on reducing the VFAs accumulation in the reactor, leading to a more stable digester operation.

The observed behaviour of the digester can be explained by aerobic respiration of soluble intermediate products. Oxygen inhibition of strictly anaerobic organisms appears to play a minor role.

Partial aeration assisted anaerobic digestion can be applied as a useful mean for enhanced waste treatment and safer biological sludge utilisation.

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References


