Bioresource Technology 102 (2011) 3768–3775

Contents lists available at ScienceDirect

Bioresource Technology

journal homepage: www.elsevier.com/locate/biortech

Effect of oxygen dosing point and mixing on the microaerobic removal of hydrogen sulphide in sludge digesters

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ARTICLE INFO

Article history:
Received 14 October 2010
Received in revised form 30 November 2010
Accepted 1 December 2010
Available online 7 December 2010

Keywords:
Biogas
Sulphide
Microaerobic
Sulphide-oxidising bacteria
Sludge digestion

ABSTRACT

Limited oxygen supply to anaerobic sludge digesters to remove hydrogen sulphide from biogas was studied. Micro-oxygenation showed competitive performance to reduce considerably the additional equipment necessary to perform biogas desulphurization. Two pilot-plant digesters with an HRT of ~20 d were micro-oxygenated at a rate of 0.25 NL per L of feed sludge with a removal efficiency higher than 98%. The way of mixing (sludge or biogas recirculation) and the point of oxygen supply (headspace or liquid phase) played an important role on hydrogen sulphide oxidation. While micro-oxygenation with sludge recirculation removed only hydrogen sulphide from the biogas, dissolved sulphide was removed if micro-oxygenation was performed with biogas recirculation. Dosage in the headspace resulted in a more stable operation. The result of the hydrogen sulphide oxidation was mostly elemental sulphur, partially accumulated in the headspace of the digester, where different sulphide-oxidising bacteria were found.

1. Introduction

Biogas is a versatile renewable energy source that can be employed for power and heat generation, as fuel for vehicles or for the synthesis of chemicals and materials (Weiland, 2010). Furthermore, biogas utilization has lower emissions of greenhouse gases in most of the cases compared to natural gas utilization. In addition, the biogas produced in the anaerobic digestion (AD) of sludge is less harmful to the environment than the biogas obtained from agricultural renewable resources (Pertl et al., 2010). As a result, AD of sludge plays an important role for the 10 million tons of sewage sludge being produced yearly in the EU (Appels et al., 2008).

Nevertheless, biogas contains several pollutants formed during the AD of organic matter, being hydrogen sulphide the main one. Hydrogen sulphide is a toxic compound that forms sulphur oxides during combustion, is highly soluble and causes corrosion reducing the lifetime of the equipments and creates problems of odours around the installations where it is produced.

The production range of hydrogen sulphide varies considerably from one process to another and depends on the amount of bioavailable sulphur compounds in the sewage sludge and the outcome of S-reducing microorganisms and methanogens (Lens et al., 1998), both competing for the same substrates. Appropriate conditioning of sewage sludge can reduce hydrogen sulphide generation (Dewil et al., 2008) but, inevitably, it has to be treated to meet the requirements of the technology employed for methane combustion (Dewil et al., 2008). The maximum permissible concentration when biogas is employed for combined heat and power production (CHP) is between 100 and 500 mg/Nm^3, depending on the manufacturer, and when utilized as fuel for vehicles the content must be lower than 5 mg/Nm^3 (Deublein and Steinhauser, 2007).

When treating biogas to remove hydrogen sulphide, physico-chemical technologies are widely employed at full scale as they are very developed and can reach very low hydrogen sulphide concentrations. However, physico-chemical processes require the use of chemicals and the disposal of the products of the reactions; as a result, they are expensive, especially for medium–low productions (Lens and Hulshoff Pol, 2000).

Nowadays, biological treatment processes to eliminate hydrogen sulphide have shown lower operational costs compared to traditional physico-chemical processes and lower or none utilization of chemicals (Syed et al., 2006).

The biological removal is based on the utilization of sulphur-oxidising microorganisms able to oxidize hydrogen sulphide to

Abbreviations: AD, anaerobic digestion; BR, biogas recirculation; CHP, combined heat and power production; DGGE, denaturing gradient gel electrophoresis; HRT, hydraulic retention time; HS, headspace; PCR, polymerase chain reaction; R1, reactor 1; R2, reactor 2; SOB, sulphide-oxidising bacteria; SR, sludge recirculation; WWTP, waste water treatment plant.

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doi:10.1016/j.biortech.2010.12.016

obtain energy when oxygen as electron acceptor is present. The biological oxidation is proposed to take place in stages through several intermediates as shown in Eq. (1) (Kelly et al., 1997):

$$SH^- \rightarrow S^0 \rightarrow S_2O_3^{2-} \rightarrow S_4O_6^{2-} \rightarrow SO_4^{2-} \quad (1)$$

The microorganisms responsible for the sulphide oxidation depend on the conditions and on the oxygen content available to carry out the oxidation. Some of them are capable of performing the oxidation in anaerobic conditions similar to those in the AD of sludge, such as *Thiomicrospira* sp. and *Thiobacillus* sp. (Tang et al., 2009). The reactions that take place are shown in Eqs. (2), (3), (4) (Madigan et al., 2009):

$$H_2S + 1/2O_2 \rightarrow S^\circ + H_2O \quad \Delta G^\circ = -209.4\ \text{kJ/reaction} \quad (2)$$

$$S^\circ + 2H_2O + 3/2O_2 \rightarrow SO_4^{2-} + 2H^+ \quad \Delta G^\circ = -587.1\ \text{kJ/reaction} \quad (3)$$

$$H_2S + 2O_2 \rightarrow SO_4^{2-} + 2H^+ \quad \Delta G^\circ = -798.2\ \text{kJ/reaction} \quad (4)$$

The predominance of elemental sulphur or sulphate as the final product of the oxidation depends on the oxygen accessibility; thus, in limited oxygen conditions (microaerobic), elemental sulphur is the main product (Janssen et al., 1995).

The biological technologies to remove hydrogen sulphide are mainly bioscrubbers (Janssen et al., 2001) and biotrickling filters (Goncalves and Govind, 2009; Ramirez et al., 2009) that employ pure cultures developed in the presence of hydrogen sulphide, oxygen and nutrients.

Micro-oxygenation of the biodigester is an alternative to the employment of additional units to biologically oxidize hydrogen sulphide due to the fact that some of the bacteria responsible of sulphide oxidation are already present in the sludge (Abatzoglou and Boivin, 2009). Several anaerobic digesters of agricultural wastes employ this process by introducing ~4–6% of air, related to the total biogas production, in the headspace (HS) of the biodigester where thiobacteria develop reaching outlet concentrations in the biogas below 200 ppmv (Weiland, 2010).

In this way, hydrogen sulphide content in the biogas was reduced by supplying oxygen to the anaerobic treatment of wastewaters with a high concentration of sulphate, both in fed-batch reactors (van der Zee et al., 2007), showing an effective competence of sulphide-oxidising microorganisms versus other microorganisms for the available oxygen, and in continuous reactors (Fox and Venkatasubbiah, 1996; Khanal and Huang, 2003; Zitomer and Shrout, 2000). In the treatment of sewage sludge, pilot-plant research (200L) showed similar removal efficiencies under microaerobic conditions when employing equivalent amounts of air or pure oxygen supplied to the HS (Díaz et al., 2010) and the limited air supply to the sludge recirculation (SR) of full-scale digesters lead to a sustained removal of hydrogen sulphide in the biogas and enhanced hydrolysis of the organic matter in the process (Jenicek et al., 2008).

Finally, several of the microorganisms related to the AD of organic matter have shown resistance to the oxygen presence further than it was often accepted, therefore, the application of microaerobic conditions did not show inhibition of methanogenesis (Estrada-Vazquez et al., 2003; Zitomer and Shrout, 1998). On the other hand, limited oxygen supply caused higher hydrolysis rates for the case of complex organic matter in the studies of Jenicek et al. (2008) and Johansen and Bakke (2006).

Literature about the limited oxygen (or air) supply to remove hydrogen sulphide has shown the feasibility of the treatment while preserving AD (Fdz.-Polanco et al., 2009; Díaz et al., 2010); conversely, there is a lack of information about the sulphide-oxidising bacteria (SOB) community and about the effect that the dosing point and mixing conditions of the reactor have on the process. A further insight on these characteristics can boost the process optimization and application.

The aim of this study is to evaluate the effect of the oxygen supply to the liquid/gas phase with biogas/sludge recirculation on the microaerobic removal of hydrogen sulphide from the biogas, on the AD performance and to describe the SOB established after limited oxygen supply.

2. Methods

2.1. Pilot-plant digesters

The treatment of sludge and the removal of hydrogen sulphide were performed in two pilot-plant scale reactors with a working volume of 200 L (250 L total volume) ran at the same time with the same feed sludge. The reactors were insulated, and the walls were heated with electric resistance. Reactor 1 (R1) was mixed with SR provided by a Bredel peristaltic pump (~50 L/h) and reactor 2 (R2) was mixed with biogas recirculation (BR) provided by an electroAD compressor (~4.5 L/min). The feed was provided from a continuously stirred tank with a Watson-Marlow peristaltic pump. Micro-oxygenation was maintained using the regulated flow of pure oxygen with a Cole-Parmer EW-32660-26 mass flow controller from an oxygen cylinder. In R1, oxygen was supplied to the SR or to the HS while in R2, oxygen was introduced with the feed sludge or into the HS. Pilot-plant diagram of R1 and R2 is shown in Fig. 1. The HS in the bioreactor (50 L) allowed the storage of ~1/4 of the daily biogas production. Finally, tygon tubing was used as the conduction material for the biogas.

2.2. Operational conditions

The pilot-plant study was developed in the sludge digesters employed for previous anaerobic/microaerobic experiments during 500 d. Digestion was performed in the mesophilic range (35 ± 1 °C) with an HRT of ~20 days. The feed consisted of mixed sludge from the Villalonquejar WWTP (Burgos, Spain) with a variable organic load (COD_T max–min [94–48] g/L). Sludge was sampled every week and conserved at 4 °C. To increase the amount of hydrogen sulphide produced during digestion, sodium sulphate was added to the feed in concentration of ~2.2 g/L.

Micro-oxygenation was performed at a flow rate of 1.8 ± 0.1 NmL/min; this represents ~0.25 NL of oxygen per L of feed sludge. Oxygen was supplied into R1 on day 41 into the SR and was changed to HS on day 81. R2 was micro-oxygenated from day 41 to 80 in the feed and from day 81 to 120 in the HS. Table 1 summarises the periods of the study.

2.3. Monitoring and experimental analysis

The pilot-plant conditions were monitored online using pressure, temperature (PT100), pH and ORP probes (Cole-Parmer EW-05993-10 and EW-27301-19, respectively). The biogas production was measured by liquid displacement in an inverted cylinder equipped with an electro-valve to release the biogas when measure volume (550 ± 10 mL) was reached.

Biogas composition was measured online by gas chromatography (GC). Micro-GC was equipped with a thermal conductivity detector (TCD) and two VARIAN modules; CP-Molsieve 5A PLOT (10 m × 32 mm, df 30 μm) for oxygen and nitrogen analysis and CP-PoraPLOT Q (10 m × 32 mm, df 10 μm) for methane, carbon dioxide and hydrogen sulphide composition. Helium was the carrier gas.

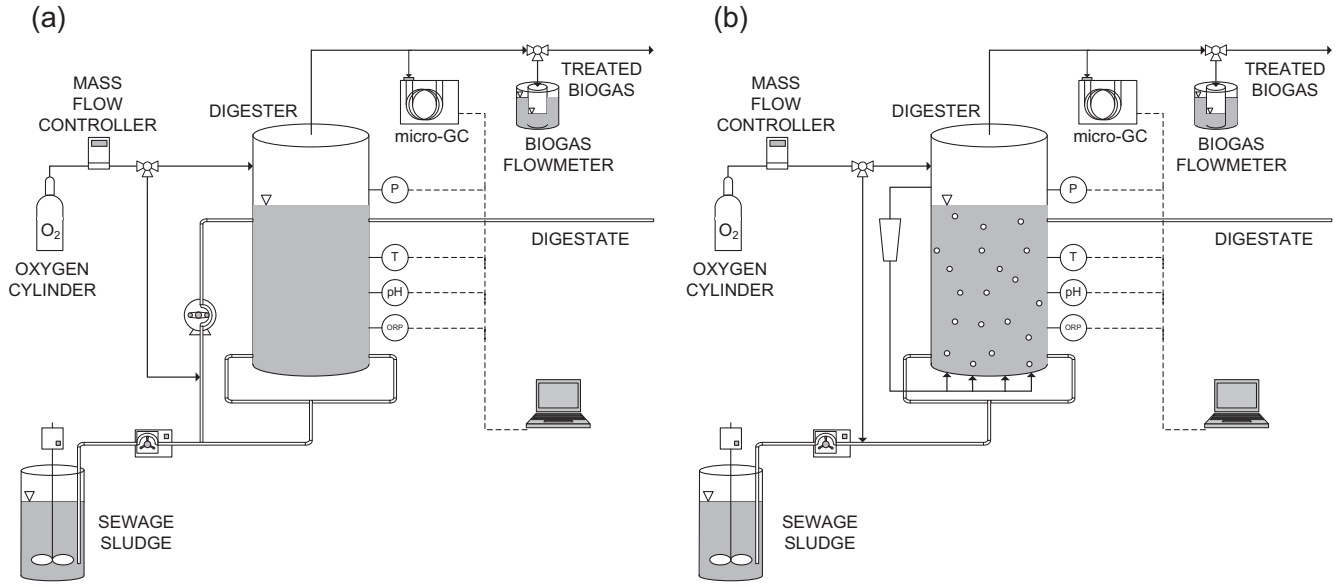

Fig. 1. Pilot-plant diagram. (1a) Reactor 1 (sludge recirculation) and (1b) Reactor 2 (biogas recirculation).

Table 1
Operational conditions during the study. AN: Anaerobic; MA: Microaerobic.

Period	R1			R2		
	1	2	3	1	2	3
Operation time (d)	40	40	40	40	40	40
Anaerobic/microaerobic	AN	MA	MA	AN	MA	MA
Mixing	SR	SR	SR	BR	BR	BR
Micro-oxygenation						
Flow rate (NmL/min)	0	1.8	1.8	0	1.8	1.8
Dosing point	–	SR	HS	–	Feed	HS

Sulphate concentration in the digesters and in the feed sludge was determined by high performance liquid chromatography (HPLC). Samples were centrifuged at 5000 rpm for 10 min, and the supernatant was filtered at 0.45 μm. Subsequently, a 1.5-mL sample was centrifuged again at 10000 rpm for 10 min, and the supernatant was filtered at 0.22 μm. Finally, the filtered sample was injected in the HPLC. The HPLC equipment has a conductivity detector (Waters Millipore TCM) and an anionic column (Waters Millipore IC-pack A-HR 4.6 x 150 mm). The method worked at 20 °C and used 5.51 g/L potassium hydrogen phthalate as the liquid carrier.

Thiosulphate was measured by HPLC according to the method described by van der Zee et al. (2007).

Sulphur content in the sludge was estimated from an elemental analysis of sulphur. Samples were dried at 95 °C, cooled and analysed in a LECO SC32 oven. Analysis was carried out by combustion of the samples at 1350 °C; as a result, the sulphur was completely oxidised to sulphur dioxide and evaluated in the detection cell (TCD). The sulphur estimation was the result of subtracting the content of the other analysed sulphur species (sulphate, thiosulphate) of the sample from the total sulphur content.Total dissolved sulphide was determined according to standard methods by the ion-selective electrode method (Clesceri et al., 1998).

Total and soluble chemical oxygen demand (COD_T and COD_S) were evaluated weekly according to standard methods.

A polymerase chain reaction (PCR) and subsequent denaturing gradient gel electrophoresis (DGGE) analyses were carried out to identify SOB. The analysis was performed as described by Lebrero et al. (2010). The samples were taken from the global liquid phase and from the HS of the digesters R1 and R2 (top, wall and liquid surface). For DGGE band sequencing, each selected band was excised, using a sterile blade, from the gel. The samples were reamplified in the thermocycler and sent to Secugen (Secugen SL, Spain) to be sequenced. The checked sequences were compared to NCBI (National Center for Biotechnology Information) (http://www.ncbi.nlm.nih.gov/) using BLAST (Basic Local Alignment Search Tool) to retrieve similar sequences and phylogenetically related species. The sequences generated in this study had been deposited in the GenBank database under accession numbers HQ392823 to HQ392831.

3. Results and discussion

3.1. Hydrogen sulphide removal in the biogas

R1, mixed with SR produced a biogas with an average hydrogen sulphide concentration in the biogas of 14,437 ± 1010 ppmv during the period 1; from day 0 to day 40.

Oxygen was supplied with a rate of 1.8 ± 0.1 mL/min into the SR of R1 during period 2 (from day 41 to 80) and into the HS during period 3 (from day 81 to 120). As a result of the oxygen introduction, the hydrogen sulphide concentration in the biogas of R1 drastically fell as shown in Fig. 2a. The average removal efficiency in period 2 was 98.8%. During this period, the hydrogen sulphide concentration was below 300 ppmv in 26 (below 100 ppmv in 21) out of 33 samples.

For period 3, the average removal efficiency was 99.8%. In this period, the concentration of hydrogen sulphide was lower than 300 ppmv in 32 (lower than 100 ppmv in 31) out of 33 samples. The introduction of oxygen in the HS showed a better performance than in the SR.

The concentration of the main biogas components, methane and carbon dioxide, showed small variances during the study (Fig. 2c); during period 1, the biogas produced in R1 had an average methane concentration of 62.6 ± 1.2 %(v/v). In period 2, this concentration was 61.3 ± 1.3 %(v/v) and; finally, in period 3 methane accounted for 62.5 ± 1.3 %(v/v) of the biogas. According to statistical *t*-Student test and *F*-test, there was not a significant difference

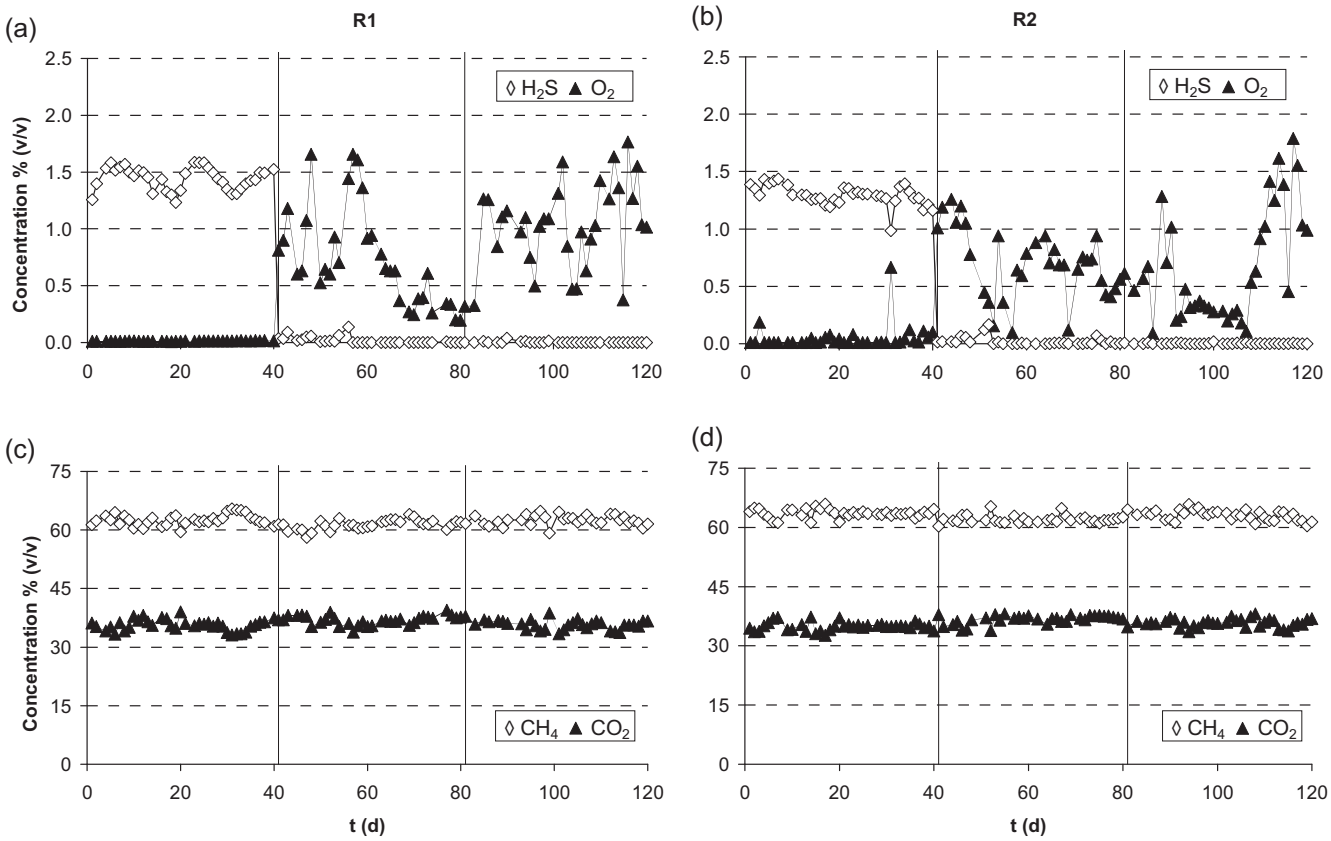

Fig. 2. Biogas composition in %(v/v). (2a) Hydrogen sulphide and oxygen concentrations in R1; (2b) hydrogen sulphide and oxygen concentrations in R2; (2c) methane and carbon dioxide concentrations in R1; and (2d) methane and carbon dioxide concentrations in R2.

between the average concentrations of methane and carbon dioxide for a 95% confidence interval in the three periods of the study. This indicates that limited oxygen supply did not reduce the energetic content of the biogas.

The reactor R2 performed sludge digestion mixed by BR. The biogas produced in R2 showed an average content of sulphide of 12,926 ± 877 ppmv during the period 1.

Micro-oxygenation started on day 41 (period 2) with a flow of 1.8 ± 0.1 NmL/min of pure oxygen introduced with the feed. Hydrogen sulphide content in biogas dropped sharply as shown in Fig. 2b. The concentration during this period was below 300 ppmv in 28 (below 100 ppmv in 20) out of 33 samples with an average removal efficiency of 98.5%.

In period 3, with oxygen supply in the HS, the hydrogen sulphide concentration was below 300 ppm in all the 36 samples measured and below 100 ppmv in 30 samples, showing an average removal efficiency of 99.8%. Again, the introduction of oxygen in the HS lead to a higher removal than the introduction with the feed.

The concentration of methane and carbon dioxide in the biogas in R2 (Fig. 2d) showed the same behaviour than in R1. There was not a significant difference between the average concentration of these components for a 95% confidence interval for *t*-Student test and *F*-test showing that micro-oxygenation did not significantly affect the methane content of the biogas. The average methane concentration in %(v/v) was 63.6 ± 1.1 in period 1, 62.1 ± 1.0 in period 2 and 63.1 ± 1.3 in period 3.

Then, in terms of removal, oxygen supply seemed to achieve a quite similar performance when introduced into HS, into the SR or with the feed. This fact shows that the mechanism of removal does not require of oxygen transfer to global liquid phase to develop. BR and SR did not show a significant difference in terms of gaseous sulphide removal from the biogas when oxygen was supplied to the HS. The main difference is the evolution of dissolved species as it is described later.

Analysis of sequenced bands by means of DGGE in samples drawn from R1 showed seven different SOB in the HS samples with a similarity of at least 92% of the closely related cultures, all belonging to the Proteobacteria division. Members found belonged to *Acidithiobacillus thiooxidans* (99% similarity), which has been employed for the aerobic treatment of H_2S in biotrickling filters, and *Arcobacter mytili* (92% similarity), which has been found in anaerobic enriched cultures for the anaerobic biooxidation of sulphide (Tang et al., 2009). Moreover, *Halothiobacillus neapolitanus* (92% similarity) and members of *Thiomonas* sp. (94% similarity) and *Thiobacillus* sp. (99% similarity) were found. *H. neapolitanus* and species belonging to those genera have been reported to utilise different sulphur compounds to produce sulphuric acid in sewer systems (Okabe et al., 2007). Finally, a 100% similarity was found for *Sulfuricuvum kujiense* that has been described by Kodama and Watanabe (2004) as a facultatively anaerobic sulphur oxidising bacterium.

On the other hand, in R2, the most closely related cultures to SOB showed at least an 86% similarity and were related to *Halothiobacillus kellyi* (86% similarity), *Arcobacter mytili* (92% similarity). Furthermore, members of *Alicyclobacillus* sp. (90% similarity) were found, some members of this genus, such as *Alicyclobacillus aeris*, have been described as ferrous- and sulphur-oxidising bacteria (Guo et al., 2009).

It is important to highlight that these SOB found in R1 and R2 were not present in the samples taken from the global liquid surface. As in R1 identified SOB were only present in the HS samples.

3.2. Evolution of the sulphur species and oxygen utilisation

A sulphur balance was performed in order to study the results of hydrogen sulphide oxidation and to explore the performance of biodesulphurisation in the different conditions studied. As a complement, oxygen utilisation in the biodigesters can be estimated from the sulphur balance assuming the stoichiometrical necessary amounts to form every specie.

The non-anionic sulphur content of the sewage sludge was calculated assuming a relation of 8.06 mg-S/gTS for the sewage sludge, 9.51 mg-S/gTS for digestate of R1 and 9.71 mg-S/dTS for the digestate of R2, estimated by the method described in the materials and methods.

The sulphur balance is shown in Fig. 3. It can be observed that R1 had a total input of 7890 mg-S/d during period 1 composed of sulphide, sulphate and the total sulphur non-anionic content of the sewage sludge. In this conventional AD represented by period 1, ~60% of the total sulphur inputs was reduced to hydrogen sulphide (~4700 mg-S/d) that distributed between gas and digested sludge for the conditions of the gas–liquid equilibrium in the digester.

During period 2 the inputs to R1 were 7990 mg-S/d. The introduction of oxygen in period 2 substantially altered the sulphur outputs of the process. The amount of sulphur transformed was ~58% (~4610 mg-S/d); however, there was a gap in the outputs of ~31% (~2500 mg-S/d, this gap is related to the formation of elemental sulphur (not analysed in this study) in microaerobic conditions. Furthermore, digestate contained ~8% of thiosulphate as a result of further oxidation of hydrogen sulphide.

In period 3, the inputs of sulphur to the biodigester were slightly lower (~7360 mg-S/d) as a result of a lower content of TS in the sewage sludge. The amount of sulphur transformed was ~56% (~4140 mg-S/d), with a gap of ~32% of the inputs (~2360 mg-S/d). The amount of thiosulphate formed during this period was ~8% of the inputs, similar to period 2.

Probably the lower sulphur reduction during period 3 compared to period 2, and lower gap in absolute value is the reason for the lower average concentration of hydrogen sulphide in period 3 compared to period 2. Then, combining this fact with the results of removal from the biogas, the point of supply did not show a significant effect on the process efficacy.

The formed elemental sulphur was partially accumulated in the top and the walls of the HS of R1 as observed when opening the reactor after 620 d of alternating anaerobic/microaerobic operation. Elemental sulphur was accumulated in form of "stalactites" in the top, with sludge in the surface between the sulphur and in the internal surface of the digester, and in layers on the walls of the digester with biomass between these layers. This fact combined with that oxygen is only in contact with the surface of the sludge when introduced in the HS, suggests a mechanism of sulphur oxidation in the HS. The appearance of SOB only in the HS supports this fact.

Proposed mechanism is that SOB develops in the top and the walls of the HS in the digester. Hydrogen sulphide dissolves in the water contained in the biogas and is easily accessible to SOB, that employ the reaction 2 under oxygen-limited conditions to produce elemental sulphur. Water condensation in the HS was the responsible mechanism of sulphur accumulation in the form of "stalactites".

For R2, the AD period 1 had a total sulphur input of ~7890 mg-S/d, during digestion ~58% of this sulphur was reduced by sulphur-reducing microorganisms leading to ~3150 mg-S/d of hydrogen sulphide in the biogas and ~1330 mg-S/d of total dissolved sulphide in the digestate.

In the period 2, oxygen was supplied with the feed to R2 and influenced the sulphur balance as a result of elemental sulphur formation. During period 2, the amount of sulphur transformed from the inputs was ~56% (~4440 mg-S/d) and the gap due to sulphur formation was 52% (~4190 mg-S/d), formation of thiosulphate during this period was negligible.

As it can be observed in Fig. 3, total dissolved sulphide concentration during this period was only ~160 mg-S/d compared with the ~1330 mg-S/d of the AD period 1, oxygen supply removed not only gaseous hydrogen sulphide but also dissolved sulphide as a result of a better contact between the gas and the liquid phases.

In the period 3, with oxygen supply to the HS of R2, total sulphur inputs were ~7360 mg-S/d. Sulphur transformed from the inputs was ~55% (4083 mg-S/d) with a gap of ~52% (~3860 mg-S/d).

The difference in the gap in the sulphur balance is the main consequence of the mixing method employed. While with SR, only gaseous sulphide was removed leading to a gap around 32%; BR removed dissolved sulphide too, leading to a gap around 52%. Therefore, BR will imply a higher oxygen consumption than biogas RC. Nevertheless, sulphide toxicity to bacteria may be prevented employing this method.

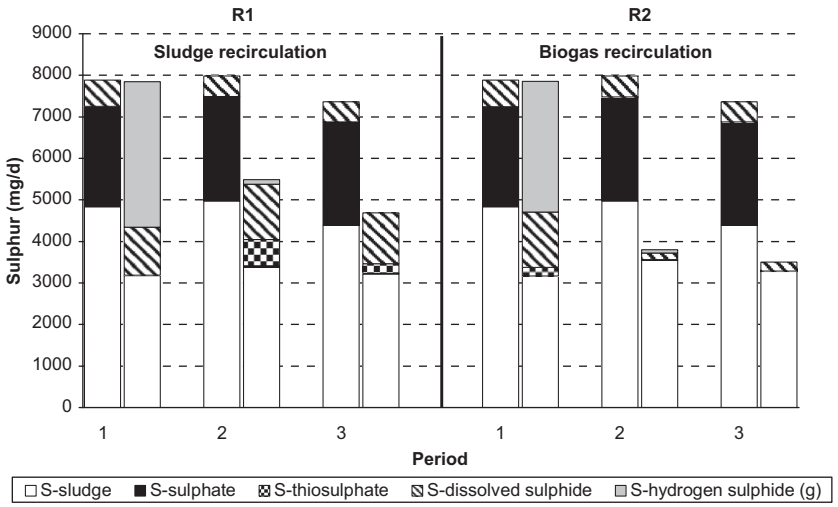

Fig. 3. Sulphur balance during the study. For each period, left column: inputs; right column: outputs.

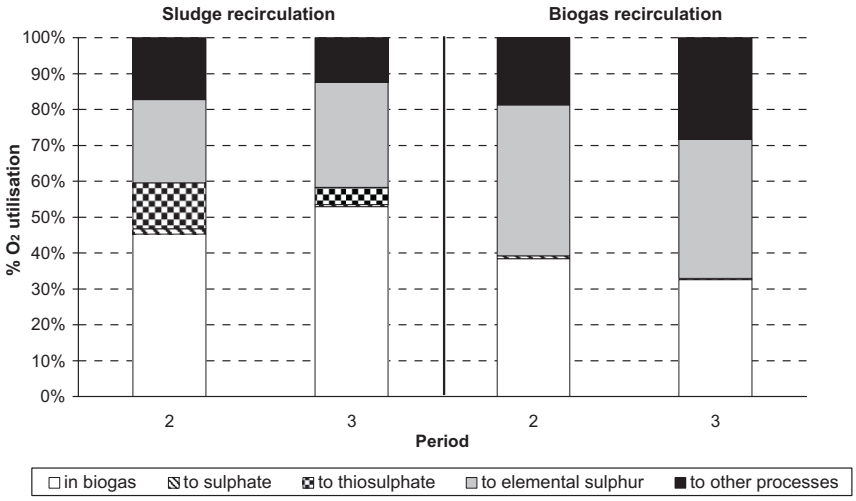

Fig. 4. Estimated oxygen utilisation in digesters.

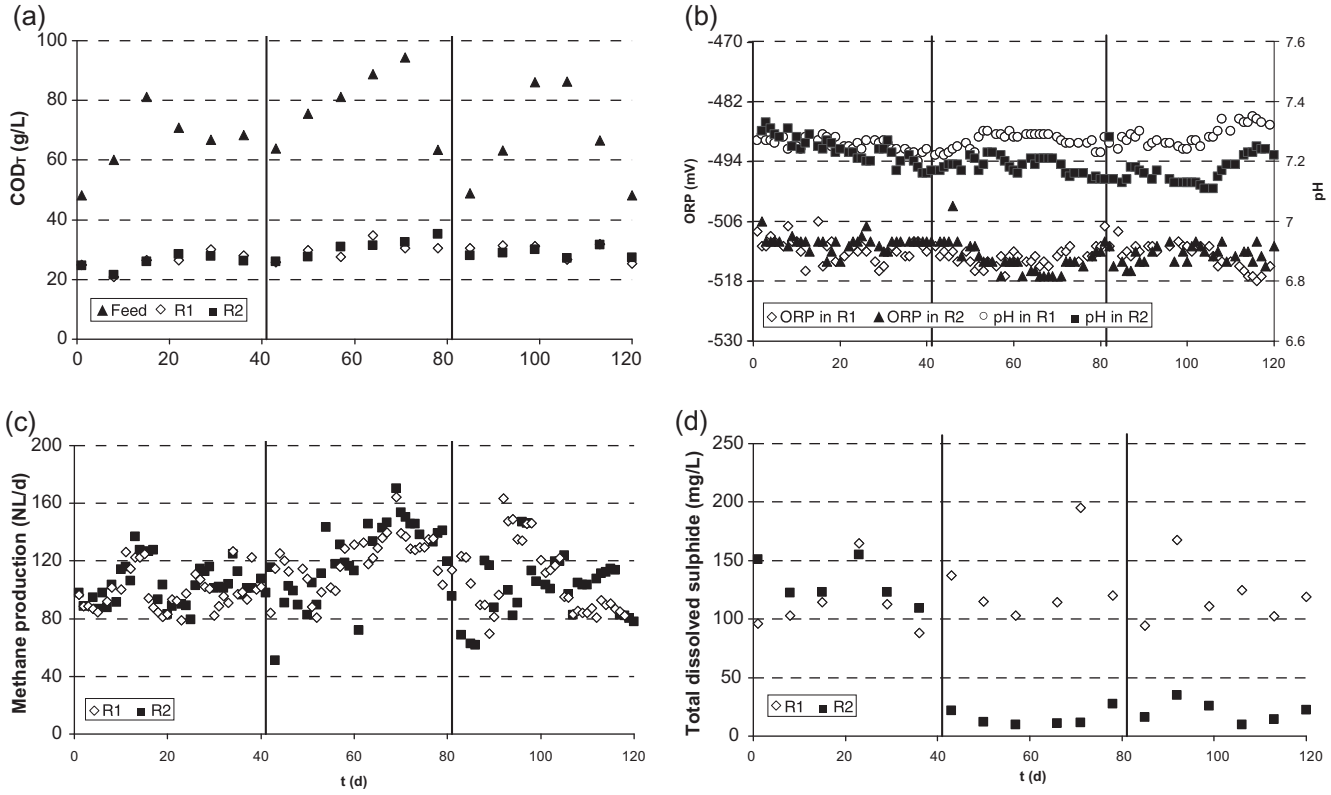

Fig. 5. Digestion behaviour during the study. (5a) COD_T in feed and digested sludge. (5b) ORP and pH in digesters. (5c) Methane production. (5d) Total dissolved sulphide concentration in digesters.

R2 showed a lower amount of accumulated elemental sulphur in the top compared to R1, probably as a result of BR that impeded sulphur deposition. The walls in the HS appeared also covered by elemental sulphur; however, it was covered by a biomass layer as a result of sludge dragging by BR.

The estimation of oxygen utilisation is shown in Fig. 4. R1 showed that ~50% of the oxygen supplied to the biodigesters was consumed while the rest left the biodigester with biogas. From oxygen that was consumed in the biodigester ~35% was employed for sulphide oxidation to form elemental sulphur and thiosulphate and the rest was employed to other non-identified processes. When oxygen was supplied to the HS (period 3) the amount employed for other processes was slightly lower than when supplied in the SR.

R2 showed a higher amount of oxygen consumed in the biogester around 60–70% from which a 30–40% was employed for sulphide oxidation. In R2 the amount of sulphur species more oxidised than elemental sulphur such as thiosulphate or sulphate was negligible. This is probably the result of a higher amount of hydrogen sulphide to be oxidised as dissolved sulphide was also removed with BR, then, a lower relation of oxygen/hydrogen sulphide impeded SOB to perform a further oxidation.

Our findings about the dosing point indicate that the supply to the HS seemed to be more profitable than to the liquid phase. This

is because of sulphur oxidation happening in the HS (with SOB developing in this area), lower contact of oxygen with global liquid phase and a lower consumption to other undesired processes and finally, avoidance of problems related with mixing because of stops in SR for maintenance.

3.3. Effect on digestion performance

Organic matter removal was not significantly affected by micro-oxygenation. Fig. 5 shows the main parameters analysed to evaluate the performance during the study.

Average COD_T removal in R1 with SR was 60% during the anaerobic period 1, 59% for period 2 and 58% for period 3. R2 with BR showed a similar performance than R1 (Fig. 5a), neither R1 nor R2 was significantly affected by limited oxygen supply. Average COD_S in digested sludge was 1.9 ± 0.5 g/L in R1 and 1.9 ± 0.3 g/L in R2 with similar performance in anaerobic and microaerobic periods, the higher variation in COD_S in R1 might be the result of the higher maintenance required by SR what resulted in worse sludge mixing. pH and ORP were not substantially altered by microaerobic conditions as shown in Fig. 5b. Digestion occurred at pH 7.1–7.3 while ORP was around −510 mV (Ag/AgCl reference electrode).

Methane production is shown in Fig. 5c. This rate varies as a result of the variable COD_T in the sewage sludge. R1 showed an average methane yield during the period 1 of 128 ± 27 mg/gCOD_T fed. During the microaerobic periods 2 and 3 this yield was 130 ± 40 and 117 ± 36 mg/gCOD_T fed, respectively. It was not found a statistical significant difference between these averages; therefore, limited oxygen supply did not affect the methane production during the digestion with SR independently of the point of supply.

A similar behaviour was found in R2. Methane yield in period 1 was 125 ± 25 mg/gCOD_T, 128 ± 34 mg/gCOD_T in period 2 and 123 ± 39 in period 3. Then, micro-oxygenation with BR did not significantly affect methane production with BR.

This observation is in agreement with the estimated oxygen consumption in the reactors (Fig. 4). Most of the oxygen consumed was employed for sulphide oxidation; thereby anaerobic degradation of organic matter and methane production were not altered by the micro-oxygenation.

Finally, the total dissolved sulphide concentration is shown in Fig. 5d. In R1 with SR, micro-oxygenation did not remove sulphide from the liquid phase as a result of the poor contact of gas and liquid and only hydrogen sulphide from the biogas was removed, dissolved sulphide was between 90 and 200 mg/L during the study for this reactor. In R2 with BR, the oxygen introduction resulted in the removal of dissolved sulphide to achieve concentration in the digestate below 40 mg/L; thereby, the BR may be employed if dissolved sulphide removal is required.

4. Conclusions

The hydrogen sulphide removal from biogas achieved in microaerobic conditions showed an efficiency higher than 98% for all the configurations studied. While sludge recirculation implied a lower consumption of oxygen than biogas recirculation, biogas recirculation could be utilised to remove dissolved sulphide from the liquid too. Besides, elemental sulphur was the main product of sulphide oxidation and was partially accumulated in the headspace; however, accumulation was lower with biogas recirculation because sulphur deposition was impeded. Sulphide oxidation occurred in the headspace were different sulphide-oxidising bacteria developed; then, the supply of oxygen to the headspace was found the optimal dosing point.

Acknowledgements

The authors thank the project CSD2007-00055, CONSOLIDER-INGENIO 2010, and the Spanish Ministry of Education and Science.

References

Abatzoglou, N., Boivin, S., 2009. A review of biogas purification processes. Biofuels Bioproducts & Biorefining-Biofpr 3, 42–71.
Appels, L., Baeyens, J., Degrève, J., Dewil, R., 2008. Principles and potential of the anaerobic digestion of waste-activated sludge. Progress in Energy and Combustion Science 34, 755–781.
Clesceri, L.S., Greenberg, A.E., Eaton, A.D., 1998. Standard Methods for the Examination of Water and Wastewater, 20th ed. American Public Health Association, Washington, DC.
Deublein, D., Steinhauser, A., 2007. Biogas from Waste and Renewable Resources: An Introduction. Wiley-VCH, Weinheim.
Dewil, R., Baeyens, J., Roels, J., Van de Steene, B., 2008. Distribution of sulphur compounds in sewage sludge treatment. Environmental Engineering Science 25, 879–886.
Díaz, I., Lopes, A.C., Pérez, S.I., Fdz-Polanco, M., 2010. Performance evaluation of oxygen, air and nitrate for the microaerobic removal of hydrogen sulphide in biogas from sludge digestion. Bioresource Technology 101, 7724–7730.
Estrada-Vazquez, C., Macarie, H., Kato, M.T., Rodriguez-Vazquez, R., Esparza-Garcia, F., Poggi-Varaldo, H.M., 2003. The effect of the supplementation with a primary carbon source on the resistance to oxygen exposure of methanogenic sludge. Water Science and Technology 48, 119–124.
Fdz.-Polanco, M., Díaz, I., Pérez, S.I., Lopes, A.C., Fdz-Polanco, F., 2009. Hydrogen sulphide removal in the anaerobic digestion of sludge by microaerobic processes: pilot plant experience. Water Science and Technology 60 (12), 3045–3050.
Fox, P., Venkatasubbiah, V., 1996. Coupled anaerobic/aerobic treatment of high-sulfate wastewater with sulfate reduction and biological sulfide oxidation. Water Science and Technology 34, 359–366.
Goncalves, J.J., Govind, R., 2009. Enhanced biofiltration using cell attachment promotors. Environmental Science & Technology 43, 1049–1054.
Guo, X., You, X.-Y., Liu, L.-J., Zhang, J.-Y., Liu, S.-J., Jiang, C.-Y., 2009. Alicyclobacillus aeris sp. nov., a novel ferrous- and sulfur-oxidizing bacterium isolated from a copper mine. International Journal of Systematic and Evolutionary Microbiology 59, 2415–2420.
Janssen, A.J.H., Ruitenberg, R., Buisman, C.J.N., 2001. Industrial applications of new sulphur biotechnology. Water Science and Technology 44, 85–90.
Janssen, A.J.H., Sleyster, R., Kaa, C.V.D., Jochemsen, A., Bontsema, J., Lettinga, G., 1995. Biological sulphide oxidation in a fed-batch reactor. Biotechnology and Bioengineering 47, 327–333.
Jenicek, P., Keclik, F., Maca, J., Bindzar, J., 2008. Use of microaerobic conditions for the improvement of anaerobic digestion of solid wastes. Water Science and Technology 58, 1491–1496.
Johansen, J.E., Bakke, R., 2006. Enhancing hydrolysis with microaeration. Water Science and Technology 53, 43–50.
Kelly, D.P., Shergill, J.K., Lu, W.P., Wood, A.P., 1997. Oxidative metabolism of inorganic sulfur compounds by bacteria. Antonie Van Leeuwenhoek International Journal of General and Molecular Microbiology 71, 95–107.
Khanal, S.K., Huang, J.-C., 2003. ORP-based oxygenation for sulfide control in anaerobic treatment of high-sulfate wastewater. Water Research 37, 2053–2062.
Kodama, Y., Watanabe, K., 2004. Sulfuricurvum kujiense gen. nov., sp. nov., a facultatively anaerobic, chemolithoautotrophic, sulfur-oxidizing bacterium isolated from an underground crude-oil storage cavity. International Journal of Systematic and Evolutionary Microbiology 54, 2297–2300.
Lebrero, R., Rodríguez, E., Martin, M., García-Encina, P.A., Muñoz, R., 2010. H2S and VOCs abatement robustness in biofilters and air diffusion bioreactors: a comparative study. Water Research 44, 3905–3914.
Lens, P.N.L., Hulshoff Pol, L.W., 2000. Environmental Technologies to Treat Sulphur Pollution: Principles and Engineering. IWA Publishing, London.
Lens, P.N.L., Visser, A., Janssen, A.J.H., Hulshoff Pol, L.W., Lettinga, G., 1998. Biotechnological Treatment of Sulfate-Rich Wastewaters. Critical Reviews in Environmental Science and Technology 28, 41–88.
Madigan, M.T., Martinko, J.M., Dunlap, P.V., Clark, D.P., 2009. Brock Biology of Microorganisms, 12th ed. Prentice Hall, Upper Saddle River, NJ.
Okabe, S., Odagiri, M., Ito, T., Satoh, H., 2007. Succession of sulfur-oxidizing bacteria in the microbial community on corroding concrete in sewer systems. Applied and Environmental Microbiology 73, 971–980.
Pertl, A., Mostbauer, P., Obersteiner, G., 2010. Climate balance of biogas upgrading systems. Waste Management 30, 92–99.
Ramirez, M., Gomez, J.M., Aroca, G., Cantero, D., 2009. Removal of hydrogen sulfide by immobilized *Thiobacillus thioparus* in a biotrickling filter packed with polyurethane foam. Bioresource Technology 100, 4989–4995.
Syed, M., Soreanu, G., Faletta, P., Béland, M., 2006. Removal of hydrogen sulphide from gas streams using biological processes – a review. Canadian Biosystems Engineering 48, 2.1–2.14.
Tang, K., Baskaran, V., Nemati, M., 2009. Bacteria of the sulphur cycle: an overview of microbiology, biokinetics and their role in petroleum and mining industries. Biochemical Engineering Journal 44, 73–94.

van der Zee, F.P., Villaverde, S., García, P.A., Fdz.-Polanco, F., 2007. Sulfide removal by moderate oxygenation of anaerobic sludge environments. Bioresource Technology 98, 518–524.
Weiland, P., 2010. Biogas production: current state and perspectives. Applied Microbiology and Biotechnology 85, 849–860.
Zitomer, D.H., Shrout, J.D., 1998. Feasibility and benefits of methanogenesis under oxygen-limited conditions. Waste Management 18, 107–116.
Zitomer, D.H., Shrout, J.D., 2000. High-sulfate, high-chemical oxygen demand wastewater treatment using aerated methanogenic fluidized beds. Water Environment Research 72, 90.