University of South Wales
Prifysgol De Cymru

THE AD CENTRE
THE WALES CENTRE OF EXCELLENCE FOR ANAEROBIC DIGESTION

Demonstrating Management Systems at Full-Scale Anaerobic Digestion Plants

Insource Energy AD Plant (Rogerstone, Wales)

inSource ENERGY

March 2014

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BIO-METHANE REGIONS

Part Reporting for Deliverable for
Task 5.3 Management Demonstrators

Co-funded by the Intelligent Energy Europe Programme of the European Union

Llywodraeth Cymru
Welsh Government

Further information related to this demonstrator can be found in:

Julie Williams, Haydn Williams, Richard Dinsdale, Alan Guwy and Sandra Esteves (2013). Monitoring methanogenic population dynamics in a full scale anaerobic digester to facilitate operational management. *Bioresource Technology* **140**: 234-242

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1. Aim of the Demonstrator

Anaerobic Digestion (AD) is increasingly being applied as a sustainable waste treatment and bioenergy generating technology throughout Europe and the World. Although AD is a proven technology for a number of substrates and designs, digesters can be susceptible to process instability for example during start-up or periods of stress due to inhibitory substances or sudden increases in organic/hydraulic loading. In addition, a number of digesters are still far from operating optimally both in terms of treatment achieved as well as biogas recovered. A typical expression of these inefficiencies is the build-up of volatile fatty acids (VFAs), which are process intermediates.

Significant understanding of the biochemical interactions occurring during AD has been developed in the last few decades. This understanding coupled with chemical analyses and sensors currently available in support of process monitoring have been summarised in the report produced as part of Task 5.2 Report within the IEE Biomethane Regions project (Esteves S., Miltner M. And Puchas K (July 2013) Monitoring Review and Guide for the Optimisation of Anaerobic Digestion and Biomethane Plants). As discussed in detail in this report the **AD process** is delivered by **complex and dynamic systems where microbiological and physico-chemical aspects are closely linked and influence process performance**. As the process is required to be delivered by an effective consortia of microbes, **process stability is dependent on the critical balance between the symbiotic growth rates and activity of the main groups of bacteria and archaea i.e. hydrolytic and acid forming bacteria, acetogens and methanogens**. The fact that digesters operate with different substrates and operating conditions and are populated by mixed bacterial and archaeal cultures with a competitive nature, makes digesters' detailed performance difficult in some cases to predict and even control.

A more in-depth understanding of the complex biochemical interactions that determine digester stability and promote enhanced performance remains a significant challenge facing the AD industry today and more monitoring efforts should be conducted both at laboratory as well as full scale to increase understanding of interactions and communities in digesters in order to improve digesters' technical, economical and environmental deliveries.

The work reported here and performed under Task 5.3 aimed therefore at demonstrating the benefits of increasing the understanding of the biochemistry within digesters and employing monitoring methodologies, which would then be followed by an appropriate management or control action. This research and demonstration programme was performed by the University of South Wales (formerly University of Glamorgan) under the IEE Biomethane Regions project Task 5.3. This task aimed at demonstrating the benefit of implementing management systems that included monitoring and control strategies to improve

the operation of AD plants operating at full-scale. To achieve an increase in methane production per plant above 10% was the target set within the project. Other aspects such as being able to increase waste throughput or increase digestate dewatering efficiency could also be part of the evaluations.

2. Introduction to the AD Plant under Evaluation

One of the AD plants evaluated within this project was the Rogerstone AD plant located in South East Wales. The plant was at that time owned by InSource Energy Limited. Its construction started in July 2010 and the plant was commissioned and started up early 2011. The technolgy was supplied by EnviTech Biogas and the main construction company was H2OK Systems. At design phase the aim was to deliver 330 kW of renewable electricity when in operation.

The plant capital costs were approximately £5M and the infrustructure benefitted from a £500k grant from the Welsh Assembly Government (WAG), administered by the Waste & Resources Action Programme (WRAP) Cymru in April 2009.

The plant was designed for a 10,000 tonnes per annum capacity to convert food waste largely from an adjacent food production plant owned by Premier Foods (RF Brookes), into energy to be used to help power its operations. The schematic of the plant is shown in Figure 1. The plant consisted of one 3090 m^3 anaerobic digester of the continuously stirrer tank reactor (CSTR) type, operated at mesophilic temperature (38^oC), which treated largely waste potato, potato sludge, depackaged food from ready meals and sludge from a dissolved air flotation (DAF) unit treating food factory wastewater arising from the adjacent factory. After one year in operation, additional waste substrates were also co-digested i.e. liquid waste from a rendering plant and delactosed whey. The plant included a pasteurisation step for all the wastes (Figure 2) and the biogas was utilised in a 500 kW combined heat and power (CHP) engine (Figure 3).

The digester was seeded with approximately 1600 m^3 of sewage sludge from a wastewater treatment plant in February 2011. The digester was fed typically with approximately 30-50 t wet weight per day of food waste. At the time of monitoring, the plant was operating on a relatively low organic loading of generally below 1.5 kg VS $m^{-3}.d^{-1}$ and fairly long retention times of around 60-100 days.

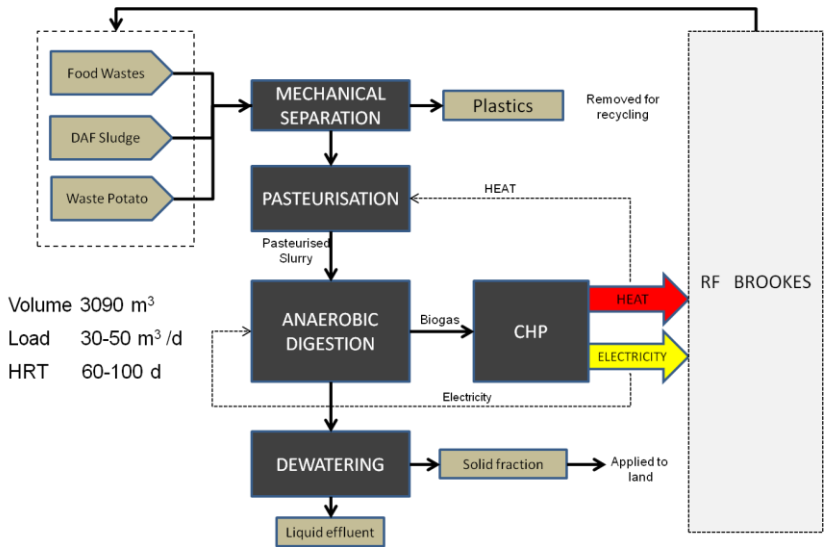


Figure 1 –AD plant's flowdiagram (Source: Insource Energy)

Figure 2 – Pasteurisation units

Figure 3 – 500 kW CHP unit

3. Monitoring and Control Methodologies Implemented

The monitoring and control support programme run from March 2011 and ended in June 2012 (i.e. over 500 days). Additional digestate dewatering evaluations as well as DNA sequencing for characterisation of Eubacteria species have been progressing through 2013 and 2014, however as the evaluations have not been completed, information has not been presented here.

Strategies for the routine monitoring and control of digesters are variable and depend on each plant individually. One general aim is being able to ensure that intermediates such as hydrogen, acetate and propionate do not accumulate resulting in acidification and process failure.

3.1 Monitoring Strategy Defined for the Demonstrator

The plant's SCADA system logged continuously a number of parameters i.e. the quantity of substrate feed added (wet weight in kg); pasteurisation and digester temperatures; biogas measurements in terms of composition of CH_4, H_2S and O_2; and power output (kW) from the CHP unit. Data was averaged weekly for graphical representation. However, no other measurements or characterisation were performed for the substrates or digester contents. Samples of digestate were normally sent out for external measurements to verify compliance for animal by products regulations (ABPR) in terms of pathogens and organic load and nutrients for the purpose of digestate liquor discharge and digestate fibre quality sent for farming land.

Therefore, as part of the work performed by USW a monitoring regime was devised for supporting the management of the plant, which included molecular techniques as well as traditional chemical analysis and the substrate flowrate and biogas data from the plant's SCADA system. The parameters monitored and related methodology is summarised in Table 1.

Weekly samples (500 ml) were collected from the Insource Energy AD Plant. During periods when VFAs were shown to be accumulating, samples were obtained daily from the digester. Samples were normally processed within 2 h for solid, pH and alkalinity determinations. Sub samples were also frozen within 2 h at -20^{o}C for subsequent DNA extraction, cation and VFA analysis.

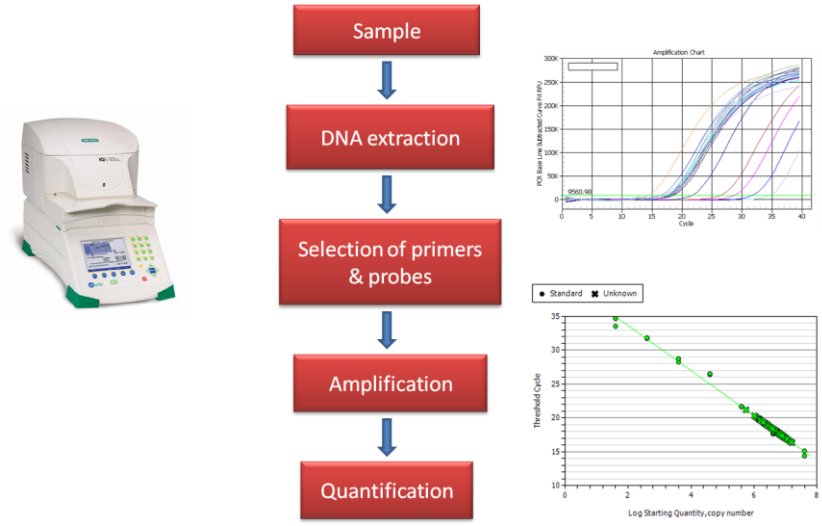
The measurements of solid content of the substrates provided an indication of the organic loading and how degradable and how much mineral content was being fed to the digester. Buffering capacity, pH, VFAs concentration indicated if acetogens and methanogens (slow growers) had been able to accompany hydrolytic and acid phase bacteria. Otherwise instability would occur with VFAs being produced in excess of the ones being converted to methane and carbon dioxide and therefore acids would accumulate and buffering capacity would reduce with potentially a reduction in pH. The level of cations being available in the digester were also important as excess could cause inhibition. Two measurements of trace elements were also performed using ICP, at the start of the operation as well as when VFAs started to increase. There are metal elements (discussed in Task 5.2 report) which are required for microbial growth and activity and if these metals are deficient digester performance can reduce.

Table 1 - Operational parameters, methodologies and sampling frequency

Matrices	Monitoring parameters	Methodologies	Sampling Frequency (2x or 3x analysis per sample)
Substrates	Solid content (total solids (TS) & volatile solids (VS))	American Public Health Association (APHA) standard methods (2005)	Weekly or for new substrates or when batches varied
	Cations (NH_4, K, Na and Ca)	Ion chromatography (Dionex ICS3000), with an IonPac CS12A column. Samples were centrifuged and then filtered through 0.45 µm pore size	Weekly or for new substrates or when batches varied
Digester Content	Solid content	As above	Weekly or daily when VFAs were high
	pH, partial alkalinity (PA), intermediate alkalinity (IA) and the Ripley ratio	pH using an electrode. PA and IA according to Ripley et al. (1986)	Weekly or daily when VFAs were high
	Individual Volatile Fatty Acids (acetic, propionic, isobutyric, n-butyric, isovaleric and n-valeric)	GC Clarus 500 with a TurboMatrix 40 Trap headspace sampler (PerkinElmer) according to Cruwys *et al.* (2002)	Weekly or daily when VFAs were high
	DNA and Microbial profiling	Genomic DNA was extracted using a PowerSoil DNA Isolation kit (Mo Bio Laboratories Inc.). DNA conc. determined based on absorbance at 260 nm using a NanoDrop 1000 Spectrophotometer (Thermo Scientific). Quantitative Polymerase Chain Reaction (qPCR) using a Biorad iQ5 system (Bio Rad Laboratories). Total EBAC estimated by targeting 16S rRNA gene sequences (Suzuki *et al.* 2000). Assays with primer and probe sets as per Yu *et al.* (2005)[a]. Calibration curves were perfromed in duplicate. DNA extracted from pure cultures were used as controls	Varied, samples were taken based other chemical data for correlation purposes and to better define control strategy. Storage of raw samples or extracted DNA samples were frozen at -20°C for analysis after a maximum of a couple of months.
	Cations (NH_4, K, Na and Ca)	As above	Weekly or Fortnightly

[a] targeting the 3 orders of methanogens (Methanobacteriales, Methanomicrobiales and Methanococcales) and 2 family level aceticlastic methanogens (Methanosarcinaceae and Methanosaetaceae)

Knowledge is still fairly limited on the microbial communities present in full scale anaerobic digesters and despite the importance of methanogens in waste mineralization as a result of their acetate and hydrogen conversion abilities, they have not been routinely monitored. Anaerobes are very difficult to measure by traditional techniques such as plate counts as they often cannot easily be cultured. Molecular methods have been used in this demonstrator using qPCR to count the number of gene sequences present in DNA extracted from a sample. The method uses "primer" and "probe" sequences unique to specific bacteria to amplify short DNA sequences only from those organisms. As the probe is incorporated into the amplified DNA it releases fluorescence that can be quantified and used to estimate the number of gene copies present in the sample. Figure 4 shows the main steps in qPCR analysis.



Fiure 4 - Main stpes in the qPCR metholodgy

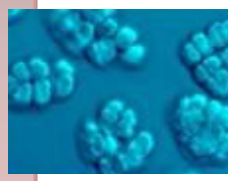
This molecular analysis has contributed to the first report of long-term monitoring of the diversity of methanogenic archaea in a full-scale digester treating food waste (Insource Energy AD plant), which in conjunction with monitoring of the VFAs and alkalinity has supported operational control and digester optimization by moderating organic loading, addition of trace elements and alkalinity. Further information can be found in an academic publication (Williams et al., 2013).

Key Microbes in Anaerobic Digesters:

Eubacteria (EBAC): Mostly acidogens e.g. *Clostridia sp*. Degrade organic material in the feed to produce VFAs and hydrogen. Become inhibited if VFAs and hydrogen are not removed by the action of methanogens.

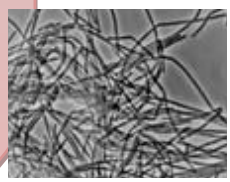
Methanosarcina sp. (MSC): Consume acetate (and hydrogen) to produce methane. High growth rate but requires high levels of acetate. Can dominate when acetate levels are high and when levels of ammonium and salts are high.

Methanosaeta sp. (MST): Consume acetate to produce methane. High affinity for acetate but slow maximum growth rate. Sensitive to inhibition by ammonia. Tends to dominate when underfeeding and low ammonia concentrations.



Methanobacter sp. (MBT): Consume hydrogen to produce methane.

Methanomicrobium sp. (MMB): Consume hydrogen to produce methane.



Methanococcus sp. (MCC): Consume hydrogen to produce methane; have some tolerance for high salt concentrarions. (Not detected in this digester).

3.2 Control Actions Implemented

Control actions can be variable and tyically range from variations in plant design (e.g. mixing, pretreatments for substrates) to regulation of substrate loading or substrate mixtures or addition of alkalinity or trace elements.

As part of the control actions specified in this demonstrator, 1000 L of EnVital mineral and trace elements (EnviTec Biogas) and 1000 L Kalic liquid lime (choice of alkali was made by the pant operator based on cost) (Tarmac Ltd) was added to the digester on day 272 to improve digester stability. At a later stage. a reduction of the input from rendering waste and whey was also suggested as the cations load was increasing significantly and mineral precipitation started to be a problem. In addition, the ammonia levels were influencing the abundance of acetate utilising methanogens, and therefore acetic acid background level was raised.

4. Monitoring and Control Results and Discussion

4.1 Substrate Solids Composition and Digester Start-up and Inital Operation

Table 2 includes the microbial profile for the inoculum (or seed) at star-up of the digester. The sewage sludge inoculum contained approximately 3.8 x 10^{10} gene copies ml^{-1} of total eubacteria. Assuming the population contained on average 6 gene copies per cell this would be equivalent to 6.4 x 10^{9} cells ml^{-1}. The methanogenic community was therefore dominated by aceticlastic methanogens from the family *Methanosaetaceae*. Methanogens from the orders *Methanomicrobiales* and *Methanobacteriales* were also present in the inoculum but at lower numbers and *Methanosarcinaceae and Methanococcales* were not detected i.e. < 200 gene copies ml^{-1}. *Methanosarcinaceae* although not detected at inoculum level, subsequently managed to proliferate to detectable levels once acetate concentrations in the digester increased and as substrates were all pasteurised, it was therefore assumed that they were present in the inoculum. *Methanosarcina sp.* have a lower affinity for acetate but higher maximum growth rate, whereas *Methanosaeta sp.* have a high affinity for acetate but a lower growth rate. This gives *Methanosaeta* a competitive advantage when the acetate concentrations are low in conventional sewage sludge digesters. The absence of *Methanococcales* in both the sewage sludge inoculum and later in this food waste digester was not surprising since these microbes grow typically within a high salt environment.

Table 2 - Microbial populations present in the sewage sludge inoculum

Microbial Target Group	Number of gene copies ml^{-1}
Total eubacteria	3.8 x 10^{10}
Methanosaetaceae	4.6 x 10^{8}
Methanomicrobiales	2.6 x10^{6}
Methanobacteriales	3.2 x 10^{6}

Figures 5 and 6 include the data gathered for microbial communities and chemical parameters in the digester throughout the monitoring period.

Over the first 100 days, the amount of substrate added to the digester generally increased from approximately 30 to 44 t wet weight d^{-1}. The typical solids composition of the substrates fed to the digester are shown in Table 3. VS introduced in the feed were reduced by approximately 86% (from approx. 6.34 to 0.88%). The biogas produced contained 55-58% methane and was used to generate between 158 and 336 kW power output over the first 10 months of operation. During the start-up period, the bicarbonate alkalinity in the digester increased from around 5000 to over 6000 mg $CaCO_3$ L^{-1}, the intermediate alkalinity decreased slightly and acetate was kept below 150 mg L^{-1}. Propionate, butyrate and other VFAs were not detected (limit of detection < 50 mg L^{-1}). This

is shown in Figure 5 in additon to cations Na, K and NH_4, increased up until 100 days and then were fairly constant up until after day 310 when additional substrates were also started to be digested.

Table 3 – Typical solid composition of substrates

Substrate	TS %	VS% ww	VS% dw
DAF sludge	6.97	6.27	91.32
Potato waste	4.37	3.89	92.21
Depackaged food waste	9.34	8.86	94.50
Rendering waste*	9.63	8.31	86.29
Delactosed whey waste*	54.20	47.20	87.10

*Only fed after approx. 310 day of plant operation

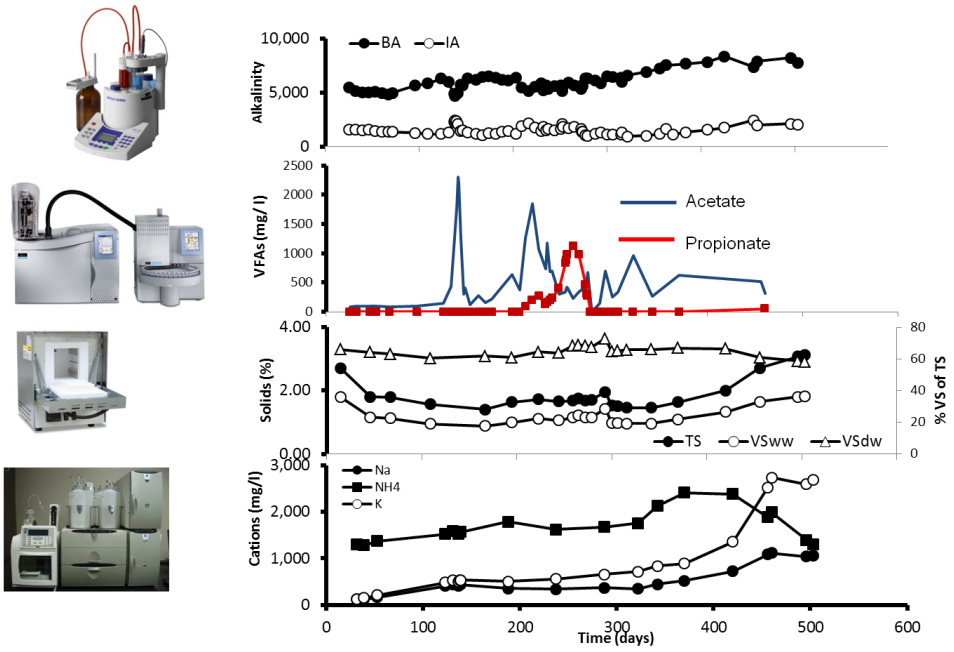


Figure 5 - Digester contents chemical monitoring over the 500 days period

As the digester started to be fed the composition of the digester contents gradually changed. Initially, the digester showed signs of being able to cope with additional feed with concentrations of VFAs below 200 mg L^{-1} and Ripley ratio below 0.3 indicating a healthy methanogenic activity. The CHP output generally corresponded well to the amount of feed added to the digester, as increases in the feeding rate were typically followed by increases in CHP output. However, reductions in feed were not always followed by proportional reductions in CHP output. This started to suggest that VFAs and residual undigested VS which had accumulated in the digester were then utilized.

Figure 6 shows the response from microbial diversity over the monitoring period and the correlation with VFAs. During the first 3 months of operation, total eubacteria in the digester increased to 5.4 x 10^{10} gene copies ml^{-1} and *Methanosaetaceae* increased to 7.4 x 10^{8} gene copies ml^{-1}. This increase suggested that the microbial populations were growing as a result of the introduction of organic substrates. The methanogenic community in the first 100 days was dominated by *Methanosaetaceae* (7.4 x 10^{8} gene copies ml^{-1}), whilst *Methanobacteriales* and *Methanomicrobiales* were present at approximately 6.5 x 10^{6} and 5.8 x 10^{6} gene copies ml^{-1}, respectively. Throughout the first year, the methanogenic population in the digester was dominated by *Methanosaetaceae*, suggesting that aceticlastic methanogenesis was the main route for the production of methane in this digester.

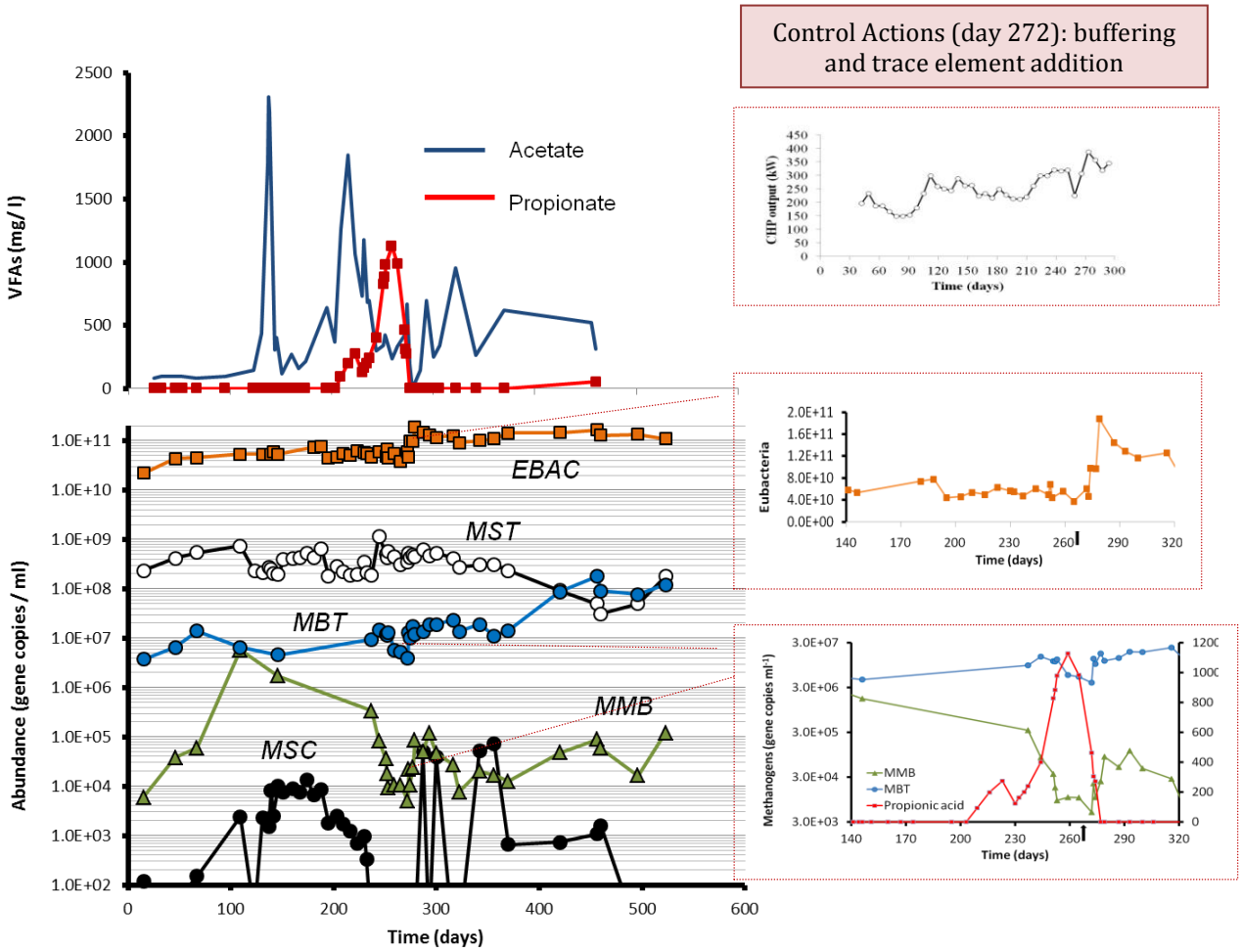


Figure 6 – VFA and microbial profile over 500 days monitoring period including the effect from trace element and alkalinity addition on populations, residual VFA and power produced

4.2 Imbalances in the Digester

Between day 109 and day 123 a sudden 69% drop in the 16S rRNA gene copy number of *Methanosaetaceae* from 7.4 x10^{8} to 2.3 x10^{8} copies ml^{-1}, followed by a

spike in acetate (maximum 2310 mg L^{-1}) was observed (Figure 7). Bicarbonate alkalinity dropped to 4700 from 6300 mg $CaCO_3$ L^{-1} and the Ripley ratio increased from 0.23 to 0.63. The pH never decreased below 7.4 from about 7.55, showing the deficiencies on relying on pH for process control. The higher acetate concentrations appeared to stimulate the numbers of aceticlastic methanogens from the family *Methanosarcinaceae* (Figure 6). *Methanosarcina* species were then able to compete better at higher acetate concentrations. When the activities of *Methanosarcina* and the recovering numbers of *Methanosaeta* brought the concentration of acetate back down below 500 mg L^{-1} the numbers of *Methanosarcinaceae* declined once again suggesting that they were no longer able to compete at the low acetate concentrations.

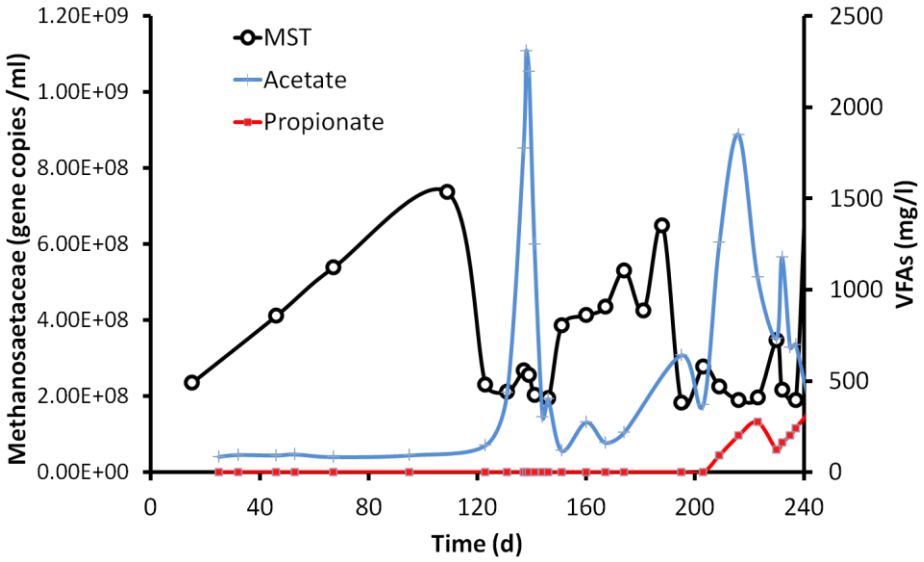


Figure 7 - Spikes in VFAs following decreases in the population of *Methanosaetaceae* (MST)

The cause of the sudden 69% reduction in the numbers of *Methanosaetaceae* over such as short period of time was not known (Figures 6 and 7). However, the drop in numbers coincided with a temporary reduction in the OLR from 0.83 to 0.64 kg VS $m^{-3}d^{-1}$ between day 105 and 119. Other possible explanations for this perturbation may be the presence of inhibitors in the feedstocks (e.g. detergents, spices, garlic or long chain fatty acids), trace elements deficiencies, elevated hydrogen, sulphide or as a result of operational actions. The 69% reduction in numbers over 14 days could not be explained by washout, and therefore the cells must have lysed and the DNA degraded making them no longer detectable by qPCR. This reduction in *Methanosaetaceae* clearly caused an imbalance in the rate of acetate consumption compared to acetate production allowing a build up of acetate (Figure 7). The comparatively low buffering capacity of the digester resulted in a drop in alkalinity and an increase in the Ripley ratio, which gave warning of a digester imbalance.

Another imbalance in the digester occurred later between day 209 and 272. There was an observed succession in VFAs with an increased level of acetic

followed by butyric and propionic acids. The concentration of propionic acid increased and reached a maximum of 1100 mg L^{-1}. Inhibition of eubacteria and the hydrogen-utilizing methanogens (*Methanomicrobiales* and *Methanobacteriales*) was observed at the same time as this increase (Figure 6). VS degradation and gas production was reduced during the propionic acid peak and unless a control action was performed, waste throughput would need to reduce at least or a possibility of digester failure was in the horizon (Figure 6). This build up of VFAs including propionate indicated a more serious imbalance of the digester; propionic acid is an effective indicator of process stress (see Task 5.2 report). Molecular monitoring was also very valuable here as it shown that reductions in the number of *Methanosaetaceae* which preceded the increase in acetate, but in this case *Methanosarcinaceae* did not grow to compensate. It is likely that whatever was inhibiting the *Methanosaetaceae* had also inhibited *Methanosarcinaceae* during this period of elevated propionate concentrations. In addition, a decline in the numbers of eubacteria and hydrogen utilizing methanogens of the orders *Methanobacteriales* and *Methanomicrobiales* was also observed (Figure 6). Much of the hydrogen flux in anaerobic digesters occurs between syntrophic associations of hydrogen-producing and hydrogen-consuming microbes. The reductions in the hydrogenotrophic capacity of the community could in theory result in increases in hydrogen concentrations, which would in turn shift the digester equilibrium towards more propionate formation and increasing inhibition. Unfortunately, hydrogen concentrations were not measured during this study, however it was possible that hydrogen levels were inhibitory.

4.3 Digester Control Based on Addition of Trace Elements and Alkalinity

The increasing VFAs and declining alkalinity, VS destruction and biogas production in conjunction with significant decreases in the microbial populations were a cause of concern and hence a decision was made to add trace elements and lime to the digester on day 272. This proved to be a successful intervention which strongly stimulated microbial growth and resulted in an immediate reduction in the VFAs and increase in VS destruction (Figures 5 and 6). Eubacterial populations were shown to increase from 5.0 $x10^{10}$ to 1.4 $x10^{11}$ gene copies ml^{-1} whilst *Methanosarcinaceae* increased from being undetectable to 5.0 $x10^{4}$ gene copies ml^{-1}. Also, the numbers of *Methanobacteriales* and *Methanomicrobiales* increased following the control action but it is not clear if it was a result of an increase in availability of limiting nutrients or decreased inhibition from inhibitors such as hydrogen, or from the additional hydrogen being available as part of the conversion of propionic acid (Figure 6). CHP output increased between day 273 and 280 following the addition of lime and trace elements to the maximum achived so far during the monitoring period.

4.4 A Following Period of Ammonium inhibition and a Resultant Shift in Methanogenic dominance

New feedstocks, including waste from a rendering plant and delactosed whey were fed to the digester from day 343 and day 411, respectively. Addition of these feedstocks caused noticeable changes in the digestate characteristics with respect to TS and VS (Figure 5). Also Na and K levels were shown to increase sharply when delactosed whey was added, reaching dissolved concentrations on day 460 of 1.1 and 2.7 g L^{-1}, respectively. Ammonium concentrations also increased in the digester between day 322 and day 370 from about 1500 to 2400 mg L^{-1} (equivalent to a free ammonia concentration of 104 to 166 mg L^{-1}, respectively). Ammonium concentrations >2250 mg L^{-1} (equivalent free ammonia of >156 mg L^{-1}) had a strong impact on the microbial community structure and activity and were shown to selectively inhibit *Methanosaetaceae* (Figures 6 and 8). Ammonia has been stated to be an inhibitor of methanogens, however, different species of methanogens have different tolerances to ammonia and as the aceticlastic methanogens declined methanogens from the orders *Methanobacteriales* (hydrogen utilisers) increased resulting in a shift of dominance from aceticlastic to hydrogenotrophic methanogens. Between day 456 and day 496, the hydrogen utilizing methanogen from the *Methanobacteriales* order became the dominant methanogen, with gene abundances between 7.7 x 10^7 and 1.8 x 10^8 copies ml^{-1} (Figures 6 and 8).

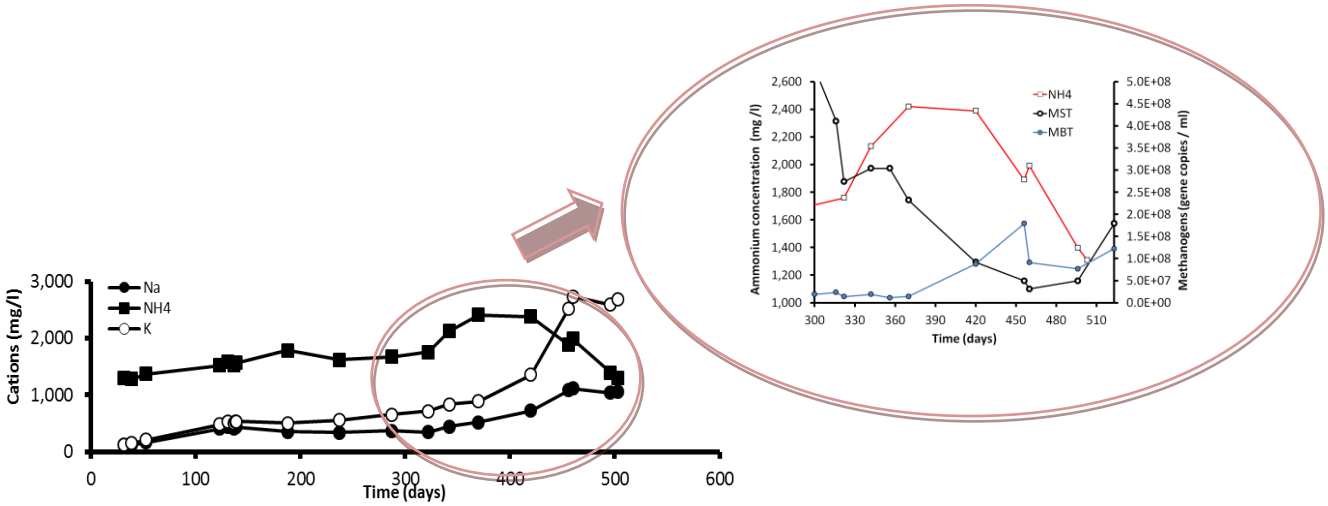


Figure 8 - Level of cations in digester contents and resultant ammonium inhibition and shift in the dominant methanogen from *Methanosaetaceae* (MST) to *Methanobacteriales* (MBT)

A reduction of input of these materials was also suggested and performed from around day 380 largely due to alkali precipitations occuring which were affecting pumps and pipes and dewatering membrane operations but also from some concern over ammonia toxicity. If aceticlastic methanogens are inhibited, VFA concentrations increase and when there are residual VFA levels then there is a sign of digester instability and a reduced waste stabilisation and energy recovery

is achieved. When the ammonium concentration later declined to below 2000 mg L^{-1} (eq. free ammonia conc. of 139 mg L^{-1}), the aceticlastic methanogens recovered whilst the hydrogenotrophic methanogens declined suggesting possible competitive interactions between these groups of bacteria.

5. Conclusions and Importance of this Demonstrator

This study highlighted the value of regular monitoring of full scale commercial digesters to track highly dynamic chemical environments as well as **microbial community structure and function**. The constant state of flux of a typical commercial digester was verified and it is important that such an understanding and demonstration is disseminated to digesters` designers and operators. The type of substrate and its inherent characteristics and loading regimes has been shown to influence the selection of methanogenic dominance. It is important to understand that the **microbial community and their interactions form the basis for the waste treatment and energy production "engine" of the plant** and unless they are monitored they are unable to be cared for, and performance will decrease. The levels of the key metabolic intermediates such as acetate and propionate were seen to spike up on several occasions indicating instability in the digester and these spikes corresponded to significant changes in the composition of the microbial community.

The results from this study have shown how routine monitoring of microbial populations in conjunction with alkalinity and VFA monitoring can provide an enhanced understanding of the complex processes occurring within the digester and can be used to help manage operational control to increase stability and improve digester performance. In addition, characterization of the composition of feedstocks in terms of TS, VS, CHN and salts helps to understand the likely changes that will occur in the chemical environment of the digester and how it may influence the composition of microbial populations. Knowledge of the types and abundance of methanogens in the digester is important in order to be able to monitor their responses to various inhibitory substances (e.g. salts, ammonia) and changes in substrate compositions.

By better understanding the microbial process that is occurring, it is possible to intervene in the process to better manage it. The objective of any management strategy would be to optimize organic degradation and the output of biogas and prevent system imbalances. It was possible to improve digestion conditions when digester health was fragile and increase biogas production by adding trace elements and alkalinity. The co-digestion of difficult substrates for example high in nitrogen and cations may be possible in a moderate way by monitoring frequently biochemical parameters in the digesters. The increase in propionate levels continued to be a good indicator of disruption in the digestion food chain. However, the level of severity of a shock and inhibition indicated by increased VFA levels (being total or propionate) was much clearer when having access to the levels of bacterial populations namely methanogens themselves. Attributing

a level of severity of a shock and evaluating the reasons behind performance decrease or defining more exactly the causes and dates when digester started to be affected could only be done by directly monitoring the microbial population themselves, as even VFA levels were shown to have a delay in response of 8-14 days as shown in Figure 7.

The addition of trace elements to digesters on a regular basis is normally costly and actual impact can only be evaluated by a thorough monitoring of the digester operation and performance. In addition, trace elements can bring a negative environmental performance to digestates with increase levels of (heavy) metals. So a better judgement of the frequency and need for these additions based on microbial communities benchmarking would be valuable.

Control actions such as the reduction of feed rate and the timing for the addition of micronutrients based on microbial abundance and diversity allowed maintenance of digester stability and an increase in power produced through increased methane generation up to 400 kW from an initial 250 kW. Pushing organic loading rates can be performed if microbial populations are being monitored and are found to be safe to do so.

Used on a regular basis, this type of monitoring proved to be a useful management tool for assessing the status of the digester with regards to its stability, predicting process trends and allowing decisive management actions to be taken.

6. References

APHA (2005) Standard methods for the examination of water and wastewater (21st ed.). American Public Health Association, Washington, DC, USA.

Cruwys, J.A., Dinsdale, R.M., Hawkes, F.R., Hawkes, D.L. (2002). Development of a static headspace gas chromatographic procedure for the routine analysis of volatile fatty acids in wastewaters. J Chromatogr A. 945, 195–209.

Esteves S., Miltner M. And Puchas K (July 2013) Monitoring Review and Guide for the Optimisation of Anaerobic Digestion and Biomethane Plants. IEE Biomethane Regions project deliverable Task 5.2 Report.
http://www.walesadcentre.org.uk/Controls/Document/Docs/D.5.2_Full%20Monitoring%20Report%20and%20Economic%20Info.pdf

Ripley, L.E., Boyle, W.C., Converse, J.C. (1986). Improved alkalimetric monitoring for anaerobic digestion of high-strength wastes. J. Water Pollut. Contr. Fed. 58, 406-411.

Suzuki, M.T., Taylor, L.T ., DeLong, E.F. (2000). Quantitative Analysis of Small-Subunit rRNA Genes in Mixed Microbial Populations via 5' – Nuclease Assays. Appl. Environ. Microbiol. 66, 4605-4614.

Williams J, Williams H, Dinsdale R, Guwy A and Esteves S (2013). Monitoring methanogenic population dynamics in a full scale anaerobic digester to facilitate operational management. Bioresource Technology 140: 234-242

Yu, Y., Lee, C., Kim, J., Hwang, S. (2005). Group- Specific Primer and Probe Sets to detect Methanogenic Communities Using Quantitative Real-Time Polymerase Chain Reaction. Biotechnol. Bioeng. 89, 670-679.

7. Acknowledgements

The authors would like to acknowledge Insource Energy (Rogerstone) specifically John Scott, Justin Strutt, Robert Hickman, Darren Smith and Patrick Taylor for the collaboration and for allowing access to the digester, the provision of samples and on-line energy data and for implementing suggested operational changes during this study. This study has been supported by the Welsh Government and IEE Biomethane Regions Project, which aims to demonstrate the impact of management systems in full-scale biogas and biomethane plants for increasing biomethane recovery. Microbial profiling methodology was devised by the Wales Centre of Excellence for Anaerobic Digestion funded by the European Regional Development Fund with also input from colleagues working on the LCRI Convergence Energy programme. The research and views described above are those of the authors and do not reflect the opinions of the funders.