BBSRC NIBB AD Network
Proof of Concept Awards: Titles and Summaries

2014 Proof of Concept Awards

POC2014001
Shotgun metabolomics in anaerobic digestion
Anaerobic digestion (AD) involves a complex network of interactions between a community of microbes which are difficult to accurately monitor. By using modern molecular techniques, it should be possible to identify key indicators of AD performance and health that in addition to helping operators maximise output from their systems can be used to populate computer-based models to improve our understanding of the AD process. One such set of indicators that could be targeted for measurement is the complement of small molecules (metabolites) produced on AD. Although the utility of metabolomics in this field has been posited a number of times, this approach requires specialist equipment and knowledge in order to be effectively executed, and is likely to require the development of specific preparation protocols for successful deployment. We propose a shotgun metabolomics approach using a state-of-the-art, ultra high resolution instrument to determine the speed, magnitude and reproducibility with which soluble metabolites change in a model AD system with the view of generating preliminary data for a larger application making use of this technology in AD.
Researchers: PI: Dr James Chong Department of Biology, University of York, Wentworth Way, Heslington, York YO10 5DD james.chong@york.ac.uk CO-I Prof Jane Thomas-Oates Department of Chemistry, University of York, Heslington, York YO10 5DD jane.thomas-oates@york.ac.uk Dr Julie Wilson Departments of Chemistry and Mathematics, University of York, Heslington, York YO10 5DD julie.wilson@york.ac.uk

POC2014011
Effective mass transfer of hydrogen into digester mixed liquor for biomethanisation of biogas CO_2
Hybrid technology combining renewables-driven electrolytic H_2 production with hydrogenotrophic methanogenesis to allow the biochemical reduction of CO_2 to CH_4 is possible and has been demonstrated. The concept has primarily been developed using purified component gas streams fed into a bioreactor in which a pure culture is maintained, resulting in a product gas with a CH_4 content >95%. The approach has also been used with biogas as one of the input gases using non-type strain methanogens which adapt naturally under non-sterile conditions; again the output gas stream can be >95% CH_4. What has proved more difficult is the in-situ conversion of bicarbonate in conventional digesters through H_2 injection: this appears to be limited by the effective mass transfer of H_2 into the system and by the need to maintain digester buffering capacity. Although CO_2 reduction involves some energy loss, this approach maximises the conversion of the available carbon in waste biomass and would provide an efficient means of in-situ biogas upgrading without additional methane slippage. The issues of energy balance can be addressed through process integration and optimisation, and a favourable overall balance can be shown when both direct and indirect energy inputs are taken into account. The concept explored in the proposed research is whether the use of a specially-designed membrane diffuser can improve the mass transfer of hydrogen into a digester, and in so doing maintain a CH_4 concentration >95% in the product biogas when using food waste at a typical biomass loading as the feedstock.
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POC2014012

Development of anaerobic biomass support particles for effective membrane cleaning

There is increasing interest in membrane technologies for anaerobic treatment of both municipal and process wastewaters. These can be configured in a number of ways using membranes of different types. Membrane fouling is one of the key issues in this type of technology and effective low-cost and low-energy in situ methods are needed to maintain the membranes clean. In conventional aerobic membrane bioreactors, which are now widespread in the water industry, the most common method used is air scouring and gas scouring using recirculated biogas has also been adopted in many prototype anaerobic systems e.g. submerged anaerobic membrane bioreactors (SAnMBR). These cleaning systems can be energy intensive, however, and unlike aerobic systems there is no advantage in circulation of large volumes of gas since there is no requirement for oxygen transfer in the equivalent anaerobic technology. The proposed research looks at the concept of developing biomass support particles of around neutral buoyancy with surface properties such that when pseudo-fluidised around the membrane they will provide a mild scouring effect. In this way the energy input will be minimised to that needed to maintain the support particles in motion. The second advantage of this system is that the support particles will permit higher concentrations of microbial biomass within the reactor and reduce the solids loading on the membrane, allowing operation at higher flux rates.

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POC2014016
Production and extraction of C3 and C4 aliphatic carboxylic acids from the anaerobic digestion of waste blood as a model substrate
This proof-of-concept project focuses on diversification of anaerobic digestion (AD) into the field of industrial biotechnology through the production and harvesting of butyric and propionic acids as intermediate bulk chemicals. These products have value and existing large-scale markets in their own right, and can also be considered for further bio-transformation and as the basis for an extended biorefinery concept. The research is based on the use of animal blood produced in abattoirs, a negative-value waste material, as a fermentation substrate since its high nitrogen content provides buffering to allow the accumulation of acid products at high concentrations without detriment to the acidogenic population due to low pH. Part of the research will focus on proving the concept that stable high rates of acid production can be achieved and that reactor operating parameters can be used to manipulate systems biology to produce a high quality output under non-sterile conditions. This builds on and will further contribute to recent advances in understanding metabolic pathways and syntrophy in anaerobic digesters that can lead to acid accumulation, and the mechanisms that control this. The second part of the work focuses on selection and preliminary testing of potential extraction methods that could be applied at large scales, ranging from conventional solvent extraction to more advanced membrane-based systems that could be used in situ in continuous processes. The research will make a preliminary assessment of issues relating to design, cost and resource requirements and environmental impacts of this novel biotechnology.
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2015 Proof of Concept Awards

POC2015001
Redesigning hydrolysis reactors for the development of high power density advanced anaerobic digestion enabling containerised electricity production from agricultural residues.
The rate limiting step of Anaerobic Digestion (AD) of cellulose feedstocks is accepted to be the bacterial hydroylsis of cellulose (Mumme, 2010). These bacteria form a predominant part of the cow microflora. Their activity in terms of hydrolysing cellulose in the cow's first fore---stomach, the rumen, is 10---30 times quicker than man---made AD plant. Oxford University is working to develop a high power density Advanced Anaerobic Digester, mimicking the mechanisms used by cows. On-going theoretical studies have led to our improved understanding of the underlying mechanism by which bacteria, growing as biofilms hydrolyse cellulose, and that conventional AD technologies are far removed from the way a cow operates. Initial work encourages us to believe that there are several pathways to achieving this High Power Density Advanced Anaerobic Digestion (AAD). A laboratory scale reactor, based on the operation of a cow's rumen has been built and will be used to test these pathways. This reactor represents the first stage in a two stage digester, and is predominantly concerned with improving the rate of hydrolysis of cellulose. The solid fraction of the biomass is retained in the reactor, whilst the interstitial liquid is recirculated. There is no agitation or stirring. Bulk mass and heat transfer are via the recirculation of the liquid. This is much more reflective of how a rumen works. We hope to demonstrate an increase in the mass of feedstock hydrolysed per unit volume of reactor compared to traditional continuous stirred tank reactors (CSTR).
Applicant: Professor Ian Thompson – University of Oxford

POC2015003
Recovery and purification fatty acids and nutrients from anaerobic digester fluids using integrated membrane freeze-thaw (MFT) processes
This project will investigate the utility of a low energy, integrated membrane freeze thaw process (MFT) for the recovery of fatty acids and nutrients (N and P) as salt crystals or concentrated fluids. The MFT process is a hybrid process that combines the ability of a RO membrane process to separate water from dilute salt solutions while freeze concentration overcomes the membrane limitations posed by high osmotic pressures for the concentrated solutions. Thus together the low energy process produces clean water and series of fractionated concentrates. It has few chemical inputs where separation principles are physically based. Preliminary calculations suggest that this process will only require 15% of the energy produced by CHP of digester gases to power the process. Using fluids from AD systems, the consortium will investigate the concentration/crystallisation process and its potential problems. There are also potential benefits for the MFT process integrated into large scale AD CHP systems and these will be investigated and the costs of the process evaluated. Finally, the quality and value products derived from the fractionation process will be assessed as chemicals or as growth media components. If successful, the study will also open the way for the potential enhancement of digester kinetics by in-situ product recovery and further process integration and optimisation.
Applicant: Dr Robert Lovitt – Swansea University

POC2015008
Microbial Enhancement of Phyto Active Compound in Digestate
The aim of the project will be to improve the quality of anaerobic digestion residues so that the growth rate of crops and plants grown on digestates will be increased. This will be achieved by investigating the ability of naturally occurring microorganisms to produce plant growth promoting factors (i.e. plant hormones such as auxins) in the digestate. The improved degradation of plant material in the anaerobic digestion process also may also release plant growth promoting compounds (i.e. humic acids/soluble lignins) and the mechanisms to increase the amount of these compounds will also be investigated. The development of the presence of plant disease biocontrol agents in digestates will also be researched. The results from this will lead to an improved product which promotes the enhanced productivity of crops, leading to greater food security and expanded markets of digestate for the UK anaerobic digestion industry.
Applicant: Professor Richard Dinsdale – University of South Wales

2016 Proof of Concept Awards

POC2016002
Novel bioelectrodes for energy positive ammonia removal from municipal wastewater
Ammonia can cause eutrophication and be toxic to fish and other aquatic wildlife. Thus the removal of ammonia from wastewater is frequently required as part of general treatment processes, to either protect receiving waters, or as a precursor to N-removal. Conventional ammonia removal technologies use oxygen, and the more ammonia that must be removed the greater the quantity of oxygen required. This is not merely expensive, but arguably unsustainable as the carbon footprint of the process is such that water companies find themselves “destroying the planet to save the river”. The objective of the work proposed here is to develop a novel microbial technology to remove ammonia-nitrogen without a need for oxygen. Rather than consuming energy the process will actually generate energy. The technology is based on the use of Anaerobic Bacteria Respiring Ammonia, a new group of microorganisms capable of donating the electrons encapsulated in ammonia to an appropriately poised electrode, a process that is of considerable fundamental interest and constitutes a new and unexpected shortcut in the nitrogen cycle. We envisage scope for this technology in the treatment of waste streams with low to medium high ammonia concentrations, where alternative methods aimed at ammonia recovery are economically less attractive.
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POC2016012
Electrode interface to control and extend metabolic outputs of AD microbial communities
AD is a well-recognised green biotechnology. It is underpinned by complex microbial communities, where different species degrade organic matter into a set of metabolic intermediates that are then used by methanogenic microbes to produce methane. Currently, we lack any direct means to control the population dynamics within this microbial world and the ensuing metabolic output types and reliability. This proposal will overcome this major limitation by developing an electrical approach to control and manipulate AD microbial communities. This approach relies on our understanding of the AD process as a thermodynamically driven conversion system, where the population size of different microbial groups are primarily determined by the availability or absence of strong electron acceptors. By using electrodes poised at appropriate redox potentials as electron sinks and donors, we will aim to foster and maintain specific methanogenic groups. This will result in a direct control of the output levels and reliability of the AD process, thereby making this biotechnology more controllable, tunable and reliable. In turn, this will reduce risk of AD investments and increase production yields. Developed further with additional research, our electrical approach will be applied to other, bespoke communities in which we control their rich metabolic repertoire towards production of desired metabolites.
Applicant: Orkun S Soyer, School of Life Sciences, University of Warwick

POC2016014

Computational Methods for Anaerobic Digestion Optimization – Act 1 (CoMAnDO1)

Anaerobic digestion (AD) is often used to treat sludge, via mixing with bacteria that biodegrade sludge, producing methane-- - rich biogas that may be harnessed via combined heat and power technology for energy recovery. However, there remains a pressing need to optimize digester mixing to maximize energy recovery.

To predict optimum digester mixing, we must determine to what extent biogas o utput is influenced by AD flow patterns; flow patterns that are determined by digester configuration, inflow mode, sludge rheology and mixing regimes. Research is lacking in this area. Traditional approaches to digester design are rooted in empiricism rather than science, and design standards focus on treated sludge quality, not gas yield and energy consumption.

The challenge is to improve digester performance and maximize biogas yield. An innovative solution is to simulate simultaneously the hydrodynamic and microbiological processes found in AD. We hypothesize there is a direct link between mixing-induced turbulence and biogas yield. Our focus is to simulate these interrelationships in a complex multiphase, non-Newtonian fluid environment. However, to achieve this, there remain several challenges to overcome; viz. computationally efficient hydrodynamic modelling and effective coupling techniques.

CoMAnDO1 will address the first of these limitations and will deliver the proof of concept necessary to develop Lattice Boltzmann (LB) modelling as a robust, rapid alternative to computational fluid dynamics (CFD) modelling to simulate the hydrodynamic environments within digesters. This will lead to a major research project in which LB and biokinetic models will be coupled to control the hydraulic and biochemical performance of AD.

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POC2016018
Metabolic Modelling meets Anaerobic Digestion (M3AD)
AD has been traditionally represented as a black (sometimes grey) box, where a relatively undefined collection of macro-scale processes lead to the degradation and conversion of a complex feedstock into a mixture of products of anaerobic metabolism. For simplicity (and sometimes lack of information or details), this modelling approach has generally overlooked the metabolic processes led by microorganisms. New powerful biomolecular tools, supported by advances in metabolic analysis software, provide now the possibility of including this microlevel in the analysis of AD processes. This PoC will attempt combining multispecies metabolic modelling (i.e. a metabolic model that includes the main metabolic features of the microorganisms in the microbial communities carrying out the process) with current models of anaerobic digestion (ADM1). Based on metagenomic information from the literature, we will build a metabolic model involving all the physiologically relevant metabolic pathways present in the most abundant species in the community, leading to CH_4, H_2 and VFA production. The metabolic model will be linked to the model presented by Poggio et al (2016), which (based on a combined biochemical and kinetic approach) combines rigorous feedstock characterisation with ADM1 to represent an AD process. Our purpose is to demonstrate the feasibility of analysing AD through a multi-scale approach, ranging from the metabolic micro-level to the kinetic macro-level, to simulate and generate predictions for the design and optimization of full-scale processes.
Applicant: Dr Claudio Avignone Rossa, Department of Microbial Sciences, School of Biosciences and Medicine, University of Surrey, Guildford GU2 7XH

POC2016019
Biobutanol production via bioelectrochemical reduction of butyric acid
This proof-of-concept project focuses on diversification of anaerobic digestion (AD) into the field of industrial biotechnology through the conversion of one of its intermediate bulk chemicals, butyric acid, into butanol using a microbial electrochemical system (MES). Butanol has many industrial applications, and it has received great interest in its use as direct replacement for petrol or as fuel additive. Butanol is superior to ethanol in this regard because it has higher energy content, higher octane number, lower volatility, is less hydroscopic and less corrosive. Therefore, it has value and an existing large-scale market. This research will focus on demonstrating that selective butanol production from butyric acid can be achieved using MES and that this MES can be integrated with the AD process for overall energy and material recovery efficiency. The operating parameters will be optimised for butanol production and this will further contribute to recent advances in understanding the mechanisms that control MES. The overall performance of the MES for butanol production from butyric acid will be evaluated in terms of electron and energy utilisation, as well as long-term stability.
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