



Synthetic Biology Research Centre Newsletter

University of Nottingham

Issue 6 December 2017

SBRC Centre Manager Update

The SBRC high throughput robotics and gas fermentation facilities are now fully operational. In combination with our growing team of experts in synthetic biology, their availability has established the SBRC as a world-leading research capability dedicated to the exploitation of C1 chassis. They are allowing the implementation of cutting edge strategies within the arena of metabolic engineering.

Thanks to the commissioned robotics capability we have implemented a wide range of automation protocols to support ongoing SBRC goals including:

- expanding our capability for controlling gene expression through the derivation and testing of an extensive range of promoter bioparts;
- the combinatorial assembly of bioparts in the requisite synthetic operons required for the production of SBRC targets;
- the evolution of the pivotal enzymes involved in the synthesis of SBRC targets;
- development of high throughput assays for the identification of CRISPR Cas9 target sequences in SBRC chassis, and;
- the isolation of plasmid replication regions that are temperature sensitive for replication.

The commissioning of our gas fermenters in continuous operation using H₂ and CO₂ as sole energy and carbon source has initiated a programme to scale-up the testing of our modified chassis for the production and accumulation of SBRC targets.

There are many other lines of enquiry underway which align with developing engineered chassis capable of being optimised for industrial production of platform chemicals from C1 feedstocks. As part of our ambition to generate effective chassis, new funding sources to future-proof our research activity are continually targeted. For example, through BBSRC funding we organised three international workshops on "Value from Waste Gas" to underpin GCRF funding applications, two Expressions of Interest for which have recently been submitted. These workshops were hosted in Mumbai, New Delhi and Shanghai. Additional grant funding has recently been secured. Of particular note are:

- **ENGICOIN:** Engineered microbial factories for CO₂ exploitation in an integrated waste treatment platform" **€6.9 million**. Jan 2018 – Dec 2022. - 12 European Partners.
- **BIOMETCHEM:** Sustainable production of added value chemicals from SynGas-derived methanol through Systems and Synthetic Biology approaches. **€1.8 million**. April 2018 - Mar 2021

The entire SBRC group, which includes researchers working on other metabolic engineering projects related to health as well as work on other industrial carbon feedstocks, currently comprises 53 PhD students including 16 BBSRC DTP studentships plus 40 post-docs and technicians. The new grant funding awards will bring six additional post-doctoral positions to the SBRC. Within the last 12 months we have welcomed 15 international visitors to work for varying periods of time in our labs. At any one time the SBRC workers number more than 110, representing a significant metabolic engineering capability for the UK and for international collaborations.

Dr Alan Burbidge, SBRC Centre Manager
alan.burbidge@nottingham.ac.uk

Jobs available at the SBRC - Nottingham

Research Associates in Synthetic Biology

We invite applications from suitably qualified candidates for research associates focusing on metabolic engineering. For more information and how to apply please visit:

<http://www.nottingham.ac.uk/jobs/currentvacancies/ref/MED439517>

Closing date 13 December 2017

Senior Research Fellow in Responsible Research and Innovation

We invite applications from suitably qualified candidates for a senior research fellow focusing on Responsible Research and Innovation. For more information and how to apply please visit:

<http://www.nottingham.ac.uk/jobs/currentvacancies/ref/MED400617>

Closing date 13 December 2017

Technician

Applications are invited to the above role to provide technical support to Synthetic Biology Research Centre (SBRC)-Nottingham research projects. For more information and how to apply please visit:

<http://www.nottingham.ac.uk/jobs/currentvacancies/ref/MED430317>

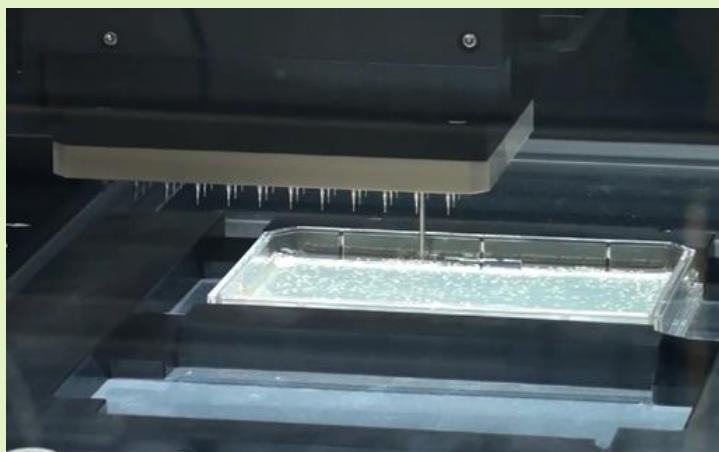
Closing date 22 December 2017

Research News

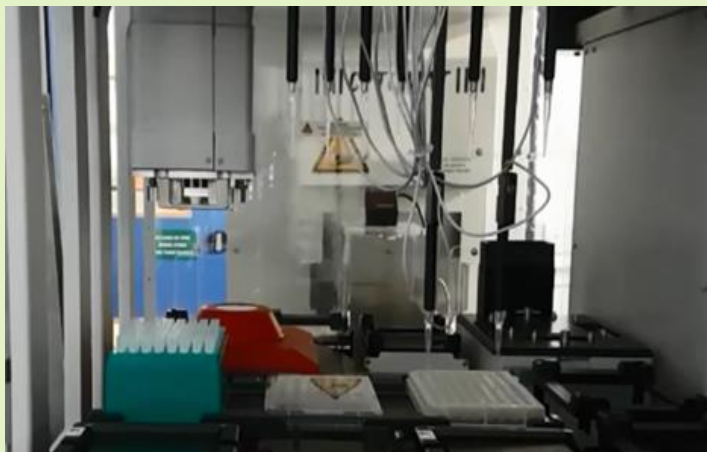
Accelerating metabolic optimisation in *Cupriavidus necator*

By Dr Magdalena Jonczyk

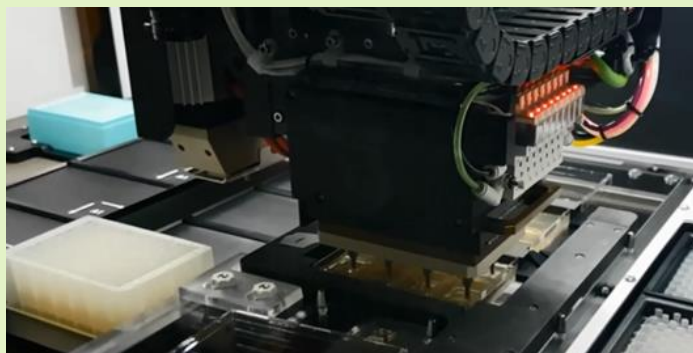
The SBRC aims to engineer microbial strains that convert greenhouse gasses into valuable platform chemicals and biofuels. *Cupriavidus necator* is an aerobic bacterium that can grow on C1 gases, such as CO and CO₂ using H₂ as an energy source to synthesize compounds of potential industrial importance. The SBRC robotic suite is being used to generate and evaluate synthetic DNA parts (e.g.: promoters, RBSs, terminators, selection markers and genes) to help accelerate the metabolic optimisation in *C. necator*. The DNA parts are designed as interchangeable blocks or modules that can be easily mixed and matched within a modular vector. A large number of randomised combinations can then be quickly screened to identify an optimal set of DNA parts. As a part of this project, a library of synthetic promoters, derived from *C. necator* native PhaC promoter, was created. The non-consensus sequence of the promoter was randomised and the pool of promoter variants transformed into *E. coli* and *C. necator*. Promoters that displayed significant difference in their activity were then identified and sequenced. Use of the robotic system enabled many more promoter variants to be analysed than would be possible manually and hence provided a comprehensive library in a short timeframe. Furthermore, work has been undertaken to produce a standard, easy to use and reliable pipeline of sample tracking and data analysis that can be applied to any future variants screening on our robotic platforms. An easily accessible large repository of data generated during the various screens could also contribute to better understanding of regulatory nodes that control metabolic traffic in *C. necator*.



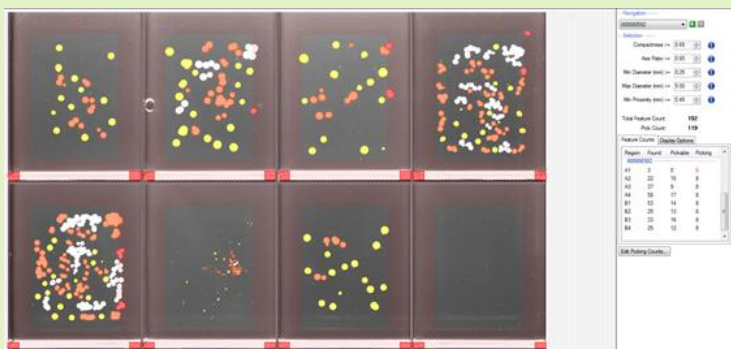
Colony picking (*E. coli*): each colony contains a unique variant of the promoter.



Cherry picking: Robot picks selected variants of the synthetic promoter.



Transformation of *C. necator* with a pool of synthetic promoters.



Colony picker (Qpix) identifies colonies to pick on an agar plate.

Aerobic Gas Fermentation Unit

By Dr Rajesh Bommareddy and Dr Yanming Wang

An efficient and safe aerobic gas fermentation system (on CO₂, H₂ and air mixture) has been developed for characterising the *Cupriavidus necator* strains that are being engineered at the SBRC. This is a parallel bioreactor system (8 x 750 ml reactors) which has been modified to suite the requirements of the process and safety needs. Flame detector, gas leak detectors are coupled to solenoid valves which shuts off the gases when triggered. Oxygen concentrations are continuously monitored and controlled below flammable range to avoid any flammability issues and addressing the O₂ mass transfer issue at the same time. A complete gas fermentation system is now commissioned. The success was demonstrated by a duplicate high cell density continuous cultures of *C. necator* (WT) growing on H₂, air and CO₂ as energy and sole carbon source respectively. Engineered *C. necator* strains producing isopropanol, 2,3-butanediol on CO₂ has also been successfully demonstrated using the gas fermentation system.



Recent Publications

1. [Multi-timescale analysis in synthetic biology: a kinetic model for 3-hydroxypropionic acid production via beta-alanine](#)
MP Dalwadi, JR King, and NP Minton
Journal of Mathematical Biology (2017) DOI: 10.1007/s00285-017-1189-3
2. [Applying asymptotic methods to synthetic biology: modelling the reaction kinetics of the mevalonate pathway](#)
MP Dalwadi, M Garavaglia, JP Webb, JR King, and NP Minton
Journal of Theoretical Biology (2017) DOI: 10.1016/j.jtbi.2017.11.022
3. [Mathematical modeling of chemical agent removal by reaction with an immiscible cleanser](#)
MP Dalwadi, D O'Kiely, SJ Thomson, TS Khaleque, and CL Hall
SIAM Journal on Applied Mathematics, 77: pp 1937–1961 (2017), DOI: 10.1137/16M1101647
4. [The effect of weak inertia in rotating high-aspect-ratio vessel bioreactors](#)
MP Dalwadi, SJ Chapman, JM Oliver, and SL Waters
Journal of Fluid Mechanics, 835: pp 674–720 (2018), DOI: 10.1017/jfm.2017.760
5. [Characterisation of a 3-hydroxypropionic acid-inducible system from Pseudomonas putida for orthogonal gene expression control in Escherichia coli and Cupriavidus necator](#)
Erik K. R. Hanco, Nigel P. Minton, and Naglis Malys
Sci Rep. 2017; 7: 1724. (2017) DOI: 10.1038/s41598-017-01850-w PMCID: PMC5431877
6. [Improving gene transfer in Clostridium pasteurianum through the isolation of rare hypertransformable variants](#)
Alexander Grosse-Honebrink, Katrin M. Schwarz, Hengzheng Wang, Nigel Minton and Ying Zhang
<https://doi.org/10.1016/j.anaerobe.2017.09.001>
7. [13C-assisted metabolic flux analysis to investigate heterotrophic and mixotrophic metabolism in Cupriavidus necator H16](#)
Swathi Alagesan Nigel P. Minton Naglis Malys Volume 14; DOI:10.1007/s11306-017-1302-z (2017)

Conferences

CALENDAR

Of Key Synthetic Biology Activities and Events

15 - 19 January 2018

Metabolic Modelling Workshop 4,
Park Inn, Nottingham

[Read more](#)

25 - 26 January 2018

IBiolC18 – The 4th Annual
Conference, Glasgow
(Sponsored by C1net)

[Read more](#)

8 February 2018

North West Innovation (KTN) –
Unlock Your Business Growth and
Productivity, Daresbury

[Read more](#)

27 February 2018

Industry 4.0 – Innovation support
event run by Innovate UK (KTN),
Ipswich

[Read more](#)

27 March 2018

Innovation South Showcase - event
run by Innovate UK (KTN),
Farnborough

[Read more](#)

18 – 19 July 2018

'Beyond the lab: developing your
Industrial Biotechnology Career',
University of York – Derwent
College, Hendrix Hall

30 September 2018

Synbiobeta
San Francisco, USA

[Read More](#)

21-23 January 2019

C1net Final Conference, East
Midlands Conference Centre,
Nottingham

C1net Conference 3

5 - 7th November 2017, Nottingham

While the UK households stoked up their bonfires polluting the atmosphere, gas fermentation scientists descended on Nottingham to further a technology to help address carbon emissions as November 5th marked the start of a 2 day conference on "Gas Fermentation". Hosted by The University of Nottingham's based BBSRC-NIBB "C1net" and organised by network manager Jacque Minton, the event was held at the East Midlands Conference Centre, Nottingham.

"Great event. Nice giveaways. Nice introduction of awards of talks and posters. Great calibre of session hosts and speakers"

Formally starting with a Welcome dinner at the Orchard Hotel, there followed 2 days packed with talks, pitches and posters. With the aim of bringing together academic and industrial scientists to commercially exploit C1 gas organisms as platforms for chemical manufacture, the conference attracted 111 attendees, 21 of whom came from industry. Delegates were mainly from the UK, with 21 from Europe and 2 from the USA. A total of 24 talks were presented, 5 of which were invited, the rest were selected from abstracts, or were Proof of Concept (POC) reports.

Eminent scientist Rolf Thauer opened proceedings as chairman and there followed high calibre key note presentations from: Volker Müller (Goethe-University, Frankfurt), Linsey Garcia-Gonzalez (VITO NV), Sean Simpson (LanzaTech, USA), Christopher Brigham (University of Massachusetts, Dartmouth, USA), Guido Saracco (Istituto Italiano di Tecnologia, Italy) and Alexander Steinbüchel (University of Münster, Germany). Amid this star cast 6 PhD students also gained valuable oral presenting experience with Vera Salgrado (Nottingham) and Rupert Norman (Nottingham) carrying away glittering prizes of novelty mugs.

Additionally, 6 pitches were made in a fast-fired session to find partners for collaborative bids. A total of 36 posters were presented with a staggering 21 from PhD students. Carrying away prizes for the best posters were Anja Wiechmann (Frankfurt) and Rhiannon Chalmers-Brown (South Wales). Outreach was also on display thanks to Louise Dynes who showcased the new SBRC & C1net stand which was used at the New Scientist Live exhibition earlier this year.



New to the Team

Dr Ruth Griffin

I am delighted and honoured to have joined the SBRC in July. Thanks everyone for making me feel so welcome!

*To complement Professor Nigel Minton's area of Health Research, I will be applying my field of vaccine research to try and develop a prophylactic and therapeutic vaccine against *Clostridium difficile*. The vaccine, designed to target each stage of pathogenesis, will comprise conserved, surface-exposed lipoprotein antigens of *C. difficile* as well as domains of toxin A and B, integrated into a liposome delivery system.*

*My PhD student, Cansu Karyal, funded by the School Of Life Sciences is currently expressing putative lipoproteins of *C. difficile* in *E. coli* to compare their reactivity to patient sera IgG by ELISA. Those that are the most immunogenic will be synthetically lipidated and incorporated into liposomes by Chemist, Dr Nicholas Mitchell. We will then test the ability of the vaccine to protect from *C. difficile* infection in vivo.*

My third year PhD student, Ronni da Silva, funded by the Brazilian Foundation, CNPq transferred to Nottingham with me and will be continuing his project on meningococcal vaccines.



From left to right – Dr Ruth Griffin, PhD student; Ronni da Silva and PhD student; Cansu Karyal



Dr Minyeong Yoo

*I have recently joined the Synthetic Biology Research Centre as a postdoctoral researcher working with Prof Philippe Soucaille to participate in the project involved in synthetic Calvin cycle. My PhD study was focused on analysis of metabolic mutants of my old friend, *Clostridium acetobutylicum* by a global and quantitative systems biology approach. It was carried out at LISBP, INSA Toulouse, France as part of the CLOSTNET research programme. In my spare time, I try to meet my cat's requirements, for instance, opening of canned food. Also, drinking beer is a not to be missed pleasure for me.*

Other News

Viva Success!

A number of SBRC students have recently passed their vivas – Many congratulations to them!



Craig Woods: PhD thesis titled 'Forward Genetics in Clostridial Acetogens as a Route to Process Improvements'. Craig's PhD involved finding condition specific essential genes in *C. autoethanogenum* using TraDIS. Craig is now working as a research fellow at SBRC Nottingham continuing to work on TraDIS and *C. autoethanogenum*.



Christian Arenas: The title of my PhD is the genetic basis of 3-hydroxypropanoate metabolism in *Cupriavidus necator* H16. During this four years project I have been doing research for the production of 3-hydroxypropanoic acid. My plans for the near future are to work as a Postdoctoral Research Associate in a European project with a focus on the production of biodegradable plastics.

Grant Success

Dr Ruth Griffin has recently been award the following grants:

2017 SfAM New Lecturer grant (£10,000)
2017 SfAM President's Fund (£1200)
2017 Microbiology Society Grant (£750)
2017 Training Grant, University of Nottingham (£715)
2017 Travel Grant, University of Nottingham (£1400)

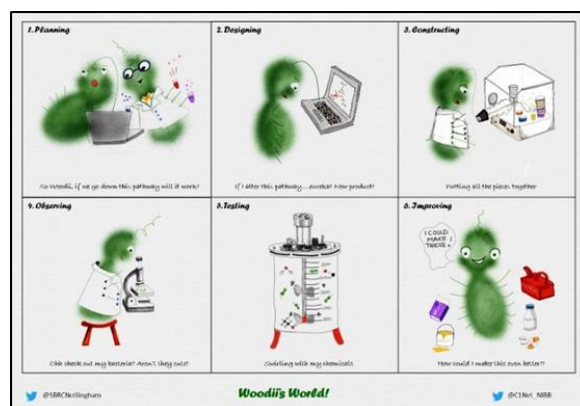
Outreach Activities

New Scientist Live! – London September 2017

New Scientist Live! Is a major science public engagement event at the Excel centre in London, which showcased a wide range of UK science innovations, with examples ranging from astronomy to microbiology. There was everything from VIP speakers such as Tim Peake to hands on demos, for example VR headsets that allowed you to experience the reality of a dementia patient.

I was lucky enough to be on the organising committee at this outreach event for the SBRC- Nottingham, which was funded both by SBRC – Nottingham and its associated network - C1net the Louise Dynes (SBRC Outreach Officer), Jacque Minton (C1net Manager), Shelly Kelly (SBRC Research Technician), Alan Burbidge (SBRC Centre Manager) and myself worked together to develop a fun and interactive stand, which would highlight the great research we do here at the SBRC- Nottingham. We designed a storyboard showing the various stages involved to go from concept to finished product and to help explain the science to everyone. We even had our own mascot Woody the Woodii, who helped walk people through each of the different steps.

We started with an interactive computer model of a biochemical pathway, developed by members of the SBRC computational team - Nicole Pearcy and Rupert Norman, which allowed people to decide how they would alter a pathway to produce the required end product. Next was a Don Whitley anaerobic cabinet that people could try, to give them an idea of what working in these cabinets was really like (especially as it was set to 37oC!).



Woody the woodii drawings by Penny Strong

With kids queuing up for this exhibit on the weekend, it was certainly a major draw to the stand. Then there was a microscope showing what these bacteria actually look like, followed by a mini bioreactor to help explain how this process could be scaled up. Finally we had some examples of potential products, such as a model tyre and fuel tank, to demonstrate where the C1 compounds could end up after the bacteria had converted them into useful chemicals.

At the event itself we were helped by many wonderful volunteers, who all demonstrated their enthusiasm and passion for science in their conversations, convincing possible sceptics about the benefits of our research. With over 30,000 visitors attending over the 4 day event, there was certainly plenty of opportunity to reach lots of people.

We had interest from a diverse range of backgrounds including; current scientists, potential future Nottingham students, children who we helped inspire with future possibilities and even artists and the media, who could help spread our research message even further. Overall I feel this was a great opportunity to highlight the wonderful cutting edge research of the SBRC – Nottingham and to educate and enter into dialogue with the general public.

***By Dr Pippa Strong, Research Fellow –
Synthetic Biology Research Centre***



Explaining the great research at the Synthetic Biology Research Centre



A visitor having a go at our anaerobic cabinet courtesy of Don Whitley Scientific Ltd



The Synthetic Biology Research Centre exhibition stand at New Scientist Live!



Prof Nigel Minton (SBRC Director) at the exhibition stand

Louise Dynes
Outreach and Communications Officer
Synthetic Biology Research Centre
Centre for Biomolecular Sciences Building
The University of Nottingham
University Park
Nottingham, NG7 2RD

