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## How next generation sequencing can fast track identification of mixed and complex samples.

**At NCIMB we identify thousands of bacterial and fungal isolates every year. Usually these arrive as pure cultures on plates, and we pick off colonies to identify the single species present.**

However, sometimes we are faced with more challenging samples, such as slime from inside a bioreactor, or samples collected from natural environments such as soil or sediments.

Within bioreactors, unwanted slimes are generally the result of microbial contamination. There could be one contaminant, or a complex mixed community which has formed a biofilm. Plating out this kind of slime to isolate individual species for identification can be a very time consuming, and ultimately frustrating, process. There is no guarantee that all the species present in mixed

environmental samples will grow quickly, or at all, under laboratory conditions. When faced with these kinds of samples, we now often suggest a 16S metagenomics approach because it provides a fast track to revealing the full make up of complex microbial communities by cutting out the requirement for plating and culturing. A 16S metagenomics approach can:

- Speed up the process by removing the time it would take to isolate and purify cultures from a range of media types/ growth conditions - the time saved could potentially amount to weeks.
- Ensure as many organisms as possible are identified including those with more complex growth requirements and non-viable organisms. Often it is the organisms with more complex growth requirements e.g. anaerobic or thermophilic organisms, that we are

sent by customers for identification, so it is important to also consider them.

16S metagenomics is based on sequencing sections of the same gene as the 16S Sanger sequencing we use for identification of bacterial isolates – the big difference is that the sequencing is undertaken in a massively parallel way. In other words, while with Sanger sequencing we sequence a single DNA fragment at a time, with NGS it is possible to sequence millions of fragments simultaneously in a single run. With our current Sanger sequencer, we could sequence just under 50,000 bases in 24 hours, whereas with our next generation sequencer, our smallest run would be 500,000,000 bases in 24 hours – in other words, our smallest NGS run in a 24-hour period would be 10,000 times what we could achieve with a Sanger sequencer in the same time.

This method can be applied to many different scenarios and gives unparalleled insight into a system under study. For complex samples it is likely to be a more cost-effective approach than isolating and identifying a large number of strains. It can also deliver results for complex samples much more quickly.

So what do the results of 16S metagenomics analysis actually look like? The results can be viewed as a simple bar chart, illustrated in *Figure 1*. For each sample there is a breakdown of the organisms present, and the relative

abundance of each organism in the sample. This can be very useful when comparing the diversity of different samples. For single complex samples a bar plot can also be useful as a way of visualising the organisms within the sample.

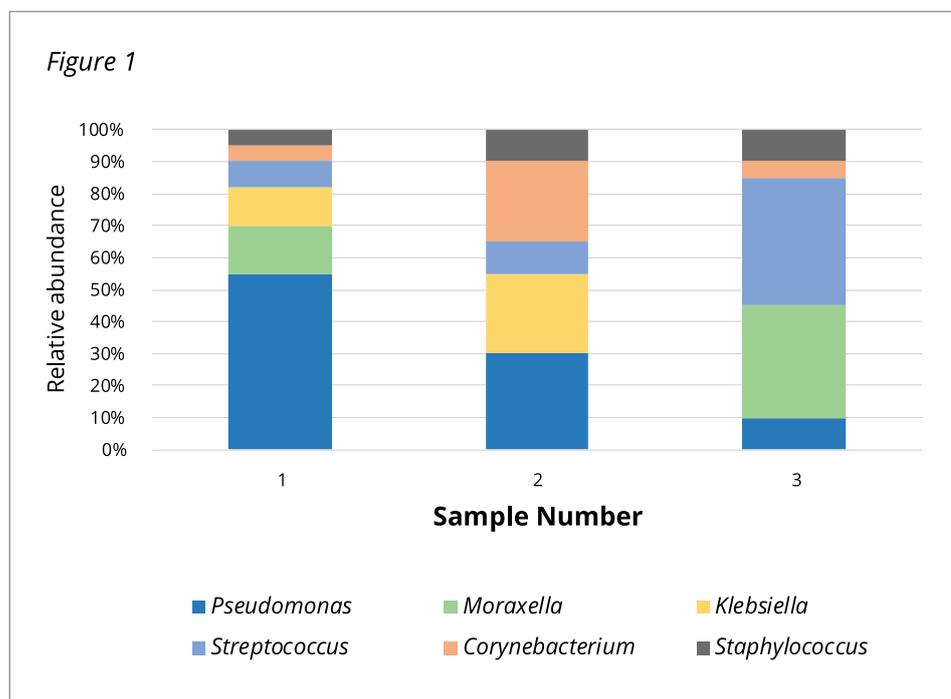
For example, in *Figure 1*, the chart shows that within sample 1, the genus *Pseudomonas* makes up the largest proportion of the sample (55%) followed by *Moraxella* (15%) then *Klebsiella* (12%), *Streptococcus* (8%) with *Corynebacterium* and *Staphylococcus* both at 5%.



#### ABOUT THE AUTHOR

Vikki Warren joined NCIMB in 2005. She leads the team of scientists responsible for delivering NCIMB's sequencing and identification services as well as sequencing additions to the National Collection of Industrial, Food and Marine Bacteria. Vikki has extensive experience of Sanger sequencing for identification of environmental isolates, and was central in the development NCIMB's microbial profiling and whole genome sequencing services, based on next generation sequencing with the Illumina platform. Vikki holds a BSc (Hons) degree in Applied Biosciences and Management, and an MSc in Instrumental Analytical Techniques; DNA Analysis, Proteomics and Metabolomics from the Robert Gordon University in Aberdeen.

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## NCIMB shares innovative MARA toxicity test experience with next generation of soil ecotoxicologists

NCIMB is sharing its experience of developing the innovative MARA toxicity tests with the next generation of soil ecotoxicologists through ARISTO: The European Industry – Academia Network for Revising and Advancing the Assessment of the Soil Microbial Toxicity of Pesticides.

This International Training Network is funded by the European Union's Horizon 2020 research and innovation programme

under Marie Skłodowska-Curie grant agreement.

The European Commission has a stringent pesticide regulatory scheme for pesticides authorisation, where risk assessment for aquatic organisms and terrestrial macro-organisms is well defined. In contrast, the assessment of the toxicity of pesticides on soil microorganisms is lagging behind. Microbial life is crucial for soil health, and the wider ecosystem, and

so the ARISTO project aims to address this gap. The program is focussed on key functional microbial groups such as ammonia oxidising microorganisms and arbuscular mycorrhizal fungi, as bioindicators of the toxicity of pesticides on soil microorganisms. In addition, issues such as toxicity of biopesticides, pesticide mixtures and pesticide transformation products will be explored as part of a holistic assessment of potential pesticide toxicity on soil microorganisms.

# NCIMB appoints Dr Edward Green as CEO

We are delighted to announce that Dr Edward Green has joined NCIMB as Chief Executive Officer. He takes over from Dr Carol Phillips who has retired after 12 years in the post. Dr Green joins NCIMB from CHAIN Biotechnology, a microbiome therapeutics company, which he founded and led since 2015. Prior to that he founded Green Biologics, a biotechnology company using bacteria to manufacture renewable chemicals.

Commenting on the appointment, outgoing CEO Carol Phillips said: "I am very pleased to be handing the reins over to somebody with so much experience in realising the potential that microorganisms have for industrial application, as this is at the

heart of NCIMB. What's more, as an NCIMB customer in his previous roles, Edward has a good understanding and appreciation of the high-quality services we strive to deliver for our clients.

"It has been a privilege to have been CEO of NCIMB, and although I will miss working with the wonderful team there, I'm looking forward to seeing how the company develops in the future".

Dr Green said: "This is a fantastic post for anyone with a passion for microbiology and its role in a cleaner, healthier and carbon-free future. There are so many issues that microorganisms have the potential to solve, and NCIMB is in a great position to contribute".



Dr Edward Green

# 40th Anniversary ushers in a new era for NCIMB Ltd with new purpose-built facility

NCIMB Ltd is the company that is home to the UK's National Collection of Industrial, Food and Marine Bacteria. It's unusual for a national culture collection to be held within a private company, but while our culture collection was first established as part of the UK Civil Service, it was transferred to Aberdeen University before being "spun out" as a limited company in 1982.

We've come a long way in 40 years, and have worked closely with different industry sectors, to develop products and services that meet their needs.

Today, NCIMB Ltd is a well-established, and highly customer-focused biotechnology company, providing innovative microbial solutions for life sciences, food & drink, energy, and environmental businesses. Our microbiology experts identify, characterise, quantify and store microorganisms to support and de-risk your process and product development.

As we turn 40, we are excited to be moving to a new purpose-built facility that will provide a fantastic foundation for continued growth and development. Of course, the culture collection still sits at the heart of our company, and continues to grow as researchers deposit new strains. We firmly believe it is an important genetic resource for the 21st century – a treasure trove of potential, available for research and development.



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## NCIMB 15251 *Lactobacillus gasseri* isolated from human breast milk

Recent additions to the National Collection of Industrial, Food and Marine Bacteria include a *Lactobacillus gasseri* strain. NCIMB 15251 *Lactobacillus gasseri* was isolated from human milk and it was deposited by Quadram Institute Bioscience.

This strain is reported to produce multiple bacteriocins, including a novel bacteriocin *gassericin M*. Bacteriocins are antimicrobial peptides produced by bacteria and archaea, and they have

been studied with respect to their role in the selection of probiotic products. The strain has been reported to exhibit antagonistic activity against different enteropathogens, and has been tested in a complex environment mimicking human colon gut conditions where it expressed seven bacteriocin genes. The authors of this study concluded that the strain was an interesting candidate for further study with respect to maintaining homeostasis in the gut environment.

You can read more about this research in the published paper:

Garcia-Gutierrez *et al.*,  
2020 Appl Microbiol Biotechnol.;  
104(9): 3869–3884.

For more information about purchasing any of our strains, or depositing your microbiome strains contact [enquiries@ncimb.com](mailto:enquiries@ncimb.com) or visit our website [www.ncimb.com](http://www.ncimb.com).

## Energy transition and Microbiologically Influenced Corrosion

NCIMB has been working with the oil and gas industry for many years, helping them to understand and monitor the risk of microbiologically influenced corrosion (MIC) in production facilities. As we transition our energy supply away from oil and gas, it is equally essential that corrosion challenges – including microbiologically influenced corrosion – are fully considered so that monitoring is effective and preventative action can be taken.

Our consultant microbiologist Dr Carol Devine attended the ICorr Aberdeen branch corrosion forum at the end of August. The theme of the event was energy transition and corrosion, and material challenges associated with the energy transition.

Topics covered included corrosion issues arising during hydrogen transport, and with offshore wind, fusion reactors and solar energy.

We understand so much more about MIC than was known in the early days of the oil and gas industry, and the techniques available to study microorganisms in the environment have developed and improved in recent years. This knowledge, and lessons learned during oil & gas production, are now available for the renewables industry. ICorr offers great opportunities to bring experts together to share their knowledge and expertise in this area and discuss key issues.



Dr Carol Devine

**Contact us** for more information on any of the stories in this edition of Collected Insights.



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