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microbiologist

The magazine of the
Society for Applied Microbiology



➤ **INSIDE**

**Plant
microbiology:**

Plant pathology
and *Pseudomonas*

To GM or not
to GM

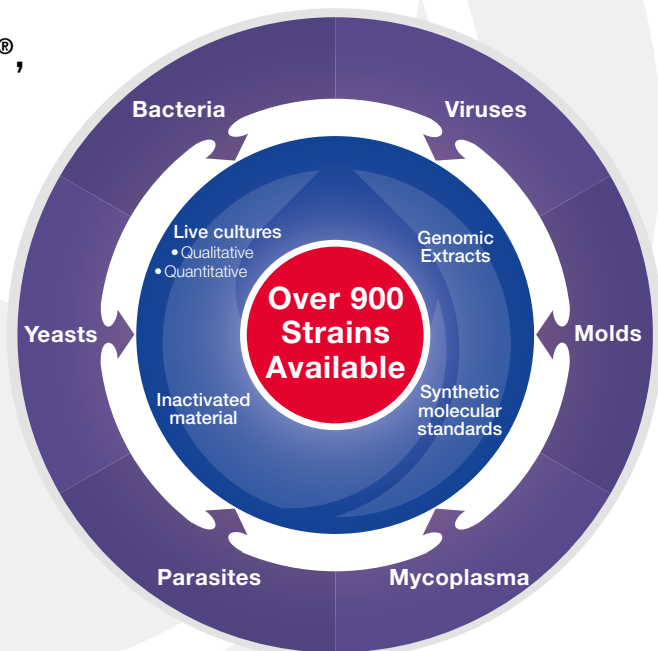
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Nancy Mendoza reviews the content of this issue

microbiologist

Plant microbiology: a niche and vital field The future depends on it...

The interactions between microbes and plants are not only fascinating, but critical to life itself. Whether dealing with diseases of crops, wild plants or trees, studying the symbiotic relationships between plants and nitrogen-fixing microbes, or exploiting the talents of agrobacteria to genetically transform plants, microbes are at the heart of ecology and food security, all over the world.

In this edition, the University of Reading's Rob Jackson describes some of the ways in which he has approached the problem of plant pathology caused by microbes, as well as microbes that are beneficial to plant growth or occur as contaminants. His work spans investigations at the molecular level through to some very interesting applications, such as the use of phage to control tree diseases.

Our second feature looks at the application of genetic modification to mitigate some of the threats to future food security. In particular, there is discussion of the work done in Norwich at The Sainsbury Laboratory where transgenic Desiree potato plants are able to resist the naturally circulating strains of *Phytophthora infestans*, which causes the devastating late blight disease. Many popular eating varieties of potato are highly sensitive to infection by this oomycete and losses to the UK farming economy are estimated at over £60 million annually. Without a new approach to controlling blight, this will only get worse as effective copper-based pesticides are phased out by the EU.

We couldn't let this issue go by without discussing teixobactin – the first member of a new class of antibiotics derived from soil bacteria. The discovery has been broadly celebrated, but there is still a good deal of work to do before we can say that the future of infection control is any brighter than before. Our third feature looks at some of the issues.

As usual, we bring you a Historical Perspective – this one is on 80 years of *Clostridium difficile* – and we highlight the career of one of our Members. We've got an article that covers the content of the *Environmental Microbiology* lecture, and a write-up of the PECs meeting, which was held on the same day. There is information about upcoming meetings, too, and we look forward to meeting many of you over the coming months.

We hope you enjoy this March 2015 edition of *Microbiologist* and please do get in touch with any feedback (communications@sfam.org.uk).

NEWS IN BRIEF

Safer GM

Scientists have created GM bacteria that are unable to survive in the wild.

http://bit.ly/GM_bacteria

Ebola vaccine

Ebola vaccine trials are beginning in West African populations.

http://bit.ly/Ebola_vaccine

Possible new antibiotic for Gram -ve infections

Mitrecin A from a new streptomycete has activity against resistant Gram-negative bacteria.

<http://bit.ly/MitrecinA>

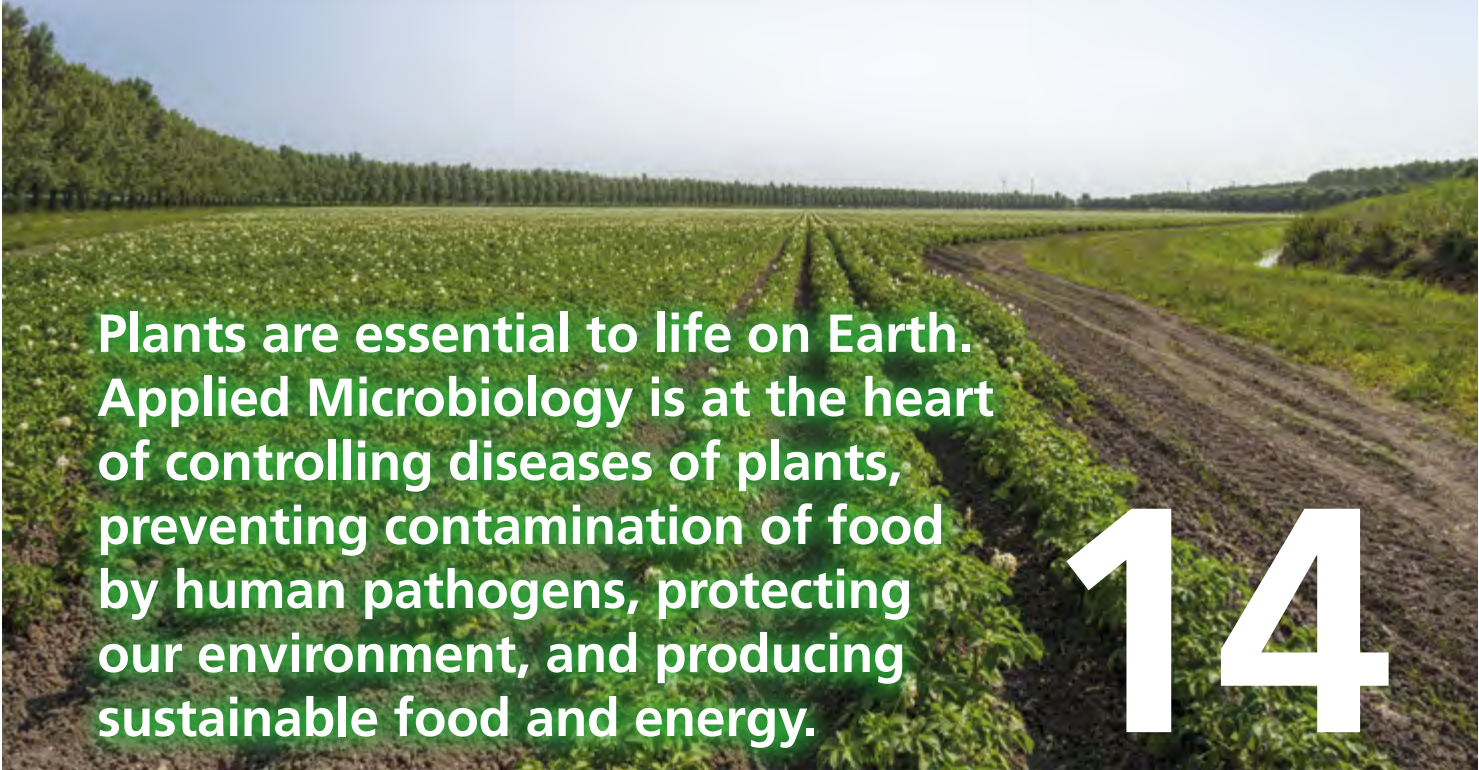
AMR could kill millions

Millions will die each year if we do nothing about the rising tide of antimicrobial resistance.

<http://bit.ly/ONeill1>



Nancy Mendoza, Editor



Plants are essential to life on Earth. Applied Microbiology is at the heart of controlling diseases of plants, preventing contamination of food by human pathogens, protecting our environment, and producing sustainable food and energy.

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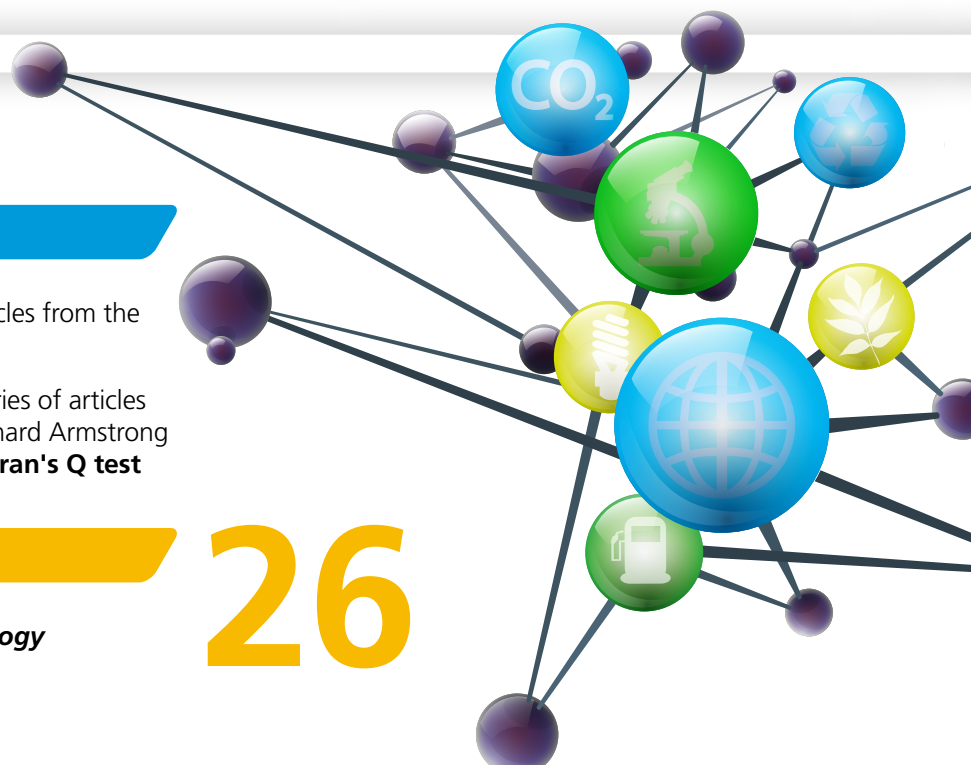
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President's column



The Broad Street Pump: just the beginning

This year is a point of change for the Society as we have said farewell to Phil Wheat, who as you will know from our last issue, retired as our Chief Executive at the end of December. This is my first opportunity through this column to thank Phil for his many contributions to the Society and to welcome Lucy Harper into her new role as Chief Executive.

As part of our farewell to Phil last December, we held a reception at the John Snow pub in London. Dr John Snow is of course widely recognized as one of the founding fathers of epidemiology and developed the theory of transmission of cholera via water. He

famously convinced the authorities to remove the handle from the Broad Street public water pump which he considered was the source of an outbreak of cholera in the Soho district of London in 1854. The site of this pump is marked by a pink granite slab in the pavement immediately outside the pub and was a subject of conversation at the event.

Visiting the site led me to wonder how John Snow would have viewed our progress in improving public health 160 years later. There is no question that he would see the development of sewerage systems and treated water supplies as legacies of his original ideas

Visiting the site led me to wonder
how John Snow would have viewed
our progress in **improving public
health 160 years later**

In our recent policy workshops, run jointly with the Society for General Microbiology, provision of a safe water supply was one of the global 'Grand Challenges' under consideration

which have contributed very significantly to the reduction of diseases like cholera and other diseases transmitted by the faecal-oral route in those countries where they exist. However, the WHO estimates that globally there are still 3–5 million cholera cases resulting in 100,000–120,000 deaths every year in developing world populations with no proper access to adequate water and sanitation resources.



John Snow memorial and pub on Broadwick Street, London.
Image: Justin Cormack CC BY-SA 2.0

In our recent policy workshops, run jointly with the Society for General Microbiology, provision of a safe water supply was one of the global 'Grand Challenges' under consideration. Whilst it could be argued this is no longer a microbiology issue but a technical infrastructure one – the microbiology issues are clear and solvable – it is certainly a cause which, as applied microbiologists, we should take every opportunity to support because of the very obvious and real benefits to public health and quality of life it provides.

It's also true that we cannot afford to be complacent in developed countries – it was about a year ago that the UK was hit by major flooding and our General Secretary, Professor Mark Fielder, was involved in providing advice to the public. He spoke to the media about pathogen contamination risks associated with urban floodwaters and advised on simple measures to prevent disease, such as handwashing. Our Winter Meeting this year '*Water, water everywhere but is it safe?*' also picked up on this issue and others involving waterborne pathogens (a full report will be available in the next edition of *Microbiologist*).

So, I think John Snow would see that his ideas on disease transmission have led to a very significant improvement in the lives of a great many people, but might reflect that there is still a great deal of work to be done.



Christine Dodd
President of the Society

Harper's Postulates:

Notes from the Chief Executive

At the end of 2014, the first report from the O'Neill Review was published, spelling out the health and economic impact of antimicrobial resistance (AMR). The second report, published last month, outlined specific actions needed to tackle this global threat. At the time of commissioning, David Cameron was quoted saying: *"If we fail to act, we are looking at an almost unthinkable scenario where antibiotics no longer work and we are cast back into the dark ages of medicine"*.

Although this does sound dramatic, the message remains: a continued global effort is needed to tackle AMR.

In the UK alone, there is now increasing activity involving scientists, clinicians, economists, politicians and the media who are all working to raise awareness and find solutions to this growing problem. The Longitude Prize is probably the highest profile from the point of view of raising awareness in the eyes of the general public. There are campaigns within the veterinary arena, and in general practice and acute care settings, promoting the stewardship of antibiotics in veterinary and human healthcare. Globally, microbiologists are looking for faster diagnostic tools

LeSPAR comprises:

- > **Biochemical Society**
- > **British Society for Antimicrobial Chemotherapy**
- > **British Pharmacological Society**
- > **Royal Society of Chemistry**
- > **Society for Applied Microbiology**
- > **Society for General Microbiology**
- > **Society of Biology**

and new antimicrobials, and they are meeting with increasing frequency to discuss this urgent issue.

I have anecdotal evidence from non-scientist family and friends demonstrating the effectiveness of the Longitude Prize in raising awareness of the ongoing threat of AMR. And in the US, at the time of writing this piece, a potential new class of antibiotic has been found to be effective against pathogens in mice, and early indications show it may be slow to develop resistance. Although in the early stages of development, there is hope that this compound will become an effective antibiotic drug against human pathogens (more on pages 18–19). So, although with caution, I think we can offer a glimmer of hope.

But the effort must continue and to that end SfAM is working with six other Learned Societies to enhance understanding and knowledge sharing on AMR between academia, industry and clinicians through the LeSPAR network.

This network represents 75,000 scientists who have come together to lead the fight against AMR. The network aims to provide a single unified voice, take action and champion best practice, as well as raising awareness of the global challenge of AMR. Visit the SfAM website for the latest LeSPAR developments (www.sfam.org.uk/LeSPAR).

As I write, we have just embarked upon a Strategy Away Day and we are in the process of setting the Society's aims and objectives for the future. Officials of the Society met for an intensive day-long event to really get to the bottom of who we are, who we want to be, what we want to be doing and how we want to be doing it.

We'll be publishing our strategy in the coming months, so keep in touch to find out more about our priorities for the future. But of course AMR will remain a priority area for SfAM and we plan to make a contribution to this global challenge facing humanity.



Lucy Harper

SfAM Chief Executive

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Plant pathology and *Pseudomonas*

Introduction

It's 5.30 am sometime in 1996 and I can sleep no more. I received confirmation the day before that I've found a virulence factor on a plasmid in the plant pathogenic bacterium *Pseudomonas syringae* that is essential for pathogenicity – and it suppresses the detection of "avirulence" genes by the plant host. I'm so excited about my PhD "breakthrough" that I want to start writing the paper. I didn't really understand the scope of things back then, so it was three years and many experiments later that the work was finally published (Jackson *et al.*, 1999).

Ten years on from that early morning paper writing, I started my Microbiology Lectureship in Reading and over the last eight years I have built up a research group. Things have moved on considerably though; global food security is a key strategic problem, research impact is now a major component of both RCUK-funded research projects and the Research Excellence Framework assessment. And despite Plant Pathology

being recognized as a key skill area, it is dangerously under threat from loss of investment both at the educational level, for teaching and training younger scientists, and from the reduction in available research funding. But I see these challenges as opportunities, both to fill key research niches and to use outreach as a tool for education and impact.

Beneficial *Pseudomonas* to promote plant growth

As much as basic fundamental biology is crucial for knowledge and idea generation, I have really come to appreciate applied science more and more. My focus remains on bacteria-plant interactions, but includes pathogens, beneficials and contaminants, both working out mechanisms at the molecular level, but also exploring applications. For example, we study the plant growth-promoting rhizobacterium (PGPR), *Pseudomonas fluorescens*.

PGPR are used as microbial inoculants, designed to promote plant growth through the provision of nutrients or growth factors, or by protection from pathogens. However, PGPR are known to have frailties, especially regarding mode of action (different mechanisms, variable expression) and robustness (their beneficial effect can diminish within days of application). This reflects our poor understanding of how bacteria live in soil and around plants, and how the environment influences them. So, to make the most of these natural resources, we need a thorough understanding of how the bacteria function.

We have been studying bacterial motility, a crucial trait required by bacteria to move around soil and roots to gain nutrients – and in turn providing maximal bacterial spread and conferment of benefits.

We found that a well-known biosurfactant, viscosin, aids *Ps. fluorescens* spreading over surfaces, including plant roots. Viscosin also antagonizes root pathogens, such as *Pythium* (Alsohim *et al.*, 2014). There is, therefore, an opportunity to examine how the viscosin system could be exploited to promote plant colonization and disease protection.





Rob Jackson, University of Reading

There is, therefore, an opportunity to examine how the viscosin system could be exploited to promote plant colonization and disease protection

FEATURES

Understanding bacterial evolution and gene regulation

More recent work with the motility system described earlier has found that immotile bacteria that cannot activate flagellum genes can evolve flagellum-based motility.

Loss of motility is a potentially catastrophic situation for the bacterium as it cannot forage for nutrients and if this happened in the wild then the strain may become extinct (or, if used as an inoculant, we see reduced or complete loss of inoculant efficacy). However, starvation selection pressure leads to the recruitment of a mutated nitrogen regulatory system to activate the flagellum system and rescue motility. This insight is important because we now know the bacterium can rapidly adapt to mutations which affect both function and robustness – the question now though is whether loss and gain of function readily happens in the natural environment in a growing season. We thus need to examine the basis of bacterial adaptations to changes in their genome and environment.

Using *Pseudomonas* to control aphid pests

One of my new research themes uses bacteria for the control of aphids, which are major horticultural pests. There are very few control products available to growers, but a few publications over the last 10 years have described some plant-associated bacteria, like

Dickeya, *Erwinia* and *Pseudomonas*, that can kill aphids (Grenier *et al.*, 2006). The mode of action seems to vary from gut occlusion preventing food digestion to production of specific insecticidal toxins. Three of my PhD students have taken a broader approach to examining the scope of this phenomenon by isolating a range of plant-associated bacteria that can kill aphids.

We found several different genera can kill aphids when ingested, but almost fortuitously, two of the best killers are species of *Pseudomonas*.


In collaboration with Chris Bass and Tim Mauchline (Rothamsted Research) we are using a combination of genomics, genetics and RNASeq to examine how the aphid responds to infection, and to also help identify bacterial genes expressed within the aphid. By analysing these, we can try to identify the mode of action the bacteria use to kill the aphid and the likelihood of resistance occurring in the aphid – for example, some pesticide resistance occurs through up-regulation of detoxification genes.

Importantly, we are also trying to determine whether the bacteria harm beneficial insects. Some trials with collaborators in Tunisia suggest there is no impact on bee larvae, but new work with evolutionary biologists and ecologists Mark Fellowes, Alice Mauchline and Louise Johnson at Reading aims to follow up on this.

Developing phage therapy as a rapid, deployable treatment for controlling epidemic tree disease

Another of my new research areas relates to developing phage cocktails to treat tree diseases.

Several members of my group work on Horse Chestnut bleeding canker and Acute Oak Decline. It has been



We found several different genera can kill aphids when ingested, but almost fortuitously, two of the best killers are species of *Pseudomonas*

illuminating to me just how tricky it can be working on a tree system, especially for infection assays. One of the things that struck me is just how slow the research response can be to trying to save our trees – in the midst of several tree epidemics, we need to be urgently considering ideas that can provide rapid treatment before the population collapses. Thus, in collaboration with Ian Jones (Reading) and Mike Brockhurst (York), we have been studying phage therapy as a possible treatment for Horse Chestnut bleeding canker disease caused by *Ps. syringae*.

Phage therapy is an alternative antimicrobial treatment to antibiotics to help clear bacterial infections (Gross, 2014). One of the benefits of using phage is the potential to use mixtures, or cocktails, of different phages on infections to reduce the chance of host resistance occurring.

We have collected several phage from Horse Chestnut and characterized them. We have been evolving phages to create new genotypes and then trialling cocktails.

Our initial observations found that a two-phage cocktail can prevent emergence of *Ps. syringae* resistance *in vitro* and also reduced disease *in vivo*. Of course, there is a requirement to understand the efficacy of phage and that the genes they carry do not pose a threat to the wider environment, so we are working towards addressing these challenges.

Microbiology and Plant Pathology Outreach and Impact

Parallel to this research work is the need to raise the profile of applied microbiology and plant pathology, by improving education and informing stakeholders. I got the bit between my teeth first as an Admissions Tutor at the University of Reading, highlighting microbiology to visitors on open days. Then I helped the British Society

for Plant Pathology to employ an outreach officer to start grass-roots outreach.

More recently, I have been working with the rapper Baba Brinkman to develop a music video for the song 'So Infectious'. This project was funded by SfAM and most of the supporting filming was done in Reading with biological science undergrads, postgrads and postdocs. The film is completed and will be available to all SfAM Members for their teaching and open days.

Of course, research impact is important these days and I am hoping these experiences, combined with my research programme and involvement with REF, will result in useful outputs for society.

Eighteen years on from that early morning paper-writing period, I still have the excitement of achieving the breakthroughs, but I especially love the academic freedom to work on different projects and with the many talented people I collaborate with in my university and research group, and elsewhere.

I thank SfAM for the invitation to write this article and for supporting the making of the rap video, and thanks to all my colleagues, young and old, for making plant pathology and microbiology such an exciting subject to work on.

FURTHER READING

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Rob Jackson
University of Reading



To GM, or not to GM, that is the question!

The current losses of food crops to disease are unacceptable and so applied microbiology has an opportunity to make a substantial difference to our future health and wellbeing. This is especially true for late blight – a disease of potatoes and tomatoes that is caused by *Phytophthora infestans* (an oomycete).

Potatoes are a key staple in many countries. Amongst starchy foods they are relatively nutrient rich and highly productive compared with cereals (though less water efficient). However, yield increases have flattened and losses to disease are significant; the cost to farmers of late blight is estimated at £60M a year in the UK alone.

Phytophthora infestans evolves remarkably rapidly, making the disease a particularly intractable problem; emerging strains have been able to overcome the resistance to infection seen previously in some potato varieties.

Plants that are susceptible to infection can be treated with fungicides, but those that are effective carry high environmental risks. The popular Bordeaux Mixture – a copper-based fungicide used to prevent late blight – is in the process of being phased out by the EU, along with other copper formulations.



With the global population set to reach nine billion by 2050 and a changing environment, we were (and still are) facing a huge challenge to be able to produce enough safe, nutritious, sustainable food for everyone

So, at present the best course of action a grower can take is to grow varieties that are relatively resistant (which unfortunately doesn't include some of our old favourites, such as the King Edward) and to use weather forecasting and other techniques to minimize the chances of an epidemic in the crop.

A case to act

Towards the end of 2008, the then Chief Scientific Advisor, John Beddington, warned that the world faced a "perfect storm" of threats to food security.

With the global population set to reach nine billion by 2050 and a changing environment, we were (and still are) facing a huge challenge to be able to produce enough safe, nutritious, sustainable food for everyone.

Already, the world has experienced rapidly rising food prices; the prices of wheat, coarse grains, rice and oilseed crops almost doubled in 2–3 years to the end of 2007. The cause of this spike was a combination of multiple, mutually reinforcing factors, including droughts in key grain-producing areas, increased use of food crops for biofuel production and rapidly rising oil prices. This made the issue all too real in the pockets of consumers.

With the current rates of improvement in production yield, it was unlikely that the numbers would ever work out. So what could be done?

To GM or not to GM?

A year after Beddington's "perfect storm", the Royal Society published their "Reaping the Benefits" report, chaired by renowned plant scientist, Professor Sir David Baulcombe. The report looked at the potential for biological sciences to contribute to increased yield and efficiency of food crop production. And so, after some years of quiet, the issue of GM crops was raised once more.

In the report's summary, it is stated that "*Genetic improvements to crops can occur through breeding or genetic modification to introduce a range of desirable traits...by increasing photosynthetic efficiency, reducing the need for nitrogen or other fertilizers and unlocking some of the unrealized potential of crop genomes.*" Later on, the authors hint at the relative velocity of genetic modification versus traditional plant breeding, which typically takes 10 years or more for a breeding cycle.

At the same time, then Secretary of State for the Environment, Food, and Rural Affairs, Hilary Benn was speaking in support of investigations to assess the potential of GM, which he said could bring about varieties that are able to fix nitrogen, or grow with less water or other inputs. Benn and others in the public sector were careful to place GM as just one tool in a toolbox, perhaps partly for fear of scaring the horses, but also because the development of GM varieties is not easy, nor is it cheap, and it is not the best solution for all problems of sustainable crop production.

A few brave scientists pushed on into field trials of GM crops during the subsequent years – some notable examples in the UK were non-commercial trials, funded by the Biotechnology and Biological Sciences Research Council (BBSRC), which stumped up the extra cash to introduce security to prevent protestors from entering trial sites and damaging the crop. One such trial took place at the Norwich Research Park, where scientists from The Sainsbury Laboratory, led by Professor Jonathan Jones, grew a GM Desiree variety, alongside non-transgenic Desiree plants.

The field trial ran from 2009–2012 and in the third year the warm and wet conditions created the perfect environment for late blight disease to arrive of its own accord (there was no inoculation).

By early August 2012, 100% of the non-transgenic plants were infected but the GM plants were fully resistant. The yield was about 5kg greater per 16 GM plants as compared with the non-transgenic variety.

Many opponents of the trial argued that blight-resistant varieties had been traditionally bred already, making the use of GM to tackle blight resistance redundant. But traditional breeders have not, after many years, been able to introduce resistance into popular eating varieties. In fact, it has been said that the most successful of the blight-resistant plants actually produces potatoes that are fairly unpleasant in texture and flavour.

It might not be too bold to suggest that here we have an example where GM is ideally suited to tackle a problem that threatens food security and the sustainability of the farming industry.

But just because it works, and just because preventing losses from blight is a good thing to do all round, doesn't mean that we will see Professor Jones' blight-resistant Desiree growing in UK fields any time soon.

Regulation in Europe – the precautionary principle

The House of Commons Science and Technology Committee is currently running an inquiry into GM foods and their regulation under the precautionary principle at European level.

In some areas of EU lawmaking, the precautionary principle is actually a statutory requirement. And in the case of GM foods this has led to regulations restricting the growth of GM foods in the UK and across Europe. This is not the case in the USA where GM crops have been grown for many years.

Andrew Miller MP, Chair of the Science and Technology Committee said: *"GM technology potentially offers an array of benefits, but concerns are being expressed that it is being held back by misuse of the precautionary principle."*

By early August 2012, 100% of the non-transgenic plants were infected but the GM plants were fully resistant. The yield was about 5kg greater per 16 GM plants as compared with the non-transgenic variety

"In this inquiry we will be looking at whether such restrictions are hampering UK scientific competitiveness and whether they are still appropriate in light of available evidence on the safety of GM."

The precautionary principle relies on an absence of scientific consensus in relation to an action or policy that has a suspected risk of causing harm to the public or environment. The current inquiry will need to assess whether there is indeed a scientific consensus that GM has a low risk of causing harm. At the time of writing, the final session of the inquiry has just taken place and a report is awaited.

Professor Jones, himself, has said that the precautionary principle should never be used without a 'post-cautionary principle'. That is to say that at a defined time point, use of the precautionary principle can be reviewed and if the caution is deemed to be excessive, it can be revoked.

Genome editing

While the issue of GM food continues to be debated at the highest levels, techniques for crop improvement are being developed and enhanced, posing further challenges in policymaking.

In October 2014, the BBSRC published a position statement (<http://bit.ly/BBGenEd>) on new and emerging techniques, which focused largely on what has been termed 'genome editing'. That is, the ability to make precise genetic changes in plants either by adding, removing or replacing DNA at specific locations, or by switching genes on or off without actually changing the DNA sequence.

These techniques are already commonly used in research and could potentially have applications in new commercial crops. For that to happen, the regulatory

process will have to accommodate these new technical developments as well as the now well-established GM protocols.

BBSRC Chief Executive, Professor Jackie Hunter, said: *"We must take advantage of the wide range of techniques available in order to use the right approach for the right circumstance, such as conventional breeding, genetic modification or newer methods like genome editing."*

"A [regulatory] system based on the crop characteristics, by whatever method it has been produced, would provide more effective and robust regulation than current EU processes."

Conclusion

Historically, it has been difficult to make a successful case for GM crops to the public and to policymakers. And that isn't so much because of a lack of evidence that the risks are low – there have now been many studies to assess the risks to health and the environment – but rather the refusal to accept the early examples that GM crops could be used for a greater good, rather than simply to improve the profit margin in crop production. Such examples include the highly popular GM tomato puree sold by Sainsbury's and Safeway, which was eventually removed from the shelves due to protests; and nutritionally enhanced 'golden rice'.

These latest developments point to an important role for GM crops in mitigating the risk of a food security crisis, which would certainly fit the criteria for providing public benefit. And it is therefore timely and appropriate for the policy process in Europe to be reviewed.

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Nancy Mendoza

Society for Applied Microbiology

Teixobactin: gold rush or muddy bank holiday?

The iChip in use

Image: Slava Epstein

The issue of antimicrobial resistance has been covered regularly in *Microbiologist* and we couldn't let this issue go by without mention of teixobactin, which, if it lives up to its promise, will be the first of a new class of antibiotics against Gram-positive organisms. News of the discovery of teixobactin, derived from soil bacteria, was reported in *Nature* on 7 January 2015.

We haven't seen a new class of antibiotics for almost three decades, and for certain diseases we are now well beyond scraping the bottom of the barrel for drugs that pathogenic microorganisms are still sensitive to. With this rising tide of resistance and the paucity of drug reinforcements, there are now common multi-drug resistant strains of key human and animal diseases, including gonorrhoea and tuberculosis, and infections involving ESBL or 'extended-spectrum beta-lactamase' resistance are widespread. We are, as UK Prime Minister David Cameron has said, facing the prospect that medicine may be "cast back into the dark ages" unless action is taken. The hope is that teixobactin will, in the end, be regarded as a 'Critically-Important Antibiotic', as defined by the WHO, and its use would be reserved for the treatment of life-threatening conditions.

Microbes themselves are, of course, a rich source of new drug candidates in the search for new antimicrobials. *SfAM* has previously reported on some of the unusual places where microbiologists are hunting for these molecules – the driest deserts, the deepest oceans and even the outer cuticle of the industrious leafcutter ant. But these examples of 'bioprospecting', though turning up the occasional nugget of a possibility, have hardly created the proverbial gold rush; the question is, will teixobactin be the one to do it?

**The hope is that
teixobactin will,
in the end, be
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'Critically-Important
Antibiotic'**

Teixobactin, though still far from the clinic, is tantalizing and could be the start of something really big. It has shown promise in mouse trials where Gram-positive infections, *Staphylococcus aureus* and *Streptococcus pneumoniae*, responded well to treatment with the drug, and there were no obvious signs of toxicity. There is also the potential that resistance to teixobactin will be slow to develop, given that it binds to lipid cell-wall precursors, just as vancomycin before it. It took 30 years of clinical use before vancomycin showed resistance, and that was probably due to horizontal transfer of self-resistance genes, which the teixobactin-producing bacteria show no signs of having in the first place. But perhaps the most exciting part of the teixobactin story, for future bioprospectors, is that a whole new method of growing soil bacteria in the lab was developed for the purpose of the study, perhaps opening the field to many new discoveries.

The 2014 *Environmental Microbiology* lecture by Professor Jim Prosser (pp26–27) dealt with some of the challenges of characterizing populations of soil bacteria, which are notoriously difficult to culture under laboratory conditions. Professor Prosser described how he turned to molecular techniques to get a snapshot of the microbial communities under our feet, but in this study the team from Northeastern University, Boston, USA, created a “subterranean hotel” for the soil bacteria to populate.

Professor Kim Lewis’ team at Northeastern created the iChip, comprising an array of ‘rooms’, and into each room, a single soil bacterium was placed. The entire chip was then buried in soil, thus providing as close to natural chemistry as possible. They were then able to test the molecules produced in each individual room, for their potential as antimicrobials.

It is estimated that this new method could enable culture studies of up to 50% of all soil bacteria, which is a massive leap on from the current ~1% that can be grown in the lab. It is of course worth noting that, despite the difficulties of culturing soil bacteria, they have been the source of many of our current antibiotics. It is perhaps unsurprising that Professor Lewis reports 25 new antibiotics discovered using this method, with teixobactin being the most promising, currently. Of course, the hope is that other antibiotics discovered in this way will be effective against important Gram-negative bacteria such as *E.coli*, *Pseudomonas* and *Klebsiella pneumoniae*, as well.

So, teixobactin is the first member of a new class of lipid II binding antibiotics and it is very likely that Gram-positive pathogens would be very slow to develop resistance to this naturally rare molecule. Early tests in mammals are promising and, of course, the next step would be to test for safety and efficacy in humans. This is all very well, but there remains the question of how



Teixobactin is produced by a new species, which has been provisionally named Eleftheria terrae.

such a molecule could be synthesized on a large scale, given that its natural producer is both rare and difficult to culture. And what would the business plan look like for its development, marketing and sales?

The current commercial involvement in this discovery is from privately held, early stage biotechnology company, NovoBiotic Pharmaceuticals. NovoBiotic is a Northeastern University spin out from Kim Lewis and colleague Slava Epstein. The company rests on the iChip technology and its ability to generate new drug candidate molecules for investigation.

The team has investigated the biochemistry of teixobactin, describing its structure, and predicting a biosynthetic gene cluster. Teixobactin is produced by a new species of β -proteobacteria, which has been provisionally named *Eleftheria terrae*. *E. terrae* has been characterized via genome sequencing, 16S rDNA and *in silico* DNA/DNA hybridization, and is thought to be from a new genus, related to *Aquabacterium*. The team has identified two non-ribosomal peptide synthetase genes, *txo1* and *txo2*, which are predicted to encode distinct catalytic domains involved in the synthesis of teixobactin, which is an unusual depsipeptide containing enduracididine, methylphenylalanine and four D-amino acids.

So, the groundwork has been laid, and we shall have to watch with interest for one of the big hitters stepping up to the plate to fund future trials and product development. Time will also tell whether the iChip technology is the breakthrough tool for bioprospecting in soil. The gold rush might yet begin...

Nancy Mendoza

Society for Applied Microbiology

HISTORICAL PERSPECTIVES

Introduction

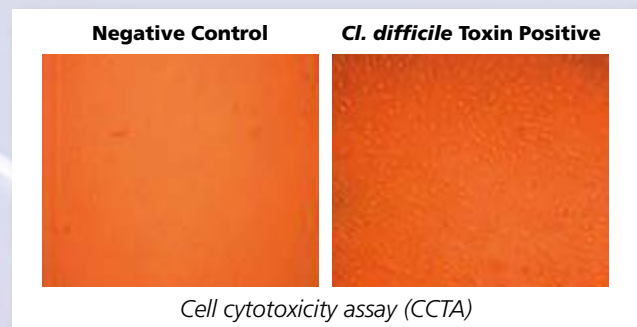
Clostridium difficile was originally recovered from the faeces of newborn infants by Hall and O'Toole in 1935. A hamster model proved essential in the early work where the toxin of *Cl. difficile* was shown to be the agent of disease. The main contributors to this work included Keighley and Larsson *et al.* in the UK, and Bartlett and Lusk *et al.* in the USA.

In 1977, Bartlett and his co-workers showed that a cell cytotoxicity assay (CCTA) used to detect toxin in the faeces of affected patients was a useful diagnostic laboratory tool to establish the presence of disease.

The most common clinical presentation of *Clostridium difficile* infection (CDI) is diarrhoea associated with antibiotic use. Diarrhoea can occur within a few days of starting therapy but can equally occur up to eight weeks after therapy has ended. Patients with mild to moderate disease usually experience diarrhoea as the only symptom with up to 10 bowel movements a day. More severe disease includes abdominal pain or cramps, fever and leucocytosis. Watery faeces with a foul odour is characteristic, however, the presence of blood is rare. Severely diseased patients also demonstrate hypoalbuminaemia and altered creatinine and lactate levels.

Rarely, diarrhoea may be absent in severely ill patients who suffer with a complication known as paralytic ileus. Any patient with unexplained fever, high white blood cell count and abdominal pain with recent antibiotic exposure should be investigated for the presence of CDI.

Elderly asymptomatic patients may represent a particular problem since many are resident in long-term care facilities where *Cl. difficile* may become endemic. Such carriers may harbour large numbers of the organism in their faeces and act as a rich reservoir of the organism.



Evolution of the laboratory diagnosis of CDI

Once it was recognized that the disease was toxin-mediated, the CCTA was recommended as the diagnostic test. Although specific for the disease, it is labour-intensive and may take overnight (and sometimes 48 hours) incubation before a positive test is recognized. Research was undertaken to find a selective culture medium for recovery of the organism from faeces and this was eventually published by George *et al.* in 1974.

Although more sensitive than CCTA, recovery of the organism by culture took 48 hours. Furthermore, the specificity of culture was found to be low because some patients carry the organism without any signs of disease and because later research showed that approximately 15–20% of *Cl. difficile* strains are non-toxicogenic.

With the realization that toxin testing is important in the diagnosis of CDI, together with the increasing unavailability of tissue culture, many laboratories called for easier methods of toxin detection. Commercially available toxin tests based on the use of ELISA technology became available as a rapid and cheaper method for toxin detection. Most laboratories rapidly moved over to using a commercially available ELISA platform or a later development, a 'flow-through' device based on immunochromatography.

Clostridium difficile: The last 80 years

It soon became obvious that the commercial ELISA kits for detection of faecal toxin, although rapid, were considerably less sensitive than the cell assay resulting in the misdiagnosis of some patients.

Department of Health Commissioned Study

A four-centre study was commissioned by the Department of Health to validate the reference methods according to clinical outcomes and to derive an optimum laboratory diagnostic algorithm for CDI.

A total of 12,441 diarrhoeal faeces samples (submitted for investigation of suspected CDI) were tested using five methods: CCTA, Toxigenic culture, GDH EIA, Toxin EIA and RT-PCR.

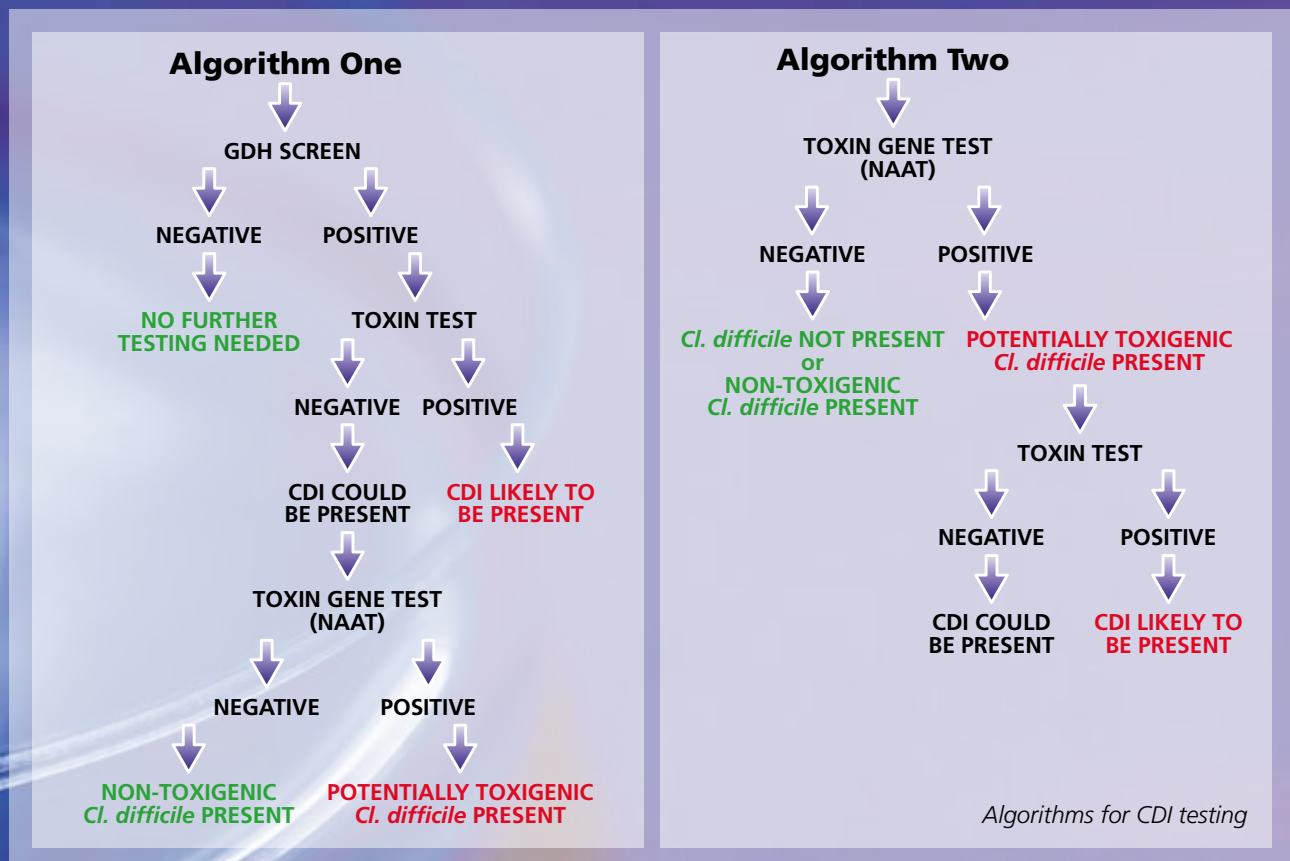
The following patient investigations were also conducted:

- **Diarrhoea (Bristol Stool Chart)**
- **Fever**
- **Abdominal cramps**
- **Evidence of colitis or pseudomembranous colitis (PMC)**
- **30-day mortality-associated and morbidity-associated laboratory measurements**
- **White blood cell count**
- **Serum albumin**
- **Serum lactate**
- **Serum creatinine**
- **Faecal lactoferrin**

Clostridium difficile was originally recovered from the faeces of newborn infants by Hall and O'Toole in 1935

The results showed that the detection of faecal toxin correlates most accurately with disease and should be regarded as the gold standard for the diagnosis of CDI.

The confidence in detecting patients with true CDI was shown to be increased by screening patients with a test for the glutamate dehydrogenase (GDH) enzyme followed by a sensitive toxin test for GDH-positive samples. Similarly, screening with a nucleic acid amplification test (NAAT) would achieve the same outcome with a faster turnaround time but is often cost prohibitive.



Once it was recognized that the disease was toxin-mediated, the cell cytotoxicity assay (CCTA) was recommended as the diagnostic test

The role of NAATs was shown to be limited to the detection of toxigenic strains in GDH-positive patients who were faecal toxin-negative. This may be useful for infection control purposes where carriers of toxigenic strains can be identified. Similarly, a negative NAAT result would rule out the likelihood of the patient having CDI and potentially negate the need for infection control measures.

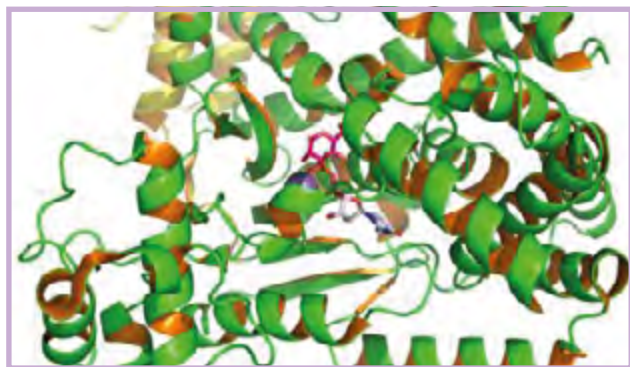
Glutamate dehydrogenase

GDH, also known as the common antigen, is constitutively produced by all strains of *Cl. difficile* independently of their ability to produce toxins.

Because of the high negative predictive value associated with GDH tests, they can be used as an accurate first stage screening test to denote the absence of the organism. The enzyme is produced in large quantities which makes it easy to detect in faeces and therefore parallels closely the results of culture but, like culture, does not give any information concerning toxigenicity. Hence, both a positive GDH test and a positive culture MUST be followed by a test for toxin in the faeces. Commercially produced tests for GDH may take the form of ELISA format or flow-through devices.

Clostridium difficile toxin

In 1982, Sullivan *et al.* separated and purified toxins A and B. Using gel electrophoresis they established that the molecular weight of toxin A is 440,000–500,000u and toxin B is 360,000–470,000u. They also found the toxins are acid-labile and heat-labile.



Structure of *Cl. difficile* glucosyl transferase Toxin B showing UDP and glucose. Reinert *et al.* (2005). *J. Mol. Biol.*, Vol. 351, No. 5, pp973–981

Investigation continued into the differences between the two toxins and in 1995 Riegler *et al.* published data showing the human colon is approximately 10 times more sensitive to the damaging effects of toxin B than toxin A. Since 2002, epidemic toxinotype III NAP1/027 strains, which produce high levels of the major virulence factors toxin A and toxin B, have emerged. Work by Lyras *et al.* has helped provide evidence that toxin B, not toxin A, is essential for virulence. These toxins can be found in the stools of 15–25% of patients with antibiotic-associated diarrhoea and in more than 95% of patients with PMC.

NAATs

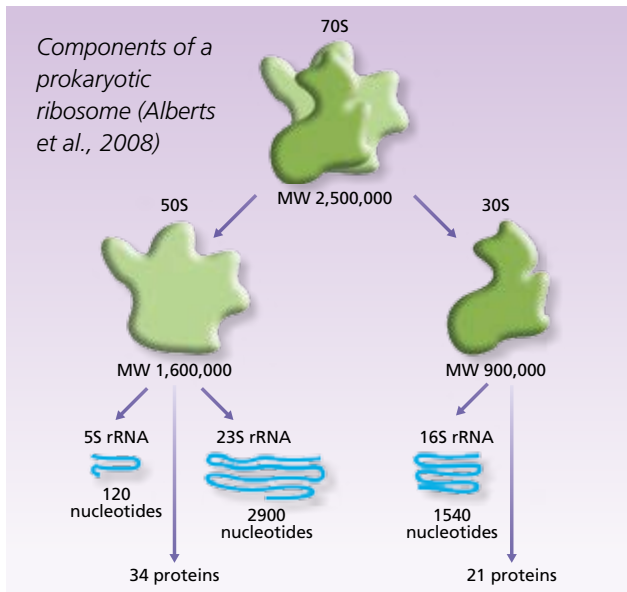
There are many forms of NAATs now available, each employing different methods of target extraction, amplification and detection. Systems can target the *tcdA*, *tcdB*, *tcdC* genes and various hypervirulence markers. However, it should be noted that the *tcdB* gene is always present in toxigenic strains while *tcdA* may be absent. Throughput, cost and ease-of-use also varies between the systems and laboratories must decide which system is most suited to their individual needs.

Lactoferrin as an inflammatory marker of infection

Lactoferrin is a by-product of white blood cells and can be detected where there is an inflammatory response. Patients who suffer from moderate to severe CDI have evidence of colonic inflammation. In such patients raised levels of lactoferrin build up at the site of infection and in severe CDI can be detected in large amounts in the faeces. In theory, this marker could be used to help diagnose severe CDI in the early stages. Lactoferrin is, however, not specific to CDI and raised levels can occur in other inflammatory conditions. Much debate revolves around the usefulness of this test and its use remains very much a local decision.

Ribotyping

The principle of prokaryotic ribotyping is based on targeting a polymorphic region in the DNA that codes for the 16S–23S RNA subunits. The ribotyping method for *Cl. difficile* was initially developed using PCR and gel electrophoresis. Ribotyping is commonly used to identify strains of *Cl. difficile* in outbreaks and to



provide data for *Cl. difficile* epidemiological studies. The Health Protection Agency (now Public Health England) began the ribotyping initiative in April 2007. Initially known as CDRNE, the network has since expanded from six to nine laboratories, including one in N. Ireland. The network was renamed in April 2009 as the *Clostridium difficile* Ribotyping Network (CDRN) for England and N. Ireland and provides a free-of-charge fingerprinting service for antimicrobial susceptibility testing and epidemiological studies.

Multilocus Variable Number Tandem Repeat Analysis (MLVA)

MLVA is often utilized as a secondary typing method following on from PCR ribotyping. It uses variable number tandem repeats (VNTR) to identify polymorphisms and is capable of discriminating between closely related *Cl. difficile* strains. It can distinguish more than 20 subtypes of *Cl. difficile* ribotype 027. An MLVA database for *Cl. difficile* has been developed by Keith Jolley (<http://pubmlst.org/cdifficile/>).

Summary

Identification of *Cl. difficile* and methods for diagnosing CDI in clinical samples has been the focus of much research over the past 80 years, while outbreaks in healthcare facilities have placed this organism in the fore of public attention. With novel methods of identification, diagnosis and epidemiology allowing rapid and effective management of patients, it is worth a final word on the availability of antibiotic treatment and the rise in antibiotic resistance. As a leading cause of antibiotic-associated diarrhoea, and an organism requiring antibiotic treatment itself, *Cl. difficile* can pose a number of challenges for effective antibiotic stewardship. Further work on resistance mechanisms and alternative treatments is needed to ensure rates of infection and associated morbidity and mortality rates continue to be effectively controlled.

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Charlotte Duncan

Pro-lab diagnostics



Governments love to 'divide and rule' so it is in no one's interest to present divided or conflicted messages



Making the case for science with a new UK Government

BIOFOCUS

With the New Year a distant memory, the General Election is beginning to loom large. There will be a new Government before we know it and, irrespective of whatever flavour it comes in, there will be huge challenges for the scientific community around the need to demonstrate value for money and even in maintaining current funding levels.

The widespread relief at a 'flat cash settlement' for science in the last funding round has slowly turned to concern that inflation has wiped out over £1 billion for UK research. At the time, the Society of Biology was one of only a few organizations to publicly voice concern. We all recognize that there are major economic challenges ahead but investment in science is part of the solution not part of the problem. We too often knock the UK research base for a failure to translate research into new products and services led by UK business. Although we must always aim for more, the reality is that we have many great examples of success and our strength in science is a beacon for overseas students and researchers. This is our historic record. Looking to the future, if we don't quickly regain lost investment, that strength may wane, and that's why we will be campaigning hard up to, and beyond, the General Election as policy for science, and its funding, starts to evolve.

We are organizing three major science events in March within the Houses of Parliament at Westminster, including a science policy debate between the main parties. All the major scientific organizations will be key partners to ensure a joined up message and we will partner with sister societies for work in the devolved nations.

We will host a dedicated page on our website with key facts and messages you can use at local events or with parliamentary candidates – the more local activity the better.

We will be seeking to present a united front for the science community, working in partnership with colleagues in chemistry, physics, mathematics and beyond. Governments love to 'divide and rule' so it is in no one's interest to present divided or conflicting messages. For us, unity starts with the bioscience

community and we urge all of our Member Organizations to engage with our policy team as much as possible to ensure biology has a single voice.

Of course, research does not take place in isolation; it is underpinned by an excellent and integrated education system, the right regulatory framework and an appropriate business environment to attract and retain investment. All of these areas, along with environmental and biodiversity policies, will be critical to assessing the new Government's overall commitment to science, evidence-led policymaking and science-based industries.

There can be no doubt that the recent Research Excellence Framework exercise has demonstrated the incredibly diverse value of science, with the life sciences faring particularly well. These messages need to be made clear to Parliamentary candidates who may well not have any background in the sciences, but not in isolation. If we invest in science but not in education and training, or try to artificially separate them, the outcome is not likely to be a good one. There remain rumours that higher education will be given to the Department for Education post-election, separating it from research. I'm sure this is something we would all have concerns about and we will be monitoring the situation closely and making our views clear.

As we celebrate our 5th birthday, we are turning our minds to the next five years. We are keen that the work we undertake on behalf of the sector reflects the priorities in our diverse Member Organizations. We are pulling together the first draft of our future plan in March with a view to finalizing it in June. If you have views on what you would like to see more of or where our priorities should lie, we would love to hear from you. Please feel free to email me:

markdowns@societyofbiology.org



Mark Downs FSB
Chief Executive, Society of Biology

2014 *Environmental Microbiology* LECTURE report



The 2014 *Environmental Microbiology* Lecture was given by Professor James Prosser from the Institute of Biological and Environmental Sciences at the University of Aberdeen.

You can watch the lecture in its entirety here:
http://bit.ly/EMI_2014

The title, "***Unimaginable, unprecedented microbial diversity: whence, so what, and can we learn from nitrifiers?***", Professor Prosser said was, on reflection, a bit long, and perhaps mildly pretentious, but nevertheless reflected what he wanted to talk about; that is, the increase in the study of microbial diversity over the past 20–25 years and what we can learn from such studies that focus on nitrifiers in the soil.

There are a number of problems with trying to characterize the diversity of microorganisms in a soil sample. The typical 1g soil sample contains more than 7 billion microorganisms, which is directly equivalent to the number of human beings on Earth. And, just as human beings interact with only a tiny subset of other individuals, the same is true for microorganisms,

perhaps with the exception of a few that might hitch a ride with a passing earthworm. Equally, it isn't possible to go out with a pair of binoculars, as a bird ecologist would, to observe the behaviours, diets and social interactions of microorganisms.

Of all the microorganisms in soil, only a very few are culturable in the lab (see pages 18–9 for a new technique that has proven fruitful in the search for new antibiotics from soil bacteria). It is also quite difficult to establish what constitutes a species as there is no reliable rule to do so for microorganisms.

Professor Prosser went on to describe the Woese Tree of Life, which, rather than relying on phenotypic analysis, focused on the 16S ribosomal RNA gene. This gene is present in every living cell; it is of sufficient length to be quite variable; and it can be sequenced to provide discrimination between different members of a microbial community. This led to the major finding that bacteria and archaea were as different from each other as they were from eukaryotes, and that the majority of diversity in eukaryotes is also in microorganisms. In fact, a view now exists that has eukaryotes evolving from archaea, giving the tree of life two rather than three original branches.

This technique of taking an environmental sample, attempting to grow it in the lab, and then carrying out 16S RNA and/or phenotypic analysis, continued as the best possible approach, until the early 1990s. There was a risk of selecting particular organisms by growth conditions, and the likelihood that researchers were not characterizing representative communities. And although a useful view had emerged – microbial communities were very diverse – there was still a need to have better information.

What happened in the early 1990s was the development of techniques to examine the genetic

Jim Prosser receives the Environmental Microbiology Lecture award from Editor, Ken Timmis and SfAM President Christine Dodd



sequences of organisms and so classify communities that way. Professor Prosser described the early work on ammonia oxidizer communities where DNA/RNA-targeted molecular analysis enabled enquiry into function, as well as phylogeny.

The basic principles are now very familiar to all molecular biologists – the cells are broken open, genetic material extracted and purified, and then PCR is used to amplify sequences of interest. For ammonia oxidisers, researchers were looking for genes that are functional for ammonia oxidation. What they ended up with was a population of gene fragments of the same length, but with differing sequences. At the time, sequencing was prohibitively expensive and so fingerprinting techniques were usually employed. In this case, it was DGGE. This is where the “unimaginable, unprecedented” of the title comes in; what was startling was that every sequence was different – the diversity was enormous! Even in groups of organisms that were relatively well known, there was far more diversity than had been observed previously. And, perhaps most interestingly, there were sequences that represented large high level groups that had never been cultivated in the lab.

These developments raised two fundamental questions:

- 1) **Where has all this diversity come from?**
- 2) **So what? Could you lose some of these taxonomic groups and have the soil function in the same way?**

These questions are still far from being answered but Professor Prosser went on to describe the approach that has been taken to address these ecological questions as they apply to nitrifiers, the ammonia oxidizers.

Ammonia appears in the soil from one of three sources:

- **Animal excreta**
- **Decomposing organic material**
- **Agricultural application of fertilizer**

Ammonia is taken in by the ammonia oxidizers, converted to nitrite, and then leaves the cells to be converted to nitrate by the nitrite oxidizers. Nitrate is then denitrified or leached – either way it's bad news: greenhouse gas emissions or groundwater pollution. Overall, there is huge loss of fertilizer – 70% in some places. So these organisms are of great environmental interest. And, it turns out, there is great diversity.

This work began with a paradox: ammonia oxidizers generally can't grow in culture under acidic conditions, but, on the whole, they are collected from acid soils, which represent around 50% of arable land in the world and can have a pH as low as 4.5. There were various ideas about how this could be so, but until the molecular techniques became available, it was impossible to find out whether soil pH selected for certain populations of ammonia oxidizers.

By interrogating genetic sequences, it emerged that there were two genera of ammonia oxidizers: *Nitrosospira* and *Nitrosomonas*. No two sequences were the same, and no environmental sample matched any cultured sample in this respect. For *Nitrosospira* in particular, the sequences could be clustered, and the clusters were clearly defined by the environment from which they came.

One important outcome from this exercise was to discover that *Nitrosomonas europaea* – the lab rat of ammonia oxidizers – was actually not typical. The majority of soil-derived sequences are actually from the *Nitrosospira* genera.

What followed was a series of experiments to look at the effects of soil pH on the ecology, distribution and community ecology of the ammonia oxidizers.

Looking at global samples, as well as local samples from the 53-year-old Craibstone pH-controlled plots, it became clear that there was an important group of archaeal ammonia oxidizers – the thaumarchaea. Activity of the *amoA* genes from these archaea was higher at low pH and fell off quite dramatically as pH increased. But for the bacterial ammonia oxidizers, the activity actually rose gradually with increasing pH.

Then there was an amazing coincidence. A PhD student was trying to enrich ammonia oxidizers from soil, and *Nitrosotalea devanattera* was discovered. *Nitrosotalea devanattera* is very acidophilic – it doesn't grow where the bacterial ammonia oxidizers grow and it belongs with a group of globally distributed acidophilic archaea. Further studies suggest that this organism is indeed responsible for acidophilic nitrification in soil.

Another interesting finding is that the speciation rate, based on *amoA* genes, remains quite high and the phenotypic analysis suggests the same.

The diversity of ammonia oxidisers is more than likely due to speciation resulting from adaptation to pH.

Professor Prosser also talked about ongoing work on drought tolerance, which looks to be far better in the bacterial species than the archaea. He concluded by saying that the lessons learned may apply more broadly in microbial ecology and the fact that we don't have species definitions is a problem – are there strong links between phylogeny and function? He also told the audience that he has learnt that you don't get answers unless you ask questions; hypothesis-driven experiments are yielding far more interesting information than work that simply seeks to survey what is out there in the microbial community in soil. This is by no means the end of the story of the ammonia oxidizers – keep watching this space...

Nancy Mendoza

Society for Applied Microbiology

JournalWATCH

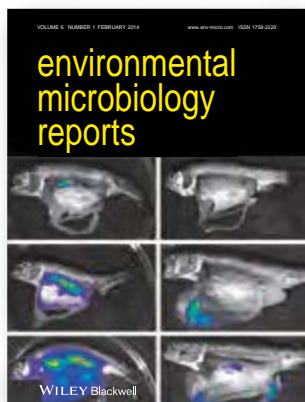
Highly read articles from the SfAM journals in 2014

Environmental Microbiology Reports

www.env-micro-reports.com

A shift in the archaeal nitrifier community in response to natural and anthropogenic disturbances in the northern Gulf of Mexico

S. E. Newell *et al.*



The Gulf of Mexico is affected by hurricanes and suffers seasonal hypoxia. The Deepwater Horizon oil spill impacted every trophic level in the coastal region. Despite their importance in bioremediation and biogeochemical cycles, it is difficult to predict the responses of microbial communities to physical and anthropogenic

disturbances. Here, we quantify sediment ammonia-oxidizing archaeal (AOA) community diversity, resistance and resilience, and important geochemical factors after major hurricanes and the oil spill. Dominant AOA archetypes correlated with different geochemical factors, suggesting that different AOA are constrained by distinct parameters. Diversity was lowest after the hurricanes, showing weak resistance to physical disturbances. However, diversity was highest during the oil spill and coincided with a community shift, suggesting a new alternative stable state sustained for at least one year. The new AOA community was not significantly different from that at the spill site one year after the spill. This sustained shift in nitrifier community structure may be a result of oil exposure.

<http://onlinelibrary.wiley.com/doi/10.1111/1758-2229.12114/abstract>

Myxobacterial community is a predominant and highly diverse bacterial group in soil niches

X-W. Zhou *et al.*

Although many molecular ecological surveys have been conducted, there is little concerning the details of specific bacterial groups, resulting in an incomplete understanding of the microorganismal composition and community structures in the environment. Myxobacteria are micropredators that are metabolically active in the soil microbial food web and have typically been considered minority components of soil bacterial communities. In this study, we surveyed the percentage of myxobacteria in a single soil sample via pyrosequencing on combined universal libraries of the V3-V4 and V6-V8 hypervariable regions of the 16S rRNA gene. Surprisingly, myxobacteria accounted for 4.10% of the bacterial community and 7.5% of the total operational taxonomic units at the 3% similarity level in the soil, containing almost all of the cultivated myxobacterial families or genera. To testify the appearance of myxobacteria in soil niches, we retrieved myxobacteria-related 16S rRNA gene sequences of 103 high-throughput sequencing data sets obtained from public databases. The results indicated that myxobacteria-related sequences were among the predominant groups in these data sets accounting for 0.4–4.5% of bacterial communities. The abundance of myxobacterial communities was correlated with site temperature, carbon-to-nitrogen ratio and pH values. Based on these results, we discuss the survival strategies of the myxobacterial community in soil.

<http://onlinelibrary.wiley.com/doi/10.1111/1758-2229.12107/abstract>

Environmental Microbiology

www.env-micro.com

Metagenomic 16S rDNA Illumina tags are a powerful alternative to amplicon sequencing to explore diversity and structure of microbial communities

R. Logares *et al.*



Sequencing of 16S rDNA PCR amplicons is the most common approach for investigating environmental prokaryotic diversity, despite the known biases introduced during PCR. Here we show that 16S rDNA fragments derived from Illumina-sequenced environmental metagenomes (*mi*tags) are a powerful alternative to 16S rDNA amplicons for

investigating the taxonomic diversity and structure of prokaryotic communities. As part of the *Tara* Oceans global expedition, marine plankton was sampled in three locations, resulting in 29 subsamples for which metagenomes were produced by shotgun Illumina sequencing (ca. 700Gb). For comparative analyses, a subset of samples was also selected for Roche-454 sequencing using both shotgun (*m454*tags; 13 metagenomes, ca. 2.4Gb) and 16S rDNA amplicon (*454*tags; ca. 0.075Gb) approaches. Our results indicate that by overcoming PCR biases related to amplification and primer mismatch, *mi*tags may provide more realistic estimates of community richness and evenness than amplicon *454*tags. In addition, *mi*tags can capture expected beta diversity patterns. Using *mi*tags is now economically feasible given the dramatic reduction in high-throughput sequencing costs, having the advantage of retrieving simultaneously both taxonomic (Bacteria, Archaea and Eukarya) and functional information from the same microbial community.

<http://onlinelibrary.wiley.com/doi/10.1111/1462-2920.12250/abstract>

Structural basis for adaptation of lactobacilli to gastrointestinal mucus

S. Etzold *et al.*

The mucus layer covering the gastrointestinal (GI) epithelium is critical in selecting and maintaining homeostatic interactions with our gut bacteria. However, the underpinning mechanisms of these interactions are not understood. Here, we provide structural and functional insights into the canonical mucus-binding protein (MUB), a multi-repeat cell-surface adhesin found in *Lactobacillus* inhabitants

of the GI tract. This study reveals functional and structural features which may affect tropism of microbes across mucus and along the GI tract, providing unique insights into the mechanisms adopted by commensals and probiotics to adapt to the mucosal environment.

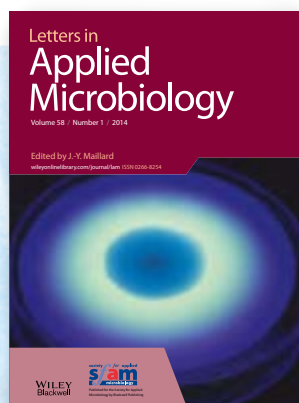
<http://onlinelibrary.wiley.com/doi/10.1111/1462-2920.12377/abstract>

Letters in Applied Microbiology

www.lettersappliedmicro.com

Mitreicin A, an endolysin-like bacteriolytic enzyme from a newly isolated soil streptomycete

M. H. Farris and A. D. Steinberg



The gene of a new protein antimicrobial, Mitreicin A, was discovered in the genome of a soil bacterium. The purified recombinant enzyme, resulting from heterologous over expression of the gene, was found to be tolerant of increased pH conditions and to have bacteriolytic activity against Gram-negative bacteria of

the medically important genera *Aeromonas*, *Escherichia*, *Salmonella*, *Shigella*, *Vibrio* and *Yersinia*. Characterization of enzymes such as Mitreicin A from previously uncharacterized bacteria provides potential options for new biocontrol agents in medically and economically important applications like therapeutics, disinfectants, food preservatives, agricultural livestock antimicrobials and inhibitors of biofilm production.

<http://onlinelibrary.wiley.com/doi/10.1111/lam.12220/abstract>

Specific capture and detection of *Staphylococcus aureus* with high-affinity modified aptamers to cell surface components

A. Baumstummeler *et al.*

Monitoring for microbial contamination of food, water, non-sterile products or the environment is typically based on culture, PCR or antibodies. Aptamers that bind with high specificity and affinity to well-conserved cell surface epitopes represent a promising novel type of reagents to detect bacterial cells without the need for culture or cell lysis, including for the capture and enrichment of bacteria present at low cell densities and for the direct detection via qPCR or fluorescent staining.

<http://onlinelibrary.wiley.com/doi/10.1111/lam.12295/abstract>

Journal of Applied Microbiology

www.journalappliedmicro.com

Fungal endophytes enhance wheat heat and drought tolerance in terms of grain yield and second-generation seed viability

M. Hubbard, J. J. Germida and V. Vujanovic



This study evaluated the impact of fungal endophyte symbiosis on the growth, ecophysiological and reproductive success of wheat exposed to heat and drought. We concluded that the tested consortium of endophytes has the potential to increase wheat tolerance for abiotic stress and improved germination

in endophyte-free second-generation seeds arising from stressed plants could be applicable to agriculture. The mechanisms by which intergenerational endophyte-mediated effects occur warrants further research.

<http://onlinelibrary.wiley.com/doi/10.1111/jam.12311/abstract>

Wild mushroom extracts potentiate the action of standard antibiotics against multiresistant bacteria

M. J. Alves *et al.*

The main objective of this study was to evaluate the capacity of wild mushroom extracts to potentiate the action of standard antibiotics, through synergisms that allow a decrease in their therapeutic doses and ultimately contribute to the reduction of resistances. This study shows that, similarly to plants, some mushroom extracts can potentiate the action of antibiotics extensively used in clinical practice for Gram-positive or Gram-negative bacteria, with positive action even against multiresistant bacteria. This signifies that mushroom extracts could decrease therapeutic doses of standard antibiotics and reduce microorganism's resistance to those drugs.

<http://onlinelibrary.wiley.com/doi/10.1111/jam.12348/abstract>

Melissa McCulloch

Wiley-Blackwell

Microbial Biotechnology

www.microbialbiotech.com

Detection of hepatitis E virus (HEV) through the different stages of pig manure composting plants

M. García, S. Fernández-Barredo and M. T. Pérez-Gracia



Hepatitis E virus (HEV) is an increasing cause of acute hepatitis in industrialized countries. The aim of this study was to evaluate the presence of HEV in pig manure composting plants located in Spain. For this purpose, a total of 594 samples were taken in 54 sampling sessions from the different stages of composting treatment in these plants

as follows: slurry reception ponds, anaerobic ponds, aerobic ponds, fermentation zone and composting final products. HEV was detected by reverse transcription polymerase chain reaction (RT-nested PCR) in four (80%) of five plants studied, mainly in the first stages of the process. HEV was not detected in any final product (compost) sample, destined to be commercialized as a soil fertilizer, suggesting that composting is a suitable method to eliminate HEV and thus, to reduce the transmission of HEV from pigs to humans.

<http://onlinelibrary.wiley.com/doi/10.1111/1751-7915.12064/abstract>

Siderophores in environmental research: roles and applications

E. Ahmed and S. J. M. Holmström

Siderophores have received much attention in recent years because of their potential roles and applications in various areas of environmental research. Their significance in these applications is because siderophores have the ability to bind a variety of metals in addition to iron, and they have a wide range of chemical structures and specific properties. For instance, siderophores function as biocontrols, biosensors, and bioremediation and chelation agents, in addition to their important role in weathering soil minerals and enhancing plant growth. The aim of this literature review is to outline and discuss the important roles and functions of siderophores in different environmental habitats and emphasize the significant roles that these small organic molecules could play in applied environmental processes.

<http://onlinelibrary.wiley.com/doi/10.1111/1751-7915.12117/abstract>

In the 40th of a series of articles about statistics for biologists, Richard Armstrong and Anthony Hilton discuss **Cochran's Q test**

StatNote40

Introduction

In a previous StatNote, an experiment was conducted in which the possible effect of two novel media supplements (S1 and S2) on increasing cell biomass was investigated (StatNote 10, Hilton & Armstrong, 2007). To carry out the experiment, three groups of 10l fermentation vessels were sterilized and filled with identical growth media, with the exception that the media in two sets of vessels was supplemented with 10ml of either medium supplement S1 or S2, the vessels then being inoculated with a culture of a bacterium.

Essentially, this experiment could be carried out using two experimental designs. First, in a fully randomized 'one-way design', treatments could be allocated at random and without restriction to the replicate vessels and the data analysed using a one-way ANOVA in a randomized design, i.e., as per StatNote 30, Hilton & Armstrong, 2012. Second, the experiment could be carried out using a 'two-way design', e.g., 30 vessels could be divided into 10 groups of 3, each group representing 'a replication' or 'block' with the intention of setting up and processing each complete block (control, treatments S1 and S2) on 10 separate occasions, the treatments being allocated to the 3 vessels within a replication independently and at random (StatNote 10, Hilton & Armstrong, 2007). To analyse these data, a two-way ANOVA is required in which each observation is classified in two ways, i.e., according to treatment and occasion. The resulting ANOVA will have three sources of variation, viz. treatments, occasions and error (Snedecor & Cochran, 1980). This design may reduce the degree of error variation present in the experiment by processing each complete block on a single occasion, thus increasing its power relative to a completely randomized design (StatNote 8, Hilton & Armstrong, 2007).

If the data were not normally distributed, the appropriate non-parametric analysis would be Friedman's ANOVA (StatNote 23, Hilton & Armstrong, 2010). Cochran's Q test is another non-parametric analysis which can be applied to a two-way design in which the data are binomial and can take only two possible outcomes, e.g., 0 or 1, alive or dead, present or absent, clean or dirty, infected or non-infected and is an extension to the binomial tests introduced in StatNote 39 (Hilton & Armstrong, 2014). This StatNote describes the application of this test in the analysis of the changes which occur in the fungal flora of forestry nursery beds after two different sterilization procedures (Warcup, 1951).

Scenario

The effect of two methods of sterilization was studied on the fungal flora of forestry nursery beds. The first method involved steam sterilization in which pipes carrying steam were buried to a depth of 20–25cm within the experimental plots under a tarpaulin cover, achieving a 100°C surface temperature for 15 minutes. The second method involved the addition of formalin to the plots as a 4% solution at a rate of 5l per m² of plot. Twenty large plots of land ('blocks') were used, each divided into three equally sized subplots, the treatments, viz. control, steam sterilization, and formalin treatment being applied at random and independently within each block. The fungi present in the top 25cm of soil three weeks after treatments were then investigated using various culture plating methods. Colonization of the soil after treatment is usually by species of the genus *Mortierella*, and hence the presence/absence of colonies of this fungus was recorded from samples of soil taken from each subplot. The data are summarized in Table 1.

Table 1. The effect of two methods of soil sterilization (steam treatment or addition of formalin) on the recolonization of forestry soil by the fungus *Mortierella*. Data are the presence/absence of colonies of the fungus isolated from samples of each plot three weeks after treatment.

Plots ('blocks')	Treatment groups		
	Untreated	Steam sterilization	Formalin sterilization
1	1	0	1
2	1	0	1
3	1	0	1
4	1	0	0
5	1	0	0
6	1	0	0
7	1	0	1
8	0	0	1
9	1	0	0
10	1	1	0
11	1	0	0
12	1	0	1
13	1	0	0
14	1	0	1
15	1	0	0
16	0	0	1
17	1	1	1
18	1	0	1
19	1	0	0
20	1	0	0

Cochran's Q test: All groups $T' = 20.21$ (2DF, $P < 0.001$); Steam versus formalin treatment $T' = 6.4$ (1DF, $P < 0.05$).

How is the analysis carried out?

Cochran's Q test is essentially an analysis of a two-way, randomized block design in which the response can only take one of two possible outcomes. The method tests two hypotheses: (1) that the 'K' treatments, where $K > 2$, have identical effects and (2) that there is a difference in the effect of individual treatments. The test statistic T' is defined as follows:

$$T' = \frac{K(K-1) \sum_{j=1}^K (X_j - N/K)^2}{\sum_{i=1}^b X_i (K - X_i)} \quad (1)$$

where K = number of treatments or groups, b = number of blocks, X_j = column total for the 'jth' treatment, b = number of plots (blocks), X_i = row total for the 'ith' block and N = grand total of all observations. For a particular significance level 'a', the value of T' has to be greater than chi-square (χ^2) with $K - 1$ degrees of freedom (DF). If the null hypothesis that all treatments are equally effective is rejected, then a pairwise comparison between the treatments can be made. Cochran's Q test does make some assumptions regarding the experimental data: (1) that the number of blocks 'b' is fairly large, (2) that blocks are randomly selected and (3) that the outcome of the experiment is binary and applies to all treatments within each block.

Interpretation of the results

The results of Cochran's Q test are shown in Table 1. The test of the hypothesis that the three treatments resulted in identical outcomes gives $T' = 20.21$, which is significant at the $P < 0.001$ level of probability, indicating highly significant differences among the treatments. A specific comparison of the steam and formalin treatment gives $T' = 6.4$, which is significant at $P < 0.05$. Hence, sterilization by formalin was not as effective as steam treatment. Normally, recolonization of the plots by fungi after sterilization occurs largely as a result of lateral spread from adjacent untreated areas together with some recolonization from the associated air. In the formalin treatment, however, it was observed that fungi were only destroyed down to a depth of 12cm and penetration of the treatment into the soil was relatively slow. Hence, it is likely that recolonization occurred rapidly in the formalin treatment area from lower unaffected areas due to poor penetration.

Conclusion

There are three methods of analysing a two-way design in randomized blocks. First, if the data are normally distributed, the appropriate analysis is a two-way ANOVA in randomized blocks (StatNote 10, Hilton & Armstrong, 2007). Second, if the data are non-parametric and are ordinal (more than 2 groups) or continuous, the appropriate analysis is Friedman's ANOVA (StatNote 23, Hilton & Armstrong, 2010). Third, if the data are binary, then Cochran's Q test should be used. The method can test whether the 'K' treatments have identical effects and whether there is a difference in the effect of individual treatments.

FURTHER READING

Snedecor, G. W. and Cochran, W. G. (1980). *Statistical Methods*. 7th edition, Iowa State University Press, Ames, Iowa.

Warcup, J. H. (1960). *Methods for isolation and estimation of activity of fungi in soil*. In: *The Ecology of Soil Fungi* (Ed D. Parkinson and J. S. Waid), Liverpool University Press.

A. C. Hilton *Pharmaceutical Sciences*

R. A. Armstrong *Vision Sciences*

Aston University, Birmingham, B4 7ET, UK.

SfAM Spring Meeting

16 April 2015

The Sheffield Hilton, Sheffield, UK

9:30 – 10:00 **Tea, coffee, trade exhibition and registration**

Chair: TBC

10:00 – 10:40 **New developments in antimicrobial resistance**

Neil Woodford, *Antimicrobial Resistance and Healthcare Associated Infections Reference Unit, PHE Colindale, UK*

10:40 – 11:20 **Current issues in medical mycology**

Elizabeth Johnson, *PHE Mycology Reference Laboratory, Public Health England South West Laboratory, Bristol, UK*

11:20 – 12:00 **What's new in the diagnosis of tuberculosis**

Tim Brown, *PHE National Mycobacterium Reference Laboratory, UK*

12:00 – 13:10 **Lunch and trade exhibition**

Case studies in clinical microbiology

Chair: TBC

13:10 – 13:35 **Study 1: *Strongyloides* sp. – an unusual treatment**

Dave Partridge, *Sheffield Teaching Hospitals NHS Foundation Trust, UK*

13:35 – 14:00 **Study 2: *Bacillus cereus* septicaemia from contaminated nutrition drips**

William Newsholme, *Guys and St. Thomas's Hospital, UK*

14:00 – 14:25 **Study 3: *Stachybotrys* sp. – a date to remember**

Stephen Wilson, *Sheffield Teaching Hospitals NHS Foundation Trust, UK*

14:25 – 14:45 **Tea and coffee**

14:45 – 15:10 **Study 4: *Legionella* in birthing pools**

Julie Samuel, *Public Health England, UK*

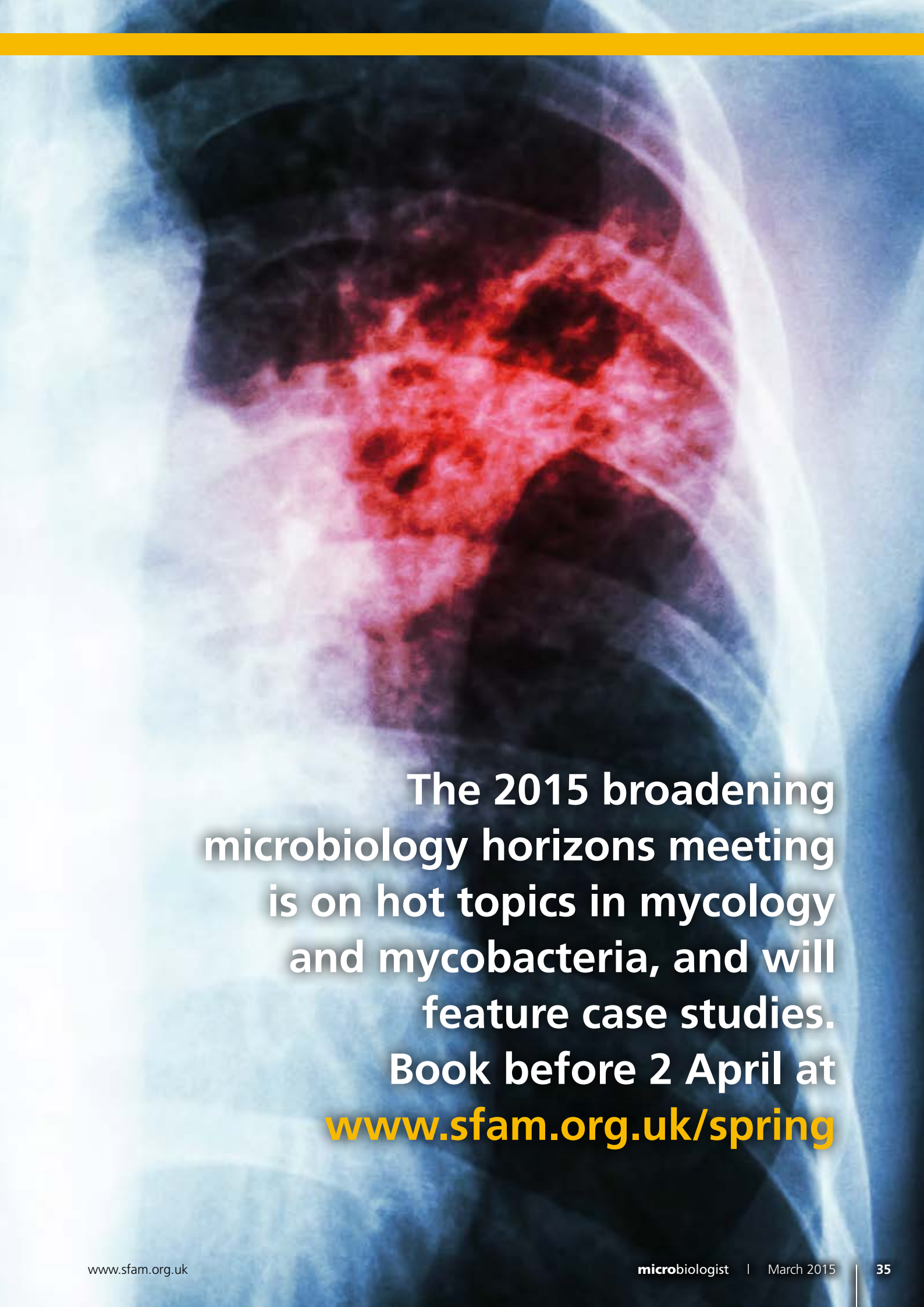
15:10 – 15:35 **Study 5: An unusual haematology case**

Dave Partridge, *Sheffield Teaching Hospitals NHS Foundation Trust, UK*

15:35 – 16:00 **Study 6: An unusual Intensive Care Unit case**

Stephen Wilson, *Sheffield Teaching Hospitals NHS Foundation Trust, UK*

16:00 **Close**



**The 2015 broadening
microbiology horizons meeting
is on hot topics in mycology
and mycobacteria, and will
feature case studies.**

**Book before 2 April at
www.sfam.org.uk/spring**

SfAM Summer Conference 2015

29 June to 2 July 2015, Intercontinental Dublin

Monday 29 June 2015

- 11:00 – 17:00 **Workshop**
 18:00 – 19:00 **Journal of Applied Microbiology Lecture**
 19:00 – 20:00 **Drinks reception and buffet**
 20:30 onwards **Quiz night**

Tuesday 30 June 2015

SESSION 1 DEVELOPING A NEW UNDERSTANDING OF FERMENTATIONS

Chair: TBC

- 09:00 – 09:35 **Use of metagenomics to study microbial fermentations**
 Danilo Ercolini, *University of Naples, Italy*
- 09:35 – 10:10 **Metabolomics approaches for studying fermentation processes**
 Eddy Smid, *Wageningen University, The Netherlands*
- 10:10 – 11:05 **Tea, coffee and trade show**
- 11:05 – 11:40 **Food poisoning and fermented foods**
 Kathie Grant, *Public Health England, UK*
- 11:40 – 12:15 **Systems engineering of cheese consortia**
 Alan Ward, *Chung Ang University, Korea*
- 12:15 – 13:15 **Lunch, trade show and posters**

SESSION 2 IMPROVING FERMENTATIONS

Chair: TBC

- 13:15 – 13:50 **Generating superior yeasts for the fermentation industry – from beer, wine and chocolate to biofuels**
 Kevin Verstrepen, *University of Leuven/VIB, Belgium*

- 13:50 – 14:25 **Development of phage-insensitive cultures for the fermentation industry**
 Paul Ross, *TEAGASC, Ireland*
- 14:25 – 14:45 **Tea, coffee and trade show**
- 14:45 – 15:20 **Bacteriocins: providing innate immunity for food systems**
 Colin Hill, *University College Cork, Ireland*
- 15:20 – 15:55 **Gene-trait matching: predicting functional properties of microbial strains**
 Sacha van Hijum, *NIZO food research, The Netherlands*
- 16:00 – 17:00 **Attended poster session A**
- 17:00 – 18:00 **Early career scientist session**
- 17:00 – 19:30 **Trade show with wine, buffet and a competition**

Wednesday 1 July 2015

SESSION 3 THE ROLE OF MICROFLORA

Chair: Tim Aldsworth

- 09:00 – 09:35 **The role of non-starter organisms in cheese ripening**
 Paul McSweeney, *University College Cork, Ireland*
- 09:35 – 10:10 **Microbiological and physicochemical characteristics of commercial-scale calcium chloride cucumber fermentations without sodium chloride**
 Ilenys Perez-Diaz, *USDA-ARS Food Science Research Unit, USA*
- 10:10 – 10:45 **Cocoa bean fermentation**
 Luc de Vuyst, *Brussels University, Belgium*
- 10:45 – 11:05 **Tea, coffee and trade show**

FERMENTED FOODS and BEVERAGES

Thursday 2 July 2015

- 11.05 – 11.40 **Sourdough fermentation**
Marco Gobbetti, *University of Bari, Italy*
- 11:40 – 12:15 **Adaptation of the starter culture *Staphylococcus xylosus* to meat products**
Régine Talon, *INRA, France*
- 12.15 – 13.15 **Lunch and networking**
- 13.15 – 13.50 **Lactic acid bacteria – friend or foe in wet cassava starch**
Andrew Graffham, *NRI, University of Greenwich, UK*
- 13:50 – 14:25 **Fermented soya bean products**
Rob Nout, *Wageningen University, The Netherlands*
- 14:30 – 15:30 **Student oral presentations**
- 15:30 – 16:30 **Attended poster session B**
- SfAM AWARD LECTURES**
- Chair: Christine Dodd,
SfAM President
- 16:30 – 16:35 **Introduction to the New Lecturer Research Grant**
Christine Dodd,
SfAM President
- 16:35 – 17.10 **SfAM New Lecturer Research Grant Lecture**
TBC
- 17:10 – 17:15 **Introduction to the W H Pierce Prize**
Christine Dodd,
SfAM President
- 17:15 – 17:45 **W H Pierce Prize Lecture**
TBC
- 17:45 – 18:15 **Annual General Meeting**
- 19:00 onwards **Drinks reception and conference dinner**

SESSION 4 TRADITIONAL FERMENTATIONS

- Chair: Christine Dodd,
University of Nottingham, UK
- 09:00 – 09:35 **Bacterial diversity and functions during production of traditional fermented West African cereal foods**
Folarin Oguntoyinbo
- 09:35 – 10:10 **Fermentation of cassava**
Linda Nicolaidis, *NRI, University of Greenwich, UK*
- 10:10 – 10:45 **Tea and coffee**
- 10:45 – 11:20 **The microbiology of Bandji, traditional palm wine of the palm tree *Borassus akeassii***
Irene Ouoba
- 11:20 – 11:55 **Craft brewing – an industry sector in fermentation**
Jerry Avis, *University of Nottingham, UK*
- 12.00 – 13.00 **Lunch and depart**

Closing date for registration:

Friday 12 June 2015

Book now at:

www.sfam.org.uk/summer

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Deadlines

Call for nominations to the Executive Committee

There will be a number of vacancies on the SfAM Executive Committee in July 2015. Nominations are invited from all Full Members of the Society for these vacancies.

Nominations must be made in writing by post or email to lucy@sfam.org.uk and received by the Society Office by **FRIDAY 1 MAY 2015**.

Should nominations exceed vacancies, election will be by a system of postal voting arranged by the Executive Committee.

Call for nominations for the W H Pierce Prize

Do you know a young microbiologist (under 40 years of age) who has made a substantial contribution to microbiology? If so, why not nominate them for this prestigious and substantial award which is now worth £3000. The award was instituted in 1984 by Oxoid to commemorate the life and works of the late W. H. (Bill) Pierce, former Chief Bacteriologist at Oxoid Ltd. and a long-time Member of the Society. The prize is presented annually at the Summer Conference. Full Members wishing to make a nomination should write in confidence to the General Secretary, Professor Mark Fielder, at the Society Office in Bedford, including a full CV of the nominee and a letter of support. Please note that application is through nomination by Full Members of SfAM only and that there are no official forms for this award.

CLOSING DATE FOR NOMINATIONS IS **FRIDAY 17 APRIL 2015**

Call for Abstracts 2015 Summer Conference

Would you like to display a poster or give a student oral presentation at the SfAM Summer Conference, taking place in Dublin, 29 June – 2 July 2015?

We are now accepting abstracts, which can be on any topic in microbiology.

DEADLINE FOR ABSTRACT SUBMISSIONS: >> **13 MARCH 2015**

REVIEW DECISIONS COMMUNICATED TO AUTHORS: >> **17 APRIL 2015**

DEADLINE FOR AUTHORS TO REGISTER: >> **24 APRIL 2015**

DEADLINE FOR VISA AND TRAVEL DOCUMENTS, WHERE RELEVANT: >> **5 JUNE 2015**

For further information please contact:

Sally Hawkes

Email: sally@sfam.org.uk

Telephone: +44 (0)1933 382191

More Summer Conference deadlines

PRESIDENT'S FUND SUBMISSIONS >> **13 MARCH 2015**

ABSTRACT SUBMISSIONS >> **13 MARCH 2015**

STUDENTSHIP APPLICATION >> **13 MARCH 2015**

E AFFILIATE BURSARY SUBMISSIONS >> **13 MARCH 2015**

W H PIERCE PRIZE NOMINATIONS >> **17 APRIL 2015**

NOMINATIONS FOR EXECUTIVE COMMITTEE >> **1 MAY 2015**

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- Hot topics in mycology
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Tuesday 13 October
**Environmental
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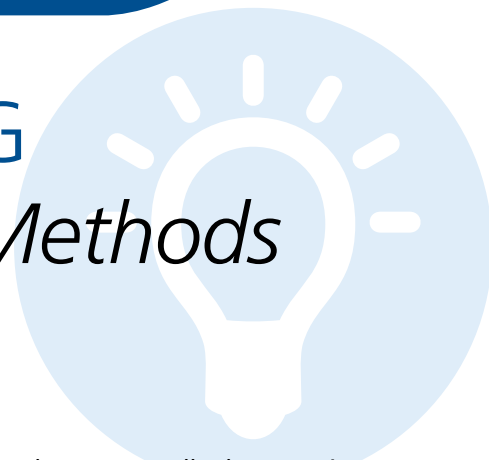
Ken Nealson
University of Southern California

Royal Society of Medicine,
London, UK

sfam.org.uk/events

PECS AUTUMN MEETING

'Current and Advanced Methods in Microbiology' Report



The 3rd annual PECS Autumn Meeting was held at the Royal Society of Medicine, with a fantastic turnout of students ready to update their knowledge with a selection of speakers from

industry and academia. First to speak was Alison Davies, from Primer Design Ltd, with an interesting insight into the many applications of qPCR, from veterinary, medicine, food quality testing and bio-threats. Moving away from PCR, Tim Nichol from Sheffield Hallam University spoke next about antimicrobial susceptibility testing. He started with an update of techniques that can be used and how they were used in his research on the antibiotic coating of orthopaedics.

After a short break for the poster session, where Student Members did a fantastic job of displaying their work, we moved on to the Student Members' session. Fittonia Elgina, explained a new biosensor-based immunoassay for the rapid detection of poultry-associated *Campylobacter* spp. Sarah Farnham, who was an SfAM Students into Work Grant beneficiary, spoke next about her work on carvacrol and thymol vapour use within food packaging to inhibit *E. coli*, with very promising results. Delveen Ibrahim, then gave an overview of her work on antibiotic resistance in *E. coli* from cattle slurry, with a surprising 61% of isolates carrying resistance to at least one antibiotic.

After lunch, James Williamson spoke about the techniques he is using to produce high value chemotherapy agent precursors from *Pantoea agglomerans*, with some interesting phenotypic changes. Closing the session was Wing Sun Faith Chung, speaking about her work on the modulation of human gut microbiota through the use of prebiotics.

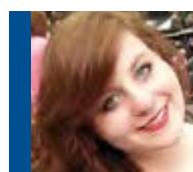


All of the Student Members gave really clear, concise presentations and did a fantastic job of speaking to a general audience as is typical at these meetings.

Andrew McBain from the University of Manchester took up the reins next, explaining some of the work that he has undertaken on biofilms, including methods for determining the efficacy of antibiotics in the more challenging environment of a biofilm. Gareth Robinson and Liz Anderson, both from the University of the West of England, shared the next slot to talk about bioluminescence and its many uses which included a look at the efficacy of household cleaning agents and chemotherapy drugs. Andrew Sails from Public Health England was the final speaker of the day, talking about what next generation sequencing can do for clinical microbiology and public health. This is an especially good technique for slow growing bacteria, allowing the study of shifts in the genome. He also described some of the challenges that need to be overcome first, such as cost and building a common language.

Finally, Christine Dodd, SfAM President, ended the day by presenting the awards for best student poster and student oral presentation, which went to Okechukwu Onianwa and Fittonia Elgina respectively.

At this point we would like to thank all of the speakers, the Student Members and the PECS Committee for making the day a really useful update, not just on new techniques that are coming through, but on more established techniques as well.



Zara Gerrard



We would like to thank all of the speakers, the Student Members and the PECS Committee for making the day a really useful update

Becoming a microbiologist: Unplanned, no regrets

Growing up in a rural village secondary school in Nigeria, I did not consider a university degree or career in microbiology because, as a kid, I had always loved nurses in their smart white uniform. I wanted then to obtain a certificate in the nursing profession through training in a school of nursing. My plans changed when I excelled in the secondary school leaving certificate examination and was advised by my aunt (a nurse at that time) to consider obtaining a university degree. I then enrolled for the higher school certificate and sat the university entrance examination. As I hated physics in secondary school, choosing a course to study in university became a problem. I could not do medicine or pharmacy as I did not have the required subject combination. I was left with the choice of biochemistry and microbiology although I had no basic knowledge of them. My plan was to change my course after the first year to pharmacy. Subsequently, I was offered admission to study Microbiology at the University of Ife (now Obafemi Awolowo University), Ile-Ife, Nigeria in 1980 and that was how my journey began. Obafemi Awolowo University is a public institution that was



established in 1962 and is situated on a vast 11,861 hectare site in Ile-Ife, Osun State, in southwest Nigeria.

During the orientation programme for fresh undergraduate students in the department of microbiology, professors gave talks on the different fields in microbiology, job prospects and microbial applications which ignited my interest in microbiology. I decided to stay and become a "MICROBIOLOGIST". In the BSc practical classes I was fascinated by the isolation and identification of microorganisms from food samples and being able to interpret how and why food was spoilt or caused poisoning. By the time I was completing the BSc programme, I had already decided I would proceed to postgraduate studies with specialization in food microbiology.

During my postgraduate studies, I received the Obafemi Awolowo University Postgraduate Fellowship Award which enabled me to complete my Master's degree (MSc Microbiology). My duties as a fellow included instructing undergraduate students during practical classes. My experience as a fellow got me really interested in teaching and research and, on completion of the Master's degree, I enrolled for a PhD. I was offered a position, while still on the PhD programme, as an Assistant Lecturer in the Department of Microbiology, Obafemi Awolowo University, where I earned all of my degrees (BSc, MSc and PhD).

Obafemi Awolowo University logo: By Femabits CC BY-SA 3.0 via Wikimedia Commons

My search for a postdoctoral position overseas for additional experience in microbiological techniques started after I obtained my PhD in 1998. Fortunately, I received a Commonwealth Fellowship Award for 6 months (October 2004 to April 2005) and this got me really excited because I was looking forward to a new experience, having had all of my previous training in Nigeria. The Commonwealth Fellowship Award offered me an opportunity to work with David Owens at the School of Food Biosciences, University of Reading, UK. During this period, I learnt new techniques, networked with other food microbiologists and most importantly, I joined the Society for Applied Microbiology (SfAM). I attended, for the first time, the Winter Meeting of SfAM in Norwich, UK, in January 2005 and have continued to attend the SfAM Summer Conferences. This has enabled me to network and stay in contact with David Owens (retired) and the University of Reading, which has helped my upward mobility in my teaching and research career.

In my teaching career, I have taught and supervised over 150 undergraduate dissertations and graduate theses. Although there are problems working in Nigerian universities due to electricity cuts, long strikes and inadequate funding, I have through dint of hard work, management of the limited resources, research grants, collaborations and occasional freebies from my mentors, published research articles in food microbiology in local and international journals.

My teaching and research outputs and administrative duties have earned me promotion from the level of assistant lecturer to the professorial cadre.

Rising to the peak of my career was not as easy and straightforward as it might look. I remember my first journal article (solely authored) published in the



World Journal of Microbiology and Biotechnology, Volume 10, 1994. The manuscript was bulky with irrelevant information. The reviewers' comments, which I still keep, were, "There are some juvenile statements that should be removed..." and "The paper should be condensed to a note without losing the importance of the findings." I had at that time thought that the quality of a manuscript had to do with the volume. The manuscript was finally reviewed and published as a short communication.

Over the years I have been inspired by my mentors and by successful publications in reputable journals. Constructive comments by manuscript reviewers have also helped me to improve on my problem identification, research design, data evaluation and scientific writing, leading to publishable research findings in my area of special interest (*Bacillus* fermentation of oil seeds).

The other arm of my duties in the university involves administrative activities. In providing administrative services, I have served and still serve in several leadership positions which include heading the department of microbiology (1,200 students, 22 academic and 7 non-academic staff).

In 2009, I was awarded the International Women's Forum (IWF) Leadership Foundation Fellows Award for the year 2009–2010. The training I received during the fellowship year at Harvard Business School, Boston, Massachusetts, USA, and Judge Business School, University of Cambridge, Cambridge, UK, has further accentuated my leadership skills and enhanced my job performance.

Overall, I have learnt many lessons of life studying microorganisms (the good and the bad). I am a proud microbiologist with no regrets. Despite the fact that my path and journey to microbiology was not planned I have remained in the university, training microbiologists – a job that I have enjoyed doing over the years and hope to continue.

Bridget Omafuvbe

Obafemi Awolowo University, Ile-Ife, Nigeria

MEMBERSHIP Benefits & Options

Benefits

The Society for Applied Microbiology is the voice of applied microbiology within the UK and was founded in 1931. Society Members play a leading role in shaping the future of applied microbiology, and enjoy many benefits, including:

- The opportunity to apply for one of our many grants or funds.
- Eligibility to win any of our awards or nominate a candidate for the SfAM Communications Award.
- Access to our five peer-reviewed journals: *Journal of Applied Microbiology* (JAM), *Letters in Applied Microbiology* (LAM), *Environmental Microbiology*, *Environmental Microbiology Reports* and *Microbial Biotechnology*.
- Free access to the entire collection of digitized back files for JAM and LAM dating back to 1938.
- A topical quarterly magazine, *Microbiologist*.
- Substantially reduced rates for attendance at SfAM meetings and conferences.
- Networking with worldwide professionals in over 80 countries
- Access to private Members' area of the SfAM website.
- Monthly email bulletins with the latest news from SfAM.
- Invitation to the annual *Environmental Microbiology* and *Journal of Applied Microbiology* lectures.
- Fostering cross disciplinary research.
- A 35% discount on the extensive Wiley-Blackwell collection of titles.

Detailed information about all these benefits and more can be found on the Society website at: www.sfam.org.uk/membership.

GRANTS & AWARDS

Many grants, awards and prizes are available to Members including the W H Pierce Memorial Prize and prizes for student oral presentations and posters at the Summer Conference. In addition to these substantial awards, the Society has funds to assist Members in their careers as microbiologists. These include the President's Fund, Conference Studentships, Sponsored Lecture Grants and the popular Students into Work Scheme.

Full details of all the Society's grants and awards, together with application forms, can be found on the website at www.sfam.org.uk/grants.

JOURNALS

The Society publishes two monthly journals: *Journal of Applied Microbiology* and *Letters in Applied Microbiology*. We also produce this quarterly colour magazine, *Microbiologist*, which contains features, topical news stories and full details of our meetings. The Society is also a partner with Wiley-Blackwell in the monthly journals: *Environmental Microbiology*, *Environmental Microbiology Reports* and *Microbial Biotechnology*. See more at www.sfam.org.uk/journals.

All Full and Student Members receive free access to the online versions of the Society's journals, and can also submit papers to our journals via an online submission service.

MEETINGS

We hold three annual meetings: the Winter Meeting is a one-day meeting with parallel sessions on topical subjects; the Spring Meeting is a one-day meeting tailored for personnel in clinical microbiology; and the Summer Conference is held every June/July and comprises a main symposium, a poster session, the AGM and a lively social programme. All Members are invited to our prestigious annual lectures held to commemorate the success of two of our journals: *Environmental Microbiology* and the *Journal of Applied Microbiology*. We also hold *ad hoc* meetings on topical subjects and enter into joint ventures with other organizations on topics of mutual interest.

WEBSITE

www.sfam.org.uk is the best source of detailed information on the Society and its many activities. It has a fully interactive Members-only area (www.sfam.org.uk/membersonly) where you can find archive issues of *Microbiologist*, exclusive SfAM documentation and much more.

Membership OPTIONS

- > **Full Ordinary** gives access to our many grants and awards, online access to the *Journal of Applied Microbiology*, *Letters in Applied Microbiology*, *Environmental Microbiology*, *Environmental Microbiology Reports* and *Microbial Biotechnology*, copies of *Microbiologist*, preferential registration rates at Society meetings, and access to the Members-only area of the website.
- > **Full Student** confers the same benefits as Full Membership at a specially reduced rate for full-time students not in receipt of a taxable salary.
- > **Associate** is only open to those with an interest in applied microbiology without it being a prime aspect of their job. For example, school teachers and those taking a career break, on maternity leave, or working temporarily in other areas. It does not provide access to any journals or Society grants and awards.
- > **Honorary** membership of the Society is by election only and this honour is conferred on persons of distinction in the field of applied microbiology. Honorary Members have access to our online journals.
- > **Retired** is available to Full Members once they have retired from their employment. Retired Members are entitled to all the benefits of Full Membership except grants and access to the Society's journals.
- > **eAffiliate:** this category of membership is open to microbiologists residing in Band I developing countries and is free of charge. It is an online only membership and provides access to the eAffiliate bursary only.
- > **eStudent:** this category of membership is open to undergraduate students only. It is an online only membership and is free of charge. This category of membership does not provide access to the Society's grants or journals.
- > **Corporate** is open to all companies with an interest in microbiology. Corporate Members benefits include:
 - Quarter page advertisement in each issue of *Microbiologist* (which can be upgraded to a larger size at discounted rates).
 - The opportunity to publish press releases, company news, etc., in each issue of *Microbiologist*.
 - FREE banner advert on the Society website with a direct link to your company site.
 - Up to three Members of company staff attending Society meetings at Members' rate (this means a 50% discount on non-Member registration rate).

Join us!

You can apply for membership online (www.sfam.org.uk/join) or offline. To apply offline, please contact the Membership & Finance Co-ordinator, Julie Wright on +44 (0)1234 326846, or email julie@sfam.org.uk.

Membership CHANGES

We would like to warmly **welcome** the following new Members to the Society.

BURKINO FASO

D. Sereme/Compaore

ETHIOPIA

*G. Alemseged
K. Birhane
G. Gebremedhn
B. Gebremeskel
M. Gebretsadkik
A. Hailessassie
K. H. Mebrahtu
T. Mesfin*

GERMANY

P. Rojas

GHANA

D. L. Narh Mensah

IRELAND

*I. Alamgir
J. McGrath
K. Murray
N. L. Pomeroy*

IVORY COAST

A. M. Alloue-Boraud Waze

NIGERIA

*I. Bilya
J. O. Ehichioya
F. N. Ezugworie
A. S. Nwankwegu
O. Ogunsade
A. A. Ojiaku
T. Ojiezeh
C. B. Okeke
U. J. Okobo
O. C. Otitoola*

SOUTH AFRICA

O. Adebo

THE NETHERLANDS

K. Venema

UK

*S. Abdullah
M. Abhishek
Y. R. Adebola
F. Afolabi
F. Ahmed
Q. Ahmed
S. Akther
A. Alaizoki
P. Almeida Powell
M. Al-Sabah
L. Altyeb Alhag
D. Alvarez
F. Alzahrani
T. Amande
A. Aryal Timilsina
P. L. Asquith
C. Atkinson
M. B. Azhar
Y. Badreldin
N. Barratt
N. Begum
G. Billenness
S. Bitou
M. Blair
C. Bourner
C. Buba
A. G. Buddie
P. Carr
R. Chai
P. Charley
F. M. A. Charlton
B. Chen
E. V. Clark
M. Connor
D. Cooke
C. J. Cooper
D. Day
A. Day
G. Divekar
J. Dobson
I. Douterelo Soler
L. Dunn
K. Emami
J. Fernandez-Piquer
W. Ferreira
G. Foot
L. Fossier
I. Garcia
V. Gill
A. Gillett
Y. Guemar
T. Guest
P. Hadjibagheri
A. Hale
N. Harris
L. Harris*

M. Hernandez Madrigal

*G. Hinch
S. Hosseini
S. Huang
R. Hull
B. Ibitoye
A. Ilyas
J. Irish
F. Islam
L. Issa
L. A. Jaafari
X. Jiang
R. S. Kanu
O. Kelly
A. Keswani
P. Khondkar
D. P. Koenig-Green
S. I. Koshin
A. Kozma
J. Kreitz
A. E. Kubala
R. Layton
R. N. Littlefair
N. H. Mansfield
C. Marshall
A. Martioucher
R. J. Masanga
E. Massone
E. Matthys
R. McBride
J. McDermott
J. N. McFadzean
O. J. McInness
R. Milner
J. Moran
C. M. More
T. Mugobhi
M. W. Naeem
R. Newbold
U. H. T. Nguyen
A. Nguyen
N. Nguyen
S. Nisa
E. M. Nwakeze
J. Nzakizwananyo
A. Ogbechie
S. Olaifa
O. K. Olarinoye
I. O. Olorunda
E. Paines
P. V. Partha Devanthi
R. Patel
D. Pearce
K. W. Pemberton
D. Perrey
N. M. Phan
A. Philpott*

*A. Pluqualec
K. D. H. Porter
D. Prudham
Y. Qi
G. Rachon
N. M. Rahimi
D. Rencricca
L. T. Rosa
F. Rozendo
E. C. Scotney
A. Sobolewska
A. J. Spicer
R. L. Spratt
L. Tetlow
S. Thomforde Kitson
K. Topley
T. T. T. Tran
S. Umaru
K. Usachova
C. S. H. Uysal
B. Vale
J. Vuong
J. Waack
J. Warden
P. Weldrick
J. Whitewood
C. Williams
R. Z. Williamson
K. Wilson
A. Wilson
K. Wray
S. Wylde
I. Yelastou
M. Zahid*

USA

*M. L. Chikindas
A. Frick
Y. Huangfu
C. Jackson
K. H. Leew*

DEATHS

We were saddened to learn of the deaths of the following Members of the Society:

*N W Le Roux,
John R Postgate,
Edmund Pike and
Sydney Neill.*

Short-term effects of biochar augmentation of a naphthalene-enriched microbial community

Following the successful completion of my undergraduate studies at Teesside University I was fortunate to be awarded the SfAM Students into Work Grant. The grant enabled me to extend my research experience by studying the effects of biochar augmentation on an indigenous microbial community enriched from contaminated sediment.

Scientific innovation and industrial development are often symbiotic; yet, industrialization has not been without consequence. Mankind is only recently facing the repercussions of rapid industrialization. The enormity of this task in a world reliant on fossil fuels is incalculable; the ongoing recovery and refinement of carbon-based fuels is often problematic and hazardous (Neff, Stout & Gunster, 2005). Sites concerned with the refinement of fossil fuels inevitably have residual consequences long after cessation of processing. Polycyclic aromatic hydrocarbons (PAHs) are prominent soil contaminants of such sites, resulting from the inefficient burning of fossil fuels. PAHs broadly encompass the most predominant organic pollutants associated with heavy industry; varying in severity and remaining *in situ* following their release.

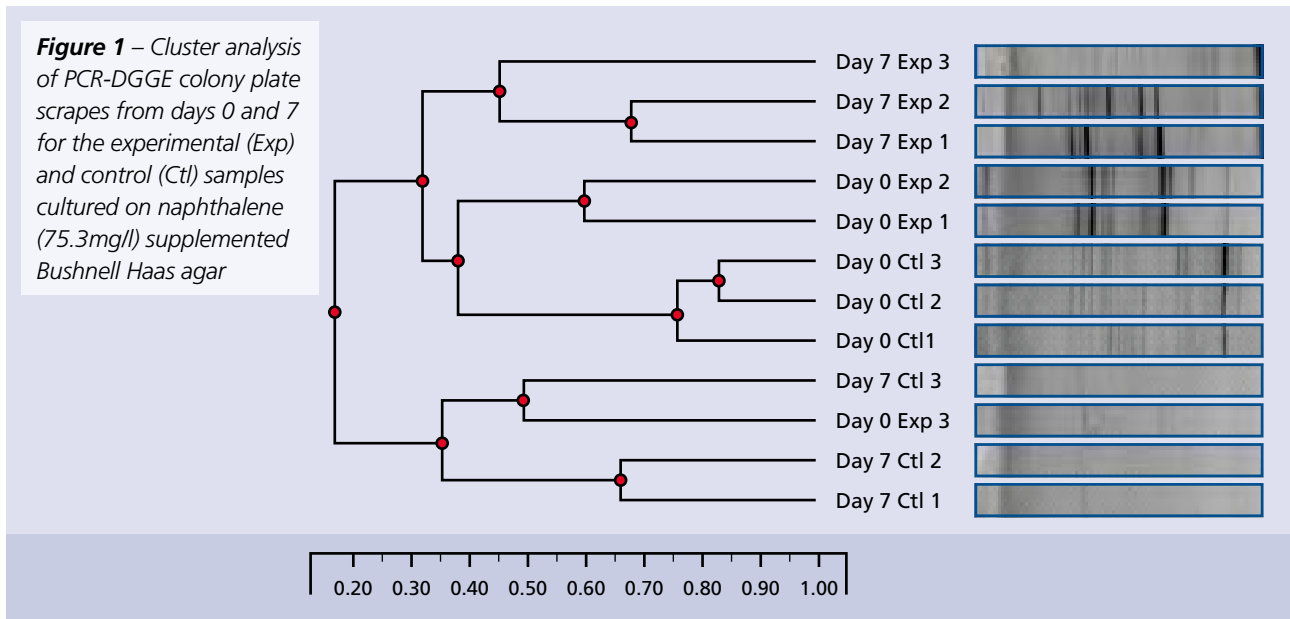
Naphthalene (C₁₀H₈) is the simplest known PAH. This strongly ordered crystalline compound is comprised of two fused benzene rings. PAH retention by soils is cause for concern due to its carcinogenic and mutagenic potential, hence its presence in brownfield sites is an inhibitory factor for their redevelopment.

Biochar is a re-emerging soil augmentation methodology. Derived from the pyrolysis of biological matter, it has been used for centuries to improve soil quality. Its effectiveness varies depending on the soil type and biomass from which the char originated. Biochar treatment of soil differs from conventional slash and burn, whereby soil retains ~3% carbon. Modern biochar treatment aims to maximize the sequestration of carbon by soil whilst minimizing ash deposition. A surge in global research now offers insight into the application of bespoke biochar to suit different soils/ environments, with promise to improve soil structure, productivity and water retention (Atkinson, Fitzgerald and Hipps, 2010).

Thus the current research builds upon two previous studies funded by SfAM using model bacterial communities to assess biochar impacts. We measured the taxonomic variations following biochar augmentation of a naphthalene-enriched microbial community from a former gasworks.

Microbial diversity was monitored by destructive sampling of triplicate microcosms containing 990µl Bushnell Haas Broth (Ramsay *et al.*, 2000), 100µl sediment enrichment and a naphthalene overlay. Microcosms were created for experimental (with 5% (w/v) biochar; +BC) and control (no biochar; -BC) samples. Destructive sampling of microcosms on days 0, 2, 4 and 7 permitted serial dilutions in saline (10⁰ – 10⁷) and spread plating (100µl) Bushnell Haas agar

The grant allowed me to extend my research experience by studying the effects of biochar augmentation on an indigenous microbial community enriched from contaminated sediment



augmented with naphthalene (75.3mg/l), 2,3-dihydroxynaphthalene (75.3mg/l), 1,2-dihydroxynaphthalene (75.3mg/l) and salicylate (75.3mg/l) using standard aseptic techniques. Plates were incubated at 25°C for seven days followed by CFU counting and scraping of viable colonies. Colony scrapes were re-suspended in 125µl sterile deionized water and used for direct PCR of the V3 region of the 16S rRNA gene (Manefield *et al.*, 2002). Successful amplification was confirmed by 1% (w/v) agarose gel electrophoresis using SYBR Safe. The remaining 15µl amplicons were loaded on 10% polyacrylamide gels with a 40–65% denaturing gradient and electrophoresed for 18 hours at 110V and 60°C. Subsequent band analysis through Phoretix 1D software (TotalLab) and application of the Shannon Weiner Index (SWI; Martin, 2002) were used to analyse shifts in diversity.

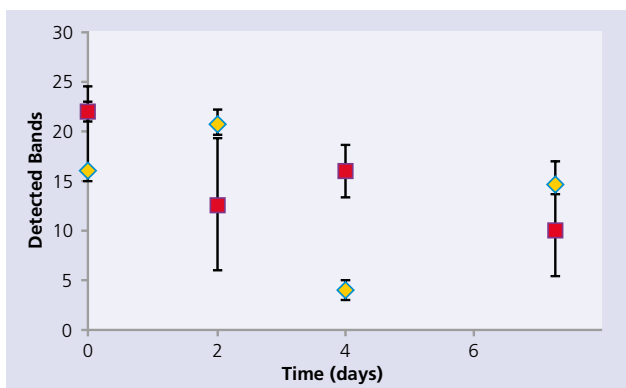


Figure 2 – Band richness detected following DGGE analysis of colony plate scrapes for the control (■) and experimental (◆) microcosms on days 0, 2, 4 and 7. Data shown is mean of experiments performed in triplicate ± standard deviation and represents bands present on naphthalene (75.3mg/l) supplemented Bushnell Haas agar only

This study's objective was to determine the effect of biochar on the structure and diversity of a microbial community enriched on naphthalene using sediments from a gasworks with a long history of PAH contamination. Soil health may be determined by its pH, water retentiveness and the diversity of resident microbiota (Doran and Zeiss, 2000). A rich microbial community may contribute to an improved soil structure and nutrient cycle thus, potentially, affecting soil water retention and productivity. Data presented within this short report represents the taxonomic variations for naphthalene-supplemented experiments only.

Despite its known limitations, e.g., DGGE bias where the composition of the bacterial assemblage may be underestimated if only highly amplified bands are prominent, the 16S rRNA gene-based analysis permitted specific analysis of changes to bacterial enrichment with naphthalene-augmented microcosms, in the presence and absence of biochar (Figure 1). SWI (H') of DGGE data (not shown) elucidated trends in diversity between experimental treatments over time. In the presence of biochar the initial biodiversity (H' =2.66) was the highest, but decreased to H' =1.89 by day 7. This downward trend was not mirrored by the control microcosms, which exhibited little change in biodiversity between day 0, H' =2.31 and day 7, H' =2.15. All microcosms showed a considerable decrease in biodiversity on day 4 (H' = 0.57). Band diversity was significantly lower on day 4 (Figure 2) dropping from an average of 21 bands to 4 hence a similarly reduced SWI (H' =0.57). This trend was also matched by a decrease in band richness (Figure 2) and the CFU counts (data not shown).

Biochar has been accepted by the United Nations as a means of alleviating climate change and organic waste produce whilst increasing crop yields (Tenenbaum, 2009). Therefore, biochar-mediated soil treatment offers a tangible means of tackling the increased

demand for land development and enhanced food yield at a time when the global population is rising and resources are diminishing. Nonetheless, its impact on the indigenous microbial communities merits further detailed research.

This SfAM funded work demonstrates how biochar supplementation may negatively impact ecosystem biodiversity. It is part of ongoing research into the potential of biochar as a multifaceted remediation technique. The work has expanded my experimental skills range providing valuable experience to assist in my progression to graduate employment. I would like to thank the SfAM and the microbiology staff of Teesside University, particularly Dr Komang Ralebitso-Senior for her continued support and encouragement.

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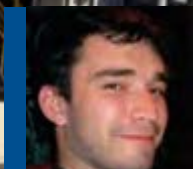
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Stephen Anderson

Petrochemical plant in The Kingdom of Saudi Arabia. Image: Secl CC BY 3.0 via Wikimedia Commons

TYPE 1 DIABETES: a gut microbiota perspective

Am I eligible – can I apply?

It is not only our Student Members who require our help. Senior microbiologists often find difficulty in funding attendance at meetings. If you are in this position you are eligible for this fund.

For further information visit
www.sfam.org.uk/presidents-fund

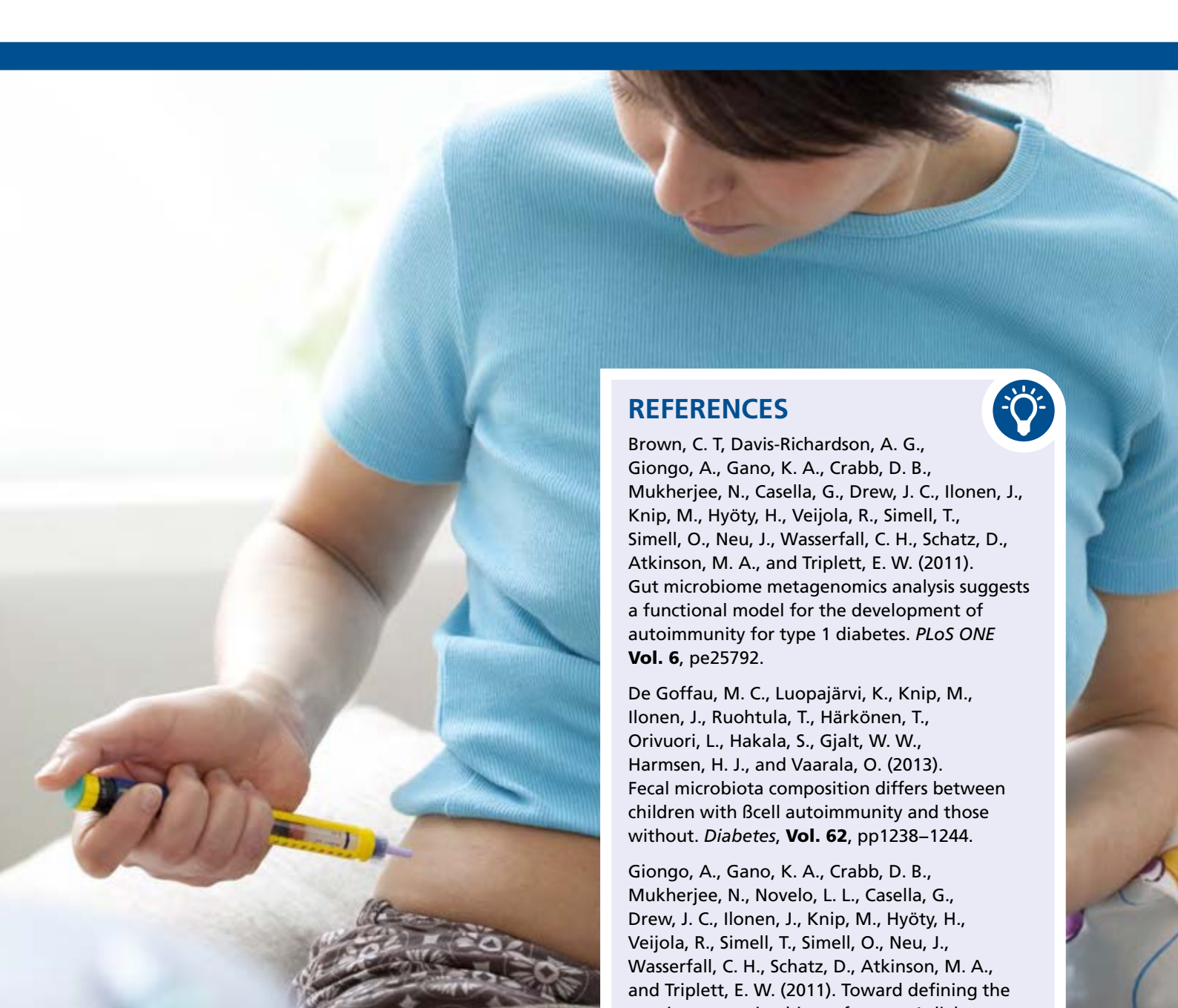
Type 1 diabetes (TD1) is considered an inflammatory disease that affects the correct activity of pancreatic beta cells. The events that trigger TD1 are still not clarified, though in the development of the disease, environmental and genetic factors have been considered. In fact, the incidence of TD1 in children during the last decades is increasing. However, significant geographical differences are observed, for instance the highest prevalence rates (estimated cases/thousands) of TD1 in childhood in 2010 were observed in Europe (112k) and in the Southeast Asia region (113.5k), in contrast to the Western Pacific region (30.5k), although the childhood population in this region is larger. In Europe, the highest incidence rates (cases per 100,000 population per year) in 2010 in children with ages 0–14-years-old were observed in Finland, reaching 57.4, followed by Sweden and Norway with 41.0 and 27.9, respectively, in contrast to Italy with a childhood population larger than Finland, that reached an incidence rate of 8.4 (Soltesz *et al.*, 2011).

Evidence suggests that
gut microbiota plays a
substantial role in the
development of (diabetes)

As highlighted by Knip and Simell (2012), these discrepancies can hardly be explained only by an increase in genetic disease vulnerability, but contributions from the environment and changes in lifestyle must certainly be considered. Among several factors, a significant body of evidence suggests that gut microbiota plays a substantial role in the development of the disease. The first report on the alterations of the gut bacteria in children that showed a high risk for the development of autoimmunity and TD1 evidenced that the microbiome of children predisposed to autoimmunity is unstable and the diversity is lower, in comparison to healthy children (Giongo *et al.*, 2011). The functional analysis of the altered bacterial communities in an autoimmunity state revealed a microbiome characterized by a poorer functional diversity, suggesting dominance of anaerobic fastidious bacteria and a significant increase in *Bacteroides*, *Veillonella* and *Alistipes* populations in contrast to healthy children who showed higher populations of *Prevotella* and *Akkermansia* (Brown *et al.*, 2011). Lower levels of *Bifidobacterium adolescentis* and *B. pseudocatenulatum* were also coupled to the autoimmunity state (de Goffau *et al.*, 2013). In clinical TD1 children the decrease in *Bifidobacterium* and *Lactobacillus* was linked with the plasma glucose value in contrast to the glycated haemoglobin (HbA1c) for which an association with the increase in *Clostridium* population and the reduction in the Firmicutes/ Bacteroidetes ratio was found (Murri *et al.*, 2013).

The differences in the dominant bacterial genera in combination with functional level description suggest an enhancement of butyrate production in the healthy microbiome, while an autoimmune microbiome is characterized by producers of short chain fatty acids (SCFA) distinct from butyrate such as propionate, acetate and succinate. A lower butyrate production will result in the disturbance of intestinal barrier functions due to a deficient mucin synthesis and a decrease in tight junction setting (Brown *et al.*, 2011).

The evaluation of changes in the gut microbiome in an autoimmunity condition clearly evidences a set of



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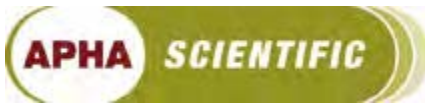
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modifications to the microbiome composition, but the microbiome action particularly linked with the destruction/protection of pancreatic beta cells still remains to be elucidated. Proteomic and metabolomic approaches would help to fill the gaps on how the host metabolism-immune system axis is affected by the changes in the microbiome. Another important question that needs to be answered regards the consistency of the alterations (composition and functionality) of the gut microbiome of TD1 children in different geographic regions. Understanding of the role of the gut microbiota on the development of TD1 is in its infancy and a significant body of work needs to be undertaken before we can intervene in the gut microbiota composition in order to gain protection and avoid TD1.

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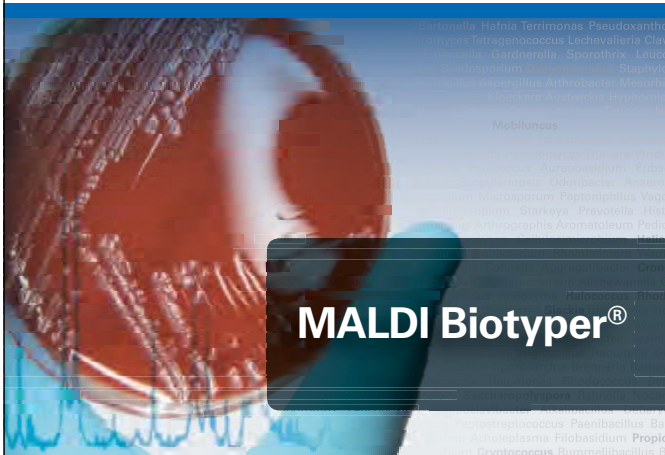
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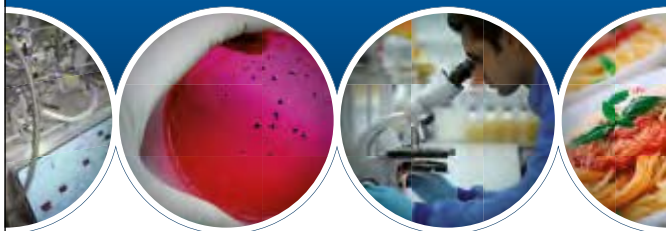


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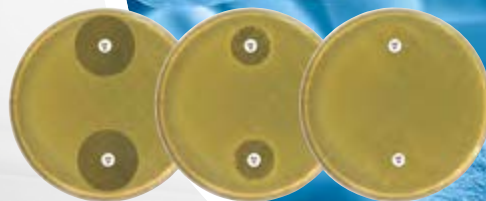
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SAS Air Samplers – A selection of robust, reliable air samplers designed for specific environmental monitoring purposes, including portable hand held units, a compressed air sampling device and an isolator specific unit. Cherwell's air samplers are easy to operate and use readily available contact plates avoiding costly, specialist consumables. Alternative Petri dish versions are also available.

Cleanroom Bio-decontamination Solutions – Suitable for use in pharmaceutical cleanrooms and other critical areas, the combination of dry fog technology and cold sterilants ensures effective and efficient bio-decontamination for surfaces, confined spaces and cleanroom suites. The range incorporates the highly effective Minncare® Dry Fog 2 and Mini Dry Fog systems plus Minncare and Actril Cold Sterilants.

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Clinical Anaerobes: Educate your staff

Now in its second print run, the publication '*An Introduction to Clinical Anaerobic Microbiology*' has proven to be a valuable educational tool. A general day-to-day reference tool, it is also being used to educate and train less experienced members of staff, providing them with an easy-to-use guide to consult as required.

This guide enables the reader to isolate and identify 12 commonly occurring clinically important anaerobic bacteria. It is printed on waterproof, tear-resistant synthetic paper to withstand regular use at the bench and is illustrated with stunningly detailed colour photographs and attractive reference tables.

Designed to fill a gap in the practical reference materials currently available to support clinical laboratory practice for microbiologists, the publication was written by Professor Michael W D Wren, MBE FIBMS, former Consultant Biomedical Scientist in the microbiology department, University College Hospital, and visiting Professor at The University of Westminster. With a foreword by Professor Brian Duerden, Emeritus Professor of Medical Microbiology at Cardiff University and input from Dr Don Whitley, Chairman and founder of DWS, considerable expert knowledge has been combined to make this the most up-to-date reference guide for the modern clinical laboratory. Images were kindly supplied by the Anaerobe Reference Unit, Cardiff.

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Lab M adds Cronobacter Sakazakii Isolation Medium to Pinnacle™ poured plates range

20 January 2015; Heywood, UK: Lab M's fast-establishing range of Pinnacle™ pre-prepared agar plates now includes Cronobacter Sakazakii Isolation Medium (CSIM) for added convenience when testing infant milk products.

Cronobacter sakazakii (formerly known as *Enterobacter sakazakii*) is a member of the Enterobacteriaceae family and has been associated with serious outbreak infections in neonates fed on infant formula milk. Pinnacle™ CSIM uses Lab M's established Harlequin™ CSIM (ISO). This media formulation is currently recommended as part of the protocol under ISO/TS 22964:2006(E) for the isolation of *Enterobacter sakazakii* from milk and milk products.

C. sakazakii appears to constitutively express high levels of α -glucosidase, which hydrolyses the chromogenic substrate 5-bromo-4-chloro-3-indolyl- α -D-glucopyranoside present in the medium, producing green to blue-green coloured colonies. The combination of sodium desoxycholate, crystal violet and elevated incubation temperature produce a very selective and specific medium. *Non-Enterobacteriaceae* may appear colourless or violet (due to their inability to hydrolyse the chromogenic substrate) or are inhibited by the selective components and incubation temperature.

For further details of Lab M's Pinnacle range of convenient ready-to-use plates and the company's extensive DCM portfolio, visit: <http://www.labm.com/pinnacle/>

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Leatherhead Food Research: Food Safety & Product Integrity

Leatherhead's food safety portfolio provides a comprehensive range of products and services to help food and drink companies maintain the highest possible standards of safety and stability in their products. We provide a broad array of microbiological food testing, analysis and consultation covering microbiological food safety, training and advice, and bespoke testing, if required.

Our main focus areas include:

- Shelf life and challenge testing (including *Cl. botulinum*)
- Microbial inactivation kinetics (to aid food processing)
- Molecular diagnostics including speciation of bacteria, yeasts and moulds, and horsemeat
- Enteric virus research and detection
- Antimicrobial screening and alternative natural preservation technologies
- HACCP training and studies including troubleshooting and audits
- Biofilm study and control
- Method development and validation of rapid detection methods
- Validating and advice on cleaning and disinfection procedures

Additionally, our Food Safety & Product Integrity Department is supported by expertise from across Leatherhead. For example, we can check the safety/shelf

life of a reformulation, whilst ensuring that it meets the required sensory and nutritional properties. We can also provide regulatory advice so that the product meets the required legislation for the countries in which it is sold.

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Medical Wire has added **Salmonella Isolation Transwab®** (Product code MW572) to its range of swab based devices for the monitoring of environmental and food contact surfaces in food manufacturing facilities. Based on the familiar Transwab® design, the kit is simple and safe to use, and includes a swab for surface sampling, together with a transport tube of bright red gel medium. The swab is used to sample the surface being tested. It is then placed in the red medium and sent to the laboratory. Upon incubation at 37°C, if *Salmonella* is present the medium will become deep black, initially around the swab bud, but spreading throughout the remaining medium. The colour change will normally be visible within 24 hours.

A recent trial undertaken at Campden BRI showed that *Salmonella enteritidis* and *Salmonella typhimurium* isolates from a range of foodstuff sources would give a positive reaction within 24 hours, while there were no false reactions with *Staphylococcus aureus* or *Escherichia coli*.

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Research project explores probiotic potential

NCIMB recently collaborated with leading probiotics manufacturer Probiotics International Ltd (PIL) in a UK Technology Strategy Board (TSB) funded research project that explored the potential of new microbial genera in the production of probiotic products.

Probiotics are live microorganisms, which when taken in adequate amounts can confer health benefits. As understanding of the complexity and importance of gut flora has increased, so has interest in probiotics for both human and veterinary use.

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However, to date, most of the species used in commercial probiotic products have come from just two genera of bacteria: *Lactobacillus* and *Bifidobacterium*.

This research project aimed to increase the health benefits offered by microorganisms by bringing NCIMB's world class culture collection together with PIL's extensive experience in probiotic product development, to explore the potential of new genera in the development of novel probiotic food supplements and functional food products.

The NCIMB culture collection includes more than 8000 strains of environmental and industrially important bacteria, as well as plasmids and bacteriophages.

The project was successful in identifying a number of bacterial strains with interesting properties, which have not previously been used in the development of probiotic products.

For more information about collaborating with NCIMB, contact Dr Carol Philips c.philips@ncimb.com

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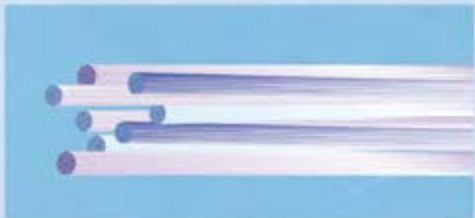
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