Microbiologist

The magazine of the Society for Applied Microbiology ■ September 2004 ■ Vol 5 No 3 ISSN 1479-2699

Tenants of the last 1.5 metres

David Reynolds looks at the varying properties of 'good bacteria'

ALSO IN THIS ISSUE:

Lichens, lichenometry and global warming

Winter Meeting 2005 preview

Suspicious packages: we review the increasing controls on the supply of dangerous pathogens

SfAM/SGM **Microarray** workshop

Peter Silley looks at Science funding

Composting: not just a load of old rot

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Microbiologist

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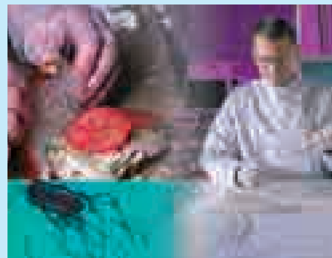

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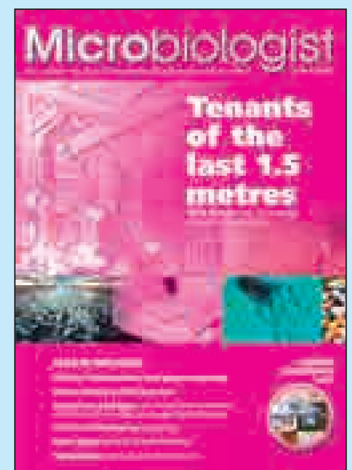

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The editor is always looking for enthusiastic writers who wish to contribute articles to *Microbiologist* on their chosen microbiological subject. Email: harperlv@aston.ac.uk

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Website: the society website is a timely source of up-to-date information on all Society matters and maintains a comprehensive archive of articles and reports on a variety of microbiological topics.

www.sfam.org.uk

Editorial

It may come as somewhat of a surprise that this is my final editorial for the *Microbiologist*. You'd think I'd know better by now but the Society is not about to let me get off that easily and at the AGM in Cork my nomination as Honorary General Secretary was approved and I took up my new position. I'm very much looking forward to my new role within the Society but it is not without some sense of sadness that I hang up my editorial cap. On my office wall I have mounted the front covers of all the issues for which I have been editor and a few issues of *SfAM News* prior to that. It represents a pictorial time-line of the development of our Society magazine over the past two years and something of which I am personally very proud. But as with all ventures of this magnitude the success of the *Microbiologist* cannot be attributed to an individual and it would be remiss of me not to take this opportunity to say a few thank-yous. Firstly, to all the authors and contributors to the *Microbiologist* over the years, without you there would be no magazine and I am grateful for the time and effort you have put into providing quality articles and reviews for me to work with. Secondly, to my editorial assistant Anouche Newman without whom the *Microbiologist* office would grind to a shuddering halt. Finally, my thanks to Pollard Creativity who have, and I know will, continue to do an excellent job in the design and production of *Microbiologist*. Although saddened to pass the reins of the magazine over I'm delighted to be placing them in the hands of Dr Lucy Harper who I know will continue to steer *Microbiologist* on to future success. Good luck!

Thanks Anthony. As the new Editor of *Microbiologist* I thought it appropriate to include a photograph of myself. Partly so you can all picture who you're contacting when you inundate me with wonderful material for the magazine but also to ensure that you don't miss seeing a cheesy photo on the inside cover!

On a more serious note, I'm honoured and thrilled to be elected as Honorary Editor of *Microbiologist*. I shall endeavour to deliver as interesting, informative and useful a magazine as my predecessor, though I have to confess that I've had a lot of help in compiling this issue. As well as help from the previous editor, my colleagues here at Aston University have been a great source of

ideas and inspiration, so without wishing to over-do the 'thank-yous' appearing in this editorial, I must say a big 'cheers' to them all (you know who you are!).

We have quite a diverse range of topics appearing in this issue, from the many and varied properties of probiotics, to the use of lichenometry in dating. We're also re-running the 'travelling microbiologist' feature, so don't forget to take your copy with you wherever you go and email your picture to me to be in with a chance of winning!

Back to the subject of probiotics, I've recently been made aware of their properties on a (little too) personal level. I had occasion to visit my friendly neighbourhood GP recently who prescribed me with some flucloxicillin. As I was aware that one of the side effects of many antibiotics is that of a 'loosened stool', I thought I'd attempt to maintain my gut flora (and hopefully lessen the severity of this particular side-effect) by drinking a well-known brand of probiotic drink — which I have to say was very pleasant on the taste-buds. This phenomenon has been investigated previously, but I thought I'd put it to the test and I'm very happy to say that it worked! I imagined that the 'good' and 'bad' bacteria were having a bit of a tug of war initially, but after a day or so I felt no side-effect from the antibiotics whatsoever. I would certainly be happy to use this as a piece of circumstantial evidence in favour of their use in combating this rather unpleasant side-effect!

Well now, it just leaves me to say I hope you enjoy this issue. All feedback is much appreciated by myself and the contributing authors, so do write and let me know what you think of this, my first issue of *Microbiologist*.

society for applied microbiology (sfam)

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Don't inhale

FROM: Gary Hogben
SUBJECT: Sick Building Syndrome

Amongst an otherwise excellent article on Sick Building Syndrome, I must challenge Dr Deisingh's description of Legionnaires Disease as "transmitted by drinking water" and the accompanying suggestion that air-conditioning systems are not implicated in disease transmission.

The drinking water theory is a controversial one propounded principally by one centre in the United States. The article refers the reader to one website (Legionella.org) where this theory is taken as established fact and no attempt seems to have been made to establish the acceptance of this theory among the wider scientific community. The website does acknowledge that the majority of scientists did not accept the drinking water hypothesis, but further suggests that new research has proved the theory.

Most scientists working in the field accept that the disease is acquired through the inhalation of droplets deep into the lung. This is not to say that occasional cases may not occur by aspiration of drinking water from the alimentary tract into the lung. Nor does it mean that the majority view will not be proved incorrect in the future. However articles should recognise the scientific consensus on issues and should avoid propounding positions of limited support as the accepted view. Dissenting positions should be presented as such.

As for air conditioning systems not been the source of disease, there are numerous outbreaks linked to these systems. Even in the case of the defining outbreak in 1976, some cases were amongst people who had only been in the hotel lobby and some outside the building. The UK regulatory authorities work hard to prevent outbreaks of Legionellosis and have an Approved Code of Practice (L8) for prevention based on the consensus viewpoint. Promulgating the drinking water theory as the state-of-the-art undermines efforts at prevention in the UK.

On a lighter note, the referenced website does recommend it's readers to heat their domestic hot water systems to 140 degrees Centigrade to remove Legionella. To be fair, it does recognise that there is a risk of scalding at this temperature!

Canned Air

FROM: Chris Collins
SUBJECT: Sick Building Syndrome

The excellent paper on sick building syndrome in the *Microbiologist* reminded me of a meeting we had quite a few years ago about the design of a new laboratory. The architects wanted to give us air conditioning ('canned air'). But our Director held out for having the heating and ventilation under the control of the occupants, not according to the calendar, mechanical gizmos or the whim of a remote administrator. So we had radiators that we could turn on or off and windows that we could open at will. There was no SBS!

Ambiguous Acronyms

FROM: Mairi Hope
SUBJECT: Erratum

The article on DGGE in the June issue was entitled 'Density Gradient Gel Electrophoresis'. I would like to point out that, as described in the text, DGGE is infact an abbreviation of **Denaturing** Gradient Gel Electrophoresis and I thought that this should be brought to the attention of your readers.

Many apologies for the editorial error Mairi and thank you for pointing this out to us.(Ed.)

Impressed in the USA

FROM: Frank Poole
SUBJECT: *Microbiologist*

Just a quick email to congratulate you on the *Microbiologist*. I picked up a copy at the ASM and was impressed both by the scientific content and the look, so much so that I applied to join you! Well done guys! I look forward in anticipation to receiving the next number.

Brilliant Bugs!

FROM: Sue Catchpole
SUBJECT: Design-a-bug
I just want to express my thanks for the Design-a-bug competition in your excellent magazine. Not only did it keep 37 very lively children quiet for several days, it also stretched their imaginations and gave them a unique insight into the fascinating world of microorganisms.

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Answers to the June 2004 Crossword puzzle.

```
 THERMOPHILE    B    C       N
 W            STATIONARY     E
CONJUGATION     C N  P   F   U
E     E         I O  SPIRALS T
L  SPIROCHETE   L R  U   C A R
L   S O        FLAGELLUM T P O
M   Y B         U A  E   O R P
E NUCLEOID COCCUS N      R O H
M   H             I    V S P I
BINARYFISSION   S C    I   H L
R   O   E       U      B   Y E
A   PLASMIDS    R   NUTRIENTS 
N   H C I       V      I   E C
E  PILI PHOSPHOLIPIDS PORES  Y
    L D E       V        N   T
 HETEROTROPH GREATER  F  D   O
 A    P M     I L   ANAEROBE P
 LAG  H E     B       C  S   L
 O    I A  M  OPTIMUM U  P   A
 P    L B  I  S       L  O S S
 H    E L  C  O MOTILITY R P M
 I      E  R  M U     A  E I  
 L M S     O  E RNA  ATP S R  
MESOPHILE  M  S E     I    I  
 S R A     E    I     V    L  
   P POPULATION N    CELLWALL 
   H E   O E               U  
   O   LOGARITHMIC   D ENZYMES
   L       S         N        
  POSITIVE  POLYSACCHARIDES   
   G
   Y
```

Word Puzzle

We were inundated with entries to our June **Crossword puzzle**, and I'm pleased to say that the lucky winner of a £30 book token was **John Caplin** of the University of Brighton. Congratulations to you!

This time we want you to re-arrange the boxes below to reveal a phrase! The wining entry, which will be picked from my lab coat pocket, will win a £30 book token. Please send your entries to the Society office by the 24th September to be in with a chance!

OGI	TUR	CUL	MIC	LWA	YS	IOL	ED!
STS	E A	AR	ROB				

A £30 book token is waiting for the person whose entry we receive first! The closing date for entries is **Friday 15th October 2004**. The answers will appear in the December 2004 issue of ***Microbiologist***.

Name: ______________________________

Address: ______________________________

Simply photocopy this page and send it to: 'Microbiologist Word Puzzle', Society for Applied Microbiology, The Blore Tower, The Harpur Centre, Bedford MK40 1TQ, UK. **Remember, you could win a £30 Book Token!**

New Members

We would like to warmly welcome the following **new members** and hope that you will participate fully in the activities of the Society.

Argentina

Dr G Font de Valdez

Australia

Mr G C Knight; Ms C M McAuley

Finland

Dr M Saarela

Greece

Miss S Iconomopoulou

India

Mr K Abdul-Muthalif; Dr D Chandra; Dr R Singh

Ireland

Dr T Beresford; Dr R Donnelly; Dr A Kelly; Ms M C Rea; Dr R Sleator

Japan

Dr Y Konagaya

Nigeria

Prof. O Famurewa; Mr F McFarlane

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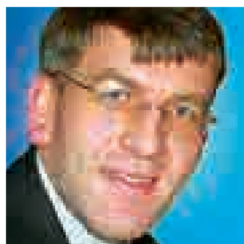
Mr D Garcia; Mr M Hassani; Mr D Sanz; Miss R Virto

United Kingdom

Miss J Angell; Miss C Ball; Dr P A Burgess; Mr C Campos-Rio; Miss J Caubillua-Barrow; Miss R Causey; Miss M Chacon-Romero; Mr J T Childers; Mr C Creen; Mr P O Curley; Dr S Dinsdale; Mr J Donnelly; Mrs G M Dreczkowski; Miss K Fletcher; Mr G J Gallimore; Mr E Giotis; Miss V Hamilton; Miss A Henein; Mr A P Hockin; Dr J V Javid; Mr A Jonas; Miss M D Jones; Dr K Jordan; Mr P S L Kwan; Ms K McCaughan; Dr R McHugh; Miss S McKenzie; Mr S McPeake; Miss S Noor; Dr D O'Connell; Mrs F Pesan; Miss H Pearce; Miss D Portner; Miss C L Rowe; Mr M A Saeed; Miss C Sexton; Miss J Shackleford; Miss G Smith; Miss M Tourna; Dr M Upton; Mrs D Zakari

USA

Ms N Campos-Gorgona; Dr C Guzman-Verri; Dr K L Kauers; Prof. J R Leadbetter



Dr Peter Silley focuses on the major funding descisions regarding science and innovation announced in the Government's early summer Spending Review

Looking back I note that this column has been a veritable "mixed bag" without any semblance of continuity and so as I enter into my last year as President of the Society I guess that there is no reason to change! It is appropriate, however, before I begin my ramblings to pay tribute to a wonderful Summer Conference in Cork in July past. For those of you who were not able to attend you missed a great meeting and if this is a sign of things to come you must already get Summer 2005 in Brighton into your diary! Thank you to all those who played their parts in a truly memorable meeting, there was excellent science and great Irish hospitality and we look forward to having the opportunity to repay that when you all come to Brighton!

I also want to take this opportunity to welcome our new Honorary Vice President, Dr Margaret Patterson. Margaret is no stranger to most of our members having been the Honorary General Secretary of the Society since 1999. I am delighted that Margaret was elected as Honorary Vice President at the recent AGM in Cork. For those of you who are unaware Margaret is Principal Scientific Officer and Project Leader in the Food Microbiology branch of the Agricultural, Food & Environmental Sciences Division, Department of Agriculture for Northern Ireland, and is also a Reader in Food Science, at The Queen's University of Belfast. Margaret has an international reputation in food irradiation and high pressure treatment of foods. We will be working together over the next 12 months to serve and promote the Society as we seek to reflect the voice of applied microbiology.

In this issue I would like to particularly focus on the major funding decisions regarding science and innovation which were announced in the Government Spending Review in early summer and on the presentation of the 10-year government science and innovation investment framework. I am indebted to Mike Withnall at the Bioscience Federation for preparing excellent briefing notes which have helped me to focus my thoughts on these important issues.

Fundamental to the government framework is the ambition that within the UK we develop world class research at the UK's strongest centres of excellence. In order to achieve that ambition it has been stated that there needs to be;

- Greater responsiveness of the publicly-funded research base to the needs of the economy and public services
- Increased business investment in R&D and increased business engagement in drawing on the UK science base for ideas and talent
- A strong and more responsive supply of scientists, engineers and technologists
- Sustainable and financially robust universities and public laboratories across the UK
- Confidence and increased awareness across UK society in scientific research and its innovative applications.

We would all applaud these commendable objectives but how will it happen and how as microbiologists can we play our part? The government's long-term objective for the UK economy is to increase the total spent on research and development from 1.9% to 2.5% of GDP by 2014. To achieve this target requires substantial growth in business R&D, which in turn requires a similarly significant growth in the underpinning investment in the public science base.

Increased funding through the DTI and the DfES at an average annual rate of 5.8% between 2004/5 and 2007/8 was announced in the Spending Review. The government intends to increase investment in the public science base at least in line with the trend growth rate of the economy through the 10-year period. But it notes that if the overall level of R&D in the economy is to reach 2.5% of GDP by 2014 it will require a challenging average rate of growth in private and public R&D spending of about 5.7%.

The government appears to differentiate two types of institutions:- a small core of leading world-class research universities and a larger number of universities with broader roles that have some pockets of excellence and maintain competitive pressure for funding on the upper tier. The funding system should enable promotion to, and relegation from, the top tier over time. The government wants to ensure the sustained health and global competitiveness of that small core, which performs a national role in making the UK a partner of choice for mobile investment and talent.

It is clear that fundamental to government plans is the need to increase the level of business investment in R&D. The aim is to increase the level from 1.25% of GDP to 1.7% by 2014. The government will commit additional resources through to 2007/8 to help bridge the funding gap between commercial application of new technologies and the underpinning research. Funding through the DTI Technology Strategy for collaborative R&D and knowledge transfer networks will rise to at least £178 million by 2007/8, and the DTI will work closely with business to pull through and exploit technologies from the UK and international research base. Universities will be encouraged to expand the commercialisation of their research and collaborative working with business through an increase in size of the Higher Education Innovation Fund to £110 million a year by 2007. Public sector research laboratories will be similarly encouraged and funded to develop their own knowledge transfer missions. Regional Development Agencies will continue to build their capacity to promote science and innovation, and each will have a Science and Industry Council by the end of 2004.

I cannot speak highly enough of the efforts of government through my local

DTI offices in Yorkshire and Humberside over recent years and I welcome any initiative to strengthen the role of regional offices as they are no doubt best placed to assess the merits of local businesses. I am not sure, however, if my experience is necessarily translated nationwide. Government must ensure, however, that the bureaucracy related to technology funding does not strangle new initiatives. We all realise the need for accountability but if the enterprise of small companies is to be harnessed then it must be realised that these companies do not have the infra-structure to handle complex paper trails. It is also crucial that we create appropriate business models that allow such small companies to work with universities and public sector laboratories without introducing severe financial penalties at the outset. Small privately owned companies are rarely cash rich and should not be seen as a short term means of funding academic research. Success should clearly be shared appropriately, but I believe there are too many instances where small companies are unable to harness the expertise available within the academic sector because of excessive financial demands being made at the outset.

Within the government framework considerable attention is rightly given to issues surrounding public confidence in science. It is acknowledged that the public needs to have confidence in the ethical and regulatory framework within which scientific advances are made. New schemes to enhance dialogue between the public, the science community and policy makers are proposed. It has been announced that the OST's Science and Society expenditure will increase from £4.25 million a year in 2005/6 to over £9 million a year by 2006/7. Clearly these are areas in which the Society is actively involved, particularly through MED-VET-NET.

The Bioscience Federation, of which we are members, has responded to this welcome framework. I will draw your attention to just a few of the comments. The programme is clearly intended to benefit the UK economy first and foremost, and in doing so it will place the science base in a much more sustainable position. The substantial investment in recent years, and continuing into the future, is very welcome, but it may well be accompanied by more central control and planning, more administration for academics, and a leaning towards shorter-term business orientated research. Each of the key ambitions is accompanied by a set of performance indicators and targets, for instance; the experience of education and health is that unless great care is taken; setting targets increases wasteful bureaucracy and skews institutional behaviour in ways that had not been intended. The government says that it will "monitor and publish a range of input, output and outcome data on a regular basis. Future public spending decisions will be made in the context of demonstrable progress and engagement with private sector stakeholders in business and the charity sector".

The report rightly identified the need to maintain a flow of talented young people into science and engineering and devoted considerable space to this, but without making any very novel recommendations. A working group has been established by the Biosciences Federation and will address the question of what core science should be included in biology curricula from primary through to university level, in order to free up time for practicals, and broader discussion of societal and ethical issues and relevance to everyday life.

Science is very much on the agenda of government and for that we must be grateful, we need to roll up our sleeves and play our part in communicating the importance of what we do.

Peter Silley

Your Society needs YOU!

SfAM is the voice for Applied Microbiology and often gets requests by journalists for background briefings or information.

Are you interested in being part of a small group which will brief the media about applied microbiology?

If so please contact **Nigel Poole** in Public Affairs at the Society Office. Phone **01344 750248** or email him at: **Sekona@btopenworld.com**

Travelling Microbiologist

Katie Hopkins writes:
"After seeing your editorial in the last Microbiologist about taking the magazine with us on holiday I took my copy with me to Mexico and the photo is from the top of 'El Castillo' at Chichen Itza. I don't look too happy to be there because the sides were rather steep and a lot of people were at the top jostling to have their photo taken where I was standing!
Is my Microbiologist the furthest travelled from Bedford?!?"

Yes Katie, your copy has travelled the furthest from the Bedford office and as such you have won a copy of the book *'Microbial Genomes'* edited by Claire M. Fraser.

The summer holidays are almost over, but I'm sure there are some of you who are taking a late break in the sun. Remember to take your copy of *Microbiologist* with you and you too could be in with a chance of winning a prize (I'm not sure what the prize will be yet, but it WILL be worth it)!

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Symposium Series — **14-29** (1985-2000)

Journal of General Microbiology — volumes **77-127** (1973-1981)

If you would like these journals please call Roy Smither on **01784 244098** or email him at: **roy@rsbiotech.freeserve.co.uk**

In Memoriam: Dr A H L Chamberlain (1947 - 2004)

It is with deep sadness that we report that Tony Chamberlain, Convener of SfAM's Bioengineering Group, died on March 5th after a long battle against cancer. Tony studied for his BSc and PhD at the University of Leeds where his major interests were in botany and he retained a deep love of and considerable knowledge about plants throughout his life. Under the guidance of Professor Len Evans, and the influence of Professor Irene Manton FRS, Tony also gained considerable practical expertise in both light and electron microscopy.

Tony was always ready to apply his academic skills to technical applications. His interests in the mechanisms of attachment of algae, initiated at Leeds, were devoted to the study of the fouling of marine surfaces, such as ships, while he worked as a postdoctoral fellow at Portsmouth University and where he further developed his skills in the study of the ultrastructure of biological material.

Tony was appointed as a lecturer in Microbiology at the University of Surrey in 1977 and quickly made his mark as a teacher and researcher. He was never happier than when taking students off the campus on field trips, visits to water works, sewage works and sites associated with his advanced courses on biodegradation and biodeterioration, his favourite being the Mary Rose at Portsmouth! He was appointed Senior Lecturer in 1989.

His research covered fundamental studies on the structure and taxonomy of groups of protists, such as the thraustochytrids and coccolithophorids, and applied studies on the mechanisms of attachment of microorganisms in the context of fouling and the contamination of food preparation surfaces and medical equipment. He believed passionately in the value of interdisciplinary studies and sought the assistance of physical chemists, spectroscopists and engineers in all his work.

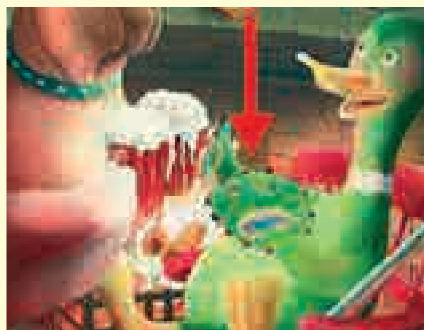
There are many who have good reason to be grateful to Tony for his professionalism and dedication to a diverse range of jobs to be done, but an even larger number of people will be thankful to have known him as a friend.

Tony was buried in Brookwood Cemetery on 12th March and a service of thanksgiving for his life was held on the same day at Knaphill Methodist Church where Peter Silley, as President, represented the Society.

Martin Adams

Spot-the-bug Competition!

In the June issue a bug was hidden amongst the pages and we asked you to find it. I am happy to say that a bottle of bubbly is on its way to **Andrew Shaw** of Don Whitley Scientific Limited who was the first to spot the bug. It was found nestling comfortably on the lap of the duck on the 'Dog and Duck' sign on page 40. Congratulations Andrew on your very prompt response – you're obviously a keen reader of *Microbiologist!*



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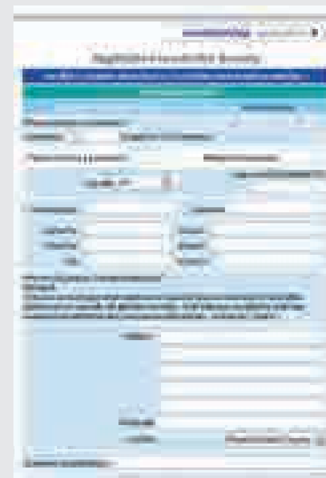


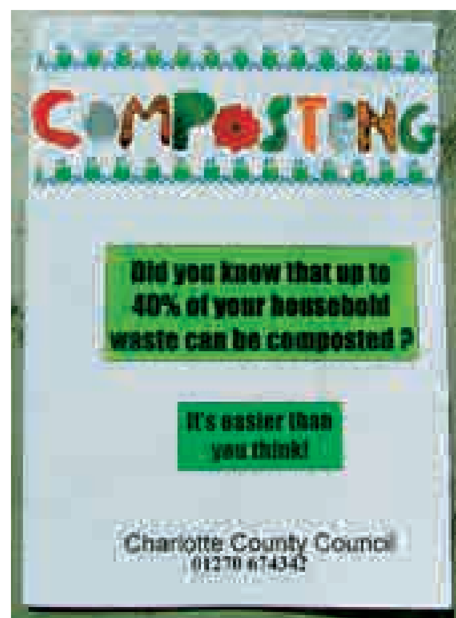
MISAC COMPETITION 2004

Composting: not just a load of old rot but a way to save the planet

THIS YEAR, the Society for Applied Microbiology (SfAM) sponsored the 16th MISAC (Microbiology in Schools Advisory Committee) competition. Officers of SfAM joined the Officers and other members of MISAC for the judging at the Institute of Biology in London. The members of SfAM involved were Meetings Secretary Dr Martin Adams (also a member of MISAC), Education Group Convener Dr Hilary Dodson and then Editor of Microbiologist, the society's magazine, Dr Anthony Hilton. There was a good response to the competition, attracting some 370 entries and involving more than 400 students from 46 schools. As is usual, there were more entries from the 11-14 age group than from the GCSE years.

As the purpose of the competition was to design an information leaflet for use by a local authority to encourage the general public to use composting, the judges looked for attractive, eye-catching styles that would first gain the attention of a potential reader and then maintain interest long enough for important information to be imparted. Many entries were of high quality. They understood the importance of these features and also demonstrated a good grasp of the scientific principles and environmental issues involved, picking out key points on the relevance of composting in environmental protection, the science behind the process and how to go about making compost. Good design included appropriate use of illustrations and effective communication used concise text tailored to the readership. One aspect of originality was evidence of the use of the entrants' own words, not entire text taken directly from other sources, e.g., the web. The competition gave opportunities for drawing and writing by hand and for using ICT skills. Although the judges felt unable to award prizes in the GCSE age group this year, several entries were highly commended and it was decided to give money awards to them and their schools. The overall results are on the MISAC web site.

MISAC and SfAM express their sincere thanks to everyone who took part, including those whose efforts were not rewarded with a prize on this occasion and the teachers who organised the preparation and submission of the entries. We hope that this has been a rewarding experience, leading to a greater interest in microbiology.



The winning entry in the 11 - 14 age group

Further Information

■ The winning entries can be seen on the MISAC website at: www.microbiologyonline.org.uk/misac.html

Dr J M Grainger
Chairman of MISAC

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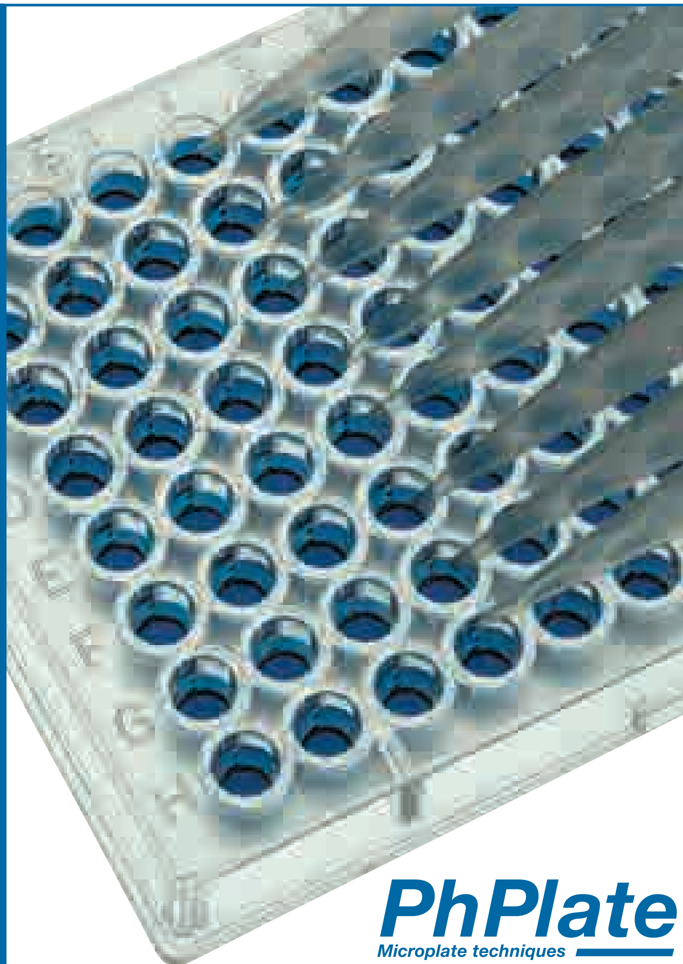

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jh18

Suspicious packages

Barry Holmes discusses the increasing controls on the supply of dangerous microorganisms and the transportation of pathogens

EUROPE HAS SEEN significant changes since its two World Wars. In recent times, following economic collapse in the USSR, many former Soviet-dominated countries regained independence, culminating in a European Union (EU) enlarged from 1st May 2004 to encompass 25 countries. Such advances yielded the promise of a prosperous and more peaceful world. Regretfully, the events of September 11th 2001 and the Madrid bombings in March 2004, show that lasting peace remains elusive. We are all now drawn in to a different kind of war in which conventional weaponry has little place. All of us are potentially in the front line, so we all have to be on our guard against the threats of terrorism, even if this amounts to no more than reporting suspicious packages left on a bus or train.

A variety of weapons could be employed by terrorists and of concern to us here are the dangerous microorganisms, those that could infect man, other animals, or crops.

It is incumbent on microbiologists to do their best to ensure that these hazardous microbes do not fall into the hands of those who would misuse them. This is not purely a responsibility of national culture collections but applies equally to any hospital or university laboratory that has acquired or isolated dangerous microorganisms and stored them away. Responding to the bio-terrorist threat has resulted in various legislative changes in various countries. Below, these changes are reviewed. Whilst the emphasis is on the UK, similar developments are or have taken place within the EU member states and other countries throughout the world.

Sending cultures overseas

Within the UK, export controls on certain dangerous microorganisms have been with us since 1993. The most recent legislation is the Export Control Act 2002, which came into force on 1st May 2004. Guidance on the impact of this legislation on those in academia or engaged in research can be found at: www.dti.gov.uk/export.control/publications/academguide.pdf.

One needs to register with the Department of Trade and Industry (DTI) prior to supplying hazardous pathogens to countries in either the EU or throughout the rest of the world. For countries outside the EU, the most common practice is for an application to be made to the DTI, by a nominated employee, for an individual export licence for each proposed shipment. Until 7th March 2003, Community General Export Authorisation EU001 permitted those registered with the DTI to export to countries such as Australia, Japan and the USA without an individual export licence. With the withdrawal of that facility, an individual export licence application must now be made even for those countries (see table 1).

Table 1. The control and Supply of dangerous microorganisms

Within the EU, one may not know the customer requesting dangerous microorganisms and there could be concerns of organisms supplied being passed on. It may thus be wise to contact the DTI (or equivalent) to check that the requestor is as claimed.

Additional organisms and toxins are likely to come under the export control regulations in the near future, including St Louis encephalitis virus, enterohaemorrhagic *Escherichia coli* and strains of *Clostridium perfringens* producing the epsilon toxin. The existing and proposed additional controlled organisms can be viewed at: http://www.australiagroup.net/en/control_list/bio_agents.htm (human pathogens) and http://www.australiagroup.net/en/control_list/animal.htm (animal pathogens).

Remember that, apart from the export controls, you also need an import licence from an increasing number of countries (e.g., Australia, Canada, New Zealand and the USA) before you ship hazardous microbes.

The deliberate releases of *Bacillus anthracis* via the postal system in the USA illustrate what can happen. Attempts to fraudulently obtain cultures of dangerous microorganisms from culture collections date back to at least 1984, when FBI agents arrested two Canadians attempting to collect cultures of *Clostridium botulinum* and *C. tetani* (Budiansky 1984). It is known that al-Qaida tried to establish a biological warfare programme in Afghanistan (Petro & Relman 2003; see also www.sciencemag.org/cgi/content/full/302/5652/1898/DCI).

Sending cultures within the UK

The two main areas of legislation are described below.

Specified Animal Pathogens Order (SAPO) 1998

Within the USA, all facilities holding dangerous pathogens must be registered, cultures can only be supplied to facilities that have also registered and the actual transfer is controlled via the Centers for Disease Control and Prevention (CDC), Atlanta, through the transfer Form EA101.

Similarly, within the UK, the supplier of dangerous organisms specified under SAPO must be in possession of a licence from the Department for Environment, Food and Rural Affairs (DEFRA); formerly the Ministry of Agriculture, Fisheries and Food (MAFF) permitting the supplier to hold these organisms. It is a requirement, under the Order, that those customers wishing to obtain such cultures must themselves hold a licence from DEFRA to do so. Thus, licence holders receiving requests for hazardous pathogens must not supply them unless or until the customer can furnish a copy of their own licence. In addition, before the cultures are despatched, DEFRA must have agreed to the issue of a transfer licence. Further information can be obtained at: www.defra.gov.uk/animalh/diseases/control/animal_pathogens.htm. It is unfortunate that with only DEFRA exercising such controls, there are no similar controls on the movement of non-animal pathogens such as *Burkholderia pseudomallei* and *Yersinia pestis*.

Anti-terrorism, Crime and Security Act 2001. Part 7 of the Anti-Terrorism, Crime and Security Act 2001 requires holders of hazardous pathogens (and toxins), listed in Schedule 5 of the Act, to be registered with the Home Office. Full details of the Act can be obtained at: www.legislation.hmso.gov.uk/acts/acts2001/20010024.htm . Registration can be achieved by email to: pathogens@homeoffice.gsi.gov.uk, or by post to: Pathogens Notifications, 6th Floor, West Wing, Home Office, 50 Queen Anne's Gate, London SW1H 9AT

or by faxing 020 7273 2773. Facilities that register can expect a visit from a police security specialist who will advise on any enhanced security measures deemed necessary.

Receiving cultures in the UK

If cultures are to be sent to you from the UK or anywhere else in the EU, if the organism is covered by the Specified Animal Pathogens Order 1998, then one must hold a licence from DEFRA beforehand. If you wish to import animal pathogens from outside the EU, then an import licence will need to be already held or obtained, as required under the Importation of Animal Pathogens Order (IAPO) 1980. For more information, again refer to: www.defra.gov.uk/animalh/diseases/control/animal_pathogens.htm.

For the majority of the animal pathogens listed under SAPO, an import licence will have to be applied for case-by-case and will not be granted unless the organisation already possesses a valid holding licence. If the hazardous pathogen is received from any part of the world, then if it is listed in Schedule 5 of the ***Anti-Terrorism, Crime and Security Act 2001***, you must register with the Home Office, Pathogens Notifications department.

Safeguards

As well as the legislation, anyone can adopt the following common sense procedures when receiving a request to supply dangerous microorganisms.

- *Do not dispatch cultures to private addresses.*
- *Do not accept electronic orders for hazardous pathogens; require all such orders to be on official headed notepaper and submitted by fax or post.*
- *Require that the customer Head of Department/Managing Director completes a form authorising which persons are able to place orders for dangerous organisms. Retain these forms on file and whenever a customer places an order for hazardous pathogens, a visual comparison can be made that the signature on the order matches an authorised signature on the authorisation form for that customer. Cultures should be despatched only to the authorised signatory of that particular order.*

I shall not enter the subject area of proper shipping requirements. However, many in the microbiological community have, in the past, taken cultures on their person to pass on to colleagues at international meetings and so on. In the present climate, however, one must strongly countenance against doing so. What customs official would be impressed by *"But it's only an Escherichia coli?"* Be warned by the experience of the American microbiologist Thomas Butler, who was arrested and tried on several charges arising from his personal carriage of specimens from more than 60 Tanzanian bubonic plague victims (Enserink & Malakoff 2003). He was found guilty on three counts (among others) of unauthorised export, making a false statement on a shipping bill and illegal transportation of hazardous materials.

Further Information

- www.defra.gov.uk/animalh/diseases/control/animal_pathogens.htm.
- www.legislation.hmso.gov.uk/acts/acts2001/20010024.htm
- www.sciencemag.org/cgi/content/full/302/5652/1898/DCI
- www.dti.gov.uk/export.control/publications/academguide.pdf.
- **Human pathogens** http://www.australiagroup.net/en/control_list/bio_agents.htm
- **Animal pathogens** http://www.australiagroup.net/en/control_list/animal.htm
- Pathogens Notifications, 6th Floor, West Wing, Home Office, 50 Queen Anne's Gate, London SW1H 9AT

References

- Budiansky, S. 1984, 'Dangerous pathogens - Illegal US bug trade halted,' *Nature*, **vol. 312**, p. 487.
- Enserink, M. & Malakoff, D. 2003, 'The trials of Thomas Butler,' *Science*, **vol. 302**, pp. 2054-2063.
- Petro, J. B.& Relman, D. A. 2003, 'Understanding threats to scientific openness,' *Science*, **vol. 303**, p. 1898.

Barry Holmes
National Collection of Type Cultures, Health Protection Agency, CPHL, London

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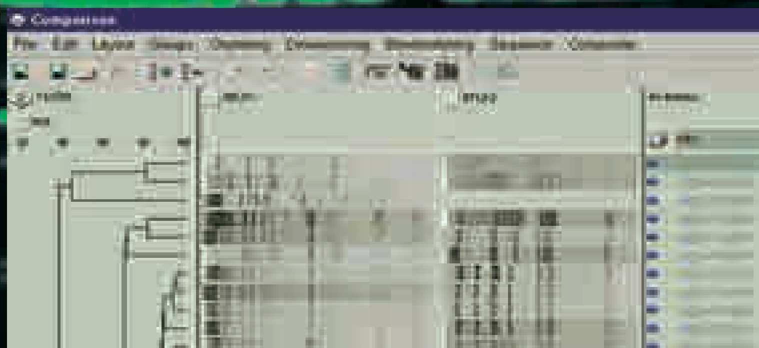

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Education in Ethiopia

In the fifth in a series of articles, **Dr Jenny Search** reports on her continuing two-year voluntary service overseas placement at Debub University in Ethiopia

THE END OF a busy academic year is approaching and I'm surprised to find myself looking forward to the summer break when I can devote more time to my research.

Our department (Applied Biology) offers a general microbiology course which is taught to third year students in our department and also to second year students in the faculty of agriculture. Last semester this course was not taught at all, but at the beginning of this semester I found out that with one other colleague we were expected to teach the course to three other departments as well as our own students.

Mr Mintesinot Ashebir and I are teaching microbiology to approximately 350 students from the departments of Animal Science, Plant Science and Horticulture. We work in two campuses 5 km apart connected by an hourly bus service, or a 20 minute bike ride. The lectures are not a problem as we can lecture two departments together. The big headache is the labs, as we only have two laboratories in the department to teach all the practicals for all the courses being offered. This logistical nightmare has been somewhat helped by the fact that, for reasons I won't go into here, the third year students started their semester about 6 weeks before the second year students which allowed us to finish most of the third year labs before the second year labs started.

The only way we could manage the large number of agricultural students was to halve the lab time from 3 hours to 1½ hours and teach two groups each afternoon. Each lab class contains about 60 students managed by the instructor and one lab technician. There is a shortage of media and chemicals which means that many of the experiments have to be carried out in large groups of up to 15 students. However, the students can individually try simple staining procedures such as Gram's and spore staining. This brought about a whole new set of difficulties as the students had forgotten how to use a microscope and insisted on using oil for all the lenses and became very excited about seeing air bubbles and the like. Slowly they are improving but it is hard to give individual attention to all the students, and when I give instructions it seems that only a tenth of the class understands (or listens to?) what I am saying!

Over the summer Mintesinot and I plan to revise the lab manual to include experiments that can be carried out by small groups of

Far left: *3rd year biology students on their field trip to the Rift Valley Lakes*

Left: *Mintesinot Ashebir teaching a microbiology lab to some of the 2nd year animal science students*

Right: *We had to stock up on bananas!*

Below right: *One of the enclosures at Arba Minch crocodile farm*

students with the equipment that is available. Mintesinot is also the colleague who I am carrying out research with into the microbiological quality of the water. We are in the process of starting the research but find it difficult to meet each other as our timetables seem to be such that we spend most of the week at opposite ends of the city in the different campuses!

Recently the third year biology students went on an educational field trip to the rift valley lakes as part of their course on Fisheries Biology. I was lucky enough to be invited to join the trip. First the students went to the lakes north of Awassa and visited a fish processing plant at Lake Zwei. The methods used are very traditional and not mechanised but there is still a huge problem in the country as a whole with over-fishing and the introduction of alien species to the lakes. After a swim in Lake Langano, one of the few "safe" lakes in Ethiopia i.e. free from crocodiles, hippos and schistosomiasis (due to a high mineral content) the students returned to Awassa for the night. The next morning I joined them to the town of Arba Minch which overlooks two lakes; Abaya and Chamo. Lake Abaya is the largest of the Rift Valley lakes measuring 1,160 km^2 are both are surrounded by mountains making the scenery spectacularly beautiful.

Arba Minch lies about 270 km south of Awassa on a fairly decent road and we set off at 6am to arrive with plenty of time to visit the lakes. Unfortunately we had a flat tyre 60 km north of Arba Minch and the driver had to return back to the closest town to get the spare repaired lest we had another before reaching our destination. The bus dropped us off at a pleasant spot where the cooks unloaded the pots and sacks of food and began to prepare lunch. One of the students wandered off and found a plantation which we went to visit. It was started ten years ago by World Vision but was handed over to local management three years ago. It is a cooperative scheme run by about 40 people who grow fruit and vegetables including bananas, mangoes, coffee, sugar cane and lettuce for export to Addis Ababa and further afield. They also grew a species of plant as feed for silk worms which produce silk for export.

We finally arrived at Arba Minch as the sun set and after negotiating accommodation for 54 students we retired for the night. The next morning we visited the shores of Lake Chamo. There, some locals (including a graduate from the Awassa College of Agriculture) talked to the students about the fish found in the lake. The talk was enhanced with the use of visual aids consisting of specimens of the fish found in the lake including tilapia, catfish and Nile perch.

After a few hours waiting for another tyre to be fixed, we visited a crocodile farm on the edge of Lake Abaya. Here the crocodiles are farmed, later to be turned into handbags and other products for export. In the 1960's the population of crocodiles in the lake were hunted almost to extinction so the farm was set up as a conservation measure to prevent illegal hunting of the crocs in the lake. We saw hundreds of crocs of different sizes, in rather small enclosures but we also saw some enormous specimens in their natural habitat from the edge of the lake. After a late lunch we stocked up on bananas, mangoes and limes and headed back to Awassa. The area seems so fertile it is difficult to imagine this area suffers from famines. In fact they are known as "green famines" as the countryside is so lush.

The trip provided me a great opportunity to see some more of this beautiful country in the company of my students and colleagues. It was good to get to know the students better. They quizzed me about everything from the view of Ethiopia in the "West" and how the education system and students compare in Ethiopia and the UK to whether I could eat a whole injera (staple grain product) in one go!

Further Information

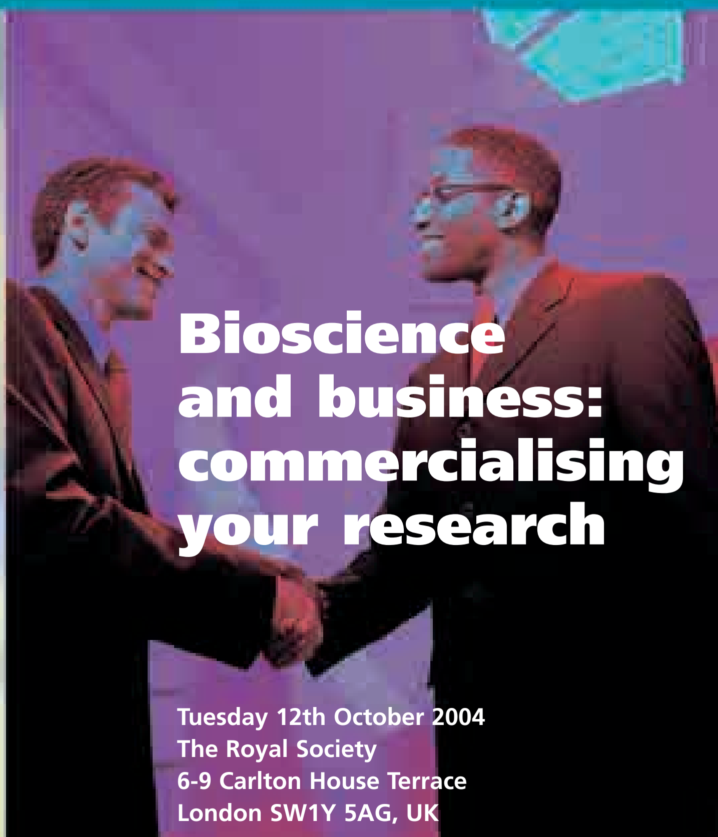
- www.neal-jenny.info
- www.vso.org.uk.
- The Faculty of Natural Sciences at Debub University: http://home.no/dufns

Jenny Search
Debub University, Ethiopia

BIOSCIENCES FEDERATION

The Biosciences Federation is holding a one-day symposium 'Bioscience and business: commercialising your research' on Tuesday **12th October 2004** at the Royal Society, London. World-class speakers will discuss some of the practical issues surrounding the commercialisation of bioscience, such as how to attract investment, developing partnerships with industry and intellectual property arrangements. SfAM members can attend at a reduced rate. For more information and a booking form call the Conference and Events Manager on **020 7581 8333** or visit the Society website at: **http://www.sfam.org.uk/meetdiary.php**

The Biosciences Federation was founded in 2002 to create a single authority within the life sciences that decision-makers can consult to assist the formulation of public policy. Its member organisations represent a membership of some 60,000 bioscientists and cover the whole spectrumfrom biochemistry and microbiology to ecology and agriculture.



EFFoST

FOOD INNOVATIONS FOR AN EXPANDING EUROPE

26 - 29 October 2004 ● Westin Hotel, Warsaw, Poland

This conference is organised by **EFFoST** in association with the **Warsaw Agricultural University** and will be administered and sponsored by **Elsevier.** The conference seeks to share and integrate advances regarding innovative aspects of dehydration, preservation and packaging and to disseminate current food chain management issues.
The conference aims to bring together the most recent innovations for the growing and extending European food chain. Plenary lectures by leading European scientists will be supplemented by contributed oral and poster presentations for which abstracts are currently invited.

Sessions include:
- Innovative Dehydration
- Innovative Preservation
- Innovative Packaging
- The Expanding European Food Chain

The language of the conference will be English.
The deadline for abstracts is 2nd July 2004.

information

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Guessing the Future: a thing of the past? Predictive Food Microbiology and Risk Assessment

SfAM JANUARY MEETING ● NORWICH, UK 12-13 January 2005

Overview

During the last few years there have been substantial advances in predictive microbiology and risk assessment. Use of predictive models facilitates the quantification of the growth of microorganisms in foods, and can be underpin risk assessments for the growth of pathogens in foods. Speakers at this challenging meeting will address important topics such as:

- Recent progress in predictive food microbiology and the ComBase Initiative
- Development of food spoilage models for use by industry
- Use of expert systems
- Risk perception and communication
- Risk characterisation and exposure assessment
- Practical approaches to risk assessment in the food and water industries

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There will be an opportunity during the meeting to present posters in any relevant subject area. Abstracts of less than 500 words, to include aims and objectives, brief methodology, results, conclusions and implications of the work, should be submitted only as a Microsoft™ Word document attachement to an email addressed to info@sfam.org.uk with the subject line 'January 2005 meeting submission'. The closing date for the submission of abstracts is Friday 22 October 2004.

For the latest information, costs and social events please visit us online at:
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Provisional Programme

Wednesday 12 January 2005

12.00–12.40 **Overview: history, background and development of PM and RA (Liase with MZ)**
Leon Gorris, University of Wageningen/Unilever Research, The Netherlands

12.40–14.00 **Lunch and posters**

14.00–14.40 **eCombase and recent developments in PM**
Jozsef Baranyi, IFR

14.40–15.20 **Expert Systems in Food Safety**
Carol Adair, Unilever Research

15.20–15.50 **Tea**

15.50–16.30 **Development of spoilage models for use by the food industry 1.**
Gail Betts, CCFRA

16.30–17.10 **Development of spoilage models for use by the food industry 2.**
Jane Sutherland, London Metropolitan University

17.10–17.50 **The link between PM and RA**
Marcel Zwietering, University of Wageningen, The Netherlands

For the latest information, costs and social events please visit us online at: **www.sfam.org.uk**

Thursday 13 January 2005

09.00–09.40 **Risk perception and communication**
Bob Mitchell, HPA

09.40–10.20 **Risk characterisation and exposure assessment**
Serve Notemans, TNO, Zeist, The Netherlands

10.20–10.50 **Coffee**

10.50–11.30 **Practical use of microbiological risk assessment by food companies**
Phil Voysey, CCFRA

11.30–12.10 **Practical risk assessment: role of eggs in the transfer of salmonella**
P Botney-Salo and A Varnam, London Metropolitan University

12.10–12.50 **Practical risk assessment: applications in the water industry**
Alan Godfree, North West Water, UK

12.10–14.00 **Lunch**

14.00–14.40 **Regulators and risk assessment: setting food safety objectives**
Paul Cook, FSA

14.40–15.20 **Where next with RA?**
Martyn Brown, Unilever research (to be invited)

15.20 **Tea and depart**

Please note that the above paper titles and speakers were correct at the time of going to press but may be subject to change.

BOOKING FORM and INVOICE

January Meeting 12 - 13 January 2005

Guessing the Future: a thing of the past?

Predictive Food Microbiology and Risk Assessment

Only ONE person per form please. If additional forms are required please photocopy this one

CLOSING DATE FOR REGISTRATIONS

Friday 24 December 2004. A **LATE BOOKING FEE of £20.00** will be applied to all bookings made after **Wednesday 1 December 2004**

FEES

	Full Members	Student, Honorary & Retired Members	Non-Members
Whole Meeting Rate: includes registration fee, full breakfast, coffee and tea breaks, lunches, Society dinner and overnight accommodation for Wed 12th Jan 2005	£200.00	£100.00	£300.00
Day Delegate Rate: includes registration fee, lunch, tea and coffee breaks.	£65.00	£55.00	£150.00
Additional accommodation per night inclusive of breakfast:			£120.00

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Charges - please tick the applicable box(es)	Amount
☐ **Whole Meeting Rate** (This includes accommodation, meals and the Society Dinner for the entire Conference):	£
☐ **Day Delegate Rate** (please tick the DAY you wish to attend): **Weds 12th:** ☐ **Thurs 13th:** ☐	£
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Please indicate any special dietary or other requirements (such as disabled access): ________________________________

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● **For all participants: The Society DOES NOT INVOICE for conference fees. Please treat your completed booking form as an invoice. Cheques must be in £ STERLING ONLY and made payable to 'The Society for Applied Microbiology'. Foreign cheques/ drafts MUST be negotiable for the full amount due. Please note that AMERICAN EXPRESS and DINERS CARDS are NOT ACCEPTED.** However the following credit and debit cards are acceptable: VISA, Mastercard, Eurocard, Delta, Electron, JCB, Maestro and Solo.

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SFAM JANUARY MEETING 2005 12 - 13 JANUARY 2005

☐ **I wish to apply for a SfAM Studentship grant to attend the 2005 SfAM January Meeting**

About this award

The Society offers Studentships to enable **student members** to attend Society meetings. These grants cover registration, accommodation, meals (where appropriate) and modest travel expenses. Preference is given to students offering a paper or poster and who have not previously received this award. To be considered for a Studentship grant, please complete this form in **BLOCK CAPITALS** and return it to the Society Office **no later than 6 weeks before the date of the meeting you wish to attend.**

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Grant authority: ____

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Your costs

Expected Travel Costs:____

Other costs - please specify:____

____

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Why do you wish to attend this meeting?

Please give your reasons:____

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Your signature:____________________________ **Date:** ____________________

(If you need more space for your answer please continue on a separate sheet)

Will you be contributing to the meeting by offering a Poster or presenting a paper? ☐ **Offering a Poster** ☐ **Presenting a Paper**

Your Supervisor's support

This section **MUST** be completed by your Supervisior or Tutor. Applications which are not supported by your Supervisor will be automatically rejected. **Please give your reasons why the applicant should receive a studentship:**

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Supervisor's name:____________________________ **Tel and extension:**____________________

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In signing this application I agree to reimburse the Society for any costs it may incur in awarding this grant should the applicant fail to attend the conference or fail to notify the Society of their inability to attend the conference within 14 days of the start of the meeting.

Please confirm your agreement by ticking the appropriate box: ☐ **I agree** ☐ **I do not agree**

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Spore forming bacteria — emerging and re-emerging issues

Old Ship Hotel, Brighton, UK ● 4th - 7th July 2005

The conference will consider recent advances in understanding in health, industrial and environmental issues associated with spore formers (following on from the 1994 meeting). It will review understanding of the taxonomy of spore formers and consider the physiological aspects, particularly those associtiated with spore structure and resistance. The health implications will be considered with respect to common infections caused by spore formers in both animals and man, the persistence of spores in food products, and also recent developments facilitating the use of spores as vaccine vehicles, probiotics and tumour targeting vectors. The environmental applications of spores will also be reviewed.

There will be sessions on:

- **Spore formers the great survivors — a taxonomy and physiology update**
- **Spore formers in food microbiology**
- **Spore formers: human health issues**
- **Environmental applications of spore formers**

There will also be an opportunity for oral and poster presentations. Further information will be published on the Society website and in the December issue of *Microbiologist.*

VENUE: The Old Ship Hotel

Dating back to 1559, The Old Ship is the oldest hotel in Brighton and stands proudly on the bustling seafront of this historical city. From its magnificent white façade to contemporary modern reception area, the hotel prides itself on its excellent customer service and comfortable, relaxed surroundings.

All 152 en-suite bedrooms have been recently refurbished and are elegantly designed with tasteful furniture and relaxing surroundings.

The contemporary Redz Brasserie is just a hop from the seafront and serves up a fine selection of gourmet dishes, accompanied by an extensive choice of fine wines.

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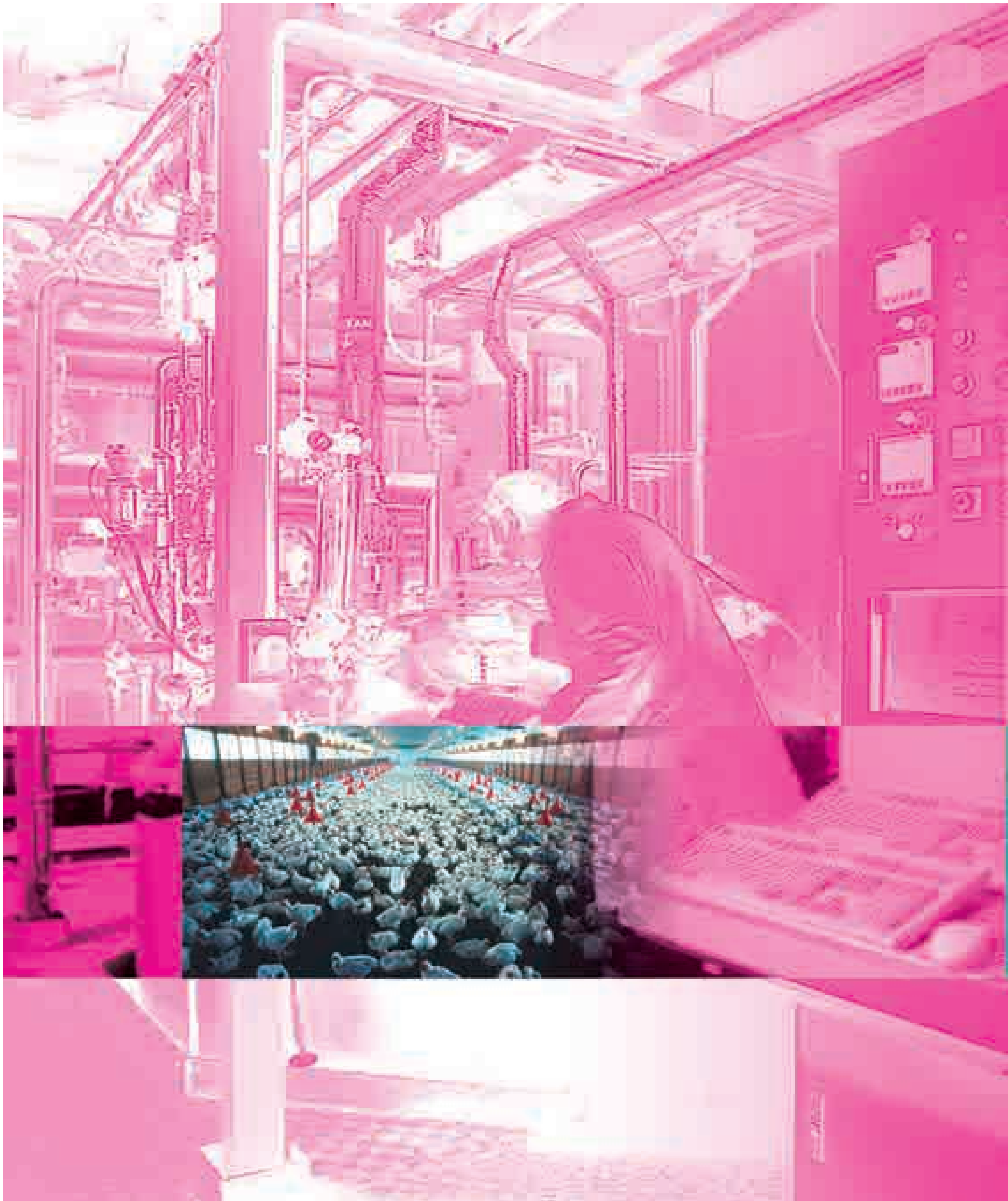

David Reynolds looks at the varying properties of 'good bacteria'

Tenants of the last 1.5 metres

BACTERIA CAN cause disease. Nothing controversial there – this has been accepted since Koch first proposed his postulates in 1876 for bovine anthrax. The 'disease' referred to here is, of course, infectious disease – whooping cough, wound infections, typhoid fever, cholera and the like. These are all readily treatable with antibiotics and fluid replacement but can bacteria do worse? Can they cause other, non-infectious diseases?

Well, yes they can - gut infections by *Salmonella* and *Campylobacter* are known to predispose to joint inflammation (Reactive Arthritis, Reiter's syndrome) and other autoimmune diseases such as Guillain-Barré syndrome. Streptococcal throat infections can lead to rheumatic fever, Mycobacteria may have a role in Crohn's disease and *Helicobacter pylori* has been shown to cause gastritis. At least there are some bacteria that do us no harm – the normal flora that exists on and within our bodies. Well… no - it is these bacteria that have the most insidious role in causing disease – they can give you bowel cancer and 1 in 20 of us will suffer the consequences by the age of 75. Is there anything we can do to defend ourselves against this enemy within?

Probiotics vs cancer

The development of colorectal cancer takes approximately 40 years in man. The initial cause of a lesion with the potential to develop into a tumour is often the result of damage to the gut lining by toxic compounds produced by the normal bowel flora – a complex interdependent community of some 1300 species of bacteria (Nelson, 2004). Carcinogenic N-nitroso compounds can be synthesised by the gut bacteria from nitrate and nitrite often found as preservatives in foods or generated by the action of the bacteria on bile produced by the liver. Bacteria can activate pro-carcinogens in the gut and release tumour promoters such as ammonia from protein in the diet (Mallett & Rowland, 1990).

But not all of the bacteria in the normal microflora are a problem and in this lies an answer. Professor Ian Rowland's group at Ulster University has shown that lactic acid bacteria (LAB – the group members include *Lactobacillus*, *Lactococcus*, *Bifidobacterium*, *Enterococcus*, *Pediococcus* & *Leuconostoc*) – part of the normal gut microflora and found in live yoghurts and probiotics (see Table 1) - produce lower amounts of these carcinogenic compounds and less of the enzymes

TABLE 1. Origins of Probiotics

Probiotics have long been used as a means of preventing disease in both man and other animals. The deliberate use of probiotics to prevent human disease dates back to the beginning of the 20th century.

Elie (Ilya) Metchnikoff working at the Pasteur Institute in Paris believed that the bacteria in fermented milk preparations helped to fight pathogenic bacteria and so extend the normal life span (*The Prolongation of Life*, Metchnikoff, 1907).

involved in generating carcinogens from non-carcinogenic complexes (Rowland, 1992). The presence of LAB in increased numbers in the gut helps to suppress the progression to tumours (Bolognani *et al.*, 1997).

Drinking a probiotic every day is not the whole answer though. Diet is also involved – the level of fat in the western diet predisposes to colorectal cancer. Studies have shown that high dietary fat affects the metabolic activity of gut bacteria as well as the levels of secondary bile acids, which may act as tumour promoters in the colon. Animal model studies indicate that it is total dietary fat, rather than the type of fat, that promotes carcinogenesis (Reddy, 1981). However, changes in diet can help – inclusion of non-digestible oligosaccharides from some plants (notably - onions, garlic, leeks, asparagus, chicory and artichoke) into the diet stimulates the growth of LAB and helps to prevent the progression to cancer. These components, collectively termed prebiotics, provide the saccharolytic LAB with nutritional advantages enabling them to out-compete carcinogen-producing proteolytic and lipolytic bacteria in the gut.

Yoghurts and probiotic drinks based on *Lactobacillus* spp., *Enterococcus* spp. or *Streptococcus* spp. are often used by people as part of a slimming diet or to help counteract such problems as acute gastritis, gastrointestinal infections, constipation, ulcerative colitis, food allergies, antibiotic-induced disorders, irritable bowel syndrome and even cardiovascular diseases. In contrast to the data on cancer prevention, the evidence for these beneficial effects has been largely circumstantial.

However, recent research

Professor Esko Nurmi (& Doppelganger) — his 1973 paper in Nature heralded in 'Competitive Exclusion' (picture by Mari Toivonen, 1998)

work carried out at Dundee University indicates that probiotics based on *Bifidobacterium* spp. can help to counteract the inflammatory bowel disease - ulcerative colitis (Kennedy *et al.*, 2002).

Probiotics vs infectious disease?

There are claims for other roles for probiotics - the use of LAB to prevent infectious disease is gaining popularity. This has been taken up more enthusiastically in the prophylactic treatment of animals than in man and products are currently available that claim to help prevent enteric disease in commercially reared pigs, calves and more particularly – *Salmonella* colonisation of broiler chickens (meat birds) and hens (laying birds). Given the extent of their use, the scientific case for their effectiveness is not as strong as might be expected.

Experimentally, dosing chicks with either single strains or mixtures of *lactobacilli* have given little protection (Watkins & Miller, 1983; Weinack *et. al.*, 1985). Using a mixture of a small number of bacterial species, Barnes *et. al.*, (1980) found that although coliform bacteria initially disappeared from the crop and caeca of probiotic-treated birds, following challenge, levels of *Salmonella Typhimurium* were 10-100 times higher than in the untreated controls.

Experience has since shown that relatively complex mixtures of bacteria (50-100 species) are necessary to obtain adequate protection against *Salmonella* (Impey *et. al.*, 1982; Nurmi, 1985; Stavric *et. al.*, 1985; Stavric, 1987). These complex mixtures of bacteria are thought to 'competitively exclude' *Salmonella* and some other bacteria from the gut.

Competitive Exclusion

During the 1970s several workers studied the problem of *Salmonella* contamination of chicken meat. The growth in the popularity of chicken meat had led to the industrialisation of chicken meat production. Systems had evolved whereby eggs were laid by hens on one farm, incubated and hatched out in near-sterile conditions in an automated hatchery, before being reared on a second farm. Some of these birds were becoming *Salmonella* positive during the rearing process. This caused little in the way of symptoms in the chickens – there was no discernible effect on growth rates, morbidity or mortality, hence meat producers were not affected by the presence of *Salmonella* in the birds.

However, at slaughter the process of evisceration was known to cause the gut contents of birds to contaminate the slaughter line and thus the meat of other birds processed on the line. If *Salmonella* positive birds were slaughtered on the line, all downstream birds and their meat would potentially be contaminated, with increased risk to the consumer.

One approach used to reduce this risk was to try to prevent *Salmonella* colonising the birds before slaughter. Chickens are increasingly resistant to *Salmonella* colonisation as they get older: 50% of one-day-old chicks can be infected using 10 cfu *S. Typhimurium* but it takes 106 cfu to colonise just 10% of the population by day 14 (Milner & Schaffer 1952). This suggested that maturation of the gut was in some way preventing *Salmonella* from colonising these older birds. One possibility was that the gut microflora was developing over time and increasing the resistance to *Salmonella*. This microflora was normally obtained by chicks from the environment and the mother hen. However, the change

from extensive farming, with eggs hatched by hens, to intensive chicken farming, with eggs hatched in incubators and an absence of adult birds, meant that development of this normal adult gut flora was delayed or prevented. Hence the partially developed gut flora of the intensively farmed chicks was less able to prevent *Salmonella* establishing itself in the guts of the birds.

The key step in what became known as **Competitive Exclusion** (CE) was to show that chicks fed the diluted minced gut contents of mature, free-range hens were more resistant to infection with *Salmonella* than were untreated birds. The contention was that the mature gut flora was a climax community that filled all of the available ecological niches in the gut and thus excluded less well-adapted bacteria such as pathogenic *Salmonella*.

This concept was demonstrated by the Finnish microbiologist Professor Esko Nurmi in a paper in *Nature* in 1973 (Nurmi & Rantala, 1973). The mechanism thought to be involved in CE has since been refined and several components are thought to be involved (see Table 2).

The impracticality of using minced adult hen gut to treat millions of chicks was not lost on Professor Nurmi and in a later paper anaerobically fermented gut contents were shown to give the same protective effects (Rantala & Nurmi, 1973). Later it was shown that even after sequential fermentation of the gut microflora, the protective effect was retained (Snoeyenbos *et. al.*, 1978, Mead & Impey, 1984). Also, fermenting the bacteria meant there was little chance of propagating any pathogenic viruses or protozoa that may have been present (Mead & Impey 1984), opening up the possibility of a commercial product.

Normal microflora - bacterial barrier at the mucosal surface of chicken caecum

TABLE 2. Possible competitive exclusion mechanisms

Direct attack	production of bacteriocins and organic acids that can damage pathogenic bacteria.
Nutrient competition	competition for the nutrients available in the gut.
Receptor competition	competition for attachment sites on the gut enterocytes.
Immune stimulation	of the non-specific immune system of the gut by the normal microflora.
Physical barrier	a bacterial mat blocking access to the mucosa

Fermentation hall — Aviguard, Microbial Developments Limited

Professor Nurmi's concept has been commercialised both in Finland (Orion Corp – Broilact) and the UK (MDL – Aviguard) with several other products available worldwide (Tecto CE, Avifree, PreEmpt & Avian Pac). These CE products contain the entire normal mature avian gut flora fermented, freeze-dried and sealed in foil sachets. They are used widely in the poultry industry and are either sprayed onto birds (method used for recently hatched birds) or given via the drinking water.

In addition to prevention of *Salmonella* infection, some of the commercial CE products have been shown to prevent gut colonisation by pathogenic, antibiotic resistant *Escherichia coli* bacteria (Weinack *et. al.*, 1981, Hofacre *et. al.*, 2002). Furthermore, work in the USA by Dr Charles Hofacre's group in the University of Georgia (Athens) has shown that one CE product helps prevent necrotic enteritis (caused by toxigenic Clostridium perfringens) in chickens (Hofacre *et. al.*, 1998a, 1998b).

Defined CE products

All of the early work on *Salmonella* prevention using CE was done with undefined or only partially defined mixtures of bacteria. This was unavoidable as many of the bacteria were uncultivable in isolation using the techniques of the time. However, it became clear that when selected bacteria were used as CE preparations they were much less effective than the mixtures derived from the entire gut flora. Single strains or mixtures of *lactobacilli* (Watkins & Miller, 1983; Weinack *et. al.*, 1985), or other bacterial species (Barnes *et. al.*, 1980) have not performed well.

Indeed, a defined product available in the USA (PreEmpt) was recently withdrawn from the market.

Later work demonstrated that relatively complex mixtures of bacteria (50-100 species) were necessary to obtain adequate protection and that they did so by establishing a balanced microflora in the chicken gut (Impey *et. al.*, 1982; Nurmi, 1985; Stavric *et. al.*, 1985; Stavric, 1987).

Attempts to produce a defined culture to prevent necrotic enteritis have met with only limited success. The Institute of Food Research (Norwich) and Veterinary Laboratories Agency (Weybridge) have worked together on a *Lactobacillus johnsonii* probiotic to exclude *Clostridium perfringens* which shows some positive effect in a colonisation model (La Ragione *et. al.*, 2004). However, there is no evidence yet that this probiotic will work to prevent infection in a necrotic enteritis disease model such as that used by Hofacre (Hofacre *et. al.*, 1998a, 1998b). *Bacillus subtilis* spores have also been used to attempt competitive exclusion of *Salmonella* and *Clostridium perfringens* (LaRagione & Woodward, 2003) but the results have been disappointing in comparison with the performance of commercial undefined CE products. To date there is no defined bacterium or group of bacteria capable of effective competitive exclusion of gut pathogens to the degree seen with undefined products.

So where does this leave us? The risk of bowel tumours is reduced if the normal bowel flora is skewed by supplementation with bacteria that produce fewer carcinogens from our food. Prevention of gut infections (at least in birds) is enhanced by the presence of a broad-spectrum bowel flora, which includes those bacteria known to generate carcinogens. This is, of course, not an issue for poultry (the usual recipient of competitive exclusion therapy), which rarely attain an age when bowel cancer becomes a significant risk to their longevity. Also, CE products are not so far available for human use and although it is unlikely that an undefined product would be allowed by the regulatory authorities, defined products remain a real possibility. However, this does rather beg the question as to who would be the source of any **Competitive Exclusion** product developed for people..?

David J Reynolds
Microbial Developments Ltd

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Jason Hinds reports on one of the joint **SfAM / SGM** Regional Meetings Schemes 2004

Microarray Workshop

'Group therapy for microarrayers?'

"I'm suffering from irregular spots!

Doughnuts make me unhappy!

Unsightly smears are the cause of my depression!"

What's the significance of it all?

COMMENTS FROM A DAYTIME TV talk show? In actual fact, these utterances are more likely to originate from scientists performing microarray experiments! The extent of such problems amongst UK microbiologists came to light during an evening microarray workshop held at the UMIST SGM meeting in September 2003. Such was their concern about the personal well-being of UK microarrayers that Drs Hinds and Dorrell prescribed a further group therapy session in the form of a SGM/SfAM sponsored workshop, held at the Hinxton Hall Conference Centre at The Wellcome Trust Sanger Institute on 12th May 2004.

DNA microarrays are a relatively new and rapidly evolving technology that are finding widespread application in microbiological research. Following the UMIST workshop, it was clear there was a real demand for a discussion forum in which researchers, with practical experience of DNA microarray technology, could get together to discuss and identify practical problems and solutions. The main objective of the recent workshop was to provide such a forum for young scientists; additional aims were to update and share the latest developments in this technology, consider how it may be exploited in microbiological research and also to encourage links and interaction between the numerous groups involved in related areas of study. Around 75 delegates, composed primarily of postgraduate and postdoctoral researchers using microbial microarrays in their research, attended the workshop.

The workshop consisted of four sessions, each having one main presentation to introduce the topic and raise the key issues for discussion based on the experience of the invited speaker. The first session on "Array Design and Construction" was presented by Richard Stabler (St George's Hospital Medical School) and followed by a session introduced by Arthur Thompson (Institute of Food Research) that highlighted the important issues in "Experimental Design and RNA Isolation." After a break to digest the information, coffee and biscuits there followed the final two sessions of the workshop. The first of these was presented by Jon Hobman (University of Birmingham) addressing the area of "Sample Labelling and Hybridisation" and followed by Jason Hinds (St George's Hospital Medical School) describing general approaches to "Data Analysis and Reporting." Following each main

presentation the audience were encouraged to actively participate in the workshop, several delegates used overheads to illustrate relevant points and there was lively discussion among the participants. This interactive approach helped to generate debate and clarify many of the issues highlighted, with much discussion continuing more informally throughout lunch.

The overall diagnosis was that radical drug treatment or electroshock therapy was not required to cure the ills of the microarrayers present. There was genuine consensus about problems and their solutions during the first three sessions, marking a dramatic improvement since the previous consultation at the UMIST meeting. However, the prognosis was less optimistic with regards to data analysis, with outbreaks of symptoms such as persistent headaches and cold sweats predicted to continue where this subject is concerned! It was agreed that future group therapy sessions to address this emerging problem would probably be required!

Jason Hinds
Bacterial Microarray Group,
St George's Medical Hospital School

Richard Armstrong describes an alternative method of dating

LICHENS, LICHENOMETRY AND GLOBAL WARMING

GEOLOGICAL AND climatic history share common themes with history, politics, and many other disciplines, that of the need to establish a timescale of events. In pre-history, timescales are critical to understanding evolution of biological, tectonic, and climatic systems that have interacted to produce the contemporary surface of the Earth. Many chronological tools have emerged to allow elapsed time to be accurately estimated for timescales ranging from decades to millions of years.

One of the most widely used techniques is 'Lichenometry', the use of symbiotic fungi in the form of lichens, growing on rock surfaces or other suitable substrates, to obtain an approximate date of the deposition of the surface. This article describes the methodology of lichenometry, the usefulness of the method in dating surfaces over the last 500 years, a period in which radiocarbon dating is relatively inefficient, and discusses how the technique has contributed to the debate over climatic change and global warming.

Lichens

Lichens are very common organisms on Earth and are found in a range of environments including the surfaces of rocks, trees, and man-made structures. A lichen is an intimate association between two quite different microorganisms, *viz.*, an alga and a fungus resulting in a

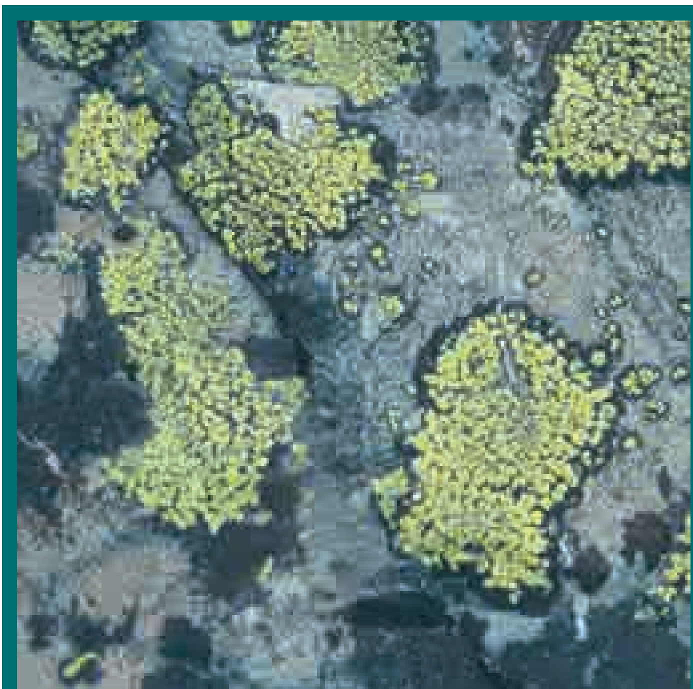

Fig. 1: *A population of yellow-green thalli of the lichen Rhizocarpon geographicum on scree boulders in the Pacific Northwest, USA. The yellow-green islands of areolae can be clearly seen as well as the black fungal hypothallus at the margin.*

'macroorganism', the lichen thallus, the morphology of which is quite unlike that of the two original constituents. The two organisms are so intimately associated that the term mutualism or symbiosis has been applied to it. Lichens were the first examples of symbiosis in which translocation between the constituent symbionts was demonstrated especially the movement of carbohydrate from the algae to the fungus. It is possible that the fungus supplies the algae with low concentrations of nutrients or vitamins but this translocation has not been convincingly demonstrated. A fuller description of lichen growth and physiology is given in a previous article in the *Microbiologist* (Armstrong, 2003, Vol 4, No.4).

There are three major types of lichen, *viz.*, the fruticose type in which the lichen thallus is attached to the substratum at a single point and forms a complex branched structure, the foliose type that comprises a series of radially arranged leaf-like lobes, and the crustose type that is tightly attached to the substratum. The foliose and crustose types of lichen grow radially over the substratum rather like a fungus on an agar plate but growth rates are very slow. Many foliose species have rates of radial extension between 2 and 5 mm per year but many crustose lichens grow much more slowly with rates of less than 0.5 mm per year. Some species grow so slowly that larger thalli growing in the Arctic may live to be over 5000 years old, thus making them candidates for oldest organism on Earth. It is this slow growth and the longevity of crustose lichens that have made them especially useful in lichenometry (Innes, 1985).

Lichenometry

Lichenometry depends on the assumption that if the lag time before colonisation of a substratum by a lichen is known and the growth of the lichen can be measured, then a minimum date can be obtained by measuring the diameter (or another property related to size) of the largest lichen at the site. The method is particularly useful in regions above and beyond the tree-line and especially in Arctic-Alpine environments where lichens grow very slowly and have great longevity. In such environments, it is possible to date deposits up to thousands of years old but, in the majority of cases, the method is most useful for dating over the past 500 years.

The 'Rhizocarpon' group of lichens

The majority of lichenometric studies have been conducted using a specific group of crustose lichens, viz., the yellow-green species of the genus *Rhizocarpon* (Fig. 1). This type of lichen is abundant in many Arctic-Alpine environments, grows very slowly (0.02 – 2 mm yr^{-1}), and lives to a considerable age. Morphologically, the lichen comprises discreet areolae that contain the cells of the alga *Trebouxia*, located on a fungal medulla which is attached to the substratum and which extends into a black algal-free marginal zone around the thallus called the hypothallus (Armstrong & Smith 1987). Within each areola there is a cortical layer 15-80 mm thick, an algal layer, and medullary tissue. The fungal hypothallus extends radially and to grow, relies on carbohydrate supplied from the areolae (Armstrong & Smith, 1987). Primary areolae near the edge of the hypothallus may develop from free-living algal cells on the substratum that are trapped by the hypothallus while secondary areolae may develop from zoospores produced within the thallus.

Different species of *Rhizocarpon* may colonise a surface at different rates, e.g., species of the section *Rhizocarpon* may colonise earlier than the species *Rhizocarpon alpicola*. However, *R. alpicola* grows faster than most members of the section *Rhizocarpon* and eventually becomes the largest lichen on the substratum (Innes, 1985).

Measurements

Several measurements of lichen thallus size have been suggested as the most useful in lichenometric studies. The use of the 'largest inscribed circle', i.e., the largest circle that can be drawn within an individual thallus was suggested by Locke *et. al.*, (1979) and has been adopted by several workers and is equivalent to using the 'shortest diameter' of the thallus. By contrast, many workers have suggested that it is the largest diameter of the thallus is the most appropriate measure. A major drawback of this method, however, is that individual lichen thalli can fuse together and therefore, be recorded as a single thallus (Armstrong, 1984). Other workers have averaged the longest and shortest axes or used both measurements to derive estimates of the surface area of the thallus (Griffey, 1977). In addition, various sampling methods have been employed. Either the single largest thallus at the site is measured and regarded as representative of age (Webber & Andrews 1973) or several of the largest deposits present are averaged (e.g., Mathews, 1974). Some workers have used the frequency distribution of thallus size as an indicator of the age of the substratum but the problem of using such distributions for dating is that they can be interpreted in several different ways (Innes, 1985).

Establishing a lichen growth curve

A lichen growth curve for an area can be established by two methods. First, directly by measuring the radial growth rate of the lichen over a period of time or indirectly, by measuring the diameter of lichen thalli on surfaces of known age. The indirect method is regarded as the most useful since it integrates' the effects of climatic change on lichen growth over long periods. By contrast, direct measurements relate only to the rates of growth over a particular, usually short, period of time. An example of the growth curve of the lichen *Rhizocarpon geographicum* obtained by direct measurement is shown in Fig 2. The growth curve appears to be non-linear, an early phase of increasing growth reaching a maximum in thalli about 2.5 – 4.5 cm diameter and then a phase of declining growth.

A variety of sources can provide information for dating a substratum and establishing a growth curve including gravestones, mine spoil heaps, abandoned farms or houses, and stone-walls or cairns. In addition, natural deposits can be used that have been dated accurately from historical events, radiocarbon dating, or the use of tree growth rings (dendrochronology). An example of a lichen growth curve relating lichen size and age derived in southeast Iceland is shown in Fig. 3 (Bradwell, 2001). This growth curve is also non-linear in shape. The relationship is often described best by a third-order polynomial function and which describes the declining growth rate apparent in larger and therefore, older thalli, also a feature of the growth curve derived by direct measurement. This is a feature of many *Rhizocarpon* growth

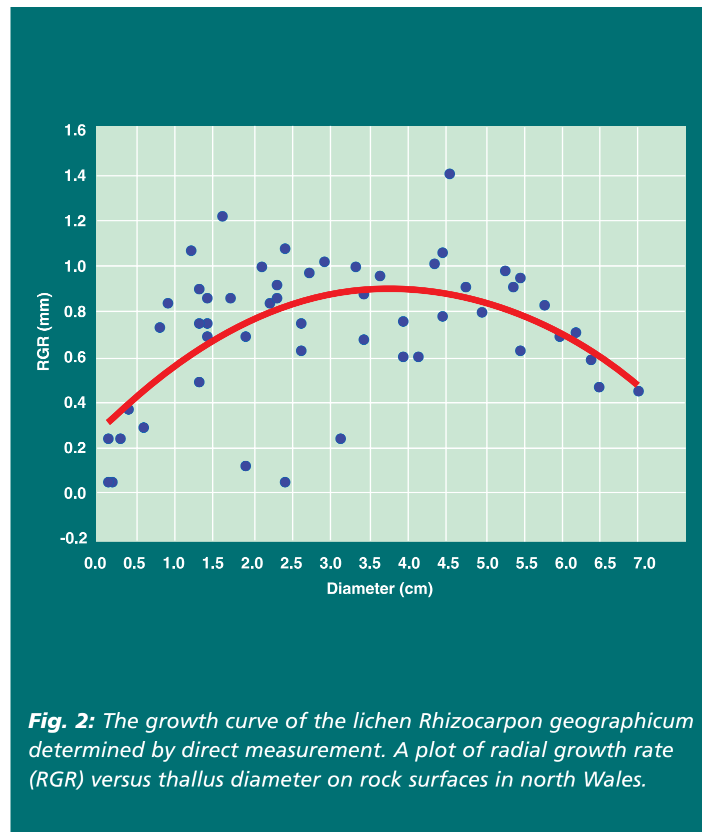

***Fig. 2:** The growth curve of the lichen Rhizocarpon geographicum determined by direct measurement. A plot of radial growth rate (RGR) versus thallus diameter on rock surfaces in north Wales.*

curves and could be due to thallus senescence (Armstrong, 1983). Another possibility is that the curve reflects a tendency for more rapid growth to have occurred in the last 100 years, i.e., environmental change over the last 300 years could have accounted for variations in lichen growth over time.

There are many uncertainties in lichenometric studies resulting from a lack of knowledge of fundamental lichen biology (Worsley, 1981). For example, there are limited data on the nature of lichen colonisation and on the influence of environmental factors on lichen growth rates. Moreover, there is a lack of reproducibility of many sampling designs, unresolved questions relating to the adequacy of using thallus diameter as an index of age, and many other difficulties associated with the methodology (Innes, 1985). Despite these problems, lichenometry, in combination with other methods, has proved to be an extremely valuable tool in dating the surfaces of deposits.

Applications of lichenometry

Lichenometry has been used in many different contexts to date surfaces. The dating of the sequences of rocks forming glacial moraines has been the most widely used application. Porter (1981) working on the moraines of Mount Rainier in Washington State, USA, (Fig. 4) used historical evidence for the past marginal positions of the glaciers and buildings together with other structures in the National Park to provide surfaces of known age from 1857 to the present. The single largest circular *Rhizocarpon geographicum* thallus was used as a measure of lichen growth. The study indicated that the moraine probably stabilised four years earlier than had been suggested by other studies.

Other applications of lichenometry including the dating of the stone images on Easter Island, stone-walls in England, river flooding, sea-level changes, and the occurrence of landslides (Innes, 1985).

Climatic change

During the last 25 years, there has been increasing concern as to the impact of possible climatic change and especially global warming due to the 'greenhouse effect'. Lichenometric studies have played an important role in this debate in providing data that support global warming. McCarroll (1993) used the size frequency distribution of lichen populations to determine the ages of boulders resulting from avalanches in south Norway. Snow avalanches reflect periods of high winter snow and rapid spring melting rather than low temperatures. It was concluded that increasing avalanche activity in south Norway was attributable to the greenhouse effect. Harrison and Winchester (2000) using a combination of lichenometry and dendrochronology studied the 19th and 20th century fluctuations of glaciers in south Chile. There was evidence for retreat of the glaciers following the 'little ice age' maximum between 1850 and 1880. Glacier retreat increased during the 1940s and the degree of synchrony exhibited by different glaciers has suggested a common climatic influence. This pattern has been repeated around the world. In central Asia, there were some small advances of the glaciers during 1908-11, 1911-34 and 1960-77 (Narama, 2002). However, significant recession also occurred in the 1900s especially in 1911-34 and 1977-78. Recession then accelerated after 1990 reflecting recent climatic

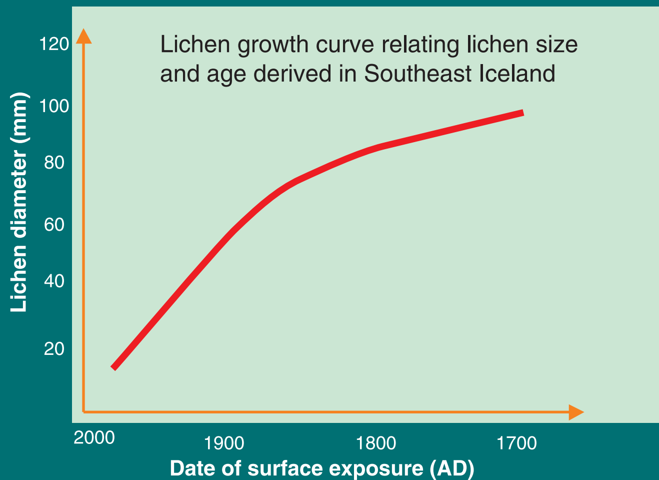

***Fig 3:** A growth curve of the lichen Rhizocarpon geographicum from southeast Iceland determined by lichenometry. The non-linear form of the curve is a notable feature of lichen growth curves from many sites around the globe.*

***Fig 4:** The terminal moraine of Emmons glacier, Mount Rainier, Washington State, USA. The boulders deposited by the retreating glacier are clearly visible and after a certain lag time become colonised by crustose lichens.*

warming in inland Asia. Records of glacier fluctuation are now compiled by the World Glacier Monitoring service and have derived estimates of global warming during the last 100 years (Oerlemans, 1994). Examination of data from all over the world has confirmed that the retreat of glaciers has occurred over the globe and can be explained by a linear warming trend of 0.66 Kelvin per century (Oerlemans, 1994).

Conclusions

Lichenometry is one of many techniques now available for estimating the elapsed time since the exposure of a substratum. Its advantages include an ability to date surfaces during the last 500 years, a time interval in which radiocarbon dating is least efficient, and provides a quick, cheap, and relatively accurate date for a substratum. Nevertheless, there are many problems associated with the methodology (Innes, 1985) with some authors (e.g. McCarthy, 1999) concluding that because of suspect methodology, most lichenometric ages are not in fact verifiably accurate. Improvements in methodology, the adoption of careful sampling regimes, and increased knowledge of the ecology of lichens are likely to improve accuracy and reliability in future (Innes, 1985). Lichenometry, together with complimentary techniques, is likely to continue to play an important role in dating a variety of surfaces and therefore in providing data that contribute to the debate regarding climatic change and global warming.

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Microbiology

European BioSafety Association

The European BioSafety Association (EBSA) was founded in June 1996. It is a not for profit organisation which aims to provide a forum for its members to discuss and debate issues of concern and to represent those working in the field of biosafety and associated activities. To date, the Association has over 300 individual members and around 20 Corporate members, representing over 15 countries in Europe, as well as the United States and Canada.

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TERMS & CONDITIONS

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2. A successful applicant cannot re-apply to the Fund for three years from the date of the award.

3. Preference will be given to applicants who are contributing to the meeting they wish to attend and/or are unable to obtain funds elsewhere.

4. Completed applications must include an abstract of any intended contribution to be made at the meeting and must be received by the Society Office not less than six weeks before the date of the event.

5. Student member applications must be supported by their supervisor and include the contact telephone number(s) and email address(es) of the supervisor or head of department who is supporting their application.

6. The maximum grant available is normally £1,000.

7. Under exceptional circumstances this maximum may be exceeded.

9. The award of this grant is at the sole discretion of the Hon President of the Society.

10. The applicant must write a short article of between 400 - 600 words within 4 weeks of the meeting, the content of which will be agreed with the Editor of sfam *Microbiologist* and will be published in the magazine. Photographs of the applicant and/or the subject of the article are desirable. These should be supplied as (a) digital files in TIFF or JPEG format at a size of not less than 4 inches square at a resolution of not less than 300 pixels per inch, or (b) original photographic prints which will be scanned and promptly returned to the applicant.

Enterobacter sakazakii; an emergent pathogen in infant formula milk.

A possible danger of consumption of breast milk substitutes are discussed by **Carol Iversen**

ALTHOUGH HUMAN BREAST milk is the most appropriate food for new born infants, breast milk substitutes and neonatal nutritional supplements are needed when breast milk is inadequate, unavailable or where viral transmission through breastfeeding is possible. The majority of these infant food products are manufactured as powders. Therefore, due to the limits of current manufacturing technologies, they are not sterile but are subject to microbiological criteria from the Codex Alimentarius Commission. A recent survey found that 2% of infant formula milk powders and 10% of dried infant foods contained *Enterobacter sakazakii* (Iversen & Forsythe, 2004).

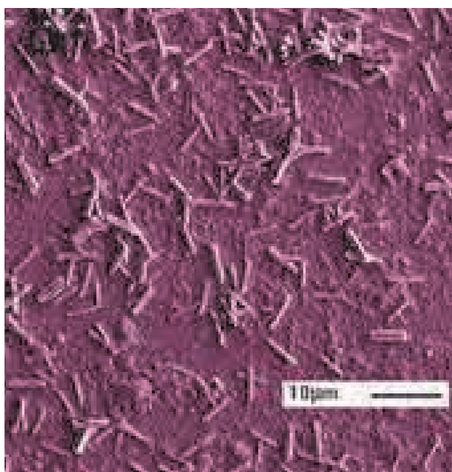
E. sakazakii has been associated with a rare form of meningitis, bacteremia and necrotizing enterocolitis (NEC), which is the most common gastrointestinal emergency in the newborn. Although *E. sakazakii* infection can be acquired by previously healthy newborn infants in the home environment, the majority of reported cases have occurred in pre-term infants in neonatal intensive care units and outbreaks have been linked to contaminated infant formula milk powder (Iversen & Forsythe, 2003).

The low reported incidence of infections demonstrates that the presence of microorganims in powdered infant formula milk is not a major health risk to the vast majority of healthy infants who consume infant formula milk each day. The bacteria are usually present at low levels and are in a dehydrated and stressed state. If these products are rehydrated according to manufacturers' instructions and either used immediately or stored in a refrigerator for no longer than 24 hours, there is little chance of infection. However if reconstituted infant formula is kept at room temperature or in a bottle warmer there is the possibility that the contaminating organisms will recover from the desiccated state and grow, therefore increasing the chance of infection. *E. sakazakii* can grow at temperatures between 5 - 47°C with mean doubling times of 21 minutes at 37°C, two hours at room temperature (18°C) and twenty hours at 6°C (Iversen, Lane & Forsythe, 2004).

It has also been demonstrated that *E. sakazakii* can adhere to materials used in infant feeding equipment (Iversen, Lane & Forsythe, 2004). Therefore once a feed has been given to the infant it is important to ensure that bottles and other equipment are immediately rinsed; and that thorough washing is followed by correct sterilization. Bacterial growth in the milk residue in a used bottle could lead to formation of a biofilm that may be more resistant to the cleaning and sterilization process. This may lead to an accumulation of bacteria in subsequent feeds.

In February 2004 a Joint FAO/WHO Workshop on *E. sakazakii and* other microorganisms in powdered infant formula was held in Geneva (http://www.who.int/foodsafety/micro/meetings/feb2004/en/). Outcomes of the meeting include the recommendation that Codex should consider the establishment of specifications for *E. sakazakii* in powdered infant formula and that validated detection methods should be promoted.

At present there is no validated isolation method for *E. sakazakii* and no specific legislation governing its permitted presence in foods. Early methods for the detection of *E. sakazakii* were based on general Enterobacteriaceae isolation using



Enterobacter sakazakii adhering to a baby feeding bottle. (JEOL-840A scanning electron microscope, JEOL Ltd., Tokyo, Japan)

Enterobacteriaceae enrichment broth and violet red bile glucose agar (VRBGA), followed by picking five colonies onto tryptose soya agar and observing yellow pigment production after 48-72 hours incubation at 25°C. Confirmatory steps included α-glucosidase activity and biochemical profile using API20E (bioMérieux UK Ltd.). This approach however has several limitations;

(a) *E. sakazakii* has the same colony morphology as other Enterobacteriaceae on VRBGA so can not be specifically selected.

(b) other Enterobacteriaceae could outgrow *E. sakazakii* during the pre-enrichment and enrichment stages, leading to relatively few *E. sakazakii* colonies on VRBGA and subsequently a reduced chance of picking the organism onto TSA

(c) not all *E. sakazakii* strains produce a yellow pigment and pigmentation can be transient on subculturing.

(d) a recent study comparing identification of *E. sakazakii* using biochemical profiles and 16S rDNA sequencing found discrepancies in strain identification between different biochemical kits. The API20E kit produced both false negative and false positive identifications. It was also found that strains identified as *E. sakazakii* by all biochemical test methods formed at least four distinct clusters when the 16S rDNA sequences were compared which may indicate separate species (Iversen *et. al.*, 2004).

To overcome these problems a chromogenic agar has been developed for the isolation and differentiation of *E. sakazakii* from food and dairy samples (Chromogenic Enterobacter Sakazakii Agar: formulation according to Druggan, Forsythe & Iversen, CM1055, Oxoid UK Ltd). *E. sakazakii* hydrolyses the substrate 5-bromo-4-chloro-3-indolyl-α,D-glucopyranoside to an indigo pigment, producing blue-green colonies (Iversen, Druggan & Forsythe, 2004). In a comparison with the method based on Enterobacteriaceae isolation and yellow pigmentation, the chromogenic agar provided results two days sooner and detected *E. sakazakii* in 67 samples compared to only 19 samples using the VRBGA method (Iversen & Forsythe, 2004).

The FAO/WHO Workshop also recommended that research should be promoted to gain a better understanding of the ecology, taxonomy, virulence and other characteristics of *E. sakazakii*. At Nottingham Trent University research is progressing in all of these areas. Our research team presented six posters at the recent American Society for Microbiology 204th Annual General Meeting in New Orleans. The topics covered included detection and isolation, desiccation survival, identification, molecular typing, and virulence factors.

I am very grateful to SfAM for the **President's Fund Grant** to support my attendance at the ASM meeting. For more information on *E. sakazakii* and other emerging food-borne pathogens please visit our website (www.foodmicrobe.com).

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Carol Iverson
Nottingham Trent University

The Society offers FULL members an opportunity to give undergraduate students of microbiology the chance to obtain work experience during the summer vacation.

Grants can be made available to ANY FULL member who is able to offer a suitable undergraduate student a work placement for a period of up to 10 weeks during summer. The grant is £160 per week for the student for a maximum of 10 weeks and up to £50 per week for lab costs for a maximum of 10 weeks. To apply, visit **www.sfam.org.uk/members/prizes.php**

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1. Any full member of the Society who can offer an undergraduate student, or a recent graduate (within 6 months of graduation) a work placement is eligible to apply for this grant. The placement can last up to a maximum of 10 weeks, normally during the summer vacation.

2. The Grant will normally provide support at the rate of £160 per week for the student and up to £50 per week for lab costs. The monies will usually be paid to the Department in which the student/graduate works unless a specific request is made for an alternative method of payment.

3. Applications should be made by the supervisor using the PDF form provided on the website or the paper form obtainable from the Society Office.

4. Successful applicants and their students/graduate must write a report on the placement within 4 weeks of completing their placement which will be published in *Microbiologist*. Photographs of the applicant and/or the work done during the placement are desirable. These should be supplied as (a) digital images at a size of not less than 4 inches square at a resolution of not less than 300 pixels per inch, or (b) original photographic prints which will be scanned and promptly returned.

5. Normally a member may not apply for a further grant until a period of two years has elapsed.

6.There is no closing date for this Grant and applications can be made any time during the year. Applicants must apply at least 6 weeks before the proposed start date.

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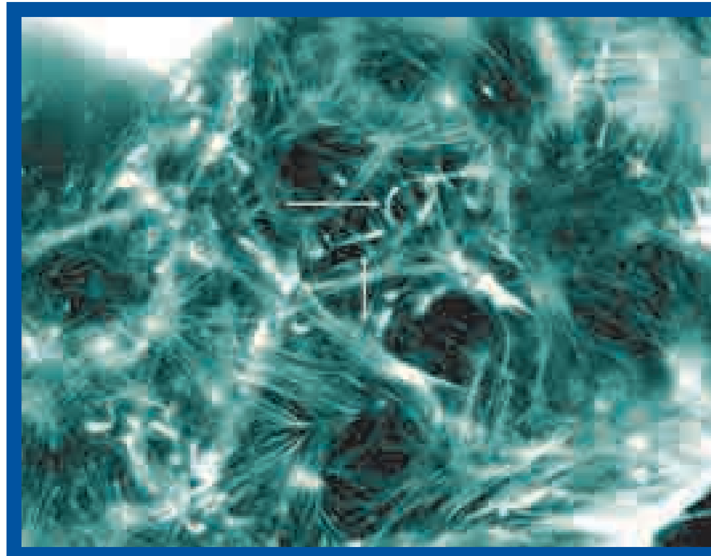
The Epidemiology and Pathogenesis of *L. monocytogenes* Infection

FOLLOWING THE DISCOVERY of *Listeria monocytogenes* in 1926 by EGD Murray as the aetiological agent of a septicaemic disease affecting rabbits in his laboratory in Cambridge; *Listeria* isolates remained somewhat of a laboratory rarity with unknown epidemiology. However, knowledge of *L. monocytogenes* increased in the 1970s and 1980s following many epidemics in Europe and North America when listeriosis was firmly established as a food-borne infection. Listeriosis presents as an extremely severe systemic infection in humans, characterised by meningitis, septicaemia and foetal death. Although relatively rare, there were 194 cases of listeriosis in England and Wales in the year 2000, resulting in 68 deaths.



Movement of L. monocytogenes in Caco-2 epithelial cells using host cell actin polymerised by the ActA protein. Arrows indicate bacteria and actin tails.

The extreme virulence of this organism is clear to see when compared to data from the same year for *Campylobacter* infections – with 86 deaths out of 360,000 people infected. *L. monocytogenes* can grow at a wide variety of temperatures, as low as 4°C — refrigeration temperature — which is why foods such as soft cheeses, patés and pre-cooked and reheated foods such as hot dogs have been widely implicated in infection.

L. monocytogenes is an intracellular parasite, and the gastrointestinal tract is thought to be the primary site for entry through epithelial cells. Once internalised, the bacteria are able to translocate to the spleen and liver where they are able to rapidly multiply in hepatocytes to form infectious foci. Bacteraemia can then follow, leading to septicaemia and meningitis. Due to *L. monocytogenes* possessing the ability to cross the placenta, neonatal death, listeric abortion or still birth can also result in pregnant women. The method by which *L. monocytogenes* infects cells has established it as a model organism for intracellular parasitism and mouse and guinea pig models of infection are available.

Uptake into non-phagocytic cells is mediated through the Internalin family of proteins, encoded by the *inl* genes. Although many Internalins have been identified, two of importance are InlA and InlB. InlA comprises 800 amino acids and mediates entry into epithelial cells, recognising E-cadherin on the epithelial cell surface. InlB has been found to mediate entry into murine hepatocytes and it is thought that the host cell receptor for InlB is the globular head of the complement factor C1q. Once internalised, listeriae need to be able to escape from the phagocytic vacuole prior to lysosomal fusion. The major virulent determinant responsible for escape from the vacuole and thus entry into the cytoplasm is listeriolysin O (LLO), encoded by the *hly* gene. LLO is a member of a large family of pore-forming haemolysins secreted by Gram-positive pathogens and is similar to Streptolysin O from *Streptococcus pyogenes*. Listeria mutants deficient in LLO fail to escape from the phagosomal vacuole. In addition to LLO, two Phospholipases C also assist in vacuolar escape. Once *L. monocytogenes* is free in the cell cytoplasm, multiplication of the organism is encouraged by more favourable conditions.

One essential aspect of any intracellular microorganism is the ability to facilitate its continued spread. To this end, several pathogens have evolved mechanisms to use the power of actin polymerisation to spread within host tissues. After lysis of the phagosomal vacuole, free-growing bacteria become coated with actin filaments. The actin coat is rearranged by the bacterium into a tail that trails the bacterium (see figure 1). This tail can be up to 40 µm long, with the length of the tail being proportional to speed. Upon contact with the plasma membrane, they cause the formation of protuberances, containing a bacterium at the tip. Internalisation by a neighbouring cell leads to the formation of a double-membrane vacuole, which is lysed by LLO, thus starting a new cycle of infection. *L. monocytogenes* mutants were isolated that were defective in intracellular motility using transposon mutagenesis. The site of the transposon insertion was found to be in the *actA* gene, which encodes ActA, a 639 amino acid protein responsible for the recruitment and polymerisation of host-cell actin. Mutants deficient in ActA form microcolonies, and are unable to spread from cell to cell, rendering them avirulent in a murine model of listeriosis.

Regulation of virulence genes is mediated through PrfA, the master regulator of virulence in *Listeria monocytogenes*. PrfA is the main switch of a regulon that includes all the virulence factors described above. Synthesis of virulence factors outside the host may represent a burden, compromising the ability of the bacterium to survive in the natural environment. PrfA therefore

regulates the expression of virulence genes in response to many extracellular stimuli, such as temperature, entry into stationary phase and oxidative stress. My area of research is concerned with the ability of *L. monocytogenes* to sense GTP levels in the cell and respond accordingly using the GTP-sensing protein CodY. GTP-sensing proteins have been shown to play major roles in other bacteria, such as *B. subtilis*, when cells make the transition between rapid growth (high GTP) and stationary phase and sporulation (low GTP).

In the last 20 years listeriosis has come from being a mere laboratory rarity to a well-publicised source of food-borne infection and major progress has been made towards understanding the life cycle of *L. monocytogenes*. However, knowledge of the pathogenic processes by which it causes disease is relatively scant. Recently, the European Listeria Genome Consortium elucidated the complete genome sequence of L. *monocytogenes*. This, combined with recent advances in microarray technology now allows much more detailed interrogation of the *Listeria* genome. This will clearly be an advantage, as many of the virulence determinants so far identified were based on overt phenotypes shown under *in vitro* conditions. The discovery of ActA not only showed the method by which *L. monocytogenes* moves intracellularly, but also gives many valuable insights into host cell actin biology. There is also evidence that *L. monocytogenes* could be a potential vector candidate for the delivery of DNA and proteins into eukaryotic cells, which would have some potential for vaccine delivery.

Undoubtedly, precise knowledge of the mechanisms by which *L. monocytogenes* enter, move and replicate within cells, and its subsequent ability to cause disease will be crucial in fully understanding the cell biology and pathogenesis of infection, in addition to the valuable insights it can provide into eukaryotic biological processes.

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An unhealthy profession?

Mark Hammond asks if microbiology is no longer a safe career choice for graduate students

As a microbiology student sixteen years ago, I had a departmental recruiting poster taped to the wall next to my desk. It read, "If you are curious, patient, and extremely bright, consider a PhD in microbiology." In 1988 a degree in microbiology seemed like a pretty good idea.

AIDS was just exploding upon an unprepared world. Lyme disease was racing through the Northeast United States and evidence was emerging that a bizarre neurologic disease might be caused by an equally bizarre infectious agent called a prion.

But now in 2004, a degree in microbiology, or even a vague interest in infectious diseases, might not be such a good idea. In fact, it might get you investigated by MI6. In the States it could even get you arrested.

After the terrorist attacks of September 11th in New York, and the later anthrax mailings, Western goverments scrambled to hastily enact a series of laws and regulations to discourage future acts of bioterrorism and misguided hoaxes. The US anthrax mailer has still not been caught and no other bioterrorists appear to be lurking on the horizon.

Deprived of new suspects, terrorists, plagues or other insidious acts of microbiology, the American secret services turned their attention to softer targets.

In the last two years the US government have used the provisions of these new anti-terrorism Acts to arrest and prosecute university professors and graduate students. None of these U.S. citizens are terrorists, but they have all been run to ground by the FBI's need to be seen doing something — even if it's the wrong thing.

Two years ago, a University of Connecticut graduate student (Tomas Foral) became the first person to be arrested under the Patriot Act. His crime: moving a 35-year-old sample collected from an anthrax-infected cow from one freezer to another freezer. This spring, world-renowned plague expert and physician Thomas Butler was sentenced to two years in prison. His crime: mislabeling a FedEx package containing plague samples, and getting into billing disputes with Texas Tech University accountants over his research funds.

In July, a federal grand jury indicted a University of Pittsburgh genetics professor, (Robert Ferrell), who apparently failed to fill out the appropriate paperwork. He faces the possibility of 20 years in prison.

The result is widespread fear among scientists. Some researchers have stopped working on certain dangerous pathogens and some universities have destroyed valuable collections rather than risk a paperwork mishap that might attract the attention of our trigger-happy security services. Cornell professor and Nobel Laureate Robert Richardson noted that before the Patriot Act thirty-eight of his colleagues were working on “select agents.” Now there are only two. Anthrax expert Paul Keim told the Los Angeles Times last October, "All of us are worried we are going to fall into some trap that we don't know about."

This assault on science and scientists is an important issue that has not been well covered outside professional journals and science magazines. It should be. When US government agents, backed by new American terrorism laws, come to view classrooms as terrorist training camps, hobbyists as dangerous lunatics, and professors as domestic terrorists, the UK is sure to follow. Perhaps a less dangerous career might be a good move for today's microbiology students?

Mark Hammond
University of London

The use of magneto-immunoassays in lateral flow and its application in microbiology.

Jacqueline M Barnett discusses the many and varied applications of lateral flow immunoassays

THE DETECTION AND quantification of many substances *in-vitro* has been possible by the utilisation of the specific high affinity interaction between an antigen and its antibody. A critical component in the development of immunoassays in terms of increased sensitivity and ease of use was the involvement of a solid phase to attach the antigen-antibody complex. Microparticles were utilised very early as a solid phase, initially in particle agglutination immunoassays where agglutination was detected by nephelometers. Further advances using coloured latex or gold microparticles as a solid phase and label resulted in the development of one step, immunochromatographic lateral flow assays, the most widely known being the over-the-counter, one step, pregnancy test, **Clearblue**™, first launched by Unipath, Bedford, UK in 1988.

In lateral flow immunoassays (LAI) a complex is formed between coloured microspheres, that flow freely by capillary action and a capture reagent that is bound to a nitrocellulose or similar membrane at the test line. Usually antibodies are bound at the capture line and on the particles, either by passive adsorption, or by specific chemistries depending on the functionality present on the surface of the particles.

In addition, lateral flow assays have been developed that detect nucleic acid, through the use of specific biotinylated probes present at the capture line. LAI are used today to detect many analytes and have applications in environmental, food, veterinary and medical microbiology. Examples include, **RapidChek**™ *Salmonella* Lateral flow Test Kit for the detection of *Salmonella* pathogen in a variety of foods including raw meat, poultry, dairy products, processed meats and fresh produce and **Pocket Diagnostics**™ for the detection of viral potato pathogens. Many lateral flow kits exist for use in medical microbiology in infectious-disease testing e.g. **QuickVue Strep A** and **QuickVue influenza** for the detection of group A *streptococcus* and influenza A and B viruses respectively in throat swabs. In addition many lateral flow kits exist for the detection of antibodies and antigens for HCV, HBV and HIV in blood samples. Lateral flow assays are accurate, rapid one-step assays that can be performed by non-laboratory personnel and are therefore ideal for point of care use. However most lateral flow assays have a limited quantitative range and give only yes/no answers above threshold values.

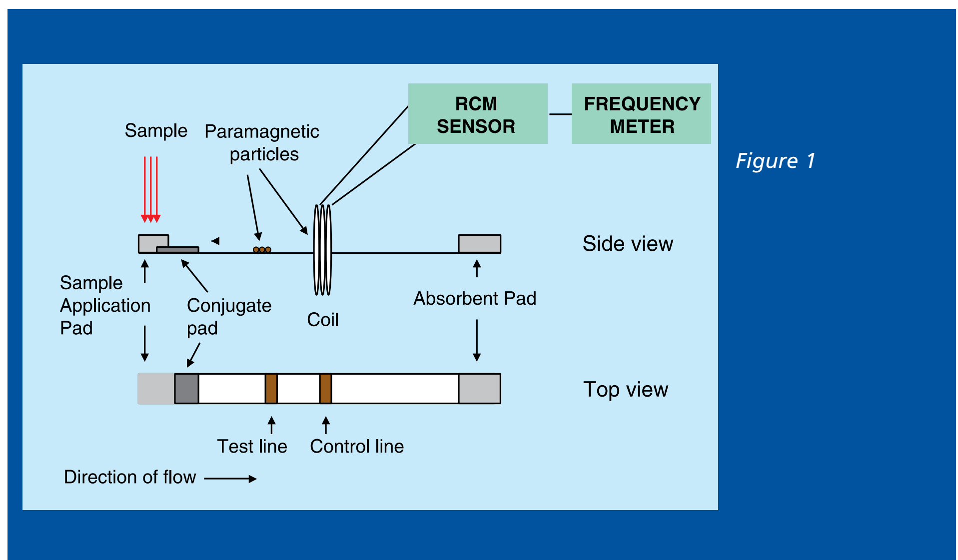

Figure 1

Magnetic particles have been used as an aid to separate micro-organisms for downstream identification for some time as described by Duncanson, (2004). Recently a new generation of immunoassays has been developed using paramagnetic particles. These particles are not permanently magnetised but are attracted to an external magnet and have been widely used not only as a solid phase to enable separation of target analyte, but also as a label. Research activity by several groups has been directed towards the development of a variety of sensors which are able to detect and quantify the particles by virtue of their magnetic properties. Advantages of this system include that the potential exists to use external magnets to accelerate reactions as described in (Luxton *et. al.*, 2004), no specific sample preparation is required, the assay strips are stable and can be read at any time and the particles present at the capture line can be quantified over a wide range using inexpensive instrumentation. In addition, the assays are not susceptible to quenching by substances present in the sample as in the case of lateral flow assays based on fluorescent particles e.g. Quantum Dot Corporation and RAMP by Response Biomedical.

Several approaches have been used to develop a sensor to quantify magnetic particles these include the use of a Force amplified biosensor, (US Naval Research Laboratory); a Maxwell bridge, (LifeAssays AB); Magnetoresistive sensor, US Naval Research Laboratory); Magnetic induction, (Quantum Design) and a Super-conducting-quantum-interference-device, SQUID. At the University of the West of England a sensor based on the effect of paramagnetic particles on the resonant frequency of a coil of wire in parallel with a capacitor (a magnetometer resonant coil, RCM), has been shown to produce a decrease in the measured frequency that is directly proportional to the number of paramagnetic particles present, (Richardson *et. al.*, 2001a). Experiments with this system, (Richardson *et. al.*, 2001b and Barnett *et. al.*, 2004) indicate that paramagnetic particles can be used in immunoassays and in lateral flow and that using the RCM sensor, analyte detection levels are equivalent to published data.

A photograph and schematic diagram of a magneto-lateral flow assay measured using the RCM sensor is shown in Figure 1 and 2.

In addition, others (LaBorde & Farrell, 2002) have reported the use of paramagnetic particles in lateral flow. In this assay paramagnetic particles are quantified by measurement of magnetic induction in a rapidly oscillating magnetic field (Quantum Design). However the cost and sensor design restricts the options available for the assay format and may limit the usefulness of this sensor in a point of care setting.

The optimum system to detect magnetic particles would be a small, relatively cheap hand device easily operated by untrained personnel. The implications for the use of such lateral flow assays in microbiology are huge. Lateral flow assays are already used in microbiology and the technology has advanced so that antibody pairs can be selected using surface plasmon resonance (SPR) and assays optimised with robotic systems (Wilson & Howell, 2002).

The use of multi-analyte detection in this system could allow a single test for the detection and quantitation of a variety of pathogens e.g. one test to detect influenza, respiratory syncytial virus and SARS. There has been much discussion in the literature regarding the use of lab-on-a-chip diagnostics and the potential for point of care use. However, lateral flow assays have proven their robustness for point of care use and the simplicity and low technical involvement in the use of quantitative multi-analyte magneto-lateral flow assays could enable a significant expansion of point of care testing.

Samples readily obtained at the point of care by untrained personnel usually contain low levels of analyte. Recently the FDA has approved a HIV test to detect HIV antibodies in lateral flow format using oral fluid, (OraSure Technologies Inc). The ability to use oral rather than blood samples will enable this test to be performed in the Third World without the need to use needles to obtain blood samples and thus removes the danger from needle stick injuries.

Developing a highly sensitive quantitative magneto-immunoassay in lateral flow format has the potential to expand point of care testing, enabling the use of samples not currently usable at the moment. Developments in sensor technology combined with the production of more highly magnetic particles may lead to increased sensitivity enabling the detection of micro-organisms in samples previously only detected by sophisticated molecular biology. Lateral flow assays with increased sensitivity could also have application in the prevention of bioterrorism.

Figure 2

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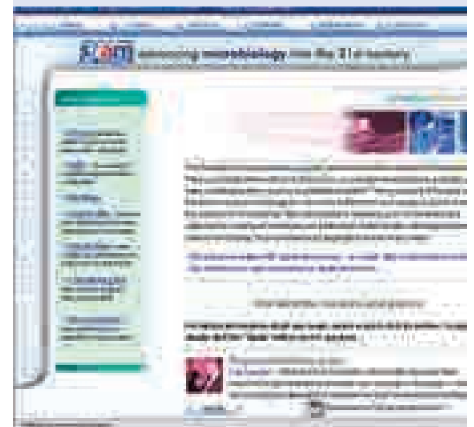

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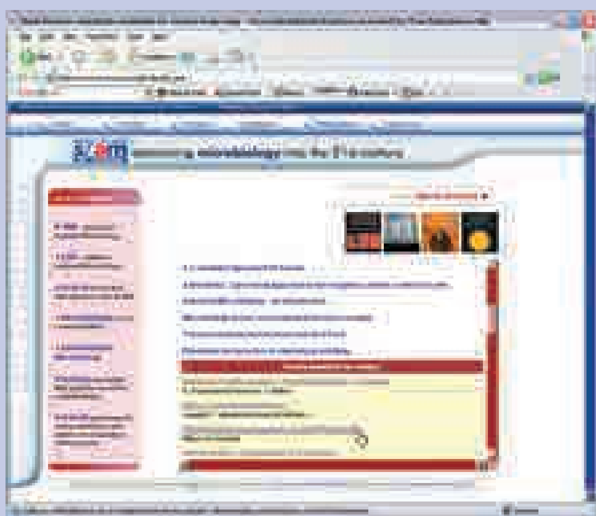

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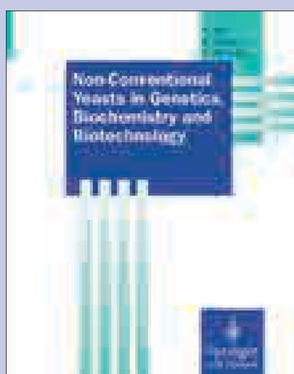

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Molecular Microbiology: Diagnostic Principles and Practice

David H. Persing, Fred C. Tenover, James Versalovic, Yi-Wei Tang, Elizabeth R. Unger, David A. Relman, Thomas J. White.
ISBN: 1-55581-221-X. 724 pp. List Price: $124.95.
Reviewed by Andrew Sails

In the past decade developments in molecular biology have led to dramatic changes in the way we detect and characterise microbial pathogens. The advent of nucleic acid probe-based assays and the subsequent development of nucleic acid amplification-based tests have led to what seems like an almost unending stream of new tests and assays being described in the literature. More recently technological advances such as the development of real-time PCR methods have facilitated the introduction of more rapid and sensitive assays for a wide range of pathogens. Application of these new developments in the routine clinical microbiology laboratory has required a new level of technical skill and understanding for laboratorians and clinicians.

Until now there has been no definitive and comprehensive reference text for this important subject. Molecular Microbiology: Diagnostic Principles and Practice attempts to fulfil this need.

The book is split into two sections, the first is diagnostic principles and the second is diagnostic applications. Diagnostic principles, reviews the technical aspects of DNA amplification, DNA probe technology, DNA sequencing, molecular fingerprinting and PCR product detection methodologies. Each of these techniques are presented and discussed in individual chapters written by outstanding experts in the field. The text of the chapters is supplemented by very good illustrations and tables which help to explain the techniques and comprehensive reference lists are also provided to direct the reader to further information. Interspersed between the technical descriptions of methods are short reflective chapters written by internationally renowned experts. These are very entertaining reviews

which introduce the reader to the background to some of the techniques presented in the accompanying chapters. In David Persing's imaginatively entitled chapter "Evolution of in Vitro Amplification Methods: the juice has been Worth the Squeeze" he reflects on the evolution and development of in vitro amplification methods. He emphasizes how proficiency testing and quality control in diagnostic molecular microbiology has become more important to ensure that accurate results are reported on patient samples. Fred Tenover from the CDC shares his thoughts on bacterial strain typing through the decades, from plasmid fingerprinting to the development of more discriminatory methods such as pulsed-field gel electrophoresis, and DNA sequence-based methods such as multi-locus sequence typing. The section ends with a very interesting chapter "Molecular subtyping for epidemiology" which argues the importance of standardisation of methods, quality assurance/quality control and makes recommendations for the interpretation of molecular subtyping data. The authors conclude that "molecular subtyping data are best interpreted in the complete context of epidemiologic, environmental, and laboratory investigations". Other chapters include technical descriptions of the recently described techniques such as multilocus sequence typing, genotyping using variable number tandem repeats, and automated ribotyping.

The second section of the book describes the application of molecular methods to the detection and characterisation of bacterial, viral, fungal and parasitic pathogens. The authors chose not to include specific laboratory protocols because they have become platform specific therefore several protocols would have to be included for each of the diagnostic applications. The detection and characterisation of bacterial pathogens section includes chapters on 16S ribosomal sequence analysis for identification of bacteria in the clinical laboratory, broad-range PCR for detection and identification of bacteria, and very topically the detection of agents associated with bioterrorism. The section on the detection and characterisation of viruses is equally authoritative and ends with chapters on emerging viral pathogens and the current and future trends in molecular virology. The sections on application of molecular methods to fungal and parasitic pathogens are a little short when compared the preceding sections however they are very well written. The recently emerged science of host genomics and pharmacogenomics is the subject for next four chapters and the book ends with chapters focussing on laboratory controls and standards, and external quality control and proficiency testing in diagnostic molecular microbiology.

Since this book arrived on my desk I have found myself "dipping into it" on a very regular basis, either to update myself on a technique or to clarify a method. I have also found myself recommending it to fellow laboratorians and clinicians. The price is slightly prohibitive to justify buying a personal copy unless you are very interested in this field however it does deserve a place on the shelf of any laboratory currently either performing or thinking about performing molecular diagnostic tests. Although this is a reference text the excellent standard of writing makes it a very enjoyable read, a rare thing for such a technical subject. The authors and editors should be commended for producing such a thorough text which should become the standard reference for the subject for many years to come.

PRIONS AND PRION DISEASE: CURRENT PERSPECTIVES

G C Telling (ed.) Horizon Bioscience 2004. ISBN: 0-9545232-6-1

Reviewed by Richard A Armstrong

The appearance of variant Creutzfeldt-Jakob disease (vCJD) a new subtype of the disease in the UK, and which may be linked to BSE, has brought prion disease to the attention of the public and media and resulted in a considerable increase in research into the basic properties of prions. Prions are proteinaceous infectious agents lacking DNA or RNA and composed largely of different forms of prion protein (PrP). The stated aim of this book is to provide a comprehensive review of the pathogenesis, molecular biology, biochemistry, cell biology, animal models, and immunology of prions. It is regarded as complimentary to an earlier volume "Prions: molecular and cellular biology" published in 1999.

The book begins (Chapter 1) with a contribution by Thompsett and Brown who review the function of normal cellular prion protein (PrP^{C}) and postulate that it is a copper binding protein. They conclude that PrP^{C} may have a role in ameliorating neuronal oxidative stress. There follows a contribution from Caughey *et al* (Chapter 2) who discuss the conversion of PrP^{C} to the scrapie form PrP^{Sc}, a process critical in the development of prion disease. They demonstrate that the pathological forms of PrP have a limited capacity of propagating themselves necessary for an infectious protein and also that PrP conversion is a highly specific process that may account for the presence of the species barrier. Brockes and Kanu (Chapter 3) then review the studies of PrP in culture including glycosylation, conformation, membrane topology, aggregation, and disulphide binding of PrP, all of which are relevant to the study of the conversion of PrP^{C} to PrP^{Sc}.

A particularly intriguing finding is that one cell may infect a neighbour by cell contact. In chapter 4, Supattapone and Rees review the role of deletion mutants of PrP in studies of the folding and neurotoxicity of PrP and conclude that different portions of the molecule are involved in prion propagation, amyloidosis, and neurotoxicity. The next two chapters review various aspects of the PrP gene including the study of PrP polymorphisms in mice (Barron and Manson) and transgenic mouse studies (Nazor and Telling). There follows a consideration of a very interesting topic by Aguzzi (Chapter 7), *viz.*, peripheral pathogenesis, which discusses the role of M-cells, Peyer's patches, and the sympathetic nervous system in the migration of prions through the body. The immunological advances relevant to PrP are discussed by Williamson (Chapter 8) and demonstrate that specific antibodies that bind to PrP may be an important means of preventing PrP replication in the body by either hindering the conversion of PrP^{C} to PrP^{Sc} or by stabilising PrP^{C}. Westaway (Chapter 9) discusses the PrP related protein 'doppel' that has a similar structure to PrP and is expressed in the testis. Interactions between PrP and doppel may occur in the body and contribute to disease.

The book concludes with a contribution by Chernoff (Chapter 10) who discusses the contribution that studies of PrP formation and propagation

in yeast cells have made to our understanding of the molecular mechanisms of amyloidosis and the general principles of protein-based inheritance.

All these contributions are relevant, topical, and well written with good illustrative material and the book certainly fulfils its stated objectives. I would have liked to see a more detailed treatment of the controversies surrounding the role of prions in disease and also of the sequences of events leading to the characteristic neuropathology of prion disease, viz., spongiform change, neuronal loss, gliosis, and the deposition of PrP aggregates. Nevertheless, all researchers with an interest in prions and the diseases they purport to cause will need to read this book.

Pathogenicity islands and the evolution of pathogenic microbes. Contemporary issues in microbiology and immunology vols. 264/I & 264/II.

J Hacker & J B Kaper (Eds.) 2002. 231/208pp.
ISBN 3-540-42681-7/3-540-42682-5.
Springer-Verlag Berlin, Heidelberg, New York. £81.00 /£77.00
Reviewed by Simon Hardy

Anyone who has read any of the Contemporary Issues in Microbiology & Immunology series will know that these are thorough, ''state of the science'' volumes. The top drawer quality is maintained in this two volume release on pathogenicity islands and the impact of their transfer. These topics have arisen from the explosion of microbial genome sequencing in the last 10 years. The idea that virulence determinants are more frequent in pathogenic isolates than commensals or environmental isolates is far from new. The microbiological literature has a long history of studies comparing selected phenotypic traits in strains from differing sources in a bid to identify virulence factors. For example does haemolysis in *E. coli* predominate in the invasive strains compared to those isolated from the bowel of healthy individuals? This type of enquiry is now being replicated within genomes, looking for the atypical stretches of bases of distinct G:C ratios as the genetic locations of the virulence traits. Pathogenicity islands (PAIS) have characteristics consistent with the interpretation that they are mobile genetic elements, able to move horizontally between species (horizontal gene transfer HGT). These characteristics include atypical G+C ratios bounded by genetically mobile IS elements, close proximity to tRNA genes and a high frequency of loss from cultures (the mechanism of this loss is not at all clear). This list is not complete and the characteristics of the PAIS from different species and how they compare is one of the key features of the books.

The editors start their introduction with the famous statement of Dobzhansky that ''nothing makes sense in biology except in the light of evolution''. The first chapter of Volume 1 is ''Evolution of prokaryotic genomes" by W. Arber (Basel) which briefly refreshes the reader of the principles of gene exchange in the context of how they contribute to molecular evolution. An entirely appropriate chapter, it is dense with interesting leads for further thought. As subsequent chapters raise the topic of genome reduction in pathogenic organisms I was hoping for but disappointed to see no discussion of possible mechanisms for reducing genome size in bacteria. In fact, the chapter does not directly address pathogenicity islands themselves but this is not necessarily a problem given the concentrated focus of the following chapters.

Volume 1 concentrates on PAIS found in human pathogens (represented by the genera Salmonella, *Escherichia (coli)*, *Shigella*, *Yersinia*, *Bordetella*, and others). The trend is similar in volume 2 but a wider perspective is provided by chapters on plant pathogens, animal pathogens (*Dichelobacter nodosus*) and fungal pathogens. The discussion on fungal genomes is particularly welcome and very useful perspective of the difficulties in genetic manipulation eukaryotic genomes present. The final chapter examines the ''Impact of integrons and transposons on the evolution of resistance and virulence'' which again extends the coverage to transfer of antibiotic resistance genes.

A cursory glance at the title might suggest that the book has little to offer those not working on PAIS or genome analysis. It is not so. The content of the chapters, perhaps not surprisingly, varies in emphasis and regularly throw up interesting perspectives outside of the remit of the title. The chapter on Bordetella includes a good review of the taxonomy of this genus. The chapter by Reeves and Wang, (Sydney) on lipopolysaccharide covers both LPS structure, biology and its genetics in *E. coli* and *Salmonella*. The overview of LPS biology, gene sequencing and its analysis is fascinating and an intriguing comparison with mobile pathogenicity islands is given.

The central theme of the book is best summarised by Dobrindt, Hentschel and the editors (Wurzburg & Baltimore) entitled ''Genome plasticity in pathogenic and non pathogenic Enterobacteria''. There central tenet is the diversity in pathogenic potential of E. coli provides an exemplar of the impact of a flexible gene pool supplying virulence factors to an organism. As the editors were early workers recognising the existence of PAIS it is to be expected that the chapter provides a compelling argument for the role of mobile gene pools in driving evolution and creating specialised pathotypes in *E. coli*.

The increasing ease with which genomes can be sequenced has resulted in numerous review articles in the literature highlighting the horizontal spread of genes across genera as well as species. Whilst the impact of such events on microbial taxonomy is potentially enormous, a less evangelical viewpoint can be adopted (eg Kurland. 2000, EMBO Reports 1; 92-5). So it is a shame that there is no chapter giving voice to more sceptical views on the extent to which horizontal gene transfer has occurred.

Books in this series provide no practical methodologies but aim to review current opinion on specific topics. Readers of this book will be confident they have read the most thorough work on the topic, and more.

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Rumen Microbiology

Burk A Dehority 2003. Nottingham University Press. ISBN: 1897676999 Price: £40.00

Reviewed by Andrew McBain

Over the past decade I have developed an interest in mammal-bacterial symbioses, yet my "hands-on" experience of this type of system has been restricted to those microbial ecosystems residing in the human large bowel, the oral cavity and on the skin.

Whilst studying human gut bacteria for my doctorate I became aware that many of the techniques used in this area of microbiology had been borrowed or adapted from the early work done by rumen microbiologists, half a century before. The opportunity to review this book has given me the chance to find out more about this fascinating system.

Herbivores, especially ruminants are able to digest cellulose and have adapted to exploit a diverse array of temperate and tropical vegetation, enabling them to become one of the worlds most widely distributed group of mammals. From a microbiologist's perspective, the rumen can be considered as a large natural fermenter in the foregut of the ruminant, holding up 200 litres of material. Microorganisms within are provided with anaerobiosis, constant temperature, mixing and pH control. Masticated substrates are fed into the system and fermentation products are either absorbed in the rumen itself or pass downstream for further digestion and absorption. Rumen microbial populations can provide up to 80% of a dairy cow's daily protein and energy requirements. From an agricultural perspective, the massive importance of this system to the human diet should not be underestimated. Rumination also contributes significantly to the production of greenhouse gases. Rumen microorganisms are highly diverse and, arguably, more than any other mammalian-associated consortium, the rumen brings together the conjoint activities of bacteria, protozoa and fungi.

"Rumen Microbiology" is primarily targeted towards undergraduate and graduate students and aims to offer a modern alternative to the influential and widely referenced book by R.E. Hungate, "The Rumen and its Microbes" (1966). "Rumen Microbiology" is apparently used as the major source of information presented to students in the rumen microbiology course at the Ohio State University. There is certainly a wealth of information in the current book. The fact that bacteria are not specifically covered until chapter seven is possibly a reflection of the microbial diversity of the rumen, and the importance of the protozoa, but it also relates to the fact that from a student's perspective, examining rumen contents for the first time, the protozoa are the most easily recognized and enumerated organisms.

There is an instructive chapter on rumen fungi. Apparently, *Neocallimastix sp*. and others contribute significantly to the breakdown of plant material. This is interesting from a human gut microbiologist's perspective, since the microflora of the human colon is considered to be overwhelmingly bacterial in composition, with only low numbers and activities of the other microbial groups.

As is required for any text concerned with mammalian-microbial interactions, the book covers fundamental issues of host physiology, such as the gross anatomy of the ruminant stomach, as well as microbial physiology and ecology. As required for a student text, there is a useful appendix describing eight simple-yet-elegant experiments that could be done in any teaching lab. Like any good teaching text, there are many clear line drawings and figures, and the lack of colour images does not significantly impact upon the books usefulness.

The writing style is clear and the overall quality of the science and the editing is good. I found the format, whereby sources of information are comprehensively referenced at the end of each chapter to be useful. This reader however, would have appreciated some information on the relative numbers of bacteria, fungi and protozoa that occur in the rumen, either in the introduction or one of the early chapters. From my perspective, one of the best ways of appreciating the significance of any host-associated flora is to know the relative abundances of microbial cells, which can outnumber host cells by orders of magnitude.

There is little information given on molecular methods for the study of rumen microbial physiology or micro-ecology. Whilst most students may benefit most from a firm grounding in basic aspects of a subject before considering advanced approaches, the pioneering molecular ecological work by rumen microbiologists could perhaps have been capitalized on in this text, even in the form of a basic introduction to the techniques available.

Retailing at $40.00 (on Amazon), it represents good value for money for a hardback text, and would serve well as a reference book for a student course, or for anyone wanting to find out more about this complex and fascinating ecosystem. It certainly succeeds in its aim to give students an insight into the "black box" of the rumen.

Modern Medical Microbiology — The Fundamentals

Stuart Clarke (2003) Hodder Arnold ISBN: 0340810440. Price: £19.99

Reviewed by Val Edwards-Jones

Modern Medical Microbiology — The Fundamentals is a text aimed at students of medical microbiology, trainee Biomedical Scientists, and undergraduate medical students. It is the culmination of a number of articles written by Dr Stuart Clarke over an eight year period.

It is divided into five different parts, Bacteriology, virology, parasitiology, mycology and current topics covering some interesting sections on emerging and re-emerging disease, antibiotics, probiotics, bioterrorism as well as pertinent infectious diseases.

The text is very easy to read and each chapter is only about 2-4pages in length with the key points in highlighted boxes. There are very few diagrams or pictures which I found made the book appear a little drab (personally I always like pictures, it helps you remember some of the horrible sequale of infection). However, the key information for each particular topic is well summarised and there are key references listed. Also, extremely useful are the listed web-site references.

This is a good basic foundation text in medical microbiology for trainee biomedical scientists and other applied microbiologists.

Microbiology, Principles and explorations, 5th edition, 2002

J.G. Black. John Wiley & Sons Inc
New York. ISBN: 0-471-38729-0
Price: £ 32.50
Reviewed by: Evdoxios Psomas

This book meets the needs of students in the health sciences and biology as well as those enrolled on other science programs who need a solid foundation in microbiology. It serves both audiences very well — in part by using an abundance of clinicaly important information to point out the principles of microbiology and by offering a wide variety of additional applications.

The book is divided into seven units and each unit is subdivided into two to six chapters each. A quite useful characteristic of the book organisation is that each unit begins with an interview with a working microbiologist. These descriptions of real-world applications of the topics covered in the unit to come, stimulate the reader's interest. Furthermore the book is full of clear, attractive drawings and carefully chosen photographs that contribute to the student's understanding of a specific subject. Illustrations amplify and enhance the narrative. In addition to narrative that is direct and accurate, the reader will find pieces of humor and personal stories that facilitate in the study of the contents. At the end of each chapter are some useful resources for the student. A chapter summary entitled "Retracing our steps" serves as a map about the facts and concepts covered in the chapter. I found the "critical thinking questions" very interesting as this section goes further than simple recall and tests the understanding of the basic fundamentals of the chapter.

The first unit introduces the reader to the fundamentals of microbiology such as microscopy, staining and the important characteristics of prokaryotic and eukaryotic microorganisms. This unit is useful as a basis for student's revision of basic concepts which may have been taught at the start of their course.

Unit two covers microbial metabolism, growth and genetics providing a lot of useful information about the essential concepts of metabolism, the growth and culturing of microorganisms and microbial genetics with an interesting chapter that deals with mutations.

The third unit is quite descriptive and takes a detailed look at the taxonomy of bacteria. In addition the sections on viruses and eukaryotic microorganisms contain enough detail to be suitable for an undergraduate level course.

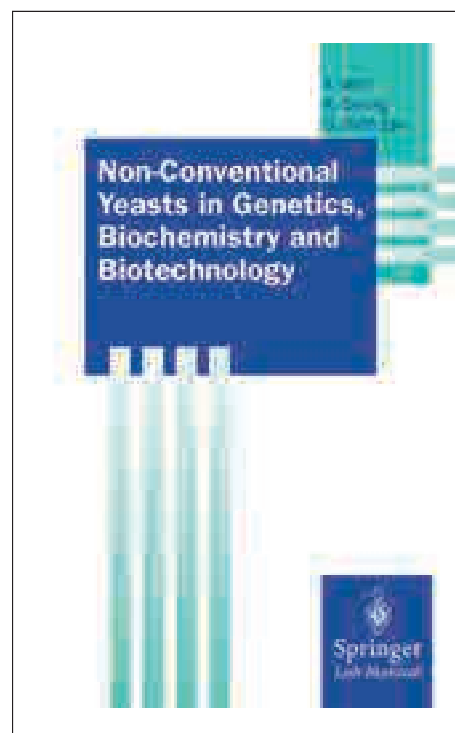
The fourth unit offers a well written review of how to control microorganisms with a chapter devoted to antimicrobial therapy especially the properties and the attributes of an antimicrobial agent.

The topic 'Host-Microbe interactions' in unit five features disease processes due to host-microbe relationships, nosocomial infections and a very well written two-chapter section about immunology.

The sixth unit is a detailed text about human infectious diseases. The first two chapters of the unit highlight diseases of the skin, eyes and urogenital system whereas the remaining four chapters cover oral and gastrointestinal diseases, those of the respiratory system and those of the cardiovascular and lymphatic systems.

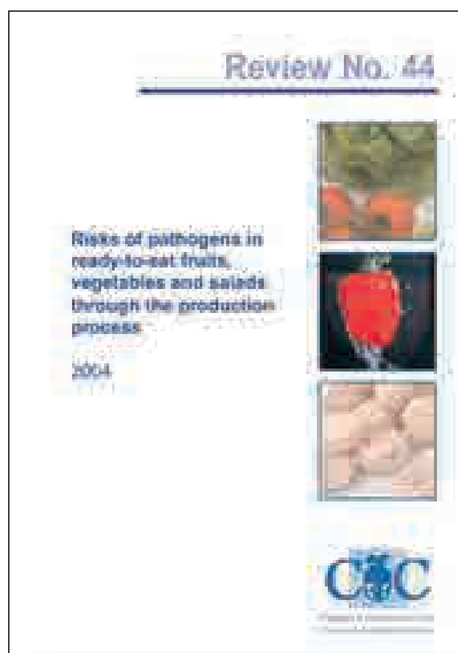
The final unit covers environmental and applied microbiology. Fundamentals of environmental microbiology with reference to air, soil and water are well described. Furthermore aspects of applied microbiology are well presented in easily digested sections. Food microbiology topics along with industrial and pharmaceutical topics are covered in the last chapter of the unit.

This textbook, unlike many other similar publications offers a clear, simple and comprehensive overview of Microbiology. For students this would be an additional source of knowledge, not only for class but also in their preparation for laboratory reports and exercises. It is an excellent publication and I recommend it highly.



Would you like to review a book for ***Microbiologist*** and get to keep it? There is an up-to-date list of titles available for review on the Society's website at:

www.sfam.org.uk/pubs/books.php



Pathogens on fresh and prepared produce

The risks posed by pathogens on fresh produce are the subject of an extensive new review from CCFRA. Risks of pathogens in ready-to-eat fruit, vegetables and salads throughout the production process looks at each of eight significant pathogens and their likely survival on fresh produce. It looks in detail at the routes of microbial contamination and points to sources of guidance on how to help to manage the risks that pathogens pose. It will be of value to anyone working in the fresh produce sector with an interest in, or responsibility for, product safety assurance.

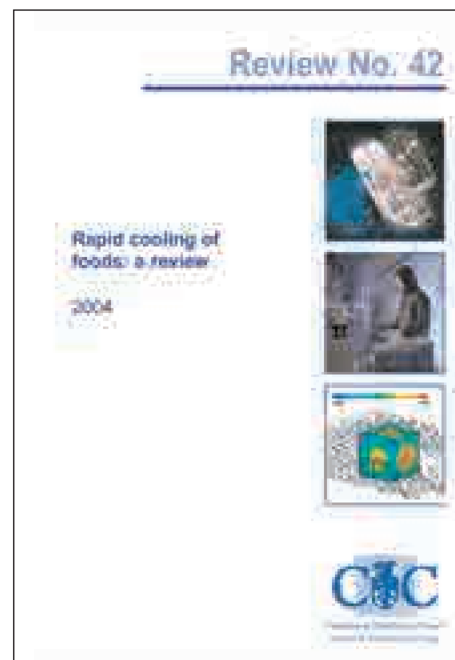
In recent years, there has been a large increase in demand for, and consumption of, fresh prepared and minimally processed fruits, vegetables and salads. Industry has responded to consumer demand by creating a wide range of fresh and minimally processed products, incorporating raw fruits and vegetables that have been cut, trimmed, peeled and washed, for consumption raw without further preparation, washing or cooking.

As with processed products, good manufacturing practice is essential in assuring the safety of these fresh prepared products, with particular attention being paid to potential routes of contamination and the risks associated with foodborne pathogens.

Edible films and coatings

The opportunities afforded by edible films and coatings in food production, preservation, packaging and new product development are the subject of an extensive new review from CCFRA. This review examines the main functions of edible films and coatings and describes their applications with a diverse range of products. The review will be of value to anyone working with products where edible films have potential applications and particularly to technical personnel and product developers who wish to understand the current and potential applications of the technology.

Although some edible films have been around and in use for several years, the technology is now more widely used as the variety of available materials and their properties has broadened. Films and coatings can now be based on polysaccharides (e.g. cellulose, starches and gums), lipids (cocoa butter and waxes) or proteins (e.g. from milk, soya, cereals). As well as providing a physical barrier and protective coating, edible films can help retain moisture and restrict its movement, limit fat migration between layers, provide a barrier to oxygen, act as an adhesive, help to trap flavour and aroma, and carry and present antioxidants or antimicrobials.



Rapid cooling of foods

This review will help technical and other product development personnel in chilled and frozen food companies to identify and potentially exploit new technologies for the rapid cooling of foods. By pulling together information from the scientific literature, the trade press and web-published articles, the review describes a range of emerging developments and novel methods for the rapid cooling of foods. These include, for example, plate and air blast chilling and freezing, immersion chilling and freezing, vacuum cooling, pressure shift freezing, dehydrofreezing, cryogenics, ice slurry methods, and novel cooling systems such as hydrogen, vortex, Peltier and heat pipe cooling.

It also outlines the importance and potential benefits of rapid cooling and discusses its theoretical basis before considering the effects of rapid cooling on the food product - with particular emphasis on meat, poultry and fish. The list of references provides useful pointers to source information, to help the reader explore areas of interest in more detail.

Further Information

Copies can be obtained from Mrs Sue Hocking, Publications Officer, CCFRA, Chipping Campden, Glos, GL55 6LD, UK
Telephone: +44(0)1386 842225
email pubs@campden.co.uk

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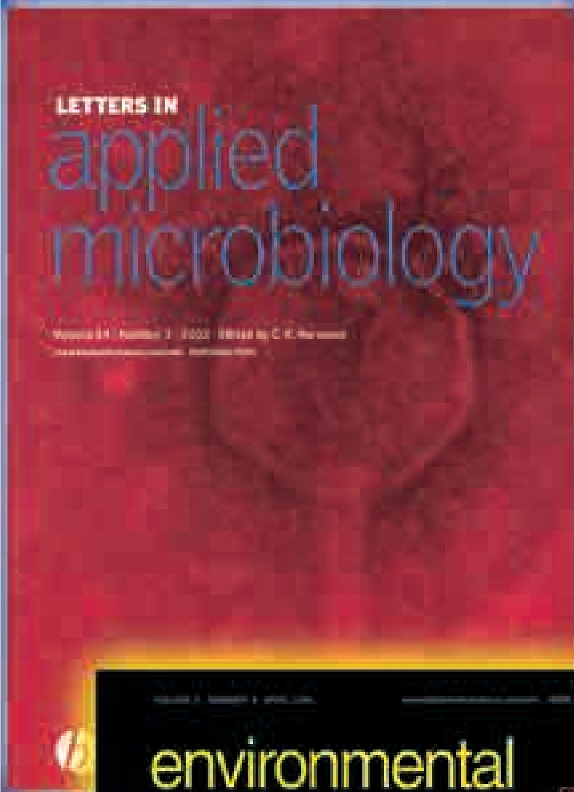

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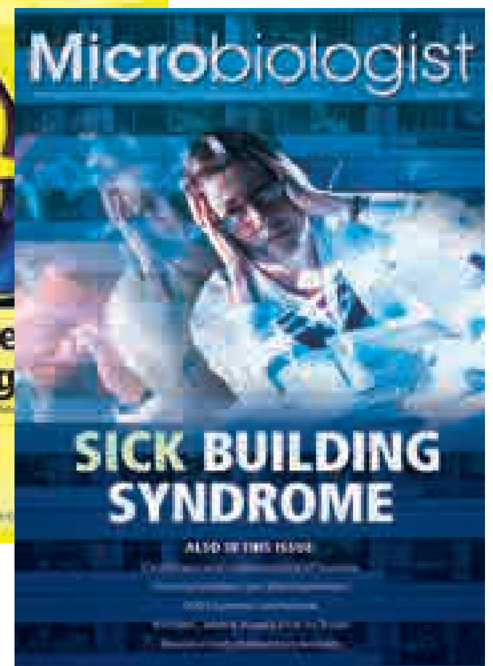

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advancing the science of microbiology

For more information about the Society, Society meetings, the benefits of membership, *Microbiologist*, or **to join us** please visit the Society website at www sfam.org.uk

The Society for Applied Microbiology was founded in 1931 and is dedicated to advancing the study of microbiology. Society members play a leading role in shaping the future of applied microbiology, and enjoy many benefits, including:

- **Reduced rates at Society meetings**
- **Access to the members areas of the Society website**
- **Generous grants and awards**
- **FREE access to three acclaimed journals**

Detailed information about all these benefits and more can be found on the Society website.

WEBSITE:
www.sfam.org.uk

The website is the best source of detailed information on the Society and it's many activities. It has a lively discussion forum and fully interactive membership areas where you can book your place at Society meetings find and advertise jobs, display your CV and much more.

CONTACT:

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Benefits of sfam membership

membership options

Full membership gives online access to the ***Journal of Applied Microbiology***, ***Letters in Applied Microbiology*** and ***Environmental Microbiology***, copies of ***Microbiologist***, preferential registration rates at Society meetings and access to the members areas of the website.

Full student membership confers the same benefits as Full Membership at a specially reduced rate for full time students not in receipt of a taxable salary.

Associate membership this new class of membership is open to all current and new Society members including existing Associate Student Members and Retired members and gives quarterly copies of ***Microbiologist*** and preferential registration rates at all Society meetings.

Honorary Membership of the Society is by election only and this honour is conferred on persons of distinction in the field of applied microbiology.

Corporate membership is open to all companies with an interest in microbiology. Corporate members benefits include:

- Online access to the Society's three journals OR hard copies of the journals.
- Half page advertisement in each quarterly issue of ***Microbiologist*** (which can be upgraded to a larger size at very attractive discounted rates).
- Full page advertisement in the Members' Handbook.
- 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).

Meetings

We hold two annual meetings. The January Meeting comprises discussion sessions with the opportunity to display posters on related work. The Summer Conference is held every July and comprises a main symposium, a poster session, the AGM and a lively social programme. We also hold occasional joint ventures with other organisations on topics of mutual interest.

Publications

The Society publishes two monthly journals: ***Journal of Applied Microbiology*** and ***Letters in Applied Microbiology.*** We also produce our own quarterly in-house colour magazine: ***Microbiologist***, which contains features, reports topical news stories and full details of our meetings. The Society is also a partner with Blackwell Publishing in the bi-monthly journal ***Environmental Microbiology.***

Online journals

Synergy is an online service provided by Blackwell Publishing that gives Full and Student Members **FREE** access to the online versions of the Society's three journals: ***Journal of Applied Microbiology***, ***Letters in Applied Microbiology*** and ***Environmental Microbiology***. Members can register for this service at http://www.blackwell-science.com. Members can also submit papers directly to our journals via an online submission service.

For more information about Synergy or online manuscript submission, please visit the website.

Grants & awards

Many 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 Lectures and the popular **Students into Work** Scheme.

Full details of all the Society's grants and awards can be found on the website together with easy-to-use online application forms.

Special interests

The Society has six very active Interest Groups:

Bioengineering, Educational Development, Environmental, Food Safety and Technology, Infection, Prevention and Treatment, Molecular Biology

Detailed information about these Groups can be found on the Society website.

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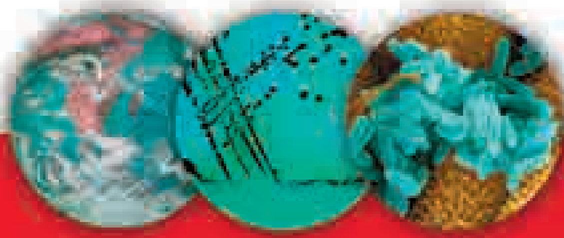

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