MICROBIOLOGY TODAY

QUARTERLY MAGAZINE OF THE SOCIETY FOR GENERAL MICROBIOLOGY VOLUME 27 NOVEMBER 2000

Contents

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February 2001 issue 2 January
May 2001 issue 30 April

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Above: Microbiology – the twilight zones
Courtesy Carlin Iverson/ Science Photo Library

Vol. 27, Part 4, November 2000

In this issue we try to answer questions such as how small IS the smallest unit of life? Is it really alive? Is it really a micro-organism?

In his overview of the twilight zones of microbiology (p. 162) John Postgate wonders if what now may seem to be biological curiosities will lead to dramatic new scientific advances.

What causes TSEs? Scientists disagree but most favour the prion protein theory discussed by Chris Bostock on pp. 164–166.

Mobile genetic elements are found widely in nature. On pp. 178–180 Nicholas West and Christoph Tang look at the role of prokaryotic transposons in adaptive evolution and their potential for exploitation by microbiologists.

Allegedly the smallest replicating molecules known, viroids cause devastation in crops, as described by Nicola Spence and Dez Barbara (pp. 168–170). Bacteria have viruses too. Gunnar Bratbak and Mikal Heldal consider the role of bacteriophages in aquatic microbial ecology on pp. 171–173.

Moving up the scale, Allan Hamilton looks at nanobacteria (pp. 182–184) and John Fry questions just how unculturable the microbes only identified by molecular biology techniques really are (pp. 186–188).

And finally, Don Cowan and Monica Grady consider the possibilities of extra-terrestrial life (pp. 174–177).

Other topics covered include the SGM's new initiative to raise the profile of microbiology (p. 200), culture collections in Cuba (p. 189) and a Lancashire attempt to interest children in microbiology (p. 195).

These articles appear in addition to all the regular features and reports of Society activities.

Articles

Regular Features

Other Items

The twilight zones of microbiology

John Postgate

Professor John Postgate, FRS, author of the well known book *Microbes and Man*, Honorary Member and former President of the Society, looks at why the 'twilight zones of microbiology' should not be ignored.

In 1674, Antoni Leeuwenhoek first described to the Royal Society his microscopic animalcules, creatures so small as to be invisible to the naked eye. His findings were received with lively interest, but they made no radical impact, because creatures close to the limits of optical resolution were already known, and his protozoa were on the margin of that limit. It was his letter of October, 1676, which astonished the Society, for the 'tiny eels' he saw, which we now call bacteria, were vastly smaller than even those earlier creatures. The subsequent story is now history. Suddenly, size-range of living things had been extended downwards by two orders of magnitude of length. It was a significant advance in our understanding of the living world, but that was all. It would be another couple of centuries before the formidable impact of those animalcules on scientific thinking began to emerge.

In the mid-nineteenth century the first hints of that impact appeared, when Robert Koch demonstrated the roles of bacteria in disease and when Louis Pasteur elucidated the function of yeasts in fermentation – and almost in passing discovered anaerobiosis. Medical and applied microbiology grew vigorously, and alongside them grew general and environmental microbiology. Long-held beliefs, such as spontaneous generation, or the roles of vapours and humours as agents of disease, withered rapidly; over the next few decades our views of ourselves and the world around us were transformed with the revelations that we eat and breathe microbes, that we are hosts both inside and outside ourselves to a menagerie of microbes; that microbes are fundamental to agriculture and many more of our economic activities; to the terrestrial cycles of the bio-elements, and even to the persistence of macroscopic life on this planet. Leeuwenhoek's letter of 1676 had seeded not just a vast explosion of both practical and scientific knowledge, but a revolution in the way we see ourselves and the biosphere.

That was the longest lag phase in the history of microbiology's intellectual impact. But within our specialized areas we can all point to recent examples of shorter lags, when an offbeat or seemingly trivial report took years, even decades, to underpin a substantial intellectual shift.

Examples feature in this issue, in which we peer into today's twilight zones of microbiology, into areas of research which seem to be out of kilter with the mainstream of thinking among microbiologists. Will their eventual illumination bring about radical revisions of our thinking?

Reflect on the history of prions. In my distant youth the scrapie agent was recognized as infectious and called a 'slow virus'; then as general virological biochemistry and molecular biology progressed it became clear that it was something exceptional. Now we are invited to regard it as a partly denatured form of a normal protein which has become infectious and catalyses or otherwise promotes further denaturation of its native form. Not happy with that? Nor am I. But the probabilities are that we shall have to come to terms with it, just as mid-twentieth-century microbiologists had eventually to accept that, contrary to the orthodoxy of the time, bacteria possessed chromosomes.

So, are prions alive? That may seem a silly question, because it has long been obvious that linguistically the borders of animate and inanimate matter are fuzzy, and that around those borders the term 'living' becomes imprecise. Yet fuzzy terminology leads to fuzzy thinking; we cannot wait for a precise definition of life, so we need an operational consensus on how elementary a biological entity can be and still be regarded as alive. Leeuwenhoek and his contemporaries had no problem here, and even when the discovery of viruses early in the twentieth century extended the size range of living things downwards yet again, there seemed to be no serious doubt that they were alive. However, in the 1930s the question became abruptly serious with the crystallization of tobacco mosaic. Well, we said, those crystals are just a special case of dormancy; or viruses are obligate parasites that have lost many of life's defining properties. On the whole viruses scored as living. But what now of viroids, fragments of infectious RNA too small to have even a plausible genome? Or self-transposing genetic elements? Are autonomous fragments of hitherto living material to be regarded as alive?

Hold it there for a moment and switch channels to another twilight area: those 'unculturable' microbes which PCR coupled with rDNA probing, and conventional c.f.u. counts on wild populations, show us are abundant in this planet, even deep beneath its surface, as well as in soils and sediments. Are they alive?

Vegetative microbes that are physiologically alive but do not multiply in conditions offered by microbiologists are commonplace in sediments, soil, old cultures and so on. As well as 'unculturable' they have been called 'non-culturable', 'dormant', 'senescent', 'moribund', 'non-viable', 'viable but non-culturable'; all names which beg questions about their physiological state. They too exist in that fuzzy zone between animate and inanimate things; therefore meticulous precision of language is needed when discussing them (especially now that interplanetary exploration is raising a serious prospect of extra-terrestrial microbiologies within the solar system). Current attempts to seek an agreed terminology for microbial cells in that state (e.g. Barer & Harwood, 1999) are laudable. Is not a rational terminology for discussing comparable non-cellular entities also needed?

The twilight zones of microbiology may seem to involve little more than biological, and sometimes philosophical, curiosities. But history tells us that, as with Leeuwenhoek's animalcules, their exploration will indeed disclose dramatic new insights, and will probably lead to scientific advances reverberating far beyond microbiology.

Further reading

Barer, M.R. & Harwood, C.R. (1999). Bacterial viability and culturability. *Adv Microb Physiol* 41, 93–137.

microbiology

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The nature of TSEs

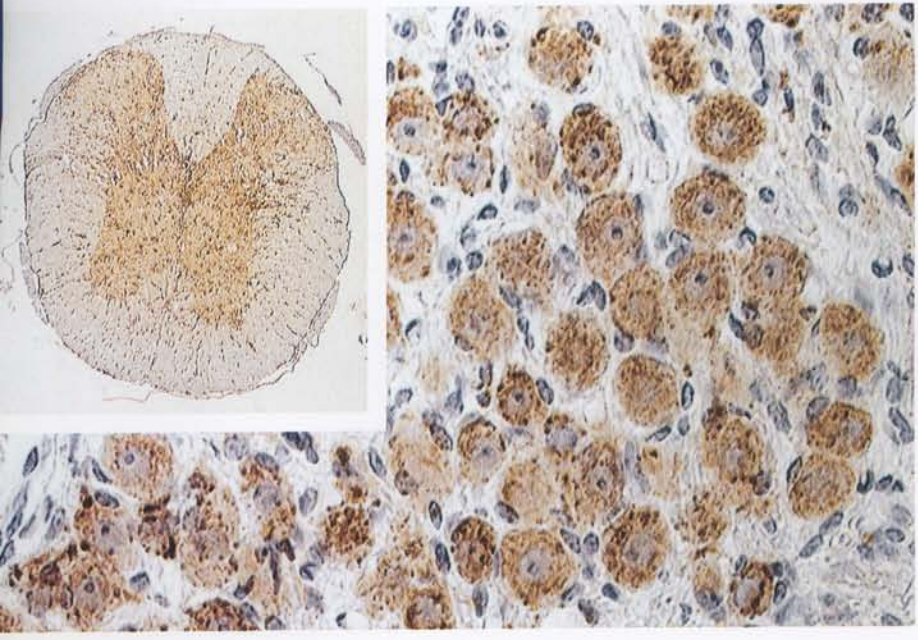
Chris Bostock

Scientists are unsure of the identity of the causative infectious agent of TSEs, but the properties are unlike those of conventional pathogens.

Creutzfeld–Jacob disease (CJD) and bovine spongiform encephalopathy (BSE or mad cow disease) both belong to a group of diseases called transmissible spongiform encephalopathies (TSEs) or prion diseases. BSE and the new variant of CJD (vCJD) have been in the news recently and are new additions to the group, but, as a whole, TSEs have been around for centuries. Scrapie in sheep was first recorded over two hundred years ago and sporadic CJD was first described in humans in the 1920s. Nevertheless, the TSEs remained a rather obscure group of diseases until 1986 when BSE first appeared. In the years since the emergence of BSE, new TSEs have also been found in exotic species of ruminants in UK zoos, exotic and domestic cats and, in 1996, vCJD was described in humans.

The TSEs are fatal once clinical signs appear, but there is a long, usually many years, incubation period between the time of first exposure to the infectious agent and the time of appearance of disease. A normal microbial infection usually elicits an immune response in its target host, but in TSEs there is no conventional immune response, although the immune system plays an important part in the development of the disease, before the infectious agent gets into the central nervous system (CNS). A common feature of all TSEs is degeneration in the brain and spinal cord and part of this process involves a normal host protein, called PrP or prion protein. During a TSE infection this protein is deposited in an abnormal form and in excessive amounts in the brain, spinal cord and many peripheral nerves and tissues (Fig. 1). The causative infectious agent has properties unlike those of conventional pathogens and may be a new class of infectious agent. *

BELOW:
Fig. 1. Abnormal prion protein can be detected as brown deposits following staining with antibodies to prion protein in a section through part of the spinal cord taken from a hamster infected orally with the 263K strain of scrapie (inset). The abnormal accumulation of prion protein can be seen at higher magnification in individual neurons in the pathway that connects the ear to the brain.
COURTESY TRICIA McBRIDE



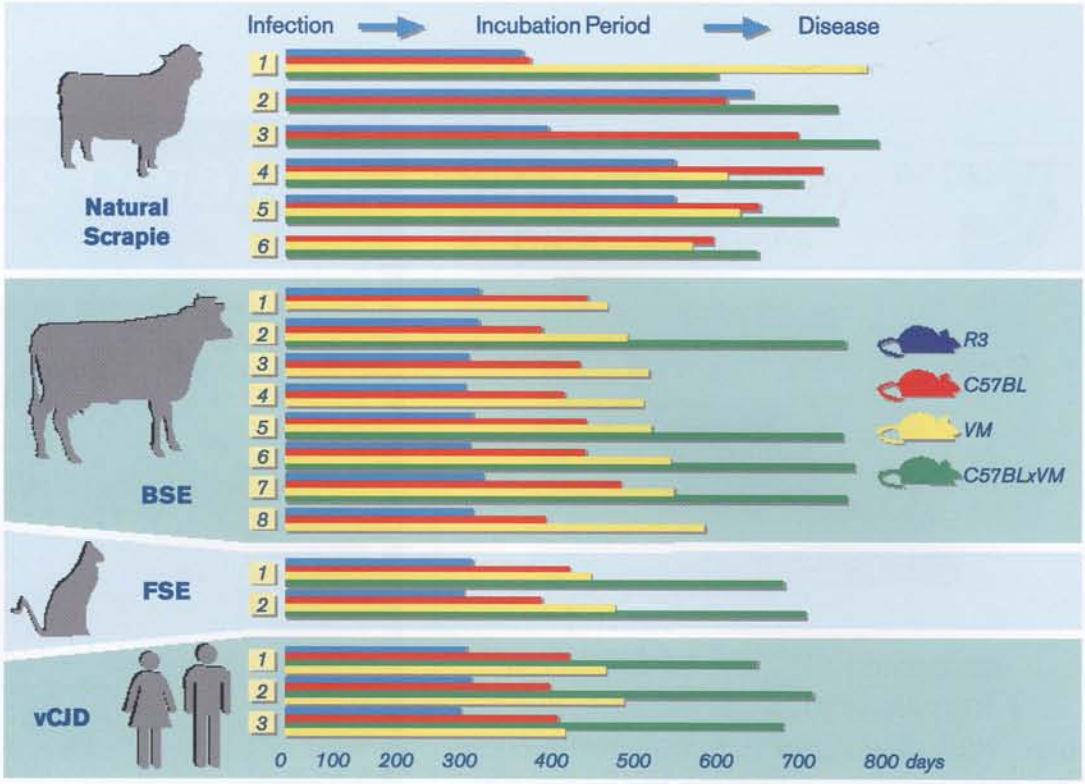
The nature of the infectious agent

There are currently three theories. One says that the infectious agent is somewhat like a small conventional virus, with genetic material coding for its own proteins. At present there is little evidence to support this theory. The second, and now most widely held theory states that it is the altered physical state of normal host prion protein, which is able to propagate itself by inducing other normal prion protein molecules to adopt the abnormal conformation. This is commonly referred to as the prion hypothesis. The third idea, called the virino hypothesis, is most easily thought of as a hybrid of these hypotheses. It suggests that there is a very small piece of genetic material, which encodes only information for its own survival and replication through interaction with a host protein, perhaps the prion protein, using it also as a protective coat. The essential, but very important difference between the prion and virino hypotheses is that in the prion hypothesis, all the information to determine the properties of the infectious agent is carried in the abnormal conformational state of the prion protein, whereas in the virino hypothesis, the information is carried in an independent 'genetic' molecule. As yet no-one has defined the precise molecular state(s) of the normal and infectious forms of the prion protein, nor produced infectious 'prions' *in vitro* from normal prion protein, as would be predicted by the prion hypothesis. But neither has anyone yet found any molecule that might fit the role of the 'virino'.

Prion protein genes

As with conventional microbes, there are different strains of TSEs, and the characteristics of an infection are determined by an interaction between a particular strain of infectious agent and a gene (or genes) in the host animal. The host gene that has the biggest effect in determining the outcome of an exposure to an infectious agent is the same gene that codes for the normal prion protein. In humans there are two common versions of the prion protein that differ in a single amino acid (whether it has valine or methionine at coding position 129), but there are also many rare mutant forms, most of which are associated with inherited susceptibility to prion disease. Sheep also have many different forms of the prion protein, none of which have so far been linked to 'inherited' scrapie and only some of which appear to affect the outcome of a scrapie infection. Two versions of the prion protein have been found in cattle but these do not appear to differ in their association with susceptibility of animals to BSE.

Much of the work that has lead to an understanding of the role of the prion protein gene in TSEs was done in laboratory mice before anything was known about the existence of the prion protein or its gene. Different lines of inbred mice were found to differ in their response to infection with scrapie and a gene, called *Sinc* (for scrapie



*inc*ubation period), was identified that controlled the incubation period. There are two versions of *Sinc*, s7 and p7, which determine, respectively, short and prolonged incubation after infection of mice with a strain of scrapie called ME7. *Sinc* turns out to be the gene that encodes the prion protein. The difference between s7 and p7 *Sinc* genes is now known to lie in the amino acids at two positions in the protein; if it has leucine at position 108 and threonine at 189 it is *Sinc* s7, whereas if it has proline at position 108 and valine at 189 it is *Sinc* p7.

Properties of different strains of infectious agents

In addition to the different versions of the prion protein gene within a species there are many different strains of the infectious agent that can infect that species. This is most clearly demonstrated by infecting the same inbred lines of *Sinc* s7 or *Sinc* p7 mice with different sources of scrapie. Different sources of scrapie can produce widely different incubation periods and result in differing patterns of damage in the brain (called lesion profile). These two features, relative incubation periods in s7 and p7 mice and the lesion profile, are classically used to define the different strains of infectious agent. Several different strains of TSE agent can be propagated in the same inbred line of mouse and thus, if the abnormal conformation of the prion protein determines the properties of the infectious agent, this means that the same normal prion protein must be able to adopt reproducibly several distinct abnormal disease-associated states.

Using the criteria of relative incubation period and lesion profile it has been possible to characterize and compare the infectious agents causing contemporary scrapie, BSE, the spongiform encephalopathies of cats (FSE), kudu and nyala, as well as classical and vCJD of humans (Fig. 2). Sources of scrapie, collected contemporaneously with the BSE epidemic, are heterogeneous, each being distinct and different from the others. This situation contrasts with the homogeneity found in several different sources of BSE sampled at different times during the epidemic and from different geographical locations. The infectious agents causing FSE and disease in exotic ruminants in zoos were found to be indistinguishable from the BSE strain, and, although classical CJD is quite distinct, the transmissible agent causing vCJD has the same strain properties as the BSE agent. This suggests that the agent that causes vCJD is the same as that which causes BSE, but as yet there is no firm evidence to say how humans became infected. From a scientific point of view, one of the interesting observations to come out of this work is that the same strain of agent (BSE) can be propagated by several species, each of which has a slightly different normal prion protein. Thus, if the conformation of the abnormal form of the prion protein determines the biological properties of a strain of infectious TSE agent, it must be independent of the structure of the normal prion protein in these different species. Different prion proteins must be able to adopt the same abnormal conformation even though they have very different structures in their normal states.

Strains of TSEs differ in several other respects in addition to incubation periods and lesion profiles. They show large differences in their resistance to inactivation by heat or chemicals. Primary BSE and a mouse-passaged strain derived from it (called 301V) are the TSE agents most resistant to inactivation by heat yet to be discovered. Nevertheless, resistance to inactivation is a property of the 'strain' and not the primary structure of the prion protein that carries it because the resistance properties of a strain remain the same when it is produced in mice differing in the type of prion protein they make (s7 v. p7).

Different strains of TSEs are associated with different biochemical and biophysical properties of the abnormal prion protein. The prion protein has two sites at which it can be glycosylated, i.e. amino acids at which sugar residues can be attached, but not all molecules of the prion protein are fully glycosylated. Thus, an individual molecule may have no added sugars, sugars added at one or other of the sites or at both sites. The relative ratios of the prion protein in these different states can be quantified and they differ predictably between different strains.

When the prion protein adopts the abnormal conformation characteristic of a TSE infection it becomes partially resistant to digestion with enzymes that degrade proteins – called proteinases. This relative resistance of the abnormal form of the prion protein to proteinase digestion is used as a diagnostic feature for infection, but the size of the resistant protein fragment has been found to differ slightly for abnormal prion proteins produced by different strains, indicating that strain-specific

ABOVE:
Fig. 2. Strains of TSE agents differ in the relative lengths of incubation period in different inbred lines of mice. R3 and C57BL are *s7* mice, VM is a *p7* mouse and C57BLxVM is the cross between the two lines. Several sources of BSE from cattle, FSE from domestic cats and vCJD from humans produce very similar patterns, indicating that the same strain of TSE agent is causing the infection. Different sources of scrapie on the other hand produce very different patterns of incubation in the different mouse lines, indicating that they are different from BSE as well as being different from each other.
BASED ON DATA PROVIDED BY MOIRA BRUCE AND COLLEAGUES

conformational differences might change the site which is accessible to the cutting enzyme.

It has been suggested that these two properties, ratios of glycosylated forms and the size of the proteinase-resistant fragment, could be used as a basis for biochemically identifying different strains. If this were possible it would provide several advantages since it would be both quicker and would not involve the use of large numbers of mice to bioassay the infectious agent. In the case of well characterized mouse passaged strains of scrapie it appears that a particular strain/mouse combination produces a characteristic narrow range of ratios of glycosylated forms (Fig. 3). The method has been used successfully to distinguish between the various forms of CJD in humans. Whether this type of biochemical typing can be made precise enough so that each strain of TSE has its own unique 'fingerprint', irrespective of the species or prion protein background, remains to be seen. In the case of natural and experimental sheep scrapie the patterns are numerous and complex and it is likely to take some time before it will be known whether the approach is feasible as a practical diagnostic method.

The observation that strains differ in the degree of glycosylation of the accumulated abnormal prion protein has lead to the hypothesis that the pattern of glycosylation may itself be the biochemical determinant of strain. It is proposed that a certain type of cell in the host might recognize individual patterns of sugars on the prion protein molecule and thus 'permit' the infectious agent to enter and replicate within it. If, in the process of replicating the agent, the cell 'stamped' its own glycosylation signal on the newly formed abnormal prion protein, it would ensure the next generation of 'infectious prions' were of its own kind, thus perpetuating the tropism and properties of the strain. This idea has been tested by infecting a single cell type with two different strains of agent. If the strain properties derive from a cell-imparted glycosylation pattern, the two originally different strains would be expected to emerge with the same strain characteristics and glycosylation patterns. Alternatively, if the determinant of strain is independent of glycosylation pattern of prion protein and the cell type in which it is replicating, the two strains should retain their original characteristics. Experiments using cells infected in culture indicate that the latter is the case and suggest that differences in glycosylation result from strain differences, and are not the cause of them.

Conclusion

Returning finally to the question of the nature of the agent. Work on strain properties of TSE agents has shown that if the prion protein is the sole component of the infectious agent, the conformational determinant of infectiousness and strain must be independent of prion protein primary structure (sequence of amino acids). A single prion protein must be capable of adopting and propagating several different stable abnormal conformations while several different prion proteins must be able to adopt a common abnormal conformation. A full understanding of how this is achieved will have to await a complete molecular description of the various forms of the prion protein.

Much of this article is based on the work of my colleagues at the Institute for Animal Health, whom I thank for providing the material that has been incorporated in the figures.

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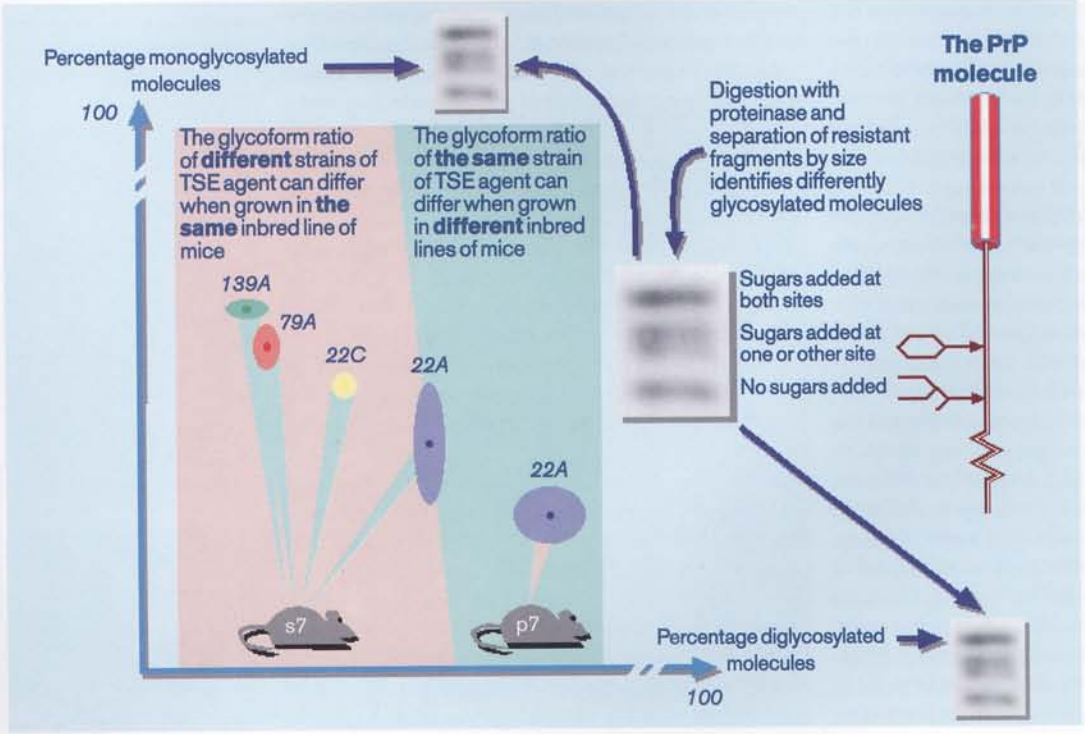
BELOW:
Fig. 3. The prion protein can have sugars added at one or other or both of two sites, the effect being to increase the size of those molecules with added sugars. These size differences can be used to separate and quantify the various forms. The percentage of the total abnormal prion protein with sugars added at two sites (diglycosylated) relative to the percentage of those molecules with sugar added at only a single site (monoglycosylated) can be constant for a particular strain of TSE agent, but differ between strains of agent, replicated in a single inbred line of mouse. The ratio can change when a strain is propagated in an inbred line of mouse with a different prion protein gene.
BASED ON DATA PROVIDED BY ROBERT SOMERVILLE AND JAMES HOPE

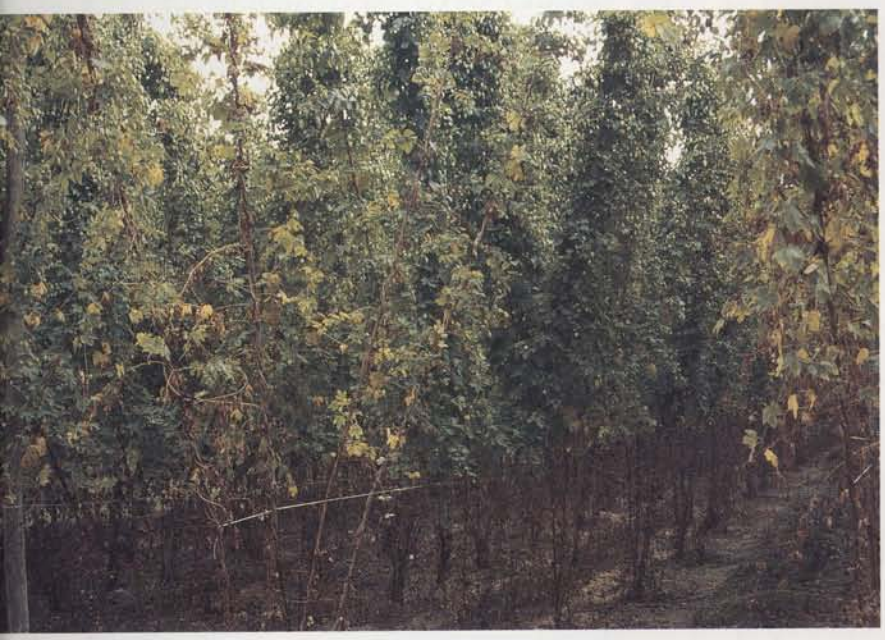


Viroids and other sub-viral pathogens of plants: the smallest living fossils?

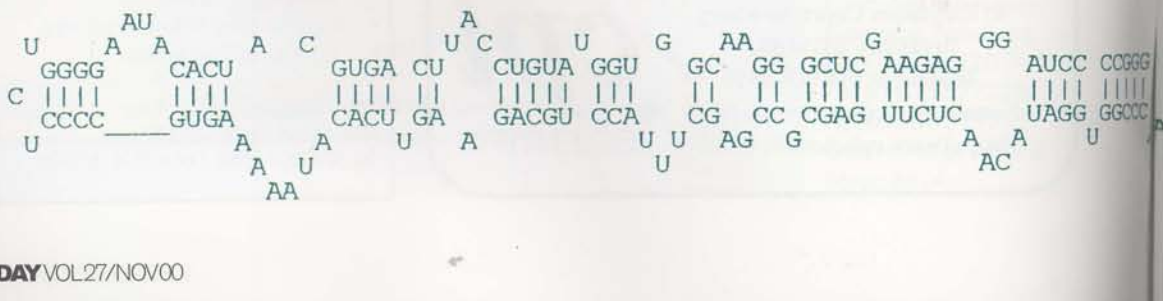
Nicola Spence & Dez Barbara

Viroids and satellite RNAs are the smallest and simplest replicating molecules known. Possibly 'living fossils' they are the cause of a wide variety of infectious diseases in plants.

Contrary to earlier belief, viruses are not the smallest causative agents of infectious diseases in plants. Single-stranded RNAs, termed viroids, exist in many higher plants, causing a variety of virus-like diseases. Viroids can be as small as 246 nucleotides or as large as 400, but their RNA is not organized into codons and they are not translated. This is in contrast to plant viruses where translation of viral genetic information is essential for virus replication. The physical structure of viroids was first shown directly by electron microscopy; under non-denaturing conditions viroids appear as small rods of approx. 37 nm, but under denaturing conditions they can be seen to be covalently closed circular molecules. Due to their high degree of internal base pairing, viroids have a characteristic rod-like secondary structure in which short helical regions are interrupted by internal and bulging loops (Fig. 1). In the majority of cases there are five structural domains in the viroid genome. Many satellite RNAs resemble viroids in size and molecular structure, but are associated with specific helper viruses on which they depend for their replication and are encapsidated with, or instead of, host viral genome components into the virus particle. However, the satellite RNA is not part of the helper virus genome and usually has no sequence similarity to it. Both types of small RNA replicate by a rolling circle mechanism, using either just plant or plant and viral components. Satellite RNAs and just two viroids are able to self-cleave from the concatameric RNA but most viroid RNAs are enzymatically cleaved.



ABOVE:
Fig. 2. Commercial hops (*Humulus lupulus*), variety Omega, with symptoms of HLVd. Omega is the only known hop variety which shows visible symptoms of this viroid.
COURTESY D. BARBARA



BELOW RIGHT:
Fig. 1. The sequence of hop latent viroid (HLVd) arranged in a thermodynamically optimized secondary structure.
AFTER PUCHTA *ET AL.* (1988)

Plant diseases caused by viroids

Crop diseases caused by viroids probably originated by chance transfer from endemically infected wild plants or by use of viroid-infected germplasm during plant breeding. Despite their extreme simplicity, viroids can cause syndromes in plants that are as varied as those caused by plant viruses but others, such as coleus latent viroid (CLVd), are totally symptomless and occur at extremely high rates of infection (up to 100%). Because viroids are not translated, their effects on plants must be a consequence of direct interactions of the viroid with the host and its environment. Although the molecular mechanisms of viroid pathogenesis are still unknown, analysis of molecular chimeras has revealed that the severity of symptoms is the result of complex interactions among three of the five viroid domains. As a group, there is nothing that distinguishes the disease symptoms produced by viroids from those induced by viruses – these include stunting, mottling, leaf distortion and necrosis. Diseases cover a wide range from the slowly developing lethal disease in coconut palms caused by coconut cadang-cadang viroid (CCCVd) to the usually symptomless hop latent viroid (HLVd; Fig. 2), and the latent viroids which are always symptomless. It is possible that many more mild or symptomless viroid infections remain to be discovered.

Viroid epidemiology

The main methods by which viroids are spread are mechanical transmission, vegetative propagation and through pollen and seed. The relative importance of these methods varies with different viroids and hosts. For example, vegetative propagation is dominant for chrysanthemum stunt viroid (CSVd). Mechanical transmission is a significant factor for others such as citrus exocortis viroid (CEVd; Fig. 3) and hop stunt viroid (HSVd; Fig. 4). Seed and pollen transmission are factors in the spread of avocado sunblotch viroid (ASBVd); seed is particularly important for some of the latent viroids. Potato spindle tuber viroid (PSTVd) is transmitted at low frequency in a non-persistent manner by the aphid *Macrosiphum euphorbiae*. However, it is doubtful if aphid transmission is of any significance in the field.

ABOVE:
Fig. 3. CEVd symptoms in a citrus species.
COURTESY D. BARBARA

Viroids move rapidly through a host plant in the manner of competent viruses, almost certainly through the phloem. The relative resistance of viroid RNA to nuclease degradation (which arises from their internally base-paired structure) probably facilitates their long-distance movement. It is also possible that viroid particles are translocated while bound to some host protein. For most viroid diseases the reservoir of inoculum appears to be within the crop itself, raising the question as to where they came from originally. A viroid present in a natural host, possibly causing no disease, might 'escape' into a nearby susceptible commercial crop and spread rapidly within it. Modern agricultural practices, such as widespread monoculture of genetically identical plants, and worldwide distribution of planting material, have probably made possible the sudden appearance and rapid spread of new viroid diseases. For example, a study of HLVd in the UK suggests that the current prevalence of this viroid in hops is a consequence of infection becoming established in the hop propagation system during the late 1970s.

Viroid detection and diagnosis

Serological tests are widely used for the detection and diagnosis of viruses, but these cannot be used for viroids as they are not immunogenic. Besides indicator plants, biophysical techniques such as two-dimensional gel electrophoresis, return gel electrophoresis, hybridization and RT-PCR of nucleic acid extracts are used for more rapid testing. Viroid identification can be difficult. In the search for viroids and viroid-like RNAs in oil palms from Central and South America affected by a fatal yellowing disease, RNAs showing viroid-like gel-electrophoretic properties were detected. However, the presence of known viroids was excluded by hybridization experiments using viroid-specific probes and the use of double-stranded RNA (dsRNA)-specific monoclonal antibodies (which do not react with viroid RNA), showed the oil palm RNAs to be dsRNA species and not circular single-stranded ones. Furthermore, since the same dsRNA pattern was found in extracts from healthy as well as from diseased oil palms, it was concluded that the dsRNAs were not associated with the disease.

Viroid disease control

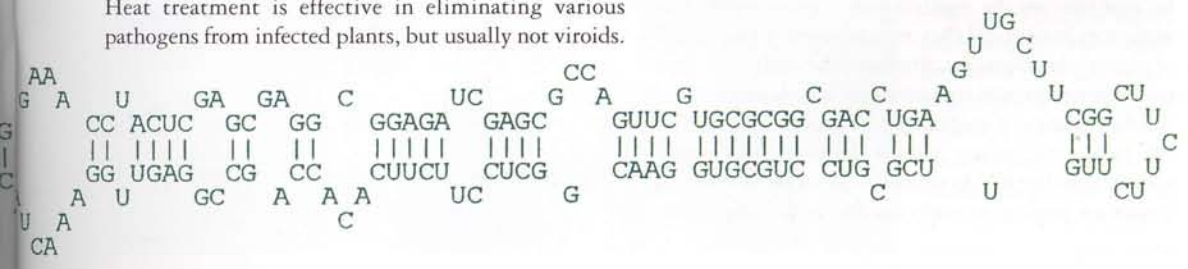
Disinfection of cutting tools is recommended to prevent viroid transmission, especially in nursery production. Heat treatment is effective in eliminating various pathogens from infected plants, but usually not viroids. However, cold treatment can be effective, e.g. storage at 4 °C for 6 months or more, followed by apical shoot-tip-culture grafting, can be used to eliminate CSVd and HLVd. Pre-inoculation with protective mild strains of viroid has proved effective to control PSTVd. It has also been shown that interference occurs not only between strains of a particular viroid but also between different viroids. In experiments in which mild and severe strains of PSTVd were inoculated to a host simultaneously, the severe strain dominated and most plants developed severe disease, even when the mild strain was in 100-fold excess in the inoculum. In other experiments, PSTVd RNA transcripts from a cloned PSTVd DNA were inoculated together with HSVd RNA. PSTVd reduced the level of HSVd RNA in infected plants. Plants inoculated with dual transcripts – two copies of a severe PSTVd strain linked to two of HSVd – developed PSTVd symptoms and only PSTVd progeny RNA could be detected. The molecular basis for this interference is not yet understood.

Recombination between viroids

Nucleotide sequence data shows that it is highly probable that recombination has taken place between different viroids, presumably during replication in mixed infections. For example, tomato apical stunt viroid (TASVd) appears to be a recombinant viroid comprised mostly of the sequence of a CEVd-like viroid but with the T2 domain replaced by the equivalent domain from a PSTVd-like viroid. Australian grapevine viroid (AGVd) appears to have originated by extensive RNA recombination as its sequence can be divided into regions each with high sequence similarity with parts of CEVd, PSTVd, apple scar skin viroid (ASSVd) and grapevine yellow speckle viroid (GYSVd).

Satellite viruses and satellite RNAs

Purified virus preparations isolated from infected plants may contain a variety of small RNAs other than the genomic RNAs. In addition, some isolates of certain plant viruses may also contain satellite viruses. The two



RIGHT:
Fig. 4. HSVd symptoms in *Humulus japonicus*.
COURTESY D. BARBARA

classes of agents can be distinguished according to the source of the coat protein encapsidating the RNA and the sizes of the RNAs. In satellite viruses the RNA encodes its own coat protein whilst small satellite RNAs become packaged in protein shells made from coat protein of the helper virus. Also, the RNAs in satellite viruses have to be large enough to encode the coat protein as opposed to the viroid-like size of the satellite RNAs. Like viroids, satellite RNAs may be responsible for serious disease despite their minimal size. In 1972, for example, a devastating outbreak of a lethal necrotic disease of field-grown tomatoes occurred in the Alsace region of France. It was quickly realized that cucumber mosaic virus (CMV) was involved, but it was not clear why necrosis occurred instead of the usual fern-leaf symptoms. However, a small RNA component was found to be present in some isolates of CMV in addition to the three genomic RNAs and the subgenomic coat protein mRNA, and this fifth RNA was not part of the viral genome. The additional small RNA (called CARNA5) present in cultures of CMV strain S was found to cause lethal necrotic disease in tomatoes when added to the CMV genomic RNAs and its presence was thought to have been responsible for the Alsace lethal necrotic disease. Similar recent outbreaks in tomatoes in southern Italy have been shown to be due to a necrogenic isolate of CARNA5. Conversely, some isolated types of CARNA5 attenuate symptoms in tomato and have been used in China for pre-inoculation as protective mild strains for the control of CMV.

Living fossils?

Viroids and satellite RNAs are of great interest because they are the smallest and simplest replicating molecules known and they may represent living fossils of pre-cellular evolution. Phylogenetic analysis of their nucleotide sequences indicates that viroids and satellite RNAs represent a monophyletic group, with all but the two self-cleaving viroids forming one cluster and the satellite RNAs another. The two self-cleaving viroids are phylogenetically distant from either cluster and may represent ancestral forms. Site-directed mutagenesis experiments indicate that viroids can evolve extremely rapidly in response to selective pressures, with fitter components of the quasi-species often becoming dominant within days or weeks. This extreme plasticity of their nucleotide sequences establishes viroids as the most rapidly evolving biological system known.

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Viruses rule the waves – the smallest and most abundant members of marine ecosystems

Gunnar Bratbak & Mikal Heldal

In his authoritative book *Marine Microbiology*, ZoBell (1946) stated that '*...since bacteriophage is generally found associated with large numbers of rapidly multiplying bacteria, it is very doubtful if the sparse bacterial population characteristic of the open ocean is conducive to the development or activity of bacteriophage*'. ZoBell's view on bacteria in sea water was based on the numbers obtained with plate counts (around 100–1000 c.f.u. ml^{-1}) and his reasoning concerning viruses was logical, but in the late 1970s the total abundance of bacteria in sea water was found to be 1000 times higher, i.e. about 10^6 cells ml^{-1}. The organisms are quite active, growing at a rate of about one division per day. The basis for ZoBell's view on viruses was thus proved wrong and we had to revise the general textbook wisdom that viruses are unimportant in natural waters. We now know that viruses outnumber bacteria by a factor of 10, at 10^7 ml^{-1}.

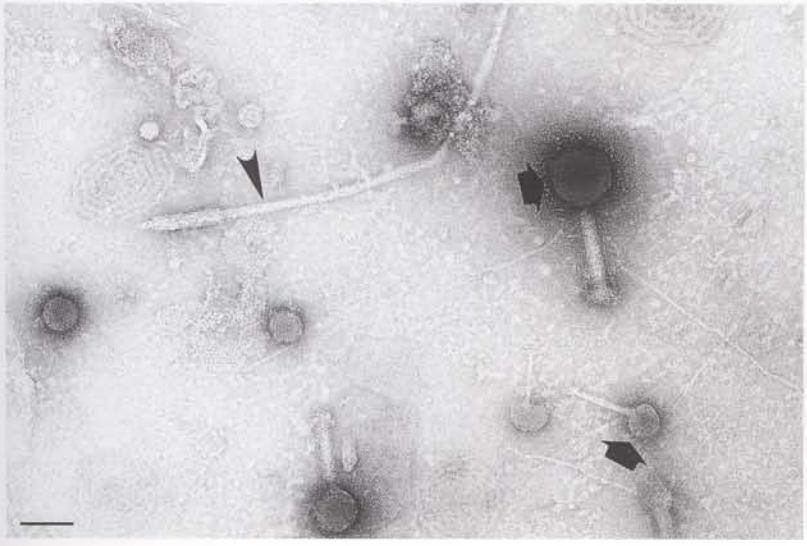
By the end of the 1980s, when much data on bacterial biomass and production and on protozoan grazing had accumulated, it appeared that bacterial production and removal were not always in balance. This could have been due to methodological inaccuracies, but it was also possible that other removal mechanisms such as cell lysis and viruses were involved. At this time centrifugation was used to harvest bacteria directly onto electron microscope grids for transmission electron microscopy (TEM) analysis and we reasoned that if viruses were present in significant numbers we could harvest them too just by increasing the speed of the centrifuge. With some trial and error, and a wrecked ultracentrifuge rotor, we came up with convincing electron microscope images of native virus communities (Fig. 1) and quantitative results on the abundance of viruses.

We did not *discover* viruses in the sea and nor was the 'novel' method we invented for counting viruses actually new. The presence of viruses, i.e. bacteriophages, in sea water has been known for a long time (at least since the 1950s) and high abundance was also reported in 1979. Unfortunately for these early researchers, their findings were not recognized and appreciated when published. Our publication appeared at a time, 10 years ago, when viruses fitted in as the last (?) piece in the puzzle. It provoked an immediate response. Publication in *Nature* gave the story publicity and the high number (billions in a teaspoon of water!) of viruses (dangerous and scary!) was considered newsworthy by the press. The notion that virus activity essentially is a question of 'sex and crime' might also have been a factor in this interest. At the time, we thought that the method we had developed for counting viruses in the transmission electron microscope was simple, elegant and new. Later we became aware of no less than five papers published between 1949 and 1986 describing procedures for counting viruses by the same approach.

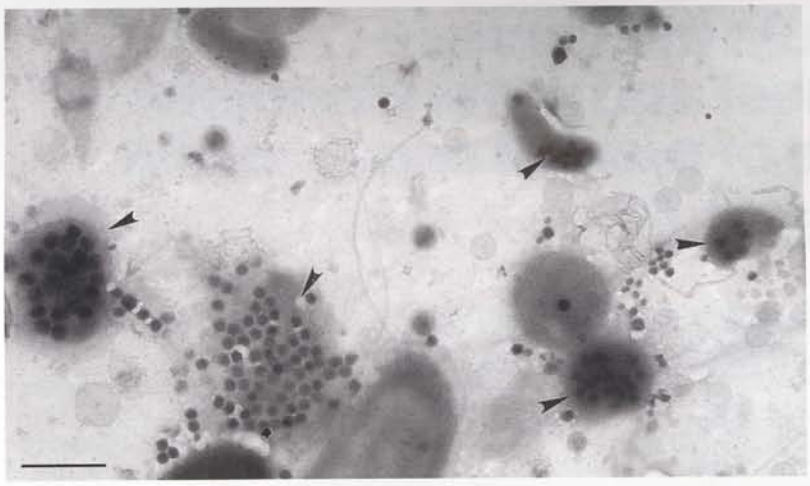
Counting viruses, was of course not enough, and in the years after the 'discovery' we and many other research groups have worked to understand their ecological significance. First of all, what we include in viral total counts are not actually viruses but Virus-Like Particles (VLPs), i.e. electron-dense particles with a hexagonal to round shape and a diameter of 30–200 nm (Figs 2 and 3). This distinction is important, as there is no way to ascertain that each and every particle we observe in the microscope really is a virus. They may in fact not be viruses at all, they may be DNA blebs, ink particles from squid, or some other particle of biological origin that has not been described yet. On the other hand, we may miss a lot of viruses that are not hexagonal or round. In our TEM preparations we sometimes observe rod-shaped and filamentous particles that may well be viruses (Figs 3 and 4), but how can we tell?

One of the first questions we asked ourselves – what do the viruses do in the ocean? – was in fact quite naïve. They obviously do the same as they do in cultures, i.e. lyse cells or make them lysogenic. Like other DNA-based entities, they struggle to survive and transfer their DNA to the next generation as best they can. The basic questions in microbial ecology – who, how many, and how fast? – may sound simple but they have nevertheless proved difficult to answer and indeed this has also been the case in virus ecology. The effects of viruses on population dynamics, community

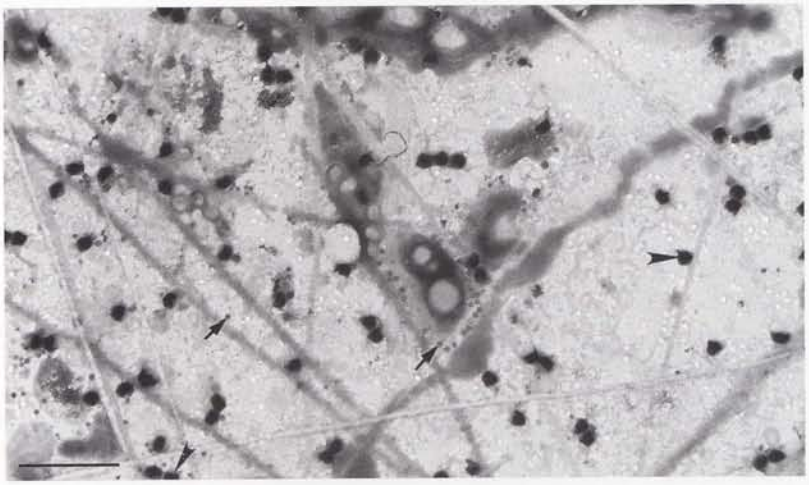
Some 10 years ago a series of letters was published in *Nature* showing that bacteriophages were both abundant and active in natural waters. These reports boosted research on the ecological significance of viruses and the field is now well established as an integral part of aquatic microbial ecology.



LEFT:
Fig. 1. Tailed bacteriophages (arrows) from coastal sea water. The wide range in head diameter indicates a diverse community. Other structures (arrowhead) may perhaps relate to filamentous phages. The particles were harvested directly onto nickel grids (400 mesh) by centrifugation (200,000 *g*, 30 min.), and negative-stained with uranyl acetate (2 % in water). Bar, 0·1 μm.
COURTESY M. HELDAL AND G. BRATBAK



RIGHT (UPPER):
Fig. 2. Five bacteria (arrowheads) at different stages of viral maturation and lysis. These virus particles may be more difficult to recognize as viruses compared with the tailed viruses in Fig. 1. Note the variation in size of virus particles and in numbers of particles per cell. Preparation was as described for Fig. 1, but the sample was positive-stained with uranyl acetate. Bar, 0·5 μm.
COURTESY M. HELDAL AND G. BRATBAK



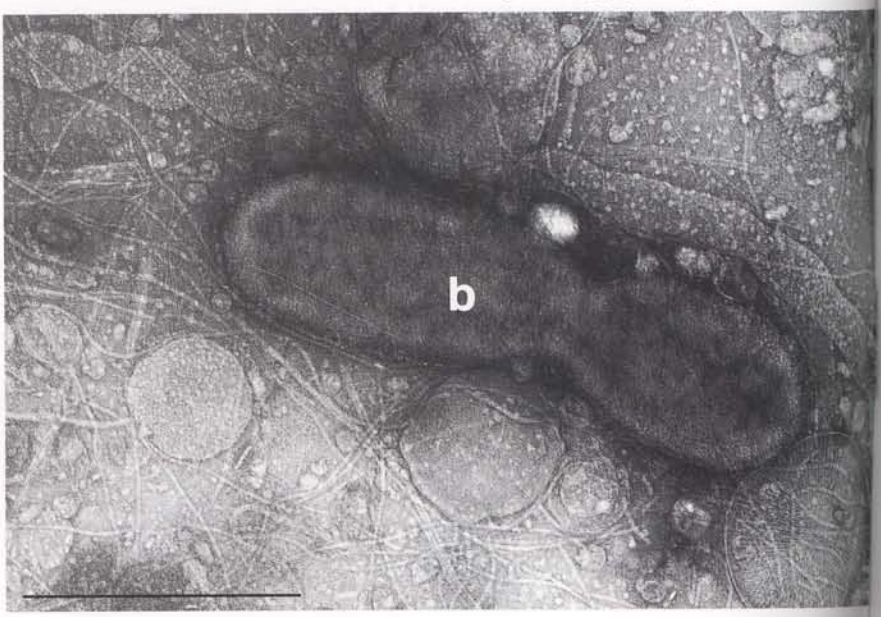
RIGHT (LOWER):
Fig. 3. Algal virus (arrowheads) and phages (arrows) in a coastal sea water sample. The thread-like structures are presumably from *Phaeocystis* sp. (small arrows), but a lot of particles of unknown identity and origin are also seen. Preparation as for Fig. 2. Bar, 1 μm.
COURTESY M. HELDAL AND G. BRATBAK

structure, diversity, nutrient flow, biogeochemical cycles and climate are far from obvious and the research requires the combined effort of many disciplines.

Based on the simple observation that bacteria are the most abundant possible hosts in the sea, it is assumed that most of the viruses are bacteriophages. The problem with the bacteria in this context is their general lack of morphological traits, which makes it difficult to distinguish one possible host population from another, and that less than 1 % of them are easily isolated and cultured in the lab. We thus know very little about individual populations in mixed natural marine communities. What we know is mainly based on the use of molecular methods and ribosomal RNA analysis. We have just begun to unravel the diversity of bacterial communities and the distribution and dynamics of individual bacterial populations. It has therefore been difficult to demonstrate how viruses affect native bacterial populations in natural environments. Some phytoplankton species are, in contrast, easily recognized by trained taxonomists and several studies have now demonstrated that phytoplankton blooms may be terminated, and the succession driven, by viral infections.

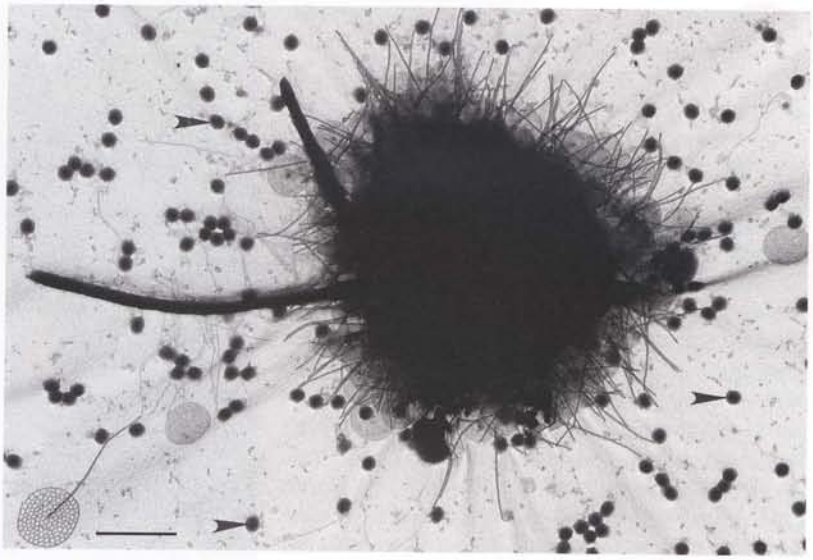
What controls viral activity in natural waters is still a matter of debate. Some experiments indicate that the majority of viruses are produced as a consequence of a lytic infection. The rate of virus production will thus be controlled by the abundance of hosts multiplied by the abundance of virus. The alternative is that the host is lysogenic, or somehow carries the virus or the virus genome in an inactivated stage. This hypothesis would explain how the virus 'survives' when the host population is gone, where it comes from when the host population returns and how the host population protects itself from being exterminated by the virus. Our guess is that both strategies and all possible intermediates are used by different populations in the bacterial community. If we look for lytic infection we will find it and if we look for lysogeny we will find that too. What is important is the question of timescale and the nexus of cause and effect, and thus the experimental design.

Population growth in natural environments is controlled and limited by food supply and by predation.



BELOW:
Fig. 4. TEM image demonstrating the diversity of particles that may be found in coastal seawater. The bacterium (b) is easily recognized, but what are all the round and filamentous particles in the background? Preparation as for Fig 1. Bar, 1 μm.
COURTESY M. HELDAL AND G. BRATBAK

These factors may be selective but they are not specific because they to some degree affect all organisms in the community, depending on the properties of the organisms involved. Those with high nutrient affinity and high growth efficiency may, for example, perform better under nutrient limitation than others. Very small, very long and very large cells may experience less grazing pressure than average-sized cells. The population control exerted by viruses is very different. Viruses are population-specific and by attacking one single population (i.e. the host) they make life easier for competing populations, which of course also may have their viruses. Thus, food limitation and predation selects for those that are able to cope with given environmental conditions, i.e. those that are most fit. The rate of virus infection depends on host-cell abundance and virus replication requires actively growing cells. Successful populations with active and abundant cells are thus more vulnerable to viral infection than those which are small or inactive. Food limitation and predation selects the winner – and the viruses kill the winner (Fig. 5).



LEFT:
Fig. 5. Blooms of the marine phytoplankton *Emiliania huxleyi* have often been observed to be terminated by viral infection and the concentration of *E. huxleyi* viruses (arrowheads) in the sea may become very high. Viruses are host-specific and the flagellate shown here does not have to worry about the surrounding *E. huxleyi* viruses. On the contrary, it may rejoice at less competition from *E. huxleyi*: the *Emiliania* bloomed, but a virus killed the winner! Preparation as for Fig. 2. Bar, 1 µm.
COURTESY M. HELDAL AND G. BRATBAK

The most frequently cited figures suggest that 20–30 % of bacterial production and 2–10 % of phytoplankton primary production is channelled through 'the viral shunt' in the microbial food web. Cell lysis implies that organic material is lost from the grazing food chain, where the organisms depend on particulate food, and becomes available to bacteria thriving on dissolved organic material and nutrients. The net effect of this is increased nutrient recycling and respiratory loss of organic carbon in the lower parts of the food chain. As a quantitative significant process, viral activity does have direct implications for the carbon budget of the ocean, and hence for the global climate. Moreover, virus infection in algae has been found to cause increased production of dimethylsulfoniopropionate (DMSP) and dimethylsulphide (DMS). When DMS escapes to the atmosphere it causes increased cloud formation, which in turn affects global radiation and thus global warming. If this was not enough, DMS also causes acid rain.

We all have viruses and know what they do to us; we try to avoid them and have doctors to cure us. We know that bacteria and algae have their pests. There are also a few other examples of the ecological significance of viruses in the sea that have attracted public attention. In 1988–89 a morbillivirus epizootic killed >18,000 seals around the North Sea and in 1990 hundreds of dolphins died of the same virus in the Mediterranean. Diseases caused by viruses like ISAV and IPNV caused significant economic loss in salmon farms before efficient vaccines became available. The obvious question then is what about all the other creatures in the sea – herring, mackerel, cod, anchovies, whales, etc.? Can food limitation, predation, fishing and hunting really explain all the large fluctuations in population density that have been observed for these species? They all have viruses and it is tempting to suggest that virus infection may play some role in controlling their lives.

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In search of a second evolutionary experiment

Don Cowan & Monica Grady

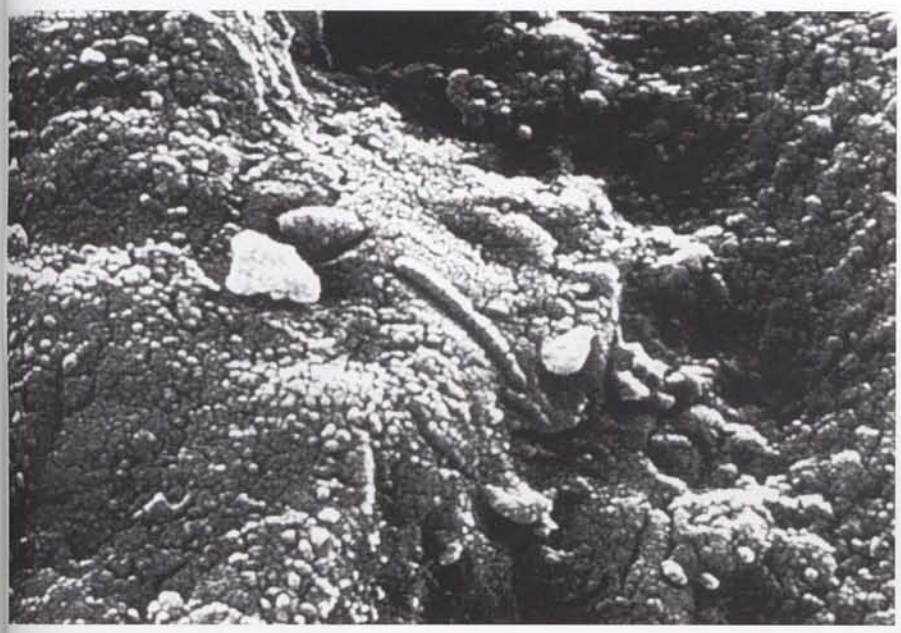
The UK Astrobiology Forum defines astrobiology as the 'formation, evolution and adaptation of life in a planetary and stellar context'. Don Cowan and Monica Grady describe current thinking on extra-terrestrial life.

Exobiology (life on other planets, now generally termed 'astrobiology') is a subject of intense interest and speculation, particularly in the public domain, with some of the most fundamental questions of science – how did life evolve, what other forms might life take and 'are we alone'? Such questions have long stimulated scientific minds, not to mention those of science-fiction writers, the media and the film industry. But despite our preoccupation with the subject, we still have little understanding of the answers. For example, the molecular and energetic processes linking prebiotic chemicals and the first simple replicating cell are subject to a number of hypotheses, but little experimental verification. Such studies are limited by two fundamental problems: we have access to only a single evolutionary experiment, i.e. our own, and the starting conditions were fixed $4{\cdot}56 \times 10^9$ years ago, leaving little if any physical evidence of the process. An ability to access and investigate a second evolutionary experiment, as we might find on a neighbouring planet, would be a major breakthrough in our understanding of the development of molecular processes and structures in our own evolution. Are there, for example, thermodynamic imperatives in the molecular basis of living systems, and will alternative evolutionary pathways have employed similar structures and processes in evolving key systems such as energy capture, compartmentalization and information storage?

Mars, Viking and meteorites

Following the unsuccessful attempts in the late 1970s by the Viking missions to identify life on the Martian surface, interest in astrobiology waned, at least amongst the scientific community. However, analyses of Martian meteorites have added impetus to the resurgence of interest in astrobiology. A collection of 17 now exists, many of which have been collected from glacial surfaces in Antarctica (Fig. 1). The Martian origin of these meteorites has been deduced partly from their young crystallization ages (implying planetary, rather than asteroidal origin) and also from the presence of gas inside that has the same elemental and isotopic composition as Mars' atmosphere (as measured by the Viking landers in 1976). The observation that at least one of the Martian meteorites, EETA79001, contained indigenous, i.e. Martian, organic material associated with carbonate minerals sparked discussion on the possibility of the meteorites containing evidence for extraterrestrial life.

The debate was reactivated in 1996 with the announcement that possible evidence of past life had been discovered in the Martian meteorite ALH 84001. A team of scientists, led by David McKay of NASA's Johnson Space Centre in Houston, described nanometre-sized features within carbonate patches in ALH 84001 and claimed to have found evidence for a primitive 'fossilized Martian biota'. Identification of the features remains controversial, since much of the evidence is circumstantial and relies on the coincidence between a number of otherwise unrelated characteristics of the meteorite (the occurrence of carbonates, organic compounds and magnetite associated with the carbonates). The most compelling 'observation', though not the most compelling scientific evidence, was NASA's electron microscopic image of a putative microfossil (Fig. 2). The features, however, are smaller by about two orders of magnitude than most common micro-organisms known on Earth. Subsequent investigation has revealed that nanobacteria might be more prevalent than previously anticipated and nanometre-sized organisms have been isolated from terrestrial sedimentary rocks. Even so, many doubts still arise as to the validity of interpreting morphological data as fossil nanobacteria, especially given that the environment and mode of formation of the host of the features in ALH 84001, the carbonates, are not fully understood. Current thinking is that the carbonates were produced at the surface of Mars



FAR LEFT:
Fig. 1. Antarctic glaciers – collection points for meteorites.
COURTESY M. GRADY

LEFT:
Fig. 2. Putative 'microfossils' in martian meteorites as revealed by electron microscopy.
COURTESY M. GRADY

in a region of restricted water flow, such as an evaporating pool of brine. This hypothesis satisfactorily accounts for the chemical and isotopic characteristics of the carbonates and is also a mechanism compatible with an environment in which micro-organisms might survive.

The debate over interpretation of the features continues, with definitive clarification probably dependent on recovery of new Martian meteorite samples, or the acquisition of appropriate material directly from a planetary source. Fortunately, there is an active programme of meteorite collection in Antarctic and other deserts, although a very small proportion of meteorites collected are of Martian origin. NASA 'sample return' missions to Mars and elsewhere are scheduled for 2008 and beyond. So, with this material available, what technologies could be best applied in the search for evidence of life? The answers differ, depending on whether the target is evidence of fossil or extant life. Numerous chemical signatures are accepted as fossil biomarkers, including polyaromatic hydrocarbons, hopanes, terpenoids, etc. Here we focus on methods applicable to the detection of extant life or life processes, such as might be applied to new meteorite samples or incorporated in a planetary lander package. One major caveat must be borne in mind when considering measurement of organic materials on planetary surfaces: to date, no organics have yet been detected, let alone have isotopic, chiral or molecular systematics been determined.

Panspermia?

Several groups of meteorites contain abundant carbon, up to several weight percent in the types classified as carbonaceous chondrites, in which it mainly occurs as organic material. There is currently no general belief that this organic matter is biogenic in origin – it is understood to be built up from simple carbon-bearing molecules, first in the interstellar medium, subsequently through processing on asteroidal parents. Comets are also rich in organic molecules and bombardment by comets and asteroids is a favoured possible mechanism by which the Earth was seeded with the starting materials necessary for life to arise. The addition to the Earth of a late-stage veneer of organic material by bombardment is very different from the transfer of biological material within the Solar System via cometary or asteroidal transport. Thus far, there is no evidence for viable biological material in meteorites, but the possibilities of its retention in meteoritic samples cannot be completely discounted. The hypothesis, promoted under the popular title of 'panspermia', has attracted considerable public and scientific notoriety, not least because of some of the more dramatic predictions of its consequences. If the three phases of 'panspermic' transfer (ejection, interplanetary transport and collision) are considered, the survival of living micro-organisms is at least hypothetically possible. Ejecta from major asteroid impacts are exposed to temperatures of a few thousand degrees for periods of only seconds before being exposed to the near-absolute-zero temperatures of space. The low thermal conductivity of most rock types would effectively protect all but the outer layer of the ejected body from significant heating. Survival of biological material during interplanetary (or interstellar) transport is an issue of molecular (in)stability, particularly with respect to the deleterious effects of chemical degradation and intrinsic and cosmic radiation damage. Biological material is also exposed to extremely low temperatures and total desiccation, over very long periods. While molecular stability over such extended periods might seem unlikely, recent reports of viable bacterial spores being recovered from ancient salt deposits and 1·25 million-year-old Antarctic ice cores suggest a lower (but not an upper) limit of microbial survival.

Finally, before a meteorite arrives at the Earth's surface, it must undergo the energetic processes of entry through the atmospheric layers and impact. Entry heating, caused by friction, melts the outermost layer of the meteoroid; the molten rock is carried away, back along the entry trajectory. Continued passage through the atmosphere melts successive outer surfaces, but the rapid removal of melt from the meteoroid prevents the inner regions from becoming hot. Eventually, friction decelerates the meteoroid such that its speed is no longer sufficient to cause melting and the surface cools. Thus meteorites are cool when they land and any organic material in the interior will be intact and unaltered by entry heating. Although there is no evidence of biogenic material in meteorites, recognition of meteorites from the Moon and Mars has re-opened the possibility of interplanetary cross-contamination by material ejected from planetary surfaces by impact.

RIGHT:
Fig. 3. The 'Dry Valleys' of Antarctica.
COURTESY D. COWAN

What is 'life'?

Any analytical method designed to identify 'life', whether applied to meteorites or Martian 'soil' samples, must take account of the possible differences in the molecular answers to fundamental questions in an alternative evolution. For example, the selection of ATP as a biomarker, appropriate on Earth because of the central function of this molecule as an energy carrier, ignores the possibility that an alternative nucleotide, a different phosphate derivative, or a different molecular structure altogether may act as a core energy transducer in an alternative evolution. Exactly how 'life' should be defined has been a matter of scientific and philosophical debate over a period of centuries. Definitions such as 'an energy-consuming, self-replicating system' suffer from the limitation that they can be successfully applied to obviously non-biological entities, such as fire!

One of the apparently fundamental properties of biological systems on Earth is symmetry. At least at the level of higher organisms, symmetry in at least one plane seems to be an evolutionary theme. While less obvious at the microbial level, structural symmetry is still evident, as seen in bacterial coccoid, bacilloid and spirilloid cellular morphologies. The impact of the ALH 84001 'microfossil' on the public and scientific consciousness may have been due largely to our instinctive association of life with symmetry. However, any assay system designed to detect symmetry in the context of 'life' must be capable of excluding those non-biological forms, such as crystals, which exhibit a symmetrical form.

Conversely, molecular asymmetry has been proposed as a useful biomarker. The selective use of chiral building blocks (i.e. D-sugars and L-amino acids) in biological systems potentially provides a mechanism for either remote or *in situ* identification of a putative biology. As always, such analyses incorporate an assumption of universality, although there are good theoretical grounds for suggesting that biological systems will tend to employ one, but not both, chiral forms of the monomeric precursors of important polymers. For example, a catalytic system capable of using both chiral forms of a molecule must either possess low specificity or must be duplicated for each form, both alternatives being of low global thermodynamic efficiency.

Biomarkers

The selection of any molecular biomarker incorporates the assumptions discussed above. However, this apart, a huge range of biomarker options exist. Table 1 shows some of the 'appropriate' targets and assay systems. Each has strengths and weaknesses, depending on the site of assay. For example, for remote planetary investigations (i.e. unmanned landers) miniaturization, sample handling and power requirements are major constraints for the development of spectroscopic technologies. Similar constraints apply to assays requiring liquid handling (such as enzyme or polynucleotide assays). However, leading-edge developments in many technological areas (such as polymer imprint detection systems) may dramatically change the feasibility of such analyses. Many of these limitations are automatically removed in the event of 'sample return' and ultra-high stringency handling facilities designed to avoid bi-directional contamination are in the planning stages both in the US and in Europe.

Table 1. Biomarkers, targets and assay systems

Biomarker	Target	Assay system	Advantages	Limitations
Metabolic activity	CO_2 release from glucose	Radiolabelling	Facile experiment, very high sensitivity	Non-biological catalysis Heterotrophic metabolism
	CO_2 fixation	Radiolabelling	Suggests autotrophic metabolism	Non-biological processes
Catalytic activity	Enzyme function	Various	Multi-enzyme array assays are feasible	High specificity of many enzymes
Biochemical intermediate	ATP	Bioluminescence	Universality?	Liquid handling required
Biological polymers	Nucleic acids	Spectroscopic	High sensitivity	High background
		Catalytic (e.g. PCR)	A positive result would be unequivocal	Liquid handling, extremely specific, very subject to contamination
	Lipid	Spectroscopic	Diagnostic signals for some lipid derivatives (e.g. using Raman spectroscopy)	Easily applicable only to aromatic molecules
	Protein	Spectroscopic	Diagnostic signals at many wavelengths	

Isotope techniques

Possibly one of the most diagnostic of techniques for the identification of biological processes is the detection of isotopic fractionation between components within a system. Abiotic systems, where fractionations are based on well understood chemical and physical reactions, tend towards equilibrium. In contrast, biological systems tend to be out of equilibrium. The carbon cycle on Earth encompasses components within the atmosphere (CO_2, CO), biosphere (organic compounds), hydrosphere (carbonate and bicarbonate anions) and lithosphere (carbonate and calcium-silicate rocks). Measurement of the isotopic composition of carbon from different components has enabled modelling of the extent of biomass turnover through sediment recycling during tectonic processes. Measurement of the end-member compositions of analogous materials on Mars is intended to enable the construction of an equivalent carbon cycle; detection of biological signatures within such a framework is then entirely possible.

Astrobiology today

In anticipation of the arrival of samples from the Martian or Europan sub-surface, what are the world's astrobiologists doing at present? Contrary to the opinion that '*astrobiology is the one subject where scientists have nothing to work on*', there are numerous avenues for research and technological development. These include (to name but a few) the modelling of Earth's early molecular evolutionary pathway, the development of new spectroscopic technologies for identification of biological molecules and the study of organisms inhabiting the most extreme biotopes on this planet as a means of understanding the range of environments which might be targeted on other planets. With respect to the latter, the world's deep-sea hydrothermal vents have yielded hyperthermophilic chemoautotrophs which may best reflect the earliest forms of life on this planet. Similarly, the deep subterranean biosphere and the cold deserts and ice-covered lakes of Antarctica (Fig. 3) are the best available analogues of possible microbiological habitats on Mars and Europa. Such sites are vital in the development and testing of chemistries and technologies which will eventually be applied to the discovery of life on other planets.

There is a resurgence of interest in the many facets of astrobiology in the UK. The recently established UK Astrobiology Network (http://ast.star.rl.ac.uk/astrobiology/panel/) has a remit to promote collaborative astrobiology research in the UK and has been assured by the UK Research Councils that research grant applications in this field will receive a fair hearing. There are now active centres of astrobiological research in the Universities of Bradford, Portsmouth, Kent, the Open University, University College London, the British Antarctic Survey and the Natural History Museum.

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Mobile genes

Nicholas West & Christoph Tang

Mobile genes and transposons are found widely in nature. This article looks at prokaryotic transposons and their potential exploitation by microbiologists.

Around 70 years ago the first observations were made regarding the existence of genes that gave rise to an increase in spontaneous mutation frequencies at other loci. These genes were originally described as 'mutation genes'. In the late 1940s, Barbara McClintock, through her now famous genetic experiments with corn, coined the phrase 'controlling elements' to describe segments of DNA that could not only alter gene activity but were also mobile in the genome. These 'controlling elements' are now known as transposable elements due to their ability to 'transpose' from one site to another. It is now recognized that transposable elements are prevalent throughout nature, being common in bacteria, plants and animals, including mammals. Transposons are particularly interesting forms of transposable elements due to their widespread distribution in nature and the extensive applications for which they have been adapted within the laboratory.

There are two general types of prokaryotic transposon, composite and non-composite. Briefly, composite transposons, such as Tn*10*, consist of a central element, which includes several genes that may confer resistance to antibiotics. This DNA fragment is mobilized through the activity of a transposase encoded within the insertion sequences flanking the element. Insertion sequences have short inverted repeats at either end that act as the recognition site for the transposase, leading to the excision of the complete unit. Non-composite transposons, such as Tn*3*, do not have insertion sequences at their extremities but simply have the inverted repeats necessary for recognition by the transposase. All the activities required for transposition, including the transposase and resolvase, are encoded by sequences in the central region.

This article covers the role of transposons in adaptive evolution and the practical applications of transposons, including some recent developments which illustrate the flexibility and versatility of this type of transposable element.

Transposons in nature

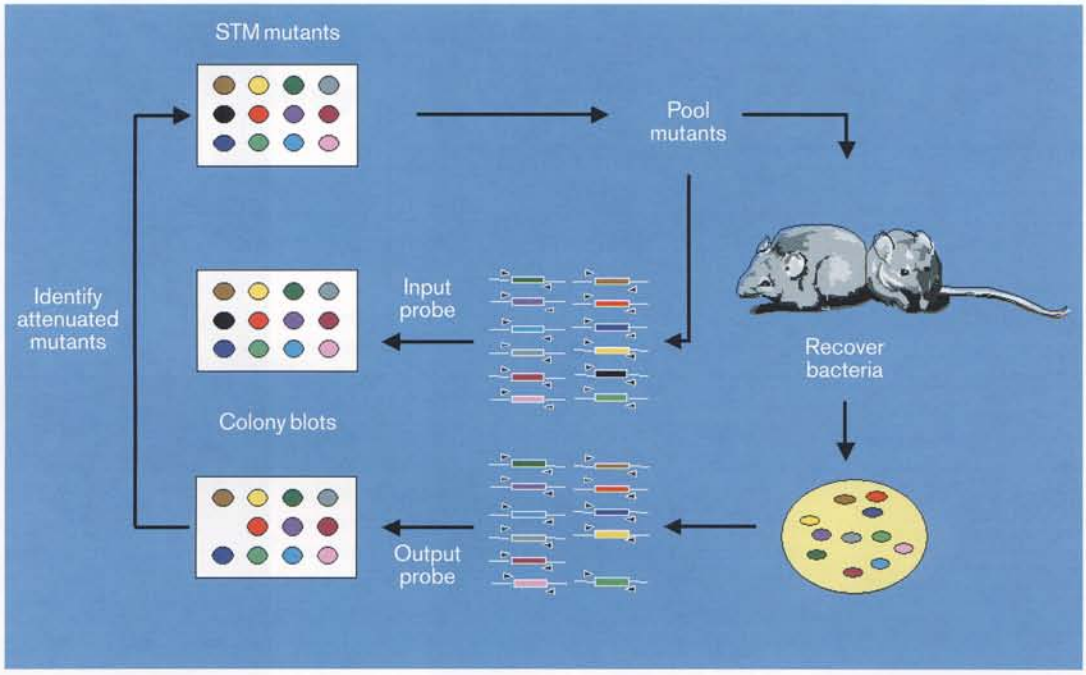
Whole bacterial genome sequencing projects have emphasized the widespread distribution of transposons in microbes. Even archaea and thermophilic bacteria, such as *Thermotoga maritima*, contain multiple copies of sequences related to transposable elements. In the human pathogen *Neisseria meningitidis* over 50 potential transposable elements belonging to two major families, IS*1016* and IS*30*, have been identified in the genome sequence. Furthermore, approximately 40% of the human genome is composed of retrotransposons, elements that utilize an RNA intermediate during mobilization. However, the contribution of transposons to adaptive evolution remains uncertain.

For intergenomic events, there are specific examples of the role of transposons in the spread of beneficial traits within bacterial populations. The best known is the emergence of antimicrobial resistance. The advent of multiresistant bacteria that are oblivious to most available antimicrobial interventions has become a major public health problem. Many of the genes responsible for resistance are carried on transposable elements, which have facilitated the transfer of resistance from commensals to pathogens. For example, vancomycin-resistant *Enterococcus faecium*, which is now prevalent in hospitals throughout developed countries, acquired resistance to this antibiotic on Tn*1546*. Transposons are in fact the predominant source of antibiotic resistance, being transferred across bacterial species and even genera.

Other examples of horizontal transfer between different species or genera further demonstrate how this type of gene 'sharing' can develop bacterial populations. For instance, the acquisition of certain genes, such as resistance to environmental metal ions, would subsequently equip the organism to exploit a new and previously unattainable environmental niche. Furthermore, due to the introduction of organic hydrocarbons into the environment, bacteria have developed mechanisms for the catabolism of many of these pollutants. The genes responsible for catabolism have been largely found on transposons. Elements have been identified with the capabilities to degrade chlorobenzoate, chlorobenzene, toluene, benzene, nylon oligomers and naphthalene, thus illustrating how naturally developed transposons may play a role in the bioremediation of environments that have become polluted.

Transposons have not only been responsible for the spread of characteristics directly related to survival, but they have also been implicated in the evolution of disease-causing bacteria. A feature of enteric pathogens, including *Escherichia coli* and *Salmonella* spp., is the presence of large genetic elements (often in excess of 10 kb) that are vital for their ability to inflict damage on their hosts. These so-called 'pathogenicity islands' are often bounded by insertion sequences, indicating that they were originally acquired by a transposition event. The implication is that transposons have caused quantum leaps in the evolution of virulence among bacterial species.

Furthermore within a population of cells derived from a single bacterium, transposition events mediate genotypic variation through intragenomic recombination, resulting in the appearance of new phenotypic traits on which selection can operate. For instance, it has been shown that expression of capsular polysaccharide, a structure required during pathogenesis by *N. meningitidis*, can be switched off by the insertion of an IS*1016* element upstream of genes encoding biosynthetic steps. A further example of the importance of intragenomic events is phase variation of fimbrial



ABOVE:
Fig. 1. In STM, an insertional mutagen (transposon) is modified by incorporation of DNA signature tags. A collection of different insertional mutants of a bacterial pathogen, each carrying a different tag, is assembled in a microtitre dish. The mutants are pooled and used as the inoculum for an appropriate animal model of infection. Following infection, bacteria are recovered from the host and the unique DNA tags are amplified alongside those present in the initial inoculum using primers that anneal to invariant sequences flanking the tags. The product of the PCR is then utilized to probe nylon membranes carrying DNA from the mutants in the inoculum. An avirulent mutant is identified by the failure to yield a signal on the membrane hybridized with the tags recovered from the animal.

expression in *Salmonella typhimurium*. The bacterium expresses one of two flagellar types, type 1 or type 2. The genetic basis of switching between these types involves the reversible inversion of the Hin element (H antigen inversion) upstream of genes encoding type 2 flagellae and a repressor of type 1 flagellar biogenesis. In one orientation, the promoter element within Hin activates transcription of type 2 biosynthetic genes. In the opposite orientation, the type 2 genes and the repressor are not expressed, allowing the synthesis of type 1 genes.

Although there are other specific examples of transposition events causing phenotypic variation, the overall contribution of transposon and insertion sequences to microbial evolution is uncertain. There are two opposing views. The 'selection hypothesis' holds that genetic flexibility and variability confer long-term fitness benefits to microbes, and this is the reason for the prevalence of transposons. There is some direct, albeit limited, experimental evidence supporting this theory from studies of the growth of *E. coli* in chemostat cultures. Strains containing the transposon Tn*10* have a competitive advantage over isogenic strains without Tn*10*; this fitness gain is lost when Tn*10* is deleted from the bacterium. Some argue that evolution is blind to the future and that long-term views on benefits are far more important in the minds of evolutionary biologists than in nature itself. They suggest that transposons exist purely as selfish genetic elements rather than as symbionts in the bacterial cell. The truth probably lies somewhere between these divergent views; many transposons may well act solely as passengers in bacteria while others enhance the survival of the cells in which they exist.

Transposons in the laboratory

Researchers have long used the natural properties of transposons and adapted them as incredibly versatile tools for the genetic analysis of bacteria. Principally, transposons have been used as insertional mutagens, which may include the construction of libraries of strains, each carrying a single transposon at a different location. The libraries can then be analysed for mutants with the desired phenotype and the affected gene(s) can be easily identified for further study by the presence of the transposon.

Natural transposons have several limitations when used in the laboratory. First, they are usually large elements that are difficult to manipulate, and second, they tend to be unstable and move to other locations. To prevent these problems, mini-transposons have been developed. These lack the resolvase gene, which is required for the recombinational events of transposition, improving their stability. This also allows a significant reduction in size, making them much more user-friendly. Transposons have now been developed for generating both reporter fusions, to monitor levels of expression of genes, and to identify gene products which are translocated to the cell surface. Transposons have also been used to generate large-scale genetic maps by introducing recognition sites for infrequently cutting restriction enzymes. However, this approach is rapidly becoming obsolete given the use of shotgun cloning for whole genome sequencing projects.

Recent developments in transposons

A major drawback of using mutant libraries was that each mutant had to be screened individually. Therefore, researchers were limited to analysing large libraries of mutants in simple assays. Signature-tagged mutagenesis (STM) was devised to overcome this (Fig. 1). In STM, each mutant is marked with a unique 40 bp DNA sequence identifier, so that it can be readily distinguished. Mutants can therefore be analysed in pools containing many individual mutants, allowing high-throughput screening of libraries in complex assays. STM has been mainly used to study the pathogenetic mechanisms of Gram-negative and Gram-positive bacteria, and fungi in animal models of disease. The results have provided unexpected insights into the genetic basis of disease and the environments that bacteria encounter during pathogenesis. For instance work on *S. typhimurium* led to the discovery of a previously unidentified genetic region containing clusters of genes required for pathogenesis. STM and similar methods do not have to be restricted to use in studies on pathogenesis; they can also be adapted to high-throughput analysis of gene function within the laboratory.

Conventional transposons are usually delivered into the organism of interest where they excise from a donor plasmid into the chromosome. For a number of microbes, this approach is not effective. Recently, *in*

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vitro mutagenesis has been successfully employed to previously intractable bacteria. In this method genomic DNA from the host bacterium is modified by combining it *in vitro* with a transposon along with its purified cognate transposase. The DNA is then returned to the host by transformation. The main constraint on this approach is that to be successful, the host bacterium must be transformable. A number of systems for *in vitro* mutagenesis are available, including Tn*7*, Tn*10* and mariner, and these have been applied to a variety of micro-organisms, including *Haemophilus influenzae*, *N. meningitidis* and *Campylobacter jejuni*.

In vitro mutagenesis has been adapted to identify essential genes. Proof-in-principle for this method was demonstrated in work on *H. influenzae*. Large fragments of genomic DNA were amplified by PCR, then subjected to *in vitro* mutagenesis. The modified products were then returned to the host by transformation. 'Essential' genes were identified by comparing the profile of insertions in the PCR products with insertions in the bacterium. This allows a systematic analysis of essential genes in transformable bacterial pathogens which may be useful for drug development, and if surface-located, for vaccine development.

Future directions

One of the major challenges facing microbiologists is how to exploit the wealth of information from whole genome sequencing. A striking feature is the large proportion of genes of unknown function. Some of these genes are conserved across genera, suggesting that they have important functions which have so far eluded researchers. There is now an urgent need for systematic analysis of gene function and central to this is the construction of ordered libraries of mutants which contain strains with deletions in each and every gene. So far, transposons have been used for making libraries of random mutants, but now they must be adapted to constructing ordered libraries that will allow large-scale and truly comprehensive analyses of gene function.

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Letter

PHOTOS IAN ATHERTON

Dear Editor

I always enjoy reading *Microbiology Today* but, as someone with a special interest in the fungi, the August 2000 issue was delightful. Although, as the articles clearly show, there is no doubt about the importance of the fungi in the biosphere and the well-being of humans, there is some anxiety about the future for the discipline of mycology. I am optimistic that the exposure of mycology in *Microbiology Today* will contribute to persuading a few young microbiologists that the fungi are an important and interesting group of organisms worthy of their attention.

You ask for names of the splendid group of photographs on the front cover. It is generally considered unwise to give names on the basis of photographs alone, but one can say with certainty that they are all fruit bodies of basidiomycetes! Top left is probably *Coprinus plicatilis*, a member of the grassland flora, although there are several similar species of these delicate and very beautiful *Coprinus*. They all have black spores and many species liquefy on maturity giving them the popular name of 'Inkcaps'. Middle left is undoubtedly *Auricularia* (*Hirneola*) *auricula-judae*, commonly known as the 'Jews' Ear', which is most frequently found on dead elder wood. These fruit bodies have the remarkable ability to dry out completely as thin tough structures which, after rainfall, can absorb water to expand considerably and continue disseminating their spores. At the bottom is a splendid photograph of a species of *Russula*. It is indeed dangerous to name species of *Russula* without additional information about habitat, spore colour and microscopic examination. However, the colours look perfect for *Russula atropurpurea* (= *R. krombholzii*), which may be mycorrhizal with a number of species of trees but is especially associated with oak.

Top right clearly has white gills and looks like an old clump of the honey fungus *Armillaria mellea* after the scales which are usually present have worn off. If this is correct then this would represent a fourth habitat, for this species is capable of parasitizing shrubs and trees and then continuing to grow and eventually fruit on the dead wood. Even if the names are not completely correct these four photographs do demonstrate the diversity of habitats of the basidiomycetes; growing on organic material in the soil, growing on and decomposing dead wood, growing symbiotically in a mutualistic association with the roots of trees and growing as a pathogen of living trees.

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Nanobacteria: gold mine or minefield of intellectual enquiry?

Allan Hamilton

Nanobacteria are very small cellular forms which seem to be closely linked with the formation of geological strata. Do they really exist? If so, their impact on our understanding of living systems is potentially huge.

Amongst microbiologists and molecular biologists there is a general consensus that the minimal size for independent viable life forms is likely to be a coccus of approximately 0·2 μm diameter with a volume of 0·06 μm^3. Perhaps not entirely coincidentally, this equates closely with the limits of the optical microscope and with the pore size of so-called sterilization filters, although it is also supported by various theoretical calculations of the minimal space required to accommodate the essential components of cellular life, as we currently understand them.

Nonetheless, from time to time there have appeared reports of various mini-forms such as granules, inclusion bodies, minicells and ultramicrobacteria. With the probable exception of ultramicrobacteria, these various mini-forms have not been extensively characterized, although they generally appear to be associated with conditions of stress and/or infection of higher organisms. The recent emergence of so-called nanobacteria, however, has dramatically re-opened the debate as to the minimal size for viable life forms and brought an urgency to research activity in subject areas as apparently diverse as infectious diseases of humans, microbiology of the deep subsurface and the origins of life.

This is a field as yet more associated with unanswered questions than with firm data and definite conclusions, but it is not a field lacking in strong characters and audacious claims. If even some of the latter can be substantiated then we are indeed sitting on a gold mine of intellectual enquiry, albeit one currently guarded by something of a minefield of circumstantial and uncorroborated findings.

To progress further, we must find the answers to questions that can be set at three different levels of enquiry, of increasing complexity and of widening relevance.

- What are the characteristics used to define nanobacteria?
- What do we know and what remains to be determined to establish their biological nature?
- What roles, if any, do they play in human disease, biomineralization and geological evolution, and the origins of life on this and other planets?

Characteristics and habitats

The major property used in characterizing the various mini-forms, i.e. size, is notoriously difficult to establish with any accuracy and free of artefactual distortion with objects in the 1 μm and less range. It is always possible, therefore, that either a single name can be used to identify two or more phenomena, or that the same basic structure can be given two names depending on the conditions of its discovery. Something of the latter may be the case with the terms ultramicrobacteria and nanobacteria. Ultramicrobacteria have been described as resting cellular life forms, of 0·3 μm diameter, derived from marine bacteria in response to stress, notably starvation. On nutrient supplementation, normal cell size and growth are re-established. More recently Jan Gottschal has proposed that ultramicrobacteria are naturally occurring organisms, again primarily found in the marine environment, which have a marked survival capacity, grow extremely slowly and retain their small size. With the exception of their larger dimension, this description of ultramicrobacteria fits closely with what is envisaged for nanobacteria. At this stage of our understanding, therefore, it might be best to consider the terms as synonymous.

The undoubted champion of nanobacteria (except that he prefers the spelling nannobacteria) is Robert Folk, a distinguished sedimentary geologist from Austin, Texas. Using techniques of acid etching and gentle gold shadowing, Folk has demonstrated the presence of tiny spherical structures (in the size range 0·05–0·2 μm) in an extensive array of geological materials. He has hypothesized that these are microfossils of previously active nanobacteria and that their activities were central to the actual formation of the geological strata in question. Folk has further claimed that evidence for extant nanobacteria is to be found in such diverse environmental samples as tap water and decaying leaves. These data have been published largely in journals and conference proceedings within the geological literature, and Folk has not yet found a particularly receptive audience within the microbiological community.

Potentially a major advance in this last regard came with a publication in *Proc Natl Acad Sci* in 1998 by the Finnish group of Kajander & Ciftcioglu. The authors demonstrated nanobacterial forms in human and cow blood, and in commercial cell culture media. They were able to grow these nanobacteria in normal culture media and to show that the cells laid down deposits of biogenic apatite on their cell envelope. Kajander & Ciftcioglu proposed on the basis of these data that nanobacteria are common organisms within the animal body and may be ultimately responsible for conditions such as tissue calcification and kidney stones. Further, they extracted 16S rRNA from their nanobacteria, and from gene sequence analysis deduced that they belonged within the α-2 subclass of the *Proteobacteria*. Unfortunately, it appears that the Finnish group had experienced a period in the scientific wilderness prior to their paper and that even since that publication their work has met with considerable scepticism in general and, as reported in *Nature* last year, downright antagonism from within the Finnish academic community.

Taking the more charitable view that such criticism and rejection as have been experienced by Folk and the Finnish group owe more to the challenging novelty of their findings than to any inherent fault in their scientific enquiry, we can say that, as a working hypothesis, nanobacteria are defined as extremely small cellular

forms, widespread in nature and closely associated with the formation of inorganic precipitates and geological strata.

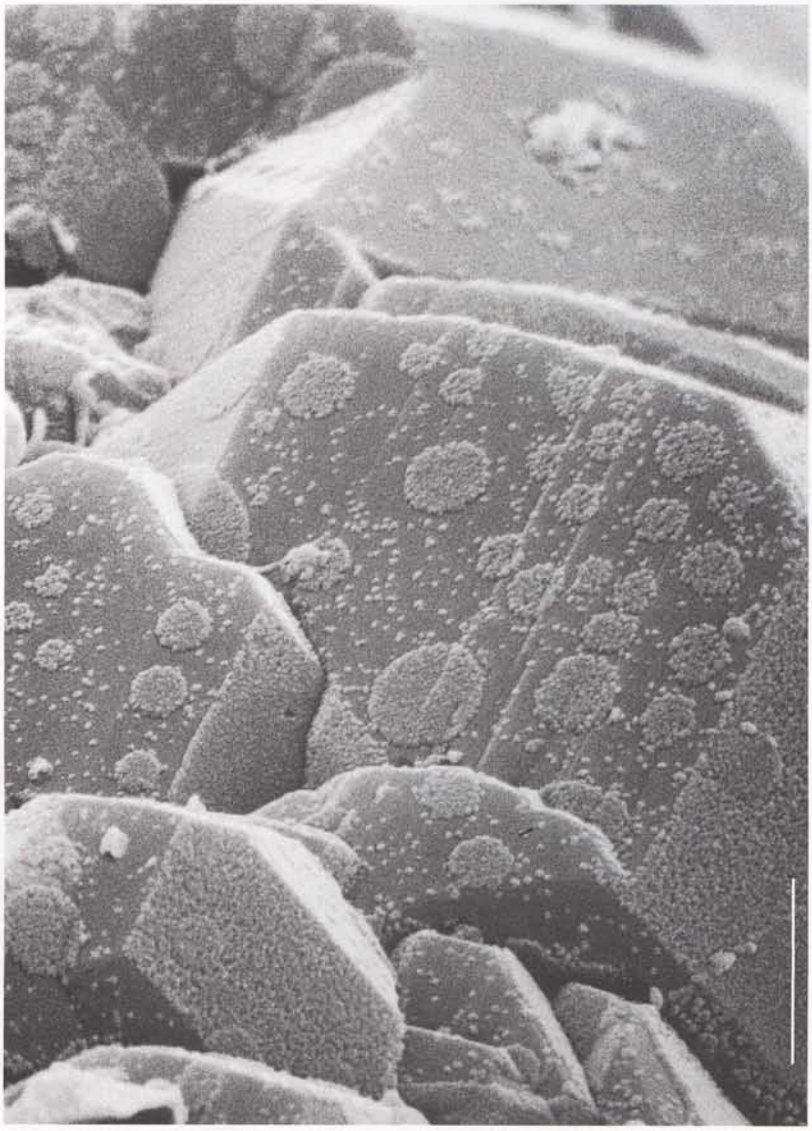
Moving to the next stage of our analysis, at least three criteria can be used to establish whether nanobacteria are true life forms. Do they demonstrate increase in biomass during incubation in nutrient media? Can evidence be obtained of the presence of nucleic acids? Can cellular structure be demonstrated? Although Folk's geological studies cannot shed any light directly on these questions, the Finnish group has claimed positive answers in each case. Is there any corroboration from other research groups on this key issue?

Another geologist, Philippa Uwins, working at the University of Queensland, Australia, has found filamentous forms associated with various rock samples. These resemble actinomycetes or fungi rather than bacteria, but their very small dimension (0·020–0·128 μm diameter) has caused them to be identified as nanobes. Uwins and her colleagues have been able to demonstrate growth of these nanobes, membrane and wall structures and the likely presence of DNA. These studies have elicited the, by now normal, expressions of scepticism, with the main criticism focussing on the diversity of size and shape which is taken to suggest that the nanobes may be simply fragmentation products of a larger cellular life form.

Nanobacteria in the deep subsurface

My own involvement with and interest in nanobacteria stems directly from the scanning electron microscopy (SEM) picture opposite. This was obtained by my colleagues Carol Devine and Iain Spark as part of a combined microbiological and geological study of reservoir souring and formation damage in the offshore oil industry. It shows fresh sandstone core material taken from a subsea depth of 13,000 feet, after 7 days incubation in nutrient medium at 90 °C. Individual structures are less than 0·1 μm in diameter and they show a striking circular aggregation pattern. These nanobacteria are not evident on core material prior to incubation, nor are they found in the liquid medium at the end of the incubation period; their presence appears to require both nutrient medium and a geological surface. Similar patterns were found after incubation at both 60 and 30 °C, but only after 28 days and 6 months, respectively. At each temperature, increasing the time of incubation gave rise to increasing biomass, as evident in SEM analyses. These nanobacterial forms, uniquely associated with core material from the deep subsurface, therefore show the standard temperature and time dependencies characteristic of cellular biological forms added to nutrient media.

While it must be freely admitted that all the evidence so far available concerning the existence and nature of nanobacteria can best be described by such euphemisms as 'preliminary' or 'indicative', this author would wager that while nanobacteria may be very small, their impact on our future understanding of living systems has the potential to be very large indeed.



ABOVE:
Circular aggregations of <0·1 μm structures on fresh sandstone core material as described in the text. Bar, 3 μm.
COURTESY CAROL DEVINE AND IAIN SPARK

The work of the Finnish group places nanobacterial forms as central to a wide range of medical conditions, and notably those associated with the formation of inorganic precipitates. Fortuitously or otherwise, the other groups whose work I have very briefly summarized have seen nanobacteria as agents at least associated with, and possibly even responsible for, many of the geological formations previously assumed to be wholly sterile and to have evolved quite independently of any biological processes. The comparatively recent demonstration that the deep subsurface is, in fact, an extensive and wholly active biosphere in its own right, has clearly shown the error of that assumption. There is even an increasingly

widespread belief that the surface-associated high-temperature characteristics of the deep subsurface constitute the ideal environment for those initial processes leading to the origins of life itself. All of which takes us neatly to NASA, meteorite ALH84001 and life on Mars!

Life on Mars?

In 1996 *Science* published a paper suggesting that a Martian meteorite, identified as ALH 84001, carried evidence of previous life on that planet. This striking claim rested on a number of features which together were taken as being compatible with life on Mars: carbonate globules, and magnetite and iron sulphide particles whose chemistries were suggestive of biological processes of formation; the presence of polycyclic aromatic hydrocarbons; and SEM and transmission electron microscope (TEM) images very closely resembling Folk's terrestrial nanobacterial fossils. Perhaps not surprisingly, this publication aroused considerable interest, including a Presidential press conference. Now, however, it has become fashionable in most scientific circles to debunk the original claims as being greatly exaggerated and largely unsustainable. In such an emotive area, where there is a great human desire to find a definite 'yes' or 'no' to a question where all the data available to us must by its nature be indirect and circumstantial, it is important to resist the temptation to take a polarized position. As one of the authors of the original paper has subsequently stated,

> *'It is important to stress that we have not found proof of life on another planet; rather, we observed features that are nearly identical to those we might expect had life once existed on Mars. This may be a fine distinction, but it is an important one'.*

The full nanobacterial story remains to be told, but its resolution and the light it may shed on other great scientific mysteries holds promise of being hugely exciting.

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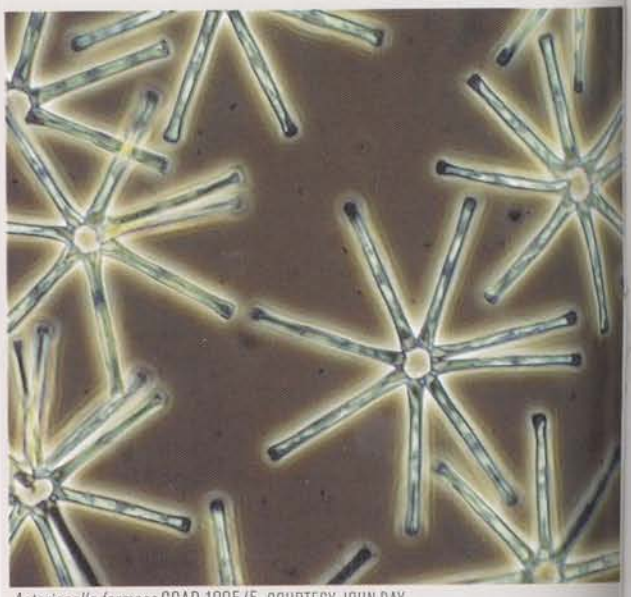
UKFCC logo competition – £250 prize

Micro-organisms, which are exceptionally diverse, are found almost everywhere and affect human society in countless ways. Thus, modern microbiology has a great impact on medicine, agriculture, food science, ecology, genetics, biochemistry and many other fields. The microbial world holds the potential to revolutionize many aspects of human life and culture collections are a key resource that is required to underpin these developments.

It is easy for those not actively involved in microbiology to overlook the importance of culture collections, but without 'biological standards' it would be impossible to perform comparative science, authenticate specimens, guarantee productivity in processes using micro-organisms/cell lines or lodge patents involving micro-organisms. In the UK there is a huge diversity of collections, both large and small, often associated with an individual scientist or research group. Such collections include those in the PHLS, the university sector, industry and the constituent collections of the UK National Culture Collection (UKNCC). The United Kingdom Federation for Culture Collections (UKFCC) aims to serve all those who are involved with or use culture collections.

The UKFCC was established on 10th April 1975 at Imperial College London. Membership of the Federation is open to all those involved with Culture Collections/Genetic Resource Centres (GRCs) and the users of cultures. Today, its membership includes users of genetic resources from both the academic and commercial sectors, as well as representatives of the constituent collections of the UKNCC. Members receive a twice-yearly newsletter, have reduced course fees on UKFCC training courses and are affiliate members of the World Federation of Culture Collections (WFCC).

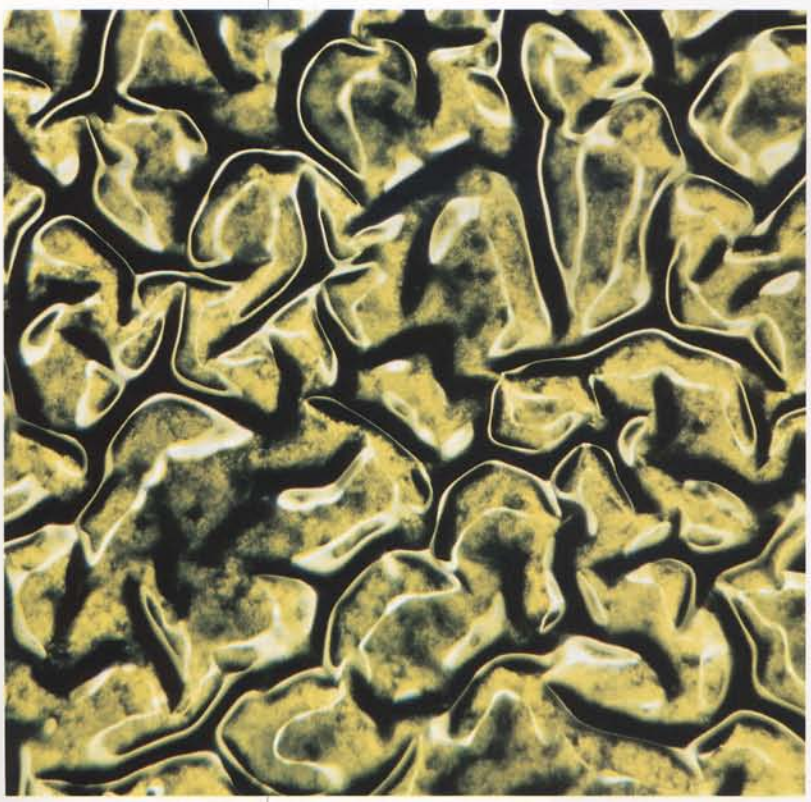
In its 25th year the UKFCC has initiated a competition (first prize £250) to design a new logo for the Federation. For further details contact the Secretary of the UKFCC: Dr John G. Day, CEH Windermere, Far Sawrey, Ambleside, Cumbria, LA22 0LP (Tel. 015394 42468; Fax 015394 46914; email jgd@ceh.ac.uk).

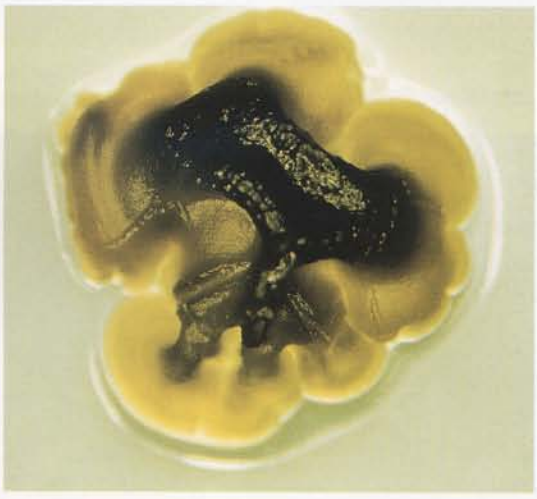


Asterionella formosa CCAP 1005/5. COURTESY JOHN DAY

Photo2000
Photographic competition

The SGM's first photographic competition was judged at the Society's meeting at the University of Exeter in September. There were 11 entries and the judging panel, made up of new President Sir David Hopwood, General Secretary Alan Vivian, Education Officer Liz Sockett, Executive Secretary Ron Fraser and Public Affairs Administrator Tracey Duncombe, had a difficult job deciding on the best shot. Eventually a consensus was reached and the winners were announced by outgoing President Howard Dalton at the Society Dinner.





First prize of £250 went to **Karen Jolly**, a second year PhD student at Leeds University, for her photograph of *Streptomyces coelicolor* producing the antibiotic undecylprodigiosin (top). Runner-up, with his two pictures of the actinomycete *Micromonospora echinospora* (above left and right) was **Paul Hoskisson** of Liverpool John Moores University.

Another competition will be held next year – start snapping now!

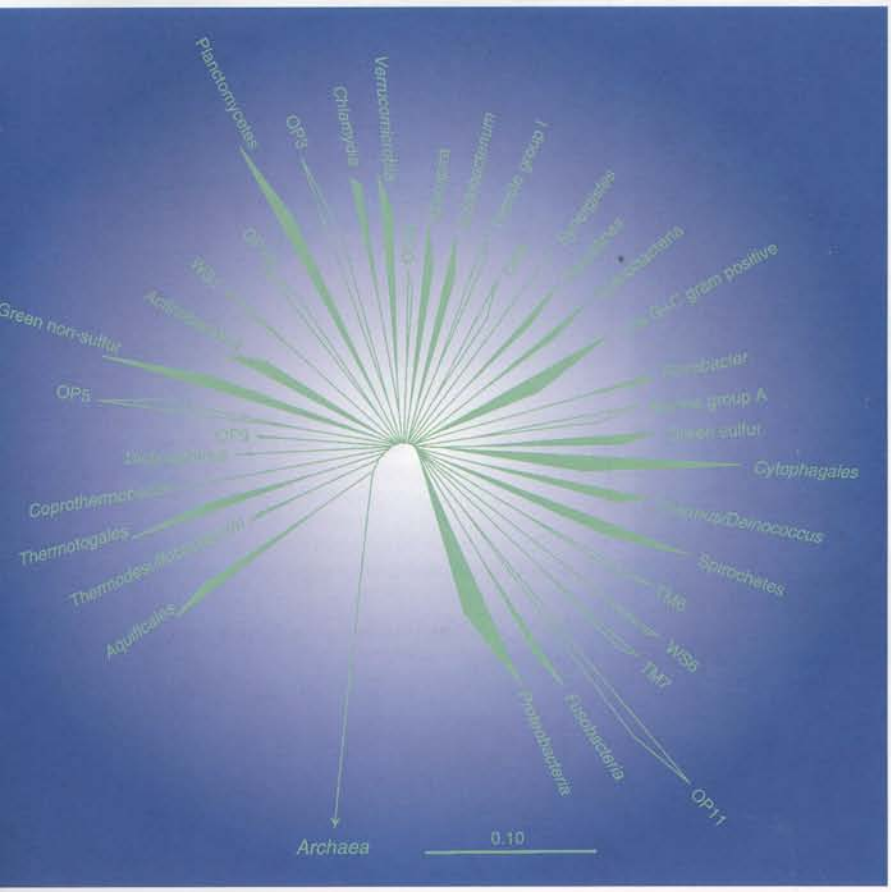
Bacterial diversity and 'unculturables'

John Fry

Many previously unknown groups of bacteria have been identified by 16S rRNA gene sequencing. John Fry speculates how unculturable many of these microbes really are.

Cataloguing and preserving the world's biodiversity is now a priority for many governments and non-governmental organizations with major interests in the environment. This is relatively straightforward for groups of organisms that are relatively large, obvious and well studied such as mammals, birds and plants. However, for smaller, less obvious organisms whose diversity is relatively poorly studied, categorizing and preserving biodiversity is exceedingly difficult. Microbes present particular difficulties, as individuals cannot be observed with the naked eye. Furthermore, bacteria cannot be identified morphologically and so less direct methods must be used.

Despite these difficulties bacteria are vitally important organisms for mankind. They are ubiquitous and it has been recently calculated that the earth's bacterial biomass is almost as great as that of plants. They play a vital role in cycling all biologically important elements and are the major agents of infectious disease. Lastly, they are posed to increase in importance to industry as the predicted biotechnological revolution develops. For all these reasons it is vital to understand the full extent of bacterial diversity on our planet and the roles of the most abundant species. We cannot do this until we can culture the dominant bacteria in all the major habitats on the globe. As currently only a small minority are in culture and new bacterial species are being described quicker than ever before, there is a lot of work to be done.



BELOW
Fig. 1. Phylogenetic tree of the recognized divisions in the domain *Bacteria*. The wedges show division-level groupings with >1 sequences, hollow wedges have no cultured representatives, solid wedges have sequences from cultured and uncultured bacteria and wedge width indicates the range of sequence diversity within a division. The diagram illustrates the position as it was known in 1998.
AFTER HUGENHOLTZ *ET AL.* (1998)

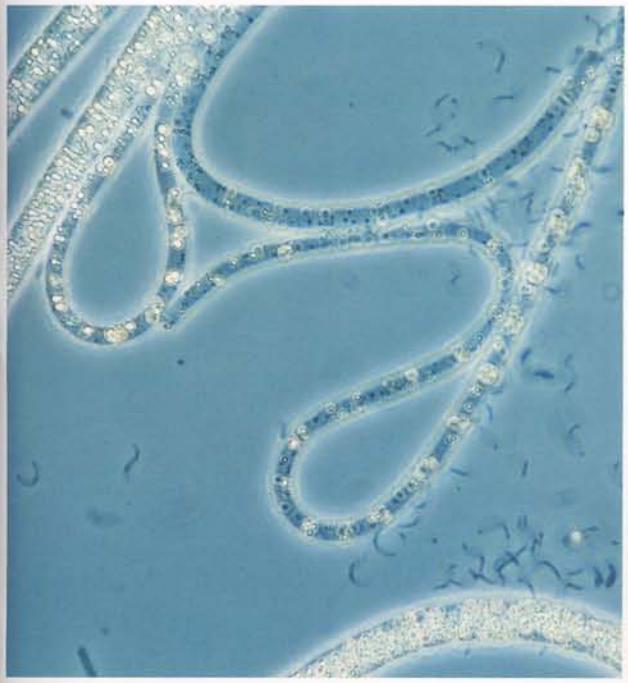
The molecular era

Traditionally, in the 1980s and before, the diversity of bacteria was assessed by identification with phenotypic tests and numerical taxonomy from collections of isolates obtained from plating on nutrient media and liquid enrichments. However, plate counts of bacteria from natural habitats, such as soil, freshwater and the sea, are much lower than direct total counts and it is accepted that <1 % of these bacteria are culturable. For this reason studying the true diversity of bacteria in nature was impossible.

This problem was overcome in 1990 when Stephen Giovannoni and David Ward's research groups first investigated bacterial diversity in the Sargasso Sea and in hot springs using molecular approaches based on sequence analysis of the highly conserved 16S rRNA gene. This was achieved in the Sargasso Sea study by extracting community DNA, PCR amplification of 16S rRNA genes, sequencing and identification of the source of these genes by phylogenetic analysis. This approach allows bacteria to be identified without culturing them and so for the first time allowed microbiologists to study the 99 % of bacteria that cannot grow on enumeration plates. Culture-independent studies of this kind are now common and many bacteria have been found that are only known from their 16S rRNA gene sequences and that do not match known cultured bacteria.

In the 10 years since 1990 many new bacteria have been identified solely from their 16S rRNA gene sequences and many of these are known to be numerically dominant in nature. Taking the marine habitat as an example, most bacteria identified by molecular methods are in the divisions *Proteobacteria* (67 %) and *Cytophagales* (25 %). These large groups are roughly equivalent to phyla. Furthermore, they contain most of the best-studied Gram-negative bacteria, which are more easily cultured than the members of the other divisions. Over 60 % of the *Proteobacteria* and over 40 % of the *Cytophagales* are culturable. Despite this many phylogenetic groups within these divisions have members only recognized by 16S rRNA gene sequences and so are perhaps unculturable. I say perhaps because many microbiologists now believe that if similar effort was put into culturing these bacteria as has been expended on culturing bacteria of medical importance over the last century, then most could be cultured.

The α-*Proteobacteria* are an important marine group, making up on average 23 % of marine bacteria, and most of these fall into two phylogenetic groupings. The first, the SAR11 cluster of 16S rRNA gene sequences,



have been found in many studies throughout the world's oceans, but have never been cultured. Conversely, many examples of the other common marine α-*Proteobacteria* group, the *Roseobacter* cluster, have recently been isolated as colonies on simple nutrient media from coastal water and their physiology has just started to be studied. This indicates that whereas some novel uncultured bacteria will be difficult to isolate, others will be easy. We must try to culture the most abundant bacteria in all environments to study their physiology, to understand their roles and to tap this large biotechnological resource.

Expanding knowledge of bacterial biodiversity

In 1990 about 10 divisions of the domain *Bacteria* were known. Now 40 have been described and this remarkable expansion in our knowledge of bacterial biodiversity has occurred entirely due to the recent explosive growth of molecular approaches (Fig. 1). Furthermore, 13 of these divisions are currently known only from sequences and have no cultured representatives. Some of these divisions of uncultured bacteria are phylogenetically extremely varied, for example WS6 and OP11 are the most diverse divisions known, showing 26 and 33% sequence divergence, respectively.

These observations further indicate the need for greater effort in growing these organisms and will be illustrated by consideration of three bacterial divisions known to be abundant from their 16S rRNA sequences but with few cultured representatives.

The *Nitrospina* division

In 1994 Erko Stackebrandt's group reported that a newly isolated Gram-negative, obligately anaerobic, heterotrophic bacterium belonged to a new bacterial group. They called this organism *Holophaga foetida* as it originated from smelly, anoxic sediment and suggested it could belong to the δ-*Proteobacteria*. At a similar time my research group in Cardiff obtained several bacterial 16S rDNA sequences from very deep, Japan Sea sediment, which also seemed to be deep branching δ-*Proteobacteria*, called the JAP504 cluster. Collections of further 16S rDNA sequences have now revealed that these bacteria belong to a new division, named the *Nitrospina* division after *Nitrospina gracilis*, which is also in this group. This division has representatives from a wide variety of marine, freshwater and terrestrial habitats and consists of 12 subgroups made up of 188 sequences, about 94% of which are from uncultured organisms. The two named cultures in this division are physiologically very different; *H. foetida* is an anaerobic heterotroph degrading a plethora of complex organic compounds and *N. gracilis* is an aerobic, nitrifying chemolithotroph. Such diversity indicates that this division is not only widespread in nature, but also as physiologically diverse as the *Proteobacteria*.

The WS6 division

The WS6 division is one with no cultured representatives. Earlier this year Norman Pace and colleagues designed PCR primers to amplify the 16S rDNA of members of this division from 12 diverse environments. They found WS6 members to be most abundant in anaerobic environments but were also present in some aerobic habitats. They found 57 different 16S rDNA clone types that increased the number in this division to 60. These clones were isolated from marine, freshwater and hot-spring sediments, contaminated aquifers and one from topsoil. They surmise that members of this division might be anaerobic, but until they are cultured this will not be confirmed. This study convincingly shows that the undiscovered bacterial diversity in the environment, from even widely distributed habitats, is almost certain to be enormous.

ABOVE LEFT:
Phase contrast micrograph of an agar-coated slide enrichment of bacteria from freshwater sediment. There are filamentous (probably *Beggiatoa* spp.)and non-filamentous bacteria present, probably representing culturable and non-cultured species.
COURTESY J.C. FRY

ABOVE RIGHT:
A sub-tropical river with sediment that is a likely source of many uncultured bacteria known only from their 16S rRNA gene sequences.
COURTESY H.G. WILLIAMS, CARDIFF

Further reading

Barnes, S.M., Takala, S.L. & Kuske, C.R. (1999). Wide distribution and diversity of members of the bacterial kingdom *Acidobacterium* in the environment. *Appl Environ Micro* 65, 1731–1737.

Dojka, M.A., Harris, J.K. & Pace, N.R. (2000). Expanding the known diversity and environmental distribution of an uncultured phylogenetic division of *Bacteria*. *Appl Environ Microbiol* 66, 1617–1621.

Giovannoni, S.J., Britschgi, T.B., Moyer, C.L. & Field, K.G. (1990). Genetic diversity in Sargasso Sea bacterioplankton. *Nature* 345, 60–63.

Hugenholtz, P., Goebel, B.M. & Pace, N.R. (1998). Impact of culture-independent studies on the emerging phylogenetic view of bacterial diversity. *J Bacteriol* 180, 4765–4774.

For access to the current range of aligned sequences of 16S rRNA genes and associated information for cultured organisms and environmental isolates go to the Ribosomal Database Project website at **http://www.cme.msu.edu/RDP/html/index.html**

The *Acidobacterium* division

The distribution and diversity of the *Acidobacterium* division, which has only one cultured member, has been investigated by similar methods. These bacteria are very widespread, they are present in many different soil types, marine and freshwater sediments, as well as in hot-spring mats and sediment. Furthermore, they sometimes form the dominant group in a habitat. In one set of arid soils they were the dominant phylogenetic group making up 51% of the 16S rDNA clones isolated, but were entirely absent from culture plates from the same habitat. Despite this, culturing some of the group might not be difficult because the one cultured member of the division, *Acidobacterium capsulatum*, is an aerobic, mesophilic, chemo-organotroph able to use a variety of carbon sources and to grow up to pH 6·0. However, other members of the division might well need specialized approaches for their culture.

Future challenges

These examples indicate that culturing many of these 'unculturable' bacteria will be an enormous task. However, the following arguments suggest that if more effort were put into growing these bacteria more of them would prove culturable. Many bacteria that are grown on plates do not match existing cultured bacteria. When effort is put into growing novel aquatic bacteria they are sometimes grown relatively easily once suitable media are developed (e.g. *Legionella* spp.). Little research effort is expended in studying unspecialized, aerobic, heterotrophic bacteria, whilst studies of specialist groups abound, even when they are hard to cultivate (e.g. methanogens, sulphate-reducing and nitrifying bacteria). In 1995 Karl Stetter's research group was the first to successfully culture a prokaryote only previously identified from a 16S rRNA gene sequence. In this case the organism was a hyperthermophile from a hydrothermal vent and isolation was by selective enrichment, *in situ* hybridization with a 16S rRNA probe and micromanipulation. Similarly, a *Thermus aquatilis*-like strain and *Synechococcus lividus* strains that were prevalent as gene sequences, but not yet isolated in culture, have been isolated from hot-spring mats by David Ward's group. So I believe that the time is right to attempt to obtain further cultures of some major groups of 'unculturable' marine bacteria. Furthermore, the *Roseobacter* and *H. foetida* examples described above add to the arguments for cultivating the uncultureables.

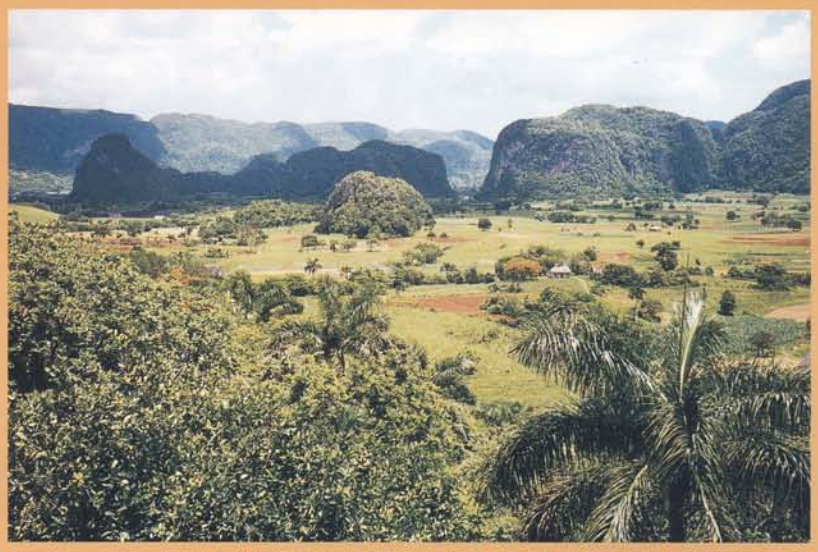
● *John C. Fry is Professor of Microbial Ecology and Deputy Director of Cardiff School of Biosciences, Cardiff University, Main Building, Museum Avenue, Cathays Park, Cardiff CF10 3TL.*
Tel. 029 2087 4190; Fax 029 2087 4305; email fry@cardiff.ac.uk

LEFT:
Part of an activated sludge aeration tank from a sewage treatment works showing inflows of settled sewage (lighter coloured inflow) and re-circulated sludge (darker coloured inflow). Once again this is a likely source of many uncultured bacteria known only from their 16S rRNA gene sequences.
COURTESY J.C. FRY

International Development Fund report

Culture collection management in Cuba

Franklin Sotolongo



UPPER:
The Viñales Valley in north-west Cuba.
COURTSEY P.M. GREEN

LOWER:
Some of the participants on the *Culture Collection Maintenance and Management* course, June 2000.
COURTSEY P.M. GREEN

Cuban Science Base

The Cuban National System for Science and Technology has sectoral entities such as health, agriculture, etc. much in the way that the UK has government departments responsible for these disciplines. These bodies are responsible for carrying out the R&D in their respective areas. National technical and scientific activities are developed as an integrated model relevant to the participating centres from each sector. In this way, they draw on each other's strengths and maximize the potential of each speciality; dealing with issues such as human resources and financial and structural capacities to reach some of the social and economic goals of the Cuban government. This integrated approach is aimed at quickly and efficiently reaching the objectives proposed through various committees within the Ministry of Science, Technology and the Environment.

This strategy has given rise to the creation of many diverse scientific institutions in Cuba, rising from only 21 in 1951 to over 200 today and including production, biotechnological, biomedical and pharmaceutical centres.

The core activities of several of these scientific institutes involves basic microbiology and it has been necessary to study techniques concerned with the preservation of micro-organisms. Personnel have been trained to set up and manage culture collections.

At the beginning of the 1990s, a directive of the Academy of Sciences of Cuba encouraged the establishment of individual culture collections in each appropriate Institution. Some thought was given as to how these should function and develop and several laboratories and individual scientists wished to set up a Cuban Federation of Culture Collections. The current strategy is aimed at establishing the requirements of such a federation in terms of logistics, co-operation and training.

Culture collection management

Workshops and national courses have taken place to promote and organize the setting up of culture collections in research and production centres within Cuba which deal with different types of micro-organisms. The first international course on *Culture Collection Maintenance and Management* was held in the Finlay Institute, Center for the Research, Development and Production of Vaccines in Havana, Cuba, from 20 to 21 June 2000. The course was co-ordinated by Dr Peter Green from the National Collection of Industrial, Food and Marine Bacteria (NCIMB), UK, assisted by Dr David Smith of CABI Bioscience and Dr John Day from the Culture Collection of Algae and Protozoa (CCAP) and some members of the National Cuban Collection Culture Group. It was attended by 60 specialists from different scientific and production facilities in Cuba.

In addition to training and education, the course also provided a great opportunity for the participants to exchange experiences. The course included formal lectures, video presentations and workshops on the following topics:

- Culture collection function and quality management
- Culture collection services and activities
- Principles and theory of freeze-drying
- Principles of cryopreservation
- Preservation techniques for bacteria, fungi, algae and protozoa
- Databases, catalogues and websites
- Demonstration of the UKNCC website
- Postal and packaging regulations for the transportation of cultures
- Acceptance and maintenance of patent cultures
- Quality assurance in culture collections
- Impact on culture collections of the Convention on Biological Diversity

The focus afforded by the meeting and the useful information and exchange of ideas will, I hope, provide the impetus for the loosely affiliated Cuban Federation to proceed on a more formal basis and will allow us to move to the next stage of policy making, planning and fund raising.

Finally, we would like to thank the UK lecturers for organizing the course and the SGM for the award from its International Development Fund, without which this meeting could not have taken place.

● *Dr Franklin Sotolongo is Assistant Director of Technical and Scientific Applications, Finlay Institute, Havana, Cuba*
email fsotolongo@finlay.edu.cu

July Council Meeting

Comings and Goings

● The President welcomed Professor Sir David Hopwood, President-elect and Dr Meriel Jones, the new Editor of *Microbiology Today*, to their first Council meeting. This was Professor Dalton's last meeting as President and he was warmly thanked for his achievements during his term of office, in which he has sought to expand the activities of the Society. Professor Jon Saunders also retired as Editor-in-Chief of *Microbiology* and his unstinting contribution to the well-being and quality of the journal during his term of office was noted. We wish him well in his new post as Dean of Science at Liverpool. Dr Pat Goodwin retires as Scientific Meetings Officer and Council recorded its appreciation for all her hard work since joining Council in 1994. Howard Jenkinson is the new Scientific Meetings Officer. Dr Dave Roberts retires as the last Publications Officer and there were warm tributes for his contribution, which has seen the creation of *Microbiology Today*, setting new standards for the Society's members' magazine. Thanks were also recorded to the three elected members who retire from Council at this time – Dr Geoff Clements, Professor Dave Rowlands and Professor Liz Wellington. For all their contributions and dedication we wish our retiring colleagues well in the future.

JGV Direct

● Council learned with pleasure that the new on-line service for the *Journal of General Virology*, described on page 151 of the August issue of *Microbiology Today*, had been successfully implemented on the SGM website (www.sgm.ac.uk/JGVDirect). Around four articles per issue will be published on-line as full text HTML, up to 13 weeks ahead of print. The project had been lead by Debbie Ollman, aided and abetted by Robin Dunford. Publication of entire issues of on-line journals continues on the journal HighWire site (http://vir.sgmjournals.org).

Support for Joint Meeting between SGM and ASM

● Council noted that plans were well in hand for the forthcoming joint meeting on *Biodegradation, Biotransformations and Biocatalysis* to be held between SGM and the American Society for Microbiology. This meeting will be held from 2 to 6 October 2001 in Puerto Rico. See p. 199 for details.

More contact with ASM

● Following her successful participation in the recent Annual Meeting in Los Angeles, Dr Liz Sockett, the Society's Education Officer, has accepted an invitation to join the ASM Education and Training Board on behalf of the Society. She will also be a member of their International Committee.

Meeting Venues and their Costs

● Council spent some time considering the difficulties of predicting numbers in advance for likely attendance at its scientific meetings, with an eye on the financial repercussions for the Society of 'getting it wrong' (which members may not be aware of!). In future, members attending meetings will be asked for their opinions of venues through the use of 'feedback forms'.

● *Alan Vivian, General Secretary*

New Members of Council and Group Committees

Council

With effect from 13 September 2000, **Professor Sir David Hopwood** (John Innes Centre) commences his 3-year term as President, **Dr Meriel Jones** (University of Liverpool) her 3-year term as Editor of *Microbiology Today* and **Professor Howard Jenkinson** (University of Bristol) his 4-year term as Scientific Meetings Officer. **Professor Chris Thomas** (University of Birmingham) commenced his 5-year term as Editor-in-Chief of *Microbiology* on 1 July 2000.

Following the recent ballot of Ordinary Members, the following have been elected to serve as Members of Council for 4-year terms, commencing on 13 September 2000:

Professor Hilary Lappin-Scott	University of Exeter
Professor Tony Nash	University of Edinburgh
Professor Ian Roberts	University of Manchester

and to serve for 1 year, with eligibility for re-election in 2001:

Dr Keith Jones	University of Lancaster

Biographies of the new Council Members appear on p. 193.

Groups

New Committee members, elected by postal ballot (Virus Group) or elected unopposed (all other Groups) are as follows:

Cells & Cell Surfaces

Dr John Armstrong	University of Sussex
Dr Nicky High	University of Manchester
Dr Brendan Kenny	University of Bristol
Dr Rod McNab	Eastman Dental Institute, London

Clinical Virology

Dr Diana Westmoreland	University Hospital of Wales, Cardiff
Dr Bernard Cohen	CPHL Colindale
Dr Sheila Cameron	Regional Virus Laboratory, Glasgow

Education

Dr Richard Jenkins	de Montfort University

Environmental Microbiology

Dr Gary Black	University of Northumbria
Dr Frans De Leij	University of Surrey
Dr Ian Thompson	NERC Centre of Ecology and Hydrology, Oxford

Fermentation & Bioprocessing

Dr Glyn Hobbs (Liverpool John Moores University) has become Group Convener

Irish Branch

Dr Cyril Carroll	National University of Ireland, Galway
Dr Sean Doyle	National University of Ireland, Maynooth
Dr John Morgan	University College Cork

Microbial Infection

Dr Sheila Patrick	Queen's University of Belfast

Physiology, Biochemistry & Molecular Genetics

Dr Bill Ashraf	University of Bradford
Dr Ian Stansfield	University of Aberdeen

Systematics & Evolution

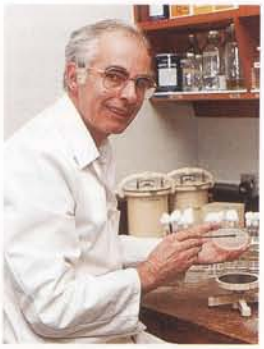
Professor Fergus Priest	Heriot-Watt University
Dr M Aquino de Muro	CABI Egham
Professor Mike Goodfellow	University of Newcastle upon Tyne

Virus

Dr Stacey Efstathiou	University of Cambridge
Dr Keith Leppard	University of Warwick
Professor James Neil	University of Glasgow
Dr Michael Skinner	Institute for Animal Health, Compton

New Honorary Members

Professor John Guest, FRS



John Guest has made many important contributions to the science and art of microbiology. With D.D. Woods he elucidated the role of vitamin B12 in methionine biosynthesis in *E. coli*. He made important contributions to Charles Yanofsky's demonstration of gene–protein colinearity and intra-codon recombination. Based in Sheffield for 35 years, he played a central role in the genetical and biochemical characterization of the citric acid cycle genes and enzymes of *E. coli* – discovering that several steps in the cycle are specified by multiple differentially regulated genes and providing a detailed molecular analysis of the pyruvate dehydrogenase complex. He discovered FNR, the oxygen-sensing transcription regulator, thus initiating an exciting field of research into the physiology of the aerobic/anaerobic interface. He also cloned and sequenced the aspartase gene that is used in the production of the sweetener, aspartame.

He has been a member of the SGM for almost 40 years and for seven years was an Associate Editor (1966–1969), then Member of the Editorial Board (1969–1973) of JGM. He organized the 1984 SGM Meeting in Sheffield and has been a strong supporter of Society journals (over 60 papers in *Microbiology*/JGM and FEMS publications out of an impressive list of over 260 published papers).

John was elected to the Royal Society in 1986 for his seminal contributions to microbial physiology. He delivered the Marjory Stephenson Prize Lecture in 1992 and the Leeuwenhoek Lecture of the Royal Society in 1995. For nearly 30 years he has been, arguably, the UK's leading *E. coli* physiologist and was certainly the first to embrace the use of molecular genetics and gene cloning technology for the analysis of central metabolic functions.

● *George Salmond*

Professor Sir John Pattison

Professor Sir John Pattison studied Medicine at the University of Oxford and spent pre-registration years at Middlesex Hospital Medical College before specializing in Clinical Virology at The London Hospital Medical College (MRCPath 1975). In the 1970s he was decisively involved in working out the causative role of parvovirus B19 for haemolytic crises in thalaessemia patients, for fifth disease, and also for intrauterine infections.

Since 1977 he has been Professor of Medical Microbiology at King's College Hospital Medical School, and since 1984 at University College of London Medical School where he became the Vice-Provost in 1994. He worked as an Honorary Consultant at various hospitals in London. He held various offices in the Royal Society of Medicine, the Society for General Microbiology, the Royal College of Pathologists, and he served as Member and Chairman of the MRC Physiological Medicine and Infection Board, later as Member of Council and Member of the Strategy Group, and most recently as Senior Clinical Advisor to the MRC Chief Executive. He also was a Member of the PHLS Board and the Board of the London School of Hygiene and Tropical Medicine, and more recently he became a Founder Fellow and Member of Council of the Academy of Medical Sciences. His most publicly visible role was that of a Member and later Chairman of the Spongiform Encephalopathy Advisory Committee (SEAC) (1995–1999) which had to advise on the difficult issues and decisions following the BSE epidemic and its possible consequences for humans. Since 2000 he has been working as Director of Research and Development in the Department of Health. He was knighted for his services to Medicine and Public Health in 1998.

● *Ulrich Desselberger*

New Group Conveners

David O'Connor

Cells & Cell Surfaces

David graduated as a zoologist in 1979 from Salford University before switching to Biochemistry through an MSc conversion course at University College London in 1980. Some deft advocacy on behalf of prokaryotes by Pat Clarke and Pauline Meadow kindled his interest in molecular microbiology, which was reinforced by PhD studies with Gwyn Humphreys and Jon Saunders in Liverpool on the genetics of restriction-modification systems. This was followed by a Royal Society Postdoctoral Fellowship (1984–1986) at the University of Geneva, where (along with skiing) he studied a class of highly cell-surface exposed bacterial lipoproteins in Ken Timmis's laboratory. In 1986, he returned to the UK to take up a New Blood Lectureship at the University of Southampton, where he is currently Reader in the School of Biological Sciences. David's research focuses on molecular aspects of bacterial stress adaptation and pathogenicity, which he is currently investigating using a range of functional genomic approaches, including proteomics.

Stephen Gillespie

Clinical Microbiology

Stephen graduated in Medicine from the Queen's University Belfast in 1980. After postgraduate training in Medicine and Medical Microbiology at the Royal Victoria Hospital, Belfast, he was appointed Mercer's Lecturer in Clinical Tropical Medicine at the London School of Hygiene and Tropical Medicine in 1985. Research interests were focussed on the interaction of acute phase proteins and *Streptococcus pneumoniae*, and field studies of parasitic infections in children at the Wellcome/KEMRI research unit in Kilifi. In 1989 he was appointed Senior Lecturer at the Royal Free Hospital School of Medicine where he continued his research interest in the molecular biology of *S. pneumoniae* and in clinical trials of anti-tuberculosis therapy in Tanzania. He was appointed Professor in 1999.

He has a passionate interest in history, is an enthusiastic but ineffective member of his village cricket team and directs the village church choir in repertoire from Vivaldi to Rutter.

Grants

Details of all Society grant schemes are now on the website at **http://www.sgm.ac.uk**
Most application forms can be downloaded.

Any enquiries should be made to the Grants Office at SGM Headquarters, Marlborough House, Basingstoke Road, Spencers Wood, Reading RG7 1AG (Tel: 0118 988 1821; Fax: 0118 988 5656; email: grants@sgm.ac.uk).

President's Fund

The President's Fund offers financial support to younger members of the Society for one of the following:

1. Travelling to present a paper or a poster on a microbiological topic at a scientific meeting
2. Attending a short course (up to two weeks)
3. Making a short research visit – larger awards are available for short research visits

1 & 2 – Smaller Awards

Maximum grants are:

- £125 for attendance at meetings/courses in the country of residence
- £200 for travel to another European country
- £300 for travel outside Europe

3 – Larger Awards (research visit)

Up to £2000 is available for making a short research visit of up to two months. The host institution may be overseas or in the country of residence.

All applicants must be resident and registered for a higher degree, or in a first postdoctoral position, in a country in the European Union. Only one application may be made to the fund during the term of a studentship or fellowship. The full rules of the scheme are published on the SGM website, from which application forms may be downloaded.

Postgraduate Conference Grants

Postgraduate Student Members of SGM currently resident and registered for a higher degree in the UK or another European Union country are eligible for a grant to cover the costs of accommodation and travel in attending ONE of the following Society meetings in 2001: Heriot-Watt University, Edinburgh, March; University of East Anglia, September; or any other SGM Group or Branch meeting. Application forms giving full details of the scheme were sent to all Student Members in the EU with their subscription invoices. The form can also be downloaded from the SGM website.

Seminar Speakers Fund 2000/2001

The purpose of the Seminar Speakers Fund is to promote talks on microbiological topics in departmental seminar programmes. Applications are invited from Higher Education Institutions where microbiology is taught for grants of up to £200 towards the travel, and if necessary, accommodation, expenses of an invited speaker. The full rules of the scheme were published on p. 92 of the May issue of *Microbiology Today*. Applications will be dealt with on a first come, first served basis during the academic year, which is defined as running from September 2000 to June 2001. Written submissions should be sent to the Grants Office at SGM Headquarters.

Vacation Studentships 2001

The Society offers a limited number of awards to enable undergraduates to work on microbiological research projects during the summer vacation. The purpose of the awards is to provide undergraduates with experience of research and to encourage them to consider a career in scientific research. The studentships provide support at a rate of £135 per week for a period of up to 8 weeks. An additional sum of up to £400 for specific research costs may also awarded. Applications on behalf of named students are now invited from SGM members in higher education institutions and research institutes. Details of the scheme are given below.

Rules

1. Applicants must be members of the Society working in a higher education institution or research institute in the UK or Republic of Ireland. Applications must be made on behalf of a named student. More than one application from a department/school will be considered, but in the case of several applications being submitted, departments/schools may be asked to rank the applicants.

2. Students must normally be in the penultimate year of their undergraduate course and registered at an institution in the UK or Republic of Ireland. Applications for students in their final year will not be considered. Medical students will be accepted at the end of their intercalated studies, but not during their elective period.

3. The research project must be on a microbiological subject. Studentships will not be awarded for projects that are part of degree work. A studentship may be held in a laboratory away from the normal place of study, but it must be located within the UK or Republic of Ireland.

4. Applications will be assessed by a Council Award Panel, based on the reports of two referees. The scheme is competitive and applications will be judged primarily on the scientific merits of the project and the suitability of the student. Once an award has been offered, it cannot be transferred to another student.

5. The awards will provide support for the student at a rate of £135 per week for a period of up to 8 weeks, and not usually less than 6 weeks. An additional sum of up to £400 for specified research costs may also be awarded. Grants are made to the institution to which the applicant belongs, not to the supervisor, on the understanding that it will administer the award.

6. It is a condition of the award that the student submits a brief report of the research at the completion of the studentship.

7. Applications must be made on the appropriate form, which is downloadable from the SGM website.

The closing date for applications is **28 February 2001**.

International Research Fellowships

Two applications were received for the first round of the new scheme and both were successful. **Professor Michael Danson**, University of Bath, has been awarded up to £1,200 to make a visit to Antarctica to carry out research into novel psychrophiles. **Dr Stacey Efstathiou**, University of Cambridge, has been awarded up to £4,273 to visit the Max von Pettenkofer Institut in Munich to gain expertise in the generation of mutant virus construction using bacterial artificial chromosomes.

The rules of the scheme were published on p. 91 of the May issue of *Microbiology Today*. The closing date for the next round is **30 November 2000**.

There are three rounds of applications each year and the closing dates for 2001 are **30 March**, **30 July** and **30 November**.

Application forms may be downloaded from the SGM website.

Public Understanding of Science Awards

Are you planning any projects to promote the public understanding of microbiology? Have you got a National Science Week event in mind? SGM can help. Grants of up to £1,000 are available to fund appropriate activities. Applications are considered on a first come, first served basis. The current funding year runs from June 2000–May 2001. See SGM website for details and an application form.

Undergraduate Microbiology Prizes

The scheme to encourage excellence in the study of microbiology by undergraduate students continues to be well received in universities in the UK and Republic of Ireland. Institutions offering an appropriate microbiology course were invited to nominate a student for an SGM prize, based on good performance in microbiology in the penultimate year of study for a BSc. The department was able to choose the type of assessed work for which the prize was awarded. Of the 70+ departments circulated, 42 made nominations. Each prizewinner will receive a certificate, a cheque for £50 and a year's free Undergraduate Membership of the Society.

Undergraduate Microbiology Prizes will be awarded annually and the invitations for nominations in 2001 will be circulated next May. Details are also available on the SGM website.

Staff News

Welcome to **Dr Catherine Tarbatt**, a new Staff Editor on JGV. Catherine comes to the Society after successfully completing a postdoc at the University of Warwick.

News of Members

The Society notes with regret the death of **Dr C.T. Calam** (member since 1945) and **Professor J.M. Macy** (member since 1979).

New Members of Council

Keith Jones

I graduated as a botanist from Nottingham University in 1963 and did my PhD on nitrogen fixation in marine cyanobacteria at Westfield College, London. I was appointed in 1966 to an assistant lectureship in microbiology at Lancaster and promoted to senior lecturer in 1981. I have also worked at the universities of Pretoria, South Florida, California, Hawaii, Naples and the University of Science and Technology, Kumasi, Ghana.

My early research was on the ecophysiology of nitrogen fixation by cyanobacteria and free-living heterotrophic bacteria in a range of temperate and tropical environments. More recently I have been investigating the distribution and behaviour of pathogens in farm and aquatic environments. Current projects include the role of birds as polluters and vectors for human disease, tracing sources of faecal pollution for bathing waters and farm slurry microbiology.

I was a committee member of the SGM Environmental Microbiology Group (1996–2000) and am currently Convener of the SfAM Environment Group. I have organized the SGM main symposia on *The Behaviour of Pathogens in the Environment* (Warwick '99) and *Waste Treatment* (Southampton '97).

Hilary Lappin-Scott

I studied Environmental Sciences at Warwick University (commencing 1977), a multi-disciplinary degree that was years ahead of its time! After a very enjoyable three years I stayed on for my PhD at Warwick on the biodegradation of chlorophenoxyalkanoic herbicides, supervised by Howard Slater. I undertook a postdoctoral position at Calgary University where I was awarded a Fellowship to research the starvation survival of microbes in the subsurface and in biofilms. I was appointed a Lecturer at Exeter University in 1990 where I furthered my research interests in biofilm formation, structure and control. I was appointed to a Readership in 1995 and a Personal Chair in 1999.

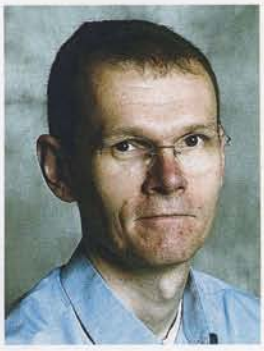
Tony Nash

My interest in infectious diseases and immunology was kindled at Queen Elizabeth College, University of London where I graduated in Biology/Biochemistry in 1971. An MSc in radiation biology at the University of Birmingham followed. This led to four excellent years in the Department of Experimental Pathology at Birmingham where I completed a PhD in Immunology, supervised by Philip Gell and the late Noel Ling, and also managed to hone my six-a-side football skills.

In 1978 I moved to Cambridge to work in Peter Wildy's group on the immunology of herpes simplex virus. This was where my interest in viral immunology and pathogenesis was firmly established. Appointed to a lectureship in Immunology, my research interests diversified to include Theiler's virus and the mechanisms of virus-induced demyelination. A stimulating sabbatical year in Michael Oldstone's laboratory at Scripps Research Institute introduced me to new molecular technologies for studying viral pathogenesis.

I moved to my present post as Professor of Veterinary Pathology at the University of Edinburgh in 1994. My current research interests are centred on understanding how gammaherpesviruses interact with their host. My six years in Edinburgh have witnessed major changes in virology research. The recent merger of Medical and Veterinary virologists has produced a major centre for research on the pathogenesis of persistent virus infections.

Outside the University my main interests are centred on my family and the fortunes of Leicester City Football Club.



Ian Roberts

Originally from Birmingham, I did my first degree in Biological Sciences at Leicester, where my interest in bacterial molecular genetics was aroused by the likes of Bill Brammar, Peter Williams and Brian Wilkins. There then followed a seemingly protracted period of adversity where I undertook a spectacularly unsuccessful PhD trying to clone germination genes from *Bacillus subtilis* in the laboratory of Derek Smith. I then returned to Leicester, first as a postdoctoral worker in the laboratory of Graham Boulnois and then two years later as a lecturer in the Department of Microbiology. Nine happy years followed surrounded by stimulating and supporting colleagues such as Peter Andrew and Dorothy Jones. During this time I became a Lister Fellow in 1994 and was awarded the Fleming Prize in 1995 from SGM and in the same year the W.H. Pierce Prize from SAB. Finally, I realized that there was more to science than the M69 between Birmingham and Leicester and in 1995 took the Chair in Microbiology at the University of Manchester. My research interests are primarily on understanding the synthesis and export of cell surface polysaccharides in Gram-negative bacteria and their role in disease.

SGM Membership Subscriptions 2001

The following rates were agreed at the AGM of the Society on 13 September 2000.

Ordinary Member	£	US$
Membership subscription (including *Microbiology Today*)	40.00	70.00
Additional concessionary subscriptions for publications:		
Microbiology	70.00	135.00
Journal of General Virology	70.00	135.00
Int J Syst Evol Microbiol	70.00	135.00

Student or Retired Member	£	US$
Membership subscription (including *Microbiology Today*)	20.00	33.00
Additional concessionary subscriptions for publications:		
Microbiology	35.00	65.00
Journal of General Virology	35.00	65.00
Int J Syst Evol Microbiol	70.00	135.00

Undergraduate Member (UK and Republic of Ireland)	£	US$
Membership subscription (including *Microbiology Today*)	10.00	NA
No concessionary subscriptions to journals are available to Undergraduate Members		

Members are reminded that their 2001 subscriptions are due for payment by **1 December 2000**.

As in previous years, no journal or meetings information will be despatched to members who are in arrears, and there will be no guarantee of provision of back numbers of journals for members who pay their subscription late.

Payment by direct debit or continuous credit card

Subscription notices were despatched recently to all members paying by direct debit or by continuous credit card arrangement. To continue your present status and journal requirements, no further action is necessary. However, if you pay by continuous credit card, you should check that the card number and expiry date on the subscription notice are correct. To change your membership status or journal requirements for 2001, or your credit card details, you should have amended your subscription notice and returned it to the membership office by **15 November 2000**. However, if you have missed this deadline, your amended notice will be accepted if it is submitted immediately.

Payment against invoice

Invoices were despatched recently to all members who pay by this method. If you did not receive one, please inform the Membership Office.

Subscriptions waived for unemployed members

As in previous years, subscriptions may be waived at the discretion of the Society for unemployed members under the age of 35 who are resident in the UK. If you are eligible and wish to benefit in this way in 2001, you should send a signed statement that you are currently unemployed to the Membership Office before **30 November 2000**. (Please note that no increase in journal requirements will be permitted.)

Income tax relief on membership subscriptions

Members who are liable for UK income tax are reminded that their annual subscriptions to the Society have been approved by the Inland Revenue as qualifying for income tax relief. Any member who would like further information or has difficulty in obtaining this relief should contact the Executive Secretary.

Going Public round-up

Popular poster competition

This year the Institute for Animal Health and BBSRC ran a competition in which primary and secondary pupils were required to design a poster to celebrate the work of Alexander Fleming in discovering antibiotics. The quality of the 900+ entries was high and the judging, assisted by SGM representative John Grainger, proved difficult. The final choice of winners was based on the accuracy of the information and an eye-catching presentation. The winning school in each category received £400 and the top competition entries were displayed in the BBSRC pavilion at the Royal Show.

SGM hits the road

External Relations Office staff have been busy this year. Events presented or attended include:

- Edinburgh International Science Festival – public symposium on vaccination and children's workshops
- SGM Warwick University meeting – public debate *Infectious disease – will we ever win?*
- Springboard Careers Day at Wembley Conference Centre
- Directions Careers Fair at Earl's Court
- Association for Science Education Area Meetings – microbiology workshops
- British Association Creating Sparks – BAYS Day microbiology workshops

Want to spread the word about science?

Science Line, the service which responds to telephone questions about science from the general public, is looking for more experts to provide the answers. When the folk who man the lines are stumped, they have to seek help. This merely involves emailing or calling the appropriate expert and asking them for information. The answers are passed on to callers without revealing the sources – contact details of experts on the Science Line database are confidential and never given to callers or the media. About one-third of the questions are about astronomy or physics, one-third biological and one-third all the rest.

If you would like to help, fill in the expert form on the website at www.sciencenet.org.uk, or send an email to scienceline@bss.org or telephone 020 8735 5015.

A Wellcome prize

The Wellcome Trust invites professional life scientists who have had no popular science books published to date to enter their latest competition. £25,000 is up for grabs towards the cost of writing a popular science book. The aim is to write an important or influential book that will not only inspire, stimulate and inform the general lay reader, but will also open up new ways of thinking about the world and set the agenda for future debate and discussion. The closing date for submissions is **2 March 2001**. Further information, the rules and guidelines can be found at www.wellcome.ac.uk

Bugs, microbes and micro-organisms

Reg England

A Microbiology Summer School for 11–16-year-old pupils from local secondary schools was held between 24 and 27 July 2000 at the University of Central Lancashire in Preston. The workshop was run by Reg England, Chris Hughes, Gill Ward and Jon Rand. The hands-on programme introduced pupils to 'the good-guys' – microbes used in food production; 'the bad guys' – microbes that cause a problem in food poisoning and health; and 'the downright ugly' – microbes found on our bodies!

At the start of the first practical exercise, pupils were presented with a menu and invited to taste various 'microbial' foods and drink (bread, Quorn™ sausages, sauerkraut, yoghurt and cheeses, all washed down with lemonade). The pupils were then allowed to look at pre-prepared plate cultures and viewed slides of the relevant microbe(s) involved. The fungal plates, in particular *Fusarium graminearum*, *Penicillium roqueforti* and *Aspergillus niger* were met with shrieks of disgust at what they had just consumed. The pupils drew each microbe (plate and slide culture) in a specially designed workbook and a prize was awarded for the best drawings. Mid-way through the first session we were joined by BBC Radio Lancashire. The reporter interviewed a few pupils, and listening to the article on the radio the following day, we are pleased to report that microbiology received a very good press from the kids.

Pupils were then introduced to microbes about their body by taking swabs from various parts of their anatomy! They streaked the swabs onto different agar media, sealed the plates with parafilm and came back the next day to see what had been caught. A similar exercise was held by asking them to place their fingers, before and after washing with soap, onto nutrient agar plates.

The following day, pupils looked at the sealed plates and were able to identify whether they had grown bacteria, yeasts or fungi by referring to a simple key in their workbook. The kids were very surprised to see the diversity and numbers of bacteria that grew on their skin. Pupils drew chosen examples of their 'body bugs' in their workbook. Each agar plate from the finger exercise was photographed using a digital camera and a colour printout was given to pupils to include in their workbook before they went home (something to show Mum and Dad).

Safe practice

This work was carried out by trained microbiologists in a university laboratory. Anyone considering organizing microbiological investigations for school students should carry out a risk assessment first. It should also be noted that in secondary schools pupils aged 11–16 may carry out L2 work (see *Safety in Science Education*, HMSO, 1996) but the document states that '*organisms may be cultured from the environment but not from environments which are likely to contain harmful organisms, for example lavatory seats or body surfaces other than fingers or hands*'. It is also recommended that culture plates are not incubated above room temperature.

The SGM External Relations Office is always pleased to give advice and has produced a factsheet on safety and GLP in schools. Email education@sgm.ac.uk or see the website www.sgm.ac.uk

The pupils then carried out Gram stains on selected 'friendly body bugs' taken from their swab plates and/or finger plates. Whoops of delight/horror were heard when they looked at the stained bugs under × 400 magnification. A few selected examples of their 'body bugs' were shown on the TV screens, linked to a Nikon microscope via a video set-up. The pupils could see each other's bugs! The hands-on element finished with the pupils looking at prepared slides of a few 'bad guys' (*Escherichia coli*, *Staphylococcus aureus*, *Streptococcus pyogenes* and *Streptococcus mutans*) down a microscope.

The workshop closed by having a few minutes chat about what they had seen and learnt in each workshop exercise, followed by prize-giving. In addition, all pupils went away with a set of SGM posters about *Microbes in Food* and several informative leaflets (many thanks to Janet for providing them at such short notice). The general opinion was that everyone (us included) had a fun time.

More photos can be seen on our microbiology website www.uclan.ac.uk/micro

Reg England is former Convener of the SGM Fermentation & Bioprocessing Group. He is based at the Department of Biological Sciences, University of Central Lancashire, Corporation Street, Preston PR1 2HE. Tel. 01772 893513; email r.england@uclan.ac.uk

ABOVE:
Pupils Gram staining their 'body bugs' (upper). Gill and Jon looking on, and looking at slide and plate cultures of 'microbial foods' (lower).
COURTESY REG ENGLAND

Communicating microbiology: The Promega Prize competition

University of Exeter, 13 September 2000

Competition was fierce during the final round of this year's Promega Prize Competition. Eight enthusiastic young members each gave a 10-minute talk followed by 5 minutes of questions. All entrants gave fluent presentations using excellent visual aids and dealt with some tough questions. Afterwards, Tracey Duncombe met up with the contestants to find out more about the importance of their research and why they had entered the competition.

Karen Mattick, PHL, University of Exeter

Filament formation by Salmonella spp. under adverse conditions

Karen let it slip that it was her supervisor who had actually entered her for the competition. '*I was giving a talk at an SGM meeting anyway, so really I didn't mind, I had nothing to lose.*' Karen is a second year PhD student investigating the on-going growth of *Salmonella* under stress conditions where growth was not previously thought possible. '*This research has public health implications as well as giving us an insight into the stress response for the organism.*'

Suzanne Strauss, PHLS, Addenbrooke's Hospital, Cambridge

A single-tube real-time nested PCR for detecting the genomes of human papillomaviruses

Suzanne was employed as a clinical scientist to perform epidemiological studies at the HPV reference library until she started her PhD this year, which is focusing on carcinogenic HPV strains. '*I'm developing type-specific, quantitative PCR to see what effect virus load has on disease progression and clearance. I didn't really know anything about the competition. I'd produced a poster for a meeting in Paris and decided to display it at an SGM meeting. The judges asked me some questions about my work and that was how I made the pre-selection group finalists.*'

Ashraful Haque, Imperial College, London

Intracellular activation of the spv and dps promoters in Salmonella typhimurium

(Unavailable for interview.)

Gina Manning, Veterinary Laboratories Agency, Weybridge

Evidence for a genetically stable clone of Campylobacter jejuni

Gina is a bit of an old hand when it comes to the Promega Prize Competition. She was one of last year's winners after giving a presentation on a different aspect of her work on *C. jejuni*. '*I presented a poster at the SGM meeting in Warwick and decided to enter again.*' After she gained her PhD from Leicester University in 1998 working on *Pneumococcus*, Gina moved to VLA to take up a permanent postdoctoral research position studying *Campylobacter*. '*C. jejuni is an important human pathogen both socially and economically. It's important to understand the epidemiology of the disease so that we can put control measures in place to control the pathogen in the food chain.*'

Amanda Smith, Institute for Animal Health, Compton

The identification of genes required for growth of Streptococcus uberis in milk

Amanda is in an auspicious position; she is a third year PhD student, has almost completed writing her thesis and has a postdoctoral position lined up to work on *Escherichia coli* O157. '*I'm looking forward to my new post, as I've always wanted to do human-related research.*' Amanda saw a poster for the competition at a previous SGM meeting and decided to present the results of her work. '*We hope to find a vaccine against S. uberis by identifying genes involved in important pathways. S. uberis is a major cause of bovine mastitis. Affected milk is not marketable and therefore has huge economic implications.*'

Martha Simpson-Holley, University of Cambridge

A functional link between the actin cytoskeleton and lipid raft domains during influenza virus budding

After giving a talk at the Warwick meeting, Martha was automatically entered for the contest. '*I've really enjoyed the experience, it's been good for building confidence.*' Martha is going into the third year of her PhD looking at the morphology of the influenza virus that is associated with clinical disease: '*this may be the form of the virus that grows in your nose when you have flu*'.

Chris Smith, University of Cambridge

HSV-1 latency in the central nervous system

Chris was one of two students selected by the Virology Group judges at the Warwick meeting. '*90% of the population are latently infected with HSV-1, but there are no drugs that target the latent virus; we can treat a cold sore but we can't stop it re-occurring. In order to develop novel anti-viral therapeutic strategies we need to gather knowledge of the virus during latency. Research in this area may also, in the future, lead to a vector for gene therapy. Latent HSV-1 does not affect neurones and is capable of long-term gene expression.*' Having finished writing his PhD thesis within 3 years, Chris, who is also a medic, is looking forward to the day in 9 months time when he'll become a 'doctor, doctor'. '*I'd love to carry on in research as well as staying in medicine.*'

Justine Fitzmaurice, National University of Ireland, Galway

Detection of verocytotoxigenic Escherichia coli using a PCR/DNA probe membrane based colorimetric detection assay

Justine started her PhD last October and has made a flying start in the communication of her research. Justine qualified for the Promega Competition after giving a talk at the Irish Branch meeting in Galway earlier this year. '*Food poisoning caused by O157 is a serious disease, which is increasing in Ireland. However, tests are not carried out routinely and it may take up to 6 days for a positive result. We have developed new probes as part of a PCR-based technique to speed up the detection of the O157 serotype to within 48 hours.*'

● Tracey Duncombe, SGM Public Affairs Administrator, is to be the new correspondent for Gradline. Please let her have your stories or news for possible publication. Articles on any topic of interest to younger scientists are welcome (email pa@sgm.ac.uk).

The names of the winners were announced at the Society Dinner that evening. Both the winners showed enthusiasm for their research work and led the audience into the background and methodology before embarking on the detailed implications of their results. The winning talks were of a very high standard that more senior microbiologists might hope to aspire to! After careful deliberation, the panel of judges, chaired by Pat Goodwin, decided to award both **Gina Manning** and **Chris Smith** with the Promega cheques for £200. The other finalists will all receive £25 from SGM and a year's free membership of the SGM. Gina and Chris will now go through to represent the society in the Promega *Young Life Scientist of the Year Award*, which will be held next year. There they will be competing against other Promega Prize winners from the Biochemical Society, the Genetical Society and the British Society for Immunology for the chance to win a trophy and £2,000 prize money.

If you are a postgrad or in the first two years of a postdoc, then it couldn't be easier to enter next year's competition. Just present a poster or an oral offered paper at a Society meeting and let the Meetings Office know that you wish to be considered. Further details of the Promega Prize competition are available on the SGM website www.sgm.ac.uk

● *Tracey Duncombe, SGM Public Affairs Administrator*

Student Membership

Student Membership of the Society is available to postgraduate students worldwide who have no taxable income. For an annual subscription of only UK£20 (US$33) Student Members can take advantage of the many benefits that this category of membership provides, such as free registration at SGM meetings and the purchase of Society publications at greatly discounted prices. In addition Student Members who are resident and registered for a higher degree in any European Union country may apply for awards from the President's Fund and Postgraduate Conference grants (see p. 192 for details) which provide financial assistance for attendance at scientific meetings.

Undergraduate Membership

Undergraduate Membership is open to students resident and registered for a first degree in the UK and Republic of Ireland. For the bargain subscription of only £10 Undergraduate Members will receive *Microbiology Today* and may attend Society meetings without payment of a registration fee. Careers events will also be held for them at different venues around the country. However Undergraduate Members will not be eligible for travel or conference grants. Information about this category of membership is being circulated to all relevant UK university departments.

Student Societies

SGM Sponsored Lecture Scheme

Grants are available to support TWO lectures on microbiological topics per academic year at Student Society meetings.

A Student Society is eligible for support if:

- It is run mainly by and for students of life sciences, either postgraduates and/or undergraduates.
- It is based in the UK or Republic of Ireland

The invited speakers will be reimbursed directly for reasonable costs of travel and accommodation. However, please note:

- The maximum claim for each lecture is £150.
- One speaker may be invited from abroad or from Ireland, but there can be no increase in the maximum claim for the lecture.
- IThe Society will be reimbursed for the costs of entertaining the speaker to dinner, including the expenses of ONE member of the committee.

Application forms are available from The Grants Office at SGM HQ (Tel. 0118 988 1821; Fax 0118 988 5656; email: grants@sgm.ac.uk).

Webwatch

Meetings

Meetings on the web

Up-to-date information on future Society meetings is available on the website: **http://www.sgm.ac.uk**

Meetings organization

The SGM meetings programmes are organized by the committees of the special interest groups, co-ordinated by the Scientific Meetings Officer, Professor Howard Jenkinson. Suggestions for topics for future symposia are always welcome. See p. 212 for contact details of Group Conveners. Administration of meetings is carried out by Mrs Josiane Dunn of the Meetings Office at SGM Headquarters, Marlborough House, Basingstoke Road, Spencers Wood, Reading RG7 1AG (Tel. 0118 988 1805; Fax 0118 988 5656; email meetings@sgm.ac.uk).

Millennium meeting

University of Warwick April 2000

Fighting Infection in the 21st Century

Symposium volume

This is available from Blackwell Science. The price is £65, with a 40% discount for members of SGM and SfAM. See p. 207 for a review of the book and details of how to order a copy.

Autumn 2000

147th Ordinary Meeting
University of Exeter
12–15 September 2000

Community Structure and Co-operation in Biofilms

Symposium Volume 59 is available from CUP. An order form for members is enclosed with this issue of *Microbiology Today*. A review of the book appears on p. 207.

Abstracts Book

The full text of the abstracts book covering other sessions at this meeting is now available as a PDF file on the SGM website.

Offered posters

Offered posters are welcome but each one should be associated with a Group. General Offered Posters will no longer be accepted. Titles and abstracts should be sent to the appropriate Convener, preferably by email. The subject content should be relevant to the remit of the Group (see website for details); it does not have to relate to the topic of the Group Symposium taking place at the particular meeting. Abstracts are required in a standard format – see website for details or contact the Events Administrator.

Future Meetings

SPRING 2001 – 148th Ordinary Meeting

Heriot-Watt University, Edinburgh 26–30 March 2001

● Main Symposium New Challenges to Health: the Threat of Virus Infection

Organizers: P.M. Goodwin, W.L. Irving, J. McCauley, D.J. Rowlands and G.L. Smith

Speakers:
C.J. PETERS (Atlanta) *Surveillance and detection of viruses*
B. GRENFELL (Cambridge) *Overview of the epidemiological impact of viruses*
R. ELLIOTT (Glasgow) *Hantavirus/bunyavirus*
B. RICHARDSON (Sydney) *Calicivirus, myxoma virus and the wild rabbit in Australia: a tale of invasions*
A. HAY (NIMR) *Influenza virus*
S. LEMON (Texas) *Hepatitis virus*
R. WEISS (London) *HIV*
T. BARRETT (Pirbright) *Morbilliviruses – dangers old and new*
C. WEISSMANN (London) *TSEs*
J.P. STOYE (NIMR) *Endogenous retrovirus/xenotransplantation*
C. BOSHOFF (London) *Gammaherpesviral infections in immunocompromised populations*
H.-D. KLENK (Marburg) The proteins of *Ebola and Marburg viruses – functions and potential roles in pathogenesis*
D. GUBLER (CDC, USA) *Dengue virus*
H. LUDWIG (Berlin) *Borna viruses*
G. DARBY (Stevenage) *Antiviral drug development and the impact of drug resistance*

● A leaflet about the meeting is enclosed with this issue. A poster to publicize the meeting is also available from the Meetings Office if you would like to display a copy on your departmental noticeboard.

Launch of new Clinical Microbiology Group

The new Clinical Microbiology Group will be launched with the following symposium:

28 March
Antibiotic resistance

SPEAKER TBC *Evolution and microbial resistance*
P. HAWKEY (Leeds) *Flow of antibiotic resistance genes in the environment*
A. VAN BELKUM (Utrecht) *Molecular epidemiology of antibiotic resistance: methods for study*
H. RINDER (Munich) *Hetero resistance: resistant bacterial sub-populations*
F.-J. SCHMITZ (Düsseldorf) *Molecular diagnosis of resistance*
K. FORBES *Studying resistance using DNA arrays*
M. GILL (Birmingham) *Novel mechanisms of resistance in Gram-positive pathogens*

29 March
a.m. Workshop on *Funding & Preparing Research*
p.m Offered Papers. Offered posters will also be welcome.

Contact the Group Convener, Stephen Gillespie (stepheng@rfc.ucl.ac.uk) for further details.

● Other Symposia and Workshops

● Wall-less organisms

Cells & Cell Surfaces Group

27 March
Organizers: I.C. Sutcliffe (iain.sutcliffe@sunderland.ac.uk) and M.J. Woodward (mwoodward@cvl.wood.gtnet.gov.uk)

● Monitoring and treatment of blood-borne viruses

Clinical Virology Group

28 March. There will also be an Offered Papers session on 29 March.
Organizers: Jeff Connell (jeff.connell@ucd.ie) and Conall McCaughey (cmccaughey@qub.ac.uk)

● Benchmarking in microbiology education

Education Group

29 March
Organizer: Trevor Cartledge (trevor.cartledge@ntu.ac.uk)

● Microbe/pollutant interactions: biodegradation and bioremediation

Environmental Microbiology Group

29–30 March
Organizer: Kirk Semple (k.semple@lancaster.ac.uk)

● Biotransformations

Fermentation & Bioprocessing and Physiology, Biochemistry & Molecular Genetics Groups

27–28 March
This session will deal with whole-cell, microbial biotransformations and will be multidisciplinary, involving biologists, chemists and biochemical engineers. It will focus on reactions currently under-represented in industrial processes.
Organizers: Neil Bruce (n.bruce@biotech.cam.uk) and Gill Stephens (g.m.stephens@umist.ac.uk)

● New enzyme targets for anti-microbials

Microbial Infection Group with Biochemical Society

26 March
a.m. *Use of structural data to characterize novel targets*
p.m. *New approaches to target identification*
Organizer: L.J.V. Piddock. (l.j.v.piddock@bham.ac.uk)

● Microbiology of nitric oxide

Physiology, Biochemistry and Molecular Genetics Group

29 March – 2 sessions:
Nitric oxide in free-living bacteria
Nitric oxide in the host–pathogen interaction
Organizer: M. Larkin (m.larkin@qub.ac.uk)

- Post-transcriptional control of gene expression

Virus Group

28–30 March
There will also be evening workshops on 26 and 29 March.
Organizer: I. Brierley (ib103@mole.bio.cam.ac.uk)

- Special symposium: Genomics: beyond the sequence

Systematics & Evolution Group with the International Committee on Systematic Bacteriology

26–27 March
Contact G. Saddler (g.saddler@cabi.org) for details

- Evening workshop for young members: Genomics

Physiology, Biochemistry and Molecular Genetics Group

Organizer: M. Larkin (m.larkin@qub.ac.uk)

Offered posters are welcome for all sessions except those of the Virus Group. Please submit titles and abstracts to the appropriate symposium organizer or Group Convener by the deadline of **17 November 2000**.

AUTUMN 2001 – 149th Ordinary Meeting

University of East Anglia
11–13 September

● Main Symposium Mycobacteria: New Developments

Organizers: M. Goodfellow, P.M. Goodwin, H.M. Lappin-Scott, G. Saddler and D. Smith

● Other Symposia

- Microbial lifestyles

Cells & Cell Surfaces Group

Organizers: J. Armitage & P. Rainey

- Lower respiratory tract infections

Clinical Microbiology Group

Organizer: S. Gillespie

- Research supervision – how to get it right

Education Group

Organizer: A. Eley
Plus supervisor training workshop: *Problem solving in supervisor-student disputes*

- Microbial interactions in aquatic environments

Environmental Microbiology Group with British Phycological Society

Organizer: G. Underwood (gjcu@essex.ac.uk)

- Bioprocess monitoring and control

Fermentation & Bioprocessing Group

Organizer: D. Mead

- Mobile genetic elements in bacterial virulence

Microbial Infection Group

Organizers: M. Barer & P. Langford

- Classification and identification of clinically significant actinomycetes

Systematics & Evolution Group

Organizer: G. Saddler

Offered posters are welcome for all sessions. Please submit titles and abstracts to the appropriate symposium organizer or Group Convener by the deadline of **11 May 2001**.

Irish Branch

Postgraduate 2001

Waterford Institute of Technology
4–5 January 2001

Offered papers from postgraduates welcome.

Organizer: Catherine O'Reilly (coreilly@wit.ie)

Functional Genomics of Microbial Pathogens

Trinity College Dublin
22–23 March 2001

How will the vast amounts of information being generated on the genome sequences of microbial pathogens be used to advance our knowledge of the biology of these organisms and to devise new methods of control? Internationally known speakers in the area of functional genomics will address these questions by discussing the new research that has become possible using whole-genome approaches with prokaryotic and eukaryotic pathogens.

D. McDEVITT (SmithKline Beecham, USA) *Genomics: new strategies for small molecule drug discovery: opportunities and hurdles*
M. PALLEN (Queen's University of Belfast) *Data mining bacterial genome sequences*
P. DORR (Pfizer, UK) *3D structure analysis coupled with high-throughput screening*
R. RAPPUOLI (Chiron SpA Italy) *Reverse vaccinology*
P. RATHOD (Catholic University of America) *Global transcriptional changes in the human malaria parasite Plasmodium falciparum*
G. WEINSTOCK (Texas) *Genomic studies of spirochaetes*
B. WREN (London School of Hygiene & Tropical Medicine) *The full Campy: post-genome analysis of a food-borne pathogen*

Offered papers from Ireland and abroad on all aspects of microbiology will be accepted as posters and a small number of young researchers will be chosen for oral presentations.

Full details of this event and a registration form are on the SGM website.

Organizer: Angus Bell (abell@tcd.ie)

Deadline for abstracts and registration **31 January 2001**

Microbial Genome environment interactions

Queen's University of Belfast
Autumn 2001

Organizers: Martin Collins (m.collins@qub.ac.uk) and Mike Larkin (m.larkin@qub.ac.uk)

For details of Irish Branch activities contact the Convener, Martin Collins (m.collins@qub.ac.uk)

Other Events

● Joint ASM/SGM Meeting

2–6 October 2001
Caribe Hilton, San Juan, Puerto Rico

Biodegradation, Biotransformation and Biocatalysis

Further details of this meeting can be found on p. 206.

● BURSARIES

Grants will be available from the SGM for young members wishing to attend this meeting. Full details of the scheme will be published in the February 2001 issue of *Microbiology Today* and on the SGM website: **www.sgm.ac.uk**

● Viral Zoonoses

9–11 January 2002
Royal College of Physicians, London

SGM Clinical Virology Group, European Society for Clinical Virology and the European Society for Veterinary Virology

Organizers: T. Wreghitt (Fax 01223 242775) & J. Best (jenny.best@kcl.ac.uk)

● Fermentation & Bioprocessing Group

Offered paper/posters are welcome for the *Biotransformations* session at Heriot-Watt (27–28 March 2001) – deadline **November 17 2000** – and the symposium on *Bioprocessing, Monitoring & Control* at UEA (10–11 September 2001) – deadline **11 May 2001**. Please contact Glyn Hobbs (email g.hobbs@livjm.ac.uk).

Promoting Microbiology:

A new strategy, a new representative – Tracey Duncombe

Until recently Janet Hurst and the other members of the External Relations Office have dealt with media and PR issues, and have done so with notable results. But as a society we have not had a clear system in place for promoting the SGM and microbiology as a whole.

With my recent appointment as Public Affairs Administrator things are set to change. We are in the process of developing a strategy to raise the profile of SGM and microbiology to both parliament and the media. We aim to be the voice for microbiology and microbiologists in the UK, and to achieve this through effective communication.

One important new undertaking is the development of a series of briefing papers aimed at Members of Parliament, science policy makers and journalists. The first paper on *Biofilms* is in production. The intention of these papers is to heighten awareness of novel research as well as providing authoritative information on microbiological matters of topical concern.

We also intend to promote SGM journals by publicizing some of the papers on subjects that are of general interest and to issue media releases about sessions at Society meetings.

As some of you may be aware, the SGM has a new Clinical Microbiology Group. The inaugural symposium will be held at the meeting at Heriot-Watt University next spring. For the first time both the Institute of Biomedical Sciences (IBMS) and the Royal College of Pathologists will accredit our symposia. This will assist clinicians and MLSOs, who are members of both organizations, to advance in their Continuing Professional Development (CPD) programme.

For those of you who don't have time to read the broadsheets on a daily basis, *Microbiology in the News*, covering a broad range of topical issues, is back on our website. News summaries will be posted fortnightly at **www.sgm.ac.uk/pa/mic_news/micro.htm**

Finally, and most importantly, a request. I am building up a database of members who are willing to speak to the media on behalf of the SGM. Individuals who wish to participate should have a strong background in the area in which they are willing to talk, preferably they should have published papers or be currently working in that field. For members who are eminent in their field I also require contacts who are willing to give an opinion on consultation documents produced by government departments. Please contact me at Marlborough House if you are able to help.
Tel. 0118 988 1800;
Fax 0118 988 5656;
email: pa@sgm.ac.uk

A Personal Profile

My first degree was in microbial biotechnology at the University of Liverpool. I gained my PhD in the bioremediation of contaminated soil in November 1999 from the same university, during which time I took part in several initiatives to raise public awareness and promote science. I spoke two years running at the British Association (BA) Festival of Science in NERC's *Stand Up Science Show*; became a mentor for a group of 14-year-old girls during GETSET '99; stood as NERC's representative during a regional final of CREST Awards; and endeavoured to keep the attention of 100 school kids during Liverpool University's *Science on Saturdays* programme. Through participation in these activities I decided that it was time to lay down my Gilson once and for all and concentrate on promoting other peoples' work, and here I am.

Excellence and opportunity explained:

A summary of the recent DTI White Paper

Much has been made of the Department of Trade and Industry's White Paper *Excellence and Opportunity – a science and innovation strategy for the 21st century* since its publication on 26 July this year. The media, in particular, have focused on one part, namely the £4 million annual fund, which is being set up to recruit 'the David Beckhams of science'. In an attempt to create a national 'brain gain', 50 leading scientists, chosen by a panel of experts from the Royal Society, the Royal Academy of Engineers and the Wolfson Foundation research charity, would be lured to Britain with salaries of up to £100,000. At present, British universities pay professors a minimum salary of around £35,000 to £40,000. Whether or not this will halt the 'brain drain' is arguable. The feeling of many scientists quoted in *THES* (28/7/00) was that it did not address the fundamental problem that some of the brightest young scientists choose not to pursue careers in academic research because of the lack of prospects.

The scheme was announced as part of a package of measures in the White Paper. It sets out how the 7 % per annum average increase in the science budget over the next three years, announced in the Government's Comprehensive Spending Review, will be allocated to maintain the UK's world-class science. However, as departmental budgets have not yet been released, it may be too early to know whether the money announced for the science base represents new funding or largely a reshuffling of existing money.

The White Paper listed a number of ways in which the government was encouraging university employers, and funding and research councils, to develop career guidance and staff development for young postdocs on fixed-term contracts. It was also examining how it could do more to help women to progress in scientific careers and how it could increase the numbers of overseas students studying science and engineering in the UK. PhD stipends are to be increased in stages to reach £9,000 outside London by 2004. In an announcement made prior to the White Paper, this increase recognized the growing problem of attracting the best students to research, and went some way towards meeting the recommendations of the UKLSC Working Party.

The continued funding to improve facilities has been welcomed by all the leading science agencies. Notably, a further £1 billion investment in science infrastructure will be available between 2002 and

2004, funded by Government and the Wellcome Trust on top of the Joint Infrastructure Fund (JIF).

Familiar statistics were used to show that Britain's science base already competes very well, but the government is clearly worried about the future of science. It was argued that we need to maintain our world lead in the fields where we excel and develop a lead in new areas, while maintaining the capacity to do science that is recognizably world-class across the board. This was the rationale for providing an additional £250 million between 2002 and 2004 in the Spending Review for research in the areas of genomics, electronic science and informatics, and in basic technology such as nanotechnology, quantum computing and bioengineering.

The White Paper acknowledged that the government has a clear role in the funding of basic curiosity-driven research. The main thrust of the findings was to address how best to assist with the transfer of research from UK universities into commercial products and services in a way that has public support and involvement.

The Paper considered that although the Research Assessment Exercise (RAE) did not penalize inter-disciplinary collaboration, more support was needed. It welcomed the guidelines for the next RAE that basic and strategic research done in confidence for business should be given equal weight alongside papers published in peer-reviewed journals. The RAE will not be used to divert research funds to support universities' applied work. Instead, a larger, permanent, stream of funding will replace the present Reach Out funding stream. The Higher Education Innovation Fund will be worth £140 million over three years. Other schemes to encourage collaboration with industry include funds to strengthen regional science and to create networks, which are to be made available through regional development agencies. Although this funding is welcomed, the UKLSC pointed out in an earlier consultation that much collaboration is carried out on a national or international scale rather than at a regional level.

The Paper also considered that it was necessary to start at the base, with better education for all children in science. It wants to beef up school labs, persuade 'science ambassadors' to encourage children – particularly girls – to take science more seriously and launch a 'science year' in September 2001. A new science curriculum, with greater emphasis on practical science, was implemented in September this year. The new £10,000 training and recruitment package for teachers intends to attract more well-qualified candidates. Several other initiatives were listed that would support continuing professional development of science teachers. The government also announced that it intended to work with the providers of science materials to schools to provide a single point of support for science teachers, so that all were aware of the resources available.

In the introductory chapter the need for public confidence in the whole notion of science to be strong and well founded was emphasized. The government saw its role in assuring consumers that risks have been properly assessed and controlled and in communicating those risks clearly and simply and at the right time. In that respect, guidelines on the use of scientific advice by government departments and agencies are to be updated and a new code of practice encouraging openness for advisory committees is to be published for consultation. However, similar guidelines and advisory committees were in place, for example, in the last two years when the public mistrust over GM foods grew, and the government was forced to draw back from its initial approach. Little was said about encouraging scientists to engage more effectively with the public other than that the government would build on existing Research Council initiatives in training scientists to communicate their work.

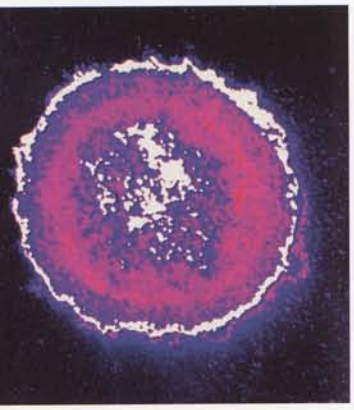
● *Tracey Duncombe, SGM Public Affairs Administrator (email pa@sgm.ac.uk)*

Acknowledgement

I would like to thank Mike Withnall, Secretary to the UK Life Sciences Committee, for permitting the use of his summary of the White Paper as the basis of this article.

Hot off the Press

Microbiology Today Editor Meriel Jones takes a look at some papers in current issues of the Society's journals which highlight new and exciting developments in microbiological research.



ABOVE:
False-colour image of HIV.
PHOTODISC

OPPOSITE PAGE TOP:
Cells lacking sphingosine-1-phosphate lyase have increased resistance to the anticancer drug cisplatin. Sphingosine-1-phosphate lyase null mutants (red line) created by homologous recombination are 25 times more resistant to cisplatin than the parental wild-type cells (yellow line). The sphingosine-1-phosphate lyase mutant also exhibits dramatically altered morphogenesis (right inset) compared to the parent (left inset).
COURTESY GUOCHUN LI, SUPRIYA SRINIVASAN, HANNAH ALEXANDER & STEPHEN ALEXANDER, UNIVERSITY OF MISSOURI, USA

Predicting the progress of AIDS

Among all the scientific work and personal tragedies of AIDS, many aspects are still not understood. One of these is how the infection can take different courses in different individuals. Although the dose and viral strains responsible for human HIV infections are highly variable, there are people known to have been infected through identical routes with the same strain. Even so, the progress of the disease has not been the same. This is even clearer in animal experiments, when there is no question but that all the animals were infected with identical amounts of virus. Simian immunodeficiency virus (SIV) causes a fatal AIDS-like disease in rhesus macaques and this has been used as a model for HIV and AIDS in humans.

Although the average time until death of a SIV-infected macaque is 1–2 years, some animals die within months while others live for many years. Experiments are carried out with small groups of animals, often four or less, and this natural variation may conceal any effect of a therapeutic trial. Researchers at the University of Pittsburgh School of Medicine, working with staff at the Tulane Regional Primate Research Center, have now reported the result of a decade of work that allows them to know in advance which animals will die rapidly after infection and which survive for a long time.

The key is the way the animal's peripheral blood mononuclear cells (PBMC) do, or do not, allow the virus to multiply. The researchers collected blood from uninfected animals, added virus to PBMC isolated from the blood and recorded how well the virus grew. The macaques were then infected with SIV and the researchers had to care for them as the disease progressed. After bringing together information on 59 macaques from vaccine and therapeutic trials, the scientists are confident that they can now predict which animals will live for a long time with a SIV infection and which ones will die rapidly. Animals with blood cells that are high producers of virus will progress to disease significantly more quickly than if their PBMC are low or intermediate producers of virus. This knowledge can be used in the design of future trials, and since it suggests that virus growth is controlled by innate characteristics of the individuals, it gives an additional factor for understanding, and combating, this lethal disease.

Seman, A. L., Pewen, W. F., Fresh, L. F., Martin, L. N. & Murphey-Corb, M. (2000). The replicative capacity of rhesus macaque peripheral blood mononuclear cells for simian immunodeficiency virus *in vitro* is predictive of the rate of progression to AIDS *in vivo*. *J Gen Virol* **81**, 2441–2449.

JGVDirect

On p. 151 of the August issue of *Microbiology Today*, an incomplete website address was printed for **JGVDirect**. The full address is:

http://www.sgm.ac.uk/JGVDirect

The SGM publishes two monthly journals, ***Microbiology*** and ***Journal of General Virology***.

The ***International Journal of Systematic and Evolutionary Microbiology*** (***IJSEM***, formerly ***IJSB***) is published bimonthly on behalf of the IUMS in conjunction with the ICSB.

The three journals are now available online.
For further information visit the journal website:
http://www.sgmjournals.org

Members may purchase SGM journals at concessionary rates. See p. 194 or contact the Membership Office for details. Information on commercial subscriptions is available from the Journals Sales Office.

An early test for cancer?

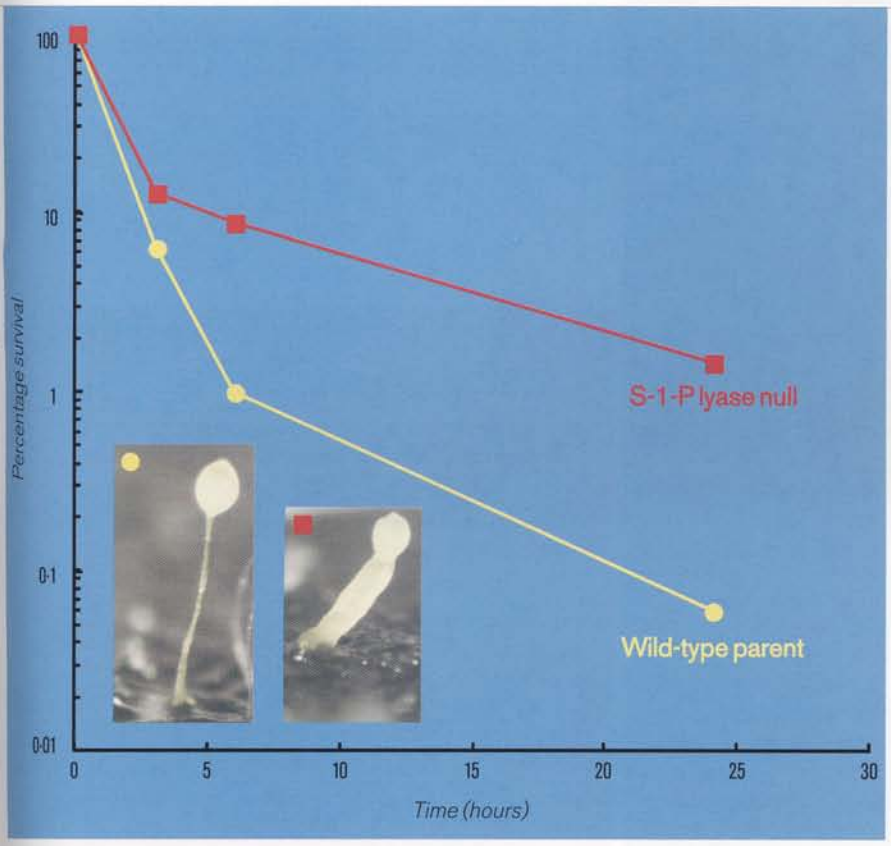
Epstein–Barr virus infects more than 90 % of the human population and has no apparent effect on the vast majority. The virus takes up lifelong harmless residence within one of the types of cell in the immune system, the resting memory B cells. The viral genes stay within the cells in an inactive condition. In a very small number of people, the virus unfortunately re-surfaces in an active state in cancerous tissues, and is considered a factor in the initiation of the disease. One of these is undifferentiated nasopharyngeal carcinoma (NPC). Scientists at the National University of Singapore have been studying the differences between patients with NPC and healthy individuals who nevertheless have antibodies to the virus in their blood.

About half of the people they examined had signs in their blood that the virus was active. The virus requires activity of a number of genes to replicate itself, and the researchers could detect activity of some, but not all of them. It was as if the virus occasionally tried to replicate itself, but was unable to complete the process. When they examined cells taken from the carcinoma itself, or the inflamed throats of some of their otherwise healthy volunteers, the situation was different. The cancer cells had signs of activity of all the viral genes required to start replication, while one gene, *BRLF1*, was inactive in the non-cancerous cells.

The body usually responds to the presence of new proteins by directing the immune system to remove them. A first step in this direction is the synthesis of proteins called antibodies that bind specifically to the new protein and mark it for destruction. When the researchers looked for antibodies to the product of the *BRLF1* gene in the blood of 53 patients with NPC, they could detect it in 83 % of them. In contrast, they could only find one positive reaction among the same number of their control volunteers.

Other scientists have suggested that the product of the *BRLF1* gene may be important in the re-activation of Epstein–Barr virus and the development of cancer. The results of this study indicate that, although this may well be true, it also betrays the presence of the active virus in the form of a unique antibody. The researchers hope that it will be possible to develop this into an early test for the disease.

Feng, P., Ren, E. C., Liu, D., Chan. S. H. & Hu, H. (2000). Expression of Epstein–Barr virus lytic gene *BRLF1* in nasopharyngeal carcinoma: potential use in diagnosis. *J Gen Virol* **81**, 2417–2423.



Resistance to anti-cancer drug cisplatin

One bizarre fact about cancer is that damage to DNA can be both its origin and cure. The chemical cisplatin, that joins together adjacent DNA bases, is a widely used treatment for cancer. Unfortunately, its effectiveness is often limited as the cancerous cells develop resistance to it. Scientists are keen to understand the exact nature of this resistance, so that they can improve cancer therapy.

Investigations of cisplatin-resistant tumour cells have spotted a large number of changes that might be the reason for their survival. It can, however, be very difficult to identify the exact molecular details. Researchers at the University of Missouri and the Max-Planck-Institut at Martinsried in Germany decided to take a different approach. They have been studying mutant cells from the slime mould *Dictyostelium discoideum* that can live despite being immersed in the cytotoxic chemical. The researchers say that the advantage of this micro-organism is that they can be confident that any resistant cells contain only a single mutation and this can be identified readily. The biochemistry and cell biology of slime moulds is very similar to that of animals, and they are multicellular for part of their life cycle. Thus, research on this organism is highly applicable to studies of human disease.

So far, the researchers have tested mutations in 10–15% of the genes in *Dictyostelium* and found seven that individually provide protection against cisplatin. Interestingly, this protection is specific to cisplatin since the *Dictyostelium* cells retained their normal sensitivity to other DNA-damaging chemicals. The function of some of these genes is currently unknown while others immediately suggest how they cause resistance. One of the latter is a mutation in the gene for sphingosine-1-phosphate lyase. This enzyme is required for the breakdown of sphingolipids, in a process that may signal the difference between death or continued life and proliferation in animal cells. The mutation certainly has a very dramatic effect on development in *Dictyostelium*. Instead of developing into an elegant oval of spores on a slender stalk, the mutants have a short, fat stalk and very few spores. Two other mutations, both in genes that might be involved in intracellular communication, also had dramatic effects on development.

The researchers point out that they are combining detailed molecular biology and cell biology in a way that promises further insights into the mechanism of cisplatin resistance. It may also suggest new drugs that could enhance the sensitivity of cancer cells to cisplatin.

Li, G., Alexander, H., Schneider, N. & Alexander, S. (2000). Molecular basis for resistance to the anticancer drug cisplatin in *Dictyostelium*. *Microbiology* 146, 2219–2227.

Any old iron?

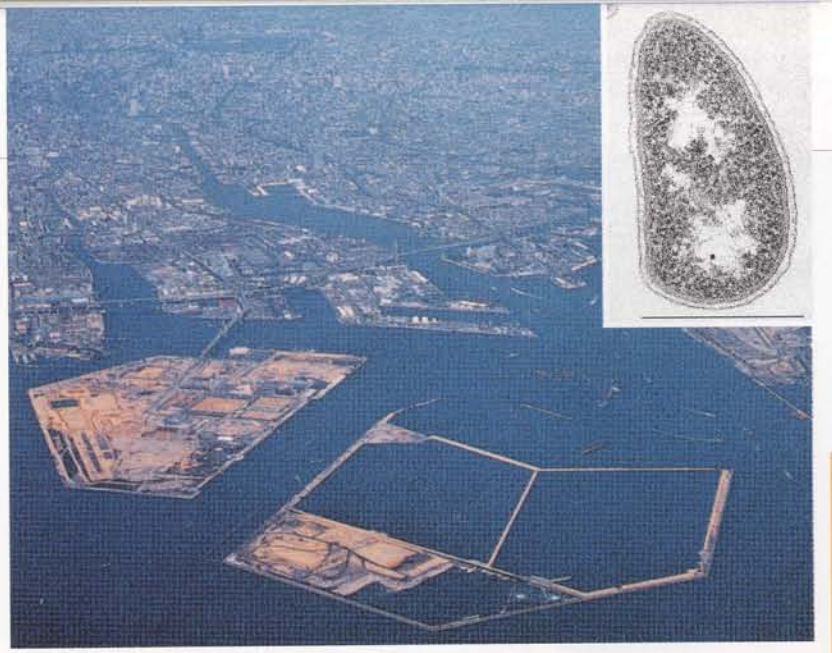
A small amount of iron is an indispensable component of all living cells. However, the very chemistry that makes it essential inside a cell also makes it virtually insoluble in the natural environment. There is around 10 million times less soluble iron in the soil environment than is required to sustain microbial life. The bacteria themselves have solved this conundrum. They synthesize and secrete chemicals, called siderophores, to dissolve and then transport the precious metal back into the cell. As a consequence, several scientists have suggested that bacteria may compete for this essential resource, perhaps through production of ever more efficient siderophores.

Dominique Joyner and Steven Lindow at the University of California in Berkley, USA, have been studying the behaviour of *Pseudomonas syringae*, commonly found on the leaves of plants. To find out if microbes really compete for iron, they wanted a way to measure its availability on the scale experienced by bacteria. To do this they joined the gene for a fluorescent green protein to a small part of one from *Pseudomonas*. This region regulated production of an enzyme required in the biosynthesis of siderophores to ensure that the cell only made them when the environment was low in iron. By putting the two together within the *Pseudomonas* cell, the researchers had a system that should have reflected the external level of iron through the amount of green fluorescence of the cells.

To check that it really worked, cultures of the bacteria were grown in liquid containing known amounts of iron and then their fluorescence was measured. This was inversely proportional to the amount of iron, giving the researchers confidence to move on to measurements of individual bacteria. They used a microscope linked to a computer to assess fluorescence from individual cells and again found a clear relationship between increasing fluorescence and decreasing iron.

When they finally looked at the surface of broad-bean leaves that had been dipped into cultures of the bacteria, things, of course, were more complicated. The leaves already had some bacteria on them, and so the researchers had to pick out the iron-sensing strain from amongst them. About 10% of these cells had a substantially higher fluorescence than the rest, which implied that some cells sensed much less iron than the majority. Thus, from a bacterium's point of view, although there was adequate iron on the leaf surface for most of them, there were a few cells suffering from a shortage. As the researchers learn more about the very small scale of the microbial environment they hope to eventually understand how such characteristics affect microbial behaviour.

Joyner, D. C. & Lindow, S. E. (2000). Heterogeneity of iron bioavailability on plants assessed with a whole-cell GFP-based bacterial biosensor. *Microbiology* 146, 2435–2445.



Waste not, want not

Waste treatment and disposal sites are good places to discover new species of microbes. They are complex, new habitats, which often contain unusual organic materials and are simply awaiting inhabitants. In addition, there is considerable scientific, commercial and regulatory interest in ensuring that rubbish decays in as harmless a manner as possible. Three recent papers in IJSEM describe new species that have been unearthed in such unsavoury surroundings.

One of the products of microbial life, especially in the oxygen-free depths of a waste site, is methane. Special pipework can be installed to channel this safely away. It is a characteristic waste product of microbes known as methanogens, which are members of the bacterial domain called the *Archaea*. Japanese scientists, studying a disposal site on an artificial island in the sea near Osaka, realized that methane was being produced despite the amount of toxic heavy metals in the waste. Very few of the known methanogens can tolerate heavy metals, so this was a good opportunity to look for them.

The researchers designed conditions that should have been ideal for these unusual organisms, added a small amount of liquid that had leached from the site and watched for growth. It turned out that getting something to grow was fairly easy, but obtaining a pure culture of it was difficult. After many attempts, they succeeded in isolating a roughly spherical organism that produced methane and grew best at 35 °C. Its growth was slowed, but not stopped, by heavy metals. To decide exactly what it was, the researchers compared the sequence of one of its genes against a database of thousands of sequences of the same gene from other bacteria. The closest match was to a member of the genus *Methanocalculus*, but it was sufficiently different to be assigned to a new species, *Methanocalculus pumilus*.

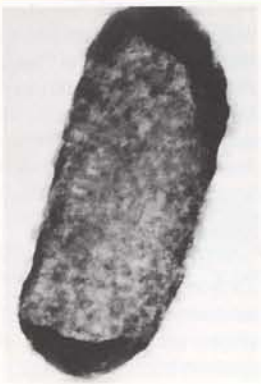
A wastewater treatment plant in Korea has turned out to be the home of a new species of *Janibacter*. This genus, within the family *Intrasporangiaceae*, has been represented by a single species until now. Scientists at the Korea Research Institute of Bioscience and Biotechnology and Sungkyunkwan University have been studying the microbial life in the soil and sludge from this treatment plant, which has to cope with toxic aromatic chemicals. The unusual characteristics of one strain, CS12^T, caught their eyes. The components of its round cells were very similar to *J. limosus*, but sufficiently different to make the researchers examine how well its DNA matched with authentic *J. limosus* DNA. The poorness of the match, along with all the other information, clinched the identity of a new species, which they called *J. terrae*.

A further species of *Janibacter*, *J. brevis*, with the useful ability to degrade trichloroethylene (TCE), a solvent used in dry cleaning and a number of industrial processes, has been reported by Japanese scientists. They isolated it from a sample of groundwater contaminated by leaks of the solvent by providing TCE as the sole source of nourishment. The glistening white bacterial colonies were made up of spherical cells and when the researchers investigated their chemical characteristics, the nearest match was to *Janibacter*.

Mori, K., Yamamoto, H., Kamagata, Y., Hatsu, M. & Takamizawa, K. (2000). *Methanocalculus pumilus* sp. nov., a heavy-metal-tolerant methanogen isolated from a waste-disposal site. *Int J Syst Evol Microbiol* **50**, 1723–1729.

Yoon, J.-H., Lee, K.-C., Kang, S.-S., Kho, Y. H., Kang, K. H. & Park, Y.-H. (2000). *Janibacter terrae* sp. nov., a bacterium isolated from soil around a wastewater treatment plant. *Int J Syst Evol Microbiol* **50**, 1821–1827.

Imamura, Y., Ikeda, M., Yoshida,, S. & Kuraishi, H. (2000). *Janibacter brevis* sp. nov., a new trichloroethylene-degrading bacterium isolated from polluted environments. *Int J Syst Evol Microbiol* **50**, 1899–1903.



TOP LEFT:
An aerial view of downtown Osaka and the Port of Osaka. The two artificial islands in the foreground are the Osaka North Port Sea-Based Solid-Waste-Disposal Site. The pentagonal island is the North Section and the the heptagonal island is the South Section. The land produced by waste-filling can clearly be seen. The organism was isolated from the land produced in the South Section. Inset: Ultrathin section of *Methanocalculus pumilus* MHT-1^T. Bar, 0·5 µm.
COURTESY DR K. TAKAMIZAWA, GIFU UNIVERSITY, JAPAN

ABOVE:
Citrobacter cells loaded with uranium. The left panel shows the cell fairly dark with deposited uranium, but some detail can still be seen, in particular fibrillar material at the cell surface. All this is obscured by the heavy uranium deposit on the cell in the right panel. This cell is loaded with more than its own weight of uranium. The cell appears black and encrusted, but a preciptate of uranyl phopshate can be seen extruding from the cell. Cells in both panels are 1–2 µm in length.
COURTESY LYNNE MACASKIE, UNIVERSITY OF BIRMINGHAM

Full metal jacket

Microbes are the great recyclers of the planet. Much of their work in the decay and dissolution of once-living material goes un-noticed, or is even unwanted. However, their abilities are becoming increasingly appreciated and exploited. Heavy metals are a very difficult type of waste material. Many are toxic and cannot be destroyed but are best concentrated so that the metal can be either re-used or put carefully out of the way. Some micro-organisms are remarkably efficient at crystallizing metals around their cells. This may be for their own protection, but can form part of strategies to clean up metal-contaminated soils.

Researchers at the University of Birmingham, UK, have now reported their most recent study of the way the bacterium *Citrobacter* deals with the heavy metal uranium. This bacterium becomes coated with uranyl phosphate if it is suspended in a solution containing uranium. The crystals on the cell are not simply due to the heavy metal sticking to all available surfaces. The researchers have been gradually building up a picture of how the bacterial cells control the deposition of the toxic metal, allowing them to live despite being covered in several times their own weight of uranium. Through collaboration with the Research and Technology section of BNFL at Preston, UK, they were able to look at the cells using atomic force microscopy, with minimal disturbance to their natural form. This showed the metallic coating particularly well. This ability to accumulate relatively large amounts of the metal is the attraction as part of a system for filtering the pollutant from water.

An enzyme called phosphatase secreted by the cells is an essential component but perhaps surprisingly, it is inhibited by uranyl ions. From detailed measurements of the exact elemental and structural composition of the deposits, the researchers think that the first protective step is association of the uranyl ions with the comparatively few phosphate groups of the lipopolysaccharide coat that always covers *Citrobacter* cells. This traps the toxic metal. The next step involves the phosphatase enzyme which the researchers think is itself associated with the lipopolysaccharide. It releases phosphate ions, which drift away to capture further uranyl ions, imprisoning them away from the vulnerable enzyme and cytoplasm of the cell. Simple chemistry converts the initial complexes into a meshwork of more insoluble sodium uranyl phosphate crystals around the cell.

One particularly intriguing aspect of this research is the level of organization it implies at the cell surface. It is another indication that the enzymes secreted by bacterial cells can be involved in carefully controlled activities, despite being on the furthest fringe of what has traditionally been thought of as a living cell.

Macaskie, L. E., Bonthrone, K. M., Yong, P. & Goddard, D. T. (2000). Enzymically mediated bioprecipitation of uranium by a *Citrobacter* sp.: a concerted role for exocellular lipopolysaccharide and associated phosphatase in biomineral formation. *Microbiology* 146, 1855–1867.

Velvet evolution

Aidan Parte

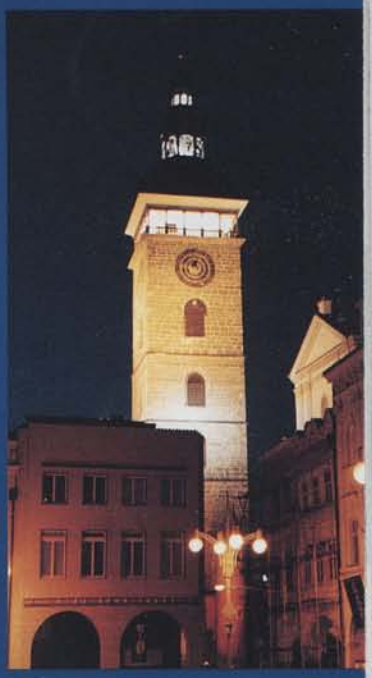
The International Society for Evolutionary Protistology (ISEP) had its 13th biennial meeting in the Czech Republic at the end of July and beginning of August 2000. ISEP serves people with interests in the taxonomy, phylogeny and evolution of protists – eukaryotes that cannot be placed among the green plants, multicellular animals or mycelial, non-zoosporic fungi.

Hosted by the Czech Academy of Sciences and organized at the local level by Julius Lukeš, the meeting saw more than 100 protistologists from all over the world descend on the ancient town of České Budějovice (Budweis in German), home of the *real* Budweiser beer, in South Bohemia. In 4 days of intense presentations, new theories on early eukaryotic evolution were expounded and accepted ones were lambasted – this was very entertaining at times. Many beautiful micrographs were shown, demonstrating the incredible diversity of protists and the skill and patience of the microscopists who study them.

What has this ISEP meeting got to do with the SGM? The IJSEM has undertaken to publish a number of papers – some invited symposium lectures and other original research papers; we hope to bundle these in a single issue to be published in 2001. Part of the reason for the change of name of the IJSB to IJSEM was to facilitate the journal's expansion into the field of protists, a natural and entirely logical change of scope. Thus, my attendance at the meeting, and publication of papers from it, was intended to give the IJSEM a profile in the evolutionary protistology market.

There was an active social programme at the meeting, too, including one for accompanying persons. There was a reception at the Academy (with local specialities such as carp), an evening in the Masné Krámy pub and an afternoon in the beautiful town of Český Krumlov, followed by a banquet in the magnificent castle gardens. All in all, I found the meeting very interesting and I hope that I learned something about a field which is largely new to me. The hospitality and efforts of Julius and his colleagues at the Academy was really appreciated.

● *Aidan Parte, Managing Editor, IJSEM*



ABOVE:
The 70 m Black Tower in Ceske Budějovice.

BELOW:
The town square in Ceske Budějovice, one of the largest squares in Europe.

PHOTOS AIDAN PARTE

MicroShorts

Joint ASM/SGM meeting

ABOVE:
Sea front gardens, San Juan, Puerto Rico.
COURTESY TRAVEL INK/ROY WESTLAKE

Biodegradation, Biotransformation and Biocatalysis

2–6 October 2001

Caribe Hilton, San Juan, Puerto Rico

Organizers: Gary Sayler, Jim Tiedje, David Gibson, Gary Toranzos and Hilary Lappin-Scott

Following on from the successful first joint meeting of the American Society for Microbiology and the SGM at the University of Aberdeen in 1995, the two societies are pleased to announce the second joint meeting. It will take place at the beautiful location of the Caribe Hotel, which is on the beach within walking distance of the old town of San Juan.

The conference will include a 3-day meeting with plenary sessions on each topic, invited speakers and an opportunity for short offered papers and posters. There are several interesting day (tropical rain forest) and night (Phosphorescence Bay) sites to visit as individuals or in organized group trips.

Further details of speakers/topics will be finalized shortly and will be available on the SGM website (http://www.sgm.ac.uk).

For further information, contact Hilary Lappin-Scott, , University of Exeter (email h.m.lappin-scott@exeter.ac.uk).

Learning and Teaching Support Network Centre for Bioscience

The LTSN is a new national network set up to promote and support high quality learning and teaching in all subject disciplines in UK higher education. The Centre for Bioscience is now in operation at Leeds University, where it is based in the School of Biochemistry and Molecular Biology. The Director is Professor Ed Wood. The main activities of the Centre are:

- Promotion of development and sharing of innovation and good practice in learning, teaching and assessment
- Formation of networks to facilitate communication between UK bioscientists of all disciplines
- Provision of a repository of materials and information to support the Centre's aims

Contact Dr Heather Sears, the specialist for microbiology, for further information (Tel. 0113 233 3001) or see **http://bio.ltsn.ac.uk**

Science on show

The new Wellcome Wing of the London Science Museum was launched in July. It occupies three strikingly decorated levels and is very different from the traditional museum in the way the exhibits are designed. Many are interactive. There is a big section on genetics. Tel. 020 7942 4455 for further information.

Royal Society

The new President of the Royal Society is to be Sir Robert May, formerly chief scientific adviser to the government.

Brilliant Careers

Channel 4 has launched a careers website for 16–24 year-olds which, according to the publicity, promises to offer '*information and advice with the distinctive Channel 4 twist*. The site provides information for those who have clear ideas about their future and those who are exploring their options. Key elements of the site include games, tests, guidance on creating successful CVs, application forms and interview techniques. The site will also feature a digital mentoring and advice service provided by career advisers and interested professionals.

The website is closely modelled on successful formats that Channel 4 has previously used and has potential to target young people who may not necessarily consult the more conventional sources of career advice. When External Relations Office staff initially contacted one of the site editors, there had been no immediate plans to include any information on biological science careers, although chemistry was represented. A meeting at Channel 4 established that they felt careers in bioscience should be included on the website but the editors need assistance from professional scientists. Which is where SGM members may be able to help . . .

The site features a bank of careers profiles of people who have followed a clear path in their chosen career and those who have taken a more unconventional approach either by choice or force of circumstance. Volunteers are asked to provide information about themselves in a short questionnaire which Channel 4 will turn into a profile. If you know someone with a suitable career path or perhaps think your own would be appropriate, please contact Jane Westwell at SGM HQ who will provide you with a questionnaire and will be happy to forward material to Channel 4. Digital mentors are also needed and if biosciences and microbiology are to be represented it is down to us. If you feel able to commit yourself to the occasional session of online mentoring, please contact Jane Westwell (j.westwell@sgm.ac.uk) for further details.

It's a record!

Dr Henry Tribe has received a certificate from Guinness confirming that his Millennium Bug, a large-scale model of *Escherichia coli* which has been displayed at many SGM meetings, is the largest model bacterium in the world.

Symposium Volume reviews

Community Structure and Co-operation in Biofilms. SGM Symposium Vol. 59

Edited by D.G. Allison, P. Gilbert, H.M. Lappin-Scott & M. Wilson
Published by Cambridge University Press (2000)

There is a 60 % discount to SGM members buying personal copies:
SGM Members £28.00/$50.00
Non-members £70.00/$125.00
Student Members £17.00

pp. 349, ISBN: 0-521-79302-5

The SGM symposium series has for long been a hallmark of the Society's commitment to scientific publishing at the highest level and has, in no small measure, contributed to its international standing. Beside the excellence of many of the individual volumes, the series as a whole has been characterized by the timeliness of the choice of subject areas to be presented. This is certainly true of this most recent volume on biofilms.

The word 'biofilm' first appeared in the title of an SGM symposium paper with my own contribution to the ecology volume in 1987. The 16 papers in the present volume neatly demonstrate the massive increase in importance of the subject area over the last 13 years. The awareness that the great majority of natural ecosystems involve mixed microbial communities growing as surface-associated aggregates, and that such sessile organisms may display differing phenotype and genotype from their planktonic cousins, has brought us to a genuine paradigm shift in our understanding of microbiology, pure and applied. The very magnitude of the difference in approach required for biofilm studies has, however, given rise to a paradox at the centre of much research in this field. While the inherent complexity of the geometrical, physical, chemical and biological heterogeneities are recognized, the demands of experimentation and interpretation often give rise to approaches that can be dangerously over-simplified. For this reason much research in biofilms has been closely concerned with methodology and input from the engineering and physical sciences has been critical to the development of a coherent body of hypotheses. Modelling has now begun to play a central role in this respect.

These unique aspects of biofilm research are clearly evident in the present volume. The 16 chapters, from most of the major groups in this country, Europe and North America, are organized into Introduction (1), Formation & structure, including detachment & modelling (7), Community interactions, including gene transfer (4), Practical applications (3) and Epilogue (1). While each article varies, according to subject matter and author, as to length and the extent to which an attempt is made to draw conclusions of more general significance, in every case the material is presented lucidly and with reference to the appropriate background studies. Key components of biofilm research that are identified, often by more than one author, are: the obligatory interdisciplinary nature of biofilm studies; cell–cell communication mediated by acyl homoserine lactones (although it is surprising how little is actually known about this aspect); the often deterministic influence of hydrodynamic and other physical forces on biological expression; the focus and rigour imposed on biological studies by the application of modelling; the constant interplay between environmental factors, physical and chemical, on the biological response of biofilm systems; the dynamic nature of biofilms at all stages of their development; the counter-intuitive conclusion that gene transfer rates are not necessarily enhanced in biofilms.

In common with all previous books in the series, this biofilm volume has set itself the difficult task of introducing the subject area to a new scientific audience, while at the same time serving as an up-to-the-minute statement of current developments, expressed at a level and depth valuable to the various experts themselves actively involved in one or other corner of this particular research field. In this it has succeeded, and shown itself to be worthy of inclusion in the pantheon of the SGM symposium series.

■ ***Allan Hamilton, University of Aberdeen***

The book can be ordered by post using the form in this issue of *Microbiology Today*. This form can also be used to order any past volumes missed by members at the time of publication. Student Members wishing to purchase Symposium Volumes should contact the Grants Office at Marlborough House for a special form.

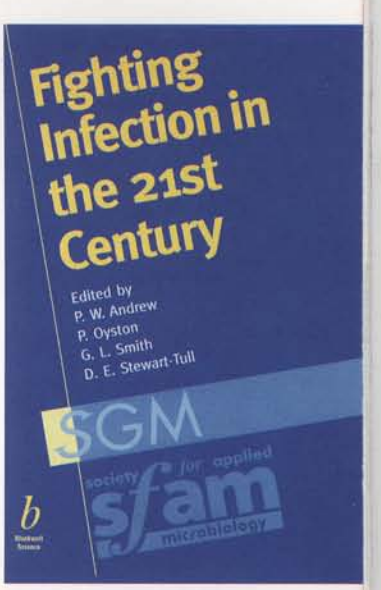
Fighting Infection in the 21st Century

Edited by P.W. Andrew, P. Oyston, G.L. Smith & D.E. Stewart-Tull
Published by Blackwell Science (2000)

Non-members £65.00
SGM members are entitled to a 40 % discount on the book. The price to them is £39.00.

pp. 272, ISBN: 0-632-0581-7-X

Advances in medical sciences during the 20th Century led to dramatic increases in life expectancy worldwide. The 21st Century began with the announcement that the human genome has been sequenced – the popular press predicts cures for all mankind's ills within the year! At the same time, the World Health Organization warns, in its recent report *Removing Obstacles to Healthy Development*, that 'an infectious disease crisis of global proportions is today threatening hard-won gains in health and life expectancy'. Infections account for 13 million deaths a year globally, one in every two deaths in developing countries. Of these, more than half are of children under 5 years old. And even among those children who survive infection, their development may be so badly impaired by disease that they will never reach their full potential as individuals and productive members of society. Knowing the human genome will not help them; understanding infectious disease sufficiently to be able to do something about it might.



Fighting Infection in the 21st Century is a timely, interesting and useful addition to the bookshelf (bed-side table?) of anyone concerned about the paradox that the incidence of infections, both ancient and emerging, is increasing relentlessly despite our ability to describe the virulence properties of many pathogens in incredible molecular and cell biological detail. There are chapters that celebrate new and exciting technologies (genome-based discovery of new vaccines, vaccine production in plants) and a chapter that proposes the re-evaluation of old approaches (probiotics and phage). There are chapters that rehearse the lessons to be learned from past experiences (live attenuated vaccines, the first antibiotic era) and others that describe in elegant detail the possibilities for new vaccines and diagnostic tests (conserved epitopes in LPS, multilocus sequence typing). And lest we forget that microbiology is an essentially multidisciplinary subject, there are chapters reminding us that the greatest advances, in numerical terms at least, are just as likely to come from better hygiene and higher quality foods as from sophisticated post-genomic molecular biology. Not surprisingly for a multi-authored book of this type, the quality of the chapters is somewhat variable in terms of novelty of ideas, level of scientific detail and ability to stimulate debate. Nevertheless, the book as a whole is a 'good read', and indeed its very heterogeneity is somehow symbolic of its inescapable message, that future successes in the fight against infection will depend heavily on '*all the approaches we can find, whether old and unfashionable or new and trendy*'.

■ ***Peter Williams, University of Leicester***

An order form may be obtained from Science Marketing Department, Blackwell Science Ltd, Osney Mead, Oxford OX2 0EL (Tel. +44 (0)1865 206013; Fax +44 (0)1865 721205; www.blackwell-science.com).

Reviews

If you would like your name to be added to our database of book reviewers, please complete the book reviewer interests form now available on the SGM website.

A classified compendium of book reviews from 1996 to the present is also available on the website.

The Variety of Life: A Survey and A Celebration Of All The Creatures That Have Ever Lived

By C. Tudge
Published by Oxford University Press (2000)
£35.00, pp. 684
ISBN: 0-19-850311-3

This book attempts the monumental task of introducing all the principal groups of organisms that have existed on Earth. Inevitably there are a few shortcomings, such as an absence of labelled diagrams to augment the textual descriptions of organism structure, some inconsistencies in the use of formal taxonomic names and imbalances in the coverage (the prokaryotes, for example, are dealt with in just 20 out of 684 pp.). Nevertheless, this book has far more strong points than weak, principally the accuracy of the information and its presentation in the context of modern concepts of phylogenetics. There is also an excellent introductory section explaining the principles of evolution and systematics. The target audience is the non-specialist and the aim is to guide the reader with no knowledge of systematics to the point at which they can access the specialist literature themselves. In this, I believe the book succeeds.

■ ***Alan Warren***
Natural History Museum, London

Alkaliphiles

By K. Horikoshi
Published by Harwood Academic (1999)
£75.00/US$120.00/EUR100.00, pp. 338
ISBN: 90-5702-458-6

This is, effectively, the third edition of a book whose origins go back to 1982 when the first version, *Alkaliphilic Microorganisms : a New Microbial World*, appeared, followed by *Microorganisms in Alkaline Environments* in 1991. The three books represent a fascinating account of Koki Horikoshi's lifelong work on alkaliphilic micro-organisms. This third work comprises eight large chapters that outline the isolation and taxonomy of alkaliphiles, their form and function, including the biochemical basis of alkaliphily, the current state of the molecular biology, and finally, around half of the book is devoted to enzymes produced by these microbes and the industrial applications. This version differs from previous books in having a more detailed consideration of alkaliphile ecology and systematics, and of course, the molecular biology, which has evolved considerably over the last two decades, culminating in the preliminary data leading to the complete genome sequence of the model alkaliphile *Bacillus* sp. C-125, currently almost complete. The book is very much for dipping into as an advanced reference source for those with specific biotechnological or molecular questions and is definitely not a beginner's guide to alkaliphiles. Indispensable reading for those with industrial leanings.

■ ***Bill Grant***
University of Leicester

The Dynamic Cell: A New Concept for Teaching Molecular Cell Biology (CD-ROM)

Edited by K. Dawson, T. Devlin, M. Klymkowsky, Y. Rochev, M. Snyder, M. Steer & J. Widom
Published by Springer-Verlag (2000)
DM1,972.00/öS12,410.00/sFr1,530.00/£737.07/US$1000.00 *for up to 20 users*
ISBN: 3-540-14723-3

This is an original approach to teaching molecular cell biology, being based around virtual reality simulations of five sites in a eukaryotic cell. At each site, the user can click on any cellular components that are visible and a menu appears with links to further information. This pops up another window containing graphics and text. Some of the animations here are excellent and would be a significant help to students trying to grasp how various processes work. The quality of some images is disappointing, however, reflecting the fact that the program is designed for a small 16-colour screen. The text is comprehensive but comes mainly as large blocks that are sometimes badly structured with numerous typographical errors. There are other small quirks, but I think most students would find the package very rewarding. It is definitely worth having a look, although the price may make you think twice.

■ ***Mike Tait***
University of Wales Swansea

Chaperonin Protocols. Methods in Molecular Biology, Vol 140

Edited by C. Schneider
Published by Humana Press (2000)
US$79.50, pp.232
ISBN: 0-89603-739-8

This is a specialized protocol book which brings together an invaluable overview of two main aspects of chaperonin research – purification of chaperonins and their cofactors and assays to monitor folding activity. Not surprisingly much of the book deals with the GroEL/ES chaperonin of *Escherichia coli*. However, there are also a number of excellent chapters on more diverse chaperonins, including those from *Archaea* and the specialized, eukaryotic TRiC complex. Anyone investigating chaperonins from other species will also find the chapter dealing with chaperonin activity *in vivo* in *E. coli* useful. Noticeable is the lack of any protocol on EM analysis of chaperonins; indeed structural aspects are only addressed indirectly. The book provides a wealth of explicitly detailed protocols (together with useful troubleshooting notes) for all those working in the field of chaperonins and protein folding or misfolding.

■ ***Sheila MacIntyre***
University of Reading

A Passion for DNA: Genes, Genomes, and Society

By J.D.Watson
Published by Oxford University Press (2000)
£18.99, pp. 256
ISBN: 0-19-850697-X

Although some are autobiographical, these essays, written periodically from the sixties through to the present, mainly present Watson's views, often radical, on the topical issues of the day: a fascinating journey through the development of molecular genetics. The writing is clear, to the point and very readable. But, apart from simply enjoying imaginary arguments with Watson over the controversial points he makes on using mice for medical research, the dangers of genetically manipulated organisms and the pros and cons of the human genome project (just three examples), I found myself planning how I could use the essays for discussion in tutorial groups at both under- and postgraduate levels. The text will not only inform and broaden student minds but also stimulate debate. I recommend it for the general reading list with which we encourage our students to expand their horizons. It also provides material for use in communicating science to the public.

■ ***Chris Thomas***
University of Birmingham

The Nucleic Acid Protocols Handbook

Edited by R. Rapley
Published by Humana Press (2000)
US$149.50, pp. 1076
ISBN 0-89603-459-3

This is a collection of 120 protocols of 3–10 pages each. The book is stronger on DNA than RNA analysis. It does not contain much on sequence evaluation (editing, phylogenetic analysis etc) which is the goal for considerable parts of nucleic acid work undertaken.

Whilst having practitioners of particular procedures providing the craftsmanship of the trade (with many excellent notes) is highly useful, in my opinion the book suffers from a lack of a systemic approach which has led to duplications, basic information being hidden in chapters (not helped by the very basic index) and a certain degree of eclecticism and over-ambition (the Human Genome Project being mentioned on half a page!). Not in vain do many authors cite *Molecular Cloning: A Laboratory Manual* by Sambrook, Fritsch & Maniatis (2nd edn, 1989) which, although lacking the progress made in the last decade, has a much preferred layout and structure. Thus, I regard the book as a very useful source of individual workers' experience rather than as a comprehensive reference book.

■ ***Ulrich Desselberger***
Addenbrooke's Hospital, Cambridge

Strange and Charmed: Science and the Contemporary Visual Arts

Edited by S. Ede
Published by Calouste Gulbenkian Foundation (UK) (2000)
Available from Turnaround Publisher Services Ltd (email: orders@turnaround-uk.com)
£10.99 + £2.00 p&p, pp. 200
ISBN: 0-903319-87-X

It is difficult to express all the reactions which this book provokes, but the persistent background question posed is 'why?' Accepting that there is a debate between science and contemporary visual arts it is difficult to decide whether artists are searching science for inspiration, using visual imagery to enable 'understanding', or any other combination of motive. Although the book is about contemporary visual arts there are relatively few photographs or other illustrations. Why the art/science relationship needs to be expressed more in words than in pictures is curious.
The images used raise serious questions about the nature of modern art. This in itself is difficult to define and further definition relating modern art to science is even more abstract. Notwithstanding the above there are many riveting images and adroit points made. This book repays close study.

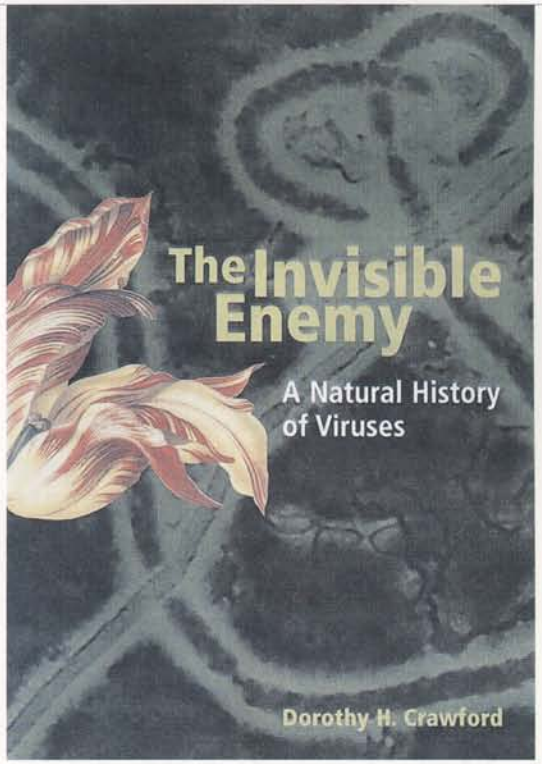
■ ***Andrew Eastabrook***
Stow on the Wold

Cellular Microbiology: Bacteria–Host Interactions in Health and Disease

By B. Henderson, M. Wilson, R. McNab & A.J. Lax
Published by John Wiley & Sons (1999)
£29.95, pp. 452
ISBN: 0-471-98681-X

A splendidly written book, which bridges the broad areas of eukaryotic and prokaryotic biology. The informative introduction, with its historic perspective, is pleasurable to read and whets the appetite for the following text. The authors have made the book accessible for a wide readership as evidenced from the nomenclature adopted and from the well balanced background information covered in the early sections. The ensuing chapters deal with several complex subject areas comprehensibly. The many useful tables and diagrams, as well as the writing style, make this an extremely readable book. Given the scope covered by the authors, I found few inconsistencies or inaccuracies. Ones that I noted are due to current ambiguities in those fields. I have bought a copy for my laboratory and it has been greatly appreciated. In my view the authors have achieved their goal and the book should be useful to students and researchers in many subject areas.

■ ***Mumtaz Virji***
University of Bristol



Glycoprotein Methods and Protocols: The Mucins. Methods in Molecular Biology, Vol. 125

Edited by A.P. Corfield
Published by Humana Press (2000)
US$119.50, pp. 528
ISBN: 0-89603-720-7

Using a consistent format, each chapter provides a specific preparative or analytical method. There is sufficient detail to follow easily and unlike many research papers, the minute details which make for success or failure, all appear to be included. Each section of the book provides a series of related methods and develops rationally from preparation to analysis, structural procedures and detection. It is a highly specialized volume and, apart from the Chapter on mucin-degrading bacteria, is probably not so useful to the microbiologist with only a fleeting interest in mucins.
As with many such volumes containing so many authors, stricter editorial control might usefully have been applied. There is no preface or introduction and many chapters contain very long lists of 'notes'. This means that the reader must continually look at the main text containing the methods, then switch to the cautionary warnings found in the notes. Perhaps a more user-friendly format could have been adopted.

■ ***Ian W. Sutherland***
University of Edinburgh

The Invisible Enemy: a Natural History of Viruses

By D.H. Crawford
Published by Oxford University Press (2000)
£14.99, pp. 288
ISBN 0-19-850332-6

The viruses here consist almost entirely of those affecting humans; plant viruses receive a brief mention in the form of flower-breaking in tulips, and viruses of other animals are considered mainly in terms of their ability to cross species barriers to us and cause new or modified disease problems. Within the human context, the treatment is very wide-ranging; a true natural history of the importance of viruses in the development of our civilization, current and emerging threats to health and different control methods. Related areas, such as transmissible spongiform encephalopathies, and the possible involvement of viruses in chronic fatigue syndrome, are also explored. Professor Crawford writes in a clear and accessible manner, which successfully conveys her fascination with the subject, and poses lots of interesting questions. Recommended reading for microbiologists; highly recommended for journalists, politicians and the public seeking understanding of big topical issues such as AIDS/HIV and BSE/CJD.

■ ***Ron Fraser***
SGM, Marlborough House

Protein Expression. A Practical Approach. Practical Approach Series no. 202

Edited by S.J. Higgins & B.D. Hames
Published by Oxford University Press (1999)
£31.95, pp. 304
ISBN: 0-19-963623-0

This book is one of the large number of specialist titles in the well-known *Practical Approach* series. This techniques-oriented series is aimed at providing background to the relevant methodology together with detailed laboratory protocols that can be easily followed in a cook-book fashion. The Editors explain that this volume on *Protein Expression* and its companion on *Post-translational Processing* are intended to update the volume on *Transcription and Translation* which was published in the series some years ago. The articles have international authorship and cover in an authoritative fashion all the commonly used cellular protein expression systems, as well as cell-free translation. The practical advice given will be especially useful for anyone wanting an up-to-date introduction to these experimental systems. The earlier volume on *Transcription and Translation* has been well used in my laboratory, and I am sure this update will become equally well thumbed.

■ ***Mike Tanner***
University of Bristol

Yeasts: Characteristics and Identification, Third Edition

By J.A. Barnett, R.W. Payne & D. Yarrow with L. Barnett
Published by Cambridge University Press (2000)
£200.00, pp. 1,139
ISBN 0-521-57396-3

The entire Dutch collection of yeasts was re-examined and over 100 characters used to describe the 706 species in this book. It benefits greatly from the unified approach of just three authors who have produced numerous keys to uniquely identify a species very rapidly. The large format is attractive and each species is usually allocated one page which includes up to five good quality (and useful) photographs. There is one similarly priced competitor which has similar coverage but different degrees of detail. I was pleased to see here a (correct) +/– growth response on glycerol for *Saccharomyces cerevisiae*! Both volumes are very good, the expert will probably want both, but this is the single volume I would recommend for any lab doing identification work because of its keys and consistency. Unfortunately, neither has yet got to grips with the onset of molecular taxonomy and there are no primer or sequence data.

■ ***Alan Wheals***
University of Bath

Viruses, Plagues and History

By M.B.A. Oldstone
Published by Oxford University Press (2000)
£10.99, pp. 228
ISBN: 0-19-513422-2

Intended to succeed and update Paul de Kruif's popular book *Microbe Hunters*, this is a modern account of successes (smallpox, yellow fever, measles and poliomyelitis) and failures to control microbial infections (AIDS, Lassa fever, Ebola fever, Hantavirus-associated diseases), told by a senior researcher in the area of viral molecular immunology and pathogenesis, in particular persistent infections. The limited success in controlling influenza is also recorded as is the story of the recognition of prions as infectious proteinaceous agents causing transmissible spongiform encephalopathies (scrapie, Creutzfeldt–Jakob disease, bovine spongiform encephalopathy, etc.). This is the paperback edition of the book which was published in 1998 and has therefore been extended by up-to-date 'Afternotes' relating to issues of bioterrorism, newly emerging viruses and zoonotic transmission.
Dr Oldstone looks back on a very prominent research career and has been and is in personal contact with many of the protagonist researchers in the areas discussed. The book is written in a lively style and makes fascinating reading. It should be in the library of every student and researcher of biomedical sciences.

■ ***Ulrich Desselberger***
Addenbrooke's Hospital, Cambridge

RNA Viruses: A Practical Approach

Edited by A.J. Cann
Published by Oxford University Press (2000)
£29.95, pp. 266
ISBN 0-19-963716-4

Like other volumes in this series, this book will be a very useful laboratory manual and its price is well within the reach of most researchers. As well as containing clear and easy-to-follow protocols for studying and using RNA viruses, each chapter gives useful background information and explanation of the principles behind the laboratory procedures. This volume not only includes techniques for studying the structure, expression and packaging of viral genomes, but also procedures for engineering viruses with desired characteristics and for studying integration and transduction of viral oncogenes. The book would benefit, however, by the addition of methods for studying virion–receptor interactions, translation and polymerases, as well as protocols for the generation of alphavirus vectors. Some of the methods, such as those for nucleic acid extraction, are a little outdated and it is unfortunate that commercially available kits are not mentioned more frequently.

■ ***Christopher Ring***
Glaxo Wellcome R&D, Stevenage

Oral Bacterial Ecology: The Molecular Basis

Edited by H.K. Kuramitsu & R.P. Ellen
Published by Horizon Scientific Press (2000)
£74.99/US$149.99, pp. 314
ISBN: 1-898486-22-0

A central theme of this book is that the oral cavity provides an ideal model system for the application of molecular techniques to investigate the complex interactions between bacteria and their environment in health and disease. What better place indeed? The mouth provides numerous different environments for growth and supports some 700 taxa. Considerable information is already available on many aspects of oral microbial physiology and ecology and this is reviewed comprehensively here. The chapter on oral innate immune responses provides a welcome host perspective. The application of molecular techniques, such as *in vivo* expression technology and differential-display PCR to study oral bacterial ecology is still in its infancy, although the potential was highlighted in several chapters. Some of the subjects have been reviewed elsewhere recently; however, the book does provide a coherent collection of chapters that workers in the oral field as well as newcomers will find very useful.

■ ***Rod McNab***
Eastman Dental Institute, University College London

The Pocket Guide to Fungal Infection

By M.D. Richardson & E.M. Johnson
Published by Blackwell Science (2000)
£14.95, pp. 114
ISBN: 0-632-05325-9

This handy slim volume wastes no space on frills but provides a succinct and very well organized overview of mycoses based on sub-divisions of types of fungal infections (dermatophytosis, opportunistic infections, etc.). Each organism or disease syndrome is subdivided under a number of common subheadings covering aspects of disease distribution, causal agents, clinical manifestations, diagnosis and management. The information is dealt with as notes and bullet points, plus a perhaps disappointingly short reference list and useful websites. The volume is very nicely illustrated with high quality colour illustrations and succeeds well in achieving its objective in summarising '*the major features of fungal infections of humans*'. The book is inexpensive and likely to be of use both in a clinical and teaching setting.
No doubt some will argue about details of the management of specific diseases – but these are always under debate.
Overall this is good value.

■ ***Neil A.R. Gow***
University of Aberdeen

Biochemistry, Third Edition

By C.K. Mathews, K.E. van Holde & K.G. Ahern
Published by Benjamin/Cummings (2000) D/B Pearson Education
£31.99, pp. 1,186
ISBN: 0-8053-3066-6

Undergraduate and postgraduate microbiology students will find this a very appealing textbook. It is up-to-date, beautifully illustrated, and has good 'further reading' bibliographies and index. It is expensive, but contains a CD-ROM 'study guide' that provides topic outlines, explains concepts and allows students to test themselves. However, the added value from the CD is not great and the academic imperative is not clear. A lot of the CD material reproduces the book. Many of the CD illustrations lack legends and cannot be displayed on the same screen as the relevant commentary. Some are pictures from the text which cannot be interpreted without reference to the book. Hyperlinks to useful websites are provided, patchily. The publishers offer continuous updating of this facility through a good supporting website. A lovely book, well worth considering.

■ ***Ian Hancock***
Newcastle-upon-Tyne

Books received

Food Biotechnology in Ethical Perspective

By P.B. Thompson
Published by Blackie A & P (1997)
£35.00, pp. 267
ISBN: 0-412-78380-0

Landmarks in Intracellular Signalling

Edited by R.D. Burgoyne & O.H. Petersen
Published by Portland Press (1997)
£20.00/US$34.00, pp. 278
ISBN: 1-85578-101-8

In Situ and On-Site Bioremediation: Vol. 4

Symposium Chairs: B.C. Alleman & A. Leeson
Published by Battelle Press (1997)
US$82.50, pp. 619
ISBN: 1-57477-029-2

Manual of Environmental Microbiology

Edited by C.J. Hurst, G.R. Knudsen, M.J. McInerney, L.D. Stetzenbach & M.V. Walter
Published by ASM Press (1997)
£59.50, pp. 894
ISBN: 1-55581-087-X

Chemokine Protocols. Methods in Molecular Biology, Vol. 138

Edited by A.E.I. Proudfoot, T.N.C. Wells & C.A. Power
Published by Humana Press (2000)
US$99.50, pp. 332
ISBN: 0-89603-722-3

Diary

january 2001

EUROPEAN SOCIETY FOR ANIMAL CELL TECHNOLOGY – UK BRANCH. ESACT-UK. 11TH ANNUAL SCIENTIFIC MEETING

Churchill College, Cambridge 4–5 January 2001

CONTACT: Julian Hanak (Meetings Secretary) for registration and programme details (Fax 01782 714167; email julian.hanak@cobrat.com) and Jon Green for Trade Exhibition (Fax 01763 263413; email jon.green@cam-antibody.co.uk)

QA-TQM-COURSE, MODULE 5: BIOLOGICAL PRODUCTION: FROM GENETICS TO DOWNSTREAM PROCESSING

Delft, The Netherlands 10–12 January 2001

CONTACT: Dr L.A. van der Meer-Lerk or Ms. G.W.J.O. Aggenbach, Biotechnology Studies Delft Leiden (BODL), The Netherlands (Tel. +31 15 2781922; Fax +31 15 2782355; email BODL@tnw.tudelft.nl; http://www.bt.tudelft.nl/bodlf.htm)

february 2001

BACTERIAL RESISTANCE: CLINICAL CHALLENGES AND EXPERT PERSPECTIVES

SCI, Belgrave Square, London 23 February 2001

CONTACT: SCI, 15 Belgrave Square, London SW1X 8PS (Tel. 020 7598 1500; Fax 020 7235 9410)

march 2001

ADVANCED COURSE ON BIOCATALYSIS

Delft, The Netherlands 5–9 March 2001

CONTACT: Dr L.A. van der Meer-Lerk or Ms. G.W.J.O. Aggenbach, Biotechnology Studies Delft Leiden (BODL), The Netherlands (Tel. +31 15 2781922; Fax +31 15 2782355; email BODL@tnw.tudelft.nl; http://www.bt.tudelft.nl/bodlf.htm)

XXXVII BRAZILIAN SOCIETY OF TROPICAL MEDICINE CONGRESS BAHIA CONVENTION

Center in Salvador, Bahia, Brasil, 11–15 March 2001

CONTACT: Eventus System, Rua Oito de Dezembro, 547, Graça, CEP 40150-000 Salvador, Bahia, Brazil (Tel. + 55 71 264 3477 Fax: + 55 71 264 0508; email: eventus@cpunet.com.br/; http://www.cpunet.com.br/eventus)

QA-TQM-COURSE, MODULE 6: FOOD TECHNOLOGY AND FOOD SAFETY

Wageningen, The Netherlands 12–14 March 2001

CONTACT: Dr L.A. van der Meer-Lerk or Ms. G.W.J.O. Aggenbach, Biotechnology Studies Delft Leiden (BODL), The Netherlands (Tel. +31 15 2781922; Fax +31 15 2782355; email BODL@tnw.tudelft.nl; http://www.bt.tudelft.nl/bodlf.htm)

april 2001

METALS & CELLS. SEB AGM

Canterbury, 2–6 April 2001

CONTACT: Dr Pamela Robinson (email pamela.robinson@ncl.ac.uk; http://www.ncl.ac.uk/sbg/robinson/metalcell.html)

MOLECULAR BIOLOGY UPDATE – A FOUR-DAY LABORATORY COURSE

Hatfield, Herts, 9–12 April 2001

CONTACT: Professor John Walker, Dept of Biosciences, University of Hertfordshire, College Lane, Hatfield, Herts AL10 9AB (Tel. 01707 284546; Fax 01707 284510; email j.m.walker@herts.ac.uk; http://www.herts.ac.uk/natsci/STC)

may 2001

ADVANCED COURSE ON DOWNSTREAM PROCESSING

Delft, The Netherlands 7–11 May 2001

CONTACT: Dr L.A. van der Meer-Lerk or Ms. G.W.J.O. Aggenbach, Biotechnology Studies Delft Leiden (BODL), The Netherlands (Tel. +31 15 2781922; Fax +31 15 2782355; email BODL@tnw.tudelft.nl; http://www.bt.tudelft.nl/bodlf.htm)

10TH INTERNATIONAL CONGRESS OF HUMAN GENETICS

Vienna, Austria, 15–19 May 2001

CONTACT: ICHG Office, c/o Vienna Medical Academy, Alser Strasse 4, A-1090 Vienna, Austria (Tel. +43 1 405 13 83 33; Fax +43 1 407 82 74; email office@ichg2001.org)

june 2001

10TH TOMASEK DAYS

Brno, Czechia, 6–8 June 2001

CONTACT: Ondrej Zahradnicek, Institute for Microbiology, St Anna's University Hospital and Medical School, Masaryk University, Brno Pekarska 53, CZ-656 91 Brno, Czechia (Tel. +42 0 5 4318309; Fax 42 0 5 4318308; email ozahrad@med.muni.cz; http://www.med.muni.cz/zahrad/strtomda.htm)

ADVANCED COURSE ON ENVIRONMENTAL BIOTECHNOLOGY

Delft, The Netherlands 20–29 June 2001

CONTACT: Dr L.A. van der Meer-Lerk or Ms. G.W.J.O. Aggenbach, Biotechnology Studies Delft Leiden (BODL), The Netherlands (Tel. +31 15 2781922; Fax +31 15 2782355; email BODL@tnw.tudelft.nl; http://www.bt.tudelft.nl/bodlf.htm)

6TH EUROPEAN CONFERENCE ON EXPERIMENTAL AIDS RESEARCH (ECEAR' 2001)

Edinburgh Conference Centre 23–26 June 2001

CONTACT: ECEAR' 2001 Conference Secretary, Division of Retrovirology, NIBSC, Blanche Lane, South Mimms, Potters Bar, Hertfordshire EN6 3QG (email ecear2001@nibsc.ac.uk; http://www.nibsc.ac.uk/ECEAR2001)

2ND EUROPEAN CELLS & MATERIALS MEETING

Davos, Switzerland 25–28 June 2001

CONTACT: Conference Secretary: Sonia Wahl (email sonia.wahl@ao-asif.ch; http://www.ao-asif.ch/events/other/ecm/index.html)

july 2001

AN INTRODUCTION TO BIOINFORMATICS – A TWO-DAY COMPUTER/LECTURE COURSE

Hatfield, Herts 3–4 *or* 10–11 July 2001

CONTACT: Dr Henry Brzeski, Dept of Biosciences, University of Hertfordshire, College Lane, Hatfield, Herts AL10 9AB (Tel. 01707 284554; Fax 01707 286137; email h.brzeski@herts.ac.uk; http://www.herts.ac.uk/natsci/STC)

RNA EXTRACTION AND ANALYSIS – A ONE-DAY LABORATORY/LECTURE COURSE

Hatfield, Herts, 5 July 2001

CONTACT: Dr Ralph Rapley, Dept of Biosciences, University of Hertfordshire, College Lane, Hatfield, Herts AL10 9AB (Tel. 01707 285097; Fax 01707 286137; email r.rapley@herts.ac.uk; http://www.herts.ac.uk/natsci/STC)

PCR METHODS AND APPLICATIONS – A ONE-DAY LABORATORY/LECTURE COURSE

Hatfield, Herts, 6 *or* 13 July 2001

CONTACT: Dr Ralph Rapley, Dept of Biosciences, University of Hertfordshire, College Lane, Hatfield, Herts AL10 9AB (Tel. 01707 285097; Fax 01707 286137; email r.rapley@herts.ac.uk; http://www.herts.ac.uk/natsci/STC)

IXTH INTERNATIONAL MEETING ON THE BIOLOGY AND PATHOGENICITY OF FREE LIVING AMOEBAE

Paris, France, 8–14 July 2001

CONTACT: Secretariat Check Up Service, 16 rue du General Faidherbe, BP 42, 94732 Nogent sur Marne cedex, France (Tel: +33 1 48 77 01 13; Fax +33 1 48 77 08 63; email amoebae2001@amoebae2001.com; http://www.amoebae2001.com)

september 2001

ESCV '01 PROGRESS IN CLINICAL VIROLOGY VII

Lahti, Finland 2–5 September 2001

CONTACT: Organizing Secretariat & Congress Office, University of Helsinki, Lahti Research and Training Centre, Kirkkokatu 16, FIN-15140 Lahti, Finland (Tel. +358 3 892 20514; Fax +358 3 892 20219; email irmeli.paasikivi@helsinki.fi; antti.vaheri@helsinki.fi; virpi.tiilikainen@helsinki.fi)

PSEUDOMONAS 2001

Brussels, Belgium 17–21 September 2001

CONTACT: Pierre Cornelis, Laboratory of Microbial Interactions, Dept of Immunology, Parasitology & Ultrastructure, Flanders Interuniversity Institute of Biotechnology, Vrije Universiteit Brussel, Paardenstraat 65, B-1640 Sint Genesius Rode, Belgium (Tel. +32 2 3590221; Fax +32 2 3590399; email pcornel@vub.ac.be; http://homepages.vub.ac.be/~pcornel/Pseudomonas2001.htm)

Comment

Archaea are really mesophiles with attitude!

Mention the *Archaea* to most microbiologists and they instinctively think of extremophiles. This is to be expected for the *Archaea* were originally recognized as a distinct group because of the common properties of various extremophiles. As this knowledge spreads into the wider culture, especially to those for whom environmental microbes are of but passing interest, a certain amount of extrapolation goes on. The *Archaea* are described as 'primitive', similar to organisms from early Earth, where the conditions are perceived as extreme compared with today. Second, the *Archaea* are still considered to be extremophiles, restricted to growth in places where sensible organisms can only be found dead. These two perceptions are self-reinforcing, producing the impression that the *Archaea* are survivors, clinging to niches resembling the conditions on Earth aeons ago.

We actually know little about the conditions on the early Earth with any certainty and almost all of what we know has been revealed in the past couple of decades. The Solar System formed around 4·56 Gyr ($4{\cdot}56 \times 10^9$ years) ago from a collapsing molecular cloud of gas and dust. Aggregation of that dust produced the Earth and planets, which continued to grow by accretion of small bodies. A combination of energy from impacts, radioactive decay and gravitational energy heated the growing planet, to the extent that it had a molten surface. As the number of small bodies available to be captured fell, the planet cooled and in due course a solid crust formed. The crustal surface of the Earth is recycled through plate tectonics and the oldest surviving bits of crust have been subjected to considerable heat, so the nature of the original crust has to be surmised from indirect evidence. As the early Earth continued to cool, the atmosphere stabilized, allowing an ocean to form. It is believed that the early Earth was completely covered in water, so no 'warm little pond' for Darwin's cradle of life. Relative age-dating of craters on the Moon show that the Earth–Moon system suffered heavy bombardment by asteroids and comets, forming craters around 4·0 Gyr ago. The impact energy generated by collisions probably evaporated much of the water on the planet. The oldest piece of sedimentary crust that is currently known is about 3·6 Gyr old and contains fossils which bear a striking resemblance to cyanobacteria. These fossils are found in cherts and a chert is formed when pieces of rock fall into a sediment, like pebbles carried into an estuary. The sediment is 3·6 Gyr old, but the fossils are found in the pieces of rock and are therefore older. So, the most plausible sequence of events would seem to be that life formed in the oceanic early Earth. The heavy bombardment did not evaporate all water, but the elevated temperature put a severe bottle-neck in evolution so that only thermophilic strains survived, which explains why the earliest branches to emerge from the 16S rDNA tree of life were thermophiles.

So far so good. The advent of molecular biology and its application to microbial ecology has revealed that the world is full of *Archaea* in perfectly normal environments. Many of these branch very deeply and disrupt the 'thermophiles-at-the-bottom' view of evolution. What is more, when the microscope of molecular biology is turned on these extremophiles, the underlying chemistry proves to be very similar to that of mesophiles, but with certain modifications to protect the mechanisms from these extreme conditions. It is difficult to see how evolution could produce vulnerable chemistry and protective mechanisms at the same time. It is more reasonable to conclude that extremophiles are adapted mesophiles and not the font from which mesophiles emerged.

This version of the story would be more convincing if we could manage to cultivate some of these lineages whose existence is known only from environmental rDNA samples. What role are they playing in the environment? Is there anything special about their chemistry? The questions are legion, but the only method we have to address them requires cultivation as a first step. Suffice it to say that we have a very poor knowledge of the extent of diversity in the *Archaea*.

The purpose of this comment is to illuminate a view of the *Archaea in toto*. For those already involved in microbial ecology there will be nothing new here, but as the field is burgeoning and funding opportunities are increasing, more people are being tempted into the water. Come on in and welcome, but it is a great help to read widely and to pay attention to the bits between the lines.

● ***Dr Dave Roberts, former Editor of Microbiology Today, is at the Natural History Museum, Cromwell Road, London SW7 5BD. Tel. 0207 942 5086; Fax 0207 942 5433; email d.roberts@nhm.ac.uk***

● Please note that views expressed in *Comment* do not necessarily reflect official policy of the SGM Council.