News in brief

Targets and mechanisms

Bacteriophage lends helping hand against bacteria

Researchers have found a novel way to kill the streptococci bacteria that cause sore throats, necrotizing fasciitis and other infections. The method exploits enzymes that are produced by bacteriophages after replication that puncture the bacterial cell wall and enable their exit from the cell. The research carried out at Rockefeller University (New York, NY, USA) is particularly promising as an anti-bacterial therapy because it could avoid the evolution of resistance, which is a major problem associated with antibiotic therapy.

The lytic enzymes produced by the bacteria are specific for the bacterial cells in which they were produced, and, therefore, they provide a way to kill pathogenic bacteria without perturbing the normal microflora of the patient, another problem that is common with antibiotics. In a report published in *Proceedings of the National Academy of Science USA*[1], 1,000 units (10 ng) of a murein hydrolase (termed lysin) isolated from the streptococcal bacteriophage C1 was shown to sterilize a culture of 10^7 Group A streptococci within 5 sec. Further, a 250-unit dose of lysin was shown to both prevent and eradicate colonization of streptococci in the oral cavity of mice.

Almost one-fifth of the population is a carrier of Group A streptococci. This rapid, lethal method of killing these bacteria is a powerful approach to eliminating or reducing streptococci infection in the upper respiratory mucosal epithelium of carriers or infected individuals, and could reduce associated diseases such as rheumatic fever, which can cause permanent heart damage.

1 Nelson, D. *et al*. (2001) Prevention and elimination of upper respiratory colonization of mice by Group A streptococci by using a bacteriophage lytic enzyme. *Proc. Natl. Acad. Sci. U. S. A.* 98, 4107–4112

Worms uncover complexity of insulin signalling

Studies of insulin-like genes encoded by the worm genome suggest that there are undiscovered members of the insulin family in humans. Researchers from Massachusetts General Hospital (Boston, MA, USA) have used genomics and genetic approaches to characterize predicted insulin-like genes in *Caenorhabditis elegans*, and have found 37 putative insulin genes[2].

The activity of DAF-2, the *C. elegans* form of the insulin-like receptor, is required for growth and reproduction of the nematode. Many of the 37 genes identified are divergent members of the insulin superfamily, and clustering of the genes is thought to suggest recent diversification. Of the 37 members of the *ins* gene family, which is implicated in reproduction, the *ins-1* gene is most similar to the human insulin gene, based on predicted protein structure and cleavage patterns. Overexpression of *ins-1,* and expression of human insulin under the control of *ins-1* regulatory elements, resulted in antagonism of the DAF-2 pathway. However, deletion of the *ins-1* coding region did not enhance the DAF-2 pathway, which suggests there is redundancy among the 37 insulin-like genes.

The discovery of such a large family of insulin-like genes in *C. elegans* suggests that there are likely to be more insulin genes and forms of insulin protein in

Patents

BMS allows emergency patent relief for Zerit

Bristol-Myers Squibb (BMS) has announced new initiatives to help fight HIV/AIDS in sub-Saharan Africa. These initiatives include reducing the prices of its two AIDS treatments, Videx (didanosine) and Zerit (stavudine), to below cost under its existing ACCESS partnership program with international agencies (such as UNAIDS, WHO, World Bank, UNICEF and the UN Population Fund). They have pledged to make the drugs available to every country in Africa interested in participating in the program at a price of 15 cents per day for Zerit and 85 cents per day for Videx.

The company has also allowed emergency patent relief on Zerit, the rights of the patent that it co-owns with Yale University. The two sides have agreed to enable the patent to be available at no cost to treat AIDS in South Africa. BMS also says it has no other patent rights in Africa that it will allow to prevent AIDS therapy there. John L. McGoldrick, Executive Vice-President of BMS said: 'This is not about profits and patents; it is about poverty and a devastating disease. We seek no profits on AIDS drugs in Africa and we will not let our patents be an obstacle.'

Value of generics set to grow as patents expire

Growth in the global market for generic pharmaceutical products will outstrip that of more expensive branded products over the next three years, according to a recent study conducted by Reuters, making it worth US$54 billion by 2004.

The high growth, predicted at 10–15% per year compared with 6–9% for branded products, will be caused by record numbers of patents expiring over the next couple of years. In the next five years, patents for 18 blockbuster drugs (worth US$37 billion in global sales) are due for expiry. One of these blockbusters, AstraZeneca's ulcer treatment Losec, was worth US$6.2 billion alone in global sales for the year 2000. In the US, the market for generics is predicted to reach US$20 billion in 2010, while strong growth in the underdeveloped markets of France and Spain will see the European market reach US$19 billion by 2004.

The cause of generic drugs might also be furthered by recent pressure in the US to have the Waxman-Hatch Act repealed, and moves in the EU to support a Roche-Bolar provision, where companies are permitted to begin preliminary testing on branded products before their patent expires. Consolidation of small companies, especially in Europe, and higher production of own-brand pharmaceuticals from former Soviet countries are also predicted.

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humans, the identification of which could help to dissect complex insulin signalling pathways.

2 Pierce, S.B. *et al*. (2001) Regulation of DAF-2 receptor signaling by human insulin and *ins-1*, a member of the unusually large and diverse *C. elegans* insulin gene family. *Genes Dev.* 15, 672–686

SIV-infected monkeys resistant to AIDS

African green monkeys do not develop AIDS despite having high levels of simian immunodeficiency virus (SIV) in their blood[3]. This finding by researchers at the Southwest Foundation for Biomedical Research (San Antonio, TX, USA) suggests that these primates could be an invaluable model for the study of natural host resistance to AIDS.

Reverse transcriptase-PCR was used to measure the levels and anatomical distribution of SIV in healthy, naturally infected monkeys. Infectious virus was isolated from plasma and peripheral blood mononuclear cells and was shown to be cytopathic in several human cell lines and macrophages. Distribution of the virus was found to be highest in the gastrointestinal tract, suggesting that this is a predominant site for viral replication. Interestingly, virus isolated from the lymph nodes was more restricted to growth in human T-cell lines, compared with virus distributed elsewhere, which readily infected macrophages in culture.

These studies demonstrate that natural host resistance to SIV is not the result of either effective control of viral replication or differences in the cell and tissue distribution compared with HIV infection, and challenges our understanding of how SIV, and its related human strain HIV, cause disease.

3 Broussard, S.R. *et al*. (2001) Simian immunodeficiency virus replicates to high levels in naturally infected African green monkeys without inducing immunologic or neurologic disease. *J. Virol.* 75, 2262–2275

Heteroclitic analogues of tumour antigens for cancer vaccines

An alternative approach to cancer vaccines was recently suggested at the *3rd Colloquium on Cancer Vaccines and Immunotherapy* in Abaco, Bahamas.

Regulatory affairs

Barr questions FDA stance on Generic and Paediatric Exclusivity

Barr Laboratories (Pomona, NY, USA) has asserted that a Food and Drug Administration (FDA; Rockville, MD, USA) response to a Congressional inquiry regarding the appropriate application of Paediatric Exclusivity and Generic Exclusivity statutes ignores their plain meaning and contains legally flawed assertions. The FDA's final interpretation of the interaction of these exclusivity statutes could have a bearing on Barr's recent Prozac patent challenge victory.

The letter, written by Melinda Plaisier (FDA Associate Commissioner for Legislation), said that the FDA had been 'unable to find any statutory language or legislative history' that would support that Pediatric Exclusivity and Generic Exclusivity 'run consecutively rather than concurrently'. Barr allege that this statement ignores the results of a previous Congressional inquiry (December, 2000) which informed the FDA that 'a proper interpretation of the two Acts together is that the language of the two provisions should read so that any Pediatric Exclusivity period which is granted does not run concurrently with a 180-day extension under Paragraph IV (Generic Exclusivity).'

'While this letter apparently expresses the views of some within the Agency [FDA], we doubt that its far reaching implications have received the full attention they deserve', said Bruce L. Downey of Barr Laboratories. While accepting that the letter is not an official ruling on their eligibility for exclusivity related to its successful Prozac patent challenge, Downey suggested 'any action by the FDA to undermine Generic Exclusivity would have an irrevocable chilling effect on the patent challenge process that results in the early introduction of generic versions of brand products.'

FDA warns Eli Lilly over manufacturing standards

Eli Lilly (Camp Hill, PA, USA) has been issued with a warning letter by the Food and Drug Administration (FDA) following a routine inspection of injectable-product manufacturing facility in Indianapolis. The inspection, a standard procedure for application for approval of their intramuscular formulation Zyprexa (olanzapine), found 'serious deviations from the Current Good Manufacturing Practice (CGMP) regulations (Title 21, Code of Federal Regulations, Parts 210 and 211), which cause your drug products to be adulterated within the meaning of Section 501 (a)(2)(B) of the Federal Food, Drug and Cosmetics Act.'

Vice-President of Manufacturing at Eli Lilly, Scott Canute responded: 'We are working in conjunction with the FDA – with a keen sense of urgency – to resolve all issues raised as a result of the FDA inspection.' Although the concerns are of the same type that sent the stock prices of Schering-Plough tumbling and ended in Federal Court settlements for Wyeth-Ayerst and Abbott Laboratories recently, Eli Lilly said it did not expect any material financial impact as it did not expect the timing of any new product launches or the manufacturing of existing lines to be affected.

Adverse reactions to Arava lead to warning being issued

The Committee for Proprietary Medicinal Products (scientific committee to the EMEA) has issued warning advice concerning the rheumatoid arthritis drug treatment Arava after it was revealed there have been 296 adverse reactions to the drug. Of these, 129 cases were considered serious, including two instances of liver cirrhosis and 15 cases of liver failure, with nine fatalities. The drug, manufactured by Aventis (Parsippany, NJ, USA), will continue to be available for the treatment of arthritis but its safety is to be monitored. Prescription and patient information has also been modified (see http://www.emea.eu.int/) and, in future, patients are to be told beforehand of its risks.

In response to the development, Aventis remarked that the number of problems was relatively low considering that approximately 200,000 people take Arava worldwide. It also commented that its drug is often taken simultaneously with other liver-threatening medications.

Epimmune (San Diego, CA, USA) is altering the natural tumour antigens to create a heteroclitic analogue, which can stimulate a potent response compared with the native antigen. Alessandro Sette, Vice-President and CSO of Epimmune claims that its data in animal models and *in vitro* using human cells shows that these analogues are up to 1000-fold more effective at activating T cells specific for tumour antigens than unaltered epitopes. However, heteroclitic analogues have previously been difficult to identify.

Sette says, 'While the importance of heteroclitic analogues has been recognized, their use has been hindered by the need to synthesize and test thousands of different types.' He says, 'Our new technology overcomes the need to screen large panels of randomly altered analogues and provides a highly efficient analogue identification process.' This technology uses a defined set of rules to alter specific amino acid sequences contained within epitopes of natural tumour antigens. These analogues are then tested for their ability to induce cytotoxic T cell responses, and then combined with native sequence epitopes from multiple tumour-associated antigens to maximize the number of T cells activated.

Miscellanous

New technology detects missed metastases

Novel diagnostic tests in Phase II trials have identified metastases in colon cancer patients that were not detected by conventional screening methods. The genetic tests, developed by the John Wayne Cancer Institute (JWCI, Santa Monica, CA, USA) and based on ORIGEN® technology (IGEN International, Gaithersburg, MD, USA), are potentially sensitive enough to detect 1–5 cancer cells among 10 million healthy cells.

Using a technique called 'sentinel node mapping', researchers at JWCI identified lymph nodes that were draining primary colorectal cancers (CRCs) and were, therefore, most likely to be invaded by micrometastases. Biopsy material from these nodes was analyzed using reverse transcriptase-PCR (rt-PCR)-analysis of three markers: β-chain human gonadotropin, hepatocyte growth factor and universal melanoma-associated antigen (uMAGE). In 12 of 26 patients tested, the ORIGEN rt-PCR-based technology detected micrometastases that were missed by conventional screening methods, and the expression of markers was consistent with that of the primary tumour[4]. Further, no micrometastases were detected in either cells from healthy individuals or lymph nodes from patients with benign tumours using the ORIGEN-based test.

In a second study, a test to detect several members of the MAGE family of cancer-related genes by rt-PCR was used to study blood from patients with melanoma, breast cancer and CRC. The test increased the identification of metastatic cells by 17%, 19% and 25% in the three cancers, respectively[5].

These tests have potential for use in the 'staging' of cancer, that is, the classification of a tumour based on its size and degree of metastasis. Earlier and more accurate staging of cancer should enable physicians to make better-informed decisions on cancer treatments, by identifying those patients who are more at risk of recurrence, and treating them more effectively.

4 Bilchik, A.J. *et al.* (2001) Molecular staging of early colon cancer of the basis of sentinel node analysis: a multicenter Phase II trial. *J. Clin. Oncol.* 19, 1128–1136

5 Miyashiro, I. *et al.* (2001) Molecular strategy for detecting metastatic cancers with use of multiple tumor-specific *MAGE-A* genes. *Clin. Chem.* 47, 505–512

Skills shortage hampers recruitment

A quarter of UK science organizations agree that the sector is experiencing a skills shortage across all disciplines, concluded a survey conducted by the Science Recruitment Group (SRG, Slough, UK) recently. The survey, questioning 200 such bodies between January and June 2000, revealed that while 25% of employers were looking to increase their science staffing levels, two-thirds expected problems finding enough graduates with relevant practical experience, even when their degree had included a year-long work placement. Scientific suppliers reported the highest shortfall, 47.4%, followed by pharmaceutical companies (35%) and clinical research organizations (35.7%). Pharmaceutical companies found problems across all areas of the industry including analysis, R&D, regulatory and formulation. Furthermore, 47% of biotechnology companies said that few graduates had the appropriate experience.

Brain drain

Compounding deficiencies in practical experience is the popularity of science graduates amongst general recruiters. Many are being drawn away to other industries such as IT by promises of higher pay and a faster career progression. A survey by IT recruitment company S.com goes some way to explaining why this is happening. A science graduate with two years of technical experience will earn 75% more for a comparable job in IT than if he or she had stayed in the science industry. 'Large pharmaceuticals' employers are not responding quickly enough to the skill shortage by positively reviewing salaries,' said Sylvia Kempsell of SRG.

Temporary staff

Incoming European legislation will soon bring rights for contract workers in the UK towards those for permanent staff. SRG has decided to offer its staff improved pay rates, sick and holiday pay entitlements and personal pensions ahead of this development. 'As a market leader in the Science Recruitment business we decided to invest in this initiative, at a cost to ourselves, to maintain our position in the market at no cost to client or members,' said Sylvia Kempsell.

GSK and WHO collaborate to complete development of new malaria treatment for sub-Saharan Africa

GlaxoSmithKline has signed an agreement with the World Health Organization to develop a new treatment for malaria called LAPDAP. The aim of the agreement is to develop LAPDAP (a combination of chlorproguanil and dapsone) as an effective oral treatment for uncomplicated malaria for use in areas of the world such as sub-Saharan Africa. Clinical trials in sub-Saharan Africa so far have shown that this therapy is effective for the treatment of malaria resistant to other standard first-line treatments such as chloroquine and sulfadoxine/pyrimethamine.

The Project Development Team for LAPDAP is being led by Peter Winstanley (University of Liverpool, UK), where the drug is currently in its final phase of development

and is anticipated to be available in some African countries as early as next year. Major contributors to this programme include the UK Department for International Development (DFID) and the University of Liverpool (UK) who devised the concept of LAPDAP. Gro Harlem Brundtland, Director General of the World Health Organization said: 'This is an important collaboration not only because it will bring a new drug to the market, but also because it includes a price structure that aims at making the drug affordable for those who need it.'

Impact of SNP genotyping could save millions by 2010

Pharmacogenomics could result in reductions in the cost of drug discovery and development of US$33 million per drug by 2010, claims a new report by Front Line Strategic Management Consulting (Foster City, CA, USA). Much of these savings would be made by employing SNP (single nucleotide polymorphism) genotyping to reduce clinical trial sample sizes and, subsequently, the overall length of clinical development. It predicts that the use of SNP genotyping will drive growth to a peak in 2005 when it will reach over US$1 billion in value.

A second major application for SNP analysis could be in the pre-screening of patients being considered for participation in clinical trials. Only using genetically 'suitable' patients could increase treatment efficacy by 32%, reported a separate survey by Front Line Strategic Management Consulting. Analysis of an already marketed drug suggested that the use of pharmacogenomics could have increased revenue from the drug by US$3.5 billion of the drug's lifetime (a 45% increase in revenue) due to decreased time in drug discovery and development. This precaution is also estimated by reducing the number of adverse drug reactions experienced by patients by as much as 25%, resulting in savings for the healthcare industry of a billion dollars per year by 2010.

Fund supports non-animal research

An anti-vivisection society, the Lord Dowding Fund (London, UK), is financially promoting several scientific and medical research programmes that have found non-animal ways of carrying out experiments that were previously carried out using animals. The Fund has awarded grants of nearly £2 million to researchers working in a wide range of fields.

Pain research

One such field to receive funding is in pain research. Strong evidence links hypersensitive gut nerves and abnormal brain processing with symptoms of pain in functional gut disorders such as irritable bowel syndrome and non-cardiac chest pain symptoms. However, analysing these results has previously been complicated by species differences in the results (e.g. using opossums, rats and primates) used in the research. Qasim Aziz (Hope Hospital, Salford, Cheshire, UK) and Paul Furlong (Aston University, Birmingham, UK) have developed a functional brain imaging technique to identify which areas of the brain are activated following both painful and non-painful stimulation of the gut in healthy people. The electrical field component of the brain's activity can be recorded by a technique know as Cortical Evoked Potentials, while the magnetic component can be recorded by Magnetoencephalography.

Hepatitis B

Nikolai Naoumov (Institute of Hepatology, University College London, UK) has developed an *in vitro* model of human liver cells infected with the hepatitis B virus that enables the immune (cytokine) response to be studied. This method, which has traditionally used ducks, woodchucks and transgenic mice, was validated and results confirmed by a parallel experiment using naturally infected human hepatocytes (liver cells).

Infertility

Christopher Barratt and Ian Brewis (University of Birmingham and Birmingham Women's Hospital, UK, respectively) are establishing and characterizing a human *in vitro* cell culture for the study of infertility caused by sperm dysfunction. Infertility research generally employs many animal specimens as more than half of the cells from which sperm cells derive die before maturity. The limited knowledge already available on mechanisms of apoptosis in sperm cells is based almost exclusively on rodent models, which are poor models for humans. It is hoped that by studying an *in vitro* culture system of the human seminiferous tubules, more might be learned about these mechanisms.

Cancers

Researchers at Sheffield University (UK) have created a bank of cultured liver cells developed from the excess healthy tissue from operations to remove tumours from the liver. The structure of the liver is such that whole sections have to be removed, not just the material that is cancerous. These cells will be used to investigate the interaction between liver endothelial cells and tumour cells. It is therefore hoped that this research will help in the understanding of how tumours spread from the colon to the liver, a common cause of death of patients with colon cancer. The cell culture will replace the animal cells and unsuitable human umbilical cord cells previously used in research into liver metastasis.

Traditionally mice have been used as model organisms in cancer research. Funding from the Lord Dowding fund has allowed Claire Lewis and colleagues at the University of Sheffield Medical School (UK) to develop a replacement novel *in vitro* assay to search for angiogenesis inhibitors for new potential anticancer drugs. The grant has also allowed the same team to isolate human endothelial cells (from blood vessels derived from surgically-removed tissues), analyse their protein expression *in vitro* and microscopically observe their activity in forming vessels *in vitro*.

New coalition to help fund stem cell research

A new coalition has been formed to ensure that federal funds are available for embryonic stem cell research. The Coalition for the Advancement of Medical Research (CAMR; Washington, WA, USA) comprises universities, scientific societies and voluntary health organizations. The goal of the coalition is to ensure that funding for stem cell research is retained and also to ensure that the public support for federal funding of stem cell research (found to be 65% in a recent public opinion poll in the US) is fed back to Congress. Founding members of this coalition include the American Society for Cell Biology, the Juvenile Diabetes Research Foundation International, Harvard University, University of Wisconsin and Washington University in St Louis.

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