launched such a course at Tel-Aviv University, which he hopes will stimulate other medical schools to follow. 'Pharmacogenomics must be incorporated within a few years... into the general MD curriculum,' he said.

The consequence of not doing so, he says, could be that any therapeutic benefits of the Human Genome Project will be severely delayed. The next generation of doctors will not have an adequate understanding of the interplay between genetics and drug metabolism, he warns.

But some experts point out that pharmacogenomics is only part of the problem. The lightning acceleration of genetics in the last decade has left the medical profession lagging behind on many fronts. 'Medical schools have been slow to take on board human genetics, as well as pharmacogenomics,' said Dan Nebert of the Cincinnati Children's Hospital Medical Center (http://www.cincinnatichildrens.org).

'The challenge in training physicians in pharmacogenomics is that there is probably inadequate attention to teaching clinical pharmacology in general, at least in US medical schools,' added Mark Ratain, Chairman of the Committee on Clinical Pharmacology and Pharmacogenetics at the University of Chicago (http://www.uchicago.edu).

Advanced pharmacogenomics

Physicians regularly practice rudimentary pharmacogenomics by asking patients for their family histories of drug sensitivity. More advanced DNA-based pharmacogenomics is already a feature of chemotherapy, where genetic analysis of cancer tissue helps oncologists choose a drug regime that will attack tumour cells most effectively.

Many genes have also been identified that have an effect on drug receptors and metabolism, says Katie Prickett, Commissioning Editor of *Pharmacogenomics*. 'CYP2D6 and apolipoprotein E status are just two of many other examples of genetic measures that could provide valuable information for drug prescribing,' she said. 'It's disappointing that genetic testing is not yet an accepted and routine part of the process in determining optimal and individualized pharmacotherapy.'

Personalized medicine is feasible, and is almost within reach, says Gurwitz, whose opinions are published in the March issue of *Trends in Pharmacological Sciences* [1]. The obstacle to further progress is the dearth of tools for rapid patient screening, he says.

'Hopefully, within 20 to 30 years, we shall be able to have genetic screening tools for choosing the most appropriate medicine for each patient, based on his or her genetics,' he said.

Reference

1 Gurwitz, D. *et al.* (2003) Education: teaching pharmacogenomics to prepare future physicians and researchers for personalized medicine. *Trends Pharmacol. Sci.* 24, 122–125

News in brief

Infectious diseases

Genetic puzzle of influenza solved

Scientists have recently solved a long-standing puzzle about how the influenza A virus assembles its genetic contents into infectious particles [1], which enables the virus to spread from cell to cell. The influenza A virus is the most virulent and potentially dangerous 'flu virus, including the strains responsible for pandemics, which kill millions or people worldwide. Scientists believe it is only a matter of time before another pandemic occurs, and therefore, these new findings about the pathogenicity of influenza A virus are crucial.

The influenza A virus genome comprises eight viral RNA segments. Although the products of all eight of these segments must be present for viral replication, little is known about the mechanism responsible for their incorporation into virions.

New research, from Yoshihiro Kawaoka's group of the University of Wisconsin-Madison School of Veterinary Medicine (http://www.uwm.edu/), has uncovered a molecular signal encoded by a single RNA strand that is crucial in the infection process. The signal recruits the necessary set of eight viral RNA strands to make a complete influenza genome within the infectious particles for the influenza A strain.

According to Kawaoka, the data indicate that the individual RNA segments each make a unique contribution towards the recruitment and assembly of disparate RNA fragments into a complete influenza genome, and therefore support the selective model of virion generation, in which specific structures in the individual RNA segments are responsible for combining the eight genetic fragments into a virion.

This insight into the genetic mechanisms that lead to 'flu infection provide a better basic understanding of how influenza and other viruses work and also has significant promise for new and better vaccines and drugs to combat the infection by exposing the genetic trick that influenza A virus uses to form infectious particles.

1 Fujii, Y. *et al.* (2003) Selective incorporation of influenza virus RNA segments into virions. *Proc. Natl. Acad. Sci. U. S. A.* 10.1073/pnas.0437772100 (http://www.pnas.org)

  PII: S1359-6446(03)02646-1

Antibacterial protein

A new antibiotic protein that kills certain types of bacteria in the gut has been discovered by researchers at Washington University School of Medicine (St Louis; http://medicine.wustl.edu) [2].

Lora V. Hooper, Instructor of Molecular Biology and Pharmacology and first author of the study, said: 'These findings were completely unexpected. We initially thought that this protein might be involved in blood vessel formation. What we discovered though, is that it is a potent killer of bacteria.'

Hooper, together with Jeffrey I. Gordon – the Dr Robert J. Glaser Distinguished University Professor and Head of Department of Molecular Biology and Pharmacology – who led the study, and co-workers, identified a protein called angiogenin (Ang4). This protein belongs to a class that was believed to be involved in blood vessel formation. However, in this study, the team found that Ang4 was released by specific cells – Paneth cells – located in the intestinal lining.

Paneth cells are known to assist in immunity by defending against infection, so the researchers examined Ang4 to determine its interaction with different microbes and found that it was selective for certain gut microbes. In particular, Ang4 production is controlled by the bacterium *Bacteroides thetaiotaomicron*, which lives in the gut. This makes Ang4 unique in that it is the first antibiotic protein whose expression is controlled by 'friendly' intestinal bacteria.

Hooper said, '…One of the functions of normal gut bacteria is to help erect an "electric fence" that protects the internal milieu from microbes [that] we encounter throughout our lives.'

Furthermore, the team found that other mouse and human angiogenins are also able to combat dangerous micro-organisms. 'These findings support the notion that the angiogenin family of proteins may represent a critical component of the body's innate defence system,' said Gordon.

2 Hooper, L.V. *et al.* (2003) Angiogenins: a new class of microbicidal proteins involved in innate immunity. *Nat. Immunol.* 4, 269–273

How *Streptococcus* avoids the immune response

The common bacterium, Group A *Streptococcus* (GAS), can evade phagocytosis and immune cells, and cause widespread human disease, such as pharyngitis and necrotizing fasciitis (flesh-eating syndrome). New research sheds light on how GAS evades destruction by the immune system. 'This is the first genome-scale look at GAS genes that are differentially expressed during interaction with the human innate immune system,' says lead investigator Frank DeLeo from the National Institute of Allergy and Infectious Diseases (http://www.niaid.nih.gov/default.htm).

The research examined the interaction between human white blood cells [polymorphonuclear phagocytes (PMNs)] and a type of GAS that causes abundant disease in North America and Western Europe. They found that GAS elicits its own genome-wide protective response to evade destruction by the human immune system [3]. GAS genes differentially regulated during phagocytic interaction with human PMNs comprise a global pathogen-protective response to innate immunity.

GAS prophage genes and genes involved in virulence, oxidative stress, cell-wall biosynthesis and gene regulation were upregulated and the resulting proteins were shown to be produced during GAS infection. The research highlighted an essential role for the Ihk–Irr two-component regulatory system in evading PMN-mediated killing and promoting cell lysis – processes that would facilitate GAS pathogenesis.

The group hope that this new knowledge in combination with earlier GAS data will inevitably spur the discovery of vaccine therapies and antibiotics that can prevent and treat different strains of this bacterium. 'We are excited about our findings and how they may lead to further investigation of therapeutics that can protect us from this major human pathogen,' said DeLeo.

3 Voyich, J.M. *et al.* (2003) Genome-wide protective response used by group A *Streptococcus* to evade destruction by human polymorphonuclear leukocytes. *Proc. Natl. Acad. Sci. U. S. A.* 10.1073/pnas.0337370100 (http://www.pnas.org)

As cunning as a virus

Herpes viruses can infect individuals for life, having the ability to hide from the host-cell immune system. Now, researchers have discovered how one herpes virus manages to escape detection – it destroys the molecules that would give it away, by turning host-cell proteins against one another [4].

Cytotoxic T cells patrol the body and eliminate cells that display viral peptides, which are presented at the cell surface thanks to major histocompatibility complex (MHC) class I molecules. Herpes viruses manage to dodge this mechanism, and researchers at the Washington University School of Medicine (http://medicine.wustl.edu/) have now worked out how.

Lybarger *et al.* looked at cells infected with γ_2-herpesvirus 68, which is closely related to the virus implicated in Kaposi's sarcoma – a cancer of blood vessels that affects some AIDS patients. In the infected cells, chaperone proteins that usually direct formation of MHC class I were joined by a viral protein, mK3. mK3 associated with, and modified, the chaperone proteins, altering their function. Instead of being transported to the cell surface, the resulting MHC class I molecules were destroyed, leaving nothing to flag the presence of the virus to cytotoxic T cells.

'These findings not only provide a better understanding of viral infections,' says Ted H. Hansen, a lead author of the study, 'they also offer novel insights into basic cellular processes in the immune system'. As commented by Lybarger, this 'represents a new strategy for blocking immune detection, and it suggests that there are probably other viral proteins that use host molecules to target MHC Class I'.

4 Lybarger, L. *et al.* (2003) Virus subversion of the MHC class I peptide-loading complex. *Immunity* 18, 121–130

Cancer

COX-2 regulator kills cancer cells

Researchers at Washington University School of Medicine (http://medschool.wustl.edu) have found a protein, cytidine uridine guanosine binding protein-2 (CUGBP2) that can destroy cancer cells [5]. The team identified CUGBP2 as a regulator in the production of cyclooxygenase-2 (COX-2), a key regulatory enzyme in the conversion of arachidonic acid to prostaglandins and other prostanoids. It is also known that the gene that produces COX-2 is switched on very early in cancer and that these prostaglandins bind to tumor cells and assist in turning on genes involved in angiogenesis.

Targets and mechanisms

Secrets from the deep

At this moment in time, tiny pumps in the fatty membranes that surround all our cells, are hard at work pumping charged ions, such as sodium, potassium and calcium, through the membrane. Until recently, the structure and function of these ion pumps have remained quite elusive. Now, researchers at The Rockefeller University (http://www.rockefeller.edu) have reported the use of palytoxin – a deadly toxin derived from coral – to elucidate the deepest secret of the ion pump: that it is just an elaborate version of an ion channel, which allows the ions to flow back down the electrochemical gradient and does not require the input of energy [10].

Lead author of the recent study, Pablo Artigas, said: 'The "pump channel" model is actually a very simple way to look at the function of ion pumps'. He added that this latest research has revealed that 'nature has once again figured out the simplest solution'.

David C. Gadsby, Head of the Laboratory of Cardiac and Membrane Physiology at Rockefeller and senior author of the study, said: 'Since the late 1950s, a handful of scientists have imagined pumps and channels as sharing some similarities, but this is the first time we've been able to establish this experimentally. By interfering with the pump's normal conformational changes, the coral toxin essentially turns them into channels.'

The researchers studied the Na^+K^+ pump, the most common of the human ion pumps, whose impaired activity is thought to result in high blood pressure. The closely related H^+K^+ pump controls the production of stomach acid and is the target of antacid drugs. Elucidating the molecular mechanisms of these pumps could pave the way for improved treatments for hypertension and heart failure and possibly other disorders.

10 Artigas, P. and Gadsby, D.C. (2003) Na^+K^+-pumpligands modulate gating of palytoxin-induced ion channels. *Proc. Natl. Acad. Sci. U. S. A.* 100, 501–505

Ceramide linked to type 2 diabetes

The precise link between increased body fat and the development of type 2 diabetes is not well understood. A recent study has shown that the saturated fat metabolite, ceramide, contributes to the development of insulin resistance in cultured cells and that excess accumulation of ceramide in the body could be the link between saturated fats and insulin resistance [11]. 'We speculated that saturated fats were not themselves responsible for antagonizing insulin action, but rather that some metabolite of the fats might themselves be responsible for blocking insulin effects,' says Jose Antonio Chavez, lead author of the paper.

These findings suggest that medication that prevents ceramide accumulation in body tissue could lessen or even prevent insulin resistance and result in a breakthrough in the treatment of type 2 diabetes. The research group will now try to replicate the results in animal models and if they are successful they can start to develop medications or therapies that target and prevent ceramide accumulation and potentially remove the need for many diabetics to take insulin.

Type 2 diabetes is the most common form of the disease, causing 90–95% of all cases in the USA and affects approximately 16 million people in the USA. It occurs typically in older people, but is also rapidly increasing in children, and accounts for almost 15% of all healthcare costs in the USA.

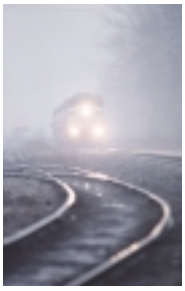
11 Chavez, J.A. *et al.* (2003) A role for ceramide, but not diacylglycerol, in the antagonism of insulin signal transduction by saturated fatty acids. *J. Biol. Chem.* 10.1074/jbc.M212307200 (epub ahead of print; http://www.jbc.org)

Principal investigator Shrikant Anant and his team found that the levels of CUGBP2 involved in the interaction between CUGBP2 and COX-2 mRNA were very low in eight types of human cancer cells. Anant explains that, 'this suggests that an important step in the development of cancer is turning down the gene responsible for production of CUGBP2, thereby reducing CUGBP2 protein levels and allowing the cancer to flourish'.

The study also reported that when CUGBP2 binds to COX-2 mRNA, it inhibits the production of COX2, resulting in tumour cell death. CUGBP2 was shown to be non-toxic to healthy cells, and restoring the level of CUGBP2 to that of normal cells caused cancer cell death, suggesting that CUGBP2 could be used to complement existing cancer therapies. The team is currently investigating whether the same effect is seen in animal models of cancer and they believe the strategy could be ready for human trials in a few years.

5 Mukhopadhyay, D. *et al.* (2003) Coupled mRNA stabilization and translational silencing of cyclooxygenase-2 by a novelRNA binding protein, CUGBP2. *Mol. Cell* 11, 113–126

Cancer: the runaway train



The function of a protein known to be involved in aberrant gene expression leading to the development of cancer has been elucidated by researchers from Saint Louis University (http://www.slu.edu) [6].

The modification of histones is important for the regulation of gene expression; to silence the expression of some genes in *Saccharomyces cerevisiae*, histone H3 must be methylated. The methylation of H3 is dependent on the ubiquitination of H2b, which is catalyzed by Rad6. Rad6 is involved in diverse biological processes and its activity in different processes is thought to be directed by different E3 enzymes, although until now the exact E3 enzyme that activates Rad6 during gene silencing was unknown.

By analyzing yeast gene deletion mutants, Ali Shilatifard (an Associate Professor of Biochemistry and Molecular Biology at Saint Louis University) and colleagues identified Bre1 as the E3 ligase

necessary for Rad6 activation resulting in H2b ubiquitination and H3 methylation. Using immunological techniques, including chromatin immunoprecipitation (ChIP), they also demonstrated that Bre1 is present with Rad6 in a complex, and they are recruited to a promoter together.

The elucidation of this molecular pathway has major implications in cancer research: 'Once we understand the normal, we will have a better understanding of where something is going wrong', said Ali Shilatifard, who describes a cancer cell as a runaway freight train: 'There may be a thousand ways to stop it. You can derail it, take all the screws from its wheels or stop giving it fuel. This is one strategy for stopping it.'

6 Wood, A. *et al.* (2003) Bre1, an e3 ubiquitin ligase required for recruitment and substrate selection of rad6 at a promoter. *Mol Cell.* 11, 267–274

MAArkers of melanoma vaccine efficacy

Melanoma is the fifth most common cancer in the USA and, if treated early, the cure rate is high. In the most advanced stage of the disease, however, the metastases spread to distant sites and are usually resistant to standard chemo- and radiotherapy. The five-year-survival rate amongst patients with advanced melanoma is 5%.

A certain subset of advanced melanoma patients (10%) survive longer than others, and researchers from the John Wayne Cancer Institute (JWCI; http://www.jwci.org) now think they know why. They took tumour samples from 35 late-stage melanoma patients, 30 of which then received immunotherapy with the cancer vaccine CANVAXIN™ (CancerVax Corp, http://www.cancervax.com). The tumour samples were analyzed for three immunogenic tumour markers known as melanoma-associated antigens (MAAs), which were contained in the cancer vaccine, and all patients were followed for up to five years.

The results indicated that the survival rate of patients with advanced melanoma could be predicted from tumour expression of the three MAAs (TYR, TRP-2 and MART-1) [7]. Patients whose tumours expressed lower levels of at least two of the MAAs assayed had a significantly worse prognosis than their counterparts, and patients with elevated expression for at least two of the MAAs had improved survival outcomes. It is thought that these antigens enhance the body's immune response to the vaccine, and as such could be used in the clinic to assess the potential success of any treatment. 'It is likely that melanoma cells with decreased expression or loss of MAA are of a selected aggressive phenotype with greater advantage to progress and escape host immunity, thus adversely affecting patients' overall survival', said Hoon, Director of Molecular Oncology at the JWCI and the study's principal investigator.

7 Takeuchi, H. *et al.* (2003) Expression of differentiation melanoma-associated antigen genes is associated with favorable disease outcome in advanced-stage melanomas. *Cancer Res.* 63, 441–448

Miscellaneous

A gene to remember

Researchers at the Emory Vaccine Research Center (http://www.emory.edu/WHSC/YERKES/VRC/) have shown that a gene called *SAP* is required to generate long-term immunity [8]. By measuring the immune responses of knockout mice that were genetically engineered to lack *SAP*, researchers found that the gene's absence impairs the immune system's 'memory', namely its ability to recognize and react to infection. This finding has implications for research on vaccines, which must engender long-term immunity.

The *SAP* gene has been identified as the genetic locus responsible for X-linked lymphoproliferative disease, a fatal immunodeficiency. Mutations in *SAP* have also been identified in some cases of severe common variable immunodeficiency disease, but the underlying cellular basis of this genetic disorder remains unclear.

In this study, the immune responses of *SAP* knockout mice were compared with those of genetically normal control mice. When infected with a virus, in this case lymphocytic choriomeningitis virus, both sets of mice mounted initial immune responses of similar magnitude. After the initial response subsided, however, the *SAP*-negative mice failed to produce significant numbers of either virus-specific plasma cells or memory B-cells, both of which are crucial components of long-term immunity. These cells are responsible for 'remembering' a particular virus and quickly rousing the immune system should it reappear. In SAP-negative mice the CD4+ T-cells apparently failed to stimulate the production of memory B-cells and plasma cells.

'What is so interesting about this gene is that it controls the generation of long-term memory, but it's not important for short-term immune responses. We haven't seen a gene that does this before,' said Rafi Ahmed, senior author of the study.

8 Crotty, S. *et al.* (2003) SAP is required for generating long-term humoral immunity. *Nature* 421, 282–287

Inheriting RNAi

The power of RNA interference (RNAi) has been extended, enabling the silencing of gene expression to be inherited in mice. The technique was developed by researchers at Stony Brook University (http://www.sunysb.edu) and Cold Spring Harbor Laboratory (http://www.cshl.org), and can be used as a complement to standard knockout methodologies in the study and treatment of many human diseases.

RNAi is a technique in which short hairpin sections of single-stranded RNA block cellular RNA by complementary binding. In a new application of the technology, the researchers genetically engineered mouse embryonic stem cells with RNAi targeted to the novel *Neil1* gene, which is implicated in DNA repair [9]. These stem cells were injected into mouse embryos, and the chimeric offspring were crossed to give mice whose cells all contained the RNAi-inducing gene. Expression of the gene was reduced by around 80% in all tissues, a phenomenon known as gene knockdown. The cells also showed a twofold increase in sensitivity to ionizing radiation, consistent with the proposed function of the gene product.

In future, the work could be adapted to selectively reduce or enhance expression in specific tissues. Such a genetic switch could have many applications in the treatment of disease.

9 Carmell, M.A. *et al.* (2003) Germline transmission of RNAi in mice. *Nat. Struct. Biol.* 10, 91–92

News in brief was written by *Matt Brown, Carrie Vicars, Joanne Clough, Morag Robertson, Linsey Stapley, Bryony Whisker, Catherine Wild and Heather Yeomans*