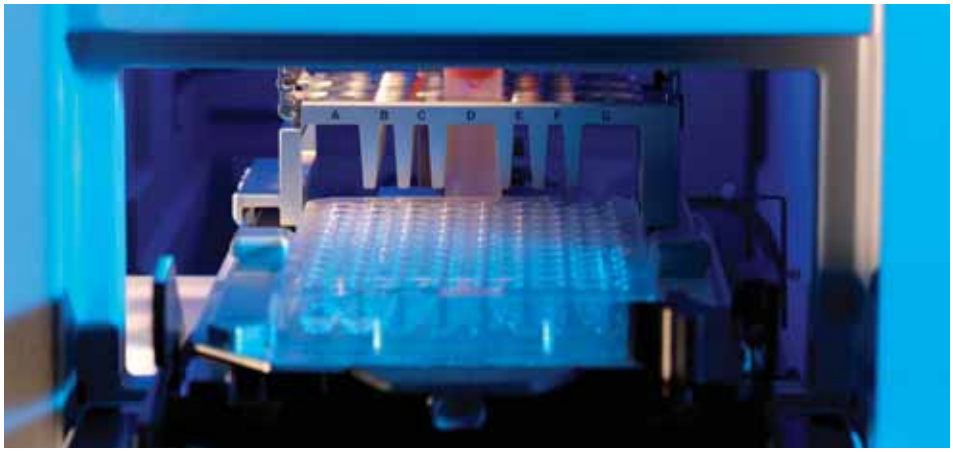
NPL
National Physical Laboratory

Health *Matters*

A National Measurement Newsletter

Spring 2008 | Issue 4



Inside:

Developing quality standards for biopharmaceuticals

NPL has teamed up with USP (United States Pharmacopeia) to improve the measurements of biopharmaceuticals, helping to ensure drugs are as effective and safe as possible.

Pharmacopoeial standards ensure the quality of medicines, including active and inactive components used to prepare a pharmaceutical product. Bodies such as the United States Pharmacopeia (USP - in Washington DC) and the European Pharmacopoeia (EP) set these standards and are key partners in the global infrastructure for controlling medicines, by complementing the licensing and inspection processes. Medicines sold must comply with these quality standards so that consumers have a guarantee for the quality of products obtained from pharmacies.

The attachment of sugar molecules to proteins (glycosylation) can affect the way the proteins interact with the human body. At present there are no general standard procedures in place in the pharmacopoeias for assessing protein glycosylation in biopharmaceutical drugs, only product specific ones. Characterisation of biopharmaceutical glycosylation (glycan analysis) is important, as incorrect glycosylation can affect the half-life and efficacy of a drug, and even lead to a toxic response. An increasing number of glycosylated biopharmaceutical products, such as antibodies, are also entering the market, hence the need for regulating their release. USP and EP have joined together to produce pharmacopoeial standards, with the help of an advisory panel, made up of representatives from NPL, LGC and the National Institute for Biological Standards and Control (NIBSC) from the UK. Four key chapters in the area of characterising protein glycosylation are now being finalised for submission later in 2008.

In addition to the written standards, an international inter-laboratory study was recently undertaken involving over 30 laboratories, to assess comparability of glycan analysis across the large range of techniques available to industry. Data from the study is currently being processed by USP, NIBSC and NPL, and it is planned that this data will inform the writing of the four chapters for the pharmacopoeias.

For further information, please contact Smita Thobhani on 020 8943 6684 or e-mail smita.thobhani@npl.co.uk

National Physical Laboratory | Hampton Road | Teddington | Middlesex | United Kingdom | TW11 0LW
Switchboard 020 8977 3222 | **NPL Helpline** 020 8943 6880 | **Fax** 020 8614 0446 | **www.npl.co.uk**

Department for **Innovation, Universities** & **Skills**

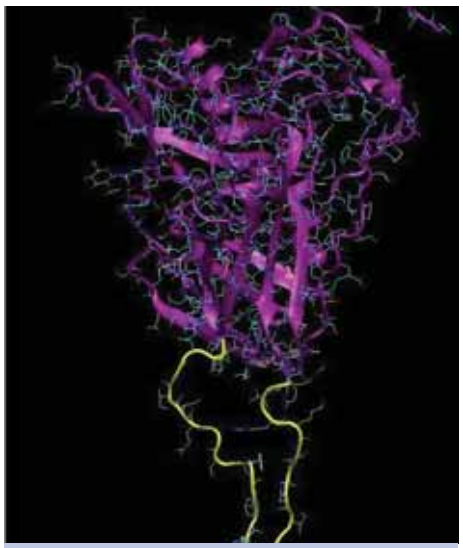
Computer fights virus: IBM's supercomputer simulates HIV fragment

NPL, IBM and the University of Edinburgh are using some of the most powerful computers in the world to help fight HIV. This collaborative project has simulated the structure of an HIV glycoprotein fragment involved in the infection of target cells, a step that will aid the identification of potential HIV inhibiting drugs.

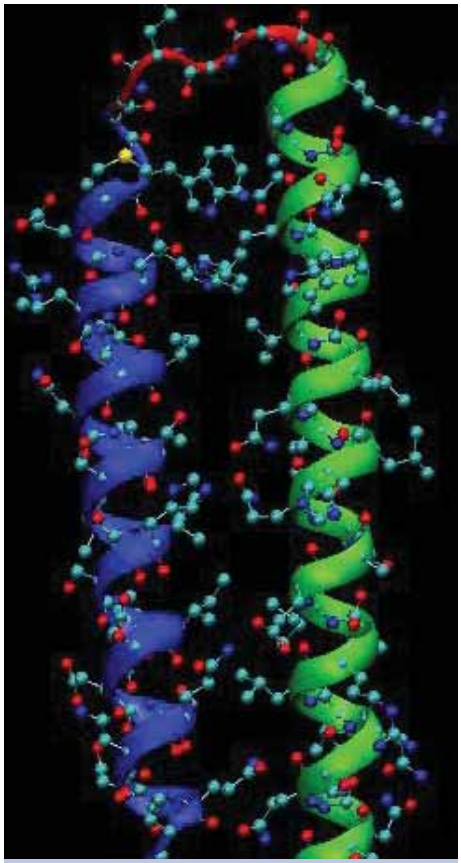
The majority of current HIV therapies are designed to treat patients who have already been infected by the virus. A new joint project between NPL, IBM and the University of Edinburgh aims to contribute to the understanding of the mechanism of virus entry. This knowledge is pivotal for developing drugs that will inhibit the fusion of HIV with cells. In particular a fragment of an HIV-1 glycoprotein is currently the target of research into HIV fusion inhibition.

In a recent collaborative project IBM's supercomputer 'Blue Gene' was able to produce molecular-scale simulations of this glycoprotein fragment, which is thought to be part of the infection process. Without the computational power of 'Blue Gene' these simulations would not have been possible on the molecular-scale. With 'Blue Gene', the sampling of the complex energy landscape of this molecule was improved.

The validation of a simulated model by experimental results is a required step in the establishment of reliable algorithms. The results from the simulations were compared with existing and newly acquired experimental outputs. In particular, circular dichroism spectroscopy measurements, performed and analysed at NPL, helped to elucidate the average content of secondary structure elements (helices, turns, sheets and other structural motifs) in the glycoprotein's peptide.

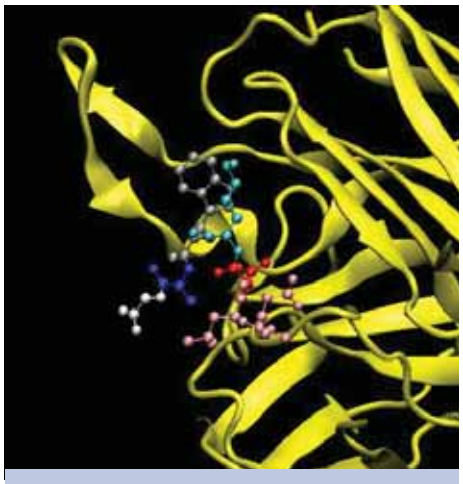


Tertiary structure of HIV glycoprotein gp120, showing the variable (V3) loop region in yellow.
Image supplied courtesy of IBM



A region of interest of HIV-1 glycoprotein.
Image supplied courtesy of IBM

Understanding the structure and behaviour of such surface proteins is a crucial step to developing drugs rationally, efficiently and rapidly; it is the first step towards identifying which drug molecules may be able to prevent the virus from infecting its target cells. This allows candidate molecules to be cherry-picked from very large drug libraries, accelerating the research by lowering the numbers of different molecules that need to be synthesised and tested in laboratory trials.



Human antibody molecule (shown in yellow) bound to a small segment of the HIV viral protein (shown in white, blue, grey, turquoise, red and pink).
Image supplied courtesy of IBM

"NPL provides the experimental validation that ultra large scale computer modelling requires, both to test theories and interpret results"

Dr. Eleonora Cerasoli,
Senior Research Scientist,
NPL Biotechnology Group

By using this supercomputer to model the structure of a glycoprotein that may be essential to the infection process, and validating the data generated with results achieved in the laboratory, researchers hope to speed up the discovery process for HIV inhibiting drugs. Researchers from IBM are now writing new algorithms to simulate the structure of drug molecules and predict how they will interact with the HIV-1 glycoprotein.

For further information, please contact Jason Crain on 020 8943 6107 or e-mail jason.crain@npl.co.uk

Single molecule genotyping

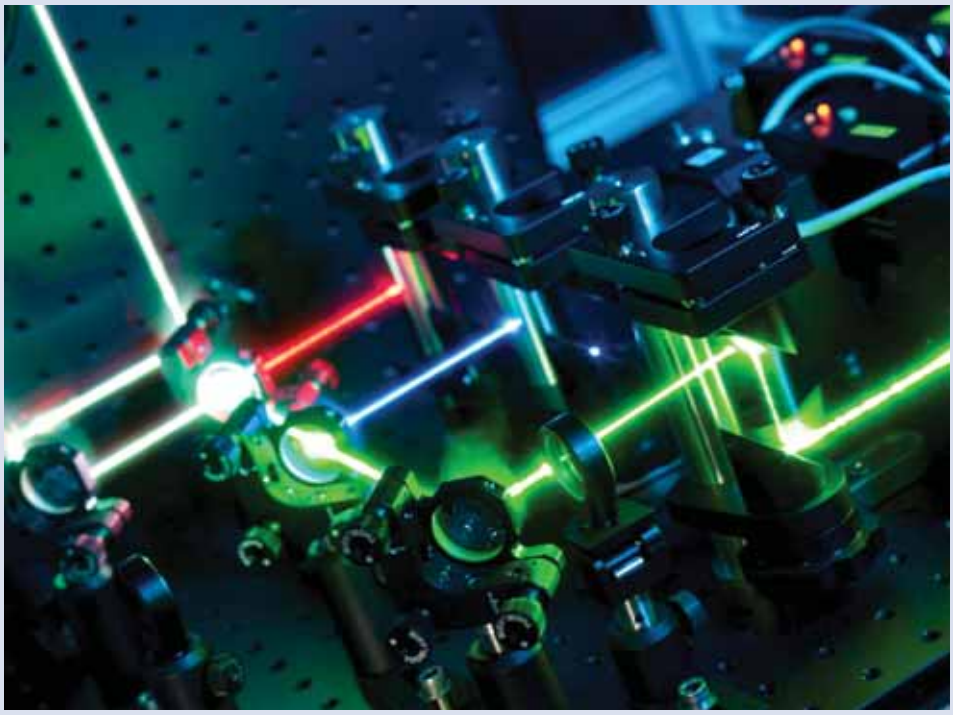
NPL has developed a new technology for measuring the frequencies of genetic variations in complex samples.

The recent completion of the human genome has given a complete "atlas" of human genes. However, it is only the genome of one, or a few, individuals. We know that in fact one in every thousand bases in the human genome (which contains billions of bases) is a site of variation between individuals. Most of these variations take the form of a change to a single base pair; e.g. a C (cytosine base) becomes a T (thymine base); these are known as Single Nucleotide Polymorphisms (SNPs or 'snips'). If such a change occurs in the coding or control regions of a gene it may cause a disease, increase the risk of developing a disease, or affect the way a particular drug is processed by the body. If it lies elsewhere, it may be genetically linked to another mutation that has such an effect. It is therefore extremely important to identify these SNPs; to determine which are correlated with disease; and to test individuals for particular SNPs that may influence their health or treatment.

The NPL technology (the single molecule imaging apparatus) relates to statistical genotyping, an approach to identifying correlations between SNPs and particular conditions. In statistical genotyping, pools of DNA from many individuals are compared and the frequencies of SNPs are measured. For example, one pool might be from healthy individuals and the other from disease sufferers. An SNP correlated with the disease will be more common in the diseased pool than in the healthy pool. The technology could also be applied to other scenarios where the frequency of a single base mutation is being monitored. For example, monitoring the frequency of a drug resistance mutation in virus samples from a patient, or a virulence mutation in isolates of influenza virus.

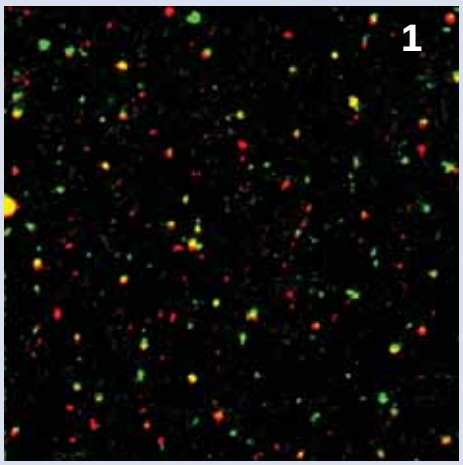
The approach consists of a labelling step followed by single molecule imaging to count the number of molecules of each genotype present in the sample - the single molecule imaging apparatus is based on total internal reflection fluorescence microscopy. By combining this with microarray technology, high throughput measurement of many alleles* will be possible.

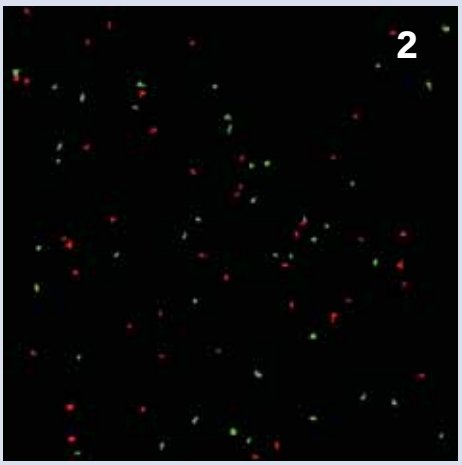
For further information, please contact Alex Knight on 020 8943 6308 or e-mail alex.knight@npl.co.uk



** Allele: one of two (or more) possible sequences of DNA found at the same position on a chromosome*

Top: Single molecule imaging apparatus based on Total Internal Reflection Fluorescence





Far left: 1) Single molecule fluorescence images of labelled DNA molecules. In this experiment, the green fluorescent molecules have one allele of the SNP (G) and the red molecules have the other (A)

Left: 2) Image processing is used to analyse the image in (1) and count the molecules of each allele. In this example, 41 green (G) and 43 red (A) objects were counted. Therefore the relative frequency of the G allele is estimated as 49%

Fidelis secondary standard radionuclide calibrator with dedicated laptop

Fidelis: Faithful radionuclide calibrations for nuclear medicine

NPL and Southern Scientific have developed 'Fidelis' - the latest generation of the NPL radionuclide calibrator. The calibrator has been developed to meet the exacting requirements of nuclear medicine departments.

Nuclear medicine is a growing field due to advances in cancer treatment and diagnostic imaging. Around 700,000 patients are injected with radioactive compounds each year in the UK. A radionuclide calibrator is used to measure the activity of every compound prior to administration to the patient. A calibrator consists of a "well-type" cylindrical ionisation chamber and an electrometer. The radioactive source is placed in the chamber well and produces a steady ionisation current across the chamber. The magnitude of the current produced is proportional to the source activity.

NPL and Southern Scientific recently developed a new generation of the calibrator. Its user-friendly computational functions and fully automatic self-testing system make it ideally suited to the needs of nuclear medicine. Each 'Fidelis' chamber is identical to the master chamber held at NPL (developed and designed by NPL). Each system is tested and calibrated against the NPL master system, so that every system is fully traceable to the UK national standards.

Data processing is performed on a dedicated laptop computer. The software includes a library of calibration factors for over 60 radionuclides commonly used in nuclear medicine. Calibration factors for additional radionuclides can easily be added. The software has been developed with reference to the requirements of the Institute of Physics and Engineering in Medicine (IPEM)/NPL good practice guide for radionuclide calibrators*. Provision has been made for regular quality assurance tests, such as daily system checks and linearity testing.

The 'Fidelis' system has been fully tested at NPL and is available from Southern Scientific. Upgrades are available for older systems.

*IPEM/NPL Measurement Good Practice Guide No. 93, *Protocol for establishing and maintaining the calibration of medical radionuclide calibrators and their quality control,* is available from NPL. To download a copy, please visit **www.npl.co.uk/guides** or for a hard copy please contact **health@npl.co.uk**

For further information, please contact Eleanor Bakhshandeiar on 020 8943 8586 or e-mail eleanor.bakhshandeiar@npl.co.uk

For sales information e-mail Southern Scientific Ltd at info@ssl.gb.com or visit www.ssl.gb.com

Radiation Dosimetry's new linear accelerator is developing fast

The new NPL clinical linac facility will conform to the National Radiotherapy Advisory Group's recent recommendations on 4D adaptive radiotherapy. The new facility will be able to calibrate the full range of energies currently in therapeutic use in the UK.

The construction of the £4m clinical linac (linear accelerator) facility, which commenced in April 2007, is on schedule, with delivery of the linac machine programmed for 10 June 2008. The linac, which will be a state-of-the-art device supplied by Elekta, is a Synergy Digital Linac with iViewGT portal imaging, MOSAIQ management system, PINNACLE 3D treatment planning system and Synergy XVI 3D x-ray volumetric imaging. This specification conforms to the recent recommendations of the National Radiotherapy Advisory Group that all new and replacement machines in UK hospitals be capable of image guided 4D adaptive radiotherapy, which can target tumours more accurately.

The NPL machine will be able to deliver seven x-ray beam energies (instead of the usual maximum of three in any one hospital machine). This feature, together with the ability to provide nine electron beam energies, will enable NPL to calibrate the full range of energies currently in therapeutic use in the UK.

Commissioning of the machine will take 12 weeks following installation on site, somewhat longer than usual because of the exceptionally wide range of x-ray beam energies being employed. Once commissioned, NPL's reference dosimetry services will be re-established in the new facility. In early Spring 2009, NPL is planning on using the new linac to provide photon and electron therapy level calibrations for customers. The session in the 'NPL Practical Course in Reference Dosimetry' covering the electron code of practice will be delivered in the new facility, early in 2009. Further new projects in the National Measurement System 2007-2010 Acoustics and Ionising Radiation Programme will develop dosimetry for small fields and IMRT (Intensity Modulated Radiation Therapy), including a new absorbed dose calorimeter specifically designed for these fields.

For further information please contact Rebecca Nutbrown on 020 8943 6473 or e-mail rebecca.nutbrown@npl.co.uk

The new linac building, construction nearly complete

Revision of standards on reference neutron fields

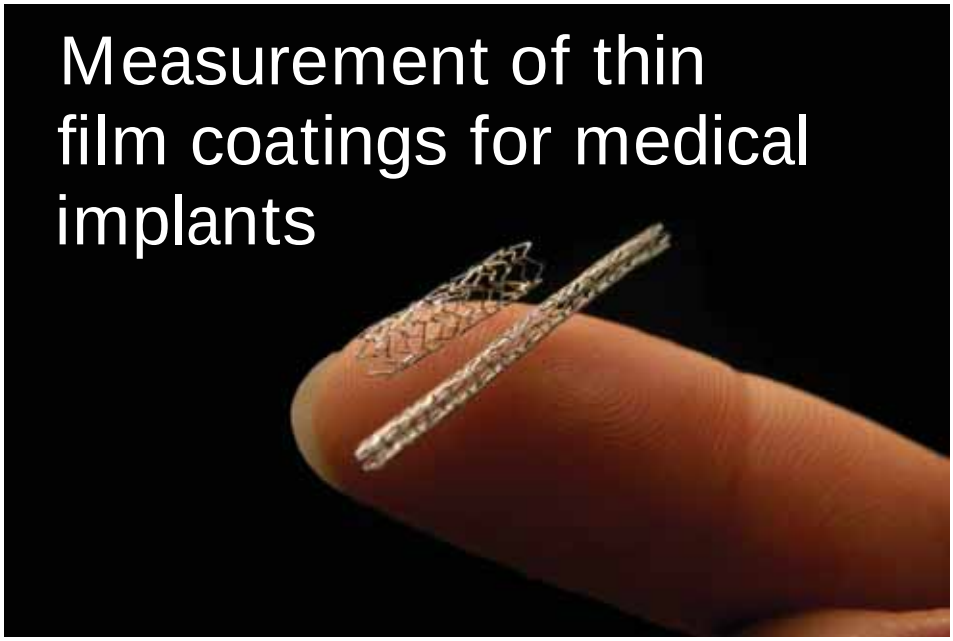
The working group that develops international standards on reference radiation fields, for calibrating radiation sensitive devices, is revising the standards for neutron fields (ISO 8529 Parts 1 to 3). This working group welcomes feedback from users of these standards on issues that need to be addressed in this revision.

Technical Committee 85 (TC85) of the International Organization for Standardization (ISO) is concerned with nuclear energy, and has a sub-committee (SC2) that is concerned with radiation protection. This sub-committee in turn has a working group (WG2) that develops standards on reference radiation, i.e. the types of fields to be used for calibration of devices that are sensitive to various types of radiation, including beta rays, x-rays, gamma rays and neutrons.

WG2 has just begun a revision of the series of international standards that deal with reference fields for neutrons - ISO 8529 Parts 1 to 3. The three parts of the standards cover the types of fields to be used, the methods available for characterising these fields, and the techniques to be used for performing calibrations. Several issues have already been identified as being in need of attention, including the possible variation of radionuclide source spectra between calibration laboratories, due to differences in source construction. The revision group would be interested to hear from users of these standards, in regards to any errors in these documents and any other issues that they feel need to be addressed in this revision.

For further information please contact David Thomas on 020 8943 6853 or e-mail david.thomas@npl.co.uk

Measurement of thin film coatings for medical implants



Left: Two stents for coronary arteries. Lower stent is unexpanded, as it would be for insertion; top stent is expanded, as it would be after insertion. Polymer coatings containing anti-rejection drugs are used on stents, to lower the likelihood of rejection by the recipient

NPL has developed transfer artefacts suitable for the verification of a system used to measure the thickness of polymer films on medical implants. This will enable better control of the release of anti-rejection drugs used on such implants.

There is a large market for implantable medical devices such as cardiac stents and prosthetic orthopaedic joints, but all these implants are subject to varying degrees of rejection by their hosts. To reduce rejection, it is common for these implants to be coated with a polymer that contains an anti-rejection drug. The total dose and dosage rate can be controlled by the thickness of the polymer coating, and the concentration of the drug. Control and measurement of the coating thickness is, therefore, very important, but is currently difficult to achieve accurately and within the production process.

Nightingale-EOS has developed a bench-top instrument capable of measuring the thickness of these polymer coatings, using beam profile reflectometry (BPR). This measurement technique has been developed from an established technology used to measure planar films, which has been adapted to work with highly curved surfaces - such as those present on stents.

Work has been carried out to develop and assess the BPR's capability and accuracy when measuring different thicknesses of polymer films. Polymer samples were developed at NPL, along with Lombard Medical, as transfer artefacts to validate the BPR when measuring curved surfaces. A range of equipment at NPL was used to measure the artefacts accurately. Results from AFM (atomic force microscope) measurements, scanning white light interferometry and three-dimensional optical microscopes were compared to those of the BPR, to demonstrate the effectiveness of this measurement technique and its application to curved surfaces.

For further information, please contact Laurence Brice on 020 8943 7054 or e-mail laurence.brice@npl.co.uk

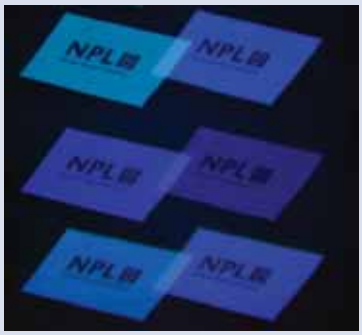
Keep your eyes out for Optical Technologies and Measurement Network events

The Optical Technologies and Measurement Network (OTMN) aims to bring together all those with an interest in optical radiation measurement, creating unique opportunities for networking and discussion of ideas, to address measurement issues that fall across industrial sectors.

The OTMN membership includes equipment developers and users, industrial companies ranging from the smallest specialist manufacturers to large multi-national companies, hospital medical physics departments, and universities.

These meetings are open to OTMN members free of charge and to non-members on payment of a registration fee of £130 (one-day meeting).

Non-members who would like to join the network and benefit from free admission to OTMN meetings, please e-mail **Fiona Jones** at **npl_clubs@npl.co.uk.** For more information about the network and membership, please visit **www.npl.co.uk/otmn**



Spatially separated images of a single NPL logo, each containing information from a different wavelength. Produced by IRIS (Image Replication Imaging Spectrometer); this technology has potential applications in ophthalmology and keyhole surgery

Mathematical representation of sparse clinical data

In collaboration with the Royal Marsden Hospital Medical School and the Royal Free and University College Medical School, NPL has carried out research into the analysis of clinical data with low numbers of data points and large uncertainties. An approach to the analysis of such data has now been successfully trialled with 30 sets of patients.

Clinical data presents challenges in its analysis, since it is often sparse (perhaps only six or eight data points), and has large standard uncertainties associated with it (usually between 10% and 20%).

Such data arises when monitoring a cancer patient by taking a time sequence of response values – from the level of a relevant enzyme or activity in serum. The response is to the initial administration of a drug – a radiopharmaceutical or a fusion protein consisting of a tumour-targeting antibody linked to an enzyme product.

Reliable analysis of this data can address three requirements of clinical drug administration and related research: the quantification of the biological decay processes specific to each patient concerned, a measure of the total absorbed dose, and the prediction of the optimal time for a further stage of drug administration.

Obtaining estimates of such parameters and the associated uncertainties requires a representation of the data in the form of a suitable mathematical model, and the propagation of the uncertainties associated with the response values through this modelling process. NPL has carried out research on both these aspects. The mathematical functions used to represent the biological decay processes constitute an age-old set of functions (a combination of exponentials with unknown parameters), but with three important differences: the parameters take biologically feasible values, the model is statistically consistent with the data, and the number of terms in the model is minimal.

Another feature of the approach adopted was obtaining a sufficiently good approximation to the parameters that, when the model is matched to the data, there is good assurance that the global solution is obtained.

The approach has been trialled satisfactorily on over 30 sets of patient data, provided by collaborators at the Royal Marsden Hospital Medical School and the Royal Free and University College Medical School.

For further information, please contact Maurice Cox on 020 8943 6096 or e-mail maurice.cox@npl.co.uk

The following OTMN meetings are of particular relevance to the Health Matters readership:

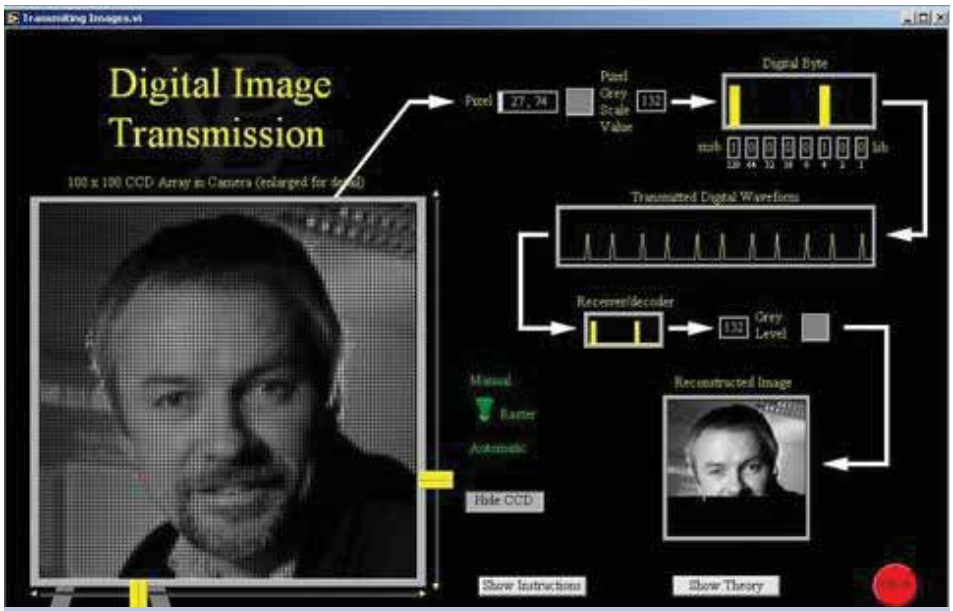
25 & 26 June 2008	***Annual OTM Network meeting*** To include "Image Capture, Reproduction and Recognition". Other topics to be announced Organiser: Mrs Fiona Jones, OTM Network Manager
9 October 2008	***"Lighting: Energy Efficiency, Health, Well-Being and Performance"*** Organiser: Dr David Loe (Consultant), Chair of the Lighting Working Group This follows on from a three-day practical Photometry & Spectroradiometry training course. For further information, please visit **www.npl.co.uk/photometrycourse**

In addition, the following meetings are planned:

17 July 2008	***FOTON – Fibre Optics Network meeting*** Organiser: Dr Neil Haigh (NWDA), Chair of FOTON WG
24 September 2008	***"Radiometric Measurement of Detectors"*** Organiser: Dr Moira Hilton (University of Reading), Chair of the Optical Standards and Traceability Theme

If you would like further information on any aspect of **Health Matters,** *please contact:*
Tel: 020 8943 6880 | **Fax:** 020 8943 7160 | **E-mail:** health@npl.co.uk

Quality of Life Division
National Physical Laboratory | Teddington | Middlesex | United Kingdom | TW11 0LW
Helpline: 020 8943 6880 | **Fax:** 020 8614 0446 | **E-mail:** enquiry@npl.co.uk



VPLab simulation showing how an image is transmitted in digital format

Physics teachers given virtual assistance

Practical experiments in physics classes take time and money to set up, and require teachers to be vigilant that pupils do not break expensive equipment. The Virtual Physical Laboratory is a piece of software designed to give students greater access to interactive experiments, without a greater cost.

The Virtual Physical Laboratory (VPLab) software is a set of over 200 interactive experiment simulations, which cover the majority of subjects that make up the Physics GCSE, AS and A2 syllabuses. This user-friendly package has proved to be very popular with teachers and pupils, who are able to use these experiments swiftly and cheaply, without the time and cost of equipment set up. Pupils can investigate these experiments according to their abilities, and at their own pace, without the fear of breaking something.

The topics covered include optics, radioactivity, x-rays, sound and waves, amongst many others. The package is very straightforward, and can be used as a demonstrator by teachers or individually by pupils. Written by John Nunn (NPL), schools and colleges in the UK and Republic of Ireland can obtain VPLab free of charge when teachers attend a demonstration, thanks to sponsorship from NPL and IOP (Institute of Physics). VPLab may be installed on the computer server in the school of the recipient, to allow multi-user access, but it should be noted that VPLab is not 'freeware' and must not be passed on to third parties.

> "I was astounded at how useful it was. Awarded 5-star = Excellent"
> *Bernard Taylor,*
> *Physics Education Magazine*

NPL and IOP are able to attend teachers events to provide these demonstrations.

To find out dates and other information about booking demonstrations, please e-mail the IOP organiser Gary Williams at gary.williams@iop.org

If you are outside of the UK and Republic of Ireland, you can purchase the software from www.vplab.co.uk

Forthcoming events

www.npl.co.uk/events

Humidity Measurement & Calibration Training Course
9 – 10 June 2008
NPL, Teddington
www.npl.co.uk/humiditytrainingcourse2008

Micro and Nano Scale Characterisation of Fibres
3 July 2008
University of Ulster, Belfast
http://www.npl.co.uk/server.php?show=ConWebDoc.2440

Nano-Molecular Analysis for Emerging Technologies III
5 – 6 November 2008
NPL, Teddington
http://conferences.npl.co.uk/nmaet

7521/SEA/2k/0508