

Micro and Nano Technology (MNT) Measurement Club

The Measurement and Characterisation of Medical Biosensors

Venue: National Physical Laboratory, Hampton Road, Teddington, Middlesex TW11 0LW

Date: 9 December 2005

Overview of Meeting

The aim of this meeting is to bring together researchers and developers of biosensor techniques to review and discuss the measurement and characterisation requirements and issues in developing robust and reliable biosensor systems for medical applications. It will include presentations describing biosensor techniques that have made a significant impact in the point of care market and the measurement challenges that lie ahead in these application areas, recent advances in medical biosensors and associated analytical techniques, and current research within the DTI's National Measurement System (NMS) Programmes supporting biosensor development and application. The meeting will also include poster presentations showing some of the latest medical biosensor related research being carried out in UK universities and other organisations.

PROGRAMME

09.30 – 10.15 REGISTRATION, COFFEE and poster session

Chair: Julie Deacon, Assistant Director, MNT Network

10.15 – 10.20 *Introduction to meeting* - Julie Deacon

10.20 – 10.45 *Blood glucose biosensors* - Jeff Newman - Cranfield University, Institute of Bioscience and Technology

10.45 – 11.10 *Medical biosensors in women's health* – Robert Porter - Unipath

11.10 – 11.30 *Sonochemically fabricated micro-electrode array based biosensors* - Séamus Higson - Cranfield University, Institute of Bioscience and Technology

11.30 – 11.50 *Standardisation of protein diagnostics measurements* – Alex Knight – National Physical Laboratory

11.50 – 12.10 *Dual Polarisation Interferometry: Molecular measurement in the life sciences*
- Marcus Swann, Fairfield Sensors

12.10 – 12.25 *Modelling and calibrating an optical biosensor* – Richard Dudley – National Physical Laboratory

12.25 – 13.45 LUNCH and poster session

13.45 – 14.05 *MultiSense dry enzyme system for simultaneous multiple parameter measurements* - John Broughall - Oxford BioSensors

14.05 – 14.25 *Modification of Nanopore proteins for single molecule detection* — Gordon Sanghera - Oxford NanoLabs

14.25 – 14.45 *Materials selection for cell based sensors* – Paul Tomlins – National Physical Laboratory

14.45 – 15.05 *Reliable interfacing of biosensors* - Pankaj Vadgama - Queen Mary, Univ. London

15.05 – 15.35 COFFEE and poster session

15.35 – 15.55 *Resonant acoustic profiling* - Matthew Cooper - Akubio

15.55 – 16.15 *Magnetic biosensors* – Richard Luxton – University of West England

16.15 – 16.35 *Terahertz biosensors* – Richard Dudley – National Physical Laboratory

16.35 – MEETING CLOSE

Presentation Abstracts and Speaker Profiles

Chairperson - Dr Julie Deacon, Assistant Director, MNT Network

Julie Deacon is a specialist in diagnostic, pharmaceutical and bio-defence markets with experience of a broad client base throughout USA, Japan and Europe. She has extensive industrial experience in technology validation, development and commercialisation, IP generation and market assessment. She is an expert in the development of systems incorporating biosensors, fluidics, optical systems, assay signal enhancement and surface plasmon resonance primarily for Pharmaceutical and Diagnostics applications. Her current role within the MNT Network is to represent leading UK Bio Nanotechnologies initiatives, to develop strategic initiatives for the industrial applications of NanoMedicine, to support facilities for MNT fabrication and to catalyse international MNT inward investments. She sits on the MNT executive panel and has recently contributed to the ESF Forward Look in Nanomedicine.

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Blood glucose biosensors

It is tempting, these days, to forget about the problems encountered when blood glucose biosensors were first being commercialised. Despite the fact that the basic principle was made public in 1962, no commercial device emerged successfully until 1975, and it was not until 1987 that a home-use product was launched. The market was established and growing. Sales of reflectance-based devices were booming, despite a number of shortcomings with this technology. Laboratories all over the world had demonstrated the advantages of biosensors, but a route to commercialisation was elusive.

So, why did it take so long for biosensors to reach such a key market? As with many innovations, it took a number of key breakthroughs to lead to a successful product. These ranged from fundamental issues concerning the measurement technique to materials technology and mass production techniques. There was, and still is, an issue concerning the measurement site, medium, frequency, sample volume and how this relates to the glycaemic control of the patient.

This presentation will examine these issues and, hopefully, see if there is anything we can we learn from this case study.

Speaker Profile - Dr J.D. Newman, Institute of Bioscience and Technology, Cranfield University

Originally from a biochemical engineering background, Jeff has over twenty years of experience in the biosensor field. Working for Unilever Research, UWE in Bristol and, for the last fifteen years at Cranfield, he has been involved in sensors for medical, environmental, defence, food and industrial applications. Advanced fabrication technologies have featured throughout his career and have led to an interest in microfluidics and a role as Cranfield's representative in the European Liquid Handling Competence Centre (LICOM). In recent years, Jeff has concentrated on developing a consultancy business within IBST. Much of this work involves working directly for companies, but it includes writing the biannual biosensor reports, which are produced for commercial sale.

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Medical biosensors in women's health

Within diagnostic there are only two diagnostic products performed in the home environment. One being glucose sensing, the other, in the form of lateral flow assays, utilised in the women health business. The talk will focus on the attribute micro and nano section of an immuno-diagnostic system utilised within a women's health product.

The talk will look at how with the correct material and structure one can improve sensitivity and specificity in an assay. We will look at how the assay is structured and performed within a non-trained user's hand.

Speaker Profile - Dr Robert Porter Unipath Ltd UK Inverness Medical Inc

I first worked at Unilever Research based at Colworth House in Bedfordshire where I was responsible for diagnostic research areas for food and medical products. Unipath was then a Unilever Company focussed on medical products in the area of infectious diseases and Women's Health, with products such as Clearblue and Clearview (home and professional versions) – the world leading pregnancy and fertility testing. My role was moved to Unipath 2000 as part of the sale to Inverness Medical Innovations where we are now at their major centre of research operations. The new main areas for Inverness Medical are in the areas of drugs of abuse and cardiology monitoring. At Unipath I was responsible for the areas of novel diagnostic technologies and the development of electrochemical immunosensors. Now I am responsible after the new business areas for cardiology monitoring.

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Sonochemically fabricated micro-electrode array based biosensors

A novel and patented procedure for the sonochemical fabrication of a new class of microelectrode array based sensor, with electrode element population densities of up to $2 \times 10^5 \text{ cm}^{-2}$, will be described. This approach involves firstly polymerising insulating ultra-thin-polymer films at conductive electrode surfaces. The electrode is then exposed to ultra-sound to cause ablation of the insulating thin polymer film. Sonochemical ablation of this film causes exposure of discrete areas of the underlying conductor. Each of these areas act as individual microelectrodes and collectively as a micro-electrode array. Microelectrode templates of this type may be used to form biosensors by firstly polymerising conductive polymers and co-entrapping enzymes or antibodies within the polymer. Model amperometric enzymatic biosensors for glucose and ethanol will be firstly described within this talk. AC impedance based antibody / antigen affinity based sensors will also be described for monitoring clinical markers such as Prostrate Specific Antigen (PSA), Neuron Specific Enolase (NSE), S-100 β and Myelin basic protein.

Speaker Profile - Séamus P. J. Higson, Institute of Bioscience and Technology, Cranfield University

Professor Higson's career to date has spanned academic departments of Chemistry, Medicine and Materials Science and this is reflected in the research interests in the group he now heads. Séamus Higson also serves within an advisory and / or consultative capacity for a number of public bodies and also acts as Technical Director for Microarray Ltd - a company formed upon science and patents originating from his laboratory. Microarray both manufactures micro-electrode arrays for use within chemical and biosensors and well as licensing patented technology to third party manufacturers.

Current research interests are primarily focussed towards practical implementation of electro-analytical science and analytical biochemistry for biomedical, environmental and industrial process control applications.

A recurrent theme in much of Professor Higson's research includes the design and fabrication of micro-electrode arrays for applications ranging from DNA analysis, (including genomic and proteomic applications), through to enzymatic biosensors and electro-analytical chemical sensors. Fabrication routes involve, for example, electrochemical polymerisation techniques, screen printing technology and sonochemical ablation approaches. Professor Higson is also author of a major text '*Analytical Chemistry*' published by the Oxford University press: (ISBN: 0 19-8502893).

Standardisation of protein diagnostics measurements

Many diagnostic devices now under development rely on fluorescence measurement for their output (for example, protein microarrays). However, there are a number of standardisation issues relating to these techniques. These include the requirement for suitable fluorescence standards, and suitable protein standards for testing and validating the devices. I will review progress in these areas at NPL and elsewhere. Potentially single molecule fluorescence measurement may offer opportunities for underpinning low-level fluorescence measurement and also offer new routes to measuring small quantities of biomolecules, and I will present current work to this end.

Speaker Profile - Dr. Alex Knight, Senior Research Scientist, National Physical Laboratory

Alex is a Senior Research Scientist in the Biotechnology group at the National Physical Laboratory. He is involved in activities in a number of areas, including fluorescence standards; spectroscopic techniques for biopharmaceutical quality control and single molecule fluorescence. He is a biochemist by training with 8 years' postdoctoral experience in single molecule biophysics. Much of his previous work has focussed on biological motor proteins and the cytoskeleton, where the single molecule techniques are particularly useful.

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Dual Polarisation Interferometry: Molecular measurement in the life sciences

Farfield Sensors have developed Dual Polarisation Interferometry (DPI) over the past decade to measure molecular changes. The company has not just developed new technology but is pioneering new measurement capabilities in the life sciences. This talk will highlight some of the technical and cultural challenges which have been met. These range from verification of the measurements generated by the technology to the use of calibration standards to their application in real measurement situations faced in the life science sector.

Speaker Profile – Dr. Marcus Swann, Farfield Sensors Ltd

Marcus is the Applications Manager for Farfield and has worked to develop applications of its proprietary technology since its concept in 1999. He has 50 publications (both academic papers and patents) to his name. Marcus is originally from a physical chemistry background gaining a PhD (Bristol) in the development of the electrochemical quartz crystal microbalance. He has since worked with a wide range of surface analytical methods and with technologies commonly used in biosensors. Much of this work has focused on simultaneously combining measurement techniques to gain greater insight into surface processes, of which Farfield's DPI is a natural extension.

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Modelling and calibrating an optical biosensor

Through the DTI's Joint Industrial Project (JIP) initiative, NPL has been working in collaboration with Farfield Instruments to bring optical and microwave modelling techniques to the Dual Polarisation Interferometry technology and investigate if primary refractive index standards can be used as a calibration route. The talk will provide an overview of how the JIP scheme operates including a review of the technical achievements made.

Terahertz biosensors

The field of Terahertz has only recently started to be explored, with applications in the medical and pharmaceutical fields exhibiting great promise and creating a lot of interest. This talk will provide a summary of what Terahertz radiation is, how it can interact with material and what potential

opportunities exist in the bio-sensing field. In particular label-free sensing, nanometer spatial resolution measurements and Theranostics opportunities will be discussed.

Speaker Profile - Dr Richard A Dudley, Senior Scientist with the Ultrafast Team at NPL.

Richard graduated with BSc & PhD in Applied Physics from Essex University in 1996 focusing on opto-electronic semiconductor devices. Joined NPL in 1997 and led development in the fields of high-speed opto-electronics, MMIC on-wafer devices, free-space electric field measurement, refractive index and Internet Calibrations. Currently, Terahertz techniques applied to the security and NDT markets are his primary focus.

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MultiSense dry enzyme system for simultaneous multiple parameter measurements

MultiSense® is a system for Point of Care use measuring multiple analytes from a single finger-stick sample of blood. It is a complete system comprising a disposable sensor strip and a hand held meter. Each sensor has an array of up to 6 microwells, the electrochemical measurement uses microband electrodes with an amperometric technique. Each well contains a cascade of enzymes to provide the specificity in the measurement, the first application of MultiSense® is for cardiac risk where a full lipid profile is obtained: Total Cholesterol, Triglycerides, HDL Cholesterol are measured and a LDL Cholesterol value is calculated by the meter. The chemistry in each microwell runs to completion, ie all the analyte in the microwell is metabolised by the enzyme system. This results in a quantitative accumulation of a reduced electron mediator which is measured as a pseudo steady state current within 2 seconds of applying a potential to the electrode. Typical current measurements are in the range of hundreds of nanoamps. Typically all the analyses are complete in under 2 minutes from application of the blood sample, c 30µl of whole blood. Unlike glucose sensors, where the measurement of blood lipids requires the separation of the plasma blood fraction from the blood cells, no separation or processing steps are required prior to application of the drop of blood to the sensor.

Speaker Profile - Dr John Broughall, Research Director, Oxford Biosensors Ltd

Trained as a microbiologist, after working in the NHS Pathology service spent my career within the IVD industry, primarily working on methods for rapid analysis. For the past 3 years has been in charge of Research at Oxford Biosensors, commercialising technology originally developed at the University of Oxford. This has involved interfacing Electrochemistry, Microfluidics and Biochemistry to produce a complete analytical system.

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Modification of Nanopore proteins for single molecule detection

Oxford NanoLabs is developing a Nanopore biochip and reader for a range of target analytes providing a discreet hand held 'point of care' device that delivers rapid results in the field. The product envisaged is a disposable chip that can be inserted into a hand held reader and used to test a patients biological sample for immediate feedback to the health care professional or end user for tailored treatment. This new type of sensor system is able to detect at the single molecule level through modification in ion channel Nanopore proteins. The development of the Nanopore Biochip will be described. The presentation will also describe the methodologies for modifying protein pores for detection of analytes ranging from small charged molecules through to proteins, antibodies and DNA detection.

Speaker Profile - Gordon Sanghera, CEO Oxford NanoLabs Ltd

Dr Sanghera has more than 15 years experience in the design, development and global launch of novel point of care biosensor devices. His extensive experience in medical device companies ranges from privately owned start-ups through to global pharmaceutical companies. Having held

both UK and US Director level positions within Abbott Laboratories, including Research Director, Manufacturing Process Development Director and most recently Worldwide Marketing Director, he has launched several generations of blood glucose biosensor systems for the consumer and medical markets, leading projects from inception through to full product launch, and has developed and validated market production processes to meet with the regulatory requirements for USA and Europe.

Materials selection for cell based sensors

Cell based sensors offer an animal 'free' route to testing or detecting potentially toxic materials especially if they can be formed into blocks of tissue that mimic that found in vivo. The success of this technology relies on providing a suitable housing in which to contain the cells and provide effective solute transport within the matrix. This requires an understanding of the structure of the housing and how it performs in terms of its permeability and mechanical resilience. This requirement is paralleled in tissue engineering where the emphasis is placed on nurturing an ever-increasing population of cells rather than using them as probes.

Speaker Profile - Paul Tomlins, National Physical Laboratory

Paul originally joined NPL's Photonics group to study non-linear effects in optical fibre and components. This work has led to current research in the use of light for optical bio-sensing applications. Areas of current interest are surface plasmon resonance (SPR) and optical coherence tomography. Paul is working closely with the biomaterials group at NPL to develop these optical methods for applications in this area.

Reliable interfacing of biosensors

Regardless of the sophistication of transducer technologies for sensing/biosensing and rapid advances in biomolecule integration, we are still left with systems that have to have direct interactive contact with the target biological sample. Accordingly, either the very strong and interfacial response of tissue and blood has to be controlled through sample bulk modification, or exposure time needs to be so restrictive that loss of surface integrity ceases to be important. The ideal approach, for devices that need to operate for any length of time within biological systems, is for the inclusion of surface modifiers which ameliorate bioreactivity to the device. This presentation will give some background of surface biocompatibility manipulation as attempted in conventional biomaterials, and then switch to illustrating polymeric film and membrane use in biosensors in order to at least partly overcome sensor response degradation, surface attachment of cells and colloids will be discussed at the chief agents responsible for this. A description will be provided of possible microfluidic strategies for interfacing sensors and for creating self-renewing barrier layers.

Speaker Profile – Professor Pankaj Vadgama, Director of the IRC in Biomedical Materials, Queen Mary, University of London

Present Appointment: Director of the IRC in Biomedical Materials, Queen Mary, University of London. Professor of Clinical Biochemistry, Barts and the London School of Medicine & Dentistry. Head of Service in the Department of Clinical Biochemistry, Barts and the Royal London NHS Trust. Prior to this he was Professor of Clinical Biochemistry, University of Manchester and Honorary Consultant Chemical Pathologist and Head of the NHS Department of Clinical Biochemistry, Hope Hospital (1988–2000), Professor of Medical Biomaterials, Manchester Materials Science Centre (1999-2000). Professor Vadgama qualified in Medicine at the University of Newcastle upon Tyne in 1971, and in Chemistry in 1976.

His particular strategy on biosensors has been to develop permselective, biocompatible and biomimetic polymeric membranes capable of stable transduction in whole blood and tissue. Both *in vivo* and *in vitro* work has been undertaken, including the use of miniaturised devices for glucose and lactate monitoring, immunosensing and interrogation of tissue-material interactions.

Current research work includes interfacial problems relating to sensor/biomaterial contact with the biomatrix, and the generalisable insights that may emerge from this. Relevant projects include: Spider silk for tissue engineering, materials for implantable electronic devices, microfluidic based separation, cell-surface interactions, biomaterial degradation dynamics, conducting polymers as biomaterials, tissue bioreactor design, cochlear implant electrodes, sensors for food microbial contamination.

Resonant acoustic profiling

Acoustic sensors that exploit resonating quartz crystals to directly detect the binding of an analyte to a receptor are finding increasing utility in the quantification of clinically-relevant analytes. We review the growth in different application areas including bacterial, viral, and oligonucleotide detection, with a focus on piezoelectric immunoassays developed by academic and commercial groups. Example data will be presented for detection of myoglobin, interleukin 1 beta (IL-1b) and enzyme co-factors. The specificity and affinity of antibody-antigen and enzyme-cofactor interactions can be rapidly and accurately determined without the need for concomitant labeling of the receptor or the analyte. The Akubio systems enable determination of protein concentration (recombinant human IL-1 b and recombinant human myoglobin) and quantification of co-factor binding (NADP⁺ and NAD⁺) to the enzyme glucose dehydrogenase. Resonant Acoustic Profiling™ is able to detect different classes of analytes in a relatively simple receptor-binding assay in less than 10 minutes. The technology shows promise for application to real-time immunoassays, biomarker detection and analyte quantification in general. The combination of this powerful technology platform with existing analyte concentration and presentation technologies should lead to simple, label-free, high sensitivity methodologies that can be used in multiple application areas of clinical chemistry and diagnostics.

Speaker Profile - Matthew Cooper PhD, MRSC, MIEEE, MAAS

Founder and Chief Scientist of Akubio Ltd. 1993 Australian Mind of the Year Finalist, 1995 George Murray Scholar at Cambridge University, 1999 Centennial Howard Florey Fellow, 2005 UK Entrepreneur of the Year Finalist. Consulted widely for biosensor, biotechnology and pharmaceutical companies in the UK, Europe and the US. Review panel member for BBSRC, EPSRC, Australian Research Council and Science Foundation Ireland. Several patents issued or pending with 50 papers in journals including Nature Cell Biology, Nature Biotechnology and Nature Drug Discovery.

Magnetic biosensors

An immuno-sensor using magnetic detection technology is described. In this device two antibodies bind to a large antigen to form a sandwich. One antibody is immobilised to the sensor surface and the other is coated on the surface of a paramagnetic particle. The antigen acts as a biological bridge cross-linking the paramagnetic particle to the sensor surface. By applying an external magnetic field the paramagnetic particles can be pulled on to the sensor surface and greatly increase the speed of reaction.

A resonant sensing coil is printed using a screen printing process on to the underside of a sensor chip, directly below antibody spotted on the reactive surface. The bound particles immobilised on

the reactive surface cause a change in the inductance of the sensing coil. This inductance change is measured using a magnetometer that contains a phase locked loop based circuit which produces a stable frequency output that is proportional to the number of immobilised particles on the reactive surface.

This presentation will examine how a number of factors determined the sensitivity of an immunoassay for CKMB, a cardiac marker. These factors include the resonant frequency of the sensor coil, the strength of the external magnetic field; the nature of the paramagnetic material in the particle and the characteristics of the antibodies used.

Speaker Profile - Dr Richard Luxton, Deputy Research Director, Faculty of the Applied Sciences, University of West England

Richard trained as a clinical biochemist for thirteen years in the Bristol Royal Infirmary before moving to the Institute of Neurology in London to study for a PhD in neuro-immunology, studying antibody affinity in the CSF of patients with multiple sclerosis compared with patients with encephalitis. At the University of the West of England he has focused on research in the area of developing new rapid diagnostic technologies for point of care diagnostics and environmental analysis. One area of research, funded by Randox Laboratories, that has been particularly successful is the development of magnetic detection technology for use in immunoassays. Using this technology a magnetic-biosensor has been developed that will perform an assay in three minutes or less. Work is also ongoing in investigating the use of this technology in DNA detection and lateral flow techniques. Other technologies that are being developed for rapid diagnostics include surface plasmon resonance techniques (using the SPREETA chip) and impedance spectroscopy in conjunction with the company Kiaku. Richard is also a University Business Fellow and a project coordinator for defra projects developing electrochemical biosensors for applications in food safety.

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Poster Presentations

Towards biosensing with DNA-functionalised single walled carbon nanotubes

Single walled carbon nanotubes (SWNT) have the potential to make ultra-sensitive biosensors due to their excellent electrical properties and nano-scale size. However, SWNTs are difficult to functionalise and manipulate. Here we present how DNA can be used to purify the SWNTs from amorphous carbon associated with the production process, and also how DNA can be used to functionalise the SWNTs with gold nanoparticles. Moreover, we show that by using dielectrophoresis, functionalised SWNTs can be positioned and trapped between micro-electrodes. Such trapped SWNTs are the basis of a generic biosensor, where biomolecules could be detected by changes in resistance as they bind to the gold-functionalised SWNTs.

J.Charles.G Jeynes^{†‡}, Ernest Mendoza[†], Johnjoe McFadden[‡], S.Ravi.P Silva[†]

Nano-Electronics Centre, Advanced Technology Institute[†] and School of Biomedical and Molecular Sciences[‡], University of Surrey

QCM characterisation of hydrogel-based molecularly imprinted polymers

We are developing protein recognition polymers (HydroMIPs) using molecular imprinting technology to produce synthetic antibody systems for protein recognition. These HydroMIPs are being tested for biosensing of proteins (using the quartz crystal microbalance) and for sample clean-up. The technology has major implications in novel diagnostics for medicine, food and the environment.

Subrayal M. Reddy, Iain Buchanan

School of Biomedical and Molecular Sciences, University of Surrey

Dielectrophoretic DEP-Wells: electrostatic characterisation of cells

Dielectrophoresis (DEP) is a phenomenon of induced movement in suspended microparticles such as cells, bacteria and viruses. Using computer models, it is possible to use the induced force spectrum to determine the electrical properties of the particles. Although the phenomenon was discovered over 50 years ago, widespread use of the technique has been inhibited by the lack of simple measurements and useful electrode structures. We have developed the DEP-Well system to overcome this; by constructing 3D electrode-bearing "wells", we have a system that allows simple, rapid measurement of populations of cells simultaneously, whilst developing a mathematical analysis system that allows the extraction of multiple population data from a single sample.

Mike Hughes, Centre for Biomedical Engineering, University of Surrey

Non-invasive glucose and lactate monitoring through reverse iontophoresis.

Iontophoresis is a non-invasive process which relies on both electro-migration and electro-osmosis to deliver drugs across the skin. More recently, studies have been undertaken to examine the use of reverse iontophoresis as a method of metabolite extraction, in particular glucose and lactate. Monitoring of these metabolites is considered crucial to the wellbeing of diabetic patients and potential patients suffering from myocardial ischemia. An *in vitro* model designed to mimic skin and internal fluid was studied to investigate the potential of using reverse iontophoresis as the basis for such a monitoring device. The work presented here includes characterisation of the skin-contacting hydrogel, and glucose extraction from the model system.

M. Farmahan, P. Connolly, G. Eccleston

Medical Devices Doctoral Training Centre, Bioengineering Unit, University of Strathclyde

Point-of-use potentiometric immunosensor for industrial and medical diagnostics

The Sortec multi-sensor approach to assay development and transfer is applicable to many market sectors: food and drink, agricultural, environmental and pharmaceutical; but particularly medical and veterinary diagnostics. The biosensors are extremely sensitive to small changes in pH, ionic strength and redox potentials close to the transducer surface; which means that many assay formats can be used: immunoassays, nucleic acid based assays, enzymatic assays, redox reactions, cell based systems and ion selective membranes. The biosensor itself is unaffected by colour, turbidity or particulates therefore the sample requires little or no preparation i.e. the sensors can be exposed directly to complex fluids including blood, serum or urine. Typically incubation times of 5-10 minutes allows detection of most small analytes (<500 daltons) and larger proteins in the ppt or fM range and lower.

***D. Purvis, O. Leonardova, N. Blair, M. Dunn, K. Wright, S. Bridgeman.
Universal Sensors Ltd, Cambridge***

Nanoparticles functionalised by the “Hand-in-Glove” method for *in-vitro* diagnostics

The convergence of nanotechnology and biology is expected to produce major advances in diagnostics, therapeutics and materials science, but until recently the range of molecules that could be attached to nanoparticles (NPs) was still very limited. At Liverpool we have developed a method that allows virtually any type of capture molecule to be attached to a NP. It also allows the number of molecules attached to each particle to be controlled. This is important because the number of capture molecules per particle helps to determine the sensitivity of diagnostic tests. Our method has become known as the ‘Hand-in-Glove’ method because it depends on a linear relationship between high molecular weight polymers (the glove) and nanometer-sized particles (the hand). It is much simpler than rival methods - just add the polymer to the particles and they are ready for use in diagnostic tests. No purification is required. And once the amount of Hand-in-Glove polymer required to coat the particles is known, production-line volumes can be prepared in seconds.

***Jenny Aveyard, Maryam Mehrabi and Robert Wilson
Centre for Bioarray Innovation, Liverpool University, Chemistry Department***

A microfluidic diagnostic chip integrating DNA extraction, amplification & detection

Most diagnostic tests require sending to analytical laboratories, which have high running costs as methods are both time consuming and are yet to be completely automated. The aim of our chip is to make genetic testing quicker, cheap, portable and available at point of care. The microfluidic chip will couple three steps; DNA extraction from biological sample, polymerase chain reaction (PCR) followed by electrochemical detection of the target.

***H. Ayers, M. Azimi, M. Zolgharni, L. Milash, W Balachandran, P Slijpcevic
Brunel University***

Electrochemically switchable DNA biosensors

A research programme at the University of Edinburgh is developing a novel concept in switchable biosensors. These are based on nano-scale biomolecules which can switch between two molecular conformations depending on a controllable ion flux. This conformation change can be enabled or disabled by the detection of target biomolecules and may be detected using optical or other techniques.

The characteristics of DNA Holliday Junctions (HJ) as ion-controlled nanoscale switches have been explored. Ion-induced switching in solution has been detected optically using fluorescence

resonance energy transfer (FRET) and ions of varying size and valence quantitatively assessed according to threshold concentration, sharpness of the switching transition, and fluorescence quenching. The principle of electrochemical control of DNA HJ switching has also been demonstrated using electrochemically-generated Zn^{2+} .

These nanoscale switchable biosensors bring specific advantages. The switching action provides a means of enhancing signal:noise ratio of signals, and they can be used to optically detect biomolecules without sample labelling. If mounted on an electronic biochip they can be used to form addressable microarrays, using locationally-specific ion flux to control array spots. This technology has potential applications in developing instrument-based devices for research and drug development, and for improved clinical diagnosis and treatment.

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(2) School of Chemistry, The University of Edinburgh

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(5) IBM T.J. Watson Research Center, Yorktown Heights, New York, 10598, USA

Optical Coherence Tomography.

Optical Coherence Tomography (OCT) is a non-invasive imaging technique that can be used to generate highly detailed three-dimensional images. The ability of OCT to detect the structural geometry of a system on a cellular level has direct applications to Biosensors. OCT can be used in-vivo to characterise the structural composition of a biosensor, being able to image individual cells, their spatial organisation and structure. At NPL, new techniques are being developed to couple OCT's spatial and geometric precision with flow rate acquisition. This new technique, known as Doppler-OCT (D-OCT), allows complex flow measurements to be taken from within a sample. This new technique will allow for the adhesion of cells to a surface to be monitored, the flow over the entire surface to be measured, and changes in either structural composition or flow to be detected, and as all data can be taken simultaneously, the interaction between all these elements can be investigated in detail.

**Paul Tomlins, Matthew Tedaldi, Bio-Photonics, National Physical Laboratory
Marco Bonesi, R.K. Wang, Cranfield University**

Impedimetric microfluidic Point-of-Care sensors with RF data transmission.

Point-of-Care cardiac marker systems are under development, incorporating the novel elements of RF technology, and impedimetric immunoassay sensing. RF data transmission from a sensor to a PDA has been demonstrated. Impedimetric sensing offers label-free easy-to-perform assays for cardiac markers, with the further advantage of rapid read-out.

Our recent work has demonstrated the capability to fabricate nanoscale interdigitated electrode-type patterns in gold, using a focussed ion beam. We have also shown the capability to nanotemplate gold, which will allow direct protein attachment.

The impedance changes due to capture of an antigen simulant by primary antibodies immobilised to a self-assembled monolayer on gold, have been measured, in a 3 electrode electrochemical cell, and reach an end-point level after about 15-20 minutes. Similar tests are proceeding for myoglobin capture and sensing on planar gold electrode sensors.

Two electrode planar sensors have been fabricated in gold/titanium on PI, with further optimisation ongoing. Microfluidic delivery of blood/serum sample to the sensor is under further investigation, with test structures being fabricated in glass, and bonding of lids by various methods being optimised. Sensor microfluidic structures will be replicated in PMMA. This work is intended to lead to commercially-realizable point-of-care cardiac marker monitoring systems.

***M. Tweedie, R. Subramanian, I. Craig, E. T. McAdams, J. A. D. McLaughlin
Nanotechnology Research Institute and the Northern Ireland BioEngineering Centre, at the
University of Ulster at Jordanstown, Belfast.***

Biosensor research at the London Centre for Nanotechnology (LCN)

Developing innovative biosensors relies on a range of skills at the frontier of physical and life sciences. The interdisciplinary structure of the LCN provides an ideal environment to develop various biosensor platform technologies, from integrated micro-fluidics silicon optical benches to biomechanical label-free assays. We present a selection of biosensor related research carried out at the LCN, including label free biomedical assays based on micro electromechanical systems (MEMS) and lymph node biopsy location based on magnetic nanoparticles. Finally, a modern approach to the sense of smell is illustrating the strong in-house theoretical activities.

R. McKendry, Q. Pankhurst, C. Renner, M. Stoneham.

***London Centre for Nanotechnology
University College London and Imperial College***