

From menace to marvel: Nitric oxide in health and disease

The BioPark Hertfordshire, Welwyn Garden City, AL7 3AX: 20th March 2009

Nitric oxide (NO), generated by the enzyme nitric oxide synthase (NOS) plays a key role in a diverse range of physiological and pathophysiological conditions within the cardiovascular, immune, reproductive and nervous systems. This meeting will examine the role of NO in neurodegenerative diseases such as Parkinson's and Alzheimer's diseases, endothelial function/dysfunction, cancer, diabetes and infection. Specific attention will be given to the methodology used by investigators to examine the roles of NO in health and disease.

- 9:00 – 9:45 **Registration**
- 9:45 – 10:00 **Introduction by the Chair:** *Dr Alan Wallace*, AstraZeneca, UK
- 10:00 – 10:30 **The nitrate-nitrite-NO pathway in health and disease**
Dr Jon Lundberg, Karolinska, Sweden
Nitric oxide (NO) is generally viewed as an autocrine or paracrine messenger, mostly regulating local intracellular processes or acting on cells in its near vicinity. The major metabolic pathway for NO involves its rapid oxidation into the higher nitrogen oxides nitrite and nitrate. Until recently, these inorganic anions have been generally considered to be inert endproducts of NO metabolism. Although, it is true that the bioactivity of NO is acutely terminated following oxidation to nitrate and nitrite, recent lines of research now suggest that a reverse pathway exists in which nitrate and nitrite are reduced stepwise into bioactive NO again. The nitrate-nitrite-NO pathway is emerging as a likely regulator of physiological functions in the gastrointestinal tract and in the cardiovascular system. In particular, it might serve as a backup system to ensure NO like bioactivity also in situations when the endogenous L-arginine/NO synthase pathway is dysfunctional. In addition, this alternative pathway can be harnessed therapeutically in prevention and treatment of disease. Finally, there is an intriguing nutritional aspect to this, since the major supply of nitrate and nitrite in our bodies comes from our everyday diet.
- 10:30 – 11:00 **Targeting eNOS coupling in human atherosclerosis**
Dr. Charalambos Antoniades, University of Athens and University of Oxford-UK
Endothelial nitric oxide synthase (eNOS), maintains its enzymatic coupling in the presence of its co-factor tetrahydrobiopterin (BH4). Under conditions of increased oxidative stress, BH4 may be oxidized leading to eNOS uncoupling. The uncoupled form of eNOS then becomes a source of superoxide radicals instead of NO. Strategies targeting eNOS coupling are usually focused on increasing BH4 bioavailability, either by increasing its synthesis or by preventing its oxidation. These strategies include antioxidants and especially the administration of 5-methyl-tetrahydrofolate, as well as direct exogenous BH4 administration. The impact of routinely used therapeutic strategies in human atherosclerosis such as statins, needs further investigation
- 11:00- 11:30 **Nitrite: Can we teach an old dog new tricks?**
Dr Alexandra Milsom, Barts & The London Medical School, UK.
Nitrite has a historical role in areas as diverse as medicine, explosives and food preservation. In medicine recent evidence has shown that the reduction of nitrite via chemical acidification and a nitrite-reductase activity of various endogenous proteins may increase nitric oxide (NO) bioavailability through the nitrate-nitrite-NO pathway. Nitrite reduction to NO is enhanced during hypoxia, a condition under which constitutive NO production in the vasculature may be reduced. Nitrite has been used successfully to protect against ischaemia-induced tissue damage in a number of ischaemia/reperfusion models. This presentation will discuss the history of nitrite, from an ancient cure for evil to a toxicological bad guy, and its future potential in the treatment of vascular disease and ischaemia/reperfusion injury.
- 11:30 – 11:40 **Speakers Photo**
- 11:40 – 11:50 **Introduction to the Biopark**
- 11:50 – 13:00 **Lunch**

- 13:00 – 13:30 **Nitric oxide in the life and death of neuronal cells**
Dr Guy Brown, Department of Biochemistry, University of Cambridge, UK
High levels of NO can be produced by inducible NO synthase expressed in glial cells in inflammatory conditions. But in addition to iNOS expression, NO-induced neuronal death normally requires either (i) hypoxia to enable NO to inhibit mitochondrial respiration, or (ii) superoxide production from microglial NADPH oxidase, reacting with NO to give neurotoxic peroxynitrite.
- 13:30 – 14:00 **The where, why and how of targeted nitric oxide delivery**
Professor Ian Megson, UHi Millenium Institute, Inverness
Since its discovery in the 1980s, nitric oxide (NO) has been heralded as a major breakthrough in disease prevention and management. However, the promise and expectations have not yet been delivered with respect to new NO donor drugs on the market and we are still heavily reliant on drugs that have been in use for decades. One possible reason for the lack of development in this area is the lack of specificity of many NO donor drugs and their global impact, as opposed to local activity. This presentation will explore some possible solutions to tackle targeted NO delivery for use in a number of cardiovascular conditions and will highlight the enormity of the task ahead to fully exploit the remarkable potential of NO in health and disease.
- 14:00 – 14:30 **S-Nitrosoglutathione Metabolism**
Dr Lea-Ann Dailey, King's College, London
- 14:30 – 15:00 **Afternoon Tea/Coffee**
- 15:00 – 15:30 **Nitric Oxide management strategies in pathogenic enterobacteria**
Dr Gary Rowley, University of East Anglia, Norwich
Nitric oxide (NO) is a highly reactive gas encountered by microbes in many environments. Some bacteria can synthesise NO themselves during cellular respiration in anaerobic conditions. Others meet it within animal tissues where macrophage cells use NO as a toxic molecule to repel the invaders. For every natural toxin, some bacteria have evolved methods to counter the effects. NO is no exception. Salmonella are able to survive and even grow and divide within macrophages. They have at least three enzymes that can metabolise NO. The advantage to the bacterium of multiple methods to counteract one toxic molecule is to give flexibility in the many environments in which the bacteria may find themselves exposed to this gas - in soil, the gastrointestinal tract, the bloodstream or the macrophage, both with and without oxygen
- 15:30 - 16:00 **Nitric oxide in intestinal barrier breakdown leading to colitis**
Professor William Roediger, University of Adelaide, Australia
Excess nitric oxide from anaerobic bacterial denitrification, together with sulphide from anaerobic fermentation of protein, is capable of inactivating free CoA in colonocytes by S-nitrosylation, producing nitrosothiols. Depletion of free CoA diminishes lipid synthesis, detoxification and beta oxidation in colonocytes. In conjunction with impaired protein synthesis by NO, cellular membranes and tight junctions of the colonic epithelial lining cells are impaired which leads to epithelial barrier breakdown. Nitric oxide production in the rectal lumen of ulcerative colitis patients is significantly greater than that observed in healthy patients. Areas of prolonged gut mucosal contact with luminal NO, i.e. distal colon and appendix, are most prone to develop colitis. Free CoA depletion can be established in experimental animals producing a colitis identical to human UC. Therapies to alter bacterial nitrate respiration and NO production in the colon of UC need to be developed in the future
- 16:00 – 16:30 **Hydrogen sulfide and the nitric oxide cross-talk: A promising future for two old gas bags?**
Dr Matt Whitemann, Institute of Biomedical and Clinical Science, Peninsula Medical School, Exeter, England
Hydrogen sulfide (H₂S) is a well known and pungent toxic gas that has recently been shown to be synthesised in man. Analogous with nitric oxide, H₂S is synthesised from amino acids (cysteine/homocysteine) by distinct enzymes (cystathionine-g-lyase and cystathionine-b-synthase) which show a marked degree of tissue specificity. In the past few years, H₂S has emerged as a novel and increasingly important mediator in the cardiovascular system and as an inflammatory mediator. Recently we showed chemical, biochemical and physiological interactions of H₂S with NO (or species derived from these gases under physiological conditions) in vitro and in vivo. The physiological and pathophysiological effects of H₂S, novel H₂S donors and the interaction of H₂S with NO in vivo will be discussed.
- 16:30 – 17:00 **Chairman's summing up**

About the Chair

Dr Alan Wallace studied Medical Biochemistry at Birmingham before moving to UMIST where his PhD concentrated on cAMP phosphodiesterases. After a postdoctoral fellowship back in Birmingham he moved into the pharmaceutical industry. Here he has worked on a wide range of projects, including a multinational search for novel NO synthase inhibitors, targeting several major diseases and involving research sites in the UK, USA, Sweden and Canada - and a collaboration with the structural biology group of John Tainer and Libby Getzoff at the Scripps Clinic in San Diego. He has presented aspects of this work at several international meetings, and his latest paper, describing how NO synthase inhibitors can achieve selectivity between isoforms, has just been published in *Nature Chemical Biology*.

About the Speakers

Dr Guy Brown, received his Ph.D. in 1986 from the Biochemistry Department at Cambridge, then had a College Research Fellowship from St Catharine's College. He then moved to the Department of Physiology, University College London, with a Wellcome Trust Post-doctoral Fellowship, and after 18 months transferred to a Royal Society Research Fellowship at the Department of Biochemistry at UCL. In 1994 he returned to Cambridge with a Royal Society Research Fellowship, and has been a Senior Lecturer since 2002. He has published about 150 scientific papers, and several books, including most recently: *The Living End: The future of death, aging and immortality*. His research has been funded by: the Medical Research Council, the Biotechnology & Biological Sciences Research Council, the European Union, the Wellcome Trust, the British Heart Foundation, and Alzheimer's Research Trust

Professor William Roediger is currently Associate Professor of Surgery; having researched the cause of IBD for 30+ years

Dr. Whiteman obtained his PhD in 1998 from King's College, London. After completing an Arthritis Research Campaign funded post-doc he moved to Singapore in 2000 to take up a position as Assistant Professor in the Faculty of Medicine, National University of Singapore (NUS) and in 2003 he joined the National Graduate School Faculty. In 2005 he was promoted to Associate Professor and then to Assistant Dean of Research in the Faculty of Medicine, NUS. For family reasons he moved to the UK in 2007 where he is a principal investigator at the Institute of Biomedical and Clinical Science, Peninsula Medical School in Exeter, Devon. Dr. Whiteman has published over 120 peer reviewed articles and several book chapters on redox signalling and nitric oxide. His more recent work has focussed on the emerging physiological and pharmacological importance of hydrogen sulfide and its interaction with nitric oxide and other gaseous mediators.

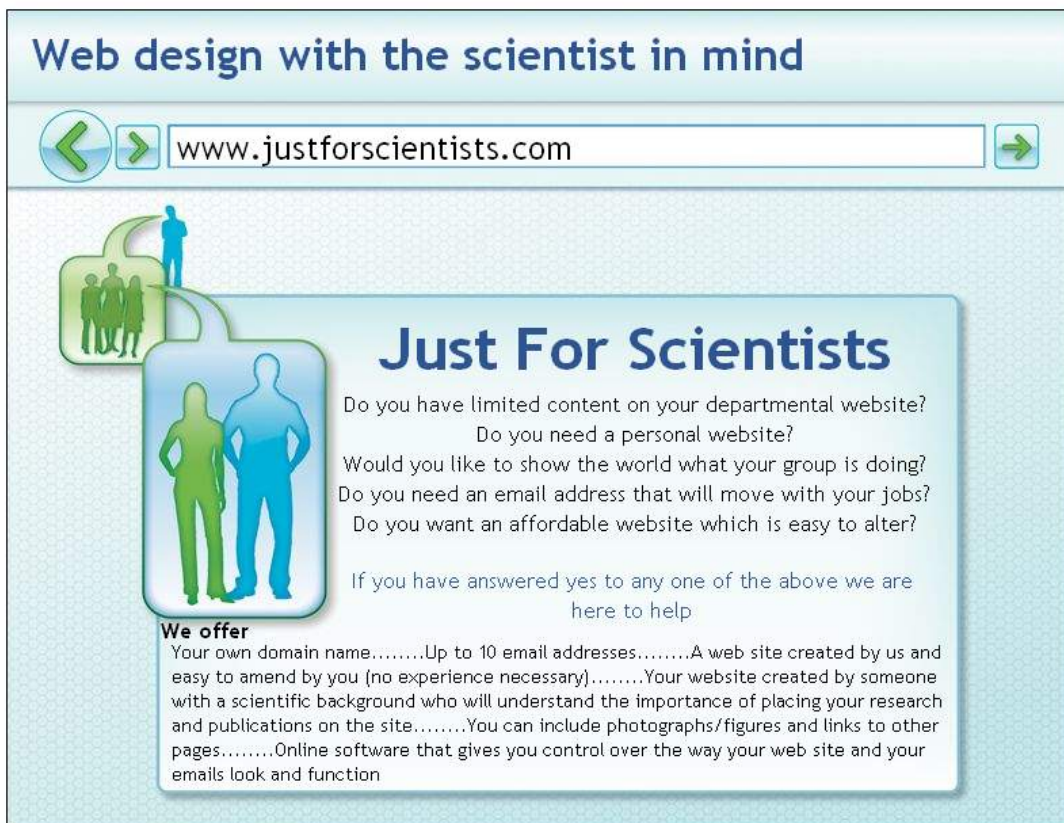
Dr. Alexandra Millsom received her PhD in 2003 in nitric oxide metabolism from the University of Wales, College of Medicine. This work received the Nitric Oxide Society Young Investigator Award. She remained in the nitric oxide field and moved to Boston, USA for a postdoctoral position at the Whitaker Cardiovascular Institute before returning to the UK to Cardiff University to continue nitric oxide-related research. She is currently a British Heart Foundation-funded post-doctoral researcher at Barts and The London, investigating the role of nitrite and nitrate in vascular function.

Dr Charalambos Antoniades has qualified Athens University Medical School in 2000, with honors. He was a Research Fellow in the 1st Department of Cardiology during the years 2000-2004 and he received his PhD degree in Cardiology by the same University in 2005. He has been a post-doctoral Research Fellow in the Department of Cardiovascular Medicine in the University of Oxford-UK since 2004. He received many Young Investigators Awards including those from the American College of Cardiology 2004, World Congress of Cardiology 2006, European Society of Cardiology (in 2006, 2007 and 2008), International Society of Heart Research 2008, and Hellenic Society of Cardiology. Now he is an independent researcher in Athens University Medical School. His research is focused on the regulation of intracellular inflammatory pathways and endothelial nitric oxide synthase in atherogenesis, and he has more than 100 full length papers in international peer-review journals.

Professor Ian Megson received his PhD at the University of St Andrews in 1993, then went on to a British Heart Foundation funded post-doctoral position and then moved to Edinburgh to take up a BHF Junior Fellowship. In 1998, he took up a lectureship at Edinburgh and progressed to Senior Lecturer in 2003. In July 2006, he accepted the LifeScan Chair of Diabetes at UHI Millennium Institute, Inverness, with the remit of starting a new research department in the area of Diabetes and Cardiovascular Science. Throughout his research career, nitric oxide has dominated: in particular, he has worked closely with chemists to develop new means of delivering nitric oxide in the right amounts and the correct place to optimize its unique beneficial qualities.

Dr Gary Rowley is a Molecular Microbiologist and my primary research interests relate to the environmental regulation of the intracellular pathogen *Salmonella Typhimurium*, including how various stresses and stimuli are integrated to achieve the desired transcriptional response. In particular I am interested in environmental conditions which induce the envelope stress response pathways and those that induce changes in virulence gene expression.

This meeting was **organised by Euroscicon** (www.euroscicon.com), a team of dedicated professionals working for the continuous improvement of technical knowledge transfer to all scientists. Euroscicon believe that they can make a positive difference to the quality of science by providing cutting edge information on new technological advancements to the scientific community. This is provided via our exceptional services to individual scientists, research institutions and industry. The event was hosted by **BioPark** (www.biopark.co.uk), a research and development centre in Welwyn Garden City providing specialist facilities and support for bioscience and health technology businesses to grow, and to develop new products and technologies



The image shows a screenshot of a web browser window. The title bar reads "Web design with the scientist in mind". The address bar contains "www.justforscientists.com". The main content area features a large graphic on the left with silhouettes of people and a speech bubble. To the right, the heading "Just For Scientists" is followed by a list of questions and a list of services offered.

Web design with the scientist in mind

www.justforscientists.com

Just For Scientists

Do you have limited content on your departmental website?
Do you need a personal website?
Would you like to show the world what your group is doing?
Do you need an email address that will move with your jobs?
Do you want an affordable website which is easy to alter?

If you have answered yes to any one of the above we are here to help

We offer
Your own domain name.....Up to 10 email addresses.....A web site created by us and easy to amend by you (no experience necessary).....Your website created by someone with a scientific background who will understand the importance of placing your research and publications on the site.....You can include photographs/figures and links to other pages.....Online software that gives you control over the way your web site and your emails look and function