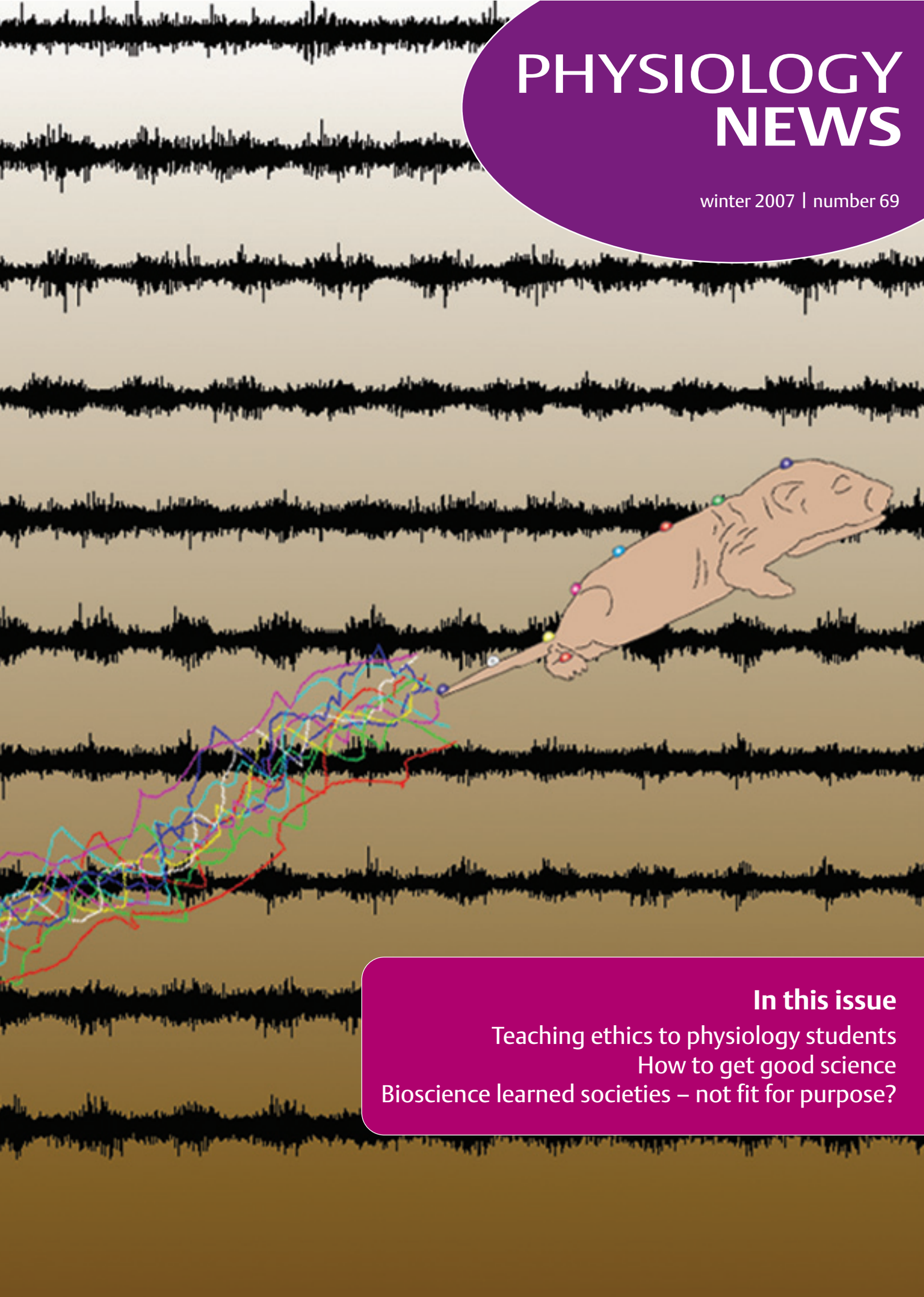
PHYSIOLOGY NEWS

winter 2007 | number 69



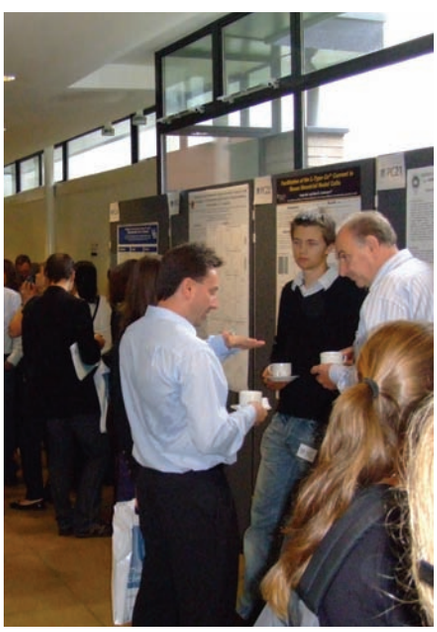
In this issue

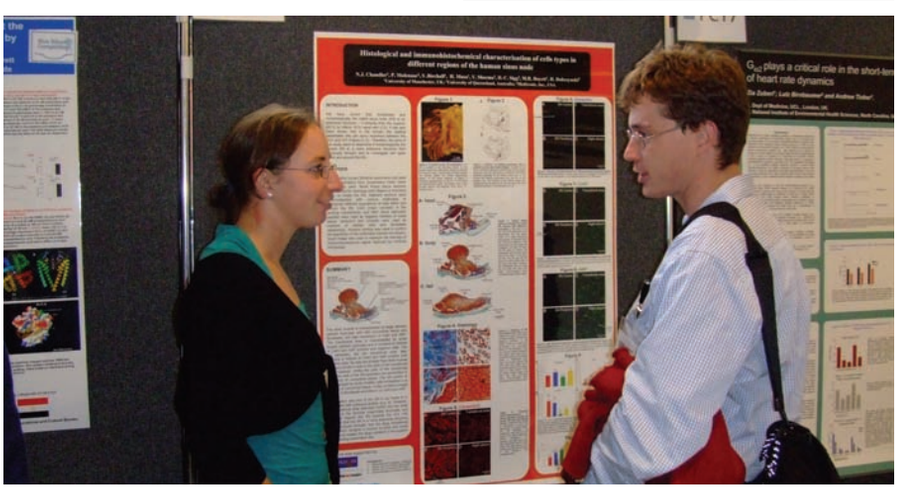
Images of Manchester

Manchester Focused Meeting

Cardiac electrophysiology: with a special celebration of the centenary of the discovery of the sinoatrial and atrioventricular nodes

5–6 September 2007





PHYSIOLOGY NEWS

The Society's dog. 'Rudolf Magnus gave me to Charles Sherrington, who gave me to Henry Dale, who gave me to The Physiological Society in October 1942'

Published quarterly by The Physiological Society

Contributions and queries
Senior Publications Executive
Linda Rimmer
The Physiological Society Publications Office
P O Box 502, Cambridge CB1 0AL, UK

Tel: +44 (0)1223 400180
Fax: +44 (0)1223 246858
Email: lrimmer@physoc.org
Web site: http://www.physoc.org

ISSN 1476-7996

The Physiological Society is registered in England as a company limited by guarantee: No 323575.
Registered office: PO Box 11319, London WC1X 8WQ
Registered Charity: No 211585.

Printed by The Lavenham Press Ltd

The Physiological Society

Advancing the science of life

It's good to talk (carefully)

The *Physiology News* editorial slot intermittently encourages scientists to get involved in public discussions about science, whether local or national.

However, there are downsides to this. One is the dilemma when you get asked to comment on something you don't know much about.

The safe option on this is, first, to admit you don't know much about it, and then second, fall back on generalities and consensus, or on things that couldn't possibly be objected to but bear a re-statement. In other words, stick to what you can say, give useful information, and avoid ranting.

The danger comes if you engage the mouth before the brain, speak off the cuff and give unvarnished personal opinions without making clear they are strictly opinions.

The more famous and eminent the scientist, the greater the danger. For the run-of-the-mill scientist speaking in a local forum, schools workshop, town meeting, Café Scientifique, or whatever, you are unlikely to come too much of a cropper if you do say something daft. You will usually be able to correct and clarify what you said, or even take back things you did not mean.

At the other end of the scale, if you are a world-famous scientist and Nobel Laureate facing a room full of journalists, and touch unhappily on a subject as explosive as race and intelligence – and note that there are multiple definitions of either word, both popular and scientific – then you can provoke a global storm of denunciation, as James Watson did recently.

It is not my purpose here to debate what Watson said, or is said to have said. Much has been written by people far better placed than I to comment (see e.g. [1]). What we can say is that if, as seems likely, we have seen a great scientist's exit from the public arena, it is a sad way for him to bow out. Most of us, I am sure, hope the fall-out will not overshadow Watson's lifetime of distinguished achievements in science.

What I do want to do here is comment on the limits of scientific authority, and where this interfaces with the continued need for scientists to speak up in public.

As a thoroughly ordinary scientific practitioner I am in the fortunate position that I will never be asked by a journalist with a tape-recorder running for my thoughts on global warming, the nature of consciousness, or when an embryo becomes a human being. But some scientists do get asked these questions, and it is then that they have to choose their words carefully. Sadly, it is common in places promoting bad science to find quotations from famous scientists speaking about things that they should have been wise enough to keep quiet about. The statements made by Kary Mullis, of PCR fame, about HIV and AIDS in his autobiography (2) are one well-known example, but there are others.

'Stick to what you know, and if you can't, stick close to what is known' – might be a good rule of thumb.

Physiology is perhaps fortunate not to touch often on areas as directly contentious as race, genetics and IQ. But it still offers opportunities for foot-in-mouth stuff. Physiologists in the public eye typically do stick close to their own ground: they can frequently be found commenting on issues around animal experimentation, and sometimes about brain scanning, or stem cells, or IVF, or even the scientific basis of drug policy, while they are less likely to be found offering their views on whether science disproves the existence of the soul.

I hope Watson's fate will not scare eminent scientists off engaging in public debates. More than ever, we need them out there explaining, and clarifying, and – perhaps most important of all – correcting common and even dangerous misconceptions. Just the weekend I wrote this, Mike Rennie was chiding *The Guardian* for multiple inaccuracies in their coverage of 'synthetic biology' (3). Long may people like him continue to speak out. But equally, long may they take care what they say.

One of the more measured online commentaries on Watson's downfall on the *Wired Science* blog (4) quoted the American author Marilynne Robinson: 'There is nothing harder than knowing what it is you don't know.' As scientists, we forget this at our peril.

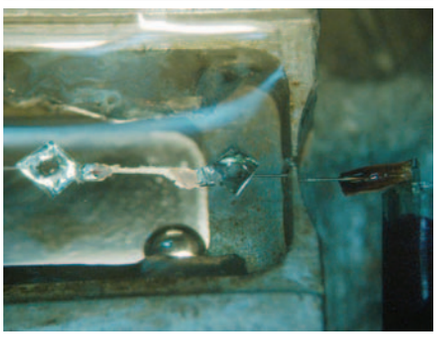
Austin Elliott

1 http://www.timesonline.co.uk/tol/comment/columnists/guest_contributors/article2702762.ece
2 Mullis, Kary. 1998. *Dancing Naked in the Mind Field.* New York: Vintage Books.
3 http://www.guardian.co.uk/comment/story/0,,2199474,00.html
4 http://blog.wired.com/wiredscience/james_watson/index.html

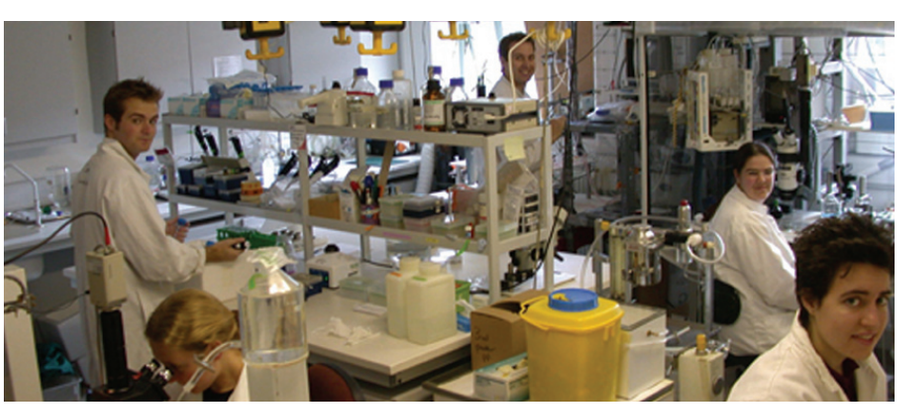
Renal cortex: physiological basis of glomerular and tubular diseases

The Department of Physiology and Pharmacology at the University of Bristol is delighted to host its first Physiological Society Focused Meeting on 17 and 18 December. The Department was formed on 1 August by the merger of the existing Departments of Physiology and Pharmacology, which had complementary research interests and expertise, focused on cardiovascular research, cell biology and neuroscience, each pursued in a collaborative, inter-disciplinary research environment. The merger has generated critical mass in each of these research areas, and increased the expertise and facilities available. The Department now comprises 35 academic staff and six independent research fellows, as well as excellent support staff; it retains Bristol's traditional strengths in *in vivo* and systems physiology, but genetic, molecular and cellular approaches are also widely used in work within the Department, which spans from the gene to the whole animal and translation to the clinic.

This Physiological Society Focused Meeting on renal cortical physiology in health and disease is organised jointly by the Renal Physiology and Epithelial & Membrane Transport Special Interest Groups and generously supported by Kidney Research UK and the Renal Association. The focus of the meeting is the regulation of renal function by epithelial and associated cells of the renal cortex, in particular the role of the glomerular and tubular epithelium and endothelium in regulating salt and water balance in health and disease. An uninterrupted layer of epithelial cells line the nephron, from podocytes in glomeruli to intercalated and principal cells in the collecting duct. Ultrastructural and molecular differences between these cell types reflect the vast differences they make to the conversion of ultrafiltrate to urine. Recent advances in animal models of human disease that inform normal function, cell-specific inducible transgenic models that highlight the role of individual molecules in normal adult physiology, cell biology techniques that provide conditionally immortalised cell types of precise origin and molecular characterisation of ion channels and transporters that underpin normal physiology and pathophysiology have provided a wealth of new models of renal cell and nephron behaviour. Investigation of individual specialised epithelial cell types in the nephron with these tools has generated an explosion in our understanding of renal epithelial cell function. This meeting will highlight this wealth of new knowledge, focusing on molecular aspects of ion and solute transport by cell types throughout the nephron, revealing how techniques used to advance our understanding of the behaviour of one nephron cell type are applicable to neighbouring cell types. The function of epithelial cell types throughout the nephron, and additional cell types such as mesangial and glomerular endothelial cells, will be discussed. We expect the meeting to provide a valuable forum for the exchange of information between physiologists investigating renal function and clinicians treating renal dysfunction.



Bristol skyline looking towards the Will Memorial Building and science buildings (*Zhe Xu*, top); muscle physiology in action (*Hamish Roots*, above); the physiology laboratory (*Dave Bates*, below left).



Bristol has strong interests in renal research, especially glomerular microvascular and renal epithelial physiology. This work is conducted within the Department of Physiology and Pharmacology, the Academic Renal Unit at Southmead Hospital in the Faculty of Medicine and Dentistry at the University and the Department of Biochemistry.

We look forward to welcoming you to the meeting, with 10 invited speakers from Europe and North America, 16 oral communications selected from submitted abstracts and an extensive poster session. The Society dinner will be held in the first class dining salon aboard the ss Great Britain, Isambard Kingdom Brunel's masterpiece of engineering refurbished in its original style from the mid 19th century. We promise you excellent science, rigorous discussion and and entertaining time.

Dave Bates & David Sheppard
Department of Physiology,
University of Bristol, UK

The Young Physiologists Bursary Scheme (YPBS) aims to encourage young physiologists to attend their first scientific meeting of The Society without the need to present an abstract. Twenty two awards were made to support young physiologists' attendance at LifeSciences2007 in Glasgow in July and here's what some of them had to say about it ...

When I heard about LifeSciences2007 it seemed like the perfect way to get an insight into many of my research fields all at once – a bit like Christmas for scientists! As I was fairly new in my lab, I was not immediately invited to the conference, but fortunately my supervisor told me of the Young Physiologists Bursary Scheme from The Physiological Society for students attending their first conference and not presenting. I applied immediately and was granted the full bursary of £250 to cover conference registration fees, travel, up to two nights accommodation and, if budgeted, conference events. My lab subsidised my accommodation and travel and, being a bit of a socialite, I was delighted to use the remaining bursary on the conference gala dinner and welcome drinks!

My lab group and I arrived in Glasgow on Sunday 8 July at the glass-fronted, towering Crown Plaza Hotel, which was conveniently adjoined to the SECC where the conference took place. The next morning we registered for the conference and were handed badges, pens, notebooks and a fully abstracted guide to the lectures. We got to work planning where we needed to be that day and each set off to our lecture of interest. This was no easy decision, however, as much time was spent deliberating which to choose when two beneficial lectures were scheduled at the same time, or one after another at opposite ends of the building! I must have looked rather amusing running between lecture theatres with my bag on my back, and notebook and pen at the ready! But it was worth it; the subjects covered were varied, the number of speakers numerous and lectures were well prepared, well timed and thoroughly interesting. I even ventured to lectures outside my area of work because of their quirky and curiosity-provoking titles and abstracts. It was great to put faces to names, hear others' findings on subjects you are researching, chat over coffee about exciting new information you had just discovered and to feel part of such a large 'team' of people with the same interests and enthusiasm.

In between lectures and lunch, opportunities arose to peruse the high quality posters submitted by students and to absorb yet more knowledge. The trade exhibition was useful too, with many scientific companies available to give advice and product information (not forgetting freebies) to anyone who strolled around the stands.

The fun didn't stop there ... the social events should not be forgotten! Welcome drinks at the Kelvingrove Art Gallery and Museum was a great way to get people talking, whether it was about science, the Scottish piper playing the bagpipes as guests mingled, or the curious taxidermy display of

'amazing creatures' on the first floor! It was a great opportunity for me to waffle on to people about my research and see them wide-eyed and intrigued rather than looking at their shoe laces or yawning!

The gala dinner on Wednesday was a more formal, extended version of the welcome reception, and after a tantalising three course dinner and short, humorous, yet rallying speeches from members of each of the organising groups of the conference, the party began! The fantastic live band started to play, and the wine provided with the meal was evidently required as a little 'Dutch courage' to enable people to let their hair down and start flinging arms and legs around and swinging partners out of sequence to what should have been an organised Scottish Ceilidh! Much fun was had by all, whether you were the offending in question, or the wise who sat giggling at the mess of ungainly, gyrating bodies on the dance floor.

All in all the conference was extremely worthwhile and I look forward to the next one. Much knowledge was acquired and shared by such an encouraging number of members from various science communities, giving myself, and undoubtedly others, the passion and persistence to continue investigating this fascinating subject we call science.

Nicola Trim

PhD student, Laboratory of Molecular Signalling, Babraham Institute/University of Cambridge, UK

A YPBS gave me the chance to attend both LifeSciences2007 and to take part in the young physiologists' symposium held prior to it. This was a small meeting focusing on advances in signalling. It enabled young scientists like me to share ideas and learn about exciting research developments in the field. This introductory meeting was on a smaller scale and facilitated interactions amongst the younger generation of scientists. I am highly appreciative of being awarded the bursary to enable me to attend both these events. I certainly made the most of what the programme had to offer; it was a very busy 4 days! I would definitely recommend other young scientists to take advantage of the bursary scheme to attend future events.

Clare Gladding

Final year PhD student, Department of Anatomy, University of Bristol, UK

I found that I had ample time to wander through and talk at the poster sessions. The availability of the abstracts at the venue ensured navigating through the posters was an easy undertaking. The talks aimed at junior scientists were an excellent addition to the conference. As someone who has not published yet, the talk on writing and publishing papers was very helpful and informative. It would be fantastic to see more of these kinds of tutorials in the future.

Ciara Doran

First year PhD student, Department of Physiology, University College Cork, Ireland

Notwithstanding the diverse symposia presented by field leaders and the special plenary lectures, LifeSciences2007 also displayed a huge collection of posters. By alternating short talks given by postdocs with longer communications given by leaders In the corresponding field, it was easy to switch from one session to another and a great opportunity to learn about other people's research. Displaying the same posters over 2 days allowed time to see and focus on each one. The different workshops and trade stands emphasized the dynamic of this meeting. Apart from the excellent science, the social events offered a more informal way to meet and discuss.

Caroline Cros

PhD student, University of Manchester, UK

The Physiological Society Plenary Lecturers at LifeSciences2007

Tom Bolton (top) gave the Annual Review Prize Lecture *Smooth muscle excitation*; Michael Joyner (centre) gave the Michael de Burgh Daly Lecture *A sympathetic view of the sympathetic nervous system and human blood pressure regulation*; and Denis Noble(above) gave the Paton Lecture *Claude Bernard, the first systems biologist, and the future of physiology.*

The main things I came away with were the need to immerse myself in science more than I currently have been to understand my areas of interest, and the complexity of signalling that has evolved, and its importance in human biology.

Michael Morgan
BSc student in Medical Genetics, University of Huddersfield, UK

It was my first time at an international scientific meeting. As a postgraduate student from Hong Kong, this was a tremendous personal achievement and the meeting was a great opportunity for me to broaden my horizons and enrich my knowledge, and so help me with the challenges in my career. For me, the most impressive event on Career Day was the Speed data session, to which The Physiological Society invited academics, clinical research officers, science teachers, and even editors of peer-reviewed journals. There was a friendly and relaxed environment for people to share past working experiences, so that participants learnt more about the possible career paths.

Lai-Ming Yung
PhD Student, Department of Physiology, Chinese University of Hong Kong, China

Young Life Scientists' Symposium at LifeSciences2007

The Young Life Scientists' Symposium (YLS2007) took place on 8 July at the University of Strathclyde in Glasgow and provided a venue for young scientists to present and interact with other researchers at the same stage in their scientific careers. YLS2007 was a satellite to the main Life Sciences2007 meeting. It brought together young scientists from the Biochemical Society, Physiological Society and British Pharmacological Society to present, discuss and network in a setting meant to encourage the participation of MSc and PhD students and post-doctoral researchers. YLS2007 was an overwhelming success! In attendance were over 160 registered delegates from all over the world including delegates from Japan, Australia, Canada, USA, Slovakia and China. More locally there were delegates present from all regions of the UK, including those from London, Oxford, Belfast, Plymouth, Leicester, Bristol, Glasgow, Dundee and Aberdeen. The theme of the symposium was Advances in signalling and there were two poster sessions showcasing almost 100 abstracts and 12 oral communications highlighting selected research in the areas of neuronal, calcium, cardiovascular and cellular signalling. There was also a full social programme allowing delegates to register and mingle at an evening reception prior to the symposium and a post symposium dinner at Modern India restaurant (generously sponsored by the Universities of Strathclyde and Glasgow respectively).

Young physiologists enjoy the welcome drinks reception at the Kelvingrove Art Gallery and Museum – a great way to get people talking.

Phil Hawkins (Babraham Institute) gave an insightful keynote lecture. His talk covered not only the scientific research of membrane lipids and associated kinases but also discussed his road to scientific research. Our other keynote speaker, Nina Balthasar (RCUK Fellow, University of Bristol), gave an equally inspiring talk relating her transition from a young scientist to independent researcher to the way in which she has built on her research programme and understanding of the field of neuronal pathways in metabolic balance.

As is the case in most scientific research these days, the posters presented at YLS2007 were not so much divided into areas based on societal membership but more due to an overall research theme. The high quality of both the oral and poster presentations made it difficult to choose the awardees for the prizes. Each society presented an award for both the best oral and poster presentation. The Biochemical Society awards were presented to Kathleen O'Hagan (University College Dublin) for oral presentation and Sanam Mustafa (University of Glasgow) for poster presentation; The Physiological Society awards were presented to Patrick Howorth (University of Bristol) for oral presentation and Lydia Jimenez-Diaz (University College London) for poster presentation; the British Pharmacological Society awards were presented to Grant Budas (Stanford University) for oral presentation and Alexandra Zahradnikova (Slovak Academy of Sciences) for poster presentation. An overall YLS2007 prize was awarded by Pfizer to Andrew McBride (University of Dundee) for his poster entitled The glycogen binding domain of the â–subunit of AMPK functions as a sensor of glycogen quality.

Finally, we would like to thank our sponsors, Institute of Pharmacy and Biomedical Sciences (SIPBS) University of Strathclyde, Institute of Biomedical and Life Sciences (IBLS) University of Glasgow, Pfizer, Stratech, AstraZeneca, Sigma-Aldrich and St. Jude Children's Research Hospital for generous funding of YLS2007. Most importantly we would like to thank the Biochemical Society, Physiological Society and British Pharmacological Society for making YLS2007 such a success. Thanks!

The YLS2007 Organising Committee
Susan Chalmers and **Marnie Olson** (University of Strathclyde), **Patrick Howorth** (University of Bristol), **Ahmed Khweir** (University of Glasgow)

Setting the pace

Otto Hutter, pictured right with Hilary Brown, gave the introductory talk at the recent Manchester meeting

Let me start by presenting my credentials as a cardiac physiologist. Early in 1946 at University College London, Sir Charles Lovatt Evans and his technician Charlie Evans set up a heart lung preparation, just so as to get things going again after the war. I was then the first, and for a while the only, post-war physiology student, and with 3 years of war-time employment at the Wellcome Physiological Research Laboratories behind me, I was recruited as a third pair of hands. My task was to defibrinate blood collected from a donor dog by whisking it with a bundle of stiff wires.

Later I sat in at Lovatt Evans' lectures. He told us how Stannius (1852) tied ligatures between the chambers of a frog heart and so identified the sinus venosus as the pacemaker, and how MacWilliam (1885) divided the heart of an eel and observed:

'The first separated portion of cardiac tissue to exhibit a rhythm is the ostial part of the sinus venosus. Next the auricular basal wall shows a rhythmic action of a slower rate. And lastly, after a long standstill the ventricle may come to exhibit a very slow rhythm.'

MacWilliam understood that in the intact heart the sinus venosus keeps the auricular rhythm in abeyance, and he interpreted the potential rhythmicity of the auricle in metabolic terms, as follows:

'The phase of exhaustion after a beat is gradually recovered from; the amount of energy for another discharge accumulates; if no stimulus arrives from the sinus venosus when the auricular tissue has recovered sufficiently, it is readily conceivable that a continued rise in excitability may result in a welling over in the form of a spontaneous discharge of energy'. At a time when electrophysiology was still in its infancy, this was quite an insightful interpretation.

Lovatt Evans told us that, by the time Stannius had focused on the sinus venosus as the seat of cardiac rhythmicity, histologists had already shown that it contains many ganglion cells and nerve fibres. Here is how one great Victorian interpreted that fact:

'We know, from evidence presented by other stripped muscle and nerve, that the contraction of the former is the result of the excitement of the latter. In like manner we know that the ganglia are centres whence that excitement originates. We are therefore justified, analogically, in seeking for the sources of contraction of cardiac muscle in the cardiac ganglia; and the experiments which have been detailed – by showing that the rhythmical contractions continue in any part of the heart which remains connected with these ganglia, while it ceases in any part cut off from them – prove that they really are the seats of the regulative power'. These were the words of the great Thomas Huxley (1854), and with such powerful advocacy it took nearly half a century for the neurogenic doctrine to be overthrown.

An early sceptic of the neurogenic theory was Michael Foster (1859; 1878). He observed how cilia beat rhythmically, and he therefore held that rhythmicity is not confined to neurones. Moreover, he found that small, ganglion-free pieces of snail's heart, and the similarly ganglion-free apex of a frog ventricle, can be made to beat spontaneously. Later Gaskell (1883, 1900) extended that observation to the apex of the tortoise heart, which is also devoid of nerve cells. And in an oft-quoted sentence of Talmudic complexity Gaskell argued:

'The automatic rhythm of the ventricle is of the same kind as that of the auricle and therefore as that of the sinus, so if the latter is due to the presence of motor ganglia so is the former; on the other hand, if the former be due to some inherent rhythmical property of the ventricular muscle then the latter is due to a somewhat better development of the same kind of property in the muscular tissue of the sinus.'

By itself that argument did not at once win the day. But once it was followed by Ransom's (1885) findings, on the octopus heart, which is also composed of striated muscle, only there the ganglia are situated outwith the heart so that they can be extirpated, and that without abolishing autorhythmicity; when there followed the finding that in the chick embryo the primodial heart beats before nerve cells reach it; and finally when the situation in the mammalian heart was resolved without recourse to a nervous connection between its chambers, then at last did the myogenic theory gain universal acceptance.

Why did the myogenic theory have such an uphill struggle? I think it was partly because it was so much against the spirit of the age. During the second half of the 19th century, Social Darwinists, like Herbert Spencer, were fond of drawing parallels between the organization of

the body biologic and that of the body politic.

For instance, the muscular tissues of the body were likened to the manual labouring class of society; the secretory glands to manufacturers, the white blood corpuscles to policemen, and so on. Finally, a nervous system dominating overall, was likened to the commanding and administrative class, which on this view, justifiably crowned a hierarchical society. So the proposition that a muscular labouring organ like the heart might have a will of its own, was an idea with revolutionary implications that was instinctively resisted.

By contrast, we know now that everywhere generation of spontaneous activity depends on what ion channels are operative in a cell; that an inward current activated by hyperpolarisation contributes to impulse formation both in cardiac pacemaker cells and in nerve cells, for instance the thalamo-cortical neurones; moreover, the systems controlling that current are similar in the sinus node and in neurones (Pape,1996). And just to round the circle, it has now been shown that the same funny current also orchestrates autorhythmicity in truly neurogenic hearts like those of the medicinal leech (Angstadt & Calabrese,1989).

So today we can see that the old distinction between subservient, labouring cardiac muscle cells and self-motivating, governing nerve cells was an illusion. Biology can no longer be traduced to defend class distinction.

But there remain some lessons for the body politic that can still be drawn validly; for instance, from the organisation of the sinus node, to which Mark Boyett and his colleagues have contributed so much.

The problem is how to protect the leading pacemaker cells, the precious centre of originality, from being weighed down by too great a load. In the heart that load is the electrotonic coupling to non-pacemaker cells: in academe, you may like to think, it is the bureaucratic load.

The solution seems to be for the leading pacemakers to keep themselves somewhat apart, and to be sheltered by other potential pacemaker cells in the atrial border zone, cells that are content most of the time just to buffer the load represented by the auricular myocardium.

Of course, there are situations where the imposition of a sizeable load is beneficial. Take the case of that potential up-start, the A-V node. There the load represented by the bundle of His keeps potential pace-maker fibres – potential troublemakers when they are not required – more subdued than would otherwise be.

Finally, the organisation of the pacemaker also teaches how vital functions are best safeguarded by multiple back-up provisions – whether in the body or in society. In the slow-mo mutation in the zebrafish, for instance, which lacks the funny pacemaker current, a slower but still life sustaining heart rate is maintained by the interplay of other current systems, like the L-type Ca current and the slow K current (Baker *et al*. 1997). One may wish that the recently failed tap water supply to Gloucester and Tewksbury had been similarly safeguarded!

But enough of such diversions! So I'll finish with a plea. When you come to lecture on cardiac rhythmicity to young students, don't start by showing them pacemaker cell action potentials with auricular and ventricular action potentials for comparison. These mean a lot to us, but to students they are just traces to be memorized. Instead, please start by presenting them with the simple classical experiments, experiments done with a piece of thread, a pair of scissors and perhaps with a thermode to produce localised changes in temperature. Such experiments they will comprehend joyfully, they will arouse their interest, fire their enthusiasm for experimental physiology, so you will then be able to build on this, even to the point of telling your student about that funny current which has become so important, about the new drugs based on it, about novel bioengineered pacemakers, and about all the other wonderful advances it was our pleasure to learn about at this symposium.

Otto F Hutter
Institute of Biomedical and Life Sciences, University of Glasgow, UK

This article is based on the introductory talk given at The Physiological Society Focused Meeting on cardiac electrophysiology in Manchester from 5 to 6 September.

References

Angstadt JD & Calabrese JD (1989). A hyperpolarization-activated inward current in heart interneurons of the medicinal leech. *J Neurosci* **9**, 2846–2847.

Baker K, Warren KS,Yellen G & Fishman MC (1997). Defective 'pacemaker' current (Ih) in a zebrafish mutant with a slow heart rate. *Proc Natl Acad Sci USA* **94**, 4554–4559.

Foster M (1859). On the beat of the snail's heart. *Rep Br Assoc Adv Sci* **29**, 160.

Foster M (1878). *A textbook of physiology*. Macmillan & Co, London.

Gaskell WH (1883) On the innervation of the heart, with special reference to the heart of the tortoise. *J Physiol* **4**, 43–127.

Gaskell WH (1900). The contraction of cardiac muscle. In *Textbook of physiology*, ed. Schaefer EA. Young J Pentland, Edinburgh.

Huxley TH (1854). On the present state of knowledge as to the structure and function of nerve. *Proc Roy Inst* **2**, 432–437.

MacWilliam JA (1885).On the structure and rhythm of the heart of fishes, with special reference to the heart of the eel. *J Physiol* **6**, 246–286.

Pape HC (1996). Queer current and pacemaker: the hyperpolarization-activated cation current in neurones. *Ann Rev Physiol* **58** 299–327.

Ransom WB (1885). On the cardiac rhythm in invertrebrata. *J Physiol* **5**, 261–372.

Stannius HF (1852). Zwei Reihen physiologischer Versuche : 1 Versuche am Froschherzen. *Arch f Anat u Wiss Med* **19**, 85–100.

What can clinical medicine give back to physiology?

John Dickinson continues his personal account with a look at an incredibly well-known phenomenon whose pathophysiology remains a 'most intriguing mystery'

All observations have shown that the common faint begins with sudden peripheral vasodilatation, usually accompanied by bradycardia. The head must be at a higher level than the heart. The faint reaction cannot be triggered in anyone lying down flat. Progressive loss of blood in a supine subject leads to a progressive fall in blood pressure and eventually to a progressive loss of consciousness. Nothing happens suddenly. But typical acute fainting can easily be brought about in normal people tilted head up by many manoeuvres such as blood loss, blood pooling with leg cuffs, hypotensive drugs, lower body negative pressure and emotional trauma. It can be aggravated by heat, fever, coughing, micturition or hypoxaemia.

The cardiovascular events are summarised in Fig.1. During the period up to the faint itself (A to B) blood pressure is maintained by progressively increasing sympathetically-induced peripheral vasoconstriction and tachycardia. But within a few seconds blood pressure falls precipitously (C). Everything reverses. Sympathetic vasoconstrictor tone decreases and there is also active neurogenic vasodilation as the faint begins. When sympathetic nervous vasoconstrictor activity has been recorded by microelectrodes impaled in an autonomic nerve trunk, the faint is seen to begin by the abrupt cessation of sympathetic vasoconstrictor nerve activity. The heart slows because of increased efferent vagal activity. Bradycardia can be prevented by blocking efferent vagal activity with atropine, but fainting still occurs because of widespread active vasodilation (Barcroft & Edholm, 1945). All changes quickly reverse if the subject lies flat, or is retransfused.

'What suddenly switches off this compensation (at the onset of a faint) to result in vasodilation and bradycardia remains one of the most intriguing mysteries in cardiovascular physiology' (Hainsworth, 1996a). One popular and current view of its aetiology – which appears in several textbooks – is that fainting is caused by 'increased activity of ... receptors located in the left ventricle ... excited by improper squeezing of the myocardium when the ventricles contract vigorously around an almost empty chamber' (Oberg & Thoren, 1972). Roger Hainsworth has criticised this explanation because 'few receptors have been shown to be excited in this way ... and people with transplanted, and therefore denervated ventricles, show similar responses to hypovolaemia' (Hainsworth, 1996b). This is where clinicians can throw light on human physiology. I can confirm an important observation made by the late Peter Sharpey-Schafer, Professor of Medicine at St Thomas' Hospital Medical School. He had noticed that congestive heart failure gives 'immunity from vaso-vagal syncope'. This crucial observation has never been refuted, though it has been largely forgotten. Several of my own patients with congestive heart failure have spent long periods propped up, or tilted head-up in bed, sometimes even sleeping in this position, without adverse effects or fainting. Sharpey also made interesting observations on blockages in the large veins in some of his patients: 'You can bleed a subject with a block in the superior vena cava in such a way that the pressure above [the block] remains exactly the same. The pressure below [the block] of course is falling, but there is a high venous pressure in the head and arm. The same thing can be done with an inferior vena

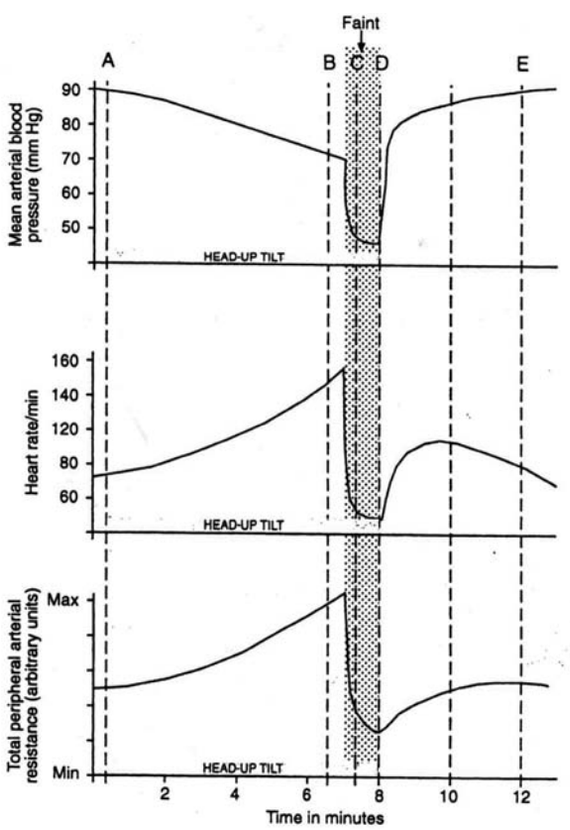


Figure 1. Graphic diagram of the sequence of events (time running from left to right) in the minutes before, during and after a faint, induced in a normal supine adult volunteer by head-up tilting in a warm room. The appropriate compensatory mechanisms were maintaining blood pressure and adequate brain blood supply until the blood pressure fell suddenly and the subject lost consciousness (see text).

caval block, maintaining a high pressure in the lower half of the body with a falling pressure in the heart and they [the patients] will faint. We suggest, therefore, that somewhere between the top of the superior vena cava and the bottom of the superior vena cava there is a reflex mechanism, if there is a reflex at all' (Sharpey-Schafer). These marvellous – but never definitively published – observations pin down a site either in the right atrium or in the large systemic veins close to the heart, where a normal or increased venous pressure prevents fainting. But what is the receptor? How does it work?

Sixty years ago David Whitteridge had set me the task of comparing cardiac fluctuations of the right atrial pressure of cats with simultaneously recorded patterns of afferent nerve discharge from low-pressure venous baroreceptors in the right atrium. I used anaesthetised artificially-ventilated open-chest cats in a steam chamber, recording afferent baroreceptor impulses in nerves coming from the right atrium to join the right vagus nerve (Fig. 2). The main burst of impulses began immediately after the P-wave of the ECG, at the time that atrial pressure was at its highest (Dickinson, 1950). I could identify the site of origin of the receptors by lightly touching the right atrial wall with a glass rod. This produced a shower of nerve impulses when the receptors at the site were those whose impulses I was recording. I could obtain an impressive torrent of impulses from many adjacent atrial receptor sites by gently pushing in the right atrial wall with my fingertip.

I remembered this experience when I began to think about the cause of fainting. By this time I was clinically experienced. I had read Sharpey's clinical observations and logical conclusions about fainting. It suddenly flashed across my mind that the mysterious faint reaction could be explained by the sudden collapse of unfilled low pressure veins and right atrium. This could signal false information to the brain – that the heart was over- rather than under-filled. Landgren had observed that carotid sinus baroreceptor discharge rate could actually increase when the artery collapsed. 'Increased impulse discharge is due to deformation of the arterial wall at low pressures' (Landgren, 1952). This phenomenon was later described as 'collapse-firing' of baroreceptors.

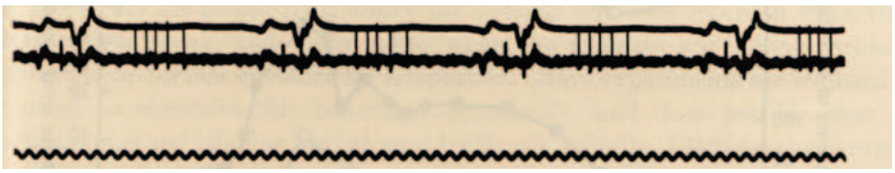


Figure 2. Two typical single fibre records from afferent nerves arising from the right atrium of a cat, and joining the right vagus nerve. Venous pressure was low in this case, and the atrial fibre discharges only two impulses between each P and QRS wave. The later burse of 5 impulses comes from an arterial baroreceptor (from Dickinson, 1950).

Mechanical activation of atrial receptors (or electrical stimulation of the cut central ends of the cardiac vagal filaments) produces reflex systemic vasodilatation and invariably slows the heart. Afferent vagal baroreceptor impulses terminate in the solitary tract nucleus in the medulla, at which site opiate receptors are present in high density. The opiate antagonist naloxone inhibits the early cardiovascular effects of blood loss in man. This region of the brain stem receives projections from the amygdala and hypothalamus and might be the common path for the initiation of emotional fainting.

Collapse-firing of low pressure baro(stretch)-receptors in the right atrium and great veins, possibly on the left side as well, plausibly and completely explains the sequence of events in the common faint. I published this hypothesis in 1993 (Dickinson). It has so far been neither criticised nor disproved. I hope that one day it will be tested by investigators who can watch human right atrial wall movements by ultrasound imaging as a faint begins. I have not yet located collaborators with equipment, facilities and volunteers – and with enough curiosity – to examine this fascinating problem. Meantime it remains an example of the way in which observations made by clinicians can suggest a solution to an interesting and well-studied physiological phenomenon.

This article and my previous one (*Physiology News* **66**, 10–12) have given examples of ways in which clinical medicine can contribute to physiology. My final piece, to be published in a future issue of *Physiology News*, will look at Hippocratic fingers. I have described many more medical mysteries from a physiological viewpoint in my recently published book (Dickinson, 2005).

John Dickinson
Wolfson Institute of Preventive Medicine, London, UK

References

Barcroft H & Edholm OG (1945). On the vasodilatation in human skeletal muscle during posthaemorrhagic fainting. *J Physiol* **104**, 161–175.

Dickinson CJ (1950). Afferent nerves from the heart region. *J Physiol* **111**, 399–407.

Dickinson CJ (1993). Fainting precipitated by collapse-firing of venous baroreceptors. *Lancet* **342**, 970–972.

Dickinson CJ (2005). *Medical mysteries*. Book Guild, Lewes.

Hainsworth R (1996a). Physiology and pathophysiology of syncope. In *Syncope in the older patient*, ed. Kenny RA, pp 15–32. Chapman & Hall, London.

Hainsworth R (1996b). The physiological approach to cardiovascular reflexes. *Clin Sci* **91**(Suppl), 43–49.

Landgren S (1952). On the excitation mechanism of the carotid baroreceptors. *Acta physiol scand* **26**, 1–34.

Oberg B & Thoren P (1972). Increased activity in left ventricular receptors during hemorrhage or occlusion of caval veins in the cat. A possible cause of the vasovagal reaction. *Acta physiol scand* **85**, 164–172.

Sharpey-Schafer EP (1952). General discussion. In *Visceral circulation* (CIBA Symposium), p 268. Churchill, London.

How to get good science

David Colquhoun considers how a university can achieve the best research and teaching, and the most efficient administration – the aim of every university vice-chancellor (president, rector or provost) in the country

Academics, like everyone else, are expected to do a good job. They are paid largely by taxpayers, and taxpayers have every right to demand value for their money. The problem is that it is very hard to measure the value of their output. Most of the ideas that have made life as comfortable as it is in the affluent West have their origins in science departments in universities, but it isn't possible to place a monetary value on, say, James Clerk Maxwell 's equations of electricity and magnetism, or on Bernard Katz's work on synaptic transmission. Still less is it possible to measure the contributions of A E Housman, Stanley Spencer or Augustus John (all UCL people, as it happens).

This paper describes one example of what happens when universities change from being run by academics to being run by managers. It describes an effect of corporatisation in the medical school of Imperial College London, but the same trends are visible in universities throughout the world. The documents on which it is based were sent to me after I'd written 'All of us who do research (rather than talk about it) know the disastrous effects that the Research Assessment Exercise (RAE) has had on research in the United Kingdom: short-termism, intellectual shallowness, guest authorships and even dishonesty' (Colquhoun, 2007).

The problem is not so much the RAE itself (the last one was done much better than the assessment described below), but rather it is the effect that the RAE has had on university managers, who try to shape the whole university in their misperception about its methods. It is another example of Goodhart's law. The problem arises when people with little understanding of scholarship, or of statistics, attempt to measure numerically things that cannot be so measured. That is a plague of our age (Colquhoun, 2006), but it is a process loved by politicians, 'human resources' people and university managers.

Imagine how you would feel if you were sent every year a spreadsheet that showed your publication score and financial viability, and showed these things for all your colleagues too. Well, you may say, there's nothing wrong with knowing how you are doing. But imagine too that your publication score is entirely automated, with no attempt to measure the quality of what you are doing. And imagine that if your grants don't cover your costs, you are in danger of being fired. And imagine that your meetings with senior colleagues consist of harassment about what journals you publish in, and how many grants you have, not a discussion of your scientific aims. Not so good, you may think. But this is exactly what has been happening at Imperial College Medical School.

Let's take a closer look at how academics are being assessed.

Imperial's 'publication score'

The publication score that appears alongside that of your colleagues is calculated thus.

Multiply the impact factor of the journal by the author position weight, and divide by the number of authors. The author position weight is 5 for the first and last author, 3 for the second author, 2 for the third author and 1 for any other position.

This index is clearly the invention of an uninformed incompetent. That is obvious for a start because it uses the impact factor. The impact factor is a (superfluous) way of comparing journals. It is the invention of Eugene Garfield, a man who has done enormous harm to true science. But even Garfield has said:

'In order to shortcut the work of looking up actual (real) citation counts for investigators the journal impact factor is used as a surrogate to estimate the count. I have always warned against this use.' (Garfield, 1998)

Garfield still hasn't understood though. As the examples below show, the citation rate is itself a very dubious measure of quality. Garfield quotes approvingly:

'Impact factor is not a perfect tool to measure the quality of articles, but there is nothing better, and it has the advantage of already being in existence and is therefore a good technique for scientific evaluation.' (Hoeffel, 1998)

And you can't get much dumber than that. It is a 'good technique' because it is already in existence? There is something better. Read the papers.

Try asking an impact factor enthusiast why it matters that the distribution of citation numbers for a given journal is highly skewed, and you will usually be met with a blank stare. One effect of the skew is that there is no detectable correlation between impact factor and citation rate (see, for example, Seglen, 1997; Colquhoun, 2003) . The easiest way to illustrate the numb-skulled nature of this assessment is with a few examples.

Publication score versus citation

Take a selection of 22 my own publications (the selection is arbitrary: it spans a range from 15 to 630 citations and omits some of the dross). Fig. 1*A* shows that the well-known lack of correlation between citations and impact factor is true for me too. Fig. 1*B* shows the same for the publication score.

The highest publication score (77.3) was for a two page perspective in *Science* , with a mere 41 citations (Sivilotti & Colquhoun, 1995). As perspectives go, it was fine. But it seems that this was 7.2 times more valuable than my best ever paper (on which I was recently asked to write a classical perspective) which has a publication score of only 10.7 (but 565 citations) (Colquhoun & Sakmann, 1985). My lowest publication score (in this selection) is 2.08. That is for a

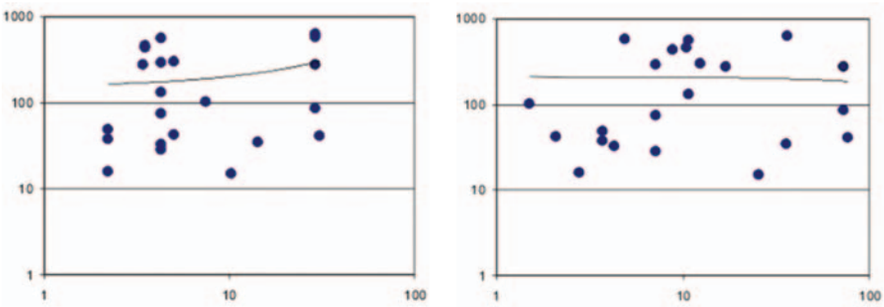


Figure 1. *A*, number of citations versus impact factor (log-log scales). *B*, number of citations versus 'publication score' (log-log scales).

Hawkes *et al*. (1992) , a mathematical paper which provides the method needed for maximum likelihood fitting of single channel recordings, without which most of my experimental work could not have been done; its mathematical difficulty may account for its modest number of citations (42) but its value for our work has been enormous after the maths was put into a computer program that can be used by the semi-numerate.

Citations versus value: a real life story

The dimwitted nature of the publication score, and also of using citation rates, can be illustrated in another way. Consider some of the background to a couple of examples; these are the real life facts that are ignored by bean counters.

Colquhoun & Sakmann (1981) got a score of 73.2 and 278 citations. It was a three page *Nature* letter, a first stab at interpretation of the fine structure of single channel openings. It wasn't bad, but since *Nature* papers are so short they mostly can't be thought of as real papers, and 4 years later we published the work properly in *The Journal of Physiology* (Colquhoun & Sakmann, 1985), the result of 6 years work (57 pages, 565 citations). For this Imperial would have awarded me a publication score of a mere 10.7.

Here is another interesting case. If we exclude chapters in *Single channel recording* (Neher & Sakmann, 1983; 1995) which apparently don't count, my most highly cited paper is Colquhoun, Neher, Reuter & Stevens (1981). This has 630 citations and a publication score of 36.6 for me, though only 14.6 for Harald Reuter. The reality behind this paper is as follows.

In the early days of gigohm seal Harald Reuter decided that he wanted to learn the method, and to achieve this he invited three of us who already had some experience of the method to spend part of the summer vacation in Bern. We had a wonderful summer there, and being somewhat overmanned it was not very stressful. It would, I think, be fair to say that all four of us did much the same amount of work. While recording we noticed a type of channel that was opened by intracellular calcium, like the calcium-activated potassium channel that was already well known in 1981. This one was a bit different because it was not selective for potassium. We hadn't expected to get a paper out of the vacation job but it seemed novel enough to write up, and 1982 being a year when anything with 'single channel' in the title, however trivial, sailed into *Nature*, and because we had a limited amount of data, we sent it there. Because we had all contributed much the same amount of work, we put the authors in alphabetical order. The analysis of the results, such as it was, was crude in the extreme (paper charts unrolled on the floor and measured with a ruler). If we hadn't seen this particular channel subtype, someone else would have done with a year or two. It just happened to be the first one of its type and so has been cited a lot, despite being scientifically trivial.

This example shows not only the iniquitous uselessness of the publication score used by Imperial; it also shows dramatically the almost equal uselessness of counting citations.

How not to get Nobel prizes

Employees of Imperial Medical School are told:

The divisional minimum benchmarks are 'To have sufficient papers in top rated journals in the speciality to ensure four publications in the RAE review period (2001–2007) with an Impact Factor of at least 5 and with no overlap with co-authors from Imperial'

The 'productivity' target for publications is "To publish three papers per annum including one in a prestigious journal with an impact factor of at least 5". Unfortunately, Dr X has published only two papers in 2006 ...

Let's see who lives up to their 'productivity' criterion.

Take, for example two scientists who command universal respect in my own field – Erwin Neher and Bert Sakmann. They got the Nobel Prize for Physiology or Medicine in 1991. In the 10 years from 1976 to 1985, Sakmann published an average of 2.6 papers per year (range 0 to 6). In 6 of these 10 years he failed to meet the publication target set by Imperial, and these failures included the years in which the original single channel paper was published (Neher & Sakmann, 1976) and also the year when Colquhoun & Sakmann (1985) was published.

In 2 of these 10 years he had no publications whatsoever.

On the other hand, a paper in 1981 in a journal with an 'unacceptable' impact factor of 3.56 has had over 15000 citations (Hamill *et al*. 1981). This paper would have earned for Sakmann a publication score of a miserable 0.71, less than 100th of our perspective in *Science*.

All this shows what is obvious to everyone but bone-headed bean counters. The only way to assess the merit of a paper is to ask a selection of experts in the field. Nothing else works. Nothing.

It seems to have escaped the attention of bean counters that this is precisely what has always been done by good grant giving agencies and search and promotion committees. Academics have always been assessed. But before HR departments and corporate-

academics got involved, it was done competently. Now a whole branch of pseudo-science has appeared which devotes itself to trying to find ways of assessing people without bothering to find out what they have done. 'Bibliometrics' is as much witchcraft as homeopathy. How long, one wonders, will it be before somebody coins the term 'bibliomics'? (Oops, a *Google* search shows I'm too late, some numbskull has already done it).

How to get good science

Universities will have to decide what sort of science they want.

They can bend their policies to every whim of the RAE; they can bow to the pressures for corporatisation from the funding council.

Or they can have creative scientists who win the real honours.

They cannot have both.

If they want to have the latter they will have to have universities run by academics. And they will have to avoid corporate and commercial pressures. They will have to resist the pressures to remove power from their best researchers by abolishing eminent departments and centralising power at a higher level. We have seen what this approach has done to the NHS, but it is a characteristic of the corporatising mentality to ignore or misuse data. They just know they are right.

It is also the box-ticking culture of managerialism that has resulted in approval of BSc degrees in anti-science (Colquhoun, 2007). Impressive sounding validation committees tick all the boxes, but fail to ask the one question that really matters: is what is being taught nonsense?

The policies described here will result in a generation of 'spiv' scientists, churning out 20 or even more papers a year, with very little originality. They will also, inevitably, lead to an increase in the sort of scientific malpractice that was recently pilloried viciously, but accurately, in the *New York Times*, and a further fall in the public's trust in science. That trust is already disastrously low, and one reason for that is, I suggest, pressures like those described here which lead scientists to publish when they have nothing to say.

Sakmann in Göttingen, 1980. He and Neher did the work *themselves*.

I wrote recently (Colquhoun, 2007) 'All of us who do research (rather than talk about it) know the disastrous effects that the Research Assessment Exercise has had on research in the United Kingdom: short-termism, intellectual shallowness, guest authorships and even dishonesty'. Now we can add to that list bullying, harassment and an incompetent box-ticking style of assessment that tends to be loved by HR departments.

This process might indeed increase your RAE score in the short term (though there is no evidence that it does even that). But, over a couple of decades, it will rid universities of potential Nobel prize winners.

David Colquhoun
Department of Pharmacology, University College London, UK

This article is an extended version of comments published in the *Times Higher Education Supplement* on 1 June 2007.

References

Colquhoun D (2003). Challenging the tyranny of impact factors. *Nature* **423** , 479.

Colquhoun D (2006). Playing the numbers game. A book review, of Does measurement measure up? *How numbers reveal and conceal the truth* by John M. Henshaw. *Nature* **442** , 357.

Colquhoun D (2007). Science degrees without the science. *Nature* **446** , 373–374.

Colquhoun D (2007). What have we learned from single ion channels? *J Physiol* **581**, 425–227.

Colquhoun D, Neher E, Reuter H, & Stevens CF (1981). Inward current channels activated by intracellular calcium in cultured cardiac cells. *Nature* **294** , 752–754.

Colquhoun D & Sakmann B (1981). Fluctuations in the microsecond time range of the current through single acetylcholine receptor ion channels. *Nature* **294** , 464–466.

Colquhoun D & Sakmann B (1985). Fast events in single-channel currents activated by acetylcholine and its analogues at the frog muscle end-plate. *J Physiol* **369** , 501–557.

Garfield E (1998). http://www.garfield.library.upenn.edu/papers/derunfallchirurg_v101(6)p413y1998english.html.

Hamill OP, Marty A, Neher E, Sakmann B, & Sigworth FJ (1981). Improved patch clamp techniques for high resolution current recording from cells and cell-free membrane patches. *Pflügers Arch* **391**, 85–100.

Hawkes AG, Jalali A, & Colquhoun D (1992). Asymptotic distributions of apparent open times and shut times in a single channel record allowing for the omission of brief events. *Philosophical Transactions of the Royal Society London* **B 337**, 383–404.

Hoeffel, C. (1998) Journal impact factors *Allergy* **53**, 1225.

Neher E & Sakmann B (1976). Single channel currents recorded from membrane of denervated frog muscle fibres. *Nature* **260**, 799–802.

New York Times. See http://www.dcscience.net/improbable.html#clone

Seglen PO (1997). Why the impact factor of journals should not be used for evaluating research. *Br Med J* **314**, 498–502.

Sivilotti LG & Colquhoun D (1995). Acetylcholine receptors: too many channels, too few functions. *Science* **269**, 1681–1682.

Comments on this article can be left at DC's Goodscience blog (http://www.dcscience.net/goodscience/?p=4) or at DC's Improbable Science blog (http://www.dcscience.net)

From lamprey swimming to human stepping: evolutionary conservation of longitudinally-propagated spinal activity

A diverse array of biomechanical systems has evolved to satisfy locomotory requirements (flight, reptation, swimming, walking, etc.) within the animal kingdom's different environments. Effectively, the ability to move from one place to another requires reconciliation between conflicting behavioural objectives, in that the seemingly straightforward demand of an animal to actively move towards a target necessitates the disruption of the fragile postural equilibrium that has to be dynamically adjusted as body displacement proceeds. Thus the successful achievement of locomotor movement requires the integrated functioning of all body segments, including the fore- and hind-limbs, trunk, head, and the synergistic action of many other muscle groups to ensure the appropriate positioning of the different body regions.

From numerous studies on a variety of vertebrate organisms, it is now well established that the basic motor patterns underlying limb and trunk movements during locomotion are driven by central networks of neurons, so-called central pattern

Jean-René Cazalets (left) and Mélanie Falgairolle.

generators (CPGs), that for the fore- and hind-limbs of quadruped animals are located within the lumbar and cervical spinal cord regions, respectively. In limbless anguilliform animals such as tadpoles, lamprey and snakes, body propulsion is driven by alternate left-right trunk muscle contractions that occur sequentially in an anterior-to-posterior direction along the body length. The CPGs that control such rhythmic trunk bending have been shown to be segmentally-distributed along the whole spinal cord axis. In limbed vertebrates, however, very few studies have explored the role of trunk muscle contractions during locomotion, although a cyclic contraction of trunk muscles in time with rhythmic limb movements has been reported in various species (cat, rat and man).

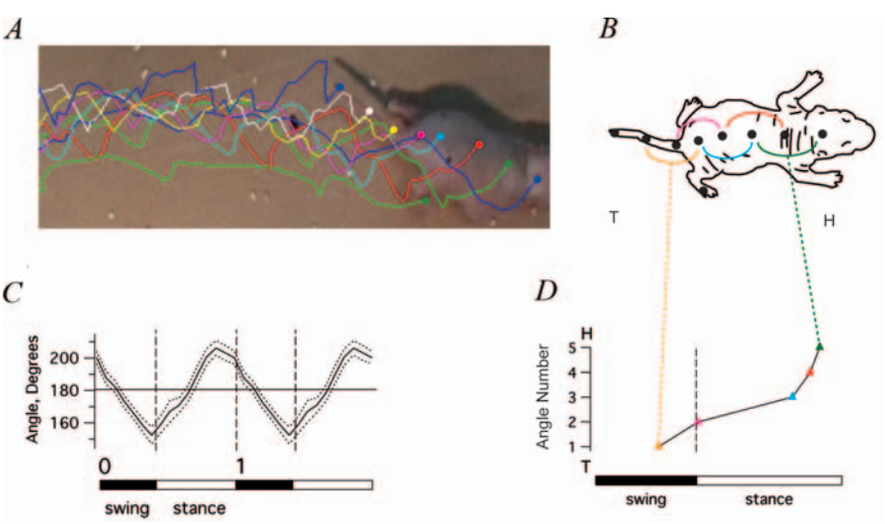


Figure 1. Two-dimensional kinematic analysis of trunk movements in newborn rat. (*A*) trajectories of dots during 4 s of walking. (*B*) Drawing indicating the position of analysed points and angles. (*C*) Mean angular change reflecting complete trunk bending during a step. (*D*) Plot of the maximum angle amplitude occurring during one cycle. Each colour corresponds to the angle in B.

We have recently begun to address a number of unresolved questions concerning the functional organization of spinal networks involved in trunk muscle activation during limb-based locomotion. What are the neural circuit substrates for coordinated hind-limb and trunk movements during actual locomotion in quadruped mammals, and to what extent have these neuronal networks been preserved during the course of evolution?

To explore the simultaneous functioning of the trunk and hind-limbs during actual locomotion, we have used both *in vivo* behavioural observations and *in vitro* electrophysiological approaches with the neonatal rat as our experimental model. In a first instance, to study coordinated back and tail movements during behaviour, rat pups were tagged with a series of ink dots placed along the dorsal axial line and were filmed during actual locomotion (Fig. 1*A*, 1*B*). By then determining the spatial coordinates of individual dots, regionally-specific body positions and angles could be computed. Since, the rat pup does not yet exhibit spontaneous locomotion at this age, forward progression was elicited by holding a tube containing nest material in front of the animal's nose. As the animal moves towards the scent, the experimenter slowly withdraws the tube so that the animal follows it. Our 2D kinematic analysis showed that there is a rhythmic sequential change in trunk curvature during each step cycle of locomotor activity (Fig. 1*C*). Moreover, this rhythmic trunk bending is propagated from tail-to-head and the bending amplitude is larger in the lower back than in the upper thoracic body region.

We next sought to identify the origin and central distribution of spinal motoneurons supplying axial (trunk) musculature in the newborn rat. This

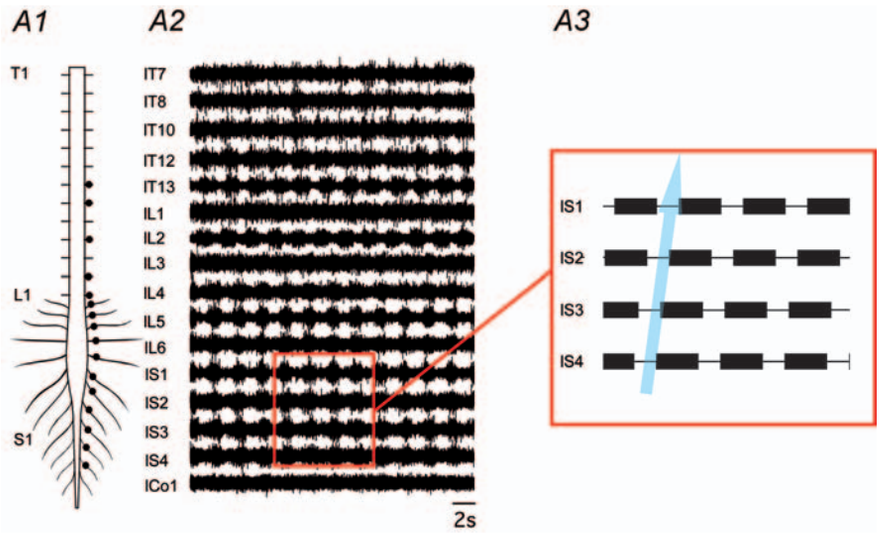


Figure 2. Simultaneous recordings of 16 ventral roots during locomotor-like activity showing a propagation wave from tail to head. *A1*, Drawing of the preparation with dots indicating electrode location. *A2*, Recordings of locomotor-like activity induced by bath-application of serotonin and excitatory amino acids. A3, Schematic representation of propagating process occurring in 4 sacral ventral roots. The arrow indicates the direction of the propagation. Horizontal black boxes in each trace indicate bursting duration during successive locomotor cycles.

was achieved by the intra-muscular injection of a retrograde marker (cholera toxin B subunit) which is taken up by motor axon terminals in the muscle and migrates into motoneuron cell bodies within the spinal cord over the ensuing 24 h. This revealed that motoneurons innervating the axial muscles are distributed along the entire spinal cord, from thoracic to coccygeal segments and including the lumbar segments where they co-localize with motoneurons that innervate the hind-limbs.

Finally, using isolated spinal cord preparations, we have analysed the functional interactions between the various cord regions during centrally-generated motor activity *in vitro*. Sequences of 'fictive' locomotion, i.e. rhythmic locomotor-related activity produced in the absence of actual movement (since the spinal cord is physically isolated from muscles and sensory inputs), were elicited by the bath-application of a mixture of the monoamine, serotonin, and excitatory amino-acids (Cazalets *et al.* 1992). The core networks that generate hindlimb locomotor activity in rat were previously found to be located in lumbar segments L1 and L2 of the spinal cord (Cazalets *et al.* 1996). To assess the global functioning of spinal circuitry and to understand how the thoracic, lumbar and sacral segments interact, we recorded from up to 16 ventral motor roots simultaneously along the thoraco-lumbo-sacral spinal cord axis (Fig. 2*A*1, 2*A*2). A major finding from this approach is that waves of rhythmic activity are propagated sequentially to the various ventral roots along the cord from sacral (posterior) to upper (anterior) thoracic segments (Fig. 2*A*3). This intra-spinal discharge in which one cord region is activated after another, has been termed 'metachronal' motor activity. *In vitro* experiments where the isolated cord was sectioned at different levels have revealed three zones (thoracic, lumbar, sacral) in which the spinal motor networks possess the intrinsic capacity for generating rhythmic motor output. This approach also provided important insights into the underlying synaptic organization. First, rhythmic burst generation continued to occur in the longitudinally-sectioned hemi-spinal cord, indicating that the central circuitry does not require cross-cord connections to generate locomotor-like output. Second, based on the relative timing of motor bursts, it appears that the travelling wave can not be explained solely on the basis of axonal conduction velocities and synaptic transmission delays. Rather, it is likely that both long multi-segmental neuronal pathways as well as local circuit interactions between adjacent segmental oscillators are involved in the coupling.

The data reported in our paper has allowed a functional diagram of interacting spinal networks to be proposed. In this model, the bursting properties of each spinal segment facilitate the transmission of motor activity along the spinal cord, while local intersegmental synaptic interactions also contribute to activity propagation. Cross-cord connections ensure bilateral coupling along the cord length, and long propriospinal pathways are also involved in coordination. The rostral part of the lumbar cord enlargement provides rhythmic locomotor-related output to rhythmic network elements in neighbouring segments that in turn convey and modulate output to their corresponding segmental motoneurons. In the intact spinal cord, the lumbar cord region probably imposes its own timing on the thoracic spinal cord generators, as it also does for the more rostral sacral segmental generators.

The first important conclusion of this study is that the rhythmic motor pattern that drives back muscle activation is centrally generated. Secondly, the metachronal nature of the propagating motor pattern is also centrally determined and remains strictly coordinated with hindlimb motor activity. Moreover, our data suggest that the networks responsible for the metachronal propagation of motor patterns during mammalian locomotion may correspond to those observed in lower vertebrates and even invertebrates, and thus appear to have been highly conserved during evolution. Interestingly, metachronal propagation is not only restricted to undulating or quadrupedal organisms. By investigating back muscle activity in humans we have recently found a comparable

sequential activation during various modes of forward bipedal locomotion (manuscript in preparation). Presumably, such a ubiquitous organization serves to assist in dynamic postural adjustments to ensure fluent body and limb movements during locomotion.

Mélanie Falgairolle
Jean-René Cazalets
Universités Bordeaux 2 and 1, CNRS, Bordeaux, France

References

Cazalets JR, Borde M & Clarac F (1996). The synaptic drive from the spinal locomotor network to motoneurons in the newborn rat. *J Neurosci* **16,** 298–306.

Cazalets JR, Sqalli-Houssaini Y & Clarac F (1992). Activation of the central pattern generators for locomotion by serotonin and excitatory amino acids in neonatal rat. *J Physiol* **455,** 187–204.

Falgairolle M & Cazalets JR (2007). Metachronal coupling between spinal neuronal networks during locomotor activity in newborn rat. *J Physiol* **580**, 87–102.

In brief

Book discount

The Physiological Society has teamed up with Wiley-Blackwell to offer Members, Affiliates and Associates of The Society a 25% discount on book purchases. For more information visit the Members' area of the web site. Access for Affiliates and Associates will be available shortly via the new web site. In the meantime please contact Sarah Ollington for further details (sollington@physoc.org)

Life science blogs

The number of life science blogs is steadily increasing and there is little guidance for newcomers on the must-read blogs in their area. *The Scientist* is therefore compiling a list of the best life science blogs. For more information and to vote on your favourite blog visit http://www.the-scientist.com

For news updates visit http://www.physoc.org/news

There goes the science bit ...

A group of young scientists hunt for the evidence behind advertising claims

Detox patches that 'draw out toxins from your body overnight' and pendants that 'balance your biofield' – almost all of us can give our own examples of science frustratingly or amusingly misrepresented in advertising. Discussing my own pseudoscience pet hates with a group of PhD and post-doctoral scientists, I realised I was far from alone in being fed up with junk science, and was thrilled when my colleagues rang companies and challenged them to back up their claims. As they shared the resulting conversations on the Voice of Young Science (VoYS) forum, the idea began to catch on, and transcripts of conversations with manufacturers and retailers ranging from obscure internet brands to big high street names began to roll in.

Amazingly, none of the companies called managed to provide any scientific evidence to back up their claims. Many made impressive statements about their products' abilities, but were unable to even explain how they worked. 'Detox patches' sold by the beauty spa *Champneys*, are designed to be stuck to your feet, and overnight turn from white to dark brown, supposedly indicating that 'toxins have been drawn out of your body.' As the 'harmful toxins' named included such essential substances as fatty acids, biologist Aarathi Prasad felt the claims required further investigation.

Several phone calls to *Champneys* and the patch manufacturer, *Trading Angel*s, informed Aarathi that the patches had 'been tested' (though they didn't know how, or with what results), and no-one could explain exactly how they worked. The fact that the patches contain wood vinegar, which turns brown on contact with moisture (for example, on sweaty feet), went unanswered.

Other companies were called to back up decisions they had made on behalf of their customers. The *Co-op* and *Sainsbury's* have both recently banned certain substances from their food and drink products (MSG and sodium benzoate respectively), implying that these may cause harm, despite a lack of any evidence to suggest this. When pressed, both companies admitted that the substances were removed due to 'customer concerns' and not any evidence of health risks.

Does it matter when companies misrepresent science? It is not just that such misleading or exaggerating can waste people's money, or even affect their health. People trust these companies. When *Sainbury's* or *Pret a Manger* decide to exclude 'obscure chemicals' because of public perceptions, they cause distrust in what scientists say, because *Pret* says otherwise. Customers expect advertisers to base claims on evidence, especially when scientific language is used (or misused) to make them sound more credible. When companies behave as if evidence does not matter, they are not only deceiving their customers, they are devaluing science for everyone.

Like the Voice of Young Science group, we need to let companies know that they are accountable for their misuse of science. If we scrutinise these claims, advertisers are forced to ensure that they have the evidence to back them up. As VoYS has shown, it only takes a phone call to challenge your own pseudoscience pet hate. If not you, then who?

VoYS has produced *There goes the science bit...* - a dossier of their amusing, eye-opening and sometimes shocking experiences, hoping to inspire others to stand up for science too. For a free copy, or more information, go to www.senseaboutscience.org or contact Alice Tuff at VoYS@senseaboutscience.org

Kate Oliver
Member, Voice of Young Science

Brain rhythms, synaptic plasticity and sleep

EEG recordings from the scalp disclose rhythmic electrical activity resulting from repetitive discharges of thousands of interconnected neurons in the forebrain. The frequency and synchrony of these rhythms change with the state of vigilance. During wakefulness and rapid-eye movement (REM) sleep fast oscillations (10–40 Hz) of low amplitude are prevalent, while during non-REM sleep slower rhythms (0.1–4 Hz) are highly synchronized over large areas of the forebrain. Associated with these state-dependent changes in global brain activity are character-istic alterations in the firing patterns of thalamic and cortical neurons (Steriade *et al.* 2001). During fast EEG rhythms, i.e. wakefulness and REM sleep, action potentials are generated as continuous trains of individual spikes (tonic mode), while during non-REM sleep they are emitted as high frequency bursts (burst mode) where action potentials are grouped together by a transient depolarizing potential (Fig. 1*A*).

Daniel Ulrich

While fast oscillations during wakefulness have been implicated in sensory processing and memory formation (Gray & Singer, 1989) the slow rhythms seem to play a role in reorganizing neural networks during sleep. There is accumulating evidence, mainly from behavioral studies, that both REM and non-REM sleep have an important influence on memory formation and learning (Walker & Stickgold, 2004). According to the 'parallel' hypothesis, non-REM and REM sleep affect different types of memories, i.e. declarative and procedural, respectively. In the 'sequential' model all forms of memory are affected by both sleep episodes. The latter possibility would be in line with the alternating occurrence of non-REM and REM episodes during sleep and the fact that most experimental memory tasks inherently consist of a combination of different types.

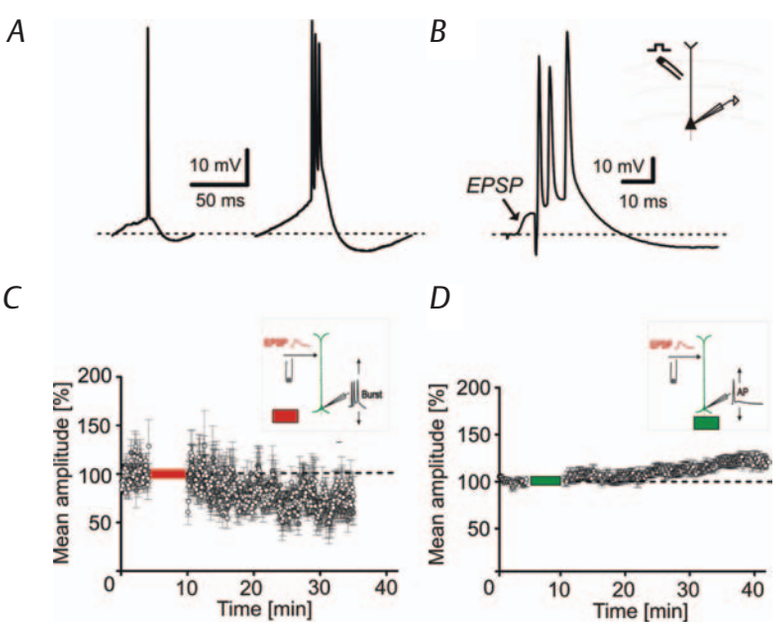


Figure 1. *A*, Action potentials can be generated as individual spikes (left) or high frequency bursts (right). *B*, The influence of single spike and burst firing on synaptic plasticity was assessed by pairing excitatory synaptic potentials (EPSPs) with single spikes or bursts. Composite EPSPs are elicited by extracellular stimulation and 'paired' repetitively with a postsynaptic action potential or spike burst (figurine). *C*, *D*, EPSP-amplitude time series illustrating the impact of single spike-pairings (green) and burst-pairings (red) on synaptic strength. The pairing periods are indicated by horizontal bars and outlined schematically in the figurine.

It is widely accepted that changes in synaptic connectivity underlie memory formation and learning (Rioult-Pedotti *et al.* 2000). In particular, long-term potentiation (LTP) and depression (LTD) are long-lasting increases or reductions in synaptic strength that are considered to be important cellular mechanisms underlying learning and memory formation. How, then, is synaptic strength influenced by the various discharge patterns observed during sleep?

Recent experiments in layer V pyramidal cells of rat somatosensory cortex *in vitro* have used a stimulation paradigm where excitatory synaptic potentials (EPSPs) were elicited in close temporal proximity with single action potentials or burst-discharges to mimic the near-synchronous activity of synaptically coupled neurons during wakefulness or sleep, respectively (Fig. 1*B*) (Birtoli & Ulrich, 2004). The results show that associations of EPSPs with single spikes predominately lead to LTP while associations of EPSPs with bursts induce an enduring reduction of the excitatory synaptic inputs (burst-LTD) (Fig. 1*C*, *D*). This suggests that firing patterns characteristic of wakefulness and sleep differentially affect synaptic plasticity. Pharmacological experiments revealed that while LTP was reliant on the NMDA (N-methyl-D-aspartate) subtype of glutamate receptors, burst-LTD depended on the concomitant activation of metabotropic group I glutamate receptors and low-threshold activated Ca^{2+} channels (Czarnecki *et al*. 2007) (Fig. 2*A*). The concurrent activation of both signalling pathways leads to a release of Ca^{2+} from internal stores and activation of

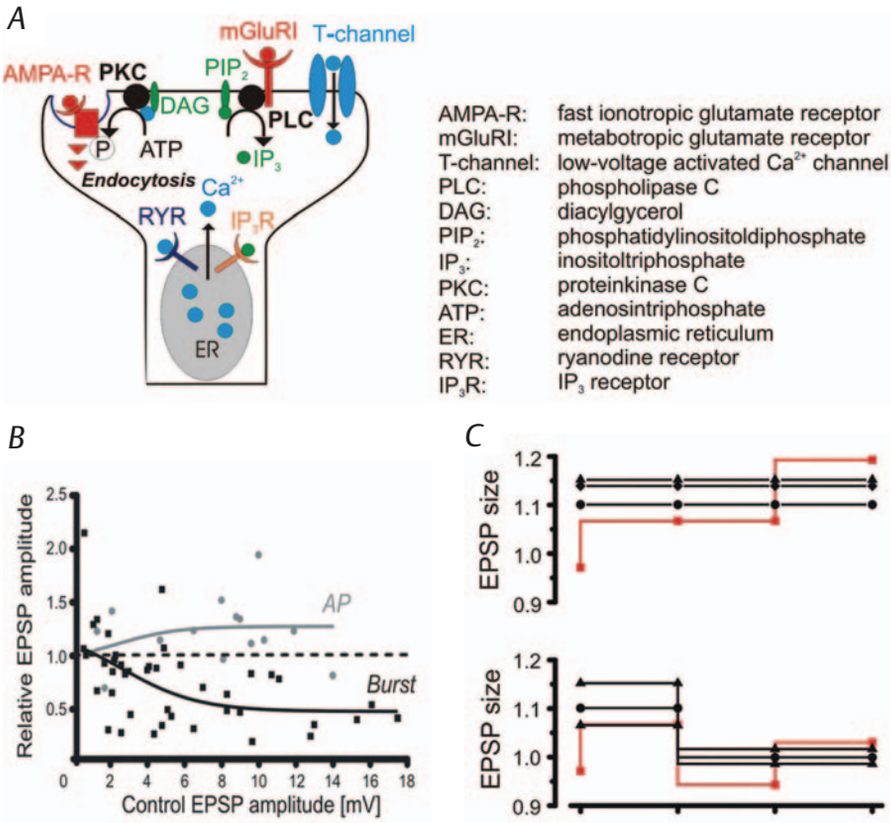


Figure 2. *A*, Schematic outline of the signaling pathway involved in burst-LTD. Coactivation of metabotropic glutamate receptors and T-type Ca^{2+} channels leads to Ca^{2+} release from internal stores and activation of proteinkinase C. Phosphorylation of AMPA receptors by this enzyme leads to their removal via endocytosis. *B*, Relationship between the initial size of the EPSP and the amount of LTP and burst-LTD. *C*, Hypothetical evolution of synaptic weights undergoing LTP (red) or no change (black) without (above) or with (below) synaptic rescaling. Note the relative synaptic weight compression in the lower graph.

protein kinase C (PKC). This enzyme promotes the endocytosis of AMPA receptors, which is the ultimate cause of a long-lasting depression of excitatory synaptic inputs (Fig. 2*A*). The concomitant requirement for presynaptic (i.e., neurotransmitter release) and postsynaptic (action potential bursts) activity explains the associative nature of burst-LTD (Birtoli & Ulrich, 2004).

Further experiments revealed that the amount of LTP is proportional to the initial EPSP size, while burst-LTD was inversely related to the baseline amplitude of the EPSP (Fig. 2*B*). Downscaling of synaptic weights has been postulated as a major mechanism by which non-REM sleep contributes to memory consolidation (Tononi & Cirelli, 2003). Indeed, it was recently found that sleep-related improvement of a trained motor task is accompanied by an overall decrease of cortical spike activity (Fischer *et al.* 2005). This may indicate that memory improvement may be associated with a relative strengthening of particular connections by reducing the overall effectiveness of synaptic inputs. Indeed, preliminary data *in vitro* suggest that burst-LTD affects homo- and hetero-synaptic inputs as expected for such a rescaling mechanism. Another potential important role of synaptic downscaling is to keep synaptic strengths within a physiological dynamic range. This would prevent synapses from being driven into saturation and thus allow their reuse over consecutive waking periods (Fig. 2*C*). It has been shown previously that LTP can be saturated and that this impedes further learning (Moser *et al.* 1998). Thus a synaptic rescaling process is likely to be of physiological relevance.

While we have shown that characteristic sleep/wake discharge patterns differentially influence synaptic strength, other factors like state-dependent changes in neuromodulator concentrations are likely to affect synaptic plasticity as well.

Overall, there is growing evidence from different fields in neuroscience for a role of sleep in reorganizing neural circuits associated with memory formation. This would add another dimension to the function of sleep that is more traditionally believed to merely subserve energy replenishment.

Daniel Ulrich
Department of Physiology, Institute of Neuroscience, Trinity College, Dublin 2, Ireland

References

Birtoli B & Ulrich D (2004). Firing mode-dependent synaptic plasticity in rat neocortical pyramidal neurons. *J Neurosci* **24**, 4935–4940.

Czarnecki A, Birtoli B, & Ulrich D (2007). Cellular mechanisms of burst firing-mediated long-term depression in rat neocortical pyramidal cells. *J Physiol* **578**, 471–479.

Fischer S, Nitschke MF, Melchert UH, Erdmann C & Born J (2005). Motor memory consolidation in sleep shapes more effective neuronal representations. *J Neurosci* **25,** 11248–11255.

Gray CM & Singer W (1989). Stimulus-specific neuronal oscillations in orientation columns of cat visual cortex. *Proc Natl Acad Sci USA* **86,** 1698–1702.

Moser EI, Krobert KA, Moser MB & Morris RG (1998). Impaired spatial learning after saturation of long-term potentiation. *Science* **281,** 2038–2042.

Rioult-Pedotti MS, Friedman D & Donoghue JP (2000). Learning-induced LTP in neocortex. *Science* **290,** 533–536.

Steriade M, Timofeev I & Grenier F (2001). Natural waking and sleep states: a view from inside neocortical neurons. *J Neurophysiol* **85,** 1969–1985.

Tononi G & Cirelli, C (2003). Sleep and synaptic homeostasis: a hypothesis. *Brain Res Bull* **62,** 143–150.

Walker MP & Stickgold, R (2004). Sleep-dependent learning and memory consolidation. *Neuron* **44,** 121–133.

Tropomyosin mutations responsible for muscle weakness in inherited skeletal muscle diseases

Several mutations have been identified in tropomyosin, a key regulatory protein in muscle fibres. These mutations specifically modify muscle fibre function, and result in general muscle weakness accompanied with various inherited skeletal myopathies

In skeletal muscle fibres, tropomyosin, a regulatory protein, coils around the actin filament, relaying the information from the Ca^{2+} sensor, the troponin complex to the actin-myosin interactions, the cross-bridges. Tropomyosin alternates between three positions: the blocked, closed and open states.

In the absence of Ca^{2+}, tropomyosin is in a position on the outer domain of actin that sterically hinders the docking of cross-bridges – the blocked state. Full activation by reversal of steric blocking involves two additional states, requiring successive tropomyosin movements away from the blocked configuration. Ca^{2+} bindings to the troponin complex cause tropomyosin movement toward the inner domain of actin, exposing sites that allow weak binding of myosin heads while still inhibiting isomerization to the strong binding state. Following this change, tropomyosin still covers an essential part of the site, leaving it inaccessible to myosin heads – the closed state. Weak-to-strong myosin binding transition induces a second tropomyosin shift, permitting cooperative binding of additional myosin heads by exposing neighbouring sites – the open state.

Three major tropomyosin isoforms are expressed, α, β and γ, which are encoded by the *TPM1*, *TPM2*, and *TPM3* genes, respectively (Perry, 2001). The α isoform encoded by *TPM1* is predominantly expressed in cardiac and fast-twitch skeletal muscle fibres. The β isoform encoded by *TPM2* is mainly expressed in slow-twitch skeletal muscle fibres. The γ isoform encoded by *TPM3* is predominantly expressed in cardiac and slow-twitch skeletal muscle fibres. A number of

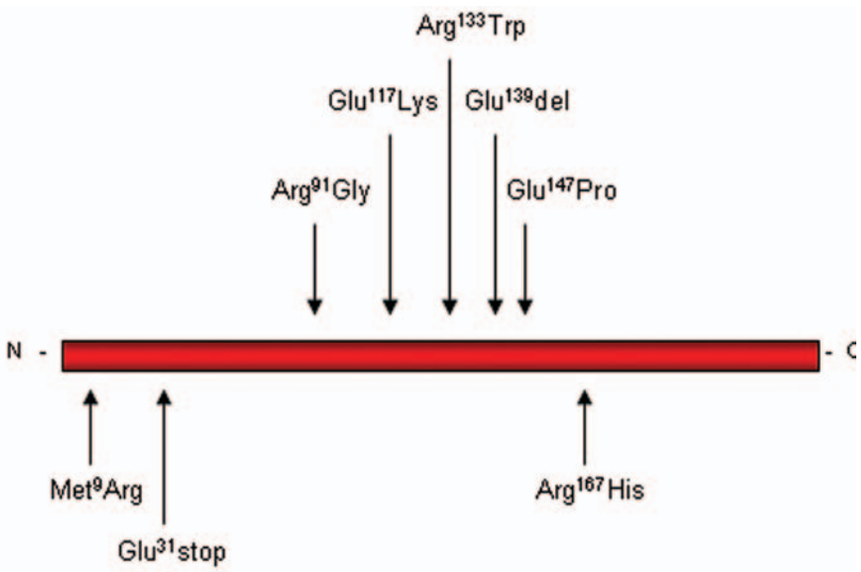


Figure 1. Mutations affecting β and γ tropomyosin isoforms. Eight mutations in the *TPM2* and *TPM3* genes have been associated with amino acids changes on the β ($Arg^{91}Gly$, $Glu^{117}Lys$, $Arg^{133}Trp$, $Glu^{139}del$, $Glu^{147}Pro$) and γ ($Met^{9}Arg$, $Glu^{31}stop$, $Arg^{167}His$) tropomyosin isoforms.

mutations in the *TPM1, TPM2*, and *TPM3* genes have been reported to cause inappropriate α, β and γ tropomyosin isoforms and inherited myopathies. Eight mutations in the *TPM1* gene induce amino acids changes on the α tropomyosin isoform ($Glu^{40}Lys$, $Glu^{54}Lys$, $Ala^{63}Val$, $Lys^{70}Thr$, $Val^{95}Ala$, $Asp^{175}Asn$, $Glu^{180}Gly$, $Glu^{180}Val$), and are not associated with skeletal myopathies but solely with cardiomyopathies. Eight mutations in the *TPM2* and *TPM3* genes have been associated with amino acids modifications on the β ($Arg^{91}Gly$, $Glu^{117}Lys$, $Arg^{133}Trp$, $Glu^{139}del$, $Glu^{147}Pro$) and γ ($Met^{9}Arg$, $Glu^{31}stop$, $Arg^{167}His$) tropomyosin isoforms and with general muscle weakness and skeletal myopathies (Fig.1).

Recent studies approach the mechanisms by which three mutations in β ($Arg^{91}Gly$, $Arg^{133}Trp$) and γ ($Met^{9}Arg$) tropomyosin isoforms cause general weakness in inherited diseases, in the absence of muscle wasting.

The $Arg^{91}Gly$ and $Arg^{133}Trp$ mutations on the β tropomyosin isoform appear to preserve the information from the Ca^{2+} sensor, the troponin complex to the cross-bridges. The Ca^{2+}-activation of the contractile proteins is unaltered as attested by the force-pCa relationships (Ochala *et al.* 2007; Robinson *et al.* 2007) in contrast to the $Met^{9}Arg$ mutation on the γ tropomyosin isoform where the force-pCa curve was shifted to the right (Michele *et al.*, 1999). The lack of effects of the $Arg^{91}Gly$ and $Arg^{133}Trp$ mutations on force-pCa relationships is surprising but may be related to their location in a region of the tropomyosin segment that is not directly associated with the troponin complex Ca^{2+} binding sites (Fig. 2). In skeletal muscle fibres, the critical interactions between tropomyosin and troponin T take place between

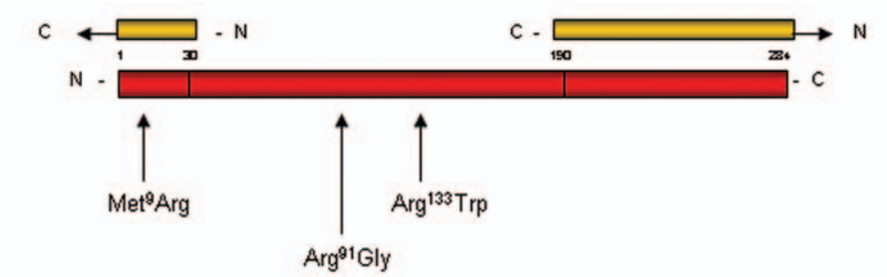


Figure 2. Tropomyosin segment. Troponin T (yellow) binds tropomyosin (red) in an overall anti-parallel manner, i.e. its C-terminal region is positioned near residue 190 of tropomyosin and the N-terminal tail of troponin T extends toward tropomyosin C-terminus, overlapping the head to tail joint of tropomyosin as well as 10–30 residues of the N-terminus of the next molecule along the filament.

tropomyosin residues 0–30 and 190–284. The $Arg^{91}Gly$ and $Arg^{133}Trp$ mutations on the β tropomyosin isoform are 60 residues away from the nearest potential Troponin T-anchoring region, in contrast to the $Met^{9}Arg$ mutation on the γ tropomyosin isoform (Fig. 2) (Perry, 2001).

The $Arg^{91}Gly$ and $Arg^{133}Trp$ mutations on the β tropomyosin isoform appear to directly modify the cross-bridge kinetics as attested by the decreased maximal apparent rate constant of force redevelopment and increased maximum unloaded shortening velocity in fibres containing $Arg^{133}Trp$ (Ochala *et al.* 2007) or increased maximal ATPase rates in fibres expressing $Arg^{91}Gly$ (Robinson *et al.* 2007). The mutations from Arg to Gly or Arg to Trp replace charges predicted to stabilize tropomyosin coiled-coil (Tajsharghi *et al.* 2007). Therefore, the $Arg^{133}Trp$ mutation may affect tropomyosin movement toward the inner domain of actin and the transition from the blocked state to the closed and open states. This would disrupt attachment and/or transition of myosin heads from the weakly to the strongly bound force-generating state, decreasing the cross-bridge attachment rate and the maximal apparent rate constant of force redevelopment. Moreover, the $Arg^{91}Gly$ and $Arg^{133}Trp$ mutations may affect tropomyosin movement from the open to the blocked state, slowing myosin attachment and promoting dissociation of myosin heads from the actin filament; thereby increasing the cross-bridge detachment rate and maximum unloaded shortening velocity or maximal ATPase rates. The combination of a decreased apparent rate of force redevelopment and an increased maximum unloaded shortening velocity in fibres expressing $Arg^{133}Trp$ induces a shortened cross-bridge duty cycle, i.e. a shorter time spent by myosin heads in a strongly bound force-generating state, resulting in a decreased force generating capacity.

In conclusion, studies on the three $Arg^{91}Gly$, $Arg^{133}Trp$ and $Met^{9}Arg$ mutations in β and γ tropomyosin isoforms suggest specific mechanisms underlying the weakness, in the absence of muscle wasting, in patients carrying these mutations. However, the mechanism is mutation-specific and results obtained from one mutation cannot be generalized to other tropomyosin mutations. Further, the results from these mutations analyses indicate the critical role of tropomyosin isoform expression in modulating muscle contraction under physiological conditions.

Acknowledgements

The project was supported by grants from the Swedish Institute and Association Française contre les Myopathies to J O, from the Association Française contre les Myopathies and Swedish Research Council (07122) to A O, and from the Swedish Research Council (08651), Association Française contre les Myopathies, Cancer Foundation and National Institutes of Health (AR045627, AR047318) to L L.

Julien Ochala[1]
Anders Oldfors[2]
Lars Larsson[1, 3]

[1]Department of Clinical Neurophysiology, Uppsala University Hospital, Sweden
[2]Department of Pathology, Sahlgrenska University Hospital, Göteborg, Sweden
[3]Center for Development and Health Genetics, The Pennsylvania State University, University Park, USA

References

Michele DE Albayya FP & Metzger JM (1999). A nemaline myopathy mutation in alpha-tropomyosin causes defective regulation of striated muscle force production. *J Clin Invest* **104,** 1575-1581.

Ochala J, Li M, Tajsharghi H, Kimber E, Tulinius M, Oldfors A & Larsson L (2007). Effects of a R133W beta-tropomyosin mutation on regulation of muscle contraction in single human muscle fibres. *J Physiol* **581,** 1283-1292.

Perry SV (2001). Vertebrate tropomyosin: distribution, properties and function. *J Muscle Res Cell Motil* **22,** 5-49.

Robinson P, Lipscomb S, Preston LC, Altin E, Watkins H, Ashley CC & Redwood CS (2007). Mutations in fast skeletal troponin I, troponin T, and beta-tropomyosin that cause distal arthrogryposis all increase contractile function. *Faseb J* **21,** 896-905.

Tajsharghi H, Kimber E, Holmgren D, Tulinius M & Oldfors A (2007). Distal arthrogryposis and muscle weakness associated with a beta-tropomyosin mutation. *Neurology* **68,** 772-775.

Muscle's silent sensors

This is a story about measurement of position sense at the elbow joint. If I cannot see my arm, where is it? The object of our studies has been to understand the changes in position sense seen after exercise and during loading of the arm. Our choice of the elbow joint is simply out of convenience. Some recent experiments using the knee joint and the quadriceps muscle have shown that the principles we have considered at the elbow joint also applies at other joints.

We have been using a property of muscle, called thixotropy, as a tool for investigating position sense (*Physiology News* **66**, 20–22). Another word for thixotropy is muscle history-dependence. This is a method which allows us, at a given muscle length, to change the levels of background activity in muscle spindles of elbow muscles by contracting them while they are held short or long and then returning them to the initial length. Muscle spindles are believed to be largely responsible for signalling to the brain muscle length and therefore elbow angle. The changes in background activity following conditioning lead subjects to think that muscle length has changed and so they make errors in limb position sense.

The way we have been doing the experiments is to use a position matching task. The experimenter places one forearm (reference) at a given angle and the subject is asked to match its position by placement of their other arm (indicator). One fact that emerged in recent experiments (Allen *et al.* 2007) is that it is necessary to consider the muscle history-dependence of both reference and indicator arms.

In the recent experiments we began to systematically control for muscle history effects in both arms. That has revealed an unexpected distribution of position errors. For these experiments, to avoid the effects of gravity, measurements were made in the horizontal plane. First, when contraction history in both arms was made the same, unexpectedly, position errors did not reduce to zero. So, for example, in Fig. 1 when both arms were conditioned by contracting elbow flexors with the arms held flexed (flexion conditioning), the subjects consistently placed their indicator arm in a position that was slightly more flexed (blue symbol) than the position adopted by the reference arm (dotted line). A second observation made was that when the reference arm was again flexion conditioned, but this time the indicator arm had its elbow extensors contracted with the arm held extended (extension conditioning), the subject thought that their reference arm was much more extended that it really was (red

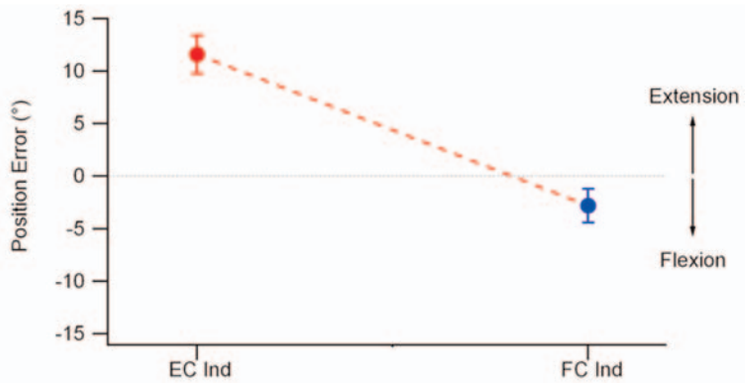


Figure 1. Position matching errors in the horizontal plane. Matching errors were calculated as the difference in position between the reference arm, set at 85°, and the indicator arm placed by the blindfolded subject in a matching position. Errors were scored as positive when the indicator was placed in an extended position relative to the reference arm, and negative when it was in the direction of flexion. Dotted line, zero error. Red symbol, position errors (± SEM) for eight subjects when the reference arm had been conditioned in a flexed position (FC), while the indicator had been conditioned with the arm held extended (EC). Blue, symbol, errors when both arms had been flexion conditioned. Dashed line indicates the trend in the errors. Data from Allen *et al.* (2007).

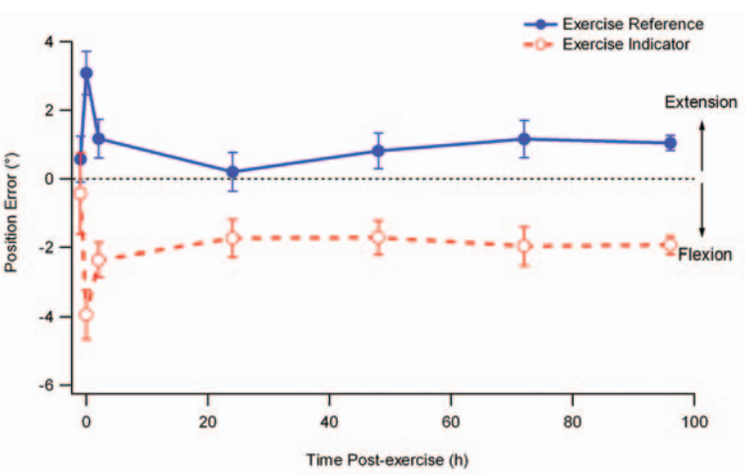


Figure 2. Position matching errors in the vertical plane after eccentric exercise. Blue symbols, mean (± SEM) errors for six subjects when the reference arm was the exercised arm, red symbols when the indicator arm was the exercised arm. Errors measured before, immediately after the exercise (0 h), at 20 h, 40 h, 60 h and 100 h. Data points have been joined by lines for clarity. Subjects held their arms, unsupported. Errors were scored as positive when the indicator arm adopted a position more extended that the reference. Errors were negative when it was more flexed. Dotted line, zero error. Data from Walsh *et al.* (2004).

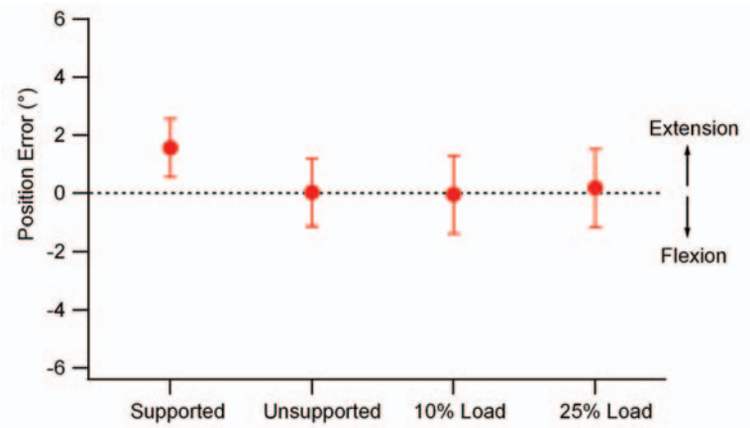


Figure 3. Position matching errors in the vertical plane. Values are means (± SEM) for 12 subjects. Matching errors in the direction of extension are shown as positive. Dotted line, zero error. For this experiment both arms had been flexion conditioned before each matching trial. The four conditions for the reference arm were: supported by the experimenter, supported by the subject, supported by the subject with a 10% maximum voluntary contraction load added, and supported by the subject with a 25% load added (from Allen *et al.* 2007).

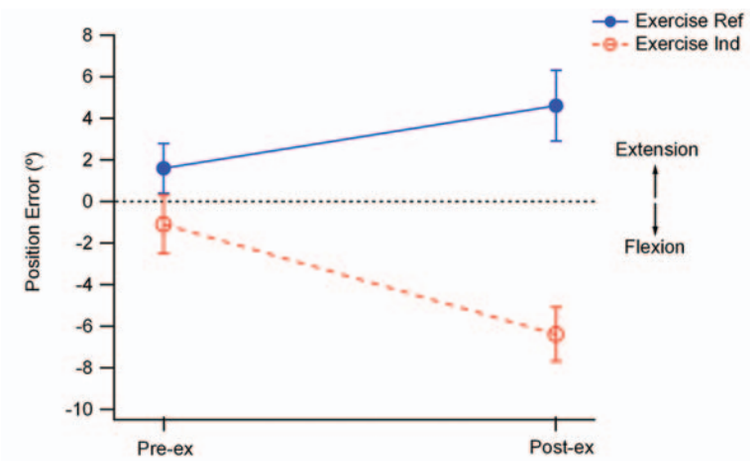


Figure 4. Position matching errors in the vertical plane after eccentric exercise. Each point is the mean (± SEM) value for 9 subjects. Before each match both arms had been flexion conditioned. Errors were scored as positive when the indicator arm was placed in the direction of extension relative to the reference, negative, when placement was in the direction of flexion. Dotted line, zero error. Pre-exercise, values before the exercise; Post-exercise, values immediately after the exercise. Blue symbols, when the exercised arm acted as the reference, red symbols, when the exercised arm acted as the indicator (from Allen *et al.* 2007).

symbol). In other words, by simply changing the form of conditioning of the indicator arm, subjects believed that arm position had shifted by over 14°. These errors can be satisfactorily explained by changes in background activity of muscle spindles of both arms, as a result of the thixotropic property of their intrafusal fibres (Allen *et al.* 2007). That conclusion led us to pay a lot more attention to the state of conditioning of the two arms.

An issue that has fascinated me for a long time is the question, how can muscle spindles act as position sensors during muscle contraction? Muscle spindles, as stretch receptors, are able to signal the length of a muscle. At the same time, because they have an inbuilt sensitivity control, the fusimotor system, they are able to change their discharge without any accompanying length change. The problem was, how did the central nervous system distinguish between activity arising from muscle length changes and from intrafusal contractions?

The current accepted view is based on a model put forward by McCloskey *et al*. (1983). The idea is that whenever the central nervous system generates activity in fusimotor neurons this is subtracted from the gross spindle discharge, to leave only the length-related signal to reach consciousness. Spindles are activated through the fusimotor system whenever we carry out a voluntary contraction.

There are reasons for thinking that what is going on is more complicated than that. Here we were guided by some experiments on the effects of eccentric exercise on position sense (Walsh *et al.* 2004). These experiments involved a position matching task in the vertical plane. After eccentric exercise of one arm this was weakened as a result of the muscle damage and fatigue following the exercise (Proske & Morgan, 2001). When the exercised arm was the indicator, the subject made significant matching errors, placing it in a position that was more flexed than the position adopted by the reference arm (Fig. 2). Conversely, when the reference arm was fatigued matching errors lay in the direction of extension. To explain the result we proposed that when an arm was fatigued, it required more effort to hold it in a given position and the effort signal provided positional information. When the indicator was fatigued, the greater effort required to support its weight was interpreted as the arm being in a more extended position, where the gravity vector acting on it was greater. So in making a match, subjects flexed their indicator to a point where the effort signals in the two arms matched, leading to errors in the direction of flexion. Predictably, errors reversed when it was the reference arm that had been exercised (Fig. 2). So we were proposing that during a muscle contraction the errors in position sense arose from a new position signal derived from a centrally generated sense of effort.

A simple and direct test of the effort hypothesis was to examine the effects of loading the arm by the addition of weights. Increasing the load should increase the effort required to support the arm and

therefore produce position errors. In this new experiment we were careful to make sure from the outset that for each trial both arms had been conditioned identically. Then, when one arm was loaded during the match, position errors seen previously (Winter *et al.* 2005) were no longer present (Fig. 3).

Presumably in the previous study the observed errors had been thixotropy-related. Subsequently we repeated the experiment on eccentric exercise, again making sure that both arms had first been identically conditioned. (Fig. 4). When the reference arm had been exercised, error lay in the direction of extension. When the indicator was exercised they lay in the direction of flexion (Allen *et al.* 2007). This was the same pattern as reported by Walsh *et al*. (2004), but now we were no longer invoking the sense of effort as responsible for the errors. If effort had played a significant role it should have shown up in the experiments of loading the arm, which it did not.

How do we explain all of this? Sticking to the simple result of Fig. 3, the conclusion is reached that in the absence of vision we are able to locate, equally accurately, the position of our arms, whether they are loaded or not, provided that beforehand they have been conditioned identically. So, by inference, whether arm muscles are contracting or not does not alter our sense of forearm position. Given that during voluntary contractions the fusimotor neurons are co-activated, the result could be interpreted as evidence in support of the operation of the M^{c}Closkey *et al*. (1983) subtraction model.

But there were bits of evidence which did not quite fit such an interpretation. Experiments on the paralysed, anaesthetised arm had shown that a large position signal of central origin was available when peripheral afferent activity had been blocked (Gandevia *et al.* 2006). Presumably with such a block in place, the command signals for fusimotor co-activation during attempted movements would still be present and any subtraction from an absent afferent signal might have been expected to lead to a negative outcome, perceived movements in a direction opposite to that which had been willed (Gandevia *et al.* 1993). That was not the case. It led us to consider another explanation and here we were attracted by propositions involving an internal forward model (Bays & Wolpert, 2007).

The idea is simple enough. As soon as a voluntary contraction of arm muscles accompanies the movement, or maintained position, an efference copy of the motor command signal to the muscle becomes available. The efference copy accesses memory stores of similar movements carried out in the past to compare the afferent feedback that had accompanied those movements with the feedback during the present movement. Only the difference signal reaches consciousness. When the anticipated and fed-back signals are the same, the posture is confirmed with no accompanying sensation of a change in position, thus the label of muscle receptors as 'silent sensors'.

Finally, it is necessary to explain the effects of exercise. We have now done exercise experiments on both arm muscles and leg muscles. The interpretation that is consistent with all of the observations is that we perceive an exercised muscle as longer than it actually is. For the arm, when the elbow flexors have been exercised, the arm is perceived as more extended (longer flexors) than is really the case. Our interpretation, in terms of the operation of an internal forward model, is that the afferent feedback for maintenance of a given position is higher, as a result of fatigue, and that is interpreted as a longer muscle.

We are now on the lookout for experiments that will test the forward model hypothesis more directly. What happens when the muscle is paralysed, that is, afferent feedback is present, but deprived of fusimotor activation? Will there be negative effects this time? Whatever the outcome, there are two important lessons from all of this. One is that the effects of muscle thixotropy on muscle spindle discharges can lead to misinterpretation of observations on limb position sense, and secondly, we should not underestimate the level of sophistication involved in the central processing of proprioceptive information.

Uwe Proske
Trevor Allen
Department of Physiology, Monash University, Clayton, Victoria, Australia

References

Allen TJ, Ansems GE & Proske U (2007). Effects of muscle conditioning on position sense at the human forearm during loading or fatigue of elbow flexors and the role of the sense of effort. *J Physiol* **580.2,** 423–434.

Bays PM & Wolpert DM (2007). Computational principles of sensorimotor control that minimize uncertainty and variability. *J Physiol* **578,** 387–396.

Gandevia SC, Killian K, McKenzie DK, Crawford M, Allen GM, Gorman RB & Hales JP (1993). Respiratory sensations, cardiovascular control, kinaesthesia and transcranial stimulation during paralysis in humans. *J Physiol* **470,** 85–107.

Gandevia SC, Smith JL, Crawford M, Proske U & Taylor JL (2006). Motor commands contribute to human position sense. *J Physiol* **571,** 703–710.

McCloskey DI, Gandevia SC, Potter EK & Colebatch JG (1983). Muscle sense and effort: motor commands and judgements about muscular contractions. In *Motor Control Mechanisms in Health and Disease*, ed. Desmedt JE, pp. 151–167. Raven Press, New York.

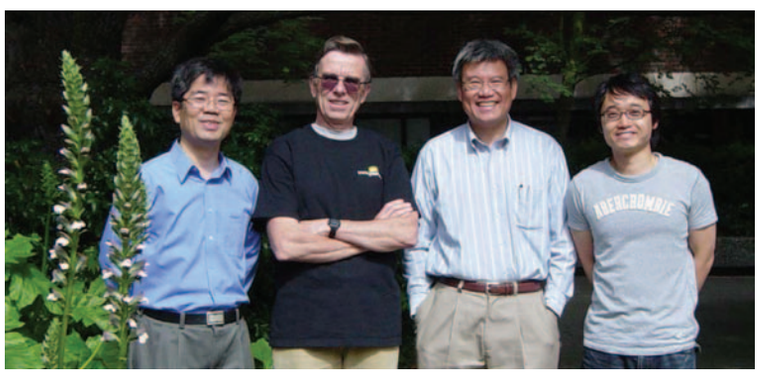
Proske U & Morgan DL (2001). Muscle damage from eccentric exercise: mechanism, mechanical signs, adaptation and clinical applications. *J Physiol* **537,** 333–345.

Walsh LD, Hesse CW, Morgan DL & Proske U (2004). Human forearm position sense after fatigue of elbow flexor muscles. *J Physiol* **558,** 705–715.

Winter JA, Allen TJ & Proske U (2005). Muscle spindle signals combine with the sense of effort to indicate limb position. *J Physiol* **568,** 1035–1046.

The pattern of intracellular Ca^{2+} signal matters in epithelial secretion

Endocrine and exocrine cells respond to external cues by releasing different secretory molecules. Seung-Ryoung Jung and colleagues analyzed how ductal epithelia encode the external inputs into Ca^{2+} signals and decode them into specific types of secretion



Duk-Su Koh (left), Bertil Hille, Toan Nguyen, and Seung-Ryoung Jung

Calcium regulates many cellular processes including muscle contraction, fertilization, gene expression, membrane excitability, and exocytosis (Berridge *et al*. 2003). To modulate a variety of target molecules selectively in a cell, the Ca^{2+} signal itself has to carry discriminatory information. Sometimes selectivity is achieved by subcellular localization of the signal, and sometimes by the pattern of the signal. In electrically excitable cells, Ca^{2+} signals are typically determined by the activity of voltage-gated Ca^{2+} channels that mediate Ca^{2+} fluxes from the extracellular space into the cytoplasm. In non-excitable cells equipped with G-protein coupled receptor (GPCR)-linked IP_3 generation, Ca^{2+} is often mobilized from internal stores by external agonists. The detailed time course of cytoplasmic free Ca^{2+} concentration ($[Ca^{2+}]_i$) is further modified by Ca^{2+} binding proteins and by transmembrane movements of Ca^{2+} ions between the cytoplasm, intracellular stores, other organelles, and the extracellular space. Ca^{2+} signals generated by GPCRs are often encoded as a graded increase of $[Ca^{2+}]_i$, with higher concentrations of agonists eliciting higher peak amplitudes of $[Ca^{2+}]_i$ ('amplitude' modulation, AM). In addition, a signal can be conveyed by oscillations of $[Ca^{2+}]_i$, where agonist concentration determines the frequency of oscillations ('frequency' modulation, FM). The oscillation frequency seems to have a significant role in modulating gene expression (Dolmetsch *et al*. 1998) and certain enzymes, including Ca^{2+}-dependent intramitochondrial dehydrogenases and protein kinase C (Hajnóczky *et al*. 1995; Oancea & Meyer, 1998). In general, these Ca^{2+}-dependent enzymes are partially activated by each Ca^{2+} spike and then deactivated slowly as Ca^{2+} declines. If the next Ca^{2+} spike occurs before the enzyme is fully deactivated, there is a cumulative increase of enzyme activity.

Pancreatic ducts collect digestive enzymes released from acinar cells, delivering them to the duodenum. Pancreatic duct epithelial cells (PDEC) lining the ducts add an important component to the digestive fluid, HCO_3^- (bicarbonate), which neutralizes acidic chyme from the stomach. Bicarbonate secretion from PDEC is mediated by ion channels and transporters (Nguyen *et al*. 1998). In addition, the cells secrete mucin, a major protein of the mucus layer, via exocytosis (Nguyen

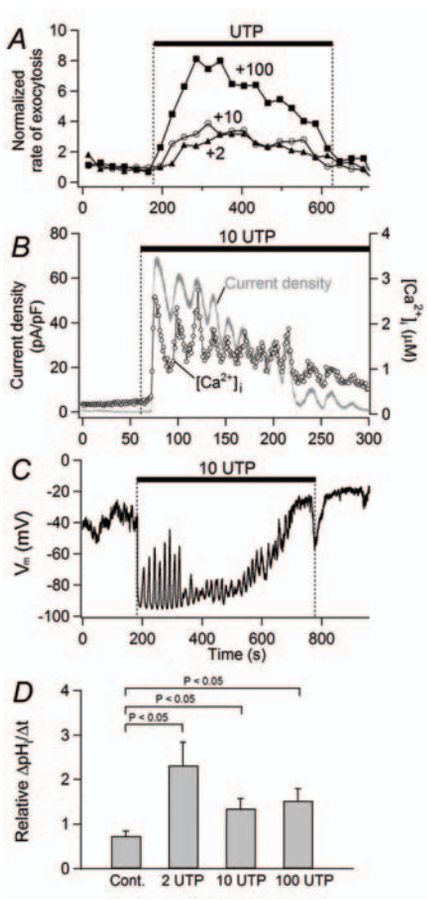


Figure 1. Differential effects of purinergic input on HCO_3^- secretion and exocytosis in pancreatic duct epithelial cells. *A*, Exocytosis rates measured with microamperometry and normalized to the baseline value. Exocytosis induced by 2, 10, and 100 mM UTP is denoted as +2 (filled triangle), +10 (open circle), and +100 (filled square), respectively. *B*, UTP (10 mM) enhances $[Ca^{2+}]_i$ oscillations (measured with fura-2) and Ca^{2+}-activated K^+ current (measured simultaneously by whole-cell patch-clamp). *C*, UTP hyperpolarizes the membrane potential. The hyperpolarization was sensitive to charybdotoxin (CTX), a blocker of Ca^{2+}-activated K^+ channel (data not shown). *D*, UTP enhances HCO_3^- secretion measured with a pH-sensitive dye (BCECF). The intracellular compartment was first alkalinized by removing external HCO_3^- and then the intracellular pH (pH_i) was allowed to recover through secretion of HCO_3^-. The rate of the pH_i decrease (DpH_i/Dt) during the reacidification, a measure of HCO_3^- secretion, is expressed relative to the rate seen in the absence of agonist. Modified from Jung *et al*. (2006).

et al. 1998). Previous studies using PDEC monolayers demonstrated that $[Ca^{2+}]_i$ elevations, induced by endogenous purinergic receptors ($P2Y_2$ and $P2Y_{11}$) promote both electrolyte (K^+ and Cl^-) and mucin secretion (Nguyen *et al*. 1998). However, the single-cell Ca^{2+} dynamics were not resolved in those studies which used populations of PDEC. Our recent single-cell measurements reveal a complex Ca^{2+} signaling system that depends on the agonist concentration: a low concentration of ATP or UTP (2 or 10 μM) evokes $[Ca^{2+}]_i$ oscillations (Fig. 1*B*), whereas a high concentration of ATP or UTP (100 μM) evokes a sustained $[Ca^{2+}]_i$ elevation (Jung et al. 2004, 2006). As measured with single-cell microamperometry, the sustained $[Ca^{2+}]_i$ elevation stimulated exocytosis well, but the $[Ca^{2+}]_i$ oscillations were much less efficacious (Fig. 1*A*), despite reaching similar peak $[Ca^{2+}]_i$ levels (1-2 μM).

What is the role of oscillatory Ca^{2+} signals? We tested the hypothesis that Ca^{2+} oscillations open Ca^{2+}-activated K^+ and Cl^- channels expressed in PDEC. Both channels should be activated by micromolar levels of Ca^{2+}. As anticipated, electrophysiological studies in the whole-cell patch configuration revealed a periodic activation of K^+ channels synchronous with the $[Ca^{2+}]_i$ oscillations (Fig. 1*B*) and periodic hyperpolarizations of the membrane (Fig. 1*C*). We identified the channel as a Ca^{2+}-activated K^+ channel of intermediate conductance (IK1/SK4 subtype) using pharmacology, single-channel recording, and immuno-histochemistry. Finally, we reasoned that the hyperpolarization might accelerate secretion of HCO_3^-. Bicarbonate secretion from single cells was assayed indirectly using a pH-sensitive dye (BCECF). A higher rate of intracellular pH decrease ($\Delta pH_i/\Delta t$) should reflect increased HCO_3^- efflux from the cytoplasm to the extracellular compartment. We found that low (2 and 10 μM) and high (100 μM) concentrations of UTP stimulated HCO_3^- secretion to a similar extent, showing the equivalence of oscillatory and sustained $[Ca^{2+}]_i$ increases in modulating HCO_3^- secretion (Fig. 1*D*). Further, the UTP effect on HCO_3^- secretion was fully blocked by an IK channel blocker, charybdotoxin, suggesting that the K^+ channel and its hyperpolarizing effect are essential for HCO_3^- secretion. The underlying mechanism(s) for the electrogenic HCO_3^- secretion is not yet clear. The hyperpolarization could favor a lower intracellular Cl^- concentration that would accelerate the activity of Cl^-/HCO_3^- exchanger indirectly. Alternatively, the hyperpolarization could increase HCO_3^- efflux through HCO_3^--permeable anion channels directly. An argument in favor of the Cl^-/HCO_3^- exchanger was that blocking it with DIDS or by removing external Cl^- eliminated the effect of UTP on HCO_3^- secretion.

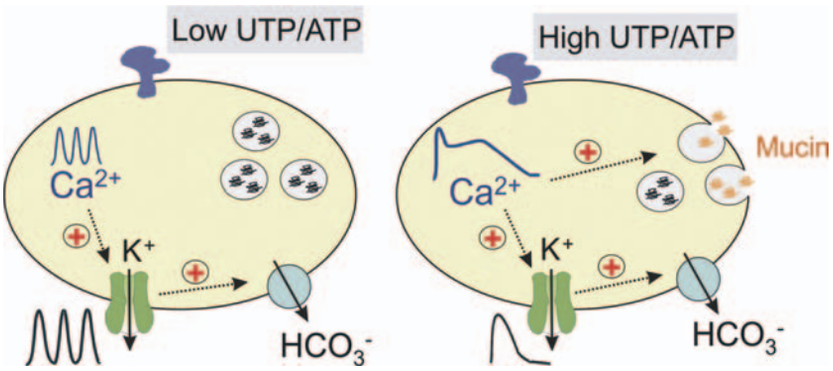


Figure 2. Transduction of stimuli into secretion in pancreatic duct epithelial cells. Low concentrations of UTP evoke $[Ca^{2+}]_i$ oscillations, and high concentrations of UTP induce a persistent $[Ca^{2+}]_i$ rise. The oscillatory Ca^{2+} signal causes HCO_3^- secretion by activating Ca^{2+}-activated K^+ channels and hyperpolarizing the membrane. The persistent $[Ca^{2+}]_i$ rise elicits both HCO_3^- secretion and exocytosis of proteins including mucin.

In summary, the intensity of external stimuli can be encoded into different patterns of $[Ca^{2+}]_i$ increases, to be decoded later into different biological outputs (Fig. 2). Low concentrations of purinergic agonists evoke mainly HCO_3^- secretion, whereas high concentrations evoke mucin secretion as well. These differential responses would be particularly relevant to the emerging autocrine and paracrine function of ATP, the major physiological activator of these receptors. The extracellular concentration of ATP depends on ATP release, diffusion, dilution, and local metabolism. Further investigation should show whether other receptors coupled to G_q and PLC (e.g. histamine H1, protease activated (PAR-2), muscarinic, or cholecystokinin receptors) alone or in combination elicit similar responses.

Physiological significance of the ductal input-output relations

Along with neutralization of acidic chyme, the ductal HCO_3^- probably also alters the rheologic properties of mucin secreted from the duct cells. The viscosity of mucin tends to increase at acidic pH. If secretion of HCO_3^- is impaired, as in cystic fibrosis, the luminal pH would fall and mucin released from PDEC might become more viscous and not be cleared from the epithelial surface. Formation of a mucin gel in the pancreatic ductal tree could lead to the blockage of the small ducts and eventual destruction of the gland. We therefore suggest that HCO_3^- and mucin secretion need to be well balanced. As UTP analogues have been advocated in the clinical

treatment of cystic fibrosis, our findings suggest that low concentrations may be preferable to high concentration of these agents as they may increase HCO_3^- secretion through activation of IK channels while stimulating less mucin production.

Seung-Ryoung Jung[1]
Toan D Nguyen[1,3]
Bertil Hille[2]
Duk-Su Koh[2,4]

Departments of [1]Medicine and [2]Physiology and Biophysics, University of Washington, Seattle, WA, USA,
[3]VA Puget Sound Health Care System, Seattle, USA
[4]Department of Physics, POSTECH, Korea

References

Berridge MJ, Bootman MD & Roderick HL (2003). Calcium signalling: dynamics, homeostasis and remodelling. *Nat Rev Mol Cell Biol* **4**, 517–529.

Dolmetsch RE, Xu K & Lewis RS (1998). Calcium oscillations increase the efficiency and specificity of gene expression. *Nature* **392**, 933–936.

Jung SR, Kim MH, Hille B, Nguyen TD & Koh DS (2004). Regulation of exocytosis by purinergic receptors in pancreatic duct epithelial cells. *Am J Physiol* **286**, C573–579.

Jung SR, Kim KJ, Hille B, Nguyen TD & Koh DS (2006). Pattern of Ca^{2+} increase determines the type of secretory mechanism activated in dog pancreatic duct epithelial cells. *J Physiol* **576**, 163–178.

Nguyen TD, Moody MW, Savard CE & Lee SP (1998). Secretory effects of ATP on nontransformed dog pancreatic duct epithelial cells. *Am J Physiol* **275**, G104–113.

Hajnóczky G, Robb-Gaspers LD, Seitz MB & Thomas AP (1995). Decoding of cytosolic calcium oscillations in the mitochondria. *Cell* **82**, 415–424.

Oancea E & Meyer T (1998). Protein kinase C as a molecular machine for decoding calcium and diacylglycerol signals. *Cell* **95**, 307–318.

Acknowledgement
Apologies to Martin Rosenberg for failing to acknowledge his award-winning photograph of Sir Eric Denton (*Physiology News* **67**, 55).

β-adrenergic receptor desensitisation – a mechanism for post-exercise reductions in cardiac function

It is widely recognised that skeletal muscle performance is reduced in response to prolonged exercise; whether the same occurs with cardiac muscle is currently a topic of scientific debate. A number of recent studies have demonstrated a modest and transitory impairment of left ventricular (LV) systolic (ejection fraction and the ratio of end systolic volume to pressure) and diastolic (ratios of early to late LV filling and early and late myocardial wall velocities) function after prolonged exercise such as marathon running and Ironman triathlons.

Keith George (left), Emma Hart and Rob Shave.

Driven largely by advances in echocardiographic technology, numerous studies have described post-exercise cardiac impairment in increasing detail. The initial studies using 2-dimensional and Doppler measures were improved upon with tissue Doppler and recently 2-dimensional speckle tracking. With the advance in technology it has been possible to demonstrate both global and regional changes in systolic function; for example, Rifai *et al.* (1999) demonstrated a 21% reduction in ejection fraction in addition to segmental wall motion abnormalities. More recently our group have demonstrated regional reductions in peak systolic 2D derived strain and strain rate (longitudinal, circumferential and radial planes) in athletes completing the Comrades ultra-marathon (Fig. 1). Similarly, evidence for reductions in both global and regional diastolic function has been presented (Neilan *et al*. 2006). The trans-mitral filling ratio and LV flow propagation velocity has been shown to be transiently impaired post-exercise (Fig. 2). These global diastolic changes have been mirrored by segmental reductions in diastolic wall velocities, strain and strain rate (George *et al*. 2006; Neilan *et al*. 2006).

It is important to recognise the limitations of some of the echo-cardiographic techniques that have been employed; trans-mitral Doppler, flow propagation velocity and ejection fraction are highly dependent upon loading and heart rate. Accordingly, assessment of these variables pre- to post- exercise warrants caution; however, the regional differences in LV function

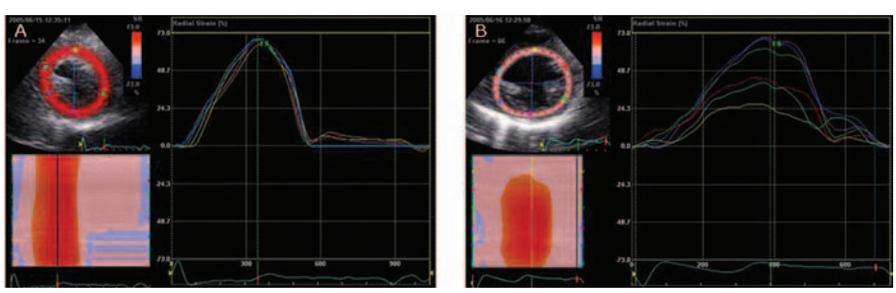


Figure 1. Individual example of peak radial strain pre- (A) and post- (B) completion of the Comrades ultra-marathon. Note the reduction in peak strain in the septal and anteroseptal wall segments but the almost identical pre and post-race peak strain values in free wall segments.

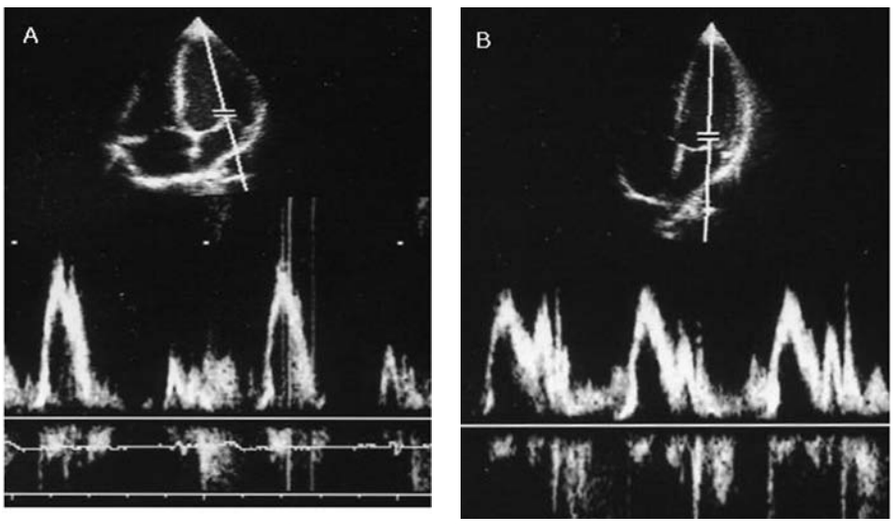


Figure 2. Individual example of trans-mitral Doppler profiles pre- (*A*) and post- (*B*) completion of the London marathon, note the reduction in early and increase in late filling velocity.

that have been shown cannot be explained by altered loading or HR. Further, when correlations are examined the changes in LV function following prolonged exercise cannot be fully explained by altered loading or HR. Whilst characterisation of post-exercise alterations in LV function is useful it does not provide a mechanistic explanation.

A number of possible mechanisms for post-exercise changes in LV function have been suggested – including minor injury to the myocardium evidenced by the release of highly cardiac specific markers of injury e.g. cardiac troponin T, impaired sequestration and re-uptake of calcium into the sarcoplasmic reticulum and de-sensitisation of the β-adrenergic receptors (β- AR). To date, however, empirical evidence supporting a role for any of these mechanisms is limited.

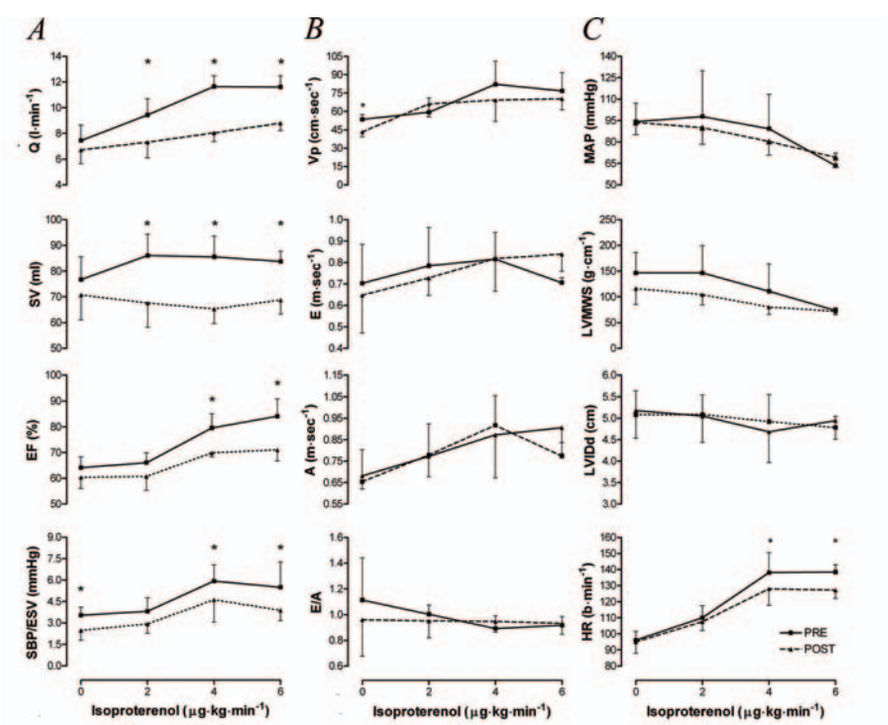


Figure 3. Left ventricular response to incremental isoproterenol infusion pre- and post-exercise. Inotropic (*A*), lusitropic (*B*) and chronotropic/ loading (*C*) response to isoproterenol pre- (continuous line) and post-exercise (dashed line). Data are means ± SD. Q, cardiac output; SV, stroke volume; EF, ejection fraction; SBP/ESV, systolic blood pressure to end systolic volume ratio; Vp, flow propagation velocity of early transmitral filling; A, peak velocity of late transmitral filling; E/A, ratio of early to late LV filling; MAP, mean arterial pressure; LVMWS, LV meridonial wall stress; LVIDd, LV internal diameter in diastole; HR, heart rate. Different from pre-exercise $^{*}p< 0.05$, $^{**}p <0.01$ (from Hart *et al*. 2006).

In our recent study we employed pharmacological manipulation to examine the role of β-AR in post-exercise changes in cardiac function (Hart *et al*. 2006). Prior to and following a 4 h bout of rowing exercise we examined the systolic and diastolic response to increasing doses of isoproterenol (a β-AR agonist). In order to partition out pre- to post-exercise changes in vagal tone, glycopyrrolate (a parasympathetic blocking agent) was administered prior to β-AR stimulation. Our data demonstrate that there is a reduction in the chronotropic and inotropic response to β-AR stimulation following 4 h of rowing (Fig. 3). We suggest, therefore, that post-exercise reductions in systolic function are likely related to a de-sensitisation of the β-AR.

Within heart failure patients, a clear link has been demonstrated between elevations in circulating catecholamines and de-sensitisation and down-regulation of the β-AR (Dzimi *et al*. 1999). It is possible that a similar mechanism may occur following prolonged exercise. Of interest, those participants within our study that demonstrated the greatest elevation in adrenaline following exercise also demonstrated the largest reduction in LV contractility as assessed by ejection fraction (r^2=0.96, P<0.05).
The cardiac β-AR subtypes β_1 and β_2 activate different signalling pathways within the myocyte (Xiao *et al*. 2004). It is possible that during prolonged exercise the sustained elevations in catecholamines initiates β_1-AR mediated apoptosis of the myocyte (Rohrer *et al*. 1999) which may increase membrane permeability and hence explain the

release of cardiac biomarkers such as cardiac troponin T observed following prolonged exercise. Conversely, stimulation of the β_2-AR has an anti-apoptotic effect and protects the myocyte against injury (Patterson *et al*. 2004). Accordingly, de-sensitisation of β_1-AR and stimulation of β_2-AR during prolonged exercise may occur so as to limit injury to the myocardium. In order to examine the role of the specific β-AR subtypes further work is required.

Over stimulation of the β-AR has been demonstrated to induce hypertrophy, at least in the rat heart (Schafer *et al*. 2000). Therefore, the sustained exposure of the β-AR to catecholamines during prolonged exercise may also be an initial stimulus for LV hypertrophy often reported in endurance trained athletes. This, however, remains unsubstantiated and warrants further research.

Although desensitisation of β-AR may be responsible for the reductions in LV systolic function post-exercise it is interesting to note that the diastolic response to increasing doses of isoproterenol did not alter following four-hours of rowing (Fig. 3). Accordingly, changes in diastolic function observed following prolonged exercise are likely related to a different mechanism. Previous studies have indicated that in some chronic heart failure models, β-AR de-sensitisation is related to systolic but not diastolic dysfunction due to an up-regulation of the sodium-calcium exchanger (Sato *et al*. 2004). Consequently, calcium is removed from the myocyte maintaining diastolic function, in spite of an impaired systolic function. Whether this is relevant in the prolonged exercise model is not clear.

The present study provides a mechanistic insight into post-exercise alterations in LV systolic function; however, the mechanisms for alterations in diastolic function following prolonged exercise are not fully understood. Further, whether post-exercise changes in LV function translate to a cardiac limitation during prolonged exercise and whether β-AR de-sensitisation is related to LV hypertrophy remains to be elucidated.

[1]Rob Shave
[2]Keith George
[1]Emma Hart

[1]Centre for Sports Medicine and Human Performance, Brunel University, Uxbridge, Middlesex, UB8 3PH

[2]Research Institute for Sport and Exercise Sciences, Liverpool John Moores University, Liverpool, L3 2ET

References

Dzimiri N. (1999). Regulation of beta-adrenoceptor signaling in cardiac function and disease. *Pharmacol Rev* **51**, 465-501.

George K, Oxborough D, Forster J, Whyte G, Shave R, Dawson E, Stephenson C, Dugdill L, Edwards B, & Gaze D. (2005). Mitral annular myocardial velocity assessment of segmental left ventricular diastolic function after prolonged exercise in humans. *J Physiol* **569**, 305-313.

Hart E, Dawson E, Rasmussen P, George K, Secher N, Whyte G, Shave R. (2006) *b*-adrenergic receptor desensitization in man: insight into post-exercise attenuation of cardiac function. *J Physiol* **577**, 717-725.
Neilan TG, Yoerger DM, Douglas PS, Marshall JE, Halpern EF, Lawlor D, Picard MH, & Wood MJ. (2006). Persistent and reversible cardiac dysfunction among amateur marathon runners. *Eur Heart J* **27**, 1079-1084.

Patterson AJ, Zhu W, Chow A, Agrawal R, Kosek J, Xiao RP, *et al*. (2004). Protecting the myocardium: A role for the beta2 adrenergic receptor in the heart. *Crit Care Med* **32**, 1041-1048.

Rifai N, Douglas PS, O'Toole M, Rimm E, & Ginsburg GS. (1999). Cardiac troponin T and I, echocardiographic [correction of electrocardiographic] wall motion analyses, and ejection fractions in athletes participating in the hawaii ironman triathlon. *Am J Cardiol* **83**, 1085-1089.

Rohrer DK, Chruscinski A, Schauble EH, Bernstein D, & Kobilka BK. (1999). Cardiovascular and metabolic alterations in mice lacking both beta1- and beta2-adrenergic receptors. *J Biol Chem* **274**, 16701-16708.

Sato M, Gong H, Terracciano CM, Ranu H, & Harding SE. (2004). Loss of beta-adrenoceptor response in myocytes overexpressing the Na+/Ca(2+)-exchanger. *J Mol Cell Cardiol* **36**, 43-48.

Schafer M, Frischkopf K, Taimor G, Piper HM, & Schluter KD. (2000). Hypertrophic effect of selective beta(1)-adrenoceptor stimulation on ventricular cardiomyocytes from adult rat. *Am J Physiol Cell Physiol* **279**, C495-503.

Xiao RP, Zhu W, Zheng M, Chakir K, Bond R, Lakatta EG, & Cheng H. (2004). Subtype-specific beta-adrenoceptor signaling pathways in the heart and their potential clinical implications. *Trends Pharmacol Sci* **25**, 358-365.

Tennis and transmission

J C Eccles has been the subject of myriad anecdotes. Fragments of the following one have been lurking in my memory since I heard it at the IUPS Meeting in Sydney in 1983. Liam Burke has confirmed it and added some background detail.

Eccles was a very keen tennis player and had a tennis court at his house in Mosman when he was Director of the Kanematsu Research Institute in Sydney. Bernard Katz and Stephen Kuffler both lived in Mosman and it was traditional for them to meet for tennis at Eccles' house on Sundays. Apparently a major reason why Eccles had hired Kuffler (who had no research experience) was because he was a good tennis player. (He had been the Austrian Junior Tennis Champion before fleeing the Nazis.) Eccles always liked to keep people involved and busy so, on one occasion, he gave Katz the job of mowing the lawn. Katz, who may never have used an electric mower before, ran over the flex and almost electrocuted himself. Eccles immediately acquired a petrol-driven mower. According to Katz, and very typical of his sense of humour, this was the moment when Eccles was converted from electrical transmission to chemical transmission (at least in the peripheral nervous system).

Perhaps other ageing physiologists have more Eccles stories to tell.

Ann Silver
Honorary Member, Cambridge

ATP synthesis during ischaemic muscle contractions

he integrity and function of skeletal muscle cells depend upon an adequate supply of adenosine triphosphate (ATP). In contracting skeletal muscle, ATP is consumed at three major sites:

- Na^+-K^+ ATPase, which restores the electrochemical gradient across the muscle membrane following depolarization;
- actin-myosin ATPase, which consumes ATP during crossbridge cycling in the process of force generation; and
- sarcoplasmic reticulum (SR) ATPase, which is responsible for calcium resequestration into the SR during muscle relaxation.

Skeletal muscle has a limited storage capacity for ATP, which is somewhat surprising given how crucial ATP is to cellular function. For example, during intense muscle contractions, where the consumption of ATP increases ~100-fold compared to rest, the ATP reserve would be exhausted in a matter of seconds. Remarkably, intracellular ATP concentration remains stable even after several minutes of maximum-intensity muscle contractions. This phenomenon can be explained by the action of three major pathways that provide ATP at a rate that satisfies the energetic demands of muscle across a range of conditions (Fig. 1).

The ability to generate ATP oxidatively becomes compromised under conditions of limited oxygen delivery or decreased functionality of mitochondria. Clinically relevant examples of these conditions include peripheral vascular disease and mitochondrial myopathy, both of which are characterized by impaired oxidative ATP synthesis. Much research has been dedicated to understanding how ATP synthesis is affected by limitations in oxygen supply and delivery to working skeletal muscle. Studies of healthy human skeletal muscle under hypoxic or ischaemic conditions have provided some insight into the energetic response to conditions in which oxidative phosphorylation is impaired.

One could anticipate that if oxidative phosphorylation is suppressed, then intracellular ATP stores might be depleted during contractions if the demand for ATP exceeds the supply from non-oxidative pathways. Some research supports this notion by demonstrating depletion of intracellular ATP stores during ischaemic muscle contractions compared to contractions with intact blood flow. However, work from our laboratory (Lanza *et al.* 2006) and others (Hogan *et al.* 1996) has shown that cytosolic [ATP] remains unchanged during free-flow and ischaemic contractions, suggesting that skeletal muscle has a strategy for balancing ATP supply and demand even when the mitochondria are rendered incapable of generating ATP.

Does skeletal muscle compensate by increasing ATP production through other synthesis pathways, for example anaerobic glycolysis? This response is biochemically intuitive, since several substrates involved in the reduction of pyruvate to lactate (e.g. pyruvate, NADH) will accumulate in the muscle if mitochondrial flux is inhibited. Although the reduction of pyruvate to lactate is not itself an ATP-generating step in anaerobic glycolysis, increased lactate production may indirectly help drive upstream reactions by maintaining a high cytosolic NAD^+/ NADH ratio. Phosphorous magnetic resonance spectroscopy (^{31}P-MRS) is an ideal tool for addressing questions related to muscle ATP production in vivo, as it is a non-invasive technique that allows quantification of ATP synthesis with a temporal resolution unmatched by more traditional techniques, such as muscle biopsy analyses (Kemp *et al.* 1994). This approach has been used to examine ATP synthesis in maximally-contracting skeletal muscle under free-flow conditions and with blood flow occluded (Conley *et al.* 1998; Lanza *et al.* 2006). In both of these studies, glycolytic flux did not increase during ischaemia, dispelling the notion, at least during brief maximal contractions with acute ischaemia, that glycolytic ATP synthesis compensates for acute limitations in mitochondrial ATP flux. Alternatively, the balance between ATP supply and demand may be met

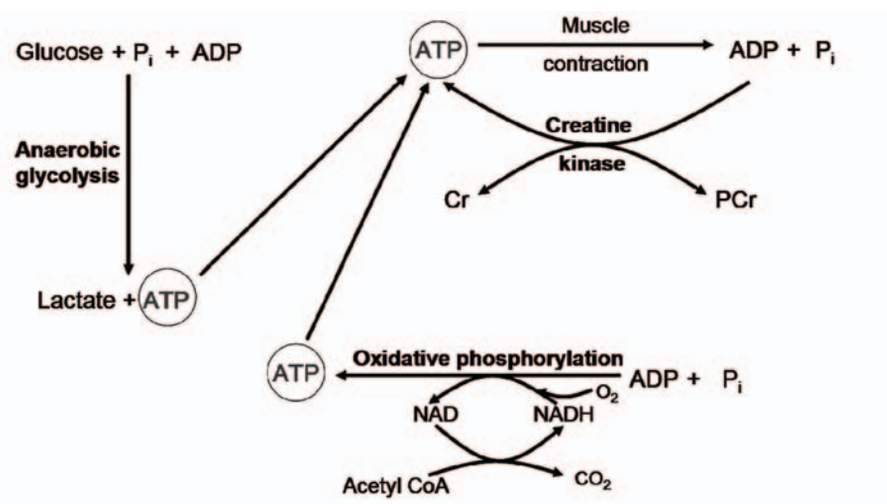


Figure 1. During muscle contractions, ATP demand by the ATPases is generally matched by ATP synthesis through three major metabolic pathways. The creatine kinase reaction phosphorylates adenosine diphosphate (ADP) to ATP at the expense of phosphocreatine (PCr). ATP is also produced via oxidative phosphorylation in the mitochondria and by anaerobic glycolysis in the cytosol.

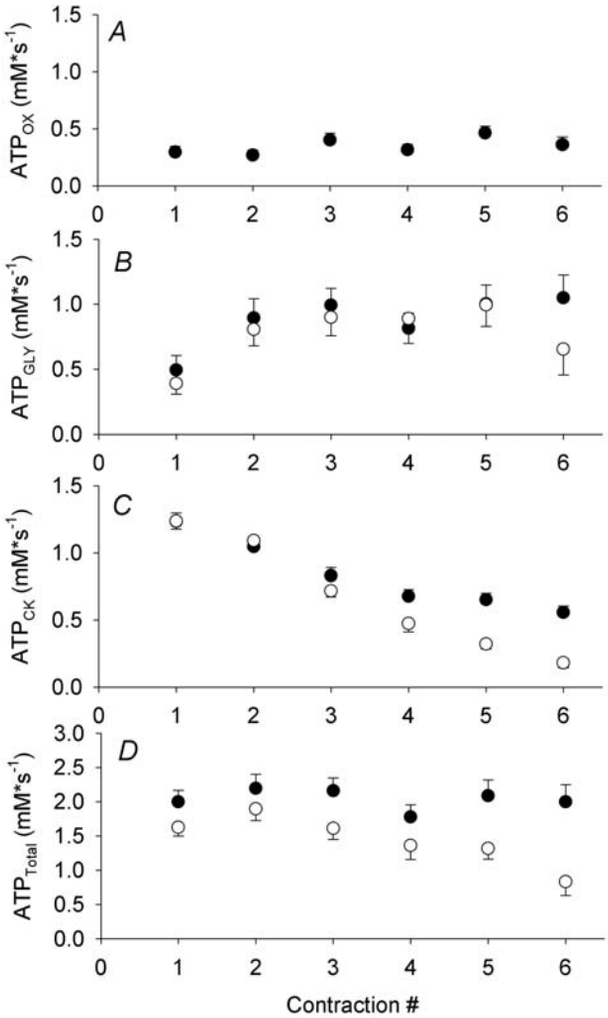


Figure 2. ATP production from oxidative phosphorylation (ATP_{OX}, *A*), anaerobic glycolysis (ATP_{GLY}, *B*), and net PCr hydrolysis (ATP_{CK}, *C*) measured by ^{31}P-MRS in human tibialis anterior muscle during free-flow (•) and ischaemic (o)contractions. Total ATP turnover (ATP_{TOT}, *D*), calculated as the sum of ATP_{OX}, ATP_{GLY}, and ATP_{CK}, was greater during free-flow compared to ischaemia.

by decreasing demand for ATP rather than with a compensatory increase in ATP supply through an alternative pathway (Lanza *et al.* 2006). At least two mechanisms by which ATP demand may decrease during contractions in oxygen-limited conditions have been proposed (Hogan *et al.* 1996; Lanza *et al.* 2006).

First, the demand for ATP may be tempered by increased contractile economy (force produced per unit of ATP consumed) when oxygen levels are not adequate to sustain oxidative metabolism. Using ^{31}P-MRS measures of ATP synthesis rates (Figs. 2*A*, *B*, *C*), along with simultaneous measures of muscle force production, we found that contractile economy was higher in vivo during ischaemic compared with free-flow contractions in human skeletal muscle (Fig. 2*D*; Lanza *et al.* 2006). Using more invasive measures of blood flow, peripheral oxygen uptake, and ATP production,

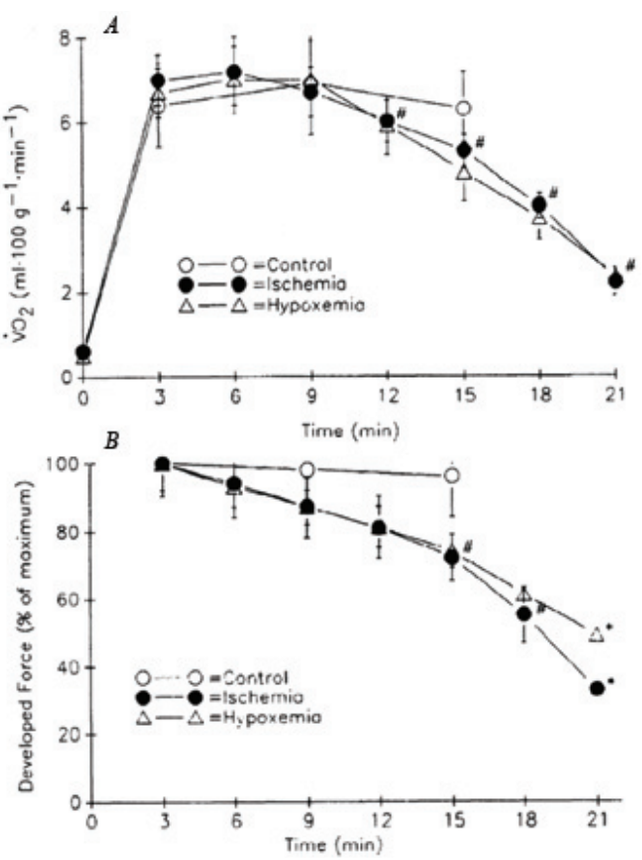


Figure 3. Mean muscle oxygen uptake (*A*) and force production (*B*) during control contractions (open circles, o) and during contractions with step reductions in oxygen delivery elicited either by decreasing the fraction of inspired oxygen (hypoxia, open triangles, Δ) or lowering pump perfusion of the muscle (ischaemia, filled circles, •). (*A*) Following an initial rise in VO_2, oxygen uptake remained constant during control contractions, whereas contractions during ischaemia and hypoxia elicited similar declines in VO_2 as contractions progressed, in comparison to control. (*B*) Developed force remained constant during control contractions, but steadily decreased during ischaemic and hypoxic contractions, with a greater decline during ischaemia than hypoxia by minutes 18 and 21. (Reproduced from Hogan *et al.* (1996), with permission.)

Krustrup *et al.* (2003) found higher mechanical efficiency and lower heat production during ischaemia than when blood flow was intact. Thus, there exists in vivo and in situ evidence to support the greater economy of ischaemic compared to oxygenated contractions. Numerous factors could contribute to increased contractile economy in the absence of adequate oxygenation; for example, changes in intracellular pH, calcium, temperature, and free phosphate have all been suggested to affect contractile economy, possibly through direct effects on the rate of cross-bridge cycling.

In addition to the metabolic savings conferred by increased contractile economy during ischaemia, the demand for ATP may also decrease consequent on the greater muscle fatigue during ischaemic compared to free-flow contractions. As contractility declines during fatigue, so too does the amount of ATP required by the ATPases, particularly myosin ATPase. It has been suggested that this may serve as a protective mechanism designed to prevent declines in cellular (ATP) and disruption of cellular homeostasis. Under ischaemic conditions, greater fatigue may develop due to the accumulation of intracellular metabolites (H^+, Pi, $H_2PO_4^-$) that are known to interfere with sarcolemmal excitability, calcium handling by the SR, and myofibrillar force generation. However, there are some intriguing data to suggest that, under certain conditions, force production is tightly coupled to oxidative ATP supply. Hogan and colleagues demonstrated this concept nicely in a perfused dog muscle preparation by imposing gradual, stepwise reductions in oxygen delivery and observing that force production decreased in parallel with declines in muscle VO_2 (Hogan *et al.* 1996; see Fig. 3*A*, *B*). Furthermore, this response occurred in the absence of any differences in muscle lactate production, ATP depletion, or accumulation of inhibitory metabolites. This behaviour in skeletal muscle is similar to that observed repeatedly in cardiac tissue. Preconditioning heart muscle with brief bouts of ischaemia leads to decreased ATP demand in concert with reduced ATP production during subsequent bouts of prolonged ischaemia. This phenomenon, known as 'myocardial hibernation', can reduce subsequent infarct size by preventing severe disruptions in cellular homeostasis. The work of Hogan and others suggests that skeletal muscle is also capable of this type of behaviour.

In summary, it appears that skeletal muscle is adept at ensuring a balance between energy supply and demand under a variety of conditions, including situations where limited oxygen delivery precludes oxidative synthesis of ATP. Although compensatory increases in non-oxidative ATP production seem logical, the results from several studies suggest that this does not occur in vivo. Instead, an energetic balance appears to be accomplished by decreasing ATP demand during contractions, potentially by increasing contractile economy or decreasing force production.

Ian R Lanza
Jane A Kent-Braun
Muscle Physiology Laboratory,
Department of Kinesiology,
University of Massachusetts, USA

References

Conley KE, Kushmerick MJ & Jubrias, SA (1998). Glycolysis is independent of oxygenation state in stimulated human skeletal muscle in vivo. *J Physiol* **511**, 935–945.

Hogan MC, Kurdak SS & Arthur, PG (1996). Effect of gradual reduction in O2 delivery on intracellular homeostasis in contracting skeletal muscle. *J Appl Physiol* **80**, 1313–1321.

Kemp GJ, Thompson CH, Barnes PR, & Radda GK (1994). Comparisons of ATP turnover in human muscle during ischemic and aerobic exercise using 31P magnetic resonance spectroscopy. *Magn Reson Med* **31**, 248–258.

Krustrup P, Ferguson RA, Kjaer M, & Bangsbo J (2003). ATP and heat production in human skeletal muscle during dynamic exercise: higher efficiency of anaerobic than aerobic ATP resynthesis. *J Physiol* **549**, 255–269.

Lanza IR, Wigmore DM, Befroy DE, & Kent-Braun JA (2006). In vivo ATP production during free-flow and ischaemic muscle contractions in humans. *J Physiol* **577**, 353–367.

Sydney Ringer

Image based on a photo by Lisa Inoue of Tokyo (1991), who draws inspiration from Ringer's life and work.

David Miller, Honorary Research Fellow at the Faculty of Biomedical & Life Sciences, University of Glasgow, has produced *A solution for the heart: the life of Sydney Ringer (1836–1910)*.

Ringer is the scientist and clinician most properly credited with 'inventing' physiological saline, most familiar as the 'drip' seen in operating theatres and hospital wards. As one of the scientific fathers of this life-saving liquid, Ringer deserves to be as well known as others whose names are associated with the great advances of medical science.

David's booklet tells about a remarkable scientist and doctor who was associated with the village of Lastingham in North Yorkshire and its church for most of his adult life. Sydney Ringer lies buried in the churchyard of St Mary's, Lastingham, North Yorkshire, together with his wife and their elder daughter, in whose memory the church was extensively restored in 1879. It gives some insight into life in Lastingham in the 19th century and Ringer's career as a leading London medical man of his day.

A plaque in honour of Sydney Ringer, sponsored by The Physiological Society, was unveiled by Dafydd Walters at St Mary's on 27 October. Photos of the occasion will be published in a future issue of *Physiology News*.

Copies of the booklet are available at http://www.physoc.org/about/history or in print via lrimmer@physoc.org.

Colonic elongation activates an intrinsic reflex that underlies slow transit and accommodation

It is generally believed that after-hyperpolarizing neurons are the only intrinsic sensory neurons in the gut wall. Recently, Smith and colleagues have demonstrated that some synaptic interneurons in the large bowel are activated by circumferential stretch and generate an ongoing motor activity even when AH neurons are silent. Further studies suggest that some descending interneurons are also mechanosensory since they are activated by longitudinal stretch rather than circumferential stretch. In particular, these mechanosensory neurons, which are NOS positive, respond to colonic elongation by releasing nitric oxide to reduce activity in intrinsic peristaltic circuits. This later reflex promotes storage by reducing the transit rate of fecal pellets down the large bowel

Transit of intraluminal contents through the human large bowel is extremely slow (≥ 30 hrs) compared to transit through other regions of the gastrointestinal tract. As intraluminal contents move down the large bowel, water and electrolytes are absorbed causing the contents to become more viscous leading to stool formation. This is dramatically illustrated in the guinea-pig large bowel, where the proximal colon contains viscous fluid chyme that is compacted into soft fecal pellets at the flexure between the proximal and distal colon (D'Antona *et al*. 2001).

A common view at the turn of the century was that an elongated colon (called dolichocolon) was a common cause of constipation. Although, this concept has been recently challenged (Muller-Lissner *et al*. 2005) it still persists in the literature: *'… the cause of constipation was colonic slow-transit … which was always associated with dolichocolon '* (Ripetti *et al.* 2006). However, an excess production of nitric oxide (NO) within myenteric neurons has been proposed from immuno-histochemical studies to have a role in the pathology underlying slow transit constipation in the human large bowel (Cortesini *et al*. 1995). Our laboratory has recently discovered a powerful intrinsic neural reflex that likely underlies colonic storage since it is activated by colonic elongation that triggers myenteric interneurons that release NO to depress motility (Dickson *et al*. 2007a, b).

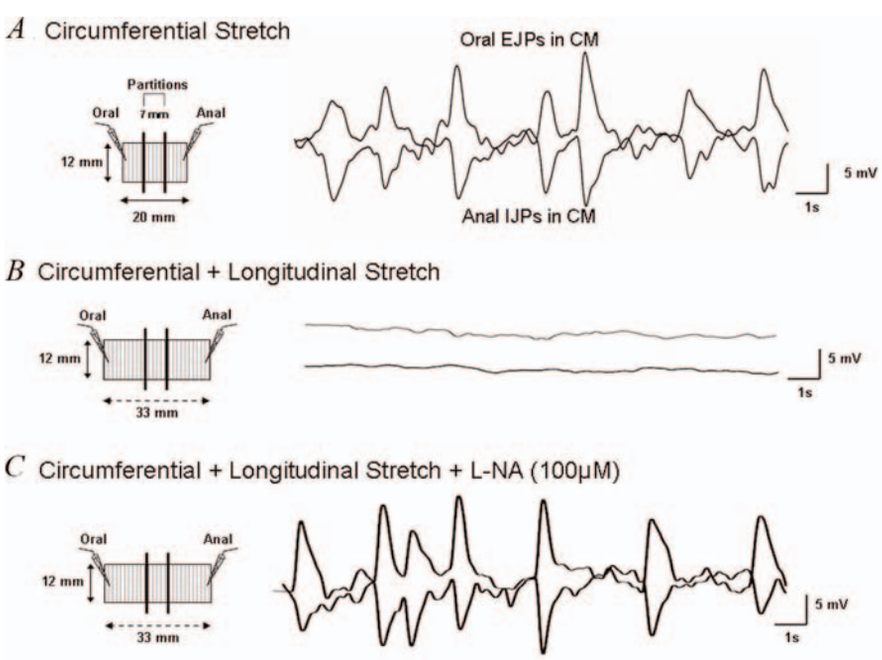


Figure 1. Effect of circumferential and longitudinal stretch. *A*, Intracellular microelectrodes were used to record from the CM at either end of a circumferential stretched preparation (20mm long) of distal colon. Circumferential stretch activated ongoing peristaltic reflex activity consisting of oral excitatory junction potentials (EJPs) that were coordinated with anal inhibitory junction potentials (IJPs) in the circular muscle (CM). *B*, Ongoing peristaltic reflex activity evoked by circumferential stretch was almost abolished by the addition of 60 % longitudinal stretch. *C*, N omega-nitro-L-arginine (100 µM) added to the middle chamber (7 mm wide) between the two recording sites restored all ongoing and evoked activity.

It is well known that there are two electrophysiological classes of myenteric neuron in the enteric nervous system of the large intestine, called originally AH and S type neurons by Hirst *et al*. (1974). After-hyperpolarizing (AH) neurons are rapidly adapting, multipolar neurons with one or two processes projecting down to the mucosa. They respond to chemical stimulation of the mucosa and smooth muscle tension (Furness *et al*. 1998; Smith, 1996; Spencer & Smith, 2004; Smith *et al*. 2005). In contrast, S (for synaptic) neurons are slowly adapting and receive fast excitatory postsynaptic potentials. They are uniaxonal excitatory and inhibitory motor neurons and ascending and descending interneurons (Lomax & Furness, 2000; Smith, 1996; Smith *et al.* 1992; Spencer & Smith, 2005). Some ascending and descending interneurons in the distal colon are stretch sensitive in that they detect circular muscle (CM) length rather than muscle tension (Smith *et al*. 2005; Spencer & Smith, 2004). In

circumferentially stretched preparations of distal colon (~20mm long) these interneurons can generate ongoing peristaltic reflex activity even when AH neurons are silent (Spencer & Smith, 2004). This neural activity consists of excitatory junction potentials (EJPs) in the CM at the oral end that are phase locked in both time and amplitude with inhibitory junction potentials (IJPs) at the anal end of the preparation (Fig. 1*A*; Spencer & Smith, 2004). The only way this can occur is if the ascending and descending mechanosensory interneurons mediating this activity are synapsing with one another to form a reverberating circuit (Fig. 2; Smith *et al*. 2005; Spencer *et al*. 2006).

Immunohistochemical coding studies of myenteric neurons in the guinea-pig distal colon have revealed 3 chemically distinct classes of ascending interneurons and 4 classes of descending interneurons (Lomax & Furness, 2000). We were therefore curious as to whether more of these chemically distinct interneurons in the colon might be mechanosensory.

Over 40 years ago Hukuhara *et al.* (1960) described a reflex activated by longitudinal stretch. They found that stretching the canine small intestine in the longitudinal axis with a hemostat produced an active and transient relaxation of the CM that inhibited ongoing rhythmic contractions, which they referred to as the 'muscular intrinsic inhibitory reflex'. They also found that removing the longitudinal muscle without apparently disrupting the myenteric plexus abolished this reflex, suggesting that the sensory elements mediating this reflex were in the longitudinal muscle (LM) layer.

Given this result, we were interested in determining the effects of longitudinal stretch on the ongoing peristaltic reflex activity generated by radial stretch of the guinea-pig distal colon (Spencer *et al.* 2002, 2006). Much to our surprise we found that the addition of longitudinal stretch did not produce a relaxation or an inhibitory junction potential in the muscle, but instead suppressed the ongoing oral EJPs and anal IJPs in the CM in a graded manner (Fig.1*B*; Dickson *et al.* 2007a, b). We referred to this reflex, which was quite different from that described by Hukuhara *et al.* (1960), as an intrinsic occult (hidden or obscure) reflex since it suppresses, rather than evokes, activity in the muscle. Furthermore, this suppression of activity by longitudinal stretch occurred even when the longitudinal muscle had been removed suggesting that the sensory processes of the neurons mediating this reflex were in the myenteric plexus or CM (Dickson *et al.* 2007a; Spencer *et al*. 2006). We were surprised to find that blocking NO synthesis between the oral and anal recording sites completely restored the amplitude and coordination of oral EJPs and anal IJPs in the CM, despite the maintained longitudinal stretch (Fig. 1*C*). This suggested that there were specific mechanosensory neurons that responded to longitudinal stretch by releasing NO to inhibit myenteric neurons in peristaltic reflex pathways (Fig. 2). From the chemical coding studies described above (Lomax & Furness, 2000), we reasoned that the neurons responding to longitudinal stretch were most likely descending NOS +ve interneurons, since the other interneurons contained mainly ACh and potential peptidergic neurotransmitters that are mainly excitatory to other myenteric neurons.

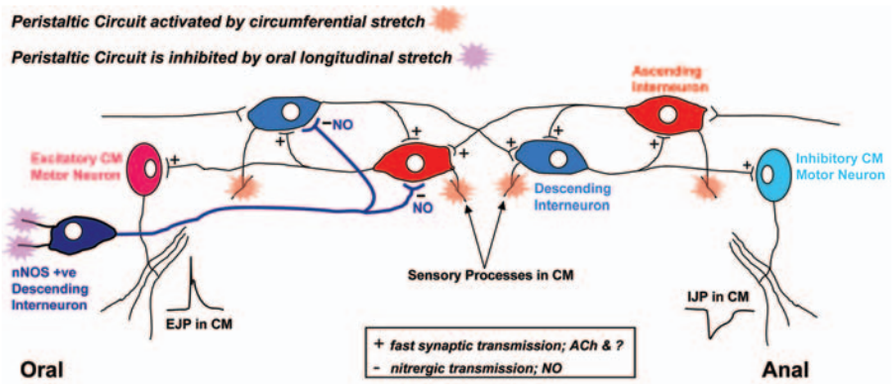


Figure 2. Myenteric circuitry activated by radial stretch and by colonic elongation. In preparations where muscle tension has been eliminated by the presence of nifedipine (1 μM; an L-type Ca^{2+} channel antagonist) circumferential stretch activates ascending and descending mechanosensory interneurons. These interneurons appear to synapse with one another, thus forming reverberating circuits that coordinate ongoing activity between ascending and descending nervous pathways. The ascending interneurons activate excitatory motor neurons to the CM. The descending interneurons activate inhibitory motor neurons to both the CM. This leads to a coordinated oral EJP and an anal IJP in CM at the same time. Orally applied longitudinal stretch (colonic elongation) activates NOS +ve descending interneurons that release nitric oxide to inhibit action potential firing in mechanosensitive ascending and descending interneurons activated by circumferential stretch. This leads to a depression of activity in intrinsic peristaltic nerve circuits.

Calcium imaging followed by immunohistochemistry of myenteric neurons in preparations that had been stretched in both axes to inhibit neurons in peristaltic nerve pathways revealed ongoing activity in a class of neurons that were anally projecting NOS +ve neurons (Dickson *et al.* 2007a). Since we had also blocked synaptic transmission in these preparations with a variety of drugs we reasoned that these must be mechanosensitive interneurons responding to colonic elongation, rather than inhibitory motor neurons. Intracellular microelectrode studies revealed that NO mediated inhibitory postsynaptic potentials (IPSPs) could be evoked in mechanosensory interneurons. The IPSPs decreased action potential firing in these interneurons and thus prevented them from activating excitatory and inhibitory motor neurons to the muscle. Later studies confirmed that this occult reflex was polarized since oral, but not anal,

longitudinal stretch inhibited junction potential activity and pellet propulsion (Dickson *et al.* 2007a, b).

When the guinea-pig distal colon is removed from the animal it is normally full of fecal pellets. Under these circumstances fecal pellet evacuation is slow and sporadic taking ~30 mins to empty all pellets compared to approximately a minute for propulsion of a single pellet down an empty segment of distal colon (D'Antona *et al*. 2001). Interestingly, we found that when the isolated colon is full of pellets it is elongated by about 40% compared to its length following the expulsion of pellets (Smith *et al*. our unpublished observations). Therefore, it would seem highly plausible to assume that the elongation of the colon produced by the pellets activates the occult reflex that slows pellet propulsion and promotes storage.

Our studies have revealed that, in addition to AH sensory neurons, distinct interneurons in the large bowel are mechanosensory and respond to either circumferential stretch or longitudinal stretch. We have described an intrinsic occult reflex that is activated by colonic elongation. Colonic elongation appears to inhibit colonic motility by activating NOS +ve descending interneurons that release NO to suppress activity in mechano-sensitive interneurons involved in peristalsis. Clinically, the intrinsic occult reflex provides a negative feedback system that would be expected to exacerbate the inhibition of motility associated with constipation in an impacted colon. It remains to be seen whether any more myenteric interneurons in the colon are also mechanosensory.

Acknowledgements

This study was funded by grants from the National Institute of Diabetes and Digestive and Kidney Diseases: RO1 DK45713 (TKS).

Terence K Smith, Eamonn J Dickson, Grant W Hennig Peter O Bayguinov, Nick J Spencer

Department of Physiology & Cell Biology, University of Nevada School of Medicine, Reno, Nevada, USA

References

Cortesini C, Cianchi F, Infantino A & Lise M (1995). Nitric oxide synthase and VIP distribution in enteric nervous system in idiopathic chronic constipation. *Dig Dis Sci* **40**, 2450-2455.

D'Antona G, Hennig GW, Costa M, Humphreys CM & Brookes SJ (2001). Analysis of motor patterns in the isolated guinea-pig large intestine by spatio-temporal maps. *Neurogastro Motil* **13**, 483-492.

Dickson E, Spencer NJ, Hennig GW, Bayguinov O, Ren J, Heredia D & Smith TK (2007a). An intrinsic occult reflex underlies accommodation and slow transit in the distal large bowel. *Gastroenterology* **132**, 1912-24.

Dickson E, Ren, Hennig GW, Spencer NJ & Smith TK (2007b). A polarized intrinsic occult reflex underlies accommodation and slow transit in the large bowel. *Gastroenterology* **A-247**, S1688.

Furness JB, Kunze WA, Bertrand PP, Clerc N & Bornstein JC (1998). Intrinsic primary afferent neurons of the intestine. *Prog Neurobiol* **54**, 1-18.

Hirst GDS, Holman ME & Spence I (1974). Two types of neurons in the myenteric plexus of duodenum in the guinea-pig. *J Physiol* **236**, 303–326.

Hukuhara T, Nakayama S & Nanba R (1960). Locality of receptors concerned with the intestino-intestinal muscular intrinsic reflexes *Jap J Physiol* **10**, 414-418.

Lomax AE & Furness JB (2000). Neurochemical classification of enteric neurons in the guinea-pig distal colon. *Cell Tissue Res* **302**, 59-72.

Muller-Lissner SA, Kamm MA, Scarpignato C & Wald A (2005). Myths and misconceptions about chronic constipation. *Am J Gastroenterol*. **100**, 232-242.

Ripetti V, Caputo D, Greco S, Alloni R & Coppola R (2006). Is total colectomy the right choice in intractable slow-transit constipation? *Surgery* **140** , 435-440.

Smith TK (1996). An electrophysiological identification of intrinsic sensory neurons that are excited by 5-HT applied to the mucosa in the guinea-pig proximal colon. *J Physiol* **495**, 102P.

Smith TK, Bornstein JC & Furness JB (1992). Convergence of reflex pathways excited by distension and mechanical stimulation of the mucosa onto the same myenteric neurons of the guinea-pig small intestine. *J Neurosci* **12**, 1502–1510.

Smith TK, Hennig GW & Spencer NJ (2005). Mechanosensory transduction in the enteric nervous system. *Physiology News* **58**, 23–25.

Spencer NJ, Hennig GW & Smith TK (2002). A rhythmic motor pattern activated by circumferential stretch in guinea-pig distal colon. *J Physiol* **545,** 629–648.

Spencer NJ & Smith TK (2004). Mechano-sensory S-neurons rather than AH neurons appear to generate a rhythmic motor pattern in guinea-pig distal colon. *J Physiol* **558** , 577–596.

Spencer NJ, Dickson EJ, Hennig GW & Smith TK (2006) Sensory elements within the circular muscle are essential for mechanotransduction of ongoing peristaltic reflex activity in guinea-pig distal colon. *J Physiol* **576**, 519-531.

Physiology in Galicia

The International Workshop on *Molecular physiology of membrane transport and cell excitability* was a wonderful opportunity not only to listen to presentations by some of the leading researchers in the field (including two Nobel Laureates), but also to catch a glimpse of Ukrainian culture and history. The workshop was held in the beautiful village of Yaremche, nestled in the Carpathian Mountains outside Ivano Frankovsk.

The lectures and student presentations were stimulating and thought-provoking. The meeting as a whole was a great combination of scientific discussion/presentations and social interaction. I would especially like to thank the professors and

participants from the Precarpathian National University for their enormous help with directions, translation, tour guiding and the wonderful cultural performance, the organizers for overall coordination and The Physiological Society for funding these valuable learning opportunities.

Brian Hagen

University of Maryland Biotechnology Institute, Baltimore, MD, USA

An integrative approach to control of oxidative phosphorylation during exercise

At the onset of exercise there is an instantaneous increase in energy (i.e. ATP) utilization within the active muscle. This increased ATP demand is accompanied by increases in both substrate-level (i.e. anaerobic) and oxidative (i.e. aerobic) phosphorylations, such that muscle ATP content remains relatively constant regardless of the exercise intensity or duration. Muscle O_2 consumption, a reflection of mitochondrial oxidative ATP production, can be estimated by pulmonary O_2 uptake. During the transition to exercise, pulmonary O_2 uptake, and muscle O_2 consumption, increase exponentially towards a new level (see Fig. 1, open circles).

As this increase is not 'instantaneous', the deficit in meeting ATP requirements by oxidative metabolism must be met by non-oxidative (substrate-level) ATP synthesis. Thus, a consequence of this delay is a metabolic disturbance within the myocyte which includes a breakdown of creatine phosphate (PCr) and glycogen, and an accumulation of creatine (Cr), ADP, lactate$^-$, protons (H^+) and inorganic phosphate (Pi). Together these changes can contribute to increased perception of effort and early fatigue. Therefore, faster activation of muscle O_2 consumption and oxidative phosphorylation is expected to reduce cellular disturbances, thereby increasing exercise tolerance and performance.

Control or limitations to the rate of increase of muscle O_2 consumption have been linked to; (i) activation of enzymes and provision of substrate (i.e. acetyl CoA and reducing equivalents) to the mitochondrial tricarboxylic acid (TCA) cycle and electron transport chain (ETC); and (ii) adjustments of muscle blood flow and O_2 delivery during the transition to exercise. In two recent publications (Gurd *et al*. 2005, 2006) we demonstrated that the kinetics of pulmonary VO_2 (and muscle O_2 consumption) during the transition to moderate-intensity exercise (i.e. an intensity domain associated with no, or only a transient, accumulation of lactate$^-$) was faster when preceded by a bout of heavy-intensity, warm-up exercise (see Fig. 1, closed circles). Therefore, using this model of prior heavy-intensity exercise, we are attempting to understand the limitations to the activation of muscle O_2 consumption and oxidative phosphorylation during the transition to exercise.

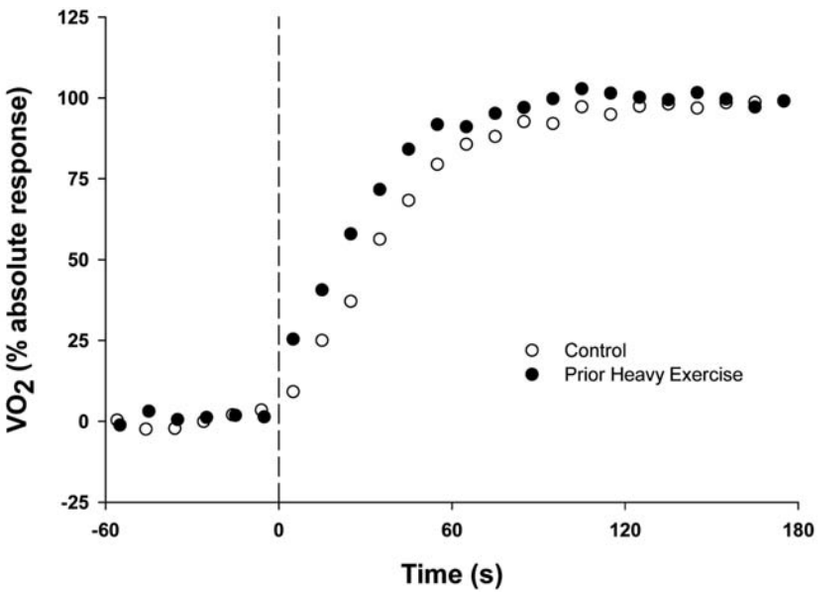


Figure 1. Typical adaptation of oxidative phosphorylation as measured at the lung during the transition to exercise (open circles) and the same response measured following heavy-intensity 'warm-up' (closed circles). Mean response of 15 subjects take from (Gurd *et al.* 2005; Gurd *et al.* 2006).

Production of ATP by oxidative phosphorylation requires the presence of O_2, reducing equivalents (NADH and $FADH_2$ (NADH for simplicity)), ADP and Pi (Fig. 2). While ADP availability, a consequence of an increase in ATP hydrolysis, likely plays a major role in activating mitochondrial oxidative phosphorylation, the availability of other substrates (O_2 and NADH) may limit the rate of activation of oxidative phosphorylation at a given ADP concentration ([ADP]) or ratio [ATP]/[ADP][Pi]. A role for the interaction between the concentrations of O_2, NADH and ADP in regulating mitochondrial oxidative phosphorylation rather than by a single substrate was reviewed by Wilson (1994). As cellular PO_2 decreases below approximately 30 torr, a given rate of mitochondrial respiration can only be sustained by an increase in [NADH] (or [NADH]/[NAD^+]) or an increase in [ADP] (or decrease in [ATP]/[ADP]), or both. We suggest that it is this 'interaction' between substrate concentrations that both limits kinetics at the onset of exercise, and is responsible for the speeding of pulmonary VO_2 kinetics (and muscle O_2 consumption) that is observed following a prior bout of heavy-intensity, warm-up exercise.

During the transition to exercise, any substrate that is not present in saturating concentrations can limit oxidative phosphorylation. Experimental data obtained from exercising humans demonstrates that intracellular PO_2 is approximately 30-40 torr at rest and decreases to less than 10 torr at relatively light-intensities of exercise (Richardson *et al*. 1995) – i.e. below 30 torr where oxidative phosphorylation begins to display a dependency on intracellular PO_2. Technical limitations have prevented an

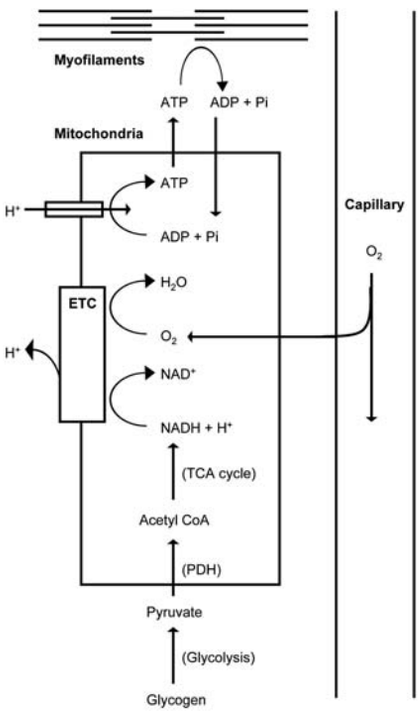


Figure 2. Schematic of oxidative phosphorylation and the delivery of oxidative substrate to the electron transport chain (ETC) and mitochondrial ATPase from the myofilament ATPase (ADP), muscle perfusion (O_2) and carbohydrate metabolism (NADH via glycolysis, pyruvate dehydrogenase (PDH) and the tricarboxylic acid (TCA) cycle).

accurate estimation of intramitochondrial [NADH] at rest or during exercise. However, there is evidence that an increase in muscle acetyl group availability via elevated flux through PDH, and subsequently, muscle NADH content can result in a faster activation of oxidative phosphorylation (Howlett *et al*. 1999). Unlike [O_2] and [NADH] which, early in the exercise transition, depend on the adaptation of blood flow and carbohydrate metabolism, respectively, [ADP] begins to increase as soon as muscle contraction is initiated and continues until a new steady-state of ATP synthesis is established. Thus, [ADP] exerts control over the rate of oxidative phosphorylation independent of the concentration of O_2 or NADH and can therefore be said to be the driving substrate for oxidative phosphorylation.

Attempts to increase only a single substrate prior to the onset of exercise experimentally have produced varying findings. Studies attempting to increase [O_2] have utilized pump perfusion and hyperoxia in animal models, and elevated central (cardiac output) or peripheral blood flow and O_2 delivery in humans, most of which failed to speed the rate of pulmonary VO_2 (or muscle O_2 consumption) (see Tschakovsky & Hughson, 1999 for review). Similarly, attempts to increase [NADH] during the transition to exercise by prior activation of the mitochondrial pyruvate dehydrogenase (PDH) complex and increased flux of carbohydrate-derived substrate into the TCA cycle (by means of dichloroacetate administration) also have yielded mixed results (see Grassi, 2005 for review). However, as discussed earlier it appears that neither O_2 nor NADH are present in saturating concentrations and thus should exert control over the rate of oxidative phosphorylation.

We recently demonstrated: i) faster pulmonary VO_2 (i.e. oxidative phosphorylation) kinetics following heavy-intensity, warm-up exercise in association with ii) greater muscle perfusion (determined by near-infrared spectroscopy (NIRS)-derived increase in total and oxygenated haemoglobin-myoglobin concentration) and iii) greater mitochondrial PDH activity which contributes to a greater muscle acetyl CoA and NADH content (determined by muscle biopsy of the quadriceps muscle group) (Gurd *et al.* 2005; Gurd *et al.* 2006). The discrepancy between this finding and that of other studies where only a single substrate was augmented (i.e. O_2 or NADH, see above) is that following heavy-intensity exercise, both O_2 and NADH availability are elevated prior to the onset of exercise and thus are immediately available to the mitochondrial ETC. Therefore, the faster rate of activation of oxidative phosphorylation seen after heavy-intensity exercise is a consequence of removing any limitation imposed by either muscle perfusion and O_2 delivery, or enzyme activation and NADH availability, or both.

An interesting observation from our recent publications was that the relative speeding of pulmonary VO_2 kinetics that was seen in our subjects was not consistent, but was dependent on their initial rate of adaptation (Fig. 3) – i.e. the speeding of kinetics was less in those subjects who presented with faster VO_2 kinetics in the 'no warm-up' condition. Many studies examining control of oxidative phosphorylation have used healthy,

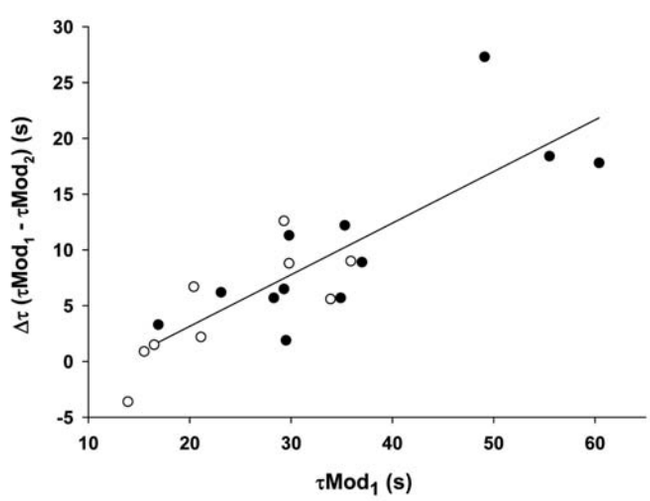


Figure 3. The relationship between τMod_1 and the change in τ ($\Delta\tau$) between Mod_1 (without 'warm-up') and Mod_2 (with 'warm-up') for the activation of oxidative phosphorylation during the transition to exercise (y-intercept = -6.4; slope = 0.47; r^2 = 0.71; $p < 0.05$). Data are taken from (Gurd *et al.*, 2005) (closed circles) and (Gurd *et al.* 2006) (open circles).

physically fit, young adults or animal models using highly capillarized, oxidative muscle preparations. In these instances the adaptation of pulmonary VO_2 kinetics and muscle O_2 consumption is relatively rapid (time constant (τ) ~ 20 s). Thus, any speeding in the adaptation of oxidative phosphorylation resulting from an intervention in healthy, physically fit, young humans or in oxidative animal muscle preparations is likely to be small and difficult to detect. The situation is likely to be exacerbated in situations where only a single substrate is elevated, such as in the previously mentioned studies where either [O_2] or [NADH] was increased alone.

Interestingly, in cases where elevating one or more substrate(s) was successful in speeding the activation of oxidative phosphorylation, full activation was still not instantaneous. This period may represent the time required for ADP to increase to appropriate levels to drive oxidative phosphorylation. Consistent with this idea is the finding that rapid increases in [ADP] alone (either by iodoacetamide-induced inhibition of the creatine kinase (CK) reaction or by using CK-knockout mice) result in a significantly faster activation of oxidative phosphorylation during the transition to exercise (for an example see Kindig *et al.* 2005).

The adaptation of oxidative phosphorylation during the transition to exercise appears to be dependant on the provision of ADP with the availability of O_2 and NADH acting as modulators. Our own data show that if, before the start of exercise, muscle perfusion and O_2 delivery and muscle substrate (pyruvate) and enzyme (PDH) activities are elevated, the rate of activation of mitochondrial oxidative phosphorylation and O_2 consumption (as determined by pulmonary VO_2) is faster compared to a control condition. As a consequence substrate-level phosphorylations are minimized, which should reduce metabolic disturbances linked to fatigue and exercise intolerance.

Brendon J Gurd[1], George J F Heigenhauser [3], Donald H Paterson[1], John M Kowalchuk[1,2]

[1]Canadian Centre for Activity and Aging, School of Kinesiology, Faculty of Health Sciences, Department of Physiology and Pharmacology

[2]Schulich School of Medicine and Dentistry, The University of Western Ontario, London, Ontario, Canada

[3]Department of Medicine, McMaster University, Hamilton, Ontario, Canada L2S 3A1.

References

Grassi B (2005). Delayed metabolic activation of oxidative phosphorylation in skeletal muscle at exercise onset. *Med Sci Sports Exerc* **37**, 1567–1573.

Gurd B, Peters SJ, Heigenhauser GJ, LeBlanc PJ, Doherty TJ, Paterson DH & Kowalchuk JM (2006). Prior heavy exercise elevates pyruvate dehydrogenase activity and speeds O_2 uptake kinetics during subsequent moderate-intensity exercise in healthy young adults. *J Physiol* **577**, 985–996.

Gurd BJ, Scheuermann BW, Paterson DH & Kowalchuk JM (2005). Prior heavy-intensity exercise speeds VO_2 kinetics during moderate-intensity exercise in young adults. *J Appl Physiol* **98**, 1371–1378.

Howlett RA, Heigenhauser GJF, Hultman E, Hollidge-Horvet & Spriet LL (1999). Effects of dichloroacetate infusion on human skeletal muscle metabolism at the onset of exercise. *Am J Physiol* 277, E18–25.

Kindig CA, Howlett RA, Stary CM, Walsh B & Hogan MC (2005). Effects of acute creatine kinase inhibition on metabolism and tension development in isolated single myocytes. *J Appl Physiol* **98**, 541–549.

Richardson RS, Noyszewski EA, Kendrick KF, Leigh JS & Wagner PD (1995). Myoglobin desaturation during exercise. Evidence of limited O_2 Transport. *J Clin Invest* 96, 1916–1926.

Tschakovsky ME & Hughson RL (1999). Interaction of factors determining oxygen uptake at the onset of exercise. *J Appl Physiol* **86**, 1101–1113.

Wilson DF (1994). Factors affecting the rate and energetics of mitochondrial oxidative phosphorylation. *Med Sci Sports Exerc* **26**, 37–43.

Non-Society scientific meetings

What do we want to see in brain imaging?
3–4 December 2007
Royal Institute of British Architects, London, UK (http://www.nyas.org/IMGconf).

BPS Winter Meeting
18–20 December 2007
Hilton Metropole, Brighton, UK
(http://www.bps.ac.uk).

Pacific symposium on biocomputing
4–8 January 2008
Fairmont Orchid, Hawaii, USA
(http://psb.stanford.edu).

International symposium on resistance arteries
17–21 February 2008
Hamilton Island, Great Barrier Reef, Queensland, Australia
(http://medicalsciences.med.unsw.edu.au).

We regularly receive information about scientific and other meetings from external organisations and post those that we think might be of interest to our Members on our web site. For the current list of forthcoming events please visit:

http://www.physoc.org/meetings

Contribution of myoglobin facilitated O_2 diffusion in respiring tissue

Sometimes scientists stumble innocently into a research question and then discover that the results do not match the accepted views. Hand wringing usually rues the naivety, while peers rush to explain away the new observations with methodology doubts. But if the observations hold, a new idea can rise.

For many, the function of myoglobin (Mb) poses no question. All biochemistry textbooks assert that Mb stores or transports oxygen. The view seems reasonable, given its structural features, its O_2 binding, and its homology with hemoglobin (Hb). Indeed, Kendrew and Perutz established a field of protein crystallography by describing the structure of these heme proteins. Monod, Wyman, and Changeux explained O_2 binding to Hb with a two state model paradigm. Given the tomes of experimental data, Mb or Hb structure or function should stand on a solid conceptual framework.

But Wyman actually puzzled over Mb function in the cell. Its concentration in terrestrial mammals seemed too low to serve as an adequate oxygen store. He proved mathematically a theory proposed by Wittenberg. Namely, Mb can compete effectively with free O_2 in delivering oxygen to the mitochondria. In essence the high O_2 carrying capacity of Mb overcomes the low solubility of free O_2. Two assumptions underpin the hypothesis: cellular O_2 in blood perfused tissue only partially saturates Mb, and cellular Mb must have sufficient mobility to compete with free O_2 diffusion. At high O_2 levels, as characterized by a fully saturated Mb, the diffusion of free O_2 will transport more efficiently than Mb. If Mb diffuses too slowly, it cannot compete effectively with free O_2 diffusion.

The Mb facilitated O_2 diffusion hypothesis, however, has languished for definitive experimental verification for over 40 years. Nevertheless, these ideas about Mb function form the cornerstone of current respiratory control models. Because ^{1}H NMR has

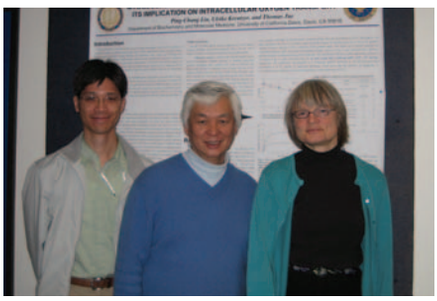


Figure 1. Ping Chang Lin (left), Thomas Jue and Ulrike Kreutzer standing in front of a Mb diffusion poster at the University of California Davis laboratory.

now detected the distinct γ CH_3 Val E11 and proximal histidyl $N_\delta H$ signals of Mb in myocardium and skeletal muscle, an opportunity has emerged to determine cellular PO_2 and to apply pulsed-field gradient techniques to map endogenous Mb translational diffusion (Kreutzer *et al.* 1992; Stejskal & Tanner, 1965).

In the basal state, ^{1}H NMR has not detected any deoxy Mb proximal histidyl NδH signal in perfused or *in situ* myocardium. In contrast, the Mb facilitated diffusion hypothesis envisions a partially saturated Mb in blood perfused tissue. Even as work doubles, the *in situ* myocardium shows no signs of Mb desaturation (Kreutzer *et al.* 2001; Zhang *et al.* 1999).

What about the other assumption in the Mb facilitated diffusion hypothesis? Solution experiments confirm that the NMR determined diffusion coefficients of 11.6 x 10^{-7} cm^2/s (Mb) and 7.53 x 10^{-7} cm^2/s (Hb) agree closely with literature reported values. In respiring rat myocardium, Mb diffusion drops by 37% to 4.24 x 10^{-7} cm^2/s. The cellular D_{Mb} matches the value predicted by the NMR rotational diffusion analysis (Wang *et al.* 1997).

Given the cellular D_{Mb}, the cellular Mb concentration, the Mb P50 (PO_2 that half saturates Mb), and K0 (the diffusion of free O_2 in the cell), the data analysis can assess the relative contribution of the O_2 flux from Mb (FO_2^{Mb}) vs. from free O_2 (FO_2^{O2}), as defined by an equipoise diffusion PO_2, the PO_2 in which Mb and free O_2 contribute equally to the cellular O_2 flux. For the rat heart, the equipoise PO_2 stands around 1.8 mm Hg. Below a PO_2 of 1.8 mm Hg, the Mb dependent contribution to the O_2 flux dominates. Because the NMR observation indicates that the basal state *in situ* myocardium operates with a PO_2 above 10 mm Hg, Mb cannot play a significant role in facilitating O_2 transport under normal physiological conditions in the myocardium.

The concentration of Mb alters significantly the equipoise PO_2. In terrestrial mammalian muscle, the Mb

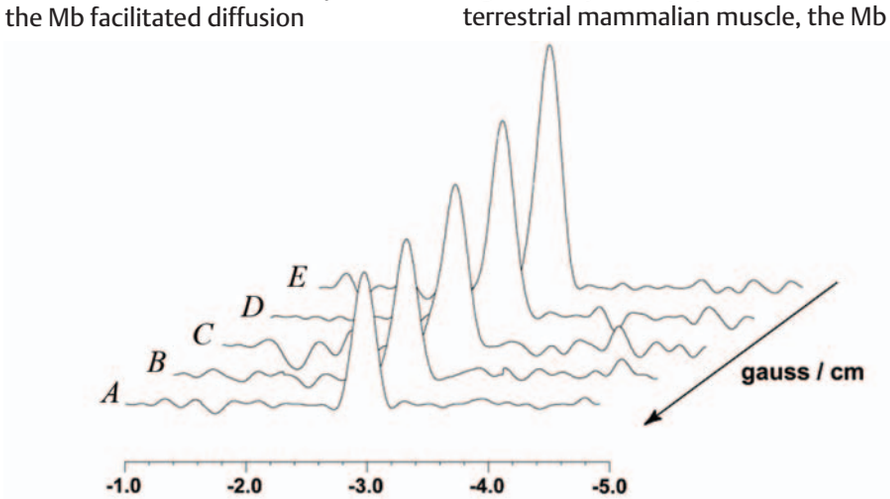


Figure 2. ^{1}H NMR diffusion-weighted spectra of MbO_2 from perfused rat heart under KCl arrest at 22 °C: a modified PG-STE sequence detects the peak intensity change in the MbO_2 γ CH_3 Val E11 signal at -2.8 ppm as a function of gradient field strength in the Y direction: *A*, 84.2 gauss/cm; *B*,74.8 gauss/cm; *C*, 56.1 gauss/cm; *D*, 37.4 gauss/cm, and (E) 4.7 gauss/cm.

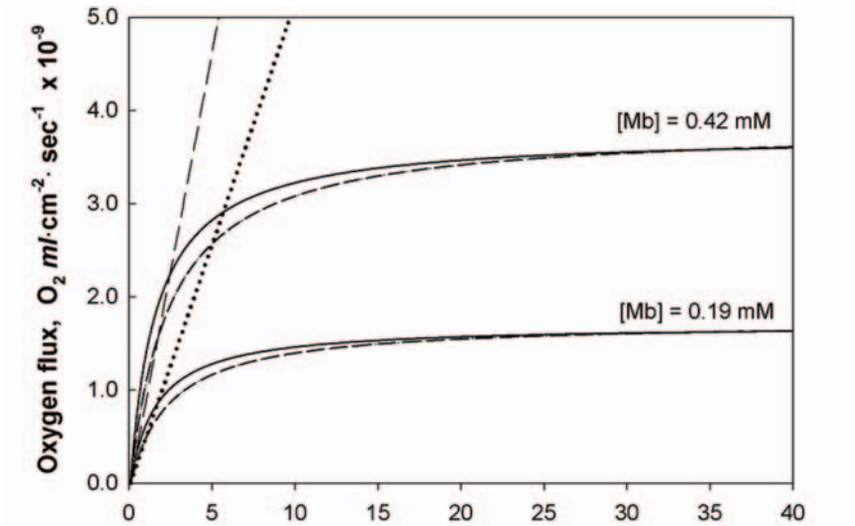


Figure 3. Plot of free O_2 flux vs. Mb facilitated O_2 diffusion as a function of PO_2 at 22 °C: the free O_2 flux increases linearly with PO_2. Two lines depict the rate of change given a cellular free O_2 diffusion constant of $K_0 = 4.28x10^{-5}$ ml O_2 cm^{-1}min^{-1}atm^{-1}and $K_0 = 2.52x10^{-5}$ ml O_2 cm^{-1}min^{-1}atm^{-1}. The two non-linear curves show the O_2 flux contribution from Mb facilitated diffusion increases as the cellular Mb concentration rises from 0.19 mM to 0.42 mM. With a P50 of 2.3 (---) and 1.5 mm Hg (—). With a P50 of 1.5 mm Hg, the equipoise PO_2 values are 1.77 (0.19 mM Mb, $K_0=2.52x10^{-5}$ ml O_2 cm^{-1}min^{-1}atm^{-1}) and 5.72 (0.42 mM Mb, $K_0=2.52x10^{-5}$ ml O_2 cm^{-1}min^{-1}atm^{-1}). With a P50 of 2.3 mm Hg, the equipoise PO_2 falls to 0.97 and 4.92 mm Hg, respectively.

concentration ranges from 0.19 mM in heart muscle to 0.42 mM in soleus muscle. Soleus muscle has then a higher equipoise PO_2 than myo-cardium. In fact, marine mammalian muscle with Mb concentration up to 25-70 g/kg tissue (approximately 1.4-4 mM) would have an equipoise PO_2 of 12-38 mm Hg. With marine mammals, Mb dependent O_2 diffusion dominates even under resting conditions.

What then is the function of Mb? Recent studies have claimed an NO scavenging role, even though other experiments have not detected the required presence of oxidized Mb (Flogel *et al.* 2001; Kreutzer & Jue, 2006; Kreutzer & Jue, 2004). Another perspective emerges from skeletal muscle experiments. At the start of muscle contraction, Mb desaturates within 30s to reach a steady state deoxygenation state, even though the cell has abundant O_2 to drive oxidative phosphorylation (Mole *et al.* 1999; Chung *et al.* 2005). MbO_2 appears to respond to a transient rather than a steady state change in energy demand.

The Mb translational diffusion coefficient also yields insight into the cellular environment. In solution, Mb diffuses isotropically; in the cell, Mb can exhibit anisotropic diffusion, since the longitudinal dimension exceeds the radial dimension by a factor of 10. However, the translational diffusion measurements show no orientation preference. Consequently, within a mean squared displacement of 3.8 μm, as determined from the measured D_{Mb}, Mb encounters no diffusion restrictions and experiences no cellular crowding, which could potentially modulate chemical reactivity. The cell contains many local fluid domains.

In summary, pulsed-field gradient NMR methods have determined the endogenous Mb diffusion in respiring rat myocardium. In myocardium, Mb cannot play a significant role to facilitate O_2 diffusion. In skeletal muscle, it may play a role at the initiation of contraction. In marine mammalian muscle, Mb contribution to O_2 flux predominates in the cell, which contains many local fluid domains.

What then is the function of Mb? Despite textbook reassurances, that simple question still challenges physiology and the search for the laws governing protein structure function relationship.

Acknowledgements

We gratefully acknowledge funding support from NIH GM 58688 (TJ), Philip Morris 005510 (TJ), and American Heart Association Western States Affiliate 0265319Y (UK) and the invaluable technical assistance of Jeff Walton and Jeff de Ropp.

Ping Chang Lin
Ulrike Kreutzer
Thomas Jue

Biochemistry and Molecular Medicine, University of California Davis, Davis, CA, USA

References

Chung Y, Mole PA, Sailasuta N, Tran TK, Hurd R & Jue T (2005). Control of respiration and bioenergetics during muscle contraction. *Am J Physiol Cell Physiol* **288**, C730–C738.

Flogel U, Merx MW, Godecke A, Decking UKM & Schrader J (2001). Myoglobin: a scavenger of bioactive NO. *Proc Natl Acad Sci* **98**, 735–740.

Kreutzer U & Jue T (2006). Investigation of bioactive NO-scavenging role of myoglobin in myocardium. *Eur J Physiol* **452**, 36–42.

Kreutzer U & Jue T (2004). The role of myoglobin as a scavenger of cellular NO in myocardium. *Am J Physiol* **286**, H985–H991.

Kreutzer U, Mekhamer Y, Chung Y & Jue T (2001). Oxygen supply and oxidative phosphorylation limitation in rat myocardium in situ. *Am J Physiol Heart Circ Physiol* **280**, H2030–H2037.

Kreutzer U, Wang DS & Jue T (1992). Observing the ^{1}H NMR signal of the myoglobin Val-E11 in myocardium: An index of cellular oxygenation. *Proc Natl Acad Sci* **89**, 4731–4733.

Mole PA, Chung Y, Tran TK, Sailasuta N, Hurd R & Jue T (1999). Myoglobin desaturation with exercise intensity in human gastrocnemius muscle. *Am J Physiol* **277**, R173–R180.

Stejskal EO & Tanner JE (1965). Spin diffusion measurements: spin echoes in the presence of a time-dependent field gradient. *J Chem Phys* **42**, 288–292.

Wang D, Kreutzer U, Chung Y & Jue T (1997). Myoglobin and hemoglobin rotational diffusion in the cell. *Biophys J* **73**, 2764–2770.

Zhang J, Murakami Y, Zhang Y, Cho Y, Ye Y, Gong G, Bache R, Ugurbil K & From AHL (1999). Oxygen delivery does not limit cardiac performance during high work states. *Am J Physiol* **276**, H50–H57.

Teaching ethics to physiology students

There are many sensitive and contentious areas within physiology such as the 'use of animals in research' or 'genetic modification', each with their own ethical issues or dilemmas. The practice of scientific research itself also has ethical implications, for example 'scientific integrity', 'data fabrication' and 'ownership of data'. We should be making our students aware of these issues and be providing training for them in ethics and ethical thinking. Indeed, the QAA biosciences benchmark statement[1] states that 'students should be confronted by some of the scientific, moral and ethical questions raised by their study discipline'; the provision of ethics training for postgraduate students is also required by the Research Councils[2]. We therefore set out to provide a comprehensive and progressive training in ethics and ethical thinking, not only to physiology students, but to all undergraduate and postgraduate students within the Faculty of Biological Sciences at the University of Leeds. Like most life scientists we have little or no training in philosophy. However, the University was awarded a HEFCE-funded Centre of Excellence in Teaching and Learning in ethics, the interdisciplinary Ethics Applied (IDEA) CETL[3]. We have therefore been able to work in partnership with applied ethicists from the CETL to design, develop and deliver truly interdisciplinary ethics teaching.

Our aim is not to produce philosophers but to train our students so that they have the ability to think through ethical issues as they arise, either in the remainder of their degree courses or subsequently in their future careers. As such, 'ethics' is probably too narrow a term, a training in critical thinking might be a more appropriate description as ethics teaching sessions provide an opportunity for students to thinking critically about a problem (but ethical issues in particular); to think

Dave Lewis - 'our aim is not to produce philosophers but to train our students so that they have the ability to think through ethical issues'.

'outside the box'; to learn how to develop and deliver rational/logical arguments including those with which they don't necessarily agree; all of which are key transferable skills that are highly sought after by employers.

When undergraduate students enter level 1, they have had little or no training in critical thinking; they also have scant discipline-specific knowledge. To cater for this we have developed sessions such as a 'life skills' seminar which considers the ethical issues surrounding alcohol promotions and cheap credit. Designed to be utilised either in freshers' week or the first 2 weeks of term when the students are facing these issues in real life, its purpose is to get them thinking/discussing a particular ethical issue without becoming bogged down by the underlying science. Having used this session to introduce them to ethical thinking, the remaining sessions in level 1 are science-based, covering both generic issues which are relevant to all students within the biological sciences (e.g. 'roles and responsibilities of a scientist' or 'plagiarism') and discipline-specific issues which students should be able to address from prior general knowledge (e.g. 'drugs in the 3rd world' or 'coaching children'). Individual programme managers are free to choose the most appropriate sessions for their particular cohort of students from the suite we have developed.

We build on these sessions at level 2, when students have acquired some subject-specific knowledge and are now more ethically aware, again covering generic issues which may arise in their degree programme or future research careers (e.g. 'scientific integrity') and discipline-specific issues (e.g. 'use of human subjects and the principles of informed consent'). These latter sessions are an opportunity not only to address the ethical issues but also to provide knowledge of the relevant UK law; this need to incorporate both ethics and law is best highlighted by sessions on the 'use of animals in scientific research'. The majority of undergraduate students in the life sciences do not use live animals in their studies; instead they undertake isolated tissue experiments or cadaver dissections. When such students are questioned, the majority have not thought about where the tissue has come from or the ethical implications of its use; the few that have, confess to blanking out such considerations. They also have little knowledge of relevant UK law and have serious misconceptions or misunderstandings about the use of animals in research, their opinions influenced by the popular and/or teen press. Student feedback has shown that, by the end of these seminars, this has been remedied; they now have both the knowledge and ethical awareness of the issues involved to enable them to make a more informed decision about the use of animals in research.

At levels 1 and 2, these ethics sessions are fully integrated within the most appropriate core modules in the different degree programmes, for example practical skills, tutorial or research orientated modules, ensuring that all students are provided with a training in ethics. Case studies, student notes and comprehensive tutor notes have been developed for each session enabling them to be delivered by a non-specialist. Our aim is to make these materials available, through the IDEA CETL[3], to colleagues at other Institutions.

By level 3, significant numbers of our students have developed an interest in

[1] The Quality Assurance Agency for Higher Education (2002) http://www.qaa.ac.uk/academicinfrastructure/benchmark/honours/biosciences.asp#1

[2] Research councils skills statement (2001) http://www.bbsrc.ac.uk/funding/training/skill_train_req.pdf

[3] Interdisciplinary Ethics Applied Centre of Excellence in Teaching and Learning (IDEA CETL). http://www.idea.leeds.ac.uk

ethics. To cater for such students, and in contrast to earlier levels where ethics teaching is provided within individual modules, we have developed an optional ethics module for physiology, pharmacology, medical sciences and neuroscience students which addresses issues including ‘research on embryos’ and ‘science and society versus the individual’. We are also trialling ethics-based final year teaching projects in local schools. Students deliver, as part of the school curriculum, a set of ethics-based teaching sessions on areas of current topical interest, evaluating ethical opinions before and after these sessions and also the effectiveness of the teaching package itself, writing the study up as their final year research project. These projects will cater for the increased public interest in ethical issues in science, provide student training in science communication and promote the public understanding of science in local schools. They are therefore a valuable alternative to wet-based laboratory projects and are particularly suitable for those students who don’t go on to research careers .

This development of ethics training is not restricted to our undergraduate programmes; we are also developing ethics training courses for all of our taught and research postgraduate students. These students come from a wide range of backgrounds, nationalities and cultures, resulting in different levels of ethical awareness and indeed ethical values. Our research suggests that they benefit most from discipline specific training delivered at a Faculty level rather than centrally delivered provision. We have therefore developed a series of ethics seminars for these students, with similar topics to those for undergraduates, but with the content altered to cater specifically for the needs of postgraduate students.

So how has this ethics teaching been received by students? Beforehand, the majority of students were unimpressed about having to attend ethics sessions and didn’t see the relevance to their studies. Afterwards, their opinions have changed. Quantitative and qualitative feedback has been very good, students enjoyed this teaching, they report that their awareness of ethical issues has increased and they requested more such sessions. We believe the success of this teaching is due to its delivery as individual sessions integrated within existing modules using topical and discipline-specific case studies. Evidence from other faculties in the University (where the CETL is also working) suggests that students do not engage with case studies they perceive as not relevant to them or where, at least at levels 1 and 2, ethics teaching is delivered as stand-alone modules.

This development of ethics training within the faculty is on-going. By the end of the 2007-2008 academic year, ethics will be firmly embedded throughout most of our under-graduate degree programmes at levels 1 and 2 and in all of our postgraduate programmes.

Our focus for the following year is to refine this teaching in response to staff and student feedback and also to develop a Faculty-wide level 3 optional ethics module which addresses the ethical issues arising from the principal areas of research within the Faculty.

Dave Lewis
Senior Lecturer, Institute of Membrane & Systems Biology, Faculty of Biological Sciences, University of Leeds and Interdisciplinary Ethics Applied Centre of Excellence in Teaching & Learning (IDEA CETL).

LETTER TO THE EDITOR

Living History

Professor Hugh Huxley of Brandeis University has pointed out to me that I made an error in my recent article (Elliott GF (2007). X-rays, twitching muscles and burning anodes. *Physiology News* **67**, 6-10) in suggesting Dr Max Perutz was responsible for the back-to- back publication of two papers (on X-ray diffraction data from living striated muscle during contraction) from King's College (London) and from Cambridge in Nature in 1965.

Hugh Huxley sent me a photocopy of a handwritten letter that I had sent to him on 7 May of that year attaching a copy of the paper the King’s College group submitted to Nature on 14 April. I had not seen this letter in 40+ years, and reading it again brought the events back to me with total clarity. As it happened, the letter was my solution to an internal problem among the KCL authors. Two of the authors, including myself, had thought it discourteous to Hugh Huxley that we had not sent him a copy of our paper when we submitted it to Nature. However, one author had wished to wait until it was accepted and had been supported in this view by other senior members of the KCL laboratory; I had found myself in the minority.

There was to be a muscle research dinner later in May that year. As the date approached without our hearing from Nature about the paper I felt increasingly uncomfortable at the prospect of talking to Hugh Huxley in such circumstances, since he had always been very friendly and helpful to me. I therefore asserted my rights as first author and sent him a copy; it was this act that initiated the negotiations that ultimately resulted in the back-to- back publication in *Nature*. I had forgotten these details because Jack Lowy carried out the actual negotiations. Jack subsequently took all the relevant copies and records with him to Denmark, including our copy of my letter, and I never saw them again.

I am happy to agree with Hugh Huxley that Max Perutz was not involved, as I had imagined and as I wrote in my article. My comment about Max was actually made with affection, since Max, now sadly missed by us all, was one of my scientific heroes and was a true gentleman in the best sense of the word. Hugh Huxley has pointed out, though, that what I wrote could be seen as a black mark on Max's memory. I am delighted that I can set the record straight and remove any implied, though unintended, criticism.

Gerald Elliott
Nuffield Laboratory of Ophthalmology, University of Oxford, UK

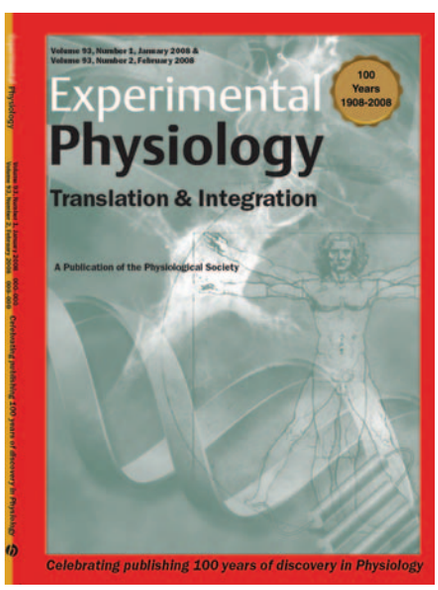
Celebrating publishing 100 years of discovery in physiology 1908–2008

Experimental Physiology
Translation & Integration

Chairman, David Paterson, welcomed the Editorial Board to Merton College, Oxford (above) for their September meeting. The meeting was well attended by Editors from the UK, USA and New Zealand, as well as representatives from Wiley-Blackwell and The Physiological Society. Plans were discussed for the journal's forthcoming centenary year.

Monthly online publication for 2008

After 100 years of predominantly quarterly and bi-monthly publication, *Experimental Physiology* will be published monthly online from 2008 to celebrate the success of the journal during its centenary year and to reflect increasing interest in it. Double issues will be printed and dispatched bi-monthly.



The Paton Lecture

The January 2008 issue, 93.1, will contain the Paton Lecture *Claude Bernard, the first systems biologist, and the future of physiology*, presented by Denis Noble at LifeSciences2007 in Glasgow in July.

Historical perspective series

Througout *Experimental Physiology*' s centenary year a series of invited articles will be published in which experts in the field bring a current perspective to a historical article published in *The Quarterly Journal of Experimental Physiology*. Contributors will include:

Uwe Proske on PBC Matthews (1962). The differentiation of two types of fusimotor fibre by their effects on the dynamic response of muscle spindle primary endings. *Q J Exp Physiol Cogn Med Sci* **47**, 324-333.

Walter St John on Fritz Buchthal (1959). Electromyography of intrinsic laryngeal muscles. *Q J Exp Physiol Cogn Med Sci* **44**, 149-159.

James FX Jones on P Alcock, JL Berry & I de Burgh Daly (1935). The action of drugs on the pulmonary cirulation. *Q J Exp Physiol* **25**, 369-392.

Roger Lemon on ASF Leyton & CS Sherrington (1917). Observations on the excitable cortex of the chimpanzee, orang-utan and gorilla. *Q J Exp Physiol* **11**, 135-222.

John Morrison on FJF Barrington (1925). The effect of lesions of the hind- and mid-brain on micturition In the cat. *Q J Exp Physiol* **15,** 81-102.

Simon Gandevia on Charles Reid (1928). The mechanism of voluntary muscular fatigue. *Q J Exp Physiol* **19**, 17-42.

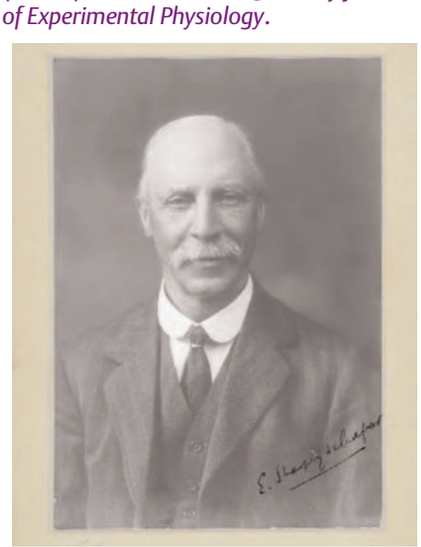
Roy Curry on CC Michel, JC Mason, FE Curry, JE Tooke & PJ Hunter (1974). A development of the Landis technique for measuring the filtration coefficient of individual capillaries in the frog mesentery. *Q J Exp Physiol Cogn Med Sci* **59**, 283-309.

The Journal of Physiology

Plagiarism in publishing: checking for duplication

Earlier in this issue (p. 41) Dave Lewis describes how Leeds University are tackling scientific ethics issues with students at all levels. This is welcome news for publishers who are finding that duplicate publication is on the increase, an observation that has been borne out recently in the Society's journals. In the past, detection of duplicated material in research papers was based largely on serendipity. Typically, the reviewer of a paper recalls that (s)he has read something recently by the same authors or on the same subject, checks up on his/her suspicions and finds that indeed parts of a previously published paper are reproduced without acknowledgement in the paper under review. New technology makes this kind of duplication all too easy but, copyright issues aside, it should be intuitively obvious that reproducing already published material, even your own, under different publishers' banners without acknowledging the primary

Denis Noble (above left) receives the Paton Lecture certificate from David Paterson; Sir Edward Sharpey-Schafer (below), founder of the *Quarterly Journal of Experimental Physiology*.



source is ethically dubious. Dave Lewis points out in his article that critical ethical thinking needs to be taught to trainee scientists. The courses offered at Leeds are a proactive part of this endeavour. Active plagiarism detection by educators and publishers, followed by appropriate sanctions against offending authors, serves to retroactively reinforce the message.

Recently search engines have started to play a part in unmasking duplicate publication. University teachers now routinely check student essays and course work against internet and database sources using duplication detection software such as *iThenticate*[1] and *Turnitin*[2] . These systems work by comparing submitted documents against the open internet and large databases of published content. Journal editors have been asking for the same facility for their publications offices, but the problem for journals has been that authors are likely to be duplicating material that is not available to plagiarism detection systems' search engines which access only material on the open internet or in their own databases of appropriate documents. In response to demand, *CrossRef*[3], the reference linking service now widely used by publishers, in partnership with *iParadigms*, the company behind *iThenticate*, has set up a pilot project, *CrossCheck*[4], to develop a database of licensed publishers' content which can be accessed by the *iThenticate* system. Wiley-Blackwell is one of eight publishers contributing material to the database for the pilot project and *The Journal of Physiology* is taking part. In the long term, it is hoped that the vast majority of publishers will add their content to the database, which will be available under specified terms and conditions to plagiarism detection systems. *CrossRef* believes that publishers will be willing to cooperate in creating the required database: 'Even if a particular publisher doesn't have a problem with plagiarized manuscripts, they should have an interest in making sure that their own published content is not plagiarized or otherwise illegitimately copied'[4]. One likely interesting feature of a final system will be that users can display the system logos on their web pages which will act as a warning to readers and future authors that duplicate publication will be sought out and is not acceptable practice.

The *Crosscheck* pilot project is running for 3 months until early in 2008. During that time Publication Office staff will test the software and feedback to the project team on the efficiency and user friendliness of the system. Based on the rise in cases of duplicate publication involving The Society's journals it is very much in our interests to take part in this initiative and to spread the message that plagiarism is regarded by the scientific publishing community as a serious ethical problem which we are taking steps to eradicate.

Carol Huxley

New Editors

Lothar Blatter, Mark Hargreaves, John Isaac, Stanley Nattel, Mu-Ming Poo, Allan Vaag, Anton Wagenmakers and Jerrel Yakel have recently joined the Editorial Board of *The Journal of Physiology*.

Jerrel Yakel received his BS from Oregon State University, and his PhD from the University of California, Los Angeles, where he studied ligand-gated ion channels and serotonin receptors in cultured hippocampal neurons and cell lines with Meyer Jackson. During a postdoctoral fellowship with the late Hersch Gerschenfeld at the Ecole Normale Superieure (Paris, France), he investigated the regulation of voltage-gated calcium channels by G protein-coupled receptors. During a second postdoctoral stage at the Vollum Institute with Alan North and Tom Soderling, he studied the function of regulation of ligand-gated ion channels. Jerry joined the National Institute of Environmental Health Sciences as an investigator in 1993, and is currently a Senior Investigator in the Laboratory of Neurobiology. His laboratory explores the function and regulation of ligand-gated ion channels, in particular the neuronal nicotinic receptor channels, in the hippocampus.



New *Journal of Physiology* Editors, clockwise from above left: Jerrel Yakel, Mark Hargreaves and Allan Vaag.

Mark Hargreaves is Professor of Physiology at the University of Melbourne, Australia. His research and teaching interests are in the physiological and metabolic responses to acute and chronic exercise, with a focus on skeletal muscle carbohydrate metabolism during exercise and the regulation of skeletal muscle GLUT4 expression. He is also a Consulting Editor for the *Journal of Applied Physiology* and an Associate Editor of *Exercise and Sport Sciences Reviews*.

John Isaac received his BSc in biochemistry with pharmacology and his PhD in neuroscience from Howard Wheal's laboratory at the University of Southampton, UK. During his postdoctoral training in Robert Malenka's laboratory at the University of California San Francisco he studied the mechanisms of synaptic plasticity in the hippocampus and barrel cortex. He started his own laboratory at the University of Bristol, UK, where he became a full professor in April 2004. In September 2004 he joined NINDS as an investigator and established the Developmental Synaptic Plasticity Unit. His group studies molecular and cellular mechanisms of developmental synaptic plasticity in the hippocampus and barrel cortex.

[1] http://www.ithenticate.com/static/home.html
[2] http://turnitin.com/login_page.asp
[3] http://www.crossref.org/webservices.html
[4] http://www.crossref.org/crosscheck.html

Joseph Francis Donegan 1893–1985

Joseph Francis Donegan was a much loved member of The Society for almost 60 years. Born in Gurteen, County Sligo in 1893, he read medicine at University College Galway where he was a brilliant student, graduating BSc (1914) and MB, BCh (1916) with first class honours and a love of physiology. His insatiable curiosity and ability led one of his teachers, J P Pye, and an external examiner, Joseph Barcroft (Cambridge) to guide him towards an academic career. Because local research facilities were limited, he was encouraged to train elsewhere.

Various travelling fellowships took him first to Cambridge, where he worked with Barcroft, Adair and Parsons on the relationship between blood pH and its O_2 and CO_2 combining power (Parson & Donegan, 1918–19). Then he worked with Thomas Milroy in Belfast on the colloid osmotic pressure of blood following haemorrhage (Milroy & Donegan, 1919). In 1919 he moved to University College London with an 1851 Exhibition Research Fellowship where he worked with Bayliss and Starling. Ernest Starling, with his interest in the contribution of venous return to cardiac output, encouraged a study of the nervous regulation of veins, a little understood topic at the time. With great technical skill, Donegan carried out the pioneer work (Donegan, 1921) that is still cited in contemporary reviews of vein physiology.

When J P Pye died in 1920 Donegan, though only 27 and relatively inexperienced, was invited to succeed him. These were difficult times in Ireland in the aftermath of a gruelling world war and civil insurrection at home. It might have been better for Donegan to remain in centres where his investigative talents would have greater scope. But his feelings for Ireland and his *alma mater* led him to accept the chair. So for a while full time research was replaced with near full time teaching but his investigative curiosity was not quelled. At the July 1924 meeting of The Society in Oxford, with Sir Charles Sherrington in the chair, he was elected to membership.

An opportunity for further research came in the summer vacation of 1927 when he visited Otto Warburg's world famous department at the Kaiser Wilhelm Institute for Biology in Berlin–Dahlem. Warburg won a Nobel Prize for Medicine and Physiology as did several others who worked there including Otto Meyerhof, Hans Krebs, Fritz Lipmann, Severo Ochoa and Hugo Theorell. The group met for sandwich lunches where they discussed their work. Donegan was so taken with the ethos of the group that he arranged to spend a sabbatical year there in 1928 at his own expense. During that year he collaborated with Hans Krebs in developing a method for peptide cleavage (Donegan & Krebs, 1929). He considered these times among the most exciting and influential in his life in both scientific and musical terms. He commemorated his time there by naming his house in Galway *Dahlem.*

When he returned to Galway, the role of intracellular potassium in excitation, a hot topic at the time, attracted his attention. This led to the John Mallet Purser Lecture he gave at Trinity College Dublin in 1942 (Donegan, 1942). Such was his interest that when he retired he set up a small laboratory in his own house to continue the work.

Donegan maintained an up-to-date knowledge in most fields of physiology by his avid reading, constant teaching and attending meetings, where he would sit in the front row and put penetrating questions to the speakers whatever the topic. Couched in simple terms with a puckish good humour and a sparkling eye, they enlivened the meetings but sometimes discomfited the authors.

In Galway where he held the chair for over 40 years, he became a legend. He had a liking for fast cars: he owned a Bugatti that was said to clear the road to the College of traffic when the approaching vroom of its engine was heard. He was a highly disciplined man of regular habits and somewhat obsessive about punctuality. His classes began and ended precisely on time. His lectures were clear, well discussed and larded with personal opinion. With a sharp tongue, he was outspoken to delinquent students, so was not often provoked. He ran the practical classes himself and came around to quiz each pair of students in turn on their work. His remit included biochemistry and pharmacology.

As a formidable hill walker he could out-walk most of his junior colleagues and contemporaries. He loved music and was very knowledgeable about it: it was said that his first attachment to Berlin was timed to coincide with the Beethoven Centenary celebrations. When in London for meetings he could usually be found the night before at concerts in the Royal Festival Hall.

When he retired in 1962 aged 70 he was elected to The Society's Committee where his wisdom was

widely appreciated. Through him The Society held its first and very successful meeting in Galway in 1965. In 1976, when he was 84, The Society elected him to Honorary membership, an honour he appreciated deeply. In 1985 he died peacefully at his home aged 93.

It is possible that Donegan's fertile mind, disciplined curiosity and experimental prowess were not exploited to the full in Galway and might have blossomed more spectacularly elsewhere. On the other hand the fun, excitement and discipline of physiology that he conveyed to generations of students, contemporaries and The Society should not be gainsaid – it was a memorable achievement by a memorable Member.

Acknowledgements
I would like to thank Sean Lavelle and Ron Whittam for their suggestions.

Ian C Roddie
Honorary Member, London

References

Donegan JF & Parsons TR (1918–19). Some further observations on blood reaction. *J Physiol* **52,** 315–327.

Donegan JF (1921). The physiology of veins. *J Physiol* **55** 226–245.

Donegan JF & Krebs HA (1929). Manometrische Messung der Peptidspaltung. *Biochem Z* **210**, 7–23.

Donegan JF (1942). Some aspects of muscle physiology. *Ir J Med Sci* Sixth Series, 561–571.

Milroy TH & Donegan JF (1919). The role of plasma proteins in diffusion. *Biochem J* **27**, 258–271.

The Benevolent Fund of The Physiological Society

I'd like to tell our Members a little bit about the Benevolent Fund. It was originally set up in 1976 '*for the purpose of assisting Members of The Society and staff and former staff (who by the nature of their employ-ment can be considered to have contributed to the advance of physiology) employed at teaching, research and industrial establishments who are in necessitous circumstances and their dependants'.*

It relies almost entirely on donations from Members of the The Society

The Fund is administered by The Physiological Society, but has separate charitable status with its own board of trustees. That means, in effect, no administrative costs so all income can be used for its primary aim – to support people in need.

Who does it support?

As its statute implies, not just Members of The Society but anyone who has contributed to physiology and is now in serious need. This includes academic staff, researchers, technicians, maintenance staff and other support staff.

What sort of help can it give?

Let me give you a few examples:

- A technician diagnosed with incapacitating motor neurone disease had to stop work and also adapt his house. He could not get any help at all from his local council and the Ben Fund obviously could not provide all the money needed (£30K plus), but it could (and did) give enough to help get a wheelchair and ramp to enable him to get in and out of the house;
- An academic staff member died prematurely without the usual staff pension. (He originally came from abroad and was never enrolled in the university's pension scheme by his employer.) The Ben Fund could not possibly substitute for a pension but it could (and did) give some immediate support to help his family through its initial financial difficulties;
- A technician in a physiology department has a son with a rare genetic disorder which requires full-time care by his parents. The Fund offered a grant to enable the son's room to be decorated so that he could live in a more stimulating environment. The Fund has also assisted with family holidays so that the family can get some much-needed time away together;
- A Member of The Society has a child who suffered from meningitis leading to profound hearing loss. The Fund paid for carpet to be fitted into their home, in order to maximize the child's residual hearing.

The main point to all of these is that the Ben Fund can act both rapidly and (even more important, perhaps) sympathetically, with minimal 'bureaucracy', at a time when help is most needed.

Where does the Ben Fund get its money from?

Partly from such activities as the raffles held at main meetings, but mainly from generous donations from Members of The Society.

So what can Society Members do to help?

Two things. One, make a donation. These can be one-off, but the most useful is some form of regular donation – it doesn't need to be much – and is even better if it is done under the Gift Aid scheme so that we can reclaim basic rate tax from the Chancellor. Forms for donations are available from Elfa at The Society's London office (see below).

The second thing you can do is to tell us if you know of anyone who needs (or even might need) help. You can do this quite confidentially – just drop a line or email to me or Elfa at the addresses below.

David Brown
Chairman, Benevolent Fund Board of Trustees, Department of Pharmacology, University College London, Gower Street, London WC1E 6BT
T: +44 (0) 20 7679 7297
F: +44 (0) 20 7679 4496
E: d.a.brown@ucl.ac.uk

Elfa Wilmot
The Physiological Society, Peer House, Verulam Street, London WC1X 8LZ
T: +44 (0) 20 7269 5713
F: +44 (0) 20 7269 5720
E: ewilmot@physoc.org

Is dual funding fit for purpose?

Ian McGrath and I attended a Parliamentary and Scientific Committee event on the 18 June, Is dual funding of our universities fit for purpose in the 21st century? Three speakers were lined up: Rama Thirunamachandran (Director of Research and Knowledge Transfer, HEFCE), Sir Keith O'Nions (Director General of Science and Innovation, Department of Trade and Industry) and Peter Cotgreave (ex-Director of the Campaign for Science & Engineering, formerly known as Save British Science.

Rama kicked off with the case for the funding councils. He emphasised that it had not been a dual funding system for some time, many different players were involved in a multi-layered funding system. The funding and research councils attempt to work very closely together to ensure that it does operate as a 'dual' rather than a 'duel' system, with the funding councils paying for the well found lab, the base on which the rest of the science funding system builds. The Research Assessment Exercises (RAEs) had proved to be a useful innovation since their inception in 1986, having raised overall research quality in the UK (shown by its correlation with a rise in the UK share of world citations) and encouraged more strategic research management in universities. However, over the years bureaucracy and game-playing has built up, and a consensus has developed that the system should be changed after the next RAE in 2008. There had also been problems with the assessment of applied/user led and multi-disciplinary research. The planned radical overhaul post-2008 will involve using a more metrics driven system, which will hopefully provide more robust indicators for international comparisons and reduce the admin load for academics. It is planned to pilot the new metrics based approach over the next few years, with it

influencing funding decisions from 2010–2011 and fully informing funding from 2014. Interestingly, some subjects – notably the arts, humanities, social sciences, mathematics and statistics – will continue to be peer review based as their publication habits (e.g. producing monographs rather than Journal papers), do not lend themselves so readily to bibliometric analysis. Rama wrapped up by saying that there will be a big debate over the next few years in moving to a long-term sustainable funding and assessment system for science, with key issues, such as the merits of concentrating/dispersing research funding and links between research and teaching, needing to be addressed.

O'Nions pointed out that most countries have a dual mode science funding system of some sort, involving project and core funding support. The basic value of the dual mode is not in question. Government policy is trying to achieve world class research in the UK, whilst increasing knowledge transfer and economic benefits and the addressing of national priorities on energy, climate change, ageing population, etc. The science funding system needs to be sustainable, support the best science, attract the best people and train them for a range of careers, be flexible and globally competitive. He asked what is the actual state of play? Research excellence in the UK is a good story to tell, the UK is very high in world rankings on citation productivity. Knowledge transfer and economic benefits are more difficult to quantify. The output of highly educated people into the broader economy is widely regarded as the biggest benefit, but income from activities such as licensing and consultancy has grown, patents granted have greatly increased, and impressive spin-out companies are now coming out of UK universities. Sustainability is a key issue, and has moved to full economic costing being used as a mechanism to ensure that the research councils pay an increased share of project support costs (now 80%, a big improvement on the previous position where they only paid marginal costs). Remaining challenges for Government are to improve the sustainability of the system still further, build improved incentives for funding to come from other areas such business, and streamline peer review.

Cotgreave then set out to try to provide a counterbalance to the rather rosy picture given by the two previous Government speakers. He agreed that no one would now argue that we do not need a dual support system. The role of the research councils in providing direct funding for specific projects, plus some of the associated support costs, was reasonably clear. The role of the funding councils was rather fuzzy, the definition of what their money is for having changed over the years, but broadly expected to cover infrastructure, and be able to be spent at university discretion to support potentially important blue skies research, research training etc. In practice it is often used to fill shortfalls in grants from other sources, for example charity funding that does not provide the same level of support costs as the research councils. It is supposed to provide funding for universities to make strategic investments, but for this to happen you need a surplus on a budget, it is simply not possible if all money is committed to existing activities. This modest surplus no longer exists; the reality is that, even with radically increased Government funding for science, the number of

institutions and individuals now chasing this funding has grown so rapidly that the share per capita has actually dropped since the 1980s. There have also been significant changes in governmental approach to science policy in the last few decades. In the 1960s, Government tended to define science policy as creating the right environment for research to take place, without directing research priorities. Now research objectives are highly strategic and directed from the centre through the funding bodies. The growing demands on the science purse have led to the invention of rationing mechanisms for apportioning research funding, at one level the RAE could be interpreted as just such a mechanism. The system seems to work well for 90% of funded research, but some vital blue sky research could be excluded.

A lively debate ensued, unsurprisingly research funding was an issue keenly felt by all the participants. Discussants were concerned by the 'cliff-face' effect of QR funding, where departments rated less than 5 get really hammered. Funding tends to be concentrated in a small number of elite institutions. Although this can help develop critical mass and increase international profiles, it can also exclude newer institutions that often make the biggest contribution to educating students from non-middle class backgrounds, students that would really benefit from being exposed to well supported research. Even elite institutions struggle; many of our top universities are trading in budget deficit each year. So-called ring fenced funding has been notoriously raided recently, notably Primary Care Trusts who took money intended for supporting academic posts in medical schools, and the DTI helping itself to the science budget. The dual funding system also has a downside in that when gaps are identified, it is often difficult to resolve who is responsible for addressing them. O'Nions sought to reassure participants on this point, saying that funding bodies spend a great deal of time and energy trying to resolve such issues. The Government speakers also received a lot of flak about how the funding mechanisms had let down foundation discipline areas such as chemistry so badly, that many departments had fallen between the support cracks and been forced to close. A stark warning from a member of the House of Lords was that biomedical sciences, so vital for this country's industry, might suffer a similar fate if its high lab and training costs were not adequately supported. O'Nions admitted that this was a complex problem, and that more needed to be understood about the real costs of supporting departments. There was a general feeling in the room that as a nation we need to stop pretending that we can have a world class education system on the cheap. We need to be realistic about what we expect to achieve, there simply isn't enough money in the system to pay for all our expectations.

A logical extension of the dual system, if the funding council stream is increasingly targeted and is to be related by metrics to research project income and outputs, is to accept that we actually have a unitary system. On being asked how far joined up thinking went, by way of an overview of all research funding, the speakers acknowledged that there was a government committee that took such an overview on which, for example, the funding councils and the DTI, responsible for Research Councils, were represented. The logic of moving to a unitary system has clearly not escaped this group, although there are obstacles to implementation such as the current devolution of funding council but not research council streams, let alone the likely opposition from many interested groups. This is potentially one of the most interesting and explosive outcomes of the current debate on how research throughout the UK is funded. Is dual funding already dead?

Liz Bell

PHYSIOLOGICAL SOCIETY MEETINGS 2008/2009

2008

Leeds, UK
17–19 March
Cardiac & Respiratory Physiology Themed Meeting with a Focused Symposium on *Determining control of the cardiovascular system in health and disease: from brain to blood vessel*.

Cambridge, UK
14–16 July
Main Annual Meeting.

Oxford, UK
9–11 September
Metabolism & Endocrinology Themed meeting with a Focused Symposiun on *Orchestration of metabolism in health and disease*.

Shanghai, China
12–16 September
International Workshop on *Latest advances in ion channel techniques applied to physiological problems*.

Beijing, China
20–22 October
Joint International Meeting of The Physiological Society with the Chinese Association for Physiological Sciences and the Canadian, Australian and American Physiological Societies.

King's College London, UK
15–17 December
Vascular & Smooth Muscle Physiology Themed Meeting with a Focused Symposium on *Vascular responses to mechanical stress: cellular cross-talk and integration*.

2009

University College Dublin, Republic of Ireland
6–10 July
Main Annual Meeting.

Woods Hole, MA, USA
September
Joint International Meeting with the Society of General Physiologists on *Basic biology and disease of muscle*.

For full details of Society Meetings and International Workshops visit http://www.physoc.org/meetings.

Scientific Meetings Calendar 2008

Meeting	Meeting Location	Meeting Date	Meeting Type	Abstract Submission and Registration Opens	Abstract Submission Closes	Online Programme Available	Published as Proceedings of The Physiological Society	Travel Grant Application Deadline
Cardiac & Respiratory Physiology Themed Meeting Focused Symposium: "Determining Control of the Cardiovascular System in Health and Disease: From Brain to Blood Vessel"	Leeds UK	17-19 Mar	Themed	14 Jan	31 Jan	29 Feb	Yes	31 Jan
Physiology 2008 Main Meeting	Cambridge UK	14-16 Jul	Main	1 Mar	31 Mar	1 Jun	Yes	31 May
Metabolism & Endocrinology Themed Meeting Focused Symposium: "Orchestration of Metabolism in Health and Disease"	Oxford UK	9-11 Sep	Themed	23 Jun	18 Jul	20 Aug	Yes	31 Jul
Latest advances in ion channel techniques applied to physiological problems	Shanghai China	12-16 Sep	International Workshop	1 Apr	1 Jun	15 Aug	No	31 Jul
Joint International Meeting The Physiological Society and the Chinese Association for Physiological Sciences, The Canadian Physiological Society, The Australian Physiological Society, and the American Physiological Society.	Beijing China	20-22 Oct	Joint International	15 Apr	15 May	15 Jul	No	31 Jul
Vascular & Smooth Muscle Physiology Themed Meeting Focused Symposium: "Vascular Responses to Mechanical Stress: Cellular Cross-Talk and Integration"	London UK	15-17 Dec	Themed	29 Sep	17 Oct	24 Nov	Yes	31 Oct

Other Key Dates

Date	Activity	Deadline
1 December 2007	Call for symposium proposals for Physiology 2009 (Main Meeting - Dublin)	31 January 2008
1 October 2007	Call for proposals to host 2009 Themed Meetings	29 February 2008
1 March 2008	Call for proposals to host Physiology 2010 (Main Meeting)	1 June 2008
July 2008	SIG Convenors meeting	N/A
March 2008	Meetings and International Committee Meeting (08.1)	N/A
October 2008	Meetings and International Committee Meeting (08.2)	N/A

Non-Society Symposium Grants
(max £1,000 each)
Deadlines in 2008
31 March
31 July
31 November

Special Symposium Grants
(max £5,000 each)
x2 awards
Deadlines in 2008
At least 2 months prior to event
(for symposium in 2008)

Introduction of registration fees

From 1 January 2008, Council have agreed that all Members and Affiliates will be required to pay registration fees to attend all Society meetings. This will be combined with substantial increases in the fees for non-Members to attend our meetings. We will also be introducing an early-bird registration deadline for all meetings, and Affiliates will continue to enjoy free registration up to that deadline.

This decision has not been taken lightly and we have set fees at as low a rate as we feel that we can. It also comes at a time of continued uncertainty about the long-term future of income sources for The Society and we believe it is a necessary step for us to take if we are to ensure the delivery of high quality meetings. These issues were raised at the 2007 Annual General Meeting in Glasgow and recently ratified by the Meetings & International Committee and subsequently Council.

We hope that you appreciate the rationale for this move and that you will be aware of the consequent improved quality of meetings.

Prem Kumar
Meetings Secretary

Registration fees for 2008
(includes the cost of the welcome reception (if applicable), conference material, lunch, tea and coffee) for the duration of the meeting, but does not include accommodation, conference dinner or any additional social events)

Fees for Themed Meetings (NO DAY RATES)

Membership status	Whole meeting *Early-bird registration*	Whole meeting *Late registration*
Retired & Honorary Members	Free	Free
Ordinary Members of The Physiological Society	£25	£50
Affiliates of The Physiological Society	Free	£30
Non-Member of The Physiological Society	£100	£200
Non-Member **student**	£50	£100

Fees for Main Meeting (NO DAY RATES)

Membership status	Whole meeting *Early-bird registration*	Whole meeting *Late registration*
Retired & Honorary Members	Free	Free
Ordinary Members of The Physiological Society	£50	£75
Affiliates of The Physiological Society	Free	£45
Non-Member of The Physiological Society	£150	£250
Non-Member **student**	£75	£125

Lister Institute Research Prizes 2008
Applications are invited from outstanding young researchers in biomedical or related biological sciences for the Lister Institute Research Prizes 2008. Up to four awards will be made on the basis of originality, quality and potential significance of the research and on the achievements of the applicant. Candidates must have more than 3 and less than 10 years' post-doctorial experience on 1 October 2008 and must have guaranteed employment for the first 3 years of the notional years of the award in any not-for-profit institution. Prize winners will receive £200,000 which may be used in any appropriate way to support their research, other than the provision or augmentation of personal salary.

For full details and application forms please visit www.lister-institute.org.uk

Completed forms must be returned no later than Friday 7 December 2007

Undergraduates – write an article and win £500!

AstraZeneca is generously supporting the career development of undergraduates studying physiology as part of their degree. From 2008 there will be two annual prizes for the best physiology-related articles written by undergraduates and published in *Physiology News*.

Undergraduate students are invited to write articles, suitable for publication in *Physiology News*, on topics which might include (but are not limited to):

- summer/final year degree projects;
- experience of attending a workshop or conference;
- what turned you on to physiology;
- topical/current physiology-related news items;
- outreach activities (e.g. school visits, science festivals);
- research groups/activities within the department;
- review of a paper that encouraged an interest in physiology.

Articles should be submitted by 1 March or 1 September 2008 and should not exceed 1000 words and can include an illustration. Submissions should be sent to education@physoc.org and will be judged on appropriateness of topic and writing style by The Physiological Society's Chief Executive Officer, the Education and Membership Manager and a member of Physiology News Editorial Board, with agreement from AstraZeneca on the winning article.

Each prize will be £500 and a day visit to AstraZeneca's headquarters in Alderley Park, Cheshire (to include UK travel, dinner and overnight accommodation).

For full details contact Mike Collis (mcollis@physoc.org).

BIOSCIENCES FEDERATION

Are bioscience learned societies 'not fit for purpose'?

Competition is an essential part of the scientific research culture. We aim to do 'internationally competitive' research and funding is on a 'competitive' basis. In reality, many laboratories seek to be first amongst equals because there are few accolades in being the second to make a discovery. This is often difficult to achieve and highly competitive laboratories frequently collaborate in order to be first together. In some modern biology this collaboration can be on a massive global scale. In general we thrive on competition and collaboration: the time to be worried is when nobody is interested in collaborating with you – it suggests that you are uncompetitive and have nothing much to offer. This competition is a major driver for the success of science but there are occasions when it is unhelpful. I don't intend here to review the major 'spats' in the biosciences but will remark only that the current (August 2007) intense competition between two F1 racing drivers is not exactly improving the morale of their team. In summary, competition can be very positive but this upside can be replaced rather quickly by a negative effect if the competition is inappropriate.

The motor racing example is relevant because the competition is inside a single team. The drivers have created an 'internal market' to be the best. They risk taking their eyes off the 'external market', which is the real competition provided by other teams. In the biosciences we sometimes behave like Lewis Hamilton and Fernando Alonso!

There are very many organisations active in the biosciences. I believe that there are at least 80 learned societies and about half of these have come together in the Biosciences Federation. But that is not the point. These societies are together whether or not they are part of an umbrella organisation: They are 'together' in Team Bioscience. 'Together' the Team faces competition for money, students, specialists and infrastructure from the arts, the humanities and other branches of science and engineering. Team Bioscience has created a competitive internal market and has sometimes lost sight of the real competition.

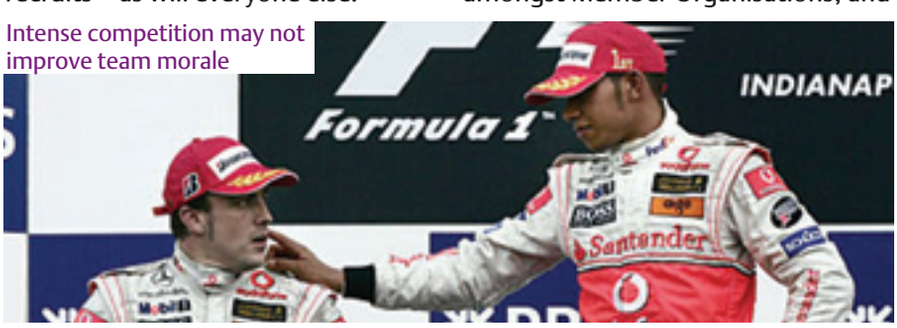
Yes, there is a little hyperbole in the last sentence – but not much! Let us consider some examples. Quite a few learned societies have staff working to get teaching resources into schools. My strong impression is that they are informed and dedicated people doing excellent jobs. But over and over again I hear that part of the motivation is to expose young people to the words that describe their society – 'microbiology', 'endocrinology', 'physiology', 'biochemistry', 'ecology' and probably all the other ologies! This is crazy because it is impossible for a single school to take on all these different resources, provided by different societies, and focused on a separate sub-discipline of the biosciences. The internal market means that 'microbiology' is successful if it gets more 'hits' than 'endocrinology'. Do we really think that this form of internal competition is helpful? What we really need is more students as a whole thinking of the biosciences as a career. What we really need is a bigger and better qualified pool of young bioscientists. When we achieve these goals both the microbiologists and the endocrinologists will get more recruits – as will everyone else.

And what do our real competitors do? The Royal Society of Chemistry is also active with young people. They also look to create teaching resources. However these are not focused on 'analytical chemistry', 'synthetic organic chemistry', or any other subset of the subject of chemistry. Their efforts are exclusively focused on chemistry because first and foremost they want to encourage more young people to become chemists. Their policy is wise.

Where else is there an internal market? Certainly in policy work – and this is partly my fault! Currently it is quite common for the Biosciences Federation to respond to an enquiry and find that several Member Organisations have produced their own response. I shall work harder to ensure that competition with Member Organisations is reduced but I can't really influence a decision of several members to 'do their own thing'. I am quite convinced that this is ineffective and therefore a waste of money. Somehow the BSF must increase its catalytic capacity to get a unified voice for the biosciences on the major topics that do and will impact on the future health of our discipline.

More broadly, we must now identify those areas where Team Bioscience has created an internal market that acts as a detriment to success in the external market. Solutions are possible for both the examples given above. The BSF was established to help provide these solutions and I detect that the landscape is changing in a way that will allow this to happen more readily than in the past. There is increasing awareness amongst Member Organisations, and

Intense competition may not improve team morale



their membership, that a structure for the biosciences that seemed to work in the mid 20th century is no longer appropriate today. Several weeks ago I was at an open meeting where Sir David King (Chief Scientific Advisor to the Government) asked in his talk if the severely fractured bioscience landscape was 'fit for purpose'! The answer depends on which purpose you consider. If the purpose is for systematists, ecologists, physiologists or plant pathologists to meet to talk about their work, the answer is a qualified yes. In the practical science context, societies are successful 'special interest groups'. The 'yes' is qualified because young people don't always relate strongly to the disciplines around which some societies are built and this may be a future problem for them. However, it is very much harder to answer 'yes' if the 'fit for purpose' question refers to outreach and to engagement with local, national and European politicians and opinion formers. These areas need Team Bioscience.

Some competition between learned societies will remain for the foreseeable future: the competition for membership is an obvious example. However, I do believe that there is an increased wish for us all to work together whenever appropriate and possible. The wish was rather theoretical 2 years ago: today I see much more desire for its implementation. I am confident that Team Bioscience will be built. Its shape and final structure is a little uncertain, but I am confident of the outcome because the need is so great.

Richard Dyer

Do learned societies behave like racing drivers ?

I re-read Richard Dyer's interesting article this morning whilst blocking the access to my local garage with my car to prevent the CEO of the Society of Endocrinology from filling hers. I then had a busy day trying to poach Biochemical Society members by offering them our highly attractive travel grants, followed by visiting local schools to convince the kids that physiology is actually the only biological science and all the rest are derivative and therefore inferior. My evening was spent loitering in the House of Commons waiting to pounce on Science Minister Ian Pearson and convince him that the HEFCE funding formula for physiology should be double that of the other biological sciences.

Joking apart, Richard Dyer's article raises many issues, some that I recognise and some that I don't. I really don't believe that societies compete for members; many of us are members of a number of societies and see no conflict between them. Regarding competition in education specifically –The Physiological Society has been a strong supporter of the BSF and a major participant in the BSF Education Committee, contributing our staff to supporting BSF-badged educational events. We are also very active on the Animal Sciences Group , the Journals Working Party and in contributing to BSF responses to Government. I also seem to recall an excellent scientific meeting we just held in Glasgow jointly with the British Pharmacological Society and the Biochemical Society. So maybe we can work closely with other societies on an increasing number of intiatives without needing to compete at all.

Some important issues that I do recognise in Richard's article are how much we collaborate, with whom and whether the BSF has a role in this. We clearly have common scientific interests with a number of other biomedical societies, and I am all in favour of working with them whenever it is mutually beneficial. However, 'horses for courses' applies here. For instance, although the term 'physiology' encompasses all living organisms, our Society is not involved in plant/microbial research and there are fewer opportunities for scientific collaboration with BSF member societies who deal with these areas of biology. However, we do actively collaborate with a number of these societies on common areas of education and publishing.

So I am in favour of working together in teams as long as the team has added value for all members.

What is the BSF's role in these collaborations ? According to its web site the BSF was established to:

- promote liaison, dialogue and interactions within the diverse community of bioscientists on common issues that relate to research and teaching;
- provide opinion and information to assist the formulation of public policy;
- promote wide and open debate, involving the wider public where appropriate, about the practical and ethical issues surrounding developments in the biosciences and their applications.

I wholeheartedly agree with all these aims and think that the BSF provides an important networking opportunity for societies which can facilitate alliances. However, I don't think that these alliances should then necessarily be badged under the BSF 'banner'.

The BSF is primarily there to provide a conduit to Government on policy matters. Should it be the only conduit and is it futile for individual societies to make their own responses to Government? To my mind the answer is both yes and no – it all depends on the issue. There are many overarching issues where there is consensus amongst the societies and where the BSF should co-ordinate the response. There are also issues where societies will disagree and will want to express the views of their members independently. This may be confusing to Government, but what's new about that? Of course Government would be easy if everyone agreed about everything. But one hopes Government has expert scientific civil servants who can enlighten them about the bioscience community and its sometimes common, sometimes diverse viewpoints.

As I said, I do recognise some important issues in Richard's article. The distinctions between biological disciplines are breaking down and it is sometimes hard to determine where, for instance, physiology ends and biochemistry begins. Lurking in this debate is the idea that working more closely with related biomedical societies could in the long term lead to merger. I can see this happening in the future, but when is a very different question. One 'Doomsday' scenario is that the publishing income that supports most of the major societies will be eroded so much by open access publishing that merger will be the only way to continue to operate. Should we be pro-active and merge whilst we are still strong ? I think these are very important questions that our Society should be discussing. But wait a minute – merging societies would mean there would be fewer CEOs; I had better dash off and let some tyres down at the BPS offices in Angel Gate ...

Mike Collis

Are you a laboratory animal? Do you have an emotional, sexual or psychic problem?

Dr Keith Cormorant will answer your problems and give honest advice on life, love, sex and the odd physiological experiment

If you are an animal employed in physiological research and would like to discuss an emotional, sexual or psychological problem please contact Dr Cormorant, an interspecies psycho-dynamic councillor specialising in psychic transference and aura balance issues in a range of animal species. Following a minor misunderstanding with DEFRA, Dr Cormorant is now joined 'on the other side' by *Physiology News* new spiritual advisor Shambo the Bull.

Worried about job security?

I am a beagle who smokes cigarettes for a living in a tobacco company. I am concerned that the new legislation banning smoking in the workplace may apply to lab animals. Will I still have a job to go to or will my company outsource my job to a Beagle in Bangalore.

Keith The Home Office has informed us there are no plans at present to extend legislation banning smoking in the workplace to dogs.

Shambo Relax, dogs are freely allowed to smoke and consume alcohol on the other side. Cannabis is also legally available and does not lead to spiritual psychosis.

Xenopus toads. In love but is there a psycho-sexual problem?

We are Xenopus tank mates; for the last 3 years we have produced oocytes for a molecular biology lab. We have just been retired from commercial oocyte production and have been re - homed, but we can no longer perform sexually without regular injections of chorionic gonadotrophin. Will we ever have tadpoles of our own?

Keith You need to relax and learn to relate to each other in a non-experimental context. There a number of web sites that may help to enhance your romantic responses (Cheap.Fake.Russian.Viagra.For.Mugs .com and Red Hot Asian Toads.com).

Shambo We are all sensual beings spiritually connected to a sensual universe. The aura can be very sensitive to disturbances, death threats by DEFRA in particular are known to affect the aura and dampen the libido. I would suggest changing to a vegetarian diet, and reading my new leaflet *Unlocking the sexual psychopath within! Applied Tantric sex therapy for the amphibian psyche* may provide you with some answers.

An insomniac mouse fears the darkness

I am a transgenic mouse with luminescent genes. This causes me problems with insomnia. When they switch the lights off in the mouse house I glow so brightly that I can't go to sleep. What can I do?

Keith Perhaps rearranging your bedding so that it covers your head may provide some relief.

Shambo I am disturbed by Keith's flippant answer; there is such a thing as Karma and the universe will appoint a spiritual nemesis to deal with Keith's attitude. There is unlimited energy available in the universe and a driving force that wants you to have the undisturbed sleep that you deserve. May I suggest reading my other leaflet *Achieving emotional stability and cosmic mastery using colonic irrigation* . Try to think positively – you won't need to switch the lights on to read it.

I am a cloned mouse. Will there be a soul for me or will I need to share?

I am a cloned mouse with a spiritual identity problem. I am identical in every way to my cage mates. My physical self has been perfectly cloned but what about my soul? Have our souls been cloned or will we have to share?

Keith I am afraid that I may have to pass you over to Shambo on this one.

Shambo Since arriving on the other side I have begun an emotionally fulfilling inter-species relationship with Dolly the sheep who assures me that every clone has its own individual soul.

Dr Keith Cormorant is a registered charity. Dr Keith Cormorant and Shambo the Bull regret that they are unable to enter into any personal correspondence. Although he harbours no spiritual grudges, Shambo is hoping to be reincarnated as administrator of the DEFRA pension fund.

David Jordan
1953–2007

David Jordan BSc, PhD, DSc (University of Birmingham), Professor of Physiology (UCL, University College London) died aged 54 on Saturday 30 June 2007 following a short illness. Born on 14 April 1953, he was the only child of Brenda and Harry Jordan. He was raised in Jarrow, Tyneside, North East England and attended Jarrow Springfield Secondary School (1964-1971). From 1971-1977 he studied at the University of Birmingham receiving a BSc in Biological Sciences (1974) and a PhD (1977) for his studies on *The termination and excitability of sinus nerve afferents* (supervisor Mike Spyer).

In 1980, following a 3 year research fellowship in the Physiology Department at the University of Birmingham, he moved to a lectureship in the Department of Physiology at the Royal Free Hospital School of Medicine (University of London). David was promoted to senior lecturer in 1988 and to reader in 1993. He spent a sabbatical (1988-1989) at the Cardiovascular Institute, University of California, San Francisco where he was a Julius H Comroe Research Fellow. Following his promotion to Professor in 1999, and the merger with UCL, David was made Head of Department on the Royal Free Campus.

Over the period 1980-1988 David's primary research association was with Mike Spyer. He was awarded his first independent research grant in 1983. In 1988 he began a research collaboration with Andrew Ramage, funded by the Wellcome Trust and British Heart Foundation, which continued until the time of his death. The many papers that flowed from their partnership have significantly increased our understanding of the role of 5-hydroxytryptamine receptors in the central nervous control of cardiovascular function.

His various review articles, book chapters and editorial activities (a member of the Editorial Board of *The Journal of Physiology* from 1989 to 1996 and a Senior Editor from 1992 to1994) exemplify David's scholarship. He was a member of the British Heart Foundation's Project Grants Committee from 1998 to 2002.

David's input to teaching, examining and committee work was considerable. He ran a number of courses in both the

science and medical degree rogrammes, and served as an undergraduate and higher degree external examiner both within the UK and overseas.

David's numerous contributions to academic life will be missed greatly by colleagues. David is survived by his parents and Ken, his partner for more than 30 years.

Michael P Gilbey

An appreciation by an old student

Physiologists of Dublin were shocked to hear of the premature death of this fine scientist. David was an authority on autonomic neuro-science, but not an authoritarian by character.

I was his first PhD student. The interaction between mentor and pupil was of an old-fashioned type and David made me feel free in the laboratory we shared. This intellectual freedom, so beloved of Carl Ludwig[1] , commenced with a digression; David's grant was for studies on central control of airways but my doctoral studies wandered into the vagal control of the heart. Obviously David agreed with Laurence Sterne[2] that digressions are the sunshine and soul of learning.

Of physiological material he was a constant reader and he had an enviable talent for critical analysis. He was drawn to papers exhibiting internal consistency, logical structure and clarity. In a phrase he relished elegance in science. He was adept at rapidly uncovering contradictions or inconsistencies. At meetings of our Society when David exposed two mutually exclusive statements, he would enquire optimistically which one was correct. David's farewell gift to me was a copy of Fulton's *Selected readings in the history of physiology*. It contains small extracts of seminal papers, little kernels heralding imminent revolutions in physiological knowledge.

Few of us stumble upon great discoveries and Ernest Starling[3] wrote that prizes and honours should not be awarded to the fortunate discoverer, since the joy of discovery contains its own reward, but should be used to console the careful scientist, the constant reader, the corrector of errors, the humble assiduous gardener of the growing tree of knowledge whose fruit will come in due season. In his brief letter to *Nature*, Starling might have been writing about my future supervisor. David Jordan had a kind heart. God rest this fine scholar.

James F X Jones

1. Carl Ludwig (1816-1895) wrote in a letter to Setchenow: 'It is mandatory to be at a place where one feels most free in spirit. Only there one is able to make the greatest progress'. Cited in Zimmer H G (1996). Carl Ludwig: the man, his time, his influence. *Pflugers Arch* **432**, R9-22.

2. Laurence Sterne (1713-1768). Author of *The life and opinions of Tristram Shandy, gentleman*.

3. Starling E H (1924). Discovery and research. *Nature* **113**, 606.

Reminiscences

I am not sure exactly when I met Dave Jordan. It was sometime in the early 1990s, a few years after I had begun researching central nerve pathways that control the cardiovascular system. Lynne Weaver (University of Western Ontario, Canada) introduced us at a conference in the US. For a number of reasons, Lynne thought Dave was an absolutely lovely man. She had spent 1979 at the University of Birmingham, where Dave had taught her how to do extracellular electrophysiology on central neurons, and he was one of the main reasons that her year in Birmingham was so much fun.

Through the 1990s, Dave and I met at conferences where we shared a number of highly enjoyable scientific conversations, drinks and dinners. As a result, I made my first visit to give a talk at the Department of Physiology at what was then the Royal Free Hospital School of Medicine in 1996. However, my relationship with Dave did not really begin to 'hot up until around the turn of the century. Although my focus until then had been spinal cardiovascular control pathways, I was becoming increasingly interested in the medulla. We began to discuss the possibility of

setting up in Dave's laboratory the method of juxtacellular labelling, which would allow us to examine the anatomy of neurons in the nucleus of the solitary tract (NTS) after they had been characterized electrophysiologically and pharmacologically. Dave had just received funding from the Wellcome Trust for a project on the intracellular recording of NTS neurons, so there was no problem in switching to a method that would produce a higher yield of filled cells.

From our first discussions about collaborating, Dave's depth of knowledge about central cardiovascular and central respiratory control, not only in standard laboratory mammals but also from a comparative perspective, was particularly impressive. He was also very good at explaining the intricacies of NTS electrophysiology and pharmacology to a novice ('There are *how many* 5-HT receptor subtypes in the NTS!') and showed great patience in the face of some pretty uninformed questions. These qualities clearly made him such a respected and well-liked teacher, who in his last years was reviewing the curricula and performance of new medical and dental schools in south east Asia. I think the fact that he was an unassuming boy from the north of England with very few airs and graces, even when he became the Professor of Physiology at the Royal Free and University College Medical School in 1999, also contributed to the empathetic relationships that he had with students and colleagues.

Dave employed Gareth Jones on his Wellcome Trust grant and, within weeks, Gareth and Dave were successfully filling NTS neurons with Neurobiotin and doing histochemistry to reveal them with ExtrAvidin-horseradish peroxidase. This success made me appreciate another two of Dave's admirable scientific qualities. One was that he was truly a technically expert electrophysiologist, able to get complex and difficult methods up and running when other less accomplished researchers could not. The second was that he was not only willing, but also able, to follow very detailed instructions given to him by a neuroanatomist with very exacting standards, i.e. me.

The auspicious beginning to our joint work made by Dave and Gareth became the basis for a major part of my grant application to the National Health and Medical Research Council of Australia in 2000. This application was funded from 2001 to 2003, with Dave as an Associate Investigator. We also received support from the Wellcome Trust in the form of a Biomedical Research Collaboration Grant (9/2002-8/2005) that facilitated a number of reciprocal visits between Dave's and my laboratories.

Because Brits think Australia is very far away and Australians think that the UK is within easy reach, I visited Dave's laboratory many more times than he visited mine. Being a kind and generous person, Dave offered me a bed at his house for the duration of my first visit and that became our pattern. Most of my fondest memories of Dave come from my stays at 108 Purves Road. We had our favourite restaurants in his neighbourhood, where we enjoyed many memorable meals after working late in the lab . At least once during every visit, we would eat a frozen Indian meal from Sainsbury's at his kitchen table, drinking a bottle of Australian (or any old) red, talking serious science and intermittently gossiping our heads off. Dave always had a balanced view of life. Regardless of how busy our schedule was (or how keen I was to keep working), he made sure that we took time off to do something non-scientific. There were visits to stately homes, museums and sights of interest. Our last interlude was a wonderful day together at Aylesbury on our way to the 2005 Physiological Society meeting in Bristol. Dave was a keen gardener, and whenever I visited in spring or summer, a tour of the garden was mandatory, as was a discussion about the neighbour who had someone come in every 6 months to scythe the weeds that invaded her backyard.

I was always touched by the fact that Dave was a loving and dutiful son, who regularly telephoned his parents. I was also highly amused that he reverted to his boyhood accent when he spoke to them. Although he never said 'Ta-ra' to me, that was the way his conversations with his Mum and Dad always ended. Dave and I also spoke often by phone and I will never forget how his voice lifted from a rather flat 'hello' when he realized he was speaking to a friend.

Dave and I worked together for 7 years. During that time, many NTS neurons were filled by Gareth, Dan and Diana; and a number of brains have yet to be cut and processed for immunohistochemistry, so Dave's input to my research will continue for many years to come.

Dave's friends and colleagues speak of him fondly and think of him often. We miss him terribly. His impact on our science and our lives has been profound.

Ida Llewellyn-Smith
Flinders University, Adelaide, Australia

Venetia Franglen

1941–2007

I first met Venetia at UCL in 1964, when she was a PhD student and I a mere MSc student, and came to know her better as a friend and colleague when I joined the Physiology Department at King's. Her work on ion and electrolyte transport in frog skin and fetal sheep and pig skin was a development of work she had started during her PhD at Chelsea College under the supervision of SE Dicker, a leading renal physiologist, who worked on neurohormones. She also worked with Richard Durbin, an eminent gastric physiologist, on ouabain binding sites in gastric mucosa.

At King's she became deeply enmeshed in student welfare and was Sub-Dean in the Medical Faculty for 5 or 6 years. She was mother confessor to successive waves of adoring students, as a student-friendly face in the Medical Faculty.

This led to Venetia's interest in curriculum development for medical pre-clinical studies. She retired from King's and with her family, husband Geoffrey (an ex-physiologist, who ran the admissions programme at St George's Hospital Medical School) and two sons, moved to Hereford to become a tutor in biology at the Open University and then Curriculum Development Facilitator at her *alma mater*, UCL.

Living in Hereford allowed her to develop her two other great interests – her family and her Christian work. She was deeply religious, but carried this lightly. She engaged actively in the life of the Cathedral as a verger and became involved in several charities, amongst which was Cancer Experience Collaborative where she became a Research Partner.

Venetia was wonderfully friendly, kind, good natured, hospitable and generous, she devoted huge amounts of her time to others, particularly Geoffrey, who was for many years, a chronic invalid. She will be greatly missed.

Richard Naftalin
With prompts from Ana Ilundain, Department of Physiology, University of Seville, Spain.

100 years ago in *J Physiol*

On the contraction of muscle, chiefly in relation to the presence of 'receptive' substances: Part I
J N Langley. 1907, **36,** 347–384

My regular trawl of The Society's publication archives from a century ago has now arrived at December 1907's volume 36 (issues 4–5) of *The Journal of Physiology*. I was initially intrigued by a paper from A E Boycott and G C C Damant of the Lister Institute of Preventive Medicine on ... *marsh-gas, hydrogen and carbon dioxide produced in the alimentary canal of goats*, since it described an 'airtight steel pressure chamber of 9500 litres capacity'! (Boycott & Damant, 1907). However, I have decided to go for a true landmark paper from that volume. This is by John Newport Langley (1852–1925) (pictured above), then Professor of Physiology at Cambridge, and is one of a series of his papers that were key in defining the idea of a 'receptive substance' – or, as we would now say, receptors.

The concept of receptive substances, like many other early discoveries in the physiology of nervous transmission, was heavily dependent on investigations of the actions of natural alkaloids like muscarine, nicotine, pilocarpine and curare. Many of these were isolated and wholly or partially purified in the late 19th century, and they were eagerly seized on by physiologists trying to work out how they exerted their effects.

In the 1907 paper Langley reports the results of experiments applying nicotine both locally (using a fine sable artist's brush to apply drops of solution) and globally to frog skeletal muscles. One of the attractions of his work is the elegant use of simple but careful observation and subsequent logical deduction. Parts of it would make good reading for modern students as an introduction to physiological research. For instance:

There can then be no doubt that the contraction [following local nicotine application] spreads throughout the muscle fibres. The outspread might be due either to conduction in the fibres, or to diffusion of nicotine along the fibres. I take the former view for the following reasons: with the stronger solutions of nicotine the outspread of the contraction seems to be too quick for diffusion to occur throughout the segment.. [and] when nicotine is placed upon a small spot of the muscle, the lateral outspread of the movement is barely appreciable, and is such as would naturally result from a trifling outspread of the nicotine... the longitudinal outspread is, as I have said, throughout the segment (p. 351).

Langley showed in the paper that the responsiveness to nicotine was highest in the regions of the muscle innervated by nerve fibres, but that nicotine acted on the muscle – or on the muscle 'receptive substance', in his terminology – rather than the nerves. Thus the nicotine receptive substance must be concentrated in the muscle at the nerve endings – in the motor endplates, as we would now say.

Langley's ideas were not universally accepted at the time, and vigorous debate ensued over the existence and role of 'receptive substances'. Maehle has written a fascinating account of how Langley presented the data contained in the 1907 (and subsequent) papers to the 7th International Physiology Conference (the forerunner of IUPS) in Heidelberg in 1907 (Maehle, 2004). Here Langley became involved in a fierce public debate with Rudolf Magnus, which raged in the physiology journals for several years.

Although Langley is widely credited with originating the idea of receptors and of chemical transmission, it would be remiss not to mention that other commentators highlight the role of his graduate student Thomas Renton Elliott (1877-1961). As was then the custom, Elliott published separately from Langley, and in a famously prophetic Phys Soc communication from 21 May 1904 (*On the action of adrenalin*) (Elliott, 1904) he hypothesised:

Adrenalin might then be the chemical stimulant liberated on each occasion when the [sympathetic nervous] impulse arrives at the periphery.

Elliott later became a close friend of Langley's immediately previous graduate student Henry (HH) Dale (1875–1968), who wrote Elliott's Royal Society biographical memoir (Dale, 1961) and always gave Elliott much credit for originating the ideas whose pursuit ultimately lead Dale to the 1936 Nobel Prize for neurotransmission . Elliott did not pursue a career in physiological research after his PhD, but instead attended medical school, later becoming the first Professor of Medicine at UCL.

Returning to Langley, the 1907 paper appeared when he was 55; he had been in Cambridge since arriving as a 19 year old undergraduate in 1871, becoming College Fellow in 1877, FRS in 1883 and succeeding Michael Foster as Professor in 1903. Langley had also been both chief editor and indeed *owner* of *The Journal of Physiology* since 1894. Langley was an experimentalist rather than a theorist. HH Dale in his obituary of Elliott quotes Langley's credo as follows (Dale, 1961):

'*Make accurate observations and get the facts*', *he would say;* '*if you do that the theory ought to make itself*'.

Like many eminent scientists of the period Langley was amazingly productive. In his case this may have reflected a lack of distractions – he lived for much of his adult life in college, only moving out when he married at 50. Despite acquiring a wife and daughter and a keen interest in gardening, he remained scientifically active as both teacher and researcher until the end of his life. His obituarist in *J Physiol,* Sir Walter (W M) Fletcher, wrote that Langley 'became unwell ... on 31 October (1925) after giving an early lecture to a large class of students and after examining the results of a long experiment done on the previous day' (Fletcher, 1926). This, note, at the age of 73 ... ! Pneumonia followed, and Langley died at his home on5 November . He had done his first teaching at Cambridge exactly 50 years previously, in 1875, as Foster's Demonstrator in Physiology. Fletcher's obituary gives an excellent flavour of Langley the man and of Cambridge intellectual life in the late 19th century.

Unsurprisingly, much has been written about Langley, Elliott, Dale and the origins of the ideas of receptors and neurotransmission, and this quick vignette has only scratched the surface. For more detail the historical articles by Max Bennett (2000) or Tilli Tansey (1991) are excellent reading.

Austin Elliott

References

Bennett MR (2000). *Neuropharm* **19**, 523–546.

Boycott AE & Damant GCC (1907). *J Physiol* **36**, 283–287.

Dale HH (1961). *Bio Mem Fellows Roy Soc* **7**, 52–74.

Elliott TR (1904). *J Physiol* **31**, xx–xxi.

Fletcher WM (1926). *J Physiol* **61**, 4–15.

Maehle A-H (2004). *Med Hist* **48**, 153–174.

Tansey EM (1991). *Clin Autonom Res* **1**, 63–72 .

Themed Meeting

Theme
Cardiac & Respiratory Physiology

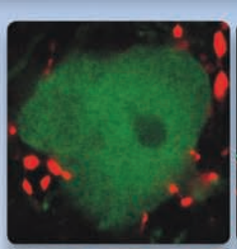
The Physiological Society

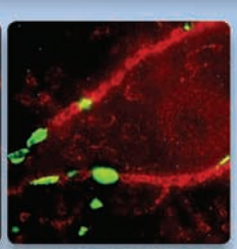
Determining Control of the Cardiovascular System in Health and Disease: From Brain to Blood Vessel

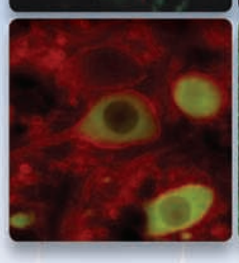
University of Leeds, UK, 17-19 March 2008

Abstract Submission and Registration	Abstract Submission	Early-Bird Registration	Registration
Opens 21 January 2008	Closes 1 February 2008	Closes 22 February 2008	Closes 10 March 2008

The Physiological Society Travel Grant Deadline is 31 January 2008







Special Lecture in memory of David Jordan

given by
Andy Ramage
(University College London, UK)

Experimental **Physiology**
Translation and Integration
Centenary **1908-2008**
Including a festschrift in honour of
Mike Spyer (University College London, UK)

Speakers

- Julian Paton
  University of Bristol, UK
- Mike Andresen
  Oregon Health & Science University, USA
- Kevin Shoemaker
  University of Western Ontario, Canada
- Michael Gilbey
  University College London, UK
- Tony Pickering
  University of Bristol, UK
- Susan Deuchars
  University of Leeds, UK
- Larry Schramm
  Johns Hopkins School of Medicine, Baltimore, USA
- David Paterson
  University of Oxford, UK
- Roger Hainsworth
  University of Leeds, UK
- Donal O'Leary
  Wayne State University, Detroit, USA
- Frederic Roche
  Université Jean Monnet, Saint Etienne, France
- Chris Matthias
  Imperial College London, UK
- Ida Llewellyn-Smith
  Flinders University, Australia
- Elspeth McLachlan
  Prince of Wales Med. Res. Inst., Sydney, Australia
- Janice Marshall
  University of Birmingham, UK
- Tom Cunnane
  University of Oxford, UK
- Sheila Gardiner
  University of Nottingham, UK
- Shaun F Morrison
  Oregon Health & Science University, USA
- John Greenwood
  Leeds General Infirmary, Leeds, UK

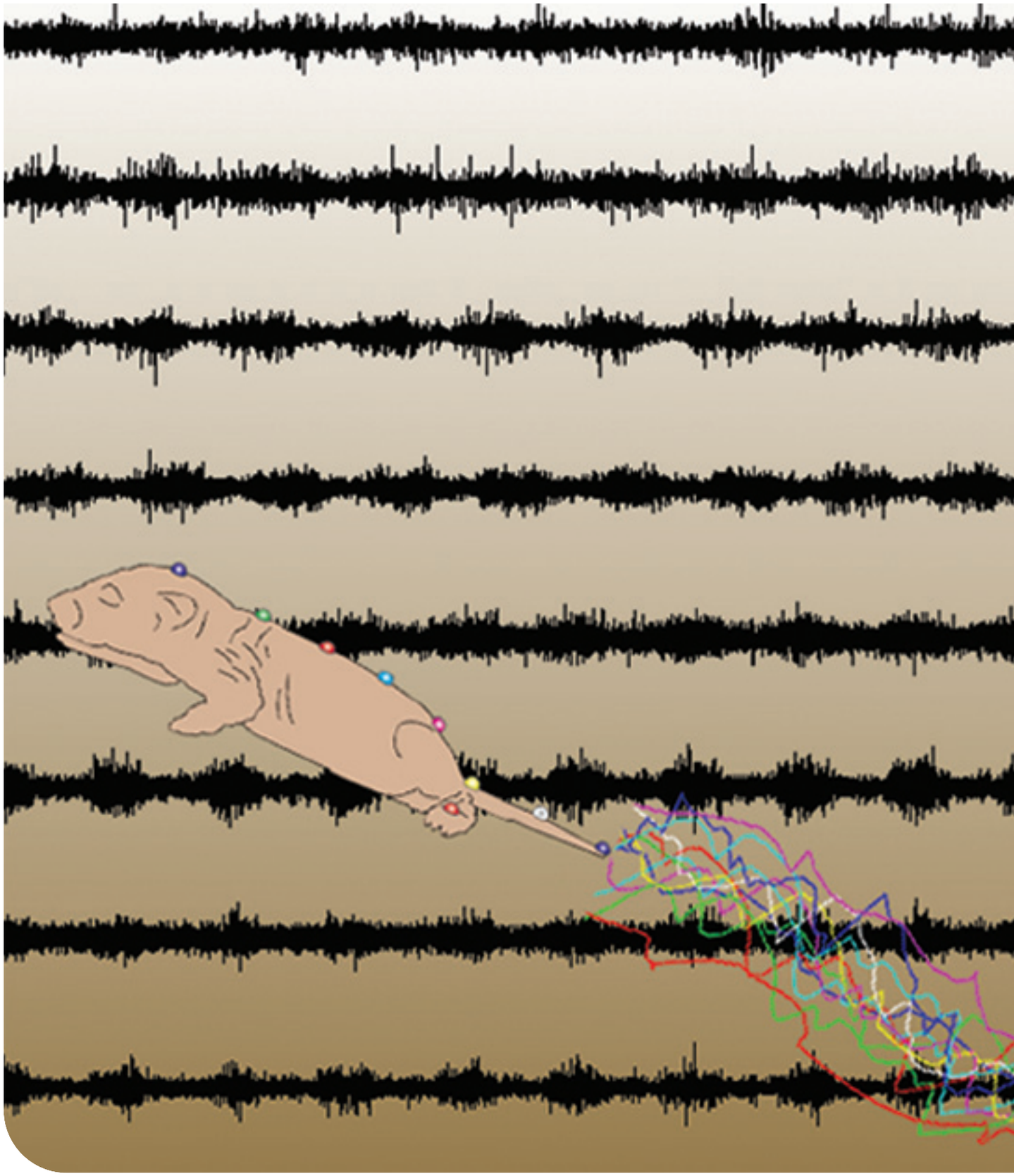
For More Information

Please visit the website, or contact

Heidi Adnum
UK Events Coordinator
t: +44 (0) 20 7269 5715
e: hadnum@physoc.org

Organisers: Jim Deuchars and Sue Deuchars (University of Leeds, UK)

http://www.physoc.org/meetings/leeds2007



Neuronal mechanisms underlying trunk bending during locomotion. Background recordings are ventral root neurograms obtained from an *in vitro* isolated neonatal rat spinal cord during fictive locomotion. Coloured lines indicate movement trajectories of the spots labelled on the rat (with corresponding colour codes) following 2D kinematic analysis of movements during an episode of locomotion (*from Falgairolle & Cazalets, p. 15*)

A publication of The Physiological Society
www.physoc.org