

THE ENDOCRINOLOGIST

THE MAGAZINE OF THE SOCIETY FOR ENDOCRINOLOGY

The magic of **MEASURING HORMONES**

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hormone actions:*

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A WORD FROM THE EDITOR...



As the spring air fast approaches, I can sense that you have been eagerly awaiting this magazine like a group of excitable Easter endocrine bunnies.

The theme for this issue is the magical way we are able to measure hormones and demonstrate how they work through laboratory techniques. It is amazing that just over 150 years ago it was first observed that reinserting the testicles of castrated chickens caused them to become more 'manly'. This gave birth to the whole concept of modern endocrinology. In the same year, a doctor called Thomas Addison described an unwell pigmented patient with diseased-looking adrenal glands on post-mortem examination.

In this fascinating issue, you will discover that we now have exquisite explanations for these early observations. We can measure tiny concentrations of hormones, are able to determine endocrine pathways using genetic techniques, and may soon have a way of directly observing cellular activity in real time.

You will read about Phil Lowry, one of our greatest living scientists who, via a series of classical experiments, discovered exactly why the patient described by Thomas Addison became pigmented. Lowry's seminal work on adrenocorticotrophin launched the concept of hormone precursors and opened the floodgates for some of our best-known clinical endocrinologists.

So, as your hormones start to burst into full bloom this springtime, I hope you enjoy this celebration of the wonders of modern endocrinology. Thanks as always to those who found the time to write for us. You will see we have introduced a new 'Letter from America', written by senior endocrinologists from across 'The Pond'. It is clear that your favourite endocrinology magazine remains in state of extremely rude health and continues to go from strength to strength.

BEST WISHES
MILES LEVY

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Editor:

Dr Miles Levy (Leicester)

Associate Editor:

Dr Tony Coll (Cambridge)

Editorial Board:

Dr Karen Featherstone (Manchester)

Dr Paul Foster (Birmingham)

Dr Paul Grant (London)

Dr Olympia Koulouri (Cambridge)

Managing Editor: **Dr Jennie Evans**

Sub-editor: **Caroline Brewster**

Design: **Third Floor Design**

Society for Endocrinology

22 Apex Court, Woodlands,

Bradley Stoke, Bristol BS32 4JT, UK

Tel: **+44 (0)1454 642200**

Email: info@endocrinology.org

Web: www.endocrinology.org

Company Limited by Guarantee

Registered in England No. 349408

Registered Office as above

Registered Charity No. 266813

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THE ENDOCRINOLOGIST ENQUIRIES

Please contact **Jennie Evans**

endocrinologist@endocrinology.org

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Please contact

advertising@endocrinology.org

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The Society welcomes news items, contributions, article suggestions and letters to the editor.

We would also like to hear your feedback on this issue of the magazine.

Deadline for news items for the Summer 2014 issue: 21 March 2014. Deadline for news items for the Autumn 2014 issue: 1 July 2014.

Society for Endocrinology

BES 201424-27 March
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**KEEPING YOU IN TOUCH AT
SOCIETY FOR ENDOCRINOLOGY BES 2014**

The Society's conference blog and Facebook page will keep you up to date with the latest news and activities at Society BES conference 2014. You can also join in the action live on Twitter using the conference hashtag #sfebes14.

Our free mobile App will also tell you everything you need to know. It is sure to complement your conference experience, with a range of features including up to the minute programme information. You will find more details at www.endocrinology.org.

**NEW
PRESIDENT
FOR THE
SOCIETY**

We're delighted to announce that Professor Sir Stephen O'Rahilly (Cambridge) will take up his Presidency of the Society for Endocrinology at the Society BES conference in Liverpool. A full interview with Steve will follow in the next issue of *The Endocrinologist*. Our huge thanks go to retiring Society President, Professor Ashley Grossman (Oxford), for all his work on behalf of the Society over the past 2 years.

**NOMINATIONS
NEEDED!**

Vacancies on the following Society committees will arise at the end of 2014. If you would like to be involved in running your Society, please consider standing for election to the Clinical, Finance, Nominations, Nurse, Programme, Public Engagement or Science committee. Full details and nomination forms can be found on the specific Committee pages at www.endocrinology.org/about/committee.html.

**NEW COMMITTEE
CHAIRS**

We are delighted to welcome the new Chair of the Society's Nurse Committee, Ms Lisa Shepherd from Birmingham Heartlands Hospital, and the new Corporate Liaison Board Chair, Dr Paul Carroll from St Thomas' Hospital, London.

We thank Mrs Nikki Kieffer and Dr John Newell-Price for their time and commitment over the last 4 years as the Chairs of these committees, as well as all the committee members who retired at the end of 2013. We are indebted to all those who give their time and expertise voluntarily.

**2014 MEDALLISTS
AND PRIZE WINNERS!**

Many Society members are world-leading endocrinologists, while others of you are at earlier stages of your careers. Our medals and prizes aim to recognise and reward excellence in endocrinology at every level.

At Society BES conference 2014, you can look forward to medal lectures from some very prestigious endocrinologists, including Bert O'Malley, Theo Visser, Mitchell Lazar, Andrew Loudon and Rob McLachlan. Last year's Dale Medal winner, Ron Evans, will also give his plenary lecture, 'Nuclear receptors, metabolism and the hunger games.'

Prizes of £500 will also be awarded to the Young Endocrinologists who have submitted the two top-scoring oral communications, and there will be additional prizes for highly commended oral and poster presentations.

So, take your seat at the Society for Endocrinology BES 2014 Awards Ceremony to celebrate all the medallists and prize winners, as well as authors receiving the Society's first ever Journal Awards.

PRIZE LECTURES 2014

We congratulate the two members who have been awarded this year's Young Endocrinologists' Prize Lectureships. They will give their lectures at 17.25 on Monday 24 March at the Society for Endocrinology BES conference in Liverpool.

The Basic Science Prize was won by Cynthia Andoniadou (London) for 'Sox2+ cells of the postnatal pituitary can differentiate into hormone-producing cells *in vivo* and have tumour-inducing potential'. The Clinical Prize winner is Channa Jayasena (London) for 'Clinical effects of kisspeptin on reproductive hormone secretion, LH pulsatility and oocyte maturation during IVF treatment'.

**SOCIETY
CALENDAR**

24-27 March 2014
**SOCIETY FOR
ENDOCRINOLOGY
BES 2014**
Liverpool

23 May 2014
**REGIONAL CLINICAL
CASES MEETING**
Dundee

15-16 September 2014
**ENDOCRINE
NURSE UPDATE**
Birmingham

October 2014
**CAREER DEVELOPMENT
WORKSHOPS**

3-5 November 2014
CLINICAL UPDATE
Manchester

see
[www.endocrinology.org/
meetings](http://www.endocrinology.org/meetings) for full details

**GRANT
AND
PRIZE
DEADLINES**

11 March 2014
SUMMER STUDENTSHIPS

15 April 2014
CONFERENCE GRANTS

27 May 2014
EARLY CAREER GRANTS

16 June-14 July 2014
**UNDERGRADUATE
ACHIEVEMENT AWARDS**

see
[www.endocrinology.org/
grants](http://www.endocrinology.org/grants) for full details of
all Society grants

HOT TOPICS

HT

SOCIETY FOR ENDOCRINOLOGY OFFICIAL JOURNALS

Society members have free access to the current content of *Journal of Endocrinology*, *Journal of Molecular Endocrinology*, *Endocrine-Related Cancer* and *Clinical Endocrinology* via www.bioscialliance.org. *Endocrine Connections* and *Endocrinology, Diabetes & Metabolism Case Reports*, the Society-endorsed case reports publication, are open access and free to all.



JOURNAL OF ENDOCRINOLOGY

Mouse model of metabolic syndrome

Patients with elevated glucocorticoid (GC) levels share many symptoms of metabolic syndrome, including hyperglycaemia, abdominal obesity, dyslipidaemia and hypertension. Conversely, patients with metabolic syndrome or glucose intolerance have elevated GC levels.

Fransson and colleagues have determined the metabolic profile of mice given corticosterone in their drinking water. This caused obesity, dyslipidaemia, fat disposition in liver and skeletal muscle and hypertension. Corticosterone-treated mice were also insulin resistant and glucose intolerant, resulting in

β -cell adaptation, whereby cell proliferation caused an increase in pancreatic volume. These effects were reversed upon cessation of corticosterone treatment.

This identifies a mouse model of metabolic syndrome where limited handling reduces the stress imposed upon the animal. This model should prove useful in studies of this condition.

Read the full article in *Journal of Endocrinology* **219** 231–241

JOURNAL OF MOLECULAR ENDOCRINOLOGY

PKC and ERK signalling mediate GH-stimulated lipolysis

Growth hormone (GH) regulates growth (an anabolic process) and lipolysis (a catabolic process). Mechanisms by which GH facilitates lipolysis and the integration of this activity with the other functions of GH are unclear.

Bergan and colleagues investigated whether GH-facilitated lipolysis is mediated by hormone-sensitive lipase (HSL), the main lipolytic enzyme in mammals and fish, and the signalling pathways involved. They found that

GH promoted HSL activation and induced HSL gene expression, via the activation of PKC and ERK signalling pathways. Interestingly JAK/STAT and PI3K/Akt signalling was inhibited during GH-mediated lipolysis.

This indicates that GH promotes lipolysis by shifting the balance between several different signalling pathways, particularly by reducing Akt signalling.

Read the full article in *Journal of Molecular Endocrinology* **51** 213–224

ENDOCRINE-RELATED CANCER

Leptin maintains cancer stem cell-like properties

Breast tumours contain cancer stem cells (CSCs), which have characteristics of normal stem cells, but may contribute to tumour formation through self-renewal and cell differentiation. CSCs are characterised by their expression of several cell surface receptors and transcription factors, including NANOG, a pro-carcinogenic factor.

Expression of the leptin receptor (LEPR) correlates with poor prognosis in breast cancer. LEPR may be involved in maintaining CSCs in a stem cell-like state. Zheng and co-workers now show that inhibition of LEPR expression

reduces NANOG expression in mammary cancer cell lines. Inhibition of LEPR also reduced CSC self-renewal and tumour outgrowth in mice.

LEPR, through its effects on NANOG, may be an effective therapeutic target. In breast cancer, this may be particularly important in tumours that are negative for known markers of tumour activity and are refractory to current clinical strategies.

Read the full article in *Endocrine-Related Cancer* **20** 797–808

ENDOCRINE HIGHLIGHTS

A summary of papers from around the endocrine community that have got you talking.

Stem cells contribute to pituitary tumour induction

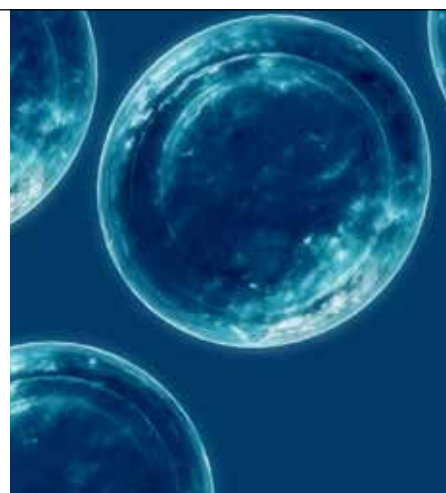
Over the past decade, increasing evidence has suggested that the pituitary contains stem cells essential not only for the development of the gland but also for tissue regeneration following cell ablation. This putative stem cell population expresses the transcription factor Sox2 and is included in the S100B-positive folliculostellate cell population.

Andoniadou and colleagues, using lineage-tracing methods, have shown that Sox2-positive cells can differentiate into the major hormone-producing cells of the pituitary, *in vivo*. Pituitary stem cells persisted in the gland in an undifferentiated state for long periods, showing that these cells contribute to pituitary gland homeostasis throughout life, rather than being transient progenitor cells.

Current hypotheses suggest that stem cells may also play a role in tumorigenesis, whereby stem cell self-renewal and differentiation contribute to tumour mass. In a mouse model of adamantinomatous craniopharyngioma, this group reports that, contrary to these hypotheses, pituitary stem cells may facilitate tumorigenesis through paracrine signalling, without directly contributing to tumour mass. These data have important implications for our understanding of the involvement of stem cells in tumorigenesis.

Read the full article in *Cell Stem Cell* **13** 433–445

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CLINICAL ENDOCRINOLOGY

UK epidemic of testosterone prescribing (2001–2010)

Suspected male hypogonadism has become a common reason for referral to endocrine services, and Gan *et al.* sought to explore this systematically.

Frequency of prescribing and cost of testosterone preparations were analysed. Over 10 years, there was a marked increase in the number of males treated (90%). This was accompanied by a 2.7-fold rise in prescription costs, reaching £11.7 million annually. Transdermal testosterone has become the most commonly prescribed form, and testosterone undecanoate is also increasingly used. All other preparations have either stayed stable or gone out of fashion.

Nevertheless, the number of people with unequivocally low testosterone (<6nmol/l) has not increased.

This raises concerns about people being treated in the absence of a robust diagnosis of hypogonadism. On the other hand, 'borderline' serum testosterone concentrations are increasingly being detected. The authors discuss pitfalls in the interpretation of testosterone levels and point out the lack of prospective data regarding long-term testosterone replacement therapy, especially in the 'borderline' hypogonadism group.

Read the full article in *Clinical Endocrinology* 79 564–570

ENDOCRINOLOGY, DIABETES & METABOLISM CASE REPORTS

Genetic testing in familial cranial diabetes insipidus

Srinivasan *et al.* describe a case of a 3-month-old baby, thought possibly to have cranial diabetes insipidus (CDI) in the context of a positive family history and parental concerns regarding polyuria and polydipsia.

Although normonatremic, his urine osmolality was low, and early stage CDI was suspected. After beginning DDAVP, he became hyponatraemic. DDAVP was discontinued and a water deprivation test performed. This showed adequate ability to concentrate urine with a urine osmolality of 563mOsm/kg

at 8h into the test and a sodium level of 136mmol/l. Sequencing the arginine vasopressin gene in the father and sister revealed the pathogenic mutation c.242T>C (p.Leu81Pro) in exon 2. This mutation was absent from the baby, and the family was reassured that there was no evidence of CDI and low future risk of developing the condition. The authors discuss the challenges of clinical/biochemical testing for CDI and advocate the use of genetic testing for selected cases.

Read the full article in *Endocrinology, Diabetes & Metabolism Case Reports* 2013 EDM130068

ENDOCRINE CONNECTIONS

FGF21 plasma levels increase in inflammatory disease

Fibroblast growth factor 21 (FGF21) regulates glucose and lipid metabolism and may have potential as a therapeutic target in conditions of insulin resistance. Produced mainly in the liver and white adipose tissue, FGF21 is released into the circulation to function as an endocrine factor.

FGF21 is elevated in inflammatory conditions in mice, and its administration can have beneficial effects in inflammatory disorders. Gariani and colleagues have extended these observations to humans by measuring serum FGF21 in

systemic inflammatory response syndrome (SIRS) and sepsis. FGF21 was elevated in both conditions with correlation to clinical state.

This suggests that FGF21 may have beneficial therapeutic effects in modulating inflammation, although the causal relationship between FGF21 and inflammatory disease, along with the potential involvement of insulin resistance, remains to be determined.

Read the full article in *Endocrine Connections* 2 146–153

Lower BAT volume in south Asian adults

Individuals of south Asian origin have a very high risk of developing type 2 diabetes compared with white Caucasians. Brown adipose tissue (BAT) is thought to have a role in energy metabolism by combusting fatty acids and glucose to produce heat and might contribute to the difference in incidence of type 2 diabetes between ethnic groups.

Bakker and co-workers measured BAT volume and activity with cold-induced ¹⁸F-FDG PET CT scans, and assessed resting energy expenditure, non-shivering thermogenesis, and serum parameters. Total BAT volume was lower in south Asians than it was in Caucasians.

Lower resting energy expenditure, non-shivering thermogenesis and BAT volumes in south Asian populations might underlie their high susceptibility to metabolic disturbances, such as obesity and type 2 diabetes. Development of strategies to increase BAT volume and activity might help prevent and treat such disorders, particularly in south Asian individuals.

Read the full article in *Lancet Diabetes and Endocrinology* doi:10.1016/S2213-8587(13)70156-6

Metabolic switch in androgen regulation of prostate cancer growth

Prostate cancer growth is driven by androgen receptor (AR) activation, by a mechanism which is poorly understood. Using emerging metabolomic techniques, Tennakoon *et al.* have shown that AR activation in prostate cancer leads to elevated glycolysis and glucose and fatty acid oxidation.

Intriguingly, this effect is regulated by the metabolic sensor protein 5'-AMP-activated protein kinase (AMPK), as its silencing blocks these metabolic changes. Furthermore, AR activation increases mitochondrial biogenesis through peroxisome proliferation-activated receptor γ co-activator 1 α (PGC-1 α) upregulation, leading to elevated intracellular ATP levels vital for cell division. PGC-1 α was found to be upregulated in a sub-population of prostate cancer clinical samples taken from patients who had clinical pathologic T stages from T2c to T3b and Gleason scores from 7 to 9 (mean 7.4).

These findings are the first to link AR activation in prostate cancer to elevations in glucose and fatty acid metabolism and AMPK activation, thus resulting in distinct growth advantages mediated through PGC-1 α . Understanding all the metabolomic pathways associated with AR signalling will lead to novel therapeutic avenues to treat prostate cancer.

Read the full article in *Oncogene* doi:10.1038/onc.2013.463

27-Hydroxycholesterol action on breast tumours

Breast cancers can be classified into several types, including oestrogen receptor (ER)-positive cancers that respond to endocrine therapy. Obesity, metabolic syndrome and, more recently, hypercholesterolaemia have been identified as risk factors for ER-positive breast cancers in postmenopausal women.

Using several mouse models of breast cancer, Nelson and colleagues investigated the effect of 27-hydroxycholesterol (27HC), a primary metabolite of cholesterol, on breast tumour growth and metastasis. 27HC increased primary breast cancer growth in an ER-dependent manner. 27HC also increased the presence of secondary metastases in the lung: an action that required liver X receptor but not ER.

Reducing cholesterol levels, or the production of metabolites such as 27HC, may have therapeutic benefit for breast cancer. Further studies are required to evaluate whether lowering cholesterol is an effective strategy for either reducing breast cancer risk or improving treatment during endocrine therapy.

Read the full article in *Science* 342 1094–1098



HOW ARE HORMONES MEASURED? A JOURNEY IN ENDOCRINOLOGY

WRITTEN BY JULIAN H BARTH



Hormones have been measured by immunoassay for more than four decades, since Berson and Yalow discovered that antibodies could be used as diagnostic agents. They were awarded a Nobel Prize and, altruistically, did not patent their findings.



Technicians at a NHS laboratory in the 1950s carry out pregnancy tests using *Xenopus* toads. Reproduced by kind permission of Audrey Peattie.

However, the story of hormone assays begins earlier, with a variety of *in vivo* biological tests. Subsequently, colourimetric assays were discovered and used to good effect, particularly for cortisol. Since 1970, immunoassays have become increasingly routine and, after a period of dominance within the clinical laboratory, it is only now after a further 40 years that a new technology, tandem mass spectrometry (TMS), is starting to emerge as the method of choice.

EARLY TESTS USING BIOLOGICAL EFFECTS OF HORMONES - BIOASSAYS

The early bioassays included the chick cockscomb test. Androgen-like material was applied to the cockscomb of a chick, and the increase in size was used as a measure of testosterone. One can only imagine the errors that might have been introduced while trying to hold the animal still in front of a lamp, whilst the shadow of the comb was outlined on a screen to measure its size.

Another qualitative test used the *Xenopus* toad, which ovulated in response to human chorionic gonadotrophin injections, and was used as an early pregnancy test. Probably more precise was the ovarian weight augmentation assay in immature rodents, used to measure follicle-stimulating hormone (FSH).

DEVELOPMENT OF MODERN IMMUNOASSAYS

Early immunoassays using antibodies from immunised animals with ¹²⁵I-labelled hormones in competitive assays were very welcome. These assays were time-consuming. In the 1970s, luteinising hormone and FSH assays took 5 days, from pipetting the samples on a Monday through to calculation and reporting of results on a Friday, but by the late 1980s a batch of 30 samples could be measured in a day.

In those days, the assays were all built in-house, and our older colleagues will remember the 'hot labs' where they iodinated their ligands, and the local university animal house where they venesected animals for their polyvalent antibodies. I remember being regaled with tales of a colleague being flung skyward at the university farm by an angry ram who was in no mood to donate his blood to science.

The antibodies were not always very precise in their detection of specific molecules, hence preparatory separation of serum samples was required, using techniques such as paper and column chromatography or organic solvent extraction to increase the specificity of the antigen available to be bound. These methods are very time-consuming and require considerable technical skill. This issue of specificity was addressed by the next milestones.

BENEFICIAL REFINEMENTS

First we saw the development of monoclonal antibody production from hybridomas (associated with another Nobel Prize). Secondly there was the development of non-isotopic labels, such as enzymes, luminescence, delayed fluorescence, polarisation fluorescence etc. Thirdly we benefited from the merging of biological with mechanical sciences as robotic instruments were developed. These instruments can pipette faster and more precisely than humans and, equally importantly, could ensure that incubations were precisely timed, so that assay numbers could be increased from 30 per day to several hundred per day without any assay drift across batches.

The increase in productivity comes with a cost. Direct assays for steroid molecules are still prone to interference from other molecules. Monoclonal antibody kits from different commercial providers are proprietary agents, and so there are variations between methods that persist even with international reference preparations. Moreover, monoclonal antibodies may not necessarily be the best tool for measuring peptide hormones that exist with many glycoforms and oligomeric forms.

IT'S ALL ABOUT SENSITIVITY: ENDOCRINE STUDIES IN MICE



WRITTEN BY PAUL LE TISSIER

The pulsatile secretion of many hormones makes single measurements of their concentration in blood a poor indicator of an animal's endocrine status. This makes studies in mice, whose small size restricts the volume of blood taken for multiple sampling, problematic. Innovations to increase the sensitivity of hormone assays, or development of models with light-emitting markers mimicking hormone secretion, may overcome this problem and make mice an even more valuable physiological model.

THE DAWN OF THE IMMUNOASSAY

Measurement of the output of endocrine glands and the concentrations of hormone in target tissue forms the basis of modern endocrinology. The development of the radioimmunoassay (RIA) by Yalow and Berson in the 1950s transformed endocrine studies, providing a technique that is sensitive, inexpensive and precise. Many readers of *The Endocrinologist* will fondly remember repeatedly adding reagents carefully to hundreds of tubes, transforming the blood samples collected over many hours into accurate, informative and illuminating hormone profiles. RIAs are still used to great effect, but in many laboratories they have been replaced with the non-radioactive enzyme-linked immunosorbent assay (ELISA), which in most cases is equally reliable and at least as sensitive.

Through the application of RIAs and ELISAs we learnt that endocrine output is dynamic and that the pattern is as important as the absolute amount of hormone in determining tissue response (for example, in the growth hormone (GH) axis^{1,2}). The sensitivity and the range of analytes have transformed our knowledge of the interactive dynamics of many endocrine axes (see Crawford *et al.*³ for one of many examples). The site of sampling has also been shown to be important; for instance, the ability to sample both hypothalamo-hypophyseal portal and peripheral blood in conscious ewes demonstrated the relationship between gonadotrophin-releasing hormone and luteinising hormone (LH) secretion.⁴

ADDRESSING ISSUES OF SAMPLE SIZE

Endocrine studies in mice, potentially powerful models through genetic modification, have been hampered by the very small quantities of blood that can be sampled, especially at the frequency required to detect the dynamic output of many hormones. The placing of catheters into specific collection sites to facilitate multiple sampling is also a technical challenge. Even highly sensitive RIAs are not capable of measuring most hormones in the small volumes of blood available, meaning that the pattern of output in many mouse models is not known, resulting in potentially misleading suppositions being made by extrapolating from data obtained in the rat or other more distant species. This issue has provided an impetus to develop new more sensitive assay methods or other ways to monitor endocrine systems.

CONTINUED ON PAGE 8...

Immunoassays have developed outside the clinical and research laboratory environment and are now used by clinical staff as near patient tests, by field toxicologists for environmental poisons, and by the lay public as pregnancy and HIV tests.

The existence of interference by molecules similar to the ones under investigation has already been mentioned but, over the years, every time an assay for a novel analyte has been produced, it has been followed by reports of antibody interference. These are usually only noted when assay results clearly diverge from the clinical picture, but our group has shown that more subtle interference occurs in at least 1:200 patient samples. The interference is quite promiscuous and not limited to a single analyte.

TANDEM MASS SPECTROMETRY - THE FUTURE?

The scaling down of mass spectrometers from the size of a double bedroom to a desk top instrument has permitted the introduction of this technology to the clinical laboratory. TMS using quadrupoles is only useful for the measurement of small molecules such as steroids, drugs and intermediary metabolites, and more sophisticated instruments are necessary for peptide hormone measurement. It is remarkable that, despite it being less than 10 years since the first reports of the use of TMS in endocrinology, *Journal of Clinical Endocrinology & Metabolism* has stated that it will only accept papers that use TMS methods for steroid analysis in future.

TMS is presently a sophisticated technique that needs skilled staff, but it will undoubtedly become a more friendly technique in the future. After all, mass spectrometry is used in airport security for the detection of volatile explosives, so it is only time before this comes to routine clinical laboratories. Inevitably, as TMS is more widely used, newer types of analytical problem will be found. We are already aware of interference by substances that co-elute in the preparatory columns and by epimers and structural isomers. So watch this space...

Endocrinology has developed in tandem with immunoassays, and both sciences have progressed in leaps and bounds since the seminal work of Berson and Yalow in 1959. The next game changer will be the analysis of hormones in real time by the patient.

JULIAN H BARTH

Consultant Chemical Pathologist, University of Leeds

Julian Barth is Director of the SAS Steroid Centre. He provides a clinical service for patients with porphyria and morbid obesity.

READER REQUEST

Jesse Olszynko-Gryn (Cambridge) is studying the history of pregnancy testing and would like to hear from readers about their memories of early laboratory protocols. If you have any stories to share, please contact jo312@cam.ac.uk.

...CONTINUED FROM PAGE 7

The sensitivities of some ELISAs have been increased by careful modification and are able to measure pulsatile hormone secretion in a 2µl blood sample for both mouse GH (sensitivity 0.9ng/ml⁵) and LH (sensitivity 117pg/ml⁶). However, there are limits to how far this technology can be adapted. The immunoassay technique developed by the Luminex Corporation⁷ or mass spectroscopy enables the sensitive measurement of hormone, but both require specialised and costly equipment, precluding their routine use in many laboratories. With Dave Grattan (University of Otago, New Zealand), Mike White (University of Manchester) and others, we have sought alternative novel methods to address these problems based on either immunoassay with electrochemical sensors or detection of proteins which are co-secreted with hormones.

'Measurement of the output of endocrine glands and the concentrations of hormone in target tissue forms the basis of modern endocrinology.'

USE OF ELECTROCHEMICAL SENSORS

The electrochemical sensors we are using are based on disposable screen printed carbon and gold electrodes. Immobilised antibodies on the electrode surface selectively bind hormone which can be measured using inexpensive portable potentiostats. This technique has extremely high sensitivity (25pg/ml for GH⁸), which in part results from limited sample dilution, since as little as 2µl of analyte is placed on the small surface area of the electrode. This should allow measurement of multiple hormones from a single 2µl sample, especially as there is scope to increase the sensitivity of these systems even further. Measurement on the electrochemical sensors can be made within 40 minutes of taking a sample, meaning that samples can be analysed in the course of an experiment, potentially reducing the number of experimental animals required. This methodology is very much in its infancy and technical advancements in both the development of ways to reversibly bind hormone and microfluidics may allow continuous monitoring of hormone output in closed loop catheter systems.

OBSERVING CO-SECRETED PROTEINS

Our alternative approach is to use light output of fluorescent or luminescent proteins which are co-packaged and secreted with hormones. In perfused tissue slices from transgenic mice with mRFP (a fluorescent protein) targeted to prolactin granules, we were able to show that fluorescent protein output mimicked that of prolactin and could be measured with a much higher time resolution.⁹ Obviously, extending this to monitoring fluorescent output with optic fibres *in vivo* is an attractive proposition, but secreted luciferases may be an alternative with even higher sensitivity; we are currently attempting to develop this with Mike White's group in Manchester. If this is successful, continuous *in vivo* monitoring of hormone output from many endocrine systems should be possible, with the additional prospect of achieving this with high spatial resolution, allowing the equivalent of hypothalamo-hypophyseal portal sampling in multiple locations.

'Through the application of RIAs and ELISAs we learnt that endocrine output is dynamic and that the pattern is as important as the absolute amount of hormone in determining tissue response.'

Both the modification of established methods and the development of new techniques should make the mouse a more useful model for endocrine studies. Endocrinologists are best placed to drive these developments, in multi-disciplinary collaborations, as we can better identify the needs and potential pitfalls. The reward will be the ability to utilise the power of mouse genetics to augment the understanding provided in the last 60 years by RIA and/or ELISA of samples from the classical models of the rat and the sheep.

PAUL LE TISSIER
University of Manchester

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MAKING YOUR CHOICE: USING CELL AND TISSUE CULTURE TECHNIQUES

WRITTEN BY KAREN FEATHERSTONE AND JULIAN DAVIS



How often do we, as research scientists, think about the models that we use before heading into the lab to perform our favourite assay on our favourite cell line? Certainly it's far easier to think about how to modify and improve an assay than it is to think about moving from a cell line to something more akin to a whole body system. But, as endocrinologists, perhaps it's time we started to think of (and study) our cells as the integrated systems that they are, responding to hormonal cues from numerous different tissues and organs.

We use a variety of culture systems in our lab to study pituitary physiology. Each system has advantages and disadvantages that dictate the nature of the experimental investigations that can be performed.

CELL LINES

The cell line is the workhorse of life sciences research. Immortalised by one of several different methods, these cells are easy to maintain in culture and continuously replicate, providing a ready source of homogeneous cells for study. They can be very useful for looking at interactions between molecules (such as protein-protein and protein-DNA interactions), defining mechanisms of cellular function, and also for assays that require large cell numbers that may not be available from other culture models. Cell lines are also ethically important, as they allow us to minimise the number of animals used to address research questions.

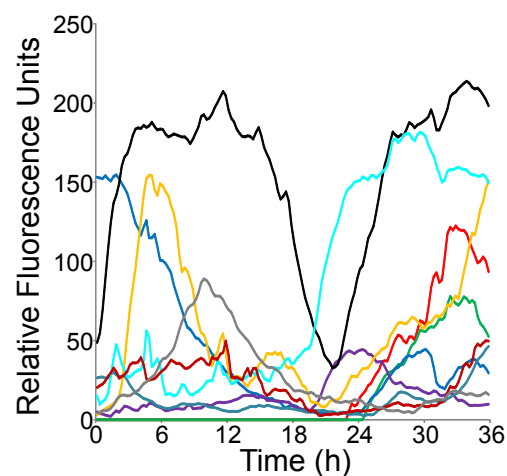
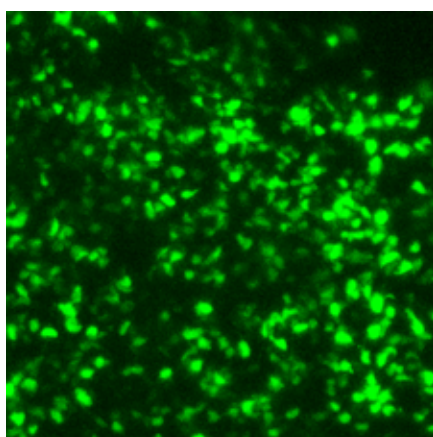
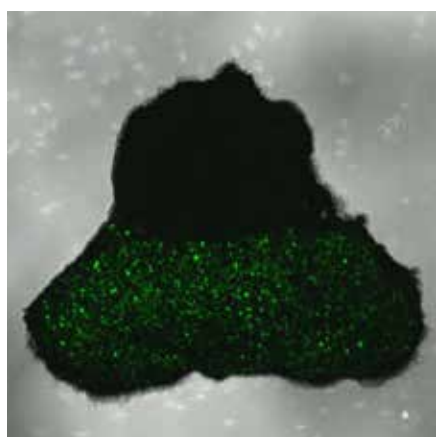
However, the behaviour of these cells may not always represent the activity of their physiological counterparts, with cell lines often misexpressing proteins and receptors, which are known regulators of that cell type *in vivo*.

PRIMARY CELL CULTURES

Primary cultures are a closer representation of the 'normal' state, often expressing receptors and proteins that are missing from cell lines. Cells are collected from the relevant organ taken from the animal of interest and maintained in culture. The collection process can be a fairly simple purification of cells from, for example, blood or bone marrow, or may involve enzymatic disaggregation of tissue.

Primary cells are more difficult to maintain in culture than cell lines and have a limited lifespan, with cells becoming less physiologically normal with longer culture periods. Unlike cell line cultures, primary cultures do not represent a clonal population of cells. This will affect most assays with results representing an averaging of the cell phenotype, unless experiments are performed that either measure the response of a single cell, or techniques to synchronise cell activity across the population are employed.

CONTINUED ON PAGE 10...



A GFP reporter expressed under the control of the human prolactin gene reveals the restriction of lactotroph cells to the anterior pituitary (low magnification view, left) and the organisation of the cells within the tissue (high magnification view, centre). Measurement of fluorescence levels over time reveals transcriptional heterogeneity in individual lactotroph cells. Pituitary tissue was taken from a 1-day-old neonatal rat.

...CONTINUED FROM PAGE 9

TISSUE EXPLANT CULTURES

Tissue explant cultures have several advantages over primary cultures. First, the tissue is minimally processed before being placed in culture. In our own work, for example, pituitary tissue is prepared as 300µm sections that are cultured on filter stages, enabling exposure of the tissue to both culture medium and air. Thus, cells are likely to express receptors and respond to stimuli as they would *in vivo*.

Secondly, the cells are maintained in their native tissue environment, with cellular interactions and interactions with the extracellular matrix at least partly maintained. This can be a key advantage when studying cells that form 3D structures *in vivo*, such as pancreatic islets where homotypic and heterotypic contacts between β cells and other endocrine cells are required for robust insulin secretion. In the pituitary, paracrine signalling is important in regulating endocrine cell behaviour. More recently, it has been suggested that the cells are also organised into networks to co-ordinate cellular activity and potentially facilitate simultaneous hormone secretion, generating pulsatile hormone release, which is characteristic of many hormone axes.

We are currently using a reporter system in which destabilised green fluorescent protein (d2EGFP) expression in cells reflects the activity of the human prolactin gene (see Figure). Our analysis of lactotroph cells in intact pituitary tissue suggests that there may be local co-ordination of transcription activity between these cells; hence local communication may have an influence on cell function and hormone secretion patterns.

Limitations of tissue explant cultures include the inability to easily manipulate the activity of the cells, with pharmacological intervention generally providing the easiest method. In contrast, genetic manipulation of cells in tissue is much more problematic, due to issues such as cell death and the potential to generate a mixed population of cells due to incomplete uptake of the introduced material. Cell death is also an issue during explant culture as tissue is unlikely to remain viable and physiologically representative for long culture periods, although some groups have been able to study explant tissue in culture for many days with maintenance of clear circadian rhythms. Finally, removal of the tissue from the animal clearly disrupts the normal external signalling cues that affect cellular activity.

REAL TIME *IN VIVO* IMAGING OF CELLULAR ACTIVITY

Monitoring cellular activity, *in vivo*, with minimal disruption, is the most informative approach to studying mammalian physiology and pathology. However, technical and ethical considerations make this a challenge. Currently, *in vivo* imaging systems exist and can monitor, for example, tumour growth. Advances with fibre-optic probes are likely to enable us to image deep inside tissues at cellular resolution. In the future, technical advances will probably enable a greater understanding of the phenotype, genetic and epigenetic make up of individual cells in real life.

KAREN FEATHERSTONE AND JULIAN DAVIS

Centre for Endocrinology and Diabetes, Manchester University

FURTHER READING

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LESSONS FROM THE ‘DIABETES HORMONE’: INSULIN’S TRIUMPHS AND CHALLENGES

WRITTEN BY ROBERT SEMPLE AND DAVID CHURCH

There are signs of cultural drift progressively separating the clinical practice of diabetology and that of endocrinology.

The former increasingly focuses on systems of care and on the ‘algorithmisation’ of the results of mega-trials, while insulin itself, the misfiring hormone at the root of diabetes, is relatively rarely measured. In contrast, endocrinology remains more firmly rooted in serial measurement of hormones.

In the face of this practical schism, it is timely to remember the long and prize-bedecked history of insulin research, scattered with landmark moments in endocrine science which have guided many key developments in endocrinology.

THE QUINTESSENTIAL HORMONE

Within 17 years of the coining of the word ‘hormone’ by Ernest Starling and others over dinner in Cambridge, Banting and Best demonstrated the apparent cure of the rapidly fatal wasting disease diabetes mellitus by administration of purified canine pancreatic extract. A Nobel Prize rapidly followed in 1923, and diabetes treatment for the next half century relied on mass-produced, animal-derived insulins. In 1958, Fred Sanger was awarded the first of his two Nobel Prizes for elucidating the amino acid sequence of insulin, while Dorothy Hodgkin described its crystal structure by applying her Nobel Prize-winning technique of X ray crystallography in 1969.

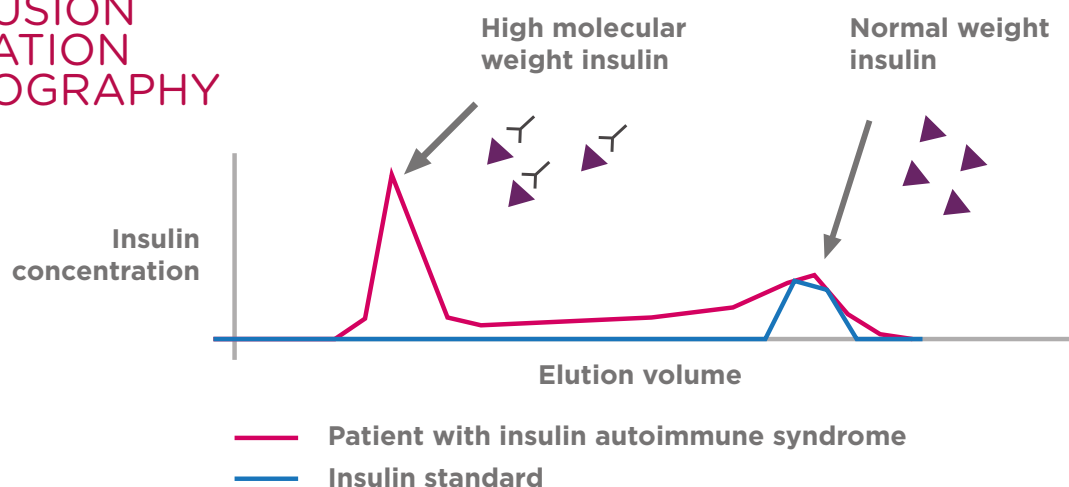
These findings, together with the advent of recombinant DNA technology, were key steps in permitting bacterial synthesis of human insulin from the 1970s, with different pharmacokinetics of therapeutic preparations generated by use of adjuncts such as protamine and zinc. The most recent phase in development of therapeutic insulins has involved genetic engineering of the insulin molecule itself, to produce analogues with altered crystalline properties, or with the ability to bind to circulating proteins. These developments have collectively yielded a flexible armamentarium of drugs with which to model endogenous insulin secretion.

ASSAYS: CHALLENGES OLD AND NEW

For nearly 40 years after its purification and first therapeutic use, determination of insulin concentration in blood was either impossible, or based on laborious bioassay of its blood glucose-lowering efficacy in model animals. However, in 1960, Berson and Yalow reported the technique of radioimmunoassay, harnessing the immense combinatorial power of the immune system, and the exquisitely sensitive detection of minute amounts of radioisotopic tracer, and applied it to biochemical detection of insulin, yielding yet another insulin-related Nobel Prize in 1977.

Immunoassays of different types remain the workhorses of endocrine laboratories to this day. However, even this powerful approach has limitations, which may have their origins either in man’s creativity in using genetically – and thus antigenically – modified insulin analogues, or in the endogenous immune response.

SIZE EXCLUSION GEL FILTRATION CHROMATOGRAPHY



ENCOUNTERING ANALOGUES

As many patients with diabetes have exogenous insulin or analogues in their circulation at the time of blood sampling, it is critical to understand the ability of the assay used to detect each species present. Insulin analogues vary in structure, with changes from native insulin ranging from alteration of a single amino acid to covalent attachment of a large hydrophobic side chain. This means that the antibodies on which immunoassays are based may detect none, or only some, of these analogues. Moreover analogues such as Levemir®, which binds to albumin in plasma via a fatty acid moiety, may exhibit a mismatch between total insulin and bioavailable insulin.

These considerations are important in biochemical evaluation of insulin-treated patients with severe insulin resistance, and in cases of possible wilful insulin misadministration. In such cases, close liaison with the laboratory is essential, with knowledge required of all insulin and analogues administered in the previous 2–3 days, along with understanding of the performance of the assay used.

Differential ability to detect different insulins is not always a problem, however. Detection of high insulin concentrations with an assay known to detect insulin analogues, with a correspondingly low level picked up by an assay selective for native insulin, is strongly indicative of exogenous analogue insulin in the circulation. This can aid clinical decision making, especially in cases of hypoglycaemia due to suspected insulin self-administration or poisoning.

ISSUES WITH ANTI-INSULIN ANTIBODIES

The immune system may also wreak havoc with the biochemical detection of insulin, either through heterophilic antibodies with sufficient affinity for capture and detection, antibodies to cause assay interference, or through appearance of high affinity anti-insulin antibodies. Anti-insulin antibodies may be measured by immunoassay; however, they are relatively common with exogenous insulin treatment, are generally of low titre and usually have no clinical importance. High affinity antibodies can sometimes form insulin-antibody 'macroinsulin' complexes in the circulation, however, leading to post-prandial hyperglycaemia and fasting hypoglycaemia. This is dubbed insulin autoimmune syndrome, or, in the case of patients naive to injected insulin, Hirata disease. The analytic challenge is to discriminate between incidental antibodies of no practical significance, and those that produce clinically significant perturbation of insulin pharmacokinetics.

A low C-peptide:insulin ratio is consistent with macroinsulin complexes in non-insulin-treated patients, but is also seen with occult insulin administration or in the presence of antibodies to the insulin receptor. An apparent increase in insulin concentrations after plasma dilution is also suggestive, and using polyethylene glycol precipitation to clear macroinsulin before re-assay of supernatant to assess recovery also has value. Insulin-antibody complexes can best be demonstrated by gel filtration chromatography, which separates insulin species according to size. The presence of complexes is demonstrated by a peak of high molecular weight insulin immunoreactivity in addition to the normal weight insulin peak (see Figure).

STILL LEADING THE WAY

Many of these immunoassay problems are common to many other tests used in endocrine practice. In future, detection of insulin using mass spectrometry may be routine, both removing possible interferences in immunoassay and allowing detection of individual analogues. However, producing quantitative mass spectroscopy-based peptide assays is technically challenging, and access to such assays in the routine hospital laboratory is currently very limited.

The measurement of hormones, historically led by the paradigm of insulin immunoassay, has shaped modern endocrine practice. At present, the uncertain nature of the perturbation of insulin action in pandemic disease, the widespread use of genetically modified insulin analogues and the wilful misuse of insulin all pose new challenges for the endocrine biochemist, and mining of the scientifically fertile seam of the 'diabetes hormone' in addressing these may yet have lessons for all of endocrinology.

ROBERT SEMPLE

*Wellcome Trust Senior Research Fellow in Clinical Science,
University of Cambridge Metabolic Research Laboratories*

DAVID CHURCH

*DRWF Sutherland-Earl Clinical Research Fellow,
University of Cambridge Metabolic Research Laboratories*



DEMONSTRATING HORMONE ACTIONS: A KNOCKOUT APPROACH



WRITTEN BY MALCOLM PARKER

Hormones control a vast array of developmental and physiological responses, and many clinical disorders result from defects in their actions.

A BLUNT INSTRUMENT

Early studies of hormone action relied on surgical endocrine ablation followed by an examination of the physiological changes, which would be expected to reflect functions of the hormone. For example, some insights into the action of oestrogens and androgens have been obtained by removal of the ovaries or testes but, even in these cases, the approach is limited, since removal of the gonads precludes studies of many aspects of reproduction.

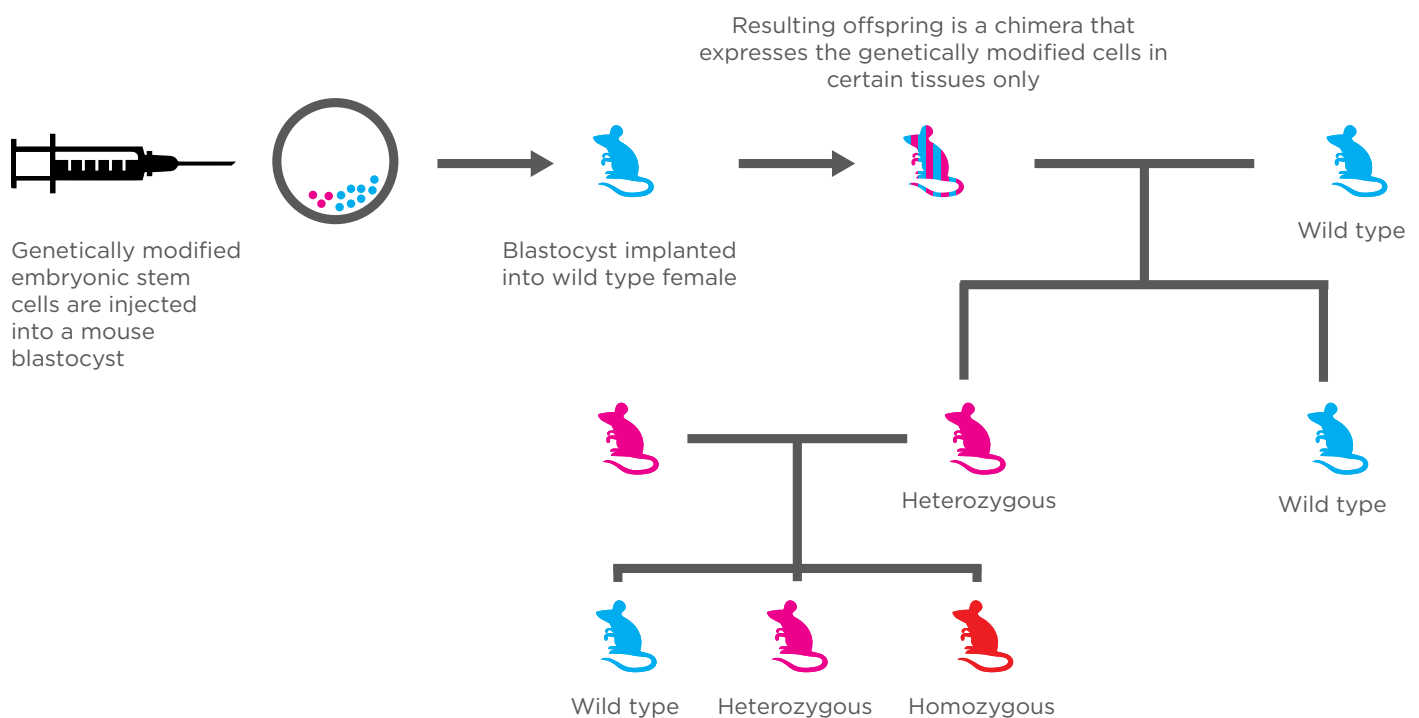
Moreover, endocrine ablation is only feasible if the source of hormone is known, if the endocrine gland or tissue is not responsible for the generation of multiple hormones, and providing its removal does not lead to widespread biological defects that in some cases may be life-threatening. Strictly speaking, demonstrating the action of a hormone depends on the

rescue of the normal biological function following endocrine ablation. Hormone replenishment, however, is often difficult to administer in a fashion that resembles normal hormone release, which may be transient or rhythmic. It is further complicated by the fact that many hormones and signalling molecules are often secreted by the same endocrine gland or tissue, and the total number may be uncertain.

A KNOCKOUT VICTORY?

So is there an alternative strategy? The ability to 'knock out' genes in embryonic cells allows the generation of homozygous null mutant mice which can be examined for the effects of a complete lack of a particular gene product. Many hormone gene knockouts have been generated in this way and the action of the hormone inferred from the phenotype of the resultant mice.

HOW TO PRODUCE A HOMOZYGOUS MOUSE



Hormone knockouts are not universally applicable, however, since small fat soluble hormones are not encoded by genes, and certain peptide hormones are generated from a single precursor protein that gives rise to multiple hormones (e.g. pro-opiomelanocortin). Making a gene knockout model is also complicated in instances where the hormone is encoded by genes that have undergone duplication, such as the mouse insulin gene, although it is possible to generate double or even triple knockouts by crossing homozygous null mice to overcome this problem.

RECEPTORS IN THE FIRING LINE

Since all hormones exert their action by binding a specific receptor, an alternative and often advantageous option is to delete the gene for the receptor rather than for the hormone. It turns out that it is often easier to identify genes that encode receptors than those for the corresponding hormones, because receptors frequently comprise families of proteins encoded by evolutionarily conserved genes. By searching for conserved DNA sequences in the human or mouse genome it is possible to identify all the potential receptor genes in a family.

‘...an alternative and often advantageous option is to delete the gene for the receptor rather than for the hormone.’

In this way, it has been discovered that as much as 10% of the human genome, more than 2,000 genes, encodes receptors for hormones and growth factors. The hormones for some of these are not known and they are referred to as orphan receptors. This approach has enabled the identification of 48 nuclear receptor genes and over 800 G-protein coupled receptors (GPCRs). All nuclear receptors and many GPCRs have been knocked out in mice and their phenotypes examined, allowing their action to be inferred, irrespective of whether their hormone has been identified.

Accordingly, complete receptor gene knockouts in mice have been extremely informative, providing insights into many endocrine pathways in humans. They have also been useful in studying the effect of gene dosage by investigating heterozygous mice.

REFINEMENTS

Nevertheless, the deletion of a gene in every type of cell or tissue may lead to complex or secondary effects, the phenotypic analysis of which is far from straightforward, and which in some cases may even be lethal (e.g. the glucocorticoid receptor). Fortunately, these problems can often be overcome by generating conditional knockouts in which the gene of interest is deleted in a specific tissue or at a specific time.

Instead of knocking out the gene in embryonic stem cells prior to generating mice, it is possible to genetically engineer mice with a normal phenotype that are then bred to produce a conditional knockout. Short DNA sequences, called loxP sites, can be inserted around a gene of interest to generate so-called ‘floxed mice’. These mice have a normal

phenotype but can be crossed with transgenic mice expressing an enzyme, Cre recombinase, that deletes the DNA between the loxP sites, thereby generating offspring that lack the gene of interest.

...IN PLACE

To knock out a gene in a specific tissue, the ‘floxed mice’ are crossed with transgenic mice that express Cre recombinase only in that tissue. This is possible by controlling the expression of the enzyme with a cell-specific promoter. For example, to delete a gene in the liver, the albumin promoter can be used to drive Cre recombinase expression only in the liver. Phenotypic examination of such mice then enables the action of the hormone or receptor in a specific tissue to be investigated.

...AND TIME

An alternative approach is to use an inducible system that allows the deletion of a gene at a specific time. In this approach, the Cre recombinase is controlled using a switch that can be off or on. Commonly, the Cre recombinase is fused to the hormone-binding domain of the oestrogen receptor, which inactivates it, but it can be switched on by tamoxifen administration. It is also possible to control Cre recombinase expression by regulatory DNA sequences, often the Tet operator, which can be switched on with tetracycline. Irrespective of the precise approach, the action of the hormone or receptor can be inferred from the phenotype of the mice.

‘...as much as 10% of the human genome, more than 2,000 genes, encodes receptors for hormones and growth factors.’

Finally, it is possible not only to knock genes out but also to generate mice with mutations in genes. Thus mutations identified in patients with endocrine disorders can be introduced into mice using the Cre-loxP technology. These mutant mice can then be used to investigate the physiological basis of the disorder, and to explore potential therapeutic approaches prior to human studies. Thus in the past 20 years there has been an exponential increase in the number of genetically modified mice that have provided valuable insights into the action of hormones and their receptors in physiology and endocrinological disorders.

MALCOLM PARKER

Professor of Molecular Endocrinology, Imperial College London

Malcolm Parker's research focuses on the function of nuclear receptor signalling in cancer, metabolism and reproduction. He obtained his PhD at the University of Leicester, and after postdoctoral research at Baylor College of Medicine, Houston, TX, USA, and the Institute of Animal Physiology in Cambridge, he was appointed group leader at the Imperial Cancer Research Fund in London. In 2000 he was appointed Head of the Institute of Reproductive and Developmental Biology at Imperial College London.

CABERGOLINE AND THE HEART: WHAT'S AN ENDOCRINOLOGIST TO DO?

WRITTEN BY MARK E MOLITCH



This is the first of a new series of musings from over the pond by senior US endocrinologists. We are pleased to introduce Mark Molitch as our inaugural trans-Atlantic correspondent.



ADVICE FOR HEALTHCARE PROFESSIONALS

Cabergoline and bromocriptine – exclude cardiac valvulopathy as determined by echocardiography before treatment. Cabergoline – echocardiography should be done within 3–6 months of starting treatment and subsequently at 6–12 monthly intervals¹

MHRA (DRUG SAFETY UPDATE, OCTOBER 2008)

In addition to the above advice from the Medicines and Healthcare Products Regulatory Agency (MHRA), the US Food and Drug Administration (FDA) has inserted similar information in the official package insert for cabergoline. So, what is an endocrinologist supposed to do?

We have these 'official' recommendations on both sides of 'The Pond' – and then we have reality. Should we be doing echocardiograms before we start treatment in all our patients with hyperprolactinaemia? Should we be doing echocardiograms every 3–12 months to monitor them?

In this short space I will try to shed some light on these issues and say what I do. I will preface this by saying I have not sought legal consultation in this regard. This may have less importance in the UK, but every morning as I dress I observe numerous commercials on CNN by lawyers asking potential 'victims' of malpractice to call their toll-free number.

The issue of cardiac valve disease and cabergoline derives from findings that high doses of cabergoline (3–6mg/day) in Parkinson's disease result in a three- to sixfold increased risk of valvular abnormalities². Leaflet and chordae thickening and stiffening are found with incomplete valvular closing and regurgitation³ and this is due to fibroblast proliferation with deposition of a plaque-like process on the valve leaflet surfaces that may also encase the chordae tendinae.^{3,4} Cabergoline and pergolide (bromocriptine to a much lesser extent) stimulate serotonin 2B receptors, resulting in activation of several mitogenic pathways, ultimately causing this overgrowth valve disorder.⁴

'We have these 'official' recommendations on both sides of 'The Pond' – and then we have reality'

Echocardiographic studies of hyperprolactinaemic patients have now been reported. However, one problem in evaluating such patients is that 'normal' individuals have pretty high rates of valvular abnormalities as assessed by echocardiography. For example, in the Framingham Heart Study of 3,589 normal middle-aged people, mitral, tricuspid or aortic regurgitation of mild or greater severity was seen in 19, 17 and 11% respectively.⁵ In the CARDIA study of 4,352 people aged 21–35 years, mitral or aortic regurgitation was found in 10.5 and 0.8% respectively.⁶

A review of the literature prior to 2010 found that most reports showed no increased risk of cardiac valve lesions detected by echocardiography in patients with hyperprolactinaemia taking conventional doses (≤ 2 mg/week) of cabergoline.⁷ A number of additional, relatively small studies have since confirmed this, although there seems to be a feeling that larger cumulative doses may be associated with some findings of uncertain clinical importance, such as valvular tenting area and calcification.^{8–15} Recently, Drake *et al.* conducted a study via the Society for Endocrinology in which they analysed data from 747 patients from 28 centres, finding no increase in the relative risk of significant valvular abnormalities. However, their comparison was the highest to the lowest quartile of cabergoline cumulative dose over approximately 5 years, and not treated patients to matched, untreated controls.¹⁶ In addition, two, large population-based studies, one with 2,381 hyperprolactinaemic patients from Denmark¹⁷ and the other with 21,039 hyperprolactinaemic patients from the UK, the Netherlands and Italy¹⁸ did not show significant increases in risk of cardiac valve disease in patients taking cabergoline.

On the basis of the above information, much of it acquired since 2008, it appears that the MHRA's 2008 recommendations are simply out-of-date and should be changed. As has been pointed out by Sherlock *et al.*,¹⁹ given the prevalence of prolactinomas in the population of between 100 and 775 per million, following the MHRA recommendations would result in 94,000 extra echocardiograms each year in the UK alone.

But...? Virtually all of these data were obtained in patients taking conventional doses of cabergoline, i.e. ≤ 2 mg/week, and the Parkinson's disease data showed a very significant risk with high doses, such as 3–5mg/day. Where is the dosing point for cabergoline for which the risk for valvular lesions begins? This is a real issue, as cabergoline was found to be effective in normalising prolactin levels in doses of ≤ 2 mg/week in only 78.7% of 150 patients by Ono *et al.*²⁰ and 83% of 122 patients by Delgrange *et al.*²¹ Ono *et al.* were then successful in normalising prolactin levels in all but one of these resistant patients with doses of cabergoline > 2 mg/week and, in some, the dose had to be increased to up to 12mg/week.²¹ These doses approximate to those used in Parkinson's patients.

Here is my approach to this conundrum. I mention to all patients with hyperprolactinaemia whom I start or continue on cabergoline that an increased risk of cardiac valve disease has been reported, but primarily in patients taking high doses for Parkinson's disease. I only do echocardiograms before and during treatment if the patient has a history of some sort of heart disease, I hear a heart murmur, or if the patient is particularly concerned, as long as the dose is kept to $\leq 2\text{mg/week}$. However, because I do not know the dosing point at which valvular risk begins to increase, in all patients requiring doses greater than 2mg/week , I ask them to do echocardiograms on a yearly basis. In those who have a trace of tricuspid or other valve regurgitation on the initial study, then the first repeat study is done at 6 months.

I believe that in this way we will be able to detect a developing lesion early in those actually susceptible to such lesions and avoid unnecessary, expensive studies in the great majority of patients we see. We now also know that, of those who do develop lesions, when the cabergoline is stopped the lesions regress in about one-half and do not progress further in the remainder.²² Therefore, I think this is a safe and cost-effective approach to this problem.

MARK E MOLITCH

Martha Leland Sherwin Professor of Endocrinology,
Northwestern University Feinberg School of Medicine,
Chicago, IL, USA

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FROM THE CHIEF EXECUTIVE'S DESK: MEASUREMENT BEYOND MATHS



WRITTEN BY LEON HEWARD-MILLS

Measuring the impact of the work we do through the Society to achieve our charitable objectives is quite a challenge.

The Society's objective is the advancement of public education in endocrinology. This is interpreted widely of course, but principally through our stated aims:

- to advance scientific and clinical education and research in endocrinology
- to encourage the best individuals into the discipline and, critically,
- to ensure a wider understanding of endocrinology

All of the above should serve to demonstrate public benefit.

Measuring this is not easy but it is important. This is not only to satisfy the Charity Commission, but also to fulfil our responsibility to our membership and the public. Of course, the financial measures are clear (e.g. the number and value of grants awarded and the financial health of the charity). Less obvious or straightforward is measurement of the public benefit directly attributable to our activities. As resources become scarcer and the call on members' time ever greater, it is more important than ever to show clearly the real impact of everything we do.

By doing this I don't mean creating a bean counters' paradise with innumerable stats and 'key performance indicator' metrics. Unnecessary measures and bureaucracy threaten to stifle the creativity and good humour (for the most part!) with which our members practise.

Some stats are essential of course – the Society's Science and Clinical Committees both measure outcomes of research activity and require detailed reports on the benefits derived from the grants awarded. But it is combined with more qualitative measures – such as practical case studies – that I believe we can demonstrate our value to the public most effectively.

I was struck by a recent article in the Addison's Disease Self Help Group newsletter (www.addisons.org.uk), describing one young person's experience of dealing with the condition. Sondra Gorick's article, *My most memorable patient*, in the Winter 2013/14 issue of this magazine (issue 110, page 22) was similarly resonant. And I was equally inspired by last year's 25-year anniversary edition of our own *Journal of Molecular Endocrinology*, effectively telling the story of the evolution of this strand of the profession. With anniversaries coming up over the next few years of the discovery of leptin and ghrelin, there is plenty more to celebrate, showing in straightforward ways the positive effect that members of the Society have had, and continue to have, on improving public health and patient outcomes.

So yes, let's keep meaningful statistical measures of progress, but as we look forward to the Society for Endocrinology BES conference in Liverpool and the many examples of clinical and research excellence that will be discussed and presented there, let's not discount the softer measures, which will help us continue our mission to advance public education regarding our discipline.

LEON HEWARD-MILLS

Chief Executive, Society for Endocrinology
and Managing Director, Bioscientifica Ltd
Email: leon.heward-mills@endocrinology.org



SCIENTIFIC CYCLES

FROM OUR SCIENCE COMMITTEE CORRESPONDENT

'The gross and net result of it is that people who spent most of their natural lives riding iron bicycles over the rocky roadsteads of this parish get their personalities mixed up with the personalities of their bicycle as a result of the interchanging of the atoms of each of them and you would be surprised at the number of people in these parts who nearly are half people and half bicycles.'

FLANN O'BRIEN (FROM THE THIRD POLICEMAN, 1967)

Howe's Cycle Shop in Cambridge has closed. After being handed down through generations of the same family for the last 173 years, the present owners have decided to call it a day. I know the Pythons are back in action and will be trying once again to mock those who repair bicycles for a living, but this is no laughing matter.

First, this bike shop was so old it opened before bikes had even been invented, the initial shop being set up to make wheels for carriages. Secondly, local legend has it that this was where Charles Darwin went to get his tyres when he was at Christ's. Thirdly, and most importantly, it was where I went for all things cyclical. Once you find a shop that suits, you will be prepared to wheel a broken derailleur past any number of seemingly appropriately equipped premises to get back to where you know things will be OK.

It's amazing how a good bike becomes an essential part of one's self. The morning ride affords ideal thinking time on the way in, and the evening ride is a handy diffuser of stress on the way home. It has also brought an appreciation of seasonality and climate that my pedestrian self never had. I suppose weather did exist in years BC ('before cycle') but I am now acutely aware of wind direction and speed, percentage chance of rain, ground temperature and relative humidity.

In a flat town with clogged roads, bikes matter. So much so that we even have DNA incorporated into the cycle path that runs south from the biomedical campus. Marked by a sculpture of the DNA double helix at either end, a series of blue, green, red and yellow stripes represent the 10,257 nucleotides of the tumour suppressor gene, BRCA2.

The third stage of this year's Tour de France will start in Cambridge and soon yellow Lycra® will be the new black. As exciting as this circus will be, cycling is best when it's simple. Contemplating the grinding vehicular traffic of the modern world, Iris Murdoch got it right when she said, 'Only the bicycle remains pure in heart'.



TONY COLL
Science Committee member

KETOCONAZOLE DISCONTINUED

FROM OUR CLINICAL COMMITTEE CORRESPONDENT

We have received notification from the Deputy Director of Patient Safety at NHS England that oral ketoconazole is to be discontinued. The drug may cause liver injury, and this is the reason why the Medicines and Healthcare Products Regulatory Agency (MHRA) has discontinued it. This decision has been ratified by the European Commission. This discontinuation was effective from 11 November 2013.

An antifungal agent, it is currently also used occasionally in the drug treatment of Cushing's disease, and is an inhibitor of P450 enzymes. We are aware that the estimated number of patients using off label ketoconazole for the treatment of Cushing's disease is around 200 in the UK.

Although discontinued for use in fungal infections, this product will remain available to specialist endocrine prescribers if necessary in the treatment of Cushing's disease.

The manufacturers Janssen Cilag produced these tablets. The specialist importers (IDIS and Pharmarama) have confirmed that they will be able to obtain unlicensed (non-Janssen Cilag) ketoconazole from abroad for individual patient use. The MHRA will not object to such import although they will issue a warning to be passed on to the prescribers drawing their attention to the European Medicines Agency/MHRA recommendations.

MHRA guidance to healthcare professionals can be found at <http://bit.ly/1eDLUd6>.

JOHN WASS
External Clinical Relations Advisor,
Society for Endocrinology



CORPORATE SUPPORTERS 2014

Platinum Supporters:
Bioscientifica Ltd

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For more information, visit www.endocrinology.org/corporate or contact amanda.helm@endocrinology.org.

You can read about their work on pages 17-18.

CORPORATE SUPPORTERS

The Society for Endocrinology operates a Corporate Supporters' scheme to strengthen our relationship with industry and further our charitable objectives.

We are delighted to highlight the activities of some of our Corporate Supporters here. We thank them for their support and contribution to scientific and clinical endocrinology. Corporate Support is vital to the Society for Endocrinology,

enabling us to further our charitable objectives and engage with endocrinologists, supporting their learning and advancing the science of endocrinology.

For further information,

visit www.endocrinology.org/corporate
or contact amanda.helm@endocrinology.org

PLATINUM SUPPORTER



Bioscientifica, the commercial subsidiary of the Society for Endocrinology, is one of the world's leading providers of high quality knowledge, information and services for the scientific, medical and pharmaceutical industries.

Bioscientifica provides business support and management services tailored to the exact requirements of our clients. Our award-winning range of services includes publishing, association management, membership development, event management, marketing and communications, website management and strategic planning. Our products and services enable our clients to build strong, mutually beneficial relationships between clinicians, scientists, nurses, medical students and pharmaceutical companies.

Profit for good: Bioscientifica exists to support the advancement of medicine and the biological sciences. Through the redistribution of our profits we are able to support the global scientific community and the drive for better public health ... purpose *beyond* profit.

Bioscientifica Ltd, Euro House, 22 Apex Court, Woodlands, Bristol BS32 4JT, UK.

Tel: +44 (0)1454 642240 Email: info@bioscientifica.com Web: www.bioscientifica.com

GOLD SUPPORTERS



Ipsen Ltd is the UK subsidiary of Ipsen, a global specialty-driven pharmaceutical company with total sales exceeding €1.2 billion in 2012. Ipsen's ambition is to become a global leader in the treatment of targeted debilitating diseases with a development strategy supported by three franchises: endocrinology, neurology and uro-oncology. Moreover, the Group has an active policy of partnerships. Research and development focus on innovative patient-driven programmes in the areas of peptides and toxins. In 2012, research and development expenditure totalled more than €250 million, greater than 20% of Group sales. The Group has a total worldwide staff of close to 4,900 employees.

Ipsen has a strong commitment to the field of endocrinology with an active research and development programme. Ipsen has supported the Society since 1997 and, in addition to corporate sponsorship, is also pleased to continue to provide financial support for the Society for Endocrinology Acromegaly Register. For more information on Ipsen, visit our website at www.ipsen.co.uk.

Ipsen Ltd, 190 Bath Road, Slough SL1 3XE, UK.

Tel: +44 (0)1753 627777 Fax: +44 (0)1753 627778 Web: www.ipsen.co.uk



Novartis Oncology strives to become the world's premier oncology company by consistently discovering and developing novel therapies that improve and extend the lives of patients living with cancer and blood disorders.

Supporting oncology healthcare professionals starts with understanding that cancer and blood disorders are a complex group of diseases fuelled by many underlying mechanisms. Research and development at Novartis utilise recent discoveries in molecular genomics, rational drug design and state-of-the-art drug discovery technologies. To support these efforts, the company invests considerably in technologies and expertise both internally and externally.

The robust research and development programme has enabled Novartis to offer a broad range of innovative therapies to help physicians meet patient needs. As an industry leader, Novartis also has one of the broadest and most comprehensive oncology pipelines in the industry.

Novartis is committed to providing patients with access to its therapies. Providing access to cancer treatments is complex, demanding collaboration with industry, governments, national health services, insurers, physicians and patient groups. Novartis works with these groups around the world to find innovative means to enhance access.

Novartis Pharmaceuticals UK Ltd, UK Head Office, Frimley Business Park, Frimley, Camberley GU16 7SR, UK. Tel: +44 (0)1276 692255 Fax: +44 (0)1276 692508 Web: www.novartis.co.uk



ViroPharma Incorporated is an international biopharmaceutical company committed to developing and commercialising novel treatment options for clinicians to address unmet medical needs in patients living with diseases that have limited clinical therapeutic options.

ViroPharma is developing a portfolio of products for rare and orphan diseases, including C1 esterase inhibitor deficiency, Friedreich's ataxia and adrenal insufficiency.

Our goal is to create new standards of care in the way serious diseases are treated, and to build international partnerships with the healthcare professionals that treat patients with these diseases.

ViroPharma Ltd, Chatsworth House, 29 Broadway, Maidenhead SL6 1LY, UK.

Tel: +44 (0)20 75721222 Fax: +44 (0)20 75721221 Web: www.viropharma.com

CORPORATE SUPPORTERS

SILVER SUPPORTERS

Since entering the field in 2002, **HRA Pharma** has been an active member of the rare endocrine diseases community, investing in, developing and supporting the enhancement of medical treatments and services for patients suffering from rare endocrine disorders.

In 2002, HRA Pharma obtained 'orphan' status in Europe for a key product indicated for the symptomatic treatment of advanced adrenal cortical carcinoma and, in 2004, the product was launched across Europe. HRA Pharma also developed Lysosafe, a free of charge therapeutic monitoring service that helps clinicians optimise the treatment of patients receiving this product.

In 2011, the company reinforced its engagement in the endocrine field by acquiring a product already well established in the UK for the management of Cushing's syndrome. HRA Pharma is currently investing in research for this disorder with a view to expanding the current knowledge base and optimising patient management.

HRA Pharma UK & Ireland Ltd, Unit 7, 3rd Floor RB Building, 557 Harrow Road, Kensal Green, London W10 4RH, UK.

Tel: +44 (0)20 89694383 Fax: +44 (0)20 89685345 Web: www.hra-pharma.com



Internis is a UK speciality pharmaceutical company engaged in the development and commercialisation of highly effective and innovative new medicines aimed at the treatment and prevention of a range of common bone disorders, such as osteoporosis and vitamin D3 deficiency.

Our business model is straightforward: 1) Identify and in-license high potential intellectual property from third parties; 2) Complete the necessary development work in collaboration with specialist scientists and research organisations; 3) Bring new, effective, high demand healthcare products to market for sale, focusing exclusively on providing highly effective, patient friendly products, demanded by medical professionals.

We understand the stringent budgetary pressures facing the NHS and are committed to providing cost-effective solutions promoting patient care, while offering high value for money. The bone therapeutic area represents a growing market as the general population ages.

Internis launched its proprietary product Accrete® D3 (calcium/colecalciferol tablets) during 2011, followed by Fultium®-D3 (colecalciferol 800IU equivalent to 20µg vitamin D3) in 2012.

Internis, Carradine House, 237 Regents Park Road, London N3 3LE, UK.

Tel: +44 (0)20 83495422 Fax: +44 (0)20 83465599 Web: www.internispharma.com



Lilly UK provides research and development, manufacturing and commercial operations on behalf of Eli Lilly and Company, a major US pharmaceutical manufacturer.

As one of the top UK pharmaceutical companies, we are dedicated to creating and delivering innovative pharmaceutical healthcare solutions that enable people to live longer, healthier and more active lives.

Lilly's research priorities are aligned with significant UK health needs including diabetes, heart disease, mental health and cancer. We focus on developing personalised, tailored therapies, using the latest scientific advances.

Lilly UK, Lilly House, Priestley Road, Basingstoke RG24 9NL, UK.

Tel: +44 (0)1256 315000 Web: www.lilly.co.uk



Pfizer is the world's largest research-based pharmaceutical company. Our goal is to discover, develop, manufacture and deliver quality, effective prescription medicines to treat and help prevent disease.

Pfizer has an excellent heritage in endocrinology and remains highly committed to its endocrine product portfolio. The company is an industry leader in long term surveillance studies in endocrinology and has an active Investigator-Initiated Research programme supporting advances in medical and scientific knowledge.

Pfizer Ltd, Walton Oaks, Dorking Road, Tadworth KT20 7NS, UK.

Tel: +44 (0)1304 616161 Web: www.pfizer.co.uk



Sandoz Biopharmaceuticals is a Novartis company and a pioneer in the development of biosimilar therapeutics, with a vision to dramatically increase worldwide patient access to essential high quality biopharmaceutical products.

Sandoz was the first company to have a biosimilar approved through the stringent EU regulatory pathway, closely followed by two more biosimilars. By incorporating quality by design with state of the art technology, Sandoz Biopharmaceuticals is committed to helping make quality healthcare affordable for more patients and to slowing the increase in escalating global healthcare costs.

Sandoz Ltd, Frimley Business Park, Frimley, Camberley GU16 7SR, UK.

Tel: +44 (0)1276 698020 Fax: +44 (0)1276 698324 Web: www.sandoz.com



RECOGNISING AND REWARDING EXCELLENCE

Since 1946, the Society has honoured individuals who have made significant achievements in endocrinology with an annual programme of prestigious medals and awards.

SOCIETY MEDALS

Society Medals are awarded to world-leading scientists and clinicians who have carried out landmark work over their lifetime, which continues to inform research and best practice in the field. The award of a Society Medal is the highest accolade granted by the Society. Medallists become part of a notable league of scientific and clinical excellence encompassing many of the leading lights in endocrinology.

Medal presentations at Society BES 2014

DALE MEDAL



2014 Winner

Dr Bert O'Malley

Baylor College of Medicine, Houston, Texas, USA



2013 Winner

Dr Ronald M Evans

The Salk Institute for Biological Studies, La Jolla, California, USA

SOCIETY FOR ENDOCRINOLOGY MEDAL



Professor Andrew Loudon

University of Manchester, Manchester, UK

EUROPEAN MEDAL



Professor Theo Visser

*Erasmus University Medical Centre
Rotterdam, The Netherlands*

HOFFENBERG INTERNATIONAL MEDAL



Professor Robert I McLachlan

Prince Henry's Institute, Melbourne, Australia

TRANSATLANTIC MEDAL



Dr Mitchell Lazar

*University of Pennsylvania,
Philadelphia, USA*

JOURNAL AWARDS NEW FOR 2014!

One way the Society enables the advancement of endocrinology is by publishing cutting-edge research in a series of high-quality journals. This year, the Society is delighted to be allocating awards on behalf of our five official journals. The Journal Awards recognise excellence in endocrine research and practice in addition to wider contribution to the field of biomedical and biological sciences.

Papers published in our journals during 2013 have been marked by an Awards panel on the basis of originality, scientific content, presentation and impact in their particular field.

Journal Awards Winners

JOURNAL OF ENDOCRINOLOGY

Jessica R Mader, Zachary T Resch, Gary R McLean, Jakob H Mikkelsen, Claus Oxvig, Ronald J Marler and Cheryl A Conover

Mice deficient in PAPP-A show resistance to the development of diabetic nephropathy. *Journal of Endocrinology* **219** 51-58. (OA)

JOURNAL OF MOLECULAR ENDOCRINOLOGY

Eva D'Amico, Stéphanie Gayral, Claude Massart, Jacqueline Van Sande, Jeremy F Reiter, Jacques E Dumont, Bernard Robaye and Stéphane Schurmans

Thyroid-specific inactivation of KIF3A alters the TSH signaling pathway and leads to hypothyroidism. *Journal of Molecular Endocrinology* **50** 375-387.

ENDOCRINE-RELATED CANCER

Xiaoli Liu, Justin Bishop, Yuan Shan, Sara Pai, Dingxie Liu, Avaniyapuram Kannan Murugan, Hui Sun, Adel K El-Naggar and Mingzhao Xing

Highly prevalent TERT promoter mutations in aggressive thyroid cancers. *Endocrine-Related Cancer* **20** 603-610. (OA)

CLINICAL ENDOCRINOLOGY

Ning Yu, Graham P Leese and Peter T Donnan

What predicts adverse outcomes in untreated primary hyperparathyroidism? The Parathyroid Epidemiology and Audit Research Study (PEARS). *Clinical Endocrinology* **79** 27-34.

ENDOCRINE CONNECTIONS

Erika Peverelli, Federica Ermetici, Sabrina Corbetta, Ettore Gozzini, Laura Avagliano, Marco A Zappa, Gaetano Bulfamante, Paolo Beck-Peccoz, Anna Spada and Giovanna Mantovani

PKA regulatory subunit R2B is required for murine and human adipocyte differentiation. *Endocrine Connections* **2** 196-207. (OA)

Plan your visit to the
Society for Endocrinology BES conference 2014 at:
www.endocrinology.org/meetings/2014/sfebes14

2013 IN REVIEW: WORKING TOGETHER TO ADVANCE ENDOCRINOLOGY

WRITTEN BY LAURA UDAKIS



2013 saw the establishment of a new era for the Society, in which we built on strong foundations to strengthen our position as a world-leading authority on hormones. The Society focused on listening to you, our members, as well as clarifying our identity and embedding our values: engage, support, advance.



ENGAGE | SUPPORT | ADVANCE

Here are our top ten achievements:

1. GROWTH OF OUR INTERNATIONAL NETWORK

Our international membership grew in 2013, expanding the Society's global network; we now have members in 60 countries around the world. Our work with international partner organisations has also increased, including our collaboration with the Endocrine Society of India, The Endocrine Society and the International Society of Endocrinology to organise International Clinical Update, held in February 2014 in Hyderabad, India.

2. BETTER UNDERSTANDING OF YOUR NEEDS

We took the opportunity to listen to you, our members, and to talk to you about the Society's future direction by seeking your feedback in creative ways, including a Diary Room at the Society for Endocrinology BES conference, focus groups, workshops, and phone and face-to-face interviews. This feedback is essential to shape Society activities to ensure your interests are kept at heart.



3. CONTINUED IMPROVEMENT OF PATIENT CARE

Enabling healthcare professionals to provide the best possible patient care is a priority for the Society. Our members are world-leading scientists, clinicians and nurses at the forefront of research and practice. In 2013 we made great progress in patient care by developing and publishing a Competency Framework for Adult Endocrine Nursing, in addition to a series of Emergency Endocrine Guidance documents for professionals who don't specialise in endocrinology.



4. STRONGER LINKS OUTSIDE ENDOCRINOLOGY



We worked with organisations representing disciplines allied to endocrinology, including the National Obesity Society, The Physiological Society and the Society of Biology. Together with 12 other learned societies, we also endorsed *Endocrinology, Diabetes & Metabolism Case Reports* – a publication to allow practitioners to share their findings and further both medical education and clinical practice.

5. FURTHER RISES IN IMPACT FACTORS

Many of you participate in the Editorial Boards that govern the Society's five journals. 2013 saw increases in the impact factors of all five journals, making them more competitive and more likely to be chosen by authors.



JOURNAL OF ENDOCRINOLOGY
3.548 4.058

JOURNAL OF MOLECULAR ENDOCRINOLOGY
3.485 3.577

ENDOCRINE-RELATED CANCER
4.364 5.261

CLINICAL ENDOCRINOLOGY
3.168 3.396

● 2012 ● 2013

Find out more about the Society's achievements in our Annual Report (included in this mailing) or visit www.endocrinology.org.

6. A LOUDER VOICE TO POLICY MAKERS

The Society of Biology organises Parliamentary Links Day on behalf of the science and engineering community, to strengthen dialogue between scientists and policy makers. We took this opportunity to highlight endocrinology's central role in many health issues and the work of endocrinologists that will help tackle these.

Some early-career members also took part in Voice of the Future, held at the House of Commons, where they put questions to a panel of policy makers on the future of UK science.

7. A CLEARER IDENTITY



We responded when you, our members, told us that you were confused about the relationship between the Society and its commercial trading company, Bioscientifica. We aimed to clarify the identities of the two organisations by giving each a distinct look, and we will continue to be open, transparent and clear about how they interact.

8. MORE TAILORED EDUCATION AND TRAINING



If your needs as members are not quite being met, we strive to address this. Consequently, we made changes to the flagship Society for Endocrinology BES conference in 2013 to include more focus on translational research, and more focal points for you to discuss the direction of the Society.

Our Young Endocrinologists' Steering Group also worked closely with the Science Committee in 2013 to expand the Career Development Workshops to a dual-track programme. This informal forum for early career researchers to discuss their work and network with more senior colleagues caters for both PhD and postdoctoral researchers, including those who are aiming to apply for fellowships or grant funding.

9. IMPROVED COMMUNICATIONS

It's essential that our members can communicate effectively with each other. You will have noticed a number of changes to *The Endocrinologist*, based on what you told us you wanted. 2013 saw the introduction of a new user-friendly design, lengthened to 32 pages, an expanded Hot Topics section, including reports on articles from around the endocrine world, new Opinion articles from our Chief Executive and Science and Clinical Committees as well as prominent members, and regular feature-length interviews with famous endocrinologists.



10. GREATER MEDIA TRAINING OPPORTUNITIES FOR MEMBERS

More of you than ever took part in Voice of Young Science media workshops in 2013. These workshops, run by Sense about Science, provide invaluable training for early career members, explaining the workings of the media and giving practical tips from journalists and advice from media-experienced scientists.

The Society has five priority places for each workshop and provides grants to cover travel costs.



LAURA UDAKIS
Communications Manager, Society for Endocrinology

ON THE RIGHT TRACK: CAREER DEVELOPMENT WORKSHOPS

The Society recently launched new Career Development Workshops (CDWs) designed to equip basic and clinician scientists with the information and skills they need to build a career in academic research. These workshops are **free** to Society members who are registered for a research degree or to postdoctoral researchers and are split into two tracks.

The next Career Development Workshops will be held in October 2014.

For more information, visit www.endocrinology.org/meetings/CDW.

Career planning ('track 1'): for PhD candidates and postdoctoral researchers covering CV and poster preparation, grant applications, and paper critique.

Fellowship and grant application ('track 2'): for more experienced postdoctoral delegates covering grant applications, project management and interview techniques.

Here, two delegates from 2013 tell us their experience of the workshops, and how the event helped their career planning and knowledge.



Delegates at CDW 2013

A PHD STUDENT'S VIEW...

I am a final year PhD student, and when I finish in a year's time I want to work in the States. I went to the CDW ('track 1') to find out how to apply for postdoctoral positions in the USA and how to make my CV more attractive to employers. I left not only feeling confident and motivated to start applying to US labs, but also with a much better CV, and the drive to apply for my own funding (an opportunity I had not realised was available to me before).

'Exceptional careers guidance – much better than ENDO equivalent'

The chance to talk to experienced faculty members during the workshop sessions (and also in the bar!) was invaluable. It showed me that the Society has a genuine interest in helping young scientists develop their careers.

I would recommend the workshop absolutely to anyone wishing to continue in academia.

LUCY BROOKS
Imperial College London

...AND FROM A SENIOR POST-DOC

I attended CDW 'track 1' back in 2008 when it was known as the Autumn Endocrine Retreat. It was great to return for 'track 2' and see familiar faces from 2008 as well as others considering that step up the academic ladder via the fellowship or direct-to-lectureship route.

'Every aspect of CDW 2013 has been excellent, and very useful for my science career'

At the outset, the workshop seemed a little daunting. We were to have our CVs assessed by successful fellowship holders, pitch our requests for funding to our peers and the faculty (including Fellows of the Academy of Medical Sciences) and undertake a mock fellowship interview based on our research ideas. The welcoming and trusting relationship that was immediately established between the delegates and faculty allowed us to be very open about not only the aspects of our careers that we felt positive about, but also the challenges we may face in our next move. Despite 'feeling the fear' on several occasions over the weekend, the experience and feedback we all received was invaluable.

It was good to have free time to network with other delegates. The faculty members also made themselves available to talk on a one-to-one basis throughout the weekend. In addition, 'track 2' delegates were offered the services of the 'track 2' faculty to coach us for specific fellowship applications. We aim to continue the tradition of previous CDW delegates, meeting up at Society for Endocrinology BES conferences and providing peer-to-peer support for the future.

If you want guidance on how to approach fellowship/grant applications or wish to discuss options outside your local environment, the Society has, through these workshops, provided the 'tools' for you to equip yourself to climb the next rung on the slippery academic ladder. This workshop would not have been so successful without the huge effort and expertise of all the faculty members.



VICTORIA CABRERA-SHARP
Royal Veterinary College, London

PUBLIC ENGAGEMENT – IGNORANCE IS BLISS?

I'm not going to tell you what public engagement is. You probably engage all the time.

You discuss muscle physiology with your hockey team, ponder over the genetic potential for unicorns with your niece (think Jurassic Park but with horses and a hint of rhinoceros), and diagnose the subjects of classic portraits with strange diseases. And sometimes, just sometimes, you'll come to some realisation about how your work fits in to the grand scheme of things.

But would the man in the street understand the significance of what you're doing? Scientists open themselves up to criticism from peers on a daily basis, but have been notoriously poor at getting to grips with what their field means to the public. Perhaps the question really is 'do you even want to know?'

The Society thinks you should. To help you, we offer grants of up to £1,000 to fund good ideas in engagement of the public with endocrinology. So far we've funded exhibitions at music festivals, patient study days, and a host of lectures across the country and the world. Those who've taken part have found it to be a truly rewarding experience, and have brought back to their labs the unique perspective that comes only from letting a multicultural, multitalented and multifaceted audience into the secrets of the science.

TOBY STEAD

Public & Media Relations Executive

ENDOCRINE CONNECTIONS: JUST WHAT ARE WE CONNECTING?

Could it be a dating site for endocrinologists?

A social network maybe? A job site?

In fact, in case you didn't know, *Endocrine Connections* is an open access journal, but one with a twist. If you have ever struggled to get your work published in traditional journals because of their restrictive scopes, you will find *Endocrine Connections* something of a relief. Its interdisciplinary nature aims to appeal to those who work in endocrinology and beyond.

Historically, research has tended to be siloed into distinct disciplines, with journals dedicating themselves to specific fields – making it easier for them to become renowned in that one specialism. *Endocrine Connections* doesn't have those boundaries. This open access journal is for those working in any of the many fields connected to endocrinology.

Consider endocrine disruptors for example; how do you classify research in this area? Endocrinology, wildlife toxicology, neurobiology, molecular biology, reproduction and development, oncology, epidemiology, ecology? On a larger scale, the birth of many new disciplines has resulted from two previously independent subjects coming together, such as ecological economics or philosophy of science.

HOW ARE WE ENGAGING?

Science festivals are a great place to get inspiration for different types of activity. The Society will be at two of the UK's largest science festivals in spring 2014 to deliver our latest event: 'Hormones and the mating game'.

We'll be exploring endocrinology's influence on courtship, relationships and libido in humans and the animal kingdom, and ask 'are we really slaves to our hormones?'

 10:00, 1 MARCH 2014,

BRIGHTON SCIENCE FESTIVAL

Speakers: Petra Boynton (London), Mike Ludwig (Edinburgh);
Chair: Anna Crown (Brighton)

 17:30, 11 APRIL 2014,

EDINBURGH INTERNATIONAL SCIENCE FESTIVAL

Speakers: Gareth Leng (Edinburgh), Richard Quinton (Newcastle upon Tyne); Chair: Philippa Saunders (Edinburgh)

Find out more at www.endocrinology.org/public/events.



Society-sponsored stand at *The Times* Cheltenham Science Festival, manned by students from Imperial College London

To find out more and submit your article to *Endocrine Connections*, visit www.endocrineconnections.com

 @EndoConnect

 www.facebook.com/EndocrineConnections

By widening the journal's scope, we remove constraints for publication, not just for authors who want their work to be easily accessible to a large relevant audience, but for readers who want an overview of an entire field in one place. For both, broadening reach ensures wide dissemination of intelligence that is of high appeal beyond the core, arguably niche, endocrinology community. *Endocrine Connections* allows the work of endocrinologists and others to have an impact across the whole spectrum of science.

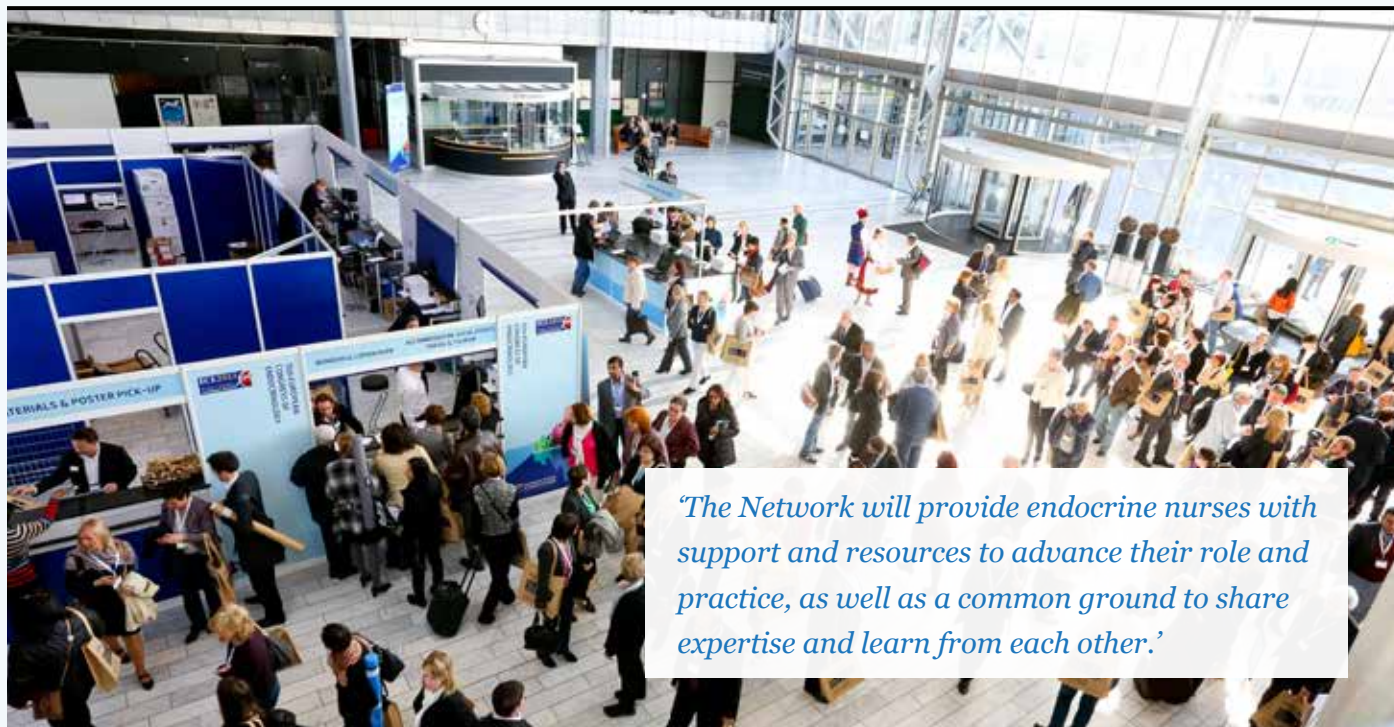
Scientific discovery is incremental and inevitably the scope of investigations creeps over boundaries between disciplines. Collaborations need to form between researchers in different fields to prevent progress from stalling. They allow scientists and clinicians to attack a challenge from all sides, focusing on problem-centred rather than discipline-centric investigation, while benefiting from the associated efficiencies: new tools, new ways of thinking, wider perspectives. Importantly, interdisciplinary research has become a priority for funding bodies. Our aim is to remove barriers to cross-disciplinary collaboration at the stage of publication.



VICTORIA MERRIMAN
Marketing Executive

ESE NURSES' NETWORK: A EUROPEAN AND INTERNATIONAL COMMUNITY

WRITTEN BY SOFIA LLAHANA



'The Network will provide endocrine nurses with support and resources to advance their role and practice, as well as a common ground to share expertise and learn from each other.'

In 2011, the Dutch Endocrine Nurses Group took the initiative of organising the first ever networking meeting for nurses during the European Congress of Endocrinology (ECE) which was held in Rotterdam, The Netherlands.

This sparked considerable interest from the European Society of Endocrinology (ESE), who are keen to support the development of endocrine nursing across Europe. And so it was that Associate Professor Pia Burman, Chair of ESE's Clinical Committee, encouraged the creation of the ESE Nurses' Working Group in early 2013. The Working Group aims to lead on nursing activities, including the creation of a European Nurses' Network. The Network provides endocrine nurses with support and resources to advance their role and practice, as well as a common ground to share expertise and learn from each other. You can find out more at www.ece-hormones.org/nurse.

One of the main activities of the ESE Nurses' Working Group is the development of the Nurses' Programme at ECE. This provides a forum for exchange and discussion of ideas related to patient care, nursing research and advancement of the endocrine nursing role across Europe and internationally.

The joint International and European Congress of Endocrinology (ICE/ECE 2012) in Florence, Italy, saw the introduction of the dedicated Nurses' Programme, which included two multidisciplinary scientific sessions and an evening networking meeting. These sessions were attended by an overwhelming number of nurses and physicians and generated very positive feedback. In Copenhagen, Denmark, at ECE 2013, we promoted advanced nursing practice, introducing a Meet the Nurse Expert session, where colleagues shared their practice and research. We also wanted to emphasise the importance of involving our patients in

decision-making; the session on congenital adrenal hyperplasia involved a physician, a nurse and a patient speaker.

The Nurses' Programme for ECE 2014 promises to be even better still! This year, the networking session will focus on improving clinicians' collaboration with patients and their families. Our patients' opinions of the services we offer are vital in helping us to continuously improve standards of care. ECE 2014 is the ideal place to establish such a collaboration, and we have invited patients and representatives from patient support groups across Europe.

We also aim to promote nursing practice and research by having a dedicated session for nursing poster presentations, where authors can discuss their work with colleagues and inspire more nurses to present their work and to be involved in research activities. The Best Nursing Poster Award will enable the winner to present their work at a future ECE. To help colleagues to prepare posters, a number of nursing posters can be found at www.ece2014.org/nurses.aspx.

Although the ESE Nurses' Network was only established within the past year, almost 150 nurses from across Europe have already joined and are actively involved in promoting and sharing best practice. UK endocrine nurses have an enormous amount of expertise to share and we certainly have a lot to learn from colleagues across Europe and internationally.

SOFIA LLAHANA
Chair, ESE Nurses' Working Group and
Society for Endocrinology Nurse Committee member

Sofia Llahana is Consultant Nurse in Endocrinology at University College Hospital, London. Other members of the ESE Nurses' Working Group are Cecilia Follin (Nurse Practitioner in Endocrinology, Sweden), Meg Keil (Nurse Practitioner at NIH, USA), Judith van Eck (Nurse Practitioner in Endocrinology, The Netherlands) and Phillip Yeoh (Endocrinology and Diabetes Manager, UK).

LISA SHEPHERD NURSE COMMITTEE CHAIR



As I take over the role of Chair of the Nurse Committee, I look back over the last 4 years of the Committee's work and see how much we have achieved. I would like to take this opportunity to thank Nikki Kieffer for leading these developments in her time as Chair. The introduction of the Competency Framework has generated interest and excitement nationally and internationally, serving as a link with our international nurse colleagues and forming a collaboration for nurse education and practice.

In this issue, Sofia Llahana describes one important partnership, in the form of the European Society of Endocrinology's Nurses' Working Group. This demonstrates how much an alliance between international nurses can accomplish.

Jean Munday discusses continued collaboration in the Society's Interdepartmental Peer Review scheme. This is something that I would encourage you to participate in, as it is an opportunity for endocrine nurses to share their practice and demonstrate their successes. The outcomes of the Peer Review process can act as tools to initiate future improvements for nurses.

I also welcome two new members of the Nurse Committee, Kate Davies and Sherwin Criseno. I am very much looking forward to working with them and the rest of the Committee in my time as Chair. Once again, we will have exciting nurse sessions at the Society for Endocrinology BES 2014 conference in Liverpool, and I look forward to seeing you all there.

LISA SHEPHERD

ENDOCRINE NURSES AND PEER REVIEW: WHAT DOES IT MEAN FOR YOU?

The Society for Endocrinology provides a peer review service for endocrine centres in the UK. As part of this process, each centre's endocrine nurse service is evaluated. As a nurse at a centre that is due to be reviewed, you may feel unsure of what is expected of you, or apprehensive about how the visit will go. You should not feel concerned, however, as the visit is designed to help you.

The review team usually consists of two consultant endocrinologists and two endocrine nurses, who want to help you showcase the good practice you already deliver, as well as highlighting areas for future development. Before the visit, the centre will be asked to provide background information, such as the numbers of patients seen, the services provided etc. As an endocrine nurse, you should ensure your part of the service is included in this response and provide copies of any leaflets, pathways, protocols, etc., for the reviewing team to see.

It is likely that you will have an opportunity to speak to the reviewing nurses alone, as well as with the team as a whole. They are on your side, and want to have the opportunity to write about the good things you do in the visit report. During the discussion, they may have ideas about how you could do things differently, or ideas for new developments you could consider. This is always with the intention of helping you to identify ways to enhance the service you provide to endocrine patients.

Many endocrine nurses work in isolation, and are managed by senior nurses who are not endocrine nurses themselves. Being part of a peer review process will give you expert evidence for your appraisal. If you know there is a need for something you currently do not deliver, it is also your chance to discuss this with the peer review team. Having the weight of the Society behind your request may help you to move your service forward.

If your centre is being peer reviewed, embrace this opportunity to demonstrate to the wider team, and management, all the things you have achieved so far, and your ideas for the future.

JEAN MUNDAY

Lead Nurse Endocrinology, Queen Alexandra Hospital, Portsmouth

For more information on the Society's interdepartmental peer review team, visit

www.endocrinology.org/about/projects/peerreview.html.



CLINICAL ENDOCRINOLOGY TRUST FUNDING FOR QUALITY IMPROVEMENT PROJECTS

GENERAL NEWS

The Clinical Endocrinology Trust (CET) is a charity which derives its income from a profit-share of the Society's official clinical journal, *Clinical Endocrinology*. The Trust has long supported UK endocrine audit projects, recent examples including the UK Acromegaly Database, the CaHASE audit of adults with congenital adrenal hyperplasia, and a British Thyroid Association study of teenage iodine status across the UK.

The Trustees now invite further applications from societies or endocrine centres in the UK. Preference will be given to projects involving multicentre collaborations, with particular interest being shown in applications related to areas of endocrinology the Trust has not supported previously.

A total sum of £50,000 is available during 2014–2015 for a number of projects judged by the Trustees to be worthy of support; their decision will be final. The Trustees require that projects are commenced within 6 months of an award being made.

Application forms are available from judith.toon@manchester.ac.uk and should be returned to Julian Davis (CET Secretary) at julian.davis@manchester.ac.uk by the closing date: 30 June 2014.

The CET looks forward to hearing from you!

AN INTERVIEW WITH... PHIL LOWRY

INTERVIEW BY MILES LEVY



I have been collecting a list of names suggested by colleagues as good people to read about in this series of interviews. Phil Lowry is a name that crops up all the time. Recognised as one of most influential basic scientists in our specialty, Lowry is the one who hypothesised that hormones such as adrenocorticotrophin (ACTH) are produced as precursors – a concept that was entirely new at the time. I am looking forward to finding out more about how this man from working class Birkenhead ended up playing a key role in modern endocrinology...

When I initially asked him to take part, Phil was refreshingly modest and pleased to be the first non-clinician involved – ‘I thought it was a wind-up at first. I must admit it would be an honour, particularly as I am from the fundamental spectrum of the Society and it may be encouraging for the troops.’

I am greeted by a smiling Phil Lowry as I pull up in the driveway of his house. He is wearing beige slacks, is tall and looks fit for his 69 years. He speaks with a subtle ‘Scouse’ accent, probably watered down from years of living in the south, and has a very down to earth manner. I have already been invited to stay for dinner, ‘We have a pheasant in the freezer which should stretch to three servings – you would have a much easier journey back [if you leave later].’ His wife Sue has even made up a bed in case I am too tired to drive back.

One of the first things we do is go into his garden where he shows me a slate sundial with an engraved message on the plinth. This is a gift from past and present members of his research group, presented as his ‘festschrift’, and he seems visibly moved when he reads the affectionate message aloud. We go into his lounge where Sue brings us tea and we make a start.

How did he discover he was good at laboratory science? ‘I just had a feel for things – I take a DIY approach to experiments; I am good at thinking on my feet and making it up as I go along,’ he says as we embark on his story.

IN THE BEGINNING

Brought up in Birkenhead, Merseyside, Phil Lowry was definitely not born with a silver spoon in his mouth. His father left school at 14 and eventually joined the police force. Phil’s mother had high hopes that her son might become a supervisor in the local soap factory and wear a white coat, a sure sign of respectability.

Phil went to the local grammar school, doing well at science. ‘I was the only one who passed A-level biology – even though my school reports commented on a lack of concentration after I discovered the fairer sex.’ He had a flair for chemistry and got chatting about careers to a local lad in a youth club, who happened to be a medical student. ‘Why not do biochemistry?’ he suggested. This was enough for Phil to apply for a BSc in Biochemistry at Leeds.

He describes his first day arriving at university digs, being greeted by his landlady standing at the door ‘with a fag in her mouth’ and being introduced to his 80-year-old fellow lodger. Despite this inauspicious start, the curriculum was perfect – it covered the three forms of pure chemistry (physical, inorganic and organic, he reminds me). Inspired by early lectures in physiology at the medical school, where basic science was applied to humans, he was a natural in the lab. ‘We were allowed to do real experiments with animals. I loved solving difficult problems relating to human physiology.’

JIMMIE DODD, □-MSH AND GRIMSBY DOCKS

A key figure in Lowry’s life was his Professor of Zoology and mentor, the late James Munro Dodd FRS (‘Jimmie Dodd’), a pioneer of comparative endocrinology. For his PhD project, Dodd wanted Lowry to find out whether the hormone that caused dogfish to change to a black colour on a dark background was similar to that found in mammals, including humans, who did not. Unwittingly, Lowry had just irreversibly embarked on a life’s work on endocrine peptides.

Phil spent much time with the fishermen of Grimsby Fish Docks, collecting the pituitary glands of countless dogfish. He recalls, ‘They cut off the head of the fish and I would hook out the neurointermediate lobe, which was the size of a matchstick head. I must have hooked out literally thousands of dogfish pituitaries. By the way, they do the best fish and chips in Grimsby!’

‘The high quality of Lowry’s GH preparation was singled out for praise by Mr Justice Morland in his report on the case that found the Government culpable for children who contracted CJD from elsewhere’

Lowry mastered column chromatography, a method for purifying chemical compounds according to molecular weight and ionic charge, which was similar to the methods used by his scientific hero Frederick Sanger. ‘I would mash up the dogfish pituitaries and let the extract trickle down each resin, using a frog skin bioassay to monitor the purification, which would eventually result in a purified white powder. I would hydrolyse a

sample to release its amino acids and analyse them to get the composition. I would then digest another sample into bits and analyse the amino acids from either end so I could piece it all together.'

In a series of classical experiments, Lowry showed that the amino acid sequences of α -melanocyte-stimulating hormone (α -MSH) in mammals and dogfish were identical apart from a single methionine/valine substitution. The larger ACTH molecule had exactly the same substitution. This was too much of a coincidence, and Lowry hypothesised that a precursor molecule was broken down into smaller components including ACTH and α -MSH.

Not only had Lowry answered Jimmie Dodd's research question, he had also proposed a fundamental hypothesis regarding post-translational modification of hormones. His first paper in 1970 was Phil's first author *Nature* paper¹ and he became Dodd's 'blue-eyed boy'.

His career as a research scientist took off and he was snapped up by the Swiss scientific arm of Novartis, CIBA (Chemical Industries Basel). The UK base was in Horsham, Sussex, where he moved with Sue, whom he had met at university whilst she was a fellow zoology PhD student. The Swiss had a good tradition of synthetic peptide chemistry: 'They were very encouraging and I got on well.' As well as developing ACTH analogues, CIBA also synthesised salmon calcitonin for use in Paget's disease, so Lowry was able to persuade them to allow him to get hold of more dogfish pituitaries to continue his passion for all matters ACTH-related.

COLLABORATION WITH CLINICAL MEDICINE

At this time, John Landon, pioneer of antibody use in clinical endocrinology, had started measuring ACTH by radioimmunoassay and needed a talented basic scientist. Sandy Scott, a PhD student, was 'getting funny results using ACTH anti-sera in detecting peptides in pars intermedia and couldn't work out what was going on'. Scott visited Jimmie Dodd for advice, who didn't know the answer but suggested he should 'go and see Phil' for help.

Lowry solved Scott's ACTH problem and was now on Landon's radar. It was not long before he became Principal Biochemist and Honorary Lecturer in Chemical Pathology at Barts. Lowry developed a sensitive analyser for amino acids that required only small amounts of material and continued work on ACTH. In 1973, with Sandy Scott, he identified a new corticotrophin-like intermediate peptide, otherwise known as CLIP.²

He concluded that ACTH was produced from a precursor (later known as pro-opiomelanocortin, POMC), and that ACTH, which comprises 39 amino acids, is processed to form α -MSH (ACTH 1-13) and CLIP (ACTH 18-39). The incontrovertible evidence that pituitary hormones were broken down into biologically active compounds changed the world of endocrinology: 'After this the floodgates opened and that was that.'

Having a chemist of Lowry's technical ability facilitated a raft of clinical publications demonstrating the presence of ACTH and other peptides in endocrine tumours. Lowry purified human pituitary hormones and raised anti-sera to ACTH, growth hormone (GH), prolactin, thyrotrophin, luteinising hormone and follicle-stimulating hormone, providing reagents for an in-house assay service that was the envy of the world. Lowry describes how visiting professors from around the globe were amused at the unglamorous laboratory environment whence such leading research emanated, 'You should have seen the look on their faces when they saw our dingy labs!'

A VERY PURE GROWTH HORMONE

Human GH (hGH) was becoming an important new treatment for children with GH deficiency – 'Bovine GH was no good.' The MRC needed a scientist to purify hGH. 'A minimum of 200 or 300 human pituitary glands were needed to extract a decent quantity of the hormone.' Lowry rose to the challenge, recalling, 'Me being me, I read all about it and really cleaned up the method.' He helped to produce therapeutic purified hGH on a large scale, and was determined that it would not be contaminated.

Alan Dickinson at the MRC Neuropathogenesis Unit in Edinburgh, an expert on 'slow viruses,' had alerted Head Office to the possibility of contamination of hGH extracted from cadaver pituitary glands, so Lowry flew to Edinburgh to plan a study there to infect a human pituitary extract with 10⁸ units of scrapie virus, to show his method could purify material to be free even of that. A neuro-degenerative disease, scrapie is now known to be closely related to bovine spongiform encephalopathy (BSE) and Creutzfeldt-Jakob disease (CJD), all caused by a common infectious prion protein.

After purification, no scrapie or pyrogens were present, and it was concluded that his GH was safe. 'Talk about Dickinson's crystal ball!' exclaims Lowry. He explains, with justifiable pride, how his GH has led to not a single case of CJD.^{3,4} The high quality of Lowry's GH preparation was singled out for praise by Mr Justice Morland in his written report on the case that eventually found the Government culpable for children who contracted CJD from elsewhere.

ADRENAL HYPERPLASIA, VASOPRESSIN AND CRF

Lowry remained fascinated by adrenal endocrinology. He published work on pro-enkephalin, a natural opiate precursor expressed in brain, adrenal medulla and adrenal medullary tumours. He became interested in the observation that, following unilateral adrenalectomy, 'the other one grew by at least 50% due to increased cell number [hyperplasia] in the cortex.'

Mirroring similar studies on pro-insulin, Lowry showed that pro- γ -MSH was activated into a mitogen by cleavage near its N-terminal via a neurally mediated mechanism after removal of one adrenal. This led to 2 more of his 15 *Nature* papers.^{5,6} He showed that pro- γ -MSH was not only important in adrenal cell hypertrophy, but also increased the effect of ACTH on the adrenal cortex by increasing the amount of mRNA involved.

Lowry did seminal work on the interaction between corticotrophin-releasing factor (CRF) and vasopressin. With PhD student Glenda Gillies (now Professor of Neuroendocrinology at Imperial College London), he developed a column of stable live pituitary cells for *in vitro* studies. Their experiments demonstrated that vasopressin and CRF act synergistically to release ACTH from the pituitary.⁷⁻⁹ I mention in passing a patient I had seen that morning, whose diabetes insipidus was unmasked with steroid replacement, which seems to get him thinking...

THE PLACENTA YEARS

Lowry's work on CRF led him to develop a specific and sensitive 'two-site' assay (he uses two teaspoons to demonstrate this!). He remembered a paper by McGuinness in 1963 suggesting that melanotrophic activity was elevated in the blood of pregnant women, reaching very high concentrations in pre-eclampsia.¹⁰ Now that Lowry could measure CRF, he decided to look at its role in the placenta. He showed that the placenta did secrete CRF, and that concentrations were very high in pregnancy, reaching levels approaching those of portal blood in stressed animals.

CONTINUED ON PAGE 28...

AN INTERVIEW WITH... PHIL LOWRY

...CONTINUED FROM PAGE 27

He found himself asking, 'But why is there no corresponding elevation in ACTH or cortisol?' With another PhD student, Dominic Behan (who a few years later co-founded Arena Pharmaceuticals in California, and was recently awarded a DSc at Reading), and using affinity purification techniques, Lowry demonstrated the presence of a large molecule secreted by the liver that was a high-affinity CRF-binding protein (CRF-BP), which neutralised the biological effect of CRF. He showed that the level of CRF-BP dropped towards term, explaining the increase in cortisol before labour.¹¹ CRF was significantly increased in pregnant women suffering from pregnancy-induced hypertension and pre-term labour, fitting with McGuinness' early observations. This led Lowry to think hard about the potential neuropeptides involved, which became an obsession.

NEUROKININS AND PRE-ECLAMPSIA

Pre-eclampsia, a sign that the mother's placenta is in distress, is well known to cause high blood pressure and vasoconstriction, with worsening disease severity. Lowry wondered if this had a neuropeptide basis. In the early 1990s, his group began to establish gene-mining techniques to look for particular areas of alteration in RNA expression. He looked at placentae from pregnancies terminated between 9 and 13 weeks, and was struck by the high expression of tachykinins, especially neurokinin B (NKB); 'I was hit like a bat out of hell'.

NKB acts on its preferred receptor (NK3) to increase heart rate, contract portal and mesenteric blood vessels and increase blood pressure. Lowry suggested that the placenta is a parasitic organ that produces NKB in times of trouble to preserve its survival, at the expense of the host pregnant mother. Increasing concentrations of this peptide in a failing placenta would allow it to cross-react with lower affinity receptors (NK1 and NK2), providing an explanation for the cerebral, platelet and lung and liver dysfunction commonly seen during pregnancy disorders.¹² 'I was so convinced of the idea that when the penny dropped I could hardly breathe.'

'Phil spent much time with the fishermen of Grimsby, "They cut off the head of the fish and I would hook out the neurointermediate lobe ... I must have hooked out literally thousands of dogfish pituitaries"'

The following part of Lowry's career is testament to his tenacity and conviction in his scientific principles. By now he was moving up the ranks. From Head of Biochemistry and Physiology soon after arriving at the University of Reading, he subsequently went on to become Dean of Science and Head of the new School of Animal and Microbial Sciences. But I am struck during the interview by how Lowry is far more interested in his scientific journey than career progression.

OVERCOMING IMMUNOASSAY ADVERSITY

For his initial placental NKB studies, he used a classical radioimmunoassay in which each plasma sample had to be extracted. However, to handle lots of blood samples, he needed to develop a specific two-site immunoassay that could be carried out directly in plasma. The new immunoassay was good at detecting NKB in the brain, but it hardly detected placental NKB at all. This was a cause of great disappointment and consternation. 'I had stuck my neck out about NKB in pre-eclampsia and was starting to think I should retract it, as I couldn't live with myself. This [inability to demonstrate NKB in the placenta with the new immunoassay] was trying to tell me something.'

He was determined to find out why he could not detect placental NKB, and conducted experiments on the placenta of a pregnant post-doc (with her consent and after delivery!). Lowry embarked on the purification of placental NKB and using 'time of flight' mass spectrometry (a way of determining the mass to charge ratio of a molecule) demonstrated placental NKB was 371 Daltons larger than expected. This suggested that the molecule 'had something stuck onto it'.

That 'something' included a chemical structure called phosphocholine (PC). Publications by other groups showed that PC transferase is highly expressed in the placenta, and that modification of tachykinins with PC would strongly change the immunological reaction, explaining why Lowry couldn't detect placental NKB with his new immunoassay. PC modification is commonly used by parasites to prevent the host immune response. Given that the placenta is a parasitic structure, it makes perfect sense that it should want to be 'radio-silent' to prevent rejection – it modifies its peptides with PC to create a Harry Potter-like invisibility cloak.

EUREKA MOMENT

Even as recently as 3 years ago, there was still something gnawing away at Lowry, as PC did not account for all the mass difference between brain and placental NKB. 'You ask my wife Sue over dinner how much better I've been sleeping in the last 3 years. It was driving me crazy, but during a holiday in Greece I suddenly realised the complete structure!'

His Greek eureka moment was to finally be able to visualise the correct structure of his proposed NKB molecule. The chemical structure of the PC-containing group that was attached to placental NKB that dawned on Lowry – whilst supposedly relaxing – is similar to platelet-activating factor and fits entirely with its molecular mass and biological function.¹³

The fact that this was still nagging away at his subconscious well after retirement shows, I think, that this man is 'the genuine article', and also suggests that he is plagued by a highly active mind.

‘Lowry suggested that the placenta is a parasitic organ that produces NKB in times of trouble to preserve its survival, at the expense of the host pregnant mother. “I was so convinced of the idea that when the penny dropped I could hardly breathe.”’

Interestingly, it is only now that Lowry’s principles are being accepted by the obstetric community. Until recently, his ideas regarding pre-eclampsia seem to have been shunned by placental academics. Perhaps there was resentment that an endocrinologist, a relative outsider to this clinical field, had dared to come up with hypotheses that concerned a condition belonging to obstetricians rather than endocrinologists and which ‘did not fit with their pet theories’. Pleasingly, only a few weeks before this interview, he was invited to give a talk at the O₂ Arena; NKB is finally being recognised as an exciting diagnostic and therapeutic area in pre-eclampsia.

PHILIP LOWRY THE MAN

We have been chatting for nearly 5 hours and it is time to eat. Over pheasant (with good red wine that Phil has chosen), I learn that he was a keen rock climber at college. He broke his elbow during an expedition during his final year, which may have explained the 2.2 he got in his degree in biochemistry – Sue enjoys telling me the latter! He listens to classical music and enjoys walking and skiing. Much of their retirement is spent in Tignes le Lac where they own an apartment. They are in good shape for their age; Phil has a family history of heart disease and is keen to avoid the fate of some family members.

Throughout our meeting, Phil strikes me as a deeply principled man. He worries about the standard of grass roots science taught at school, is concerned about the lack of social mobility, berating a recent speech given by Boris Johnson. He is very grateful for his career and for how far the boy from Birkenhead has come. ‘I have to pinch myself when I look back.’ After all, he got to wear a white coat, just as his mum wanted!

As I drive back to Leicester, I wonder in how much of a chasm not being actively involved in research must leave someone like Phil. Since our meeting he has emailed me about the diabetes insipidus patient I mentioned in passing, explaining how his early findings on vasopressin and ACTH, and the discovery by others of glucocorticoid receptor binding regulatory elements in the vasopressin gene, are likely to explain this phenomenon. Phil’s research group seem to have been an extension of his family. That visit to see the sundial when we first met, to read the message of deep affection from members of his research group, suggests a deep bond with his close colleagues.

I reflect how our great UK endocrine institutions are a product of the symbiotic relationship between clinicians and pioneering scientists such as Lowry. He followed where the science took him, and stuck to his core research ideas rather than following trends. I wonder whether he would have been given the freedom to do this in today’s environment, where large funding favours common disease for health economy benefits, rather than rare disease that may have unforeseen fundamental scientific relevance.

I feel I have met a true diamond in the rough, a scientist who could hold his own in the company of his heroes Sanger and Crick. As I head up the M1, I wonder how long it will be until endocrinology is graced with the likes of a Philip Lowry again.

MILES LEVY
Editor, The Endocrinologist

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MANAGING TYPE 2 DIABETES: TOWARDS A STRATIFIED APPROACH

WRITTEN BY EWAN PEARSON



Diabetes is a highly heterogeneous condition, yet currently all patients with type 2 diabetes are treated the same. The hope is that a stratified approach would allow subclassification of patients by aetiology, likelihood of progression and risk of complications, as well as the targeting of therapy to these subgroups.

Diagnosis of diabetes is easy – someone simply needs to have a fasting blood glucose over 7mmol/l, or glycated haemoglobin (HbA1c) greater than 6.5%. However, this simplicity ignores the multiple mechanisms by which people can develop hyperglycaemia. Whilst there is some attempt to stratify diabetes into different types, e.g. type 1 diabetes, type 2 diabetes, gestational diabetes, the current approaches are largely based upon clinical criteria.

To be defined as having ‘type 2 diabetes’ one simply needs to not have type 1 diabetes or any other obvious aetiology. This means that type 2 diabetes is a mixed bag of conditions. It seems reasonable to assume that some of these subconditions will behave differently to others – whether in terms of rate of progression, response to therapy or risk of complications.

A ROLE FOR PHARMACOGENETICS

The modern concept of stratified medicine attempts to identify subgroups that behave differently by careful clinical phenotyping and biomarker discovery. One aspect of stratification is pharmacogenetics – the use of genetics to target drug treatment. There are examples of pharmacogenetics currently in mainstream clinical use. The two main areas are adverse drug reaction (e.g. abacavir hypersensitivity, azathioprine toxicity) and cancer treatment (aided by the ability to sequence tumour tissue and target therapy directly at the cancer tissue itself). Outside these areas, there are limited examples of practical use of genetics to guide therapy. I would argue that diabetes is one key area where genotyping is established to guide therapy – in the field of monogenic diabetes.

Although maturity onset diabetes of the young (MODY) was defined on clinical grounds from the 1970s, it was not until the mid-1990s that the genetic aetiology was dissected out. Knowing the genetic aetiology was important for genetic counselling of family members, but did not alter the diabetes therapy. In 2003, Andrew Hattersley and I published a trial of metformin and sulphonylureas in patients with MODY due to an HNF1A mutation compared with patients with ‘type 2 diabetes’.¹ The HNF1A patients were over four times more sensitive to the hypoglycaemic effect of sulphonylureas than patients with type 2 diabetes. This trial established that patients with this genetic aetiology of diabetes should be treated with sulphonylureas not metformin. This is a clear example of pharmacogenetics that now is in mainstream clinical practice.

ADDRESSING TYPE 2 DIABETES

What about stratification in ‘type 2 diabetes’? Here, there has been a lot of progress in recent years to identify genetic variants associated with response. A proof of principle study was carried out in Dundee, using electronic health record data linked to a biobank of 10,000 patients with diabetes. We showed that patients who do not metabolise sulphonylureas very well, due to the presence of two loss of function mutations in the cytochrome P450 gene *CYP2C9*, are 3.4 times more likely to achieve a treatment target than those who have normal *CYP2C9*. However, even despite this relatively large effect, the clinical utility of knowing somebody’s *CYP2C9* genotype before prescribing sulphonylureas has yet to be established.

So, will we be able to stratify effectively in type 2 diabetes and, if so, when? I believe that our over-simplified approach to the diagnosis and management of diabetes makes it highly likely that we will be able to dissect out different subgroups of ‘type 2 diabetes’. We need much better phenotypic characterisation of patients, better trials designed to assess multiple drugs in the same individual, and the application of ‘-omics’ technologies: not just genomics, but metabolomics, proteomics and metagenomics.

There are a number of initiatives focusing on exactly these challenges, in particular, the IMI-DIRECT (www.direct-diabetes.org) and ABPI/MRC MASTERMIND projects. I believe these and other projects will deliver, and that multiple diabetes subtypes will be discovered. This means that the future of diabetes care is set to become far more complex, but that hopefully we will be able to better target the rapidly increasing number of therapies available to particular groups of patients.

EWAN PEARSON

*Professor of Diabetic Medicine, Medical Research Institute,
University of Dundee*

*Ewan Pearson is Professor of Diabetic Medicine at the University of Dundee.
His research interests are the genetic and physiological determinants of drug
response in diabetes.*

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IMAGES IN ENDOCRINOLOGY

Here are the latest highlights from our journal Cover Art Competition, showcasing the best images in endocrinology.

COVER IMAGE FROM ENDOCRINE- RELATED CANCER, OCTOBER 2013

A confocal image of a mammary carcinoma from a medroxyprogesterone acetate-induced mouse breast cancer model transplanted into a GFP-BALB/c mouse (stroma depicted in green) treated with antiprogesterins (to induce tissue remodelling) and doxorubicin (red autofluorescence).

Credit: G Sequeira & C Lanari, Laboratorio de Carcinogénesis Hormonal, Insitituto de Biología y Medicina Experimental, Buenos Aires, Argentina.



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Tostran[®] (testosterone) 2% Gel Prescribing Information

Please refer to the Summary of Product Characteristics (SPC) before prescribing.

Presentation: Tostran 2% Gel, contains testosterone, 20 mg/g. **Indication:** Replacement therapy with testosterone for male hypogonadism when testosterone deficiency has been confirmed by clinical symptoms and laboratory analyses. **Dose:** The starting dose is 3 g gel (60 mg testosterone) applied once daily to clean, dry, intact skin, on the abdomen or to both inner thighs. Adjust dose according to clinical and laboratory responses. Do not exceed 4 g of gel (80 mg testosterone) daily. Apply after washing, bathing or showering. Do not apply to the genitals. Do not use in women, or children under the age of 18 years. **Contraindications:** Known or suspected carcinoma of the breast or the prostate. Hypersensitivity to any of the ingredients. **Special warnings and precautions for use:** Not to be used to treat non-

specific symptoms suggestive of hypogonadism if testosterone deficiency has not been demonstrated and if other aetiologies have not been excluded. Not indicated for treatment of male sterility or impotence. Pre-examine all patients to exclude a risk of pre-existing prostatic cancer. Perform regular monitoring of breast and prostate. Androgens may accelerate the development of subclinical prostatic cancer and benign prostatic hyperplasia. Oedema with/without congestive heart failure may be a serious complication in patients with pre-existing cardiac, renal or hepatic disease. Discontinue immediately if such complications occur. Use with caution in hypertension, ischemic heart disease, epilepsy, migraine and sleep apnoea as these conditions may be aggravated. Care should be taken with skeletal metastases due to risk of hypercalcaemia/hypercalcuria. Androgen treatment may result in improved insulin sensitivity. Inform the patient about the risk of testosterone transfer and give safety

instructions. Health professionals/carers should use disposable gloves resistant to alcohols.

Interactions: Androgens can increase the anticoagulant effect of anticoagulants. Concurrent administration with ACTH or corticosteroids may increase the likelihood of oedema. **Side-effects:** *Very common:* application site reactions (including paresthesia, xerosis, pruritis, rash or erythema). *Common:* increased haemoglobin and haematocrit, increased male pattern hair distribution, hypertension, gynaecomastia, peripheral oedema, increased PSA. May cause irritation and dry skin. Consult SPC for further details of side-effects. **Pack Size and Price:** Packs containing one or three 60 g metered-dose canisters per pack. Price £28.67 per canister. **Legal Category:** POM. **Marketing Authorisation Holder:** ProStrakan Limited, Galabank Business Park, Galashiels, TD1 1QH, UK. Marketing Authorisation Number: PL16508/0025 ©ProStrakan. ®Registered Trade Mark. Date of PI Preparation: November 2013.

Adverse events should be reported. Reporting forms and information can be found at www.mhra.gov.uk/yellowcard.
Adverse events should also be reported to ProStrakan Ltd on 01896 664000.

References: 1. Tostran[®] Summary of Product Characteristics, June 2010. 2. Testogel[®] Summary of Product Characteristics, November 2006.
Date of preparation: February 2014. **Job code:** M015/1224